



Research note

Notes on helminth parasites of tuna fishes (Scombridae) in Brazil

Notas sobre los helmintos parásitos del atún (Scombridae) en Brasil

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Abstract. Four species of Scombridae from the coast of Rio de Janeiro, southwest Atlantic Ocean were found to be parasitized with species of Didymozoidae (Digenea): *Thunnus atlanticus* (Lesson) a new host record for *Nephrodidymotrema ahi* Yamaguti, 1970, *Didymocystis neothunni* (Yamaguti, 1970), and *Didymozoon longicolle* (Yamaguti, 1970); *Thunnus obesus* (Lowe) parasitized by *Didymozoon longicolle* and reported as a new host for *Nephrodidymotrema ahi*; *Thunnus albacares* (Bonaterre) parasitized by *Nephrodidymotrema ahi*, and *Katsuwonus pelamis* (Linnaeus) by *Didymozoon longicolle*. Measurements, figures, and parameters of infection are presented.

Key words: *Thunnus atlanticus*, *Thunnus albacares*, *Thunnus obesus*, *Katsuwonus pelamis*, Digenea, Didymozoidae, Brazil.

Resumen. Cuatro especies de Scombridae de la costa de Río de Janeiro, suroeste del océano Atlántico, se reportan parasitados con diferentes especies de Didymozoidae (Digenea): *Thunnus atlanticus* (Lesson) un registro de nuevo huésped para *Nephrodidymotrema ahi* Yamaguti, 1970, *Didymocystis neothunni* (Yamaguti, 1970) y para *Didymozoon longicolle* (Yamaguti, 1970); *Thunnus obesus* (Lowe) parasitado por *Didymozoon longicolle*, se indica como nuevo huésped para *Nephrodidymotrema ahi*; *Thunnus albacares* (Bonaterre) parasitados por *Nephrodidymotrema ahi* y *Katsuwonus pelamis* (Linnaeus) por *Didymozoon longicolle*. Se presentan las mediciones, las figuras y los parámetros de la infección.

Palabras clave: *Thunnus atlanticus*, *Thunnus albacares*, *Thunnus obesus*, *Katsuwonus pelamis*, Digenea, Didymozoidae, Brasil.

The importance of research concerning parasite fauna of Brazilian tuna is due to the large number of specimens exported. In this paper, 4 species of Scombridae from the coast of Rio de Janeiro, which represent important economic resources, were studied for helminth parasites: *Thunnus albacares* (Bonaterre), *T. atlanticus* (Lesson), *T. obesus* (Lowe), and *Katsuwonus pelamis* (Linnaeus). Studies have confirmed the great migratory potential of tuna from different oceans (Arrizabalaga et al., 2002; Hallier, 2005; Arreguil et al., 2006). Madhavi and Ham (2000) affirmed that high vagility and endothermy require high metabolic energy which is met through foraging on large quantities of food items comprising crustaceans, fish, mollusks, and polychaetes that serve as intermediate and paratenic hosts for didymozoid and other helminth parasites. In studies

involving helminth parasites of Scombridae from Brazil, in the southwest Atlantic Ocean (Kohn et al., 2001; Kohn and Justo, 2008; Justo et al., 2008, 2009; Justo and Kohn, 2005, 2009, 2010), the most prevalent and abundant species belong to Didymozoidae Monticelli, 1888, which is in agreement with results reported by other authors in different parts of the world (Yamaguti, 1970; Ishii, 1935; Nikolaeva, 1985; Pozdnyakov, 1996; Madhavi and Ham 2000; Munday et al., 2003; Mladineo et al., 2011). The Didymozoidae, found predominantly in tropical and subtropical waters, are one of the most enigmatic groups of parasites of marine fish (Nikolaeva, 1985). These new data increase current knowledge regarding helminth parasites of tuna from the southwest Atlantic Ocean and expand the known geographic distribution.

