A healthy but depleted herd: Predators decrease prey disease and density.

Lopez, L.K., Cortez, M.H., DeBlieux, T., Menel, I., O'Brien, B., Cáceres, C.E., Hall, S.R., Duffy, M.A.

Ecology.

Appendix S1

Contents

Section S1 Supplemental empirical methods

Section S1.1 Quantifying susceptibility to predation of different clones

Our mesocosm experiment used nine genotypes of *Daphnia dentifera* that differed in feeding rates and susceptibility to infection by the parasite *Metschnikowia bicuspidata*: BD 05-42, BD 08-46(81), BD 19-64(73), CB 24-68, DW 22-58(84), DW 29-75, IL 14-43, ML 30-82, and ML 32-84. Clones with "BD" at the start are from Beaver Dam Lake, the "CB" clone is from Canvasback Lake, the "DW" clones are from Downing Lake, "IL" from Island Lake, and "ML" from Midland Lake; these lakes are in Greene and Sullivan Counties, Indiana.

We quantified the susceptibility of these nine genotypes to predation by *Chaoborus punctipennis*. To do this, we reared individuals from each *D. dentifera* genotype at 20 °C. When they reached 8 days old, we placed individuals in 150 mL beakers filled with 100 mL of filtered lake water at densities of 1, 2, 5 or 10 animals/beaker. All individuals in a beaker were from the same genotype, and all individuals used in this study were uninfected (and had not been exposed to parasites). We placed a single 3rd or 4th instar *C. punctipennis* (collected from lakes in Indiana and starved 48 hours prior to the feeding trials) into each beaker. Beakers were kept in the dark at 20 °C for 16 hours, at which point the number of surviving *D. dentifera* was recorded. Susceptibility to predation was then quantified by estimating predator attack rates, as described in the multi-clone model section below.

Section S1.2 Quantifying selectivity of predation of infected vs. uninfected prey

We quantified the susceptibility of uninfected (susceptible) and infected *D. dentifera* individuals to predation by *Chaoborus punctipennis*. This was part of a larger study on the effect of infection on host behavior; full results of that study will be reported elsewhere. To do this, we reared individuals of a single genotype (known as "Mid-37", which comes from Midland Lake, Indiana) at 20 °C. All individuals were maintained at 20 °C under an 18:6 light:dark schedule. When the individuals were 9 days old, the population was split into a control population and an exposed population. For the control population, groups of 20 individuals were transferred to 120 mL beakers with 100 mL of lake water. For the exposed population, groups of 20 individuals were transferred to 120 mL beakers and exposed to *Metschnikowia bicuspidata* spores at a dose of 200 spores per mL. Animals were moved to beakers with clean (spore-free) water after 24 hours. Every three days thereafter, animals were moved to beakers with clean water, taking care not to transfer any offspring.

Beginning the day after infection, and every third day after that, beakers of 10 *Daphnia* from each population were exposed to a third or fourth instar *Chaoborus*. For the "control" treatment, all individuals in each beaker were from the control population (and therefore not exposed to parasites). For the "exposed" treatment, all individuals in each beaker were from the exposed population but not (yet) showing obvious signs of infection (especially asci in the hemolymph). For the "infected" treatment, all individuals in each beaker were from

the exposed population and showing visible signs of infection; because of delays between when individuals are exposed and when they develop asci, we did not have "infected" beakers until 10 days post-exposure. For the predation trials, beakers were kept in the dark at 20 °C for 15 hours, at which point the number of surviving *D. dentifera* was recorded. Susceptibility to predation was then quantified by estimating predator attack rates, as described in the multi-clone model section below.

Section S1.3 Additional methods related to mesocosm experiment and genotyping

Each tank received 70 individuals of the following clonal lines: BD 05-42, BD 08-46(81), BD 19-64(73), CB 24-68, DW 22-58(84), DW 29-75, IL 14-43, ML 30-82, and ML 32-84. These are the same genotypes that were used in the "Quantifying susceptibility to predation of different clones" experiment (above). Table S3 (below) contains estimates of the per spore susceptibility, feeding rates, and susceptibility to predation for each of these genotypes. These genotypes do not show a tradeoff between susceptibility to predation and susceptibility to parasitism (Figure S1a).

Each of these genotypes was known to have a unique multilocus genotype using a set of microsatellites developed by Fox (2004) and that we have used in prior studies (Strauss et al., 2017). We were therefore able to genotype the individuals that were preserved in ethanol during our routine sampling, following the general methods outlined in Allen et al. (2010). We analyzed individuals that had been preserved on weeks 2, 6, and 9 to determine evolutionary changes in the population that resulted from shifts in clonal frequencies.

Individuals were first rinsed in deionized water to remove ethanol. We then digested tissue and extracted DNA by incubating each individual in a 5% Chelex solution. This solution contained 0.8 g Chelex resin (200-400 mesh), 8 mL TE buffer, and 8 mL molecular-grade water. The Chelex solution was vortexed, then 150 µL was immediately pipetted into a sterile 1.5 mL microcentrifuge tube. A single *Daphnia* individual was added, then the tube was again briefly vortexed. Samples were then incubated at 50 °C for at least 3 hours. We then raised the temperature to 99 degrees C for 10 minutes. Samples were then briefly vortexed at a low speed, then centrifuged at 8000 rpm for 2 mL. 70 µL of the supernatant was then pipetted to a new microcentrifuge tube and stored in the freezer until PCR.

We then amplified four microsatellite loci using PCR, using primers that were designed by Fox (2004): Dgm106, Dgm109, Dgm112, and Dgm113. The genotypes we used in this study have unique microsatellite genotypes at these four loci. Each PCR reaction contained 6 µL Qiagen multiplex PCR mastermix, 1.2 µL primer mix (2 mmol each), 3.8 µL ddH20, and 1 µL DNA sample. PCR was run on a SimpliAmp Thermal Cycler. Cycling was initiated with one cycle at 95 °C for 15 minutes, followed by 30 cycles (94 °C for 30 s, 58 °C for 180 s, 72 °C for 90 s) and a final extension at 72 °C for 10 minutes. Amplified DNA was diluted (1 µl amplified DNA and 10 µl ddH20). Samples were sent to the Roy J. Carver Biotechnology Center at the University of Illinois Urbana-Champaign (Urbana, IL,

USA) for microsatellite fragment analysis. We called alleles using GeneMapper software (Version 5: Applied Biosystems, Foster City, CA, USA). Because the genotypes we used in this experiment had unique combinations of alleles at these four loci, we were able to use this information to assign each individual to one of the genotypes that we used to start the experiment.

Section S2 Supplemental empirical results

Section S2.1 Evolution in prey populations

As stated in the results section in the main text, the mean susceptibility of the populations evolved over time via clonal selection. For mean susceptibility to parasitism, there was a significant effect of time ($F_{1,71} = 112.0$, $p < 0.0001$) and parasitism ($F_{1,39} = 4.86$, $p = 0.0334$) but not of predation ($F_{3,39} = 2.16$, $p = 0.1079$). None of the interaction effects were significant (predation*parasitism: $F_{3,39} = 1.52$, $p = 0.225$; predation*time: $F_{3,71} = 0.475$, $p = 0.701$; parasitism*time: $F_{1,71} = 0.135$, $p = 0.715$) though the three way interaction was marginally significant (predation*parasitism*time: $F_{3,71} = 2.18$, $p = 0.0979$).

For mean susceptibility to predation, there was a significant effect of time ($F_{1,71} = 17.06$, $p = 0.0001$) and parasitism ($F_{1,39} = 4.46$, $p = 0.0411$) but not of predation ($F_{3,39} = 0.446$, $p = 0.7217$). None of the two-way interaction effects were significant (predation*parasitism: $F_{3,39} = 0.283$, $p = 0.837$; predation*time: $F_{3,71} = 1.19$, $p = 0.319$; parasitism*time: $F_{1,71} = 0.34$, $p = 0.562$) though the three way interaction was significant (predation*parasitism*time: $F_{3,71} = 5.499$, $p = 0.0019$).

Table S1: Results of the pairwise comparison of total host density across the experiment for the different predator × parasite treatments, using the emmeans package. The number indicates the predation level (0, 0.1, 0.5, or 1 *Chaoborus* predator per L). The two parasitism treatments are indicated by "No parasites" and "Parasites". The 1 predator per L predation treatments with and without parasites did not differ significantly from one another. However, these highest predation treatments (that is, 1.0 predator per L with and without parasites) differed significantly from all of the other treatments; none of those other treatments differed significantly from one another. Put differently, every significant contrast involves one of the two 1 predator per L predation treatments being compared with something other than the other 1 predator per L predation treatment.

Contrast	Estimate	SE	df	lower.CL	upper.CL	t.ratio	p.value
0 Chaob No parasites - 0.1 Chaob No parasites	0.49	0.34	39	-0.6	1.59	1.44	0.834
0 Chaob No parasites - 0.5 Chaob No parasites	0.08	0.33	39	-0.97	1.12	0.23	1
0 Chaob No parasites - 1 Chaob No parasites	**2.01**	**0.33**	**39**	**0.97**	**3.05**	**6.16**	**8.13E-06**
0 Chaob No parasites - 0 Chaob Parasites	0.41	0.33	39	-0.63	1.46	1.27	0.904
0 Chaob No parasites - 0.1 Chaob Parasites	0.31	0.33	39	-0.74	1.35	0.95	0.979
0 Chaob No parasites - 0.5 Chaob Parasites	0.35	0.33	39	-0.69	1.4	1.08	0.957
0 Chaob No parasites - 1 Chaob Parasites	**2.51**	**0.33**	**39**	**1.47**	**3.56**	**7.7**	**6.43E-08**
0.1 Chaob No parasites - 0.5 Chaob No parasites	-0.42	0.34	39	-1.51	0.68	-1.22	0.921
0.1 Chaob No parasites - 1 Chaob No parasites	1.52	0.34	39	0.42	2.61	4.43	0.002
0.1 Chaob No parasites - 0 Chaob Parasites	-0.08	0.34	39	-1.17	1.02	-0.23	1
0.1 Chaob No parasites - 0.1 Chaob Parasites	-0.18	0.34	39	-1.28	0.91	-0.54	0.999
0.1 Chaob No parasites - 0.5 Chaob Parasites	-0.14	0.34	39	-1.23	0.96	-0.41	1
0.1 Chaob No parasites - 1 Chaob Parasites	**2.02**	**0.34**	**39**	**0.93**	**3.12**	**5.9**	**1.83E-05**
0.5 Chaob No parasites - 1 Chaob No parasites	**1.93**	**0.33**	**39**	**0.89**	**2.98**	**5.93**	**1.69E-05**
0.5 Chaob No parasites - 0 Chaob Parasites	0.34	0.33	39	-0.71	1.38	1.04	0.965
0.5 Chaob No parasites - 0.1 Chaob Parasites	0.23	0.33	39	-0.81	1.28	0.71	0.996
0.5 Chaob No parasites - 0.5 Chaob Parasites	0.28	0.33	39	-0.77	1.32	0.85	0.989
0.5 Chaob No parasites - 1 Chaob Parasites	**2.44**	**0.33**	**39**	**1.39**	**3.48**	**7.47**	**1.31E-07**
1 Chaob No parasites - 0 Chaob Parasites	**-1.6**	**0.33**	**39**	**-2.64**	**-0.55**	**-4.89**	**4.35E-04**
1 Chaob No parasites - 0.1 Chaob Parasites	**-1.7**	**0.33**	**39**	**-2.75**	**-0.66**	**-5.21**	**1.59E-04**
1 Chaob No parasites - 0.5 Chaob Parasites	**-1.66**	**0.33**	**39**	**-2.7**	**-0.61**	**-5.08**	**2.44E-04**
1 Chaob No parasites - 1 Chaob Parasites	0.5	0.33	39	-0.54	1.55	1.54	0.781
0 Chaob Parasites - 0.1 Chaob Parasites	-0.11	0.33	39	-1.15	0.94	-0.32	1
0 Chaob Parasites - 0.5 Chaob Parasites	-0.06	0.33	39	-1.11	0.98	-0.19	1
0 Chaob Parasites - 1 Chaob Parasites	**2.1**	**0.33**	**39**	**1.05**	**3.14**	**6.43**	**3.45E-06**
0.1 Chaob Parasites - 0.5 Chaob Parasites	0.05	0.33	39	-1	1.09	0.14	1
0.1 Chaob Parasites - 1 Chaob Parasites	**2.2**	**0.33**	**39**	**1.16**	**3.25**	**6.76**	**1.24E-06**
0.5 Chaob Parasites - 1 Chaob Parasites	**2.16**	**0.33**	**39**	**1.12**	**3.2**	**6.62**	**1.91E-06**

Section S3 Supplemental methods for models

Section S3.1 Model assumptions

The model given in equations (1.a-c) in the main text makes the following assumptions: (i) all spores are identical, regardless of which clone released them; (ii) all clones are equally exposed to the spores (which matches the experiment); (iii) the predators have a Type 1 functional response; (iv) infected individuals release more spores if they die due to infection than if they die due to consumption by predators (because they have less time to develop within the host, as is supported by prior studies (Auld et al., 2014)); (v) susceptible and infected individuals have unequal uptake rates (which is supported by prior studies, e.g., Penczykowski et al. (2022); Searle et al. (2016)); (vi) there is no recovery from infection, i.e., infection is lethal; (vii) predators have higher attack rates on infected individuals than susceptible individuals. The model includes the removal of liquid for destructive sampling, which occurs at rate λ, but we set $\lambda = 0$ in the model analyses because the removal rate is negligible; see below for details.

