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Diagnosis of Feline Viral Infection

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Veterinarians, as well as physicians, constantly search for new, improved, or more rapid and accurate methods for disease diagnosis. Such attempts to improve diagnostic acumen may lead to misunderstanding or misinterpretation of the results of ill-advised laboratory tests. Before requesting any laboratory test, it might be well to repeat the following:

"Why did I request this test? Now that I have the results, I have no idea why I requested it nor do I know what the normal values should be, let alone the abnormal. My books are too old to mention this test. Did I hear a seminar or read an article about this test? I have no such recollection and wouldn't know where to look. Even if I found the reference, I am not sure I would know why I requested the test for this patient. Perhaps I should call the laboratory and ask what the results mean. No, the lab has spent hours running the test, the client has been charged for it, and it might upset them if I exposed my ignorance. I'd better file this report and continue to treat the patient in the usual manner as I would have had I not ordered this test."†

Diagnosis of the virus-induced diseases of the cat involves an understanding of the epizootiology of the diseases in question, the pathophysiologic changes induced by these diseases, and the clinical signs produced by those pathophysiologic changes. In short, it is necessary to know how the virus is spread and, thus, the likelihood of the disease appearing in a particular patient (history), the physical signs of the disease (physical examination), and to utilize specific clinicopathologic tests such as blood counts, immunologic tests, and so on, indicative of pathophysiologic change associated with the disease in order to confirm a diagnosis. There is no "battery" of laboratory tests that will establish a diagnosis without an adequate history and physical examination; yet, a few appropriate tests, properly conducted, may well reasonably confirm a tentative diagnosis that was based on history and clinical signs. It would be unreasonable, for example, to require virus isolation from oropharyngeal swabs of every patient with signs of upper respiratory infection. Calicivirus could probably be isolated from 70 to 80

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†With apologies to Lancet, 2:890, 1957.

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per cent of such patients, but is the calicivirus isolated pathogenic or non-pathogenic? The virus isolated would have to be matched against a battery of antibodies and ultimately specific pathogen-free cats would have to be exposed to the isolate in order to establish its pathogenicity. The patient would have recovered months before this time-consuming and expensive laboratory work had been completed. This may seem a ludicrous example, yet many veterinarians almost routinely request feline infectious peritonitis (FIP) antibody titers on their patients without an adequate understanding of the extreme vagaries of this test. Veterinarians should not have to ask themselves, "Why did I request this test?" after the test has been completed; rather they should ask, "What specific test will either confirm or rule out the tentative diagnosis of the problems exhibited by the patient?".

The cat suffers from a wide variety of viral infections, including parvovirus (panleukopenia), several herpesviruses (rhinotracheitis, pseudorabies, and the cell-associated herpesvirus causing cystitis and urethritis), several species of caliciviruses, retrovirus (feline leukemia), rhabdovirus (rabies), rotovirus, reovirus, and syncytia-forming virus. Many of these viral infections may co-exist in the same host. Calicivirus and herpesvirus infections frequently occur simultaneously. Feline leukemia virus infection produces an immunosuppression, rendering the infected cat highly susceptible to feline infectious peritonitis, calicivirus, or herpesvirus infection as well as bacterial or other infections. Secondary or simultaneous bacterial, protozoan, chlamydial, mycoplasma, or fungal infections may also occur with many viral infections of the cat.

It is not the purpose of this article to describe the pathophysiology and disease signs associated with each of these possible disease entities. That material is well covered in other texts.\textsuperscript{3,7} Gross and histopathologic features, although often necessary for "the final diagnosis," will not be described in detail in this article. In the following pages, specific tests such as fluorescent antibody tests, ELISA, and so forth, will be described briefly, and the methods of diagnosis of the specific diseases will be discussed.

Table 1 lists the common, and some uncommon, viral infections of the cat, the primary methods used to confirm a clinical diagnosis, and other tests that may be useful in diagnosis.