Fish, 149 specimens, were collected and examined from January 2004 to April 2007: 38 individuals of *T. albacares*, 34-76 cm total body length and 0.550-7.8 kg

weight [15 males (39.4%) and 23 females (60.6%)]]; 45 individuals of *T. atlanticus*, 45-82 cm total body length and 1.3-6.0 kg [16 males (35.5%) and 29 females (64.5%)]]; 35 individuals of *T. obesus*, 42-80 cm total body length and 1.2-8.0 kg [21 males (60%) and 14 females (40%)]]; and 44 individuals of *K. pelamis*, 26-73 cm total body length and 1.0-8.0 kg [18 males (41%) and 26 females (59%)]. Fishes were obtained from local fishermen from the coastal zone of the state of Rio de Janeiro, off Cabo Frio, Brazil (22°52'46" S, 42°01'07" W). Helminths were released from cysts and fixed with or without compression in AFA (alcohol 93%, formalin 5%, acetic acid 2%), stained in alcoholic-acid carmine, dehydrated in an alcohol series, cleared in methyl salicylate, and mounted in Canada balsam. Measurements of slightly compressed worms are in micrometers, with the mean in parentheses followed by the number of specimens measured in brackets, where applicable. Parameters of infection as proposed by Bush et al. (1997) are given in Table 1. Confidence intervals (95%) were calculated assuming a binominal distribution, using software Quantitative Parasitology 3.0 (Rózsa et al., 2000). Representative specimens were deposited in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC), Rio de Janeiro, Brazil.

Nephrodidymotrematinae Yamaguti, 1970

Nephrodidymotrema ahi Yamaguti, 1970 (Figs. 1A, B)

Hosts and site of infection: encysted in pairs in kidney of *T. atlanticus* and *T. obesus* (new host records) and *T. albacares*.

Material examined: 28 specimens in three of 38 *T. albacares*, 21 specimens in 5 of 45 *T. atlanticus*, and three specimens in 2 of 35 *T. obesus*.

Voucher specimens deposited: CHIOC n° 37139 a-d, 37140 a-b, 37141 a-b, 37142.

Description (based on 9 specimens): round cysts containing a pair of worms (1 male and 1 female) are easily visualized by the bright yellow color of the cyst in the kidneys of the hosts. The body of both the male and female are divided into 2 free anterior regions and 1 posterior region fused on one side, such that male and female cannot be separated.

Male. Anterior region free, elongated, widened in the anterior third (Figs. 1A, B), measuring 1 900-3 200 (2 600) [n= 3] long by 220-400 (300) [n= 3] wide. Oral sucker terminal, 75-150 (96) [n= 4] long by 45-105 (64) [n= 4] wide, directly followed by the globular pharynx, 35-45 (39) [n= 4] in diameter. Short esophagus surrounded by gland cells, 100-120 [n= 2] long. Caeca extended into the posterior region. Posterior region of the body completely fused with posterior region of female. Single testis elongated and convoluted, situated in posterior region near base of the anterior margin. In 1 specimen, observation confirmed the testis to be divided into 2 branches: one long, bifurcated and the other short, with no bifurcation. Vas deferens distended near the intestinal bifurcation.

Female. Anterior region free and elongated, similar to the male (Figs. 1A, B), but differs in the presence of well-differentiated metraterm, 1 000-2 800 (1 800) [n= 4] long by 250-400 (316) [n= 4] wide. Oral sucker terminal, 62-75 (71) [n= 5] long by 40-65 (50) [n= 5] wide, directly followed by the globular pharynx, 30-80 (44) [n= 6] long by 27-55 (41) [n= 6] wide. Posterior region of female subglobular, with irregular grooves clearly observed in uncompressed worms (Fig. 1A), measuring 3 000-5 900 (4 200) [n= 8] long by 3 500-5 500 (4 600) [n= 8] wide. Ovary branched, 50-100 (63) [n= 8] wide. Vitellarium branched, 30-70 (52) [n= 9] wide. The uterus occupies all available space of posterior region. Opening of muscular metraterm ventral to oral sucker. Eggs 17-20 (19) [n= 9] long by 10-12 (12) [n= 9] wide.

