

Endoparasites of horses from the Formiga city, located in center-west region of the state of Minas Gerais, Brazil

Endoparasitas de equinos provenientes do município de Formiga, localizado na região Centro-oeste do estado de Minas Gerais, Brasil

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Abstract

With the aim of studying the endoparasite fauna of horses from the Formiga city, located in center-west region of the state of Minas Gerais, 25 animals that were naturally infected with helminths were evaluated. By means of parasitological necropsies, different endoparasites were found. The subfamily Cyathostominae presented the highest incidence, followed by *Trichostrongylus axei*, *Oxyuris equi*, *Triodontophorus serratus*, *Strongyloides westeri*, *Strongylus edentatus*, *Habronema muscae*, *Parascaris equorum*, *Probstmayria vivipara*, *Strongylus vulgaris*, *Gasterophilus nasalis*, *Anoplocephala magna* and *Anoplocephala perfoliata*. In the present study, if the species *Probstmayria vivipara* was not considered in the prevalence, the frequency of Cyathostominae was equivalent to 94.85%. The results obtained in this study allowed us to detect and identify different species of helminths in horses, and confirmed the high incidence of nematodes belonging to the subfamily Cyathostominae in the center-west region of Minas Gerais.

Keywords: Endoparasites, helminth, horse, parasite.

Resumo

Com o objetivo de estudar a fauna de endoparasitas de equinos da Região Centro-Oeste do Estado de Minas Gerais, 25 animais naturalmente infectados por helmintos foram avaliados. Por meio de necropsias parasitológicas, diferentes endoparasitas foram identificados. A sub-família Cyathostominae apresentou maior incidência, seguido por *Trichostrongylus axei*, *Oxyuris equi*, *Triodontophorus serratus*, *Strongyloides westeri*, *Strongylus edentatus*, *Habronema muscae*, *Parascaris equorum*, *Probstmayria vivipara*, *Strongylus vulgaris*, *Gasterophilus nasalis*, *Anoplocephala magna* e *Anoplocephala perfoliata*. No presente estudo, se não for considerada a espécie *Probstmayria vivipara* na prevalência, a frequência de Cyathostominae é equivalente a 94,85%. Os resultados obtidos neste estudo, permitiu detectar e identificar diferentes espécies de helmintos em equinos, bem como confirmar a elevada incidência de nematódeos pertencentes à sub-família Cyathostominae na Região Centro-Oeste de Minas Gerais.

Palavras-chave: Helmintos, equinos, parasitas.

Introduction

Brazil, with 5.5 million horses, has the largest herd in Latin America and the third largest herd worldwide. The state of Minas Gerais is where the highest number of horses in Brazil are reared, with approximately 800,000 animals (IBGE, 2010).

This animal species presents a great variety of helminths, comprising several families and genera, among which the following stand out: small strongyles or Cyathostominae, large strongyles

(*Strongylus vulgaris*, *Strongylus equinus* and *Strongylus edentatus*), as well as *Parascaris equorum*, *Oxyuris equi*, *Strongyloides westeri*, *Trichostrongylus axei*, *Gasterophilus* spp, *Habronema* spp, *Dictyocaulus arnfieldi* and *Anoplocephala* spp. (MOLENTO, 2005).

The main clinical signs/symptoms of equine helminth infections are weakness, rugged hair coat, slow growth, cramps and diarrhea. These are considered to be precursors of damage to organs of the digestive tract and severe disorders in enzymatic and hormonal processes (ASSIS & ARAÚJO, 2003).

In this context, helminth infections in horses can lead to direct or indirect losses to the national economy, mainly relating

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to late growth of the animals, inferior performance of athletic horses or even the death of these animals (HERD, 1990; ASSIS & ARAÚJO, 2003).

To minimize these losses, it is of the utmost importance to have knowledge of the biology and occurrences of these endoparasites, since much greater intensity and diversity of helminths is commonly diagnosed in horses (MOLENTO, 2005).

In order to achieve this goal, studies have been conducted to evaluate the rates of helminthic infections in this animal species in different regions of the world, such as the United States (REINEMEYER et al., 1984; TOLLIVER et al., 1987), Poland (GAWOR, 1995), Australia (BUCKNELL et al., 1996), Japan (YOSHIHARA et al., 1994), South Africa (MATTHEE et al., 2000) and Brazil (BARBOSA, 1995; SOUTO MAIOR et al., 1999; RODRIGUES et al., 2000; MARTINS et al., 2001; DA SILVA ANJOS & DE LURDES A RODRIGUES, 2006).

The primary goal of the present study was to identify and quantify the endoparasites found in naturally infected horses, originating from the center-west region of the state of Minas Gerais, Brazil.

