Modeling a bacterial ecosystem through chemotaxis simulation of a single cell

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Abstract We present in this paper an artificial life ecosystem in which bacteria are evolved to perform chemotaxis. In this system, surviving bacteria have to overcome the problems of detecting resources (or sensing the environment), modulating their motion to generate a foraging behavior, and communicating with their kin to produce more sophisticated behaviors. A cell's chemotactic pathway is modulated by a hybrid approach that uses an algebraic model for the receptor clusters activity, an ordinary differential equation for the adaptation dynamics, and a metabolic model that converts nutrients into biomass. The results show some analysis of the motion obtained from some bacteria and their effects on the evolved population behavior. The evolutionary process improves the bacteria's ability to react to their environment, enhancing their growth and allowing them to better survive. As future work, we propose to investigate the effect of emergent bacterial communication as new species arise, and to explore the dynamics of colonies.

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1 Introduction

Artificial life research has made progress in the study of adaptive behavior through computational models of artificial organisms. Remarkably simple chemical reactions can perform movements toward attractants, and are in principle capable of modulating the behavior of artificial organisms. We will demonstrate in this work that a simple process of evolved bacterial chemotaxis can explain the emergence of more complicated behaviors seen in bacterial population dynamics. In terms of bacterial chemotaxis, this issue can be explored by looking at the influence of signaling network parameters on the spatiotemporal dynamics of bacteria that migrate toward chemical attractants and away from repellents. Cell chemotaxis is one of the simplest behaviors known, and it likely is one of the first behaviors to have existed in the history of life on earth. In bacterial chemotaxis such as that of *Escherichia Coli* (*E. Coli*), a bacterium typically tends to swim in a random walk, punctuated with periods of straight swimming sprints (or runs) interrupted by brief tumbles that causes the bacterium to change direction when the concentration of attractant or repellant is uniform or undetectable. In response to attractant gradient, this random walk becomes biased, and the bacteria tumble less frequently when encountering increasing attractant concentrations (i.e., they swim longer runs), and tumble more frequently when the attractant concentration decreases [1].

Phosphorylation cascade in a chemotactic pathway was simulated by a lot of works, in [2] a theoretical analysis of a full system of ODEs included phosphorylation

cascade, and other works focusing on chemotaxis bacteria [3, 4]. More recently, a bacterial chemoreceptor model was developed in [5]. The present work presents a bacterial ecosystem where individual E. Coli's chemotactic pathway is simulated by modeling signal processing by mixed chemoreceptor clusters using a rapid equilibrium (algebraic) model, adaptation through methylation simulated by ordinary differential equations (ODEs), and *E. Coli*'s characteristic flagellar motor-induced runs and tumbles [6]. The metabolism is modeled as a set of intracellular chemical reactions. The goal of this study is to demonstrate the importance of allowing to recycle dead cell matter released in the environment.

1.1 Bacterial chemotaxis

The chemotaxis process consists of three stages: chemoreception, signaling, and adaptation. Methyl-accepting chemotaxis proteins are located along the cell surface. These proteins act as chemoreceptors and bind with chemicals in the environment. If a nutrient attractant is detected outside of the cell, through MCP, the level of production of protein CheA decreases because the receptors state shifts to the off state. It has been shown that the activity of the receptor cluster depends on the local ligand concentration and the methylation level according to the MWC (Monod-Wyman-Changeux signal processing) model [7, 8]. CheA binds with phosphate in the cell. The phosphate group is transferred from the active CheA to the response regulator CheY. The concentration of CheYp modulates the motor and its behavior makes the cell run or tumble.