Section S3.2 Estimating model parameter values

Details about how each parameter value was estimated are given below. The values are summarized in Table S3.

Clone reproduction rates, G_i: We cannot estimate the functions defining clonal reproduction rates because we do not have estimates of the growth rates and competitive abilities for all nine genotypes. However, we do not need that information for our analyses. Specifically, our calculations of the basic reproduction number $\mathcal{R}_0$ and the reproduction number (R) are independent of the reproduction functions because we compute them using the empirically measured prey densities. Also, the sensitivity of total prey density to predation (dN^*/dP) can be computed without a parameterization of the reproduction rates, provided we make the reasonable assumption that the average per capita growth rate decreases with increased density; see Section S6 for details.

Estimating predator attack rates on susceptible individuals, a_i: Attack rates were estimated from the predation trials with different clones (Section Section S1.1) where predators were offered different numbers of uninfected individuals of a particular genotype. These experiments used all nine genotypes, but predators were only offered individuals from a single genotype at a time, as indicated in Section S1.1 above.

Let $S(t)$ be susceptible prey density at time t in a consumption experiment, T be the total length of the experiment, $S(0) = S_0$ be the initial density, S_e be the density eaten at the end of the experiment, and P be the predator density. We assume the predator has a Type 1 functional response, which means the change in prey density is given by the differential equation $\dfrac{dS}{dt} = -a_i SP$ where a_i is the attack rate on susceptible individuals of

clone i. Solving this differential equation yields $S(T) = S_0 \exp(-a_i PT)$. This means that the density of prey eaten is given by

$$S_e = S_0 - S(T) = S_0(1 - \exp(-a_i PT)).$$

This derivation is a special case of the derivation for Type 2 functional responses originally derived in Royama (1971) and Rogers (1972) and reviewed in Rosenbaum and Rall (2018).

The model was fit to the consumption data for clone i in the following way. The values of T and P are known quantities and the same across all replicates. Let n be the total number of replicates across all prey density treatments for clone i. At the end of the experiment, an individual prey is either dead or alive. Thus, the model is fit to the predation data using a likelihood function that assumes the data are binomially distributed. For replicate j, let $S_{0,j}$ denote the initial density and $S_{e,j}$ denote the observed number of prey eaten at the end of the experiment. For a given parameter $\hat{a}_i$ and a given replicate, the expected number of eaten prey is $\hat{S}_{e,j} = S_{0,j}(1 - \exp(-\hat{a}_i PT))$. From this, the expected probability an individual is eaten in replicate j is $\hat{\rho} = \hat{S}_{e,j}/S_{0,j}$. Assuming a binomial distribution, the likelihood of the parameter $\hat{\rho}$ given the data in all replicates is

$$\mathcal{L}(S_{e,1}, ..., S_{e,n}, S_{0,1}, ..., S_{0,n}|\hat{\rho}) = \prod_j c(S_{0,j}, S_{e,j})(\hat{\rho})^{S_{e,j}}(1 - \hat{\rho})^{S_{0,j} - S_{e,j}} \tag{S1}$$

where $c(S_{0,j}, S_{e,j})$ denotes combinations of $S_{0,j}$ individuals taken $S_{e,j}$ at a time. The negative log likelihood is

$$NLL = -\sum_j \ln[c(S_{0,j}, S_{e,j})] + S_{e,j} \ln(\hat{\rho}) + (S_{0,j} - S_{e,j}) \ln(1 - \hat{\rho}) \tag{S2}$$

The negative log likelihood is maximized at the parameter value satisfying $\partial NLL/\partial\hat{\rho} = 0$. Differentiating and rearranging terms yields

$$0 = -\sum_j S_{e,j}/\hat{\rho} + \sum_j (S_{0,j} - S_{e,j})/(1 - \hat{\rho}) \tag{S3}$$

$$\Rightarrow 0 = -(1 - \hat{\rho}) \sum_j S_{e,j} + \hat{\rho} \sum_j (S_{0,j} - S_{e,j}) \tag{S4}$$

$$\Rightarrow \hat{\rho} = \sum_j S_{e,j} \Big/ \sum_j S_{0,j}. \tag{S5}$$

Substituting using $\hat{\rho} = \hat{S}_{e,j}/S_{0,j} = 1 - \exp(-\hat{a}_i PT)$ and solving for $\hat{a}_i$ yields the estimate for $\hat{a}_i$ that maximizes the negative log likelihood,

$$\hat{a}_i = -\frac{1}{PT} \ln\left(1 - \frac{\sum_j S_{e,j}}{\sum_j S_{0,j}}\right) \tag{S6}$$

Estimating predator attack rates on infected individuals, ωa_i: As explained above, we estimated predator attack rates on susceptible individuals of each clone (a_i). We did not estimate predator attack rates on infected individuals of each clone. Instead, we assume (i) the attack rates on infected individuals of clone i are a multiplicative factor of the attack rates on the susceptible individuals of clone i and (ii) the multiplicative factor is the same for all *Daphnia* genotypes. This means that we assume the attack rates on infected individuals of clone i are ωa_i, where ω determines if infected individuals experience increased predation rates ($\omega > 1$), equal predation rates ($\omega = 1$), or reduced predation rates ($\omega < 1$) relative to susceptible individuals.

The factor ω was estimated from the predation trials with susceptible and infected individuals of the Mid 37 clone; see Section S1.2 for details. The predation trials measured the number of *Daphnia* (out of 10) that were consumed. We converted these values to the proportion of individuals killed (i.e., number killed divided by 10). Because we could not detect signs of infection until day 10, we classified the replicates based on two factors. Factor 1 was infection status, which includes three treatments: (i) "control", meaning not exposed to spores, (ii) "exposed", meaning exposed to spores, but showing no visible signs of infection under a dissecting microscope, and (iii) "infected", meaning exposed to spores and showing visible signs of infection. Factor 2 was time block, which includes two treatments: (i) Block 1 is all measurements on days 1, 4, and 7 post exposure and (ii) Block 2 is all measurements on days 10 and 13 post exposure.

The data was analyzed using a two-way ANOVA in R; see accompanying code. Infection status was a significant factor ($F_{2,127} = 7.5$, $p < 0.001$), time block was not ($F_{1,127} = 0.76$, $p > 0.35$), and there was no significant interaction ($F_{1,127} = 0.53$, $p > 0.45$). We used the Tukey-Kramer Method to perform a multiple comparison test of means between infection status treatments. The differences between the mean for "infection" and the means for "exposed" and "control" were statistically significant (estimated difference between infected and control: 0.22, 95% CI: 0.06-0.38, $p = 0.004$; estimated difference between infected and exposed: 0.21, 95% CI: 0.05-0.37, $p = 0.006$) whereas the difference between the means for "exposed" and "control" was not statistically significant (estimated difference: 0.011, 95% CI: -0.09-0.12, $p > 0.95$); see Table S2.

We estimated ω using the data from the "control" and "infected" treatment. We did not use the data from the "exposed" treatment (exposed individuals who are not visibly infected) because our model does not include an exposed class and assumes all individuals are visibly infected immediately after infection. To estimate ω, we used equation (S6) to estimate the attack rates on susceptible and infected individuals,

$$\hat{a}_S = \frac{-1}{PT} \ln\left(1 - \frac{\sum_j N_{e,j}}{\sum_j N_{0,j}}\right) = 0.017/\text{pred/hr} \tag{S7}$$

$$\hat{a}_I = \frac{-1}{PT} \ln\left(1 - \frac{\sum_j N'_{e,j}}{\sum_j N'_{0,j}}\right) = 0.043/\text{pred/hr} \tag{S8}$$

where $P = 1$ individual is the number of predators in each replicate and $T = 15$ hours

is the length of the experiment. In the first equation, $\hat{a}_S$ is the estimated attack rate on susceptible individuals, $N_{0,j} = 10$ is the number of susceptible individuals at the start of replicate j, and $N_{e,j}$ is the number of susceptible individuals eaten in replicate j. Similarly, in the second equation, $\hat{a}_I$ is the estimated attack rate on visibly infected individuals, $N'_{0,j} = 10$ is the number of visibly infected individuals at the start of replicate j, and $N'_{e,j}$ is the number of visibly infected individuals eaten in replicate j. The values of $\sum_j N_{e,j} / \sum_j N_{0,j}$ and $\sum_j N'_{e,j} / \sum_j N'_{0,j}$ are the mean fractions of susceptible (control) and visibly infected individuals eaten, respectively. The values for those fractions are listed under the category "mean" in Table S2.

The ratio of the attack rates is $\hat{a}_I / \hat{a}_S = 2.5$. In addition, the difference between the attack rates is $\hat{a}_I - \hat{a}_S = 0.0258$/pred/hr. If we set the attack rates for infected individuals of clone i to $\hat{a}_i + 2.5$, that would be a doubling of the attack rate for the clone most susceptible to predation (BD05-42) and a quadrupling of the attack rate for the clone least susceptible to predation (CB24-68). Based on this and as a conservative estimate we set $\omega = 2$, which means the attack rate on infected individuals is twice that of susceptible individuals.

Table S2: Comparison of the mean fraction of individuals eaten during in the predation trials with susceptible and infected individuals of clone Mid 37. Groups were determined based on Tukey-Kramer Method comparisons; the differences between the mean for "infection" and the means for "exposed" and "control" were statistically significant (estimated difference between infected and control: 0.22, 95% CI: 0.06-0.38, $p = 0.004$; estimated difference between infected and exposed: 0.21, 95% CI: 0.05-0.37, $p = 0.006$) whereas the difference between the means for "exposed" and "control" was not statistically significant (estimated difference: 0.011, 95% CI: -0.09-0.12, $p > 0.95$).

Treatment	Mean	Group
Control	0.227	a
Exposed	0.24	a
Infected	0.475	b

Prey mortality rates, m_i and μ_i: We assume all prey clones have the same disease-induced morality rate. Disease-induced mortality occurs approximately 20 days after exposure, so the mortality rate is estimated to be 0.05/day = 0.0021/hour. Uninfected individuals can live for multiple months, which means that the rate of mortality due to factors other than disease (m_i) is much smaller. Consequently, we assume that the natural morality rate is negligibly small and set $m_i = 0$.

Liquid removal rate, λ: Liquid removal rate was 2L of 60L once per week. Thus, the average removal rate per hour is $2/60/7/24$ hr^{-1} = 0.0001986 hr^{-1}. This value is negligible compared to the spore degradation rate (0.0083 hr^{-1}) and prey mortality rate (0.0021 hr^{-1}). Consequently, we do not include it in our analyses.

Prey filtering rates, f_{S_i} and f_{I_i}: The filtering rates for susceptible individuals were

estimated in lab experiments (C.E. Cáceres and I. Menel, unpubl. data). Based on prior work on *D. dentifera* (Penczykowski et al., 2022; Searle et al., 2016), we assume that the filtering rates for infected individuals are half of the filtering rates for susceptible individuals ($f_{I_i} = 0.5 f_{S_i}$).

