

Impact of host sex and group composition on parasite dynamics in experimental populations

C. P. TADIRI^{1*}, M. E. SCOTT² and G. F. FUSSMANN¹

¹Department of Biology, McGill University, 1205 Avenue Docteur Penfield, Montreal, QC H3A 1B1, Canada

²Institute of Parasitology and Centre for Host-Parasite Interactions, McGill University (Macdonald Campus), 21,111 Lakeshore Drive, Ste. Anne-de-Bellevue, QC, H9X 3V9, Canada

(Received 13 August 2015; revised 22 December 2015; accepted 29 December 2015; first published online 18 February 2016)

SUMMARY

To better understand the spread of disease in nature, it is fundamentally important to have broadly applicable model systems with readily available species which can be replicated and controlled in the laboratory. Here we used an experimental model system of fish hosts and monogenean parasites to determine whether host sex, group size and group composition (single-sex or mixed-sex) influenced host-parasite dynamics at an individual and group level. Parasite populations reached higher densities and persisted longer in groups of fish compared with isolated hosts and reached higher densities on isolated females than on isolated males. However, individual fish within groups had similar burdens to isolated males regardless of sex, indicating that females may benefit more than males by being in a group. Relative condition was positively associated with high parasite loads for isolated males, but not for isolated females or grouped fish. No difference in parasite dynamics between mixed-sex groups and single-sex groups was detected. Overall, these findings suggest that while host sex influences dynamics on isolated fish, individual fish in groups have similar parasite burdens, regardless of sex. We believe our experimental results contribute to a mechanistic understanding of host-parasite dynamics, although we are cautious about directly extrapolating these results to other systems.

Key words: Epidemic dynamics, host-parasite dynamics, guppies, *Gyrodactylus*.

INTRODUCTION

Infectious diseases are important drivers of ecological interactions and evolution (Boots *et al.* 2009; Schulenburg *et al.* 2009), and are of general concern in the context of disease mitigation and conservation biology (Scott, 1988; Smith *et al.* 2005, 2009). Traditional microparasite models focus on infectious disease from the host point of view by dividing hosts into Susceptible, Infected and Recovered sub-populations (SIR) (Anderson and May, 1979; Grenfell and Harwood, 1997; Hagensars *et al.* 2004; Brooks *et al.* 2008; Ben-Zion *et al.* 2010). Although these models effectively describe epidemics/epizootics of those microparasites for which their numbers per host are irrelevant and/or difficult to quantify, they are less applicable to those microparasites where the size of parasite population within a host is a key to understanding host-parasite population dynamics. Recently, a metapopulation framework has been applied to disease dynamics in order to incorporate spatial structuring of the host population (Arino and Van den Driessche, 2006; Colizza and Vespignani, 2008; Apolloni *et al.* 2014), but in such approaches the unit of the patch is a host population, and the parasite population per host is still overlooked even

though dynamics of infection within a host can be affected by individual host characteristics and can have direct impacts on individual host health, on host movement and on the rate of transmission. Macroparasite models, on the other hand, directly consider the parasite population but even these models often do not capture the dynamics in parasite numbers within individual hosts (May and Anderson, 1979; Rosà *et al.* 2003; Cornell *et al.* 2004). Furthermore, not all parasites fit neatly into the micro- or macroparasite conceptual framework. Together, these limitations have led to the call for a unifying framework which considers both host and parasite populations (Gog *et al.* 2015). One possible approach applies traditional metapopulation theory to parasite population dynamics, but views individual hosts (rather than local host populations) as patches that can be colonized by the parasite (Grenfell and Harwood, 1997). To our knowledge, this approach has not yet been developed theoretically nor investigated experimentally, perhaps because very few parasites allow for the possibility of tracking their dynamics over time without destructive sampling. The use of model systems which can experimentally test how characteristics of individual hosts can influence parasite populations at both the individual host and host population levels are thus of fundamental importance.

Gyrodactylus spp. (Monogenea) are ectoparasites which feed on the epithelial cells and mucus of many marine and freshwater teleost fish species

* Corresponding author. McGill University Stewart Biology Building, 1205 Avenue Docteur Penfield, Room W3/2, Montréal, QC H3A 1B1, Canada. E-mail: christina.tadiri@mail.mcgill.ca

(Bakke *et al.* 2007). They attach to the epidermis of their host via specialized hooks and are directly transmitted primarily by jumping to a new host during contact (Scott and Anderson, 1984; Kearn, 1994). *Gyrodactylus* spp. are viviparous, with an unusual method of reproduction: the developing embryo contains within itself a second developing embryo, which allows for rapid population growth of the parasite on an infected host (Kearn, 1994; Bakke *et al.* 2007). Gyrodactylid infection can result in high rates of mortality (Van Oosterhout *et al.* 2003), and induce a temporary refractory period in surviving hosts (Scott and Robinson, 1984; Scott, 1985a). As such, gyrodactylids cause epidemic outbreaks, making their population dynamics typical of microparasites (Anderson and May, 1979; May and Anderson, 1979) despite being helminth parasites. Furthermore, because they are ectoparasites they can be observed over time without sacrificing the host. Thus this model system has been useful for studying parasite dynamics on individual hosts within a host population (Scott, 1985b; Cable and van Oosterhout, 2007b; Richards *et al.* 2010; Johnson *et al.* 2011; Tadiri *et al.* 2013), and holds potential for furthering our understanding of host-parasite population dynamics.