1.1.1 MWC model

We applied the MWC model for a mixed receptor cluster [7, 8]. The probability A that receptor cluster is active is dependent on ligand concentration and the methylation state of the receptors and calculated as

$$A = 1/(1 + e^{F}), \tag{1}$$

where $F = F_{\text{on}} - F_{\text{off}}$, and $F^{\text{on/off}}$ is the free energy of the cluster to be on/off as a whole. Hence, the average activity per receptor in the cluster is A. The total free-energy difference in the mean-field approximation is $F = n_r f_r(m)$, which is just the sum of the individual free-energy differences between the receptor on and off states.

$$f_r(m) = f_r^{\text{on}} - f_r^{\text{off}} = \epsilon_r(m) + \log\left(\frac{1 + [S]/K_r^{\text{on}}}{1 + [S]/K_r^{\text{off}}}\right), \tag{2}$$

where $[S]$ is the ligand concentration, and $K_r^{\text{on/off}}$ is the dissociation constant for the ligand in the on and off state, respectively.

1.1.2 Adaptation model

Adaptation is modeled according to the mean-field approximation of the assistance-neighborhood (AN) model [7, 9]. It is assumed that both enzymes work at saturation ([CheR]= 0.16, [CheB]= 0.28) [10]:

$$dm/dt = a(1 - A)\ [\text{CheR}] - bA\ [\text{CheB}] \tag{3}$$

The average methylation level evolves in time as

$$m(t + \Delta t) = m(t) + kV\Delta t \tag{4}$$

The parameter k indicates the adaptation rate relative to the wild-type adaptation rate V that is the rate of receptor methylation (see Eq. 3) [6].

1.1.3 Kinase activity

Both ligand binding and receptor methylation affect the activity of CheA. CheA kinase activity [6] is calculated as (varrint into [0,1])

$$\text{CheA} = \text{CheA}_{\text{tot}}\text{A}K_A/(\text{A}K_A + K_Y\text{CheY}_{\text{tot}}), \tag{5}$$

where A is the probability that receptor cluster is active, CheY_{tot} is the total CheY concentration that is equal to 9.7 according to [10], and K_A=5 and K_Y=100 are the rate constants according to [6].

1.1.4 CheY phosphorylation

The concentration of CheYp is obtained as a function of active CheA from the steady-state equation [11].

$$\text{CheY} = \text{CheY}_{\text{tot}}K_Y A/(K_Y\text{CheA} + K_Z\text{CheZ} + g_y), \tag{6}$$

where CheY_{tot} is the total CheY concentration, and CheZ is the total CheZ concentration, CheA is the active [CheA], $k_y = 100\,\mu\text{M}^{-1}\text{s}^{-1}$, $k_Z = 30/[\text{CheZ}]s^{-1}$, Y = 0.1 are the rate constants according to [11], [12].

1.1.5 The CCW motor bias

The CCW motor bias depends on CheYp concentration in the following form [13].

$$mb = \frac{mb_0}{\text{CheY}(1 - mb_0) + mb_0} \tag{7}$$

2 Bacterial metabolism

The metabolism is responsible for essential reproduction, growth, and development cycles, which are affected by gene expression and bacterial movement. An organism's genome contains instructions that encode the ability to metabolize one or more substrates present in the environment. Metabolism of a food either accelerates or

decelerates a bacterium's replication rate by a factor that is positive (nutrient) or negative (toxin), respectively. In this model, every bacterium is represented by a genome that encodes basic properties such as motion, energy absorption, toxin removal, and waste production. These properties are adjusted at each time step as the genome is expressed, and mutations are applied after each division. Forrest and Jones' simulation [14] allows for simple material cycling through absorption while agent is created and release agents die and their bodies deteriorate. We adopted a similar approach where the material within dying bacteria is released in the medium and available as a source of energy for surrounding bacteria. Metabolism is calculated as the organism's total energy (sum of constant basal energy and energy foraged via metabolized nutrients) and subtracted from the energy spent to tumble or run. This metabolic model allows bacteria to stabilize their energy consumption and eventually reach a splitting threshold. The bacterial energy cycle is implemented as follows:

$$\Delta M_t = (M_0 + A(M_F + M_W) + M_T + mb_0 M_M + M_S)\Delta t \tag{8}$$

- ΔM_t is the total metabolic expenditure (stepwise energy depletion rate);
- M_0 is the basal metabolic level (initial level at birth, equal to 25);
- A is the bacteria nutrient absorption rate (genetically encoded and described in the next section);
- M_F is the food source energetic value (2 units);
- M_W is the energetic value extracted from waste consumption (+1 unit);
- M_T is the metabolic cost of consuming toxins (−2);
- M_M is the metabolic cost of movement (−1 unit);
- mb_0 is the tumble frequency as a result of bacterial chemotaxis;
- M_S is the metabolic cost of cell division (Mt/2).

