Guidelines on Feline Infectious Diseases

FELINE INFECTIOUS PERITONITIS

October 2008

The following recommendations have been formulated by the European Advisory Board on Cat Diseases.

The European Advisory Board on Cat Diseases is an independent panel of 17 veterinarians from ten European countries, with an expertise in immunology, vaccinology and/or feline medicine. The ABCD was set up to compile guidelines for the prevention and management of major feline infectious disease in Europe based on current scientific knowledge and available vaccines.

This work would not have been possible without the financial support of Merial.

ABCD GUIDELINES ON:

7. FELINE INFECTIOUS PERITONITIS 3

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7. Feline Infectious Peritonitis

7.1 Biology of the virus

7.1.1 Virus properties

Feline coronavirus (FCoV) belongs to the family *Coronaviridae* of the Order *Nidovirales* [de Vries et al, 1997]. These viruses are large, spherical, enveloped, positive-sense single-stranded RNA viruses [Lai and Holmes, 2001].

With a genome of 27 to 32 kb, encoding an ~750-kDa pp1ab replicase polyprotein, four structural proteins (S for Spike, M for Matrix, N for nucleocapsid, and E for Envelope) and up to five accessory non-structural proteins, coronaviruses (CoVs) are the largest RNA viruses known to date [Brown and Brierly, 1995; de Vries et al, 1997].

A significant characteristic of these viruses is their capability to undergo recombination [Lai, 1996; Lai and Holmes, 2001].

FCoV, together with canine coronavirus (CCoV) and transmissible gastroenteritis virus (TGEV) of pigs, belongs to group I of the coronaviruses, defined by both antigenic and genomic properties.

Feline coronavirus may itself be subdivided serologically and by nucleotide sequencing into two types. Type I virus is the most prevalent [Hohdatsu et al, 1992; Addie et al, 2003; Vennema, 1999; Kummrow et al, 2005; Shiba et al, 2007]. Type II virus is less common and results from recombination between type I feline coronavirus and canine coronavirus involving the spike gene [Herrewegh et al, 1998]. Most research studies have been conducted on type II since, unlike type I virus, it can be readily propagated in cell cultures [Pedersen et al, 1984]. Both types of virus can induce FIP. Previously, feline coronavirus strains have also been subdivided into two distinct “biotypes”: Feline Enteric Coronavirus (FECV) and Feline Infectious Peritonitis Virus (FIPV) [Pedersen, 1987]. However, since all FCoV may induce systemic infection as demonstrated by RT-PCR studies such descriptions are perhaps best avoided and have not been used in these guidelines.

Feline coronavirus is an enveloped virus that can survive up to seven weeks in a dry environment [Scott, 1988]. Therefore, FCoV can be transmitted indirectly readily, e.g. via litter trays, shoes, hands and clothes. Indirect transmission may also occur at cat shows. However, FCoV can be readily inactivated by most household detergents and disinfectants.

7.1.2 Epidemiology

Feline coronavirus infection is extremely common in domestic cats and wild felidae may also be seropositive. Infection is particularly common in multi-cat households where the seroprevalence may reach 90 to 100% [Horzinek et al, 1979; Addie and Jarrett, 1992; Sparkes et al, 1992; Addie, 2000; Kummrow et al, 2005; Herrewegh et al, 1995; Foley et al, 1997; Kiss et al, 2000]. A substantial proportion of FCoV infected cats go on to develop FIP, a fatal disease [Pedersen, 1995b] that is especially common in multi-cat environments [Addie & Jarret, 1992]. In some studies up to twelve percent of FCoV infected cats subsequently die from FIP [Addie et al, 1995a; Fehr et al, 1995]. The prevalence of FIP will depend on the population of cats, particularly their age, and local differences are likely to apply.

Some breeds of cats (e.g. Persians) and individual lines within breeds are more likely to be affected by FIP. [Kiss et al, 2000; Pesteanu-Somogyi et al, 2006]. Age is an important risk factor for FIP and 70% of cats that develop disease are less than one year old [Rohrer et al, 1993; Hartmann, 2005]. However, the disease has been observed in cats up to 17 years of age. It has also been suggested that the prevalence of FIP is higher in sexually-entire cats [Pesteanu-Somogyi et al, 2006].

Since any stress experienced during FCoV infection may predispose a cat to develop FIP e.g. surgery, visit to a cattery, moving, co-infection with FeLV [Poland et al, 1996; Rohrer et al, 1993], stress management is an important part of control.

In breeding catteries, kittens usually become infected at a young age, often prior to weaning. The mother is often the source of infection, particularly if the litter has been reared in isolation. The exact age at which kittens become infected appears to vary. It

may not occur until 5-6 weeks of age, associated with the loss of maternally derived immunity, but in some situations very early infection (as early as 2 weeks of age) has been detected [Lutz et al 2002].

Faeces are the major source of FCoV and the major mode of transmission is believed to be the faecal-oral route, with litter boxes representing the main source of infection in groups of cats. Contamination via saliva may occur in groups of cats in close contact or sharing feeding bowls [Addie & Jarrett, 2001]. Transplacental transmission has been described from a queen that developed the disease during pregnancy [Pastoret & Henroteaux, 1978] but is rare [Addie & Jarrett, 1990].

