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IDENTIFICATION OF GASTROINTESTINAL PROTOZOA OF SUMATERA ELEPHANT (*Elephas maximmus sumatranus*) IN LOMBOK WILDLIFE PARK

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ABSTRACT: The protozoan parasites have been reported to infect Sumatran elephants. Gastrointestinal protozoa could potentially be a factor in the decline in the Sumatran elephant population in Indonesia. This study aims to identify the presence of gastrointestinal protozoa in Sumatran Elephant (Elephas maximus sumatranus) in the Lombok Wildlife Park, North Lombok Regency, Indonesia. This research has used fresh fecal samples from 5 Sumatran elephants. The examination of feces samples using native, sedimentation, and floating methods. The research results have identified the presence of gastrointestinal protozoa in 2 of the 5 Sumatran elephant feces examined. The gastrointestinal protozoa found were Eimeria spp with dimensions of 16.30 x 20.93 µm and 25.21 x 38.49 µm.

Keywords: *Eimeria* spp., Lombok Wildlife Park, Sumatran Elephant.

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INTRODUCTION

Lombok Wildlife Park is a conservation institution that conserves several species of animals, such as Elephants, Orangutans, Hippos, Crocodiles, Siamang, Langur, Macaca, Birds, Komodo, Sun Bears, and various other types of animals (LPW, 2024). Lombok Wildlife Park is also the only ex-situ conservation

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institution for the Sumatran elephant (*Elephas maximus sumatranus*) on Lombok Island. The existence of Sumatran elephants in the Lombok Wildlife Park needs to be preserved because the elephant population has decreased. Sumatran elephants are included in the Red List Data Book of the International Union for Conservation of Nature and Natural Resources (IUCN) (Gopala *et al*., 2011).

The decline in the population of Sumatran elephants (*Elephas maximus sumatranus*) has occurred in Indonesia, this population decline started from the narrowing of habitat, illegal hunting, a conflict between humans and elephants, as well as infection with various diseases such as viruses, bacteria, and parasites. Soehartono *et al*. (2007) reported that the Sumatran elephant population decreased by around 35% to 2,400-2,800 individuals. In 2014, the natural population of Sumatran elephants is estimated to continue to decline until only 1,800 individuals remain. The decline in the elephant population is caused by disturbances in elephants which can also occur as a result of various diseases such as viruses, bacteria, and parasites which can result in the death of these animals (Fowler and Mikota, 2006). One of the diseases that often attacks Sumatran elephants is gastrointestinal parasites which have not yet been reported on Lombok Island.

Protozoan parasites from two protozoan families *Eimeriidae* and *Ophryoscolecidae* have been reported to infect Sumatran elephants (*Elephas maximus sumatranus*) in Lampung with a prevalence of 41.8% for elephants aged 1-3 years, 47.2% for elephants aged 19-36 years (Herdaus *et al.*, 2015). Protozoal infections have a big role in affecting elephant health because they can cause a decrease in nutrient absorption and death. Astiti *et al*., (2011) stated that gastrointestinal parasites including protozoa can infect the digestive tract of animals, resulting in a decrease in nutrient absorption and causing growth delays (Rahmi *et al*., 2010). The presence of gastrointestinal protozoa in elephants can cause infection to other animals because *Eimeria* in the form of cysts will be able to survive in the environment in tropical climates such as in Indonesia so that it can infect other animals. Parasitic infections have become a serious obstacle to livestock health and productivity in tropical and sub-tropical regions of the world (Kumar *et al*., 2016).

Identification of protozoa in Sumatran elephants is one way to take action to break the life cycle of protozoa because some protozoa such as *Eimeria* which have infected elephants in Indonesia have various types and specific hosts. Dubey (2018) stated that Eimeria species are generally considered to have specific hosts and more than 11 Eimeria species are considered common in buffalo and cattle. Data regarding gastrointestinal protozoa in Sumatran elephants in Lombok Wildlife Park has not been reported, so research was carried out to identify the presence of gastrointestinal protozoa in Sumatran elephants (Elephas maximus sumatranus) in Lombok Wildlife Park which is the only ex-situ conservation institution for Sumatran elephants. Data about protozoa can be used as initial data regarding the health status of Sumatran elephants in Lombok Wildlife Park.

