Modeling continuous levels of resistance to multidrug therapy in cancer

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Abstract

Multidrug resistance consists of a series of genetic and epigenetic alternations that involve multifactorial and complex processes, which are a challenge to successful cancer treatments. Accompanied by advances in biotechnology and high-dimensional data analysis techniques that are bringing in new opportunities in modeling biological systems with continuous phenotypic structured models, we study a cancer cell population model that considers a multi-dimensional continuous resistance trait to multiple drugs to investigate multidrug resistance. We compare our continuous resistance trait model with classical models that assume a discrete resistance state and classify the cases when the continuum and discrete models yield different dynamical patterns in the emerging heterogeneity in response to drugs. We also compute the maximal fitness resistance trait for various continuum models and study the effect of epimutations. Finally, we demonstrate how our approach can be used to study tumor growth regarding the turnover rate and the proliferating fraction, and show that a continuous resistance level may result in a different dynamics when compared with the predictions of other discrete models.

Keywords: Multidrug resistance, Tumor growth, Phenotype structured model, Epimutation

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1. Introduction

The biological mechanisms responsible for the emergence of drug resistance and its propagation often involve a multifactorial and complex process of genetic and epigenetic alternations [1–3], that arise through a series of genetic and non-genetic changes [4–7]. Such changes can be due to drug administration (drug induced resistance) [8, 9], or they can emerge independent of therapy due to intrinsic mechanisms. Cancer cells may develop simultaneous resistance to structurally and mechanistically unrelated drugs, leading to multidrug resistance (MDR) [1, 2, 10]. The complex dynamical nature of MDR is one of the most challenging obstacles to successful treatment.

The complexity of the mechanisms underlying drug resistance has encouraged its study through mathematical modeling. Such models aim at providing quantitative tools for testing therapies that circumvent or at least delay the unfortunate consequences of drug resistance. Examples include the models of Goldie and Coldman [11–13] that are based on resistance due to point mutations. These works were proceeded by many studies considering stochastic models (including branching process and multiple mutations) to study MDR and optimal control of drug scheduling [14–17]. Alternative approach includes continuum deterministic models using ordinary differential equations, for example, modeling kinetic resistance [18] and point mutations [19], and partial differential equations, where spatial heterogeneity and vascularization can be readily incorporated [20, 22]. For additional approaches see [17, 23–28].

In addition to the aforementioned modeling approaches, the advance of biotechnology in collecting data characterizing the phenotype is bringing in new opportunities of mathematical modeling of biological systems. The most recent technology allows cytometry data to be collected up to O(50) dimensions, Methylation profiles in the scale of O(1000), and gene-expression profile in the scale of O(10000) [29, 31]. In particular, recent advances in single cell RNA sequencing technologies has enabled a new high-dimensional definition of cell states, that is on the order of 20,000 protein encoding genes that compose the
transcriptome [32, 35]. The high-dimensionality of the data makes it practically impossible to consider a meaningful model on the original space in which the data is collected. Thus, various dimension reduction techniques, such as, principal component analysis [36, 37], t-distributed stochastic neighbor embedding [30, 38, 39], diffusion maps [40, 41], and machine learning techniques [42, 43], have been employed to reduce the dimensionality and to identify only the critical directions. In contrast to classical biology and modeling approaches, where cell types are classified into discrete states and differentiation is considered as a stepwise process of binary branching decision, the new technologies and data analysis enabled considering cell differentiation as a continuous process that can be mapped into a continuum of cellular and molecular phenotypes [31, 33, 40]. In other words, the high-dimensional configuration space is mapped into a continuous trait in a lower-dimensional space. Figure 1 shows two examples of high-dimensional cell data mapped into a continuous trait in a lower dimensional space using stochastic neighbor embedding (viSNE) [38] and diffusion mapping [41]. This reveals the continuous phenotypic trait space where resistance can be locally characterized. For instance, the left figure shows that relapsed leukemia cells are associated with high expression of CD34, and the ALDH1 in the right figure is related to cancerous stem cells in mammary gland and breast cancer [44]. This opens the door to mathematical models that assume a continuous trait space [45, 46].

Among continuous phenotypic structured models, recent studies in [47–52] consider a continuous trait variable that represents the level of cytotoxic drug resistance. This framework allows to explicitly model the heterogeneous response to drugs and effectively study the selection dynamics under microenvironmental constraints and chemotherapy. The asymptotic distributions on the resistance trait space are obtained in [47], and the following works in [49, 51] extend it to include mutations and epimutations. The distribution of resistance levels can then be translated to therapeutic recommendation. The effectiveness of a combination of cytotoxic and cytostatic drugs when cytotoxic resistance emerge is studied in [47]. An optimal combination therapy to eliminate the most resis-
Figure 1: High-dimensional cell data projected into a lower dimensional continuous trait space, where the reduced dimensions are obtained by dimension reduction techniques. Figures are reproduced from the data provided in [38] and [41]. Figure (a) shows the CD34 expression level of 41 dimensional data [38] mapped into two dimensions by stochastic neighbor embedding (viSNE), and the relapsed leukemia cells are located at where CD34 is highly expressed. Figure (b) shows the ALDH1 expression level of 4773 dimensional data [41] mapped into three dimensions by diffusion mapping, where ALDH1 is related to cancerous stem cells in mammary gland and breast cancer [44].

tant clones is proposed in [52]. Moreover, [52] extends the framework that was restricted to solid tumor that is radially symmetric with a fixed boundary [48] to an asymmetric tumor growth model with moving boundary. However, this framework is limited to a single trait variable to a cytotoxic drug.

In this paper we extend the framework of [52] to multi-dimensional resistance trait. We compare our approach that allows for a continuous drug response to more traditional approaches that assume a discrete response to drugs. The paper is organized as follows. In section 2, we introduce a mathematical model for MDR assuming continuous trait variables. We parameterize our model as an extension of a discrete resistance state model in section 2.1 and compute the maximal fitness trait of resistance in section 2.2 for different types of continuum models. This allows us to characterize the cases when the solutions of the continuous models are qualitatively different than the corresponding discrete models. Section 2.3 presents simulation results for the different cases of cytotoxic and cytostatic drugs studied in 2.2. The impact of mutations and epimutations is studied in section 2.4. In section 3 we simulate tumor growth and resistance dy-
dynamics subject to MDR on different types of tumors characterized by turnover rates and the proliferating ratios. Our simulations correspond to the discrete MDR models studied by Komarova and Wodarz (2005) \[53\] and Gardner (2002) \[54\]. We observe that a combination therapy with multiple cytotoxic drugs is also effective in high turnover tumors using relatively high dosages. Increasing the dosage in low turnover tumor is effective only for certain drug uptake functions. In addition, the drug response function plays a key role in determining the tumor growth dynamics when combination therapy is administered using cell-cycle nonspecific cytotoxic drugs, such as Cyclophosphamide and Doxorubicin. Conclusions and future directions are discussed in section 4.

