

“Community Structure Of The Gastrointestinal Helminths Of The Red Snapper *Lutjanus Campechanus*, Poey, 1860 (Perciformes: Lutjanidae), On The Tamiahua Coast, Veracruz”

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Abstract

The present study captured 71 hosts of *Lutjanus campechanus* from December 2015 to November 2016, observing a sex ratio of 1:0.5, an average total length (TL) of 31.53 cm, an average height (H) of 10.44 cm, and an average weight (W) of 455.25 g. The average age of the *L. campechanus* individuals captured was 2.76 years. The present study registered 430 parasites, 118 nematodes, 272 acanthocephalans, and 27 platyhelminths along the length of the gastrointestinal tract. The intestine presented the highest number of acanthocephalans, while the nematodes and platyhelminths were the most abundant in the stomach. The months in which the highest presence of helminths was recorded were October, December, and January, with June, July, and August the months presenting the lowest abundance. The prevalence of each group was 85%, 67%, and 38% for acanthocephalans, nematodes, and platyhelminths respectively, while the median intensity of infection and average abundance were higher for the acanthocephalans, with 3.83 and 4.45, respectively. In terms of the association of variables, the nematodes and platyhelminths did not show a correlation with size (TL cm, A cm), weight, or sex. Only the acanthocephalans presented a correlation with size (TL cm, A cm) and weight (W g).

Keywords: endoparasites, community, red fish, coastal region

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I. Introduction

A multitude of parasites are present in fish of habitual consumption (Vidal *et al.*, 2002; Fuentes-Zambrano *et al.*, 2003; CECOPECA, 2012; Morales-Serna and Pérez-Ponce de León, 2012). Chappuis and Loutan (2006) describe how the consumption of fish presents a high risk for the transmission of parasites to humans. Recent years have seen the discovery of emerging diseases related to the consumption of fish (Keiser and Utzinger, 2005; Torres *et al.*, 2007).

Helminths have become the subject of increasing research interest in recent decades, given that they are responsible for both significant economic losses in the aquaculture sector and the majority of parasitic zoonoses (diseases caused by consumption of parasitized fish) found in many regions of the world (Lamothe-Argumedo and Osorio-Saraia, 1998; Ogata *et al.*, 1998; Quijada *et al.*, 2005; Aragort-Fernández, 2006; Luque, 2008). However, these organisms play a valuable role, regulating the abundance of wild animals and providing stability to the trophic networks within ecosystems (Morales-Serna and Pérez-Ponce de León, 2012).

Although many parasite species infect fish, only a few helminth species are zoonotic, namely that they cause health problems in humans.

By volume, the red snapper occupies 23rd place in Mexico's fishery production, with approximately 8,532 tons, while, by value, it lies in eleventh place. The average annual growth rate for production was 1.43% over the last ten years. In 2013, 322 tons of this species was captured in the state of Veracruz (CONAPESCA, 2013). The species is distributed across the western Atlantic, the Gulf of Mexico, and the east coast of the United States, where its range runs north up to Massachusetts, including the Florida coast, although it is rarely seen in North Carolina. Although it can be found at depths of 10 to 190 m, it is most commonly found at depths of 30 to 130 m (Jiménez-Badillo *et al.*, 2006).

Fuentes-Zambrano *et al.* (2003), in research conducted in the *Laguna de la Restinga* National Park, Margarita Island, Venezuela, found total of eight parasite species present in *Lutjanus griseus*. The protozoans *Amyloodinium ocellatum*, *Trichodina sp.*, and *Cryptobia sp.*, the crustaceans *Lernanthropus rathbuni* and *Caligus bonito*, and the platyhelminth *Euryhaliotrema sp.* were all found in the gills, while *Argulus sp.* was found in the

skin and *Siphodera vinaledwardsii* in the pyloric caecum and intestines. *A. ocellatum* was considered the species with most potential for developing acute pathological states in *L. griseus* individuals held in captivity.

