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Neha Gupta Post Graduate Scholar, Department of Veterinary Medicine, GBPUA&T, Pantnagar, Uttarakhand, India

VS Rajora

Professor, Department of Veterinary Medicine, GBPUA&T, Pantnagar, Uttarakhand, India

Kapil Kumar Gupta Ph.D., Scholar, IVRI, Izatnagar, Bareilly, Uttar Pradesh, India

Niddhi Arora

Associate Professor, Department of Veterinary Medicine, GBPUA&T, Pantnagar, Uttarakhand, India

Corresponding Author: Kapil Kumar Gupta Ph.D., Scholar, IVRI, Izatnagar, Bareilly, Uttar Pradesh, India

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## Evaluation of novel polyherbal formulation for the Clinico-therapeutic management of Diarrhea in calves

### Neha Gupta, VS Rajora, Kapil Kumar Gupta and Niddhi Arora

#### Abstract

In the present investigation therapeutic efficacy of polyherbal formulation were evaluated in diarrhoeic calves of upto 6 weeks of age of either sex at Instructional Dairy Farm (IDF) of the university. Polyherbal formulation (PHF) was prepared by mixing equal proportions of extract of dried fruits of Aegle marmelos, roots of Zingiber officinale, leaves of Dalbergia sissoo, seeds of Eugenia jambolana and dried fruits of Emblica officinalis. PHF mixture was fed to the diarrhoeic calves @ 10 gm, BID for 5 days. The collected faecal samples were examined for E. coli and Salmonella by culture and biochemical method and Toxocara and Eimeria by flotation method using saturated salt solution. The therapeutic efficacy of polyherbal formulation was evaluated in diarrhoeic calves based on clinical examination, clinical profiles, haematological study, biochemical analysis and percent recovery. Twelve, diarrhoeic calves selected for therapeutic studies were divided in 2 equal groups of 6 calves each. Six healthy calves served as control and kept in group I. The animals of group II were treated with tab Tariflox OZ (500 mg Ornidazole and 200 mg Ofloxacin) @ 10 mg/kg body weight twice a day orally along with intravenous fluid (Ringer lactate) according to dehydration status of calves, whereas group III was treated with PHF along with intravenous fluid. Both the treatments were given for a maximum period of 5 days in both the groups or till recovery which is earlier. All animals of both the groups were recovered after therapy of their respective group however the duration of therapy required for complete recovery is more in case of group III. Further it has been observed that therapy with polyherbal formulation along with intravenous fluid administration is more economic (Rs. 129.37 per calf) as compared to therapy applied in group II (Rs. 180.06 per calf). Evaluation of clinical profile score indicates fluid therapy along with antibiotic is more superior than fluid therapy with polyherbal formulation. On the basis of these findings it can be concluded that poor farmers should go for polyherbal therapy along with fluid administration in case of calf diarrhea however for the financially strong farmers antibiotic administration is not the limiting factor so they can adopt alternate antibiotic therapy with consideration of side effect and antimicrobial resistance.

Keywords: Diarrhoea, intravenous, polyherbal, ringer lactate

#### Introduction

Calf diarrhea is the most frequent problem in young animals and causes a huge loss of production throughout the world <sup>[1]</sup> and every practical effort should be made to minimize productive losses and mortality. In dairy cattle herds the prevalence and incidence risk for neonatal calf diarrhoea has recently been reported to be 19.1 and 21.2%, respectively <sup>[2]</sup>. Neonatal calf diarrhoea is chief cause of morbidity and mortality in young calves <sup>[3]</sup>. Calves suffer from various diseases in which death associated with diarrhoea vary from 2% to 20% <sup>[4]</sup>. Both Infectious and non-infectious factors can lead to severe diarrhoea in calves. Some noninfectious causes of diarrhoea include poisoning like molybdenum, stress conditions, imbalanced diet, and sudden change of feed, administration of large quantity of laxative and excess use of non-milk carbohydrates in milk replacer. The most important infectious enteric pathogens identified to cause diarrhoea in neonatal calves are enterotoxic Escherichia coli (ETEC), Clostridium perfringens type C, Cryptosporidium parvum, Bovine rotavirus (BRV) group A, Bovine coronavirus (BCV), Bovine viral diarrhoea (BVD) virus, and Salmonella spp., either single or in combination <sup>[5]</sup>. Among these organisms *Escherichia coli* is the most common and most important cause for the calf diarrhea as "white" scour <sup>[6]</sup>. Among the various serotypes, the most familiar cause of neonatal calf diarrhoea is enterotoxigenic E. coli (ETEC) <sup>[7]</sup>. The patho-physiology of diarrhoea is multifaceted in nature characterized by imbalance of fluid, electrolyte and acid-base status. Dehydration, anorexia and pyrexia are