\section*{LABORATORY TESTS}

\subsection*{Virus Isolation}

Although the isolation of a specific virus from a diseased patient conclusively demonstrates that the patient was indeed infected with that virus, it does not conclusively demonstrate that the disease signs observed were indeed caused by that virus infection. Calicivirus, for example, can often be isolated from oral swabs of a cat with hypertrophic gingivitis; it is only too easy to assume that calicivirus infections cause hypertrophic gingivitis in the cat, an assumption that has no sound basis in fact.

It is seldom necessary to attempt virus isolation to confirm a diagnosis in the living individual patient; however, in order to establish the cause of outbreaks of disease in a cattery, pet shop, or multiple-cat establishment,
**Table 1. Laboratory Diagnosis of Feline Viral Diseases**

| DISEASE                              | Primary Diagnostic Tests† | Other Tests                  |
|--------------------------------------|---------------------------|------------------------------|
| DNA Viruses                          |                           |                              |
| Panleukopenia                        | CBC                       | DFA, VI, EM                  |
| Herpesvirus infections               |                           |                              |
| Rhinotracheitis                      | DFA, VI                   | Cyto                         |
| Pseudorabies                         | DFA                       | VI                           |
| Cell-associated herpesvirus          |                             |                              |
| RNA Viruses                          |                           |                              |
| Rabies                               | DFA                       | Cyto, VI                     |
| Calicivirus infections               | DFA, VI                   |                              |
| Reovirus infections                  | Cyto, VI                  |                              |
| Rotavirus infections                 | EL                        | EM, VI                       |
| Feline leukemia complex              | Cyto, IFA, EL             | VI                           |
| Feline infectious peritonitis        | CBC, Cyto                 | VN, FA-antibody, EL-antibody |
| Feline enteric coronavirus           | VI                        | FIP antibody titer           |
|                                      |                           | may cross-react              |
| Syncytia-forming virus               | VI                        |                              |

*CBC = complete blood count including differential count and serum protein; cyto = cytologic examination of blood or impression smears, and so forth; DFA = direct fluorescent antibody test for antigens; EL = ELISA; EM = electron microscopy; IFA = indirect fluorescent antibody test for antigen; VI = virus isolation; VN = virus neutralization; FA-antibody = fluorescent test for antibody; EL-antibody = ELISA for antibody.

†The demonstration of the virus by isolation, DFA or IFA test, ELISA, electron microscopy, and so forth is obviously essential for a "final diagnosis" but may not be necessary for an accurate clinical diagnosis.

virus isolation may be an extremely useful tool. Virus isolation attempts may well be warranted at necropsy if the cause of death was questionable.

Samples submitted from the living patient for virus isolation should be taken during the acute stage of the disease. Samples from necropsies should be collected as soon as possible after death and certainly not more than a few hours after death. Sample selection depends on the suspected clinical disease and may be oropharyngeal swabs, tracheal washings, feces, effusions, biopsied tissues, organs, or those specimens collected at necropsy. It is essential that samples for virus isolation be collected as aseptically as possible. Samples collected should be placed in individual sterile vials without preservative, stored at 20°C (freezer temperature), and shipped to the laboratory on dry ice. Swabs or very small samples should be placed in transport media (with antibiotics) to prevent excessive drying.

Most viral isolation attempts in the laboratory will be on cell cultures; however, the type of cells used and the technique used will vary with the pathogen that is suspected. It is essential that the laboratory technician know what particular virus is being sought. An oropharyngeal swab from a cat with typical signs of rhinotracheitis (herpesvirus infection) may contain both herpesvirus and calicivirus. If such a swab is placed on cultured cells, the calicivirus will often destroy the cell sheet before the typical cytopathic effect and nuclear inclusion of the herpesvirus can be recognized. This example
of an absolute waste of time and effort could have been avoided had the technician known what was suspected.

Laboratories engaged in virus isolation procedures are widespread throughout the country. A practitioner desiring to utilize the virus isolation services provided should call and discuss the case, the samples needed, and the best method for transporting the samples to the laboratory. Virus isolation as a diagnostic technique is valueless when attempted in a haphazard manner.