Susceptible cats are most likely to be infected with FCoV from asymptomatic cats. Although transmission of infection from cats with FIP may occur, it is important to note thatthis does not necessarily lead to disease. Indeed, transmission of FIP is considered unlikely under natural conditions although it has been demonstrated experimentally.

Folllowing natural infection with FCoV cats begin to shed virus in the faeces within one week [Pedersen et al, 2004] and shedding continues for weeks to months. A small proportion of cats may shed virus for life (also called carriers) [Addie & Jarrett, 2001] and at high levels [Horzinek & Lutz, 2000]. Whilst a cat remains infected, faecal excretion of virus appears to be continuous [Addie & Jarrett, 2001].

7.2 Pathogenesis

Most cats infected by FCoV either develop an asymptomatic infection or show minor signs of enteritis. Only a proportion (see above) of these cats goes on to develop FIP, a pyogranulomatous disease [Pedersen et al, 1981; Pedersen, 1987].

The precise cause of FIP is unclear but there are two main hypotheses. First, that a mutation occurs which favours viral replication in monocytes and macrophages [Poland et al, 1996; Vennema et al, 1998; Cornelissen et al, 2007 Haijema et al, 2004; Rottier et al, 2005]. This has been called the internal mutation theory although no consistent mutation has yet been identified. In support of this hypothesis is the presence of highly virulent strains of FCoV that are capable of consistently inducing FIP, albeit under experimental

conditions [Poland and Venemma 1996]. The second hypothesis for the development of FIP is that any FCoV can cause FIP but that the viral load and the cat's immune response determines whether or not FIP will develop [Addie et al, 1995, Dewerchin et al, 2005; Dye & Siddell, 2007; Meli et al, 2004, Rottier et al, 2005; Kipar et al, 2006]. It is likely that both factors, namely viral genetics and host immunity, play a role in the development of FIP.

FIP occurs in two major forms: an effusive form which is characterized by polyserositis (e.g. thoracic and abdominal effusion) and vasculitis as a consequence of injury of blood vessels wall by extravasating macrophages [Kipar et al, 2005] and a non-effusive form characterized by granulomatous lesions in organs. These two forms probably reflect clinical extremes of what is in reality a continuum, with many cats having signs and lesions consistent with both forms.

A rare nodular enteric form described in young cats with diarrhoea and vomiting was associated with intestinal pyogranulomatous lesions [Van Kruiningen et al, 1983; Harvey et al, 1996].

All forms of FIP are lethal and the disease progression may be the consequence of severe immunodepression by T-cell depletion [de Groot-Mijnes et al, 2005].

Whether a cat develops the wet or dry form of the disease is thought to depend on strength of the T-cell-mediated immune response, which is probably the only efficient immune response against disease progression [Pedersen, 1987; Cornelissen et al, 2007]. The wet forms are presumed to be the consequence of a weak cell-mediated immune response [Pedersen, 1987].

Attempts to identify a tissue distribution of FCoV that is diagnostic for FIP have proved difficult. In cats with FIP, virus replicates to high titres in monocytes and can be found in many organs [Kipar et al, 2005]. In asymptomatic cats, FCoV is mainly confined to the intestine. However a low-level monocyte-associated viraemia can also be detected by RT-PCR [Gunn-Moore et al, 1998b; Herrewegh et al, 1995; Meli et al, 2004] and a high-level of replication has also been demonstrated in organs of asymptomatic cats, at least within

the first month after an experimental infection with FCoV type I [Meli et al, 2004]. A significant difference in viral replication in haemolymphatic tissues has been demonstrated between cats that died from FIP and healthy long-term infected cats [Kipar et al, 2006].

Monocytes and macrophages remain infected by FCoV even in the presence of high levels of antibodies. The mechanism of this immune evasion has not yet been elucidated but one hypothesis could be an escape from antibody-dependent lysis due to absence of viral antigens on the surface of infected cells triggered by FCoV specific antibodies [Dewerchin et al, 2006; Cornelissen et al, 2007]. The direct consequence may be a quiescent infection state and a long incubation period. Activation of monocytes and perivascular macrophages may lead to the development of typical widespread pyogranulomatous and vasculitis/perivasculitis lesions in various tissues and organs, including lung, liver, spleen, omentum, and brain of cats with FIP [Kipar et al, 2005; Berg et al, 2005].

7.3 Immunity

It remains to be determined how some cats are protected from developing FIP. It has been suggested that cats developing a successful CMI response do not develop FIP, whereas cats that develop a predominantly humoral response are likely to develop disease [Pedersen 1987]. Hypergammaglobulinaemia [Ward et al, 1974; Paltrinieri et al, 1998] is common in cats with FIP. Also a profound depletion of T cells from the blood [de Groot-Mijnes et al, 2005] as well as from lymphoid tissues has been described [Haagmans et al, 1996; Paltrinieri et al, 2003; Dean et al 2003].

