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# Canine Parvovirus Type 2

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Chao-Nan Lin and Shu-Yun Chiang

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## Abstract

Canine parvovirus (CPV) enteritis is characterized by intestinal hemorrhage with severe bloody diarrhea. The causative agent, CPV-2, was first identified in the late 1970s. CPV is a nonenveloped, linear, single-stranded DNA virus with a genome of approximately 5 kb, and it belongs to the genus *Parvovirus*, together with feline panleukopenia virus, mink enteritis virus, raccoon parvovirus, and porcine parvovirus. An antigenic variant, CPV-2a, identified within a few years after the emergence of CPV-2, and another variant, CPV-2b, began appearing in the canine population in 1984. In 2000, a novel antigenic variant, CPV-2c, was first detected in Italy. This chapter focuses on the history, viral evolution, epidemiology, pathogenesis, clinical signs, diagnosis, vaccination, and prevention of CPV-2.

**Keywords:** canine parvovirus type 2, CPV-2, viral evolution, antigenic variants, epidemiology, pathogenesis

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## 1. Introduction

Canine parvovirus (CPV) infection is characterized by clinical gastroenteritis with severe hemorrhagic diarrhea and is a common infectious disease in younger dogs [1]. Gastroenteritis caused by CPV type 2 (CPV-2) infection, especially that caused by the newer variants of CPV-2, may progress rapidly [2, 3]. Dehydration is the rapid onset of the loss of bodily fluids caused by severe vomiting, diarrhea, or hemorrhagic diarrhea. Death occurs in as early as a few days after disease onset. This chapter documents the history, viral evolution, epidemiology, pathogenesis, clinical signs, diagnosis, vaccination, and prevention of CPV-2.

### 1.1. History

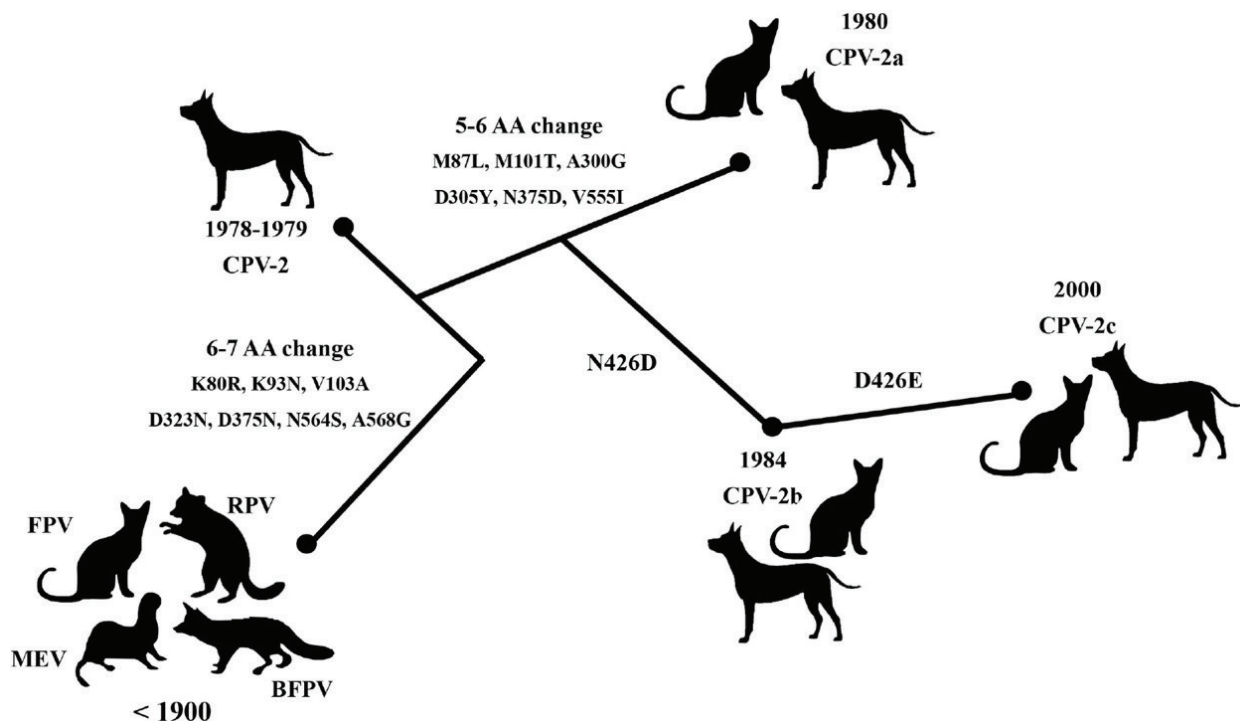
Canine parvoviral enteritis is characterized by intestinal hemorrhage with severe bloody diarrhea [4]. The causative agent, CPV-2, was first identified in the late 1970s [5]. CPV is a nonenvel-