The guppy (*Poecilia reticulata*) is the host for *Gyrodactylus turnbulli* (see Harris and Lyles, 1992). Guppies are a common sexually dimorphic ovoviparous tropical fish, used as a model species for many ecological studies including exploration of male–female interactions, mate-choice and parasitism (Houde and Torio, 1992; Kolluru *et al.* 2009), and shoaling behaviour (Croft *et al.* 2003; Richards *et al.* 2010). In many guppy populations, females harbour more parasites than males (Gotanda *et al.* 2013; Stephenson *et al.* 2014; Dargent *et al.* 2015), and the tendency of females to shoal more tightly together than males may facilitate parasite transmission especially in grouped female fish (Croft *et al.* 2003; Richards *et al.* 2010). Also, guppy populations vary widely in their ability to resist parasites (Cable and van Oosterhout, 2007b; Dargent *et al.* 2013). Thus, the guppy-gyrodactylid system provides a unique opportunity for experimentally testing how heterogeneity among hosts can influence parasite population dynamics both at the individual host level and at the host and parasite population level. Although the effects of sex and number of guppies on parasite population growth have been studied in separate experiments (Richards *et al.* 2010; Johnson *et al.* 2011; Stephenson *et al.* 2014; Dargent *et al.* 2015), the direct comparison between parasite dynamics on isolated hosts and groups has not been made, nor have the combined effects of grouping and sex on parasite epidemic dynamics been investigated.

The goals for this experiment were to determine whether host sex, group size and group composition

influenced host-parasite dynamics at the level of individual and grouped hosts. We expected parasite populations to reach higher numbers, and persist for longer in groups of fish when compared with isolated fish due to greater availability of hosts. We also expected higher parasite burdens on females than males, both on isolated fish and in single-sex groups due to greater size and possibly lower resistance of females (Dargent *et al.* 2015). For mixed-sex groups, however, our null expectation was that heterogeneity among fish would have an averaging effect on parasite population growth. Although we found that parasites reached higher densities on isolated females than males, this difference did not persist in groups, and heterogeneity in group composition did not influence parasite dynamics.

MATERIALS AND METHODS

Source and maintenance of fish

Animal Care Approval was obtained per McGill University Ethics Guidelines (AUP 2009-5759). Guppies obtained from the Guanapo River and Lower Lalaja tributary in Trinidad (10°38'23"N, 61°14'54"W and 10°39'14"N, 61°15'18"W) were bred to the F3 generation, keeping track of maternal lines, in the McGill University Phytotron. The room was maintained at 27 ± 1 °C with a 12-h light-dark cycle and the fish were raised in common-garden conditions in an Aquaneering Inc. (San Diego, California, USA) flow-through system. Fish were raised on controlled amounts of TetraMin® Tropical Flakes (Tetra Werke, Melle Germany). In order to mimic a history of natural infection, F3 fish were exposed to our isogenic laboratory culture of *G. turnbulli* (identified by S. King) from birth.

Experimental design

The experiment consisted of two parts, conducted simultaneously. The first part was a 2×2 factorial design used to test the effects of host sex (male *vs* female) and host group size (1 *vs* 8) on parasite dynamics. As treatments, we established groups of 8 males (4 replicates), groups of 8 females (4 replicates), isolated males (8 replicates), and isolated females (8 replicates). The second part of the experiment tested the effect of host heterogeneity in sex on parasite dynamics. This part consisted of 4 replicates each containing a group of 8 fish (4 males and 4 females), and data were compared with the homogenous sex groups from the first part of the experiment.

Experimental protocol

In order for fish to overcome infection-acquired resistance and regain susceptibility to *Gyrodactylus*

spp. (Scott and Robinson, 1984; Scott, 1985a; Cable and van Oosterhout, 2007b) parasites were eliminated from adult F3 fish by treating them in a 25 g L⁻¹ salt water bath for 15 min (Schelkle *et al.* 2011) 2 months before the start of the experiment. One week later, fish were anaesthetized in 0.02% Tricaine methanesulfonate (MS-222), buffered to a neutral pH with sodium bicarbonate and scanned using a dissection microscope to confirm the absence of parasites. Seven weeks later, adult F3 fish were again scanned for parasites and weighed to the nearest 0.001 g, measured for standard length (SL) to the nearest 0.01 cm with a calliper, and marked for identification with visible implant elastomer dye (Northwest Marine Technologies Inc., Shaw Island Washington, USA) which has been shown to have no impact on fish health or behaviour (Croft *et al.* 2003, 2004). Fish were then assigned to treatments/replicates in a way that would distribute size, population of origin and maternal lines evenly across treatments/replicates and groups of fish were acclimated with one another for 1 week prior to infection.

A total of 112 fish (56 males and 56 females) were used for this experiment, with an SL of 2.34 ± 0.03 cm for females and 1.61 ± 0.01 cm for males and weights of 0.29 ± 0.01 g for females and 0.08 ± 0.002 g for males. Each group of 8 fish was housed in a tank with 6 L of water and each isolated fish was housed in a tank with 1.8 L of water. Each tank was considered an experimental unit for analyses at the population level. Fish were fed daily with TetraMin[®] Tropical Flakes mixed with conditioned water into a paste and delivered through a glass precision syringe to each tank according to the number and sex of fish in each tank. A low food availability regime was used to prevent compensation of innate resistance through additional food acquisition (Kolluru *et al.* 2006; Tadiri *et al.* 2013).

To begin infections on isolated fish, a heavily infected fish was taken from our isogenic lab culture of *G. turnbulli* and anaesthetized in 0.02% MS-222. Scales with parasites were removed from the donor fish and placed on an anaesthetized recipient until 3 parasites had transferred to the recipient fish (Scott, 1982). To introduce infection to a group of fish, a juvenile pet-store guppy (sex undetermined) from a naïve laboratory stock was infected with 3 parasites as above and added to the experimental tank for 4 or 6 days when 3 parasites had naturally transferred to the experimental fish in the group, at which time the juvenile pet-store guppy was removed (defined as 'Day 0' for each tank). This procedure eliminated the potential bias that might have occurred by initiating the epidemic on a male or a female in the mixed groups.

Parasites on each fish were counted every second day for 36 days or until no parasites were found in

a tank on two consecutive counting days. In groups of fish, the first day of infection was noted separately for each group (Day 0 in all cases) and for each individual within the group, based on the day that it was first infected. If a fish in a group died, it was left in the tank for one day in order to allow transmission to other guppies (Scott and Anderson, 1984; Gheorghiu *et al.* 2007) and then removed.

