Special Reports

Use of serologic testing to assess immune status of companion animals

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Summary: At the November 1997 meeting of the AVMA Council on Biologic and Therapeutic Agents, the Council recommended that the *JAVMA* publish an article on the current status of the use of serologic testing in an effort to assist practitioners who must make decisions regarding vaccination of companion animals (ie, dogs, cats, and horses). It is anticipated that the peer-reviewed article provided here will be of benefit to veterinarians and will facilitate their attempts to maintain animal health through the knowledgeable use of vaccines.

In recent years, use of serologic assays, primarily ELISA, to assess immunity of vaccinated animals has increased. This has become a standard procedure for the poultry and swine industries, in which repeated testing, along with use of sophisticated data management software, has enabled producers to determine the degree of immunity in their animals. Analysis of results of these tests provides a rational basis for determining whether revaccination is required as well as an early warning of susceptibility to disease. This report examines whether similar procedures could be of benefit in decision making for vaccine use in dogs, cats, and horses.

Effective vaccines induce immunity sufficient to confer prolonged protection in a vaccinated animal. Because vaccine-induced immunity does not last indefinitely, it must be boosted at appropriate intervals to ensure that vaccinated animals do not inadvertently become susceptible to disease. This is especially important in companion animals with long life spans, such as horses and many breeds of dogs and cats. If revaccination is delayed, an animal's immune response may wane such that it may become susceptible to disease. On the other hand, too frequent revaccination may be inefficient and increase the risk of adverse effects. Therefore, revaccination ideally should be timed to ensure that immunity does not wane below protective levels while concomitantly ensuring that an animal is not subjected to unnecessary booster vaccinations.

Several reasons could be used to suggest that vaccination even with good vaccines cannot always be relied on. For example, immune responses are biological phenomena. As such, they have a normal distribution in the population. Some animals that receive an effective vaccine will mount poor immune responses and will not be protected. It is for this reason that pet owners must be advised that vaccines cannot be absolutely guaranteed to provide protection. Yet, although we know that animals vary greatly in response to vaccines, vaccinated animals are rarely tested to ensure that they have mounted a protective immune response.

Another reason for vaccination failure involves time of administration of vaccines. It is critical that young animals are vaccinated at a time when passive maternal immunity will not interfere with their immune responses. Carefully conducted studies have identified the earliest time when, in general, vaccines will be effective. Nevertheless, veterinarians rarely take into account variation among animals for duration of maternal immunity. Modified-live virus vaccines rely on limited replication of the agent within a vaccinated animal to trigger a protective immune response. These vaccines may contain remarkably little antigen. This small antigenic mass makes these vaccines highly susceptible to interference from antibodies. When an animal has maternal or endogenous antibodies at the time of vaccination, the vaccine agent may be neutralized before it can replicate. A high concentration of antibodies in an animal implies that the animal is probably protected but also could indicate that it may not be possible to stimulate an additional immune response in that animal.

Finally, it is reasonable to assume the efficacy of vaccines produced by various manufacturers will vary. It cannot be assumed that the duration or degree of immunity induced by one brand of a specific vaccine will be identical for all other brands of that vaccine.

Without a comprehensive effort to monitor vaccine efficacy and duration of immunity, annual revaccination has the advantage of simplicity and ensures that owners bring pets in for regular examinations. Although duration of vaccine-induced immunity may be variable, manufacturers and veterinarians have conformed to this practice. Indeed, until recently, only rabies vaccines were evaluated for duration of immunity. Other vaccines were simply tested by challenging vaccinated animals months or weeks after administration. The USDA has altered its rules to require that new vaccines must protect animals for the period claimed on the vaccine label. However, although the minimum duration of immunity must be determined, it is not required that manufacturers establish maximum duration of immunity. Unfortunately, few studies have been

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performed to determine duration of immunity for vaccines designed for animals. Because it has been accepted that immunity will persist for most animals for at least 1 year (provided the animal is not challenge exposed to the agent), this assumption has rarely been critically assessed. In fact, given the amount of information known about the immune system, it is likely that many vaccines provide immunity for periods much longer than 1 year. In support of this, investigators examined the persistence of antibodies against panleukopenia, herpesvirus, and calicivirus in vaccinated cats and documented that antibodies against those diseases persisted at protective concentrations for at least 3 years.[1] In another study,[2] researchers documented that antibodies against canine distemper can persist for 8 to 10 years in dogs that are not challenge exposed to the agent.

