TOWARD FASTER BACTERIAL MICRO-ACTUATORS

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ABSTRACT

In recent studies, magnetotactic bacteria (MTB) have been used as autonomous mechanical actuators and biocarriers [1]. In many aspects, bacterial actuators surpass Micro Electro Mechanical Systems (MEMS) which make them the ideal candidates for motion in microfluidic systems. Although great progress has been made in combining biological systems and engineering techniques, increasing the speed of these bacteria is a prerequisite in order to improve their motion under low Reynolds numbers hydrodynamics [2]. Our work in this paper shows a noticeable increase in the mean speed of MTB when cultured in enriched ferrous iron media. Raising the concentration of iron from 33 µMol to 50 µMol, enhanced the average velocity of MTB by more than 50µm/s.

KEY WORDS: Magnetotactic bacteria, Velocity, Magnetosome, Ferrous iron

INTRODUCTION

When it comes to designing micro-scaled propulsion systems, nature offers a wide range of molecular motors that best fit all possible environments. Even the most advanced engineered microdevices don't reach the level of efficiency given by biological systems. For example, the behavior of certain bacteria in viscous fluids promises a great deal of advancements in nanotechnological applications [1]. As well as providing adapted propulsion, these bacteria fulfill energy requirements which are major concerns in designing artificial micro-actuators. Nonetheless, even bacterial actuators present their own set of challenges. Slow motion, poor thrust and controllability are the most common issues encountered in low Reynolds hydrodynamics [2]. To solve the controllability problem, we introduced magnetotactic bacteria [1][2][4]. MTB are motile aquatic prokaryotes which contain biogenic magnetite (Fe_3SO_4) encapsulated in a lipid bilayer and arranged to form a chain of magnetosomes along the polar axis (cf. Figure 1). This chain of magnetosomes acts as a single magnetic dipole (or compass) and allows the bacteria to align themselves with the earth magnetic field [3]. When an external magnetic field is applied, bacteria are attracted to the source by mean of magnetotaxis [1].

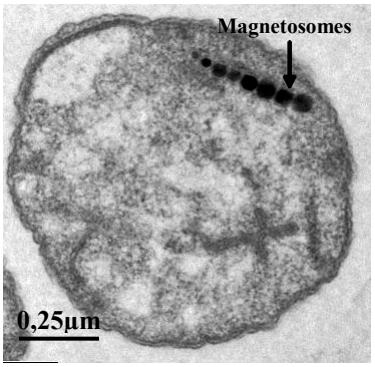

Figure 1: Transmission Electron micrograph of a thin-sectioned cell of MC-1 showing a chain of magnetosomes.

In this study, we used *Magnetococcus sp.* MC-1 strain, a magnetic gram negative coccus that has a length of ~1–2 µm and an average swimming speed of ~200µm/s in normal conditions [4]. This bacterium contains magnetosomes averaging 72 by 70 nm in size [3]. Controlling the size, magnetism and composition of the ferromagnetic nanoparticles embedded in the MTB will have a tremendous impact on biomedical technologies and industry. By changing magnetosomes properties, values of magnetism, coercivity and magnetic moment, could raise. Moreover, it is essential to improve our knowledge of all the parameters that influences bacterial motion in order to get accurate predictive models and implement propulsion systems.

To broaden the use of these bacteria to the desired applications, we need to increase the average velocity of bacteria so it compensates the loss of speed due to higher viscosity and temperatures [2][4]. Various conditions could be tested such as culture media, temperature, O_2 concentrations, etc [4]. In this experiment, MTB were cultured in different concentrations of iron (II) sulfate. We emitted the hypothesis that increasing the iron concentration will affect the magnetosomes size and hence, amplify the swimming speed.

METHODS

Culture growth conditions

Culture of MC-1 strain was grown in liquid medium under microaerobic conditions. Two growth conditions were tested: 33µMol and 50µMol of ferrous sulfate heptahydrate $FeSO_4 \cdot 7H_2O$ (F8048, Sigma-Aldrich) added to the culture medium prior to inoculation. MC-1 culture was incubated for 48h without agitation at room temperature.

