ELECTRONIC SUPPLEMENTARY INFORMATION FOR
Influence of myosin activity and mechanical impact on keratocyte polarization

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Fig. S1 Model results of a keratocyte with a non-uniform $\rho_{\text{cyt}}$ distribution as initial condition (case A2). The rest of the unknowns exhibit a uniform distribution at the initial time. (a) $\rho_r$ distribution on the membrane and $\rho_{\text{cyt}}$ (top half of the cell) and $\rho_m$ (bottom half of the cell) distributions on the cytosol at times $t_0 = 0$, $t_1 = 35$, $t_2 = 70$, and $t_3 = 300$ s. Arrows in the bottom half represent the velocity field $\mathbf{u}$. The dash-dotted line represents the axis of symmetry of the cell traced by the cell’s center of mass. The cytosolic distributions are symmetric with respect to the axis of symmetry. Time evolution of (b) the cell area (black solid line) and perimeter (red dashed line); (c) the velocity of the cell’s center of mass $u_{\text{cell}}$; (d) $\rho_{\text{cyt}}$ and $\rho_r$ at the front edge of the cell at the axis of symmetry, denoted as $\rho_{\text{F cyt}}$ (red dashed line) and $\rho_{F r}$ (black dashed line), respectively; (e) $\rho_{\text{cyt}}$ and $\rho_r$ at the rear edge of the cell at the axis of symmetry, denoted as $\rho_{\text{R cyt}}$ (red solid line) and $\rho_{R r}$ (black solid line), respectively; and (f) cellular shape factor ($SF$, red solid line) and polarization factors $P_t$ (black solid line) and $P_m$ (black dashed line). Vertical dotted lines in the graphs indicate the times $t_1$, $t_2$, and $t_3$. The horizontal green dash-dotted lines in (d) and (e) represent the densities $\rho_{I \text{ cyt}}^F$ and $\rho_{II \text{ cyt}}^F$, which separate regions I and II, and II and III, respectively, in the $f_p$ diagram (see Fig. 2 in the main text).
Fig. S2 Model results of a keratocyte with a non-uniform $\rho_m$ distribution as initial condition (case A3). The rest of the unknowns exhibit a uniform distribution at the initial time. The layout of the figure is analogous to Fig. S1. (a) $\rho_r$, $\rho_{cyt}$, $\rho_m$ and $u$ distributions at four different times. Time evolution of (b) cell area and perimeter; (c) $u_{cell}$; (d) $\rho_{cyt}^F$, $\rho_{cyt}^B$, $\rho_m^B$, and $\rho_m^F$; and (e) cellular shape factor ($SF$), $P_r$, and $P_m$. 

**Fig. S2**

- **(a)** $\rho_m$-polarized initial condition (case A3)
- **(b)** Area ($\mu m^2$) and Perimeter ($\mu m$) evolution over time.
- **(c)** $u_{cell}$ variation over time.
- **(d)** Front and Rear densities over time.
- **(e)** Polariz. Factor (-) and Shape Factor (-) evolution over time.
Fig. S3 Model results of a keratocyte with a non-uniform $\rho_T$ distribution as initial condition and a total amount of RhoA proteins $N_\rho=460.35$ (case A4). The rest of the unknowns exhibit a uniform distribution at the initial time. Note that $N_\rho=488.9$ in the rest of the cases shown in the paper. The layout of the figure is identical to Fig. S1. The cell achieves a polarized and motile steady state, which is slightly different from the steady state of cells with $N_\rho=488.9$; compare with Fig. 4 (case A1) in the main text.
Fig. S4 Comparison between cells with different levels of myosin inhibition. On the left column, results corresponding to $\bar{\eta}_{\text{myo}} = 12\, \text{pN\,\mu m}$ (case B1). On right column, results corresponding $\bar{\eta}_{\text{myo}} = 30\, \text{pN\,\mu m}$ (case B2). The layout of the figure is analogous to subpanels (b)–(f) in Fig. S1. Time evolution of (a) cell area and perimeter; (b) $u_{\text{cell}}$; (c) $\rho_{\text{F cyt}}$, $\rho_{\text{F R cyt}}$, $\rho_{\text{F R}}$, and $\rho_{\text{F}}$; and (d) $\text{SF}$, $P_{\text{R}}$, and $P_{\text{m}}$. High levels of myosin inhibition (left column) impede the attainment of a steady polarized state, while low levels of myosin inhibition (right column) allow for cell polarization and motion.
spatially-localized downregulated myosin activity ($\bar{\eta}_{\text{myo}} = 45$, case C3)

