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Coccidiosis: A Parasitic Disease of Significant Importance in Rabbits

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ABSTRACT

Rabbits are considered an important and healthy source of animal protein all over the world. They are susceptible to important diseases that can reduce their productivity, causing severe economic losses. Coccidiosis is one of the important protozoon diseases caused by *Eimeria* species. Rabbits are highly susceptible to coccidiosis, especially after weaning time. Coccidiosis in rabbits has two forms, namely hepatic and intestinal. Affected animals indicated the symptoms of diarrhea, reduced appetite, dehydration, and weight loss as well as liver and intestinal lesions. Diagnosis is based on the detection of the infective stages of the protozoon in feces or affected tissues. Prevention and control are achieved by adopting hygienic measures and using different anticoccidial drugs. The use of natural alternatives for the prophylaxis of coccidiosis in rabbits indicated promising results. Vaccine production trials are still under investigation. Accordingly, this review article aims to shed light on coccidiosis in rabbits considering pathology, diagnosis, and control.

Keywords: *Eimeria*, Intestine, Liver, Rabbits, Treatment

INTRODUCTION

Rabbits (*Oryctolagus cuniculus*) are regarded as a potential source of animal protein for human consumption. The meat of rabbits is recommended for human consumption more than other sources of proteins due to its high nutritious protein, calcium, phosphorus, and linoleic acid, with low fat and cholesterol contents (Nistor et al., 2013). In addition to the commercial use of rabbits, they can be used for wool production and in medical research as laboratory animals, and they are raised as pets for hobby purposes (Al-Mathal, 2008).

Rabbits are susceptible to dangerous viral, bacterial, and parasitic diseases that drastically affect their production. Coccidiosis or eimeriosis is one of the most important and widely distributed parasitic diseases in rabbits (Grès et al., 2003; El-Shahawi et al., 2012; Okumu et al., 2014; Yin et al., 2016; Bachene et al., 2019; Hamid et al., 2019). The disease affects the intestine, liver, and bile duct of the animal and is associated with digestive disorders (Manjunatha et al., 2019). Coccidiosis is a highly contagious disease that has negative impacts on the domestic rabbit industry worldwide (Pakandl, 2009; Geru et al., 2017). It seriously impairs growth performance parameters, causes high morbidity and mortality rates (Abdel-Megeed et al., 2005), and reduces the carcass weight by more than 23% (Barriga and Arnoni, 1981). Coccidiosis causes annual losses in the rabbit industry in the USA amounting to 127 million dollars, and similar losses may happen worldwide (Chapman, 2009). The prevalence of coccidiosis in rabbits is varied and influenced by geographical location, season, as well as management factors like housing and rearing conditions, and the use of preventive coccidiostats (Chowdhury and Fraser, 2008). Furthermore, the prevalence of coccidiosis in weaned rabbits is higher than that in growing and adult ones (El-Ghoneimy and El-Shahawy, 2017). Duszynski and Couch (2013) stated that the rate of coccidial infection ranged from 64% to 100% all over the world.

The disease is caused by an intracellular ubiquitous protozoon parasite of the genus *Eimeria* (Jing et al., 2012; Mäkitaipale et al., 2017). Mixed infection with more than one *Eimeria* spp. is common (Jithendran and Bhat, 1996). There are 15 *Eimeria* spp. that affect the intestinal tract and one species (*E. stidea)* affecting the liver and bile duct (Li and Ooi, 2009). *E. intestinalis*, *E. magna*, *E. piriformis*, *E. perforans*, *E. media*, *E. agnotsa*, *E. exigua*, *E. flavescens*, *E. irresidua*, *E. coecicola*, *E. vejdovskyi*, *E. roobroucki*, *E. oryctolagi*, *E. nagpurensis*, and *E. matsubayashi* invade the small intestine (Soulsby, 1968). The highly pathogenic *Eimeria* spp. in rabbits are *E. intestinalis* and *E. flavescens*, the moderately pathogenic spp. are *E. magna*, *E. irresidua*, and *E. piriformis*, and the low pathogenic or nonpathogenic spp. are *E. exigua*, *E. media*, *E. coecicola*, and *E. perforans* (Jithendran, 1995). The rapid spread of infection, due to the direct and short life cycle of the parasite, is characteristic of coccidiosis (García-Rubio et al., 2017). The severity of coccidial infection depends on the number of ingested oocysts, age, and the immune status of the animal (Miller and Fowler, 2003). In the early stage of infection with coccidiosis, sudden diarrhea, or death may occur without any clinical signs.

