Apicomplexa of Systemic Pathology

Toxoplasma, Neospora, Sarcocystis

Toxoplasma gondii
Feline coccidian of zoonotic importance

A. Morphology
- Oocyst
  - Small, sub-spherical, smooth coat, no polar cap, single embryo when passed
  - Sporulated oocyst contains 4 sporocysts with 2 sporozoites each = 8 sporozoites total
- Intracellular parasite of intestinal epithelium and various host tissues

B. Terminology Review as it relates to Toxoplasma gondii
- Definitive host – species in which the adult (or sexual) form of the parasite occurs
  - Felids are definitive hosts for T. gondii
- Facultative intermediate / paratenic host – species which supports the multiplication (asexual) stage of a parasite, can carry parasites through to definitive host, but are not required for life-cycle of the parasite
  - Any warm-blooded animal
- Accidental host – “dead end” host, parasites are not passed on and can’t complete life-cycle
  - Humans are accidental hosts when infected with T. gondii
- Endodyogeny – single division forming 2 daughter cells inside cytoplasm of a mother cell
  - Occurs in the cat and paratenic host (only replicative form in paratenic host)
- Schizogony – multi-nuclear division followed by a cytoplasmic division forming multiple daughter cells
  - Occurs only in the cat, intestinal epithelium
- Tachyzoite – fast growing, infective stage
  - Localize in neural and muscle cells, fast growing; will develop into bradyzoites
- Bradyzoite – slow growing, infective stage
  - Localize as cysts in tissue (neuron, skeletal mm); remains semidormant (chronic infections)

C. Life Cycle Rather Complex

1. Cat to Cat
   a. Direct Life Cycle (homoxenous) --- Definitive host – Felids only
   b. Transmission -- fecal-oral, ingestion of oocyst – felid hosts only
   c. Invasion -- Sporozoites excyst from oocyst and invade enterocyte; some sporozoites invade deep tissue (CNS, muscle, viscera) multiply and disseminate as tachyzoites, eventually forming bradyzoite cysts.
   d. Asexual reproduction (in intestinal cells)
      i. Endodyogeny and schizogony
      ii. Approximately 5 cycles of endodyogeny and / or schizogony
      iii. Causes none to mild pathology in the cat.
   e. Sexual reproduction (only occurs in the Felids)
      i. Final generation of “zoites” exit the enterocyte, infect other enterocytes, and go through gametogony (production of gametes)
      ii. Macrogamete (egg)
         1. Some final zoites remain a single cell and become a macrogamete (egg), within a macrogamont.
      iii. Microgametes (sperm)
         1. Other final zoites go through multi-nuclear division, cytoplasmic division, and develop 2 flagella (bi-flagellate) on each gamete; thus forming a microgamont
         2. Exflagellation – when microgametes exit the microgamont in search of a macrogamete.
iv. Fertilization – a microgamete fuses with a macrogamete forming a zygote
v. A cyst wall forms around the zygote and the immature oocyst exits the macrogamont into the lumen of the host’s gut and is passed in the feces.
f. Dissemination
   i. Oocysts (unsporulated) exit the host in the feces and contaminate the environment.
      1. Felids are the only hosts to pass oocysts (b/c of sexual fertilization).
      2. Prepatent period (oocyst ingestion to oocyst passing): 19 - 48 days
      3. Oocysts highly resistant and remain infectious for many months

2. Paratenic Host to Cat
   a. Facultative Indirect Life Cycle (heteroxenous)
   b. Paratenic host – any warm-blooded animal infected by ingesting sporulated oocysts from infected cat feces
   c. Transmission to cat
      i. Cat ingests of a tissue cyst from a paratenic host = carnivorism
   d. Bradyzoites released from tissue cysts develop into tachyzoites
   e. Invasion, Asexual Reproduction (intestinal epithelium), Sexual Reproduction (unsporulated oocysts); some sporozoites invade deep tissue
   f. Dissemination – unsporulated oocysts passed in feces, sporulate in environment
   g. Prepatent period (Ingest tissue cyst from paratenic host to passing oocysts) = 3 to 10 days vs the 19-48 days in the cat-to-cat cycle

