



EDITOR'S
CHOICE

The scaling of parasite biomass with host biomass in lake ecosystems: are parasites limited by host resources?

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The standing crop biomass of different populations or trophic levels reflects patterns of energy flow through an ecosystem. The contribution of parasites to total biomass is often considered negligible; recent evidence suggests otherwise, although it comes from a narrow range of natural systems. Quantifying how local parasite biomass, whether that of a single species or an assemblage of species sharing the same host, varies across localities with host population biomass, is critical to determine what constrains parasite populations. We use an extensive dataset on all free-living and parasitic metazoan species from multiple sites in New Zealand lakes to measure parasite biomass and test how it covaries with host biomass. In all lakes, trematodes had the highest combined biomass among parasite taxa, ranging from about 0.01 to 0.25 g m⁻², surpassing the biomass of minor free-living taxa. Unlike findings from other studies, the life stage contributing the most to total trematode biomass was the metacercarial stage in the second intermediate host, and not sporocysts or rediae within snail first intermediate hosts, possibly due to low prevalence and small snail sizes. For populations of single parasite species, we found no relationship between host and parasite biomass for either juvenile or adult nematodes. In contrast, all life stages of trematodes had local biomasses that correlated positively with those of their hosts. For assemblages of parasite species sharing the same host, we found strong relationships between local host population biomass and the total biomass of parasites supported. In these host–parasite biomass relationships, the scaling factor (slope in log-log space) suggests that parasites may not be making full use of available host resources. Host populations appear capable of supporting a little more parasite biomass, and may be open to expansion of existing parasites or invasion by new ones.

Quantifying and comparing the standing crop biomass of organisms at different trophic levels has provided key insights into how energy and matter flow through an ecosystem (Lindeman 1942, Odum 1957). In particular, the ratio of total biomass between two consecutive trophic levels has been used to estimate the efficiency of energy transfer from resources to consumers. Typically, the biomass of a trophic level is only about one-tenth of the biomass at the level below, giving rise to the pyramidal shape of biomass distribution in food webs reported in many classical studies (Lindeman 1942, Odum 1957, 1971). The contribution of parasites to total ecosystem biomass is widely assumed to be negligible, because of their small body sizes and cryptic existence (Polis and Strong 1996, Loreau et al. 2005). Parasites can occupy multiple positions in food webs, and indeed a given parasite species can change trophic levels from one life stage to the next (Lafferty et al. 2008); however, as they usually feed on hosts from mid- to high trophic levels, only a small fraction of the total biomass in the system is available to parasites (Sukhdeo 2012). Therefore, despite parasites having indirect effects on species interactions that are disproportionate to their size (Mouritsen and Poulin 2002, Hatcher and Dunn 2011), their direct contribution to ecosystem energetics is often assumed to be minimal.

Only very few recent studies have quantified the biomass of parasites across an entire ecosystem and compared it to that of free-living species. For example, Kuris et al. (2008) found that in three estuaries on the Pacific coast of North America, trematodes, the most common parasite taxon in these systems, achieved biomasses in the order of 0.8 to 2 g m⁻²; other parasite taxa occurred at much lower biomasses. Remarkably, the biomass of trematodes exceeded that of birds by at least three-fold, and was only slightly lower than that of fish and polychaetes in all three estuaries (Kuris et al. 2008). In three fishless Californian ponds, trematodes had total biomass values around 0.1 g m⁻², an order of magnitude lower than that of frogs and snails, but equal to or superior to that of any order of insects or crustaceans (Preston et al. 2013). Most of the trematode biomass in these ecosystems is localized within the intermediate hosts, i.e. juvenile stages of the parasites account for more biomass than adult worms inside their definitive vertebrate hosts (see also Sukhdeo 2012). It remains to be seen whether the findings summarized above are generally applicable to other types of ecosystems.

Another issue regarding parasite biomass is the extent to which it is resource limited. There is insufficient empirical evidence to determine whether parasites are subject to the

constraints of the classical biomass pyramid model, with their total biomass a roughly fixed fraction of their hosts' biomass. Higher host densities should support greater parasite population sizes (Anderson and May 1978, Arneberg et al. 1998), a pattern seen in the lake ecosystems that are the subject of the present study (Laguerre and Poulin 2015a). However, although density is a component of biomass, the inverse relationship between individual body mass and density means that population density and biomass are not necessarily correlated. Indeed, in our study systems, host density is only marginally correlated with host biomass across different species (Supplementary material Appendix 1, Fig. A2). Thus, not only is biomass different from density as a measure of host resource availability, but we can also compare the slope of the host–parasite biomass relationship to theoretical predictions (see below), something not possible for the host–parasite density relationship.

