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Nested assemblages resulting from host size variation: the case of endoparasite communities in fish hosts

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Abstract

Nested species subsets are a common pattern in many types of communities found in insular or fragmented habitats. Nestedness occurs in some communities of ectoparasites of fish, as does the exact opposite departure from random assembly, anti-nestedness. Here, we looked for nested and anti-nested patterns in the species composition of communities of internal parasites of 23 fish populations from two localities in Finland. We also compared various community parameters of nested and anti-nested assemblages of parasites, and determined whether nestedness may result simply from a size-related accumulation of parasite species by feeding fish hosts. Nested parasite communities were characterised by higher prevalence (proportion of infected fish) and intensities of infection (number of parasites per fish) than anti-nested communities; the two types of non-random communities did not differ with respect to parasite species richness, however. In addition, the correlation between fish size and the number of parasite species harboured by individual fish was much stronger in nested assemblages than in anti-nested ones, where it was often nil. These results were shown not to be artefacts of sampling effort or host phylogeny. They apply to both assemblages of adult and larval parasites, which were treated separately. Since species of larval parasites are extremely unlikely to interact with one another in fish hosts, the establishment of nestedness appears independent of the potential action of interspecific interactions. The species composition of these parasite communities is not determined from within the community, but rather by the extrinsic influence of host feeding rates and how they amplify differences among parasite species in probabilities of colonisation or extinction. Nested patterns occur in parasite communities whose fish hosts accumulate parasites in a predictable fashion proportional to their size, whereas anti-nested communities occur in parasite communities whose fish hosts do not, possibly because of dietary specialisation preventing them from sampling the entire pool of parasite species available locally. Thus, nestedness in parasite communities may result from processes somewhat different from those generating nested patterns in free-living communities.

Keywords: Comparative analysis; Infracommunity structure; Null models; Phylogeny

1. Introduction

Null models are used in ecology to derive statistically random patterns that serve as null hypotheses against which patterns observed in nature can be compared. The use of null models to detect species assembly rules is currently at the core of community ecology (Gotelli and Graves, 1996; Weiher and Keddy, 1995, 1999). Because these rules are seen as statistical patterns that emerge from the background noise, they are easier to distinguish when replicate communities are available. For this reason, parasite communities represent ideal models to study assembly rules. Each host individual in a population harbours a replicate infracommunity (sensu Bush et al., 1997) of parasite species, drawn from the larger pool of species forming the component parasite community, i.e. all parasite species found in a host population. The ease with which several infracommunities can be sampled down to the last individual parasite allows strong statistical tests to be used in the search for assembly rules. In recent years, one of the most popular approaches to the study of parasite community structure has involved the search for a nested pattern in infracommunity composition within the parasite component community (Guégan and Hugueny, 1994; Poulin, 1996; Worthen and Rohde, 1996; Rohde et al., 1998). Nestedness is one of many possible departures from a random assembly of species that was first applied to the study of insular or fragmented communities of free-living organisms (Patterson and Atmar, 1986; Wright and Reeves, 1992; Worthen, 1996; Wright et al., 1998). In the context of parasite communities, a nested pattern occurs when the parasite

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species found in depauperate infracommunities represent non-random subsets of progressively richer ones; it implies that the distributions of different parasite species among host individuals are not mutually independent. Significant nestedness has been observed in ectoparasite communities of fish, though it does not appear to be a very common pattern when using the rather conservative Bonferroni correction (Guégan and Hugueny, 1994; Worthen and Rohde, 1996; Rohde et al., 1998).

Despite its apparent usefulness as a means of detecting non-random structure in the species composition of parasite communities, our understanding of nestedness is far from complete. First, most comprehensive studies to date have focused on metazoan ectoparasite assemblages of fish (Worthen and Rohde, 1996; Rohde et al., 1998). Nestedness analyses have only been performed for very few communities of endoparasitic helminths, from two mammalian host populations (Poulin, 1996) and a handful of fish host populations (Rohde et al., 1998). It is therefore not clear whether nested patterns are frequent in types of communities other than those found on the external surfaces of fish, where interspecific interactions are notably weak (Rohde, 1991). Second, we know very little about the mechanisms that may generate nested patterns in parasite communities. In communities of free-living animals, differences among species in probability of local extinction or colonisation are often invoked as processes generating nestedness among insular communities (Patterson and Atmar, 1986; Patterson, 1990; Bolger et al., 1991). The same mechanisms could operate in parasite communities, with the probability of local extinction corresponding to the exclusion of a parasite species from a host. This does not necessarily need to involve interspecific interactions among the parasite species. The presence of nestedness, while indicating that parasite species do not occur at random among infracommunities (i.e. among host individuals in a population), could even be promoted by extrinsic factors. For instance, Guégan and Hugueny (1994) found that the fish size correlated with ectoparasite species richness (i.e. number of parasite species per host) along a sequence of nested infracommunities. Although their analysis has been criticised (Worthen and Rohde, 1996), nestedness might be driven by host biology as well as by interactions from within the infracommunities. Host size, via feeding rates, could amplify intrinsic differences among parasite species in probabilities of colonisation or extinction. One way to address this issue could be to compare nestedness patterns between communities of adult endohelminths living in the host's gut, and communities of larval helminths living in other host tissues. Interspecific interactions among parasites are likely to occur in the former type of communities, because their members exploit similar resources in the same general habitat, but are unlikely in the latter type of communities, which consist of species often encysted and unlikely to compete with one another for host resources.