Argáez-García *et al.* (2010) conducted helminthological analysis on the digestive tracts of *Lutjanus griseus* individuals collected from the Celestùn coastal lagoon and the marine area offshore of the communities of Chelem and Progreso in the state of Yucatán, Mexico.

The same researchers also compiled a list of the intestinal helminths registered for this host species in the Gulf of Mexico and the Caribbean region, finding 20 helminth species reported for Yucatán. Of these species, twelve were newly registered for *L. griseus* and the other eight species pertain to previous records for the Gulf of Mexico and the Caribbean Sea, thus increasing to 44 the number of species for this host in the region. These authors suggest a similarity between the intestinal helminth fauna of *L. griseus* in Yucatán and that observed for the Atlantic coast of the United States, the Gulf of Mexico, and the Caribbean Sea.

Moravec *et al.* (2014), in a study carried out in the northern section of the Gulf of Mexico that pertains to the state of Florida, USA, found and described the three following parasitic nematode species pertaining to the family Philometridae and infecting the gonads of the following fish of the family Lutjanidae: *Philometra longispicula*, found in *L. Campechanus* and *L. vivanus*; *P. latispicula* found in *L. griseus*; and *P. synagris* found in *Lutjanus synagris*. These parasite species are mainly characterized by the size of their spicules (378–690 µm, 135–144 µm, and 186–219 µm, respectively).

Montoya-Mendoza *et al.* (2014) captured 52 examples of *L. campechanus* in the Santiaguillo Reef, which pertains to the Veracruz Coral Reef System National Park, finding a total of 21 parasitic helminth species, including: nine trematodes (seven adults and two metacercariae); four nematodes (three adults and one in larval state); four acanthocephalans (one adult and three juveniles); two cestodes (both in larval state); and two monogeneans. Sixteen of these 21 species corresponded to new reports and seven are species already reported with a 40% prevalence level and an average intensity of 4.1. The monogenean *Euryhaliotrema tubocirrus* was the species that presented the highest prevalence of 78.8%. It is suggested that the community composition of parasites is associated with the host's feeding habits, with 18 of these 21 parasite species transferred trophically.

II. Material And Methods

The samples were collected via commercial capture by the independent and associated fisherfolk of the community of Tamiahua, Veracruz, with one sampling effort conducted per month, from December 2015 to October 2016, with a sample size of 71 *L. campechanus* specimens.

The organisms captured were transported to the Biology Laboratory of the Faculty of Biological and Agro-livestock Sciences at the *Universidad Veracruzana* for correct taxonomic identification.

The morphometric values (TL, H, and W) of the hosts were then registered. In order to evaluate the sex ratio and sexual maturity, the gonads were extracted and the sex determined (Nicolisky, 1963, Compagno, 1984, Barrera-Lara, 2013), with the sagittal otoliths then extracted, polished, and rinsed and the bony structures then reviewed (Tello-Macas, 2014).

The examination involved the extraction of the gastrointestinal tract, after which the parasites were collected and quantified and their location in the host (stomach or intestine) recorded (Lamothe, 1997; Vidal *et al.*, 2002).

The fixing and preservation of each parasite species was carried out in accordance with the established conventional techniques (Lamothe, 1997; Vida *et al.*, 2002). The specific fixation process conducted depended on the group of parasites sampled. The platyhelminths were flattened and fixed in Bouin's solution for 24 hours, removed, washed in alcohol at 70%, stored in small vials, stained with Gomori trichrome in alcoholic solution, rinsed with methyl salicylate, and mounted using Entellan Merk or Canada balsam. The nematodes were fixed in hot alcohol at 70%, stored in vials with clean cold alcohol at 70%, and then rinsed with glycerin. The acanthocephalans were placed in distilled water at 4°C for four to eight hours or more, prior to fixation, in order to expose the proboscis. Small acanthocephalan specimens were fixed, flattened between two slides with Bouin's solution for 24 hours, removed, washed with alcohol at 70%, and stored in jars. The samples were then stained with Gomori trichrome, for periods of 30 minutes to an hour or more depending on the thickness and size of the specimen, until attaining a pale pink color, and then rinsed with methyl salicylate and mounted in Canada balsam.

