Stochastic within-host dynamics of antibiotic resistance: Resistant survival probability, size at the end of treatment and carriage time after treatment

Peter Czuppon[1,2,3,*], Troy Day[4], Florence Débarre[2], François Blanquart[3]

[1] Institute for Evolution and Biodiversity, University of Münster, 48149 Münster, Germany

[2] Institute of Ecology and Environmental Sciences of Paris, Sorbonne Université, UPEC, CNRS, IRD, INRA, 75252 Paris, France

[3] Center for Interdisciplinary Research in Biology, CNRS, Collège de France, PSL Research University, 75005 Paris, France

[4] Department of Mathematics and Statistics, Department of Biology, Queen's University, Kingston, ON K7L 3N6, Canada

[*] corresponding author: p.czuppon@uni-muenster.de

Keywords: antibiotic resistance, biostatic drug, biocidal drug, within-host dynamics, carriage time, stochastic dynamics

Abstract. The use of an antibiotic often leads to the emergence and spread of bacterial strains resistant to this antibiotic. Experimental and theoretical studies have investigated the drug dose that minimizes the risk of resistance evolution over the course of treatment of an individual, showing that the optimal dose will either be the highest or the lowest drug concentration possible to administer; however, no analytical results exist that help decide between these two extremes. To address this gap, we develop a stochastic mathematical model of bacterial within-host dynamics under antibiotic treatment. We explore various scenarios of density regulation (bacterial density affects cell birth or death rates), and antibiotic modes of action (biostatic or biocidal). We derive analytical results for the survival probability of the resistant subpopulation until the end of treatment, the size of the resistant subpopulation at the end of treatment, the carriage time of the resistant subpopulation until it is replaced by a sensitive one after treatment, and verify these results with stochastic simulations. We find that the scenario of density regulation and the drug mode of action are important determinants of the survival of a resistant subpopulation. Resistant cells survive best when bacterial competition reduces cell birth and under biocidal antibiotics. Compared to an analogous deterministic model, the population size reached by the resistant type is larger and carriage time is slightly reduced by stochastic loss of resistant cells. Moreover, we obtain an analytical prediction of the antibiotic concentration that maximizes the survival of resistant cells, which may help to decide which drug dosage (not) to administer. Our results are amenable to experimental tests and help link the within and between host scales in epidemiological models.

1 Introduction

Bacterial pathogens resistant to antibiotics are a major public health challenge responsible for more than a million deaths per year (Murray et al., 2022). Ecological and evolutionary principles have guided the design and evaluation of strategies to reduce the use of antibiotics while still eradicating pathogenic bacteria in patients (Read et al., 2011). Much theoretical and experimental research on antibiotic resistance has focused on the optimal drug dose (e.g. Day and Read, 2016; Scire et al., 2019; Hansen et al., 2020) and the optimal prescription regimen in hospitals (e.g. Bergstrom et al., 2004; Kouyos et al., 2013; Abel zur Wiesch et al., 2014; Tepekule et al., 2017) to limit the evolution and spread of resistance (reviewed in Opatowski et al. (2011); Blanquart (2019); Niewiadomska et al. (2019)).

The question of the optimal drug dose and duration to limit the evolution of resistance has received much attention. "Hitting early and hitting hard" (Ehrlich (1913); Fleming (1964)) may limit the emergence of drug resistance (e.g. Olofsson and Cars, 2007; Cantón and Morosini, 2011; Mouton et al., 2011; Ankomah and Levin, 2014). More recent studies challenge this view (Read et al., 2011; Huijben et al., 2013; Day and Read, 2016) based on the result that the probability of emergence of drug resistance is maximized at an intermediate concentration. A low antibiotic concentration prevents the emergence of resistance by allowing the maintenance of the sensitive strain, which impedes growth of the resistant strain through competition. On the other hand, a high antibiotic concentration also prevents the emergence of resistance by quickly eradicating the bacterial population, limiting the input of resistance mutations, and directly limiting the growth of the resistant subpopulation. Thus, the emergence of drug resistance is most likely at an intermediate concentration where the sensitive population is eradicated, which frees resistance from competition ("competitive release", Day et al. (2015)), allowing the resistant subpopulation to grow. Depending on the treatment window, which describes the feasible range of drug concentration and is defined by clearance of the infection-causing bacteria and considerations on drug toxicity, a low or a high drug dose may be best to limit the emergence of resistance. This line of argument applies to symptomatic infections caused by pathogenic bacteria, which are the direct target of antibiotic treatment. Yet, it also applies to commensals and opportunistic pathogens that are carried asymptomatically, which are not the target of the antibiotic treatment but under 'bystander selection' (Tedijanto et al., 2018).

The survival or extinction of a small resistant subpopulation emerging during treatment is governed by the random processes of cell division and death. How stochasticity impacts the emergence of drug resistance has been studied theoretically (Day and Read, 2016; Blanquart, 2019; Scire et al., 2019) and empirically (Alexander and MacLean, 2020). However, a complete analytical treatment of the stochastic within-host dynamics of resistance during and after treatment is missing. Such analytical treatment is important to characterize the drug concentration maximizing the probability of emergence of resistance, the size of the resistant subpopulation within the host and, in the case of commensal bacteria, how long a treated host carries and sheds resistance after treatment. These results can in turn provide insight on the optimal drug dose and inform between-host models describing the shedding and transmission of the resistant strain to other hosts. A fully stochastic treatment of the within-host evolution of drug resistance also requires that we specify how bacterial cells compete, and whether the drug impedes cell division or actively kills cells (biostatic or biocidal). Both are important determinates of the probability of emergence of resistance (the probability that a resistant cell appears and establishes a substantial subpopulation). However, little attention has previously been paid to these aspects of the life cycle of bacterial cells. Here, we analyze different models of within-host dynamics and treatment and compute the survival probability of a resistant subpopulation until the end of treatment, the size of the resistant subpopulation at the end of treatment, and the carriage time of the resistant subpopulation in a host after treatment.

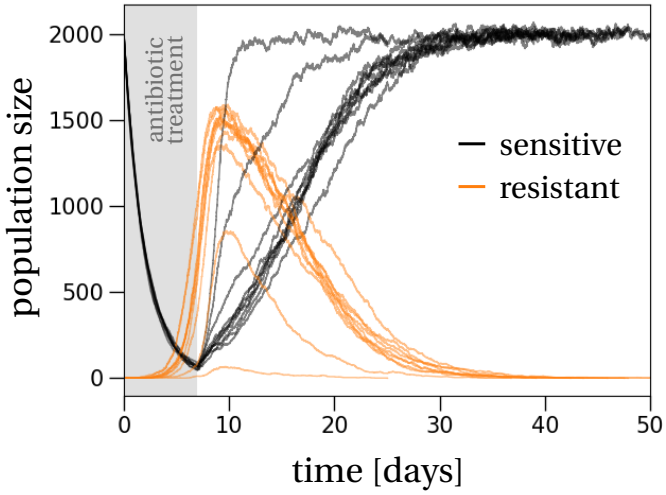

Figure 1: **Population dynamics of the resistant (orange) and sensitive (black) strains.** The sensitive subpopulation declines almost deterministically during treatment, which is administered for seven days (gray shaded region). At the same time the resistant subpopulation, if it survives, increases in size. There is strong variation between the resistance growth curves because of stochastic birth and death events. Non-surviving trajectories are not shown. After treatment, both subpopulations grow until the overall pathogen population reaches the carrying capacity, which happens approximately two days after the end of treatment when the resistant subpopulation reaches its maximum. The overall size of the resistant subpopulation at that point shows variation among the ten sample trajectories, again because of demographic stochasticity. This variation carries over to the time at which the resistant subpopulation is outcompeted by the sensitive subpopulation in the absence of antibiotic treatment.

