Two new and two redescribed species of *Anonchotaenia* (Cestoda: Paruterinidae) from South American birds

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Abstract: Morphological examination of novel specimens of paruterinid cestodes from passerine birds from Brazil and Chile and of museum specimens from Paraguay revealed two new species: *Anonchotaenia prolixa* sp. n. from *Elaenia albiceps chilensis* Hellmayr from Chile, and *Anonchotaenia vaslata* sp. n. from *Tyrannus melancholicus* (Vieillot) (type host) and *Myiodynastes maculatus* (Statius Muller) from Paraguay. The generic diagnosis of *Anonchotaenia* Conn, 1900 is amended, prompted by the presence of the armed cirrus and the elongated cirrus sac of *A. prolixa*. Two species were redescribed: *Anonchotaenia brasiliensis* Fuhrmann, 1908 from *Tachyphonus coronatus* (Vieillot) and *Thraupis cyanoptera* (Vieillot) (new host records) from Brazil, and *Thraupis sayaca* (Linnaeus) and *Volatinia jacarina* (Linnaeus) from Paraguay (new host and geographic records); and *Anonchotaenia macrocephala* Fuhrmann, 1908 from *Tachycineta leucorrhoa* (Vieillot) (new host record) from Brazil, *Tachycineta meyeni* (Cabanis) from Chile (new host and geographic record) and *Stelgidopteryx ruficollis* (Vieillot) from Paraguay (new host and geographic record). Scanning electron microscopy of *A. brasiliensis* and *A. macrocephala* revealed less microtrix variation than has been reported for other cyclophyllidean taxa. Sequence data were generated for nuclear *ssr*- and *lsr*-DNA and mitochondrial *rrnL* and *cox1* for *A. prolixa*, *A. brasiliensis*, and *A. macrocephala*. Maximum likelihood and Bayesian inference analyses supported each species as distinct, but revealed cryptic diversity among *A. brasiliensis* specimens from different host families. New host records of *A. brasiliensis* and *A. macrocephala* prompted a formal assessment of host specificity. *Anonchotaenia prolixa* was found to be oioxenous ($HS_s = 0$), *A. vaslata* and *A. macrocephala* were found to be metastenoxenous ($HS_s = 3.000$ and 3.302 , respectively), whereas *A. brasiliensis* was found to be euryxenous ($HS_s = 5.876$). *Anonchotaenia brasiliensis* has been found parasitising several species of different passerine families that participate in mixed-species foraging flocks in the Atlantic Forest. A diversity of species of other families join these flocks and are among the substantial number of South American passerine species yet to be examined for cestodes.

Keywords: *Anonchotaenia prolixa*, *Anonchotaenia vaslata*, *Anonchotaenia brasiliensis*, *Anonchotaenia macrocephala*, new species, new host record, Brazil, Chile, Paraguay, new geographical record

This article contains supporting information (Table S1, S2) online at <http://folia.paru.cas.cz/suppl/2014-5-441.pdf>

Anonchotaenia Cohn, 1900 is one of 22 genera of Paruterinidae that is characterised by an unarmed scolex, short, wide proglottides, vermiform oncospheres and a uterus without septa (Georgiev and Kornyshev 1994, Phillips et al. 2012). It contains 27 species that parasitise a wide diversity of passeriform hosts and has a cosmopolitan distribution (Voge and Davis 1953, Dollfus 1959, Singal 1963, Ulmer and James 1976, Olsen et al. 1978, Shinde 1984, Schmidt 1986, Sharma and Mathur 1987, Mariaux 1991). The latest contributions to the systematics of *Anonchotaenia* include erection of the subgenus *Anonchotaenia* (*Paranonchotaenia*) Mariaux, 1991 and the description of

two new species from the Ivory Coast (Mariaux 1991). Most recently, sperm ultrastructure and spermiogenesis was characterised in *Anonchotaenia globata* (von Linstow, 1879) by Yoneva et al. (2010). Species delimitation within *Anonchotaenia* is challenging because the few available morphological characters are easily obscured or distorted in contracted material. Testis number per proglottis historically has been considered the most reliable character for delimiting species of *Anonchotaenia*, although this character is known to vary between conspecifics and even within an individual's strobila (Mariaux 1991). The addition of molecular data to phylogenetic analyses has been

suggested to clarify species boundaries within the genus (Mariaux 1991).

South American *Anonchotaenia* species are poorly known, with only six species in total reported from the continent and all but two of these species described with brief treatments prior to 1909 (Fuhrmann 1901, 1908). As part of the present study, fieldwork conducted near the Comau Fjord, Chile in 2008 and near São Paulo, Brazil in 2011 led to the collection of cestode specimens from a diversity of avian orders, but predominantly Passeriformes. Examination of this material combined with the study of museum specimens resulted in the description of two new species of *Anonchotaenia* as well as the redescription of *Anonchotaenia brasiliensis* Fuhrmann, 1908 and *Anonchotaenia macrocephala* Fuhrmann, 1908. *Anonchotaenia brasiliensis* and *A. macrocephala* are the first paruterinid taxa to be examined with scanning electron microscopy (SEM). Sequences from the newly collected specimens are some of the first molecular sequences of paruterinids and were the basis for phylogenetic analyses. Host specificity had not been previously measured for members of Paruterinidae prompting the first formal assessment of host specificity for the family to assess trends of host spectrum breadth, overlap of host spectrum and host specificity in comparison to other cestode groups.

MATERIALS AND METHODS

Specimen sampling and abbreviations

Cestodes studied included newly collected material from Brazil and Chile, which were obtained as part of a collaborative effort focused on a global assessment of cestode diversity funded by the National Science Foundation (NSF) Planetary Biodiversity Inventory (PBI) project 'A Survey of the Tapeworms (Cestoda: Platyhelminthes) from Vertebrate Bowels of the Earth' (NSF PBI award Nos. 0818696 and 0818823). In Brazil, birds were sampled at two localities. At Estação Biológica de Boracéia, Salesópolis, São Paulo, Brazil (23°38'S; 45°52'W, elevation 946 m) during 24–28 November 2011, 164 adult birds, including 46 passerine species and one species each from Columbiformes, Piciformes and Trogoniformes, were captured by mist netting in semi-deciduous forest. At Fazenda Nabor, Tremembé, São Paulo, Brazil (22°55.45'S; 45°34.25'W, elevation 540 m) during 30 November–3 December 2011, 47 adult birds, including 14 species of Passeriformes, six of Pelecaniformes, three of Cuculiformes, two of Accipitriformes, two of Columbiformes, two of Piciformes and one species each of Charadriiformes, Anseriformes, Psittaciformes and Strigiformes, were collected by shooting in rice fields and surrounding shrublands.

At both localities, bird specimens were identified by skilled ornithologists. Bird specimens collected at Estação Biológica de Boracéia were deposited at the Museu de Zoologia Universidade de São Paulo, São Paulo, Brazil and bird specimens collected from Fazenda Nabor were deposited at the Museu Nacional Rio de Janeiro, Brazil. In Chile, 87 birds, including 19 species from 15 families were captured by mist netting and shooting at Huinay Station, Comau Fjord, Los Lagos Region, Chile (42°22'47"S; 72°22'54"W, elevation ca 20 m). Avian no-

menclature follows the IOC World Bird Names (Gill and Donker 2012).

Museum abbreviations used here are: MHNG-PLAT – Natural History Museum of Geneva, Geneva, Switzerland; MZUSP – Museu de Zoologia Universidade de São Paulo, São Paulo, Brazil; MNRJ – Museu Nacional Rio de Janeiro, Brazil; UFRJ – Universidade Federal do Rio de Janeiro, Brazil; USNPC – United States National Parasite Collection, Smithsonian's National Museum of Natural History, Washington, D.C.. Specimens collected through the PBI project are indicated by the number assigned specimens (PBI-_) and are listed in the Global Cestode Database (tapewormdb.uconn.edu). Collectors' authorities for specimens obtained during the Expédition du Muséum de Genève au Paraguay are given as EMGP.

Specimen preparation and morphological examination

Newly collected cestodes underwent different preparations: a subset of worms had the two to three most terminal proglottides removed and fixed in 95% ethanol for molecular work and the remaining complete worms and fragments were fixed in hot 5% formalin and subsequently transferred to 70% ethanol for storage. For light microscopy, a subset of worms were stained with hydrochloric carmine, dehydrated in an ascending ethanol series, cleared in methyl-salicylate, and mounted in Canada balsam for morphological examination.

Measurements were taken using an optical retical on a Nikon Eclipse 80i microscope. Measurements are given in micrometres (µm) unless otherwise stated. Measurements of the testes, cirrus sac, vitellarium and ovary were taken only from fully developed mature and early pregravid proglottides. Extent of the cirrus sac is reported as the percentage the cirrus sac that extends across the proglottis width. Metrical and meristic data are presented as the range, followed by the mean, standard deviation and the number of measurements or counts taken (n) in parentheses. Illustrations were made with the aid of a drawing tube and Adobe Illustrator® and Adobe Photoshop®. The terms used for the developmental stages of proglottides follow Georgiev and Vaucher (2001).

Scolecex of worms from *Tachyphonus coronatus* (Vieillot), *Tachycineta leucorrhoa* (Vieillot), *Thraupis cyanoptera* (Vieillot) and *Ammodramus humeralis* (Bosc) collected in Brazil in 2011 were prepared for SEM with the following procedure: they were hydrated in a graded filtered ethanol series, post-fixed in 1.5% osmium tetroxide overnight, washed in distilled water, dehydrated in a graded filtered ethanol series, transferred to hexamethyldisilazane (Ted Pella, Inc., Redding, CA, USA) and allowed to air dry in a fume hood. The specimens were then mounted on aluminium stubs on double-sided adhesive carbon tabs (Ted Pella, Inc.). Specimens were sputter-coated with ca 35 nm of gold/palladium and examined with a LEO/Zeiss DSM982 (Zeiss, Oberkochen, Germany) digital field emission scanning electron microscope at the Electron Microscopy Laboratory at the University of Connecticut, Storrs, CT, USA. Scolecex examined with SEM were retained in the personal collection of Janine N. Caira (University of Connecticut).

During the present study, several specimens from passerine hosts in South America were determined to represent *Anonchotaenia* but could not be definitively identified to species (Table S1). These specimens have been included here because they represent new host records and additional diversity of *Anonchotaenia*.