Table 1. Prevalence (P), mean intensity (MI) and intensity range (IR) of species of Didymozoidae from *Thunnus atlanticus*, *T. albacares*, *T. obesus*, and *Katsuwonus pelamis* from Cabo Frio, Rio de Janeiro State, Brazil

	<i>Nephrodidymotrema ahi</i>			<i>Didymozoon longicolle</i>			<i>Didymocystis neothunni</i>		
	P % (CI)	MI (CI)	IR	P % (CI)	MI (CI)	IR	P % (CI)	MI (CI)	IR
<i>T. atlanticus</i>	11.1 (3.7-24.06)	4.20 (1.80-8.8)	1-12	8.9 (2.47-21.23)	30.00 (2.00-69.50)	2-92	4.4 (0.54-15.15)	8.00 (2.00-8.00)	2-14
<i>T. albacares</i>	10.5 (2.94-24.81)	7.0 (1.50-15.25)	1-18	-	-	-	-	-	-
<i>T. obesus</i>	11.4 (3.2-26.74)	2.00 (1.25-2.50)	1-3	11.4 (3.2-26.74)	17.50 (5.00-25.50)	4-30	-	-	-
<i>K. pelamis</i>	-	-	-	25.0 (14.2-39.69)	16.18 (9.82-29.45)	2-58	-	-	-

CI= Confidence interval.

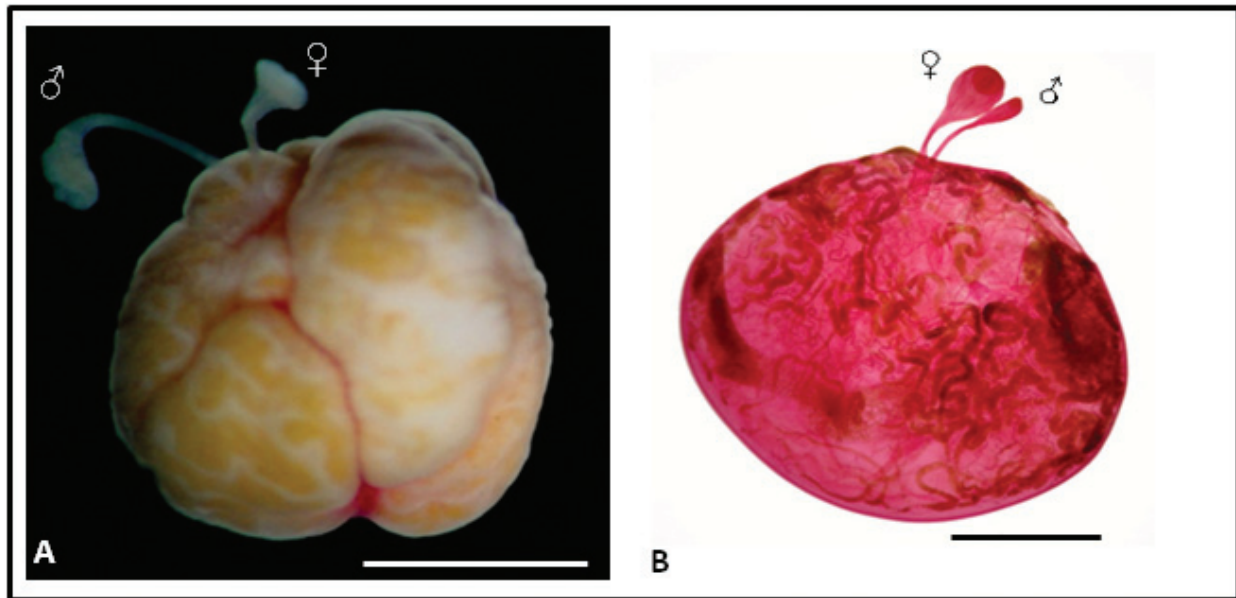


Figure 1. Photomicrographs of *Nephrodidymotrema ahi* Yamaguti, 1970. A, male and female, without compression, showing the irregular grooves on the posterior region of the female. Bar: 2 mm. B, compressed specimens stained in alcoholic-acid carmine. Bar: 2 mm.

Remarks. The genus *Nephrodidymotrema* is characterized by the complete fusion of the posterior regions of the male and female and by the peculiar habitat, the kidney.

