Mathematical models of budding yeast colony formation and damage segregation in stem cells

Dissertation

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Abstract

This dissertation consists of two chapters.

In Chapter 1, we present an individual-based model to study budding yeast colony development. Budding yeast, which undergoes polarized growth during budding and mating, has been a useful model system to study cell polarization. Bud sites are selected differently in haploid and diploid yeast cells: haploid cells bud in an axial manner, while diploid cells bud in a bipolar manner. While previous studies have been focused on the molecular details of the bud site selection and polarity establishment, not much is known about how different budding patterns give rise to different functions at the population level. In this chapter, we developed a two-dimensional agent-based model to study budding yeast colonies with cell-type specific biological processes, such as budding, mating, mating type switch, consumption of nutrients, and cell death. The model demonstrates that the axial budding pattern enhances mating probability at the early stage and the bipolar budding pattern improves colony development under nutrient limitation. Our results suggest that the frequency of mating type switch might control the trade-off between diploidization and inbreeding. The effect of cellular aging was also studied through our model. Based on the simulations, colonies initiated by an aged haploid cell show declined mating probability in the early stage and recover as the rejuvenated offsprings become the majority. It was shown that
colonies initiated by aged diploid cells do not show disadvantage in colony expansion due to the fact that young cells contribute the most to colony expansion.

In Chapter 2, we present a continuous population model of transport type to study stem cell aging. Stem cells are important for living creatures in that they generate all specialized tissues and give rise to the entire body at early life stage, and also serve as a repair system to replenish adult tissues while maintaining the stem cell pool for the lifetime of the living creature. Persistent division leads to stem cells suffering from the accumulation of molecular damages, which are commonly recognized as drivers of aging. As research on stem cells continues to advance, interesting questions arise as rapidly as new discoveries. How do stem cells respond to internal and external signals and regulate self-renewal and differentiation? How do stem cells cope with damage accumulation and maintain fitness? In this research, we propose a novel model to integrate stem cell proliferation and differentiation with damage accumulation in stem cell aging process. A system of two structured PDEs are used to model stem cells (including all multiple progenitors) and TD cells (terminally differentiated cells). It is assumed that cell cycle progression is continuous while division is discrete, and damage segregation takes place at division. Regulations from TD cells and stem cells population are incorporated through negative feedbacks on stem cell proliferation and symmetric division. Aging effect is added through the inhibition from damage accumulation on stem cell proliferation and self-renewal. Analysis and numerical simulations are conducted to study steady state populations and stem cell damage distributions under different damage segregation rules. Our simulations suggest that equal distribution of damage between stem cells in symmetric renewal and less damage retention in stem cell in asymmetric division are favorable rules, which
reduce the death rate of stem cells and increase TD cell populations. But asymmetric damage segregation in stem cells leads to less concentrated damage distribution in stem cells population, which may be more stable to sudden increase of damage. Compared to feedbacks solely from TD cells, adding feedbacks from stem cells will reduce oscillations and population overshoot in the process of population convergence to steady state. Moreover, adding the regulation that slows down the proliferation of stem cells with high level of damage and increases their tendency to differentiate can improve the fitness of stem cells by increasing the percentage of stem cells with less damage in the stem cell population.
Dedicated to my family.
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Chapter 1: Modeling Budding Yeast Colonies: an
Agent-based Approach

1.1 Introduction

Budding yeast *Saccharomyces cerevisiae* has been an ideal model system to study many biological processes crucial to the development of uni-cellular or multi-cellular organisms, such as cell polarization, cytokinesis and cell aging. It became a favorable model system because of its experimental tractability and the existing extensive studies over the decades. Yeast cells exist in haploid and diploid forms and they form colonies via sexual or asexual reproduction depending on the environmental cues [5]. Both haploid and diploid yeast cells can reproduce asexually by budding, in which a small bud emerges from the mother cell, enlarges until reaching a certain size, and then separates from the mother cell. The haploid cells have two mating types $a$ and $\alpha$, and they mate with their mating partners of the opposite mating type to form a diploid cell of type $a/\alpha$. Under extreme conditions such as stress or starvation, diploid cells can undergo sporulation, by entering meiosis and producing four haploid spores [5, 6]. The life cycle of budding yeast is illustrated in Fig. 1.1.

Yeast budding is an important process to understand cell polarization and symmetry breaking. Studies using both experimental or modeling approaches have been
extensively conducted on yeast budding [7, 8, 6, 9]. During budding, a new daughter cell emerges from a mother cell through polarized cell growth [6]. Haploid cells bud in an axial manner in which both mother and daughter cells have their bud sites adjacent to their previous division sites; diploid cells bud in a bipolar manner in which mother cells have a new bud site either adjacent or opposite to the previous division site, whereas daughter cells mostly choose a new bud site opposite to their birth site [5, 6, 10]. This budding event involves a key polarized protein, Cdc42 GTPase, which is highly conserved from yeast to human and plays a central role in polarity establishment [11, 12, 13]. Cdc42 localizes and interacts with other players in the pathway that ultimately lead to polarized growth and the emergence of a bud [6, 8]. This polarization is oriented by spatial cues that are distinct in each cell type [10]: proteins such as Bud3 and Bud4 are thought to function as a transient spatial cue.
in the axial budding pattern [14, 15], while Bud8 and Bud9 are the spatial cues in the bipolar budding pattern [16]. Previous efforts have been made to understand bud emergence at the molecular and mechanistic level; however, not much is known about why the haploid and diploid cells bud in different patterns. A long-standing speculation is that different budding patterns give rise to different biological functions specific to each cell type [8, 17, 7]: the axial budding pattern may facilitate mating by generating a tighter cluster of cells with opposite mating types, and the bipolar budding pattern may maximize the expansion of the colony, allowing a wider nutrient search in new territory.

An interesting feature of haploid budding yeasts is their ability to switch the mating type. Homothallic haploid yeast strains are able to switch between two mating types during mitotic growth [5, 18] and generate a colony that is a mixed population of both haploid and diploid cells. Mating type switch has an advantage of allowing haploid cells to change their mating type in daughter cells to generate a compatible mating partner, but it may come at a cost of forming diploid cells between closely related cells (mother-daughter or siblings), resulting in inbreeding which reduces genetic variation and fitness of offsprings [5]. How the cells balance these benefits and costs from mating type switch is yet unclear and could be related to the frequency of mating type switch [18, 5, 19, 20].

An unavoidable factor affecting all the processes discussed above, as well as almost all the other biological functions, is aging. Budding yeast renders itself as a useful tool to study the evolutionary conserved aspects of eukaryotic aging [21]. Individual yeast cells divide limited times before they die, and the number of cell divisions is defined as their replicative age [22]. It is known that certain cellular functions or processes
are associated with replicative age, for example, the mortality rate, cell cycle length, cell size and the sensitivity to environment (such as response to mating pheromone and nutrients) \[22\]. It was also observed that the cellular spatial order declines with replicative age, and interestingly, by tracking individual yeast cells, experiments showed that the probability of normal budding decreases with age \[2, 1\]. While it is still elusive whether the change of budding pattern is the cause or consequence of aging, a natural but unanswered question is how this loss of correct orientation in old cells impacts the colony at the population level.

Mathematical modeling has served as a useful tool to successfully address many important questions regarding cell polarization. Similar to the previous experimental works on yeast, most modeling works for budding yeast are on the molecular level to understand the pathways and mechanisms in cell polarization \[23, 24, 9\]. Modeling works that study yeast from the population point of view is very limited. In \[25\], an agent-based model was proposed to study the effects of different budding patterns and growth inhibition (induced by crowding effect) on colony morphology at the single-cell level. Their simulations demonstrated that growth inhibition and polar budding pattern are the most significant factors driving colony expansion. In \[26, 27\], agent-based models were proposed to simulate yeast colony growth, which includes a size-controlled module to govern cell proliferation and a cell-cell interaction module to arrange spatial positions of cells; the authors discussed the influence of cell-cell cohesion force and budding patterns on the colony shape and size \[26\], and they studied a variety of diameter growth time and reproduction time to better match the exponential growth in experiments \[27\]. However, the studies in \[25, 26, 27\] did not consider the intrinsic difference between budding patterns of haploid and diploid
cells, nor did they discuss how budding patterns and cell types affect the growth of colonies. In addition, the existing models did not include the interaction between cells and their living environment.

In this chapter, we present a novel and more comprehensive agent-based type model to study how the budding patterns in yeast cells affect colony growth. Our model incorporates many important biological processes in yeast cells, colony spatial arrangement through cell-cell mechanical iterations, and cell-environment interactions. To be more specific, the key biological processes include budding, mating, mating type switch, changes in cell cycle length and cell size due to aging, and cell death; cell-cell mechanical interaction is modeled through a contractive component due to cell adhesion and a repulsive component due to elastic compression [28, 29, 30, 31]; a nutrient field is introduced and the nutrient is consumed by cells while growth inhibition is induced if the nutrient level is too low. Our major findings include that (1) mating type switch frequency controls the trade-off between diploidization and inbreeding; (2) axial budding pattern in haploid yeast cells facilitates mating at an early stage of colony expansion; (3) bipolar budding is necessary for a branched colony under limited nutrient; (4) mating efficiency is lower in aged colonies but colony expansion does not depend on the overall age of the colonies. It is worth remarking that our modeling framework is not restricted to budding yeast and could be applied to study other systems, such as fungi, bacteria and stem cells. The chapter is structured as follows. A detailed description of the model is given in Section 2. In Sections 3 we present and analyze the results. Conclusions and discussions are given in Section 4. Supporting figures and texts can be found in Appendix A.
1.2 Model description

In nature, yeast cells bud and mate in a three-dimensional space and change their shapes during these processes. In our model, the cross sections of yeast cells on a two-dimensional domain are considered and their shapes remain spherical before and after budding and mating. This simplification would not significantly impact our conclusions since the focus of our study will be on the total population of yeast colonies and the overall spatial distribution.

Our model considers both haploid cells, which are of either $a$ or $\alpha$ mating type, and diploid cells, which are of $a/\alpha$ type. Each cell is viewed as a single agent and carries its own biological and physical information (summarized in Fig. 1.2). As time progresses with discrete time steps, these information will be updated with certain rules which will be further explained in details in the remainder of this section. During each time step, cells may experience budding, cell death, mating (haploid cells) or mating type switch (haploid cells); cell size and cell cycle length may change depending on the age of cells or other factors, and cells’ location may be rearranged due to budding or mating when the number of cells changes. In the extracellular space, there is a nutrient field which is initially set to be uniform and is updated at each time step due to the consumption by cells. In the meanwhile, cell cycle length may be prolonged by nutrient deficiency. The agent-based algorithm is summarized in a flow chart in Fig. 1.3.
Biological characteristics:
cell age
cell cycle length
cell size
cell budding angle
cell type and mating info

Physical quantities:
cell location
cell velocity
surrounding nutrient info

Cell-cell adhesive and replusive forces

$V_j$

Figure 1.2: A schematic of the agent-based model, with the key biological and physical quantities.

Cell death

The lifespans of yeast cells can be measured by either their replicative potential (replicative lifespan) or the maximal survival time of a non-dividing cells (chronological lifespan) [21, 22]. In the literature both lifespans are used to study different aspects of aging: the replicative lifespan is associated with the total number of cell division, and the chronological lifespan is related to the physical time. Only replicative lifespan is considered in this chapter because we are mainly interested in the budding event and budding patterns.

It is known that during budding, yeast cells undergo asymmetric division, in which mothers give rise to daughter cells with full lifespan capacity [22]. Therefore, in our model, upon cell division the age of the mother cell is increased by one, while the initial age of daughter cell is set to be zero. In experiments, the replicative lifespan is measured by counting the number of total bud scars [2] [1], and the average replicative age is approximately 30–50 cell divisions [1].
Figure 1.3: Overview of the processes within a single cell cycle. $P_d$ and $P_b$ are the probabilities of cell death and normal budding (axial for haploid cells and bipolar for diploid cells), respectively. $P_s$ is the frequency of mating type switch. $P_m$ is the frequency of successful matings. The simulation stops when the maximal time or the maximal population is attained.
The death probability, denoted by $P_d(a)$, represents the probability that a cell with age $a$ dies before reaching age $a + 1$. Although this important quantity is not directly observable, its companion, the survival fraction $F_s(a)$ of the population, can be measured in experiments. Since the survival fraction $F_s(a)$ can be viewed as the probability that cells survive up to an age greater than $a$, $F_s(a)$ and $P_d(a)$ are related by the following formula

$$F_s(a) = (1 - P_d(a))F_s(a - 1) \quad \text{for } a \geq 1,$$

with $F_s(0) = 1 - P_d(0)$. Thus

$$F_s(a) = \prod_{i=0}^{a} (1 - P_d(i)), \quad \text{for } a \geq 1.$$

As cells bud many times, their death probability becomes higher [22]. In our model the death probability $P_d(a)$ is assumed to take the following form

$$P_d(a) = 1 - e^{-k_1a}, \quad (1.1)$$

and thus the corresponding survival fraction is

$$F_s(a) = e^{-k_1 \sum_{i=0}^{a} i} = e^{-\frac{k_1(a+1)}{2}}, \quad (1.2)$$

which is a sigmoid function. This is consistent with the shape of survival curves measured in experiments, regardless of the cells’ genetic background [22].

Previously, Jazwinski and Wawryn [1] measured the survival fraction of a population of haploid yeast cells through different ages. Using the experimental data in [1], the value of $k_1$ is estimated to be 0.006 for haploid cells. As for diploid cells, there is no available data of survival fraction to our knowledge; however, it has been reported that diploid cells are longer-lived than haploid cells [32], so $k_1 = 0.004$ is chosen for
diploid cells. These estimated survival fractions are shown in Fig. A.1A in Appendix A.

**Budding patterns**

Yeast cells undergo polarized division by budding at specific sites determined by their cell types. Wild type haploid cells bud in an axial manner: mother cells form new buds adjacent to previous bud site and daughter cells bud next to their birth site. On the other hand, wild type diploid cells bud in a bipolar manner: mother cells can choose a new bud site either adjacent (proximal pole) or opposite to (distal pole) the previous bud site and daughter cells bud at the site opposite to the birth site (distal pole) [14, 15, 16]. The schematic diagram of bud sites is shown in Fig. 1.4.

Figure 1.4: Budding patterns for haploid and diploid cells. Haploid cells bud in an axial manner: both mother and daughter cells have bud sites adjacent to the previous division site. Diploid cells bud in a bipolar budding pattern: mother cells have a new bud site adjacent to their daughters or on the opposite end of the cell, whereas daughter cells mostly choose a new bud site on the opposite end of the cell.
Interestingly, as a cell ages, its budding pattern, a representation of the cellular spatial order, appears to be disrupted with a manifestation of budding randomly at a higher frequency [2, 1]. Based on single-cell observations in [2], which tracked the budding patterns throughout lifespans of cells, we assume the probability of random budding to be an increasing function of the replicative age $a$:

$$P_b(a) = 1 - e^{-k_2a},$$

where $k_2$ is estimated to be 0.024. The estimated probability of random budding for haploid cells is shown in Fig. A.1B in Appendix A. Due to the lack of experimental data, diploid cells are assumed to have the same random budding probability as haploid cells.

**Cell size and cell cycle length**

Morphological and physiological changes were observed during the aging process of yeast [21, 22]. For example, cell size and cell cycle length were shown to gradually increase with successive divisions [22]. It was shown in [4] that the average radius of diploid cells increases from 3.5$\mu$m to 5.5$\mu$m from birth to death. Cells of the first generation are usually small and require a long cell cycle to reach a critical size to bud. In our model, the radius of a newborn diploid cell is set to be 3.5$\mu$m and increases by $\frac{1}{a_d}$ at each division after its cell size reaches the critical size 4.375$\mu$m. Thus, the radius function for diploid cells can be formulated as

$$r_d(a) = \begin{cases} 
3.5 & \text{if } a = 0, \\
4.375 + a/a_d & \text{if } a > 0,
\end{cases}$$

where $a_d = 30$. Since the average cell size of diploid cells is approximately 1.25 times that of the haploid cells [3], the radius of a newborn haploid cell is assumed to be
2.8\mu m and increases by \frac{1}{a_h} at each division after its cell size reaches the critical size 3.5\mu m. Similarly, the radius function for haploid cells takes the form

\[ r_h(a) = \begin{cases} 2.8 & \text{if } a = 0, \\ 3.5 + a/a_h & \text{if } a > 0, \end{cases} \quad (1.5) \]

where \( a_h = 25 \). The comparison of changes in cell size with respect to age is shown in Fig. A.1C in Appendix A.

Cell cycle lengths of budding yeast cells have been shown to increase with age [22], with possibly the exception of newborn cells because they have a longer G1 phase before the initiation of budding. While cell cycle lengths vary from cell to cell and depend on the strain background and growth media, the model is simplified by ignoring the variations among the cells and the stochasticity due to other factors. In addition, the cell cycle lengths of haploid and diploid cells are assumed to be the same because no significant difference in average generation time has been observed in experiments [3]. Thus, for both types of cells, the cell cycle length \( \lambda(a) \) is modeled as

\[ \lambda(a) = \begin{cases} g_0 & \text{if } a = 0, \\ g_1 & \text{if } a > 0. \end{cases} \quad (1.6) \]

Based on the experimental data in [3, 4], the parameters are chosen as follows: \( g_0 = 101.25 \text{ mins} \), \( g_1 = 90 \text{ mins} \) and \( g_{11} = g_{12} = \frac{2}{3} \). The estimated curve for cell cycle length is shown in Fig. A.1D in Appendix A.

**Mating and mating type switch**

Mating is a process in which a haploid \( a \) cell and a haploid \( \alpha \) cell come into physical contact and once successful, these two cells fuse into a diploid \( a/\alpha \) cell. It is known that haploid cells of opposite mating types tend to mate to form a diploid cell [33]. Yeast cells select their mating partners and preferentially mate with the cell that
produces the highest level of pheromone \[34, 35\]. However, cells become less sensitive to mating pheromone and become sterile as they grow old. Experiments showed that the frequency of successful matings dropped significantly when one mating partner was relatively old \[35, 22\]. Interestingly, when cells of different ages mate, the replicative age of the zygote is set to be that of the older haploid cell, indicating that age is a dominant phenotype \[35\].

