Effect of Geometry and Bacterial Collisions on the Motion of Micro Bio Robots

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***Abstract*—Microbiological organisms, such as bacteria, are highly adaptable, thriving in extreme environments where humans cannot survive unaided. By harnessing these biomolecular motors, it is possible to create hybrid micro-bio-robots (MBRs) by engineering molecular networks in swimming bacteria and attaching them to inorganic microstructures. In this paper, we investigate MBR transport in a suspension of swimming bacteria, each driven by flagellar motors controlled by a biomolecular network. MBRs are propelled by collisions between free-swimming bacteria and microstructure boundaries. First, we use micro-gears and micro-chevrons to experimentally demonstrate that a MBR exhibits biased trajectories according to its geometry. Second, we develop a mathematical model to describe the dynamics of a microstructure propelled in a suspension of motile bacteria. This model will allow us to predict MBR trajectories and to develop improved strategies for MBR design and control.**

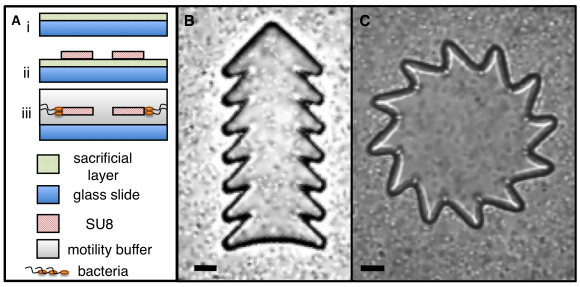

Fig. 1. (A) MBR microfabrication and design. (i) Glass slide spin coated with dextran (ii) SU8 patterning via photolithography (iii) MBRs release in a semi-dilute suspension of swimming bacteria and bacteria collide with walls. (B) Micro-chevron with boundary comprised of 14 teeth. (C) Micro-gear with boundary comprised of 12 teeth. Each scale bar is 15μm

I. Introduction

One of the significant challenges in microscale engineering is the development of controllable actuation methods for miniaturized systems. In nature, transport on the microscopic scale is achieved through biomolecular motors embedded in the cell bodies of bacteria. These biomolecular motors are self-powered and untethered making them capable of operating in hazardous or inaccessible environments. Previously, microorganisms have been shown to produce useful work in micro-engineered systems, and have been adapted to create an integrated micro-bio-robotic system [1]. Micro Bio Robots (MBRs) are controllable actuators driven by bacteria, with potential for cell manipulation, microassembly, and *in vivo* procedures, such as targeted cell drug delivery.

Recent studies have shown microscale transport by which a passive micro-object is actuated when immersed in a suspension of motile bacteria [2, 3, 4]. The object's motion results from collisions between swimming bacteria in solution and microstructure walls. Additionally, if the shape of the micro-object is asymmetric, geometrically biased trajectories can be achieved in bacterial suspensions. Experimentally, this has been demonstrated using micro-gears [2, 3, 4].

This work proposes developments toward an integrated MBR platform which relies on actuation provided by cell-wall collisions in a bacterial suspension. In particular, both an experimental framework and a mathematical model are under development in order to demonstrate the potential of propelling microstructures via stochastic collisions. Additionally, we examine the effect of microstructure shape on the overall trajectory of a MBR.

II. Micro Bio Robot Design and Experiments

The experimental platform used in this work builds upon the design of microbiorobots proposed in [5]. Microstructures were fabricated from SU8 negative photoresist and patterned via standard photolithographic methods (Figure 1A(i-ii)). Two types of microstructure geometries were designed in order to characterize MBRs driven via stochastic collisions. Both asymmetric micro-gears measuring 70μm in diameter (Figure 1B), and micro-chevrons measuring 80μm $\times$ 40μm were fabricated (Figure 1C). Each type of structure measured 10μm in thickness. Micro-gears and micro-chevrons were released in a small petri dish containing a semi-dilute suspension of *Serratia marcescens* and motility buffer (Figure 1A(iii)). Subsequent stochastic collisions between swimming bacteria and the out-of-plane microstructure walls resulted in MBR motion. In order to exploit a propulsion mechanism dependent on collisions between microstructure boundaries and free-swimming bacteria, an excess amount of surfactant was introduced to the solution, which prevented bacteria from attaching to the microstructure.

