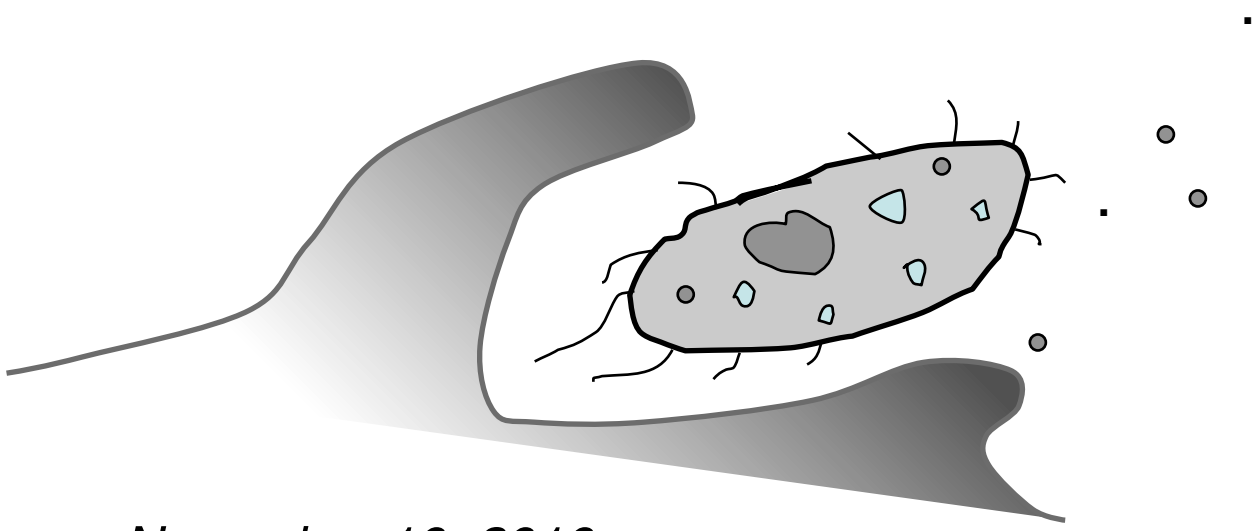
How one cell eats another

Physical principles in phagocytic uptake



IoSSB Symposium *November 10, 2010*

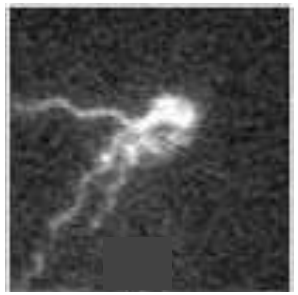
Robert Endres
Imperial College London

http://www3.imperial.ac.uk/biologicalphysics
r.endres@imperial.ac.uk

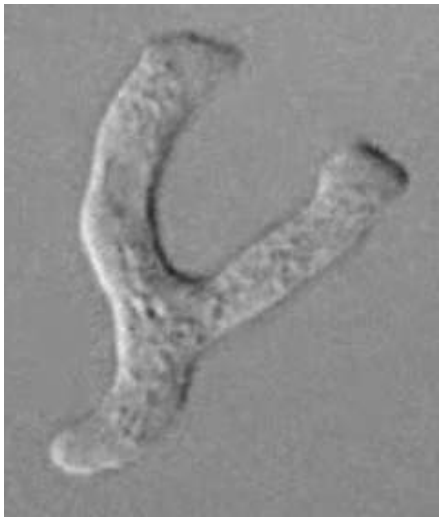
Where physics meets biology….

Chemosensing and motility

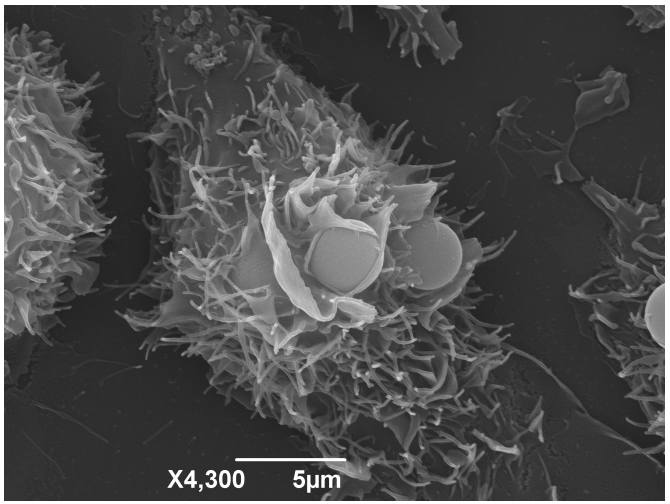
bacteria



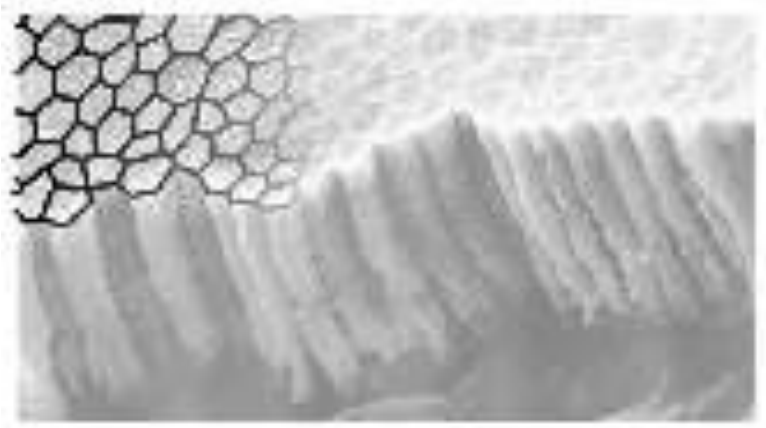
amoeba



Eating, uptake, and destruction



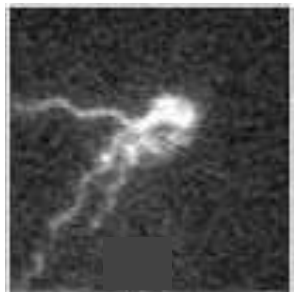
Cell packing



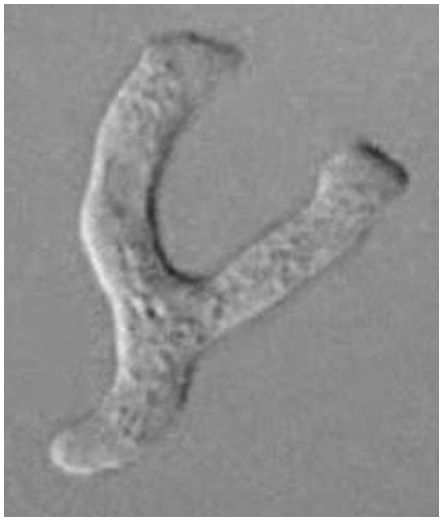
Where physics meets biology….