2. Models of multidrug resistance

Let us consider a cancer growth model under multidrug therapy that depends on an \(M\)-dimensional phenotype variable \(\theta = (\theta_1, ..., \theta_M) \in \Gamma \cong \Pi_{i=1}^M \Gamma_i\). The phenotype variable in the \(i\)-th direction \(\theta_i \in \Gamma_i = [0, 1]\) characterizes the resistance level to the \(i\)-th drug or the \(i\)-th drug mechanism, where \(\theta_i = 0\) and \(\theta_i = 1\) represents the fully-sensitive cells and fully-resistant cells to drug \(i\), respectively. The value of \(\theta_i\) can be obtained by normalizing the expression level of a gene or a gene cluster that is linked to the cellular levels of drug resistance and proliferative potential, such as ALDH1, CD44, CD117, or MDR1 \[38, 55–57\]. The governing equations follows the dynamics of the density of proliferating cells, \(n_P = n_P(t, \theta)\), and quiescent cells, \(n_Q = n_Q(t, \theta)\), as

\[
\partial_t n_P(t, \theta) = ((1 - w)R(t, \theta) - D - C_P(t, \theta) - q) n_P(t, \theta) + pn_Q(t, \theta) + w \int_{\Gamma} M(\theta, \vartheta) R(t, \vartheta) n_P(t, \vartheta) d\vartheta,
\]

\[
\partial_t n_Q(t, \theta) = q n_P(t, \theta) + (-p - D_Q - C_Q(t, \theta)) n_Q(t, \theta).
\]

The first term on the RHS of Eq. (1) is a growth term, \(R(t, \theta)\), which we assume depends on the resource level \(s_0(t)\) with the proliferation rate function \(\varphi(\theta)\) as \(R(t, \theta) = \varphi(\theta)s_0(t)\). Also, we assume an exponential growth by considering a constant apoptosis rate \(D\) for the proliferating cells and \(D_Q\) for the quiescent
cells. To consider a logistic growth, we substitute both terms with a density-dependent apoptosis term \( d\rho(t) \), where \( \rho(t) \) is the total number of cells

\[
\rho(t) = \int n_P(t, \theta) + n_Q(t, \theta) d\theta,
\]

and \( d \) is a constant that determines the cell capacity.

The net effects of the cytotoxic drugs on the proliferating and quiescent cells are denoted as \( C_P(t, \theta) \) and \( C_Q(t, \theta) \), respectively. These terms depend on the marginal drug effects, \( C_i = C_i(t, \theta; c_i(t)) \), the cell death rate due to the \( i \)-th drug, which is assumed to be a function of the drug concentration \( c_i(t) \). We either consider \( C_i(t, \theta) = \mu_i(\theta)c_i(t) \), where \( \mu_i(\theta) \) is the drug uptake function of the \( i \)-th drug, or the exponential kill model \[54\], \( C_i(\theta_i) = e^{-a_i(\theta_{max}-\theta_i)c_i(t)} \), where \( C_i \) represents the probability of the cell death due to the \( i \)-th drug. The net drug effect is modeled as \( C_P(t, \theta) = \Phi(C_1, \ldots, C_M) \), where \( \Phi \) is the overall drug effect function that can be taken for the cytotoxic drugs as

\[
C_P(t, \theta) = \Phi(C_1, \ldots, C_M) = 1 - \prod_i (1 - C_i), \tag{3}
\]

and similarly for \( C_Q \). The form \[3\] is valid when \( C_i \) is the probability of death due to the \( i \)-th drug \( (C_i \leq 1) \), and assuming that the drug effects are independent. Dependency between the drugs can be imposed through different choices of \( \Phi \), e.g., Copula functions \[58\] that are used to describe the dependence between random variables using multivariate probability distributions with prescribed marginal distribution functions. In addition to the cytotoxic drugs, we consider cytostatic drugs, which we assume delay the proliferation according to

\[
R(t, \theta) = \frac{\phi(\theta)s_0(t)}{1 + \Phi(C_1, \ldots, C_M)}.
\]

The net cytostatic drug effect delays the progression of the proliferating cells through the cell cycle. We assume an additive \( \Phi \):

\[
\Phi(C_1, \ldots, C_M) = \sum_i C_i.
\]

Proliferating cells enter the quiescent state at a rate \( q \) and quiescent cells return to the cycling compartment at a rate \( p \). These rates regulate the proliferating portion \( \delta(t) \doteq \int n_P d\theta/\rho(t) \). To balance a fixed ratio of proliferating
cells, namely the proliferating index $\delta^*$, the transfer rate $q$ can be computed as

$$q = (\max_\theta R(\theta) - D + D_Q)(1 - \delta^*) + p(1 - \delta^*)/\delta^*.$$ 

The last term in Eq. (1) is a mutation term. We assume that mutations occur at a rate $w$ during the proliferation cycle. The mutation is modeled as an integral term with a kernel function $M(\theta, \vartheta)$. $M(\theta, \vartheta)$ represents the probability of a mother trait $\vartheta$ mutating to a daughter trait $\theta$ that is taken as an asymmetric exponential function with mutation range $\ell$, i.e., $M(\theta, \vartheta) = M_0 \exp \left[ ((\theta - \vartheta)^2 / \ell^2) \right]$ for $\theta \geq \vartheta$, and zero otherwise. Here, $M_0$ is a normalizing constant. This model represents a mutation that gradually increases the resistance level through multiple mutations. A rare mutation that confers a complete drug resistance in a single step can be imposed with a discrete kernel function \[50\] and a smaller value of $w$.

2.1. Multidrug resistance models parameterized with a binary level of resistance

In this section, we simplify the model given by Eq. (1) to a model that assumes a binary trait space. In this case, cells are either fully-sensitive or fully resistant with respect to each drug, i.e., $\theta_i \in \{0, 1\}, \forall i$. To compare the discrete- and continuous-trait models, we parameterize the proliferation and drug function with the parameters related to the microenvironment selection as follows. We denote the proliferation rate of the fully-sensitive cells ($\theta = 0$) as $\gamma$, and assume that the proliferation rate of the fully-resistant cells ($\theta = 1$) is reduced by $\eta$. With a normalized constant resource level ($s_0 = 1$),

$$R(0) = \varphi(0) = \gamma, \quad R(1) = \varphi(1) = \gamma - \eta.$$ 

We scale the drug dosage $c(t)$ to represent the drug effect on the fully-sensitive cells and assume that the fully-resistant cells do not respond to the drug. This yields a drug uptake function for which $\mu(0) = 1$ and $\mu(1) = 0$. Hence, the effect of the cytotoxic drug $C(t, \theta) = c(t)\mu(\theta)$ boils down to

$$C(t, 0) = c(t), \quad C(t, 1) = 0.$$ 

See Table 1 for a summary of the fitness parameters.
parameters | biological meaning
---|---
$\gamma$ | maximum proliferation rate
$\eta$ | reduced proliferation due to resistance (selection gradient)
c($t$) | maximum apoptosis rate of sensitive cells due to drug

Table 1: Parameters of the proliferation and drug effect that yield the microenvironmental selection process [51].

The resulting model can be written as a dynamical system. For instance, we consider a single ($M = 1$) cytotoxic drug affecting the proliferating cells. There exists two cell states: sensitive cells, $n_S(t) = n_P(t, \theta = 0)$, and resistant cells, $n_R(t) = n_P(t, \theta = 1)$. In this case, the resulting system is

$$
\dot{n}_S = ((1 - w)\gamma - D - c(t)) n_S,
\dot{n}_R = w \gamma n_S + ((\gamma - \eta) - D) n_R,
$$

(4)

where, $D = d\rho(t)$, and $\rho(t) = n_S(t) + n_R(t)$. In the case of a single cytostatic drug affecting the proliferating cells, the dynamics follows

$$
\dot{n}_S = \left( (1 - w) \frac{\gamma}{1 + c(t)} - D \right) n_S,
\dot{n}_R = w \frac{\gamma}{1 + c(t)} n_S + ((\gamma - \eta) - D) n_R.
$$

(5)

In case of $M$ drugs, the resulting model will involve $2^M$ discrete cell state variables.