Cytology

Histologic examination of biopsy or necropsy specimens for characteristic lesions, including inclusion bodies, is a time-honored and effective method of virus disease diagnosis. Cytologic examination of blood or tissue smears, effusions, conjunctival or other tissue scrapings can often be equally effective in disease diagnosis. Conventional staining and microscopic examination of such material can be done in the office laboratory, or air-dried smears may be sent to a commercial laboratory for special staining or immunofluorescent examination.

Electron Microscopy

Electron microscopy provides a useful means for the rapid diagnosis of some feline virus diseases, particularly the enteric disorders caused by agents that are difficult to culture (for example, feline enteric coronavirus and feline rotavirus). A high density of virus must be present in the sample in order to demonstrate virus by electron microscopy, and this limits its usefulness for clinical diagnosis.

Virus Neutralization Test

Although complement-fixing (CF) and hemagglutination-inhibiting (HAI) antibodies may appear in cat serum in response to some feline virus diseases, serum-neutralizing (SN) antibody tests are the most commonly performed of the "conventional" antibody tests. The SN test is based on the fact that specific antibodies (neutralizing antibodies) in the patient's serum will "neutralize" the specific antigen (virus) that was responsible for that antibody production. Dilutions of the patient's serum are added to a known amount of the virus and the mixture is incubated for a period of time that depends on the virus being used. After incubation, the mixture is inoculated onto cell cultures. If neutralizing antibody was not present in the mixture, typical cytopathology caused by the unneutralized virus appears. If neutralizing antibody was present in amounts sufficient to neutralize the virus, no cytopathologic changes are observed. The end point is determined as the greatest serum dilution that reduces the cytopathic effect of the virus by 50 per cent. In most instances, in order to use the SN test as an aid in diagnosis, serum must be drawn early in the course of the disease and again 3 weeks later. A fourfold or greater increase in SN antibody in the second sample is an indication that the patient was indeed exposed to the virus in question. Cats recovering from panleukopenia or calicivirus infection often develop a high SN antibody response, whereas cats recovering from herpesvirus infections develop only a slight SN antibody response. The fact that two samples must be collected at 3-week intervals, the varying test parameters
depending on the antigen in question, and often the vagaries of the test itself severely limit the usefulness of the SN test (or CF or HAI) in diagnosis of viral disease. However, the SN test can be extremely useful in determining the response of cats to vaccination against panleukopenia, calicivirus, and herpesvirus infection and possibly feline leukemia if there are questions regarding the efficacy of the vaccine or the cat’s ability to respond.

**Immunofluorescent Tests**

Immunofluorescent tests have added a new dimension to diagnosis of viral disease. These tests can be used to detect specific viral infections in infected cells in cell cultures, in tissue sections, and in impression smears, and also to detect specific viral antibodies. The direct or indirect procedure may be used. The direct fluorescent antibody technique involves the application of a specific, fluorescein-labelled antibody to the specimen. If that specimen contains the viral antigen, a fluorescein-labelled antigen-antibody complex is formed and will be visible using a fluorescent microscope with the proper filters. The classic example of the application of the direct fluorescent antibody (FA) technique is in the diagnosis of rabies. A fresh section of the hypothalamus is flooded with fluorescein-labelled antibody and examined under fluorescent light. The labelled rabies antibody attaches to any rabies virus present in the specimen and the fluorescence of the labelled antibody-antigen complex is easily visible. If no antigen (rabies virus) is present, the labelled antibody is removed in the washing process and no fluorescence is visible—this is a negative test. This test has reduced the amounts of time spent in the diagnosis of rabies and increased the accuracy of the diagnosis. The FA technique may be used in the diagnosis of panleukopenia (intestinal epithelium, cell culture cells), calicivirus (oropharyngeal cells), herpesvirus infections (oropharyngeal or conjunctival smears), and pseudorabies (brain and/or tonsil sections) as well as rabies.

