

Overview of coccidiosis in sheep: History of Disease Incidence in the World and Life Cycle

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Abstract

Sheep are regarded as being one type of potential livestock that are widely raised in various regions around the world for both purposes of producing meat and wool. In particular in the Southeast Asian areas, such a stock has been playing a significant role in providing animal protein for human consumption in one hand. However, on the other hand health, genetic and environmental factors are critical for animal husbandry factors to success. One of main diseases in sheep population in the region has been an internal parasitism due to coccidian infestation that had proved to be economically detrimental as the infestations can cause health problems ranging from substantial weight gain losses to culminating in death of the livestock. This article reports on literature reviews regarding various cases of coccidiosis in several countries and also matters related to a life cycle of the disease causative agent. The main highlight of this review has been aimed at a formulation of appropriate and effective disease control strategy in combating coccidiosis in small ruminant population in the country.

Keywords : Sheep, coccidiosis, livestock, animal diseases

INTRODUCTION

Coccidiosis is often manifested as highly contagious enteritis. This condition is actually caused by an acute coccidian invasion, which is mostly in forms of intestinal mucosa destruction by protozoa of the genera *Eimeria* or *Isospora*. Clinical signs may include diarrhea (Keton and Navarre, 2018), fever, in appetite, weight gain loss, emaciation, and even in extreme cases it might be culminating in death. Diarrhea is most commonly encountered in lambs (Khodakaram-Tafti, and Hashemnia, 2017). The existence of a predilection site for coccidian infestation in sheep is however still being studied, particularly for its pathogenesis (Barrel, 1997).

Cases of coccidian infestation had been reported in sheep in 1879 and since then have been reported worldwide. Coccidia are known as protozoan unicellular organisms of the phylum Apicomplexa that parasitize most major lineages of vertebrates as well as invertebrates (Fayer, 1980). Sporozoites in blood (Haemosporidiides), generally not called coccidia, but they are

known as Haemosporidia instead. The later consists of the genus Plasmodium, Haemoproteus and Leucocytozoon.

Coccidiosis in livestock is generally caused by the genus *Eimeria* (Shivaramaiah *et. al.*, 2014; Sufi *et. al.*, 2017), so it is also called eimeriosis. Another name for coccidia that was used previously was Psorospermim which was discovered by Lieberkuhm in 1841 and by Leukart as cocsidium (Gondipon, 1988)

There are approximately 17 species of *Eimeria* in sheep. There are 9 genera of *Eimeria* identified in sheep in New South Wales, Australia, namely *E. ashata*, *E. arloingi*, *E. crandalis*, *E. faurei*, *E. granulosa*, *E. ninakohlyakimovae*, *E. intricata*, *E. parva*, and *E. Pallida*. In the UK, it is reported that the species that mostly attack sheep are *E. crandalis*, *E. faurei*, *E. parva*, *E. weybridgensis* and *E. ovina*. In Africa the species of most commonly encountered are *E. ovina*, *E. weybridgensis*, *E. marsica* and the pathogenic ones are *E. ashata* and *E. ninakohlyakimovae* (Gondipon, 1988).

Eimeria nina-kohlyakimovae was originally the name for coccidia in goats, while sheep were given another name, namely *E. ovinoidalis* (da-Silva, 2014; Paul *et. al.*, 2020). The morphology of *E. ninakohlyakimovae* and *E. ovinoidalis* is almost the same, but there is no cross infection between sheep and goats (Gondipon, 1988).

The name *E. arloingi* is a species of coccidia in goats, but some authors use this name in sheep (Mohamaden *et. al.*, 2018). There is a species very similar to *E. Arloingi* commonly called as *E. Ovina* in sheep (Soekardono, 1989). *Eimeria arloingi* cannot infect sheep; meanwhile *E. ovina* cannot infect goats. In England, two forms of *E. arolingi* were found, namely the large one called *E. weybridgensis* and another of which a small one called *E. ovina* (Gondipon, 1988).

