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Determination of the morphology of *Eimeria* spp. in beef cattle in Bandung Regency, West Java Province, Indonesia

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ABSTRACT

Background: *Eimeria* spp. include endoparasites that can infect the digestive tract of cattle. Coccidiosis can reduce productivity, cause economic losses, increase susceptibility to other diseases, and even cause death; therefore, the urgency of the government's role in giving special attention to this case. A high potential case was found in farms with poor livestock management systems. Coccidiosis often does not show clinical symptoms; therefore, breeders and veterinarians have difficulty recognizing early symptoms in the field.

Aim: The purpose of the research is to identify and characterize the morphology of *Eimeria* spp.

Methods: Fresh fecal samples were obtained from 106 beef cattle from Bandung Regency. Examination of fecal samples using the floating method with saturated sugar. The shape of the oocyst was observed using an Optilab microscope and oocyst measurements were performed using Image Raster software.

Result: The examination results showed that 51 stool samples out of the 106 samples tested positive for *Eimeria* spp. The samples revealed eight different species of *Eimeria*.

Conclusion: Eight *Eimeria* spp. were identified from the samples taken from the Bandung regency and they were *Eimeria bovis*, *Eimeria zuernii*, *Eimeria alabamensis*, *Eimeria cylindrica*, *Eimeria auburnensis*, *Eimeria wyomingensis*, *Eimeria pellita*, *Eimeria canadensis*. The prevalence of beef cattle coccidiosis in Bandung Regency was 48.11%. Morphological determination can be used to diagnose *Eimeria* spp. based on shape, size, shape index, and characteristics.

Keywords: Beef cattle, Coccidiosis, *Eimeria*, Fecal, Morphological determination.

Introduction

Livestock sector development programs support the fulfillment of animal protein needs and improve the prosperity of farmers. Beefs are a source of animal protein. Data from the Bandung Regency Animal Husbandry Service shows a high population of beef cattle, namely 22,647 heads (in 2021), 22,332 heads (in 2022), and 20,812 heads (in 2023) (West Java Province Central Statistics Agency, 2023). Cattle with a maintenance management system that does not have the maximum impact on cattle health. Cattle are often malnutrition, easily infected with parasites and have reduced productivity. Coccidiosis is caused by *Eimeria* spp. which attacks the digestive tract of cattle, young cattle have acute diarrhea that can lead to death (Dauguschies and Najdrowski, 2005; Bruhn *et al.*, 2011). Coccidiosis is the most difficult condition

to control in the field (Dauguschies and Najdrowski, 2005). Coccidiosis is highly likely to be found on farms with poor rearing management systems. Direct transmission through feed, water, and other materials contaminated by sporulating oocysts. Coccidiosis can reduce productivity and economic losses, and the level of vulnerability to infection with different diseases is increasing.

Therefore, special attention from the government is required. According to Ibrahim *et al.*

(2015), coccidiosis is subclinical and can lead to sudden death.

Coccidiosis cattle in Sumedang regency, West Java was reported with a positive percentage of *Eimeria* spp. amounting to 14.29% of the total 105 samples (Ninditya *et al.*, 2024), in West Java 24% of the total 50 samples (Ekawasti *et al.*, 2021) and 22.4% of the

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total 394 samples and showing the highest prevalence compared to other gastrointestinal parasites (Ananta et al., 2014). Coccidiosis in dairy cattle in Bandung was obtained, with a positive prevalence of *Eimeria* spp. amounting to 179 samples (44.75%) from a total of 400 samples. Factors that influence the coccidiosis are age of livestock, medication, and type of bedding (Sufi et al., 2016). *Eimeria* spp. found in dairy cattle in Bandung included *Eimeria bovis* (42.5%), *Eimeria wyomingensis* (39.1%), *Eimeria bukidnonensis* (32.4%), *Eimeria pellita* (26.3%), *Eimeria auburnensis* (19.6%), *Eimeria zuernii* (17.3%), *Eimeria cylindrica* (3.9%), *Eimeria canadensis* (3.9%), *Eimeria brasiliensis* (3.4%), and *Eimeria alabamensis* (1.1%). Mixed coccidiosis infections in cattle usually contain 2–5 *Eimeria* spp. (Sufi et al., 2017). Coccidiosis in cattle in Central Java was reported with a positive percentage of *Eimeria* spp. amounting to 15.33% of the total 455 samples. *Eimeria* spp. found in cattle in Central Java include *E. bovis*, *E. auburnensis*, *E. bukidnonensis*, *E. canadensis*, *E. zuernii*, and *E. cylindrica* (Hamid et al., 2016). In cattle, there are approximately 20 species of *Eimeria* spp., pathogenic species can elicit clinical signs and symptoms such as *E. bovis*, *E. zuernii*, and *E. auburnensis* (Ekawasti and Wardhana, 2010; Kawahara, 2010). Nonpathogenic *Eimeria* spp. causes tissue harm, increases sensitivity to infection with other infectious diseases, but does not cause death (Ekawasti and Wardhana, 2010).

