Differential impacts of shared parasites on fitness components among competing hosts

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Abstract

Effects of parasites on individual hosts can eventually translate to impacts on host communities. In particular, parasitism can differentially affect host fitness among sympatric and interacting host species. We examined whether the impact of shared parasites varied among host species within the same community. Specifically, we looked at the impacts of the acanthocephalan Acanthocephalus galaxii, the trematodes Coitocaecum parvum and Maritrema poulini, and the nematode Hedruris spinigera, on three host species: the amphipods, Paracalliope fluviatilis and Paracorophium excavatum, and the isopod, Austridotea annectens. We assessed parasite infection levels in the three host species and tested for effects on host survival, behavior, probability of pairing, and fecundity. Maritrema poulini and C. parvum were most abundant in P. excavatum but had no effect on its survival, whereas they negatively affected the survival of P. fluviatilis, the other amphipod. Female amphipods carrying young had higher M. poulini and C. parvum abundance than those without, yet the number of young carried was not linked to parasite abundance. Behavior of the isopod A. annectens was affected by M. poulini infection; more heavily infected individuals were more active. Paracorophium excavatum moved longer distances when abundance of C. parvum was lower, yet no relationship existed with respect to infection by both M. poulini and C. parvum. The differential effects of parasites on amphipods and isopods may lead to community-wide effects. Understanding the consequences of parasitic infection and differences among host species is key to gaining greater insight into the role of parasite mediation in ecosystem dynamics.

Keywords

host behavior, host fitness, multispecies infection, parasites, survival

1 | INTRODUCTION

Community structure and dynamics are affected by the direct interactions of competition and predation, as well as indirect interactions, such as trophic cascades, keystone predation, and parasite mediation (Hatcher & Dunn, 2011; Holt & Pickering, 1985; Park, 1948; Price et al., 1986). Host responses to parasitism can vary widely and affect growth, behavior, reproduction, aging, and ability to respond to stressful conditions (Bedhomme, Agnew, Vital, Sidobre, & Michalakis, 2005; Brown & Pascoe, 1989; Cox, 2001; Thomas, Guegan, & Renaud, 2009; Thompson, Redak, & Wang, 2001). The extent of these impacts can also vary greatly among hosts, both inter- and intraspecifically (Shaw & Dobson, 1995). These variations among and within host species may affect competition and predation dynamics, eventually impacting...
the structure of the entire community (Rauque et al., 2011; Smith, Acevedo-Whitehouse, & Pedersen, 2009; Tompkins, Dunn, Smith, & Telfer, 2011).

Direct effects of parasitism on hosts can include alteration of feeding rates (Rivero & Ferguson, 2003), behavior (Lefèvre et al., 2009; Poulin, 1995; Thomas, Renaud, Demee, & Poulin, 1998), stress response (Bedhomme et al., 2005; Brown & Pascoe, 1989), survival (Lehmann, 1992), and ability to compete for resources (Park, 1948; Price, Westoby, & Rice, 1988). Parasite can also have direct effects on reproduction, through the total or partial castration of the host via gonad destruction or reduced energy stores needed for egg production (Rauque et al., 2011), or indirect ones through a reduction in pairing success or parental care (Bollache, Gambade, & Cézilly, 2001; Lefebvre, Fredensborg, Armstrong, Hansen, & Poulin, 2005; Rauque et al., 2011; Read, 1988). Changes in host behavior, even subtle, can in turn have community-wide repercussions (Lefèvre et al., 2009; Poulin, 1995; Thomas et al., 1998). Behavioral modification can include changes in activity levels (Kunz & Pung, 2004; Leung & Poulin, 2006; Webster, 1994), position in the water column (Hansen & Poulin, 2005; Rauque et al., 2011), aggression (Mikheev, Pasternak, Taskinen, & Valtonen, 2010), boldness (Reisinger, Petersen, Hing, Davila, & Lodge, 2015), and photophilia (Bauer, Trouvé, Grégoire, Bollache, & Cézilly, 2000; Rauque et al., 2011). Alterations of host behavior may lead to increased vulnerability to predation (Kunz & Pung, 2004) and reduced ability to compete for resources (Mikheev et al., 2010; Reisinger et al., 2015). Consequently, parasites may influence the outcome of competition among hosts and impact community dynamics.

Furthermore, different parasite species can vary greatly in their impacts on hosts. For instance, the pairing success of male Gammarus pulex amphipods was affected differently by infection with Pomphorhynchus laevis than Polymorphus minutus, both acanthocephalan parasites (Bollache et al., 2001). Many parasite species are known to infect a variety of hosts, many of which may compete strongly with each other within their ecosystem. Differential impacts of parasites on competitors will affect their relative competitive abilities (Price et al., 1986). As species vary in their susceptibility and tolerance to parasites, the presence or absence of parasite species may dictate the coexistence of species or the complete absence of a species within an ecosystem (Greenman & Hudson, 2000; Hatcher, Dick, & Dunn, 2006). If two species are equal competitors, the presence of a parasite that infects only one of them may change this interaction. If the host is negatively affected by the parasite, it may give the competitor the advantage. The more tolerant host may also be able to act as a reservoir for the parasites, maintaining a high level of parasitism within the system (Arneberg, Skorping, Grenfell, & Read, 1998). To better understand the outcomes of competition and predation in the presence of shared parasites, it is important to understand the different impacts parasites may have on particular hosts.

Impacts of parasites on their hosts are usually studied in simple one parasite — one host species context. However, multispecies infections are not uncommon (Alizon, de Roode, & Michalakis, 2013; Hughes & Boomsma, 2004; Lagrue & Poulin, 2008a; Lange, Reuter, Ebert, Muylaert, & Decaestecker, 2014; Pedersen & Fenton, 2007; Thumbi et al., 2014). Furthermore, although each infection event is often independent, the presence of multiple parasite species within a host may have synergistic or antagonistic effects compared to the presence of one parasite alone (Alizon et al., 2013; Lagrue & Poulin, 2008a; Lange et al., 2014). However, the effects of diverse within-host parasite assemblages can often be very difficult to predict (Alizon, 2013). Interactions among parasites may affect their respective virulence and the survival of the host (Alizon, 2013; Balmer, Sternarn, Schützau, & Brun, 2009; Lange et al., 2014; de Roode, Culleton, Cheeseman, Carter, & Read, 2004). The overall virulence of a combination of parasites can be higher than that of the most virulent parasite, lower than the least virulent one or reach some intermediate level (Alizon et al., 2013). Yet, despite the importance of interactions among shared parasites and multispecies infections, little is understood about how shared parasites may shape host communities.

