

Pilot Scale Study on the Prevalence and Associated Risk Factors of Canine Distemper and Canine Parvovirus in Ahmadabad and Gandhinagar Cities of Gujarat, India

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Abstract

Canine parvovirus (CPV) and canine distemper virus (CDV) infections are the most common and contagious viral infections of dogs. The present study investigated the burden of CPV and CDV infections among the domestic dogs in Ahmadabad and Gandhinagar cities of Gujarat, India with the specific objective of determining the case prevalence and the associated risk factors of the disease. Faecal samples were collected from dogs (n = 507) attended at different private and government veterinary polyclinics, from 2020 to 2021. Data regarding sex, age and season were recorded with respect to prevalence case. Faecal samples were tested for CPV using Ubio Quick VET Canine Parvovirus antigen rapid test kit and (nasal and ocular secretion) were tested for CDV antigen using canine distemper virus antigen rapid test kit. The results of the present study revealed an overall case prevalence of 8.68% for both infections, while 5.71% and 11.63% Case prevalence were reported for CPV and CDV, respectively. The Case prevalence among dogs examined according to sex was 6.82% for CPV and 11.36% for CDV in male and 3.23% for CPV and 12.26% for CDV in female. Pups had the higher Case prevalence of both infections than those of adults. Case prevalence of DV infection was high in the winter season (14.40%), while the lowest (3.03%) in monsoon season. For CPV also Case prevalence (7.67%) was higher in winter season while no preva-

lence reported for CPV in summer. Age, sex and seasonal variations may be considered as possible risk factors in the Case prevalence of CPV and CDV infections. Thus, the identification of the potential risk factors associated with these diseases may be helpful for designing their prevention and control strategies.

Key words: CPV, CDV, dog, Epidemiology, Prevalence, Risk factors

Companion animals, like dogs serve an important meaning in our communities. Number of viruses can cause acute and severe illness in dogs, out of that canine parvovirus (CPV) and canine distemper virus (CDV) are highly contagious and virulent (Nandi and Kumar, 2010). Both CPV and CDV are highly infectious and serious disease attacking the respiratory, gastrointestinal and nervous systems of dogs (Mylonakis *et al.*, 2016; AVMA, 2022). CPV is a small (diameter of 25nm), non-enveloped DNA virus belonging to the family Parvoviridae, and is frequently reported in India (Kulkarni *et al.*, 2019). CPV causes acute hemorrhagic enteritis and myocarditis in dogs with high morbidity (100%) and mortality up to 10% in adult dogs and 91% in pups (Nandi and Kumar, 2010). The infection is spread from infected dogs to naïve dogs by direct or indirect contact with their infected faeces. The CPV can affect all breeds of dogs at any age; however, puppies (6 weeks to 4 months) are most affected (Nandi and Kumar, 2010). The disease is characterized by two prominent clinical forms; enteritis with vomiting

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and diarrhea and myocarditis followed by heart failure (Mylonakis *et al.*, 2016). Early symptoms are depression, loss of appetite, vomiting, high fever and severe diarrhea. Rapid dehydration is a sign of danger, and dogs may continue to vomit and have diarrhea until death, usually within 3 days after onset of symptoms (Mylonakis *et al.*, 2016). Within an infected litter, 70% pups will die by heart failure at the age of 8 weeks and the remaining 30% will have pathological changes that may result in death after months or years (Nandi and Kumar, 2010). Diagnosis can be done by ELISA, PCR and viral antigen detection method (Gupta and Singh, 2016) but antigen detection from fecal sample is the most common diagnostic assay for CPV (Marulappa and Kapil 2009). No specific treatment is available for treatment of CPV, therapeutic measures including symptomatic treatment and supplementation with immune boosting agents. Isolation of infected dogs is recommended to minimize spread of infection (Schultz and Larson, 2007).

CDV, a relatively large (150–250 nm) single-stranded RNA virus with a lipoprotein envelope, is a Morbillivirus in the family Paramyxoviridae (Degene and Zebene, 2019). Canine distemper virus (CDV) is considered as acute, highly infectious, and polysystemic infection of domestic dogs distributed worldwide. Its fatality rate is second after the rabies. CDV is common in young dogs, especially in their first year of life but many cases are also seen in adults (Degene and Zebene, 2019). The transmission is through aerosolization of respiratory excretion and other body excretions and secretions (e.g., urine) can also result in infection transmission to susceptible ones. Transmission through placenta is also reported in domestic animals (Alfano *et al.*, 2022). The virus often enters through the upper respiratory tract and grows in macrophages before spreading to the tonsils and local lymph nodes, where it can replicate for two to four days' post infection. During a week's time, CDV multiplies in lymphoid organs of lamina propria and small intestine, Kupffer's cells, mesenteric lymph nodes, and spleen. Dogs with the neurologic condition may experience hyperkeratosis (thickening) of the footpads and nose, the term "hardpad disease" has become another common name for CDV response that

