Diagnosis, Hematologic Profile, and Treatment of Cystoisosporiasis in Domestic Dog

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Abstract

Cystoisosporiasis infection is caused by *Cystoisospora* spp. or *Isospora* spp. in dogs. *Cystoisospora* spp. are protozoa ordo Eucoccidiorida. Cystoisosporiasis is considered more risky in puppies. Clinical manifestations of cystoisosporiasis tend to be watery diarrhea to melena in puppies which can also be concurrent with vomiting, and tenesmus, whereas in adult dogs known as symptomatic. A female domestic dog, aged >1 yo was kept indoors and had not been vaccinated or anthelmintic drugs. The dog has changed in the consistency of slightly soft feces with a frequency of defecation 1–2 times a day. Diagnose performed using microscopic fecal examination with the floating method found the presence of *Cystoisospora* spp. oocysts, followed by the calculation of oocysts per gram (OPG) using a two-chamber McMaster to determine the degree of *Cystoisospora* spp. infection. Blood examination showed leukocytosis, granulocytosis, and hypochromic anemia. The treatment was administered of 15 mg/kg BW sulfadiazine orally every 12 hours for 7 days, 14 mg/kg BW metronidazole every 12 hours for 7 days, and 300 mg Hemafort® every 24 hours for 7 days. After 7 days of treatment, we reported normal fecal consistency and negative parasitic manifestation. Prevention was done by paying attention to treatment, immediately identifying the presence of oocysts, educating owners to avoid animal distress, and maintaining a good hygiene environment.

Keywords: cystoisosporiasis, *Cystoisospora* spp., protozoa

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INTRODUCTION

Cystoisosporiasis infection is an infection caused by *Cystoisospora* spp. or *Isospora* spp. in dogs (Tilley *et al*., 2021). Cystoisosporiasis is considered the most frequent disease in puppies, which are more likely to have clinical manifestations, whereas adult dogs are usually asymptomatic. Clinical symptoms of protozoan infection include watery diarrhea to melena which may also be accompanied by vomiting, and tenesmus. Death due to excessive dehydration may also occur (Garanayak *et al*., 2017).

The prevalence in Malaysia of parasitic infections in the gastrointestinal tract of dogs is 88.3% and 34.3% of the parasites that cause parasitic infections are protozoa *Entamoeba* spp., *Cryptosporidium* sp., *Giardia* sp. and *Isospora* sp. (Ngui *et al*., 2014). According to study by Glantiga *et al*. (2016), protozoan infection in the gastrointestinal tract of dogs in Indonesia is 10%. Based on the maintenance system, the prevalence of penned dogs was 9.6%, while those released dogs had a prevalence of 10.41%. Infected and released dogs will contaminate the environment with oocysts, which will lead to higher transmission to other dogs (Unal and Gokpinar, 2020). Dogs kept in kennels can also be infected due to a lack of hygiene in the kennel environment (Ilić *et al*., 2021).

Cystoisosporiasis harms animal health, especially in puppies at approximately 4 months of age because the immune system in puppies is still vulnerable and there is no immunity to Coccidia from the mother, so proper diagnosis, treatment, and prevention are needed (Unal and Gokpinar, 2020). Diagnosis of cystoisosporiasis is generally performed by performing microscopic examination of fecal samples qualitatively and quantitatively. Common qualitative fecal examination techniques are the native method and the floating method. Quantitative examination methods indicate the number of oocysts in the host present in each gram of feces using McMaster (Demelash *et al*., 2016; Purnama *et al*., 2021).

Treatment of cystoisosporiasis can be given sulfonamide antibiotics for 10–14 days or until the dog is asymptomatic and fecal examination is negative for oocysts. According to Garanayak *et al*. (2017), dogs treated with sulfonamide antibiotics combined with metronidazole for 5 days can show complete clinical recovery along with the elimination of oocysts in feces. Prevention can be achieved by paying attention to treatment that eliminates oocysts in feces to disrupt the life cycle (Gomez *et al*., 2018). It is important to identify the presence of coccidian oocysts in the gastrointestinal tract of dogs as soon as possible to prevent possible disease transmission (Garanayak *et al*., 2017). This case study observed the diagnostic technique, hematological profile, and treatment in dogs infected with the protozoan *Cystoisospora* spp.