2 Model

We study the population dynamics of a bacterial population within a host. We first describe how a drug initially clears the sensitive population, potentially allowing the establishment and rise of a resistant subpopulation. This first phase applies both to the bystander antibiotic exposure of bacteria that are carried asymptomatically and to the direct treatment of infections (symptomatic disease). A single resistant cell is introduced during treatment by mutation or transmission from another host. This resistant subpopulation may or may not establish in the host. If it does establish, it grows logistically until the end of treatment. Following this establishment phase, we model the dynamics leading to the extinction of the resistant subpopulation after treatment. When bacteria are carried asymptomatically, no further treatment is prescribed after the end of the antibiotic course. Sensitive strains can re-establish, either because they were not fully eradicated or because they are reintroduced by transmission. The sensitive strain eventually competitively excludes the resistant subpopulation because of resistance being associated with a cost. When bacteria caused an infection, it is reasonable to assume that a second course of antibiotic will be applied if the bacterial load is still high at the end of the first course. We do not model this scenario here.

Overall, the model is a stochastic description of the transient establishment, peak and extinction of a resistant subpopulation within the treated host (Fig. 1).

2.1 Bacterial population dynamics

We study two different *models* of density regulation of the bacterial population (Table 1). In the first model, population density affects the birth rate, which we refer to as 'birth competition'. For example, limited resource availability may impede cell division. In the second model, population density increases the death rate, which we refer to as 'death competition'. For example, bacterial cells may secrete bacteriocins that increase the death rate of cells in the population. Both models of density regulation yield the same deterministic population dynamics, as long as the overall population size remains below the carrying capacity. In the stochastic model formulation and analysis, however, the exact form of density regulation matters as we will show below.

Additionally, we distinguish between two *modes of action* of antibiotics: biostatic and biocidal. A biostatic drug, e.g. tetracycline or erythromycin, reduces the birth (division) rate of the cells, a biocidal drug, e.g. ciprofloxacin or streptomycin, increases the death rate. The deterministic dynamics are the same for the two modes of action if the antibiotic concentration is small enough. They differ, however, for larger concentrations because the birth rate cannot be smaller than zero, while the death rate can, in principle, increase without bounds as the antibiotic concentration increases (Fig. A in the Supplementary Information (SI)). Interestingly, the stochastic dynamics always depend on the mode of action because the variance of the underlying stochastic process is different for the two modes of action.

The two density regulation models and two modes of action define four *scenarios*. We start by describing the dynamics of the sensitive strain in the bacterial population. The overall birth and death rate of sensitive cells are denoted by $\lambda_S(x_S, x_R)$ and $\mu_S(x_S, x_R)$, where x_S and x_R denote the densities of the sensitive and resistant subpopulations, respectively. These rates, defining the stochastic process, vary depending on the scenario. For simplicity, we mostly present results from the main scenario whereby bacterial growth is modeled by birth competition and the host receives a biocidal antibiotic treatment. In this main scenario the rates are given by:

$$\lambda_S(x_S, x_R) = \max\big(\beta_S - \gamma_S(x_S + x_R), 0\big) \quad \text{and} \quad \mu_S(x_S, x_R) = \delta_S + \alpha_S(c), \tag{1}$$

where β_S denotes the birth rate of the sensitive strain, δ_S the intrinsic death rate of the sensitive strain, γ_S the competition parameter of the sensitive strain and $\alpha_S(c)$ the effect of the antibiotic treatment on the sensitive strain, where the drug is administered at concentration c (more details in the next section). The basic death rate comprises both cell death and the outflux of the host compartment colonized by bacteria. The dynamics of the resistant strain are defined analogously. We assume that resistance comes at a cost (Andersson and Hughes, 2010) that is mediated through a reduced birth rate $\beta_R < \beta_S$, an increased death rate $\delta_R > \delta_S$, or through less competitiveness $\gamma_R > \gamma_S$. Being (partly) resistant to the antibiotic reduces the rate by which the resistant type is affected by the antibiotic, i.e. $\alpha_R(c) < \alpha_S(c)$.

The description of the four scenarios at the individual cell level is summarized in Table 1. In SI, Section G we additionally study another model of density regulation, where the antibiotic interacts with the competition process described by the density-dependent terms, and in SI, Section I we study an alternative model with an explicit host immune response instead of competition between the two bacterial subpopulations.

2.2 Antibiotic response curve

We assume that the antibiotic affects the population dynamics of both the sensitive and the resistant strains. The *Minimum Inhibitory Concentration* (MIC) is the concentration at which the maximal growth rate, measured during the exponential phase, where density regulation can be neglected, is

		birth competition	death competition
biostatic	$\lambda_j(x_S, x_R)$	$\max\left(\beta_j - \gamma_j(x_S + x_R) - \alpha_j(c), 0\right)$	$\max\left(\beta_j - \alpha_j(c), 0\right)$
	$\mu_j(x_S, x_R)$	δ_j	$\delta_j + \gamma_j(x_S + x_R)$
biocidal	$\lambda_j(x_S, x_R)$	$\max\left(\beta_j - \gamma_j(x_S + x_R), 0\right)$	β_j
	$\mu_j(x_S, x_R)$	$\delta_j + \alpha_j(c)$	$\delta_j + \gamma_j(x_S + x_R) + \alpha_j(c)$

Table 1: **Birth and death rates in the four studied scenarios.** The overall birth and death rates, denoted by λ_j and μ_j, are composed of the birth, death and competition processes that occur at rates β_j, δ_j and γ_j, respectively. Additionally, the effect of antibiotics, denoted by $\alpha_j(c)$, affects either the birth or the death rate, depending on the type of drug administered. The variable c denotes the concentration of the antibiotic and the index j indicates the strain-specificity, $j = S$ or $j = R$ for antibiotic-sensitive or -resistant cells.

zero. For the sensitive strain, it is denoted mic_S and given by: $\beta_S - \delta_S - \alpha_S(\text{mic}_S) = 0$. The MIC of the resistant strain, mic_R, is defined analogously and is larger than the MIC of the sensitive strain. The resistant strain is unaffected by treatment, i.e. fully resistant, when mic_R equals infinity.

The antibiotic response curve, denoted $\alpha_j(c)$ (j either S or R), defines the effect of the antibiotic as a function of its concentration. In line with empirical studies (Regoes et al., 2004; Yu et al., 2016; Lee et al., 2020; Salas et al., 2022), we assume a sigmoid function relating the antibiotic-mediated death rate to the antibiotic concentration, specifically:

$$\alpha_j(c) = (\psi_{j,\max} - \psi_{j,\min}) \frac{\left(\frac{c}{\text{mic}_j}\right)^{\kappa}}{\left(\frac{c}{\text{mic}_j}\right)^{\kappa} - \frac{\psi_{j,\min}}{\psi_{j,\max}}}. \tag{2}$$

The function increases from zero when the antibiotic concentration is zero ($c = 0$), to $\psi_{j,\max}$ when $c = \text{mic}_j$, and to $\psi_{j,\max} - \psi_{j,\min}$ (note that $\psi_{j,\min} < 0$) when the antibiotic concentration is much larger than mic_j.

