Coronaviridae

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Glossary

Viruses of this family are enveloped with a positive-sense, single-stranded, linear RNA molecule as genome. The term "corona" refers to the halo of spikes extending outwards from the envelope. These viruses infect the respiratory, gastrointestinal tracts and the CNS of many mammals, including humans, and birds.

Viral Characteristics
- Virions are enveloped (80 - 120 nm in diameter), with club-shaped surface spikes (about 20 nm from the envelope surface) that give the appearance of a crown (see Fig. 24.1).
- The nucleocapsid has helical symmetry. This feature is unique to the coronaviruses, as most positive-sense RNA viruses have icosahedral nucleocapsids.
- The spike protein is associated with attachment to target cells, which is usually species specific, and is antigenic.
- Coronaviruses replicate in the cytoplasm and bud into cytoplasmic vesicles from which the virions obtain the envelope.
- Coronaviruses have the largest genome of any RNA virus (26 - 32 kb in size).
- The genome is positive-sense ssRNA that is nonsegmented. The genome has a 5’ cap and a 3’ polyadenylated (poly A) tail.
- Coronaviruses have a high frequency of mutation and a high frequency of recombination, resulting in rapid strain formation within an individual.
- In the cytoplasm, the genomic RNA is copied to a complementary negative-sense RNA strand. This is used as the template for more genomically positive-sense RNA strands and for the formation of viral mRNAs of various sizes (all have a common 3’ end), known as subgenomic RNAs. The production of subgenomic RNAs is characteristic of coronaviruses.
- These viruses can be propagated in cell culture but often with difficulty.
- They are labile in the environment.
Classification
The family Coronaviridae has two genera, Coronavirus and Torovirus. The genus Coronavirus is divided into three groups on the basis of several features, including the presence or absence of a hemagglutinin-esterase (HE) protein and the number and arrangement of non-essential genes.

Important viruses in these genera are as follows:

**Coronavirus**
- Group 1
  - Porcine transmissible gastroenteritis virus
  - Porcine epidemic diarrhea virus
  - Feline infectious peritonitis virus
  - Canine coronavirus
- Group 2
  - Bovine coronavirus
  - Porcine hemagglutinating encephalomyelitis virus
- Group 3
  - Infectious bronchitis virus
  - Turkey coronavirus

**Torovirus**
- Bovine torovirus (Breda virus-Iowa): Associated with severe diarrhea in neonatal calves.
- Equine torovirus: Isolated from a horse with diarrhea in Switzerland, although it was probably not the etiological agent responsible for the disease. The isolate was antigenically related to bovine torovirus.

Coronaviruses cause human respiratory infections, including the common cold and recently a disease called severe acute respiratory syndrome (SARS) that was first recognized in Asia in 2003. It spread to a number of countries in Europe, the Americas and Asia but was finally contained by strict control measures. The fatality rate was ~ 3%. The virus is genetically and antigenically different from other previous known animal or human coronaviruses and has not yet been assigned to a genus.

**Coronavirus Transmissible Gastroenteritis**

**Cause**
Transmissible gastroenteritis virus. There is only one antigenic type. The virus is serologically related to porcine respiratory coronavirus, canine coronavirus, and feline infectious peritonitis virus. The virus may remain viable on premises for up to three days.

**Occurrence**
Although transmissible gastroenteritis (TGE) virus only causes serious disease in pigs; it can infect dogs subclinically. The disease, which is highly contagious and destructive, occurs frequently in swine worldwide. The majority of outbreaks occur during the colder months of the year.

**Transmission**
The virus is present in feces and nasal secretions and may also be present in the milk of infected sows. Spread is by direct and indirect contact.