3. Cat to Paratenic Host
   a. Facultative Indirect Life Cycle (heteroxenous) -- Paratenic host – Any warm blooded animal, including felids
   b. Transmission to paratenic host
      i. ingestion of an oocyst from the feces of a felid (the definitive host)
   c. Invasion -- “Zoites” enter intestinal cells and lymph cells and transform in to tachyzoites.
      i. Tachyzoites - rapidly dividing zoites via endodyogeny (2 daughter cells / division)
   d. Asexual reproduction – extra-intestinal cells
      i. Tachyzoites are dispersed throughout the body of the paratenic host
         1. The rapid destruction of host cells during the tachyzoite phase causes the acute/ severe disease
      ii. Eventually tachyzoites transform into bradyzoites
         1. Bradyzoites
            a. Slowly dividing zoites via endodyogeny (2 daughter cells / division)
            b. Stationary (= non-disseminating) and form tissue cysts.
         2. Tissue cysts
            a. Cause more chronic pathology in the paratenic (and human) hosts
            b. Occur in and cause damage to the brain, liver, striated muscles
            c. Remain viable for the life of the paratenic host.
   e. General Notes
      i. Tachyzoites may be transplacentally transmitted.
      ii. Thus mother ingestion of oocyst from cat feces – Toxoplasmosis in fetus

4. Paratenic Host to Paratenic Host
   a. Transmission
      i. ingestion of a tissue cyst from a paratenic host (any warm blooded animal) = carnivorism
   b. Invasion and Asexual reproduction is the same as “Cat to Paratenic Host” above
   c. General Notes
      i. Tachyzoites may be transplacentally transmitted.
      ii. Thus mother ingestion of raw meat with tissue cyst – Toxoplasmosis in fetus

D. Pathogenesis
   1. Intestinal phase in felids -- minimal.
   2. Systemic disease (extra-intestinal phase) in felids, paratenic hosts or humans – explosive multiplication of tachyzoites cause massive destruction of host cells, also acute immune response. Tissue cysts cause tissue damage, and a source for recrudescence of acute disease.
E. Clinical Disease (Cat only, see DZ in paratenic hosts below)
   1. Intestinal Disease – usually no complaint (oocyst found on routine fecal); some cats develop self-limiting small bowel diarrhea
   2. Systemic Disease -
a. Fever, anorexia, vomiting, diarrhea, myositis, uveitis, enlarged lymph nodes, pneumonia (especially for FIV+ cats), encephalitis, nephritis, death.

F. Diagnosis (Cat only, see DZ in paratenic hosts below)
1. Intestinal Disease – oocysts in feces – use Zinc Sulfate as oocysts may distort in other solutions
2. Systemic Disease – serologic tests (good to rule-out Toxoplasmosis if cat is seronegative)

G. Treatment (cats)
1. Pyrimethamine plus triple sulf drugs used against intestinal phase and acute phase.
2. Clindamycin and Ponazuril also used.
3. Clindamycin used in systemic disease
4. No good treatment for tissue cysts

H. Control (cats)
1. Sanitation
   a. Clean litter box daily (sporulation in 1-3 days).
   b. No raw meat for cats
   c. Don’t let cat outside to defecate or hunt

I. Non-felid paratenic hosts
1. Dogs -- Systemic toxoplasmosis, ingest oocyst from cat feces or tissue cyst from prey.
   a. Fever, respiratory signs
2. Sheep & Goats -- Systemic toxoplasmosis & Congenital toxoplasmosis, ingest oocyst from cat feces
   a. Systemic – CNS signs
   b. Congenital – abortion
   c. Toxovac S48 – vaccine
3. Cattle -- Congenital toxoplasmosis (abortion, but very rare), ingest oocyst from cat feces.
4. Horse -- Systemic toxoplasmosis, low pathology, ingest oocyst from cat feces
5. Rodents -- Systemic toxoplasmosis, ingest oocyst from cat feces or tissue cyst from prey.
   a. Decreased fear of cats
   b. Major source of infection for cats.
6. Swine -- Systemic toxoplasmosis, ingest oocyst from cat feces or tissue cyst from prey.
   a. Fever, respiratory signs
   b. Highly prevalent in free-range pigs
   c. Important source of infection for humans
7. Poultry -- Systemic toxoplasmosis, ingest oocyst from cat feces.
   a. Prevalent in free-range and back-yard chickens
   b. Important source of infection for humans