The binary models (4) and (5) yield an outcome where either the sensitive cells $n_S$ or the resistant $n_R$ cells dominate the population asymptotically depending on the fitness parameters. In particular, for Eq. (4), with fixed values of $\gamma$ and $\eta$, if the drug dosage is low, $c(t) < \eta - w\gamma$, the sensitive cells dominate, but if the drug dosage increases as $c(t) \geq \eta - w\gamma$, the resistant cells dominate the population. The same holds for Eq. (5) with a threshold $(\eta - w\gamma)/(\gamma - \eta)$. If the mutation during treatment is negligible ($w = 0$) [53], the thresholds become $\eta$ and $\eta/(\gamma - \eta)$ for models (4) and (5), respectively.

To connect between models with binary traits and models with continuous traits, we extend the binary models assuming that the proliferation and drug
effects are smooth and monotone with respect to $\theta$. This assumption (although
may not always hold) makes it possible to classify continuum scenarios and
helps in identifying cases in which the continuous traits dynamics is qualita-
tively different than the corresponding binary models. Since we only consider
proliferating cells, the transfer terms to the quiescent cells are removed from
Eq. (1), and we simulate
\[
\partial_t n(t, \theta) = (R(\theta) - D - C(t, \theta)) n(t, \theta).
\] (6)

Starting from the proliferation, we assume that cells that are resistant to
cytotoxic drugs use their resources to develop and maintain the drug resistance
mechanism\,[59, 60], that is, $\varphi'(\theta) < 0$. On the domain of $\theta \in [0, 1]$, the pro-
liferation function $R(\theta) = \varphi(\theta)$ can be characterized according to its concavity.
We consider three sample cases: $\varphi(\theta) = \gamma - \eta + \eta(\theta - 1)^2$, $\varphi(\theta) = \gamma - \eta \theta$, and
$\varphi(\theta) = \gamma - \eta \theta^2$. The cytotoxic drug effect $C(\theta) = c(t) \mu(\theta)$ can be modeled
similarly. Assuming that apoptosis decreases with an increased level of resis-
tance, we have $\mu'(\theta) < 0$. Accordingly, we consider three characteristic cases:
$\mu(\theta) = (\theta - 1)^2$, $\mu(\theta) = (1 - \theta)$, and $\mu(\theta) = (1 - \theta^2)$. The models we consider
are summarized in Table 2 and Figure 2.

| $\varphi(\theta)$ | concave up | linear | concave down |
|-----------------|-----------|--------|-------------|
| (1) $\gamma - \eta + \eta(\theta - 1)^2$ | (2) $\gamma - \eta \theta$ | (3) $\gamma - \eta \theta^2$ |
| $\mu(\theta)$ | (i) $(\theta - 1)^2$ | (ii) $(1 - \theta)$ | (iii) $1 - \theta^2$ |

Table 2: Classification of the continuous proliferation and drug effect functions depending on
the concavity. We consider three cases for both $R(\theta) = \varphi(\theta)$ and $C(\theta) = \mu(\theta)c(t)$ denoted as
case {1, 2, 3} and {i, ii, iii}, respectively.

2.2. Differentiating models with binary traits from models with continuous traits

To demonstrate the difference between models that are based on binary traits
and continuous-traits models, we compute the trait that achieves the maximal
fitness of Eq. (6) under different microenvironment conditions. We denote such
Figure 2: Models of proliferation rate $\varphi(\theta)$ and drug uptake $\mu(\theta)$ considering a continuous resistance trait space on $\theta \in [0, 1]$. We assume that the proliferation rate reduces from $\gamma$ to $\gamma - \eta$ as the resistance level increases, and the drug effect reduces from 1 to 0.

trait with the maximal growth rate as $\theta_M(c(t), \eta, \gamma) \doteq \arg \max_{\theta} (R(\theta) - C(\theta))$. Our choices of $R(\theta)$ and $C(\theta)$ in Section 2.1 yield nine cases that are presented in the following list. We comment that among the nine cases, six cases resemble the discrete model in a sense that the maximal fitness trait is binary, either fully-sensitive or fully-resistant, while three cases allow intermediate trait levels. This demonstrates that in certain circumstances, continuum models are necessary. We first consider the single cytotoxic drug setup that is comparable to the binary model (4). The results are summarized in Table 3.

- **Case (3,i).** The maximal growth rate is achieved at $\theta_M = c(t)/(\eta + c(t))$ that changes its value from $\theta_M(c = 0, \cdot, \cdot) = 0$ to $\lim_{c \to \infty} \theta_M(c, \cdot, \cdot) = 1$. This case allows an intermediate maximal fitness trait for any drug dosage $c(t) \in \mathbb{R}_+$. 

- **Case (3,ii).** The maximal growth rate is achieved at $\theta_M = c(t)/(2\eta)$. This model increases the maximal trait linearly in terms of the drug dosage when $c(t) \leq 2\eta$. For $c(t) > 2\eta$, the maximal fitness occurs at $\theta_M = 1$.

- **Case (3,iii).** The maximal growth rate is either achieved at $\theta_M = 0$ when $c(t) < \eta$, or at $\theta_M = 1$ when $c(t) > \eta$. Since the phenotype distribution

\[1\] For simplicity, we compute the maximal fitness trait following the assumption that mutations during treatment are negligible ($w = 0$) [53].
asymptotically converges to a delta function centered at \( \theta = 0 \) or \( \theta = 1 \), the overall quality of the solution is similar to the binary-trait model. We also remark that there exists a critical drug dosage at \( c(t) = \eta \) that yields multiple fitness traits.

- Case (2,i). This model is similar to the case (3,ii), but opposite in the sense that the maximal growth rate is achieved at \( \theta_M = 0 \) for \( c(t) < \eta/2 \), and increases as \( \theta_M = (2c(t) - \eta)/2c(t) \) for \( c(t) \geq \eta/2 \).

- Cases (2,ii), (2,iii), (1,i), (1,ii), and (1,iii). These models also yield a solution that is either concentrated at \( \theta_M = 0 \) or \( \theta_M = 1 \), similar to case (3,iii), that is, \( \theta_M = 1, \eta > c \), where \( 1_A \) is an indicator function on \( A \).

| \( \varphi \) | Case (1) | Case (2) | Case (3) |
|-----|--------|--------|--------|
| (i) | \( \theta_M = 1_{c>\eta} \) | \( \theta_M = \max\left(0, \frac{2c - \eta}{2c}\right) \) | \( \theta_M = \frac{c}{\eta + c} \) |
| (ii) | \( \theta_M = 1_{c>\eta} \) | \( \theta_M = 1_{c>\eta} \) | \( \theta_M = \min\left(\frac{c}{2\eta}, 1\right) \) |
| (iii) | \( \theta_M = 1_{c>\eta} \) | \( \theta_M = 1_{c>\eta} \) | \( \theta_M = 1_{c>\eta} \) |

Table 3: The selected trait with maximal growth rate \( \theta_M = \theta_M(c, \eta, \gamma) \) depending on the cytotoxic drug concentration \( c \) and the resource parameters \( \gamma \) and \( \eta \).

In addition to cytotoxic drugs, we also consider the drug uptake models in Table 2 for a single cytostatic drug that is comparable to the binary model (5). The maximal fitness traits for the different choices of proliferation rate functions and drug uptake functions are summarized in Table 4.

