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Prevalence of *Cryptosporidium* and *Eimeria* Protozoa in Large Ruminants of Marathwada Region of Maharashtra State

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ABSTRACT

To appraise the occurrence of *Cryptosporidium* and *Eimeria* in cattle and buffalo calves below 3 months of age, a total of 407 faecal samples (buffalo, 136 and cow, 271) were collected from the Parbhani region of Maharashtra state. For detection of *Cryptosporidium* oocysts, Modified Ziehl-Neelsen staining method, while for *Eimeria* oocysts fecal floatation technique with saturated salt solution was employed. The prevalence of *Cryptosporidium* was 3.67% in buffalo calves and 4.79% in cow calves. Whereas overall prevalence of *Eimeria* spp. was 30.14% and 30.62% in buffalo and cow calves, respectively. The seasonal prevalence showed higher level of infection of both *Cryptosporidium* and *Eimeria* in both buffalo and cattle claves during summer season hence care during this season is important. Sex wise prevalence shows higher prevalence of *Cryptosporidium* and *Eimeria* in female calves, of both the host. The occurrence of *Cryptosporidium* oocysts was higher predominantly during 0-15 days in buffalo calves and 16-30 days in cattle calves. The prevalence of *Eimeria* was higher in 16-30 days buffalo calves and 1-15 days in cow calves. Breed-wise prevalence was higher in non-descript buffalo calves and cow calves, needs attention of these calves as they may also be source of infection for other breed calves. Management as regards weaning and confined housing have shown considerable effect on the prevalence of both the infections in calves.

HIGHLIGHTS

- *Cryptosporidium* and *Eimeira* are prevalent in calves of large ruminants in Marathwada.
- Age has been found a significant risk factor for these infections.
- However, sex, breed and management practices were non-significant risk factors.

Keywords: *Cryptosporidium*, *Eimeria*, young calves, weaning, housing management

Two apicomplexan parasites *viz. Cryptosporidium* and *Eimeria,* with certain common characteristics such as causing intestinal tract infection in cattle and buffalo calves, are responsible for huge losses in terms of morbidity and mortality (Hatam-Nahavandi *et al*., 2019, Sultana *et al*., 2020). Both these parasites are among the most common and important entero-pathogens in calves (Delling and Daugschies, 2022). *Cryptosporidium* spp. is an intracellular but extra-cytoplasmic, tissue dwelling protozoan parasite which localizes in the gastro-intestinal tract of vertebrates including human, domestic and wild animals and also birds (Sweeny *et al.,* 2012). *Cryptosporidium* infection is well known as a major cause of morbidity and mortality particularly in immune-compromised hosts and young animals (Ryan *et al*., 2014; Thomson *et al*., 2017). However, very few published reports of cryptosporidiosis in animals are available from India (Venu *et al*., 2012; Jyothimol and Ravindran, 2015;

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Hingole *et al*., 2017). *Cryptosporidium* spp. is presently gaining attention as an important pathogen mainly due to its dominant involvement in worldwide water borne outbreaks (Pollock *et al*., 2014). Among the wide spectrum of hosts, cattle is considered to be a primary reservoir host for *Cryptosporidium* oocysts (Jokar *et al*., 2021). In infected animals, clinical Cryptosporidiosis is commonly associated with severe enteritis characterized by acute, watery, or steatorrheic diarrhea and colic while asymptomatic infections are common (Mitchell *et al.,* 2012). Upon recovery of infected animals, acts as carriers and remains main source of infection to the susceptible individuals (Bangoura and Daugschies, 2019).

