multilevel modeling of morphogenesis:
cell based morphodynamics
Cell-based morphodynamics

cell shape, cell movement and multicellular development

- **single cells**
  Mutual interaction cell shape and internal dynamics
  keratocyte chemotactic movement
  amoeboid movement (Dictyostelium).
- **“what about the horse part” “from single cells to multicellular organism”**
  through signaling, chemotaxis and differential adhesion

(from data intensive to behavior intensive models)
chemotaxis: modeling internal dynamics at different levels of detail

In CPM model chemotaxis can be implemented as ‘extend phyllopodia preferentially in direction of gradient’

How does the cell do this?

Interaction of small g proteins and actin network

Well studied in Keratocytes

importance of mutual feedback between cell shape and gene regulation

importance of biochemical detail ONLY apparent through this interaction
relevant small G protein interactions

**a** Small G-protein cross-talk

Cdc42 → Rac → Rho

- **Sidebranching** (Arp2/3)
- **Capping** protein
- **Contraction** protein

bistability in space due to fast diffusion inactive form
actin dynamics and cell wall dynamics
Table 1  Parameter estimates relevant to the small G-proteins and their interactions

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Meaning</th>
<th>Values</th>
<th>Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>$C^*$</td>
<td>typical level of active Cdc42</td>
<td>1</td>
<td>µM</td>
</tr>
<tr>
<td>$R^*$</td>
<td>typical level of active Rac</td>
<td>3</td>
<td>µM</td>
</tr>
<tr>
<td>$\rho^*$</td>
<td>typical level of active Rho</td>
<td>1.25</td>
<td>µM</td>
</tr>
<tr>
<td>$C_{tot}$</td>
<td>total level of Cdc42</td>
<td>2.4</td>
<td>µM</td>
</tr>
<tr>
<td>$R_{tot}$</td>
<td>total level of Rac</td>
<td>7.5</td>
<td>µM</td>
</tr>
<tr>
<td>$\rho_{tot}$</td>
<td>total level of Rho</td>
<td>3.1</td>
<td>µM</td>
</tr>
<tr>
<td>$I_C$</td>
<td>Cdc42 activation input rate</td>
<td>3.4</td>
<td>µM s$^{-1}$</td>
</tr>
<tr>
<td>$I_R$</td>
<td>Rac activation input rate</td>
<td>0.5</td>
<td>µM s$^{-1}$</td>
</tr>
<tr>
<td>$I_\rho$</td>
<td>Rho activation input rate</td>
<td>3.3</td>
<td>µM s$^{-1}$</td>
</tr>
<tr>
<td>$\beta_\rho$</td>
<td>Rho level for half-max inhibition of Cdc42</td>
<td>1.25</td>
<td>µM</td>
</tr>
<tr>
<td>$\beta_C$</td>
<td>Cdc42 level for half-max inhibition of Rho</td>
<td>1</td>
<td>µM</td>
</tr>
<tr>
<td>$n$</td>
<td>Hill coefficient of Cdc42-Rho mutual inhibition response</td>
<td>3</td>
<td>–</td>
</tr>
<tr>
<td>$\alpha_C$</td>
<td>Cdc42-dependent Rac activation rate</td>
<td>4.5</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$\alpha_R$</td>
<td>Rac-dependent Rho activation rate</td>
<td>0.3</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$d_C$, $d_R$, $d_\rho$</td>
<td>decay rates of activated small G-proteins</td>
<td>1</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$D_m$</td>
<td>diffusion coefficient of active small G-proteins</td>
<td>$1 \times 10^5$</td>
<td>nm$^2$ s$^{-1}$</td>
</tr>
<tr>
<td>$D_{mc}$</td>
<td>diffusion coefficient of inactive small G-proteins</td>
<td>$1 \times 10^7$</td>
<td>nm$^2$ s$^{-1}$</td>
</tr>
<tr>
<td>Parameter</td>
<td>Meaning</td>
<td>Values</td>
<td>Units</td>
</tr>
<tr>
<td>-----------</td>
<td>-------------------------------------------------------------------------</td>
<td>-------------</td>
<td>-----------</td>
</tr>
<tr>
<td>$A^*$</td>
<td>typical Arp2/3 concentration</td>
<td>2</td>
<td>$\mu$M</td>
</tr>
<tr>
<td>$F^*$</td>
<td>typical filament density</td>
<td>0.278</td>
<td>nm$^{-1}$</td>
</tr>
<tr>
<td>$B^*$</td>
<td>typical barbed end density</td>
<td>$1.7 \times 10^{-5}$</td>
<td>nm$^{-2}$</td>
</tr>
<tr>
<td>$P^*$</td>
<td>typical edge density of barbed ends</td>
<td>0.05</td>
<td>nm$^{-1}$</td>
</tr>
<tr>
<td>$\mu_C, \mu_R$</td>
<td>Cdc42 and Rac-dependent Arp2/3 activation</td>
<td>0.16</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$d_A$</td>
<td>activated Arp2/3 decay rate</td>
<td>0.1</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$D_A$</td>
<td>diffusion coefficient of Arp2/3</td>
<td>$1 \times 10^6$</td>
<td>nm$^2$s$^{-1}$</td>
</tr>
<tr>
<td>$n_0$</td>
<td>Arp2/3 nucleation rate</td>
<td>60</td>
<td>$\mu$M$\times$nm$s^{-1}$</td>
</tr>
<tr>
<td>$K_m$</td>
<td>saturation constant for Arp2/3 nucleation</td>
<td>2</td>
<td>$\mu$M</td>
</tr>
<tr>
<td>$l$</td>
<td>scale factor converting units of $F$ to concentration</td>
<td>255</td>
<td>$\mu$M$\times$nm</td>
</tr>
<tr>
<td>$k$</td>
<td>scale factor converting concentration to units of $B$</td>
<td>$1.06 \times 10^{-4}$</td>
<td>nm$^{-2}$$\mu$M</td>
</tr>
<tr>
<td>$v_0$</td>
<td>actin filament growth rate (free polymerization)</td>
<td>500</td>
<td>nm$s^{-1}$</td>
</tr>
<tr>
<td>$d_F$</td>
<td>actin filament turnover rate</td>
<td>0.03</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$\kappa_{max}$</td>
<td>barbed end capping rate</td>
<td>2.8</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$\kappa_{rac}$</td>
<td>max reduction of capping by Rac</td>
<td>2.1</td>
<td>s$^{-1}$</td>
</tr>
<tr>
<td>$K_R$</td>
<td>Rac level for half-max reduction of capping</td>
<td>3</td>
<td>$\mu$M</td>
</tr>
<tr>
<td>$r$</td>
<td>reduction of capping close to the edge</td>
<td>0.14</td>
<td>–</td>
</tr>
</tbody>
</table>
Shapes itself into a walking keratocyte and Walks! (and at the correct speed)
Can reorient itself: polarity and/vs rotation and/vs shape
feedback internal dynamics and cell shape
faster internal polarity change because of cell shape
changes (which are caused by internal polarity
change)
HOWEVER, internal dynamics more complex WHY?
Feedback through PIP network smooths out gradient
Feedback through PIP network causes faster adaptation