Reasons to Test

Because we cannot always assume that a vaccinated animal is protected or that revaccination is absolutely necessary, many veterinarians are seeking methods for objectively determining immunity and risk of infection in animals prior to revaccination. It would be desirable to have simple, rapid, inexpensive tests available for measuring an animal's immune status. These tests could be used to differentiate between animals that are susceptible and require revaccination and those that do not. They also could be used to compare efficacy of vaccines from various manufacturers.

It is commonplace to test sera obtained from vaccinated swine or poultry to ensure that they are protected against disease. Thus, serum samples are routinely and repeatedly obtained from flocks to test for antibodies against pathogens that cause disease, such as avian influenza, Newcastle disease, or infectious bursal disease. If antibody titers are satisfactory, revaccination is not necessary; however, when antibody titers are low, the flock is reimmunized until birds achieve a protective titer. This procedure also permits managers to use the experience of others in the poultry industry when selecting the most effective brand of vaccine.

In general, detection of an antibody titer in serum obtained from an animal indicates that the animal has encountered the agent and an immune response has developed. However, the precise interpretation of a positive serologic response depends on specific circumstances. For example, a high antibody titer to parvovirus in an unvaccinated puppy suggests that vaccination must be delayed until maternal immunity has waned. On the other hand, detecting a high antibody titer to parvovirus in a mature dog after vaccination implies that vaccination has been successful.

Vaccination carries certain risks but rarely causes serious adverse effects. Thus, benefits of vaccination usually greatly outweigh risks. Nevertheless, any procedures that may improve this risk-to-benefit ratio should be welcomed. The major benefit of routine serologic testing would be to identify susceptible animals or to identify animals that do not require initial or booster vaccinations. Ideally, vaccination should be used to protect only those mature animals that require revaccination and then only for diseases for which the benefits of vaccination are clearly apparent.

Limitations to Serologic Testing

A simple test to determine an animal's immune status is a desirable goal; however, such a procedure has a number of limitations. Antibodies are not always protective, and for some diseases, antibody titers cannot serve as indicators of protection. An animal becomes immune to infection through many mechanisms. Thus, B-cell responses and subsequent antibody production are primarily protective against organisms exposed to body fluids. These include extracellular organisms, such as *Leptospira* spp, and bacterial toxins that are dissolved in body fluids, such as tetanus toxin, that are exposed to the immune system during transmission between cells. Conversely, for diseases in which organisms invade the body via the gastrointestinal or respiratory tract, surface antibodies of the IgA class, rather than serum antibodies, play a critical role in defense. Thus, high concentrations of serum antibodies against *Bordetella pertussis* in a dog cannot be used as a guarantee of protection, because serum antibody concentrations are poorly correlated with immunity for this disease.

For viral diseases, neutralizing antibodies and complement may play a less important role than T-cell–mediated immunity. Most antibodies do not penetrate cells, and persistent intracellular pathogens, such as viruses or intracellular bacteria, are not exposed to antibodies. Theoretically, antibodies are ineffective against such organisms, and high concentrations of antibodies may not be protective. These pathogens may replicate within cells regardless of the concentration of antibodies in the extracellular environment. This is well documented for herpesvirus or coronavirus infections, in which the viral carrier state is associated with persistent detection of virus despite a high concentration of antibodies.[3] In general, the higher the serum antibody titers, the greater the protection; however, the correlation between the 2 is not always absolute. It cannot always be assumed that an animal with low serum antibody titers is unprotected or that an animal with high antibody titers is resistant to infection. It must be strongly emphasized that interpretation of serologic results must account for the limitations of each assay and that results of assays must be interpreted conservatively.