The objectives of our study were to examine potential impacts of different parasite species on three species of hosts in the same community. We examined the effects of parasites on invertebrates used as intermediate hosts by four parasite species, all of which are transmitted trophically to their definitive host. Parasite effects on host fecundity, behavior, and survival have been previously examined in one of the host species, the amphipod Paracalliope fluviatilis (Lagrue & Poulin, 2008a; Rauque et al., 2011). It serves as host to two trematode species, Coitocaecum parvum and Maritrema poulini, and the acanthocephalan Acanthocephalus galaxii. Coitocaecum parvum, and A. galaxii use fish as definitive hosts while M. poulini is an avian parasite (Hine, 1977; MacFarlane, 1939; Presswell, Blasco-Costa, & Kostadinova, 2014). Less is known about the impacts of these parasites on other hosts in the community. Additionally, potential effects of multiple infections are not well understood. Two other crustacean species are commonly found coexisting with P. fluviatilis and serve as hosts to some of the same parasites. Paracorophium excavatum, another amphipod, is larger than P. fluviatilis but they both occur in sympatry (Ruiz-Daniels, Beltran, Poulin, & Lagrue, 2012). Paracorophium excavatum is also host to three parasites, including the trematodes C. parvum and M. poulini, but also the fish nematode Hederis spinigera (Lagrue & Poulin, 2008b; Luque, Bannock, Lagrue, & Poulin, 2007; Luque et al., 2010; Ruiz-Daniels et al., 2012). The prevalence and abundance of C. parvum and M. poulini have previously been found to be higher in P. excavatum than in P. fluviatilis (Ruiz-Daniels et al., 2012). The isopod Austrodotea annectens is also found in the same area and is an intermediate host for M. poulini (Hansen & Poulin, 2005; Presswell et al., 2014). Many of these parasites reach a relatively large size and/or abundance within their hosts, suggesting potential impacts on host survival and behavior (Rauque et al., 2011). Our specific objectives were to (i) determine whether parasite effects varied among hosts within the same community and (ii) test whether multispecies infections had synergistic or antagonistic effects compared to the presence of single parasite infection (Alizon et al., 2013; Lagrue & Poulin, 2008a; Lange et al., 2014). As these hosts are all competing for resources and share a variety of parasites, a better understanding of the impacts of parasitism on each host is necessary to understand how parasites affect population dynamics in this community.
2 | METHODS

2.1 | Sample collection

We collected samples of naturally infected amphipods and isopods from the littoral zone of Lake Waihola, South Island, New Zealand (46°01′14S, 170°05′05E) between February and September 2016. Sampling for survival tests occurred over 3 days, 9 February, 21 March, and 1 May 2016. Sampling for the behavioral tests occurred over 3 days, 13 May, 1 June, and 6 September 2016, due to seasonal variation in host abundance. Sampling for pairing behavior occurred on 21 March 2016 for P. fluvialilis (as described below) but paired isopods were collected during the entire sampling period as they occurred far less frequently. Data on the size, sex, and the prevalence and abundance of parasites in each host species from each sample event were pooled for examination of intraspecific and interspecific variation. Animals were caught using dip-nets and transported to the laboratory in lake water. Amphipods and isopods were transferred and maintained separately by species in 10 L tanks containing aerated lake water. Animals were kept at room temperature (14 ± 1°C) and maintained separately by species in 10 L tanks containing aerated lake water. Animals were kept at room temperature (14 ± 1°C) with aquatic plants (Myriophyllum triphyllum and Elodea canadensis) for food, and under a controlled photoperiod (12-hr dark and light).

All amphipods used in behavioral and fecundity trials were subsequently dissected within a week of collection as keeping these amphipods in the laboratory for long periods of time can affect amphipod survival (Lagruè & Poulin, 2007; Lagruè, Poulin, & Keeney, 2009; Poulin, 2003). All isopods used in behavioral trials were dissected within a month of collection. If individuals did not die during trial, they were killed in 70% ethanol and rinsed in distilled water before dissection. In our study, prevalence was defined as the percentage of infected hosts, abundance was defined as the number of parasites per host including zeroes, and mean abundance as the mean number of parasites among a specific sample of hosts.

2.2 | Survival tests

Within 6 hr after sampling, 266 P. fluvialilis, 210 P. excavatum, and 390 A. annectens were separated into individual wells of tissue culture plates. According to host size, P. fluvialilis were maintained in 96-well microplates with 300 μL of water per well, P. excavatum were maintained in 24-well plates with 500 μL of water per well, and A. annectens were kept in 12-well plates with 1 mL of water per well. All individuals were maintained at the same temperature (14 ± 1°C) and photoperiod (12-hr dark and light) but no food was added. Well plates were checked daily for any dead individuals. If a female released young, the number of young was recorded and they were removed from the well as they could have provided an additional food resource to the focal animal through cannibalism. If individuals could be dissected the same day, they were left in lake water. For dissection occurring more than 24 hr after death, the individuals were immediately preserved in 70% ethanol until dissection. The total body length of each individual was determined by measuring from the anterior tip of the cephalic capsule to the posterior end of the uropods. Sex was determined for each individual when possible. Isopods shorter than 7–8 mm in body length were impossible to sex due to the lack of secondary sexual characters and thus considered juveniles. Egg presence and number were also recorded. Individuals were then dissected to identify and count parasites.

2.3 | Behavioral tests

Amphipods and isopods were individually isolated (in wells of culture plates, as described above) within 6 hr of sampling and left for 12 hr to acclimate to their new environments. Individuals were subsequently filmed to record velocity and activity levels. Infection status of each individual was unknown during filming and subsequent video analysis. Fine sand was added to the wells of P. excavatum and A. annectens to simulate natural conditions as both species are benthic and may use sand to burrow. One hour prior to recording, plates were moved to the filming studio to allow the animals to adjust to the temperature (18 ± 1°C) and lighting changes. Paracalliope fluvialilis and P. excavatum were filmed for 5 min under a dissecting microscope (Olympus SZ61, 0.65× magnification) due to their small size. Well plates containing A. annectens were filmed for 5 min using a Canon digital camera (1200D). Activity levels (distance moved from center (mm), mean velocity (mm/ms), highly mobile duration (more than 60% of the animal (measured by pixels altered) has moved within the sample period (0.05 s), which is then calculated as a proportion of the entire sample to give a measure of high-speed movement), and mobile duration (more than 20% of the animal (measured by pixels altered) has moved within the sample period (0.05 s), which is then calculated as a proportion of the entire sample to give a measure of movement) were calculated for each individual over 5 min using EthoVision XT (Noldus Information Technology 2015). Male and female amphipods were combined as no difference in behavior between sexes was observed (P. fluvialilis: distance moved from center, Kruskal–Wallis tests Z = 1.3, p = .19; mean velocity, Kruskal–Wallis tests Z = 1.3, p = .19; highly mobile duration, Kruskal–Wallis tests Z = 1.2, p = .23; and mobile duration, Kruskal–Wallis tests Z = −0.93, p = .35; P. excavatum: distance moved from center, Kruskal–Wallis tests Z = −0.42, p = .67; mean velocity, Kruskal–Wallis tests Z = −0.40, p = .68; highly mobile duration, Kruskal–Wallis tests Z = 0.58, p = .58; and mobile duration, Kruskal–Wallis tests Z = 0.014, p = .99).

