10  ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

11  PREVALENCE OF INTESTINAL HELMINTHS AND TREATMENT OF OSTENSIVE

12  POLICING HORSES FROM THE MILITARY BRIGADE OF RIO GRANDE DO SUL,

13  BRAZIL

14

15  PREVALENCIA DE HELMINTOS INTESTINALES Y TRATAMIENTO DE CABALLOS

16  POLICÍAS OSTENSIVOS DE LA BRIGADA MILITAR DE RIO GRANDE DO SUL,

17  BRASIL

18  Sandra Márcia Tietz Marques[1*], Carolina Rigotto Murari[2] & Isabele Colla Lazzari Royes[3]

19

20  [1]  Laboratório de Helmintoses, Departamento de Patologia Clínica Veterinária,
21  Faculdade de Veterinária, UFRGS, Porto Alegre, RS, Brasil. Av. Bento Gonçalves, 9090,
22  Porto Alegre.

23  [2]  Programa de Pós-graduação FAVET, Faculdade de Veterinária, UFRGS, Porto
24  Alegre, RS, Brasil.

25  [3]  Laboratório de Reprodução Animal, Faculdade de Veterinária, UFRGS, Porto
26  Alegre, RS, Brasil.

27  *Corresponding author: E mail: santietz@gmail.com

28

29  Running title: Prevalece of helminths in horses
30  Tietz Marques *et al.*

31

32  Sandra Márcia Tietz Marques:  https://orcid.org/0000-0002-7541-9717

33  Carolina Rigotto Murari:  https://orcid.org/0009-0004-9577-7417

34  Isabele Colla Lazzari Royes:  https://orcid.org/0000-0001-9787-1213

35    ABSTRACT

36    This study describes the prevalence of intestinal helminths in 98 adult working military

37    horses of the Brazilian Sport Horse and Crioulo breeds from the Military Brigade of Rio

38    Grande do Sul, Brazil, before and after treatment with a formulation containing ivermectin

39    and praziquantel. Fecal samples were collected and sent to the laboratory of the Faculty

40    of Veterinary Medicine at the Federal University of Rio Grande do Sul. Parasitological

41    diagnosis was obtained using the eggs per gram of feces (EPG) count method and

42    coproculture for identification of infective larvae. The prevalence of parasitized horses

43    was 40.8% (40/98), with 62.5% (25/40) males and 37.5% (15/40) females. Positive

44    samples showed eggs from the Strongylidae family Baird, 1853 and, in 20%, eggs of

45    *Parascaris equorum* Goeze, 1782. After anthelmintic treatment and repeated fecal

46    examinations, 13.3% (13/98) of the tests showed Strongylidae eggs, and 86.7% (85/98)

47    were negative. Infective larvae of *Gyalocephalus capitatus* Looss, 1900, *Cylicocyclus*

48    *radiatus* (Looss, 1900) Chaves, 1930, and *Trichostrongylus axei* Looss, 1905 were

49    recovered from coproculture. The recorded efficacy was 67.5%. Parasitological

50    examinations should be performed periodically to determine parasitic load and select the

51    therapeutic protocol.

52    **Keywords**: Helminth – Horse – Intestinal nematodes – Fecal egg counts – Strongylidae

53    – Treatment

54

55    RESUMEN

56    Este estudio describe la prevalencia de helmintos intestinales en 98 caballos militares

57    de trabajo adultos de las razas Caballo Deportivo Brasileño y Criollo de la Brigada Militar

58    de Rio Grande do Sul, Brasil, antes y después del tratamiento con una formulación que

59    contiene ivermectina y praziquantel. Se recolectaron muestras fecales y se enviaron al

60    laboratorio de la Facultad de Medicina Veterinaria de la Universidad Federal de Rio

61    Grande do Sul. El diagnóstico parasitológico se obtuvo utilizando el método de recuento

62    de huevos por gramo de heces (HPG) y coprocultivo para la identificación de larvas

63    infectivas. La prevalencia de caballos parasitados fue de 40,8% (40/98), con 62,5%

64    (25/40) machos y 37,5% (15/40) hembras. Las muestras positivas mostraron huevos de

65    la familia Strongylidae Baird, 1853 y, en 20%, huevos de *Parascaris equorum* (Goeze,

66    1782). Tras el tratamiento antihelmíntico y repetidos análisis fecales, el 13,3% (13/98)

67    de las pruebas mostró huevos de Strongylidae, y el 86,7% (85/98) fueron negativos. Se

68    recuperaron larvas infectivas de *Gyalocephalus capitatus* Looss, 1900, *Cylicocyclus*

69    *radiatus* Looss, 1900, Chaves, 1930, y *Trichostrongylus axei* Looss, 1905 mediante

coprocultivo. La eficacia registrada fue del 67,5%. Se deben realizar exámenes parasitológicos periódicamente para determinar la carga parasitaria y seleccionar el protocolo terapéutico.

