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for Theoretical Physics**

QUANTITATIVE LIFE SCIENCES SECTION

## **MATHEMATICAL MODELS OF PROLIFERATIVE CONTROL**

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# **Abstract**

During development, tissues and organs achieve a finite size that can overcome injuries, even in adulthood, by means of control strategies. On the other hand, when cancer arises, key feedback mechanisms that allow for control in normal tissues fail, leading to uncontrolled cell proliferation. In this dissertation, we reviewed the work of Lander et al.[1] on the effects of different feedback controls on a three stages cell lineage model of self-renewing tissue dynamics. Later, we proposed the same three stages cell lineage model as a minimal representation of tumor heterogeneity in the context of colorectal cancer (CRC); we found the optimal drug treatment that targets the specific cell types in the lineage and controls CRC cells' proliferation, this was done by using optimal control theory, where we set the controller as the death rates of the cell types.

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# 1

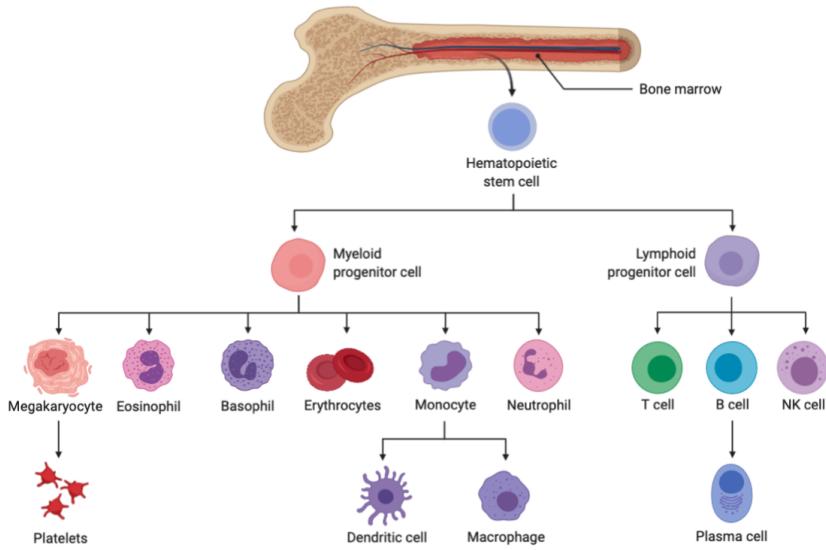
## Introduction

A cell lineage is the developmental history of a differentiated cell. In other words, it is a family tree that tracks the different stages in the cellular differentiation process. A cell division event can result in the birth of daughter cells equipped with transcriptional profiles different than those of the mother-cell and consequently performing different functions. Cell lineages can be a potential target of control, such that groups of cells, i.e. ideally tissues, can achieve disturbance rejection (catch-up growth), parametric robustness (variation to parameters, as for example final tissue size or robustness of the final state with respect to the initial conditions), stability and rapid regeneration, in order to attain an “optimal” biological behaviour of specific tissue .

An example of a cell lineage is represented in figure 1.1. In this picture the lineage starts with the Hematopoietic stem cell, a type of somatic stem cell that can differentiate into all types of blood cells. The multipotent hematopoietic stem cell can differentiate into the Myeloid and Lymphoid progenitor cells, which then give rise to other either terminally or non terminally differentiated cells (e.g. Monocyte, Megakaryocyte and B cell), where terminally differentiated cells represent the last stage in the lineage.

In some adult tissues and organs, there are adult stem cells (also called somatic stem cells) which can boost tissue self-renewal and are usually inactive until a damage of some kind in the tissue occurs. Upon injury somatic stem cells can restore a normal functioning tissue by activating cell proliferation. To achieve that goal, the somatic stem cells -through differentiation- give rise to a controlled number of terminally differentiated cells that build the tissue, maintaining tissue homeostasis.

Feedback interactions among cell types within a tissue in general are mediated by biochemical events typically relying on binding of a set of specialized molecules called



**Figure 1.1:** Bone marrow cell lineage. Figure taken from <https://blog.cellsignal.com/immunology-what-cells-have-a-myeloid-lineage-and-how-are-they-identified>

ligands to a corresponding set of cognate molecules called receptors, where ligands can either be soluble or membrane-bound. These processes can enable not only regeneration from an injury but also coordinated immune responses to recover from infections, thus maintaining tissue integrity. During cancer, key elements of this feedback fail, disrupting tissue function and immunity, and giving rise to unregulated cellular growth [2, 3].

The origin of cancer is the accumulation of genetic, epigenetic, and transcriptional alterations in a microevolutionary process that occurs over many years. Almost all tumors become very heterogeneous containing different types of proliferative and differentiated cells. This heterogeneity can be intratumoral or intertumoral (among tumors in different patients within a specific type of cancer), and may be responsible of tumor progression, relapse, metastasis and resistance to therapy [4].

Different models have been postulated in order to describe how tumor heterogeneity arises; such as clonal evolution, cancer stem cell (CSC), and phenotype plasticity models [5, 6].

The CSC or cancer-initiating cell (CIC) model postulates the existence of a differentiation hierarchy that starts with a cancer stem cell may be originating from normal stem cells or from progenitors. In this framework, CSCs are the only cells capable of extensive self-renewal, in contrast with non-CSCs transit-amplifying cells that have limited proliferative capacity and cannot regenerate cancer stem cells. Recent evidence challenges this constraint and suggests that differentiated cells can also rarely switch back to CSCs in a process called dedifferentiation [7]. The intimate internal architecture of

a tumor cell population might in principle have an impact on how tumors will react to therapy. For example in tumors that are fueled by CSCs, eliminating CSCs using targeted therapies will induce tumor regression, whereas in the clonal evolution model, which explains heterogeneity as a serial acquisition of mutations, removing the bulk of the tumor will reduce its progression. It is important however to notice that these two hierarchical models are not necessarily mutually exclusive.

CSCs, like normal tissue stem cells, are defined by their functional properties and are capable of long-term repopulation that enhances tumor progression; whereas more committed tumor cells, as transit-amplifying cells, contribute only transiently to tumor growth. Importantly, CSCs are thought to be more prone to the development of drug resistance, motivated in part by the intrinsic transcriptional profiles, by the limited rate of proliferation and by extrinsic regulation by the tumor microenvironment (e.g., hypoxia, inflammation, vascular niche, and cancer-associated fibroblasts) [4]. Having said all that, targeting CSCs is important to develop a rational approach to therapy.

A biological context where cell lineages and the role of stem cells is critical is the intestine. The inner lining of the intestine is known as a rapidly renewing tissue, which growth and homeostasis are promoted by a complex cell lineage led by intestinal stem cells (ISCs). The high turnover rate of the intestinal tissues (among other causes) results in possible malignant transformations and colorectal cancer (CRC) [8]. CRCs are thought to present heterogeneous cellular hierarchy organizations, reminiscent of a normal tissue homeostasis, with possible CSC at the top. Such cellular architecture, is believed to directly affect metastasis formation and therapy response.

In metastatic CRC, standard of care consists in the combination of chemotherapy with targeted therapies against specific molecular targets (VEGF and/or EGFR), with the exclusion of patients with tumors carrying RAS mutations where such treatments are not beneficial. For patients with wildtype RAS all first-line approved therapies are available and mostly beneficial except for the later emergence of relapse associated to poor prognosis [9]. Such relapse is associated to the so called minimal residual disease, i.e. a small cell fraction refractory to therapy that is left behind after the treatment and might fuel recurrence. The role of different cell lineages and/or cell types behind the response to therapy has not yet been investigated.

Motivated by these observations, in this work we investigate - from a theoretical point of view- how different therapeutic strategies can affect tumor progression in CRC; for instance, finding the optimal times at which drugs that target specific cell types, can be administrated. To do so, we use a variational approach to optimal control problems in order to find an optimal controller of cell proliferation that simulates the action of

drugs in a compartment toy model of population dynamics composed of three stages (cell types).

This document is organized as follows: in chapter 2, we review some basic concepts of control theory and introduce the necessary conditions for optimal control; later, in the same chapter, we reproduce some results obtained by Lander et al.[1] in the study of the dynamics of a three stages cell lineage and the effect of feedback in regulating cell proliferation upon injury. In chapter 3, we introduce the model to simulate the action of drugs in a minimal three stages cell lineage, as a representation of tumor growth control, we find the optimal controller that minimizes the side-effects of drugs and the total number of cells within the tumor. Finally, in chapter 4 we conclude and give some ideas to explore in our future work.

# 2

## Control theory in cell proliferation

### 2.1 Basics of control theory

In engineering, it is said that A controls B when it enables B to perform the desired task, to achieve some specific goals in the presence of uncertainties and disturbances. Hence, a system is under control if it provides the desired response/output by varying the input with some mechanism, the controller.

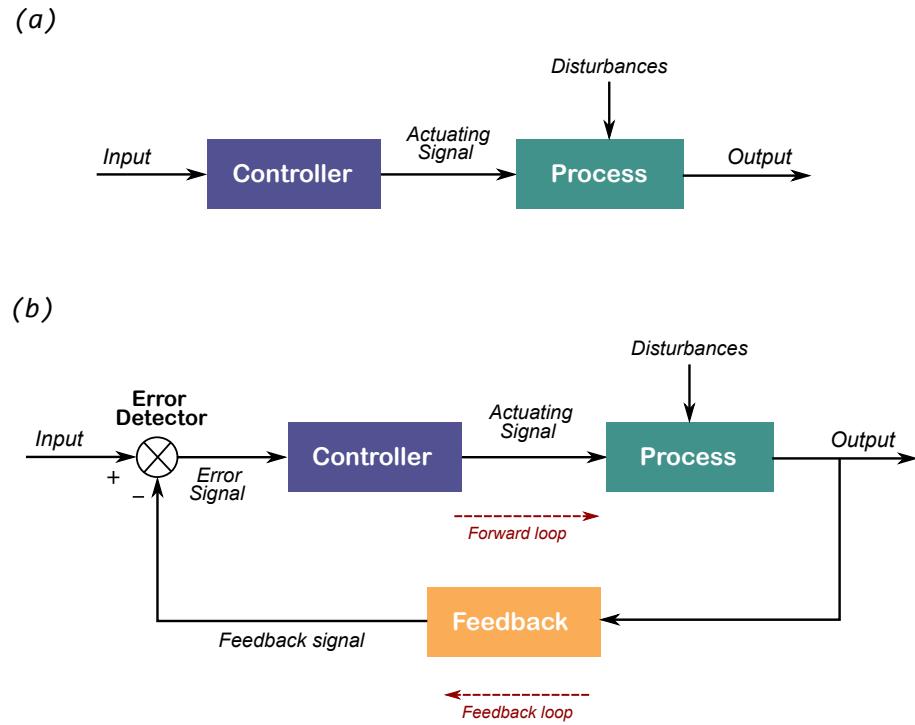
#### 2.1.1 Open-loop and closed-loop control systems

Based in some parameters, control systems can be classified as follows:

- **Type of the signal:** continuous time and discrete time control systems.
- **Number of inputs and outputs:** SISO Single Input and Single Output (SISO), and Multiple Inputs and Multiple Outputs (MIMO).
- **Feedback path:** open-loop and closed-loop control systems.