2.4 | Fecundity and pairing probability

Offspring carried in the brood pouch of gravid females (from the survival and behavioral tests) were counted and recorded with their corresponding body length and parasite burdens. If any young were released during survival tests, the number of young was recorded and matched with the female’s corresponding parasite burden upon death. The parasite burden of females without any young was also compared to those with young to examine whether a relationship existed between the probability of having young and a female’s parasite burden.

Additionally, a subset of paired P. fluvialilis and A. annectens were identified and individually separated into tissue culture microtest
tubes within 12 hr of capture. The paired individuals consisted of a male claspers a female in a precopulatory pair. Nonpaired individuals were also collected during the same sampling event and separated into individual tubes. Amphipods were dissected as described above within 24 hr of capture. Isopods were euthanized and preserved in ethanol and dissected as described above.

2.5 | Statistical analysis

Statistical analyses were performed in JMP® 12 (SAS Institute Inc 2015) and R statistical software (http://www.R-project.org). Size differences between sexes were examined using a Kruskal–Wallis test. The relationship between host size and parasite abundance was assessed using Spearman’s correlations. Differences in parasite abundance and prevalence among host species and between sexes of individual parasite species were examined using a Kruskal–Wallis test and contingency analysis, respectively. The relationship between either survival (days before death) or behavioral measures, and parasite abundance of C. parvum, M. poulini, A. galaxii, and H. spinigera, as well as multispecies combinations, was analyzed separately for each host species using a negative binomial regression, with host size being included as an additional explanatory variable and their interactions when significant. Logistic regression was used to relate parasite abundance with both the likelihood of being paired in P. fluviatilis and A. annectens by sex and the probability of having young in female P. fluviatilis and P. excavatum. The number of young carried by a female was related to its body size and parasite abundance using negative binomial regression.

3 | RESULTS

Parasite prevalence and abundance varied greatly among host species (Table 1). The highest number of individual of M. poulini per host was 74 (in A. annectens), 8 for C. parvum (in P. excavatum), and 1 for both A. galaxii (in P. fluviatilis) and H. spinigera (in P. excavatum). Multiple infections were found in both amphipod species, with C. parvum and M. poulini co-infecting 1.65% of P. fluviatilis and 20.7% of P. excavatum, and M. poulini and H. spinigera co-infecting 3.7% of P. excavatum. One P. excavatum (0.34%) was infected with three parasite species (C. parvum, M. poulini, and H. spinigera). Acanthocephalus galaxii was not found sharing the same individual host with any other parasites within our sample. The abundance and prevalence of M. poulini varied significantly among all hosts (abundance: Kruskal–Wallis tests, \( Z^2 = 967, p < .0001 \); prevalence: Contingency analysis, \( Z^2 = 1033.6, p < .0001 \)), with the abundance being highest in P. excavatum and lowest in P. fluviatilis (Table 1). A synopsis of the key results of this study is summarized in Table 2.

Amphipod sex ratio was approximately 3:1 females to males for P. fluviatilis and 2.5:1 females to males for P. excavatum. Isopod sex ratio was approximately 1:1 among individuals that were large enough to be sexed. Size differences between sexes were found in P. fluviatilis (Kruskal–Wallis test, \( Z = 12.68, p < .0001 \)) but not P. excavatum (\( Z = -0.47, p = .64 \)), or A. annectens (\( Z = -1.62, p = .10 \)). Sex and size were both related to parasite infection in P. fluviatilis. As observed in

| Species | Sex | N | Size (mm) | AG | CP | HS | MP | Total |
|---------|-----|---|-----------|----|----|----|----|-------|
| P. fluviatilis | Female | 687 | 2.0 ± 0.02 | 0.9 | 19 | 5.8 | 24 | 0.009 ± 0.004 | 0.26 ± 0.026 |
| | Male | 220 | 2.6 ± 0.03 | 3.3 | 23 | 12 | 34 | 0.002 ± 0.003 | 0.29 ± 0.045 |
| | Female | 21 | 4.2 ± 0.40 | 0.3 | 24 | 3.3 | 91 | 92 | 0.033 ± 0.012 | 0.085 ± 0.006 |
| | Male | 21 | 4.6 ± 0.44 | 0.3 | 24 | 3.3 | 91 | 92 | 0.033 ± 0.012 | 0.085 ± 0.006 |
| P. excavatum | Female | 83 | 8.4 ± 0.18 | – | – | 86 | 86 | 0.006 ± 0.001 | 0.35 ± 0.030 |
| | Male | 24 | 8.8 ± 0.29 | – | – | 88 | 88 | 0.006 ± 0.001 | 0.35 ± 0.030 |
| A. annectens | Female | 21 | 8.4 ± 0.18 | – | – | 86 | 86 | 0.006 ± 0.001 | 0.35 ± 0.030 |
| | Male | 24 | 8.8 ± 0.29 | – | – | 88 | 88 | 0.006 ± 0.001 | 0.35 ± 0.030 |
| Hosts                  | Parasites                  | A. galaxii (acanthocephalans) | C. parvum (trematode) | H. spinigera (nematode) | M. poulini (trematode) | Multiple infection (M. poulini and C. parvum) | Multiple infection (M. poulini and H. spinigera) |
|------------------------|----------------------------|--------------------------------|------------------------|-------------------------|------------------------|-----------------------------------------------|--------------------------------------------------|
| **Paracalliope fluviatilis** (amphipod) | Prevalence | Low | Low | - | Low | Low | - |
|                        | Abundance     | Low | Low | - | Low | Low | - |
|                        | Survival      | No effect | Reduced | No effect | Reduced | Reduced | Reduced |
|                        | Activity      | Reduced | No effect | No effect | No effect | No effect | No effect |
|                        | Fecundity     | No effect | More with eggs | No effect | More with eggs | No effect | No effect |
|                        | Pairing - ♀   | Higher | Lower | - | No effect | No effect | No effect |
|                        | ♂             | No effect | No effect | No effect | No effect | No effect | No effect |
| **Paracorophium excavatum** (amphipod)  | Prevalence    | - | Low | Low | High | Low | Low |
|                        | Abundance     | - | Low | Low | High | Low | Low |
|                        | Survival      | No effect | No effect | No effect | No effect | No effect | No effect |
|                        | Activity      | Reduced | No effect | No effect | No effect | No effect | No effect |
|                        | Fecundity     | More with eggs | No effect | More with eggs | No effect | Fewer with eggs | |
|                        | Pairing - ♀   | - | - | - | - | - | - |
|                        | ♂             | - | - | - | - | - | - |
| **Austridotea annectens** (isopod)   | Prevalence    | - | - | - | High | - | - |
|                        | Abundance     | - | - | - | High | - | - |
|                        | Survival      | - | - | - | Improved | - | - |
|                        | Activity      | - | - | - | Increase | - | - |
|                        | Fecundity     | - | - | - | No effect | - | - |
|                        | Pairing - ♀   | - | - | - | No effect | - | - |
|                        | ♂             | - | - | - | Higher | - | - |
previous studies, abundance and prevalence of *M. poulini* (Kruskal–Wallis test, $Z = 3.1$, $p = .002$; contingency analysis, $\chi^2 = 8.9$, $p = .003$) and *A. galaxii* (Kruskal–Wallis test, $Z = 6.3$, $p = .012$; contingency analysis, $\chi^2 = 5.3$, $p = .021$) were higher in male than female *P. fluviatilis* (Rauque et al., 2011). However, no difference was found between males and females of *P. fluviatilis* in the abundance or prevalence of *C. parvum* (all $p$-values $>.05$). *Paracalliope fluviatilis* size was positively related to *C. parvum* abundance (Spearman $\rho = 0.10$, $p = .0025$) and multiple infection of *C. parvum* and *M. poulini* (Spearman $\rho = 0.078$, $p = .019$) but not related to *A. galaxii* or *M. poulini* abundance (all $p$-values $>.05$).