MATERIALS AND METHODS

Study Period and Location

This study was carried out in November 2022 at the Parasitology Laboratory and Teaching Veterinary Hospital, Brawijaya University, Malang.

Clinical History

A female domestic dog, aged >1 yo had not been given vaccines or anthelmintic drugs but had been given anti-ectoparasite drugs. The dog has changed consistency slightly soft with a frequency of defecation 1–2 times a day.

Physical Examination

The dog had a normal body temperature of 38°C, weighed of 11 kg, and was rated 4 out of 9 based on Body Condition Score (BCS) (WSVA Global Nutrition Committee, 2013). The abdominal examination resulted in no abdominal distension, no abdominal pain, and normal. Stool examination found soft stool consistency with a score of 5 out of 7 based on Englar's (2019) stool assessment which is very moist but has a different shape, the stool is more in the form of a pile than a rod, then leaves residue on the ground.

Supporting Examination

Supporting examinations carried out were hematological examination and fecal examination at the Brawijaya University Teaching Animal Hospital. A hematology examination was performed using Veterinary Hematology Analyzer Rayto 7600 with normal hematology values based on Weiss and Wardrop (2010). Stool examination using the floating method using saturated sugar solution (Dubey and Almeria, 2019) followed by oocysts per gram (OPG) calculation using McMaster chamber. McMaster two Chamber calculation formula (University of Saskatchewan, 2021):

$$\frac{Total\ oocysts}{0{,}3}\ x\ \frac{V}{m} =$$

Total oocysts = all oocysts that can be counted in all counting rooms. If counting rooms are used: 1 counting room = divisor 0.15; 2 counting rooms = divisor 0.3; 3 counting rooms = divisor 0.45; V stands for the volume of liquid; m stands for the weight of feces mixed.

Treatment

Treatment carried out on dogs confirmed with Cystosisosporiasis was administered sulfonamide sulfadiazine at a dose of 15 mg/kg BW 15 mg/kg orally every 12 hours for 7 days, nitromidazole metronidazole at a dose of 14 mg/kg BW every 12 for 7 days, and Hemafort® blood-boosting vitamins containing Ferrous at a dose of 300 mg orally every 24 hours for 7 days.

RESULTS AND DISCUSSION

Dog were diagnosed with cystosisosporiasis due to infection from *Cystoisospor*a spp. or *Isospora* spp. which was confirmed based on fecal examination using the floating method and found oocysts with two sporocysts found in Figure 1. The oocysts found had a slightly oval shape with two sporocysts. These oocysts are characteristic of *Cystoisospora* spp. or *Isospora* spp. (Saari, 2019). The examination was continued using McMaster for the OPG calculation. Counting the number of oocysts per

Table 1. Hematological profile of dogs with cystoisosporiasis

Parameters	Result	Unit	Normal value (Weiss and Wardrop, 2010)	Interpretation
Leukocytes	22.1	10^3/µl	6–17	Leukocytosis
Lymphocytes	1.4	10^3/µl	1–4.8	Normal
Monocytes	0.4	10^3/µl	0.15–1.35	Normal
Granulocytes	20.3	10^3/µl	3.5–14	Granulocytosis
Erythrocytes	5.44	10^6/µl	5.5–8.5	Anemia
Hemoglobin	8.7	g/dL	12–18	Hemoglobinemia
Hematocrit	40.6	%	37–55	Normal
MCHC	21.4	g/dL	32–36	Hypochromic
MCH	16	pg	19.5–24.5	Hypochromic
MCV	74.6	fL	60–77	Normal
RDW-CV	15.5	%	12–16	Normal
RDW-SD	49.1	fL	35–56	Normal
Platelets	370	10^3/µl	200–500	Normal
MPV	8.8	fL	6.7–11	Normal
PDW	8.1	fL	0.0–50	Normal
PCT	0.325	%	0.0–2.9	Normal
P-LCR	29.4	%	13–43	Normal

Mean Corpuscular Hemoglobin Concentration (MCHC), Mean Corpuscular Hemoglobin (MCH), Mean Corpuscular Volume (MCV), Hematocrit (HCT), Red Distribution Width (RDW), Mean Platelet Volume (MPV), Platelet Distribution Width (PDW), Platelet crit (PCT).

