ISSN Versión impresa 2218-6425 ISSN Versión Electrónica 1995-1043

RESEARCH NOTE/NOTA CIENTÍFICA

FIRST RECORD OF *ANISAKIS SP*. (NEMATODA, ANISAKIDAE) L3 INFECTING THE BODY CAVITY OF *ATLANTORAJA PLATANA* (CHONDRICHTHYES, RAJIDAE)

PRIMER REGISTRO DE *ANISAKIS SP*. (NEMATODA, ANISAKIDAE) L3 EN LA CAVIDAD CORPORAL DE *ATLANTORAJA PLATANA* (CHONDRICHTHYES, RAJIDAE)

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Neotropical Helminthology, 2015, 9(2), jul-dec: 359-365.

ABSTRACT

This communication is the first record of the presence of a third stage larva of *Anisakis sp.* infecting *Atlantoraja platana.* The hosts were collected from fishery landings at processing plants of San Antonio Oeste (40° 44' S 64° 57' O) and San Antonio Este ports (40° 49' S 64° 57' O), Rio Negro province, Argentina. They were found in the visceral cavity near the epigonal organ, a lymphomyeloid tissue closely associated with gonads and only in cartilaginous fish. The high concentrations of urea in the body fluid and tissues of elasmobranch hosts made an inhospitable environment to the colonization of helminthes. The results produced by this work constitute the first report of L_3 of *Anisakis sp*. in the body cavity of an elasmobranch, in particular *A. platana*, and show the capability of this anisakid to survive in the visceral mass of these hosts.

Keywords: *Anisakis* sp. - *Atlantoraja platina* - Epigonal organ - L_3 - Nematodes.

RESUMEN

En la presente nota se registra por primera vez el tercer estadio larval de *Anisakis* sp. infectando a *Atlantoraja platana* (Günther, 1880). Los hospederos fueron obtenidos en desembarques pesqueros de plantas de procesado de los puertos de San Antonio Oeste (40° 44' S 64° 57' O) y San Antonio Este (40° 49' S 64° 57' O), Provincia de Río Negro, Argentina. Las larvas fueron colectadas en la cavidad visceral de los peces, cerca del órgano epigonal, un tejido linfomieloide estrechamente asociado a las gónadas y exclusivo de los peces cartilaginosos. Hay evidencias documentadas que las altas concentraciones de urea en tejidos y fluídos corporales tornan inhabitable el medioambiente celómico para ser colonizado por helmintos. Los resultados expuestos en este trabajo constituyen el primer reporte de L_3 de *Anisakis sp. en la cavidad corporal de un elasmobranquio, en particular A. platana*, y demuestran la capacidad de este anisákido para sobrevivir en la masa visceral de estos hospederos.

Palabras clave: *Anisakis sp.* L_3 - *Atlantoraja platana* - *Epigonal organ* - Nematodes.

INTRODUCTION

The anisakid nematodes parasitize elasmobranches as larvae or adults and are generally internal parasites (Caira, 1990) besides they can be found at integumentary level. The location where larval nematodes can be found range from the uterus (Benz *et al.*, 1987) and ovaries (Rosa-Molinar *et al.*, 1983; Aragort *et al.*, 2002) to superficial body tissues (Ruyck & Chabaud, 1960).

Anisakids use fish as paratenic hosts to close their complex life cycles (Guagliardo *et al.*, 2009). There are relatively few records of nematodiasis in rays (Mc Vicar 1977; Romera, 1993; Tanzola *et al.*, 1998; Borucinska & Heger, 1999; Sanmartín *et al.*, 2000, Knoff *et al.*, 2001; Aragort *et al.*, 2002; Santos *et al.*, 2004; Alvarez *et al.*, 2006; Díaz Andrade *et al.*, 2008). The present communication is the first report of the third stage larva (L_3) of *Anisakis sp*. infecting the body cavity of *Atlantoraja platana* (Günther, 1880). This skate is an endemic species from the Southwest Atlantic Ocean and one of the most commonly taken as bycatch in the coastal fisheries at the San Matías Gulf (Northern Patagonia, Argentina). Its IUCN Red List Category is "Vulnerable A4bd"(http://www.iucnredlist.org/details/63110/0).

