12

Competition and Diversity

12.1 Phage Worlds

12.1.1 *Consider a restriction- modification system in an E.coli where the restriction enzyme cut a certain sequence with efficiency 1/min, whereas the counteracting methylation happens 0.5/min. Consider a foreign phage that infect the bacteria, and which have 4 sites that can be cut/methylated. What is the probability δ that the phage DNA survive intact to be methylated at all 4 sites.*

Answer Rate to be cut is double that of being methylated, thus the probability for each site to be cut first is $C = 2/3$ and to be methylated first is $1 - C = 1/3$. To survive, all four sites have to be methylated first. This occurs with probability $\delta = (1 - C)^4 = (1/3)^4 = 1/81 = 0.012$.

12.1.2 *Repeat the above calculation if the invading phage express a protein that within 1 minute can prevent the restriction enzyme to work.*

Answer Let $r_{\mathrm{m}} = 0.5\,\mathrm{m}^{-1}$ be the methylation rate and $\mathrm{r}_c = 1\,\mathrm{m}^{-1}$ be cutting rate. The chance that a site survives unmodified until time t is the probability that none of the reactions occur during $[0, t]$:

$$P(t) = (1 - (r_{\mathrm{c}} + r_{\mathrm{m}})d\mathrm{t})^{t/d\mathrm{t}} = \exp(-(r_{\mathrm{m}} + r_{\mathrm{c}}) \cdot t)$$

The probability to be either cut or modified within time t is thus:

$$(1 - P(t)) = 1 - \exp(-(r_{\mathrm{m}} + r_{\mathrm{c}}) \cdot t)$$

Of this the fraction $r_c/(r_c + r_m)$ will be cut before being methylated. Thus the probability for a given site to be cut becomes:

$$C = (1 - P(t))\frac{r_c}{r_c + r_m} = (1 - e^{-(r_m+r_c)\cdot t}) \cdot \frac{r_c}{r_c + r_m} = (1 - e^{-1.5}) \cdot \frac{2}{3} = 0.52$$

for $t = 1$ min. The probability of all four sites surviving is the probability that none of the four site is cut within the 1 minute time interval:

$$(1 - C)^4 = 0.48^4 = 0.054$$

Thus the protective protein gives a factor of ~ 4.5 better infection probability.

12.1.3 *Repeat question 6.3.1 and 6.3.2 if the invading phage has 8 DNA sites that can be cut / methylated.*

Answer Without any protection, the survival becomes $(1/3)^8 = 0.00015$. With protective protein, the survival becomes $(1 - C)^8 = 0.003$ Thus the protective protein gives a factor of ~ 20 better infection probability.

12.1.4 *Consider the idealized equations for two bacteria species a and b with each their restriction modification system. The bacteria compete while exposed to one phage that acquire methylation from only the last bacteria it has successfully infected:*

$$\begin{aligned}
\frac{da}{dt} &= a \cdot (1 - a - b) - a \cdot (p_a + \omega_a \cdot p_b) \\
\frac{dp_a}{dt} &= a \cdot (p_a + p_b \cdot \omega_a) \cdot \beta - p_a \cdot (a + b + 1/\tau) \\
\frac{db}{dt} &= b \cdot (1 - a - b) - b \cdot (p_b + \omega_b \cdot p_a) \\
\frac{dp_b}{dt} &= b \cdot (p_b + p_a \cdot \omega_b) \cdot \beta - p_b \cdot (a + b + 1/\tau)
\end{aligned}$$

where p_a and p_b is the phage population of phages that have been methylated in a, respectively b last, β is the number of phage particles released per infection and τ is the life time of phages in the environment. Simulate the system for $\tau = 10$, $\beta = 100$ and relative deficiency of restriction modification systems $\omega_a = 1$ and $\omega_b = 0.1$ respectively. Discuss the result in terms of bacteria that use phages to compete with each other. Hint: Start with $a = 0.1$, $p_a = 1$ and fixed $b = p_b = 0$ and simulate until steady state using time-steps $dt = 0.001$. Then introduce $b = 0.000001$ and simulate again until steady state.