At the level of an individual host, both the average and the maximum biomass of metazoan parasites (all species combined) scale positively with host body mass, among individual hosts of the same species (Mouillot et al. 2005, Muñoz and George-Nascimento 2008) as well as across different host species (George-Nascimento et al. 2004, Poulin and George-Nascimento 2007). Based on a rigorous framework stemming from the metabolic theory of ecology (Brown et al. 2004), it is possible to calculate how host body size and metabolic rate determine the amount of space and energy available for parasites (Hechinger 2013). Both host and parasite body sizes are expected to influence the standing stock biomass of parasites per individual host, but to different extent depending on whether the parasites live within the host's tissues or on its external surface areas. Specifically, parasite biomass should increase with host body size to the $\approx 3/4$ power for endoparasites and to the $\approx 5/12$ power for ectoparasites (see Hechinger 2013 for details). These predicted scaling relationships apply equally to a single parasite species or to an assemblage of parasite species all infecting the same host, since multiple parasites sharing a host are still faced with finite resources constrained by the host's size and its size-specific metabolism (Hechinger 2013). They also apply specifically to carrying-capacity biomass, or the maximum mass of parasites that an individual host can support in principle. However, not all individuals in a host population are infected by parasites, and those that are infected can harbour vastly different numbers of parasites. Therefore, the above scaling factors ($\approx 3/4$ power for endoparasites, $\approx 5/12$ power for ectoparasites; Hechinger 2013) represent the theoretical maximum for the relationship between total population-level parasite biomass and host population biomass (i.e. the sum of body masses across all host individuals). The difference between this upper limit and the observed scaling factor can provide a rough estimate of how much host resources are unused by parasites, and how this varies among parasites at different life stages or from different taxa.

The goals of the present study are to quantify parasite biomass, contrast it to the biomass of free-living animals, and test its dependence on host biomass, across all metazoan species from four New Zealand lake ecosystems. First, we quantify the standing crop biomass of all parasite and free-living populations in all four lakes, and determine whether the relative magnitude of parasite biomass is comparable to

what has been previously measured in estuaries (Kuris et al. 2008) and frog-dominated ponds (Preston et al. 2013). We also identify the contribution of different helminth life stages, in intermediate or definitive hosts, to total parasite biomass. Second, we test the extent to which parasite biomass is constrained by host biomass at the population level, by quantifying the relationship between parasite biomass (either single-species or that of a mixed-species assemblage) and host biomass across sampling localities, for different parasite taxa and life stages. Based on observed patterns of infections (many uninfected hosts, and many infected hosts harbouring few parasites), we did not expect parasite species sharing the same host population to greatly influence each other's achieved biomass. Therefore, we predicted the total biomass of a multi-species parasite assemblage sharing a host population would scale differently (higher power) with increasing host biomass than the biomass of single parasite species. Our findings add to the still very small, but hopefully growing, number of studies of parasite biomass in the context of whole-ecosystem energetics.

Methods

Field sampling and parasite recovery

We investigated the entire community of free-living and parasitic metazoans from the littoral zone of 4 small-to-medium sized lakes on the South Island of New Zealand (Table 1) (for name, location and characteristics of each lake, see Supplementary material Appendix 1, Fig. A1 and Table A1). In each lake, we sampled 4 square areas (15×15 m) with one side of the square along the shore, distant by 123 to 2250 m from each other and selected to represent all habitat types (substrate, macrophytes, riparian vegetation, etc.) present within each lake. This gave us 16 study sites ($4 \text{ lakes} \times 4 \text{ sampling sites per lake}$). Each site was sampled in three seasons (September 2012, January and May 2013) to assess temporal changes in the communities, and

Table 1. Taxonomic composition (numbers of taxa) of the free-living and parasite communities in the four New Zealand lakes. For parasites, the numbers in parentheses indicate the number of different life stages considered.

Taxon	Lake Hayes	Tomahawk Lagoon	Lake Tuakitoto	Lake Waiholā
Free-living				
Fish	4	5	8	10
Crustaceans	11	10	11	16
Insects	23	18	22	11
Mites	3	0	2	1
Gastropods	4	3	3	2
Bivalves	1	1	2	2
Annelids	4	5	5	4
Nemertean	1	0	0	0
Turbellarians	1	1	1	1
Cnidarians	1	1	2	1
Parasites				
Trematodes	13 (21)	15 (24)	14 (23)	16 (25)
Cestodes	–	–	1 (1)	1 (1)
Nematodes	1 (1)	2 (2)	3 (3)	5 (6)
Acanthocephalans	–	–	1 (1)	2 (3)
Mites	1 (1)	–	1 (1)	–

on each occasion we sampled all fish, benthic and demersal invertebrates, zooplankton and all metazoan parasites on or within these organisms. Our study excludes birds (and their parasites), which could not be sampled for ethical and conservation reasons; no amphibians occur in the lakes studied.

