

# Robustness Analysis of a Self-Oscillating Molecular Network in Dictyostelium Discoideum

L. Ma  
Electrical & Computer Engng  
The Johns Hopkins University  
Baltimore, MD 21218  
lma@jhu.edu

P.A. Iglesias  
Electrical & Computer Engng  
The Johns Hopkins University  
Baltimore, MD 21218  
pi@jhu.edu

## ABSTRACT

During the interphase between growth and full aggregation-competence, cells of *Dictyostelium discoideum* develop the ability to undergo synchronous periodic activities. Spontaneous light-scattering oscillations can be measured a few hours after the initiation of development [1, 3]. A model of molecular network that can account for the spontaneous oscillations of adenosine, 3', 5'-cyclic monophosphate (cAMP) observed in the field of chemotactic *D. discoideum* has been proposed by Laub and Loomis [2].

In their model of the aggregation network, pulses of cAMP are produced when ACA is activated after the binding of extracellular cAMP to the surface receptor CAR1. Ligand-bound CAR1 activates the protein kinase ERK2 that transmits the signal to ACA. When cAMP accumulates internally, it activates the protein kinase PKA. ERK2 is inactivated by PKA and no longer inhibits the cAMP phosphodiesterase REG A. A protein phosphatase activates REG A such that REG A can hydrolyze internal cAMP. CAR1 is phosphorylated when PKA is activated, leading to loss-of-ligand binding. When the internal cAMP is hydrolyzed by REG A, PKA activity is inhibited by its regulatory subunit, and protein phosphates returns CAR1 to its high-affinity state. Changes in the enzymatic activities are modeled as a set of nonlinear differential equations with kinetic constants  $k_n$  ( $n = 1 - 14$ , unit:  $min^{-1}$ ):

$$\begin{aligned}d[ACA]/dt &= k_1[ERK2]-k_2[ACA] \\d[PKA]/dt &= k_3[\text{internal cAMP}]-k_4[PKA] \\d[ERK2]/dt &= k_5[CAR1]-k_6[ERK2][PKA] \\d[REG A]/dt &= k_7-k_8[REG A][ERK2] \\d[\text{internal cAMP}]/dt &= k_9[ACA]-k_{10}[REG A][\text{internal cAMP}] \\d[\text{external cAMP}]/dt &= k_{11}[ACA]-k_{12}[\text{external cAMP}] \\d[CAR1]/dt &= k_{13}[\text{external cAMP}]-k_{14}[CAR1][PKA]\end{aligned}$$

Spontaneous oscillation appears at nominal parameter value:  $k_1 = 1.4$ ,  $k_2 = 0.9$ ,  $k_3 = 2.5$ ,  $k_4 = 1.5$ ,  $k_5 = 0.6$ ,  $k_6 = 0.8$ ,  $k_7 = 2.0$ ,  $k_8 = 1.3$ ,  $k_9 = 0.3$ ,  $k_{10} = 0.8$ ,  $k_{11} = 0.7$ ,  $k_{12} = 4.9$ ,  $k_{13} = 20$ ,  $k_{14} = 1.5$ . In [2] it is claimed that each of these 14 parameters is 25-fold robust. Using numerical bifurcation analysis package AUTO, however, we found that the network is far less robust than claimed. The bifurcation diagrams illustrate that Hopf bifurcations occur for each parameter, i.e. the spontaneous oscillation exists in a limited

range of each parameter. Use the robustness indicated by the ratio between the maximum value and the minimum value of this range, we demonstrate that only 4 out of 14 parameters are 25-fold robust, namely  $k_1$ ,  $k_4$ ,  $k_7$  and  $k_{10}$ . Four parameters ( $k_2$ ,  $k_6$ ,  $k_{12}$ ,  $k_{14}$ ) are less than 10-fold robust and are called sensitive parameters. The most sensitive parameter is  $k_{12}$ , which is only 5-fold robust. Six parameters are 10-fold robust but not 25-fold robust. They are referred to as medium-sensitive parameters ( $k_3$ ,  $k_5$ ,  $k_8$ ,  $k_9$ ,  $k_{11}$ ,  $k_{13}$ ).

Bifurcation analysis shows that when the parameters are varied, the period of the network does not deviate much from the nominal period  $\sim 7$  min. This is consistent with the result in [2]. However, we found that the amplitude is sensitive to the variations in most of the parameters. Twofold changes in 6 parameters ( $k_2$ ,  $k_4$ ,  $k_7$ ,  $k_{10}$ ,  $k_{12}$ ,  $k_{14}$ ) make the oscillation attenuate to zero, which is infinity-fold decrease. And twofold changes in 3 parameters ( $k_1$ ,  $k_5$ ,  $k_{11}$ ) lead to more than 9-fold decrease. This contradicts the conclusion in [2] stating that twofold changes in the kinetic constants make little difference in the amplitude of the oscillation. Thus the network can be seen as robust only in the sense of frequency change.

## ACKNOWLEDGMENTS

This work was supported in part by the National Science Foundation, under grant DMS-0083500, and the Whitaker Foundation. Discussions with Dr. Wilson J. Rugh are gratefully acknowledged.

## REFERENCES

- [1] G. Gerisch and U. Wick. Intracellular oscillations and release of cyclic AMP from *Dictyostelium* cells. *Biochem. Biophys. Res. Commun.*, 65(1):364–370, 1975.
- [2] M. T. Laub and W. F. Loomis. A molecular network that produces spontaneous oscillations in excitable cells of *Dictyostelium*. *Mol. Biol. Cell*, 9:3521–3532, Dec. 1998.
- [3] W. Roos, C. Scheidegger, and G. Gerisch. Adenylate cyclase activity oscillations as signals for cell aggregation in *Dictyostelium discoideum*. *Nature*, 266(17):259–261, March 1977.