

HAEMATOLOGICAL AND BACTERIOLOGICAL STUDIES IN NEONATAL LAMBS WITH REFERENCE TO NEONATAL LAMB MORTALITY

M. Naveed, M. T. Javed, A. Khan and R. Kausar¹

Department of Veterinary Pathology, ¹DVM student, Faculty of Veterinary Science, University of Agriculture Faisalabad-38040, Pakistan

ABSTRACT

The study was conducted on 100 neonates born to ewes of Pak-Karakul breed during breeding season starting from February to March 1998. The haematological parameters showed non-significant differences between neonates those died and survived and between those having different birth weights and also between those born to ewes of different parities. All the haematological parameters showed no difference in neonates of two sexes except eosinophils which were higher ($P < 0.05$) in male neonates. From a total of seven neonates died within a week of birth, three had diarrhoea and four had signs of pneumonia. Of the three cases of diarrhoea, single organism was isolated from one while in other two mixed cultures were obtained. From four cases of pneumonia, single organism was isolated from three, while in one case mixed culture was obtained. From a total of 16 morbid neonates (dead neonates are also included) seven showed diarrhoea and nine pneumonia. From the cases of diarrhoea (listed in morbid neonates), mixed culture was obtained. However, *E. coli* was isolated from four cases with other organism and *Bacillus* was isolated from two cases with other organisms. From the morbid cases of pneumonia, single isolate was obtained from two cases of the five, while in other three mixed culture was obtained. *Bacillus* and *Pseudomonas* were isolated as single isolate from the three cases of neonates died of respiratory sings.

INTRODUCTION

Many infectious and noninfectious conditions are deterrent in the development of sheep industry in Pakistan. Neonatal period is very critical in which infectious and noninfectious conditions result into bountiful losses. Young ones of ruminants are born with zero to low immunity while the environment it faces is contaminated with different infectious agents. Various bacterial agents involved in neonatal lamb mortality are *E. coli* causing enteritis (Sawyer *et al.*, 1977), *Cl. perfringens* type D causing lamb dysentery and deaths of neonates (Snodgrass and Gray, 1977; Eales *et al.*, 1986). Chaarani *et al.* (1991) isolated haemolytic and non-haemolytic strains of *E. coli* from enteritis in neonates and Sawyer *et al.* (1977) isolated Clostridial spp. from lambs suffering from enteritis. Sawyer *et al.* (1977) isolated *pasteurella haemolytica*, *Staphylococcus aureus* and *Corynebacterium pyogenese* from dead lambs. While Streptococci, Staphylococci, *Fusiformis necrophorus* and Clostridial spp. were the agents isolated from cases of navel ill-septicaemia, resulting in mortality.

Keeping in view, the threat of bacterial infections in neonatal life this study was carried out to investigate various bacterial agents involved in neonatal lamb mortality. Haematological studies on neonates were also

carried out and effort was made to find out any possible correlation between haematology with birth weight and sex of lamb, parity of the dam and neonatal lamb mortality.

MATERIALS AND METHODS

The study was carried out at Sheep and Goat Development Centre, Rakh Khairwala, District Layyah, on 100 Pak-Karakul neonatal lambs, after lambing to one month of age. These lambs were randomly selected during lambing season, starting from February to April 1998.

All the neonatal lambs were kept under identical conditions of feeding and management. The lambs were housed collectively in sheds having shelter and open space. The neonatal lambs were allowed to suckle dams *ad libitum* during first week, thereafter, twice daily, i.e., early morning and in the evening, in addition green fodder was also offered.

Age and parity of the dam along with sex and birth weight of the lambs were recorded. Neonates under study were monitored for any clinical sign due to disease condition up to one month of age.

Blood samples with EDTA were collected from jugular vein from each neonate within 24-36 hours of age

for haematological studies. RBC counting and TLC were done with the help of improved Neubaur counting chamber (Coles, 1988). Haemoglobin was estimated by acid haematin method as described by Benjamin (1979) using Sahli's apparatus. The PCV was determined by microhaematocrit method (Coles, 1988). The procedures of Coles (1988) for preparation of slides for differential leukocyte count was followed.

Samples for bacteriological examination were obtained from neonates having nasal discharge and/or diarrhoea. Cotton swabs were used to obtain samples from nares and throat keeping aseptic precautions. Faecal samples from the rectum were collected from cases of diarrhoea. Morbid tissues from dead neonates were collected and samples for bacterial isolation were taken, keeping aseptic precautions. Samples collected for bacteriological studies including nasal and throat swabs and faecal materials from the rectum were cultured in nutrient broth. The primary cultures thus obtained were transferred on nutrient agar, blood agar and MacConkey's agar plates. Each of the isolate was identified on morphological, cultural and biochemical characters as described by Cruickshank *et al.* (1974).