Generation of nucleotide data

Sequence data for almost complete small subunit nuclear ribosomal RNA gene (*ssrDNA*) (= 18S rDNA), domains D1–D3 of large subunit nuclear ribosomal RNA gene (*lsrDNA*) (= 28S rDNA), partial large mitochondrial ribosomal RNA subunit (*rrnL*) (= 16S rDNA) and partial cytochrome *c* oxidase subunit I (*cox1*) were generated *de novo* for newly collected cestode specimens, i.e. two outgroup taxa and six *Anonchotaenia* representatives (GenBank accession numbers: KF685910–KF685939; Table S2). Tissue samples for DNA extraction consisted of two or three of the most terminal proglottides or a subset from the middle of the strobila of complete worms, while the scolex and remaining strobila of each worm were prepared as whole mounts as previously described. Voucher specimens were deposited in the MZUSP, MHNG and the USNPC. For details of genomic DNA extraction, PCR, sequencing, sequence editing and sequence identity verification, see Scholz et al. (2013).

Phylogenetic analyses

Outgroup selection consisted of *Biuterina* sp. (PBI-96 from host PBI-GAB-106 *Andropadus latirostris* Strickland from Franceville, Haut Ogooue, Gabon, 1°36'57"S; 13°34'55"E) and *Dictyterina cholodkowskii* (Skrjabin, 1914) (PBI-173 from host PBI-CHIN-049 *Lanius tephronotus* (Vigors) from Yuzhong, Gansu, China, 35°46'33"N; 104°02'27"E), two species of paruterinid genera that are characterised by the possession of hooks and therefore easily distinguished morphologically from the taxa of interest in this study and formed part of the sister group to the ingroup in a wider analysis of Cyclophyllidea of currently unpublished data.

Sequences were aligned using MAFFT v. 6.611b (Katoh et al. 2005) with 1 000 cycles of iterative refinement and the *genafpair* algorithm. The alignment was improved by eye using MacClade (Maddison and Maddison 2005). Positions that could not be unambiguously aligned were excluded from any subsequent analyses. Modeltest v. 3.7macX (Posada and Crandall 1998) was used to select models of evolution using the Akaike Information Criterion. Phylogenetic analyses were conducted using two approaches: Maximum likelihood (ML) and Bayesian inference (BI). Bayesian Inference trees were constructed using MrBayes v. 3.2.1 (Huelsenbeck and Ronquist 2001). Likelihood settings for *ssrDNA* and *lsrDNA* were set to nst = 6, rates = inv, and for *rrnL* and *cox1* were set to nst = 6, rates = gamma, equivalent to the GTR+I and GTR+G models, respectively. Parameters were estimated separately for each partition. Four chains (heated chains temp = 0.2) were run for 5 000 000 generations and sampled every 1 000 generations. A total of 500 000 generations were discarded as burn-in, at which point the average standard deviation of split frequencies was < 0.01. The frequency with which a particular clade was present in the posterior distribution is represented by the clade's posterior probability (pp).

Maximum likelihood analyses were performed in PAUP* v. 4b10 (Swofford 2003) using successive approximation: model parameters were estimated based on a starting tree determined by neighbour-joining. A heuristic search was performed implementing the estimated model parameters using nearest-neighbour-interchange branch swapping. Model parameters were estimated on the best tree and a heuristic search performed using subtree-pruning-regrafting branch swapping implementing the estimated model parameters. After estimating model parameters, heuristic searches using tree-bisection-reconnection branch swapping were performed until the topology remained unchanged. Un-

corrected 'p' distances were calculated for *rrnL* and *cox1* gene fragments using Geneious Pro v. 5.6.4 created by Biomatters and available at <http://www.geneious.com>.

Measurement of host specificity

Host specificity at the species level was assessed using the Index of Phylogenetic Host Specificity (Caira et al. 2003) and standardised terminology for categories of host specificity proposed by Caira et al. (2003) was followed.

RESULTS

Paruterinidae Fuhrmann, 1907

Genus *Anonchotaenia* Cohn, 1900

Anonchotaenia (*Paranonchotaenia*) *prolixa* sp. n.

Figs. 1A, 2

Description (based on one complete worm and one fragment of strobila): Body ribbon-like, up to 28 mm (n = 1). Maximum width 700 (n = 2), at level of early gravid proglottides (Fig. 1A). Mature proglottides acraspedote, 4 to 8 times wider than long; pregravid and gravid proglottides slightly craspedote, as wide as long. Scolex rounded, 565 (n = 1) in diameter, with 4 muscular suckers, 225–250 (240 ± 12, n = 4) in diameter (Fig. 3A). First proglottides appear 0.78 mm (n = 1) from posterior margin of suckers. Mature proglottides appear 3.74 mm (n = 1) posterior to appearance of first proglottides (Fig. 1A). Genital pores irregularly alternating in short series (e.g. 2, 1, 2, 2, 1, 1, 2, 1, 2, 2, 1, 1, ...), opening equatorial along lateral edge of proglottis. Genital atrium with infundibular orifice, pyriform, narrow, deep. Genital ducts pass between osmoregulatory canals. Osmoregulatory canals classically disposed; dorsal osmoregulatory canals 7.5–22 (12 ± 7, n = 4) in diameter; ventral osmoregulatory canals 20–50 (32 ± 13, n = 4) in diameter, with transverse anastomoses along posterior margin of each proglottis.

Testes spherical, slightly oval to oval in contracted proglottides, 4–6 [4 (14%), 5 (66%), 6 (20%)] (5 ± 0.6, n = 59) in number, 28–58 (47 ± 10, n = 10) long, 40–53 (48 ± 4, n = 10) wide; arranged in single transverse dorsal row, occasionally overlapping osmoregulatory canals (Fig. 2B), persist in pregravid proglottides. Cirrus sac very large, claviform, thick-walled, muscular (Fig. 2C); 172–225 (192 ± 14, n = 31) long and 38–60 (47 ± 6, n = 31) wide in mature proglottides, 245–270 (255 ± 12, n = 5) long and 55–62.5 (59 ± 3, n = 5) wide in post-mature or early pregravid proglottides; crossing and surpassing osmoregulatory canals, reaching over one third of proglottis width, length extending 30–40% (36 ± 3%, n = 10) across width of proglottis. Cirrus cylindrical, finely and densely armed on its entire length with small spines approximately 3 in length, occasionally straight but usually with one or two loops at mid-length when invaginated (Fig. 2E). Internal vas deferens thin, making a few loops in proximal part of cirrus sac. External vas deferens coiled, ventral to cirrus sac.

Fig. 1. Drawings of whole worms. **A** – *Anonchotaenia prolixa* sp. n., holotype (MHNG-PLAT-64401), from *Elaenia albiceps chilensis*; **B** – *Anonchotaenia vaslata* sp. n., holotype (MHNG-PLAT 37770), from *Tyrannus melancholicus*. Abbreviations: PO – paruterine organ; VO – vermiform oncospheres.

Ovary oval, medial, 65–95 (79 ± 9 , $n = 10$) long, 58–103 (85 ± 13 , $n = 10$). Vitellarium compact, round, 48–65 (57 ± 6 , $n = 10$) long, 50–70 (59 ± 5 , $n = 10$), aporal to ovary. Mehlis' gland indistinct. Seminal receptacle fusiform, not clearly distinct from vagina. Vagina conspicuous, walls well-demarcated (Fig. 2C), opening posterior to male pore. Copulatory portion wide in mature

proglottides (Fig. 2B), decreasing in width in pregravid proglottides (Fig. 2D,F), tapering to conductive portion, passing posterior and parallel to cirrus sac, rarely crossing. Conductive portion making one or two very big loops along aporal end of cirrus sac.

Uterus circular to oval, sac-like, dorsal to ovary in post-mature and early pregravid proglottides, expanding to fill median field, usually not overlapping cirrus sac in pregravid proglottides (Fig. 2D,F). Uterine wall thickening as proglottis matures. Paruterine organ pyramidal, consisting of uniform fibrillar tissue, developing as thickening of anterior uterine wall, directed anteroposteriorly to uterus, appearing approximately 150–163 proglottides posterior to first mature proglottides (Fig. 1A). Eggs initially spherical. Oncospheres and embryophore transform into vermiform shape in pregravid proglottides approximately 40–42 proglottides posterior to appearance of paruterine organ (Fig. 1A), gradually filling uterus, eggs passing into paruterine cavity not observed (Fig. 2F). Embryonic hooks not observed.

Type and only host: *Elaenia albiceps chilensis* Hellmayr, white-crested elaenia (Passeriformes: Tyrannidae).

Type and only locality: Huinay Station, Comau Fjord, Los Lagos Region, Chile ($42^{\circ}22'47''S$; $72^{\circ}22'54''W$, 21 m).

Site of infection: Small intestine.

Prevalence and intensity of infection: 8.3% (1 host infected of 12 examined), 2 worms.

Type specimens: Holotype (MHNG-PLAT-64401); 1 paratype (MHNG-PLAT-64403).

Etymology: *prolixa* (Latin) = extended or elongated; referring to the unusually long cirrus sac.

Remarks. *Anonchotaenia* (*Paranonchotaenia*) *prolixa* sp. n. is unique from the other 27 species of *Anonchotaenia* (Table 1) in the possession of an armed cirrus and a cirrus sac that extends into the median field of the proglottis. The new species has genital ducts that pass between the osmoregulatory canals as in members of the subgenus *Anonchotaenia* (*Paranonchotaenia*), including *Anonchotaenia malaconoti* Mariaux, 1991, *Anonchotaenia prionopos* Mariaux, 1991 and *Anonchotaenia dendrocitta* (Woodland, 1929). *Anonchotaenia prolixa* sp. n. is unique from the *Anonchotaenia* species that possess fewer than 7 testes and for which the subgeneric position is unclear (Table 1): *Anonchotaenia prolixa* sp. n. possesses larger suckers (225–250 μm) than *Anonchotaenia mexicana* Voge et Davis, 1953 (121–198 μm), *Anonchotaenia ranae* (Ulmer et James, 1976) (138–175 μm) and *Anonchotaenia zonotrichicola* Dollfus, 1959 (170–200 μm), possesses a smaller scolex (565 μm in diameter) than *Anonchotaenia sbesteriometra* Joyeux et Baer, 1935 (740 μm), and a larger cirrus sac (172–225 $\times$ 38–60 μm) than *A. zonotrichicola* (46–54 $\times$ 22 μm) and *Anonchotaenia zanthopygiae* Yamaguti, 1956 (60–100 $\times$ 18–22 μm).

