









Two treatment protocols for parasitic otitis caused by *Rhabditis* spp. in Gir cattle

[Resultado de dois protocolos de tratamento para otite parasitária em gado Gir]

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ABSTRACT

Parasitic otitis in cattle is normally caused by nematodes of the genus *Rhabditis* spp. and is a common problem in Gir breed. Parasitic otitis can cause premature culling and even death of animals. Despite its importance, there is no effective standardized treatment protocol for this condition. The objective of this study was to evaluate two treatments for parasitic otitis caused by *Rhabditis* spp. in Gir cattle. The external left ear of the animals was washed with 2% boric acid alcohol, while the right ear received topical application of Triclorfon, allantoin and zinc oxide paste. The treatments were performed weekly in the first month and monthly over the next two months. Monthly, the ears of all animals were washed under pressure with a physiological solution before treatment. The recovered solution was analyzed, and the number of nematodes counted. Neither treatment was effective in reducing the number of nematodes recovered or the otitis score, but weekly treatment seemed more promising, independent of the drug used.

Keywords: *Rhabditis* spp., nematodes, bovine, ear, otitis, *Raillietia*.

RESUMO

A otite parasitária em bovinos é provocada normalmente por nematóides do gênero *Rhabditis* spp. e é um problema comum na raça Gir. A otite parasitária pode causar o descarte prematuro e até a morte de animais. Apesar de sua importância, não existe um protocolo de tratamento eficaz padronizado para essa condição. O objetivo deste estudo foi avaliar dois tratamentos para otite parasitária causada por *Rhabditis* spp. em gado Gir. Animais afetados tiveram a orelha externa esquerda lavada com álcool bórico a 2%, enquanto a orelha direita recebeu aplicação tópica de triclorfon, alantoina e pasta de óxido de zinco. Os tratamentos foram realizados semanalmente no primeiro mês e mensalmente nos dois meses seguintes. Mensalmente, as orelhas de todos os animais foram lavadas sob pressão com solução fisiológica antes do tratamento. A solução recuperada foi analisada, e o número de nematóides contado. Nenhum dos tratamentos foi eficaz na redução do número de nematóides recuperados ou do escore de otite, mas o tratamento semanal parece ser mais promissor, independentemente da droga utilizada.

Palavras-chave: *Rhabditis* spp., nematóides, bovinos, orelha, otite, *Raillietia*

INTRODUCTION

Parasitic otitis caused by *Rhabditis* spp. is a common condition in Gir cattle and is widely distributed in Brazil and other tropical countries (Odongo and D'Souza, 1989; Duarte *et al.*, 2001; Leite *et al.*, 2012). Parasitic otitis can also be caused by mites of the genus *Raillietia*, and secondary infection by bacteria and fungi can occur, causing inflammation, auditory canal

stenosis, and purulent discharge. This condition causes great distress; animals repeatedly shake their heads, and without treatment, progress to internal otitis can lead to neurological symptoms (Leite *et al.*, 2013). Besides its negative impact on the animal's well-being, parasitic otitis has a great economic impact due to the cost of treatment, use of manpower, and potential culling of the animal. Consequently, it is a persistent problem for dairy producers (Vieira *et al.*, 2006; Leite *et al.*, 2012).

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The Gir breed has been commonly used as foundation stock for Girolando breeding for milk production in Brazil and other tropical climate countries. This breed stands out mainly because of its resistance to heat stress and ectoparasite infestation, and higher than average milk production compared to other Zebu breeds (Costa *et al.*, 2015). However, due to the morphological characteristics of the head and ears of the Gir, it is predisposed to parasitic otitis by *Rhabditis* spp. and mites of the genus *Raillietia* (Duarte *et al.*, 2001).

A wide variety of methods and products are used for the treatment of parasitic otitis caused by *Rhabditis* spp., presenting good results with the use of nematophagous fungi associated with ivermectin and dimethyl sulfoxide (Sobral *et al.*, 2019; Ferraz *et al.*, 2019; Sobral *et al.*, 2022). However, other protocols tested have shown inefficiency in parasitic control or inconsistent results (Verocai *et al.*, 2009; Leite *et al.*, 2013, Barbosa *et al.*, 2016). This study aimed to evaluate the efficacy of two types of drug treatments for parasitic otitis caused by *Rhabditis* spp. in Gir cattle.

