S1: Hardware considerations

Operational amplifier selection

DStat relies on three op-amps: the control and buffer amplifiers in the potentiostat (U1 and U2 in Fig. 2(c) in the main text) and the transimpedance amplifier in the current measurement circuit (U3 in Fig. 3(b) in the main text). In DStat, these amplifiers are the LMP7702 (Texas Instruments, Dallas TX, USA) for control and buffer and LMP7721 (Texas Instruments) for transimpedance, respectively. These models were carefully selected on the basis of a number of criteria. To keep polarization error small, the buffer was chosen to have a minuscule input bias current (~200 fA; a function of its CMOS input stage), allowing it to maintain a negligible reference electrode current. To avoid problems with potential inaccuracies, control and buffer amplifiers with large open loop gains and small voltage noise densities and input voltage offsets were selected, as a closed loop gain error exists that is the reciprocal of the open loop gain (for an inverting amplifier with gain of -1), and potential accuracy is also affected by the amplifiers' voltage noise densities and input voltage offsets. Additionally, the control and buffer amplifiers' bandwidths should be sufficiently high to accommodate the potential roles of the potentiostat—a bandwidth of a few hundred kilohertz may be ample for typical experiments at macroelectrodes, but several megahertz may be required for fast scanning experiments or electrochemical impedance spectroscopy. In DStat, the LMP7702 fulfils the requirements for both amplifiers while reducing design complexity by containing two amplifiers in the same physical package. Further, the LMP7702 has an open loop gain of approximately 10^6, an input bias voltage of ± 32 µV, and an input referred voltage noise density of 9 nV$/\sqrt{\text{Hz}}$ (at 1 kHz), allowing potential accuracy to tens of microvolts, and with a unity gain bandwidth of 2.5 MHz, potential control can be maintained at high enough frequencies for DStat's intended uses.

In the transimpedance measurement circuit, the input bias current of the op-amp is summed with the current at the working electrode input and can cause a large offset error, especially at high gain. For example, a simple op-amp such as the Texas Instruments LM741 can have an input bias current as high as 500 nA, a non-trivial error for low-current voltammetry; if this amplifier were used, the measurement circuit would cause output saturation at gains over 3 MΩ (for a ±1.5 V output range) without an applied current, making measurements impossible. Further, input bias current can vary significantly between op-amps of the same model, which complicates calibration, and electronic compensation requires manual tuning, increases noise, and may not be able to fully remove input bias current. To overcome these challenges, we chose the internally-compensated LMP7721, which has an extremely small input bias current of 3 fA (~1 electron every 53 µs), effectively eliminating bias current error.

DAC sample rate and reconstruction filtering

As described in the main text, DACs are an integral part of most modern potentiostat control circuits. The MAX5443 (Maxim Integrated, San Jose CA, USA) DAC was selected for DStat to allow implementation of appropriate filters and to minimize aliasing. DACs are discrete time devices—only able to change value at finite time intervals. The rate at which the value can be updated is crucial for several reasons. Most significantly, the Nyquist-Shannon sampling theorem reveals that in order to reconstruct a continuous signal of a given frequency from discrete samples, samples must be output at at least double the frequency of the signal. Additionally, for correct reconstruction, the output must be band-limited to remove frequencies above the Nyquist frequency (half the sample rate) or else artifacts will be observed as mirrored and normal spectral duplicates of the output spectrum (known as images), at multiples of the sampling frequency, in the frequency domain. That is, if the output is not band-limited, images will be found at $nF_s \pm f$, where n is a positive integer, F_s is the sample frequency, and f is the frequency of a signal in the intended output spectrum. The reconstruction filter's function is mainly to remove these images and should ideally have the response of a rectangular function in the frequency domain, such that all frequencies below the Nyquist frequency are passed completely and all higher frequencies are completely blocked. Unfortunately, it is not possible to have a filter with an infinitely high stopband rolloff; therefore, the sample frequency must be sufficiently high to allow a gap between the highest frequency component of the output spectrum and the Nyquist frequency. In addition, most DACs operate with a zero-order hold, where a given output voltage is maintained until the next value is set, resulting in a staircase-like output that results in an attenuation of high frequencies corresponding to $|\frac{\sin(2\pi F_s)}{2\pi F_s}|$. This can be corrected by analogue or digital filtering, but the simplest way to ensure accuracy at high frequencies is to use a sample frequency much higher than the highest desired output, since the attenuation is relatively small at low frequencies. Using a high sample frequency also relaxes the requirements for the reconstruction filter since there is a large gap between the highest output frequency and the Nyquist frequency.