J. Zoonosis (Human infections)
1. Systemic Toxoplasmosis and Congenital Toxoplasmosis
2. Ingestion of oocysts from cat feces, unwashed vegetables, or unclean hands
3. Ingestion of tissue cyst from undercooked meat (mainly mutton, goat, free-range pork, free-range chicken)
4. Transplacental Transmission (Congenital Toxoplasmosis)
5. Immunocompetent (systemic toxoplasmosis)
   a. 1st exposure – “flu-like” illness that may last for weeks (Fever, myalgia, sore throat, lymphadenopathy)
   b. Future exposures -- immune-protected, no issues.
6. Immuno-deficient Adult (systemic toxoplasmosis)
   a. Elderly & those with Immunosuppressive diseases
   b. Severe disease (Respiratory, CNS, etc.)
   c. 1st exposure prior to immune-suppression, then recrudescence may cause severe DZ
   d. 1st or future exposure after immune-suppresssion may cause severe acute DZ
7. Immuno-deficient Fetus (Congenital toxoplasmosis)
   a. Severe DZ (congenital malformation, mental retardation, death)
   b. If mother’s 1st exposure occurs during pregnancy.
      i. Transplacental transmission is greatest in 3rd trimester.
      ii. But more severe defects if transplacental transmission occurs in the 1st Trimester.
   c. 1st exposure prior to pregnancy, then immune system controls toxoplasmosis, unless mother is immune-deficient. – can get antibody tested.
   d. Avoid cat feces, have someone else clean litter box daily (sporulation in 1-3 days)
Avoid uncooked meat, unclean hands, unclean vegetables, unclean knives and cutting boards.

**Neospora caninum**

**Canine coccidian causes bovine abortions and canine neurologic disease**

A. **Morphology**

- Oocyst
  - small, subspherical, smooth coat, no polar cap, single embryo when passed
  - Sporulated oocyst contains 2 sporocysts with 4 sporozoites each = 8 sporozoites total
- Intracellular parasite of intestinal epithelium and various host tissues.
- Form thick walled bradyzoite cysts in tissues

B. **Life Cycle Rather Complex**

1. **Intermediate Host to Dog**
   a. Obligate Indirect Life Cycle (heteroxenous)
      i. Definitive host – Canids only (Dog, Coyote, Wolf, etc.)
      ii. Intermediate Host – Cattle and other animals
   b. Transmission -- carnivorism, ingestion of tissue cysts
   c. Invasion -- Zoites excyst from tissue cyst and invade enterocyte
   d. Asexual reproduction (in intestinal cells – no pathology)
      i. Endodyogeny [single division forming 2 daughter cells] and
      ii. Schizogony [multi-nuclear division followed by cytoplasmic division forming multiple daughter cells]
   e. Sexual reproduction (only occurs in the Canids)
      i. Final generation of “zoites” exit the enterocyte, infect other enterocytes, and go through gametogony (production of gametes)
      ii. Macrogamete (egg)
         1. Some final zoites remain a single cell and become a macrogamete (egg), within a macrogamont.
      iii. Microgametes (sperm)
         1. Other final zoites go through multi-nuclear division, cytoplasmic division, and develop 2 flagella (bi-flagellate); thus forming a microgamont
         2. Exflagellation – when microgametes exit the microgamont in search of a macrogamete.
      iv. Fertilization – a microgamete fuses with a macrogamete forming a zygote
      v. A cyst wall forms around the zygote and the immature oocyst exits the macrogamont into the lumen of the host’s gut and is passed in the feces.
   f. Dissemination
      i. Oocysts (unsporulated) exit the host in the feces and contaminate the environment ~4 days post ingestion, then sporulate after 1-3 days
         1. Canids are the only hosts to pass oocysts.
         2. Oocysts highly resistant and remain infectious for many months.
   g. Transmission / Invasion in dog (details)
      i. Intestinal –some zoites go into intestinal epithelium to complete sexual cycle and generate oocysts in feces
      ii. Systemic – other zoites get into deeper tissue to complete asexual cycle, producing tachyzoites and bradyzoite tissue cysts (predilection for neural tissue = neurologic disease)
      iii. Congenital – in pregnant bitches, some tachyzoites move through placenta to puppies