MATERIAL AND METHODS

Males specimens of *A. platana* were collected from fishery landings at processing plants of San Antonio Oeste (40° 44' S 64° 57' O) and San Antonio Este ports (40° 49' S 64° 57' O), Rio Negro province, Argentina. The specimens were taxonomically identified following Coller (2012). Two of fifteen (13.3% prevalence) result parasitized by anisakid larvae. These findings proceed from histopathological examination.

Small pieces of the male reproductive system were fixed in Bouin's solution in seawater for at least 24 h. Afterwards, all material was dehydrated through a graded series of ethanol and embedded in Paraplast®. Sections of 4-5 µm thick were stained by Masson's trichromic stain and hematoxilyn-eosin. Selected sections were photographed using an Olympus BX51 light microscope equipped with an Olympus C-7070 digital camera.

Following the criteria of Oshima (1972) the larvae were identified as third stage (L_3) of *Anisakis sp.* and were characterized by a cuticle with slight ripples, number of muscular cells (41-56 muscular cells per quadrant) and hypodermic cords with "Y-shape" associated with the excretory gland (Figs. 1 y 2). The maximum diameter of the larvae were between 0.35 – 0.37 mm.

RESULTS AND DISCUSSION

The parasites were found in visceral cavity of two males of *A. platana* (one juvenile and other sexually mature). In the juvenile specimen, larvae were found near the epigonal tissue adjacent to the testes. In this case, inflammatory tissue was evident (Fig. 3). In the mature host, larvae were found near the epigonal organ bordering the genital ducts and no inflammatory tissue was detected (Fig. 4). Despite the presence of larvae, the cytoarchitecture of reproductive and lymphomyeloid structures of the hosts was preserved.

The larvae stages of *Anisakis sp.* present in fish tissues can be determined, in cross section, by the structure of the digestive tract, the morphology of the hypodermic cords and the association of these structures with the excretory gland (Oshima, 1972). However, few characters of the adults are present in larvae, so the specific determination of the

parasite, based on the larvae, is complex (Dick & Choudhury, 1995; Guagliardo *et al.,* 2009).

The general pathology of fish specify that the major diseases caused by nematodes have targeted organs as intestine, liver, heart, body cavity, muscle, gonads (especially ovary) and swim bladder (Williams & Jones, 1994; Dick & Choudhury, 1995). Borucinska & Heger (1999) associated the presence of granulomas scattered throughout different organs (spleen, stomach, spiral valve, kidney and gill septa) in *Isurus oxyrinchus* (Rafinesque, 1891) with nematoda larvae and were attributed likely to drancunculoids.

Santos *et al.* (2004) reported the occurrence of a *Pseudoanisakis* (Ascaridida: Acanthocheilidae) from the intestines of two rajids: *Rioraja agassizii* (Müller & Henle, 1841) and *Psammobatis extenta* (Garman, 1913) from Brazilian southwestern Atlantic waters. Alvarez *et al.* (2006) recorded six species of nematode, among them *A. simplex*, all of them in the lumen of stomach and intestine.

Knoff *et al.,* (2001) reported L_3 of *Anisakis* sp. in the stomach and spiral valve of 263 elasmobranchs of Brazil.

Guagliardo *et al*. (2009) found L_3 of *Anisakis sp*. in a bony fish *Seriolella porosa* (Guichenot, 1848) and Tanzola & Guagliardo (2004) reported L_3 of several anisakids in the intestinal lumen of cartilaginous fish, but no invasion of these in the body cavity. The *Anisakis* larvae found in the present study are morphologically similar and share the range of measurements with those parasitize silver warehou, *S. porosa,* from the same geographical area, San Matias Gulf (Guagliardo *et al.,* 2009).

