Supplementary Material

Bimodal Analysis of Mammary Epithelial Cell Migration in Two Dimensions

Alka A. Potdar, Jenny Lu, Junhwan Jeon, Alissa M. Weaver and

Peter T. Cummings

The supplementary material given below provides additional supporting information that is considered too detailed for the manuscript itself. In each case, simulations are performed to support our interpretation of the experiments.

Effect of Criteria r1, r2, and r3 for Change of Mode

We include here a comparison (Fig. S1 a, b) of an artificially created cell trajectory (using cellular dynamics simulation based on the bimodal model and parameters extracted from bimodal analysis) with an analysis of this artificial trajectory with the bimodal analysis. The cellular dynamics simulation scheme used here has the same basic algorithm that was used to simulate bacterial migration[1], modified to have a bimodal motion with experimentally determined turn angle, directional-mode and re-orientation-mode distributions. We can see that all the turns (known independently from simulation) in the artificial trajectory are picked up by the r3 analysis technique.

[1] Frymier, P. D., R. M. Ford, and P. T. Cummings. Cellular-dynamics simulations of bacterial chemotaxis. *Chem. Eng. Sci.* 48: 687-699, 1993.

The less-stringent r1 criterion (directional-mode characterized by only one frame) reduces the duration of the re-orientation-mode (Fig. S2) and picks up more directional and re-orientation modes. The mean directional-mode time and re-orientation-mode times are reduced with r1 criterion. The same is found experimentally: for example for neuT cells with r1 criterion, the directional-mode time is 2.42±0.63 minutes and re-orientation-mode time is 1.86±0.50 minutes, compared with 3.5±0.7 and 4.3±1.2 minutes respectively using the r3 criterion.

Impact of Sampling Frequency on Average Speed

The squared difference in the speed at $\Delta t_{sampling} = 0.5$ minute sampling interval and speed at any higher sampling interval ($\Delta t_{sampling} > 0.5$ minute) reduces as $\Delta t_{sampling}$ approaches 0.5 minutes. The measured speed is found to increase as the sampling time interval is decreased. A simple random walk simulation we performed illustrates this observation. We perform a simulation with time step Δt_{sim}= 0.5minute. The lattice spacing is $\ell = 0.5\mu\text{m}$, and in the random walk we move one lattice spacing per step, so the actual speed is $v_{actual} = \ell / \Delta t_{sim} = 0.5\mu m / 0.5$minute, or $v_{actual} = 1$ μm/minute. We know from the theory of random walks that the displacement of the random walker after n steps will be $r = A(n\Delta t_{sim})^{\alpha}\ell$, where at short enough times $\alpha = 1$ (corresponding to ballistic motion) and at long times $\alpha = 0.5$ (corresponding to diffusive motion) and A is a pre-factor to match the units, which at long time is simply related to the diffusion coefficient. Suppose the measurement interval is $\Delta t_{sample} = m\Delta t_{sim}$. For example, a sampling time of 2.5 minutes corresponds to $m = 5$. We expect that the measured speed will be:

$$v_m^{measured} = \frac{r_m}{t_{sample}} = \frac{A(m\Delta t_{sim})^{\alpha}\ell}{m\Delta t_{sim}} = Am^{\alpha-1}(\Delta t_{sim})^{\alpha}\frac{\ell}{\Delta t_{sim}} = Am^{\alpha-1}(\Delta t_{sim})^{\alpha}v_{actual} \qquad (1)$$

If $m=1$, we can assume we are in the ballistic regime, so equation 1 gives $v_1^{measured}=v_{actual}$. And, indeed, we see from Fig. S3 that the sampled speed is in fact the actual speed. However, for $m>1$ (which in the experiment corresponds to sampling times greater than 0.5 minute, such as 2.5, 5 and 15 minutes), equation (1) predicts that the sampled speed will be less than the actual speed. In fact, the curve in Fig. S3 has the same characteristics as the experimental curve (Fig. 11). Thus we see from Fig. S3 that although the sampled speed does not appear to asymptote to a value at short sampling times, in fact it is becoming the exact result. This gives us some confidence that our measured speed at the 0.5 minute interval is a reasonable estimate of actual cell speed.