Analyzed separately, the size of female *P. fluviatilis* was positively related to *C. parvum* abundance (Spearman’s $\rho = 0.11$, $p = .0044$) but male size was not (Spearman’s $\rho = 0.03$, $p = .63$). Separating the sexes did not change the lack of relationship between size and the abundance of *A. galaxii* and *M. poulini* for females or males (all $p$-values $>.05$).

In contrast, female *P. excavatum* had a higher abundance and prevalence of *M. poulini* (Kruskal–Wallis test, $Z = -2.24$, $p = .025$; contingency analysis, $\chi^2 = 5.76$, $p = .016$) and *C. parvum* (Kruskal–Wallis test, $Z = -1.70$, $p = .089$; contingency analysis, $\chi^2 = 2.59$, $p = .11$) than males. *Paracorophium excavatum* size was positively related to *M. poulini* abundance (Spearman’s $\rho = 0.19$, $p = .0014$). However, size was not related to *H. spinigera* abundance, *C. parvum* abundance, or multiple infections (all $p$-values $>.05$). Analyzing the sexes separately, both male and female *P. excavatum* size, is related to *M. poulini* abundance (females, Spearman’s $\rho = 0.41$, $p < .001$; males, Spearman’s $\rho = 0.25$, $p = .021$).

Isopods were infected by *M. poulini* only. The abundance of *M. poulini* in *A. annectens* varied greatly between sexes, with males having the highest abundance, followed by females, and then juveniles (ANOVA, $F_{2.544} = 95.24$, $p < .001$. Tukey’s HSD post hoc test, all $p < .001$). Size was positively related to *M. poulini* abundance (Spearman’s $\rho = 0.33$, $p < .001$).

### 3.1 | Host survival and parasite infection

The survival (i.e., number of days before death) of *P. fluviatilis* was negatively related to *M. poulini* and *C. parvum* abundance and host size (Table 4, Figure 1). There was no interactive effect between either parasite abundance and size (Table 4). Survival was not related to the abundance of *A. galaxii*. However, there was an interactive effect between host size and *A. galaxii* abundance (Table 4).

No relationship was found between host survival and *M. poulini*, *C. parvum*, or *H. spinigera* abundance in *P. excavatum* (Table 4, Figure 1). Intriguingly, there was a positive relationship between *M. poulini* abundance and survival in *A. annectens*, as well as a relationship between host size and parasite abundance with an interactive effect between size and abundance (Table 4, Figure 1).

### 3.2 | Host behavior

The distance moved from the center in *P. fluviatilis* depended on the abundance of *A. galaxii* (Table 4). While all other measures of behavior in *P. fluviatilis* (velocity, high mobile duration, and mobile duration) were unaffected by parasite abundance (*A. galaxii*, *M. poulini*, or *C. parvum*), *Paracalliope fluviatilis* size did influence host behavior, as larger hosts moved further and had longer highly mobile durations (Table 4).

In *P. excavatum*, the distance moved from the center of the well plate was negatively related to the abundance of *C. parvum* with a significant interactive effect between host size and *C. parvum* abundance.
Additionally, individuals who spent more time highly mobile tended to have a lower abundance of *C. parvum* or to be smaller, with a trend toward an interactive effect between *C. parvum* and size (Table 4). Mobile duration (time spent moving) of *P. excavatum* was negatively related to size and tended to be positively related to *M. poulini* abundance with a trend toward an interactive effect between *M. poulini* abundance and size (Table 4). However, there was no relationship between or distance moved or high mobile duration and the abundance of *M. poulini*, *H. spinigera*, or size (Table 4). Mobile duration was not related to the abundance of *A. galaxii* or that of *C. parvum*. Velocity (mm/ms) was not related to the abundance of any parasite or size (Table 4). No relationship was found between velocity (ms/s) or distance moved from center and *M. poulini* abundance or size (Table 4).

Mobile duration (time spent in motion) of *A. annectens* was negatively related to *M. poulini* abundance but not to host size, although there was a significant interaction effect (Table 4, Figure 3). High mobile duration was not related to *M. poulini* abundance, but it was related to size (Table 4). No relationship was found between velocity (ms/s) or distance moved from center and *M. poulini* abundance or size (Table 4).

### 3.3 | Fecundity and pairing probability

A subset of 141 *P. fluviatilis* had young (mean ± SE, 3.6 ± 0.16 per female). The number of young carried was not related to either *M. poulini* or *C. parvum* abundance (all p-values > .05). The number of young carried was positively related to amphipod size (Z = 5.6, p < .001), with larger females carrying more young. There was no relationship between multispecies infection abundance, that is, *C. parvum* with *M. poulini*, and the number of young (Z = 0.065, p = .95).