Efforts have been made to improve taxonomy to reduce taxonomic redundancy, by recording, storing and re-listing coccidians. Based on Coccidia of the World compiled by Don Duszynski, Steve Upton, and various universities, there are more than 2000 species named coccidia spread over 8-13 families. This proves that the diversity of coccidia has not been well described. This may also lead to a lacking in our understanding to both pathogenicity of the diseases and difficulties in setting up our disease control measures (Moekti, 2021 unpublished data). Furthermore, not only by utilizing anti-protozoan medication against the diseases has been proved to be ineffective due to the protozoan high ability in progressing their drug resistance (da-Cunha *et. al.*, 2010), but also by combating the diseases through vaccinations, will face huge huddles as any vaccine development will be inappropriate measures as producing protozoan vaccines is seemingly to be cumbersome process due to both their complex life cycle and also their presence of unlimited gene copies encoding for their dominant antigenic determinants, leaving an immunogenic selection for vaccine candidates closed to impossible to approach (Moekti, 2021, personal communication). Therefore, this following literature review is primarily attempted to aiming at finding of most appropriate and effective disease control strategies in order to overcoming a health problem with vast complexity measures of its causative agent pathogenicity.

DISEASE PREVALENCE IN VARIOUS REGIONS AROUND THE WORLD

Yan *et al.* (2021), reported an epidemiological study of *Eimeria* coccidia in Kazakh sheep from a Kazakh × Texel cross in 2019, a total of 7599 sheep feces samples were collected from Zhaosu County and Nilka County in Ili Kazakh Autonomous Prefecture in four seasons which were taken in spring, summer, autumn, and winter. Infectious parasites were identified by the saturated salt floating method, and the intensity of infection was calculated by the modified McMaster method. To evaluate the difference in the number of fecal oocysts (FOC) for coccidia per sample, the evaluation was carried out using SPSS 19.0. The results show that there are *Eimeria* in two districts of Ili locations. Most coccidia infections in these two areas are mild to moderate. The log mean (FOC) of coccidia infections in Zhaosu was significantly lower than in the Nilka. The average log (FOC) of coccidia infection was highest in spring, followed by autumn, and lowest in summer and winter. Gregory *et al.* (1980) received coccidiosis diagnosis report on the cases that had been affecting sheep flocks in England and Wales. The data had been recorded by the Veterinary Investigation Centers over a period of 1978 to 1979. These data suggested that there was an increase in the number of clinical coccidiosis cases in both places.

A research work of El-Alfy *et al.* (2020), which was mainly based on sheep faecal examination by using a standard floatation technique involving 184 samples collected from sheep population in the Dakahlia governorate, Egypt, had shown that the presence of coccidian oocysts from *Eimeria* species in 68.4% of the total samples examined. A coccidian prevalence rate was found to be higher in young sheep than that in adult ones. Eleven species of coccidia were identified i.e. *E. ahsata*, *E. rawensis*, *E. crandalis*, *E. faurei*, *E. granulosa*, *E. intricata*, *E. marsica*, *E. ovinoidalis*, *E. pallida*, *E. parva*, and *E. webybridgensis*.

Meanwhile, a research work from Wang *et al.* (2010) on coccidian infestation in both sheep and goats populations in Heilongjiang Province, North-eastern China conducted over a period of January to June 2009. From 508 samples comprising 309 sheep and 199 goat faeces respectively, where coccidian oocysts were detected by employing a flotation technique in almost all the samples (462 out of 508 or 90.9%). Sporulation conditions and oocyst morphology were used in the identification of coccidian species. The overall prevalence of coccidian infestation was 90.9% (detected in 462 out of overall 508 samples); whereas prevalence rates within species of animals, sheep and goats were 92.9% (287 out of 309) and 87.9% (179 out of 199) respectively. There were 13 *Eimeria* species were identified namely *E. bakuensis*, *E. faurei*, *E. parva*, *E. ahsata*, *E. crandalis*, *E. granulosa*, *E. intricata*, *E. pallida*, *E. chistenseni*, *E. caprina*, *E. alijeivi*, *E. apsheronica* and *E. arlongi*.

A point prevalence of *Eimeria* infestation in sheep and goats in the village of Geneffe, Suez Governorate, Egypt that was monitored over a period of March 2015 to February 2016, collating in a total of 277 animals, which comprised 142 sheep and 135 goats, resulted in 57.7% and 60% respectively. Faecal examinations by a microscopy and followed by a polymerase chain reaction (PCR) indicated that *Eimeria* infestations in sheep in area were significantly higher in summer (75%) than those in autumn (74.2%); whereas a prevalence rate in winter appeared to be the lowest (38.2%) amongst other seasonal cases. *Eimeria* species that were mostly identified in sheep in the area were *E. crandalis*, *E. granulosa*, *E. ovina*, *E. parva*, *E. faurei*, *E. ovinoidalis*, *E. intricate*, *E. Pallida*, *E. arloingi*, and *E. ahasta* (Mohamaden *et al.*, 2018).

