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EIMERIA INFECTION IN CALVES UNDER LOCAL FARMING SYSTEM IN AND AROUND ASELLA, OROMIA REGIONAL STATE, ETHIOPIA

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ABSTRACT

Bovine coccidiosis is one of the most common livestock diseases worldwide, and usually affects cattle under one-year-old. High infection rates occur from environments that were already contaminated with infected animals. A study on the prevalence, species and risk of occurrence of *Eimeria* species in calves was conducted at Asella, Oromia Regional State, Ethiopia. Management systems, breed, age, sex, and site were considered as variables for the surveillance. For this, fecal samples were collected from 384 randomly selected calves of age less than or equal to 12 months. Sporulation was applied at room temperature for 10-14 days. The flotation technique was applied on fecal samples and these were examined using direct microscopic examination to detect presence of *Eimeria* species. *Eimeria* oocysts weredetected in 72.4% of the total samples with ranges of 68-100% prevalence in all factors considered. Ten different *Eimeria* species were identified: *E. bovis* (44.5%), *E. zuernii* (26.3%), *E. auburnensis* (10.9), *E. canadensis* (9.4%), *E. ellipsoidalis* (5.7%), *E. subspherica* (6.5%), *E. cylindrical* (3.1%), *E. alabamensis* (2.6%), *E. wyomingensis* (2.6%) and *E. bukidnonensis* (2.1%) in decreasing order of prevalence. Of the 278 positive calves, 52.2% were infected with single species, while 47.8% were infected with 2-4 of identified species. Significantly lower prevalence in calves fed colostrums (71.2%) than those not fed colostrum (100%) ($\chi 2 = 6.3$; $p < 0.05$) were observed in early birth. However, no statistical association in infection was observed within all other risk factors considered ($p > 0.05$). Infection with one or more species was found to be insignificant in sex, feeding system, and cleaning frequency ($p > 0.05$). The present finding showed the role of early colostrum feeding in a remarkable reduction of coccidiosis. However, the majority of assessed factors had a great contribution for the contamination and maintenance of *Eimeria* on the farm and its primes at 68.4%-100%, with high risk of infection to subsequent calves. Moreover, the occurrence of infection, with single (52.2%) to multiple of four (1.4%) pathogenic *Eimeria* spp. in positive cases, indicated the downside of substandard management systems, making calves prone to subclinical cases thus hampering growth and health, with considerable economic losses. Therefore, early feeding with sufficient amount of colostrum, practicing good management in calves and implementation of proper hygienic measures in their environments should be recommended to reduce direct and indirect economic losses from the infection.

Key words: *Eimeria*, epidemiology, prevalence, calves, infection, sporulated oocyst, Asella

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INTRODUCTION

Bovine coccidiosis is one of the most common livestock diseases worldwide and usually affects cattle under one-year-old. It is occasionally seen in yearlings and even adults, especially if massive infections are acquired [1, 2]. It is a protozoal infection caused by the *Eimeria* genus of several species, which comprises *E. bovis, E. zuernii, E. ellipsoidalis, E. cylindrica, E. subspherica, E. canadensis, E. alabamensis*, and *E. auburnensis* in several classes of livestock, particularly in dairy calves [2]. The *Eimeria* species cause disease primarily in animals up to two years old and it is particularly common in calves between three weeks and six months of age [2]. Oral-fecal transmission in a contaminated environment having sporulated oocysts is the most common infection route. Usually, the oocysts that pass through the feces of infected animals require appropriate environmental conditions that favor sporulation.

Dairy environments with high temperatures prevent them from acting as a source of infection [3]. High infection rate occurs when animals are placed in environments contaminated by infected animals, which happens either indoors from bedding or outdoors around drinking or feeding troughs. However, not all coccidian species cause disease. The two most common pathogenic species are *E. bovis* and *E. zuernii* [4] but *E. alabamensis, E. auburnensis,* and *E. wyomingensis* may also cause disease in calves [5].

The diagnosis of the disease involves clinical symptoms, post-mortem examination, scrapings of the intestinal mucosa, fecal examination, laboratory sporulation test, and morphological characteristic according to availability [2, 3]. A few studies were conducted in the central part of Ethiopia with 68.1% of calves reported with coccidiosis [6] and 24.9% of cattle infected in a retrospective laboratory report [7]. However, the epidemiological status of *Eimeria* and the risk of infection in calves managed under traditional farming systems at Asella is not yet known. Hence, this study aimed at performing an epidemiological survey of *Eimeria* species in calves at Asella.