Independent variables

Our independent test variables were sex (male *vs* female), group size (isolated *vs* grouped), and group composition (homogenous *vs* heterogeneous).

In addition, to account for variability in the size of fish at the beginning of the experiment (Cable and van Oosterhout, 2007a; Tadiri *et al.* 2013), we calculated the relative condition index (K_n) of each guppy based on its weight (W) and SL relative to all other fish of the same sex in the experiment. For each sex, a least squares regression of $\text{Log}(SL)$ and $\text{Log}(W)$ was performed, and the slope (b) and intercept ($\log(a)$) for the line of best fit were obtained. K_n was then calculated for each individual fish as $K_n = W/(a \times SL^b)$ (Le Cren, 1951; Peig and Green, 2010) using the sex-specific parameters. Average K_n was also calculated for each group of fish.

Definition and calculation of dependent variables

Peak parasite burden (maximum number of *G. turnbulli*), time to peak parasite burden, persistence of infection (last day of infection minus first day of infection) and host mortality were recorded for isolated fish, for each individual in a group, and for the population of grouped fish. In addition, asynchrony in when individual fish within groups became infected was recorded as the delay from when infection was introduced into the population. Maximum prevalence (per cent of infected fish in groups) and time to maximum prevalence over the course of the experiment were also recorded for groups of fish.

Statistical analysis

All analyses were done using R Language and Environment for Statistical Computing version 3.1.0 (R Development Core Team, 2014). Generalized Linear Mixed-Effects Models (GLMMs) were constructed to determine the effects of host sex, host group size (isolated *vs* group of 8), fish size (either W and SL or K_n , and average of the group for group-level response variables) and group composition (homogeneous or heterogeneous) and the interactions thereof on host mortality, peak parasite burden, time to peak and persistence on isolated fish, on individuals in groups and in the group as whole. For each response variable, models were

fitted to the distribution of the variable and models for individual fish-level response variables were nested within the random variable tank. Models using *SL* and *W* as metrics for size were not significant, so all final models used only *Kn*. Final models were produced using the stepAIC function to select the combination of factors which produced a model with the lowest Akaike information criterion (AIC). In all cases, the level of significance was set at $P < 0.05$, and all values reported are means and standard errors.

RESULTS

Basic parasite dynamics

A total of 28 fish (13.2%) died over the course of the experiment, and mortality did not significantly differ between group sizes ($P = 0.271$) or between sexes ($P = 0.433$).

In all but two grouped tanks, parasites reached 100% prevalence within 14 days as additional fish became infected asynchronously (Fig. 1b, c, e, and f). In tanks, parasite numbers increased and reached distinct population peaks (Fig. 2). The rate at which fish became infected (delay to infection) did not significantly differ among groups (data not shown). Group composition (females, males, mixed sex) had no impact on peak prevalence, time to infection or time to peak prevalence (data not shown).

Table 1 gives a full overview of the outcomes of our GLMMs and results are explained below in detail.

Individual vs grouped fish

Peak total parasite population on groups of fish was higher (123.5 ± 40.0) than on isolated fish (26.4 ± 6.0) ($P < 0.001$). Parasite populations also persisted longer ($P = 0.001$) on groups of fish (24.5 ± 1.3 days) than on isolated fish (17.0 ± 1.5 days). Overall, isolated fish had lower peak burdens than individual fish in groups ($P = 0.015$), but there was an interaction between sex and grouping, with isolated females having higher peak burdens (34.9 ± 10.1) than individual females within groups (17.5 ± 2.6) (Fig. 3). No difference in parasite time to peak or persistence on an individual fish was found between isolated fish or individual fish in single-sex or mixed groups.

There was a significant interaction of *Kn* and grouping (isolated *vs* in a group of 8) ($P = 0.01$), with the effect of *Kn* on parasite burden being stronger on isolated fish than on individual fish within single-sex or mixed groups (Fig. 4).

Male vs female hosts

Parasites reached higher peak burdens on isolated females (34.9 ± 10.1) than on males (15.4 ± 5.0)

($P = 0.006$). There was an interaction of sex and *Kn* ($P = 0.038$) on peak parasite burden both for isolated fish and individual fish in groups, with *Kn* having a positive impact on parasite load for males but not for females (Fig. 4). Parasite numbers peaked later ($P = 0.001$) (Fig. 3) on females (9.4 ± 0.7 days) than on males (6.6 ± 0.5 days) and the infection persisted longer ($P = 0.033$) on females (17.4 ± 0.9 days) than on males (13.6 ± 0.7 days). Infection also persisted longer on fish with a higher *Kn* ($P = 0.0479$), regardless of sex.

At the group level, there was no difference in time to peak prevalence, parasite population peak burden, time to peak population burden or parasite persistence in a tank between male and female groups.

Group composition: single-sex vs mixed sex groups

We found no differences between mixed-sex groups and single-sex groups (or individual fish within them) for any of the response variables.

DISCUSSION

Our investigation of parasite dynamics on isolated (single host patch) and grouped (multiple host patches) fish confirms that metapopulation theory is compatible with our model system (Grenfell and Harwood, 1997; Hanski, 1999), as the presence of multiple patches and connectivity among them allowed the parasite total population to grow larger and persist longer than on single isolated fish. There was no difference in time between when fish first became infected and when parasite burden peaked or in duration of infection between isolated fish and individual fish in a group, but time to peak parasite numbers in the tank and duration of infection in the tank were prolonged in groups compared with isolated fish. In this aspect, dynamics on each fish were similar but occurred asynchronously due to consecutive infection, leading to longer persistence of the overall parasite populations. We found that fish characteristics in the form of sex and *Kn* impacted parasite dynamics in isolation, but that these differences were not observed in grouped fish.