2.3. Simulation of continuum model in cytotoxic and cytostatic resistance

In this section, we simulate the model (6) for the cases shown in Table 2 and compare the results with the binary models (4)–(5). For the numerical simulations, we consider the maximal proliferation rate as \( \gamma = 0.66 \) per day,
Table 4: The selected trait with maximal growth rate $\theta_M = \theta_M(c, \eta, \gamma)$ depending on the cytostatic drug concentration $c$ and the resource parameters $\gamma$ and $\eta$. We remark that $\theta_M$ are taken as 0 or 1 in cases (2,i) and (3,ii) similar to Table 3.

| $\varphi$ | Case (1) | Case (2) | Case (3) |
|-----------|----------|----------|----------|
| (i)       | $\theta_M = 1_{c>\eta\gamma}$ | $\theta_M = \frac{\eta}{2\gamma} - \sqrt{\frac{C_{2i}}{\eta\gamma}}$ where $C_{2i} = \frac{\eta}{2\gamma} - 1$ | $\theta_M = \frac{C_{1i}}{2} - \sqrt{\frac{C_{1i}^2}{4} - \frac{\eta}{\gamma}}$ where $C_{1i} = 1 + \frac{1}{c} + \frac{2}{\gamma}$ |
| (ii)      | $\theta_M = 1_{c>\eta\gamma}$ | $\theta_M = 1_{c>\eta\gamma}$ | $\theta_M = C_{1ii} - \sqrt{C_{1ii}^2 - \frac{\eta}{\gamma}}$ where $C_{1ii} = 1 + \frac{1}{c}$ |
| (iii)     | $\theta_M = 1_{c>\eta\gamma}$ | $\theta_M = 1_{c>\eta\gamma}$ | $\theta_M = 1_{c>\eta\gamma}$ |

corresponding to a cell cycle of approximately 25 hours [61, 62]. We also assume that the reduction in proliferation of the resistant cells is $\eta = 0.132$ per day based on the experiments of non-small lung cancer cells exposed to Erlotinib [59], where the growth rate of resistant cell is reduced by approximately 70%.

Experiments with HL60 leukemic cells exposed to vincristine [63] and calculation in [51] further support this assumption. We assume a logistic growth by $D = d\rho(t)$, where the apoptosis constant that represents the average death rate is taken as $d = 0.66 \cdot 10^{-8}$. This corresponds to a cell capacity of $10^8$ [63] assuming a solid tumor of size $1\text{cm}^3$ prior to angiogenesis [64] and a tumor cell volume $10^{-9} \sim 3 \cdot 10^{-8}\text{cm}^3$ [65, 66].

Figure 3: Total number of sensitive and resistant cancer cells in log scale using the binary-trait model (4) for different dosages of cytotoxic drug. The outcome is asymptotically binary, where either the sensitive or resistant cells dominate depending on the drug dosage with a threshold $c_1 = \eta = 0.132$.  

\[0 100 200\]
\[0 5 10\]
\[0 100 200\]
\[0 5 10\]
\[0 100 200\]
\[0 5 10\]
Figure 4: The dynamics of the resistance profile of the cancer cells in the continuous-trait model. The drug dosages are considered from \( c_1 = 0 \) to 0.8 and the shown results are at time \( t = 30, 60, \) and 90. Cases (3,i), (3,ii), and (2,i) yield a distribution with an intermediate resistance level of maximal fitness, where the maximum trait occurs at \( \theta_M(c_1) = c_1 + 0.132 + c_1, \) \( \theta_M(c_1) = c_1 + 0.264, \) and \( \theta_M(c_1) = 2c_1 - 0.132 + c_1, \) respectively. Cases (2,ii) and (1,iii) result in a distribution that is similar to the binary-trait model, either concentrated at the fully sensitive or fully resistant trait (see Table 3).

In Figure 3, we first present the result of the binary-trait model showing that either the fully-resistant or the fully-sensitive cells survive depending on the drug dosage \( c_1(t) \) compared to \( \eta = 0.132. \) The total number of sensitive and resistant cells, \( n_S(t) \) and \( n_R(t), \) are plotted in log scale with a constant drug dosage up to time \( t = 200. \) We observe that when \( c_1 = 0 < \eta, \) the sensitive cells dominate at \( t = 200, \) however, when the drug dosage increases to \( c_1 \geq 0.4 > \eta, \)
the resistant cells dominate. When $c_1 = 0.2 > \eta$, but close to $\eta$, the resistant cells will eventually dominate.

In contrast, Figure 4 shows the cancer cell density $n(t, \theta)$ of the continuous-trait model (6) subject to cytotoxic drug for cases (3,i), (3,ii), (2,i), (2,ii), and (1,iii). We vary the constant cytotoxic drug dosage from $c_1 = 0$ to $0.8$ and compute the solution up to time $t = 90$. Case (3,i) always yields an intermediate level of maximal fitness trait of resistance level $\theta_M(c_1) = \frac{c_1}{\eta + c_1}$. Case (3,ii) also yields intermediate levels of $\theta_M(c_1) = \frac{c_1}{2\eta}$ when $c_1 \leq 2\eta = 0.264$, and $\theta_M(c_1) = 1$ otherwise. Alternatively in case (2,i), $\theta_M(c_1) = 0$ when $c_1 < \eta/2 = 0.066$, and $\theta_M(c_1) = \frac{2c_1 - \eta}{2c_1}$ otherwise. These simulations are consistent with Table 3.

Moreover, we observe that the transition from the sensitive to the resistant trait is faster in cases (3,ii) and (2,i) compared with case (3,i), and even more rapid in cases (2,ii) and (1,iii). In particular, cases (2,ii) and (1,iii) result in a distribution that is either concentrated at the fully sensitive or fully resistant trait with a threshold $c_1 = \eta = 0.132$.

Figure 5: Total number of cancer cells $\rho(t)$ up to $t = 100$ simulated with the binary model (4) and continuous model (6). As the cytotoxic drug is increased, $t^*_\rho$ is delayed. The total number of cells at $t_s$ when the tumor growth slows down is monotonically reduced as the drug dosage increases in the continuum case (3,i), while it is not in the binary model and case (1,iii). In particular, the dynamics is identical in the binary model when the dosage is relatively high as $c_1 > 0.132$.

In addition to the resistance trait density, the following quantities of interest are computed. We denote the time that the tumor size $\rho(t) = 5 \cdot 10^7$ as

$$t^*_\rho \doteq \min \{ t \mid \rho(t) \geq 5 \cdot 10^7 \}.$$
In addition, the full cell capacity is approximately computed as \( \rho(t_s) \), where 
\[ t_s = \min \{ t \geq t^*_\rho | \rho'(t)/\rho(t^*_\rho) \leq 0.01 \} \], the time when tumor growth slows down.

Figure 6: Comparison between the binary model (4) and continuous model (6) regarding the time \( t^*_\rho \) and cell capacity \( \rho(t_s) \) in terms of cytotoxic drug dosage \( c_1 \). The binary model yields an identical result when the drug dosage is \( c_1 \geq 0.3 \), while the results of the continuum models change gradually. Moreover, \( t^*_\rho \) varies depending on the choice of continuum models and the measured time is shown to be more sensitive to the choice of the drug effect function than to the proliferation function.