Materials and Methods

The present study was conducted between January 2012 and January 2013. In this study, the gastrointestinal contents of 25 horses that had been used as control groups (not medicated) in four anthelmintic studies conducted at the Animal Health Research Center (CPPAR) of the School of Agrarian and Veterinary Sciences (FCAV), São Paulo State University (UNESP), in Jaboticabal, São Paulo, were analyzed. Eleven animals were acquired in January 2012 and additional groups of six and eight were slaughtered in December 2012 and January 2013, respectively. Only male horses over the age of 36 months that originated from the municipality of Formiga, located in the center-west region of the state of Minas Gerais, were selected for this study. This municipality is located between the latitude: 20° 27' 52" S, longitude: 45° 25' 35" W, altitude of 841m, in an area of 1503.8 km². The municipality contains 1973 head of horses in accordance with IBGE (2010).

The animals selected had not previously been treated with any anthelmintic drug during the six months that preceded the present investigation, and presented egg counts (strongyle eggs) greater than 500 eggs per gram of feces (GORDON & WHITLOCK, 1939).

After selection, all the animals were transferred to CPPAR/FCAV/UNESP, Jaboticabal, São Paulo, where they underwent an acclimatization period of 14 days, receiving commercial food, hay and water *ad libitum*. Once this period was over, all the animals were subjected to 24 hours of fasting and were then sacrificed in accordance with the ethical procedures described in law number 11,794, of October 8, 2008, which regulates the use of animals in experiments in Brazil (BRASIL, 2008). The method used for sacrificing them was as described in the Note Guidelines on Euthanasia of the American Veterinary Medical Association (AVMA, 2007). Each animal received pre-anesthetic medication consisting of xylazine (2 g) at a dosage of 0.3 mg/kg, administered intravenously. After the animals had been sedated, sodium thiopental at a dosage of 4 mg/kg was administered intravenously. After the

animals had been confirmed to be totally unconscious (absence of palpebral and masticatory reflexes), they were bled by means of transection of the great vessels of the neck, so that their death occurred through hypovolemia.

After death, all the digestive tracts of the horses were removed, and the anatomical segments (stomach, small intestine and large intestine) were isolated and separated by means of double ligatures. These segments were then opened with the aid of an enterotome, and all the mucosa were washed with running water in order to remove all of the content and parasites present. The total content extracted from each segment was subjected to a second washing in running water and was percolated through a sieve (Tyler 48, 0.297 mm opening) for conservation of the helminth samples, which were then stored in plastic jars, with the addition of heated 10% formaldehyde at 80 °C. After all the washing procedures, the mucosa of the stomachs of each animal were subjected to digestion in a hydrochloric pepsin solution that had previously been heated to approximately 37 °C (99 °F) for a minimum of five hours (HERLICH, 1956; VICH, 2001; DUNCAN et al., 2002).

During the necropsies, all the lungs and livers were dissected and visually inspected, with the objective of determining the number of endoparasites possibly present in these organs (VICH, 2001; DUNCAN et al., 2002).

Following storage and conservation with formaldehyde, a 10% aliquot of each content was separated, after homogenization, so that endoparasites could be collected with the aid of stereomicroscopes. The helminthes were identified at genus and species level using optical microscopes (100-400x amplification), in accordance with the taxonomical criteria described by Costa (2012).

The present study was submitted to the ethics committee for animal experimentation of the Animal Health Research Institute (IPESA), Formiga, Minas Gerais, and was approved under the protocol number PD 010/2012.

Results and Discussion

From the parasitological necropsies and subsequent characterization using optical microscopy, it was possible to identify different endoparasites. The cyathostominae were the most prevalent among the necropsied horses (found in 100% of the animals), followed by *Trichostrongylus axei* (68%), *Oxyuris equi* (64%), *Triodontophorus serratus* (56%), *Strongyloides westeri* (28%), *Strongylus edentatus* (28%), *Habronema muscae* (20%), *Parascaris equorum* (20%), *Probstmayria vivipara* (16%), *Strongylus vulgaris* (12%), *Gasterophilus nasalis* (8%), *Anoplocephala magna* (8%) and *Anoplocephala perfoliata* (8%) (Table 1).

The high levels of occurrence of Cyathostominae found in the present study corroborate previous studies in which similar occurrence rates for this helminth subfamily in horses in different regions of the world were found (MATTHEE et al., 2000). This evidence is of great concern from a clinical point of view, given that the state of Minas Gerais has the largest horse herd in Brazil (IBGE, 2010) and that high rates of infection by Cyathostominae are considered to be one of the main causes of intestinal cramps in this animal species (HERD, 1990). The necropsied animals presented an average helminthic burden of 193,443.48 parasites,

not found by previous authors. This considerably diminished the mean percentage intensity of Cyathostominae diagnosed among the horses. In the present study, if *Probstmayria vivipara* is not considered in the prevalence, the frequency of Cyathostominae is equivalent to 94.85%.

Strongyloides westeri was found in the small intestines of 28% of the necropsied animals (Table 1). This was a higher proportion than what was described by Gawor (1995) in Poland and Martins et al. (2001) in Brazil (state of Rio de Janeiro), with incidences of 4%, 5% and 6.7% for this parasite, respectively. Even though this species was identified in 28% of all the animals examined, the mean intensity of infection was the fifth smallest amongst the thirteen species diagnosed in this study (Table 1).