The parameter $\psi_{j,\max} = \beta_j - \delta_j$ is the maximal growth rate of strain j. This parameter is cancelled by antibiotic-induced death when the concentration is at the MIC. The parameter $\psi_{j,\min}$ is negative and corresponds to the maximal decline rate of strain j, i.e., the value $\beta_j - \delta_j - \alpha_j(\infty)$. The parameter κ is the steepness of the antibiotic response curve. Fig. 2 shows the resulting pathogen growth rate under antibiotic treatment, i.e. $\beta_j - \delta_j - \alpha_j(c)$, for our default parameter values. For simplicity, we neglect pharmacokinetics and model a constant antibiotic concentration during treatment.

cell

2.3 Stochastic simulations

In the stochastic simulations, we keep track of bacterial counts, denoted by X_j (j either S or R) and not densities, x_j, as introduced above. To translate between counts and densities, we define the parameter K that can be understood as the volume that the population inhabits. The relationship between densities and counts is then given by $x_j = X_j/K$.

Unless stated otherwise, the simulation results are obtained from 10^6 independent runs. All simulations are written in the C++ programming language and use the GNU Scientific Library. Code and data to reproduce the figures, which have been generated with python, have been deposited at `https://github.com/pczuppon/AMRWithinHost`.

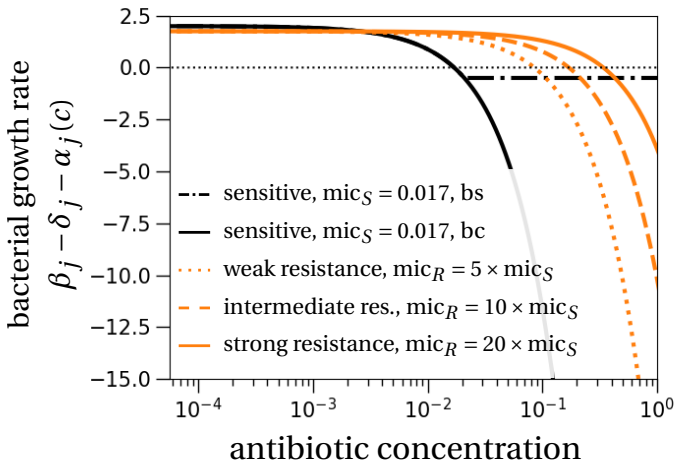

Figure 2: **Bacterial growth rate without density-dependent effects for different antibiotic concentrations.** The curves show the effect of a biostatic (bs) and biocidal (bc) drug. For a biostatic drug, we plot $\max\big(\beta_j - \alpha_j(c), 0\big) - \delta_j$ instead of the axis label $\beta_j - \delta_j - \alpha_j(c)$. To maintain clarity of the plot, we only show the biostatic drug effect on the sensitive strain, i.e., resistant strain growth curves all correspond to a biocidal drug effect. The effect of a biostatic drug on the different resistant strains would yield a curve with a minimal growth rate of $-\delta_R$. Parameters for the antibiotic response curve are motivated by estimates for ciprofloxacin from Regoes et al. (2004): $\beta_S = 2.5$ per day (d^{-1}), $\beta_R = 2.25$ (d^{-1}), $\delta_S = \delta_R = 0.5$ (d^{-1}), $\kappa = 1.1$, $\psi_{j,\text{min}} = -156 \times \log(10)$ (d^{-1}), $\psi_{j,\text{max}} = \beta_j - \delta_j$ (d^{-1}).

3 Results

Our aim is to quantify the dynamics of the resistant subpopulation during and after treatment of a host. We compute the probability of survival of a resistant subpopulation until the end of treatment, the size of the resistant subpopulation at the end of treatment and the carriage time of the resistant strain after the end of treatment. Together, these quantities provide a comprehensive picture of the stochastic population dynamics of a resistant subpopulation within a host (Fig. 1).

3.1 Survival probability of the resistant strain during treatment

We compute the survival probability of a resistant strain emerging during treatment. We assume that the resistant subpopulation establishes from a single resistant cell appearing during treatment. This single resistant cell could arise from a *de novo* mutation, or equivalently from transmission from another host during treatment, or from standing genetic variation. For clarity, in the main text we just present the results of resistance evolution from standing genetic variation, that is when one resistant cell is present at the beginning of treatment. Results about resistance dynamics from *de novo* emergence are discussed in SI, Section F. Qualitatively, the results of standing genetic variation and *de novo* emergence of resistance during treatment are very similar.

The dynamics of the resistant subpopulation emerging from a single resistant cell are well described by a stochastic birth-death process. For small numbers of cells, a deterministic description through an ordinary differential equation is not appropriate because stochastic fluctuations cannot be ignored. We use a branching process in a time-heterogeneous environment (Kendall, 1948; Haccou et al., 2005; Uecker and Hermisson, 2011) to approximate the probability of survival until the end of treatment.

This is the probability of having at least one resistant cell in the bacterial population at the end of treatment, which we refer to as *emergence* of resistance. Emergence, or survival, therefore does not imply establishment. We say that resistance *established* if the resistant type has risen to a density large enough that the probability of stochastic loss is negligible. A similar argument has been used in Day and Read (2016) to distinguish between emergence and establishment (note that they refer to emergence as occurrence and to establishment as emergence).

In the following, we qualitatively compare the predictions for the survival probability in the four scenarios. Theoretical predictions are derived in Section B in the SI.

In Fig. 3, we plot the survival probabilities of the resistant subpopulation for different MICs of the resistant strain and in the different models of density regulation and antibiotic modes of action as defined in Table 1. The survival probability of the resistant subpopulation is maximal for intermediate antibiotic concentrations c, in line with previous results (Day and Read, 2016; Scire et al., 2019). The relationship quantitatively depends on the model of density dependence. The survival probability is always higher for birth rather than death competition (compare y-axes between the different rows in Fig. 3). This is explained by a larger variance of the stochastic process when the density affects the death rate. Intuitively, a larger variance increases stochastic fluctuations, which reduces the survival probability for a population with large birth and death rates compared to a population with the same deterministic growth rate but small birth and death rates (Parsons et al., 2018).

By the same argument, one would expect that biocidal antibiotic treatment results in a smaller survival probability than biostatic treatment. Interestingly though, we observe that in all scenarios, there is barely a difference between the two antibiotic modes of action (compare the blue and orange curves and symbols in Fig. 3). In fact, survival probabilities tend to be larger for biocidal drugs. The largest difference is for highly resistant strains (right column on Fig. 3). This is explained by the much stronger competitive release caused by biocidal drugs: For biostatic drugs, the sensitive population will reach its minimal growth rate for concentrations slightly above its MIC. In contrast, for biocidal drugs, increasing the concentration continues to increase the death rate of sensitive strains and competitive release (Fig. 2, black solid line). This deterministic effect dominates the stochastic effect explained above.

3.2 Predicting the antibiotic concentration maximizing the resistant survival probability

As just outlined, the survival probability exhibits a maximum at intermediate antibiotic concentrations. In the context of treatment of symptomatic infections, the exact location of this antibiotic concentration would inform whether the "hit hard" strategy is optimal to limit resistance evolution.

