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Metazoan parasites as biological tags for stock discrimination of whitemouth croaker *Micropogonias furnieri*

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Examination of 248 adult specimens of whitemouth croaker *Micropogonias furnieri* from five localities along the Brazilian coast revealed 8735 parasites belonging to 41 metazoan species. Samples from Ceará to Bahia and Rio de Janeiro to Santa Catarina showed a high level of correct assignment (92 and 87%, respectively) and cross assignment (*i.e.* almost all specimens misidentified in Ceará were assigned to Bahia and almost all specimens misidentified in Bahia were classified as Ceará), so samples were pooled in the northern and south-eastern samples, and Rio Grande do Sul was considered a southern area. Eight parasite species were characteristic of the northern localities, five species were found just in the area associated with south-eastern localities and two species were characteristic of the southern area providing first evidence of stock discreteness. The multivariate discriminant analysis successfully discriminated three groups of localities associated with three stocks of *M. furnieri* in Brazil: a northern stock associated with Ceará and Bahia, a south-eastern stock related to Rio de Janeiro and Santa Catarina and a southern stock in the area of Rio Grande do Sul, which could be considered as the northern limit of the stock associated with the Common Fishing Zone of Uruguay and Argentina.

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Key words: Brazil; fisheries management; stocks.

INTRODUCTION

Micropogonias furnieri (Desmarest), the whitemouth croaker (Sciaenidae), has a latitudinal distribution along the Atlantic Ocean coast of America from Veracruz, México (20° 20' N) to El Rincón, Argentina (41° 00' S) (Juareguizar *et al.*, 2003). It supports both industrial and local fisheries in Venezuela, Argentina, Uruguay and Brazil (Vizziano *et al.*, 2002; Gómez & Guzmán, 2005). *Micropogonias furnieri* is an important resource with reported landings for 1995–2000 amounting to 28.1% of local catch and 16.7% of the industrial landings in the marine coastal system of southern Brazil (Vasconcellos *et al.*, 2007). It was the most frequently landed species (4070 t) in the State of São Paulo, representing 17.1% during the last 4 years

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(Mendonça & Miranda, 2008). Also, *M. furnieri* is the most important resource for the Uruguayan and Argentinean artisanal and industrial coastal fishery associated with the Rio de la Plata fisheries (Norbis & Verocai, 2005).

For management purposes, *M. furnieri* is considered as an unitary stock caught in the Rio de la Plata and Maritime front (Arena & Rey, 2000), but its stock structure is not well known. Studies based on morphometric and meristic characters (Figueroa & Díaz de Astarloa, 1991) as well as allozyme analysis (Maggioni *et al.*, 1994) did not reveal more than one stock in the main fishing area (southern Brazil, Uruguay and Argentine), but Norbis & Verocai (2005) claimed the existence of two groups, but not two discrete stocks, based on morphometric and age analysis of sagitta otoliths from fish caught in the Rio de la Plata coastal area. Norbis & Verocai (2005) concluded that the two groups found during the spawning season were members of a unitary stock. Haimovici & Umpierre (1996) suggested the presence of two groups (but not stocks) in southern Brazil, based on their migratory behaviour. Most recently, Vasconcellos & Haimovici (2006) suggested the existence of at least two stocks in southern Brazil, with a boundary in the area of Cabo Santa Marta Grande, but the separation between a southern Brazilian stock and the exploited stock in the Common Fishing Zone of Uruguay and Argentina remains uncertain. The status of the *M. furnieri* population along the coast of Brazil is therefore unclear.

Due to the importance of *M. furnieri* from both the Brazilian and Argentinean fisheries, a clear definition of the population structure is a pre-requisite for a rational management of this resource. Currently, the stock structure of *M. furnieri* is not clear, and contradictory results have been reached using different techniques. Here, data on metazoan parasites of *M. furnieri*, caught along a latitudinal gradient of *c.* 29° and >3500 km of coastline, extending from Fortaleza (*c.* 3° 44' S) to Rio Grande do Sul (*c.* 32° 15' S) were analysed in order to test whether more than one stock or discrete populations of *M. furnieri* are present in the area under study.