The parasites were then identified in line with Yamaguti (1961), Moravec (1998), Anderson (2000), Gibson *et al.* (2002), and Vidal *et al.* (2002).

The prevalence (%) was recorded and the formula $P=c/d(100)$ then applied on the data obtained, where c = number of parasitized fish per parasite species and d = number of fish of the same species reviewed in a single sample.

Intensity of median infection: Average intensity of a particular parasite species in the infected hosts. Total number of parasites = n , divided among the number of infected fish in the sample = d , defined via the formula $I=n/d$.

Average abundance: Average number of parasites per host examined in a sample (including non-infected individuals). $A=n/h$, where n =Total number of parasites and h = total number of hosts examined.

Association among variables: In order to determine the possible correlations among the gastrointestinal parasites to the size, weight, sex, and age of the host, a normality test was carried out to establish which statistical test fit the data, resulting in the use of a Pearson correlation coefficient and a T test. The analysis was conducted in the R statistics program.

III. Results

Seventy-one examples of *L. campechanus* were analyzed, corresponding to 64.78% females and 35.21% males at a ratio of 1:0.5. Maximum and minimum total length (TL) values of 50.7 and 21.6 cm, respectively, were recorded, giving an average of 31.53 cm, while the maximum and minimum height (H) observed were 18 and 7.3 cm, respectively, giving an average of 10.44 cm. The average weight (W) of the red snappers sampled was 455.25 g, with maximum and minimum values ranging from 1,846 to 155 g. The age of the *L. campechanus* individuals was from one to seven years, with an average value of 2.76 years.

It was found that 4.20% were parasite free, while 95.80% of the red snapper individuals were infected with helminths, with three parasite phyla identified in the *L. campechanus* gastrointestinal tracts examined, which corresponded to a total of 424 parasites, comprising 118 nematodes, 272 acanthocephalans, and 34 platyhelminths. The site in the host (stomach or intestine) in which the helminth species were located was then established.

The taxonomic identification revealed five families, six genera, and seven helminth species (Table 1).

Table 1. Record of the helminth species found in *Lutjanus campechanus* (Sto = stomach and Int = intestine).

Phylum	Class	Family	Genus	Species	Microhabitat
Platyhelminthes	Digenea	Lecithasteridae	<i>Aponurus</i>	<i>Aponurus laguncula</i> (Looss, 1907)	Sto
			<i>Lecithochirium</i>	<i>Lecithochirium floridense</i> (Manter, 1934) Crowcroft, 1946	Sto
		Lepocreadiidae	<i>Preptetos</i>	<i>Preptetos trulla</i> (Linton, 1907) Bray and Cribb, 1996	Int
Acanthocephala	Palaeacanthocephala	Illiosentidae	<i>Dollfusentis</i>	<i>Dollfusentis chandleri</i> (Golvan, 1969)	Sto/Int
		Isthmosacanthidae	<i>Gorgorhynchoides</i>	<i>Gorgorhynchoides bullocki</i> (Cable and Mafarachisi, 1970)	Sto/Int
				<i>Gorgorhynchoides sp.</i>	Sto/Int
Nematode	Chromadorea	Cucullanidae	<i>Cucullanus</i>	<i>Cucullanus pargi</i> (González-Solis, Tuz-Paredéz and Quintal-Loria, 2007)	Sto/Int

In terms of monthly variation, the helminth species *Dollfusentis chandleri*, *Gorgorhynchus bullocki*, and *Cucullanus pargi* were more abundant in March–June (dry season), July–October (rainy season), and November–February (northerly winds) than the species *Aponurus laguncula*, *Lecithochirium floridense*, and *Preptetos trulla*.

The infection parameters (prevalence %, median intensity of infection, and average abundance) were subject to a general evaluation for each helminth species (Table 2), namely for each group of parasites, with analysis then conducted for each site of infection (stomach and/or intestine). The highest prevalence levels were found for *Dollfusentis chandleri* (74.64%), *Cucullanus pargi* (70.42%), and *Gorgorhynchus bullocki* (59.15%).