Despite having explicit expressions for the survival probability (SI, Section B), we were not able to analytically find the concentration that maximizes the survival probability of the resistant subpopulation during treatment. However, in the case of biocidal treatment we can derive an implicit solution that depends on the demographic parameters, which can be evaluated numerically. Under birth competition and biocidal treatment, the concentration maximizing the survival probability of the resistant subpopulation is found by solving the following equation (details in SI, Section C):

$$\frac{\alpha_S'(c)}{\alpha_R'(c)} - 1 = \frac{\beta_R(\rho_R(c) - \rho_S(c))}{\rho_R(c)(\delta_R + \alpha_R(c))} + \frac{\beta_R(\rho_R(c) - \rho_S(c))^2}{x_S(0)\gamma\rho_R(c)(\delta_R + \alpha_R(c))}, \tag{3}$$

where $\alpha_k'(c)$ denotes the derivative of the antibiotic response curve with respect to the antibiotic concentration, and $\rho_k = \beta_k - \delta_k - \alpha_k(c)$ is the exponential growth rate under treatment of strain $k \in \{S, R\}$. A similar condition can also be derived in the scenario of a biocidal drug and death competition (SI, Section C).

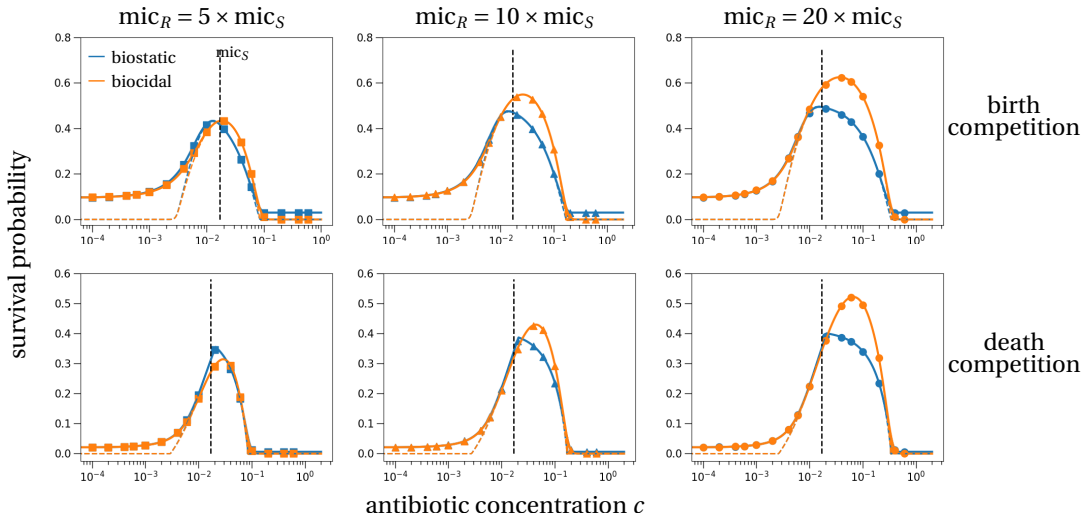

Figure 3: **Survival probability of the resistant subpopulation for varying drug concentrations, MIC values, drug types and model assumptions.** At the beginning of treatment the population consists of the sensitive strain at its carrying capacity and a single resistant cell. Then treatment, either with a biostatic (blue) or biocidal (orange) antibiotic, is applied for seven days (solid lines) or infinitely long (colored dashed lines). The survival probability of the resistant subpopulation obtained from stochastic simulations (symbols) agrees perfectly with our theoretical predictions (Section B in the SI). The different rows show different models (Table 1): top row = birth competition; bottom row = death competition. The columns show survival probabilities for different values of the resistant MIC as a multiple of the sensitive MIC: left = low, middle = intermediate, right = high resistant MIC. Parameter values are the same as in Fig. 2 and additionally $\gamma = 1$, $K = 1{,}000$ and $X_S(0) = (\beta_S - \delta_S)K/\gamma$.

The left-hand side of Eq. (3) reflects how much more the antibiotic affects the growth of the sensitive strain compared to the resistant strain. This difference can be positive or negative. For low concentrations, it will be positive as the sensitive strain is more affected by the antibiotic than the resistant strain. As soon as the antibiotic response for the resistant strain has a steeper negative slope than for the sensitive strain, the left-hand side will become negative. It is not possible to generally predict when this will be the case, but one can expect this to be at concentrations above the MIC of the sensitive strain (at least for sigmoid antibiotic response curves). The right-hand side is always positive as long as the selection coefficient (the growth rate difference $\rho_R(c) - \rho_S(c)$) is positive, which is always the case in a neighborhood of the maximizing concentration. Still, it is not straightforward to predict when these fractions are large and no general prediction of the maximizing concentration is possible.

For biostatic drugs, we are not able to derive a similar condition. For death competition, however, we find an upper bound for the concentration maximizing the survival probability. Denoting this concentration by $\tilde{c}$, it is given by the lowest concentration that ensures no further sensitive cell replication (details in SI, Section C), i.e.

$$\tilde{c} = \inf\{c : \beta_S - \alpha_S(c) = 0\}. \tag{4}$$

Importantly, this implies that this threshold concentration is independent of the level of resistance of the resistant strain. The maximum of the survival probability remains at the same concentration, as can be verified visually in Fig. 3 (lower row, blue curves). Note that if the intrinsic death rate of sensitive cells, δ_S, is zero, we find $\tilde{c} = \text{mic}_S$, which implies that the maximum of the resistant survival

probability is at a concentration equal to or less than the MIC of the sensitive type (note that in Fig. 3 we have $\delta_S > 0$). In the scenario of treatment with a biostatic drug and birth competition, no analytical prediction on the survival-maximizing concentration seems possible.

3.3 Size of resistant subpopulation at end of treatment

We now study the *size* of the resistant subpopulation when it survives treatment. The probability of survival is useful to know if any resistant subpopulation is able to establish in the host during treatment. Yet, the amount of shedding of resistant cells will depend not only on the possible emergence but also on the absolute size reached by the resistant subpopulation. Note that the resistant subpopulation may still continue to grow after treatment has ended because the total carrying capacity has not been reached at that time (Fig. 1).

The resistant subpopulation size at the end of treatment is typically larger in a stochastic model than in the deterministic counterpart (compare solid and dotted lines in Fig. 4). Precisely, because we condition on survival of the resistant subpopulation, the stochastic trajectory will increase faster initially than the deterministic trajectory. This is true until a certain threshold of cells is reached. This threshold is defined by the survival probability being equal to one for a resistant subpopulation of that size. From that threshold level on, the dynamics are well described by the deterministic system of ordinary differential equations. The detailed mathematical analysis is stated in Section D in the SI. The difference between the stochastic and deterministic predictions is largest for small differences between the resistant and sensitive strain because this is where the survival probability of the resistant strain is smallest, which speeds up the initial stochastic resistance establishment dynamics the most (compared to deterministic dynamics).

In Fig. 4, we compare the stochastic prediction of the size of the resistant subpopulation at the end of treatment (solid lines) and the deterministic prediction (dotted lines) with simulation results (symbols). The two modes of action of antibiotics affect the resistant subpopulation size at the end of treatment differently. We consistently find that the size of the resistant subpopulation at the end of treatment is larger for biocidal drugs than for biostatic ones. This is again explained by the stronger competitive release under biocidal drugs, which kill sensitive cells. The sensitive subpopulation therefore declines faster (Fig. A.1 in the SI). Consequently, the resistant subpopulation grows faster than compared to the biostatic treatment. This difference in resistant subpopulation sizes for the two drug modes of action is most apparent for a model with birth competition (upper row in Fig. 4). For death competition, the deterministic dynamics of the sensitive cells agree for both drug modes of action up to values close to the sensitive MIC (Fig. A.1 in the SI). For larger concentrations, biocidal treatment results in larger resistant growth, which agrees with the pattern from the other model (birth competition).