Another apicomplexan parasite *Eimeria* spp. Which cause coccidiosis is one of the most common and important disease of cattle worldwide. Bovine coccidiosis has been reported widely and is usually most common and important in calves younger than 1 year (Thomson *et al*., 2017). Calves reared under conventional management systems are prone to exposure and become infected early in life. Several earlier reports indicated that under natural conditions, mixed species infections are much more common than mono-species coccidiosis (Bangoura and Daugschies, 2019). Among more than 20 species of *Eimeria* identified in cattle worldwide *E. bovis, E. zuernii* and *E. auburnensis* were recorded as most commonly prevalent and the two species *i.e. Eimeria bovis* and *E. zuernii* are considered to be the most pathogenic species of coccidiosis in cattle and buffaloes, responsible for outbreaks of clinical coccidiosis in calves (Dubey *et al*., 2018). *Eimeria spp.* develop only in the intestinal epithelial cells, causing mucosal damage and nutrient losses which lead to malnutrition, weakness, anemia, diarrhea and haemorrhagic faeces (Nisar-Khan *et al*., 2013). Important clinical signs observed are thin watery diarrhea, occasionally containing blood during clinical *Eimeria* spp. infections, while during sub-clinical chronic infections obvious signs are fatigue, fever, and reduced appetite (Keeton and Navarre, 2018). Not only the clinical from of coccidiosis with its severe health consequences but also its sub-clinical form in terms of huge economic losses due to reduced growth and productivity are considered hindrance in successful dairy enterprises worldwide (de Souza Rodrigues *et al*., 2017).

Although the infections with both these protozoan parasites lead to fewer deaths, serious economic losses can occur due to significant morbidity amongst calves, reduced production potential and the costs involved in the treatment of infected animals (Nisar-Khan *et al*., 2013). Epidemiological survey of parasitic diseases facilitates in formulating and adopting effective control measures is an important tool in controlling losses. Hence it was advised to periodically conduct and report the epidemiological picture of important diseases across the area (Nain *et al*., 2017). With this principle several reports on prevalence and seasonal variation in the intensity of parasites of cattle and buffaloes has been observed in different parts of the country over the period of time (Bhat *et al*., 2013; Gupta *et al*., 2016; Das *et al*., 2015; Bhangale, 2020). However in view of the very few reports from the state of Maharashtra, current study was conducted in Marathwada region of the state.

MATERIALS AND METHODS

Investigations were conducted to study the prevalence of *Cryptosporidium* spp. and *Eimeria* spp. in cattle and buffalo calves ageing up to 3 months along with its correlation with season, sex and breed of cattle and buffalo calves in and around Parbhani region during 2017-18.

Location

The research work was undertaken at Department of Veterinary Parasitology and Department of Animal Biotechnology, College of Veterinary and Animal Sciences, (MAFSU) Parbhani.

Parbhani is situated at 19^{o} 16’ North latitude 76^{0} 47’ East longitudes and at an altitude of 423.46 m above mean sea level in Marathwada division of Maharashtra state. Agro climatically this region falls in assured rainfall zone of Maharashtra state. The mean average annual precipitation is about 742 mm, mostly received between June to September from south-west monsoon with uncertain and scanty winter rains. The mean maximum temperature varies from 28.62^{o}C in winter (December) to about 40.9^{o}C in summer (May), whereas the mean minimum temperature varies from 12.1^{o}C to 24.9^{o}C during winter and summer respectively. Thus, Parbhani has cold winter and dry hot summer. The mean minimum and maximum relative humidity varies between 25 to 63 and 85 to 96 percent respectively.

The variables selected for recording the prevalence of *Cryptosporidium* and *Eimeria* were season, age, sex and breed along with some management practices. Summer, winter and monsoon seasons defined as per WMO norms for the Marathwada region were considered. Young cattle and buffalo calves up to 3 months of age were included in the study. The common breeds of cattle in this region are Red Kandhari, Holstein Friesian and Non-descript while those of buffalo are Marathwadi and Non-descript only. In addition to these variables, management practices of young claves were also included in the current study to study their impact on the prevalence of these two important intestinal protozoa.

