

Digenean parasites of three species of cathartid birds from Formosa Province, Argentina

Digeneos parásitos de tres especies de aves catártidas de la provincia de Formosa, Argentina

Chiberry Lu^{1*}, Díaz Analía², Pagano Luis^{3†} and Drago Fabiana B.⁴

ABSTRACT: The aims of this paper were to increase the knowledge of the diversity of helminth parasites from cathartid birds from Argentina, and to analyse the role of the parasites found as indicators of their diet. Six specimens of three species of vultures captured in Formosa Province, *Coragyps atratus*, *Cathartes burrovianus* and *Cathartes aura* were analysed. Helminths and hosts diet were studied. The helminthological examination revealed the presence of *Petasiger segregatus* (Echinostomatidae) in *C. atratus* and *C. burrovianus*, and *Strigea vaginata* (Strigeidae) in *C. atratus* and *C. aura*. The findings of *S. vaginata* parasitizing *C. aura*, and *P. segregatus* parasitizing *C. burrovianus*, constitute the first records of any helminth in both hosts in Argentina. The examination of the stomach contents revealed the presence of marsupials, snakes and insects (Trichoptera, Formicidae and Muscidae). *Petasiger segregatus* could be a good indicator of the bird's diet; and the findings of *P. segregatus* in *C. atratus* and in *C. burrovianus* allow to infer that these birds have ingested fishes or amphibians some time before being caught. Instead, *Strigea vaginata* has a wide range of intermediate hosts, thus this strigeid species would not be a good indicator of the diet of its definitive hosts.

Keywords: Argentina, Cathartidae, diet, Echinostomatidae, Strigeidae.

RESUMEN: Los objetivos de este trabajo fueron incrementar el conocimiento de los helmintos parásitos de las aves catártidas de Argentina y analizar el rol de los parásitos encontrados como indicadores de su dieta. Se analizaron seis ejemplares de tres especies de jotes capturados en la provincia de Formosa, *Coragyps atratus*, *Cathartes burrovianus* y *Cathartes aura*. Se estudiaron los helmintos y la dieta de los hospedadores. El examen helmintológico reveló la presencia de *Petasiger segregatus* (Echinostomatidae) en *C. atratus* y *C. burrovianus*, y *Strigea vaginata* (Strigeidae) en *C. atratus* y *C. aura*. Los hallazgos de *S. vaginata* parasitando a *C. aura* y de *P. segregatus* parasitando a *C. burrovianus*, constituyen los primeros registros de algún helminto en ambos hospedadores en Argentina. El examen del contenido estomacal reveló la presencia de marsupiales, ofidios e insectos (Trichoptera, Formicidae y Muscidae). *Petasiger segregatus* podría ser un buen indicador de la dieta de las aves, y los hallazgos de *P. segregatus* en *C. atratus* y *C. burrovianus* permiten inferir que estas aves se han alimentado de peces o anfibios un cierto tiempo antes de la captura. En cambio, *Strigea vaginata* tiene un amplio rango de hospedadores intermediarios, por lo tanto, no sería un buen indicador de la dieta de las aves que actúan como sus hospedadores definitivos.

Palabras clave: Argentina, Cathartidae, dieta, Echinostomatidae, Strigeidae.

Although six species of cathartid birds [*Cathartes aura* (Linnaeus), *Cathartes burrovianus* Cassin, *Cathartes melambrotus* Wetmore, *Coragyps atratus* (Bechstein), *Sarcoramphus papa* (Linnaeus) and *Vultur gryphus* Linnaeus] inhabit Argentina, the information about their helminthofauna is scarce. At present, only

three species of digeneans were reported parasitizing the black vulture *C. atratus*: *Strigea vaginata* (Brandes, 1888) (Strigeidae), *Petasiger segregatus* (Dietz, 1909) (Syn. *Paryphostomum segregatum*) and *Petasiger* sp. (Echinostomatidae) (Drago and Lunaschi, 2011).

¹Laboratorio de Ecología Molecular, Centro Austral de Investigaciones Científicas (CCT-Tierra del Fuego – CONICET), Bernardo Houssay 200, V9410, Ushuaia, Argentina.