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oped, linear, single-stranded DNA virus with a genome of approximately 5 kb, and it belongs to the genus *Parvovirus*, together with feline panleukopenia virus (FPV), mink enteritis virus, raccoon parvovirus, and porcine parvovirus [6]. CPV-2 is distinct from CPV-1, also known as canine minute virus, which belongs to the genus *Bocavirus*. Indeed, CPV-2 is believed to have originated from FPV or a closely related FPV-like parvovirus of wild carnivores (**Figure 1**) [7, 8]. Various hypotheses for how this may have occurred have been suggested, including direct mutation from FPV and contact between cats and dogs kept as companion animals within the same home [8].

## 1.2. Viral evolution

Shortly after CPV was first identified in 1978, the original virus, CPV-2, was subsequently replaced in the dog population by strains carrying small antigenic variations (termed 2a, 2b, and 2c) of the VP2 protein that could be distinguished by monoclonal antibodies and genetic analysis. An antigenic variant, CPV-2a, identified within a few years after the emergence of CPV-2 [9, 10], and another variant, CPV-2b, began appearing in the canine population in 1984 [11]. A novel antigenic variant, Glu-426 mutant (now termed CPV-2c), was first detected in Italy in 2000 [12]. New antigenic variants of CPV-2 have been observed in epidemics around the world and are soon replacing the original CPV-2. CPV-2a variant shows several substitutions within the VP2 protein, including Met87Leu, Ile101Thr, Ala300Gly, and Asp305Tyr. Furthermore, CPV-2b variant has been identified to contain an additional amino acid change, Asn426Asp [13, 14]. These two antigenic variants further evolved into new CPV-2a and -2b



**Figure 1.** Cartoon of the evolution of canine parvovirus type (CPV-2). CPV-2 is believed to have originated from FPV or a closely related FPV-like parvovirus of wild carnivores (modified from Truyen [8]).

types, with amino acid substitutions at residue 297 (Ser to Ala), during the 1990s [15]. Antigenic variant CPV-2c was identified only one residue substitution (Asp426Glu) as differ from CPV-2a and -2b [12].

In addition to the amino acid substitution described above, amino acid substitutions Tyr324Ile, Gln370Arg, and Thr440Ala in the surface of the VP2 were also noted in the recent year [16]. Review of CPV-2 sequences from GenBank showed that this Ile324 variant of CPV-2a is also found in several countries, including Korea [17, 18], China [19–25], Thailand [26], Uruguay [27, 28], Japan [29], Taiwan [30, 31], and India [32, 33]. Surprisingly, aside from the Uruguayan strains, this Ile324 CPV-2a variant is only distributed in Asian countries. Previous study has shown that residue 324 of VP2 is subject to positive selection among all carnivore parvoviruses [34]. This residue is adjacent to the residue 323, which together with residue 93 is known to be affected in host range and tropism of canine transferrin receptor binding [35]. The mutation of VP2 residue 323 may affect interactions between residues in neighboring loops of either the same VP2 molecule or the threefold-related VP2, greatly reducing replication in canine cells [36]. The Gln370Arg change is unique in the Taiwanese [85] and Chinese CPV-2c strains [22, 25]. This substitution is also observed in Chinese panda parvovirus [37]. Residue 370 is located between residues 359 and 375, which constitutes a flexible surface loop of the capsid protein that is adjacent to a double Ca<sup>2+</sup>-binding site. They were found to be essential for virus infectivity. Changes in them are correlated with the ability of the virus to hemagglutinate erythrocytes [38]. In addition, VP2 position 440 is located near a major antigenic site. The Thr440Ala change was found in China, the USA [39], Italy [14], Argentina [40], Uruguay [27], India [33], and Taiwan [85].