Early in 1965 Marth and Sherick conducted their study on coccidiosis caused by *E. Ashata* infestation amongst nursing lambs in Illinois, the United States of America, where they then reported a similar pattern coccidiosis cases found in 200 heads of lambs. As shown in their

findings that a disease prevalence rate of higher than 30% with a mortality rate ranging from four to six per cent. More similar patterns of coccidian infestation were also reported after the disease survey on sheep and goats carried out in Senegal (Gondipon, 1988).

As previously stated that most of internal parasitism, in particular in small ruminants can cause in enormous economic losses to livestock industry in some countries (Kumar *et al.*, 2013). An internal parasitism attack rate, which was indicated by the presence of faecal oocysts in sheep population in certain countries, can be as high as 85% (Gondipon, 1988). Moreover, Leon *et al.*, (2019) reported that a prevalence rate of coccidiosis in sheep appeared to be significantly higher ($P>0.05$) than that found in cattle, as there had been proved by them that in the Colombian North-eastern Mountain, the prevalence of parasitic infestation in sheep (63%) was higher than that found in cattle (50.5%).

A disease survey of ruminants in Malaysia found that 15.7% of enteritis cases were caused by coccidia (Zakaria *et al.*, 2019). The protozoan coccidiosis was then considered as being an important enteric disease of sheep and goats in the region. This was mainly because of such a condition could lead to various clinical manifestations such as diarrhea, inefficient weight gain, and even ultimately culminating in death. Effective in prevention and disease control measures for coccidiosis had proved to resulting in much greater achievement of productive performances. Adversely, if it were left untreated, such a disease would have caused in inefficient production and substantial economic losses. Paul *et al.* (2020) conducted a cross-sectional survey over a period between March and December 2019 to investigate disease prevalence, risk factors, and infestation rates of gastrointestinal parasitism due to strongyles and coccidia in selected small goat herds in Negeri Sembilan, Malaysia. A total of 257 blood and faecal samples together with farm management records were collected from four farms within Negeri Sembilan regions. Gastrointestinal parasites were then examined by using a routine sodium chloride flotation method, and were then followed by using a McMaster technique in order to enumerate faecal worm ova (eggs) and/or coccidian oocysts per gram output (EPG/OPG). The severity of infestation was classified as mild (50-799), moderate (800-1200), or severe (>1200). Packed cell volume (PCV) was also determined by a microhematocrit centrifugation to enable the investigators to classify any anaemic or non-anaemic conditions. Coprological examinations revealed an overall prevalence of 78.6% (CI = 72.74–83.44) and a herd rate prevalence of 100% of strongyle and coccidia infestation in goats from Negeri Sembilan with higher infection rate in A-Lenggeng flocks (95.6%) than B-Senawang (87.3%), D-Mendom (80.6%), or C-Seremban (60.0%). *Eimeria* coinfection (4.3%; CI = 2.41 to 7.50). Quantitative analysis has revealed different EPG/OPG patterns ($P<0.05$) in different categories of goats. In total 40.1% mild, 6.6% moderate, and 19.8% severe coccidia infestations among goats. There was an increased chance of infection with strongyle and coccidia among female (OR = 3.2) and adult (OR = 11.0) goats from a small farmer herd in Negeri Sembilan. In conclusion, gastrointestinal parasitism due to both strongyles and coccidia tends to occur with high frequency among small goats, and there is a higher risk of infection among adult and female cattle.

CLASSIFICATION OF COCCIDIA AND SPOROCYSTS

Coccidia is a term that has not been defined precisely, but generally, coccidia is protozoa that belong to the class Sporozoa. Sporozoites in the blood (Hemosporidii) are generally

not called coccidia but are known as haemosporidia, which consist of the genera that are *Plasmodium*, *Haemoproteus*, and *Leucocytozoon*.