In the indirect fluorescent antibody (IFA) procedure, virus (antigen)-specific antiserum is flooded over the specimen, the sample is washed, and fluorescein-labelled antiglobulin against the antibody used in the first step is added. The preparation is washed again and then examined under a fluorescent microscope. The classic example of the application of the IFA test in the diagnosis of feline virus infection is the IFA feline leukemia test. An air-dried blood smear from a cat suspected of being infected with feline leukemia virus (FeLV) is treated with acetone to break down the cell wall of the neutrophils and flooded with rabbit (or bovine) serum from an animal immunized against the p27 antigen of the feline leukemia virus. After allowing adequate time for the formation of antigen-antibody complexes, if antigen was indeed present in the smear, the preparation is washed and fluorescein-labelled antirabbit (or bovine) immunoglobulin is added. The preparation is again washed and then examined under a fluorescent microscope. If the p27 antigen of the feline leukemia virus was present in the neutrophils or platelets in the blood smear, the rabbit (or bovine) origin anti-p27 would form an antigen-antibody complex that would not be removed by the first wash. The fluorescein-labelled antirabbit (or bovine) globulin would then adhere to the rabbit (or bovine) globulin complexed with the p27 antigen in the preparation, and the typical fluorescence would be visible
in the neutrophils or platelets on examination under a fluorescent microscope. If no p27 antigen of the feline leukemia virus were present in the neutrophils or platelets in the air-dried slide, no complexes would be formed and the excess rabbit (or bovine) anti-p27 antibody as well as the fluorescein-labelled antirabbit (or bovine) globulin would be eliminated in the washing process and typical fluorescence would not be observed.

If the reagents are carefully prepared and standardized and the tests are carefully performed by experienced technicians, the FA and IFA tests provide a relatively rapid and accurate means of identifying virus antigens in the diagnosis of rabies, panleukopenia, rhinotracheitis, calicivirus infection, pseudorabies, and feline leukemia virus infections.

Immunofluorescent tests can also be used to detect viral antibodies. A modification of the indirect fluorescent antibody technique is used. A cell culture is infected with a known amount of a specific virus, the unknown cat serum is flooded over the preparation, allowed to react, and the preparation was washed. Fluorescein-labelled anti-cat globulin is then added, allowed to react, and the preparation is washed again. If the unknown cat serum contained antibodies against the specific virus, antigen-antibody complexes will be formed in the first step and the fluorescein-labelled anti-cat globulin will adhere to the antibody of the antigen-antibody complex in the second step. Thus, specific fluorescence would be observed in a positive test. If no specific antibody were present in the unknown serum, that serum as well as the fluorescein-labelled anti-cat globulins would be washed off in the washing procedures and no fluorescence would be observed. By utilizing multiple preparations, dilutions of the unknown serum, and establishing positive and negative controls, this type of immunofluorescent test can be used to establish the antibody titer of an unknown serum. Modifications of the immunofluorescent test for antibody are used in determining anti-feline infectious peritonitis (anti-FIP) antibody titers and anti-feline oncornavirus cell membrane antigen (anti-FOCMA) antibody titers in cats. Unfortunately, the tests are difficult to standardize and, in the case of FIP, cross-reactions occur with feline enteric coronavirus antibody. The use of immunofluorescent tests to determine serum antibody titers is of questionable value in the diagnosis of viral disease in the cat.

Enzyme-Linked Immunosorbent Assay

The enzyme-linked immunosorbent assay (ELISA) can be adapted to detect either antigen (as in the feline leukemia virus antigen test) or antibody (as in the feline infectious peritonitis antibody test). The ELISA does not require a fluorescent microscope or other highly specialized equipment for reading. The test can be rapidly performed in an office laboratory and, depending on the type of test being performed, is accurate and economical. The ELISA results can be roughly quantitated based on color change, which eliminates the need for serial dilutions of the unknown sera. The ELISA can be quantitated more accurately by using a special spectrophotometer (ELISA reader); however, this is seldom necessary in the office laboratory.