Per spore probability of infection, p_i: For the exposure experiments, each individual was put in 0.045L of lake water and exposed to a spore density of 200,000 spores/L for 24 hours. We model spore density in the experiment using the differential equation

$$dZ/dt = -f_i S Z \tag{S9}$$

where Z is the density of spores with initial condition $Z(0) = 200,000$ spores/L, f_i is the filtering rate of clone i in units of L/hr, and $S = 1/0.045$ indiv/L is the density of a single individual in 0.045L of lake water. The solution to the differential equation is $Z(t) = Z(0)\exp(-f_i S t)$, with units of spores/L. This means that the density of spores consumed by the individual in 24 hours is

$$Z(0) - Z(24) = Z(0)(1 - \exp(-f_i 24/0.045))\text{spores/L}. \tag{S10}$$

The total number of spores taken up in 24 hours is computed by multiplying by 0.045L. If the fraction of infected individuals across replicates is F_i, then the per spore probability of infection is

$$p_i = F_i / \underbrace{[Z(0)(1 - \exp(-f_i 24/0.045))0.045]}_{\text{spores consumed}}. \tag{S11}$$

Table S4 shows the values used to compute p_i.

Genotype-specific spore yield (burst size), χ_i: Spore yield from infected prey was estimated in lab experiments (C.E. Cáceres and I. Menel, unpubl. data). The value of χ_i for genotype BD19-64 was not measured. Because of this, we used the average of the other genotypes for its value.

Reduction in spore burst size, x_i: We did not measure within-host proliferation of spores in this study. The model in Auld et al. (2014) was used to estimate the reduction in burst size for infected individuals that were consumed by predators. In that study, the within-host density of spores was modeled as $dz/dt = r(1 - z/\sigma)$ where r is the maximum exponential replication rate of spores, σ is the maximum burst size, and $z(0) = z_0$ is the initial within-host density of spores. Thus, the within-host spore density is given by

$$z(t) = \frac{\sigma z_0 e^{rt}}{\sigma - z_0 + z_0 e^{rt}}. \tag{S12}$$

To use the model, we need to estimate r, z_0 and σ for each clone. We note a few things about this. First, spore density will never reach the maximum burst size (σ) in finite time. Consequently, we do not want to use the measured burst sizes (χ_i) as the estimate

for σ to avoid underestimating the burst size for individuals that are not consumed by predators ($P = 0$ treatment). Second, the parameterized model and empirical data in Auld et al. (2014) show that spore density reaches half of its maximum value about 13 days past exposure. Third, for the two different parasites studied in Auld et al. (2014), the estimated values of r were nearly identical.

We estimated the values of r, z_0 and σ for each clone in the following way. First, we assume the value of r was the same for all clones and set to the value ($0.5/\text{day} = 0.0208/\text{hr}$) estimated in Auld et al. (2014). Second, the values of z_0 and σ were chosen such that (i) the within-host density of spores is half of its maximum on day 13 (i.e., hour 314) and (ii) the burst size for an infected individual that dies on day 20 (i.e., hour 480) is equal to the measured values of χ_i. The first condition was motivated by the data and model in Auld et al. (2014) (see previous paragraph). The second condition is consistent with our assumption that infected individuals die 20 days after exposure. The values of z_0 and σ are given by solving the equations $z(312) = \sigma/2$ and $z(480) = \chi_i$, which yields

$$z_0 = \frac{\chi_i(e^{312r} + e^{480r})}{e^{480r}(1 + e^{312r})}, \quad \sigma = \frac{\chi_i(e^{312r} + e^{480r})}{e^{480r}}. \tag{S13}$$

For an infected individual of clone i that dies at time T, the reduction in burst size is given by $z(T)/\chi_i$. The average time to mortality in the absence of predators is $T = 1/\mu_i = 480$ hours. Consistent with our model assumptions, $z(480)/\chi_i = 1$, which means there is no reduction in spore burst size in the absence of predators. When predators are present at density P, the average time to mortality for an infected individual of clone i is $T = 1/(\mu_i + \omega a_i P)$. Thus, the reduction in spore burst size is $x_i = z(T)/\chi_i$ where $T = 1/(\mu_i + \omega a_i P)$. The estimates of z_0, σ, and x_i for all clones and predator density treatments are given in Table S5.

Spore degradation rate, δ: The spore degradation rate is taken from Strauss et al. (2015). The value in that study is $0.2\ \text{day}^{-1} = 0.0083\ \text{hr}^{-1}$.

Table S3: Summary of estimated parameters values for all prey clones

Parameter	Prey Genotype								
	BD05-42	BD08-46(81)	BD19-64(73)	CB24-68	DW22-58(84)	DW29-75	IL14-43	ML30-82	ML32-84
p_i	$5.54{\cdot}10^{-4}$	$5.64{\cdot}10^{-4}$	0	$5.25{\cdot}10^{-4}$	$6.88{\cdot}10^{-4}$	$5.77{\cdot}10^{-4}$	$1.17{\cdot}10^{-3}$	$8.58{\cdot}10^{-4}$	$4.93{\cdot}10^{-4}$
f_{S_i}	$1.44{\cdot}10^{-4}$	$1.74{\cdot}10^{-4}$	$9.94{\cdot}10^{-5}$	$1.552{\cdot}10^{-4}$	$2.46{\cdot}10^{-4}$	$2.65{\cdot}10^{-4}$	$1.54{\cdot}10^{-4}$	$5.8{\cdot}10^{-5}$	$2.78{\cdot}10^{-4}$
f_{I_i}	$7.2{\cdot}10^{-5}$	$8.7{\cdot}10^{-5}$	$4.97{\cdot}10^{-5}$	$7.76{\cdot}10^{-5}$	$1.23{\cdot}10^{-4}$	$1.33{\cdot}10^{-4}$	$7.7{\cdot}10^{-5}$	$2.9{\cdot}10^{-5}$	$1.39{\cdot}10^{-4}$
μ_i	0.00208	0.00208	0.00208	0.00208	0.00208	0.00208	0.00208	0.00208	0.00208
a_i	0.0197	0.0134	0.0116	0.0062	0.0144	0.0082	0.016	0.0157	0.0096
ω	2	2	2	2	2	2	2	2	2
χi	78231	36990	83034 †	70578	89909	146128	108163	41667	92610
x_i for $P = 0.1$	0.047	0.1098	0.1501	0.4558	0.0937	0.2968	0.0741	0.0773	0.2213
x_i for $P = 0.5$	0.004	0.0059	0.0071	0.0188	0.0055	0.0116	0.0049	0.005	0.0091
x_i for $P = 1$	0.0026	0.0032	0.0035	0.0065	0.0030	0.0048	0.0028	0.0029	0.0041

† Value was not measured. Listed value is the average of the values for the other genotypes

Parameter values are defined in Table 1 of the main text.

The per spore probabilities of infection (p_i) are computed in equation (S11)

Filtering rates for infected individuals (f_{I_i}) are half of that for susceptible individuals (f_{S_i})

Attack rates (a_i) are computed using equation (S6)

Increase in attack rate for infected individuals (ω) is computed using equations (S7) and (S8)

Reductions in burst size (x_i) are computed using equations (S12) and (S13)

Table S4: Values used to calculate per spore probabilities of infection

	Prey Genotype								
	BD05-42	BD08-46(81)	BD19-64(73)	CB24-68	DW22-58(84)	DW29-75	IL14-43	ML30-82	ML32-84
Fraction infected (F_i)	0.3684	0.45	0	0.375	0.7619	0.6842	0.8333	0.2353	0.6111
Filtering rate (f_{S_i})	$1.44{\cdot}10^{-4}$	$1.74{\cdot}10^{-4}$	$9.94{\cdot}10^{-5}$	$1.55{\cdot}10^{-4}$	$2.46{\cdot}10^{-4}$	$2.65{\cdot}10^{-4}$	$1.54{\cdot}10^{-4}$	$5.8{\cdot}10^{-5}$	$2.78{\cdot}10^{-4}$
Spores consumed ($Z(0) - Z(24)$)	665	798	465	714	1107	1186	710	274	1240
Infection probability (p_i)	$5.54{\cdot}10^{-4}$	$5.64{\cdot}10^{-4}$	0	$5.25{\cdot}10^{-4}$	$6.88{\cdot}10^{-4}$	$5.77{\cdot}10^{-4}$	$1.17{\cdot}10^{-3}$	$8.58{\cdot}10^{-4}$	$4.93{\cdot}10^{-4}$

Values are computed using equations (S9)-(S11)

Table S5: Values used to calculate reduction in burst size for consumed infected individuals

	Prey Genotype								
	BD05-42	BD08-46(81)	BD19-64(73)	CB24-68	DW22-58(84)	DW29-75	IL14-43	ML30-82	ML32-84
Estimated burst size (χ_i)	78231	36990	83034$*$	70578	89909	146128	108163	41667	92610
Maximum burst size (σ_i)	$8.06{\cdot}10^4$	$3.81{\cdot}10^4$	$8.55{\cdot}10^4$	$7.27{\cdot}10^4$	$9.26{\cdot}10^4$	$1.51{\cdot}10^5$	$1.11{\cdot}10^5$	$4.29{\cdot}10^4$	$9.54{\cdot}10^4$
Initial density (z_0)	121	57	128	109	139	226	167	64	143
Reduction for $P = 0.1$ (x_i)	0.047	0.1098	0.1501	0.4558	0.0937	0.2968	0.0741	0.0773	0.2213
Reduction for $P = 0.5$ (x_i)	0.004	0.0059	0.0071	0.0188	0.0055	0.0116	0.0049	0.005	0.0091
Reduction for $P = 1$ (x_i)	0.0026	0.0032	0.0035	0.0065	0.0030	0.0048	0.0028	0.0029	0.0041

Values were computing using equations (S12) and (S13).

Section S3.3 Computing the parasite reproduction numbers

The basic reproduction number ($\mathcal{R}_0$) and the reproduction number ($\mathcal{R}$) are computed using the next generation matrix (NGM) (Van den Driessche and Watmough, 2008; Diekmann et al., 2010). The reproduction number is equal to the basic reproduction number whenever all individuals in the population are susceptible (i.e., $N = S + I = S$ because $I = 0$). Throughout this section we assume loss due to volume removal is negligible ($\lambda = 0$).

We start by giving the equations for $\mathcal{R}_0$ and $\mathcal{R}$ when we have measurements of total density (N_i) and infected density (I_i) for each clone. For a single-clone population with total density N_i, the basic reproduction number is

$$\mathcal{R}_{0,i}(N_i) = \frac{\chi_i\,(m_i + \mu_i + \omega x_i a_i P)}{(m_i + \mu_i + \omega a_i P)} \frac{p_i f_{S_i} N_i}{(\delta + f_{S_i} N_i)} \tag{S14}$$

The basic reproduction number for a multi-clone system with densities $N_1, ..., N_n$ is

$$\mathcal{R}_0(N_1, ..., N_n) = \sum_i \frac{\chi_i\,(m_i + \mu_i + \omega x_i a_i P)}{(m_i + \mu_i + \omega a_i P)} \frac{p_i f_{S_i} N_i}{(\delta + \sum_j f_{S_j} N_j)}. \tag{S15}$$

The reproduction number is similarly computed to be

$$\mathcal{R}(N_1, ..., N_n, I_1, ..., I_n) = \sum_i \frac{\chi_i\,(m_i + \mu_i + \omega x_i a_i P)}{(m_i + \mu_i + \omega a_i P)} \frac{p_i f_{S_i} (N_i - I_i)}{(\delta + \sum_j [f_{S_j}(N_j - I_j) + f_{I_j} I_j])} \tag{S16}$$

where the densities of susceptible individuals are $S_i = N_i - I_i$.

To get the equations for $\mathcal{R}_0$ and $\mathcal{R}$ in equations (2.a) and (2.b) of the main text, we write the formulas in terms of the total density of all clones ($N = \sum_i N_i$), the total density of infected individuals ($I = \sum_i I_i$), and the clone frequencies ($q_1, ..., q_n$). In particular, the formulas for $\mathcal{R}_0$ and $\mathcal{R}$ are rewritten using $N_i = p_i N$, $I_i = p_i I$, and $S_i = p_i(N - I)$. Note that equations (2.a) and (2.b) of the main text assume that the frequency distributions for susceptible individuals and infected individuals are the same.

