



Identification of ecto and endoparasites in domestic pigeons (Columba livia) from the urban area of Villavicencio, Meta, Colombia

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ABSTRACT

Objetive. Determine the presence of internal and external parasites in common pigeons (*Columba livia*) in the urban area of the city of Villavicencio, Meta, during the transition from summer to winter in 2017. Materials and Methods. 72 pigeons from three communes of the city were captured and subjected to visual inspection for external parasites, at the same time, samples of fecal material were obtained for stool analysis using the Sheather's method. Blood samples were smeared and Giemsa stained for hemotropic agents. Results. Two species of external parasites were found in 100% of the samples: Mallophaga lice (*Columbicola columbae*) and pigeon fly (*Pseudolynchia canariensis*). Within the internal parasites, two protozoa were found: *Haemoproteus* spp. (9/34) in blood smear and *Eimeria* spp. (26/72) in feces, followed by nematodes: *Ascaridia* spp. (3/72) and *Capillaria* spp. (10/72) and cestodes: *Raillietina* spp. (1/72). No association of parasitism was found with the sex of the pigeon or the sampling commune. **Conclusions**. The presence of internal parasites was low, except for *Eimeria* spp., these data represent important information on the potential risk for animal and human health, especially for commercial populations of birds and native avifauna. These results indicate that sanitary and control programs are required in the pigeon populations of the city.

Keywords: Columba livia; Columbicola columbae; parasitism (Source: DeCs).

RESUMEN

Objetivo. Determinar la presencia de parásitos internos y externos en palomas comunes (Columba livia) del área urbana de la ciudad de Villavicencio, Meta, durante la transición verano a invierno del año 2017. Materiales y Métodos. Se capturaron 72 palomas de tres comunas de la ciudad y sometidas a inspección visual para parásitos externos, a la vez se obtuvo muestras de material fecal para análisis coprológico mediante el método de Sheather's. Se realizaron frotis con las muestras de sangre y tinción con Giemsa para agentes hemotrópicos. Resultados. En el 100% de las muestras se encontraron dos especies de parásitos externos: piojos malófagos (Columbicola columbae) y mosca de paloma (Pseudolynchia canariensis). Dentro de los parásitos internos se encontraron dos

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protozoarios: *Haemoproteus* spp. 26.5 % (9/34) en frotis sanguíneo y *Eimeria* spp. 36% (26/72) en heces, seguido de los nematodos: *Ascaridia* spp. 4.2% (3/72) y *Capillaria* spp. 13.8% (10/72) y cestodos: *Raillietina* spp. 1.38% (1/72). No se encontró asociación del parasitismo con el sexo de la paloma o la comuna de muestreo. **Conclusiones.** La presencia de parásitos internos fue baja, excepto para *Eimeria* spp., estos datos representan información importante del riesgo potencial para la salud animal y humana, especialmente para poblaciones comerciales de aves y la avifauna nativa. Estos resultados indican que se requieren programas sanitarios y de control en las poblaciones de palomas de la ciudad.

Palabras clave: Parasitosis; Columbicola columbae; parásitos (Fuente: DeCs).

INTRODUCTION

Domestic pigeons (Columba livia domestica) are urban feral birds adapted to life in diverse niches. They are distributed throughout all continents, except for Antarctica (1). According to Del Hoyo et al (2), this species originated in a large area of Eurasia and Africa (2).

A risk analysis of invasive species in Colombia catalogs domestic pigeons within the "high risk" category. As such, they should be controlled and be subject to management actions, environmental education and specific legislation to prevent and mitigate risk (3,4). Despite this, little has been done to stop population growth in many urban and rural areas, thus becoming an actual plague in the whole national territory.

Many economic losses are attributed to pigeons, including damages to stored food, to buildings and facilities, and competition for territory and food with native avifauna (5,6). However, the most severe effects of their presence are those related to pigeons being a potential source of transmission of zoonotic agents that affect public health such as histoplasmosis, ornithosis, salmonellosis, cryptococcosis, campylobacteriosis and chlamydiosis (7,8,9,10). Several diseases affect pigeon health, with internal and external parasitism among the most frequently reported conditions (11,12,13). Besides, pigeons can be a reservoir for parasitic, bacterial and viral infections for other birds (14,15). Due to pigeon population growth in Villavicencio, their close contact with humans, and commercial operations with other bird species and with native avifauna, the goal of this study was to identify the main internal and external parasite that affect this species in three districts of the urban area of said city.