Chemosensing and motility

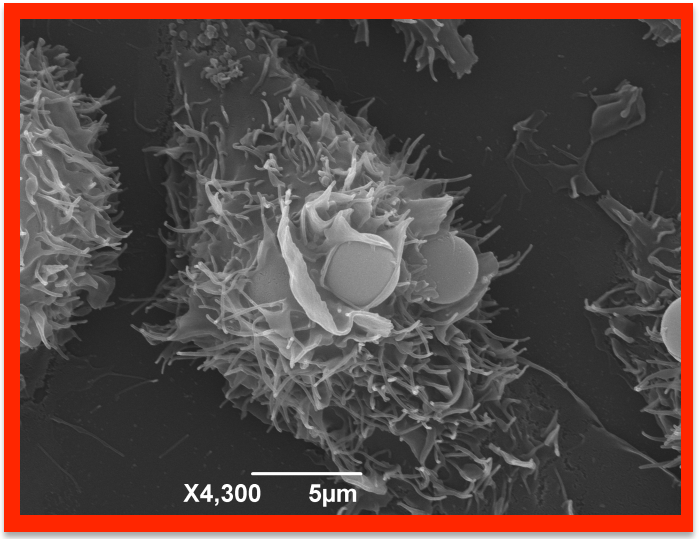
bacteria



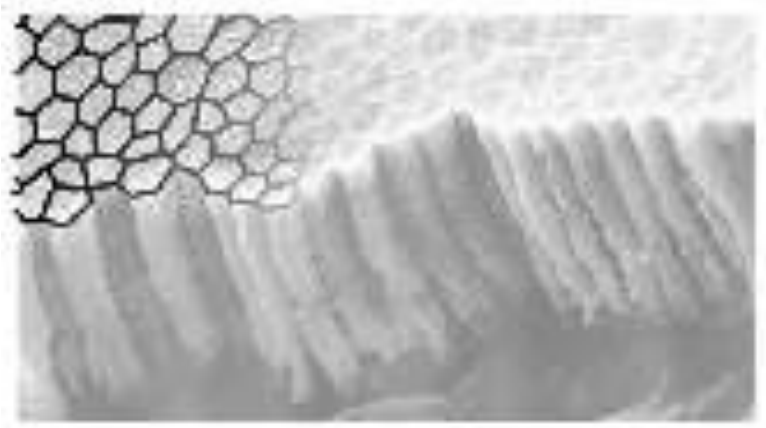
amoeba



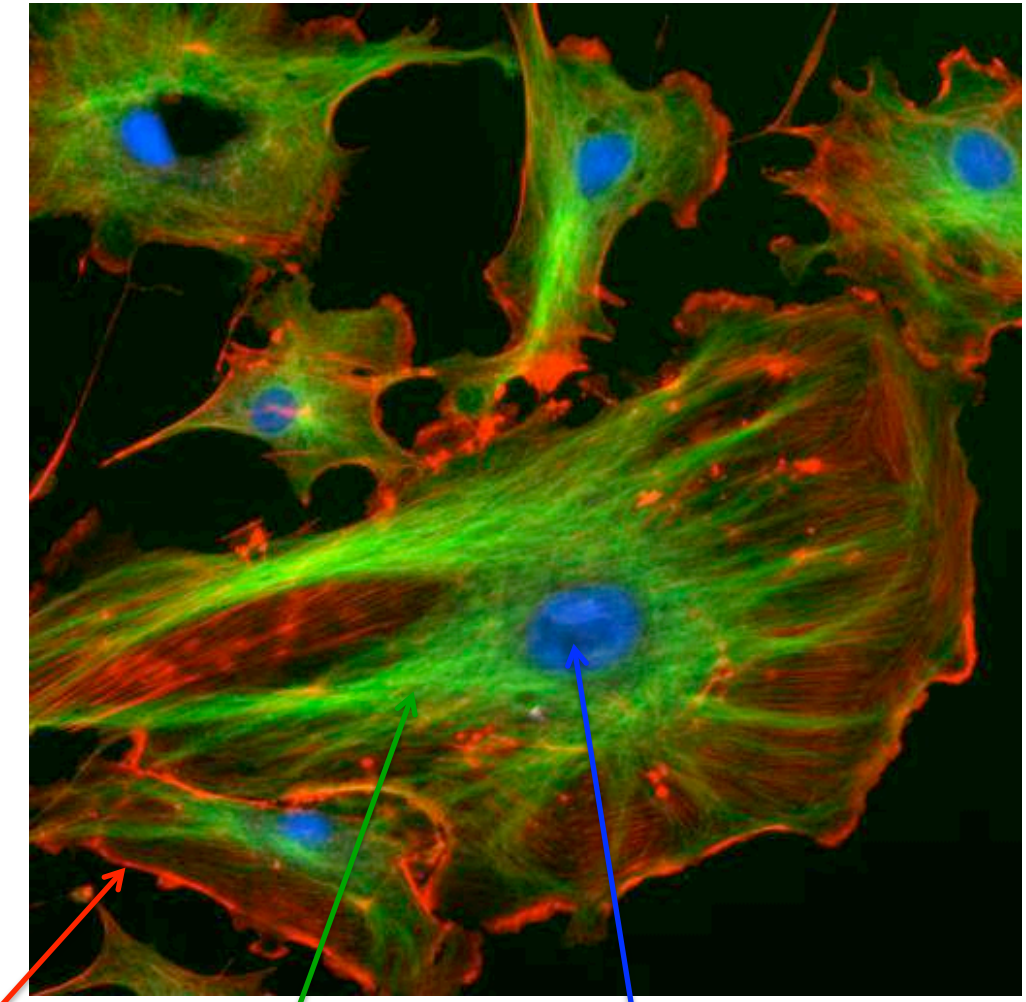
Eating, uptake, and destruction



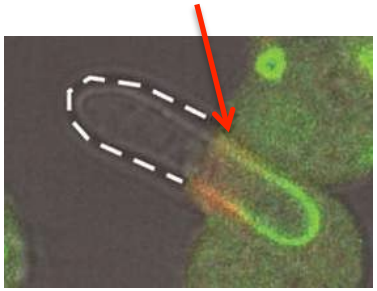
Cell packing

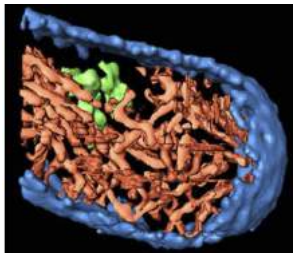


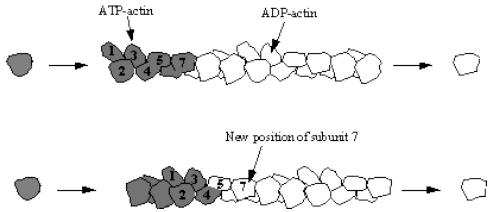
Cell cytoskeleton – very dynamic and versitile for cell-shape changes in phagocytosis, migration, adhesion, and packing



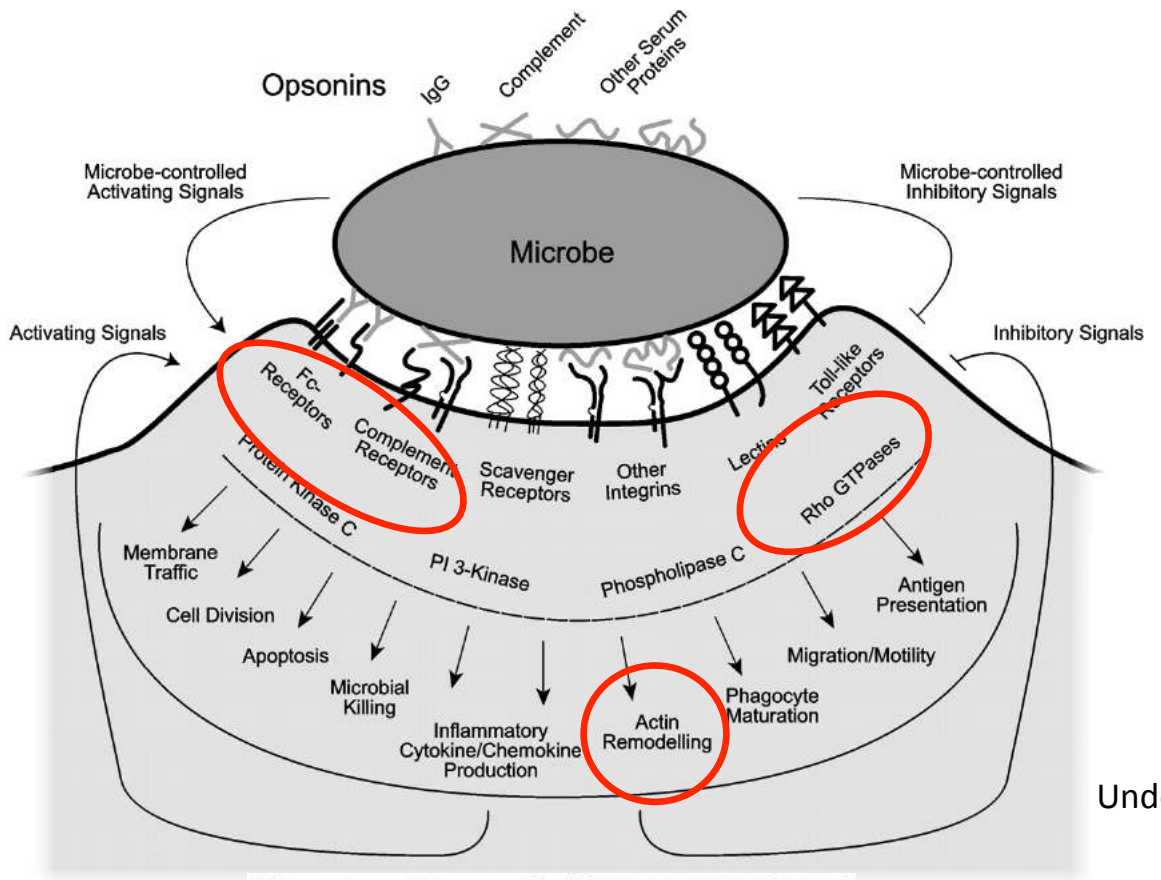
actin





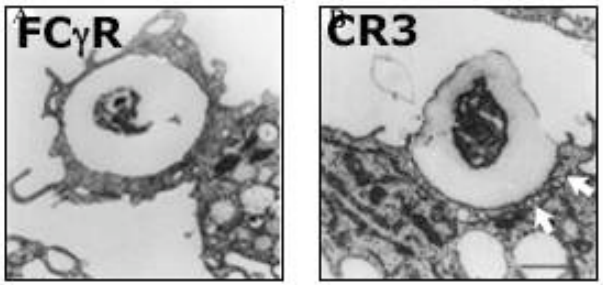


Daunting signalling complexity in phagocytosis



Underhill & Ozinsky (2002)

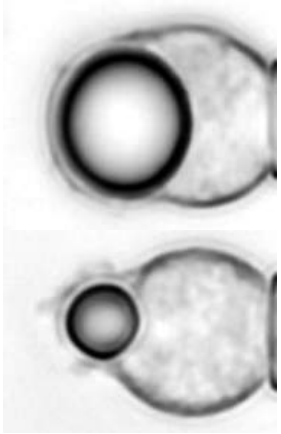
about 140 different molecular species are involved



Allen & Aderem (1996)

Geometric and biophysical aspects of phagocytosis

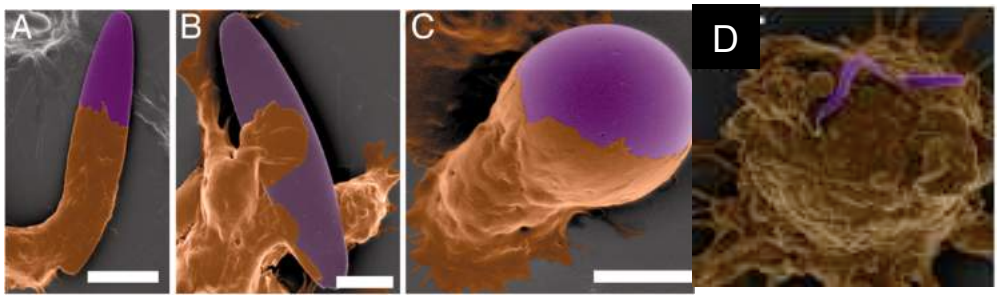
1. Particle-size independence



Herant et al. (2006)