Collection of faecal samples

Faecal samples were collected directly from calf's rectum in a separate clean labelled container. Formed as well as diarrheic samples were collected in quantity of about 5 grams with the help of well lubricated plastic hand gloves. Calves from Instructional Livestock farm complex of COVAS Parbhani, Cross breeding farm of VNMKV, Parbhani as well as calves from adjoining villages were included in this study. Faecal samples were then transferred into glass vials/plastic vials, labelled and were brought to laboratory of Dept. of Parasitology, College of Veterinary and Animal Sciences, Parbhani and processed further.

Processing of fecal samples

The faecal samples were first examined by direct smear method and then were further processed for the presence or absence of the coccidia oocysts by saturated salt Floatation Technique. For detection of *Cryptosporidium* spp. oocysts thin faecal smears were prepared, fixed with methanol spirit and were stained with Modified Ziehl-Neelsen Stain (MZN).

Totally 407 samples of buffalo (136) and cow calves (271) were examined for the presence of *Eimeria* spp. And *Cryptosporidium* spp. Oocysts. The data regarding each calf including identification (farmer's name, location) and other description (age, sex, breed, weaned or un-weaned, whether belongs to organized or unorganized farm), was gathered during sampling.

The prevalence of coccidiosis was determined by using the formula:

Prevalence (%) =

$$\left[\frac{\text{Number of infected individuals (p)}}{\text{Total number of samples examined (n)}}\right]\times 100$$

STATISTICAL ANALYSIS

The data obtained from various parameters were analysed by employing Chi-Square test and significance was presented as non-significant, significant (at 5%) and highly significant (at 1%).

RESULTS AND DISCUSSION

On examination of 407 samples from both the hosts, the prevalence of important intestinal protozoa *viz*. *Cryptosporidium* spp. and *Eimeria* spp. was observed as 4.42% and 30.46%, respectively. The prevalence for *Cryptosporidium* was 3.67% in buffalo calves and 4.79 % in cow calves. However the overall prevalence of *Eimeria* spp. was 30.14 % and 30.62 % in buffalo and cow calves, respectively (Table 1).

The seasonal prevalence noted in both the host species was non- significant amongst the three seasons. In buffaloes the per cent prevalence was 0.00 and 5.95, 19.35 and 34.52, 22.58 and 26.19 for the *Cryptosporidium spp.* and *Eimeria* spp. infections during monsoon, winter and summer seasons, respectively. However, for cow calves, seasonal prevalence was 1.96 and 0.00, 11.76 and 50.00, 13.72 and 18.18 for the *Cryptosporidium* spp*, Eimeria* spp. infections during monsoon, winter and summer seasons, respectively (Table 2).

In buffaloes prevalence of *Cryptosporidium* was higher in males while that of *Eimeria* was higher in female calves than males. The prevalence noted in male buffalo calves was 8.47 and 28.81 per cent, while in female buffalo calves it was 0.00 and 31.16 per cent for *Cryptosporidium, Eimeria* infection, respectively. In cow calves, the infections of *Cryptosporidium* and *Eimeria* showed non-significant difference between males and females, though the magnitude of infections was higher in females as compared to males (Table 3).

There found non-significant variation among the different age groups *i.e.* both the parasites were equally distributed and infects to all age groups up to 90 days of age. In case

Table 1: Overall prevalence of gastrointestinal protozoa in cattle and buffaloes

Sl. No.	Host	Total examined	Prevalence of *Cryptosporidium* p(%)	Positive for *Eimeria* p(%)
1	Buffalo calves	136	05 (3.67)	41 (30.14)
2	Cattle calves	271	13 (4.79)	83 (30.62)
	Total	407	18 (4.42)	124 (30.46)
	Significance		Non-significant	Non-Significant

Table 2: Seasonal prevalence of *Cryptosporidium* spp., *Eimeria* spp. and other GI parasites in buffalo and cow calves