Recently, Poulin and Guégan (2000) exposed another departure from random species assembly in fish ectoparasite communities, one that is the exact opposite of nestedness. They labelled it anti-nestedness; it consists of parasite species that are, more often than not, absent from infracommunities richer than the most species-poor one in which they occur (see Fig. 1 in Poulin and Guégan, 2000). This definition is formulated as the opposite of nestedness, but anti-nestedness could be defined more simply as a situation in which parasite species are more dispersed than random among infracommunities. Poulin and Guégan (2000) observed consistent differences between nested and anti-nested parasite communities: nested communities are characterised by overall parasite prevalence and mean total intensity of infection that are significantly higher than in anti-nested communities. No differences in species richness were observed between nested and anti-nested assemblages. These findings suggest that there may exist a continuum from anti-nested to nested communities, depending on the commonness and abundance of available species (Poulin and Guégan, 2000). If indeed the pattern of species occurrence among host individuals is dependent on basic parameters of the parasite component community, then species interactions may play a role in creating nested patterns.

In this paper, we search for the occurrence of nested patterns among communities of both adult endoparasites and larval endoparasites, from a large number of fish populations (i.e. a large number of independent parasite component communities). Our objectives are three-fold. First, we present the first application of nestedness analyses to a large sample of communities of internal parasites of vertebrates. Second, following Poulin and Guégan (2000), we look for a continuum from anti-nested to nested communities, and for the same differences in overall prevalence and mean intensity of infection between the two extreme types of communities. Third, we explore the general mechanism(s) responsible for the establishment of nestedness in parasite community structure. The use of assemblages of both adult and larval endohelminths is particularly relevant for this last objective: consistent patterns emerging from both types of communities are unlikely to be due to the effects of interspecific interactions, because the latter are extremely unlikely in communities of larval endohelminths.

2. Methods

2.1. Fish collection and nestedness analyses

Data on parasite communities were obtained by one of us (E.T.V.) in two localities, the northeastern Bothnian Bay of the Baltic Sea and Lake Yli-Kitka (NE Finland) in 1978–1979. Fish samples were obtained monthly or bimonthly from the catches of commercial fishermen at both localities. Each fish was measured (total length). They were then dissected and examined for internal metazoan parasites following standard techniques. All parasite individuals found in each fish were identified to the species level and counted. Adult helminths found in the gut, and larval helminths found in the body cavity or other organs, were tabulated separately. Diplostomid metacercariae were excluded from the analyses, because all of them could not be identified to the species level. The list of helminth species found in fish from the Bothnian Bay is given in Valtonen et al. (2001); those from Lake Yli-Kitka are available from the authors.

We included only component communities consisting of at least three different parasite species, since nestedness is meaningless for communities of one or two species. For each component community, we computed the index of nestedness, *N*, first proposed by Patterson and Atmar (1986). This index corresponds to the sum, across all parasite species, of the instances where a parasite species is absent from infracommunities richer than the most species-poor one in which it occurs. This was done using only data from fish with at least one parasite (i.e. excluding uninfected fish from the computations). For each component community, the observed *N* value was compared with the *N* values of 1000 randomly generated presence/absence matrices, produced using the algorithm RANDOM1 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 the fish sample of the host species being considered. The proportion of simulated *N* values that were lower than or equal to the observed *N* value gave the RANDOM1 *P*-value, which was used as a measure of departure from the structure expected from random assembly (Guégan and Hugueny, 1994; Hugueny and Guégan, 1997). When the RANDOM1 *P*-value is ≤ 0.05, the infracommunities are significantly nested; when the *P*-value is ≥ 0.95, they show a significant anti-nested pattern. In other words, as the *P*-value increases, the structure of the infracommunities becomes less and less nested and increasingly anti-nested.

2.2. Further statistical analyses

For each component community, we also computed and recorded (i) the number of fish examined; (ii) the species richness of the component community; (iii) the mean infracommunity species richness; (iv) prevalence, or the percentage of infected fish in the total sample; (v) the mean total intensity of infection, with all parasite species pooled together; (vi) the coefficient of variation of fish length among the individual infected fish in the sample, i.e. the standard deviation divided by the mean, as a measure of the relative variability in fish sizes; and (vii) the Spearman rank correlation coefficient, and its associated *P*-value (corrected for ties), between infracommunity richness and fish length among infected fish of the same population.

The RANDOM1 algorithm may overestimate the occurrence of nested or anti-nested patterns, and the use of Bonferroni corrections has been recommended for the assessment of nestedness across several communities (Worthen and Rohde, 1996). We therefore used Bonferroni corrections when determining the frequency of nestedness or anti-nestedness across all communities. However, in subsequent analyses, we used less conservative procedures for two reasons. The first is that the Bonferroni approach has been criticised by statisticians in recent years, because it often leads to the incorrect acceptance of the null hypothesis when multiple comparisons are in fact independent of one another (Rothman, 1990; Perneger, 1998). The second reason is that we believe the RAMDOM1 *P*-values most likely form a continuum, and we considered them all rather than only focusing on extreme values.

We analysed the data in two ways. First, we compared nested and anti-nested assemblages with respect to the seven parameters described above. Assemblages were classified as tending-toward-nested if their RANDOM1 *P*-value was ≤ 0.15, and as tending-toward-anti-nested if it was ≥ 0.85. We used these less rigorous cut-off points because of the small number of communities in our analyses. Comparisons were performed using two-tailed *t*-tests on log-transformed data. We also checked for associations among the different continuous variables using product-moment correlation coefficients computed on log-transformed data; the number of fish examined, in particular, may act as a confounding variable (Gregory and Blackburn, 1991).

