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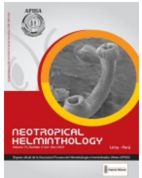

ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

HELMINTH COMMUNITY OF BACKYARD CHICKENS (*GALLUS GALLUS DOMESTICUS* LINNAEUS, 1758) IN SEROPÉDICA, RIO DE JANEIRO, BRAZIL

COMUNIDADE DE HELMINTOS DE FRANGOS DE QUINTAL *(GALLUS GALLUS DOMESTICUS LINNAEUS, 1758)* EM SEROPÉDICA, RIO DE JANEIRO, BRASIL

COMUNIDAD DE HELMINTOS DE POLLOS CASEROS (*GALLUS GALLUS DOMESTICUS* LINNAEUS, 1758) EN SEROPÉDICA, RIO DE JANEIRO, BRASIL

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ABSTRACT

The domestic chicken, *Gallus gallus domesticus* Linnaeus, 1758, is an important component of the agribusiness segment, and Brazil is one of the world's largest broiler producers and exporters. The present study aimed to characterize the composition and structure of the helminth community of backyard chickens, *G. g. domesticus*, in Seropédica, Rio de Janeiro, Brazil. Fifty-five adult chickens were studied. The overall helminth species richness was 12. The nematodes *Capillaria* sp. and *Heterakis gallinarum* (Schrank, 1788), recovered from small intestine and cecum, respectively, presented the highest prevalence and mean abundance. In addition, these species presented the highest values of frequency of dominance. The helminth species *Amoebotaenia cuneata* (von Linstow, 1872) – *Raillietina tetragona* (Molin, 1958); and *Davainea proglottina* (Davaine, 1860) – *A. cuneata*; and *H. gallinarum* – *Capillaria* sp. showed significant positive correlation between their abundance and prevalence. *Gongylonema ingluvicola* Ransom, 1904 and *H. gallinarum* showed significant correlation between host sex and helminth abundance, while there was no correlation between host sex and helminth prevalence. The knowledge of helminth community structure in free-range chickens is important to adopt better measures for control and prevention of helminth infections.

Keywords: birds – Cestoda – Nematoda – parasite ecology – prevalence – parasite richness

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RESUMO

As galinhas domésticas *Gallus gallus domesticus* Linnaeus, 1758, são um importante componente do segmento do agronegócio, e o Brasil é um dos maiores produtores e exportadores de frangos de corte. O presente estudo teve como objetivo caracterizar a estrutura da comunidade componente de helmintos de galinhas de fundo de quintal, *G. g. domesticu*s, em Seropédica, Rio de Janeiro, Brasil. Foram estudados 55 frangos adultos. A riqueza total de helmintos foi de 12 espécies. Os nematoides *Capillaria* sp. e *Heterakis gallinarum* (Schrank, 1788), encontrados no intestino delgado e ceco, respectivamente, foram as espécies mais prevalentes e com maior abundância média e foram consideradas espécies centrais. Além disso, essas espécies apresentaram alta frequência de dominância e dominância relativa média. Os pares de espécies de helmintos *Amoebotaenia cuneata* (von Linstow, 1872) - *Raillietina tetragona* (Molin, 1958); *Davainea proglottina* (Davaine, 1860) - *A. cuneata* e *H. gallinarum* - *Capillaria* sp. mostraram correlação positiva significativa entre abundância média e prevalência parasitária. *Gongylonema ingluvicola* Ransom, 1904 and *H. gallinarum* mostraram correlação significativa entre sexo do hospedeiro e abundância de helminto, enquanto que não houve correlação no sexo do hospedeiro e a prevalência destes. O conhecimento da estrutura da comunidade de helmintos em frangos criados no quintal é importante para a adoção de medidas de controle e prevenção de helmintos.