Figure legends for supplemental figures:

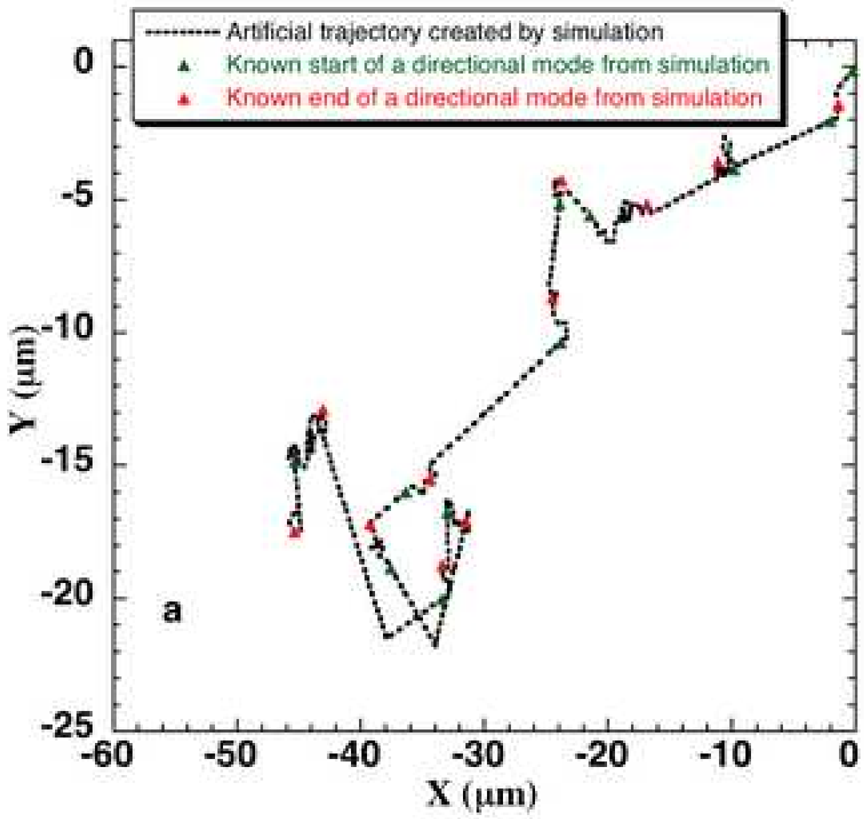
Figure S1: a) An artificial trajectory of a cell created using cellular dynamics simulation with the known directional and re-orientation modes indicated. b) Analyzed artificial trajectory with (r3 criterion and $\phi_{cut} = 45°$ cut-off) with the two modes indicated.

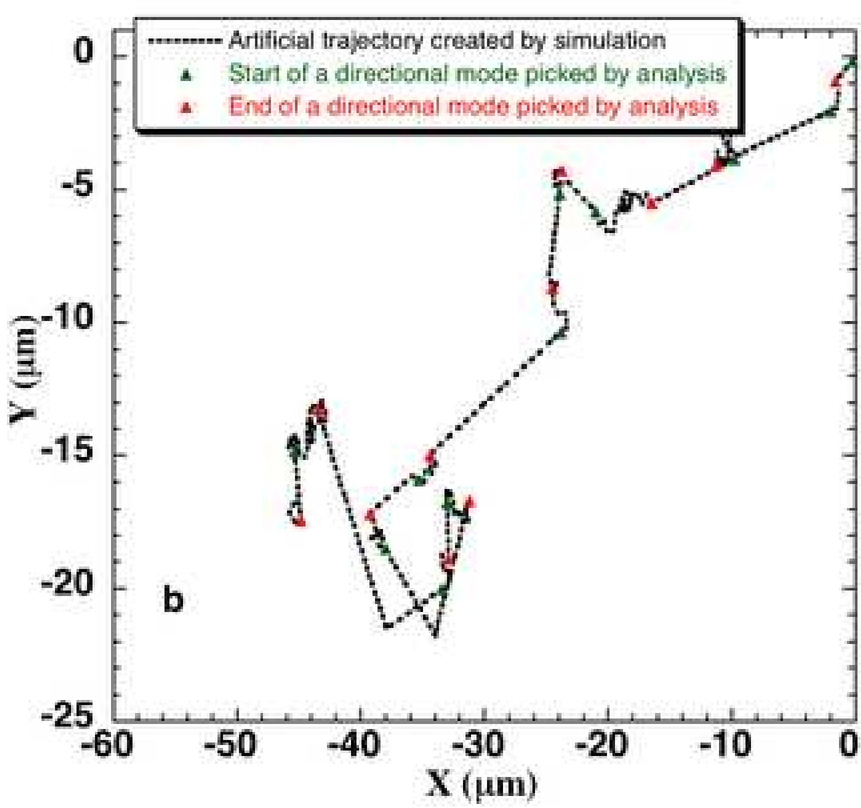
Figure S2: The directional-mode (mode I) and re-orientation-mode (mode II) located in a neuT cell trajectory using $\phi_{cut} = 45°$ and ‘r1 criteria’. The change with respect to r3 criteria (Fig. 3b) has been highlighted in the square inset.

