



Control of *Giardia* infections with ronidazole and intensive hygiene management in a dog kennel

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ABSTRACT

Infections with the intestinal protozoan parasite *Giardia* in dogs and cats are common. Clinical signs vary from asymptomatic to small bowel diarrhea and associated discomfort. The control of infections in dogs is frequently a frustrating issue for animal owners and veterinarians. Drugs with antiprotozoal activity such as fenbendazole and metronidazole are recommended, however, they do not show 100% efficacy and superinfections occur regularly. Ronidazole is currently the drug of choice for the treatment of *Tritrichomonas foetus* in cats and there is now limited information available about its efficacy against *Giardia* spp. In the kennel investigated, dogs regularly showed loose feces and the presence of *Giardia* (assemblage C, renamed as *G. canis*) cysts. An elimination strategy of this parasite involving strict hygiene management and disinfection of the enclosures with 4-chlorine-M-cresol, oral treatment with ronidazole (30–50 mg/kg BW bid for 7 days) and two shampoos (containing chlorhexidine) at the beginning and the end of the treatments was implemented for a group of 6 dogs. As a control another group of 7 dogs was transferred to the disinfected enclosures and shampooed, but left untreated. Dog feces were tested for the presence of *Giardia* cysts (SAF concentration technique) or *Giardia* antigen with a commercial ELISA (NOVITEC®) and a quick immunochromatography-based test (SensPERT®) before and between 5 and 40 days after the last treatment. All ronidazole-treated dogs were negative for *Giardia* cysts and antigen up to 26 days after the last treatment, while between 1 and 5 of the control animals tested positive in each of the test series. At this point, also dogs of the control group were again moved into clean enclosures, shampooed twice and treated with ronidazole. Five, 12 and 19 days after the last treatment, the dogs in the control group tested negative for *Giardia* cysts and antigen. However, all animals had again positive results at later time points in at least one of the three applied diagnostic techniques within 33–61 days after treatment. Furthermore, all dogs had episodes of diarrhea (for 1–4 days) within 14–31 days after treatment and unformed feces during the whole experiment. The positive effect of ronidazole against *Giardia* infections in dogs could be confirmed in this study. In particular, the combination of ronidazole treatment combined with the disinfection of the environment and shampooing of the dogs was highly effective in reducing *Giardia* cyst excretion and may therefore constitute an alternative control strategy for canine giardiasis.

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1. Introduction

Giardia is an intestinal protozoan with a broad host range in wild and domestic mammals. Although the adverse consequences of *Giardia* infection and its pathogenic potential are best recognized in humans

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(Thompson, 2004), it is also a well known causative agent of diarrhea in dogs and cats. Diarrhea is common in both animal species, with many possible causes: non-infectious (stress, disturbances in water balance, nutritional and immune status, malnutrition, neoplasia, inflammatory disease) and infectious (bacterial, parasitic, or viral infections) causes, but also any combination of the above (Payne and Artzer, 2009). Since stress has an effect on the function and the immunological reactions in the gut, it is not surprising that high *Giardia* prevalences were identified among animals housed in stressful situations such as dog rescue shelters (Upjohn et al., 2010) or kennels (Scaramozzino et al., 2009).

Molecular tools are commonly used for the genetic characterization of *Giardia* isolates. Currently, seven *Giardia* genotypes, designated assemblages and in some cases assigned distinct species names, are recognized. Dogs are infected by parasites of four assemblages (A, B, C, D), of which assemblages C and D (also defined as *G. canis*) are found exclusively in dogs, while parasites of assemblages A and B (also defined as *G. enterica*) are zoonotic (Covacin et al., 2011; Thompson, 2004; Thompson and Monis, 2011).

Giardia cysts are therefore frequently found in routine diagnostic examination of dog feces, also from asymptomatic dogs (Covacin et al., 2011). In a recent study performed with 878 shelter dogs (Upjohn et al., 2010), the apparent prevalence of *Giardia* was 9.9% and the true prevalence, based on the known sensitivity and specificity of the ELISA test, was 21.0%, which is in the same range as found in previous studies. Since stress has an effect on the function and the immunological reactions in the gut, it is not surprising that high *Giardia* prevalences were identified among animals housed in stressful situations such as dog rescue shelters (Upjohn et al., 2010) or kennels (Scaramozzino et al., 2009). In addition, fecal samples that were graded concerning their consistency, confirmed previous results, i.e. a weak association between fecal consistency and infection with *Giardia* in dogs.

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in vitro (Cedillo-Rivera et al., 2002), while azythromycin, an azalide, has been used for the treatment of only one dog (Zygner et al., 2008). Therefore, further experiments are required to confirm the efficacy of these drugs against *Giardia* infection (Geurden and Olson, 2011). Ronidazole and tinidazole are also nitroimidazoles, and while the latter has recently been approved in the United States for the treatment of giardiasis in humans, ronidazole has been used for treatment of blackhead disease, caused by *Histomonas meleagridis* in turkeys. In addition, ronidazole is currently the drug of choice against *Tritrichomonas foetus* in cats (Gookin et al., 2006). A high antiprotozoic effect was demonstrated in vitro against *G. duodenalis* with an approximately fivefold higher activity than metronidazole (Boreham et al., 1985). The same authors also reported good efficacy of ronidazole against *Giardia* sp. in mice (Boreham et al., 1986).

Although several compounds are effective against *Giardia*, control programs combining drug treatment with cleaning and disinfection of the environment to reduce the environmental infection pressure are recommended (Geurden and Olson, 2011). Studies showed that calves as well as dogs re-excreted cysts shortly after the end of antiprotozoic treatment if no hygienic measures were implemented (Geurden et al., 2006; Villeneuve et al., 2000). In addition, thorough shampooing of companion animals is recommended after treatment to prevent reinfection through fecal material on the fur (Payne et al., 2002; Zajac et al., 1998). The aim of the present study was to assess the efficacy of ronidazole against *Giardia* infections in a dog kennel.

2. Materials and methods

2.1. Facility, animals, management

In the animal facilities of the Veterinary Faculty of the University of Zurich, beagle dogs are housed in groups of 2–4 in pens of 1.45 m × 4.5 m in size with access to an outside run of 1.45 m × 5.5 m. Some adjacent pens share a common outside run of 3 m × 11 m with a concrete floor. Pens are enriched by installations allowing dogs to jump and use the space tridimensional, as well as to rest and retreat.

The total number of dogs in the facility varies between 12 and 42 with an age range between puppies and 6 years. Dogs are tested regularly (every 3 months) for the presence of parasites in their feces by sedimentation/flotation and by SAFC-technique (Eckert et al., 2008). For individual fecal samples, dogs are isolated overnight. Occasionally, some dogs presenting *Toxocara canis* infections are treated with an anthelmintic compound. Most of the dogs occasionally show loose feces but only in some of them *Giardia* cysts are detected. Prior to the start of this study, *Giardia* of the 'dog genotype' assemblage C was identified by PCR/sequencing of part of the 18S rRNA gene (Hopkins et al., 1997). Daily cleaning of the pens with a cleaning agent (Allzweckreiniger 681, Kärcher AG, CH-8108) is combined with the use of a disinfectant (Incidin® PLUS, Ecolab GmbH, 4132 Muttentz) twice a week in dosages according to the instructions of the manufacturers. The dogs are fed once

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