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Clinical infection is characterized by apathy, diarrhea, dehydration, reduced appetite, and weight loss resulting in death (Ogolla et al., 2018).

Prevention and control of coccidiosis in rabbits are achieved through careful management, enhancing rabbits' immunity, vaccination, and using synthetic anticoccidial drugs. Housing in large, dry, sunny, and disinfected rooms is very essential to fight coccidial infection (Pilarczyk et al., 2020). Although synthetic anticoccidials were effective, their hazardous use in the field resulted in some drawbacks. Therefore, there is a shift toward using novel approaches that pose a minimal risk to human or animal health. Different kinds of herbal extracts (Indrasanti et al., 2017; Sorour et al., 2018; Rivero-Perez et al., 2019) and acidifiers (Shkromada et al., 2019) showed successful preliminary results in treating coccidiosis in rabbits. Efficient vaccines are very important for the prevention of rabbit coccidiosis (Song et al., 2017), but so far, no vaccine is available for rabbits.

Given the above information, the aim of the present review is to shed light on coccidiosis in rabbits considering pathology, diagnosis, and control.

Parasite life cycle

Infection usually occurs through the ingestion of contaminated feed and water containing sporulated oocysts (i.e., infective oocysts consist of four sporocysts, each containing two sporozoites). Upon entering the gut, due to the effects of gastric and pancreatic juices, the oocysts walls rupture and the sporozoites invade the intestinal epithelial cells and then pass via the mesenteric lymph nodes and hepatic portal circulation to the liver where they enter the epithelial cells of the bile duct becoming trophozoites and then schizonts. The recognition and invasion processes may occur via the action of sugar residues in *Eimeria* sporozoites (John et al., 1999), followed by the release of merozoites (merogony stage) that form four generations in the asexual stage of the parasite. Lastly, male (micro) and female (macro) gametocytes combine sexually (gametogony stage) to form zygotes that develop into non-sporulated oocysts. The non-sporulated oocysts pass in the bile and are shed in the feces of the infected rabbits about 18 days after infection. Under favorable environmental conditions, sporulated oocysts are formed within three days (Gardiner et al., 1998).

Susceptibility

All breeds of domestic are highly susceptible to coccidiosis, and those of 1-4 months old more susceptible than adults (González-Redondo et al., 2008; Papeschi et al., 2013; Bachene et al., 2018). Suckling rabbits cannot be infected with coccidiosis before three weeks of age. It has been found that the production of *E. flavescens* and *E. intestinalis* oocysts in suckling rabbits increases with age (Pakandl and Hlásková, 2007). More susceptibility in young rabbits may be due to reduced immunity resulting from weaning stress, feeding and reproductive status (Drouet-viard et al., 1997a; Al-Mathal, 2008). Also, it was found that female rabbits were more infected than males (Faraj, 2017).

Types of rabbit coccidiosis

Hepatic coccidiosis

Hepatic coccidiosis involves the liver and bile ducts of rabbits causing cirrhosis and cholestasis (Singla et al., 2000). Hepatic coccidiosis is associated with severe economic losses in rabbitries and is caused by *E. stiedae* that is one of the most pathogenic species of *Eimeria* (Xin et al., 2016; Al-Taee and Al-Zubaidi, 2017). The oocysts of this species of *Eimeria* were first detected in the bile of infected rabbits in 1674 (Duszynski and Couch, 2013). The most adverse effects of hepatic coccidiosis are reduced growth and feed utilization and increased mortality rate in young rabbits (Hanptman et al., 2001). Like other *Eimeria* spp., *E. stiedae* penetrates the intestinal wall and migrates to the bile ducts where it reproduces (Kraus et al., 1984). Hepatic coccidiosis is mostly chronic and subclinical infection specifically in adults who are carriers and sources of infection (Barriga and Arnoni, 1981; Al-Mathal, 2008; Pakandl, 2009). Some of the affected animals displayed symptoms of anorexia, polydipsia, brown watery diarrhea, dehydration, icteric membranes, poor feed conversion, growth retardation, wasting of the back and hindquarters, coarse hair, abdominal distension, and even death especially in young rabbits with severe infection (Erdogmus and Eroksuz, 2006; Lakshmanan et al., 2011; Al-Saeed et al., 2017). High mortality results from high doses of oocysts, while morbidity results from diarrhea and reduction in body weight (Renaux et al., 2003). Several studies have described the post-mortem findings of hepatic coccidiosis. On gross examination, enlarged and cirrhosed liver with multiple distributed whitish nodules containing creamy thick exudate, and distended gall bladder has been reported. Moreover, histologic findings included cholangitis, bile duct hyperplasia, hemorrhage, dilatation and congestion of the central veins as well as necrosis of hepatocytes and hepatic fibrosis (Sanyal and Sharma, 1990; Cam et al., 2008; AL-Naimi et al., 2012; Sorour et al., 2018).

