Supplementary lnformation

Percolation of noneqilibrium assemblies of colloidal particles in active chiral liquids

Pragya Kushwaha[1], Sayan Maity[1], Anjaly S. Menon[2],
Raghunath Chelakkot[3], Vijayakumar Chikkadi
Indian Institute of Science Education and Research Pune,
Pune 411008, India
[2] *Department of Applied Physics,*
Aalto University School of Science,
Puumiehenkuja 2, 02150 Espoo, Finland
[3] *Indian Institute of Technology Bombay,*
Powai, Mumbai 400076, India

CONTENTS

I. TRAJECTORIES OF BACTERIA

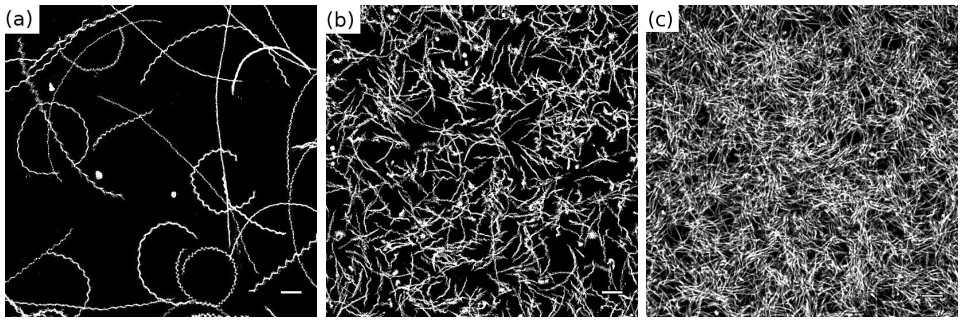

FIG. S1. Trajectories of bacteria at three different concentrations of bacteria corresponding to $c_b = 0.01b_0$, $5b_0$ and $10b_0$ from left to right, respectively.

The Figs. S1(a)-(c) depicts the trajectories of bacteria at three different concentrations c_b = $0.01b_0$, $1b_0$, and $10b_0$, respectively. The concentration used in all the our experiments is $10b_0$, and the definition of b_0 is given in the main text. The form of the trajectories is circular at low densities, however, this features diminishes with increasing concentrations due to interactions between swimmers. Despite the non-circular nature of trajectories at high densities, the colloid-bacteria interactions gives rise to unbalanced tangential forces on the colloids, leading to persistent rotation of particles [1]. This is evident in the supplementary movies SV1 and SV2.

II. RELAXATION TIME-SCALE OF THE LARGEST CLUSTER

To quantify the time scale of long lived clusters, we estimate the relaxation time of the largest cluster. This is done by calculating the time needed for the largest cluster to lose 50% of the original particles. Denoting the number of particles in the largest cluster at time $t = 0$ as $N_l(0)$ and the number of original particles that persist in the largest cluster after a time interval t as $N_l(t)$. The variation of the ratio $N_l(t)/N_l(0)$ with time is illustrated above in Fig.S2(a) at several densities of colloidal particles. The dashed line is drawn at $N_l(t)/N_l(0) = 0.5$. The crossing of this line by the curves is considered relaxation time of the largest cluster. The Fig.S2(b) shows the relaxation timescale at varying densities of colloids ϕ. The dotted line shows an exponential fit to the data.