cause severe clinical illness at 2-3 weeks and mortality by 3–4 weeks. Puppies with distemper develop pneumonia, conjunctivitis, rhinitis, diarrhoea and tracheitis (Degene and Zebene, 2019). Canine distemper is considered for diagnosis of any febrile condition with multisystemic manifestations. There is no specific treatment for canine distemper and is best prevented by vaccination.

Rapid diagnosis of CPV and CDV infections are especially important in dogs in order to isolate infected dogs to prevent secondary infection spread to susceptible animals. The present study aimed to determine the case prevalence and unveil the associated risk factors of CPV and CDV in the Ahmadabad and Gandhinagar cities of Gujarat, India.

Materials and Methods

Study location:

This Study was conducted in the Ahmadabad and Gandhinagar cities of Gujarat, India covered three major seasons: summer (April–June), monsoon (July–September) and winter season (October–March) in each year and the temperature range was between 10 to 45°C.

Study Design:

To determine the risk factors (sex, age, and season) for susceptibility to CPV and CDV, a cross-sectional study with 507 dogs (Ahmadabad and Gandhinagar have approximately 1.5 lakh pet dogs) was conducted. Animals were divided into age and sex categories. There were 155 females and 352 males among them. There were 321 puppies (under 1 year) and 186 adults (over 1 year), respectively. The diagnosis was separated into three seasons for the years 2020 and 2021; winter, summer, and monsoon. Samples (Faecal, nasal secretion and ocular secretion) were obtained from different private and public veterinary polyclinics in the cities of Ahmedabad and Gandhinagar. These animals represented mixed clinical signs such as lethargy, loss of appetite, abdominal pain and bloat, fever or low body temperature (hypothermia), vomiting, diarrhea, nasal discharge and coughing. Gujarat Veterinary Research and Diagnostic Centre (GVRDC), in Ahmadabad, handled all

diagnostic procedures.

Laboratory Diagnosis

Diagnosis of CPV was carried out using standard antigen detection kits from UbioQuickVET Canine Parvovirus Antigen Rapid test (Sensitivity-99.8%, Specificity-99%) as per manufacturer's protocol. Briefly, the fecal samples were collected directly from the rectum of the animal with the help of a sterile cotton swab and stored at 2-8°C till tested. Then, swab containing sample was inserted into the assay diluent tube (500µl) and mixed with diluent by agitation for antigen extraction. Then, 4-5 drops of extracted specimen were applied to drop on the sample window of lateral flow strip containing monoclonal anti-CPV antibodies and results were recorded within 10 min.

Diagnosis of CDV was carried out using standard antigen detection kit (qualitative), bioQuickVET Canine Distemper Virus Antigen Rapid Test (Sensitivity-99%, Specificity-98.7%). Briefly, samples (Nasal/ ocular secretion) were collected directly from the animal with the help of a sterile cotton swab and then swab containing sample was inserted into the assay diluent tube (500µl) and mixed with the diluent for few seconds to allow antigen extraction. Then 2-3 drops of extracted specimen applied to drop on the sample window of lateral flow strip containing monoclonal anti-CDV antibodies and results were recorded after 10 minutes.

Statistical Analysis

The collected data was submitted to logistic regression analysis, and the findings were expressed as percentages and displayed as tables

(Table I, and II). Odds ratio at 95% confidence interval was utilized to evaluate the risk factors. $P < 0.05$ values were regarded as measure of significance.

Results and Discussion

The overall case prevalence for both CPV and CDV infection among dogs studied from the Ahmadabad and Gandhinagar cities of Gujarat, India was 8.68%. The overall prevalence reported for CPV was 5.71% (Table I).

