OBSERVATIONS ON CLINICAL TREATMENT TRIALS OF INDUCED ESCHERICHIA COLI DIARRHOEA IN BUFFALO NEONATAL CALVES

M. T. Javed, A. Khan, Abid Hussain and Babar Niaz

Department of Veterinary Pathology,

University of Agriculture, Faisalabad-38040, Pakistan

ABSTRACT

The study was conducted on 24 calves of one-week-age, randomly divided into four groups, of six calves each. First three groups were experimentally infected with *E. coli* and given treatment in various combinations, while group four was taken as uninfected untreated control. Results obtained on different parameters were, green to yellow-white diarrhoea with variable consistency from watery to semisolid. Generally in all groups during first two days, calves passed faeces around 6-10 times a day and this frequency lowered in about 2-4 days. During first 3-4 days of treatment, all calves appeared weak, dull, depressed with cold skin and wetness of the muzzle, while oral mucosa was pale and eyes were sunken. Overall mean respiration rate of treatment groups was slower (P<0.05) than control group. However, pulse rate showed no difference between treatment and control group. Overall mean rectal temperature was lower (P<0.05) in calves treated with antibiotic alone and with antibiotic + electrolytes, while body weight was higher (P<0.05) in calves treated with antibiotic but was lower along with creatinine (P<0.05) in calves treated with antibiotic + electrolytes and those treated with electrolytes alone.

Keywords: Diarrhoea, E. coli, Buffalo calves, antibiotics, respiration, pulse, temperature, body weight

INTRODUCTION

It has been reported that calf mortality of about 20 per cent can reduce net profit of up to 38 per cent and can result in shortage of milk and meat supplies (Khan and Khan, 1996). The incidence of calf mortality in cattle and buffalo has been reported to range from 29.1-39.8 per cent in Pakistan (Afzal et al., 1983) and 14.2-22.5 per cent in India (Khera, 1981). In the first month of age, diarrhoea accounts for 3.6-6.3 per cent mortality (Khan and Khan, 1991) in an overall mortality of 7.1 and 11 per cent (Khan and Khan, 1996) in cow and buffalo neonates, respectively. The diarrhoeic calves show extreme metabolic acidosis, haemoconcentration and hypofunction of both kidney and liver (Maach et al., 1992). Treatment of experimentally infected calves with enterotoxigenic E. coli (09;K30;K99) showed that caloric oral rehydration solution with glutamine is more effective in correcting plasma extracellular fluid and blood volume (Brooks et al., 1997). It has also favourable effect on hyponatraemia and metabolic acidosis than standard ORS. Michna et al.(1996) used 8.4 per cent sodium bicarbonate solution intravenously and found improvement in the general health of the calves in 3-24 hours while Holck et al. (1994) reported that treatment with danofloxacin (1.25mg/Kg i.m.) resulted in significant improvement in clinical condition, increased weight gain and fewer mortalities. Aly et al. (1996) treated diarrhoeic calves

with 20% ampicillin in combination with fluid therapy and observed great improvement in clinical symptoms, blood picture and blood serum constituents. As evident from the above studies various remedial measures have been suggested in overcoming the problem in calves. However, present study was aimed to see the effect of various treatments on clinical condition of the buffalo calves and also to evaluate different treatment protocols for treatment of diarrhoea.

MATERIALS AND METHODS

The study was carried out on 24, one-week-old male buffalo calves. These calves were randomly divided into four groups, having six calves in each. Faecal samples of these calves were examined for parasites and pathogenic *E. coli* and only negative calves to these were included in the study.