Research results in Central Java identified the presence of *Eimeria* spp. where almost all of the animals sampled in the research were raised using traditional farming methods (Hamid et al., 2016). This indicates that maintenance management is related to the presence of *Eimeria* spp.

Poor maintenance management has an impact on high infection of *Eimeria* spp. *Eimeria* infection opens the door for the entry of other disease agents. The importance of coccidiosis control strategies, especially the correct identification of inappropriate control strategies for *Eimeria* spp. in increasing cases of coccidiosis, oocysts can spread infection and can continually contaminate the environment (Gupta et al., 2016; Ekawasti et al., 2021). One method of detection of coccidiosis is through conventional techniques (float test), which is used to observe the morphology under a microscope.

Determination of coccidiosis using conventional methods by observing microscopic morphology is limited to the genus level. This identification method cannot be used as a reference for determining the species of *Eimeria* because each species has the same shape, structure, and size. Identification to determine morphological variations in cattle in Bandung regency has not been carried out. The importance of diagnostic methods in controlling coccidiosis in the field. Therefore, it is essential to carry out research to determine the morphological diversity of *Eimeria* spp.

because the pathogenicity of each species of *Eimeria* varies.

Materials and Methods

Fecal samples were collected from 106 beef cattle from three sub-districts (Cicalengka, Cikancung, and Nagreg), Bandung Regency, West Java Province, Indonesia. The sampling method with purposive sampling, was collected randomly from door to door, then recording the GPS coordinates of each sampling point location using a Geographic Information System approach. Samples were collected from farm to farm from January 2024 to March 2024. A total of 106 samples were categorized based on the age of beef cattle, namely < 1 year old six samples, aged 1–2 years, as many as 80 samples, aged >2 years as many as 20 samples. Overall, fecal has a normal texture. One fecal sample was obtained from one cattle. Fecal samples were taken from the cattle rectum, stored in a plastic bag, and then stored at 4°C until laboratory examination. Laboratory examinations are conducted in several laboratories, such as the Veterinary Technology Laboratory, Vocational College, Universitas Gadjah Mada, and the Parasitology Laboratory, Faculty of Veterinary Science, Universitas Gadjah Mada.

Microscopic examination

Feces were examined using a float test by adding saturated sugar with a specific gravity of 1.2 (Matsubayashi et al., 2005). One gram of stool sample was centrifuged at $800 \times g$ for 5 minutes. After removing the supernatant, adding a sugar solution with a specific gravity of 1.2 and centrifugation was carried out. After covering the tube's surface with a cover glass and moving it to an object glass, use an Optilab microscope to observe the sample. According to Soulsby (1986), the recognized species were determined. Qualitative microscopic analysis was performed using a Miconos Optilab microscope at a magnification of 10–100 to identify the presence of positive and negative *Eimeria* spp. oocysts. Next, measure the length and width to determine the shape index using Image Raster software.

Ethical approval

This research was approved by the Ethics Committee of the Faculty of Veterinary Medicine, Gadjah Mada University, Indonesia (Approval Number: 146/EC-FKH/int./2023).

Results

Beef cattle fecal samples from Bandung Regency were tested using the flotation method with the addition of saturated sugar and then observed under an optical microscope. Laboratory examination results showed morphological variations in *Eimeria* spp. oocysts, which can be seen in (Fig. 1).