A simple way of identification that is commonly used is the examination of oocysts, especially, coccidian ones that are morphologically round, slightly round, oval, or elliptical and the size depends on the species (Cama and Mathison, 2015). The oocyst of *E. intricata* is the largest oocyst among all coccidia species in sheep with a size of 47 x 32 µm, *Eimeria* spp. were found in 74.11% (581/784) of the samples in Erzurum Province, Turkey. Identified *Eimeria* species were as follows: *E. parva* (59.68%), *E. ovina* (51.67%), *E. faurei* (47.80%), *E. ahsata* (39.27%), *E. granulosa* (36.62%), *E. punctata* (28.42%), *E. pallida* (26.09%), *E. ovinoidalis* (18.34%), *E. crandallis* (16.79%), *E. intricata* (15.76%), *E. weybridgensis* (11.36%) and *E. marsica* (6.20%) (Akyuz *et. al.*, 2019). Oocysts of the genus *Eimeria* can be distinguished from other genera of Sporozoa by the number of sporocysts present in sporulated oocysts. Coccidial oocysts are tough structures, which contribute to resistance to mechanical damage, often isolated from the feces or urine of their hosts. The presence of these oocysts and allows the parasite to survive and remain infective for a long time. The diagnosis of coccidiosis, species description, and systematic all depend on the characterization of the oocyst (Berto *et. al.*, 2014). In the genus *Eimeria*, there are four sporocysts and each sporocyst contains two sporozoites (Helke *et. al.*, 2006).

LIFE CYCLE

Coccidia of the genus *Eimeria* are host specific and each species has its own predilection for the digestive tract. Each species infects a specific cell type of the gut. In most coccidia in sheep, the giant schizont stage (about 300 µm in diameter) appears as white patches on the intestinal mucosa (Ammar *et. al.*, 2019). The infective form is an oocyst that has sporulated and the way of infection is through the digestive tract.

The mechanism of infection is food containing oocysts, after entering the intestinal lumen, sporozoites will come out of the oocysts and enter the epithelial cells of the digestive tract. The process of releasing sporozoites from oocysts requires CO₂ and the enzyme trypsin and bile. The process of releasing sporozoites from oocysts can be observed in the laboratory by applying 5% alkaline trypsin to a warm glass object. In sheep coccidia, it is known that the concentration of CO₂ and the contact time required for excystation varies according to the coccidia species (Gondipon, 1988).

Bile fluid will facilitate the entry of the trypsin enzyme into the oocyst through the microfil and digest the "sporocystic plug" so that the motile sporozoites will exit the cell. These sporozoites also secrete a type of enzyme to destroy the "sporocystic plug". The free sporozoites measuring 10 x 1.5 µm, are transparent and can perform contraction, elongation and gliding movements. Sporozoites that enter the cell enlarge and undergo a "round up" process, where this stage is the beginning of the asexual reproduction process or the process of schizogony. Sporozoites that develop in cells and go through a "round up" process become trophozoites which undergo nuclear division and become schizonts which are the first generation of schizonts. The division of the nucleus in the process of asexual reproduction is thought to be mitotic. Schizonts develop by repeated divisions and eventually become elongated merozoites (Mehlhorn and Heydom, 1976). This stage is referred to as the first stage merozoites. These motile merozoites will leave the host cell and penetrate into other cells, then develop into second generation schizonts (Dubey, 2020). The number of merozoites produced is in accordance with the number

of nuclei in the mature schizont. This schizont formation process will continue for several generations (Minchin, 2003).

At the end of the schizogony process that has been repeated, merozoites enter the cell and become gametes consisting of micro- and macro-gametes, which in this phase is called the gametogony phase or sexual reproduction phase (Figure 1). The macrogamete is the female form and the microgamete is the male form. Macrogametes develop into macrogametes and enlarge in the nucleus and spread around the cytoplasm, and large granules are found in the periphery of the cell. Meanwhile, microgametes develop into microgametes by repeated division of the nucleus. The nucleus will spread in the plasma but eventually form like a coma and gather at the periphery of the cell. The number of macrogametes is much more than that of microgametes, but at the end of the development of both the number of microgametes exceeds the number of macrogametes (Walker *et al.*, 2013). Microgametes are flat-shaped with a length of 5 μm , will come out of the cell and move with the help of flagella to reach cells containing macrogametes for fertilization to occur. The result of fertilization is a zygote which will form the cyst wall and eventually become an oocyst (Chapman *et al.*, 2013).