MATERIALS AND METHODS

From September 2003 to June 2006, 248 specimens of *M. furnieri*, ranging from 23 to 69 cm total length (L_T) (mean \pm s.d. 48.7 ± 9.6 cm) were obtained from local fishermen in five Brazilian localities (Fig. 1 and Table I). In addition, a previous sample ($n = 34$) taken in 1999 from Pedra de Guaratiba, State of Rio de Janeiro, was included in this study. Fish were frozen (-18°C) until examination, L_T (to the nearest cm) and sex were determined after thawing. All specimens were examined first for metazoan ectoparasites (skin, gills and mouth cavity) and then for metazoan endoparasites. All viscera, including heart and blood vessels were examined. To quantify parasites, each organ was dissected separately and washed in running water and all the material retained on a 154 μm mesh was examined stereomicroscopically. Parasites were fixed, preserved and stained with standard techniques (Amato *et al.*, 1991). Prevalence, abundance and mean abundance were calculated according to Bush *et al.* (1997). Univariate analyses were performed to evaluate the infections at the infrapopulation levels. Significance of the differences in mean L_T of the fish host was evaluated by ANOVA. Non-parametric tests were used to evaluate significance in mean abundance and prevalence of infection. Multivariate discriminant analysis (MDA) at infracommunity level was used to test whether metazoan parasite communities could be a good predictor for localities. Analyses were performed following the recommendation of Wilkinson (1990) using SYSTAT (version 8.0; SPSS Inc.; www.spss.com) as the statistical tool. Data were transformed by $\log_{10}(x + 1)$.

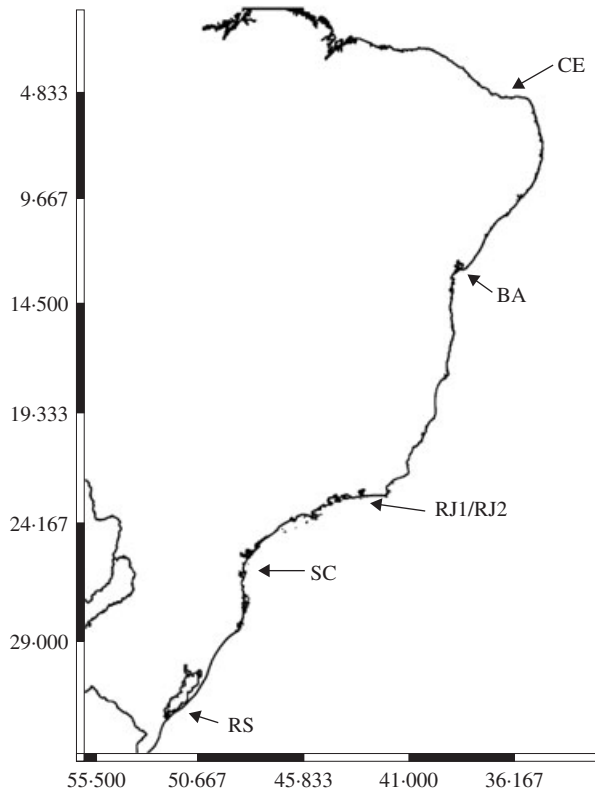


FIG. 1. Brazilian littoral showing approximate position of the localities where samples were taken (see Table I).

RESULTS

A total of 8735 parasites belonging to 41 metazoan taxa (24 identified to species; Table II), comprising, three Monogenea, one Aspidogastrea, 12 Digenea, four larval Cestoda, one larval Acanthocephala, three adult Acanthocephala, five larval Nematoda, five adult Nematoda, one Hirudinea, four Copepoda and two Isopoda, were

TABLE I. Locality, geographic co-ordinates, sample size (n) and mean \pm s.d. total length (L_T) of the specimens of *Micropogonias furnieri* from the studied localities

Locality (code)	Latitude and longitude	n	L_T (cm)
Fortaleza, Ceará (CE)	3° 40' S; 38° 30' W	50	35.36 \pm 12.48
Ilhéus, Bahia (BA)	14° 48' S; 30° 01' W	52	45.62 \pm 4.38
Pedra de Guaratiba, Rio de Janeiro 1 (2003–2004) (RJ 1)	23° 01' S; 43° 38' W	59	52.75 \pm 5.01
Pedra de Guaratiba, Rio de Janeiro 2 (1997) (RJ 2)	23° 01' S; 43° 38' W	34	53.97 \pm 5.77
Florianópolis, Santa Catarina (SC)	27° 47' S; 46° 25' W	50	53.01 \pm 2.93
Cassino Beach, Rio Grande do Sul (RS)	32° 20' S; 52° 00' W	36	54.54 \pm 4.60

TABLE II. Prevalence (Prev.) and mean \pm s.d. abundance of infection (MA) of the metazoan parasites of *Micropogonias furnieri* in Brazil (for locality codes see Table I).