Figure 5 compares the dynamics of the total number of cancer cells \( \rho(t) \) using the continuous model (6) and binary model (4) up to \( t = 100 \). The times \( t^*_\rho \) and \( t_s \) are delayed as the cytotoxic drug dosage increases. However, in the binary model, the results are essentially identical when the dosage is relatively high as \( c_1 > \eta = 0.132 \). Moreover, the tumor size of approximate full capacity \( \rho(t_s) \) in the continuum case (3,i) is gradually reduced as the drug dosage increases, which is not the case in the binary-trait model and case (1,iii). The results of \( t^*_\rho \) and \( \rho(t_s) \) with respect to the cytotoxic drug dosage \( c_1 \) shown in Figure 6, where the distinction between the binary and continuum models are more apparent. The binary model yields an identical result after the drug dosage increases above \( c_1 \geq 0.3 \), while the continuum models show a gradual change depending on the drug dosage. We observe that with our model parameters the results are more sensitive to the choice of the drug effect function (case i, ii, iii) than to the proliferation function (case 1, 2, 3).

The case of a cytostatic drug comparing the continuous model (6) and binary model (5) is shown in Figures 7 and 8. The resistance trait distribution considering cases (3,i), (2,ii), and (1,iii) are plotted in Figure 7. The interme-
Figure 7: The cancer cell distribution using continuum model \(6\) for different dosages of cytostatic drug at time \(t = 30, 60, 90\). The case (3,i) shows a smooth transition of intermediate maximal resistance trait as \(\theta_M(c_1) = 3 + 1/2c_1 - \sqrt{4 + 3/c_1 + 1/4c_1^2}\). On the other hand, cases (2,ii) and (1,iii) show maximal trait either at the most sensitive or the most resistant trait depending on the drug dosage threshold \(c_1 = 0.25\) (see Table 4).

Figure 8: Comparison between the binary model \(5\) and continuous model \(6\) regarding the time \(t^*_\rho\) and cell capacity \(\rho(t_s)\) with respect to the cytostatic drug dosage \(c_1\). In this case, the binary model also yields a gradual change regarding the drug dosage, still it varies from the results of different continuum models.

diate resistance level of maximal fitness is achieved in case (3,i) for all drug dosages \(c_1\) at \(\theta_M(c_1) = C_{1i}/2 - \sqrt{C_{1i}^2/4 - 5}\), where \(C_{1i} = 6 + 1/c_1\), similar to the results of using cytotoxic drugs. We also observe a binary outcome either at the most sensitive or the most resistant trait depending on the drug dosage threshold \(c_1 = \eta/\gamma - \eta = 0.25\). The time \(t^*_\rho\) and approximate capacity \(\rho(t_s)\)
are shown in Figure 8. In contrast to the cytotoxic drug case, the binary model also shows a gradual change as a function of the drug dosage. Still, the results obtained by the binary and continuous models are different.

2.4. Epimutation in drug resistance

In this section, we investigate the effect of epimutation on the drug resistance dynamics of cancer cells. Phenotypic variants in cancer cell populations emerge not only from genetic mutations, but also due to epimutations. Epimutations are heritable changes in gene expression that do not alter the DNA, but contribute to the phenotypic instability [67–71]. Recent experiments demonstrate that such non-genetic instability and phenotypic variability allows cancer cells to reversibly transit between different phenotypic states [63, 72, 73] and contributes to development of resistance to cytotoxic drugs [75, 76]. In the continuous phenotypic models, epimutation can be readily modeled as a diffusion term assuming that random epimutations yield infinitesimally small phenotypic modifications [51, 77, 78]. The dynamics of proliferating cells in Eq. (6) with an epimutation rate $\nu$ can be written as

$$\partial_t n(t, \theta) = \left( R(\theta) - d\rho(t) - C(\theta) \right) n + \nu \frac{\partial^2 n}{\partial \theta^2}. \quad (7)$$

The asymptotic distribution of the continuum model with epimutation for the case (3,i) is derived in [51]. Here, we study the effect of epimutation in different continuum models.

Figure 9 shows the resistance trait density $n(t, \theta)$ with epimutation using Eq. (7) corresponding to cases (3,i) and (1,iii) when the rate of epimutation is $\nu = 10^{-2}$. Although the maximum fitness trait is similar to the results without epimutations in Figure 3, the phenotypic instability yields a significantly more heterogeneous population, not only in case (3,i), where the maximal fitness trait is intermediate, but also in case (1,iii), where the distribution becomes a Dirac-delta function at the boundary trait without epimutations.

We now study the effect of epimutations on the time $t^*_\rho$ that the tumor size reaches a certain size in different models subject to cytotoxic drugs. In
Figure 9: The cancer cell distribution using continuum model (7) with nonzero epimutation rate $\nu = 10^{-2}$. The results shown are for different drug dosages at times $t = 30, 60, 90$. While the maximal resistant traits are similar to the results without the epimutations as in Figure 4, the cell population is significantly more heterogeneous.

Figure 10: The comparison of $t_\rho^*$ using the binary models (4)–(5) with mutation rate $w = 10^{-2}$ compared with the model with no mutations ($w = 0$). In general, mutations result with an earlier relapse due to an increased portion of resistant cells, when the drug dosage is sufficiently high, i.e., $c_1 \geq 0.2$ with a cytotoxic drug and $c_2 \geq 0.5$ with a cytostatic drug.

In particular, we compare epimutations with regular mutations. Figure 10 shows the time of relapse using the binary models (4)–(5) with and without mutations of rate $w = 10^{-2}$ initiated from $n_S(0) = 0.99$ and $n_R(0) = 0.01$. In general, mutations accelerate the relapse time by increasing the proportion of resistant cells under a sufficiently high dosage. We remark that this is similar in the continuum models, when using the asymmetric mutation kernel $M(\theta, \vartheta)$ described section 2. However, Figures 11 and 12 show that epimutations in the continuum model (7) often delay the relapse time. We consider two ini-
Figure 11: The comparison of $t^*_{\rho}$ using the epimutation model (7) subject to cytotoxic drugs. (a) and (b) correspond to different amount of preexisting resistance, modeled by the initial conditions $n_a(\theta)$ and $n_b(\theta)$, respectively. While mutations in the discrete case accelerate the relapse time, epimutations in the continuum models often delay the relapse time, especially with the initial condition $n_a$. With the initial condition $n_b$, epimutations accelerate the relapse in case (i), but in case (iii) only for a certain range of the drug dosage.

Initial conditions: (a) $n_a(\theta) = n_0 \exp[-\theta^2/l_0]$, where we set $l_0 = 0.0739$ and $n_0$ so that $\int_{0.5}^{1} n(t = 0, \theta) d\theta = 0.01$ and $\rho(0) = 1$; and (b) a linear distribution $n_b(\theta) = -0.98\theta + 0.99$, which has a larger population of resistant cells.

In Figure 11, using the epimutation model (7) subject to cytotoxic drugs, we observe that $t^*_{\rho}$ is delayed with the initial condition $n_a$, especially in case (iii) with a larger rate $\nu$. However, epimutations with initial condition $n_b$ accelerate the relapse in case (i), and also for a certain range of drug dosages in case (iii). For a higher cytotoxic dosage $c_1$ in case (iii), the relapse time is again delayed. Similarly, Figure 12 shows the effect of epimutations on the continuum model (7) subject to cytostatic drugs. Compared with the cytotoxic drugs, resistance to cytostatic drugs is less affected by epimutation especially when starting with the initial condition $n_a$. However, an earlier relapse is observed with the initial condition $n_b$ in both models (i) and (iii).
Figure 12: The comparison of $t^*_p$ using the epimutation model (7) subject to cytostatic drugs. (a) and (b) correspond to different amounts of preexisting resistance, modeled by the initial conditions $n_a(\theta)$ and $n_b(\theta)$, respectively. Compared with the cytotoxic drugs, resistance to cytostatic drugs is less affected by epimutations especially with the initial condition $n_a$. However, an earlier relapse is observed with the initial condition $n_b$.