The effects of aliasing are shown in Fig. A(a) where the sine wave outputs from DStat and CheapStat at 70 Hz with an amplitude of 50 mV are compared. CheapStat has a relatively low sample frequency of approximately 2 kHz and doesn't employ a reconstruction filter, resulting in obvious jumps between samples. These images manifest as peaks at multiples of the sampling frequency in the frequency domain (Fig. A(b), left). This can be problematic for voltammetry since the sudden voltage changes will result in transient spikes in the non-faradaic current at the working electrode, distorting the measurement, depending on the cell capacitance, and desired measurement speed. These aliases are a natural part of a DAC's output and are also visible in DStat's (raw) output recorded without applying the reconstruction filter (Fig. A(b), right—green curve). To compensate for the images, DStat's DAC employs an active 4th order low pass filter with a cutoff frequency of 35 kHz, constructed around a MAX4477 (Maxim Integrated) dual op-amp. This leaves ample usable bandwidth with the DAC's maximum attainable sample rate of ~100 kHz while removing the majority of the images visible in the frequency domain (Fig. A(b), right—red curve) resulting in the smooth curve in Fig. A(a). The filter also prevents voltage glitches that can occur when switching a DAC's output from being applied to the cell.

Power supply constraints

DStat was designed to be a self-contained instrument that is capable of operation with only connectors for electrodes and a computer and power connection over USB. Thus, DStat can be used anywhere a computer can be operated, even in the field using a laptop on battery power. DStat's analogue components were chosen to be compatible with the 5 V supplied by the USB connection, but the digital components of the instrument require 3.3 V. In DStat, this is supplied by a low-dropout regulator (TPS79933, Texas Instruments). A high precision voltage reference (MAX6126, Maxim Integrated) provides an accurate 3 V source with which to reference analogue components. The reference is also used to power a 1.5 V line, formed using a precision voltage divider (MAX5491, Maxim Integrated) and a low noise op-amp buffer (OPA350, Texas Instruments) which serves as a signal ground for the analogue circuitry. This restricts the full potential range of the potentiostat to ±1.5 V (relative to the signal ground), sufficient for most aqueous voltammetry applications.

Reducing microcontroller loading

As described in the main text, DStat relies on an ATxmega256A3U microcontroller (Atmel Corporation, San Jose CA, USA) to manage the DAC and ADC, and to switch the measurement resistors. Criteria evaluated in selecting this component included the interrupt controller and event and timing control, features that allow tasks to be performed concurrently without overloading the CPU. The interrupt controller is a common feature found on microcontrollers that allows the normal program flow to be interrupted by certain signals, either from external sources (from other chips on the board, over USB) or from internal peripherals. When an interrupt is triggered, the main program is suspended, a section of code known as an interrupt handler (specific to each interrupt) is executed, and then the main program resumes from where it was suspended. In DStat, an interrupt can be triggered when the external ADC signals that data is ready, sending the data to the computer, which allows the CPU to handle other tasks rather than constantly polling the ADC to check if data is ready. An interrupt is also used to set new DAC values, triggering when a timer overflows (see below) indicating that a new value should be set. The interrupt controller of the xmega series microcontrollers is particularly flexible in that it permits different priorities to be set for each interrupt. In DStat, this is used to allow data to be read from ADC before a subsequent measurement occurs, without being interrupted by DAC writes, which are not timing critical.

A second common microcontroller feature is the timer/counter (TC). TCs are peripherals that store a number that can be incremented or decremented without use of the CPU and will overflow when it reaches its minimum or maximum value, continuing to count from the other value. A TC's current value can be read or set explicitly and can be used as either a timer or a counter, depending on its source. When used as a timer, the TC increments automatically at a fraction of the CPU frequency, and by setting the timer's maximum value, the time between overflows can be controlled. When used as a counter, the TC is incremented manually when called by the CPU or can be incremented by the overflow of a timer (a special feature of the xmega series). DStat uses TCs to perform many functions without constantly loading the CPU. For example, (a) timers are used to keep track of when the value of the DAC should be changed and for providing elapsed time values for measurements, and (b) counters are used to store the current DAC value, incrementing or decrementing automatically when the DAC timer overflows, to produce a linear voltage output for linear scan voltammetry and cyclic voltammetry.