METHOD

This type of research is descriptive observational. Observations regarding the presence of gastrointestinal protozoa were carried out on the population of

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Sumatran elephants (*Elephas maximus sumatranus*) located in Lombok Wildlife Park (LWP), North Lombok Regency, North Lombok Regency, West Nusa Tenggara Province, Indonesia in January 2020.

The population samples for this research were feces from 5 Sumatran elephants. The sampling method in the research is the census method. Census sampling or census method is a sampling technique in which all members of the population are used as samples. The samples for this research were feces from 5 Sumatran elephants. A 200-gram sample of fresh feces from a Sumatran elephant was taken and put into a plastic bottle to which 2% potassium dichromate was then added until the feces sample was submerged and labeled (Herdaus *et al*., 2015). Sumatran elephant feces samples were then immediately taken using a cool box to the Microbiology and Parasitology Laboratory, Faculty of Veterinary Medicine, Universitas Pendidikan Mandalika for examination. Examination of gastrointestinal protozoa in Sumatran elephant feces was carried out using native, sediment, and floating methods. Identification of oocysts in feces using a modified sugar flotation method has been reported to be very sensitive for detecting *Eimeria* spp (Ekawasti *et al*., 2019).

The native method is carried out by taking a lump of Sumatran elephant feces, then placing it on a glass object, dropping a drop of water using a plastic pipette and flattening it, covering it with a covered glass, examining it with a microscope at 10x magnification (Taylor *et al.*, 2007). The sedimentation method is carried out by taking ± 2 grams of elephant feces and putting it in a mortar, then adding a little water to the feces and homogenizing it. The suspension is poured into a centrifuge tube until it is ¾ of the tube high. Next, it was centrifuged for five minutes and the clear liquid above the precipitate was discarded, then saturated NaCl was added to the precipitate ¾ of the tube high and homogenized. After that, it was centrifuged again at a speed of 1600 rpm for five minutes. The centrifuge tube was placed on the tube rack perpendicularly, then saturated NaCl was added using a pasture pipette until the surface of the liquid in the centrifuge tube became convex and left for 3 minutes, then a glass object was attached to the convex surface of the centrifuge tube carefully, then immediately reversed. Finally, it was covered with a cover glass and observed under a microscope (Taylor *et al.*, 2007).

The floating method is carried out by taking 3 grams of Sumatran elephant feces, putting it in a beaker, and adding 30 ml of water so that the concentration is 10%, then stirring until homogeneous. Next, it is filtered to remove large parts, then the results are collected in another beaker. The filtrate was put into a centrifuge tube until ¾ of the volume of the tube, then centrifuged at a speed of 1,500 rpm for 2-3 minutes, the centrifuge tube was removed, the supernatant was discarded so that the sediment remained, and then a floating solution was added to ¾ of the tube volume, then stirred until the sediment was homogeneous. This suspension was centrifuged at a speed of 1,500 rpm for 2-3 minutes. The centrifuge tube is carefully removed from the centrifuge and then placed on the test tube rack in an upright position. Next, the floating liquid is added slowly using a Pasteur pipette until the surface of the liquid is convex, and waited for 1-2 minutes to give the parasites a chance to float to the surface. The glass cover is

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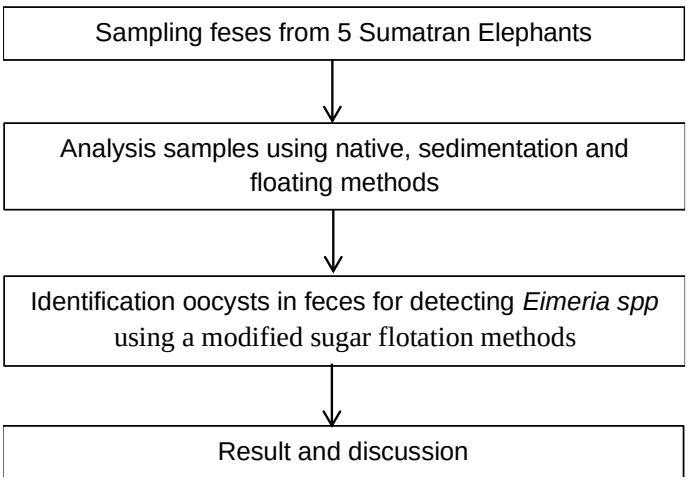
taken, then touched to the convex surface of the liquid, and then placed on top of the object glass. Examination was carried out with a microscope with 400X magnification (Taylor *et al*., 2007). Data from the microscopic examination were analyzed descriptively regarding the morphology of intestinal protozoa in Sumatran elephants (*Elephas maximus sumatranus*) in Lombok Wildlife Park, North Lombok Regency, presented in the form of tables and figures.

