Lab on a Chip

Electronic Supplementary Information (ESI)

Bacterial Chemotaxis Enabled Autonomous Sorting of Nanoparticles of Comparable Sizes

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S. I. Selective attachment of bacteria to particles through specific and non-specific interactions

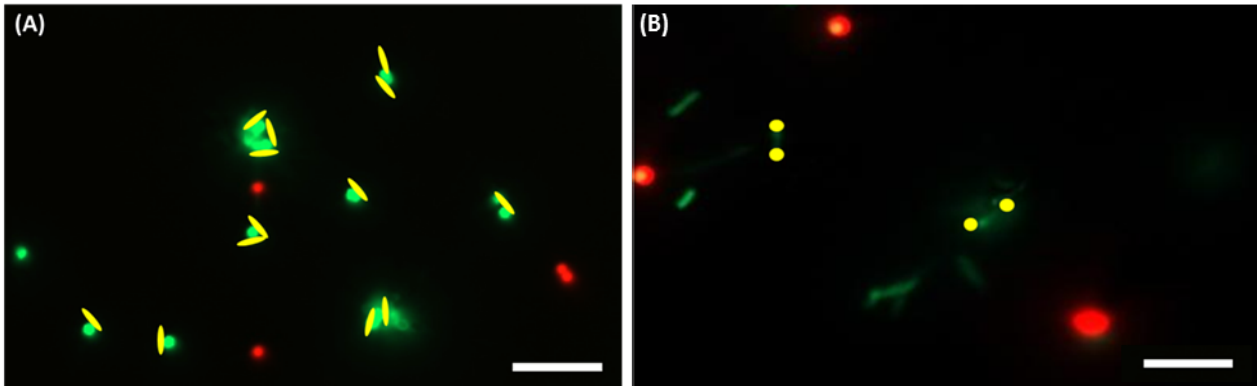

Fig. S1 (A) Representative micrograph images of 1040 nm streptavidin-coated particles (green), 1000 nm uncoated polystyrene particles (red), and biotinylated antibody coated *E. coli* MG1655m (false colored in yellow). *E. coli* selectively attached on the surface of the 1040 nm particles via streptavidin-biotin binding. (B) Representative micrograph images of 1000 nm positively charged particles (yellow), 1000 nm neutrally charged polystyrene particles (red), and biotinylated antibody coated *E. coli* MG1655m (green). Negatively charged bacteria selectively attached on positively charged particle via electrostatic interactions, while neutrally charged particles remain unattached. All scale bars are 10 µm.

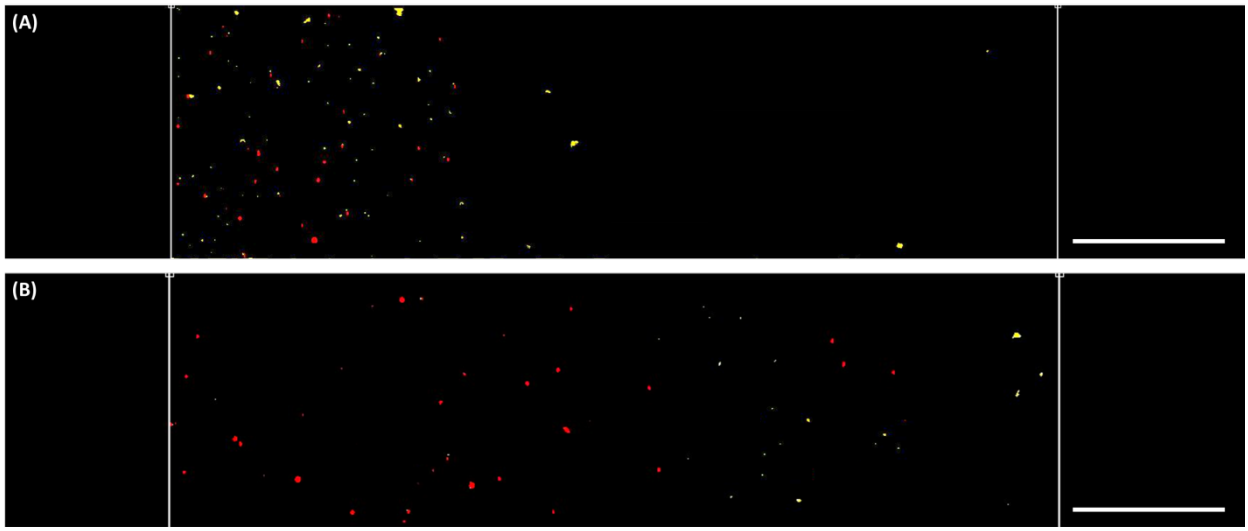

Fig. S2 Representative images of the work area in the microfluidic sorting device. **(A)** At t = 0 minutes, the 1000 nm particle-bacteria assemblies (yellow) and the freely diffusing neutrally charged 1000 nm particles (red) reside in the left-half of the device center channel. (B) At t = 35 minutes, bacteria carrying 1000 nm particles have migrated up the chemical gradient and reside in the right-half of the device center channel. All scale bars are 100 µm.

