Use of bait containing triclabendazole against *Fasciola gigantica* in a herd of captive wild impala (*Aepyceros melampus*)

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Abstract

Although the efficacy of triclabendazole treatment against *Fasciola gigantica* has been shown before in cattle and buffaloes, there appears to be no published report on the efficacy of triclabendazole in impala or other antelope species. As part of a health monitoring programme at Mulungushi International Conference Centre, Lusaka, Zambia, a coprological examination was undertaken to investigate the helminthological status of captive impala (*Aepyceros melampus*). Of 39 fresh coprological samples, 46% contained *F. gigantica*. The source of infection was identified to be a fountain within the grounds of the conference centre. *Lymnaea natalensis*, collected from the study site, were induced to shed cercariae, and were thus confirmed as the snail intermediate host. In managing this disease, triclabendazole at 6 mg/kg was administered together with the feed bait. Water from the pond was drained; vegetation within it cleared and the pond allowed to dry for 1 week before water was replaced. Three weeks post-treatment, faecal examination revealed that the *Fasciola* had been cleared. To the best of our knowledge, this is a first case of *Fasciola* spp. infection reported in captive wild impala in Zambia and provides evidence that triclabendazole may be delivered to free-ranging antelope using medicated bait.

Introduction

Of the many species of game held in captivity in Zambia, impala (*Aepyceros melampus*) are the most abundant antelopes. There are increasing concerns worldwide, particularly in cases where management of wildlife species can influence dynamics of disease spillover into domestic animals or threaten wildlife conservation efforts through increased parasite transmission (Hines *et al.*, 2007; Phiri *et al.*, 2007). Helminths have been reported in impala (Ezenwa, 2004a; Nalubamba & Mudenda, 2012; Nalubamba *et al.*, 2012). While these are tolerated by wild animals, with insignificant effects on their health, helminth burden may increase when animals are kept in captivity (Ezenwa, 2004b).

There is limited information on fasciolosis in impala. However, with increased captive wildlife in private game ranches in southern Africa, awareness, knowledge, treatment and prevention of this disease in naturally infected wildlife are needed. Therefore, the present study reports on fasciolosis in captive wild impala and explores how feed baited with an anthelmintic can be used to manage the disease. Furthermore, other studies of triclabendazole treatment in domestic and wild ruminants are compared. Fasciolosis is a parasitic disease of a wide range of mammals, which is caused by *Fasciola gigantica*. Aquatic snails are the intermediate hosts. This disease presents either as an acute or chronic form. The acute form is commonly associated with sudden death, while the chronic form often shows no specific clinical signs. Knottenbelt (1990) identified acute fasciolosis due to *F. gigantica* as the main disease problem associated with smaller game farms, especially where impala had been introduced recently. Fasciolosis has also been detected at slaughter in a semi-aquatic antelope, the Kafue lechwe (*Kobus leche kafuensis*) (Phiri *et al.*, 2011).

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Materials and methods

Collection and examination of faecal samples

The study was carried out in impala in August and September, 2014 at the Mulungushi International Conference Centre (MICC) located within the city of Lusaka, Zambia. Impala are medium-sized antelopes, which normally weigh between 40 and 65 kg. The centre had three water-points, with one of them being a concrete-lined fountain containing aquatic grass. Impala had been at the site for over a decade and there was no record of de-worming. No supplementary feeding was practised. Four impala were reported to have died a few months previously and no post-mortems were performed to establish the cause of death.

A coprological examination (faecal egg counts) of 39 samples from impala of mixed ages was undertaken to investigate their helminthological status. Only the top un-contaminated faecal pellets were sampled during morning hours. Faeces were processed using the Modified McMaster counting technique and sedimentation methods (MAFF, 1980) at the Department of Clinical Studies, School of Veterinary Medicine, The University of Zambia.

