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Supporting Online Material for

Ecological Context Influences Epidemic Size and Parasite-Driven Evolution

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Materials and Methods

Field sampling

We studied host evolution and ecologically driven variation in epidemic size in seven lake populations: Beaver Dam, Canvasback, Downing, Hale, Island, Midland, and Scott Lakes (Greene and Sullivan Counties, Indiana). Lakes were sampled, on average, every 8 days (range: 2-15 days). As in previous studies (*15, 16, 21*), we estimated infection prevalence visually on live hosts, using a dissecting microscope at 25-50x magnification. Infections are clearly visible through the transparent body of the host.

Infection assays

We assayed the infection risk of isofemale lines established from each of the seven populations. These assays were conducted following established protocols (*13, 14, 18*). Individuals were raised under standard conditions for several generations in the lab prior to use in the assays; this protocol minimizes environmental effects. We assayed 9-21 (mean: 15.4) isofemale lines per time period (pre-epidemic vs. post-epidemic) per lake. For logistical reasons, we used two temporal blocks to assay the isofemale lines from four lakes (Beaver Dam, Canvasback, Hale, and Midland).

We used *D. dentifera* that were 6-9 days old for the assays. Six individuals from a given isofemale line were placed in a beaker containing 100 ml of a 50:50 mixture of filtered lake water and Artificial *Daphnia* Medium (ADaM; *28*). There were eight replicate beakers per isofemale line; therefore, we exposed a total of 48 individuals per isofemale line to *Metschnikowia*. Animals were fed 10,000 cells ml^{-1} of the green algae *Ankistrodesmus falcatus* and exposed to 200 *Metschnikowia* spores ml^{-1} for approximately 24 hours, after which they were moved to beakers containing clean water. Animals were fed 20,000 cells ml^{-1} for the remainder of the experiment. Individuals were moved to clean beakers halfway through the experiment. Nine days after exposure,

individuals were examined for infections; by this time, infections are clearly visible, but hosts have not yet begun to die from virulent effects of the parasite (*14*).

Infection risk in this system depends on the rate at which hosts encounter spores (driven by host feeding rate) and the ability of spores to infect after they are consumed (per-spore susceptibility). The infection assays cannot distinguish between these two mechanisms.

Epidemiological modeling: methods

We used a trait-based epidemiological model parameterized for this system (see Table S1 for a complete description of parameters/variables). The model tracked changes through time of the density of host clones and free-living yeast spores and incorporated a parameterized tradeoff between transmission rate and fecundity among host clones (*13*). Since parasite-driven evolution of host populations is sensitive to tradeoff strength and shape (*6, 7*), it was essential to explore model predictions for this particular tradeoff. We varied productivity as a parameter governing density-dependence of birth rates (K), and we altered intensity of fish predation directly as a mortality rate (m), incorporating data on selectivity of predation on infected hosts (*15*).

We followed the dynamics of clones with differing susceptibility whose dynamics are given by:

$$\frac{dS_i}{dt} = b_i(S_i + fI_i)\left(1 - \frac{\sum_j S_j + I_j}{K}\right) - nS_i - mS_i - \beta_i S_i Z \qquad (1.a)$$

$$\frac{dI_i}{dt} = \beta_i S_i Z - nI_i - vI_i - \theta m I_i \qquad (1.b)$$

$$\frac{dZ}{dt} = \sigma(n + v + e\theta m)\sum_j I_j - dZ - \sum_j \beta_j S_j Z \qquad (1.c)$$

where S_i indicates susceptible hosts of clones i, I_i indicates infected hosts of clone i, and Z indicates free-living infectious stages of *Metschnikowia* (“spores”). Susceptible hosts (Eq. 1.a) increase as a result of density-dependent births, where K modulates that density

dependence and serves as our measure of environmental productivity. Maximal birth rate, b_i, is a function of transmission rate (susceptibility), β_i, based on a tradeoff quantified for our *D. dentifera-Metschnikowia* system (*13*; see Table S1). Fecundity of infected hosts is reduced compared to uninfected hosts, governed by the proportion constant f (*15, 29*). Susceptible hosts die as a result of non-selective mortality (n) and fish predation (m). They also become infected via free-living spores, at clone-specific rate β_i. Infected hosts (Eq. 1.b) increase following parasite transmission and are lost due to non-selective mortality (n) and fish predation (m). Fish prey selectively on infected hosts (*15*), as captured by θ (where $\theta > 1$ indicates selectivity). Infected hosts also die from direct virulent effects of the parasite (*14, 15, 30*). Free-living infective stages (spores; Z; Eq. 1.c) increase upon release from dead infected hosts. These dead, infected hosts contain σ spores; however, only a fraction of them (e) are released following predation. Spores are lost at rate d (e.g., due to sinking out of the water column) and due to uptake by susceptible *Daphnia*.

