ELSEVIER

Parasite community structure within and across host populations of a marine pelagic fish: how repeatable is it?

Juan T. Timi[a,*], Robert Poulin[b]

[a]Laboratorio de Parasitología, Departamento de Biología, Facultad de Ciencias Exactas y Naturales, Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Universidad Nacional de Mar del Plata, Funes 3350 (7600) Mar del Plata, Argentina
[b]Department of Zoology, University of Otago, P.O. Box 56, Dunedin, New Zealand

Abstract

The geographical variation in parasite community structure among populations of the same host species remains one of the least understood aspects of parasite community ecology. Why are parasite communities clearly structured in some host populations, and randomly assembled in others? Here, we address this fundamental question using data on the metazoan parasite communities of different host size-classes of four distinct populations of a small pelagic fish, the Argentine anchovy, *Engraulis anchoita*, from the South West Atlantic. Within each fish sample, fish length was correlated with both the total intensity of parasites and species richness among infracommunities. More importantly, average fish length correlated with mean infracommunity richness and mean total intensity across the fish samples, indicating that the characteristics of parasite assemblages in a fish population are strongly influenced by the size of its fish in relation to those in other populations. Nested subset patterns were observed in about half of the fish samples. This means that the presence or absence of parasite species among fish individuals is often not random; however, no repeatability of nestedness among component communities was observed. Average fish length did not influence directly the likelihood that a parasite assemblage was significantly nested. However, variables influenced by average fish length, namely mean infracommunity richness and mean total intensity, determine the probability that a nested hierarchy will be observed; host size may thus indirectly affect parasite community structure either itself or via its influence on host movement and feeding patterns. To some extent, this apparent link may be due to the sensitivity of nestedness analyses to the proportion of presence in a presence/absence matrix; this in itself is a biological feature of the parasite community, however, which is associated with mean host length.
© 2003 Australian Society for Parasitology Inc. Published by Elsevier Ltd. All rights reserved.

Keywords: Nested subset analyses; *Engraulis enchoita*; Infracommunity structure; Host size

1. Introduction

Parasite communities are the product of acquisitions and losses of parasite species during the evolutionary history of their hosts (Poulin and Rohde, 1997). Probably because the number of species coexisting in a community may be the most important characteristic of that assemblage (Poulin and Rohde, 1997), during the past few decades parasitologists have attempted to identify the determinants of species richness in parasite communities, and have shown that several environmental factors and host traits are likely determinants of parasite colonisation and extinction rates (Guégan et al., 1992; Sousa, 1994; Poulin, 1995; Rohde et al., 1995).

On an ecological time scale, however, the central question remains whether parasite communities are random collections of species or structured and repetitive sets of species, i.e. whether parasite assemblages are predictable or not. This question is commonly addressed by using nested subset analyses, which are useful tools for revealing an important type of non-random community structure based on species richness (presence–absence data) (Worthen, 1996; Wright et al., 1998). However, most previous studies have focused on metazoan ectoparasite assemblages of fish (mainly monogeneans), a few studies have been carried out on endoparasitic helminths (Poulin, 1996; Rohde et al., 1998) and only one has analysed nestedness considering larval endohelminths (Poulin and Valtonen, 2001), but no study has been conducted on the whole metazoan parasite fauna of fish.

Further, the majority of earlier studies have compared several host species. This large-scale approach is necessary

---

* Corresponding author. Tel.: +54-223-4752426; fax: +54-223-4753150.
E-mail address: jtimi@mdp.edu.ar (J.T. Timi).

to identify and understand general patterns and processes, but the results can be confounded by the inherent differences among host species. Interspecific differences in host life traits such as growth, physiology, ecology and evolutionary history, do not provide a common set of extrinsic factors acting to shape the distribution of parasite species among individual hosts. In the present paper, this variability is avoided by focusing on parasite assemblages from different populations of a single host species. Thus, if differences are found they should be due to the same ecological variables acting among hosts of both different age or size and/or geographic origin, i.e. the extent to which parasite communities are structured by evolutionary events becomes negligible in relation to the influence of ecological processes.

The purpose of this study is two-fold. First, we aim to determine whether or not the organisation and species richness of parasite infracommunities can be predicted, in terms of nestedness, among four populations of a small pelagic fish, the Argentine anchovy, *Engraulis anchoita*, in the South West Atlantic. Second, we search for general patterns of nestedness by comparison across parasite assemblages of anchovies grouped by both size-classes and geographic origin of the samples, as a way to assess the repeatability of parasite community structure across comparable samples of the same host species.

2. Materials and methods

A total of 2,045 specimens of *E. anchoita* were examined for parasites. Fish samples were collected during six research cruises of R.V. Cap. Oca Balda and R.V. Dr. E. Holmberg (Instituto Nacional de Investigación y Desarrollo Pesquero, Argentina) during the spring: OB 11/93 (October 1993), OB 13/93 (November–December 1993), OB 08/94 (October 1994), H 07/95 (September 1995) and OB 14/95 (October 1995), and autumn: H 04/94 (May 1994). Pre-stratified random sampling designs and midwater trawl nets were used to collect samples. This sampling scheme covered the Southwest Atlantic shelf from 34°S to 46°S. Fish were fixed in buffered 10% formalin, measured and examined under a stereoscopic microscope. Parasites were recovered from the gills, branchial and body cavities, swim bladder and gut following standard techniques.