For practitioners, the division between antibody- and cell-mediated immune responses is less clear. Although it is true that antibodies are not the primary protective mechanism against viruses, high antibody concentrations in the extracellular environment will serve to inhibit the spread of viruses between cells and, thus, promote host resistance. In diseases such as distemper of dogs or rhinopneumonitis of horses, many protective cell- and antibody-mediated processes contribute to host defenses. Although antibodies may not be protective against all diseases, antibody titers often are the only practical indicator available. Cellular immunity cannot be rapidly or cheaply measured, except in certain restricted circumstances.

Serologic tests are subject to error and may yield misleading results. Some insensitive serologic assays

may yield false-negative results, which would cause an animal to be vaccinated unnecessarily. This is of much less consequence than nonspecific tests that yield false-positive results, falsely implying that an animal is protected. Errors of this nature would, of course, lead to failure to vaccinate a susceptible animal. Understanding this limitation is central to the use of serologic assays for determining disease resistance.

Serologic tests measure the current status of an animal and cannot predict future protective status with accuracy. This is especially important when assessing relative risk and amount of challenge exposure. The immune system has a finite protective ability. Whether it can protect an animal will depend, in part, on the amount of challenge exposure by an infectious agent. Thus, an antibody titer sufficient to protect an animal against a low level exposure may well be insufficient to protect against an unanticipated heavy challenge. Results of serologic tests must be interpreted conservatively to minimize the risk of leaving an animal unprotected. It must be remembered that an animal that has mounted an immune response after vaccination will possess memory cells. Although antibody titers may have declined to low or undetectable concentrations, those memory cells remain. Thus, during challenge exposure to infectious organisms, that animal will mount a more rapid and efficient response, which may be sufficient to prevent development of clinical disease.

Serologic Assays

Serologic assays can be used to diagnose disease or assess immunity. Although many serologic assays (eg, virus neutralization [VN], immunofluorescence, and hemagglutination inhibition [HI] tests) have been used to measure antibody concentrations against infectious agents, the only practical assays suitable for veterinary practitioners are ELISA. Thus, a simple assay, such as a clinic-based ELISA, may be used to detect antibodies against canine parvovirus (CPV) in sera of pregnant bitches and their offspring to study the response of puppies to vaccination. With an easily accessible procedure for CPV antibody determination, veterinarians should be able to gauge the response of puppies after vaccination. It is not surprising that ELISA are used routinely to determine the protective status of poultry flocks or pig herds.

The crucial part of these tests is to establish a baseline or cutoff point. Animals with values greater than the cutoff point are adequately protected, whereas values less than the cutoff value would indicate animals that are not protected and are in need of revaccination. The cutoff value can be established 2 ways. First, challenge-exposure studies can be used. Vaccines are administered at various doses, and antibody titers are determined before animals are challenge exposed to infectious agents. The antibody titer of animals protected after a minimal vaccine dose serves as the reference point for the protective cutoff value. These studies are often performed by vaccine and test kit manufacturers. The second method is to monitor disease incidence in a specified population in relation to antibody titers. This is usually accomplished during a prolonged period of monitoring. Using this approach, some poultry and swine farms have established their own cutoff values instead of only relying on values provided by vaccine and test kit manufacturers. Each veterinarian could use the same method, although mass testing of herds or flocks has economic advantages and benefits for maintaining consistency of testing that might not be available for separately tested animals.

Interpretation of antibody titers, especially those determined by use of nonstandardized assays, may be difficult. Laboratories that offer these serologic tests are often reluctant to provide an interpretation for practitioners. Conversely, practitioners may have little experience in relating serologic results to protective status. In addition, test procedures are not standardized and lack quality-control standards. As pointed out previously, errors in interpretation of serologic test results may cause inappropriate decisions to be made and place animals at unnecessary risk of disease. Ultraconservative interpretation of serologic results can be modified as experience is gained. Practitioners should consult with laboratory personnel about results of serologic tests. Combining analysis of serologic test results with knowledge about an animal and its environment will enable practitioners to make the best recommendations possible for vaccination of each animal. Conversely, feedback from practitioners can assist laboratories as they refine testing techniques and interpretation of test results.

