

HELMINTHOLOGIA, 53, 4: 391 - 395, 2016

Research Note

Report of the fourth specimen of *Amphiorchis solus* (Simha & Chattopadhyaya, 1970) Platt, 2002 46 years after the original description

M. R. WERNECK^{1*}, L. S. MEDEIROS²

¹BW Veterinary Consulting. Address: Rua Ponciano Eugênio Duarte 203, Centro, Zip Code: 11680-000, Ubatuba, São Paulo State, Brazil, *E-mail: max@bwvet.com.br; ²Aquatic Mammals Foundation (FMA). Address: Av. Tancredo Neves 5655, Jabotiana, Zip Code: 49095-000, Aracaju, Sergipe State, Brazil

Article info

Received January 25, 2016
Accepted March 30, 2016

Summary

The aim of this paper was to describe the occurrence of the fourth specimen of *Amphiorchis solus* (Simha & Chattopadhyaya, 1970) Platt, 2002 (Digenea: Spirorchiidae) in a green turtle (*Chelonia mydas* L. 1758) found in northeastern Brazil. *Amphiorchis solus* is a parasite that has only been reported in green turtles in India, Costa Rica and Brazil. Although the original description was made in 1970, only four specimens (including the one described herein) have been reported in the literature. In this note, we present the second occurrence of *A. solus* in Brazil, broadening information on the species and providing the first photographic record of this parasite.

Keywords: *Amphiorchis solus*; Brazil; *Chelonia mydas*; Digenea; Green sea turtle; Spirorchiidae; spirorchiids

Introduction

Spirorchiids (family Spirorchiidae Stunkard 1921) are a group of trematodes that inhabit the circulatory and lymphatic system of turtles. The family has approximately 100 species grouped into 19 genera, ten of which are parasites of sea turtles (Smith 1997, Platt, 2002). The genus *Amphiorchis* Price, 1934 is exclusively reported in sea turtles and six species are currently accepted for this genus: *A. amphiorchis* Price, 1934; *A. lateralis* Oguro, 1938; *A. indicus* Mehrotra, 1973; *A. caborjoensis* Fischthal & Acholonu, 1976; *A. indicum* Simha & Chattopadhyaya, 1980; and *A. solus* (Simha & Chattopadhyaya, 1970) Platt, 2002 (Smith, 1997; Platt, 2002). *Amphiorchis solus* has exclusively been described in green turtles (*Chelonia mydas* L. 1758). Although the species was first described more than 45 years ago, only three specimens have been reported in the literature (Simha & Chattopadhyaya, 1970; Santoro *et al.*, 2006; Werneck *et al.*, 2011).

The aim of the present note was to report the occurrence of the

forth specimen of *A. solus*, broadening morphometric and morphological information on this species through photographic images.

Materials and Methods

In August 2015, a juvenile green sea turtle (curved carapace length: 37.0 cm) was discovered in Mangue Seco in the state of Bahia on the northeastern coast of Brazil (11° 31' 24.924"S and 37° 23' 57.948"W). The turtle was found dead on the beach with diffuse algae distributed on the carapace as well as eggs from a leech *Ozobranchus* sp. (Hirudinea: Ozobranchidae) and barnacles on the carapace and plastron. Severe indentation of the plastron and lateral plates of the carapace were found, indicating cachexia. Moreover, skin tumors compatible with fibropapillomatosis were found in the eyes, cervical region, inguinal region and base of the right anterior fin.

Necropsy revealed hypotrophy, paleness and a gelatinous consistency of the pectoral muscle as well as blackish spots measuring

Table 1. Morphometric data, in millimeters, of *Amphiorchis solus* (Simha & Chattopadhyaya, 1970) Platt, 2002 (Digenea, Spirorchidae) from *Chelonia mydas* Testudines, Cheloniidae)