Nephrodidymotrema ahi, type and only species of the genus, was described in 1970 by Yamaguti, isolated from kidneys of *T. albacares* (= *Neothunnus macropterus*) from Hawaii, Pacific Ocean. In 1996, Pozdnyakov reported this species parasitizing *T. alalunga* and *T. albacares*, also in the Pacific Ocean. Specimens of *N. ahi* were collected in the same site of the type host, *T. albacares*, and of 2 new hosts, *T. obesus* and *T. atlanticus*. The morphology and measurements of these specimens agree with those given in the original description. Yamaguti (1970) reported finding only a single testis, and mentioned that the distal end of the testis was bifid only in the type specimen. In the present paper, among the specimens studied, only 1 presented 2 testes: one long, divided into 2 branches, and one short with no bifurcation.

Didymozoinae Monticelli, 1888

Didymocystis neothunni (Yamaguti, 1970) Pozdnyakov, 1996 (Figs. 2A, B)

Host and site infection: Encysted in pairs on tongue of *T. atlanticus* (new host record).

Material examined: 16 specimens in 2 of 45 *T. atlanticus*.

Voucher specimens deposited: CHIOC: 37128 a-d, 37129.

Description (based on 10 specimens): Round cysts containing 2 identical hermaphroditic parasites. Body divided into 2 distinct regions: a spatulated anterior region

attached at the ventral part of posterior region of the body, measuring 3100-3900 (3700) long and maximum width 600-1000 (900) at the esophageal level. Oral sucker muscular and subglobular, with ventroterminal aperture, 220-275 (253) [n= 4] long by 160-225 (152) [n= 4] wide. Pharynx poorly developed. Esophagus 950-1 800 (1 337) [n= 4] long. Caeca narrow in the anterior region and enlarged in the posterior region of body. Posterior region of body hemispheric, with greater length than width, ventrally flattened and dorsally convex, measuring 3 500-4 200 (4 140) long by 2 800-4 500 (3 600) wide. Testes tubular and sinuous, 1 on each side near anterior margin of the posterior region of the body. Vas deferens extends forward in the anterior region alongside the metraterm. Ovaries paired, tubular and very thin. Vitellarium consisting of a single tubular gland, long, unbranched, across the entire body's posterior region. Uterus fills all available space of posterior region of the body. Opening of muscular metraterm ventral to oral sucker. Eggs embryonated, 17-20 (18) [10] long by 12-13 (12) [10] wide.

Remarks. Yamaguti (1970) erected *Univitellodidymocystis* by the type species *U. neothunni* isolated from *T. albacares* (= *Neothunnus macropterus*) and *T. obesus* (= *Parathunnus sibi*) from Hawaii, characterized by the presence of a single, unbranched vitelline gland extending the entire length of the posterior region of the body. Pozdnyakov (1990) considered *Univitellodidymocystis* as synonymous with *Didymocystis* and later (Pozdnyakov, 1996) proposed the new combination *Didymocystis*

