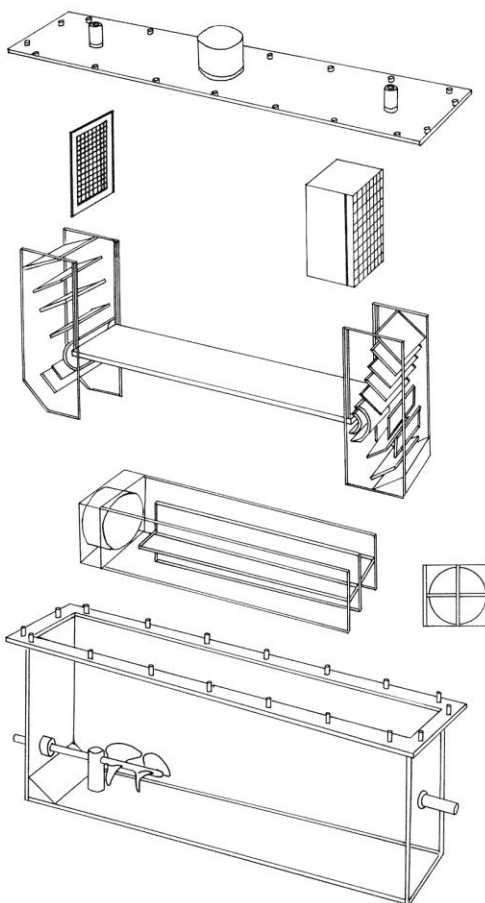


Fig. S1 Steffensen-type swimming respirometer for intermittent-flow respirometry. Panel A shows the different components of the respirometer, including the baffles, flow straightener and honeycomb used