Palavras-chaves: aves – Cestoda – Nematoda – ecologia parasitária – prevalência – riqueza parasitaria

RESUMEN

Las gallinas domésticas, *Gallus gallus domesticus* Linnaeus, 1758*,* son un componente importante del segmento de la agroindustria y Brasil es uno de los mayores productores y exportadores de pollos. El presente estudio tuvo como objetivo caracterizar la estructura comunitaria de helmintos de pollos criados en corrales, *G. g. domesticus*, en Seropédica, Rio de Janeiro, Brasil. Se estudiaron 55 aves adultas. La riqueza total de helmintos fue de 12 especies. *Capillaria* sp. y *Heterakis gallinarum* (Schrank, 1788), que se encontraron en el intestino delgado y ciego intestinal, respectivamente, fueron las especies más prevalentes con mayor abundancia promedio y fueron consideradas especies centrales. Además, estas especies mostraron los valores mayores de frecuencia de dominancia y de dominancia relativa media. Los pares de especies de helmintos, *Amoebotaenia cuneata* (von Linstow, 1872) - *Raillietina tetragona* (Molin, 1958); *Davainea proglottina* (Davaine, 1860) - *A. cuneata* y *H. gallinarum* - *Capillaria* sp. mostraron una correlación positiva significativa entre la abundancia media y la prevalencia de parásitos. *Gongylonema ingluvicola* Ransom, 1904 y *H. gallinarum* mostraron una correlación significativa entre el sexo del huésped y la abundancia de helmintos, mientras que no hubo correlación entre el sexo del huésped y la prevalencia de helmintos. El conocimiento de la estructura de la comunidad de helmintos en los pollos criados en corrales es importante para la adopción de medidas de prevención y control de helmintos.

Palabras clave: aves – Cestoda – Nematoda – ecología parasitaria – prevalência – riqueza parasitaria

INTRODUCTION

The domestic chicken, *Gallus gallus domesticus* Linnaeus*,* 1758, is an important component of the agribusiness segment, and Brazil is one of the world's largest broiler producers and exporters (ABPA, 2020). There is high demand for chicken meat and eggs due to high nutritional value and affordable price (Geraldo *et al*., 2020). In the past decade, the market of organic, healthy, and sustainable products and contingent of consumers concerned about animal welfare have both increased (Miao *et al.*, 2005; Alsaffar, 2016).

Chickens under free-range production typically show signs of calmness and comfort (Bogdanov, 1997). However, chickens reared freely are more exposed to helminths than those kept in confinement (Cardozo & Yamamura, 2004; Lozano *et al.*, 2019), since they have direct contact with soil, and free access to different areas and food items (Gomes *et al.*, 2009). A high burden of gastrointestinal helminths can decrease productivity by increasing mortality, causing economic losses to breeders (Ruff, 1999).

In Brazil, many studies have reported the occurrence of helminths in *G. g. domesticus* in different regions and breeding systems (Grisi & Cravalho, 1974; Quadros *et al.*, 2015; Silva *et al.*, 2016; Silva *et al.*, 2018; Brandão-Simões *et al.*, 2020). However, little is known about the helminth community structure of *G. g. domesticus* in this condition (Silva *et al.*, 2016, 2018). Studies of parasites through an ecological approach, to detect possible patterns in the organization and composition of parasite populations, can be relevant to understand the distribution of the pathogen species in different breeding systems. Moreover, it can assist helminthic control strategies and contribute to develop biosafety programs. Here, we characterize the composition and structure of the helminth component community of backyard chickens in Seropédica, Rio de Janeiro, Brazil.

MATERIALES AND METHODS

Host and parasitological procedures
Fifty-five sexually mature *G. g. domesticus* were purchased dead during 1999-2000. The study was carried out in Seropédica (22° 44′ 29″S, 43° 42′ 19″W), state of Rio de Janeiro, Brazil. The chickens were subjected to procedures in accordance with standard international parasitological guidelines (Yazwinski *et al.*, 2003). The gastrointestinal tract, eyeball, trachea, lungs, kidneys, and bursa of Fabricius were examined. The helminths were washed in saline solution (0.85% NaCl) and fixed in AFA (2% acetic acid, 3% formaldehyde and 95% ethanol). For microscopic studies, nematodes were cleared in lactophenol and cestodes were stained with chlorhydric carmine. The helminth parasites were identified according to Yamaguti (1961), Khalil *et al.* (1994), Vicente *et al.* (1995) and Shmidt (1986), along with specific papers.

Data analysis
To analyze the helminth community structure, we considered prevalence, mean intensity and mean abundance of each species using the procedures described by Bush et al. (1997). Statistical analyses were performed only for the parasite species with prevalence higher than 10%.