2. **Dog to Intermediate Host**
   a. Transmission
      i. Ingestion of a sporulated oocyst from the feces of a canid (the definitive host)
   b. Invasion -- Zoites enter intestinal cells and lymph cells and transform into tachyzoites.
      i. Tachyzoites - rapidly dividing zoites via endodyogeny (2 daughter cells / division)
c. Asexual reproduction – extra-intestinal cells
   ii. Tachyzoites divide and are dispersed throughout the body of the intermediate host
       1. The rapid destruction of host cells during the tachyzoite phase causes the acute/severe disease
       2. Can be transplacentally transmitted to calf fetus
       3. Eventually tachyzoites transform into bradyzoites
   ii. Bradyzoites
       1. Slowly dividing zoites via endodyogeny (2 daughter cells/division)
       2. Stationary (= non-disseminating) and form tissue cysts.
       3. Tissue cysts
          a. cause more chronic pathology in the intermediate hosts
          b. occur in and cause damage to the CNS, lungs, heart, striated muscles, etc.
          c. remain viable for the life of the intermediate host.

d. Transmission / Invasion (details) – after ingestion of sporulated oocyst from dog feces
   iii. Intestinal
       1. No replication of invasion of neighboring enterocytes; penetrate intestinal lining
   iv. Systemic
       1. Zoites go to deeper tissue with production of tachyzoites and eventually bradyzoite tissue cysts (neural, muscle, myocardial, placenta, fetus)
   v. Congenital
       1. Some may go transplacental to calf
       2. trans-generational infection
          a. Cows infected in utero will in-turn infect their calves, without reinfection with sporulated oocyst from dog feces
          b. Seropositive calves are less likely to have abortions

3. Intermediate Host to Intermediate Host’s Offspring
   a. Congenital Neosporosis -- Transplacental transmission
   b. Transmission
      i. “Re-activated” bradyzoites invade the placenta and fetus
      ii. Tissue cyst of pregnant dog to in utero puppies
      iii. Tissue cyst of pregnant cow to in utero calf
   c. Trans-generational (cows)

4. Dog to Dog (ingestion of sporulated oocyst does not result in oocyst production)
   a. Ingestion of sporulated oocyst, parasite goes systemic or transplacental (as in intermediate hosts)
   b. Only ingestion of bradyzoite cysts will the parasite go into enterocytes, sexual reproduction and oocyst production

C. Pathogenesis
   1. Intestinal Phase in Canids – no pathology
   2. Systemic Disease (Extra-Intestinal Phase) in canids or intermediate hosts – Explosive multiplication of tachyzoites causes massive destruction of host cells, also acute immune response. Tissue cysts cause tissue damage and is source for transplacental transmission.

D. Clinical Disease
   1. Intestinal Neosporosis – no clinical signs - oocysts found on a routine fecal (Canids only)
   2. Systemic Neosporosis
      a. Dogs
         i. Multi-system DZ due to tachyzoites and bradyzoites throughout the body and the immune reaction and inflammation that follows
         ii. Neurologic signs, nodular dermatitis, pneumonia, urine incontinence, fecal incontinence, nephritis, myocarditis, polymyositis (inflammation and pain of muscles).
   3. Congenital Neosporosis
      a. Dogs
         i. Puppy litter-mates dying with signs of polyradiculitis (inflammation of the nerve roots, especially of the hind limbs)
         ii. Puppy with signs of paralysis of the rear limbs at 3-8 weeks post-birth
         iii. Puppy with flaccid hind limb paresis
      b. Cattle
         i. Abortions (10-20% of abortions caused by N. caninum)
1. Abortions usually occur with 1st post-infection pregnancy; autolysis of fetus
2. Later pregnancies usually go to term, but calves are infected (maintenance of dz in herds)
   ii. Trans-generational infections (Seropositive calves give birth to seropositive calves –without reinfection via sporulated oocyst ingestion)
   iii. Decreased milk production and decreased weight gain