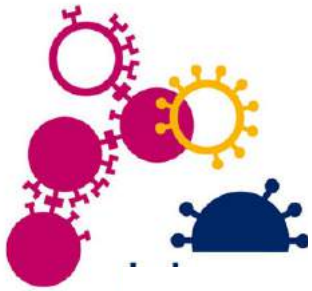
2. Shape dependence

3. Elastic properties



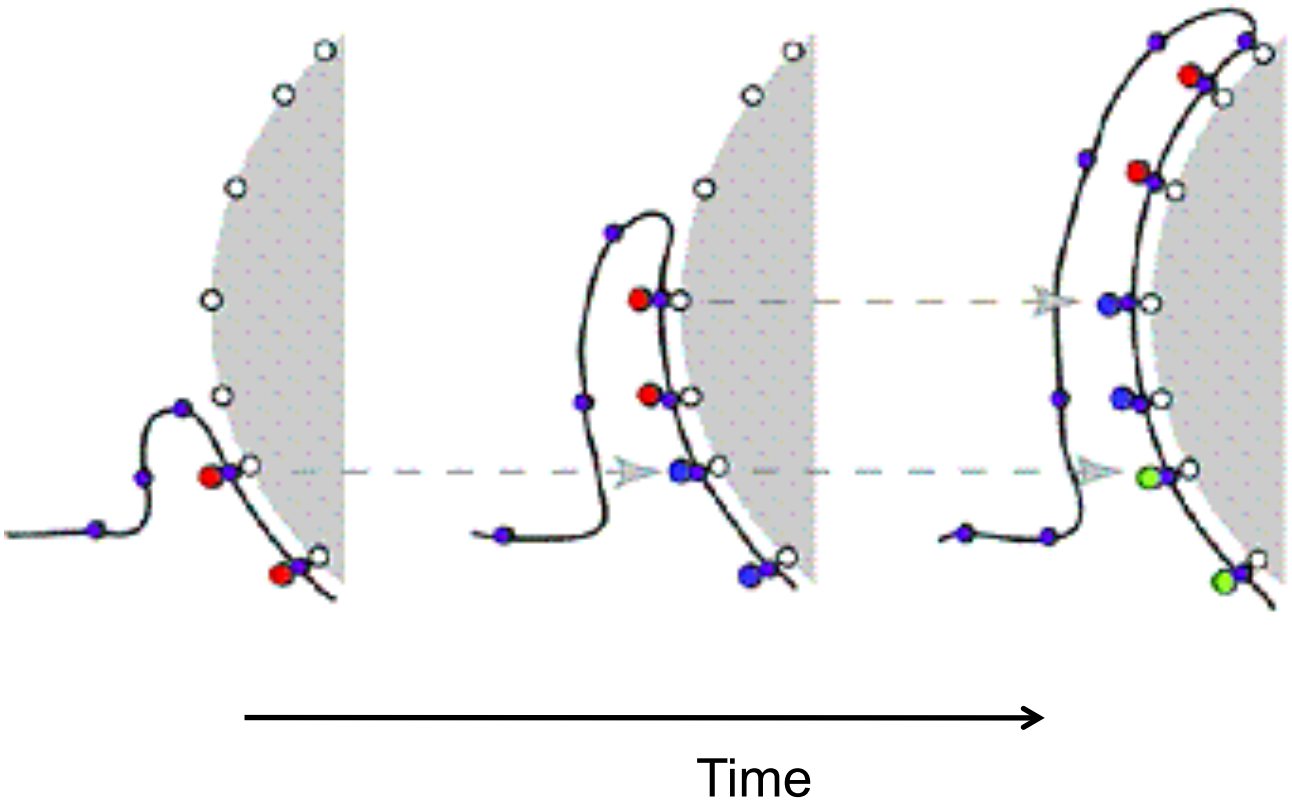
Champion et al. (2006,2009)

4. Ligand density



Conceptual Zipper mechanism

for explaining dependence on ligand density



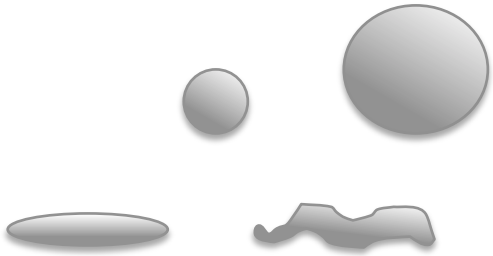
Griffin et al. (1975), Swanson (2008)

Can the Zipper mechanism explain?

Geometric requirements of particle?

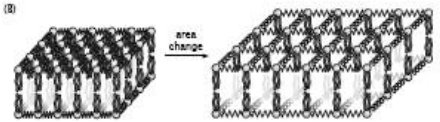
Size independence

Shape dependence



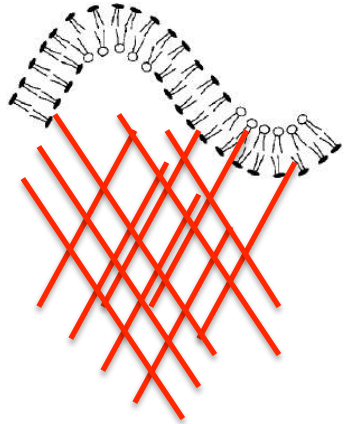
Energetic requirements of cell?

Ligand-receptor binding



Surface tension

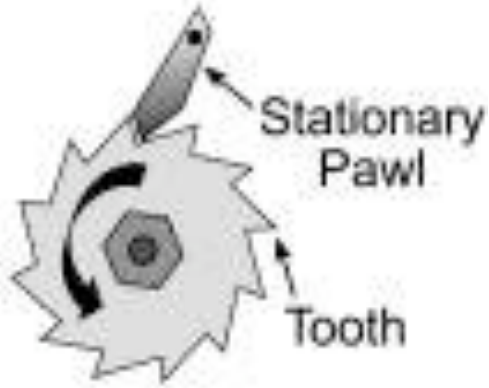
Membrane bending



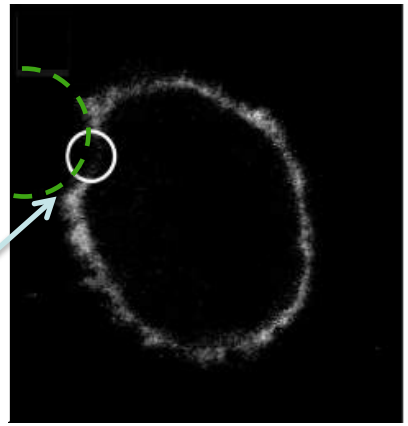
Remodelling of cytoskeleton

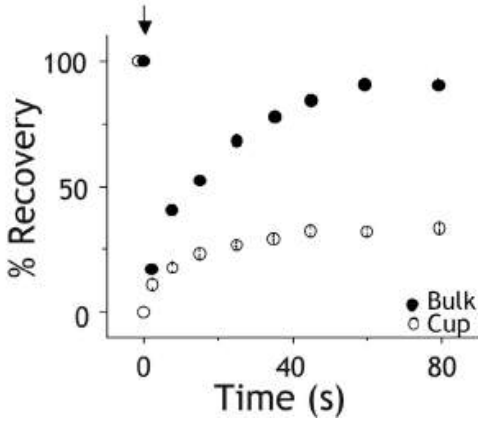
Mechanistic idea for modelling Zipper mechanism

Ratchet



1. **Unidirectional** engulfment, even **without pauses**
2. **Immobilization** of proteins and lipids in cups from FRAP

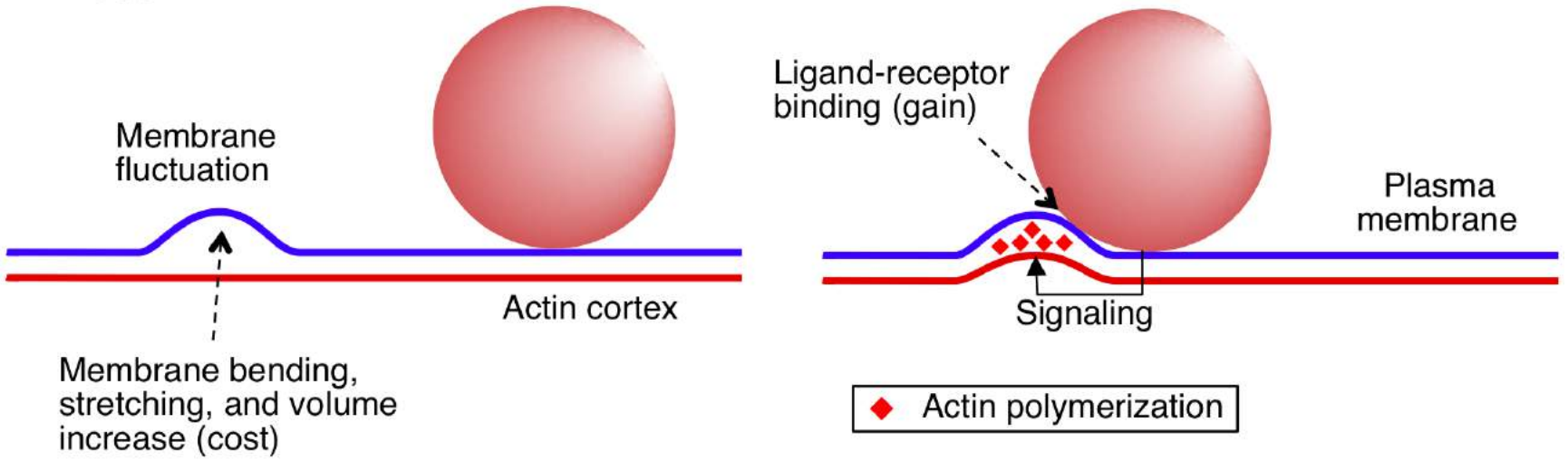




Corbett-Nelson et al. (2006)