Infection with enteropathogenic *E. coli*, isolated from field cases of diarrhoea, was administered orally to induce diarrhoea to these calves. For this purpose 2mL broth culture having coliform counts of 1010 colony forming units (CFU) of *E. coli* (Brooks *et al.*, 1996) was administered to groups, one through three, while the calves of group four were taken as uninfected control. The first three groups (infected with E. coli) were given treatment as shown below:

Group	Exp. Infection	Treatment
1	•	Kanamycin (@ 10 mg/kg body weight) i.m. Rehydration solution (200 mL/day) i.v.
2	+	Kanamycin (@ 10 mg/kg body weight) i.v.
3	+	Rehydration solution (200 mL/day) i.v.
4		No treatment

The composition of the rehydration solution used for the treatment is given below:

NaCl	10 gm
Glucose	50 gm
KCL	04 gm
NaHCO ₃	30 gm
Distilled water	4 Liter

All the calves were kept under identical conditions of feeding and management. They were given ½ litre milk, thrice daily and water ad libitum. The study was conducted during October and the calves were kept in sheds during the night, while in the open under the sun in the noon hours.

Body weight of individual calves in each group was recorded daily by using large weighing scale. Clinical signs exhibited by the calves and mortality during the study period was recorded for which calves were monitored daily. Pulse rate was recorded twice a day, i.e., at morning and evening from ventral coccygeal artery. Rectal temperature and respiration rates were recorded twice a day. Development of colour, odour, frequency and nature of diarrhoea, i.e., watery, mucoid, blood mixed etc. was also recorded.

Data thus obtained were subjected to analysis of variance technique and means were compared by LSD and DMR on personal computer by using SAS 6.2.1 statistical software package (Anonymous, 1996). Pearson correlation coefficients between various parameters were also obtained.

RESULTS AND DISCUSSION

Neonatal calf diarrhoea is considered the major cause of losses in Livestock (Khan and Khan, 1996) among the various disease conditions, *E. coli* diarrhoea is the important cause of mortality (Khan and Khan, 1991) in Pakistan. Diarrhoea results in blood acid base imbalance, electrolyte depletion and more importantly hypovolaemia, consequently resulting into shock and death (Roussel and Kasari, 1990). During present study

the local pathogenic *E. coli* strain was administered orally to induce diarrhoea in three groups of calves, which started within 4-6 hours of experimental infection in all calves.

Colour and consistency of diarrhoea observed during present study (Table 1) was in complete agreement as reported by Morin et al. (1976) who also observed watery and yellowish diarrhoea and stated that the colour and consistency remain the same, whatever etiology of diarrhoea. Frequency of diarrhoea was high (10 times) initially (at day 1-3) but improved subsequently with the start of the treatment (Table 1). The statistical analysis of respiration, pulse, temperature and weight also suggests improvement in these parameters at day four, as was observed for other clinical parameters (Tables 3,4). The response to treatment was early in calves treated with antibiotic + electrolytes as compared with other groups. This was in line with the findings of Aly et al. (1996) who also reported significant improvement in clinical symptoms in calves treated with antibiotic and fluid. The signs observed in calves due to diarrhoea were almost same as reported by Naylor (1987). The improvement in visible picture of the calves was complete at day six. Overall lower respiration and body weight of calves treated with antibiotic than other treatment groups (Tables 3.4) indicate relatively poor response to antibiotic alone. The temperature in all the groups remained close to the normal till day two of the treatment but it decreased from day three onward (Table 4). However, increased body temperature, pulse and respiration rates in otherwise debilitating, anorexic and lethargic calves have been reported by Sridhar et al. (1988). Hegazy et al. (1996) also reported an initial increase in body temperature in calves suffering from diarrhoea. The close to normal temperature without rise, during present study was probably due to the early start of treatment, furthermore, the experiment was conducted during the winter months. Therefore, the early start of treatment and winter season were probably responsible for normal temperature during first two days and decrease afterwards that was only observed in calves treated with antibiotics. This again suggests that treatment with antibiotics alone is probably not that effective.

Mortality occurred in calves given antibiotic only and out of six, two (33%) calves died, one at day two and second at day five (Table 2). This suggests that treatment of diarrhoea without electrolytes is not useful and probably correction of acidosis and electrolytes level is of more value in saving calves. These findings are in complete agreement with Brooks et al. (1996) who stated that ORS provide significant nutritional support, effective rehydration, correction of acid base imbalance and electrolytes disturbances. They further stated that all this

is very important and useful in young animals. Similarly, Sadiek and Schlerka, (1995) reported that oral or intravenous use of solution of sodium bicarbonate, sodium chloride and glucose gave good results.