7.3.1 Passive immunity

As in coronavirus infections of other species, maternally derived antibody (MDA) usually gives protection until about 5-6 weeks of age [Addie & Jarrett, 1992]. Levels of MDA decline and become undetectable by 6-8 weeks of age [Pedersen et al 1981].

7.3.2 Active immune response to feline coronavirus infection

7.3.2.1 Cell-mediated immunity

Cats that did not develop disease after experimental coronavirus infection displayed a greater CMI compared to those that did develop disease [Pedersen & Floyd, 1985, de Groot-Mijnes et al, 2005]. Studies that measured cytokine responses in blood or lymphatic tissues revealed decreased IL-12 responses, and low levels of IFN-gamma expression [Kiss et al, 2004; Gelain et al 2006; Kipar et al 2006], indicative of impaired cellular immune responses, although results were not always consistent.

7.3.2.2 Humoral immunity

Cats may be reinfected only weeks after they have overcome a first episode of feline coronaviruses because natural immunity is short-lived. [Addie et al, 2003].

The role of humoral immunity in protection against FIP is controversial. Clearance of natural infections has been associated with antibodies directed against the FCoV spike protein [Gonon et al, 1999], suggesting that, in natural infection, humoral immunity may have a role in protection. However, the role of humoral immunity in natural infections is unknown. It has been proposed that antibodies, especially those directed against the spike protein, can be detrimental. In cats with pre-existing antibodies an enhanced form of disease has occurred in experimental infections, typified by an earlier development of disease and a shortened disease course leading to a more rapid death. This phenomenon was observed in experimental cats that acquired their antibodies through passive or active immunization [Pedersen & Boyle 1980; Weiss & Scott 1981]. Furthermore, in a study in which cats were immunised with a recombinant vaccinia virus expressing the coronaviral S protein, cats became severely ill 7 days after challenge with the virulent, FIP-causing mutant. In contrast, the unvaccinated control cats survived for more than 28 days [Vennema et al, 1990]. This antibody-dependent enhancement (ADE) is likely mediated by opsonisation of the virus facilitating viral uptake by macrophages via Fc receptor-mediated attachment [de Groot and Horzinek, 1995; Corapi et al, 1992]. However, the role of ADE in natural infection is not clear since in field studies cats were most likely to develop FIP on first exposure to FCoV [Addie et al, 1995a, 1995b, 2003].

7.4 Clinical signs

The clinical presentation of FIP is extremely variable and this is reflected in the marked variability in the distribution of the vasculitis and pyogranulomatous lesions.

FIP has previously been classified as occurring in effusive and non-effusive (wet and dry) forms. This has some value in recognizing clinical presentations of FIP and contributing to diagnosis but it is clear that there is considerable overlap between the two forms. In cases with predominantly non-effusive features, investigation of possible accumulation of sub-clinical, small amounts of effusion can be helpful to provide samples for diagnostic testing.

Fever refractory to antibiotics, lethargy, anorexia and weight loss are common non-specific signs but occasional cases remain bright and retain body condition.

Ascites is the most obvious clinical manifestation of the effusive form [Holzworth, 1963, Wolfe & Griesemer 1966]. Thoracic and pericardial effusion may occur in combination with abdominal effusion. In a smaller proportion of cases effusion is restricted to the thorax and those cats usually present with dyspnoea. Serositis can involve the tunica vaginalis of the testes leading to scrotal enlargement. Non-effusive (or dry) FIP frequently represents a major diagnostic challenge. Non-specific signs of pyrexia, anorexia and lethargy may be the only signs, particularly in the early stages of disease. More specific signs will depend on the organs or tissues involved in the vasculitis and pyogranulomatous lesions. Abdominal organs are a common site for lesions. Renal involvement may lead to renomegaly detectable on palpation. Mural intestinal lesions in the colon or ileocaecoecolic junction occasionally occur and may be associated with chronic diarrhoea and vomiting. There may also be palpable enlargement of the mesenteric lymph nodes and this may be misinterpreted as neoplasia [Kipar et al, 1999]. Ocular involvement is common, leading to a variety of changes, such as iris colour, dyscoria or anisocoria secondary to iritis, sudden loss of vision and hyphaema. Keratic precipitates can also be seen and may appear as “mutton fat” deposits on the ventral corneal endothelium [Davidson, 2006]. The iris may show swelling, a nodular surface,

and aqueous flare may be detected. On ophthalmoscopic examination chorioretinitis, fluffy perivascular cuffing (representing retinal vasculitis), dull perivascular puffy areas (pyogranulomatous chorioretinitis), linear retinal detachment and fluid blistering under the retina may be seen. Neurological signs are reported in around 10% or more of cats with FIP [Rohrer et al, 1993]. They reflect focal, multifocal, or diffuse involvement of the brain, the spinal cord and meninges. The most commonly reported signs are ataxia, hyperaesthesia, nystagmus, seizures, behavioural changes and cranial nerve defects [Kline et al, 1994; Timman et al, 2008]. Cutaneous signs have recently been reported occurring as multiple nodular lesions caused by pyogranulomatous-necrotising dermal phlebitis [Cannon et al, 2005] and skin fragility [Trotman et al, 2007]. A diffuse pyogranulomatous pneumonia is seen in some cases leading to severe dyspnoea [Trulove et al, 1992].

