

# Adaptation to extreme stress under the growth-survival fitness trade-off

Nandita Chaturvedi <sup>\*1</sup>, Charuhansini Tvishamayi<sup>1</sup>, and Shashi Thutupalli <sup>†1</sup>

<sup>1</sup>Simons Center for the Study of Living Machines, National Center for Biological Sciences (TIFR), Bangalore, India

August 27, 2025

## Abstract

Microbial adaptation to extreme stress, such as starvation, antimicrobial exposure, or freezing often reveals fundamental trade-offs between survival and proliferation. Understanding how populations navigate these trade-offs in fluctuating environments remains a central challenge. We develop a quantitative model to investigate the adaptation of populations of yeast (*Saccharomyces cerevisiae*) subjected to cycles of growth and extreme freeze-thaw stress, focusing on the role of quiescence as a mediator of survival. Our model links key life-history traits—growth rate, lag time, quiescence probability, and stress survival—to a single underlying phenotype, motivated by the role of intracellular trehalose in the adaptation of yeast to freeze-thaw stress. Through stochastic population simulations and analytical calculation of the long-term growth rate, we identify the evolutionary attractors of the system. We find that the strength of the growth-survival trade-off depends critically on environmental parameters, such as the duration of the growth phase. Crucially, our analysis reveals that populations optimized for growth-stress cycles can often maintain viability alongside growth-optimized populations even in the absence of stress. This demonstrates that underlying physiological trade-offs do not necessarily translate into fitness trade-offs at the population level, providing general insights into the complex interplay between environmental fluctuations, physiological constraints, and evolutionary dynamics.

## 1 Introduction

Adaptation to extreme stress in microbial systems often occurs at a cost to their ability to grow quickly under ambient conditions [1, 2]. Specifically, an increase in survival under oxidative stress, glucose limitation, freeze-thaw, and heat exposure redirects resources from growth pathways to those dedicated to damage reduction and repair [3]. While trade-offs are not universal across all environmental niches [4, 5], they are a common outcome of adaptation to extreme stressors. These evolutionary trade-offs directly influence the emergence of generalists or specialists depending upon the extremity and frequency of environmental change [6, 7, 8].

Here we present a model of adaptation of *Saccharomyces cerevisiae* to repeated cycles of growth and nutrient exhaustion followed by freeze-thaw stress, based upon experiments covered in previous work [9]. Evolution under these conditions produces a clear fitness trade-off: freeze-thaw survival increases while the growth rate in ambient conditions decreases. Our model simulates the evolution of a yeast population through these repeated cycles and explicitly addresses the roles of quiescence and lag time.

The evolved cells in our experimental system are smaller, denser and lighter than the wild type cells and show higher intracellular trehalose concentration in stationary phase [9]. Trehalose, a dimer of glucose plays a crucial role both in entry to and exit from quiescence, as well as cellular response to extreme stress. Trehalose has been observed to bind to the cell wall and protect protein molecules which

---

<sup>\*</sup>nandita@ncbs.res.in

<sup>†</sup>shashi@ncbs.res.in

prevents damage from freezing or desiccation [10, 11, 12, 13, 14, 15]. Total intracellular trehalose shows correlation with the density of individual cells both in stationary phase cultures and during continuous growth. Immediately upon exit from quiescence, cells metabolize trehalose to re-enter the cell cycle. Cells that lack trehalose initiate growth more slowly and frequently exhibit poor survivability [16]. Thus, cells with more trehalose exhibit a shorter lag phase when re-introduced to rich media [17]. A shorter lag phase has also been linked to a higher adaptability to changing environments [18]. This establishes a dual trade-off: not only between growth rate and stress survival, but also between growth rate and lag time, a phenomenon previously observed in *E. coli* [19].

Further, when yeast is subject to freeze-thaw stress without allowing the population to reach stationary phase, the population crashes, suggesting that quiescence acts as a helpful step to the evolution of freeze-thaw tolerance. Recent work in yeast has shown that stress response often follows a general strategy rather than evolving to specific stress conditions [1, 2, 20]. For instance, yeast evolved under aerobic glucose limitation show few trade-offs with growth in other carbon-limited environments [2]. While the fitness of the evolved strain increases in comparison to yeast growing in rich media, these fitness improvements can arise from multiple mutational paths. This general stress response is most evident at the level of the phenotype and includes producing higher levels of stationary phase trehalose, thickening of the cell wall, a reduction in size [9], and is underpinned by large-scale, genome-wide transcriptional reprogramming [20, 21]. Therefore, while our model is grounded in a specific experimental system, its results can generalize to any microbial population evolving under cycles of growth and stress exposure.

Our model highlights the role of quiescence in adapting to extreme stress. Yeast populations undergo a diauxic shift following glucose depletion, where cells reduce their growth rate to readjust their metabolism for the subsequent phase of slow, respiratory growth. After nonfermentable carbon sources have been consumed, the population enters stationary phase. Stationary phase cultures are complex and heterogeneous, composed of a large fraction of quiescent, long-lived, largely daughter and young mother cells, and a nonquiescent subpopulation of cells [22, 23, 24, 25, 17]. The non-quiescent subpopulation eventually perishes under conditions of long term starvation. Quiescence is a distinct, growth-arrested phase of the cell cycle [22, 26, 27].

Thus, quiescence of yeast cells in conditions of starvation is a milder stress response which could prime them for harsher conditions, enabling survival during long periods of starvation [28]. Quiescent cells share several key characteristics with response to extreme stress, including smaller, denser cells with thicker cell walls [29, 25], and also show increased resistance to diverse environmental stresses [28, 30, 31]. However, the exact relationship between quiescence – a separate cell cycle phase involving stochastic switching – and canonical cellular stress response remains unclear [30, 23]. Distinct from stress-resistant cells, quiescent cells show growth arrest and stochastic switching between the growth cycle and quiescence. The dynamics of entry into and exit from quiescence has been shown to be a noisy process that couples deterministic memory effects of preceding cell cycle with stochastic switching [32]. Yet, the large scale rearrangement of gene expression, metabolic state and cellular processes that takes place in quiescence may show similarity to stress response in yeast [33, 3, 34, 35].

We therefore identify intracellular trehalose produced in stationary phase as a primary phenotypic marker. We posit that this intracellular trehalose concentration determines the population’s growth rate, survival probability, lag time, and each cell’s likelihood of going into quiescence. Our model integrates growth, lag and survival-mediated selection with the stochastic switch to quiescence.

In Section 2 we introduce our model of microbial evolution under growth-starvation-stress cycles that treats the switch to quiescence as a stepping stone to stress-resistance. The model qualitatively captures the evolution of yeast populations under cycles of growth, starvation, and freeze-thaw stress. We specifically confirm its predictions for the evolutionary trajectory of the central phenotype, the amount of intracellular trehalose in stationary phase, with experimental results. In Section 3 we calculate the long term growth rate of a population under growth-stress cycles and use it to calculate the optimum phenotype for different evolution programs. We find that despite the growth-survival trade-off, populations evolved under growth-stress cycles can coexist with those evolved in the absence of stress.

## 2 Trehalose Mediated Quiescence, Lag Time, Growth and Survival

We highlight a few features of the experiments in [9] to motivate our model: (1) The survival fraction for yeast undergoing several rounds of growth followed by freeze-thaw stress shows a gradual increase from about 2% to 70% over 25 cycles. Since the increase is gradual, this indicates that the adaptation to freeze-thaw is not through a single sweeping mutation or rearrangement of biological pathways, but rather through a series of changes, either genetic or otherwise, that allow for incremental fitness increases. (2) The population shows survival and subsequent adaptation when freeze-thaw is applied after it reaches stationary phase. If the population is frozen in its exponential phase, it collapses within a few cycles and no survival is seen. (3) The evolved strain of yeast after about 25 cycles is characterized by a shortened lag phase after stress, a slightly lower growth rate and higher levels of trehalose production in stationary phase. (4) The cells of the evolved strain are smaller, lighter but denser, and show less budding, similar to key characteristics of quiescent cells.

