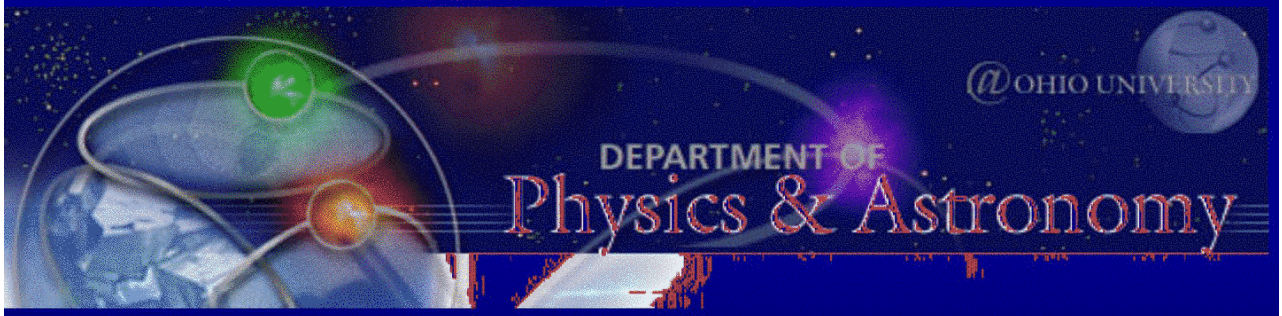
# Optimal Ion Channel Clustering

J.W. Shuai and P. Jung




Quantitative Biology Institute at Ohio University

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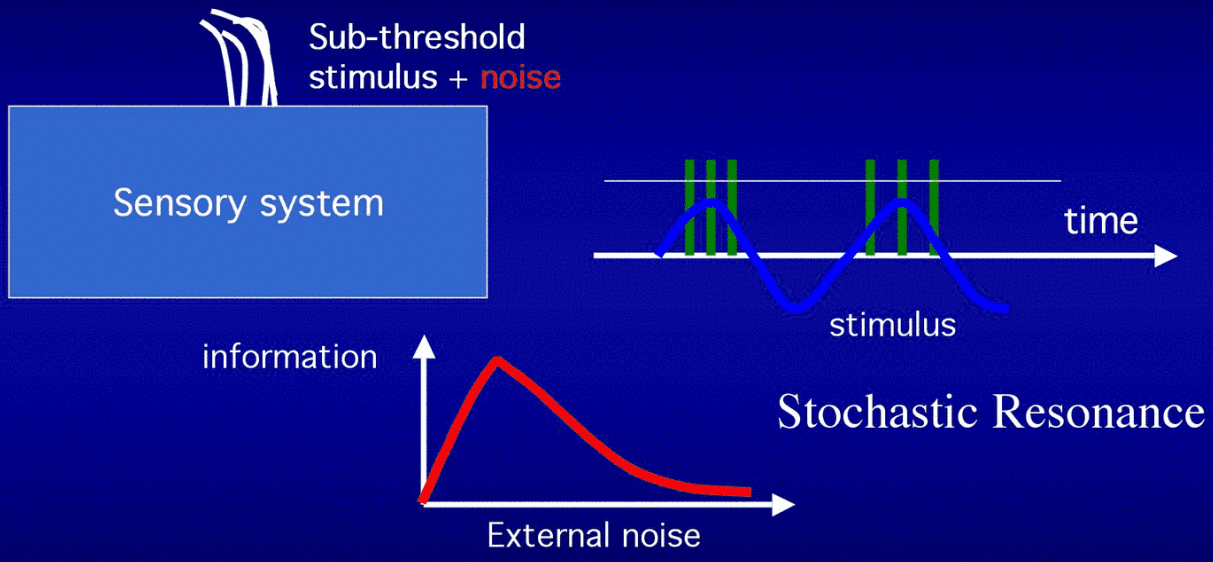
# Motivation




Stochastic Resonance

Review: Gammaitoni, Hanggi, Jung, Marchesoni, Rev.Mod. Phys. 1998
Longtin, Bulsara, Moss, PRL, 1991
Douglas, Wilkins, Pantazelou, and Moss., Nature, 1993
Levin and Miller, Nature 1996

# Size, Noise and Optimal Design

Do living systems exploit fluctuations to optimize their functions (stochastic resonance, coherence resonance) ?

If yes, how ?