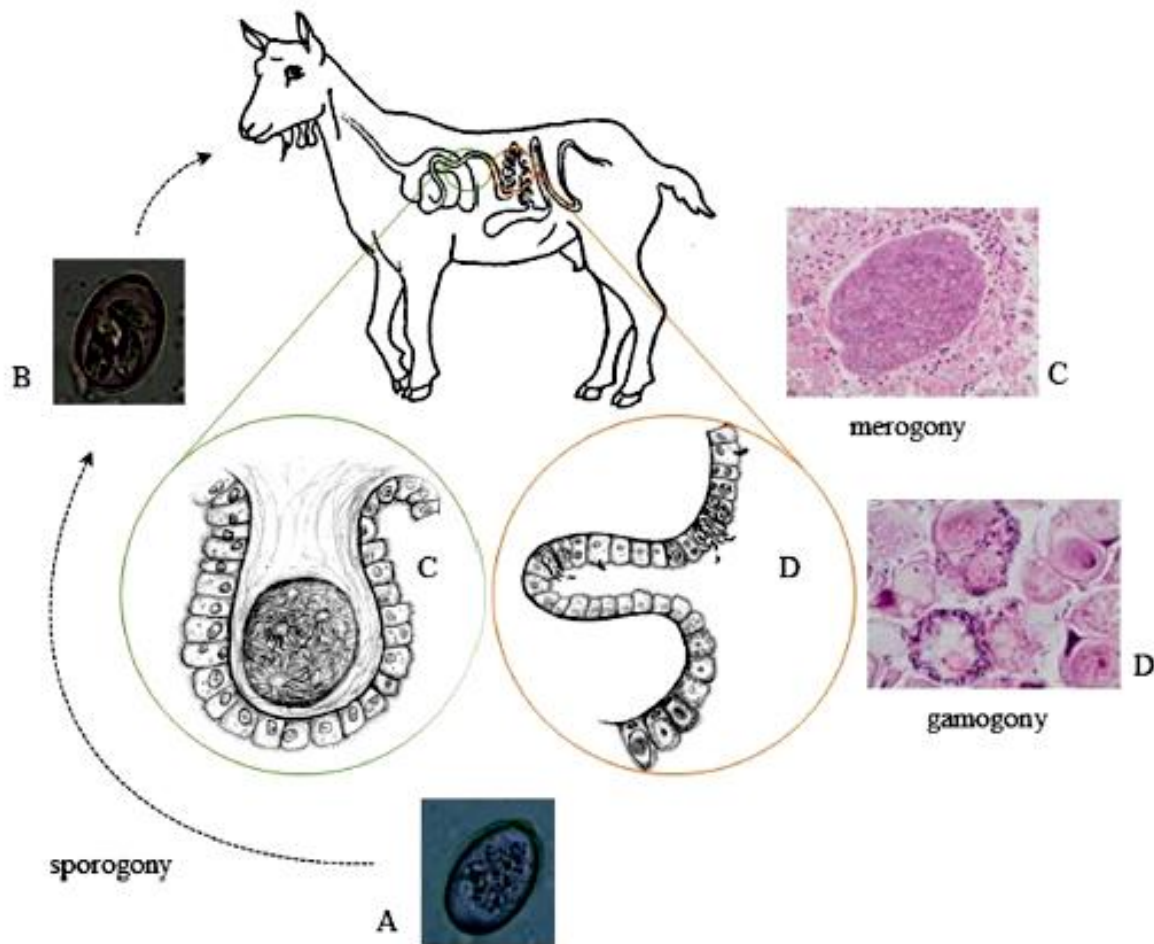


Figure 1. Eimeria Life Cycle (adopted from da-Silva, 2014) A. unsporulated oocyst; B. Sporulated oocyst; C. Macromeronts in intestine; D. Gamogony.