We found a higher abundance of *M. poulini* (logistic regression, $\chi^2 = 17.1, p < .001$) and *C. parvum* ($\chi^2 = 4.2, p = .04$) in female *P. fluviatilis* with young (Figure 2). Consistently, the prevalence of *M. poulini* was higher in females with young (Contingency analysis, $\chi^2 = 21, p < .0001$). However, the prevalence of *C. parvum* was higher in females with young (logistic regression, $\chi^2 = 8.8, p = .0031$). There was no relationship between the presence of young and *A. galaxii* or multispecies infection abundance or prevalence (all p-values > .05). There was a relationship between the size of a female *P. fluviatilis* and the likelihood of having young ($\chi^2 = 38, p < .001$), with the mean size of females with young (1.8 ± 0.024 mm) being significantly smaller than those without (2.1 ± 0.025 mm).

A subset of 71 *P. excavatum* had young (mean = 6.24 ± 0.01). The number of young *P. excavatum* carried was not related to the abundance of *H. spinigera*, *M. poulini*, *C. parvum*, or female size, and there was no interactive effect (all p-values > .05). Females with a higher abundance of *C. parvum* ($\chi^2 = 4.0, p = .046$) and *M. poulini* ($\chi^2 = 4.7, p = .031$) were more likely to have young (Figure 2). However, we found no relationship between the abundance of *H. spinigera* and the likelihood of having young in *P. excavatum* (logistic regression, $\chi^2 = 1.4, p = .24$; Figure 2). There was no relationship between multispecies infections by *C. parvum* with *M. poulini* ($\chi^2 = 1.2, p = .27$); however, there was a trend of higher abundance of *H. spinigera* and *M. poulini* in *P. excavatum* without young than those with young ($\chi^2 = 3.1, p = .076$). We were unable to examine any relationships between the fecundity of *A. annectens* and the abundance of *M. poulini* due to a low sample size.

### DISCUSSION

Understanding impacts of shared parasites on a variety of sympatric host species is necessary for making predictions on the potential
TABLE 3  Parasite abundance (mean ± SE) and prevalence (percentage of sample infected) in paired and unpaired Paracallophium fluviatilis and Austridotea annectens (listed with sample size per group). All samples were collected from Lake Waihola, South Island, New Zealand between February and September 2016

| Host               | Parasite          | Males                  | Females         |
|--------------------|-------------------|------------------------|-----------------|
|                    |                   | Paired | Non-paired | χ² | p      | Paired | Non-paired | χ² | p      |
|                    |                   | n = 64 | n = 156    |    |        | n = 60   | n = 627    |    |        |
| Paracallophium     | A. galaxii        | 0.016 ± 0.016 | 0.040 ± 0.016 | 0.88 | .35    | 0.05 ± 0.028 | 0.0048 ± 0.0028 | 7.0 | .0083 |
| fluviatilis        |                   | 1.6%  | 3.8%       | 0.58 | .45    | 4.9%  | 4.8%       | 6.5 | .011  |
| C. parvum          | 0.19 ± 0.068      | 0.29 ± 0.050 | 0.85 | .36    | 0.15 ± 0.11 | 0.25 ± 0.026 | 1.5 | .23    |
|                    | 17%  | 25%        | 2.1   | .15    | 6.6%  | 20%       | 9.5 | .0021  |
| M. poulini         | 0.19 ± 0.08       | 0.13 ± 0.044 | 0.79 | .37    | 0.12 ± 0.060 | 0.065 ± 0.014 | 2.3 | .13    |
|                    | 14%  | 11%        | 0.29   | .59    | 9.8%  | 5.4%      | 1.3 | .25    |
| M. poulini + C.    | 0.097 ± 0.001     | 0.077 ± 0.004 | 0.35 | .56    | 0.097 ± 0.013 | 0.040 ± 0.0009 | 0.74 | .39    |
| parvum             | 3.1%  | 3.8%    | 0.075   | .78    | 1.6%  | 1.2%      | 0.045 | .83   |

|                    | Austridotea       | M. poulini | 15 ± 3.6 | 21 ± 3.5 | 1.2 | .28 | 17 ± 6.7 | 11 ± 3.0 | 0.88 | .35 |
| annectens          |                   | n = 13     | n = 30    |    |        | n = 11   | n = 27    |    |        |
|                    |                   | 100%  | 87%       | 3.1  | .081*  | 91%  | 74%       | 1.5 | .22   |

Significant differences through logistic regressions are shown in bold, Trends followed by *.

role of parasites in ecosystem structure and functioning. We found that the impacts of parasites varied among host and parasites species within our study community. Survival rates varied among amphipods, with one showing a reduced lifespan when infected by either of the two shared parasites, whereas survival of the other amphipod species was not affected. We also found evidence that when a host species is infected by multiple parasites, the effects of each parasite may have been antagonistic to one another, with a net neutral effect on the host.

Infection levels varied among hosts within the community. Paracorophium excavatum had higher abundance of M. poulini than P. fluviatilis (Table 1), which is consistent with previous studies (Presswell et al., 2014; Ruiz-Daniels et al., 2012). Variation in host size or other biological characteristics may lead to these differences (Grutter & Poulin, 1998; Johnson, Bush, & Clayton, 2005; Ruiz-Daniels et al., 2012; Saad-Fares & Combes, 1992). Paracorophium excavatum is a larger amphipod that has a more benthic habitat than P. fluviatilis as it is often found burrowing in the sand. However, we only found a positive correlation between the size of P. excavatum and one species of parasite, contrary to previous studies (Presswell et al., 2014; Ruiz-Daniels et al., 2012). The lack of difference in prevalence and abundance of C. parvum between amphipod species may have been due to the low overall abundance and prevalence of C. parvum within the community, making it more difficult to detect any effect of this parasite on its hosts. Additionally, as discussed more thoroughly below, we found no relationship between the abundance of M. poulini and the survival of P. excavatum, suggesting that interspecific differences in abundance between the two amphipods may be due to higher parasite-induced mortality in P. fluviatilis following infection and/or parasite accumulation.

The impact of parasites on survival varied among the three host species. The amphipod P. fluviatilis was shown to have reduced survival when infected with both C. parvum and M. poulini (Table 4, Figure 1). Individuals with multispecies infections also had a lower survival than individuals with no parasites. Reduced survival incurred by hosts may be due to energetic costs of infection. It has been previously suggested that the negative effect of M. poulini (previously referred to as Microphallus sp.) on P. fluviatilis is a direct consequence of high parasite abundance (Rauque et al., 2011). Our results suggest that parasite-induced mortality may influence differences in parasite abundance between amphipod species rather than size differences between the two host species. Paracorophium excavatum survival was not impacted by the abundance of any of its three parasites (Table 4, Figure 1). The stark contrast in response to similar parasites by the two amphipod species may be due to a difference in the virulence of the parasite between hosts as documented in several prior studies (Jensen, Thomas Jensen, & Mouritsen, 1998; Park, 1948; Thomas et al., 1995). The positive relationship between M. poulini abundance and survival in A. annectens was surprising although it may be an effect of host size (Table 4, Figure 1). The abundance of M. poulini in this isopod may be directly linked to exposure over time and as age is linked to size, a relationship between both is not surprising. Additionally, larger individuals may be able to survive longer, due to a higher resource base to draw upon, therefore creating the appearance of a positive relationship between parasite abundance and survival.

