

Occurrence of *Amphiorchis indicus* Mehrotra, 1973 (Digenea, Spirorchiiidae) infecting Green turtle *Chelonia mydas* Linnaeus, 1758 (Testudines, Cheloniidae) in Brazil

Werneck, MR.^{a*} and Silva, RJ.^b

^aBW Consultoria Veterinária, Rua Ponciano Eugênio Duarte, 203, Centro, CEP 11680-000, Ubatuba, SP, Brazil

^bDepartamento de Parasitologia, Instituto de Biociências, Universidade Estadual Paulista – UNESP, Botucatu, SP, Brazil

*e-mail: max@bwwet.com.br

Received March 19, 2012 – Accepted May 11, 2012 – Distributed February 28, 2013

(With 1 figure)

The family Spirorchiiidae was created by Stunkard (1921) and includes approximately 100 species distributed among 19 genera described parasitising terrestrial and aquatic chelonians (Platt, 2002). Ten of these genera are exclusive to sea turtles (Smith, 1997).

The genus *Amphiorchis* Price 1934 was created to house the species *Amphiorchis amphiorchis* Price, 1934, found in a specimen of *Chelonia mydas* Linnaeus, 1758. There are currently five other species accepted for this genus: *A. lateralis* Oguro, 1938, *A. indicus* Mehrotra, 1973, *A. caborojoensis* Fischthal & Acholonu 1976, *A. indicum* Simha & Chattopadhyaya, 1980 (Smith, 1997) and *A. solus* n. comb. (Simha and Chattopadhyaya, 1970) (Platt, 2002). The present study reports the occurrence of *A. indicus* in *C. mydas* for the first time in Brazilian waters.

The hosts had their circulatory system examined according to the method described by Snyder and Clopton (2005) and simplified by Werneck et al. (2006). The helminths (n = 5) were fixed in AFA (Alcohol-Formalin-Acetic Acid) solution under cover slip pressure, stained with chloridric carmine and analysed on a computerised system for image analysis (QWin Lite 3.1 – Leica). Morphometrical values (in micrometres) are shown as mean and range (minimum-maximum). The helminths collected during the study were deposited in the Coleção Helmintológica do Instituto de Biociências (CHIBB) of the Universidade Estadual Paulista, Botucatu, in the state of São Paulo, Brazil (Numbers 4046, 4048, 4991-4995). All collections were authorised by federal licenses for activities for scientific purposes (SISBIO 12421-1 and 12431-2).

Prevalence, mean infection intensity and mean abundance values were determined by species based on Bush et al. (1997).

1. Results

A total of 348 specimens of *C. mydas* underwent necropsy between 2008 and 2010 and specimens of *A. indicus* were found in six individuals, with a prevalence rate of 1.7% and mean intensity of 2.3 ± 0.8 (1 to 6). The specimens were found in the gastrointestinal tract (n = 8) of two hosts and the liver (n = 6) of four hosts. Hosts positive for *A. indicus* were from the southern coast of Rio de Janeiro state (n = 2) and the northern coast of São Paulo state (n = 4), Brazil. Table 1 displays the results of the morphometric analysis (n = 5).

2. *Amphiorchis indicus* Mehrotra, 1973 (Figure 1, Table 1)

2.1. Description

Body small, with thin extremities, wider in the testicular region, surface without spines; Oral sucker small and terminal;



Figure 1. *Amphiorchis indicus* Mehrotra, 1973 (Digenea: Spirorchiiidae) from green turtle *Chelonia mydas* Linnaeus, 1758 (Testudines: Cheloniidae), from Brazil. Scale bar 1 mm.

Table 1. Morphometric data, in millimetres, of *Amphiorchis indicus* Mehrotra, 1973 (Digenea: Spirorchiidae) from marine turtles (Testudines: Cheloniidae).