Figure 2.1 shows the two types of control systems based on the feedback path. In the **open-loop system**, subfigure (a), the input is applied to a controller which produces an actuating signal; this signal is then given as an input to the process or system (that we want to control) which then produces an output. A common example of this control system is a central heating boiler controlled only by a timer, here the process is the building temperature and the timer gives the signal of switching on/off the boiler in order to apply heat for a constant -previously determined- time which doesn't take into account the temperature of the building.

The disadvantages of open-loop systems are low accuracy and reliability, since in absence of a feedback mechanism any change in the output cannot be corrected auto-



**Figure 2.1:** Classification of control systems based on the feedback path. (a) An Open-loop system is one in which the actuating signal provided by the controller is independent of the desired output. (b) In a Closed-loop system, the output is fed back to the input. Thus, the actuating signal is dependent on the desired output. Figure adapted from [https://www.tutorialspoint.com/control\\_systems/control\\_systems\\_quick\\_guide.htm](https://www.tutorialspoint.com/control_systems/control_systems_quick_guide.htm).

matically, therefore it is sensitive to noise or unknown disturbances.

In a **closed-loop system**, figure 2.1 (a), its output is adjusted automatically until get the desired response. This is possible due to a mechanism that fed back the output to the input and an error detector produces an error signal (difference between actual and desired value of the process variable) that then is applied as feedback to generate an actuating signal by the controller to bring the system's output to a desired value, also called **set point**.

An advantage of these systems, compared with the open-loop ones, is that the controller has a way to measure the effects of unknown disturbances or noise in the output, leading to the ability of change its influences in the process by giving a correct actuating signal.

Using feedback control leads to increase robustness to perturbations, automatically reduce errors and improve stability. One way to explore the consequences of feedback and measure robustness is by calculating *sensitivity coefficients*, which are defined

as:

$$S = \frac{df}{d\theta} \frac{\theta}{f'} \quad (2.1)$$

here  $S$  is the sensitivity,  $f$  is a state variable (output of the system), and  $\theta$  is some parameter of the process. A system is said to be more robust when it achieves low values of sensitivity coefficients. For instance, a well-regulated system in biology means that its parameters' sensitivities are less than 1.

Despite the advantages of feedback, there are some situations in which its effectiveness is unsatisfactory. One of these is the presence of a large time delay within the process, and the other is the occurrence of very large disturbances. Since disturbances enter the process before the correction action of feedback can be done, large magnitudes of it may cause an unacceptable upset in a process.

There are two **types of feedback**: Positive and negative feedback. The first one tends to increase the magnitude of a small disturbance and accelerate the process, whereas the second reduces the error signal and tends to slow down the process.

### Controller types

All controllers have a specific use depending on which they are best suited. Two main types of them are continuous and discontinuous controllers. The basic types of continuous controllers are:

- **Proportional controllers:** give an output to the process that proportional to the error signal  $e(t)$ ,  $u(t) = K_P e(t)$ , where  $K_P$  is the proportional gain.

This type of controller works under small and smooth deviations between the input and output. Also, if the proportional gain is too high, the system can become unstable but if it is too small then the actuating signal may be too small when responding to system disturbances.

- **Derivative controllers:** are those whose output is a derivative of the error signal,  $u(t) = K_D e(t)$ , where  $K_D$  is the derivative gain.

This type of controller improves the transient response of the system. Its actuating signal predicts the system's behavior improving settling times and stability. One of its drawbacks is that they amplify the noise signals produced in the system.

- **Integral controllers:** give an output that is an integral of the error signal,  $u(t) = K_I \int_0^t e(\tau) d\tau$ , where  $K_I$  is the integral gain.

When there is a steady state then the steady-state error is always zero, thus these kinds of controllers are able to return the controlled variable back to the exact

set point [10]. However, they can make the system unstable for an oscillatory response.

Usually, these controllers are used together in different settings, in order to compensate for their disadvantages. Their common combinations are Proportional Derivative (PD), Integral Derivative (ID), and Proportional Integral Derivative (PID) controllers.

### 2.1.2 Deterministic optimal control

The goal of optimal control problems is to find a controller  $u(t)$  that minimizes a certain functional called cost or performance measure that is expressed as:

$$J = \phi(x(t_f), t_f) + \int_{t_0}^{t_f} C(x(\tau), u(\tau), \tau) d\tau, \quad (2.2)$$

where  $x(t) \in \mathbb{R}^n$ ,  $u(t) \in \mathbb{R}^m$ ,  $C : \mathbb{R}^n \times \mathbb{R}^m \times \mathbb{R} \rightarrow \mathbb{R}$  and  $\phi : \mathbb{R}^n \times \mathbb{R} \rightarrow \mathbb{R}$ . The function  $C$  is called *running cost* whereas  $\phi$  is the *terminal cost*. Here the final time  $t_f$  may not be fixed.

In general, the optimal controller  $u^*(t)$  causes the system

$$\dot{x} = f(x(t), u(t), t), \quad t_0 < t < t_f \quad (2.3)$$

to follow the optimal trajectory from an initial state  $x(t_0) = x_0$ , i.e., minimize the cost (2.2). Often the controller  $u(t)$  is constrained and to find it we must compute the functional derivative:

$$\frac{\delta J}{\delta u} = 0$$

But, this problem is difficult to solve since  $x$  depends implicitly on  $u$ . So, instead we can reformulate the problem as an constrained optimization problem by writing an augmented functional:

$$\tilde{J} = \phi(x(t_f), t_f) + p(t_0)^\top (x(t_0) - x_0) + \int_{t_0}^{t_f} \{C(x, u, \tau) + p(\tau)^\top [f(x, u, \tau) - \dot{x}]\} d\tau \quad (2.4)$$

and getting rid of the dependency of  $x$  on  $u$ . Here the components of  $p(\tau) \in \mathbb{R}^n$  are the Lagrange multipliers. With this we can therefore calculate the functional derivatives (2.1.2) and:

$$\frac{\delta \tilde{J}}{\delta u} = \frac{\delta \tilde{J}}{\delta x} = \frac{\delta \tilde{J}}{\delta p} = 0, \quad (2.5)$$

to obtain the necessary conditions for optimal control.

Let us consider the case of autonomous ordinary differential equations. Then, equation (2.4) becomes:

$$\tilde{J} = \phi(x(t_f)) + p(t_0)^\top (x_0 - x(t_0)) + \int_{t_0}^{t_f} \{C(x, u) + p(\tau)^\top [f(x, u) - \dot{x}]\} d\tau \quad (2.6)$$

and its differential (variation or first variation) is:

$$\begin{aligned} \delta\tilde{J} = & \frac{\partial\phi}{\partial x}\Big|_{x(t_f)} \delta x(t_f) - p(t_0)^\top \delta x(t_0) + \int_{t_0}^{t_f} \left[ \frac{\partial C}{\partial x} \delta x + \frac{\partial C}{\partial u} \delta u \right] d\tau + \\ & + \int_{t_0}^{t_f} p^\top(\tau) \left[ \frac{\partial f}{\partial x} \delta x + \frac{\partial f}{\partial u} \delta u - \frac{d}{dx}(\delta x) \right] d\tau + \\ & + \int_{t_0}^{t_f} [f(x, u) - \dot{x}]^\top \delta p d\tau \end{aligned} \quad (2.7)$$

where we have used the property  $\delta dy = d(\delta y)$ . Integrating by parts the term  $\frac{d}{dx}(\delta x)$  and regrouping terms, the variation  $\delta\tilde{J}$  is:

$$\begin{aligned} \delta\tilde{J} = & p(t_0)^\top \delta x(t_0) - p(t_f)^\top \delta x(t_f) - p(t_0)^\top \delta x(t_0) + \frac{\partial\phi}{\partial x}\Big|_{x(t_f)} \delta x(t_f) + \\ & + \int_{t_0}^{t_f} \left[ \frac{\partial C}{\partial x} + p^\top(\tau) \frac{\partial f}{\partial x} + \dot{p}(\tau)^\top \right] \delta x d\tau + \\ & + \int_{t_0}^{t_f} \left[ \frac{\partial C}{\partial u} + p^\top(\tau) \frac{\partial f}{\partial u} \right] \delta u d\tau + \\ & + \int_{t_0}^{t_f} [f(x, u) - \dot{x}]^\top \delta p d\tau \end{aligned} \quad (2.8)$$

Recalling that

$$F[f] = f(x_0), \quad x_0 \in (x_1, x_2) \rightarrow \quad F_{\hat{\delta}}[f] = \int_{x_1}^{x_2} \hat{\delta}(x - x_0) f(x) dx$$

and

$$\delta F[\rho; \eta] = \int \frac{\delta F}{\delta \rho}(x) \eta(x) dx$$

where  $\eta = \delta\rho$  and  $\hat{\delta}$  is the  $\delta$ -function; we can derive the expressions for equations (2.5):

$$\frac{\delta\tilde{J}}{\delta p} = f(x, u) - \dot{x} = 0 \quad (2.9)$$

$$\frac{\delta\tilde{J}}{\delta x} = \left[ \frac{\partial\phi}{\partial x} - p(t) \right] \hat{\delta}(t - t_f) + \frac{\partial C}{\partial x} + p(t)^\top \frac{\partial f}{\partial x} + \dot{p}(t) = 0 \quad (2.10)$$

$$\frac{\delta\tilde{J}}{\delta u} = \frac{\partial C}{\partial u} + p^\top(\tau) \frac{\partial f}{\partial u} = 0 \quad (2.11)$$

Defining the hamiltonian:

$$\mathcal{H} = C(x, u) + p(t)^\top f(x, u),$$

and considering that the terms with  $\delta$ -functions  $\hat{\delta}$  in (2.10) give the boundary conditions. Equations (2.9) to (2.11) would give us the **necessary conditions for optimal control** when the state is fixed at the initial time and free at the terminal time:

$$\dot{x}_i(t) = \frac{\partial \mathcal{H}}{\partial p_i} \quad (2.12a)$$

$$\dot{p}_i(t) = -\frac{\partial \mathcal{H}}{\partial x_i} \quad (2.12b)$$

$$0 = \frac{\partial \mathcal{H}}{\partial u_i} \quad (2.12c)$$

with boundary conditions:

$$x(t_0) = x_0 \quad (2.13a)$$

$$p_i(t_f) = \left. \frac{\partial \phi}{\partial x_i} \right|_{t=t_f} \quad (2.13b)$$

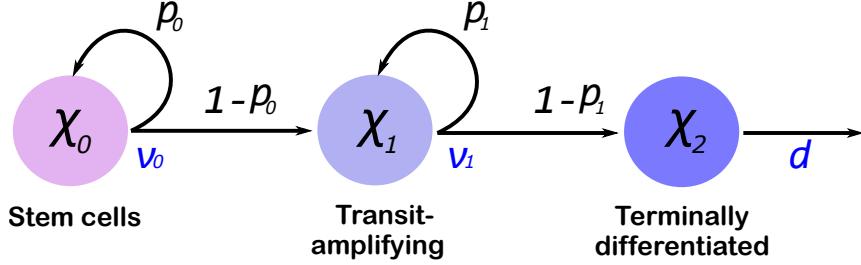
for  $i = 1, 2, \dots, n$ . The Hamilton's equations (2.12a) and (2.12b), along with the *transversality* conditions (2.13) constitute a two-point boundary value problem, where the state equation (2.12a) is solved forward in time while the costate equation (2.12b) is solved backwards. Finally, equation (2.12c) determines the optimal control  $u^*(t)$ .