**Clinical & Pathologic Features**
The severity of the disease depends to a large extent on the level of the sow’s immunity. In previously unexposed swine herds, TGE is highly fatal to pigs less than 10 days old and usually spreads rapidly through the whole herd. Young pigs have a severe diarrhea with a watery, whitish or whitish-green stool. Vomiting is fairly common. Dehydration is especially marked, and deaths occur in 2 - 5 days after the onset of clinical signs.
The TGE virus selectively multiplies and destroys absorptive epithelial cells of the villi, giving rise to villous atrophy and impaired absorption (malabsorption). The disease in adult animals may include elevated temperature, poor appetite, mild diarrhea and depression. Vomiting may also occur in some animals. In herds where the virus is endemic, due to pre-existing immunity in most individuals, clinical signs are milder and mortality is relatively low. Clinical signs are often seen in these pigs during the post-weaning period when passive immunity has declined. Although clinical signs of respiratory infection are not common, the virus can be recovered from lung tissue. The virus has been shown to persist in the intestine of pigs for extended periods of time.

**Diagnosis**
- Clinical specimens: Portions of jejunum and ileum with contents.
- Because various other agents cause clinically similar gastroenteritis, confirmation of diagnosis by laboratory means is recommended.
- The laboratory method most often used to diagnose TGE is the fluorescent antibody examination of cryostat sections or scrapings of affected intestine.
- The virus can be cultivated in cell cultures of swine origin, but may produce little or no discernible cytopathology.

**Prevention**
- Both live attenuated and killed vaccines are available for the immunization of sows prior to farrowing. Their value seems to depend on their capacity to produce colostral and colostrum-derived immunity.
- Stronger and long lasting immunity is achieved via natural infection, and one empirical procedure has been to feed infectious intestine and feces to pregnant sows about a month before farrowing.
- Application of strict sanitary measures to prevent spread to susceptible swine.

**Porcine Epidemic Diarrhea**

**Cause**
Porcine epidemic diarrhea virus.

**Occurrence**
Porcine epidemic diarrhea virus affects swine in Asia and Europe, but not in the Americas.

**Transmission**
Infection is by the oral or nasal route. Spread is by direct or indirect contact.

**Clinical & Pathologic Features**
The virus infects the epithelial cells of the villi of the small intestine, causing histopathologic lesions similar to but milder than those of TGE.
The incubation period is about 1 - 4 days. The principal clinical sign is watery diarrhea and all ages of pigs may be affected. The clinical disease is somewhat similar to TGE but differs in that spread of the virus throughout the herd is much slower, vomiting is not a predominant clinical sign, and mortality is lower. In addition, older pigs are occasionally more severely affected than younger ones.

**Diagnosis**
- Clinical specimens: Portions of small intestine with contents.
- The virus can be propagated in cell cultures if trypsin treatment (to cleave the fusion protein enabling the virus to infect the next cell) of inoculum is utilized between transfers.
- Diagnosis is usually accomplished by fluorescent antibody examination of cryostat sections of affected intestine or by immune electron microscopic examination of feces or intestinal contents.
- Paired serum samples are tested for antibodies with an ELISA or an indirect immunofluorescence assay.

**Prevention**
- Vaccines are not available.
- Strict sanitary measures to prevent spread, along with deliberate infection of pregnant sows have reduced losses.

**Feline Infectious Peritonitis**

**Cause**
Feline coronavirus.

**Occurrence**
This frequently occurring, widespread disease affects domestic and wild felidae. The morbidity rate is low, and although a number of cats in a household or colony may be affected, most cases are sporadic.
Transmission
The virus is present in blood and exudates of infected animals. Transmission requires close contact with carrier or clinically infected cats, and is thought to occur by the oral route and through inhalation of aerosol droplets.

Pathogenesis
Infection is thought to begin in the intestinal epithelium and regional lymph nodes with spread to target organs via infected macrophages. Development of disease depends on the degree and nature of pre-existing immune responses. Two forms of the disease, "wet" and "dry", are described but a definite distinction between them cannot always be made. It is suggested that the cell-mediated response accounts for the dry or non-effusive form, while the wet or effusive form develops in the absence of a sufficient cell-mediated response. Antigen-antibody complexes are considered responsible for the lesions in the wet form.

