INCIDENCE AND THERAPEUTIC MANAGEMENT OF *Eimeria species* INFECTION IN MURRAH BUFFALO CALVES

Siddhant Bendigeri¹, Rupesh Verma^{1,*}, Giridhari Das¹, Suman Kumar¹, Arun Mourya² and Rupanjali Saiyam¹

Received: 06 July 2020 Accepted: 27 June 2023

ABSTRACT

The present study was undertaken to examine the incidence of *Eimeria* species infection in Murrah buffalo calves and evaluate the therapeutic management of the disease. In November 2019, 9 to 10 buffaloes calves up to 3 months of age showed symptoms of diarrhoea, loss of appetite, and poor weight gain at an organised dairy farm. Based on history, faecal samples were collected from 20 buffaloes calves and subjected to standard qualitative coprological examination and quantitative analysis using a modified McMaster technique. Out of 20 faecal samples, 90% were positive for coccidia oocysts. Among positive samples, 11% were heavily infected and 16% had a moderate level of infection of Eimeria spp. Six species of Eimeria were identified, E. bareillyi was the most often encountered species, accounting for 37.39%, followed by E. bovis (22.76%), E. zuernii (14.63%), E. ellipsoidalis (13.82%), E. canadensis (13.82%), and E. cylindrica (6.50%). Blood examination of mild and severely affected calves revealed a decrease in the mean values of hemoglobulin, total erythrocyte count, total leucocyte count, platelets and an increase in monocytes as compared with healthy calves. The affected calves were treated with combination of Sulphadiazine and Trimethoprim (Biotrim IV injection) along with supportive treatment for five successive days. All the affected animals responded well to the treatment and showed marked reduction in clinical symptoms as well as oocyst output.

Keywords: *Bubalus bubalis*, buffaloes, buffalo calves, oocyst per gram, *Eimeria* spp., haematological alterations, therapeutic management

INTRODUCTION

Animal husbandry plays a vital role in the rural economy of India, and buffalo (*Bubalus bubalis*) farming is a crucial component of this industry. According to the 20th All India Livestock Census, there are over 109 million buffaloes in the country, contributing more than 56% of total milk production. Maximizing the profitability of dairy production relies on efficient and successful

¹Department of Veterinary Parasitology, College of Veterinary Science and Animal Husbandry, Nanaji Deshmukh Veterinary Science University, Madhya Pradesh, India, *E- mail: vrupesh77@gmail.com ²Department of Veterinary Medicine, College of Veterinary Science and Animal Husbandry, Nanaji Deshmukh Veterinary Science University, Madhya Pradesh, India growth of calves to breeding age. However, newborn calves are highly vulnerable to infections and diseases due to their low immunity (Eğlenti et al., 2020), which can result in neonatal mortality and/or suboptimal growth. Among different calf diseases in the buffalo population, coccidiosis is a leading cause of diarrhoea, dehydration, anorexia, reduced body weight, anaemia, and increased susceptibility to other diseases, including death (Coetzer and Justin, 2004; Saravanan et al., 2017; Jayalakshmi et al., 2018). The disease is caused by various species of the apicomplexan protozoan Eimeria, which invade and destroy cells lining the intestinal tract of the host, leading to intestinal damage and potential loss of production (Soulsby, 1982). The infection is disseminated by the consumption of sporulated oocysts deposited in contaminated feed, water, and surfaces in animal dwelling areas (Taylor and Catchpole, 1994). The disease is more prevalent in animals kept in confinement, such as those raised under intensive farming conditions, and is associated with poor hygiene, overcrowding, and other stress conditions like weaning or changes in feed (Abede et al., 2008). The magnitude of infection depends on the amount of sporulated oocysts consumed and is more common in calves between 3 to 6 months old (Verma et al., 2018). Therefore, the present investigation aims to determine the incidence of *Eimeria* species infection in Murrah buffalo calves and evaluate the therapeutic management of the disease

MATERIALS AND METHODS

In November 2019, 9 to 10 buffaloes calves up to 3 months of age exhibited symptoms of diarrhoea, loss of appetite, and poor weight gain at an organised dairy farm located at Adhartal, Nanaji Deshmukh Veterinary Science University (NDVSU), Jabalpur (latitude 23° 10' N and longitude 79° 56' E), Madhya Pradesh, India. To investigate the possible causes of these symptoms, fresh faecal samples were collected from 20 buffaloes calves and transported to the Department of Veterinary Parasitology, College of Veterinary Science and Animal Husbandry, Jabalpur and stored 4°C for further analysis. In addition, blood samples were also collected from affected animals for haematological examination. Qualitative examination of each faecal sample was performed using the floatation method to evaluate the incidence of infections. Coccidial oocysts per gram (OPG) of faeces were quantified using the modified McMaster technique (Soulsby, 1982; Sloss et al., 1994). Faecal samples that tested positive were then subjected to sporulation in a 2.5% potassium dichromate solution, as described by Verma et al. (2018). The identification of Eimeria species was carried out based on the key described by Soulsby (1982); Levine (1985); Duszynski and Wilber (1997).