A total of eight *Eimeria* spp. based on their morphology namely *E. bovis*, *E. zuernii*, *E. alabamensis*, *E. cylindrica*, *E. auburnensis*, *E. wyomingensis*, *E. pellita*, and *E. canadensis* could be identified (Fig. 1).

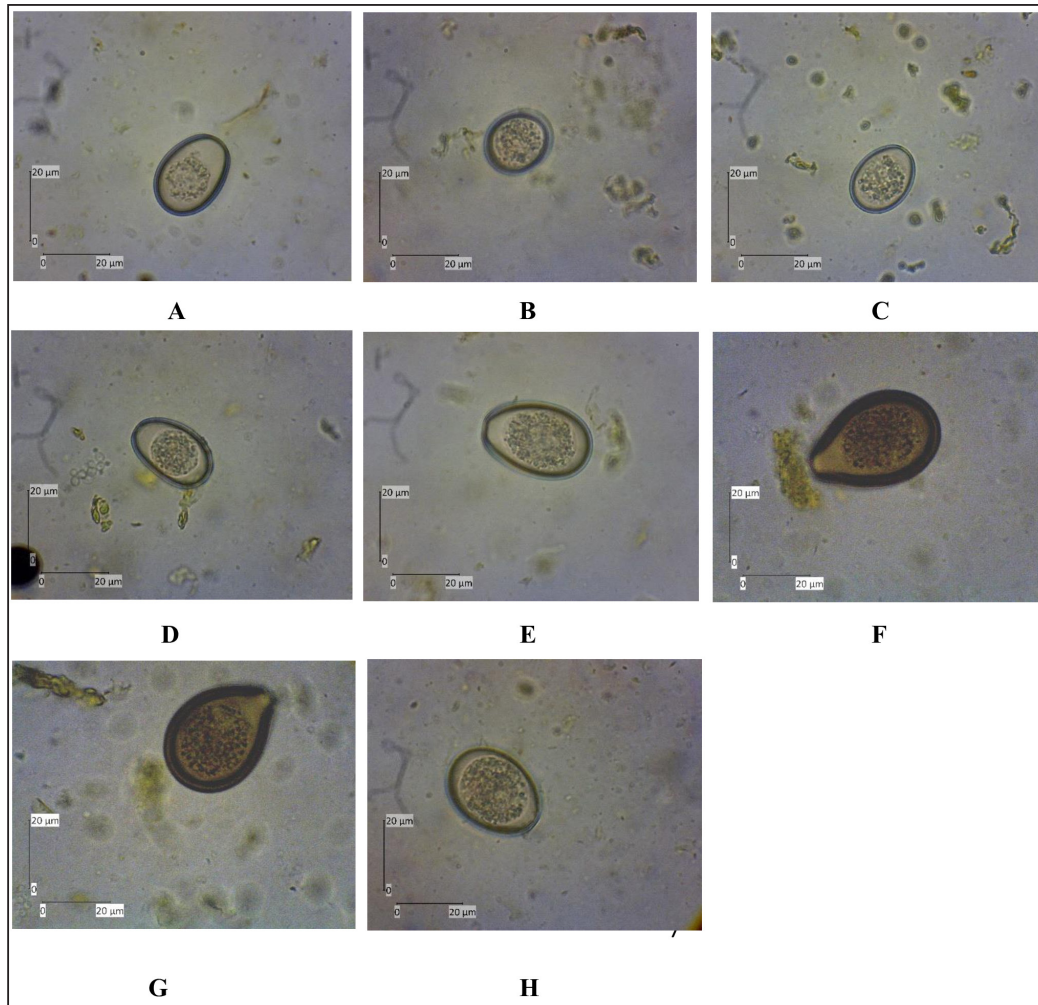


Fig. 1. Morphological variation of eight *Eimeria* spp. at 100 × magnification: A. *Eimeria bovis*, B. *Eimeria zuernii*, C. *Eimeria alabamensis*, D. *Eimeria cylindrica*, E. *Eimeria auburnensis*, F. *Eimeria wyomingensis*, G. *Eimeria pellita*, H. *Eimeria canadensis*.