to produce near-laminar flow. Panel B shows the assembled respirometer with an external motor powering the propeller. Arrows indicate flow direction.

A)



B)

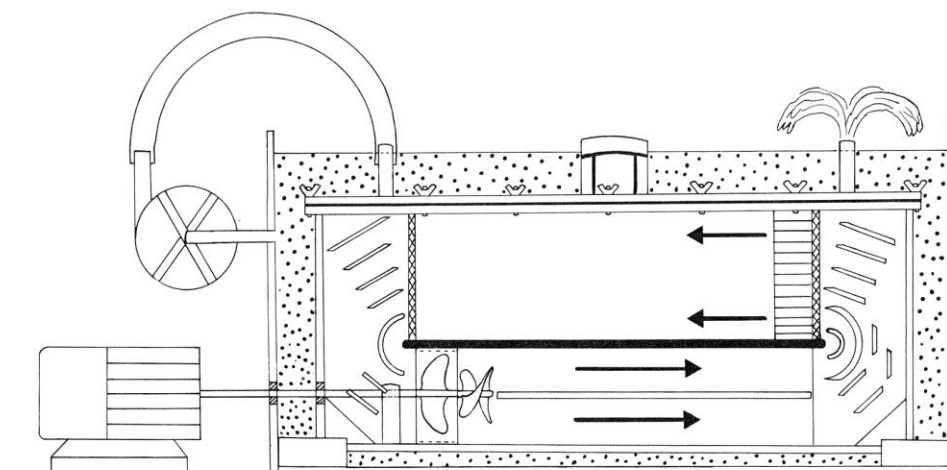
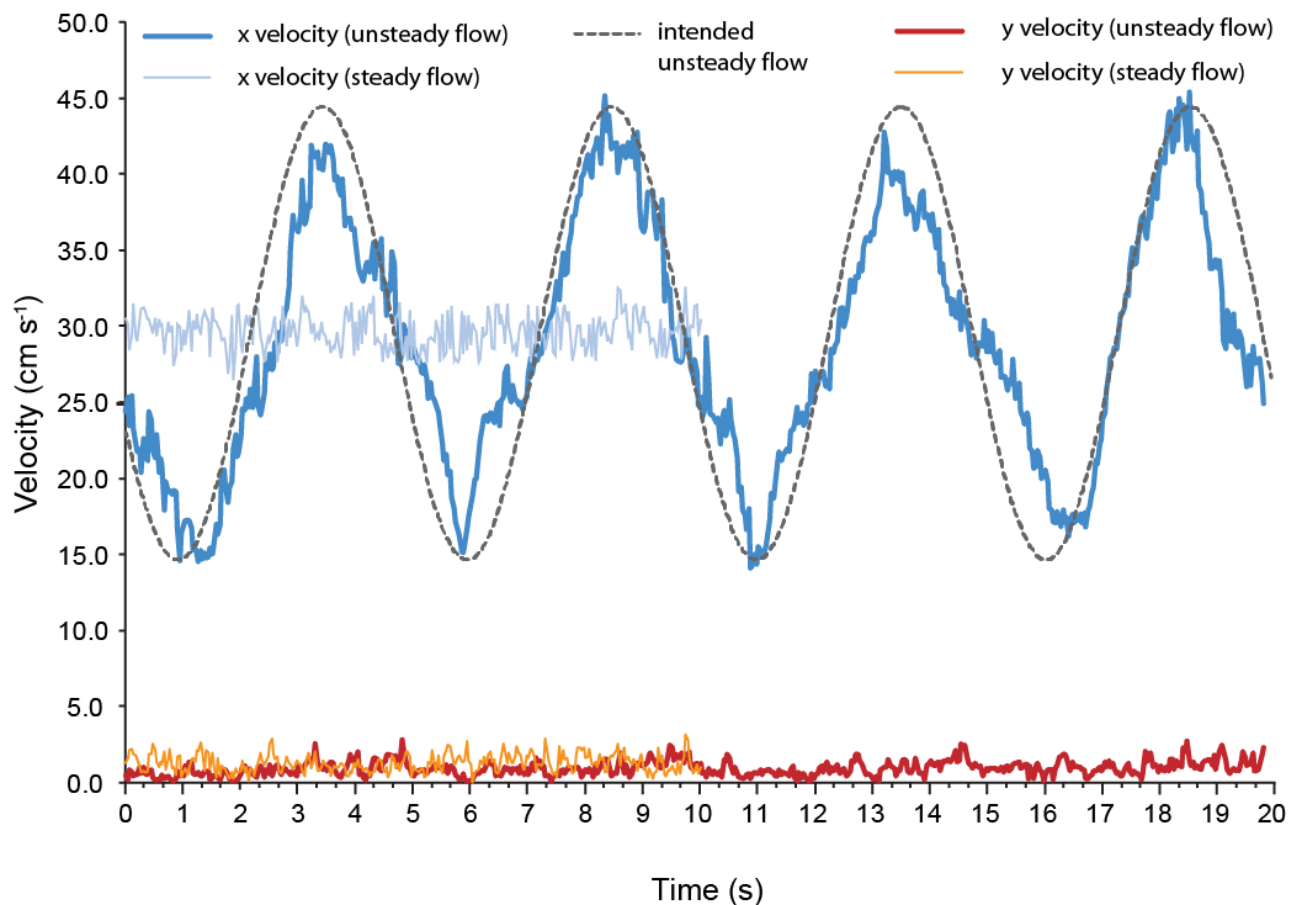