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commonly noticed in diarrhoeic calves which results into decreased plasma volume and extracellular fluid volume, which in turn leads to decreased cardiac output, peripheral perfusion, and oxygen delivery (hypovolemic shock). Diarrhoea causes hypoglycemia <sup>[8]</sup>, hyperproteinaemia, hyperalbuminaemia [9] and electrolyte imbalance. Therapeutic intervention involves use of rehydration therapy in combination with antibiotics but indiscriminate and frequent use of antibiotics cause development of resistance in pathogen and also it is costly and their efficacy is controversial. Hence use of herbal remedy is another alternate approach for the management of diarrhoea. The earliest mention of diarrhoea is found in Veda, particularly in Atharva Veda. Herbal medication and poly-herbal formulations are being most popular as they are low cost and safe <sup>[10]</sup>. There is an everincreasing demand for herb-based therapeutics due to their easy availability, no side effect, affordable prices and no chances of resistance development. The present work is therefore, undertaken to evaluate the efficacy of polyherbal formulation in the treatment of calf diarrhoea.

#### **Material and Methods**

#### Preparation of polyherbal formulation (PHF):

Polyherbal formulation comprised of equal parts of dried roots of *Zingiber officinale* (carminative and antispasmodic), fruits of *Aegle marmelos* (astringent), leaves of *Dalbergia sissoo* (anti *E. coli*), seeds of *Eugenia jambolana* (anti E. coli) and fruits of *Emblica officinalis* (immune-potentiator) powder.

All ingredients, mentioned above, were collected and from nearby surroundings and sundried. Then these ingredients were grinded individually and mixed in equal proportions to make formulation. (Fig 1)



Fig 1: Herbs and their extracts

#### **Experimental design**

#### Selection and grouping of animals

Twelve (12) clinical cases (divided in 2 equal groups of 6 calves each) in crossbred calves of either sex of up to six weeks of age suffering from diarrhoea at Instructional Dairy Farm (IDF) of G. B. Pant University of Agriculture and

Technology, Pantnagar were used during present investigation. Six healthy calves served as control in group I. These calves were maintained under similar husbandry practices in separate cages and were fed whole milk @ 10% of their body weight, twice daily. The crossbred calves with diarrhoea were selected and grouped as described below:

Groups (No. of calf=6)	Health status	Therapeutic strategy
Ι	Healthy control	_
II	Calves with diarrhoea	Tariflox OZ* + IV fluid X 5 days
Ш	Calves with diarrhoea	Polyherbal formulation*** + IV fluid X 5 days

Tariflox OZ\*: Ofloxacin-Ornidazole, marketed by CIPLA LTD. Oral, @ 10mg/kg b. wt. BID

Polyherbal formulation\*\*\*: Oral, @ 10 gm BID

Rehydration therapy was given to each diarrhoeic calf of both groups according to their fluid requirement. For rehydration therapy intravenous infusion of Ringer lactate (RL) was used.

#### **Clinical examination**

Detailed clinical examination was carried out before (0 day)

and after (5<sup>th</sup> day) therapy as per the standard procedure <sup>[4]</sup>. Clinical manifestations *viz*. faecal consistency, mucous membrane appearance, skin elasticity, enophthalmos and extremity temperature were recorded using the standard criteria <sup>[11]</sup> given in Table 1.