	Gupta and Mehrotra (1981)	Present report
Host	<i>Eretmochelys imbricata</i>	<i>Chelonia mydas</i>
Locality	Gulf of Mannar (India)	São Paulo and Rio de Janeiro States (Brazil)
Site of infection	Liver	Oesophagus, stomach, small intestine, large intestine and liver
Number of parasites	22	5
Body length	2.69-3.517	4.48 ± 0.62 (3.59-5.04)
Body width	0.246-0.396	0.327 ± 0.04 (0.29-0.39)
Oral sucker length	0.059-0.077	0.095 ± 0.021 (0.070-0.122)
Oral sucker width	0.045-0.069	0.066 ± 0.012 (0.054-0.083)
Acetabulum length	0.041-0.082	0.116 ± 0.04 (0.077-0.167)
Acetabulum width	0.077-0.110	0.098 ± 0.039 (0.055-0.148)
Esophagus length	0.295-0.426	0.438 ± 0.102 (0.312-0.550)
From the end of right caecum until the final extremity of the body	0.196-0.278	0.584 ± 0.108 (0.448-0.713)
From the end of left caecum until the final extremity of the body	0.418-0.639	0.948 ± 0.222 (0.615-1.077)
Anterior testis length	0.139-0.278	0.568 ± 0.160 (0.329-0.679)
Anterior testis width	0.155-0.221	0.261 ± 0.049 (0.197-0.317)
Posterior testis length	0.393-0.557	0.597 ± 0.094 (0.493-0.691)
Posterior testis width	0.196-0.278	0.266 ± 0.025 (0.236-0.287)
Ovary length	0.172-0.311	0.284 ± 0.040 (0.231-0.324)
Ovary width	0.155-0.196	0.212 ± 0.030 (0.185-0.239)
Mehlis' gland length	0.069-0.118	0.121 ± 0.015 (0.099-0.135)
Mehlis' gland width	0.049-0.077	0.076 ± 0.014 (0.063-0.094)
Cirrus pouch length	0.069-0.094	0.108 ± 0.026 (0.083-0.137)
Cirrus pouch width	0.094-0.135	0.092 ± 0.012 (0.073 -0.100)
External seminal vesicle length	0.055-0.110	0.138 ± 0.027 (0.103-0.172)
External seminal vesicle width	0.073-0.139	0.110 ± 0.045 (0.055-0.163)
Yolk reservoir length	0.094-0.172	0.056 ± 0.019 (0.042-0.085)
Yolk reservoir width	0.049-0.118	0.120 ± 0.029 (0.097-0.161)
Egg length	0.131-0.164	0.167 ± 0.041 (0.138-0.197) n = 3
Egg width	0.164-0.205	0.020 ± 0.0001 (0.020-0.020) n = 3
Uterus length		0.525 ± 0.082 (0.402-0.579)

esophagus short and sinuous, approximately one tenth of total body length; Pharynx absent; Ceca small, sinuous, terminating asymmetrically, right cecum longer than left, ceca covered with vitellaria throughout practically their entire length, denser in the region between the end of the esophagus and anterior testicle and between the ovarian region and posterior testicle; Excretory vesicle terminal, Y-shaped; two voluminous testicles in tandem, the anterior of which occupies the area between the acetabulum and outer seminal vesicle and the posterior of which is found after the yolk reservoir, contour irregular, slightly oval, anterior testicle slightly larger than the posterior; External seminal vesicle occupying areas to the left of the cirrus sac and immediately posterior to the anterior testicle, with

irregular shape; Cirrus sac with irregular shape, occupying median region between the external seminal vesicle and ovary; Genital pore in median region immediately anterior to the ovary; Uterus short, slightly sinuous, located to the left between the ovary and cecum; Eggs observed; Ovary oval shaped, located between Mehlis' gland and cirrus sac; Mehlis' gland posterior to the intracecal ovary; Vitellaria densely distributed throughout the body, exhibiting small follicles extending from the cecal bifurcation to the extremity of the right cecum; Fields interrupted only in region of the testicle and ovary.

2.2. Remarks

Amphiorchis indicus was found in the liver of a sea turtle specimen *Eretmochelys imbricata* Linnaeus 1758 in

India (Gupta and Mehrotra, 1981). According to Smith (1997) this species was first identified in a *C. mydas*, making the validity of the host species discussed. The results of the present study confirm the occurrence of *A. indicus* in *C. mydas*.

This study is the second record of this helminth and the first record in sea turtles on the coast of Brazil. The only previous report of *A. indicus* involved the aforementioned specimen of *E. imbricata* in India, in which 22 specimens of the helminth were found (Gupta & Mehrotra, 1981). Based on these two isolated descriptions, the prevalence of this parasite can be considered low.

