

Helminth Parasites of Lane Snapper, *Lutjanus synagris* from Santiagoullo Reef, Veracruz, Mexico

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Abstract

In 51 specimens of lane snapper, *Lutjanus synagris*, captured in Santiagoullo Reef, Veracruz Reef National Park System, State of Veracruz, in the Southern Gulf of Mexico, a total of 25 helminth species were recovered, as follows: 9 digeneans (8 adults, and 1 metacercaria), 7 monogeneans, 6 nematodes (4 adults, and 2 larvae), 2 cestodes (both larvae), and 1 acanthocephalan (juvenile). Out of the 25 species, 11 are new host records; 2 have prevalence > 50%, and mean intensity > 4.7; *Haliotrematoides cornigerum* (monogenean) had the highest prevalence, 94.11%, followed by *Euryhaliotrema tubocirrus* with prevalence of 66.67%. Richness ($S = 25$) and diversity (Shannon index $H' = 2.13$) at component community, and endoparasites infracommunity level ($S = 6.27 \pm 2.5$, Brillouin index $H = 1.07 \pm 0.42$), and ectoparasites infracommunity level ($S = 3.6875 \pm 1.87$, Brillouin index $H = 0.74 \pm 0.4$), were similar to those found in other marine fish. Results suggests that the host feeding habits determine the endoparasites composition, while the ectoparasites composition is associated to the environmental conditions.

Keywords: *Lutjanus synagris*, richness, diversity, parasites

1. Introduction

Demersal species known as lane snapper is distributed from North Carolina in the US, to Brazil, including the Gulf of Mexico and the Caribbean Sea (Allen, 1985). It has high biological and commercial relevance (Freitas et al., 2011), with growing demand for its value for regional fisheries in Mexico (Jiménez-Badillo et al., 2006; Arreguín-Sánchez & Arcos-Huitrón, 2011), and the Caribbean (Landínez et al., 2009). Considering only the Southeast US and the Colombian Caribbean, 48 parasite species have been recorded for *Lutjanus synagris*, a figure similar to that found in *L. griseus* for the same region (Argaéz-García et al., 2010). Helminth species registered for *L. synagris*: trematodes, *Hamacreadium mutabile*, *Helicometrina nimia*, *Lecithochirium floridense*, *L. microstomum*, *L. parvum*, *Metadena globose*, and *Stephanostomum casum* in the southeast USA (Overstreet et al., 2009); *Siphodera vinaledwardsii*, *M. globosa*, *Paracryptogonimus neoamericanus*, *S. casum*, *Lepocreadium trulla*, *H. mutabile*, *H. gullela*, *Prosogonotrema bilabiatum*, *Aponurus laguncula*, and other as Didymozoidae, *Pseudopecoelus* sp., *Xystretum* sp., *Lasiotocus* sp., *Megalomyzon* sp., for the Colombian Caribbean (Velez, 1987; Cortés et al., 2009a); the monogeneans: *Haliotrematoides longihamus*, *H. magnigastrohamus*, *H. heteracantha*, *H. cornigerum*, *Euryhaliotrema longibaculum*, *E. tubocirrus*, *E. torquecirrus* in hosts from Cuba (Kritsky & Boeger, 2002; Kritsky, Yang, & Sun, 2009), and the nematodes: *Capillaria* sp., *Contracecum* sp., *Cucullanus* sp., *Raphidascaris* sp., in hosts from Colombia Caribbean (Cortés et al., 2009b). Parasite species found in lane snapper are unknown in México, and only *Mesostephanus appendiculatoides* has been recorded (Pérez-Ponce de León et al., 2007). This is why, we are addressing lane snapper parasites in this paper, as the composition, richness, and diversity characteristics of parasite communities are unknown, contrasting with other tropical marine fish where some baseline data has been published (Sánchez-Ramírez & Vidal-Martínez, 2002; Aguirre-Macedo et al., 2007; Montoya-Mendoza et al., 2014b).

Considering the previous parasite records for Lutjanids, it was expected that lane snappers would have a helminth community as rich and diverse as that of other reef-associated fish (Rohde & Heap, 1998; Justine et al., 2012). Herein we are describing the helminth community of *L. synagris* in terms of species richness and diversity.