Based on the geographical location and season of collection, four distinct populations of fish were identified by using the parasites of adult anchovies (Total length > 140 mm) as biological tags: the autumn north Bonaerense (35–37°S) and three spring populations, north Bonaerense (34–40°S), south Bonaerense (40–43°S) and Patagonian (43–46°S) (Timi, 2003). Despite both autumn and spring samples from the north Bonaerense zone being caught in the same area, parasitological evidence indicated that autumn samples correspond to a different population, which inhabits other zones than fish caught during spring in these area (Timi, 2003).

Following Angelescu (1982), fish from each population were grouped into size-classes representing the following developmental stages: primary juveniles (36–60 mm total length), secondary juveniles (61–100 mm), pre-adults (101–120mm) and adults (>120 mm). Adults were arbitrarily subdivided into two groups based on length, adults < 150 mm (121–149 mm) and adults > 150 mm.

Following Bush et al. (1997) prevalence and mean intensity of each parasite species were calculated for each host population. In the analyses, fish size-classes were considered as separate component communities. For each host population, as well as for each component community, the following variables were calculated: (1) the number of hosts examined, (2) the mean host total length, (3) the coefficient of variability in fish length, i.e. the standard deviation divided by the mean, (4) the total number of parasite individuals, (5) the component community species richness, (6) the mean infracommunity species richness, (7) the total prevalence, i.e. the percentage of fish infected with at least one of the parasite species, (8) the total mean intensity, with all the parasite species pooled together, (9) the relationship between host length and both infracommunity richness and infracommunity total intensity. All the relationships among variables were evaluated by means of non-parametric Spearman rank correlation tests, with correction for ties.

For each component community, the index of nestedness, N, was computed (Patterson and Atmar, 1986). The index corresponds to the sum, across all parasite species, of the instances where a parasite species is absent from infra-communities richer than the most species-poor one in which it occurs. This was done with data either including or excluding uninfected fish. For each component community, the observed N value was compared with the N values of 1,000 randomly generated presence/absence matrices, produced using the algorithm RANDOM 1 of Patterson and Atmar (1986); in these Monte Carlo simulations the probability of each parasite species being included in an infracommunity was set equal to its observed prevalence in each fish sample. Nestedness was also calculated at the level of the host species by evaluating the sum, across all parasite species, of the instances where a parasite species is absent from component communities richer than the most species-poor one in which it occurs.

Due to the fact that the number of component communities included in the analyses across samples was low, when RANDOM 1 probabilities were not significantly correlated with any variable, the values of these variables were compared between nested and non-nested assemblages using Mann–Whitney U tests.

Variations in nestedness can have methodological (non-biological) explanations, for example, N depends on matrix size, which in turn is commonly correlated with matrix fill (Wright et al., 1998). The influence of both matrix size (the number of cells in the matrix, calculated as component community species richness × sample size) and matrix fill

(measured as the proportion of one or presences in a matrix) on nestedness was also evaluated by correlation analyses and Mann–Whitney *U* tests. Nestedness, when quantified as a single number from a presence–absence matrix, measures community-wide differences in the frequency of occurrence or "distribution" of species (Wright and Reeves, 1992). Total prevalence and mean total intensity underestimate and thus do not represent the actual frequency of occurrence of parasite species in each host. For this reason, three additional, more representative variables were calculated for each component community: the average of either prevalences, mean intensity or indices of aggregation (the ratio of the variance to the mean number of parasites per host) of component populations (e.g. for each parasite species).

3. Results

A total of 13 metazoan parasite species were found in the pooled samples, with an unequal distribution among the four host populations (Table 1). Two additional digenean species were also found, *Cardiocephaloides* sp. in the eyes,

and *Rhipidocotyle* sp. in the musculature, both at the metacercarial stage; these species were not included in the analyses because most fish were not searched for these two parasites.

Host body size, as well as the various community descriptors, was also variable among the host populations. A great influence of host length was observed, judging by the highly significant correlations between host length and both infracommunity species richness and mean total intensity within all host populations (Table 2).

Fish from each population were grouped into size-classes representing distinct developmental stages (Fig. 1, Table 3). The sample of primary juveniles from the autumn north Bonaerense anchovy population had only one parasite species, so it was not included in nested subset analyses; secondary juveniles from the spring Patagonian (SP) population were also excluded because of the small sample size ($n = 5$).

3.1. Effects of host length on community structure

With the exception of the component community richness, most community descriptors showed a relationship

Table 1
Taxonomic composition, prevalence (*P*) and mean intensity (MI) of metazoan parasites of *Engraulis anchoita* in four host populations