### 1.3. Epidemiology

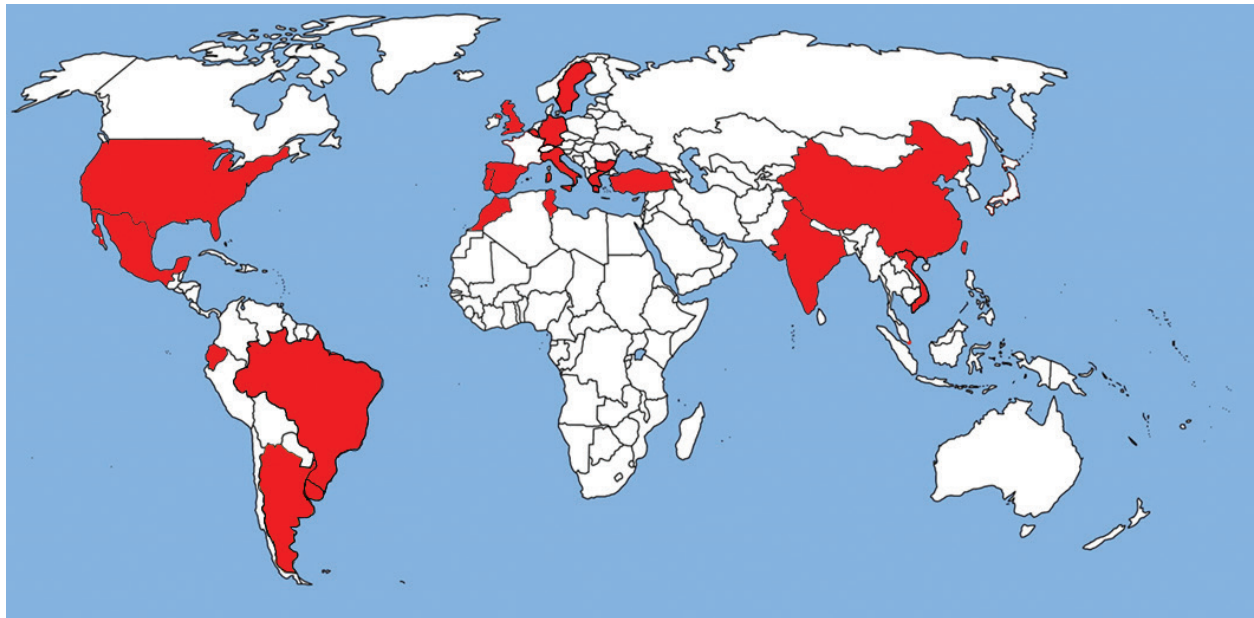
According to the epidemiology surveillance of CPV-2, the distribution of these three antigenic variants (-2a, -2b, and -2c) is summarized in **Table 1**. Previous study has revealed that the oldest CPV-2c variant was isolated in 1996 in Germany [41]. Epidemiological surveillance in

Country	Positive of strain detected			Reference
	CPV-2a	CPV-2b	CPV-2c	
<b>Europe</b>				
Italy	+	+	+	[2]
Portugal	+	+	+	[16]
Spain	+	+	+	[78]
France	+	+	+	[79]
UK	+	+	+	[41]
Belgium	+	-	+	[79]
Germany	+	+	+	[79]
Greece	+	+	+	[43]

