Evaluating the frequency and common drivers of within-host priority effects during coinfection

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A current frontier in disease ecology is understanding how interactions among parasite species influence their epidemics. Interactions among parasites can result when prior infection by one parasite alters host susceptibility to a second parasite, generating priority effects among parasite species within host individuals. The increasing number of laboratory studies that aim to measure priority effects highlights growing interest among disease ecologists to understand these processes. Yet, laboratory studies, which are implemented at the scale of host individuals, are poorly suited to understand parasite epidemics, which occur at the scale of host populations, posing a key challenge for disease ecologists. One way to overcome this challenge is to mark and then repeatedly recapture sentinel hosts in the field, then analyze the resulting data using longitudinal regression models (Fenton *et al.* 2014; Hellard *et al.* 2015; Halliday *et al.* 2017, 2018). The purpose of this project was to implement this approach across empirical systems to test two broad questions:

(1) Is disease risk influenced by infection sequence during natural epidemics (i.e., do parasites generally experience/exhibit priority effects?
(2) To what extent do these within-host priority effects depend on traits of individual parasite species?

To evaluate the role of within-host priority effects during parasite epidemics, we compiled longitudinal datasets of coinfection in host individuals across 13 host and 110 parasite species, including over 25,000 observations of host plants, primates, ungulates, small mammals, birds, and invertebrates. To evaluate the role of within-host priority effects, we performed time-until

event analysis, specifically testing whether infection sequence among co-occurring parasites influenced their risk of infection (e.g., Halliday *et al.* 2017, 2019). Surprisingly, the sequence of infection predicted fewer than 10% of the more than 1000 pairwise combinations of potentially interacting parasites. This rarity of within-host priority effects may result from a lack of natural variation in infection sequence, indicating that within-host priority effects may be a less common driver of parasite epidemics than previously thought, and highlighting the need for experimental manipulations of parasite infection in the field (e.g., Ezenwa & Jolles 2015; Pedersen & Fenton 2015; Halliday *et al.* 2017). Showing weak support for ecological theory, parasites with a high degree of niche overlap were slightly more sensitive to priority effects than parasites with a low degree of niche overlap, and this effect was stronger for parasites with narrow niche requirements than with wide niche-requirements. However, in contrast with theory, the frequency of priority effects could not be explained by a parasite's impact on its host. Together, these results indicate that priority effects may be less commonly detected during natural epidemics than previously expected, and that within-host priority effects among parasites may not necessarily follow the same ecological rules of community assembly as their free-living counterparts.

The database:

We compiled a database of longitudinal surveys of host individuals (i.e., mark-recapture data), where hosts were unmanipulated (sentinels were ok), and surveyed for infection by multiple parasites. This study focused on interspecific interactions among parasites rather than intraspecific interactions among strains of the same parasite species. The final database includes plants, invertebrate, and vertebrate hosts (Table 1).

Table 1. Contributing authors and associated host datasets

Contributors	***Host species***	***Host taxon***
T. Aivelo	*Microcebus rufus*	Primate
P. Beldomenico	*Hydrochoerus hydrochaeris*	Rodent
A. Blackwell	*Homo sapiens*	Human
V. Ezenwa, A. Jolles, E. Gorsich	*Syncerus caffer*	Wild ungulate
AM Hämäläinen, JH Rakotoniaina, E. Kaesler, C. Kraus, P. Kappeler	*Microcebus murinus*	Primate
C. Fichtel, C. Defolie	*Eulemur rufifrons*	Primate
C. Freeman-Gallant	*Geothlypis trichas*	Bird
F. Halliday	*Festuca arundinacea*	Plant
M. Huffman	*Pan troglodytes*	Primate
N. Olifers, AM Júnior, EP de Araujo , R de Cassia Bianchi, APN Gomes, PHD Cançado, ME Gompper, & PS D'Andrea	*Nasua nasua & Cerdocyon thous*	Carnivore
M. Soldanova	*Lymnaea stagnalis*	Invertebrate
C. Taylor	*Microtus agrestis*	Rodent