III. Multi-Scale Model

In order to model the interactions that govern the motion of a passive micro-object immersed in a bath of motile bacteria, we propose a multi-scale model which couples interactions

arising from molecular reactions occurring within the cell, to single-cell dynamics, and finally to cell-object interactions. Free-swimming bacteria are modeled using *Escherichia coli* as a reference bacterium.

A. Chemotactic Network

Chemotactic signaling pathways in *E. coli* are responsible for the physical behavior of the flagellar bundle in each bacterium. In order to model each flagellum in simulation, we consider the molecular mechanisms of the chemotaxis network which involve chemical reactions of kinases, proteins, and enzymes. These reactions govern transmission of cellular messages, control of the flagellar motor, and adaptation to different concentrations of chemical gradients. In particular, chemoreceptors called methyl-accepting chemotaxis proteins (MCPs), control directed cell locomotion through the regulation of the kinase CheA [6]. The flagellar motor can rotate in either a clockwise or counterclockwise direction, and is controlled through the phosphorylation of a response regulator by CheA [6]. Our simulation will model this chemoreceptor signaling pathway in order to demonstrate the cell's physical behavior when swimming through spatial chemical gradients.

B. Free-Swimming Bacteria

Operating in low Reynolds number environments, *E. coli* propel themselves by rotating helical flagellar filaments [7]. Motile *E. coli* can operate in either a "run" or a "tumble" state. When the flagella rotates in a counterclockwise direction (when viewed from behind), the cell swims with near constant linear velocity, and its trajectory is called a run. Conversely, when the flagella rotates in a clockwise direction, the flagellar filaments unbundle and the cell is subject to a random reorientation [7]. In the absence of chemical gradients, the process of switching states between a run and a tumble may be modeled by a Poisson process [8]. The rate for a cell to change state from a run to a tumble is $10s^{-1}$, whereas the rate for a cell to change state from a tumble to a run is $1s^{-1}$ [9].

In our simulation, each cell is modeled as a prolate spheroid of length $\ell = 3\mu m$, and is described by a position vector $\mathbf{r}$, and an orientation vector $\hat{\mathbf{e}}$, in the free-swimming direction [10]. We prescribe an effective thrust to each swimming cell which is linearly related to the free-swimming cell velocity through the mobility coefficient acting in the $\hat{\mathbf{e}}$ direction. Typically, we apply the maximum propulsive force that a cell may exert, which is approximately $0.45pN$ [11]. The propulsive force is characterized in both the run and tumble states by the following:

$$f_0 = \begin{cases} v_0/m_{\parallel} & \text{if cell is running} \\ 0 & \text{if cell is tumbling} \end{cases}$$

The resulting equations of motion of a swimming cell are described by Equation (1), where $m_{\parallel}$ and $m_{\perp}$ represent the cell's resistance to translation with respect to $\hat{\mathbf{e}}$.

$$\begin{bmatrix} \frac{dx}{dt} \\ \frac{dy}{dt} \end{bmatrix} = \begin{bmatrix} m_{\perp} & 0 \\ 0 & m_{\parallel} \end{bmatrix} \begin{bmatrix} F_x \\ F_y \end{bmatrix}, \quad \mathbf{F} = f_0 \hat{\mathbf{e}} \tag{1}$$

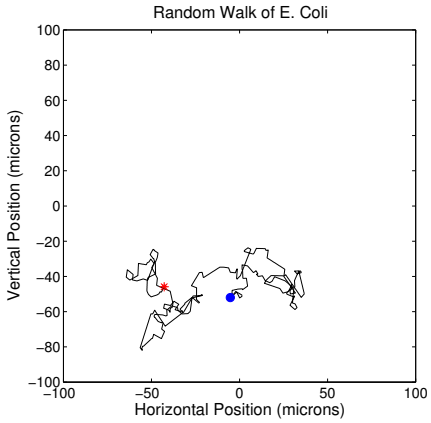

Fig. 2. Random walk trajectory of single swimming bacterium generated from our simulation. The cell moves from the blue marker to the red marker.