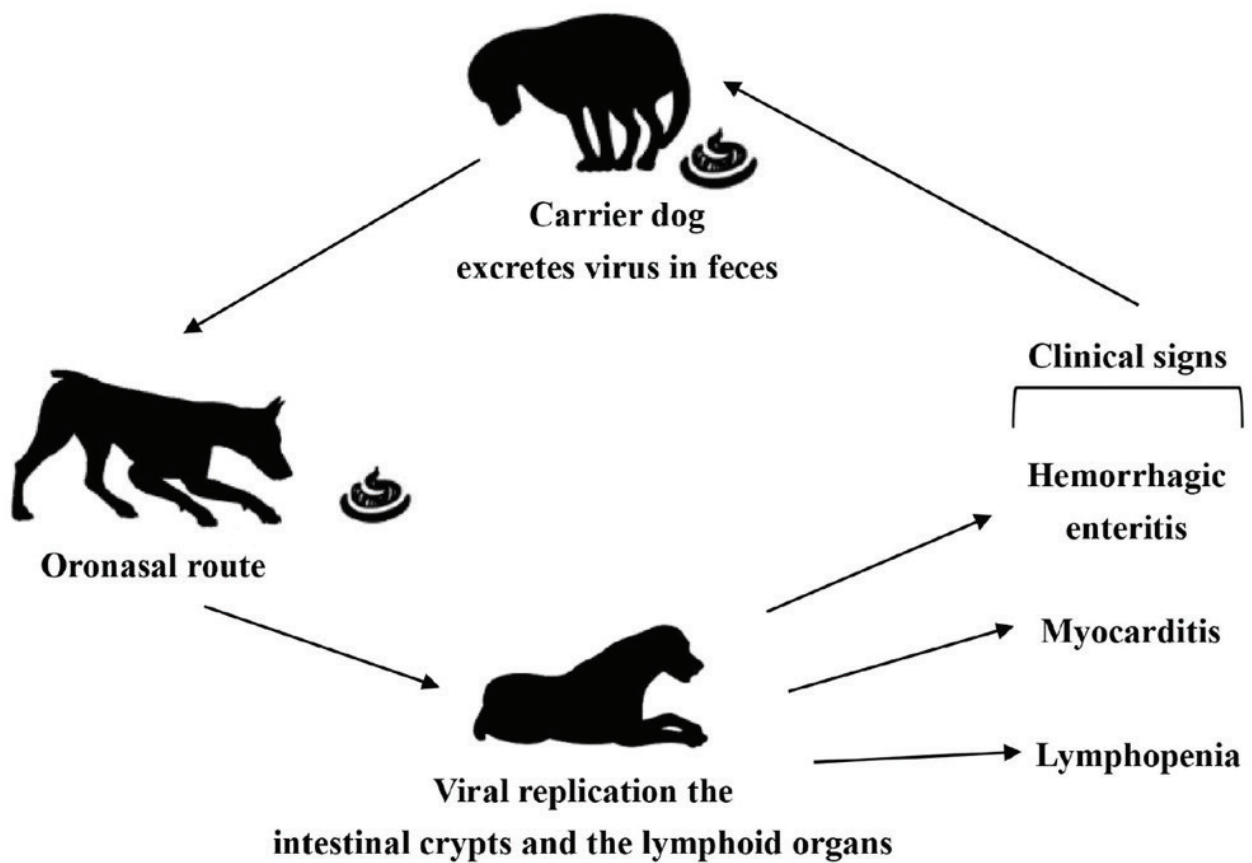
Country	Positive of strain detected			Reference
	CPV-2a	CPV-2b	CPV-2c	
Switzerland	+	+	+	[41]
Czech public	+	+	-	[79]
Romania	+	-	-	[79]
Hungary	+	-	-	[80]
Bulgaria	+	+	+	[81]
Turkey	+	+	+	[82]
<b>Africa</b>				
Tunisia	+	+	+	[47]
Morocco	+	+	+	[53]
<b>North America</b>				
USA	+	+	+	[39]
<b>South America</b>				
Uruguay	+	+	+	[83]
Argentina	+	+	+	[50]
Brazil	+	+	+	[84]
Ecuador	+	+	+	[51]
Mexico	-	-	+	[52]
<b>Asia</b>				
India	+	+	+	[55]
Taiwan	+	+	+	[85]
Korea	+	+	-	[18]
Japan	+	+	-	[15]
China	+	+	+	[19]
Thailand	+	+	-	[26]
Vietnam	+	+	+	[54]
<b>Oceania</b>				
Australia	+	+	-	[86]