***A standard measurement of within-host priority effects across systems*:**

To facilitate comparisons across systems, data were analyzed using a common analytical method (described in Halliday et al 2017). Briefly, to detect priority effects, we recorded the sequence of infection by each parasite on each host individual. Time in days since the first survey of each host individual was used as a proxy for exposure to parasite propagules. To model within-host priority effects, we constructed a series of models following Halliday et al (2017). These models explicitly measure priority effects by testing whether the sequence of infection on an individual host influences the rate of infection by each parasite. Each model included one dependent variable pertaining to one parasite species ("the focal parasite"). To explicitly model priority effects, we used Cox Regression with Firth's Penalized Likelihood to reduce bias from factors exhibiting (quasi)complete separation (Heinze & Schemper 2002) from the R package, coxphf (Ploner & Heinze 2015), to estimate the probability of a host transitioning from uninfected to infected. Specifically, the dependent variable in each model was time to infection, modeled as the transition rate from uninfected to infected as a function of exposure time. We modeled time to infection resulting from a baseline rate of infection shared by all individuals and modified by the infection status of the host by each other parasite during the previous survey (treated as a time-varying coefficient). Exponentiated coefficients are interpreted as multiplicative changes in infection rate, providing a standardized estimate of priority effects across study systems. Statistically significant ($p<0.05$) positive and negative coefficients were interpreted as evidence of facilitative and antagonistic priority effects, respectively, whereas non-significant coefficients were interpreted as lacking sufficient evidence of a priority effect occurring.

Our hypotheses:

Hypothesis 1 – Niche overlap. Priority effects are expected to occur more commonly among species with a high degree of niche overlap (Vannette & Fukami 2014). A host comprises the entire niche available to parasites during infection (Kuris *et al.* 1980; Rynkiewicz *et al.* 2015), and thus coinfecting parasites often exhibit high niche overlap (Sousa & Zoologist 1992; Graham 2008; Seabloom *et al.* 2015). The degree of niche overlap among parasites may additionally vary depending on whether parasites share common vectors, employ similar feeding strategies, or share infection sites. We hypothesized that such parasites would therefore more commonly experience priority effects.

Hypothesis 2 – Parasite virulence. Priority effects are expected to occur when early arriving species have a high impact on shared resources with later arriving species (Vannette & Fukami 2014). Parasites require host resources for survival, growth, and reproduction (Lafferty & Kuris 2002). We hypothesized that early-arriving parasites that were more damaging to their hosts would more commonly experience priority effects.

Hypothesis 3 - Host breadth. Priority effects are expected to occur when the late arriving species have high requirements for shared resources with early arriving species (Vannette & Fukami 2014). Late arriving species with narrow niche requirements may therefore experience the strongest priority effects. For free-living species, the breadth of niche requirements may manifest as generality in a species' use of habitat types or tolerance to environmental conditions. For parasites, the breadth of niche requirements may be related to host specificity, defined as the ability to infect multiple unrelated host taxa (Barrett *et al.* 2009; Park *et al.* 2018). We hypothesized that late-arriving host specialists would therefore more commonly experience priority effects.

Analytical methods

Data analysis was performed using R version 3.5.2 (R Core Team 2015). To test whether parasites that shared similar traits more commonly experienced priority effects (Hypothesis 1), we used a series of generalized linear mixed models using the lme4 package, version 1.1-20 (Bates *et al.* 2014), treating the source dataset and host species (Table 1) as random intercepts in each model. The dependent variable in each model was whether or not a pairwise combination of parasites exhibited a priority effect (0,1), and the independent variables where whether or not parasites were the same type (e.g., both macroparasites), shared a common infection site (e.g., both gastrointestinal), or shared the same transmission mode (e.g., both tick-vectored). Data for every trait was not available for every parasite species (Table S1), so in order to maximize the number of studies contributing to the analysis, each independent variable was therefore tested in a separate analysis. We used inverse-variance weighting based on the number of surveys per host to give more explanatory weight to studies with a greater number of surveys of each host individual. However, because this weighting strongly influenced the results of the analyses, we report results from both weighted and unweighted analyses.

To test whether early-arriving parasites that were more damaging to their hosts would more commonly experience priority effects, we again tested whether or not a pairwise combination of parasites exhibited a priority effect, this time as a function of host virulence. We coded host virulence as 1 if the parasite had few reported adverse effects on the host, 2 if the parasite was known to moderately reduce host fitness, and 3 if the parasite was known to seriously impact host survival (e.g., infections frequently leading to host mortality).

To test whether late-arriving host specialists more commonly experienced priority effects, we used a similar, weighted generalized linear mixed model in lme4, testing whether or not a pairwise combination of parasites exhibited a priority effect as a function of the host specificity of the late-arriving parasite (generalist = known to infect more than one host species; specialist = known to infect only a single host species).

