Motility of keratocyte cells: asymptotic and numerical analysis via a phase field model

Matthew Mizuhara

The Pennsylvania State University
Adviser: Leonid Berlyand
Other collaborators: Volodymyr Rybalko, Lei Zhang

June 11, 2015

Motile cells

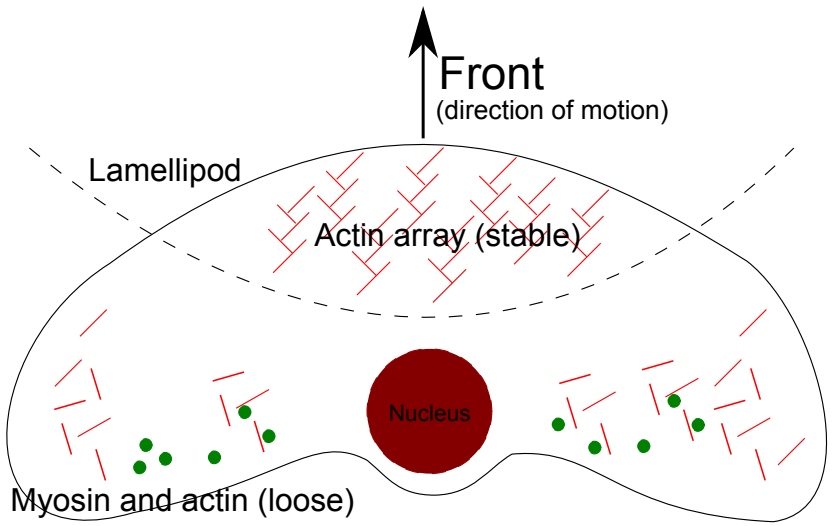
Keratocyte cells:[1] Crawl along a substrate; integral part of corneal wound healing (e.g., after LASIK or PRK surgery)

Contributing mechanisms - Several biological/physical processes interact to initiate and maintain movement:

(i). Actin polymerization

(ii). Surface tension of the membrane

(iii). Volume preservation



[1] Typical length scale: $100\mu m$

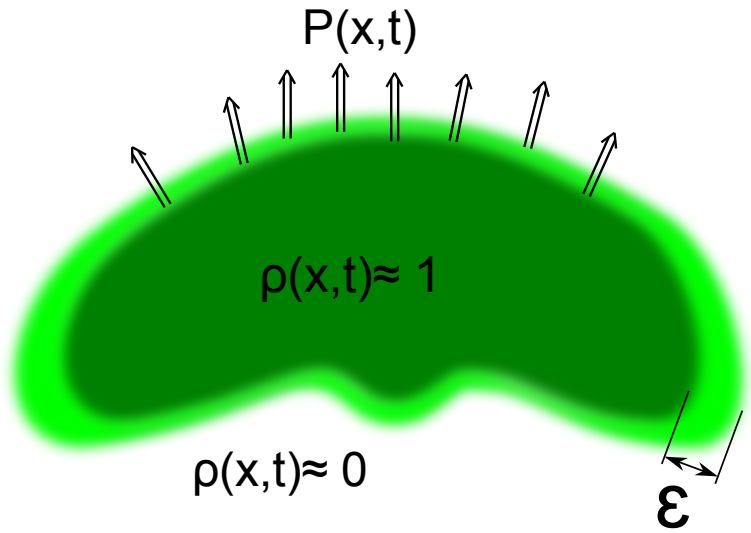
Phase-field model

Aronson, et al. (2011) proposed a *phase-field model*.
Phase-field variable, $\rho(x,t)$ - describes location of cell; takes value 1 inside and 0 outside; smooth ε-width transition at the boundary.
$\Omega \subset \mathbb{R}^2$ - bounded smooth domain (large substrate)

$$\begin{cases} \dfrac{\partial \rho_\varepsilon}{\partial t} = \Delta \rho_\varepsilon - \frac{1}{\varepsilon^2} W'(\rho_\varepsilon) - P_\varepsilon \cdot \nabla \rho_\varepsilon + \lambda_\varepsilon(t) \\ \dfrac{\partial P_\varepsilon}{\partial t} = \varepsilon \Delta P_\varepsilon - \frac{1}{\varepsilon} P_\varepsilon - \beta \nabla \rho_\varepsilon \end{cases} \quad \text{in } \Omega \qquad (1)$$