The morphometric analysis of the specimens reveals larger body length and width, larger distance between the end of the right cecum and posterior extremity and larger posterior testicle length in comparison to the data described by Gupta and Mehrotra (1981).

There are few studies on the occurrence of parasites from the family Spirorchiidae in Brazil, with only five species reported thus far: *Learedius learedi* Price, 1934 (Werneck et al., 2006) and *Monticellius indicum* Mehra, 1939 (Werneck et al., 2008a) in juvenile specimens of *C. mydas*; *A. solus* in an adult specimen of *C. mydas* (Werneck et al., 2011); and *Carettacola stunkardi* (Martin & Bamberger, 1952) Dailey, Fast & Balazs, 1991 and *A. caborjoensis* in *E. imbricata* (Werneck et al., 2008b).

This is the first report of *A. indicus* found in sea turtles from the Brazilian coast, thus providing important information about helminth fauna in this region and broadening the geographical distribution of this species. This report also contributes to the knowledge of morphometrical data of the species.

Acknowledgements – We would like to thank the Fundação de Amparo à Pesquisa do Estado de São Paulo - FAPESP (Proc. 07/59504-7) for the financial support.

References

BUSH, AO., LAFFERTY, KD., LOTZ, JM. and SHOSTAK, AW., 1997. Parasitology meets ecology on its own terms: Margolis et al. revisited. *Journal of Parasitology*, vol. 83, p. 575-583.

GUPTA, NK. and MEHROTRA, V. 1981. On two blood flukes (trematoda) of the family Spirorchiidae Stunkard, 1921 from Indian marine turtles. *Acta Parasitologica Polonica*, vol. 28, no. 2, p. 11-20.

PLATT, TR., 2002. Family spirorchiidae Stunkard. In GIBSON, DI., JONES, A. and BRAY, RA. (Eds.). *Keys to the Trematoda*. London: CABI Publishing. p. 453-467.

SMITH, JW., 1997. The blood flukes (Digenea: Sanguinicolidae and Spirorchiidae) of cold-blooded vertebrates: part. 2. *Helminthological Abstracts*, vol. 6, no. 10, p. 329-344.

SIMHA, SS. and CHATTOPADHYAYA, DR. 1970. A new genus and species of a blood flukes, *Squaroacetabulum solus*, from the ventricle of the heart of a marine turtle, *Chelone mydas*. *Zoologischer Anzeiger*, vol. 184, p. 290-294.

SNYDER, S. and CLOPTON, R., 2005. New methods for the collection and preservation of spirorchiid trematodes and Polystomatid manogeneus from turtles. *Comparative Parasitology*, vol. 72, no. 1, p. 102-107. <http://dx.doi.org/10.1654/4155>

WERNECK, MR., BECKER, JH., GALLO, BMG. and SILVA, RJ., 2006 *Learedius learedi* Price 1934 (Digenea, Spirorchiidae) in *Chelonia mydas* Linnaeus 1758 (Testudines, Cheloniidae) in Brazil: case report. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia*, vol. 58 no. 4, p. 550-555. <http://dx.doi.org/10.1590/S0102-09352006000400016>

WERNECK, MR., GALLO BMG. and SILVA RJ., 2008a. First report of *Monticellius indicum* Mehra, 1939 (Digenea: Spirorchiidae) infecting *Chelonia mydas* Linnaeus, 1758 (Testudines: Cheloniidae) from Brazil. *Brazilian Journal of Biology*, vol. 68, no. 2, p. 455-456. PMID:18660980. <http://dx.doi.org/10.1590/S1519-69842008000200033>

-, 2008b. Spirorchiids (Digenea: Spirorchiidae) infecting a Hawksbill sea turtle *Eretmochelys imbricata* (Linnaeus 1758) from Brazil. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia*, vol. 60, no. 3, p. 663-666. <http://dx.doi.org/10.1590/S0102-09352008000300021>

WERNECK, MR., GALLO, B., LIMA, EHDM. and SILVA, RJ., 2011. Occurrence of *Amphiorchis solus* Simha & Chattopadhyaya, 1970 (Digenea, Spirorchiidae) infecting Green turtle *Chelonia mydas* Linnaeus, 1758 (Testudines, Cheloniidae) in Brazil. *Comparative Parasitology*, vol. 78, no. 1, p. 200-203.

