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Research Article

Evaluation of Faecal Microbiota Transplantation as Adjunctive Therapy in Parvo Viral Enteritis in Dogs

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Abstract

The present study was aimed to evaluate the therapeutic role of faecal microbiota transplantation in dogs affected with parvoviral enteritis. Group II dogs were treated with standard therapy, whereas Group III dogs received faecal microbiota transplantation (FMT) in addition to the standard therapy adopted in Group II dogs. Among the hemogram, recorded significant increase ($p < 0.05$) in mean values of Hb (g/dl) and PCV (%) in Group III, which were increased non-significantly ($p > 0.05$) in Group II. The pre and post therapeutic serum biochemical parameters revealed significant improvement ($p < 0.05$) in SGPT (IU/L), albumin (g/dl) and potassium (mmol/l) in both Group II and Group III. The recovery pathway revealed that two dogs (18.18%) were completely recovered by 96 hours and eight dogs (72.73%) by 120 hours in Group II whereas seven dogs (63.64%) in Group III recovered by 72 hours and nine dogs by 96 hours (81.82%). The survival percentage in Group II and Group III was 72.75% and 81.82% respectively with the average duration of recovery as 4.75 ± 0.05 and 3.22 ± 0.05 days. The pre and post therapeutic clinical scores before and 120 hours after therapy in Group II dogs were 14.91 ± 0.46 vs 1.38 ± 0.26, while the corresponding scores in Group III were15.36 ± 0.34 vs 0.11 ± 0.11, suggesting significant clinical and quick response was noticed in dogs received FMT following therapy.

Keywords: Canine Parvo Viral (CPV); Faecal Microbiota Transplantation (FMT)

Introduction

Canine parvo viral (CPV) enteritis is a highly contagious, virulent, acute and fatal gastrointestinal viral disease that targets primarily the pups and collapses the body's posthaste branching organs causing bloody diarrhoea amidst a high mortality and morbidity rate [1]. It is one of the common causes of morbidity and mortality in young dogs globally. CPV belongs to the genus protoparvo virus, family parvoviridae, a single- stranded DNA virus that infects rapidly dividing cells of the gastrointestinal tract, bone marrow, lymphoid tissue and cardiac myocytes [2]. CPV dogs witness dysbiosis in the gut microflora that in turn impairs enterocyte nutrition, immunological modulation, gastrointestinal motility and protective barrier function [3]. Faecal microbiota transplantation (FMT) is a lesser explored procedure which involves introducing a homogenised liquid suspension of faeces from a healthy donor into the patient's gastrointestinal (GI) tract to restore the normal gut microbiota.

Materials and Methods

The dogs presented to the Small Animal Medicine Ward, Veterinary Clinical Complex, NTR College of Veterinary Science,

Gannavaram from January 2024 to December 2024 with the symptoms of foul- smelling diarrhoea, dehydration, vomiting, inappetance/anorexia and congested mucous membranes were selected and screened with rapid immunichromatigraphic test kit (BioNote®) for parvoviral enteritis. Dogs which had parasitic infection and mixed etiology were excluded from the current study. Dogs infected with CPV were randomly allocated into two groups and were subjected to therapeutic trials.

Therapeutic trials

A total of twenty-two dogs affected with canine parvoviral enteritis (confirmed with rapid immunochromatographic test kit) were selected and subjected to therapeutic trial. The dogs were randomly divided into two groups i.e., Group II and Group III each comprising of eleven animals and treated with two different therapeutic regimens. All the dogs were treated with Inj. Amoxicillin-Sulbactum @ 20 mg/kg bid, iv along with inj. ondansetron @ 0.2 mg/kg, iv or im,tid, inj.pantaprazole @ 1 mg/kg iv sid for 3 days, inj.tranexamic acid @25 mg/kg, iv bid and supportive fluid therapy (RL, DNS and vetplasma®). The dosages of antiemetic, proton pump inhibitor, tranexamic acid and fluid therapy were modified according to the clinical condition. All the dogs were observed at 12-24 hour intervals till the resolution of clinical symptoms and haemato-biochemical alterations were documented before and after therapy.