Snail sampling and cercarial shedding

Twenty snails were removed from the fountain and surrounding vegetation using gloved hands, and identified according to morphological features of the shell (Brown & Kristensen, 1989) before being transported in fountain water to the laboratory. Shedding of cercariae was performed as described by Phiri *et al.* (2007). Living and unstained cercariae were identified based on morphological characteristics, swimming behaviour and resting position, as described by Frandsen & Christensen (1984).

Treatment of the animals with baited feed

Triclabendazole 10% m/v (ECO Animal Health, London, UK) was used to treat impala at an estimated dose of 6 mg/kg. The weight of each animal was estimated at 50 kg on average, with a herd size of about 50 antelopes; 150 ml of the drug was added to 10 litres of water, and the solution mixed with 25 kg of game-feed pellets (Tiger Brands, Sandton, South Africa). To ensure easy access of impala to the feed, the bait was placed at three sites, taking into account three groups, namely territorial males, a bachelor herd and a group comprising adult females and their young. The current method of choice to determine efficacy of an anthelmintic in veterinary medicine is to compare infection levels before and after treatment. Infection levels can be measured either by worm counts (method of choice) or faecal egg counts (FEC). Faecal samples are thus collected from at least ten animals with positive counts (Presidente, 1985).

Efficacy of triclabendazole

A faecal egg count reduction test (FECRT) was carried out according to Kochapakdee *et al.* (1995) to determine the efficacy of triclabendazole. Faecal egg counts before and after treatment were evaluated. The test dealt with arithmetic means and no untreated control group, because each host served as its own control. The following formula was employed:

$$\text{FECR} = (1/n) \sum \{100 \times [1 - (\text{Ti}_2/\text{Ti}_1)]\},$$

where Ti_2 is post-treatment and Ti_1 is pre-treatment eggs per gram (epg) in host i from a total of n hosts.

Data analysis

Results for worm infection were recorded as FECs. For an animal to be considered positive for fasciolosis, it needed to have at least 1 epg. Descriptive statistics were used to describe the infection. Prevalence was the proportion of positives from the whole herd. No correlation was determined as it was difficult to establish age, weight and sex when faeces were being collected. Variation was recorded as standard error of the mean. For cercarial shedding, a binary method was used to describe whether shedding was present or not. The result of FECRT was given as the difference between before- and after-treatment FECs.

Results and discussion

Of 39 faecal samples, 46% were positive for *F. gigantica* (mean EPG ± SEM: 138.7 ± 33; range 0–750) while 26% were positive for strongyle-like helminths (mean EPG ± SEM: 69.3 ± 27.2; range 0–600) which could not be identified without culturing. Snails were identified as *L. natalensis* and 16 (80%) shed gymnocephalous cercariae. This is a common form represented within the *Fasciola* species, having two suckers of equal size and developing within rediae. Identification of *Lymnaea* snails was mainly based on colour, turreted appearance and position of the aperture when facing the observer.

Animals were able to eat shortly after baited-feed was placed at each site. To avoid re-infection, grass growing in the fountain was removed. The pond was drained of water and left to dry for a week before refilling it with fresh water. Three weeks after treatment, 39 coprological samples were also collected and examined as described previously. No *F. gigantica* eggs were detected in faecal samples, signifying a 100% FECR. However, 35.5% (11/39) of the samples tested positive for strongyle helminth eggs (mean EPG ± SEM: 62.9 ± 29.1; range 0–750).

This is a first case of *F. gigantica* reported in impala. Furthermore, this present study confirmed that anthelmintics can be delivered via bait to free-ranging antelopes. The impala herd was at risk since the animals were mostly found grazing on the aquatic grass around the fountain and drinking water from it. Infected snail intermediate hosts were also present in that environment.

