Note 1: Resolution of mass accumulation rate sensor

How precisely can we measure the mass accumulation rate of a cell? To measure the mass accumulation rate, we will measure the cell size k times, once every Δt minutes, and fit a model to explain how it varies over time. Here we will assume the total duration of the measurements ($k\Delta t$) is short enough that a line is an appropriate model. So the problem becomes, how precisely can we know the slope of a line?

Fortunately, least-squares slope estimates can be written as a linear combination of the observed size values, Y, as follows [5]:

$$\begin{bmatrix}\text{intercept}\\ \text{slope}\end{bmatrix} = (X^T X)^{-1} X^T Y$$

Here X is a $k \times 2$ matrix, where the first column is filled with ones, and the second column corresponds to the evenly-spaced times at which the cell size is measured. For simplicity, we assume the times are mean-centered, yielding the following time vector:

$$\begin{bmatrix}\frac{-k+1}{2}\Delta t & \frac{-k+3}{2}\Delta t & \frac{-k+5}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix}$$

which we will generally simply denote as $\begin{bmatrix}\frac{-k+1}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix}$. Plugging in this definition of X gets us to the coefficient vector relating the measured sizes to the slope estimator (specifically, this coefficient vector is the second row of $(X^T X)^{-1} X^T$).

$$\begin{bmatrix}\text{intercept}\\ \text{slope}\end{bmatrix} = \left(\begin{bmatrix}1 & \dots & 1\\ \frac{-k+1}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix} \begin{bmatrix}1 & \frac{-k+1}{2}\Delta t\\ \vdots & \vdots\\ 1 & \frac{k-1}{2}\Delta t\end{bmatrix} \right)^{-1} \begin{bmatrix}1 & \dots & 1\\ \frac{-k+1}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix} \begin{bmatrix}y_1\\ \vdots\\ y_k\end{bmatrix} \tag{1}$$

$$\begin{bmatrix}\text{intercept}\\ \text{slope}\end{bmatrix} = \left(\begin{bmatrix}k & 0\\ 0 & \Delta t^2 \frac{k^3-k}{12}\end{bmatrix} \right)^{-1} \begin{bmatrix}1 & \dots & 1\\ \frac{-k+1}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix} \begin{bmatrix}y_1\\ \vdots\\ y_k\end{bmatrix}$$

$$\begin{bmatrix}\text{intercept}\\ \text{slope}\end{bmatrix} = \frac{1}{k\Delta t^2 \frac{k^3-k}{12}} \begin{bmatrix}\Delta t^2 \frac{k^3-k}{12} & 0\\ 0 & k\end{bmatrix} \begin{bmatrix}1 & \dots & 1\\ \frac{-k+1}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix} \begin{bmatrix}y_1\\ \vdots\\ y_k\end{bmatrix}$$

$$\text{slope} = \frac{k}{k\Delta t^2 \frac{k^3-k}{12}} \begin{bmatrix}\frac{-k+1}{2}\Delta t & \dots & \frac{k-1}{2}\Delta t\end{bmatrix} \begin{bmatrix}y_1\\ \vdots\\ y_k\end{bmatrix}$$

$$\text{slope} = \frac{1}{\Delta t \frac{k^3-k}{12}} \begin{bmatrix}\frac{-k+1}{2} & \dots & \frac{k-1}{2}\end{bmatrix} \begin{bmatrix}y_1\\ \vdots\\ y_k\end{bmatrix}$$

Since the slope estimate is a linear combination of observed size values $Y = [y_1\ y_2\ \dots\ y_k]^T$, errors also propagate linearly. If all the size measurements have independent and identically distributed errors with mean zero and root-mean-square-error (RMSE) σ_ϵ, then the slope RMSE is σ_ϵ times the magnitude of the coefficient vector, $\frac{1}{\Delta t \frac{k^3-k}{12}} \begin{bmatrix}\frac{-k+1}{2} & \dots & \frac{k-1}{2}\end{bmatrix}$. The magnitude of $\begin{bmatrix}\frac{-k+1}{2} & \dots & \frac{k-1}{2}\end{bmatrix}$ is $\sqrt{\frac{k^3-k}{12}}$, therefore

$$\sigma_{\text{slope}} = \frac{\sigma_\epsilon \sqrt{12}}{\Delta t \sqrt{k^3-k}} \approx \frac{\sigma_\epsilon \sqrt{12}}{\Delta t k^{1.5}} \tag{2}$$

It is worth note that we could parameterize this instead in terms of total time transiting the array, $T = k\Delta t$, with k measurements occurring at evenly-spaced increments throughout this interval.

$$\sigma_{\text{slope}} \approx \frac{\sigma_\epsilon \sqrt{12}}{T \sqrt{k}}$$

In this form, it is clearly seen that the standard error scales inversely proportional to the total measurement duration, with a $\sqrt{k}$ dependence on the number of measurements made during that interval (in direct analogy to the central limit theorem).