Answer First let us discuss the various terms, all expressed in time units of the bacterial growth rate:

$\mathrm{d}a/\mathrm{d}t = a$ is normal exponential growth in non-saturated conditions.
The factor $(1 - a - b)$ takes into account that there are limits on how many bacteria are sustainable, given external conditions like, for example, food supply or space.
The term $a \cdot p_a$ describes predation of a phage with modification compatible with restriction system of bacteria a, on bacterial population a.
The term $\omega_a \cdot a \cdot p_b$ describes predation of a phage without methylation modification that would make it possible to bypass restriction system of a, on the population of a. Thus $\omega_a < 1$ describes the efficiency of the restriction-modification system on new phages.
β is (proportional to) the phage burst size.
$\frac{1}{\tau}$ describes the death rate of phages due to external causes.

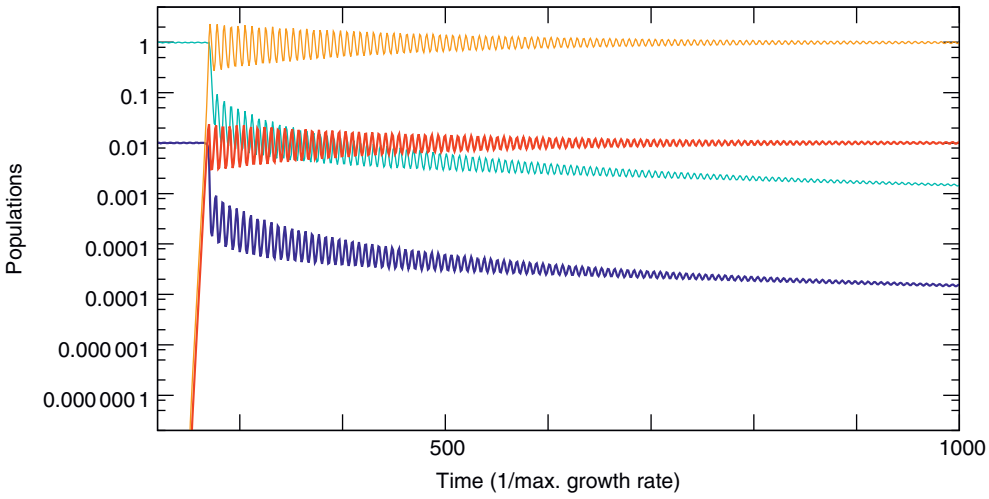

Figure 12.1 Simulation results where the "red" strain with $\omega = 0.1$, is introduced at time $t = 250$ to a "blue" strain with $\omega_b = 1$. The orange and cyan trajectories follow the corresponding phage populations. The simulation is done with burst size $\beta = 100$ and basal phage survival set by $\tau = 1$. The damped oscillations are not a robust feature of the model: oscillations disappear with low carrying capacity (here set to 1). With unlimited growth the system oscillates in analogy with the Lotka–Volterra equations. Notice that the blue strain persistently declines, a decline that is halted for any $\omega_b < 1$.

The resulting trajectories are seen in Fig. 12.1. One can see that even the relatively weak RMsystem with $\omega \sim 0.1$ allows the invading bacteria to displace the other bacteria, effectively using the phage as a weapon. Noticeably, if both bacteria have an RM system, they can co-exist.

12.2 Phage-bacteria ecology

12.2.1 *A more complete set of equations for a bacteria–phage ecosystem, where bacteria and phages are also removed by external sources, is:*

$$\frac{\mathrm{d}b}{\mathrm{d}t} = \Lambda_B \cdot \frac{b}{1 + b/b_{\max}} - \eta' \cdot b \cdot p - \delta'_b \cdot b$$
$$\frac{\mathrm{d}p}{\mathrm{d}t} = \beta \cdot \eta' \cdot b \cdot p - \delta' \cdot p$$

Use a saturation concentration of bacteria $b_{\max} \sim 10^8\,\mathrm{ml}^{-1}$*, and a maximum growth rate* $\Lambda_B \sim 0.0004\,\mathrm{s}^{-1}$*. Estimate* η' *from diffusion-limited localization of a phage with* $D \sim 4\,\mu\mathrm{m}^2\,\mathrm{s}^{-1}$ *to a bacterium with radius* $\sim 1\,\mu m$*. Reduce the number of parameters by rescaling* $t \to t \cdot \Lambda_B$ *etc.*