In addition to symptomatic and supportive therapy, dietary recommendations were made for all the affected dogs. The recommendations included withholding food and water for at least 24 hours or until the cessation of the emesis (in severe cases) followed by introduction of easily digestible diets in small increments.

Dogs in Group II were treated with antibiotic, symptomatic and supportive therapy including IV fluids, whereas Group III dogs were treated with antibiotic, symptomatic, supportive therapy and faecal transplantation as detailed below.

Faecal microbiota transplantation

Donor selection

A healthy donor was selected according to the following criteria as outlined by Pereira., *et al*. [4]. The faecal donor for the procedure was a client owned, intact female Labrador dog aged 3.5 years which had no prior history of gastrointestinal disease and absence of use of antibiotics for the past two years and fed on home-made diet with daal, rice, eggs, chicken and curd. The donor dog was promptly vaccinated and dewormed. The animal was naturally born and colostrum fed. The animal was a having a normal body condition score and faecal score. Haemato-biochemical parameters of the donor were analysed to rule out any latent diseases.

The donor faeces were examined for parasites, enteropathogens and a normal microbiota using culture procedures. Stool was collected daily basis in the early hours into a sterile plastic vial and was immediately processed in the laboratory within 2 hours of collection.

Diagnostic assessment of donor

A Complete blood count, serum biochemical profile and faecal tests were performed. Furthermore, Rapid antigen tests for canine parvovirus, distemper virus, and tests for haemoprotozoan were carried out.

Selection of recipient

Recipients or CPV positive dogs were chosen regardless of age, sex and breed. Single FMT was carried out in CPV dogs on the day of presentation (day 0).

Processing of the faecal suspension

Freshly evacuated faeces from donor were collected in the morning and processed within two hours and the suspension was stored at -20 ◦ C for further use on the same day. About 10g of freshly collected faeces were mixed with 10 ml of 0.9 per cent sodium chloride solution and then homogenously blended for approximately 5 minutes, sample was strained with a strainer to achieve a homogeneous mixture devoid of solids. The suspension was then loaded in 20 ml syringes attached with a red rubber catheter and stored at -20 ◦ C. To administer the enema, a small amount of lubricant was applied to the catheter and the catheter was completely introduced in the proximal segment of colon (Figure 1-3).

Procedure of faecal transplantation

The lower abdomen of recipient dogs was lifted at about 45° angle, the suspension via a syringe linked to a catheter was administered per-rectal and the contents were deposited in the

Figure 1: Donor dog used for FMT.

Figure 2: Fresh faecal sample collected form donor.

Figure 3: Diluted and filtered faeces.

proximal segment of the colon. The animal was kept stable during subsequent 30 minutes thereafter. The procedure was executed after initiation of the symptomatic therapy, on day 0 (day of presentation) only (Figure 4). The pet owners were contacted via telephone during clinical trials to report any undesirable effects and any recurrence of gastroenteritis, chiefly diarrhoea.

Figure 4: FMT to CPV affected dog.

Clinical scoring of various parameters of group II and III

The efficacy of therapy was evaluated based on clinical assessment (resolution of clinical signs and clinical scoring) and improvement in haemato-biochemical parameters.

Clinical scoring

The dogs of Group II and Group III were allotted a clinical score [5] at each visit from the day of presentation to recovery.

Statistical analysis

The data obtained were subjected to statistical analysis by using computer software SPSS 29. The mean values of different parameters between control and the affected group before and after therapy were compared at one per cent and five per cent level of significance using t- test.