The largest resistant subpopulation is reached for an antibiotic concentration close to the MIC of the sensitive strain in all studied parameter sets and scenarios (compare dashed vertical line with the maximum of the curves in Fig. 4). For the survival probability the concentration maximizing resistant survival was in some cases clearly shifted away from the sensitive MIC. For the size of the resistant subpopulation, this shift away from the sensitive MIC is much smaller. Although we cannot rule out that this is a result of our chosen parameterization, we observe the same shift in all parameter sets (Section H in the SI).

3.4 Carriage time of resistant subpopulation within a host after treatment

So far, we have studied the within-host dynamics of a bacterial population, commensal or pathogenic, during antibiotic treatment. Following treatment, the resistant subpopulation will, if it established,

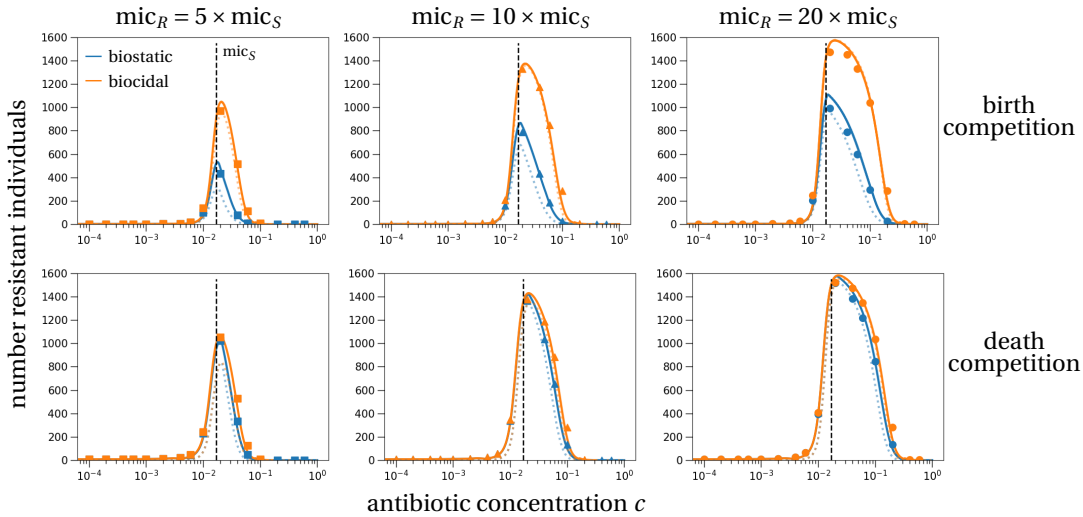

Figure 4: **Size of the resistant subpopulation at the end of treatment** if the resistant subpopulation survives. At the onset of treatment there is exactly one resistant cell in the population and treatment lasts for seven days. The top row shows the results for the model of birth competition, the lower row corresponds to death competition. Dotted lines show the deterministic prediction of the resistant subpopulation size, solid lines correspond to the stochastic prediction that incorporates a 'correction' due to conditioning on survival. Symbols are the mean resistant subpopulation sizes of the stochastic simulations that were conditioned on survival of the resistant subpopulation. Blue and orange colors correspond to biostatic and biocidal treatment, respectively. Parameters are as in Fig. 3.

still be present in the host and can therefore be transmitted to other hosts. Here, we study the carriage time of a resistant commensal strain in a host after treatment has ended. The analysis of this last phase is less relevant for infections by pathogenic bacteria because in this case treatment would be continued, possibly with a different antibiotic, to reduce the pathogen load and cure the patient.

The carriage time of a commensal resistant subpopulation can be interpreted as the time it takes a sensitive strain to re-establish within a host after antibiotic treatment has ended. At the end of treatment, the antibiotic concentration is set to $c = 0$ and we have $\alpha_S(0) = \alpha_R(0) = 0$. In the absence of antibiotics the resistant subpopulation will likely be replaced by the sensitive strain because the resistant strain has a fitness disadvantage compared to the sensitive strain. The size of the resistant and sensitive subpopulations at the end of treatment (previous section) are the initial conditions for this post-treatment phase. If the sensitive subpopulation has been eradicated during treatment, we assume that it starts with a single cell that is introduced from the host environment immediately after treatment has ended. In reality, the time until re-introduction of sensitive cells is a stochastic process governed by the influx rate from the environment.

We estimate the extinction time of the resistant subpopulation, conditioned on its extinction. To characterize this phase, we apply a timescale separation of the fast ecological and slow evolutionary dynamics, which reduces the problem to a single dimension (Constable and McKane, 2015; Parsons and Rogers, 2017). We follow the frequency dynamics of the resistant type in the population, while the overall population size is assumed to remain (approximately) constant. A timescale separation of the population size and the frequency dynamics applies if the difference between the resistant and sensitive strains is small or even negligible compared to the ecological rates (Czuppon and Traulsen, 2018; Gerlee, 2022). In our standard parameter set, the evolutionary rate is proportional

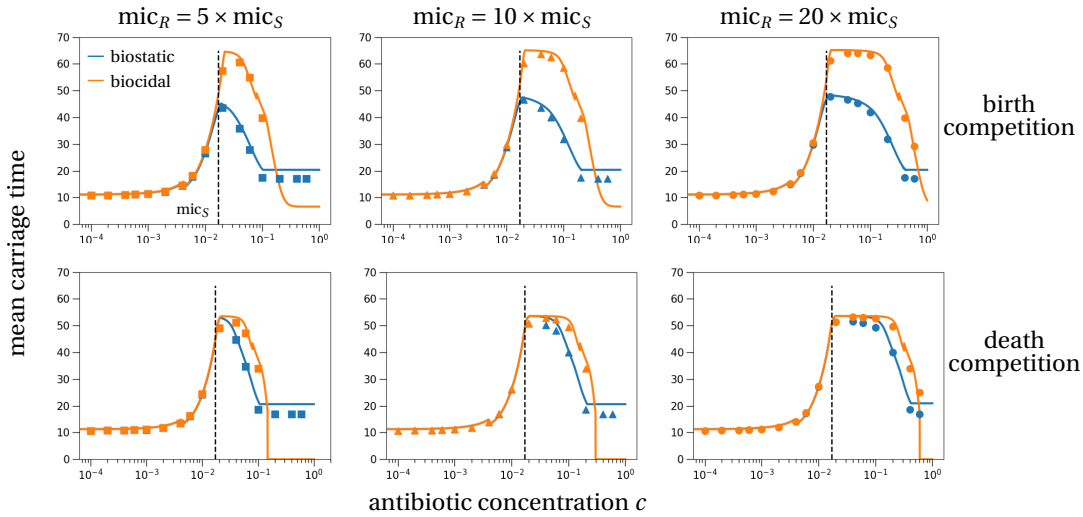

Figure 5: **Mean carriage time of the resistant subpopulation in a host** (including the treatment time $\tau = 7$). The carriage time is set to zero if the resistant subpopulation did not survive antibiotic treatment. Discontinuities are a result of discontinuities in the estimate of the resistant subpopulation size at the end of treatment (discussion in Section D in the SI). The theoretical predictions (lines) are derived in Section E. Symbols show the average carriage time of stochastic simulations. The color coding and figure structure is the same as in the previous figures. Parameters are the same as in Fig. 3.

to the cost of resistance $\beta_S - \beta_R$. This cost is about 10% of the ecological rate $\beta_k - \delta_k$. We first outline the deterministic population dynamics and then comment on the differences with a fully stochastic version. The deterministic population dynamics unfold as follows (Fig. 1): first, on the fast timescale, the overall population size, which is the sum of sensitive and resistant cells, will increase and approach the carrying capacity very quickly. Once at carrying capacity, the population will slowly move towards the extinction boundary of the resistant strain due to its fitness disadvantage. In other words, the population size remains approximately constant while the frequency of the sensitive strain increases. With demographic fluctuations, which are due to stochasticity in the birth and death processes, extinction of the resistant strain is not certain as the sensitive strain can go extinct on its way to re-establishment. Still, we assume that eventually the sensitive strain will replace the resistant subpopulation. Biologically, this is motivated by a constant influx of sensitive cells, so that one of these repeated establishment attempts will eventually be successful. We therefore condition the stochastic process on extinction of the resistant subpopulation. This conditioning results in a lower boundary of the carriage time of the resistant strain, i.e., the carriage will in reality be at least as long as our estimate. Applying results from one-dimensional stochastic diffusion theory (e.g. Ewens, 2004; Otto and Day, 2007; Czuppon and Traulsen, 2021), we compute the mean extinction time of the resistant population in this situation (mathematical details are stated in SI, Section E).