In conclusion, compared with regular mutations that give advantage to tumor growth under drug administration, epimutations have more diverse effects that can either promote or slow down tumor growth depending on other circumstances, including the drug uptake function and the initial conditions.

3. Simulating tumor growth under multidrug therapy

In this section we demonstrate how our continuous phenotype structured modeling framework can be used to study MDR. The impact of the tumor’s turnover rate and the proliferating fraction of cancer cells have been studied within a discrete phenotype framework by Komarova and Wodarz (2005) [53] and by Gardner (2002) [54]. Here, we compare the results obtained with our approach with the conclusions of [53, 54].
3.1. Multidrug resistance: tumor turnover rate

The impact of the turnover rate in tumor growth and resistance dynamics has been studied by Komarova and Wodarz (2005) \[53\]. Their model assumes two discrete states for \( M \) cytotoxic drugs, adding to \( 2^M \) discrete resistance levels. The model assumes a constant growth rate \( R \), a constant death rate \( D \), and is independent of the cell-cycle. Komarova and Wodarz conclude that when comparing tumors of identical sizes at detection, high turnover tumors \((R \approx D)\) have a higher probability of treatment failure than low turnover tumors \((R \ll D)\). Moreover, a combination therapy \((M > 1)\) is less likely to have an advantage over single-drug therapy in tumors with high turnover rates. In contrast, in the continuum models we show that depending on the proliferation and drug response functions, a combination therapy to high turnover tumor can be more effective than a single drug treatment. This is the case with relatively higher dosages when the drug uptake follows model (i). In addition, increasing the dosage in low turnover tumors is effective in delaying the tumor relapse when the drug uptake follows model (i), but not in model (iii).

The simulation we present is computed using the continuum model \((6)\) with the different drug response functions in Table 2. As in \[53\], we assume a constant proliferation rate \( R = 1 \), and model the high and low turnover tumor by setting \( D = 0.9 \) and \( D = 0.1 \), respectively. The cytotoxic drug effect is taken as \( C_P(\theta) = c(t)\Phi(\theta) \), where we consider a single parameter \( c \) for the drug dosage, and \( \Phi(\theta) = 1 - \prod_{i=1}^{M}(1 - \mu_i(\theta)) \) with the uptake functions \( \mu_i(\theta) \). We consider the drug dosages around \( c(t) \approx 0.1 \) in high turnover tumors and \( c(t) \approx 0.9 \) in low turnover tumors.

Figure 13 presents the cell density in the resistance trait space using the continuum model \((6)\) subject to a combination therapy using two cytotoxic drugs \((M = 2)\). We consider a high turnover tumor with the uptake functions of cases (i,i), (i,iii), and (iii,iii), and set the drug dosage as \( c = 0.1, 0.2, 0.4 \). The distributions shown are cancer cell densities in log scale, \( \log(n(t,\theta_1,\theta_2)) \), at time \( t = 100 \). The marginalized distribution in each resistance trait is similar to the results of section 2.3 where case (iii) yields more localized distributions near
Figure 13: Phenotype distribution in the continuum resistant space using two drugs with the drug uptake functions of cases (i,i), (i,iii), and (iii,iii) computed using Eq. (6). The distributions shown are cancer cell densities in log scale, \( \log(n(t, \theta_1, \theta_2)) \), at time \( t = 100 \) for drug dosages \( c = 0.1, 0.2, \) and 0.4. The distribution is more localized near \( \theta_1 = 0 \) or 1 in case (iii) compared with case (i).

\( \theta = 1 \) in relatively higher dosages compared to case (i).

Figure 14: The total number of cancer cells \( \rho(t) \) using the continuum model (6) and case (i) with \( M = 1, \ldots, 5 \) cytotoxic drugs. As the drug dosage \( c \) and the number of drugs \( M \) are increased, the relapse time is delayed. Increasing the number of drugs to \( M \geq 2 \) is effective not only in low turnover rates but also in the high turnover rates with relatively high dosages \( c \geq 0.2 \).
We now compare the responses of high and low turnover tumors with respect to the number of drugs $M$ in the continuous models. Figure 14 shows the total number of cells $\rho(t)$ up to $t = 100$ for an increasing number of drugs $M = 1, \ldots, 5$, and increasing drug dosages. We choose case (i) for the drug uptake function. As expected, we observe a delayed growth with an increased number of drugs and increased dosages. While increasing the number of drugs is not effective in high turnover tumors in the model of [53], it is effective in the continuum model (6) with the drug update model (i) and high dosages $c \geq 0.2$. Figure 15 compares the total number of cells in four different continuum models, combining the drug effect (case (i), (iii)) and the turnover rate ($D = 0.9, 0.1$). We observe that increasing the drug dosage over a certain threshold is less likely to delay the relapse time in low turnover tumor for which the drug uptake follows case (iii). It is effective in drug uptake case (i).

Finally, Figure 16 shows the effect of increasing the number of drugs assuming a logistic growth model by taking $D = d\rho(t)$ in Eq. (6). In this case, the dynamics does not depend on the turnover rate $d$ except that the cell capacity changes. The results are shown for $d = 10^{-8}$, and we remark that taking $d = 9 \cdot 10^{-8}$ shows essentially no difference. However, the relapse does depend on the choice of a continuum model. Increasing the number of drugs delays the
relapse in both cases (i) and (iii), but more so in case (iii) compared with (i).

We conclude that in addition to the turnover rate, the drug uptake function of the continuum model is also important in controlling the outcome of the treatment. In particular, a combination therapy with multiple drugs is effective not only in low turnover tumors, but also in high turnover tumors with the drug uptake case (i). Moreover, a high cytotoxic drug dosage in low turnover tumor with case (iii) is less effective than case (i). The drug uptake function is often more important than the turnover rate in determining the outcome of the tumor growth and relapse, particularly with a logistic growth condition.

3.2. Multidrug resistance: heterogeneity due to the proliferating index

Gardner (2002) proposed an individually tailored model based on the tumor cell kinetics of patients following heterogeneous colonies of proliferating and quiescent cells. This study considered multidrug resistance to six specific drugs, including two cell-cycle specific (CS) cytotoxic drugs, 5-Fluorouracil and Methotrexate, that only affect the proliferating cells; two cell-cycle non-specific (nCS) cytotoxic drugs, Cyclophosphamide and Doxorubicin, that kill
both proliferating and quiescent cells; and two cytostatic drugs, Tamoxifen and Herceptin. The model assumed discrete levels of resistance in addition to the parameters of cell division rates, apoptotic rates, response to drugs, and evolution of drug resistance. It then used the discrete model to predict drug combinations and schedules that are likely to be effective in reducing the tumor size.

Figure 17: The drug effect $C_i(\theta_i)$ at resistance level $\theta_i \in \{0, 0.5, 1\}$ of the six drugs used in \cite{54}. The drugs include two CS cytotoxic drugs: 1) 5-Fluorouracil, and 2) Methotrexate; two nCS cytotoxic drugs: 3) Cyclophosphamide, and 4) Doxorubicin; and two cytostatic drugs: 5) Tamoxifen, and 6) Herceptin. The exponential kill models can be categorized into the continuum models of cases (ii) and (iii).

