FELINE INFECTIOUS DISEASES
UPDATE

Feline Specialist Seminar

Cornell University

August 7 - 11, 1989

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FELINE INFECTIOUS PERITONITIS

I. REVIEW OF THE DISEASE

A. Overview

Feline infectious peritonitis (FIP) is THE most feared disease today in breeding catteries. It is less common and of less concern in the general pet population. There are at least five reasons why FIP is so feared in breeding catteries.

First, there is no effective treatment for FIP. Once clinical disease develops, nearly 100% of cats will eventually die of the disease. Second, there is no preventative vaccine. Despite extensive efforts to develop effective and safe vaccines, no commercial vaccine is available to protect cats. Third, there is no diagnostic test available to accurately diagnose FIP. While there are several coronavirus antibody tests available, none are specific for FIP, and a positive test only indicates previous exposure to a coronavirus, possibly feline infectious peritonitis virus (FIPV). Fourth, there is no practical, effective way to handle a cattery enzootic. It is not possible or practical to identify potential virus-carrier cats, and the coronavirus antibody tests that are available do not predict what will happen to an individual cat, whether antibody positive or antibody negative. Lastly, the incubation period may be weeks, months, or even years. Cats may become infected with the virus without showing clinical disease, then after a protracted period of time, clinical FIP may develop. This makes it extremely difficult to live with chronic FIP in a cattery. Kittens may become infected when young, but clinical disease may not occur until after the kittens are sold and enter other households or catteries.

B. Pathogenesis

The incubation period from the time of initial exposure to virus and the development of classical signs of FIP may be weeks, months, or even years. Generally, it is two to three weeks. We have an experimental case in a controlled, isolation situation where the incubation period was 8 months from the time of exposure to virus to the development of clinical signs.

Local infection or primary infection occurs in the pharyngeal and lung epithelium, and perhaps the intestinal epithelium as well. Minimal clinical disease occurs during the primary infection, often just a transient fever for one to a few days.

Antibodies against FIPV first appear in serum by day 7 to day 10 after infection. Antibodies allow infection of macrophages with virulent FIPV through a process of immune enhancement. Infected macrophages in turn transport the virus throughout the body. Secondary infection then occurs in many tissues, with macrophages attaching to and migrating through the walls of veins. A perivascular reaction occurs, leading to development of a pyogranuloma, the basic lesion of FIP within tissues. In wet FIP there is an exudative reaction at the vessel walls, with exudative fluid accumulating in the peritoneal and/or the thoracic cavities.

As the disease progresses, there is an insidious increase in clinical signs due to anorexia, weight loss, persistent fever, and depression. Death occurs in most affected cats, with the duration of the disease lasting from a few days to several months.

C. Wet vs. Dry Forms of FIP

The relative incidence of wet and dry forms of the disease has changed since FIP was first described. In early reports, most cases that were diagnosed were the wet form of the disease, i.e., there was a pronounced accumulation of fluid in body cavities, especially the abdominal and thoracic cavities. The wet form may have actually been more common, or it may be that the dry form of the disease was not diagnosed.

At present, the dry form of FIP is more common than the wet form. This may be due to more accurate diagnosis of dry cases, or it may be an actual increased incidence of the dry form.

It should be pointed out that the wet and dry forms of FIP are merely variations of the same disease process. The same virus strain may produce both types, while other strains tend to produce only dry cases. An individual cat may have transient wet form early in the disease, then the disease progresses to the dry form only.

D. Virus Shed

The period when FIPV is shed after infection extends from about day 1 to day 10 after infection. Clinical disease usually is not present when the cat is shedding virus. By the time most infected cats develop clinical disease they are no longer shedding virus in their feces or secretions. Virus may persist within the cat in an intracellular location, but most cats have sufficient neutralizing antibodies so that virus is neutralized as soon as it is released from the infected cell. Although it has not been documented, it is possible that transfer of infected cells from one cat to another could occur in some cases even after they no longer are shedding extracellular virus. If reinfection of a FIP antibody-positive cat occurs, infection with clinical disease may proceed without virus being shed from the cat.

Isolation of virus from clinical cases of FIP is very difficult. In the vast majority of cases, diligent virus isolation attempts are fruitless, even with strains of virus that replicate readily in cell culture. In an occasional case, FIPV may be isolated using special techniques such as co-cultivation of peripheral blood leukocytes with regular cell cultures.

E. Relationship of FIP to Feline Leukemia Virus (FeLV) infection and Feline Immunodeficiency Virus (FIV) infection

All three viruses are distinct and produce clinical disease by themselves, without help from other virus. Both FeLV and FIV are immunosuppressive viruses. Thus they can predispose cats to other opportunistic infections, and they can cause a recrudescence of latent infections.

Originally, 40 to 50 % of FIP cases occurred in FeLV-positive cats. Today, far fewer cases occur in FeLV-positive cats. Many catteries experience FIP without either FeLV or FIV being present in the cattery. FeLV, if present, may exacerbate a mild or subclinical infection of FIP, causing a severe and fatal infection.

There does not appear to be a predisposition of FIV to cause FIP. The incidence of feline coronavirus antibody-positive cats is the same in FIV-negative and FIV-positive cats.

II. REVIEW OF THE VIRUS

A. Properties of FIP Virus

FIPV is a coronavirus, a pleomorphic enveloped virus containing single-stranded RNA. Coronaviruses contain three major proteins, N, E1, and E2. The N or nuclear protein occurs in the core of the virus and is composed of only polypeptides. The E1 and E2 proteins are glycosolated proteins associated with the envelope of the virus. The E2 glycoprotein makes up the projections or peplomers extending outward from the envelope, and is associated with attachment to receptors on susceptible cells.

Replication of FIPV occurs at the endoplasmic membranes as the virus buds into vacuoles within the cell cytoplasm. Virus remains cell-associated, only being released outside the cell after the cell is destroyed. Replication of virus is rapid, with the complete cycle requiring less than 24 hours.

FIPV appears to survive in the environment much longer than was originally thought. Studies in our laboratory have shown that infectious virus can be recovered from contaminated dry surfaces for 3 to 7 weeks. The amount of infectious virus recovered decreases with time. Relatively large quantities of virus are required to establish infection in a susceptible cat, and, therefore, in natural situations, FIPV probably is not infectious to cats longer than 2 or 3 weeks after an environment, cage, food or water dish is contaminated.

