Factors associated with parasite aggregation levels in fishes from Brazil

Fatores associados aos níveis de agregação parasitária em peixes do Brasil

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Abstract

The degree of parasite aggregation is determined by a number of factors that are not well understood. In total, 3,746 fish from 73 species and their parasites were analyzed to determine associations between the degree of aggregation by taxon and specific characteristics of the fish. A multiple linear regression (95% confidence interval - CI) was used to show that in most taxa, the degree of parasite aggregation was significantly higher in freshwater fish and schooling fish but varied according to taxon in relation to gender, habitat and feeding habits. Parasite aggregations were also significantly increased in fish with a large number of parasitic larvae and greater body length in all evaluated taxa. Because the coefficients of determination of the models were lower than 60% for all taxa, other factors may be involved in parasite aggregation. Conversely, marked significance was observed for the tested variables in relation to taxon dependence, which indicates that additional studies should be performed for these factors using comprehensive databases with larger samples per parasite species.

Keywords: Epidemiology, parasite ecology, multiple regression analysis.

Resumo

O nível de agregação parasitária é determinado por um conjunto de fatores não muito bem elucidados. Foram analisados 3.746 peixes pertencentes a 73 espécies e seus respectivos parasitos, com o objetivo de verificar a associação entre o nível de agregação por táxon e algumas características dos peixes. Por meio da técnica de regressão linear múltipla (IC 95%), observou-se que o nível de agregação de parasitos foi significativamente maior em peixes dulcícolas e formadores de cardume, na maioria dos táxons, mas variou conforme o táxon em relação ao sexo, o hábitat e o hábito alimentar. A agregação parasitária também aumentou significativamente em peixes com maior proporção de larvas parasitas e com maior comprimento do corpo em todos os táxons avaliados. Tendo em vista que os coeficientes de determinação dos modelos para todos os táxons foram inferiores a 60%, outros fatores podem estar envolvidos na agregação de parasitos. Por outro lado, as variáveis testadas assumem importância diferenciada na dependência do táxon, sinalizando para a necessidade de se continuar pesquisando esses fatores em bancos de dados mais abrangentes e com amostras maiores por espécies de parasitos.

Palavras-chave: Epidemiologia, ecologia parasitária, análise de regressão múltipla.

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Introduction

The aggregate spatial distribution pattern of parasites is a common characteristic among various types of hosts and metazoan parasites and is considered a law of parasite ecology (CROFTON, 1971; VON ZUBEN, 1997; POULIN, 2007a). This distribution pattern can be observed in practice, with many hosts harboring few or no parasites and few hosts harboring many parasites (SHAW & DOBSON, 1995; POULIN, 2007b).

Because of the heterogeneous size of parasitic infrapopulations, parasite spacial distribution is significant in the epidemiology of parasitic diseases and is observed in two distribution patterns: homogeneous distribution, which is associated with the mortality and density of parasites and the mortality of hosts induced by the parasites; and aggregate distribution, which is associated with heterogeneous host susceptibility to infection, parasite reproduction in the host and different abilities of hosts to eliminate parasites through immune responses or other mechanisms (ANDERSON & GORDON, 1982).

The inherent characteristics and habits of hosts can also contribute to different degrees of parasite aggregation. An approach to assessing aggregation processes in species of fish parasites with the potential for use as a discriminator of stocks or populations of hosts was developed by Lester (2012), who analyzed aggregations according to parasite species by determining the relationship between parameters of aggregation and aspects of parasitic biology, such as the number of hosts participating in the biological cycle.

Conversely, Poulin (2013) conducted a meta-analysis with data on 410 samples compiled from several published articles to explain possible variations in parasite aggregation. In this study, the author used a mixed-effects model to analyze the different degrees of aggregation between host samples, and the parasite species, host species and study of origin were included as random effect variables. Thus, the effect of systems or researchers was also considered. The parasite taxon, developmental stages (larval/adult) and host size were variables included as fixed effects.

The approach of Poulin (2013) was based on knowledge (from experimental evidence) that changes in the host size in the studied samples and differences between parasitic stages (young and old) and their relationship with parasite aggregation could be confirmed in natural samples. As a result, Poulin (2013) observed that only the host sample size variable, which was included in the model as a confounding variable, was significant and explained 8% of the variability in aggregation. The author also observed that the combination of parasite species, host species and study of origin explained nearly two-thirds of the variability that was not explained by the fixed effects. Although Poulin (2013) considered the idiosyncrasies of the systems or researchers by using the mixed-effects model, the characteristics of studies that utilize databases can also produce different results compared with studies developed with data generated by the research group itself in a single location.