Data thus obtained on various parameters was analysed by using one way analysis of variance technique and means were compared by using DMR test on personal computer, by using SPSS computer software package (Anonymous, 1996).

RESULTS AND DISCUSSION

Haematology

The results of haematology during present study revealed slightly lower values in neonates those died as compared to healthier, although the difference was non-significant (Table 1). This blood picture probably reflects an acute death of the neonates without significant changes in blood picture or the haematological variation does not occur early in neonatal life. Similarly, Eales (1986) reported no change in haematology in neonates suffering from watery mouth disease. However, Adam (1992) reported significant difference in haematological values with higher ($P < 0.05$) values of neutrophils and monocytes in ill calves which might be due to specie difference.

The effect of sex of neonates also revealed non-significant difference on haematological parameters except eosinophils that were higher ($P < 0.05$) in males (Table 2). Similarly, Khan and Khan (1996) reported non-significant difference in haematological parameters in the two sexes of buffalo neonates.

During present study, the effect of birth weight on haematological parameters was also non-significant

(Table 3). However, the values of PCV and lymphocytes were relatively lower in neonates of less than 3 kg. This suggests that haematological variation probably do not depend on birth weight of neonates.

The effect of parity of the dam during present study also showed non-significant difference, however, most of the blood parameters were relatively higher in neonates of dams at their 4th and 5th parity (Table 4). Similarly, Khan and Khan (1996) also reported non-significant difference between blood parameters in buffalo neonates born to dams of different parity.

Bacteria Isolated From Morbid Neonates

The organisms isolated during present study from neonates showing the signs of diarrhoea include *E. coli*, *Bacillus*, *Pseudomonas* and *Proteus* (Table 5). These were almost similar as recovered from dead neonates (Table 5). Sawyer *et al.* (1977) isolated *E. coli* from cases of diarrhoea which was also isolated from most of the cases of diarrhoea during present study. Janke *et al.* (1990) also observed *E. coli* in most cases of diarrhoea. The organisms isolated from cases of pneumonia during present study were *E. coli*, *Streptococci*, *Proteus*, *Staphylococcus* and *Bacillus*. This suggests high prevalence of these organisms in the area causing disease in neonates. These findings were in line with those of Sawyer *et al.* (1977) who reported *E. coli* and *Staphylococcus* from pneumonia in neonates. Present findings agreed with Mason and Corbould (1981) who recovered *E. coli* from both rectal and nasal samples. It can be deduced from the present findings that *E. coli* is the main organism responsible for disease involving both gastrointestinal tract and respiratory system.

Bacteria Isolated From Dead Neonates

The organisms isolated during present study from neonates died off diarrhoea were *E. coli*, *Bacillus* spp., *Streptococcus* spp. and *Corynebacterium* spp. (Table 5). This was in line with the findings of Radostitis (1986) and Khan and Khan (1996). They also had the view that intestinal lesions were not characteristic of any single etiological agent of diarrhoea in neonates.

The organisms isolated from neonates died off pneumonia were *E. coli*, *Streptococci*, *Pseudomonas* and *Bacillus*. The gross and histopathological picture were non-specific for these organisms. However, all cases revealed almost a similar picture of these organs both grossly and microscopically.

ACKNOWLEDGMENTS

Financial support provided by the Pakistan Science Foundation under Research Project No. P-AU/AGR (199) is highly acknowledged.

Table 1: Comparison of RBC, PCV, Hb, erythrocytic indices, TLC and DLC (Means \pm SD) of neonatal lambs those survived or died.

Parameters	Lambs	
	Died	Live
RBC ($10^6/\mu\text{L}$)	7.53 \pm 1.22	7.66 \pm 1.02
PCV (%)	39.57 \pm 5.82	40.60 \pm 7.81
Hb (g/dL)	10.90 \pm 1.29	11.20 \pm 1.62
MCV (fl)	54.16 \pm 14.62	54.03 \pm 13.85
MCH (pg)	14.85 \pm 3.32	14.87 \pm 3.18
MCHC (%)	28.00 \pm 4.97	28.02 \pm 4.67
TLC ($10^3/\mu\text{L}$)	7.34 \pm 2.20	8.99 \pm 3.04
Neutrophils (%)	69.17 \pm 11.05	68.16 \pm 12.45
Lymphocytes (%)	29.83 \pm 10.94	30.72 \pm 13.34
Eosinophils (%)	1.25 \pm 0.50	2.13 \pm 1.60
Monocytes (%)	1.00 \pm 0.59	1.50 \pm 1.10

Table 2: Effect of sex on RBC, PCV, Hb, erythrocytic indices, TLC and DLC (Means \pm SD) of neonatal lambs.