Variables	<i>Amphiorchis solus</i> (Simha & Chattopadhyaya, 1970)				Presente report
	Simha & Chattopadhyaya (1970)	Werneck et al. (2011)			
Host	<i>C. mydas</i>	<i>C. mydas</i>	<i>C. mydas</i>	<i>C. mydas</i>	
Locality	Camp Mandapam Gulf of Manar, India	Barra Nova Beach, State of Ceará, Brazil	Mangue Seco, state of Bahia, Brazil		
Site of infection	Heart	Heart	Heart	Heart	
Number of parasites	1	1	1	1	
Body length (mm)	3.96	2.86		2.85	
Body width anterior part*	0.81	0.55		0.54	
Body width posterior part**	-	-		0.44	
Oral sucker length	0.17	0.16		0.11	
Oral sucker width	0.16	0.14		0.09	
Acetabulum length	0.25	0.32		0.29	
Acetabulum width	-	0.23		0.29	
Esophagus length	1.17	0.97		1.01	
Esophagus width	-	-		0.079	
Anterior testis length	0.58	0.31		0.35	
Anterior testis width	0.51	0.31		0.25	
Posterior testis length	0.26	0.20		0.13	
Posterior testis width	0.27	0.17		0.25	
External seminal vesicle length	0.23	0.18		0.11	
External seminal vesicle width	-	0.091		0.083	
Cirrus pouch length	0.22	0.26		0.16	
Cirrus pouch width	-	-		0.066	
Ovary length	0.32	0.14		0.22	
Ovary width	0.30	0.13		0.15	
Distance from:					
Genital pore to posterior end	-	-		0.45	
Intestinal caeca bifurcation to anterior end	-	-		1.1	
Intestinal cecal end to posterior end	-	-		0.12	

Legend: * at the level to cecal bifurcation; ** at the level to anterior testis

one mm in diameter with multifocal distribution, concentrated more in the small intestine and to a lesser degree in the large intestine. The heart was examined using the method simplified by Werneck *et al.* (2006). Twenty three specimens of *Learedius learedi* Price, 1934 and one specimen of *A. solus* were found.

The helminths were found dead and fixed in a 70% ethanol, stained with carmine and cleared with eugenol. Morphometric data were acquired with the aid of an image analysis program (ImageJ, National Institutes of Health). Drawings were made using a drawing tube. The helminths were deposited in the Helminthological Collection of the Biosciences Institute (CHIBB), São Paulo State University (UNESP), Botucatu, São Paulo State, Brazil (*A. solus* number: 7843 and *L. learedi* number: 7844). A taxonomic key by Platt (2002), the original description (Simha & Chattopadhyaya, 1970) and a study by Werneck *et al.* (2011) were used for morphological and morphometric comparisons (Table 1).

Results and Discussion

Description (Fig. 1 and 2): Parasite small, with tapered anterior extremity (Fig. 2A) and slightly rounded posterior extremity (Fig. 2F), marked constriction in equatorial region (Fig. 2B) and small spines covering entire body; Oral sucker subterminal, esophagus long, sinuous and ending in a ceca bifurcation, ceca loop anteriorly then turn posteriorly (Fig. 2A and 2B); Ceca run parallel to side of body curvemediad at level of acetabulum, returning to lateral region of body when approaching anterior testis (Fig. 2C), terminating blindly posterior to posterior testis and anterior to excretory vesicle; Acetabulum rounded (Fig. 2B), positioned in equatorial region, coinciding with body constriction; Two testes, anterior testis larger and sinuous, resembling an S shape (Fig. 2C), posterior testis smaller and slightly oval in shape (Fig. 2D); Ovary lobulate, intercecal and located between cirrus sac and posterior testis (Fig. 2D – E); External seminal vesicle small, oval and intracecal, located posterior to anterior testis, in transverse position in relation to body (Fig. 2F); Cirrus sac elongate and immediately anterior to ovary, diagonal to body and (where visualization was possible) ventral to ovary, terminating in genital pore near posterior edge of ovary (Fig. 2F); Vitelline reservoir easily visible (Fig. 2D), occupying area between ovary and posterior testis; Excretory vesicle V-shaped; Vitellarium (Fig. 1 and 2B – F) composed of small follicles that run from cecal bifurcation to region of posterior testis and interrupted in region of ovary, ventral to cecae; Uterus (Fig. 2F – black arrow) with tubular structure occupying area lateroposterior to ovary, with trajectory turned toward medial region of body, at which point increasing in diameter, passing dorsally through the vitelline reservoir and terminating near region of genital pore; no eggs found.