Second, we performed a parallel series of analyses aimed at verifying whether the phylogenetic relationships of the host species influenced the results of the above tests. Associations between the above variables and the RANDOM1 *P*-values were determined in a comparative analysis across component communities, i.e. across fish populations and species. We controlled for the potentially confounding influence of fish phylogeny by using the phylogenetically independent contrasts method (Felsenstein, 1985), the most widely used and reliable method in comparative biology (Diaz-Uriarte and Garland, 1998; Ackerly, 2000; Oakley and Cunningham, 2000). This is an important step in the analysis, because if some taxonomically constrained trait of fish hosts promotes the establishment of nested patterns, then the different host species would not represent statistically independent points in the analyses. The method simply consists in deriving phylogenetic contrasts (i.e. differences between the species values) among all sister clades in a reconstructed fish phylogeny, and using them in standard parametric analyses. The contrasts were obtained from log-transformed data using the CAIC, version 2.0, computer package (Purvis and Rambaut, 1994). Here, RANDOM1 *P*-values were treated as a continuous variable. Contrasts between the values of sister taxa were computed such that contrasts in RANDOM1 probabilities were always positive; contrasts in other variables were either positive or negative, depending on whether they varied in the same direction as those in RANDOM1 probabilities. The fish phylogeny was inferred from the cladistic and taxonomic scheme presented in Nelson (1994); populations of the same species but from

Table 1
Data on communities of adult endoparasites used in the comparative analyses across fish species from Finland[a]

Fish species	No. examined	No. infected	Coefficient of variation in fish length	Component community richness	Mean infracommunity richness	Mean total intensity of infection	Fish length vs. richness[b]	RANDOM1 probability (observed *N*)
Bothnian Bay								
Leuciscus leuciscus	60	17	0.093	4	1.12	13.6	− 0.256	1.000 (1)
Rutilus rutilus	157	30	0.134	5	1.10	5.2	0.022	1.000 (5)
Esox lucius	49	48	0.328	9	2.98	41.7	0.574**	0.069 (96)
Osmerus eperlanus	272	262	0.142	4	2.05	25.8	0.411**	0.000 (198)
Coregonus albula	139	111	0.146	3	1.04	15.4	0.302*	0.133 (2)
Salmo salar	14	14	0.248	6	2.07	60.8	0.690*	0.075 (5)
Salmo trutta	51	48	0.337	8	1.63	35.7	0.391*	0.200 (59)
Lota lota	83	77	0.190	11	2.84	33.6	0.124	0.001 (229)
Gadus morhua	13	13	0.324	5	2.15	62.8	0.377	0.045 (2)
Gasterosteus aculeatus	393	55	0.116	3	1.07	4.7	− 0.009	1.000 (4)
Pungitius pungitius	281	38	0.126	3	1.11	4.9	0.098	0.413 (1)
Myoxocephalus scorpius	30	11	0.142	4	1.18	4.4	0.149	1.000 (4)
Pomatoschistus minutus	18	17	–	5	1.47	24.4	–	0.164 (6)
Zoarches viviparus	65	40	0.153	5	1.13	4.2	− 0.046	0.366 (10)
Gymnocephalus cernuus	224	139	0.189	6	1.26	3.8	− 0.122	1.000 (134)
Perca fluviatilis	139	126	0.215	6	1.71	6.0	− 0.104	0.128 (124)
Lake Yli-Kitka								
Esox lucius	60	56	0.202	8	2.09	12.3	0.259	0.062 (129)
Coregonus albula	80	53	0.104	4	1.42	2.4	0.201	0.068 (14)
Coregonus lavaretus	315	238	0.127	7	1.59	10.4	0.198*	0.012 (337)
Lota lota	21	21	0.205	7	3.43	39.1	− 0.183	0.892 (48)
Pungitius pungitius	90	37	0.187	3	1.05	7.7	0.294	0.403 (1)
Gymnocephalus cernuus	307	100	0.484	5	1.07	2.6	0.100	1.000 (21)
Perca fluviatilis	197	134	0.335	5	1.48	5.2	0.415**	1.000 (148)

[a] The RANDOM1 probability indicates whether infracommunities are structured according to a nested (≤0.05), random ($0.05 < P < 0.95$) or anti-nested (≥0.95) pattern; the observed index of nestedness, *N*, is also shown.

[b] Spearman rank correlation coefficient; $*P < 0.05$, $**P < 0.001$.

different locations were treated as sister species. All analyses follow the procedures recommended by Garland et al. (1992). Correlations between the contrasts in RANDOM1 *P*-values and the other seven variables described above, or between selected pairs from these seven variables, were computed as forced through the origin (see Garland et al., 1992, for justification). Residuals of a regression forced through the origin were used as estimates of contrasts in a dependent variable corrected for the independent variable, if and when necessary (Garland et al., 1992).

3. Results

3.1. Adult endoparasites

Only four component communities were excluded because they contained two or fewer parasite species, leaving a total of 23 component communities of adult endoparasites to be included in the analyses: 16 fish species from the Bothnian Bay, and seven from Lake Yli-Kitka (Table 1). Of these, four were significantly nested, and seven were significantly anti-nested; the number of nested communities dropped to two after a Bonferroni correction, whereas the number of anti-nested communities remained the same.