Fig. 2. *Anonchotaenia prolixa* sp. n., holotype (MHNG-PLAT-64401), from *Elaenia albiceps chilensis*. **A** – scolex; **B** – mature proglottides; **C** – terminal genital ducts; **D** – early pre-gravid proglottides; **E** – everted armed cirrus; **F** – late pre-gravid proglottides.

In light of the unique characters of *A. prolixa* sp. n., i.e. the possession of an armed cirrus and a cirrus sac that extends into the median field of the proglottis that differentiates this species from its congeners, the generic diagnosis proposed by Georgiev and Kornyshev (1994) was amended (as follows):

Genus *Anonchotaenia* Cohn, 1900

Amended diagnosis: Scolex without rostellum or rostellar hooks. Suckers muscular, deep. Mature proglottides considerably wider than long (seven to ten times or more), acraspedote; gravid proglottides with almost equal length and width, craspedote. Genital pores alternating. Genital ducts ventral to or between osmoregulatory canals.

Testes occupying all median field. Cirrus sac may reach osmoregulatory canals or cross into median field. Cirrus unarmed or exceptionally armed. Vitellarium compact, round, slightly poral, median or slightly aporal. Ovary compact, oval, poral. Seminal receptacle round or slightly oval, near point where osmoregulatory canals and genital ducts cross. Uterus spherical or oval. Developing paruterine organ aporal and dorsal to uterus; paruterine organ anterior to uterus in last gravid proglottides. Eggs with vermiform oncospheres. In Passeriformes (different families), sometimes (probably accidentally) in Apodiformes (Apodidae, Trochilidae), Ciconiiformes (Threskiornithidae) and amphibians. Cosmopolitan.

Table 1. Key characters of *Anonchotaenia* spp. (Cestoda: Paruterinidae).

Species	Authors	Locality	Host*	Scolex diameter	Sucker diameter	Position of GD**	Testis number ¹	Cirrus sac extent	Cirrus sac length (MP)	Cirrus sac width (MP)
<i>Anonchotaenia antirina</i> Singal, 1963	Singal (1963)	Delhi, India	<i>Passer domesticus indicus</i> Jardine et Selby (Passeridae)	490	195	ventral	4–5 (5)	NR	43–55	25–31
<i>Anonchotaenia arhyncha</i> Fuhrmann, 1918	Fuhrmann (1918)	New Caledonia	<i>Zosterops lateralis griseonota</i> Gray (Zosteropidae)	480	200–220	ventral	4–7	-	80–90	18
<i>Anonchotaenia brasiliensis</i> Fuhrmann, 1908	Present study	São Paulo, Brazil	<i>Tachyphonus coronatus</i> (Vieillot) (Thraupidae)	497–696 (628)	223–271 (247)	ventral	4–8 (6)	NR	62.5–89 (73)	20–30 (25)
<i>Anonchotaenia castellani</i> Fuhrmann et Baer, 1943	Fuhrmann and Baer (1943)	El Banno, Ethiopia	<i>Eurocephalus rueppeli rueppeli</i> Bonaparte (Laniidae)	600–700	280–300	ventral	9–10	-	103–115	25–28
<i>Anonchotaenia clelandi</i> (Johnston, 1912)	Georgiev (1992)	Sydney, Australia	<i>Zosterops carulescens</i> (Latham) (= <i>Zosterops lateralis carulescens</i>) (Zosteropidae)	424	172–185 (180)	ventral	3–7 (5)	R	52–58 (56)	25–30 (28)
<i>Anonchotaenia dendrocityta</i> (Woodland, 1929)	Southwell (1930)	India	* <i>Dendrocityta rufia</i> (Corvidae)	600–800	300	between	10–12	-	160	-
<i>Anonchotaenia gaugi</i> Singh, 1952	Singh (1952)	India	<i>Turdoides striata somervillei</i> (Sykes) (Leiothrichidae)	1250	330–360	-	12–13 (13)	-	129–155	39–45
<i>Anonchotaenia globata</i> (von Linstow, 1879)	von Linstow (1879)	Kaliningrad Zoo, Kaliningrad, Russia (formerly Königsberg, Germany)	<i>Parus major</i> Linnaeus (Paridae)	500–700	140–300	ventral	4–8, (4/5)	NR	60–128	18–36
<i>Anonchotaenia indica</i> Singh, 1964	Singh (1964)	Mukteswar-Kumaun, India	<i>Muscicapa sundara</i> Hodgson (= <i>Niltava sundara</i>) (Muscicapidae)	890–997 (995)	312–401 (372)	ventral	4	NR	110–150	26–36
<i>Anonchotaenia jeandorsti</i> Dollfus, 1959	Dollfus (1959)	Bagua Grande, Amazon, Peru	<i>Tyrannus melancholicus melancholicus</i> Vieillot (Tyrannidae)	810	380–395	ventral	5	-	110	28–30
<i>Anonchotaenia lambi</i> (Voge et Davis, 1953)	Georgiev and Korniyushin (1993)	Durango, Mexico	<i>Streptoprocne semicollaris</i> (De Saussure) (Tyrannus)	1120	353–365 (359)	ventral	5–8 (7)	-	102–118 (111)	23–29 (27)
<i>Anonchotaenia longiovata</i> Fuhrmann, 1901	Fuhrmann (1908)	South America	<i>Icterus cayennensis</i> Linnaeus (= <i>Icterus cayennensis</i>) (Icteridae), <i>Agelaius curaeus</i> (Molina) (= <i>Curaeus curaeus</i>) (Icteridae), <i>Loxops</i> sp. (Fringillidae), <i>Plegadis guarauna</i> (Vieillot) (= <i>Plegadis chihi</i>) (Threskiornithidae)	340–485	126–239	ventral	7–12 (8/9)	NR	67–96 (82)	23–29 (26)
<i>Anonchotaenia macrocephala</i> Fuhrmann, 1908	Present study	São Paulo, Brazil	<i>Tachycineta leucorrhoa</i> (Vieillot) (Hirundinidae)	994	412–428 (419)	ventral	4–8 (6)	R	104–178 (137)	25–34 (30)
<i>Anonchotaenia malaconoti</i> Mariaux, 1991	Mariaux (1991)	Ivory Coast	<i>Malaconotus blanchoti</i> Stephens (Malaconotidae)	805–903 (854)	307–366 (341)	between	5–10 (7/8)	NR	106–135 (121)	22–31 (27)

(continued)

Table 1. Continued.

Species	Authors	Locality	Host*	Scolex diameter	Sucker diameter	Position of GD**	Testis number ¹	Cirrus sac extent	Cirrus sac length (MP)	Cirrus sac width (MP)
<i>Anonchotaenia mexicana</i> Voge et Davis, 1953	Voge and Davis (1953)	Durango, Mexico	<i>Pipilo erythrophthalmus griseipigum</i> Van Rossem (= <i>Pipilo maculatus griseipigum</i>) (Emberizidae)	522–612 (578)	121–198	-	5–10 (7/8)	*	90–132 (101)	19–27 (23)
<i>Anonchotaenia oriolina</i> Cholodkovsky, 1906	Cholodkovsky (1906)	Russia	<i>Oriolus galbula</i> (Linnaeus) (= <i>Oriolus oriolus</i>) (Oriolidae)	600	-	-	15 or more	*	*	*
<i>Anonchotaenia piriformis</i> Fuhrmann, 1918	Fuhrmann (1918)	Canala, New Caledonia	<i>Pachycephala 'morariensis'</i> (Pachycephalidae)	-	-	-	12	*	90	16
<i>Anonchotaenia pritonopos</i> Mariaux, 1991	Mariaux (1991)	Ivory Coast	<i>Prionops plumata</i> (Shaw) (= <i>Prionops plumata</i>) (Prionopidae)	683–830 (750)	229–273 (254)	between	5–8 (6/7)	R	76–107 (91)	20–28 (24)
<i>Anonchotaenia prolifica</i> sp. n.	Present study	Comau Fjord, Chile	<i>Elaenia albiceps chilensis</i> Hellmayr (Tyrannidae)	565	225–250 (240)	between	4–6 (5)	C	172–225 (192)	38–60 (47)
<i>Anonchotaenia quisquali</i> Rausch et Morgan, 1947	Rausch and Morgan (1947)	Ohio, USA	<i>Quiscalus versicolor</i> Vieillot (= <i>Quiscalus quiscula versicolor</i>) (Icteridae)	580	200	ventral	9 (5 aporal)	*	120	45
<i>Anonchotaenia ranae</i> (Ulmer et James, 1976)	Ulmer and James (1976)	Iowa, USA	<i>Rana pipiens</i> (Schreber) (= <i>Lithobates pipiens</i>) (Ranidae: Anura)	320–380 (353)	138–175 (155)	-	3–10 (8)	NR	50–62.5 (60)	17.5–20 (20)
<i>Anonchotaenia sbesteriometra</i> Joyeux et Baer, 1935	Joyeux and Baer (1935)	Indochina	<i>Motacilla cinerea caspica</i> Gmelin (= <i>Motacilla cinerea cinerea caspica</i>) (Motacillidae)	740	240	-	4–6	*	108	18
<i>Anonchotaenia singhi</i> Shinde, 1984	Shinde (1984)	Maharashtra, India	<i>Passer domesticus</i> Linnaeus (Passeridae)	118–119	43–45	-	14–16	*	51–52	*
<i>Anonchotaenia terricoli</i> (Sharma, 1947)	Sharma (1947)	Jhushi, Allahabad, India	' <i>Turdoides terricolor terricolor</i> ' (Liothrichidae)	860–1204	240–258	-	12–16	*	125–155	28–36
<i>Anonchotaenia trochili</i> Fuhrmann, 1908	Fuhrmann (1908)	Brazil	<i>Eupetomena macrura</i> (Gmelin) (= <i>Eupetomena macroura</i>) (Trochilidae)	250	100	-	14	*	60	*
<i>Anonchotaenia vastata</i> sp. n.	Present study	Jeju, San Pedro, Paraguay	<i>Tyrannus melancholicus</i> Vieillot (Tyrannidae)	760–980 (878)	325–360 (346)	ventral	7–10 (8)	R	133–153 (141)	33–48 (42)
<i>Anonchotaenia yadavi</i> Sharma et Mathur, 1987	Sharma and Mathur (1987)	Jaipur, Rajasthan, India	<i>Pycnonotus jocosus abuensis</i> (Whistler) (Pycnonotidae)	350–1000	110–230	ventral	2–9	R	19–110	5–98
<i>Anonchotaenia zanthopygiae</i> Yamaguti, 1956	Yamaguti (1956)	Siriya, Aomori Prefecture, Japan	<i>Zanthopygia narcissina narcissina</i> (Temminck) (= <i>Ficedula narcissina narcissina</i>)	550–650	220–250	-	5–7 (6)	NR	60–100	18–22
<i>Anonchotaenia zonotrichicola</i> Dollfus, 1959	Dollfus (1959)	Cheayane, Azangaro Puno, Peru	<i>Zonotrichia capensis peruvienis</i> (Lesson) (Emberizidae)	510–570	170–200	-	4–7	*	46–54	22

* original host name followed by the current name if different in parentheses; ** in relation to osmoregulatory canals; MP – mature proglottides; GD – genital ducts; C – crossing and surpassing; R – reaching but not crossing; NR – not reaching; ¹ – usually numbers are in parentheses.