Within a colony whose haploid cells have one dominant mating type, the chances of forming a diploid cell can be enhanced by mating type switch, a process in which haploid \(a\) cells and \(\alpha\) cells switch their mating types. The ability to switch mating type is restricted to cells that have budded at least once \[18, 5\]. This process is regulated by the HO gene, which may be activated in mother haploid cells during the G1 phase \[20\]. Mating type switch is not a rare event: previous experiments suggested that mating type switch occurs with a high frequency, usually greater than 50\% \[5\]. Hence even if a colony starts with one single haploid cell, both \(a\) and \(\alpha\) cells will be present in that colony. The high frequency of mating type switch and the tendency of mating between haploid cells with opposite mating types result in the prevalence of diploid cells in a colony. On the other hand, the high frequency of mating type switch may also lead to inbreeding, which reduces genetic variation \[18, 19, 20\].

Based on biological observations, the following simplified rules are used during the mating process in our model (Fig. 1.5): (i) the frequency of successful mating, denoted by \(P_m\) if two haploid cells of opposite mating types are in direct contact, will drop as cells age:

\[
P_m(a) = 0.75 - \frac{a}{k_3},
\]  

(1.7)
where $k_3$ is estimated as 80; (ii) newborn cells are not allowed to switch mating types, while experienced cells have a constant mating type switch frequency $P_s$; (iii) cells will preferentially choose the youngest of neighbors of the opposite type to mate; (iv) a newly formed diploid cell has a circular shape and has the same age as the older haploid cell prior to mating, and its volume is the sum of two mated cells; (v) inbreeding is defined as mating between mother and daughter cells or among siblings.

Figure 1.5: Mating type switch follows certain rules: (1) only experienced cells can switch mating type; (2) mating type switch occurs during the late G1 phase and the switched cells come in pairs; (3) mating type switch occurs at a high frequency. Inbreeding is defined as mating between mother and daughter cells or among siblings.

**Nutrient field**

The growth of individual yeast cells and the expansion of the colony depend on nutrient supply. Decrease in nutrient concentration will slow down cell growth by prolonging cell cycle length [36, 37]. The level of nutrient also affects cell-cell adhesion
and cell-media adhesion because nutrient depletion may activate certain genes to express corresponding cell-wall proteins that are essential for cell filamentous growth [28].

In our model, a nutrient field $u$, as a function of space $\vec{x}$ and time $t$, is introduced across the domain. The change of nutrient concentration is due to diffusion of nutrient and consumption by live cells. The consumption rate is assumed to be highly localized around cells and decreases exponentially with the distance from cells. The dynamics of nutrient concentration is described by

$$\frac{\partial u}{\partial t} = D \Delta u - \sum_{k=1}^{N(t)} c_r e^{-\frac{|\vec{x} - \vec{x}_k|}{c_d}} u,$$

where $\vec{x}_k$ denotes the coordinates of the center for the $k$-th cell, $N(t)$ is the total number of cells at time $t$, $c_r$ is the consumption rate of a single cell, and $c_d$ controls the degree of local consumption. The diffusion coefficient $D$ is selected depending on the growth media: larger values of $D$ for more liquid media and smaller values for more solid media. The initial nutrient field is assumed to be homogeneous with a value $U_0$.

To account for the growth inhibition induced by nutrient depletion for each individual cell, the cell cycle lengths are assumed to depend on local nutrient concentration $u_{loc}$, and therefore $g_0$ and $g_1$ in Eq. (1.6) are replaced by $g_0 f(u_{loc})$ and $g_1 f(u_{loc})$, respectively, where $f$ is a decreasing function. It is reasonable to assume that $f$ is 1 when nutrient is rich, decreases slowly under nutrient consumption, and drops rapidly when the nutrient supply is very limited. Therefore in our model $f$ is defined by

$$f = \max\{f_1, 1\},$$

where

$$f_1(u_{loc}) = \begin{cases} c_u^1 - c_u^2 u_{loc} & \text{if } u_{thd} \leq u_{loc} \leq U_0, \\ c_u^3 - c_u^4 \log(u_{loc}) & \text{otherwise}, \end{cases}$$
where $c_1, c_2, c_3, c_4$ and $u_{thd}$ are constants.

In the numerical implementation, the average local nutrient concentration for a cell centered at $\vec{x}$ with radius $r$ can be approximated by

$$u_{loc}(\vec{x}) \approx \frac{1}{n} \sum u_m (1 - H(|\vec{x}_m - \vec{x}|/r - d_{nur})),$$

where $u_m$ is the nutrient concentration at a grid point $\vec{x}_m$, $d_{nur}$ is the range that a cell can sense nutrient, $n$ is the total number of $m$ such that $|\vec{x}_m - \vec{x}|/r \leq d_{nur}$, and $H$ denotes the Heaviside function.

As a cell grows older its response to environment becomes less sensitive. To model this effect, the nutrient sensing distance, $d_{nur}$, is assumed as a decreasing function of age of the following form

$$d_{nur}(a) = \frac{d_{nur}^0}{1 + a/k_4},$$

where the constants $d_{nur}^0$ and $k_4$ are taken as 2 and 45, respectively. The choice of these parameters are random due to lack of experimental data.

**Cell spatial arrangement**

The spatial distribution of a colony of cells depends on the response of cells to forces exerted by their neighboring cells. For example, yeast cells in physical contact can form adhesive bonds, which result in adhesive force, by certain proteins located on the surface of cell walls [28, 29]. Yeast cells can also resist the compression by other cells due to the incompressibility of their cell wall [30, 31].

Many models are proposed to model cell-cell adhesive and repulsive forces [25, 38, 39, 40]. The repulsive force is mainly designed to avoid overlap between two agents in the model. Experiments have shown that that linear elastic constitutive equation can be used to describe cell wall material of a yeast cell [30, 31]. In this chapter, forces
between cells are modeled by linear contractile-repulsive springs as in [25]. Consider the \(i\)-th cell centered at \(\vec{x}_i\). The repulsive force between the \(i\)-th cell and the \(j\)-th cell centered at \(\vec{x}_j\) is assumed to increase with the overlap \(\Delta d = (r_i + r_j) - d_{ij}\), where \(d_{ij} = |\vec{x}_i - \vec{x}_j|\) and \(r_i, r_j\) are the corresponding radii of the cells. Then this repulsive force is given by

\[
\vec{F}_{r_{i,j}} = \begin{cases} 
  k_r \Delta d \frac{\vec{x}_i - \vec{x}_j}{d_{ij}} & \text{if } d_{ij} < r_i + r_j, \\
  0 & \text{otherwise},
\end{cases}
\]

where \(k_r\) is a spring stiffness constant. The adhesive force is assumed to be proportional to the overlap between a cell and its neighbors, and is defined by

\[
\vec{F}_{a_{i,j}} = \begin{cases} 
  -k_a \Delta d \frac{\vec{x}_i - \vec{x}_j}{d_{ij}} & \text{if } d_{ij} < r_i + r_j, \\
  0 & \text{otherwise},
\end{cases}
\]

where \(k_a\) is a spring stiffness constant.

Thus the overall force exerted on the \(i\)-th cell centered at \(\vec{x}_i\) is given by

\[
\vec{F}_i = \sum_{j=1,j\neq i}^{N(t)} \left( \vec{F}_{r_{i,j}} + \vec{F}_{a_{i,j}} \right).
\]

According to the Newton’s second law, the acceleration \(\vec{a}_i\) is proportional to force \(\vec{F}_i\). Assuming that initial velocity of the cell is 0, the instantaneous velocity is \(\vec{V}_i = \vec{a}_i \Delta t\) and

\[
\vec{V}_i = \alpha \vec{F}_i \Delta t,
\]

where \(\alpha\) is taken to be constant for simplicity. Thus the current position \(\vec{x}_i^{n+1}\) of the \(i\)-th cell can be approximated by

\[
\vec{x}_i^{n+1} = \vec{x}_i^n - \vec{V}_i \Delta t \quad \text{for } n \geq 0,
\]

where \(\vec{x}_i^n\) denotes the previous position of the \(i\)-th cell, and the time step \(\Delta t\) is chosen as 1.8 mins in our simulations, which is sufficiently small compared to cell cycle length.
1.3 Results

All the simulations presented in this section were based on model assumptions in Section 1.2 unless otherwise stated. The simulations were based on the algorithm presented in A.5 in Appendix A, and the values of all the parameters can be found in Table A.1 in Appendix A.

1.3.1 Mating type switch frequency controls the trade-off between diploidization and inbreeding

Mating type switch allows haploid cells to divide and change their mating type to generate a compatible mating partner. A single homothallic haploid cell will generate a colony with a mixed population, which contains both diploid $a/\alpha$ cells and haploid cells of $a$ and $\alpha$ types. Diploidization is advantageous because diploid cells are better than haploid cells at coping with DNA damage. However, mating type switch is likely to come with a cost [19]. For example, mating type switch may cause replicative delays, and the presence of switching mechanisms increases DNA replication errors. In addition, the formation of diploid $a/\alpha$ cells from closely related cells (mother-daughter or siblings) results in inbreeding and reduces the genetic variation, which is the primary selective force. While many homothallic strains of yeast cells switch their mating type at a very high frequency (about 70% of the total cell divisions), it is not understood why the switch does not happen at an even higher frequency, such as 100% [18, 5, 20].

To study the benefit and cost of mating type switch in budding yeast colonies, we examined two corresponding indicators: the percentage of diploid cells and the percentage of inbreeding among all mated pairs. In our simulations, three frequencies,
50%, 70% and 90%, of mating type switch were tested based on a simple setting: a colony starts with a single haploid cell, and this cell will bud and its offsprings are likely to switch their mating types, which eventually leads to mixed types of cells (a sample colony is shown in Fig. A.2 in Appendix A). 1000 samples were simulated for each mating type switch frequency and the statistics are summarized in Fig. 1.6. It can be seen that, in wild type axial budding haploid cells, higher mating switch frequency leads to higher percentage of diploid cells (Fig. 1.6A left panel) but meanwhile also leads to higher percentage of inbreeding (Fig. 1.6A right panel). Fig. 1.6B shows that this observation does not depend on the budding pattern of these haploid cells, and the same conclusion also holds for random budding haploid cells (new buds emerging at random positions). These results reveal the trade-off between diploidization and inbreeding controlled by the frequency of mating type switch, and may explain why the mating type switch frequency for wild type cells is approximately 70% instead of 100%. In the remainder of this chapter, the mating type switch frequency is set to be 70%, unless otherwise specified.

1.3.2 Axial budding pattern in haploid yeast cells facilitates mating

Different budding patterns are thought to contribute to different biological functions specific to each cell type [8, 17, 7]. Some researchers believe that axial budding pattern helps generate a tighter cluster of cells and facilitates the mating of haploid cells of opposite mating types to form diploid cells. Using our model, this hypothesis was tested by comparing colonies with axial budding haploid cells (new buds emerging adjacent to the previous bud site) and colonies with random budding haploid
Figure 1.6: Effect of the mating type switch frequency on diploid cell percentage and inbreeding percentage. Each bar represents the average value (± standard deviation).
cells (new buds emerging at random positions). The time of first mating and the percentage of diploid cells in a colony were used as a quantitative measure to assess the mating efficiency.

In 1000 simulations performed for each budding pattern, we found that although the total populations for both budding patterns show exponential growth, these two budding patterns lead to significantly different mating features. On average, the first mating happens earlier in the axial budding colonies as shown in Table 1.1. Moreover, the axial budding colonies show significantly higher percentage of diploid cells than the random budding ones (Fig. 1.7A). At 12 hours, over 70% of the population are diploid cells in the axial budding colonies, while the percentage for the random budding colonies is only 50%. These results support that axial budding pattern facilitates mating, especially at an early stage of colony growth.

<table>
<thead>
<tr>
<th>Budding pattern</th>
<th>mean</th>
<th>median</th>
<th>min</th>
<th>max</th>
<th>standard derivation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Axial</td>
<td>3.6</td>
<td>4.1376</td>
<td>3.3</td>
<td>10.2</td>
<td>1.126</td>
</tr>
<tr>
<td>Random</td>
<td>5.1</td>
<td>4.8867</td>
<td>3.3</td>
<td>10.5</td>
<td>1.753</td>
</tr>
</tbody>
</table>

Table 1.1: The time of first mating.

Another interesting and unexpected result from the simulations is that axial budding colonies show significantly lower percentage of inbreeding during the early stage of colony development, compared to random budding colonies with the same mating type switch frequency (Fig. 1.7B). At 4.5 hours, among all pairs of mated cells, about 65% are mother-daughter or siblings in random budding colonies, and this percentage
Figure 1.7: Comparisons of colonies with axial and random haploid budding patterns. (A) Percentage of diploid cells as time evolves. (B) Percentage of imbreeding calculated from the same simulations in (A).

is only around 45 for axial budding colonies. However, this difference becomes smaller as colonies grow.

1.3.3 Bipolar budding is necessary for a branched colony under limited nutrient

It was hypothesized that bipolar budding is important for maximizing the spread of a colony to reach out for nutrient in new territory [17, 7, 8]. Diploid cells require the BUD8 protein for bipolar budding: bud8Δ mutants do not bud in a bipolar manner but instead bud adjacent to their birth scars in a pattern similar to haploid axial budding [10]. By comparing wild type cells to bud8Δ mutants, it was shown that bipolar budding is necessary for colony spread and agar invasion [11].

To understand the relationship between budding patterns in diploid cells and the spread of colonies, colonies with bipolar and random budding diploid cells were studied via our model. In order to assess the differences of these colonies, two indicators
that measure the overall spread were introduced. The first indicator, called the colony radius and denoted by $R$, is the radius of the minimal covering circle of the colony (see Fig. A.3 in Appendix A for illustration). Larger colony radius implies a wider spread and higher efficiency in nutrient search. The second indicator, called the colony sparseness and denoted by $\sigma_D$, is defined as the ratio between the area of the minimal covering circle and the total actual area of the colony:

$$\sigma_D = \frac{\pi R^2}{\text{area of the colony}}.$$ 

Larger colony sparseness implies sparser distribution of cells inside the minimal covering circle and less competition for nutrient from neighboring cells. In the simulations, these two types of colonies start with four diploid cells, and will contain only diploid cells because sporulation is not considered in this chapter. For each of the situations under different budding patterns and initial nutrient settings, 1000 samples were simulated and data are recorded when colonies grow to 25, 50, 75, 100, 125 and 150 cells (shown in Fig. 1.8).

When the initial nutrient is abundant (by setting $U_0 = 2$), for both bipolar and random budding colonies, the colony radius increases on average as colonies grow. The bipolar budding colonies have, on average, slightly larger colony radius and sparseness than the random budding ones (Fig. 1.8). However, these advantages for the search of nutrient is gradually lost as colonies grow, since only little statistical difference was detected in these indicators for colonies consisting of 150 cells. Our simulations also agree with the modeling results in [25], which suggested that simply switching budding pattern from non-polar to polar does not necessarily lead to significant increase in colony size. Since only little advantage of the bipolar budding pattern was observed
under rich nutrient condition, we tested a decreased initial nutrient level $U_0 = 1$, which represents a poor nutrient condition.

Fig. 1.9A shows that for bipolar budding colonies, when the initial nutrient level $U_0$ decreases, both the colony radius and sparseness increase on average. With rich initial nutrient $U_0 = 2$, the colony radius on average increases about 150% as the population grows from 25 cells to 150 cells, while the colony sparseness remains almost constant; on the other hand, when the initial nutrient level is lowered to $U_0 = 1$, the increase in colony radius is over 200%, and the colony sparseness also increases more than 30%. These observations support that the bipolar budding pattern enhances colony development through better nutrient search. Another noticeable observation is that the curves of colony radius and sparseness has shifted significantly as $U_0$ decreased from 1.1 to 1, suggesting the possible existence of certain threshold of nutrient, below which the bipolar budding is far more advantageous.
Figure 1.9: Colony radius and sparseness in bipolar and random budding colonies as population grows. In these simulations, we set the initial nutrient level $U_0 = 1, 1.1, 1.25$ or 2.
Figure 1.10: Sample simulations of colonies with 150 cells under different nutrient levels. (A) Bipolar budding under rich nutrient $U_0 = 2$. (B) Bipolar budding under limited nutrient $U_0 = 1$. (C) Random budding under rich nutrient $U_0 = 2$. (D) Random budding under limited nutrient $U_0 = 1$.

However, the situation is different for the random budding colonies. As shown in Fig. 1.9B, nutrient limitation does not cause an active spread of the colony and there is little increase in colony radius and sparseness with various $U_0$. Previously, Jönsson and Levchenko [25] found that the colony area increased by 20% when neighbor inhibition varied from weak to strong, even when all divisions were non-polar. This difference between our results and theirs in [25] may be explained by the different approaches used to model the growth inhibition: in [25] the growth inhibition was modeled to be proportional to the number of neighboring cells, while in our model the growth of the cells is inhibited as their surrounding nutrient is consumed by themselves and neighboring cells. This growth inhibition due to nutrient consumption
is negligible under rich nutrient condition but will be more pronounced when the overall nutrient is limited.

Besides the measures of colonies, we are also interested in the actual colony morphology. Under rich nutrient condition $U_0 = 2$, colonies of both budding patterns exhibit approximately round shape except that the peripheries of the bipolar budding colonies are not as smooth and have lightly irregular extensions (Fig. 1.10A and Fig. 1.10C). Under poor nutrient condition $U_0 = 1$, bipolar budding colonies have a tight core and finger-like branches to reach out for nutrient (Fig. 1.10B), while random budding colonies are still relatively compact with emerging small branches at the periphery (Fig. 1.10D). More samples of colony morphology are shown in Fig. A.4 in Appendix A. Our simulations suggest that only bipolar budding cells give rise to a colony morphology with finger-like extensions under limited nutrient. Our results are consistent with the previous experiments of the lab strain S288C, which showed smooth colony structure and was specifically selected to be non-flocculent with a minimal set of nutritional requirements [41, 42].

