

SEMESTER ONE 2007
INFECTION and IMMUNITY
GRADUATE ENTRY PROGRAMME
PARASITOLOGY PRACTICAL 6 – Dr TW Jones
POULTRY COCCIDIOSIS

Objectives

After this class I expect you to be able to :

1. Describe the principal ante and post mortem signs of poultry coccidiosis caused by *Eimeria tenella* infection
2. Describe how you would confirm a suspected case of fatal coccidiosis in poultry.
3. Recognise and describe the gross pathological changes characteristic of caecal coccidiosis in poultry.
4. Identify and distinguish between schizont, merozoite and oocysts stages of *Eimeria* in stained gut smear-preparations from poultry.
5. Identify and distinguish between schizont and merozoite stages of *Eimeria* in stained tissue sections from poultry.
6. Recognise and distinguish between the sporulated and unpopulated form of oocysts of *Eimeria tenella*

This class requires you to make observations on a variety of materials and to answer questions related to that material. The format and type of the question used in this class are typical of those that will be used in new format spot examination so it is important that you record your observations and answers a permanent way so that you can use them as a revision aid.

Activities

1. Watch the video on poultry coccidiosis and then answer the following questions
 - a. Which symptoms can be used as indicators of acute coccidiosis?
 - Behavioural changes
 - Blood in the faeces
 - b. Which form of poultry coccidiosis is shown in the video?
 - Caecal
 - c. Which other organ system can be affected in other forms of poultry Coccidiosis?
 - The intestine
 - d. What are the principal signs of coccidiosis seen at this post-mortem?
 - Distended, blood-filled caecae
 - e. What are the two types of stained preparations used to look for evidence of coccidian infection?
 - Tissue scrapes and impression smears
 - f. Which stages in the life cycle of *Eimeria* were found in the stained smear?
 - Schizonts, merozoites & oocysts
 - g. In the stained sections what are the names of the stages are associated with the earlier stages of infection?
 - Schizonts and merozoites

- h. Under which circumstances do you think you would use stained histological sections as routine diagnostic technique?
 - Research on poultry coccidiosis
 - Testing the efficacy of anti-coccidial agents
2. Now examine a stained tissue smear from an infected chicken under the microscope. You should be able to identify the following parasitic stages
 - a. Enlarged host gut cells containing multinucleate schizont stages appearing as oval blue bodies with dark purple spots within an enlarged host cell - you might also see the cell nucleus as a pink-purple body attached to the periphery of the host cell. – **what would these have developed into if they were still in the animals?**
 - Merozoites
 - b. Enlarged host gut cells containing merozoite stages appearing as spindle-shaped cells with a purple nucleus within an enlarged host cell – **what would have happened next in the host and what impact would this have on the host?**
 - The merozoites would have burst out of the host cell and then infected new cells
 - Large scale release of merozoites is the principal cause of disease
 - c. Free merozoites – **where do you think these have come from?**
 - Host cells containing merozoites that have burst during smear preparation
 - Merozoites already present in the lumen
 - d. Oocysts – appearing as clear, unstained oval bodies - **why are these unstained?**
 - The thick water-proof wall of the oocyst prevents the entry of aqueous stains such as Geimsa's stain
 - e. You should also look for the presence of non-parasitic structures in the smear including
 - Host gut cells – intact and damaged
 - Red blood cells – **how do these relate to other red blood cells that you have seen?**
 - Avian rbc's are nucleated
 - i. Gut bacteria – **which types can you see?**
 - A range of rods and cocci

3. Now examine under the microscope the stained tissue section of caecum from a chicken infected with *Eimeria tenella*. Because of the small size of the individual protozoa you will need to use the $\times 100$ oil immersion objective to see the fine details but you will be able to see other structures and groups at lower magnifications. As a general rule you should use the lower powered objectives first of all to find the area of interest and then switch to the highest power. Be careful to avoid getting oil on the lower powered objectives (10X & 40X). Also PLEASE wipe the oil from the slide once you have finished and return it to the tray.

Use the following table to help you identify the different stages in the tissue section. Not all stages will be present in every section, **as a minimum you must make sure that you can identify the schizont and merozoite stages in your section**

<i>Stage</i>	<i>Description</i>
Schizont	Found within a clear parasitophorous vacuole inside the host cell, range of sizes, dense blue-purple central mass of parasite nuclei
Merozoites,	Spindle-shaped structure found within a clear parasitophorous vacuole inside the host cell. Large number of merozoites per cell, sometimes seen in the lumen of the gut
Gametes	Macrogametes - easily visible as a single, round structure with large pink-orange peripheral granules within the host cell, single central nucleus Microgametes. – difficult to spot and less frequently encountered than the macrogametes, appear as mass of small, comma.-shaped dark blue bodies within the host cell
Zygotes	Oval pink bodies with a thin outer wall and a single central nucleus similar to macrogamete but peripheral granules absent.
Oocysts	Unstained oval bodies with a thick outer wall that does not stain, contents often lost during sectioning. In the gut cells or lumen

Now answer the following questions

- a. What kind of changes can you see to the structure of the gut and how do these relate to the ante and post mortem signs of poultry coccidiosis seen in the video?
 - Destruction of the villus structure related to mass emergence of merozoites from host cells – gives rise to haemorrhage into the lumen of the caecum
 - b. Which of the stages in the life cycle do you think are responsible for the damage seen to the gut wall?
 - The merozoite stages
 - c. Why do the schizont stages look different in the section when compared to their appearance in the stained smear?
 - In the stained smear the cells containing the schizonts have been compressed during the drying process so that the contents can be difficult to distinguish,
 - In sections the cells will have been cut into slices so that it is easy to see the contents but what you see depends on where the cell was cut
 - d. Why do you think that infected cells are usually found in groups rather than distributed throughout the gut?
 - Merozoites from one cell infect neighbouring cells rather than distant cells in the gut
4. Now look at the photograph of a sporulated and un-sporulated oocyst stage and answer the following questions
- a. What is the epidemiological significance of each of these forms of the oocyst

- In the case of *Eimeria tenella* unsporulated oocysts are not infective and have to spend 24-48 hours outside the host before they sporulate and are infective for other birds
- b. Which form would you expect to find in freshly collected faecal samples?
 - Unsporulated oocysts
- c. How reliable do you think faecal examination for the presence of oocysts is as a means of diagnosing acute coccidiosis?
 - Oocyst development occurs at 5-7 days after infection after the more pathogenic asexual phase involving merozoites

Stop and think

1. If you were in charge of a poultry unit, which diagnostic techniques would you consider using for
 - a. Routine health monitoring
 - Oocyst screening – won't detect acute coccidiosis but is the only non-invasive/lethal method that is readily available
 - b. Evaluating the effectiveness of control regimes
 - Oocyst screening with possible pm screening of slaughtered birds
 - c. Dealing with an outbreak of acute coccidiosis
 - PM examination
 - Oocyst contamination levels in the environment
2. Which other food animal species can be affected by coccidiosis?
 - Cattle, sheep & pigs (*Isospora* is a coccidian)
3. Which members of the coccidia which are zoonotic?
 - *Toxoplasma* and some species of *Sarcocystis*