Supporting Material for

Differential Dynamic Microscopy: a High-Throughput Method for Characterizing the Motility of Microorganisms

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Supporting Figures



Figure S1: SW *E.coli*. Effect of using different forms of the speed distribution: Schulz (black circles), Log-normal (red squares) $P_L(v) = \exp\left[-\left(\ln(v/v_g)\right)^2/2\sigma_g^2\right]/v\sigma_g\sqrt{2\pi}$, or Gaussian (green triangles) $P_G(v) = \exp\left[-(v-\bar{v})^2/2\sigma^2\right]/\sigma\sqrt{2\pi}$, on the fitting parameters versus q using Eqs. 2, 4 & 6. See main text. Note that in the case of a Log-normal distribution, the arithmetic mean and standard deviation are $\bar{v} = v_g \exp(\sigma_g^2/2)$ and $\sigma = \bar{v}\sqrt{\exp(\sigma_g^2) - 1}$ respectively, where v_g and $\exp(\sigma_g)$ are the geometric mean and geometric standard deviation.



Figure S2: Comparison of SW *E.coli* experiments (Fig. 2-4) and straight swimmers simulations. Simulated data were generated using a depth of field δ =40µm, a Schulz distribution for *P*(*v*), and motility parameters obtained from experiments (Figure 4). (a) ISFs versus delay time at several *q* values in µm⁻¹. Symbols and lines are results for experiments and simulations respectively. (b) Results of the speed distribution *P*(*v*) obtained from fitting the *g*(*q*, τ) functions assuming a Schulz (black), Log-normal (red) or Gaussian (green) distributions for experiments (lines) and simulations (dotted lines). The *P*(*v*) distributions were calculated from the mean speed $\bar{\nu}$ and width σ averaged over the range 0.6 < *q* < 1.7 µm⁻¹ to avoid noisy values at higher *q* in the case of the Gaussian distribution (see Fig. S1).



Figure S3: *C. reinhardtii.* The reconstructed ISFs, $f(q, \tau)$, plotted against (a) τ and (b) $q\tau$. The q value increases, by step of $\approx 0.004 \ \mu m^{-1}$, from red to blue end of the spectrum colour in the range $0.18 < q < 0.89 \ \mu m^{-1}$. (a) The characteristic time of the fast process is independent of q, while its amplitude decreases with q. (b) The slow process scales with $q\tau$ confirming the ballistic motion of this process.

Details for creation of smooth swimmer Δ *che***Y mutant strain**:

P1 phage transduction (1) was used to create a smooth swimming (AB1157 $\Delta cheY$) using the appropriate *E. coli* K-12 single knockout mutant from the KEIO collection (2). Kanamycin (final concentration 30 mg/l) was added to all growth media for AB1157 $\Delta cheY$.

Supporting References:

1. Miller, J.H. 1972. *Experiments in Molecular Genetics*. Cold Spring Harbor Laboratory Press.

2. Baba, T., T. Ara, M. Hasegawa, Y. Takai, Y. Okumura, M. Baba, K.A. Datsenko, M. Tomita, B.L. Wanner and H. Mori. 2006. *Construction of Escherichia coli K-12 in-frame, single-gene knockout mutants: the Keio collection*. Syst. Biol., 2, 1–11.