We finally tested whether niche-overlap lead to stronger priority effects among late-arriving host specialists by analyzing the interaction coefficient for each pairwise combination of parasites as a dependent variable. Because more closely related species tend to exhibit higher niche-overlap, we used the phylogenetic relatedness of parasites as a metric of niche-overlap, and modeled its interaction with host specificity of the late-arriving parasite and their main effects as independent variables in this analysis. Parasites were defined as "closely related" if they shared at least the same order, and "distantly related" if they were from different orders or more distantly related.

Results

Contrary to expectations, we found surprisingly limited evidence of priority effects across our database. Specifically, out of 1128 potential pairwise interactions, we detected only 98 interactions resulting from prior infection (i.e., priority effects). 64 were facilitative (prior infection by one parasite species facilitated subsequent infection by another species), 34 interactions were antagonistic (prior infection by one parasite species inhibited infection by a subsequent species), and the remaining 1030 lacked sufficient evidence to support a priority effect occurring. This surprising result may stem from a lack of variation in infection sequence among study systems. Early infections were often dominated by one or a few parasite species (Fig. 1), potentially resulting from variation in parasite phenology (Halliday *et al.* 2017) or variation in parasite transmission rates (Clay *et al.* 2019). Together, these results point to the need for studies that systematically manipulate infection sequence during natural epidemics,

potentially through the use of pesticides or anti-parasite treatments of hosts (Pedersen & Fenton 2015).

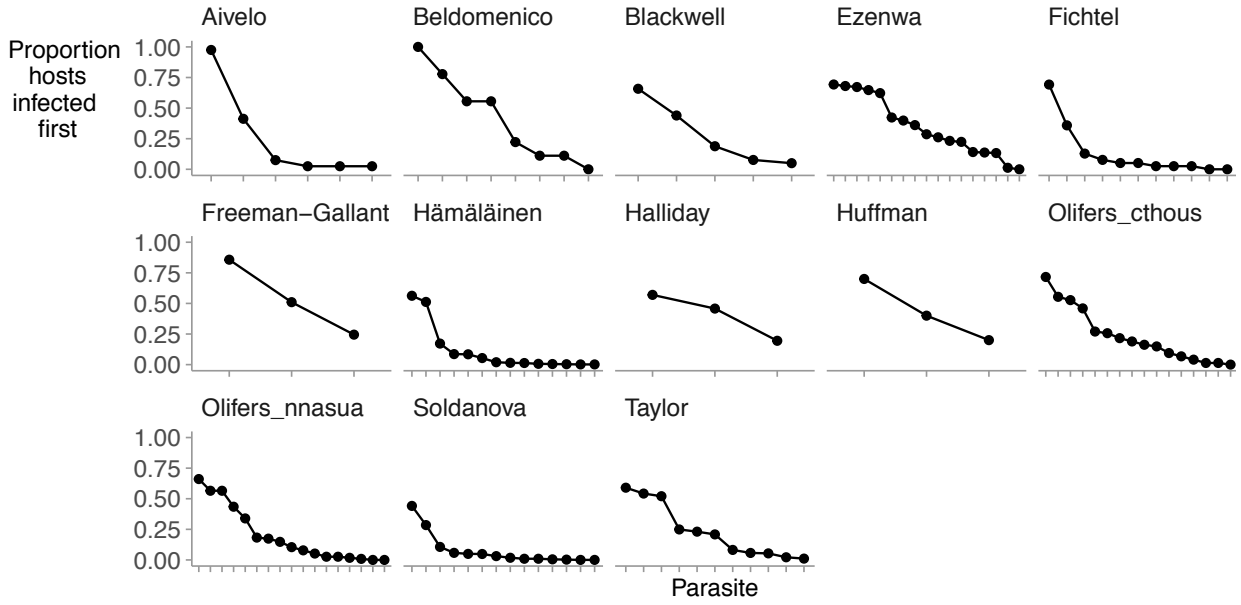

Fig 1. In most study systems, early infections were dominated by one or a few parasites. This lack of variation in infection sequence may explain why fewer priority effects were observed than expected based on laboratory studies. Each point represents a single parasite species, and the y-axis represents the proportion of hosts whose first infection was associated with that parasite. Values on the y-axis sum to >1 due to simultaneous infections.