Table 2. Infection parameters by helminth species

Especies	Microhábitat	% prevalencia	Abundancia promedio	Intensidad de infección
<i>Aponurus laguncula</i> (Looss, 1907)	Est	19.71	0.202	0.197
<i>Lecithochirium floridense</i> (Manter, 1934)	Est	14.08	0.002	0.14
<i>Preptetos trulla</i> (Linton, 1907)	Int	8.45	0.086	0.084
<i>Dollfusentis chandleri</i> (Golvan, 1969)	Est/Int	74.64	0.768	0.746
<i>Gorgorhynchus bullocki</i> (Cable y Mafarachisi, 1970)	Est/Int	59.15	0.608	0.591
<i>Gorgorhynchoides</i>	Est/Int	32.39	0.333	0.323
<i>Cucullanus pargi</i> (González-Solis, Tuz-Paredéz y Quintal-Loria, 2007)	Est/Int	70.42	0.724	0.704

Especies Microhabitat %prevalencia Abundancia promedio Intensidad de infección Species
Microhabitat %prevalence Average abundance Intensity of infection

The values obtained via the Pearson correlation reveal a slight correlation between the acanthocephalan *Dollfusentis chandleri* and the height and weight of the red snapper (Table 3).

Table 3. Pearson correlation showing the correlation values between helminths and the variables TL (total length), H (height), P (weight), and age

Ratio		Statistic (t)	Degree of freedom (df)	p-value	Coefficient of correlation
<i>Aponurus laguncula</i>	TL (cm)	-1.0098	69	0.3161	-0.1206773
	H (cm)	-0.70841	69	0.4811	-0.08497444
	W (g)	-0.65117	69	0.5171	-0.07815156
	Age	-0.76451	69	0.4472	-0.09164842
<i>Lecithochirium floridense</i>	TL (cm)	1.2432	69	0.218	0.1480131
	H (cm)	1.4813	69	0.1431	0.1755579
	W (g)	1.9252,	69	0.05833	0.2257782
	Age (cm)	2.2073	69	0.03062	0.2568117
<i>Prepetos trulla</i>	TL (cm)	1.9839,	69	0.05124	0.2323045
	H (cm)	1.7335	69	0.08747	0.2042882
	W (g)	1.7957	69	0.07692	0.2112949
	Age (cm)	2.2073	69	0.03062	0.2568117
<i>Dollfusentis chandleri</i>	TL (cm)	4.143	69	9.57E-05	0.4463245
	H (cm)	4.8957	69	6.19E-06	0.5077487
	W (g)	4.8985	69	6.13E-06	0.5079653
	Age (cm)	3.4351	69	0.001006	0.3821546
<i>Gorgorhynchoides bullocki</i>	TL (cm)	2.8844	69	0.005227	0.3280293
	H (cm)	3.3212	69	0.001435	0.3712527
	W (g)	3.296	69	0.001551	0.3688226
	Age (cm)	1.9398	69	0.05649	0.2274071
<i>Gorgorhynchoides sp.</i>	TL(cm)	1.1104	69	0.2707	0.1324973
	H (cm)	1.262	69	0.2112	0.150205
	W (g)	1.336	69	0.1859	0.1587979
	Age (cm)	1.4729	69	0.1453	0.1745872
<i>Cucullanus pargi</i>	TL(cm)	1.5176,	69	0.1337	0.1797196
	H (cm)	1.9446	69	0.0559	0.227942
	W (g)	2.4521	69	0.01674	0.283116
	Age (cm)	2.4954	69	0.01498	0.287712

A T test was carried out for the ratios between the helminths and the sex of the red snapper individuals, with no statistically-significant differences found between the variables (Table 4).