E. Diagnosis
   1. Dogs
      a. Puppy – classic flaccid hind limb.
      b. Serology, molecular tests
      c. Organism on biopsy, necropsy of litter mates
   2. Cattle
      a. Diagnostic arrays, immunological and molecular tests
      b. Antibody tests for whole milk

F. Treatment
   1. Dogs
      a. No drugs available to kill tissue forms.
   2. Cattle
      a. No drug therapy available
      b. Treatment of lactating cows is problematic (check withdrawal times)

G. Epidemiology, Control
   1. Epidemiology: worldwide
   2. control wild and domestic canid populations
   3. cull seropositive cows

H. Zoonosis --- Is not zoonotic

Sarcocystis cruzi
Canine coccidian of bovine deaths

A. Morphology
   • Sporocyst
     o Thin-walled oocyst sporulates and ruptures before exiting in the feces, thus sporocysts are seen in the feces
     o small, oval, smooth coat, no polar cap; 4 sporozoites

B. Life Cycle Rather Complex
   1. Cattle infects Dog
      a. Obligatory Indirect Life Cycle (heteroxenous) --- Definitive host – Dog
      b. Transmission -- carnivorism, dog ingests sarcocyst in cattle muscle
      c. Invasion -- Bradyzoites from sarcocyst from muscle invades intestinal cells
      d. (Asexual reproduction – none occurs in the dog.)
      e. Sexual reproduction (only occurs in Dogs)
         i. Zoites go through gametogony (production of gametes)
         ii. Macrogamete (egg)
            1. Some final zoites remain a single cell and become a macrogamete (egg), within a macrogamont.
            iii. Microgametes (sperm)
               1. Other final zoites go through multi-nuclear division, cytoplasmic division, and develop 2 flagella (bi-flagellate); thus forming a microgamont
               2. Exflagellation – when microgametes exit the microgamont in search of a macrogamete.
         iv. Fertilization – a microgamete fuses with a macrogamete forming a zygote
         v. A cyst wall forms around the zygote and the immature oocyst exits the macrogamont into the lumen of the host’s gut
         vi. The oocyst sporulates within the gut lumen, then ruptures, releasing its 2 sporocysts into the gut lumen.
f. Dissemination
   i. Sporocysts exit the host in the feces and contaminate the environment.
      1. Dogs are the only hosts to pass sporocysts.
      2. Sporocysts are infectious when shed, are very resistant and remain infectious for
         several months if kept cool and moist.

2. Dog to Cattle
   a. Intermediate host – Cattle
   b. Transmission
      i. Cattle ingests a sporocyst from the feces of a dog (the definitive host)
   c. Invasion -- Sporozoites enter vascular endothelial cells
   d. Asexual reproduction – vascular endothelial cells and muscle cells
      i. Zoites go through a few cycles of schizogony, then disperse throughout the body to muscle
         cells.
         1. The destruction of host cells during the tachyzoite phase causes the acute/ severe
            disease with immune reaction and inflammation.
      ii. Bradyzoites go through schizogony
         1. Sarcocysts (muscle cysts), full of bradyzoites develop within muscle cells and remain
            viable for the life of the intermediate host.

C. Pathogenesis
   1. Intestinal Phase in Canids – no pathology
   2. Systemic Disease (Endothelial and Muscle Phase) in cattle – Zoites cause destruction of host cells,
      also acute immune response.
   3. Multisystemic dz due to zoites throughout the vascular endothelium and muscles, w/ immune
      reaction and inflammation

D. Clinical Disease
   1. Intestinal Sarcocystosis – no pathology in the dog
   2. Immune status of the host and the dose of sporocysts important factors of development of clinical dz
   3. Systemic Sarcocystosis -- cattle only (4-6 weeks post ingestion of sporocyst)
      i. Protracted fever, lymphadenopathy, anorexia, cachexia, muscle spasms, myositis, hyper-
         excitability, diarrhea, hyper-salivation, weakness, hair loss around eyes, neck and tail switch,
      ii. Pregnant cows – abortions, still births
      iii. Condemnation of carcass at inspection due to Sarcocysts