Answer $\lambda \sim 0.0004\,\mathrm{s}^{-1}$ corresponds to an exponential growth rate of a factor of 2 every 1700 s, i.e. the generation time $t_{\mathrm{gen}} = \ln(2)/\lambda = 1700\,\mathrm{s}$.
A phage locating any surface point on a bacterium with radius $\epsilon = 1\,\mu\mathrm{m}$ would use:

$$\tau = \frac{(1/B)}{4\pi D\epsilon} = \frac{B_{\max}}{B} \cdot \frac{10000\,\mu\mathrm{m}^3}{12 \cdot 1\,\mu\mathrm{m} \cdot 4\,\mu\mathrm{m}^2\,\mathrm{s}^{-1}} = \frac{B_{\max}}{B} \cdot 200\,\mathrm{s}$$

using a unit of density $B_{\max} = 10^{11}\,\mathrm{l}^{-1} = 10^{-4}\,\mu\,\mathrm{m}^3$, which corresponds to one unit per $10\,000\,\mu\,\mathrm{m}^3$. Thus one phage per $B_{\max}$ will give an infection rate of $0.0012\,\mathrm{s}^{-1}$, thus $\eta = 0.0012\,\mathrm{s}^{-1}/B_{\max} = \eta'/B_{\max}$.

Measuring both bacteria $b = B/B_{\max}$ and phage density $p = P/B_{\max}$ and using $\eta' = 0.0012\,\mathrm{s}^{-1}$ one obtains:

$$\frac{\mathrm{d}b}{\mathrm{d}t} = \lambda \cdot \frac{b}{1 + b} - \eta' \cdot b \cdot p - \delta_{\mathrm{B}} \cdot b$$
$$\frac{\mathrm{d}p}{\mathrm{d}t} = \beta \cdot \eta' \cdot b \cdot p - \delta \cdot p$$

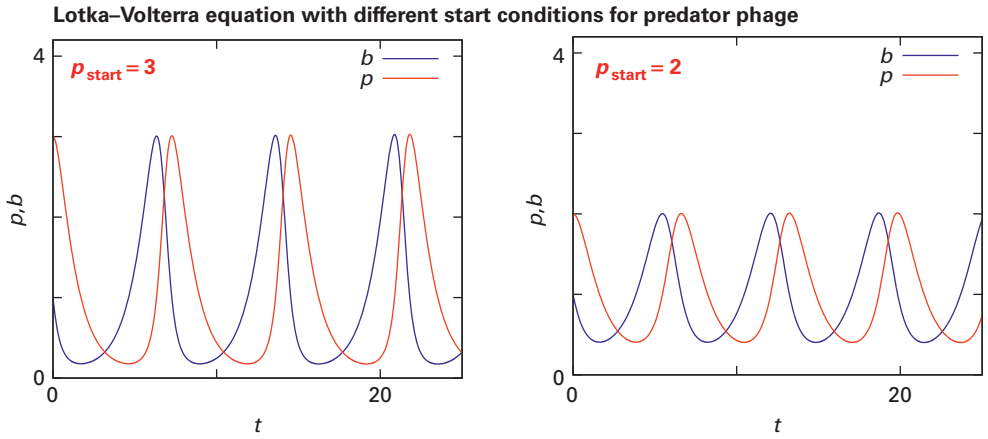

Figure 12.2 Lotka–Volterra equation with different starting conditions.

an equation where the two timescales $1/\eta' = 200$ s and $1/\lambda = 1700$ s are comparable. Finally rescaling the time with maximum growth rate $t' = \lambda t$:

$$\frac{\mathrm{d}b}{\mathrm{d}t} = \frac{b}{1+b} - \frac{\eta'}{\lambda} \cdot b \cdot p - \frac{\delta_\mathrm{B}}{\lambda} \cdot b$$
$$\frac{\mathrm{d}p}{\mathrm{d}t} = \beta \cdot \frac{\eta'}{\lambda} \cdot b \cdot p - \frac{\delta}{\lambda} \cdot p$$

an equation with three parameters, the burst size, the infection rate and the overall dilution rate (that in fact could be absorbed in the overall growth as is done in text). The rate constant $\frac{\eta'}{\lambda} \sim 8$. The dilution rate δ_B is in chemostats typically, $0.2\,\mathrm{h}^{-1} = 1/(18000\,\mathrm{s}) \sim 0.00005\,\mathrm{s}^{-1}$ in the chemostat by Levin (1977), corresponding to $\delta_\mathrm{B}/\lambda \sim 0.1$.