Fig. S5 Spatially-localized downregulation of myosin activity (case C3). Model results of a keratocyte with a non-polarized initial condition (see, e.g., $t_0$ in Fig. 3(a) in the main text). Myosin activity is reduced such that $\bar{\eta}_{\text{myo}} = 45 \text{pN} \mu\text{m}$ in the front half of the cell for $t < t_{\text{myo}} = 40 \text{s}$. Otherwise, $\bar{\eta}_{\text{myo}} = 60 \text{pN} \mu\text{m}$. The layout of the figure is analogous to Fig. S1. (a) $\rho_{\Gamma}$, $\rho_{\text{cyt}}$, and $\rho_{\text{m}}$ distributions at four different times. Myosin activity is downregulated in the striped region (front half of the cell). Time evolution of (b) cell area and perimeter; (c) $u_{\text{cell}}$; (d) $\rho_{\text{F, cyt}}$, $\rho_{\text{F, r}}$, $\rho_{\text{P, cyt}}$, and $\rho_{\text{P, r}}$; and (e) SF, $P_r$, and $P_m$. The sharp transition of the plots at $t_{\text{myo}}$ is due to the sudden change of $\bar{\eta}_{\text{myo}}$, which recovers its standard value ($\bar{\eta}_{\text{myo}} = 60 \text{pN} \mu\text{m}$). The cell is unable to attain a steady polarized state. A higher level of myosin downregulation is required to achieve a polarized and motile steady state (see Fig. 7 in the main text).
spatially-localized upregulated myosin activity ($\eta_{\text{myo}}=80$, case C4)

Fig. S6 Spatially-localized upregulation of myosin activity (case C4). Model results of a keratocyte with a non-polarized initial condition (see, e.g., $t_0$ in Fig. 3(a) in the main text). Myosin activity is increased such that $\eta_{\text{myo}} = 80\text{pN}\mu\text{m}$ in the rear half of the cell for $t < t_{\text{myo}} = 40\text{s}$. Otherwise, $\eta_{\text{myo}} = 60\text{pN}\mu\text{m}$. The layout of the figure is identical to Fig. S5. (a) $\rho_r$, $\rho_{\text{cyt}}$, and $\rho_m$ distributions at four different times. Myosin activity is upregulated in the striped region (rear half of the cell). Time evolution of (b) cell area and perimeter; (c) $u_{\text{cell}}$; (d) $\rho_{\text{cyt}}^F$, $\rho_{\text{cyt}}^R$, $\rho_m^F$, and $\rho_m^R$; and (e) $SF$, $P_r$, and $P_m$. The cell achieves a steady polarized state.
Fig. S7 Influence of the pushing object radius ($R_{\text{imp}}$) on cell polarization by mechanical impact. Time evolution of (a) $\rho_F^{\text{cyt}}$ and $\rho_F^{\Gamma}$; (b) $\rho_R^{\text{cyt}}$ and $\rho_R^{\Gamma}$; (c) $P_R$; (d) $P_m$; and (e) cellular shape factor for three simulations with $R_{\text{imp}} = 1.5$ (blue dotted line), 2.25 (black solid line), and 3.0 $\mu$m (red dashed line). The vertical dotted line in the graphs indicates the time $t_{\text{imp}}$, time at which the pushing force vanishes. The influence of the radius $R_{\text{imp}}$ is very small.
**cell polarization by mechanical impact: object velocity influence**

\[ u_{\text{imp}} = 0.01 \, \mu m/s \]

\[ t_1 = 20 \, s \quad t_2 = 40 \, s \quad t_3 = 80 \, s \quad t_4 = 100 \, s \]

\[ u_{\text{cell}} = 0 \]