Previous records: Ventricle of heart in *C. mydas* from Gulf of Manar, India (Simha & Chattopadhyaya, 1970) and Brazil (Werneck *et al.*, 2011 and present report) as well as intestine of *C. mydas* in Costa Rica (Santoro *et al.*, 2006)

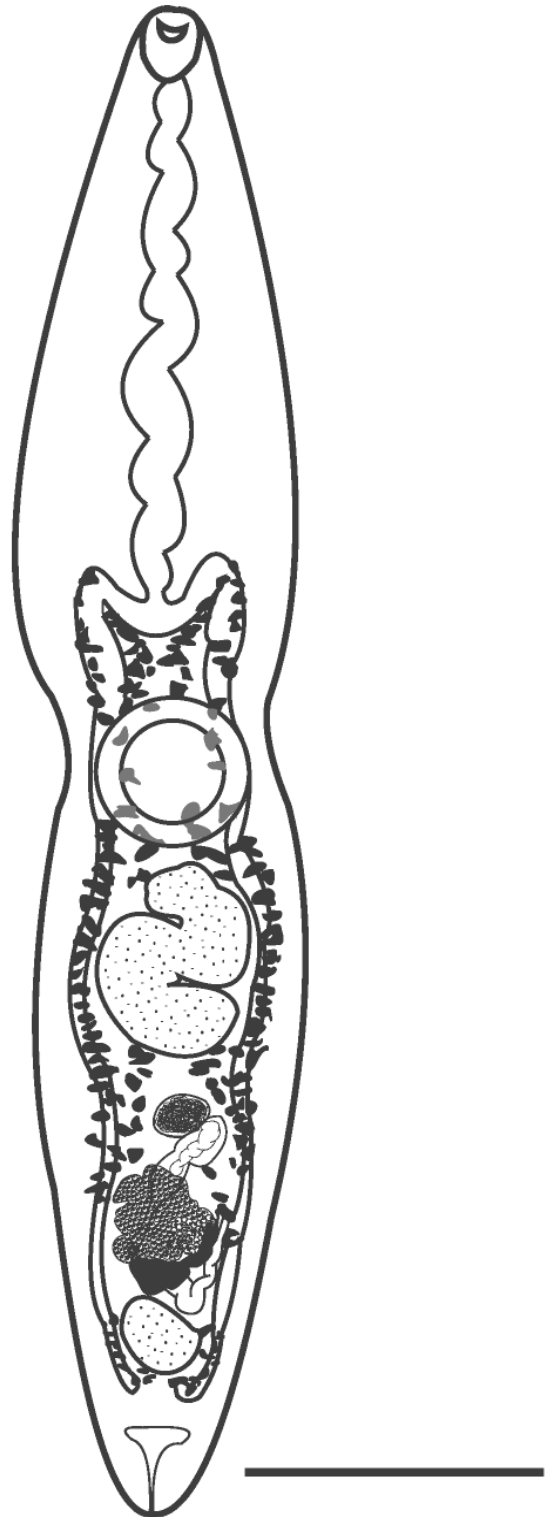


Fig. 1. *Amphiorchis solus* (Simha & Chattopadhyaya, 1970) Platt, 2002 (Digenea: Spirorchidae) found in *Chelonia mydas* Linnaeus 1758 (Testudines, Cheloniidae) from Brazil. (scale bar = 500 μ m)