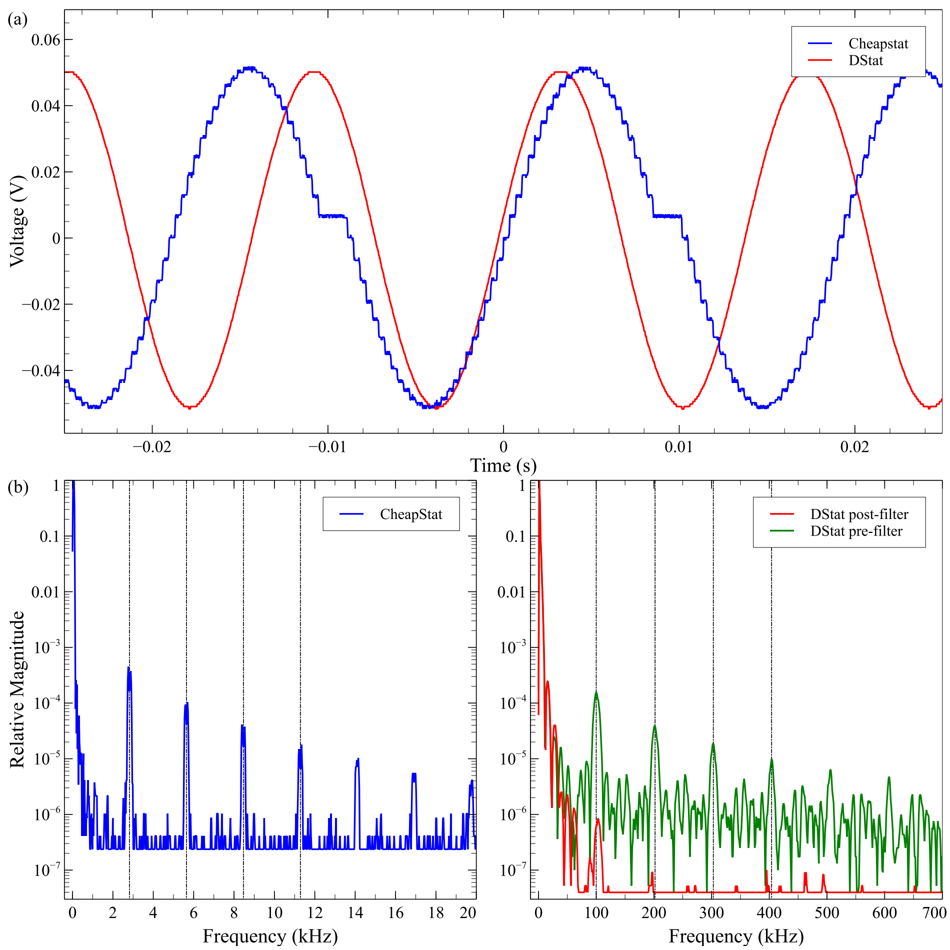

Figure A. Comparison of DAC-programmed control signals in DStat and CheapStat for a sine wave output at 70 Hz with an amplitude of 50 mV. (a) Time domain output of DStat (red) and CheapStat (blue). The CheapStat output shows harmonic distortion from a small delay between periods as well as visible aliasing. (b) Frequency domain output of the CheapStat (left) and DStat (right). Dashed lines indicate the first four harmonics of the sample frequency. Both potentiostats show images due to aliasing, without a reconstruction filter. A 4th order low pass filter with a cutoff of 35 kHz eliminates the majority of images in DStat. (green - without filter, red - with filter)