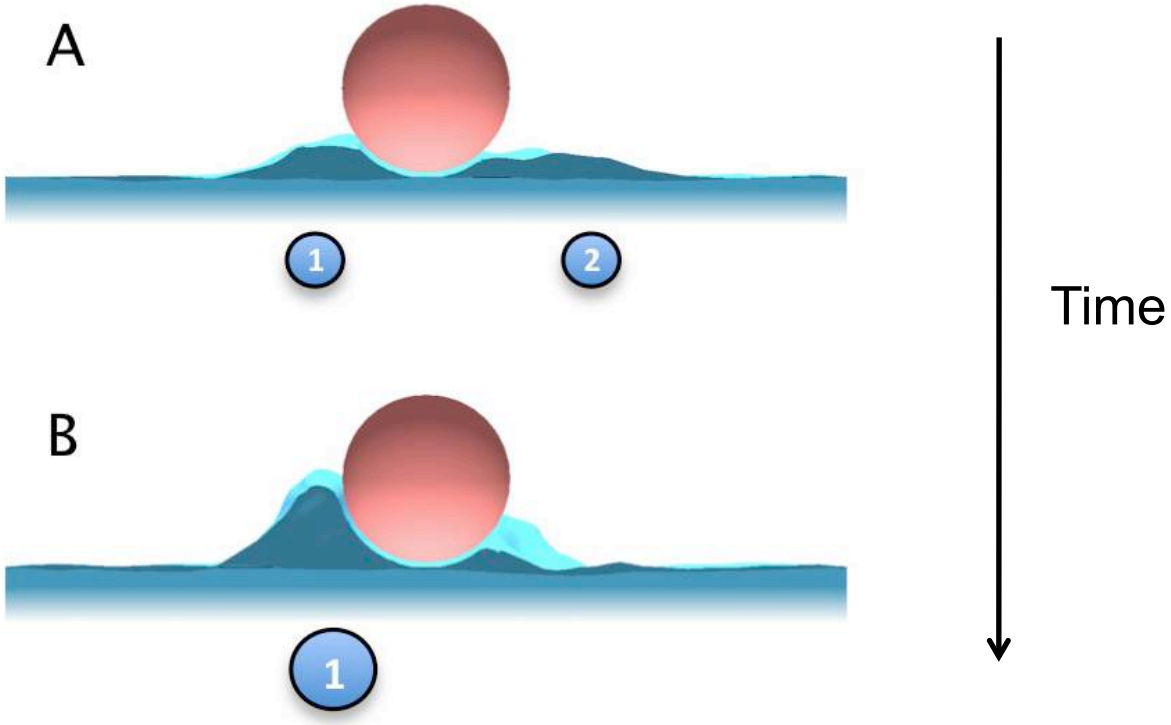
3. Ratchets used before in modelling actin-driven motility

Implementation of zipper mechanism



Ligand-receptor binding induces actin polymerization, making membrane deformation effectively irreversible → **ratchet**.

Ratchet model in action



① Membrane deformation near particle is stabilized.
② Membrane deformation far from particle is retracted later.

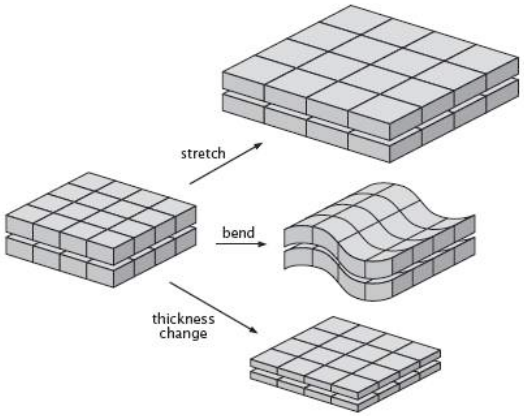
Model for cell membrane

Cell-membrane energy:

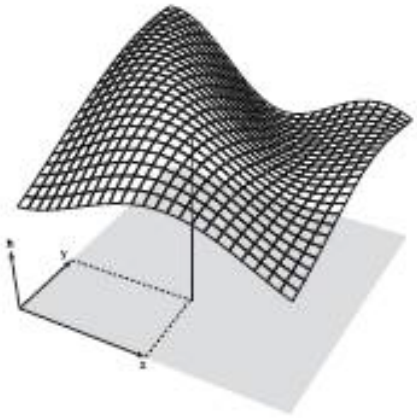
bending — membrane stretching — vesicle swelling — Ligand-receptor binding

$$E = \frac{\kappa_B}{2}\oint (\underbrace{C_1 + C_2}_{=2\overline{C}} - C_0)^2 dA + \Sigma A + PV + E_{LR}$$

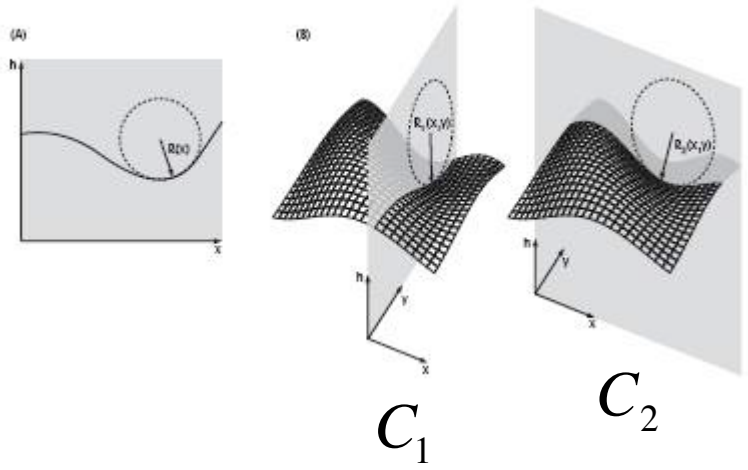
membrane deformations



height profile $h(x,y)$



membrane curvature



C_1 C_2

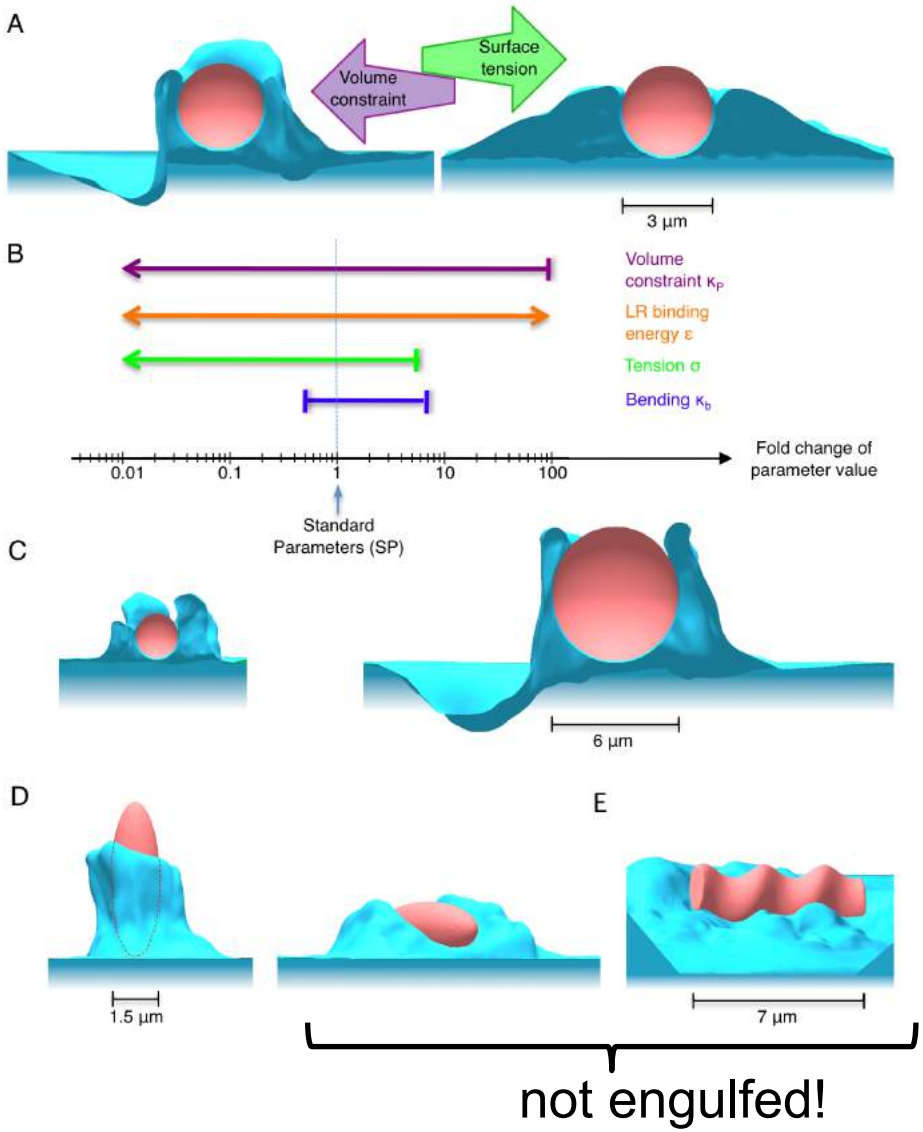
Engulfment for wide range of parameters

Cup shape

Parameter range

Particle size

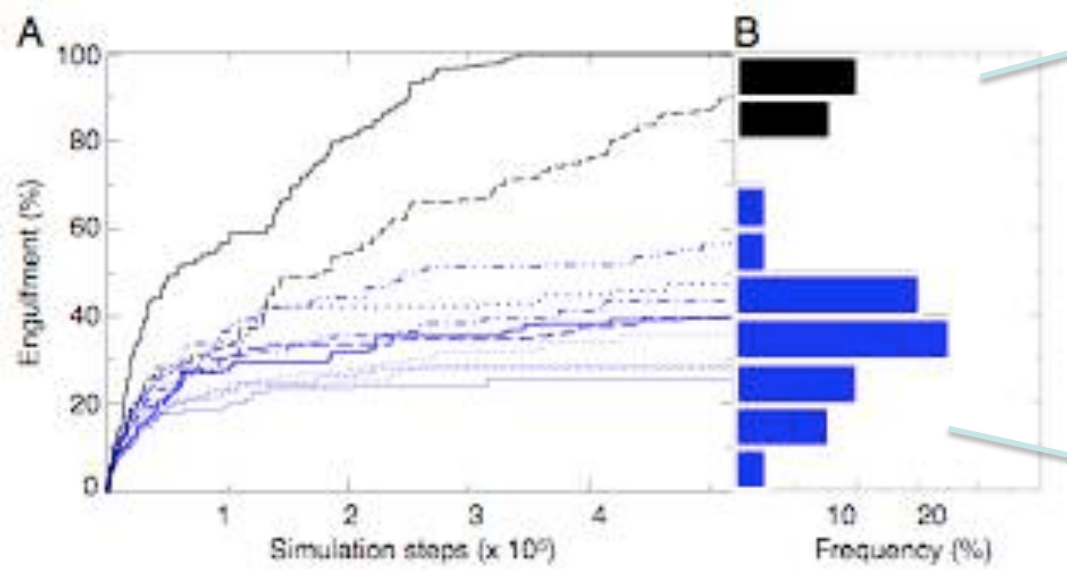
Particle shape
and orientation

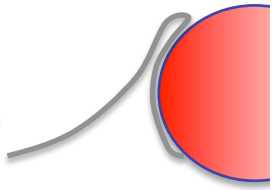


Bistability: a means of decision making?