( HOWEVER: in round cell SLOWER reorientation to external signal!)
Feedback through PIP network enable resolving conflicting signals

polarization through noise instead of gradient
Feedback through PIP network maintains cell integrity when bumping in wall
Feedback through PIP network maintains cell integrity when bumping in obstacle
conclusions

Multilevel modeling makes things simpler!

Understanding of complexity at one level needs understanding of multilevel interactions

PIP network inhibits reorientation in static round cell

BUT speeds up response to cell shape AND reorientation in flexible cell AND Maintains cell integrity
Amoeboid cell movement, e.g. lymphocytes, Dictyostelium

two-dimensional excitable waves govern self-organized morphodynamics of amoeboid cells
Taniguchia,1, .. Sawai, PNAS 2013

visualised

modeled (excitable medium)
elastic cell

1. Inhibition of actin polymerization or PI3Kinase activity reduces the of wave nucleation and simplifies the wave patterns. (A) PIP3 (PHcrac-
Very simple model for Keratocyte
AND Amoeboid movement
duration of local, directional memory
(== actin network persistence)
Ioanna Niculescu and Rob de Boer Plos comp biol 2015

Simple extension of CPM model with periphery constraint
No representation of internal dynamics.
Only memory of previous movement
builds up from spontaneous
membrane fluctuations