S2: Analysis of noise measurements

Fig. 4 in the main text shows a gradual decrease in noise reduction as a function of increasing gain in both experiments (blue diamonds) and simulations (green circles). This can be explained by recognizing two different types of noise sources in the system: voltage noise and current noise. Voltage noise is noise introduced after the current to voltage conversion and is produced by the op-amp of the transimpedance amplifier (U3), as thermal noise in the gain resistor, and as part of the ADC's analogue circuitry. The thermal noise contribution to the voltage noise increases with the square root of the resistance of R_M, while the other components are invariant with gain. Current noise is noise introduced before the transimpedance amplifier's input, including the op-amp's inherent current noise as well as noise coupled into the input line from external sources (e.g., environmental noise from nearby traces on the circuit board). Current noise is amplified by the transimpedance amplifier and its contribution to the output noise is therefore proportional to the gain. The data in Fig. 4 in the main text shows the relationship between the two noise types. At low gain, amplification of current noise is low and voltage noise dominates, resulting in a linear decrease in noise as gain increases. At higher gains, the current noise is amplified significantly and begins to reduce the signal-to-noise ratio enhancement with increasing gain. The practical significance of the noise behaviour is that DStat's signal to noise ratio increases with increasing gain settings throughout the entire range of available measurement resistors and since measurements are never limited by the ADC, the noise response curve can be used to select an appropriate gain for a given signal.

Finally, the observation that the simulation (green circles in Fig. 4 in the main text) indicates slightly lower noise than the experimental data (blue diamonds in Fig. 4 in the main text) can be explained by recognizing that the simulation did not account for noise coupling to the transimpedance amplifier inputs. Thus, the excess noise in the experimental data likely originates from sources beyond the current measurement circuit and could potentially be improved by changes in the circuit board layout, or shielding of the entire DStat.

S3: Potentiostatic circuit stability

As with all amplifier circuits, certain operating conditions can cause instability of a potentiostatic circuit which may manifest as overshoot, poorly-damped ringing, or in extreme cases full-scale oscillations at the CE output. Instability occurs when the phase shift in the circuit's feedback loop (the path between the output and the inverting input of U1 in Fig. 2 in the main text) is sufficient to produce positive rather than negative feedback. Many electrochemical cells are especially prone to introducing instability for two reasons: (1) the electrical double layer at the surface of the WE adds a capacitance between the RE and ground, adding phase shift, and (2) extra resistance at the RE increases the phase shift of any capacitance in the feedback loop. The double layer capacitance is proportional to the WE area, with specific capacitances typically ranging between 10 and 40 $\mu F/cm^2$, and is also dependant on the cell potential [1].

While DStat was not designed for large capacitive electrodes, it is capable of operating stably with a purely capacitive load and with well-damped ringing when a 10 kΩ resistance is added at the RE terminal. Stability was assessed by connecting a capacitive load of 1.12 µF between the RE and WE terminals and applying a 1 kHz square wave with an amplitude of 100 mV from DStat's DAC to the potentiostatic circuit. The output of the potentiostatic circuit was measured at the RE terminal with a TDS 2001C oscilloscope (Tektronix, Inc., Oregon, USA). The output with no resistance at the RE terminal (RE and CE shorted) as well as with a RE resistance of 10 kΩ is plotted in Fig. A. The square wave was reproduced without evidence of instability when no resistance was added to the RE and showed some ringing with resistance added to the feedback loop, quickly decaying to the intended output value within 120 µs.