The comparison between our theoretical prediction and the simulation results in Fig. 5 shows that the timescale separation in our competitive Lotka-Volterra model captures well the simulation results, even for this relatively large evolutionary rate compared to the ecological processes. The carriage time is mainly determined by the frequency of the resistant strain in the overall bacterial population at the end of treatment. The carriage time is typically larger after biocidal treatment than after biostatic treatment, especially for antibiotic concentrations larger than the MIC of the sensitive strain (vertical dashed line), which is explained by the larger relative frequency of the resistant strain in the bacterial

population at the end of treatment (Fig. D.2 in the SI). This is because under biocidal treatment the sensitive population size is smaller than under biostatic treatment, because of the increased death rate, and the resistant subpopulation is, on average, larger (Fig. 4).

Stochastic effects do not reduce much the carriage time compared to a deterministic analysis (Fig. E.1 in the SI). Only for death competition, which results in larger variance of the stochastic process, or when the cost of resistance is smaller than in our default parameter set (Fig. E.2 in the SI), are stochastic effects important and the carriage time substantially reduced.

4 Discussion

We developed a fully stochastic model describing the within-host dynamics of antibiotic resistance in a treated host. We analyzed the survival probability of the resistant strain until the end of treatment and the resistant subpopulation size at the end of treatment. These results apply to commensal bacteria as well as to pathogens. Additionally, we compute the time after treatment until a commensal is replaced by an antibiotic-sensitive strain due to competition, which we refer to as the carriage time.

Generally, the survival probability, size of resistant subpopulation, and carriage time are all maximized at an intermediate antibiotic concentration (Figs. 3-5) in line with previous studies (Day and Read, 2016; Scire et al., 2019). We obtain new explicit analytical results for the probability of survival and derive predictions for the antibiotic concentration maximizing this probability. We find that in the scenario of biostatic treatment and death competition, this maximizing concentration is very close to, not necessarily below, the MIC of the sensitive strain. The distance between these two concentrations scales with the intrinsic growth rate of the sensitive strain and is independent of the level of resistance of the resistant strain. The smaller the intrinsic death rate, the closer is this maximizing concentration to the sensitive MIC. In the biocidal treatment scenarios, the condition cannot be evaluated in all generality and depends in particular on the choice of the antibiotic response curve (Eq. (3)). We find that the concentration maximizing the resistant survival probability is always close to the sensitive MIC. This was true in our default parameter set, for a broad range of sigmoid antibiotic response curves (Section H in the SI), in an alternative model where the bacterial population is limited by the host's immune response instead of intraspecific competition (Section I in the SI) and in previous models (e.g. Day and Read, 2016; Blanquart, 2019; Scire et al., 2019). If patients are generally prescribed antibiotic doses at or above the MIC of the sensitive drug, this implies that to limit the emergence, "hitting hard" (at the maximum tolerable dose) is best in the case of biostatic drugs and seems often to be best in the case of biocidal drugs. More importantly, this is also true to limit the establishment of resistance, i.e., a substantial growth of the resistant subpopulation during treatment.

One originality of our model is that we describe in detail the bacterial life cycle including density regulation on birth vs. death, and antibiotic effect on birth vs. death (but see Marrec and Bitbol (2020) for similar models). These considerations are particularly important in stochastic models. One interesting result is that the probability of emergence is larger when competition reduces the birth rate. Indeed, a bacterial subpopulation with low birth and low death rates has a higher chance of survival than one with high birth and high death rates, because the former strategy reduces the variance of the stochastic process, which reduces the stochastic loss when the resistant subpopulation is small. Another interesting result is that biocidal drugs lead to stronger competitive release than biostatic drugs. This is because biocidal drugs can eliminate the sensitive strain faster by driving the death rate to very large values, while the birth rate, which is affected by biostatic drugs, can only be reduced to zero. As a consequence, the probability of emergence of resistance, resistant subpopulation size and overall carriage time of resistant pathogens are larger for biocidal drugs.

In our parameterization, we assumed high drug resistance (large MIC differences between resistant and sensitive cells), which leads to large survival probabilities (up to 60%) and limited stochastic

effects. We also assumed a large cost of resistance ($\sim 10\%$), which reduced stochastic effects in the post-treatment phase. All stochastic effects would be stronger for smaller differences between the drug-sensitive and -resistant cells, i.e., weak resistance and weak cost of resistance. Precisely, the survival probability would be smaller and the differences generated by different antibiotic modes of action and density dependence would be larger. Generally though, a smaller cost of resistance will prolong the carriage time (Fig. E.2 in the SI). We also re-emphasize that the 'death' rate comprises both cell death and the outflux of the host compartment colonized by bacteria. For example for the human gut, it could be of the order of $\delta = 1$ day^{-1} (Asnicar et al., 2021), as in our parameterization ($\delta_S = \delta_R = 0.5$).

Our model has several limitations. We model a single antibiotic course of fixed duration (e.g. seven days). Variation in treatment length will likely affect the quantitative values, but not the qualitative pattern of the studied quantities. For example, compare the dashed and solid colored lines in Fig. 3 that correspond to an infinite treatment and a seven day treatment, respectively. Importantly the concentration maximizing the risk of resistance evolution remains unchanged. The last phase examining the duration of carriage of the resistant strain is less relevant if we consider pathogenic bacteria. In this case, as the total bacterial population size quickly recovers to pre-treatment values after the end of treatment (Fig. 1), it is likely that the host would undergo a second antibiotic course to cure the disease. However, bystander exposure to antibiotics in carriage (not infection), where our post-treatment phase applies, is the most common context of exposure for several important bacterial species (Tedijanto et al., 2018). Last, we assume that the antibiotic concentration is constant throughout treatment. In reality, the antibiotic concentration might fluctuate in time, which would impact the probability of emergence and the final population size. Pharmacokinetic-pharmacodynamic models have been studied in related contexts, but none directly compared resistance emergence and establishment to a constant concentration with a single drug (Chakrabarti and Michor, 2017; Yu et al., 2018; Nyhoegen and Uecker, 2023).

Our theoretical work suggests several interesting future experiments. Some of our findings have in fact already been tested experimentally. A larger resistant subpopulation size at the beginning of treatment increases the probability of survival and establishment of the resistant strain and the subpopulation size at the end of treatment (Alexander and MacLean (2020) and Eq. (B.2) in the SI). Another study investigated the population dynamics of a pathogen population under different drug modes of action and found that biocidal treatment reduces the population size more strongly than biostatic treatment (Coates et al. (2018) and our Fig. A.1 in the SI). Based on our theoretical results, further *in vitro* experiments could be conducted to characterize the probability of emergence of resistance depending on drug concentration, the model of density regulation and the drug mode of action. We predict that the differences between the two drug modes of action are strongest when density affects the birth rate (Fig. 4).