Season	Buffalo calves			Cattle calves		
	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeria*	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeria*
Monsoon	31	00 (0)	6 (19.35)	51	1 (1.96)	6 (11.76)
Winter	84	05 (5.95)	29 (34.52)	44	0 (0)	22 (50.00)
Summer	21	00 (0)	6 (28.57)	176	12 (6.81)	55 (31.25)
Total	136	05 (3.67)	41 (30.14)	271	13 (4.79)	83 (30.62)
Significance		Non –significant	Non –significant		Non –significant	Significant

Table 3: Sex-wise prevalence of *Cryptosporidium* spp., *Eimeria* spp. and other GI parasites in buffalo and cow calves

Sex	Buffalo calves			Cattle calves		
	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeira*	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeira*
Male	59	5 (8.47)	17 (28.81)	124	4 (3.22)	36 (29.03)
Female	77	0 (0)	24 (31.16)	147	9 (6.12)	47 (31.97)
Total	136	5 (3.67)	41 (30.14)	271	13 (4.79)	83 (30.62)
Significance		Significant	Non-significant		Non –significant	Non-significant

Table 4: Age-wise prevalence of *Cryptosporidium* spp., *Eimeria* spp. and other GI parasites in buffalo and cattle calves

Age group	Buffalo calves			Cattle calves		
	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeria*	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeria*
1-15 days	22	2 (9.09)	9 (40.9)	52	1 (1.93)	18 (34.61)
16-30 days	41	3 (7.37)	17 (41.46)	98	12 (12.24)	41 (14.28)
31-60 days	32	0 (0)	8 (25.00)	57	0 (0)	14 (24.56)
61-90 days	41	0 (0)	7 (17.07)	64	0 (0)	10 (15.62)
Total	136	05 (3.67)	41 (30.14)	271	13 (4.79)	83 (30.62)
Significance		Non-Significant	Non-Significant		Highly-Significant	Highly-Significant

of *Cryptosporidium,* though non-significant variation was observed, but its prevalence was noted higher in 1-15 days and followed in 16-30 days age group. Similarly the prevalence of *Eimeria* spp. was noted higher in the 1-30 days age groups compared with 30 days and above age group. However, in cow calves, significantly higher prevalence of *Cryptosporidium* was recorded in the age group of 16-30 days (12.24%) and the prevalence of *Eimeria* was significantly higher in 0-15 days (34.61%). From the data of the present study, it can be assessed that *Cryptosporidium* and *Eimeria* are the parasites of age group 1 month (Table 4).

As far as distribution of *Cryptosporidium* amongst breeds of cattle is concerned, only non-descript buffalo calves were found positive (4.62%) and no occurrence in Marathwadi buffalo calves. Similar type of trend was observed for *Eimeria* infection in which prevalence was on higher side in non-descript buffalo calves as compared to Marathwadi buffalo calves. Among 3 breeds of cow under study *i.e.* Holstein Friesen, Non-Descript and Red Kandhari, it was found that infections with both *Cryptosporidium* and *Eimeria* were *at par* among the 3 breeds, still the prevalence was comparatively lower in Red Kandhari breed. Thus, from the data it can be assessed that most vulnerable breed for the all the type of infection was Non Discript cattle (Table 5).

The C*ryptosporidium* infection was higher (3.93%) in un-weaned buffalo calves as compared to weaned calves (0.00%). Similarly prevalence of *Eimeria* was higher (31.49%) in un-weaned buffalo calves compared to weaned calves (11.11%). No differences were noted in the prevalence pattern of *Cryptosporidium* and *Eimeria* infection in the weaned and un-weaned cow calves. The percent prevalence noted was 4.34 and 8.69 per cents in weaned cow calves and 4.83 and 7.25 per cents in un-weaned cow calves, respectively (Table 6).

The prevalence of both types of protozoan infections in buffalo calves showed no significant variation among the organized and un-organized farms. The *Cryptosporidium* spp. Infection was found null on organized farms (0.00%), while *Eimeria* infection was also comparatively lower in buffalo calves on organized farms as compared to un-organized farms. Cow calves from organized farms were found infected with *Cryptosporidium* to the extent of 7.27%, which was higher than that recorded from un-organized farms *i.e.* 3.44 per cent. However there was no substantial difference in infection rate of *Eimeria* spp. among cow calves from both organized (25.45%) and un-organized farms (26.43%) respectively (Table 7).