If the ELISA is to be used to measure antibody levels, the test is generally performed using the patient's serum; however, if the ELISA is used to measure virus antigen (p27 of the feline leukemia virus, for example),
the test may be performed using the patient’s serum, whole blood, bone marrow, effusates, saliva, and so on.

The ELISA basically uses a “sandwich” technique. In testing for the presence of virus antigen—feline leukemia, for example—the walls of the microtiter plate are coated with antibody of bovine origin against the p27 core antigen of the feline leukemia virus (monoclonal antibody). After incubation, the excess antibody is washed away. This is the lower piece of the sandwich. The unknown sample (serum, and so on) is then added to the well, allowed to incubate, and the well is washed again. The unknown constitutes the “filler” of the sandwich. If the specific antigen (p27) is present in the unknown, it will complex with the antibody and not be washed away. An enzyme-antibody conjugate is then added as the top piece of the sandwich and allowed to incubate, and the well is washed to remove any enzyme-conjugated antibody that did not complex to form the “sandwich.” Then one adds an enzyme substrate that, when acted upon, the enzyme present in the top piece of the sandwich will change color. A positive test records a color change; a negative test does not change color. The feline leukemia ELISA, which tests for the presence of the p27 antigen in the unknown sample, is quite sensitive and specific when the test is properly conducted. A similar test, obviously using different antisera in the sandwich, can be used to test for rotavirus in feces; with proper modification, it is possible that other ELISAs will be developed to test for herpesvirus or calicivirus in saliva, panleukopenia virus in serum, and so on.

To test for antibody in serum using the ELISA, the sandwich is reversed. The lower piece of the sandwich is the viral antigen (for example, FIP virus), the “filler” is the unknown serum, and the top piece is an enzyme-conjugated anti-cat globulin. An enzyme substrate is added to produce the color change. The FIP antibody test is perhaps the most often used of the ELISAs measuring antibody. An ELISA test for feline leukemia antibody to whole virus and possibly FOCMA has been developed; however, it is a very crude test that is of questionable significance. An ELISA that measures anti-gp-70 (feline leukemia virus envelope antigen) antibody has also been developed. ELISA tests may be developed to measure other antibodies (for example, anti-FOCMA, anti-panleukopenia, anti-herpesvirus antibodies, and so on) if the demand for such tests is great enough.

**DIAGNOSIS OF FELINE VIRAL DISEASE**

The following paragraphs are devoted to discussion of methods used in the diagnosis of specific feline viral diseases. A tentative diagnosis is based on the history and clinical signs exhibited by the patient. The fact that this statement is not repeated for each disease in the following discussion should not be interpreted to deprecate the significance of the clinical examination in diagnosis of feline viral disease. A final diagnosis of a specific viral disease is based upon a combination of factors, including the history, characteristic clinical signs, and virus isolation or identification by other means. Virus identification, short of actual isolation, may embrace cytologic or histopathologic examination of smears, scrapings, or tissue sections, including the
finding of typical inclusion bodies, fluorescent antibody or ELISA identification of viral antigens, or the demonstration of virus by electron microscopy. Virus isolation or identification by the means described merely indicates the presence, in the patient, of the particular virus isolated or identified. Such isolation or identification does not prove that the problems of the patient are a direct result of damage produced by the virus isolated. It is essential that such isolation procedures be combined with the observation of characteristic clinical signs or pathologic lesions in order to confirm a diagnosis.

It is extremely difficult to evaluate the significance of serum antibody levels, even the demonstration of a rising antibody titer, as determined by virus neutralization, ELISA, or immunofluorescent techniques. The significance of such test results is dependent upon a variety of factors, including the specificity of the test reagents (antibodies against FIP and FECV cannot be differentiated), possible previous exposure to the agent, and the effect of vaccination. Under certain circumstances, the determination of serum antibody levels may be of value in disease diagnosis when correlated with the patient’s history and the clinical signs exhibited; however, as a general rule, such tests are of very limited value in clinical diagnosis. The determination of serum antibody levels is of much greater value in epizootiologic surveys and in assaying response to vaccination.