Entry into quiescence is a stochastic process, while the efficiency of a cell's exit from quiescence is expressed in the duration of the lag phase upon re-entry into rich media. The duration of the lag phase is also determined by the amount of intracellular trehalose produced in stationary phase, as trehalose is preferentially metabolized to re-enter the cell cycle.

We assume that a single phenotype is able to capture different aspects of a cell's fitness. We take the primary phenotype to be the amount of intracellular trehalose in the cell when the population enters stationary phase. It is well known that higher trehalose levels correspond to lower cellular damage under physical stress, as well as higher survival fractions under freezing and desiccation. The cellular mechanisms that produce trehalose as part of a broader and general stress response show a trade off with the pathways that promote cell division. Thus, as trehalose production under stressful conditions increases, growth rate of the cell upon reentry into the growth phase decreases. All of this is also observed in our experimental results, and we consider that the stationary phase trehalose levels in the cells can determine the cell's growth rate, quiescence probability, survival probability, and lag time upon reentry into the cell cycle.

### 2.1 Methods

We start with a population of  $N$  individual cells whose phenotype, given by  $\phi$ , is drawn initially from a Gaussian distribution with mean  $\phi_0$  and a narrow peak with standard deviation  $\sigma_\phi^0$ . We assume the dependence of each cell's growth rate (or fecundity per unit time in the simulation) on stationary phase trehalose concentration to be quadratic,

$$g(\phi) = g_0(1 - \phi/\tau_g)^2 \quad (1)$$

where  $g_0$  is the highest achievable growth rate and  $\tau_g$  the amount of trehalose that inhibits growth to effectively zero. Note that our population does not typically reach  $\phi$  values close to  $\tau_g$  during the course of the simulation, and we set  $g(\phi) = 0$  for  $\phi > \tau_g$ . At each time step in the growth process, we calculate the number of offspring for each cell by applying a ceiling function to  $g(\phi)$  calculated according to the above equation, and add them to the population. We consider here a quadratic function for  $g(\phi)$  so as to allow the growth rate to fall faster with increasing  $\phi$  values at lower values and slower as  $\phi$  reaches higher values. Each offspring is also subject to a small mutation, which acts to change the trehalose concentration of the offspring with respect to its parent. This mutational effect on offspring phenotype is drawn from a Pearson Distribution at a mutation rate  $m$  for each offspring. The Pearson Distribution is a continuous probability distribution with a long tail defined by its kurtosis. Our distribution has a mean at zero, mutational standard deviation  $\sigma_m$ , a kurtosis  $k_m$  and no skew. A kurtosis  $k = 3$  is a Normal distribution, with outliers becoming more probable with increasing kurtosis. For our simulations we use  $m = 0.8$  and  $k_m = 5$ . Most mutations have no effect upon the offspring's phenotype, preserving the value inherited from the parent cell. Simultaneously, some large effect mutations, both positive and negative are possible.

For each division, we assume a constant amount of a finite resource,  $r_d$ , is consumed, starting with  $r_0$  resource units. Cells also consume a smaller amount of the resource,  $r_l$  while they are non-growing in lag phase. The growth process continues till the resource runs out and the population faces resource exhaustion. For our simulations we use  $r_d = 0.5$ ,  $r_l = 0.1$  and  $r_0 = 10^5$ . Upon starvation, yeast forms a

heterogeneous population of quiescent and non-quiescent cells. We model the switch to quiescence as a stochastic process, where the probability of each cell going into quiescence is given by

$$p_q(\phi) = \frac{1}{(1 + \exp^{-(\phi - \tau_q)/\gamma_q})}, \quad (2)$$

where  $\tau_q$  is the trehalose concentration at which probability of quiescence is 50% and  $\gamma_q$  is a parameter quantifying how quickly the probability rises to 1 as  $\phi$  increases. The functional form, as well as the  $\tau_q$  values we chose allows for probability of quiescence to gradually increase with  $\phi$  and saturate at a high value. For each cell we decide the fate of the cell by comparing this probability with a randomly generated number. If the probability given by Equation 2 is higher than the random number the cell goes into a quiescent state.

Note that the phenotype of each cell changes from mutations during the growth process. The phenotype corresponds to trehalose concentrations produced by the cell eventually in the stationary phase, which also determines its quiescence probability. Thus mutations accrued in growth process can bias a cell towards or away from quiescence, although no trehalose is produced in the growth phase.

Entry upon quiescence is followed by exposure to extreme stress, which in our experiments is freeze-thaw. Upon exposure to extreme stress, we assume that the non-quiescent population entirely perishes. Among the quiescent cells, the cells with higher trehalose concentration have a higher probability of survival. We model the probability of survival,

$$p_s(\phi) = \frac{1}{(1 + \exp^{-(\phi - \tau_s)/\gamma_s})}, \quad (3)$$

where  $\tau_s$  is the trehalose concentration at which probability of survival under freeze thaw is 50% and  $\gamma_s$  is a parameter quantifying how quickly the probability rises to 1 as  $\phi$  increases. Again, we choose  $\gamma_s$  and  $\tau_s$  such that the probability of survival is close to zero for low values of  $\phi$  and then increases relatively quickly in an interval given by  $\gamma_s$ , representing a switch-like transition to having enough trehalose production in each cell to lend the cell resistance to freeze-thaw. In general we expect that  $\gamma_s < \gamma_q$  and  $\tau_s > \tau_q$  since the stress is harsher to cells than nutrient scarcity in stationary phase. Thus, both quiescence and the freeze thaw process shift the mean phenotype of the population to higher values since cells with lower phenotype amounts perish. The functions for growth rate, quiescence probability and survival probability are shown in Figure 1 (a).

When the surviving population is allowed to thaw and re-introduced into a growth medium, the cells with higher trehalose concentrations have a shorter lag time, and begin to grow earlier as trehalose is crucial for re-entry into the cell cycle [16]. Longer lag times mean less resource available for growth because a small amount of resource is consumed even in the lag phase and the total amount consumed before growth depends on the length of the lag phase. We assume the simplest possible functional form, a linear relationship between the lag time and trehalose amounts,

$$l(\phi) = l_0(1 - \phi/\tau_l), \quad (4)$$

where  $l_0$  represents the maximum lag time possible, and  $\tau_l$  the phenotype value at which the lag time reduces to zero. We set  $l(\phi) = 0$  for all  $\phi > \tau_l$  for the sake of our simulation, although  $\tau_l$  is set large enough that  $\phi$  does not exceed it in the simulations. In each growth cycle, a phenotype value thus means a shorter lag phase, and more time available for growth, while simultaneously causing a lower growth rate. Lastly, we model the dilution process of the experimental setup by randomly selecting a fraction  $d$  of the population. This can introduce additional stochasticity into the evolution process.

## 2.2 Results

When the population starts growth after exit from lag phase with lower values of mean trehalose, the effect of a decreasing growth rate is not sufficient to stop the increase in mean trehalose of the population caused by a shorter lag phase. In the growth phase, cells with higher trehalose are able to produce a larger number of offspring and take over the population.