Hosts exhibited different behavioral responses related to the abundance of parasites. Interestingly, P. fluviatilis only demonstrated behavioral changes when infected with A. galaxii. We did not observe behavioral effects of the other parasite species as seen in a previous study (Rauque et al., 2011), and observations of parasite-induced behavioral modification in this amphipod species have been mixed (Poulin, 2001). The inconsistency with the first study could be due to the behavioral aspects measured (phototaxis versus activity levels) (Rauque
et al., 2011). In contrast, P. excavatum movement decreased when infected with an increasing abundance of C. parvum (Table 4, Figure 3). Interestingly, this relationship disappears when the host is co-infected with M. poulini, suggesting an antagonistic relationship, rather than the additive effect of parasites that is often assumed. Previous studies have shown a similar trend, with photophilia increasing with C. parvum and A. galaxii infection but negated by co-infection with M. poulini (Table 4), possibly due to a subtle manipulative effect being impaired by M. poulini (Rauque et al., 2011). The negative relationship between movement and abundance of C. parvum appears nonadaptive for the parasite, as increased movement can increase risk of predation, allowing C. parvum to be passed on to its fish definitive host, mainly the common bully (Gobioomorphus cotidianus). The common bully can use nonvisual methods, such as olfactory, tactile, or lateral-line prey detection, to find prey and the lack of moment in Paracorophium excavatum may allow them to avoid being depredated by this fish (Rowe, 1999; Rowe, Nichols, & Kelly, 2001). The decrease in activity levels may be due to reduced energy available due to parasite infection. Parasites had intriguing impacts on the fecundity and probability of pairing in the various hosts. Female P. fluviatilis and P. excavatum carrying young had a higher abundance of M. poulini. Previous studies have indicated that "handicapped" P. fluviatilis females, that is, with artificially impaired swimming performance, were more likely to be paired (Sutherland, Hogg, & Waas, 2007), and therefore, females with parasites may also have been easier to pair with, explaining the higher likelihood of having young in females with a higher parasite abundance. However, the abundance of both C. parvum and M. poulini was not linked with the probability of male or female P. fluviatilis being found in pairs, contrary to previous studies (Rauque et al., 2011). An alternative hypothesis may suggest that females may choose to invest more into reproduction while infected; therefore, more infected female amphipods would be carrying young (Agnew, Koella, & Michalakis, 2000). Yet, we did not find any relationship between the number of young carried and the abundance of any parasites.

Multispecies infections have important implications for the fitness of individuals and the dynamics of populations. In our study...
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Coitocaecum parvum abundance; (a) distance moved from center versus the abundance of Coitocaecum parvum. Regression lines represent the direction of relationships. All animals were collected were taken during a 5-min behavioral observation. Regression lines in (a) = 63; z = −2.0, p = .049) (b) mobile duration (time moving) of individual compared to their Maritrema poulini abundance in Paracorophium excavatum (n = 84; negative binomial regression, z = −2.0, p = .049). All measures were taken during a 5-min behavioral observation. Regression lines represent the direction of relationships. All animals were collected from Lake Waihola, New Zealand between February and September 2016.

FIGURE 3 Host behavior measures compared to parasite abundance; (a) distance moved from center versus the abundance of Coitocaecum parvum in Paracorophium excavatum (n = 84; negative binomial regression, z = −2.0, p = .049) (b) mobile duration (time moving) of individual compared to their Maritrema poulini abundance in Austridotea annectens (n = 63; z = 2.88, p = .004). All measures were taken during a 5-min behavioral observation. Regression lines represent the direction of relationships. All animals were collected from Lake Waihola, New Zealand between February and September 2016.

co-infections in amphipods; for example, co-infected Gammarus pulex showed a mixed response compared to individuals with single-species infections (Cezilly, Gregoire, & Bertin, 2000). As multispecies infections are more common in P. excavatum than P. fluviatilis and appear to potentially be more important to the impacts of parasites on this host species, these co-infection effects may have impacts on the host population overall.

Our study was based on natural infections rather than an experimental approach thus limiting our interpretations, as we cannot directly address causality or the direct mechanisms of these interactions. However, if combined with what is known of the mechanistic bases of parasite-induced host modifications (see Introduction section), it remains a strong approach for examining the effects of parasites on their hosts (Poulin, 2001).

Interestingly, the effects of parasites on survival and behavior varied greatly between amphipod hosts. Not only does P. excavatum have a much higher abundance and prevalence of M. poulini but there appears to be no decreased survival with a high abundance of either trematode parasite (Table 2). If P. fluviatilis and P. excavatum are competing for the same resources, this may give P. excavatum an advantage. A more tolerant host, such as P. excavatum, may also be able to act as a reservoir for the parasites, maintaining a high level of parasitism within the system (Arneberg et al., 1998). This may increase infection risk for P. fluviatilis with possible consequences for behavior, reproduction, and/or survival. The more tolerant species may become the stronger competitor within the ecosystem, and thus, the parasite could mediate the interaction between the two hosts and alter the outcome of competition (Greenman & Hudson, 2000).

Variation in how parasites affect their different host species has the potential to have community-wide effects. As our species vary in their susceptibility and tolerance to parasites, the presence or absence of a parasite species within a system may dictate the coexistence of species or the extirpation of a particular species (Begon, Bowers, Kadianakis, & Hodgkinson, 1992; Greenman & Hudson, 2000; Hatcher et al., 2006; Hudson, Dobson, & Lafferty, 2006). The differential effects on amphipods and isopods may lead to community-wide effects. Understanding the consequences of parasitic infection and differences between host species is key to gaining greater insight into the role of parasite mediation in ecosystem dynamics.

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CONFLICT OF INTEREST

None declared.
AUTHOR CONTRIBUTIONS

OCF, RP, and CL conceived and designed the experiments. OCF and CL conducted fieldwork. OCF performed the experiments. OCF analyzed the date and wrote the manuscript; other authors provided editorial advice.

REFERENCES

Agnew, P., Koella, J. C., & Michalakis, Y. (2000). Host life history responses to parasitism. Microbes and Infection, 2, 891–896.

