Volume 5, issue 8, 2016 • e-ISSN: 1857-8187 • p-ISSN: 1857-8179

Research Article

Distribution of Antibody Titers to CPV-2 Among Dogs of Various Age-Groups and Locations in Kosovo

Healthcare

Keywords: dogs, syndrome, dog breeds, Illyrian shepherd, English setters, German shepherd, Belgian shepherd and Pekingese, HI titers, CPV-2, age-groups, etc.

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Abstract

Blood samples were taken from 124 unvaccinated, clinically healthy dogs of different ages (from 6 to 156 months). Serum samples were tested by using hemagglutination inhibition (HI) test and the tests were performed at the Institute for Food Safety and Veterinary (ISUV), Tirana, Albania. Titers of HI antibodies to CPV-2 varied from 0 to 1:2048. Out of 124 dogs tested, 96 (77.42%) had HI titers of 1:16 or greater (a criteria that establishedthe positivity) and 28 of them had HI titers less than 1:16 that meant a negative result. The seroprevalence in communities of Kamenica, Rahovec, Peja, Prishtina and Prizeren was 78.0%, 76.5%, 64.2%, 75.0% and 100%, respectively. It showed a high prevalence of CPV-2 antibodies in each community involved in the study as well as that at the nation level 96 (77.42%). Such a finding indicates that the prevalence of anti-CPV-2 antibodies is high, and as a consequence, the CPV-2 itself is widespread all-over the country. Dogs that were studied were divided in five different groups of ages; 1) 6-12 months old, 30 of the total from whom 19 (63.34%) were positive and 11 (36.7%) were negative; 2) > 12-24 months old, 34 of the total, from whom 26(76.47%) were positive and 8 (23.53%) were negative; 3) >24-48 months old, 44 of the total, from whom 36 (81.8%) were positive and 8 (18.2%) were negative; 4) >48-96 months old, 13 of them in total, from whom 12 (92.3%) were positive and 1 (7.7%) was negative and finally 5) >96 months old, 3 of the total, 3 (100%) were positive.The older the age-group, the higher rate of dogs having HI antibody titers was the result.

Introduction

Almost 4 decades ago, simultaneously worldwide, as the causative of syndromes of an previously unknown disease in dogs, it was isolated canine parvovirus type 2 (CPV-2, canine parvovirus type 2) (Eugster et al. 1978; Appel et al., 1978; Burtonboy et al., 1979; Ganon and Povey, 1979; Johnson and Spradbrow, 1979, McCandlich et al., 1979).

Canine parvovirus type 2 (CPV-2) for many countries of the world and Europe continues to be one of the most troubling health problems of dogs (McCaw et al, 1998; Pollock and Carmichael, 1990; Carmichael, 2005; Decario, N et al., 2007). The disease first appeared with two syndromes; a non suppurative myocarditis associated with heart failure in dogs 4-8 weeks (Języka et al., 1979; Hayes et al 1979; Carpenter et al., 1980) and a severe enteritis accompanied by vomiting, diarrhea and death in puppies and adult dogs (Appel et al., 1978, 1979; Osterhaus et al., 1980; Meunier et al., 1981). However, thanks to its high contagious properties and sustainability of the virus to environmental factors today, especially parvoviral enteritis, is a concern of the first hand to the importance of viral diseases in dogs.

To date, in Europe and wider there are identified 3 serotypes of CPV-2 (CPV-2a, 2b and recently the 2c) (Decario, N et al., 2007). Two types of CPV-2, namely CPV-2a and CPV-2c have been recently isolated in Albania and genetically characterized (CAVALLI et al., 2014).

What is the situation regarding the spread out of the virus in Kosovo? Little can be said. There are reports of clinical symptoms similar to CPV-2 in dogs associated with high mortality. To the best of the authors´ knowledge up to currently in Kosovo there is neither a laboratory confirmation of CPV-2 in dogs nor any serologic evidence regarding antibodies to CPV-2.

Materials and Methods

Serologic survey for detection of HI antibodies to CPV-2

It were tested 124 never-vaccinated, clinically healthy dogs of various age, breed and sex. The age ranged from 6 to 156 months. Only seven of dogs were confined as puppies belonging to the age group equal or greater than 6 to 12 months old and 116 others were from greater than 12 to 156 months old. A good majority of them was mixed breed (98), while 13 were Illyrian shepherd, 9 English setters, 1 German shepherd, 1 Belgian shepherd and 1 Pekingese. Out of 124 dogs, 65 (52.4%) were males and 59 (47.5%) were females. Geographical coverage of dogs that were serologically tested included Kamenica (41), Rahovec (47), Peja (14), Prishtina (12) and Prizeren (10).