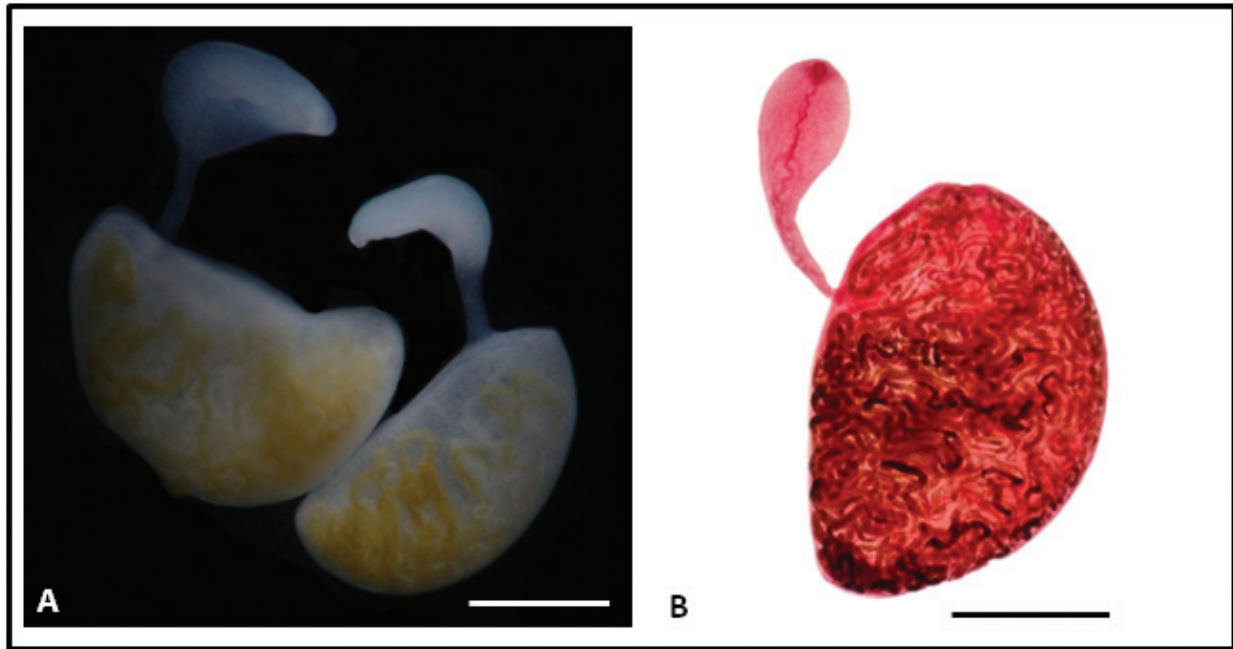


Figure 2. Photomicrographs of *Didymocystis neothunni* (Yamaguti, 1970) in saline. A, without compression. Bar: 2 mm. B, compressed specimen stained in alcoholic-acid carmine. Bar: 1 mm.

neothunni. In the revision “Helminths of Scombrid-like Fishes of the World’s Oceans”, Pozdnyakov (1990) referred to *Thunnus thynnus* (Linnaeus) as a host for *D. neothunni*, but made no further reference to it in papers that were published later. In a study of the parasites of *T. thynnus*, Mladineo et al. (2008, 2011) did not refer it as a host for *D. neothunni*. The earlier reference (Pozdnyakov, 1990) seems to have been a mistake because *T. thynnus* has never been indicated as a host for this species in subsequent publications. The current specimens present the same morphometrical features as in the original description, and constitute a new report in *T. atlanticus*, and a new geographical report from Brazil, southwest Atlantic Ocean. From the same species of host and locality, another species, *Didymocystis lamotheargumedei* Kohn and Justo, 2008, was described recently.

Didymozoon longicolle Ishii, 1935 (Fig. 3)

Hosts and site of infection: Encysted in pairs on branchial filaments of *T. atlanticus* (new host record), *T. obesus*, and *K. pelamis*.

Material examined: 117 specimens in three of 45 *T. atlanticus*, 69 specimens in 4 of 35 *T. obesus*, and 177 specimens in 11 of 44 *K. pelamis*.

Voucher specimens deposited: CHIOC: 37133, 37134 a-b, 37135, 37136, 37137 a-c.

Description (based on 10 specimens): Cylindrical cysts, bright yellow in color, longitudinally disposed in gill

filaments, containing 2 hermaphroditic individuals similar in form and size. Body divided into 2 regions. Anterior region filiform, wide in the esophageal region, measuring 600-3 600 (1 800) [n= 9] long by 100-300 (250) [n= 9] wide. Oral sucker terminal, oval and muscular or rather poorly developed, and 72-102 (88) [n= 7] long by 50-87 (70) [n= 7] wide, directly followed by pharynx, 35-62 (50) [n= 3] long by 35-55 (45) [n= 3] wide, and a long esophagus. Posterior region cylindrical, conical anteriorly and rounded in the posterior extremity, 3 800-11 000 (6 900) long by 220-700 (500) wide. 2 testes tubular, tortuous, located on the anterior margin of the posterior region, 1 on each side. Vas deferens parallel to the metraterm in anterior region of body. Ovaries formed by 2 elongate sinuous branches originating at different levels near the anterior margin. Vitellarium single, tubular, and sinuous. Uterus fills all available space of posterior region of the body without forming an egg reservoir. Metraterm strongly muscular, with genital pore lateral to oral sucker. Eggs 15-17 (16) [n= 10] long by 12 (12) [n= 10] wide.