Fish were sampled using a combination of gear types following a standardized protocol to achieve estimates that represented as accurately as possible actual fish diversity and density (see Supplementary material Appendix 1 for full details). Two fyke nets were set overnight along the edges of the sampling area, perpendicular to the shore, and two 15 m long multi-mesh gillnets were deployed in the same place during the day. These were used to capture all fish swimming in and out of the area, i.e. both residents and visitors to the area. In addition, a standard, fine-mesh purse seine net was dragged across the whole area to capture sedentary resident fish not captured by passive gear like fyke nets or gillnets. All fish caught were identified to species, counted, and a subsample was returned to the laboratory for weighing and dissection. In each site and in each season, 6 samples of benthic invertebrates, distributed haphazardly across the sampling area, were taken using a standard Surber sampler net with a 0.1 m² horizontal metal frame fitted with a 250 µm mesh collecting net. In addition, 6 samples of demersal invertebrates, living on or near the substrate but not captured in Surber nets, were sampled using a rectangular dip net (30 cm wide and 22 cm high opening) with a 250 µm mesh net; each sample consisted of a fast, 2 m-long sweep of the net along the lake bottom without dredging the substrate. All invertebrate samples were preserved in ethanol for later identification, counting and dissection. Finally, four plankton samples were taken per site and per season in each lake. Sampling was done at night by towing a conical net (25 cm mouth diameter) made of fine nylon mesh (90 µm mesh size) through the water for a 3 m horizontal distance. Samples were distributed haphazardly across the sampling site. Zooplankton were fixed in 70% ethanol for later identification and count. The volume of water from which zooplankton were obtained was estimated as the product of tow length and the area of the net opening. Density m⁻³ was then converted to density m⁻² by projection of the number of individuals per plankton taxon contained in 1 m³ of lake water onto the flat surface necessary to contain that 1 m³ of water based on water depth at each sampling site.

Overall, >660 000 individuals of free-living species were sampled, counted and identified. In the laboratory, large subsamples of each fish (>600 individuals total) and invertebrate (>40 000 individuals total) species were dissected carefully following a standardised protocol for parasite recovery and identification. In the case of small and transparent invertebrate species, individuals were instead pressed between glass plates and scanned under a microscope for parasite detection (see Supplementary material Appendix 1 for full details). From these dissections, >370 000 parasite individuals were recovered and identified.

Biomass measurements

All biomass data used in the following analyses are available from the Dryad Digital Repository: <<http://dx.doi.org/10.5061/dryad.523t1>> (Laguerre and Poulin 2015b). For

each free-living taxon and for each life stage of each parasite taxon, biomass (g m⁻²) was calculated for each of the 16 study sites (4 lakes × 4 sampling sites per lake) by multiplying density (no. individuals m⁻²) by the average body mass of one individual (wet weight of whole organism, including bones, exoskeleton, shell, etc., after removal of excess ethanol/water). For parasites, density was calculated by summing up all parasite individuals of a particular species and life stage across all infected hosts from a sample taken over a given surface area; these estimates were then averaged across all samples (and seasons) from the same site.

All fish were weighed to the nearest 0.01 g, allowing calculation of a mean individual mass for each fish species and for each site. Most free-living invertebrates were large enough to be weighed individually (e.g. isopods, chironomids, odonates, large Trichoptera larvae, adult hemiptera, molluscs, leeches, etc.). Invertebrates varied little in size intraspecifically and by weighing a subsample of individuals for each taxon (to the nearest 0.01 mg) we calculated the mean body mass of an individual for all those invertebrate taxa. For small free-living invertebrates, which also varied little in size intraspecifically (amphipods, small Trichoptera larvae, oligochaetes, planktonic crustaceans, etc.), we pooled 5, 10 or 20 conspecific individuals (depending on individual body size) from random subsamples, weighed them as a group, and from the total mass calculated the average body mass of one individual.

Parasites were too small to be individually weighed and body measurements indicated that they varied little in size within life stages of each species. We thus calculated body volume for the subsamples of parasite individuals measured during host dissection based on the most appropriate formula for each species' shape (e.g. adult nematodes and acanthocephalans, trematode rediae and sporocysts = cylinder; adult trematodes = flattened ellipsoid; encysted juvenile trematodes [metacercariae] = spheres). We verified that body dimensions of parasites had not been affected by preservation and storage methods (frozen in fish tissue, ethanol) and represented those of live individuals (see Supplementary material Appendix 1 for details). Body volume was calculated for each life stage of each species and their volume was converted to mass assuming their density equalled that of water. We could thus calculate a mean individual body mass for each life stage of each species.