Additionally, the orientation of the cell is formulated by Equation (2), where θ_r is the cell's orientation during a run and θ_t is the cell's orientation during a tumble.

$$\begin{bmatrix} \theta_r(t) \\ \theta_t(t) \end{bmatrix} = \begin{bmatrix} \theta_r(t-1) \\ \theta_r \pm \mathcal{N}(62, 26) \end{bmatrix} \tag{2}$$

When a tumbling event is detected, we determine change in orientation based on previous experimental observations of tumbling *E. coli* in a plane [12, 13]. The change in cell orientation during a tumble event is chosen from a normal distribution with a mean of 62° and a standard deviation of 26°, with an equal probability that the cell will rotate in the positive or negative direction [12, 13, 7]. A representative trajectory exhibiting the run and tumble dynamics of a single cell is presented in Figure 2.

IV. Results

A. Unidirectional Motion

The experimental platform developed in this work resulted in the creation of microstructures propelled along geometrically biased trajectories via bacterial suspensions. Micro-gears were observed to rotate in a direction dependent on the chirality of the gear's teeth (Figure 3). Micro-gears possessing teeth oriented in the counterclockwise direction, rotated clockwise. Conversely, micro-gears possessing teeth oriented in the clockwise direction, rotated counterclockwise. Similarly, micro-chevrons were observed to translate unidirectionally along the structure's symmetric axis (Figure 4). Overall trajectories included both rotation and translation due to the stochastic nature of collisions. Since the rate of collisions over time in a semi-dilute suspension is not constant, experimental trajectories exhibited some curvature.

V. Discussion

This work explores MBRs propelled by suspensions of motile bacteria. By designing asymmetrically shaped micro-objects and exploiting a propulsion mechanism dependent on

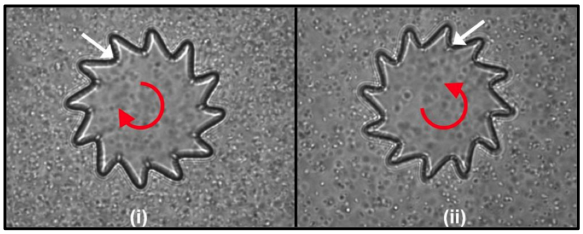

Fig. 3. Collisions between swimming bacteria in solution and micro-gear boundaries result in unidirectional rotation of the microstructure (i) Clockwise rotation direction observed for micro-gears exhibiting counterclockwise oriented teeth (ii) Counterclockwise rotation direction observed for micro-gears exhibiting clockwise oriented teeth

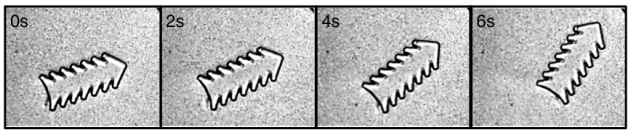

Fig. 4. Collisions between free swimming bacteria in solution and micro-chevron boundaries result in unidirectional translation along structure's symmetric axis.

collisions, resultant microstructure trajectories may be geometrically biased. We have implemented strategies to model free-swimming bacteria dynamics, including an-depth characterization of run and tumble behavior. Currently, we are developing methods to model cell-wall collisions and hydrodynamic interactions governing the behavior of swimming cells near surfaces. Through formulating a comprehensive mathematical model that describes MBR motion originating at the cellular level, we will be able to predict trajectories and to develop feedback control laws in order to drive MBRs. This will allow for applications such as exploring miniature environments and delivering micro-cargoes.

Acknowledgments

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