The validity of our prediction on the concentration maximizing the resistant survival probability in Eq. (3) can also be tested. To this end, one needs to first expose resistant bacteria at different antibiotic concentrations and study the location of the concentration that maximizes the survival probability. To evaluate Eq. (3) through experiments one could proceed as follows. The exponential growth rates (ρ_S, ρ_R) can be measured for different antibiotic concentrations. This will also provide an estimate for the antibiotic response curve (α_S, α_R). The derivatives of the antibiotic response curves can then be evaluated numerically, or alternatively, can be approximated by interpolation of two values of α_j (optimally measured slightly above and below the empirically found concentration that maximizes resistant survival). Our prediction that biostatic drugs always have a maximizing concentration at (or slightly above) the sensitive MIC, irrespective of the level of resistance, can even be tested without knowledge of the demographic parameters.

The within-host dynamics of antimicrobial resistance underpin the between-host epidemiolog-

ical dynamics of resistance. In fact, the dynamics of colonization by resistant strains and the slow dynamics of clearance of the resistant strain after treatment are key determinants of the intermediate equilibrium frequency reached by the resistant strain in a host population (Davies et al., 2019). Here, we produced mathematical results that help bridge the gap between the two scales. All quantities derived here (probability of survival, population size reached, carriage time) are relevant to epidemiological dynamics and determine the total shedding of resistance. Stochasticity implies that not all events of transmission of a resistant strain lead to survival and establishment of a resistant subpopulation; when establishment is successful, however, a greater resistant subpopulation is reached than in the equivalent deterministic model. These two effects may approximately compensate in terms of shedding of resistance: accounting for stochasticity leads to fewer hosts colonized by resistant strains, but to more resistance transmission per successfully colonized host. The carriage time is reduced by stochasticity, but this reduction is small for our choice of parameters (highly resistant strains with a strong cost; Figs. E.1 and E.2).

In conclusion, we developed a fully stochastic mathematical description of the emergence of a resistant subpopulation in hosts treated by antibiotics. This work could motivate experiments studying how drug resistance evolves from small populations of resistant cells, and better epidemiological models explicitly linking the within and between host scales.

Acknowledgements

P.C. has received funding from the European Union's Horizon 2020 research and innovation program under the Marie Skłodowska-Curie grant agreement PolyPath 844369. F.B. is funded by the ERC StG 949208 EvoComBac. F.D. is funded by ANR-19-CE45-0009-01 TheoGeneDrive. We are grateful to the High Performance Computing service of the University of Münster for providing computational resources on the computer cluster PALMA II. We are also thankful to the INRAE MIGALE Bioinformatics Facility (MIGALE, INRAE, 2018; Migale Bioinformatics Facility, `https://doi.org/10.15454/1.5572390655343293E12`) for providing computational resources during the early phase of this project.

References

Abel zur Wiesch, P., Kouyos, R., Abel, S., Viechtbauer, W., and Bonhoeffer, S. Cycling empirical antibiotic therapy in hospitals: Meta-analysis and models. *PLoS Pathogens*, 10(6):e1004225, 2014. doi: 10.1371/journal.ppat.1004225.

Alexander, H. K. and MacLean, R. C. Stochastic bacterial population dynamics restrict the establishment of antibiotic resistance from single cells. *Proceedings of the National Academy of Sciences*, 117 (32):19455–19464, 2020. doi: 10.1073/pnas.1919672117.

Andersson, D. I. and Hughes, D. Antibiotic resistance and its cost: is it possible to reverse resistance? *Nature Reviews Microbiology*, 8(4):260–271, 2010. doi: 10.1038/nrmicro2319.

Ankomah, P. and Levin, B. R. Exploring the collaboration between antibiotics and the immune response in the treatment of acute, self-limiting infections. *Proceedings of the National Academy of Sciences*, 111(23):8331–8338, 2014. doi: 10.1073/pnas.1400352111.

Asnicar, F., Leeming, E. R., Dimidi, E., Mazidi, M., Franks, P. W., Al Khatib, H., Valdes, A. M., Davies, R., Bakker, E., Francis, L., Chan, A., Gibson, R., Hadjigeorgiou, G., Wolf, J., Spector, T. D., Segata, N., and Berry, S. E. Blue poo: impact of gut transit time on the gut microbiome using a novel marker. *Gut*, 70(9):1665–1674, 2021. doi: 10.1136/gutjnl-2020-323877.

Bergstrom, C. T., Lo, M., and Lipsitch, M. Ecological theory suggests that antimicrobial cycling will not reduce antimicrobial resistance in hospitals. *Proceedings of the National Academy of Sciences*, 101 (36):13285–13290, 2004. doi: 10.1073/pnas.0402298101.

Blanquart, F. Evolutionary epidemiology models to predict the dynamics of antibiotic resistance. *Evolutionary Applications*, 12(3):365–383, 2019. doi: 10.1111/eva.12753.

Cantón, R. and Morosini, M.-I. Emergence and spread of antibiotic resistance following exposure to antibiotics. *FEMS Microbiology Reviews*, 35(5):977–991, 2011. doi: 10.1111/j.1574-6976.2011.00295.x.

Chakrabarti, S. and Michor, F. Pharmacokinetics and drug interactions determine optimum combination strategies in computational models of cancer evolution. *Cancer Research*, 77(14):3908–3921, 2017.

Coates, J., Park, B. R., Le, D., Şimşek, E., Chaudhry, W., and Kim, M. Antibiotic-induced population fluctuations and stochastic clearance of bacteria. *eLife*, 7, 2018. doi: 10.7554/elife.32976.

Constable, G. W. and McKane, A. J. Models of genetic drift as limiting forms of the Lotka-Volterra competition model. *Physical Review Letters*, 114(3), 2015. doi: 10.1103/physrevlett.114.038101.

Czuppon, P. and Traulsen, A. Fixation probabilities in populations under demographic fluctuations. *Journal of Mathematical Biology*, 77(4):1233–1277, 2018. doi: 10.1007/s00285-018-1251-9.

Czuppon, P. and Traulsen, A. Understanding evolutionary and ecological dynamics using a continuum limit. *Ecology and Evolution*, 11(11):5857–5873, 2021. doi: 10.1002/ece3.7205.

Davies, N. G., Flasche, S., Jit, M., and Atkins, K. E. Within-host dynamics shape antibiotic resistance in commensal bacteria. *Nature Ecology & Evolution*, 3(3):440–449, 2019. doi: 10.1038/s41559-018-0786-x.

Day, T. and Read, A. F. Does high-dose antimicrobial chemotherapy prevent the evolution of resistance? *PLoS Computational Biology*, 12(1):e1004689, 2016. doi: 10.1371/journal.pcbi.1004689.

Day, T., Huijben, S., and Read, A. F. Is selection relevant in the evolutionary emergence of drug resistance? *Trends in Microbiology*, 23(3):126–133, mar 2015. doi: 10.1016/j.tim.2015.01.005.

Ehrlich, P. Chemotherapeutics: Scientific principles, methods, and results. *The Lancet*, 182(4694): 445–451, 1913. doi: 10.1016/s0140-6736(01)38705-6.

Ewens, W. *Mathematical Population Genetics. I. Theoretical Introduction*. Springer, New York, 2004.

Fleming, A. Penicillin. Nobel lectures, physiology or medicine 1942–1962. Elsevier Publishing Company, 1964.