We found weak support for the hypothesis that parasites with high niche overlap would more commonly experience priority effects. Specifically, when weighted by the number of samples per host individual, parasites of the same type (e.g., both macroparasites), parasites that infected the same site (e.g., gastrointestinal parasites), and parasites with the same transmission mode (e.g., both tick vectored), all experienced significantly more priority effects than parasites of different types, different sites, and different transmission modes ($p= 0.006$, $p<0.001$, and $p = 0.004$, respectively; Fig 2). However, these effects were of small magnitude and were not statistically significant when the same analyses were performed with unweighted data ($p = 0.98$, $p = 0.48$, $p = 0.68$, respectively). We found no support for the hypothesis that early-arriving parasites that were more damaging to their hosts would more commonly experience priority effects. Even though the weighted regression indicated that parasite virulence significantly predicted the priority effects ($p < 0.001$), the direction of this effect was not consistent with our prediction, with moderately virulent parasites exhibiting the highest rate of priority effects (Fig

2). The effect of host virulence on the probability of observing a priority effect was not significant when the analysis was performed with unweighted data (p = 0.25).

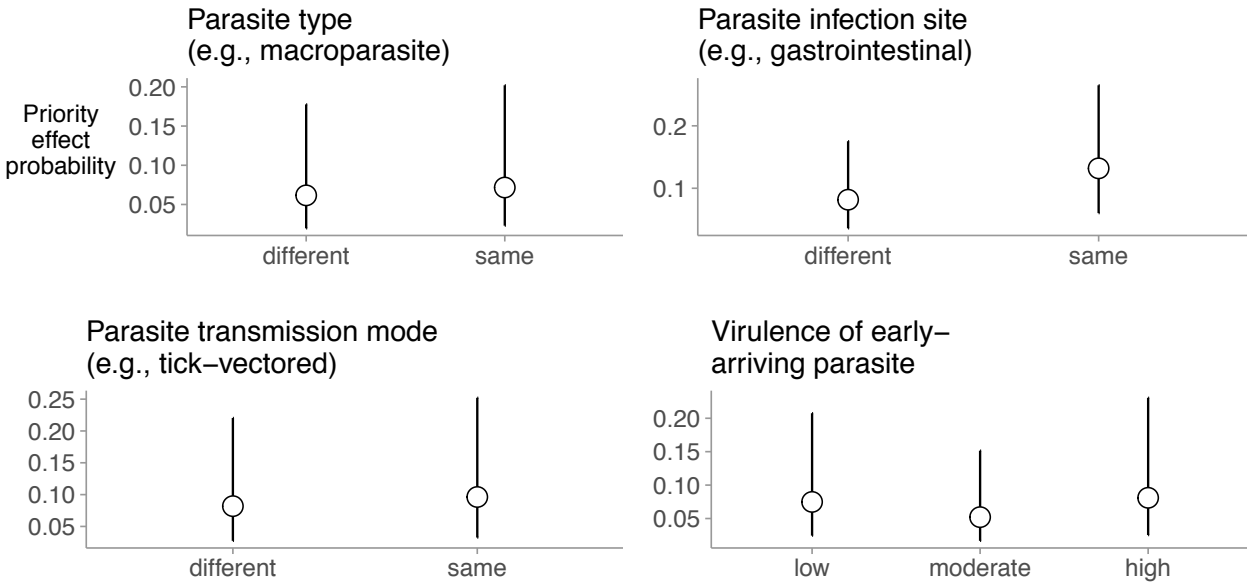

Fig 2. Priority effects were somewhat more commonly observed among parasites of the same type, infection site, or transmission mode ($p<0.05$ when inversely weighted by number of samples per host; $p>0.05$ when unweighted), lending some support to hypothesis one. In contrast with hypothesis one, priority effects were no more commonly observed when early arriving species had greater or smaller impacts on their hosts, though weighted regression indicates that moderately viulent parasites may exhibit fewer priority effects than low- or highly virulent parasites.