Diseases of Dogs

Canine distemper—Dogs are protected against canine distemper virus (CDV) by multiple immune mechanisms, including antibody- and cell-mediated responses. Dogs develop high concentrations of antibody to CDV after successful vaccination or infection. These antibody concentrations can be measured by means of ELISA, indirect fluorescence, and VN tests.[4-6] These antibodies clearly play a role in resistance to canine distemper. In a study,[7] investigators measured antibody titers to CDV in dogs with known immunization status. For nonvaccinated dogs less than 12 months old, they found that about half had a titer of ≥ 1:8. For dogs vaccinated against CDV, more than three fourths had a titer of ≥ 1:16. Protection appeared to be associated with an animal developing an IgG titer of > 1:50 (as determined by immunofluorescent antibody testing) within 3 weeks after vaccination. Other investigators[8] found that CDV-infected dogs with lesions of the nervous system appeared to have an impaired antibody response to the virus. For dogs that completely recovered from infection, antibodies were observed early after infection and antibody concentrations correlated with lack of lesions. In another study,[9] antibody titers were measured in dogs exposed to virulent CDV. Dogs surviving infection developed antibody titers of 1:100 within 14 days after exposure, as determined by use of VN tests. Dogs that failed to develop this antibody titer became ill and died. Similarly, it was reported in another study[10] that high amounts of antiviral IgG correlated with recovery from disease and that an inability to sustain substantial antiviral antibody response was characteristic of dogs that developed fatal encephalitis. Krakowka et al[11] documented that the ability of dogs to mount a vigorous antibody response

distinguished resistant from susceptible animals. Similarly, other investigators[4] reported that three fourths of dogs arriving at their laboratory without CDV antibodies developed respiratory tract disease, and a fourth of all dogs that did not have CDV antibodies at the time of arrival died. Conversely, for dogs with antibodies to CDV, only a fourth developed respiratory tract disease, and less than 5% died. The fact that passive immunization can be used successfully to protect dogs against infection with CDV also confirms the importance of antibodies for resistance to this disease.[12-14] One group of investigators[15] reported that antibodies against CDV, determined by use of VN tests, were protective and that dogs with a titer of ≥ 1:30 were protected, whereas those with a titer of ≤ 1:20 tended to be susceptible. Other investigators[16] documented that specific antibodies were highly effective for neutralizing CDV as well as preventing intercellular spread of CDV. Cells infected with CDV are lysed by antibody and complement.[17,18]

The importance of antibodies for determining whether a dog is protected against CDV is also emphasized by the fact that inactivated CDV vaccines, which are usually somewhat ineffective for providing substantial protection, stimulate an extremely low and transient antibody response.[19] It appears that successful viral replication in a dog is necessary if that dog is to develop a substantial and protective antibody response.

Persistence of antibodies to CDV after vaccination without challenge exposure was examined in dogs imported to Iceland, an island where canine distemper is not found.[2] Investigators found that two thirds of dogs vaccinated 8 to 10 years previously had antibody titers of ≥ 1:45. In the group for which mean interval since last vaccination was 6.25 years, 22 of 30 (73%) dogs had titers of ≥ 1:16. Although some dogs had low antibody titers, it is possible that these may have been inadequately vaccinated during the initial vaccination.

Although antibody titers are important when assessing immunity to CDV, it must be pointed out that cell-mediated responses also play a role in immunity to this disease.[20-25] Although multiple mechanisms are involved in resistance to CDV, detecting high antibody titers would indicate that an animal will likely be resistant to CDV.[26] Measurement of antibodies against CDV for dogs would, therefore, be a useful prognostic tool.

Canine adenovirus-1—Maternal antibodies can protect puppies against canine adenovirus-1[27]; therefore, antibodies must be protective for dogs. Antibody measurement would be useful in determining immune status of dogs to canine adenovirus-1.