1.3.4 Mating efficiency is lower in aged colonies but colony expansion does not depend on the overall age of the colony

It is known that cellular functions decline as a cell ages. As consequences or causes, increased cell size, cell cycle length and death probability, disruption of regular budding pattern, as well as lower sensitivity to mating pheromone are manifestations of aging. Experiments have shown that daughter cells from older mothers have shorter lifespan [22]. However, granddaughters of an old mother cell show a gradual
restoration to a normal lifespan, suggesting that some aging factors might be diluted to recover rejuvenation \[22\].

To accurately reflect the effect of the ages of mother cells as observed in biological experiments, we included a variable \(a\) representing age in some probabilities or parameters introduced in Section \[1.2\] (i) Daughter cells born from older mothers have higher death probability and are more likely to bud randomly. Accordingly Eqs. \[1.1\] and \[1.3\] are modified to:

\[
P_d(a, a_M) = 1 - e^{-k_1(1+a_M/c_1)a} \quad \text{and} \quad P_b(a, a_M) = 1 - e^{-k_2(1+a_M/c_2)a},
\]

where \(a_M\) is the age of the mother cell. (ii) The initial size of a daughter cell increases with the age of its mother: for diploid daughter cells, Eq. \[1.4\] is changed to \(3.5(1 + a_M/c_3)\mu m\), and for haploid daughters, Eq. \[1.5\] is modified as \(2.8(1 + a_M/c_4)\mu m\). The initial cell cycle length decreases with the age of its mother and \(g_0\) in Eq. \[1.6\] is modified to be \(g_0/(1 + a_M/c_5)\). (iii) The frequency of successful matings of haploid cells is influenced by the ages of their mother cells, so that Eq. \[1.7\] is modified to

\[
P_m(a, a_M) = 0.75 - \left(\frac{a}{k_3} + \frac{a_M}{c_6}\right).
\]

Here \(c_1, c_2, \ldots, c_6\) are scaling parameters.

First, we tested how aging affects yeast colony by comparing two colonies: colonies initiated by a single haploid cell of age 0 (referred to as young colonies), and colonies initiated by a single haploid cell of age 30 (referred to as old colonies). Within each of these two colonies, both axial budding and random budding patterns are considered. It can be seen in Fig. \[1.11A\] that young colonies always have significantly larger populations than old colonies regardless of the budding patterns of the haploid cells. This result is expected, as the death probability increases with replicative age and the
offsprings of old mothers need a few generations to fully rejuvenate. Fig. 1.11B shows that mating occurs later in old colonies compared to young colonies: at 4.5 hours, the diploid cell percentage of old colonies is almost zero, while that of young colonies is over 20% on average. Fig. 1.11B also shows that the old colonies tend to have a lower percentage of diploid cells, indicating lower mating efficiency in old colonies. Interestingly, although young colonies generally have higher mating efficiency, axial budding in haploid cells still shows its advantage: as time approaches 10 hours, the diploid cell percentage in an old colony with axial budding haploid cells (blue bars in Fig. 1.11B) exceeds that in a young colony with random budding haploid cells (green bars in Fig. 1.11B).

Young and old colonies initiated by four diploid cells of age 0 and of age 30 were also studied under limited nutrient. The first observation is that old colonies need a
longer time to reach a certain population size, since offsprings of old mothers need several generations to rejuvenate. Fig. 1.12A shows that if diploid cells bud in a bipolar manner, old colonies exhibit slightly larger colony radius and sparseness on average than young colonies; however, greater variance and more outliers are observed in old colonies. This may be explained by that most of the cells in a colony are young cells, and colony expansion is due to cell divisions at the periphery, which is also occupied by young cells (old cells reside in the core of the colony). On the other hand, if diploid cells bud in a random manner, almost no difference was observed between young and old colonies (Fig. 1.12B).

In summary, our results suggest that the age of the colony determines the mating efficiency and how quickly the cell population increases, but does not affect the overall spatial distribution of cells.

1.4 Conclusions and discussions

In this chapter, a two-dimensional agent-based model was developed to study budding yeast colonies with cell-type specific biological processes. Our model considers processes such as budding, mating cell death, consumption of nutrient and mating type switch. We investigated the roles of budding patterns, mating type switch frequency and growth inhibition induced by nutrient depletion in yeast colony development. Our findings reveal that axial budding pattern enhances mating efficiency at an early stage of colony development, and bipolar budding pattern improves colony expansion under nutrient limitation. Our results also suggest that mating type switch frequency might control the tradeoff between efficient diploidization and inbreeding.
Figure 1.12: Comparison of colony radius and sparseness between old and young colonies with different budding patterns under limited nutrient $U_0 = 1$. 
The effect of cellular aging was also studied. Based on the simulations, colonies initiated by an aged haploid cell show declined mating probability in an early stage of colony development but later recover as the rejuvenated offsprings become the majority. It was also shown that colonies initiated by aged diploid cells do not show disadvantage in colony expansion due to the fact that young cells contribute the most to colony expansion.

Our model can be extended to take into account intracellular signaling pathways and cellular responses. For simplicity and due to the lack of sufficient information, our model focused on a set of conceptual agent-based rules based on statistical results of experimental observations. However, one may also include the Cdc42 pathway or the cell cycle pathway for each individual cell to achieve a more realistic model. Another possible direction to extend our model is to include more detailed morphological changes induced by mating pheromone or nutrient depletion. In the current model, mating is only allowed when two cells of opposite types have direct contact, while in reality, cells may be able to sense mating pheromone over a longer distance and make projection toward the mating partners. A more realistic field of mating pheromone described by reaction-diffusion equations as in [43] could be included. Elongated cellular morphology due to nutrient depletion may also be considered. The incorporation of cell morphological change may need to shift the current simple computational approach to a more involved numerical method, such as the level set or phase field methods [44], and the computational cost will increase drastically. Therefore, a more feasible first step would be using the current framework but making the directional growth possible as in [45], without considering the detailed cell shape change.
One major limitation of the agent-based approach is that the cell population size is restricted to a relatively small scale due to a high computational cost. For a relatively large population, it is often beneficial to consider on-lattice agent modeling or continuous model governed by continuum equations [46, 47]. However, these modeling approaches cannot capture some important biological phenomena at the small population scale, as studied in this chapter.

In conclusion, our model is simple, but captures many essential characteristics of yeast colony development and our statistical results show good agreement with previous experiments and have verified some existing hypotheses. It can be extended to further understand the development of yeast colonies. It is worth noting that the model proposed here can serve as a framework to study multicellular organisms, especially systems such as tissues with stem cell lineage [48, 49, 50].
2.1 Introduction

Stem cells are characterized by their ability to give rise to a variety of cell types while retaining a steady stem cell pool, through self-renewal and differentiation [51]. When a stem cell divides, each progeny has the potential to remain as a stem cell (self-renewal) or becomes a cell with a more specialized function (differentiation) (Fig. 2.1A). Stem cells can be found in most mammalian tissues, participating in tissue repair in response to damage and maintaining tissue homeostasis [52, 53]. Available data suggest that most stem cells are able to switch between symmetric and asymmetric divisions, and the balance between these modes is controlled by various internal and external signals to produce appropriate numbers of stem cells and differentiated cells [51, 54]. Research suggests that the decline in adult tissue maintenance and the increase in cancer formation might be a consequence of stem cell aging [55]. Although age-related manifestation in stem cell population and function differ across tissues and organisms, decline in regenerative capacity due to depletion or dysfunction of stem cells is a hallmark [56].

Protein aggregates, dysfunction organelles, and DNA damages are commonly identified as factors of aging [52, 53]. To slow down the accumulation of aging factors,
a hypothesis suggests that mitotic cells (actively dividing cells) might asymmetrically segregate damages away from the cell whose fate is to become a new stem cell [57]. The asymmetric inheritance of cellular components in dividing cells was first observed in yeast, and has been extensively studied over the past decades. In yeast, carbonylated proteins, extrachromosomal ribosomal DNA circles and dysfunction mitochondria are retained by a mother cell during asymmetric division, while its daughter is rejuvenated with little damage [58, 59, 60]. Recent evidence suggests that stem cells may employ a similar mechanism to protect one progeny from aging [61, 62, 63, 64, 65, 66, 67] (Fig. 2.1B). Asymmetric partition of damaged proteins in stem cell division was observed in adult flies’ intestine and germline [62]: regulated by the signals from respective niches, the long-lived intestinal stem cells segregate the altered molecules to differentiating enteroblasts, protecting the stem cell pool which replenishes the short-lived intestinal cells; while the germline stem cells retain damages, protecting the differentiated progenies that give rise to new organisms. Asymmetric distribution of damaged proteins and mitochondria was also demonstrated in mammalian stem cells [64]: by tracking young and old organelles during division of human mammary epithelium stem-like cells, it was observed that high levels of old mitochondria were associated with differentiated daughters while damage-free daughters maintained stem cell traits. Asymmetric damage distribution was also found in murine neural stem cells in vitro [65]: during division of embryonic and young adult neural stem cells, altered proteins are inherited predominantly by the daughter that is destined to differentiate. Despite these emerging findings, it still remains elusive how stem cells cope with damage accumulation and how this is related to stem cell proliferation and differentiation.
Driven by the lack of knowledge of precise mechanisms regulating stem cell maintenance and differentiation in the aging process, mathematical models have been employed to address key questions and provide quantitative insights into stem cell renewal and differentiation, and decline of cellular functions in the aging process. In the following paragraphs we briefly review mathematical models proposed to study stem cell population and aging, which fall into two categories: individual-based modeling and continuous population modeling ([68, 69]).

Individual-based modeling simulates individuals or agents that have a unique set of state variables and usually interact with each other and the local environment. Some advantages of this approach include that it can take stochastic effects into consideration to describe phenomena on the level of individual cells, and that detailed molecular dynamics and cell-cell interaction can be incorporated. A few recent papers
consider various mechanisms associated with aging through individual-based computational models. A detailed summary can be found in a comprehensive review paper [69]. Assuming that cell proliferation is negatively affected by telomere shortening, an agent-based stochastic model is proposed to study telomere-dependent stem cell replicative aging [70]. In their model, cell division is simulated in discrete time interval as a random process with probability decreasing linearly as telomere is shortening. This model provides good approximation of the qualitative growth of cultured human mesenchymal stem cells. In [71], authors modeled mutation accumulation in large populations of stem cells with a discrete-time branching process where each division produces 0, 1 or 2 stem cell daughters, each of which randomly accumulates a mutation. This model demonstrated that symmetric division can reduce the risk of accumulating phenotypically silent heritable damage in individual stem cells, and has most pronounced effect on rapidly cycling tissues with increasing rates of damage accumulation.

However, a major drawback of individual-based modeling is the computational inefficiency, especially when the population is large. When a population is large and homogeneous, continuous population model using ODEs or PDEs are more appropriate to describe the population dynamics. Based on the properties of the state variables, there are two subcategories of this modeling approach. If a state variable (such as maturation stage or mutation class) is discrete, a multi-compartment model, in which the cell population is divided into hierarchical subpopulation, is suitable. If a state variable is better viewed as a continuous one, a structured-population model will be more appropriate. Next we briefly review a few representative continuous population models.
In [72], a maturity-structured two-compartment model is proposed to study the regulation of mammalian red blood cell production. The model consists of two transport equations describing population densities of mitotic cells and post-mitotic cells, and an ODE describing hormone dynamics. Mitotic cells divide and differentiate to replenish post-mitotic cells, while post-mitotic cells, by affecting the growth factor concentration (hormone), have influence on both self-renewal rate and the progression of maturity of mitotic cells. Theoretical and numerical analysis showed that a perturbation of blood-donation type leads to damped oscillatory return to normal status, and that an elevated random peripheral destruction of red blood cells leads to sustained oscillations.

In [73], the authors used a three-compartment ODE model to study the dynamics of stem cells, transit amplifying cells and terminally differentiated cells in the olfactory epithelium of mice. They identified conditions on parameters for the stability of the system when negative feedback loops are present either as Hill functions or in more general forms. Their analysis suggested that two factors, autoregulation of the proliferation of transit amplifying progenitor cells and low death rate of terminally differentiated cells, enhance the stability of the system.

The literature for other modeling works can be found in [74, 75, 76, 77] for PDE models, and [78, 79, 80, 81, 82, 83] for ODE models, and many more. However, no aging factors were introduced to any of these mentioned models and the death rates are always assumed to be constants. In [84], a system of PDEs is used to model mutation accumulation hierarchy and differentiation hierarchy of cells with stem cells on the top level and to examine cancer development and growth. In their model, maturity is treated as a continuous variable, while the number of mutation accumulation
and telomere shortening are treated as discrete cell classes. The boundary conditions describe transition among different cell classes at division: cells lose telomeres and acquire mutations. A normal cell is defined to be cancerous, if it acquires a certain number of mutations. The theoretical analysis and numerical simulations show that the more mutation classes and higher proliferation rate are sufficient to explain the faster growth of the cancer cell population.

In this research, we propose a novel model to integrate stem cell proliferation and differentiation with damage accumulation in stem cell aging process. A system of hyperbolic PDEs is constructed to model two compartments in cell lineage: mitotic cells (called stem cells) and post-mitotic cells (called TD cells). It is assumed that cell cycle progression of stem cells is a continuous process while stem cell division is discrete. The boundary conditions of the PDEs model the stem cell renewal and differentiation at division when damage segregation takes place. Cell death is modeled as an outcome of damage accumulation. Stem cell proliferation and differentiation are regulated by feedbacks from the population of TD cells and stem cells. Aging effect is added to the model through the inhibition of damage accumulation on stem cell proliferation and self-renewal. Analysis and numerical simulations are carried out to compare the effects of different regulation strategies and damage segregation rules on population dynamics and stem cell fitness. Our simulations suggest that equal distribution of damage between stem cells in symmetric renewal and less damage retention in stem cell in asymmetric division reduce the death rate of stem cells and increase TD cell populations. However, asymmetric damage segregation in stem cells leads to less concentrated damage distribution in stem cells population, which may be more stable to sudden increase of damage. Compared to feedbacks solely from
TD cells, adding feedbacks from stem cells to themselves will reduce oscillations and population overshooting in the process of population convergence to steady state. Moreover, adding the regulation that slows down the proliferation of stem cells with high level of damage and increases their tendency to differentiate can improve the fitness of stem cells by increasing the percentage of stem cells with less damage in the stem cell population. This chapter is structured as follows. The general description of our model is given in Section 2.2. In Section 2.3 a simple model without feedback regulations is presented to analyze the relation between population dynamics and various parameters. In Section 2.4 two more complex models with feedbacks from TD cells and stem cells are proposed to study different regulation mechanisms and the effect of segregation rules. Supporting analysis and figures are shown in Appendix B.

2.2 Model Description

Although molecular details regarding asymmetric segregation remain obscure, we consider a conceptually simple mathematical model. In the model, we group all mitotic cells (stem cells and multiple progenitor cells) as one compartment, call them stem cells for simplicity, and denote the population density by $S$. The other compartment consists of post-mitotic cells, or terminally differentiated cells, which we call TD cells, with the population density $T$.

Two state variables are considered: cell cycle progression $p$ and damage level $d$. When the amount of damage ($d$) accumulates and reaches a certain threshold $d^*$, the stem cells die by an apoptosis-like process as a result of aging via damage accumulation. Cell cycle progression is an indicator variable, for which a stem cell
divides when cell cycle progression \( (p) \) increases to a threshold \( p^* \), unless damage has already reached a threshold \( d^* \) in which case the stem cell is removed before division. For TD cells we assume the upper bound for \( p \) is infinity, since they will not proceed a division process. We also assume that their aging process is similar to stem cells: when the amount of damaged protein reaches a certain threshold \( d^c \), the TD cells are removed by an apoptosis-like process.

By conservation law, a system of PDEs of transport type are derived to model the evolution of population densities \( S \) and \( T \):

\[
\frac{\partial S}{\partial t} + \frac{\partial}{\partial p} (V_p S) + \frac{\partial}{\partial d} (V_d S) = 0 \tag{2.1}
\]

\[
\frac{\partial T}{\partial t} + \frac{\partial}{\partial p} (U_p T) + \frac{\partial}{\partial d} (U_d T) = 0 \tag{2.2}
\]

where \( V_p, V_d, U_p, U_d \) are all positive functions.

To ensure conservation of population in the direction of \( d \), we impose no-flux conditions on the boundary \( d = 0 \):

\[
S(t, p, 0) = 0, \quad \text{for } t > 0, \quad p \in [0, p^*] \tag{2.3}
\]

\[
T(t, p, 0) = 0, \quad \text{for } t > 0, \quad p \in [0, \infty). \tag{2.4}
\]

We also assume

\[
\lim_{p \to \infty} T(t, p, d) = 0. \tag{2.5}
\]

The boundary conditions at \( p = 0 \) describe the reproduction process and damage segregation. Before stating the conditions, we first make some assumptions: the renewal and differentiation process takes place at cell division \( (p = p^*) \); cell fate decisions/division types are given by \( 0 \leq \delta_1, \delta_2, \delta_3 \leq 1 \) with \( \delta_1 + \delta_2 + \delta_3 = 1 \) in the following manner: with probability \( \delta_1 \) the daughter cells are two stem cells, with
probability $\delta_2$ are two terminally differentiated cells, and with probability $\delta_3$ are one stem cell and one TD cell; upon the completion of a cell cycle, the cell cycle progression $p$ will be reset to zero and the damaged protein are inherited from mother to daughters; the damage inheritance can be described by transition kernels $r_i(d, d')$.