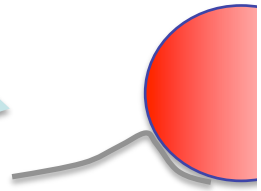
Simulation

Experiment





Mechanical bottleneck at half-engulfment



Trajectories for different values of membrane-surface tension

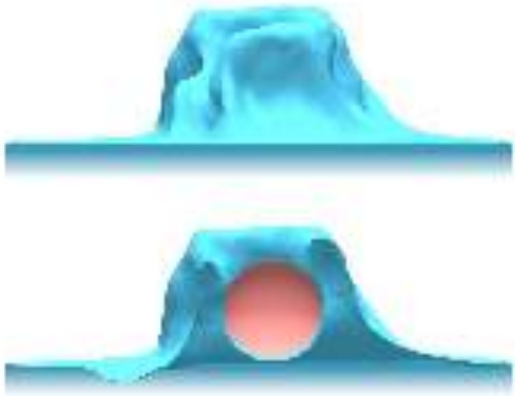
Bimodal distribution from experiments with WT-FcR after 10 min.

Van Zon *et el*. (2009), Zhang *et al*. (2010)

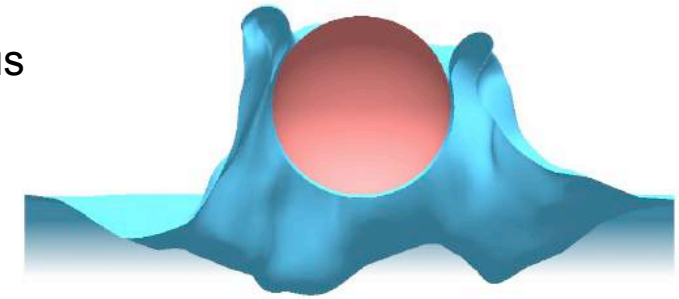
Energetic requirements:

Active zipper

Small,
1.5 μm-radius
particle



Large,
3 μm-radius
particle



→ Active zipper easily engulfs small and large particles

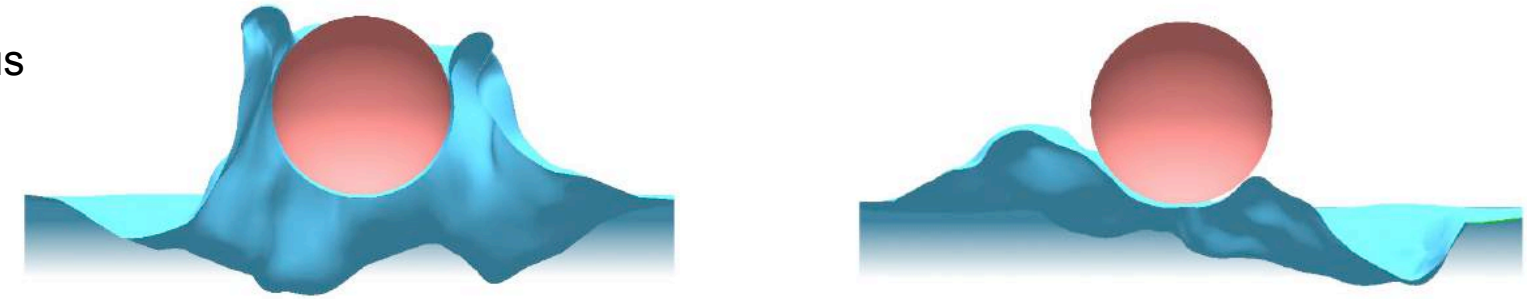
Energetic requirements:

Active zipper

Passive zipper

Small,
1.5 μm-radius
particle

Large,
3 μm-radius
particle

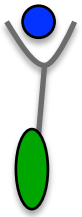


→ Active zipper easily engulfs small and large particles

→ Passive zipper ONLY engulfs small particles - slowly with highly variable cups

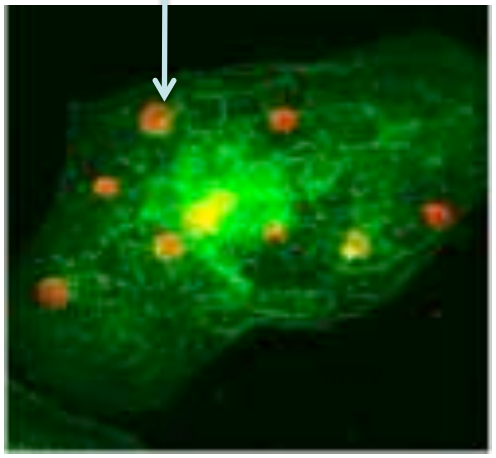
3D imaging with confocal microscopy

Fcγ receptor

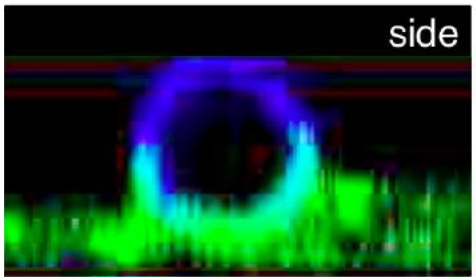


GFP

Latex particle



COS-7 cell



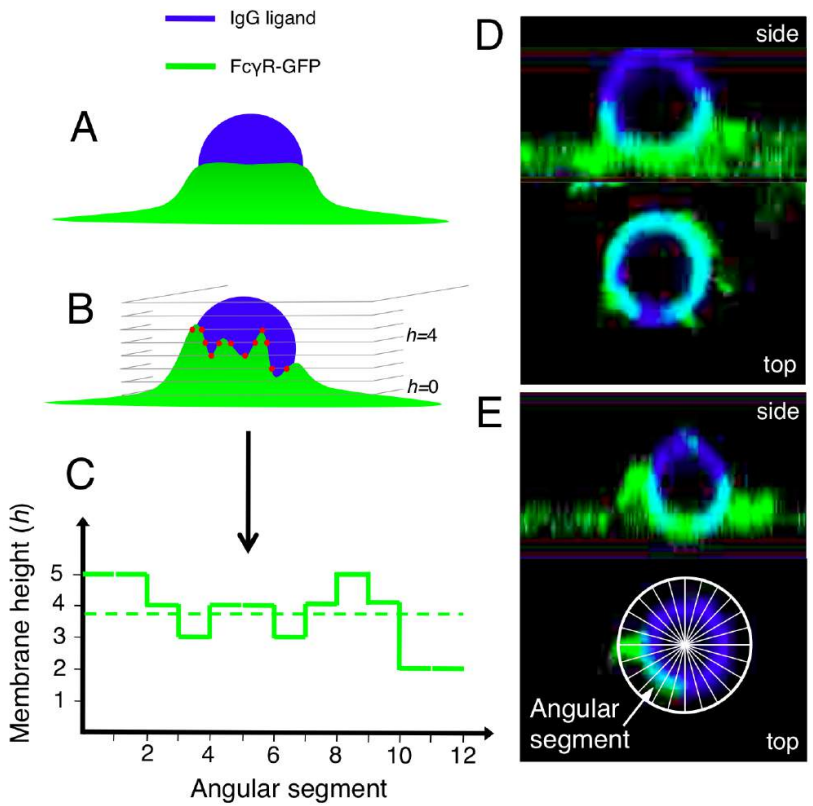
Phagocytic cup

Experimental implementation of two zippers

Active zipper: cells expressing wild-type Fcγ receptor

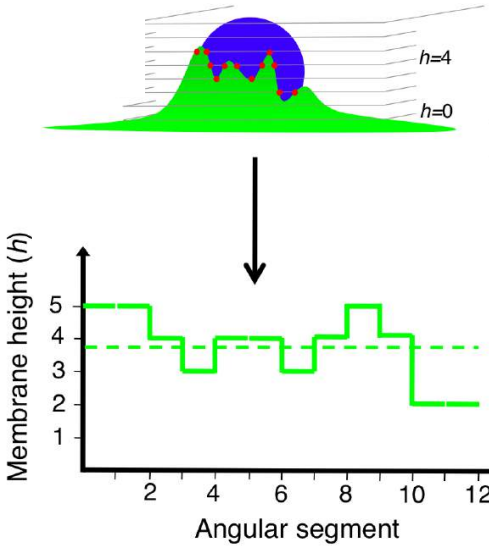
Passive zippers
- cells expressing signalling-dead mutant receptor
- cells transfected with cytochalasin D