**Table 1.** Detection of the canine parvovirus variants in the world.

Europe shows that CPV-2c is predominant currently in Italy, Germany, and Spain and is also extensively co-distributed with CPV-2a or -2b in Portugal [42], Belgium, France, Greece [43], Bulgaria [44], Sweden [45], Turkey [46], and the United Kingdom. In recent years, CPV-2c has also been widespread in Tunisia [47], the USA [39], Uruguay [48], Brazil [49], Argentina [50],



**Figure 2.** Distribution of canine parvovirus type 2c (CPV-2c) variants around the world. Red color represents that countries reporting CPV-2c case.



**Figure 3.** Transmission and clinical symptoms of canine parvovirus type 2 (CPV-2). Transmission of CPV-2 occurred by the fecal-oral route, after exposure to CPV-2 in feces and/or vomit, or viral-contaminated fomites.



Ecuador [51], Mexico [52], and Morocco [53]. Surprisingly, the CPV-2c variant has not been prevalent in Asia since the first identified in Vietnam in 2004 [54]. Only a few CPV-2c strains have been reported in India [55], China [19, 22, 25], and Taiwan [85]. To date, either CPV-2a or -2b has been prevalent in Asian countries [15, 17–19, 22, 25, 26, 29–33, 55–59]. Phylogenetic analysis demonstrated that the recent CPV-2c isolate from Taiwan shares a common evolutionary origin with Chinese strains of CPV-2c, as classified into novel Asian CPV-2c variants (Phe267Tyr, Tyr324Ile, Gln370Arg, and Asp426Glu) [85]. **Figure 2** summarizes the distribution of the CPV-2c variant around the world.

#### 1.4. Pathogenesis

Transmission of CPV-2 occurred by the fecal-oral route, after exposure to CPV-2 in feces and/or vomit, or viral-contaminated fomites (**Figure 3**). The severity of clinical symptoms depends on the factors such as viral strain, host immunity, and the presence of the coinfection with other pathogens. In naturally [60] or experimentally [61] infected dogs, many dogs never develop overt clinical symptoms, especially when the dogs have high level of the maternally derived antibodies (MDAs) [61]. Higher titers of MDA in young dogs are protective against infection by CPV-2. Infected dogs with severe clinical symptoms of disease had lower levels of hemagglutination inhibition (HI) titers than did animals without clinical signs following