The research flow diagram is describe as follows:



RESULTS AND DISCUSSION

Based on the results of microscopic laboratory examinations in identifying gastrointestinal protozoa in 5 samples of Sumatran elephant (Elephas maximus sumatranus) feces located in Lombok Wildlife Park (LWP) North Lombok Regency, West Nusa Tenggara Province using native, sedimentation and floating methods, gastrointestinal protozoa could be found. type Eimeria spp. In 2 samples of Sumatran elephant feces. The results of the examination are presented in Table 1 and Figure 1.

Table 1. The results of the feces examination of Sumatran elephant (*Elephas maximus sumatranus*).

Number	Type of sample	Results	Species of Protozoa
1.	Feces of Elephant 1	Negative	-
2.	Feces of Elephant 2	Positive	*Eimeria spp*
3.	Feces of Elephant 3	Positive	*Eimeria spp*
4.	Feces of Elephant 4	Negative	-
5.	Feces of Elephant 5	Negative	-

The research results in Table 1 show that *Eimeria* spp was found in Sumatran elephant feces samples no. 2 and 3. The results of the microscopic examination in Table 1 which shows the presence of *Eimeria* spp protozoa of 40% in 2 of 5 Sumatran elephant feces samples shows that the *Eimeria* spp infection in the Wildlife Park (LWP) is almost the same as the presence of *Eimeria* spp in Sumatran elephants in Aek Nauli Elephant Conservation Camp (ANECC), North Sumatra, which has identified the presence of *Eimeria* spp in 2 Sumatran elephant feces samples out of 4 feces samples examined (Melia *et al*., 2020).

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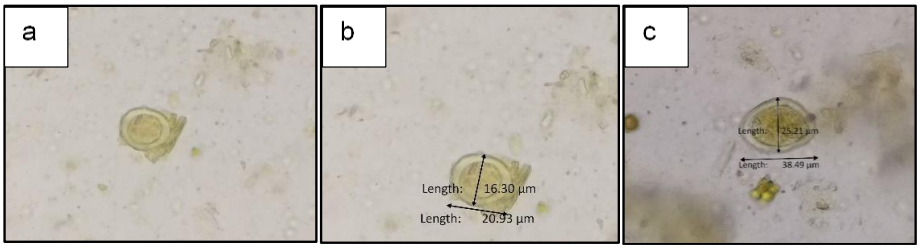

Figure 1. Morphology of *Eimeria* spp on a microscope with 400x magnification; (a) morphology of *Eimeria* spp without size, (b), morphology of *Eimeria* spp with the smallest size, (c) morphology of *Eimeria* spp with the largest size.

The results of protozoan identification in Sumatran elephants at Lombok Wildlife Park (LWP) (Figure 1) show that *Eimeria* spp has an oval-shaped morphology with the smallest size being 16.30 x 20.93 µm, while the largest size is 25.21 x 38.49 µm. The morphology of *Eimeria* spp with the smallest size in this study is almost the same as *Eimeria bovis* found in cattle in Madura which has a size of 25.4 × 17.2 µm, while the largest size is almost the same as *Eimeria canadinensis* with a size of 29.4 × 20.3 µm (Hastutiek *et al*., 2022). The morphological similarities of eimeria that have been found in research with *Eimeria bovis* and others make it possible for contamination from the environment, considering that Lombok Island is one of the islands that supplies beef cattle. Since 2013 in East Lombok have been around 504 livestock farmers, 238 of 504 livestock farmer groups were livestock groups (Kholik *et al*., 2023).