Parasite species	SI	Localities													
		CE		BA		RJ1		RJ2		SC		RS			
		Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA		
Trematoda															
<i>Aponurus pyriformis</i>	Stomach	1.0	0.1 \pm 0.4	7.7	0.8 \pm 3.5	5.8	0.8 \pm 0.3	4.9	0.6 \pm 0.8	4.0	0.6 \pm 0.3	2.7	0.6 \pm 0.9		
<i>Aponurus laguncula</i>	Stomach	7.5	0.8 \pm 1.2	38.5	2.3 \pm 6.2	—	—	—	—	4.0	0.4 \pm 0.3	2.7	0.4 \pm 0.7		
<i>Aponurus</i> sp.	Stomach	2.5	0.2 \pm 0.3	13.5	0.3 \pm 0.7	—	—	—	—	—	—	5.6	0.2 \pm 0.7		
<i>Bucephalus varicus</i>	Intestine	—	—	5.8	0.8 \pm 0.3	—	—	—	—	—	—	—	—		
Didymozoidae larvae*	Intestine	—	—	48.8	1.2 \pm 2.2	3.4	0.3 \pm 0.4	—	—	—	—	—	—		
Hemitiuridae gen. sp.	Intestine	—	—	9.6	0.2 \pm 0.5	—	—	—	—	—	—	—	—		
<i>Lecithaster falcatus</i>	Intestine	—	—	13.5	0.3 \pm 1.1	—	—	—	—	—	—	—	—		
<i>Lecithochirium microstomum</i>	Intestine	—	—	—	—	15.3	0.5 \pm 2.6	24.4	0.4 \pm 0.5	—	—	—	—		
<i>Lobatostoma ringens</i>	Intestine	7.5	0.6 \pm 0.4	3.8	0.8 \pm 0.4	22.3	0.4 \pm 0.7	12.2	0.1 \pm 0.3	6.0	0.1 \pm 0.2	13.9	0.1 \pm 0.3		
<i>Opecoeloides catarinensis</i>	Intestine	—	—	—	—	—	—	—	—	4.0	0.4 \pm 0.2	8.3	0.4 \pm 0.5		
<i>Opecoeloides polynemi</i>	Intestine	5.0	0.5 \pm 0.6	26.9	0.8 \pm 1.7	—	—	—	—	—	—	—	—		
<i>Opecoeloides stenosomae</i>	Intestine	7.5	0.8 \pm 1.3	3.8	0.4 \pm 0.3	16.9	0.6 \pm 1.3	7.3	0.7 \pm 1.2	6.0	0.8 \pm 0.3	8.3	0.8 \pm 1.3		
<i>Pachycreadium gastrocotylum</i>	Intestine	5.0	0.3 \pm 1.2	—	—	11.9	0.6 \pm 2.3	9.8	0.9 \pm 1.6	6.0	0.8 \pm 0.3	36.1	0.8 \pm 1.4		
Monogenea															
<i>Encyrtillabe spari</i>	Mouth	2.5	0.4 \pm 0.9	1.9	0.2 \pm 0.5	16.9	0.2 \pm 0.3	12.2	0.1 \pm 0.2	8.0	0.8 \pm 1.3	13.9	0.8 \pm 0.5		
<i>Macrovalvirema sinaloense</i>	Gills	35.0	0.9 \pm 1.3	38.5	0.9 \pm 1.6	74.6	5.1 \pm 4.8	63.4	0.9 \pm 1.0	42.0	1.2 \pm 2.2	55.6	1.2 \pm 2.3		