MATERIALS AND METHODS

Sample location and size. A cross-sectional epidemiological study was conducted in districts 2, 3 and 4 of Villavicencio, Meta, which have the oldest architecture and the largest pigeon population in the city (Figure 1). The sample was designed through probability sampling in Epidat 3.1, based on a population of 14718 pigeons, according to unpublished data by González et al (personal communication), with a sampling fraction of 70%.



Figure 1. Presence of pigeons in a public place, district 2.

To determine the common pigeon (*Columba livia*) population in public roads of Villavicencio's urban area, we took into account the information obtained by González et al (personal communication) regarding an estimated population of 14718 birds.

Specifically, out of that report, we only took into account the districts with the greatest pigeon population density, namely districts 2, 3 and 4, which amount to a population of 10343 pigeons, i.e., 70.3% of the total population. We start from the assumption that, according to the study by González et al (personal communication), 10% (p) of the pigeon population have some kind of parasite, and 90% do not (q): The following formula was used to determine the sample size, with a probability of success of 5%, a confidence interval of 95% and an accuracy of 5%:

$$n_0 = \frac{Z^2 * p * q * N}{(N-1) * e^2 + Z^2 * p * q}$$

Where:

- n= Estimated sample size
- Z= Standard normal distribution value
- N= Population size
- p= Estimated proportion
- q= 1 P
- e= Sampling error

Once values are replaced:

$$n_0 = \frac{1,96^2 * 0,05 * 0,95 * 14718}{(14718 - 1) * 0.05^2 + 1.96^2 * 0.05 * 0.95} = 72$$

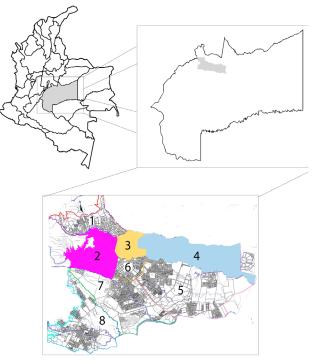
These 72 pigeons were captured in the three districts mentioned above in one climatic period during the dry season's transition to the wet season (Table 1, Figure 2).

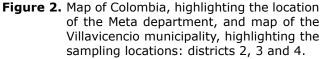
Table 1. San	npling distribution.
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District	Pigeon population	% of total population	Sample size
2	6.328	61.18 %	44
3	2.205	21.32 %	15
4	1.810	17.50 %	13
Total	10.343	100 %	72

Capture and necropsy. Pigeons were captured in the transition period from the dry season to the wet season of 2017 —i.e., from April to June—, by using a nylon net. They were transported

in cages to the Histopathology Laboratory of the University of Los Llanos, where they were sacrificed with 10% ketamine, via IM, with a dose of 40 mg/kg. Sex was identified through direct observation of the gonads. During the necropsy, feathers were carefully inspected, collecting ectoparasite specimens visible on feathers and skin. The identification of ectoparasites was made with a stereo microscope (Nixon brand, model SMZ 445), using the conventions developed by Clayton & Price (16), Atkison & Hunter (17) and Graciolli & Carvalho (18).





Sample collection. Before euthanasia, blood was extracted by puncturing the bronchial vein; and thick blood smears were done on microscope slides to identify hemoparasites through Giemsa stain. 5 g of feces per bird were collected from portions of the animals' small and large intestines and put in sterile and refrigerated (4-8°C) glass containers until being processed.

Direct smear. A fecal smear, diluted in 0.9% saline solution and Lugol's Iodine, was observed directly through the microscope, searching for trophozoites, cysts, eggs and oocysts of intestinal parasites.