Although peak parasite total populations were higher on groups than on isolated fish, they were not 8 times higher, and the existence of additional hosts lowered the average parasite burden per fish for female hosts. The addition of multiple hosts presumably provided the parasite with more options if their host mounted an immune response, died, or became overcrowded with parasites (Bagge *et al.* 2004), and thus allowed it to reach a population growth rate closer to the parasite's innate reproductive potential. However, parasite population growth and dispersal were likely constrained due to trade-offs between carrying capacity, reproductive

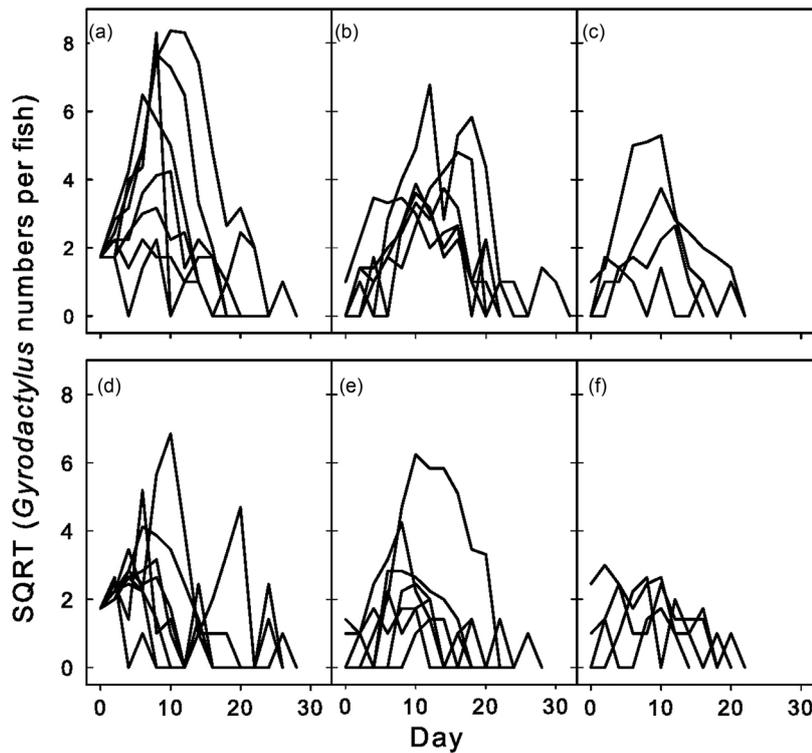


Fig. 1. Parasite population dynamics on individual isolated females (a), individual females in a sample all-female tank (b), and individual females in a sample mixed-sex tank (c), as well as individual isolated males (d), individual males in a sample all-male tank (e) and individual males in a sample mixed-sex tank (f). Data are square-root transformed for graphing purposes, but were not transformed for analysis. For all panels, 'Day 0' indicates the day on which at least 3 parasites were first found in the tank.

potential and the cost of migrating. Our study would indicate that the costs of transmission and the parasite's own reproductive potential may have had a greater impact on parasite dynamics than overall quality of the host (carrying capacity). Of course, these inferences are limited by the fact that our epidemics were run in a highly controlled, experimental setting and began with only 3 parasites. It is possible that host abundance and sex could have a greater impact if more parasites had been introduced.

Consistent with our hypothesis, parasites reached higher burdens and persisted longer on isolated female guppies compared with isolated male guppies. One reason could be that females from the populations we used have been shown to be less resistant to parasites than males (Dargent *et al.* 2015). Another reason could be that the larger size of females compared with males provided more resources for the parasite in terms of food, space and ability to move to another region of the host to avoid local defence reactions (Poulin and Rohde, 1997). Previous work has shown that larger guppies harbour more gyrodactylids than smaller ones (Cable and van Oosterhout, 2007a) and that the parasites disperse more rapidly through a group of fish when introduced on a fish with a higher Kn (Tadiri *et al.* 2013). In this study, we found a positive relationship between Kn and peak

parasite burden on isolated males, but not on isolated females (which were overall larger than males). However, despite differences in parasite dynamics between the sexes observed at the individual level, we did not find a difference in parasite burden between individual grouped males and grouped females, nor did we find any effect of Kn on parasite burden for grouped fish, indicating that there was also an effect of group size on individual burden.

In contrast to previous reports of higher transmission in female than male groups (Richards *et al.* 2010) and higher transmission in male than female groups (Richards *et al.* 2012), we did not observe any differences in peak prevalence, time to first infection or time to peak prevalence between our single-sex groups. In both previous studies, the measure of transmission was the number of non-focal fish that became infected within 3 days of introduction of a focal fish infected with either 30 (Richards *et al.* 2012) or 100 gyrodactylids (Richards *et al.* 2010). This contrasts with our protocol in that we explored transmission from an initial population of 3 parasites to the time of peak prevalence in populations of smaller feeder guppies at higher density compared with the larger ornamental guppies kept at lower density. Richards *et al.* (2012) suggested that transmission may be a function of initial parasite load and the impact it has on shoaling

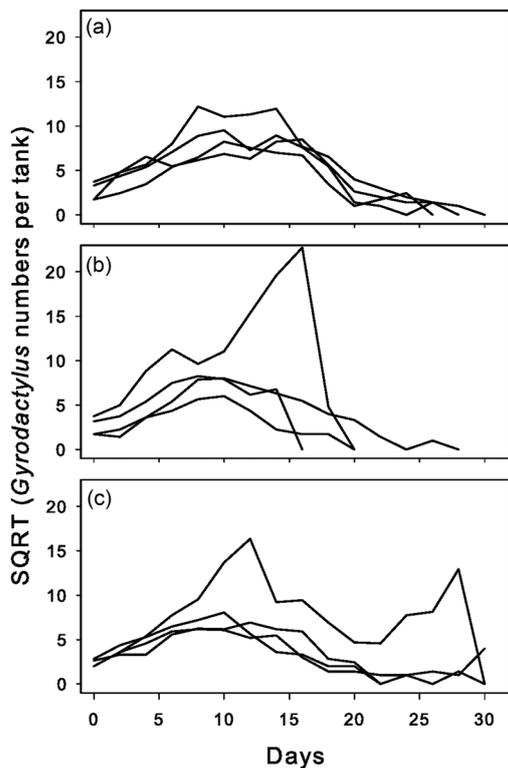


Fig. 2. Total parasite population numbers over the course of the experiment in group tanks for male groups (a), female groups (b) and mixed-sex groups (c). Data are square-root transformed for graphing purposes, but were not transformed for analysis. For all panels, 'Day 0' indicates the day on which at least 3 parasites were first found in the tank.

behaviour or courtship displays but given the number of differences between our experiment and the two previous studies, it is difficult to attribute the different findings to a single factor.