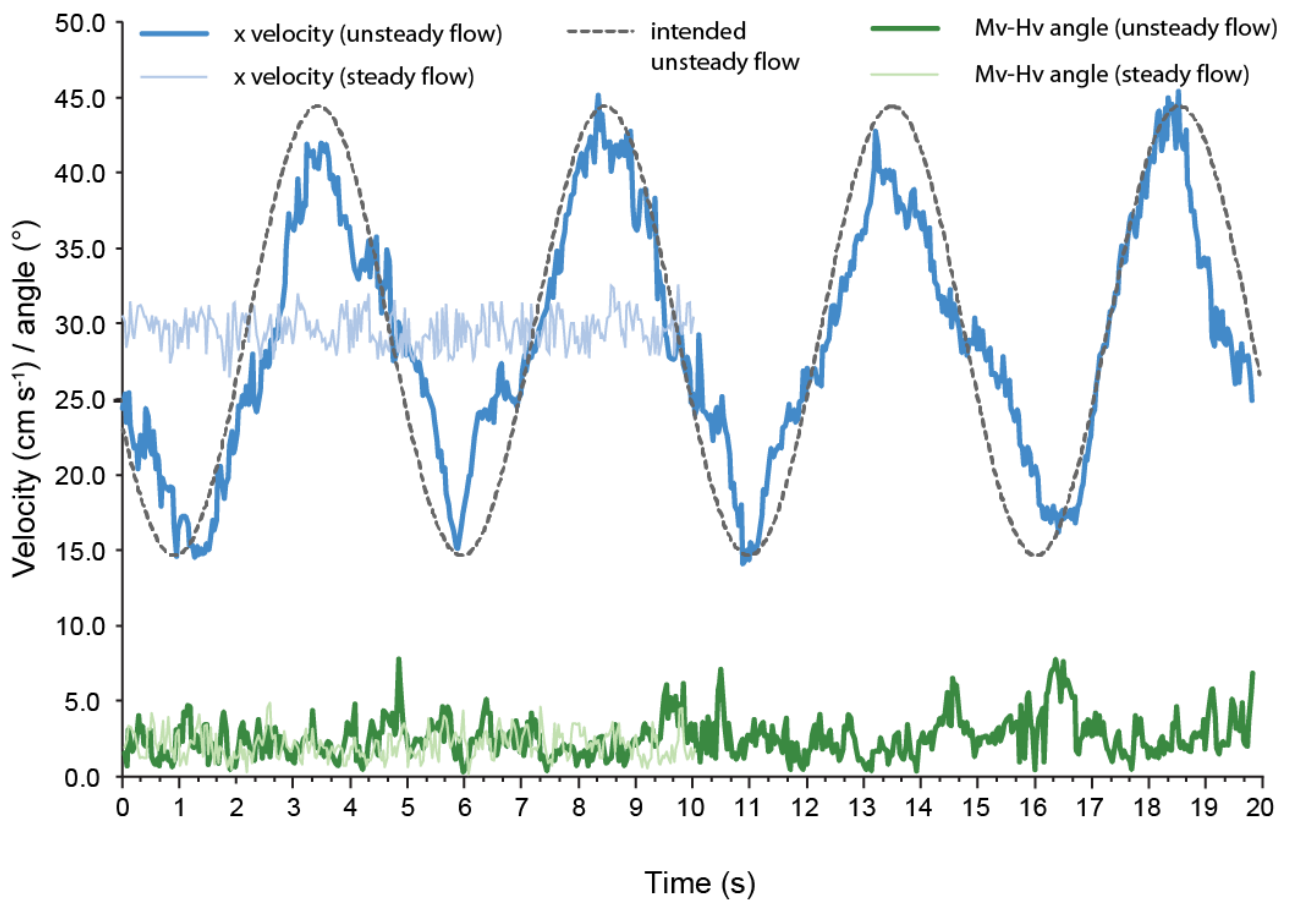
Fig. S2 Example of flow characteristics in steady flow versus high unsteady flow for an average size fish (14.8 cm total length). Water flow velocity (cm s^{-1}) was obtained by tracking passive particles (neutrally buoyant expandable polystyrene beads with diameter < 1 mm, Foamex Polystyrene, Revesby, Australia) in the test section of the swim chamber at 0.033 Hz. Particles were tracked using the manual object tracking plugin MtrackJ for ImageJ (Meijering et al., 2012). Particles were tracked for 20 s in high unsteady flow (mean speed = 29.6 cm s^{-1} or 2 BLs^{-1} ; amplitude = 1 BLs^{-1} ; period = 5 s): the thick blue line indicates flow velocity in the x axis and the thick red line indicates absolute flow velocity in the y axis. The dashed grey line indicates the intended flow velocity. For comparative purposes, particles were tracked for 10 s in steady flow (mean speed = 29.6 cm s^{-1} or 2 BLs^{-1}): the thin blue line indicates flow velocity in the x axis and the thin orange line indicates absolute flow velocity in the y axis. Measures were obtained by averaging values obtained three times on the same video for both steady and unsteady flow; we averaged absolute values of velocity in y. Noise is partly due to small scale variation in flow speed and partly due to magnification of errors from the digitizing process (Walker, 1998). Descriptive statistics are presented in Table S1. Data are deposited in the figshare repository (DOI: 10.6084/m9.figshare.789064).

