QUORUM SENSING IN SINGLE CELLS OF NEUROSPORA CRASSA

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ABSTRACT

The use of a microwell microfluidic device allows separating single cells and tracking single cells data. The measurement of single cell fluorescent intensity trajectories in the microwell device supported a deterministic quorum sensing model identified by ensemble methods for clock phase synchronization. A strong inference framework was used to test the communication mechanism in phase synchronization of quorum sensing versus cell-to-cell contact, and the results lent support for quorum sensing.

KEYWORDS: Circadian Rhythms, Microwell, Quorum Sensing, Genetic Network

INTRODUCTION

Collective behavior occurs on a variety of scales of biological organization. The study of synchronization of biological oscillators constitutes building blocks of understanding collective behavior. Previously, much attention has been paid to the cell communication at the macroscopic level (Duvall and Taghert, 2012). However, there is a lack of knowledge of how cells communicate at single cell level. This research focus on how single cells of *Neurospora Crassa* communicate and synchronize their circadian rhythm through exchanging signal molecules.

EXPERIMENT

A microwell microfluidic device to trap individual cells is constructed to test the quorum sensing model versus the contact model. Each well is 10 μm deep and 10 μm in diameter to trap one conidial cell of average size. In the experiment, two populations of cells that were 12 hours out of phase initially were mixed and then put into the microwell device. The cells were observed and tracked over 10 days, and their fluorescent intensities were recorded. Then the mixed cell data were separated into two separate clusters (CCG_1 and CCG_2) based on their initial phase difference (from 0 to 20 h). In addition, the genetic networks of *Neurospora Crassa* and ensemble modeling method (Deng, et al., 2016; Yu, et al., 2007) were also used to study how single cells communicate and synchronize their circadian rhythm through quorum sensing mechanism (Figure 1a and 1b).

RESULTS AND DISCUSSION

The averages of the single cell trajectories in CCG_1 and CCG_2 were calculated and labeled as CCG_1_obs and CCG_2_obs, respectively. The result showed that the trajectories of CCG_1_obs and CCG_2_obs oscillated at a period around 20 h and they synchronized with each other after 80 h (Figure 1c). The simulation results showed that the trajectories of CCG_1_obs and CCG_2_obs were in good agreement with their model ensemble averages for the quorum sensing model but had a problem in fitting the direct contact model (Figure 2).

CONCLUSION

Using fluorescent trajectory data of single cells from the microwell experiment, a strong inference framework was established to test a quorum sensing hypothesis versus a contact hypothesis for communication using ensemble methods. The results that isolated single cells showing phase synchronization provided strong evidence for the quorum sensing hypothesis and some information about the communication parameters that quantify quorum sensing.

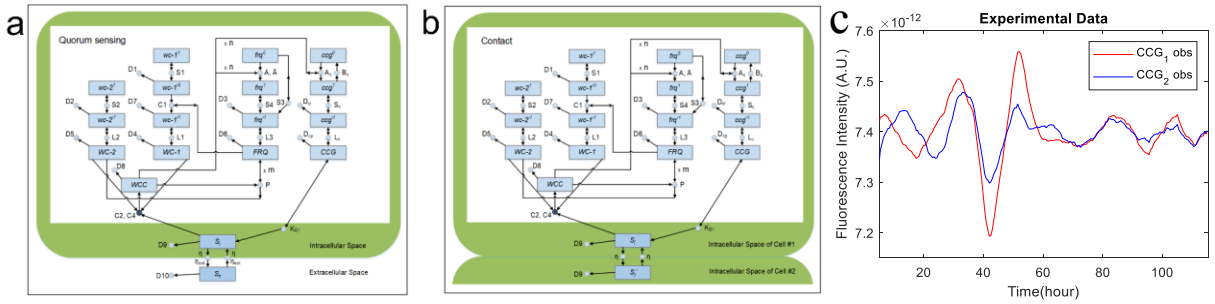

Figure 1: Genetic networks and experimental data. Quorum sensing and contact models for clock synchronization in single cells: (a) Quorum sensing model. (b) Contact model. (c) Plots of the experimental data show that trajectories were fully synchronized after 80 hours.

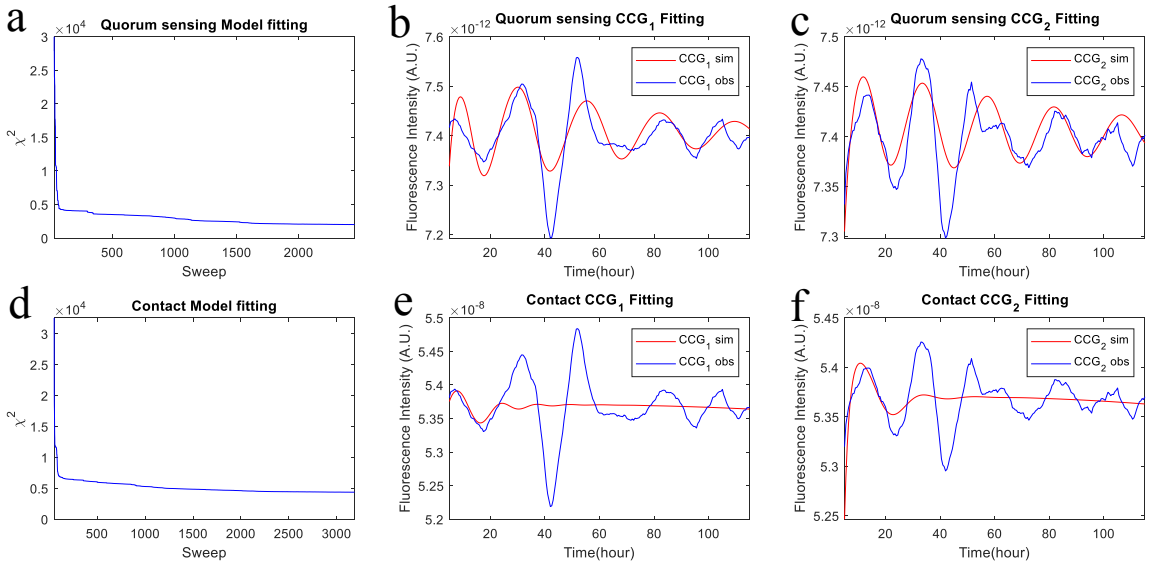

Figure 2: CCG_1 and CCG_2 trajectories fitted to an ensemble of deterministic models. (a) As a control on the MCMC experiment, the chi-squared statistic χ^2 was plotted as a function of sweep in quorum sensing model. (b and c) CCG_1 and CCG_2 average trajectories were in good agreement with their model ensemble averages for the quorum sensing model. (d) As a control on the MCMC experiment, the chi-squared statistic χ^2 was plotted as a function of sweep in contact model. (e and f) The simulation demonstrated that the measured fluorescence could fit the contact model for only the first oscillation.

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