Clinical profile (Rectal temperature, respiration rates, pulse rates and heart rate) were recorded as per following criteria (Table 2).

#### **Table 1:** Criteria for recording clinical manifestation

Clinical manifestation	Assessment							
Chinical mannestation	Observation	S	Observation	S	Observation	S	Observation	S
Faecal consistency	Firm	+	Semisolid	++	Loose	+++	Watery	++++
Mucous membrane appearance	Normal	+	Dry	++				
Skin elasticity (Time elapsed to normal after tenting)	Wthin 2 seconds (Normal)	+	2-4 seconds	++	5-10 seconds	+++	>10 seconds	++++
Enophthalmos	Normal	+	Sunken	++				
Extremity temperature	Warm	+	Cool	++	Very cold	+++		

 $\overline{S}$  = Degree of severity

Table 2: Criteria for recording clinical profile

Clinical profiles		As	sessment			
Chincal profiles	Observation	S	Observation	S	Observation	S
Rectal temperature	Normal (101.5-102 °F)	+	High	++	Subnormal	-
Respiration rate	Normal (26-50/min)	+	High	++	Low	-
Pulse rate	Normal (100-120/min)	+	High	++	Low	-
Heart rate	Normal (48-84/min)	+	High	++	Low	-

S= Degree of severity

#### Haematobiochemical analysis

All the blood samples which were collected, evaluated immediately within 2 hours of collection of samples. Haematological (Hb, PCV, TEC, TLC, DLC, MCV, MCH, MCHC) and biochemical (Blood glucose, serum total protein, serum albumin, serum globulin, albumin: globulin ratio, Blood urea nitrogen and Creatinine) parameters were studied as per standard laboratory procedures.

#### Collection and analysis of faecal sample

The faecal material (4-5 gm) was directly picked up by introducing index finger in rectum with the gloved hands in sterilized culture bottles and shifted to the laboratory in ice box. Samples were kept in deep freezer till further processing. Two sets of faecal samples, one for detection of the presence of parasitic egg/ova/segment of gastrointestinal helminthes and another for isolation of *E. coli* and *Salmonella* were collected from each diarrhoeic cattle calf. A small amount of faeces on a clean glass slide was placed and diluted with a

drop of water and mixed thoroughly with wooden stick or glass rod. A cover slip was placed and examined under microscope. Diagnosis of helminth and coccidian infections was done with the floatation method by using the saturated sodium chloride solution <sup>[12]</sup>. Floatation solution was prepared by dissolving sodium chloride (table salt) to boiling water until the salt no longer dissolved and settled to the bottom of the pot. Two to five grams of faeces was placed in a cup. Flotation solution was added, directly, to the faeces and mixed thoroughly with a spatula. The mixture was strained through a tea strainer into a second cup. The contents of the second cup were poured into a test tube and the flotation solution was further added until a meniscus was formed. A cover slip was placed over the meniscus and allowed to remain there for 30 minutes. The cover slip was removed from the test tube, placed on the slide and examined under the microscope (100X magnification). Bacterial identification was done by culture method and confirmed by biochemical tests. (Fig 2)

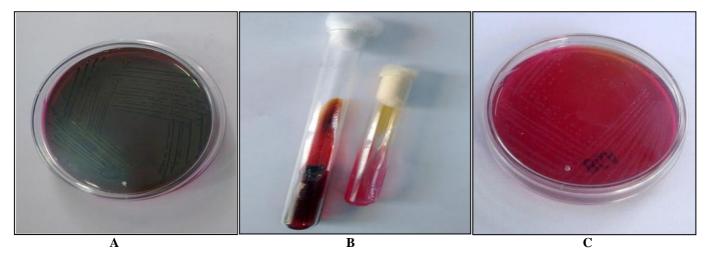


Fig 2: Isolation and confirmation of bacterial pathogens (A) *E. coli* on EMB agar (B) Salmonella on BGA (C) Biochemical Identification by kit (HiMedia)

#### **Evaluation of fluid deficit**

Fluid deficit was calculated either by clinical approach by assessment of degree of dehydration on the basis of clinical signs (skin elasticity, sunken face and eyes) along with skin fold test <sup>[4]</sup> or by laboratory approach by considering PCV and

total protein (TP). The total fluid deficit in milliliters was computed using the following formula.

