PREVALENCE OF *Balantidium sp* IN BALI CATTLE AT DIFFERENT AREAS OF BALI

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ABSTRACT

Balantidiasis is a protozoal zoonotic disease caused by Balantidium sp. which can be transmitted from animals to humans reversibly. It can cause a serious health problem, both in animal and human, including anorexia, persistent diarrhea, haemorraghic diarrhea, and dehydration, which can lead to death. In order to decrease the incidence of balantidiasis, especially in cattle, the study about pathology and epidemiology of Balantidium sp. infection should be conducted. The aim of this study is to determine the prevalence of Balantidium sp. infection in bali cattle (Bos sondaicus) in Bali Province and related risk factor including the origin of cattle, rainfall factor, maintenance system, sex and age of the cattle. This study was an observational study using cross-sectional design. A total of 320 feces of bali cattles had been used in this study, which were derived purposively from eight area in the province of Bali. Feces examination was conducted using float concentrations method in Laboratory of Parasitology, Faculty of Veterinary Medicine, Udayana University. Data were then analyze using descriptive statistic, continued by Chi-square (χ^2) test to determine the correlation between risk factor and the prevalence of balantidiasis. Moreover, odds ratio test was performed to measure the intensity or the degree of association between a risk factor and prevalence of Balantidium sp infection. The result showed that the prevalence of Balantidium sp infection in bali cattle (Bos sondaicus) in Bali Province was 17.19% in general. There was a significant relationship between the origin of cattle ($p<0.001$; OR,4.0; 95% CI, 2.07-7.85), rainfall factor ($p<0.001$; OR,2.9; 95% CI, 1.54-5.45), maintenance system ($p<0.001$; OR,8.9; 95% CI, 4.40-18.32), and sex ($p<0.05$; OR,1.9; 95% CI, 1.06-3.43) with the prevalence of Balantidium sp infection in bali cattle (Bos sondaicus) in Bali Province. But there was no correlation between the age of cattle with the prevalence of Balantidium sp infection in bali cattle (Bos sondaicus) in Bali Province ($p>0.05$). It is concluded that cattle are highly suspectible to balantidiasis irrespective of origin of cattle, environment rainfall, maintenance system, and sex.

Keywords : prevalence, Balantidium sp., bali cattle, Bali province.

Introduction

Bali cattle is an important animal as a source of protein and potential to fulfill the community needs. Bali cattle have a high economic potential than other beef cattle in Indonesia due to various advantages such as high fertility rates of 80%, the carcass percentage of 56-60% and the daily weight gain up to 0.35 kg/day. It is a major cause of the increasing demand for beef each year, proved by Central Statistical Agency report (2015) that showed the increase number of beef production of 6283 tons in 2009 to 9041 tons in 2014. However, there is some obstacle faced by farmers, one of which is a disease both infectious and non-infectious. Protozoan diseases have great importance in ruminants and other animals. Among the protozoan diseases balantidiasis caused by *Balantidium sp*, is a common disease of ruminants (cattle, buffaloes, sheep and goats), pig, monkey,chimpanzee, orangutan, guinea pig and man (Levine, 1994). *Balantidium sp* naturally inhibitats in the caecum, colon and rectum of of

apparently healthy animals, but under certain circumstances it produces clinical disease (Schuster and Ramirez-Avila, 2008). Infection occurs through ingestion of food or water contaminated with cysts of *Balantidium sp*. Cyst is the infective form of *Balantidium sp*, non-reproductive, and has a function to survive. Once the cyst reaches the small intestine, trophozoites are produced which called the excystation. The trophozoites then colonize the large intestine, where they live in the lumen and feed on the intestinal flora (Urquhart, *et al*., 1996; Ahmad, 2011). *Balantidium sp* fundamentally affects the colon and causes clinical manifestation from asymptomatic to serious dysenteric forms (Lazar, *et al*., 2004). *B. coli* also produces hyaluronidase (Tempelis and Lysenko, 1957) which potentially enhancing its ability to invade the intestinal mucosa, causing enteritis where the clinical features are manifested by loose faeces to watery persistent foetid diarrhea, dehydration, loss of appetite, retarded growth, loss of body condition and reduced production performance of the animals (Roy, *et al.*, 2011). The epidemiology of *Balantidium sp* infection is influenced by three factors, which are the host, the agent and the environment. Host factor consist of the resistance, species, breed, age, immune system, nutrition status, and sex of bali cattle. The agent factor in this case is pathotype, infectivity, pathogenicity, virulence, immunogenicity, and viability of *Balantidium sp*. While feeding, temperature, humidity, and maintenance system are included as an environment factor (Budiharta and Suardana, 2007). Bali has an area of $\pm$5621 km^2 which divided into lowland area (altitude of 0-500m above sea level, average air temperature of 27.2°C) and plateau area (altitude of above 500m, average air temperature of 18.9°C) (Suweta, 1982). There has no study on the prevalence of *Balantidium sp* infection in bali cattle that ever been done. The present study was undertaken to study the prevalence of *Balantidium sp* infection (balantidiasis) in cattle in different areas of Bali Province.