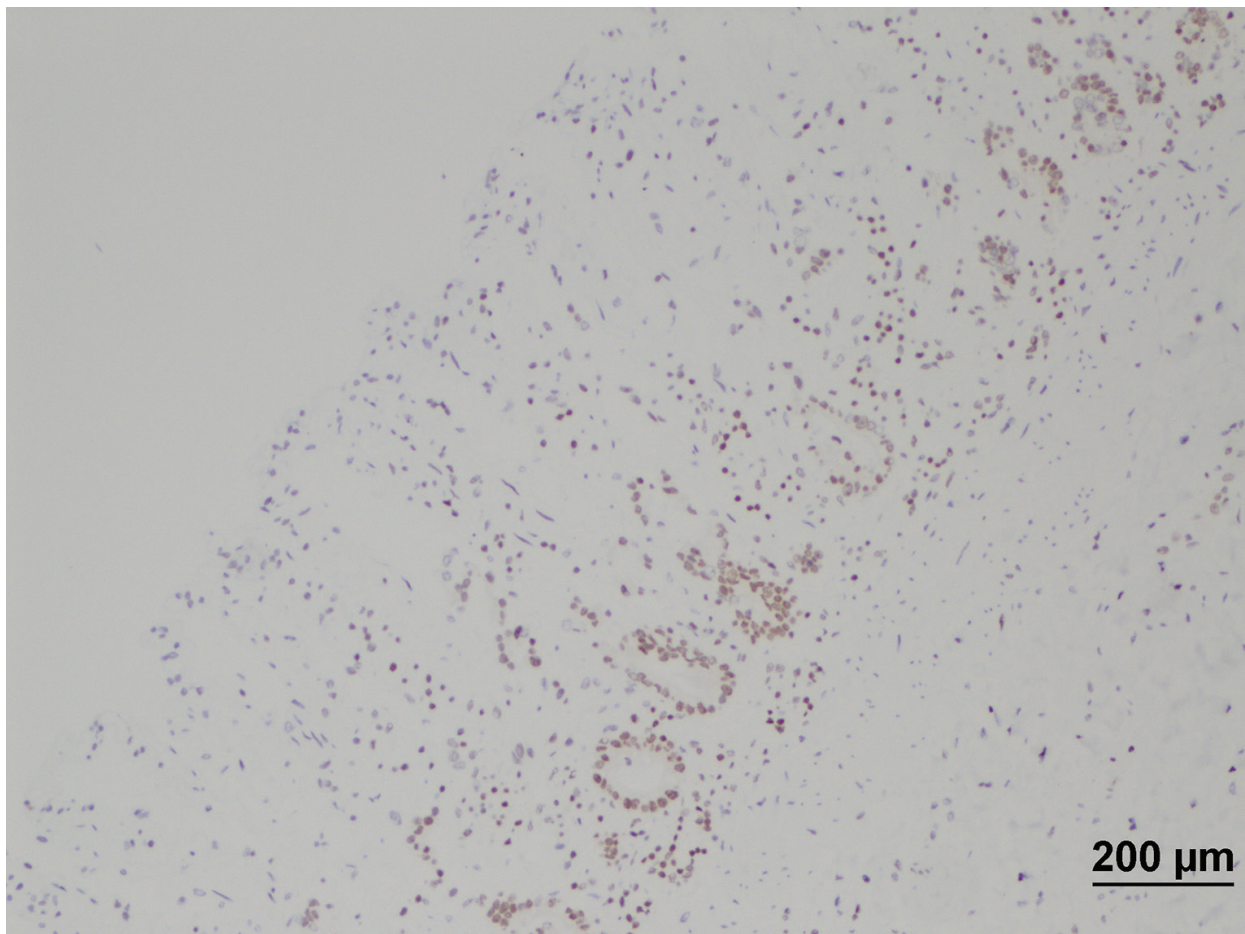
**Figure 4.** Mucosal hemorrhage in canine parvovirus type 2 (CPV-2)-infected dog. A 4-month-old puppy suffered from CPV-2 which showed lethargy, inappetence, and bloody diarrhea (from Professor Ming-Tang Chiou, Department of Veterinary Medicine, National Pingtung University of Science and Technology, Taiwan).



CPV-2 infection [61]. In the lowest HI titers group, CPV-2 shedding was noted up to 45 days post infection [61]. The incubation period of CPV-2 infection is 1–2 weeks. The affected gastrointestinal tissues include epithelium of the tongue, oral cavity, esophagus, and intestinal tract. CPV-2 replicates and destroys epithelial cells of the intestinal crypts causing malabsorption and increased intestinal permeability. CPV-2 shedding in feces was detected up to 45 and 54 days in experimentally [61] and naturally infected dogs [60], respectively. The period could also potentially correlate with the severity of the clinical status of the animals [62]. Viral shedding from infected dogs with bloody diarrhea was observed up to 63 days compared to 54 days in infected dogs with diarrhea (no dogs with hemorrhagic diarrhea or vomiting were tested) [60].