Host population	North Bonaerense				South Bonaerense		Patagonian	
	Autumn ($n = 585$)		Spring ($n = 715$)		Spring ($n = 504$)		Spring ($n = 241$)	
	P	MI ± S.D.	*P*	MI ± S.D.	*P*	MI ± S.D.	*P*	MI ± S.D.
Monogenea								
Pseudanthocotyloides heterocotyle (van Beneden, 1871) (g)	0.68	1.00 ± 0.00	16.92	1.86 ± 1.69	13.69	1.59 ± 1.29	9.13	1.50 ± 0.80
Mazocraes australis (Timi et al., 1999) (g)	0.34	1.50 ± 0.71	3.78	1.59 ± 1.29	2.58	1.31 ± 0.48	5.81	1.86 ± 2.21
Digenea								
Lecithochirium microstomum (Chandler, 1935) (sb)	42.05	2.48 ± 2.62	64.34	5.44 ± 12.09	59.13	8.91 ± 29.21	20.33	10.24 ± 17.34
Parahemiurus merus (Linton, 1910) (s)	6.84	1.42 ± 1.01	12.73	1.40 ± 0.99	3.97	1.45 ± 0.82	–	–
Cestoda								
Scolex polymorphus (Müller, 1784) plerocercoid (pc)	24.10	4.80 ± 7.43	79.16	24.40 ± 42.10	92.46	23.48 ± 25.84	69.71	15.71 ± 14.88
Pseudophyllidea plerocercoid (m)	–	–	0.14	1.00 ± 0.00	0.20	1.00 ± 0.00	–	–
Bothriocephalus sp. (pc)	–	–	–	–	0.20	3.00 ± 0.00	1.24	4.67 ± 5.51
Nematoda (third stage larvae)								
Anisakis simplex (Rudolphi, 1809; det. Krabbe, 1878) (m)	–	–	2.24	1.75 ± 1.29	4.17	1.62 ± 1.46	34.44	2.18 ± 1.73
Contracaecum sp. (m)	10.60	1.45 ± 0.74	41.68	2.65 ± 2.37	56.55	3.08 ± 2.65	68.46	3.31 ± 2.36
Hysterothylacium aduncum (Rudolphi, 1802) (m)	6.15	1.75 ± 1.20	33.01	15.95 ± 17.03	37.50	13.99 ± 31.66	70.12	33.33 ± 51.71
Terranova sp. (m)	–	–	0.84	1.00 ± 0.00	0.20	1.00 ± 0.00	–	–
Acanthocephala								
Corynosoma australe (Johnston, 1937) cystacanth (m)	0.51	1.33 ± 0.58	3.78	1.30 ± 0.87	0.59	1.33 ± 0.58	0.41	1.00 ± 0.00
Copepoda								
Nothobomolochus cresseyi (Timi and Sardella, 1997) (g)	1.20	1.00 ± 0.00	0.56	1.00 ± 0.00	1.58	1.00 ± 0.00	0.83	1.00 ± 0.00

n, Sample size; g, gills; sb, swim bladder; s, stomach; pc, pyloric caeca; m, mesenteries.

Table 2
Community descriptors of metazoan parasite assemblages of *Engraulis anchoita* from four host populations

Host population	North Bonaerense		South Bonaerense	Patagonian
	Autumn	Spring	Spring	Spring
Number of examined hosts	585	715	504	241
Total length (mm) ± S.D.	98.53 ± 25.37	128.76 ± 22.94	123.81 ± 31.34	149.80 ± 26.23
Coefficient of variation in fish length (%)	25.7	17.8	25.3	17.5
Total number of parasites	1,515	18,974	17,327	9,577
Component community richness	9	12	13	10
Mean infracommunity richness ± S.D.	0.92 ± 0.96	2.59 ± 1.41	2.73 ± 1.23	2.80 ± 1.45
Total prevalence	59.66	93.85	98.41	89.26
Total mean intensity ± S.D.	4.34 ± 5.83	28.27 ± 48.05	34.93 ± 46.12	44.34 ± 52.75
Fish length vs. infracommunity richness[a]	0.43*	0.68*	0.65*	0.69*
Fish length vs. total infracommunity intensity[a]	0.17*	0.57*	0.43*	0.75*

*$P < 0.001$.
[a] Spearman rank correlation coefficient.

with host size, increasing among larger fish in the four anchovy populations (Table 3). Similar results were found in correlation analyses between mean host body length and community descriptors across the 15 assemblages. No significant correlation was observed between mean total length and component community richness (Spearman rank correlation coefficient, $Rs = 0.38$, $n = 15$, $P = 0.16$), but mean infracommunity richness, total prevalence and mean total intensity increased significantly with host size (all $Rs > 0.67$, $n = 15$, $P < 0.01$) (Fig. 2).

Within each component community, host length was significantly correlated with infracommunity richness and total intensity in some assemblages, but with no clearly defined pattern (Table 3). The exclusion of uninfected fish changed the significance of only three correlations (infracommunity richness of secondary juveniles from both the autumn and spring north Bonaerense and the pre-adults from the Patagonian populations).
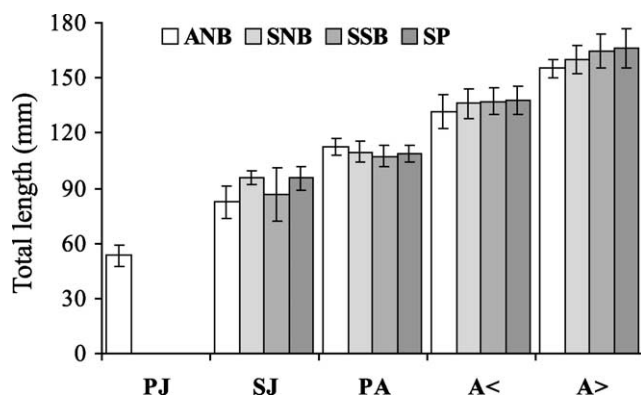

Fig. 1. Mean total length (± S.D.) of *Engraulis anchoita*, from four host populations divided in size-classes. PJ, primary juveniles; SJ, secondary juveniles; PA, pre-adults; A < , adults < 150 mm; A > , adults ≥ 150 mm; ANB, autumn north Bonaerense population; SNB, spring north Bonaerense population; SSB, spring south Bonaerense population; SP, spring Patagonian population.

3.2. Effects of sample size and host size variability on community structure

With the exception of a significant correlation between sample size and component community species richness ($Rs = 0.61$, $n = 15$, $P = 0.016$), the number of examined hosts did not relate significantly with any of the following community descriptors: mean infracommunity species richness, prevalence or mean total intensity (all $Rs < 0.24$, $n = 15$, $P > 0.39$). Neither of these variables nor component community species richness was correlated with the coefficient of variation in fish length (all $Rs < 0.23$, $n = 15$, $P > 0.40$).