As the population undergoes a complete round of growth and nutrient exhaustion, we have a resultant heterogeneous population, with cells with higher  $\phi$  having a higher probability of entering quiescence. Thus, the distribution of quiescent cells now has a higher mean amount of trehalose production than the entire population taken together. A fraction of these quiescent cells are able to

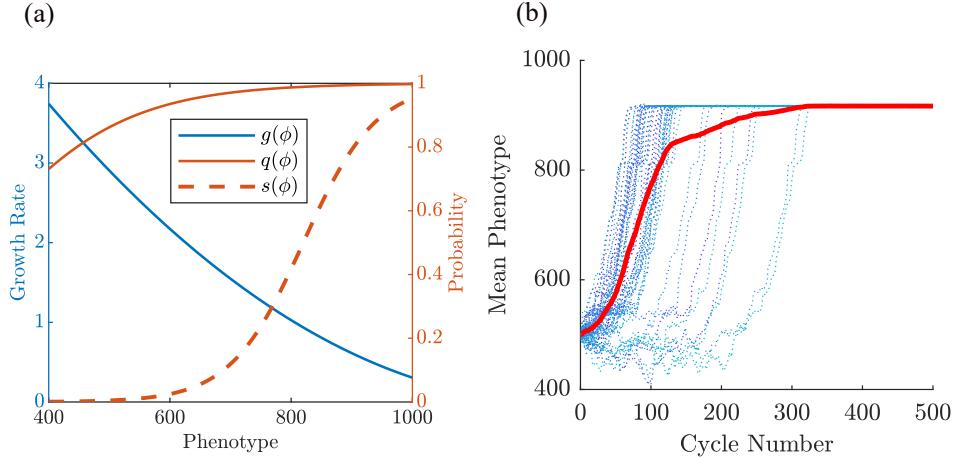


Figure 1: (a) Growth rate, quiescence probability and survival probability as functions of the phenotype. (b) The mean phenotype of the population first increases stochastically before settling to an equilibrium value. The red curve is the average over 100 replicate populations, and the blue trajectories represent individual replicates. As can be seen, while the trajectory is stochastic, the final equilibrium phenotype remains the same across replicates. Here  $m = 0.8$ ,  $\sigma_m = 3$ ,  $k_m = 5$ ,  $l_0 = 30$ ,  $\tau_l = 1000$ ,  $g_0 = 40$ ,  $\tau_g = 1200$ ,  $r_0 = 10^5$ ,  $\tau_q = 400$ ,  $\gamma_q = 100$ ,  $\tau_s = 850$ ,  $\gamma_q = 50$ .

survive freeze-thaw stress. The cells that are now able to survive are now a larger proportion of the entire population because of this upward shift in  $\phi$ . Hence, our simulation includes both a component of trehalose-dependent stochastic switching to quiescence, as well as growth, lag and survival mediated selection for trehalose levels. The trade-off between the decrease in growth rate and the positive effect on lag time and survival from increasing trehalose sets the final equilibrium value of trehalose in the simulations. The effect of a lower growth rate begins to dominate at larger trehalose values, causing the mean trehalose of the population to stagnate.

We see an increase in the phenotype and survival as the evolution process progresses, before settling at an equilibrium value. This is shown in Figure 1 (b). The blue curves represent individual replicate populations, while the red curve represents the average over 100 replicates. The behavior of populations is highly stochastic in the early stages of the evolutionary process, due to small numbers of cells that survive freeze-thaw stress, but the equilibrium value they settle to remains conserved across all replicates.

Figure 2 (a) and (b) show the evolution of the populations mean phenotype and the resultant survival fractions over time. We compare them to the trehalose evolution curve (c) and the survival fraction curve (d) obtained from the experimental evolution of yeast under freeze thaw. Both sets of curves show qualitative agreement. The rapid increase in the survival rate is caused by the large tailed mutational distribution we use which biases evolution towards large jumps. We note also the delay in increase of both the survival curves, which arises, in the case of our simulation, from the sigmoidal dependence of the survival probability upon trehalose concentration.

We investigate the effect of increasing  $g_0$  on equilibrium values of the phenotype and survival fraction in Figure 3 (a) and (b). Increasing  $g_0$  increases the number of offspring at each time point, leading to the resource being exhausted faster, thus less time is spent by the population in its growth phase. This causes the effect of selection from growth to decrease, allowing the phenotype to increase to higher values and biasing the population towards maximizing survival. Thus, we see an increase in both the equilibrium value of the phenotype as well as survival fraction as  $g_0$  increases.

Similarly, Figure 3 (c) and (d) show that decreasing the rate at which the lag time decreases as a function of the phenotype, controlled by increasing  $\tau_l$ , also leads to an increase in the equilibrium values of the phenotype and survival fraction. We find that increasing the value of  $\tau_l$  leads to the lag time increasing at each value of the phenotype. This means that a larger proportion of the resource is consumed by non-growing cells, leading to the population having less time in the exponential growth phase before the resource is exhausted.

Changing the mutation rate and variance of mutation effects only affect the dynamics of the climb to equilibrium, leaving the equilibrium value of the phenotype unaffected.

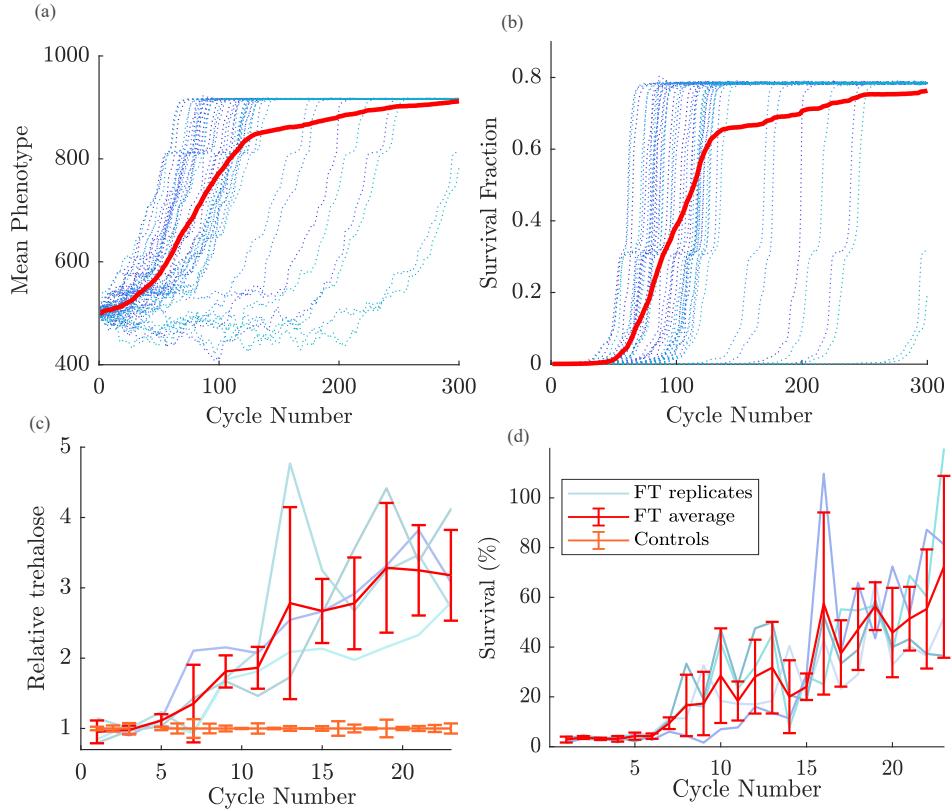


Figure 2: (a) Mean phenotype of the population and (b) survival fraction with cycle number for 100 replicate populations. For comparison, (c) is the mean trehalose amount at stationary phase in the yeast populations as a function of cycle number. Four replicates and their average is shown. (d) Survival fraction of yeast populations under freeze-thaw stress increases from about 2% to 70% in as little as 25 cycles of freeze-thaw. Figures (c) and (d) are reproduced from [9].

