## **Post Mortem Examination of Wild Animals**

Cleve Main Veterinary Pathologist Email: cmain@agric.wa.gov.au

**ABSTRACT:** Be aware that some diseases are transmissible to man; salmonellosis, chlamydiosis, cryptococcosis, avian influenza and Australian bat lyssa virus, just to name a few. Protect your self and others always wear protective clothing and gloves. Clean up and disinfect when you have finished. Don't transfer the pathogen to other animals

Know your subject (not always possible) and learn to know what is normal. Then when something is abnormal you should notice it. Be aware that post mortem change alters the appearance of the subject. Post mortem change is often mistaken for a real pathogenic change by the inexperienced.

If the animal is still alive – note its gait, stance, ability to stand or fly. Observe its appetite and water consumption. Note the frequency or absence of urination and presence of diarrhoea.

If the animal is dead - examine the animal externally – note injury wounds etc. Note the presence and nature of any discharge, oral, aural, ocular, nasal etc.

Necropsy techniques – be methodical and systematic. Note colour abnormalities, bruising, haemorrhage, or staining. Examine every organ and take a wide range of samples. Don't fall into the trap of zeroing in on one organ and ignoring others. Don't make up your mind at the onset that you are dealing with a particular disease and ignore the possibility that you might be wrong or that there may be more than one abnormality or disease present. Describe but don't interpret what you see.

#### **Presentation Notes**

Almost everything discussed in this presentation is contained in Dr Karrie Rose's excellent manual on wild life health investigations (Wildlife Health Investigation Manual). Use it as your guide, even your bible and you can't really go wrong. I commend it to you and urge you to read it even before you need to use it.

When dealing with diseased animals always remember that some diseases are zoonoses; that is, they are transmissible to humans. Avian Influenza has been in the news forthe past year or two. People in Asia who had very close contact with infected birds have died from the disease. Two people in Australia have died from Australian bat lyssavirus infection. One was a bat handler and the other person was bitten by an infected bat. The Australian bat lyssavirus is related to but distinct from the classic rabies virus. Cryptococcosis, psittacosis and salmonellosis are common everyday diseases of birds and animals.

Wear protective clothing, gloves and coveralls at least when handling sick wildlife. Rubber boots are desirable because they can be washed and disinfected easily. Always clean your boots, gloves etc before having contact with healthy animals and don't tempt fate by trying to treat bats with neurological signs.

Just as you would try to avoid close contact with a person with a bad cold, avoid breathing in aerosols and moisture droples from animals with respiratory disease. Similarly beached whales and dolphins may be suffering from pneumonia. The spray from their blowholes may be loaded with pathogens.

Australia is free from Highly Pathogenic Avian Influenza. However some of the less important strains of the virus have been detected in wild birds. We cannot afford to be complacent and people such as yourselves are an important link in our sustained efforts to remain free of Avian Influenza.

General surveillance is the most effective and efficient way of detecting new diseases or incursions of exotic diseases that cause significant clinical signs and/or deaths. General surveillance relies heavily on networks that include people in the poultry and other bird industries, wildlife officers, bird watchers and members of the general public. All need to be alert for unusual disease events and promptly report such events to aveterinarian.

Notes on reporting and sample collection from dead birds for the purposes of avian influenza exclusion have been provided for this conference.

Veterinarians are a diverse group and many of us have not had contact with some of the wild life species that you deal with. From a personal point of view, I have sometimes had to ask myself what a specific organ or piece of tissue is that I have been asked to examine. Less frequently I have had to decide whetherthat piece of tissue is normal for the animal being examined or whether it is diseased or neoplastic. We don't have text books for every species and sometimes need to seek additional information on the animal we are dealing with. The more information you supply to the laboratory the more able we are to help you.

From your own point of view make every effort to know what is normal; normalbehaviour, normal eating habits, normal movements and normal appearance of the animal and its internal organs.

When you have a sick animal in your care, pay particular attention to its gait, head carriage, attempts to fly and its general state of awareness. If you are familiar with the species, that knowledge should help you decide whether its appetite is normal. You will also know whether it should drink regularly or only occasionally. If it has diarrhoea note the colour, consistency and smell. Take samples. If it does not pass faeces or urine, note that as well.

Note external abnormalities and try to assess their age.

If the animal that has been presented is dead, make a thorough external examination first. How long has it been dead? Has rigor mortis set in? What is its body condition? Are there are wounds or injuries? If so are they old or recent? Did they occur after the animal died, eg predator or scavenger damage?