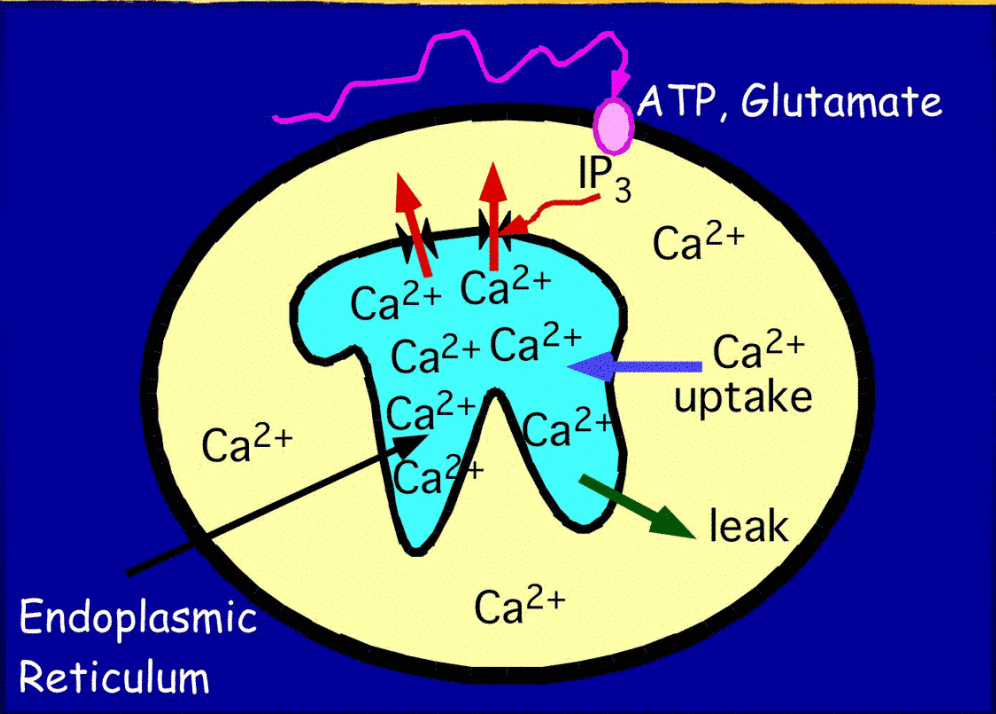
- Size <--> noise level --> size tuning

Neurons: (Shuai and Jung (2001), Schmid et al. (2001))

Intracellular $Ca^{2+}$ signaling (Shuai and Jung (2002))

Coupled bistable systems (Pikovsky, 2002)

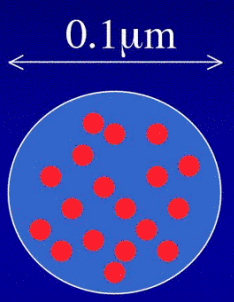
# Intracellular $Ca^{2+}$ Release



# Questions

- Optimal sizes of single clusters ?
- Optimal cluster configurations ?
- Biologic function: Is there a benefit from the clustering

# $IP_3$ Receptor Cluster




$IP_3$ - binding site

$Ca^{2+}$ binding site

G. DeYoung
J. Keizer
(1992)

$L_{\text{diff}}(0.1\text{s}) \approx 1\mu\text{m}$

Eliminate fast time scales ! (Li and Rinzel 1994)

Calcium equilibrates instantaneously within cluster !

# Stochastic Li-Rinzel Model

(Li and Rinzel 1994, Shuai and Jung 2002)

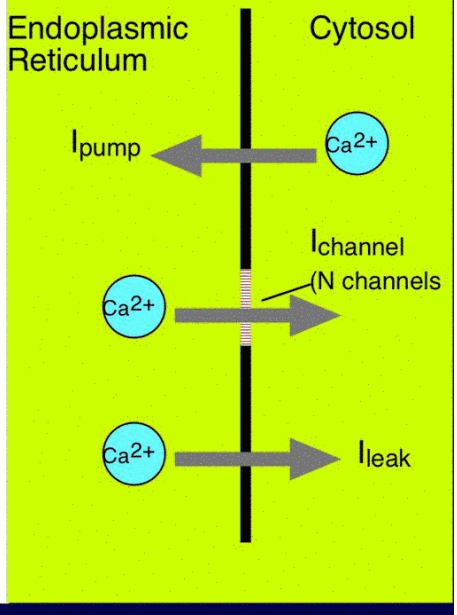
$$\frac{d[Ca^{2+}]}{dt} = I_{\text{channel}} + I_{\text{Pump}} + I_{\text{Leak}}$$

$$I_{\text{channel}} = -c_1 v_1 n_\infty^3 m_\infty^3 \frac{N_{open}}{N}\left([Ca^{2+}] - [Ca^{2+}]_{ER}\right)$$