Parameters	Lambs	
	Male	Female
RBC ($10^6/\mu\text{L}$)	7.79 \pm 1.14	7.49 \pm 0.91
PCV (%)	40.00 \pm 7.65	40.97 \pm 7.65
Hb (g/dL)	11.10 \pm 1.46	11.22 \pm 1.71
MCV (fl)	52.31 \pm 13.54	55.68 \pm 14.06
MCH (pg)	14.52 \pm 3.12	15.19 \pm 3.21
MCHC (%)	28.30 \pm 4.23	27.75 \pm 5.07
TLC ($10^3/\mu\text{L}$)	8.75 \pm 3.20	8.91 \pm 2.82
Neutrophils (%)	66.12 \pm 12.45	70.28 \pm 11.93
Lymphocytes (%)	32.66 \pm 13.79	28.75 \pm 12.26
Eosinophils (%)	2.66 \pm 1.82 A	1.50 \pm 1.03 B
Monocytes (%)	1.69 \pm 1.31	1.15 \pm 0.55

Values with different capital letters in a row are statistically significant at $P < 0.05$.

Table 3: Effect of birth weight on RBC, PCV, Hb, erythrocytic indices, TLC and DLC (Means \pm SD) on neonatal lambs.

Parameters	Birth Weight (Kg)		
	2-3	3-4	4 & above
RBC ($10^6/\mu\text{L}$)	7.71 \pm 0.98	7.30 \pm 1.19	7.77 \pm 0.97
PCV (%)	38.42 \pm 5.86	42.33 \pm 9.28	41.15 \pm 7.71
Hb (g/dL)	11.06 \pm 1.62	11.24 \pm 1.50	11.20 \pm 1.64
MCV (fl)	50.43 \pm 9.83	59.80 \pm 17.38	53.73 \pm 13.72
MCH (pg)	14.58 \pm 3.07	15.78 \pm 3.75	14.52 \pm 2.87
MCHC (%)	29.06 \pm 4.53	27.36 \pm 5.47	27.55 \pm 4.26
TLC ($10^3/\mu\text{L}$)	8.94 \pm 3.10	8.28 \pm 2.65	9.05 \pm 3.14
Neutrophils (%)	69.68 \pm 10.72	67.12 \pm 15.64	67.87 \pm 11.55
Lymphocytes (%)	29.14 \pm 11.45	31.24 \pm 16.51	31.39 \pm 12.42
Eosinophils (%)	1.66 \pm 1.00	2.75 \pm 2.12	1.72 \pm 1.27
Monocytes (%)	1.36 \pm 1.20	1.20 \pm 0.44	1.60 \pm 1.04

Table 4: Effect of parity on RBC, PCV, Hb, erythrocytic indices, TLC and DLC of neonatal lambs

Parity	Birth weight(Kg)	RBC ($10^6/\mu\text{L}$)	PCV (%)	Hb (g/dL)	
1	3.85±0.67	7.66±1.16	41.64±9.41	10.97±1.76	
2	3.84±0.53	7.61±1.14	40.87±6.19	11.13±1.41	
3	3.62±0.62	7.62±1.04	42.08±6.64	11.06±1.58	
4	3.11±0.60	7.70±1.02	41.45±9.20	11.56±1.86	
5	3.64±0.75	7.86±0.86	38.00±5.45	11.12±1.68	
6	3.71±0.70	7.55±0.99	39.80±8.72	11.26±1.80	
7	3.57±0.96	7.46±0.54	31.60±6.42	11.06±0.93	
	MCV (fl)	MCH (pg)	MCHC (%)		
1	55.81±17.38	14.69±4.01	27.05±5.29		
2	54.80±11.11	14.82±2.55	27.28±4.40		
3	56.10±11.89	14.69±3.02	26.42±3.05		
4	55.53±18.29	15.53±4.26	28.49±5.57		
5	48.63±9.54	14.33±3.04	29.65±3.63		
6	53.55±15.04	15.08±2.84	28.96±5.67		
7	48.04±10.30	14.88±1.98	31.46±2.98		
	TLC ($10^3/\mu\text{L}$)	Neutro. (%)	Lympho. (%)	Eosino. (%)	Mono. (%)
1	8.90±3.26	68.69±13.39	29.62±13.80	2.12±2.10	1.14±0.37
2	8.64±3.20	65.36±15.70	33.93±15.95	1.20±0.44	1.00±0.02
3	8.90±3.14	73.50±10.07	25.40±11.11	1.66±0.57	1.75±1.50
4	9.27±2.83	69.73±10.34	28.55±11.03	2.33±1.33	2.00±0.16
5	7.86±3.60	67.57±10.53	32.14±11.13	1.00±0.04	1.00±0.50
6	9.01±2.60	68.40±11.96	30.10±12.31	2.00±1.41	1.20±0.44
7	9.34±2.71	62.50±11.19	38.20±15.75	5.00±0.24	3.01±0.14