DNA Virus Infections

Panleukopenia. At the height of disease, there is a pronounced leukopenia, and the few remaining cells are predominantly mononuclear (lymphocytes). Virus can be isolated from the intestinal epithelium, mesenteric lymph nodes, spleen, and perhaps kidneys or identified by fluorescent antibody technique. Intracytoplasmic inclusion bodies are short-lasting and difficult to demonstrate in tissues of a patient that died of panleukopenia. Feline salmonellosis or Escherichia coli infection may also produce a leukopenia; however, the neutropenia tends to be less profound in these infections. The panleukopenic form of feline leukemia closely resembles, in all features, panleukopenia. Panleukopenia vaccination history and ELISA for feline leukemia are valuable in differential diagnosis.

Herpesvirus Infections

Rhinotracheitis. Typical clinical signs of sneezing, conjunctivitis, and rhinitis with profuse mucopurulent ocular and nasal discharges mark severe rhinotracheitis. The presence of corneal ulcers, which are uncommon, with the other signs described is almost pathognomonic of herpesvirus infection. Milder clinical cases may be confirmed by isolation of virus from ocular and oropharyngeal swabs. Calicivirus overgrowth from oropharyngeal swabs may make the isolation of the herpesvirus difficult. Both herpesvirus and calicivirus infections persist in the cat for long periods. Fluorescent antibody may be used to identify herpesvirus from ocular swabs. Conjunctivitis with ocular discharges may be seen with reovirus type 3 infections in cats, Chlamydia psittaci, or Mycoplasma spp. infections. Conjunctival scrapings stained with Giemsa may demonstrate Chlamydia or Mycoplasma. Reovirus can be isolated from the conjunctiva and produces cytoplasmic inclusions in cell cultures.8
Pseudorabies. Naturally occurring cases of pseudorabies have not been described in cats in the United States; however, the cat is susceptible to the disease. The cat may be more resistant to pseudorabies virus than the dog, in which the disease is uniformly fatal. Identification of virus by immunofluorescent testing (brain and tonsil) is preferable to animal inoculation or virus isolation in the diagnosis of pseudorabies.

Cell-Associated Herpesvirus Cystitis. The role of the cell-associated herpesvirus, a calicivirus, and a syncytial virus in the production of cystitis and/or urethritis in the cat is currently unclear. Isolation of virus from urine or bladder mucosal scrapings is difficult and unwarranted as a clinical diagnostic procedure.

RNA Virus Infections

Calicivirus Infection. Many antigenically distinct strains of calicivirus have been isolated from cats. Several of these appear to be nonpathogenic. Different strains have been implicated in a wide variety of disease syndromes, including oral ulcers (tongue and palate), severe and often fatal interstitial pneumonia, a "limping" syndrome, and perhaps cystitis, although the latter is of questionable significance. In the oral and the pneumatic forms of the disease, virus can be isolated from oral swabs and lung tissue, respectively. Serologic testing is necessary to identify the strain of virus involved and is not warranted for clinical diagnosis in the oral ulcerative form of the disease. The virus persists for long periods in recovered cats, so care must be taken in interpreting results of oropharyngeal swabs. An immunofluorescent test is available for identifying calicivirus from oral swabs; however, as with virus isolation, great care must be used in evaluating these tests.

In the limping syndrome, calicivirus (two distinct strains) has been isolated from blood of affected cats. The clinical signs are so characteristic and short-lasting that attempts to isolate virus seem unnecessary for the diagnosis of the clinical disease entity.

Feline Leukemia

Feline leukemia may be manifested as a malignant, proliferative (lymphosarcoma, lymphocytic or myelogenous leukemia, and so on) or degenerative (nonregenerative anemia, thymic atrophy, and so on) disease. Abortion, fetal resorption, or infertility may occur in queens infected with feline leukemia virus (FeLV). Immune-mediated disease (hemolytic anemia, glomerulonephritis) may be initiated by an FeLV infection. The FeLV is immunosuppressive, and this may lead to an increased severity of signs produced by a wide variety of other simultaneous infections in the FeLV-infected cat.