12.2.2 *Simulate the Lotka–Volterra equations:* $\mathrm{d}b/\mathrm{d}t = b - b \cdot p$ *and* $\mathrm{d}p/\mathrm{d}t = \beta \cdot b \cdot p - p$*, starting with different initial conditions,* $p = 3, b = 1$ *respectively* $p = 2, b = 1$.

Answer Use simple direct integration with time steps $\mathrm{d}t = 0.001$ and iterate from $t = 0$ to $t = 25$ to obtain the results shown in Fig. 12.2.

12.2.3 *Compare the standard Lotka–Volterra equation with two versions that include saturation terms on the prey population: (1)* $db/dt = b/(1+b) - b \cdot p$, $dp/dt = \beta \cdot b \cdot p - p$ *and (2)* $db/\mathrm{d}t = b \cdot (1-b) - b \cdot p$, $dp/\mathrm{d}t = \beta \cdot b \cdot p - p$. *In all cases start the simulation with* $p = 3, b = 1$.

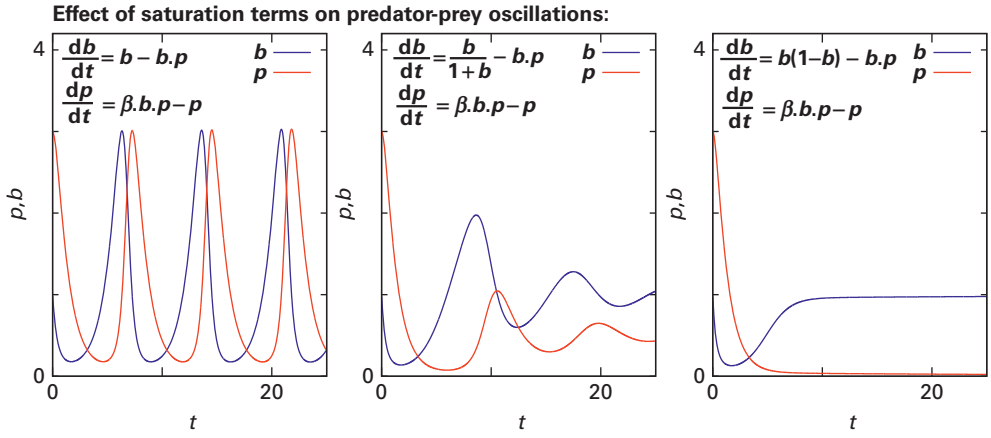

Figure 12.3 Effect of saturation on bacterial density.

Answer Use simple direct integration with time steps $\mathrm{d}t = 0.001$ and iterate from $t = 0$ to $t = 25$ to obtain the results shown in Fig. 12.3.

12.2.4 The time delay due to latency can, in principle, be implemented as a third state in Eqs. 12.17, which in rescaled units then translates to

$$\begin{aligned}
\frac{\mathrm{d}B}{\mathrm{d}t} &= \frac{B}{1+B} - \frac{1}{2}B - \eta \cdot B \cdot P \\
\frac{\mathrm{d}I}{\mathrm{d}t} &= \eta \cdot B \cdot P - I \\
\frac{\mathrm{d}P}{\mathrm{d}t} &= \beta \cdot I - \delta \cdot P - \eta \cdot I \cdot P
\end{aligned}$$

where we implicitly parameterize the latency time with an I state with lifetime $\tau = 1$. Simulate these equations with $\eta = 0.1$, $\beta = 100$ and death rate $\delta = 1$. Start the simulation with $B = 10^{-6}$ and $P = 0$ at time $t = 0$, and introduce phage $P = 10^{-6}$ at time $t = 30$.