²Instituto de Limnología Dr. Raúl A. Ringuelet (CCT-La Plata CONICET), Boulevard 120 y 62, 1900. La Plata, Provincia de Buenos Aires, Argentina.

³Universidad Nacional de La Plata, Facultad de Ciencias Naturales y Museo, División Zoología Vertebrados, Paseo del Bosque S/N° (1900) La Plata, Argentina.

⁴Universidad Nacional de La Plata, Facultad de Ciencias Naturales y Museo, División Zoología Invertebrados, Paseo del Bosque S/N° (1900) La Plata, Argentina - Comisión de Investigaciones Científicas de la Provincia de Buenos Aires (CIC).

† The author deceased on 19/03/2020.

The cathartid birds are scavengers taking almost any animal food but also catch small preys such as nesting birds, reptiles, fishes and insects (Thiollay, 1994). In Argentina, the knowledge about their diet is scarce, being most of the studies fragmentary and based on a small number of specimens (see De la Peña, 2019a).

Bird diets can be assessed using a wide range of techniques, which vary greatly from the direct killing of birds to inspect their stomach contents through to non-invasive and repeatable observations. Indirect methods include analyses of faeces or regurgitated food remains, tissue collection for stable isotopes or fatty-acid analyses (Barrett *et al.*, 2007). A less explored method to study the diet of birds is the parasitological or coproparasitological examination. The complex life cycles of parasites may be integrated into intricate food webs and give some clues on food web structure and on the food preferences and foraging strategies of hosts (Marcogliese and Cone, 1997). The most used method, the analysis of stomach contents, is often inefficient in identifying food remains found in the gut. The studies based on gut contents only reflect the last hours of feeding prior to capture. Instead, helminths can stay for months or years within a bird host as evidence of long-term trophic relationships (Calegario-Marques and Amato, 2010). Therefore, helminths found in the gut and other organs open up a new dimension in the study of avian diet. Because a substantial number of helminths that parasitize birds have a heteroxenous cycle, transmission occurs when the bird (definitive host) preys on an intermediate host infected with the immature stage of the parasite. In this case, the finding of a helminth inside the bird's body is evidence that the potential intermediate host belongs in its diet. Therefore, the knowledge of the parasitic helminths of birds could help to solve their trophic jigsaw (Calegario-Marques and Amato, 2010).

The aims of this paper were to increase the knowledge of the diversity of helminth parasites from cathartid birds from Argentina, and to analyse the role of the parasites found as indicators of their diet.

Six cathartid birds were hunted with a shotgun in June and September 2012, and May 2016 in La Marcela farm, Pirané, Formosa Province, Argentina (26° 17' 35" S; 59° 06' 38" W). The authorization was provided by the *Ministerio de la Producción y Ambiente* of Formosa Province. The analysed birds were *C. aura* (turkey vulture, n=1), *C. burrovianus* (lesser yellow-headed vulture, n=3) and *C. atratus* (black vulture, n=2). Birds were dissected in the field; their viscera were fixed in 10% formalin and transported to the laboratory of the *Museo de La Plata* (UNLP). The viscera were examined under a stereoscopic microscopy Stemi 2000-C Zeiss and the helminths were removed. The digeneans were stained with a 1:6 dilution in 96% ethanol of hydrochloric carmine, dehydrated in a graded ethanol

series (70%, 96%, 100%), cleared in xylene, and mounted in Canada balsam. Line drawings were made using a drawing tube. Taxonomic identification was made using specific literature (Dubois, 1968; Kostadinova *et al.*, 2002; Lunaschi and Drago, 2009; Tkach *et al.*, 2016). For echinostomatids, the following relative proportions were calculated after Kostadinova (2005): FO%, length of forebody as a proportion of body length; T%, length of post-testicular field as a proportion of body length; U%, length of uterine field as a proportion of body length. The contents of dissected stomachs were studied by examination under a stereoscopic microscope Stemi 2000-C Zeiss and stored in 70% ethanol or 10% formalin. Vertebrates and invertebrates prey items in each stomach were identified up to the lowest possible taxon. Particularly, for remains of mammals, hairs were studied under light microscopic observation, using a Standard 25 Zeiss optical microscope, and compared with specific publications (Palacio, 2009). The pattern of cuticle scales was studied according to Quadros and Monteiro-Filho (2006), through impression of the hairs on a layer of colourless nail polish, and the morphology of hairs medulla through temporary mounts cleared in hydrogen peroxide 30% or lactophenol. Helminths were deposited in the Helminthological Collection of the *Museo de La Plata*, Argentina (MLP-He 7743-7744-7745).

