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Effect of Filgrastim in a Severe Leucopenia associated Parvoviral Enteritis in Rottweiler

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A B S T R A C T

Filgrastim (Colstim®) is a recombinant Methionyl Human Granulocyte Colony-Stimulating Factor (r-metHuG-CSF) analog used to stimulate the proliferation and differentiation of granulocytes in humans undergoing chemotherapy with granulocyte values below 0.5 G/l likely to be threatened by infections, including sepsis possibly with a fatal outcome. A four month old female Rottweiler pup was presented to the TVCC, RIVER with the history of vomiting and melena for the past two days. Further enquiry revealed that the pup had properly been immunized against all infectious diseases. Clinical examination showed a pink and moist conjunctival mucosa, rectal temperature of 38.7º C, hear rate 150 bpm, and capillary refill time of 3 seconds. The ECG showed a normal study. Laboratory examination revealed Hb - 15.6 g %, PCV - 45.5%, RBC count - 5.87 millions/mm^3, WBC count - 1000 cells/ mm^3, platelets - 4.21 lakhs/ mm^3, MCV - 77.6 fl, MCH - 26.5 pg, MCHC - 34.2%. Absolute count of Neutrophils - 500 cells/ mm^3, Lymphocytes - 450 cells/ mm^3, Monocytes - 40 cells / mm^3 and Eosinophils - 10 cells/ mm^3. However, serum biochemistry showed normal values but with an increase in Creatine kinase of 567 U/L. The dog was treated with Filgrastim @ 10 µg/ kg b.wt. S.c along with other supportives. After 5 days of therapy the dog recovered uneventfully.

Introduction

Canine Parvovirus (CPV) emerged as a new pandemic disease in dogs with first report from United States in 1978. In subsequent years it was widely reported from Canada, Australia, England, Asia and European countries. In India, the disease was first reported in Madras in 1981 by Balu and Thangaraj. Parvovirus infection in dogs, especially in the young ones, may result in a severe and fatal disease (Greene, 1984). Glickman *et al*., (1985), Greene and Decaro (2012) claimed that Doberman Pincher, Rottweiler, Labrador retriever, German shepherd, Alaskan sled dog, American Staffordshire terriers and English springer spaniels were significantly susceptible to CPV enteritis. CPV spreads rapidly from dog to dog via oronasal exposure to contaminated faeces. CPV localizes predominantly in the gastrointestinal epithelium lining the tongue, oral and esophageal mucosae and small intestine and lymphoid tissue, such as thymus, lymph nodes and bone marrow. It can also be isolated from the lungs spleen, liver, kidney

and myocardium (Greene, 1984). CPV also destroys mitotically active precursors of circulating leukocytes and lymphoid cells. In severe infections, the results are often neutropenia and lymphopaenia.

Filgrastim (Colstim®) is a recombinant Methionyl Human Granulocyte Colony-Stimulating Factor (r-metHuG-CSF) analog used to stimulate the proliferation and differentiation of granulocytes in humans undergoing chemotherapy with granulocyte values below 0.5 G/l likely to be threatened by infections, including sepsis possibly with a fatal outcome (Novotny *et al*., 1995). Filgrastim is a non-glycosylated, 175 amino acid containing protein, which is produced recombinantly by *E. coli.* Filgrastim has a molecular weight of 18.8 kDa. It regulates the production and release of functional neutrophils from bone marrow within 24 hours of administration. Filgrastim results in increase in peripheral blood neutrophil counts with minor increases in monocytes.

Materials and Methods

A four month old female Rottweiler pup was presented to the TVCC, RIVER with the history of vomiting and melena for the past two days. Further enquiry revealed that the pup had properly been immunized against all infectious diseases.

Clinical Assessment

Clinical examination showed a pink and moist conjunctival mucosa, rectal temperature of 38.7º C, heart rate 150 bpm, and capillary refill time of 3 seconds (Fig. 1). The ECG showed a normal study (Fig. 2) (Table 1).

Laboratory Assessment

The following table 2 shows the hematological values of CPV affected pup on Day I, II and III respectively and table 3 shows the serum biochemical values.