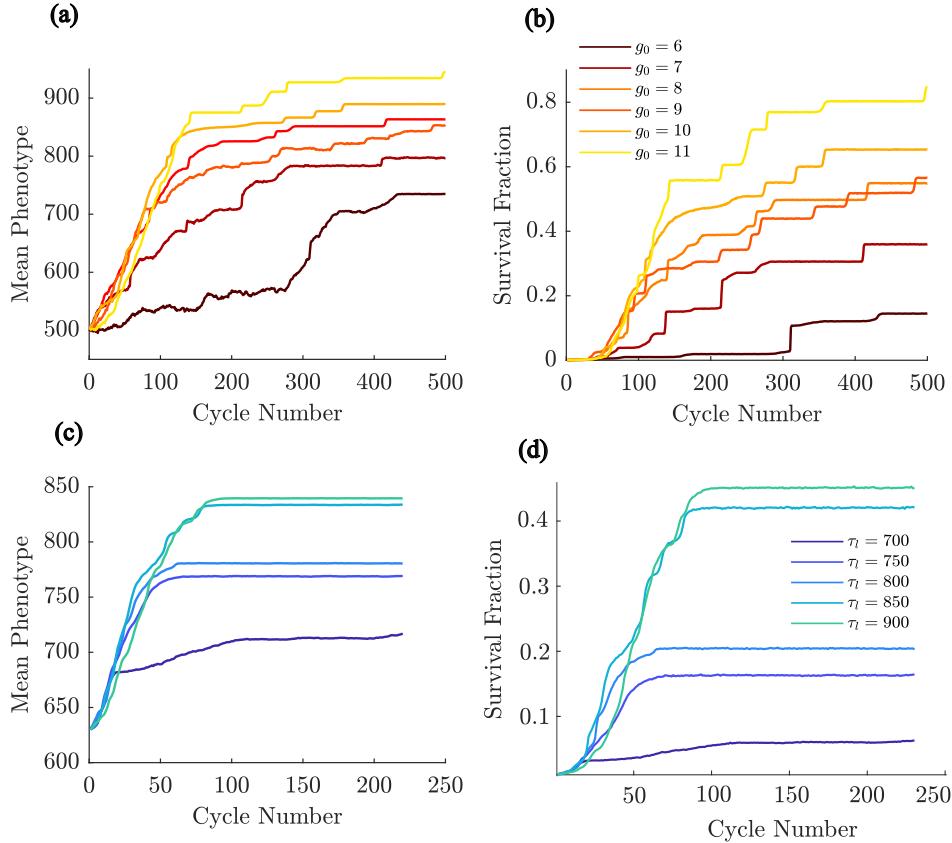


Figure 3: Mean phenotype and survival fraction for changing  $g_0$  (figure (a) and (b)) and  $\tau_l$  (figure (c) and (d)). Each line represents the mean value taken over 40 replicates. As can be seen in (a) and (b), increasing  $g_0$  increases final equilibrium phenotype as well as survival. Two competing effects come into play, a higher  $g_0$  means a larger population level variation at each time point for selection to occur. It also means that the total time for growth reduces if the total resource is kept constant. However, increasing  $g_0$  implies also a higher selection from growth. The effect of decreasing growth time dominates. For (a) and (b),  $m = 0.8$ ,  $\sigma_m = 3$ ,  $k_m = 5$ ,  $l_0 = 30$ ,  $\tau_l = 1000$ ,  $r_0 = 10^5$ ,  $\tau_g = 1200$ ,  $\tau_q = 400$ ,  $\gamma_q = 100$ ,  $\tau_s = 850$ ,  $\gamma_q = 50$ . In (c) and (d) we can see increasing the phenotype value at which lag effectively drops to zero also increases the final phenotype and survival. This is because increasing  $\tau_l$  means that the lag time is higher at the same phenotype. Since this leaves less time for growth, the contribution of selection from growth decreases, allowing the equilibrium value of the phenotype as well as survival to climb higher. For (c) and (d),  $m = 0.8$ ,  $\sigma_m = 3$ ,  $k_m = 5$ ,  $l_0 = 30$ ,  $r_0 = 10^5$ ,  $g_0 = 20$ ,  $\tau_g = 1200$ ,  $\tau_q = 400$ ,  $\gamma_q = 100$ ,  $\tau_s = 850$ ,  $\gamma_q = 50$ .

Hence we find that the amount of time spent in the growth phase, the maximum growth rate, among other parameters can affect the equilibrium value of the phenotype and survival by changing the strength of the trade-off. We write down the exact form of this dependence in Section 3 by estimating the long term growth rate of the population.

### 3 Long Term Growth Rate and Comparing Fitness

We calculate the long term growth rate of the evolving population to help us to analytically recover some of the results of our model. Consider a population of cells evolving under growth-stress cycles given by the distribution  $n(\phi, t)$ . As in the last section, the growth rate, probability of survival under stress and lag time all depend on the phenotype of each cell. Through the growth phase, the number of cells with phenotype  $\phi$  at time  $(t + 1)$  is given by  $g(\phi)n(\phi, t)$ , where  $g(\phi)$  is the growth rate at phenotype  $\phi$ . If the population is allowed to grow for  $T_g$  time till the resource runs out, at the end of the growth phase the number of cells at each phenotype is

$$n(\phi, T_g) = g(\phi)^{T_g - t_l(\phi)} n(\phi, 0), \quad (5)$$

where  $t_l(\phi)$  is the lag time of a cell with phenotype  $\phi$ . We measure time in units of generation time, so  $g(\phi)$  is only a numerical factor.

The population is then exposed to extreme stress, such as freeze-thaw, and the fraction of cells at each phenotype that survive the stress is  $s(\phi)$ . Thus after one cycle of lag-growth-starvation-stress and dilution, the population has a distribution

$$n(\phi, T_g + T_s) = ds(\phi)g(\phi)^{T_g - t_l(\phi)} n(\phi, 0). \quad (6)$$

where  $T_s$  is the duration of stress exposure and  $d$  is the dilution rate. Note that we do not explicitly account for transition to quiescence here for simplicity of calculation, which means that  $s(\phi)$  does not act upon a smaller quiescent population, but rather on the entire population. The survival process in this calculation can be thought of as an effective process that combines quiescence and survival from our simulations. After  $c$  cycles, then, we have a population distribution

$$n(\phi, c(T_g + T_s)) = [ds(\phi)g(\phi)^{T_g - t_l(\phi)}]^c n(\phi, 0). \quad (7)$$

Thus, the subpopulation with phenotype  $\phi$  grows exponentially as the number of cycles tends to infinity if  $ds(\phi)g(\phi)^{T_g - t_l(\phi)} > 1$  and dies out exponentially if  $ds(\phi)g(\phi)^{T_g - t_l(\phi)} < 1$ . At large cycle numbers, the growth rate of the population is given by  $\Lambda = \frac{1}{C} \log \frac{n(\phi, C(T_g + T_s))}{n(\phi, 0)}$ . Thus, the effective long term growth rate of the subpopulation with phenotype  $\phi$  under growth-stress cycles can then be written as

$$\Lambda_s = \log [ds(\phi)g(\phi)^{T_g - t_l(\phi)}] \quad (8)$$

At long times, the phenotype that maximizes the long term growth rate dominates the population. Thus, finding the  $\phi$  values at which  $\Lambda_s$  is maximum will give us the optimum phenotype for a population evolved under growth-stress cycles. Further, the rate at which the  $\Lambda_s$  falls off from this maxima can give us an estimate of the variation in the population.

Figure 4 shows a comparison of the equilibrium phenotype values from the simulation with the optimal phenotype that maximized the long term growth rate in equation 8. As was seen in the last section, the equilibrium phenotype increases with both an increase in  $g_0$  and  $\tau_l$ . Note that we made the assumption above that the total time of growth,  $T$ , remains constant across cycles. This assumption does not hold in the simulations as the time of growth decreases with increasing population size as the evolution progresses forward. Growth time in our simulations drops from a relatively high value in the beginning to a lower equilibrium value. For the growth time dynamics, see Supplementary Information. We use the mean of the growth time from simulations as  $T$  in 8 to calculate the optimum phenotype. Since we are comparing a finite resource growth process (in the simulations) to a finite time growth process (in the analytic calculation), some difference in the two predictions is seen. However, the analytical prediction agrees with the values from the simulation to a large degree.