Total fluid deficit (ml) = % Dehydration × Body weight (kg) × 10 Or Total fluid deficit (L) =  $BW \times 60/100 \times PD/100$ 

Where plasma deficit (PD) is calculated as-Plasma deficit (PD) = 100A (1-C/B) or PD% =  $(1-N/M) \times 100$ 

(A= body weight in kg, B= Normal percent PCV and C = Percent PCV in dehydrated animal, M= measured total proteins and N= normal total proteins in g/dL)

#### Therapeutic protocol

The diarrhoeic calves were divided in 2 groups (II and III) consisting of 6 calves in each group to evaluate therapeutic efficacies as per the experimental design given below. Six healthy calves served as control in group I.

#### **Therapeutic evaluation**

The efficacy of the therapy to treat diarrhea in calves was assessed by summation of the evaluation score computed as follows:

- 1. The score obtained by dividing total number of cases showing either disappearance of clinical manifestation or significant variation in each clinical profile toward normalcy by number of such manifestation on day 0 post treatment. The score so obtained was multiplied by the factor for day of such disappearance *viz.* 0.2, 0.4, 0.6, 0.8 and 1.0 for day 5, 4, 3, 2 and 1 respectively to obtain daily evaluation score.
- 2. The score recorded by dividing the number of cases recovered on each day by the number of diarrhoeic cases on the 0 day and multiplying it with the factor for day of such disappearance *viz.* 0.2, 0.4, 0.6, 0.8 and 1.0 for day 5, 4, 3, 2 and 1 respectively to obtain recovery score.
- 3. Recovery rate in % was calculated by dividing number of cases recovered by total number of clinical cases on day 0 multiplied by 100 in respective group.
- 4. Therapy gradation was obtained by summation of daily evaluation score of clinical manifestation, clinical profile and recovery score.

#### Statistical analysis

Statistical difference between respective means for various parameters was evaluated using appropriate statistical tests. One way ANOVA technique was employed to compare means as per the method described by Snedecor and Cochran (1994) <sup>[13]</sup>. Statistically significant difference was considered at 5% level.

#### **Results and Discussion**

Therapeutic efficacy of both groups was assessed on the basis of recovery manifested by degree of resolution of clinical manifestations, clinical profiles, haematological profiles, biochemical profiles and percent recovery of diarrhoeic calves.

#### **Clinical manifestation**

Detailed analysis of results revealed that normal skin elasticity and extremity temperature becomes retained by day 4 post treatment in both the group. Enophthalmos completely resolved by day 3 post treatment in both groups. Firm fecal consistency and normal mucus membrane were appeared by day 4 and 5 in group II and III respectively. Appetite becomes normal by day 3 and 4 post treatment in group II and III respectively. Individual therapeutic evaluation score card revealed superiority of group II treatment over group III (Table 3). Loss of skin elasticity and sunken eyes (enophthalmos) in diarrhoeic calve occurs due to fluid loss through watery feaces <sup>[4]</sup>.

<b>Table 3:</b> Total therapeutic evaluation score for different clinical
manifestation

Parameters	Group II	Group III
Faecal consistency	0.632	0.599
Appetite	0.750	0.600
Skin elasticity	0.840	0.760
Mucous membrane appearance	0.666	0.599
Enophthalmos	0.800	0.733
Extremity temperature	0.698	0.865
Sub total	4.386	4.156