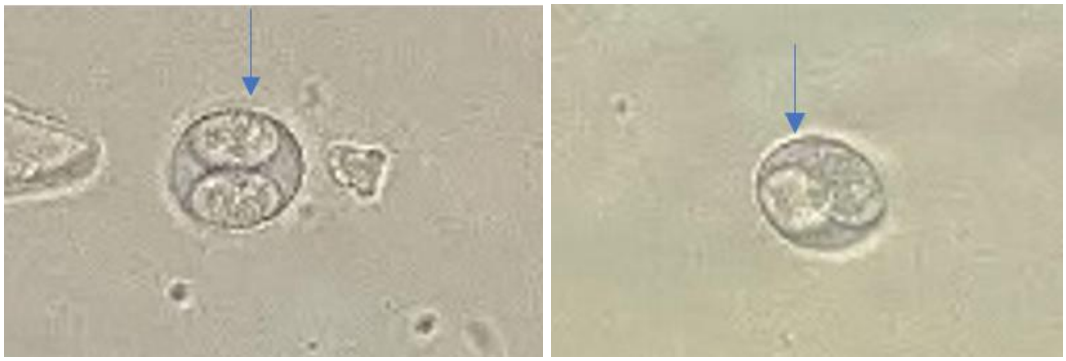

Figure 1. Oocyst with two sporocysts (→) *Cystosisospora* spp. or *Isospora* spp. at 400× magnification found using the floating method.

gram of feces if below 500 oocysts is classified as low infection and more than 500 oocysts as moderate infection while counting the number of oocysts per gram of feces of more than 1000 oocysts is generally considered a severe infection (Permatasari *et al*., 2020). Based on the calculation of oocysts using the McMaster chamber, 50 OPG were obtained, as shown in Figure 2, and included in low infection.

The results of the hematological examination using the Veterinary Hematology Analyzer Rayto 7600 was reported leukocystosis, granulocytosis, and hypochromic anemia (Table 1). Leukocytosis can occur due to infection which encourages white blood cells to work against the infection (Purnama *et al*., 2021). An acute inflammatory leukogram in dogs is characterized by a total leukocyte count usually ranging from 20,000 to 30,000 /µL of blood (Weiss and Wardrop, 2010). Granulocytes are sometimes called granular leucocytes, polymorphonuclear leucocytes, or PMNs (Purnama *et al*., 2019). Granulocytes consist of 3 types: neutrophils, eosinophils, and basophils. Neutrophils have the function of attacking bacteria. Eosinophils function in cooperation with the immune response and also fight parasites. Basophils have the function of fighting allergic reactions by releasing histamine

(Kristanto and Septiyani, 2023). Granulocytosis is often a common picture of acute inflammation that can be caused by bacteria, parasites, or other foreign bodies (Valenciano and Cowell, 2020).

Figure 2. Microscopic examination of a fecal sample showing two oocysts in a McMaster two chamber at 100× magnification. (→) *Cystoisospora* spp. or *Isospora* spp. oocysts.

The red blood cell value is below the normal limit of 5.44 10^6/µL, resulting in anemia in Table 1. Non-regenerative anemia is a relatively common condition in dogs including inflammatory diseases, chronic renal disease, nutritional anemia, or bone marrow diseases (Mizuno *et al*., 2022). Nonregenerative anemia has also been reported in dogs with positive *Cystoisospora* spp. (Zoto *et al*., 2022) and also found in dogs infected with blood parasites (Thongsahuan *et al*., 2022). Hemoglobin, hematocrit, MCH, and MCHC values showed a decrease with MCV values still within normal limits. Mean Corpuscular Volume is a calculation of the average size of red blood cells (Majid *et al*., 2023). The normal condition of MCV is called normocytic (Wicaksono *et al*., 2022). A low MCH value indicates that the amount of hemoglobin in the red blood cells has decreased below the normal value (Septiyani *et al*., 2023). The MCHC value is decreased and is called a hypochromic condition. Based on the hemoglobin, hematocrit MCV, MCH, and MCHC values, it can be concluded that the dog has hypochromic normocytic anemia. Hypochromic anemia often occurs due to a lack of albumin in hemoglobin, it can also occur due to hemorrhage (Tvedten, H., 2022).