7.5 Diagnosis

Diagnosis of FIP *intra vitam* is extremely challenging. In addition, a definitive diagnosis may not always be possible, e.g., because of the invasiveness of biopsies in a sick cat. Difficulties in definitively diagnosing FIP arise from an absence of non-invasive confirmatory tests in cats with no effusion. Presence of effusion should first be ruled out because obtaining effusion and analysis is very useful and relatively non-invasive. In cats with no effusion, several parameters, including the background of the cat, history, presence of clinical signs, laboratory changes, and antibody titres [Rohrer et al, 1993] should be used to help to inform the decision about appropriate further diagnostic procedures.

7.5.1 Haematology

Haematology results are often altered in cats with FIP, but the changes are not pathognomonic. White blood cell counts can be decreased or increased. Lymphopenia is commonly present; however, lymphopenia in combination with neutrophilia is generally common in cats as a typical “stress leukogram” and can occur in many other diseases. However, a normal lymphocyte count makes FIP less likely. A mild to moderate non-

regenerative anaemia is also a common, but non-specific, finding, which may occur in almost any chronic disease of the cat.

A very common laboratory finding in cats with FIP is an increase in total serum protein concentration caused by a rise in globulins, mainly γ-globulins [Paltrinieri et al, 2001; 2002]. In one study hyperglobulinaemia was found in about 50% of cats with effusion and 70% of cats without effusion [Sparkes et al, 1994]. Following experimental infection, an early increase of α_2-globulins is seen, while γ-globulins and antibody titres increase just prior to the onset of clinical signs [Pedersen 1995; Gunn-Moore et al, 1998]. Serum total protein levels in cats with FIP can reach very high concentrations of up to 120 g/l (12 g/dl) or higher. In some studies, the albumin to globulin ratio was found to have a significantly higher diagnostic value than either total serum protein or γ-globulin concentrations, because a decrease in serum albumin also may occur through a decrease in production [Shelly et al, 1988; Rohrer et al, 1993; Hartmann et al, 2003]. Low albumin is usually associated with protein loss caused by glomerulopathy secondary to immune complex deposition or by extravasation of protein-rich fluid during vasculitis [Hartmann et al, 2003]. An optimum cut-off value (maximum efficiency) of 0.8 was determined for the albumin to globulin ratio [Hartmann et al, 2003]. Serum protein electrophoresis may show both polyclonal and monoclonal hypergammaglobulinaemia as well as increases in acute phase proteins. Other laboratory parameters (liver enzymes, bilirubin, urea, creatinine) can be variably elevated depending on the degree and localization of organ damage, but are generally not helpful in establishing an etiological diagnosis. Hyperbilirubinemia and icterus are often observed and frequently are a reflection of hepatic necrosis [Hartmann et al, 2003]. Sometimes, bilirubin is increased without evidence of haemolysis, liver disease, or cholestasis; this unusual change is otherwise only observed in septic animals. Bilirubin metabolism and excretion into the biliary system is compromised in these cats due to high levels of TNF-α that inhibit transmembrane transport. Thus, high bilirubin in the absence of haemolysis and elevation of liver enzyme activity should raise the suspicion of FIP. Recent research has focused on the diagnostic value of acute phase reaction parameters including α_1-acid

glycoprotein (AGP), a serum acute phase protein that is elevated in cats with FIP [Duthie et al, 1997; Paltrinieri, 2008]. High serum AGP levels (>3 mg/ml) can support the diagnosis of FIP [Paltrinieri et al, 2007a], but levels also rise in other inflammatory conditions and thus, these changes are not specific. Additionally, AGP may also be high in asymptomatic cats infected with FCoV, especially in households where infection is endemic [Paltrinieri et al, 2007a].

7.5.2 Tests on effusion fluid

If there is effusion, the most important diagnostic step is to sample the fluid, because tests on effusion have a much higher diagnostic value than tests that can be performed on blood. Only about half of the cats with effusion suffer from FIP [Hirschberger et al, 1995]. Although effusions of clear yellow colour and sticky consistency are often called “typical”, the presence of this type of fluid in body cavities alone is not diagnostic. Sometimes the fluid has a totally different appearance and some cases of FIP with pure chylous effusion have been reported [Savary et al, 2001]. Usually the protein content is very high (>35g/dl) and consistent with an exudate, whereas the cellular content is low (< 5000 nucleated cells/ml) and approaches that of a modified transudate or pure transudate. Cytology of the effusion in cats with FIP shows a variable picture but often consists predominantly of macrophages and neutrophils. Electrophoresis in effusions is a diagnostic tool with a high positive predictive value if albumin/globulin ratio is < 0.4 and a high negative predictive value if the ratio is > 0.8 [Shelly et al, 1988]. Major differential diagnoses of cats with similar effusions include inflammatory liver disease, lymphoma, heart failure, and bacterial peritonitis or pleuritis.

“Rivalta’s test” is a very simple, inexpensive method that does not require special laboratory equipment and can be easily performed in private practice. This test was originally developed by the Italian researcher Rivalta around 1900 and was used to differentiate transudates and exudates in human patients. This test is very useful in cats to differentiate between effusions due to FIP and effusions caused by other diseases

[Hartmann et al, 2003]. Not only the high protein content, but high concentrations of fibrinogen and inflammatory mediators lead to a positive reaction.