MATERIALS AND METHODS

Study area

The study was conducted in Arsi Zone of Oromia Regional state, in and around Asella town located 175 km from Addis Ababa, south east of Oromia Original State, Ethiopia. The agro-ecology and livestock population in the study area are mid altitude with a mixed farming system as described in Arsi Plan Economic Development Office (APEDO) [8].

Study animals and the considered risk factors

The study constituted 384 randomly sampled calves of age less than or equal to 12 months, and 1-3 calves per owner were sampled. Animal husbandry practices such as management systems including cleanings of cow udder and feeding and watering troughs, colostrum feeding time, house cleaning frequency, housing system and cleaning were collected from owners through interviews. Breed, age, and sex of calves were considered as risk factors.

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Samples

Fecal samples were collected directly from the rectum into clean universal bottles preserved with 2.5% potassium dichromate [1]. Each sample was clearly labeled and transported under cold chain to Asella Regional Veterinary Laboratory.

Laboratory procedures and data analysis

Laboratory investigation was performed according to Kaufmann [9] with application of sporulation for 10-14 days at room temperature. Using overtime stand flotation technique with no centrifugation, microscopic examination was carried out and the sporulated oocysts and the oocysts were characterized by using shape and morphological appearances of the respective species [1, 10]. All the data obtained were analyzed using STATA Version 7.0 and SPSS 16.0 to determine the association of risk factors. Chi-square was analyzed for the considered variables of the study. *p value* (<0.05) was considered as the significance point of *Eimeria* infection.

RESULTS

Out of the total 384 fecal samples examined, 278 (72.4%) tested positive for *Eimeria*. The prevalence of *Eimeria* was 68.4% in male and 75.8% in female calves. As shown in Table 1, the infection rates were significantly different between presence (71.2%) and absence (100%) of colostrum feeding at an early age ($p < 0.05$). However, no statistical significant differences ($p > 0.05$) in occurrence of *Eimeria* infection were found.

From the 278 positive calves, 10 species of *Eimeria* were identified: These were *E. bovis* (44.5%), *E. zuernii* (26.3%), *E. auburnensis* (10.9%), *E. canadensis* (9.4%), *E. subspherica* (6.5%), *E. ellipsoidalis* (5.7%), *E. cylindrica* (3.1%), *E. alabamensis* (2.6%), *E. wyomingensis* (2.6%), and *E. bukidnonesis* (2.1%) in the decreasing order. As shown in Table 2, differences in prevalence of *E. bovis*, *E. auburnensis*, *E. canadensis*, and *E. ellipsoidalis* were observed to be significantly lower in the colostrum feeding states ($p < 0.05$) but not in the others factors ($p > 0.05$).

The prevalence of *Eimeria* species by breed, sex, and age is shown in Table 3. The sex did not influence the prevalence of isolated species ($p > 0.05$) but significant differences ($p < 0.05$) were observed for *E. zuernii*, *E. auburnensis*, *E. ellipsoidalis*, *E. subspherica*, and *E. cylindrica* in relation to breed, and for *E. zuernii* and *E. wyomingensis* in relation to age groups.

The prevalence of *Eimeria* species according to housing system, house cleaning frequency and udder cleaning are described in Table 4. Differences in prevalence were not observed for housing system and house cleaning frequency ($p > 0.05$). However, high prevalence of *E. auburnensis*, *E. subspherica*, and *E. alabamensis* ($p < 0.05$) were observed in calves suckling un-cleaned udder.

Out of 278 positive calves for *Eimeria* species, 47.8% of them were infected at a rate of 2-4 species. The remaining 52.2% were infected with single species (Fig. 1). Calf infection with two, three, and four *Eimeria* species were 41.1%, 5.0%, and 1.4%, respectively.