In the present study, a low Case prevalence rate was observed for CPV. The above finding was in accordance with those of Phukan *et al.* (2010). Wazir *et al.* (2013) also reported a low prevalence (6.93%) in Jammu. However, Sayed-Ahmad *et al.* (2021) reported higher prevalence of CPV (59.7%). Other studies conducted by Archana *et al.* (2010), Roy *et al.* (2010), and Khare *et al.* (2019) also reported comparable higher prevalence of 45.30% and 65.04% in Jabalpur and Chhattisgarh, respectively. Such high prevalence might be due to occurrence of prevalent infection and rapid transmission in the population under study. The dissimilarity in the reporting of prevalence of CPV infection might be due to the diagnostic tools used among different studies and wide variation in the number of samples tested as well as different geographically regions under various studies. The ELISA based antigen detection is the most commonly used diagnostic test for parvovirus (Schmitz and Coenen, 2009), however, the present study used lateral flow-based antigen detection test because of its rapidity, easy result interpretation and less turnaround time. The main clinical signs of CPV disease are bloody diarrhea,

Table I: Univariable logistic regression for risk factors for CPV in dogs

Risk Factor	Category	No. +ve/total (%)	OR	95% CI		P-value
				Lower	Upper	
Sex	Male	24/352 (6.82)	2.1501	0.4963	1.6095	0.13
	Female	05/155 (3.23)				
Age	Pup	19/ 321 (5.92)	0.9576	0.4315	2.1255	0.92
	Adult	10/186 (5.38)				
Season#	Winter	24/313 (7.67)	0.3563	0.1601	0.7932	0.01
	Monsoon	05/99 (5.05)				
	Summer	00/95 (0.00)				

Table II: Univariable logistic regression for risk factors for CDV in dogs

Risk Factor	Category	No. +ve/total (%)	OR	95% CI		P-value
				Lower	Upper	
Sex	Male	40/352 (11.36)	0.8938	0.4963	1.6095	0.71
	Female	19/155 (12.26)				
Age#	Pup	46/ 321 (14.33)	0.4580	0.2400	0.8740	0.02
	Adult	13/186 (6.99)				
Season	Winter	46/313 (14.70)	0.6973	0.4674	1.0404	0.08
	Monsoon	03/99 (3.03)				
	Summer	10/95 (10.52)				

(p< 0.05)

vomiting, and dehydration. Present findings are according with the previous studies (Thomson and Gagnon, 1978, Prittie, 2004) and Khare *et al.* (2019) indicated that significant association between season and prevalence of the infection in studied area. Disease was more prevalent (7.67%) in winter duration in comparison with monsoon (5.05%) (Table I). This higher prevalence in winter season may be related with stress due to change in weather (Subramanian *et al.* 2021). Present study has not been reported any case of CPV in summer, which might be associated with high temperature during summer which may reduce the survival of virus in environment and further spread to susceptible host. Kalita *et al.* (2022) also reported that higher prevalence during winter and early spring due to significant seasonal diurnal temperature differences and changeable climates during these seasons. Also, low temperature supports survival and rapid multiplication of virus (Qi *et al.*, 2020). In contrast, Sayed-Ahmed *et al.* (2021) reported that the higher prevalence of CPV during in summer (77.1%) followed by spring (55.5%), autumn (25%) and winter (16.6%). However, many studies have reported that CPV infection rates are higher in spring and autumn (Qi *et al.*, 2020). Such findings were possibly due to increased outdoor activities of dogs and human during spring and autumn (Zhuo *et al.*, 2015 and Zhao *et al.*, 2016).

Present study could not find significance difference in the Case prevalence of CPV according to age (Table I). However, these findings are dissimilar to the previous findings, where in CPV infection is reported to be more prevalent

in young puppies compared to adults (Nandi and Kumar, 2010). This higher prevalence rate below 6 months age has been reported in various earlier studies (Kalita *et al.*, 2022; Kelman *et al.*, 2020; Mokhtari *et al.*, 2018). The higher occurrence of CPV in puppies is attributed towards high the affinity of the virus for accept intestinal crypt cells in weaning pups. Further, higher mitotic index of these cells and microbiome due to changes in weaning pups makes them more susceptible (Hasan *et al.*, 2018). In contrast, Phukan *et al.* (2010) reported highest incidence in the age group of 7-12 months followed by 1-6 month, 13 months and above. The variation in the prevalence among different age groups might be due to improper timing of vaccination, skipping booster doses and improper maintenance of cold chain for storage of vaccines (Behera *et al.*, 2022).