B. FIPV vs. FECV

There are differences of opinion concerning the relationship between FIPV and feline enteric coronavirus (FECV). It is the authors belief that these are not different viruses, but merely biotypes of the same virus. There are minor differences in the genome of the two viruses, and they do have a different ability to infect cells. FIPV has a greater affinity to infect macrophages, whereas FECV has little affinity or ability to infect these macrophages. There is a direct correlation between the ability to infect macrophages, with immune enhancement, and the virulence of the strain of FIPV.

C. FIPV and Other Coronaviruses

FIPV belongs to an antigenic cluster of viruses that includes transmissible gastroenteritis virus (TGEV) of swine, canine coronavirus (CCV), and human bronchitis virus or human coronavirus (HCV) strain 229E. Cats can become infected with TGEV, CCV, or HCV-229E, with development of antibodies, but without development of clinical disease. Antibodies against these three viruses do not resulted in immune enhancement or sensitization of cats to subsequent infection with FIPV.

D. Type 1 and Type 2 of FIPV - Do They Really Exist?

Again there is a difference of opinion, but it is the author's view that these are different biotypes of virus, not different types of FIPV. There is no evidence to date that multiple serotypes of FIPV exist. There is, however, a wide range of strains of coronaviruses that infect the cat, including variations in virulence and ability of virus to replicate in cell cultures.

III. DIAGNOSIS

The diagnosis of clinical FIP depends on clinical signs, clinical pathological findings, an evaluation of abdominal and/or thoracic fluid if present, serological assays for the presence of coronavirus antibodies, and histopathological findings.

Clinical signs associated with FIP include the following: (1) a persistent, non-responsive fever, (2) a gradual weight loss, (3) progressive anorexia and depression, and (4) various other clinical signs, depending on location of lesions. With fluid accumulation in the abdominal cavity, moderate to marked ascites occurs. If the fluid is in the thoracic cavity, progressive dyspnea occurs. Involvement of the kidneys can result in signs of renal failure and toxicity, while involvement of the central nervous system produces various neurological signs. Liver disease is a common occurrence in FIP.

Clinical pathological results may include an elevated total serum protein, elevated liver enzymes, elevated bilirubin, and an altered electrophoretic pattern of the serum globulins. Leukocyte counts are variable and thus not predictive of FIP.

Evaluation of abdominal and/or thoracic fluid, if present, should always be done. In most cases of wet FIP, this evaluation of fluid can reasonably confirm the diagnosis of FIP. Fluid is generally characterized as having a viscid, egg-white consistency, with flecks of fibrin floating within the fluid. FIP fluid usually will clot on standing, and it usually will have a high specific gravity (above 1.018 and often above 1.030).

Considerable controversy exists concerning the serologic tests for FIP. The available tests are antibody assays which detect antibodies against various coronaviruses. As such, they are not specific for FIP, but when used and interpreted appropriately, they can be helpful to the process of diagnosing FIP. None of these tests, however, are diagnostic in themselves -- that is, one cannot confirm absolutely the diagnosis of FIP based solely on a positive coronavirus antibody titer. Positive coronavirus antibody tests, however, are consistent with FIP, but some severe cases of clinical FIP will have negative antibody titers.

At present, histopathology is the most accurate method of confirming a diagnosis of FIP. Lesions consisting of typical pyogranulomatous reactions are diagnostic for FIP. Samples examined may include biopsies of affected organs, or tissues with lesions collected at necropsy.

IV. SEROLOGICAL TESTS

A. Tests Available

There are several antibody tests available for assisting in the diagnosis of FIP. Most tests are enzyme-linked immunosorbent assays (ELISA). These may be heterologous tests (using TGEV or CCV), or they may be homologous assays (using FIPV) as the test antigen. Some of these assays are kinetic ELISAs (KELA) which quantitate the reaction. FIP virus neutralization assays can also be used to measure the titer of antibody present in the test serum.

B. How Accurate are the FIP Tests?

As mentioned above, as far as the author is aware, the available tests are not diagnostic by themselves. They only measure coronavirus antibody, not antibody specific for a certain strain of FIPV.

C. Antibody Titer - What does it mean?

The height of coronavirus titers are greatly overemphasized. Coronavirus antibody titers in healthy cats have little predictive or diagnostic value. They are either "positive" or "negative", and will only tell if cats have been infected with a coronavirus sometime in the past.

The level of titer in a sick cat has more predictive value. A titer >1:400 is consistent with FIP, but still is not diagnostic. A titer <1:400 indicates that the disease in question probably is not FIP in most cases, but some cases of fatal FIP have negative or low titers. When in doubt, rely on clinical signs and clinical pathological results rather than the results from coronavirus antibody tests.

D. To Test, or not to Test? That is the Question!

The coronavirus antibody tests should be used in many cases, but they should not be over-interpreted. They should be used as screening tests to determine the presence of coronavirus in a group of cats, and they also should be used as an AID (and only an aid) in diagnosing clinical FIP.

Repeat testing in a FIP antibody-positive cattery or multicat household is not recommended or warranted. Titers will fluctuate naturally, and owners may believe that active FIP will result in a cat that has a slight increase in titer.

V. IMMUNOLOGY OF FIP

A. Humoral Immunity

Serum virus neutralizing antibodies first appear about 7 to 10 days after infection. The antibody titer gradually increases until it reaches a maximum titer at 5 to 6 weeks after infection. Non-neutralizing antibodies also may be present, and these probably contribute to the hypergammaglobulinemia that is usually present in FIP. This hypergammaglobulinemia appears to be an over-stimulation of gammaglobulins in a futile attempt to provide protection against the fatal disease process.

B. Cell-Mediated Immunity

Cell-mediated immunity is believed be play an essential role in those cats that develop an effective immune response against FIPV. Many cats exposed to attenuated or low virulent strains of FIPV will develop high neutralizing antibody titers, but these cats may not be protected against subsequent exposure to homologous or heterologous strains of virulent FIPV. Much research is needed to elucidate cell-mediated immunity in cats against many disease agents, including FIPV.