Section S4 Supplemental results for models

Section S4.1 Partitioning effects of ecology and evolution on changes in $\mathcal{R}_0$ and $\mathcal{R}$

Motivation: Figure 2 shows that the calculated values of $\mathcal{R}_0$ and $\mathcal{R}$ changed between weeks 0 and 2. These changes are due to ecological changes (i.e., changes in prey density) and evolutionary changes (i.e., changes in clone frequencies). Our goal here to assess whether changes in the values of $\mathcal{R}_0$ and $\mathcal{R}$ were primarily driven by ecological changes, evolutionary changes, or both.

Method: We use the Geber Method (Hairston Jr et al., 2005) to partition the changes in $\mathcal{R}_0$ and $\mathcal{R}$ into contributions from ecological change (i.e., change in prey densities) and

contributions from evolutionary change (i.e., changes in clone frequencies). The formulas for the Geber Method are set up in the following way. Measurements are taken at time points $t_1, ..., t_m$. Let N_{t_k} denote the total prey density at time t_k, I_{t_k} denote the total density of infected individuals at time t_k, and $\vec{q}_{t_k} = (q_{1,t_k}, ..., q_{n,t_k})$ denote the vector of clone frequencies at time t_k. Following Hairston Jr et al. (2005), the contribution of ecological processes to changes in $\mathcal{R}_0$ is computed using the formula,

$$\Delta_{eco}(t_k, t_{k+1}) = \frac{\mathcal{R}_0(N_{t_{k+1}}, \vec{q}_{t_k}) - \mathcal{R}_0(N_{t_k}, \vec{q}_{t_k})}{2(t_{k+1} - t_k)} + \frac{\mathcal{R}_0(N_{t_{k+1}}, \vec{q}_{t_{k+1}}) - \mathcal{R}_0(N_{t_k}, \vec{q}_{t_{k+1}})}{2(t_{k+1} - t_k)}. \quad \text{(S17)}$$

The contribution of evolutionary (i.e., changes in clone frequencies) to changes in $\mathcal{R}$ is computed using the formula,

$$\Delta_{evo}(t_k, t_{k+1}) = \frac{\mathcal{R}_0(N_{t_k}, \vec{q}_{t_{k+1}}) - \mathcal{R}_0(N_{t_k}, \vec{q}_{t_k})}{2(t_{k+1} - t_k)} + \frac{\mathcal{R}_0(N_{t_{k+1}}, \vec{q}_{t_{k+1}}) - \mathcal{R}_0(N_{t_{k+1}}, \vec{q}_{t_k})}{2(t_{k+1} - t_k)}. \quad \text{(S18)}$$

The formulas for the contributions of ecological and evolutionary processes to changes in $\mathcal{R}$ are identical, except that all instances of $\mathcal{R}_0(N_{t_k}, \vec{q}_{t_j})$ are replaced with $\mathcal{R}(N_{t_k}, I_{t_k}, \vec{q}_{t_j})$. The denominators of both formulas include $t_{k+1} - t_k$ to account for uneven inter-sampling times. Consequently, both formulas report the effects of ecology and evolution in terms of standardized rates.

In the formulas, $\mathcal{R}_0(N_{t_k}, \vec{q}_{t_k})$ are "real" values of $\mathcal{R}_0$ computed from the empirical densities and frequencies measured at the same time point. In comparison, $\mathcal{R}_0(N_{t_{k+1}}, \vec{q}_{t_k})$ and $\mathcal{R}_0(N_{t_k}, \vec{q}_{t_{k+1}})$ are hypothetical values of $\mathcal{R}_0$ computed from densities and frequencies measured at different time points. Equation (S17) represents the effect of ecology because it computes the changes in $\mathcal{R}_0$ that would occur if the total prey density could change from N_{t_k} to $N_{t_{k+1}}$, but the clone frequencies were held fixed at the values at the first time point ($\vec{q}_{t_k}$) or the second time point ($\vec{q}_{t_{k+1}}$). Positive and negative values mean that changes in prey density caused $\mathcal{R}_0$ to increase and decrease, respectively. Likewise, equation (S18) represents the effect of evolution because it computes the changes in $\mathcal{R}_0$ that would occur if the clone frequencies could change from $\vec{q}_{t_k}$ to $\vec{q}_{t_{k+1}}$, but the total prey densities were held fixed at the values at the first time point (N_{t_k}) or the second time point ($N_{t_{k+1}}$). Positive and negative values mean that changes in prey density caused $\mathcal{R}_0$ to increase and decrease, respectively. Ecological processes have larger effects than evolutionary processes when Δ_{eco} is larger in magnitude than Δ_{evo}, and vice versa.

Results: We applied equations (S17) and (S18) to the values of $\mathcal{R}_0$ and $\mathcal{R}$ computed at weeks 0 and 2 of our experiment. We did not apply the equations to later weeks because the estimates of $\mathcal{R}_0$ and $\mathcal{R}$ may not be accurate due to changes in predator densities (which affects the parameter x_i). The results are shown in Figure S3. Positive ($\Delta_{eco} > 0$) and negative ($\Delta_{eco} < 0$) values in Figure S3a,b mean the changes in prey densities caused $\mathcal{R}_0$ and $\mathcal{R}$ to increase and decrease, respectively. Positive ($\Delta_{evo} > 0$) and negative ($\Delta_{evo} < 0$) values in Figure S3c,d mean the changes in clone frequencies

caused $\mathcal{R}_0$ and $\mathcal{R}$ to increase and decrease, respectively. In Figure S3e,f, values above and below the dashed purple line imply that ecological processes have larger and smaller effects, respectively, than evolutionary processes.

For the lower predation treatments, the increases in prey densities caused the values of $\mathcal{R}_0$ and $\mathcal{R}$ to increase (all dots of all shades of blue have positive values in Figure S3a,b). In addition, the effects of ecology were generally larger in magnitude than the effects of evolution (all dots of all shades of blue are above the dashed purple line in Figure S3e,f). The only exceptions are two replicates of the 0.5 predator/L treatment (one blue dot on and one blue dot below the purple line in Figure S3f).

For the highest predation treatments, the changes in $\mathcal{R}_0$ and $\mathcal{R}$ were small in magnitude between weeks 0 and 2. As a consequence, the effects of changes in prey densities and changes in clone frequencies are also small in magnitude (all black dots are close to 0 in Figure S3a-d). Overall, this means that ecology and evolution had small effects of roughly similar magnitude on the changes in $\mathcal{R}_0$ and $\mathcal{R}$ in the highest predation treatment.

To explore if our results were sensitive to the assumption that predators had higher attack rates on infected prey than susceptible prey, we computed $\mathcal{R}_0$ and $\mathcal{R}$ assuming $\omega = 1$ and applied the Geber Method equations (S17) and (S18). Figure S4 shows that the values of $\mathcal{R}_0$ and $\mathcal{R}$ change in similar ways when predators are assumed to have equal attacks rates on susceptible and infected prey. In addition, Figure S5 shows that our results from the Geber Method are qualitatively unchanged. One important difference between the values of $\mathcal{R}_0$ in Figures 2 and S4 is that none of the values of $\mathcal{R}_0$ are below 1 when predators have equal attack rates on susceptible and infected prey (all points at week 0 above the purple line in Figure S4a). Because outbreaks did not occur in the highest predation treatments and the values of $\mathcal{R}_0$ are below 1 for those treatments when predators are assumed to have higher attack rates on infected prey, this is indirect support for the assumption that predators have higher predation rates on infected prey.

Section S4.2 Healthy herds hypothesis: predictions about total prey density

Motivation: The healthy herds hypothesis predicts that for a prey population that is suppressed by both a parasite and a predator, if regulation of the prey population by the parasite is greater than regulation by the predator, then predator removal can cause the prey population to decrease. In this case, the predator-prey interaction suppresses the parasite, which allows the prey population to remain at higher abundances than would be possible in the absence of the predator.

Our experimental results do not allow us to determine if the above prediction applies to our system. This is because (i) the parasite went extinct in all of the highest predation treatments and (ii) the variation in *Daphnia* densities across the lower predation treatments is similar to the variation in *Daphnia* densities in each of the lower predation treatments. Here, we use the mathematical model to assess whether our parameter estimates provide indirect support for the above hypothesis.

Model and Approximations: We analyze a single-clone version of the multi-clone model (1.a-c) from the main text. We analyze a single-clone version of the model because an equivalent analysis of a multi-clone model would require a full parameterization of the reproduction functions for each clone (G_i), which we do not have. To facilitate the analysis, we convert the model from a SI form that tracks susceptible (S) and infected (I) prey densities to an NY form that tracks the total density of prey (N), infection prevalence (i.e., the proportion of infected prey, $Y = I/N$), and spore density (Z). The equation for infection prevalence is derived using the quotient rule from calculus: $dY/dt = d(I/N)/dt = (1/N)(dI/dt) - (I/N)(dN/dt)$.

The model equations are

$$\begin{aligned}
\frac{dN}{dt} &= \overbrace{Ng(N)}^{\text{reproduction}} \overbrace{-\overline{m}N}^{\text{non-disease mortality}} \overbrace{-\overline{a_S}(1-Y)NP - \overline{a_I}YNP}^{\text{predation}} - \overbrace{\overline{\mu}YN}^{\text{disease mortality}} \\
\frac{dY}{dt} &= \overbrace{\overline{pf}(1-Y)Z}^{\text{infection}} - \overbrace{(\overline{m}+\overline{\mu})Y}^{\text{non-disease \& disease mortality}} - \overbrace{\overline{a_I}YP}^{\text{predation}} - \frac{Y}{N}\frac{dN}{dt} \\
\frac{dZ}{dt} &= \overbrace{(\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P)NY}^{\text{shedding}} - \overbrace{\overline{f}NZ}^{\text{uptake}} - \overbrace{\delta Z}^{\text{degradation}}
\end{aligned} \tag{S19}$$

The notation is similar to the clonal model from the main text with the key difference being that overlines denote values averaged across the population. In particular, $g(N)$ is the per capita growth rate of the population (averaged across all clones); $\overline{m}$ is the average mortality rate due to causes other than disease and predation; $\overline{a_S}$ and $\overline{a_I} = \omega\overline{a_S}$ are the average predation rates for susceptible and infected individuals, respectively; $\overline{\mu}$ is the average mortality rate due to disease; $\overline{pf}$ is the average rate at which susceptible individuals become infected, which accounts for the differences in the filtering rates and probabilities of infection across clones; $\overline{\chi m}$ is the average excretion rate for individuals who die due to causes other than disease and predation; $\overline{\chi\mu}$ and $\overline{\chi a_I x(P)}$ are the average excretion rates for individuals who die due to disease and predation, respectively; and $\overline{f}$ is the average filtering rate. To simplify the notation, the liquid removal rate (λ) has been absorbed into the non-disease mortality rates and the spore degradation rate. The average values are computed using the clonal frequencies, $q_i, ..., q_n$. For example, $\overline{a_I} = \sum_i a_{I_i} q_i$ and $\overline{\chi\mu} = \sum_i \chi_i \mu_i q_i$. Specific choices for the frequencies are discussed later.

We note the following about model (S19). First, all of the assumptions about the clonal model (1.a-c) in the main text also apply to model (S19) with one exception. The one exception is that model (S19) assumes susceptible and infected individuals have equal uptake rates. This assumption is necessary in order for the analytical calculations to remain tractable. Second, we assume the attack rates on infected individuals are twice the attack rates on susceptible individuals for all clones ($\omega = 2$). To simplify the equations, we use the more condense notation $\overline{a_I} = \omega\overline{a_S}$ and $\overline{\chi a_I x(P)} = \omega\overline{\chi a_S x(P)}$. Third, the function g defines the growth of the population and accounts for intraspecific competition. We assume

g is a decreasing function, i.e., $g'(N) < 0$. We cannot parameterize the per capita growth rate function $g(N)$ because we do not have estimates of the growth rates and competitive abilities for all of the prey clones.

Fourth, the notation $\overline{\chi a_I x(P)}$ is used to explicitly denote that the release rate of infected individuals who are consumed by predators depends on predator density. In particular, the reduction in burst size for infected individuals of clone i that are consumed (x_i) is computed by replacing the value of t in equation (S12) with the average lifespan of an infected individual, $1/(\mu_i + \omega a_i P)$, which yields

$$x_i(P) = \frac{1}{\chi_i} \frac{\sigma_i z_{0,i} \exp(r/[\mu_i + a_{I,i}P])}{\sigma - z_{0,i} + z_{0,i} \exp(r/[\mu_i + a_{I,i}P])} \tag{S20}$$

where r is the maximum replication rate of spores within an infected individual (assumed to be the same for all clones) and for clone i, χ_i is the burst size of infected individuals that are not consumed, σ_i is the maximum burst size, μ_i is the disease-induced mortality rate, and $a_{I,i}$ is the predation attack rate on infected individuals.