Box 1. Rivalta's test

To perform this test, a transparent reagent tube (volume 10 ml) is filled with approximately 7-8 ml distilled water, to which 1 drop of acetic acid (98%) is added and mixed thoroughly. On the surface of this solution, 1 drop of the effusion fluid is carefully layered. If the drop disappears and the solution remains clear, the Rivalta's test is defined as negative. If the drop retains its shape, stays attached to the surface or slowly floats down to the bottom of the tube (drop- or jelly-fish-like), the Rivalta's test is defined as positive.

The Rivalta's test had a high positive predictive value (86%) and a very high negative predictive value for FIP (96%) in a study in which cats that presented with effusion were investigated (prevalence of FIP 51%) [Hartmann et al, 2003]. Positive Rivalta's test results can occur in cats with bacterial peritonitis or lymphoma. Those effusions, however, are usually easy to differentiate through macroscopic examination, cytology, and/or bacterial culture.

7.5.3 Cerebrospinal Fluid

Analysis of cerebrospinal fluid (CSF) from cats with neurological signs due to FIP lesions may reveal elevated protein (50 - 350 mg/dl with a normal value of less than 25 mg/dl) and pleocytosis (100 - 10000 nucleated cells/ml) containing mainly neutrophils, lymphocytes, and macrophages [Li et al, 1994; Rand et al, 1994; Foley et al, 2003], which is, however, a relatively non-specific finding. Many cats with neurological signs caused by FIP have normal CSF.

7.5.4 Antibodies

Antibody titres measured in serum can contribute to the diagnoses if interpreted with care. A high percentage of healthy cats are FCoV antibody-positive and most of those cats will never develop FIP. Thus, antibody titres must be interpreted with extreme caution; it has been contended that more cats have died of false interpretation of FCoV antibody test results than of FIP [Pedersen, 1995a]. There is no "FIP antibody test", all

that can be measured is antibodies again FCoV. Methodology (and thus, antibody titre results) may vary significantly between laboratories. It is important to realize that the presence of antibodies does not indicate FIP and absence of antibodies does not exclude FIP. Low or medium titres do not rule out FIP and approximately 10% of the cats with clinically manifest FIP have negative results [Hartmann et al, 2003]. In cats with fulminant FIP, titres may decrease terminally [Pedersen, 1995a]. This is either because large amounts of virus in the cat's body bind to antibodies and render them unavailable to bind antigen in the antibody tests or because the antibodies are lost into the effusion when protein is extravasated in vasculitis. Very high titres can be of certain diagnostic value and increase the likelihood of FIP [Hartmann et al, 2003].

Measuring antibodies in fluids other than blood has been investigated [Boettcher et al, 2007; Foley et al, 1998]. However, interpretation of titres in effusion and cerebrospinal fluid is even more complicated than titres in blood and measurement of antibodies there is therefore not recommended.

7.5.5 FCoV Reverse-transcriptase polymerase chain reaction (RT-PCR)

FCoV RT-PCR in blood is sometimes used as diagnostic tool for the diagnosis of FIP. At the time of writing no PCR has been developed which can definitively diagnose FIP, and FCoV RT-PCR in blood is not recommended for the diagnosis of FIP. This is because it is not possible to distinguish between the FIP-inducing mutant and the non-mutated FCoV [Fehr et al, 1996]. Furthermore, positive FCoV RT-PCR results occur not only in cats with FIP but also in healthy carriers that did not develop FIP for a period of up to 70 months [Gunn-Moore et al, 1998b; Meli et al, 2004; Gamble et al, 1997; Herrewegh et al, 1997], and negative FCoV RT-PCR also occurs very commonly in cats with FIP [Hartmann et al, 2003]. Another approach is to measure messenger RNA by RT-PCR in blood with the rationale that levels of messenger RNA may correlate with the level of replication of FCoV and thus, be correlated with the presence of FIP. However, validity is unclear at present, 5 to 50 % of healthy were positive in that test [Simons et al, 2005; Can-Sahnak et al, 2007], and so far, the test is not available in Europe.