Remarks. The morphometric characteristics of *D. longicolle* isolated from *T. atlanticus* (new host), *T. obesus*, and *K. pelamis* agree with those of the original description (Ishii, 1935) from specimens isolated from the gills of *K. pelamis* (= *K. vagans*), *Thunnus orientalis* Temminck and Schlegel, and *Scomber japonicus* Houttuyn from Japan and with the description of specimens isolated from the gills of *T. albacares* (= *Neothunnus macropterus*) and *T.*



Figure 3. Photomicrographs of *Didymozoon longicolle* Ishii, 1935 stained in alcoholic-acid carmine. Bar: 0.5 mm.

obesus (= *Parathunnus sibi*), from Hawaii (Yamaguti, 1970). Nikolaeva and Parukhin (1968) and Baudin-Laurencin (1971) reported *D. longicolle* as a parasite of *T. albacares* from the Gulf of Mexico. This species was also reported by Williams Jr. and Bunkley-Williams (1996) from *S. japonicus* (Massachusetts) and *T. albacares* (Gulf of Mexico). This genus was reported in Brazil by Alves and Luque (2006) from *K. pelamis* as *Didymozoom* sp. Recently, *D. longicolle* was reported in *T. alalunga* from the Balearic Sea (Mele et al., 2010). However, the prevalence of *D. longicolle* in *T. alalunga* reported by that study was higher (63.3%) than those observed in the present study (*T. atlanticus*: 8.9%; *T. obesus*: 11.4%, and *K. pelamis*: 25%) and that reported by Mladineo et al. (2008) (*T. thynnus*: 1.1%). As well, *D. longicolle* was considered by Meles et al. (2010) to be a dominant species.

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Literature cited

- Alves, D. R. and J. L. Luque. 2006. Ecologia das comunidades parasitárias de cinco espécies de escombrídeos (Perciformes: Scombridae) do litoral do Estado do Rio de Janeiro, Brasil. *Revista Brasileira Parasitologia Veterinária* 15: 167-181.
- Arreguil, I., H. Arrizabalaga and J. M. de la Serna. 2006. Preliminary approach to the experimental design of tagging campaigns for movement rates estimation of east atlantic bluefin tuna. *Collective Volume of Scientific Papers ICCAT*, 59(3): 769-788
- Arrizabalaga, H., V. L. Rodas, V. O. Zárate, E. Costas and A. González-Garcés. 2002. Study on the migrations and stock structure of albacore (*Thunnus alalunga*) from the Atlantic Ocean and the Mediterranean Sea based on conventional tag release - recapture experiences. *Collective Volume of Scientific Papers ICCAT* 54:1479-1494.
- Baldin-Laurencin, F. 1971. Crustacés et helminthes parasites de l'albacore (*Thunnus albacares*) du Golfe de Guinée. *Documents scientifiques. Centre de Recherche Océanographiques Abidjan (ORSTOM)* 2:11-30.
- Bush, A. O., K. D. Lafferty, J. M., A. Lotz and W. Shostak. 1997. Parasitology meets ecology on its own terms: Margolis et al., revisited. *Journal of Parasitology* 83:575-583.
- Hallie, J. P. 2005. Movements of tropical tunas from the tuna