Regarding *Parascaris equorum* infection, an occurrence rate of 20% was diagnosed (Table 1), thus corroborating the results found by Gawor (1995) and Martins et al. (2001), who observed percentages of 26% and 20%, respectively. The low parasitism by this helminth species (mean intensity of 5.48 helminths/necropsied horse) found in the present study can be correlated with the ages of the animals selected, since the infection rate due to this parasite diminishes considerably in animals over 12 months of age (BOWMAN, 2006).

In the large intestine, *Oxyuris equi* was found parasitizing 64% of the animals. This result was similar to what was described by Martins et al. (2001) (53.3%), but divergent from what was described by Bucknell et al. (1996) and Matthee et al. (2000), who found 7.0% and 29.0% respectively.

Among large strongyles, the most prevalent species was *Strongylus edentatus*, which parasitized 28% of the animals, and the least prevalent was *S. equi*, which was not diagnosed in any of the horses. The absence of this parasite differs from the results found by Matthee et al. (2000), Bucknell et al. (1996) and Gawor (1995) who, in other countries, found occurrences of 3.0%, 3.0% and 14.0% for this species, respectively. In Brazil, Souto Maior et al. (1999) and Martins et al. (2001) also found divergent results regarding the occurrence rate of this parasite: 12.1% and 36.7%, respectively. These data coincide with a study conducted by Rodrigues et al. (2000) in the state of Rio de Janeiro, in which *S. equi* was not found in any of the horses used in the research.

For *S. vulgaris*, the diagnosed occurrence rate (12% of the horses examined) was considerably lower than what was found in studies conducted in other countries (GAWOR, 1995; ENGLISH, 1979; MATTHEE et al., 2000), in which occurrence rates of

74.0%, 88.0% and 100.0%, respectively, were observed. In studies conducted in Brazil, Martins et al. (2001) and Souto Maior et al. (1999) also found occurrences that were greater than in the present study (56.7% and 84.8%, respectively). According to Cunha (1992), even though large strongyles (*S. equi*, *S. edentatus* and *S. vulgaris*) presented lower occurrence rates than other helminth species, these present greater pathogenicity to the animals and were the main agents responsible for disorders such as anemia, cramps and even death.

Nematodes of the species *Triodontophorus serratus* were found in 56% of the horses, thus differing from the results obtained by Souto Maior et al. (1999) and Rodrigues et al. (2000), who diagnosed frequencies of this genus in the large intestine of 16.5% and 10.8% of the animals, respectively. On the other hand, Martins et al. (2001) described a similar occurrence rate for *Triodontophorus serratus* in adult horses (40%).

For *Trichostrongylus axei*, an occurrence rate of 68% was diagnosed in the necropsied horses. This was the helminth species with the fifth greatest intensity (1,337.80), and is thus higher than what was found by Collobert-Laugier et al. (2000), who observed a prevalence of 12.4%. According to Briggs et al. (2004), infection by this species at low rates mostly presents asymptotically, but when these rates are high, it can lead to severe and prolonged aqueous diarrhea and consequent debilitation of the animal.

Helminths of the species *Habronema muscae* were found parasitizing 20% of the horses, thus differing from the results found by Gawor (1995), in Poland and in the United States (8%), and from the results of a study conducted in Brazil by Paiva (1988), who found an occurrence rate of 95.45% in the state of Mato Grosso do Sul. Both the adults and the larvae of this parasite may have clinical relevance. The adults can form gastric ulcers, and larvae of this parasite can be involved in cutaneous habronemiasis, thus leading to formation of a pruriginous lesion that is very difficult to treat (PAIVA, 1988).

Among the 25 horses analyzed, 24% presented six genera/species of helminths concomitantly, in different anatomical segments, followed by 20% with five genera/species present (Table 3). The maximum coinfection index diagnosed was eight genera/species of helminths parasitizing one animal.

Visual inspection of the livers and lungs, during the necropsies on the animals of the present study, did not reveal the presence of any parasites.

Table 3. Distribution of cases of endoparasites coinfections in equines from Formiga, Minas Gerais State, Brazil.

| Number of endoparasites different found in the same animal | Number of equines parasitized | Percentage (%) |
|--|-------------------------------|----------------|
| 1 | 0 | 0.00 |
| 2 | 4 | 16.00 |
| 3 | 1 | 4.00 |
| 4 | 4 | 16.00 |
| 5 | 5 | 20.00 |
| 6 | 6 | 24.00 |
| 7 | 4 | 16.00 |
| 8 | 1 | 4.00 |

Conclusions

In the present study, it was possible to detect and identify different helminths in the anatomical segments of the digestive tract of horses, with the aid of parasitological necropsies and subsequent characterization under a microscope. All the animals were derived from the Formiga city, located in center-west region of the state of Minas Gerais, thus demonstrating the high diversity and distribution of these parasites in horses, which has also been widely confirmed in the worldwide literature. Moreover, it was possible to diagnose high incidence of Cyathostominae, which are considered to be among the most important parasites in horses, from an epidemiological and clinical point of view.

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