Diagnosis of the malignant proliferative disease caused by the FeLV is based on the demonstration of typical malignant cells in blood smears (Figs. 1 and 2), effusions (Fig. 3), peritoneal washings, biopsies, or necropsy specimens.

Diagnosis of the degenerative diseases caused by FeLV infection may present problems. No easily identifiable malignant cells are present; thus, diagnosis is based on characteristic changes that may appear on blood or
Figure 1. A large lymphoblast in a peripheral blood smear. Significant numbers of such obviously malignant cells justify a diagnosis of lymphocytic leukemia (acute lymphoblastic leukemia).

Figure 2. Large, bizarre cells with eccentric nuclei from a blood smear of a cat with reticulendotheliosis.

Figure 3. Large lymphoblastic-appearing cells from the thoracic exudate of a cat with an anterior mediastinal lymphosarcoma. Note the *Haemobartonella felis* on the few red cells in the exudate—an incidental finding with no significance.
bone marrow smears (anemia, pancytopenia, and so on) and identification of the virus or viral antigen by ELISA, indirect fluorescent antibody (IFA) test, or virus isolation. Although there has been considerable argument on the relative value of the ELISA versus the IFA test for FeLV virus p27 antigen, when carefully conducted on properly prepared specimens, there is very close correlation in results obtained by these tests. A positive ELISA test may indicate a transient FeLV infection that may not be apparent when the IFA test is used. Both tests will be positive in the case of a cat persistently infected with FeLV.

It is essential to realize that a positive ELISA, IFA test, or even virus isolation merely indicates that the patient was infected with FeLV at the time the test was performed. Such a positive test does not indicate that the patient is suffering from feline leukemia. It is very important that the results of the test be correlated with clinical signs and other laboratory tests before a diagnosis of specific disease is made. One must realize also that a negative ELISA, IFA test, or virus isolation indicates only that the virus or the p27 antigen was not demonstrable in the specific sample being tested; the virus or viral antigen may well be present elsewhere in the body. If, for example, a clinical diagnosis of mediastinal lymphosarcoma has been made on the basis of clinical findings and typical cytology seen in a thoracic effusate, it is superfluous to perform an ELISA or IFA test on the patient. A negative blood test performed on the patient could easily be misinterpreted to mean either that the cytologic evaluation is in error or that the patient is not capable of spreading the leukemia virus. Both of these assumptions are likely to be incorrect. It is also important to completely evaluate the disease problems that may be observed in an ELISA or IFA test-positive patient. Such problems may or may not be a direct result of the FeLV infection.

Testing for the presence of antibodies, either anti-FOCMA or anti-gp70 (neutralizing antibody), should be confined to epizootiologic surveys or perhaps for determining response to vaccination. Such tests are of extremely limited value in the clinical patient and results are apt to be misinterpreted.

Feline Infectious Peritonitis

Feline infectious peritonitis (FIP) may produce a wide variety of clinical signs. Diagnosis may be reasonably confirmed by hemogram and, in the case of the effusive form of the disease, by examination of the exudate. The hemogram will often demonstrate a marked rise in serum protein (globulin) and an absolute lymphopenia, in the face of leukopenia or leukocytosis, at the height of disease. The exudate is rather characteristic and contains a high number of neutrophils (Fig. 4).