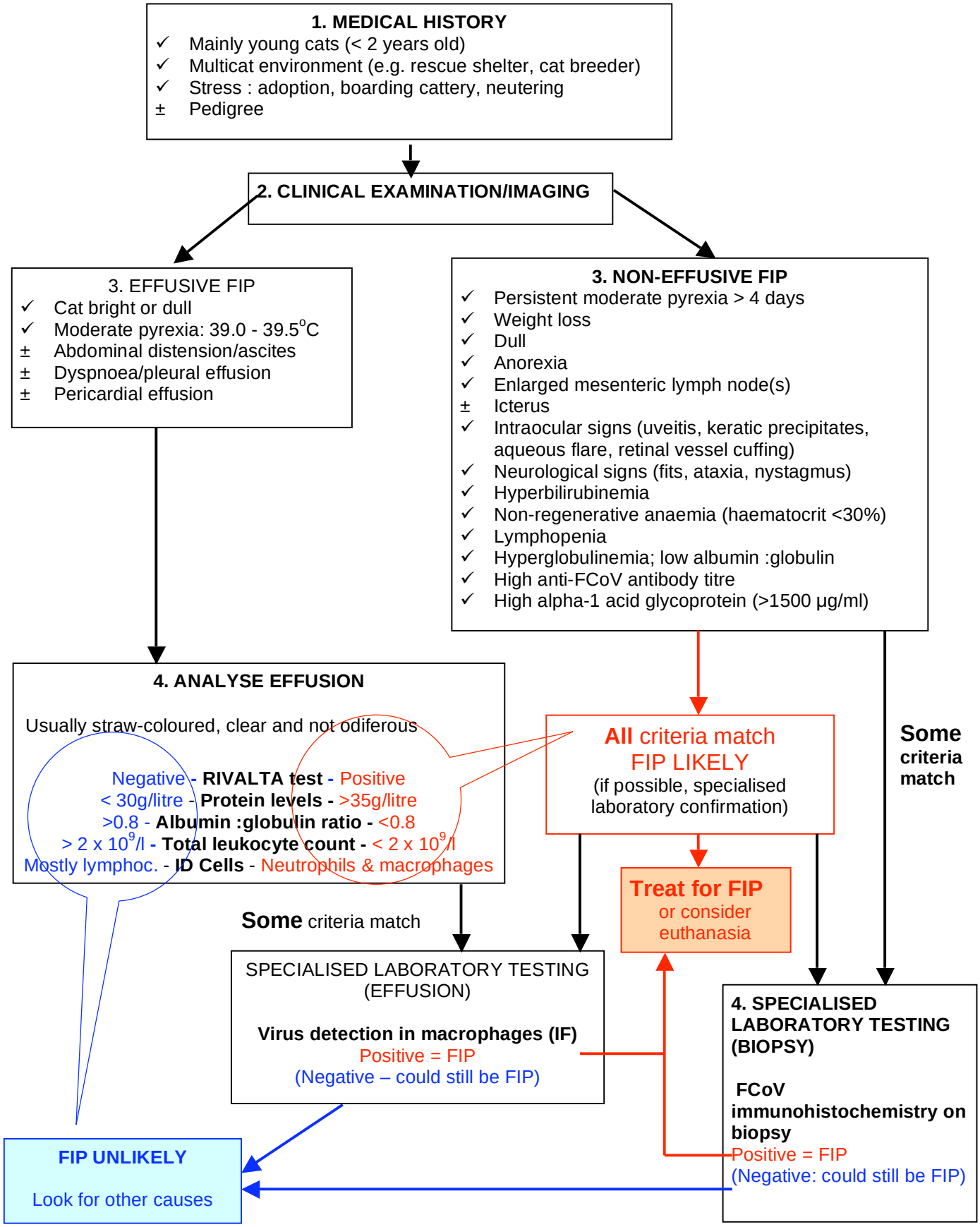
PCR in effusion or CSF has also been discussed as a diagnostic tool. However, data on the value of these approaches are not yet available.

7.5.6 Immunostaining of FCoV antigen in macrophages

Methods to detect the virus itself include the search for the presence of FCoV antigen in macrophages using immunofluorescence (in effusion macrophages) or immunohistochemistry (in tissue macrophages). While FCoV may be present systemically in cats without FIP, only in FIP will there be sufficiently large amounts of virus in macrophages to obtain positive staining. In a recent study in which a large number of cats with confirmed FIP and controls with other (confirmed) diseases were investigated (n = 171, prevalence of FIP 64%), positive immunofluorescence staining of intracellular FCoV antigen in macrophages of the effusion was 100 % predictive of FIP [Hartmann et al, 2003]. Unfortunately, the negative predictive value is not very high (57%), which can mainly be explained by low numbers of macrophages on effusion smears (even though cats have FIP) resulting in negative staining [Hartmann et al, 2003]. Immunohistochemistry can be used to detect the expression of FCoV antigen in tissue, and it also proved to be 100% predictive of FIP if positive [Tammer et al, 1995; Kipar et al 1998b]. However, invasive methods (e.g. laparotomy or laparoscopy) are usually necessary to obtain appropriate tissue samples. When the diagnostic sensitivity between true-cut biopsy (TCB) and fine-needle aspiration (FNA) of liver and kidney tissue obtained at necropsy was compared, the sensitivity of FNA was similar to TCB, but a higher sensitivity in the liver *versus* kidneys was observed [Giordano et al, 2005]. The value of ultrasound-guided FNA to diagnose FIP *in vivo*, however, still has to be investigated.

Therefore, there are 2 diagnostic strategies to obtain a definitive diagnosis of FIP. If there is effusion, immunofluorescence staining of FCoV antigen in effusion macrophages can diagnose FIP. If there is no effusion, tissue samples of affected organs have to be obtained. Histology is confirmative or immunohistochemical staining of FCoV antigen in tissue macrophages can be used to diagnose FIP. A diagnostic algorithm is provided in figure 7-1.