C. Immune Enhancement

Immune enhancement of infection has been documented with certain strains of FIPV. These strains of virus "sensitize" cats to subsequent exposure to FIPV rather than provide protection as is the norm for most viral infections. This immune enhancement is associated with enhanced infectivity of large monocytes or macrophages, probably by enabling immune complexes of virus-antibody to attach to Fc receptors on the surface of the macrophages, followed by infectivity of these macrophages without inactivation of the virus. The infected macrophages then transport the virus throughout the body as an intracellular viremia, thus rendering circulating neutralizing antibodies useless to control the infection. Immune enhancement results in decreased incubation time - as short as 1 to 2 days - after exposure to virulent FIPV.

D. Role of the Macrophage

Infectivity of macrophages appears to be the key to the ability of FIPV to become systemic and thus to develop perivascular lesions and clinical FIP. Serum antibody to coronavirus appears to enhance the infectivity of macrophages by the virulent strains of FIPV, but not the avirulent strains such as the strains of FECV.

VI. ATTEMPTS AT IMMUNIZATION

For many years investigators have attempted to develop effective and safe vaccines for FIP. Many different approaches have been tried, but because of the uniqueness of the pathogenesis and immunity of this disease, commercial vaccines have not been developed.

Attempts to develop inactivated whole-virus vaccines have been unrewarding to date. Similarly, attempts to develop subunit vaccines have failed. Cats vaccinated with inactive products may develop an antibody response, but these cats are not protected against challenge with virulent FIPV. Usually these vaccinated cats have experienced an immune enhancement of infection rather than protection.

Attempts to shown efficacy from heterologous live-virus vaccines using CCV, TGEV, or HCV-229E have failed. Cats vaccinated with these viruses develop neutralizing antibody titers against the vaccine virus, but there does not appear to be any protection provided against FIP.

Research to develop modified live-virus (MLV) or attenuated FIP vaccines has met with more success than with inactivated vaccines. Virulent strains of FIPV have been attenuated by rapid passage in cell cultures, and experimental vaccines have been prepared from these attenuated strains of virus. However, these attenuated experimental vaccines have had varying degrees of safety and varying degrees of efficacy. Some vaccinated cats have develop solid and complete protection against challenge with highly virulent strains of FIPV, while other vaccinated cats develop high neutralizing antibody titers but without any degree of protection. The reasons for this inconsistent protection are unknown.

VII. TREATMENT

Treatment for FIP is almost invariably discouraging. There are no specific antiviral compounds available for treatment of affected cats, although research is ongoing to evaluate various antiviral compounds. Treatment then is aimed at support and possibly alteration or modification of the immune response in the cat since FIP is an immune-mediated disease. There is an urgent need for more effective treatment for this disease.

Good nursing care is generally the most import aspect of therapy for FIP. A small percentage of cats do survive this infection, and this occurs only in cats that continue to eat. Once debilitation sets in, the outcome of FIP is invariably fatal.

Cats that develop only localized and limited pyogranulomatous lesions may survive the disease. This is especially true where cats only have ocular involvement. These cats usually have very high antibody titers. Local or subconjunctival therapy with corticosteroids appears to provide beneficial results in cases limited to ocular FIP.

While antibiotics are generally prescribed for cats with FIP, there is no indication that they are of any help. Bacterial infections do not occur in most cases of FIP, and antibiotics have no effect on the virus.

Various immune modulators have been suggested from time to time for therapy of cats with FIP. Since FIP is an immune mediated disease, and since there appears to be an aberrant immune response following infection, immune modulation therapy theoretically might be beneficial. Unfortunately, to date, we have not been able to establish a beneficial regime of therapy using immune modulators. If therapy is to be effective, it may require combination therapy, perhaps with some interferon product and an immune modulator. Further work must be conducted in this area.

VIII. WHAT DOES THE FUTURE HOLD FOR FIP?

There is urgent need for an effective vaccine, for an effective treatment, and for an accurate diagnostic test. While research to date has not been successful in any of these areas, one has to be optimistic that at some point success will come.

One area of possible success in vaccine development may be in the use of recombinant carrier vaccines. It may be possible to stimulate an effective cell-mediated immune response without sensitizing the cat by use of such a product.

Until success occurs in at least one of the above three areas, prevention, treatment, or diagnosis, FIP will continue to be a frustrating disease to deal with, and it will continue to be the most feared disease experienced by cattery owners. It will require a breakthrough in one of these areas in order to develop an effective way of managing a FIP-positive cattery or multicat household.