TABLE II. Continued

Parasite species	SI	Localities											
		CE		BA		RJI		RJ2		SC		RS	
		Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA
<i>Pterinotrematoides mexicanum</i>	Gills	12.5	0.6 ± 1.8	36.5	0.7 ± 1.2	71.2	6.6 ± 10.5	87.8	1.1 ± 1.7	5.0	1.2 ± 1.8	58.3	1.2 ± 2.2
<i>Callitetrarhynchus gracilis*</i>	Mesenteries	—	—	—	—	5.8	0.6 ± 1.2	9.8	0.1 ± 0.3	6.0	0.6 ± 0.3	2.8	0.6 ± 1.3
<i>Nybelinia</i> sp.*	Mesenteries	—	—	5.8	0.6 ± 0.8	—	—	—	—	—	—	—	—
<i>Pterobothrium heteracanthum*</i>	Mesenteries	2.5	0.2 ± 0.8	5.8	0.3 ± 1.1	18.6	0.9 ± 6.1	22.0	2.8 ± 2.6	12.0	0.2 ± 0.3	22.2	0.2 ± 0.3
<i>Scolex polymorphus</i>	Intestine	—	—	—	—	—	—	2.4	0.3 ± 0.6	—	—	—	—
<i>Acanthocephala</i>	Intestine	—	—	1.9	0.2 ± 0.3	—	—	—	—	4.0	0.4 ± 0.1	—	—
<i>Acanthocephala</i> gen. sp.	Intestine	—	—	—	—	—	—	—	—	—	—	—	—
<i>Corynosoma australe*</i>	Mesenteries	7.5	0.7 ± 2.6	—	—	15.3	0.3 ± 9.8	34.1	6.1 ± 4.9	1.0	0.1 ± 0.1	58.3	0.1 ± 0.3
<i>Dolifusentis chandleri</i>	Intestine	1.0	0.8 ± 0.6	5.8	0.1 ± 0.5	—	—	2.4	0.3 ± 0.5	—	—	—	—
<i>Rhadinorhynchus</i> sp.	Intestine	—	—	—	—	—	—	—	—	2.0	0.2 ± 0.3	2.8	0.2 ± 0.6
Nematoda													
<i>Anisakis</i> sp. larvae*	Mesenteries	2.5	0.2 ± 1.0	—	—	—	—	—	—	—	—	—	—
<i>Contracaecum</i> sp. larvae*	Mesenteries	—	—	—	—	11.9	0.3 ± 0.8	—	—	—	—	—	—
<i>Cucullanus</i> sp.	Intestine	25.0	1.3 ± 3.6	63.5	2.5 ± 3.1	3.6	2.8 ± 1.7	7.3	6.8 ± 9.3	76.0	7.4 ± 10.4	58.3	7.4 ± 11.4
<i>Dichelyne</i> (C.) <i>sciaenidicola</i>	Intestine	22.5	1.6 ± 3.1	86.5	4.2 ± 5.4	78.0	2.3 ± 3.6	14.9	13.8 ± 21.8	78.0	19.9 ± 22.9	75.0	19.9 ± 24.3

TABLE II. Continued

Parasite species	SI	Localities											
		CE		BA		RJI		RJ2		SC		RS	
		Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA	Prev. (%)	MA
<i>Hysterothylacium</i> sp. larvae*	Mesenteries	27.5	0.4 ± 1.0	32.7	0.8 ± 1.5	15.3	0.5 ± 0.4	14.6	1.0 ± 1.4	26.0	1.5 ± 0.3	36.1	1.5 ± 1.7
<i>Procamallanus pereirai</i>	Intestine	—	—	13.5	0.4 ± 1.0	—	—	—	—	—	—	—	—
<i>Pseudocapillaria magalhaesi</i>	Coelomic cavity	—	—	—	—	—	—	12.2	0.3 ± 0.7	—	—	—	—
<i>Raphidascaris</i> sp. larvae*	Mesenteries	2.5	0.2 ± 0.3	26.9	2.3 ± 6.8	—	—	—	—	2.0	0.4 ± 1.2	—	—
<i>Raphidascaris</i> sp. adult	Intestine	—	—	46.2	2.6 ± 5.3	—	—	—	—	—	—	—	—
<i>Terranova</i> sp. larvae*	Mesenteries	17.5	1.6 ± 3.9	67.4	3.5 ± 4.8	11.9	0.2 ± 0.4	7.3	0.9 ± 1.6	6.0	0.8 ± 0.3	8.3	0.8 ± 1.3
Hirudinea	Gills	7.5	0.2 ± 1.4	1.9	0.2 ± 0.3	3.4	0.3 ± 0.2	7.3	0.6 ± 0.8	6.0	0.6 ± 0.4	8.3	0.6 ± 1.4
Copepoda	Gills	—	—	—	—	—	—	2.4	0.3 ± 0.6	—	—	—	—
<i>Bomolochus paucus</i>	Gills	—	—	—	—	16.9	0.3 ± 0.8	24.4	0.5 ± 0.8	14.0	0.2 ± 0.5	2.8	0.2 ± 0.3
<i>Caligus haemulonis</i>	Gills	7.5	0.2 ± 0.3	3.8	0.6 ± 0.3	8.5	0.1 ± 0.4	9.8	0.9 ± 1.2	6.0	0.6 ± 0.4	27.8	0.6 ± 1.2
<i>Clavellotis dilatata</i> *	Gills	2.5	0.2 ± 0.3	—	—	3.4	0.7 ± 0.3	7.3	0.9 ± 1.3	2.0	0.2 ± 0.3	5.6	0.2 ± 0.4
<i>Neobrachiella chevreuxii</i> *	Gills	—	—	—	—	—	—	—	—	—	—	—	—
Isopoda	Mouth	2.5	0.4 ± 0.2	23.8	0.4 ± 1.2	1.2	0.3 ± 1.0	12.2	0.6 ± 0.8	2.0	0.2 ± 0.3	8.3	0.2 ± 0.3
Cymothoidae gen. sp.	Mouth	—	—	26.9	0.9 ± 2.9	—	—	—	—	—	—	—	—
<i>Gnathia</i> sp.	Mouth	—	—	—	—	—	—	—	—	—	—	—	—