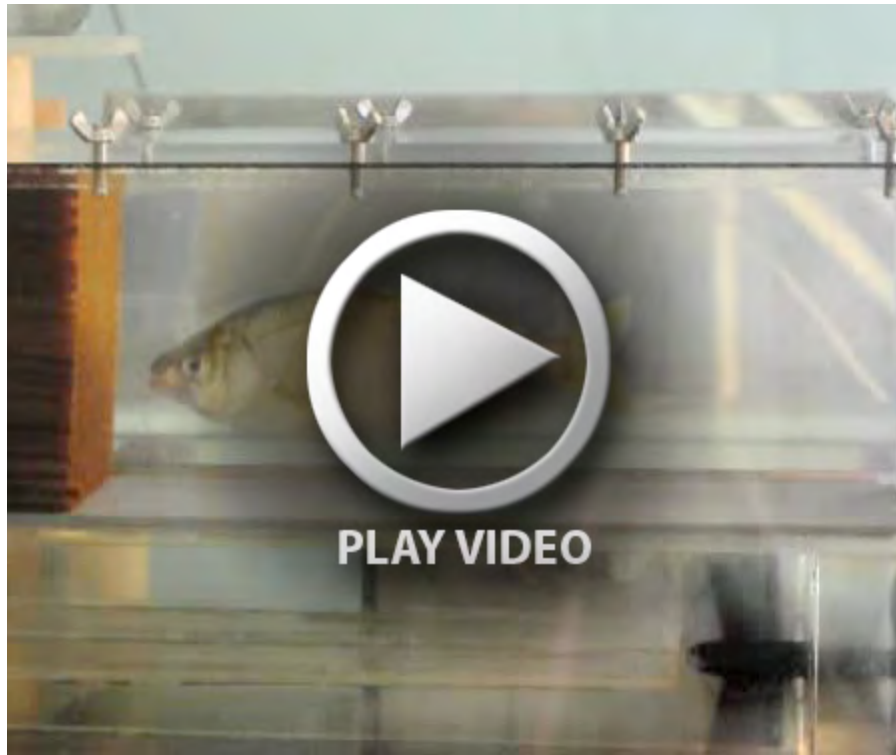


Meijering, E., Dzyubachyk, O. and Smal, I. (2012). Methods for cell and particle tracking. *Methods Enzymol.* **504**, 183-200.

Walker, J. A. (1998). Estimating velocities and accelerations of animal locomotion: A simulation experiment comparing numerical differentiation algorithms. *J. Exp. Biol.* **201**, 981-995.

Fig. S3 Same as Fig. S2, but showing (in green) the change in absolute vector angle of tracked passive particles at 0.033 Hz relative to the expected flow direction (i.e. measured vector vs. horizontal vector angle; Mv-Hv angle). Particles were tracked using the manual object tracking plugin MtrackJ for ImageJ (Meijering et al., 2012). Measures were obtained by averaging absolute values obtained three times on the same video for both steady and unsteady flow. The thick green line indicates the absolute Mv-Hv angle for particles tracked over 20 s in high unsteady flow (mean speed = 29.6 cm s^{-1} or 2 BLs^{-1} ; amplitude = 1 BL s^{-1} ; period = 5 s). The thin green line indicates the absolute Mv-Hv angle for particles tracked over 10 s in steady flow (mean speed = 29.6 cm s^{-1} or 2 BLs^{-1}). The mean change in absolute Mv-Hv angle was 1.98° (range $0.00^\circ - 4.76^\circ$) for steady flow and 2.47° (range $0.18^\circ - 7.67^\circ$) for high unsteady flow. Other than velocity fluctuations in the x axis, flow conditions were similar in the two treatments and approximated near-laminar flow. Descriptive statistics are presented in Table S1. Data are deposited in the figshare repository (DOI: 10.6084/m9.figshare.789064).





Movie 1. *Cymatogaster aggregata* swimming in high-amplitude unsteady flow. Fish exhibit high variation in fin beat period (T): T increases as flow velocity decreases, and decreases as flow velocity increases.

Table S1 Intended vs. observed flow characteristics (mean \pm s.d.) for steady (mean speed = 29.6 cm s^{-1} or 2 BLs^{-1}) and high unsteady (mean speed = 29.6 cm s^{-1} or 2 BLs^{-1} ; amplitude = 1 BLs^{-1} ; period = 5 s) flow visualized in Figs S1 and S2.

Descriptor	Steady flow		High unsteady flow	
	<i>Intended</i>	<i>Observed</i>	<i>Intended</i>	<i>Observed</i>
Velocity in x (cm s^{-1})	29.60 ± 0.00	29.63 ± 1.18	29.60 ± 0.00	28.98 ± 7.98
Amplitude (cm s^{-1})	N/A	N/A	14.80 ± 0.00	15.03 ± 2.01
Velocity in y (cm s^{-1})	0.00 ± 0.00	-0.24 ± 0.84	0.00 ± 0.00	0.10 ± 0.70
Absolute velocity in y (cm s^{-1})	0.00 ± 0.00	1.27 ± 0.59	0.00 ± 0.00	0.93 ± 0.49
Mv-Hv angle ($^{\circ}$)*	0.00 ± 0.00	-0.37 ± 1.37	0.00 ± 0.00	0.28 ± 2.04
Absolute Mv-Hv angle ($^{\circ}$)*	0.00 ± 0.00	1.98 ± 0.92	0.00 ± 0.00	2.47 ± 1.36
Period (s)	N/A	N/A	5.00 ± 0.00	4.98 ± 0.28

* Vector angle relative to intended flow direction.