To allow a coupling between parasite biomass and the biomass of the host in which they occur, we had to treat different life stages of the same parasite species as separate 'populations'. Distinct stages of parasites with complex life cycles exploit completely different host species and infect them via different transmission routes. As one of our goals is to determine if parasite biomass is constrained by host biomass, host biomass must represent the resources available to a particular set of transmission stages. Therefore, different parasite life stages are treated here as separate 'parasite populations'; each corresponds to a unique parasite life stage-by-site combination. For each site and each sampling season, we first averaged the biomass values of each host or parasite population across all samples. Then, we calculated the mean biomass per site across all three seasons.

In most cases, a parasite population (as defined above) exploited a single host species. However, in cases where the

parasites used two or more host species, the main host was defined as the species harbouring the most parasite individuals. If the second most-important host species harboured at least a quarter of the number of parasites found in the main host, it was combined with the main host to calculate mean host biomass for that parasite population. In the rare cases where the third most-important host species harboured at least a quarter of the number of parasites found in the second most-important host species, it was also included in calculations of mean host biomass.

Data analysis

To quantify the standing crop biomass of all parasite and free-living populations, we summed the biomasses of all taxa belonging to the same higher taxonomic group for each sampling site, before averaging them across the four sites in each of the four lakes. This yielded a single average biomass ($\pm$ SD) per lake for each higher taxonomic group of parasites and free-living animals. For trematodes (the main parasite taxon in our systems), we also did this for each life stage, to identify the contribution of different life stages, in intermediate or definitive hosts, to total parasite biomass.

To test the extent to which parasite biomass is constrained by host biomass at the population level, we only included life stages of parasite taxa for which there were at least 10 data points, i.e. 10 unique parasite species–life stage–site combinations. We regressed log-transformed parasite biomass against log-transformed host biomass across sampling localities, for different parasite higher taxa and life stages. The fitted slope of each of these relationships corresponds to the power at which parasite biomass scales with host biomass. We obtained them from mixed-effects models implemented in JMP ver. 11.0 (SAS Inst., Cary, NC, USA), with log-transformed parasite biomass as response variable and log-transformed host biomass as fixed factor. This allowed us to account for any phylogenetic influences or idiosyncrasies of particular localities, and for the non-independence and correlated structures in the data. It also allowed us to calculate the proportion of the total variance unexplained by the fixed effects that could be accounted for by the following random effects: lake and sampling site (nested within lake), and parasite species.

Finally, we performed similar analyses by considering the biomass of the total assemblage of parasite species using a host population. We focused on the two most widely used and spatially widespread host species in our lake systems: the snail *Potamopyrgus antipodarum* and the fish *Gobiomorphus cotidianus*. These are the only two host species present at all sites, with a broad range of local biomasses, and parasitized at all sites. For each of the 16 sampling sites, we summed the biomasses of all trematodes using the snail as their first intermediate host, and all parasites of any taxa infecting the fish. Then, as above, we regressed log-transformed parasite total biomass against log-transformed host biomass across sampling localities. The two relationships were fitted using ordinary least squares regressions, from which we obtained their slope.

Results

Parasite versus free-living biomass

The lake communities we studied were dominated, in terms of species richness, by fish, crustaceans and insects as free-living animals, and by trematodes as parasites (Table 1). In total, we recovered 35 parasite species, most occurring in more than one lake (for complete list of these parasites, see Supplementary material Appendix 1, Table A2).

Absolute and relative biomasses of different higher taxa of free-living and parasitic animals varied among lakes (Fig. 1).

