



## Factors Associated with the Occurrence of Canine Parvoviral Enteritis in Dogs

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### ABSTRACT

A prospective study was conducted to identify the risk factors associated with the incidence of Canine Parvoviral Enteritis (CPE) in dogs. Total of 120 animals screened using PCR assay, 72.50 percentage of animal were found positive for Canine Parvo Virus (CPV). Incidence in history of unvaccinated and vaccinated dogs was 79.69 and 64.29 per cent respectively. Age-wise predisposition of CPE indicated that the highest incidence was observed in both less than 3 months (78.08 %) and 3 to 6 months of age group (77.42 %) followed by 6 to 12 months of age group (34.50 %). Incidence of CPE in scheduled and unscheduled vaccination was 30.00 and 83.33 per cent respectively. In this study, unvaccinated status, unscheduled vaccination and young age groups are found to be significant risk factors associated with the occurrence of CPE.

**Keywords:** Incidence, CPE, unvaccinated dogs, unscheduled vaccination

Canine Parvo Virus (CPV) is an acute contagious viral disease of dog's especially young ones with high morbidity and mortality. It occurs as an intestinal form with haemorrhagic foul smelling diarrhoea and vomition with severe dehydration, while the cardiac form is seldom seen. The incidence of CPE is reported from various continents viz., Africa, Asia, Australia, America and Europe (Nandi and Kumar, 2010; Miranda and Thompson, 2016). Though the vaccination is the first step in controlling any viral diseases of dogs, CPV vaccine does not ensure 100 % protection (Coyne, 2000). In general, HI titre of 1:80 is considered as a protective titre in puppies against CPV infection and vaccination will turn into immunization only when the titre level is below 1:10.

There are varieties of hindering factors contributing the

occurrence of CPE in vaccinated animals which include MDA neutralization, lack of seroconversion variability in individuals, immune status of puppy, vaccination schedule, efficacy of vaccine, mutant virulence over the prototype vaccines and poor vaccine storage (Singh *et al.*, 2013; Schultz, 2006; Nandi and Kumar, 2010). Though the various reasons are enlisted for CPE occurrence in vaccinated animals, neutralization of CPV by MDA and improper or irregular vaccination scheduled animals are the major reasons behind this outbreak (Singh *et al.*, 2013). The associated risk factor analysis in CPE in vaccinated animals is seeking its demand to understand the disease

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epidemic which may help to make an appropriate disease control strategy. This manuscript aimed to elucidate the role of vaccination and age as risk factors in the occurrence of CPE in vaccinated animals.

### MATERIALS AND METHODS

In this prospective analysis, a total of 120 animals of varying breed and sex were included with the ailment of haemorrhagic foul-smelling diarrhoea and vomition with severe dehydration which was presented to Madras Veterinary College Teaching Hospital, Chennai, India. A structured questionnaire regarding age and vaccination history was collected to categorize the suspected animals into an age-wise category such as less than 3 months, 3 – 6 months and 6 - 12 months of age group and vaccinated and unvaccinated groups. Among vaccinated animals scheduled vaccination to consist of one primary and 3 boosters at 21 days interval (WSAVA guidelines) and unscheduled vaccination consist of animals which do not follow the above protocol (Day *et al.*, 2010).

From the suspected animals, stool samples were collected and processed for molecular confirmation. The DNA extraction from stool samples was performed by hot and boiling method (Schunck *et al.*, 1995) and PCR was done with a published primer of Buonavoglia *et al.*, 2001. The PCR was performed targeting H gene forward sequence (CAGGTGATGAATTTGCTACA) and reverse (CATTTGGATAAACTGGTGGT). The positive samples will yield a PCR product size of 630 bp which is specific for CPV 2 confirmed cases (Fig. 1). In statistical analysis, Pearson chi square test and Odds ratio was calculated using SPSS package (Version 20.0).

### RESULTS AND DISCUSSION

In this epidemiological study, 72.50 % CPV incidence was reported from the 120 suspected stool samples (Table 1). This is in agreement with Miranda *et al.* (2015) and Markovich *et al.* (2012), who has also documented 77.5% and 78% of CPV incidence by using PCR assay respectively. But, Mittal *et al.* (2015) and Godsall *et al.* (2010) documented the lower incidence of 6.74% and 58% by PCR respectively. The possible reason behind the higher incidence might be due to the framed animal inclusion criteria which are less than one year age group of animals with suspicion of haemorrhagic gastroenteritis

(Miranda *et al.*, 2015). The lower incidence might be due to the inclusion of all similar systemic illness and which makes a decrease in the incidence pertaining to the study (Godsall *et al.*, 2010).

**Table 1:** Age-wise incidence of Canine Parvoviral Enteritis

Age group	Total no. of screened animals	Total no. of positive animals (%)	$\chi^2$ - value
< 3 months	73	57 (78.08)	(11.348)**, P = 0.003
3 – 6 months	31	24 (77.42)	
> 6 – 12 months	16	6 (34.50)	
<b>Total</b>	<b>120</b>	<b>87 (72.50)</b>	

On the age-wise incidence of CPV were analyzed and the higher positivity was encountered in both less than 3 months (78.08 %) and 3 to 6 months of age groups (77.42%) and 34.50% of incidence in 6 to 12 months of age group (Table 1). The increased susceptibility of less than 6 months of age dogs for CPE were in agreement with other researchers (Umar *et al.*, 2015; Thomas *et al.*, 2014; Strepita *et al.*, 2013) who documented the increased chance of CPV in less than 3 months followed by 3 – 6 months and 6 – 12 months of age group. Due to the affinity of CPV to the mitotic intestinal cells and gut flora and fauna changes especially during weaning time may facilitate the increased chances of CPV in less than 3 months of age group. Similarly, sudden fall in MDA level after 3 months of age makes 3 - 6 months age group as vulnerable for CPV infection in endemic areas (Strepita *et al.*, 2013). On contradictory with the present study, Phukan *et al.*, 2010 and Carmichael *et al.* (1983) documented the highest incidence of CPV with 7-12 months followed by 1 – 6 months, and above 12 months age groups. The possible variation in these results might be due to improper vaccination schedule, lack of booster vaccination and improper storage of vaccines leads to vaccination failure in 7 -12 months as more prone to the disease (Carmichael *et al.*, 1983).