Finally, in the calculations that follow, it is useful to know that

$$\frac{dx_i}{dP} = -x_i(P) \frac{r a_{I,i}(\sigma - z_0)}{[\sigma - z_{0,i} + z_{0,i} \exp(r/[\mu_i + a_{I,i}P])] \left[a_{I,i}P + \mu\right]^2} < 0. \tag{S21}$$

Biologically, this means that increases in predator density cause greater decreases in the spore burst size of consumed infected individuals of clone i. This occurs because greater predator density means an infected individual is more likely to die sooner after infection, which results in less time for within-host replication of the spores. Because increased predator density leads to decreased spore burst sizes of consumed infected individuals for all clones, the average burst size for consumed infected individuals also decreases with increased predator density. Said mathematically, $d\overline{\chi a_I x(P)}/dP < 0$ because $\overline{\chi a_I x(P)} = \sum_i \chi_i a_{I_i} x_i(P) q_i$ and the previous equation shows $dx_i(P)/dt < 0$ for all i.

Responses in total prey density to increased predator density: Let $p^* = (N^*, Y^*, Z^*)$ be an equilibrium of model (S19) where the prey and parasite stably coexist. Our goal is to compute how the total population size at equilibrium, N^*, changes in response to increased predation (P). We cannot directly compute the total population size at equilibrium because we do not have estimates for the growth function $g(N)$. Nonetheless, we can compute how the total prey population size at equilibrium changes as predator density is varied. This is done by computing the derivative $\partial N^*/\partial P$. If $\partial N^*/\partial P$ is negative, then increased predator density results in lower prey density. This outcome does not support the healthy herds hypothesis because it suggests that prey density will be highest when the predator is absent. If $\partial N^*/\partial P$ is positive, then increased predator density results in higher prey density. This outcome supports the healthy herds hypothesis because it suggests that prey density will be lower when the predator is absent in comparison to when the predator is present.

To compute the derivative, set all equations in model (S19) equal to zero and simplify the second equation using $dN/dt = 0$. Solving the dZ/dt equation for the equilibrium spore

density yields $Z^* = (\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P)N^*Y^*/(\overline{f}N^* + m)$. Substituting Z^* into the second equation and solving for the equilibrium infection prevalence, Y^*, yields

$$Y^* = 1 - \frac{\overline{f}N^* + \delta}{N^*\overline{pf}} \cdot \frac{\overline{\mu} + \overline{m} + \overline{a_I}P}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P}. \tag{S22}$$

Substituting Y^* into the dN/dt equation yields

$$0 = g(N^*) - \overline{m} - \overline{a_S}P + (\overline{a_S}P - \overline{a_I}P - \overline{\mu})\left(1 - \frac{\overline{f}N^* + \delta}{N^*\overline{pf}} \frac{\overline{\mu} + \overline{m} + \overline{a_I}P}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P}\right). \tag{S23}$$

Implicitly differentiating the equation with respect to P and solving for $\partial N^*/\partial P$ results in

$$\begin{aligned}\frac{\partial N^*}{\partial P} =& \frac{1}{g' + (\overline{a_S}P - \overline{a_I}P - \overline{\mu})\left(\frac{\overline{\mu}+\overline{m}+\overline{a_I}P}{\overline{\chi\mu}+\overline{\chi m}+\overline{\chi a_I x(P)}P}\frac{\delta}{\overline{pf}(N^*)^2}\right)} \times \\ &\left[\overline{a_S}(1-Y) + \overline{a_I}Y + (\overline{a_S}P - \overline{a_I}P - \bar{\mu})\frac{\overline{f}N^* + \delta}{\overline{pf}N^*} \cdot \frac{\overline{a_I}(\overline{\chi\mu} + \overline{\chi m}) - \overline{\chi a_I x(P)}(\overline{\mu} + \overline{m})}{(\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P)^2}\right. \\ &\left. -(\overline{a_S}P - \overline{a_I}P - \overline{\mu})\frac{\overline{f}N^* + \delta}{\overline{pf}N^*}\frac{(\overline{m} + \overline{\mu} + \overline{a_I}P)}{(\overline{\chi m u} + \overline{\chi m} + \overline{\chi a_I x(P)}P)^2}\frac{d\overline{\chi a_I x(P)}}{dP}P\right].\end{aligned} \tag{S24}$$

With some algebraic manipulation, the equilibrium conditions $dZ/dt = 0$ and $dY/dt = 0$ can be combined and rearranged to yield

$$1 = \frac{\overline{f}N + \delta}{\overline{pf}N} \cdot \frac{\overline{m} + \overline{\mu} + \overline{a_I}P + \overline{pf}Z}{\overline{\chi m} + \overline{\chi\mu} + \overline{\chi a_I x(P)}P}. \tag{S25}$$

Substituting into equation (S24) produces

$$\frac{\partial N^*}{\partial P} = \overbrace{\frac{1}{g' + \underbrace{(\overline{a_S}P - \overline{a_I}P - \overline{\mu})}_{\text{Factor 1.1}} \underbrace{\left(\frac{\overline{\mu} + \overline{m} + \overline{a_I}P}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P} \frac{\delta}{\overline{pf}(N^*)^2} \right)}_{\text{Factor 1.2}}}}^{\text{Term 1}} \times$$

$$\Bigg[\overbrace{\overline{a_S}(1-Y)}^{\text{Term 2}} + \overbrace{\overline{a_I}Y}^{\text{Term 3}} + \overbrace{-\overline{a_I} \cdot \underbrace{\frac{\overline{\chi\mu} + \overline{\chi m}}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P}}_{\text{Factor 4.1}} \cdot \underbrace{\frac{\overline{\mu} - \overline{a_S}P + \overline{a_I}P}{\overline{\mu} + \overline{m} + \overline{a_I}P + \overline{pf}Z}}_{\text{Factor 4.2}}}^{\text{Term 4}} \tag{S26}$$

$$+ \overbrace{\underbrace{\frac{\overline{\chi a_I x(P)}(\overline{\mu} + \overline{m})}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P}}_{\text{Factor 5.1}} \cdot \underbrace{\frac{\overline{\mu} - \overline{a_S}P + \overline{a_I}P}{\overline{\mu} + \overline{m} + \overline{a_I}P + \overline{pf}Z}}_{\text{Factor 5.2}}}^{\text{Term 5}}$$

$$+ \overbrace{\underbrace{\frac{\overline{\mu} + \overline{m} + \overline{a_I}P}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P}}_{\text{Factor 6.1}} \cdot \underbrace{\frac{\overline{\mu} - \overline{a_S}P + \overline{a_I}P}{\overline{\mu} + \overline{m} + \overline{a_I}P + \overline{pf}Z}}_{\text{Factor 6.2}} \underbrace{\frac{d\overline{\chi a_I x(P)}}{dP}P}_{\text{Factor 6.3}}}^{\text{Term 6}} \Bigg].$$

The magnitudes and signs of the factors and terms in equation (S26) are listed below.

Term 1: Term 1 is likely to be negative in most systems for three reasons. First, g' is assumed to be negative. Second, Factor 1.2 is positive. Third, Factor 1.1 is likely to be negative in most systems. The reasoning is that Factor 1.1 can be written as $(\overline{a_S}P - \overline{a_I}P - \overline{\mu}) = -(g(N) - \overline{m} - \overline{a_S}P)/Y$ using the equilibrium condition $dN/dt = 0$. The sum $g(N) - \overline{m} - \overline{a_S}P$ is negative only if (i) prey density is sufficiently large such that $g(N) \approx 0$, (ii) infection prevalence is very high ($Y \approx 1$), and (iii) the predation rates for susceptible individuals are much larger than those for infected individuals ($\overline{a_S} \gg \overline{a_I}$). Because these conditions are unlikely to be met in most empirical systems, we expect that Factor 1.1 will be negative. This results in Term 1 being negative.

Term 2: Term 2 is positive. It is smaller in magnitude when predators have lower attack rates on susceptible individuals (smaller $\overline{a_S}$) and infection prevalence is high (Y closer to 1).

Term 3: Term 3 is positive. It is smaller in magnitude when predators have lower attack rates on infected individuals and infection prevalence is low (Y small).

Term 4: Term 4 is always smaller in magnitude than $\overline{a_I}$ and it is likely to be negative in most systems. The justification is the following. First, Factor 4.1 is positive and less than 1 (because the numerator is smaller than the denominator). Factor 4.1 is larger if infected individuals have smaller burst sizes (small x).

Second, Factor 4.2 is smaller than 1 in magnitude and likely to be positive. To see this, use the equilibrium conditions $dN/dt = 0$ and $0 = dS/dt = Ng(N) - \overline{m}N(1-Y) - \overline{a_S}(1-Y)NP - \overline{pf}ZN(1-Y)$ to rewrite Factor 4.2 as

$$\frac{\overline{\mu} - \overline{a_S}P + \overline{a_I}P}{\overline{\mu} + \overline{m} + \overline{a_I}P + \overline{pf}Z} = \frac{(g(N) - \overline{m} - \overline{a_S}P)/Y}{(g(N) - \overline{m}(1-Y) - \overline{a_S}P(1-Y))/Y + g(N)/(1-Y) - \overline{m}N - \overline{a_S}P} \tag{S27}$$

$$= \frac{g(N) - \overline{m} - \overline{a_S}P}{g(N) - \overline{m} - \overline{a_S}P + g(N)Y/(1-Y)}. \tag{S28}$$

The text about Term 1 explains why the numerator and denominator are likely to be positive in most systems. Factor 4.2 must be smaller than 1 in magnitude because the numerator of the previous equation is smaller than the denominator.

Term 5: Term 5 is likely to be positive in most systems. This is because Factor 5.1 is positive and Factor 5.2 is likely to be positive in most systems (see explanation about Factor 4.2).

Term 6: Term 6 is likely to be negative in most systems. This is because Factor 6.1 is positive, Factor 6.2 is likely to be positive in most systems (see explanation about Factor 4.2), and Factor 6.3 is negative (see equation (S21) and surrounding text).

Altogether, for most systems we expect $\overline{\mu} - \overline{a_S}P + \overline{a_I}P > 0$, which implies Term 1 is negative, Terms 2, 3, and 5 are positive, Term 6 is negative, and Term 4 is negative and smaller in magnitude than $\overline{a_I}$. In this case, the healthy herds hypothesis is supported, i.e., equation (S26) is positive, only if Terms 4 and 6 are large in magnitude and Terms 1, 2, and 5 are small in magnitude.

Responses in total prey density to increased spore density: To determine how equilibrium prey density is affected by the parasite, we compute how the equilibrium prey density, N^*, changes with increased spore density, Z^*. This is done by computing the derivatives

$$\frac{dN^*}{dZ^*} = \frac{\partial N^*}{\partial \delta} \Bigg/ \frac{\partial Z^*}{\partial \delta} \tag{S29}$$

where $\frac{\partial N^*}{\partial \delta}$ and $\frac{\partial Z^*}{\partial \delta}$ define how the prey and spore equilibrium densities respond to small increases in the spore degradation rate. If $\partial N^*/\partial P$ is negative, then increased spore density results in lower prey density. If $\partial N^*/\partial P$ is positive, then increased spore density results in higher prey density.

The derivative $\partial N^*/\partial \delta$ is computed by implicitly differentiating equation (S23) and solving, which yields

$$\frac{\partial N^*}{\partial \delta} = \underbrace{\frac{1}{g' + (\overline{a_S}P - \overline{a_I}P - \overline{\mu})\left(\frac{\overline{\mu}+\overline{m}+\overline{a_I}P}{\overline{\chi\mu}+\overline{\chi m}+\overline{\chi a_I x(P)}P}\frac{\delta}{\overline{pf}(N^*)^2}\right)}}_{\text{Factor 1}} \cdot \underbrace{\frac{\overline{a_S}P - \overline{a_I}P - \overline{\mu}}{\overline{pf}N^*}}_{\text{Factor 2}} \cdot \underbrace{\frac{\overline{\mu} + \overline{m} + \overline{a_I}P}{\overline{\chi\mu} + \overline{\chi m} + \overline{\chi a_I x(P)}P}}_{\text{Factor 3}} \cdot \tag{S30}$$

Factor 1 is expected to be negative in most systems; see the text about Term 1 for equation (S26). For the same reason, Factor 2 is expected to be negative for most systems. Factor 3 is always positive. In total, we expect $\partial N^*/\partial \delta$ to be positive for most systems.