- associated baitboat fishery of Dakar and from betyp and historical tagging operations in the Atlantic Ocean. *Collective Volume of Scientific Papers ICCAT* 57:76-99.
- Ishii, N. 1935. Studies on the family Didymozoidae Monticelli, 1888. *Japanese Journal of Zoology* 6:279-335.
- Justo, M. C. N. and A. Kohn. 2009. Trematoda, Digenea, Didymozoidae, *Coeliotrema thynni* Yamaguti, 1938: First record in Brazil with three new host records. *Check List* 5:436-438.
- Justo, M. C. N. and A. Kohn. 2005. Didymozoidae (Digenea) parasites of Scombridae (Actinopterygii) from Rio de Janeiro coast, Brazil. *Revista Brasileira de Zoociências* 7:333-338.
- Justo, M. C. N. and A. Kohn. 2010. Trematoda, Digenea, Didymozoidae, *Wedlia retrorbitalis* (Yamaguti, 1970) and *Wedlia submaxillaris* (Yamaguti, 1970): first record in South America and the Atlantic Ocean. *Check List* 6:387-389.
- Justo, M. C. N., R. Tortelly, R. C. Menezes and A. Kohn. 2008. First record in South America of *Didymosulcus palati* and *Didymosulcus philobranchiarca* (Digenea, Didymozoidae) with new host records and pathological alterations. *Memórias do Instituto Oswaldo Cruz* 103:207-210.
- Justo, M. C. N., R. Tortelly, R. C. Menezes and A. Kohn. 2009. First record in South America of *Koellikerioides internogastricus* (Digenea, Didymozoidae) with new host record and pathological alterations. *Veterinary Parasitology* 161:158-161.
- Kohn, A. and M. C. N. Justo. 2008. *Didymocystis lamotheargumedoi* n. sp. (Digenea: Didymozoidae) a parasite of three species of scombrid fishes. *Revista Mexicana de Biodiversidad* 9:9S-14S.
- Kohn, A., A. L. Santos and M. D. F. Baptista-Farias. 2001. Report of *Didymocystis wedli* Ariola, 1902 (Digenea; Didymozoidae) from *Thunnus albacares* in Brazil. *Memórias do Instituto Oswaldo Cruz* 96:951-954.
- Madhavi, R. and B. K. Ram, 2000. Community structure of helminth parasites of the tuna, *Euthynnus affinis*, from the Visakhapatnam coast, Bay of Bengal. *Journal Helminthology* 74:337-342.
- Mele, S., P. Merella, D. Macias, M. J. Gómez, G. Garippa and F. Alemany. 2010. Metazoan gill parasites of wild albacore *Thunnus alalunga* (Bonaterre, 1788) from the Balearic Sea (western Mediterranean) and their use as biological tags. *Fisheries Research* 102:305-310.
- Mladineo, I, J. Žilić and M. Čanković. 2008. Health survey of Atlantic Bluefin Tuna, *Thunnus thynnus* (Linnaeus, 1758), reared in Adriatic cages from 2003 to 2006. *Journal of the World Aquaculture Society* 39:281-289.
- Mladineo, I., T. Šegvić and M. Petrić. 2011. Do captive conditions favor shedding of parasites in the reared Atlantic bluefin tuna (*Thunnus thynnus*)? *Parasitology International* 60:25-33.
- Munday, B.L., Y. Sawada, T. Cribb and C.J. Hayward. 2003. Diseases of tunas, *Thunnus* spp. *Journal of Fish Diseases* 26:187-206.
- Nikolaeva, V. M. 1985. Trematodes Didymozoidae fauna, distribution and biology. *In Parasitology and Pathology of Marine Organisms of the World Ocean*, NOAA, W. J. Hargis, Jr. (ed.). *Technical Report*, NMFS 25:667-672.
- Nikolaeva, V. M. and A. M. Parukhin. 1968. To the study of fish helminths in the Gulf of Mexico. *In Investigations of Central American Seas*, E. B. Jankovskaya (ed.). *Naukova Dumka*, Kiev 2:126-149.
- Pozdnyakov, S. E. 1990. [Helminths of Scombrid-like Fishes of the World's Oceans]. DVO ANSSSR, Vladivostok. 82 p. (In Russian)
- Pozdnyakov, S. E. 1996. [Trematodes suborder Didymozoata]. *Tikhookeanskii Nauchno-Issledovatel'skii Rybokhozyaistvennyi Tsent*, Vladivostok. 319 p. (In Russian)
- Rósza, L., Reiczigel, J. and G. Majoros. 2000. Quantifying parasites in samples of hosts, *Journal of Parasitology* 86:228-232.
- Williams, Jr. E. and L. Bunkley-Williams. 1996. Parasites of offshore big game fishes of Puerto Rico and the Western Atlantic. *Antillean College Press*. 382 p.
- Yamaguti, S. 1970. The digenetic trematodes of Hawaiian fishes. *Keigaku Publishers Co., Tokyo*. 436 p.