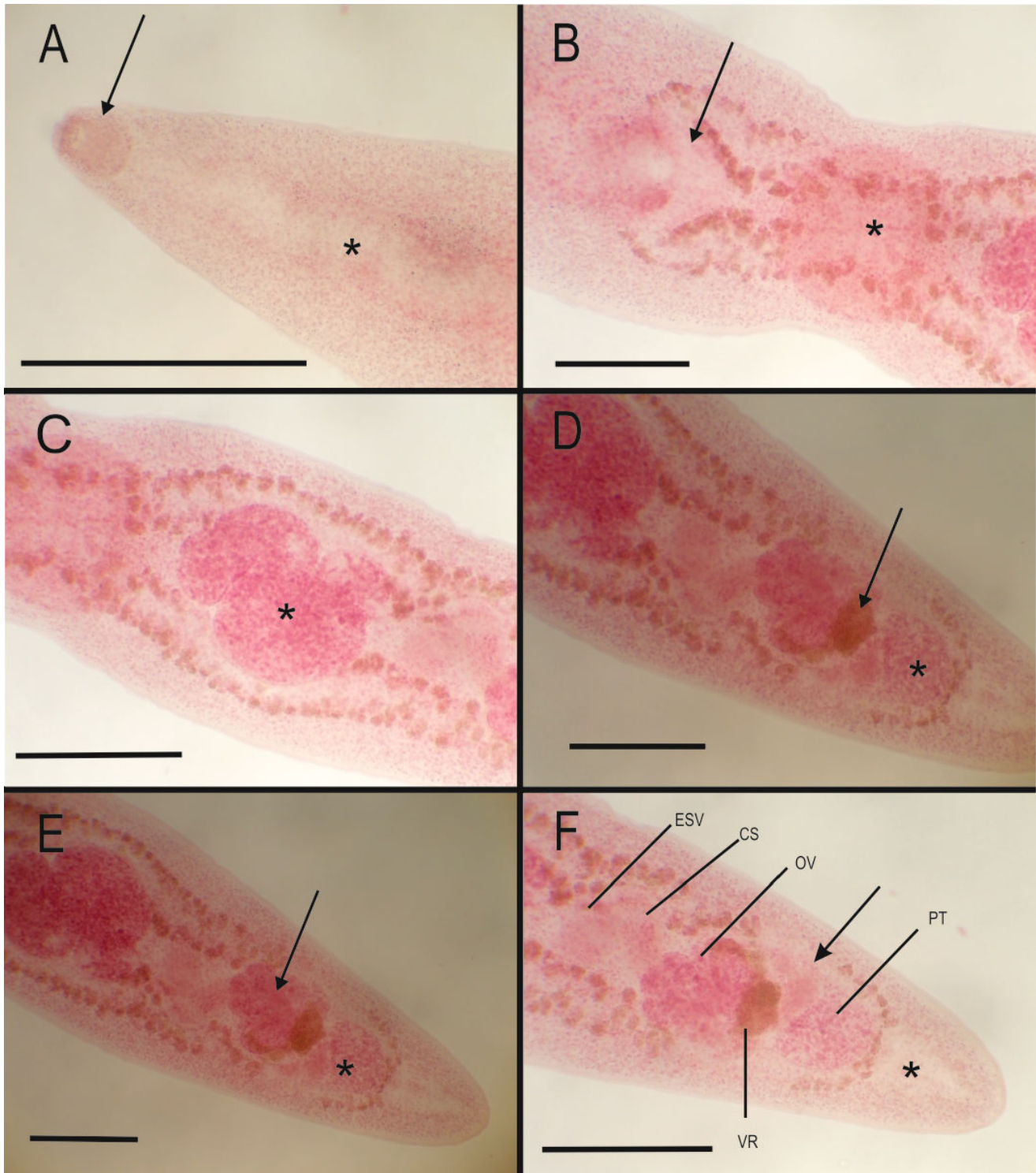


Fig. 2. *Amphiorchis solus* (Simha & Chattopadhyaya, 1970) Platt, 2002 (Digenea: Spirorchidae) found in *Chelonia mydas* Linnaeus 1758 (Testudines, Cheloniidae) from Brazil.

(A) Oral sucker (black arrow) and esophagus (asterisk) (scale bar = 500 μ m). (B) Intestinal bifurcation (black arrow) and acetabulum (asterisk) (scale bar = 250 μ m). (C) Anterior testis (asterisk) (scale bar = 250 μ m). (D) Dorsal view- Vitelline reservoir (black arrow) and posterior testis (asterisk) (scale bar = 200 μ m). (E) Dorsal view- Ovary (black arrow) and posterior testis (asterisk) (scale bar = 200 μ m). (F) Ventral view- Uterus (black arrow) and excretory vesicle (asterisk); ESV= external seminal vesicle, CS= Cirrus sac, OV= Ovary, PT= Posterior testis, VR= vitelline reservoir (scale bar = 250 μ m)

Remarks

Amphiorchis solus was originally described as *Squaroacetabulum solus* by Simha & Chattopadhyaya (1970) based on a single specimen collected from the heart of a green sea turtle from the Gulf of Manar, India. The acetabulum (“...whitish, situated anterior to the equatorial plane, is nearly square and the corners are slightly drawn outwards...”) and position of the pre-ovarian and post-ovarian vitelline follicles were what most piqued the interest of the authors and led them to institute the genus.

Platt (2002) considered the genus *Squaroacetabulum* to be synonymous with the *Amphiorchis*. Moreover, the author suggested that *A. solus* could be synonymous with *A. caborojoensis*, due to structural similarities and the shape of the acetabulum (“slightly squared ventral sucker”), but stressed that specimens of *S. solus* (*A. solus* n. comb.) were not available for evaluation and validation. Forty adult female turtles were evaluated during the nesting season in Costa Rica by Santoro *et al.* (2006), with the occurrence of only one specimen of *A. solus* (prevalence: 2.5%), which was collected from the intestine, but no morphological or morphometric data were provided.