$$I_{\text{Pump}} = -\frac{v_3 [Ca^{2+}]^2}{k_3^2 + [Ca^{2+}]^2}$$

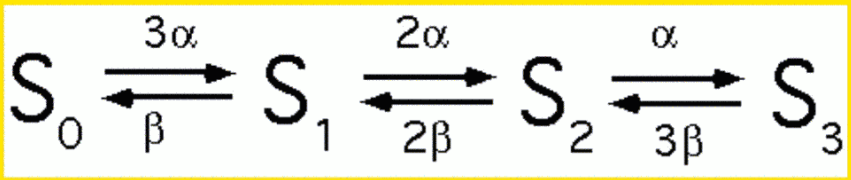
$$I_{\text{Leak}} = -c_1 v_2\left([Ca^{2+}] - [Ca^{2+}]_{ER}\right)$$

$\frac{N_{open}}{N}$: fraction of releasing channels




# Stochastic Cluster Kinetics

Keep track of number of channels in all states!




Markov-Monte-Carlo scheme

$$p(m: S_1 \rightarrow S_2) = \binom{N_1}{m}(2\alpha\Delta t)^m (1 - 2\alpha\Delta t)^{N_1 - m}$$

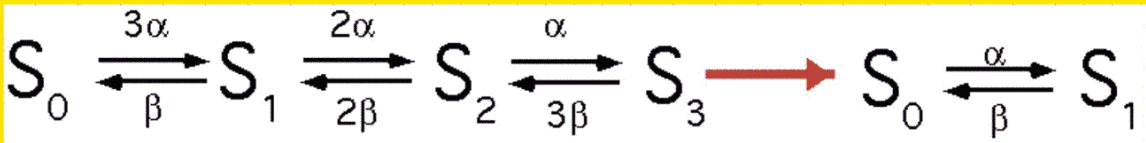
$\alpha([IP_3])$ activation

$\beta([c])$ inactivation

(Clay and DeFelici 1983, Chow and White 1996, Schneidman 1998)

# Langevin Approximation

(Fox and Liu, 1994)




Master Equation for Probability of open channels:

$$\frac{\partial P(N,t)}{\partial t} = G^{+}(N-1)P(N-1,t) - \left(G^{+}(N) + G^{-}(N)\right)P(N,t) + G^{-}(N+1)P(N+1,t)$$

$$G^{+}(N) = (N_t - N)\alpha_h$$

$$G^{-}(N) = N\beta_h \,.$$

# Kramers-Moyal Expansion

$$n = N / N_t \quad , \quad \rho(n) \equiv P\left(N_t n\right) \quad , \quad \varepsilon = 1 / \mathrm{N_t}$$

$$g^{\pm}(n) = G^{\pm}(N) / N_t$$

$$\frac{\partial \rho(n,t)}{\partial t} = \sum_{i=1}^{\infty} \frac{1}{i!} \varepsilon^{i-1} \left(-\frac{\partial}{\partial n}\right)^{i} M_i(n)\rho(n,t)$$

$$M_i(n) = g^{+}(n) + (-1)^{i} g^{-}(n) = (1-n)\alpha_h + (-1)^{i} n\beta_h$$

# Langevin Equation (large N)

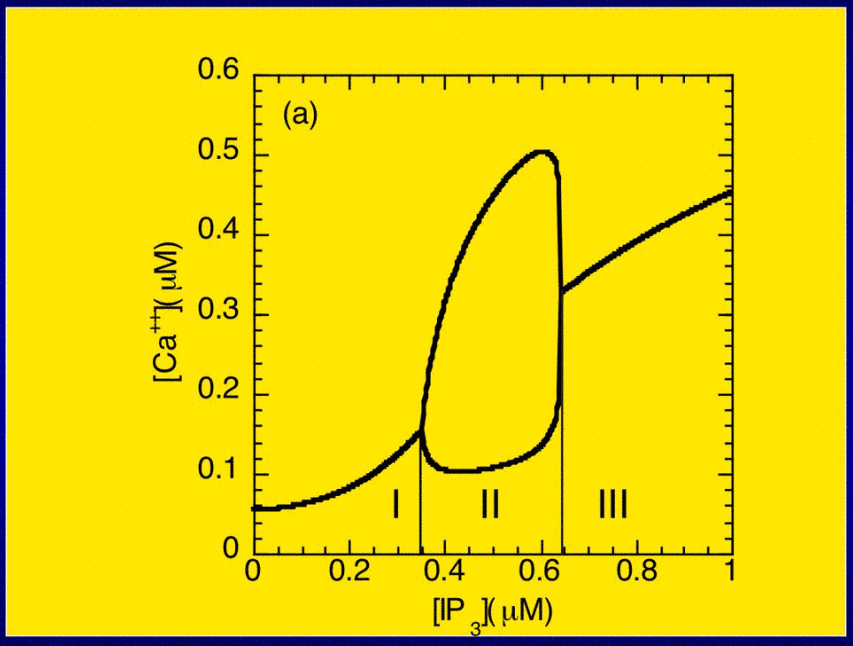
$$\frac{dc}{dt} = -c_1 v_1 m_\infty^3 n_\infty^3 h^3 (c - c_{ER}) - c_1 v_2 (c - c_{ER}) - \frac{v_3 c^2}{k_3^2 + c^2}$$