Figure 7-1. Diagnostic approach to FIP



7.6 Management of cats with FIP

Any cat in a hospital is potentially a source of FCoV infection. Therefore, routine hygiene measures should be taken to avoid inadvertent spread of FCoV infection between cats. In a case of a cat with FIP, this cat is highly likely to shed FCoV and therefore strict precautions to avoid infection of other cats is particularly important. However, any in-contact cats within the cat's own home will probably have already been exposed to the FCoV from the cat, so there is no particular benefit in isolating the cat at home. It remains controversial whether FIP-inducing mutants are shed from a cat with FIP.

In situations where a cat with FIP has been euthanased and there is no cat left in that household it is recommended to wait 2 months before obtaining a new cat. If other cats in that household remain, they most like carry FCoV. Before introducing a new cat into that household, several things have to be considered including environment, number and density of cats as well as their age, and it is prudent to wait at least several months in any case before introducing new cats.

7.6.1 Treatment

Treatment (or euthanasia) should only be considered after every effort has been made to obtain a definitive diagnosis. Once FIP is established, in most cats it is fatal. There have been reports of occasional cats surviving up to several months after diagnosis of FIP. It is not clear whether this improvement was caused by treatment. There have even been some very occasional reports of cats that have "recovered" from FIP, but in these cases a definitive diagnosis of FIP had not been obtained.

As FIP is caused by inflammatory and inappropriate immune-responses to FCoV, supportive treatment is aimed at suppressing that inflammatory and inappropriate immune-response, usually with corticosteroids. There are, however, no controlled studies that indicate whether corticosteroids have any beneficial effect or not. Occasional cases treated with corticosteroids have shown improvement for up to several months, but these are anecdotal reports.

Numerous other treatments have been tried, but data from only one controlled field study have been published. In this placebo-controlled study of 37 cats treatment with feline interferon omega showed no benefit compared to the placebo [Ritz et al, 2007]. Other drugs (table 1) have been used, but there are no controlled studies to support their efficacy.

Table 7-1. Drugs that have been suggested for use in FIP

Substance	**Comment**	**ABCD recommendation (EBM level)**
ANTIVIRALS		
Ribavirin	works in vitro but toxic in cats	not recommended (2)
vidarabin	works in vitro but toxic in cats	likely ineffective (4)
human interferon-α SC high dose	although human interferon-a has in vitro effect on FCoV, SC treatment didn't work in an experimental trial	ineffective (2)
human interferon-α PO low dose	no trials only acts as immune-stimulant if given orally immune-stimulation should be avoided in cats with FIP	contraindicated (4)
feline interferon-ω	one placebo-controlled study of naturally occurring cases	ineffective (1)
IMMUNOSUPPRESSANTS		
prednisolone/ dexamethasone (immuno-suppressive doses)	no controlled studies, some cats have improved during treatment and survived for several months , but does not cure FIP	currently supportive treatment of choice (3) if effusion is present, dexamethasone IT or IP may be helpful
pentoxyfylline	aimed at treating the vasculitis some veterinarians in practice have tried, but there are no published studies or case reports	Requires studies (4)
Ozagrel hydrochloride	thromboxane synthesis inhibitor aimed at treating the inflammatory response, only used in 2 cases with beneficial effect	may have some beneficial effect, controlled studies needed (3)
cyclosporine A	aimed to immune-suppress (lower the corticosteroid dose), No published studies	not recommended because more directed against cellular immunity than humoral (lack of data) (4)
cyclophosphamide	aimed to immune-suppress (lower the corticosteroid dose), No published studies	might be considered in combination with glucocorticoids (4)
chlorambucil	aimed to immune-suppress (lower the corticosteroid dose), No published studies	might be considered in combination with glucocorticoids (4)
azathioprine	toxic in cats (!), aimed to immune-suppress (lower the corticosteroid dose), No published studies	not recommended (4)
Salicylic acid (aspirin) platelet inhibitory dosage	aimed at treating the inflammatory response as well as the vasculitis. No published studies	may have some beneficial effect, but side effects possible if used in combination with high steroids

EBM level 1 = confirmed by placebo-controlled double-blind field study
EBM level 2 = shown in a controlled experimental study
EBM level 3 = supported by case series
EBM level 4 = only based on expert opinion

The prognosis for cats with FIP is very poor. In one recent published study the median survival after diagnosis was 9 days. Factors that indicate a short survival time are low lymphocyte count, high bilirubin, presence of high amount of effusion. Cats that show no improvement within 3 days are unlikely to show any benefit from treatment and euthanasia should be considered.

7.7 Vaccination

Many attempts have been made to develop effective and safe vaccines to protect cats against FIP. Unfortunately most of these studies failed, with ADE observed in several trials. At present there is only one commercial vaccine available (Primucell©, Pfizer). This vaccine is available in the USA and some European countries.

Primucell® contains a temperature sensitive mutant of the type 2 FCoV strain DF2. The vaccine is administered intranasally and aims at inducing local mucosal immune responses through the induction of IgA and cell mediated immunity. However the vaccine also induces seroconversion, although titres are generally low. There is considerable controversy regarding the safety and efficacy of this vaccine. The vaccine contains a type-2 strain, whereas type-1 coronaviruses are more prevalent in the field. Different studies on the efficacy of vaccination in inducing protection against disease have been performed, both under experimental and field conditions.