Nested and anti-nested assemblages (based on the less stringent criteria RANDOM1 *P*-value ≤0.15 and ≥0.85, respectively) did not differ with respect to number of fish examined, component community richness, or the coefficient of variation in fish length within samples (all $P > 0.21$). Nested assemblages tended to have infracommunities that had higher species richness, on average by 0.5 species, than anti-nested assemblages ($t = 2.109$, d.f. $= 16$, $P = 0.051$). However, nested assemblages were clearly characterised by higher prevalences and mean total intensities of infection, and stronger correlations between fish length and infracommunity richness, than anti-nested assemblages (Fig. 1). The number of fish examined correlated significantly with prevalence, mean infracommunity richness and mean total intensity of infection (all $P < 0.025$). After correcting for the influence of the number of fish examined (using residuals instead of raw values in the *t*-tests), though, the differences reported above between nested and anti-nested assemblages remained valid. Not surprisingly, there were also many pairwise correlations between various community parameters (richness, prevalence and intensity measures), suggesting that these parameters are not independent of one another. For instance, prevalence and mean total intensity correlated positively across assemblages ($r = 0.609$, $P = 0.002$) and the fact that nested and anti-nested assemblages differed with respect to both these parameters is a reflection of their covariation.

In the comparative analyses using phylogenetically independent contrasts, the number of fish examined did not correlate with any of the other variables except for prevalence ($r = -0.504$, $P < 0.025$). We thus used residuals of the prevalence vs. number of fish examined regression as corrected estimates of prevalence in subsequent tests.

Contrasts in RANDOM1 *P*-values did not correlate significantly with contrasts in any of the other variables. However, the majority of contrasts in both prevalence and mean intensity of infection (17 out of 22 contrasts in each case) are negative (Fig. 2). Since contrasts in RANDOM1

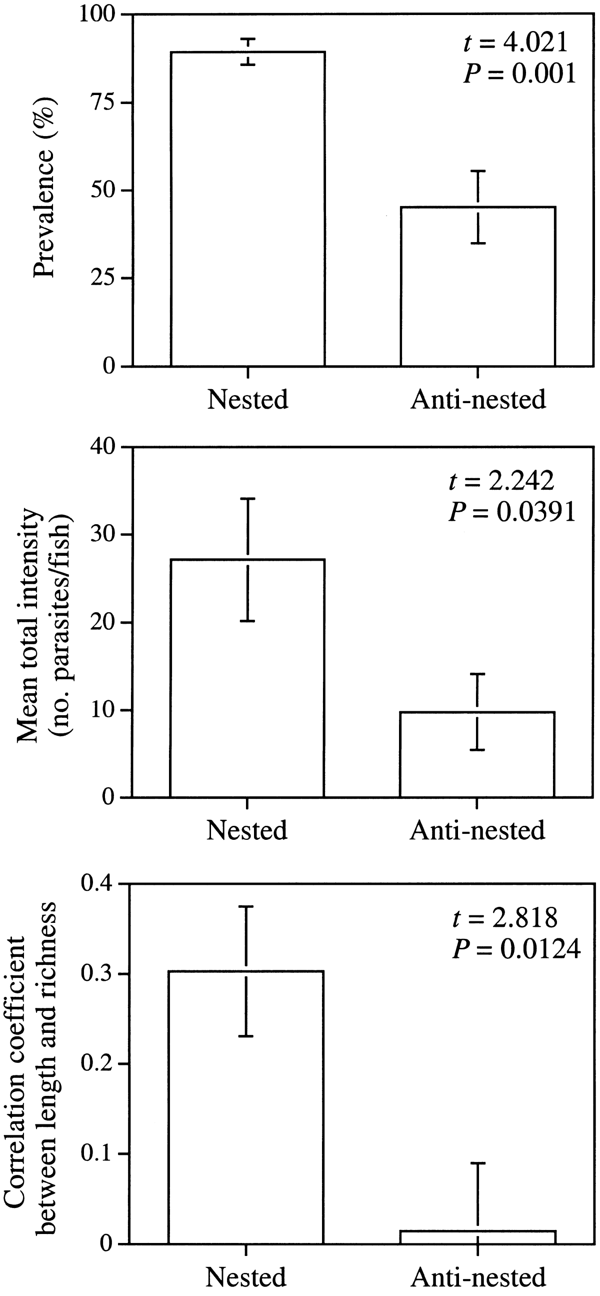

Fig. 1. Mean (±standard error) prevalence, total intensity of infection and Spearman correlation coefficient between fish length and infracommunity richness, for 10 nested and eight anti-nested assemblages of adult internal parasites of fish species from Finland. The results of two-tailed tests shown in the figure were obtained using log-transformed data.

