Serological Response of Pups to the Selected Canine Vaccines and Vaccination Schedules against Canine Parvovirus

Doo Kim¹, Seok-young Jeoung, So-jeo Ahn, Jong-ho Jung and Son-il Park

Department of Veterinary Medicine, Kangwon National University, Chunchon 200-701, Korea

Abstract: This study was undertaken to provide the appropriate vaccination protocol of canine parvovirus (CPV) vaccine for the companion dogs in Korea. A total of 120 healthy pups (20 pups per group) at 6 weeks of age were randomly assigned to one of four commercially available vaccines [C, G, K, and V groups] and one of vaccination schedules [V2 and V4 groups]. The serological responses to the CPV component of the vaccines were determined by measuring HI titers. The maternal antibodies were declined to under the protective level at 6 weeks of age. Therefore, it was considered that vaccination of pups for CPV should be started at 6 weeks of age. And when the combination vaccine was used, the immunogenicity of V vaccine was superior to the other vaccines and optimum vaccination schedule was 3 times vaccination with 3 weeks-interval starting vaccination at 6 weeks of age. Although pups were vaccinated at 6 weeks of age, the geometric mean CDV titers of pups in all groups by 9 weeks of age were under the protective level. So, hygienic measures including avoiding to exposure to the high risk areas were needed to prevent CPV infection in this period.

Key words: canine parvovirus, vaccine, vaccination schedule, HI titer

Introduction

Canine parvovirus (CPV) infection in dogs has been associated with outbreaks of acute hemorrhagic enteritis characterized by bloody diarrhea, vomiting, depression, leukopenia, and dehydration. The causative organism, CPV type 2 (CPV-2), was first identified in 1979. Since then, two variants of the virus have been identified: CPV type 2a (CPV-2a) in 1980 and CPV type 2b (CPV-2b) in the mid 1980s. Currently, more than 80% of all cases of CPV infection in the United States are a result of infection with CPV-2b.

At the present time, nearly all adult dogs are immune because of vaccination or natural exposure. Although vaccination has reduced the number of clinical cases, parvoviral enteritis is still an important disease, especially in pups. The continuance of the disease might be the result of the mutation of the virus; but it is not likely, because vaccines made from CPV-2 produce an antibody response that protects dogs challenge with CPV-2a and CPV-2b. And pups are at risk for infection because maternal antibodies interfere with response to vaccines but do not protect the pup from natural disease.

Even with adequate treatment, the mortality for dogs with CPV infection is high. For this reason, extensive efforts have been directed at preventing the disease through vaccination. The original vaccines incorporated feline panleukopenia virus or mink enteritis virus, and viruses that are antigenically similar to CPV-2 were of limited efficacy. Killed virus and modified-live CPV vaccines were subsequently developed; however, efficacy varied widely. In addition, it was found that maternally derived antibody titers that were too low to provide protection from naturally acquired infection were high enough to prevent immunization. Thus, dogs were susceptible to infection for as long as 10 weeks, while passive (i.e., maternally derived) immunity waned and before active immunity could be induced.

In some instances, pups could not be actively immunized with conventional, commercial vaccines until at least 18 weeks of age. Thus, veterinarians recommend administration of multiple CPV vaccine doses. However, a pup would develop clinical CPV enteritis if exposure occurs during the window of susceptibility. The window of susceptibility exists because modified live CPV is less immunogenic than naturally occurring strains. This is a result of the viral attenuation process, which reduces CPV host infectivity. The more passages CPV undergoes in tissue culture, the less infective it is in vivo. To more effectively overcome interfering levels of maternally derived antibodies, a new vaccine was developed containing a low-passage CPV strain with inherently greater host infectivity.

To further enhance its immunogenicity, the low-passage CPV strain was produced in a high-titer dosage (i.e. with an increased amount of CPV). High-titer canine parvovirus vaccines are intended for use in the one segment of the canine population that continues to be vulnerable to clinical CPV enteritis - pups four to 18 weeks of age.

Despite high-titer vaccines were available, manufactures of these commercial vaccines recommended that pups be vaccinated until they are 16 weeks of age or older. However, recently after high-titer vaccines are available commonly, vaccination schedules have been changed so that most pups receive their last dose of CPV vaccine at 12 weeks of age.

Recently in Korea, CPV infection in the companion dogs has been controlled after use of high-titer CPV live vaccines.

¹Corresponding author.
E-mail: kimdoo@kangwon.ac.kr
However, CPV infection is quite common in kennels and breeding farms. And difference in efficacy of vaccines is anticipated because of use of various vaccines and vaccination protocols. Thus, this study was undertaken to compare the efficacy of four commercially available combination vaccines and three different vaccination schedules in inducing an active immune response in pups against CPV.