**Palabras clave**: Caballo – Helminto – Nematodos intestinales – Recuento de huevos fecales – Tratamiento


INTRODUCTION

Gastrointestinal parasitosis in horses have a cosmopolitan distribution and cause a wide spectrum of health effects, ranging from asymptomatic infections to sudden death. There are 29 genus and 83 species of nematodes; the majority includes 19 genus and 64 species, and belong to the family Strongylidae. The genus *Strongylus* Müller, 1780 includes three species: *S. equinus* Müller, 1780, *S. edentatus* Looss, 1900, and *S. vulgaris* Looss, 1900, which exhibit higher pathogenicity, yet their prevalence is low due to the widespread availability of anthelmintics (Kaplan & Nielsen, 2010; Marama *et al*., 2025). Ascarids and small strongyles (cyathostomins) are the most common parasites worldwide (Saeed *et al.*, 2019), with frequent infections involving 15 to 25 species and global prevalence rates of up to 100% across all age groups (Bellaw & Nielsen, 2020). Emerging cyathostomin larvae from the intestinal wall can lead to larval cyathostominosis syndrome, resulting in diarrhea, weight loss, lethargy, colic, edema, hypoproteinemia, and mortality in up to 50% of hospitalized cases. Although the relative importance of each species remains unknown, the development of species-specific DNA methods provides a means to establish the pathogenicity of different species (Ghafar *et al.*, 2023).

In Brazil, prevalence rates are significant, with regional variations ranging from 60% to 100% (Molento *et al.*, 2024), and critical records are present in the states of Rio Grande do Sul (Piccoli *et al.*, 2015; Lignon *et al.*, 2020; Menetrier *et al.*, 2020; Marques *et al.*, 2025), Paraná (Godéski & Pedrassani, 2018), Minas Gerais (Barbosa *et al.*, 2018), and Amazonas (Ferreira *et al.*, 2024). Recent studies in Australia (Abbas *et al.*, 2021; Abbas

97   *et al.*, 2023; Saeed *et al.*, 2019), Germany (Boelow *et al.*, 2023), Denmark (Beasley *et*

98   *al.*, 2020), the United States (Nielsen *et al.*, 2024; Nielsen *et al.*, 2025), Mexico (Romero

99   *et al.*, 2020), Romania (Bulgaru *et al.*, 2021), Nigeria (Alaba *et al.*, 2022; Ogben *et al.*,

100  2022), Ethiopia (Marama *et al.*, 2025), and Peru (Almeyda & Porras, 2019) have reported

101  prevalence rates oscillating from 46% to 93%.

102  Anthelmintic drugs are widely used to treat nematodes in equines; however, resistance

103  to nearly all of them is attributed to continuous and indiscriminate use, representing a

104  challenge for their control (Nielsen *et al.*, 2018; Abbas *et al.*, 2021). Alternative programs,

105  including treatment strategies based on egg shedding levels and epidemiological factors,

106  have been proposed to address anthelmintic resistance (Kaplan & Nielsen, 2010; Saeed

107  *et al.*, 2019). Usually, 10% to 30% of horses contribute 80% of the eggs deposited on

108  pastures, known as high egg excretors and their treatment can help reduce overall

109  anthelmintic use, allowing most horses to avoid frequent treatments and keep parasite

110  refugia (nematode populations not exposed to anthelmintics) (Nielsen *et al.*, 2024).

111  Although several diagnostic tests are used for fecal egg counts, there is no consensus

112  on their methodologies, and the modified McMaster technique (Gordon & Whitlock

113  method or fecal egg count) remains the most widely method used in equine parasitology

114  (Ghafar *et al.*, 2021). This study aimed to collect fecal samples to measure helminth

115  parasitic loads before and after broad-spectrum anthelmintic treatment, dosed quarterly

116  according to the military unit's protocol, in working military horses from the Military

117  Brigade in Porto Alegre, Rio Grande do Sul, Brazil.

118

119  MATERIAL AND METHODS

120  A total of 98 horses, 60 males and 38 females, all adults, of the Brazilian Sport Horse

121  and Crioulo breeds, belonging to the 4th RPMon – Regimento Bento Gonçalves of the

122    Military Brigade in the state of Rio Grande do Sul, Brazil were subjected to parasitological

123    examinations. The stool samples were collected from the rectal ampulla of 20 animals

124    per week by military handlers and sent to the Helminthology Laboratory at the Faculty of

125    Veterinary Medicine, Federal University of Rio Grande do Sul. Coprological diagnosis

126    was made using the modified Gordon and Whitlock method (eggs per gram of feces -

127    EPG); in the laboratory, the samples were kept refrigerated at 4°C and analyzed within

128    24 hours (Hoffmann, 1987). The five EPG ranges were: (1.) Zero; (2.) 100-400; (3.) 500–

129    800; (4.) 800-1000; (5.) above 1100, according to Nielsen *et al.* (2019).

130    Coprocultures were performed on 14 fecal samples, corresponding to EPG above 500.

131    The identification of infective larvae from coprocultures, conducted before and after

132    treatment, enabled the identification of $L_3$ larvae with morphological characteristics of the

133    Subfamily Cyathostominae, based on observation of the number and shape of intestinal

134    cells, total larva length, presence or absence of sheath, and tail sheath aspect (Madeira

135    de Carvalho *et al.*, 2004; Madeira de Carvalho *et al.*, 2008). To evaluate the product's

136    efficacy, the fecal egg count reduction test (FECRT) was performed, using the formula

137    by Barbosa *et al.* (2018): Efficacy (%) = (EPG (pre-treatment) − EPG (post-treatment) /

138    EPG (pre-treatment) × 100.

139

140    **Ethic aspects**: For this study formal consent is not required.