Fig. 3. *Anonchotaenia vaslata* sp. n., holotype (MHNG-PLAT 37770), from *Tyrannus melancholicus*. **A** – scolex; **B** – terminal genital ducts; **C** – mature proglottides; **D** – pre-gravid proglottides; **E** – gravid proglottides with eggs passing into the paruterine organ.

Type species: *Anonchotaenia clava* Cohn, 1900, a junior synonym of *A. globata* (von Listow, 1879).

Other species: See Table 1 for the list and key measurements of valid *Anonchotaenia* spp.

***Anonchotaenia (Anonchotaenia) vaslata* sp. n.**

Figs. 1B and 3

Description (based on five complete worms and one detached scolex from *Tyrannus melancholicus* (Vieillot)): Body ribbon-like, up to 31 mm ($n = 5$) long, maximum width 810–900 (853 ± 35 , $n = 5$) at level of pregravid proglottides (Fig. 1B). Formation of primordial genital organs appearing before external segmentation, which becomes distinct at level of late pre-mature proglottides.

Proglottides slightly craspedote, wider than long when mature (Fig. 3C); craspedote when pregravid and gravid, wider than long, only most terminal gravid proglottides as long as wide (Fig. 3E). Scolex rounded, 760–980 (878 ± 80 , $n = 6$) in diameter, with 4 round, muscular suckers, 325–360 (346 ± 10 , $n = 24$) in diameter (Fig. 3A). First proglottides appear 0.37–1.01 mm (0.8 ± 0.3 , $n = 5$) from posterior margin of suckers. Mature proglottides appear 2.14 mm ($n = 1$) posterior to appearance of first proglottides (Fig. 1B). Genital pores irregularly alternating (e.g. 3, 2, 1, 1, 1, 1, 1, 1, 2, 1 ...), opening in anterior third of lateral margin of mature proglottides and equatorial along lateral edge of post-mature and gravid proglottis. Genital atrium with infundibular orifice, narrow, deep

Table 2. Key measurements of specimens of *Anonchotaenia vaslata* sp. n. from various hosts from Paraguay.

Catalog number	Host species	Locality	Scolex diameter	Sucker diameter	Testis number	CS length MP	CS width MP	CS length PP	CS width PP
MHNG-PLAT 37966	<i>Tyrannus melancholicus</i> (Viellot) (Tyrannidae)	Paraguay	1 075 n = 1	405–425 (411 ± 10) n = 4	-	135–150 (143 ± 6) n = 5	-	-	-
MHNG-PLAT 39505	<i>Myiodynastes maculatus</i> (Statius Muller) (Tyrannidae)	Paraguay	1 075 n = 1	390–420 (403 ± 13) n = 4	7–10* (8 ± 1) [7(43%), 8 (43%), 9 (7%), 10 (7%)]	135–150 (141 ± 8) n = 4	50 n = 4	155–175 (163 ± 9) n = 4	40–45 (44 ± 3) n = 4

CS – cirrus sac; MP – mature proglottides; PP – pregravid proglottides; * n = 14 from 1 worm.

extended tubular middle part, flat base. Genital ducts pass ventrally to osmoregulatory canals. Osmoregulatory canals classically disposed; dorsal osmoregulatory canals 14–18 (15 ± 2, n = 4) in diameter; ventral osmoregulatory canals 35–43 (40 ± 4, n = 4) in diameter, with transverse anastomoses along posterior margin of each proglottis.

Testes spherical, slightly oval to oval in contracted proglottides, 45–55 (49 ± 4, n = 10) by 40–55 (49 ± 4, n = 10), arranged in single transverse dorsal row, occasionally overlapping osmoregulatory canals; 7–10 [7 (33%), 8 (44%), 9 (9%), 10 (14%)] (8 ± 1 n = 36) in number (Fig. 3C). Cirrus sac pyriform, tapering porally, rounded aporally, muscular, thick-walled (Fig. 3B), 133–153 (141 ± 6, n = 10) by 33–48 (42 ± 4, n = 10) in mature proglottides (Fig. 3C), 118–170 (143 ± 19, n = 8) by 38–45 (41 ± 2, n = 8) in early pregravid proglottides, reaching but not crossing osmoregulatory canals; length extending 16–18% (16 ± 1%, n = 10) across width of proglottis. Cirrus unarmed, cylindrical, 10–13 (12 ± 1, n = 10) in diameter. Internal vas deferens forming several coils in aporal portion of cirrus sac. External vas deferens large, wide, prominent, forming coiled mass occupying poral field of proglottis.

Ovary oval, medial, 50–73 (64 ± 6, n = 10) by 113–130 (120 ± 5, n = 10). Vitellarium compact, circular to oval, 38–60 (49 ± 6, n = 10) by 45–75 (62 ± 9, n = 10), aporal to ovary. Mehlis' gland indistinct. Seminal receptacle tubular to fusiform. Vagina conspicuous, walls well-demarcated, thick, opening posterior to male pore, separated into copulatory and conductive parts, passing posterior and ventral to cirrus sac and external vas deferens, lumen of copulatory portion crenulated (Fig. 3B).

Uterus in post-mature and early pregravid proglottides circular to oval, sac-like, occupying center of median field, dorsal to vitellarium, expanding to occupy posterior median field in pregravid and gravid proglottides. Uterine wall thickening as proglottis matures. Paruterine organ developing from anterior wall of uterus, conical, consisting of uniform fibrillar tissue, appearing 94 proglottides posterior to first mature proglottides (Fig. 1B); initially directed anteriorly or antero-laterally, curving dorsally in pregravid and gravid proglottides (Fig. 3D,E), curving 63 proglottides posterior to appearance (Fig. 1B). Eggs passing into paruterine cavity observed in terminal proglottis

of one worm (Fig. 3E). Gravid proglottides with all eggs completely within paruterine organ not observed. Eggs initially spherical. Oncospheres and embryophore transform into vermiform shape in late pregravid proglottides, appearing approximately 69 proglottides posterior to first appearance of paruterine organ, passing into paruterine cavity in vermiform state. Embryonic hooks present in central portion of vermiform oncospheres.

Type host: *Tyrannus melancholicus* (Viellot), tropical kingbird (Passeriformes: Tyrannidae).

Type locality: Jejui, San Pedro, Paraguay (24°06'S; 56°27'W, elevation 96 m).

Site of infection: Small intestine.

Type specimens: MHNG-PLAT 37770: Holotype (in black brackets) and 1 paratype on the same slide, 4 paratypes (3 complete worms and 1 detached scolex) on a separate slide.

Additional material examined: 1 complete worm (MHNG-PLAT 37966) from *T. melancholicus* from Arroyo Tapicuarai, San Pedro, Paraguay (24°36'S; 56°45'W); 1 complete worm in 4 fragments (MHNG-PLAT 39505) from *Myiodynastes maculatus* (Statius Muller), streaked flycatcher (Passeriformes: Tyrannidae) from Carapegua, Paraguari, Paraguay (25°44'S; 57°14'W).

Etymology: *Vas* (Latin) – vessel or duct + *lata* (Latin) – broad or wide; referring to the wide external vas deferens observed in this species.

Remarks. The new species presents a combination of characters unique from the other 28 species of *Anonchotaenia* (Table 1): wide, prominent external vas deferens rather than thin, coiled external vas deferens embedded within dense tissue, a paruterine organ that is conical but curves dorsally (even in relaxed proglottides) rather than a cylindrical, conical and straight, or otherwise shaped paruterine organ, and a long, narrow genital atrium with a flat base rather than short and wide with a rounded base.

Anonchotaenia (*Anonchotaenia*) *vaslata* sp. n. possesses genital ducts that pass ventrally to the osmoregulatory canals rather than between the canals as in the four species of *Anonchotaenia* (*Paranonchotaenia*): *A. dendrocitta*, *A. malaconoti*, *A. prionopos* and *A. prolixa*. *Anonchotaenia vaslata* belongs to a group of 15 *Anonchotaenia* species that have fewer than 11 but more than 6 testes per proglottis (Table 1). *Anonchotaenia vaslata* usually possesses 7–8 testes per proglottis unlike *Anonchotaenia cas-*

Fig. 4. Drawings of whole worms. **A** – *Anonchotaenia brasiliensis* from *Tachyphonus coronatus*; **B** – *Anonchotaenia macrocephala* from *Tachycineta leucorrhoa*. Abbreviations: O – oncospheres; PO – paruterine organ; VO – vermiform oncospheres.

tellani Fuhrmann et Baer, 1943 (usually 9–10), *Anonchotaenia clelandi* (Johnston, 1912) (usually 5), or *A. globata* (usually 4–5). Testes of the new species occur in a single transverse row rather than clustered in two groups as in *Anonchotaenia yadavi* Sharma et Mathur, 1987 and *Anonchotaenia quiscali* Rausch et Morgan, 1947.