SI, site of infection; *, long-lived parasites.

collected from the 282 specimens of *M. furnieri*. Of the whole fish sample, 93.97% were parasitized with at least one parasite species.

Table I shows the characteristic of the samples for each locality. An ANOVA showed that mean L_T differed significantly between localities ($F_{5,276}$, $P < 0.001$). A Tukey's test demonstrated that fish from Rio de Janeiro, Rio Grande do Sul and Santa Catarina were significantly larger than those from Ceará and Bahia. For the whole sample, all specimens were larger than the size at first maturity that is 19.2–20.4 cm L_T according to Vizziano *et al.* (2002).

Only a few parasite species (three from Rio de Janeiro, two from Rio Grande do Sul and one for Santa Catarina and Bahia) showed a significant correlation (Pearson correlation coefficient, r , $P < 0.05$ for all significant relationships) between L_T and abundance [$\log_{10}(x + 1)$]. Therefore, parasite counts were not adjusted for L_T and the analyses included the whole sample rather than those of similar host age groups (Oliva & Ballón, 2002).

The parasite species found in *M. furnieri* for each locality, prevalence and mean abundance are given in Table II. There is no evidence of a geographic tendency in the population descriptors, except for a few species such as the copepod *Caligus haemulonis* that showed a lower prevalence in the southern locality, but the digenean *Pachycreadium gastrocotylum* and the larval acanthocephalan *Corynosoma australe* increased in prevalence from north to south. From these data (Table II) it is evident that the parasite fauna of *M. furnieri* shows qualitative and quantitative differences along the area under analysis.

The MDA of abundance data, for the whole sample, suggested a good discrimination function (correct assignment 71%, Wilks' λ *c.* 0.041; $F_{205,1178}$, $P < 0.001$). Because samples from Ceará and Bahia showed high levels of correct assignment (92 and 87% respectively) and cross assignment (misidentified fish from Ceará

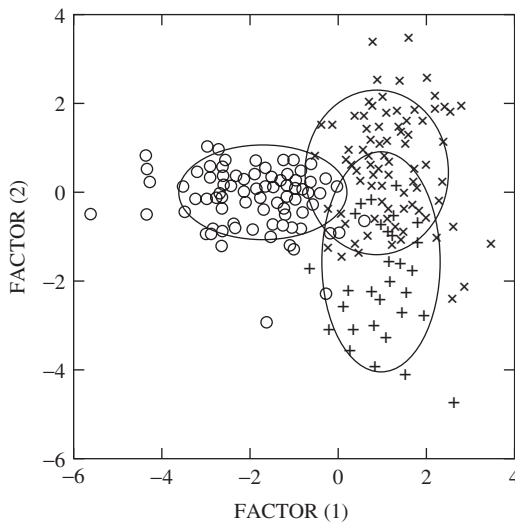


FIG. 2. Plot of multivariate discriminant analyses of northern stock *Micropogonias furnieri* (Ceará and Bahia) (○), south-eastern stock (Rio de Janeiro and Santa Catarina) (×) and southern stock (Rio Grande do Sul) (+).