FELINE LEUKEMIA VIRUS INFECTION

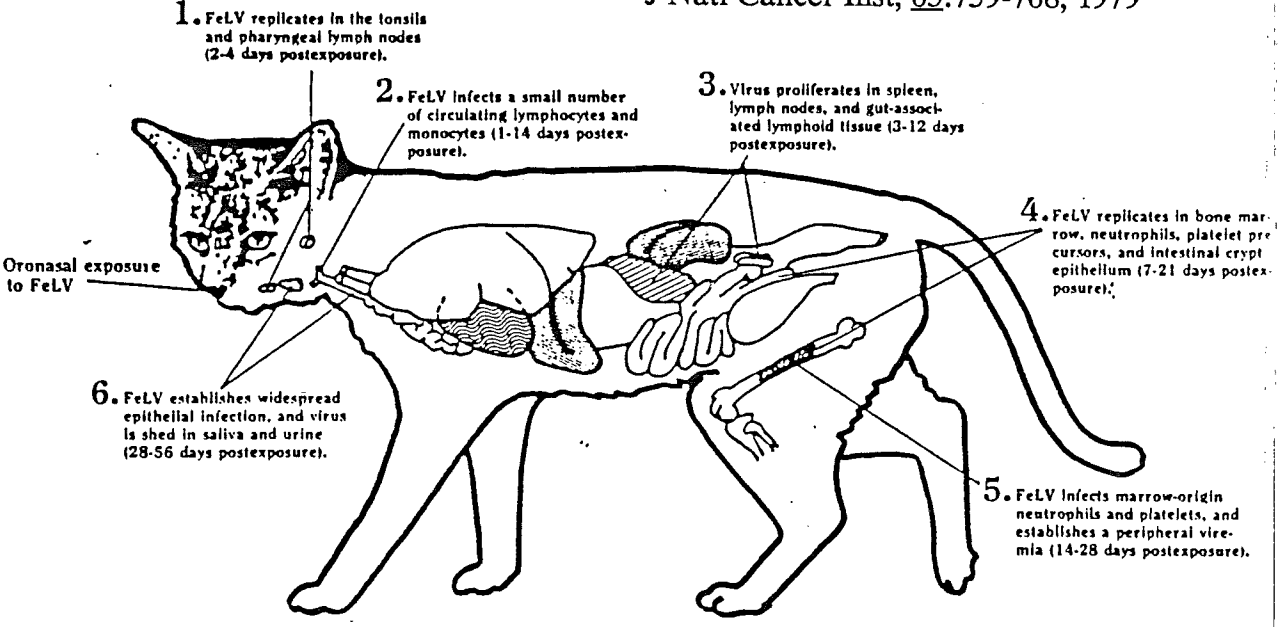
I. INTRODUCTION

Feline leukemia virus (FeLV) has received considerable research attention in recent years. From the isolation and identification of a virus as the cause of the disease in 1964 until the introduction of the first commercial vaccine in 1985 a wealth of scientific information has been accumulated which has advanced the science of both feline medicine and human medicine. This discussion will address the current knowledge of FeLV vaccines, vaccine recommendations, and the commercial FeLV diagnostic tests and how they apply to FeLV vaccination.

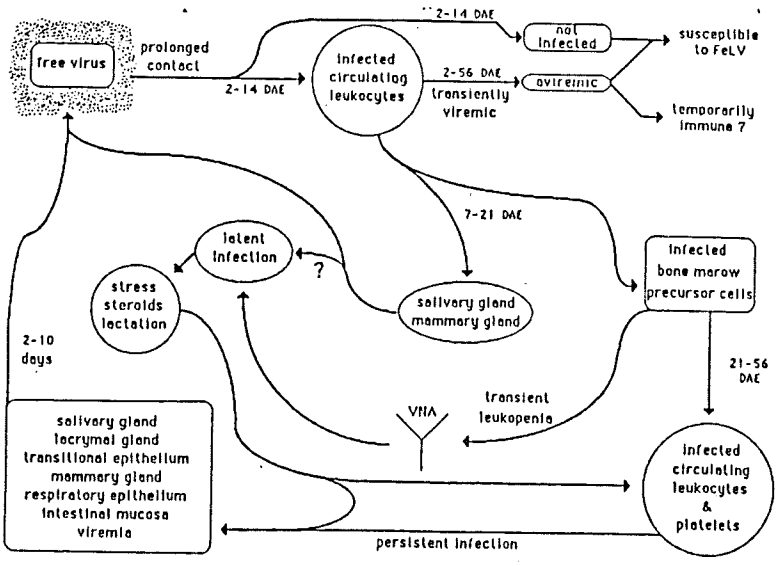
II. PATHOGENESIS

A. Rojko studies

Rojko et al., Pathogenesis of FeLV infection. J Natl Cancer Inst, 63:759-768, 1979



B. Lopez diagram



III. IMMUNITY

There is fairly uniform agreement that virus neutralizing antibodies must be present in a cat in order to protect that cat against virus infection with FeLV and the development of persistent viremia. Antibodies stimulated in the cat against gp70 are virus neutralizing antibodies. There is not uniform agreement amongst scientists concerning the necessity or role of FOCMA antibodies in preventing the development of the various forms of neoplasia associated with FeLV infection.

IV. FELV VACCINES

A. Leukocell (Norden)

The first commercial vaccine (Leukocell, Norden Laboratories) was approved in 1985, and is an inactivated "subunit" vaccine containing an adjuvant. It contains at least 2 antigens, the gp70 envelope antigen and the cell membrane antigen FOCMA (Feline Oncornavirus Cell Membrane Antigen).

Leukocell vaccine is prepared from suspension cultures of a persistently infected cell line of transformed lymphoid cells (FL-74) with subgroups A, B, and C of FeLV. Cells are grown in a fermenter in such a way that expression of antigen subunits are maximized. These subunit antigens are clarified and inactivated, preservatives are added, the antigens are quantitated and standardized, adjuvants are added, and potency release tests are run.

Recently, Norden has release a two-dose, subcutaneous vaccine, "Leukocell-II", which is reported to have a six-fold increase in antigenic mass of FeLV antigens.

According to the manufacturer, Leukocell aids in the prevention of:

1. Persistent viremia
2. Lymphoid tumors
3. Diseases associated with FeLV infection

B. Covenant (Diamond Scientific)

Covenant FeLV vaccine, introduced in 1988, is a chemically inactivated, whole-virus vaccine. It is prepared by propagating the "LUNA" strain of FeLV in a nontransformed chronically infected normal feline cell line, standardizing each serial to optimize the whole virus titer, chemical inactivation of the virus, and addition of the adjuvant "CASCADE". Diamond recommends for primary immunization two doses of Covenant 3-4 weeks apart starting at 10 weeks of age, with annual revaccinations using a single dose of vaccine.

C. Safety

Safety of Leukocell was evaluated in experimental situations and in a fairly extensive field trial prior to licensure of the vaccine. Although the vaccine appears to be safe from producing FeLV or FeLV-related disease in vaccinated cats, it is clear that the vaccine is not without side effects in some cats. During the field trial, the following postvaccination reactions were reported: "9.2% rate of injection site pain or stinging; 3.6% rate of lethargy, inappetence, or pyrexia occurring 6 to 12 hours postvaccination and lasting 6 to 24 hours; and a 0.4% incidence of hypersensitivity

reactions characterized by vomiting, myxedema, or cyanosis." Because of these adverse reactions, the manufacturer made two changes in the production process, an addition of another purification step, and the removal of a preservative. Reported reaction rates in serials of vaccine produced after these changes are substantially reduced. Reports from veterinarians confirmed this reduction in reactivity of the host to the vaccine.