Results and Discussion

Therapeutic studies were undertaken in 22 dogs diagnosed as canine parvovirus infection and were randomly allocated into two

Parameter	0	1	2	3
Activity	Active	Mildly lethargic	Moderately lethargic	Lateral recumbency and comatose
Appetite	Normal diet	Able to digest semi-solid food	Able to digest bland liquid diet	Complete anorexia with polydipsia
Vomiting (frequency)	0	1-2	3-5	>5
Faecal consistency	Solid	Semisolid	Watery	Watery/bloody
Defaecation (frequency/ day)	1	2-3	4-5	>5
Dehydration (%)	<5	5-8	8-10	>12
Total clinical score	0-3	4-5	6-8	9 or more
Clinical significance	Clinically insignificant	mild	moderate	severe

Table 1: Scoring criteria of affected dogs according to various parameters [5].

groups, each comprising of eleven dogs. Dogs of Group II received standard medical therapy, whereas Group III dogs were treated with faecal microbiota transplantation in addition to the treatment adopted in Group II dogs. All the dogs in both groups were treated with analogous symptomatic (antiemetic, proton pump inhibitor and tranexamic acid) and supportive medication (Ringer's Lactate and colloids) in conjunction with Amoxicillin-Sulbactam. The doses of antiemetics, tranexamic acid, and hydration therapy were modified according to the requirement and adjusted based on clinical recovery. Venn., *et al*. [6], Rafiq., *et al*. [7] and Lomiya., *et al*. [8] recommended the administration of antibiotics, anti-emetics, proton pump inhibitors and vigorous fluid therapy (crystalloids and colloids) for the treatment of canine parvoviral enteritis, which enhanced survival rates and facilitated quick recovery.

The efficacy of treatment was evaluated by the alleviation of clinical symptoms foul-smelling diarrhoea, emesis and dehydration that were often observed in all the dogs enrolled in the treatment by monitoring vital signs and based on recovery change in haematological and biochemical parameters.

Group II dogs treated with standard medical therapy (SMT) exhibited vomiting 6-8 times and foul-smelling, blood-tinged diarrhoea 7-9 times along with anorexia, lethargy and moderate dehydration on the day of presentation. Upon therapy the vomiting frequency was reduced to 5-6(24 h), 2-4(48 h), 1-3(72 h), 0-2(96 h) times and completely subsided in all dogs by 120 hours. Whereas the frequency of diarrhoea decreased from 4-7 times at 24 hours, 3-5 at 48 hours, 0-2 at 72 hours and resolved completely by 96 hours.

Similarly, the frequency of vomiting and foul -smelling diarrhoea was 6-9 and 7-9 respectively in Group III dogs (FMT) with the signs of anorexia, depression and moderate dehydration. Upon therapy with faecal microbial transplant in-addition to the standard medical therapy, the vomiting frequency was reduced to 0-1(24h) and completely subsided in all dogs by 48 hours. While episodes of diarrhoea were diminished with improvement in faecal constancy to 0-2(24h), 0-1(48h) and had resolution of diarrhoea by 72 hours. Following therapy, gradual improvement in the frequency of vomiting and diarrhoea (3-4 days) with complete resolution by 4-5 days in dogs of Group II, while the same resolved quickly by 1-2 days completely with amelioration of symptoms by 2-3 days in Group III dogs. These findings were in agreement with the observations of Kalita., *et al*. [9] who reported quick resolution of diarrhoea in dogs treated with FMT in comparison to symptomatic therapy alone (48 hours vs 96 hours). The advantages and rapid resolution of symptoms in FMT treated dogs were also reported in previous studies [4,10,11]. Response to the FMT in CPV enteritis could be due to reconstitution of the intestinal microbiota and its corresponding metabolites [12]. On the contrary, Heilman., *et al*. [13] reported perpetuation of diarrhoea in CPV infected dogs treated with faecal microbial transplantation due to bacterial dysbiosis caused by inflammation, hypersecretion and hypermotility and changes in osmolality and PH.

