SOAR Project Proposal, Spring 2021

Canine Parvovirus-2 Prevention Study

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Abstract

Canine Parvovirus-2 is a highly contagious viral gastrointestinal disease that has a fatality rate of greater than 90%. Past research of canine parvovirus-2 (CPV-2) shows that the implementation of at least one prevention measure can be successful in preventing parvovirus infection. However, very little research has focused on approaching prevention measures from a holistic perspective where multiple variables are employed. Therefore, the present study will examine what combination of best practices is the most successful at preventing canine parvovirus infection in puppies ages 4-12 weeks. This study focuses on three breeds of livestock guardian dogs: Kangal, Boz, and Anatolian Shepherd that currently reside at Blackleaf Livestock Company in Montana. The study will take place in the summer of 2021, where approximately 6 batches of puppies will be evaluated using a combination of different preventative protocols.

Keywords: canine parvovirus, prevention, puppies

Process and Methods

Background

Canine Parvovirus-2 is a highly contagious viral gastrointestinal disease that has a fatality rate of greater than 90% when prevention measures are not in place or properly utilized (Horecka et al., 2020). Vaccination and other prevention protocols are important because once a puppy is infected there is no treatment for the disease leading to a high mortality rate. Therefore, preventing parvovirus infection is imperative. In addition, vaccinations are cheap and effective when a vaccination schedule is followed. However, locations with a higher viral load in the soil due to a large dog population create a hardier environment that predisposes puppies to a higher and more acute rate of infection (Nandi & Kumar, 2010). The purpose of this prevention study is

to find what combination of prevention protocols prove to be the most effective with the puppies at the Blackleaf Guardian Ranch in Montana.

Studies have shown that proper vaccination is a critical component of preventing canine parvovirus infection, starting as soon as 4 weeks of age, and revaccinating every 2-3 weeks until 20 weeks old (Decaro et al., 2020; Mylonakis et al., 2016). While working with a veterinarian on a vaccination schedule, it is encouraged to track the antibody levels in the blood to determine the optimal time for re-vaccination, contributing to an even lower rate of infection (Decaro et al., 2020). Although there are a few different vaccination types, modified live virus vaccines are the most effective. They take less time to create an immune response and are recommended for routine use (Day et al., 2016). While vaccinating is extremely important in creating an immune response to the virus, the environment that the puppy is whelped and raised in has a significant impact on their chance of contracting the virus.

Locations with large dog populations, like shelters and breeding kennels, where an outbreak has occurred in the past acts as the perfect environment to foster canine parvovirus-2 by providing hosts for continued infection through shedding the virus in feces (Cavalli et al., 2018). Research by Nandi & Kumar (2010) found that a large amount of virus is shed in dog stool during the 2 weeks post-exposure. The average infectious dose for an unvaccinated dog is 1,000 viral particles while an infected dog sheds 35 million viral particles, or 35,000 times the typical infectious dose per ounce of stool. Because of the viral load in a fecal sample, there is an important need for isolation to prevent the contraction of CPV-2. Therefore, isolating puppies while following the vaccination sequence is a very important step in correct prevention. While isolated, one should also disinfect all surfaces regularly.

Utilizing a 0.75% sodium hypochlorite solution (bleach) on all equipment and exposed surfaces significantly reduces the spread of the canine parvo virus when used in combination with handwashing and proper use of gloves, gowns, hair caps, and booties (Mazzaferro, 2020; Mylonakis et al., 2016). Because the virus is not enveloped, it can overcome winter temperatures, last 7 months in shaded areas, and 5 months in areas with sun exposure. However, there is no way to completely disinfect contaminated dirt and grass. In indoor cases, the virus loses infectivity within one month. When assuming that no reintroduction of the virus takes place, disinfection has proven to be effective indoors while proving somewhat effective outside (Nandi & Kumar, 2010).

Additional research that would be beneficial to this study is to evaluate the susceptibility of each breed of dog residing at the ranch: Kangal, Boz, Anatolian Shepherd, Great Pyrenees, Komondor, and Akbash. This is because previous research has provided a relationship between certain breeds and their susceptibility to parvovirus (Behera et al., 2015). Furthermore, one could test if mix breeds are less susceptible due to the combination of different genetics. Studies have also shown that the incidence of infection and death was higher among males than females (Horecka et al., 2020; Behera et al., 2015).

Following the evidence stated in the introduction above, this research will be guided by the following research question: By vaccinating and following best care practices for canine parvovirus-2, there will be a reduction in illness and elimination of death among highly susceptible puppies ages 4-12 weeks old when compared to a control group without preventative measures.

Two relevant hypotheses that can be derived from this question are as follows. First, through administering a booster vaccination to bitches before conception, puppies will be less

likely to develop an infection. Second, by providing a cement floored environment isolated from other animals that is routinely cleaned with a bleach solution, we will see a subsequent decrease in infection.

Participants

Approximately 6 batches of puppies will be utilized for this study (n=40). The puppies of three breeds of livestock guardian dogs: Kangal, Boz, and Anatolian Shepherd will be used in this study. These dogs all reside at the Blackleaf Livestock Company ranch in Montana, where they are raised and trained to protect livestock from predators such as grizzly bears, wolves, and coyotes.