Intestinal coccidiosis

There are about 15 species of *Eimeria* that are known to induce pathology in the intestine of rabbits causing intestinal coccidiosis. The most common *Eimeria* spp. that cause intestinal coccidiosis in rabbits are *E. magna*, *E.*

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irresidua, *E. media*, and *E. perforans* (Fox, 1984). The parasite colonizes distinct parts of the intestine and the mucosa at different depths (Pakandl, 2009). Intestinal coccidiosis often is observed in rabbits aged from six weeks to five months old. Older animals can acquire immunity after recovery and become carriers (Kulisic et al., 2006). This type of coccidial infection results in atrophy of the intestinal villi, malabsorption of nutrients, hypoproteinemia, electrolyte imbalance, dehydration, anemia (Dakshinkar and Dharmadhikari, 1985; Hana et al., 2011) and is manifested by diarrhea, weight loss, and mortality (Lebas et al., 1986). Affected rabbits suffer from weakness, gnashing teeth, dirty anus, weight loss, soft to watery hemorrhagic diarrhea, dehydration, and thirst (Fioramonti et al., 1982). Inflammation and edema in the ileum and jejunum associated with mucosal bleeding and ulcerations were observed (Coudert et al., 1995; Oncel et al., 2011). There are differences in pathogenicity among intestinal coccidial spp. (Jithendran, 1995). These differences in pathogenicity can be determined through the experimental infection of animals with different *Eimeria* species.

Laboratory diagnosis

Laboratory diagnosis of hepatic and intestinal coccidiosis depends on the analysis of feces of suspected rabbits. Microscopic identification of *Eimeria* spp. oocysts through the fecal analysis of suspected animals is very important (Pakandl et al., 2008). Developmental stages of *E. stiedae* have been detected in stained impression smears from the liver (Al-Rukibat et al., 2001; Sivajothi et al., 2016). Histopathological examination of the liver tissues, bile duct, or intestine is also used for the detection of different developmental stages of the parasite (Sivajothi et al., 2016). The oocyte detection site under a microscope is a guide to determining *Eimeria* species. Immuno-diagnosis of *E. stiedae* was investigated in previous studies (Zayed and Kutkat, 1998; Kandil et al., 2000). Serological diagnosis of *E. stiedae* using ELISA was reported (Abu-El-Ezz et al., 2010; Wei et al., 2020). Identification of *Eimeria* spp. using molecular assays such as multiplex PCR assay was reported (Oliveira et al., 2011; Yan et al., 2013). Hassan et al. (2015) detected *E. stiedae* schizonts using PCR 12 days after the experimental infection of rabbits, and this occurred prior to the development of lesions or shedding of the oocysts in feces.

It should be noted that the presence of oocysts in fecal samples does not confirm the presence of clinical disease. It has been reported that rabbits with high-intensity infection showed no clinical symptoms of coccidiosis (Pilarczyk et al., 2020). The induction of symptoms may depend on the virulence and pathogenicity of the infecting *Eimeria* spp. (Pakandl, 2009).

Prevention and treatment

Management practices

Coccidiosis in rabbits is aggravated by poor hygienic conditions and high stocking densities that encourage the spread of protozoa (González-Redondo et al., 2008). Rabbits raised in groups are more affected than those kept alone (Sharma et al., 2016). Accordingly, the first steps for preventing the occurrence and spread of coccidiosis in a rabbitry are proper hygiene and husbandry practices as well as strict biosecurity measures (Pakandl et al., 2008; Schlolaut et al., 2013). Control of coccidial infection using common disinfectants is difficult as oocysts have a remarkable ability to survive under exogenous environmental conditions (Chapman et al., 2013).