The helminthological study revealed the presence of two species of digeneans in the intestine: *P. segregatus* and *S. vaginata* (Table 1, Fig. 1). The examination of the stomach contents of studied birds revealed the presence of vertebrates (ophidians and mammals) and insects (Table 1).

The specimens of *S. vaginata* can be distinguished by the combination of the following characters: plump body, distinctly bipartite; hindbody curved dorsally and without true neck; oral sucker terminal; pharynx present; ventral sucker equatorial or post-equatorial; ovary oval; vitellarium follicular, with few follicles in forebody and densely distributed in hindbody; testes *in tandem* and scarcely lobed; copulatory bursa large; genital pore terminal, shallow genital atrium; and genital cone enormous occupying almost half of hindbody.

The specimens of *P. segregatus* can be distinguished by the combination of the following characters: body elongate; tegument armed with spines; forebody long (FO% = 24–30%); head collar reniform, with a deep ventral incision, and with 27 collar spines (4 angle spines on each lappet, lateral spines in single row; dorsal spines in double row); oral sucker spherical; ventral sucker with a deep cavity; pharynx present, oesophagus long; caeca long extending up to posterior end; testes *in tandem*, contiguous; post-testicular field very long (T% = 30–32%), cirrus sac oval, anterior

Table 1. Diet and parasites of cathartid birds from Formosa Province recorded in the present study.

Host birds	Food items (Vertebrates)	Food items (Invertebrates)	Parasite species
<i>Cathartes aura</i> 19/09/2012	marsupials	Trichoptera larvae (Insecta)	<i>S. vaginata</i> (1 specimen)
<i>Cathartes burrovianus</i> 22/06/2012	marsupials ophidians	-----	<i>P. segregatus</i> (9 specimens)
<i>Cathartes burrovianus</i> 22/06/2012	marsupials	Formicidae (Insecta-Hymenoptera)	<i>P. segregatus</i> (1 specimen)
<i>Cathartes burrovianus</i> 22/06/2012	ophidians	-----	<i>P. segregatus</i> (11 specimens)
<i>Coragyps atratus</i> 19/06/2016	marsupials	Muscidae* larvae (Insecta- Diptera)	<i>S. vaginata</i> (1 specimen)
<i>Coragyps atratus</i> 19/06/2016	marsupials	-----	<i>P. segregatus</i> (3 specimens) <i>S. vaginata</i> (21 specimens)

* associated with rotting meat

and dorsal to ventral sucker; internal seminal vesicle voluminous; ovary oval, pre-equatorial; uterus very short (U% = 1,6–1,7%); and vitellarium follicular, extending from ventral sucker up to posterior end.

Strigea vaginata is widely distributed in the Neotropical region, and has been reported parasitizing *C. atratus* from Argentina; *Cariama cristata* (Linnaeus), *Cathartes burrovianus urubitinga* von Pelzeln, *Cathartes* sp., *C. atratus*, *S. papa* and *Spizaetus ornatus* (Daudin) from Brazil; *Caracara plancus* (Miller), *Cercibis oxycerca* (von Spix), and *Theristicus caudatus* (Boddaert) from Colombia; *Cathartes aura aura* (Linnaeus) from Cuba; and *Amazonetta brasiliensis* (Gmelin), *Rupornis magnirostris* (Gmelin), *Buteo* sp. and *C. atratus* from Venezuela (Dubois and Macko, 1972; Fernandes et al., 2015). The finding of *S. vaginata* parasitizing *C. aura* constitutes the first record of any helminth in this host species from Argentina.