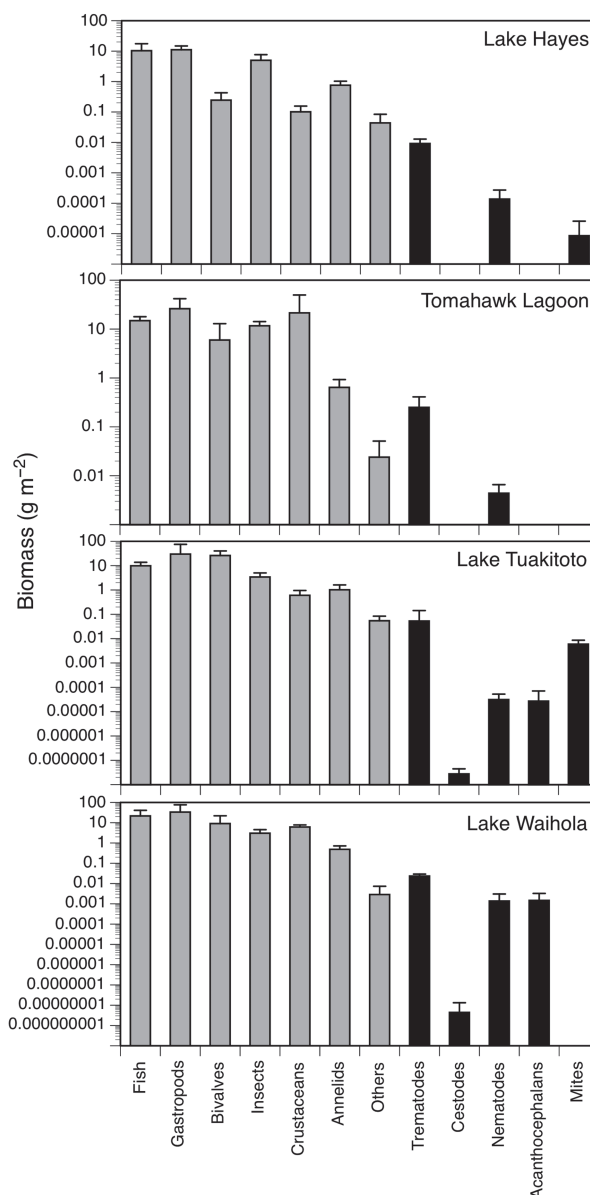


Figure 1. Mean ($\pm$ SD) total biomass of all free-living (grey bars) and parasitic (black bars) taxa in 4 lakes from the South Island, New Zealand. Note the logarithmic scale on the y-axis. Each value is the mean of 4 sampled sites per lake (see text); biomass values for parasitic helminths include only aquatic life stages, and not adult stages in bird definitive hosts for the species to which this applies. 'Others' include free-living mites, nemerteans, turbellarians and cnidarians.

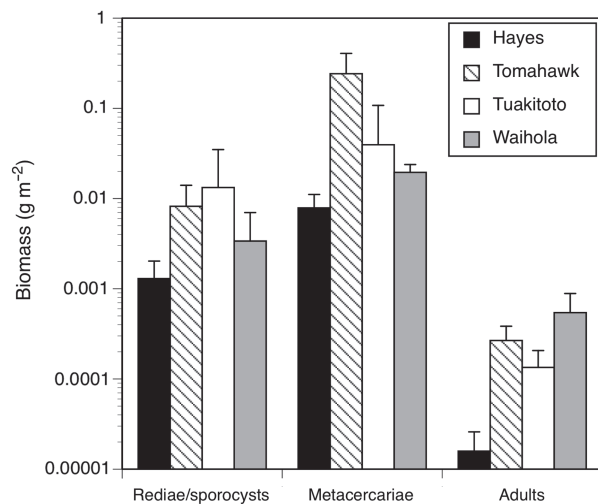


Figure 2. Mean (+ SD) total biomass of trematodes at three distinct life stages, in 4 lakes from the South Island, New Zealand. Note the logarithmic scale on the y-axis. Each value is the mean of 4 sampled sites per lake (see text); biomass values for adult stages include only those occurring in fish, and not those in bird definitive hosts for the species to which this applies.

Among parasite taxa, only trematodes and nematodes were found in all four lakes. In all lakes, trematodes had the highest biomass among parasites, ranging from about 0.01 to 0.25 g m⁻² (Fig. 1). Although trematode biomass approached or even surpassed the pooled biomass of minor free-living taxa (mites, nemerteans, turbellarians and cnidarians), it never approached that of any of the more important free-living groups, which had biomasses 1–2 orders of magnitude larger than that of trematodes.

In all four lakes, the life stage contributing the most to total trematode biomass was the metacercarial stage in the second intermediate host (Fig. 2). This was followed in all cases by sporocysts or rediae within the snail first intermediate host, with adult worms in fish definitive hosts contributing the least biomass.

Parasite–host biomass regressions

These analyses include only life stages of parasite taxa for which there were at least 10 data points, i.e. 10 unique parasite species–life stage–site combinations. They are therefore

limited to all life stages of nematodes and of trematodes (for complete list of these parasites, see Supplementary material Appendix 1, Table A2). For both juvenile and adult nematodes, we found no significant relationship between host biomass and parasite biomass. In contrast, all three life stages of trematodes had local biomasses that correlated positively and significantly with those of their hosts (Table 2). The slopes of these relationships in log-log space, corresponding to the scaling of parasite biomass with host biomass, were always lower than the theoretically expected value of 0.75; however, that value fell within the 95% confidence intervals of all three slope estimates (Table 2). In some cases, the identity of the parasite species as well as the locality in which they were sampled accounted for a substantial fraction of the variance not explained by host biomass (Table 2). Indeed, although for each of the three life stages, positive relationships between host biomass and parasite biomass were found in more than half of the species when these were tested separately, it did not apply to all species (Supplementary material Appendix 1, Table A2).