The CPV affected dogs in Group II and III exhibited dehydration varying from five per cent to eight per cent. However, the hydration status was improved by 72 hours in Group II and by 48 hours in Group III. Similarly, the appetite was restored completely by 72 hours of therapy in Group III while the same took 120 hours in Group II and restoration of appetite was quicker in Group III dogs treated with FMT+SMT than Group II dogs those received SMT alone. Similar findings were reported by Sruthi and Ajay [14] who documented that all the survived animals from canine parvoviral enteritis regained their normal appetite in 5-7 days of initiation of treatment and were active, alert and bright in appearance.

S. No	Clinical signs	Group II						Group III					
		BT	24H	48 H	72H	96 H	120H	BT	24H	48H	72 H	96 H	120H
1	Vomiting	6-8	5-6	2-4	1-3	0-2	_	6-9	0-1	_	_	_	_
2	Foul smelling blood- tinged diarrhoea	7-9	4-7	3-5	0-2	Nil	Nil	7-9	0-2	0-1	_	_	_
3	Dehydration	Improved by 72 hours						Improved by 48 hours					
4	Appetite	Improved by 5-7 days						Improved by 3 days					
5	Number of dogs died	_	_	3	_	_	_	_	_	2	_	_	_

Table 2: Resolution of clinical signs in Group II and Group III dogs during therapy.

Dogs of both the groups had significant rise in mean ± SE values of temperature, heart rate and respiratory rate prior to therapy, which were significantly reduced ($p < 0.05$) following therapy. Similar results were reported by Bastan., *et al*. [15] and Patel., *et al*. [16] who opined that these changes might be due to the effect of catecholamine and other compensatory mechanisms in the vital organs during the course of the disease and were returned to the normalcy as a response to therapy.

Significant reduction in haemoglobin, PCV, TLC, TEC, neutrophils, lymphocytes and monocyte were recorded in affected dogs when compared to healthy. After therapy the Group II dogs manifested a non-significant elevation in the mean values of Hb, PCV, monocytes and eosinophils, whereas significant increase was observed in the mean values of total erythrocyte count, TLC, lymphocytes and significant decrease in mean values of neutrophils. Cases of Group III showed significant increase in the mean values of haemoglobin, PCV, total erythrocyte count, TLC and lymphocytes and a significant decrease in the mean values of neutrophils following therapy. No significant difference was seen in the mean values of eosinophils and monocytes before and after therapy. Though, the dogs in both groups showed an overall improvement in all the haematological parameters by the end of therapy, significant elevation in haemoglobin and PCV was recorded in group III dogs. These findings gained support by Shruti and Ajay, [14] who reported significant alteration in haematological parameters after therapy. On the contrary Patel., *et al*. [16] documented non-significant difference in the mean values of Hb, RBC, WBC, lymphocytes, monocytes, eosinophils, neutrophils, basophils and PCV in CPV affected dogs after therapy. However, improvement in haematological parameter in both the groups in the present study suggested the both the treatments had positive affect on haematological parameters.

The serum biochemical parameters in Group II dogs showed significant decrease in mean SGPT levels but remained within reference range, whereas the mean values of albumin, potassium was increased significantly following therapy. But non-significant difference in the mean levels of total protein, sodium and chloride was observed after therapy. In the present study significant

improvement was recorded in serum SGPT (IU/L) albumin (g/dL) and potassium (mmol/L) concentrations in Group II dogs after therapy while insignificant variation was recorded in serum total protein (g/dL), sodium(mmol/L) and chloride concentrate (g/dL).

Similarly, Group III dogs showed significant reduction in mean SGPT levels within the reference limits and significant increase in the mean values of albumin, potassium, post therapeutically. But non-significant difference in the mean levels of total protein, sodium and chloride was observed after therapy.

Correction of dehydration, electrolyte imbalance and use of antibiotics might have resulted in improvement in serum biochemical parameters. Irrespective of therapeutic regimen administration of intravenous fluid therapy was also recommended by previous researchers [17].