The derivative $\partial Z^*/\partial\delta$ is computed using the Jacobian-based framework developed by Bender et al. (1984) and (Yodzis, 1988). Let $\mathbf{J}$ be the Jacobian of model (S19) and let $\mathbf{M}$ denote the submatrix of the Jacobian where the third row and column have been removed. After some straightforward algebraic manipulation, the derivative simplifies to

$$\frac{\partial Z^*}{\partial \delta} = \left(-\frac{\partial}{\partial \delta}\frac{dZ}{dt}\right)\frac{(-1)^{3+3}|\mathbf{M}|}{|\mathbf{J}|} \tag{S31}$$

$$= \frac{-Z^*N^*g'(N^*)(\overline{pf}Z^* + \overline{m} + \overline{\mu} + \overline{a_I}P)}{|\mathbf{J}|} < 0. \tag{S32}$$

where $|\mathbf{J}|$ and $|\mathbf{M}|$ are the determinants of the Jacobian and the submatrix. The determinant of the Jacobian is negative, i.e., $|\mathbf{J}| < 0$, because we assume the equilibrium is stable. We also assume $g' < 0$. Consequently, $\partial Z^*/\partial\delta$ is always negative and increased spore degradation always causes the equilibrium spore density to decrease.

Combining the above, we predict that increases in equilibrium spore density will cause a decrease in prey density ($dN^*/dZ^* < 0$) for most systems.

Interpretation: All of the following results assume $\overline{\mu} - \overline{a_S}P + \overline{a_I}P > 0$. We expect this condition to be met in most systems. The only exceptions are systems where prey density is very close to its carrying capacity ($g(N) \approx 0$), infection prevalence is very high ($Y \approx 1$), and the predation rates for susceptible individuals are much larger than those for infected individuals ($\overline{a_S}$ much larger than $\overline{a_I}$). In combination, this would mean that the predator and parasite suppress prey density to very low densities and most of the prey population is infected by the parasite; we expect this situation to be rare in natural systems. Thus, we focus on systems where $\overline{\mu} - \overline{a_S}P + \overline{a_I}P > 0$.

Recall that total prey density increases with increased predator density ($\partial N^*/\partial P > 0$; equation (S26) positive) only if the negative Terms 4 and 6 are large in magnitude and the positive Terms 2, 3, and 5 are small in magnitude. From this, we predict that increased prey density with increased predator density is more likely to occur if

(i) Predators have higher attack rates on infected individuals than susceptible individuals ($\overline{a_I} > \overline{a_S}$)

(ii) Consumed individuals have smaller spore burst sizes ($x_i < 1$) and spore burst size decreases with increased predation ($d\overline{\chi a_i x(P)}/dP < 0$). The latter means that infected individual life span decreases as predator density increases.

(iii) Infection prevalence is low (Y closer to 0 rather than 1)

Our results about responses to increased spore density (dN^*/dZ^*) show that increased spore density always causes prey density to decrease.

Let us denote the presence and absence of predators and parasites using plus and minus signs. For example, $N^*(-\text{parasite},+\text{predator})$ is the equilibrium prey density when the predator is present and the parasite is absent. Combining the above yields the following predictions about prey density in the presence/absence of parasites and predators.

First, consider systems where (i) predators have a sufficiently higher attack rate on infected prey than susceptible prey ($\overline{a_I} > \overline{a_S}$), (ii) consumed prey have sufficiently smaller burst sizes than prey that are not consumed ($x < 1$), and (iii) infection prevalence is sufficiently low (Y sufficiently small). Then, increased predator density increases prey density and increased parasite density decreases prey density. This means

$$\begin{aligned} N^*(-\text{parasite},-\text{predator}) &> N^*(-\text{parasite},+\text{predator}) \\ &> N^*(+\text{parasite},+\text{predator}) > N^*(+\text{parasite},-\text{predator}) \end{aligned} \tag{S33}$$

That is, prey density is highest in the absence of the predator and parasite, lower when only the predator is present, even lower when both the parasite and predator are present, and lowest when only the parasite is present. In this case, the healthy herds hypothesis is supported because predation causes an increase in prey density by reducing the suppressing effects of the parasite.

Now consider systems where conditions (i)-(iii) are not met. In these systems, increased predator density decreases prey density and increased parasite density decreases prey density. This means

$$N^*(-\text{parasite},-\text{predator}) > N^*(+\text{parasite},-\text{predator}) > N^*(+\text{parasite},+\text{predator}) \tag{S34}$$

and

$$N^*(-\text{parasite},-\text{predator}) > N^*(-\text{parasite},+\text{predator}) > N^*(+\text{parasite},+\text{predator}). \tag{S35}$$

That is, prey density is highest in the absence of the predator and parasite, lowest when the predator and parasite are both present, and intermediate when either the parasite or the predator is present. Note that without a complete parameterization of the prey growth function, $g(N)$, we cannot determine if prey density is higher when only the predator is present or only the parasite is present. In this case, the healthy herds hypothesis is not supported because predation always causes a decrease in prey density.

Predictions for empirical system: In order to make predictions about the *D. dentifera-M. bicuspidata-C. punctipennis* system, we applied the above theory in the following way. We computed the sign of $\partial N^*/\partial P$ by evaluating the negative of the numerator of equation (S26). We only used the numerator because we cannot compute the denominator of equation (S26) without a parameterization for the prey growth rate, $g(N)$. The negative sign was used to account for the fact that the denominator of equation (S26) is expected to be negative. Note that because the denominator of equation (S26) is always negative, equation (S26) only changes sign when the numerator of equation (S26) changes sign.

The average parameter values in equation (S26), i.e, parameter values with overlines, were computed by averaging the estimated parameter values for our system. Averages were computed using three different sets of frequencies: (i) equal frequencies for all prey clones, meaning $q_i = 1/9$, (ii) the observed frequencies in the mesocosms in the "with parasites" treatment at week 9, and (iii) the observed frequencies in the mesocosms in the "no parasites" treatment at week 9. Because we do not know the equilibrium infection prevalence (Y^*) for our system, we computed the sign of equation (S26) for all values of Y^* between 0 and 1. Note that Z^* is determined by Y^* because $dY/dt = 0$ implies $Z^* = (\overline{m} + \overline{\mu} + \overline{a_I}P)Y^*/[\overline{pf}(1 - Y^*)]$. This allows us to determine if the sign of equation (S26) is fixed or changes as equilibrium infection prevalence increases from low to high values.

Figure S6 shows the relationships between the sign of $\partial N^*/\partial P$ and equilibrium infection prevalence (Y^*). Positive values (parts of the curves above the dashed purple line) imply that increased predator density will lead to increases in total prey density. Negative values (parts of the curves below the dashed purple line) imply that increased predator density will lead to decreases in total prey density. As expected, all of the curves have negative slope because increased predator density leads to decreases in total prey density when infection prevalence is sufficiently high. With the exception of the treatment with zero predator density (lightest blue color), all curves are above zero for sufficiently low infection prevalence (all curves above dashed purple curve on the left side of each panel).

We predict that the addition of the predator *C. punctipennis* will likely lead to decreased density of *D. dentifera*. Our reasoning is the following. Figure S6 shows that increased predator density leads to increased total prey density only if equilibrium infection prevalence is sufficiently low (curves above dashed purple line only on the left side of each panel). Here, "sufficiently low" means less than 5% because most of the curves become negative for $Y^* > 0.05$. For all replicates in the treatments with predator densities of 0/L, 0.1/L, and 0.5/L, infection prevalence was 10% or higher at the end of the experiment. This suggests that equilibrium infection prevalence is above 5% in those treatments. That in turn suggests that increasing predator density from 0/L to 0.5/L will decrease total prey density. Infection prevalence must eventually go to zero as predator density increases to larger values; this is a logical result and consistent with the observed zero infection prevalence in the treatments with predator densities of 1/L. Thus, we predict that the relationship between predator density and total prey density is u-shaped, where total prey density decreases as predator density increases from 0/L to 0.5/L and then total prey density increases for some range of predator densities between 0.5/L and 1/L. Because the magnitudes of the curves in Figure S6 are relatively small when they are above 0, we expect only small increases in prey density as predator density increases from 0.5/L to 1/L. In total this means we expect the relationship between predator density and total prey density to be u-shaped, with the highest prey density occurring when predator density is zero.

To explore if our results were sensitive to the assumption that predators had higher attack rates on infected prey than susceptible prey, we repeated the above analysis assuming $\omega = 1$. The results are shown in Figure S7. A key difference is that increased

predation decreases prey density at all levels of predation and for any level of infection prevalence in the prey (all curves in Figure S7 below the dashed purple line). Thus, we predict that if predators have equal attack rates on susceptible and infected prey, then we expect a negative relationship between total *Daphnia* density and predator density, with the highest prey density occurring when predator density is zero.

Effects of variation in resource availability: As noted in the main text, resource availability varied over time in our experiments. As explained below, our model suggests that variation in resource availability was unlikely to qualitatively affect our experimental results.

Equation (S26) defines how equilibrium prey density responds to changes in predator density. In our model, variation in resource availability is realized as variation in the prey per capita growth rate, $g(N)$. Variation in prey per capita growth rate would qualitatively alter our results only if equation (S26) were to change sign. Changes in the sign of equation (S26) require a change in the sign of either the numerator or denominator. The sign of the denominator of equation (S26) is unlikely to change with variation in prey growth rates because (i) for the high prey densities in our experiment, variation the prey growth rate will not alter the assumption that prey growth rate is a decreasing function of prey density, i.e, $g'(N) < 0$, and (ii) the other terms in the denominator of equation (S26) are negative. The sign of the numerator of equation (S26) could change as prey growth rates are varied. However, a change in sign would only occur if the variation in prey growth rates decreased prey densities to such low levels that infection prevalence dropped below 5% (see left side of each panel in Figure S6). Given that infection prevalence was greater than 10% at the end of the experiment, the variation in resources is unlikely to cause a large enough change in prey density that the infection prevalence drops by more than half. In total, our model suggests that the variation in prey per capita growth rates caused by variation in resource availability is unlikely to have affected the negative relationship between total prey density and parasite level.

Connections with results in Packer et al. (2003): Here we show that sufficiently low infection prevalence is also a necessary condition for increased prey density with increased predation in the density-dependent direct transmission model on page 789 of Packer et al. (2003).

After making the notation consistent with this paper, the total prey density for the Packer et al. (2003) model is

$$N^* = I^* + S^* = \frac{(b_S - a_S P)(\mu + a_I P)}{\beta(a_I P - b_I)} + \frac{\mu + a_I P}{\beta} \tag{S36}$$

where b_S and b_I are the exponential reproduction rates of susceptible and infected individuals, a_S and a_I are the predator attack rates on susceptible and infected individual, μ is the disease-induced mortality rate, and β is the transmission parameter. The densities are positive only if $a_I P - b_I > 0$. Biologically, this means that the exponential reproduction

rate for infected individuals (b_I) is less than the per capita mortality rate due to predation ($a_I P$).

Differentiating the equation with respect to predator density, P, and algebraic simplification yields

$$\frac{\partial N^*}{\partial P} = \frac{-a_S(\mu + a_I P)}{\beta(a_I P - b_I)} + \frac{a_I(b_S - a_S P)}{\beta(a_I P - b_I)} - \frac{a_I I^*}{(a_I P - b_I)} + \frac{a_I}{\beta} \tag{S37}$$

$$= \frac{-a_S}{a_I P - b_I} S^* + \frac{a_I}{\mu + a_I P} I^* - \frac{a_I}{a_I P - b_I} I^* + \frac{a_I}{\mu + a_I P} S^* \tag{S38}$$

$$= N^* \left(\frac{-a_S}{a_I P - b_I}(1 - Y^*) + \frac{a_I}{\mu + a_I P} - \frac{a_I}{a_I P - b_I} Y^* \right). \tag{S39}$$

The second term in the parentheses is positive and the first and third terms in the parentheses are negative. If infection prevalence is sufficiently high (Y^* closer to 1), then the third term will be larger than the second term and the whole equation will be negative. Thus, increased prey density with increased predation ($\partial N^*/\partial P > 0$) is only possible if infection prevalence (Y^*) is sufficiently low.