If the population undergoes cycles of growth, starvation and dilution without exposure to stress, we can write the long term growth rate as  $\Lambda_g = \log [dg(\phi)^{T_g - t_l(\phi)}]$ . Figure 5 (a) shows  $e^{\Lambda_g} = dg(\phi)^{T_g - t_l(\phi)}$

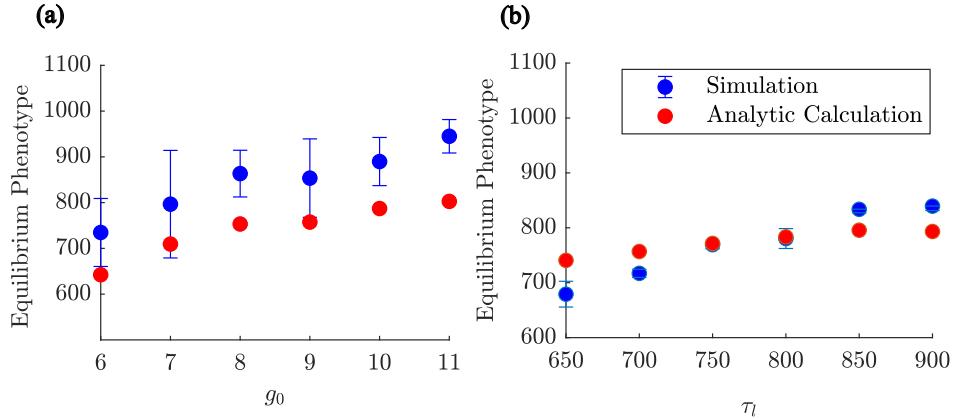


Figure 4: We compare the equilibrium values of the population’s mean phenotype (in blue) with the optimum phenotype  $\phi$  that maximizes equation 8 (red) for different values of  $g_0$  and  $\tau_l$ . Each point from the simulations represents the mean and standard deviation from 20 different replicates. The difference between the two curves can be explained by the assumption that the growth time remains constant during the duration of evolution. We use the mean value of  $T$  from simulations to determine the optimum phenotype analytically. This leads to a difference of the equilibrium phenotype value from the analytically predicted value. To see how the growth time varies with cycle number, see Supplementary Information. Unless otherwise specified, here we use  $m = 0.8$ ,  $\sigma_m = 3$ ,  $k_m = 5$ ,  $l_0 = 30$ ,  $\tau_l = 1000$ ,  $r_0 = 10^5$ ,  $g_0 = 20$ ,  $\tau_g = 1200$ ,  $\tau_q = 400$ ,  $\gamma_s = 100$ ,  $\tau_s = 850$ ,  $\gamma_q = 50$ .

and  $e^{\Lambda_S}$  as a function of  $\phi$ . Since  $s(\phi)$  increases with  $\phi$ , while  $t_l(\phi)$  and  $g(\phi)$  decrease with  $\phi$ , both functions have a unique maximum. The position of this maximum is different for the case with and without stress, representing the varying degrees of the growth-survival trade-off.

The long term growth rate is an effective tool to compare two populations evolved under different conditions; under solely growth cycles and under growth-stress cycles. We calculate the value of the phenotype which maximizes the long term growth rate for each evolution program,  $\phi_{G0}$  and  $\phi_{S0}$ , respectively. We then compare  $\Lambda_G$  and  $\Lambda_S$  of both optimal phenotypes in the two cases. This is shown in Figure 5. The three different point color schemes, blue, green and red, represent three different values of  $T$ , while the different shades in each color represent optimum phenotypes for different  $g_0$  values, with  $g_0$  increasing from lower left to upper right quadrant. The same color circle and diamond show the performance of  $\phi_{G0}$  and  $\phi_{S0}$  evolved with the same  $g_0$  and  $T$  with and without stress respectively.

Note that when the long term growth rate is negative, the strain is expected to go extinct. As can be seen, for most parameter pairs, the performance of the population remains in the upper right quadrant under growth cycles (circular points), *i.e.* populations of both  $\phi_{G0}$  and  $\phi_{S0}$  grow well under only growth cycles. When each circular point is compared with its counterpart under growth-stress cycles (diamond points), you see a shift towards lower  $\Lambda$  for both phenotypes.

Some populations, with larger  $g_0$  values, remain in the upper right quadrant, implying both  $\phi_{G0}$  or  $\phi_{S0}$  are able to grow under both growth-stress, only with reduced rates (with  $\Lambda_i(\phi_{S0})$  typically decreasing less than  $\Lambda_i(\phi_{G0})$ ). Thus, two populations of  $\phi_{G0}$  and  $\phi_{S0}$  are generalists with different emphasis on growth and survival. This implies that in cases where the population variance is large enough, coexistence may be possible.

Some populations under growth-stress cycles shift to the lower left quadrant, implying neither  $\phi_{G0}$  or  $\phi_{S0}$  are able to grow under stress. The counterparts for these populations under solely growth cycles are in the first quadrant, implying that both  $\phi_{G0}$  or  $\phi_{S0}$  are able to grow, and even possibly coexist. This occurs at lower values of  $g_0$ , where the low fecundity does not allow the population to recover its numbers after the significant decrease in its population size upon exposure to stress. Increasing both  $g_0$  and  $T$  can help the population recover and reduce this effect.

A few populations under growth-stress fall in the second quadrant, showing survival of  $\phi_{S0}$  under stress conditions and extinction of  $\phi_{G0}$ . In other words, a strain evolved under stressful conditions survives under growth-stress cycles, while one evolved under growth cycles does not. However, the counterparts of these populations under solely growth cycles exist in the upper right quadrant. This shows that even though a population of  $\phi_{S0}$  evolves to exist under stress conditions when  $\phi_{G0}$  cannot,