CPV—Serologic assays, including ELISA[28] and indirect fluorescent antibody (IFA), HI, and VN tests, have been developed for the diagnosis of CPV infection.[29] In general, the IFA test is best used for diagnostic purposes, whereas the other tests can be used for determining immunity. Disease attributable to CPV infection is associated with viral invasion and destruction of the gastrointestinal tract. Cell-mediated immune responses do not play a role in antiviral defenses within the intestinal lumen, where IgA is of critical importance. However, once CPV organisms have gained access to the body, T- and B-cell responses are important. In a study,[30] investigators found that CPV antibody titers of ≥ 1:80 were detected in 125 of 176 (71%) CPV-vaccinated dogs. Antibodies to CPV are transferred from immune bitches to puppies through the placenta and via colostrum.[31] Colostral transfer accounts for approximately 90% of the maternally derived CPV antibody concentration. After suckling, antibody titers of puppies are, in general, about half that of their dam's titer. Puppies with titers of ≥ 1:80 (determined by use of HI tests) were immune to oronasal challenge with virulent CPV organisms. Dogs with titers of ≤ 1:100 are not protected, whereas dogs with titers of 1:200 to 1:800 may be protected, and dogs with titers of ≥ 1:1,600 are protected. It is not uncommon for dogs successfully vaccinated against CPV to develop titers of > 1:25,000, as determined by use of VN tests. After parenteral vaccination against CPV, seronegative dogs developed substantial antibody titers as early as 2 days after vaccination, and maximal titers developed within a week.[32] Immunity was associated with persistence of antibody titers of > 1:80 (HI test). In contrast, puppies with titers of < 1:40 remained susceptible. Thus, measurement of serum antibody concentrations appears to be a useful tool for determining protective status and the need for vaccination against CPV.

Leptospirosis—The antibody response of dogs to leptospiral vaccines can be monitored by use of the microscopic agglutination or leptospiricidal activity tests. Protection (determined by use of the hamster passive-protection test) is directly correlated with serum antibody concentrations.[33] Given that leptospiral bacterins may be responsible for many of the adverse effects of vaccines administered to dogs, serologic testing of dogs for leptospiral antibodies could be of enormous benefit.

Borreliosis—Commercially available antibody test kits (ELISA) are currently available to test for borreliosis (Lyme disease). However, they are primarily used for disease diagnosis rather than assessing immune status. Clearly, dogs that lack antibodies to *Borrelia burgdorferi* will be susceptible to infection.

Respiratory tract infections—Similar to gastrointestinal tract diseases, locally produced surface IgA, rather than serum antibodies, is responsible for resistance to infection in the respiratory tract of dogs.[34] Because multiple etiologic agents are involved, including *B bronchiseptica*, canine parainfluenza, and canine adenovirus-2, serum antibody concentrations are not good predictors for resistance of dogs to respiratory tract infections.[35] This lack of association between serum antibody concentrations and resistance in the respiratory tract is reinforced by the observation that maternal antibodies do not interfere with development of resistance in the airways.

Rabies—Rabies vaccines licensed for use in animals have label indications for duration of protection of 1 or 3 years. Several years ago, the USDA permitted some vaccine companies to market rabies vaccines on which the efficacy was determined on the basis of antibody titers rather than on challenge-exposure studies. Unfortunately, it was found that serologic test results

were not predictive of resistance to challenge exposure, and those vaccines had to be withdrawn.[35]

Diseases of Cats

Antibody titers against common viral pathogens of cats can be determined by use of ELISA or VN tests. Measurement of serum antibody concentrations may be especially appropriate for cats that have had an adverse reaction to a vaccine. However, similar to dogs, the correlation between antibody titers and protection has not been completely analyzed. A low antibody titer may not always mean that a cat is susceptible to infection, and a high antibody titer does not guarantee resistance. Persistence of antibodies in cats after vaccination has been investigated by measuring antibody titers in a group of specific-pathogen-free cats that were vaccinated as kittens with 2 doses of an inactivated vaccine against feline panleukopenia virus (FPV), feline herpesvirus (FHV), and feline calicivirus (FCV).[1] The response to FPV was especially strong, with high titers developing within months after vaccination and persisting for at least 6 years. In contrast, antibody titers to FHV and FCV (determined by use of VN tests) never reached high concentrations and declined slowly during the subsequent 6 years. Titers against FHV persisted for at least 3 years, whereas titers against FCV persisted for at least 4 years. However, titers varied substantially among cats, with some cats becoming seronegative to FHV by 4 years and to FCV by 5 years.