Present study did not find any significant association between Case prevalence and sex of the animal. Previous reports also suggest the similar findings that there was no impact of sex on the occurrence of CPV (Banja *et al.*, 2002; Sanjukta *et al.*, 2011). However, Males (6.82%) are more susceptible than females (3.23%). Reason behind this high incidence in males may be due to biased number of tested dogs (69.4%). The present findings are in accordance with Gombac *et al.*, (2008). Other studies reported higher prevalence of CPV in males (86.21%) than females (13.79%) (Bahera *et al.*, 2015). Thomas *et al.* (2014) also reported a higher incidence of CPV (78.26%) in male dogs. The higher incidence in males was also reported by Phukan *et al.*, (2010), Srinivas *et al.* (2013), Tajpara *et al.* (2009), Deka

et al. (2013). Due to specific behavioral patterns and pet owners' wish for keeping male dogs, there may be a higher probability of exposure and a higher prevalence of CPV in male dogs (Deka *et al.* 2013).

In the present study, overall case prevalence of CDV was 11.63%. Finding of the present study are similar to Dorji *et al.*, (2020), wherein reported prevalence was 11.3%. However, The prevalence (observed in this study is not in accordance with study conducted in Madras city of India where in 1.35% animals were positive for CDV (Alex and Dhanapalan, 1994). However, varied prevalence of CDV has been recorded by different workers 6.1 % by Gauveia *et al* (1987), 21.7% by Ek-Kommonen (1997), 56% by Guo *et al* (1986), 42% and 74% by Moeller *et al.*, (1993). The degree of specificity of the diagnostic method, the phase of infection, immunological status of the animal at the time of the tests, and the geographical location all are contributing factors to this wide range in CDV prevalence reporting. Also, these variations observed in the prevalence are difficult to explain due to the different study design areas under study and differences in the diagnostic methods as well as objective of the study.

Pups are more susceptible than adults for CDV and the present study indicated the significant associate between age and prevalence ($P < 0.5$) (Table II). Young dogs, especially neonates and recently weaned dogs, are generally more susceptible to CDV infection, demonstrating a relationship between susceptibility and age (Headley and Graça, 2000). Present findings are similar to the results of previous work (Ek-Kommonen *et al.*, 2003) and are in agreement with the affirmation that neonates, recently weaned (Krakowka and Koestner, 1976), or young dogs (Swango 1989), are more affected by CDV. This clearly establishes a relationship between susceptibility and age. The susceptibility of neonates or young dogs to CDV is related to the start of the steady reduction of maternal antibodies soon after birth (Headley and Graça, 2000), this being directly proportional to the growth rate of these animals (Evermann and Kennedy, 2011). In contrast Dorji *et al.* (2020) reported no significant differences in the seroprevalence of CDV among different

age groups.

There was no significant association found between prevalence of CDV with season around year. However, animals were more susceptible to CDV in winter (14.70%) followed by Summer (10.52%) and monsoon (3.03%) (Table II). One another study revealed that more CDV cases are reported in winter months (Alex and Dhanapalan, 1994). The lower temperature favors viral survival (McDermott *et al.*, 2023), and possibly induces immuno-depression in neonates and recently weaned animals. The lack of sufficient maternal antibodies, their scarcity, the existence of another severe condition concurrently, or any combination of these factors may make this imposed immune suppression more pronounced. In contrast, Dorji *et al.*, (2020) reported higher sero-positivity to CDV during the summer season compared to winter. Similarly, Patronek *et al.*, (1995) also recorded higher occurrence of CDV in summer than winter season.

Regarding the sex of animal, there was no observable difference in case prevalence between males (11.36%) and females (12.26%) (Table II). Findings are similar to Dorji *et al.*, (2020) that there are no significant differences in the seroprevalence of CDV among different sexes were reported. Headley and Graça, (2000) also reported that significant differences in susceptibility among different sexes. These findings support the idea that host sex has no bearing on the prevalence of CDV. (Gorham, 1966). Some study reported that male dogs may be at higher risk of exposure, probably because of their aggressive and roaming behaviors (McGreevy *et al.*, 2018). However, different results were obtained in other studies. More males than females (60:40%) were infected in Madras City, India (Alex and Dhanapalan, 1994), in -contrast (Guo *et al.*, 1986) reported the high prevalence in females than males. The present study revealed the infection induced by both CPV and CDV are more prevalent in young pups than adult during the winter season. Regarding the sex of animal both viral infections are more prevalent in males in comparison to females.

Conclusion

The case prevalence studies in some way benefi-

cial to track the infection among the susceptible population. Furthermore, epidemiological intelligence system is lacking in these study area with respect to canine infectious disease. There are no specific antiviral drugs to treat CPV and CDV. Therefore, the present study was conducted to determine the case prevalence of CPV and CDV infection in Ahmedabad and Gandhinagar city of Gujarat, for which effective control measures should be designed considering the view of different risk factors.

Competing interest:

The authors declare that they have no known competing financial interests.

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