Systematic Analysis of the Robustness in Complex Reaction Networks of Bacteria

Hiroyuki Kurata hkurata@bio.t.u-tokyo.ac.jp
Kazunari Taira taira@nair.go.jp

Department of Chemistry and Biotechnology, The University of Tokyo, Tokyo 113-8656, Japan

Keywords: robustness, reaction networks, bacteria

1 Introduction

Barkai and Leibler [1] demonstrated a robust property of adaptation behavior in bacterial chemotaxis, indicating that the robust adaptation was a consequence of the network's connectivity and the chemotaxis system did not require the fine-tuning of biochemical parameters. However, they did not answer the crucial question on whether it is possible to isolate such a subsystem from the whole system composed of heterogeneous and interactive networks and to analyze it separately. If the interaction among subsystems is sensitive, one cannot analyze smaller subsystems separately. If their interaction shows a robust property, one can extract the subsystems out of the whole system, analyzing them one by one. In *Escherichia coli* heat shock response [2], σ^{32} (encoded the *rpoH* gene) plays a major role in controlling expression of the heat shock protein genes encoding chaperones and proteases. The level of active σ^{32} is regulated by complex mechanisms: chaperone-mediated regulation of σ^{32} activity and stability, thermoregulated-translation induction of the *rpoH* mRNA, and protease-mediated σ^{32} degradation. The numerical framework model clarifies that complexity in σ^{32} regulation generates a robust property of *E. coli* heat shock response, thereby increasing the robustness of the interconnected factors among subsystems. Complexity seems to disturb isolating a smaller subsystem out of the whole biological system. Actually, complexity generates the robustness among subsystems, thereby making it possible to extract a smaller subsystem out of the whole system and analyze it separately.

2 Methods and Results

Complexity, performance (yield and efficiency), and robustness are defined to analyze heat shock response system. Complexity in the σ^{32} regulation is generated from three processes, (1) feedback control: chaperone (DnaK)-mediated sequestering σ^{32} away from binding to RNAP core enzyme, and chaperone (DnaK)-mediated degradation of σ^{32} and its nascent polypeptideon the *rpoH* mRNA, (2) feedforward control: heat-induced translation of the *rpoH* mRNA, (3) autogenous control (named because it seemed self-loop σ^{32} degradation): degradation of σ^{32} by σ^{32}-expressed FtsH protease. The degree of complexity is adjusted by changing the combination of these three processes. Performance of the *E. coli* heat shock response system is characterized by yield and efficiency. Since the aim of heat shock response is considered to reduce free unfolded proteins, yield is defined by: Yield $= 1 - \frac{\text{free } \mathrm{P_{un}}}{\text{Total P}}$, where ($\mathrm{P}_{un}$= unfolded protein, P=protein). An excess amount of chaperone refolds proteins sufficiently, but it loads the system. Efficiency is defined by: Efficiency $= 1 - \frac{\text{free DnaK}}{\text{Total DnaK}}$, where free DnaK neither involves refolding process nor binding to σ^{32}. Efficiency means how efficiently heat shock response is regulated by a minimum amount of chaperone (DnaK).

Calculations involving differential equations and simultaneous nonlinear equations were performed by the Runge-Kutta method and by the Newton-Raphson method, respectively. Computer programs

in C language and Message-Passing Interface were executed on sixty-four CPUs of a super parallel computer SR2201 (HITACHI, TOKYO). Heat shock occurred at 50 min. The mathematical simulation sampled the yield and efficiency at 50 min and 150 min at low and high temperatures, respectively, when the response reached the steady state.

To determine the robustness in the heat shock response, the two-dimensional feedback control parameter space, consisting of the binding association constant of DnaK and σ^{32} and the degradation rate of DnaK-bound σ^{32}, were searched to provide the required performance (efficiency > 0.8 and yield > 0.995). Figure 1 shows that complexity in σ^{32} regulation enlarges the feedback control parameter space. By comparing B with A, the addition of feedforward control increased the feedback control parameter space, *i.e.*, increasing the robust property. In C, the addition of autogenous control, consisting of two parameters, binding association constant between σ^{32} and FtsH and degradation rate of FtsH-bound σ^{32}, further enlarged the feedback control parameter space. It had been hard to understand the self-loop-like function that σ^{32}-expressed FtsH protease directly degrades σ^{32}, but system analysis showed the capability of autogenous control to increase the robustness for the feedback control parameters. Note, however, that the addition of the autogenous control cannot enhance the robustness without the feedforward control (data not shown). These simulations predicted that complexity in σ^{32} regulation generates the robust property of the interaction between σ^{32} and DnaK, thereby clarifying that the interaction between the heat shock response and other subsystems is robust (data not shown).

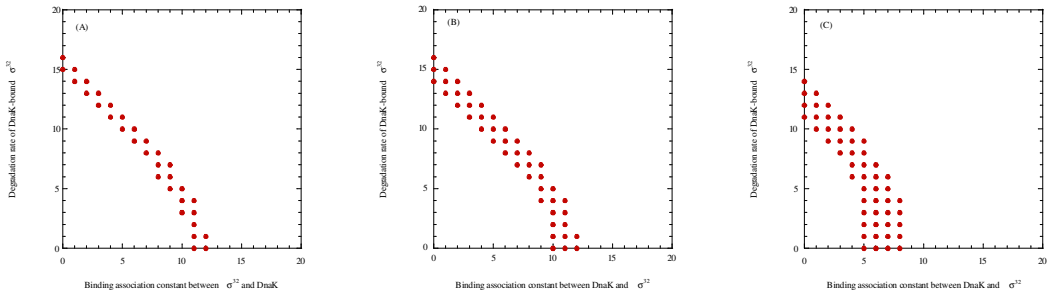

Figure 1: Two-dimensional space of the feedback control parameters. The whole space as follows: the x-axis (binding association constant between σ^{32} and DnaK) is $10^4 \times 2^x M^{-1}$ ($x = 0, 1, 2, 3, \ldots, 19$), the y-axis (degradation rate of DnaK-bound σ^{32}) is $0.01 \times 2^y \,\mathrm{min}^{-1}$ ($y = 0, 1, 2, 3, \ldots, 19$), is searched to provide the performance (efficiency > 0.8 and yield > 0.995), while the feedforward control parameter (translation efficiency) and the autogenous control parameters are varied. The efficiency and yield were calculated at low and high temperature. (A) Regulation by feedback control. (B) Regulation by feedback and feedforward controls. The translation efficiency is employed to provide the most enlarged space for the feedback control parameters. (C) Regulation by feedback, feedforward, and autogenous controls. The feedforward and autogenous control parameters are used to provide enlarged space for the feedback control parameter.

References

[1] Barkai, N. and Leibler, S., Robustness in simple biochemical networks, *Nature*, 387(6636):913–917, 1997.

[2] Yura, T., Regulation and conservation of the heat-shock transcription factor σ^{32}, *Genes to Cells,* 1:277–284, 1996.