THE ROLE OF RIBOSOME AUTOCATALYSIS AND FEEDBACK IN RESOURCE COMPETITION

Fiona A Chandra* and Domitilla Del Vecchio
Massachusetts Institute of Technology
Cambridge, MA 02139

Abstract

Resource competition, and primarily competition for ribosomes, can lead to unexpected behavior in genetic circuits as "hidden" interactions arise. However, ribosomes are also autocatalytic since they are partially made of proteins and autocatalysis can have detrimental effects on a system's stability and/or robustness. Additionally, there are known feedback regulations on ribosome synthesis such as inhibition of rRNA synthesis via ppGpp. Here, we develop a minimal model of ribosome and protein synthesis, which includes autocatalysis and feedback, to investigate conditions under which these regulatory actions may have a significant effect in situations of increased ribosome demand.

Keywords

Ribosome, Autocatalysis, Resource competition.

Introduction

Ribosomes as a limited resource have gained renewed attention lately with studies on resource limitations in synthetic biology (Stanton, 2014). Competition for ribosomes, can lead to unexpected behavior in genetic circuits as "hidden" interactions arise (Del Vecchio, 2014, Qian, 2015). The expression of a constitutive gene may decrease when the expression of another gene is increased due to sequestration of common resources such as RNA polymerases and ribosomes (Gyorgy, 2015).

Current models assume a constant production of ribosomes and these models describe the experimental results well (Gyorgy, 2014). However, ribosomes are also autocatalytic since they are partially made of proteins.

Autocatalysis has been shown to have detrimental effects on a system's stability and/or robustness (Chandra, 2011). In this paper, we explore the effects of the autocatalysis of ribosome production on the system's steady state behavior and stability using minimal models of protein translation. We also explore the effects of feedback regulation on ribosome production.

Our modeling results show that while the effects of increasing synthetic demand on the total concentration of ribosomes vary between the system with or without autocatalysis, the effect on the free ribosomes and thus on constitutive genes is negligible. This suggests that the previous model of resource competition using a constant ribosome pool is indeed good enough for the purposes of studying resource competition. However, autocatalysis changes the stability of the system high demand. We also see that feedback regulation ameliorates the effects of resource competition.

Model

We start with a minimal model describing the translation of free ribosomes (R) and non-ribosomal proteins (P). The production of both R and P depends on R, and R is sequestered by binding to different mRNA molecules in the cells, including mRNA of ribosomal proteins (m_{RT}), mRNA of the synthetic gene which

* To whom all correspondence should be addressed

constitutes the demand (m_{PT}), and other cellular mRNA (m_C), yielding

$$\dot{R} = \frac{Vk_t R m_{RT}}{K_D + R} - \delta \frac{R}{K_D + R}(m_{RT} + m_{PT} + m_C) - \delta R$$
$$\dot{P} = \frac{k_t R m_{PT}}{K_D + R} - \delta P \qquad (1)$$

Description of other parameters is given in Table 1. The non-autocatalytic model is similar to Eq. (1) except R is produced at a constant rate Vr.

We also explore the effects of negative feedback, adding inhibition by R on its own translation (the equation for P remains the same as in Eq. (1)):

$$\dot{R} = \frac{Vk_t R m_{RT}}{(K_D + R)(K_I + R)} - \delta R - \frac{\delta R}{K_D + R}(m_{RT} + m_{PT} + m_C) \qquad (2)$$

Table 1. Model parameters

Parameter	Description
k_t	Translation rate
δ	Dilution rate
K_D	Effective translation dissociation constant
K_I	Effective inhibition constant
V	Maximal production rate

Results

At low demand, the change in free ribosome level due to increasing *mPT* remains very similar for both autocatalytic and non-autocatalytic systems. At higher demand, while R in the non-autocatalytic system continuously approaches zero, the autocatalytic system bifurcates, giving rise to a steady state at $R=0$ (Figure 1). In particular, when

$$Vk_t m_{RT} - \delta(m_{RT} + m_C + K_D) > \delta m_{PT} \qquad (3)$$

Eq. (1) has a stable positive equilibrium ($R>0$) and an unstable one at $R=0$. Otherwise, Eq. (1) goes through a transcritical bifurcation to a stable equilibrium at $R=0$ and an unstable negative ($R<0$) equilibrium. When there is feedback (Eq. (2)), the bifurcation point is given by:

$$Vk_t m_{RT} - K_I \delta(K_D + m_{RT} + m_C) < K_I \delta m_{PT} \qquad (4)$$

With strong feedback (low K_I), free ribosome concentration decreases more slowly and the bifurcation shifts to a higher demand. In comparing the three cases, we adjust V such that the initial ribosome level is the same, therefore V is much higher in Eq. (2), which also shifts the bifurcation point to a much higher demand m_{PT}.

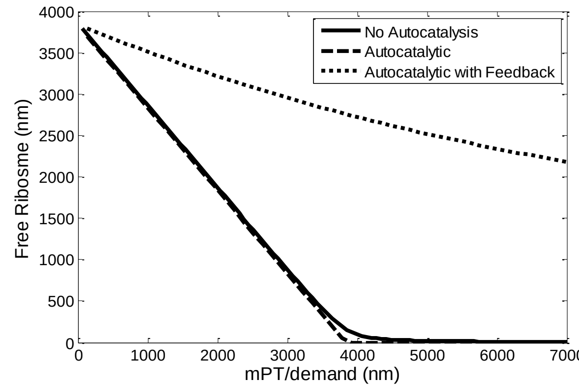

Figure 1. Free ribosome concentration as demand (m_{PT}) increases.

There is a more significant difference in total ribosome level between the three cases. As expected, total ribosome level in the non-autocatalytic case remains constant. In the autocatalytic case, however, while the total ribosome concentration doesn't change significantly at low demand, it immediately drops to zero close to the bifurcation point. Conversely, for the system with feedback, the total ribosome level actually increases (Figure 2). Since the presence of feedback increases the total ribosome concentration as demand increases, effects of ribosome competition are also reduced compared to those predicted by a model with constant ribosome production.

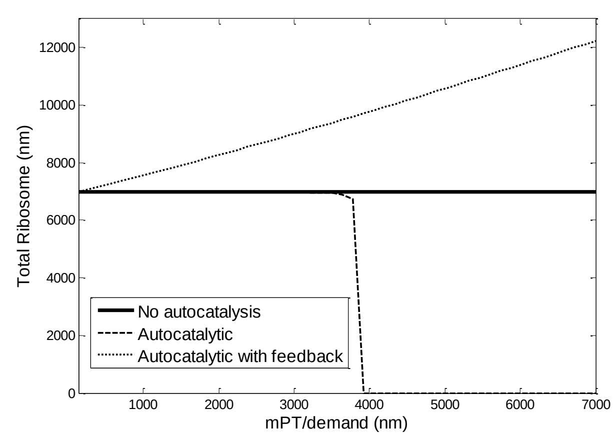

Figure 2. Total ribosome level as demand (m_{PT}) increases

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