Cup height and variability

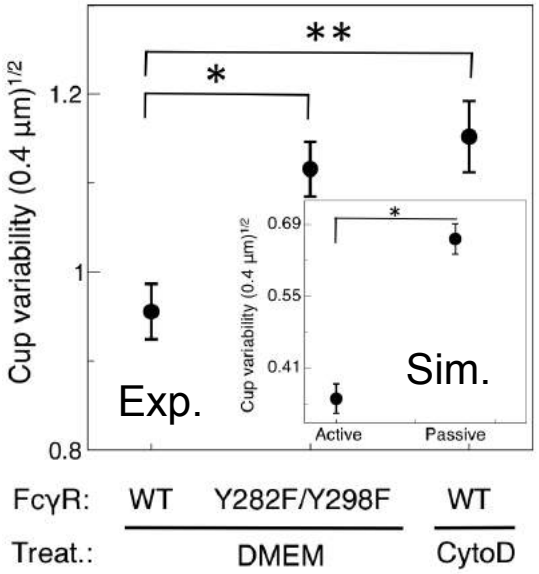


Variability of cup shape

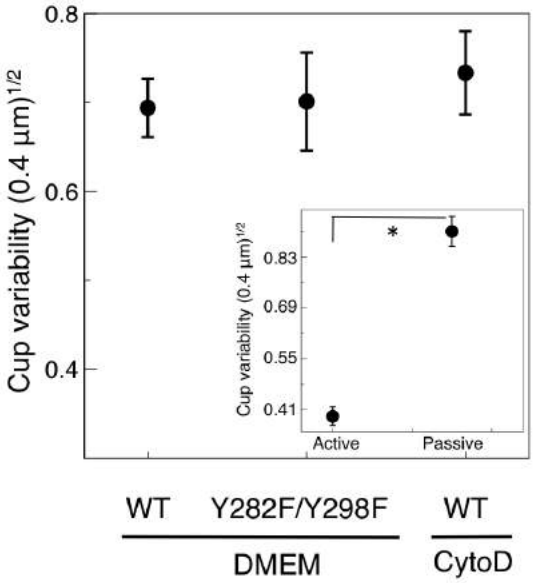
Variability



20-40% Engulfment



40-60% Engulfment

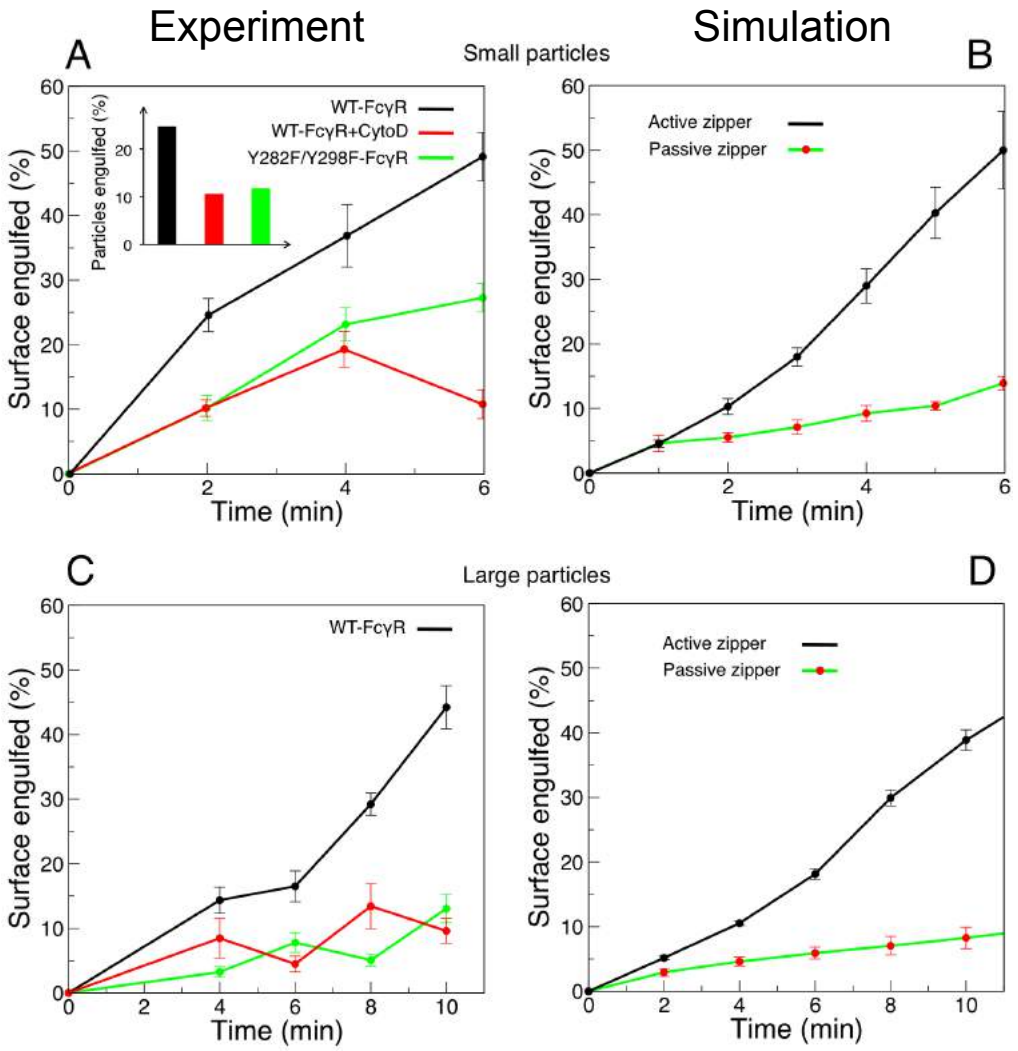


Passive zipper produces more variable cup shapes than active zipper initially.

Engulfment time

Small particle

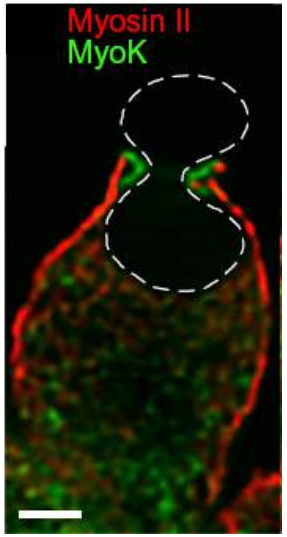
Large particle

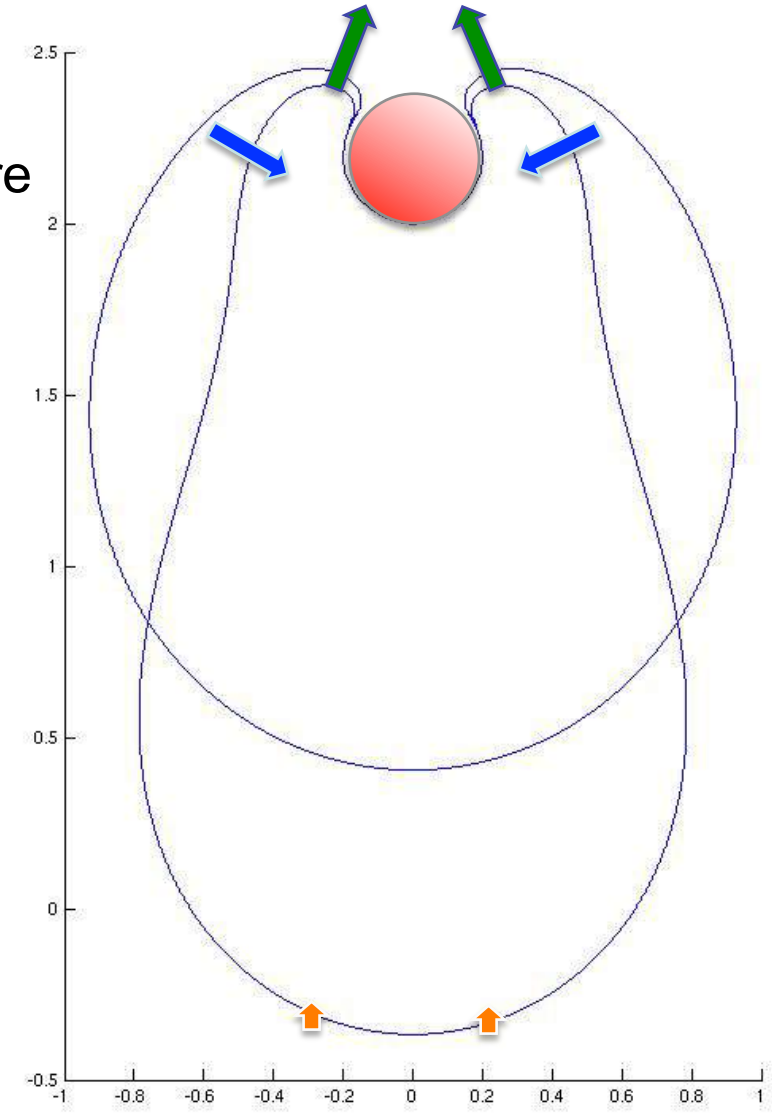


Active zipper engulfs significantly faster than passive zipper

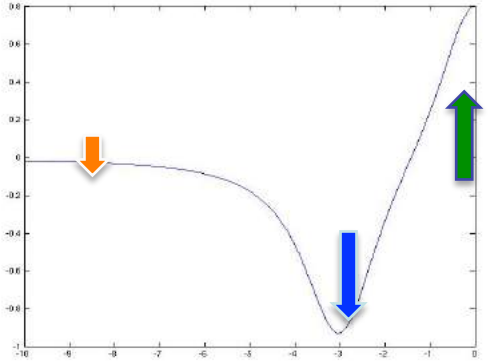
Outlook: role of contraction in cup closure and shape