Alizon, S. (2013). Co-infection and super-infection models in evolutionary epidemiology. Interface Focus, 3, 20130031.

Alizon, S., de Roode, J. C., & Michalakis, Y. (2013). Multiple infections and the evolution of virulence. Ecology Letters, 16, 556–567.

Arneberg, P., Skorping, A., Grenfell, B., & Read, A. F. (1998). Host densities as determinants of abundance in parasite populations. Proceedings of the Royal Society of London. Series B: Biological Sciences, 265, 1283–1289.

Balmer, O., Stearns, S. C., Schützau, A., & Brun, R. (2009). Intraspecific competition between co-infecting parasite strains enhances host survival in African trypanosomoses. Ecology, 90, 3367–3378.

Bauer, A., Trouvé, S., Grégoire, A., Bollache, L., & Cézilly, F. (2000). Differential influence of Pomphorhynchus laevis (Acanthocephala) on the behaviour of native and invader gammarid species. International Journal for Parasitology, 30, 1453–1457.

Bedhomme, S., Agnew, P., Vital, Y., Sidobre, C., & Michalakis, Y. (2005). Prevalence-dependent costs of parasite virulence. PLoS Biology, 3, e262.

Begon, M., Bowers, R. G., Kadianakis, N., & Hodgkinson, D. E. (1992). Disease and community structure: The importance of host self-regulation in a host–host-parasite model. American Naturalist, 139, 1131–1150.

Bollache, L., Gambade, G., & Cézilly, F. (2001). The effects of two acanthocephalan parasites, Pomphorhynchus laevis and Polymorphus minutus, on pairing success in male Gammarus pulex (Crustacea: Amphipoda). Behavioral Ecology and Sociobiology, 49, 296–303.

Brown, A. F. & Pascoe, D. (1989). Parasitism and host sensitivity to cadmium: An acanthocephalan infection of the freshwater amphipod Gammarus pulex. Journal of Applied Ecology, 26, 473–487.

Cézilly, F., Gregoire, A., & Bertin, A. (2000). Conflict between co-occurring manipulative parasites? An experimental study of the joint influence of two acanthocephalan parasites on the behaviour of Gammarus pulex. Parasitology, 120, 625–630.

Cox, F. E. (2001). Concomitant Infections, parasites, and immune responses. Parasitology, 122(Suppl), S23–S38.

Greenman, J. V., & Hudson, P. J. (2000). Parasite-mediated and direct competition in a two-host shared macroparasite system. Theoretical Population Biology, 57, 13–34.

Grutter, A. S., & Poulin, R. (1998). Intraspecific and interspecific relationships between host size and the abundance of parasitic larval gnathiid isopods on coral reef fishes. Marine Ecology Progress Series, 164, 263–271.

Hansen, E. K., & Poulin, R. (2005). Impact of a microphallid trematode on the behaviour and survival of its isopod intermediate host: Phylogenetec inheritance? Parasitology Research, 97, 242–246.

Hatcher, M. J., Dick, J. T. A. A., & Dunn, A. M. (2006). How parasites affect interactions between competitors and predators. Ecology Letters, 9, 1253–1271.

Hatcher, M. J., & Dunn, A. M. (2011). Parasites and competitors. Parasites in ecological communities: From interactions to ecosystems (1st ed., pp. 20–86). Cambridge: Cambridge University Press.

Hine, P. M. (1977). Acanthocephalus galaxii n.sp. Parasitic in Galaxias maculatus (Jenyns, 1842) in the Waimeha Stream, New Zealand. Journal of the Royal Society of New Zealand, 7, 51–57.

Holt, R. D., & Pickering, J. (1985). Infectious disease and species coexistence: A model of Lotka-Volterra Form. American Naturalist, 126, 196–211.

Hudson, P. J., Dobson, A. P., & Lafferty, K. D. (2006). Is a healthy ecosystem one that is rich in parasites? Trends in Ecology & Evolution, 21, 381–385.

Hughes, W. O. H., & Boomsma, J. J. (2004). Let your enemy do the work: Within-host interactions between two fungal parasitic of leaf-cutting ants. Proceedings of the Royal Society B: Biological Sciences, 271, S104–S106.

Jensen, T., Thomas Jensen, K., & Mouritsen, K. N. (1998). The influence of the trematode Microphallus claviformis on two congenic intermediate host species (Corophium): Infection characteristics and host survival. Journal of Experimental Marine Biology and Ecology, 227, 35–48.

Johnson, K. P., Bush, S. E., & Clayton, D. H. (2005). Correlated evolution of host and parasite body size: Tests of Harrison’s rule using birds and lice. Evolution, 59, 1744–1753.

Kunz, A. K., & Pung, O. J. (2004). Effects of Microphallus turgidus (Trematoda: Microphallidae) on the predation, behavior, and swimming stamina of the grass shrimp Palaeomonetes pugio. Journal of Parasitology, 90, 441–445.

Lagrange, C., & Poulin, R. (2007). Life cycle abbreviation in the trematode Coitocaecum parvum: Can parasites adjust to variable conditions? Journal of Evolutionary Biology, 20, 1189–1195.

Lagrange, C., & Poulin, R. (2008a). Intra- and interspecific competition among helminth parasites: Effects on Coitocaecum parvum life history strategy, size and fecundity. International Journal for Parasitology, 38, 1435–1444.

Lagrange, C., & Poulin, R. (2008b). Lack of seasonal variation in the life-history strategies of the trematode, Coitocaecum parvum: No apparent environmental effect. Parasitology, 135, 1243–1251.

Lagrange, C., Poulin, R., & Keeney, D. B. (2009). Effects of clonality in multiple infections on the life-history strategy of the trematode Coitocaecum parvum in its amphibid intermediate host. Evolution, 63, 1417–1426.

Lange, B., Reuter, M., Ebert, D., Muylaert, K., & Deceaestecker, E. (2014). Diet quality determines interspecific parasite interactions in host populations. Ecology and Evolution, 4, 3093–3102.

Lefebvre, F., Fredensborg, B., Armstrong, A., Hansen, E., & Poulin, R. (2005). Assortative pairing in the amphipod Paracalliope fluviatilis: A role for parasites? Hydrobiologia, 545, 65–73.

Lefèvre, T., Lebarbenchon, C., Gauthier-Clerc, M., Missé, D., Poulin, R., & Thomas, F. (2009). The ecological significance of manipulative parasites. Trends in Ecology and Evolution, 24, 41–48.

Lehmann, T. (1992). Ectoparasite impacts on Gerbillus andersoni allenbyi under natural conditions. Parasitology, 104, 479–488.

