Neotrop. Helminthol., 8(2), 2014

ISSN: 2218-6425 impreso / ISSN: 1995-1043 on line

aphia
ASOCIACIÓN PERUANA DE HELMINTOLOGÍA E INVERTEBRADOS AFINES

ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

PARASITE DIVERSITY OF *OSTEOGLOSSUM BICIRRHOSUM*, AN OSTEOGLOSSIDAE FISH FROM AMAZON

DIVERSIDAD DE PARÁSITOS DE *OSTEOGLOSSUM BICIRRHOSUM*, UN PEZ OSTEOGLOSSIDAE DE LA AMAZONÍA

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Suggested citation: Rodrigues, MNG, Dias, MKR, Marinho, RGB & Tavares-Dias, M. 2014. Parasites diversity of *Osteoglossum bicirrhosum*, an Osteoglossidae fish from amazon. Neotropical Helminthology, vol. 8, n°2, jul-dec, pp. 383-391.

Abstract

We investigated the ecological relationships between parasites and the host silver arowana *Osteoglossum bicirrhosum* Cuvier, 1829 from Amazonian basin, in Brazil. A total of 1,570.539 parasites belonging to different taxa were found including *Ichthyophthirius multifiliis*, *Piscinoodinium pillulare* (Protozoa), *Gonocleithrum aruanae*, *Gonocleithrum planacrus*, *Gonocleithrum coenoideum* (Monogenoidea) and *Camallanus acaudatus* (Nematoda); all had an aggregated dispersion. The Brillouin diversity index was 0.11 ± 0.21, evenness was 0.08 ± 0.15 and species richness was 3.2 ± 0.5. There was positive correlation between the intensity of *I. multifiliis* and host size, as well as between the intensity of *Gonocleithrum* spp. and the relative condition factor of hosts. This is the first report on parasitic diversity indices in wild *O. Bicirrhosum.* The parasites community was characterized by low species diversity and species richness, and by high prevalence of ectoparasites species.

Keywords: Amazon, condition factor, fish, parasites.

Resumen

Este estudio investigó las relaciones ecológicas entre los parásitos y el hospedero arauana *Osteoglossum bicirrhosum* Cuvier, 1829 de la cuenca del Amazonas, en Brasil. Un total de 1,570.539 parásitos pertenecientes a diferentes taxones se encontraron como *Ichthyophthirius multifiliis*, *Piscinoodinium pillulare* (Protozoa), *Gonocleithrum aruanae*, *Gonocleithrum planacrus*, *Gonocleithrum coenoideum* (Monogenoidea) y *Camallanus acaudatus* (Nematoda), que tenían una dispersión agregada. El índice de diversidad de Brillouin fue 0,11 ± 0,21, la uniformidad fue 0,08 ± 0,15 y la riqueza de especies fue 3,2 ± 0,5. Hubo correlación positiva entre la intensidad de *I. multifiliis* y el tamaño del hospedero, así como entre la intensidad de *Gonocleithrum* spp. y el factor de condición relativa de los hospederos. Este primer informe sobre el índices de diversidad parasitarias en *O. bicirrhosum* muestra que la comunidad de parásitos se caracterizó por una baja diversidad y riqueza de especies, y por la alta prevalencia de especies de ectoparásitos.

Palabras clave: Amazonas - factor de condición - peces - parásitos.

INTRODUCTION

Among the fish of Amazon River and in the Rupununi and Oyapock Rivers system is the silver arowana *Osteoglossum bicirrhosum* Cuvier, 1829 (Osteoglossidae), a large Osteoglossiformes that can reach over 1 m of length and 5 kg of body weight. *O. bicirrhosum* is a benthopelagic fish, with diurnal and sedentary habits and omnivore with a tendency to carnivory. In adult age, it feed on aquatic and terrestrial invertebrates (insects, spiders and decapods) and on fish, but when juveniles it feed on phytoplankton, zooplankton and seeds. During the Amazonian flood season it is more abundant in the flooded forest (Soares *et al*., 2011; Tavares-Dias *et al*., 2014), but in the drainage season it inhabits marginal rivers and lakes and that is when it is caught. This fish has a great important for the local fishery, due to its use in feeding of the human populations (Tavares-Dias *et al*., 2014), and the fry are very popular worldwide as ornamental fish (Chaves *et al*., 2005; Ortiz & Iannacone, 2008). In China, *O. bicirrhosus* is known as dragon fish or god fish and their fry reach high market values.