## 2.2 Control in cell lineages

This section reviews the work of Lander et al. [1], which will be useful to understand some features of cell lineage dynamics and the biological implications of the assumptions over parameter values and feedback interactions.

### 2.2.1 Open loop three stages unbranched cell lineage

To understand how feedback controls tissue development and regeneration, let's start by studying an open-loop model of an unbranched cell lineage (see figure 2.2) with pools of three different types of cells: **stem cells**,  $\chi_0$ , which give rise to the so-called **transit-amplifying cells** (TA cells),  $\chi_1$ , that are more differentiated progenitor cells



**Figure 2.2:** Three stages cell lineage model for cell proliferation.  $p$ -parameters correspond to the replication probabilities,  $v$ -parameters are the cell cycle speeds and  $d$  is the death rate of Terminally Differentiated cells.

and which in turn differentiate into the **Terminally Differentiated cells** (TD cells),  $\chi_2$ , which typically characterizes the tissue and do not undergo self-renewal.

Figure 2.2 shows a graphical representation of the model in which stem and TA cells replicate with probability  $p_i$ , where  $i = 0, 1$  respectively.  $p_i = 1$  means that the cell is renewing itself all the time,  $p_i = 0$  means that every division gives rise only to daughter cells that are different, and  $p_i = 0.5$  means that on average every division gives rise to a differentiated cell and the other that undergoes renewal. Therefore,  $1 - p_i$  corresponds to the fraction of cells that differentiate into cells of the next stage. The rate parameters  $v$  are the corresponding cell cycle speed, which is defined as  $v = \ln(2)/\lambda$  where  $\lambda$  is the duration of the cell cycle, i.e., the time that it takes for a cell to grow and divide into two daughter cells. Finally, the parameter  $d$  is the death rate of the TD cells.

Then, if the abundance of cells at each stage in the lineage is large and their divisions are asynchronous, one can consider a continuous model in time and cell abundance, that can be described by the following system of ordinary differential equations (ODEs):

$$\begin{cases} \dot{\chi}_0(t) = (2p_0 - 1) v_0 \chi_0(t), \\ \dot{\chi}_1(t) = 2(1 - p_0) v_0 \chi_0(t) + (2p_1 - 1) v_1 \chi_1(t), \\ \dot{\chi}_2(t) = 2(1 - p_1) v_1 \chi_1(t) - d \chi_2(t), \end{cases} \quad (2.14)$$

here  $\dot{\chi}_i(t) = \frac{d}{dt}\chi_i(t)$  and the term  $(2p_i - 1)$  accounts for the fact that with probability  $p_i$  the cell divides into two daughter cells of the same type with the death of the parent. In the same way  $2(1 - p_i)$  refers to the probability that the daughter cells belong to the other type (differentiate).

Re-scaling the above system of equation by  $\tau = t v_1$ ,  $\xi = v_0/v_1$ ,  $\delta = d/v_1$ , we get:

$$\begin{cases} \dot{\chi}_0(\tau) = (2p_0 - 1) \xi \chi_0(\tau), \\ \dot{\chi}_1(\tau) = 2(1 - p_0) \xi \chi_0(\tau) + (2p_1 - 1) \chi_1(\tau), \\ \dot{\chi}_2(\tau) = 2(1 - p_1) \chi_1(\tau) - \delta \chi_2(\tau). \end{cases} \quad (2.15)$$

By doing a quick inspection of equations in (2.15) we can observe the following:

- If  $p_i > 0.5$  for any  $i$ , then  $\chi_2 \rightarrow \infty$ .
- If  $p_i < 0.5$  for all  $i$  and  $\delta = 0$ , then  $\chi_0$  and  $\chi_1$  will eventually run out, so no more cells are available to give rise to TD cells and the tissue will reach a *final state* with a fixed number of TD cells; otherwise, if  $\delta \neq 0$  terminally differentiated cells will also run out  $\chi_2 \rightarrow 0$ .
- If  $p_0 = 0.5$  and  $p_1 < 0.5$ :  $\chi_2 \rightarrow \infty$  when  $\delta = 0$ , and if TD cells die but not at an appreciable rate, i.e.,  $\delta \ll 1$ , the system will reach a *steady state* (see (2.16) and also the time-dependent solution with  $t \rightarrow \infty$ ).
- If  $p_0 < 0.5$  and  $p_1 = 0.5$  stems cells will run out and TA cells will achieve a constant abundance. When  $\delta = 0$  TD cells blow out; otherwise, the system will reach a *steady state* solution provided that the death rate is small. This is the case where TA cells behave as stem cells, which means that being "stem" is a functional property, also context dependent, rather than an intrinsic characteristic.

*Steady state solution:*

For the case in which none of the cell types runs out, i.e., when  $p_0 = 0.5$ ,  $p_1 < 0.5$  and small  $\delta$ , the steady state solution ( $\dot{\chi}_0 = \dot{\chi}_1 = \dot{\chi}_2 = 0$ ) is:

$$\chi_{1S} = \frac{\xi}{1 - 2p_1} \chi_0^*, \quad \chi_{2S} = \frac{2\xi}{\delta} \frac{1 - p_1}{1 - 2p_1} \chi_0^* \quad (2.16)$$

where  $\chi_0^*$  is the initial amount of stem cells. Equation (2.16) shows that the output of the system (number of Terminally Differentiated cells  $\chi_2$ ) is sensitive to the systems parameters  $d$ ,  $v_0$ ,  $p_1$  and the initial abundance of stem cells  $\chi_0^*$ . For instance, using equation (2.1) the sensitivity coefficients for the parameters  $v_0$  and  $p_1$  are:

$$S_{p_1} = \frac{p_1}{1 - 3p_1 + 2p_1^2}, \quad S_{v_0} = 1 \quad (2.17)$$

where  $S_{p_1} \geq 1$  for  $p_1 \in [1 - \frac{1}{\sqrt{2}}, 0.5]$ . Sensitivities greater or equal than 1 are undesirable in biological systems that are well-regulated meaning that the actual lineage dynamics need to be somehow under control strategies.

*Time-dependent solution:*

The general solution of the system of ODEs (2.16) with  $p_0 = 0.5$ ,  $p_1 < 0.5$ ,  $\delta \neq 0$  and initial conditions  $\chi_0^*, \chi_1^*, \chi_2^*$  is:

$$\begin{aligned}\chi_0(\tau) &= \chi_0^*, \\ \chi_1(\tau) &= \frac{\xi\chi_0^*}{1-2p_1}[1-e^{-(1-2p_1)t}] + \chi_1^*e^{-(1-2p_1)t}, \\ \chi_2(\tau) &= \frac{e^{-\delta t}}{\delta(2p_1-1)(2p_1-1+\delta)}\{\delta\chi_2^*[(2p_1-1)(2p_1-1+\delta)]+ \\ &\quad 2\xi\chi_0^*(p_1-1)[1-2p_1-\delta e^{(2p_1-1+\delta)t}+(2p_1-1+\delta)e^{\delta t}]- \\ &\quad 2\delta\chi_1^*[(p_1-1)(2p_1-1)(-1+e^{(2p_1-1+\delta)t})]\}.\end{aligned}$$

By taking the limit of  $t \rightarrow \infty$  we recover the results in (2.16).

*Disturbance in the dynamics:*

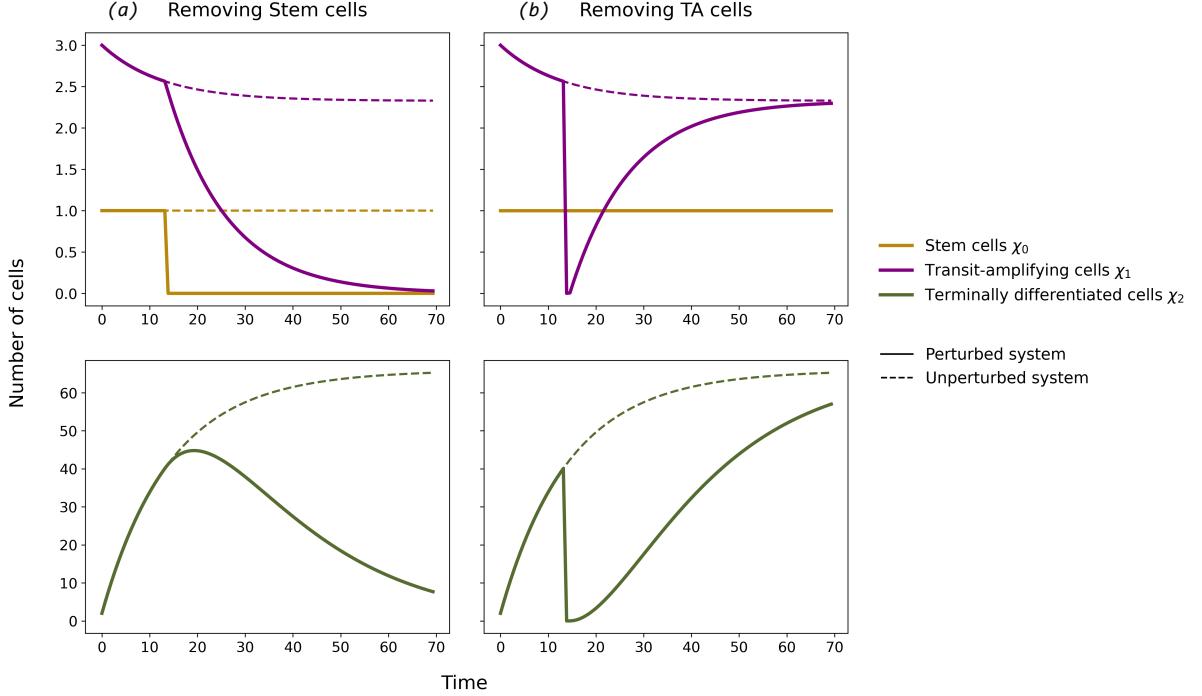
To study what would happen to the system's dynamics for the case of  $p_0 = 0.5$ ,  $p_1 < 0.5$  and  $\delta \neq 0$  when we remove stem or TA cells at some time  $t_p$ , we introduce a term  $-\nu I(t - t_p) \chi_i$  into the ODE of  $\chi_i$  for  $i = 0, 1$ , where  $I(t - t_p)$  is an impulse function that can be modeled numerically by defining a tent function, and  $\nu$  is the strength or rate at which cells are removed.