#### **Clinical profiles**

In group II, Complete resolution of all clinical profiles *viz* rectal temperature, respiration, pulse rate and heart rate was recorded on 4<sup>th</sup> day after treatment while respiration rate was observed normal on 3<sup>rd</sup> day post treatment. While in group III complete resolution of all clinical entities towards normalcy was recorded on day 5 post treatments. Rectal temperature, respiration rate, pulse rate and heart rate decreased significantly (P < 0.05) after treatment as compared to their pretreatment values at day 0 (Table 4). Therapeutic evaluation score suggested that group III therapeutic regimen is more effective as compared to group III. (Table 5)

**Table 4:** Mean ± S.D. of clinical profiles in diarrhoeic calves of different therapeutic groups

Diarrhoea in calves								
Groups	oups Rectal temperature		Respirat	Respiration rate Pulse rate			Heart	rate
	Before	After	Before	After	Before After		Before	After
	Treatment	Treatment	Treatment	Treatment	Treatment	Treatment	Treatment	Treatment
Ι	100.2 <sup>a</sup>	100.2 <sup>a</sup> ±0.44 29.33 <sup>a</sup> ±0.88		80.50 <sup>a</sup> ±4.26		66.66 <sup>a</sup> ±5.90		
II	103.01 <sup>b</sup> ±0.18	101.30 <sup>a</sup> ±0.37	50.66 <sup>b</sup> ±2.33	$35.66^{a}\pm1.62$	121.00 <sup>b</sup> ±5.06	$86.50^{a}\pm1.58$	89.16 <sup>b</sup> ±1.51	$80.00^{a}\pm1.36$
III	$102.98^{b} \pm 0.14$	$101.08^{a}\pm0.32$	$50.50^{b}\pm1.17$	$34.83^{a}\pm1.68$	122.33 <sup>b</sup> ±3.75	$84.83^{a}\pm4.72$	$88.83^{b} \pm 3.01$	$75.33^{a}\pm2.15$

 Table 5: Total therapeutic evaluation score for different clinical profiles

Parameters	Group II	Group III
Rectal temperature	0.799	0.665
Respiration rate	0.799	0.766
Pulse rate	0.832	0.766
Heart rate	0.832	0.766
Sub total	3.262	2.963

#### Haematological profiles

In both the group, significant (P < 0.05) decrease was recorded in the mean values of haemoglobin (Hb), total erythrocyte count (TEC), packed cell volume (PCV), total leucocyte count (TLC) and neutrophils whereas lymphocyte counts increased significantly (P < 0.05) after treatment in diarrhoeic calves when comparison was done to their values before treatment. However, no significant change was observed in mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), monocytes, basophils and eosinophils post treatment (Table 6). The calculated value of therapeutic evaluation score was 0.42 and 0.50 in group II and III respectively. It indicates superiority of group III therapy over group II i.e. polyherbal formulation plus fluid over antibiotic plus fluid.

Table 6: Mean ± S.D. of haematological profiles in diarrhoeic calves of group II and III with control group I

