



**ORIGINAL RESEARCH PAPER**

**Zoology**

**Anthelmintic Efficacy Of Ivermectin, Fembendazol And The Fembendazol And Triclorfon Association In Equine**

**KEY WORDS:** horse, helminths, parasites, anthelmintic resistance

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**ABSTRACT** The objective of this study was to evaluate the anthelmintic efficacy of fembendazole and ivermectin and of the association fembendazol and trichlorfon in horses raised in the municipality of Janaúba, MG. Egg counts per gram of faeces were measured in 32 animals. In property A was tested as Ivermectin bases and an association of fembendazole and trichlorfon, and in Property B, tested fembendazole after ten days of antihelmintic administration. The EPG counts were used to calculate the reduction test on egg count per gram of faeces (FECRT). (LCL) at 95% and FECRT > 95% for macrocyclic lactones and > 90% for benzimidazoles / organophosphates were created to establish an adequate criterion. 90 and 80% LCLS were selected for resistance to ivermectin and fembendazole / fembendazole and trichlorfon, respectively. Resistance to fembendazole on property B was suspected. The hypothesis of resistance to ivermectin and fembendazol + trichlorof in property A was detected.

**INTRODUCTION**

The success of animal production is based on genetics, nutrition and health management techniques, with gastrointestinal nematodes being identified as one of the main problems in production systems (Gentil and State, 2012). These are responsible for causing various clinical signs ranging from minor abdominal discomfort fulminant to episodes of cramps and even death in horses (Canever et al., 2013).

Probably due to the prolonged transmission period and the intense use of anthelmintics in Brazil, the rate of resistant gastrointestinal nematodes in equines is worrisome. It is also possible that high levels of ultraviolet light in subtropical latitudes result in mutations in grazing livestock larvae allowing them to survive anthelmintic treatment (Molento et al., 2008).

The *Cyathostomideos* (or small *Strongylus*) are the most problematic nematodes which affect horses of all ages, because of its high prevalence, ability to acquire anthelmintic and potential of pathogenicity resistance (Stratford et al, 2014; Kooyman et al, 2016). Three classes of anthelmintics are currently licensed for their control, benzimidazoles (fembendazole), tetrahydropyrimidines (pirantel) and macrocyclic lactones (ivermectin and moxidectin), with no new classes expected in the near future, it is essential to determine the efficacy of Anthelmintics available to inform future control programs (Stratford et al., 2014).

Anti-helminth resistance is defined as a significant increase in the number of individuals in a given population capable of withstanding doses of a chemical compound that has proven to be lethal to most individuals in a normally sensitive population of the same species. This ability to survive future exposures of a drug can be transmitted to its offspring (Holsback et al., 2016).

The objective of this study was to verify the anthelmintic efficacy of ivermectin, fembendazole, and the combination of fembendazol and trichlorfon in equine nematodes from two properties in the city of Janaúba, Minas Gerais.

**MATERIAL AND METHODS**

The experiment was approved by the ethics committee on experimentation and animal welfare of UNIMONTES CEEBEA 076/2014.

The study was conducted in two properties in the municipality of Janaúba-MG. Thirty - eight pure and mestizo adult male and female horses from the Mangalarga Marchador and Quarter Horse breeds were used, of which 19 were from the "A" farm and 13 were from the "B" farm. The animals were identified and the groups were divided according to the number of eggs per gram of faeces (EPG).

The criteria adopted for the selection of the animals were: the positive diagnosis for gastrointestinal nematodes by the EPG count equal to or greater than 100. The choice of animals was made by those who would not have been treated with any anthelmintic drug for a minimum and antecedent period Of 45 days, the minimum time required for the drug to be eliminated from the animal's organism.

In the A property, the horses were divided into two groups, containing seven animals in group I, and six in group II, being group I: treated with ivermectin and group II with fembendazol + trichlorfon. In property B, only one group with six animals was made, which was treated with fembendazole.

The anthelmintics were administered with a syringe containing the paste for each animal orally. The animals treated with Provermin

paste (fembendazol) at a dose of 7.5 mg per kg of body weight, for Supramec Gel (ivermectin) the dose was 200 mg per kg of body weight and for Provermin Plus (fembendazol + trichlorfon) the dose was 7.5 mg of fembendazole per kg of body weight and 22.5 mg of trichlorfon per kg of body weight in a single dose. The doses used were recommended according to the manufacturers of each product.

Stool samples were collected directly from the rectal ampulla of each animal, and up to 48 hours after collection the animals were wormed. After 14 days on each farm, other faecal samples were collected from the same animals to verify the effectiveness of the vermifuge used. The counting of OPG was done according to the technique described by Gordon and Whitlock (1939) modified (Ueno and Gonçalves, 1998). The reduction test in egg count per gram of faeces (FECRT) was calculated by following The formula described by Coles et al., (1992):

$$FECRT (\%) = \frac{(\text{day 0 EPG} - \text{EPG day 14}) \times 100}{\text{EPG Day 0}}$$

It was used to estimate the 95% confidence limits for each property to bootstrap analysis. The cutoff limits for determining the efficacy of each of the three anthelmintic classes used following the recommendation of arithmetic means thresholds of the population with RCOF > 95% for macrocyclic lactones and > 90% for benzimidazole / organophosphates were designed to establish adequate efficacy (Kaplan and Nielsen, 2010), in addition, the confidence limits of 95% lower (LCLS) were included to provide a more accurate indication of the data range (Vidyashankar et al. 2007) LCLS 90 and 80% Were selected for resistance to ivermectin and fembendazole / fembendazole and trichlorfon, respectively. Therefore, if the percentage of RCOF and LCL is below these designated cut-off points, anti-helminth resistance will be indicated. Alternatively, if either the percentage of RCOF or LCL is below the cut-off point, resistance will be suggested.