Figure 2.3 shows the results of this simulation with initial conditions  $\chi_0^* = 50$ ,  $\chi_1^* = 150$  and  $\chi_2^* = 100$ . Here we can note that an open-loop three stages system is not able to recover thus loosing all cell types if we remove completely the stem cells. On the contrary, if we target only TA cells, the system can catch up and achieve steady state, since TA cells are then regenerated by the differentiation of stem cells. This resembles to the behavior of CSCs when there is not dedifferentiation of early progenitors.

## 2.2.2 Feedback control in three stages unbranched cell lineage

In some tissues, control of proliferation and differentiation is mediated by feedback signals, mostly substances secreted by cells in the same tissue, as for example the negative growth regulators named *chalone*s [11] that can act at specific lineage stages. Many of those factors are members of the transforming growth factor beta (TGF $\beta$ ) superfamily; for instance, in the olfactory epithelium neural lineage, secreted molecules as GDF11 and Activin $\beta$ B slow the rates at which their target cells divide while increasing the differentiation probability [1, 12].

The open-loop three stages system studied before can achieve steady state even af-



**Figure 2.3:** Effect of a disturbance at a time  $t_p \approx 13.863$  on the dynamics of the three stages lineage in the case of stem cells replication probability  $p_0 = 0.5$  and system's parameters  $p_1 = 0.4725$ ,  $\xi = 0.128$  and  $\delta = 0.0372$ . The disturbance was introduced using a tent function to simulate a removal of cells at a time  $t_p$ . Two cases are shown: column (a) corresponds to a removal of only stem cells and column (b) only of TA cells. Number of cells are relative to the initial number of stem cells and time is expressed in units of  $\ln(2)/v_1$ .

ter complete removal of TD cells (initial condition for  $\chi_2$  equal to zero), as figure 2.4 (dashed lines) shows. In this simulated injury, initial conditions for  $\chi_0$  and  $\chi_0$  are the steady state values in (2.15). In this scenario, the regeneration time scale is similar to that over which TD cells turn over. Therefore, it is natural to wonder what is the best biological plausible control strategy to improve this regeneration speed?

#### Negative feedback on $v_1$ :

At first, it seems that one way to improve the regeneration speed would be having a feedback on the cell division rate of the TA cell,  $v_1$ . In biology, the feedback due to secreted growth factors is often introduced in terms of Hill functions [13], since these particular functions describe, at equilibrium, the relation of the ratio of bounded to total receptors as a function of the ligand concentration [14]. Thus, a monotonic decreasing hill function can represent negative feedback on the parameter  $v_1$ :

$$v_1 \rightarrow \frac{v_1}{1 + h\chi_2}$$

So, for large values of  $\chi_2$  the cell division rate would be very small such that it stops the production of TD cells quickly, achieving the steady state faster than without feed-

back.

The system of ODEs for  $p_0 = 0.5$  are now:

$$\begin{cases} \dot{\chi}_0(\tau) = 0 \rightarrow \dot{\chi}_0(\tau) = \chi_0^* \\ \dot{\chi}_1(\tau) = \xi \chi_0^* + (2p_1 - 1) \frac{1}{1+h\chi_2} \chi_1(\tau) \\ \dot{\chi}_2(\tau) = 2(1 - p_1) \frac{1}{1+h\chi_2} \chi_1(\tau) - \delta \chi_2(\tau) \end{cases} \quad (2.18)$$

and the corresponding steady state values are:

$$\chi_{1S} = \frac{\xi \chi_0^*}{1 - 2p_1} \left[ 1 + h \frac{2(1 - p_1) \xi \chi_0^*}{\delta(1 - 2p_1)} \right], \quad \chi_{2S} = \frac{2\xi}{\delta} \frac{1 - p_1}{1 - 2p_1} \chi_0^* \quad (2.19)$$

Figure 2.4 (a) shows the dynamics of the system following an injury (i.e. regeneration) where all TD cells have been removed at the same time, for the cases of absence of feedback and when its output  $\chi_2$  feeds back onto  $v_1$ . The parameters used correspond to the greatest improvement of regeneration speed (ratio between the regeneration times with and without feedback) and are consistent with the progenitor cells abundance -stem and TA cells- being no more than 50% of the tissue mass [1]. From the figure, it can be seen that the feedback on  $v_1$  enables the system to regenerate faster but at the expense of high values of TA cells  $\chi_1$  (almost equal to  $\chi_2$ ). This result however is undesirable since normally, as for example in the Olfactory Epithelium, the progenitor load is lower than 10% of the tissue mass.

### Negative feedback on $p_1$ :

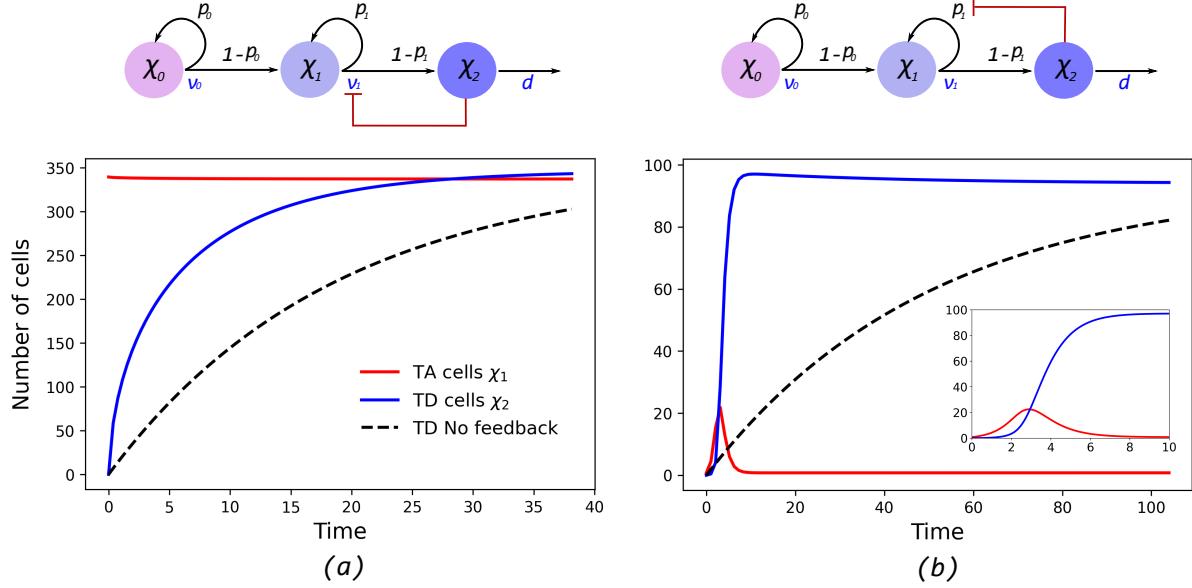
what would be the effect if the output feeds back only onto the replication probability  $p_1$ ? Using again the hill function:

$$p_1 \rightarrow \frac{p_1}{1 + g\chi_2}$$

the steady state solution is now:

$$\chi_{1S} = \frac{\delta}{2g} \left[ 2p_1 - 1 + \sqrt{(1 - 2p_1)^2 + 4\frac{g}{\delta} \left( \xi \chi_0^* + \frac{g}{\delta} \xi^2 \chi_0^{*2} \right)} \right], \quad \chi_{2S} = \frac{\chi_{1S} + \xi \chi_0^*}{\delta}. \quad (2.20)$$

where the second solution of the quadratic equation for  $\chi_{1S}$  has been neglected since it corresponds to a negative value due to the expression inside the radical that is always greater than  $2p_1 - 1$  with  $p_1 < 1$ . Analogously to the previous section, a regeneration simulation was performed using this new set of equations. Figure 2.4 (b) shows the results for this experiment in which the feedback now allows the progen-



**Figure 2.4:** Return to steady state of the system after removal of all Terminally Differentiated cells, without feedback (dashed lines) and with feedback on (a)  $v_1$  and (b)  $p_1$  (solid lines). Cell numbers are relative to the initial number of stem cells and time is expressed in units of  $\ln(2)/v_1$ . This corresponds to the initial conditions  $\chi_0 = 1$ ,  $\chi_1 = \chi_{1S}$  and  $\chi_2 = 0$ . Parameter values are  $p_0 = 0.5$ , (a)  $p_1 = 0.495$ ,  $\xi = 0.128$ ,  $\delta = 0.0372$ ,  $h = 0.0734$  and (b)  $p_1 = 0.942$ ,  $\xi = 0.506$ ,  $\delta = 0.0138$ ,  $g = 0.0449$ . The value of  $p_1$  for the case without feedback in (b) was obtained from equation (2.16) using (2.20).

itor load to be significantly lower than the 50% of the tissue mass and eliminate the constraint ( $p_1 < 0.5$ ) on TA cells' self-replication probability. In this setting, regeneration is characterized by a rise of TA cells proliferation (red curve) followed by a burst of TD cells production, achieving an explosive regeneration after a perturbation (blue curve); then as the number of TA cells decreases, the value of TD cells reaches its steady state.

One - among others [1] - tradeoff of the strategy of feeding back onto  $p_1$  is the loss of regeneration speed when the initial conditions change to a milder perturbation. For instance, figure 2.5(a) shows that having 75% of loss in TD cells implies a very long regeneration time, which is not a good strategy for an organism in the real world. Then, even if feeding back on  $p_1$  enhances the regeneration speed and the number of progenitors required for recovery, the behavior (in the absence of feedback, dashed lines) of faster tissue renewal when fewer cells are removed is lost.

#### Feedback on all the parameters:

As seen before, feeding back on  $p_1$  and  $v_1$  are not good strategies alone. Let's see what is the behavior of the system if the output feeds back on all the parameters.