Anonchotaenia vaslata possesses a longer cirrus sac (133–153 μm) than *Anonchotaenia arhyncha* Fuhrmann, 1918 (80–90 μm); this further differentiates the new species from *A. brasiliensis* (62.5–89 μm), *Anonchotaenia longiovata* Fuhrmann, 1901 (67–96 μm), *A. ranae* (50–62.5 μm), *A. yadavi* (19–110 μm), *A. castellani* (103–115 μm), *A. zanthopygiae* (60–100 μm), and *A. clelandi* (52–58 μm). The cirrus sac of *A. vaslata* reaches the osmoregulatory canals unlike *Anonchotaenia antirina* Singal, 1964, *A. brasiliensis*, *A. indica* Singh, 1964, *A. longiovata*, *A. ranae*, and *A. zanthopygiae* and is further differentiated from *A. globata* in this respect.

The new species possesses a larger scolex (760–980 μm) than *A. quiscali* (580 μm), *A. mexicana* (522–612 μm), *A. zanthopygiae* (550–650 μm) and *A. globata* (500–700 μm), further differentiating the new species from these taxa. The new species differs from *Anonchotaenia lambi* (Voge et Davis, 1953) in the possession of a smaller scolex (760–980 μm rather than 1120 μm) and a shorter cirrus sac (102–118 μm rather than 133–153 μm). *Anonchotaenia vaslata* possesses larger suckers (325–360 μm) than *A. mexicana* (121–198 μm) as well as a wider cirrus sac (33–48 μm rather than 19–27 μm in *A. mexicana*).

Material included here from *T. melancholicus* (MHNG-PLAT-37966) and *M. maculatus* (MHNG-PLAT 39505) is morphologically consistent with the type specimens (Table 2); *M. maculatus* represents an additional host record for *A. vaslata*.

Additional specimens examined in this study were found not to be conspecific with *A. vaslata* (Table S2). Specimens from Paraguay (MHNG-PLAT 37807 and MHNG-PLAT 39512) resemble *A. vaslata* in the form of the external vas deferens, paruterine organ, and genital atrium but differ in the possession of a smaller scolex (410–530 μm rather than 760–1075 μm), smaller suckers (170–195 μm rather than 325–425 μm) and a shorter cirrus sac (92.5–110 μm rather than 133–153 μm). Specimens from Bolivia (MHNG-PLAT 83085) resemble *A. vaslata* in the form of the external vas deferens, paruterine organ and genital atrium, but differ in the possession of a higher number of testes per proglottis (8–12, usually 10 rather than 7–10, usually 7 or 8). For these reasons, this material has not been included in the type series of the new species.

Anonchotaenia (Anonchotaenia) brasiliensis Fuhrmann, 1908
Figs. 4A, 5, 6A–C, 7

Redescription (based on eight complete worms and one scolex prepared for SEM from *T. coronatus* from Bra-

Fig. 5. *Anonchotaenia brasiliensis*: Brazilian material from *Tachyphonus coronatus* (A–E) and Paraguayan material from *Thraupis sayaca* (F). **A** – scolex; **B** – mature proglottids; **C** – terminal genital ducts; **D** – gravid proglottids with eggs passing into the paruterine organ; **E** – pre-gravid proglottids; **F** – gravid proglottids with eggs in the paruterine organ.

zil): Body ribbon-like, 31–47 (38 ± 6 mm, $n = 6$) in length, maximum width 593–696 (636 ± 33 , $n = 6$), at level of pre-gravid proglottids (Fig. 4A). External segmentation lacking at level of developed mature proglottids (Fig. 5C). Mature, post-mature and early pregravid proglottids slightly craspedote, wider than long (Fig. 5C); late pre-gravid proglottids slightly craspedote, as wide as long (Fig. 5E). Scolex rounded, 497–696 (628 ± 70 , $n = 7$) in diameter, with 4 round, muscular suckers, 222–271

(247 ± 13 , $n = 28$) in diameter (Figs. 5A, 6A). Sucker proximal and distal surfaces covered by acicular filitriches (Fig. 6B,C). First proglottids appear 1.0–1.4 mm (1.2 ± 1 , $n = 7$) from posterior margin of suckers. Mature proglottids appearing approximately 3.56 mm ($n = 1$) from posterior margin of suckers (Fig. 4A). Genital pores irregularly alternating in short series (e.g. 2, 1, 1, 2, 1, 3, 1, 2, 2, 1, 1, 1, 1, 1 ...); opening equatorial along lateral edge of proglottis. Genital atrium with infundibular

Fig. 6. Scanning electron micrographs: *Anonchotaenia brasiliensis* from *Tachyphonus coronatus* (A–C) and *Anonchotaenia macrocephala* (D–F). **A** – scolex; **B** – proximal surface of a sucker; **C** – distal surface of a sucker; **D** – scolex; **E** – proximal surface of a sucker; **F** – distal surface of a sucker.

orifice, tubular. Genital ducts pass ventral to osmoregulatory canals. Osmoregulatory canals classically disposed; dorsal osmoregulatory canals 8–18 (11 ± 2 , $n = 23$) in diameter; ventral osmoregulatory canals 17–25 (21 ± 2 , $n = 18$) in diameter; transverse anastomoses along posterior margin of each proglottis.

Testes spherical, circular to oval in contracted proglottides, 4–8 [4 (2%), 5 (30%), 6 (45%), 7 (19%), 8 (4%)] (6 ± 2 , $n = 112$) in number; arranged irregularly across proglottis in single plane, sometimes overlapping; 29–51 (43 ± 6 , $n = 38$) by 24–46 (37 ± 6 , $n = 38$), occasionally overlapping margins of ventral osmoregulatory canal (Fig. 5C). Cirrus sac pyriform, tapering slightly porally and aporally, muscular, thick-walled (Fig. 5B), 62.5–89 (73 ± 7 , $n = 25$) by 20–30 (25 ± 2 , $n = 25$) in mature proglottides (Fig. 5C), 65–89 (76 ± 5 , $n = 25$) by 20–35 (28 ± 4 , $n = 25$) in early pregravid proglottides, not reaching osmoregulatory canals, length extending 11–20% ($14 \pm 2\%$, $n = 40$) across width of proglottis. Cirrus unarmed, cylindrical. Internal vas deferens forming several coils in aporal portion of cirrus sac. External vas deferens

forming coiled mass, occupying anterior poral portion of median field of proglottis.

Ovary round to oval, poral, 36–62 (50 ± 7 , $n = 28$) by 45–94 (67 ± 10 , $n = 28$). Vitellarium compact, circular, 31–48 (38 ± 4 , $n = 28$) by 34–53 (45 ± 5 , $n = 28$), medial. Mehlis' gland not distinct. Seminal receptacle fusiform. Vagina tubular, opening posterior to male pore, not clearly separated into copulatory and conductive parts, passing posteriorly to cirrus sac, sometimes overlapping slightly, intertwining with external vas deferens (Fig. 5B). Uterus circular to oval, sac-like, dorsal to vitellarium, expanding to occupy entire medial field in pregravid (Fig. 5E) and gravid proglottides (Fig. 5D,F). Paruterine organ conical, consisting of uniform fibrillar tissue, developing as thickening of anterior uterine wall, appearing approximately 145 proglottides posterior to first mature proglottides (Fig. 4A), initially directed anteriorly or anterolaterally; in late pregravid proglottides broadening, joining uterus to form spherical shape (Fig. 5E). Eggs initially spherical. In pregravid proglottides oncospheres and embryo-phore transform into vermiform shape approximately 98

Table 3. Key measurements of type material and specimens of *Anonchotaenia brasiliensis* from various localities and host species and *A. cf. brasiliensis* from Brazil.

Catalog number	Host species	Locality	Scolex diameter	Sucker diameter	Testes number	CS extent	CS length MP	CS width MP	CS length PP	CS width PP
MHNG-PLAT 40213 (type material)	<i>Cacicus haemorrhous</i> (Linnaeus) (Icteridae)	Brazil	-	-	-	8–11% (10 ± 1%) n = 6	53–75 (65 ± 8) n = 6	33–40 (36 ± 3) n = 6	68–78 (72 ± 4) n = 6	38–43 (40 ± 2) n = 6
MZUSP 7191, MZUSP 7192, MZUSP 7193	<i>Thraupis cyanoptera</i> (Viellot) (Thraupidae)	Brazil	536–559 (547 ± 16) n = 2	202–241 (228 ± 14) n = 8	-	-	74–86 (80 ± 4) n = 10	23–31 (27 ± 3) n = 10	78–106 (92 ± 9) n = 10	21–35 (27 ± 4) n = 10
MHNG-PLAT 39504, MHNG-PLAT 39531	<i>Thraupis sayaca</i> (Linnaeus) (Thraupidae)	Paraguay	425–600 (512 ± 61) n = 8	160–240 (200 ± 23) n = 32	5–8* (7 ± 0.8) [5 (2%), 6 (32%), 7 (42%), 8 (24%)]	11–18% (15 ± 2%) n = 10	69–95 (79 ± 8) n = 10	25–35 (28 ± 3) n = 10	65–88 (73 ± 7) n = 10	20–33 (26 ± 4) n = 10
MHNG-PLAT 39494, MHNG-PLAT 82656	<i>Volatinia jacarina</i> (Linnaeus) (Thraupidae)	Paraguay	335–440 (392 ± 38) n = 6	154–183 (167 ± 7) n = 24	-	-	-	-	57–89 (70 ± 12) n = 7	17–22 (20 ± 2) n = 7
MZUSP 7284 (<i>Anonchotaenia cf. brasiliensis</i>)	<i>Ammodramus humeralis</i> (Bosc) (Emberizidae)	Brazil	424–562 (465 ± 65) n = 4	179–222 (196 ± 13) n = 16	4–8** (6 ± 1) [4 (1%), 5 (26%), 6 (46%), 7 (21%), 8 (6%)]	9–13% (11 ± 1%) n = 10	64–81 (72 ± 5) n = 10	25–33 (29 ± 2.5) n = 10	64–89 (76 ± 7) n = 10	21–30 (26 ± 3) n = 10

CS – cirrus sac; MP – mature proglottides; PP – pregravid proglottides; * n = 50 from 3 worms; ** n = 200 from 4 worms.

Fig. 7. *Anonchotaenia brasiliensis*, syntype: MHNG-PLAT 40213 from *Cacicus haemorrhous*. **A** – pregravid proglottides; **B** – terminal genital ducts.

proglottides posterior to appearance of paruterine organ (Fig. 4A); in late pregravid proglottides embryophore condensing into round shape with coiled vermiform oncospheres approximately 64 proglottides posterior to ap-

pearance of paruterine organ (Fig. 4A). Oncospheres pass into paruterine organ in coiled state (Fig. 5D). Embryonic hooks not observed.