2. Material and Methods

2.1 Sampling Procedures

From October, 2012 to March, 2013, 51 specimens of *L. synagris* were collected for helminthological examination. Fish were captured with fishing-baited hooks and longlines at 10-20 m depth in Santiaguillo Reef (19°08'30.00" N y 95°48'00.41" W), located in the Veracruz Reef National Park System, state of Veracruz, Mexico, in the Southern Gulf of Mexico. Fish specimens were kept in plastic containers with ice and transported to the lab for examination within 24 hours post-capture. Tissues and organs were reviewed using a stereomicroscope. The external examination included skin, scales, fins, gills, eyes, nostrils, mouth, and anus. Gills were removed and analyzed separately in Petri dishes with seawater. Internal examination included mesenteries, liver, kidney, and gonads, and the whole digestive system was placed in Petri dishes with 0.75% saline for examination. Helminths were fixed with hot 4% formalin and preserved in 70% ethyl alcohol.

For taxa identification, monogeneans, digeneans, cestodes, and acanthocephalans were stained using either Mayer's paracarmine, Gomori's triple stain, or Erlich's hematoxylin and then dehydrated in a graded alcohol series, cleared with clove oil, and mounted whole in Canada balsam. Nematodes were studied on temporary slides and cleared in glycerin, after which they were preserved in 70% alcohol. In order to study sclerotized structures, some specimens of monogeneans were fixed with ammonium picrate (Vidal-Martínez et al., 2001). Voucher specimens were deposited at the National Helminths Collection (*Colección Nacional de Helmintos*) (CNHE), Institute of Biology of the National Autonomous University, Mexico City. Prevalence (percentage of infected hosts) and mean intensity (mean number of parasites per infected fish) were calculated following Bush et al. (1997).

2.2 Sample Size

Helminth communities were analyzed at the component community (all helminths in all individuals of *L. synagris* examined), and infracommunity (helminths in each single fish examined) levels (Holmes & Price, 1986). Helminth species richness observed was one measure of component community structure adopted. Sampling adequacy for the component community was evaluated with a similar procedure at the helminth parasites community as *L. campechanus* (Montaya-Mendoza et al., 2014b), and it consisted in using a randomized (100x) sample-based species accumulation curve computed in EstimateS (version 8.0 RK Colwell, <http://viceroy.eeb.unconn.edu/estimates>) (Moreno & Halffter, 2001). For the component community, we examined the asymptotic richness based on the Clench's model equation (Soberon & Llorente, 1993), as well as the final slope of the randomized species accumulation curve (Jiménez-Valverde & Hortal, 2003). Clench's model is described by the following function:

$$V2 = (a \times VI) / [1 + (b \times VI)] \quad (1)$$

Where, $V2$ is the observed richness, VI is the number of hosts examined, and a and b are curve parameters, a equals the new species adding rate, and b is a parameter related to the curve shape. These values were calculated iteratively using the EstimateS and Statistica (StatSoft, Inc., Tulsa, Oklahoma) software as in Jiménez-Valverde and Hortal (2003). The slope of the cumulative species curve was calculated as $a / (1 + b \times n)^2$, where a and b are parameters above and n is the number of hosts examined from a given component. Clench's model equation allows estimating the total number of species in a component as a/b . To calculate the number of rare species missing at the component community level, the nonparametric species-richness estimator bootstrap was calculated from data observed, as recommended by Poulin (1998). The Shannon index of diversity (H'), was calculated for the component community as in Magurran (2004). Descriptors of infracommunities included the mean number of helminth species per fish, the mean number of helminth individuals per fish, and the mean value of the Brillouin's diversity index per fish (H).

3. Results

All 51 *L. synagris* were sexually mature adults (23 males and 28 females), total fish length 22.2-49.4 cm (33.8 ± 4.3); fish weight, 157-1074 g (552.6 ± 177.1), and all were parasitized. Twenty-five helminth species were recorded. The analysis of cumulative species curves for component community suggested that the inventory of the helminth species was near completed and the slope of cumulative species curve was 0.03. Thus, an asymptote was reached, and richness estimated by Clench's model was 26.18 species ($a = 6.440828$, $b =$

0.245964; $a/b = 26.18$); the value of the nonparametric species-richness estimator bootstrap ($S_b = 26.79$) confirms, indeed, that most, if not all, helminth species from the component community were recovered. Out of the 25 species of helminths, 9 were digeneans (8 adults, and 1 metacercaria), 7 were monogeneans, 6 were nematodes (4 adults, and 2 larvae), 2 were cestodes (both larvae), and one was acanthocephala (1 juvenile) (Table 1).