See Fig. 2.2 for illustration. Thus we impose the following boundary conditions for $S$ and $T$ cells at $p = 0$:

$$V_pS(t, 0, d) = \int_0^{d^*} \delta_1V_pr_1(d, d')S(t, p^*, d')dd' + \int_0^{d^*} \delta_3V_pr_3(d, d')S(t, p^*, d')dd' \quad (2.6)$$

$$U_pT(t, 0, d) = \int_0^{d^*} \delta_2V_pr_2(d, d')S(t, p^*, d')dd' + \int_0^{d^*} \delta_3V_pr_4(d, d')S(t, p^*, d')dd' \quad (2.7)$$

where $r_i(d, d')$'s are transition kernels describing how daughters with damage $d$ come from the mothers with damage $d'$ and satisfying the conservation conditions:

$$\int_0^{t_d} \int_0^{t_d} r_i(d, d')dd'dd' = 2, \quad i = 1, 2,$$

$$\int_0^{t_d} \int_0^{t_d} r_j(d, d')dd'dd' = 1, \quad j = 3, 4,$$

$$\int_0^{t_d} \int_0^{t_d} d r_i(d, d')dd'dd' = d', \quad i = 1, 2,$$

$$\int_0^{t_d} \int_0^{t_d} d r_3(d, d')dd'dd' + \int_0^{t_d} \int_0^{t_d} d r_4(d, d')dd'dd' = d'.$$

Then the populations of stem and TD cells at time $t$ are given by the integrals

$$P_S(t) = \int_0^{d^*} \int_0^{p^*} S(t, p, d)dpdd \quad \text{and} \quad P_T(t) = \int_0^{d^*} \int_0^{\infty} T(t, p, d)dpdd.$$

One important remark is that to be realistic we regard the stem cell population $P_S(t)$ as extinct and stop the propagation once its value drops below a certain small number, which can take different values according to scaling. For example, if there is no scaling of the populations, then we can naturally take this number to be 1.
Figure 2.2: Stem cells (mitotic cells) and TD cells (post-mitotic cells) are modeled as two compartments. Stem cells renew themselves and replenish TD cells. The cell cycle progression ($p$) and damage accumulation ($d$) are modeled as continuous processes. Stem cell division and damage segregation take place at the end of stem cell cell cycle ($p = p^*$). Cells die when damage reaches lethal threshold ($d^*$ or $d^c$).
In the sequel we will study population dynamics, starting with models in simple situations in Section 2.3 and then with more complex models in Section 2.4.

### 2.3 Models with constant parameters

In this section we make some simplifying assumptions: (a) cell cycle progression speeds \( V_p = v_p \) and \( U_p = u_p \), as well as damage accumulation rates \( V_d = v_d \) and \( U_d = u_d \), are constant; (b) the division rule is fixed, i.e., the proportions of three types of divisions \( \delta_i \) are fixed numbers; (c) the damage segregation rules in each division type is also fixed: damage is partitioned with portions \( \alpha_1 \) and \( \alpha_2 \) between stem cells, \( \beta_1 \) and \( \beta_2 \) between TD cells, and \( \gamma_1 \) and \( \gamma_2 \) between stem and TD cells, where \( \alpha_1 + \alpha_2 = 1, \beta_1 + \beta_2 = 1 \) and \( \gamma_1 + \gamma_2 = 1 \) (Fig. 2.3). Under these assumptions, the transition kernels in the boundary conditions are Dirac delta functions:

\[
\begin{align*}
    r_1(d, d') &= \delta \left( \frac{d}{d'} - \alpha_1 \right) + \delta \left( \frac{d}{d'} - \alpha_2 \right) \\
    r_2(d, d') &= \delta \left( \frac{d}{d'} - \beta_1 \right) + \delta \left( \frac{d}{d'} - \beta_2 \right) \\
    r_3(d, d') &= \delta \left( \frac{d}{d'} - \gamma_1 \right) \\
    r_4(d, d') &= \delta \left( \frac{d}{d'} - \gamma_2 \right)
\end{align*}
\]

And the boundary conditions in (2.6) and (2.7) become

\[
\begin{align*}
    S(t, 0, d) &= \frac{\delta_1}{\alpha_1} S(t, p^*, \frac{d}{\alpha_1}) + \frac{\delta_1}{\alpha_2} S(t, p^*, \frac{d}{\alpha_2}) + \frac{\delta_3}{\gamma_1} S(t, p^*, \frac{d}{\gamma_1}) \\
    T(t, 0, d) &= \frac{v_p}{u_p \beta_1} \delta_2 S(t, p^*, \frac{d}{\beta_1}) + \frac{v_p}{u_p \beta_2} S(t, p^*, \frac{d}{\beta_2}) + \frac{v_p}{u_p \gamma_2} S(t, p^*, \frac{d}{\gamma_2}).
\end{align*}
\]  

(2.8) (2.9)

To simplify the analysis, we make the following change of variables:

\[
\tilde{p} = \frac{p}{v_p}, \quad \tilde{d} = \frac{d}{v_d}
\]
Figure 2.3: In the simple model in Section 2.3, the proportions of three types of division are constant $\delta_i$, and the damage segregation rules are fixed, i.e., $\alpha_i, \beta_i, \gamma_i$ are constants.

The domain of new variables is $0 \leq \tilde{p} \leq t_p = \frac{p^*}{v_p}$ and $0 \leq \tilde{d} \leq t_d = \frac{d^*}{v_d}$. Since we assume constant velocity of cell cycle progression $v_p$, the biological meaning of the parameter $t_p$ is the duration of one cell cycle of stem cells. Similarly, $t_d$ means the time needed to accumulate damage from zero to lethal threshold. With the new variables, (2.1) and (2.2) become

$$\frac{\partial \tilde{S}}{\partial t} + \frac{\partial \tilde{S}}{\partial \tilde{p}} + \frac{\partial \tilde{S}}{\partial \tilde{d}} = 0 \quad (2.10)$$

$$\frac{\partial \tilde{T}}{\partial t} + \frac{u_p}{v_p} \frac{\partial \tilde{T}}{\partial \tilde{p}} + \frac{u_d}{v_d} \frac{\partial \tilde{T}}{\partial \tilde{d}} = 0 \quad (2.11)$$

where $\tilde{S}(t, \tilde{p}, \tilde{d}) = S(t, p, d)$ and $\tilde{T}(t, \tilde{p}, \tilde{d}) = T(t, p, d)$. For simplicity, we will drop all the tildes on the notations. The dynamics of stem cells are independent of TD cells, while the dynamics of TD cells are determined by stem cell through boundary condition (2.9). Once the behavior of stem cell is determined, the behavior of TD cell can be easily derived. So we mainly focus on the dynamics of stem cells.
2.3.1 Analytic solution

Using the method of characteristics, the analytic solution can be derived and determined by initial and boundary conditions. Due to the complexity of boundary conditions, no closed form of $S(t, p, d)$ can be obtained, and the solution is expressed in a recursive form. Within the first cell cycle, i.e., $0 \leq t \leq t_p$, $S(t, p, d)$ can be solved as (the derivation can be found in Appendix B.1).

$$S(t, p, d) = \begin{cases} 
S(0, p-t, d-t) & \text{if } t \leq p \text{ and } t \leq d \\
0 & \text{if } t > d \text{ and } d < p \\
H(t, p, d, \alpha_1, t_d)\frac{\delta_1}{\alpha_1}S\left(0, t_p - (t - p), \frac{d-p}{\alpha_1} - (t - p)\right) & \text{if } t > p \text{ and } p < d,
\end{cases}$$

(2.12)

$$H(t, p, d, \omega, t_d) = \begin{cases} 
1 & \text{if } \frac{d-p}{\omega} < t_d \text{ and } \frac{d-p}{\omega} - (t - p) > 0; \\
0 & \text{otherwise}.
\end{cases}$$

(2.13)

For the time beyond the first cell cycle, i.e., $t > t_p$, by tracing back the process of damage accumulation by one cycle, $S(t, p, d)$ is solved as

$$S(t, p, d) = H(t_p, p, d, \alpha_1, t_d)\frac{\delta_1}{\alpha_1}S\left(t - t_p, p, \frac{d-p}{\alpha_1} - (t_p - p)\right)$$

(2.14)

$$+ H(t_p, p, d, \alpha_2, t_d)\frac{\delta_1}{\alpha_2}S\left(t - t_p, p, \frac{d-p}{\alpha_2} - (t_p - p)\right)$$

$$+ H(t_p, p, d, \gamma_1, t_d)\frac{\delta_3}{\gamma_1}S\left(t - t_p, p, \frac{d-p}{\gamma_1} - (t_p - p)\right).$$

Applying (2.14) iteratively and eventually (2.12), we can obtain the density of the stem cell population at any time.

2.3.2 Analysis of population dynamics

Due to the complexity of the recursive form of the density function, it is generally difficult to obtain the long-term behavior of the stem cell population directly from
the population density. We can use an alternative approach to analyze the effects of parameters on the long-term behavior of the stem cell population.

Among all parameters, we are especially interested in the role played by the damage segregation rules in population dynamics. Since the segregation rule is fixed, after sufficiently long time, the cellular damage at the end of cell cycle, temporarily assuming no death, converges to a limit damage band

\[
\left[ \frac{t_p}{1 - \omega_1}, \frac{t_p}{1 - \omega_2} \right],
\]

where \( \omega_1 \) and \( \omega_2 \) are the minimum and the maximum of \( \alpha_1, \alpha_2, \gamma_1 \), respectively. The derivation of damage limit band can be found in Appendix B.2. Without loss of generality, in the following discussion, we assume \( \alpha_1 \leq \alpha_2 \).

The population dynamics turns out to depend on the proportions of three division types of stem cells and the position of the lethal threshold \( t_d \) with respect to the limit damage band \( \left[ \frac{t_p}{1 - \omega_1}, \frac{t_p}{1 - \omega_2} \right] \). Before proceeding, we first introduce the self-renewal fraction

\[
f_r = \frac{2\delta_1 + \delta_3}{2} = \frac{1 + \delta_1 - \delta_2}{2}.
\]

Our findings are discussed in the following.

**Proposition 2.1.** Stem cells become extinct, if the renewal fraction \( f_r < 1/2 \) or the limit damage band is completely above the lethal threshold, i.e., \( t_d < \frac{t_p}{1 - \omega_1} \).

**Proposition 2.2.** Assume that the limit damage band lies completely below the lethal threshold, i.e., \( t_d > \frac{t_p}{1 - \omega_2} \), and

\[
\int_0^{t_p} \int_0^{\omega_2 t_d + p} S(0, p, d) \, dd \, dp > 0.
\]

Then stem cell population blows up to infinity if the renewal fraction \( f_r > 1/2 \), or is eventually conserved if \( f_r = 1/2 \).
The proofs of Propositions 2.1 and 2.2 can be found in Appendices B.3 and B.4 respectively. In the uninteresting situations shown in Propositions 2.1 and 2.2, either no cell can survive after sufficiently long time or damage accumulation does not affect the cells.

Next we consider the more intriguing intermediate situations as follows, based on the following assumptions on the renewal fraction and the lethal threshold:

\[ f_r \geq \frac{1}{2} \quad \text{and} \quad \frac{t_p}{1 - \omega_1} \leq t_d < \frac{t_p}{1 - \omega_2}. \]  

(2.15)

We point out that under the above assumptions in (2.15), there are many situations of damage segregation rules and relative positions of \( t_d \) with respect to the limit damage band, and more importantly not all of the situations are biologically meaningful. Here, we will focus on one situation and other situations can be discussed in a similar manner. We assume that the damage retention in stem cells through asymmetric division is smaller than the damage segregation portions in symmetric stem cell renewal, i.e.,

\[ \gamma_1 \leq \alpha_1 < \alpha_2. \]  

(2.16)

The biological meaning behind this setting is that we believe that asymmetric division is a favorable mechanism for stem cell lineage to clear out the damage. It is uninteresting to have the lethal threshold \( t_d \) to lie below \( \frac{t_p}{1 - \alpha_1} \), i.e., all asymmetrically renewing stem cells are destined to die. Thus we further assume

\[ f_r > \frac{1}{2} \quad \text{and} \quad \frac{t_p}{1 - \gamma_1} \leq \frac{t_p}{1 - \alpha_1} < t_d < \frac{t_p}{1 - \alpha_2}. \]  

(2.17)

The conditions \( f_r = \frac{2 \delta_1 + \delta_3}{2} > \frac{1}{2} \) and \( 0 < \delta_1 + \delta_3 \leq 1 \) form the triangular region shown in Fig. 2.4. Naturally one wishes to obtain a partition of the space of the parameters,
the triangular region for \((\delta_1, \delta_3)\), to categorize different types of population dynamics, for chosen parameters \(t_p, t_d, \alpha_i, \gamma_i\). However, it is in general very difficult to achieve this goal by purely using rigorous mathematical analysis. But we managed to find a condition to guarantee extinction of stem cells, as well as a condition for population blow-up, stated in the following result whose proof is given in Appendix B.5.

**Proposition 2.3.** Assume that \((2.17)\) holds.

(a) For integers \(m \geq 1\), define

\[
n(m) = \min \left\{ n \in \mathbb{N} : t_d < \frac{1 - \gamma_1^n}{1 - \gamma_1} t_p \alpha_2^n + \frac{1 - \alpha_2^n}{1 - \alpha_2} t_p \right\}.
\]

For any fixed \(m \geq 1\), if there exists a combination of \(\delta_i\) such that \((2f_r)^n(m) - \delta_1^n(m) < 1\), then the stem cell population becomes extinct eventually.

(b) If \(\delta_2 = 0\), the stem cell population blows up.

Both (a) and (b) yield subspaces of the triangular region \((2\delta_1 + \delta_3 > 1\) and \(0 < \delta_1 + \delta_3 \leq 1\), showing distinct population dynamics (see Fig. 2.4 solid red regions for (a) and solid solid green lines for (b)). However in the remaining part of the triangular region we fail to describe the population dynamics mathematically and thus have to turn to numerical simulation. In the numerical experiments, we choose equidistributed sample points in the triangular region, simulate population evolution, and record the population dynamics. Underlying each simulation, we are particularly interested in the choices of parameters that give rise to unbounded populations, since we will discuss various negative feedback regulations to stabilize the stem cell population. Under the assumption \((2.17)\), interesting results obtained from simulations are summarized in the following, with an illustration in Fig. 2.4 where the green...
dots represent the samples whose population blew up and the red dots represent the samples whose population went to zero.

i. For fixed segregation rules, the region of population blow-up expands as damage accumulation speed slows down.

As shown in Fig. 2.4A, when we decrease the damage accumulation speed, i.e., $t_d$ is prolonged from 8.33 to 12.5 under fixed segregation rules $\alpha_1 = 0.3, \gamma_1 = 0.1$, not only the solid red region given by Proposition 2.3 shrinks, the number of red dots generated by numerical simulations also becomes fewer, which implies that there are more combinations of parameters $(\delta_1, \delta_3)$ that give rise to unbounded populations of stem cells.

ii. For fixed damage accumulation speed, the region of population blow-up expands as damage retention in asymmetric division decreases or damage distribution in self-renewal becomes more equal.

Under fixed damage accumulation ($t_d = 10$), although the solid red regions given by Proposition 2.3 are the same, the simulations show that the region of population blow-up expands as damage retention $\gamma_1$ in asymmetric division decreases (Figs. 2.4B) or as damage distribution $(\alpha_1, \alpha_2)$ in self-renewal becomes more equal (Figs. 2.4C).

In conclusion, for most of the combinations of parameters, the stem cell populations in the models without feedback regulation blow up to infinity or diminish to zero. Although the no-regulation assumption is not realistic, the toy model not only provides us guidance on the selection of parameters in the following discussions, but also reveals that population dynamics are results of all factors: the stem cell cell cycle progression, damage accumulation, fractions of divisions and damage segregation rules. In the next section, we will consider the combinations of parameters that
guarantee population exponential growth and study feedback regulations that could lead the population to steady state.

2.4 Models with feedbacks

Biological evidence shows that some mammalian stem cells can switch between symmetric and asymmetric divisions in response to external and internal regulations [51, 54]. For example, both epidermal [85] and neural [86] progenitors change from primarily symmetric division that expand the stem-cell pool during embryonic development to primarily asymmetric in mid to late gestation. It is also observed that nervous [87] and haematopoietic [88] stem cells in adults can divide symmetrically to replace lost cells through injury, although they divide asymmetrically under steady-state conditions.

Feedback regulations affecting the processes of cell division and differentiation have been documented in the mouse olfactory epithelium, skeletal muscle, bone, and the hematopoietic system [82]. Healthy cell lineages are highly regulated to promote the rapid regeneration in embryonic development or after an injury and to maintain tissue homeostasis under normal conditions. In particular, two types of feedbacks have been proposed [89, 82]: the long-range feedbacks responding to the population of terminally differentiated cells, and short-range feedbacks acting in an autocrine fashion in stem cells (Fig. 2.5).

Feedbacks regulate stem cell population through two fundamental parameters: the speed of stem cell division (or cell cycle progression speed), and the types of division (differentiation vs renewal). Possibly mediated by regulatory molecules, the
Figure 2.4: The solid green line and green dots represent the combinations of \((\delta_1, \delta_3)\) whose corresponding population blew up, while the solid red region and red dots represent the situation where population went to zero. The solid regions and lines are from Proposition 2.3 and the dots are from simulations. In A1-A3, \(\alpha_1 = 0.3, \gamma_1 = 0.1\) \(t_p = 5\) and \(t_d\) is tested for \(8.33, 10\) and \(12.5\). In B1-B3, \(t_p = 5, t_d = 10, \alpha_1 = 0.3\) and \(\gamma_1\) is tested for \(0.3, 0.2,\) and \(0.1\). In C1-C3, \(t_p = 5, t_d = 10, \gamma_1 = 0.3\) and \(\alpha_1\) is tested for \(0.1, 0.25,\) and \(0.4\).
underlying mechanisms are not fully understood. Mathematical models with different feedbacks can help to explain which types of feedback best fits the observations in biological systems. Before stating our more complex models, we make the following assumptions based on the analysis from the simple model in Section 2.3: (i) the self-renewal fraction $f_r > \frac{1}{2}$; (ii) damage accumulation speed is slower than stem cell cell cycle progression, and the relative importance of renewal and differentiation forces stem cell population to blow up without any feedback regulation.