Diagnosis

The faecal sample was collected for the CPV diagnosis by Polymerase Chain Reaction (PCR) and it was positive (Fig. 3) by using primer pair H_{for} / H_{rev} that amplify a large fragment of the capsid protein-encoding gene (VP2) of CPV-2 (Buonavoglia *et al*., 2001). Primer H_{for} (5' CAG GTG ATG AAT TTG CTA CA3') and H_{rev} (5' CAT TTG GAT AAA CTG GTG GT 3'), located at nucleotide position 3556-3575 and 4166-4185 of the CPV genome respectively yield 630bp product.

Sequence Analysis

For the sequencing PCR products were purified and sequenced from Eurofins Genomics India Pvt. Ltd., Bengaluru, Karnataka. It was analyzed and compared with the available CPV sequences in the gene bank using NCBI BLAST http://blast.ncbi.nlm.nih.gov/blast.cgi (Altschul *et al*., 1997). It had 100% identity with the corresponding Canine parvovirus 2a isolate OB4 VP2 capsid protein gene, partial sequence (Gene bank accession number JX683402.1)

Clinical Management and Outcome

The dog was treated with Inj. Filgrastim @ 10 µg / kg b.wt. subcutaneously (Morris and Dobson, 2001) along with supportive therapy includes table 4.

Results and Discussion

In CPV infected pups death can occur at any age, depending on when maternally derived antibody (MDA) wanes.

Table.1 Clinical assessment

(Table 1) P wave		QRS complex		PR interval (sec)	ST segment	QT interval (sec)	T wave Amplitude (mV)	T wave Amplitude
Amplitude (mV)	Duration (sec)	Amplitude (mV)	Duration (sec)					
0.15	0.04	1.2	0.04	0.08	Elevated	0.12	0.2	Positive

Table.2 Hematological values of CPV

PARAMETER	Day I	Day II*	Day III**	Normal value
Hemoglobin (g/dL)	15.6	13.2	11	12-18
PCV (%)	45.5	40.1	32.3	37-55
RBC (million/mm^3)	5.87	4.9	4.43	5.5-8.5
WBC (cells /µL)	**1000**	**5800**	**7800**	6000-17000
Platelets (lakhs/µl)	4.21	4.20	4.42	2.0- 5.0
Neutrophils (cells/ mm^3)	500	4118	7332	3000 – 11,500
Lymphocytes (cells/ mm^3)	450	1624	312	1000 – 4800
Monocytes (cells/ mm^3)	40	58	78	150 – 1350
Eosinophils(cells/ mm^3)	10	-	78	100 – 1250
MCV (fL)	77.6	81.8	73.1	60 – 77
MCH (pg)	26.5	26.9	24.8	21 – 32
MCHC (%)	34.2	32.9	34.0	32 – 36

(* Day II - After 24 hours of Filgrastim Injection
** Day III - after the recovery from the disease)

Table.3 Serum biochemical values

Parameter	Observed Values	Normal Value
BUN (mg/dl)	12	12-25
Creatinine (mg/dl)	0.6	0.5-1.5
Protein Total (g/dl)	5.3	5.2-8.2
Albumin (g/dl)	2.6	2.3-4.0
Globulin (g/dl)	2.7	2.5-4.5
Potassium (Mmol/L)	4.6	3.8-5.8
Glucose (mg/dL)	105	74-143
ALP U/L	81	23-212
CK U/L	567	10-200

Table.4 Supportive therapy

Drug	Dose (per Kg B.wt)	Dosage
Inj. Lactate ringer solution	10ml	BID
Inj. Dextrose 5%	10ml	BID
Inj. Metronidazole	25mg	BID
Inj. Astymin-3	1ml	BID
Inj. Intralipid®	1 ml	BID
Inj. Ondansetron	0.1 mg	BID
Inj. Ranitidine	2 mg	BID
Inj. Cefotaxime	40 mg	BID

After 5 days of therapy the dog recovered uneventfully.