The Spearman rank correlation coefficients between host length and mean infracommunity richness were not correlated with sample size ($Rs = 0.22$, $n = 15$, $P = 0.44$) nor with the coefficient of variability in fish length ($Rs = -0.20$, $n = 15$, $P = 0.47$). Similar results were observed for the coefficients between host length and mean total intensity in relation to both sample size ($Rs = 0.02$, $n = 15$, $P = 0.95$) and coefficient of variability ($Rs = 0.11$, $n = 15$, $P = 0.68$).

3.3. Nested subset analyses

Infracommunities in seven of the 15 component communities were nested with a probability level of $P < 0.05$, and four of these with $P < 0.01$. The exclusion of uninfected fish from all the matrices yielded the same values of N but with slight reductions in some RANDOM 1 probabilities (Table 4), with no changes in the significance of nestedness. No anti-nestedness cases were found (all $P < 0.65$) (Table 4).

All component communities from the autumn north Bonaerense population were non-nested, whereas among the spring samples, all adult ≥150 mm fish groups had significantly nested parasite assemblages. Nestedness among other size-classes was variable among host

Table 3
Community descriptors of metazoan parasite assemblages of *Engraulis anchoita* from four host populations, divided in size-classes (component communities)

Host population/size-class	No. examined	No. infected	CV[a]	Component community richness	Mean infracommunity richness ± S.D.	Total prevalence	Mean total intensity ± S.D.	Fish length vs. richness[b]	Fish length vs. intensity[b]
Autumn north Bonaerense									
Primary juveniles	30	1	11.0	1	0.03 ± 0.18	3.33	1	–	–
Secondary juveniles	308	155	10.7	8	0.67 ± 0.77	50.32	3.70 ± 5.73	0.24**	0.16*
Pre-adults	118	92	4.1	7	1.24 ± 0.92	77.97	4.52 ± 5.99	0.15	−0.04
Adults < 150 mm	114	89	6.7	9	1.39 ± 1.08	78.07	5.12 ± 5.93	0.07	0.08
Adults ≥ 150 mm	15	12	3.1	4	1.87 ± 1.50	80.00	5.67 ± 5.16	0.13	0.04
Spring north Bonaerense									
Secondary juveniles	90	73	3.8	7	1.19 ± 0.95	80.00	6.51 ± 14.49	0.26*	0.22
Pre-adults	207	181	5.2	11	1.73 ± 1.03	87.44	15.70 ± 39.19	0.34**	0.41**
Adults < 150 mm	257	257	6.0	11	3.10 ± 1.10	100	32.10 ± 35.20	0.30**	0.01
Adults ≥ 150 mm	161	161	4.8	10	3.66 ± 1.17	100	46.05 ± 71.64	0.15*	0.31**
Spring south Bonaerense									
Secondary juveniles	131	124	17.0	6	1.88 ± 0.87	94.66	19.67 ± 18.11	0.09	−0.31**
Pre-adults	118	117	5.3	9	2.05 ± 0.98	99.15	20.49 ± 25.71	0.30**	0.31**
Adults < 150 mm	130	130	5.3	10	3.16 ± 0.95	100	41.02 ± 58.77	0.20*	−0.15
Adults ≥ 150 mm	125	125	5.6	11	3.80 ± 0.97	100	57.26 ± 54.61	0.14	0.28**
Spring Patagonian									
Secondary juveniles	5	2	6.8	3	0.80 ± 1.09	40.00	3.50 ± 2.12	–	–
Pre-adults	48	26	4.3	8	0.77 ± 0.83	54.17	5.08 ± 11.27	0.25	0.54**
Adults < 150 mm	31	31	5.4	7	2.58 ± 0.88	100	25.81 ± 27.47	0.33	0.09
Adults ≥ 150 mm	157	157	6.5	10	3.53 ± 0.96	100	55.02 ± 56.63	0.71**	0.24**

*$P < 0.05$, **$P < 0.01$.
[a] Coefficient of variation in fish length (%).
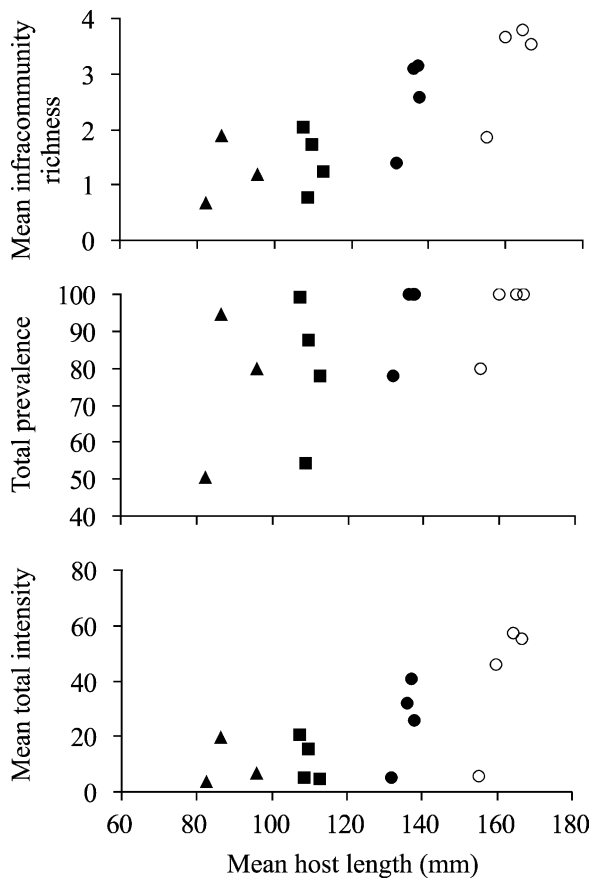[b] Spearman rank correlation coefficient.