### 2.4.1 Feedbacks from TD cells

The long-range feedbacks, which act through signals secreted by differentiated cells and inhibit stem cell division and self-renewal, have been biologically observed in numerous tissues, including muscle [90], bone [91], skin [92], nervous system [93] and hematopoietic systems [94]. Despite this wealth of data, there is less understanding of
the exact mechanisms of feedback regulation. A significant number of mathematical
models have been developed to explore the possible mechanisms behind feedback
regulations \[72, 78, 73, 79, 75, 80, 77, 81, 82, 95, 83, 96\]. The dynamics of signaling
molecule \(s(t)\) can be described by a simple ODE as follows:

\[
\frac{d}{dt} s(t) = \alpha - (\mu + \beta P^m) s(t), \tag{2.18}
\]

where \(\alpha\) is a constant synthesis rate, and the degradation is proportionally to the level
of \(s\) and affected by the total cell population \(P\). Since the dynamics of the signaling
molecules take place on a faster time scale than the process of cell proliferation and
differentiation, mathematical models always assume that the feedback signal fulfills
a quasi-steady state assumption \[72, 74, 73, 78, 79, 80, 77, 81, 95, 82, 83, 96\]. By
properly rescaling \(s\), the quasi-steady state of \(s\) is given by a Hill function

\[
\tilde{s} = \frac{1}{1 + (kP)^m}, \tag{2.19}
\]

where \(k\) is a regulation constant to account for the sensitivity to the cell population
and \(m\) is the Hill exponent. The function in (2.19) reflects the assumption that
the signal intensity achieves its maximum when in the absence of certain cells and
decreases asymptotically to zero if the number of cells increases. We mention that Hill
functions are widely used to describe ligand-receptor interactions, which also makes
them natural choices to model the actions of secreted feedback factors \[81\].

According to biological evidence, we may model the feedbacks from TD cells in
the following way: when stem cell or TD cell population is small (in the early stage
of development or with drastic loss due to injury), symmetric division predominates
over asymmetric division; during the stable stage (mid and late gestation or tissue
homeostasis), stem cells switch to asymmetric division. Such feedbacks may be added to the model by modifying division fractions $\delta_i$ as follows:

$$\delta_i(P_T) = \frac{\delta^0_i}{1 + (k^T_i P_T)^{m_T}},$$  \hspace{1cm} (2.20)

where $i \in \{1, 2\}$, $\delta^0_i$ are maximum fractions of certain division, $k^T_i$ are regulation constants, $m_T$ is the Hill exponent and $P_T$ is the TD cell population.

Recent studies on cell cycles of neural stem cells have shown evidence that terminally differentiated cells may also provide a source of signaling molecules that inhibit the cell cycle progression of stem cells [97]. Therefore, besides the above regulation on cell fate decisions, negative feedback from TD cells can also be added to stem cell proliferation $V_p$ (cell cycle progression speed), i.e., excessive number of TD cells may slow down the proliferation of stem cell, and hence reduce the population of both stem cells and TD cells. Thus, $V_p$ can be modified as

$$V_p(P_T) = \frac{v_p}{1 + (k^T_v P_T)^{m_T}},$$  \hspace{1cm} (2.21)

where $v_p$ is the maximum cell cycle progression speed and $k^T_v$ is a regulation constant.

In summary, in our first model with feedback regulation, TD cell population simultaneously regulates the stem cell division fractions and proliferation speed (see Fig. 2.6).

Among all parameters, we are most interested in the effect of segregation rules $\alpha_i, \beta_i, \gamma_i$ on population dynamics. A set of reasonable estimations of the parameters other than $\alpha_i, \beta_i, \gamma_i$ are chosen based on appropriate biological and mathematical assumptions and are shown in Table 2.1. The detailed reasoning of the selection of parameters is provided in Appendix B.6. In brief, we assume that the initial
Figure 2.6: Overpopulation of TD cells slows down stem cell proliferation speed and inhibits stem cell symmetric renewal and differentiation.

setting is that stem cells are expanding, the initial cell cycle progression is fast, and the symmetric renewal predominates in three types of division. In the rest of this subsection, we will focus on the discussion of the segregation rules $\alpha_i, \beta_i, \gamma_i$.

In the simulations, we consider the following situations: $\alpha_1$ varies from 0.1 to 0.5, indicating that the symmetry in damage segregation increases between two stem cells; $\beta_1$ varies from 0.1 to 0.5, indicating that the symmetry in damage segregation increases between TD cells; $\gamma_1$ varies from 0.1 to 0.9, indicating that the damage retention in stem cells increases in asymmetric division. Under different segregation rules, we are interested in several aspects: the death rates of TD cells and stem cells; the fraction of three types of division in steady state; the population sizes of TD cells and stem cells and the population ratio of TD cells versus stem cells, the dynamics of population evolution, and the damage distribution of stem cell population. Simulation results are analyzed as follows:
<table>
<thead>
<tr>
<th>Parameters</th>
<th>Meaning</th>
<th>Value</th>
</tr>
</thead>
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<tr>
<td>$v_p$</td>
<td>Maximum cell cycle progression speed of stem cells</td>
<td>0.2</td>
</tr>
<tr>
<td>$v_d$</td>
<td>Constant damage accumulation speed of stem cells</td>
<td>0.06</td>
</tr>
<tr>
<td>$u_d$</td>
<td>Constant damage accumulation speed of TD cells</td>
<td>0.02</td>
</tr>
<tr>
<td>$\delta^0_1$</td>
<td>Maximum fraction of symmetric renewal of stem cells</td>
<td>0.6</td>
</tr>
<tr>
<td>$\delta^0_2$</td>
<td>Maximum fraction of symmetric differentiation of stem cells</td>
<td>0.3</td>
</tr>
<tr>
<td>$\delta^0_3$</td>
<td>Maximum fraction of asymmetric division of stem cells</td>
<td>0.1</td>
</tr>
<tr>
<td>$k^T_T$</td>
<td>Regulation constant</td>
<td>$10^{-8}$</td>
</tr>
<tr>
<td>$k^T_T$</td>
<td>Regulation constant</td>
<td>$0.5 \times 10^{-8}$</td>
</tr>
<tr>
<td>$k^T_T$</td>
<td>Regulation constant</td>
<td>$0.5 \times 10^{-8}$</td>
</tr>
<tr>
<td>$m_T$</td>
<td>Hill exponent</td>
<td>2</td>
</tr>
</tbody>
</table>

Table 2.1: The choices of parameters, estimated based on biological evidence and previous modeling works, with details in Appendix B.6.

i. Damage distribution between TD cells in symmetric differentiation does not affect population dynamics.

The parameter $\beta_1$, which determines how damage is distributed between two TD cells in the symmetric differentiation of stem cells, does not affect either stem cell population or TD cell population much. This result is biologically meaningful, since the replenishment of TD cells is efficient with respect to loss of TD cells due to damage accumulation. In the following part, we will focus on the case that $\beta_1 = 0.5$. More results when $\beta_1 = 0.1$ and 0.3 can be found in Appendix B.7.

ii. More equal distribution of damage in symmetric renewal and less damage retention in asymmetric division result in lower death rate of stem cells and less symmetric division in steady state.
According to our simulations, the segregation rules do not have much impact on the death rate of TD cells, which ranges from \(2.130 \times 10^{-4}\) to \(3.345 \times 10^{-4}\). However, the death rate of stem cells changes dramatically as we vary the segregation rules. From Table 2.2 we can see that the death rate of stem cells increases as \(\alpha_1\) decreases or \(\gamma_1\) increases. The smallest death rate of stem cells is attained when damage is equally distributed between stem cells in symmetric renewal and damage retention is minimal in asymmetric division. To maintain the steady stem cell population and to replenish the short lived TD cells, higher death rate should be associated with fast turnover of stem cells. Indeed, when we examine the fractions of three types of division, we find that higher death rate of stem cells is always associated with higher fraction of symmetric renewal (Table 2.3). These results suggest that to maintain stabilized populations stem cells may have different mechanisms that involve different damage segregation rules and division rules.

iii. More equal distribution of damage in symmetric renewal and less damage retention in asymmetric division result in larger TD cell population.

Fig. 2.7 compares the TD cell population and the population ratio \(P_T/P_S\) in steady state under these different segregation rules. For fixed \(\gamma_1\), as damage segregation \(\alpha_i\) between stem cells becomes more symmetric, TD cell populations increase, while the ratios \(P_T/P_S\) are almost constant with slight decrease. For fixed \(\alpha_1\), as damage retention \(\gamma_1\) in asymmetric division increases, both TD cell populations and the population ratios decrease. Interestingly, when \(\alpha_1 = 0.5\), although the populations of TD cells are very close for the cases \(\gamma_1 = 0.1, 0.3\) and \(0.5\), the population ratios differ dramatically. Compared to \(\gamma_1 = 0.1\), one needs a larger stem cell pool to generate a similar number of TD cells when damage retention is relative higher, i.e., \(\gamma_1 = 0.5\).
<table>
<thead>
<tr>
<th>$\gamma_1$</th>
<th>$\alpha_1$</th>
<th>0.1</th>
<th>0.2</th>
<th>0.3</th>
<th>0.4</th>
<th>0.5</th>
</tr>
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<tr>
<td>0.1</td>
<td>1.096 x 10^{-4}</td>
<td>6.586 x 10^{-5}</td>
<td>2.913 x 10^{-5}</td>
<td>7.536 x 10^{-6}</td>
<td>7.646 x 10^{-8}</td>
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<tr>
<td>0.3</td>
<td>1.215 x 10^{-4}</td>
<td>8.962 x 10^{-5}</td>
<td>4.631 x 10^{-5}</td>
<td>1.200 x 10^{-5}</td>
<td>1.514 x 10^{-7}</td>
<td></td>
</tr>
<tr>
<td>0.5</td>
<td>1.909 x 10^{-4}</td>
<td>1.399 x 10^{-4}</td>
<td>8.515 x 10^{-5}</td>
<td>3.162 x 10^{-5}</td>
<td>6.343 x 10^{-6}</td>
<td></td>
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<tr>
<td>0.7</td>
<td>2.655 x 10^{-4}</td>
<td>2.398 x 10^{-4}</td>
<td>2.115 x 10^{-4}</td>
<td>1.769 x 10^{-4}</td>
<td>1.565 x 10^{-4}</td>
<td></td>
</tr>
<tr>
<td>0.9</td>
<td>3.171 x 10^{-4}</td>
<td>3.101 x 10^{-4}</td>
<td>2.873 x 10^{-4}</td>
<td>2.599 x 10^{-4}</td>
<td>2.658 x 10^{-4}</td>
<td></td>
</tr>
</tbody>
</table>

Table 2.2: Death rate of stem cells in steady state under different segregation rules.

Moreover, it would be very difficult to find explicit formulas for the populations, since the population sizes in steady state are affected by all parameters as observed in the simulations. However, when steady state exists, we can give an upper-bound estimate of the populations of stem cells and TD cells, with details given in Appendix B.8. When the death rate of stem cells is rather small in steady state, our upper-bound estimation of TD cell population is very close to the real value. An example of such situation is also given in Appendix B.8.

iv. Higher damage retention results in oscillations in population evolution.

Given $\beta_1 = 0.5$, Fig. 2.8 shows the comparison of population dynamics among 9 combinations of $\alpha_1$ and $\gamma_1$. Oscillations start to appear as we increase the damage retention $\gamma_1$ in stem cells during asymmetric division (Figs. 2.8G,H,I). Also the oscillations become more severe when the damage distribution among stem cells in symmetric renewal becomes more symmetric (Fig. 2.8). Moreover, population overshooting before steady state is observed in all cases that $\alpha_1 \neq 0.5$. 

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Table 2.3: Symmetric renewal fraction $\delta_1$ in steady state under different segregation rules.

<table>
<thead>
<tr>
<th>$\gamma_1$</th>
<th>$\alpha_1$</th>
<th>0.1</th>
<th>0.2</th>
<th>0.3</th>
<th>0.4</th>
<th>0.5</th>
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<tr>
<td>0.1</td>
<td></td>
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<td>0.27</td>
<td>0.23</td>
<td>0.21</td>
<td>0.20</td>
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<tr>
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<td>0.20</td>
</tr>
<tr>
<td>0.5</td>
<td></td>
<td>0.37</td>
<td>0.33</td>
<td>0.28</td>
<td>0.23</td>
<td>0.21</td>
</tr>
<tr>
<td>0.7</td>
<td></td>
<td>0.42</td>
<td>0.40</td>
<td>0.37</td>
<td>0.34</td>
<td>0.33</td>
</tr>
<tr>
<td>0.9</td>
<td></td>
<td>0.45</td>
<td>0.45</td>
<td>0.43</td>
<td>0.42</td>
<td>0.42</td>
</tr>
</tbody>
</table>

Figure 2.7: TD cell population and population ratio for different combinations of $\alpha_1$ and $\gamma_1$ in steady state. The parameters used in these simulations are shown in Table 2.1.
Figure 2.8: Sample population dynamics under different combinations of $\alpha_1$ and $\gamma_1$. The red curves stands for TD cell population and the blue curve for stem cell population. The parameters used in these simulations are listed in Table 2.1. To eliminate the influence of the initial condition, a perturbation of reducing population of TD cells by 20% is introduced after the populations reach the steady state.
The above analysis i-iv suggests that more equal distribution of damage in symmetric renewal and less damage retention in asymmetric division are favorable segregation rules in four aspects: populations converge to steady states faster without oscillations or severe population overshooting; the death rate of stem cells is much smaller than that of TD cells; asymmetric division predominates in three types of division in steady state; not only the size of TD cell population is larger, the population ratio $P_T/P_S$ is also larger.

However, the asymmetric damage segregation between stem cells in self-renewal may have benefits that cannot be seen from the analysis of total populations as integrated results. Our PDE modeling approach allows us to investigate more details of population density and damage distribution of stem cells.

v. Asymmetric damage segregation in stem cells leads to less concentrated damage distribution in stem cells.

Table 2.4 shows a few samples of segregation rules, and Fig. 2.9 gives the corresponding stem cell population densities and damage distributions at the end of cell cycle $p = p^*$. As we studied in Section 2.3.2, damage segregation rules determine the limit damage band after sufficiently long time. In particular, the less symmetric the segregation rules are, the wider the limit damage bands will be. Although asymmetric segregation rules may result in a greater percentage of stem cells inheriting more damage and accelerated death, it leads to a higher percentage of healthier stem cells with less damage. This can be observed in the samples shown in Fig. 2.9 and Table 2.4. On the one hand, in Table 2.4, the symmetry of damage segregation in stem cell population increases from case A to case D. On the other hand, Fig. 2.9 shows
that the damage distributions at the end of cell cycle for stem cells become more concentrated, as such symmetry increases. The concentrated damage distribution could be a disadvantage, since it is less resistant to external perturbations. In this sense, we think that some stem cells may sacrifice the lower death rate for more diversified damage distribution, in order to protect the stem cell pool from possible unfavorable situation that may lead to sudden increase in damage.

2.4.2 Feedbacks from stem cells

The maintenance of the stem cell pool is not only affected by signaling from mature cells, but also by the stem cell pool itself. Stem cells reside in the so-called stem cell niche, where both cellular and non-cellular components interact in order to control stem cell proliferation and differentiation [98, 99, 100, 101]. The regulations from the stem cell population include two aspects: a negative control of stem cell proliferation as a result of contact inhibition; and a self-renewal inhibition factor secreted by the stem cell niche, in which case the more stem cells there are, the less likely stem cells will divide symmetrically. Experiments also show that stem cells inheriting the
Figure 2.9: Population density and damage distribution at the end of cell cycle for stem cells. The damage segregation rules in cases A-D are shown in Table 2.4. All the other parameters are in Table 2.1. The damage distributions are re-scaled for comparison. Continued.
majority of damage protein aggregates during asymmetric division have an increased cell cycle length and higher tendency to differentiate [63, 65, 66].

In addition to feedback regulations from mature cells, we are also interested in how the dynamics of populations would change if regulations from stem cells are added. Many modeling works (see e.g. [73, 82]) only consider the feedbacks from stem cell populations, due to the simplicity of their models. However, we include damage as a state variable in our model and could simulate feedbacks from both the stem cell population and the stem cell cellular damage level.

Thus, in our model we assume that the excessive stem cell population and elevated stem cell cellular damage slow down stem cell proliferation and modify $V_p$ in (2.21) as

$$V_p(P_T, P_S, d) = \frac{v_p}{1 + (k^{T}_{P} P_T)^m + (k^{S}_{P} P_S)^m} f(d),$$

(2.22)
Figure 2.10: Inhibitions on stem cell proliferation and symmetric renewal and differentiation are from both stem cells (blue lines) and TD cells (red lines).

where

\[ f(d) = a_1 + \frac{b_1}{1 + e^{-k_1^d(d-d_1^0)}} \]

is a decreasing function of \( d \) with sigmoid shape, in which \( k_1^d, d_1^0, a_1 \) and \( b_1 \) are constants.

We also assume that the excessive stem cell population inhibits stem cell symmetric division, and that the elevated stem cell cellular damage promotes stem cell differentiation via decreasing stem cell self-renewal. As a result, we modify \( \delta_1 \) and \( \delta_2 \) in \text{(2.20)} in the same way as above:

\[ \delta_1(P_T, P_S, d) = \frac{\delta_1^0}{1 + (k_{1T}^d P_T)^{m_T} + (k_{1S}^S P_S)^{m_S}} g(d), \quad (2.23) \]
\[ \delta_2(P_T, P_S, d) = \frac{\delta_2^0}{1 + (k_{2T}^d P_T)^{m_T} + (k_{2S}^S P_S)^{m_S}} g(d), \quad (2.24) \]

where

\[ g(d) = a_2 + \frac{b_2}{1 + e^{-k_2^d(d-d_2^0)}} \]

is another function of sigmoid shape with constants \( k_2^d, d_2^0, a_2 \) and \( b_2 \).
### Table 2.5: The choices of additional parameters, estimated based on biological evidence and previous modeling works, with details in Appendix B.9.