Answer Simulation is shown in Fig. 12.4. In panel B we examine the equations:

$$\begin{aligned}
\frac{\mathrm{d}B}{\mathrm{d}t'} &= \frac{B}{1+B} - \frac{1}{2}B - \eta \cdot B \cdot P \\
\frac{\mathrm{d}I}{\mathrm{d}t} &= \eta \cdot B \cdot P - I/(1+B) \\
\frac{\mathrm{d}P}{\mathrm{d}t} &= \beta \cdot I/(1+B) - \delta \cdot P - \eta \cdot I \cdot P
\end{aligned}$$

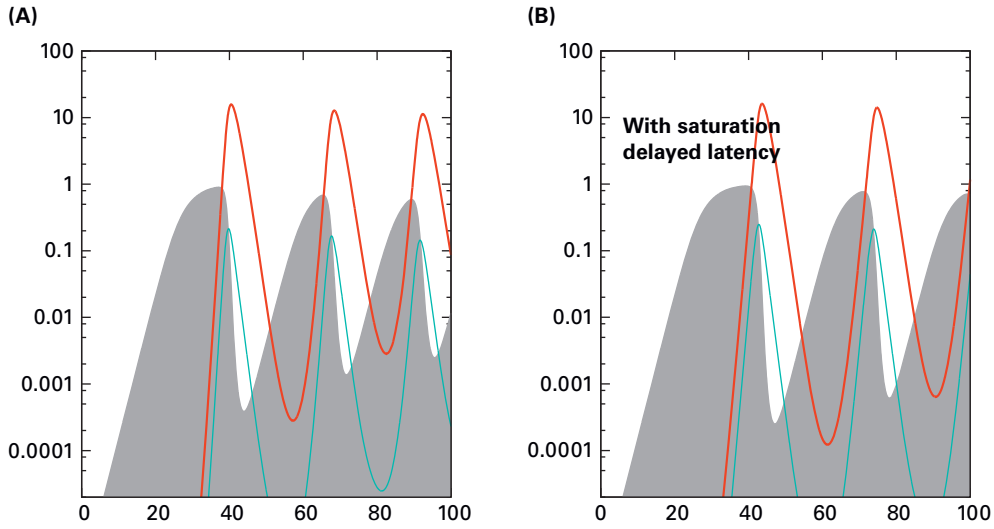

Figure 12.4 Effect of including delay associated with infected bacteria, which produce phages after (an exponentially distributed) delay τ.

thus implementing a latency that is further delayed as the bacterial population approaches 1. In both cases one obtains damped oscillations.

12.2.5 Bacterial species may limit their own growth more than others, because they will partly depend on different food resources. Such inhibition may be modeled through:

$$\frac{\mathrm{d}B}{\mathrm{d}t} = \frac{B}{0.5+B}\cdot(1-B-R) - 0.8\cdot B$$
$$\frac{\mathrm{d}R}{\mathrm{d}t} = \frac{0.2\cdot R}{0.5+R}\cdot(1-B-R) - 0.1\cdot R$$

Solve these equations numerically by starting both populations at size 0.000 001 and follow them until time $t = 200$. Notice that stable co-existence is indeed possible, and notice that the slow grower in the end dominates the population.

Answer Simulate the equations using timesteps $\mathrm{d}t = 0.001$. Results are shown in Fig. 12.5

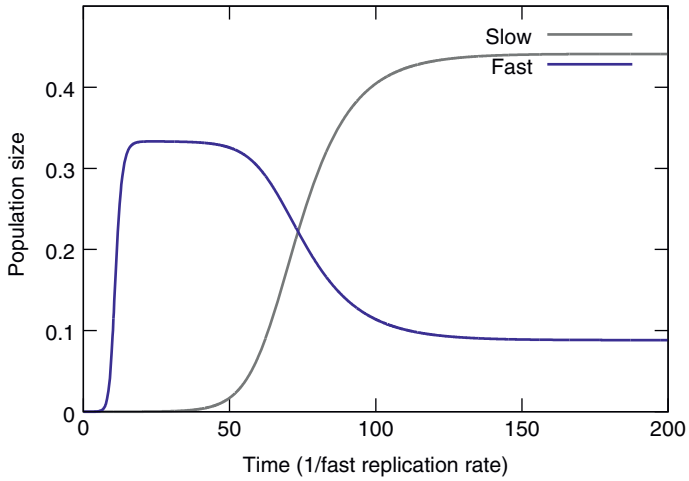

Figure 12.5 The dominance of the slow grower, provided: (1) that it is an even slower "dier" and (2) that the fast grower inhibits its own growth.