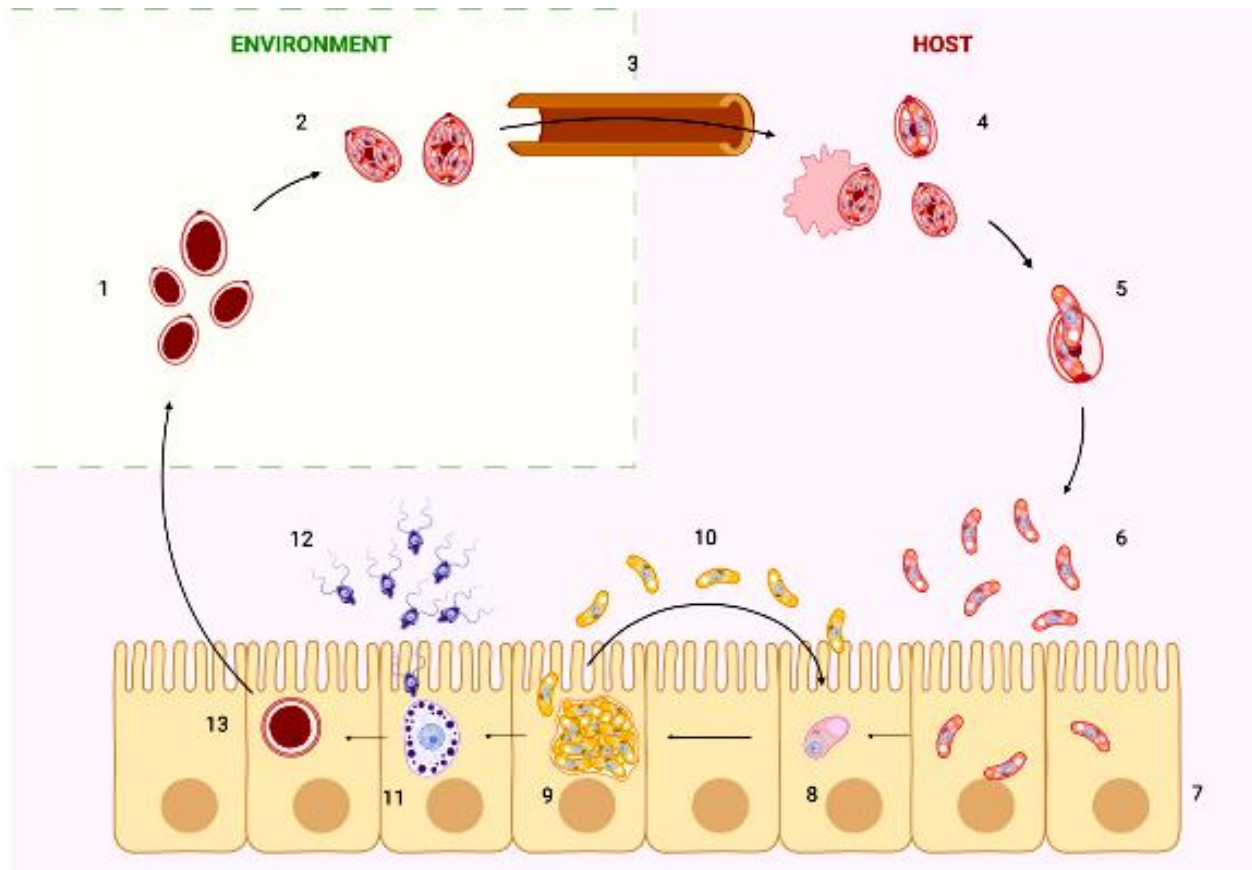


Figure 2. *Eimeria coccidian* life cycle. 1. Unsporulated oocysts; 2. Sporulated oocysts; 3. Gastrointestinal tract; 4. Mechanical rupture of oocyst and sporocyst release; 5. Excystation of sporozoites; 6. Released sporozoites; 7. Enterocytes; 8. Trophozoite; 9. Mature schizont; 10. Merozoites and re-infection of enterocytes; 11. Macrogamete; 12. Microgamete; 13. Zygote (Felici *et. al.*, 2021).

This sporogony process is influenced by several environmental factors (Figure 2), including humidity, temperature and oxygen (Waldenstedt *et. al.*, 2001). The optimum temperature for sporulation to occur is 30°C and at -12°C sporulated oocysts can only live for two weeks, while unsporulated oocysts will die within 96 hours (Gondipon, 1988). There are also researchers who say that the optimum temperature for sporulation is 20-25°C. The time required to sporulate to the infective stage is a special trait of each coccidia species, so this trait is used in identification. Under conditions of oxygen and appropriate humidity and optimum temperature, oocysts can sporulate (Felici *et. al.*, 2021) and become infective within 24-28 hours (Gondipon, 1988).

In general, oocysts are very resistant to changes in environmental factors, but in countries with four seasons, in the dry season oocysts are likely to die due to too high temperatures and low humidity (Lopez-Orosio *et. al.*, 2020). Drying of oocysts at 40°C will inhibit sporulation and eventually the oocysts will die. In the life cycle of coccidia, oocysts are a form of resistant and infective stages so that this stage is very important in the spread of coccidiosis disease.

CONCLUSION

Recognition of coccidiosis as a disease and its spread and life cycle is very crucial to be thoroughly understood in order to set appropriate effective preventive measures for controlling the disease. In other words that if the disease pathogenicity and its related aspects are fully understood, then appropriate action and prevention, treatment, control and even eradication of the disease can be carried out effectively.

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