Fig. 2. Relationships between mean length of *Engraulis anchoita* and mean infracommunity richness, total prevalence or mean total intensity, across 15 assemblages of metazoan parasites. Triangles: secondary juveniles; squares: pre-adults; black circles: adults < 150 mm; white circles: adults ≥ 150 mm.

populations (Table 4), being more common in both north and south spring Bonaerense populations.

RANDOM 1 probabilities were not significantly correlated with sample size (Rs = −0.43, $n = 15$, $P = 0.11$) nor with the coefficient of variation in fish length within each sample (Rs = −0.38, $n = 15$, $P = 0.16$); however, although no differences between nested and non-nested communities were observed in terms of fish length variability (two-tailed Mann–Whitney U test, $U = 17.00$, $P = 0.23$), nested assemblages were detected in larger samples than non-nested ones (Mann–Whitney U test, $U = 9.50$, $P = 0.029$). Similarly to sample size, the matrix size was not significantly correlated with RANDOM 1 probabilities (Rs = −0.47, $n = 15$, $P = 0.08$), but matrix sizes of nested assemblages were significantly higher than those of non-nested ones (Mann–Whitney U test, $U = 10$, $P = 0.037$). To avoid the effect of matrix size on nestedness, the sample of adults > 150 mm from the autumn north Bonaerense population, which has the lowest sample size ($n = 15$), was excluded from subsequent analyses. After excluding this sample no differences in sample size (two-tailed Mann–Whitney U test, $U = 9.5$, $P = 0.053$) nor matrix size (two-tailed Mann–Whitney U test, $U = 10$,

$P = 0.073$) were found between nested and non-nested communities.

No significant correlations were observed between RANDOM 1 probabilities and both mean host length (Rs = −0.36, $n = 14$, $P = 0.20$) and component community richness (Rs = −0.49, $n = 14$, $P = 0.78$), also no differences between nested and non-nested assemblages were found for both mean host length (two-tailed Mann–Whitney U test, $U = 17.0$, $P = 0.38$) and component community richness (two-tailed Mann–Whitney U test, $U = 9.5$, $P = 0.053$). On the other hand RANDOM 1 probabilities were significantly correlated with mean infracommunity species richness (Rs = −0.77, $n = 14$, $P < 0.01$), total prevalence (Rs = −0.73, $n = 14$, $P < 0.001$) or mean total intensity (Rs = −0.75, $n = 14$, $P < 0.01$). This indicates that an increase in either species richness, prevalence and number of parasites, increases the probability that a nested hierarchy will be observed. This is not surprising since most of these variables were significantly and positively correlated with each other (mean infracommunity richness vs. total prevalence, Rs = 0.95, $P < 0.01$; mean infracommunity richness vs. mean total intensity, Rs = 0.97, $P < 0.01$ and total prevalence vs. mean total intensity, Rs = 0.96, $P < 0.01$).

The matrix fill was significantly correlated with RANDOM 1 probabilities (Rs = −0.59, $n = 14$, $P = 0.026$) and also with either mean host length, mean infracommunity richness, total prevalence and mean total intensity (all Rs > 74, $n = 14$, $P < 0.01$).

The average of either prevalences, mean intensity or the indices of aggregation of component populations proved to be correlated significantly with matrix fill (all Rs > 0.63, $n = 14$, $P < 0.01$). RANDOM 1 probabilities were also significantly correlated with both averaged prevalence and averaged mean intensity (Rs = −0.58, $n = 14$, $P = 0.026$ and Rs = −0.62, $n = 14$, $P = 0.018$, respectively), whereas no relationship was observed with averaged aggregation (Rs = −0.42, $n = 14$, $P = 0.14$).

Considering the whole sample, the component communities (e.g. all the 16 size-classes from the four host populations, including primary juveniles from the autumn north Bonaerense population) were nested ($N = 23$) with a probability level of $P = 0.001$. In other words, parasite species were not randomly distributed among the 16 host samples, but instead they occurred as a nested subset pattern.

4. Discussion

Mean infracommunity richness, total prevalence and mean total intensity increased with host size, within each component community and across assemblages. This is a common feature of fish–parasite systems, due to larger fish having higher feeding rates and/or larger surfaces for parasite attachment, as well as having been exposed to

Table 4
Within component community (host size-classes) nestedness analyses of metazoan parasites of *Engraulis anchoita* from four host populations, and data of matrix size and fill