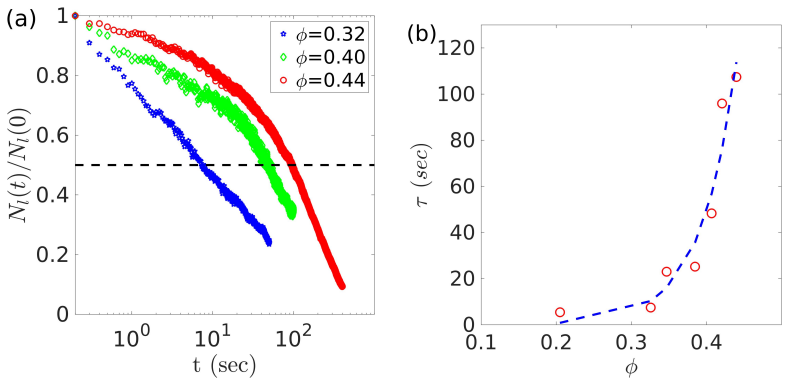

FIG. S2. The relaxation time of the largest cluster. (a) The number of original particles in the largest cluster after a time interval t. $N_l(0)$ is number of particles in the largest cluster at $t = 0$ and $N_l(t)$ is the number of original particles that persist in the largest cluster after a time interval t. (b) The relaxation time-scale of the largest cluster is shown at several densities of colloidal particles. The dashed curve is an exponential fit.

III. ACTIVITY OF BACTERIA INFERRED FROM THE DYNAMICS OF COLLOIDS

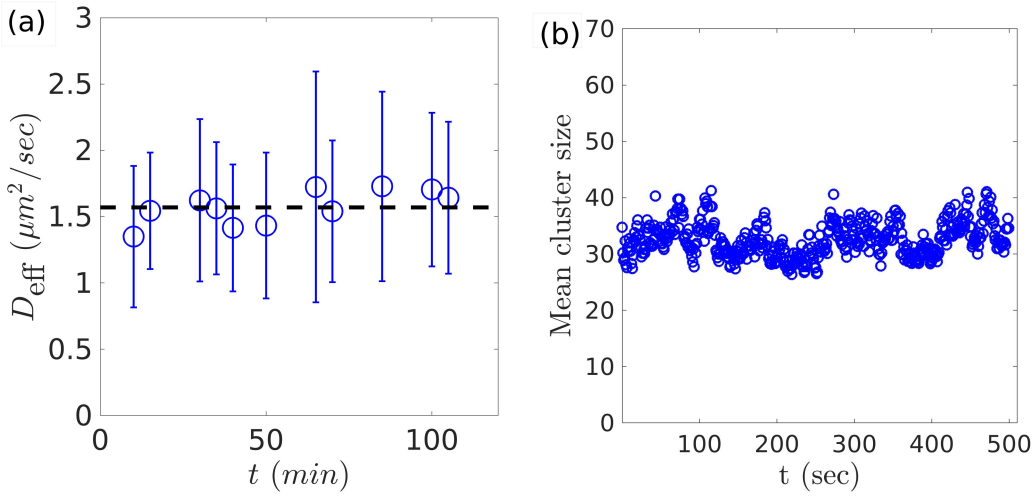

FIG. S3. (a) The effective diffusion constant D_{eff} of colloidal particles in active liquids at a bacteria concentration $c_b = 10b_0$. The area fraction of colloids is $\phi = 0.005$. The dashed line indicates the mean value. The error bars are obtained from the standard deviations of D_{eff}. (b) The mean cluster size of colloidal aggregates over a duration of 500 s at $\phi \sim 0.4$ and $c_b = 10b_0$. This data was measured after 90 minutes from the start of the experiment.

We analyze the constancy of bacterial motility over the duration of our experiments by investigating the dynamics of colloids. The experiments in the manuscript are performed at a bacteria concentration of $c_b = 10b_0$, where $b_0 = 6 \times 10^9$ cells/ml. At such high densities, it is difficult to measure directly the velocities of individual swimmers. Therefore, we measured the effective diffusivity of colloidal particles at an area fraction of $\phi = 0.005$. The effective long time diffusion constant is defined as $D_{\text{eff}} = \frac{1}{4}\delta\langle\Delta r^2(\tau)\rangle/\delta t$ [2]. The time scale of our observation δt is chosen such that it is sufficient to observe diffusive motion. This data is shown in Fig. S3(a) over a duration of 100 min. The variation in the diffusivity D_{eff} is small. The Fig. S3(b) shows the mean cluster size of colloidal aggregates over a time duration of 500 s at $\phi \sim 0.4$ and $c_b = 10b_0$. This data was measured after 90 minutes from the start of the experiment. The mean cluster size fluctuates about a mean during our measurement timescale. Even though we have not measured the velocity of bacteria directly, the small variation in D_{eff} and the fluctuations of mean cluster size about a mean provide strong evidences of the constant activity of bacteria over the duration of our experiments.