12.3 Phage games and optimal choice of lysogeny

12.3.1 *Compare the two phage, two host scenario explored in Eq. (12.21) for the case where there are three phages, each in their host. Plot the optimal strategy as a function of p_{lysis} and $p_{lysogeny}$ when these two vary between 0.02 and 0.98. Comment on the limitation of the used life/death approximation from the perspective of the prediction that the probability of choosing lysis decreases with increasing phage number.*

Answer The equation now reads

$$\begin{aligned} 1-P = & q^3 \cdot (1-p_{\text{lysis}}) \\ & + 3q^2(1-q) \cdot (1-p_{\text{lysis}}) \cdot (1-p_{\text{lysogeny}}) \\ & + 3q(1-q)^2 \cdot (1-p_{\text{lysis}}) \cdot (1-p_{\text{lysogeny}})^2 \\ & + (1-q)^3 \cdot (1-p_{\text{lysogeny}})^3 \end{aligned}$$

For each value of p_{lysis} and p_{lysogeny}, the value of $1-P$ is calculated for all $q = 0.01, 0.02, 0..3...0.99, 1.00$. The minimum value of $1-P$ is selected and its associate q value is plotted as a function of p_{lysis} and p_{lysogeny} in Fig. 12.6. One can see that lysis is, in general, disfavored more when there are more phages. This reflects the increased weight on selecting more and

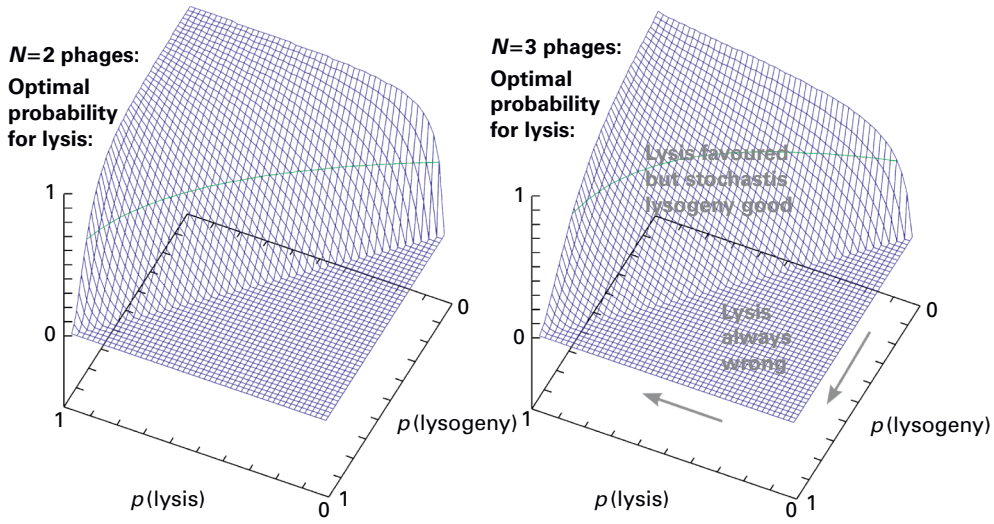

Figure 12.6 N = 2 and N = 3 phage calculation of optimal strategy for lysis frequency as a function of survival for each of the alternatives.

more independent future fates, as options for these increases. However, this repression of lytic growth would in reality be counteracted by exponential growth of a successful lytic strategy, which is not included in our present binary life/death approximation.

12.3.2 Estimate the phage decay rate, given that a burst of $\beta = 100$ phages on average allows just one phage to find a host when the density of hosts is $7000\,\mathrm{ml}^{-1}$ [696]. Assume a diffusion rate of phages of $4\,\mu\mathrm{m}^2\,\mathrm{s}^{-1}$ [676] and assume that a phage needs to come within $1\,\mu\mathrm{m}$ of the center of a bacterium to infect it.