Statistical analysis

Data analysis was performed in Microsoft Excel spreadsheet program version 2010 (Microsoft, Redmond WA, USA) to derive the arithmetic means, minimum and maximum values, and prevalence expressed as a percentage.

RESULTS AND DISCUSSIONS

In the present study, out of 20 faecal samples, 90% were positive for coccidia oocysts. Among positive samples, 11% were heavily infected (OPG >14000) and 16% had a moderate

level of infection (OPG >3000) of Eimeria spp. Our results are consistent with previous studies (Dubey, 2018; Maurya et al., 2020) who stated that the Eimeria infection is more damning to younger animals than adults and infection decrease with growing age due to immunological maturity acquired after repeated exposure. Prevalence of coccidiosis depends upon the age and immune status of animals, climatic conditions, and farm management. The affected buffalo calves exhibited clinical signs such as inappetence, weakness, rough body coat, dehydration, dry muzzle, sunken eyeballs, pale and congested conjunctival mucous membrane, foul-smelling diarrhoea, and smudging of the perineum and tail with faeces. The animals also showed reluctance to move and lack of response to environmental stimuli.

Clinical examination revealed subnormal temperature, increased pulse, and respiration rates. These findings are consistent with previous studies conducted by Saravanan et al. (2017); Gopalakrishnan et al. (2017); Jayalakshmi et al. (2018). In bovine *Eimeria* infection, the schizonts (2nd and/or 3rd generations) and gametogony stages grow deep into the mucosa, which can cause sloughing of the epithelium, associated haemorrhage, and tissue destruction. However, haemorrhagic diarrhoea (dysentery) was not observed in the current finding as reported by Mundt et al. (2005); Verma et al. (2018) in cattle (<3 month) which might be due to Eimeria zuernii infection. Under natural conditions, the disease with single species of *Eimeria* is rare and mixed infections are more common. The present study identified six Eimeria species based on their morphological and micrometric features, as detailed in Table 1. Species identification was also determined based on sporulation time. The prevalence of each Eimeria species was assessed,

with E. bareillyi (37.39%) being the most often detected and dominating species. This was followed by E. bovis (22.76%), E. zuernii (14.63%), E. ellipsoidalis (13.82%), E. canadensis (13.82%), and E. cylindrica (6.50%) (Figure 1). Several Eimeria species that were originally described in cattle have been reported in water buffaloes (Dubey, 2018). Among these, E. bareillyi is a buffalo-specific species that is non-transmissible to cattle and is frequently reported as the most pathogenic species in India and other parts of the world (Sanyal et al., 1985; Dubey et al., 2008; Maurya et al., 2020). In our study, we also found that E. bovis and E. zuernii were among the most prevalent species (22.76%) and 14.63%, respectively). While these species are known to be highly pathogenic to cattle, clinical coccidiosis associated with these species has also been reported in buffaloes worldwide (Bahrami and Alborzi, 2013; El-Alfy et al., 2019).

The examination of blood samples from both mildly and severely affected animals revealed a significant decrease in the mean value of haemoglobin, total erythrocyte count, total leukocyte count, and platelets, as well as an increase in monocytes, when compared to apparently healthy calves (Table 2). These findings are consistent with previous studies by Anwar et al. (1999); Jayalakshmi et al. (2018). The observed variations in haematological parameters in the present study may be attributed to the intensity of infection, immune status of the animal, and the specific species of Eimeria involved in the infection. In the present study, clinically affected animals were treated with Biotrim IV injection (Zydus AHL), a combination of Sulphadiazine BP (Vet) 200 mg and Trimethoprim IP 40 mg, at a dosage of 1 ml per 10 kg body weight twice a day for three consecutive days. To prevent dehydration and electrolyte imbalance, intravenous

			Long	jitudinal and h	Longitudinal and horizontal diameter	er	
S.No.	S.No. <i>Eimeria</i> species	Prevalence	Length (Max-Min)	Average length	Width (Max-Min)	Average width	Sporulation time
1	E. bareillyi	37.39%	32.82-27.66	30.33	23.06-17.80	20.62	3-4
2	E. bovis	22.76%	30.59-25.24	28.49	22.82-15.27	20.08	2-4
ю	E. zuernii	14.63%	21.45-19.66	21.45	21.45-16.87	19.66	3-4
4	E. ellipsoidalis	13.82%	28.40-22.50	25.15	23.65-19.20	20.62	3-5
5	E. canadensis	6.50%	33.50-30.39	31.5	20.88-19.04	20.88	3-5

Table 1. Morpho-metric identification and prevalence and of Eimeria species.