Because the presence of a variable can change the effects of another variable when they are analyzed simultaneously (MEDRONHO et al., 2009), certain explanatory variables that are considered in descriptive studies on fish parasite ecology and have the potential to explain the aggregation of parasites must be tested simultaneously. The present study aimed to evaluate whether parasite aggregation in certain taxa of fish parasites is associated with characteristics of the host, such as gender, size, habitat, schooling, feeding habits, aquatic environment and parasite development stage. This approach is intended to add new information related to determinants of change in the degree of aggregation characteristic of parasitic systems.

Materials and Methods

Data sources

The data analyzed in the present study belong to a database consisting of 73 species of fish (Table 1), with 54 marine and 19 freshwater species and a total of 3,983 specimens. The hosts and their parasites were collected between 1991 and 2009 on the coast of the state of Rio de Janeiro (21° - 23° S and 42° - 45° W) and in the Guandu River (22° 48' 2" S, 43° 37' 35" W). Fish and their parasites were identified at the Laboratory of Fish Parasitology of the Department of Animal Parasitology of the Federal Rural University of Rio de Janeiro (*Universidade Federal Rural do Rio de Janeiro* - UFRRJ) using the same methodological criteria throughout the collection period as previously described by Luque et al. (2004) and Azevedo et al. (2011). Fish are classified in the database according to species, gender, schooling, aquatic environment, habitat, feeding habits and body length. Parasite abundance was classified according to the development phase as either larvae or adult. Because of immature gonads, the gender of 237 specimens could not be identified, and they were excluded from the present study, which analyzed 3,746 individuals.

In the present study, because of the small number of samples of certain species, the parasites were grouped at higher taxa as follows: Nematoda, Monogenea, Trematoda, Cestoda, Acanthocephala, Hirudinea and Crustacea. Parasites from two taxa, Myxozoa and Mollusca, were excluded because only two host species were parasitized by species from these groups.

Variables studied

The explanatory variables used in the study were the host's gender (male/female), schooling (yes/no), aquatic environment (marine/freshwater), habitat (benthic/benthopelagic/pelagic), feeding habits (carnivore/planktivore/omnivore), and size (cm) and the parasite's development stage (larva/adult). The aggregation indices of higher taxa were considered the outcome variables.

Statistical analysis

The aggregation indices for taxa and parasitic development stages were calculated using the dispersion index by dividing the variance by the parasitic mean (VON ZUBEN, 1997). The dispersion index was chosen for the statistical analysis because it is the most accepted and widely used parameter (WILSON et al., 2002),

Table 1. Fish species that compose the analyzed database.