We found that parasites peaked earlier on males than females, despite having similar burdens in groups. One possibility for the lower parasite growth rate in grouped females could be that females increase investment in parasite resistance (rather than growth) when grouped at a high density, where infection is more likely to occur, an effect observed in many invertebrate systems (Wilson and Cotter, 2008), and potentially also in ours (Pérez-Jvostov *et al.* 2015). While we did not find a significant difference in somatic growth between isolated and grouped females as Pérez-Jvostov *et al.* (2015) did, this could have been an issue of power, since there were only 8 isolated females and changes in weight were much less drastic than differences in parasite loads.

We also found no effect of group composition (homogenous *vs* heterogeneous) on parasite dynamics, as our mixed-sex groups did not differ from all-male or all-female groups, nor did individuals within these groups. This finding is inconsistent with theoretical work that suggests heterogeneity would promote asynchrony in local population dynamics

and therefore parasite persistence (Hagenaars *et al.* 2004; Singh *et al.* 2004; Colizza and Vespignani, 2008). However, since parasite dynamics were similar between single-sex groups of males and females in our study, mixing the sexes in our system may not have generated the heterogeneity in individual hosts that we had expected and can thus explain why we found no influence of heterogeneity on parasite dynamics. Similar results have also been reported in mice (Scott, 1991), where grouping susceptible and resistant strains together resulted in similar nematode burdens among mice of both strains, but that increasing transmission rates effected a distinction between the two strains (Scott, 2006). However, those studies did not investigate parasite dynamics in single-strain groups, and our results indicate that grouping, rather than group composition, has the greatest impact in homogenizing parasite dynamics.

Although this study set out with the intention of determining how host heterogeneity may influence parasite population dynamics, we found that group composition and factors which influenced parasite dynamics on fish in isolation (Kn and sex) had almost no effect on parasite dynamics on fish in groups or at the group level. These findings indicate that factors associated with grouping fish become more relevant than the effects of the individual host characteristics sex and Kn of individual hosts for both individual and group-level outcomes, but we are cautious about over-generalizing these interpretations, given that our study comes with the limitations of using a specific experimental system.

Our ability to detect some biologically important differences may have been limited by having only 4 replicates per treatment. The relatively small size of the fish tanks probably limited our ability to detect differences in parasite dynamics that would have been driven by host behaviours including shoaling of females but not males. We did not know the infection history of individual fish, other than the fact that they had been previously exposed to parasites, and as such could not explore any possible impact of differences in acquired resistance to parasites (Scott and Robinson, 1984; Scott, 1985a; Richards and Chubb, 1998) or of an interaction between sex and acquired resistance. Finally, this study only looked at two host traits (sex and size) and it is possible that other host characteristics, such as Major Histocompatibility Complex (MHC) profiles (Fraser *et al.* 2009), colour (Houde and Torio, 1992) or population of origin (Van Oosterhout *et al.* 2003; Dargent *et al.* 2013), could have a stronger impact on parasite dynamics.

Metapopulation theory, while compatible in our system in the sense that additional hosts allowed for asynchronous dynamics to promote parasite persistence, predicts that heterogeneity in patch quality prolongs persistence due to greater asynchrony in

Table 1. Outputs of generalized linear mixed-effects models for response variables. For two-way comparisons, the comparison is explained in parentheses next to the variable name

	Estimate (\pm S.E.)	<i>z</i> -value	<i>P</i> -value
(a) Outcome: parasite peak burden on individual patches (111 D.F.)			
Group size (isolated <i>vs</i> grouped)	-30.8 (\pm 12.9)	-2.392	0.015
Sex (male <i>vs</i> female)	-4.8 (\pm 1.7)	-2.754	0.006
<i>Kn</i> ^a	-1.8 (\pm 1.1)	-1.536	0.124
Group size \times sex	34.7 (\pm 19.3)	1.798	0.072
Sex \times <i>Kn</i>	3.7 (\pm 1.8)	2.076	0.038
Group size \times <i>Kn</i>	29.4 (\pm 12.2)	2.411	0.016
Sex \times group size \times <i>Kn</i>	-33.0 (\pm 18.2)	-1.809	0.070
(b) Outcome: parasite peak burden in tanks (27 D.F.)			
Group size (isolated <i>vs</i> grouped)	-2.3 (\pm 0.5)	-4.717	<0.001
Sex (males <i>vs</i> females)	0.6 (\pm 0.5)	1.1	0.300
Sex (females <i>vs</i> mixed)	-0.1 (\pm 0.5)	-0.175	0.861
Sex (males <i>vs</i> mixed)	0.5 (\pm 0.5)	0.905	0.366
Group size \times sex	1.2 (\pm 0.7)	1.855	0.064
(c) Outcome: time to peak burden on individual patches (110 D.F.)			
Sex (males <i>vs</i> females)	-2.8 (\pm 0.9)	-3.241	0.002
(d) Outcome: parasite persistence on individual patches (111 D.F.)			
Sex (males <i>vs</i> females)	25.6 (\pm 11.9)	2.151	0.033
<i>Kn</i>	22.4 (\pm 7.8)	2.881	0.048
Sex \times <i>Kn</i>	-26.7 (\pm 15.5)	-1.722	0.089
(e) Outcome: parasite persistence in tanks (27 D.F.)			
Group size (isolated <i>vs</i> grouped)	-7.5 (\pm 2.1)	-3.574	0.001

^a *Kn* is the relative condition index based on weight (*W*) and standard length (*SL*) of each fish relative to all other fish of the same sex in the experiment. See section Materials and Methods for calculation.