IV. VARIATION OF MOTILITY OF BACTERIA DUE TO INTERACTIONS WITH COLLOIDAL BEADS

All the measurements reported in the manuscript were done with an objective of 10x magnification and NA = 0.30 in bright-field mode. The resolution is not sufficient to extract the bacteria tracks in the current data. A detailed analysis of variation in the motility of bacteria due to varying density of colloidal particles is deferred for future investigations.

Here we present results of preliminary investigation of the influence of colloidal particles on bacteria motion. These studies are motivated by earlier studies of a similar systems [3]. We have calculated the velocity distribution of bacteria for a fixed configuration of colloidal cluster shown in Fig. S4(a). The concentration of the bacteria is $c_b = 1b_0$, which is small compared to the concentration, $c_b = 10b_0$, used in the manuscript. At dilute concentrations, we track the swimmer trajectories and measure their velocities. This is shown in Fig. S4(b). The histogram in orange shows the velocity distribution $P(v)$ of bacteria with colloidal particles, which is compared with $P(v)$ without colloidal particles. Evidently, there is an enhancement in the velocities of bacteria in the presence of colloids. These are similar to

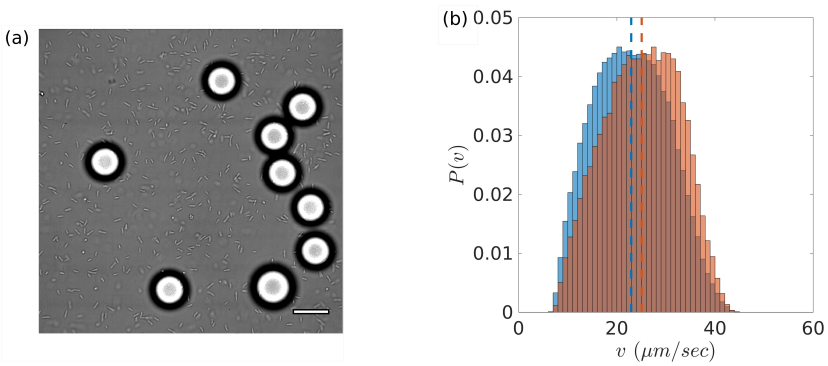

FIG. S4. Variation in the motility of bacteria due to the presence of colloidal particles. (a) Bright-field image of bacteria and stuck colloidal particles taken at a magnification of 63X. The size of the colloidal particles is $\sigma = 15\mu m$, the concentration of the bacteria is $c_b \sim 1b_0$, and the scale bar is of $15\mu m$. (b) The normalised histogram of velocity distribution $P(v)$ is shown for the bacteria suspension with colloidal particles in orange and without colloidal particles in blue. The average velocity of bacteria without colloidal particles is $v = 22.926 \pm 7.476$, and in the presence of beads $v = 25.059 \pm 7.56$. The vertical lines in the figure denote these mean values of the velocities.

the results reported in earlier studies [3].

V. SUPPLEMENTARY VIDEOS INFORMATION

SV1: Spinning motion of half coated polystyrene particle of 15 μm in the bacterial bath. The video is accelerated 10X times and the scale bar is $30\mu m$.

SV2: Trajectories of colloids in a cluster are shown in different colors. The video is accelerated 5X times, scale bar: 20um.

SV3: PS $15\mu m$ particles in chiral bacterial bath. The video is played 5X times faster, scale bar is $50\mu m$.

[1] D. Grober, I. Palaia, M. C. Uçar, E. Hannezo, A. Šarić, and J. Palacci, Unconventional colloidal aggregation in chiral bacterial baths, Nature Physics , 1 (2023).

[2] X.-L. Wu and A. Libchaber, Particle diffusion in a quasi-two-dimensional bacterial bath, Physical review letters **84**, 3017 (2000).

[3] S. Kamdar, S. Shin, P. Leishangthem, L. F. Francis, X. Xu, and X. Cheng, The colloidal nature of complex fluids enhances bacterial motility, Nature **603**, 819 (2022).