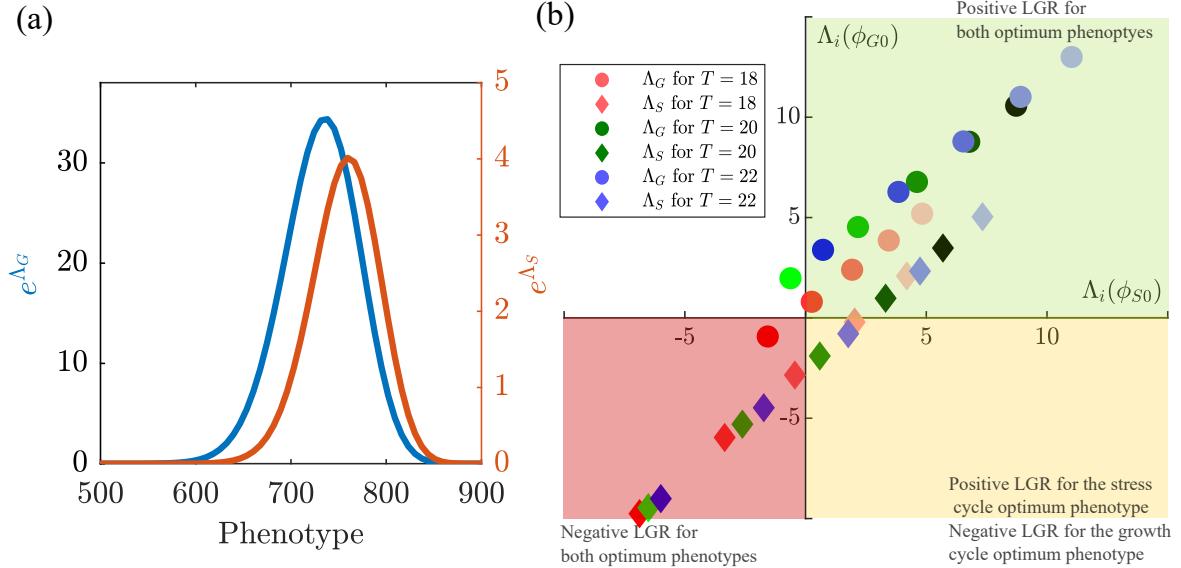


Figure 5: (a) A plot of  $e^{\Lambda_S}$  and  $e^{\Lambda_G}$  shows that the value of  $\phi$  at which they are maximized, as well as their values at the optimum are different. (b) The optimal phenotype in each experimental setup (with and without stress) was calculated for different values of  $T$  and  $g_0$ . Red, blue and green series show the three values of  $T$  chosen while  $g_0$  was chosen to be 6, 8, 10, 12 and 14 for each  $T$  value (increasing as you go from lower left to upper right in each series). The long term growth rate of these ‘growth type’ and ‘survival type’ optimal strains with phenotypes  $\phi_{G0}$  and  $\phi_{S0}$ , respectively, was then calculated with and without stress growth cycles. The same color circle and diamond show the performance of  $\phi_{G0}$  and  $\phi_{S0}$  evolved with the same  $g_0$  and  $T$  with and without stress, so can be compared to each other. Note that when the long term growth rate is negative, the strain is expected to go extinct. As can be seen, for most parameter pairs, the performance of the population remains in the upper right quadrant for under growth cycles (*i.e.* populations of both  $\phi_{G0}$  and  $\phi_{S0}$  grow well under only growth cycles. When each point is compared with its counterpart under stress cycles (diamond points), you can see a shift towards lower  $\Lambda$  for both phenotypes. Some points remain in the upper right quadrant, implying both  $\phi_{G0}$  or  $\phi_{S0}$  are able to grow under stress, only with reduced rates (with  $\Lambda_i(\phi_{S0})$  typically decreasing less than  $\Lambda_i(\phi_{G0})$ ). Some points are shifted to the lower left quadrant, implying neither  $\phi_{G0}$  or  $\phi_{S0}$  are able to grow under stress. A few points are in the lower right quadrant, showing survival of  $\phi_{S0}$  under stress conditions and extinction of  $\phi_{G0}$ . In other words, a strain evolved under stressful conditions survive under stress cycles, while that under growth cycles do not.

under solely growth cycles the two populations may coexist. Hence, despite the growth-survival trade-off, populations evolved under growth-stress may not show significant trade-offs with their fitness under only growth cycles.

## 4 Discussion

Growth-survival trade-offs, observed across microbial species, help explain key ecological phenomena like co-existence, population heterogeneity, and the preference for resource-scarce environments [36, 37]. We present a model of adaptation to extreme stress under such a trade-off. To our knowledge, this is a novel effort to model the role of quiescence in evolutionary adaptation to extreme stress. This approach is supported by evidence that stress response in yeast may follow a general strategy rather than evolving to specific stress conditions [1, 20, 38]. Studies in bacteria suggest that exposure to lower concentrations of antibiotics may prepare cells to take on the challenge of lethal concentrations [39, 40]. Further, the existence of the growth-lag trade-off suggests that starvation in stationary phase may be perceived a mild stress [41]. These observations support our assumption that quiescence, a

response to the relatively mild stress of starvation, may provide a helpful step towards resistance to harsher stress.

Previous work has demonstrated that yeast populations increase their survival from 2% to 70% over the course of 25 generations [9]. These evolved strains show key characteristics of quiescent cells including smaller, denser cells, increased cell wall thickness. Our model captures this evolutionary trajectory qualitatively at the phenotypic level. It also provides a framework to explore its dependence on the frequency and strength of environmental change, as well as on features of the population such as its maximum growth rate and the nature of the variation of lag time with the population phenotype.

Our model assumes that the growth rate, probability of quiescence, probability of survival as well as lag time of each cell depends on a single phenotype. In the case of yeast, we propose that this phenotype corresponds to the amount of intracellular trehalose produced by the cell once it reaches stationary phase. Previous work with *E. coli* shows that phenotypic heterogeneity in lag times could be a strategy to break the limitations of the growth-survival trade-off [42]. However, here we take a more holistic view by factoring in both the switch to quiescence, the exit from quiescence (through lag time) and their interaction with growth through dependence on the same phenotype. We take the functional form of this dependence to be the simplest assumptions that are biologically realistic; linear in the case of lag time, quadratic for growth rate and sigmoidal for the two probabilities. We are not aware of any attempt to quantify the dependence of these characteristics upon trehalose concentrations in yeast populations experimentally. Our results are nevertheless robust to changes in these functional forms. They depend only on the presence of a trade-off in the form of lag time and growth rate decreasing while the probability of survival and quiescence increase with the phenotype. Our results also are characteristic of selection from a threshold like selection pressure, and we expect similar behavior in many systems that show such a feature [43, 44, 45, 46].

We model mutational effects drawn from a long-tailed Pearson distribution. The model is agnostic to the source of such mutations, which could be genetic or epigenetic in nature. Further, the framework does not speculate on the mechanism through which the memory of stress, or of intracellular trehalose produced in stationary phase can be inherited across generations. Interestingly, the whole genome sequencing of the evolved yeast strains in [9] does not show any mutations conserved across replicate lines. This is also supported by previous work [47]. Yet, a convergent phenotype emerges within the short period of 25 generations, suggesting an epigenetic stress response. This response seems to still be inheritable, as evidenced by the evolved phenotype persisting even in the absence of continued stress. It is possible that if quiescence, a generalized stress response mechanism, facilitates the adaptation to freeze thaw, that the adaptation mechanism is one that involves genetic assimilation [48, 49].

We assume a stochastic switch to quiescence, with the probability of quiescence increasing with increasing intracellular trehalose. In this way our system is similar to stochastic switching to a persistent state in bacterial populations, although for us probability of switching is biased by trehalose concentration in stationary phase [50, 51, 52]. However, our model is distinct from most models of bacterial persistors since we postulate that our phenotype is inheritable and over time lends higher survival fractions. There is, however, some support for the idea that that persistence frequency in bacteria is also an evolvable trait that adapts to both the duration and frequency of the antibiotic application[53, 54]. We assume that the subset of quiescent cells is then subject to probabilistic survival upon exposure to stress. This is a reasonable assumption given previous evidence that quiescence leads to higher stress tolerance [55]. Yet this assumption remains experimentally unconfirmed, and it is not certain whether the same quiescent subset of the population in stationary phase is selected from by subsequent stress.

Although the model presented here allows for a more sophisticated treatment of the biological processes involved in adaptation, we found that a simpler toy framework is also sufficient to capture the dependence of equilibrium survival on trade-off strength. Details of such a simpler framework which relies on a growth and survival fitness function is presented in Supplementary Information. In our model the trade-off strength is determined by the interplay between growth rate, quiescence and survival probabilities and lag time. In the toy model it is a more direct feature, the distance between the two fitness optima and selection strengths of the fitness functions.

We calculate the long term growth rate of a population adapting to growth-stress cycles and use it to calculate the optimum phenotype for different evolution programs. Our analysis reveals that the duration and nature of the growth process affects directly the optimum phenotype, final survival, as well as the long term growth rate. We are also able to compare the fitnesses of populations that

are optimum under growth cycles and under growth-stress cycles. We calculate the long term growth rate of these 'growth-type' and 'stress-type' populations both with and without stress. We find that depending on the evolution program, the populations can show the possibility of coexistence, or show a trade-off in their overall fitness. The populations show the possibility of coexistence both with and without stress at large fecundity,  $g_0$ , indicating that their overall fitness may not show a strong trade-off if they are able to recover their populations in the growth phase after exposure to stress, where the population size decreases drastically.

For a smaller range of lower values of  $g_0$  and time of growth,  $T$ , we see that 'stress-type' populations may survive both types of environments, while 'growth-type' populations do not survive cycles with stress exposure. Thus, for some evolution paradigms, populations that do not show a significant decrease in their fitness in growth-only cycles while still increasing their fitness in cycles with stress exposure exist. This is because the overall fitness and long term growth rate depend not only on the relative strengths of the growth and survival functions but also on other characteristics of growth such as time,  $T$  and maximum growth rate  $g_0$ . Several experimental studies have shown that fitness trade-offs are not ubiquitous in microbial systems [4, 56]. Our results demonstrate that the absence of more obvious fitness trade-offs does not preclude the existence of an underlying growth-survival trade-off.

Although previous work has suggested that coexistence and trade-offs at the fitness level are context dependent [57], our calculations allow for a more systematic treatment of the different cases possible. Our results from calculations of the long term growth rate can easily be verified by evolving populations under different evolution programs, with varying growth times for instance.