Lower (P<0.05) body weight of calves treated with antibiotic alone than control and other treatment groups (Table 4) further strengthen the evidence of mortality in this group. It has been observed that calves can die losing as little as 6.5 per cent of their body weight, however, the average loss of body weight resulting in calf mortality is reported to be 13.4 per cent (Groutides and Michell, 1990). It has been observed that even plain ORS do not lead to improvement in the body weight but glutamine solution and high energy ORS do (Brooks et al, 1997).

The solution used during present study for intravenous therapy consisted sodium chloride, potassium chloride, sodium bicarbonate, glucose and water. Sodium chloride and potassium chloride were used to check the depletion of these ions, while sodium bicarbonate to correct the acid base imbalance and glucose to provide the energy. Roussel and Kasari, (1990) have recommended the same combination of electrolytes through intravenous route as treatment of diarrhoea and present study was carried to evaluate its response under local conditions for the treatment of *E. coli* diarrhoea. The combination of these electrolytes during present study has proven very useful alone or in combination with antibiotic.

Table 1: Development, nature, frequency and colour of diarrhoea in different groups of calves.

Calves			Diarrhoea						
	Nature	Frequency	Colour	Time/hours after infection					
Group 1	,								
1	Semisolid	7.	Greenish	3.00 hrs					
2	Watery	10	Sulphur + blood flakes	4.30					
3	Watery	9	Yellow	3.20					
4	Watery + semisolid	8	Dirty yellow	3.40					
5	Watery	10	Yellow	2.50					
6	Watery	8	Greenish yellow	6.30					
Group 2									
1	Watery	9	Yellow	3.30					
2	Watery	8	Sulphur yellow	4.00					
3	Semisolid	7	Greenish	3.05					
4	Watery	8	White+blood flakes	2.30					
5	Watery	7	Dirty yellow	5.00					
6	Watery	9	Yellow	3.15					
Group 3			× .						
1	Semisolid	6	Greenish + blood flakes	4.00					
2	Watery	8	Whitish yellow	3.20					
3	Semisolid	7	Greenish	2.30					
4	Semisolid	9	White	4.00					
5	Watery	10	Yellow	4.30					
6	Watery	8	Greenish yellow	6.15					
Group 4									
	misolid + solid	3-5	Whitish yellow	and the second					

Table 2: Recovery pattern of calves during treatment trials in different groups.

Groups	Died							- FIG. 1	Recovered								
	0	1	2	3	4	5	6	7	Ō		1	2	3	4	5	6	7
				4										_			
G-1	-	-	-	-	-	-	-	-	-		•	•	3	3	-	-	-
G-2			1	-		1	•	-					-	-	3	1	-
G-3					-	•	-							2	2	2	
G-4	-			-	-				-			-	*	- 1	-	-	

Table 3: Respiration and pulse rate of calves of treatment groups and control group in overall and at different days.