Earlier Hingole *et al.* (2017) reported higher prevalence rates of Cryptosporidium in cow and buffalo calves from Mumbai as 34.48% in cow calves and 38.56% in buffalo calves. Reports from elsewhere in the country also reported higher prevalence of cryptosporidium in cow calves than buffalo calves which corroborate with the findings of present study (Venu *et al*., 2012, Daniels

Table 5: Breed-wise prevalence of *Cryptosporidium* spp., *Eimeria* spp. and other GI parasites in buffalo calves

Buffalo calves				Cattle calves			
Breed	**Total examined**	**Positive for *Cryptosporidium***	**Positive for *Eimeria***	**Breed**	**Total examined**	**Positive for *Cryptosporidium***	**Positive for *Eimeria***
Marathwadi	28	0 (0)	6 (21.42)	HFxD (CB)	19	1 (5.26)	1 (5.26)
Non-descript (ND)	108	5 (4.62)	35 (32.40)	Non-descript (ND)	173	11 (6.35)	58 ()33.52
Total	136	05 (3.67)	41 (30.14)	RK (Red Kandhari)	79	1 (1.26)	24 (30.37)
				Total	271	13 (4.79)	83 (30.62)
Significance		Non-Significant	Non-Significant			Non-significant	Non-significant

Table 6: Prevalence of *Cryptosporidium* spp., *Eimeria* spp. and other GI parasites in buffalo calves (Management type 1-weaned/ unweaned)

Management type I	Buffalo calves			Cattle calves		
	Total examined	**Positive for *Cryptosporidium***	**Positive for *Eimeria***	**Total examined**	**Positive for *Cryptosporidium***	**Positive for *Eimeria***
Weaned	9	0 (0)	1 (11.11)	23	1 (4.34)	2 (8.69)
Un-weaned	127	5 (3.93)	40 (31.49)	248	12 (4.83)	81 (32.66)
Total	136	05 (3.67)	41 (30.14)	271	13 (4.79)	83 (30.62)
Significance		Non-significant	Non-significant		Non-significant	Significant

Table 7: Prevalence of *Cryptosporidium* spp., *Eimeria* spp. and other GI parasites in buffalo calves (Management type II-organized/ un-organized)

Management type II	Buffalo calves			Cattle calves		
	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeria*	Total examined	Positive for *Cryptosporidium*	Positive for *Eimeria*
Organized farm	17	0 (0)	3 (17.64)	55	4 (7.27)	14 (25.45)
Un-organized farm	119	5 (4.20)	38 (31.93)	216	9 (3.44)	69 (31.94)
Total	136	05 (3.67)	41 (30.14)	271	13 (4.79)	83 (30.62)
Significance		Non-significant	Non-significant		Non-significant	Non-Significant

et al., 2015). Contrarily, trend of higher prevalence rate of cryptosporidiosis in buffaloes than cattle was noted elsewhere in the country by Bhat *et al*. (2012), Mourya *et al*. (2013) and Das *et al*. (2018). The prevalence rates recorded in the present study however are lower in magnitude, even though the number of samples considered in the present study were strictly from age group 1-90 days which is most vulnerable age group for cryptosporidial infection. However these results compel to plan the strategies at this stage so that prevalence rate will be under control and further damage can be prevented.

The prevalence rate of *Eimeria* spp. infection in cow calves (31.25%) in the current study showed a wide difference between the earlier studies conducted from same area. Prior studies by Jyoti *et al*. (2012) reported 54.55% from Punjab, while Nain *et al*. (2017) reported it as 58.84% from Haryana region. The wide variation in the prevalence rates reported may be due to difference in the spatio-temporal attributes of those studies.