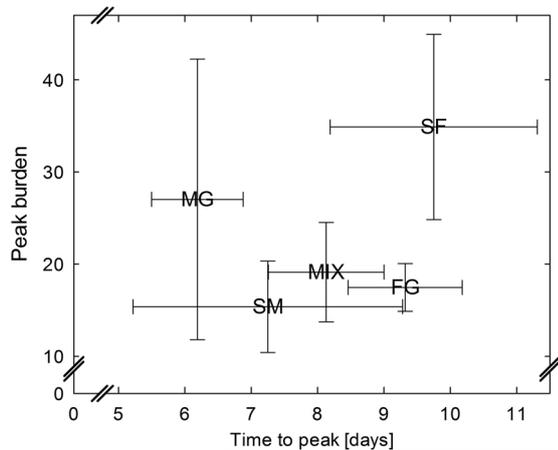


Fig. 3. Mean peak burden (\pm S.E.) *vs* mean time to peak (\pm S.E.) for individual fish in each treatment. Abbreviations: SF, single (isolated) females, SM, single (isolated) males, FG, female groups, MG, male groups, MIX, mixed groups.

local patch dynamics (Dennis and Eales, 1997; Thomas *et al.* 2001; Fleishman *et al.* 2002; Bonte *et al.* 2003; Schooley and Branch, 2007; Franzén and Nilsson, 2010). Our study has shown that the ability of a parasite to move from host to host (connectivity) may override individual host differences in the absence of connectivity, thus rendering the expectation of persistence over heterogeneous patches weaker for our system. This study served as the first step towards conceptualizing a theory that incorporates dynamics within individual hosts

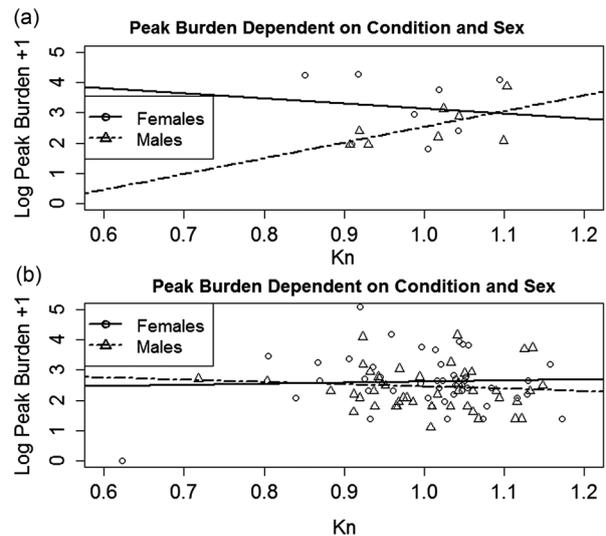


Fig. 4. Interaction of *Kn* and sex for isolated (A) and grouped (B) fish. Log(peak burden + 1) used for graphing purposes, but not for statistical analysis.

rather than focusing solely on infection status of individuals (like microparasite models) or the total parasite populations (like macroparasite models), and further investigation into these dynamics is necessary to develop a more unifying framework for parasite population growth and dissemination.

ACKNOWLEDGEMENTS

We would like to thank Stanley King for identification of the parasite. We would also like to thank Mark Romer and

Claire Cooney for the management of the McGill Phytotron.

FINANCIAL SUPPORT

Funding for this research was provided by a Fonds de recherche du Québec – Nature et technologies (FQRNT) grant (57516) and National Sciences and Engineering Research Council (NSERC) Discovery grants were awarded to M. E. S. and G. F. F. Research at the Institute of Parasitology is supported by a FQRNT regroupement grant to the Centre for Host–Parasite Interactions.