Material examined: From *Tachyphonus coronatus* (Viellot), ruby-crowned tanager (PBI BRE-094) (Passeriformes: Thraupidae): 8 complete worms and 1 scolex prepared for SEM (MZUSP 7213, 7214) from Boracéia Biological Station, Salesópolis, São Paulo, Brazil (23°38'S; 45°52'W, elevation 946 m).

Additional material examined: From *Cacicus haemorrhous* (Linnaeus), red-rumped cacique (Passeriformes: Icteridae): 2 syntypes and 3 slides of longitudinal sections (MHNG-PLAT 40213) from 'San Paulo', Brazil (GPS data not specified); from *Thraupis cyanoptera* (Viellot), azure-shouldered tanager (PBI BRE-070) (Passeriformes: Thraupidae): 3 contracted worms (MZUSP 7191–7193) and 1 detached scolex prepared for SEM from Boracéia Biological Station, Salesópolis, São Paulo, Brazil (23°38'S; 45°52'W; elevation 946 m); from *Thraupis sayaca* (Linnaeus), sayaca tanager (Passeriformes: Thraupidae): 2 worms with contracted strobila (MHNG-PLAT 39504) from Carapegua, Paraguari, Paraguay (25°44'S; 57°15'W; elevation 92 m); 1 complete worm, 5 worms with incomplete strobila, 14 fragments of strobila, 1 detached scolex (MHNG-PLAT 39531) from San Lorenzo, Central, Paraguay (25°18'S; 57°30'W, elevation 146 m); from *Volatinia jacarina* (Linnaeus), blue-black grassquit (Passeriformes: Thraupidae): 3 worms with incomplete strobila and 2 fragments of strobila (MHNG-PLAT 39494) from Carapa, Canendiyu, Paraguay (24°08'24"S; 55°40'12"W, elevation 143 m); 3 complete worms and 3 fragments of strobila (MHNG-PLAT 82656) from Bahia Asunción, Distrito de la Capital, Paraguay (27°16'22"S; 57°37'37"W, elevation ca 61 m).

Prevalence and intensity of infection: From

Fig. 8. *Anonchotaenia macrocephala* from *Tachycineta leucorrhoa*. **A** – scolex; **B** – terminal genital ducts; **C** – late pre-gravid proglottides; **D** – mature proglottides; **E** – early pre-gravid proglottides; **F** – gravid proglottides with eggs passing into the paruterine organ.

T. coronatus, 14% (1 host infected of 7 examined), 25 worms; from *T. cyanoptera*, 25% (1 host infected of 4 examined), 7 worms.

Remarks. The original description of *A. brasiliensis* by Fuhrmann (1908) was very brief and included measurements and information about few, yet essential, characters without illustrations. The present redescription expands the range and provides information on additional morphological features with illustrations and scanning electron micrographs (see Figs. 5 and 6A–C for the morphology of newly collected specimens and Fig. 7A,B for

syntype morphology). *Anonchotaenia brasiliensis* is now characterised by its possession of five to eight, but usually six, testes per proglottis, a scolex 497–696 µm in diameter, suckers 223–271 µm in diameter, a cirrus sac 62–89 µm in length in mature proglottides, and a conical paruterine organ.

Prior to this study, *A. brasiliensis* had only been reported from *C. haemorrhous* and *Hemignathus virens* (Gmelin) (see Fuhrmann 1908, van Riper 1991). Material included here from *T. coronatus*, *T. cyanoptera*, *T. sayaca* and *V. jacarina* is morphologically consistent with

Fig. 9. *Anonchotaenia macrocephala* from *Progne 'purpurea'*, syntype: MHNG-PLAZ 40225. **A** – scolex; **B** – terminal genital ducts.

specimens from the type host, *C. haemorrhous* (Table 3). These species represent new host genus records and Thraupidae is a new host family record for *A. brasiliensis*. Specimens from the MHNG extend the geographic range of the species beyond Brazil to include areas in Paraguay.

Specimens from *Ammodramus humeralis* (Bosc), grassland sparrow (Passeriformes: Emberizidae) (MZUSP 7284) from Boracéia Biological Station, Salesópolis, São Paulo, Brazil (23°38'S; 45°52'W, elevation 946 m) are morphologically indistinguishable from *A. brasiliensis*, but the results of molecular analyses suggest they represent a species distinct from *A. brasiliensis*. As a result, these specimens were not included in the redescription based on the results of the molecular phylogeny and are referred to as *A. cf. brasiliensis* PBI-228.

Anonchotaenia (Anonchotaenia) macrocephala

Fuhrmann, 1908

Figs. 4B, 6D–F, 8–10

Redescription (one complete worm, two fragments of strobila and one scolex prepared for SEM from *T. leucorhoa* from Brazil): Body ribbon-like, up to 59 mm long ($n = 1$), maximum width 878–1011 (963 ± 59 , $n = 3$), at level of pregravid proglottides (Fig. 4B). Proglottides acraspedote, wider than long in mature, post-mature and young pregravid (Fig. 8D,E); slightly craspedote, as wide as long or longer than wide in late pregravid and gravid proglottides (Fig. 8C,F). Scolex rounded, massive, 994 ($n = 1$) in diameter with 4 round, muscular suckers, 412–428 (419 ± 7 , $n = 4$) in diameter (Figs. 6D, 8A). Sucker proximal and distal surfaces covered by acicular filitriches (Fig. 6E,F). First proglottides appear 1.9 mm ($n = 1$) from posterior margin of suckers. Mature proglottides appear approximately 3 mm ($n = 1$) from posterior margin of suckers (Fig. 2B). Genital pores irregularly alternating in short series (e.g. 3, 2, 1, 1, 1, 1, 1, 2, 1, 2, 1, 2, 1, 1 ...), opening equatorial along lateral edge of proglottis. Genital atrium with infundibular orifice, pyri-

form. Genital ducts pass ventrally to osmoregulatory canals. Osmoregulatory canals classically disposed; dorsal osmoregulatory canals 6–12 (9 ± 2 , $n = 12$) in diameter; ventral osmoregulatory canals 22–57 (31 ± 10 , $n = 12$) in diameter; transverse anastomoses along posterior margin of each proglottis.

Testes spherical, slightly oval to oval in contracted proglottides; 4–8 [4 (4%), 5 (17%), 6 (36%), 7 (32%), 8 (11%)] (6 ± 1 , $n = 95$) in number, arranged in single dorsal transverse row, 29–55 (43 ± 7 , $n = 37$) by 31–57 (44 ± 7 , $n = 37$), not overlapping marginal osmoregulatory canals (Fig. 8D), persist in pregravid proglottides. Cirrus sac pyriform, tapering porally, rounded aporally, muscular, thick-walled (Fig. 8B), 104–178 (137 ± 19 , $n = 24$) by 25–34 (30 ± 3 , $n = 24$) in mature proglottides, 93–125 (113 ± 9 , $n = 24$) by 23–31 (28 ± 2 , $n = 24$) in early pregravid proglottides, not reaching osmoregulatory canals; length extending 10–17% ($13 \pm 1\%$, $n = 28$) across width of proglottides (Fig. 8D,E). Cirrus unarmed, cylindrical. Internal vas deferens forming several coils in aporal portion of cirrus sac. External vas deferens appearing as bulbous mass in mature proglottides, developing into coiled, tubular mass in pregravid proglottides, occupying poral portion of median field.

Ovary round to oval, poral, 44–65 (54 ± 7 , $n = 17$) by 57–105 (89 ± 13 , $n = 17$). Vitellarium compact, circular, medial, 30–50 (39 ± 6 , $n = 17$) by 35–55 (43 ± 6 , $n = 17$). Mehlis' gland not distinct. Seminal receptacle fusiform. Vagina narrow, opening posterior to male pore, not clearly separated into copulatory and conductive parts, passing posteriorly to cirrus sac (Fig. 8B). Uterus circular to slightly oval, sac-like, occupying median field in post-mature and early pregravid proglottides. Paruterine organ consisting of uniform fibrillar tissue, developing as thickening of anterior uterine wall, initially conical, directed anteriorly (Fig. 8E), appearing approximately 150 proglottides posterior to first mature proglottides

Fig. 10. *Anonchotaenia macrocephala* from *Progne 'purpurea'*, syntype: MHNG-PLAZ 40225. **A** – mature proglottides; **B** – early pregravid proglottides; **C** – late pregravid proglottides.

(Fig. 4B). In pregravid proglottides pressing posteriorly into anterior uterine wall to form conical pocket inside uterine margins, but not invading uterus (Fig. 8C), approximately 105 proglottides posterior to appearance of paruterine organ (Fig. 4B). Eggs passing into paruterine cavity observed in terminal proglottis (Fig. 8F). Gravid proglottides with all eggs completely within paruterine cavity not observed. Eggs initially spherical. In pregravid proglottides oncospheres and embryophore transform into vermiform shape approximately 112 proglottides posterior to appearance of paruterine organ (Fig. 4B), pass into paruterine cavity in vermiform state (Fig. 8F). Embryonic hooks not observed.

Material examined: From *Tachycineta leucorrhoa* (Vieillot), white-rumped swallow (PBI BRE-211) (Passeriformes: Hirundinidae): 1 complete worm, 2 fragments of strobila, and 1 scolex prepared for SEM (MZUSP 7313, MZUSP 7314) from Boracéia Biological Station, Salesópolis, São Paulo, Brazil (23°38'S; 45°52'W, elevation 946 m).