141

142    RESULTS AND DISCUSSION

143    In the pre-treatment coprological analysis, the overall prevalence of parasitized horses

144    by the EPG method was 40.8% (40/98), with 62.5% (25/40) and 37.5% (15/40) for males

145    and females, respectively. The positive samples showed eggs from the Strongylidae

146    family, and in 20%, eggs of *P. equorum* Goeze, 1782. After anthelmintic treatment and

147  repetition of fecal exams, 13.3% (13/98) of the exams showed Strongylidae eggs, and

148  86.7% (85/95) exhibited negative results. Infective larvae of *Gyalocephalus*

149  *capitatus* Looss, 1900, *Cylicocyclus radiatus* Looss, 1900 Chaves, 1930,

150  and *Trichostrongylus axei* Looss, 1905 were recovered from the coproculture, in similar

151  proportions (30-35%). After deworming, no *P. equorum* eggs were detected in the

152  coprological exams.

153  The anthelmintic treatment performed on all animals was the formulation with 1.55%

154  ivermectin and 7.75% praziquantel, according to the manufacturer's recommendation.

155  Two weeks after the application of the anthelmintic, new collections and fecal exams

156  were performed, resulting in 13.3% (13/98) of horses positive and 86.7% (85/95) of

157  animals negative. The results are shown in Table 1.

158  **Table 1**. Parasitological examination results (PG) of military working horses from the 4th

159  RPMon – Bento Gonçalves Regiment of the Military Brigade (RS), Brazil, before and

160  after treatment with an ivermectin- and praziquantel-based anthelmintic.

| EPG values | Before treatment (%) | After treatment (%) |
| --- | --- | --- |
| Zero | 58 (59.18) | 85 (86.73) |
| 100-400 | 26 (26.53) | 9 (9.18) |
| 500-800 | 7 (7.14) | 3 (3.06) |
| 900-1000 | 2 (2.04) | 0 (0) |
| ≥ 1100 | 5 (5.10) | 1 (1.02) |

161

162  The average efficacy recorded was 67.5%. The *in vivo* test has been used for many

163  years, mainly based on procedures described in the guidelines of the World Association

164  for the Advancement of Veterinary Parasitology (WAAVP) published by Coles *et al.*

165  (1992), whose test is based on fecal egg counts (EPG) before and after treatment in the

same animals. Anthelmintic efficacy is estimated by comparing egg counts in feces before and after treatment, with the time defined according to the tested group. The evaluation of this equine population was conducted two weeks post-treatment. The reduction test, according to WAAVP recommendations (Coles *et al.*, 1992), is considered the method of choice for monitoring anthelmintic efficacy due to its easy execution and interpretation, being performed with a sequence of fecal egg count exams. Regarding the calculation of EPG counts, it is a phenotypic test whose count directly depends on the host effect and is considered indirect, as it reflects the egg-laying by females, which depends on the host's immune response (Fortes & Molento, 2013). Furthermore, the use of arithmetic means of egg counts in the feces of the same animals before and after anthelmintic administration, instead of randomly, can provide more reliable results (Dobson *et al.*, 2009). The test evaluation can be impaired due to variations in the correlation between fecal egg counts and adult parasite burden among different parasite species. Anthelmintic resistance is widely common in small strongyles, and recent studies have documented the increasing incidence of resistance to the macrocyclic lactone class of drugs (Barbosa *et al.*, 2018; Abbas *et al.*, 2021).

Several European countries have implemented legal restrictions on the use of anthelmintics, such as prescription-only use, aiming to reduce the intensity of anthelmintic treatment and decrease the selection pressure for resistance; however, the long-term results of this approach have not been fully evaluated (Nielsen *et al.*, 2018; Nielsen, 2022). The sanitary management of working military horses is rigorous, with quarterly deworming supervised by career military veterinarians. The acquisition of medications and supplies follows the official supply protocol, ensuring predictability for both the herd and the police personnel. Therefore, routine health evaluations are essential for logistical planning of new products and materials.

According to Aromaa *et al.* (2018), different types of hygienic management affect each parasite differently: pasture management is critical for strongylids, while hygiene of stalls/facilities is more relevant for *P. equorum*, highlighting the need for customized control strategies. However, Almeyda & Porras (2019) investigated gastrointestinal parasite rates in 384 Thoroughbred horses in the province of Chincha, Peru, and found a prevalence of *Strongylus* spp. of 74.22%, identifying that animal age, humidity, and rainy seasons (typical climatic conditions in the region) were factors associated with this prevalence. Romero *et al.* (2020) investigated horses from different regions in central Mexico with a prevalence of 47.24%, with *Strongylus* spp. being the most prevalent (23.85%), followed by *Trichostrongylus* spp. (21.56%), and concluded that breed and place of origin showed a significant association with helminth infection.