Concentration technique. Fecal samples were processed by means of the qualitative flotation method in Sheather's solution. One gram of fecal sample and 15 mL of saturated sugar solution with a density of 1.27 were put in a tube, which was filled until forming a meniscus, on top of which a cover slip was placed. After letting it settle for 10 minutes, the coverslip was placed on a slide and observed through a binocular microscope at magnification levels of 10X, 20X, 40X and 100X. (Zeiss-Primo Star, Göttingen, Germany) (19).

Coccidia sporulation. Samples positive for coccidia (at least 5 coccidia per field) underwent the sporulation process in a 2.5% (w/v) solution of potassium dichromate ($K_2Cr_2O_7$). Feces were well mixed in petri dishes and kept at room temperature in a dark, ventilated place to ease sporulation. Coccidia species were determined according to the oocysts morphological characteristics and their time of sporulation (20).

Giemsa stain. Blood smears were done by using fresh blood samples. Slides were previously fixed with ethanol, and later covered with the stain solution, diluted 1:10, for 12 minutes, flushed with abundant water, and observed in the

microscope at 100x magnification, searching for hemoparasites. Hemoparasites were identified by following Soulsby's technique (21).

Statistical analysis. Results were analyzed by applying Fisher's exact test and the chi-squared test (X^2) with a probability of 95% (α = 0,05), associating the parasitic species, the pigeon's sex, and the district. Data were analyzed by using the statistics program SPSS, version 23.0 for Windows and Epidat 3.1.

Ethical aspects. This project was approved by the Bioethics Committee of the Faculty of Agricultural Sciences and Natural Resources of the University of Los Llanos, according to Record 003 of April 18th, 2017.

RESULTS

Out of the 72 captured pigeons, 19 were females, and 53 were males. 100% of the pigeons showed the presence of two types of external parasites: Mallophaga lice (*Columbicola columbae*) and pigeon flies (*Pseudolynchia canariensis*) (Figure 3). The first ones were mainly found in the remiges and the chest and abdomen areas, while the second ones did not have a differentiated distribution over the pigeons' bodies.

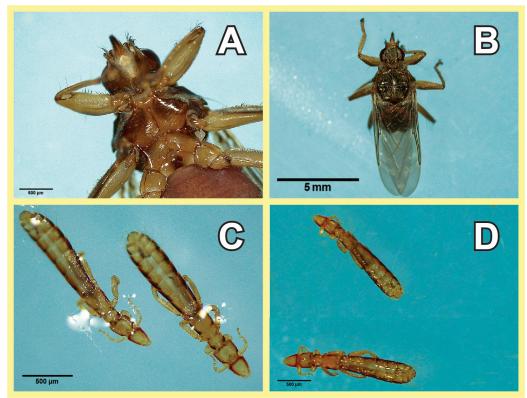


Figure 3. Ectoparasites detected in birds of the Columbidae family. (A and B) Pigeon louse fly (*Pseudolynchia canariensis*); (C and D) Mallophaga lice (*Columbicola columbae*).

In the macroscopic examination of the intestinal tract, adult parasites were collected in 58.33% (42/72) of the pigeons. 43.05% (31/72) of those parasites were nematode parasites, 9.72% (7/72) segmented parasites matching cestodes, and in 6.94% (5/72) of the birds a multiple association of nematodes and cestodes was found (Figure 4).

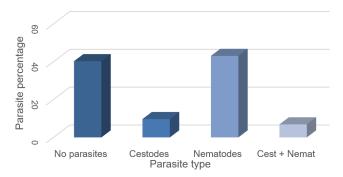


Figure 4. Count of adult parasites in the intestines of Villavicencio pigeons.

Parasite eggs were found in the feces samples of 30 pigeons, classified according to egg type as: type 1: one egg type; type 2: two egg types; type three: three egg types (Figure 5).

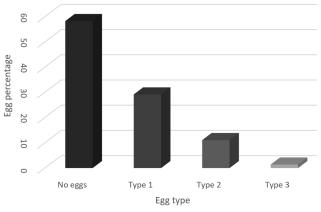


Figure 5. Count of parasite egg types in Villavicencio pigeon feces.