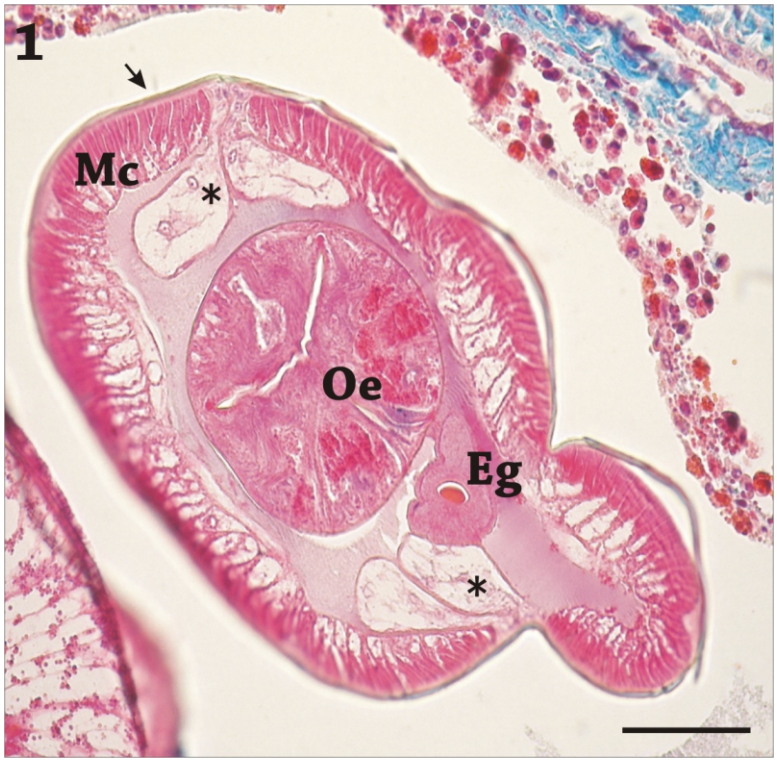

Figure1. High magnification of the anterior part of a third larva stage of *Anisakis sp.* Arrow shows the cuticle. Asterisks indicate the hypodermic cordons. **Mc:** muscular cells; **Oe:** oesophagus; **Eg:** excretory gland. Scale bar: 70 µm.

The body cavity of elasmobranch is rarely parasitized by nematodes (Caira & Healy, 2004). Moravec & Little (1988) reported two species of micropleurid nematodes in bull sharks and Caira & Healy (2004) found larvae of ascarid nematodes in elasmobranchs from the Gulf of California. Williams (1964) and Williams *et al.* (1970) stated that the high concentrations of urea in the body fluid and tissues of elasmobranch hosts made an inhospitable environment to the colonization of helminthes, such as acanthocephalans. This statement could be extrapolate to explain the few records of larval helminths parasitizing the body cavity and tissues of cartilaginous fishes to date (Díaz Andrade *et al.,* 2008).

Base on this background, the results exposed in this work constitute the first report of L_3 of *Anisakis sp*. in the body cavity of an elasmobranch, in particular *A. platana*, and show the capability of this anisakid to survive in the visceral mass of these hosts.

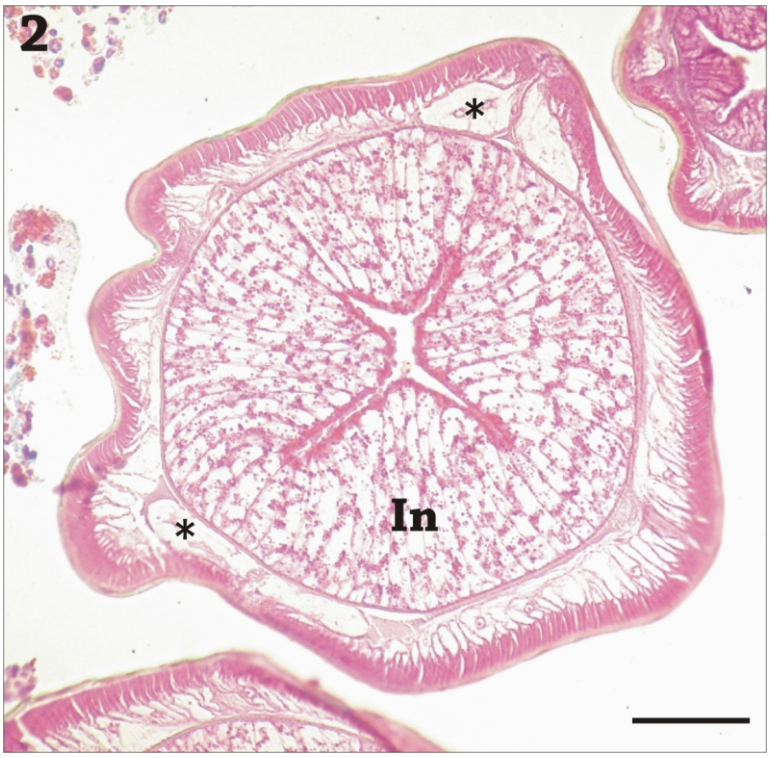

Figure2. High magnification of the middle part of a third larva stage of *Anisakis sp*. Asterisks indicate hypodermic cordons. **In:** intestine. Scale bar: 60 µm.