Anticoccidial drugs

Prevention of coccidiosis in rabbits using coccidiostats is regulated by Regulation (EC) No 1831/2003 of the European Parliament and of the Council on additives for use in animal nutrition. Globally, synthetic anticoccidial drugs, either ionophores or synthetic chemicals, remain the mainstream pharmaceuticals that are used for the control of rabbit coccidiosis (Pakandl, 2009). Anticoccidial drugs should be broad-spectrum, highly effective with a good therapeutic index, and easily administered for short time. Coccidiostats in rabbits prevent the developmental stages (schizogony and gamogony stages) of the parasite inside the host. Coccidiostats are usually added to the feed of animals; however, coccidiocidal drugs are added to water. It is preferable to prevent coccidial infection before its occurrence as the treatment is usually not very successful when clinical signs of coccidiosis appear (Pakandl, 2009). A previous study by Peeters et al. (1981) showed that concordant infection with hepatic and intestinal *Eimeria* spp. could be treated with narasin, while hepatic coccidiosis could be prevented by clopidol/methylbenzoquate, robenidine, and salinomycin (Peeters et al., 1982). Several reports recommended the use of toltrazuril for either prevention or treatment of coccidiosis in rabbits (Mikhail et al., 1981; Redrobe et al., 2010; Qamar et al., 2013; El-Ghoneimy and El-Shahawy, 2017). In a study by Vereecken et al. (2012), treatment with diclazuril, salinomycin, and robenidine showed significant improvement in both growth performance and parasitological parameters in infected rabbits compared to non-treated animals. However, using 25 ppm toltrazuril/liter of drinking water for two days was successful in treating the clinical hepatic coccidiosis (Singla et al., 2000), increasing body weight and lowering the mortality rate in rabbits infected with mixed intestinal and hepatic *Eimeria* spp. compared to control animals (Balicka-Ramisz et al., 2014). Treatment with toltrazuril immediately reduced signs and oocysts shedding, allowing the development of immunity against reinfection (Peeters and

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Greeroms, 1986). Treatment using diclazuril is recommended for eliminating oocyst shedding in rabbits worldwide (Vanparijs et al., 1989a, b; Peeters and Geeroms, 1989; Polozowski, 1993; Vereecken et al., 2012). Diclazuril and sulfachlorpyridazine were efficacious in treating rabbit coccidiosis (Ogolla et al., 2018). Studies presented variable results following the prophylactic and therapeutic use of sulphonamides in treating coccidiosis (Joyner et al., 1983; Polozowski, 1993; Redrobe et al., 2010; Qamar et al., 2013). Kolabskii et al. (1973) and Ogolla et al. (2018) reported the effectiveness of sulphachloropyrazine in controlling clinical coccidiosis in rabbits. Successful control of hepatic coccidiosis using sulphaquinoxaline was also reported (Magray et al., 2010). Amprosol, bifuran, and sulpha-based drugs have been used for the prevention of rabbit coccidiosis (Bhat et al., 2010). Trimethoprim-sulfamethoxazole showed moderate to satisfactory efficacious results in the treatment of field infection (Ogolla et al., 2018). It has been documented that sulfonamides protected against experimental *E. magna* and *E. media* infections better than colistin and trimethoprim (Bachene et al., 2019). Amprolium could not treat intestinal and hepatic coccidiosis (Laha et al., 1999; Ogolla et al., 2018). On the other hand, superior effects have been demonstrated following the prophylactic use of amprolium in intestinal coccidiosis especially when applied concurrently with other anticoccidials (Qamar et al., 2013; Laha et al., 2015; El-Ghoneimy and El-Shahawy, 2017). Some anticoccidials such as sulphonamides, salinomycin, and robenidine were toxic for pregnant does and kids, while few others used for poultry were recommended for rabbits (Ogolla et al., 2018).