Velocity acquisition experimental setup

Approximately 500 cells were washed by suspending the cells in 100 µl PBS-1X buffer at room temperature. The cells were deposited on a microscope slide, a cover slip placed over the drop using 450µm separators to form a pool in the center. The slide was then placed on a microscope stage (AxioImager Z1, Carl Zeiss Imaging solutions) under dark-field illumination. An electromagnet was positioned close to the bottom of the slide, under the field of view of the microscope. The electromagnet consists of a tip mounted on a computer controlled motorized x-y stage (PI Physik instrumente). The

electrical current was set to deliver an electromagnetic field of 20 Gauss. Image acquisition was made by AxioCamMr (Zeiss) using a 20X LD Ediplan lens magnifier. The exposure time was set to 200ms allowing the visualization of the trail left by bacterial motion. All measurements were made with AxioVision v.4.6.0 software.

RESULTS AND DISCUSSION

MC-1 is most likely one of the fastest aquatic bacteria with displacement speeds reaching up to 300µm/s in controlled laboratory conditions [1]. Previous work done by our laboratory showed that, at room temperature and in normal conditions, MC-1 has an average velocity of ~180-200µm/s [4] (cf. *Figure 2*). The swimming speed was calculated after cultivating bacteria in liquid medium containing 33µMol of ferrous sulfate. In this experiment, we increased iron (II) sulfate concentration by 1,67-fold (compared to the initial condition). The other parameters remained unchanged. Preliminary results showed a shifted Gaussian distribution to higher velocities as depicted in *Figure 3*. The swimming speed peeked at more than 360µm/s and the average displacement speed was ~240-280µm/s. Compared to the initial condition, increasing ferrous sulfate concentration enhanced the average speed of MC-1 (more than 50 µm/s). Knowing the central role played by this mineral in biomineralization of magnetite [5], it is possible that increasing the concentration of ferrous sulfate had an effect on the size of magnetosomes. Although, the link between magnetosomes size and bacteria swimming speed remains unclear, higher concentrations of iron (II) sulfate had clearly an effect on MC-1 displacement speed. Further work should be done to investigate the phenomenon. TEM micrographs of magnetosomes obtained with this new condition should complete the work and show whether higher concentration of ferrous sulfate increased the size of magnetosomes.

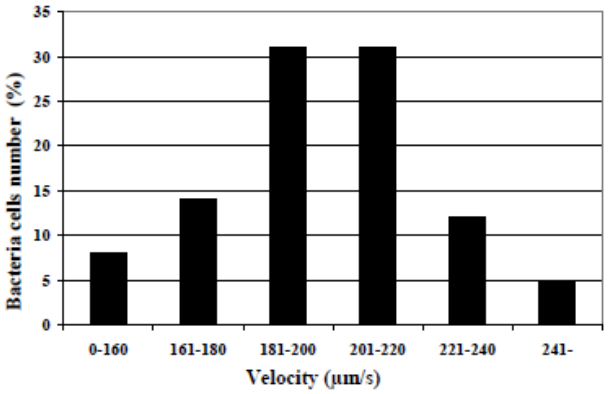

Figure 2: Swimming speed Distribution of MC-1 when cultivated in 33µMol of ferrous sulfate ($FeSO_4$). Cells were suspended in PBS buffer [4].

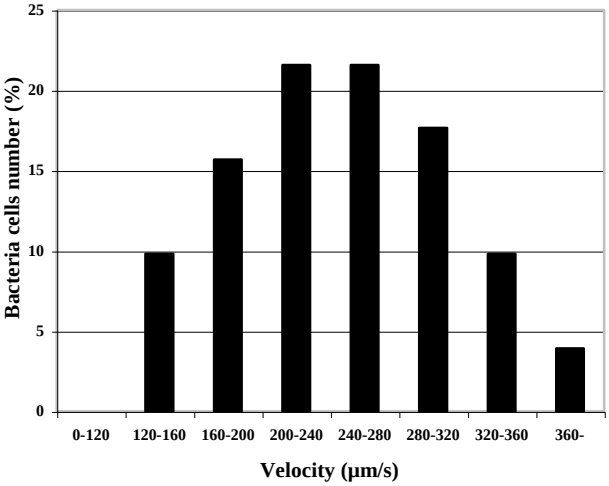

Figure 3: Swimming speed Distribution of MC-1 when cultivated in 55µMol of ferrous sulfate ($FeSO_4$). Cells were suspended in PBS buffer.

CONCLUSION

MC-1 bacteria cultivated in liquid medium containing higher concentrations of ferrous sulfate were faster than the previous culture. Raising the speed limit will improve bacterial motion in microfluidic systems as well as open the door to other biomedical applications.

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