As a result of research, eight species of *Eimeria* have been found to have different morphologies. *E. bovis* (ovoid, $25.68 \times 17.4 \mu\text{m}$, with micropyle), *E. zuernii* (sub-spherical, $17.73 \times 15.36 \mu\text{m}$, without micropyle), *E. alabamensis* (pear shaped, $20.69 \times 15.98 \mu\text{m}$, without micropyle), *E. cylindrica* (cylindrical, $24.14 \times 13.89 \mu\text{m}$, without micropyle), *E. auburnensis* (ellips, $33.09 \times 20.40 \mu\text{m}$, with micropyle), *E. wyomingensis* (ovoid, $39.63 \times 27.66 \mu\text{m}$, with micropyle), *E. pellita* (ovoid, $38.19 \times 28.39 \mu\text{m}$, with micropyle), and *E. canadensis* (ovoid/ellips, $30.28 \times 21.86 \mu\text{m}$, with micropyle) (Table 1).

51 stool samples out of the 106 samples tested positive for *Eimeria* spp., representing a 48.11% prevalence. The prevalence of *E. bovis* was 78.43% (40 samples), followed by *E. zuernii* 31.37% (16 samples), *E. alabamensis* 43.14% (22 samples), *E. cylindrica* 33.33% (17 samples), *E. auburnensis* 27.45% (14

samples), *E. wyomingensis* 13.73% (7 samples), *E. pellita* 11.76% (6 samples), and *E. canadensis* 11.76% (6 samples).

A total of eight *Eimeria* spp. were identified from cattle less than 1-year-old and from cattle 1–2 years old, namely *E. bovis*, *E. zuernii*, *E. alabamensis*, *E. cylindrica*, *E. auburnensis*, *E. wyomingensis*, *E. pellita*, and *E. canadensis*. Cattle older than 2 years old contained two species of *Eimeria*, namely *Eimeria cylindrica* and *Eimeria auburnensis* (Fig. 2).

The research showed that 51 samples (106 samples) were positive for *Eimeria* spp., with a single type of infection at 33.33% and multiple infections at 66.67%. A single infection is the infection of only one species of *Eimeria*. Multiple infections are caused by more than one species of *Eimeria* spp. Stool samples positive for *Eimeria* spp. in beef cattle aged < 1 year showed that all samples had multiple infections (100%), those aged