Leung, T. L. F., & Poulin, R. (2006). Effects of the trematode Maritrema novaezelandensis on the behaviour of its amphibid host: Adaptive or not? Journal of Helminthology, 80, 271–275.

Luque, J. L., Bannock, L. M., Lagrange, C., & Poulin, R. (2007). Larval Hysterophryum sp. (Nematoda, Anisakidae) and trematode metacerariae from the amphibid Paracorophium excavatum (Corphidae) in New Zealand. Acta Parasitologica, 52, 146–150.

Luque, J. L., Vieira, F. M., Herrmann, K., King, T. M., Poulin, R., & Lagrange, C. (2010). New evidence on a cold case: Trophic transmission, distribution and host-specificity in Hedriris spinigera (Nematoda: Heduridae). Folia Parasitologica, 57, 223–231.

MacFarlane, W. V. (1939). Life cycle of Coitocaecum anaspidis Hickman, a New Zealand digenetic trematode. Parasitology, 31, 172.

Mikheev, V. N., Pasternak, A. F., Taskinen, J., & Valtonen, E. T. (2010). New evidence on a cold case: Trophic transmission, distribution and host-specificity in Hedriris spinigera (Nematoda: Heduridae). Folia Parasitologica, 57, 223–231.

Noldus Information Technology. (2015). EthoVision XT 9.0.

Park, T. (1948). Interspecies competition in populations of Tribolium confusum Duval and Tribolium castaneum Herbst. Ecological Monographs, 18, 265–307.

Pedersen, A. B., & Fenton, A. (2007). Emphasizing the ecology in parasite community ecology. Trends in Ecology and Evolution, 22, 133–139.
FRIESEN, P. W., Westoby, M., & Rice, B. (1988). Parasite-mediated competition: Some predictions and tests. _The American Naturalist_, 131, 554–555.

Price, P. W., Westoby, M., Rice, B., Atsatt, P. R., Fritz, R. S., Thompson, J. N., & Mobley, K. (1986). Parasite mediation in ecological interactions. _Annual Review of Ecology and Systemsatics_, 17, 487–505.

Rauque, C. A., Paterson, R. A., Poulin, R., & Tompkins, D. M. (2011). Do different parasite species interact in their effects on host fitness? A case study on parasites of the amphipod _Paracalliope fluviatilis_. _Parasitology_, 138, 1176–1182.

Read, A. F. (1988). Sexual selection and the role of parasites. _Trends in Ecology & Evolution_, 3, 97–102.

Reisinger, L. S., Petersen, I., Hing, J. S., Davila, R. L., & Lodge, D. M. (2015). Infection with a trematode parasite differentially alters competitive interactions and antipredator behaviour in native and invasive crayfish. _Freshwater Biology_, 60, 1581–1595.

Rivero, A., & Ferguson, H. M. (2003). The energetic budget of _Anopheles stephensi_ infected with _Plasmodium chabaudi_: Is energy depletion a mechanism for virulence? _Proceedings of the Royal Society B: Biological Sciences_, 270, 1365–1371.

de Roode, J. C., Culleton, R., Cheesman, S. J., Carter, R., & Read, A. F. (2004). Host heterogeneity is a determinant of competitive exclusion or coexistence in genetically diverse malaria infections. _Proceedings of the Royal Society B: Biological Sciences_, 271, 1073–1080.

Rowe, D. K. (1999). Factors influencing the abundance of the common bully, _Gobiomorus cotidianus_ Mc Dowall, in small, North Island, New Zealand, lakes. _Fisheries Management and Ecology_, 6, 377–386.

Rowe, D. K., Nichols, S., & Kelly, G. R. (2001). Depth distribution and abundance of the common bully, _Gobiomorus cotidianus_ (Cottidae), in three oligotrophic New Zealand lakes, one of which is turbid. _Environmental Biology of Fishes_, 61, 407–418.

Ruiz-Daniels, R., Beltran, S., Poulin, R., & Lagrue, C. (2012). Do parasites adopt different strategies in different intermediate hosts? Host size, not host species, influences _Coitocaeum parvum_ (Trematoda) life history strategy, size and egg production. _Parasitology_, 140, 1–9.

Saad-Fares, A., & Combes, C. (1992). Abundance/host size relationship in a fish trematode community. _Journal of Helminthology_, 66, 187–192.

SAS Institute Inc. (2015). JMP (R) Version 12.

Shaw, D. J., & Dobson, A. P. (1995). Patterns of macroparasite abundance and aggregation in wildlife populations: A quantitative review. _Parasitology_, 111, S111–S127.

Smith, K. F., Acevedo-Whitehouse, K., & Pedersen, A. B. (2009). The role of infectious diseases in biological conservation. _Animal Conservation_, 12, 1–12.

Sutherland, D. L., Hogg, I. D., & Waas, J. R. (2007). Is size assortative mating in _Paracalliope fluviatilis_ (Crustacea: Amphipoda) explained by male-male competition or female choice? _Biological Journal of the Linnean Society_, 92, 173–181.

Thomas, F., Guegan, J. F., & Renaud, F. (2009). _Ecology and evolution of parasitism_. Oxford, UK: Oxford University Press.

Thomas, F., Renaud, F., Demee, T., & Poulin, R. (1998). Manipulation of host behaviour by parasites: Ecosystem engineering in the intertidal zone? _Proceedings of the Royal Society B: Biological Sciences_, 265, 1091–1096.

Thomas, F., Renaud, F., Roussel, F., Cezilly, F., Meeûs, T. De., & Meeus, T. D. (1995). Differential Mortality of Two Closely Related Host Species Induced by One Parasite. _Proceedings of the Royal Society of London. Series B: Biological Sciences_, 260, 349–352.

Thompson, S. N., Redak, R. A., & Wang, L.-W. (2001). Altered dietary nutrient intake maintains metabolic homeostasis in parasitized larvae of the insect _Manduca sexta_ L. _Journal of Experimental Biology_, 204, 4065–4080.

Thumbi, S. M., Bronsvoort, B. M. D. C., Poole, E. J., Kiara, H., Toye, P. G., Mbole-Kariuki, M. N., ... Woolhouse, M. E. J. (2014). Parasite co-infections and their impact on survival of indigenous cattle (ed TA Smith). _PLoS ONE_, 9, e76324.

Tompkins, D. M., Dunn, A. M., Smith, M. J., & Telfer, S. (2011). Wildlife diseases: From individuals to ecosystems. _Journal of Animal Ecology_, 80, 19–38.

Webster, J. P. (1994). The effect of _Toxoplasma gondii_ and other parasites on activity levels in wild and hybrid _Rattus norvegicus_. _Parasitology_, 109, 583–589.