Morphological studies of parasites in *O. bicirrhosum* from central Amazon (Brazil) described *Caballerotrema aruanense* Thatcher, 1980 (Thatcher, 1980), *Gonocleithrum aruanae* Kritsky & Thatcher, 1983; *Gonocleithrum coenoideum* Kritsky & Thatcher, 1983; *Gonocleithrum cursitans* Kritsky & Thatcher, 1983; *Gonocleithrum planacroideum* Kritsky & Thatcher, 1983; *Gonocleithrum planacrus* Kritsky & Thatcher, 1983; *Gonocleithrum planacroideum* Kritsky & Thatcher, 1983 (Kritsky & Thatcher, 1983) and *Camallanus acaudatus* Ferraz & Thatcher, 1990 (Ferraz & Thatcher, 1990). Recently, infections by non-identified larvae of Nematoda, *C. aruanense*, *Sebekia* sp., *Telethecium nasalis* Kritsky, Van Every & Boeger 1996, *G. aruanae* and *Argulus* sp. were reported for wild *O. bicirrhosum* from central Amazon (Pelegrini *et al*., 2006; Lemos *et al*., 2012; Tavares-Dias *et al*., 2014). Vazquez *et al.* (2007) reported *G. cursitans*, *Trichodina* sp. and *C. acaudattus* in *O. bicirrhosum* farmed in Peruvian Amazon. However, has not been studied the diversity from parasitic community and infracommunity of *O. bicirrhosum* population.

Studies on the parasitic community of fish allow us to obtain important information not only about the host, but also about the environment in general. Environmental changes are useful to explain the presence or absence of certain species of parasites, besides explaining the rates of parasitism in hosts (Takemoto *et al.*, 2009; Silva *et al.*, 2011; Tavares-Dias *et al*., 2014). Moreover, since most parasites have a complex life-cycle, they can be indicators of changes occurring in the environment and in the fish community (Takemoto *et al*., 2009; Silva *et al*., 2011; Tavares-Dias *et al*., 2014), because frequently the transmission of parasites involve predator-prey interactions. Therefore, parasites can also provide information on the host population structure, environmental stressors, trophic interactions and other conditions. This study investigated the diversity of parasites in *O. bicirrhosum* of a tributary from Amazon River system, in eastern Amazon, Brazil.

MATERIAL AND METHODS

Fish and sampling site

From April to October 2010, 117 specimens of silver arowana *O. bicirrhosum* (34.0 to 56.5 cm and 246.0 to 1254.0 g) were caught at Preto River basin (00° 10'38”S and 051º 33'034'W), in the municipality of Magazão, State of Amapá (eastern Amazon, Northern Brazil) for parasitological analysis. All fish were caught with appropriate nets (Ibama, 2007). The Preto River basin) is a tributary from Amazon River system levels of dissolved oxygen ranging from 2.84 to 5.59 mg/L; pH from 4.84 to 5.55; ammonia from 0.56 to 0.68 mg/L and phosphate from 0.13 to 0.16 mg/L. The features of these aquatic parameters may be related to natural phenomena of increased organic matter in the tributary waters and/or may suffer the influence of the logging activity in the area (Silva *et al.*, 2001). Regional vegetation consists of plants characteristics of floodplain forests and periodically flooded herbaceous fields, composed mainly of various macrophyte species.