3 Genetic representation

In *E. Coli* chemotaxis, flagellar rotation is generated by an intracellular system of moderate complexity modulated by sensing the medium in which the bacterium lives. This system exhibits properties such as sensing, adaptation, memory, and motor modulation. To adjust these properties in order to simulate bacterial population behaviors, we use a genome that encodes factors involved in various levels of the metabolic pathway responsible for chemotaxis. Each gene in the genome is composed of two different types of encoding as is shown in Fig. 1. The first type is a binary encoding that is used in four genomic loci. It is used to encode variable levels of the bacterium's ability to detect nutrients or toxins with the same receptor. The signals

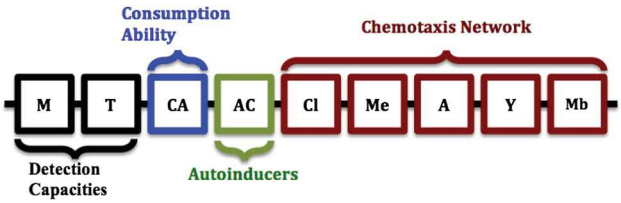

Fig. 1 The bacterium's genome

collected by these receptors constitute the environmental information fed to the chemical pathway generating chemotaxis. The values in M and T increase with the detected nutrient concentration. A is the bacterium's nutrient assimilation rate (consumption capacity), which affects its metabolism. The gene (AC) encodes a bacterium's sensitivity to detect autoinducer molecules from its own bacterial strain, allowing it to communicate with its own kind by secreting autoinducer molecules. The second type of encoding uses real numbers and is employed in the remainder of the genome. The following loci constitute the building blocks of the bacteria's chemotactic pathway: Cluster activity (CI), kinase activity, methylation level (Me), CheY phosphorylation (Y), and motor bias (Mb), which are employed in Eqs. (1–7).

4 Experimental results

The set of experiments presented here have been designed to answer the question of whether an agent-based simulation evolving simple bacterial chemotaxis can explain the emergence of more complicated behaviors at the population level. The work presented here makes a distinction between three types of chemical substances: (1) nutrients diffusing from multiple points in the environment, (2) nutrients released from dead bacteria (or waste), and (3) toxic compounds. Although the environment has been discretized, bacteria are free to move in the continuous two-dimensional space by translating their location. Each bacterium has one cell, with identical size, shape, and chemotactic network controlling its movements. The reader is invited to see [15] for the remaining parameters used in the setup of the chemotactic network. All runs presented in Fig. 2 (left) show a fast population growth in the first twenty simulation cycles, fueled by abundant nutrients and low levels of toxicity. This increase is followed by a high reproduction regime (or split operations) compensated by as many death events, which leads to a constant population size for about 200 generations. From generation 200 to 300, the sharp drop in population size is due to two factors: a high death rate caused by the depletion of food resources is compound with bad MCP and toxin avoidance abilities.