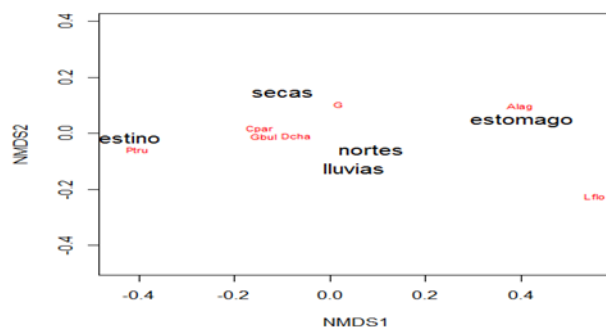
Table. 4. A) Acanthocephalan-host sex ratio, B) Nematode-host sex ratio, and C) Platyhelminth-host sex ratio

Ratio		Statistic (t)	Degree of freedom (df)	p-value
<i>Aponurus laguncula</i>	Sex	0.9279	64.48	0.3569
<i>Lecithochirium floridense</i>	Sex	-1.3718	64.48	0.1771
<i>Prepetos trulla</i>	Sex	-1.2342,	30.156	0.2267
<i>Dollfusentis chandleri</i>	Sex	-0.68457	49.914,	0.4968
<i>Gorgorhynchoides bullocki</i>	Sex	0.16802	43.098	0.8674
<i>Gorgorhynchoides sp.</i>	Sex	0.29452,	66.381	0.7693
<i>Cucullanus pargi</i>	Sex	0.33381	44.081	0.7401

Multidimensional scaling (MDS) analysis was conducted, thus establishing proximities for the helminth species *Aponurus laguncula*, *Lecithochirium floridense*, *Prepetos trulla*, *Dollfusentis chandleri*, *Gorgorhynchus*

bullocki, *Gorgorhynchoides* sp, and *Cucullanus pargi*, by season (dry, rainy, or northerly winds) and site of infection (stomach and/or intestine) (Figure 1).

Fig. 1. MDS for the helminth species, in terms of season and site of infection



IV. Discussion

Helminths are a polyphyletic group, in that they comprise representatives of four phyla, platyhelminths, nematodes, acanthocephalans, and annelids, which are a key component in the biological diversity of our planet. Further to structuring and linking trophic networks (Arias-González and Morand, 2003; Hudson *et al.*, 2006) in ecosystems and providing complementary information on their lifecycles and life histories, these organisms may be used as bioindicators of the environmental health of a certain site (Vidal *et al.*, 2002; Monks *et al.*, 2013). The present study was able to identify three of the four abovementioned phyla (platyhelminths, nematodes, and acanthocephalans), which were found distributed along the length of the gastrointestinal tract (Montoya-Mendoza *et al.*, 2014). The coexistence of the three groups of helminths in the red snapper sampled by the present study can be explained by the spatial position of these fish in the area and their peculiar feeding habitat (Pérez-Ponce de León *et al.*, 2000). These are carnivorous predators, both generalists and opportunists, which feed on fish, shrimp, crab, crustaceans, various benthic invertebrates, cephalopods, and planktonic organisms (Pérez-Díaz *et al.*, 2007). Red snapper carry out their foraging activity over extensive areas, most of the time at night (Starck and Davis 1966, Parrish 1987). The highest abundance of helminths presented in the months of October, December, and January, the same months corresponding to the season featuring low temperatures in the region. Similarly, the abundance of helminths increases depending on the site of infection (stomach and/or intestine), which is the case with the acanthocephalans, given that they are the most abundant group both for the entire sampling season and by site of infection.

The acanthocephalans correlate slightly with the host's height, namely that the larger the size and the older the age of the host, the higher the likely abundance of acanthocephalans, thus coinciding with the work of both Rodríguez-Alvarado (2014) and Sánchez-Ceballos (2010). In contrast with the remaining helminth groups (nematodes and platyhelminths), no correlation was found with any of the variables mentioned above.

In terms of their correspondence with the parameters of infection, the following prevalence results were obtained for each helminth group: acanthocephalans, 85%; nematodes, 67%; and platyhelminths 38%. This suggests that, in accordance with Lamothe (1997) and Vidal *et al.* (2002), the sample size is adequate for acanthocephalans and nematodes, while further sampling is required for the platyhelminths.

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