E. Diagnosis
   1. Dogs – sporocysts found on a routine fecal exam
   2. Cattle – serology, necropsy

F. Treatment
   1. Dogs
      a. No drugs available to kill tissue forms.
   2. Cattle
      a. Amprolium may provide some prophylactic protection.
      b. Treatment against sarcocysts is ineffective

G. Epidemiology and Control
   1. Distributed worldwide
   2. Control wild and domestic canid populations
   3. don’t let dogs have access to raw meat, offal or dead animals

H. Zoonosis --- Is not zoonotic

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**Sarcocystis neurona**  
Opossum coccidian of horse neuropathy (Equine Protozoal Myeloencephalitis –EPM)

A. Morphology  
- Sporocyst (not oocyst in opossum feces)  
  - Thin walled oocyst sporulates and rupture before exiting in the feces, thus sporocysts are seen in the feces  
  - Small, oval, smooth coat, no polar cap; 4 sporozoites

B. Life Cycle  
1. Sylvatic Life Cycle  
   a. Obligatory Indirect Life Cycle (heteroxenous)  
      i. Opossum -- definitive host; sexual / intestinal stages, passes sporocysts  
      ii. Other small mammals and birds – intermediate host (IH), asexual / muscle stages, sarcocysts in muscles  
      iii. IH ingests sporocysts form opossum feces and opossum ingests sarcocyst in bird muscle.  
2. Accidental Host -- Aberrant, dead-end host  
   a. Horse  
      i. Ingests sporocyst from opossum feces (contaminated food or water)  
      ii. Asexual stages / systemic stages  
      iii. Organism disseminates throughout body, especially to neural tissue, and muscle tissue

C. Pathogenesis  
1. Systemic Disease but with a predilection for neural tissue (neurons and leukocytes of the brain and spinal cord) in horses – Zoites cause destruction of host cells, and acute immune response / inflammation.

D. Clinical Disease  
1. Equine Protozoal Myeloencephalitis (EPM)  
   a. Spinal Cord involvement  
      i. Gait abnormalities, unilateral muscle atrophy (gluteal), myopathy, asymmetric weakness, and ataxia  
      ii. Demarcated spontaneous sweating, loss of reflexes, cutaneous hyper-sensation.  
   b. Cranial nerve involvement  
      i. Seizures, visual deficits, behavioral abnormalities  
   c. Brain involvement  
      i. Depression, head tilt, facial paralysis, muscle atrophy (masseter), dysphagia  
   d. Without treatment, may progress to recumbency and death

E. Diagnosis  
1. Horses  
   a. Observation of Clinical Signs, mostly neurologic and muscle atrophy.  
      i. Unfortunately there is a broad spectrum of disease agents that induce similar clinical signs  
   b. Serology  
      i. Serum IgG against *S. neurona* indicates exposure; may also give a false-positive due to cross-reactivity to another non-pathogenic *Sarcocystis* species  
      ii. Seropositive + neurological signs strongly supportive of EPM  
      iii. Paired serology testing with CSF and serum more predictive of active infection  
      iv. Western Blotting and ELISAs use *S. neurona* specific antigens (less cross-reactivity)  
      v. IFA uses whole cell antigen (more cross-reactivity)  
      vi. Parasite antigen or antibody in spinal fluid is diagnostic.  
   c. PCR detects *S. neurona* DNA (= active infection).  
   d. Post-mortem demonstration of organism in CNS lesions.
F. Treatment
   1. Horses
      a. Ponazuril, didazuril, pyrimethamine, sulfadiazine.
      b. Long treatment period. Be vigilant of side-effects.
      c. Improvement in 60-70%, complete recovery in up to 20%, relapse in 20%.

G. Epidemiology
   a. no breed predilections
   b. confined to the Americas (range of opossum distribution)
   c. Estimated ~50% of horses have been exposed to S. neurona but <1% develop EPM

H. Control
   1. Prevent access of opossums to horse-feeding / watering areas
   2. Prevent access of opossums to stored horse feed
   3. Fallen fruit should be removed from horse pastures. Why?

I. Zoonosis --- Is not zoonotic

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