Alternatives to anticoccidial drugs

Anticoccidial drugs and are relatively inexpensive and showed successful results. However, increase in consumer demand for the production of organic products, the potential development of resistant strains of parasites toward drugs (Pakandl, 2009), and the presence of antibiotic residues in meat created a potential need for searching for natural and safe alternatives to anticoccidial chemicals. Hence, several studies investigated the effects of natural alternatives such as sulfur and sulfates (including copper sulfate), tannic acid, bismuth compounds, thymol, camphor, alum, volatile oils, and garlic, oregano, sage, caraway, cinnamon, basil, and rosemary on rabbit coccidiosis (Kowalska et al., 2012). Oral prophylactic administration of garlic was effective in the amelioration of *E. stiedae* infection (Toulah and Al-Raw, 2007; Abu-Akkada et al., 2010; Indrasanti et al., 2017). Adding of a mixture of garlic oil and oregano to rabbit feed can also help in disease prevention (Kowalska et al., 2012; Nosal et al., 2014). Other plant extracts such as banana stem extract could decrease the number of *E. stiedae* oocysts *in vitro* (Indrasanti et al., 2015). However, Matekaire et al. (2005) stated that both banana root (*Musa paradisiaca*) and sulphadimidine sodium treatments caused a significant decrease in oocyst excretion in rabbits. Neem extract improved body weight and liver function in experimentally infected rabbits with *E. stiedae*, and these results were similar to those in the toltrazuril treated group (Ahmed et al., 2014). Cervantes-Valencia et al. (2015) demonstrated that the hydroalcoholic extract of *Curcuma longa* at doses of 25 and 40 mg/kg body weight reduced the fecal *Eimeria* sp. excretion in naturally infected rabbits. Artemisinin liquid extract, cinnamon, and clove essential oils could also be used for protection against *E. stiedae* infection in rabbits compared to toltrazuril (Sorour et al., 2018). Furthermore, it has been demonstrated that 25 and 50 mg *Salix babylonica* hydroalcoholic extract per kg of body weight successfully reduced the intestinal *Eimeria* oocyst count per gram of feces in rabbits (Rivero-Perez et al., 2019). The effects of herbal antioxidants (*Psidium guajava*) as inhibitors of *E. intestinalis*, *E. magna*, *E. flavescens*, and *E. stiedae* sporozoites were evaluated *in vitro*, and preliminary results showed that these herbs were the best substitutes to chemical anticoccidials (Cedric et al., 2017). A dose of 20 g *Calotropis procera* dried leaves powder per kg pelleted ration of rabbits reduced the number of oocysts in feces with the absence of alterations in the intestine and liver tissues of rabbits with mixed infections (Seddek et al., 2015). Aloe vera and liquid paraffin showed varied efficacy in the treatment of rabbit coccidiosis (Ogolla et al., 2017). Shkromada et al. (2019) proved that water treatment of rabbits with an acidifier concentrate (a mixture of benzoic, acetic, and propionic acids) reduced *Eimeria* invasion, increased body weight, and improved feed conversion. In addition, compared to anticoccidial drugs, this composition was not toxic, improved digestion processes, and inhibited the development of conventionally pathogenic microflora.

Vaccines

Vaccinations using oral or spray dispersion of precocious live lines of *E. magna* oocysts in the nest boxes (Drouet-Viard et al., 1997a, b; Licois, 2004), and vaccination against *E. magna* and *E. media* (Akpo et al., 2012) showed satisfactory preliminary results. Some efforts have been directed toward the production of recombinant vaccines against coccidiosis in rabbits (Hanada et al., 2003; Abdel-Megeed et al., 2005; Song et al., 2010). A recent trial for the preparation of a vaccine against hepatic coccidiosis was carried out. Using immuno-proteomic analysis of sporozoite proteins of *E. stiedae*, the immuno-reactive proteins were recognized by the sera of infected rabbits, which may be helpful for the production of a vaccine (Song et al., 2017). However, production of a vaccine on a commercial and field scale is still distant and limited, may be due to the high cost of production and the time required for the processes of

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optimization, registration, safety assessment, and distribution to customers (Song et al., 2017). Vaccination is promising and still requires extensive research and development to be applied in the near future.

CONCLUSION

As coccidiosis is considered a very important parasitic disease in rabbits, future studies should focus on finding novel approaches for the prevention and control of such a significant threat.

DECLARATIONS

Competing interests

The author has no conflict of interest.

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