The direct smear of small intestine fecal sample was negative in 69 out of 72 samples, and only one sample contained *Ascaridia* spp. and two contained *Capillaria* spp. As for the birds' large intestine, 78% (56/72) of the samples were negative, 1.39% (1/72) containing *Ascaridia* spp., 2.8% (2/72) containing *Capillaria* spp. and 16.67% (12/72) *Eimeria* spp, with 1.4% (1/72) of samples having the presence of *Capillaria* spp. and *Eimeria* spp. (Figure 6- Table 2).

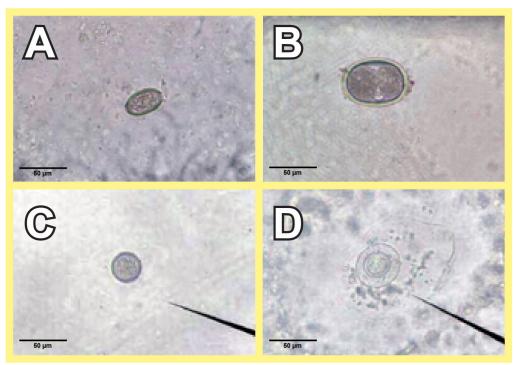


Figure 6. Photomicrographs of endoparasites detected in Columbidae-family birds. (A) *Capillaria* spp egg; (B) *Ascaridia* spp egg; (C) Coccidia oocyst (D) *Raillietina* spp. egg.

Parasite	Small intestine	% of population	Large intestine	% of population	Total positives
Ascaridia spp.	1/72	1.39%	1/72	1.39%	2.8% (2/72)
<i>Capillaria</i> spp.	2/72	2.78%	2/72	2.78%	5.6% (4/72)
<i>Eimeria</i> spp	0/72	0%	12/72	16.67%	16.67(12/72)
Eim. + Capil.	0/72	0%	1/72	1.39%	1.39%(1/72)
Negative	69/72	95.83%	56/72	77.77%	-
Total	72	100%	72	100	26.38%(19/72)

Table 2. Number and percentage of parasites found through small and large intestine direct smears in Villavicencio pigeons.

The flotation technique resulted in 66 out of 72 small intestine samples being negative for intestinal parasite eggs, and *Raillietina* spp., *Ascaridia* spp., *Capillaria* spp. and *Eimeria* spp were identified. Among the large intestine samples, 48 out of 72 samples were negative; and *Capillaria* spp., *Eimeria* spp. and 4/72 associations *Eimeria* plus *Capillaria* were found (Table 3).

Overall, the prevalence of intestinal endoparasites by type of parasite was 33.3% (24/72) of *Eimeria* spp., 11.1% (8/72) of *Capillaria* spp., 1.39% (1/72) of *Ascaridia* spp., and 1.39% (1/72) of *Raillietina* spp. As for blood protozoa, 26.5% (9/34) of *Haemoproteus* spp. were found in blood smears (Figure 7 - Table 4).

Table 3.	Number and percentage of parasites detected through Sheather's technique in small and large intestine
	fecal matter in Villavicencio pigeons.

Parasite	Small intestine	% of population	Large intestine	% of population	Total positives
<i>Raillietina</i> spp.	1/72	1.39%	0/72	0	1.39%(1/72)
<i>Ascaridia</i> spp	1/72	1.39%	1.39% 0/72 0		1.39%(1/72)
<i>Capillaria</i> spp.	1/72	1.39%	3/72	4.17%	5.55%(4/72)
<i>Eimeria</i> spp	3/72	4.17%	17/72	23.61%	27.7%(20/72)
Eim. + Capil.	0/72	0	4/72	5.56%	5.55%(4/72)
Negative	66/72	91.66%	48/72	66.66	-
Total	72	100%	72	100%	41.6%(30/72)

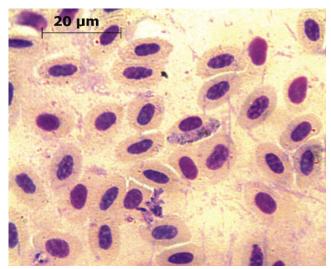


Figure 7. *Haemoproteus columbae* gametocyte, see arrow.