### 1.5. Clinical signs

The clinical signs of CPV-2 infection include fever, lethargy, inappetence, vomiting, diarrhea or bloody diarrhea (**Figure 4**), and dehydration. Myocarditis usually occurs up to the first 4 weeks of age or infected in utero, which may result in signs of sudden death



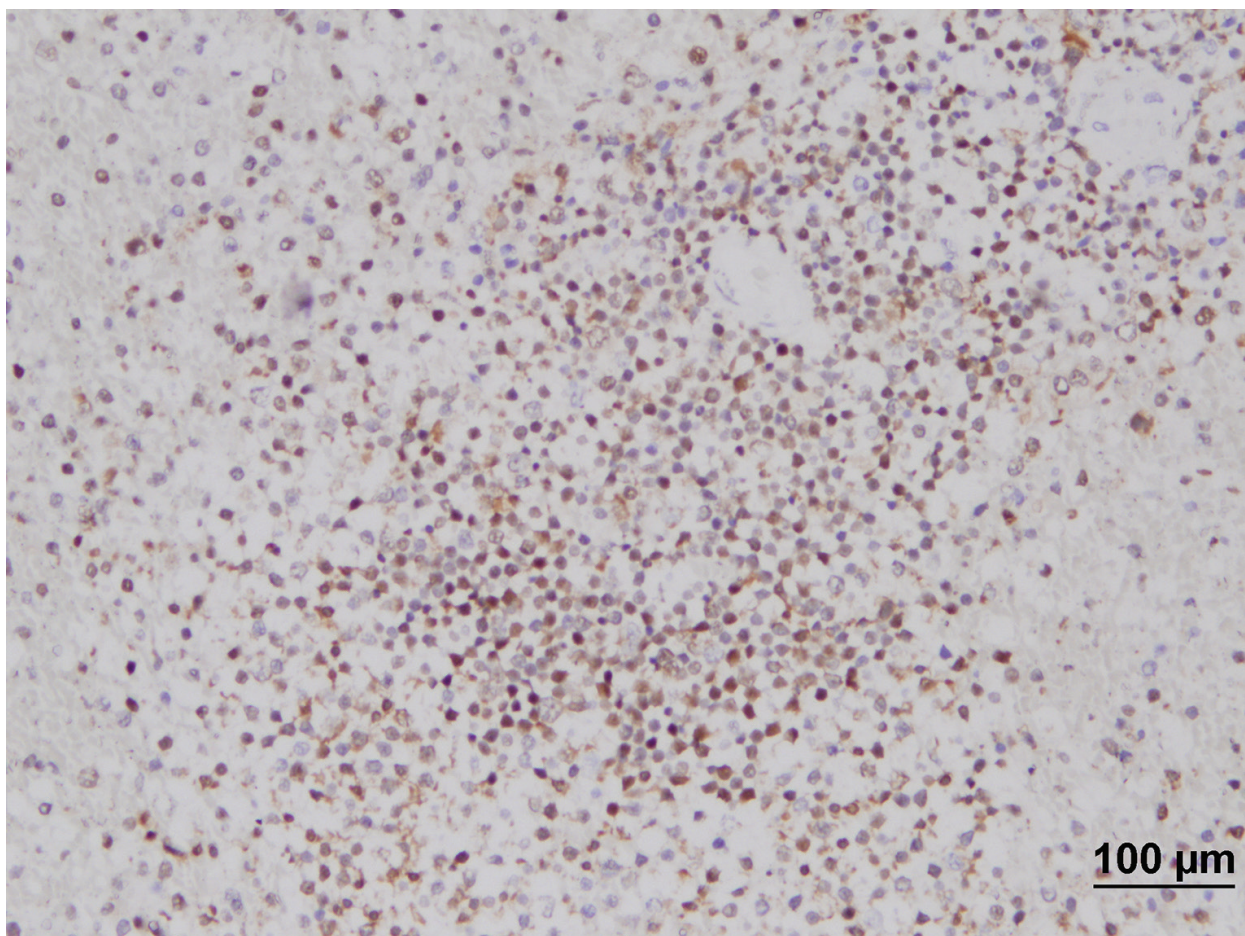
**Figure 5.** Detection of canine parvovirus type 2 (CPV-2) antigens in intestinal tract using in situ hybridization. CPV-2 immunolabeling is seen in the crypt epithelium (from Professor Ming-Tang Chiou, Department of Veterinary Medicine, National Pingtung University of Science and Technology, Taiwan).