Methods

This study was an observational study using cross-sectional design. A total of 320 feces of bali cattles had been used in this study, which were derived purposively from eight area in the province of Bali. Sample in each area of Bali Province were selected randomly irrespective of maintenance system, sex and age of the cattle. The origin of cattle was classified into two groups, namely, lowland (0-500 meter asl) and plateau (above 500 meter asl). Morover, the rainfall of which bali cattle originally from was grouped into dryland and wetland. The maintenance system was also divided into two categories, namely, free stalls (with outdoor access) and tie stalls (without outdoor access). According to age, cattle were categorized into two groups, namely, young ($\leq$ 18 months) and adult (> 18 months). The research was carried out for a period from June to July 2015. Stool examination was conducted at the Laboratory of Parasitology, Faculty of Veterinary Medicine Udayana University. The collected faecal samples were examined by float concentrations method for determining the appearance of *Balantidium sp* infection by the cysts or trophozoites characteristic morphological features as described by Soulsby, (1982) and Levine, (1994) (Figure.1 and Figure.2).

Fig.1. Cyst of *Balantidium sp* in cattle

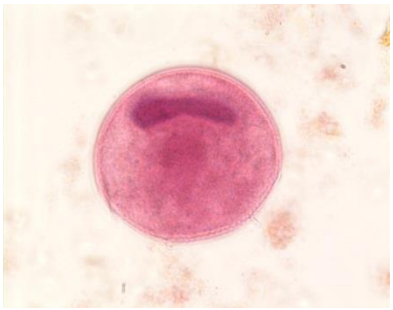
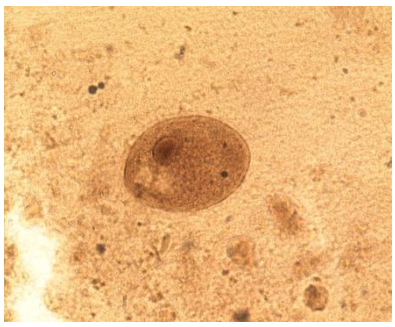


Fig.2. Trophozoit of *Balantidium sp* in cattle



The data obtained were reported descriptively. Statistical analysis was done by using the Statistical Package for the Social Sciences (SPSS-17.0 version) software (Sampurna and Nindhia, 2013). Chi-square (χ^2) test was performed for testing the significance of several prevalence of *Balantidium sp* infection at 1% or 5% level of significance with the risk factor such as origin of cattle, rainfall factor, maintenance system, sex and age of the cattle. In addition, odds ratios test was performed to measure the intensity or the degree of association between a risk factor and prevalence of *Balantidium sp* infection.

Results and discussion

Overall prevalence of *Balantidium sp* infection in cattle

During the study period, a total of 320 cattle spread over eight regions in Bali Province were examined through faecal examination, of which 55 (17.19%) were found to be infected with *Balantidium sp* (Table 1). The prevalence of balantidiasis in cattle has never been reported in Indonesia. However, some study have reported the presence of *Balantidium sp* infection swine in Central Java, Indonesia which was 5 swine were positive out of 60 swine (8.3%) (Sulistiningari, 2003), and reached 38% in pigs in Baliem and Arfak Mountains, Papua (Yuliari, *et al*., 2013). In addition, another study mentioned that balantidiasis cases was high enough in pigs in Bali, which reached 62% (Damriyasa, *et al*., 2002). The high prevalence of *Balantidium sp* infection in pigs in Bali was suspected to have an impact on the high prevalence of *Balantidium* sp in cattle, since the disease is a zoonotic

disease that can be transmitted from one species to another. The prevalence of *Balantidium sp* in cattle in Bali was lower than in Lahore, Pakistan, which reached 20% in 2008 (Tarrar, *et al*., 2008) and 25% in 2009 (Bilal, *et al*., 2009). Other studies have also reported that the prevalence of *Balantidium sp* reached 45.03% in Mymensingh, Bangladesh (Roy, *et al*., 2011), and reached 45.45% in India (Palanivel, *et al*., 2005). However, the result of this study was higher when compared to balantidiasis prevalence in Punjab, India which only 2.52% (Singh, *et al*., 2012).

The variations between the present and previous findings might be due to difference in the sample size, selection of samples, technique of sample examination, geographical locations/ topography, climatic condition, management and nutritional factors of the animals (Roy, *et al*., 2011).