Our model is also reminiscent of recent work done on the population dynamics of bacteria in stochastic feast-famine cycles [58, 59]. Previous work which models growth and death processes both with growth-yield and growth-death trade offs shows that the ratio of the growth rate and death rate is an effective fitness of the population [58]. This is similar to our expression for the long term growth rate which combines survival probability with the growth rate. Further, they also find the duration of the feast and famine periods to be important to the eventual fate of the population. However, here we also take into account the added complexity of lag time along with growth and survival, all determined by a single phenotype. Other work has examined the optimal persistence strategy for bacteria in feast-famine cycles with the stochastic application of antibiotics [60] to find that 'triggered' dormancy, such as what we have included in this model with quiescence, may be the optimal strategy when the stress of the antibiotic treatment is very severe.

## References

- [1] Jared W Wenger, Jeffrey Piotrowski, Saisubramanian Nagarajan, Kami Chiotti, Gavin Sherlock, and Frank Rosenzweig. Hunger artists: yeast adapted to carbon limitation show trade-offs under carbon sufficiency. *PLoS genetics*, 7(8):e1002202, 2011.
- [2] Anna Zakrzewska, Gerco Van Eikenhorst, Johanna EC Burggraaff, Daniel J Vis, Huub Hoefsloot, Daniela Delneri, Stephen G Oliver, Stanley Brul, and Gertien J Smits. Genome-wide analysis of yeast stress survival and tolerance acquisition to analyze the central trade-off between growth rate and cellular robustness. *Molecular biology of the cell*, 22(22):4435–4446, 2011.
- [3] Kiyan Shabestary, Cinzia Klemm, Benedict Carling, James Marshall, Juline Savigny, Marko Storch, and Rodrigo Ledesma-Amaro. Phenotypic heterogeneity follows a growth-viability tradeoff in response to amino acid identity. *Nature Communications*, 15(1):6515, 2024.
- [4] Albert F Bennett and Richard E Lenski. An experimental test of evolutionary trade-offs during temperature adaptation. *Proceedings of the National Academy of Sciences*, 104(suppl\_1):8649–8654, 2007.
- [5] Angus Buckling, Michael A Brockhurst, Michael Travisano, and Paul B Rainey. Experimental adaptation to high and low quality environments under different scales of temporal variation. *Journal of Evolutionary Biology*, 20(1):296–300, 2007.
- [6] Maryl Lambros, Ximo Pechuan-Jorge, Daniel Biro, Kenny Ye, and Aviv Bergman. Emerging adaptive strategies under temperature fluctuations in a laboratory evolution experiment of escherichia coli. *Frontiers in microbiology*, 12:724982, 2021.

- [7] Luis López-Maury, Samuel Marguerat, and Jürg Bähler. Tuning gene expression to changing environments: from rapid responses to evolutionary adaptation. *Nature Reviews Genetics*, 9(8):583–593, 2008.
- [8] Mikhail Tikhonov, Shamit Kachru, and Daniel S Fisher. A model for the interplay between plastic tradeoffs and evolution in changing environments. *Proceedings of the National Academy of Sciences*, 117(16):8934–8940, 2020.
- [9] Charuhansini Tvishamayi, Farhan Ali, Nandita Chaturvedi, Nithila Madhu-Kumar, Zeenat Rashida, Chandan Muni Reddy, Ankita Ray, Stephan Herminghaus, and Shashi Thutupalli. Convergent cellular adaptation to freeze-thaw stress via a quiescence-like state in yeast. *eLife*, July 2025.
- [10] Christoffer Olsson and Jan Swenson. Structural comparison between sucrose and trehalose in aqueous solution. *The Journal of Physical Chemistry B*, 124(15):3074–3082, 2020.
- [11] Juan S Aranda, Edgar Salgado, and Patricia Taillandier. Trehalose accumulation in *saccharomyces cerevisiae* cells: experimental data and structured modeling. *Biochemical Engineering Journal*, 17(2):129–140, 2004.
- [12] Hugo Tapia, Lindsey Young, Douglas Fox, Carolyn R Bertozzi, and Douglas Koshland. Increasing intracellular trehalose is sufficient to confer desiccation tolerance to *saccharomyces cerevisiae*. *Proceedings of the National Academy of Sciences*, 112(19):6122–6127, 2015.
- [13] Juan Carlos Argüelles. Physiological roles of trehalose in bacteria and yeasts: a comparative analysis. *Archives of microbiology*, 174:217–224, 2000.
- [14] Anita Panek. Function of trehalose in baker’s yeast (*saccharomyces cerevisiae*). *Archives of Biochemistry and Biophysics*, 100(3):422–425, 1963.
- [15] L Diniz-Mendes, E Bernardes, Pedro Soares De Araujo, AD Panek, and VMF Paschoalin. Preservation of frozen yeast cells by trehalose. *Biotechnology and Bioengineering*, 65(5):572–578, 1999.
- [16] Lei Shi, Benjamin M Sutter, Xinyue Ye, and Benjamin P Tu. Trehalose is a key determinant of the quiescent metabolic state that fuels cell cycle progression upon return to growth. *Molecular biology of the cell*, 21(12):1982–1990, 2010.
- [17] Monika Opalek, Bogna Smug, Michael Doeblei, and Dominika Wloch-Salamon. On the ecological significance of phenotypic heterogeneity in microbial populations undergoing starvation. *Microbiology Spectrum*, 10(1):e00450–21, 2022.
- [18] Dominique Chu and David J Barnes. The lag-phase during diauxic growth is a trade-off between fast adaptation and high growth rate. *Scientific reports*, 6(1):25191, 2016.
- [19] Markus Basan, Tomoya Honda, Dimitris Christodoulou, Manuel Hörl, Yu-Fang Chang, Emanuele Leoncini, Avik Mukherjee, Hiroyuki Okano, Brian R Taylor, Josh M Silverman, et al. A universal trade-off between growth and lag in fluctuating environments. *Nature*, 584(7821):470–474, 2020.
- [20] Shay Stern, Tali Dror, Elad Stolovicki, Naama Brenner, and Erez Braun. Genome-wide transcriptional plasticity underlies cellular adaptation to novel challenge. *Molecular Systems Biology*, 3(1):106, 2007.
- [21] Nitnipa Soontorngun. Reprogramming of nonfermentative metabolism by stress-responsive transcription factors in the yeast *saccharomyces cerevisiae*. *Current genetics*, 63:1–7, 2017.
- [22] Claudio De Virgilio. The essence of yeast quiescence. *FEMS microbiology reviews*, 36(2):306–339, 2012.
- [23] Isabelle Sagot and Damien Laporte. The cell biology of quiescent yeast—a diversity of individual scenarios. *Journal of Cell Science*, 132(1):jcs213025, 2019.
- [24] Linda L Breeden and Toshio Tsukiyama. Quiescence in *saccharomyces cerevisiae*. *Annual Review of Genetics*, 56(1):253–278, 2022.