Answer The capturing rate for phages with bacteria is given by:

$$\eta = 4\pi D\epsilon\rho = 3 \cdot 10^{-7}\,\mathrm{s}^{-1}$$

when using $D = 4\mu m^2\,\mathrm{s}^{-1}$, $\epsilon = 1\,\mu\mathrm{m}$ and $\rho = 7000\,\mathrm{ml}^{-1} = 7 \cdot 10^6\,\mathrm{l}^{-1} = 7 \cdot 10{-}9\,\mu\mathrm{m}^2$. Thus one phage is on average captured in about $1/\eta = 3\,000\,000\,\mathrm{s}$ or 800 hours. Assuming a decay rate δ for the phage particle, each phage particle will either decay or be captured, with respective probabilities:

$$P(\text{decay}) = \frac{\delta}{\delta + \eta} \quad \text{and} \quad P(\text{infect}) = \frac{\eta}{\delta + \eta}$$

If burst size is $\beta = 100$, the average number of infections becomes:

$$n(\text{infect}) = \beta \cdot \frac{\eta}{\delta + \eta}$$

which has to be equal to 1 to secure marginal propagation:

$$\beta \cdot \eta = \delta + \eta \Rightarrow \delta = (\beta - 1) \cdot \eta \sim 100 \cdot 3 \cdot 10^{-9} = 30\,000\,\text{s}$$

thus we estimate that each free T4 phage particle "survives" 8 hours on average. If we overestimated the infection rate η by the assumption that all phages in the vicinity of a bacterium are captured, a reduced η implies a correspondingly longer estimate for the lifetime of the free phage. As mentioned earlier, phages in oceans "live" about 1-2 days.

12.3.3 *Simulate the long-term (500 updates) development of a phage population that grows with rate $\Omega = 2$ during good times, but is exposed to events of size $\omega = 10^{-12}$ with frequency $p = 0.1$. Use the Kelly optimum value of x, $x = 0.01$ and $x = 0.9$, and compare outcomes. Repeat the simulation for smaller disasters, with $\omega = 10^{-2}$ and $\omega = 0.5$.*

Answer Starting with population 1, at each timestep assign it to be good with probability 0.9, and otherwise to be bad:

If good, then update the logarithm of the population by adding $\log(\Omega (1 - x) + x)$.
If bad, then update the logarithm of population by adding $\log(\omega(1 - x) + x)$.
Results are shown in Fig. 12.7.

12.3.4 *Consider a temperate phage that follows the Kelly optimal lysogeny frequency $x^* = p\Omega/(\Omega - 1) - (1 - p)\omega/(1 - \omega)$ with its associated fitness $\Lambda(x) = (1 - p) \cdot \log(\Omega \cdot (1 - x) + x) + p \cdot \log(\omega(1 - x) + x)$. Show that its fitness changes by mutating to become virulent is:*

$$\Delta\Lambda = \Lambda(0) - \Lambda(x^*) \sim p \cdot log(\omega/p) \tag{12.1}$$

in the limit where $\Omega >> 1 >> \omega$. Interpretation: the fitness loss at relatively frequent disasters $p > \omega$ comes from treating the probability of disasters as if it was as small as the severity of disasters.

Answer Fitness change is:

$$\begin{aligned}\Delta\Lambda &= \Lambda(x = 0) - \Lambda(x) \\ &= (1 - p) \cdot \log(\Omega) + p \cdot \log(\omega)\end{aligned}$$