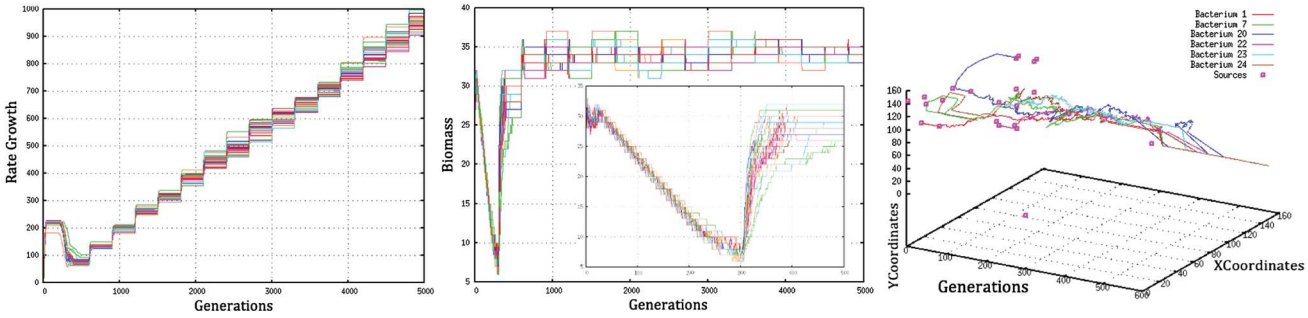

Fig. 2 *Left-hand* the 'Growth rate' runs, which we have replicated 30 times with quantitatively the same results, representing the optimal values of the whole of the bacteria for 5,000 steps. *Middle* the energy of the evolved population of bacteria for 30 runs at 5,000 generations. *Inside* the figure, a zoom in the same runs for 500 generations. *Right-hand* path realized by some bacteria in 2D space for the first 600 cycles

From generation 300, and at every 300 cycles, the growth rate often increases and is the result of the combined effect of individually improved genomic traits via mutation. The number of species varies greatly during an experiment, indicating that a high population turnover is taking place due to bacterial death and division. It is important to keep in mind that the speed of the whole population dynamics is not solely due to food abundance in the environment or to population size, but is for the most part due to evolved skills. We monitored the changes in total population energy for all 30 runs [Fig. 2, middle], where resources are being consumed, resulting in a decrease in the population's collective energy at the beginning of the run. Since cell divisions happen at very high frequency, this process accounts for most of the energy depletion in the first 300 generations. The simulation parameters have been adjusted so that within thirty generations, after most of the population died of starvation, a few survivors remain. In iteration 300, when new nutrient resources are added to the environment, the bacteria feed on it and multiply until there is no more nutrients in the environment, at which point their energy decreases again but more slowly than before. After thousands of time steps, the surviving bacteria are more efficient energy consumers and are able to keep a more stable energy level. This effectiveness is due to improved nutrient detection sensitivity (MCP), and to a greater extent to improved consumption ability. We also present some examples of trajectories from a few simulated swimming bacteria in Fig. 2-right. Each bacterium responds to the presence of nutrient sources with long runs and short tumbles, or by random walks in the absence of detectable traces of nutrients (Steady State). Several characteristic angles formed between two periods of long straight swimming (a 'run') marked by a period of abrupt change in orientation (a tumble) can be seen in the figure. These successive angles illustrate how bacteria can progress from their initial positions toward a preferred, nutrient-rich zone. Two characteristic evolutionary phases have been observed in this system: in the beginning (20) of the first 300 cycles, the bacteria first execute long runs from a nutrient source to another, but switch their oscillation bias to transition to a random walk when all resources are depleted.

5 Discussion

The results shown above demonstrate that a simple simulation of single-celled organisms with biological mechanisms such as simple chemical reactions allows us to model more complicated emergent behaviors at the population level. We observed in the results a healthy growth rate (or increasing bacterial count) for several hundreds of epochs, as long as the resources are abundant in the environment, and the population's collective lifespan increases because the evolved bacteria consume less energy, thanks to their optimized metabolism and their increased ability at gathering resources. This system is thus favorable to the emergence (or adaptation) of efficient sensory apparatus to detect food and avoid toxins, allowing individuals to better avoid deadly situations and improve reproduction allowing the population to thrive. When bacteria are moving, consuming, and splitting, their chemotaxis network is optimized in order to control their movement.