Procedures for collection and analysis of parasites

Collected fish were weighed (g), measured for total length (cm), each individual was macroscopically evaluated as for body surface, mouth, eyes, opercula and gills were removed to collect ectoparasites. Gastrointestinal tract was removed to collect endoparasites. All parasites were collected, fixed, stained for identification (Eiras *et al*., 2006) and quantified (Tavares-Dias *et al*., 2001a,b). The ecological terms adopted were those recommended by Rohde *et al.* (1995) and Bush *et al.* (1997).

The Brillouin index (*HB)*, evenness (*E*) and species richness were calculated for the parasite component community, by using the Diversity software (Pisces Conservation Ltd., UK). The dispersion index (ID) and the discrepancy index (D) were calculated using the Quantitative Parasitology 3.0 software, in order to detect the distribution pattern of each parasite infracommunity (Rózsa *et al.*, 2000) in species with prevalence 10%. The significance of ID for each parasite species was tested using *d*-statistic (Ludwig & Reynolds, 1988).

Data of body weight (g) and total length (cm) were used to calculate the factor of relative condition (Kn) of fish (Le-Cren, 1951), which was compared to the standard value (Kn = 1.0), by the t-test. Spearman correlation coefficient (rs) was used to determine correlations of host length with the *HB*, *E*, species richness and parasites abundance (Zar, 2010).

The potential hydrogen (pH), the temperature and the levels of dissolved oxygen (DO) were determined using specific digital devices. The mean water temperature was 29.8 ± 0.9; mean dissolved oxygen was 3.1 ± 0.7 and the mean pH was 5.01 ± 0.7.

RESULTS

From 117 specimens of silver arowana *O. bicirrhosum* necropsied, 98.3% had their gills parasitized by protozoans *Ichthyophthirius multifiliis* Fouquet, 1876 (Ciliophora) and *Piscinoodinium pillulare* (Schäperclaus, 1954) Lom, 1981 (Dinoflagellida); *Gonocleithrum aruanae*, *Gonocleithrum planacrus* and *Gonocleithrum coenoideum* (Monogenoidea) and their intestine by *Camallanus acaudatus* (Camallanidae). The highest dominance was of protozoans *I. multifiliis* and the lowest dominance was of *C. acaudatus* (Table 1). These

Table 1. Parasites on *Osteoglossum bicirrhosum* from Amazon river system in Northern Brazil. TNP: Total number of parasites; RD: Relative dominance.

Parameters	*I. multifiliis*	*P. pillulare*	*Gonocleithrum* spp.	*C. acaudatus*
Examined fish	117	117	117	117
Parasitized fish	114	15	115	8
Prevalence (%)	97.4	12.8	98.3	6.8
Mean intensity	12,300.2	10,916.6	39.6	1.6
Mean abundance	11,985.8	1399.6	38.9	0.1
Range of intensity	1,316-35,037	3,445-34,428	3-356	1-3
TNP	1.402,225	163,749	4,552	13
RD	0.893	0.104	0.003	-

parasites showed a typical aggregated distribution pattern the higher discrepancy values for infection by *P. pillulare* (Table 2).

The Brillouin diversity index (*HB*) was 0.11 ± 0.21, evenness (E) was 0.08 ± 0.15 and mean species richness was low (3.2 ± 0.5 parasites per host). The length of the hosts show not correlation with the *HB* (rs = -0.071, p = 0.449), species richness (rs = 0.065, p= 0.491) and *E* (rs = -0.072, p = 0.449). Hosts parasitized by four parasite species predominated, *Gonocleithrum* spp. and *I. multifiliis*.

The Kn was not different (p=0.989) from standard Kn (Kn = 1.00), and there are a week positive correlation from Kn of the hosts with the intensity of monogenoideans species (Figure 1). There was also a week positive correlation of the body length and weight with the intensity of *I. multifiliis* on the fish gills (Figure 2). However, no correlation from intensity of *I. multifiliis* (rs = 0.017, p = 0.861) with the Kn of the hosts was observed.