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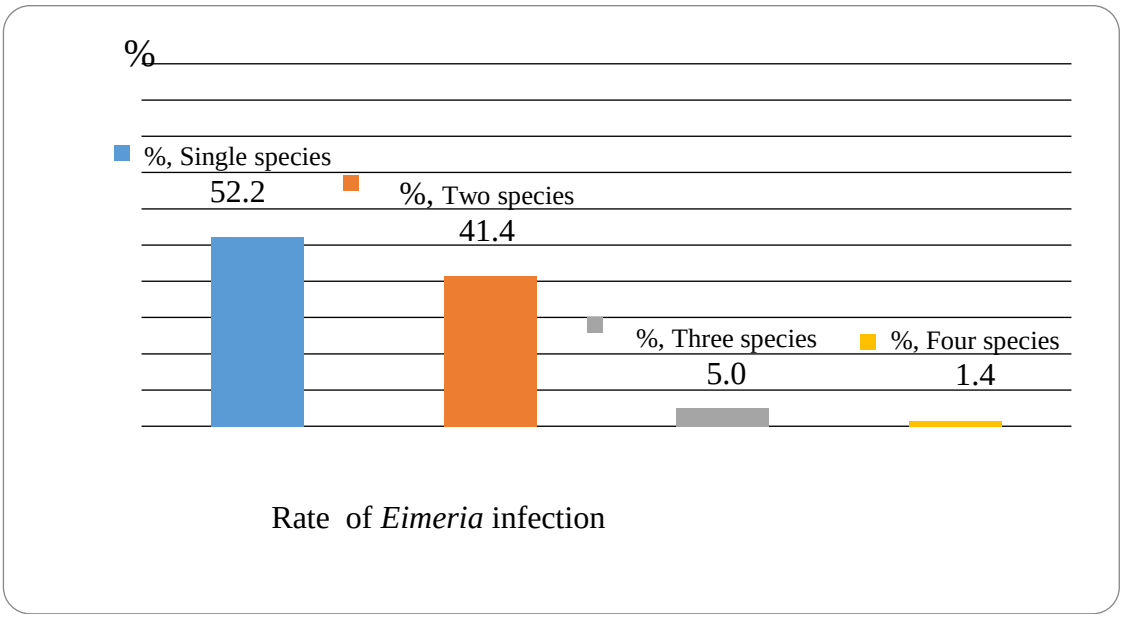

Figure 1: Rate of *Eimeria* infection in studied dairy calves which are infected

The independent samples test showed no association ($p > 0.05$) of the risk of sex, feeding systems, and cleaning frequency on number of infections, conditional on the calf being infected. However, the association could not be analyzed for other factors due to zero values in the cell for breed, age, housing system, cleaning of udder, and colostrum feeding (Table 5).

DISCUSSION

The epidemiological investigation of *Eimeria* is found to be important to implement the control and prevention strategies in dairy calves' management. Thus, the fact that overall 72.3% of calves tested positive for *Eimeria* showed the prevalence and the burden of infection in the study area. Moreover, the insignificant differences in the occurrence of the agent according to breed, sex, age, feeding system, housing systems, house cleaning frequency, and udder cleaning states before suckling also shows a high incidence of *Eimeria* in the area. Although the sample sizes were not proportional in colostrum feeding states due to random sampling, significant difference in prevalence related to colostrum feeding showed the role of protection by passive immunity in *Eimeria* infection. On the other hand, only 16 calves in total did not receive colostrum; however, all of them were infected. The overall prevalence of *Eimeria* infection in the present study was compatible with the 68.1% finding of coccidiosis in dairy calves in central Ethiopia [6], 67.4% in Kenya [11] and 70% in South Africa [12]. These studies show the widespread occurrence of the agent in the calves' environment. The present finding was also higher than reports from Saudi Arabia [13], Japan [14, 15], the Netherlands [16], and Poland [17]. The difference could be due to differences in the calves' management systems, agro-ecology of the areas, and the awareness of breeders on the importance of colostrum feeding in the study areas. This study was performed under traditional calves' management system. Radostits *et al*. [2], Davis and Drackley [18], and Seifert [19]

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indicated poor farming system as a risk factor for predisposing to *Eimeria* infection while the improved dairy calves' management systems reduce the risk and improve control of coccidiosis.

The detection of several *Eimeria* species in the study area showed the high risk. *Eimeria* species also maintained and circulated in dairy animals and in the environment under sub-clinical conditions, which became a source of infection for the subsequent new calves, particularly the neonates. Hiko and Wondimu [20] emphasized that sub-clinical parasitic conditions in dairy animals result in reduced animal performance and increased susceptibility to other diseases. The highest prevalence was observed for *E. bovis* (44.5%) followed by *E. zuernii* (26.3%) and *E. auburnensis* (10.9%) which was similar to a report by Speer [21]. These species are usually the most frequently seen coccidia in outbreaks of coccidiosis throughout the world [2]. Despite that, most of the studied calves were found to be infected with *Eimeria* species, although clinical symptoms were not observed during the study period. This resulted in subclinical infections especially due to the high proportion of the pathogenic species such as *E. bovis* and *E. zuernii* observed in the present study. The impacts are underestimated, since infection negatively affects animal productivity, growth and physiological performance; contributing to economic losses in the livestock sector. Moreover, infection continues to promote the shedding of oocysts and environmental contamination. Such environment will act as the potential source of infection for the calves [19]. The multiple infections with 2-4 *Eimeria* species in this study was smaller than those reported with seven different species in Ethiopia [6], in USA [22]) and in the Netherlands [16], as well as with five *Eimeria* species in Canada [23]. The status of multiple infections with age, housing system, lower udder cleaning during milking, and absence of colostrum feeding in this report showed the risk for infection. Similar patterns were suggested by Radostits *et al*. [2] and Seifert [19].

CONCLUSION

Based on the present results, early colostrum feeding showed remarkable reduction of coccidiosis. However, the majority of other factors assessed were found to contribute to the contamination and maintenance of *Eimeria* on the farm. The occurrence of multiple pathogenic *Eimeria* spp. in the studied dairy calves indicates that substandard management systems make calves prone to a subclinical form of infection. This disease hampers growth and health of the most susceptible animals especially young calves under six months old. This may lead to considerable economic losses at the dairy farm. Therefore, besides early feeding with enough amount of colostrum, practicing good management in calves and implementation of proper hygienic measures in their environments were recommended to reduce direct and indirect economic losses from the infection.

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Table 1: The status of *Eimeria* infection according to studied factors

Studied Factors		Number examined	Positive No. (%)	$\chi 2$	P-value
Breed	Local	156	116 (74.4)	0.5	0.4
	Cross	228	162 (71.1)		
Sex	Female	207	157 (75.8)	2.6	0.1
	Male	177	121 (68.4)		
Age (months)	Less than 6	213	157 (73.7)	0.4	0.5
	7-12	171	121 (70.8)		
Feeding system	Ground	131	96 (73.3)	0. 1	0.7
	Troughs	253	182 (71.9)		
Housing system	Single	64	44 (68.7)	0.5	0.4
	Group	320	234 (73.1)		
Cleaning Frequency (day)	Once	338	245 (72.5)	2.2	0.1
	Twice	46	33 (71.7)		
Udder Cleaning	Present	210	146 (69.5)	1.9	0.1
	Absent	174	132 (75.9)		
Colostrum feeding	Present	368	262 (71.2)	6.3	0.0
	Absent	16	16 (100.0).		
Total		384	278 (72.3)		

Table 2: Factors related to feed and feeding system on the prevalence of *Eimeria* species

Eimeria species	Total No. of examined (n=384)	Feeding system		Colostrum feeding	
		Ground (n=131)	Troughs (n=253)	Present (n=368)	Absent (n=16)
	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)
E. bovis	171 (44.5)	56 (42.7)	115 (45.5)	160 (43.5)	11 (68.8)*
E. zuernii	101 (26.3)	32 (24.4)	69 (27.3)	98 (26.6)	3 (18.8)
E. auburnensis	42 (10.9)	16 (12.2)	26 (10.3)	37 (10.1)	5 (31.3)*
E. canadensis	36 (9.4)	16 (12.2)	20 (7.9)	30 (8.2)	6 (37.5)*
E. ellipsoidalis	22 (5.7)	6 (4.6)	16 (6.3)	16 (4.3)	6 (37.5)*
E. subspherica	25 (6.5)	10 (7.6)	15 (5.9)	22 (6.0)	3 (18.8)
E. cylindrica	12 (3.1)	5 (3.8)	7 (2.8)	12 (3.3)	0 (0.0)
E. alabamensis	10 (2.6)	4 (3.1)	6 (2.4)	10 (2.7)	0 (0.0)
E. wyomingensis	10 (2.6)	3 (2.3)	7 (2.8)	9 (2.4)	1 (6.3)
E. bukidnonesis	8 (2.1)	5 (3.8)	3 (1.2)	8 (2.2)	0 (0.00)

* Statistically significant ($p < 0.05$)

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Table 3: Prevalence of *Eimeria* species according to breed, age, and sex of animals

Eimeria Species	Breed		Sex		Age (months)	
	Local (n=156)	Cross (n=228)	Female (n=207)	Male (n=177)	< 6 (n=213)	7-12 (n=171)
	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)
E. bovis	68 (43.6)	103 (45.2)	92 (44.4)	79 (44.6)	99 (46.5)	72 (42.1)
E. zuernii	30 (19.2)	71 (31.1*	57 (27.5)	44 (24.9)	65 (30.5)	36 (21.1)*
E. auburnensis	23 (14.7)	19 (8.3)*	27 (13.0)	15 (8.5)	21 (9.9)	21 (12.3)
E. canadensis	17 (10.9)	19 (8.3)	24 (11.6)	12 (6.8)	22 (10.3)	14 (8.2)
E. ellipsoidalis	4 (2.6)	18 (0.1)*	14 (6.8)	8 (4.5)	10 (4.7)	12 (7.0)
E. subspherica	18 (11.5)	7 (3.1)*	17 (8.2)	8 (4.5)	11 (5.2)	14 (8.2)
E. cylindrica	1 (0.6)	11 (4.8)*	4 (1.9)	8 (4.5)	9 (4.2)	3 (1.6)
E. alabamensis	10 (6.4)	0 (0.0)	6 (2.9)	4 (2.3)	3 (1.4)	7 (4.1)
E. wyomingensis	3 (1.9)	7 (3.1)	4 (1.9)	6 (3.4)	2 (0.9)	8 (4.7)*
E. bukidnonesis	3 (1.9)	5 (0.1)	4 (1.9)	4 (2.3)	3 (1.4)	5 (2.9)

