The regulation of ribosome efficiency by nutrients and growth

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***Short Abstract* — (100 words) In microorganisms, ribosomal content and growth usually scale linearly, suggesting that translation is an efficient, rate-limiting process. However, we found that ribosomal content is not universal for a given growth rate: under phosphorus (P) limitation, cells produce the same number of proteins but have fewer ribosomes compared to carbon (C) or nitrogen (N) limitation. By combining experimental measurements and mathematical modeling, we discovered that the inefficiency in ribosome usage arises from premature translational termination. We propose a "gate control model": cells maintain a high influx of translational initiation but gate elongation. This regulation might support faster adaptation upon nutrient fluctuations.**

***Keywords* — Ribosome efficiency, Gate control model, Translational premature termination**

I. Extended Abstract

Resource allocation is a fundamental problem faced by organisms in evolution, which shapes phenotypes from animal behaviors to cellular physiology (Stoebel, Dean, & Dykhuizen, 2008). For single-cell organisms like *Escherichia coli*, how to allocate the limited amount of nutrients to different cellular components is critical to survival and fitness (Li, Burkhardt, Gross, & Weissman, 2014). It has long been known that *E. coli* cells exhibit a linear relationship between growth rate and ribosomal content, with increasing number of ribosomes supporting increasing growth rates (Bremer & Dennis, 2012; Schaechter, Maaløe, & Kjeldgaard, 1958). Linearity follows naturally if ribosomal content is always limiting for steady-state growth. This "efficient ribosome" hypothesis (Maaløe & Kjeldgaard, 1966) predicts that ribosomal level should reflect growth rate in all nutrient environments, and represents a simple strategy to achieve optimality in proteome allocation (You et al., 2013).

Surprisingly, however, we find that the ribosomal content of *E. coli* depends both on growth rate and nutrient limitation. At the same growth rates, phosphorus (P-) limited cells produce the same amount of protein but contain significantly fewer ribosomes than under carbon (C-) or nitrogen (N-) limitation. This inefficient ribosomal usage under C/N-limitation is due to an accumulation of mRNA-free ribosomes. We investigated the mechanism underlying the difference in ribosomal activity by building a macroscopic model based on flux-balance equations to capture the global dynamics of ribosomes. The model suggested that a large fraction of ribosomes prematurely terminate and do not complete translation in C- and N-limited cells. We then used ribosome profiling to seek experimental evidence for premature termination, and built a microscopic model to quantify the behaviors of a ribosome along the transcript. Together, these results support the presence of a high rate of premature termination (up to 50% under C- and N-), which is especially prevalent early in translation.

While not optimized for efficient ribosomal usage, this mechanism might promote rapid acceleration of growth when nutrients become abundant. Our macroscopic model predicted that C/N-limited cells should recover faster than P-limited cells upon nutrient repletion, and we experimentally validated this prediction.

II. Conclusion

Microorganisms are generally thought to outcompete their neighbors by optimizing growth and using cellular machineries efficiently. However, our finding that *E. coli* grows sub-optimally with inefficient ribosome usage suggests that cells do not always optimize growth for the current condition, but rather for future transitions when the extra ribosomes can quickly restore growth. These results add an unexpected nutrient-dependent layer of regulation to the well-characterized bacterial growth laws, and indicate a previously unappreciated role for premature termination in global protein production and growth rate.

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