We found strong evidence that later arriving parasites with narrow host breadth (e.g., specialists) more commonly experienced priority effects than parasites with wider host breadth (e.g., generalists; weigthed $p < 0.001$, unweighted $p = 0.007$; Fig. 3 - inset). Furthermore, among host specialists, more closely related parasites tended to exhibit more antagonistic interactions ($p = 0.003$). Together, these results indicate that priority effects among parasites may be less common than expected based on laboratory studies**,** and that priority effects are rarely consistent with predictions from ecological theory**.**

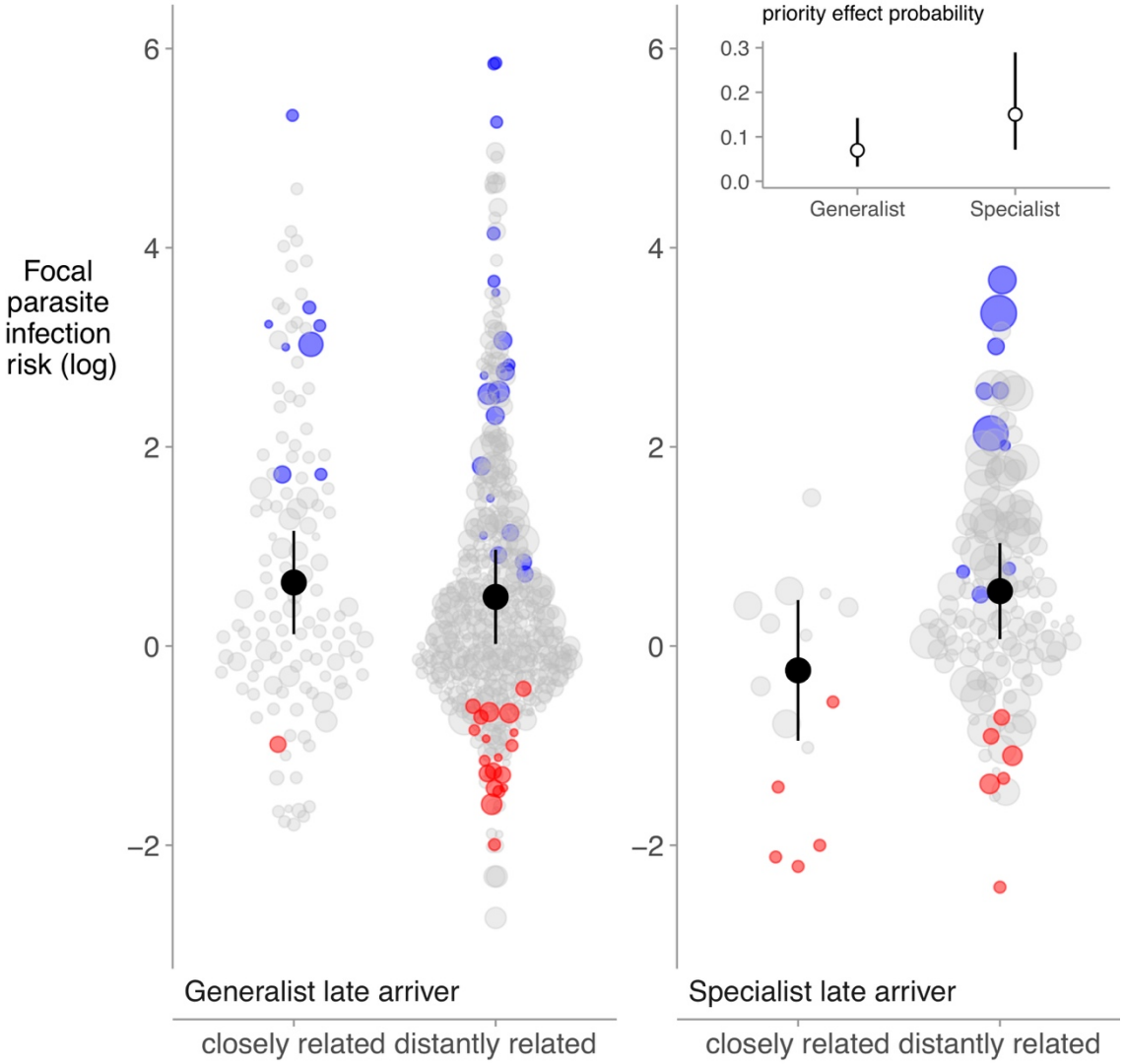

Fig 3. Priority effects were more commonly observed when late-arriving parasites were host specialists, supporting hypothesis two. Among specialists, more closely related parasites tended to experience more competition. Black circles are model estimated means, error bars are 95% confidence intervals; colored points are estimates from each individual pairwise combination of studies, with size corresponding to the number of samples per host individual. Blue indicates significant facilitation, red indicates significant antagonism, and grey indicates insufficient evidence for significant priority effect. The inset shows the model estimated probability of a priority effect on the y-axis as a function of host specificity.