Host population/size-class	RANDOM 1 probability (observed N)	RANDOM 1 probability[a] (observed N)	Matrix size	Proportion of 1	Averaged prevalence ± S.D.	Averaged mean intensity ± S.D.	Averaged coefficient of aggregation ± S.D.
Autumn north Bonaerense							
Secondary juveniles	0.651 (157)	0.414 (157)	2,464	0.084	8.40 ± 14.22	1.76 ± 1.35	3.45 ± 5.88
Pre-adults	0.257 (129)	0.280 (129)	826	0.177	17.67 ± 18.82	2.01 ± 1.36	3.78 ± 4.79
Adults < 150 mm	0.188 (183)	0.205 (183)	1,026	0.155	15.50 ± 16.94	1.87 ± 1.36	3.02 ± 3.74
Adults ≥ 150 mm	0.544 (5)	0.544 (5)	60	0.467	46.67 ± 9.43	2.42 ± 0.14	2.30 ± 0.55
Spring north Bonaerense							
Secondary juveniles	0.244 (81)	0.242 (81)	630	0.170	16.98 ± 20.41	2.56 ± 2.92	7.38 ± 15.31
Pre-adults	0.019 (563)	0.032 (563)	2,277	0.157	15.72 ± 24.36	2.83 ± 4.36	14.83 ± 31.84
Adults < 150 mm	0 (908)	0 (908)	2,827	0.282	28.23 ± 33.26	4.28 ± 7.42	7.22 ± 12.21
Adults ≥ 150 mm	0.006 (563)	0.006 (563)	1,610	0.366	36.65 ± 35.02	6.19 ± 9.94	20.95 ± 34.81
Spring south Bonaerense							
Secondary juveniles	0 (97)	0 (97)	786	0.314	31.42 ± 35.47	4.50 ± 6.25	5.43 ± 6.39
Pre-adults	0.014 (200)	0.007 (200)	1,062	0.228	22.79 ± 31.38	3.66 ± 5.24	6.83 ± 11.09
Adults < 150 mm	0.090 (413)	0.090 (413)	1,300	0.316	31.61 ± 36.51	5.80 ± 9.16	21.51 ± 53.33
Adults ≥ 150 mm	0 (290)	0 (290)	1,375	0.345	34.54 ± 40.14	6.75 ± 10.20	12.37 ± 22.23
Spring Patagonian							
Pre-adults	0.427 (35)	0.397 (35)	384	0.096	8.85 ± 8.39	2.57 ± 3.34	5.54 ± 11.52
Adults < 150 mm	0.151 (49)	0.151 (49)	217	0.369	36.87 ± 28.42	6.93 ± 9.37	9.32 ± 14.24
Adults ≥ 150 mm	0.036 (415)	0.036 (415)	1,570	0.353	35.35 ± 34.49	7.88 ± 11.76	14.78 ± 24.30

[a] Calculated on infected fish only.

parasites for longer (Price and Clancy, 1983; Guégan et al., 1992; Poulin, 2000). Larger hosts are also expected to harbour richer parasite faunas because they provide a greater variety of niches and can sustain a greater absolute number of parasites (Morand and Poulin, 1998). Neither the number of examined fish per sample nor variability in host length influenced most community descriptors in the analyses across assemblages. Their relationship with component community species richness could be due to the latter being highly influenced by rare species, which are more likely to be found in larger samples. This effect, however, is diluted at the infracommunity level; thus no confounding influences of sampling effort are acting on our analyses.

In one of the most extensive studies to date, Rohde et al. (1998) showed that fewer than one-third of parasite assemblages studied were nested, and concluded that parasite assemblages in marine fish were commonly unstructured and unpredictable, at least in terms of hierarchical species assembly rules. Rohde et al. (1998) defended their conclusion by arguing that other authors found nested patterns in fish–parasite assemblages only because they used fish samples including juvenile and adult hosts together. The examination of developing parasite communities is likely to generate apparent nestedness because of differential colonisation probabilities among parasites. This is confirmed in the present paper, however, the tendency for fish–parasite communities to be unstructured seems to depend on the scale of the study: if pooling juvenile and adult fish in an analysis yields nestedness, there is a predictable structure of the parasite community within a real host population, despite no structure being found in some portions (host age groups) of this assemblage.

The findings of the present paper agree with those of Poulin and Valtonen (2001) in that when larger fish tend to harbour richer infracommunities, the infracommunities tend to be nested. Therefore, community structure is a consequence of the way in which hosts accumulate parasite species rather than of ecological processes acting within assemblages, and largely depends on the host size/age structure of the samples. The local environmental conditions and geographic distribution patterns of both parasites and other hosts involved in their life cycle, as well as the occurrence of host habitat shifts, can reinforce the effects of host size or lead to nestedness.

The rarity of nestedness in parasite communities was supported by Poulin and Guégan (2000) but these authors, by pooling the proportion of both nested and anti-nested assemblages, showed that the commonness of non-random structural patterns reached more than a half of the communities analysed. In the present paper infracommunities in seven of the 15 component communities were significantly nested. Earlier workers have often used Bonferroni corrections to adjust RANDOM 1 probabilities, but given the independence of our sample, we chose not to do this (see Moran, 2003).

Although a high proportion of the parasite infracommunities of *E. anchoita* showed a non-random structure, no repeatability of nestedness among component communities was observed. Theoretically, nested patterns of species composition can be generated by either one or both of the main processes governing community dynamics: local extinction of species and colonisation (Patterson, 1990).

Differential extinction, as a result of interspecific competition, is rare in host–parasite systems (Rohde et al., 1995; Poulin, 1996). Density-dependent extinction rates cannot be a cause of nestedness because the time scale for structuring parasite infracommunities is the life span of the host, therefore extinction of parasite communities has to be considered at the level of the component population or suprapopulation (sensu Bush et al., 1997), in which the loss of a parasite species in an individual host is compensated by the immigration of new parasites (Guégán and Hugueny, 1994). Because of the dispersal requirements of most parasite species, eggs or larvae leave the host, and thus the density of parasite infrapopulations depends on the immigration and persistence of parasite individuals, especially for intermediate hosts harbouring long-lived larval stages of parasites, such as those in the present study.