Table 5: Bacteria isolated from various tissues/organs of morbid and died neonatal lambs

Morbid Neonates				
S.No.	Sex	Disease	Sample/tissue	Organisms
1.	Male	Diarrhoea	Faecal	<i>E. coli</i> , Bacillus spp.
2.	Male	Diarrhoea	Faecal	<i>Pseudomonas</i> spp., <i>E. coli</i>
3.	Female	Diarrhoea	Faecal	<i>E. coli</i> , Proteus spp.
4.	Male	Pneumonia	Nasal/Throat	<i>E. coli</i>
5.	Male	Pneumonia	Nasal/Throat	Streptococci spp.
6.	Female	Pneumonia	Nasal/Throat	Proteus spp., <i>E. coli</i>
7.	Male	Diarrhoea	Faecal	<i>E. coli</i> , Bacillus spp.
8.	Female	Pneumonia	Nasal/Throat	Staphylococcus spp., Bacillus spp., <i>E. coli</i>
9.	Male	Pneumonia	Nasal/Throat	<i>E. coli</i> , Bacillus
Dead Neonates				
1.	Male	Diarrhoea	Intestine	<i>E. coli</i> , Bacillus spp.
2.	Female	Diarrhoea	Intestine	<i>E. coli</i>
3.	Male	Pneumonia	Lungs/Trachea	<i>E. coli</i> , Streptococci spp.
4.	Male	Pneumonia	Lungs/Trachea	<i>Pseudomonas</i> spp.
5.	Female	Pneumonia	Lungs/Trachea	<i>Pseudomonas</i>
6.	Female	Diarrhoea	Intestine	<i>Corynebacterium</i> , Streptococci spp.,
7.	Male	Pneumonia	Lungs/Trachea	Bacillus spp.

REFERENCES

- Adams, R., R.B. Gary, B.M. Aldridge and K.G. Oddle, 1992. Haematological values in newborn beef calves. *Am. J. Vet. Res.*, 53 (6):944-950.
- Anonymous, 1996. SPSS, 7.5.1. Release, Standard Version. Sterling Technologies, Inc., 444 N, Michigan Avenue, Chicago, IL 60611.
- Benjamin, M.M., 1979. Outline of Veterinary Clinical Pathology. 2nd ed., The Iowa State University Press, Ames, Iowa, USA.
- Chaarani, B., R.A. Robinson and D.W. Jonson, 1991. Lamb Mortality in Meknes Province (Morocco). *Preventive Vet. Med.*, 10: 283-289.
- Coles, E.H., 1988. Veterinary Clinical Pathology. 4th ed., W. B. Saunders Co., London.
- Cruikshank, R., J. P. Duguid, B.P. Marmion and R.H.A. Swain, 1974. Medical Microbiology, 12th ed. Vol. I. The English Language Book Society and Churchill Livingstone.
- Eales, F.A., J. Small, J.S. Gillmour, R.H. Armstrong and G.D. Gittus 1986. A simple system for recording lamb mortality used to improve flock management. *Vet. Rec.*, 118 (9): 22-230.
- Janke, B.H., D.H. Francis, J.E. Collins, M.C. Libal, D.H. Zeman, D.D. Johnson and R.D. Neiger, 1990. Attaching and effecting E.coli infection as a cause of diarrhoea in young calf. *J. Am. Vet. Med. Assoc.*, 196: 897-901.
- Khan, A. and M.Z. Khan, 1996. Neonates calf mortality in Pakistan. III. Immunoglobulin in relation to mortality in buffalo and cow neonates. *Buffalo J.*, 12 (2): 243-252.
- Mason, R.W. and A. Corbould, 1981. Colisepticaemia of lambs. *Aut. Vet. J.*, 57 (10): 458-460.
- Radostits, O.M., 1986. Neonatal diarrhoea in ruminants. In: Current Veterinary Therapy, 2. Food Animal Practice. (Howard, J.L., Ed.). W.B. Saunders Company, Philadelphia, pp: 105-112.
- Sawyer, M., C. H. Willadsen, Osburn and T.C. McGuire, 1977. Passive transfer of colostral immunoglobulins from the ewes to lamb and its influence on neonatal lamb mortality. *J. Am. Vet. Med. Assoc.*, 171 (12): 1255-1259.
- Snodgrass, D.R. and E.W. Gray, 1977. Detection and transmission of 30 nm virus particle (Astrovirus) in faeces of lamb with diarrhoea. *Archiv Virology*, 55: 287-291.