- [25] Anthony D Aragon, Angelina L Rodriguez, Osorio Meirelles, Sushmita Roy, George S Davidson, Phillip H Tapia, Chris Allen, Ray Joe, Don Benn, and Margaret Werner-Washburne. Characterization of differentiated quiescent and nonquiescent cells in yeast stationary-phase cultures. *Molecular biology of the cell*, 19(3):1271–1280, 2008.
- [26] Bertrand Daignan-Fornier and Isabelle Sagot. Proliferation/quiescence: the controversial “aller-retour”. *Cell Division*, 6:1–4, 2011.
- [27] James R Valcourt, Johanna MS Lemons, Erin M Haley, Mina Kojima, Olukunle O Demuren, and Hilary A Coller. Staying alive: metabolic adaptations to quiescence. *Cell cycle*, 11(9):1680–1696, 2012.
- [28] Paul K Herman. Stationary phase in yeast. *Current opinion in microbiology*, 5(6):602–607, 2002.
- [29] Chris Allen, Sabrina Büttner, Anthony D Aragon, Jason A Thomas, Osorio Meirelles, Jason E Jaetao, Don Benn, Stephanie W Ruby, Marten Veenhuis, Frank Madeo, et al. Isolation of quiescent and nonquiescent cells from yeast stationary-phase cultures. *The Journal of cell biology*, 174(1):89–100, 2006.
- [30] Margaret Werner-Washburne, Edward Braun, Gerald C Johnston, and Richard A Singer. Stationary phase in the yeast *Saccharomyces cerevisiae*. *Microbiological reviews*, 57(2):383–401, 1993.
- [31] Tomás Alarcón and Henrik Jeldtoft Jensen. Quiescence: a mechanism for escaping the effects of drug on cell populations. *Journal of The Royal Society Interface*, 8(54):99–106, 2011.
- [32] Xia Wang, Kotaro Fujimaki, Geoffrey C Mitchell, Jungeun Sarah Kwon, Kimiko Della Croce, Chris Langsdorf, Hao Helen Zhang, and Guang Yao. Exit from quiescence displays a memory of cell growth and division. *Nature communications*, 8(1):321, 2017.
- [33] Siyu Sun and David Gresham. Cellular quiescence in budding yeast. *Yeast*, 38(1):12–29, 2021.
- [34] Yoav Kaplan, Shaked Reich, Elyaqim Oster, Shani Maoz, Irit Levin-Reisman, Irine Ronin, Orit Gefen, Oded Agam, and Nathalie Q Balaban. Observation of universal ageing dynamics in antibiotic persistence. *Nature*, 600(7888):290–294, 2021.
- [35] Gabrielle Woronoff, Philippe Nghe, Jean Baudry, Laurent Boitard, Erez Braun, Andrew D Griffiths, and Jérôme Bibette. Metabolic cost of rapid adaptation of single yeast cells. *Proceedings of the National Academy of Sciences*, 117(20):10660–10666, 2020.
- [36] Manlu Zhu and Xiongfeng Dai. Shaping of microbial phenotypes by trade-offs. *Nature Communications*, 15(1):4238, 2024.
- [37] Frank J Bruggeman, Bas Teusink, and Ralf Steuer. Trade-offs between the instantaneous growth rate and long-term fitness: consequences for microbial physiology and predictive computational models. *Bioessays*, 45(10):2300015, 2023.
- [38] Leonie J Jahn, Christian Munck, Mostafa MH Ellabaan, and Morten OA Sommer. Adaptive laboratory evolution of antibiotic resistance using different selection regimes lead to similar phenotypes and genotypes. *Frontiers in microbiology*, 8:816, 2017.
- [39] Diarmaid Hughes and Dan I Andersson. Selection of resistance at lethal and non-lethal antibiotic concentrations. *Current opinion in microbiology*, 15(5):555–560, 2012.
- [40] Linus Sandegren. Selection of antibiotic resistance at very low antibiotic concentrations. *Upsala journal of medical sciences*, 119(2):103–107, 2014.
- [41] Michael Manhart, Bharat V Adkar, and Eugene I Shakhnovich. Trade-offs between microbial growth phases lead to frequency-dependent and non-transitive selection. *Proceedings of the Royal Society B: Biological Sciences*, 285(1872):20172459, 2018.
- [42] Stefany Moreno-Gámez, Daniel J Kiviet, Clément Vulin, Susan Schlegel, Kim Schlegel, G Sander van Doorn, and Martin Ackermann. Wide lag time distributions break a trade-off between reproduction and survival in bacteria. *Proceedings of the National Academy of Sciences*, 117(31):18729–18736, 2020.

- [43] Ayşe Nisan Erdoğan, Pouria Dasmeh, Raymond D Socha, John Z Chen, Benjamin E Life, Rachel Jun, Linda Kiritchkov, Dan Kehila, Adrian WR Serohijos, and Nobuhiko Tokuriki. Neutral drift upon threshold-like selection promotes variation in antibiotic resistance phenotype. *Nature Communications*, 15(1):10813, 2024.
- [44] Shimon Bershtain, Adrian WR Serohijos, and Eugene I Shakhnovich. Bridging the physical scales in evolutionary biology: from protein sequence space to fitness of organisms and populations. *Current opinion in structural biology*, 42:31–40, 2017.
- [45] Jacob A Moorad and Timothy A Linksvayer. Levels of selection on threshold characters. *Genetics*, 179(2):899–905, 2008.
- [46] Robert A Krebs and Volker Loeschke. Estimating heritability in a threshold trait: heat-shock tolerance in drosophila buzzatii. *Heredity*, 79(3):252–259, 1997.
- [47] David B Berry, Qiaoning Guan, James Hose, Suraiya Haroon, Marinella Gebbia, Lawrence E Heisler, Corey Nislow, Guri Giaever, and Audrey P Gasch. Multiple means to the same end: the genetic basis of acquired stress resistance in yeast. *PLoS genetics*, 7(11):e1002353, 2011.
- [48] Alexander V Badyaev. Stress-induced variation in evolution: from behavioural plasticity to genetic assimilation. *Proceedings of the Royal Society B: Biological Sciences*, 272(1566):877–886, 2005.
- [49] Vidyanand Nanjundiah. Phenotypic plasticity and evolution by genetic assimilation. *Origination of organismal form: Beyond the gene in developmental and evolutionary biology*, 2:245, 2003.
- [50] Thomas K Wood, Stephen J Knabel, and Brian W Kwan. Bacterial persister cell formation and dormancy. *Applied and environmental microbiology*, 79(23):7116–7121, 2013.
- [51] Edo Kussell, Roy Kishony, Nathalie Q Balaban, and Stanislas Leibler. Bacterial persistence: a model of survival in changing environments. *Genetics*, 169(4):1807–1814, 2005.
- [52] Nathalie Q Balaban, Jack Merrin, Remy Chait, Lukasz Kowalik, and Stanislas Leibler. Bacterial persistence as a phenotypic switch. *Science*, 305(5690):1622–1625, 2004.
- [53] Bram Van den Bergh, Joran E Michiels, Tom Wenseleers, Etthel M Windels, Pieterjan Vanden Boer, Donaat Kestemont, Luc De Meester, Kevin J Verstrepen, Natalie Verstraeten, Maarten Fauvart, et al. Frequency of antibiotic application drives rapid evolutionary adaptation of escherichia coli persistence. *Nature microbiology*, 1(5):1–7, 2016.
- [54] Ofer Fridman, Amir Goldberg, Irine Ronin, Noam Shores, and Nathalie Q Balaban. Optimization of lag time underlies antibiotic tolerance in evolved bacterial populations. *Nature*, 513(7518):418–421, 2014.
- [55] Monika Opalek, Hanna Tutaj, Adrian Pirog, Bogna J Smug, Joanna Rutkowska, and Dominika Włoch-Salamon. A systematic review on quiescent state research approaches in s. cerevisiae. *Cells*, 12(12):1608, 2023.
- [56] Gregory J Velicer and Richard E Lenski. Evolutionary trade-offs under conditions of resource abundance and scarcity: experiments with bacteria. *Ecology*, 80(4):1168–1179, 1999.
- [57] Minna Pekkonen, Tarmo Ketola, and Jouni T Laakso. Resource availability and competition shape the evolution of survival and growth ability in a bacterial community. *PLoS One*, 8(9):e76471, 2013.
- [58] Yusuke Himeoka and Namiko Mitarai. Dynamics of bacterial populations under the feast-famine cycles. *Physical Review Research*, 2(1):013372, 2020.
- [59] Rintaro Niimi, Chikara Furusawa, and Yusuke Himeoka. Population dynamics of generalist/specialist strategies in the feast-famine cycle. *bioRxiv*, pages 2025–05, 2025.
- [60] Silja Borring Låstad and Namiko Mitarai. Triggered and spontaneous dormancy in bacteria during feast-famine cycles with stochastic antibiotic application. *bioRxiv*, pages 2025–02, 2025.