Clinical & Pathologic Features
Feline infectious peritonitis (FIP) is a progressive debilitating febrile disease affecting cats of all ages, although most cases occur in cats six months to two years of age. The incubation period may be as short as two weeks or as long as several months. In the wet or effusive form, the initial clinical signs consist of anorexia, high temperature, and depression followed by progressive emaciation. The abdomen is often enlarged as a result of the accumulation of fibrinous fluid. There is a pyogranulomatous vasculitis due to precipitation of immune complexes associated with serous membranes. Dyspnea is a common sign when fluid accumulates in the thoracic cavity. In the dry form, there is little or no fluid buildup and a febrile response may be the only initial overt sign of infection. Ocular involvement (anterior uveitis) and CNS dysfunction are more commonly seen with the dry form. Both forms are generally fatal, but the wet form proceeds much more rapidly. Experimentally, a more rapid and severe development of clinical disease (accelerated FIP) occurs in cats with preexisting FIP antibody. This form has been attributed to the enhancement of viral infection mediated by Fc receptors present in macrophages ("antibody-dependent enhancement", ADE).

There is considerable variation in the virulence of viral isolates, and most cats with serologic evidence of FIP never develop the disease. However, a confirmed diagnosis is, with rare exception, eventually fatal. Most cats with clinical FIP die several weeks to months after diagnosis. The very closely related feline enteric coronavirus (FeCV) is endemic in cats in which it may cause a mild enteric infection. Evidence suggest that virulent FIP viruses arise as mutants of FeCV. The FIP virus is also closely related to transmissible gastroenteritis virus of swine and canine coronavirus.

Diagnosis
- Clinical specimens: Abdominal and thoracic fluids, lung, kidney, liver, spleen and brain.
- The disease is often diagnosed by gross and histologic examination. In the wet form, there are varying quantities of a characteristic straw-red fluid with fibrin in the abdominal and/or thoracic cavities and fibrinous adhesions are present on visceral organs, particularly the spleen and liver. Multifocal grayish-white granulomatous-like lesions are usually noted on the surfaces of various organs. These granulomatous-like lesions are usually larger and penetrate the tissue more deeply in cats with the dry form of FIP. Fibrinonecrosis and pyogranulomas are observed upon histological examination. The dry form is difficult to diagnose clinically. Lesions most commonly affect the eyes, brain, liver and kidneys. X-ray or ultra sound can be used to detect small amounts of fluid in the chest or abdominal cavities.
- Virus infected cells can be demonstrated in cryostat sections of affected tissues by immunofluorescence. Infected cells are usually restricted to or near the surface of affected tissues, and in and around the granulomatous lesions. Failure to sample appropriate areas may result in false negative results.
- Ante-mortem diagnosis is difficult and particularly so in the dry form. Hyperproteinemia is suggestive. Positive immunofluorescence results on cells collected from effusions are definitive.
- Serologic test (ELISA, indirect immunofluorescence assay) results are considered of little value in diagnosis, although it is claimed by some that high titers are suggestive of FIP.

Prevention
- Vaccines are available but their value according to many practitioners is questionable. They are probably of no value in cats already exposed.
- Disinfection of premises and isolation of seropositive cats. Only admit seronegative cats.

Canine Coronavirus Infection

Cause
Canine coronavirus. Because the virus can be recovered from many normal dogs, some investigators have questioned its
Pathogenicity. Most dogs from shows and in kennels have antibodies.

**Occurrence**
Canine coronavirus is worldwide in distribution. The virus is highly contagious, affecting dogs of all ages, but the disease in puppies is more severe.

**Transmission**
Virus is shed in the feces and infection occurs mainly by ingestion. Transmission is by direct and indirect contact.

**Clinical & Pathologic Features**
Initially, clinical signs are anorexia, depression, and loose stools, followed by vomiting and diarrhea. Feces often contain mucus (seldom blood) and have a fetid odor. Affected puppies may become dehydrated. Asymptomatic infections are common, especially in older dogs. Recovery usually occurs in 1 - 2 weeks and fatal infections in uncomplicated cases are rare. Gross necropsy lesions are those of a nonspecific enteritis; microscopic lesions consist of atrophy and fusion of intestinal villi.