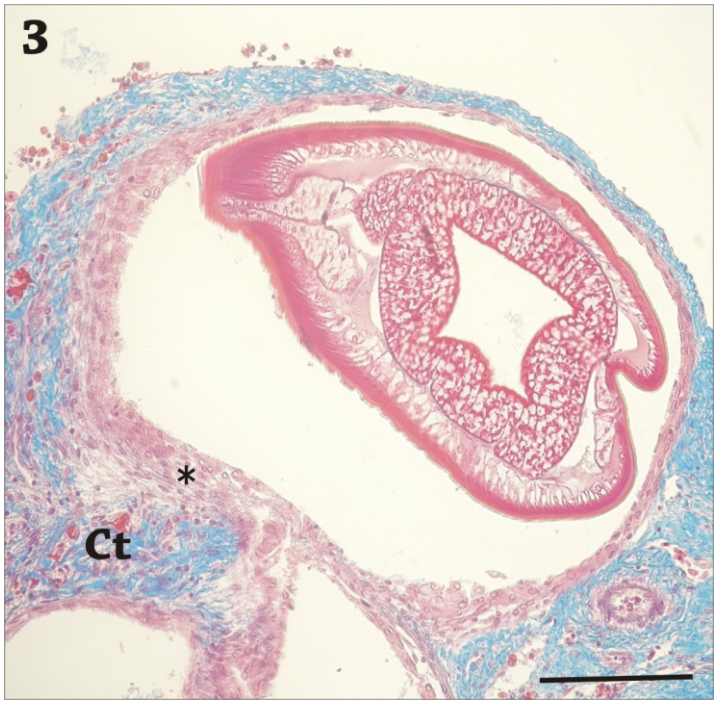

Figure3. Transverse section of a third larva stage of *Anisakis sp*. in visceral tissue of a juvenile male of *Atlantoraja platana*. Asterisk depicts inflammatory tissue. **Ct:** connective tissue. Scale bar: 130 µm.

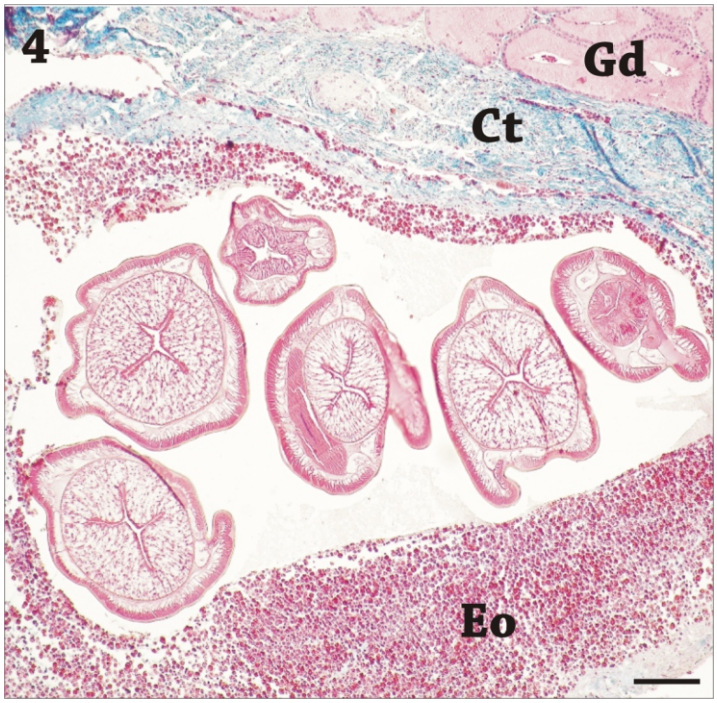

Figure4. General view of a third larval stage of *Anisakis sp*. in visceral tissue of a mature male of *A. platana*. **Eo:** epigonal organ; **Ct:** connective tissue; **Gd:** genital ducts. Scale bar: 150 µm.

ACKNOWLEDGEMENT

We thank to San Antonio Oeste and San Antonio Este fish-processing plants for providing us the samples. This work was supported by the SGCyT-UNS, PGI: 24/B173.

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Received June 29, 2015.
Accepted September 26, 2015.