Vector field, $P(x,t)$ - polar orientation of the actin filaments

This model is accurate and simpler than previous models (no matching BC at interface)

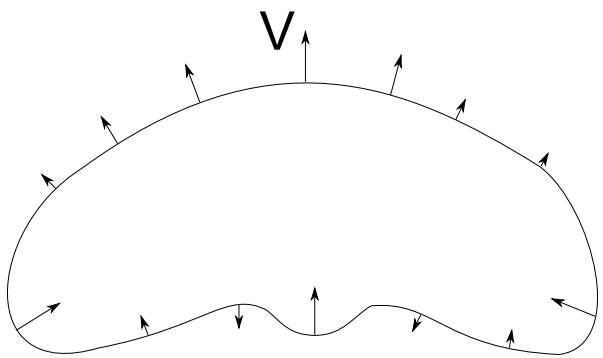


Sharp interface limit equation

In recent work of Berlyand, et al. (2014) sharp interface limit ($\varepsilon \to 0$) of (1) derived:

$$V = \kappa + \frac{\beta}{c_0}\Phi(V) - \frac{1}{|\Gamma(t)|}\int_{\Gamma(t)}\left(\kappa + \frac{\beta}{c_0}\Phi(V)\right) ds. \tag{2}$$

V - inward normal velocity, κ - curvature, β - physical parameter,
$c_0 = \sqrt{3/2}$ (depends only on double-well potential W)
Φ - known non-linear function
Equation (2) *is an equation for the evolution of a planar curve* Γ *(i.e., given* Γ*, one computes* κ *and* V*)*



Two regimes: Subcritical $\beta < \beta_{cr}$, supercritical $\beta > \beta_{cr}$

Analytical results

Theorem 1

Let $0 < \beta < \beta_{cr}$. Given $\Gamma_0 \in W^{1,\infty}$, there exists $T = T(\Gamma_0) > 0$ such that a family of curves $\Gamma(t) \in H^2$ exists for $t \in (0, T]$ satisfying the evolution equation (2) with $\Gamma(0) = \Gamma_0$.

Proof idea: Write the evolution equation as a PDE and prove existence for smooth (H^2) data. Passing to $W^{1,\infty}$ requires uniform estimates (independent of H^2 initial data) based on a maximum principle argument and classical results on Hölder continuity of solutions to quasilinear PDEs.

Remark: $W^{1,\infty}$ initial conditions $\Rightarrow$ corners

Theorem 2

Let $0 < \beta < \beta_{cr}$. There are no traveling wave solutions of (2) other than trivial stationary circles.

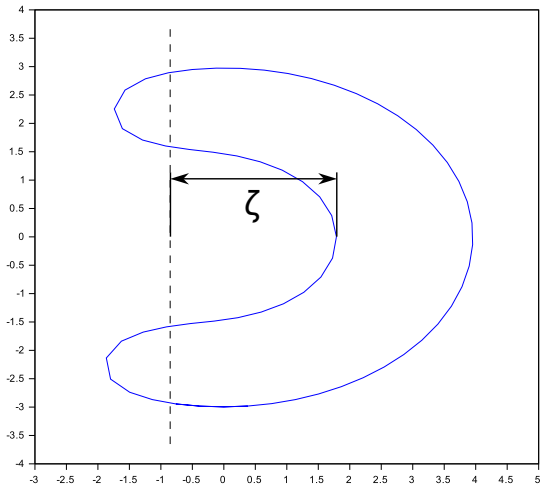
No moving traveling waves $\Rightarrow$ can we still observe *transient motion* of curves for subcritical β?

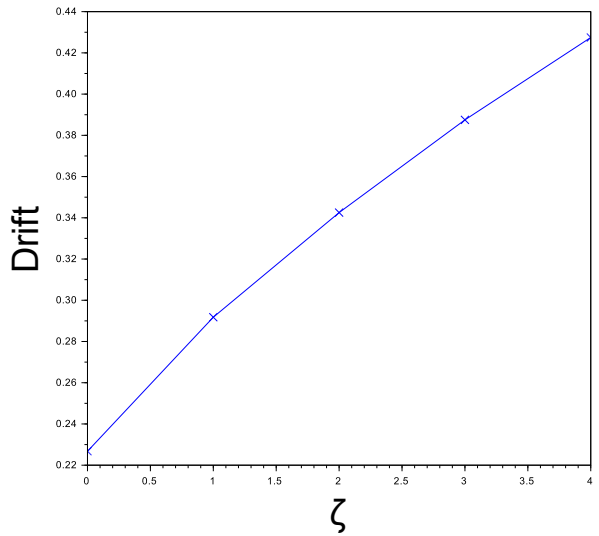
Numerical Results

Main difficulty: Discretizing curve gives rise to coupled system of non-linear equations $\Rightarrow$ hard to solve and preserve volume