FeLV—Kittens that had antibodies against FeLV and feline oncornavirus-associated cell membrane antigen (FOCMA) are protected against infection and oncogenesis by virulent FeLV.[36] All kittens with titers > 1:8 (determined by VN tests) were protected against a known FeLV challenge that was fatal to 95% of kittens that did not have detectable antibody titers.[3] Because of this, ELISA has been used as the basis for deciding whether to vaccinate against FeLV. Cats with antibodies against the FeLV gp70 protein or FOCMA are considered immune and do not need to be vaccinated.[37] Testing serum of cats prior to vaccination has proven to be a successful and feasible procedure and supports the concept that this could be successfully extended to other viral diseases.

Respiratory tract disease—Cats that are carriers of FHV may have relatively high concentrations of antibodies (titer of ≥ 1:96; VN test), whereas cats that are noncarriers may be seronegative or have decreasing titers.[3] Cats develop cell-mediated responses against FHV and FCV after vaccination.[38,39] Efficacy of vaccines against FHV, FCV, and FPV can be readily measured by serologic tests. Vaccinated cats develop substantial persistent antibody titers to all of these viruses.[40] Thus, serologic assays may work well to monitor these diseases, although their interpretation may be colored by the potential for development of a carrier state in cats with FHV infections.

FPV—Antibodies are protective against FPV and can be used to measure immune status. In general, cats with antibody titers against FPV of ≥ 1:8 (VN test) appear to be protected against clinical disease.

Feline immunodeficiency virus (FIV)—Cats infected with FIV will mount antibody responses and produce virus neutralizing antibodies. However, unlike the other diseases of cats described here, antibodies to FIV are not protective.[41] Specific-pathogen-free cats immunized with a synthetic peptide developed high antibody titers to the purified virus.[42] Immunized and control cats were challenged with FIV and then monitored for 12 months. Despite detection of antibodies, immunization with the specific peptide failed to prevent FIV infection. Therefore, results of antibody assays for FIV are not of value when assessing whether cats will be protected from infection and, thus, cannot be used to predict immune status.

Feline infectious peritonitis—Feline infectious peritonitis is a disease associated with development of immune-complex mediated hypersensitivity.[43] Attempts to correlate in vitro antibody activity (VN tests) with in vivo protection have revealed that antibodies produced during the course of this disease will not necessarily protect cats against infection and cannot be used to determine immune status. An ELISA is commercially available but is used primarily for diagnostic and prognostic purposes. Results of assays are only indicative of exposure to FCV and do not specifically detect antibody against FCV organisms responsible for disease.

Diseases of Horses

Horses should also be subjected to serologic testing to determine whether they are immune to specific infections or would benefit from vaccination. However, similar to dogs and cats, little effort has been made to correlate antibody titers with protection or the need for initial or booster vaccination.

Equine arteritis virus—After vaccination with an inactivated whole-virus vaccine, antibody concentrations detected by use of ELISA preceded development of a virus-neutralizing response.[44] After vaccination with an equine viral arteritis vaccine, horses develop titers as high as 1:5,120 (VN test). This antibody titer decreased rapidly, but revaccination 2 months after initial inoculation elicited a prompt antibody response, and titers persisted for at least 6 months. A titer of 1:43 (VN test) appeared to be the 50% protective dose, which was confirmed on the basis of clinical signs and viremia.[45]

Tetanus—Tetanus is an excellent example of a protective immune response mediated entirely by antibodies. Thus, serum antibody concentrations should provide a direct measure of protection. Tetanus antitoxin antibody concentrations were measured in ponies.[46] Results of that study revealed that protective concentrations (≥ 0.01 U/ml of serum) were maintained for at least 20 months after vaccination.