Two of the 25 helminth species appeared as frequent with prevalence values $> 50\%$ and mean intensity > 4.7 helminths per infected fish (Table I). Nine out of the 25 were rare with a prevalence $< 6\%$ and mean intensity < 1.9 , and fourteen species hold an intermediate position. Monogeneans *H. cornigerum* (prevalence 94.11%), *E. tubocirrus* (66.67%), *H. heteracantha* (43.14%), and *E. longibaculum* (43.14%) reached the highest prevalences recorded in this study. Subtle differences were observed in helminth infections when comparing male and female fish; only in the number of parasites in males ($n = 1121$) it was higher than in females ($n = 819$). No significant correlation was found between the total number of species (S) or the total number of helminths (N), when compared to the host size (total host length vs. S , $r = 0.039$; vs. N , $r = 0.099$) and weight (weight vs. S , $r = 0.037$; vs. N , $r = 0.055$). However, a highly significant correlation ($r = 0.77$) was found when comparing prevalence and mean intensity of the helminth species.

Table 1. Prevalence, mean intensity, and site of infection of helminth parasites in lane snapper, *Lutjanus synagris*, from Santiaguillo Reef, Veracruz, Mexico

Species	CNHE	Site	n (% prevalence)	mi (\pm SD)	Range
Trematoda					
<i>Lecithochirium floridense</i> (Manter, 1934)	10209	sto	2 (3.92)	28 \pm 38.18	1-55
<i>Prepetos trulla</i> (Linton, 1907)	10210	int	12 (23.53)	3.41 \pm 2.87	1-10
<i>Siphodera vinalwardsii</i> (Linton, 1901)	10211	int	20 (39.22)	5.75 \pm 8.13	1-35
<i>Metadena adglobosa</i> Manter, 1947*	10212	int	7 (13.73)	6 \pm 8.96	1-26
<i>Metadena crassulata</i> Linton, 1910*	10213	int	15 (29.41)	2.93 \pm 2.84	1-11
<i>Lepocreadium</i> sp.*	10214	int	2 (3.92)	1 \pm 0	1-1
<i>Opechona</i> sp.*	10215	int	2 (3.92)	1 \pm 0	1-1
<i>Stephanostomum</i> sp.	10216	int	1 (1.96)	5	-
Trematoda ^(mt)	-	fins	9 (17.65)	2.56 \pm 2.12	1-7
Monogenea					
<i>Haliotrematoides cornigerum</i> (Zhukov, 1976)**	10217	gills	48 (94.11)	18.48 \pm 22.6	1-135
<i>Haliotrematoides heteracantha</i> (Zhukov, 1976)	10218	gills	22 (43.14)	2.68 \pm 1.89	1-8
<i>Haliotrematoides longihamus</i> (Zhukov, 1976)**	10219	gills	14 (27.45)	2.21 \pm 1.71	1-7
<i>Haliotrematoides magnigastrohamus</i> (Zhukov, 1976)**	10220	gills	16 (31.37)	3.43 \pm 3.71	1-15
<i>Euryhaliotrema longibaculum</i> (Zhukov, 1976)**	10221	gills	22 (43.14)	3.27 \pm 2.21	1-8
<i>Euryhaliotrema tubocirrus</i> (Zhukov, 1976)	10222	gills	34 (66.67)	4.74 \pm 4.11	1-19
<i>Euryhaliotrema torquescirrus</i> (Zhukov, 1976)	10223	gills	21 (41.18)	2.67 \pm 1.91	1-8
Cestoda					
<i>Callitetrarhynchus</i> sp. ^{(p)*}	10224	int	11 (21.57)	3.27 \pm 3.07	1-10
Tetraphyllidea gen. sp. ^(p)	10225	Int	24 (47.06)	7.21 \pm 13.15	1-54
Nematoda					
<i>Cucullanus pargi</i> González-Solís, Tuz-Paredes y Quintal-Loria, 2007*	10227	int	10 (19.61)	2.4 \pm 1.65	1-6
<i>Hysterothylacium reliquens</i> Norris y Overstreet, 1975*	10228	Int	19 (37.25)	2.42 \pm 1.54	1-6
<i>Anisakis</i> sp. ^{(l)*}	10229	mes	1 (1.96)	1	-
<i>Contraecum</i> sp. ^(l)	10230	int	3 (5.88)	1 \pm 0	1-1
<i>Procammallanus</i> (<i>Spirocammallanus</i>) sp.*	10231	int	2 (3.92)	1.5 \pm 0.71	1-2
Phyllometridae gen. sp.*	10232	cau fn	1 (1.96)	1	-
Acanthocephala					
<i>Gorgorhynchoides</i> sp. ^{(l)*}	10226	mes	2 (3.92)	1 \pm 0	1-1