We can use equation (2) generally to estimate the resolution of any sytem measuring rates of mass or volume increase, but specifically in the case of a serial SMR array, it also provides a convenient way to express the effect of the flow rate, which controls the trade-off between mass accumulation rate resolution and throughput. As we increase the flow rate, we decrease Δt proportionally, which decreases the mass accumulation rate resolution. Simultaneously, the throughput goes up directly proportionally to flow rate. An added effect is that faster flow rates yield larger *mass* error, σ_ϵ, as the cell spends a smaller amount of time in the cantilever and therefore cannot filter out as much frequency noise. For white-noise-dominated resonant frequency measurements (here corresponding to flow rates faster than what we've utilized in this paper for large-channel devices, e.g. $\Delta t < 2$ minutes), we expect that σ_ϵ will scale roughly inversely proportional to the square root of Δt:

$$\sigma_\epsilon = \frac{\alpha}{\sqrt{\Delta t}} \tag{3}$$

Plugging this into (2) suggests that as we increase the flow rate, the mass accumulation rate error is expected to scale with throughput ($1/\Delta t$) to the three-halves.

$$\sigma_{\text{slope}} = \frac{\alpha\sqrt{12}}{\Delta t^{1.5}\sqrt{k^3 - k}} \tag{4}$$

We have illustrated this resolution-noise trade-off in Supplementary Figure 14. For slower flow, σ_ϵ may not be dominated by white frequency noise but instead by flicker (pink) or brown noise, and therefore (3) will sizably underestimate the actual mass noise magnitude.

Note 2: Mass sensitivity scales with frequency$^{3/2}$ for varied cantilever lengths

The cantilever resonant frequency f is given by $f = \frac{1}{2\pi}\sqrt{\frac{k}{m_{\text{eff}}}}$, where k is the spring constant and m_{eff} is the effective mass of the cantilever. m_{eff} is proportional to the cantilever length l [6], and k is proportional to $1/l^3$ [7], therefore $f \propto \frac{1}{l^2}$.

We can similarly determine how the cantilever mass sensitivity [8], s depends on length: $s \propto \frac{f}{m} \propto \frac{1}{l^3}$. Combining these two facts, we find s will be proportional to $f^{3/2}$, when all dimensional parameters other than the length of the cantilever are kept constant.

Note 3: Explanation of peak matching algorithm

We attempt to identify all the peaks (up to twelve, one in each cantilever) that we believe originate from the same cell. To do this, we use a heuristic approach in which we build "cells", collections of peaks that we believe belong to the same cell. At each cantilever in turn, starting at the second cantilever, we try to match the observed peaks at that cantilever with the previously observed cells. To match peaks to their corresponding cells, we define a cost function, detailed below, representing our assumptions about how cells both grow and flow through the device. We then try to find a way of pairing the already-observed cells (from the first n cantilevers) with the peaks observed at cantilever n+1 that minimally violates our expectations. Additionally, we also include the possibility that a cell is not observed at a particular cantilever, possibly due to simultaneously entering the cantilever at the same time as another cell, or adhering to the device walls. We include this possibility by adding fictitious cells ("gaps") such that a previously-observed cell can be assigned to a gap if there are no peaks at sensor n that are likely to originate from that cell. Similarly, a peak in cantilever n+1 can be assigned to a gap if it doesnt appear to clearly appear to correspond to an existing cell.

Pseudo-code for our matching approach is given below (variables are denoted in blue):

```
Initialize each peak in sensor 1 as its own cell, put them all in cellList

For each sensor n in 1:(numberOfSensors-1)
   peaksToBeAssigned = all peaks in sensor n+1

   costs = matrix( number of rows = length(cellList),
                   number of columns = length(peaksToBeAssigned) )
   Pad costs with extra rows and columns for 'unassigned' cells
   Set entries of costs for assigning a cell to 'unassigned' to gapCost
   Set entries of costs for assigning 'unassigned' to 'unassigned' to 0

   For each r in 1:length(cellList)
      For each c in 1:length(peaksToBeAssigned)
         costs[r,c] = -log( P(peaksToBeAssigned[c] | cellList[r]) )

   Find optimal assignment for costs via Hungarian algorithm

   Any peaks in peaksToBeAssigned that were assigned to existing cells in the cellList
   should be concatenated onto the end of their corresponding cell.

   Any entries in peaksToBeAssigned NOT assigned to existing cells are added to the
   cellList as cells containing only one peak.
```