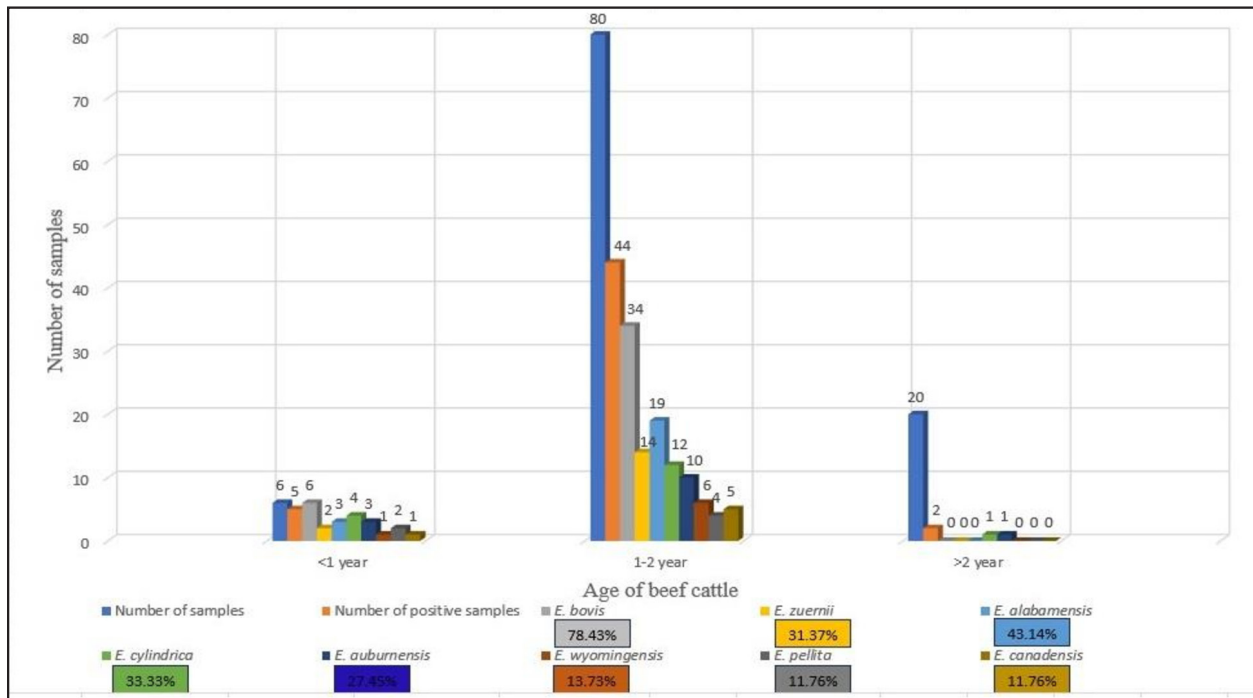


Fig. 2. *Eimeria* spp. prevalence and identification based on the age and prevalence of cattle.

Table 1. Determination of *Eimeria* spp. based on length, width, and shape index.

Species	Research in Bandung				Soulsby (1986)		
	Shape	Micropyle	Size (µm)	Shape index	Length (µm)	Width (µm)	Shape Index
<i>Eimeria bovis</i>	Ovoid	Present	25.68 × 17.4	1.48	23–34	17–23	1.35–1.48
<i>Eimeria zuernii</i>	Sub-spherical	No	17.73 × 15.36	1.15	15–22	13–18	1.15–1.22
<i>Eimeria alabamensis</i>	Pear shaped	No	20.69 × 15.98	1.29	13–24	11–16	1.18–1.44
<i>Eimeria cylindrica</i>	Cylindrica	No	24.14 × 13.89	1.74	16–27	12–15	1.67–1.80
<i>Eimeria auburnensis</i>	Ellips	Present	33.09 × 20.40	1.62	32–46	20–25	1.60–1.84
<i>Eimeria wyomingensis</i>	Ovoid	Present	39.63 × 27.66	1.43	37–44.9	26.4–30.8	1.40–1.46
<i>Eimeria pellita</i>	Ovoid	Present	38.19 × 28.39	1.35	36.1–40.9	26.5–30.2	1.35–1.36
<i>Eimeria canadensis</i>	Ovoid/ellips	Present	30.28 × 21.86	1.38	28–37	20–27	1.37–1.40

1–2 years had a single infection (34.09%), and multiple infections (65.91%), and those aged >2 years had a single infection (100%) (Fig. 3).

Discussion

Diagnosis of *Eimeria* spp. in the field is limited to the genus level because each species has almost the same shape and size. The determination of each

species was based on its shape, size, shape index, and characteristics. In Bandung Regency was reported variation of species *Eimeria* spp. in dairy cattle namely *E. bovis*, *E. wyomingensis*, *E. bukidnonensis*, *E. pellita*, *E. auburnensis*, *E. zuernii*, *E. cylindrica*, *E. canadensis*, *E. brasiliensis*, and *E. alabamensis* (Sufi et al., 2017). When compared to the author's research, Sufi et al. (2016, 2017) research was also carried out in the

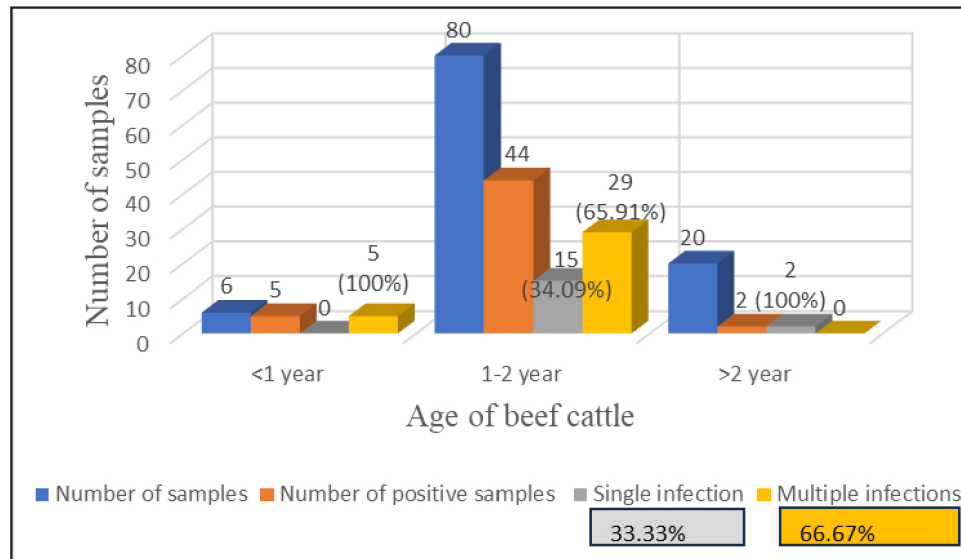


Fig. 3. Prevalence of *Eimeria* spp. according to the type of infection.