Parameters	I	II		II	[
Farameters		<b>Before Treatment</b>	After Treatment	<b>Before Treatment</b>	After Treatment
Haemoglobin (g/dL)	10.93 <sup>a</sup> ±0.70	12.68 <sup>b</sup> ±0.21	11.20 <sup>a</sup> ±0.34	12.65 <sup>b</sup> ±0.54	10.46 <sup>a</sup> ±0.27
PCV (%)	37.00 <sup>a</sup> ±0.57	46.83 <sup>b</sup> ±2.12	$38.50^{a}\pm0.84$	$47.00^{b}\pm1.75$	$38.66^{a}\pm1.42$
TEC (10 <sup>6</sup> /µL)	$7.73^{a}\pm0.52$	10.21 <sup>b</sup> ±0.36	$8.36^{a}\pm0.20$	$10.18^{b} \pm 0.42$	8.25 <sup>a</sup> ±0.17
MCV (fL)	48.76 <sup>a</sup> ±2.71	$45.90^{a}\pm1.79$	46.15 <sup>a</sup> ±1.58	46.39 <sup>a</sup> ±1.95	47.03 <sup>a</sup> ±2.30
MCH (pg)	$14.26^{a}\pm0.88$	12.49 ±0.48	13.40 <sup>a</sup> ±0.44	12.59 <sup>a</sup> ±0.90	12.71 <sup>a</sup> ±0.42
MCHC (%)	29.54 <sup>a</sup> ±1.82	$27.34^{a}\pm1.24$	29.20 <sup>a</sup> ±1.30	27.25 <sup>a</sup> ±2.01	27.23 <sup>a</sup> ±1.15
TLC (10 <sup>3</sup> /µL)	$11.40^{a}\pm0.28$	18.30 <sup>b</sup> ±0.73	$11.60^{a}\pm0.21$	18.31 <sup>b</sup> ±0.62	11.38 <sup>a</sup> ±0.53
Neutrophils (%)	38.00 <sup>a</sup> ±1.03	$54.50^{b}\pm1.47$	39.50 <sup>a</sup> ±1.72	$54.33^{b}\pm1.28$	43.50 <sup>a</sup> ±0.42
Lymphocytes (%)	$51.83^{a}\pm0.90$	$34.00^{b} \pm 0.96$	$52.50^{a}\pm1.25$	33.83 <sup>b</sup> ±2.52	$47.50^{a}\pm1.45$
Monocytes (%)	$4.50^{a}\pm0.34$	$4.66^{a}\pm0.88$	3.83 <sup>a</sup> ±0.60	$4.66^{a}\pm0.88$	$4.16^{a}\pm0.60$
Basophils (%)	$0.33^{a} \pm 0.21$	$0.83^{a}\pm0.30$	0.33 <sup>a</sup> ±0.33	$0.83^{a}\pm0.40$	0.66 <sup>a</sup> ±0.21
Eosinophils (%)	$5.33^{a}\pm0.42$	$6.00^{a}\pm1.00$	3.83 <sup>a</sup> ±0.65	6.33 <sup>a</sup> ±1.11	4.16 <sup>a</sup> ±0.65

#### **Biochemical profiles**

In both group, biochemical analysis showed a significant (P < 0.05) elevation in blood glucose concentration, serum sodium and serum chloride concentration and a significant (P < 0.05) decrease was recorded in serum total protein level along with albumin, blood urea nitrogen (BUN) and creatinine post treatment as compared to their pretreatment levels. However, mean values of A: G ratio and serum potassium did not reveal

any significant alteration during pre and post treatment period. In group II no significant difference was recorded in globulin while in group III globulin was significantly decreases in post treatment when compared with pre treatment value (Table 7). Therapeutic evaluation score for biochemical profile was found as 0.70 and 0.80 in group II and III respectively which indicates superiority of polyherbal formulation over antibiotics.

Table 7: Mean ± S.D. of biochemical profiles in diarrhoeic calves of group II and IV with control group I