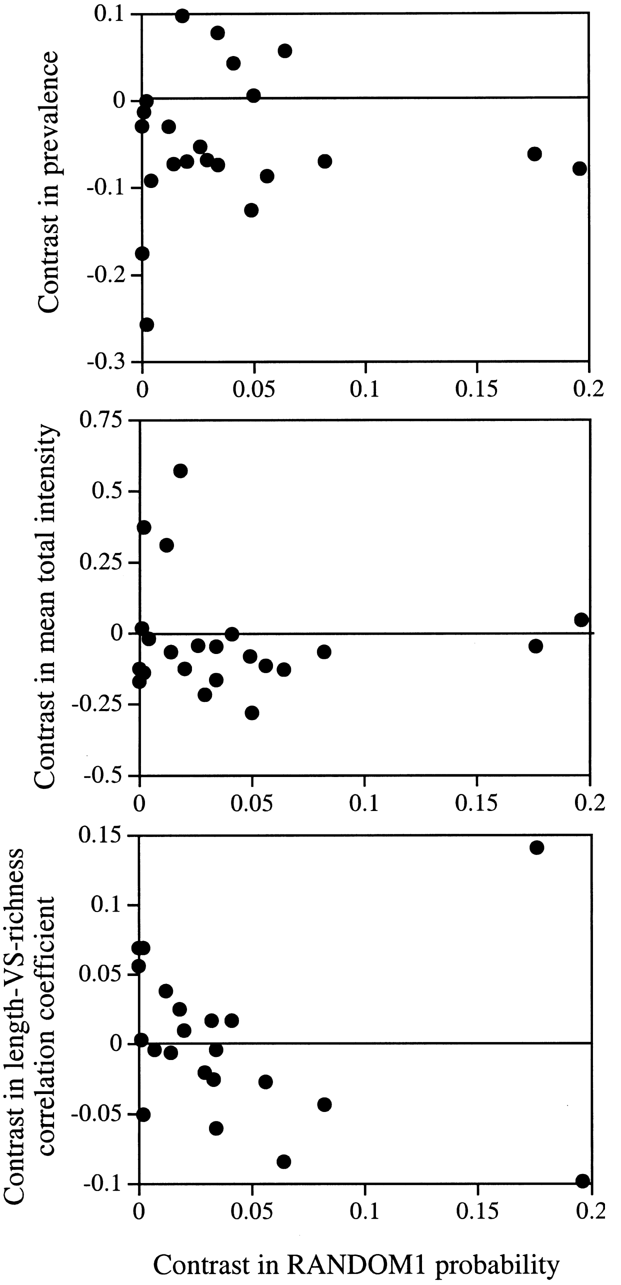

Fig. 2. Relationship between the RANDOM1 probability of an assemblage being nested, and prevalence (top), mean total intensity of infection (middle) and the Spearman correlation coefficient between fish length and infracommunity richness (bottom). Data are phylogenetically independent contrasts among assemblages of adult internal parasites of fish species from Finland. Contrasts in prevalence have been corrected for fish sample size (see text).

probabilities have been computed to be positive, this indicates a negative association between RANDOM1 *P*-values and both prevalence and intensity. Indeed, there are more negative contrasts in these two variables than are expected by chance ($\chi^2 = 6.55$, d.f. = 1, $P < 0.01$). There was also a weak tendency for contrasts in prevalence and mean intensity to be positively associated with one another ($r = 0.363$, $P < 0.10$).

Contrasts in the Spearman correlation coefficient between fish length and infracommunity richness were not significantly related with contrasts in RANDOM1 probabilities; however, after removing an outlier point from the analysis (the point in the top right-hand corner in Fig. 2), a relatively strong negative correlation was observed ($r = -0.618$, $P < 0.01$). Interestingly, this point corresponds to the contrast between the two populations of *Perca fluviatilis* and not to a basal contrast in the phylogeny. Removing the other outlier point instead (the one in the lower right-hand corner in Fig. 2) had no effect on the initial correlation result. Finally, there was a positive correlation between contrasts in the correlation coefficient between fish length and infracommunity richness and contrasts in the coefficient of variation in the fish length ($r = 0.613$, $P < 0.01$): the more variable the fish sizes within a sample, the stronger the Spearman correlation between fish length and infracommunity richness.

3.2. Larval endoparasites

After the exclusion of nine component communities because they contained two or fewer parasite species, a total of 18 component communities of larval endoparasites were included in the analyses: 14 fish species from the Bothnian Bay and four from Lake Yli-Kitka (Table 2). Of these, three were significantly nested, and seven were significantly anti-nested; the number of nested communities dropped to two after a Bonferroni correction, whereas the number of anti-nested communities remained the same.

There were no differences between nested and anti-nested assemblages (based on the less stringent criteria RANDOM1 *P*-value ≤ 0.15 and ≥ 0.85, respectively) in terms of number of fish examined, component community richness, mean infracommunity richness and the coefficient of variation in fish length within samples (all $P > 0.22$). As for adult parasite communities, nested assemblages displayed higher prevalences and mean total intensities of infection, and stronger correlations between fish length and infracommunity richness, than anti-nested assemblages (Fig. 3). There were no correlations between the number of fish examined and any of the other parameters, and therefore sampling effort is not a confounding variable in these results. As before, there were some correlations among the various community descriptors, but fewer than for adult endoparasites; in particular, prevalence and mean intensity of infection were strongly correlated ($r = 0.762$, $P = 0.0002$).

Using phylogenetically independent contrasts, the number of fish examined did not correlate with any of the other variables (all $P > 0.15$). Contrasts in RANDOM1 *P*-values did not correlate significantly with contrasts in component community richness, mean infracommunity richness or the coefficient of variation in fish length. However, contrasts in RANDOM1 *P*-values correlated negatively with contrasts in prevalence ($r = -0.896$,

Table 2
Data on communities of larval endoparasites used in the comparative analyses across fish species from Finland[a]