MATERIALS AND METHODS

This research was approved by the Ethics Committee for Animal Experimentation of the Universidade Vila Velha under protocol number 306/2014. The study was conducted at a property in the municipality of Anchieta, Espírito Santo state. The location has a humid, tropical climate and average temperature of 27 °C. We used twenty Gir animals, aged between 4 and 15 years, that were kept on pasture and received supplementation of corn silage and concentrate. The last deworming was done more than 60 days before the start of the study.

All animals received two treatments (T1 and T2), one in each ear. In the T1 group (performed in the left ear), the treatment consisted of washing the ear canal using 50 mL of 2% boric alcohol with the aid of a syringe and an adapted macro dropper as described by Leite and Faccini

(1994). The T2 group (performed in the right ear) received a paste applied topically to the external ear canal. This paste contained a Neguvon®-based ointment (Triclorfon, Bayer Health Care®, Germany) mixed with Alantol® (Allantoin/Zinc Oxide, Vetnil®, Brazil) in a 1:1 ratio. In the first month, the treatments were done weekly, and then treatments were done every month for another two months, totaling three months of treatments and seven applications.

It was decided that the experiment should be performed with a positive control because of animal well-being and not deny treatment to any animal. Furthermore, studies have shown that *Rhabditis* spp. infestation is not related to climatic factors with no variation in time (Duarte *et al.*, 2001; Leite *et al.*, 2013).

Parasite quantification was done initially, and then monthly by washing the external ear canal with 50mL of saline solution, totaling four samples. The recovered liquid was stored in Falcon tubes. The tubes were transported to the laboratory, then centrifuged at 3000 rpm for five minutes. The supernatant was discarded and the remaining 3mL was homogenized and 10µL was pipetted on a microscope slide. *Rhabditis* spp. were counted under a 4x objective microscope. The otitis score was also evaluated as defined by Leite *et al.* (2013) with minor modifications, as described in Table 1. The first recording, to quantify initial nematode infestation and otitis score, occurred before the beginning of treatment (D0), and other collections were then made monthly, always before the next application of the treatment.

The data obtained were tabulated in spreadsheets and statistical analysis was performed using SAEG software (Viçosa, Brazil) using the chi-square test to compare otitis scores between ears (T1/T2) and between monthly and weekly treatments, and the Tukey test to compare the amount of *Rhabditis* spp. recovered at each collection time, between treatments T1 and T2 and at the different intervals of administration of the interventions.

Table 1. Otitis clinical classification (score 0 to 3) used for Gir cattle. Adapted from Leite *et al.* (2013)

Otitis score	Description
(0) Negative	Ear and external auditory canal with intact skin, clear aspect, and a lack of secretion. Cerumen in small quantities with a yellowish or brown color and dry aspect.
(1) Subclinical	Normal general aspect. Absence of visible alterations. Presence of parasites only detectable by parasitological diagnosis.
(2) Mild	Normal general aspect but with some degree of distress. Small quantity of yellowish fluid secretion.
(3) Severe	Distress with repeated head movements. Auditory canal stenosis with large amounts of purulent secretion with a strong odor.

RESULTS

All animals presented *Rhabditis* spp. in both ears during the entire observation period, but no sample presented mites of the genus *Raillietia*.

No statistical differences were observed between the time intervals and the number of *Rhabditis* spp. recovered, or the otitis score ($p>0.05$; Fig. 1 and Fig. 2). The mean number of *Rhabditis* spp.

recovered before the beginning of treatment was 49 ± 46.8 (mean \pm standard deviation) and 45 ± 42.1 for groups T1 and T2, respectively. After one month of weekly treatments, the mean number of *Rhabditis* spp. recovered was 32 ± 29.5 and 26 ± 25.7 for group T1 and T2, respectively. At the end of the treatment period 51 ± 57.7 and 39 ± 64.4 mean *Rhabditis* spp. were recovered from groups T1 and T2, respectively (Fig. 1).