Table 1. Prevalence of *Balantidium sp* infection

Risk Factor	Category	No. of sample examined (N=320)	No.of sample positive for *Balantidium sp*	Prevalence (%)	Overall Prevalence (%)
Origin of Cattle	Plateau	160	42	26.25	17.19
	Lowland	160	13	8.12	
Rainfall	Wetland	160	33	24.37	
	Dryland	160	16	10.00	
Maintenance system	Free stalls	279	33	11.83	
	Tie stalls	41	22	53.66	
Sex	Male	116	27	23.20	
	Female	204	28	13.72	
Age	(≤ 18 months)	142	22	15.49	
	(> 18 months)	178	33	18.54	

Prevalence of *Balantidium sp* infection in cattle in relation to origin of cattle

The origin of cattle has considered to had an effect on the prevalence of *Balantidium sp* infection. A total of 320 cattle was used in this study, which were took from plateau area as much as 160 cattle, and the rest of 160 cattle were took from lowland area. The result revealed that the prevalence of balantidiasis in plateau area was 26.25%, whereas in lowland area the prevalence of *Balantidium sp* infection was just 8.12% (Table.1).

Chi-square (χ^2) test results showed there was a very significant correlation (P <0.001) between the origin of cattle and the prevalence of *Balantidium sp* infection (Table 2). Odds ratio test revealed that cattle in plateau area were 4.025 times more likely to be suspectible to infection than that cattle in lowland area (Table 2). Results of this study was contrast with the theory of the optimum temperature for *Balantidium sp* growth which tend to lowland area (20°C-40°C; an average of 30°C) (Clark and Diamond, 2002), while the average temperature of plateau area in Bali is 18.9°C. However, this result indicates that *Balantidium sp* are capable to adapt with plateau area temperature so that do not disrupt its reproductive cycle. It supported by research report indicated that *Balantidium elongatum*, a gastrointestinal endoparasit of amphibians, was able to adapt well at low temperatures (18°C) (Poljansky and Sukhanova, 1965).

Prevalence of *Balantidium sp* infection in cattle in relation to rainfall factor

This study revealed that rainfall condition in which cattle was maintained had an effect on the prevalence of *Balantidium sp* infection. The prevalence of balantidiasis in wetland area was 24.37%, whereas in dryland area the prevalence of *Balantidium sp* infection was just 10.00% (Table 1). Significantly (p<0.05) higher prevalence of balantidiasis in cattle was observed in wetland compared to dryland and cattle in wetland were 2.901 times more prone to infection that that of dryland (Table 2).

The results were similar with previous research stated that the prevalence of *Balantidium sp* infection was higher in the high rainfall and high humidity area. The highest prevalence in wetland may be due the high humidity, heavy rain fall and higher chance of contamination of feeds that enhance the rate of infection of *Balantidium sp* (Roy, *et al*., 2011). Until nowadays, there were no report on the optimum humidity and rainfall for *Balantidium sp* growth. Nevertheless, there has so many studies proved that heavy rainfall is appropriate for the parasite growth especially gastrointestinal protozoa (Azhar, *et al*., 2002; Mamun, *et al*., 2011; Roy. *et al*., 2011).

Prevalence of *Balantidium sp* infection in cattle in relation to maintenance system

During this study, it was observed that the prevalence of *Balantidium sp* infection was higher in cattle reared with free stalls system (53.66%)) than that of animals reared with tie stalls system (11.83 (Table 1). Chi-square (χ^2) test results showed there was a very significant correlation (P <0.001) between the maintenance system of cattle and the prevalence of *Balantidium sp* infection (Table 2). Odds ratio test revealed that cattle reared with free stalls system were 8.982 times more likely to be suspectible to infection than that cattle reared with tie stalls system (Table 2).
Cattle reared with free stalls system have a higher risk of *Balantidium sp* infection, as transmission of *Balantidium sp* due to contamination of the food by infective cysts. A free cattle is tend to seek for food in already contaminated grass, whereas cattle

in the tie stalls is fed properly. Moreover, the farmer of tie stalls management system, usually maintain hygienic condition and fed the animals in the stall without grazing here and there which reduce the chance of ingestion of contaminated feed leading to lower prevalence of *Balantidium sp* infection. Maintenance system is very important factor, directly affect the epidemiology of balantidiasis due to its life cycle and transmission model. The swallowed cysts will develop into tropozoit and binary division will occurs in small intestine. The tropozoit then will turn into cysts and come out with feces (Urquhart, *et al*., 1996; Schuster and Ramirez-Avila, 2008; Koopowitz, *et al*., 2010; Ahmad and Rasad, 2011). A free range cattle has a bigger opportunities to consume contaminated grass. Poor and unconscious farmer do not practice to maintain proper hygienic condition, compared to tie stalls management which tend to practice proper hygiene, disposal of faeces and waste products. This supported by Roy, *et al*. (2011) who reported that the prevalence of *B. coli* infection was higher in buffaloes reared in normal floor/muddy floor (46.21%) than that of animals reared in concrete floor (36.84%).