References

Allen, M. R., R. A. Thum, and C. E. Cáceres. 2010. Does local adaptation to resources explain genetic differentiation among daphnia populations? Molecular Ecology 19:3076–3087.

Auld, S. K., S. R. Hall, J. Housley Ochs, M. Sebastian, and M. A. Duffy. 2014. Predators and patterns of within-host growth can mediate both among-host competition and evolution of transmission potential of parasites. The American Naturalist 184:S77–S90.

Bender, E. A., T. J. Case, and M. E. Gilpin. 1984. Perturbation experiments in community ecology: theory and practice. Ecology 65:1–13.

Diekmann, O., J. Heesterbeek, and M. G. Roberts. 2010. The construction of next-generation matrices for compartmental epidemic models. Journal of the Royal Society Interface 7:873–885.

Fox, J. A. 2004. New microsatellite primers for daphnia galeata mendotae. Molecular Ecology Notes 4:544–546.

Hairston Jr, N. G., S. P. Ellner, M. A. Geber, T. Yoshida, and J. A. Fox. 2005. Rapid evolution and the convergence of ecological and evolutionary time. Ecology Letters 8:1114–1127.

Packer, C., R. D. Holt, P. J. Hudson, K. D. Lafferty, and A. P. Dobson. 2003. Keeping the herds healthy and alert: implications of predator control for infectious disease. Ecology Letters 6:797–802.

744 Penczykowski, R. M., M. S. Shocket, J. H. Ochs, B. C. Lemanski, H. Sundar, M. A. Duffy,
745 and S. R. Hall. 2022. Virulent disease epidemics can increase host density by depressing
746 foraging of hosts. The American Naturalist 199:75–90.

747 Rogers, D. 1972. Random search and insect population models. The Journal of Animal
748 Ecology pages 369–383.

749 Rosenbaum, B., and B. C. Rall. 2018. Fitting functional responses: Direct parameter esti-
750 mation by simulating differential equations. Methods in Ecology and Evolution 9:2076–
751 2090.

752 Royama, T. 1971. A comparative study of models for predation and parasitism. Population
753 Ecology 13:1–91.

754 Searle, C. L., M. H. Cortez, K. K. Hunsberger, D. C. Grippi, I. A. Oleksy, C. L. Shaw,
755 S. B. de la Serna, C. L. Lash, K. L. Dhir, and M. A. Duffy. 2016. Population density,
756 not host competence, drives patterns of disease in an invaded community. The American
757 Naturalist 188:554–566.

758 Strauss, A. T., D. J. Civitello, C. E. Cáceres, and S. R. Hall. 2015. Success, failure and
759 ambiguity of the dilution effect among competitors. Ecology Letters 18:916–926.

760 Strauss, A. T., J. L. Hite, M. S. Shocket, C. E. Cáceres, M. A. Duffy, and S. R. Hall. 2017.
761 Rapid evolution rescues hosts from competition and disease but—despite a dilution
762 effect—increases the density of infected hosts. Proceedings of the Royal Society B:
763 Biological Sciences 284:20171970.

764 Van den Driessche, P., and J. Watmough. 2008. Further notes on the basic reproduction
765 number. Mathematical Epidemiology pages 159–178.

766 Yodzis, P. 1988. The indeterminacy of ecological interactions. Ecology 69:508–515.

767 Section S5 Supplementary Figures

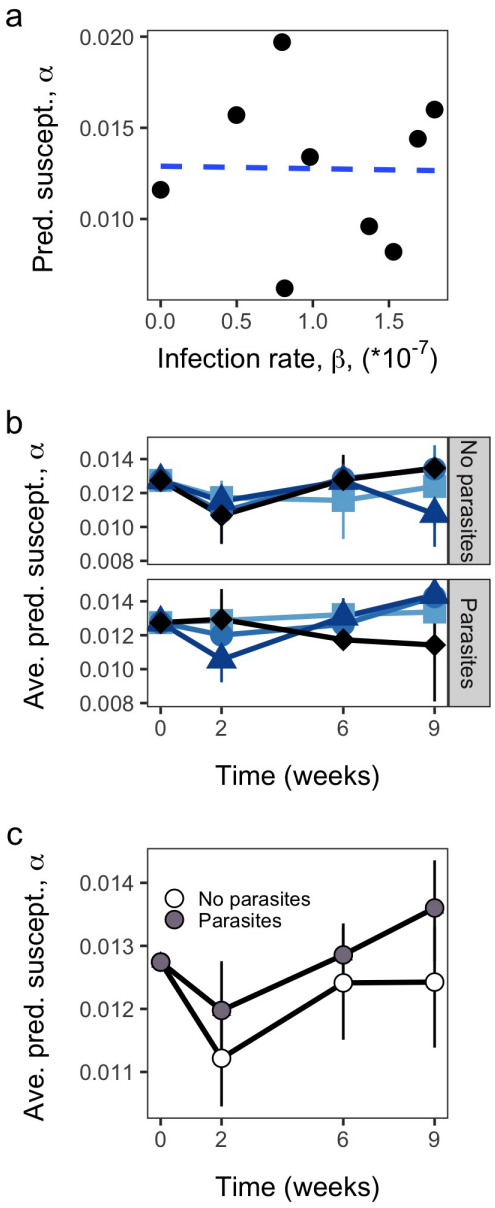

Figure S1: (a) The 9 genotypes used in this study spanned a wide range of susceptibility to parasitism and predation, but there was no trade-off between susceptibility to these two natural enemies. (b) Predation evolved over the course of the experiment but there was not a clear relationship between predation treatment and overall predation susceptibility. (c) Instead, populations that were exposed to parasites were not as resistant to the predation, even though there is no trade-off. Error bars in b&c are standard error. The colors and symbols in b are the same as for figures in the main text: light blue squares are 0 predators per L, medium light blue circles are 0.1 predators per L, medium dark blue triangles are 0.5 predators per L, and black diamonds are 1.0 predators per L.

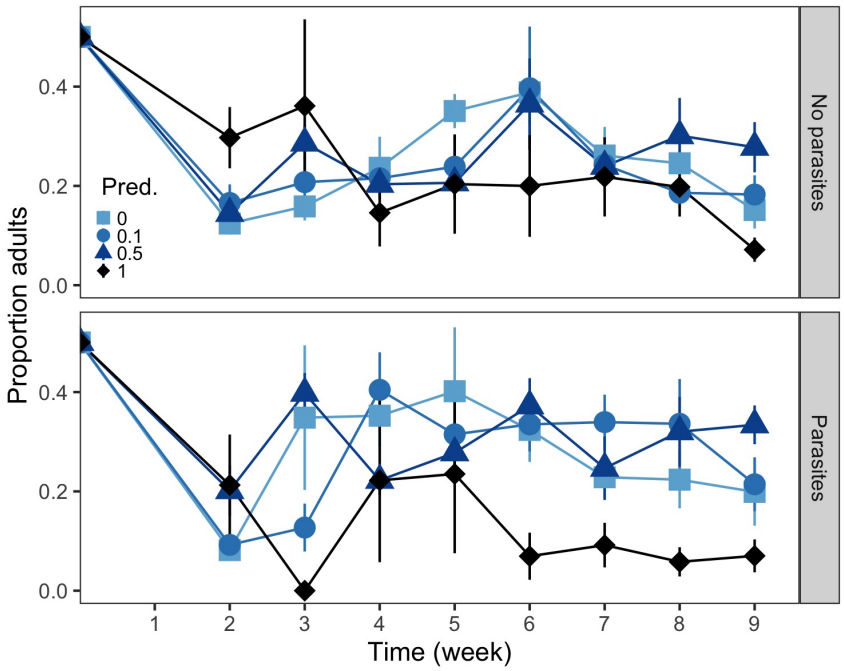

Figure S2: Stage structure of the populations varied over time. Populations that experienced high mortality (e.g., the highest predation treatment in the + parasite treatment) were dominated by juveniles.

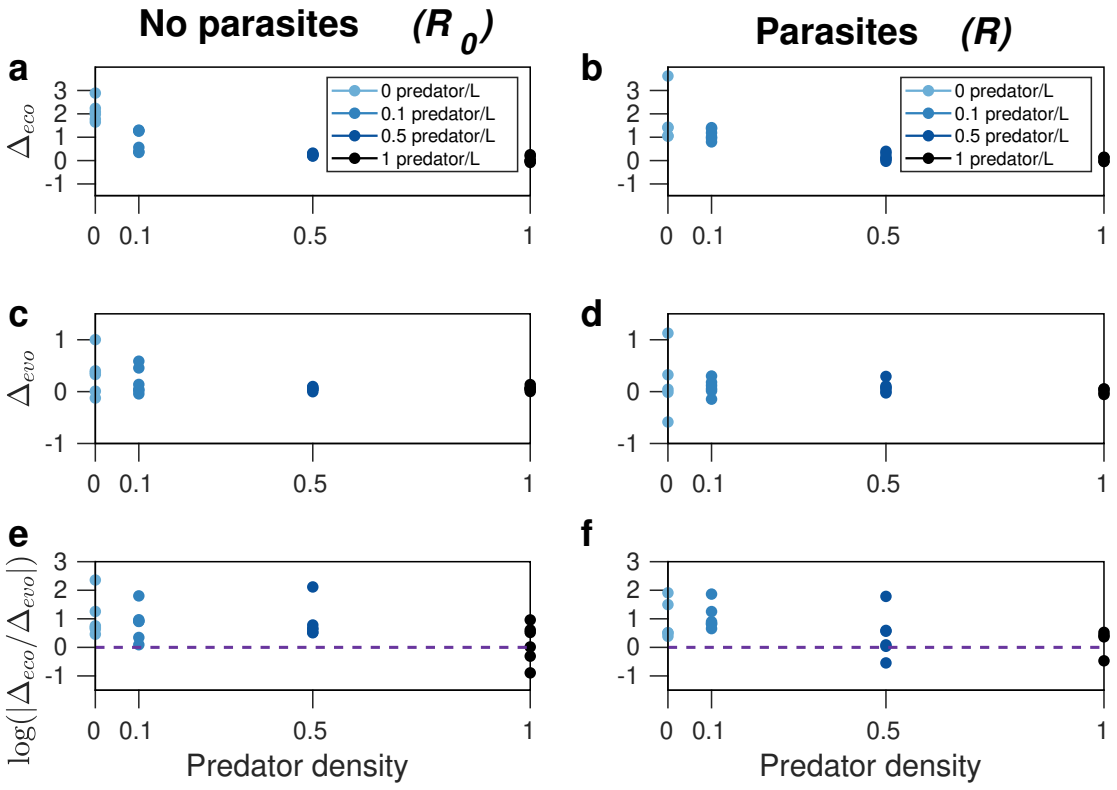

Figure S3: Ecological processes had larger effects than evolutionary processes on changes in parasite reproduction numbers between weeks 0 and 2. Top row: effect size of ecological changes (i.e., changes in prey density) on $\mathcal{R}_0$ (left) and $\mathcal{R}$ (right); middle row: effect size of evolutionary changes (i.e., changes in clone frequencies) on $\mathcal{R}_0$ (left) and $\mathcal{R}$ (right); bottom row: ratio of the effect sizes of ecological and evolutionary changes on a base 10 logarithmic scale. Effect sizes for $\mathcal{R}_0$ and $\mathcal{R}_0$ were computed using equations (S17) and (S18). Each point on the figure is one experimental replicate. Panels on the left show the treatments without the parasite and those on the right show those with the parasite. In the top row, positive and negative values indicate that changes in prey densities increased and decreased the reproduction number, respectively. In the middle row, positive and negative values indicate that changes in clone frequencies increased and decreased the reproduction number, respectively. In the bottom row, values above the dashed purple line indicate ecological changes had effects larger in magnitude than evolutionary changes and values below the line indicate evolutionary changes had effects larger in magnitude than evolutionary changes.