Initially, reactions to Covenant after vaccination appeared to be similar to those experienced with Leukocell after its introduction. Steps were taken by Diamond to reduce reactions caused by the vaccine, and it appears that there is now little difference in safety between Leukocell and Covenant.

D. Efficacy

In the experimental trial conducted by the manufacturer of Leukocell (25 vaccinates and 10 nonvaccinates), 80% of the vaccinates did not develop persistent viremia after challenge with virulent FeLV, whereas 70% of the nonvaccinates did develop persistent viremia. 92% of the vaccinates remained tumor free during the course of the observation period, whereas 60% of the controls developed tumors.

Pedersen et al. (1985) stated that "Leukocell failed to perform as well as claimed by the manufacturer." "Only 13% of cats given two doses and 50% of cats given three doses of Leukocell developed good ELISA FeLV-gp70 antibody titers." "Most vaccinated cats developed FOCMA antibodies after two or three doses of Leukocell." "With respect to the actual efficacy of Leukocell in preventing latent or active FeLV infections, two doses of Leukocell given according to the manufacturer's instructions failed to induce any immunity whatsoever against experimental FeLV infection." "The level of protection provided by three doses of Leukocell appeared to be considerably below the 80% level reported by the manufacturer."

Drs. Janet Scarlett and Roy Pollock at Cornell are currently conducting a two-year study on the efficacy of three-dose Leukocell in a very large colony of cats with a high incidence of persistently infected cats. This study involved approximately 45 cats in each of three groups housed together in one large room. One group consisted of Leukocell-vaccinated FeLV-negative cats, a second group consisted of placebo-vaccinated FeLV-negative cats, and the third group was made up of persistently-infected FeLV-positive cats. Fifteen weeks after the start of the vaccination program, the FeLV positive group of cats was housed with the vaccinated cats. At the conclusion of one year, 15 of 40 remaining placebo-vaccinated cats became FeLV positive, while 5 of 40 remaining Leukocell-vaccinated cats became FeLV positive. This provides an efficacy of approximately 67 percent. The efficacy at the conclusion of two years is currently being evaluated. (Results reported at CRWAD, Chicago, Nov. 1988.)

Diamond Scientific reported at the introduction of Covenant that 105 of 124 vaccinated cats (85%) in a total of 8 trials were protected against a challenge with a virulent strain of FeLV, while 50 of 77 (65%) of nonvaccinated control cats developed persistent viremia and death associated with FeLV infection.

Once cats have been infected with FeLV the vaccines will not alter the course of events. The FeLV vaccines do not cause adverse effects when given to FeLV-positive cats, but because of their lack of beneficial effects on these cats, FeLV-positive cats should not be vaccinated.

The FeLV vaccines have been evaluated singularly and with other feline vaccines such as rabies, panleukopenia, herpesvirus, and calicivirus. While the manufacturers claim lack of complications or adverse effects from the simultaneous vaccination of cats with these vaccines and the FeLV vaccines, several clinicians believe they do see more adverse reactions with simultaneous vaccination. These clinicians use a staggered vaccination schedule in which FeLV vaccine is administered alone 2 weeks from when other vaccines are given.

E. **Vaccine guidelines and recommendations**

The following are guidelines and recommendations for FeLV vaccination

1. **The decision of whether or not to vaccinate is left up to the veterinarian and the owner. Cats which have the greatest potential for infection should be vaccinated (e.g. show cats, shelter cats, negative cats going into multiple cat households, outdoor cats).**

2. **Vaccination should begin at 9 to 10 weeks of age, with a second dose of vaccine given 3 to 4 weeks later. Annual revaccinations are recommended.**

3. **Before vaccination or at the time of vaccination, if there is any question about the FeLV status of the cat to be vaccinated, the cat should be tested for FeLV.**

4. **If the initial blood test is positive for FeLV antigen, do not vaccinate or discontinue the vaccination program and retest in 3 to 4 weeks. Also do not vaccinate cats that are pregnant.**

5. **If the second test is negative the cat probably has experienced a transient viremia and may now be naturally immune; however, vaccination should be initiated or resumed to further booster immunity.**

6. **If the second FeLV test is positive, the cat is persistently viremic and should be handled accordingly. Vaccination of positive cats has no detrimental or beneficial effects.**

There is no evidence that Leukocell or Covenant FeLV vaccines can reverse an established FeLV viremia, or alter the clinical course in viremic cats. Neither is there evidence that the vaccines produce any greater untoward effects in FeLV-positive than FeLV-negative cats.

Under natural conditions, many FeLV-exposed cats experience a transient viremia followed by an immune response that eliminates viremia, the cat reverts to FeLV-negative status and is thought to be FeLV-immune. However, since the actual anti-FeLV or anti-FOCMA antibody titers in these cats are seldom known, it seems most prudent to proceed with immunization and boost whatever naturally acquired immunity may be present.

F. Other considerations

Leukocell and Covenant are first generation vaccines which provide for the first time the possibility of preventing FeLV infection by any practical means other than environmental isolation of susceptible cats. According to the manufacturers, approximately 80-85% of vaccinated cats remained healthy after an experimental challenge with a large dose of virulent FeLV in conjunction with corticosteroid-induced immunosuppression. Protection, therefore, is not complete.

The ability of Leukocell or Covenant to prevent latent FeLV-negative infections or the effect of vaccinating latently infected cats are unknown. Furthermore, the actual clinical significance of latent FeLV infection remains incompletely understood. Latent FeLV infections are not detected by routine ELISA or IFA procedures and therefore it is certain that some FeLV-negative, but latently FeLV-infected cats, will be vaccinated.

The FeLV vaccines themselves will not produce a FeLV-positive test, nor do they contain infectious FeLV.

Finally, while Leukocell and Covenant provide useful aids in reducing the incidence of FeLV infection in cats, their use should not provide a false sense of security. Protection is not absolute. Vaccination should add to, but not replace, existing test and removal or isolation programs for FeLV-infected cats.

V. FELV DIAGNOSTIC TESTS

There are several tests available to detect current or previous FeLV infection. Most tests detect the presence of virus in the blood, saliva, or tears, but tests are also available to detect antibodies to the virus as an indication of previous infection or vaccination.

A. Types of tests

There are 2 basic types of tests used to detect the p27 core antigen of FeLV.