We simulated the dynamics of this host-parasite system to determine the infection prevalence and mean transmission rate (susceptibility) of the population. For the results presented in Figure 3, simulations were run with eleven clones with transmission rates evenly spaced between maximal ($8.5 * 10^{-6}$ L spores $^{-1}$ day^{-1}) and minimal ($0.5 * 10^{-6}$ L spores $^{-1}$ day^{-1}) values. Given this initially uniform distribution among clones, mean transmission rate started at $4.5 * 10^{-6}$ L spores $^{-1}$ day^{-1}. We explored four different predation rates (m = 0.02, 0.05, 0.10 and 0.15 day^{-1}) and a range of productivities, manipulated by changing K. The proportion infected was integrated over the simulation as a metric of epidemic size, and the average transmission rate (susceptibility) of the population at the end of the simulation (t = 200 days) was determined by weighting the transmission rate of each clone by its frequency. We focus on finite-time simulations to match our observations of the seasonal dynamics of susceptibility, but our qualitative results agree with an equilibrium based analysis of our model (not shown) and similar

ones (*7*). Our results were robust to changes in initial conditions (i.e., the initial distribution of the resistance trait among host clones and number of host clones in the population), length of the simulations, and the shape of the fecundity-resistance tradeoff that we assumed (linear, convex, or concave).

Epidemiological modeling: Results

This mathematical model supported our empirical findings. Our simulations confirmed that low predation intensity and high productivity fuel large epidemics that select for increased resistance (Fig. S1). Conversely, high predation and low productivity scenarios had small epidemics and experienced selection for increased susceptibility.

Table S1.

Model parameters and variables.

Parameter/ Variable	**Units**	**Definition**	**Value (*Reference*)**
b_i	day^{-1}	Maximum birth rate of susceptible clone *i*	$5712.11\beta_i + 0.241$ (*13*)
d	day^{-1}	Loss rate of spores	0.05
e	--	Fraction of spores released by predation	0.5 (*31*)
f	--	Proportional fecundity of infected clones	0.75 (*15*)
I_i	hosts L^{-1}	Density of infected clone *i*	--
K	host L^{-1}	Inverse of strength of density dependence of birth rate; a metric of productivity	1-10 (*14, 15*)
m	day^{-1}	Mortality rate from fish predation, susceptible hosts	0-0.24(*32*)
n	day^{-1}	Non-selective (background) mortality rate, susceptible hosts	0.05 (*15*)
S_i	hosts L^{-1}	Density of susceptible hosts	--
t	day	Time	--
v	day^{-1}	Enhanced death rate due to infection (virulence)	0.05 (*14, 15*)
Z	spores L^{-1}	Density of parasite spores	--
β_i	L spores^{-1} day^{-1}	Transmission rate (susceptibility) of host clone *i*	$0.5\text{-}8.5*10^{-6}$ (*14, 15, 18*)
θ	--	Selectivity of predation on infected hosts	9 (*15*)
σ	spores host^{-1}	Density of spores per infected host	15,000