<table>
<thead>
<tr>
<th>Parameters</th>
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<tr>
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<td>Regulation constant</td>
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</tr>
<tr>
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<td>Hill exponent</td>
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</tr>
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<td>$a_1$</td>
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<tr>
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</tr>
<tr>
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<tr>
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As a continuation of Section 2.4.1, our analysis still focuses on the discussion of segregation rules. In addition to the parameters in Table 2.1, the values of more parameters are given in Table 2.5 for which the detailed reasoning can be found in Appendix B.9.

To examine the change in population dynamics after introducing feedbacks from stem cells, the same combinations of segregation rules as in Section 2.4.1 are considered: $\alpha_1$ varies from 0.1 to 0.5, $\beta_1$ varies from 0.1 to 0.5, and $\gamma_1$ varies from 0.1 to 0.9. In addition to previously considered factors (the death rates of TD cells and stem cells, the fraction of symmetric renewal in steady state, the TD cell population, the population ratio $P_S/P_T$ and dynamics of population evolution), we also analyze the distribution of damage at the end of cell cycle, which is reasonable since we include
Figure 2.11: TD cell population and population ratio for different combinations of $\alpha_1$ and $\gamma_1$ in steady state. The parameters used in these simulations are shown in Table 2.5.

the feedback regulation of cellular damage level. The results are presented in the following paragraphs.

i. More equal distribution of damage in symmetric renewal and less damage retention in asymmetric division are still favorable segregation rules and lead to larger TD cell populations and greater population ratios.

Similar to the model in Section 2.4.1, damage distribution $\beta_i$ between TD cells does not affect the results much. Since the regulation is stronger when we consider feedbacks from both stem cells and TD cells, the stabilized population of TD cells is smaller than that in Section 2.4.1 under the same parameters. It may not be meaningful to directly compare the sizes of populations in different models, since we do not have real experimental data. However, similar trends are observed: for fixed $\alpha_1$, as damage retention $\gamma_1$ in stem cell increases, both the TD cell population and the
Figure 2.12: Sample population dynamics under different combinations of $\alpha_1$ and $\gamma_1$. The red curve stands for TD cell population and the blue curve for the stem cell population. The parameters used in these simulations are shown in Table 2.5. To eliminate the influence of the initial condition, perturbation of reducing population of TD cells by 20% is introduced after the populations reach the steady state.
population ratio \( P_S/P_T \) decrease; for fixed \( \gamma_1 \), as damage segregation between stem cells becomes more symmetric, the TD cell population increases, but the population ratio is almost unchanged (Fig. 2.11). It is worth mentioning that the population ratio increases a little bit for each damage segregation configuration, after adding feedbacks from stem cells. This means that the regulation is more efficient, since one needs a smaller stem cell pool generate the same number of TD cells. Another interesting observation quite different from Fig. 2.7A is that when \( \alpha = 0.5 \), the cluttering phenomena do not appear for populations of TD cells with \( \gamma_1 = 0.1, 0.3, 0.5 \). Less damage retention shows great advantage to have larger TD cell population, when stem cells distribute damage equally in symmetric self-renewal (Fig. 2.11A). This advantage does not exist, if feedbacks are solely from TD cells.

Two other notable consequences of adding feedbacks from stem cells are that the oscillations in population dynamics disappear (even when \( \gamma_1 \) is large) and that the population overshooting problem resolves when \( \alpha_1 \) is small (Fig. 2.12).

ii. Adding feedbacks from stem cells promotes stem cell health by slowing down cell cycle progression of stem cells with high damage and hence increasing their death rates.

Compared to Section 2.4.1 the death rate of TD cells does not change much and ranges from \( 2.130 \times 10^{-4} \) to \( 2.942 \times 10^{-4} \). But the death rate of stem cells shown in Table 2.6 in each segregation configuration increases a bit, compared to the data in Table 2.2. One reason is that the feedbacks from cellular damage level slow down the cell cycle progression speed of stem cells with high level of damage and the prolonged cell cycle length results in more damage accumulation and leads to increased death of this kind. It may not be a bad mechanism to have a greater death rate of such stem
cells, since this will increase the percentage of stem cells with low level of damage and promote the overall health of the stem cell pool.

iii. Including feedbacks from stem cell cellular damage level improves the health of the stem cell population by increasing the percentage of stem cells with low damage.

Since the feedbacks from stem cells described by (2.22) and (2.23) also depend on stem cell cellular damage level, we want to examine the effect of such regulations on damage distribution at the end of cell cycle by comparing the models in Sections 2.4.1 and 2.4.2 (Fig. 2.9 and Fig. 2.13). Similar to Section 2.4.1, the damage distributions at the end of cell cycle become more concentrated, as the degree of symmetry in damage segregation increases. However, compared to the model in Section 2.4.1, stem cells in this subsection have better fitness, since under same segregation rules, there are more cells with less damage at the end of cell cycle in steady state. This effect becomes most significant when the segregation rule is symmetric, taking Fig. 2.9D2 and Fig. 2.13D2 as examples. This result suggests that slowing down the cell

Table 2.6: Death rate of stem cells in steady state under different segregation rules.
cycle progression of stem cells with high level of damage and promoting such stem cells to differentiate indeed can improve the overall health of the stem cell population.

In conclusion, more equal distribution of damage between stem cells in symmetric renewal and less damage retention in stem cell in asymmetric division are still favorable segregation rules resulting in higher population size, greater population ratio and lower death rate of stem cells, when both the feedback regulations from TD cells and stem cells are present. However, adding feedback regulations from stem cells can reduce oscillations and population overshooting in some situations with unfavorable damage segregation rules, say, in the situation where $\alpha_1$ is small and $\gamma_1$ is large. Besides that, including feedbacks from stem cell cellular damage level can improve the health of the stem cell population by increasing the percentage of stem cells with low level of damage.

2.5 Conclusions and discussions

In this chapter, we proposed three models to study damage segregation in stem cells without and with different regulation mechanisms. This is the first time to use the continuous approach to study damage accumulation aging theory in stem cells. Our models integrate stem cell renewal and differentiation with damage accumulation and segregation. In the first model, we did not include any regulation. With mathematical analysis and numerical simulations, we found that stem cell population either blows up or becomes extinct for most choices of parameters. In the second model, we included the inhibitions from TD cells on stem cell proliferation and symmetric divisions and studied the influence of segregation rules on population dynamics. We
Figure 2.13: Population density and damage distribution at the end of cell cycle for stem cells. The damage segregation rules in cases A-D are shown in Table 2.4. All the other parameters are in Table 2.1 and Table 2.5. The damage distributions are re-scaled for comparison. Continued.
found that less damage retention (smaller $\gamma_1$) and more equal distribution of damage between stem cells ($\alpha_1$ tends to be 0.5) might be favorable segregation rules which result in greater TD cell population and population ratio, smaller stem cell death rate, more asymmetric stem cell division in steady state, and faster and more stable convergence to steady state. One unique feature of our PDE models is that they allow us to discover the advantage of unequal distribution of damage between stem cells that cannot be seen from the analysis of total population which is an integrated result. We found that with less equal distribution of damage between stem cells, the population distributions are less concentrated and have more portions in lower damage region, which increases the resistance of sudden damage elevation and enhances system stability. In the third model, besides the regulation from TD cells, we added feedbacks from stem cells and cellular damage level. In this model, excessive stem cell population would induce inhibition to slow down stem cell proliferation and reduce stem cell symmetric renewal and differentiation; and the stem cells with higher level damage have slower cell cycle progression and reduced tendency to self-renew.
Compared to the model with regulation solely from TD cells, two improvements are that adding new regulations reduces population overshooting and oscillations and also shifts the damage distribution curve to the low damage region.

Our models have some limitations. The first limitation is that parameters in Table 2.1 and Table 2.5 are not estimated from real data due to lack of experiments. Different kinds of stem cells have different dynamics and mechanisms to cope with damage. For example, experiments observed that, intestine stem cells of adult flies segregate damage to differentiated progenies to protect the stem cell pool, which needs to be maintained to replenish short-lived intestine cells; while germline stem cells and neuroblasts tend to retain damage during division, since the differentiated daughters give rise to new organism [62]. Hence, to better understand the mechanisms of how stem cells cope with damage accumulation, it is better to choose the specific type of stem cells and work with real experiments. The second limitation is that our model only contains two compartments. In reality, stem cells have a small population size and mainly stay quiescent; and the progenitors divide fast and replenish TD cells [102, 103]. In the future, we may divide the stem cell compartment into two compartments and consider a three-compartment model. With three compartments, more regulations and damage segregation mechanisms can be considered.

In conclusion, our two-compartment PDE models of transport type are a brand new attempt to study the mechanisms of stem cell coping with damage accumulation in the aging process. Despite the limitations mentioned above, our models discussed the advantages and disadvantages of different damage segregation rules without or with feedback regulations. We believe that with more available data in the future, our models can be better modified to serve as a tool to explore stem cell aging.
Appendix A: Supporting Information for Chapter 1

A.1 Estimation of parameters

The parameters in the survival fraction in Eq. (2), the random budding probability in Eq. (3), the cell sizes in Eqs. (4) and (5) and the cell cycle length in Eq. (6) are estimated using existing biological data in references [1, 2, 3, 4]. The estimated curves are shown in Fig. A.1.
Figure A.1: (A) Estimated survival fractions using Eq. (2). The blue curve for haploid cells is estimated through Fig. 1 in [1]. Since there is no available data for diploid cells, we simply estimated it to be the red curve based on the fact that diploid cells are longer-lived than haploid cells. (B) Estimated random budding probability using Eq. (3). The curve for haploid cells is estimated through Fig. 3 in [2]. Due to lack of data, we use the same curve for diploid cells. (C) Estimated cell size using Eqs. (4) and (5) and the data in [3, 4]. (D) Estimated cell cycle length using Eq. (6) and the data in [3, 4].
Apart from the above estimated parameters shown in Fig. A.1 we selected the other parameters based on reasonable assumptions, while the experimental data are lacking. All the parameters are given in the Table A.1.
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Table A.1: Parameters used in simulations and their references.
A.2 A sample colony generated by a single haploid cell

Due to mating type switch, a single homothallic haploid cell will generate a colony with a mixed population, which contains both diploid $\alpha/\alpha$ cells and haploid cells of $a$ and $\alpha$ types. A sample simulation is shown in Fig. A.2.
Figure A.3: Minimal covering circles for diploid colonies. (A) A sample colony of bipolar budding diploid cells. (B) A sample colony of random budding diploid cells.

A.3 Demonstration of minimal covering circles

The minimal covering circle, which is the minimal circle covering the colony, is introduced to measure colony expansion and sparseness in Section 4.3 in the main text. Two sample colonies of diploid cells, budding bipolarly and randomly, are shown in Fig. A.3.
A.4 Samples of bipolar/random budding colonies under rich/poor nutrient conditions

Colonies generated by diploid cells which bud bipolarly and randomly show different colony morphologies under different nutrient conditions. Our simulations suggest that only bipolar budding colonies give rise to a colony morphology with finger-like extensions under limited nutrient $U_0 = 1$. Bipolar budding colonies under rich nutrient $U_0 = 2$ or random budding colonies in any nutrient condition do not show extended branches. This phenomenon is seen in most of the simulations, and Fig. A.4 shows some sample simulations.
Figure A.4: Samples of bipolar and random budding colonies with 150 cells under rich ($U_0 = 2$) and poor ($U_0 = 1$) nutrient conditions.
A.5 Numerical methods

A.5.1 Dynamics of agents

In our model, cells can grow, divide, die and mate according to rules defined in
Section 2 in the main text. Each of these processes changes the configuration of
the cells. In this chapter, we use the following computational scheme to update the
agents:

1. Initialize the configuration of cells (age, size, type, position, cell cycle length
   and budding angles) and record all these variables.

2. Set time $t = 0$ and choose a sufficiently small time step $\Delta t$.

3. Advance one time step. Label all dead cells and exclude them from further bio-
   logical processes. Update the nutrient field and the biological features affected
   by the field for each cell.

4. For the live cells, at the end of a cell cycle:

   (a) New cells are born from the cells that just have finished one cell cycle; for
       the mother cells, increase their replicative age and update the list of cell
       features according to the defined mathematical rules; for newborn cells,
       set up their initial configurations and record mother-daughter relations.

   (b) For haploid cells, check the possibility of mating type switch.

5. In a haploid colony, remove mated haploid cells, create the new diploid cell and
   set up its initial configuration.

6. Rearrange cell position based on cell-cell repulsive and adhesive forces.
7. Calculate and record needed statistical results every 1.5 hours.

8. Repeat 3–7 until certain stopping criterion (maximum time or maximum population size) is reached.

### A.5.2 Numerical scheme solving the evolution of the nutrient field

The nutrient field is defined across a square mesh of points. During each time step, nutrient on each grid point is consumed by nearby cells and diffuses over the domain. The method of lines is applied for the discretization of the nutrient equation (8). Second order central difference is used for approximating the Laplace operator and the forward Euler method is applied for solving the temporal evolution of the system. The discretized nutrient equation is

\[
\frac{u_{i,j}^{n+1} - u_{i,j}^n}{\Delta t} = D \left( \frac{u_{i+1,j}^n - 2u_{i,j}^n + u_{i-1,j}^n}{\Delta x^2} + \frac{u_{i,j+1}^n - 2u_{i,j}^n + u_{i,j-1}^n}{\Delta y^2} \right) - c_r \sum_{k=1}^{N(t)} e^{-\frac{\sqrt{(a_k-x_i)^2+(b_k-y_j)^2}}{c_d}} u_{i,j}^n,
\]  

(A.1)

where \((a_k, b_k) = \vec{x}_k\) is the location of the \(k\)-th cell. The initial condition is \(u(x, y, 0) = U_0\). No-flux conditions \(\frac{\partial u}{\partial x} = 0, \frac{\partial u}{\partial y} = 0\) are imposed on the boundary of the domain. Our calculations are carried out by MATLAB.
Appendix B: Supporting Information for Chapter 2

B.1 Derivation of Equation (2.12)

After a change of variable, (2.10) together with initial condition and boundary condition (2.8) can be solved by the method of characteristics. For the first cell cycle, i.e. $0 < t < t_p$, we have three cases:

1. If $t \leq p$ and $t \leq d$, the solution is determined by the initial condition

$$S(t, p, d) = S(0, p - t, d - t); \quad \text{(B.1)}$$

2. If $t > d$ and $d < p$, the solution is determined by the boundary condition on $d = 0$

$$S(t, p, d) = S(t - d, p - d, 0); \quad \text{(B.2)}$$

3. If $t > p$ and $p < d$, the solution is determined by boundary condition on $p = 0$

$$S(t, p, d) = S(t - p, 0, d - p). \quad \text{(B.3)}$$

Considering the biological background, we assume that $t_p < t_d$. Applying the boundary condition (2.8), the solution $S(t, p, d)$ can be expressed as
\[ S(t, p, d) = \begin{cases} 
S(0, p - t, d - t) & \text{if } t \leq p \text{ and } t \leq d \\
0 & \text{if } t > d \text{ and } d < p \\
H(t, p, d, \alpha_1, t_d) \frac{\delta_1}{\alpha_1} S\left(0, t_p - (t - p), \frac{d - p}{\alpha_1} - (t - p)\right) & \text{if } t > p \text{ and } p < d, \\
+ H(t, p, d, \alpha_2, t_d) \frac{\delta_1}{\alpha_2} S\left(0, t_p - (t - p), \frac{d - p}{\alpha_2} - (t - p)\right) & \\
+ H(t, p, d, \gamma_1, t_d) \frac{\delta_3}{\gamma_1} S\left(0, t_p - (t - p), \frac{d - p}{\gamma_1} - (t - p)\right) &
\end{cases}
\] (B.4)

where the function \( H \) is given by

\[ H(t, p, d, \omega, t_d) = \begin{cases} 
1 & \text{if } \frac{d - p}{\omega} < t_d \text{ and } \frac{d - p}{\omega} - (t - p) > 0; \\
0 & \text{otherwise.}
\end{cases} \] (B.5)

For the time that is greater than one cell cycle, i.e. \( t > t_p \), we can trace back the process of damage protein synthesis by one cycle, which is given the following formula

\[ S(t, p, d) = H(t, p, d, \alpha_1, t_d) \frac{\delta_1}{\alpha_1} S\left(t - t_p, p, \frac{d - p}{\alpha_1} - (t_p - p)\right) + H(t, p, d, \alpha_2, t_d) \frac{\delta_1}{\alpha_2} S\left(t - t_p, p, \frac{d - p}{\alpha_2} - (t_p - p)\right) + H(t, p, d, \gamma_1, t_d) \frac{\delta_3}{\gamma_1} S\left(t - t_p, p, \frac{d - p}{\gamma_1} - (t_p - p)\right). \] (B.6)

Applying Eq. (B.4) and (B.6) iteratively, we can obtain the density of the stem cell population at any time.