The alleviation of clinical symptoms and improvement of haemato-biochemical parameter in both the groups, indicate that the implemented therapy regimen was effective in treating canine parvoviral enteritis in dogs. These findings align with those of Haritha., *et al*. [18] and Lomiya., *et al*. [8] who documented the efficacy of antibiotics in treating canine parvoviral infection.

The observed improvement in clinical signs and hematobiochemical parameters in FMT treated dogs suggests that FMT therapy might play a beneficial role as on adjunct therapy in the management of CPV infection. The aforementioned findings align with those of Pereira., *et al*. [4], who emphasized the efficacy of faecal transplantation along with standard treatment in CPV affected dogs. Thus, serving as an alternative approach and facilitated a rapid resolution of diarrhoea and reduced duration of hospitalization.

Survival of eight (72.75%) dogs in Group II and nine (81.82%) in Group III suggested that FMT might be beneficial in reducing mortality in dogs affected by CPV. These findings were in agreement with the observations of Pereira., *et al*. [4] recorded highest survival rate (78.78%) in dogs treated with FMT than standard medical therapy (63.63%). The outcome of therapy gained support by the reports of Burton., *et al*. [19] who concluded that FMT was a safe therapeutic option without any adverse side effects in dogs with CPV infection. In addition to the lower death rate Group III animals showed earlier clinical recovery with the average duration of therapy as 4.75 ± 0.05 in Group II and 3.22 ± 0.05 dogs in Group III.

Group	Total dogs treated	Number of dogs recovered			Total died	Number of dogs survived	Average number of days taken for recovery
		72 hr	96hr	120hr			
		Number	Number	Number			
II	11	0	2(18.18%)	8(72.73%)	3(27.25%)	8(72.75%)	4.75 ± 0.05
III	11	7(63.64%)	9(81.82%)	0	2(18.18%)	9(81.82)	3.22 ± 0.05

Table 3: Recovery pathway in Group II and Group III dogs affected with canine parvoviral enteritis (Mean ± SE).

Canine parvoviral enteritis modifies the microbiota in canine intestines; hence, restoring or augmenting the original microbiota is a successful strategy with higher success rate, lower costs and shorter inpatient periods [20]. Restoring gut microbiota in dogs with CPV enteritis by supplementing FMT helped to rebuild the intestinal barrier, to reduce inflammation and diarrhoea, to boost immune defence and supported faster recovery in the present study.

Mean ± SE of the clinical scores before therapy in Group II and Group III dogs were 14.91 ± 0.46 and 15.36 ± 0.34 while the same were 1.38 ± 0.26 and 0.11 ± 0.11 respectively after therapy. Thought significant improvement in Mean ± SE of the clinical scores were observed in both the groups after therapy the recovery rate was faster in Group III compared to Group II. These findings align with the observations of Kalita., *et al*. [9] who documented that FMT treated dogs had better clinical scores than dogs treated with symptomatic therapy alone.

The gut microbiota contributed positively to the general health of the host through several functions *viz.* development and support of the immune system, production of metabolites

Days	Parameter	Activity	Appetite	Vomiting (frequency)	Faecal consistency	Defecation Frequency/ day	Dehydration (%)	Score
BT	Group II	2.18	3	2.82	3	2.82	1.09	14.91 ± 0.46
	Group III	2.36	3	2.91	3	2.91	1.18	15.36 ± 0.34
24H	Group II	2	3	2.82	2.73	2.18	0.82	13.64 ± 0.43
	Group III	1.09	3	1	1	1	0.82	7.82 ± 0.12
48H	Group II	1	3	1	2	1	0.73	8.73 ± 0.14
	Group III	0	0.55	0	0	0	0	0.55 ± 0.31
72H	Group II	1	2	1	2	0.75	0	6.75 ± 0.16
	Group III	0	0.56	0	0	0	0	0.56 ± 0.29
96H	Group II	1	1	0.63	1	0	0	3.63 ± 0.18
	Group III	0	0.22	0	0	0	0	0.22 ± 0.15
120H	Group II	0.25	1.13	0	0	0	0	1.38 ± 0.26
	Group III	0	0.11	0	0	0	0	0.11 ± 0.11

Table 4: Pre and post therapeutic clinical score in CPV affected dogs (Group II and III).

