## **TITERS**

Protecting Your Dog With This Simple Test Makes Good Sense



By Dr Don Hamilton DVM



fter three decades of veterinary practice, I believe vaccination to be a troublesome aspect of medicine. My own observations, along with insights from those who taught me, indicate that vaccination is fraught with misunderstanding. Furthermore, vaccination, in my experience, is likely responsible, at least in part, for the vast majority of chronic disease we see in human and non-human animals in the modern world. For this reason, I do not generally recommend vaccines.

However, there is also an inherent risk in not vaccinating. Vaccination does in fact prevent some acute illnesses like parvovirus and distemper virus in dogs and panleukopenia virus in cats. While puppies are at much greater risk than adult animals for contracting infectious diseases, adult animals are not without risk. I have seen unvaccinated adult cats, for example, who became infected with panleukopenia virus, and some of these died. I do not recall seeing canine parvovirus in an adult dog, but it can occur. I have seen canine distemper in an adult dog, although thankfully not a fatal case, so far, in an adult. Vaccination, therefore, has a certain benefit. It's the risk that's less well understood, but my experience is that this risk is high.

There are larger questions here, including Darwin's "survival of the fittest," which implies that diseases are in fact strengthening factors for species. If we view the species as that which must be kept alive rather than the individual, then we must not vaccinate. However, it is not so simple to ignore the individual, and in this case, many animal guardians choose to vaccinate. As I understand, though, vaccination protects against the acute illness by inducing a chronic one in its stead. The more vaccinations we give (or receive), generally the greater the impact of the chronic disease.

#### **Titer Testing**

Today, many animal caregivers recognize the basis of the danger of overvaccination, and many will choose not to vaccinate except as required by law, preferring the risk of exposure to that of vaccine induced disease. Others, however, choose not to risk infection, preferring to vaccinate as wisely as possible. For those guardians, we look for ways to minimize the damage while maximizing the protection. Titer testing is one way to do this.

A titer is a measurement of the amount of antibody in serum to a given antigen. An antigen is a substance that can induce an immune response, and usually refers to antibody production. Proteins and other substances on bacterial and viral outer surfaces, for example, induce antibody production following infection or vaccination. Antibodies are proteins that the body produces to help fight infections; these bind to foreign antigens, facilitating their destruction and removal from the body. Antibodies are also called immunoglobulins.

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Titer testing involves using a known antigen (such as canine distemper virus) and mixing this with serum to see if the serum contains antibodies that will bind to the antigen. This binding is called agglutination. If agglutination occurs, then the laboratory technician will dilute the se-



rum and repeat the test. This step is repeated until the serum is diluted to the point that agglutination no longer occurs. The last dilution at which agglutination happens is then said to be the titer against distemper, for example.

Historically, titer testing was done at two-fold dilutions, in the sequence 2,4,8, 16, and so on. The resultant titer would then be represented as a ratio, such as 1:16 or 1:32, meaning that agglutination still occurred when the serum was diluted to 1/16 or 1/32 of full strength. The higher the second number, the more dilute the serum could be and still agglutinate the antigen, therefore the more antibody was present in the serum and the greater the protection.

In practice, there is generally a recognized minimum titer that provides adequate protection, and anything over that is gravy, so to speak. Additionally, today the titers are tested more mathematically, so the older two-fold dilution is not always how titers are reported. For example, protective titers for canine distemper virus (CDV), canine parvovirus (CPV) are generally 1:5 or greater. This will depend upon the laboratory performing the test, however, so any result must be compared with that laboratory's standards. For any of these viral diseases, though, as long as a dog has a titer with the second number equal to or higher than that of the lab's protective titer, you can rest assured that he or she is protected.

#### **Long Term Protection**

Now, however, this brings a second question, and that is, does an animal have to maintain such a titer to remain protected? The short answer is no. However, in response to client requests (or due to their own knowledge), many progressive veterinarians now offer titer testing in lieu of routine vaccination. The idea is that, perhaps three years after the last vaccine, the veterinarian would take a blood sample and test

a dog for parvo or distemper. Should one of these titers fall below the protective level, then the veterinarian would recommend a booster vaccine.

While this is a great step forward from routine boosters, and especially from annual routine boosters, it is mostly unnecessary. Immunological science informs us that, once an animal has ever had a protective antibody level, as indicated by a protective titer, that animal is almost certainly protected for life, at least for the major diseases (CDV, CPV, and canine hepatitis virus). Thus, once you run a titer test that indicates a protective level, you need not concern yourself further with that virus. Canine hepatitis is thankfully rare today so most guardians don't test for this.