Resolution: Develop *splitting scheme* which alternatively resolves 1) non-linearity 2) volume preservation

Experiment: Fix $\Gamma(0)$. Compute $\Gamma(t)$ propagating via (2) for $0 < \beta < \beta_{cr}$; compare to $\beta = 0$ (volume preserving curvature motion)





Observations: Curves propagating via (2) with $0 < \beta < \beta_{cr}$

(i) converge to a steady-state circle

(ii) transient motion; distance proportional to 1) β and 2) asymmetry ζ

Conclusions

- Aronson, et al. proposed a *phase-field model* to describe keratocyte cell motility
- Berlyand, et al. derived the *sharp interface limit equation* $\Rightarrow$ non-linear, non-local equation describing evolution of planar curves
- We investigate this sharp interface limit equation in the subcritical $\beta < \beta_{cr}$ regime:
  - **Analytical results:** existence of solutions, non-existence of traveling waves
  - **Numerical results:** convergence to steady-state circle, drift distance depending on physical parameter β and initial geometry

Thank you

Thank you for your attention!

This work is partially supported by the Department of Defense (DoD) through the National Defense Science & Engineering Graduate Fellowship (NDSEG) Program

Appendix

Splitting scheme

Given a curve Γ, discretize it by N points: $p_i = (x_i, y_i)$, $i = 1, \ldots, N$.
Naive discrete approximation:

$$V_i = \kappa_i + \beta\Phi(V_i) - \frac{1}{|\Gamma|}\sum_{j=1}^{N} \kappa_j + \beta\Phi(V_j)dS, \tag{3}$$

$\kappa_i, |\Gamma|, dS$ - discrete curvature, curve length, and line element, resp.
Non-linearity and **area preservation** $\Rightarrow$ level-set methods not available
Introduce the following *splitting scheme:*

1. Fix an auxiliary parameter $C \in \mathbb{R}$
2. Use an iteration method to solve

$$\tilde{V}_i = \kappa_i + \beta\Phi(\tilde{V}_i) - C \quad \textit{(local equation)} \tag{4}$$

3. Temporarily update Γ by the velocity profile $\tilde{V}$: $\tilde{p}_i = p_i + \tilde{V}_i \mathbf{n}_i$, $\mathbf{n}_i$ - discrete inward pointing normal. Compute change in area
4. Adjust auxiliary parameter C; repeat (2)-(3) until area is preserved

Parametrization of curve

Let $\tilde{\Gamma}_0$ be a C^4 smooth reference curve in a small neighborhood of Γ_0 with curvature κ_0 and let $\tilde{\Gamma}_0$ be parametrized by arc length parameter $\sigma \in I$. Then

$$\Gamma(\sigma, t) = \tilde{\Gamma}_0(\sigma) + u(\sigma, t)\nu(\sigma),$$

with ν the normal vector to $\tilde{\Gamma}_0$ and u solves

$$u_t - \frac{S}{1-u\kappa_0}\Phi\left(\frac{1-u\kappa_0}{S}u_t\right) = \frac{S}{1-u\kappa_0}\kappa(u) - \frac{S}{(1-u\kappa_0)L(u)}\left(\int_I \Phi\left(\frac{1-u\kappa_0}{S}u_t\right) S d\sigma + 2\pi\right),$$

where $S = \sqrt{u_\sigma^2 + (1-u\kappa_0)^2}$, and $L(u)$ is the total arc length of u.

