The FIP Jigsaw-Puzzle

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Feline infectious peritonitis is a systemic disease caused by a mutated form of the feline enteric coronavirus (FCoV). Although the prevalence of enteric coronavirus infection is high, around 90% in catteries, only 5% of these cats, at the most, will go on to develop FIP.1

The pathogenesis of FIP is related to an aberrant immunological and inflammatory response to this virulent mutated virus (FIPV). There are two forms of FIP. The “wet” form occurs as a result of inflammation of serosal surfaces (e.g. pleura, peritoneum) and is associated with the presence of body cavity effusions. The “dry” form is characterised by granulomatous lesions in organs like the kidney, intestines, abdominal lymph nodes, liver, eyes and CNS.2 The wet form is more common, with ascites usually present.3 The wet and dry forms represent two extremes of one disease, and patients may present with clinical signs and lesions anywhere on the continuum between them. FIP is invariably fatal.

There is unfortunately no simple definitive test for FIP. The diagnosis of FIP can be compared to making a jigsaw puzzle, where various puzzle pieces need to be fitted together to complete the picture.

The important corner pieces

Signalment: Although FIP can occur in cats in of any age, 50-70% of FIP cases occur in cats under one year of age.

Effusion: Ascites, thoracic and/or pericardial effusion characterise the wet form of FIP. It should be kept in mind however that less than half of effusions in cats are caused by FIP, and it is the characteristics of the effusion, not the mere presence, that make up this important piece of the FIP puzzle.1

Results from effusion analysis, in particular total protein, albumin/globulin (A/G) ratio and PCR, have a higher diagnostic value than tests performed on blood.5 The following are typical for an FIP effusion:
Effusion: Immunohistochemical staining can be definitive proof of FIP. 

- Tissue sections: FIP lesions exhibit a typical histopathological pattern, which is considered to be the gold standard test. The demonstration of FCoV in organ biopsies using immunohistochemistry is also 100% predictive for FIP. These techniques involve invasive sampling but are the only way to confirm FIP in animals without effusions. False negatives may occur.

- Rivalta test: This is a simple, inexpensive test that is useful to demonstrate a high content of inflammatory proteins in the effusion. A transparent tube of 10-20 mL is filled with 7-8 mL of distilled water and a drop of 98% acetic acid is added and mixed well in order to acidify the solution. A drop of effusion is placed carefully onto the surface of the mixture. If this drop disappears and the mixture remains clear, the result is negative. If the drop remains formed and slowly sinks to the bottom of the mixture like a jellyfish, the test is positive. A negative result has been found to have a high predictive value for the absence of FIP. A positive result could indicate FIP, but may also be seen in effusions due to lymphoma or bacterial infection.6

- Serum albumin: globulin ratio: The serum A/G has a higher diagnostic value than serum total protein or gamma globulin concentrations. Cats with an A/G ratio >0.8 are highly unlikely to have FIP, cats with an A/G ratio <0.6 are highly likely to have FIP.4

- Hyperbilirubinaemia: This is a common finding in FIP cats and FIP is the most common cause of this change in cats under 3 years of age.2,4 Usually there are no concurrent increases in liver enzyme activity. The increase in bilirubin may result in icterus.

PCR in effusions and tissues: Detection of the FCoV virus in effusion macrophages or in tissue sections (dry form) using immunohistochemistry provides definitive proof of FIP. 

- Effusion: Immunohistochemical staining can be used to demonstrate the presence of the virus within macrophages in the fluid and should be performed on all effusions fitting the criteria for FIP. The sample required is at least 12 mL of effusion fluid in EDTA tubes (i.e. at least three filled 4 mL EDTA collection tubes), which should be submitted as soon as possible. (This test is performed in the Pathology Laboratory at the Faculty of Veterinary Science in South Africa, and commercial laboratories can forward samples to them.) Positive results are 100% predictive for the presence of FIP, however a negative result does not rule out FIP (the negative predictive value is only 57%). This is because false negatives can occur due to insufficient numbers of macrophages in the sample examined.6

- Pieces that complete the picture, particularly in the dry form, where the ‘effusion’ piece is not present

Clinical signs:3

- Wet form: Moderate pyrexia, distended abdomen, dyspnoea
- Dry form: Variable and non-specific but include moderate refractory pyrexia, weight loss, lethargy, intraocular changes (uveitis), neurological signs and enlarged abdominal lymph nodes.