Alaba *et al.* (2022) investigated the prevalence, severity, and predisposing factors (age, sex, and breed) of gastrointestinal parasites in 56 polo horses in Ibadan, Nigeria, and demonstrated a prevalence of 92.9%, dominated by *Strongylus* spp. (89.3%), showing no association between EPG prevalence and the age, sex, and breed of the horses. However, there was a higher occurrence and more severe infections in adult horses, females, and exotic breeds. Meanwhile, Ogbein *et al.* (2022) tested fecal samples from 108 horses in Jos (Nigeria), consisting of 25 males and 83 females, over a six-month period covering three months in the dry season and three months in the rainy season, with an overall prevalence of 82.41%, of which 51.85% showed single infections and 30.56% mixed infections. The most relevant gastrointestinal parasites in this study were *Strongylus* spp., *Strongyloides westeri* Ihle, 1917, *Trichonema* spp. K. Möbius, 1888, *P. equorum*, and *Triodontophorus* spp. Looss 1900. The fecal egg count (EPG) prevalence was zero (3.70%), low (46.30%), medium (24.07%), and high (9.26%), and had a higher expression in young animals, during the rainy season, and in animals with poor body condition scores (84.62%). For these authors, under the rearing conditions of the horses,

217 different factors such as inadequate nutrition, incorrect management practices, health

218 status, and other physiological factors can affect the horses' immune system, making

219 them more susceptible to gastrointestinal parasitic infections. In the context of the Military

220 Brigade, the health of the horses is the foundation of operational efficiency. Unlike leisure

221 horses, policing horses face a workload that demands perfect physical and metabolic

222 condition, where any parasitosis can compromise the performance and safety of the

223 man-horse duo. The facilities for the horses and the vehicles that transport them to the

224 working locations have to be qualified to preserve health and avoid stress during

225 displacements.

226 In Germany, Boelow *et al.* (2023) evaluated the prevalence and potential severity of

227 nematodiasis through fecal exams of 1,067 horses on German farms, with 46.5% for

228 strongylids. Marama *et al.* (2025) described the strong dependence on anthelmintics for

229 parasitic control in horses in Ethiopia, with growing concern about drug resistance, and

230 therefore conducted a study to estimate helminth prevalence, assess risk factors, and

231 evaluate anthelmintic efficacy. Standard parasitological methods were applied to 382

232 samples to detect parasite eggs or larvae, allocated into three treatment groups:

233 fenbendazole, ivermectin, and an untreated control group. Fecal egg counts were

234 performed on the day of treatment and 14 days later. The overall prevalence of

235 gastrointestinal helminths was 72%, and the identified parasites included *Strongylus*

236 species (63.87%), *S. westeri* (4.71%), *O. equi* (4.45%), *P. equorum* (5.5%), and most

237 animals presented mild (51.0%) or moderate (38.2%) infections, while 10.7% had severe

238 infections. The anthelmintic efficacy study demonstrated resistance to fenbendazole in

239 horses, while parasites in donkeys remained susceptible to both fenbendazole and

240 ivermectin, reinforcing the importance of gastrointestinal parasite control in equines and

241 the judicious use of anthelmintics to control resistance and maintain effective parasitic

242 control.

243    Piccoli *et al.* (2015) evaluated the occurrence of intestinal helminths in working horses

244    (N=131) used in carts for recyclable waste collection and compared them with leisure

245    horses (N=145) raised in Porto Alegre, Rio Grande do Sul. Positivity was 73% (202/276),

246    with 64.8% (94/145) and 82.4% (108/131) for leisure and working horses, respectively,

247    with a higher frequency of eggs from the Strongylidae family and low occurrence of *P.*

248    *equorum*, *S. westeri*, and *Anoplocephala* spp. Menetrier *et al.* (2020) recorded 94%

249    (64/68) gastrointestinal infection in horses treated at the university hospital in Porto

250    Alegre, Rio Grande do Sul, with predominance of Strongyloidea eggs (78%), *P. equorum*

251    (7.81%), and *O. equi* (9.4%). The study by Godéski & Pedrassani (2018) reinforces an

252    important pattern in equine parasitology: ubiquity versus intensity. Even with 100%

253    prevalence (N=35), the average EPG of 102 is low, suggesting a balance between the

254    horses' immune system and herd management. There was 100% predominance of

255    nematodes from the order Strongylida and 71.4% by *P. equorum* in animals up to 2

256    years; for horses aged 3 to 15 years, 96.4% presented parasitism by nematodes from

257    the order Strongylida. The hatched larvae from the coprocultures of the eggs identified

258    small strongyles. A study conducted by Lignon *et al.* (2020) in draft horses in the city of

259    Pelotas, southern Rio Grande do Sul, recorded a rate of 90.2% (74/82) for some

260    helminth, presenting an average count of 739.2 EPG, with higher prevalence of infections

261    by parasites from the Strongylidae family (74.3%). In northern Brazil, Ferreira *et al.*

262    (2024) studied horses raised on farms in the state of Pará. The result, through EPG,

263    revealed 66.67% (20/30) eggs of parasites from the Strongylidae family.