Contrary to the observations of current study, Bhat *et al*. (2012) reported highest prevalence of *Cryptosporidium* spp. in buffalo calves during monsoon season followed by pre-monsoon and lowered during post- monsoon season. Morsy *et al*. (2014) also reported seasonal prevalence of *Cryptosporidium* infection, which was higher in spring (58%) followed by winter (55%), autumn (50%) and lowest during summer (43%).

While prevalence of *Cryptosporidium* spp. in cow calves showed non-significant seasonal variation, however, numerically it was higher in summer season as compared to winter and monsoon. Also, Hingole *et al*. (2017) reported prevalence of cyrptosporidum highest in winter (40.5%) followed by that in monsoon (35.63%) and summer (29.41%) with similar trend among buffalo calves without any statistically significant difference between seasons. Seasonal dynamics for cryptosporidiosis in bovines has been projected through earlier reports with wide variation (Bhat *et al*., 2012; Mohanty *et al*., 2012). The higher prevalence during drier months could be attributed to the possibility of shedding of cryptosporidial oocysts due to heat stress by adult animals and feed scarcity during summer season. Sample size could also be the factor for this trend since few numbers of samples were obtained and examined during winter and monsoon.

As regards prevalence rate of *Eimeria* spp. in cow and buffalo calves, it was found that the prevalence was more during winter season followed by summer and least during the monsoon season. The probable reason for least prevalence during monsoon could be that, animals are getting ample feed and fodder during this season, which reduces two way stress *i.e.* one from dry and hot summer in the form of THI and other from scarcity of feed, fodder and clean ample quantity of drinking water. However Filho *et al*. (2016), Jahanzaib *et al*. (2017) reported higher prevalence during monsoon season. Climatic factors such as temperature, humidity etc. which influence the viability *vis-a-vis* longevity of free living lifecycle stages (oocysts) and thus has direct impact on environmental contamination and source of infection (Hingole *et al*., 2017).

In the present study out of 136 Buffalo calves examined 77 were female, 59 males however, 5 Buffalo calves found positive for *Cryptosporidium* infection were all belongs to male sex not a single case reported from the female calves. It may be due to the usual practice in the region of letting male buffalo calves for sale in early life and keeping female calves for future production purposes which eventually also receive better nutritive and managemntal care than males. This may predispose these male calves to various infections. Contrary to our observations, Bhat *et al*. (2012)

both reported higher prevalence in females as compared to males. However prevalence of cryptosporidium was more than female cow calves compared to males. Similar observations were recorded by Venu *et al*., 2012). Few other studies from India even tough recorded marginally higher prevalence of cryptosporidiosis in male as compared to female calves however no gender discrimination in the prevalence of cryptosporidiosis in bovines in India was found (Bhat *et al*., 2012; Hingole *et al*., 2016).

In the present study prevalence of *Eimeria* infections was slightly higher in female buffalo calves as compared to male buffalo calves or in other words it was of about to equal magnitude. Contrary to this observation, Heidari *et al*. (2014), Jahanzaib *et al*. (2017), Nain *et al*. (2017) reported prevalence significantly higher in male calves as compared to females. Likewise in case of cow calves, no significant difference was found for prevalence rate between two sexes. This is in agreement with the observations reported by Jyoti *et al*. (2012), Heidari *et al*. (2014), Sultana *et al*. (2017), Birile *et al.* (2017) and Teketel *et al*. (2017). As regards to Indian farming practices, the better management of females than males keeping in view their economic importance may be the rationalizing factor for this variation (Nain *et al*., 2017). However, Dawid *et al*. (2012) and Teketel *et al*. (2017) opined that sex had no influence on the occurrence of coccidian infection as calves of both sexes have equal chance of accessing the oocysts or no difference on protective immunity for the disease.