Bandung area, but with different subjects (using dairy cow). Other previous research, in central Java reported *Eimeria* spp. in cattle, namely *E. bovis*, *E. auburnensis*, *E. bukidnonensis*, *E. canadensis*, *E. zuernii*, and *E. cylindrica* (Hamid *et al.*, 2016). In Java, was reported *E. bovis*, *E. ellipsoidalis*, *E. alabamensis*, *E. zuernii*, *E. auburnensis*, and *E. cylindrica* (Ekawasti *et al.*, 2019). The author's research, Hamid's research and Eka's research were both carried out in the Java area. The author's research is in West Java province, Hamid's research is in Central Java province, while Eka's research covers a wider province in Java. No previous research publications have been found regarding *Eimeria* on beef cattle in the Bandung area (West Java). This research from sample cattle in Bandung regency obtained eight *Eimeria* spp. were *E. bovis*, *E. zuernii*, *E. alabamensis*, *E. cylindrica*, *E. auburnensis*, *E. wyomingensis*, *E. pellita*, and *E. canadensis*. Various species have different levels of pathogenicity. Pathogenic species include *E. bovis*, *E. zuernii*, and *E. auburnensis* (Ekawasti and Wardhana, 2010; Kawahara, 2010). *E. bovis* and *E. zuernii* have high pathogenicity and can cause death, especially in young cattle (Rehman *et al.*, 2011; Koutny *et al.*, 2012). Non-pathogenic species impact tissue damage and increase sensitivity to other infections (Ekawasti and Wardhana, 2010).

Previous research in Bandung Regency, *E. bovis* in dairy cattle had the highest prevalence of 42.5%, other species were *E. wyomingensis*, *E. bukidnonensis*, *E. pellita*, *E. auburnensis*, *E. zuernii*, *E. cylindrica*, *E. canadensis*, *E. brasiliensis*, and *E. alabamensis* (Sufi *et al.*, 2017).

Then in Java, *E. bovis* in beef cattle had the highest prevalence of 10.4%, other species were *E. ellipsoidalis*,

E. alabamensis, *E. zuernii*, *E. auburnensis*, and *E. cylindrica* (Ekawasti *et al.*, 2019). This research shows that the *E. bovis* species has the highest prevalence in the farms (78.43%) compared to other *Eimeria* spp. *E. bovis* is one of the most common *Eimeria* pathogens with the highest frequency in the field.

Eimeria spp. can infect cattle alone or in mixed infections (Ekawasti *et al.*, 2019), in mixed infections usually 2-5 *Eimeria* spp. can infect one cattle simultaneously (Sufi *et al.*, 2017).

According to the study, 71.2% of cattle had multiple *Eimeria* spp. infections (Morgoglione *et al.*, 2020). This research showed the same results, all research samples positive for *Eimeria* spp. had higher mixed infections (66.67%) than single infections (33.33%). The results of the research showed that cattle with age category <1 year had a coccidiosis percentage of (83.33%), 1–2 years (55%), and >2 years (10%). Based on age category, cattle aged <1 year have the highest percentage of infection, which is related to the immune system not being optimal.

