

Research article

Parasitism in the armadillo *Zaedyus pichiy* (Cingulata: Chlamyphoridae) from San Juan Province, Argentina

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Abstract: The objective of this study was to describe parasitism in the piche armadillo *Zaedyus pichiy* (Desmarest, 1804) from San Juan Province, Argentina. An adult male *Z. pichiy* found run over on provincial route 510 (province of San Juan, Argentina) was analysed. The armadillo was examined externally in search of ectoparasites. Subsequently, the gastrointestinal contents were extracted and transported to the laboratory for endoparasite search. The presence of the tick *Amblyomma pseudoconcolor* (Acari: Ixodidae) and nematodes *Cyclobulura superinae* (Spirurida: Subuluridae), *Aspidodera scoleciformis*, and *Aspidodera fasciata* (Spirurida: Aspidoderidae) was determined. Our work provides new information on parasitism in this armadillo species in San Juan, Argentina.

Keywords: *Amblyomma pseudoconcolor*, *Aspidodera fasciata*, *Aspidodera scoleciformis*, *Cyclobulura superinae*, nematode, tick

Introduction

Xenarthrans are mammals almost exclusively distributed in the Neotropical Region, except for *Dasypus novemcinctus* Linnaeus, 1758, which ranges as far as the central United States (Nigro et al., 2021). In Argentina, two orders are represented, totalling 17 species: order Pilosa, with three species of terrestrial habits, arboreal and adapted to myrmecophagy, and order Cingulata, known as armadillos, equipped with a protective shell or bony armour, comprising 14 species (Nigro et al., 2021; Bauni et al., 2021). Within armadillos, there are two families, Dasypodidae with 4 species and Chlamyphoridae with 10 species (Bauni et al., 2021).

The genus *Zaedyus* Ameghino, 1889, is characterised by its relatively small body and ears (Nigro et al., 2021). The armadillo *Zaedyus pichiy* (Desmarest, 1804) (Chlamyphoridae) is distributed in Argentina and Chile (Superina & Abba, 2014). In Argentina, *Z. pichiy* is distributed across Buenos Aires, Chubut, Córdoba, La Pampa, La Rioja, Mendoza, Neuquén, Río Negro, San Juan, San Luis, and Santa Cruz provinces (Bauni et al., 2021). This species inhabits arid or semi-arid environments in firm and sandy soils. Is carnivorous-omnivorous, with a preference for carrion, but also consumes ants, beetles, scorpions, spiders, dipteran larvae, and vertebrates such as lizards and small mammals (Superina & Abba, 2014).

Information on parasite diversity in *Xenarthra* is represented in southern, western, and eastern Argentina (Daciuk et al., 1981; Ezquiaga et al., 2009; Navone et al., 2010; Monferrán & Silverio-Reyes, 2014; Ezquiaga et al., 2019; Ezquiaga et al., 2021; Gallo et al., 2021), mentioning nematodes of the Aspidoderidae and Subuluridae families (Ezquiaga et al., 2019) and ectoparasites, mainly Ixodidae ticks of the genus *Amblyomma* Koch, 1844 (Nava et al., 2017).

Nematodes of the Aspidoderidae family are parasites mainly of xenarthrans, didelphimorphs, hystriognathous and sigmodontine rodents (Santos et al., 1990; Anderson et al., 2009). The genus *Aspidodera* Railliet & Henry, 1912 includes 12 species: *Aspidodera scoleciformis* (Diesing, 1851); *Aspidodera subulata* (Molin, 1860); *Aspidodera fasciata* (Schneider, 1866); *Aspidodera binansata* Railliet & Henry, 1913; *Aspidodera raillieti* Travassos, 1913; *Aspidodera ansirupta* Proença, 1937; *Aspidodera vazi* Proença, 1937; *Aspidodera lacombeae* Vicente, 1964; *Aspidodera soganderesei* Jiménez-Ruiz, Gardner & Varela-Stokes, 2006; *Aspidodera kinsellai* Jiménez, Carreno, & Gardner, 2013; *Aspidodera lanfredi* Chagas-Moutinho, Sant'Anna, Oliveira-Mendes & De Souza, 2014 and *Aspidodera serrata* Cesário, Gomes, Maldonado, Olifiers, Jiménez & Bianchi, 2021 (Cesário et al., 2021).