References

Barrett, L.G., Kniskern, J.M., Bodenhausen, N., Zhang, W. & Bergelson, J. (2009). Continua of specificity and virulence in plant host–pathogen interactions: causes and consequences. *New Phytol.*, 183, 513–529.

Bates, D., Mächler, M., Bolker, B. & Walker, S. (2014). Fitting Linear Mixed-Effects Models using lme4. *J. Stat. Softw.*, 67, 1–48.

Clay, P.A., Cortez, M.H., Duffy, M.A. & Rudolf, V.H.W. (2019). Priority effects within coinfected hosts can drive unexpected population-scale patterns of parasite prevalence. *Oikos*, 128, 571–583.

Ezenwa, V. & Jolles, A. (2015). Opposite effects of anthelmintic treatment on microbial infection at individual versus population scales. *Science (80-.).*

Fenton, A., Knowles, S.C.L., Petchey, O.L. & Pedersen, A.B. (2014). The reliability of observational approaches for detecting interspecific parasite interactions: comparison with experimental results. *Int. J. Parasitol.*, 44, 437–45.

Graham, A.L. (2008). Ecological rules governing helminth-microparasite coinfection. *Proc. Natl. Acad. Sci. U. S. A.*, 105, 566–70.

Halliday, F.W., Heckman, R.W., Wilfahrt, P.A. & Mitchell, C.E. (2019). Past is prologue: host community assembly and the risk of infectious disease over time. *Ecol. Lett.*, 22, 138–148.

Halliday, F.W., Umbanhowar, J. & Mitchell, C.E. (2017). Interactions among symbionts operate across scales to influence parasite epidemics. *Ecol. Lett.*, 20, 1285–1294.

Halliday, F.W., Umbanhowar, J. & Mitchell, C.E. (2018). A host immune hormone modifies parasite species interactions and epidemics: insights from a field manipulation. *Proc. R. Soc. B Biol. Sci.*, 285, 20182075.

Heinze, G. & Schemper, M. (2002). A solution to the problem of separation in logistic regression. *Stat. Med.*, 21, 2409–2419.

Hellard, E., Fouchet, D., Vavre, F. & Pontier, D. (2015). Parasite-Parasite Interactions in the Wild: How To Detect Them? *Trends Parasitol.*

Kuris, A.M., Blaustein, A.R. & Alio, J.J. (1980). Hosts as Islands. *Am. Nat.*, 116, 570–586.

Lafferty, K.D. & Kuris, A.M. (2002). Trophic strategies, animal diversity and body size. *Tree*, 17, 507–513.

Park, A.W., Farrell, M.J., Schmidt, J.P., Huang, S., Dallas, T.A., Pappalardo, P., *et al.* (2018). Characterizing the phylogenetic specialism-generalism spectrum of mammal parasites. *Proc. R. Soc. B Biol. Sci.*, 285, 20172613.

Pedersen, A.B. & Fenton, A. (2015). The role of antiparasite treatment experiments in assessing the impact of parasites on wildlife. *Trends Parasitol.*, 31, 200–11.

Ploner, M. & Heinze, G. (2015). coxphf: Cox regression with Firth's penalized likelihood. *R Found. Stat. Comput.*

R Core Team. (2015). R: a language and environment for statistical computing | GBIF.ORG.

Rynkiewicz, E.C., Pedersen, A.B. & Fenton, A. (2015). An ecosystem approach to understanding and managing within-host parasite community dynamics. *Trends Parasitol.*, 31, 212–221.

Seabloom, E.W., Borer, E.T., Gross, K., Kendig, A.E., Lacroix, C., Mitchell, C.E., *et al.* (2015). The community ecology of pathogens: Coinfection, coexistence and community composition. *Ecol. Lett.*, 18, 401–415.

Sousa, W.P. & Zoologist, A. (1992). Interspecific Interactions among Larval Trematode Parasites of Freshwater and Marine Snails Interspecific Interactions Among Larval Trematode Parasites of Freshwater and Marine Snails1. *Interactions*, 32, 583–592.

Vannette, R.L. & Fukami, T. (2014). Historical contingency in species interactions: Towards niche-based predictions. *Ecol. Lett.*, 17, 115–124.