264    Routine diagnostic methods can be evaluated for parameters such as sensitivity,

265    specificity, accuracy, and precision. There are numerous variables that can affect

266    accuracy in egg counting, including biological ones such as: uneven distribution of

267    parasite eggs in a fecal sample; composition of parasite species within the animal, and

268    the number of internal parasites present, which can affect egg production (Denwood *et*

269    *al*., 2023; Kaplan & Vidyashankar, 2012). Modern anthelmintic treatment programs in

270    equines in North America use surveillance-based methods, in which only horses that

271    contribute the highest number of parasite eggs to the pasture, and consequently increase

272    infection pressure, are treated with an anthelmintic drug (Nielsen *et al.*, 2019; Rendle *et*

273    *al.*, 2019). The American Association of Equine Practitioners currently recommends two

274    fecal egg count methods to determine the number of strongylid parasite eggs: a modified

275    McMaster technique and a modified Wisconsin technique. Both techniques require

276    specialized equipment, including a flotation solution with density, a microscope, and a

277    specialized counting slide or microscope slides (Nielsen *et al.*, 2019); however, these

278    routine methods are widely used, and their cost is well-supported by breeders.

279    The control of endoparasites is one of the main challenges of equine health management

280    and requires constant vigilance, based on diagnosis and monitoring of therapy and

281    resistance to anthelmintics. The reviewed studies demonstrate that the issue is global,

282    with reports on different continents involving several drug classes, which highlights the

283    extent and severity of the phenomenon. Comparative analyses of international guidelines

284    show that there is no absolute consensus on surveillance and control protocols, with

285    regional differences prevailing related to the importance attributed to certain species and

286    the availability of diagnostic resources, which reinforces the complexity of the scenario.

287    In addition to scientific effort and technological innovations, addressing anthelmintic

288    resistance in equines also depends on consistent public policies and extension programs

289    that guide producers and veterinarians (Bulgaru *et al.*, 2021).

290    The development of new drugs is extremely expensive and is expected to be more

291    expensive than older drugs. Therefore, it seems clear that the "global deworming"

292    approach that has consolidated over the last 40 to 50 years needs to change, and animal

293    producers need to develop a new vision for parasite control and production sustainability

294    (Bellaw & Nielsen, 2020; Ghafar *et al.*, 2023). Furthermore, parasitologists must improve

295 the study design and data analysis methods used for diagnosing anthelmintic resistance,

296 especially for the fecal egg count reduction test. One critical point is the lack of a standard

297 for comparing data between different studies (Ghafar *et al*., 2021). The view of Kaplan &

298 Vidyashankar (2012) highlights that we cannot just "wait for a new drug," as resistance

299 tends to evolve faster than pharmaceutical innovation. It is necessary to adopt molecular

300 methods that are more sensitive and cost-effective to become routine and early in

301 identifying important parasites.

302 MacDonald *et al.* (2023) reviewed data on the Efficacy Reduction Period (ERP) in

303 cyathostomins against the three main classes of anthelmintics: macrocyclic lactones,

304 tetrahydropyrimidines, and benzimidazoles in 54 studies published over three decades

305 (1972-2022). Until early 2022, there was no consensual definition for ERP, with eight

306 definitions identified in the literature, which makes it difficult to compare the studies. It is

307 not clear whether the reduction of ERP of these anthelmintics to such levels is due to the

308 development of anthelmintic resistance (AR) or biological factors related to horses,

309 cyathostomin species, and/or the environment. The ERP for other anthelmintics

310 (fenbendazole and pyrantel) was often not reported due to already established

311 resistance against these drugs. Nielsen *et al.* (2025) reported increasing incidence of

312 resistance to the macrocyclic lactone class of drugs in cyathostomin parasites. Several

313 European countries have implemented legal restrictions on the use of anthelmintics,

314 allowing only under medical prescription, to reduce treatment intensity and decrease

315 selective pressure for drug resistance, however, the long-term results of this approach

316 have not been comprehensively evaluated. In Danish horses parasitized by *S. vulgaris*,

317 it was selected 299 horses from 30 herds and four collaborating veterinary clinics. All

318 horses with fecal egg counts were treated with ivermectin. The efficacy of ivermectin and

319 the effective remission period were determined following current guidelines.

320 Coproculture and PCR were used for the detection of *S. vulgaris*. Fecal egg count

321 reduction tests based on egg counts by the McMaster method suggested inconclusive
322 efficacy of ivermectin in two equine operations and total efficacy in all other populations,
323 while the automated system suggested resistance to ivermectin in six operations and
324 inconclusive results in another 8. The ERP of ivermectin was determined at least 8 weeks
325 with both methods in all cases. The prevalence of *S. vulgaris* was 2.7% and 5.7% with
326 coproculture and PCR, respectively, and all samples were negative with both methods
327 at 8 and 24 weeks after treatment. Overall, ivermectin efficacy was high, although some
328 results suggest that a reduction in efficacy may be occurring, justifying deeper
329 monitoring. ERP estimates exceeded 8 weeks, suggesting no reduction. The two egg
330 counting techniques showed general agreement, but the automated system detected
331 more positive results at low egg count levels, leading to lower efficacy estimates in some
332 populations.