Although some experimental studies have indicated that vaccination protects against disease, results have not been consistent. Preventable fractions between 0 and 75 % have been reported [Hoskins et al, 1995; McArdle et al, 1995; Scott et al, 1995; Gerber et al 1990]. The results of field studies examining the efficacy of protection have been equally contradictory. No difference in the development of FIP between the vaccinated and placebo group was found when the vaccine was used in Persian breeding colonies [Fehr et al 1995]. In a double-blind trial including 609 cats, no differences between the vaccinated and placebo group were found during the first 150 days after vaccination. However, after 150 days, fewer FIP cases occurred in the vaccinated group compared to the placebo group (1 against 7). In another trial, a preventable fraction of 75% was

found when the vaccine was tested in a very large cat shelter in the USA [Postorino Reeves, 1995]. In the latter study all kittens were seronegative prior to vaccination. Therefore it can be concluded that Primucell® might not be effective in seropositive cats that have already been exposed to FCoV. Since Primucell® is licensed for use from 16 weeks of age and is not effective in younger cats [Lutz et al 2002], most kittens (and especially those living in breeding colonies and multiple cat households) have already been infected and are seropositive. This is an important practical limitation for its use. The ADE that was a feature of some experimental vaccine trials has not been observed in field studies, suggesting that the vaccine can be considered safe.

7.7.1 Primary vaccination course

ABCD does not consider the FIP vaccine as a core vaccine. Vaccination can be considered in kittens that are unlikely to have been exposed to FCoV, e.g. from an early weaning programme particularly if they enter an FCoV endemic environment.

If immunization is considered, a primary vaccination course consisting of 2 doses of the vaccine 3 weeks apart from an age of 16 weeks onwards should be given. Vaccination before 16 weeks was not shown to give protection against infection [Lutz et al 2002]. Therefore there are two particular problems in breeding catteries; firstly most kittens are already seropositive at the age of vaccination and secondly FCoV infection occurs much earlier than 16 weeks [Lutz et al, 2002, Addie & Jarrett 1992].

7.7.2 Booster vaccination

In cats of which the lifestyle has justified primary vaccination, annual boosters may be considered. Although studies on the duration of immunity are lacking, it is thought to be short lived and regular boosters are recommended to maintain immunity.

7.8 FIP control in specific situations

FIP is a problem of cats kept in groups, particularly in breeding catteries and in rescue situations. Since the most important route of transmission is fecal-oral transmission, Hygiene is the foremost method of FIP control in any multi-cat environment. FCoV

infection is maintained in a household or cattery by continual cycles of infection and re-infection [Foley et al, 1997, Addie et al, 2003] with the source of infection being the cat litter tray. FIP is rarely a problem amongst cats leading a natural, indoor-outdoor, lifestyle. The goal in every cat household has to be to reduce the FCoV pressure and risk of transmission. This can be done by avoiding large numbers of cats in single households, keeping small group groups of cats of not more than 3 (well-adapted) cats per room, observing strict hygiene, and providing outdoor access to enable the cats to bury their faeces. If the latter is not possible, enough litter boxes should be provided, cat litter boxes have to be cleaned frequently, and litter trays should be in different rooms from food bowls.

7.8.1 Breeding catteries

Breeding catteries are high-risk situations for FIP. Today, in most European countries, there are few catteries in which FCoV is not endemic. In some catteries, attempts have been made to control the spread of FCoV by segregation. A policy of separating cats which are shedding high amounts of FCoV from low shedders and negative cats has been suggested for reducing transmission within a cattery but the value of this approach is controversial. High shedders can be detected using RT-PCR screening of faeces but multiple sampling (optimum 4 times over 3 weeks) may be necessary for this to be reliable and this presents practical difficulties. Virus shedding occurs over several months is life-long , in some cats, especially in multi-cat households.

Kittens typically develop FIP in the post-weaning period [Cave et al, 2002], therefore many breeders are unaware that they have endemic FCoV infection, since FIP deaths usually occur once the kittens are in the new household. Most kittens are protected from FCoV infection by maternally derived antibodies until they are between 5 and 6 weeks of age. It has been reported that it may be possible to prevent FCoV infection of young kittens by isolating pregnant queens 2 weeks before birth and removing kittens from their mother to a clean environment when they are 5-6 weeks old and maintaining them there until they go to a new home [Addie & Jarrett, 1990, 1992 and 1995]. For this

technique to work, the breeder is required to strictly follow strong hygiene methods. However, controversy exists about the efficacy of this method.

Although documented in rare cases, transplacental transmission of FCoV does not appear to be a problem [Addie & Toth, 1993].

7.8.2 Rescue and boarding catteries

Strict hygiene precautions should be enforced at all times to attempt to minimise viral spread and to keep virus load at a minimum. Ideally, cats should be kept separately. New catteries should be designed with infectious disease control and stress reduction as a priority.

Vaccination of a cat that is unlikely to have been exposed to FCoV, and is entering a boarding or rescue cattery may be considered.

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