Haematology: Anaemia (both regenerative and non-regenerative) is present in about 50% of cases. A recent study found microcytosis in one third of cats with FIP, 40% of which were not anaemic.4 This suggests that microcytosis in a non-anaemic cat can increase the suspicion of FIP if other pieces of the puzzle are also present. Lymphoedema is a common finding.2

- The confusing or unhelpful “blue sky” pieces

Coronavirus antibody titres: The antibodies detected in these tests are against feline coronavirus and are NOT specific for FIP. The vast majority of cats with FCoV titres do not have FIP. More cats have probably been euthanased based on a positive anti-

- Cell count: Low (<2x10⁹ /L) with the cell population consisting of a mixture of non-degenerate neutrophils and macrophages with lower numbers of lymphocytes

- Protein: High (>35 g/L) due to the presence of gamma globulins. If possible, the albumin concentration of the fluid should be determined (this can be performed on a bench-top analyser, do not attempt if the fluid is very viscous and thick). The globulin fraction is calculated by subtracting the albumin from the total protein concentration. The A/G ratio can then be calculated. An A/G ratio <0.4 has a high predictive value for the presence of FIP and a ratio of >0.8 a high predictive value for the absence of FIP.3

- Rivalta test: This is a simple, inexpensive test that is useful to demonstrate a high content of inflammatory proteins in the effusion. A transparent tube of 10-20 mL is filled with 7-8 mL of distilled water and a drop of 98% acetic acid is added and mixed well in order to acidify the solution. A drop of effusion is placed carefully onto the surface of the mixture. If this drop disappears and the mixture remains clear, the result is negative. If the drop remains formed and slowly sinks to the bottom of the mixture like a jellyfish, the test is positive. A negative result has been found to have a high predictive value for the absence of FIP. A positive result could indicate FIP, but may also be seen in effusions due to lymphoma or bacterial infection.6

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PCR in effusions and tissues: Detection of the FCoV virus in effusion or tissue means in theory that there is systemic spread – i.e. the virus is FIPV and FIP disease is present. Studies suggest that this is a useful test to run, particularly on effusions, where it could replace the immunofluorescence test. A consensus as to whether this is an important corner piece or just a useful piece of the FIP puzzle has not yet been reached in the current literature, but updates are expected in the next few years. The RT-PCR test itself needs to be run to high technical standards in order to provide a good diagnostic performance.7

- The confusing or unhelpful “blue sky” pieces

Coronavirus antibody titres: The antibodies detected in these tests are against feline coronaviruses and are NOT specific for FIP. The vast majority of cats with FCoV titres do not have FIP. More cats have probably been euthanased based on a positive anti-
body titre than have died from FIP. Conversely, some cats with acute FIP will not have an antibody titre and some patients with terminal disease may test negative as dsall antibody is all bound to viral antigen. The diagnosis of this disease should never ever be based on antibody titres alone. Very high titres of > 1:1600 can be used as a very small piece of the puzzle only.

PCR in blood: Routinely offered PCR detection of coronavirus in blood is not useful, as the test cannot distinguish between FCoV and FIPV. In addition, false negatives occur commonly. A mutation in the spike protein (a protein on the envelope that assists with cell invasion) of the FCoV has recently been identified which appears to be associated with systemic spread of the virus (and therefore may be associated with FIPV).

A RT-PCR which detects this mutation is commercially available in Europe, but the presence of the mutation is still not 100% specific for the presence of FIP. PCR of faeces should only be used to identify FCoV shedders for the purposes of controlling viral spread in catteries.