REFERENCES

- Anderson, R. M. and May, R. M.** (1979). Population biology of infectious diseases: Part I. *Nature* **280**, 361–367.
- Apolloni, A., Poletto, C., Ramasco, J. J., Jensen, P. and Colizza, V.** (2014). Metapopulation epidemic models with heterogeneous mixing and travel behaviour. *Theoretical Biology and Medical Modelling* **11**, 3.
- Arino, J. and Van den Driessche, P.** (2006). Disease spread in metapopulations. *Nonlinear Dynamics and Evolution Equations* **48**, 1–13.
- Bagge, A. M., Poulin, R. and Valtonen, E. T.** (2004). Fish population size, and not density, as the determining factor of parasite infection: a case study. *Parasitology* **128**, 305–313.
- Bakke, T. A., Cable, J. and Harris, P. D.** (2007). The biology of gyrodactylid monogeneans: the ‘russian-doll killers’. In *Advances in Parasitology*, Vol. **64** (ed. J. R. Baker, R. M. and Rollinson, D.), pp. 161–376, 459–460. Academic Press, Cambridge, MA.
- Ben-Zion, Y., Cohen, Y. and Shnerb, N. M.** (2010). Modeling epidemics dynamics on heterogeneous networks. *Journal of Theoretical Biology* **264**, 197–204.
- Bonte, D., Lens, L., Maelfait, J.-P., Hoffmann, M. and Kuijken, E.** (2003). Patch quality and connectivity influence spatial dynamics in a dune wolfspider. *Oecologia* **135**, 227–233.
- Boots, M., Best, A., Miller, M. R. and White, A.** (2009). The role of ecological feedbacks in the evolution of host defence: what does theory tell us? *Philosophical Transactions of the Royal Society B: Biological Sciences* **364**, 27–36.
- Brooks, C. P., Antonovics, J. and Keitt, T. H.** (2008). Spatial and temporal heterogeneity explain disease dynamics in a spatially explicit network model. *American Naturalist* **172**, 149–159.
- Cable, J. and van Oosterhout, C.** (2007a). The impact of parasites on the life history evolution of guppies (*Poecilia reticulata*): the effects of host size on parasite virulence. *International Journal for Parasitology* **37**, 1449–1458.
- Cable, J. and van Oosterhout, C.** (2007b). The role of innate and acquired resistance in two natural populations of guppies (*Poecilia reticulata*) infected with the ectoparasite *Gyrodactylus turnbulli*. *Biological Journal of the Linnean Society* **90**, 647–655.
- Colizza, V. and Vespignani, A.** (2008). Epidemic modeling in metapopulation systems with heterogeneous coupling pattern: theory and simulations. *Journal of Theoretical Biology* **251**, 450–467.
- Cornell, S. J., Isham, V. S. and Grenfell, B. T.** (2004). Stochastic and spatial dynamics of nematode parasites in farmed ruminants. *Proceedings of the Royal Society of London B: Biological Sciences* **271**, 1243–1250.
- Croft, D., Krause, J. and James, R.** (2004). Social networks in the guppy (*Poecilia reticulata*). *Proceedings of the Royal Society of London B: Biological Sciences* **271**(Suppl 6), S516–S519.
- Croft, D. P., Albanese, B., Arrowsmith, B. J., Botham, M., Webster, M. and Krause, J.** (2003). Sex-biased movement in the guppy (*Poecilia reticulata*). *Oecologia* **137**, 62–68.
- Dargent, F., Scott, M. E., Hendry, A. P. and Fussmann, G. F.** (2013). Experimental elimination of parasites in nature leads to the evolution of increased resistance in hosts. *Proceedings of the Royal Society of London B: Biological Sciences* **280**. doi: 10.1098/rspb.2013.2371.
- Dargent, F., Rolshausen, G., Hendry, A. P., Scott, M. E. and Fussmann, G. F.** (2015). Parting ways: parasite release in nature leads to sex-specific evolution of defence. *Journal of Evolutionary Biology* **29**, 23–24.
- Dennis, R. L. H. and Eales, H. T.** (1997). Patch occupancy in *Coenonympha tullia* (Muller, 1764) (Lepidoptera: Satyrinae): habitat quality matters as much as patch size and isolation. *Journal of Insect Conservation* **1**, 167–176.
- Fleishman, E., Ray, C., Sjögren-Gulve, P., Boggs, C. L. and Murphy, D. D.** (2002). Assessing the roles of patch quality, area, and isolation in predicting metapopulation dynamics. *Conservation Biology* **16**, 706–716.
- Franzén, M. and Nilsson, S. G.** (2010). Both population size and patch quality affect local extinctions and colonizations. *Proceedings of the Royal Society of London B: Biological Sciences* **277**, 79–85.
- Fraser, B. A., Ramnarine, I. W. and Neff, B. D.** (2009). Selection at the MHC class IIB locus across guppy (*Poecilia reticulata*) populations. *Heredity* **104**, 155–167.
- Gheorghiu, C., Cable, J., Marcogliese, D. J. and Scott, M. E.** (2007). Effects of waterborne zinc on reproduction, survival and morphometrics of *Gyrodactylus turnbulli* (Monogenea) on guppies (*Poecilia reticulata*). *International Journal for Parasitology* **37**, 375–381.
- Gog, J. R., Pellis, L., Wood, J. L. N., McLean, A. R., Arinaminpathy, N. and Lloyd-Smith, J. O.** (2015). Seven challenges in modeling pathogen dynamics within-host and across scales. *Epidemics* **10**, 45–48.
- Gotanda, K., Delaire, L., Raeymaekers, J. M., Pérez-Jvostov, F., Dargent, F., Bentzen, P., Scott, M., Fussmann, G. and Hendry, A.** (2013). Adding parasites to the guppy-predation story: insights from field surveys. *Oecologia* **172**, 155–166.
- Grenfell, B. and Harwood, J.** (1997). (Meta)population dynamics of infectious diseases. *Trends in Ecology and Evolution* **12**, 395–399.
- Hagenaars, T. J., Donnelly, C. A. and Ferguson, N. M.** (2004). Spatial heterogeneity and the persistence of infectious diseases. *Journal of Theoretical Biology* **229**, 349–359.
- Hanski, I.** (1999). *Metapopulation Ecology*. Oxford University Press, Oxford.
- Harris, P. D. and Lyles, A. M.** (1992). Infections of *Gyrodactylus bullatarudis* and *Gyrodactylus turnbulli* on guppies (*Poecilia reticulata*) in Trinidad. *Journal of Parasitology* **78**, 912–914.
- Houde, A. E. and Torio, A. J.** (1992). Effect of parasitic infection on male color pattern and female choice in guppies. *Behavioral Ecology* **3**, 346–351.
- Johnson, M. B., Lafferty, K. D., van Oosterhout, C. and Cable, J.** (2011). Parasite transmission in social interacting hosts: monogenean epidemics in guppies. *PLoS ONE* **6**, e22634.
- Kearn, G. C.** (1994). Evolutionary expansion of the Monogenea. *International Journal for Parasitology* **24**, 1227–1271.
- Kolluru, G. R., Grether, G. F., South, S. H., Dunlop, E., Cardinali, A., Liu, L. and Carapiet, A.** (2006). The effects of carotenoid and food availability on resistance to a naturally occurring parasite (*Gyrodactylus turnbulli*) in guppies (*Poecilia reticulata*). *Biological Journal of the Linnean Society* **89**, 301–309.
- Kolluru, G. R., Grether, G. F., Dunlop, E. and South, S. H.** (2009). Food availability and parasite infection influence mating tactics in guppies (*Poecilia reticulata*). *Behavioral Ecology* **20**, 131–137.
- Le Cren, E. D.** (1951). The length-weight relationship and seasonal cycle in gonad weight and condition in the perch (*Perca fluviatilis*). *Journal of Animal Ecology* **20**, 201–219.
- May, R. M. and Anderson, R. M.** (1979). Population biology of infectious diseases: Part II. *Nature* **280**, 455–461.
- Peig, J. and Green, A. J.** (2010). The paradigm of body condition: a critical reappraisal of current methods based on mass and length. *Functional Ecology* **24**, 1323–1332.
- Pérez-Jvostov, F., Hendry, A. P., Fussmann, G. F. and Scott, M. E.** (2015). An experimental test of antagonistic effects of competition and parasitism on host performance in semi-natural mesocosms. *Oikos*. doi: 10.1111/oik.02499.
- Poulin, R. and Rohde, K.** (1997). Comparing the richness of metazoan ectoparasite communities of marine fishes: controlling for host phylogeny. *Oecologia* **110**, 278–283.
- R Development Core Team** (2014). *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria.
- Richards, E. L., van Oosterhout, C. and Cable, J.** (2010). Sex-specific differences in shoaling affect parasite transmission in guppies. *PLoS ONE* **5**, e13285.
- Richards, E. L., van Oosterhout, C. and Cable, J.** (2012). Interactions between males guppies facilitates the transmission of the monogenean ectoparasite *Gyrodactylus turnbulli*. *Experimental Parasitology* **132**, 483–486.
- Richards, G. R. and Chubb, J. C.** (1998). Longer-term population dynamics of *Gyrodactylus bullatarudis* and *G. turnbulli* (Monogenea) on adult guppies *Poecilia reticulata* in 50-l experimental arenas. *Parasitology Research* **84**, 753–756.
- Rosà, R., Pugliese, A., Villani, A. and Rizzoli, A.** (2003). Individual-based vs. deterministic models for macroparasites: host cycles and extinction. *Theoretical Population Biology* **63**, 295–307.