Species	Family	Number of hosts	Environment	Mean body size (cm)
Aluterus monoceros	Monacanthiidae	39	marine	31.2
Anchoa marinii	Engraulidae	95	marine	9.8
Anchoa tricolor	Engraulidae	103	marine	11.1
Archosargus rhomboidalis	Sparidae	29	marine	31.3
Astyanax bimaculatus	Characidae	40	freshwater	9.9
Astyanax parahybae	Characidae	40	freshwater	10.4
Astronotus ocellatus	Cichlidae	35	freshwater	20.4
Balistes capriscus	Balistidae	33	marine	35
Balistes vetula	Balistidae	18	marine	47.8
Brervoortia aurea	Clupeidae	42	marine	29.6
Caranx hippos	Carangidae	60	marine	43.9
Caranx latus	Carangidae	55	marine	33.3
Centropomus undecimalis	Centropomidae	30	marine	35.2
Cephalopholis fulva	Serranidae	30	marine	20.6
Cichla ocellaris	Cichlidae	26	freshwater	26.4
Chaetodipterus faber	Ephippidae	110	marine	27.7
Cynoscion guatucupa	Sciaenidae	73	marine	32.6
Cyphocharax gilbert	Curimatidae	60	freshwater	16.3
Dactylopterus volitans	Dactylopteridae	78	marine	22.9
Diapterus rhombeus	Gerreidae	32	marine	17.4
Euthynnus alletteratus	Scombridae	17	marine	43.9
Genidens barbus	Ariidae	63	marine	43.8
Geophagus brasiliensis	Cichlidae	50	freshwater	15.5
Genypterus brasiliensis	Ophidiidae	21	marine	42.7
Gymnothorax moringa	Muraenidae	30	marine	70.4
Gymnotus carapo	Gymnotidae	30	freshwater	36.5
Haemulon steindachneri	Clupeidae	80	marine	19.6
Hoplosternum littorale	Callichthyidae	100	freshwater	19.7
Harengula clupeola	Haemulidae	35	marine	20
Hypostomus affinis	Loricariidae	31	freshwater	27.8
Leporinus copelandii	Anostomidae	30	freshwater	34.8
Leporinus conirostris	Anostomidae	18	freshwater	36.6
Lophius gastrophysus	Lophiidae	30	marine	41.9
Loricariichthys castaneus	Loricariidae	32	freshwater	27.9
Macrodon ancylodon	Sciaenidae	31	marine	30
Menticirrhus americanus	Sciaenidae	115	marine	28.4
Merluccius hubbsi	Merluccidae	31	marine	38.5
Micropogonias furnieri	Sciaenidae	100	marine	33.2
Mugil liza	Mugilidae	34	freshwater	34.1
Mugil platanus	Mugilidae	150	marine	54.8
Mullus argentinae	Mullidae	100	marine	17.8
Mylossoma aureum	Characidae	17	freshwater	15.7
Oligoplites palometa	Carangidae	84	marine	38.2
Oligoplites saliens	Carangidae	36	marine	36.8
Oligoplites saurus	Carangidae	37	marine	29
Oligosarcus hepsetus	Characidae	40	freshwater	16.6
Pagrus pagrus	Sparidae	90	marine	29.5
Paralichthys isosceles	Paralichthyidae	36	marine	31.2
Paralonchurus brasiliensis	Sciaenidae	93	marine	21.1
Parona signata	Carangidae	31	marine	37.5
Peprilus paru	Stromateidae	81	marine	23.7
Percophis brasiliensis	Percophidae	60	marine	43.6

Table 1. Continued...

Species	Family	Number of hosts	Environment	Mean body size (cm)
Pimelodus maculatus	Pimelodidae	40	freshwater	23
Priacanthus arenatus	Priacanthidae	58	marine	37.7
Pomatomus saltatrix	Pomatomidae	55	marine	46.6
Prionotus punctatus	Triglidae	80	marine	29.3
Pseudopercis numida	Pinguipedidae	58	marine	47.7
Pseudopercis semifasciata	Pinguipedidae	66	marine	38.7
Rhamdia quelen	Heptapteridae	30	freshwater	32.7
Sarda sarda	Scombridae	24	marine	45.4
Sardinella brasiliensis	Clupeidae	35	marine	18.2
Sciadeichthys luniscutis	Ariidae	69	marine	35.5
Scomber japonicus	Scombridae	100	marine	25.8
Scomberomorus brasiliensis	Scombridae	12	marine	46.4
Selene setapinnis	Carangidae	53	marine	29.4
Sphyraena guachancho	Sphyraenidae	36	marine	36.4
Tilapia rendalli	Cichlidae	30	freshwater	22.1
Trachelyopterus striatulus	Auchenipteridae	60	freshwater	19.2
Trichiurus lepturus	Trichiuridae	55	marine	122.8
Tylosurus acus acus	Belonidae	31	marine	72.5
Uraspis secunda	Carangidae	21	marine	39.9
Urophycis brasiliensis	Phycidae	26	marine	28.5
Urophycis mystacea	Phycidae	46	marine	26.4

although it is somewhat dependent on the parasite prevalence in large samples (POULIN, 2007b).

A univariate analysis was performed, consisting of calculating the mean aggregation indices for each species and the mean aggregation for each category of explanatory variable except for host size and larvae proportion.

Data normality was tested using the Shapiro-Wilk test. The non-parametric Wilcoxon test for unpaired samples and Kruskal-Wallis test were used to compare two or more than two independent groups, respectively (KATZ, 2006a). In this study, the groups were formed by the categories of explanatory variables. Pearson's linear correlation test was used to evaluate the correlation between the aggregation and proportion of larvae and the body length of the host. Subsequently, bivariate analyses were performed to better understand the behavior of the variables.

Thereafter, all of the variables were submitted to a multiple linear regression model fitting. The stepwise technique was used to select variables and obtain the most parsimonious model (final model) to explain variations in the degree of parasite aggregation (VENABLES & RIPLEY, 2002; DOHOO et al., 2003; KATZ, 2006b).