Among the colonisation-dominated communities analysed here, nestedness was common in spring samples, but autumn assemblages showed no departures from randomness. Autumn samples showed the lowest values of prevalence and mean intensity (total and for each parasite species) in comparison with spring ones (Tables 1 and 2); although autumn samples contained smaller fish on an average than those in spring samples (Fig. 1), latitudinal differences proved more important than host size in determining differences among parasite assemblages of *E. anchoita* (see Timi, 2003). Rohde et al. (1998) showed that poor assemblages of parasites have a low probability of being nested, prevalence being the best predictor of nestedness in their study. Both lower prevalences and intensities are also characteristic features of other non-nested parasite communities (Poulin and Guégan, 2000; Poulin and Valtonen, 2001).

Among spring samples from three host populations, nestedness was observed in those component communities with the highest values of total prevalence, mean total intensity and mean infracommunity richness. Nestedness also seems to be highly related to host size; in fact most nested assemblages showed significant relationships between host length and both richness and total intensity. All assemblages of adult fish ≥150 mm were significantly nested, showing the highest values of total prevalence, mean total intensity and mean infracommunity richness. Matrix fill was also associated with the probability of observing a nested pattern. However, as this variable is simply a reflection of real biological parameters (prevalences of the different parasite species and infracommunity richness values), this association merely confirms that nestedness is more likely to occur when prevalence, intensity and

infracommunity richness values are high. However, despite the similar coefficients of variability in fish length with respect to other size-classes, nestedness could be related to variability in host age. Growth studies of *E. anchoita* have shown that whereas secondary juveniles are 0–1 years old and pre-adults are 1–2 years old, adult fish larger than 120 mm can range from 1 to 6 years old (Hansen and Madirolas, 1996), therefore the age variability in samples of adult fish is not well represented by size variability.

Engraulis anchoita is a planktivorous species (Angelescu, 1982) that feeds mainly by filtering, but also by actively catching some larger prey; prey that are actively caught by smaller fish are retained by filtering in older anchovies (Angelescu, 1982). Although the addition of "new" prey (intermediate hosts of "new" parasites) results from fish growth, large fish still feed on the same zooplankters as juveniles, resulting in a stable composition of diet items through the fish ontogeny. This could be linked with a sequential enrichment of parasite species with host age, and therefore with host size-dependent nested subset patterns.

As postulated by Worthen and Rohde (1996), Rohde et al. (1998) and Poulin and Guégan (2000), it is concluded that differences in colonisation rates among parasite species might be the cause of nestedness in parasite assemblages of marine fish. However, when analysing a single host species, capable of harbouring a fixed set of parasite species, each one with characteristic transmission rates, if differences are observed among host groups in terms of nestedness, other variables must also be acting.

The distribution patterns of marine parasites are determined mainly by temperature–salinity profiles and their association with specific masses of water (Esch and Fernández, 1993). As with distribution, transmission of parasites is potentially affected by abiotic conditions both directly or indirectly, mainly via effects on intermediate host populations (Janovy et al., 1997). Thus the causes of differences in nestedness patterns among anchovy populations should be related to the predominant environmental conditions in the regions inhabited by each host population. This is supported by the finding that differences in parasite assemblages between autumn and spring samples from the same area are not due to seasonality (Timi, 2003).

The distribution of zooplankton in the Argentine sea may be assigned to three water masses: coastal to the north to Península Valdes, middle shelf and outer shelf, the outer shelf influenced by the Falklands (Malvinas) current of subantarctic waters. Each water mass has its characteristic composition of plankton species (Ramírez et al., 1989). This distribution pattern results in anchovies from each zone having different diets (in quantity and composition), which is reflected in variations in diet specificity and increase in prey size (mainly due to the presence of euphausiids and hyperiids) as both latitude and distance from the coast increase (Angelescu, 1982).

The latitudinal gradient of environmental conditions and its influence on the distribution of both zooplankton and definitive hosts could therefore be the main determinant factor of the differences in the parasite assemblages of *E. anchoita* among host populations. These differences are probably reinforced by the migratory behaviour of the fish. Whereas Patagonic anchovies live in a region influenced by subantarctic waters throughout the year, the Bonaerense populations develop an annual migratory cycle between coastal waters during spring–summer for reproductive purposes, and off-shore subantarctic waters, from late summer through winter for feeding (Angelescu, 1982). Anchovies start migrating when they reach the pre-adult stage, and visit the subantarctic waters once a year, where they feed on a different planktonic community, resulting in a sequential enrichment of parasite species with host age. In other words, anchovies go through both a habitat and diet shift once per year, enhancing the probability that age-dependent nested subset patterns develop; this could explain the higher proportion of nested assemblages in both Bonaerense populations.

Correlation analyses and comparisons across assemblages also allowed some general patterns to be recognised. Whereas an increase in host length variability does not significantly bias nestedness values across assemblages, the influence of the number of examined fish on nestedness needs to be considered. After removing the smallest sample, no significant differences between nested and non-nested communities were found; however, after excluding uninfected fish, which reduced markedly the matrix size, no changes in the significance of nestedness were observed. Therefore if an artefactual element of sampling effort or matrix size exists, it is small. Surprisingly, neither correlations nor differences of host size in relation to nestedness were found, despite the influence of this variable on parasite community structure. This lack of relationship could be due to the obscuring effect of the local conditions acting on the different host populations. However, the probability of finding nestedness increased with total prevalence, total mean intensity and mean infracommunity richness across assemblages of a single species, confirming previous findings (Rohde et al., 1998; Poulin and Guégan, 2000; Poulin and Valtonen, 2001). Most of these variables were positively correlated with each other because assemblages characterised by a high number of parasite individuals per infracommunity are expected to have a high proportion of fish harbouring infracommunities (Poulin and Guégan, 2000).