Note. Life stages: *mt*, metacercaria; *p*, plerocercoid; *j*, juvenile; *l*, larva. *, New host record. **, New record for Mexico. Site: sto, stomach; int, intestine; mes, mesenteries, cau fn; caudal fin. n, number of hosts infected. mi, mean intensity.

A total of 1940 individual helminths were collected; infections ranged from 5 to 155 helminth individuals per infected host. Richness of the component community was $S = 25$ and the Shannon index diversity value was $H' = 2.13$. Richness in infracommunities ranged from 2 to 12 species of helminths per fish, three hosts were infected by 2, 4 had 3, 8 had 4, 6 had 5, 7 had 6, 4 had 7, 9 had 8, 6 had 9, 1 had 10, 2 had 11 and 1 host had the maximum of 12 parasites species. As it can be observed, about one half of examined hosts ($n = 30$) were infected with 6 to 12 helminth species. The average number of parasites species per individual host was 6.27 ± 2.5 , while the average number of helminth individuals per host was 38.03 ± 29.47 . The value of Brillouin's index for each infracommunity ranged from 0.14 to 1.79 with an average value of 1.07 ± 0.42 . Fourteen helminth species inhabit the intestine (Table 1); intestines of only 3 hosts were free of parasites, i. e., 48/51 (94%) hosts had intestinal helminths. Twelve hosts (25%) had a single helminth species and 36 (75%) had concurrent infections of intestinal helminths. Up to a maximum of 7 intestinal helminth species co-occurred in the infracommunities. A total of 592 intestinal individual helminths were collected; the number of helminth individuals per host ranged from 1 to 68 with an average of 12.3 ± 15.54 . The number of intestinal helminth species ranged from 1 to 7 per examined host and the average was 2.7 ± 1.5 . Brillouin's index for intestinal species was from 0 to 1.13 with an average of 0.49 ± 0.36 . Seven helminth species inhabit the gills (Table 1); the branchial of only 3 hosts were free of parasites, i.e., 48/51 (94%) hosts had branchial helminths. Five hosts (10.4%) had a single helminth species and 43 (89.4%) had concurrent infections of branchial helminths. Up to a maximum of 7 branchial helminth species co-occurred in the ectoparasite infracommunities. A total of 1321 branchial individual helminths were collected; the number of helminths per host ranged from 1 to 146 with average of 27.5 ± 27.4 . The number of ectoparasite helminth species ranged from 1 to 7 per examined host and the average was 3.6 ± 1.8 . Brillouin's index for branchial species was from 0 to 1.5 with average of 0.66 ± 0.44 .