**B.2 Derivation of the limit damage band**

Suppose a stem cell currently has damage level \( d^0 \) at the beginning of some cell cycle. We want to record the damage in the cycles that follow. At the end of the first cycle, the damage accumulates to \( d^0 + t_p \). After division, a stem daughter cell inherits damage \((d^0 + t_p)\ell_1 \) with \( \ell_1 \in \{ \alpha_1, \alpha_2, \gamma_1 \} \), whose damage gets to \((d^0 + t_p)\ell_1 + t_p = d^0\ell_1 + t_p(\ell_1 + 1)\) at the end of the second cycle. After division, a stem daughter
cell inherits damage \( (d^0 \ell_1 + t_p(\ell_1 + 1))\ell_2 \) with \( \ell_2 \in \{\alpha_1, \alpha_2, \gamma_1\} \), and the damage accumulates to \( (d^0 \ell_1 + t_p(\ell_1 + 1))\ell_2 + t_p = d^0 \ell_1 \ell_2 + t_p(\ell_1 \ell_2 + \ell_2 + 1) \) at the end of the third cycle. By induction, one easily sees that the damage in a stem daughter cell (if not dead yet) at the end of the \( m \)th cycle is

\[
d^0 \prod_{i=0}^{m-1} \ell_i + t_p \prod_{i=0}^{m-1} \ell_k \quad \text{with} \quad \ell_0 = 1 \quad \text{and} \quad \ell_i \in \{\alpha_1, \alpha_2, \gamma_1\} \text{ for } i \geq 1.
\]

Let \( \omega_1 \) and \( \omega_2 \) be the minimum and the maximum of \( \alpha_1, \alpha_2, \gamma_1 \), respectively. In the above, considering a fixed segregation portion \( \ell_i = \omega_j \) for all \( i \geq 1 \), we get the damage level

\[
d^0 \omega_j^{m-1} + t_p \frac{1 - \omega_j^m}{1 - \omega_j}, \quad j = 1, 2.
\]

The limits as \( m \to \infty \) yields the limit damage band

\[
\left[ \frac{t_p}{1 - \omega_1}, \frac{t_p}{1 - \omega_2} \right].
\]

**B.3 Proof of Proposition 2.1**

First suppose \( f_r < 1/2 \). Consider any fixed time \( t \gg t_p \). By (B.6), we have

\[
0 \leq S(t + t_p, p, d) \leq \frac{\delta_1}{\alpha_1} \left( t, p, \frac{d - p}{\alpha_1} - (t_p - p) \right) + \frac{\delta_1}{\alpha_2} \left( t, p, \frac{d - p}{\alpha_2} - (t_p - p) \right) + \frac{\delta_3}{\gamma_1} \left( t, p, \frac{d - p}{\gamma_1} - (t_p - p) \right).
\]

Then by integration and substitutions

\[
0 \leq P_S(t + t_p) = \int_0^{t_p} \int_0^{t_d} S(t + t_p, p, d) \, dd \, dp \\
\leq (\delta_1 + \delta_1 + \delta_3) \int_0^{t_p} \int_0^{t_d} S(t, p, d) \, dd \, dp \\
= (2f_r) P_S(t).
\]
By the continuity of $P_S(t)$, it attains a maximum $P_{\text{max}}$ in a time interval $[t_0, t_0 + t_p]$ for a fixed $t_0$. Repeated application of the above inequality, in view of the arbitrariness of $t$, implies that for any $n \geq 1$, and for any $t$ in $[t_0 + nt_p, t_0 + (n+1)t_p]$, we have

$$P_S(t) \leq (2f_r)^n P_{\text{max}},$$

which clearly establishes that $\lim_{t \to \infty} P_S(t) = 0$.

Next suppose the limit damage band exceeds the lethal threshold, i.e. $t_d < \frac{t_p}{1-\omega_1}$. The stem cell population always diminishes to zero, since after sufficiently long time the offsprings of stem cells with initial zero damage, even in the slowest way of accumulating damage, i.e. $\ell_i = \omega_1$ for every $i$, would gain damage to reach $t_d$ and die, and other offsprings in the evolution gain more damage and would die sooner. This result is true, no matter what the proportions of three division types are.

### B.4 Proof of Proposition 2.2

By the assumption that $t_d \geq \frac{t_p}{1-\omega_2}$, no cell will die due to damage accumulation and $f_r$ completely determines the evolution of stem cell population. If $f_r > 1/2$, the portion of generated stem cells through divisions is greater than 1, which means that the stem cell population is increasing to infinity. Indeed, for any $t$, let $\omega \in \{\alpha_1, \alpha_2, \gamma_1\}$, by the definition of $H$ and change of variables,

$$I(\omega) := \frac{1}{\omega} \int_0^{t_p} \int_0^{\omega^{2t_d+p}} H(t_p, p, d, \omega, t_d) S\left(t - t_p, p, \frac{d-p}{\omega} - (t_p - p)\right) dd dp$$

where

$$I(\omega) = \frac{1}{\omega} \int_0^{t_p} \int_0^{\omega^{t_d+p}} S\left(t - t_p, p, \frac{d-p}{\omega} - (t_p - p)\right) dd dp$$

$$= \int_0^{t_p} \int_0^{\omega^{t_d-p+p}} S\left(t - t_p, p, d\right) dd dp$$

$$\geq \int_0^{t_p} \int_0^{\omega^{2t_d+p}} S\left(t - t_p, p, d\right) dd dp$$

(B.7)
By (B.6) and (B.7),

\[
P_S(t) \geq \int_0^{t_p} \int_0^{\omega_2 t_d + p} S(t, p, d) \, dd \, dp
\]

\[
= \delta_1 I(\alpha_1) + \delta_1 I(\alpha_2) + \delta_3 I(\gamma_1)
\]

\[
\geq 2f_r \int_0^{t_p} \int_0^{\omega_2 t_d + p} S(t - t_p, p, d) \, dd \, dp
\]

By repeating this procedure, if \( t = nt_p + \tilde{t} \) and \( \tilde{t} \in [0, t_p) \), then we have

\[
P_S(t) \geq (2f_r)^n \int_0^{t_p} \int_0^{\omega_2 t_d + p} S(\tilde{t}, p, d) \, dd \, dp
\]

If

\[
\int_0^{t_p} \int_0^{\omega_2 t_d + p} S(0, p, d) \, dd \, dp > 0
\]

then the population will blow up.

Intuitively, after sufficient long time, there is no death by damage accumulation since \( t_d \geq \frac{t_p}{1 - \omega_2} \). Hence in the situation that \( f_r = 1/2 \) one of the offsprings after stem cell division remains a stem cell and the stem cell population is conserved.

**B.5 Proof of Proposition 2.3**

We assume that (2.17) holds:

\[
f_r > \frac{1}{2} \text{ and } \frac{t_p}{1 - \gamma_1} \leq \frac{t_p}{1 - \alpha_1} < t_d < \frac{t_p}{1 - \alpha_2},
\]

and remark that Fig. B.1 can be helpful to visualize some part of the proofs below.

We first prove (a). For integers \( m \geq 1 \), define

\[
n(m) = \min \left\{ n \in \mathbb{N} : t_d < \frac{1 - \gamma_1^m/t_p \alpha_2^n}{1 - \gamma_1} + \frac{1 - \alpha_2^n}{1 - \alpha_2} t_p \right\}.
\]
Figure B.1: In the simple model, the cell lineage can be modeled through discrete process.

Let $P_S(m t_p)$ denote the stem cell population at the end of the $m$th cycle, when the damage in cells is bounded from below by $d_m = \frac{1 - \gamma^m}{1 - \gamma_1} t_p$. The damage in stem daughter cells (if not dead yet) after $n$ cycles would be greater than

$$d_m \prod_{i=0}^{n-1} \ell_i + t_p \sum_{i=0}^{n-1} \prod_{k=i}^{n-1} \ell_k \quad \text{with } \ell_0 = 1 \text{ and } \ell_i \in \{\alpha_1, \alpha_2, \gamma_1\} \text{ for } i \geq 1. \quad (B.8)$$

Then at the end of the $(m+n(m))$th cycle, i.e. taking $n = n(m)$ in $\text{(B.8)}$, if $\ell_i = \alpha_2$ for all $0 \leq i \leq n(m)$, the cells with damage in $\text{(B.8)}$ would die, in view of the condition in the definition of $n(m)$.

Thus, for the possible live cells at the end of the $(m+n(m))$th cycle, there should be at least one $\ell_i = \gamma_1$ or $\alpha_1$ for $1 \leq i \leq n(m)$. Clearly the proportion of such stem daughter cells in the population is

$$(2 f_r)^n(m) - \delta_1^n(m).$$

Suppose there exists a combination of $m$ and $\delta_i$ such that $(2 f_r)^n(m) - \delta_1^n(m) < 1$. It is easy to see the sequence $\{(2 f_r)^n - (\delta_1)^n\}$ is increasing in $n$, under the assumption
Also it is clear that the sequence \( \{n(m)\} \) is non-increasing in \( m \), i.e. \( n(m + 1) \leq n(m) \). Hence

\[
P_S((m + n(m))t_p) \leq P_S(mt_p)((2f_r)^{n(m)} - \delta_1^{n(m)}).
\]

Then applying the above inequality with \( m \) replaced by \( m + n(m) \), one gets

\[
P_S((m + n(m) + n(m + n(m)))t_p) \\
\leq P_S((m + n(m))t_p)((2f_r)^{n(m+n(m))} - \delta_1^{n(m+n(m))}) \\
\leq P_S((m + n(m))t_p)((2f_r)^{n(m)} - \delta_1^{n(m)}) \\
\leq P_S(mt_p)((2f_r)^{n(m)} - \delta_1^{n(m)})^2
\]

Repeating this argument yields a sequence of population decreasing to zero. Therefore (a) is proven, as we consider stem cells to be extinct once the population of stem cells drops below a certain small number.

Next we work with (b) and suppose \( \delta_2 = 0 \). Let \( N_0(m) \) denote the number of live stem cells at the end of the \( m \)th cycle whose damage segregation history contains no \( \alpha_2 \), and \( N_1(m) \) the number of live stem cells whose damage segregation history contains exactly one \( \alpha_2 \). First we note that \( \{N_0(m)\} \) is a constant sequence, in view of the definition of \( N_0(m) \) and that \( \delta_1 + \delta_3 = 1 \). Also \( N_1(m+1) = \delta_1 N_0(m) + N_1(m) = \delta_1 N_0(m) + \delta_1 N_0(m-1) + N_1(m-1) = 2\delta_1 N_0(1) + N_1(m-1) = \cdots = (m-1)\delta_1 N_0(1) + N_1(1) \). If the initial population is nonzero, then \( N_0(1) > 0 \) and we obtain a strictly increasing subpopulation of stem cells, from which (b) follows.

### B.6 Parameters in the model with feedback from TD cells

Although there is no concrete biological data in stem cell division types or specific regulation mechanisms to our knowledge, we discuss the selection of parameters in
Table 2.1 referring to both the biological observations [85, 86, 87, 88] and the previous mathematical models [72, 74, 73, 78, 79, 75, 80, 77, 81, 95, 82, 83, 96]. In this part, we assume that damage segregates symmetrically between stem cells and between TD cells, i.e. $\alpha_1 = 0.5, \beta_1 = 0.5$, while the damage retention in asymmetric division is low, i.e. $\gamma_1 = 0.25$.

It is generally accepted that the population of mitotic cells is much smaller than that of terminally differentiated cells, although it is very difficult to identify and count stem cells and progenitors [98, 99, 100, 101]. Due to lack of experimental data, we consult some existing modeling works for a reasonable range for the ratio of mitotic cells vs TD cells. In the simulations of [81, 96], populations of post-mitotic cells are about 4-10 folds of that of mitotic cells.

Many works suggest that regulation parameters $k_t^i$ and $k_T^v$ are closely related to the TD cell population in steady state [78, 75, 77, 72]. These modeling works mainly focus on the blood system, which is one of the most well studied systems and serves as a paradigm for understanding stem cells. The regulation parameter $k$ in those models ranges from $10^{-7}$ to $10^{-10}$, which corresponds to red blood cell population of size $10^7 \sim 10^{10}$. However, some other works that do not specify the type of stem cells may choose some other ranges. For example, [81] chose $10^{-3}$ for regulation constant and the resulted population magnitude is about $10^3$. In our simulations, we choose $10^{-8}$ as the scale for the magnitudes of $k_t^i$ and $k_T^v$.

On the other hand, mitotic cells are assumed to have a smaller death rate than post-mitotic cells [56]. However, there is no precise description of death rates due to two reasons: the death rate of stem cells is not observable, and the death rate of TD cells is tissue specific. In various modeling works, the assumed death rate of

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TD cells ranges from $10^{-1}$ to $10^{-4}$ of the total population \cite{72, 74}. And analysis and simulations in \cite{72, 74, 73} suggest that a large death rate of TD cells result in instability of population. In our model, the death rates are measured in the following way:

\begin{align}
    r_S &= \int_0^{p^*} \frac{v_d S(t, d^*, p)}{P_S} \, dp \\
    r_T &= \int_0^{\infty} \frac{u_d T(t, d^*, p)}{P_T} \, dp
\end{align}

(B.9) \hspace{1cm} (B.10)

where the numerators are population out-fluxes due to death and the denominators are total populations. By properly choosing $v_p, v_d, u_d$, we wish to achieve that $P_T/P_S$ is no less than 5, $r_T$ is about $10^{-4}$, and $r_S$ is reasonably small, compared to $r_T$.

It was observed that stem cells mainly undergo asymmetric renewal and differentiation when stem cell pool is expanding or when TD cells suffer great loss, and enter into an inactively dividing stage and then mainly undergo asymmetric division in the steady state \cite{85, 86, 87, 88}. These biological obeservations can be used in the following two aspects in our model.

On the one hand, since the feedbacks we consider are negative regulations, $\delta_1^0$ provides the maximum fraction of symmetric renewal. By the property of the Hill function, $\delta_1(P_T)$ can switch between large and small values in response to small and large populations of TD cells. But as we increase the initial $\delta_1^0$, population overshoot and oscillations appear before convergence (Fig. B.2). Moreover, the population ratio $P_T/P_S$ decreases and death rates of stem cell increases as $\delta_1^0$ increases (Table B.1).
Figure B.2: Comparison of different initial division fractions. The initial division fractions are listed in Table B.1. The other parameters are $k_1^{T} = 10^{-8}, k_2^{T} = 0.5 \times 10^{-8}, k_3^{T} = 0.5 \times 10^{-8} \alpha_1 = 0.5, \beta_1 = 0.5, \gamma_1 = 0.25, v_p = 0.2, v_d = 0.06, u_d = 0.02, m_T = 2$. To eliminate the influence of initial condition, perturbation of reducing population of TD cells by 20% is considered after population research steady state.

<table>
<thead>
<tr>
<th>Cases</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\delta_1^{0}$</td>
<td>0.6</td>
<td>0.7</td>
<td>0.8</td>
</tr>
<tr>
<td>$\delta_2^{0}$</td>
<td>0.3</td>
<td>0.2</td>
<td>0.1</td>
</tr>
<tr>
<td>$\delta_3^{0}$</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
</tr>
<tr>
<td>Dynamics</td>
<td>Fig. B.2A</td>
<td>Fig. B.2B</td>
<td>Fig. B.2C</td>
</tr>
<tr>
<td>$P_T/P_S$</td>
<td>5.664</td>
<td>4.241</td>
<td>3.622</td>
</tr>
<tr>
<td>$r_s$</td>
<td>$1.191 \times 10^{-7}$</td>
<td>$7.021 \times 10^{-5}$</td>
<td>$1.365 \times 10^{-4}$</td>
</tr>
<tr>
<td>$r_T/r_s$</td>
<td>1979.1</td>
<td>3.311</td>
<td>1.738</td>
</tr>
<tr>
<td>$\delta_1$</td>
<td>0.20</td>
<td>0.16</td>
<td>0.17</td>
</tr>
<tr>
<td>$\delta_2$</td>
<td>0.20</td>
<td>0.11</td>
<td>0.05</td>
</tr>
<tr>
<td>$\delta_3$</td>
<td>0.60</td>
<td>0.73</td>
<td>0.78</td>
</tr>
</tbody>
</table>

Table B.1: Comparison of initial division fractions.
On the other hand, these biological observations also suggest choosing appropriate regulation parameters $k_T^1$, $k_T^2$, and $k_v^T$ in Eq. (2.20) and Eq. (2.21) such that asymmetrical division will predominate in three types division of stem cells in the steady state. In our simulations, we found that equal strengths of feedback on stem cell symmetric renewal $\delta_1$ and cell cycle progression $V_p$ would result in oscillations (Fig. B.3A and C). Stronger regulation on $V_p$ relative to $\delta_1$ is needed to obtain stabilized population evolution (Fig. B.3B and D). We also found that the relative strengths of regulation on symmetric renewal $\delta_1$ and symmetric differentiation $\delta_2$ play a role in determining the death rate of stem cells. To obtain a reasonably small death rate of stem cells, stronger regulation on $\delta_2$ is also needed (Table B.2).

The modeling work [73] suggests that a low Hill exponent in the feedback is needed to avoid oscillations, where the authors observed unstable steady states for Hill exponent $m \geq 3$ through direct simulations in their ODE models. We also found

<table>
<thead>
<tr>
<th>Cases</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>D</th>
</tr>
</thead>
<tbody>
<tr>
<td>$k_T^1$</td>
<td>$10^{-8}$</td>
<td>$10^{-8}$</td>
<td>$0.5 \times 10^{-8}$</td>
<td>$10^{-8}$</td>
</tr>
<tr>
<td>$k_T^2$</td>
<td>$10^{-8}$</td>
<td>$10^{-8}$</td>
<td>$0.5 \times 10^{-8}$</td>
<td>$0.5 \times 10^{-8}$</td>
</tr>
<tr>
<td>$k_v^T$</td>
<td>$10^{-8}$</td>
<td>$0.5 \times 10^{-8}$</td>
<td>$0.5 \times 10^{-8}$</td>
<td>$0.5 \times 10^{-8}$</td>
</tr>
<tr>
<td>Dynamics</td>
<td>Fig. B.3A</td>
<td>Fig. B.3B</td>
<td>Fig. B.3C</td>
<td>Fig. B.3D</td>
</tr>
<tr>
<td>$r_S$</td>
<td>-</td>
<td>$2.408 \times 10^{-4}$</td>
<td>-</td>
<td>$1.191 \times 10^{-7}$</td>
</tr>
<tr>
<td>$r_T/r_S$</td>
<td>-</td>
<td>3.119</td>
<td>-</td>
<td>1979.1</td>
</tr>
<tr>
<td>$\delta_1$</td>
<td>-</td>
<td>0.13</td>
<td>-</td>
<td>0.20</td>
</tr>
<tr>
<td>$\delta_2$</td>
<td>-</td>
<td>0.07</td>
<td>-</td>
<td>0.20</td>
</tr>
<tr>
<td>$\delta_3$</td>
<td>-</td>
<td>0.80</td>
<td>-</td>
<td>0.60</td>
</tr>
</tbody>
</table>

Table B.2: Comparison of regulation parameters.
Figure B.3: Comparison of different regulation parameters. The values of $k^T_1$, $k^T_2$, $k^T_v$ are listed in Table B.2. The other parameters are $\alpha_1 = 0.5$, $\beta_1 = 0.5$, $\gamma_1 = 0.25$, $v_p = 0.2$, $v_d = 0.06$, $u_d = 0.02$, $\delta^0_1 = 0.6$, $\delta^0_2 = 0.3$, $m_T = 2$. 