Despite the fact that we found no repeatability of parasite community structure across samples of the same host species, one of our main results was that parasite species of the 16 component communities from the four host populations occurred as a nested subset pattern at the largest scale in our study. In other words, species in depauperate component communities, irrespective of host size and geographical origin, were non-random subsets of

the species that occurred in progressively richer assemblages, indicating that there are some general, large-scale processes structuring parasite communities at the level of host species. There is therefore a need for more studies of nestedness at this spatial level to reveal how common these patterns are and also what processes govern their dynamics.

Acknowledgements

References

Angelescu, V., 1982. Ecología trófica de la anchoíta del Mar Argentino (Engraulidae, *Engraulis anchoita*). Parte II. Alimentación, comportamiento y relaciones tróficas en el ecosistema. Contrib. Inst. Nac. Invest. Des. Pesq., Mar del Plata. No. 409.

Bush, A.O., Lafferty, K.D., Lotz, J.M., Shostak, A.W., 1997. Parasitology meets ecology on its own terms: Margolis et al. revisited. J. Parasitol. 83, 575–583.

Esch, G.W., Fernández, J.C., 1993. A Functional Biology of Parasitism. Chapman & Hall, London.

Guegán, J.-F., Hugueny, B., 1994. A nested species subset pattern in tropical fish: host as major determinant of parasite infracommunity structure. Oecologia 100, 184–189.

Guégan, J.-F., Lambert, A., Lévêque, C., Combes, C., Euzet, L., 1992. Can host body size explain the parasite species richness in tropical freshwater fishes? Oecologia 90, 197–204.

Hansen, J.E., Madirolas, A., 1996. Distribución, evaluación acústica y estructura poblacional de la anchoíta, resultados de la campaña del año 1993. Rev. Invest. Des. Pesq. 10, 5–21.

Janovy, J. Jr., Snyder, S.D., Clopton, R.E., 1997. Evolutionary constraints on population structure: the parasites of *Fundulus zebrinus* (Pisces: Cyprinodontidae) in the South Platte River of Nebraska. J. Parasitol. 83, 584–592.

Moran, M.D., 2003. Arguments for rejecting the sequential Bonferroni in ecological studies. Oikos 100, 403–405.

Morand, S., Poulin, R., 1998. Density, body mass and parasite species richness of terrestrial mammals. Evol. Ecol. 12, 717–727.

Patterson, B.D., 1990. On the temporal development of nested subset patterns of species composition. Oikos 59, 330–342.

Patterson, B.D., Atmar, W., 1986. Nested subset and the structure of insular mammalian faunas and archipelagos. Biol. J. Linn. Soc. 28, 65–82.

Poulin, R., 1995. Phylogeny, ecology, and the richness of parasite communities in vertebrates. Ecol. Monogr. 63, 283–302.

Poulin, R., 1996. Richness, nestedness, and randomness in parasite infracommunity structure. Oecologia 105, 545–551.

Poulin, R., 2000. Variation in the intraspecific relationship between fish length and intensity of parasitic infection: biological and statistical causes. J. Fish Biol. 56, 123–137.

Poulin, R., Guégan, J.-F., 2000. Nestedness, anti-nestedness, and the relationship between prevalence and intensity in ectoparasite assemblages of marine fish: a spatial model of species coexistence. Int. J. Parasitol. 30, 1147–1152.

Poulin, R., Rohde, K., 1997. Comparing the richness of metazoan ectoparasite communities of marine fishes: controlling for host phylogeny. Oecologia 110, 278–283.

Poulin, R., Valtonen, E.T., 2001. Nested assemblages resulting from host size variation: the case of endoparasite communities in fish hosts. Int. J. Parasitol. 31, 1194–1204.

Price, P.W., Clancy, K.M., 1983. Patterns in number of helminth parasite species in freshwater fishes. J. Parasitol. 69, 449–454.

Ramírez, F.C., Mianzán, H.W., Santos, B., Viñas, M.D., 1989. Synopsis on the reproductive biology and early life history of *Engraulis anchoita*, and related environmental conditions in Argentine waters. Zooplankton. In: Second IOC Workshop on Sardine/Anchovy Recruitment Project (SARP) in the Southwest Atlantic. Montevideo, Uruguay. Workshop Report No. 65. Annex V. pp. 4–6.

Rohde, K., Hayward, C.J., Heap, M., 1995. Aspects of the ecology of metazoan ectoparasites of fishes. Int. J. Parasitol. 25, 945–970.

Rohde, K., Worthen, W.B., Heap, M., Hugueny, B., Guégan, J.-F., 1998. Nestedness in assemblages of metazoan ecto- and endoparasites of marine fish. Int. J. Parasitol. 28, 543–549.

Sousa, W.P., 1994. Patterns and processes in communities of helminth parasites. Trends Ecol. Evol. 9, 52–57.

Timi, J.T. Parasites of Argentine anchovy in the Southwest Atlantic: latitudinal patterns and their use for discrimination of host populations. J. Fish Biol., 63, 90–107.

Worthen, W.B., 1996. Community compòsition and nested-subset analyses: basic descriptors for community ecology. Oikos 76, 417–426.

Worthen, W.B., Rohde, K., 1996. Nested subset analyses of colonization-dominated communities: metazoan ectoparasites of marine fishes. Oikos 75, 471–478.

Wright, D.H., Reeves, J.H., 1992. On the meaning and measurement of nestedness of species assemblages. Oecologia 92, 416–428.

Wright, D.H., Patterson, B.D., Mikkelson, G.M., Cutler, A., Atmar, W., 1998. A comparative analysis of nested subset patterns of species composition. Oecologia 113, 1–20.