The nematodes of the Subuluridae family include several genera, one of which is *Cyclobulura* Quentin, 1977, parasites of xenarthrans (Quentin, 1977; Anderson et al., 2009) with two species, *Cyclobulura lainsoni* Quentin, 1977, and *Cyclobulura superinae* Navone, Ezquiaga & Jiménez, 2010 (Anderson et al., 2009; Navone et al., 2010).

Our objective is to report the first record of parasitism by both ecto and endoparasites in the armadillo *Z. pichiy* from San Juan Province, Argentina.

Materials and methods

In April 2023, a male armadillo *Z. pichiy* was examined externally and internally. The specimen was found dead, run over on provincial route 510 (31°17'2.1"S; 67°13'45.7"W) 25 km from the town of Chucuma, Valle Fértil Department, San Juan Province, Argentina. An external examination was carried out; a male tick in the adult stage was collected and stored in

70° alcohol. The gastrointestinal contents were extracted and transported in containers with 70° alcohol to the laboratory to search for endoparasites. In the laboratory, the intestine and stomach were analysed for nematodes using a binocular stereoscopic microscope. The nematodes were preserved in 70° ethyl alcohol in Eppendorf tubes. For the observation of the nematodes, semi-permanent preparations were made using lactophenol solution. The identification was carried out using a light microscope (Arcano Xsp optical binocular). The tick was observed by direct observation under a magnifying glass. For tick identification, was followed Nava et al. (2017), and nematode identification followed Navone et al. (2010) and Cesário et al. (2021). Drawings were made using a camera lucida attached to a light microscope. Measurements are in μm , with mean ± 1 SD and range in parentheses unless otherwise noted.

The specimens were deposited in the Parasitological Collection of the Center for Wildlife Rehabilitation, Environmental Education and Responsible Recreation (Parque Faunístico) (*Amblyomma pseudoconcolor*: CRFSJ_59A; *Aspidodera scoleciformis*: CRFSJ_59B; *Aspidodera fasciata* CRFSJ_59C; *Cyclobulura superinae* CRFSJ_59D).

Ethical aspects

All applicable international, national, and/or institutional guidelines for the care and use of animals were strictly followed. All sample collection protocols complied with current Argentine laws. All animal procedures performed in this research were carried out in accordance with the ethical standards of the institution or organisation where the study was conducted.

Results

One adult male tick of *Amblyomma pseudoconcolor* Aragão, 1908 (Acari: Ixodidae) was recorded. The characteristics that allowed it to be identified are: body outline oval rounded, scapulae rounded; cervical grooves short, comma-shaped; marginal groove complete. Scutum ornate, with small pale spots adjacent to the marginal groove; Genital aperture located at level of coxa II, U-shaped. Legs:

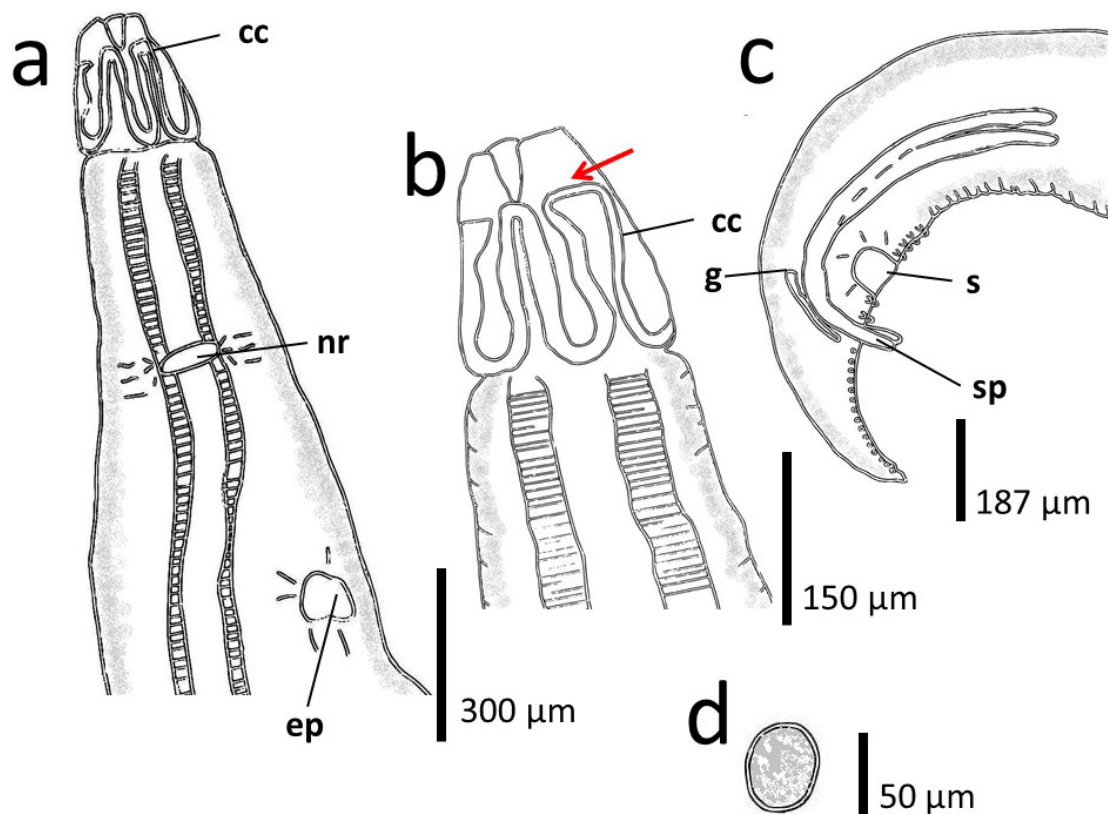


Fig. 1. *Aspidodera scoleciformis*. (a); female, anterior extremity, lateral view, (b); males, anterior extremity, lateral view, (c); males, posterior extremity, lateral view, (d); eggs. Cephalic cords = cc, nerve ring = nr, excretory pore = ep, sucker = s, spicule = sp, gubernaculum = g. Detail of the square-shaped cephalic cord at the labial root (red arrow).

coxa I with two distinct, short, triangular spurs, subequal in size; coxae II-IV with a triangular spur each; trochanters with spurs. Spiracular plates comma-shaped.

In the large intestine, 5 female and 2 male nematodes of *Aspidodera scoleciformis* and 6 females and 3 males of *Aspidodera fasciata* were recorded. In addition, 8 females and 2 males of *Cyclobulura superinae* in the small intestine were recorded.

Nematoda Rudolphi, 1808

Spirurida Railliet, 1915

Aspidoderidae Freitas, 1956

Aspidodera Railliet & Henry, 1912

Aspidodera scoleciformis (Diesing, 1851) (Fig. 1)

General: They present an expansion called cofia, which has cephalic cords with recurrent ramifications.

Cephalic cords present in a square shape at the labial root. Mouth with three lips. Esophagus with posterior bulb. The nerve ring and excretory pore are located in the middle of the esophagus. Females with the vulva located in the anterior third, approaching the middle of the body. Males without absent caudal alae.

Males (Based on 2 specimens) (µm): Total length 6693 ± 4.2 (6690–6696), maximum width 407 ± 1.4 (406–408), esophagus length (excluding bulb) 1473 ± 4.2 (1470–1476), esophagus width 76.5 ± 2.1 (75–78), bulb length 294 ± 1.4 (293–295), bulb width 316 ± 1.4 (315–317), cord length 61 ± 1.4 (60–62), distance to excretory pore from anterior region 831.5 ± 2.1 (830–833). Spicules of equal size, 752 ± 2.8 (750–754), distance to sucker from anterior region 6240 ± 2.1 (6239–6242), sucker length 86.5 ± 2.1 (85–88), sucker width 57.5 ± 0.7 (57–58), gubernaculum length 195.5 ± 0.7 (195–196). Presence of cloacal papillae: 4 precloacal before the sucker, 2 pairs posterior to the sucker and between 15 and 23 postcloacal pairs.