#### **Repeating Titers**

So now, what do you do with this information? For adult animals, whether a year old or older, if you've never run a titer, you could have your veterinarian do so. If you see protective levels, forget about booster vaccinations for the viruses I've mentioned in the paragraph above. These are the primary canine viruses for which titers have value and which are life threatening diseases, other than rabies. For most other organisms, either vaccination is of questionable value, the disease isn't as dangerous, or titers are not as useful.

For example, circulating antibody is not as important for protection against upper respiratory pathogens like kennel cough (bordetella/parainfluenza virus) or the uncommon canine coronavirus. Immune suppressive intracellular viruses and bacteria like the Lyme disease spirochete (Borrelia burgdorferi), the feline leukemia virus, the feline immunodeficiency virus, and feline infectious peritonitis virus depend more upon cell mediated immunity and immunosuppression, and vaccination doesn't work well, nor do titers



accurately indicate protection. (I mention these feline viruses here as classic examples of viruses which require immune suppression to infect their hosts.)

The initial studies for the first Lyme disease vaccine, for example, required immunosuppressive doses of steroids to even induce short term infection in the dogs used in the investigation. Without the steroids, researchers could not successfully infect animals with the Lyme spirochetes. This, to me, strongly suggests that there is much more to the Lyme disease picture than a simple organism/host and organism/vaccine relationship. I don't recommend Lyme vaccination, and titer testing is also not too valuable.

#### Leptospirosis

Leptospirosis, while uncommon in most areas, can cause a severe infection occasionally. This is a complex situation due to the fact that we aren't dealing with only one antigenic organism. The Leptospira bacteria are spirochetes, and their outer protein and fat coating provide the antigens. However, these antigens are different among different strains of the bacteria. These strains are designated as serovars, which belong to various serogroups. Each serovar and serogroup is antigenically distinct, and as a consequence, vaccination and titer testing requires knowing which serovar/serogroup in your region is problematic, as there is little cross-protection between them.

For example, vaccination against the canicola serogroup will provide little protection against the icterohemorrhagiae serogroup. There are five main serovars that affect dogs: canicola, icterohemorrhagiae, grippotyphosa, pomona, and bratislava. For the first four, the serogroup to which they belong has the same name; bratislava belongs to the serogroup Australis.

Leptospira can be transmitted directly from one animal to another, or from the environment. For the latter situation, the organisms require moist conditions and moderate temperatures. As a consequence, leptospirosis primarily occurs in warm, moist regions and in rainy seasons. Outdoor dogs living in such climates are obviously the most at risk.





Titer testing isn't as widely available as for distemper, parvovirus, and rabies, and only IgG ELISA testing adequately evaluates vaccination. The bottom line is that immunization and protection requires using a vaccine that contains the serovars known to exist in your region. Titer testing, if available, will show immunity if IgG antibodies are present, and must also correlate to the serovars in the vaccine and in your region.

Titer longevity and immunity is not generally as good with bacterial as with viral vaccines, thus if you need to vaccinate for leptospirosis, you may need an occasional booster vaccine, especially in young dogs. Older dogs show less susceptibility to leptospirosis, probably due to exposure at some time. Essentially, if you have outdoor dogs with access to ponds or other slow to non-moving water sources, in warmer areas of the country, and leptospirosis is endemic in your region (you can find this through your health department), you may wish to consider immunization. The disease is uncommon, but can be severe. Titer testing will probably become more available and useful over time.

#### Rabies

Rabies vaccination and titer testing has its own complications, particularly because rabies affects humans and, as a consequence, mandatory vaccination laws represent our attempt to prevent rabies in humans rather than being primarily about protecting animals. This leads to more stringent requirements than are medically necessary.

Titer testing is essentially as accurate as for distemper and parvovirus in establishing protection, which is life long in most animals. Unfortunately, at this time titers are not accepted in lieu of booster vaccination, but hopefully this will change before too long. Furthermore, there have been one or two cases wherein vaccinated animals who have been experimentally challenged with rabies virus developed rabies despite sero-conversion. While this is an anomalous situation, its existence leads those who favor stringent vaccination requirements to strongly oppose

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using titers as a means of assessing protection. As a consequence, most states and provinces require rabies vaccinations in adult animals, at one to three year intervals, even though this is not necessary. However, there is no more certainty of protection with routine boosters than with titer testing, so these requirements are not based upon scientific studies. Basically, they're based upon guesswork.

Essentially, with rabies vaccine, we face a conflict between medical requirements and legal requirements. For legal requirements, you must check with your health department or your local veterinarian. Most commonly, vaccination is required at three or four months of age, one year later, and then every one to three years, with three years (triennial) being most common. Large studies have shown that triennial vaccination is as effective as annual vaccination, so requirements for more frequent vaccination are pointless.