Figure 3 explains how the chemotaxis network responds to intracellular and environmental changes. Four internal states have been isolated in this response: *Steady State*, *Fully Inactive*, *Adapted*, and *FullyActive State*. For each of these states, the protein concentrations have been measured and compared to [6]. The *Steady State* is characterized by the bacteria performing a random walk and exploring the environment with initial parameter values of CheA =0.0164, methylation = 1.92, CheY = 1.92, and motor bias = 0.65. When the bacteria detect food sources, they enter a consumption regime where transmembrane

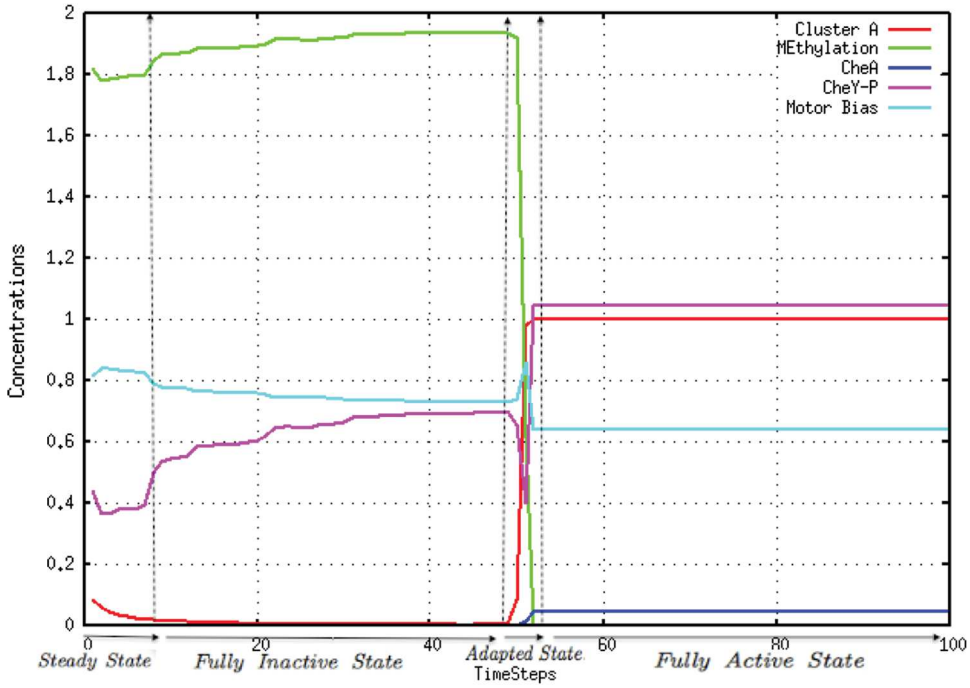

Fig. 3 Graph of variations in concentrations of proteins and enzymes used inside bacteria during the chemotaxis process

receptors sense the changes of attractant concentrations and become inactive. As the attractant binds to the receptors, it inhibits autophosphorylation activity of CheA. The CheY phosphorylated by the groups received from CheA (CheYp) diffuses to the flagellar motors and induces changes in motor rotation, causing the bacterium to run (move forward). The increase of attractant concentration switches the receptor to the 'off' state (i.e., *Fully Inactive State*), resulting in an initial fast decreases of kinase activity (CheA) (to 0.002) and hence CheY level, and causes longer runs (i.e., mb 0.75). The decrease of ChA activity is followed by a slow CheR-dependent adaptation. In the *Adapted State*, both *on* and *off* states are equiprobable, and both enzymes CheR and CheB are working for methylation and demethylation simultaneously. In this regime, methylation increases receptor ability to stimulate CheA activity. A removal of attractants shifts the system to the *on* state (or *Fully Active State*) that activates CheA autophosphorylation (0.047) and hence the downstream CheY phosphorylation.

6 Conclusion and future work

Our model was designed to model growth and behavior of bacterial ecosystems by simulating a group of bacterial cells in a stepwise fashion. To analyze and visualize the evolved behaviors, we present data that characterize bacterial positions, energy, and state in the cellular reproduction cycle. These results demonstrate that bacteria are able to evolve through mutation. The constructed model of chemotactic *E. coli* employed a hybrid model for simulating chemical pathways, with a mix of algebraic, ODE, and stochastic components instead of a fully stochastic model with an evolutionary algorithm to evolve a population of bacteria.

In future work, we will build upon the existing chemotaxis network to generate more diversified behaviors and capable bacteria that can emerge as distinct species with different behaviors to eventually form colonies. This platform will allow us to test our model with different environmental and non-environmental conditions.

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