A cat exposed to FIP virus will usually respond by producing antibodies that may be demonstrated by the ELISA or IFA, or by virus neutralization tests. The cat that develops clinical FIP may demonstrate an antibody titer significantly greater than 1:1600. Unfortunately, there are a number of variables that make interpreting the test extremely difficult, and misinterpretation is extremely common. The tests are not well standardized and a fourfold variation in reported titers may occur; thus, one laboratory may report a 1:400 titer, another a 1:1600, and third laboratory may report a 1:6400 titer on three samples drawn from the same patient at the same time.
Antibodies against feline enteric coronavirus (FECV) cross-react with antibodies against FIP and the ELISA and IFA tests do not distinguish between the two. A cat infected with FECV does not often develop an antibody titer greater than 1:1600, even though it may; conversely, a cat with clinical FIP may not evidence a titer greater than 1:400. Cats infected with FIPV, but not diseased, may develop titers greater than 1:6400. Whether or not cats with high FIP antibody titers but without disease signs are more likely to be FIP virus shedders than cats with lower antibody titers has not be conclusively demonstrated. Taking all of these factors into consideration, it becomes obvious that ELISA and IFA test antibody titers are without significance in FIP diagnosis unless the results are very carefully correlated with the clinical signs of disease and results of other laboratory tests (such as CBC and cytology).

Isolation of FIPV is difficult and often unrewarding, so virus isolation is not a viable means of confirming clinical FIP.

**Feline Enteric Coronavirus Infection**

Feline enteric coronavirus (FECV) infection is widespread in the cat population. The incidence of infection is much greater where cats are congregated, such as catteries, multiple cat households, pet shops, humane shelters, and similar facilities. The disease produced by FECV infection tends to be most severe, although rarely fatal, in 5 to 10-week-old kittens; it tends to be mild in kittens over 12 weeks of age. FECV infection in adult cats is usually inapparent.9

Diagnosis of FECV infection is based on clinical signs, a moderate rise in antibody titer using the FIP antibody titer tests, and virus isolation or identification by electron microscopy of feces.

**Rotovirus Infection**

Rotovirus has been implicated as a cause of diarrhea in young kittens. Widespread incidence of rotovirus infection has been indicated by serologic surveys.6 Virus isolation or identification by ELISA or electron microscopy of feces may be useful in diagnosis. There is a dearth of clinical reports of rotovirus infection in cats.
Reovirus Infection

Reovirus type 3 has been incriminated as causing a mild conjunctivitis and lacrimation in cats. The virus can be isolated from the conjunctiva and oropharynx. The virus produces cytoplasmic inclusion bodies in cell culture and also in infected conjunctival cells.

Rabies

Although rabies may be diagnosed by fluorescent antibody staining of corneal impression smears or skin biopsies, these techniques are usually not warranted because of the public health significance of feline rabies. Rabies virus antigen is detected in the brain most consistently by direct fluorescent antibody (DFA) examination, although conventional stains and virus isolation by animal inoculation (young mice) may be used. DFA examination will demonstrate virus in the brain if the cat is excreting rabies virus in the saliva and before neurologic signs are apparent. The veterinarian's responsibilities and the procedures for shipping suspect animal heads to the laboratory are described in detail in other texts.3,7

Atypical strains of rabies virus do appear outside the United States and may not produce strong immunofluorescence with the conjugates commonly used in the United States.2

Syncytia-Forming Virus (Syncytial Virus)

Syncytia-forming virus has been isolated from normal cats and cats with cystitis, respiratory disease, enteritis, peritonitis, and neoplasms. The virus is widespread in the cat population; however, its significance as a pathogen has not been established. The virus can easily be isolated in cell culture, but inoculation of any of the virus isolates has not produced disease in cats. The virus can be recovered from inoculated cats for long periods of time.

SUMMARY

A diagnosis of a specific viral disease in the cat involves a combination of an accurate history, careful observation of disease signs, demonstration of characteristic clinical pathologic changes, and isolation or identification of the virus. Isolation or identification of a virus from the patient does not establish that the disease observed was caused by the virus so isolated or identified; correlation and proper interpretation of all findings are necessary to establish a diagnosis.

Virus identification may involve office laboratory tests, such as cytology or ELISA, or more specialized procedures. Whether specimens are to be sent out for specialized tests or office laboratory procedures are to be used, the veterinary practitioner must not only know what specimens are required but must also understand the test and be able to properly interpret the results in light of the patient's observed condition.

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