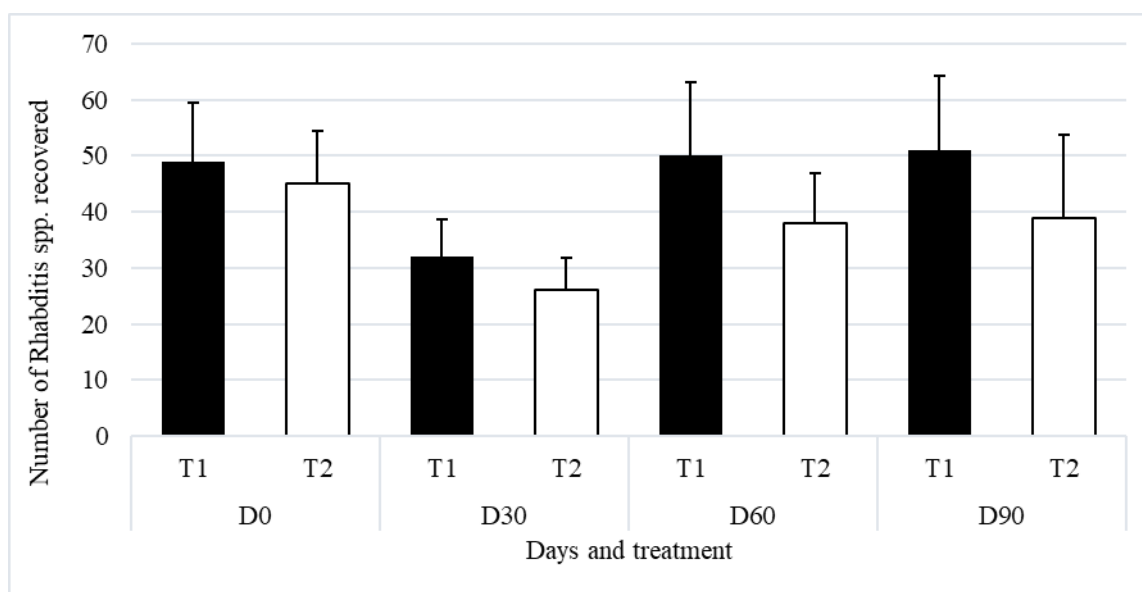


Figure 1. Number of nematodes of *Rhabditis* spp. recovered at each experimental time interval after washing the external ear canal of Gir cattle with a saline solution after 0, 30, 60 and 90 days of treatment with Treatment 1 (T1; otitis treated with 50 mL of 2% boric alcohol) and Treatment 2 (T2; otitis treated with topical paste containing Triclorfon, allantoin and zinc oxide in a 1:1 ratio).

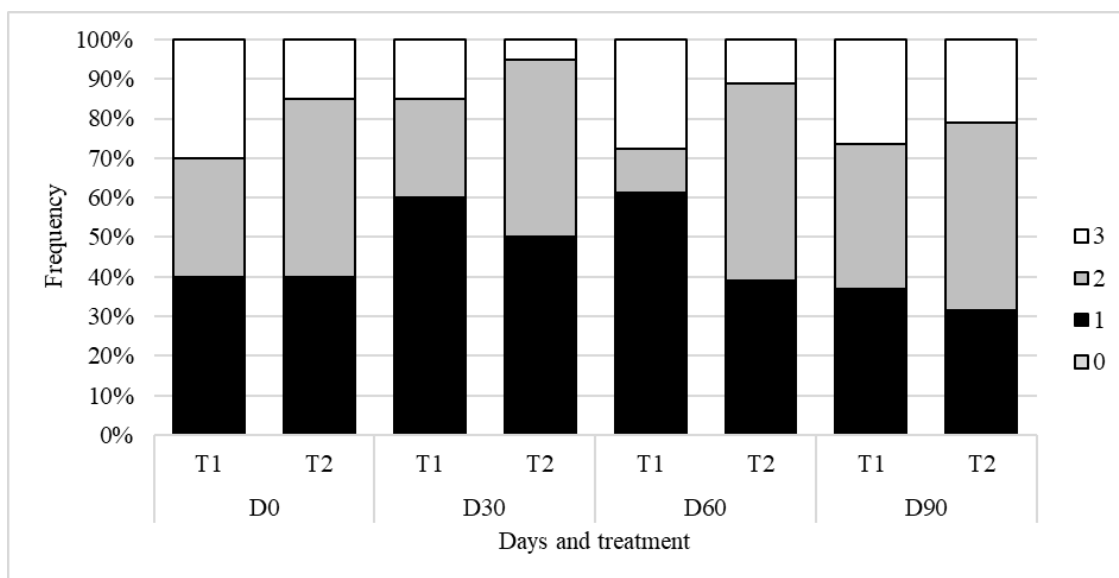


Figure 2. Otitis score (0 to 3) frequency observed in Gir cattle with parasitic otitis after 0, 30, 60 and 90 days of treatment with Treatment 1 (T1; otitis treated with 50 mL of 2% boric alcohol) and Treatment 2 (T2; otitis treated with topical paste containing Triclorfon, allantoin and zinc oxide in a 1:1 ratio).