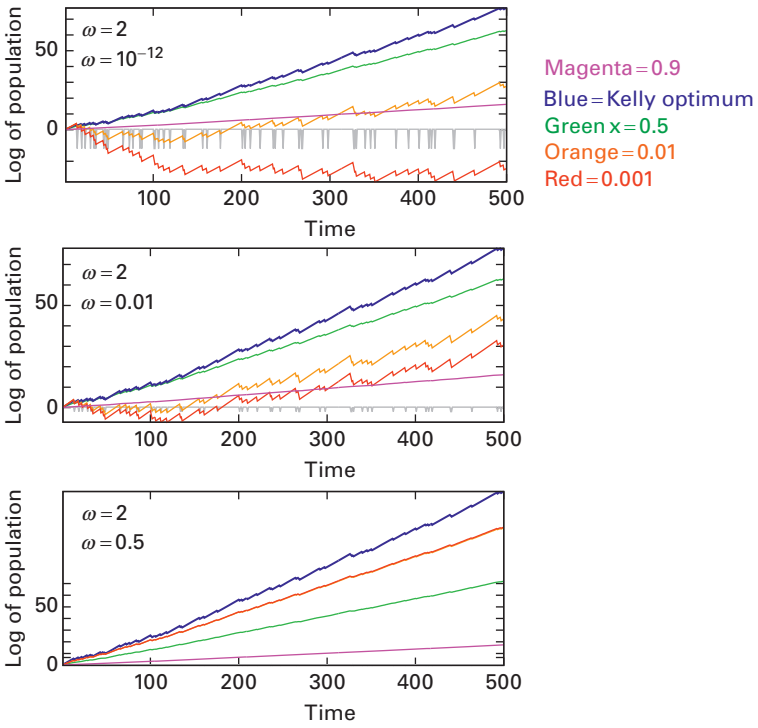

Figure 12.7 Simulation of phage growth when exposed to disasters of size ω with frequency $p = 0.1$.

$$
\begin{aligned}
&- ((1-p)\cdot\log(\Omega\cdot(1-x)+x) - p\cdot\log(\omega(1-x)+x)) \\
&= -((1-p)\cdot\log((1-x)+x/\Omega) - p\cdot\log((1-x)+x/\omega))
\end{aligned}
$$

into which we insert $x = p\Omega/(\Omega-1) - (1-p)\omega/(1-\omega)$ to obtain:

$$
\begin{aligned}
\Delta\Lambda &= -(1-p)\cdot\log\left(1 - \frac{p\Omega}{\Omega-1} + \frac{(1-p)\omega}{1-\omega} + \frac{p}{\Omega-1} - \frac{(1-p)\omega}{\Omega(1-\omega)}\right) \\
&\quad - p\cdot\log\left(1 - \frac{p\Omega}{\Omega-1} + \frac{(1-p)\omega}{1-\omega} + \frac{p\Omega}{\omega(\Omega-1)} - \frac{(1-p)}{(1-\omega)}\right) \\
&= -(1-p)\cdot\log\left((1-p) + (1-p)\frac{\omega}{1-\omega} - \frac{(1-p)\omega}{\Omega\cdot(1-\omega)}\right) \\
&\quad - p\cdot\log\left(p + \left(-\frac{p\Omega}{\Omega-1}\right) + \frac{p\Omega}{\omega(\Omega-1)}\right) \\
&= -(1-p)\cdot\log\left((1-p)\cdot\left(1 + \frac{\omega}{1-\omega}\left(1 - \frac{1}{\Omega}\right)\right)\right)
\end{aligned}
$$

$$
\begin{aligned}
&- p \cdot \log\left(p \cdot \left(1 + \frac{\Omega}{\Omega - 1}\left(\frac{1}{\omega} - 1\right)\right)\right) \\
&= -(1-p) \cdot \log\left((1-p) \cdot \frac{\frac{1}{\omega} - \frac{1}{\Omega}}{\frac{1}{\omega} - 1}\right) - p \cdot \log\left(p \cdot \frac{\frac{1}{\omega} - \frac{1}{\Omega}}{1 - \frac{1}{\Omega}}\right)
\end{aligned}
$$

which for $\omega << 1$ and $\Omega >> 1$ can be approximated by:

$$
\begin{aligned}
\Lambda &= -(1-p) \cdot \log\left((1-p) \cdot (1+\omega)\right) - p \cdot \log\left(\frac{p}{\omega}\left(1 + \frac{1}{\Omega}\right)\right) \\
&\sim +p \cdot \log(\omega/p)
\end{aligned}
$$

Thus Λ is negative (lose fitness) if $p > \omega$: the phages lose population if the frequency of disasters is larger than their magnitude. Notice that the magnitude of a disaster is quantified as the probability that a given lytic phage survives it, and it thus could indeed be compared to the probability that such a event occurs.