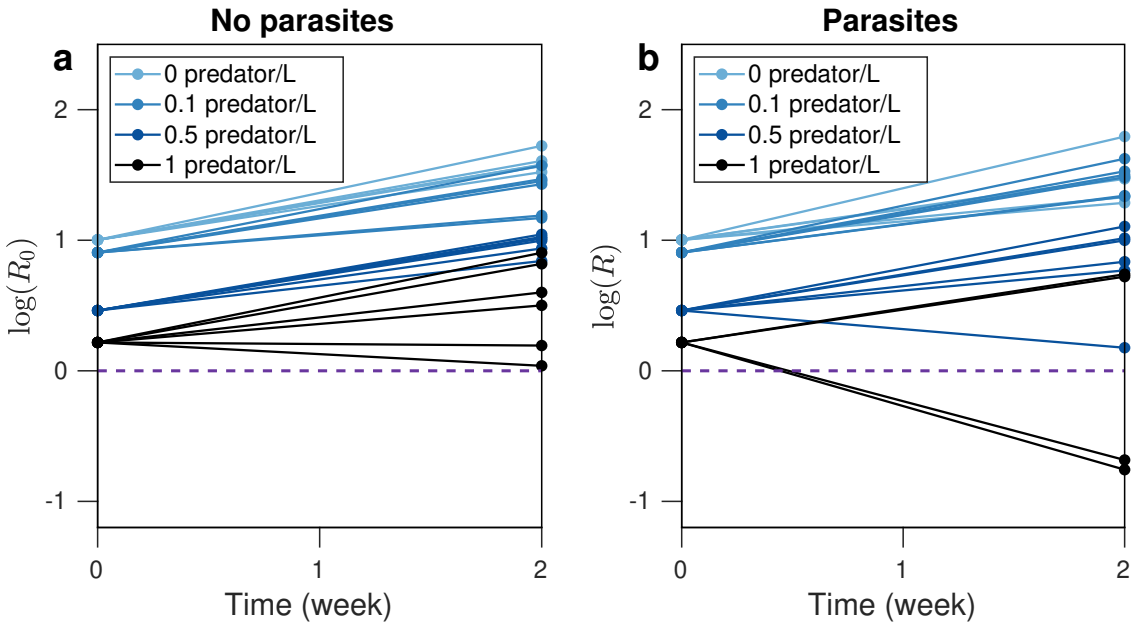

Figure S4: Parasite's basic reproduction number ($\mathcal{R}_0$) and reproduction number ($\mathcal{R}$) when calculated assuming the predator has equal attack rates on susceptible and infected prey ($\omega = 1$). Values of $\mathcal{R}_0$ and $\mathcal{R}$ were computed using equations (2.a) and (2.b), the estimated parameter values with $\omega = 1$, and the measures clone frequencies and prey densities at weeks 0 and 2. Each point represents an estimated value of $\mathcal{R}_0$ and $\mathcal{R}$ for a particular tank, with individual tanks connected by lines. The line coloring indicates the predation treatment (lightest blue = no predation, darkest blue = highest predation).The dashed line indicates $\mathcal{R}_0 = \mathcal{R} = 1$.

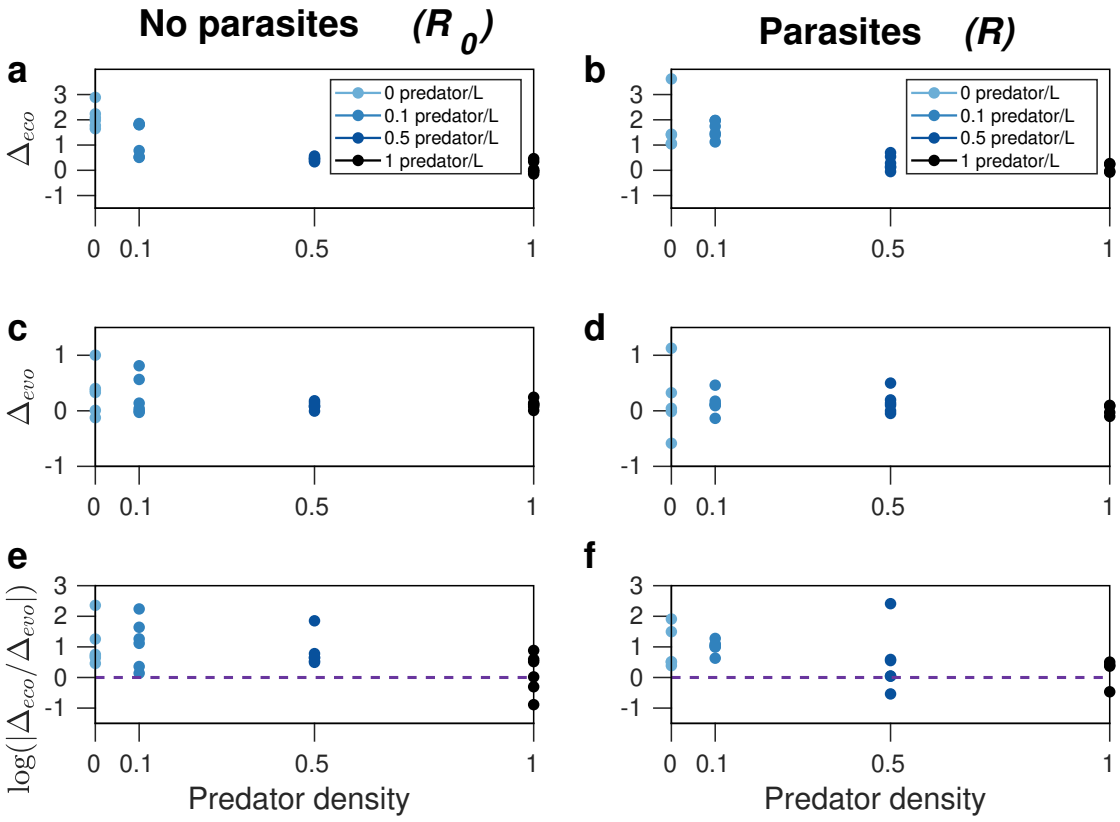

Figure S5: Predicted effects of ecological and evolutionary processes on changes in parasite reproduction numbers between weeks 0 and 2, computed assuming predators have equal attack rates on susceptible and infected prey ($\omega = 1$). Top row: effect size of ecological changes (i.e., changes in prey density) on $\mathcal{R}_0$ (left) and $\mathcal{R}$ (right); middle row: effect size of evolutionary changes (i.e., changes in clone frequencies) on $\mathcal{R}_0$ (left) and $\mathcal{R}$ (right); bottom row: ratio of the effect sizes of ecological and evolutionary changes on a base 10 logarithmic scale. Effect sizes for $\mathcal{R}_0$ and $\mathcal{R}_0$ were computed using equations (S17) and (S18), where the values of $\mathcal{R}_0$ and $\mathcal{R}_0$ were computed assuming $\omega = 1$; see Figure S4. Each point on the figure is one experimental replicate. Panels on the left show the treatments without the parasite and those on the right show those with the parasite. In the top row, positive and negative values indicate that changes in prey densities increased and decreased the reproduction number, respectively. In the middle row, positive and negative values indicate that changes in clone frequencies increased and decreased the reproduction number, respectively. In the bottom row, values above the dashed purple line indicate ecological changes had effects larger in magnitude than evolutionary changes and values below the line indicate evolutionary changes had effects larger in magnitude than evolutionary changes.

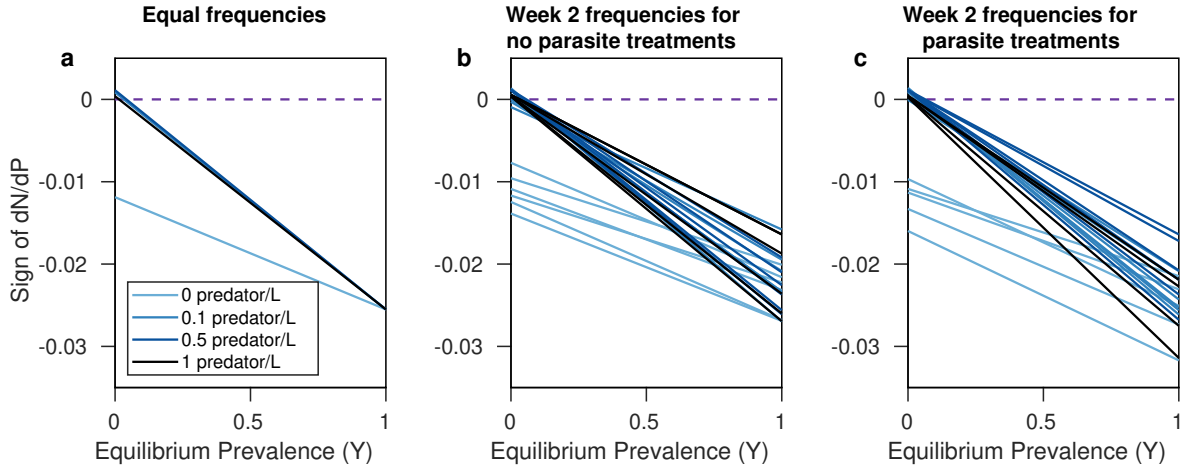

Figure S6: When predators have higher attack rates on infected individuals, for the *Daphnia* system, we predict that increased predation leads to increased total prey density only if infection prevalence is sufficiently low. In all panels, each curve shows the sign of $\partial N^*/\partial P$, which was computed using the estimated parameter values and the negative of the numerator of equation (S26); see text for additional details. Value above zero (purple line) indicate that prey density increases with increased predation and values below zero indicate that prey density decreases with increased predation. (a) Predictions when all clones are present at equal frequencies (which matches the conditions of the start of the experiment). (b) Predictions when the clone frequencies match the observed frequencies at week 2 in the treatments without parasites. (c) Predictions when the clone frequencies match the observed frequencies at week 2 in the treatments with parasites.

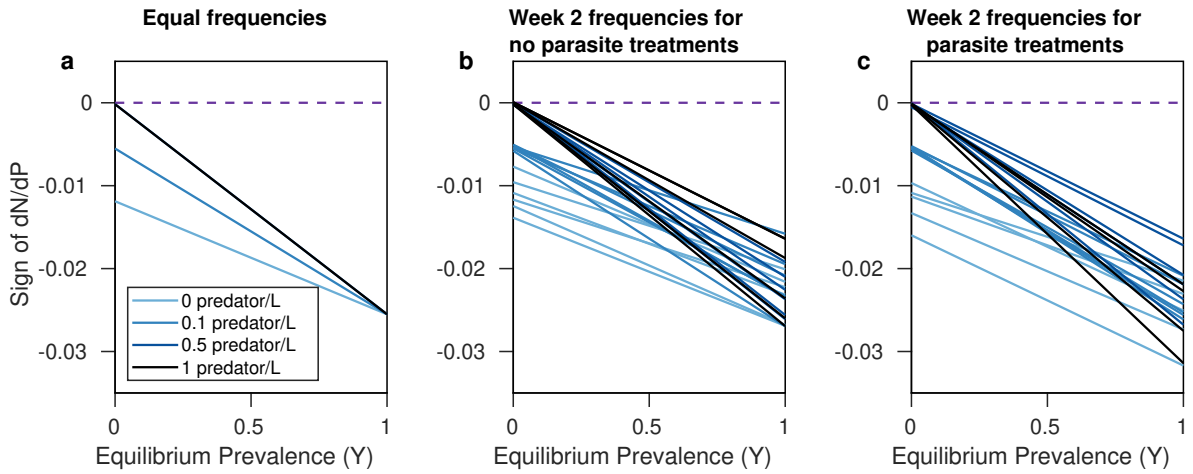

Figure S7: If the predator has equal attack rates on susceptible and infected prey, then we predict that increased predation leads to decreased total prey density in the *Daphnia* system; this outcome is predicted for all levels of infection prevalence. In all panels, each curve shows the sign of $\partial N^*/\partial P$, which was computed using $\omega = 1$, the estimated parameter values, and the negative of the numerator of equation (S26); see text for additional details. Value above zero (purple line) indicate that prey density increases with increased predation and values below zero indicate that prey density decreases with increased predation. (a) Predictions when all clones are present at equal frequencies (which matches the conditions of the start of the experiment). (b) Predictions when the clone frequencies match the observed frequencies at week 2 in the treatments without parasites. (c) Predictions when the clone frequencies match the observed frequencies at week 2 in the treatments with parasites.