Parameters	Ι	П			III
Parameters		<b>Before Treatment</b>	After Treatment	<b>Before Treatment</b>	After Treatment
Glucose (mg/dL)	$60.00^{a}\pm2.78$	$40.66^{b}\pm1.02$	53.50 <sup>a</sup> ±2.12	37.50 <sup>b</sup> ±1.92	$48.00^{a}\pm1.87$
Total Protein (g/dL)	$8.20^{a}\pm0.04$	10.61 <sup>b</sup> ±0.39	$8.00^{a} \pm 0.17$	10.90 <sup>b</sup> ±0.49	8.06 <sup>a</sup> ±0.19
Albumin (g/dL)	3.35 <sup>a</sup> ±0.18	4.76 <sup>b</sup> ±0.22	3.65 <sup>a</sup> ±0.15	$4.46^{b}\pm0.16$	3.61 <sup>a</sup> ±0.09
Globulin (g/dL)	4.85 <sup>a</sup> ±0.17	5.85 <sup>a</sup> ±0.57	4.35 <sup>b</sup> ±0.27	6.43 <sup>a</sup> ±0.57	4.45 <sup>a</sup> ±0.27
A:G Ratio	$0.70^{a}\pm0.06$	$0.87^{a}\pm0.12$	$0.86^{a} \pm 0.07$	0.73 <sup>a</sup> ±0.09	$0.83^{a} \pm 0.06$
BUN (mg/dL)	$20.76^{a}\pm0.61$	30.96 <sup>b</sup> ±1.16	$24.98^{a}\pm0.20$	30.96 <sup>b</sup> ±0.66	24.11 <sup>a</sup> ±0.51
Creatinine (mg/dL)	1.43 <sup>a</sup> ±0.12	2.48 <sup>b</sup> ±0.22	$1.60^{a} \pm 0.08$	2.95 <sup>b</sup> ±0.15	$1.60^{a}\pm0.12$
Sodium (mEq/L)	$141.50^{a}\pm3.01$	123.83 <sup>b</sup> ±1.90	137.33 <sup>a</sup> ±2.10	124.83 <sup>b</sup> ±2.49	139.00 <sup>a</sup> ±2.39
Potassium (mEq/L)	$4.80^{a}\pm0.17$	4.68 <sup>a</sup> ±0.27	4.65 <sup>a</sup> ±0.20	4.61 <sup>a</sup> ±0.39	4.68 <sup>a</sup> ±0.27
Chloride (mEq/L)	$102.00^{a}\pm1.59$	87.16 <sup>b</sup> ±2.34	99.33 <sup>a</sup> ±1.94	86.50 <sup>b</sup> ±2.97	$101.66^{a}\pm 2.09$

#### **Recovery rate**

Recovery score was estimated based on number of diarrhoeic calve day 5 post treatment in different therapeutic groups as shown in (table 8). Two diarrhoeic calves each in group II and III recovered from diarrhoea on day 2 post treatment. 3 calves in group II and 2 calves in group III recovered on day 3 post

treatment. All the treated calves of group II and III recovered on day 4 and day 5 post treatments respectively. Recovery rate in both group were 100% because of rehydration therapy by intravenous fluid. Calculated evaluation score for both groups are 0.632 and 0.565 respectively.

Table 8: Recovery rate based on number of diarrhoeic calves post -treatment in different groups

Dorrow of the other and	Groups			
Days post treatment	II	IV		
0	6	6		
1	6	6		
2	4	4		
3	1	2		
4	0	1		
5	0	0		
Total score	100%	100%		

#### **Cost of treatment**

The cost of treatment calculated per calf to be rupees 60.06 and rupees 9.37 excluding rehydration therapy and rupees

180.06 and rupees 129.37 including rehydration therapy in group II and III respectively (Table 9). The evaluation of economy of treatment cost was calculated in terms of number

of calves recovered from diarrhoea in each group. Total cost of treatment per calf in group III by using PHF and I.V. fluid costs rupees 129.37 as total cost of treatment per calf showed 100% recovery. In group II, calves were treated by tab Tariflox OZ with I.V. fluid costs rupees 180.06 as total cost of treatment per calf was the most expensive and showed equally effective results (100% recovery) as group III. Group III treated with PHF and I.V. fluid was found to be most suitable than group II (both showed 100% recovery) as it was light in economics as well as less danger of emergence of antibiotic resistance and prolong withdrawal period.

Table 9: Economics	of various	treatments
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Particulars	Groups				
Paruculars	II	III			
No. of doses used/group	34	38			
Cost*/dose (rupees)	10.60	1.48			
Total cost of treatment/group (rupees)	360.40	56.24			
No. of calves recovered	6	6			
Cost of treatment/calf (rupees)	60.06	9.37			
Average quantity of RL administered <i>iv</i> /group	12	12			
Cost** of <i>iv</i> RL/group (rupees)	720	720			
Cost of rehydration therapy/group (rupees)	720	720			
Cost of rehydration therapy/calf (rupees)	120	120			
Total cost of treatment/calf (rupees)	180.06	129.37			

\*Zingiber officinale, Dalbergia sissoo, Aegle marmelos, Emblica officinalis and Eugenia jambolana @ rupees 200,150,100,150 and 140 per kilogram respectively.