The frequency of dominance and the Berger-Parker index of each parasite species were calculated according to Rohde *et al.* (1995) and Magurran (2004).

Species richness, helminth richness at the infracommunity level and Brillouin's diversity index (log10) were calculated. The parasite infracommunity species were classified as: a) central species (present in more than one-third of the hosts); b) secondary species (present in one- to two-thirds of the hosts); and c) satellite species (present in less than a third of the hosts) (Bush & Holmes, 1986).

Mann-Whitney *U* test values and chi-square analysis were used for comparison of parasite abundance and prevalence, respectively, and between host sex (Zar, 1999). These analyses were performed following the recommendations of Wilkinson (1990) using the SYSTAT™ statistical software.

The possible relationships between prevalence and abundance between pairs of concurrent species were determined using the chi-square test and Spearman rank correlation coefficient, respectively (Ludwig & Reynolds, 1988). Statistical significance was considered at $P < 0.05$. All ecological terminology was used according to Bush *et al.* (1997).

Ethic aspects
All national and institutional guidelines for use of animals were followed. All chickens were purchased died.

RESULTS

The overall helminth richness was 12 species. The helminth richness at the infracommunity level ranged of 1 to 10, with a mean of 5.2 ± 2.2. Fifty-three hosts (96%) presented two or more helminth species.

A total of 10,708 specimens were collected, with mean abundance of 194.7 ± 283.9. Nematodes were the most abundant, with 7 species representing 60.7% of all parasite specimens collected. All chickens examined were parasitized by at least one species of nematode and 85.7% (47) were parasitized by cestodes. Trematodes were not found. The nematode *Capillaria* Zeder, 1800 was not identified to the species level as cestode *Raillietina* Fuhrmann, 1920 due to the low quality of the morphological characteristics, such as scolex and mature proglottids, of the specimens collected.

The nematodes *Capillaria* sp. and *Heterakis gallinarum* (Schrank, 1788), recovered from the small intestine and cecum, respectively, presented the highest values of prevalence and mean abundance and were considered central species (Table 1). In addition, these species presented the highest values of frequency of dominance and Berger-Parker index (Table 2); while the cestode *Raillietina* sp. presented the highest value of mean intensity and was considered a secondary species along with the cestode *Amoebotaenia cuneata* (von Linstow, 1872) and the nematodes *Gongylonema ingluvicola* Ransom, 1904 and *Oxyspirura mansoni*. (Cobbold, 1879) (Table 2). Secondary species were found in the small intestine, crop and eye. *Tetrameres confusa* Travassos, 1917 showed the lowest values of prevalence, mean intensity and abundance (Table 1). The Brillouin index for helminth infracommunities had mean value of 0.43 ± 0.19 and maximum diversity of 0.78.

Table 1. Prevalence (P), mean intensity (MI), and mean abundance (MA) followed by standard error, community status (CS) and site of infection (SI) of helminth parasites of *Gallus gallus. domesticus* from Seropédica, Rio de Janeiro, Brazil.

Helminth species	P (%)	MI	MA	CS*	SI
CESTODA					
Amoebotaenia cuneata	41.8	45.4±99.9	19.0±67.6	S	Small Intestine
Davainea proglottina	30.9	37.2±53.0	11.5±33.7	Sa	Small Intestine
Raillietina tetragona	32.3	16.5±29.2	5.4±18.2	Sa	Small Intestine
Raillietina echinobothrida	27.3	17.5±22.4	4.8±13.9	Sa	Small Intestine
Raillietina sp.	43.6	81.9±253.3	35.7±170.3	S	Small Intestine
NEMATODA					
Ascaridia galli	21.8	37.6±65.7	8.2±33.6	Sa	Small Intestine
Capillaria sp.	90.9	46.0±83.7	41.8±80.9	C	Small Intestine
Cheilospirura hamulosa	25.4	8.9±13.2	2.2±7.6	Sa	Ventricle
Gongylonema ingluvicola	49.1	7.0±8.2	3.4±6.7	S	Crop
Heterakis gallinarum	72.7	63.1±122.8	45.9±108.1	C	Cecum
Oxyspirura mansoni	56.4	27.7±36.2	15.6±30.4	S	Eye
Tetrameres confusa	23.6	4.1±4.4	1.0±2.7	Sa	Proventriculum

* Central species (C). Secondary species (S) and Satellite species (Sa).