According to Bush *et al*. (2001), *Eimeria* belongs to *Phylum: Apicompleka, Class: Sporozoa, Order: Eucoccidlorida, Sub Order: Eimeria*. The *Eimeria* spp. that have been identified in Figure 1 shows *Eimeria* spp. at the oocyst stage which is found in a non-sporulating state. This sporulation time is the time it takes for the oocysts to come out with the feces and will occur within 2-4 days. Raslina et al. (2018) and Taylor et al. (2016), stated that the time required for these changes varies according to temperature, but under optimal conditions, it usually takes 2-4 days. *Eimeria* spp. only requires a single host to complete the entire life cycle process (Soulsby, 1986). The life cycle of *Eimeria* spp. can occur inside the host's body (endogenous) and outside the host's body (exogenous), the eimeria life cycle consists of several phases, namely schizogony, gametogony, and sporogony.

The presence of *Eimeria* spp in Sumatran elephants in Lombok Wildlife Park (LWP) can be caused by several factors, such as seasonal conditions, temperature, humidity, and management of the elephant enclosure. The tropical climate in Lombok with cold or rainy winters can be a factor in the existence of *Eimeria* spp. where humid conditions when it rains are very good conditions for sporulation. The sporulating oocysts that survive in the environment also favor bacterial transmission (Daugschies and Najdrowski, 2005).

Oocysts excreted in feces require moist environmental conditions and low temperatures for sporulation, while high temperatures can inhibit the optimal temperature for sporulation of most *Eimeria* spp. Under various environmental conditions, it is between 29-33 ℃ (Graat *et al*., 1994). Other factors can also be caused by the feed or drinking water given to elephants, the feed given to

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elephants in Lombok Wildlife Park is rice straw, feed, or drinking water that has been contaminated by *Eimeria* spp. through feces is very beneficial for the protozoa *Eimeria spp*., so that oocysts can speculate and become infective. Infections from animals that ingest oocysts along with contaminated feed and drinking water are highly infectious and can easily spread throughout animal populations. This situation is supported by Hasbullah *et al*. (1990) reported that cow feces samples that were positive for coccidial oocysts were 12.9% and 26.7% on university farms obtained during grazing in pastures and loose housing.

Eimeria spp has been identified in Sumatran elephants in Lombok Wildlife Park and could pose a threat to the health and sustainability of these elephants because *Eimeria* spp. can cause damage to the intestinal epithelium and connective tissue of the intestinal mucosa, accompanied by bleeding in the intestinal lumen which can cause a decline in the elephant's health and can even cause death. Lassen and Jarvis (2009) revealed that the clinical symptoms of *Eimeria* spp. with high pathogenicity in the form of bloody diarrhea, fever, abdominal pain, tenesmus, anemia, dehydration, anorexia, and can cause death. However, these symptoms were not found in the elephants that were observed. The elephants that were observed showed normal conditions. In this condition, it can be said that *Eimeria* spp. those who infected the elephant were not in a state of high pathogenicity.

The habit of elephants when defecating in any place with poor sanitary conditions supported by the tropical climate is very likely to cause infection of *Eimeria* spp. of Sumatran elephants in the Lombok Wildlife Park so regular sanitation and treatment greatly influences the health status of the elephants. The warm and humid tropical climate in Indonesia has been stated to be a risk factor for *Eimeria* infection, this is demonstrated by the occurrence of *Eimeria* spp. widespread in Indonesia with a prevalence of 65.4% on cattle farms (Ekawasti *et al*., 2021). The incidence of *Eimeria* infection can be reduced if the Lombok Wildlife Park increases biosecurity measures and better management practices, such as maintaining cleanliness, including sanitation, and monitoring food management. Additionally, due to the large number of species that can infect elephants, it is important to identify which species are more frequently encountered for control and treatment measures.

CONCLUSION

Protozoa of the *Eimeria spp* have been identified in 2 of the 5 feces of Sumatran elephants (*Elephas maximus sumatranus*) in Lombok Wildlife Park, North Lombok Regency, West Nusa Tenggra Province, Indonesia. The *Eimeria* spp found had an oval shape with the smallest size being 16.30 x 20.93 µm, while the largest size was 25.21 x 38.49 µm. The incidence of *Eimeria* infection in Sumatran elephants may be reduced by increases in biosecurity measures and better management practices.

SUGGESTION

The authors hopes that this research can be continued periodically and on other mammal spesies in Lombok Wildlife Park so that the health of existing animal can be maintained.

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