Equine influenza—Equine influenza vaccines have been regarded as having limited, short-lived efficacy. For example, Burrows et al[47] obtained serum samples from ponies vaccinated with 4 commercially available equine influenza vaccines. They found that little or no antibody was detected after the first inoculation (on the basis of results of HI tests); second and subsequent annual revac-

cinations produced peak titers between 7 and 14 days after inoculation.[47] Titers rapidly decreased between 14 and 28 days after vaccination and less rapidly thereafter. Serologic responses to 4 equine influenza vaccines were monitored in seronegative ponies, using HI and single-radial-hemolysis tests.[48] Four weeks after the initial vaccination, responses were barely detectable, using the HI test. After revaccination, responses to A/Miami/63 virus were low or undetectable, but responses to A/Prague/56 virus were higher (17/20 ponies with titers of ≥ 1:16). Using the sensitive single-radial-hemolysis test, investigators found a dose-related antibody response to both viruses. Titers after revaccination were two- to fivefold higher than after initial vaccination. They also detected a high rate of decrease in titers after vaccination, because titers in ponies with the highest antibody concentrations had declined to low or undetectable concentrations 14 weeks later. Ponies immunized with inactivated equine influenza vaccines document a clear association between antibody titers (measured by single-radial-hemolysis tests) and protection.[49,50] Therefore, given the inadequacies of current equine influenza vaccines, it appears that routine serologic testing may be of major benefit when determining the protective status of horses and the need for revaccination.

Equine herpesvirus—As pointed out previously during the discussion of FHV, antibody titers do not have a good correlation with immunity to herpesvirus infections. For example, cell-mediated and antibody responses of horses to equine herpesvirus (EHV)-1 were examined.[51] After vaccination and revaccination with a modified-live EHV-1 vaccine, horses had minimal increases in EHV-1 antibody titers (VN test). However, these same horses had a marked increase in the cell-mediated immune response, as measured by the lymphocyte transformation test. Samples from vaccinated foals and mares were tested to determine antibody concentrations and the degree of protection afforded.[52] Investigators found that titers against EHV-1 were 1:8 or less (VN test) in most foals, and all foals were not protected. The ability of EHV vaccines to stimulate cellular and antibody responses to EHV-1 and EHV-4 has been evaluated in healthy horses.[53] Comparison of results of lymphocyte blastogenesis tests indicated that horses given modified-live EHV-1 vaccines had substantial increases in responses to EHV-1 and EHV-4. Responses to EHV-1 and EHV-4 for horses given an inactivated-virus bivalent vaccine were less. Both vaccines induced major increases in antibody titers against EHV-1 and EHV-4 (VN test and ELISA). Serum concentrations of antibodies to EHV-1 and EHV-4 (ELISA) were significantly higher in horses that received a bivalent vaccine, compared with responses for horses that received a monovalent vaccine. Thus, vaccination with modified-live EHV-1 vaccines can stimulate cellular and antibody responses that cross-react with EHV-4.

Conclusions

Managers of poultry and swine operations have embraced widespread use of a panel of ELISA to determine whether their animals are susceptible to infection and require vaccination. They usually obtain serum samples from a statistically determined number of animals and extrapolate the results to the remainder of the flock or herd. These tests are relatively inexpensive and lend themselves to automation and computer-based analysis. They have the additional benefit of identifying infectious agents against which their animals have developed resistance. Thus, testing can save money for producers by preventing unnecessary vaccinations.

For companion animal practitioners, conditions are different, although the basic principal is the same. Thus, the unit of interest would be each companion animal rather than an entire flock or herd. Therefore, testing of each animal would be required, and decisions would have to be made on the basis of information and circumstances for each animal. Once reliable data are acquired, careful and conservative use of serologic assays for companion animals could provide veterinarians with the ability to identify animals that must be vaccinated or, conversely, animals that can be safely left unvaccinated. Routine serologic testing could also provide badly needed data on the efficacy of various vaccines, timing for administration of vaccines, passive immunity, and persistence of immunity for companion animals. Analysis of antibody titers may provide important information about the degree of protection animals have developed against numerous infectious diseases, such as those mentioned previously. However, they probably would be less useful for determining the degree of protection against diseases, such as rabies, respiratory tract infections, FIV and FCV infections, and EHV-1 and EHV-4 infections, although test results could be used to monitor responses to vaccination.

Given the success of routine serologic testing for determining protection in the swine and poultry industries, it is reasonable to assume the same techniques could be applied to companion animals. Better, more cost-effective vaccines and vaccination schedules could be established, unnecessary revaccination could be eliminated, and clients could be provided with a scientifically based rationale for use of vaccines. The end result would be an improvement in the overall health of animals.

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