The frequencies of the different otitis scores (on a scale of 1 to 3) are presented in Fig. 2. The mean otitis scores in the T1 group were 1.90, 1.55, 1.67, and 1.76, for the initial score, and then after one, two and three months of treatment, respectively. In T2 group, the mean score was 1.75 before treatment and 1.55, 1.67, and 1.76 after one, two and three months of treatment. There was no statistical difference in the otitis score between treatments at any moment or between monthly and weekly treatments.

DISCUSSION

Both proposed treatments were ineffective in reducing the number of *Rhabditis* spp. recovered from the ears and in reducing the otitis score, independently of the frequency of the two treatments offered. This result is similar to Leite (2013) and Vieira *et al.* (2006) who both describe an unsuccessful outcome using Triclorfon® paste and Leite and Faccini (1994) had no success with boricated alcohol. The treatment for parasitic otitis caused by *Rhabditis* spp. is controversial and presents inconsistent results (Odongo and D'Souza, 1989; Verocai *et al.*, 2009; Leite *et al.*, 2013; Barbosa *et al.*, 2016).

It is important to clarify that the authors decided to perform the study only with a positive control

due to animal well-being implications, in order not to leave any animal untreated. Additionally, no correlation was observed between temporal or climatic changes and infestation by *Rhabditis* spp. in studies that followed the dynamics of auricular parasitosis in Gir cattle for up to one year in Minas Gerais (Duarte *et al.*, 2001; Leite *et al.*, 2013), a state next to where this study was performed.

All animals (100%) in the current study were parasitized by *Rhabditis* spp., which is higher than the proportion reported by Leite *et al.* (2013), who reported a prevalence of 52% in Gir cattle between 13 and 33 months of age, and Duarte *et al.* (2001) who found 60%. This is probably because all animals enrolled in the study were older with at least 4 years old, and the frequency of infestation by *Rhabditis* spp. increases with age (Duarte *et al.*, 2001; Leite *et al.*, 2013). In the present study, the parasitic otitis was caused by rhabditiform nematodes while no mites of the genus *Raillietia* spp. were observed. This is similar to results reported by Duarte *et al.*, (2001) and Leite *et al.* (2012), who found that parasitic otitis by *Rhabditis* spp. was much more common than that caused by *Raillietia* spp. The ineffective results of both topical treatments in the current study could be caused by many factors. More than 50% of the ears presented purulent secretions (scores 2 and 3) at most of

the recorded treatment intervals, demonstrating the presence of a secondary infection. In this case, the use of antibiotics would be indicated to reduce the otitis score.

Between D0 and D30, when the treatment was performed weekly, the T1 and T2 groups presented a reduction of 35% and 42% in recovered *Rhabditis* spp., respectively, but no statistical difference was found. Between D30 and D60, the treatment was made monthly, and the number of recovered *Rhabditis* spp. increased by 56% and 46% in T1 and T2, respectively. Again, no statistical difference was found. Based on these results, weekly treatment is promising, and it may require a longer period of treatment and a larger number of animals to observe a statistical difference. This is especially true when we observe the large standard deviation found, what accounted for the non-statistical significance between treatments and intervals. Thereafter, future studies are needed to address these observations.

Another possible cause of treatment failure is the high frequency of reinfection (Duarte *et al.*, 2001; Leite *et al.*, 2013). Because the entire herd was not treated, it is possible that the animals were re-infected between treatments. Thus, it is possible that the treatment protocol should address all animals in the herd.

CONCLUSION

In conclusion, both treatments were ineffective in reducing the number of *Rhabditis* spp. and in reducing the otitis score in Gir cattle. Other studies are necessary to develop a treatment protocol because parasitic otitis is a challenge for Gir breeders. This study should specifically address the frequency of treatments, the inclusion of antibiotics if a secondary infection is present, and the necessity of treating the entire herd.

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