Table 2. Frequency of dominance and Berger Parker Index values (BPI) of the helminth species from *Gallus gallus domesticus* in the municipality of Seropédica. Rio de Janeiro, Brazil.

Helminth species	Frequency of dominance	BPI
CESTODA		
Amoebotaenia cuneata	5	0.08±0.16
Davainea proglottina	2	0.05±0.12
Raillietina tetragona	2	0.04±0.10
Raillietina echinobothrida	3	0.04±0.14
Raillietina sp.	7	0.12±0.24
NEMATODA		
Ascaridia galli	1	0.02±0.09
Capillaria sp.	13	0.25±0.22
Cheilospirura hamulosa	0	0.01±0.03
Gongylonema ingluvicola	0	0.03±0.06
Heterakis gallinarum	15	0.23±0.24
Oxyspirura mansoni	7	0.12±0.21
Tetrameres confusa	0	0.01±0.03

Table 3. Mann-Whitney *U* test values (Z, value of the normal U test approximation) and Chi square (^{2}) to evaluate the relationship between the sex of *Gallus domesticus* and the abundance and prevalence of the components of its parasitic community (*P*= level of significance).

Helminth species	*Z*	*P*	χ^2	*P*
CESTODA				
Amoebotaenia cuneata	-0.18	0.86	0.15	0.70
Davainea proglottina	-0.72	0.47	0.02	0.89
Raillietina tetragona	-0.37	0.71	0.02	0.89
Raillietina echinobothrida	-1.53	0.13	0.23	0.63
Raillietina sp.	-0.47	0.64	0.09	0.76
NEMATODA				
Ascaridia galli	-0.26	0.79	0.22	0.64
Capillaria sp.	-1.85	0.06	0.01	0.94
Cheilospirura hamulosa	-0.71	0.48	0.06	0.81
Gongylonema ingluvicola	-2.11*	0.04	0.96	0.33
Heterakis gallinarum	-2.01*	0.04	0.02	0.89
Oxyspirura mansoni	-1.41	0.16	0.09	0.76
Tetrameres confusa	-0.85	0.40	0.12	0.73

*Significant Value

Gongylonema ingluvicula and *H. gallinarum* showed significant correlation between host sex and helminth abundance, while there was no correlation between host sex and helminths prevalence (Table 3).

The following pairs of concurrent helminths showed significant positive correlation between their abundance and prevalence, respectively: *Amoebotaenia cuneata* – *Raillietina tetragona* (Molin, 1858) (*rs* = 0.37; $P = 0.006/^{2} = 8.59$; $P = 0.004$), *Davainea proglottina* (Davaine, 1860) – *A. cuneata* (*rs* = 0.46; $P < 0.001/^{2} = 3.53$; $P = 0.049$); and *H. gallinarum* – *Capillaria* sp. ($r_s = 0.39$; $P = 0.003/^{2} = 5.6$; $P = 0.024$). The pair *R. tetragona* – *Ascaridia galli* showed significant correlation only between abundance ($r_s = 0.37$; $P = 0.005/^{2} = 2.22$; $P = 0.136$).

DISCUSSION

Several studies of the characteristics of the helminth communities of *G. g. domesticus* have been performed worldwide (Schou *et al.*, 2007; Idika *et al.*, 2016; Slimane, 2016; Berhe *et al.*, 2019; Sarba *et al.*, 2019, among others). In Brazil, most studies have focused only on the description of the helminth fauna (Costa & Freitas, 1959; Grisi & Carvalho, 1974; Gomes *et al.*, 2009; Siqueira & Marques, 2016). More recently, knowledge about the helminth community structure of chickens raised in different production systems has increased, mainly in the state of São Paulo (Silva *et al.*, 2016; 2018). In contrast, there are few reports of the helminth community structure of *G. g. domesticus* in the state of Rio de Janeiro (Grisi & Carvalho, 1974; Gomes *et al.*, 2009). The composition and structure of the helminth community of a specific host population correspond to a pool of available parasite species in a specific locality. Moreover, the structure and composition of a helminth community can vary due to biotic and abiotic factors, such as environmental changes, parasite control, host behavior and age, among others (Poulin, 2007; Santoro *et al.*, 2012; Simões *et al.*, 2016; Cardoso *et al.*, 2019).