Replacing the parameters with their correspondent Hill functions:

$$v_0 \rightarrow \frac{v_0}{1 + (j\chi_2)^{nj}}, \quad v_1 \rightarrow \frac{v_1}{1 + (h\chi_2)^{nh}}, \quad p_0 \rightarrow \frac{p_0}{1 + (k\chi_2)^{nk}}, \quad p_1 \rightarrow \frac{p_1}{1 + (g\chi_2)^{ng}},$$

the nondimensionalized system of ODEs becomes:

$$\begin{cases} \dot{\chi}_0(\tau) = (2\frac{p_0}{1+(k\chi_2)^{nk}} - 1) \frac{\xi}{1+(j\chi_2)^{nj}} \chi_0(\tau) \\ \dot{\chi}_1(\tau) = 2(1 - \frac{p_0}{1+(k\chi_2)^{nk}}) \frac{\xi}{1+(j\chi_2)^{nj}} \chi_0(\tau) + (2\frac{p_1}{1+(g\chi_2)^{ng}} - 1) \frac{1}{1+(h\chi_2)^{nh}} \chi_1(\tau) \\ \dot{\chi}_2(\tau) = 2(1 - \frac{p_1}{1+(g\chi_2)^{ng}}) \frac{1}{1+(h\chi_2)^{nh}} \chi_1(\tau) - \delta \chi_2(\tau) \end{cases} \quad (2.21)$$

In the case of  $nj = nh = nk = ng = 1$  and  $j, h, k, g \neq 0$  the Jacobian that is used for linearize the system of equations (2.21) to evaluate the stability of steady states is:

$$J = \begin{pmatrix} f_{00}(\chi_2) & 0 & \chi_0 f'_{00}(\chi_2) \\ f_{10}(\chi_2) & f_{11}(\chi_2) & \chi_0 f'_{10}(\chi_2) + \chi_1 f'_{11}(\chi_2) \\ 0 & f_{21}(\chi_2) & -\delta \end{pmatrix} \quad (2.22)$$

where:

$$f_{00}(\chi_2) = \left(2\frac{p_0}{1+k\chi_2} - 1\right) \frac{\xi}{1+j\chi_2}, \quad f_{10}(\chi_2) = 2\left(1 - \frac{p_0}{1+k\chi_2}\right) \frac{\xi}{1+j\chi_2},$$

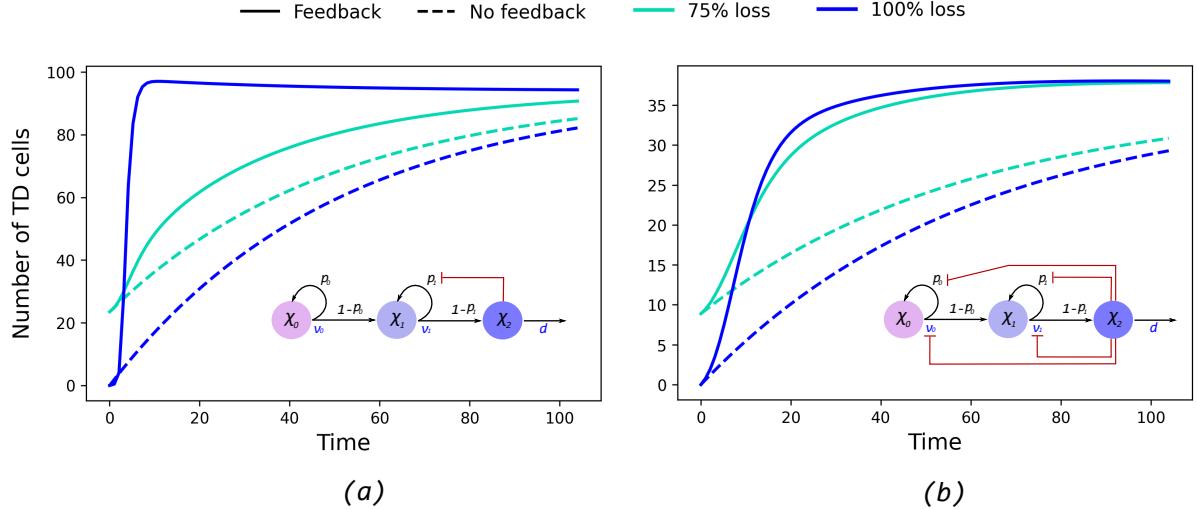
$$f_{11}(\chi_2) = \left(2\frac{p_1}{1+g\chi_2} - 1\right) \frac{1}{1+h\chi_2}, \quad f_{21}(\chi_2) = 2\left(1 - \frac{p_1}{1+g\chi_2}\right) \frac{1}{1+h\chi_2},$$

and the functions  $f'_{xy}$  their corresponding derivatives. Also, it is important to notice that Hill coefficients equal to 1 correspond to an amount of growth factor signaling, in stem and TA cells, proportional to the system's output (number of cells producing the growth factor). [1]

For this system there are three fixed points:

1. The trivial solution (reached for  $p_0 < 0.5$ ):  $\chi_{0S} = \chi_{1S} = \chi_{2S} = 0$ .
2. Stem cells are extinguished and TA cells behave as stem cells (reached for  $p_0 > 0.5$ ):

$$\chi_{0S} = 0, \quad \chi_{1S} = \frac{\delta(2p_1 - 1)(g - h + 2hp_1)}{g^2}, \quad \chi_{2S} = \frac{2p_1 - 1}{g}.$$



**Figure 2.5:** Return to steady state of Terminally Differentiated cells after 75% (cyan lines) and 100% (blue lines) of depletion, with feedback (solid lines) and without feedback (dashed lines) for two different feedback strategies. Initial conditions correspond to the steady state values  $\chi_{0S} \neq 0$  (1 in figure (a)),  $\chi_{1S}$  and 0.7 (or 1)  $\chi_{2S}$ . Parameters in both cases are: (a) same as in figure 2.4(b), and (b)  $p_0 = 0.507$ ,  $p_1 = 0.546$ ,  $\xi = 0.965$ ,  $\delta = 0.0116$ ,  $g = 1.258$ ,  $h = 1.03$ ,  $j = 1.683$ ,  $k = 0.0394$ .

3. All three populations are present (reached for  $p_0 > 0.5$ ):

$$\begin{cases} \chi_{0S} = \frac{\delta(2p_0-1)(k-j+2jp_0)(g-k-2gp_0+2kp_1)}{2k^2\xi(g-k-2gp_0+kp_1)}, \\ \chi_{1S} = \frac{\delta(2p_0-1)(-g+k+2gp_0)(-h+k+2hp_0)}{2k^2(-g+k+2gp_0-kp_1)}, \\ \chi_{2S} = \frac{2p_0-1}{k}. \end{cases} \quad (2.23)$$

Which one of the steady states is in fact reached is determined by the eigenvalues of the Jacobian (2.22) (see appendix A1.1).

In this case, it is possible for the system to achieve fast regeneration with low progenitor loads from different starting conditions. Figure 2.5(b) shows the improvement of regeneration speed in this situation, where the system reaches steady state as fast as a massive injury (100% loss) when there is 75% loss of TD cells.

# 3

## Optimal control of tumor growth

In this chapter, we apply the necessary conditions for optimal control to the minimal three stages cell lineage as a model of control of tumor growth in Colorectal cancer (CRC). This problem is aimed to investigate how an optimal drug therapy, that affects directly the death rate parameters of cancer cells, modify the proliferation of CRC cells.

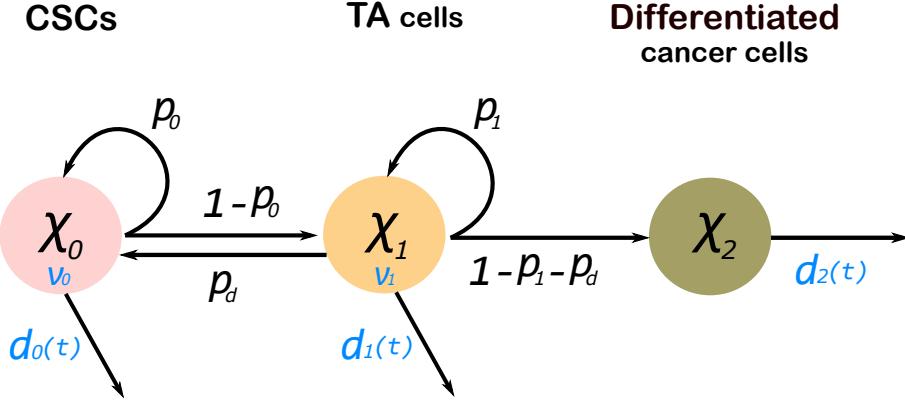
### 3.1 A dynamical model of tumor growth

Since CRC is thought to present heterogeneous cellular hierarchy organizations, a good starting point to model the problem of control of cancer cell population growth is the open-loop cell lineage system seen in chapter 2. For this problem, we introduce a dedifferentiation probability of TA cells into CSCs and time-dependent death rates. In this model, the death rates simulate the action of drugs that target specific types of tumor cells; therefore, they are the control variables in the dynamics that we want to find in order to obtain an optimal therapy strategy; i.e., the optimal times at which drugs can be administrated.

Figure 3.1 shows a cartoon of the model in which stem cancer cells  $\chi_0$  and TA cells  $\chi_1$  replicate with probability  $p_i$ ,  $i = 0, 1$ , and differentiate with probabilities  $1 - p_0$  and  $1 - p_1 - P_d$  respectively, where  $p_d$  is the dedifferentiation probability. Parameter  $d_i(t)$  is the death rate of cell type  $i$ .

The system of ODEs that describes the dynamics of the model in figure 3.1 is:

$$\begin{cases} \dot{\chi}_0(t) = (2p_0 - 1) v_0 \chi_0(t) + 2 v_1 p_d \chi_1(t) - d_0(t) \chi_0(t), \\ \dot{\chi}_1(t) = 2(1 - p_0) v_0 \chi_0(t) + (2p_1 - 1) v_1 \chi_1(t) - d_1(t) \chi_1(t), \\ \dot{\chi}_2(t) = 2(1 - p_1 - p_d) v_1 \chi_1(t) - d_2(t) \chi_2(t), \end{cases} \quad (3.1)$$



**Figure 3.1:** Three stages model for a colorectal cancer heterogeneity cell dynamics. Again,  $\chi_i$   $i = 0, 1, 2$  are the cell abundance of each cell type,  $v$ -parameters are the cell cycle speeds,  $p_j$  parameters with  $j = 0, 1$  correspond to the replication probabilities of CSCs and TA cells respectively, whereas  $p_d$  is the dedifferentiation probability. Finally,  $d$ -parameters are unknown time-dependent death rates that simulate the action of drugs administrated at certain times and target specific cell type populations.

Performing the same nondimensionalization as in section 2.2.1,  $\tau = t v_1$ ,  $\xi = v_0/v_1$  and  $u_i(\tau) = d_i(\tau)/v_1$  ( $i = 0, 1, 2$ ), we get:

$$\begin{cases} \dot{\chi}_0(\tau) = (2p_0 - 1) \xi \chi_0 + 2 p_d \chi_1 - u_0 \chi_0 = f_0, \\ \dot{\chi}_1(\tau) = 2(1 - p_0) \xi \chi_0 + (2p_1 - 1) \chi_1 - u_1 \chi_1 = f_1, \\ \dot{\chi}_2(\tau) = 2(1 - p_1 - p_d) \chi_1 - u_2 \chi_2 = f_2, \end{cases} \quad (3.2)$$

## 3.2 Optimal control of CRC cell proliferation

An optimal drug therapy strategy is the one that minimizes the tumor density or - as in our case- the total number of tumor cells and the drugs' side effects on the patient; for instance, giving high amounts of drugs can lead to poisoning and/or other complications. Therefore, in order to find the optimal controller  $u^* \in \mathbb{R}^3$ ,  $u^*(\tau) = (u_0^*(\tau), u_1^*(\tau), u_2^*(\tau))$  for this scenario, let's define the cost:

$$J = \int_0^T \left( \|x(\tau)\|_1 + \frac{\gamma}{2} \|u(\tau)\|_2^2 \right) d\tau, \quad (3.3)$$

where  $x(\tau) = (\chi_0(\tau), \chi_1(\tau), \chi_2(\tau))$ . Here the running cost is composed of the total number of cells and a quadratic controller which represents possible side effects. The goal is to minimize this performance objective, where the weight factor  $\gamma$  was introduced in order to give more importance into minimize the side-effects compared to the tumor size.