The governing system in \cite{54} assumes three discrete drug resistance levels, $\theta_i = \{0, 0.5, 1\}$, for each of the six drugs, and it is similar to Eqs. (1)-(2):

\begin{align*}
\dot{n}_P &= ((1 - w) R - C_P - q) n_P + p n_Q + wM(n_P), \\
\dot{n}_Q &= q n_P + (-p - D_Q - C_Q) n_Q.
\end{align*}

Here $n_P$ and $n_Q$ are defined on 3\(^6\) discrete resistance levels. In addition, $C_P$ includes the effect of apoptosis of proliferating cells of rate $D$, the quiescent cells die as a result of necrosis of rate $D_Q$, and $M$ denotes the mutation term similar to Eq. (1) \cite{54}. The transfer rates from the quiescent cells to the proliferating cells to balance a fixed ratio of proliferating cells $\delta^*$ is $q = (R - D + D_Q)(1 - \delta^*) + p(1 - \delta^*)/\delta^*$. We denote the CS cytotoxic drugs as $C_1$ and $C_2$, the nCS cytotoxic drugs as $C_3$ and $C_4$, and the cytostatic drugs as $C_5$ and $C_6$. The drug effects are modeled using the exponential
kill model \[81\] as \(C_i(\theta_i) = R \left[ 1 - e^{-a_i(\theta_{max} - \theta_i)c_i(t)} \right]\) for the CS cytotoxic drug \((i = 1, 2)\), \(C_i(\theta_i) = 1 - e^{-a_i(\theta_{max} - \theta_i)c_i(t)}\) for the nCS cytotoxic drug \((i = 3, 4)\), and \(C_i(\theta_i) = z_i \left[ 1 - e^{-a_i(\theta_{max} - \theta_i)c_i(t)} \right]\) for the cytostatic drug \((i = 5, 6)\), where \(\theta_{max} = 1\) and the domain of resistance trait is taken at three discrete levels \(\theta_i \in \{0, 0.5, 1\}\). The net drug effects are taken as
\[
C_P(t, \theta) = 1 - (1 - D) \prod_{i=1}^{4} (1 - C_i(\theta_i; c_i(t))) , \\
C_Q(t, \theta) = 1 - \prod_{i=3}^{4} (1 - C_i(\theta_i; c_i(t))) , \\
R(t, \theta) = \frac{\varphi(\theta)}{1 + \sum_{i=5}^{6} C_i(\theta_i; c_i(t))} .
\]

Figure 17 shows the three discrete levels of drug effect using the dosages \(c_1, \ldots, c_6\) from \[54\] (see Appendix A). We note that although Gardner considers three levels of resistance, the cells with sensitive levels \(\theta_i = 0\) and \(\theta_i = 0.5\) of \(i = 1, 2, 3, \) and \(6\) have similar response to the drug. Moreover, the exponential kill model of \(C_1, C_2, C_3,\) and \(C_6\) based on the concavity can be classified as our case (iii), and \(C_4\) and \(C_5\) as case (ii). In the following simulations, we assume that the proliferation \(R\) and the drug effects \(C_i\) in Eqs. (1)–(2) follow the models as in Table 2 with the net drug effect as in (9), and compare the results with the discrete model (8). See Appendix A for the model parameters.

Figure 18 compares the result of the discrete model (8) and the continuum model (1)–(2), in particular with regards to the drug \(C_2\). Shown is the cell distribution on the resistance trait space of drug \(C_2\) in log scale when using no drug, a single drug \(c_2\), and all 6 drugs. Here, the continuum model is taken as the exponential kill model that can be classified as cases (iii) and (ii). As expected from the shape of the uptake function in Figure 17, the distribution in the \(\theta_2\) trait space is concentrated at the boundary traits, similarly to the discrete model. However, the continuum model predict emerging cells with intermediate levels of resistance, and the degree of heterogeneity in the resistance level can be quantitatively computed.

\[2\] \(n_i(t, \theta_2) = \int_{\Gamma_i} n_P(t, \theta) + n_Q(t, \theta)d\theta_i\), where \(\theta_i^c\) is the vector of \(\theta\) except the \(i\)-th index \(\theta_i\) and \(\Gamma_i^c\) is its domain.
Figure 18: The cell distribution in the resistance trait space of $C_2$ in log scale, $\log(n(t, \theta_2))$, using no drug, a single drug of $C_2$, and all drugs. The plots compare the discrete model (top) and the continuum model (bottom). Due to the shape of the exponential kill model (case (iii)), the cell distribution of the continuum model is concentrated at the boundary traits similarly to the discrete model. However, the continuum model reveals the cell distribution in the intermediate levels and the degree of heterogeneity in the resistance trait.

Figure 19 compares the sensitivity of the tumor size with respect to the drug dosage between the continuum model (1)–(2) and the discrete model (8). For comparison, we plot the normalized total number of cells in log scale at time $t = 200$ that is normalized by the mean. Here, two drugs are applied, either $(C_1, C_3)$ or $(C_3, C_6)$, with different weighted dosages $\omega_i c_i$, where $\omega_i = 0, 0.2, ..., 1$. The results show that the tumor size $\rho(t)$ in the continuum model is more sensitive to the drug dosage, with variation of a larger order of magnitude compared with the results of the discrete model. In addition, the effects of drugs $C_1$, $C_3$, and $C_6$ in the discrete model are binary depending on whether the drug is applied ($\omega_i \geq 0.2$) or not ($\omega_i = 0$). In contrast, the continuum model shows a gradual decay when increasing the dosage. Figure 19 also shows the total number of cells when all six drugs are applied. We observe that $\rho(t)$ significantly depends on the choice of model, as the tumor size varies by two orders of magnitudes.
Figure 19: Comparison of normalized total number of cells \( \log(\rho(t)) \) at \( t = 200 \) when two drugs, either \([C_1, C_3]\) or \([C_3, C_6]\), are applied in difference dosages \( \omega \). Top: three discrete levels of resistance (8). Bottom: the continuum model (1)–(2). In the discrete model, the effects of drugs \( C_1, C_3, \) and \( C_6 \) are binary depending on whether the drug is applied or not, while the continuum models show gradual changes. The figure on the right shows the results of using all six drugs, where the tumor size significantly depends on the choice of model (two orders of magnitude) around \( t = 200 \).

Figure 20: Comparison of the mean resistant trait \( E[Q_i(t, \theta_i)] \) to the \( i \)-th drug. Each column corresponds to different types of drug: (a) CS cytotoxic, (b) nCS cytotoxic, and (c) cytostatic drug. Using the discrete model (8), the mean resistance level increases to \( \theta_i = 1 \) in all drugs, i.e., the cancer cell population is dominated by cells that are resistant to all six drugs. In the continuum model (1)–(2), resistance to CS cytotoxic drugs and cytostatic drug develops faster in case (i) compared with (iii), while the resistance to nCS cytotoxic drug arises faster in case (iii).
Figure 20 compares the mean resistance level \( E[Q_i(t, \theta)] \) up to \( t = 200 \) when all 6 drugs are applied. While the mean resistance level in \( \theta_i \) implies the dominating resistance to the \( i \)-th drug, we observe distinct results in different models. First, using the discrete model (8), the resistance level in each drug eventually converges to the most resistant cells \( \theta_i = 1 \). This implies that the surviving cancer cells are only the ones that are fully resistant to all six drugs. However, the continuum model (1)–(2) shows a more gradual increase of resistance. Moreover, the resistance to nCS cytotoxic drugs develops more rapidly in case (iii) than in case (i). On the other hand, resistance to CS cytotoxic drugs and to cytostatic drugs is more sensitive to the drug application in case (i) that in case (iii). We finally comment that \( E[Q_i(t, \theta_i)] \) shows similar dynamics when using drugs with the same mechanism, that is, the results with drugs \( C_1, C_3, \) and \( C_5 \) are similar to \( C_2, C_4, \) and \( C_6 \), respectively.