Fig. S8 Influence of the pushing object velocity \( (u_{\text{imp}}) \) on cell polarization by mechanical impact. \( \rho_F \), \( \rho_{\text{cyst}} \), and \( \rho_m \) distributions at times \( t_1 = 20 \), \( t_2 = 40 \), \( t_3 = 80 \), and \( t_4 = 100 \, s \) for two simulations with (a) \( u_{\text{imp}} = 0.01 \) and (b) \( u_{\text{imp}} = 0.12 \mu m/s \). Time evolution of (c) \( \rho_F \), (d) \( \rho_{\text{cyst}} \), and \( \rho_m \); (e) \( P_F \); (f) \( P_{\text{cyst}} \); (g) \( SF \); and (h) \( u_{\text{cell}} \) for four simulations with \( u_{\text{imp}} = 0.01 \) (blue dotted line), 0.04 (yellow dash-dotted line), 0.08 (black solid line), and 0.12 (red dashed line). The vertical dotted line in the graphs indicates the time \( t_{\text{imp}} \). The sharp transition of the plots at \( t_{\text{imp}} \) is due to the sudden removal of the pushing force. Small \( u_{\text{imp}} \) does not produce a steady polarized state. As \( u_{\text{imp}} \) increases, the cell polarizes faster.
Influence of the application time ($t_{\text{imp}}$) of the pushing object on cell polarization by mechanical impact. The layout of the figure is analogous to Fig. S8. (a) $\rho_F$, $\rho_{\text{cyt}}$, and $\rho_m$ distributions at four different times. Time evolution of (b) $\rho_F^{\text{c}}$, (c) $\rho_{\text{cyt}}^{\text{c}}$, (d) $P_r$; (e) $P_m$; (f) $SF$; and (g) $u_{\text{cell}}$ for three simulations with $t_{\text{imp}} = 25$ (blue dotted line), 35 (red dashed line), and 85 s (black solid line). The vertical dotted lines in the graphs indicate the corresponding $t_{\text{imp}}$: 25 (blue), 35 (red), and 85 s (black). Short $t_{\text{imp}}$ cannot induce a steady polarized state. The results show that there is a threshold of $t_{\text{imp}}$ to achieve cell polarization. Once the cell reaches that threshold, the influence of $t_{\text{imp}}$ is small (compare $P_r$ and $P_m$ for $t_{\text{imp}} = 35$ and 85 s).
Fig. S10 Influence of myosin inhibition on cell polarization by mechanical impact. Myosin activity is downregulated in the entire cell for $t < t_{\text{myo}} = 100\,\text{s}$. The pushing force vanishes at $t_{\text{imp}} = 85\,\text{s}$. The layout of the figure is analogous to Fig. S8. $ho F_{\text{cyt}}$, $ho F_{\Gamma}$, and $\rho_m$ distributions at times $t_{\text{imp}} = 85$, $t_{\text{myo}} = 100$, $t_3 = 110$, $t_4 = 290$, $t_5 = 310$, and $t_6 = 320\,\text{s}$ for two simulations with (a) $\bar{\eta}_{\text{myo}} = 12\,\text{pN\,\mu m}$ (case D1) and (b) $\bar{\eta}_{\text{myo}} = 30\,\text{pN\,\mu m}$ (case D2). Time evolution of (c) $\rho_F^F$, (d) $\rho_F^R$, (e) $P_F$, (f) $P_m$, (g) SF, and (h) $u_{\text{cell}}$ for three simulations with $\bar{\eta}_{\text{myo}} = 12$ (blue dotted line), 30 (red dashed line), and 60pN\,\mu m (black solid line). We stop the simulations with $\bar{\eta}_{\text{myo}} = 12$ and 30pN\,\mu m at $t = 600\,\text{s}$ rather than 300s so that the simulations can achieve a steady state. The vertical dotted lines in the graphs indicate the times $t_{\text{imp}}$, $t_{\text{myo}}$, $t_3$, $t_4$, $t_5$, and $t_6$. Strong levels of myosin inhibition impede the polarization of the cell.
Fig. S11 | Estimation of the threshold level for cell polarization induced by external stimuli. We computed $P^\Gamma$, $P^m$, and $SF$ at the time the external stimuli vanish, denoted as $P^\Gamma$, $P^m$, and $SF^*$, respectively, for cases C2, C3, C4, C5, D0, D1, D2, and the cases of cell polarization by mechanical impact with $u_{imp} = 0.01\mu m/s$ and $t_{imp} = 25s$ and $u_{imp} = 0.04\mu m/s$ (see Fig. S8), and $t_{imp} = 35s$ (see Fig. S9). (a) $P^\Gamma$; (b) $P^m$; and (c) $SF^*$ for the cases analyzed. Blue circles represent cells that achieve a steady polarized state. Red crosses represent cells that do not polarize. We define the region of no polarization (red shaded region) as the points $P^\Gamma < P^\Gamma_{NO,max}$, $P^m < P^m_{NO,max}$, and $SF > SF_{NO,min}$, respectively, where $P^\Gamma_{NO,max}$ is the maximum $P^\Gamma$, $P^m_{NO,max}$ the maximum $P^m$, and $SF_{NO,min}$ the minimum $SF^*$ of non-polarized cells (red crosses). Likewise, we define the region of cell polarization (blue shaded region) as the points $P^\Gamma > P^\Gamma_{POL,min}$, $P^m > P^m_{POL,min}$, and $SF < SF_{POL,max}$, respectively, where $P^\Gamma_{POL,min}$ is the minimum $P^\Gamma$, $P^m_{POL,min}$ the minimum $P^m$, and $SF_{POL,max}$ the maximum $SF^*$ of polarized cells (blue circles). If there is a threshold level for cell polarization, the blue and red shaded regions can not overlap. The hypothesis of a threshold level for cell polarization only holds for $P^\Gamma$. The threshold is between $P^\Gamma_{NO,max} = 0.1$ and $P^\Gamma_{POL, min} = 0.13$. 

threshold level for cell polarization

- Cell polarization
- No polarization

(a)

(b)

(c)