Table 2. Haematological parameters in healthy, diseased and mild infected calves as observed.

3.100.	Dougan of our	Healthy calve	Mild infected calves	DISEASE CALVES
1	rarameters	(Mean±SE); (N=5); (OPG = O)	(Mean±SE); (N=3); (OPG ≥3000)	$ \pm SE ; (N=5); (OPG = O) (Mean\pm SE); (N=3); (OPG \ge 3000) (Mean\pm SE); (N=2); (OPG \ge 14000) (Mean\pm SE); (Mean\pm SE$
	WBC count (×10 ³ / μ 1)	14.40 ± 1.50	12.67 ± 0.66	9.50 ± 2.50
2	RBC count (×10 ⁶ / μ l)	$7.80{\pm}1.15$	8.33 ± 0.88	6.00 ± 1.00
3	Haemoglobin (gm/dl)	$9.40{\pm}1.77$	8.33±0.88	6.50 ± 0.50
4	PCV (%)	29.60±5.60	31.33 ± 2.84	$23.00{\pm}1.00$
5	MCV (fl)	37.20±1.98	39.67±3.28	$37.00{\pm}7.00$
9	MCH (pg)	$12.00 {\pm} 0.54$	12.67 ± 0.66	$13.00{\pm}2.00$
7	MCHC (g/dl)	$31.60{\pm}0.40$	33.67 ± 0.33	$35.00{\pm}1.00$
8	Platelets (×10 ⁵ / μ l)	$4.40{\pm}1.07$	1.00 ± 0.00	1.00 ± 0.00
6	Lymphocytes (%)	26.80±5.43	29.33 ± 5.54	$23.50{\pm}1.50$
10	Monocytes (%)	$3.80{\pm}0.86$	5.00 ± 0.00	$7.00{\pm}2.00$
11	Granulocytes (%)	69.40±5.22	65.67±5.54	69.50 ± 3.50

2-6

14.36

18.12-12.48

25.84

30.49-22.96

4.87%

E. cylindrica

9



Figure 1. Microscopic observation of *Eimeria* species oocysts before and after sporulation.

- a. E. bareillyi (unsporulated); b. E. bareillyi (sporulated); c. E. bovis (unsporulated);
- d. E. bovis (sporulated); e. E. canadensis (unsporulated); f. E. canadensis (sporulated);
- g. E. cylindrica(sporulated); h. E. ellipsoidalis (unsporulated); i. E. ellipsoidalis (sporulated);
- j. Eimeria zuernii (sporulated); k. Eimeria zuernii (unsporulated).

administration of dextrose normal saline (DNS) at 250 to 300 ml and Ringer's lactate (RL) at 250 to 300 ml was performed, along with a multivitamin preparation (MVI) at 2 ml. Mildly affected calves were treated with Biotrim DS Bolus (Zydus AHL), which contains Sulphadiazine BP (Vet) 2000 mg and Trimethoprim IP 400 mg per bolus, at a dosage of 1/2 bolus per 50 kg body weight orally twice a day for three days to control infection. Affected animals showed marked improvement after posttreatment and there was a marked reduction in oocyst output. Sulpha drugs such as sulphadiazine, sulphadimidine and sulphaquinoxaline have been shown to have potent anti-coccidial activity against a variety of Eimeria species and are therefore widely used to treat acute coccidiosis in calves (Chakrabarti and Jha, 2016; Saravanan et al., 2017; Gopalakrishnan et al., 2018; Maurya et al., 2020). These drugs act by inhibiting the synthesis

of folic acid, which is essential for the growth and development of *Eimeria parasites*. However, the emergence of drug-resistant strains of *Eimeria* has become a growing concern, and there is a need for alternative treatment strategies to control the disease.

CONCLUSION

The results of this study demonstrate a high incidence of *Eimeria* species infection in buffalo calves, with *E. bareillyi* being the most identified species. The findings also highlight the potential impact of the infection on the health and growth of the animals, as evidenced by the decrease in blood parameters. However, the successful therapeutic management of the infected animals with a combination of Sulphadiazine and Trimethoprim provides valuable information for the treatment and prevention of the disease. This investigation underscores the importance of regular monitoring, proper hygiene, and appropriate management practices to minimize the spread of *Eimeria* species infection in buffalo calves and optimize the productivity of the dairy industry.