Sex wise distribution of *Balantidium sp* infection in cattle

Sex of cattle had an effect on the prevalence of *Balantidium sp* infection. This study showed that the prevalence of balantidiasis in cattle was significantly higher ($p<0.05$) in male cattle (23.20%) than female cattle (13.72%) (Table 1). Calculated odds ratio revealed that male cattle were 1.907 times more prone to infected with *Balantidium sp* than that female cattle (Table 2). It is supported by previous research stated that male buffalo (3.8%) had a higher prevalence of balantidiasis than female buffalo (3.4%) (Islam, 2000). Mamun, *et al*. (2011) also reported that the male buffaloes (37.11%) were more susceptible to infected by *Balantidium sp* than female buffaloes (Asif, *et al*., 2007). However, other research indicated that gender did not affect balantidiasis prevalence in cattle (Singh, *et al*., 2012) which supported to the earlier finding of Azhar, et al. (2002) who reported both male and female buffalo, have the same chance of being infected with the *Fasciola sp*.

On the other hand, some research has found an opposite result. Study conducted by Bachal, *et al*. (2002) found the prevalence of *Balantidium sp* infection in the female buffalo (48.30%) was higher than the male buffalo (45.12%). Roy, *et al.* (2011) found similar result and reported a higher prevalence of balantidiasis in female buffalo (47.32%) compared to male buffalo (38.46%).

The sex of the animal is the most common comparison that is widely used as a risk factor of animal diseases. This various findings can not be explained with certainty. In general, disease susceptibility can not only be measured by analyzing a single risk factor. There is so many other factors can interact with sex as a risk factor. The various results between the present and previous findings possibly may be due to the alteration in the physiological condition of cattle during pregnancy, lactation and parturition (hormonal influences) and lower feed supplements for production which may lead to decreasing of resistance (Roy, *et al*., 2011). Higher levels of prolactin and progesterone hormones make the female individual more susceptible to any infection (Lloyd, 1983). Meanwhile, male cattle are also suspectible to stress for a poorly understood reason, which is a predisposes factor of infections (Mamun, *et al*., 2011).

Age wise distribution of *Balantidium sp* infection in cattle

According to age, cattle were categorized into two groups, namely, young cattle (≤ 18 months) and adult cattle (> 18 months) (Batan, 2006). Result of this study showed that 22 young cattle were positive of balantidiasis out of 142 young cattle (15.49 %), while in the group of adult cattle, 33 cattle were positive of balantidiasis out of 178 cattle (18.54%) (Table 1). Meanwhile, the chi-square (χ^2) test revealed that there were no correlation between age of cattle and the prevalence of balantidiasis ($P>0.05$) (Table 2). These results was different with earlier findings of Roy, *et al*. (2011) who reported that age was significantly influence the prevalence of balantidiasis. Another research also reported also mentions that the prevalence of infection Balantidium sp. buffalo is highest in the highest prevalence in buffaloes of 5 years and above age group (4.0%) than in animals 2-5 years (3.48%) and below 2 years of age (3.24%) (Islam, *et al*., 2000). The variation of prevalence of *Balantidium sp* infection in different age groups of cattle is consider as the result of various stress condition such as pregnancy, lactation, parturition, transportation of goods, exhausted immune system and management system are responsible for the higher prevalence of *B. coli* in adult animals (Roy, *et al*., 2011).

Table 2. Chi-square (χ^2) and odds ratio test for identifying the risk factors of *Balantidium sp* infection in cattle in Bali

Risk Factor	Category	χ^2-value	p	Odds ratio = exp(β)	95% C.I for odds ratio
Origin of Cattle	Plateau Lowland	18.464	0.000	4.025	2.065-7.846
Rainfall	Wetland Dryland	11.614	0.001	2.901	1.545-5.447
Maintenance system	Free stalls Tie stalls	43.946	0.000	8.982	4.403-18.323
Sex	Male Female	4.739	0.029	1.907	1.060-3.429
Age	(≤ 18 months) (> 18 months)	0.515	0.473	-	-

Conclusion

Based on the results of this research, it can be conclude that the prevalence of *Balantidium sp* infection in bali cattle (*Bos sondaicus*) in Bali Province was 17.19% in general. Bali cattle are highly suspectible to balantidiasis irrespective of origin of cattle, environment rainfall, maintenance system, and sex. However, the age of cattle has no effect on prevalence of *Balantidium sp* infection in bali cattle (*Bos sondaicus*) in Bali Province.

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