Petasiger segregatus possesses a distribution restricted to the Neotropical region, and was found mainly parasitizing cathartid birds (*C. atratus*, *Coragyps atratus foetens*, *C. aura*, *Cathartes aura ruficollis*, *C. b. urubitinga* and *S. papa*) from Argentina, Brazil, Venezuela, British Guiana and Paraguay (Fernandes et al., 2015). This echinostomatid was also recorded parasitizing ichthyophagous birds, specifically *Phalacrocorax brasilianus* from Argentina and Brazil (Fernandes et al., 2015). The finding of *P. segregatus* parasitizing *C. burrovianus* constitutes the first record of any helminth in this host species from Argentina.

The reports on the feeding of *C. aura* in Argentina include snakes, birds, wild and domestic mammals and fruits. Among wild mammals, several species of Canidae, Cervidae, Dasypodidae, Didelphidae, Mustelidae, Myrmecophagidae, Procyonidae, and Tayassuidae were mentioned (Di Giacomo, 2005; De la Peña, 2019a). In this research, some larval insects (Trichoptera) are added as part of the diet of these birds.

The lesser yellow-headed vulture, *C. burrovianus* is a scavenger that feeds mainly on vertebrates, the reports in Argentina include carcasses of medium-sized wild mammals, run over pets, snakes, lizards and fishes. Among wild mammals, several species of Canidae, Caviidae, Mustelidae, Myrmecophagidae and Procyonidae were mentioned (Di Giacomo, 2005; De la Peña, 2019a). In this research, marsupials and insects (Formicidae) are added as part of the diet of these birds.

The black vulture, *C. atratus*, is a general scavenger taking almost any animal food, also catches small preys; the reports in Argentina include fishes, reptiles, birds, wild and domestic mammals, and insects. Among wild mammals, several species of Canidae, Caviidae, Cervidae, Dasypodidae, Tapiridae, Tayassuidae and Procyonidae were mentioned (De la Peña, 2019a; Di Giacomo, 2005). In this research, marsupials are added as part of the diet of these birds.

On the other hand, the finding of helminth species with heteroxenous life cycles can provide information about the diet of their host. The life cycle of *P. segregatus* was studied by Lie and Basch (1967), who found snails (*Biomphalaria* spp.) naturally infected with cercariae, and experimentally obtained metacercariae in tadpoles (*Lithobates catesbeianus*) and fishes (*Carassius auratus auratus* and *Danio rerio*), and adults in birds (*Coragyps atratus foetens*). In addition, they suggested that *P. segregatus* can use other host groups as second intermediate host, because *C. a. foetens* does not feed on fish. However, later findings of *P. segregatus* in ichthyophagous birds (*P. brasiliensis*) (Fernandes et al., 2015), and reports of cathartids that occasionally feed on large fishes, allow to assume that fish are the most suitable hosts for this echinostomatid species. Consequently, *P. segregatus* could be a good indicator of the bird's diet.