Materials

The materials needed for this research are: CPV-2 live vaccine for following a strict vaccination schedule, parvovirus test kits for verifying infection in puppies, bleach solution for disinfection whelping pens, chemical spray bottles, hoses, overalls, boots, and gloves.

Procedure

The project will last over the course of the summer of 2021, where puppies will be followed for 12 weeks. We are actively trying to determine if this research is exempt from Institution Animal Care and Use Committee (IUCAC) approval from the United States Department of Agriculture Licensing and Registration Under the Animal Welfare Act.

All puppies will be vaccinated every two weeks, starting at 4 weeks until 12 weeks of age. The vaccination that will be utilized is a modified live virus that is given subcutaneously between the scapula on the back by trained personnel. This experiment will not include non-vaccinated control because not vaccinating results in a death rate as high as 100%. Therefore all puppies will be vaccinated and only additional protocols will be evaluated during this research.

To verify that prevention protocols were successful, each puppy will be tested with a parvo virus test kit.

The table below shows the experimental groups of this study. “Vaccination Only” indicates that the puppies will only be vaccinated without any other prevention protocols but in all other groups this will be a control. “Vaccination Booster” indicates that the bitches will be vaccinated prior to conceiving, “Isolation and Disinfection” indicates that the puppy is vaccinated, kept in isolation, and the area disinfected once daily. Finally, “Isolation, Disinfection, & Booster” group indicates that all protocols will be employed. A 0.75% sodium hypochlorite solution will be applied to all equipment and surfaces in contact with the puppies every 24 hours while the vaccination protocol is being completed (approximately 8 weeks).

Table 1: Assignment of Study Groups

Puppies:	Vaccination Only:	Vaccination Booster:	Isolation & Disinfection:	Isolation, Disinfection, & Booster:
Anatolian Shepherd	✓	✓		
Boz			✓	✓
Kangal	✓		✓	

Based on previous experience, the Boz breed is more susceptible to parvovirus infection, therefore the Boz batches will be in the combined protocol group. The Anatolian Shepherd has shown a higher resistance to infection, so those batches will be in the booster only prevention groups. The Kangal breed is moderately affected and is assigned to the isolation and disinfection group.

To document the success of each prevention group, signs and symptoms of CPV-2 will be closely monitored and recorded. The most common characteristics of this disease are lethargy,

loss of appetite, abdominal pain and bloating, fever or hypothermia, vomiting, and severe diarrhea. A parvo test will be obtained for suspected cases, and a death count will be kept. This procedure was discussed with Dr. Jennifer Fischer, DMV of Valley City Veterinary Clinic. She agreed with the prevention protocols and encouraged the vaccination of bitches before breeding.

Timeline

The following timeline illustrates the intended schedule for research at the Blackleaf Livestock Guardian ranch by Bynum, Montana. The following schedule is based off the birth of each batch of puppies as time point zero.

Table 2: Timeline

Weeks by batch age:	Goal:
Spring Break	Booster shots on adult Boz and Anatolian Shepherd breeds
1-3	Establish mentorship with ranch vet, order live virus vaccine, gather supplies
4-12	Intervention period when procedure for each batch is strictly followed
13	Analysis of data

Budget

The table below lists the anticipated expenses of this research project. The Blackleaf Livestock Company will continue to pay for vaccine. The majority of supplies will already be available at the ranch.

Table 3: Budget

ID	Category	Source	Total
1	Supplies: Parvo virus test kit ($7/test x 40 tests = $280), bleach ($10)	SOAR	$290
2	Parvovirus live vaccine	Blackleaf Livestock Company	$600
3	Payment to Primary Researcher	SOAR	$1,000
4	Total	SOAR	$1,890

Dissemination

This research will be presented on at the Valley City State University Scholarship Symposium and Science Seminars Series, a poster will be created and posted in Rhoades Science Center, presentations at veterinary offices and the North Dakota Veterinary Technician Association annual meeting, and any other opportunities that may present themselves.

Educational Objectives

I am intending to pursue a Doctor of Veterinary Medicine. I want to be comfortable evidence-based information so that I can share my experience with future clients, specifically with regards for this virus because of its preventable nature and subsequent large fatality rate without prevention. I have a moral and ethical obligation to find a solution for the puppies at this ranch where I have worked the past two summers and have had firsthand experience with sick and dying puppies. By doing this research project, I will become more comfortable with

constructing and carrying out research in the future. Finally, this will provide a way of showing my commitment to the advancement of veterinary medicine.

Integrative Nature of work

The integration of this research will be putting into practice the information and skills I have learned in the classroom into a real-life situation. While at the ranch last summer I recognized an issue with severe parvovirus infections in puppies. Immediately, my scientific thought process that was learned in microbiology, research methods, statistics, chemistry, and anatomy and physiology classrooms started to kick in. What could I do to prevent infection? Researching this topic became easy as I was able to combine my researching skills with my interest in finding an answer to help the problem. In concert with over 250 hours of vet clinic work, two summers of ranch work, and three years of undergraduate studies at VCSU my research plans started to take shape. Now I am eagerly waiting to combine my theory with my practice. If this research goes as planned and a solution is discovered, I will seek to publish this research and share it with veterinarians to help prevent parvovirus outbreaks in the future.

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