The heart of this approach is how we define a cost function representing our prior assumptions about device behavior (e.g. cells take approximately two minutes to transit from one cantilever to the next, and can't possibly show up at cantilever 2 before appearing at cantilever 1) and cell behavior (over such a short time period, cell mass usually changes roughly linearly, and the rate of change is unlikely to be extremely large). To represent these assumptions, we use a probabilistic model of seeing a peak of a particular mass and time at sensor n+1, given the previous n peaks we've already decided are part of the cell's trajectory. Using the negative log of the probability gives us a cost function for which minimizing the cost corresponds to maximizing the likelihood of the data.

We model the probability of observing a peak of mass m_{n+1} and at time t_{n+1}, conditioned on the peak occurring at sensor n+1 and having observed previous peaks of masses $m_{1:n}$ at times $t_{1:n}$, as follows:

$$P(m_{n+1}, t_{n+1} | m_{1:n}, t_{1:n}) = P(m_{n+1} | t_{n+1}, m_{1:n}, t_{1:n}) P(t_{n+1} | m_{1:n}, t_{1:n})$$

We then assume t_{n+1} depends only on t_n, and is normally distributed with mean $t_n + \mu_{\Delta t}$ and variance $\sigma^2_{\Delta t}$, where $\mu_{\Delta t}$ and $\sigma^2_{\Delta t}$ are specified by the user *a priori*. We further posit that m_{n+1} should be related to t_{n+1} and the previous data via the following relation

$$m_{n+1} = \beta_1 t_{n+1} + \beta_0 + \epsilon$$

where β_1 is a random variable corresponding to the slope implied by the previous datapoints, β_0 is the y-intercept, and ϵ is random instrument noise. If we mean-center the time values ($\sum_{i \in 1:n} t_i = 0$), then β_0 and β_1 become uncorrelated, and we can thus express the mean and variance of m_{n+1} as

$$\mu_{m_{n+1}} = t_{n+1}\mu_{\beta_1} + \mu_{\beta_0}$$
$$\sigma^2_{m_{n+1}} = t^2_{n+1}\sigma^2_{\beta_1} + \sigma^2_{\beta_0} + \sigma^2_\epsilon$$

Furthermore, if β_1, β_0 and ϵ are assumed normal, then m_{n+1} is normally distributed with the above parameters.

While it is straightforward to obtain frequentist estimates of β_1 and $\sigma^2_{\beta_1}$ when we have already seen many datapoints, we cannot estimate these quantities easily with only one or two datapoints. To mitigate this we use Bayesian estimators, which are shaped by a prior distribution when only one or a few datapoints are available, and shaped more by the data when more data becomes available. The conjugate prior for β_1 is normal (assuming the mass sensor error parameter σ_ϵ is already known) and is specified by hyper-parameters $\overline{\mu}_{\beta_1}$ and $\overline{\sigma}^2_{\beta_1}$. The posterior distributions for β_1 is also normal, with variance and mean as follows:

$$\sigma^2_{\beta_1} = \frac{1}{\frac{1}{\overline{\sigma}^2_{\beta_1}} + \frac{t_{1:n} \cdot t_{1:n}}{\sigma^2_\epsilon}}$$

$$\mu_{\beta_1} = \sigma^2_{\beta_1}\left(\frac{\overline{\mu}_{\beta_1}}{\overline{\sigma}^2_{\beta_1}} + \frac{t_{1:n} \cdot m_{1:n}}{\sigma^2_\epsilon}\right)$$

We also assume that since σ_ϵ is known, μ_{β_0} is just the mean mass from the n previous observations, and $\sigma^2_{\beta_0} = \sigma^2_\epsilon / n$. Using these parameters, we can then write the cost function as:

$$\text{Cost}(m_{n+1}, t_{n+1} | m_{1:n}, t_{1:n}) = -\log\left[N(m_{n+1} | \mu_{m_{n+1}}, \sigma^2_{m_{n+1}})\right] - \log\left[N(t_{n+1} | t_n + \mu_{\Delta t}, \sigma^2_{\Delta t})\right] \quad (5)$$

where $N(x|\mu, \sigma^2)$ is the normal density function evaluated at x with mean μ and variance σ^2. Examples of this cost function for simulated cells are shown in Supplementary Figure 4, demonstrating how this cost function narrows as more and more data is observed.