Dogs affected by cystoisosporiasis were treated with sulfonamide group antibiotics combined with metronidazole, and the animals showed recovery of clinical symptoms along with elimination of oocysts in feces (Garanayak *et al*., 2017). According to Madani *et al*. (2018), treatment using sulfadiazine can effectively eliminate protozoa and is considered more effective than the antiprotozoal dicrazuril. Sulfonamides are effective against the asexual stage of coccidian development during merogony or schizogony and work as coccidiostatic drugs. Sulfadiazine is potentially more rapidly effective when combined with other drugs such as trimethoprim, ormetoprim, pyrimethamine, metronidazole, and diaveridine as they synergize their anticoccidial activity by blocking parasite metabolic pathways and show less toxicity in animals (Lai *et al*., 2020; Purnomo *et al*., 2022). Administration of blood supplements such as Hemafort® contains ferrous fumarate. Ferrous fumarate is a supplement that replaces iron in animals deficient or with iron deficiency anemia (Papich, 2016).

After one week of treatment, there was a good change in the clinical symptoms of the dog. Stool assessment after 7 days of treatment included a score of 2 out of 7. A stool with a score of 2 is characterized by shaped stool but not hard and flexible stool, with no stool residue on the ground when lifted. A stool with a score of 2 is considered normal stool (Englar, 2019). Good changes in clinical symptoms indicate that the drug is working well. To determine whether there is still infection or not, a re-examination through stool examination with the native and float method confirmed no infection.

Cystoisospora spp. infecting dogs have not been reported as zoonotic, but other species of *Cystoisospora* spp. namely, *Cystoisospora belii* are pathogenic to humans. Therefore, to prevent cystoisosporiasis infection in humans, which is to

check the health of their pets by immediately identifying the presence of oocysts (Garanayak *et al*., 2017). Educating owners is also important for successful therapy, as is outdoor maintenance. Although the effect of outdoor rearing is not significant, it can increase exposure to *Cystoisospora* spp. (Maheswari *et al*., 2023). Keeping the environment and animals clean is also important. Disinfection with chemicals such as ammonia 10% can increase the efficacy of cleaning the cage (Saari, 2019). Pregnant females should be treated and bathed before giving birth to remove sporulated oocysts on their hair coat (Troccap, 2019). Prevention can be done by paying attention to treatment to eliminate oocysts in feces to disrupt the life cycle (Gomez *et al*., 2018). The combination of febantel and pyrantel can also be used as a preventive measure. The combination of febantel and pyrantel eliminates oocysts from protozoa (Papich, 2016) because it has an ovicidal ability that can damage the structure of the oocyst wall by binding to proteins in the oocyst wall (Nurhasanah and Murlina, 2020).

CONCLUSION

Cystoisosporiasis infection caused by *Cystoisospora* spp. Cystoisosporiasis in this case only experiences changes in fecal consistency and leads to asymptomatic. Confirmation of the diagnosis was obtained from the microscopic examination of fecal samples for the presence of *Cystoisospora* spp. oocysts and following McMaster which included low infection from *Cystoisospora* spp. Treatment using sulfadiazine and metronidazole for a week in dogs can effectively eliminate oocysts and restore clinical symptoms.

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AUTHORS' CONTRIBUTIONS

SKR: Conceptualization and drafted the manuscript. KAF and SKR: Performed sample evaluation. SKR: Prepared of table and figures. All authors have read, reviewed, and approved the final manuscript.

COMPETING INTERESTS

The authors declare that they have no competing interests.

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