The variables development stage and food type were included in the aggregation analysis only for the taxa whose cycles are indirect. For the analysis, the variable development stage was transformed into a proportion, and the proportion of larvae was used in the models.

Because of the low number of observations, the following categories were not analyzed in the Hirudinea taxon: pelagic (habitat variable) and planktivore (feeding habits variable).

All of the statistical calculations were performed with R software for Windows, version 2.15.2 (R DEVELOPMENT CORE TEAM, 2014) using a 5% significance level.

Results

Table 2 shows that the three highest values for mean parasite aggregation were observed for the taxon Trematoda, followed by the taxa Nematoda and Cestoda. Female hosts exhibited greater aggregation than males except for in Hirudinea and Crustacea. Host species that live in schools also presented greater aggregation than species without this characteristic in all taxa. Parasites of marine fish presented greater aggregation in Nematoda, Trematoda, Acanthocephala, Hirudinea and Crustacea. Parasites of benthopelagic fish showed greater aggregation in Nematoda, Trematoda, Acantocephala, Cestoda, Hirudinea and Crustacea. Parasites of carnivorous hosts of the Trematoda, Cestoda and Hirudinea taxa showed greater aggregation compared with other taxa.

The degree of parasite aggregation had a positive and significant correlation with larval stage in the following taxa: Nematoda [0.08 (0.04; 0.11)], Acanthocephala [0.08 (0.04; 0.13)] and Cestoda [0.28 (0.25; 0.32)] and the adult stage in Trematoda [0.14 (0.11; 0.17)] and Cestoda [0.29 (0.25; 0.32)]. Host size exhibited the same correlation type in the following taxa: Nematoda [0.18 (0.14; 0.21)], Trematoda [0.65 (0.63; 0.67)], Acanthocephala [0.23 (0.19; 0.27)], Cestoda [0.68 (0.66; 0.70)] and Crustacea [0.27 (0.23; 0.30)].

A bivariate analysis (Table 3) was performed on the results of association tests and were later complemented by the multivariate analysis. Tables 4 and 5 show the results of the initial and final models. To fit the multiple linear regression models (Table 5), an increased degree of aggregation was associated with female hosts in the Monogena and Cestoda and with male hosts in the Crustacea taxon. In other taxa, the gender variable was not included in the final regression model and excluded because of a lack of significance.

Table 2. Distribution of the means of the degrees of parasite aggregation in fish from the State of Rio de Janeiro.

Variables	Categories	Total means by taxon. and means and their respective confidence intervals (95%) by taxon according to the categories of explanatory variables.						
		Nematoda (29.10)	Monogenea (19.11)	Trematoda (63.84)	Acanthocephala (6.01)	Cestoda (23.06)	Hirudinea (2.20)	Crustacea (16.16)
Sex	Male	27.44* [24.11; 30.77]	16.10 [14.75; 17.45]	40.18 [35.77; 44.59]	5.52* [5.10; 5.94]	13.96* [12.27; 15.63]	4.79 [4.03; 4.85]	16.41 [15.01; 17.79]
	Female	30.12 [26.87; 33.35]	19.98 [18.24; 21.74]	71.72 [60.55; 82.89]	6.73 [6.29; 7.17]	29.50 [25.54; 33.44]	3.86 [3.30; 4.06]	16.11 [14.71; 17.49]
School	Yes	33.39 * [30.46;36.32]	18.51 * [17.21; 19.83]	66.43* [59.94; 74.92]	6.65* [6.28; 7.02]	23.07 * [20.36 25.76]	5.88* [5.00; 5.74]	17.49* [16.32; 18.64]
	No	15.14 [11.67;13.81]	15.99 [14.33; 16.67]	13.39 [12.23; 14.53]	4.11 [3.94; 4.26]	18.25 [16.16; 20.34]	1.41 [1.36; 1.44]	12.01 [10.29; 13.71]
Environment	Marine	33.55* [30.73; 36.35]	16.58* [15.35; 17.83]*	64.32* [56.97; 71.69]	6.43 [6.08; 6.78]	21.68 [19.21; 24.13]	4.94 [4.13; 4.87]	16.67* [15.55; 17.79]
	Freshwater	10.51 [7.71; 13.29]	27.01 [24.72; 29.32]	18.24 [15.88; 20.56]	3.42 [3.26; 3.58]	30.84 [27.54; 34.14]	3.18 [2.89; 3.43]	11.98 [10.20; 13.76]
Habitat	Benthic	17.58 [15.80; 19.32]	26.55** [24.05; 29.09]	11.52** [11.05; 11.99]	2.45** [2.32; 2.58]	8.46** [7.55; 9.35]	3.22** [3.02; 3.42]	12.52** [11.00; 14.03]
	Benthic pelagic	38.01 [33.24; 42.76]	15.45 [14.12; 16.78]	92.25 [78.54; 105.94]	11.80 [11.14; 12.44]	41.92 [37.00; 46.82]	5.52 [4.91; 6.11]	24.48 [22.69; 26.27]
	Pelagic	29.28 [25.31; 33.25]	10.52 [9.81; 11.21]	68.49 [63.72; 73.28]	3.75 [3.47; 4.00]	7.26 [6.57; 7.93]	†	6.68 [5.99; 7.37]
Feeding habits	Carnivorous	28.86** [26.41; 31.29]	7.51** [7.05; 7.97]	133.90** [110.51; 157.29]	4.73** [4.42; 5.00]	50.20** [42.82; 57.58]	12.20** [10.97; 13.45]	12.30** [10.79; 13.79]
	Planktivorous	1.93 [1.88; 1.98]	1.95 [1.80; 2.0]	65.17 [55.90; 74.44]	1.01 [1.00; 1.02]	1.50 [1.48; 1.48]	†	14.92 [13.33; 16.55]
	Omnivorous	31.57 [28.26; 34.84]	22.74 [21.22; 24.28]	26.49 [24.87; 28.11]	7.10 [6.68; 7.52]	8.60 [7.97; 9.19]	2.65 [2.55; 2.71]	17.93 [16.60; 19.24]