In Buffalo calves the age wise prevalence of *Cryptosporidium* occurred in the same manner as like cow calves and it was only present in the calves up to 30 days age. In the 31 days and above age up to 90 days it was totally absent. Bhat *et al*. (2012) observed that the infection was highest in the 0-1 month age group in comparison to other groups and rate of infection decreased with advancing age. Susceptibility of different age groups of cattle and Buffalo dairy calves to the *Cryptosporidium* spp.

In case of cow calves, *Cryptosporidium* spp. infection was found up to 30 days only and its prevalence was more in 16 to 30 days age group however one case was also reported below 15 days in present study. However, *Cryptosporidium* spp. infection was recorded in calves upto 03 months of age. Similarly the age related pattern of *Cryptosporidium* spp. in cow calves in earlier reports by Bhat *et al*. (2012), Venu *et al*. (2012) and Hingole *et al*. (2016) substantiated that the *Cryptosporidium* spp. infection is more prevalent in 1 month aged calves. It is therefore evident that *Cryptosporidium* spp. infection is of young calves only. However, Ouchene *et al*. (2014) also reported the prevalence in adult cows to the extent of 18.78% along with young calves (26.11%) and Huang *et al*. (2014) reported *Cryptosporidium* spp. infection in pre-weaned calves and adults older than 2 year.

In the present study non-significant variations are recorded in the prevalence rate of coccidiosis in buffalo calves. Immediately after birth in the age group 1-15 days prevalence was higher and showing rising trend in the 16-30 days age group, followed by sharp fall afterwards till the calves reaches 90 days of age. Recently, Nain *et al*. (2017) reported that calves of 0-3 months of age had significantly higher prevalence rate of 63.14% than those of age group 3-6 months 43.47%. These reports indicated that animals of any age are susceptible to coccidia infections but their susceptibility tends to decrease in older animals resulting in lower prevalence of the parasites in adult buffaloes (Jahanzaib *et al*., 2017). In the context of these observations resent study cannot be taken into account because it was having observations to a limited age up to 90 days only. Still it has shown the trend as observed in the above studies. It is therefore evident that coccidiosis in buffalo calves is a disease observed since birth and persistent even up to two and half years of age, though its magnitude and intensity shows declining trend, therefore management along with preventive care is the only key for maintaining health status and protection from coccidiosis.

In case of cow calves, even though the prevalence of 34.61% was recorded from calves below 15 days of age in the present study, the literature articulates that the oocysts shedding from cow calves has variable patterns. There appears two clear patterns *i.e.* one as coccidial infections predominates in the age below six months and another thought as coccidial infections predominates in the age above six months. In consistent with present study, Jahanzaib *et al*. (2017) reported higher prevalence of coccidiosis in less than 6 month of age as compared to higher age group. Similar observations were reported by Khan *et al*. (2013) where they observed that the young calves are more susceptible to coccidiosis than adults due to immature immune system or due to the non-development of immunity in calves. On the contrary

Sultana *et al*. (2017) reported that the coccidial infections are higher in age group above 1 year but there prevalence starts from 6 month above to one year. Similarly Ouchen *et al*. (2014), Teketal *et al*. (2017) also supported that coccidial infections in cattle calves occur more in the age group above 6 month.

Remarkably, after examination 28 Murrah buffalo calves not a single case was found infected with *Cryptosporidium*. Similarly quantum of *Eimeria* infection was also significantly less in Murrah buffalo calves as compared to non-descript buffalo calves. Thus it can be inferred that Murrah buffalo calves may have inbuilt protection from *Cryptosporidium* and *Eimeria* spp. infection as compared to non-descript calves. Also, it is incidental that, even after examination of 79 faecal samples of Red Kandhari calves, only one was found positive for *Cryptosporidium* spp. infection, indicating resistance level of these cattle. Ebiyo and Haile (2022) observed more incidence of *Cryptosporidium* spp. infection in crossbred cows as compared to indigenous cows. However, the prevalence of *Eimeria* was also recorded to higher extent in non-descript cows as compared to Red Kandhari cows and was lowest in crossbred cows.