$$\frac{dh}{dt} = \alpha_h (1 - h) - \beta_h h + \xi(t)$$

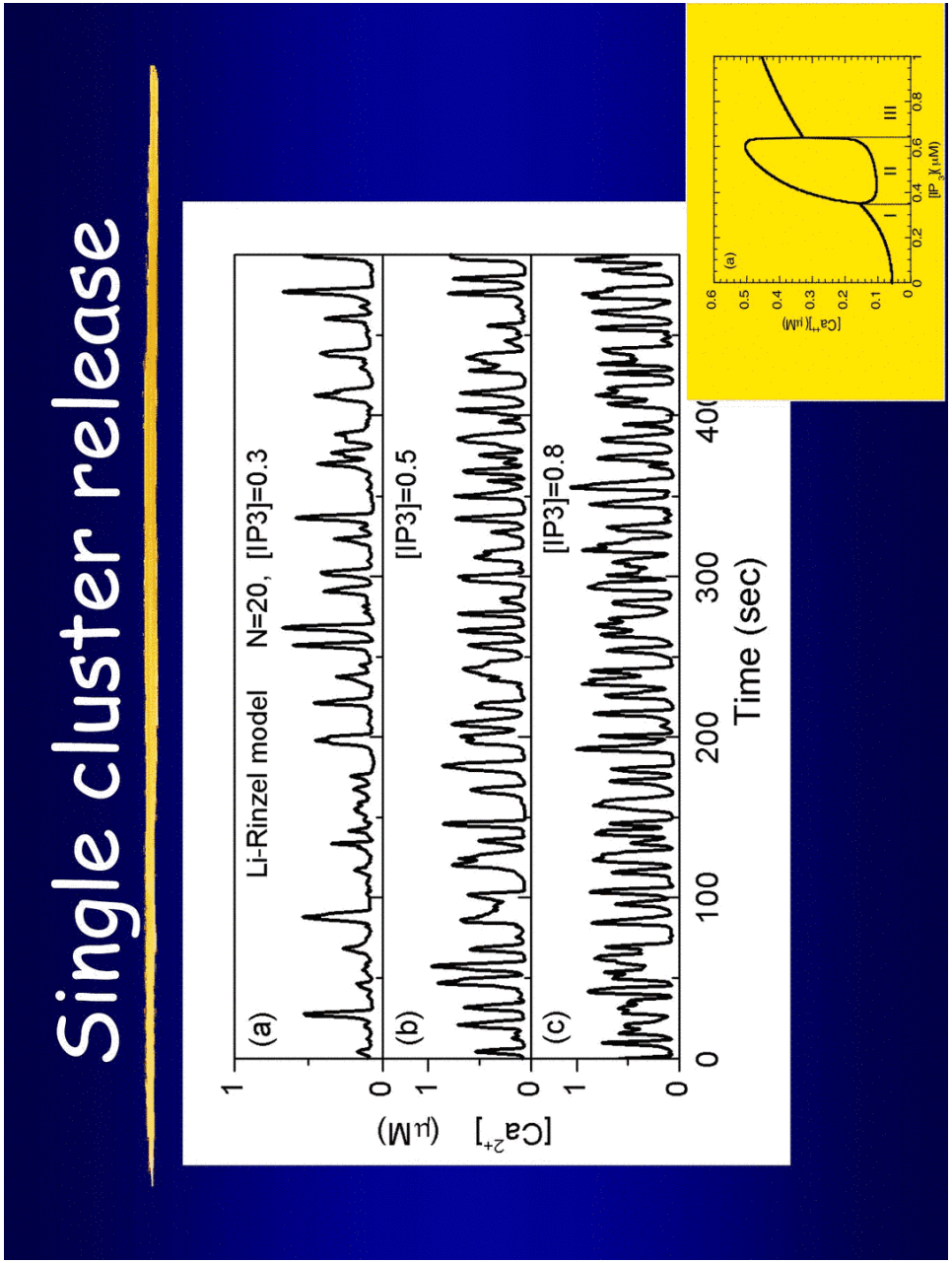
$$\langle \xi(t) \xi(t') \rangle = \frac{\alpha_h (1 - h) + \beta_h h}{2 N_t} \delta(t - t')$$