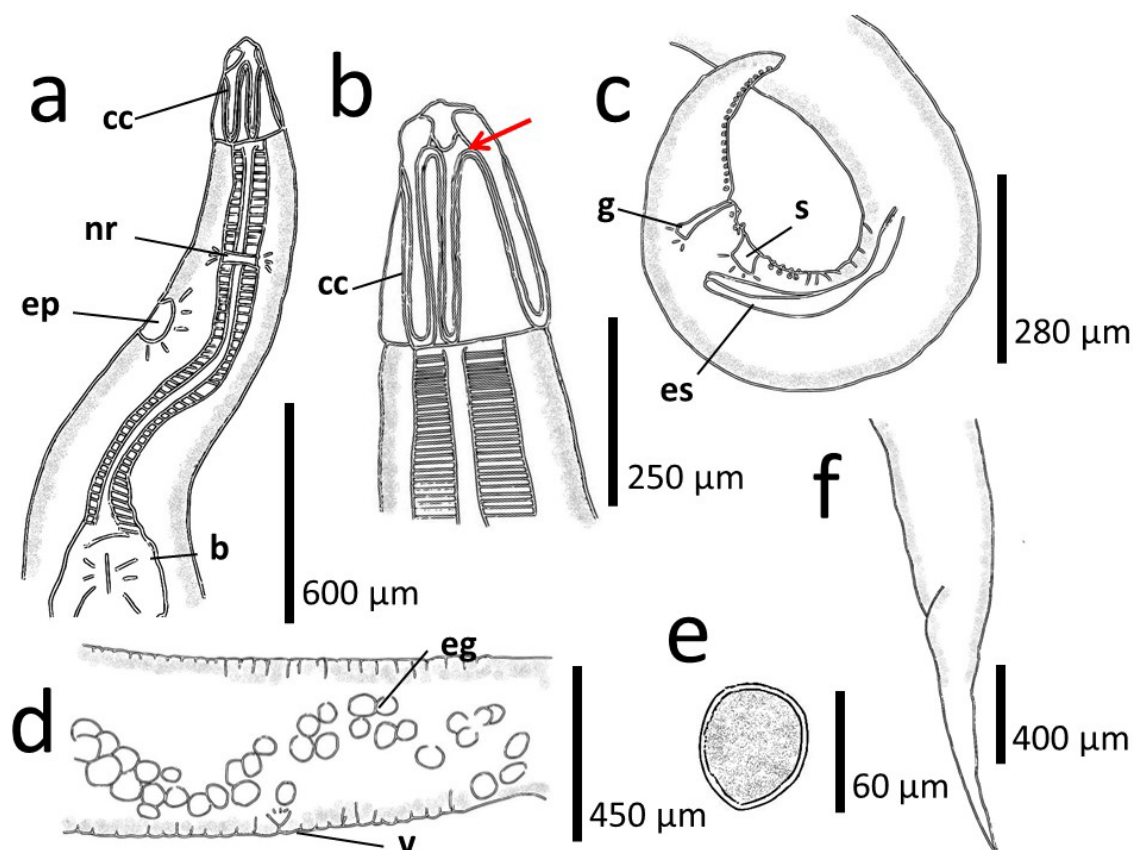


Fig. 2. *Aspidodera fasciata*. (a); female, anterior extremity, lateral view, (b); males, anterior extremity, lateral view, (c); males, posterior extremity, lateral view, (d); female with eggs, medium extremity, lateral view, (e); eggs, (f); female, posterior extremity, lateral view. The cephalic cords are observed = cc, nerve ring = nr, excretory pore = ep, bulb = b, vulva = v, eggs = eg, gubernaculum = g, sucker = s, spicule = sp. Detail of the round-shaped cephalic cord at the labial root (red arrow).