Table 4. Parasite prevalence in pigeons studied in
three districts of Villavicencio.

Parasite group	Species	n	Prevalence percentage
Cestodes	Raillietina spp.	1/72	2.04%
Nematodes	Ascaridia spp	3/72	6.12%
	<i>Capillaria</i> spp.	10/72	16.33%
Intestinal protozoa	Eimeria spp.	26/72	65.31%
Blood protozoa	Haemoproteus spp.	9/34	26.47%

The identified intestinal parasites were related to the pigeon's sex by using the frequency analysis of contingency tables and by correlating the categorical variables. Their statistical significance was assessed by using the chi-squared test (X^2), with a confidence of 95%. No relation was found between the presence of a given parasitic species and the pigeon's sex (Table 5). By relating the identified intestinal parasites to the sampled districts (2, 3 and 4), using the frequency analysis of contingency tables, and correlating the categorical variables, no relation was found between them (Table 6).

Parasite	Sex		- X ²	Р	Drovalance ratio	Confidence interval	
Falasite	Female	Male	- X-	F	Prevalence ratio	confidence interval	
Ascaridia spp.	2	1	2.614	0.106	5.579	0.536 - 58.064	
Capillaria spp.	1	7	0.893	0.344	0.398	0.052 - 3.030	
Raillietina spp	0	1	0.894	0.344	0.000	-	
Eimeria spp	12	20	0.364	0.547	1.673	1.028 - 2.724	
Eim.+ Capil.	0	5	1.926	0.165	0.000	-	
Total	15	34					
gl=1; P= 95%							

Tabla 6. Bivariate (chi-squared) statistical analysis between intestinal parasites and Villavicencio districts.

Davaalta	District		- X ²	Р	Ducus lon co untio	Confidence interval	
Parasite	2	3	4	- X-	Р	Prevalence ratio	Confidence Interval
Ascaridia spp.	3	0	0	1.993	0.369	-	-
<i>Capillaria</i> spp.	5	3	0	2.788	0.248	1.760	0.476 - 6.496
Raillietina spp.	1	0	0	0.648	0.723	-	-
Eimeria spp.	18	7	7	0.199	0.905	1.140	0.597 - 2.178
Eim.+ Capil.	2	3	0	5.319	0.070	4.400	0.811 - 23.857
Total	29	13	7				

GL= 2; P= 95%

DISCUSSION

Domestic pigeons (*Columba livia*) have become a severe problem in big cities worldwide, given their uncontrolled population growth. The city of Villavicencio, where there are no official control programs, is not alien to this problem, which is why we intended to identify ecto and endoparasites present in the pigeons of three city districts. It is known that some infectious agents are significant to public health or to animal production systems. Among those most well-known, one can include histoplasmosis, Newcastle disease, psittacosis, cryptococcosis, coccidiosis, toxoplasmosis, pseudotuberculosis and salmonellosis (7,8,9,10).

The presence of only two species of ectoparasites in the pigeons object of this study was considered low, and even lower if it is compared with seven species identified in pigeons of the San Martín de Porres market in Lima – Peru (22), eight species in Iran (23) and 60 species in other studies (24). Prevalence figures for ectoparasitosis in *C. livia* domestica can vary according to the season and to climatic and ecogeographic factors related to the hosts' range (25,26).

In nature, only some arthropods carry pathogens. Therefore, vertebrate hosts are more often exposed to hosts free of them. However, even if no pathogens are transmitted, hematophagy causes irritation and distracts hosts, which often results in behavioral defenses (such as grooming) (27,28). Literature on this topic points out that *Haemoproteus columbae* and its vector, the fly *Pseudolynchia canariensis*, are distributed worldwide, although having a more prominent presence in tropical areas (29). In our study, 26.54% of pigeons were infected with this hemoparasite (Figure 7), which entails a prevalence greater than the one reported in

Zaria (Nigeria), i.e., 15.2% (30), similar to a study in Italy that reported 29.4% (31), and lower than reports from South Africa, -96.9%(29) — and Sao Paulo, Brazil — 100% (32) It is important to note that Haemoproteus columbae has not been reported in non-columbid hosts, and therefore these might be its specific hosts (33). Possibly, the high prevalence of this infection depends on the population density of *Haemoproteus* spp. and its vector, as well as the population density of feral and wild pigeons, which are generally high in suburban and urban studies (32,34,35). However, given that the natural populations of C. livia have not been studied much, there are not enough data to make a comparison (29).