Table 2. Dispersion index (DI), *d* statistic and discrepancy index (D) for the parasite species on *Osteoglossum bicirrhosum* from Amazon river system in Northern Brazil.

Parasites	DI	*d*	D
I. multifiliis	1.522	391.4	0.321
P. pillulare	3.089	433.5	0.906
Gonocleithrum spp.	3.223	75.8	0.369

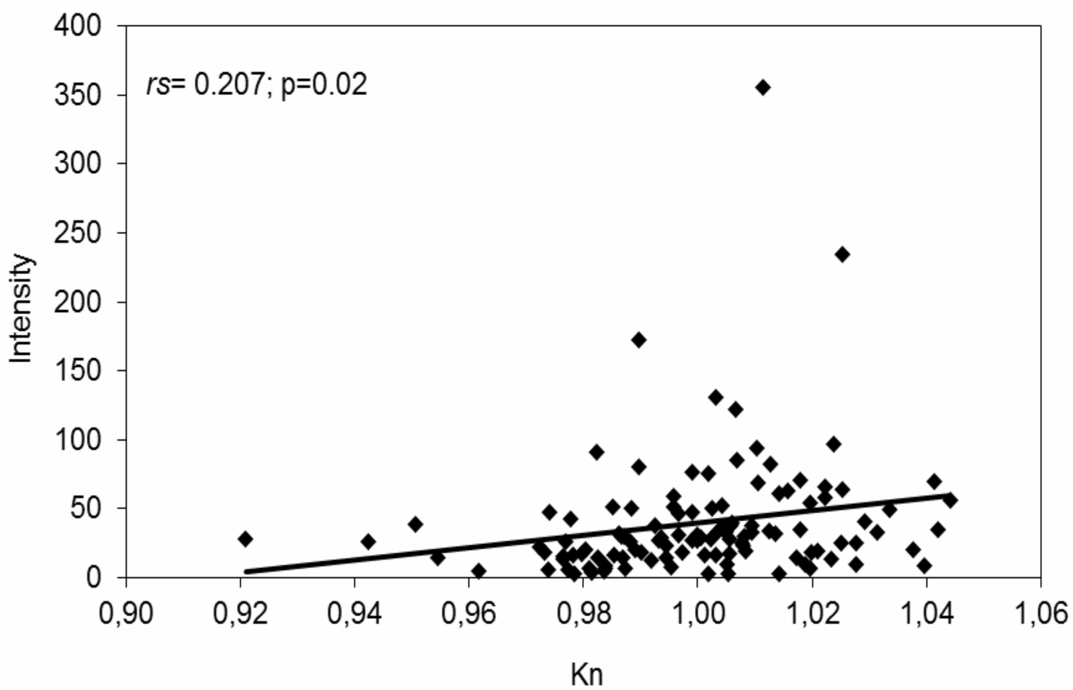

Figure 1. Correlation between the relative condition factor (Kn) and the intensity of *Gonocleithrum* spp. on the gills of *Osteoglossum bicirrhosum* (N=115) from Amazon river system in Northern Brazil.