Females (Based on 2 specimens) (μm): Total length 6813 ± 4.2 (6810–6816), total width 521 ± 1.4 (520–522), esophagus length (excluding bulb) 1702 ± 2.8 (1700–1704), esophagus width 66.5 ± 2.1 (65–68), bulb length 339 ± 1.4 (338–340), bulb width 339 ± 1.4 (338–340), distance to nerve ring from anterior region 321.5 ± 2.1 (320–323), cord length 149.5 ± 0.7 (149–150), distance to vulva from anterior region 3292 ± 2.8 (3290–3294), egg width 50 (50) and egg length 50 (50).

Aspidodera fasciata (Schneider, 1866) (Fig. 2)

General: They present an expansion called cofia, which has cephalic cords with recurrent ramifications. Cephalic cords present in a round shape at the labial root. Mouth with three lips. Esophagus with posterior bulb. The nerve ring and excretory pore are located in

the middle of the esophagus. Females with the vulva located in the anterior third, approaching the middle of the body. Males with caudal wings absent.

Males (Based on 2 specimens) (μm): Total length 4882 ± 2.83 (4880–4884), total width 341 ± 2.1 (340–343), esophagus length (excluding bulb) 1178 ± 2.1 (1177–1180), esophagus width 48 ± 0.7 (48–49), bulb length 195.5 ± 0.7 (195–196), bulb width 213.5 ± 2.1 (212–215), distance to nerve ring from anterior region 732 ± 3.5 (730–735), cord length 212 (212), distance to excretory pore from anterior region 831 ± 2.1 (830–833). Spicules of equal size, 309 ± 1.4 (308–310), distance to sucker from anterior region 6012 ± 3.5 (6010–6015), sucker length 97 ± 0.7 (97–98), sucker width 48 ± 0.7 (48–49), gubernaculum size 136 ± 1.4 (135–137). Caudal papillae numerous and sessile. Presence of cloacal papillae: 4 precloacal before the sucker, 2 pairs posterior to the sucker and between 20 and 23 postcloacal pairs.