Serum electrophoresis: Electrophoresis patterns may show either a monoclonal or polyclonal increase in gamma globulins, and are not specific for FIP. An alpha-2 globulin increase may be present, representing the increase in acute phase proteins, but is also a non-specific change. Additionally the serum A/G ratio has a higher diagnostic value than the gamma globulin concentration. Electrophoresis is therefore not a useful puzzle piece for the diagnosis of FIP.

Acute phase proteins: Acute phase proteins like serum amyloid A, haptoglobin and alpha(1)-acid glycoprotein (AGP) are elevated in cats with FIP, but increases are also expected in any systemic inflammatory disease. AGP however has been investigated in detail and increases above 1.5-2 mg/mL can potentially discriminate cats with FIP from those without FIP but with clinical signs consistent with the disease. This suggests that AGP is a useful puzzle piece but this test is unfortunately not available in South Africa.

Conclusion
The diagnosis of FIP is not always straightforward and a combination of findings, or pieces of the puzzle, need to be considered together. Finding more pieces will give a clearer picture, and some pieces make a bigger contribution than others.

FIP is NOT contagious:
Cats shed FCoV, which has a possibility of mutating to FIPV in specific cats - those with immunocompromise or immature immune systems, and those in high stress, multi-cat environments.

FIP is an SYSTEMIC disease - not an ENTERIC disease - and virus is not shed in the faeces.
1. Which one of the following statements about FIP is INCORRECT?  
   a. FIP is caused by the intestinal corona virus FCoV.  
   b. FIP is caused by a virulent mutation of the intestinal corona virus.  
   c. FIP is caused by an aberrant response of the host to the mutated corona virus.  
   d. FIP is caused by the mutated virus FIPV.  
   e. FIP virus is present in 90% of the feline population.

2. Which one of the following statements regarding the prevalence of FIP is INCORRECT?  
   a. The prevalence of enteric corona virus can reach 90% in breeding catteries.  
   b. Less than 5% of cats infected with FCoV will go on to develop FIP.  
   c. 50 – 70% of FIP occurs in cats older than one year of age.  
   d. Pure breed cats appear to be more susceptible.  
   e. High density population in shelters is a risk.

3. Which one of the following statements regarding effusions in FIP is INCORRECT?  
   a. Less than half of the effusions occurring in cats are due to FIP  
   b. Ascites and pleural effusion characterise the wet form of FIP.  
   c. The effusion due to FIP has a characteristic appearance  
   d. Tests performed on the effusion may be more diagnostic than those performed on blood.  
   e. Effusions in a young cat are almost diagnostic for FIP.

4. Which one of the following clinical signs is NOT typical of the dry form of FIP?  
   a. Intra-ocular changes such as uveitis  
   b. Refractory pyrexia  
   c. Dyspnoea  
   d. Enlarged abdominal lymphnodes  
   e. Neurological signs

5. Which of the following statements regarding FIP effusions is INCORRECT?  
   a. The cell count of the effusion is generally low (<2x10^9/L).  
   b. The fluid is generally clear and straw coloured.  
   c. The fluid had a high protein content making it quite viscous.  
   d. The major contribution to the protein content is albumin from vascular leakage  
   e. The Rivalta test is a simple test to show high protein content of fluid.

6. Which of the patterns of effusion characteristics listed below is MOST UNLIKELY to be FIP?  
   a. An effusion with a high cell count, a predominance of macrophages, low protein concentration, A/G ratio >0.8 and negative Rivalta test  
   b. An effusion with a low cell count, predominance of cells other than neutrophils and macrophages, a low protein concentration and a A/G ratio of >0.8 and a positive Rivalta test  
   c. An effusion with a low cell count, a predominance of cells other than neutrophils and macrophages, a high protein concentration, and A/G ratio of >0.6 and a positive Rivalta test  
   d. An effusion with a low cell count, a predominance of cells other than neutrophils and macrophages, low protein concentration (<30 g/L), A/G ratio >0.8 and negative Rivalta test is

