# Stress-Induced Division of Labor Underlies Bacterial Colony Branching

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***Short Abstract* — Branching patterns of *Pseudomonas* colonies have been shown to depend on the secretion of biosurfactant. Here we show that starvation induces the differentiation of a *Pseudomonas* population to slow-growing cells that produce biosurfactant as a public good and fast-growing non-producers. Mathematical modeling indicates that this division of labor strategy is critical for explaining the diverse morphologies of branching patterns observed. We propose that the division of labor of branching bacterial colonies optimizes nutrient utilization and maximizes the population fitness by balancing between cell growth and motility. This collective mechanism for colony branching implies the role of population heterogeneity in pattern formation.**

***Keywords* — Patterning, division of labor.**

## I. Introduction

THE branching pattern is one of the most prevalent patterns found in nature [1]. How branching patterns emerge in living organisms has been a long standing question. The study of branching morphogenesis in bacterial colonies serves as a step stone to the understanding of branching pattern formation in multicellular organisms and also allows us to rationally design and control branching patterns using synthetic biology.

A number of mechanistic models for branching pattern formation in bacterial colonies have been proposed [2-5], but they are either not tested by experiments or not able to account for the diverse patterns under different experimental conditions. Based on mathematical modeling and experimental observations, here we show that under stress, *Pseudomonas aeruginosa* cells employ a division-of-labor strategy, which is necessary for explaining the various complex branching patterns observed in experiments.

## II. Results

The formation of branching patterns of *Pseudomonas* colonies has been shown to depend on the secretion of biosurfactants (such as rhamnolipids), which facilitates local cell swarming [6]. The expression of the rhamnolipid synthesis operon *rhlAB* is regulated by quorum sensing signaling and nutrient availability [7]. Flow cytometry of *Pseudomonas aeruginosa* PA14 cells expressing the *rhlAB* reporter showed that when nutrient was abundant, surfactant synthesis was turned off. Under starvation, however, a subpopulation of Pseudomonas cells with high biosurfactant expression quickly emerged. The metabolic burden of biosurfactant synthesis suggests that the population differentiates into cells producing biosurfactant as a public good at the expense of cell growth (Producers) and non-producers with higher proliferation rate (Growers).

Based on this observation, we developed a diffusion-reaction model that incorporates nutrient consumption, surfactant production, and the spatial-temporal dynamics of both subpopulations, while the transition rates between the two cell states are modulated by environmental conditions.

Simulations reveal that the division-of-labor scheme is essential for reproducing the wide range of branching patterns of *Pseudomonas* colonies under different conditions, as well as the unexpected observation of colony expansion against a nutrient gradient. Future work is needed to determine the evolutionary significance of this strategy and we propose that division-of-labor underlies the efficient adaptation of bacterial colony to various environments and ensures optimal resource exploitation which is manifested as colony branching.

## III. Significance

Division of labor and cell differentiation within communities of unicellular organisms were proposed to be the prelude to the emergence of multicellularity [8]. The population-level regulation during the morphogenesis of colony branching patterns may shed light on the design principles of pattern formation and provides insights to the control of heterogeneous cell communities such as biofilms and tumors.

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