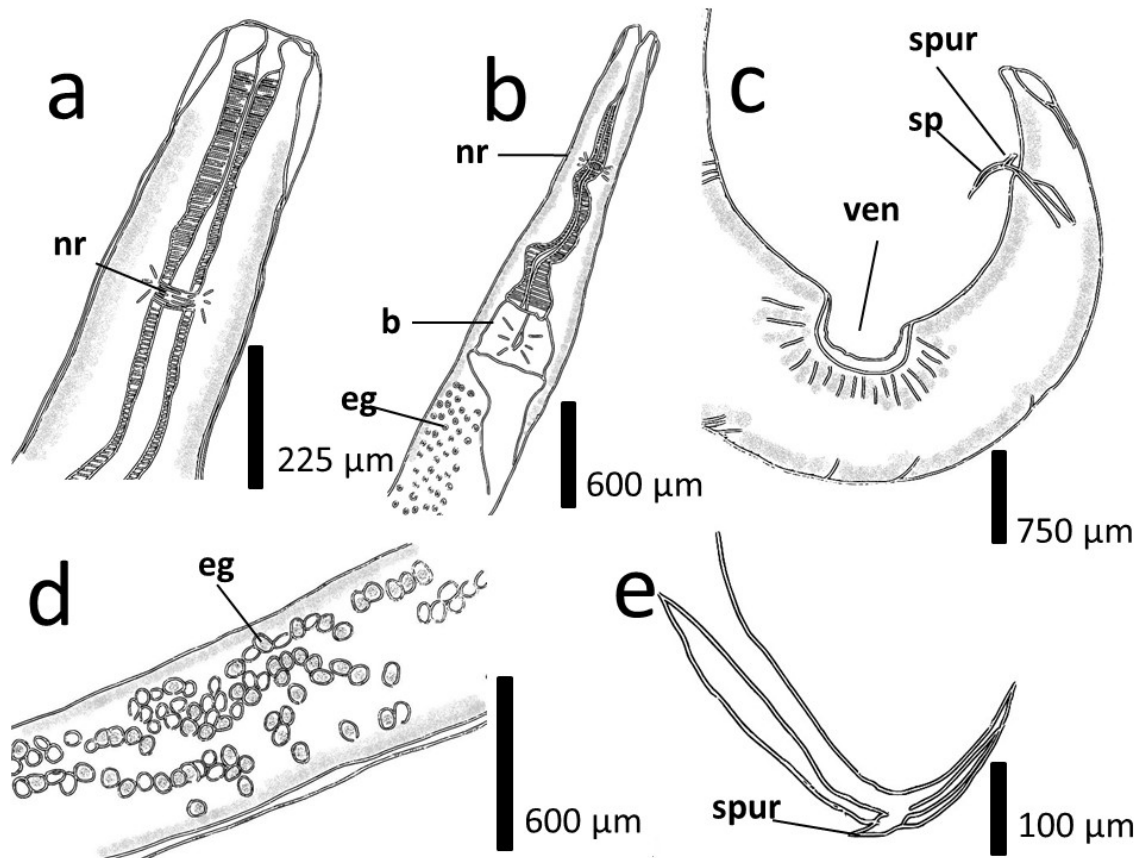


Fig. 3. *Cyclobulura superinae*. (a, b); female, anterior extremity, lateral view, (c); male, posterior extremity, lateral view, (d); female with eggs, medium extremity, (e); spicule and spur. Nerve ring = nr, bulb = b, eggs = eg. Sucker = s, spicule = sp.

Females (Based on 2 specimens) (μm): Total length 6701 ± 1.4 (6700–6702), total width 452 ± 2.8 (450–454), esophagus length (excluding bulb) 1244 ± 6.36 (1240–1249), esophagus width 44 ± 1.4 (43–45), bulb length 223 ± 4.9 (220–227), bulb width 271 ± 1.4 (270–272), cord length 249.5 ± 0.7 (249–250), distance to excretory pore from anterior region 732.5 ± 3.5 (730–735), distance to vulva from anterior region 2724 ± 2.1 (2723–2726), egg width 74.4 ± 0.7 (74–75) and egg length 61.4 ± 0.7 (61–62).

Subuluridae Yorke & Maplestone, 1926

Cyclobulura superinae Navone, Ezquiaga, Notarnicola & Jiménez, 2010 (Fig. 3)

General: White nematodes, buccal capsule divided into an anterior buccal part and a posterior pharyngeal part. Small buccal cavity, triangular in cross-section.

Simple oral opening without lip lobes. Present spurs on the spicules.

Males (Based on 2 specimens) (μm): Total length 11325 ± 35.3 (11300–11350), total width 534.5 ± 6.3 (530–539), esophagus length (excluding bulb) 1700 ± 1.4 (1700–1702), esophagus width 116 ± 1.4 (115–117), bulb length 339 ± 1.4 (338–340), bulb width 247 ± 2.8 (245–249), distance to nerve ring from anterior region 452 ± 2.8 (450–454), distance to excretory pore from anterior region 683 ± 4.2 (680–686). Spicules of equal size, spicule size 1073 ± 5.6 (1070–1078), spur size 49.4 ± 0.7 (49–50), distance to sucker from anterior region 10664 ± 6.3 (10660–10669), sucker width 97.4 ± 0.7 (97–98), sucker length 195 ± 0.7 (195–196). Presence of caudal papillae: 1 pair in the middle sector where the sucker is located, 3 precloacal and 4 postcloacal pairs.