Table S1. Parasites and their associated traits

Parasite	*Type*	*Virulence*	*Specialist*	*Route*	*Mode*	*Helminth*	*Intracellular*
Anaplasma centrale	microparasite	Low	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Ascaris lumbricoides	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Anaplasma marginale	microparasite	Medium	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Anaplasma phagocytophilum	microparasite	Medium	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Acanthocephala	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Amblyomma cajennense	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Amblyomma ovale	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Amblyomma parvum	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Apicomplexa	microparasite	Low		Complex		Not Helminth	Intracellular
Ascaridae	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Ascaridida	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Aspidoderidae	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Australapatemon burti	macroparasite	Medium	Generalist	Complex	Ingestion	Helminth	Not intracellular
Australapatemon minor	macroparasite	Medium	Generalist	Complex	Ingestion	Helminth	Not intracellular
Babesia	microparasite	Low	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Ballantidium	microparasite	Low		Direct	Ingestion	Not Helminth	Not intracellular
Bartonella	microparasite	Medium	Generalist	Complex	Flea vectored	Not Helminth	Intracellular
Brucella abortus	microparasite	Medium	Generalist	Direct	Ingestion	Not Helminth	Intracellular
Callistoura	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Capillaria	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Cestoda	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular

Parasite	*Type*	*Virulence*	*Specialist*	*Route*	*Mode*	*Helminth*	*Intracellular*
Conoidasida	microparasite	Low		Direct	Ingestion	Not Helminth	Intracellular
Conoidasida	microparasite	Low		Direct	Ingestion	Not Helminth	Intracellular
Colletotrichum cereale	microparasite	Low	Generalist	Direct	Rain splash	Not Helminth	Not intracellular
Cooperia	macroparasite	Low	Generalist	Direct	Ingestion	Helminth	Not intracellular
Cotylurus cornutus	macroparasite	Medium	Generalist	Complex	Ingestion	Helminth	Not intracellular
Cowpox virus	microparasite	Medium	Generalist	Direct	Direct contact	Not Helminth	Intracellular
Dicrocoeliidae	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Diphyllobothriidae	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Diplostomum pseudospathaceum	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Dipylidiidae	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Ehrlichia ruminantium	microparasite	Medium	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Anaplasma	microparasite	Low	Specialist	Complex	Tick vectored	Not Helminth	Intracellular
Echinocleus hydrochaeri	macroparasite	Low	Specialist	Complex	Ingestion	Helminth	Not intracellular
Echinoparyphium aconiatum	macroparasite	Medium	Generalist	Complex	Ingestion	Helminth	Not intracellular
Echinoparyphium recurvatum	macroparasite	Medium	Generalist	Complex	Ingestion	Helminth	Not intracellular
Echinostoma revolutum	macroparasite	Medium	Generalist	Complex	Ingestion	Helminth	Not intracellular
Eimeria boliviensis	microparasite	Medium	Specialist	Direct	Ingestion	Not Helminth	Intracellular
Eimeria hydrochaeri	microparasite	Medium	Specialist	Direct	Ingestion	Not Helminth	Intracellular
Eimeria	microparasite	Medium	Specialist	Direct	Ingestion	Not Helminth	Intracellular
Entamoeba	microparasite	Medium		Direct	Ingestion	Not Helminth	Not intracellular

Parasite	*Type*	*Virulence*	*Specialist*	*Route*	*Mode*	*Helminth*	*Intracellular*
Fasciolidae	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Siphonaptera	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Giardia lamblia	microparasite	Medium	Generalist	Direct	Ingestion	Not Helminth	Not intracellular
Haemonchus	macroparasite	Medium		Direct	Ingestion	Helminth	Not intracellular
Ancylostomatidae	macroparasite	Low	Specialist	Direct	Free-living	Helminth	Not intracellular
Hymenolepis	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Hypoderaeum conoideum	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Laelapidae	ectoparasite	Low		Direct	Free-living	Not Helminth	Not intracellular
Lemuricola	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Lemuricola	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Lemurostrongylus	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Leucocytozoon	microparasite	Low	Generalist	Complex	Blackfly vectored	Not Helminth	Intracellular
Listrophorus	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Phthiraptera	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Metagonymus	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Hystrichopsyllidae	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Moliniella anceps	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Myobiidae	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Neotrichodectes pallidus	ectoparasite	Low	Specialist	Direct	Free-living	Not Helminth	Not intracellular
Notocotylus attenuatus	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Oesophagostomum	macroparasite	Medium	Generalist	Direct	Ingestion	Helminth	Not intracellular