7. Which one of the following statements regarding immunohistochemical staining for FIP is INCORRECT?  
   a. Immunohistochemical staining demonstrates presence of virus in the plasma  
   b. Immunohistochemical staining demonstrates presence of virus in the macrophages in the effusion  
   c. Immunohistochemical staining demonstrates presence of virus in the macrophages in tissue biopsies  
   d. Immunohistochemistry is 100% predictive and specific for FIP  
   e. Immunohistochemistry is not 100% sensitive - false negatives can occur

8. Which of the following statements regarding FeCoV titres is INCORRECT?  
   a. You can get false negative titres in the terminal phase of the disease  
   b. Serum corona titres are a good screening test for FIP infection  
   c. Serum corona titres test for feline corona virus not FIPV  
   d. Serum titres are only significant if titres are very high (1:1600) with a classical clinical presentation  
   e. Titres can be tested in blood and effusion fluid.
9. Which one of the following statements is INCORRECT?
   a. False negatives occur frequently with PCR in blood
   b. Routine PCR can distinguish between FCoV and FIPV
   c. Serum electrophoresis generally shows a polyclonal or even a monoclonal gammopathy, but is not specific for FIP
   d. Acute phase proteins are elevated in cats with FIP – but are also once again, non specific.
   e. AGP (alpha 1 – acid-glycoprotein) is elevated to a greater degree in cats with FIP versus cats with other causes of systemic inflammation

10. A problem with FIP in a cattery can be managed by doing which one of the following?:
   a. Testing the blood of all the cats for FCoV titres
   b. Doing PCR on blood of all the cats
   c. Doing PCR of the faeces of all the cats to detect shedders, and separating.
   d. Euthanising cats who are positive shedders
   e. Doing electrophoresis to check for acute phase proteins in all the cats

Management of FIP in High Density Households and Catteries

Breeding catteries are high risk environments and FCoV is endemic in many or most. The virus is transmitted primarily via the faecal-oral route and thus hygiene is most important. Corona virus is maintained by continued cycles of infection and re-infection. Young cats are predominantly infected - 40% of infections occur in cats 6m-2yrs old. The incidence of FIP is <4% if cats are older than 3 years.

The virus can stay viable for long periods in the cat litter. Gross and microscopic litter dust contains high numbers of virus particles. Cats contaminate their paws and fur as they use the litter box and then ingest virus particles when they groom themselves. Cats with an indoor outdoor lifestyle seldom get FIP. The risk of transmission is reduced if smaller groups of cats, ≤ 3, are kept per room, have separate airflow, and have outdoor access to bury their faeces.

Faecal shedding:
Attempts to control viral spread by segregation of faecal shedders and non-shedders has been suggested - but is still controversial.

- Faecal PCR is used to determine if the cat is shedding FCoV and non-shedders and shedders are separated.
- Shedders will be retested after 3 months as most natural infections will stop shedding after this time.
- 15% of cats are persistent shedders and these will need to be permanently separated.
- Cats with immunosuppressive conditions or other illness shed more virus for longer:
  - FIV: Shedding is 100X increased, and duration is prolonged.
  - FeLV: Proportion of cats become persistent shedders.
  - Stressed individuals = susceptible individuals.
  - Shelter environment amplifies shedding massively (10⁶) due to stress.
  - Sick animals and kittens shed higher levels.
- The primary stage of infection lasts 7-18 months - when shedding is at the highest levels.

Prevention of infection of Kittens:
Kittens typically develop FIP signs post weaning and after rehoming.

Isolation and early weaning:
Most kittens are protected from FCoV infection from maternal derived immunity after until they are 5-6 weeks of age. Separating the queen from other cats and removing their kittens to a clean environment at 5-6 weeks of age will help prevent transmission. Good hygiene is essential to prevent transmission of viruses on food and litter items and clothing.

References: