PREVALENCE AND MORPHOLOGICAL IDENTIFICATION OF *EIMERIA* SPECIES ON BALI CATTLE IN SUMBAWA DISTRICT, WEST NUSA TENGGARA

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ABSTRACT: This study aims to determine the prevalence and morphological identification of *Eimeria* spp. from Bali cattle feces on traditional farms in Sumbawa District. A total of 300 samples were identified and 238 samples were positive, resulting in a prevalence of *Eimeria* spp. of 79.33%. Based on age category, the highest prevalence of *Eimeria* spp. was found in cattle aged 6 months-2 years (88.76%; 79/89), followed by cattle aged <6 months (88%; 88/100) and the lowest prevalence in cattle aged >2 years (63.96%; 71/111). Female Bali cattle had the highest prevalence of *Eimeria* spp. (80.55%; 145/180) than male Bali cattle (77.50%; 93/120). Based on livestock systems, the highest prevalence of *Eimeria* spp. was found in semi-intensive (89.15%; 74/83), then extensive (80.76%; 105/130) and the lowest in intensive (67.81%; 59/87). Six different *Eimeria* species were identified: *E. bovis* (33.19%), *E. auburnensis* (21.42%), *E. zuernii* (15.96%), *E. alabamensis* (12.18%), *E. ellipsoidalis* (10.50%), and *E. canadensis* (6.72%). Preventing and controlling coccidiosis infection in cattle can be done with good hygiene management.
Keywords: Bali cattle, *Eimeria* species, traditional farms, morphologi.

INTRODUCTION

Bali cattle is one of several cattle varieties in Indonesia that can adapt to the tropical environment. Bali cattle are the result of domestication from wild Bali cattle, which are known to have the advantages of good fertility (80–85%) and a high carcass percentage (56%). (Hilmiati et al., 2021; Nurany et al., 2022). Therefore, Bali cattle are prioritized by the Ministry of Agriculture to meet the needs of high-protein food and achieve meat self-sufficiency in Indonesia (Andi et al., 2017). However, Bali cattle are reported to have low productivity in some

breeding centers such as Sumbawa Island, West Nusa Tenggara Province (Besung et al., 2019; Adnyana et al., 2021). Factors affecting the low productivity of Bali cattle in Sumbawa include slow cow growth, high calf mortality (10-27%) and high prevalence of gastrointestinal parasites (65.38% - 90.99%) (Sriasih et al., 2021).

Gastrointestinal parasites including protozoa are reported to infect cattle worldwide. *Eimeria* spp. are the most significant gastrointestinal protozoa in Bali cattle and can cause coccidiosis (Nurany et al., 2022). The prevalence of *Eimeria* spp. was reported to be (54%) in Lopok District, Sumbawa Regency (Hamdani et al., 2021) and (23.68%) in Dompu District, Sumbawa Island (Sriasih et al., 2018). Coccidiosis in livestock occurs due to the multiplication of *Eimeria* spp. in the intestinal wall relatively quickly, resulting in rupture of the intestinal epithelium. As a result, nutrient absorption in the intestine is disrupted resulting in diarrhea containing blood, fibrin and intestinal tissue (Daugschies & Najdrowski, 2005; Bangoura et al., 2022). In addition, livestock will also experience anemia, depression, reduced appetite, weight loss, and weakness (Heidari & Gharekhani, 2014). The economic loss caused by *Eimeria* spp. infection in cattle is estimated at USD 400 million worldwide (Ekawasti et al., 2019). This is attributed to the cost of anticoccidial treatment, losses due to morbidity and mortality. The incidence of coccidiosis can be caused by the age of the animal when infected, the number of oocysts ingested, the presence of other microorganism infections, farm management and *Eimeria* species (Alcala-Canto et al., 2020).

Eimeria spp. are specific host parasites and as many as 20 species of *Eimeria* infecting cattle have been reported worldwide (Hamid et al., 2019). Five species have been reported to be pathogenic including *E. alabamensis*, *E. auburnensis*, *E. ellipsoidalis*, *E. bovis* and *E. zuernii* (Daugschies & Najdrowski, 2005). Only *E. bovis* and *E. zuernii* species are highly pathogenic as they can cause clinical coccidiosis while other species are mildly or moderately pathogenic (Das et al., 2015). *Eimeria* spp. can be identified conventionally through a floating method of examination by observing under a microscope. Identification of *Eimeria* spp. oocysts is based on shape and size. Oocysts of *Eimeria* spp. are round, elliptical and oval with smooth, transparent, homogeneous and yellowish or colorless surfaces. Oocysts are 10-50 μm in size depending on the *Eimeria* species (Ekawasti & Wardhana, 2019).

The rearing system of Bali cattle in Sumbawa Regency is carried out extensively, semi-intensively and intensively. The risk of coccidiosis is higher in extensive and semi-intensive rearing systems because it can be infected from *Eimeria* spp oocysts in the environment. Environmental factors such as climate change, temperature, humidity, rainfall intensity, geographical altitude, sanitation and livestock population density can affect protozoan infection (Nurany et al., 2022). This study aimed to determine the prevalence and morphological identification of *Eimeria* species infecting Bali cattle in Sumbawa Regency based on oocyst morphology examination.

METHOD

This study was a discriptif observasional on Bali cattle in Sumbawa District with a population of 252,943 heads. The sample used in this study was Bali cattle

feces. Bali cattle were categorized by age (< 6 months), (6 months - 2 years) and (> 2 years) which were randomly selected.

Sampel Size

The sample size used in this study was n = 4PQ / L2. 95% confidence interval, prevalence rate (P) =75%, error rate (L)=5% and Q=1-P so that the sample size (n) is 300 samples (Trushfield, 2007).

Collection of Fecal Samples

Feces were taken directly through the rectum as much as 20-50 grams. The feces were then put into a sample pot, given a marker number that was adjusted to the cattle data including age, sex, feces consistency, livestock system, time and place of collection. Samples were then taken using a cool box to the Laboratory of Veterinary Parasitology Division, Faculty of Veterinary Medicine, Airlangga University, Surabaya. Samples were stored in a refrigerator at 4°C during the study.

Microscopic Examination

Fecal examination to identify *Eimeria* spp. oocysts was carried out using the floating method. The first step is to make a fecal suspension with a ratio of 1 g of feces diluted in 9 ml of distilled water. After that, filtration was carried out and the filtrate from the filtration was placed in a centrifuge tube, then centrifuged at 1500 rpm for 5 minutes. The centrifuged supernatant was discarded and 10 ml of saturated sugar was added. Next, centrifugation was carried out at 1500 rpm for 5 minutes, the tube was transferred to the tube rack and saturated sugar was added until it was full with the liquid surface becoming convex. The sentifuse tube was covered with cover glass for 5 minutes and then oocysts were qualitatively identified as positive and negative with a Nikon® E100 microscope, Japan at 100-400x magnification. The identification of *Eimeria* spp. oocysts was based on morphology (Florião et al., 2016) and then the prevalence of *Eimeria* spp. in Bali cattle was calculated. Calculation of the prevalence of *Eimeria* spp. was calculated based on the formula (Rehman et al., 2011).

$$\text{Prevalence (\%)} = \frac{\text{Number of samples infected (n)}}{\text{Total samples examined (N)}} \times 100$$

RESULTS AND DISCUSSION

The results of this study obtained *Eimeria* spp. oocysts from the feces of Bali cattle in Sumbawa Regency using the floating method. A total of 300 samples were identified and 238 positive samples were obtained, resulting in a prevalence of *Eimeria* spp. of 79.33%. *Eimeria* species found based on morphological identification included *E. auburnensis*, *E. bovis*, *E. zuernii*, *E. canadensis*, *E. ellipsoidalis* and *E. alabamensis*. The morphology of the six identified *Eimeria* spp. oocyst species has different characteristics (Figure 1) (Table 1).

The results obtained in this study were higher than previous studies. Sriasih et al. (2018) reported the prevalence of *Eimeria* spp. in Bali cattle in Dompu Regency, West Nusa Tenggara as 23.68%. and Hamid et al. (2019) in beef cattle in West Nusa Tenggara Province was 78.43%. Studies on Bali cattle in Bali Province found the prevalence of *Eimeria* spp. was 58.8% (Rahmawati et al., 2018), 32.23% (Saputri et al., 2018) and 24.19% in Bali cattle at the Superior Livestock Breeding

Center, Denpasar (Nurany et al., 2022). Various factors can influence the prevalence of *Eimeria* spp. including differences in sampling time, number of samples, feed source, farm management, season, temperature, humidity and geographical conditions (Gupta et al., 2016).
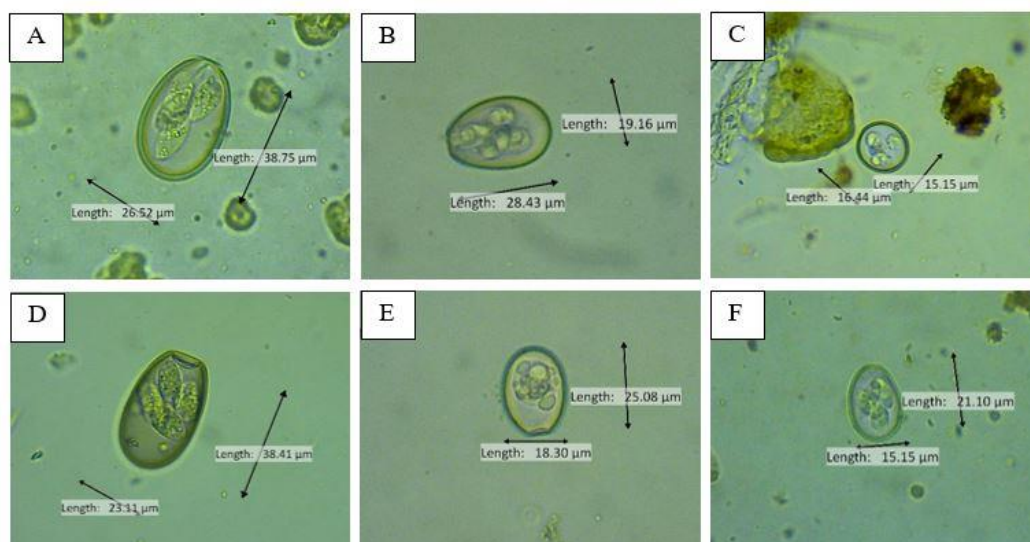
Figure 1. Morphology of six oocysts *Eimeria* species in Bali cattle at 400X magnification: (A) *E. auburnensis*. (B) *E. bovis*. (C) *E. zuernii*. (D) *E. canadensis*. (E) *E. ellipsoidalis* (F) *E. alabamensis*

Table 1. Morphology of Oocysts *Eimeria* species from Bali Cattle in Sumbawa Regency.

Species	Size (µm)		Shape	Characteristics
	Length (range)	Width (range)		
E. auburnensis	38.75 (36.0-45.0)	26.52 (24.0-26.0)	ovoid	Double-layered walls, wide and crimped micropiles
E. bovis	28.43 (27.0-32.0)	19.16 (20.0-23.0)	ovoid	Double-layered walls, micropiles present
E. zuernii	16.44 (16.0-22.0)	15.15 (14.0-19.0)	sperical	Double-layered and smooth walls, no micropiles
E. canadensis	38.41 (33.0-38.0)	22.11 (22.0-26.0)	ovoid	Double-layered walls, wide micropiles and inward invaginations
E. ellipsoidalis	25.08 (19.0-27.0)	18.30 (15.0-20.0)	ellips	Thin wall, no micropile
E. alabamensis	21.10 (17.5-22.5)	15.15 (12.5-18.0)	ovoid	Smooth wall, no micropiles

Based on data from the Central Bureau of Statistics of Sumbawa Regency (2022), Sumbawa Regency has a tropical climate that is influenced by the rainy season and dry season. The rainy season runs from November to April and the dry season runs from May to October. Samples for this study were taken from November to December, which is the beginning of the rainy season, resulting in a high prevalence of *Eimeria* spp. Previous research reported by Asfaw et al. (2016) which found a higher prevalence of coccidiosis in the rainy season (67.7%) than in the dry season (57.3%). According to Gupta et al. (2016) the rainy season is significantly associated with a higher prevalence of *Eimeria* spp. in cattle because

it is a favorable condition in terms of temperature and humidity for sporulation of *Eimeria* spp. oocysts. In addition, the high prevalence of *Eimeria* spp. in the rainy season can be attributed to the presence of precipitating factors such as stress, bad weather, wet conditions and high levels of fecal contamination in feed and water sources (Das et al., 2015).

Factor associated with the high prevalence of *Eimeria* spp. is management of livestock in Sumbawa Regency. Most farmers use conventional livestock systems with characteristics of intensive to semi-intensive animal raising (Hilmiati, 2019). The results of this study showed that the prevalence of *Eimeria* spp. was higher in the semi-intensive (89.15%;74/83) and extensive (80.76%;105/130) livestock systems while the intensive system had a lower prevalence (67.81%;59/87) (Table 2).

Table 2. **Prevalence of *Eimeria* spp. in Bali Cattle Based on Livestock System**

Livestock system	Number of Samples	Number of Positif Samples	Prevalence
Ekstensive	130	105	80.76%
Semi-intensive	83	74	89.15%
Intensive	87	59	67.81%

Similar results were reported by Kertawirawan (2013) who obtained coccidiosis prevalence of 85.71% in Bali calves reared with traditional cage system and poor sanitation, 57.14% in semi-intensive model and 0% in intensive cage model. Study conducted by Asfaw et al. (2016) in Ethiopia and Makau et al. (2017) in Kenya reported the prevalence of *Eimeria* spp. in calves reared with semi-intensive systems of 71.7% and 32.8%.

The prevalence of *Eimeria* spp. is higher in extensive and semi-intensive livestock systems than in intensive livestock systems. This is due to several factors such as population density in pastures, oxygen and lighting levels, cage hygiene, feeding patterns and drinking water sources (Bangoura et al., 2007; Rehman et al., 2011). According to Tomczuk et al. (2015) larger numbers of animals and crowding are predisposing factors for coccidiosis. Herds living in groups will be infected at the same time if they are under the same field conditions. This is because the continuous release of oocysts from subclinically or clinically infected cattle will contaminate the environment and cause coccidiosis in other cattle (Sufi et al., 2017). Intensive rearing systems have a lower prevalence due to smaller herds and better sanitation. Cattle are housed for 24 hours, making it efficient for feeding, disease control, bathing and cleaning (Ekawasti et al., 2021).

The prevalence of *Eimeria* spp. related to the age of the livestock was also observed in this study. The lowest prevalence (63, 96%; 71/111) was found in cattle aged >2 years while cattle aged <6 months and 6 months to 2 years showed relatively higher prevalence (88%; 88/100 and 88.76%; 79/89) (Table 3).

Table 3. Identification of *Eimeria* species in Bali Cattle Based on Age.

Age	Number of Samples	Number of Positif Samples	Prevalence
<6 month	100	88	88%
6 month – 2 year	89	79	88.76%
>2 year	111	71	63.96%

The results of this study are in accordance with studies (Dong et al., 2012; Ekawasti et al., 2019; Hamid et al., 2019; Hastutiek et al., 2022) which reported a higher prevalence of *Eimeria* spp. in cattle aged < 2 years. Young cattle are more susceptible to *Eimeria* spp. infection than mature cattle because younger animals generally have underdeveloped immunity to coccidia infection. In addition, immunity is highly specific to only one species of *Eimeria*, and therefore cannot provide cross-protection against subsequent infection with other species (Bangoura et al., 2022). Meanwhile, adult cattle (>2 years old) already have immunity to *Eimeria* spp. infection as a result of previous exposure.

Based on gender, the prevalence of *Eimeria* spp. was higher in cows (80.55%; 145/180) than in bulls (77.50%; 93/120) (Tabel 4). Similar results were reported in Bangladesh with the prevalence of *Eimeria* spp. in dairy cows was 61.28% and bulls were 49.03% (Candra Deb et al., 2022). Hiko & Rorisa (2016) also reported the prevalence of *Eimeria* spp. in Ethiopia was higher in cows (75.8%) than in bulls (68.4%). The higher prevalence found in cows could be attributed to physiological stress conditions in cows due to pregnancy and breeding (Sufi et al., 2017).

Table 4. Prevalence of *Eimeria* species. in Bali Cattle Based on Gender

Gender	Number of Samples	Number of Positif Samples	Prevalence
Bulls	120	93	77.50%
Cows	180	145	80.55%

Coccidiosis in cattle can be caused by more than one *Eimeria* species. In this study, six *Eimeria* species were identified, namely *E. bovis* (33.19%), *E. auburnensis* (21.42%), *E. zuernii* (15.96%), *E. alabamensis* (12.18%), *E. ellipsoidalis* (10.50%), and *E. canadensis* (6.72%) (Tabel 4). Previous studies reported that *E. bovis*, *E. zuernii*, *E. auburnensis*, *E. ellipsoidalis*, *E. canadensis* and *E. alabamensis* were the species frequently identified in cattle (Enemark et al., 2013; Heidari et al., 2014; Saravia et al., 2021; Hastutiek, et al., 2022). *E. bovis* is the most dominant species found in this study compared to other species because in this study 18 cows were found to have diarrhea and emaciation. According to Lopez-Osorio et al. (2020) *E. bovis* is one of the most common pathogenic species affecting livestock worldwide, causing clinical coccidiosis such as diarrhea containing blood and fibrin while some other species can cause subclinical coccidiosis. Therefore, prevention and control of coccidiosis is necessary as there is no vaccine for coccidiosis in livestock (Bangoura et al., 2022).

Prevention and control of coccidiosis can focus on livestock management including hygiene management practices. *Eimeria* spp. oocysts can survive for a long time in the environment under moist conditions (Campbell, 2021). Therefore, timely cleaning of manure, use of clean paddocks and pastures that are not overcrowded with livestock are management practices that can prevent *Eimeria* spp. infection (Keeton & Navarre, 2018).

CONCULSION

Based on the research results, it can be concluded that (1) The prevalence of *Eimeria* spp. in Bali cattle in Sumbawa District is high, with a prevalence of 79.33%; (2) Species were identified are *E. bovis*, *E. zuernii*, *E. auburnensis, E.

canadensis, E. alabamensis, and *E. ellipsoidalis*; (3) The prevalence of *Eimeria* spp. was high in Bali cattle raised semi-intensively (89.15%); calves aged 6 months-2 years (88.76%); and cows (80.55%). Calves have a higher prevalence of *Eimeria* spp. because immunity has not yet developed against coccidia infection, efforts to prevent and control coccidiosis infection in livestock can be made with good hygiene management practices.

RECOMMENDATION

The prevalence of *Eimeria* spp. on Bali cattle in Sumbawa Regency is relatively high, especially in young cattle that are raised traditionally. Therefore, it is necessary to provide counseling and treatment for coccidiosis so that the productivity of Bali cattle can increase.

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