Few studies have been done in Zambia on fasciolosis; most are in cattle (Phiri *et al.*, 2005, 2007; Yabe *et al.*, 2008; Munyeme *et al.*, 2012) and fewer are on captive impala (Nalubamba & Mudenda, 2012; Nalubamba *et al.*, 2012) and Kafue lechwe antelope (Phiri *et al.*, 2007). Another study at a private game ranch did not find trematode eggs (Nalubamba *et al.*, 2012). These studies have shown a cattle–wildlife interaction and resultant bi-modal transmission of a wide range of parasitic helminths

between them. Furthermore, non-species-specific Bovidae helminth parasites easily infect multiple host species (Boomker *et al.*, 1986; Waruiru *et al.*, 1995; Phiri *et al.*, 2007, 2011).

In Zambia, trematode infections in wild and domestic ruminants are highly prevalent in southern, central and western provinces (Phiri *et al.*, 2005). However, the present study has shown that infections can also be maintained outside those provinces as long as suitable conditions exist. In order to reduce the risk of further infection or re-infection among the impala, triclabendazole was administered through feed, and post-treatment evaluation was done 3 weeks later. Triclabendazole, being a flukicide of choice for both immature migratory flukes and mature adult flukes in bile ducts (Fairweather & Boray, 1999), eliminated the flukes, as no trematode eggs were detected after treatment. The snail habitat was mechanically destroyed and left dry for several days, thereby reducing the likelihood of snail survival and involvement in the transmission cycle. Although efficacy of triclabendazole treatment against *F. gigantica* has been shown before in cattle (Suhardono *et al.*, 1991) and buffaloes (Sanyal & Gupta, 1996), there appears to be no published report on the efficacy of triclabendazole in impala or other antelope species. The recommended dose of triclabendazole in naturally infected cattle is 12 mg/kg given orally every 8 weeks for 1 year (Suhardono *et al.*, 1991). Surprisingly, the therapeutic dose used in cattle (i.e. 12.0 mg/kg body weight) was only 19–23% effective. In wildlife affected by *Fascioloides magna*, such as deer, 11 mg/kg has been used effectively (Quereshi *et al.*, 1994). In the present study, half the dose at 6 mg/kg was efficacious. However, Sanyal & Gupta (1996) reported an even higher dose of intraruminal administration of triclabendazole, at 24.0 and 36.0 mg/kg, in buffaloes experimentally infected with *F. gigantica*. Integrated control of fasciolosis has also been documented with other flukicides, such as rafoxanide, diamphenetide, oxyclozanide, albendazole, closante and clorsulon. When single oral doses of clorsulon (12–30 mg/kg of body weight) and albendazole (17–46 mg/kg) were given per deer, the efficacy was 92% and 67%, respectively (Foreyt & Drawe, 1985). Ivermectin has also been used in deer (Garris *et al.*, 1991). Therefore, control measures can be employed aimed at reducing infections through use of anthelmintics, eliminating the intermediate host through use of molluscicides or mechanical removal, and reducing parasite transmission through good management practices.

Good nutrition, containing protein, plays a role in resistance and resilience to gastrointestinal parasite infection in wild ruminants (Ezenwa, 2004b). Unlike sheep, which are known for expelling *Fasciola* after several months, many other ruminants harbour it for years (Pleasance *et al.*, 2011). Therefore, it is recommended that newly introduced animals are dewormed, especially if they are sourced from a fasciolosis-endemic area. Nalubamba *et al.* (2012) documented *Haemonchus* spp., *Trichostrongylus* and *Strongyloides* spp. as being the most predominant parasites during coprological examination in captive wild impala conducted at a game ranch in Zambia. Anthelmintic treatment can be administered for more than 3 days at a time to achieve maximum benefit. Nonetheless, anthelmintic treatment must be approached

with caution as there are some reports of anthelmintic resistance in antelopes (Nalubamba & Mudenda, 2012).

In conclusion, the present study found that *Fasciola* spp. infection was prevalent among impala kept at MICC in Lusaka, outside known endemic areas. It was also found that triclabendazole was efficacious in treating the infection. For monitoring levels of parasitic infections, determining quantitative faecal egg counts becomes an important tool for identifying herd-health problems.

Conflict of interest

None.

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