Result on asymptotic equation

Consider the 1d model:

$$\begin{cases} \dfrac{\partial \rho_\varepsilon}{\partial t} &= \partial_x^2 \rho_\varepsilon - \dfrac{W'(\rho_\varepsilon)}{\varepsilon^2} + P_\varepsilon \partial_x \rho_\varepsilon + \dfrac{F(t)}{\varepsilon}, \quad x \in \mathbb{R}^1 \\ \dfrac{\partial P_\varepsilon}{\partial t} &= \varepsilon \partial_x^2 P_\varepsilon - \dfrac{1}{\varepsilon} P_\varepsilon + \beta \partial_x \rho_\varepsilon. \end{cases} \tag{5}$$

Note: Problem now exists on $\mathbb{R}$ and $F(t)$ is an arbitrary function.

Let θ_0 be the standing wave solution $\theta_0'' = W'(\theta_0)$.

Theorem. *Let ρ_ε and P_ε solve (5) on $[0, T]$ with "nice" initial data. For β sufficiently small, we have $\rho_\varepsilon = \theta_0\left(\frac{x - x_\varepsilon(t)}{\varepsilon}\right) + \varepsilon \rho_\varepsilon^{(1)}$ where $x_\varepsilon(t)$ is the interface location between 0 and 1 phase. Moreover $x_\varepsilon(t) \to x_0(t)$ with*

$$-c_0 \dot{x}_0(t) = \beta \Phi(\dot{x}_0(t)) + F(t), \Phi(-V) := \frac{1}{\beta} \int \psi |\theta_0'|^2 dy$$

Proof outline

Approximate ρ_ε and P_ε as

$$\rho_\varepsilon \approx \theta_0\left(\frac{x-x_\varepsilon(t)}{\varepsilon}\right)+\sum_{i=1}^{N}\varepsilon^i\theta_i\left(\frac{x-x_\varepsilon(t)}{\varepsilon},t\right), \quad P_\varepsilon \approx \sum_{i=0}^{N}\varepsilon^i\psi_i\left(\frac{x-x_\varepsilon(t)}{\varepsilon},t\right).$$

Expand $x_\varepsilon(t)$ in an ε-series; substitute and match powers of ε:

$$\begin{aligned}
\theta_0'' &= W'(\theta_0)\\
-\theta_1'' + W''(\theta_0)\theta_1 &= -V_0\theta_0' + \Psi_0\theta_0' + F(t)\\
-\theta_2'' + W''(\theta_0)\theta_2 &= -V_1\theta_0' - V_0\theta_1' - \frac{W'''(\theta_0)}{2}\theta_1^2 + \Psi_0\theta_1' + \Psi_1\theta_0'
\end{aligned}$$

and

$$\begin{aligned}
\Psi_0'' - V_0\Psi_0' - \Psi_0 &= -\beta\theta_0',\\
\Psi_1'' - V_0\Psi_1' - \Psi_1 &= -\beta\theta_1' + V_1\Psi_0' + \dot{\Psi}_0,\\
\Psi_2'' - V_0\Psi_2' - \Psi_2 &= -\beta\theta_2' + V_1\Psi_1' + V_2\Psi_0' + \dot{\Psi}_1.
\end{aligned}$$

Eigenvalue of linearized Allen-Cahn

Since $\theta_0'' = W'(\theta_0)$ then θ_0' is a zero eigenfunction of the operator

$$\mathcal{L}(f) = f'' - W''(\theta_0)f.$$

The Fredholm alternative implies that $g \in \operatorname{Im}\mathcal{L}$, if and only if $g \in (\operatorname{Ker}\mathcal{L})^{\perp}$. One can show $(\operatorname{Ker}\mathcal{L})^{\perp} = \operatorname{span}\{\theta_0'\}$. In other words, the equations for θ_i have solutions if and only if

$$\int(-V_0\theta_0' + \Psi_0\theta_0' + F(t))\theta_0' = 0,$$

$$\int(-V_1\theta_0' - V_0\theta_1' - \frac{W'''(\theta_0)}{2}\theta_1^2 + \Psi_0\theta_1' + \Psi_1\theta_0')\theta_0' = 0.$$

This gives the correct interface equation. The result follows by completing estimates on energy.