Many studies have reported the prevalence of coccidiosis in cattle from different countries (Heidari *et al.*, 2014). The frequency of coccidiosis varies across nations; the prevalence of *Eimeria* spp. is 96% in the US, 83.67% in Austria, 75.5% in Colombia, 70% in the UK, 47.09% in Pakistan, 30% in Kenya, 29.2% in Ethiopia, 22.1% in South Korea, and 11.97% in India (Rehman *et al.*, 2011; Peter *et al.*, 2015; Alemnew *et al.*, 2017; Lee *et al.*, 2018). The research demonstrates the prevalence of *Eimeria* spp. is (48.11%), previous research on fecal samples of cattle in Sumedang regency, West Java shows a prevalence of (14.29%) (Ninditya *et al.*, 2024), beef cattle in West Java (22.4%) (Ananta *et al.*, 2014), dairy cattle in Bandung (44.8%) (Sufi *et al.*,

2017), beef cattle in Central Java (15.33%) (Hamid *et al.*, 2016), and beef cattle in Java (52.3%) (Ekawasti *et al.*, 2019). This difference includes livestock management, geographical conditions, food sources, feeding behavior, detection methods, and sampling time (season) (Eckert *et al.*, 2005; Gupta *et al.*, 2016). Stress, dosage challenges, and immune status can influence coccidiosis in cattle. Any treatment, control, or prevention program must coincide with problem management. All equipment, including drinking and eating containers must be cleaned to prevent fecal infection. The change in diet is carried out gradually considering economic aspects and avoiding overcrowding of livestock (Gupta *et al.*, 2016). One aspect of livestock management is the type of cage bedding. Most cattle in Bandung were positive for *Eimeria* spp. and kept in wooden cages. According to Bangoura *et al.*, (2011) and Rehman *et al.*, (2011), cage bedding type affects the frequency of oocyst excretion in the environment. The incidence of coccidiosis in cattle is higher in cages without cement than in cages using cement. Because the cage base with cement is easier to clean, it reduces the risk of infection with *Eimeria* from the environment. Most cattle in Bandung were positive for *Eimeria* spp., with more than five individuals housed in a single pinfold. These conditions facilitate coccidiosis transmission. Environmental cleanliness significantly influences the spread of coccidiosis. In beef cattle, providing adequate nutrition in both quantity and quality impacts the cattle's immune system. Cattle have higher resistance to *Eimeria* spp. infection. The cattle's overall body condition has a body condition score of 3–3.5, that range means that the cattle have sufficient nutrition.

Transmission of *Eimeria* spp. via ingestion of sporulating oocysts. Infected livestock shed oocysts of *Eimeria* spp. in fecal form, which are resistant to environmental conditions and most disinfectants. Oocysts have a long lifespan, and afterward, can have the ability to spread illness over the surrounding area (Lucas *et al.*, 2014; Ekawasti *et al.*, 2021). Direct or indirect infection in cases of coccidia occurs by the fecal-oral pathway and is detected by finding oocysts. According to Gupta *et al.* (2016), coccidiosis is among the top five diseases in the livestock industry in terms of economic impact. Acute diarrhea causes approximately 75% of all deaths, resulting in financial losses. Calves raised in traditional systems have the highest prevalence; these calves contact the infection at a young age (Fadly, 2012). *Eimeria* spp. has a specific host, in cattle more than 20 *Eimeria* spp. have been found (Heidari *et al.*, 2014). *E. bovis* and *E. zuernii* are pathogenic species, that can cause morbidity and death, affect the physiology of intestinal absorption, and cause diarrhea with fibrin and blood (Ekawasti *et al.*, 2021). Only a few fecal samples were used in this study, and morphological identification was limited to the species level. Further research is recommended by increasing

the number of samples and widening the scope of sampling locations. Other than that, further research is essential to determine drug resistance and the level of pathogenicity of species so that livestock productivity increases through good livestock management practices. In conclusion, eight *Eimeria* spp. were identified in the samples taken from the Bandung regency were *E. bovis*, *E. zuernii*, *E. alabamensis*, *E. cylindrica*, *E. auburnensis*, *E. wyomingensis*, *E. pellita*, and *E. canadensis*.

The prevalence of beef cattle coccidiosis in Bandung Regency was 48.11%. Morphological determination can diagnose *Eimeria* spp. based on shape, size, shape index, and characteristic.

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Conflict of interest

The authors declare that they have no conflict of interest.

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Author contributions

FRH, RWN, DP, SI, FE, and VIN designed this study. FRH, FE, and VIN field research and sample analysis in the laboratory. All authors wrote, edited, read, and approved the final manuscript.

Data availability

All information supporting the discoveries of this consideration is accessible inside the manuscript.

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