Using equation (3.3) the hamiltonian, defined in section 2.1.2, is:

$$\begin{aligned}\mathcal{H} &= \|x\|_1 + \frac{\gamma}{2} \|u\|_2^2 + \lambda^\top f(x, u) \\ &= \chi_0 + \chi_1 + \chi_2 + \frac{\gamma}{2}(u_0^2 + u_1^2 + u_2^2) + \lambda_0 f_0 + \lambda_1 f_1 + \lambda_2 f_2\end{aligned}\tag{3.4}$$

where  $\lambda$  is the vector of Lagrange multipliers. The quadratic controller written in this form implies that there are three different kinds of drugs that kill each type of cell independently with the same efficacy. From the necessary conditions for optimal control in the same section, we have:

$$\begin{cases} \dot{\lambda}_0(\tau) = -1 - [(2p_0 - 1)\xi - u_0]\lambda_0 - 2(1 - p_0)\xi\lambda_1, \\ \dot{\lambda}_1(\tau) = -1 - 2p_d\lambda_0 - (2p_1 - 1 - u_1)\lambda_1 - 2(1 - p_1 - p_d)\lambda_2, \\ \dot{\lambda}_2(\tau) = -1 + u_2\lambda_2, \end{cases}\tag{3.5}$$

$$u_i = \frac{\lambda_i \chi_i}{\gamma}, \quad (i = 0, 1, 2)\tag{3.6}$$

and the boundary conditions:

$$\lambda_i(T) = 0,\tag{3.7a}$$

$$\chi_i(0) = \chi_0^{(i)},\tag{3.7b}$$

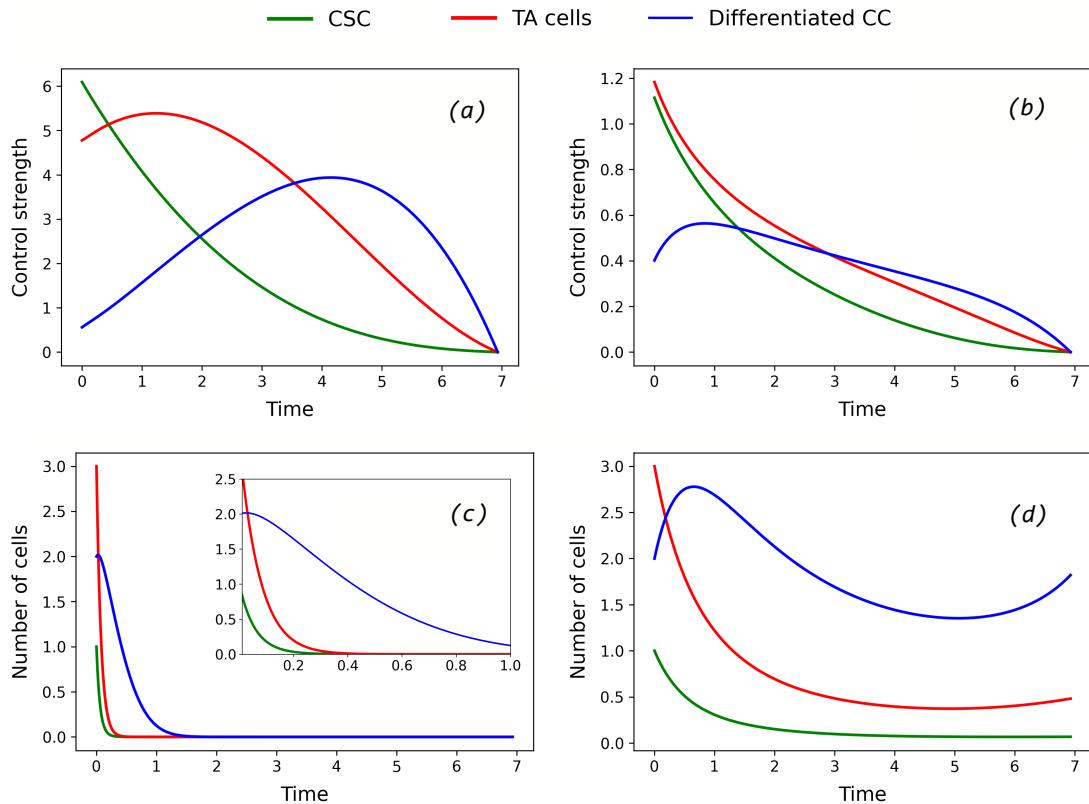
for  $i = 0, 1, 2$ . The optimal control law in (3.6) is also proportional to the Lagrange multipliers as in the linear-quadratic regulator (see appendix A1.2), but with the difference that in this case the controller is also proportional to the state values. Equations (3.2), (3.5), (3.6) and (3.7) form a nonlinear two-point boundary value problem that cannot be solved analytically. Therefore, we must use numerically iterative techniques as for example: steepest descent, variation of extremals, quasilinearization [15] or Forward-Backward Sweep Method [16] in order to solve it.

In this project we used the Forward-Backward Sweep Method to find the open-loop optimal control; i.e., the optimal control history associated with a set of initial conditions. This technique uses Runge-Kutta 4th order forward and backward methods to integrate the state and costate equations, (3.2) and (3.5), respectively, starting with an initial guess of the controller and then updating it using the control law (3.6) after the integration.

### Case of zero dedifferentiation probability

Figure 3.2 shows the first and tenth update of the controller  $u$  for the case of zero dedifferentiation probability ( $p_d = 0$ ) and parameters  $p_0 = p_1 = 0.51$ ,  $\xi = 1$  and  $\gamma = 10$ . In this case, since  $p_i$ , ( $i = 0, 1$ ) is greater than 0.5 the solution without control grows exponentially, as seen in section 2.2.1.

A parameter  $\gamma = 10$  was chosen in order to give more importance to the minimization of the controller  $u$  - amount of drugs - over the number of cancer cells. This also allows for the use of bigger step sizes in the Runge-Kutta integrator and still guaranteeing the convergence of the solutions (see appendix A1.3) since, for the normal integration with  $\gamma = 1$ , it will be needed smaller step sizes to not observe a very high increment in the exponential solution. A better study of this issue is pending, for instance using a different algorithm.



**Figure 3.2:** Control strength and number of cells for non-optimal control (a,c) and optimal control (b,d). The optimal control was achieved after 10 iterations whereas the non optimal at the first updating of the controller  $u$ . The parameters used in this simulation were:  $p_0 = p_1 = 0.51$ ,  $p_d = 0$ ,  $\xi = 1$ ,  $\gamma = 10$  and initial conditions  $\chi_0 = 1$ ,  $\chi_1 = 3$  and  $\chi_2 = 2$ . Cell numbers are relative to the initial number of CSC and time is expressed in units of  $\ln(2)/v_1$ .

Figure 3.2(a) shows that for the first update of  $u$ ; i.e., the non optimal controller, increasing the amount of drugs that kills differentiated cells until later times, compared

to TA cells, during treatment and then decreasing it leads to a faster decrease of cell numbers (see figure 3.2(c)). This is done at the cost of higher amount of drug (control strength) compared with the optimal control in figure 3.2(b).

Both cases, optimal and non-optimal controllers, agree in the strategy to reduce the amount of CSCs, namely, a high initial control strength followed by a gradual reduction of no drugs until the end of the treatment.

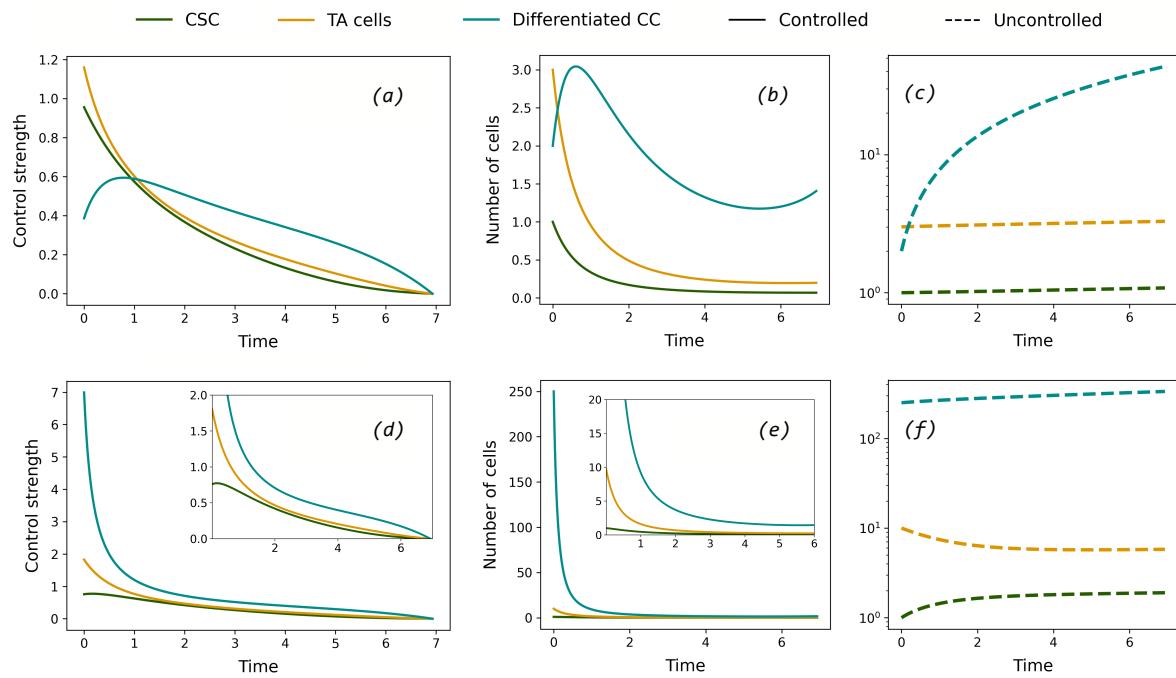
The optimal control for this choice of parameters and initial conditions was able to reduce the final number of CSCs, but for TA and differentiated cancer cells this strategy, with lowered control strength, allows for an increase of cell numbers after a time of  $5 \ln(2)/v_1$  (see blue and red curve in figure 3.2(d)).

### **Case of non zero dedifferentiation probability**

For the case when the dedifferentiation probability is  $p_d \neq 0$ , we obtained the results shown in figure 3.3, where the value of  $p_d$  is ten times smaller than  $p_1$  since this is a rare event. We used different values of  $p_0$  and  $p_1$  that are smaller than 0.5 and still have exponential solutions for cell types' population abundances at large times (see subplot c), a result that is different from what we saw in section 2.2.1.