Classical result

Gage proved that a convex curve moving under area preserving curvature motion will converge to a circle exponentially.
Let $L = L(t)$ be the length of the curve at time t, A - area, k - curvature, N - normal vector

1. $L_t = -\int \langle V, kN \rangle ds$
2. $L_t \leq 0$ for all t (the curve is length shortening)
3. $\left(\frac{L^2}{A} - 4\pi\right) \leq Ce^{\frac{-2\pi}{A}t}$

Using the Bennesen inequality,

$$\left(\frac{L^2}{A} - 4\pi\right) \geq \frac{\pi^2}{A}(r_{out} - r_{in})^2$$

where r_{out} is the radius of the encompassing circle and r_{in} is the radius of the inscribed circle, the result follows.

It is important to note that he also must prove that convex curves remain convex and singularities do not develop.

Small β

We expect for small values of β that a similar result should hold. We are currently working to prove that (2) gives rise to length shortening flow.

If we apply similar methods as Gage, we compute the following

$$L_t = -\int k^2 + \frac{4\pi^2\nu^2}{L} + \beta \int \left(\frac{2\pi\nu}{L} - k\right)\Phi \leq -\theta + \beta\theta^{1/2}.$$

Here, $\theta = -\int k^2 + \frac{4\pi^2\nu^2}{L}$ is a measure of how "far" we are from a circle. If length is decreasing then $\theta \to 0$. This would imply that $\beta \to 0$. Thus, new techniques are required to complete this result.

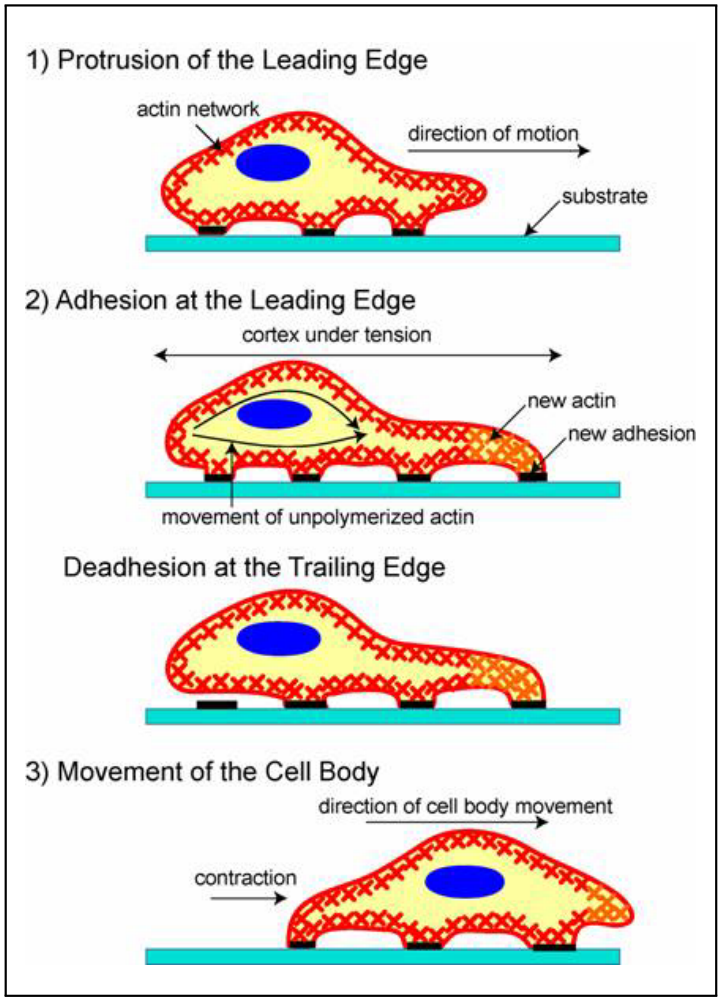
Cyclic view of motion

Orientation - choice of direction based on external cues (chemical gradients, mechanical deformations)

Protrusion - extension of cell membrane in direction of motion. Polymerization of actin filaments - backbone of protrusion

Adhesion - attachment of a cell to the extra-cellular matrix (ECM)

Retraction - contraction of actomyosin cytoskeleton



Plan of Computational Approach

Consider

$$V = \kappa + \frac{\beta}{c_0}\Phi(V) - \frac{1}{|\Gamma(t)|}\int_{\Gamma(t)}\left(\kappa + \frac{\beta}{c_0}\Phi(V)\right).$$