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similar phenomena in our model, where oscillations appear in the simulations with \( m_T > 4 \). Thus we need \( m_T \in \{1, 2, 3\} \). To make the best estimation of \( m_T \), we also need to consider all biological observations. Our simulations show that when \( m_T = 1 \) the death rate of stem cell is only about one third of that of TD cells (Table B.3), while when \( m_T = 3 \) there is population overshoot before convergence to steady state although the death rate of stem cell decreases to \( 10^{-15} \) (Table B.3). Hence we choose \( m_T = 2 \) in our model.

After a large number of trials, we arrive at the best choice of the parameters in Table 2.1 with consideration of all biological and mathematical reasons. Under these parameters, the resulted death rates are \( r_S = 1.191 \times 10^{-7} \) and \( r_T = 2.357 \times 10^{-4} \). The populations in steady state are \( P_S = 2.496 \times 10^7, P_T = 1.414 \times 10^8 \) with the ratio \( P_T/P_S = 5.664 \). But we need to point out that all our assumptions are pieced together

### Table B.3: Comparison of Hill exponents.

<table>
<thead>
<tr>
<th>Cases</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>( m_T )</td>
<td>Fig. B.4A</td>
<td>Fig. B.4B</td>
<td>Fig. B.4C</td>
</tr>
<tr>
<td>Dynamics</td>
<td>( 1.515 \times 10^8 )</td>
<td>( 1.414 \times 10^8 )</td>
<td>( 1.101 \times 10^8 )</td>
</tr>
<tr>
<td>( P_T )</td>
<td>4.116</td>
<td>5.664</td>
<td>7.519</td>
</tr>
<tr>
<td>( P_T/P_S )</td>
<td>( 9.586 \times 10^{-5} )</td>
<td>( 1.191 \times 10^{-7} )</td>
<td>( 1.686 \times 10^{-15} )</td>
</tr>
<tr>
<td>( r_S )</td>
<td>( 2.371 \times 10^{-4} )</td>
<td>( 2.357 \times 10^{-4} )</td>
<td>( 2.282 \times 10^{-4} )</td>
</tr>
<tr>
<td>( r_T )</td>
<td>( 2.474 )</td>
<td>( 1979.1 )</td>
<td>( 1.35 \times 10^{11} )</td>
</tr>
<tr>
<td>( r_T/r_S )</td>
<td>0.24</td>
<td>0.20</td>
<td>0.26</td>
</tr>
<tr>
<td>( \delta_1 )</td>
<td>0.17</td>
<td>0.20</td>
<td>0.26</td>
</tr>
<tr>
<td>( \delta_2 )</td>
<td>0.59</td>
<td>0.60</td>
<td>0.48</td>
</tr>
</tbody>
</table>
Figure B.4: Comparison of different Hill exponents. The value of the parameters: 
\( k_T^1 = 10^{-8}, k_T^2 = 0.5 \times 10^{-8}, k_v^T = 0.5 \times 10^{-8} \alpha_1 = 0.5, \beta_1 = 0.5, \gamma_1 = 0.25, v_p = 0.2, v_d = 0.06, u_d = 0.02, \delta_1^0 = 0.6, \delta_2^0 = 0.3 \). The Hill exponents in panels A, B and C are \( m_T = 1, 2, 3 \).

through different biological and mathematical researches, more precise estimations of parameters need real experimental data.

B.7 TD cell population and population ratio when \( \beta_1 = 0.1 \) and 0.3, see Fig. B.5

B.8 Estimation of populations in the models with feedbacks from TD cells

First we recall the assumptions: \( d^* \leq d^c \), \( V_d = v_d \), \( U_p = u_p \) and \( U_d = u_d \) are constants, and \( \delta_i \) and \( V_p \) have feedbacks from TD cells described by (2.20) and (2.21).

The populations of stem and TD cells at time \( t \) are give by

\[
P_S(t) = \int_0^{d^*} \int_0^{p^*} S(t, p, d) dpdd \\
P_T(t) = \int_0^{d^c} \int_0^{\infty} T(t, p, d) dpdd.
\]
Figure B.5: TD cell population and population ratio in steady state for different combinations of $\alpha_1$ and $\gamma_1$. 

- $\beta_1 = 0.1$
- $\beta_1 = 0.3$
Under the above assumptions, we integrate (2.1) and (2.2) in $p$ and $d$ to get

\[
\frac{dP_S}{dt} = -v_d \int_0^{p^*} S(t, p, d^*) - S(t, p, 0) \, dp - V_p(P_T) \int_0^{d^*} S(t, p^*, d) - S(t, 0, d) \, dd
\]

(B.13)

\[
\frac{dP_T}{dt} = -u_d \int_0^\infty T(t, p, d^*) - T(t, p, 0) \, dp + u_p \int_0^{d^*} T(t, 0, d) \, dd
\]

(B.14)

By boundary conditions (2.3), (2.4), (2.8), and (2.9), we rewrite the above equations as

\[
\frac{dP_S}{dt} = -v_d \int_0^{p^*} S(t, p, d^*) \, dp - V_p(P_T) \left[ \int_0^{d^*} S(t, p^*, d) \, dd - \frac{\delta_1(P_T)}{\alpha_1} \int_0^{d^*} S(t, p^*, \frac{d}{\alpha_1}) \, dd \right]
\]

\[
- \frac{\delta_1(P_T)}{\alpha_2} \int_0^{d^*} S(t, p^*, \frac{d}{\alpha_2}) \, dd - \frac{\delta_3(P_T)}{\gamma_1} \int_0^{d^*} S(t, p^*, \frac{d}{\gamma_1}) \, dd
\]

\[
\frac{dP_T}{dt} = -u_d \int_0^\infty T(t, p, d^*) \, dp + V_p(P_T) \left[ \frac{\delta_2(P_T)}{\beta_1} \int_0^{d^*} S(t, p^*, \frac{d}{\beta_1}) \, dd \right]
\]

\[
+ \frac{\delta_2(P_T)}{\beta_2} \int_0^{d^*} S(t, p^*, \frac{d}{\beta_2}) \, dd + \frac{\delta_3(P_T)}{\gamma_2} \int_0^{d^*} S(t, p^*, \frac{d}{\gamma_2}) \, dd
\]

Making a change of variable in $d$ and noticing that $d^* \leq d^c$, we obtain a system of ODEs, called I:

\[
\frac{dP_S}{dt} = -v_d \int_0^{p^*} S(t, p, d^*) \, dp + V_p(P_T) \left[ (\delta_1(P_T) - \delta_2(P_T)) \int_0^{d^*} S(t, p^*, d) \, dd \right]
\]

(B.15)

\[
\frac{dP_T}{dt} = -u_d \int_0^\infty T(t, p, d^*) \, dp + V_p(P_T) \left[ (1 + \delta_2(P_T) - \delta_1(P_T)) \int_0^{d^*} S(t, p^*, d) \, dd \right]
\]

(B.16)

Consider a related system II given by the following equation and (B.16):

\[
\frac{dP_S}{dt} = V_p(P_T) (\delta_1(P_T) - \delta_2(P_T)) \int_0^{d^*} S(t, p^*, d) \, dd
\]

(B.17)

Then under the same initial condition, the solution to system I, called system is no larger than that to system II, by comparison theorems for ODEs.

Now we assume that both systems I and II have steady state solutions, denoted by and $(\bar{P}_S, \bar{P}_T)$ and $(\tilde{P}_S, \tilde{P}_T)$, respectively. Then $\tilde{P}_T$ can be obtained by solving

\[
\delta_1(P_T) - \delta_2(P_T) = 0.
\]

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Assuming appropriate values of parameters to guarantee solvability of the last equation, we have the upper-bound estimate

\[
\bar{P}_T \leq \tilde{P}_T = \left( \frac{\delta_1^0 - \delta_2^0}{\delta_2^0 k_1^T m_T - \delta_1^0 k_2^T m_T} \right) \frac{m_T}{n_T}.
\]  

(B.18)

Next we continue to estimate \( \bar{P}_S \). First we observe that in the steady state both \( S(t, p, d) \) and \( T(t, p, d) \) are constant along characteristic curves and refer the reader to Fig. B.6A for an illustration of the situation. Since in system II there is no outflux of stem cells on the boundary \( d = d^* \), we have

\[
\tilde{P}_S = p^* \int_0^{d^*} S(t, p^*, d) \, dd
\]

On the other hand, by the intermediate value theorem for integrals, we have for some \( h \in [h_1, h_2] \) that

\[
\tilde{P}_T = h \int_0^\infty T(t, p, d^*) \, dp
\]

where

\[
h_1 = d^* - \frac{\eta_2}{1 - \omega_2} \Delta d \quad \text{and} \quad h_2 = d^* - \frac{\eta_1}{1 - \omega_1} \Delta d
\]

with \( \eta_1 = \min\{\beta_1, \beta_2, \gamma_2\} \), \( \eta_2 = \max\{\beta_1, \beta_2, \gamma_2\} \) and \( \Delta d = \frac{p^*}{V_p(\tilde{P}_T)} v_d \). Using that \( \tilde{P}_T \) is a stead state population, we have by (B.15)

\[
u_d \tilde{P}_T = h V_p(\tilde{P}_T) \int_0^{d^*} S(t, p^*, d) \, dd = h V_p(\tilde{P}_T) \tilde{P}_S / p^*
\]

and thus the estimate

\[
\bar{P}_S \leq \tilde{P}_S = \frac{\nu_d p^*}{h V_p(\tilde{P}_T)} \tilde{P}_T \leq \frac{\nu_d p^*}{V_p(\tilde{P}_T) h_1} \tilde{P}_T,
\]  

(B.19)

where \( \tilde{P}_T \) is given in (B.18).

Since in Section 2.4.1 we intentionally choose the parameters such that the death rate of stem cells is small, i.e. the outflux \( v_d \int_0^{d^*} S(t, p, d^*) \, dp \) is small, the upper bound
Figure B.6: A. Demonstration of upper bound estimation of the population. B. A sample simulation shows that our population estimation could be very close to the real solution \((\alpha_1 = 0.5, \beta_1 = 0.5, \gamma_1 = 0.25)\) and the other parameters are listed in Table 2.1 and Table 2.5.

in \((B.18)\) is very close to the real TD cell population in the steady state. However, the estimation for stem cells may not be sharp, due to the variation of the segregation rules. As an example, with the parameters in Table 2.1 we have that the estimated populations are \((\bar{P}_S, \bar{P}_T) = (\frac{6}{13} \sqrt{2} \times 10^7, \sqrt{2} \times 10^8)\) and the steady state populations in the simulation are \((\bar{P}_S, \bar{P}_T) = (2.496 \times 10^7, 1.414 \times 10^8)\) (Fig. B.6B).

B.9 Parameters in the model with feedbacks from TD cells and stem cells

Inspired by the analysis in Section B.6, we selected the most reasonable parameters in Table 2.5 based on both biological and mathematical reasons as follows.

Similar to Section B.6, due to the consideration of reasonable population ratio and death rate of stem cells, we still assume that the feedbacks from stem cell population on cell cycle progression \(V_p\) and symmetric differentiation \(\delta_2\) are stronger than that
on symmetric renewal $\delta_1$. That is, we intentionally choose smaller $k^S_v, k^S_2$, compared to $k^S_1$. And the Hill exponent $m_S$ is chosen to be 2.

To decide the correct magnitude of $k^S_1$, we simulate the population evolution with $k^S_1$ ranging from $10^{-6}$ to $10^{-8}$. We found that the relative magnitude of $k^T_1$ and $k^S_1$ has little influence on the population ratio. To effectively model the regulation from the stem cell population, $10^{-7}$ is the best choice, according to our simulations under the choice of parameters in Table 2.1.

According to experiments, mitotic cells inheriting more damage protein aggregates have an increased cell cycle length and tendency to differentiate [63, 65]. To model these observations, we assume that the multipliers $f(d)$ and $g(d)$ decrease in $d$ and have sigmoid shape. Parameters $a_i$ and $b_i$ in the multipliers determine their maximum and minimum strengths, $d_i^0$ locate the threshold in damage of the transition and $k^d_i$ control the stiffness of the transition. Due to lack of experimental data or previous modeling works to refer to, we assume $f(d)$ and $g(d)$ take the same form and range between $[0.4, 1.1]$ with a transition at $d = 0.75$ (Fig. B.7A).

With a large number of simulations, we present the best choice of the parameters in Table 2.5 and conclude that this set of parameters is our best estimations. Under these parameters, the resulting death rates are $r_S = 2.560 \times 10^{-7}, r_T = 2.314 \times 10^{-4}$ for stem cells and TD cells, respectively. The populations in steady state are $P_S = 9.593 \times 10^6, P_T = 6.709 \times 10^7$ with the ratio $P_T/P_S = 6.994$ (Fig. B.7B).

**B.10 Numerical Scheme**

Consider functions $V_p, V_d, U_p, U_d$ are dependent on $p, d$ only, Eq. (2.1) and Eq. (2.2) can be rewrite as the following system, assuming $W = [S, T]^t$: 104
Figure B.7: A. Sigmoid shape of \( f(d) \) \((a_1 = 1.1, b_1 = -0.7, k_1^d = 20, d_1^0 = 0.75)\). B. A sample simulation with \( \alpha_1 = 0.5, \beta_1 = 0.5, \gamma_1 = 0.25 \) and parameters in Table 2.1 and Table 2.5.

\[
\frac{\partial}{\partial t} W + \frac{\partial}{\partial p} F(W) + \frac{\partial}{\partial d} G(W) = 0 \tag{B.20}
\]

where

\[
F(W) = [f_1(W), f_2(W)]^t = [V_p S, U_p T]^t
\]

and

\[
G(W) = [g_1(W), g_2(W)]^t = [V_d S, U_d T]^t
\]

In the following simulation, we use third-order WENO scheme and third-order TVD Runge-Kutta time integrator.

As \( U_p, U_d, V_p, V_d \) are all positive, the numerical approximation \( W_{i,j} \) to the exact solution \( W(p_i, d_j, t) \) satisfies the following ODE system:

\[
\frac{dW_{i,j}(t)}{dt} = - \frac{\hat{F}_{i+1/2,j} - \hat{F}_{i-1/2,j}}{\Delta p} - \frac{\hat{G}_{i,j+1/2} - \hat{G}_{i,j-1/2}}{\Delta d} \tag{B.21}
\]

where \( \hat{F}_{i+1/2,j} \) is called numerical flux, the design of which is the key ingredient for a successful scheme. For the third-order WENO scheme, the numerical flux \( \hat{F}_{i+1/2,j} \) is
defined as follows:
\[ \hat{F}_{i+1/2,j} = \omega_1 \hat{F}_{i+1/2,j}^{(1)} + \omega_1 \hat{F}_{i+1/2,j}^{(2)} \]  
(B.22)

where \( \hat{F}_{i+1/2,j}^{(m)} \) for \( m = 1, 2 \), are the two second-order accurate fluxes on two different stencils given by
\[ \hat{F}_{i+1/2,j}^{(1)} = -\frac{1}{2} F_{i-1,j} + \frac{3}{2} F_{i,j} \]
\[ \hat{F}_{i+1/2,j}^{(2)} = \frac{1}{2} F_{i,j} + \frac{1}{2} F_{i+1,j} \]  
(B.23)

The nonlinear weights \( \omega_m \) are given by
\[ \omega_m = \frac{\alpha_m}{\sum_{k=1}^{2} \alpha_k}, \quad m = 1, 2 \]  
(B.24)

where
\[ \alpha_k = \frac{\gamma_k}{(\varepsilon + \beta_k)^2} \quad k = 1, 2 \]  
(B.25)

and
\[ \beta_1 = (F_{i,j} - F_{i-1,j})^2 \quad \beta_2 = (F_{i+1,j} - F_{i,j})^2 \]  
(B.26)

and
\[ \gamma_1 = \frac{1}{3}, \quad \gamma_2 = \frac{2}{3} \]  
(B.27)

The parameter \( \varepsilon \) insures the denominator never gets 0, and is fixed at \( \varepsilon = 10^{-6} \) in the computation in this report. Similar construction can be applied to the direction of \( d \).

The time concretization is implemented by a third-order TVD Runge-Kutta method:
\[ W^{(1)} = W^n + \Delta t L(W^n, t^n) \]  
(B.28)
\[ W^{(2)} = \frac{3}{4} W^n + \frac{1}{4} W^{(1)} + \frac{1}{4} \Delta t L(W^{(1)}, t^n + \Delta t) \]
\[ W^{n+1} = \frac{1}{3} W^n + \frac{2}{3} W^{(2)} + \frac{2}{3} \Delta t L(W^{(2)}, t^n + \frac{1}{2} \Delta t) \]

where \( L \) denotes the RHS of Eq.(71).
A CFL condition is needed for stability:

\[
\alpha \frac{\Delta t}{\min\{\Delta p, \Delta d\}} < 1
\]

(B.29)

where \( \alpha = \max\{U_p, U_d, V_p, V_d\} \).
Bibliography