1. *Indirect immunofluorescent assay (IFA, slide test, or Hardy test).* This was the first test developed for FeLV, and detects virus in blood leukocytes and platelets. A blood smear is tested using antibodies specific for the p27 core antigen. A positive test indicates a high probability of persistent viremia and persistent shedding of virus in the saliva and other secretions.

2. *Enzyme-linked immunosorbent assay (ELISA, kit test).* There are several kit tests designed for rapid in-house testing for FeLV. These tests detect virus by identifying reaction to the p27 core antigen in blood, serum, plasma, saliva, or tears. They are very sensitive and detect minute quantities of virus. They also detect early or primary viremia that occurs in all cats that are infected with FeLV, even those that develop an effective immune response. Some cats that develop sequestered virus but not persistent viremia may also test positive with the ELISA tests, but negative with the IFA test. Most if not all of these tests use mouse monoclonal antibody produced against the p27 antigen, and this mouse monoclonal antibody usually is obtained from a single laboratory.

There are several ELISA tests that use some form of well in which the tests are run. The antibody is adhered to the plastic well, then the test serum is added and allowed to react such that a specific antigen-antibody reaction occurs. An enzyme that is tagged to the antigen-antibody complex, and substrate is added that produces a color reaction that is detected visually. Because of the great sensitivity and the possible variability in running and interpreting the tests, it is imperative that positive and negative controls be run at the same time as the test samples. If the controls do not react properly, then the whole assay is void.

A variation of the ELISA test is the CITE test in which the monoclonal antibodies are impregnated into a disc at the top of an absorbent chamber. This unit contains a disposable filter top through which the whole blood is filter. After appropriate washes the reaction appears as blue dots on the membrane. Positive and negative controls are incorporated into the membrane.

B. Lopez/Jacobson studies

A comparative evaluation of the sensitivity and specificity of seven commercial FeLV ELISA kit tests has been done by Drs. Noel Lopez and Richard Jacobson at the New York State Diagnostic Laboratory, in conjunction with the Feline Health Center. The results of this comparison were included in Dr. Lopez's PhD thesis (May 1989), and will be published in the next few months.

1. References

a. Lopez, NA et al. Sensitivity and specificity of seven blood test kits for FeLV infection. JAVMA 195 (Sept. 1989).
b. Lopez, NA et al. False positive reactions associated with anti-mouse activity in serotests for feline leukemia. JAVMA 195 (Sept. 1989).
c. Lopez, NA et al. Sensitivity, specificity, and predictive values of ClinEase-Virastat saliva test for FeLV infection. Cornell Vet. (in press, 1989).

2. Sensitivity

The studies on the sensitivity (the probability of correctly identifying true-positive animals) of the 7 kit tests evaluated by Lopez et al. indicated that all seven kits were highly sensitive. All kits were 100% sensitive in detecting sera from cats presented to the NYSCVM that were IFA positive for FeLV. However, when tested against purified FeLV p27 antigen, considerable variation occurred in the sensitivity. Three kits detected 100% of samples with as little as 28 ng/ml of antigen, while one kit was unable to detect 225 ng/ml. The sensitivity of other kits was intermediate.

3. Specificity

Studies on the specificity (the probability of correctly identifying true-negative animals) of the 7 test kits resulted in more clearly defined differences than for sensitivity. FeLV IFA-negative cats were correctly identified by all of these kits, unless anti-mouse antibodies were present. Three kits correctly identified FeLV IFA-negative, anti-mouse-positive samples as "negative", while 4 kits called these sera "positive".

4. Cat anti-mouse antibodies

Approximately 0.14% - 0.57% of the cat population has anti-mouse antibodies in their bloodstream. Approximately 1 in 283 cats tested by ELISA will result in "FeLV false positive" results, unless the manufacturer of the kit uses specific steps to avoid these false positive results. Sera from these cats contain cat anti-mouse antibodies which reaction to the mouse anti-FeLV monoclonal antibodies in the ELISA kits. The reason cats contain these anti-mouse antibodies is not known -- but certain vaccines will elicit this anti-mouse antibody response.

5. Saliva test

ClinEase-Virastat saliva test (Synbiotics Corp.) had high sensitivity (>92%) in relation to both IFA and plasma ELISA, with 34/34 IFA-positive cats testing positive, and 39/42 (93%) ELISA-positive cats testing positive by this saliva test. Concerning specificity, 55/65 (85%) of IFA-negative cats and 55/60 ELISA (92%) tested negative by the saliva test.

6. Discordant results

The serum ELISA almost always agrees with IFA-positive results, but about 6% of serum ELISA-positive cats will test IFA negative.