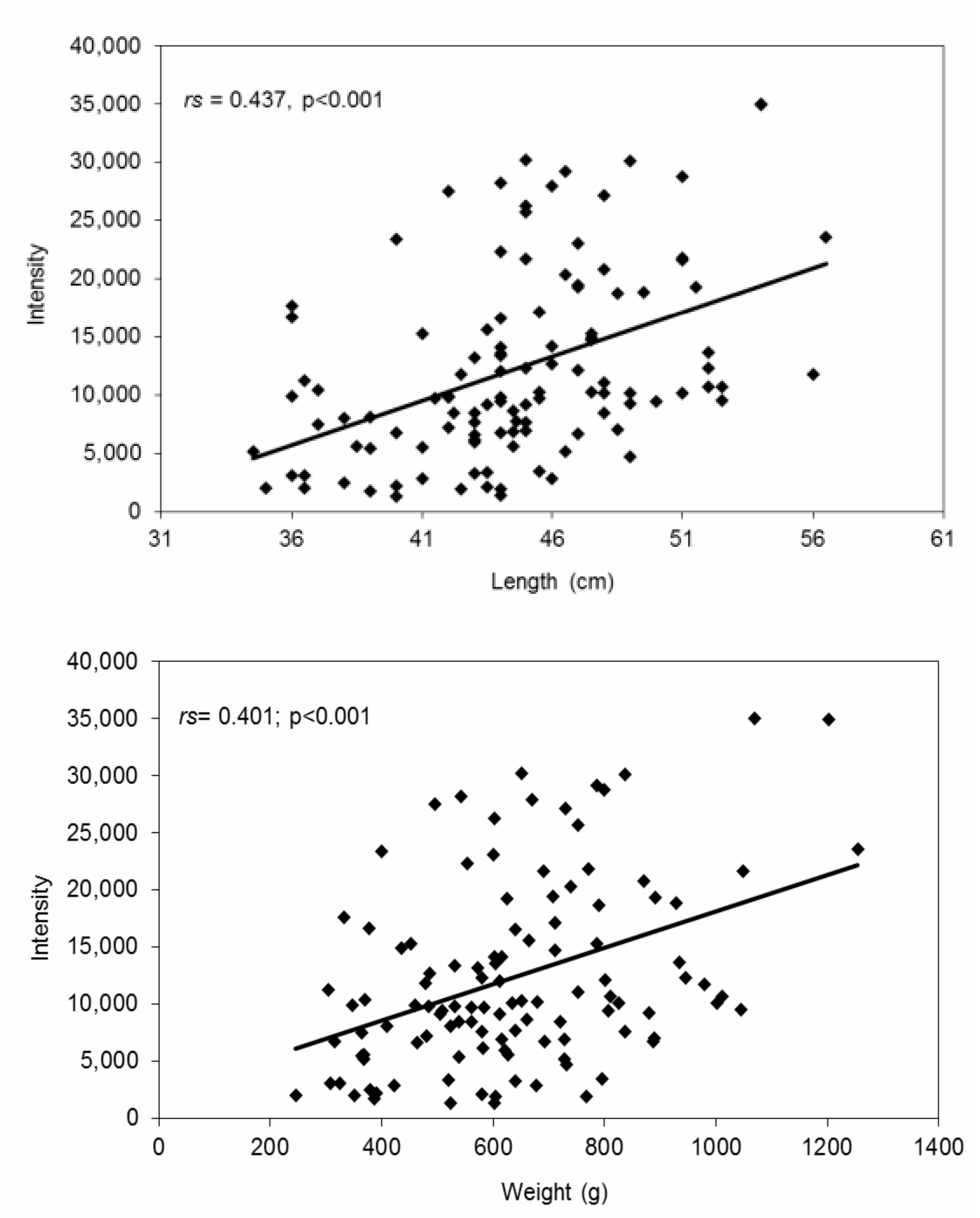

Figure 2. Correlation between the intensity of *Ichthyophthirius multifiliis* and the total length and body weight on *Osteoglossum bicirrhosum* (N=114) from Amazon river system in Northern Brazil.