2 parameters: strength $\lambda$
and duration $\text{Max}$
method
\[
\Delta \mathcal{H}_{\text{Act}}(u \rightarrow v) = \frac{\lambda_{\text{Act}}}{\text{Max}_{\text{Act}}}(\text{GM}_{\text{Act}}(u) - \text{GM}_{\text{Act}}(v))
\]

\[
\text{GM}_{\text{Act}}(u) = \sqrt[3]{15 \times 17 \times 15 \times 18 \times 20}
\]

\[
\text{GM}_{\text{Act}}(v) = \sqrt[4]{17 \times 16 \times 19 \times 11}
\]
Duration determines mode of movement

- limited duration
- long duration
- sensitive to chemotaxis
lymphocyte movement through skin
conclusions

Duration of local memory of protrusion sufficient to model difference between keratocyte and amoeboid movement

Keratocytes very robust (like extended model with PIP network)

Why?

Efficient Movement within tight tissue by small cell shape fluctuations
“How to compute an organism
Multilevel modeling of Morphogenesis
bridging levels of organization

Model premises

• Target morphogenesis ss (not only pattern formation)
• Cell basic unit (growth, division, movement, ...)
• Cell is NOT point, bead, homunculus
• Cells are deformable highly viscuous objects
• Genes act through cells 'with a dynamics of their own’

use CPM as simple but basically correct representation of a cell
Finding Sufficient Conditions for complex behavior using only (subset of) known processes allowing many (open set) different observations

**explicit 2-level model for implicit multilevel behavior**

**Dd morphodynamics:**

From single cells (amoebae) to multicellular 'individuals' with 'new' ways of sensing and metamorphosis to groups of those

Dictyostelium phylogeny

Early offshoot:
shares protein domains otherwise exclusive for plants, fungi, and animals
Lifecycle Dictyostelium

- Mature fruiting body
- Spores
- Free-living amoebae
- Aggregation induced by starvation
- Slug formation
- Cell division
- Germination
- Fruiting-body formation
- Migration

Images showing the lifecycle stages with time stamps (1 min 28 sec, 1 min 44 sec, 2 min 0 sec).
Goldbeter-Martel model of cAMP signaling

Models for cAMP signalling in Dictyostelium

\[ \frac{d\rho}{dt} = -f_1(\gamma) \rho + f_2(\gamma)(1 - \rho), \]
\[ \frac{d\beta}{dt} = s_1(\rho, \gamma) - \beta, \]
\[ \frac{d\gamma}{dt} = s_2(\beta - \gamma), \]

where

\( \rho \) = fraction of receptor in active state,
\( \beta = \frac{[cAMP]_{extracellular}}{[cAMP]_{intracellular} + \frac{K_R}{1 + \gamma}}, \)
\( \gamma = \frac{[cAMP]_{extracellular}}{[cAMP]_{intracellular} + \frac{K_R}{1 + \gamma}}, \)
\( t = k_1 \times \text{time}, \)

and

\[ f_1(\gamma) = \frac{1 + \kappa \gamma}{1 + \gamma}, \]
\[ f_2(\gamma) = \frac{L_1 + \kappa L_2 \gamma}{1 + \gamma}, \]
\[ \Phi(\rho, \gamma) = \frac{\lambda_1 + \gamma^2}{\lambda_2 + \gamma^2}, \]
\[ Y = \frac{\rho \gamma}{1 + \gamma}. \]

The parameters appearing in system (1)–(3) are explained and estimated in tables I and II; refer also to fig. 2.

Parameter set A in table II was used by Martiel and Goldbeter [16] to model autonomous oscillations of cAMP in stirred suspensions of Dictyostelium cells. The numerical solution of the
Martiel & Goldbeter, 1987 (cont’d)

\[
\begin{align*}
R & \xrightleftharpoons[k_{-1}]{k_1} D \\
R + P & \xrightleftharpoons[d_1]{a_1} RP \\
D + P & \xrightleftharpoons[d_2]{a_2} DP \\
RP & \xrightleftharpoons[k_{-2}]{k_2} DP \\
2RP + C & \xrightleftharpoons[d_3]{a_3} E \\
E + S & \xrightleftharpoons[d_4]{a_4} ES \xrightarrow[k_4]{k_4} E + P_i \\
C + S & \xrightleftharpoons[d_5]{a_5} CS \xrightarrow[k_5]{k_5} C + P_i \\
P_i & \xrightarrow[k_i]{k_i} \\
P_i & \xrightarrow[k_e]{k_e} P \\
P_i & \xrightarrow[v_i]{v_i} S \xrightarrow[k']{k'}
\end{align*}
\]
Parameter estimates of Goldbeter-model (Tyson 1989)

Table II
Model parameters.