Fish species	No. examined	No. infected	Coefficient of variation in fish length	Component community richness	Mean infracommunity richness	Mean total intensity of infection	Fish length vs. richness[b]	RANDOM1 probability (observed *N*)
Bothnian Bay								
Clupea harengus	314	28	0.095	5	1.04	1.3	− 0.072	1.000 (3)
Esox lucius	49	10	0.281	4	1.10	4.1	0.058	1.000 (2)
Osmerus eperlanus	332	66	0.132	6	1.14	2.4	0.159	1.000 (26)
Coregonus albula	164	56	0.101	4	1.16	2.1	0.056	1.000 (15)
Salmo salar	14	10	0.161	3	1.50	62.6	− 0.132	0.339 (0)
Salmo trutta	51	39	0.331	5	1.21	18.7	− 0.196	0.177 (7)
Lota lota	83	78	0.195	8	2.42	7.2	0.150	1.000 (246)
Gadus morhua	13	11	0.330	4	1.46	8.9	0.635*	0.736 (8)
Gasterosteus aculeatus	360	281	0.099	7	1.95	3.7	0.494**	0.071 (608)
Pungitius pungitius	328	63	0.142	5	1.14	1.6	− 0.143	1.000 (27)
Myoxocephalus scorpius	30	29	0.146	5	2.28	33.5	0.334	0.003 (1)
Zoarches viviparus	65	25	0.168	5	1.20	2.1	0.007	0.772 (11)
Gymnocephalus cernuus	163	116	0.162	8	1.40	7.2	0.229*	0.123 (167)
Perca fluviatilis	128	65	0.212	5	1.05	4.8	− 0.033	0.643 (7)
Lake Yli-Kitka								
Rutilus rutilus	165	24	0.431	3	1.00	2.9	–	1.000 (0)
Coregonus lavaretus	247	240	0.151	3	1.02	87.3	− 0.003	0.017 (0)
Gymnocephalus cernuus	228	219	0.430	5	1.43	110.6	0.398**	0.000 (24)
Perca fluviatilis	171	163	0.329	3	1.06	54.6	0.060	0.292 (1)

[a] The RANDOM1 probability indicates whether infracommunities are structured according to a nested (≤ 0.05), random ($0.05 < P < 0.95$) or anti-nested (≥ 0.95) pattern; the observed index of nestedness, *N*, is also shown.

[b] Spearman rank correlation coefficient; $*P < 0.05$, $**P < 0.001$.

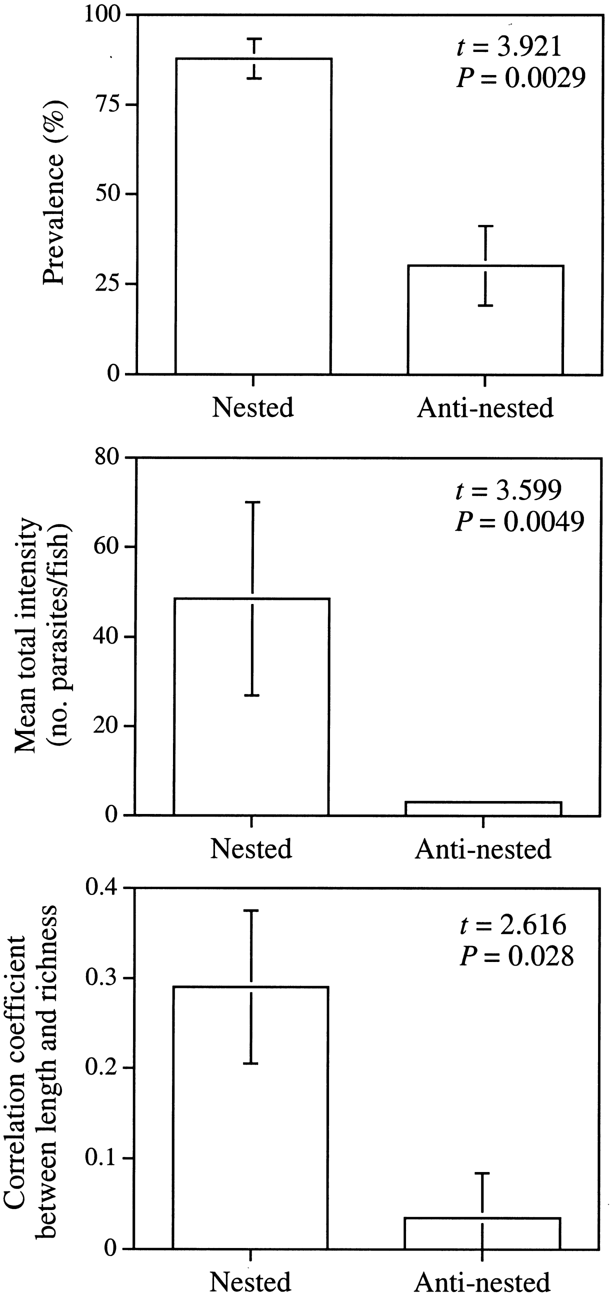

Fig. 3. Mean (±standard error) prevalence, total intensity of infection and Spearman correlation coefficient between fish length and infracommunity richness, for five nested and seven (only six in the bottom histogram) anti-nested assemblages of larval internal parasites of fish species from Finland. The results of two-tailed tests shown in the figure were obtained using log-transformed data.

$P < 0.001$), mean total intensity of infection ($r = -0.707$, $P < 0.001$) and the Spearman correlation coefficient between fish length and infracommunity richness ($r = -0.484$, $P < 0.05$). Most contrasts in both prevalence and intensity were negative (Fig. 4), clearly indicating a negative association with the contrasts in RANDOM1 probabilities computed to be positive. Contrasts in prevalence and intensity were also positively correlated with each other ($r = 0.745$, $P < 0.001$). The weaker relationship between contrasts in the correlation coefficient between fish length and infracommunity richness and contrasts in RANDOM1 probabilities, on the other hand, was entirely dependent on one outlier (the point in the bottom right-hand corner in Fig. 4): without it the correlation disappears. The likelihood of detecting a correlation between host length and infracommunity richness does not appear to depend on the range of fish sizes in a sample, as contrasts in the Spearman correla-