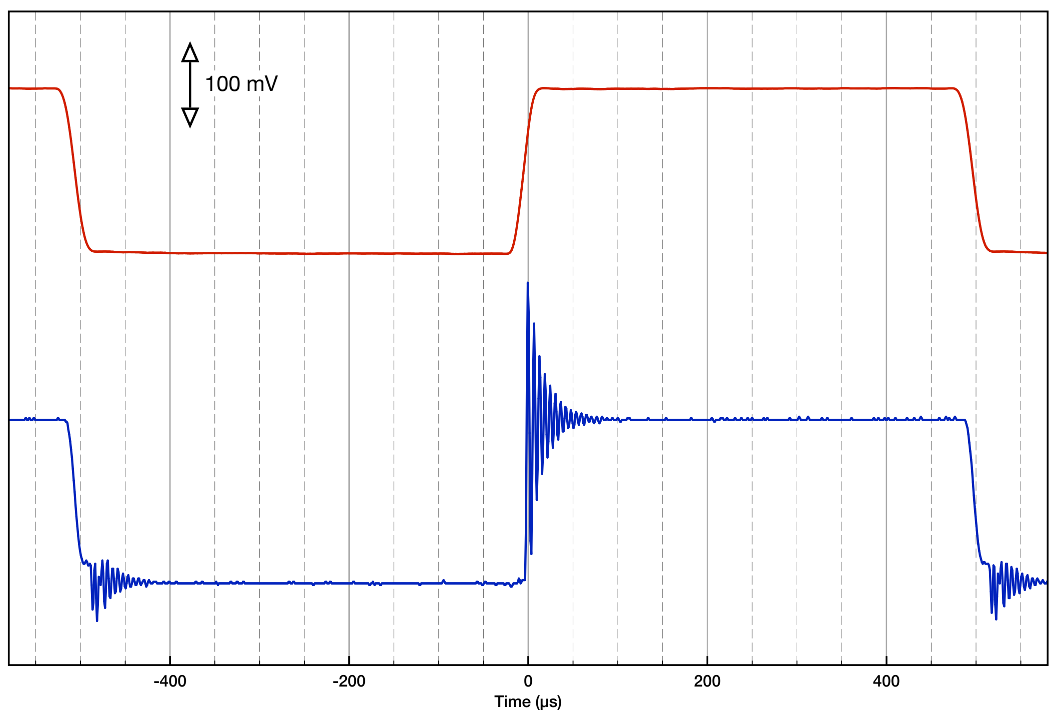

Figure A. DStat potentiostatic circuit square wave (1 kHz) performance under 1.12 µF capacitive load. The output measured at the RE terminal with CE and RE terminals shorted is plotted in red (top trace) and shows no signs of instability. The output measured at the RE terminal with a 10 kΩ resistor between CE and RE terminals is plotted in blue (bottom trace) and shows some well-damped ringing, decaying within 120 µs.

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S4: Potentiometry

pH 3, 7, and 11 calibration standards were purchased from Fisher Scientific Company (Ottawa ON, CA). pH measurements of each standard were collected using a combination glass pH electrode with integrated Ag/AgCl reference electrode (Beckman Coulter Canada, Mississauga ON, CA) with DStat and an Accumet AR50 benchtop meter (Thermo Fisher Scientific, Waltham MA, USA). The results are shown in Fig. A.