\*\*@ rupees 60 per litre

#### **Therapy gradation**

Therapy gradation was done on the basis of therapeutic evaluation procedure explained in this study. Tab Tariflox OZ along with I.V. fluid (group II) was graded superior for diarrhoeic calves with a total score of 9.40 followed by PHF and I.V. fluid (group III) with total score of 8.984. Those calves, who could not recover even after end of the study, were treated with suitable anthelmentics (Table 10).

Various phytochemical like alkaloids, tannins, steroids, flavonoids, terpenoids and cardiac glycoside are present in rhizome of Zingiber officinale which impart antimicrobial activity against *E.coli* and *Salmonell* spp. <sup>[14]</sup>. Besides, it also has strong anti-oxidant effect and removes free radicals from tissues <sup>[15]</sup>. Antidiarrhoeal property of this herb is due to inhibition of gastrointestinal motility and secretion, antispasmodic effect of GIT musculature and reduced gastric emptying <sup>[16]</sup>. Fruit pulp of Aegle marmelos contains various phytochemicals like steroids, terpenoids, flavonoids, phenolic compounds, lignin, fat and oil, inulin, proteins, carbohydrates, alkaloids, cardiac glycosides and flavonoids [17]. Unripe fruit extract of Aegle marmelos possess antidiarrhoeal and gastroprotective properties. Antidiarrhoeal activity is due to its ability of decreasing intestinal transit time and secretions <sup>[18]</sup>. Leave extract of *Dalbergia sissoo* exerts analgesic, antiinflammatory, antipyretic, antimicrobial, antidiarrhoeal, antiulcerogenic, anti-spermicidal activity [19]. Seeds of Emblica officinalis is rich source of vitamin C which is essential for the restoration of the mucosal barrier function in intestine as it is also helpful in the formation of collagen. Besides, it is a necessary nutrient for the immune system. The clinical observations reported herein demonstrate a positive response to vitamin C supplementation in newly born calves affected with neonatal calf diarrhoea <sup>[20]</sup>. Fruits of Eugenia jambolana contain jambolin which has digestive, carminative and antipyretic property <sup>[21]</sup>.

Although the recovery percent of both group was 100%, therapeutic evaluation reflect that treatment with tab Tariflox OZ and intravenous fluid with a total score of 9.40 was graded superior treatment for diarrhoeic cases followed by PHF and I.V. fluid with a total score of 8.984. But due to non

or less availability of antibiotics in remote area and also because of problem of antimicrobial resistance, polyherbal formulation therapy is more convenient and cost effective as compared to conventional antibiotic therapy. Also, in light of percent efficacy and danger associated with prolong withdrawal period, the PHF along with intravenous fluid may be a most suitable and cheap therapeutic alternative for the treatment of calf diarrhea. On the basis of results of present investigation, the use of PHF along with intravenous rehydration therapy may be recommended for successful treatment of diarrhea in calves.

<b>Table 10:</b> Therapy gradation on the basis of profile wise evaluation
score

Profiles	Gro	Groups	
	II	III	
Clinical examination	4.386	4.156	
Clinical profiles	3.262	2.963	
Haematological profiles	0.42	0.50	
Biochemical profiles	0.70	0.80	
Recovery rate	0.632	0.565	
Total score	9.40	8.984	
Therapy gradation	Ι	Π	

#### Conclusion

Above description suggest that ethnoveterinary approach may be an alternate way to treat the calf diarrhea. Although it requires somewhat longer duration of therapy as compared to conventional antimicrobial therapy, it is more economic than earlier one so beneficial for poor farmers in rural areas. Besides, various problems associated with antimicrobial use like antimicrobial resistance, withdrawal period, side effects are not of significant concern with herbal therapy. So, ethnoveterinary approach may be promoted for not only treating calf diarrhea but also for as many diseases as possible.

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#### Declaration

Ethical approval has been taken from the institutional animal ethic committee prior to conducting research.

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