Let $f(V) := \frac{1}{c_0}\left(\Phi(V) - \frac{1}{|\Gamma|}\int_{\Gamma(t)}\Phi(V)\right)$.
Perturbation theory motivates iterative scheme:

$$V_0 = \kappa - \frac{1}{|\Gamma(t)|}\int_{\Gamma(t)}\kappa,$$

$$V_1 = V_0 + \beta f(V_0),$$

$$\vdots$$

$$V_n = V_0 + \beta f(V_{n-1}).$$

Convergence for Small β

Observe

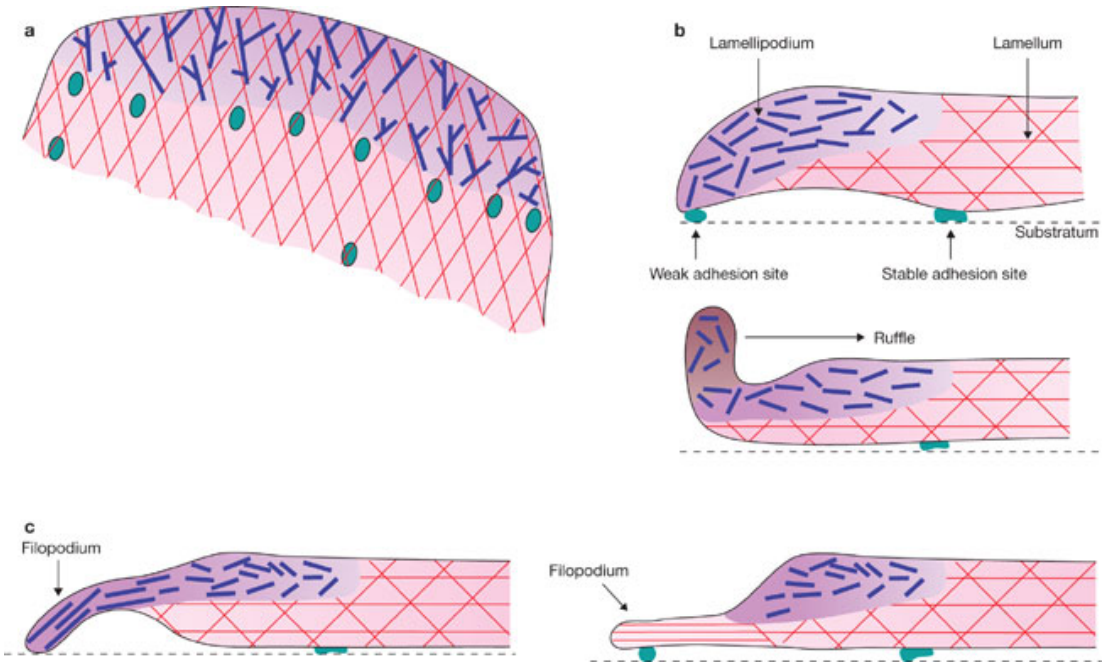
$$\begin{aligned}
\|V_n - V_{n-1}\|_{L^1} &= \|\beta(f(V_{n-1}) - f(V_{n-2}))\| \\
&= \left\| \frac{\beta}{c_0} \left(\Phi(V_{n-1}) - \Phi(V_{n-2}) - \frac{1}{|\Gamma|} \int_{\Gamma(t)} \Phi(V_{n-1}) - \Phi(V_{n-2}) \right) \right\| \\
&\leq \frac{2\beta}{c_0} \left(\int |\Phi(V_{n-1}) - \Phi(V_{n-2})| \right) \\
&\leq \frac{2\beta}{c_0} |\Phi'(V)|_{L^\infty} \|V_{n-1} - V_{n-2}\|_{L^1}
\end{aligned}$$

So for β small enough this iterative scheme converges.

Lamellipodia

The *lamellipod* is the leading edge of the cell which drives locomotion.

The *dendritic-nucleation hypothesis* predicts a precise pattern of *angular branching* of *actin filaments*.



The major conclusion from this model is that cell motion is dictated by an array of progressing filaments, not by individual filaments.

Contraction

Another key protein in cell motility is *myosin*, responsible for contraction.

Essentially, it uses energy from the ATP hydrolysis to enact a force on existing actin filaments.

Contraction

Another key protein in cell motility is *myosin*, responsible for contraction.

Essentially, it uses energy from the ATP hydrolysis to enact a force on existing actin filaments.

Actin disassembly leads to piling of "unorganized" actin filaments at the rear of the cell on which myosin can act, effectively pulling the rear of the cell forward.