333 Advances in equine parasite diagnosis involve molecular techniques that facilitate a
334 deeper understanding of the epidemiology of different parasite species in a given horse
335 population. Next-generation sequencing of target DNA regions has revolutionized the
336 identification of gastrointestinal nematodes in production animals, potentially providing
337 the relative abundance of prevalent species (Ghafar *et al.*, 2023). The use of ITS-2 region
338 sequencing to characterize the nemabiome represents the most modern in parasitology;
339 while EPG reveals how much infection exists, Nielsen (2022)'s work with ITS-2 amplicon
340 sequencing identifies nematode species that the common microscope cannot
341 differentiate. However, these advances in molecular biology are well-suited to unravel
342 the helminthic fauna, but are unfeasible in laboratory conditions due to the high cost of
343 equipment and necessary infrastructure, in addition to the high cost of parasitological
344 exams for equine breeders. The breeder needs to know the parasitic burden of their herd
345 and which treatment to perform, having difficulty affording high-cost exams. This
346 precision veterinary medicine is still not accessible in most countries.

347    The most prevalent parasites were strongylids. The prevalence of negative animals

348    reached 86.73% of the herd. There was no detection of *P. equorum* post-treatment.

349    Infective larvae of *Gyalocephalus capitatus*, *Cylicocyclus radiatus*, and *Trichostrongylus*

350    *axei* were recovered from the coproculture.

351

352    **Author contributions: CRediT (Contributor Roles Taxonomy)**

353    **SMTM** = Sandra Márcia Tietz Marques

354    **CRM** = Carolina Rigotto Murari

355    **ICLR** = Isabele Colla Lazzari Royes

356    **Conceptualization:** SMTM

357    **Data curation:** SMTM

358    **Formal Analysis:** SMTM, CRM, ICLR

359    **Funding acquisition:** SMTM

360    **Investigation:** SMTM, CRM, ICLR

361    **Methodology:** SMTM, CRM, ICLR

362    **Project administration:** SMTM

363    **Resources:** SMTM, CRM

364    **Software:** SMTM, CRM, ICLR

365    **Supervision:** SMTM, ICLR

366    **Validation:** SMTM, CRM, ICLR

367    **Visualization:** SMTM, CRM, ICLR

368 **Writing – original draft:** SMTM, CRM, ICLR

369 **Writing – review & editing:** SMTM, CRM, ICLR

370

371 BIBLIOGRAPHIC REFERENCES

372 Abbas, G., Ghafar, A., Hurley J., Bauquier J., Beasley A., Wilkes E.J.A., Jacobson C.,

373     El-Hage C., Cudmore L., Carrigan P., Tennent-Brown B., Gauci C.G., Nielsen M.K.,

374     Hughes K.J., Beveridge I., & Jabbar A. (2021). Cyathostomin resistance to

375     moxidectin and combinations of anthelmintics in Australian horses. *Parasites &*

376     *Vectors*, *14*, 597.

377 Abbas, G., Ghafar, A., Bauquier, J., Beasley, A., Ling, E., Gauci, C.G., El-Hage, C.,

378     Wilkes, E.J.A., McConnell, E., Carrigan, P., Cudmore, L., Hurley, J., Beveridge, I.,

379     Nielsen, M.K., Stevenson, M.A., Jacobson, C., Hughes, K.J., & Jabbar, A.

380     (2023). Prevalence and diversity of ascarid and strongylid nematodes in Australian

381     Thoroughbred horses using next-generation sequencing and bioinformatic

382     tools. *Veterinary Parasitology*, *323*, 110048.

383 Alaba, B., Olajide, E., Omotosho, O., & Okemiri, D. (2022). Prevalence, severity and

384     predisposing factors of gastrointestinal parasite Infection in polo horses in Ibadan,

385     Nigeria. *Journal of Applied Veterinary Sciences, 7*, 80-85.

386 Almeyda, M.D., & Porras, E.G. (2019). Gastrointestinal parasitism in purebred equines

387     in Chincha, Ica-Peru 2016. *Brazilian Journal of Health Review*, *2*, 3086–3090.

388 Aromaa, M., Hautala, K., Oksanen, A., Sukura, A., & Náreaho, A. (2018). Parasite

389     infections and their risk factors in foals and young horses in Finland. *Veterinary*

390     *Parasitology: Regional Studies and Reports*, *12*, 35-38.

391 Barbosa, F.C., Oliveira, W.J., Costa, P.C., & Mundim, A.V. (2018). Eficácia anti-
392     helmíntica da ivermectina em equinos: exames coproparasitológicos e
393     hematológicos. *Ciência Animal Brasileira*, *19*, 1-12.

394 Beasley, A.M., Kotze, A.C., Barnes, T., & Coleman, G.T. (2020). Equine helminth
395     prevalence and management practices on Australian properties as shown by
396     coprological survey and written questionnaire. *Animal Production Science*, *60*,
397     2069–2079.

398 Bellaw, J.l., & Nielsen, M.K. (2020). Meta-analysis of cyathostomin species-specific
399     prevalence and relative abundance in domestic horses from 1975-2020: emphasis
400     on geographical region and specimen collection method. *Parasites & Vectors*, *13*,
401     509.

402 Boelow, H., Krücken, J., & von Samson-Himmelstjerna, G. (2023). Epidemiological study
403     on factors influencing the occurrence of helminth eggs in horses in Germany based
404     on sent-in diagnostic samples. *Parasitology Research, 122*, 749-767.