For the same initial conditions used in figure 3.2; i.e.,  $\chi_0 = 1$ ,  $\chi_1 = 3$  and  $\chi_2 = 2$ , we got almost the same strategy as in the case when  $p_d \neq 0$ , in which the strength of the drugs that kill CSCs and TA cells is higher at the start of the treatment and decreases in time until the end of it. However, in this case, the decrease in control strength for TA cells is faster than the previous case when  $p_d \neq 0$ , this is due to the lower value of probabilities  $p_0$  and  $p_1$ . The control strategy for  $\chi_2$  remains the same. The convergence plots for this scenario can be seen in appendix A1.3 figures A1.1 (d-f). The optimal control obtained was able to not only reduce the final number of CSCs but also for TA cells. The number of differentiated cancer cells still increases cell numbers after a time of  $5 \ln(2)/v_1$ .

A slightly different situation occurs when the initial conditions change and the parameters remain the same. The optimal control strategy for initial values of cell numbers  $\chi_0 = 1$ ,  $\chi_1 = 10$  and  $\chi_2 = 250$  is one in which the best control strategy (see figure 3.3d) comprises of giving the highest amount of drugs at the beginning of the treatment following by a reduction of the doses - abrupt reduction for differentiated cells - of drugs until the end of it. This leads to a fast reduction of cell numbers for every type, which means that if the initial conditions are so that  $\chi_2 > \chi_1 > \chi_0$  then the optimal drug therapy strategy shown here will reduce effectively the tumor size.



**Figure 3.3:** Control strength and number of cells for the optimal control of model (3.2). The optimal control was achieved after 15 iterations. The parameters used in this simulation were:  $p_0 = 0.41$ ,  $p_1 = 0.31$ ,  $p_d = 0.031$ ,  $\xi = 1$ ,  $\gamma = 10$  with the initial conditions  $\chi_0 = 1$ ,  $\chi_1 = 3$  and  $\chi_2 = 2$  for figures a,b and c, and  $\chi_0 = 1$ ,  $\chi_1 = 10$  and  $\chi_2 = 250$  for figures d,e and f. Cell numbers are expressed relative to the initial number of CSC and time is expressed in units of  $\ln(2)/v_1$ . Figures c and d are the solutions of the state equations when there is no control; these semi-log plots show that the solutions have exponential behavior for times larger than  $4 \ln(2)/v_1$ .

# 4

## Conclusions

The concept of control is fundamental in engineering. It is used to describe how some processes or dynamical systems can be modified or influenced in order to achieve some specific goals that are important in different applications, ranging from the cruise control present in some automobiles to medicine, pharmaceutical, and food industry, where high-performance controllers have been implemented for dosage, where indicating specific amounts of drugs or products, plays an important role.

In control theory, feedback is an important feature that allows improving the performance of control systems in the presence of noise and disturbances. Biology is a perfect example of great control engineering, as feedback regulation is present in many biological systems, that range from molecules to cells to ecosystems. In this context, examples of feedback processes are gene regulation and glucose regulation in the bloodstream through the production of insulin and glucagon by the pancreas [17].

During development, organ and tissue achieve a finite size by means of feedback mechanisms; for instance, the secretion of specialized substances by cells, most of them members of the transforming growth factor beta (TGF $\beta$ ) superfamily, perform negative feedback control on specific cell types, inhibiting their proliferation by targeting some parameters involved in their growth dynamics as division or differentiation rates. On the other side, since cell lineages track the different stages in cellular differentiation, they can be used to model the different feedback interactions among cell types in the lineage (still present in adult tissues) that enable final size and tissue homeostasis.

In this work, we reviewed different feedback strategies in a three stages cell lineage dynamics (Stem, TA and TD cells) that can lead to achieve steady state upon injury, a characteristic of self renewing tissues. In this case, we have seen that following an

injury in which all TD cells have been removed, targeting the cell cycle rate of TA cells  $v_1$  is not a good strategy because even if it enables the system to regenerate faster, it does it at the cost of high values of TA cells, which is not desirable since normally progenitor load remains at lower values, typically below 10% of tissue mass.

When the feedback was performed only on the TA cells' replication probability  $p_1$ , the rapid regeneration was still present and the problem of high progenitor load went away, also allowing to eliminate the hard constraint of  $p_1 < 0.5$ , that was present in the open-loop system and in the previous feedback strategy. Despite this useful characteristic, feeding back on  $p_1$  only has a tradeoff that occurs for a milder perturbation; i.e., when removing lower amount of TD cells the regeneration time becomes longer, which is not what really happens in organisms in the real world. As last, feeding back on all parameters in the cell lineage model improved the failures of the previous strategies. It achieved fast regeneration with low progenitor loads for different initial conditions; i.e., different degrees of perturbation (massive and milder injury).

Unregulated cellular growth arises when key elements of feedback control in tissue fails. This unregulated process is also characteristic of cancer, which its origin is the accumulation of genetic, epigenetic and transcriptional alterations. Therefore, optimal control theory - with the method of calculus of variations - allowed us to find the optimal drug strategy for the control of unregulated proliferation of cancer cells.

Motivated by the evidence of heterogeneous cellular hierarchies in Colorectal Cancers (CRCs), with characteristics similar to the normal tissue; i.e. different type of cells can interact among them by feedback mechanisms in order to maintain their population, which is also believed to enable resistance to therapy; we modeled the growth cell dynamics using the open-loop three stages cell lineage and introducing death rate parameters that simulated the action of different drugs that target specifically each cell type in the lineage.

We studied two cases of this model, zero and non zero dedifferentiation probability  $p_d$ . We found that for the same initial conditions, and replication probabilities  $p_0$  and  $p_1$  that lead to an exponential solution in the model, the optimal control strategy is similar in both cases. This strategy consists in applying the highest dose of drugs that affect CSCs and TA cells at the beginning of the treatment and rapidly reducing the amount of it until a certain time,  $2\ln(2)/v_1$ , following a slow reduction until the end of the treatment. Whereas, to kill differentiated cancer cells the highest control strength is applied later in time but still near the beginning of the treatment, and then reducing it gradually with a different rate and profile compared to CSC and TA cells control. The result of this optimal therapy was to decrease the total number of CSC and TA cells at the end of the treatment while allowing a small increase in differentiated cells.

In both cases, a finite amount of TA and differentiated cells remained at the end of therapy.

When  $p_d \neq 0$ , a change in the initial conditions such that  $\chi_2 > \chi_1 > \chi_0$  led to an optimal control which applies the maximum strength at the very beginning of the treatment for all cell types. This enabled a very fast decrease in cell numbers at early times, keeping a small number of cells remaining.

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# A1

## Appendix

### A1.1 Stability of the ODE's steady state solutions with feedback on $v_0$ , $v_1$ , $p_0$ and $p_1$

The system of equations where all the Hill coefficients are equal to 1 is:

$$\begin{cases} \dot{\chi}_0(\tau) = f_{00}(\chi_2) \chi_0(\tau) \\ \dot{\chi}_1(\tau) = f_{10}(\chi_2) \chi_0(\tau) + f_{11}(\chi_2) \chi_1(\tau) \\ \dot{\chi}_2(\tau) = f_{21}(\chi_2) \chi_1(\tau) - \delta \chi_2(\tau). \end{cases} \quad (\text{A1.1})$$

where:

$$f_{00}(\chi_2) = \left(2 \frac{p_0}{1+k\chi_2} - 1\right) \frac{\xi}{1+j\chi_2}, \quad f_{10}(\chi_2) = 2 \left(1 - \frac{p_0}{1+k\chi_2}\right) \frac{\xi}{1+j\chi_2},$$

$$f_{11}(\chi_2) = \left(2 \frac{p_1}{1+g\chi_2} - 1\right) \frac{1}{1+h\chi_2}, \quad f_{21}(\chi_2) = 2 \left(1 - \frac{p_1}{1+g\chi_2}\right) \frac{1}{1+h\chi_2},$$

and

$$f'_{00}(\chi_2) = \frac{\xi [j - 2(j+k)p_0 + 2jk(1-2p_0)\chi_2 + jk^2\chi_2^2]}{(1+j\chi_2)^2(1+k\chi_2)^2},$$

$$f'_{10}(\chi_2) = \frac{2\xi [j(p_0-1) + kp_0 + 2jk(p_0-1)\chi_2 - jk^2\chi_2^2]}{(1+j\chi_2)^2(1+k\chi_2)^2},$$

$$f'_{11}(\chi_2) = \frac{h - 2(g+h)p_1 + gh\chi_2[2(1-2p_1) + g\chi_2]}{(1+g\chi_2)^2(1+h\chi_2)^2},$$

$$f'_{21}(\chi_2) = \frac{2\{gp_1(1+h\chi_2) + h(1+g\chi_2)[p_1 - (1+g\chi_2)]\}}{(1+g\chi_2)^2(1+h\chi_2)^2}.$$

To find the stability of each of the steady state solutions of (A1.1), we must find the eigenvalues of the ODE's Jacobian evaluated at the stationary points  $(\chi_{0S}, \chi_{1S}, \chi_{2S})$  (i.e., linearizing around the point):

$$J = \begin{pmatrix} f_{00}(\chi_2) & 0 & \chi_0 f'_{00}(\chi_2) \\ f_{10}(\chi_2) & f_{11}(\chi_2) & \chi_0 f'_{10}(\chi_2) + \chi_1 f'_{11}(\chi_2) \\ 0 & f_{21}(\chi_2) & -\delta \end{pmatrix} \quad (\text{A1.2})$$

**Stability of the trivial solution**  $\chi_0 = \chi_1 = \chi_2 = 0$ :

For the trivial solution the jacobian becomes:

$$J|_{(0,0,0)} = \begin{pmatrix} (2p_0 - 1)\xi & 0 & 0 \\ 2(1 - p_0)\xi & (2p_1 - 1) & 0 \\ 0 & 2(1 - p_1) & -\delta \end{pmatrix} \quad (\text{A1.3})$$

which its eigenvalues are:

$$\lambda_1 = (2p_0 - 1)\xi$$

$$\lambda_2 = (2p_1 - 1)$$

$$\lambda_3 = -\delta$$

This solution is locally stable if and only if  $p_0 < 0.5$  and  $p_1 < 0.5$ .

**Stability of the solution**  $\chi_0 = 0, \chi_1 = \chi_{1S}, \chi_2 = \chi_{2S}$ :

The jacobian for this solution is:

$$J|_{(0,\chi_{1S},\chi_{2S})} = \begin{pmatrix} f_{00}(\chi_{2S}) & 0 & 0 \\ f_{10}(\chi_{2S}) & f_{11}(\chi_{2S}) & \chi_{1S} f'_{11}(\chi_{2S}) \\ 0 & f_{21}(\chi_{2S}) & -\delta \end{pmatrix} \quad (\text{A1.4})$$

its eigenvalues are:

$$\lambda_1 = f_{00}(\chi_{2S})$$

$$\lambda_2 = \frac{1}{2} \left[ f_{11}(\chi_{2S}) - \delta - \sqrt{f_{11}^2(\chi_{2S}) + 2\delta f_{11}(\chi_{2S}) + \delta^2 + 4f_{21}(\chi_{2S})f'_{11}(\chi_{2S})\chi_{1S}} \right]$$

$$\lambda_3 = \frac{1}{2} \left[ f_{11}(\chi_{2S}) - \delta + \sqrt{f_{11}^2(\chi_{2S}) + 2\delta f_{11}(\chi_{2S}) + \delta^2 + 4f_{21}(\chi_{2S})f'_{11}(\chi_{2S})\chi_{1S}} \right]$$

where replacing the steady state solution  $\chi_{2S} = \frac{2p_1-1}{g}$  into the equations:  $f_{11}(\chi_{2S}) = 0$ , the first eigenvalue is negative if:

$$\frac{2p_1-1}{g} > \frac{2p_0-1}{k}, \quad (\text{A1.5})$$

and since  $p_0 > 0.5$ , then to meet this condition  $p_1$  should be greater than 0.5 as well.