Grisi & Cravalho (1974) reported 13 species of helminths in *G. g. domesticus* in Seropédica, similar to the present study, in which we found 12 species in the same locality. However, some species differed between the studies. The species *Syngamus trachea* (Montagu, 1811), *Heterakis brevispiculum* Gendre, 1911, *Capillaria collaris* (Linstow, 1873) and *C. obsignata* Madsen, 1945 were not found in our study, although we found nematodes belonging to the genus *Capillaria.* Unfortunately, the specific identification was not possible. Moreover, we collected two species of cestodes (*Davainea proglottina* and *Amoebotaenia cuneata*) that were not reported by Grisi & Carvalho (1974). Neither study found trematodes. In contrast, Silva *et al.* (2016, 2018) reported trematodes in different regions of São Paulo state. These differences have also been observed in chickens from African and Asian countries (Berhe *et al.*, 2019; Chege *et al.*, 2015; Junaidu *et al.*, 2014; Schou *et al.*, 2007). Additionally, nematodes and cestodes have been reported with more frequency than trematodes in the component helminth community in chickens by many researchers around the world (Sarba *et al.*, 2019; Idika *et al.*, 2016; Slimane, 2016; Hussen *et al.*, 2012). Variation in environmental conditions can favor or disfavor the presence of intermediate hosts responsible for cestode and trematode transmission and can also influence the occurrence of these species in the helminth community of the definitive host.

In the present study, all chickens were infected with the nematodes *H. gallinarum* and *Capillaria* sp., and both were the more prevalent (central species), with high frequency of dominance and mean values of Berger-Parker index in the helminth community. Similar findings were also observed by Gomes *et al.* (2009), Siqueira & Marques (2016) and Silva *et al.* (2018) in chickens reared in extensive, semi-intensive and/or intensive systems. Parasites with a direct life cycle have a higher probability of infecting definitive hosts since there is no intermediate host. In addition, the high fertility of these female nematodes increases the likelihood of host infection due to more eggs eliminated in the environment. The chickens in the present study were raised free outdoors and allowed to find food by foraging in the surface soil layer. Thus, these factors can partly explain the high prevalence of nematodes in the helminth community.

The influence of host sex on the helminth abundance has been observed in different helminth community structures (Simões *et al.*, 2014; Wendt *et al.*, 2018). The influence of female chickens was observed regarding two nematodes species (*H. gallinarum* and *Capillaria* sp.). However, this result must be viewed with caution, since the proportion of female chickens used in the study was much greater than of males (10: 1). In addition, many studies of the helminth community of *G. g. domesticus* have not reported significant differences in helminth infection rate between male and female animals (Poulsen *et al.*, 2000; Abdelqader *et al.*, 2008; Ebrahimi *et al.*, 2014).

The positive correlation of abundance and prevalence between pairs of cestodes and nematodes may be related to the similarities of their life cycle. The cestodes *R. tetragona*, *A. cuneata* and *D. proglottina* need an intermediate host, such as an ant, beetle or fly; earthworm; and slug respectively (Acha & Szyfres, 2003; Taylor *et al.*, 2017). One of the food items collected by the chicken was small animals found in the soil. Similarly, this can explain the positive correlation with nematodes, which have a direct life cycle and oral infection.

Free-range chickens are exposed to more parasites than those reared in intensive systems (Ruff, 1999; Lozano *et al.*, 2019) due to the direct contact with soil and intermediate hosts. Tapeworms are frequently encountered in the chickens in these production systems and most species cause low pathogenicity (Ruff, 1999). However, *R. tetragona* and *D. proglottina* are more pathogenic, reducing the performance of parasitized animals (Ruff, 1999; Nnadi & George, 2010). Moreover, parasitism by *Capillaria* spp. can also cause production losses (Ruff, 1999). Seasonality and other collection areas should be considered in future studies to measure the heterogeneity and stability of the parasite helminth community.

Knowledge of the helminth community structure in backyard chickens can be relevant to allow breeders to adopt better measures for control and prevention of helminths. Moreover, other variables like climate conditions, hygiene, age, and animal density should also be considered since they may cause influence in the parasitism. In addition, this study can also serve as a baseline for future parasitological studies on other bird species.

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