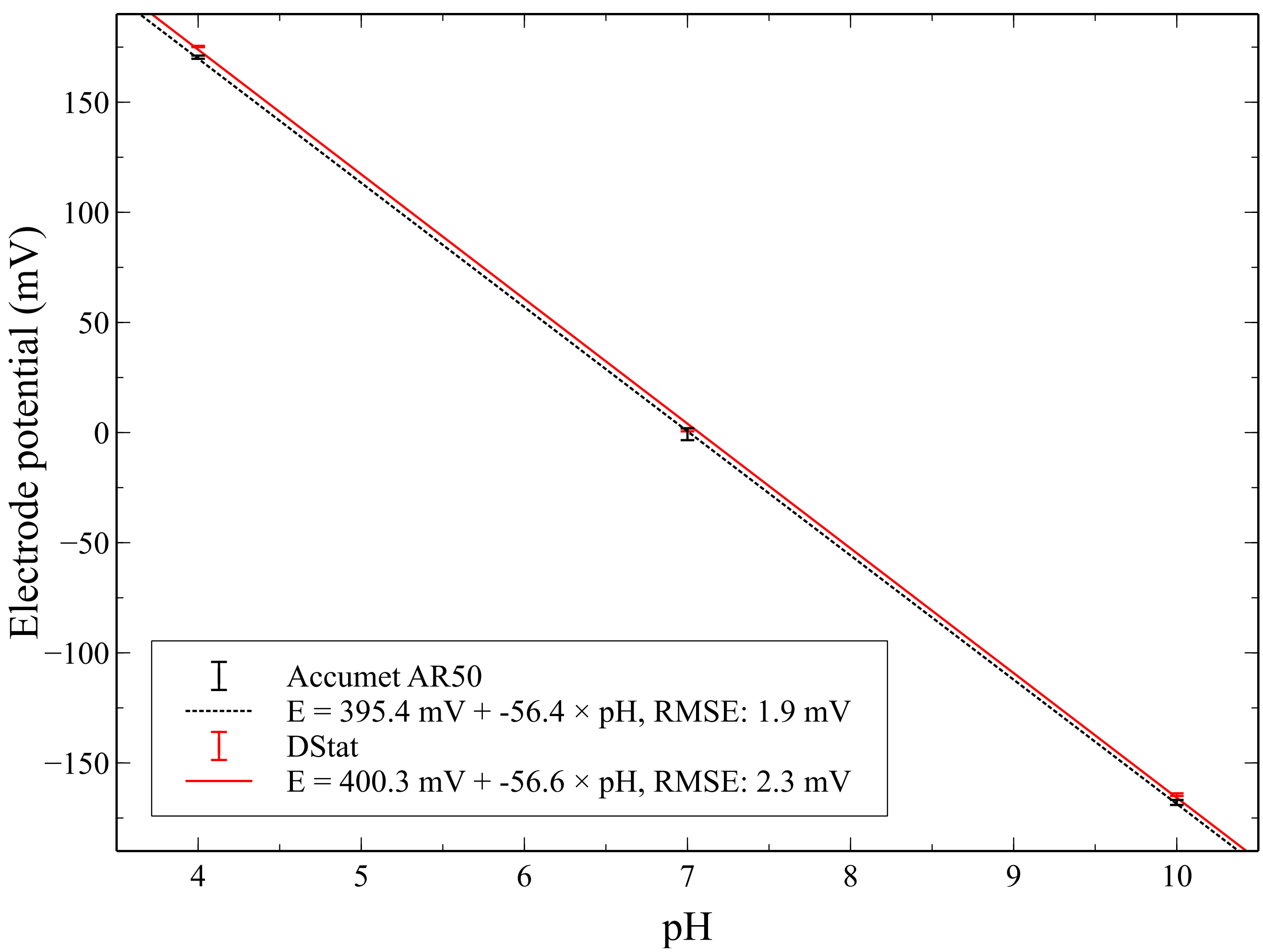

Figure A. Comparison of DStat and Accumet AR50 measurements of pH standard solutions with a combination glass pH electrode. Error bars are ±1 s.d. (n=3). Accumet AR50 measurements are plotted as black bars and DStat measurements as red bars. Linear least squares fit for the Accumet AR50 is shown as a dashed black line and DStat's is shown as a solid red line. RMSE: Root Mean Square Error

S5: Integration of DStat with Dropbot

DMF device fabrication

Digital microfluidic (DMF) bottom plates were formed with chromium driving electrodes on glass substratesas described previously [1] but with one modification: the use of Fluoropel 1604V (Cytonix LLC, Beltsville, MD, USA) in place of Teflon AF as the hydrophobic coating. Fluoropel 1604V was diluted 1:3 with PFC 110 (Cytonix LLC), to produce a 1% Fluoropel solution. The solution was spin-coated (30 s, 2000 rpm, 500 rpm/s) onto the devices after the Parylene coating step and baked for 10 min at 140 °C. Hybrid DMF top plates bearing electrochemical cells embedded in a DMF-counter-electrode were formed using methods similar to those described by Shamsi et al. [2]. Differences included the use of Fluoropel instead of Teflon-AF for the coating (as above), and the use of a three-electrode cell configuration in place of a two-electrode cell. The cell used here is similar to that described by Dryden et al. [3], comprising a 400 µm-radius circular gold-plated working electrode surrounded by a 30 µm wide annular region containing a counter electrode (270° of annulus) and a silver-plated reference electrode (90° of annulus).

Device operation

Each DMF device was assembled from a bottom plate and a top plate separated by two pieces of 3M Scotch double-sided tape (St. Paul, MN, USA) with a total spacing of approximately 180 µm. Droplet actuation was performed by applying sinusoidal signals of approximately 100 V_{rms} at 10 kHz between the top plate electrodes and electrodes on the bottom plate with the open-source DropBot DMF control system [4]. A 1.5 µL droplet of 10 mM potassium hexacyanoferrate(II) was dispensed from a reservoir and then driven to the electrochemical cell (on the top plate). A cyclic voltammogram (2 scans between -600 mV to 200 mV at 50 mV/s) was acquired with DStat and the droplet was driven away from the electrodes. The interplay between DropBot and DStat is illustrated in Figure 6 in the main text, and video of the whole process is included in S6 Video. (In the video, the recording of the DStat measurement is accelerated 4x for brevity.)

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