Fig.1 Shows the affected animal

Fig.2 Electrocardiogram of Parvovirus infected dog

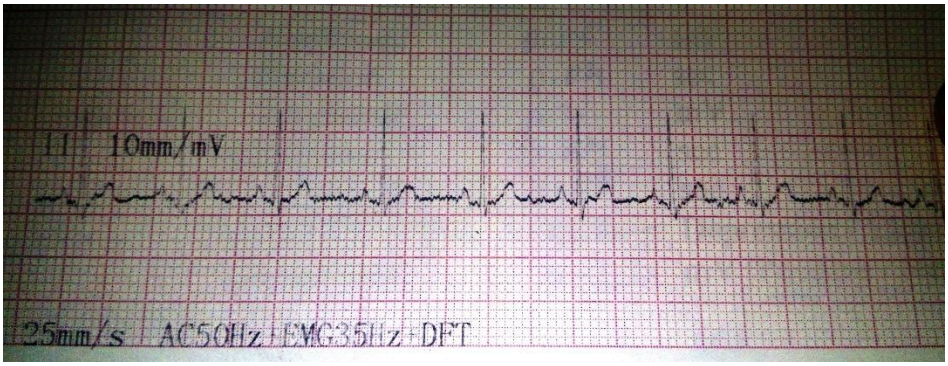
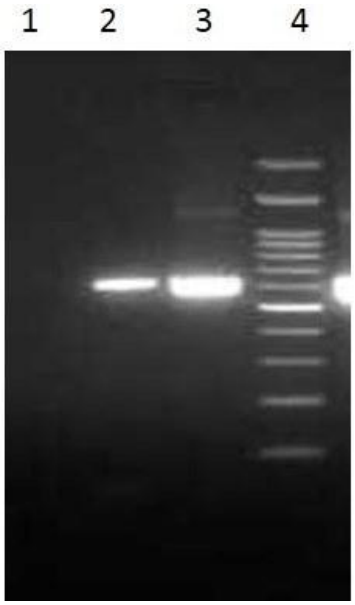


Fig.3 1. Negative control, 2. Sample, 3. Positive control (vaccine), 4. DNA marker (100bp)



It causes acute hemorrhagic gastroenteritis in dogs and is prone to genetic evolution mainly due to the mutations in VP2 gene (Mohan *et al*., 2010). In this case, although the dog vaccinated properly, occurrence of the disease noticed and the leukocytes count (1000 cells /µL) was very low. To overcome this Inj. Filgrastim at the dose rate of 10 µg/kg body weight (dose rate 5-15 µg/kg) was administered. After 24 hours of the injection the leucocyte count was elevated to 5800 cells /µL. Greene and Decaro (2012) reported that those pups dying from the disease generally have TLC equal to or less than 1030 cells/µl and have persistent lymphocytopenia, monocytopenia and eosinopenia within the first 3 days of hospitalization. Even though Rottweiler is a predisposing breed to CPV (Glickman *et al*., 1985), the administration of Filgrastim will improve the survival rate if it is administered at the early stage of the disease along with other supportive therapy.

References

Altschul, S.F., Madden, T.L., Schaffer, A.A., Zhang, J., Zhang, Z., Miller, W. and Lipman, D.J. 1997. Gapped BLAST and PSI-BLAST: a new generation of protein data-base search programs. *Nucleic Acids Res.*, 25: 3389-3402.

Balu, P.A. and T.M. Thangaraj. 1981. Canine viral gastroenteritis – A clinical report. *Indian J. Vet Med.*, 1: 73.

Buonavoglia, C., Martella, V., Pratelli, A., Tempesta, M., Cavalli, A. and Buonavoglia, D. 2001. Evidence for evolution of canine parvovirus type-2 in Italy. *J. Gen. Virol*., 82: 3021–3025.

Glickman, L.T., L.M. Domanski., G.J. Patronek and F. Vistainer. 1985. Breed related risk factors for canine parvovirus enteritis. *J. Am. Vet. Med. Assoc.*, 187: 589-594.

Greene, C.E. 1984. Canine viral enteritis. Infectious diseases of the dog and cat. Philadelphia, W.B. Saunders, pp. 220-37.

Greene, C.E. and Decaro, N. 2012. Canine viral enteritis. In: Greene CE, editor. Infectious diseases of the dog and cat 4th ed. St. Louis: Elsevier; pp. 67-80.

Mohan Raj, J., Mukhopadhyay, H.K., Thanislass, J., Antony, P.X. and Pillai R.M. 2010. Isolation, molecular

characterization and phylogenetic analysis of Canine parvovirus. *Infect. Genet. Evol.*, 10(8): 1237-1241.

Morris, J. and Dobson. J. 2001. Small animal oncology, Black well sci. ltd. Pp 288

Novotny, J., M. Zvarova, L. Prazakova, M. Jandlova and L. Konvickova. 1995. G-CSF in the treatment of patients with chronic aplastic anemia with severe neutropenia, *Vnitr Lek.*, 41(10): 692-5.

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