DISCUSSION

The parasitic community of *O. bicirrhosum* showed low diversity, consisting of two Protista, two Monogenoidea and one Nematoda. This highest richness of ectoparasites (five species) reflects environmental conditions that were favorable to their transmission, because they did not need intermediate hosts. The low richness of endoparasites was due to the living habits of *O. bicirrhosum*, which occupies the second trophic level in the food chain (Soares *et al*., 2011; Tavares-Dias *et al*., 2014). In contrast, Lemos *et al*. (2012) reported that the community of *O. bicirrhosum* consisting of *T. nasalis*, G. *aruanae* and *Argulus* sp., while Tavares-Dias *et al*. (2014) reported only monogenoidean *G. aruanae*. The diversity of species in parasites communities is result, among all factors, from interactions between the evolutionary history and the host ecology (Takemoto *et al*., 2009; Silva *et al*., 2011; Tavares-Dias *et al*., 2014). Infection by *I. multifiliis*, *P. pillulare* and monogenoideans *Gonocleithrum* spp. presented aggregated dispersion, a pattern of parasitic distribution that is common in different fish species and caused by heterogeneity of hosts and parasites (Poulin, 2013). This was the first study concerning to pattern of parasites distribution for this important Amazonian fish.

Protozoans are common and dangerous parasites of farmed fish (Tavares-Dias *et al*., 2001a; Tavares-Dias *et al*., 2010). However, they may also infect fish from natural environment, and high infection rates occur due to aggregation and life habit of hosts. In *O. bicirrhosum* gills, the high levels of infection by *I. multifiliis* were found and the parasitic intensity increased with body growth of hosts. Bigger fish have a greater gill surface area for the multiplication of these protozoans, besides spending more time looking for food, which increases their exposure to parasites in the environment (Omeji *et al*., 2010). Hence, in these ectoparasites populations, there is generally a positive relationship between the body size and infection intensity.

Low of infection levels by *P. pillulare* were observed on the gills of wild *O. bicirrhosum*, but these levels were higher than the described for wild *Carnegiella martae* Myers, 1927 (Tavares-Dias *et al*., 2010). In contrast, these infection levels were lower than the reported for farmed fish (Tavares-Dias *et al*., 2001a), as expected. Environment features and different species of hosts, which differ in susceptibility to infection regarding the immune system, behavior, microhabitat and others factors, caused such differences in these infection levels by protozoans. Therefore, is necessary to understand the epidemiology features of *P. pillulare*, including the factors that can to affect its transmission among the fish hosts. This is the first ecological survey reporting the complete parasite fauna of *O. bicirrhosum* from a natural habitat. Moreover, this is the first record of *I. multifiliis* and *P. pillulare*.

On the gills of *O. bicirrhosum* from Preto River, in eastern Amazon, the levels of infection by monogeneans *Gonocleithrum* spp. were similar to this host from central Amazon parasitized by G. *aruanae* (Lemos *et al*., 2012). However, they were lower than the infection levels by monogeneans *Cosmetocleithrum* spp. on the gills of *Oxydoras niger* Valenciennes, 1821 from central Amazon (Silva *et al*., 2011). Monogeneans of the genus *Gonocleithrum* (*G. aruanae*, *G. coenoideum*, *G. cursitans*, *G. planacroideum*, *G. planacrus* and *G. planacroideum*) were first described in *O. bicirrhosum* from Solimões river basin, in Brazil (Kritsky & Thatcher, 1983), but the life cycle of these parasites is unknown. Monogenoideans are parasites with a direct life cycle and the rates of reproduction related to environment conditions, because they are often found in fish from lentic environments, which facilitate their transmission (Takemoto *et al*., 2009; Silva *et al*., 2011; Tavares-Dias *et al*., 2014). Moreover, infection rates by such parasites may vary with the physiological and immunological status of hosts.

The nematodes are endohelminths commonly found in wild populations because they make a significant part of the parasitic fauna of different fish from several environments (Pelegrini *et al*., 2006; Takemoto *et al*., 2009; Silva *et al*., 2011; Luque *et al*., 2011). In *O. bicirrhosum*, a bentophelagic fish (Santos & Brasil-Sato, 2006; Soares *et al*., 2008), the levels of infection by *C. acaudatus* were low and similar to those described for the benthic fish *O. niger* from the Solimões River infected by *Cucullanus grandistomis* Ferraz & Thatcher, 1988 (Silva *et al*., 2011a). Pelegrini *et al.* (2006) reported a high prevalence and intensity of unidentified Nematoda larvae (73.3%) for *O. bicirrhosum* from central Amazon. The *O. bicirrhosum* is an omnivorous fish that feeds on aquatic and terrestrial invertebrates (insects, spiders and decapods) and is secondarily carnivore (Soares *et al*., 2008); hence, it was expected a high rate of parasitism by nematode. However, low parasitism by *C. acaudatus* in *O. bicirrhosum* of this was due to the lack of infectants forms in the environment, because they are trophically parasites transmitted.