Residuals

Modify the expressions

$$\rho_\varepsilon = \tilde{\rho}_\varepsilon + \varepsilon^\alpha u_\varepsilon, \text{ and } P_\varepsilon = \tilde{P}_\varepsilon + \varepsilon^\alpha Q_\varepsilon.$$

Plugging in these expressions into the original equation, we get

$$\frac{\partial u_\varepsilon}{\partial t} = \frac{u_\varepsilon''}{\varepsilon^2} - \frac{V_0 u_\varepsilon'}{\varepsilon} - \frac{W''(\theta_0)u_\varepsilon}{\varepsilon^2} - \frac{W'''(\theta_0)u_\varepsilon\theta_1}{\varepsilon} + \frac{\Psi_0 u_\varepsilon'}{\varepsilon} + \frac{Q_\varepsilon\theta_0'}{\varepsilon} + R_\varepsilon(t,y). \tag{6}$$

and

$$\frac{\partial Q_\varepsilon}{\partial t} = \frac{Q_\varepsilon''}{\varepsilon} - \frac{V_0 Q_\varepsilon'}{\varepsilon} - \frac{Q_\varepsilon}{\varepsilon} + \frac{\beta u_\varepsilon'}{\varepsilon} + \varepsilon^{N-\alpha} m_\varepsilon(t,y). \tag{7}$$

Our goal is to obtain bounds on u_ε and Q_ε.

Proof Outline

Proof idea:1. Write $u_\varepsilon(y,t) = \theta_0'(y)[\nu_\varepsilon(t,y) + \xi_\varepsilon(t)]$ where $\theta_0'\nu_\varepsilon$ is orthogonal to θ_0'.
This *factoring* of u_ε is useful because it allows us to use a modified **Poincaré inequality**.
2. Then, can obtain a bound

$$\frac{d}{2dt}\|u_\varepsilon\|_{L^2}^2 + \frac{1}{2\varepsilon^2}\int(\theta_0')^2(\nu_\varepsilon')^2 dy \le C\xi_\varepsilon^2 + \frac{1}{\varepsilon}\left[\int Q_\varepsilon(\theta_0')^2(\nu_\varepsilon+\xi_\varepsilon)dy - \int \psi_0'(\theta_0')^2 dy\xi_\varepsilon^2\right] + \int R_\varepsilon\theta_0'(\nu_\varepsilon+\xi_\varepsilon)dy.$$

3. To estimate u_ε, only must bound $\frac{1}{\varepsilon}$ terms. (Use that ψ_0 solves $\psi_0'' - V_0\psi_0' - \psi_0 = -\beta\theta_0'$.)
4. Bound the remaining terms to get the energy estimates:

Proof End

Define energies

$$\mathcal{E}_\varepsilon(t) = \int (\theta_0')^2 \nu_\varepsilon^2 dy + c_0 \xi_\varepsilon^2 + \frac{1}{c_p \beta^2 \varepsilon} \int B_\varepsilon^2 dy + \frac{1}{\varepsilon} \int D_\varepsilon^2 dy$$

$$\mathcal{D}_\varepsilon(t) = \frac{1}{8\varepsilon^2} \left\{ \int (\theta_0')^2 (\nu_\varepsilon')^2 dy + \int B_\varepsilon^2 dy + \int (B_\varepsilon')^2 dy + \int D_\varepsilon^2 dy + \int (D_\varepsilon')^2 dy \right\} + \left(\frac{\beta}{8} - c\beta^2 \right) \dot{\xi}_\varepsilon^2.$$

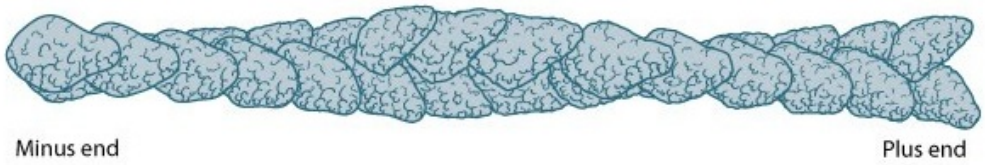
Then can show

$$\dot{\mathcal{E}}_\varepsilon + \frac{1}{2}\mathcal{D}_\varepsilon \leq c\mathcal{E}_\varepsilon + c\varepsilon \mathcal{E}_\varepsilon^{3/2} + c\varepsilon \mathcal{E}_\varepsilon^2 + c\varepsilon^6 \mathcal{E}_\varepsilon^3 + c(\varepsilon + \varepsilon^2 \mathcal{E}_\varepsilon^{1/2} + \varepsilon^4 \mathcal{E}_\varepsilon^{3/2}) \mathcal{D}_\varepsilon.$$