Amphiorchis solus was again reported in the heart of a green sea turtle on the coast of Brazil, where only one specimen was found (Werneck *et al.*, 2011). The authors compared this specimen with specimens of *A. caborojoensis* and determined that the species are distinct and valid.

The morphometric analysis of the specimen revealed that the width of the acetabulum, dimensions of the oral sucker and external seminal vesicle, width of the anterior testis, length of the posterior testis and length of the cirrus sac were larger in comparison to previously published data (Table 1). Moreover, new data are presented herein on the width of the esophagus and cirrus sac as well as the distances from the genial pore and intestinal ceca to the posterior extremity and from the cecal bifurcation to the anterior extremity. However, such differences are not surprising, as this is only the third specimen of the species with known morphometric data and the findings merely reflect possible individual variation.

One of the most striking differences is the circular shape of the acetabulum, as also reported by Werneck *et al.* (2011), which differs from the original description of a square shape, explaining why the genus originally received the name *Squaroacetabulum* (Simha & Chattopadhyaya, 1970). Another interesting point regards the vitelline follicles, which are less dense in the present specimen and very different from those reported by the authors of the original description of the species. Moreover, a relatively voluminous tubular structure was found in the specimen described herein, which occupied the left side of the lateroposterior region of the ovary (Fig. 2F – black arrow). In our opinion, this is the uterine loop,

which is described for the first time for the species. Unfortunately, no eggs were found in the current specimen.

Based on the present findings, *A. solus* may be considered a rare species with low prevalence and intensity rates. Such information is supported by the few studies on this species (Simha & Chattopadhyaya, 1970; Santoro *et al.*, 2006; Werneck *et al.*, 2011; present report). The lack of specimens impedes broader morphological knowledge on the species and only reports of further individuals will contribute to a better understanding of the internal and surface structures of this parasite.

Acknowledgements

The authors wish to express their special thanks to Dr. Thomas Platt for critical reading and suggestions to improve the manuscript. This paper employed data generated by the Regional Program for Stranding and Abnormal Activity Monitoring in the Sergipe-Alagoas Basin carried out by the Fundação Mamíferos Aquáticos and Petrobras, in a partnership with Projeto Tamar/ICMBio, as a mitigating measure of the Federal Environmental Licensing conducted by the Brazilian environmental Agency IBAMA.

References

- PLATT, T.R. (2002): Family Spirorchidae Stunkard. In: GIBSON, D.I., JONES, A., BRAY, R.A. (Eds) *Keys to the Trematoda*. Volume 1. London, UK: CABI Publishing, pp. 453 – 467
- SANTORO, M., GREINER, E.C., MORALES, J.A., RODRÍGUEZ-ORTÍZ, B. (2006): Digenetic trematode community in nesting green sea turtles (*Chelonia mydas*) from Tortuguero National Park, Costa Rica. *J. Parasitol.*, 92(6): 1202 – 1206. DOI: 10.1645/GE-866R.1
- SIMHA, S.S., CHATTOPADHYAYA, D.R. (1970): A new genus and species of a blood flukes, *Squaroacetabulum solus*, from the ventricle of the heart of a marine turtle, *Chelone mydas*. *Zool. Anz.*, 184: 290 – 294
- SMITH, J.W. (1997): The blood flukes (Digenea: Sanguinicolidae and Spirorchidae) of cold-blooded vertebrates: Part 2. *Helminthol. Abst.*, 66: 329 – 344
- WERNECK, M.R., BECKER, J.H., GALLO, B.G., SILVA, R.J. (2006): *Learedius learedi* Price 1934 (Digenea, Spirorchidae) in *Chelonia mydas* Linnaeus 1758 (Testudines, Cheloniidae) in Brazil: case report. *Arq. Bras. Med. Vet. Zootec.*, 58(4): 550 – 555. DOI: 10.1590/S0102-09352006000400016
- WERNECK, M.R., GALLO, B.M.G., LIMA, E.H.S.M., SILVA, R.J. (2011): Occurrence of *Amphiorchis solus* Simha & Chattopadhyaya, 1970 (Digenea, Spirorchidae) infecting Green turtle *Chelonia mydas* Linnaeus, 1758 (Testudines, Cheloniidae) in Brazil. *Comp. Parasitol.*, 78(1): 200 – 203. DOI: 10.1654/4435.1