Take note of any discharges from body orifices, but remember that after death, sphincters and muscle tone relaxes allowing urine to leak and stomach content to regurgitate.

The is no single correct method to carry out a post mortem examination. Once again use Karrie Rose's manual as your guide. What ever method you choose, always be methodical. Examine and/or gently feel all organs as much as possible. Take care not to contaminate the organs with dirty knives, scissors and scalpels. Avoid contamination of organs with intestinal content.

Be very gentle with gut samples that you intend to submit for laboratory examination. Take a wide range of samples even if you see something that really stands out. It might be abnormal but it may not have been responsible for the death of your subject.

I have lost count of the number of times that some one submitting samples has limited the range of samples to one or two organs because they had already made up their minds what the problem was. So often we have found that they were wrong but were not able to provide a diagnosis because critical samples were not provided.

Do not make up your mind an animal has pneumonia or enteritis because its lungs were dark red or the intestinal content was fluid. Keep an open mind and just describe what you see - dark red lungs on left side or red intestinal content.

On some occasions several abnormalities may be present. Some of them may be a consequence of another. Some of them may be coincidental or maybe some of them in combination contributed to the animals sickness or death.

Don't assume that an animal merely died because it was savaged by a dog or drowned. That might have been why it died but it may have been lethargic or unable to escape because of disease.

Sample collection is important. The range of samples and the manner in which they are submitted are critical.

You should not expect a diagnosis if the organs are already decomposed or have not been adequately fixed in formalin.

Always provide an adequate history and epidemiological data. Photos and movies are extremely helpful, even those taken with a mobile phone.

Unfixed tissue will easily slip through the narrowneck of a container. Once it is fixed and hardened into shape, it is impossible to remove without cutting open the container or smashing the bottle.

Avoid the temptation to submit large samples in formalin. They can be immersed for a week but are still red in the middle when they are cut open. Red colouration means it is not fixed and therefore autolytic changes will have taken place.

Try very hard to get formalin solution into the lumen of the intestine. It is very fragile and is one of the first organs to decompose.

There is no benefit in submitting fist sized samples for histopathological examination. Firstly they will not fixed properly in formalin and secondly we will only look at one or two small areas.

It is much better to place your samples in a large (leak proof) container of 10% formalin than several small ones. The ratio of formalin solution to tissue should be at least 10:1.

For bacterial culture, use swabs in transport medium unless you can get them to the laboratory very quickly.

Submission of "fresh" tissues are acceptable but they need to be collected aseptically and chilled as soon a possible. Tie off the ends of loops of intestine.

Submit each tissue sample in a clean and separate container or plastic bag. Some viruses are sensitive to freezing, so avoid doing so if you can.

Tissue samples for virological testing or biochemical analysis should also be submitted in separate clean containers.

The Animal Health Laboratories do not do blood counts or other haematological tests. We do carry out tests on blood, plasma and serum for bacterial and viral antibodies, chemical analyses, and for viral particles.

We prefer blood in lithium heparin tubes, not EDTA. Some tests cannot be done on plasma or serum. PCR for bacterial or viral antigen requires white blood cells.

Laboratory staff should not be exposed to biological hazards. A big "no no" is submission of syringes with needles attached. If the needle is removed then the contents may leak out creating another sort of biological hazard. If the blood clots in the syringe we will injure the cells in trying to remove it.

Paper work. Provide as much information as you can on your submission sheet. State the sex, age, diet and population data. State clearly what samples you have submitted. Label each container clearly and make sure it matches what you have written on the sheet. Try and be specific as to what you want the laboratory to do. What information are you hoping the laboratory tests will provide?

Finally - take the time to prepare clean and well labelled samples in leakproof containers. Laboratory staff are only to willing to help you - we are only a phone call away.

## **BIOGRAPHY:**

Cleve Main is a veterinary pathologist with the Department of Agriculture and Foods, Animal Health Laboratories. His area of expertise is diagnostic pathology, which has a strong bias towards exclusion of animal diseases exotic to Australia. Although the majority of his work involves domestic livestock, he has a keen and active interest in wildlife diseases. Cleve is a member of the Management Committee of the Australian Wildlife Health Network and also the State wildlife disease surveillance coordinator for the network.

#### **Appendix:**

# Protocol for dealing with wild bird deaths in Western Australia Avian Influenza (Bird flu)

Chief Veterinary Officer
Department of Agriculture and Food
3 March 2006

#### **Background**

One of the objectives of surveillance for exotic animal diseases such as highly pathogenic avian influenza (HPAI) is to give a greater level of confidence that the disease is not present in Australia, or alternatively, to detect the disease at theearliest possible time and trigger a response to control or eradicate the incursion.