If $\mathcal{E}_\varepsilon(t) \leq c$ for $t \in [0, t*)$ then $\dot{\mathcal{E}}_\varepsilon \leq c\mathcal{E}_\varepsilon$. For small ε the result holds.

Actin protein

The cells on which we focus use *actin treadmilling* as a means of transportation (e.g. fish keratocytes).
Actin is a globular (spherical) protein which exists in two forms: G-actin and F-actin.



Actin Filament

G-actin is a monomer of actin ($\sim$ 5.4 nm) containing a polarity: one end is barbed and the other is pointed (analogy is an ice-cream cone).

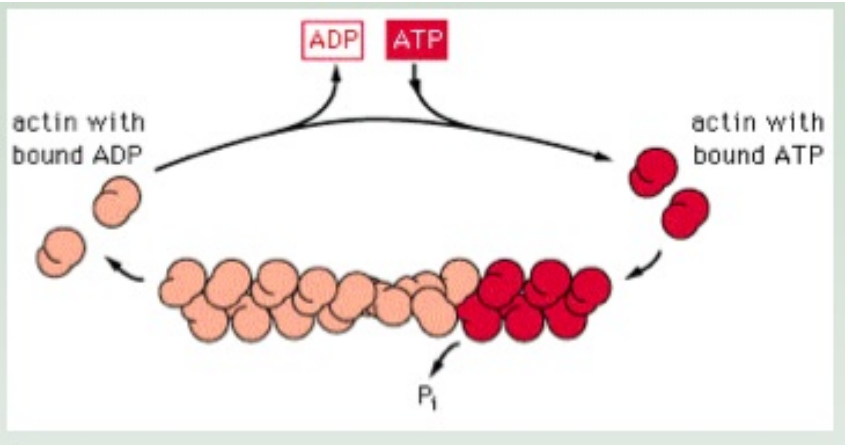
F-actin is a filament of actin formed by assembly of G-actin. The barbed end assembles and disassembles monomers two orders of magnitude faster than the pointed end (due to ATP binding).

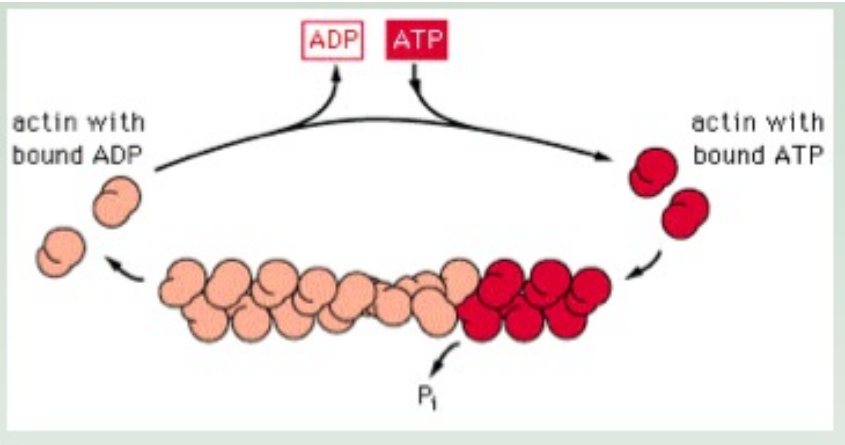
ATP capping and polarity

In the absence of *nucleotide hydrolysis*, the barbed and pointed ends of F-actin polymerize and depolymerize at the same rates.

However, G-actin binds to ATP at the barbed end and the following hydrolyzation occurs:

$$ATP \rightarrow ADP + P_i + \Delta E.$$





ADP-actin has the tendency to dissemble at the pointed end resulting in a net treadmill effect.

ADP-ATP re-charge occurs in the plasma membrane resulting in a basic mechanism for cell motility.

This cycle turns **chemical energy** into **mechanical energy**.