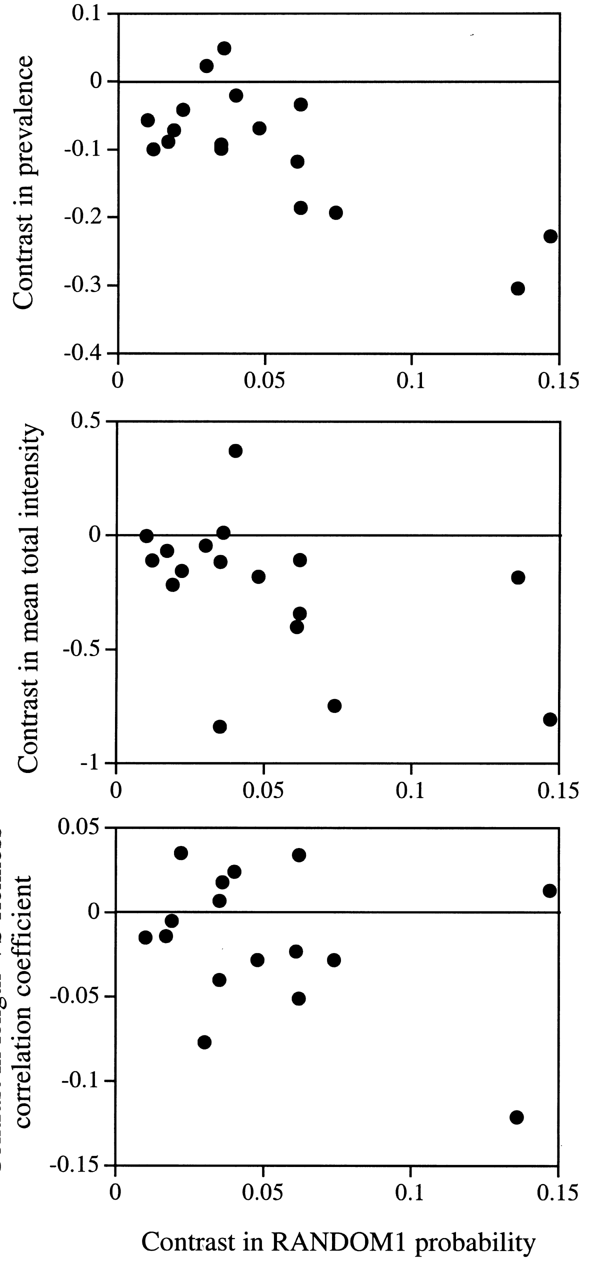

Fig. 4. Relationship between the RANDOM1 probability of an assemblage being nested and prevalence (top), mean total intensity of infection (middle) and the Spearman correlation coefficient between fish length and infracommunity richness (bottom). Data are phylogenetically independent contrasts among assemblages of larval internal parasites of fish species from Finland.

tion coefficient between fish length and infracommunity richness were not related to contrasts in the coefficient of variation in fish length ($r = 0.072$, $P > 0.50$).

4. Discussion

The occurrence of nested arrangements in species composition has been investigated in several types of animal and plant communities (Patterson and Atmar, 1986; Bolger et al., 1991; Patterson and Brown, 1991; Wright and Reeves, 1992; Cook, 1995; Worthen, 1996; Wright et al., 1998). In particular, external parasites living on fish have received much attention (Guégan and Hugueny, 1994; Worthen and Rohde, 1996; Rohde et al., 1998; Matejusová et al., 2000). The latest study on these parasites revealed a continuum in community organisation from nestedness to anti-nestedness that was linked to recent models of species coexistence (Poulin and Guégan, 2000). Here, we observed the same continuum among communities of internal helminth parasites of fish. The proportion of nested and anti-nested assemblages among the ones studied were approximately the same as those reported for communities of ectoparasites of fish (Rohde et al., 1998; Poulin and Guégan, 2000). This finding is in itself interesting. It indicates that nested patterns are roughly as infrequent in the non-interactive, colonisation-dominated ectoparasite communities of fish (Rohde, 1991) as they are in communities of adult endoparasites where density-dependent interspecific interactions are commonplace (Poulin, 2001). Our results also confirmed the suggestion of Poulin and Guégan (2000) that anti-nestedness is as uncommon as nestedness in parasite communities.

There were clear differences between nested and anti-nested assemblages with respect to the proportion of fish infected by parasites, or prevalence, and the mean total intensity of infection. These differences were not simply due to the confounding influences of sampling effort or host phylogeny, as they remained after the appropriate corrections were made. Interestingly, whether an assemblage is nested or not appears linked more with the overall abundance and commonness of parasites than with the richness of the parasite species pool locally available. Our results on endoparasites of fish mirror those of Poulin and Guégan (2000) on fish ectoparasites, for which these authors proposed several explanations. A non-biological explanation may be that the power of the algorithm to detect nestedness depends on how ‘full’ the presence–absence matrix is, i.e. on the general prevalence of the parasite species in a fish population (Wright et al., 1998). There is thus a methodological as well as an ecological explanation for the differences we observed, and determining the relative importance of each will require greater resolution (see Cam et al., 2000).