<table>
<thead>
<tr>
<th>Name</th>
<th>Definition</th>
<th>Values used in calculations*</th>
<th>Set A</th>
<th>Set B</th>
<th>Set C</th>
<th>Set D</th>
<th>Set E</th>
</tr>
</thead>
<tbody>
<tr>
<td>( L_1 )</td>
<td>( k_{-1}/k_1 )</td>
<td>10</td>
<td>=</td>
<td>=</td>
<td>=</td>
<td>=</td>
<td>=</td>
</tr>
<tr>
<td>( L_2 )</td>
<td>( k_{-2}/k_2 )</td>
<td>0.005</td>
<td>0.005</td>
<td>0.005</td>
<td>0.005</td>
<td>0.005</td>
<td>0.005</td>
</tr>
<tr>
<td>( c )</td>
<td>( k_2/k_D )</td>
<td>18.5</td>
<td>=</td>
<td>=</td>
<td>=</td>
<td>=</td>
<td>=</td>
</tr>
<tr>
<td>( c )</td>
<td>( [ATP]/K_m )</td>
<td>3</td>
<td>=</td>
<td>=</td>
<td>=</td>
<td>=</td>
<td>=</td>
</tr>
<tr>
<td>( a )</td>
<td>( [ATP]/K_m )</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>45</td>
<td>=</td>
</tr>
<tr>
<td>( \lambda_1 )</td>
<td>( \left( \frac{V_m/K_m}{\alpha} \right) \left( \frac{K_m}{R_f} \right) )</td>
<td>( 10^{-4} )</td>
<td>( 10^{-3} )</td>
<td>( 10^{-3} )</td>
<td>( 10^{-5} )</td>
<td>6.7 ( \times 10^{-4} )</td>
<td>=</td>
</tr>
<tr>
<td>( \lambda_2 )</td>
<td>( \left( \frac{1 + \alpha K_m}{K_m} \right) \left( \frac{K_m}{R_f} \right) )</td>
<td>0.26</td>
<td>2.4</td>
<td>2.4</td>
<td>2.4</td>
<td>2.4</td>
<td>=</td>
</tr>
<tr>
<td>( r_1 )</td>
<td>( \left( \frac{V_m/K_m}{\alpha} \right) \left( \frac{1}{\alpha} \right) \left( \frac{a}{1 + a} \right) )</td>
<td>690</td>
<td>950</td>
<td>950</td>
<td>360</td>
<td>80</td>
<td>=</td>
</tr>
<tr>
<td>( r_2 )</td>
<td>( k_{-2}/k_2 )</td>
<td>0.033</td>
<td>0.05</td>
<td>0.05</td>
<td>0.13</td>
<td>0.13</td>
<td>0.39</td>
</tr>
<tr>
<td>( s )</td>
<td>( s_I/s_2 )</td>
<td>23</td>
<td>47</td>
<td>47</td>
<td>47</td>
<td>47</td>
<td>28</td>
</tr>
<tr>
<td>( \epsilon )</td>
<td>( k_2/k_1 )</td>
<td>0.014</td>
<td>0.019</td>
<td>0.019</td>
<td>0.005</td>
<td>0.005</td>
<td>0.005</td>
</tr>
<tr>
<td>Time-scale</td>
<td>( 1/k_1 )</td>
<td>0.0067</td>
<td>0.01</td>
<td>0.01</td>
<td>0.01</td>
<td>0.024</td>
<td>0.024</td>
</tr>
<tr>
<td>Space-scale</td>
<td>( (k_2 D)^{1/2}/k_1 )</td>
<td>10</td>
<td>8.2</td>
<td>4.5</td>
<td>8.2</td>
<td>4.1</td>
<td>=</td>
</tr>
</tbody>
</table>

*From Martiel and Goldbeter [16]. NA = not available, in which case units of the quantity are given with no numerical value.
**Units are the same as in column giving experimental range. When all four parameter set assume the same value of a parameter, the symbol = is used.
Set A: used by Martiel and Goldbeter to model cAMP oscillations in well-stirred cell suspensions.
Set B: used by Martiel and Goldbeter to model cAMP signal-relaying in well-stirred cell suspensions.
Set C: used in this paper to calculate spiral waves in the full three-component model.
Sets D and E: used in this paper to calculate spiral waves in the model two-component model.