TABLE III. Discriminant analysis classification showing the numbers and percentages of *Micropogonias furnieri* classified in each zone (rows correspond to group memberships): northern (Ceará and Bahia), south-eastern (Rio de Janeiro and Santa Catarina) and southern (Rio Grande do Sul)

Region	Northern	South-eastern	Southern	% correct*
Northern	96	2	4	94
South-eastern	20	102	21	71
Southern	2	8	27	73
Total	118	112	52	80

*Percentage of correctly classified fish per zone.

assigned to Bahia and misidentified fish from Bahia assigned to Ceará), the localities were pooled and considered as northern samples. Similarly, samples from Rio de Janeiro showed low levels of correct assignment but high level of cross assignment. In addition, and following Castello *et al.* (1997) who indicated that the continental shelf between 29 and 34° S corresponds to a transitional zone between neritic Patagonia and southern Brazil, samples from Santa Catarina (27° S) were pooled with samples from Rio de Janeiro and identified as the south-eastern sample. The new MDA showed an overall discrimination of 80%. (Wilk's λ *c.* 0.256; $F_{78,482}$, $P < 0.001$) (Fig. 2 and Table III).

DISCUSSION

According to Vasconcellos & Haimovici (2006), *M. furnieri* is currently heavily overfished, and the population structure is not well understood. Levy *et al.* (1998) suggested that some morphological and population dynamic characters of *M. furnieri*, between 23 and 33° S, pointed to the existence of two partially isolated populations, but allozyme analysis showed a low degree of genetic heterogeneity that did not support the hypothesis of two partially isolated populations in the area studied. The discrepancy between morphological and population dynamics analysis and the genetic analyses of the same populations (Levy *et al.*, 1998) could be explained by the Féral (2002) argument in which the number of polymorphic loci and alleles per locus is often too low to characterize all genetic patterns or to assign parentage with confidence.

According to Carozza *et al.* (2004), morphometric, morphological, genetic and reproductive studies suggested the potential existence of four population groups from the Brazilian coast to the south of Buenos Aires Province (Argentina). Recently, Vasconcellos & Haimovici (2006) suggested that morphological and life cycle characteristics, in addition to historical trends in fisheries (catch per unit effort) supported the existence of at least two stocks in southern Brazil, being Cabo de Santa Marta (29° S) the border for both stocks. The separation between the stock in southern Brazil and the stock exploited in the Common Fishing Zone of Uruguay and Argentina is less conclusive. With regard to the population structure in Argentine waters, Volpedo & Cirelli (2006) suggested the existence of two stocks, based on otolith chemistry. A northern stock associated with the fishing grounds in Samborombón Bay and Partido de la Costa (*c.* 36–37° S) in the southern area of the

Rio de la Plata and a southern one associated with El Rincón and San Blás Bay (c. 39–40° S).

Eight parasite species are characteristic of the northern localities (Ceará and Bahia), five species were found only in the area associated with south-eastern localities (Rio de Janeiro and Santa Catarina) and two species were characteristic of the southern area (Rio Grande do Sul) (Table II). Qualitative differences in metazoan parasites can provide first evidence of stock discreteness. In addition, species common to all localities (or Rio de Janeiro and Santa Catarina) showed clear quantitative differences. The larval acanthocephalan *Corynosoma australe* was found in all localities, but there were higher prevalences in the most southern locality. This agreed well with data from Braicovich & Timi (2008) who suggested that this parasite is a good tag for southern populations of the Brazilian flathead *Percophis brasiliensis* Quoy & Gaimard in the south-west Atlantic Ocean.

As suggested by Oliva & Ballón (2002), when multivariate analysis generate high levels of cross assignment between closely associated localities, those localities can be pooled and considered as a unit. The MDA successfully discriminated three groups of localities that can be associated to three stocks of *M. furnieri* on the Brazilian coast. These were a northern stock associated with Ceará and Bahia, a south-eastern stock related to Rio de Janeiro and Santa Catarina and a southern stock in the area of Rio Grande do Sul, which could be considered as the northern limit of the stock associated with the Common Fishing Zone of Uruguay and Argentina.

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