Additional material examined:

From *Progne purpurea* [sic] (most likely *Progne subis* (Linnaeus), purple martin) (Passeriformes: Hirundinidae): 2 syn-

types (MHNG-PLAT 40225) from Brazil (exact locality is unknown); from *Progne tapera* (Linnaeus), brown-chested martin (Passeriformes: Hirundinidae): 1 syntype (MHNG-PLAT-40266) from Brazil (exact locality is unknown); from *Progne chalybea* (Gmelin), grey-breasted martin (Passeriformes: Hirundinidae): 3 syntypes (MHNG-PLAT 40227) from Brazil (exact locality is unknown); 4 complete worms and 4 fragments of strobila (MHNG-PLAT 37772) from Jejuí, San Pedro, Paraguay (24°06'S; 56°27'W, elevation 96 m); 4 complete worms and 2 fragments of strobila (MHNG-PLAT 39528) from Caicisa, Itapua, Paraguay (26°19'12"S; 55°13'11"W, elevation 324 m); 2 worms with contracted strobila and 1 fragment of strobila (MHNG-PLAT 39529) from Caicisa, Itapua, Paraguay (26°19'12"S; 55°13'11"W, elevation 324 m); 1 complete worm (MHNG-PLAT 39519) from Santa Maria, Itapua, Paraguay (26°53'59"S; 55°49'12"W, elevation 206 m); from *Hirundo* sp. (Passeriformes: Hirundinidae): 1 syntype (MHNG-PLAT 40229) from Brazil (exact locality is unknown); from *Hirondella* sp. [sic] (Passeriformes: Hirundinidae): 2 syntypes (MHNG-PLAT-40577) from Brazil (exact locality is unknown); from *Tachycineta meyeni* (Cabanis), Chilean swallow (PBI-CHI-043): 5 worms (MHNG-PLAT 83087) from Huinay Station, Comau Fjord, Los Lagos Region, Chile (42°22'47"S; 72°24'54"W, elevation 22 m); from *Stelgidopteryx ruficollis* (Vieillot), southern rough-winged swallow: 3 complete worms and 7 fragments of strobila (MHNG-PLAT 39521), 3 complete worms and 4 fragments of strobila (MHNG-PLAT 39522), and 2 worms with incomplete strobila and 2 fragments of strobila (MHNG-PLAT-39523), all from San Benito (Pastoreo), Itapua, Paraguay (26°48'35"S; 55°43'11"W, elevation 207 m).

Prevalence and intensity of infection: 1 *T. leucorrhoa* infected of 1 examined, 2 worms; from *T. meyeni*, 16.7% (1 host infected of 6 examined), 2 worms.

Remarks. The original description of *A. macrocephala* is brief and included measurements and descriptions of few, yet essential, characters. Fuhrmann (1908) described *A. macrocephala* as possessing 10–13 testes per proglottis, but examination of the type material showed the syntypes to actually possess 6–10 testes per proglottis (Table 4, Fig. 9). This redescription shifts the range of testes per proglottis from 10–13 to 6–10 and contributes information about additional morphological features with illustrations and scanning electron micrographs (see Figs. 6D–F, 8 for morphology of newly collected specimens and Figs. 9A,B, 10A–C for syntype morphology). Fuhrmann (1908) does not mention the change of paruterine organ shape we observed in pregravid proglottides, although the description included an illustration of a proglottis with a conical paruterine organ projecting anteriorly from the uterus. This illustration resembles the shape of the paruterine organ we observed in post-mature proglottides of newly collected material and the unique shape of the paruterine organ was observed in pregravid proglottides of the type material (Fig. 10C). *Anonchotaenia macrocephala* is

Table 4. Key measurements of specimens of *Anonchotaenia macrocephala* from various localities and host species.

Catalog number	Host species	Locality	Scolex diameter	Sucker diameter	Testes number	CS extent	CS length MP	CS width MP	CS length PP	CS width PP
MHNG-PLAT 40225 (Type material)	<i>Progne 'purpurea'</i>	Brazil	650–740 (695) n = 2	295–385 (350 ± 30) n = 8	-	14–23% (17 ± 3%) n = 8	73–118 (90 ± 15) n = 8	23–35 (27 ± 4) n = 8	83–153 (103 ± 22) n = 8	25–30 (28 ± 2) n = 8
MHNG-PLAT 40266 (Type material)	<i>Progne tapera</i> (Linnaeus)	Brazil	710 n = 1	310–330 (320 ± 9) n = 4	-	15–16% (15%) n = 2	95–108 (102 ± 6) n = 6	28–33 (31 ± 2) n = 6	90–108 (98 ± 9) n = 3	28–30 (29 ± 1) n = 3
MHNG-PLAT 40229 (Type material)	<i>Hirundo</i> sp.	Brazil	660–790 n = 2	240–415 (319 ± 83) n = 8	6–9* (8 ± 1) [6 (5%), 7 (46%), 9 (19%)]	11–19% (13 ± 3%) n = 8	60–93 (79 ± 11) n = 8	20–28 (26 ± 3) n = 8	78–88 (82 ± 4) n = 5	28–30 (29 ± 1) n = 5
MHNG-PLAT 40577 (Type material)	<i>Hirondella</i> sp.	Brazil	700–720 n = 2	255–310 (293 ± 18) n = 8	7–10** (9 ± 1) [7 (7%), 8 (29%), 10 (21%)]	13–16% (14 ± 2%) n = 4	-	-	68–73 (70 ± 2) n = 4	18–23 (22 ± 3) n = 4
MHNG-PLAT 40227, MHNG-PLAT 39528, MHNG-PLAT 39529, MHNG-PLAT 39519	<i>Progne chalybea</i> (Cabanis)	Paraguay	760–975 (858 ± 73) n = 9	290–360 (326 ± 17) n = 34	5–8*** (7 ± 1) [5 (2%), 6 (19%), 7 (48%), 8 (31%)]	-	-	-	72–125 (102 ± 6) n = 16	21–30 (27 ± 3) n = 16
MHNG-PLAT 39522, MHNG-PLAT 39523	<i>Stelgidopteryx ruficollis</i> (Viellot)	Paraguay	840–1000 (900 ± 47) n = 9	305–460 (361 ± 43) n = 36	5–7 ^a (6 ± 1) [5 (20%), 6 (50%), 7 (30%)]	-	-	-	-	-
MHNG-PLAT 83087	<i>Tachycineta meyeni</i> (Cabanis)	Chile	670–900 (748 ± 97) n = 5	305–410 (357 ± 32) n = 12	5–7 ^b (6 ± 1)	-	-	-	-	-

CS – cirrus sac; MP – mature proglottides; PP – pregravid proglottides; * n = 37; ** n = 14; *** n = 54; ^a n = 10; ^b n = 3.

characterised by a scolex 650–994 µm in diameter, a cirrus sac 60–178 µm in length in mature proglottides, and a paruterine organ that turns inward and expands towards the uterus rather than protruding from it.

Anonchotaenia macrocephala was originally described from *Progne 'purpurea'*, *P. tapera*, *P. chalybea*, *Hirundo* sp., and *Hirondella* sp. Of the syntypes available at the MHNG, the most intact specimens are from *Progne 'purpurea'* (MHNG-PLAT-40226); this host species name seems to be the latinisation of the common name purple martin (*Progne subis*). Voucher material from *T. leucorhoa*, *T. meyeni*, and *S. ruficollis* represent new host genera and species records for *A. macrocephala* and extend the geographic distribution of the species beyond Brazil to include areas in Paraguay and Chile.

Molecular analysis

The combined molecular dataset included a total of 6328 aligned characters (*ssrDNA*: 2692 characters, *lsrDNA*: 2119 characters, *rrnL*: 917 characters, *coxI*: 600 characters).

Pairwise uncorrected p-distances between the *coxI* gene fragments were as follows: between specimens of *A. prolixa* (PBI-147) and those identified as *A. brasiliensis* (PBI-204+212) averaged 14.3% ± 0.6%; between *A. brasiliensis* (PBI-204+212) and *A. macrocephala* (PBI-156+235) averaged 13.5% ± 0.4%; and between *A. brasiliensis* (PBI-204+212) and *A. cf. brasiliensis* (PBI-228) averaged 11.7%. The pairwise distances between the *rrnL* gene fragments were as follows: between specimens identified as *A. brasiliensis* (PBI-204+212) and *A. prolixa* (PBI-147) averaged 21.6% ± 0.7%; between *A. macro-*

cephala (PBI-156+235) and *A. prolixa* (PBI-147) averaged 22.2%; between *A. brasiliensis* (PBI-204+212) and *A. macrocephala* (PBI-156+235) averaged 18.9% ± 0.8%; and between *A. brasiliensis* (PBI-204+212) and *A. cf. brasiliensis* (PBI-228) averaged 15.6% (Table 5).

The log-likelihood of the topology produced by the ML analysis of the combined dataset was -14462.43. The harmonic means of estimated marginal likelihood values from the two BI runs were -14107.63 and -14113.62. The topologies of the BI and ML analyses were identical, thus only the results from the BI analysis are shown; all nodes were supported by pp ≥ 0.99 (Fig. 11). *Anonchotaenia prolixa* (PBI-147) placed basal to other representatives of *Anonchotaenia*. Specimens of *A. macrocephala* (PBI-156 and PBI-235) formed a clade distinct from specimens of *A. brasiliensis*. *Anonchotaenia brasiliensis* (PBI-204) was more closely related to *A. brasiliensis* (PBI-212) than to *A. cf. brasiliensis* (PBI-228). The long branch-length separating *A. cf. brasiliensis* (PBI-228) from *A. brasiliensis* (PBI-204+212) and the comparatively large pairwise genetic distance between them (11.7% *coxI*, 19.0% *rrnL*; see above) suggests these to be separate species.

Host specificity

The host data for each paruterinid used in the assessment of host specificity was derived from type and voucher specimens. The HS_s values calculated for each cestode species and the results of the determinations of relative host specificity for each species are provided in Table 6. The four species of paruterinids examined in this study exhibit host specificity index values ranging from HS_s = 0 (*A. prolixa*) to HS_s = 5.876 (*A. brasiliensis*).

Table 5. Uncorrected pairwise distances of *cox1* and *rrnL* gene fragments

<i>cox1</i>	1	2	3	4	5	6	7	8
1. <i>A. brasiliensis</i> (PBI-204)								
2. <i>A. brasiliensis</i> (PBI-212)	0.009							
3. <i>A. cf. brasiliensis</i> (PBI-228)	0.118	0.116						
4. <i>A. macrocephala</i> (PBI-156)	0.139	0.138	0.129					
5. <i>A. macrocephala</i> (PBI-235)	0.133	0.131	0.128	0.011				
6. <i>A. prolixa</i> sp. n. (PBI-147)	0.148	0.148	0.134	0.141	0.139			
7. <i>Biuterina</i> sp. (PBI-96) outgroup	0.154	0.151	0.158	0.143	0.138	0.136		
8. <i>Dictyterina cholodkowskii</i> (PBI-173) outgroup	0.154	0.155	0.134	0.139	0.139	0.144	0.123	
<i>rrnL</i>	1	2	3	4	5	6	7	8
1. <i>A. brasiliensis</i> (PBI-204)								
2. <i>A. brasiliensis</i> (PBI-212)	0.008							
3. <i>A. cf. brasiliensis</i> (PBI-228)	0.160	0.152						
4. <i>A. macrocephala</i> (PBI-156)	0.194	0.179	0.189					
5. <i>A. macrocephala</i> (PBI-235)	0.196	0.185	0.187	0.01				
6. <i>A. prolixa</i> sp. n. (PBI-147)	0.211	0.21	0.226	0.22	0.223			
7. <i>Biuterina</i> sp. (PBI-96) outgroup	0.239	0.239	0.257	0.263	0.264	0.275		
8. <i>Dictyterina cholodkowskii</i> (PBI-173) outgroup	0.212	0.255	0.27.8	0.288	0.289	0.193	0.243	

Table 6. Host specificity data for *Ananchotaenia* spp. from South America.