Gerlee, P. Weak selection and the separation of eco-evo time scales using perturbation analysis. *Bulletin of Mathematical Biology*, 84(5):52, 2022. doi: 10.1007/s11538-022-01009-3.

Haccou, P., Jagers, P., and Vatutin, V. A. *Branching Processes: Variation, Growth, and Extinction of Populations*. Cambridge Studies in Adaptive Dynamics. Cambridge University Press, 2005. doi: 10.1017/CBO9780511629136.

Hansen, E., Karslake, J., Woods, R. J., Read, A. F., and Wood, K. B. Antibiotics can be used to contain drug-resistant bacteria by maintaining sufficiently large sensitive populations. *PLoS Biology*, 18(5): e3000713, 2020. doi: 10.1371/journal.pbio.3000713.

Huijben, S., Bell, A. S., Sim, D. G., Tomasello, D., Mideo, N., Day, T., and Read, A. F. Aggressive chemotherapy and the selection of drug resistant pathogens. *PLoS Pathogens*, 9(9):e1003578, 2013. doi: 10.1371/journal.ppat.1003578.

Kendall, D. G. On the Generalized "Birth-and-Death" Process. *The Annals of Mathematical Statistics*, 19(1):1–15, 1948. doi: 10.1214/aoms/1177730285.

Kouyos, R., Klein, E., and Grenfell, B. Hospital-community interactions foster coexistence between methicillin-resistant strains of staphylococcus aureus. *PLoS Pathogens*, 9(2):e1003134, 2013. doi: 10.1371/journal.ppat.1003134.

Lee, C. Y., Cheu, R. K., Lemke, M. M., Gustin, A. T., France, M. T., Hampel, B., Thurman, A. R., Doncel, G. F., Ravel, J., Klatt, N. R., and Arnold, K. B. Quantitative modeling predicts mechanistic links between pre-treatment microbiome composition and metronidazole efficacy in bacterial vaginosis. *Nature Communications*, 11(1), 2020. doi: 10.1038/s41467-020-19880-w.

Marrec, L. and Bitbol, A.-F. Resist or perish: Fate of a microbial population subjected to a periodic presence of antimicrobial. *PLoS Computational Biology*, 16(4):1–19, 2020. doi: 10.1371/journal.pcbi.1007798.

Mouton, J. W., Ambrose, P. G., Canton, R., Drusano, G. L., Harbarth, S., MacGowan, A., Theuretzbacher, U., and Turnidge, J. Conserving antibiotics for the future: New ways to use old and new drugs from a pharmacokinetic and pharmacodynamic perspective. *Drug Resistance Updates*, 14(2):107–117, 2011. doi: 10.1016/j.drup.2011.02.005.

Murray, C. J., Ikuta, K. S., Sharara, F., Swetschinski, L., Aguilar, G. R., Gray, A., Han, C., Bisignano, C., Rao, P., Wool, E., ..., and Naghavi, M. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. *The Lancet*, 399(10325):629–655, 2022. doi: 10.1016/s0140-6736(21)02724-0.

Niewiadomska, A. M., Jayabalasingham, B., Seidman, J. C., Willem, L., Grenfell, B., Spiro, D., and Viboud, C. Population-level mathematical modeling of antimicrobial resistance: a systematic review. *BMC Medicine*, 17(1), 2019. doi: 10.1186/s12916-019-1314-9.

Nyhoegen, C. and Uecker, H. Sequential antibiotic therapy in the laboratory and in the patient. *Journal of The Royal Society Interface*, 20(198), 2023. doi: 10.1098/rsif.2022.0793.

Olofsson, S. K. and Cars, O. Optimizing drug exposure to minimize selection of antibiotic resistance. *Clinical Infectious Diseases*, 45:S129–S136, 2007. doi: 10.1086/519256.

Opatowski, L., Guillemot, D., Boëlle, P.-Y., and Temime, L. Contribution of mathematical modeling to the fight against bacterial antibiotic resistance. *Current Opinion in Infectious Diseases*, 24(3): 279–287, 2011. doi: 10.1097/qco.0b013e3283462362.

Otto, S. P. and Day, T. *A Biologist's Guide to Mathematical Modeling in Ecology and Evolution*. Princeton Univ. Press, Princeton, NJ, 2007.

Parsons, T. L. and Rogers, T. Dimension reduction for stochastic dynamical systems forced onto a manifold by large drift: a constructive approach with examples from theoretical biology. *Journal of Physics A: Mathematical and Theoretical*, 50(41):415601, 2017. doi: 10.1088/1751-8121/aa86c7.

Parsons, T. L., Lambert, A., Day, T., and Gandon, S. Pathogen evolution in finite populations: slow and steady spreads the best. *Journal of The Royal Society Interface*, 15(147):20180135, 2018. doi: 10.1098/rsif.2018.0135.

Read, A. F., Day, T., and Huijben, S. The evolution of drug resistance and the curious orthodoxy of aggressive chemotherapy. *Proceedings of the National Academy of Sciences*, 108:10871–10877, 2011. doi: 10.1073/pnas.1100299108.

Regoes, R. R., Wiuff, C., Zappala, R. M., Garner, K. N., Baquero, F., and Levin, B. R. Pharmacodynamic functions: a multiparameter approach to the design of antibiotic treatment regimens. *Antimicrobial Agents and Chemotherapy*, 48(10):3670–3676, 2004. doi: 10.1128/aac.48.10.3670-3676.2004.

Salas, J. R., Gaire, T., Quichocho, V., Nicholson, E., and Volkova, V. V. Modelling the antimicrobial pharmacodynamics for bacterial strains with versus without acquired resistance to fluoroquinolones or cephalosporins. *Journal of Global Antimicrobial Resistance*, 28:59–66, 2022. doi: 10.1016/j.jgar.2021.10.026.

Scire, J., Hozé, N., and Uecker, H. Aggressive or moderate drug therapy for infectious diseases? Trade-offs between different treatment goals at the individual and population levels. *PLoS Computational Biology*, 15(8):e1007223, 2019. doi: 10.1371/journal.pcbi.1007223.

Tedijanto, C., Olesen, S. W., Grad, Y. H., and Lipsitch, M. Estimating the proportion of bystander selection for antibiotic resistance among potentially pathogenic bacterial flora. *Proceedings of the National Academy of Sciences*, 115(51), 2018. doi: 10.1073/pnas.1810840115.

Tepekule, B., Uecker, H., Derungs, I., Frenoy, A., and Bonhoeffer, S. Modeling antibiotic treatment in hospitals: A systematic approach shows benefits of combination therapy over cycling, mixing, and mono-drug therapie. *PLoS Computational Biology*, 13(9):e1005745, 2017. doi: 10.1371/journal.pcbi.1005745.

Uecker, H. and Hermisson, J. On the fixation process of a beneficial mutation in a variable environment. *Genetics*, 188(4):915–930, 2011. doi: 10.1534/genetics.110.124297.

Yu, G., Baeder, D. Y., Regoes, R. R., and Rolff, J. Combination effects of antimicrobial peptides. *Antimicrobial Agents and Chemotherapy*, 60(3):1717–1724, 2016. doi: 10.1128/aac.02434-15.

Yu, G., Baeder, D. Y., Regoes, R. R., and Rolff, J. Predicting drug resistance evolution: insights from antimicrobial peptides and antibiotics. *Proceedings of the Royal Society B: Biological Sciences*, 285 (1874):20172687, 2018. doi: 10.1098/rspb.2017.2687.