4. Discussion

Adding the previous known parasites records for *L. synagris* and those reported in the present investigation, the updated helminths inventory for this fish reached 59 helminth species. This research is adding 11 new host records and 4 new location records. The number of parasite species on *L. synagris* is higher if compared with the number of species found in other Lutjanids; for example, 44 species have been reported in *Lutjanus griseus* from the Gulf of Mexico and Caribbean region (Argaéz-García et al., 2010). This larger inventory of parasites covers a systematic sampling involving several locations and a much larger sample size. Nevertheless, an accumulation curve suggested that our parasites inventory is near completed concerning the site studied with 42.3% (25/59) in this helminths survey. Trematodes and nematodes species are the main components of intestinal helminth parasites of marine fish most studied in northern temperate zones (Zander et al., 1999; Fernández et al., 2005). The same taxonomic groups of helminths have also been recorded as the most frequent in marine fish in the southern Gulf of Mexico (Moravec et al., 1997; Sánchez-Ramírez & Vidal-Martínez, 2002; Rodríguez-González & Vidal-Martínez, 2008; Argáez-García et al., 2010; Espínola-Novelo et al., 2013; Montoya-Mendoza et al., 2014a, 2014b). The number of trematodes and nematodes species in *L. synagris* was larger than that of cestodes and acanthocephalans. Proportions among parasite groups indicate the relevance of intermediate hosts (Palm & Overstreet, 2000; 2002; Sánchez-Ramírez & Vidal-Martínez, 2002). Lane snapper are carnivorous, and feed on crustaceans such as stomatopods, penneids, and portunids, and other as gastropods (Doncel & Paramo, 2010; Rosa et al., 2015); consumption of these organisms facilitates parasitic infections using them as intermediate hosts (Deardorff & Overstreet, 1981; Aguirre-Macedo et al., 2007; Lagrue et al., 2011). Presence of living cestode, nematode, and acanthocephalan larvae suggest that lane snappers are at intermediate level in the marine food web, just as are other fish in the Western Atlantic (Luque & Poulin, 2004).

Host specificity seems to be an important ectoparasites trait in structuring the communities of helminths of *L. synagris*. Four out of the seven helminth parasite species reached maximum prevalence in this survey. The highest prevalence and mean intensity was recorded by *H. cornigerum*, *E. tubocirrus*, *H. heteracantha* and *E. longibaculum*, with previous records in different snapper such as *L. analis*, *L. apodus*, *L. cyanopterus*, *L. mohogoni*, *L. synagris*, *Ocyurus chrysurus*, and *Rhomboplites aurorubens* (Zhukov, 1976; Kritsky & Boeger, 2002; Kritsky, 2012). This number of monogeneans in *L. synagris*, is higher if compared to others Lutjanids (Zhukov, 1976), and similar to *Caranx hippos* from Veracruz (Montoya-Mendoza et al., 2008), Venezuela (Boada et al., 2012), and Brazil (Luque & Alves, 2001). Richness and diversity of ectoparasites infracommunity in *L. synagris*, can be explained particularly from host-parasite relationship and the environmental conditions prevailing in coral reef systems (Justine et al., 2012), and considering that reefs in southeastern Gulf of Mexico are part of the Barrera Reef, running from the Caribbean Sea to the coast of Veracruz (Jordán-Dahlgren & Rodríguez-Martínez, 2003), providing larger distribution areas for Lutjanids and their ectoparasites (Zhukov, 1976; Montoya-Mendoza et al., 2014a).

These data shows that the helminth community of *L. synagris* is as rich and diverse as those of other marine fish in temperate (Châari et al., 2015) and tropical zones (Luque & Poulin, 2007; Madhavi & Triveni Lakshmi, 2012). This community is also similar to others described from marine fish in the southern Gulf of Mexico and the Caribbean (Sánchez-Ramírez & Vidal-Martínez, 2002; Aguirre-Macedo et al., 2007; Espínola-Novelo et al., 2013; Montoya-Mendoza et al., 2014a, 2014b). Infracommunity richness and diversity were not different from those recorded for marine fish at temperate latitudes (Madhavi & Sai Ram, 2000; Châari et al., 2015), the tropical Atlantic (Sánchez-Ramírez & Vidal-Martínez, 2002; Espínola-Novelo et al., 2013; Montoya-Mendoza et al., 2014b), and the Australian tropic (Rohde & Heap, 1998; Justine et al., 2012). The composition, richness, and diversity of the helminth communities of *L. synagris* are mainly associated to their feeding habits as reef inhabitants. Now, the intermediate host distribution is a factor to be considered, as fish feed on the most abundant prey, together with environmental stability in tropical latitudes. Therefore, the fish feeding habits, and the intermediate host abundance, and the habitat continuum are important factors determining the parasite community structure. A further potential factor to explain the composition of these communities is host switching among sympatric, related species in this area, as 60% or more of intestinal parasite species found in this study have been registered in other Lutjanids (Kritsky & Boeger, 2002; Overstreet et al., 2009; Argáez-García et al., 2010; Kritsky, 2012; Montoya-Mendoza et al., 2014a, 2014b).

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