When similar analyses were done using the biomass of the total assemblage of parasite species using a host population, we found strong relationships between local host population biomass and the biomass of parasites supported (Fig. 3). The overall biomass of all trematodes using the snail *Potamopyrgus antipodarum* increased across sites with host biomass with a slope of 0.958 in log-log space (95% CI = 0.590–1.325; $R^2 = 0.690$, $p < 0.0001$). For parasites of the fish *Gobiomorphus cotidianus*, the slope was even steeper at 1.222 (95% CI = 1.140–1.304; $R^2 = 0.987$, $p < 0.0001$). These slopes lie above the theoretically expected value of 0.75, although that for trematodes in *P. antipodarum* included 0.75 within its 95% confidence interval. Ratios of total parasite biomass to host population biomass varied across the 16 localities, but not substantially; expressed as a percentage, the average ratio for snails was 0.1% (range 0.02–0.23%); for fish, it was 2.2% (range 0.7–5.2%).

Discussion

Recent evidence indicates that parasite biomass in natural communities is not as negligible as previously thought, and that parasites can play important roles in ecosystem energetics (Kuris et al. 2008, Preston et al. 2013). Our findings from lake communities add to those from earlier studies on

Table 2. Slopes of the relationships between log-transformed parasite biomass and log-transformed host biomass, in which each data point corresponds to a unique parasite species–life stage–site combination. Tests of significance and the proportions of the variance in parasite biomass unaccounted for by host biomass that can be explained by the identity of the parasite species or the site (nested within lake) of sampling, were obtained from mixed-effects models.

								% variance explained		
Taxon and life stage	Host	No. data points	No. different species	Slope	95% confidence intervals	R ²	p	Parasite species	Site [lake]	Lake
Trematodes										
rediae/sporocysts	1st intermediate	96	15	0.595	0.292–0.898	0.136	0.0002	49.6	11.3	1.6
metacercariae	2nd intermediate	139	16	0.655	0.548–0.762	0.513	<0.0001	82.1	1.3	2.8
adults	definitive	50	6	0.606	0.405–0.807	0.433	<0.0001	8.3	3.8	37.2
Nematodes										
juveniles	intermediate	21	3	0.340	–	0.122	0.1208	3.6	10.2	58.9
adults	definitive	16	3	0.144	–	0.002	0.8659	1.1	59.4	7.4

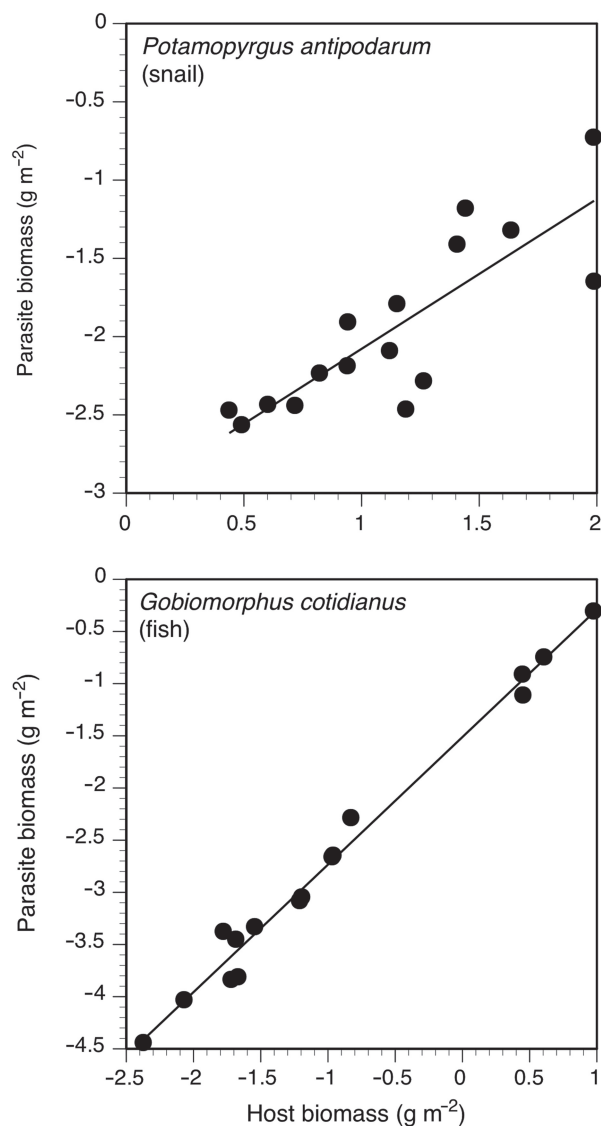


Figure 3. Total parasite biomass, pooled across all species using the host population, as a function of local host population biomass, for trematodes using the snail *Potamopyrgus antipodarum* as first intermediate host, and for all parasites infecting the fish *Gobiomorphus cotidianus*. Each point represents an estimate from a different sampling site ($n = 16$) in lakes from the South Island, New Zealand.

ponds and estuaries to further confirm the biomass contributions of parasites. In addition, we quantified how local parasite biomass scales with varying host biomass among sites, for both single parasite species and entire mixed-species parasite assemblages. These relationships shed new light on how available host biomass constrains parasite biomass, and suggest that host populations may be a little 'underutilized' by their parasites.