Gardner (2002) [54] presents the effect of different drug combinations particularly to cancer cells with different proliferating proportions \( \delta(t) \). Figure 21 shows simulations of the total number of tumor cells \( \rho(t) \) with a highly proliferating index \( (\delta^* = 0.5) \) and a low proliferating index \( (\delta^* = 0.05) \). We demonstrate that the drug response function plays a key role in determining the tumor growth dynamics using certain combination therapies that often involve the nCS cytotoxic drugs (\( C_3 \) and \( C_4 \)). In general, the drug combinations that includes CS cytotoxic drugs (\( C_1 \) and \( C_2 \)) are more effective in highly proliferating tumors. In the discrete model (8), the drug combinations without the CS cytotoxic drugs show no difference. However, in the continuum model (1)–(2), the highly proliferating cancer cells show disadvantage under drug combinations without CS cytotoxic drugs, which reveals a possible internal dependency between the drugs.

We observe that the choice of continuum model is critical to the emerging

\[ E[Q_i(t, \theta_i)] \triangleq \int \theta_i \, Q_i(t, \theta_i) d\theta_i, \] where \( Q_i(t, \theta) = \frac{\int_{\Gamma^c_i} n_P(t, \theta) + n_Q(t, \theta) d\theta_i}{\rho(t)} \) and \( \theta^c_i \) is the vector of \( \theta \) except the \( i \)-th index \( \theta_i \) and \( \Gamma^c_i \) is its domain.
Figure 21: Total number of cells $\rho(t)$ using different drug combinations with either high or low proliferating index, that is, $\delta^* = 0.5$ or 0.05. The drug combination that includes CS cytotoxic drugs ($C_1$ and $C_2$) are more effective in highly proliferating cells. The drug effect of combinations without CS cytotoxic drugs is independent of the proliferating index in the discrete model (8). In contrast, highly proliferating cancer cells show certain disadvantages in the continuum model (1)–(2).

drug response. For an effective individually-tailored cancer modeling, these results stress the importance of identifying an appropriate model depending on the drug response of each individuals.

4. Conclusion

In this paper we propose a mathematical model for multidrug resistance, assuming a continuous resistance phenotype space. The multidrug resistance trait variable represents the level of resistance to various drugs including cell-cycle specific and nonspecific cytotoxic drugs, as well as cytostatic drugs. We classify the proliferation and drug uptake functions and identify the cases where the continuum model results in an intermediate maximal fitness resistance, i.e., the cases in which the continuum and discrete models are essentially different. Thus, by observing the proliferation and drug effects, we can predict when the continuum models are different than the corresponding discrete models. We study the effect of epimutation on the cytotoxic and cytostatic resistance traits.
In contrast to standard mutations that are associated with an early relapse, epimutations may either accelerate or delay the relapse time. We demonstrate such effects on different continuum models, initial preexisting resistance ratios, and types of drugs.

We use our approach to revising the works of Komarova and Wodarz (2005) [53] and the Gardner (2002) [54]. Following [53], we study the impact of the turnover rate on tumor growth and drug response. We verify the effectiveness of a combination therapy with multiple cytotoxic drugs in low turnover tumors and also in high turnover tumors with a drug uptake function of case (i) under high drug dosages. Increasing the cytotoxic drug dosage delays the relapse in tumor that the drug uptake follows case (iii), but not in low turnover tumor with case (i), thus in particular in such cases, the dosage should be carefully chosen. Moreover, the choice of a drug uptake function is shown to have a higher impact than the turnover rate under a logistic growth condition. These results provide new insights on the dynamics beyond what is accessible by (and in certain cases even contradictory to) the discrete-trait model of [53].

The second example we studied followed [54] by considering three different types of drugs: cell cycle specific and nonspecific cytotoxic drugs, and cytostatic drugs. We demonstrated that the size of the tumor is more sensitive to the drug dosage in the continuum models compared with the model of [54]. In addition, a drug combination without the cell cycle specific cytotoxic drug shows no disadvantage in highly proliferating tumors in the discrete model, which is not the case in the continuum models. We conclude that the dynamics of the cancer cell population including the time of relapse and the resistance profile significantly depends on the choice of (continuum) models, in addition to the turnover rate and the proliferation index. Thus, it is critical to select appropriate multidrug resistance models depending on the drug response of each individuals, to accomplish an effective individually-tailored cancer modeling framework and a corresponding optimal drug therapy.

Our future work includes deriving a continuum model from high-dimensional data that will be preprocessed with data analysis techniques. In addition, mod-
eling the dependency structure of multiple drugs and investigating its effect on the resistance dynamics is another challenging topic. Finally, due to its dimensionality, simulation of multidrug resistance model requires developing an efficient numerical method that balances computational cost and accuracy. This will be addressed with adaptive numerical methods that take advantage of the underlying low dimensional structure of the solution.

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Appendix A. Parameters of simulation

The parameters for the simulation in section 3.2 are taken from [54] as following.

- Maximum proliferation rate of highly proliferating cells is $\gamma = 1/30$, and for less proliferating cells, it is $\gamma = 1/50$. In addition, reduced proliferation due to resistance is assumed that the cell cycle is delayed by approximately 20 hours [82–85].

- Transfer rate from quiescent to proliferating cells: $p = 1/20 \text{day}^{-1}$ [85–87].

- Proliferating proportion: $0.05 \leq \delta^* \leq 0.5$ [82–85]. We take $\delta^* = 0.15$ unless otherwise stated.

- Necrosis rate of the quiescent cells: $D_Q = 1/100 \text{day}^{-1}$ [88].

- $c_i(t) = \left\{ \begin{array}{ll} \frac{\tilde{e}_i}{\lambda_i} \left(1 - e^{-\lambda_i t}\right) + c_i^{prev}, & t \leq d_i \\ \frac{\tilde{e}_i}{\lambda_i} e^{-\lambda_i t} \left(1 - e^{-\lambda_i t}\right) + c_i^{prev}, & t > d_i \end{array} \right.$, where $c_i^{prev}$ is the amount of drug built up from previous drug applications and the parameters for drug administration are as follows [89–91].
- Periods of drug administration: $\lambda_i \big|_{i=1}^4 = 21$, $\lambda_5 = 1$, $\lambda_6 = 7$.
- Duration of drug administration: $d_i \big|_{i=1}^4 = 0.1\text{h}$, $d_5 = 2\text{h}$, $d_6 = 1/3\text{h}$.

- Drug dosage scaled for $a_i = 2$ and $z_i = 1$: $\bar{c}_1 = 5$, $\bar{c}_2 = 0.005$, $\bar{c}_3 = 0.0009$, $\bar{c}_4 = 0.00012$, $\bar{c}_5 = 0.01$, $\bar{c}_6 = 0.01$.

  - Mutation rate: $w = 10^{-6}$ [20].

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