405 Bulgaru, A., Lupu, D., Dinu, H., Negru, E., & Danes, M. (2021). Parasitological
406     investigations in an arabian horse breeding farm in romania. *Scientific Works. Series
407     C. Veterinary Medicine, 67*, 28-33.

408 Coles, G.C., Bauer, C., Borgsteede, F.H., Geerts, S., Klei, T.R., Taylor, M.A., & Waller,
409     P.J. (1992). World Association for the Advancement of Veterinary Parasitology
410     (W.A.A.V.P.) methods for the detection of anthelmintic resistance in nematodes of
411     veterinary importance. *Veterinary Parasitology*, *44*, 35-44.

412 Denwood, M.J., Kaplan, R.M., McKendrick, I.J., Thamsborg, S.M., Nielsen, M.K., &
413     Levecke, B. (2023). A statistical framework for calculating prospective sample sizes
414     and classifying efficacy results for faecal egg count reduction tests in ruminants,
415     horses and swine. *Veterinary Parasitology*, *314*, 109867.

416    Dobson, R.J., Sangster, N.C., Besier, R.B., & Woodgate, R.G. (2009). Geometric means

417       provide a biased efficacy result when conducting a faecal egg count reduction test

418       (FECRT). *Veterinary Parasitology*, *161*, 162-167.

419    Ferreira, S.M., Macedo, J.J.P., Seade, G.C.C., Camargo Júnior, R.N.C., & Silva, W.C.

420       (2024). Pesquisa de parasitas gastrointestinais em equinos do Baixo Amazonas,

421       Amazônia Oriental, Brasil. *Revista Colombiana de Ciência Animal*, *16*, e1022.

422    Fortes, F.S., & Molento, M.B. (2013). Resistência anti-helmíntica em nematoides

423       gastrintestinais de pequenos ruminantes: avanços e limitações para seu

424       diagnóstico. *Pesquisa Veterinária Brasileira*, *33*, 1391-1402.

425    Ghafar, A., Abbas, G., King, J., Jacobson, C., Hughes, K.J., El-Hage, C., Beasley, A.M.,

426       Bauquier, J., Wilkes, E.J.A., Hurley, J., Cudmore, L., Carrigan, P., Tennent-Brown,

427       B., Nielsen, M.K., Gauci, C., Beveridge, I., & Jabbar, A. (2021). Comparative studies

428       on faecal egg counting techniques used for the detection of gastrointestinal parasites

429       of equines: A systematic review. *Current Research in Parasitology & Vector-borne*

430       *Diseases, 1*, 100046.

431    Ghafar, A., Abbas, G., Beasley, A., Bauquier, J., Wilkes, E.J.A., Jacobson, C.,

432       McConnell, E., El-Hage, C., Carrigan, P., Cudmore, L., Tennent-Brown, B., Hurley,

433       J., Nielsen, M.K., Gauci, C.G., Beveridge, I., Hughes, K.J., & Jabbar, A. (2023).

434       Molecular diagnostics for gastrointestinal helminths in equids: Past, present and

435       future. *Veterinary Parasitology*, *313*, 109851.

436    Godéski, A., & Pedrassani, D. (2018). Helmintos em equinos de cabanha da cidade de

437       São José dos Pinhais - PR. *Saúde e Meio Ambiente*, *7*, 22-30.

438    Hoffmann, R.P. (1987). *Diagnóstico de parasitismo veterinário*. Sulina editora, 156 pp.

439    Kaplan, R.M., & Nielsen, M.K. (2010). An evidence-based approach toe quine parasite

440        control: It ain't the 60s anymore. *Equine Veterinary Education*, *22*, 306–316.

441    Kaplan, R.M., & Vidyashankar, N.A. (2012). An inconvenient truth: global worming and

442        anthelmintic resistance. *Veterinay Parasitology*, *186*, 70-78.

443    Lignon, J.S., Martins, N.S., Mueller, A., Siegert, F., De Leão, M.S., Camassola, J.L.T.,

444        Pellegrin, T.G., Antunes, T.A., Pappen, F.G., & Pinto, D.M. (2020). Prevalência de

445        nematódeos intestinais em equinos de tração na cidade de Pelotas/RS, Brasil.

446        *Veterinária e Zootecnia*, *27*, 1-6.

447    MacDonald, S.L., Abbas, G., Ghafar, A., Gauci, C.G., Bauquier, J., El-Hage, C., Tennent-

448        Brown. B., Wilkes, E.J.A., Beasley, A., Jacobson, C., Cudmore, L., Carrigan, P.,

449        Hurley, J., Beveridge, I., Hughes, K.J., Nielsen, M.K., & Jabbar, A. (2023). Egg

450        reappearance periods of anthelmintics against equine cyathostomins: The state of

451        play revisited. *International Journal for Parasitology: Drugs and Drug Resistance*, *21*,

452        28-39.

453    Madeira de Carvalho, L.M., Fazendeiro, M.I., & Afonso-Roque, M.M. (2008). Estudo

454        morfométrico das larvas infectantes (L$_3$) dos estrongilídeos (Nematoda:

455        Strongylidae) dos equídeos. 4. Estudo das populações de ciatostomíneos de

456        equídeos bravios e domésticos através do método de análise dos morfotipos de L3

457        de *Cyathostomum* sensu latum. *Acta Parasitológica Portuguesa*, *15*, 65-70.