Cell shape for given cell-pressure profile





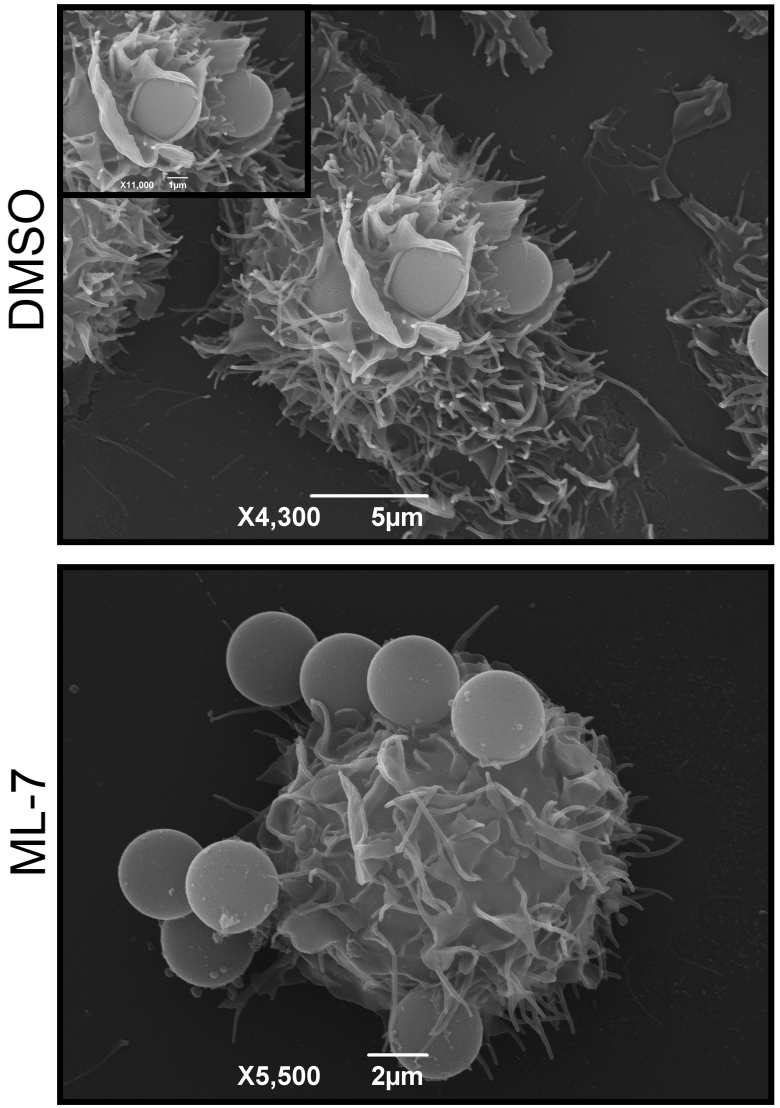
Applied pressure profile



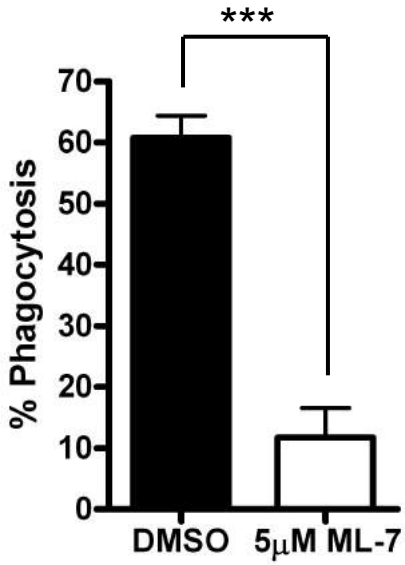
Contour length

Need to calculate pressure from model for acto-myosin cytoskeleton and signalling

Myosin inhibitor ML-7 turns phagocytosis off

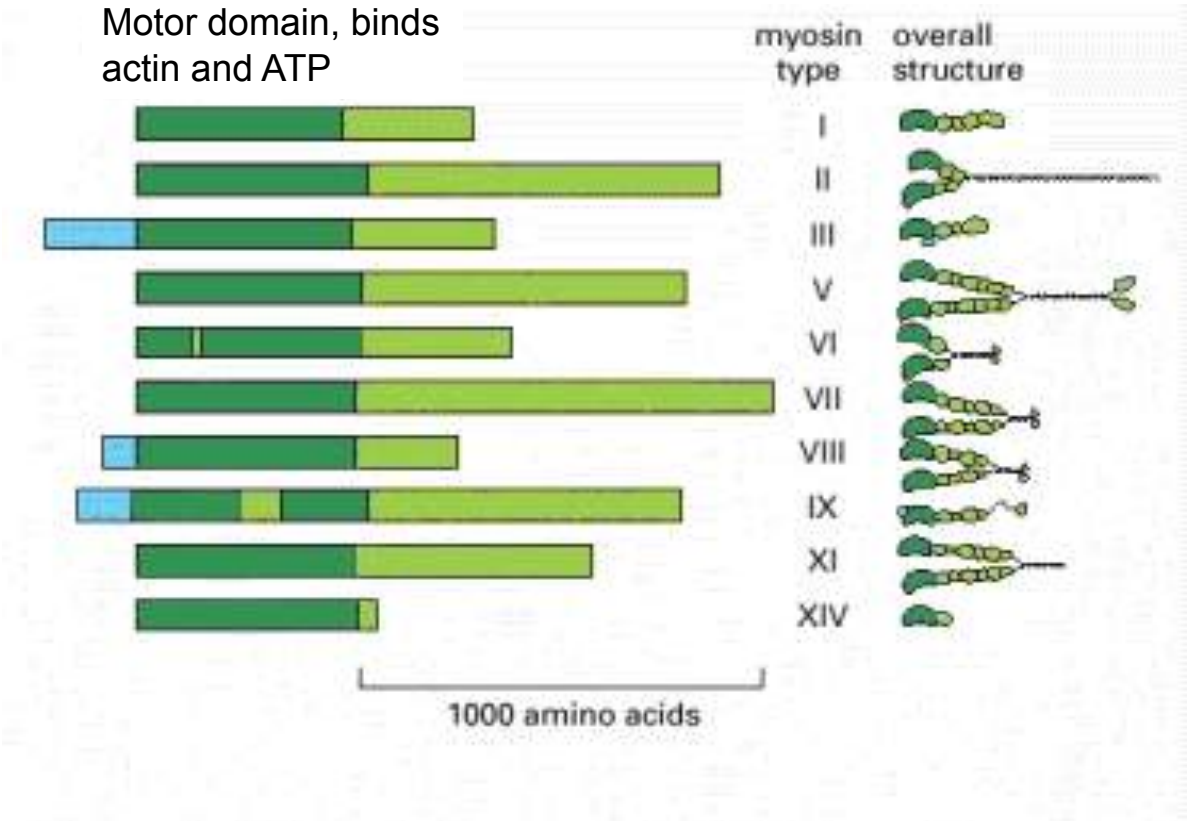


ML-7 is an inhibitor of myosin-light-chain kinase → perturbation of myosin-II



Domain structure of myosins

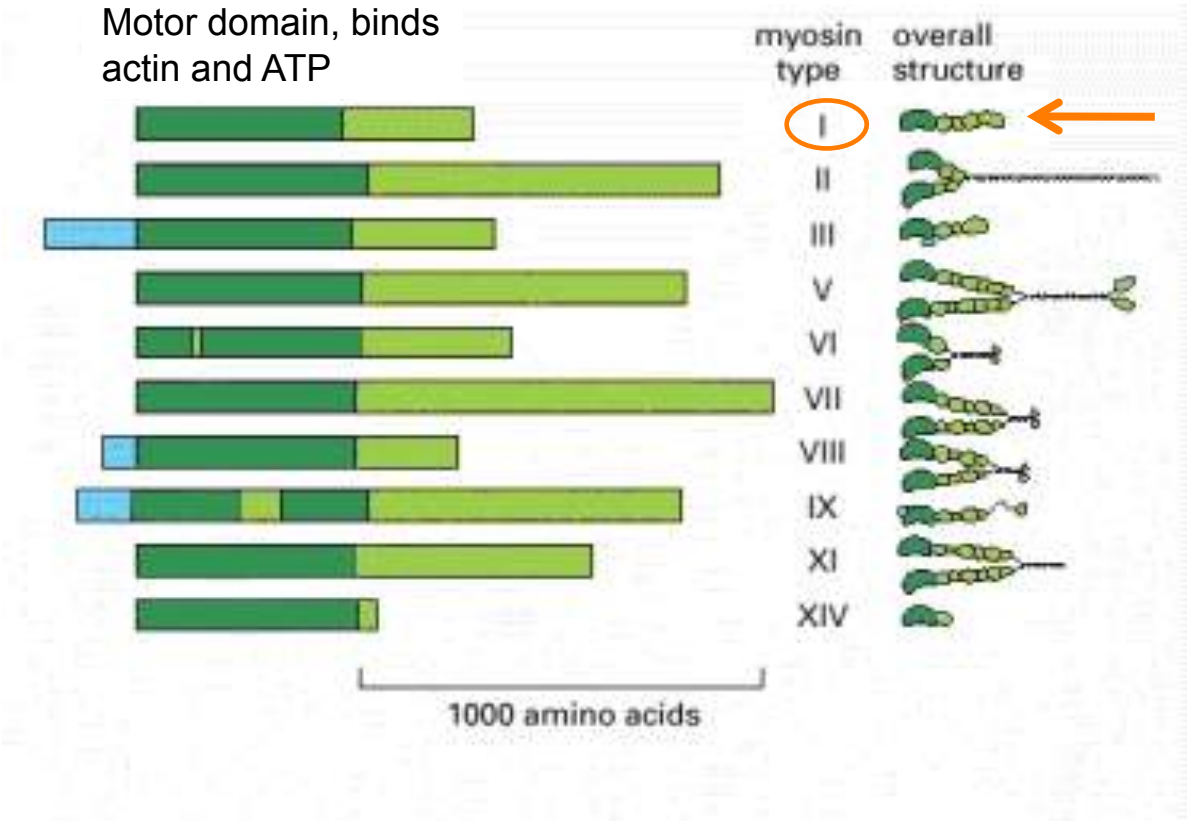
Motor proteins show diverse N- and C-terminus tails



Regulate membrane tension, purse-string contraction, pseudopod extension, transport, e.g. of membrane, and cell elasticity.