The eigenvalues  $\lambda_2$  and  $\lambda_3$  can be written as:

$$\begin{aligned}\lambda_2 &= \frac{1}{2} \left[ -\delta - \sqrt{\delta^2 - \frac{2g(2p_1-1)}{p_1(g+h(2p_1-1))}\delta} \right] \\ \lambda_3 &= \frac{1}{2} \left[ -\delta + \sqrt{\delta^2 - \frac{2g(2p_1-1)}{p_1(g+h(2p_1-1))}\delta} \right]\end{aligned}$$

since  $p_1 > 0.5$  both results are negative if the expression under the square root is positive; otherwise, the solution oscillates towards the steady state value if (A1.5) is met.

Hence, to obtain the steady state for  $p_0 > 0.5$  and  $\chi_{0S} = 0$ ,  $\chi_{1S}$ ,  $\chi_{2S} \neq 0$ , the parameters of the model should meet condition (A1.5). Contrarily, the system will reach steady state number 3 (see main text) when  $p_0 > 0.5$ .

## A1.2 Linear-quadratic regulator

The steps shown here follow the derivations of Kirk [15].

Regulator problems are special case of tracking problems, in which the goal is to maintain the system  $x(t) \in \mathbb{R}^n$  close to the desired state  $r(t) \in \mathbb{R}^n$  in the interval  $[t_0, t_f]$  using a performance measure:

$$J = \frac{1}{2} \|x(t_f) - r(t_f)\|_2^2 + \int_{t_0}^{t_f} \|x(t) - r(t)\|_2^2 dt$$

The regulator case arises when the desired state values are zero ( $r(t) = 0$ ). When the dynamics of the system are described by a set of linear differential equations and the cost by a quadratic function is called the linear-quadratic regulator problem.

The system is described by a set of linear state equations:

$$\dot{x}(t) = A(t)x(t) + B(t)u(t), \quad x(t_0) = x_0, \quad (\text{A1.6})$$

where  $u(t) \in \mathbb{R}^m$  is the controller and  $A(t)$  and  $B(t)$  are matrices of sizes  $n \times n$  and  $n \times m$  respectively. The cost or performance measure to be minimized is:

$$J = \frac{1}{2}x^\top(t_f)Px(t_f) + \frac{1}{2} \int_{t_0}^{t_f} [x^\top(t)Qx(t) + u^\top(t)Ru(t)] dt, \quad (\text{A1.7})$$

here  $Q$  and  $P$  are real symmetric positive semi-definite matrices while  $R$  is a real symmetric positive definite matrix.

Using the necessary conditions for optimal control in section 2.1.2 where the states and controls are not bounded and  $X(t_f)$  is free. The hamiltonian is:

$$\mathcal{H}(x, u, p, t) = x^\top Qx + u^\top Ru + p^\top [Ax + Bu] \quad (\text{A1.8})$$

and the equations to solve are (A1.6) and:

$$\dot{p} = -Qx - A^\top p, \quad p(t_f) = Px(t_f) \quad (\text{A1.9})$$

$$u = -R^{-1}B^\top p. \quad (\text{A1.10})$$

Replacing (A1.10) in (A1.6), and joining the 2n linear ODEs:

$$\begin{pmatrix} \dot{x} \\ \dot{p} \end{pmatrix} = \begin{pmatrix} A & -BR^{-1}B^\top \\ -Q & -A^\top \end{pmatrix} \begin{pmatrix} x \\ p \end{pmatrix} \quad (\text{A1.11})$$

This system of equations can be represented as  $\dot{X} = \Lambda X$  and solved by taking the Laplace transform. So the solution is:

$$X(t_f) = \mathcal{L}^{-1}\{(sI - \Lambda)^{-1}\}X(t)$$

where  $s \in \mathbb{R}$  and  $\mathcal{L}^{-1}\{(sI - \Lambda)^{-1}\}$  is called the state-transition matrix  $\Phi(t_f, t)$ . Explicitly:

$$\begin{pmatrix} x(t_f) \\ p(t_f) \end{pmatrix} = \begin{pmatrix} \phi_{11}(t_f, t) & \phi_{12}(t_f, t) \\ \phi_{21}(t_f, t) & \phi_{22}(t_f, t) \end{pmatrix} \begin{pmatrix} x(t) \\ p(t) \end{pmatrix} \quad (\text{A1.12})$$

here  $\phi_{ij}$ ,  $i, j = 1, 2$  is a  $n \times n$  matrix. Using the boundary conditions of (A1.9) in (A1.12) we obtain:

$$x(t_f) = \phi_{11}(t_f, t)x(t) + \phi_{12}(t_f, t)p(t) \quad (\text{A1.13})$$

$$Px(t_f) = \phi_{21}(t_f, t)x(t) + \phi_{22}(t_f, t)p(t) \quad (\text{A1.14})$$

Substituting (A1.13) in (A1.14) and solving for  $p$ , yields

$$p(t) = K(t_f, t)x(t) \quad (\text{A1.15})$$

where  $K(t_f, t) = (\phi_{22} - P\phi_{12})^{-1}(\phi_{21} - P\phi_{11})$ . Substituting this result into equation (A1.10) the optimal control is:

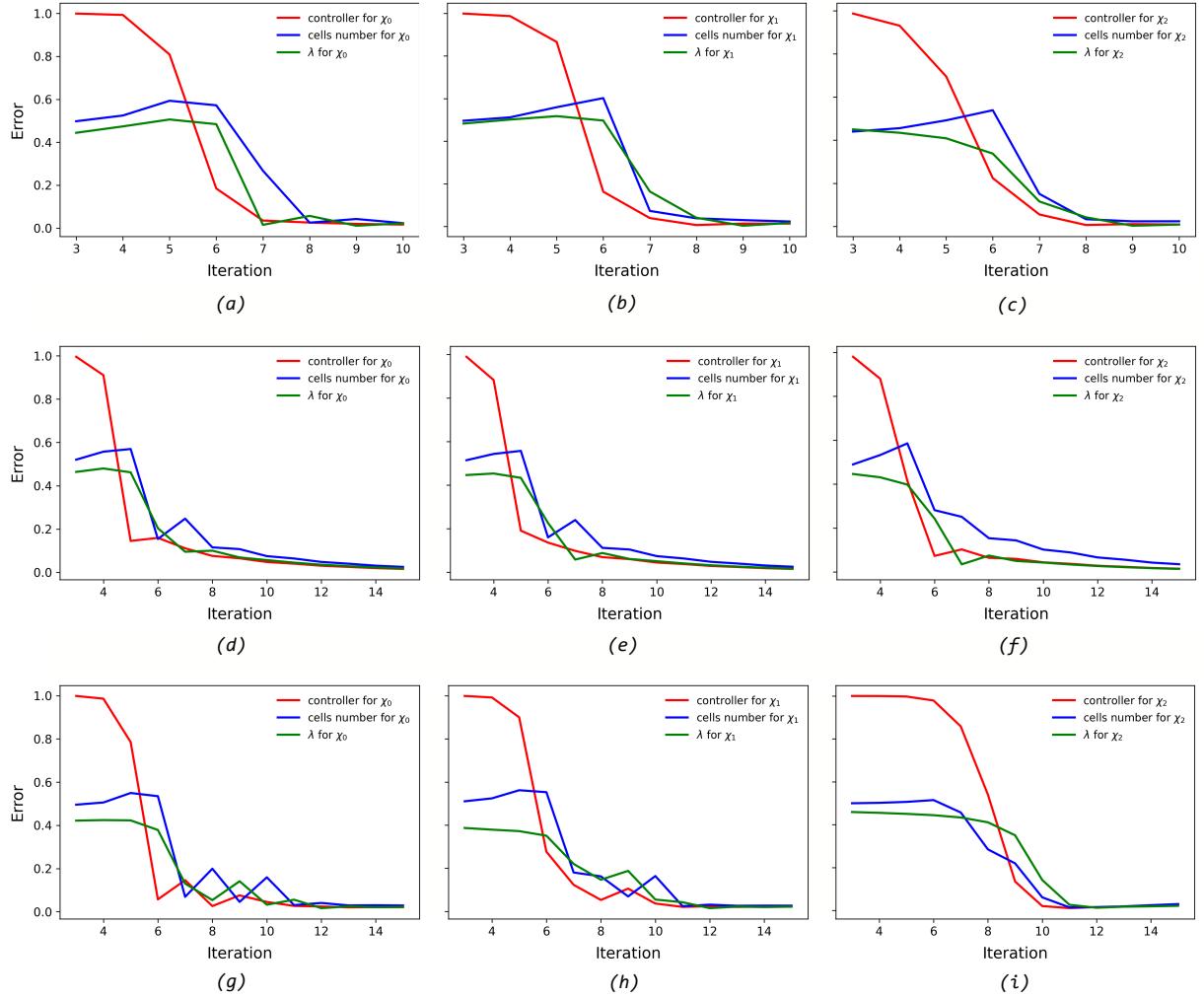
$$u^*(t) = -R^{-1}B^\top Kx(t) \quad (\text{A1.16})$$

An alternative approach to find  $K(t_f, t)$ , instead of computing the state-transition matrix, is to solve the *Riccati* matrix ODE:

$$\dot{K} = -Q - A^\top K - KA + KBR^{-1}B^\top K, \quad (\text{A1.17})$$

with the boundary condition  $K(t_f) = P$ . That comes from using (A1.15) and expanding  $\dot{p} = \dot{K}x + K\dot{x}$  in equation (A1.9), replacing (A1.6) and solving for  $\dot{K}$ . The *Riccati* matrix ODE can be solved numerically from  $t = t_f$  and proceeding backwards in time to  $t = t_0$ .

### A1.3 Error plots of the solutions for the optimal control of tumor growth



**Figure A1.1:** Convergence plots for the solutions in section 3.2. Red curves are the errors for the controller, blue curves for the cell abundance and green curves for the Lagrange multipliers  $\lambda$  of each cell type. Plots (a-c) correspond to the solution in figure 3.2 c and d. Plots (d-i) correspond to scenario of figure 3.3. Figures (d-f) are for the case of initial conditions  $\chi_0 = 1$ ,  $\chi_1 = 3$ , and  $\chi_2 = 2$ , and figures (g-i) for initial conditions  $\chi_0 = 1$ ,  $\chi_1 = 10$ , and  $\chi_2 = 250$ .