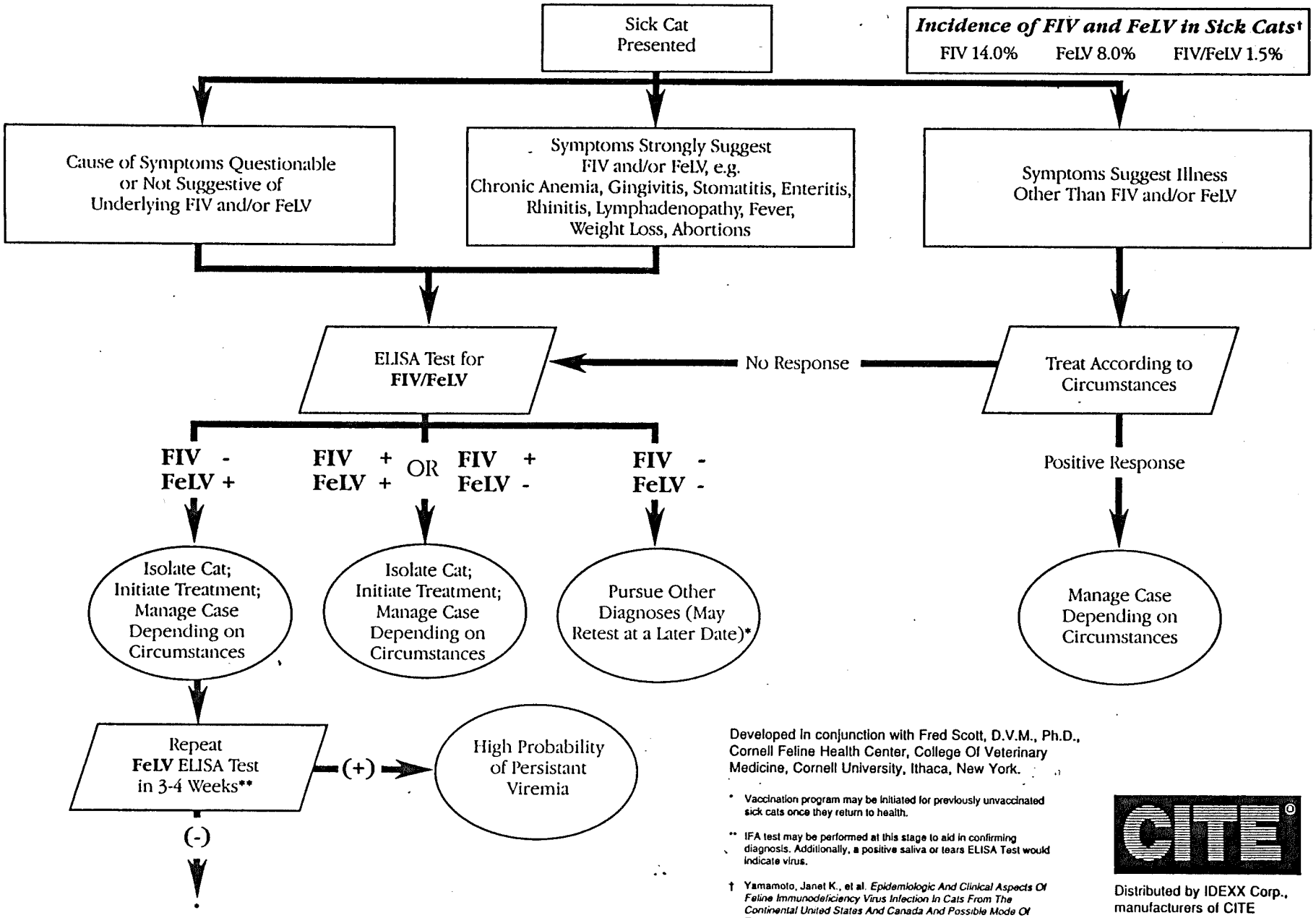
The high sensitivity of ClinEase-Virastat make it a good screening test to rule out FeLV, but the relative large number of IFA- and serum ELISA-negative cats that test positive by saliva ELISA requires that these saliva ELISA "positive" cats be retested by another test before they are declared "persistently viremic".

C. FeLV tests recommendations

We suggest use of one of the ELISA kits as a prevaccination or diagnostic screening test for FeLV. These tests can detect cats in the incubation period of the infection prior to the bone marrow stage of infection and a positive immunofluorescence (IFA) test. The ELISA will also pick up those "test-discordant" cats which remain persistently ELISA-positive but IFA-negative. ELISA kits occasionally can give false positive reactions due to operator error or slight non-specific reactions. ELISA-positive cats should always be rechecked in 3-4 weeks, and in our opinion, persistently ELISA-positive cats should be tested by the IFA test before they are condemned or declared persistently viremic.

The saliva ELISA test can be used as a screening test, but "positive" cats should be retested with another test, preferably the IFA test.

THE SICK CAT: Diagnostic/Treatment Protocol



Developed In conjunction with Fred Scott, D.V.M., Ph.D., Cornell Feline Health Center, College Of Veterinary Medicine, Cornell University, Ithaca, New York.

* Vaccination program may be initiated for previously unvaccinated sick cats once they return to health.

** IFA test may be performed at this stage to aid in confirming diagnosis. Additionally, a positive saliva or tears ELISA Test would indicate virus.

† Yamamoto, Janet K., et al. *Epidemiologic And Clinical Aspects Of Feline Immunodeficiency Virus Infection In Cats From The Continental United States And Canada And Possible Mode Of Transmission.* J. Am. Vet. Med. Assoc. 1989; 194: 213-220.

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Canine and feline immunization guidelines

This report, prepared by the AVMA Council on Biologic and Therapeutic Agents, summarizes information available on important diseases affecting dogs and cats, and includes recommendations for immunization to control these diseases. The revision was necessary to account for new knowledge and new products. The guidelines are for the usual situation of one or a few animals in a single environment. The veterinarian must use judgment and modify the guidelines for animals with special risks, for example, those in kennels, catteries, and shows. Some of these factors are discussed in the text. The Council recommends only those vaccines licensed by the USDA. Every effort was made to make the guidelines consistent with currently available products. The specific manufacturer's recommendations supplied with the vaccine should be consulted, particularly with new or revised products.

An animal's reaction to an invading organism is to mount an immunologic response. If the animal previously has been exposed to the organism, either naturally or by vaccination, it usually will be protected. The degree of protection will depend on a number of factors, but humoral antibody is one of the most important defense mechanisms. This antibody may be of several types, including IgM (rapidly responding antibody), IgA (local antibody), or IgG (long-acting antibody). The antibody will usually combine with the infectious agent to render it noninfectious. The degree of protection often depends on the amount of specific antibody present. Therefore, it is important to maintain a high antibody titer, and this may be accomplished by periodic exposure of the animal to the antigen. Thus, following completion of an initial vaccination regimen, revaccinations are important. Special consideration should be given to geriatric patients because of potential decrease in immunocompetence.

Animals are exposed to a number of infectious agents throughout their lives, but they are most susceptible to infection in early life. Thus, providing early protection through maternal antibody by immunization of the dam prior to breeding and vaccination of offspring at the proper time (when the maternal antibody has been depleted) is essential to a good immunization program. It is difficult to predict the exact time that maternal antibody will be depleted, so the initial immunization regimen usually requires a number of doses, beginning as early as possible. It is also difficult to predict the kind of infectious agents to which an animal will be exposed. For this reason, multivalent vaccines containing immunogens for most of the common infectious diseases have been developed. For licensure, these vaccines have to be effective and safe and, therefore, their wide-scale use is recommended.