Nematodes Camallanidae have a high degree of diversity and specialization in the tropics. *Camallanus anabantis* Pearse, 1933 infects *Anabas testudineus* Bloch, 1792 throughout the year and has a distinct annual cycle, with an annual generation. Hosts' invasion occurs during spring and summer, and during fall and winter, the parasites grow and develop (De, 1993). In this study, carried out from April to October, infections by *C. acaudatus* in *O. bicirrhosum* were only by adult parasites and no nematode was found from April to June. Therefore, future studies on the seasonal pattern of *C. acaudatus* are necessary for knowledge from life cycle of this nematode that is still totally unknown. In South America, fish there have been few investigations on the subject.

The Kn considered an important quantitative indicator of the degree of well-being (Guidelli *et al.*, 2011; Silva *et al*., 2011; Lemos *et al*., 2012) was not affected by parasitism in *O. bicirrhosum*. However, was found increase in Kn with the intensity of monogenoideans *Gonocleithrum* spp. due to its aggregate distribution pattern. Probably, fish with a better Kn are more able to withstand the intensity of infections by these monogeneans species and therefore there are no pathological effects as expected. Similarly, in *O. niger* the intensity of monogenoideans *Cosmetocleithrum* spp. showed a positive correlation with the Kn, which indicates that the parasitism did not affect the health of the host (Silva *et al*., 2011) and the same occurred for *Gyrodactylus gemini* Ferraz, Shinn & Sommerville, 1994 and *Procamallanus* (*Spirocamallanus*) *inopinatus* Travassos, Artigas & Pereira, 1928 in *Semaprochilodus insignis* Jardine, 1841 from Coari Lake (Silva *et al*., 2011). In contrast, negative correlation between Kn and the abundance of *Anacanthorus penilabiatus* Boeger, Husak & Martins, 1995 and *Mymarothecium* spp. was reported for *Piaractus mesopotamicus* Holmberg, 1887 (Lizama *et al*., 2007), as well as between the abundance of monogenoideans and the Kn of *Leporinus lacustris* Amaral-Campos, 1945 and *Leporinus elongatus* Valenciennes, 1850 (Guidelli *et al*., 2011), indicating effects negative of these parasite on hosts. Since the response of hosts to parasites varies with the stress intensity provoked by the parasites species, life cycle their and time of exposure to they; thus, the host-parasite interactions are complex.

The majority of parasite species of *O. bicirrhosum* were represented by ectoparasites, which are favoured by the lentic habitat and has not parasitic specificity (*I. multifiliis* and *P. pillulare*). Parasite abundance of *I. multifiliis* and *Gonocleithrum* spp. increased with the increase of host length, indicating that this was a factor influencing such infections. However, as no correlations was observed between the host length and the diversity indices, then this indicates that factors other than host body size are more important determinants of diversity and species richness among the host population. Diet composition may be important for *C. acaudatus* infection, since it is a heteroxenous nematode with hosts related to food web links.

ACKNOWLEDGEMENTS

The present work was developed according to the principles adopted by Brazilian College of

Animal Experiments (COBEA) and under the license of ICMBio. M. Tavares-Dias was supported by a Research fellowship from Conselho Nacional de Pesquisa e Desenvolvimento Tecnológico (CNPq).

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Received September 29, 2014.
Accepted November 26, 2014.