REFERENCES

- Abede, R., A. Wossene and B. Kumsa. 2008.
 Epidemiology of *Eimeria* infections in calves in Addis Ababa and Debre Zeit dairy farms, Ethiopia. *Int. J. App. Res. Vet. M.*, 6(1): 24-30.
- Anwar, A.H., S.I. Kazmi and M.N. Khan. 1999. Effect of experimentally induced coccidiosis on some blood parameters of buffalo calves. *Pakistan Journal of Biological Sciences*, 2(3): 1024-1026. DOI: 10.3923/ pjbs.1999.1024.1026
- Bahrami, S. and A.R. Alborzi. 2013. Prevalence of subclinical coccidiosis in river buffalo calves of southwest of Iran. *Acta Parasitol.*, 58(4): 527-30. DOI: 10.2478/s11686-013-0167-1
- Chakrabarti, A. and B.K. Jha. 2016. Winter coccidiosis in a calf - A case report. *International Journal of Agricultural Science and Research*, 6(1): 279-82.
- Coetzer, J. and R. Justin. 2004. *Infectious Diseases* of *Livestock*, 2nd ed. Oxford University Press, Cape Town, South Africa.
- Dubey, J.P. 2018. A review of coccidiosis in water buffaloes (*Bubalus bubalis*). Vet. Parasitol., **256**(30): 50-57. DOI: 10.1016/j. vetpar.2018.04.005
- Duszynski, D.W. and P.G. Wilber. 1997. A guideline for the preparation of species descriptions

in the Eimeriidae. *J. Parasitol.*, **83**(2): 333-336. DOI: 10.2307/3284470

- Eğlenti, N., S. Kozat and V. Denizhan. 2020. Investigation of immunoglobulin (IgE, IgA, IgG, IgM) concentrations in calves naturally infected with coccidiosis. *Journal* of Istanbul Veterinary Sciences, **41**(1): 1-7. DOI: 10.30704/http-www-jivs-net.691671
- El-Alfy, E., I.E. Abbas, Y. Al-Kappany, M. Al-Araby, S.A. Abu-Elwafa and J.P. Dubey.
 2019. Prevalence of *Eimeria* Species in water buffaloes (*Bubalus bubalis*) from Egypt and first report of *Eimeria bareillyi* oocysts. *J. Parasitol.*, **105**(5): 748-754. DOI: 10.1645/19-58
- Gopalakrishnan, A., U. Dimri, V. Joshi, V.R. Kundave, Y. Ajith and M.I. Yatoo. 2017. A clinically rare occurrence of rectal mucosal prolapse associated with tenesmus in a calf caused by *Eimeria* sp. *Journal of Parasitic Diseases*, 41(3): 723-725. DOI: 10.1007/ s12639-016-0877-z
- Jayalakshmi, K., M. Sasikala, B. Dhivya, S. Yogeshpriya, M. Veeraselvam, S. Krishnakumar and P. Selvaraj. 2018. Winter Coccidiosis in a buffalo calf. *Indian Vet. J.*, 95(1): 75-76.
- Levine, N.D. 1985. *Veterinary Protozoology*. Iowa State University Press, Ames, Iowa, USA. p. 150-202.
- Maurya, P.S., S. Sahu, S. Rawat and V. Jaiswal. 2020. Simultaneous Infection in a buffalo calf with *Toxocara vitulorum* and *Eimeria bareillyi*: A case report. *Research Journal for Veterinary Practitioners*, 8(1): 1-3. DOI: 10.17582/journal.rjvp/2020/8.1.1.3
- Mundt, H.C., B. Bangoura, M. Rinke, M. Rosenbruch and A. Daugschies. 2005. Pathology and treatment of *Eimeria zuernii*

coccidiosis in calves: investigations in an infection model. *Parasitol. Int.*, **54**(4): 223-230. DOI: 10.1016/j.parint.2005.06.003

- Sanyal, P.K., N.S. Ruprah and M.B. Chhabra. 1985. Attempted transmission of three species of *Eimeria* Schneider, 1875 of buffalo-calves to cow-calves. *Indian J. Anim. Sci.*, 55: 301-304.
- Saravanan, S., G. Ponnudurai and K.M. Palanivel. 2017. Haemorrhagic enteritis due to mixed coccidian infection in a young Murrah calf. *Trends Biotechnol.*, **10**:1466-1468.
- Sloss, M.W., R.L. Kemp. and A.M. Zajac. 1994. Veterinary Clinical Parasitology, 6th ed. Iowa State University Press, Iowa, USA.
- Soulsby, E.J.L. 1982. *Helminths, Arthopods and Protozoa of domesticated Animals*, 7th ed. Bailliere and Tindall, London, UK.
- Taylor, M.A. and J. Catchpole. 1994. Coccidiosis of domestic ruminants. *Applied Parasitology*, 35: 73-86.
- Verma, R., G. Das, R. Saiyam and S. Bendigeri. 2018. Clinical coccidiosis in calves and its treatment. *Journal of Entomology* and Zoology Studies, 6: 2964-2967. DOI: 10.13140/RG.2.2.17932.62088