Avian Influenza has generated enormous media attention because of the small number of human deaths that have resulted from infection with the H5N1 avian influenza virus and the theoretical risk of the virus mutating to cause a human influenza pandemic. These factors have resulted in public and industry fears about the risk of the H5N1 virus entering Australia in wild birds.

Whilst there are always risks associated with any disease, it is important in relation to the H5N1 virus to remember that

- Waterfowl are the natural hosts and normal spreaders of avian influenza.
- Australia is not on the flight path of any of the migratory waterfowl.
- Waders and shorebirds that do migrate from Asia to Australia can carry the avian influenza virus but are not efficient spreaders of the disease.
- There is little chance of migratory birds bringing avian influenza to Australia and infecting Australian birds or poultry.
- The international view is that it is not feasible to control avian influenza in wild bird
- populations and that control should not be attempted.
- The Department of Health considers migratory birds in Australia to be a negligible avian influenza risk to humans.
- The Department of Health advises that people working in the poultry and associated industries, should be vaccinated as a matter of course with the normal human flu vaccine before each influenza season that is, in about March or April each year.
- The human flu vaccine does not protect against infection with avian influenza virus. However, it is thought that vaccination for human flu will reduce the chance of bird flu viruses and human flu viruses combining to form a new strain of human virus.

#### Surveillance

General surveillance is the most effective and efficient way of detecting new diseases or incursions of exotic diseases that cause significant clinical signs and/or deaths. General surveillance relies heavily on networks that include people in the poultry and other bird industries, wildlife officers, bird watchers and members of the general public. All need to be alert for unusual disease events and promptly report such events to aveterinarian.

Respiratory distress and/or death in birds can be caused by a number of diseases, including diseases that occur naturally in Australia. The following guidelines are provided to assist organisations and institutions to target the investigation of unusual disease events in wild birds so as to demonstrate that it is not due to an exotic disease, or detect an exotic disease if it is present.

## Wild birds

A number of organisations, including DAWA, the Perth Zoo, the Department of Conservation and Land Management (CALM), Murdoch University, and local governments, as well as veterinary clinics and bird refuges receive calls about ill health and deaths in wild birds.

Wild birds may be grouped according to heir susceptibility to HPAI. Water fowl eg ducks, geese and swans, are the most likely to become infected with HPAI, followed by water birds eg waders, and then pigeons and parrots.

## Clinical signs of avian influenza include:

- noticed sick one day and dead the next
- depressed, weak and staggering
- watery eye and nasal discharge
- diarrhoea
- nervous signs such as muscle weakness, paralysis, head tilt, tremors

## DAWA veterinary investigation

Significant numbers of juvenile wild birds die due to natural causes. However, juvenile waterfowl are susceptible to avian influenza and a cluster of deaths in such birds can be a first indicator of the presence of pathogenic avian influenza.

A DAWA veterinarian should be contacted and investigate cases where:

- there is an unexplained cluster of deaths of 6 or more juvenile wild waterfowl, 3 or more adult waterfowl, 5 or more other types of water birds, 8 or more pigeons/parrots; and
- the deaths occur within three to five days or within one locality such as a lake.

Cases which meet the criteria should be reported to the Duty Pathologist at the Department of Agriculture and Food on 93683351 or the Emergency Disease Hotline on 1800675888.

## Handling and collecting dead birds for investigation

Dead birds that fit the criteria and are collected for investigation should be:

- fresh:
- double bagged i.e. sealed in a plastic bag and then sealed inside another plastic bag;
- kept cold until delivery eg placed in an esky with a cold brick;
- not put in a food fridge due to the risk of salmonella;
- delivered to a DAWA office after consulting the District Veterinary Officer or duty pathologist.

The esky should be thoroughly cleaned and disinfected after the birds are removed.

## Personal hygiene

- Be aware that normal personal hygiene measures such as hand washing are the most effective means of reducing the risk of infection.
- The Department of Health advises that because avian influenza has not been reported in Australia, people do not need to take any additional precautions.
- Use rubber or plastic gloves when inspecting and handling sick or dead birds.
- Use additional precautions such as rubber boots, overalls and a mask if there is suspicion of avian influenza or when working in an enclosed shed.
- Wash hands and equipment with soap or disinfectant.