Medically, as with distemper, parvovirus, and adenovirus, rabies vaccination generally provides long duration of immunity, and probably lifelong immunity, once adequate seroconversion occurs. Essentially, once a dog seroconverts, he is likely protected for life. In the United States, titer testing for rabies is widely available, either through commercial veterinary lab-



oratories such as IDEXX or Antech, or directly through Auburn University or Kansas State University (KSU) laboratories. The commercial labs send the serum to the university laboratories for rabies testing, so you can save some money if your veterinarian sends the sample directly to the university labs. In Canada, samples can be sent via IDEXX or University of Guelph; both send the samples to KSU. I understand it is less expensive through Guelph.

The gold standard test for rabies antibodies is the rapid fluorescent focus inhibition test (RF-FIT), sometimes also called the fluorescent antibody virus neutralization test (FAVN); more recently an ELISA (Enzyme-linked immunosorbent assay) test has been developed, which correlates favorably with the RFFIT. In either assay, the cutoff for protection has been set at 0.5 IU/ml. This is much greater than is probably need-



ed, as 0.1 to 0.2 IU/ml have been shown to be 100% protective in some studies.

As rabies vaccines are inactivated (killed) virus vaccines, a booster is often needed for the maximum protection. Medically, the best practice would be to give two vaccines, two to four weeks apart for the initial protection, even though this is not what the law or most veterinary organizations recommend. In either case, if you run a titer test a month following the vaccine and your dog has a protective titer, she is likely protected for life.

If you are in an area where rabies is endemic, you may wish to consider the two-vaccine initial series, followed by a titer test. With a positive titer, you're highly likely to have lifetime protection. Sometimes the protection following one vaccine is not as durable, but as rabies is relatively uncommon, for most dogs this is probably medically (not legally) adequate. I realize these recommendations are not in line with the law, and so you must be mindful of legal requirements, but I suggest you work in your community and state/province to change the requirements to reflect modern immunological science.

#### **Core Vaccines**

Finally, what about the other vaccines in puppies? Generally, for those who choose to vaccinate, I recommend one distemper virus vaccine at 12 weeks and one parvovirus vaccine at 14 weeks, given as single virus vaccines, if available. If the pup's mother was heavily vaccinated, you may need to delay these vaccines another week or two.

Two weeks after the parvovirus vaccine, test for parvovirus and distemper titers. If these are strongly positive, you should have no fur-



ther concerns. If they are not strongly positive, you can retest a month later, and if they persist at positive levels, the dog is probably protected for life. A concern here is that the maternal antibodies (MA), which pass from the colostrum (and a small amount through the umbilical cord) into the puppy, will test positive with titer testing, though these should be at pretty low levels by 12-16 weeks of age. Thus, testing prior to twelve to sixteen weeks may show a positive titer that reflects MA rather than that induced by vaccination. Therefore, if the second titer is less than the first one, this may reflect only MA, and you should repeat the vaccine one more time and retest in another month. While this is a bit costly, you can minimize vaccination and maximize protection, as well as your certainty of protection, with this method.

You must use caution regarding potential exposure to other dogs until you have established protection. There are new in-clinic tests for the main dog viruses (CDV, CPV, and CAV) now, called TiterCHEK and VacciCheck, so titer testing will be simplified when clinics obtain these kits, as results are available within about 30 minutes. Hopefully a kit for rabies will also be available soon. These kits will greatly reduce vaccination exposure in the future. TiterCHEK currently does not test for CAV.

Adenovirus-2 (CAV) vaccine, while recommended as a core vaccine by many organizations, is not as necessary for most dogs, in my opinion. This vaccine protects against the canine hepatitis virus (Adenovirus-1) and the upper respiratory virus, which is less severe than the hepatitis virus. While the canine hepatitis virus is extremely severe, it's not so common these days, and for most dogs, may be unnecessary. It's included in any of the combination products, however, and if you cannot use separate parvovirus and distemper virus vaccines, you may use one combination vaccine at 12 weeks, test a month later, and repeat parvovirus vaccine if necessary. In this case, you'll probably have

successfully immunized your dog against adenovirus-2. The more vaccines and the more antigens your dog receives, however, the greater the likelihood of complications like autoimmune disease, so you do want to minimize these as much as possible.

Dr Don Hamilton has been practicing veterinary medicine since 1979 and practicing homeopathy since 1988. His practice is now exclusively homeopathy. He is the author of Homeopathic Care for Cats and Dogs: Small Doses for Small Animals. Dr. Hamilton has concerns about our interactions with companion animals and is interested in how these interactions affect animals, as well is in what these interactions suggest about modern culture and our own needs as humans. Dr. Hamilton believes that all animals and plants have an inherent right to exist that is independent of their usefulness to humans.





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