(*) Significant Wilcoxon Test. (**) Significant Kruskal-Wallis Test. (†) Excluded category.

Table 3. Bivariate analysis of the parasite aggregation in fish from the State of Rio de Janeiro.

Variables	Regression coefficients and their respective confidence intervals (95%). according to the taxa						
	Nematoda	Monogenea	Trematoda	Acanthocephala	Cestoda	Hirudinea	Crustacea
Sex (Ref. Male)	2.67 [-2.26; 7.61]	3.88* [1.40; 6.37]	31.54* [19.40; 43.68]	1.22* [0.40; 2.04]	15.54* [10.32; 20.76]	-0.93 [-1.91; 0.04]	-0.30 [-2.54; 1.94]
School (Ref. Yes)	-20.64* [-26.52; -14.75]	-2.52 [-5.72; 0.68]	-54.04* [-68.76; - 39.31]	-2.53* [-3.57; -1.50]	-4.82 [-11.18; 1.54]	-4.47* [-5.46; -3.47]	-5.48* [-8.15; -2.80]
Environment (Ref. Marine)	23.05* [-29.09; -17.00]	10.42* [6.86; 14.00]	-46.09* [-61.72; -30.45]	-3.01* [-4.44; -1.59]	9.15 [-4.69; 23.00]	-1.77* [-2.79; -0.75]	-4.69* [-8.62; -0.77]
Habitat (Ref. Benthic)							
Benthic pelagic	20.43* [14.93; 25.93]	-11.10* [-13.89; -8.32]	80.73* [67.32; 94.13]	9.35* [8.52; 10.17]	33.46 * [27.67; 39.25]	2.29* [1.33; 3.26]	11.96* [9.49; 14.43]
Pelagic	11.70* [4.77; 18.63]	-16.03* [-19.57; -12.49]	56.96* [40.43; 73.49]	1.30* [0.32; 2.28]	-1.20 [-8.14; 5.74]	†	-5.84* [-8.78; -2.91]
Feeding habits (Ref. carnivorous)							
Planktivorous	-26.93* [-37.39; -16.47]	#	-68.77* [-93.46; -44.08]	-3.72* [-5.68; -1.76]	-48.70* [-61.99; -35.41]	†	#
Omnivorous	2.71 [-3.00; 8.42]	#	-107.45* [-121.35; -93.55]	2.37 * [1.44; 3.29]	-41.60* [-47.00; -36.20]	#	#
Body length	0.70* [0.56; 0.83]	-0.05 [-0.12;0.01]	6.83* [6.57; 7.09]	0.11* [0.09; 0.13]	2.46* [2.35; 2.56]	0.17 * [0.12; 0.21]	0.44* [0.38; 0.50]
Larval stage	13.25* [7.54; 18.96]	#	-0.22 [-22.47; 22.02]	2.44* [1.16; 3.71]	44.91* [39.04; 50.78]	#	#

(*) p-value < 5%. (†) Category not analized. (#) Variable not analized.