The distribution of lice (*Columbicola columbae*) in pigeons' feathers and body matches the one discovered by Harbison & Boughton (36), who found a higher number of adult lice on the abdomen, wings and tail, while immature lice congregate in large numbers on the head and neck. The prevalence of *Columbicola columbae* was similar to the one found in the Canary Islands (37), although greater than the 82.8% reported in Lima (38), 41.3% in Iran (39), 64% in Colombia (40), and 82% in Libya (41).

The prevalence of *Eimeria* spp. oocysts in 65.3% of the samples was higher than reports from the Canary Islands, -50% (37)-, 55% in Envigado, Colombia (40), and lower than the 86.05% reported in Brazil (42), 55-89% in Poland (43) and 90% reported in the region of Jabal al Akhdar in Libya (44). Coccidiosis does not usually evidence any clear clinical manifestations. Pigeons seem healthy but are less active. In some cases, aqueous diarrhea can be observed. In juvenile pigeons, the disease is acute. Sick pigeons have ruffled and brittle feathers, weight loss and bloody diarrhea. Juvenile mortality fluctuates between 5% and 30%. Besides, in some cases, growth inhibition and balance disorders can be observed (45). Many factors can contribute to the differences of Coccidia prevalence among different areas, being climatic conditions, agricultural practices and pigeon breeds some of the most important ones (46).

Among cestodes, *Raillietina* is considered the most common genus in pigeons. In our study, we found a prevalence of 2.0%, which is lower than the 7.8% reported in Venezuela (47) and the 44% found in the study by Foronda et al (37) in Nigeria, where these parasites' high

prevalence was attributed to sampling during the rainy season, when its activity is higher, as its eggs develop faster under high temperature and humidity conditions (48). In our study, sampling was carried out during the transition from the dry to the wet season, with high temperatures but low humidity, which probably did not favor the rapid eclosion of eggs. Taking into account that the dynamics of different parasites can change depending on environmental conditions, the importance of this helminth should not be underestimated, as pigeons can be a source of transmission for backyard and commercialoperation poultry (21).

The low presence of nematode eggs observed with the flotation technique (*Ascaridia* spp= 4.16% and *Capillaria* spp= 13.8%) contrasts with the prevalence reported in Libya -22% (41)— and Brazil -32.56% (42)—. Our study showed mixed gastrointestinal parasitism, which matched the studies by Msoffe et al (49), Abed et al (50) and El-Dakhly et al (51). Likewise, Pérez-García et al (40) found three pigeons with *Capillaria* spp. and *Ascaridia* spp., while five showed *Capillaria* spp. and coccidia oocysts. *Capillaria* sp. prevalence (16.3%) was higher than the 4.32% observed by Kommu et al (52) and 7.63% reported in Nigeria (53).

The lack of difference in parasitism associated to each sex found in this study matches studies carried out in Turkey, which also found that seasons were significant (54). Although Colombia, due to its geographic location, does not have defined seasons, it could be speculated that parasite infestations should be different in the dry season and the wet season, an idea that should be addressed in future studies. Likewise, the lack of differences in parasitism between districts suggests similar epidemiological conditions related to habitat, food, and contact with other birds.

Lastly, the internal and external parasites reported in other studies were identified in Villavicencio pigeons as well, although in a smaller amount. It would be necessary to carrying out longitudinal studies to know more precisely the times of the year with the greatest parasitic infestations. A massive parasite elimination strategy through water could help prevent the transmission of these parasites to native fauna, commercial-operation birds, and humans. This strategy could be reinforced by restricting pigeon feeding in these districts of the city, thus allowing a decrease in these birds' population.

Conflicts of interest

The authors declare having no conflicts of interest.

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