It is also possible that the frequency of anti-nestedness may be overestimated by the use of RANDOM1 *P*-values. If the observed index of nestedness, *N*, equals zero, then the assemblage is by definition perfectly nested (Patterson and Atmar, 1986). It is still possible in such cases to obtain RANDOM1 *P*-values approaching 1.0 if the simulated *N* scores are all, or almost all, equal to zero; this can lead to apparent anti-nestedness when it does not exist (W. Worthen and K. Rohde, personal communication). Poulin and Guégan (2000) did include cases in which $N = 0$ in their analyses, and some of their results may have been weakened had these been excluded. In our data set, however, of all 15 cases where the RANDOM1 *P*-value is greater than 0.85, there is only one in which the observed index of nestedness equals zero; this type of error is thus not likely to affect our analyses. This argument could be extended to cases where *N* is very small, though; if we remove cases where $N < 10$ from our analyses (i.e. most of the $P = 1.0$ in Tables 1 and 2), the direction of our results does not change but their statistical significance is greatly lowered. Caution is therefore needed when dealing with the anti-nested end of the nestedness continuum, where not all high *P*-values indicate true anti-nestedness.

Our other key result suggests an influence of fish biology on nested patterns, and thus community processes are not alone in structuring parasite assemblages. The finding that the likelihood of observing a significant correlation between fish size and infracommunity richness is closely linked with the RANDOM1 *P*-value has very important implications for our understanding of parasite community structure. When larger fish tend to harbour richer infracommunities, the infracommunities tend to form a nested pattern (low RANDOM1 *P*-value). When fish size does not covary with infracommunity richness, the infracommunities are randomly structured and even tend toward being anti-nested. This result was weakened somewhat when phylogenetic influences were removed. Still, the meaning of this may be that fish size drives the nestedness/anti-nestedness continuum via its effect, or lack of it, on infracommunity structure and species composition. Within the same fish population, larger fish tend to have richer parasite infracommunities because they eat more and have been accumulating parasites for longer than small fish. They thus sample the available parasite species at a higher rate and have done so for longer than small fish. When this happens, larger fish harbour most parasite species occurring in their habitat, both common and rare ones, whereas small fish are likely to harbour only the common parasites. The resulting structure of infracommunities is therefore one that departs from randomness, but without requiring the action of competitive or other species interactions. The effect of fish size could be to amplify intrinsic differences among parasite species in colonising abilities. The fact that this was also found in communities of larval endohelminths, where species interactions are unlikely anyway, reinforces the argument that fish size and its effect on species accumulation are sufficient to generate nestedness. This was first suggested by Guégan and Hugueny (1994) for a component community of ectoparasites on a freshwater fish, although their analysis has been questioned (Worthen and Rohde, 1996). This may be a major difference between communities of parasites and those of free-living organisms: in the former, the habitat (host) actively samples locally available species, whereas in the latter the habitat passively receives species from the regional pool.

The fact that, among assemblages of larval endohelminths, the coefficient of variation in fish length does not correlate with the Spearman correlation between size and infracommunity richness, or with the RANDOM1 *P*-value, indicates that the likelihood of observing nestedness is not merely due to the relative variability of fish sizes in a sample. In other words, it is not a statistical artefact, but is instead dependent on real biological processes. However, there was a relationship between variation in fish length and the Spearman correlation between fish size and infracommunity richness among assemblages of the adult endohelminths. Such correlations are always easier to detect when fish sizes are more variable (e.g. Poulin, 2000). There may, therefore, be a small artefactual element in the detection of nested patterns, whereby characteristics of the sample of hosts (or habitats, in the case of free-living organisms) can determine whether or not a pattern emerges from the data.

How would anti-nestedness, the other departure from random assembly observed here in parasite communities, be produced if hosts were the main structuring force in the communities? A simple explanation, consistent with the general absence of correlations between fish length and parasite species richness in anti-nested assemblages, would be that individual fish show dietary specialisations. If different subsets of conspecific fish feed on different prey and are thus only exposed to a small number of the locally available parasite species, fish size becomes less important in determining species richness. An anti-nested structure, as described in Fig. 1 of Poulin and Guégan (2000), would be a likely result.

The above discussion suggests that nested patterns are more a consequence of the way in which the host or habitat accumulates parasite species than of ecological processes acting among those parasite species. This does not mean that species interactions are of no importance in structuring communities of internal helminths. Nestedness analysis focuses only on the presence or absence of different species from infracommunities. It is possible that the intensity of infection of the different species depends on the presence of others, or that their resource usage and position in the host gut varies according to which species co-occur with them. These are also important indices of species interactions in parasite communities (Poulin, 2001). The main thing our results suggest is that the species composition of infracommunities may be determined by fish size, an extrinsic factor, with species interactions not important in determining this particular aspect of the communities. In ectoparasite communities on fish, host specificity can influence nestedness and other aspects of parasite community structure (Matejusová et al., 2000); this factor could also be important for endoparasite communities, in addition to the effect of fish size.

What of communities of organisms in general? Nested patterns are believed to appear across insular or fragmented habitats mainly as a result of differential colonisation or extinction probabilities among the available species (Patterson and Atmar, 1986; Patterson, 1990; Bolger et al., 1991). Differential extinction, in particular, is usually related to habitat area. Large areas maintain all species above their minimum viable population size, and increasingly small areas only contain the non-random subset of species that can tolerate a smaller or less predictable resource base (Patterson, 1990; Bolger et al., 1991). In such communities, the presence of a nested pattern is indicative of the intrinsic properties of the species present. The species richness vs. area relationships that emerge from the studies of free-living communities are to some extent similar to the species richness vs. host length relationships of parasite communities. If host size, through its effect on host feeding rates, is sufficient to generate a nested pattern, by amplifying differences among parasite species in colonisation or extinction probabilities, then the usefulness of nestedness analyses for parasite community studies will be limited. Nestedness can still serve as a useful descriptor of the composition of parasite communities (Worthen, 1996), but it may not reflect the intrinsic forces shaping parasite communities.

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