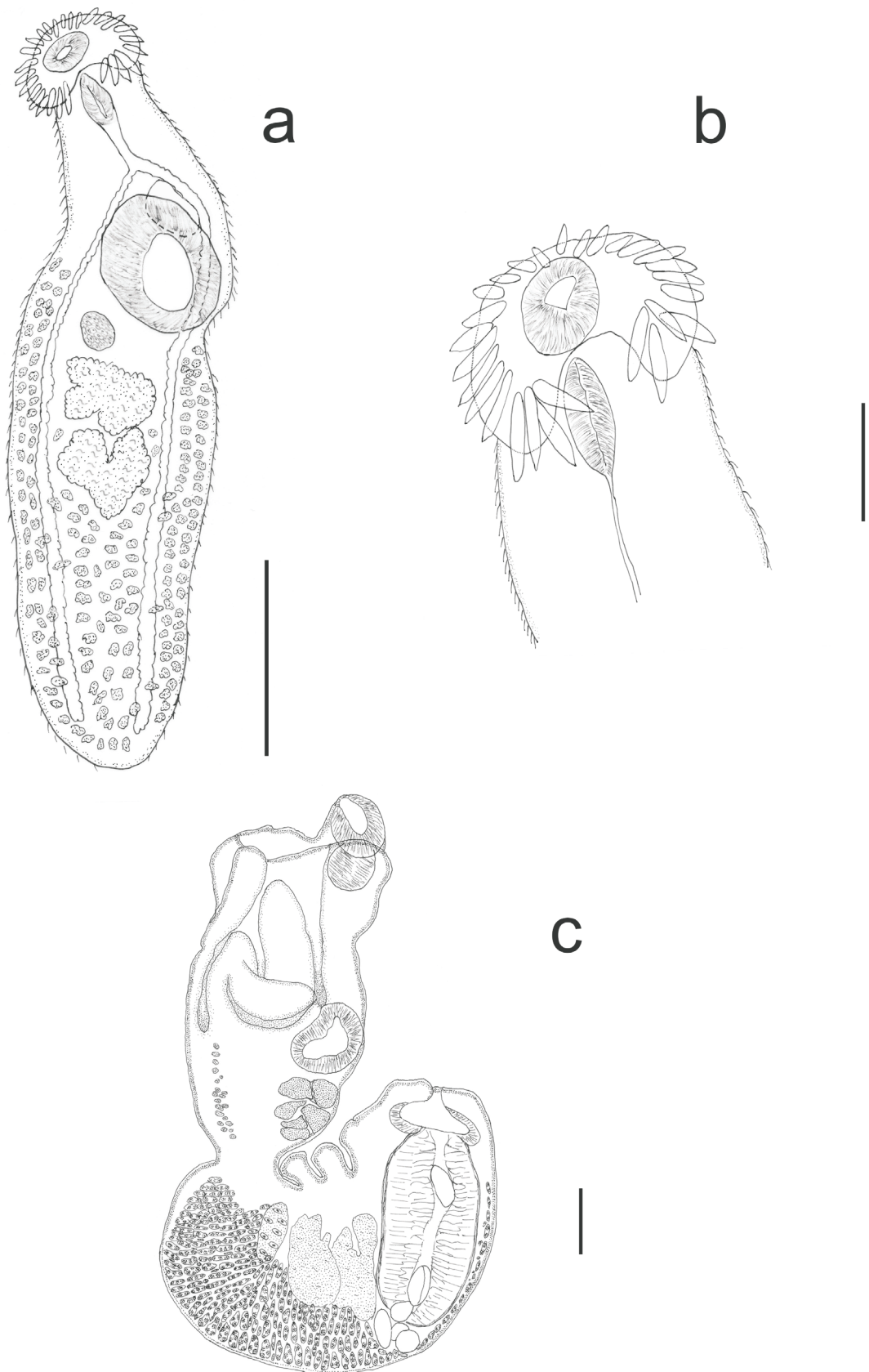


Figure 1. a. *Petasiger segregatus* from *Cathartes burrovianus*, specimen *in toto*. Scale bar = 500 μ m; b. *Petasiger segregatus* from *Coragyps atratus*, anterior end. Scale bar = 200 μ m; c. *Strigea vaginata* from *Coragyps atratus*, specimen *in toto*. Scale bar = 200 μ m.

Therefore, the findings of *P. segregatus* in *C. atratus* and in *C. burrovianus* allow to infer that these birds have ingested fishes or amphibians some time before being caught.

The life cycle of *S. vaginata* is very complex, including mesocercariae in frogs and metacercariae in fish, snakes and mammals (Dubois, 1968). For example, the finding of *S. vaginata* in *A. brasiliensis* was considered accidental by Dubois (1968) and Lunaschi and Drago (2013), because these anatids often eat fruits, roots, and some invertebrates such as insects and molluscs (De la Peña, 2019b). However, the rest of the birds mentioned above as hosts of *S. vaginata*, prey on a wide variety of potential intermediate hosts. Therefore, the broad range of intermediate hosts of *S. vaginata* allows birds with different feeding habits to acquire this parasite. In this context, this strigeid species would not be a good indicator of the diet of its definitive hosts.

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