Parasite	*Type*	*Virulence*	*Specialist*	*Route*	*Mode*	*Helminth*	*Intracellular*
Opisthioglyphe ranae	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Opisthorchis	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Oesophagostomum stephanostomum	macroparasite	Medium	Generalist	Direct	Ingestion	Helminth	Not intracellular
Oxyuridae	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Oxyuridae	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Oxyuridae	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Pararhabdonema	macroparasite	Low	Generalist	Direct	Ingestion	Helminth	Not intracellular
Paryphostomum radiatum	macroparasite	Low	Generalist	Complex	Ingestion	Helminth	Not intracellular
Physalopteridae	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Plagiorchis elegans	macroparasite	Low	Specialist	Complex	Ingestion	Helminth	Not intracellular
Plasmodium	microparasite	Medium	Specialist	Complex	Mosquito vectored	Not Helminth	Intracellular
Protozoophaga obesa	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Strongyloides	macroparasite	Low	Generalist	Direct	Free-living	Helminth	Not intracellular
Caenorhabditis	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Strongyloidae	macroparasite	Low	Generalist	Direct	Ingestion	Helminth	Not intracellular
Chromadorea	macroparasite	Low				Helminth	Not intracellular
Enterobius	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Panagrellus	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Puccinia coronata	microparasite	Low	Specialist	Complex	Airborne	Not Helminth	Not intracellular
Rhizoctonia solani	macroparasite	Medium	Generalist	Direct	Rain splash	Not Helminth	Not intracellular

Parasite	*Type*	*Virulence*	*Specialist*	*Route*	*Mode*	*Helminth*	*Intracellular*
Schistosoma	macroparasite	Medium	Generalist	Complex	Free-living	Helminth	Not intracellular
Schistosomatidae	macroparasite	Medium	Generalist	Complex	Free-living	Helminth	Not intracellular
Strongyloides fuelleborni	macroparasite	Low	Generalist	Direct	Ingestion	Helminth	Not intracellular
Strongiloides	macroparasite	Low	Generalist	Complex	Free-living	Helminth	Not intracellular
Strongyloidae	macroparasite	Low	Generalist	Complex	Free-living	Helminth	Not intracellular
Strongyloidae	macroparasite	Low	Generalist	Complex	Free-living	Helminth	Not intracellular
Strongyloidae	macroparasite	Low	Generalist	Complex	Free-living	Helminth	Not intracellular
Strongiloides chapini	macroparasite	Low	Generalist	Complex	Free-living	Helminth	Not intracellular
Subulura	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Subulura	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Theileria mutans	microparasite	Low	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Theileria parva	microparasite	High	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Theileria sable	microparasite	Low	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Theileria sp (buffalo)	microparasite	Medium	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Trichuris trichiura	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Theileria velifera	microparasite	Low	Generalist	Complex	Tick vectored	Not Helminth	Intracellular
Mycobacterium tuberculosis	microparasite	Medium	Specialist	Direct	Airborne	Not Helminth	Intracellular
Thysanotaenia	macroparasite	Low	Specialist	Complex	Ingestion	Helminth	Not intracellular
Ixodida	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Trematoda	macroparasite	Low		Complex		Helminth	Not intracellular
Trichinellidae	macroparasite	Low		Complex	Ingestion	Helminth	Not intracellular
Trichobilharzia szidati	macroparasite	Low	Generalist	Complex	Free-living	Helminth	Not intracellular

Parasite	*Type*	*Virulence*	*Specialist*	*Route*	*Mode*	*Helminth*	*Intracellular*
Trichodectes canis	ectoparasite	Low	Generalist	Direct	Free-living	Not Helminth	Not intracellular
Trichostrongyloidea	macroparasite	Low		Direct	Ingestion	Helminth	Not intracellular
Trichuris	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Trichuris cutillasae	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Trichuris	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular
Trypanosoma	microparasite	Medium	Generalist	Complex	Tsetse fly vectored	Not Helminth	Intracellular
Trichuris trichiura	macroparasite	Low	Specialist	Direct	Ingestion	Helminth	Not intracellular

Complex parasites sequentially infect different hosts over the course of their lifecycle