Some of the observed variation in biomasses of different higher taxa of free-living and parasitic animal may come from the fact that some taxa (i.e. fish) were weighed fresh whereas others were weighed after storage in alcohol. Preservation in alcohol leads to loss of body weight due to dehydration and leaching of organic compounds, and the relative amount of weight lost differs among higher taxa (Mills et al. 1982,

Leuven et al. 1985). However, alcohol preservation typically reduces body weight by 10–15% or less for the major taxa we studied (molluscs, insects, crustaceans), and given the magnitude of some of the differences we reported (note log-scale in Fig. 1), weight loss due to alcohol storage is very unlikely to affect our main findings.

As in other aquatic ecosystems in which parasite biomass has been quantified and compared to that of free-living taxa, trematodes were the most important group of parasites. Their total biomass in our study lakes, which ranged from about 0.01 to 0.25 g m⁻², is comparable to that found by Preston et al. (2013) in three fishless Californian ponds, but roughly one order of magnitude lower than what Kuris et al. (2008) found in three estuaries on the Pacific coast of North America. To some extent, this may be due to differences in the way biomass was measured in different studies (dry mass in Preston et al. 2013; wet mass in Kuris et al. 2008 and the present study). There is also the fact that adult trematodes using birds as definitive hosts were not included in our estimates, although they can be considered as part of the same ecosystem. However, based on bird counts and dissection of a few dead aquatic birds recovered over the past few years (unpubl.), it is unlikely that these missing trematodes account for much of the difference between trematode wet biomass estimates from freshwater systems (present study) and those from coastal marine systems (Kuris et al. 2008). Higher species diversity of trematodes and higher prevalences of infection in snail first intermediate hosts in the marine systems than in our lakes are more likely explanations for the discrepancy.

Contrary to previous studies (Kuris et al. 2008, Preston et al. 2013), we found that encysted metacercariae within their second intermediate hosts were the life stage contributing the most to total trematode biomass, in all four lakes, and not the asexually multiplying sporocysts or rediae within snail first intermediate hosts, as seen elsewhere. There are several potential reasons for this different pattern. Despite its very high densities, the snail *Potamopyrgus antipodarum*, which is the only snail we found acting as first intermediate host of trematodes, is small-bodied (shell length 2–5 mm) and short-lived compared to the dominant snail hosts in previously studied systems (*Cerithidea californica* in Kuris et al. 2008; *Helisoma trivolvis* in Preston et al. 2013). Also, *P. antipodarum* is only infected at very low prevalences by the various trematode species. Thus, infected snails are few, and each harbours only a small amount of trematode tissue due to its small size. This represents only a modest standing stock biomass at this particular life stage. In contrast, the free-swimming dispersal stages, or cercariae, produced in snail hosts and released continuously or at intervals, can far exceed that biomass if summed up over a long period of time, such as a year (Kuris et al. 2008, Thieltges et al. 2008, Morley 2012, Preston et al. 2013). Because cercariae in our study systems can accumulate (as metacercariae) in relatively long-lived hosts like fish, their biomass can surpass that of the life stage which produced them, and account for much of total trematode biomass.

From an epidemiological perspective, higher host densities should increase parasite transmission success and lead to greater parasite population sizes (Anderson and May 1978, Arneberg et al. 1998), an expectation realised in

our lake ecosystems (Laguerre and Poulin 2015a). From an energetic perspective, we may also expect host biomass to determine the biomass of any given parasite species (George-Nascimento et al. 2004, Poulin and George-Nascimento 2007). At an individual host level, carrying-capacity parasite biomass should increase with host body size to the $\approx 3/4$ power for endoparasites (Hechinger 2013). However, not all host individuals in a population harbour parasites, and those that are infected do not always harbour many parasites. Therefore, the $\approx 3/4$ power represents the theoretical upper limit for the population-level relationship between parasite biomass and host biomass, and it is unlikely to ever be realised in nature. We found no significant relationship between local parasite biomass and host biomass for both juvenile and adult nematodes; in contrast, we found positive relationships for all three life stages of trematodes. It is unclear why the two taxa of parasites should respond differently to changes in host biomass; admittedly, our analyses of nematode biomass were based on much fewer data points than those on trematodes. For all three trematode life stages, the host–biomass versus parasite–biomass relationships had slopes that were slightly lower, but not significantly different from those predicted for the maximum carrying-capacity (i.e. scaling power of $\approx 3/4$ corresponds to a slope of 0.75 in log-log space). This suggests, however only weakly, that the host population as a whole is somewhat underexploited by parasites, i.e. that a slightly greater parasite biomass could theoretically be supported by the available energy from the hosts. In our systems, the size (and therefore the biomass) of parasite populations may be constrained more by transmission dynamics and other epidemiological processes than by limited host resources. It must be pointed out that our conclusions rest solely on estimates of standing-stock biomass, and not on productivity. It is possible that productive host species, with a high turnover of biomass per unit time, represent better resources for parasites and can support greater parasite biomass. This remains an important issue for future empirical studies to explore.