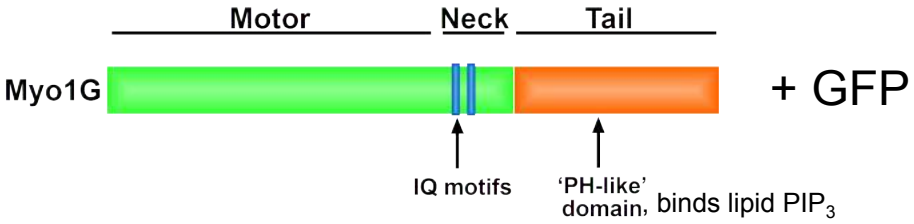
Domain structure of myosins

Motor proteins show diverse N- and C-terminus tails

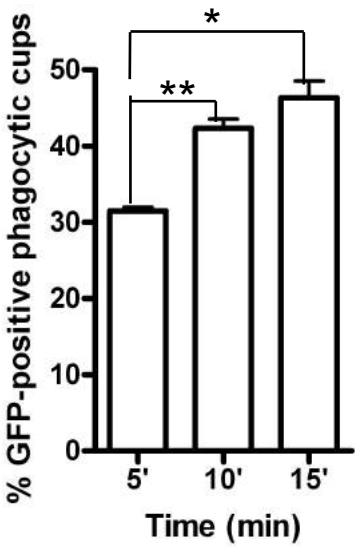


Regulate membrane tension, purse-string contraction, pseudopod extension, transport, e.g. of membrane, and cell elasticity

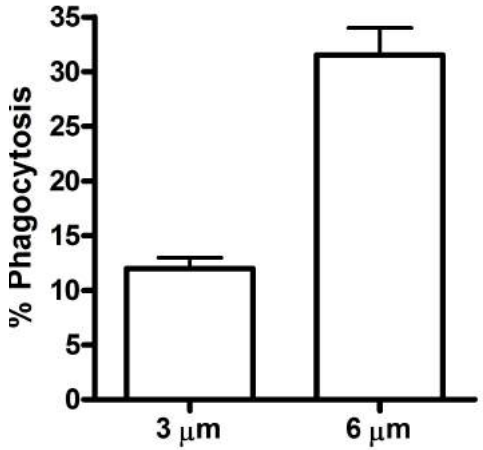
Myosin 1G – new role in phagocytosis



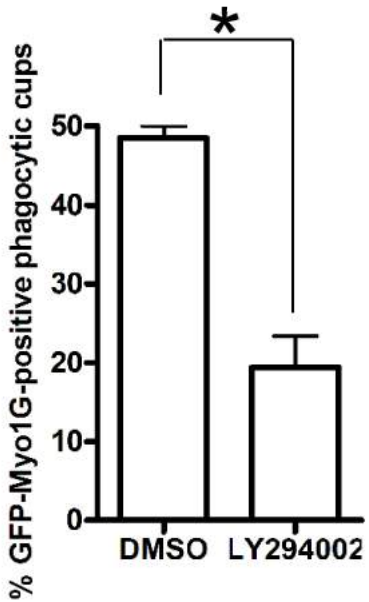
Recruited to cups



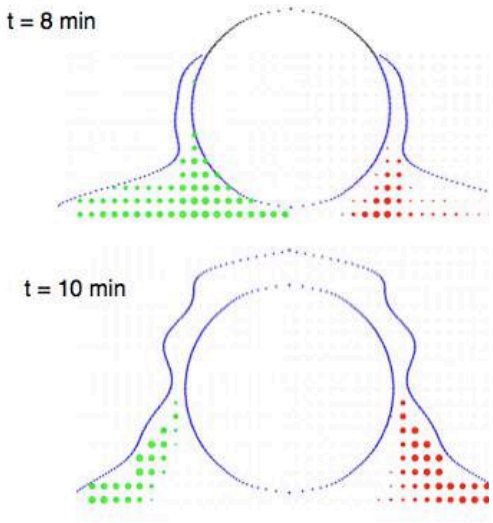
Required for uptake of large particles



Downstream of kinase PI3K

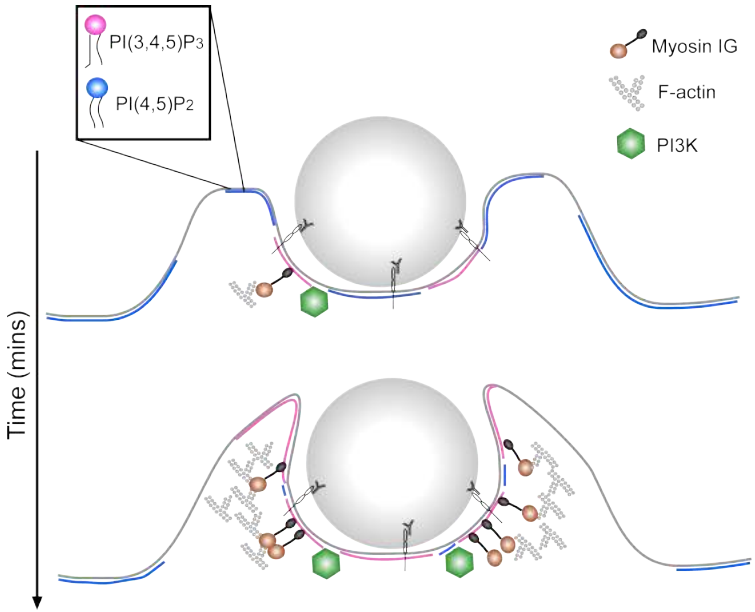


Myo1G and actin colocalise



Myosin 1G – dynamic linking of actin and lipids

Conceptual model: PI3K produces PIP_3 lipids, a second messenger for Myo1G and cup closure



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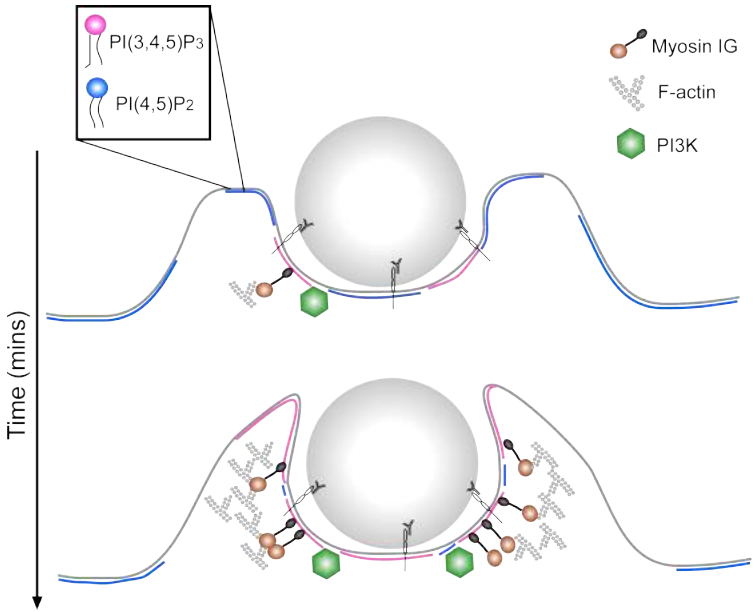
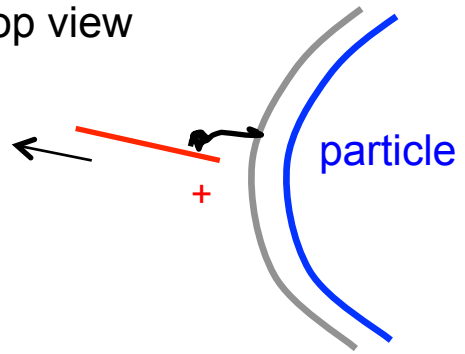
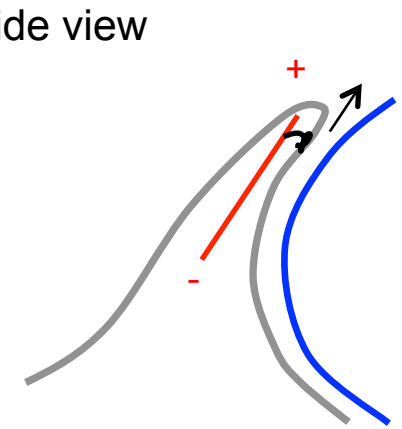


Role of Myo1G motor?

1) Radial force for cup closure

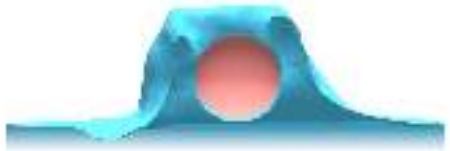


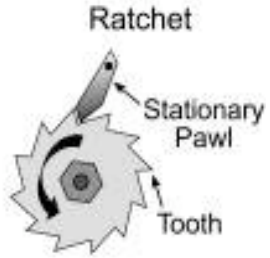
2) Transport of PIP_3 to cup rim



Summary of phagocytosis work

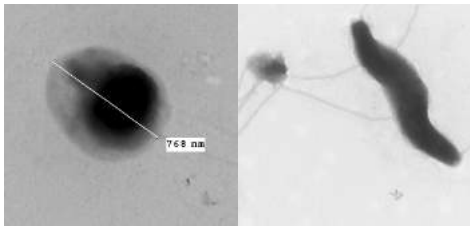
- Implemented a 3D ratchet model for Zipper mechanism.





- Particle size does not matter for active zipper, but for passive zipper.

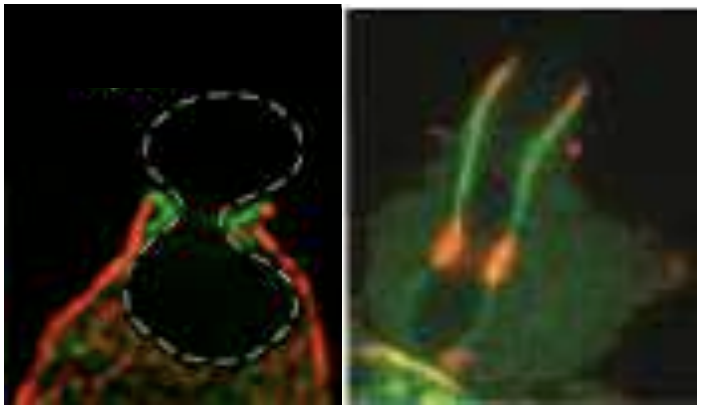
- Biochemical signalling pathways added by evolution for additional robustness?



Coccoid and helical shapes

- Shape and orientation matter for active zipper: Potentially important for host-pathogen interactions and efficient drug design.

- Current model does not close cups well, contraction by motor proteins necessary? Wave-like nature of actin polymerization?



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- Thierry Soldati (Geneva)

Tollis S, Dart A, Tzircotis G, Endres RG, BMC Syst Biol 4: 149 (2010); http://arxiv.org/abs/1011.0370.

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BBSRC bioscience for the future

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