Cestode species	No. host species	No. host genera	No. host families	No. host orders	No. host classes	Rank	HS _s Index Value	Host specificity category (Caira et al. 2003)
<i>Ananchotaenia prolixa</i> sp. n.	1	1	1	1	1	1	0	oioxenous
<i>Ananchotaenia vaslata</i> sp. n.	2	2	1	1	1	1,001	3.000	metastenoxenous
<i>Ananchotaenia macrocephala</i>	6	3	1	1	1	2,003	3.302	metastenoxenous
<i>Ananchotaenia brasiliensis</i>	6	5	3	1	1	75,1497	5.876	euryxenous

DISCUSSION

Morphology

Morphological distinction amongst *Ananchotaenia* species can be challenging due to the scarcity of morphological characters and their tendency to become easily obscured or distorted in contracted material. In this study, we complement the set of historically useful characters for species delimitation with some previously disregarded additional characters. Paruterine organ shape is easily distorted by contraction in short proglottides and has not been considered reliable for species delimitation within *Ananchotaenia* (Fuhrmann 1908, Joyeux and Baer 1935, Rausch and Morgan 1947), although this character has been informative of species boundaries for other paruterinid genera (Georgiev and Korniyushin 1994).

Contrary to expectation, paruterine organ shape was distinguishable and remarkably consistent within each of the four *Ananchotaenia* spp. examined here (Figs. 3D,E, 4D,E, 5D–F, 7A, 8C,E,F, 10B,C). *Ananchotaenia brasiliensis* and *A. prolixa* had paruterine organs with a similar shape, a conical paruterine organ that maintained this shape in pregravid proglottides. *Ananchotaenia macrocephala* had a paruterine organ that was initially conical but in pregravid proglottides pressed posteriorly towards the anterior uterine wall forming a conical pocket inside the anterior margin of the uterus although not invading

it. *Ananchotaenia vaslata* had a paruterine organ that was initially conical but curved dorsally in pregravid proglottides. These differences were present in contracted and relaxed proglottides, suggesting that these differences were not an artefact of proglottis contraction, but rather represent species-level variation.

Among the species of *Ananchotaenia* examined here, landmarks of maturation of proglottides along the length of the strobila were observed and compared. The development of mature proglottides, the appearance of the paruterine organ, and the appearance of vermiform oncospheres occurred at a similar distance or number of proglottides posterior to the scolex with the number of proglottides between landmarks increasing with the length of the worm (Figs. 1, 4). Previous reports have suggested that the proglottides of some *Ananchotaenia* species mature earlier in the strobila than other paruterinid genera (Fuhrmann 1908, Rausch and Morgan 1947, Saxena and Baugh 1978), but few studies have reported variation in proglottis maturation along the length of the strobila among paruterinids. Future examination across the other 21 paruterinid genera will determine the variability of the location of these landmarks in comparison to *Ananchotaenia*.

Scanning electron microscopy is a standard tool used in species descriptions across a wide array of cestode taxa (see Chervy 2009) and until now has not been used to view

Fig. 11. Bayesian inference of the concatenated four-gene dataset depicting the phylogeny of species of *Anonchotaenia*. The tree was constructed using MrBayes v. 3.2.1 under the GTR+I (*ssrDNA*, *lsrDNA*) and GTR+G (*rrnL*, *coxI*) models; 5 million generations, 500 000 generations burn-in. The branch length scale indicates number of substitutions per site. Posterior probabilities are shown at nodes.

members of Paruterinidae. Within Cyclophyllidea, SEM has been used mostly to view scolex microthrix patterns of taxa that bear a rostellum, such as species of Dilepididae, Davaineidae, Hymenolepididae, Taeniidae and Anoplocephalidae (e.g. Mariaux and Vaucher 1990, Casado et al. 1994, Cielecka et al. 1994, Bâ et al. 1995). Our examination of scoleces of *A. brasiliensis* and *A. macrocephala* showed a consistent pattern of acicular filitriches across all scolex surfaces of each of the two species (Fig. 6). This is considerably less variation than has been reported for other cyclophyllidean taxa. It will be interesting to compare microthrix patterns across a broader spectrum of paruterinid taxa including both those with and without a rostellum to determine its utility as a taxonomic character among paruterinids.

Molecular analysis

This study provides the first published DNA sequence data of paruterinids beyond *ssrDNA* sequences of *Lyuterina nigropunctata* (Spasskaya et Spasskii, 1971) (AJ555173, AJ555174, Foronda et al. 2004) and a single translated *coxI* sequence of *A. globata* (JX310719) by presenting a four-gene phylogenetic analysis of six newly sampled paruterinid species (Fig. 11). Within the clade of *Anonchotaenia* representatives, specimens of *A. macrocephala* form a clade distinct from those identified as *A. brasiliensis* and *A. cf. brasiliensis*. The distinction of these species is further supported by the greater average genetic distance between these morphologically distinct species (on average $13.5\% \pm 0.4\%$; Table 5) than the genetic distances between conspecifics (0.9% in *A. brasiliensis* and 0.11% in *A. macrocephala*; Table 5).

Of the specimens identified as *A. brasiliensis*, those found in tanagers (PBI-204 and PBI-212) are each other's closest relatives showing little genetic divergence (0.8–0.9%; Table 5), whereas *A. cf. brasiliensis* (PBI-228) from an American sparrow represents a distinct lineage to the former (Fig. 11) despite being morphologically indistinguishable. The pairwise distances between *A. brasiliensis* (PBI-204 + PBI-212) and *A. cf. brasiliensis* (PBI-228) are 13 times (*coxI*) and 19 times (*rrnL*) as large as the distance between *A. brasiliensis* conspecifics (Table 5) indicating that *A. cf. brasiliensis* (PBI-228) may represent a distinct species. More specimens of *A. cf. brasiliensis* are necessary to determine if these specimens represent a cryptic species of *Anonchotaenia*.

Our analyses did not contradict the erection of *A. proluxa* as a new species. Interspecific genetic distances between previously described *Anonchotaenia* species (*A. brasiliensis* and *A. macrocephala*) were similar to those between these species and *A. proluxa* (Table 5). The inclusion of additional sequences of the other 26 *Anonchotaenia* spp. is needed to determine their relationship to *A. proluxa*.

Host associations

Members of Tyrannidae (tyrant flycatchers) are interesting hosts for *Anonchotaenia* species not only in terms of the diversity of tyrannids, but also because multiple *Anonchotaenia* species can parasitise a single tyrannid species. In this study, we found species of Tyrannidae host a diversity of morphologically distinct forms of *Anonchotaenia* and at least one tyrannid species is host to more than one *Anonchotaenia* species; *T. melancholicus*

has been reported to host both *Anonchotaenia jeandorsti* Dollfus, 1959 and *A. vaslata*. Tyrannidae is the most diverse avian family in the Western Hemisphere and contains approximately 100 genera and 430 species (Rheindt et al. 2008). This study includes cestode specimens from only three species of three tyrannid genera, a minute proportion of the described diversity within the family of which most species have yet to be examined for cestodes. Concentrating efforts on examining the diversity of tyrannids for cestodes and tracking cestode presence and absence among that diversity will contribute towards understanding the breadth of tyrannid species that host *Anonchotaenia*, but also the frequency by which the host spectrum of different *Anonchotaenia* species overlaps.

Even though patterns of paruterinid host associations are poorly understood, these associations appear to be closely tied to host diet. Intermediate hosts are unknown for most paruterinid species, but are suspected to be terrestrial insects in most cases. *Anonchotaenia brasiliensis* has been reported from three unrelated host families (Icteridae, Fringillidae and Thraupidae) whose members consume insects in addition to fruits and seeds, and frequently participate in mixed-species foraging flocks in the South American Atlantic forest (Develey and Peres 2000). These foraging flocks are composed predominately of omnivores and insectivores (Dario and De Vincenzo 2011) and members of these flocks exhibit common behaviours that increase foraging efficiency, such as prey flushing (Sridhar et al. 2009), which may explain the broad spectrum of host associations exhibited by *A. brasiliensis*.

Anonchotaenia species have been reported from a variety of passerine families (see Table S2) and exhibit a wide spectrum of host specificity (Table 6). *Anonchotaenia prolixa* was found in a single host species and by definition is oioxenous ($HS_s = 0$), although this could be an artefact of low sampling. *Anonchotaenia vaslata* and *A. macrocephala* both have been reported in multiple genera of a single host family and were metastenoxenous ($HS_s = 3.000$ and $HS_s = 3.302$, respectively). *Anonchotaenia macrocephala* has been found in more genera than

A. vaslata and thus had a higher index value. *Anonchotaenia brasiliensis* was the least host specific species evaluated here. It has been reported from multiple host families and was euryxenous ($HS_s = 5.876$). Host specificity of cestodes has been most thoroughly evaluated for species that parasitise elasmobranchs, partially because of the strict host specificity exhibited by most elasmobranch cestodes (Caira and Jensen 2001, Caira et al. 2003). Host specificity has not been formally assessed for most cyclophyllideans, which comprise over half of cestode diversity (> 3 500 species). In the present study, the HS_s values of the non-oioxenous species ranged from 3.000–5.876 (Table 6). These taxa exhibit a more relaxed pattern of host specificity than exhibited by the cestode taxa most often evaluated in formal assessments of host specificity (i.e. elasmobranch cestodes). It is possible that additional paruterinid species parasitise multiple avian host species and genera and, in some cases, multiple host families.

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