Table 4. Initial model of the multivariate linear regression analysis of parasite aggregation in fish from the State of Rio de Janeiro.

Variables/ metric	Categories	Regression coefficients and their respective confidence intervals (95%) according to the taxa						
		Nematoda	Monogenea	Trematoda	Acanthocephala	Cestoda	Hirudinea	Crustacea
Sex (Ref. Male)		-1.43 [-6.29; 3.44]	3.83 [1.38; 6.28]	4.00 [-4.54; 12.54]	0.37 [-0.33; 1.08]	4.93 [1.33; 8.53]	-0.22 [-1.14; 0.71]	-2.69 [-4.76; -0.63]
School (Ref. yes)		-13.82 [-20.33; 7.31]	-8.83 [-12.44; -5.21]	-93.42 [-104.96; -81.88]	1.61 [0.65; 2.58]	-34.88 [-39.81; -29.95]	-6.75 [-7.82; -5.67]	-14.16 [-16.99; -11.33]
Environment (Ref. marine)		-16.84 [-23.66; -10.03]	11.87 [8.00; 15.75]	72.36 [60.48; 84.24]	-8.27 [-9.61; -6.92]	30.21 [20.70; 39.71]	1.61 [0.50; 2.72]	-2.4 [-6.21; 1.40]
Habitat (Ref. benthic)	Benthic pelagic	13.16 [7.23; 19.09]	-13.36 [-16.55; -10.18]	-16.99 [-27.26; -6.72]	11.56 [10.64; 12.48]	-0.39 [-4.76; 3.98]	-0.68 [-1.70; 0.33]	3.19 [0.57; 5.82]
	Pelagic	32.57 [22.57; 42.57]	-3.33 [-8.80; 2.14]	-21.88 [-39.46; -4.29]	6.52 [5.12; 7.92]	-41.66 [-48.33; -34.99]	†	-10.02 [-12.94; -7.11]
Feeding habits (Ref. carnivorous)	Planktivorous	-20.73 [-32.82; -8.64]	#	168.97 [147.62; 190.31]	-1.36 [-3.19; 0.46]	44.93 [34.70; 55.15]	†	#
	Omnivorous	27.57 [19.84; 35.30]	#	-4.28 [-18.28; 9.72]	5.99 [4.85; 7.13]	31.60 [-37.04; -26.15]	#	#
Body size (cm)		0.69 [0.52; 0.86]	0.15 [0.07; 0.23]	8.39 [8.09; 8.69]	0.03 [0.01; 0.06]	2.23 [2.11; 2.34]	0.30 [0.24; 0.35]	0.34 [0.28; 0.40]
Larval stage		7.19 [1.07; 13.31]	#	49.29 [33.21; 65.38]	2.58 [1.46; 3.70]	25.14 [20.74; 29.54]	#	#
R² ajusted		8%	7%	53%	32%	56%	14%	17%

(†) Category not analized. (#) Variable not analized.

Table 5. Final model of the multivariate linear regression analysis of parasite aggregation in fish from the State of Rio de Janeiro.

Variables/metric	Categories	Regression coefficients and their respective confidence intervals (95%) according to the taxa						
		Nematoda	Monogenea	Trematoda	Acanthocephala	Cestoda	Hirudinea	Crustacea
Sex (Ref. Male)		-	4.00 [1.53; 6.47]	-	-	4.93 [1.33; 8.53]	-	-2.64 [-4.70; -0.57]
School (Ref. Yes)		-13.74 [-20.24; -7.23]	-12.76 [-16.21; -9.32]	-93.13 [-104.65; -81.60]	1.77 [0.77; 2.76]	-34.88 [-39.81; -29.95]	-6.57 [-7.60; -5.55]	-14.49 [-17.28; -11.69]
Environment (Ref. marine)		19.17 [-23.63; -12.71]	14.60 [10.76; 18.43]	71.98 [60.14; 83.83]	-8.17 [-9.52; -6.82]	30.21 [20.70; 39.71]	1.68 [0.58; 2.77]	-
Habitat Ref.benthic)	Benthic pelagic	12.03 [6.19; 17.86]	-16.50 [-19.59; -13.41]	-17.15 [-27.41; -6.89]	11.40 [10.48; 12.32]	-0.39 [-4.76; 3.98]	-	2.78 [0.24; 5.32]
	Pelagic	30.82 [20.95; 40.68]	-18.29 [-21.97; -14.60]	-22.18 [-39.75; -4.61]	7.09 [5.69; 8.49]	-41.66 [-48.33;-34.99]	†	-9.96 [-12.88; -7.05]
Feeding habits (Ref. carnivorous)	Planktivorous	-21.52 [-33.60; -9.45]	#	169.32 [147.99; 190.65]	-1.67 [-3.50; 0.16]	44.93 [34.70; 55.15]	†	#
	Omnivorous	27.80 [20.09; 35.51]	#	-3.92 [-17.90; 10.06]	6.31 [5.17; 7.44]	-31.60 [-37.04 -26.15]	#	#
Body size (cm)		0.70 [0.54; 0.87]	0.09 [0.01; 0.16]	8.41 [8.11; 8.71]	0.05 [0.03; 0.07]	2.23 [2.11; 2.34]	0.28 [0.23; 0.33]	0.35 [0.29; 0.41]
Larval stage		-	#	49.54 [33.47; 65.62]	1.36 [-2.69; -0.04]	25.14 [20.74; 29.54]	#	#
R² ajusted		8%	6%	53%	31%	56%	14%	17%