Parameter	Group II (Before therapy	Group II (After therapy)	Group III (Before therapy)	Group III (After therapy)
Clinical score	14.91 ± 0.46	1.38 ± 0.26*	15.36 ± 0.34	0.11 ± 0.11*

Table 5: Mean ± SE of Clinical score of pre and post therapeutic mean values of Group II and Group III dogs.

*: Significant at $p < 0.05$ between pre and post therapy values.

NS: Not significant ($P > 0.05$).

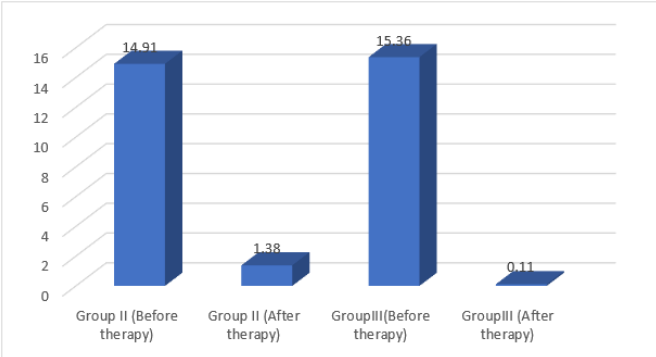

Figure 5: Mean ± SE of Clinical score of pre and post therapeutic mean values of Group II and Group III dogs.

with nutritional value or signalling function, maintenance of homeostasis (e.g., maintenance of the intestinal barrier) and resistance to colonization with pathogenic bacteria [21]**.** Among the survivors of CPV affected dogs with FMT correlated a more rapid cure of diarrhoea ($P < .001$) and reduced hospitalization duration ($P = .001$; median, 3 days for STD 1 FMT; median, 6 days for STD) in comparison to standard treatment. Further non-significant difference was noticed in mortality rate between STD group and FMT (36.4% 12/33 vs 21.2% 7/33) respectively [4]. Faecal microbiota transplantation (FMT) could restore impaired microbial flora and treat intestinal illnesses due to the colonization of commensal bacteria [22]. Faecal microbiota transplantation (FMT) was superior than metronidazole in the treatment of acute enteritis, particularly in restoring dysbiosis [23]. Faecal microbiota transplantation (FMT) offered benefits in the treatment of canine *Clostridium difficile* infection (CDI), inflammatory bowel disease

(IBD), canine parvovirus diarrhoea and acute diarrhoea, while also enhancing the intestinal microbiota in dogs with acute haemorrhagic diarrhoea syndrome (AHDS) [24]. Faecal microbiota transplantation should be regarded as an additional therapy for canine parvovirus enteritis, acute diarrhoea, and chronic enteropathy (Winston., *et al*. 2024). Parvo viral enteritis altered the microbiota in canine intestines; hence, restoring or enhancing the original microbiota was considered an effective approach, and patients who had STD+FMT had a more rapid resolution of diarrhoea offering superior success rate, reduced expenses, and shortened hospitalization duration [20]. The objectives of this study were to evaluate the efficacy of faecal transplantation as an adjunctive therapy in parvoviral enteritis in dogs.

The faster resolution in clinical scores and shorter duration of therapy observed in FMT treated dogs suggest that FMT could serve as an effective adjuvant therapy in dogs with CPV infection.

Conclusion

Present study showed that Faecal microbiota transplantation (FMT) serves as a valuable adjunctive treatment for dogs with canine parvovirus by accelerating the recovery and shortening recovery times. FMT helps rapid restoration of the gut with beneficial commensal microbes, restoring the gut barrier and immune balance in CPV affected dogs.

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