- Schelkle, B., Doetjes, R. and Cable, J.** (2011). The salt myth revealed: treatment of gyrodactylid infections on ornamental guppies, *Poecilia reticulata*. *Aquaculture* **311**, 74–79.
- Schooley, R. and Branch, L.** (2007). Spatial heterogeneity in habitat quality and cross-scale interactions in metapopulations. *Ecosystems* **10**, 846–853.
- Schulenburg, H., Kurtz, J., Moret, Y. and Siva-Jothy, M. T.** (2009). Introduction. Ecological immunology. *Philosophical Transactions of the Royal Society B: Biological Sciences* **364**, 3–14.
- Scott, M. E.** (1982). Reproductive potential of *Gyrodactylus bullatarudis* (Monogenea) on guppies (*Poecilia reticulata*). *Parasitology* **85**, 217–236.
- Scott, M. E.** (1985a). Dynamics of challenge infections of *Gyrodactylus bullatarudis* Turnbull (Monogenea) on guppies, *Poecilia reticulata* (Peters). *Journal of Fish Diseases* **8**, 495–503.
- Scott, M. E.** (1985b). Experimental epidemiology of *Gyrodactylus bullatarudis* (Monogenea) on guppies (*Poecilia reticulata*): short- and long-term studies. In *Ecology and Genetics of Host-Parasite Interactions* (ed. Rollinson, D. and Anderson, R. M.), pp. 21–38. Academic Press, New York.
- Scott, M. E.** (1988). The impact of infection and disease on animal populations: implications for conservation biology. *Conservation Biology* **2**, 40–56.
- Scott, M. E.** (1991). Heligmosomoides polygyrus (Nematoda): susceptible and resistant strains of mice are indistinguishable following natural infection. *Parasitology* **103**, 429–438.
- Scott, M. E.** (2006). High transmission rates restore expression of genetically determined susceptibility of mice to nematode infections. *Parasitology* **132**, 669–679.
- Scott, M. E. and Anderson, R. M.** (1984). The population dynamics of *Gyrodactylus bullatarudis* (Monogenea) within laboratory populations of the fish host *Poecilia reticulata*. *Parasitology* **89**, 159–194.
- Scott, M. E. and Robinson, M. A.** (1984). Challenge infections of *Gyrodactylus bullatarudis* (Monogenea) on guppies, *Poecilia reticulata* (Peters), following treatment. *Journal of Fish Biology* **24**, 581–586.
- Singh, B. K., Rao, J. S., Ramaswamy, R. and Sinha, S.** (2004). The role of heterogeneity on the spatiotemporal dynamics of host–parasite metapopulation. *Ecological Modelling* **180**, 435–443.
- Smith, K. F., Dobson, A. P., McKenzie, F. E., Real, L. A., Smith, D. L. and Wilson, M. L.** (2005). Ecological theory to enhance infectious disease control and public health policy. *Frontiers in Ecology and the Environment* **3**, 29–37.
- Smith, K. F., Acevedo-Whitehouse, K. and Pedersen, A. B.** (2009). The role of infectious diseases in biological conservation. *Animal Conservation* **12**, 1–12.
- Stephenson, J. F., van Oosterhout, C., Mohammed, R. S. and Cable, J.** (2014). Parasites of Trinidadian guppies: evidence for sex- and age-specific trait-mediated indirect effects of predators. *Ecology* **96**, 489–498.
- Tadiri, C. P., Dargent, F. and Scott, M. E.** (2013). Relative host body condition and food availability influence epidemic dynamics: a *Poecilia reticulata*–*Gyrodactylus turnbulli* host–parasite model. *Parasitology* **140**, 343–351.
- Thomas, J. A., Bourn, N. A. D., Clarke, R. T., Stewart, K. E., Simcox, D. J., Pearman, G. S., Curtis, R. and Goodger, B.** (2001). The quality and isolation of habitat patches both determine where butterflies persist in fragmented landscapes. *Proceedings of the Royal Society of London B: Biological Sciences* **268**, 1791–1796.
- Van Oosterhout, C., Harris, P. D. and Cable, J.** (2003). Marked variation in parasite resistance between two wild populations of the Trinidadian guppy, *Poecilia reticulata* (Pisces: Poeciliidae). *Biological Journal of the Linnean Society* **79**, 645–651.
- Wilson, K. and Cotter, S. C.** (2008). Density-dependent prophylaxis in insects. *Phenotypic Plasticity of Insects: Mechanisms and Consequences* (eds T. Ananthakrishnan & D. Whitman), pp. 381–420. Science Pub Inc, Plymouth, UK.