(†) Category not analized. (-) Variable excluded. (#) variable not analized.

Freshwater schooling host species significantly influenced the increased parasite aggregation in most taxa. Parasite aggregation was higher in the Monogenea, Trematoda and Cestoda in benthic host species, Acanthocephala and Crustacea in benthopelagic species and Nematoda in pelagic species. In the Nematoda and Acanthocephala, the increased parasitic aggregation was significantly associated with omnivorous fish species, whereas in the Cestoda and Trematoda, it was associated with planktivorous species. The larval development stage remained in the final model in the Trematoda, Acanthocephala and Cestoda and contributed to increased parasite aggregation; however, this parameter was only calculated for species that develop in an indirect cycle.

Discussion

Studies on the degree of parasite aggregation should use a comparative approach to obtain results that provide a better explanation of the factors that determine this characteristic of

parasitic populations. Fish are considered a good model for studies on the aggregate distribution of parasites as well as other types of ecological aspects because they are easily obtained and consequently provide sufficient samples for statistical analyses (LUQUE et al., 2013).

Studies on the structure of parasitic fish communities in Brazil have provided results on parasite aggregation (LUQUE et al., 1996; ISAAC et al., 2000; LUQUE & ALVES, 2001; LUQUE et al., 2008); however, they have not tested the association of parasite aggregation with any biological characteristics of the hosts, which drastically decreases the possibility of extracting patterns that might contribute to explaining the variability of aggregation. Furthermore, this information is included in studies that describe parasitic communities with no comparative purpose.

In the present study, problems detected in previous analyses were minimized. Such problems were attributed to databases that were compiled from different sources and used in these analyses (LUQUE et al., 2004). Differences in the methods used to detect and identify parasites in different sources may cause reading variations in the results. Furthermore, different host species included in the same data set often are originated from different geographical areas. The availability of different parasite species has a strong regional influence because of variation between regions, which adds another element of fluctuation to these datasets. Therefore, the consistency of the results of the present study was improved because the dataset included fish from the same geographical location, and all of the hosts and parasites were examined and identified using the same standards of a single research group. In addition, the present study tested the significance of biotic variables as determinants of the degree of parasite aggregation, which had not been tested to date.

The results of the bivariate and multivariate analyses are complementary and demonstrate the active strength of one variable acting on another when they are analyzed simultaneously, which can change the significance status. In addition, these results corroborate the importance of regression analyses to evaluate true associations because such analyses minimize the action of confounding variables and thus prevent erroneous conclusions (MEDRONHO et al., 2009). Thus, the multifactorial nature of variations in the population dynamics of parasites, which is expressed as parasite aggregation, becomes evident.

The influence of a fish species' ability to form schools on the degree of parasite aggregation is presented as a determining factor in parasite diversity; however, the various results require additional investigation (MORAND et al., 2000). The formation of schools might be expected to allow greater access of parasitic groups to their hosts because schools increase the size of the resource to be explored from a macroecological perspective, and schooling can influence the abundance of certain parasites, susceptibility of hosts to infection and parasite aggregation values, which was observed in the present study for all parasite taxa except Acanthocephala.