It is therefore inferred that Coccidiosis can occur in indigenous and crossbred cattle to the sizable extent. However variation in infection rate based on breed factor was reported non-significant earlier by Yamral *et al*. (2016). Also Khan *et al*. (2013) from Faisalbabd, Pakistan and Jahanzaib *et al*. (2017) from Lahore, Pakistan in their respective studies concluded that the breed was not a risk factor for coccidiosis in calves.

Since dairying is predominantly adopted as a supportive enterprise to land based farming or farm-labour in drought hit Marathwada region, organised management is seldom seen. This often encompasses absence of proper weaning care. Also as regards weaning practice in cow as well buffalo calves, there are no specific difference between these two species therefore the observations from current study are applicable to both these species of livestock. The higher prevalence of *Cryptosporidium* spp. in un-weaned buffalo and cow calves compared to weaned calves indicates weaned calves have shown somewhat immunity to the infection. It could be attributed to two important factors, firstly as the weaned calves receive calculated milk as per their body requirement, while sucking calves are not allowed to consume sufficient quantity of milk from their dam which affects their growth and health. Secondly, weaned calves are kept in separate pens distant from adults which minimize the odds of exposure to oocysts discharged by adults. Earlier, Huang *et al*. (2014) reported the similar trend of higher prevalence rates of intestinal protozoa in pre-weaned calves than any other age groups. These observations suggested that *Cryptosporidium* spp. infection is a problem that occurs before the weaning age which restricts here to comment whether the prevalence is related with management practice pertaining to weaning or un-weaning.

Remarkably higher prevalence of Cryptosporidium was recorded in cow calves from organized farm than those from un-organized farm. On the other hand, for buffalo calves the prevalence was nil on organised farm and 4.20% on un-organised farm. Venu *et al*. (2012) reported the lower prevalence rate of cryptosporidiosis in bovines maintained at well organised farms compared to those maintained in small and un-organized farms. There is a strong association between housing system and risk of *Cryptosporidium* infection where closed hosing with non-cemented floor and feeding without proper toughs were considered important conducive factors due to increased chance of feed getting contaminated (Bahrami and Alborzi, 2013). Also absence or inability of imparting requisite hygienic practices such as cleaning and dung removal let the area moist and damp which supports the propagation of oocysts (Venu *et al*. 2012; Bahrami and Alborzi, 2013). Several factors contributes to the variations in prevalence are breed, husbandry and management system, age, nursing conditions of the calves, season as well as the sanitary conditions inside and around the farms. However, some of these factors may act individually or collectively to increase the risk of transmission and prevalence of *Cryptosporidium* spp. infection between calves (Venu *et al*., 2012).

Present study reported non-significant difference between *Eimeria* spp. prevalence in calves from organised and un-organised farm. However, Jahanzaib *et al.* (2017) reported higher prevalence in stall-fed, confined calves as compared to grazing animals having out-door access and proper feeding system to avoid fecal contamination of feed. It was also reported that high concentration of ammonia, CO_2 and moisture present due to lack of aeration in pens and groups or accumulation of faeces on the ground due to unsuitable

slatted floor more prevalent in organized farms are major risk factors for calves to contract the oocysts (Thomson *et al*., 2017). In addition to poor sanitation, poor nutrition the chances of contamination are more in organized farm as compared to unorganized sector where high stocking density increases the chance of contamination with *Eimeria* oocysts (Hatam-Nahavandi *et al*., 2019).

CONCLUSION

Direct linkage of both these infections to the management practices and their prevalence ranging from 17.02% to 43.53% in this region suggest that there is much scope for improvement in management practices for the calves. Therefore looking towards the losses due to clinical as well as sub-clinical infections of these important entero-protozoa in dairy animals, proper preventive measures need to be formulated at farm as well as community level so that effective check on the spread of these parasites of economic importance can be possible.

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