**RESULTS AND DISCUSSION**

In the A property, the bases ivermectin and fembendazol + trichlorfon were applied in 13 of the 19 evaluated animals. The mean FECRT was 100% for ivermectin and 96.15% for fembendazole + trichlorfon. The LCL was 100% and 98.72% for ivermectin and fembendazol + trichlorfon respectively (Table 1).

Resistance to fembendazole was tested on property B in 6 animals and the samples were obtained in 13 animals in total. A reduced efficacy was recorded, with the mean FECRT percentage of 89.94. On the property in question, the average percentage of FECRT was below the designated limit of 90%. The calculated LCL was 99.13% for the animals treated with fembendazole.

In property A, there was no resistance and no suspicion in any of the bases used. Fembendazole associated with trichlorfon increased efficacy, since the union of active antiparasitic principles increases the spectrum and the period of action of the same. The group of animals treated with ivermectin was 100% effective, that is, of greater efficiency in egg reduction in feces when compared to fembendazol + trichlorfon, that is because ivermectin is a base of long persistence in the organism (Molento et al., 2012)

Kaplan et al., (2003) determined the efficacy of 99% for ivermectin in equines, but for fembendazole it was 31% effective, a result different from that found in this study.

On property B, a linear reduction in the EPG count 83.33 and 75 with 10 and 15 days respectively in the animals treated with fembendazole was observed. Despite the considerable reduction of eggs in the fembendazole group, resistance is suggested, since only the LCL (96.16%) was above the designated cutoff point of 80% while the FECRT (81.07%) was below the 90% point selected for resistance rating.

In the study by Sanna et al. (2016), there was no evidence of anthelmintic resistance in the 74 animals used on the island of

Sardinia in Italy. Fembendazol had 99.5% efficacy on day 7 and 99% on day 14. Cyathostomides were predominant (96.4%) in equines, also presented by (Molento et al., 2012 and Peregrine et al., 2014). The absence of drug resistance in the examined population of horses on the island of Sardinia could be related to the frequency of anthelmintic treatments on the Island, in addition to their insularity (ie natural limits prevent the introduction of resistant parasitic populations). The suspected resistance to fembendazole in the Janaúba region of Minas Gerais may also be related to the semi-arid climate of the region, which favors the larval mutation through ultraviolet rays, increasing their resistance. Also, animals are rarely vermifuged inappropriately, as the creations are mostly backyard without proper parasitological management.

Canever et al. (2013) conducted a study on 11 equine farms in the states of Paraná, São Paulo, Minas Gerais and Rio de Janeiro in 2009 and 2010, reporting the failure of fembendazole, pyrantel, ivermectin, moxidectin and abamectin against *Cyathostomides* on most properties. Efficacy variations were (0-61%) for fembendazole, (59-99%) pyrantel, (89-100) ivermectin and (96-100) moxidectin.

Traversa et al., (2007) studying 16 farms in Abruzzo, Lazio, Calabria and Apulia in Italy during 2005 and 2006 revealed suspicion of resistance in 27 (39.1%), 16 (23.1% %) And none (0%) of the 69 horses treated for fembendazol, pyrantel and avermectins-milbemycins, respectively. As confirmed in this study, confirmed resistance to fembendazole was found in 37.5% of the 16 farms examined and was suspected in two cases (12.5%), most of them occurring in the Lazio region. Resistance to pyrantel was found in two farms (12.5%) and in one suspect case (6.2%). Resistance to macrocyclic lactones was also observed. Microscopic examination of cultured larvae collected by the Baermann technique also showed that the genus was exclusively *Cyathostomideos*.

It is necessary that the creators are in warning about the inappropriate use of anthelmintic products, using when necessary, paying attention to the bases, and associate the control with management alternatives to slow the development of anthelmintic resistance.

Table-1. Means of egg count per gram of faeces (EPG), egg count per gram feces (FECRT) test and confidence limit (LCL) in horses wormed with ivermectin, fembendazol + trichlorfon or fembendazol on days 0 And 10 and 15

Anthelmintic group	horses	horses recruited (OPG ≥ 100)	OPG day 0	OPG day 10	RCOF day 10 (95%)	lcl day 10 (95%)
Property A						
Ivermectin	19	7	1592.86	0	100	100
Fembendazol + trichlorfon		6	1275	33.33	96.15	98.72
Property B						
Fembendazol	13	6	1658.33	83.33	89.94	99.13

**CONCLUSIONS**

Fembendazole resistance was suspected at property B. The hypothesis of resistance to ivermectin and fembendazol + trichlorfon at property A was ruled out.

**ACKNOWLEDGMENTS**

To the Foundation for Research Support of the State of Minas Gerais (FAPEMIG) for financial support.

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