S.II. Design of the microfluidic sorting platform

The optimum width of the work area for the sorting platform was determined through computational modeling of the 3D bacterial random walk. The maximum possible range of travel of *E. coli* bacteria in presence of a chemoattractant source was modeled over time. To this end, the motility of bacteria was modeled as two distinct states of run and tumble. In the run state, the flagellar motors of *E. coli* synchronously rotate in the counterclockwise direction. This causes the flagellar filaments to coalesce, thus forming a bundle and generation of a propulsive force. Through this process, the bacterium propels itself forward at a constant speed (~30 μm/s). In an isotropic environment, this *E. coli* run lasts on average for about 0.9 s followed by a tumble. Bacterial tumbling occurs when one or more of the bacterium's flagellar motors rotate in the clockwise direction. This process causes the disruption of the flagellar bundle. During a tumble, under thermal diffusion effects, the bacterium randomly reorients itself before beginning a new run cycle. The duration of a tumble lasts for about 0.1 s on average. The successive occurrence of runs and tumbles leads to stochastic motion (i.e. random walk) of bacteria in three dimensions. Random walks can be modeled as a two-state Markov chain with state duration distributions occurring based on an exponential distribution:

$$f(t,\lambda_i) = \lambda_i e^{-\lambda_i t}$$

In the presence of a chemical attractant gradient, the bacterium biases its random walk by extending when moving up the chemoattractant gradient. The extended run period lasts for an average of 1.3 s. Through this biased random walk mechanism, bacteria are able to direct their movement towards chemical attractant source. Our simulation results show that in presence of gradient generated by diffusion of 0.4% (w/v) casamino acids from a point source, *E. coli* travels 2025 μm ± 790 μm over a 45 minutes period, as depicted in Fig. S3. Thus, to achieve an effective chemotaxis-based sorting, we set a limit of 1500 μm for the lateral dimension of the bacteria chemotaxis-enabled microfluidic sorting device.

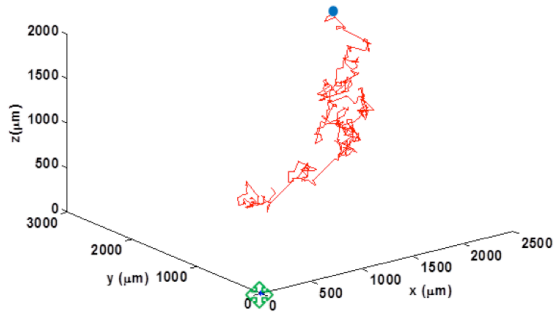

Fig. S3 Representative bacterial trajectory in presence of a chemo-attractant gradient with 0.4% (w/v) casamino acids as a chemo-attractant point source. The blue solid circle on the bacterial path represents the starting point and the green diamond at coordinates (0,0,0) represents the location of the chemical attractant source.

S. III. Throughput of the bacteria chemotaxis-enabled microfluidic sorting platform

The maximum particulate concentration permissible for effective chemotaxis-based sorting was empirically determined to be 1.8×10^{10}/ml. Considering the work area volume ($500\times4000\times300\ \mu m^3$) and the required sorting time of up to 45 min, the throughput for the current design was estimated to be:

$$\text{Throughput} = \text{work area volume} \times \text{concentration of processed mixture} \times \text{process time}$$

$$= 6.0\times10^{-4}\text{ml}\times1.8\times10^{10}\left(\frac{\text{particles}}{\text{ml}}\right)\times\frac{1}{45\text{ minutes}} = 2.4\times10^{5}\ \frac{\text{particles}}{\text{min}}$$

The throughput of our bacteria chemotaxis-enabled sorting method is within the range of other microfluidic based sorting techniques (10^3-10^9 cells/min)[1]. This throughput can further be improved by increasing the length of the work area or by multiplexing.

References

1 D. R. Gossett, W. M. Weaver, A. J. Mach, S. C. Hur, H. T. K. Tse, W. Lee, H. Amini and D. Di Carlo, *Anal. Bioanal. Chem.*, 2010, **397**, 3249–67.