Materials and Methods

Experimental animals
1) Efficacy of vaccines
To compare the efficacy of four commercially available combination vaccines and three vaccination schedules in inducing an active immune response in the companion dogs against CPV, a total of 120 healthy pups at 6 weeks of age were included in this study. These pups were presented for vaccination by owner at 9 local animal clinics between March, 2002 and October, 2002. After owner’s were in compliance with participation in this study, pups at six weeks of age (20 pups per group) were randomly assigned to one of six groups shown in Table 1. Each pup was reared in owner’s house and managed according to the ownership. The pups received preventive and therapeutic medical care as deemed necessary by the veterinarians at local clinics. This included anthelmintics and rabies vaccination. The eleven pups failed to complete the study and were excluded during the trial.

2) Change of maternal antibodies
Seven healthy mixed breed pups were used to observe the declining pattern of maternal antibodies. These pups were born from a bitch who was vaccinated 2 times with the commercial combination vaccine containing CPV at 6 month interval before pregnant, and were reared for 7 weeks with the dam. Pups were weaned at 7 weeks of age and managed in individual cage and provided with commercial dog food and fresh water ad libitum during the experimental period of 17 weeks of age. Blood were collected every week for entire period. In this study, antibody titers of 1:80 or greater using hemagglutination inhibition (HI) were considered to be protective.

Vaccines
The two commercial combination vaccines (G and K) were manufactured in Korea and two vaccines (C and V) were imported from USA. Each vaccine was contained modified-live CPV, canine distemper virus, canine adenovirus type 2, and canine parainfluenza virus in a lyophilized form and Leptospira canicola-icterohaemorrhagiae bacterin in a liquid form that was used as the vaccine diluent. All vaccines were purchased by the investigators. At six weeks of age, each pup in each group was vaccinated with one of the vaccines according to Table 1. Revaccination was administered at 8, 9, 10, 12, and 14 weeks of age according to Table 1. Vaccines were administered subcutaneously in the dorsal aspect of the neck or thorax. Postvaccinal adverse effects were not observed in all vaccine groups.

Hemagglutination inhibition (HI) test
The HI test was based on the method described by Carmichael et al. Briefly, after heat inactivation at 56°C for 30 minutes, the sera were treated with pig erythrocytes to remove non-specific inhibitors of viral hemagglutination. Serial two-fold dilutions of the treated sera were prepared in U-bottomed microtiter plates and then an equal volume (50 µl) of virus suspension, containing 8 hemagglutination units of CPV, was added to each well. When the serum-virus mixtures had been incubated for 60 minutes at room temperature, 50 µl of 0.5 percent pig erythrocytes was added to each well and the plates were kept at 4°C for four hours.

The antibody titer was considered to be the reciprocal of the highest dilution that completely inhibited hemagglutination. In each test, known negative and positive sera with moderate and high titers were included. Results were only accepted when the titers of the standard sera fell within specific limits (a two-fold change for the positive sera). Seroconversion was considered to be a four-fold or greater change in titer or an increase from a negative value (< 8) to a positive value (8 or more).

Statistical analysis
Prior to statistical analysis, all titers were converted to natural logarithms and geometric mean CPV HI titers were determined for each sample period. And the week that each pup seroconverted, the overall percentage of pups in each group that had seroconverted at each sample period, and the mean and standard deviation of the week of seroconversion for each group were calculated.

A repeated-measures analysis of variance (ANOVA) was used to compare between-group titers or differences in regard to number of pups that had seroconverted at the time of each vaccination, using Tukey’s multiple comparison test. A values of p less than 0.05 were considered significant. All analyses were performed with computer software package SAS (version 8.1 for Windows).

Table 1. Numbers of pups, number of vaccinations, and interval of vaccination in each vaccine group

<table>
<thead>
<tr>
<th>Groups</th>
<th>Number of pups</th>
<th>Number of vaccination</th>
<th>Interval of administration (Weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td>C</td>
<td>20(18)*</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>G</td>
<td>20(18)</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>K</td>
<td>20(17)</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>V (or V3)</td>
<td>20(20)</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Interval</td>
<td>V 2</td>
<td>20(19)</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>V 4</td>
<td>20(17)</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td>6 groups</td>
<td>120(109)</td>
<td></td>
</tr>
</tbody>
</table>

*The number in parenthesis is number of pups which were provided all data and finally used for statistical analysis.