**Diagnosis**
- Clinical specimens: Fresh feces and intestine.
- Diagnosis is best accomplished by the electron microscopic demonstration of coronavirus in feces or by the fluorescent antibody staining of cryostat sections of intestine.
- The virus, which is antigenically related to transmissible gastroenteritis of pigs and feline infectious peritonitis, can be isolated in cell cultures of canine origin.
- Histopathologic lesions of atrophy and fusion of intestinal villi are suggestive.

**Prevention**
- Inactivated vaccines are available, usually in combination with other viral and bacterial agents. Puppies are generally vaccinated twice during the first 1 - 2 months of life and thereafter on a yearly basis.
- Sound sanitation and the use of effective disinfectants (sodium hypochlorite) help control the disease in kennels.

**Feline Coronavirus Infection**
Feline coronavirus causes subclinical infections in cats and mild gastroenteritis in kittens. Feline infectious peritonitis virus is considered to have derived from strains of feline coronavirus.

**Bovine Coronavirus Infection**

**Cause**
Bovine coronavirus.

**Occurrence**
Bovine coronavirus infection occurs in cattle throughout the world.

**Transmission**
Virus is shed in the feces. Spread is by direct and indirect contact. The mode of infection is ingestion. Dams may carry the viruses and infect neonatal calves.

**Clinical & Pathologic Features**
Coronavirus affects calves in the first three weeks of life. Another virus of bovine neonates, rotavirus, is seldom found in calves over 7 days of age. Both viruses produce disease characterized clinically by profuse, watery diarrhea of sudden onset leading to extreme dehydration. Morbidity is often near 100% and mortality may range from none to >50%. Damage to the intestinal mucosa including loss of villous absorptive cells results in malabsorption. Bovine coronavirus has also been incriminated as a cause of "winter dysentery" in adult cattle.

**Diagnosis**
- Clinical specimens: Fresh feces and segments of intestine including spiral colon, ileum, and jejunum.
- There are several convenient and rapid methods used to diagnose rotavirus and coronavirus infections, including the electron microscopic examination of distilled water lysates of feces and fluorescent antibody staining of fecal smears and frozen sections of intestine.
- Commercial kits (ELISA and Latex agglutination) are available commercially for detection of rotavirus.
- Both rotaviruses and coronaviruses can be cultivated in cell cultures of bovine origin but often with some difficulty.

**Prevention**
- Commercial vaccines are available for bovine rotavirus and coronavirus but their efficacy is questionable. These may
be administered to heifers or cows before mating.

- Sound management practices including cleaning, disinfection and the use of footbaths, and provision of colostrum.

**Porcine Hemagglutinating Encephalomyelitis Virus Infection**
*(Vomiting and Wasting Disease)*

**Cause**
Porcine hemagglutinating encephalomyelitis virus. Only one serotype has been identified.

**Occurrence**
Porcine hemagglutinating encephalomyelitis virus (PHEV) infection is widespread in North America and Europe.

**Transmission**
Spread is by contact and aerosol droplets.

**Pathogenesis**
The virus first replicates in the tonsils and epithelial cells of the upper respiratory tract, followed by spread via peripheral nerves to the central nervous system. Damage to the vagal ganglion results in gastric irregularities including vomiting.

**Clinical & Pathologic Features**
Swine less than two weeks old are more susceptible, but most infections are subclinical. Clinical disease that occurs in some pigs consists of anorexia, rapid loss of weight, and depression. Vomiting and constipation may follow. Neurologic signs, such as muscle tremors, incoordination, and paddling movements, may develop later in the course of the disease. Mortality rate may reach 100% in baby pigs.