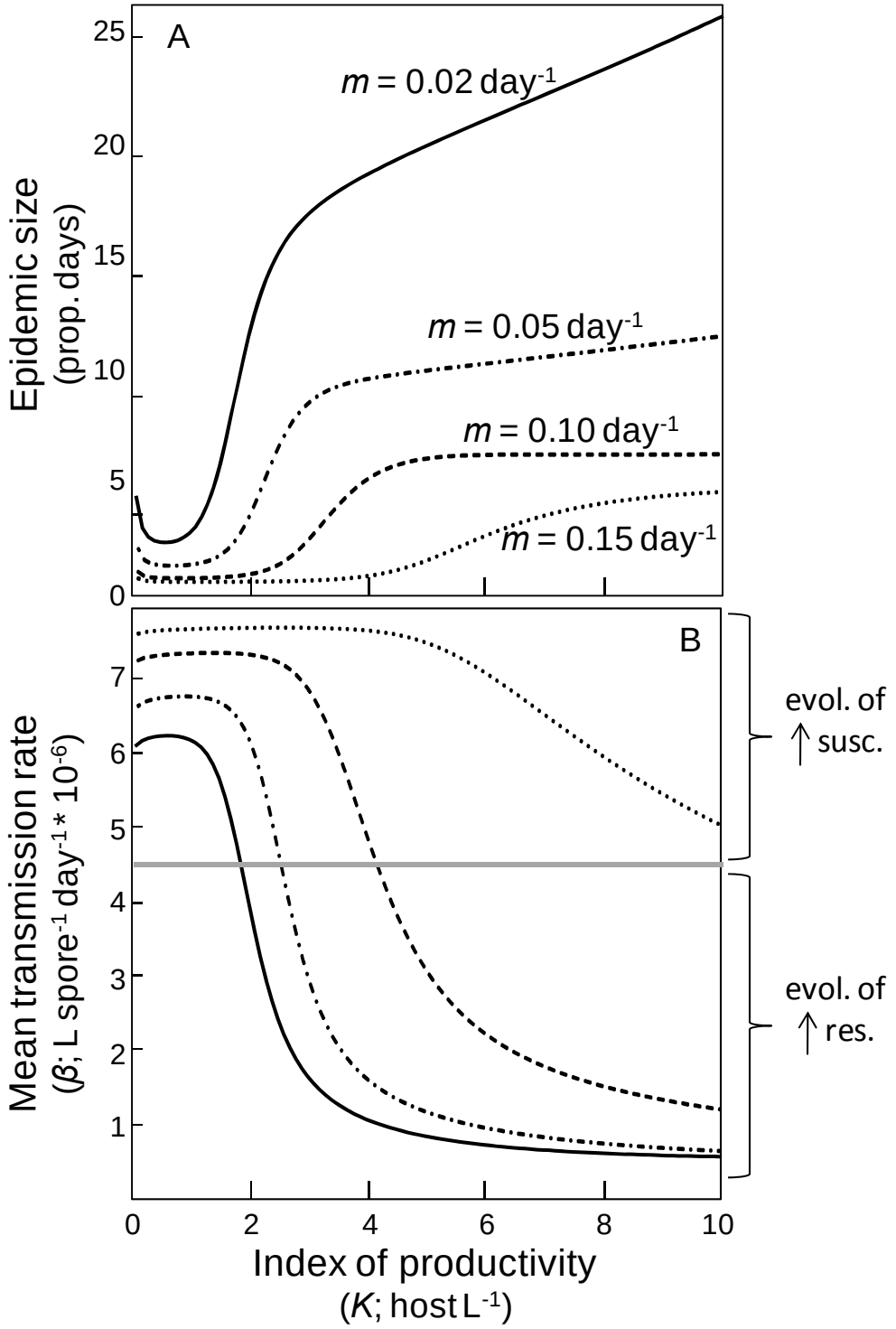
Table S2.

Ecological context, epidemic size and evolutionary responses of hosts. This table contains the data used to make Figure 2. The "Evolutionary response" column indicates the results of the analyses presented in Figure 1; "NS" means no significant evolutionary change, "IS" indicates evolution of increased susceptibility, and "IR" indicates evolution of increased resistance. "Pre-epidemic mean" and "post-epidemic mean" columns indicated the average proportion infected for genotypes assayed for the lake. Total phosphorus (TP), total nitrogen (TN), adult size, and integrated prevalence were determined from the field survey. Integrated prevalence is the area under the time series of infection prevalence for each lake. Integrated prevalence is strongly correlated with maximum infection prevalence ($r = 0.89$, $p = 0.008$).

Lake	Evolutionary response	Pre-epidemic mean	Post-epidemic mean	Change in susceptibility (pre – post)	TP (µg P L^{-1})	TN (µg N L^{-1})	Adult size (mm)	Integrated prevalence (prop. days)
Beaver Dam	NS	0.42	0.37	0.05	14.30	336.79	1.27	1.90
Canvasback	IS	0.238	0.302	-0.064	7.16	221.66	1.20	7.11
Downing	IS	0.469	0.537	-0.068	7.59	230.58	1.19	4.38
Hale	IS	0.344	0.484	-0.140	19.45	312.36	1.15	8.68
Island	IR	0.269	0.133	0.136	6.42	335.93	1.22	19.16
Midland	IR	0.26	0.218	0.042	26.92	435.29	1.30	20.13
Scott	IR	0.396	0.289	0.107	20.02	381.21	1.29	26.52

Figure S1. Simulation results showing effects of two ecological factors, productivity and predation intensity, on A) epidemic size (quantified as area under the infection prevalence curve) and B) susceptibility (quantified as mean transmission rate). Simulations were run for four different predation intensities (m = 0.02 (solid lines), 0.05 (dot-dashed lines), 0.10 (dashed lines) and 0.15 (dotted lines) day^{-1}). At high predation and low productivity, epidemics are relatively small and the population evolves higher susceptibility. As predation decreases and/or productivity increases, epidemics grow larger and the population evolves higher resistance. Initial mean susceptibility was 4.5 * 10^{-6} L $spore^{-1}$ day^{-1}, as denoted by the horizontal gray line. This line demarks the border between the evolution of increased susceptibility ("susc.", top) and the evolution of increased resistance ("res.", bottom).

Figure S1.


A
m = 0.02 day-1
m = 0.05 day-1
m = 0.10 day-1
m = 0.15 day-1
Epidemic size
(prop. days)
0
5
10
15
20
25
B
Mean transmission rate
(β; L spore-1 day-1 * 10-6)
1
2
3
4
5
6
7
evol. of
susc.
evol. of
res.
0
2
4
6
8
10
Index of productivity
(K; host L-1)

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