458    Madeira de Carvalho, L.M., Fazendeiro, M.I., & Afonso-Roque M.M. (2004). Estudo

459        morfométrico das larvas infectantes (L3) dos estrongilídeos (Nematoda:

460        Strongylidae) dos equídeos - 1. Género *Cyathostomum* s.l. *Acta Parasitológica*

461        *Portuguesa*, *11*, 23-32.

462    Marama, A., Terefe, G., Waktole, H., Ashenafi, H., Ayana, D., & Megersa, B. (2025).

463        Occurrence of gastrointestinal helminthiasis and efficacy of anthelmintics in equines

464    in Shashemene and Asella districts, Oromia, Ethiopia. Ethiopia. *Discover Animals*,

465    *2*, 1-17.

466    Marques, S.M.T., Royes, I.C.L., Tondin, M., Dutra, B.C.M., & Peitz, L. (2025).

467    Estrongilídeos (Nematoda: Strongylidae) em cavalos segregados em piquetes e

468    sem tratamento anti-helmíntico. *Revista Agrária Acadêmica*, *8*, 41-52.

469    Menetrier, L.C., Marques, S.M.T., & De Mattos, M.J.T. (2020). Multiparasitismo em

470    cavalos provenientes de pequenas propriedades na cidade de Porto Alegre/RS -

471    relato de caso. *Revista Agrária Acadêmica*, *3*, 14-24.

472    Molento, M.B., Pires, L.S.A., Dall'Anese, J., Yoshitani, U.Y., & Almeida, T. (2024).

473    Prevalence and risk factors of gastrointestinal helminths infection in Brazilian horses:

474    A retrospective study of a 12-year (2008–2019) diagnostic data. *Research in*

475    *Veterinary Science*, *173*, 105272.

476    Nielsen, M.K. (2022). Anthelmintic resistance in equine nematodes: current status and

477    emerging trends. *International Journal for Parasitology: Drugs and Drug Resistance*,

478    *20*, 76-88.

479    Nielsen, M.K., Branan, M.A., Wiedenheft, A.M., Diganantonio, R., Garber, L.P., Kopral,

480    C.A., Phillippi-Taylor, A.M., & Traub-Dargatz, J.L. (2018). Parasite control strategies

481    used by equine owners in the United States: a national survey. *Veterinary*

482    *Parasitology*, *250*, 45-51.

483    Nielsen, M.K., Sauermann, C.W., & Leathwick, D.M. (2019). The effect of climate,

484    season, and treatment intensity on anthelmintic resistance in cyathostomins: A

485    modelling exercise. *Veterinary Parasitology*, *269*, 7–12.

486 Nielsen, M.K., Slusarewicz, P., Kuzmina, T.A., & Denwood, M.J. (2024). US-wide equine
487      strongylid egg count data demonstrate seasonal and regional trends. *Parasitology*,
488      *151*, 579–586.

489 Nielsen, M.K., Bartholdy, I.D., Kristensen, K.S., Borrye, J.C., Meilvang, K.S.S., Rendtorff,
490      C.E.K., Hjortflod, M.D., Fuglbjerg, V., Karlsson, M., Petersen, H.H., Toft, K.,
491      Thamsborg, S.M., & Pihl, T.H. (2025). Ivermectin performance against equine
492      strongylids: Efficacy, egg reappearance periods, and fecal egg counting method
493      comparison. *Veterinary Parasitology*, *336*, 110465.

494 Ogbein, K.E., Dogo, A.G., Oshadu, D.O., & Edeh, E.R. (2022). Gastrointestinal parasites
495      of horses and their socio-economic impact in Jos Plateau - Nigeria. *Applied*
496      *Veterinary Research, 1*, e2022010.

497 Piccoli, C., Marques, S.M.T., Appel, G., Silveira, G.B., Loos, D.E., & Mattos, M.J.T.
498      (2015). Helmintos intestinais em cavalos de trabalho e de lazer de Porto Alegre/RS.
499      *Science and Animal Health*, *3*, 56-64.

500 Romero, C., Heredia, R., Miranda, L., & Arredondo, M. (2020). Prevalence of
501      gastrointestinal parasites in horses of Central Mexico. *Open Journal of Veterinary*
502      *Medicine*, *10*, 117-125.

503 Saeed, M.A., Beveridge, I., Abbas, G., Beasley, A., Bauquier, J., Wilkes, E., Jacobson,
504      C., Hughes, K.J., El-Hage, C., O'Handley, R., Hurley, J., Cudmore, L., Carrigan, P.,
505      Walter, L., Tennent-Brown, B., Nielsen, M.K., & Jabbar, A. (2019). Systematic review
506      of gastrointestinal nematodes of horses from Australia. *Parasites & Vectors*, *12*, 188.

507 Rendle, D., Austin, C., Bowen, M., Cameron, I., Furtado, T., Hodgkinson, J., McGorum,
508      B., & Matthews, J.B. (2019). Equine de-worming: a consensus on current best
509      practice. *UK-Vet Equine*, *3*, 1-14.