Regarding vaccination status of dogs increased positivity noted in unvaccinated history dogs (79.69 %) than vaccinated (64.29%) (Table 2). The increased positivity of CPV in the unvaccinated population was documented many of the researchers (Houston *et al.*, 1996; Parrish, 1999; Singh *et al.*, 2013; Miranda and Thompson, 2016). In this study, more than 50 percent of CPV incidence was

**Table 2:** Vaccination status-wise incidence of Canine Parvoviral Enteritis

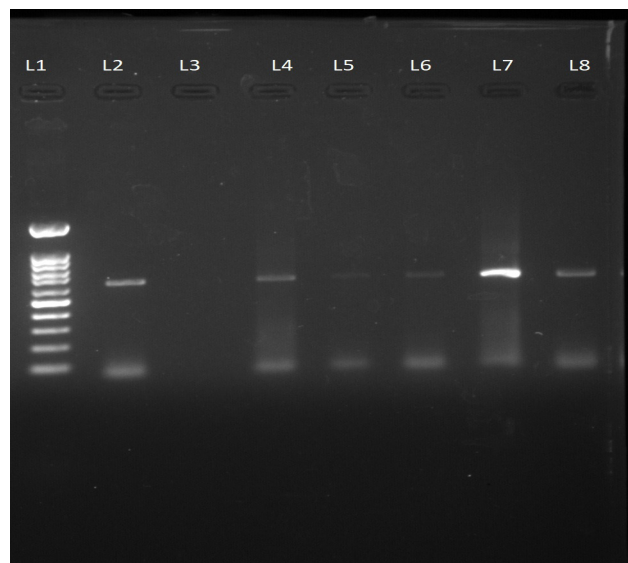
Category	Total no. of animal screened	Total no. of positive animals	Percentage positivity	Odds ratio	95 % Confidence interval	P value
<b>Total no of animal screened (n = 120)</b>						
Vaccinated animals	56	36	64.29	2.179	0.9620 – 4.940	(3.554)NS P = 0.059
Unvaccinated animals	64	51	79.69			
<b>Within the vaccinated animals (n = 56)</b>						
Scheduled	20	6	30.00	11.667	3.188 – 42.692	(15.929)** P = 0.000
Unscheduled	36	30	83.33			
<b>Within unscheduled animals (n = 36)</b>						
Primary (P) alone	14	13	92.86	1	—	—
P + First booster (B1)	11	9	81.82	0.346	0.027 – 4.418	(0.711) <sup>NS</sup> P = 0.399
P + B1 + Second booster (B2)	11	8	72.73	0.593	0.078 – 4.498	(0.259) <sup>NS</sup> P = 0.611

observed in vaccinated animals and this might be due to improper or irregular vaccination, use of the poor quality vaccine and improper storage and/or handling of vaccine. The variation in the vaccination coverage might be due to adopting vaccination schedule based on endemic status, variation in incidence based on a location-wise, sampling frame of individual study and awareness among pet owners regarding the clean management and vaccination.

In this study increased incidence of CPV in unscheduled vaccination (83.33 %) was noted than scheduled vaccinated animals (30.00 %) (Table 2). The high incidence of CPV in unscheduled vaccinated animals was also reported by Castro *et al.*, 2007 and Srinivas *et al.*, 2013. Castro *et al.*, 2007 documented about 93.47 % incidence of CPV in unscheduled vaccinated animals.

Unscheduled vaccinated animals further identified as Primary, Primary + first booster and Primary + first booster + second booster and revealed 92.86, 81.82 and 72.73 percent respectively (Table 2). Among the unscheduled vaccinated animal category, higher incidence observed in primary followed by one and two booster injected animals. There are many hindering factors might responsible for the variation in the incomplete vaccinated animal group incidence *viz.*, MDA neutralization, a sudden change in diet at weaning, immunity of individual dogs, nosocomial infections and co-infection. The above factors are almost in significant reasons despite MDA neutralization in individual animals play a major role in CPV occurrence (Parrish, 1999; Miranda and Thompson, 2016). Even after

scheduled vaccination 30% of incidence was observed in our study. The possible reason behind this, 10% of puppies are not enough to get a protective titre due to the persisting maternal antibodies interference role (Miranda and Thompson, 2016).



**Fig. 1:** Agarose gel (1.5%) electrophoresis showing H primer PCR amplicon from CPV confirmed dog faeces. L1 – 1.2 kb size ladder, L2 – Positive control, L3 – Negative control, L4- 8 – Positive samples for CPV 2 PCR product showing 630 bp size

## CONCLUSION

In this epidemiological study, out of 120 animals, 72.50% incidence of CPV was documented by using PCR assay.



Unvaccinated animals, less than 6 months of age group and unscheduled vaccination are found to be significant risk factors associated with the occurrence of CPV. However using this descriptive study data, analytical oriented epidemiology studies are warranted to know the more risk factors associated with a disease outbreak in a different location.

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