The feeding habits of hosts may also be associated with the degree of parasite aggregation, specifically for endoparasites that are generally transmitted at a trophic level. Because of the amplitude of their trophic spectrum, which is diversified and less specialized than that of fish with more restricted diets (carnivore and planktivore), omnivorous fish would be expected to have access

to a greater number and diversity of parasites, which would also influence the degree of aggregation according to diet. However, this behavioral pattern was not observed in the present study, which might have been caused by biological differences in the set of hosts studied in our sample. This explanation can also be applied to differences found in aggregation values, which may be related to different feeding behaviors among fish from different habitats. Benthic fish have a more generalist diet and pelagic fish a more specialized diet; this behavior is consistent with our results, which showed that benthic fish exhibited a greater degree of aggregation in most taxa.

Host size, which is an indirect measure of the degree of susceptibility to parasitic infections, can act as a determinant of variations in the degree of parasite aggregation (POULIN, 2013). However, when analyzing size variations in fish from the studied samples, Poulin (2013) did not observe significant effects on the variability of parasite aggregation, either in the entire sample or within a particular group of parasites. In the present study, however, the host size variable was evaluated as a determinant of parasite aggregation between the sizes of fish from different samples and not within each sample, and the results were significant. This variable initially explains most of the parasite distribution, abundance and diversity in a particular host (LUQUE et al., 2004). The association between the degree of aggregation and body length was expected because hosts with greater length should be able to host a greater number of parasites and because body size is a good measure of total nutrients or energy available in a host to support a parasite species (LUQUE et al., 2004). Therefore, the values of parasite abundance may be higher in hosts with greater total length, thereby increasing the possibility of a higher degree of aggregation.

A similar situation may occur for differences in the degree of aggregation between different parasitic stages (larvae and adults). However, Poulin (2013) did not observe significant effects of the development stage on aggregation levels, which is inconsistent with the results of Lester (2012), who associated the discrepancies with differences in sample size and number of species, which were higher in his study, and a more rigorous assessment of species-specific effects. In the present study, aggregations significantly increased with increases in the proportion of larvae, which indicates that the development stage is a determinant of aggregation. Because of different levels of host specificity, which is greater in larvae than in adults, a greater degree of aggregation was expected for parasites in the larval stage.

In the present study, the factors that could be involved in aggregation were evaluated for each higher taxon, whereas Poulin (2013) tested the development stage and taxon as possible determinants of aggregation. Furthermore, the characteristics of the databases used in the analyses are quite different. Thus, methodological issues may partly explain the different results observed among studies. Moreover, different results may also be a result of the biological characteristics of each parasite species because aggregation as a function of the development stage can be influenced by the particular characteristics of the different biological cycles. Therefore, a comparative analysis by parasite species and an analysis by higher taxa would be required to clarify this issue.

Regarding host gender and its possible association with the degree of parasite aggregation, the results are heterogeneous, and there are no clearly defined standards that could be attributed to differences in biological and behavioral aspects between male and female hosts. Because there are no studies on the biology and population dynamics of most hosts, it is difficult to establish whether these differences may be true standards in the distribution of parasites or just stochastic results as discussed in studies on the ecology of parasitic communities of marine fish (LUQUE et al., 1996; LUQUE & ALVES, 2001).

Except for the body size and development stage, the tested variables showed a significance degree of the taxon dependence. On a larger scale, other factors may influence the aggregation of fish parasites in the region because the correlation coefficients for all taxa and stages were lower than 60%. Macroecological changes may influence parasite abundance (LUQUE & POULIN, 2008), and according to Braga (2001), the coastal area of Rio de Janeiro is strongly influenced by upwelling systems and subtropical convergence, which are important for the feeding and reproduction of marine organisms (ODEBRECHT & CASTELLO, 2001). This oceanographic transition zone may represent the northern limit of species from cooler regions or the southern limit for more tropical species, and this zone also receives migratory birds and aquatic mammals from the two hemispheres that may act as definitive hosts of several parasite species of marine fish. The differences between the degrees of parasite aggregation of freshwater and marine fish also reinforce the possibility of macroecological influences on aggregation. Although Marcogliese & Cone (1997) and Luque & Poulin (2008) did not detect differences between the parasite diversity of marine and freshwater fish, the possibility that different environmental conditions may influence the degree of aggregation cannot be discounted.

Although parasite aggregation is considered an intrinsic property of parasitic processes, the results of the present study highlight the need to expand investigations by using more comprehensive databases to analyze the combined action of factors with a significant potential influence on the degrees of aggregation and evaluate the role of these factors in the transmission of parasitic diseases.

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