A similar conclusion emerges from the analyses we performed on spatial variation in the biomass of whole mixed-species parasite assemblages as a function of local host biomass. These analyses were restricted to two widespread host species with rich parasite faunas: the snail *P. antipodarum* and the fish *G. cotidianus*. The parasites and hosts in these cases represent successive trophic levels, with the parasite consumers achieving a biomass that is only 0.1% (for snails) or 2.2% (for fish) of that of the host resource. Because host biomass is calculated using host density, it is not possible to disentangle the respective effects of host density and host biomass on parasite biomass; therefore, we only use the slopes of the parasite–host biomass relationship as an indication of the extent to which available host resources are used. If parasite species utilizing the same host share available host resources, then their pooled carrying-capacity biomass should also scale with host body size to the $\approx 3/4$ power (Hechinger 2013). However, the slopes observed tended to be greater than 0.75, significantly so in the case of the fish host. This indicates that, at least in the case of the fish host, the biomasses of different parasite species are additive, such that one species' use of host resources does not constrain the biomass that others can achieve in the same

host population. Various observations support this conclusion. For instance, out of 10 385 individual snails dissected for this study, we found low infection prevalences by all species of trematodes co-occurring at any given site. Among species using snails as first intermediate hosts (life stage = sporocyst or redia), we found no case of two or more species infecting the same individual snail; among those using snails as second intermediate hosts (life stage = metacercaria), we observed 10 cases where a snail harbouring many individuals of one trematode species also had 1–2 individuals from a different species. There is thus no strong evidence of competition among parasite species for host resources whereby one species might limit the abundance or biomass of another.

In the case of the fish host, the parasites accounting for most of the biomass are four trematode species occurring as encysted metacercariae in various host tissues. An earlier study of one of these species found that the size achieved by an individual parasite is totally unaffected by the number of con- or heterospecific metacercariae in the same host (Herrmann and Poulin 2011). Typically, after a brief initial growth phase immediately following infection of the fish host, metacercariae encyst and probably derive very little further energy from the host. Space is the main resource they use, and this would only become a limiting factor at unusually high intensities of infection. Again, this supports a scenario in which parasites do not compete for host resources, and achieve biomasses independently of those of other co-occurring parasites. Similarly, an earlier study on endoparasites of fish has also found that the species richness of parasites per host correlated positively with their total biomass (Mouillot et al. 2005), suggesting that the biomasses of different parasite species are not greatly influenced by the simultaneous presence of other parasites in the same host.

These findings are relevant to a long-standing debate in parasite community ecology regarding whether or not parasite communities are saturated with species, i.e. whether host individuals or host populations can support additional species of parasites (Kennedy and Guégan 1994, 1996, Poulin 1996, Morand et al. 1999, Norton et al. 2004). In general, the relationship between the maximum number of parasite species seen in an individual host and the number of parasite species available locally is linear: the more species are present locally, the more can successfully establish in individual hosts (Poulin 1996, Norton et al. 2004). Co-existence of multiple species of parasites in a host population can be facilitated by all species having independent patterns of aggregated distribution among individual hosts, which reduces the likelihood of interspecific interactions (Morand et al. 1999). Our study focuses on total parasite biomass and not parasite species richness, and the unit of study is the host population, not host individuals. Despite these differences, our results parallel those of earlier studies by suggesting a slight deficit between what can theoretically be supported by hosts and what we find exploiting them. Our data on the scaling of parasite biomass with host biomass also indicate limited interspecific effects among parasites, at least in the fish *G. cotidianus*, and support the unsaturated nature of parasite communities. This suggests that available host resources could not only support the population expansion of any existing parasite species, but also the invasion of new parasites.

Our findings contribute to the growing appreciation of parasites as non-trivial players in ecosystem energetics. To existing data from estuaries and fishless ponds, we add data on fish-dominated lakes. The small discrepancies between the patterns we observed and those reported from other systems highlight the idiosyncrasies of energy flow and biomass production in different types of systems, and call for future efforts to target other type of communities (e.g. terrestrial habitats). Our results also suggest that a portion of host biomass appears to be unexploited by parasites, and that host resources may not be the main limiting factor preventing an increase in parasitism in natural ecosystems.

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Supplementary material (Appendix ECOG-01720 at <www.ecography.org/appendix/ecog-01720>). Appendix 1.