Groups

Days

	-20			Viene de la companya	15.4			27 704
	0	1	2	3	4	5	6	Overall
₹es	piration		10000	XXX 500		555		202
	а	Bbc	Bbc	Bbc	С	b	bc	В
1	44.00	33.33	32.83	28.83	27.33	35.33	31.50	32.86
	±10.58	±2.07	±2.86	±3.71	±3.05	±5.51	±7.78	±6.07
	а	Cb	Cab	ABab	ab	ab	ab	С
2	34.67	22.00	26.00	33.00	26.50	27.00	24.00	27.41
	±8.33	±3.52	±2.31	±8.45	±2.12	±4.24	±2.01	±6.66
	а .	ABab	BCbc	Bbc	bc	abc	С	В
3	40.67	34.67	29.50	29.67	26.33	33.33	23.50	31.41
8	±6.43	±7.50	±4.18	±5.99	±5.13	±6.35	±4.95	±6.98
	A	AA	3.550.00.00.00.00.00.00.00.00.00.00.00.00	A	500.000	0.775.57.50		•
4	32.00	41.33	44.00	39.33	37.33	39.33	38.00	39.33
1.0	±4.00	±8.32	±4.00	±8.08	±10.06	±12.05	±6.00	±8.11
Pul	se Rate					ATT. 1.1.TELET. TO	matana.	
	ab	Aa	ab	ABb	ABb	ABb	ab	Α
1	58.00	76.67	65.00	56.00	52.00	54.67	45.50	62.14
	±19.08	±15.81	±11.98	±5.66	±7.21	±3.21	±14.85	±13.41
	110.00	B	B	AB	В	B	211.00	2.0.11
2	53.33	58.33	54.00	50.75	46.00	47.00	56.00	53.23
	±8.33	±1.86	±10.58	±3.78	±8.49	±7.07	±0.01	±7.00
	a	Bab ab	Aab Bc	ABabc bc	B	27.01	20.01	27.00
3	60.67	59.33	58.50	59.00	41.33	49.33	48.50	55.59
	±7.57	±6.47	±4.46	±5.37	±5.51	±12.42	±4.99	±8.60
	11.51	B. 47	A A	A	AB	112.72	14.55	20.00
4	52.00	56.67	64.67	58.00	58.67	64.67	52.67	58.19
7	±8.00	±13.01	±7.57	±12.16	±6.11	±4.16	±15.27	±9.79
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Values in each row with different small letters and in each column with different capital letters are statistically different (P<0.05). Each figure represent mean ± standard deviation.

Table 4: Rectal temperature and body weight of calves of treatment groups and control group in overall and at different days.

Grou	ps			Days				
	0	1	2	3	4	5	6	Overall
Rec	tal Temperat	ture			7			
	a	ab ABab	Bb b	Bb ab	В			
1	101.87	101.63	100.68	100.23	100.40	100.20	101.50	100.89
	±0.42	±1.41	±0.72	±0.15	±0.53	±0.01	±0.42	±0.99
	ab	ab Bb	Bb b	Bb a	В			
2	101.53	101.23	100.43	100.40	100.10	100.20	102.40	100.83
	±1.03	±1.14	±0.44	±0.54	±0.14	±0.02	±0.01	±0.93
	ab	ab Aab	Bab b	Aab a	AB .			
3	101.20	101.07	101.43	100.77	100.33	101.13	101.90	100.08
	±0.69	±0.63	±0.48	±0.87	±0.58	±0.78	±0.14	±0.70
	_0.00	AB	A	A	Α			
4	101.67	101.66	101.33	101.73	101.13	101.73	101.47	101.53
30	±0.81	±1.10	±0.90	±0.83	±0.58	±0.30	±0.92	±0.72
Roc	ly Weight	2			(1707)(1707)	(and a c min)	77,700,700	
500	AA	Α		Α				
1	40.00	40.00	41.33	41.33	41.33	41.00	41.00	40.79
•	±3.61	±3.23	±2.88	±2.88	±3.21	±3.61	±5.66	±2.85
	10.01	B B	B	12.00	C	20.01	20.00	
2	33.67	33.67	34.50	34.50	34.50	34.50	34.00	34.14
-	±4.04	±3.60	±1.73	±1.73	±2.12	±2.12	±1.01	±2.47
	14.04	A A	A	20	A			
3	41.00	41.00	42.67	42.67	42.33	42.00	40.50	41.89
	±3.41	±3.09	±2.88	±2.88	±4.62	±4.36	±2.12	±3.04
	10.71	AB AB	A	22.00	В	_1,00		
4	38.00	37.33	37.67	39.33	37.67	38.33	36.67	37.86
7	±5.00	±7.02	±7.09	±3.21	±5.03	±5.03	±6.50	±4.84

Values in each row with different small letters and in each column with different capital letters are statistically different (P<0.05). Each figure represents mean ± standard deviation.

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