Although the risk of adverse effects vs the benefit of using any product must be considered, in most instances the benefits far outweigh the risk. General contraindications for most vaccines are that they should not be used in animals that are obviously ill, pregnant, or undergoing a course of immunosuppressive treatment.

Feline diseases and recommendations for immunoprophylaxis (Table 2)

All cats should be immunized against feline panleukopenia, feline viral rhinotracheitis, and feline calicivirus infection. Cats should also be immunized against rabies, and vaccination of cats against chlamydiosis and leukemia should be done as required.

Feline panleukopenia—Feline panleukopenia, a highly contagious and devastating viral disease of cats, is caused by the feline parvovirus. By inducing cytolytic effects on actively mitotic cells, feline parvovirus causes severe leukopenia, enteritis, dehydration, and high mortality. Recovery from natural infection results in lifelong immunity.

Inactivated and MLV vaccines are available for parenteral vaccination, and certain MLV strains may be administered by the intranasal route. If administered after maternal antibodies have waned, these vaccines result in rapid and complete protection for at least 1 year and probably longer.

Feline viral rhinotracheitis—Feline viral rhinotracheitis, a highly contagious upper respiratory disease of cats, is caused by feline herpesvirus-1. Recovery from natural infection results in immunity against systemic disease, but not necessarily against local infection. Latent infection, with intermittent shedding of virus, frequently occurs.

Inactivated and MLV vaccines for parenteral administration as well as MLV vaccines for intranasal administration are available. Vaccines given parenterally in 2 doses at least 3 weeks apart, or as a single dose intranasally, should induce protection

Table 2—Feline vaccination recommendations

Disease	Type of vaccine	Route of administration	Age at first vaccination (wk)	Age at second vaccination (wk)	Revaccination intervals (mo)
Panleukopenia	MLV	SC or IM	8 to 10	12 to 16	12
	Inactivated	SC or IM	8 to 10	12 to 16	12
	MLV-IN	IN	8 to 10	12 to 16	12
Viral rhinotracheitis	MLV	SC or IM	8 to 10*	12 to 16	12
	Inactivated	SC or IM	8 to 10	12 to 16	12
	MLV-IN	IN	8 to 10	12 to 16	12
Calicivirus infection	MLV	SC or IM	8 to 10*	12 to 16	12
	Inactivated	SC or IM	8 to 10	12 to 16	12
	MLV-IN	IN	8 to 10	12 to 16	12
Pneumonitis (chlamydiosis)	Modified live	SC or IM	8 to 10	12 to 16	12
Rabies†	MLV	IM	12	64	12
	Inactivated	IM	12	64	12 or 36
Feline leukemia	Inactivated subunit	SC or IM	9	12 and 24	12
	Inactivated whole virus	IM	10	13 to 14	12

*For further details, *see* Rabies Compendium, *JAVMA*, Jan 15, 1989, pp 188-192. †May be performed earlier, but at risk of increased maternal antibody interference.

MLV = modified-live virus; IN = intranasal.

for at least 1 year. Annual revaccination is recommended. A rapid anamnestic immune response occurs in vaccinated cats after exposure to virulent feline herpesvirus-1. Intranasal vaccination may result in mild sneezing and ocular and nasal discharge 4 to 7 days after vaccination.

Feline calicivirus infection—Feline calicivirus infection is an acute respiratory and ulcerative disease caused by 1 of several strains of feline calicivirus. Recovery results in good immunity against most strains, but persistent infection may be associated with continuous shedding of small amounts of virus from the oropharynx.

Inactivated and MLV vaccines in combination with feline viral rhinotracheitis vaccine are available for parenteral vaccination, and combination MLV vaccines for intranasal administration are also available. The guidelines for feline viral rhinotracheitis vaccines apply to feline calicivirus vaccines.

Feline pneumonitis (feline chlamydiosis)—Feline pneumonitis (feline chlamydiosis) is an acute to chronic respiratory infection caused by a strain of *Chlamydia psittaci*. Attenuated virus vaccines of cell culture origin are available for parenteral vaccination, alone or in combination with other feline vaccines. A single vaccination affords adequate protection for at least 1 year.

Rabies—Feline rabies, an acute encephalomyelitis caused by a rhabdovirus, results primarily from exposure of cats to virus-infected wildlife such as skunks, raccoons, foxes, or bats. There is a high geographic correlation between skunk rabies and feline rabies. There are approximately 200 feline rabies cases per year in the United States and inasmuch as these cases pose significant risks to human beings, cats routinely should be vaccinated for rabies, especially in areas of enzootic wildlife rabies.

Inactivated virus vaccines of mouse brain and cell culture origin are available, and a high egg-passage cell culture-adapted MLV strain is also approved for use in cats. For MLV rabies vaccines, only approved strains should be used, because vaccine-induced rabies has resulted in some cats being vaccinated with other strains of virus.

Feline leukemia virus infection—Feline leukemia virus infection is a contagious oncornaviral disease of cats that may result in a non-neoplastic leukemia-related disease (such as anemia, immunosuppression, or panleukopenia-like disease, as well as neoplastic disease such as lymphosarcoma, fibrosarcoma, and leukemia). Whereas some cats develop an effective immune response, infection of other cats results in persistent viremia, with shedding of virus and development of neoplastic disease.

Inactivated subunit and whole-virus vaccines are available. Cats virus-negative for FeLV may be vaccinated when 9 to 10 weeks old, with repeat vaccinations 3 to 4 weeks later. A third dose may be required in 3 months. Annual revaccinations are indicated.

Feline infectious peritonitis—Feline infectious peritonitis (FIP) is a prevalent and usually fatal disease caused by a coronavirus. Subclinical infections with FIP virus or a closely related enteric coronavirus stimulate antibody, but do not necessarily provide protection against the clinical FIP. Coronaviral antibody may enhance clinical disease if there is reexposure to virulent FIP virus. There is a great need for an effective vaccine against FIP.

Feline immunodeficiency virus—Feline immunodeficiency virus (FIV), originally called feline T-lymphotropic lentivirus, is a prevalent infection of cats. After a subclinical, persistent infection of several years' duration, impaired immune functions may predispose the cat to various secondary infections, which may prove fatal. A vaccine for FIV is not available.