---

\[ \text{spiral} \]
simplify dynamics - add cells:

Dd life cycle by excitable medium and differential adhesion

excitable medium + differential adhesion
Lifecycle of Dd by chemotaxis and adhesion

aggregation

streams

orientation

culmination
Dd morphodynamics:
multiple causes and multiple effects

<table>
<thead>
<tr>
<th>Aggregation</th>
<th>streams if wave propagation dep on density faster movement in streams</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mount(slug) slug</td>
<td>cell sorting by differential adhesion AND chemotaxis slug shape attractor of energy minimization vs inward movement (wave shape) taxis (thermo- photo-taxis) via NH3 effect on excitability) slug shape and wave shape bi-directional mutant direction of movement vs momentum</td>
</tr>
<tr>
<td>culmination</td>
<td>needs dynamic cell differentiation downward movement of stalk cells caused by peristalsis caused by upward movement of spore cells pressure waves and wave shape self-correcting and self-terminating</td>
</tr>
</tbody>
</table>
stream formation requires density dependent speed of cAMP wave propagation (i.e. fast internal cAMP dynamics)

Why streams?

Fig. 4. Plot of velocity (each point averaged over 10 simulations) against the cell–medium bond energy for prespore amoebae. Given that the amoebae adhere to each other the group will always move faster than a single amoeba. Parameters are as described in the legend to Fig. 1. □, Group of amoebae; ●, single amoeba.
Cell Sorting much faster in *moving slug* than in *fixed mount*

**WHY?**

Prestalk cells (Yellow) stronger adhesion
Prespore cells (Green)

\[ J_{yy} < J_{gg} < J_{yg} \]

Same chemotactic response

Savill and Hogeweg 1997
EQUAL chemotaxis speeds up sorting
(moving slug instead of fixed mount also faster sorting
(cf Kafer, “go against the flow” (binf4))
Movement Dd slugs: measured bead displacement and calculated force fields
cf. Rieu, Baranth, Maeda and Sawada 2005

displacement field

outward directed forces!

stress field
similar forces in model Dd slugs?

Note:
forces are (emergent) observables instead of model ingredients!

Can be measured (like in experiments)
cf From energy to cellular forces in the Cellular Potts Model: An algorithmic approach EG Rens, L Edelstein-Keshet - PLoS Computational Biology, 2019

Perpendicular forces expected because:
- wave shape (most concave in middle of slug)
- sideward push because of pressure gradient
conclusions

- Using simplifications which allows multilevel modeling we “can go for the horse part”
- Development as trajectory of dynamical system model minimizes regulation within cells
- Assumption of CPM seem very suitable to describe biological cells
- Relatively few parameters need to be specified; large set of 'new' observables
- Treating forces as observables rather than model assumption allow close comparison with experimental measurements

BUT WHAT ABOUT THE GENES?
Evolutionary “testing” of the model

who wants to be a stalk?, cf Queller
how to come become another dictyosteloid?

*multiple levels needed to understand complexity*
Who want to become a stalk?

Evolution of cooperation and why cheaters do not take over
single gene greenbeard effect

Who depends on phase in cell cycle
Cell adhesion gene csA binds to csA
on agar csA knockouts become spores because wildtype cells
have more adhesion → go to front - become stalk
BUT
in soil csA knockouts are left behind during aggreg. phase

conclusion: who wants to become a stalk

Simple optimality reasoning often flawed
Important role of non-inheritable behaviour
  stochasticity
  environmental heterogeneity
  selforganization
from Dictyostelium to other discyosteliids
Polysphondinum

Polysphondylium violaceum

A.R. Swanson, A Guide to the Common Dictyostelid Slime Molds of Great Smoky Mountains National Park

continuous redifferentiation prestalk-stalk sidebranches (polyshondininium)