Figure S3: Mean speed versus time trend for a random walk simulation.

Supplemental FigureS1_PotdarAA

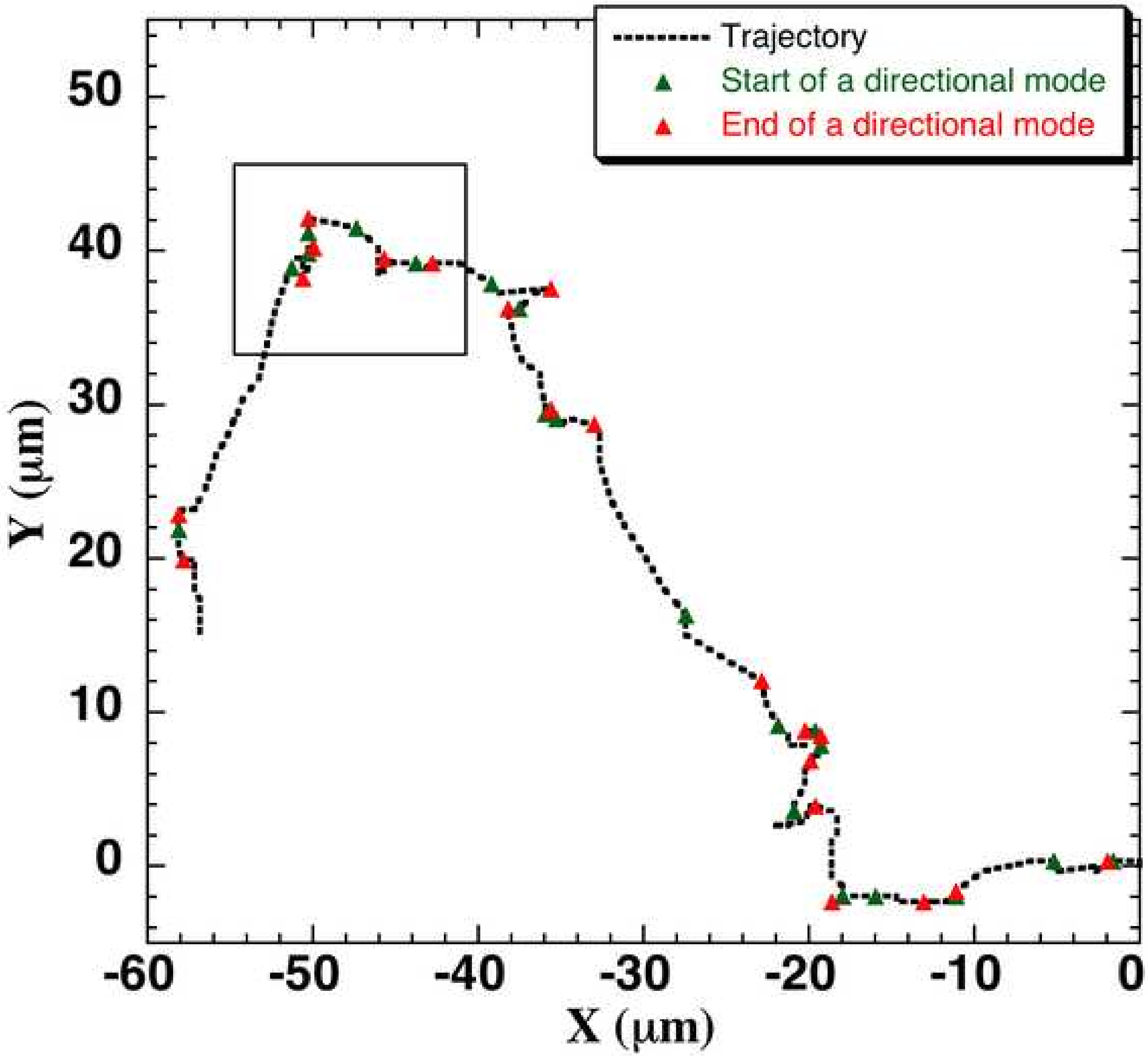
Click here to download high resolution image





Supplemental FigureS2_PotdarAA

Click here to download high resolution image



Supplemental FigureS3_PotdarAA
Click here to download high resolution image