or congestive heart failure. Cerebellar hypoplasia has been rarely reported in dogs with utero infection. CPV-2c shows almost the same clinical signs as CPV-2a and CPV-2b, such as anorexia, vomiting, acute gastroenteritis, and hemorrhagic diarrhea. However, CPV-2c infection has been reported to be indicative of a more severe disease induced by this variant [51, 63].

### 1.6. Diagnosis

The most common abnormalities found on the CBC are leukopenia, lymphopenia, and neutropenia. Some disease dogs develop anemia as a result of gastrointestinal blood loss. Electrolyte and coagulation abnormalities have been reported in dogs with parvoviral enteritis. The diagnosis of CPV-2 infection has relied on probe-based real-time polymerase chain reaction (PCR) [64–69], SYBR green-based real-time PCR [62, 70–72], conventional PCR [47, 68], electron microscopy [73], and methods provided as commercial kits [74]. CPV-2 antigens were found in crypt epithelium (**Figure 5**) or affected tissues such as spleen (**Figure 6**) using in situ hybridization.



**Figure 6.** Detection of canine parvovirus type 2 (CPV-2) antigens in spleen using in situ hybridization. CPV-2 immunolabeling is seen in lymphocyte (from Professor Ming-Tang Chiou, Department of Veterinary Medicine, National Pingtung University of Science and Technology, Taiwan).

## 1.7. Vaccination and prevention

Immunization is the most effective method for the prevention of CPV-2 enteritis. However, the initial immunization should be of concern regarding the interference of the MDA. The second concern is whether the currently available CPV-2 vaccine provides adequate protection against CPV-2c infection. VP2 encodes a viral capsid protein that is the major structural protein of CPV-2 and is involved in the host-immune response [75]. Therefore, a small number of mutations may result in increased pathogenicity [51]. Several studies have demonstrated the efficacy of the current CPV-2 vaccine against CPV-2c infection [76, 77]. By contrast, some evidence suggests that dogs with the complete vaccination program still suffer from CPV-2c [63]. Therefore, the efficacy of the current vaccine against prototype CPV-2c and/or novel CPV-2c variant remains to be evaluated, especially in regard to the amino acid substitutions observed in the novel CPV-2c variant as compared to the prototype of CPV-2c. Parvoviruses are extremely stable in the environment and can be transmitted via indirect contact, an important factor in their maintenance in populations. Several disinfectants had been reported that parvoviruses can be inactivated with a 1:30 dilution of household bleach, potassium peroxymonosulfate, and accelerated hydrogen peroxide. These disinfectants will also inactivate other viruses.

## 2. Conclusion

CPV-2 is most likely one of the most common infectious diseases in younger dogs. CPV-2 is constantly mutating, leading to the evolution of novel variants. All countries should continue surveillance and monitor the events associated with CPV-2 disease. The need to update current vaccines remains to be assessed.

## Author details

Chao-Nan Lin\* and Shu-Yun Chiang

\*Address all correspondence to: [cnlin6@mail.npust.edu.tw](mailto:cnlin6@mail.npust.edu.tw)

Department of Veterinary Medicine, College of Veterinary Medicine, National Pingtung University of Science and Technology, Pingtung, Taiwan, ROC

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