In sum, the cost depends on the new data (m_{n+1}, t_{n+1}), the previously observed data ($m_{1:n}$, $t_{1:n}$), and five user-defined parameters:

parameter	description
σ	sensor RMS error
$\overline{\mu}_{\beta_1}$	prior expectation for mean mass accumulation rate
$\overline{\sigma}^2_{\beta_1}$	prior expectation for mass accumulation rate variance
$\mu_{\Delta t}$	expected average time between sensors
$\sigma^2_{\Delta t}$	expected variance in time between sensors

Additionally, there is one more parameter for the cost of a gap, yielding six parameters in total controlling the matching process.

It is worth note that by simply choosing the best matching between the previously-observed cells and the newly-observed peaks at every step, we do not properly take into account uncertainty in the matching process. While we have not undertaken this task here, future work to do so may utilize Murty's algorithm [9] to obtain not only the optimal assignment (as provided by the Hungarian algorithm), but a ranked set of the best assignments (e.g. the top 50 assignments). This would allow one to check which assignments are tenuous and which are very certain.

Note 4: Comparison of measurement precision between SMRs and quantitative phase microscopy (QPM)

Comparisons of SMR and QPM mass measurements cannot be made directly because the two methods exploit different physical principles. QPM requires computing an unwrapped phase shift function from image data to yield optical thickness. Optical thickness is integrated over the area of a cell, and the result is multiplied by a constant to convert it into dry mass units. The constant is based on an average refractive increment of mostly globular proteins [10]. On the other hand, SMR measurements are based on the change in resonant frequency of an oscillating cantilever caused by a cell passing through an embedded microfluidic channel. The frequency shift is divided by a sensitivity constant (Hz/pg) to obtain buoyant mass. The sensitivity constant is a device parameter and independent of the properties of the analytes. It is obtained by direct calibration with particles of known buoyant mass.

Supplementary Figures 13A and 13C show mass measurements of *E.coli* cells with similar interdivision times made by QPM (left panel) and SMR (right panel). The left panel shows dry mass versus time for three *E. coli* cells measured by Mir et al. using QPM [3]. Buoyant mass versus time for 11 *E. coli* cells measured by SMR is shown on the right. Buoyant mass error for these cells is 0.22 fg, based on repeat measurements of a single inert polystyrene particle similar in size and density to these cells (1.36 μm diameter, 1.05 g/mL). One way to compare the precision of the two methods is to convert the SMR buoyant mass to dry mass. This is possible using a method we validated in a previous study [11]. Briefly, we measured the buoyant mass of *E. coli* cells in two fluids with different densities. The first fluid was a standard phosphate-buffered saline solution. The second fluid was identical to the first, except the water in the formula was replaced by heavy water (D_2O). Using the method of Archimedes, we found the density of *E. coli* biomass (*E. coli*'s dry density) to be 1.45 g/mL. This constant can be used to convert buoyant mass to dry mass, as shown on the right axis in Supplementary Figure 13C. The conversion produces good agreement between the *E. coli* dry mass obtained by the two methods. Converting buoyant mass error to dry mass error yields an error of 0.63 fg, approximately 30 times smaller than values produced by QPM.

Another way to assess the precision of the two methods is to compare the relative uncertainties of both techniques. Because they are unitless, relative uncertainties can be compared directly. In Mir et al. [3], measurements of a $\sim$1.5 pg cell dry mass have a standard deviation of $\sim$0.0196 pg, yielding a relative uncertainty of 1.3%. SMR measurements of similarly-sized cells with an average buoyant mass around 0.3 pg have a standard deviation of 0.20 fg, or 0.06% relative uncertainty, about 20 times better precision than QPM. Both the dry mass conversion and relative uncertainty approaches give similar values.

We also find that serial and single SMR measurements of mass accumulation rates exhibit less variation than those measured by QPM. Supplementary Figures 13B and 13D compare mass accumulation rate measurements obtained by QPM and SMR. The left panel (Supplementary Figure 13B) shows dry mass accumulation rate (referred to as growth rate) vs. dry mass of 20 cells measured in reference [3]. Supplementary Figure 13D shows two analogous datasets produced by the SMR method - one taken with a single SMR device at low throughput and another taken on a serial SMR array with a higher flow rate.

How does the noise of the single SMR compare to the serial SMR array results shown in Figure 6 and Supplementary Figure 13? The uncertainty in SMR buoyant mass measurements depends on the flow rate of cells through the device. By varying the flow rate, the system can be optimized for throughput or measurement precision. The noise level in the serial SMR array measurements (Figure 6) is about 3 times higher than the single SMR in Supplementary Figure 13C because of the faster flow rate. This could be reduced at the expense of lower throughput.

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