**Diagnosis**
- Clinical specimens: Tonsil, lungs, stomach, small intestine, brain, spinal cord, and acute and convalescent sera.
- Clinically PHEVI may resemble some of the neonatal diseases of pigs, such as TGE, colibacillosis, and clostridial enterotoxemia; the absence of diarrhea and the irregularity of vomiting may help in differentiation. The neurologic signs may be confused with pseudorabies, hog cholera, erysipelas and salt poisoning.
- Laboratory diagnosis is required to confirm PHEV infection. This is most conveniently accomplished by fluorescent antibody staining of frozen sections of tissue (brainstem) and virus isolation.
- The virus can be isolated in early passage of porcine cell cultures in which it produces syncytia.
- Hemadsorption can be demonstrated on infected cells. The virus agglutinates chicken, sheep, rabbit, rat, mouse, and hamster erythrocytes.
- Histopathologic examination of brainstem may reveal non-suppurative encephalitis.

**Prevention**
- The virus is widespread and most infections are subclinical.
- There is no vaccine available and no control measures are practiced.

**Infectious Bronchitis**

**Cause**
Infectious bronchitis virus. Neutralization tests in chicken embryos have shown there are many antigenic types of the virus. They are not related antigenically to other coronavirus species.

**Occurrence**
This highly contagious disease of chickens occurs worldwide.

**Transmission**
The virus is present in respiratory discharges and transmission is by direct and indirect contact and aerosol.

**Clinical & Pathologic Features**
Infectious bronchitis is a highly contagious disease of sudden onset and high morbidity. The disease is most severe in chicks and young birds; older birds are susceptible, although the disease is mild. Mortality may be high in baby chicks infected with nephrotropic strains.

The cardinal clinical signs are coughing and gasping. Changes include cloudiness of the air sacs, exudative bronchitis, and excess serous or catarrhal exudate in the trachea. Principal loss in affected flocks is the lowered egg production. The egg-laying capacity of survivors may be permanently impaired; eggs may be misshapen, rough, and soft-shelled. Some strains of the virus are nephrotropic and cause interstitial nephritis with sudden death.
Diagnosis

- Clinical specimens: Trachea, lungs, and kidneys.
- The virus can be cultivated in susceptible chicken embryos and in chicken epithelial cell cultures. Serotype identification is accomplished by virus neutralization tests with specific antisera.
- The changes occurring in the inoculated embryos are usually seen after several passages. They are characterized by death or dwarfing, curling of the embryo, and crystal urate deposits in the meso-nephrons.
- Fluorescent antibody tests on tracheal scrapings from infected birds have been used for rapid diagnosis.

Prevention

- Vaccination is practiced widely. A live attenuated virus is usually administered to birds at 1 - 2 weeks of age via drinking water with revaccination 3 - 4 weeks later, often with a killed vaccine injected subcutaneously. Since there are numerous types of virus, the vaccine used should include the appropriate type(s) for a given area.

Coronal Enteritis of Turkeys
(Blue comb disease)

Cause
Turkey coronavirus.

Occurrence
This highly infectious disease of turkeys occurs widely.

Transmission
The virus is shed in the feces and spread is by direct and indirect contact.

Clinical & Pathologic Features
The onset is sudden with anorexia, diarrhea, and marked dehydration. Sick birds may show darkening of the head. The mortality rate varies depending on the age and may approach 100% in young poults. Catarrhal enteritis with villous atrophy is the principal lesion seen in necropsied birds.

Diagnosis

- Clinical specimens: Feces and intestine.
- Diagnosis is usually based on clinical signs and gross and microscopic lesions.
- Electron microscopic examination (negative staining) of intestinal contents and fluorescent antibody examination of frozen sections of intestine provide for rapid diagnosis.
- The virus can be propagated in embryonated turkey eggs.

Prevention

- Affected birds and those that have recovered should be isolated. Strict sanitary measures are important in reducing losses.
- Antimicrobial drugs may be indicated to treat secondary bacterial infections.
- Vaccines are not available.

Glossary

Hemagglutinin-esterase (HE) protein: This protein causes hemagglutination of red blood cells and may also initiate binding to target cells.
Polyadenylation: This is the addition of tracts of adenosine polymers to the 3N ends of messenger RNAs in eukaryotic cells.

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