* Statistically significant ($p < 0.05$)

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Table 4: Factors related to housing and cleaning methods on the prevalence of *Eimeria* species

Eimeria species	Housing system		House cleaning frequency/day		Udder cleaning	
	Single (n=64)	Group (n=320)	Once (n=338)	Twice (n=46)	Present (n=210)	Absent (n=174)
	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)	Positive No. (%)
E. bovis	35 (54.7)	136 (42.5)	153 (45.3)	18 (39.1)	93 (44.3)	78 (44.8)
E. zuernii	17 (26.6)	84 (26.3)	89 (26.3)	12 (26.1)	61 (29.0)	40 (22.9)
E. auburnensis	3 (4.7)	39 (12.2)	39 (11.5)	3 (6.5)	13 (6.2)	29 (19.7)*
E. canadensis	7 (10.9)	29 (9.1)	33 (9.8)	3 (6.5)	15 (7.1)	21 (12.1)
E. ellipsoidalis	3 (4.7)	19 (5.9)	17 (5.0)	5 (10.7)	12 (5.7)	10 (5.7)
E. subspherica	1 (1.6)	24 (7.5)	22 (6.5)	3 (6.5)	2 (0.9)	23 (13.2)*
E. cylindrica	3 (4.7)	9 (2.8)	11 (3.3)	1 (2.2)	9 (4.3)	3 (1.7)
E. alabamensis	0 (0.0)	10 (3.1)	10 (2.9)	0 (0.0)	2 (0.9)	8 (4.6)*
E. wyomingensis	1 (1.6)	9 (2.8)	8 (2.4)	2 (4.4)	7 (3.3)	3 (1.7)
E. bukidnonesis	1 (1.6)	7 (2.2)	7 (2.1)	1 (2.2)	4 (1.9)	4 (2.3)

*Statistically significant ($P < 0.05$)

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Table 5: Frequency of *Eimeria* positive calves as infection with one or more species

Risk Factors		Number examined	Rate of *Eimeria* observation No. (%)				$\chi 2$	*P-value*
			One	Two	Three	Four		
Breed	Local	156	62 (39.7)	50 (32.1)	4 (2.5)	0 (0.0)	-	-
	Cross	228	53 (36.4)	65 (28.5)	10 (4.4)	4 (1.8)		
Sex	Female	207	84 (40.6)	59 (40.6)	11 (5.3)	3 (1.5)	7.33	0.1
	Male	177	61 (34.4)	56 (31.6)	3 (1.7)	1 (0.6)		
Age (months)	< 6	213	86 (40.3)	56 (26.2)	14 (6.5)	1 (0.4)	-	-
	7-12	171	59 (34.5)	59 (34.5)	0 (0.0)	3 (1.8)		
Feeding system	Ground	131	48 (36.6)	43 (32.8)	3 (2.3)	2 (1.5)	2.13	0.7
	Troughs	253	97 (38.3)	72 (28.5)	11 (4.4)	2 (0.7)		
Housing system (pen)	Single	64	24 (37.5)	13 (20.3)	7 (10.9)	0 (0.0)	-	-
	Group	320	121 (37.0)	102 (31.0)	7 (2.2)	4 (1.2)		
Udder Cleaning	Present	220	84 (40.0)	53 (25.2)	9 (4.3)	0 (0.0)	-	-
	Absent	174	61 (35.0)	62 (35.6)	5 (2.9)	4 (2.3)		
Cleaning frequency (day)	Once	338	132 (39.0)	101 (29.0)	13 (3.9)	3 (0.8)	3.83	0.4
	Twice	46	13 (28.3)	14 (30.4)	1 (2.2)	1 (2.1)		
Colostrum feeding	Present	368	141 (38.0)	106 (28.8)	14 (3.8)	1 (0.2)	-	-
	Absent	16	4 (26.6)	9 (56.3	0 (0.0)	3 (18.8)		
Total		384	145 (37.8)	115 (29.9)	14 (3.6)	4 (1.0)		

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