Females (Based on 2 specimens) (μm): Total length 13823.5 ± 33.5 (13800–13847), total width 606.5 ± 9.1 (600–613), esophagus length (excluding bulb) 1915.5 ± 22 (1900–1931), esophagus width 87.5

± 3.5 (85–90), bulb length 337.5 ± 3.5 (335–340), bulb width 271 ± 1.4 (270–272), distance to nerve ring from anterior region 452 ± 2.8 (450–454), distance to vulva from anterior region 10318.5 ± 26 (10300–10337), egg width 54.5 ± 0.7 (54–55) and egg length 66 ± 1.4 (65–67).

Discussion

In the province of San Juan, 5 species of Cingulata armadillos are registered (Bauni et al., 2021): *Chaetophractus vellerosus* (Gray, 1865), *Chaetophractus villosus* (Desmarest, 1804), *Chlamyphorus truncatus* Harlan, 1825, *Tolypeutes matacus* (Desmarest, 1804), and *Z. pichiy*. So far, only the armadillo *C. vellerosus* has been studied in San Juan, reporting the nematodes *A. scoleciformis* and *C. superinae* (Ezquiaga et al., 2019).

In the present study, we analyse the parasitic fauna of *Z. pichiy* recorded in the locality of Valle Fértil. Our results from the analysis of this host indicated the presence of three species of nematodes: *C. superinae*, *A. scoleciformis* and *A. fasciata*. The analysis of a unique *Z. pichiy* specimen impedes the obtaining of conclusions on the ecological parasitic indexes.

The literature indicates that *C. villosus* has been studied in the provinces of Chubut, Santa Cruz, Tierra del Fuego, and La Rioja, where *C. superinae*, *A. fasciata*, and *A. scoleciformis* have been recorded (Navone et al., 2010; Gallo et al., 2021). *Chaetophractus vellerosus* has been studied in Buenos Aires, Córdoba, La Rioja, Mendoza, San Juan, and San Luis, with reports of *A. scoleciformis* and *C. superinae* (Ezquiaga et al., 2019). In *Z. pichiy*, *C. superinae*, *Aspidodera* sp., *A. fasciata*, and *A. scoleciformis* have been recorded in Mendoza, Chubut, Santa Cruz, and Tierra del Fuego provinces (Ezquiaga et al., 2009; Navone et al., 2010; Gallo et al., 2021). Lastly, in *T. matacus*, *A. scoleciformis* and *A. fasciata* were recorded in Catamarca (Monferrán & Silverio-Reyes, 2014).

These antecedents allow our work to be the first record on the parasitism of *Z. pichiy* in the province of San Juan, Argentina, recording the nematodes *C. superinae*, *A. scoleciformis*, and *A. fasciata*.

Regarding ectoparasites recorded in armadillos from Argentina, antecedents mention records in Patagonia, Argentina, reporting in *C. villosus* the ticks

A. pseudoconcolor and *Amblyomma auricularium* (Conil, 1878) (Daciuk et al., 1981; Guglielmone & Nava, 2006; Ezquiaga et al., 2021) and in *Z. pichiy* also from Patagonia, Argentina *A. pseudoconcolor* (Gallo et al., 2021). Due to this background, our results mention the first record of the tick *A. pseudoconcolor* in *Z. pichiy* for San Juan, Argentina.

Later studies in more armadillos will allow us to elucidate the parasitic ecological data and thus be able to compare with other regions of Argentina.

Conflicts of interest

No potential conflict of interest has been reported by the authors, reviewers, or subject editor.

Author's contributions

(using [CrediT](#) )

Gabriel N. Castillo: conceptualisation, data curation, formal analysis, investigation, methodology, project administration, resources, software, supervision, validation, visualisation, writing—original draft, writing—review and editing; Cynthia J. González-Rivas: data curation, formal analysis, investigation, supervision, validation, writing—original draft, writing—review and editing; Lucas C. Zúñiga: funding acquisition, writing—original draft; Ruben Fernandez-Reinoso: funding acquisition, writing—original draft; Leandro F. Castillo: funding acquisition, writing—original draft; Rodolfo O. Noriega: funding acquisition, writing—original draft.

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