The criteria for the collection of sera for each dog available were the age equal or older than 6 months old and the dog should never be vaccinated. As the test to detect anti-CPV-2 antibodies was the hemagglutination-inhibition (HI) test performed according to an already well established method (Carmichael et al., 1980; Kumar et al., 2004). The serum samples for HI test were heat-inactivated at 56 ° C for 30 minutes and serial 2-fold dilutions were made of each serum in PBS, in 96-well U-bottom microtiter plates using 25 µl droppers and diluters. A total of 10 units of hemagglutination of canine parvovirus antigen with a hemaglutination (HA) titer of 1:1024/0.025 ml was added to each serum dilution and plates were mixed. The CPV-2 antigen used was prepared from a fecal sample of a 9-week old female puppy from Kamenica diagnosed of CPV-2 enteritis by HA test carried out at ISUV. After standing for 1 hour at room temperature, 50 µl of a 0.5% pig RBC suspension was added to each serum-virus mixture and incubated at 4°C until erythrocyte controls had formed clear buttons, usually after 3 hours. Each test series included erythrocyte, virus and serum controls.

Results and Discussion

Out of 124 sera tested with HI test, 96 (77.42%) resulted positive (titers ≥1:16 were considered positive), what meant that those dogs had already contracted the virus. HI titers varied from 1:16 to 1:2048. Out of 96 HI positive dogs, 48 (38.7%) were males and also 48 (38.7%) were females, being in a perfect equality of positivity. Thus no difference was shown between positive male dogs and positive female dogs. The overall seroprevalence of 77.42% in this study was similar high to another serologic survey carried out in dogs in Albania 2004, which was 83.9% (KUSI, 2004).

The seroprevalence in communities of Kamenica, Rahovec, Peja, Prishtina and Prizeren was 78.0%, 76.5%, 64.2%, 75.0% and 100%, respectively. It showed a high prevalence of CPV-2 antibodies in each community involved in the study as well as that at the nation level (Table 1). Nonetheless, no significant difference was found between the communities involved in the study. Such a finding indicates that the prevalence of anti-CPV-2 antibodies is high, and as a consequence, the CPV-2 itself is widespread all-over the country.

Table 1. *Seroprevalence in each district involved in the study and overall*

Communities	**No. of dogs tested**	**No. of dogs having a HI titer ≥ 1:16**	**Seroprevalence in %**
Kamenica	41	32	78.0
Rahovec	47	36	76.5
Peja	14	9	64.2
Prishtina	12	9	75.0
Prizeren	10	10	100.0
Total	**124**	**96**	**77.42**

As the main task of the serological survey was to investigate the spread of anti-CPV-2 antibodies among the dog population in Kosovo, we did not focus on the puppies less than 6 months old in order to avoid the maternal derived immunity and thus compromise the seroprevalence. However, it has to be noted that we found high level of HI antibodies to CPV-2 in all age-groups involved in the study. As shown in the Table 2, dogs that were studied were divided in five different groups of ages; **1**) 6-12 months old, 30 of the total, from whom 19 (63.34%) were positive and 11 (36.7%) were negative; **2**) > 12-24 months old, 34 of the total, from whom 26(76.47%) were positive and 8 (23.53%) were negative; **3**) >24-48 months old, 44 of the total, from whom 36 (81.8%) were positive and 8 (18.2%) were negative; **4**) >48-96 months old, 13 of the total, from whom 12 (92.3%) were positive and 1 (7.7%) was negative and finally **5**) >96 months old, 3 of the total, 3 (100%) were positive.

Table 2. *Distribution of anti-CPV-2 antibodies by age-group*

Age-group (months)	No. of dogs tested	No. of dogs having HI titers ≥ 1:16	No. of dogs having HI titers < 1:16
6 – 12	30	19	11
> 12 – 24	34	26	8
> 24 – 48	44	36	8
> 48 – 96	13	12	1
> 96	3	3	0
T O T A L (%)	**124 (100%)**	**96 (100%)**	**28 (100%)**

The older the age-group, the higher rate of dogs having HI antibody titers was the result. This way, the survey supported the fact that adult dogs today are generally immune as a result of previous contact with natural infection of CPV-2 or as a result of vaccination (Mason et al., 1987; Pollock et al., 1993).

In conclusion, based on the serologic, the situation allows little space for discussion about the necessity of strict and regular vaccination of puppies, considering every dog population nationwide.

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