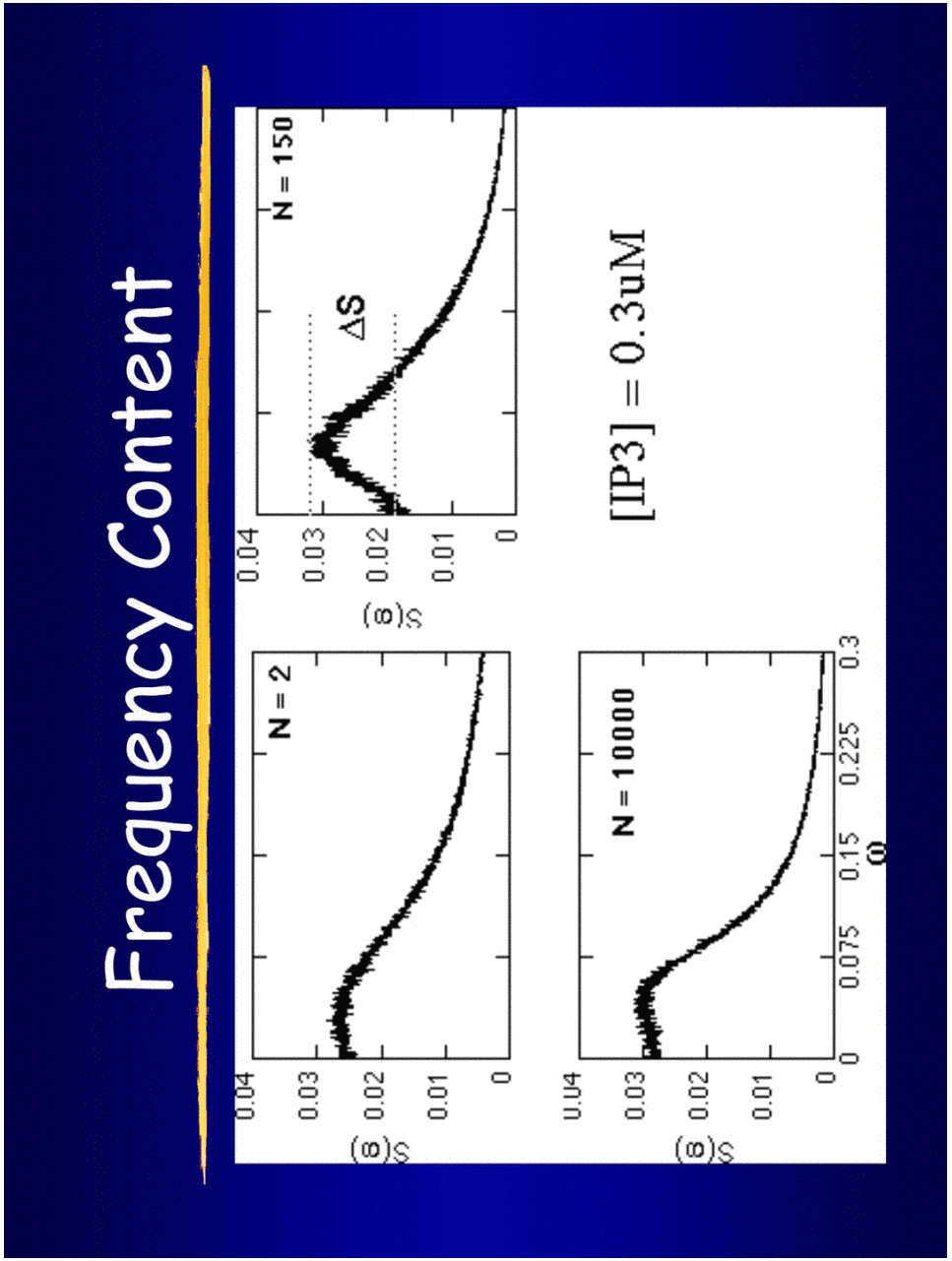
$$\langle \xi(t) \rangle = 0$$

# Calcium Oscillations

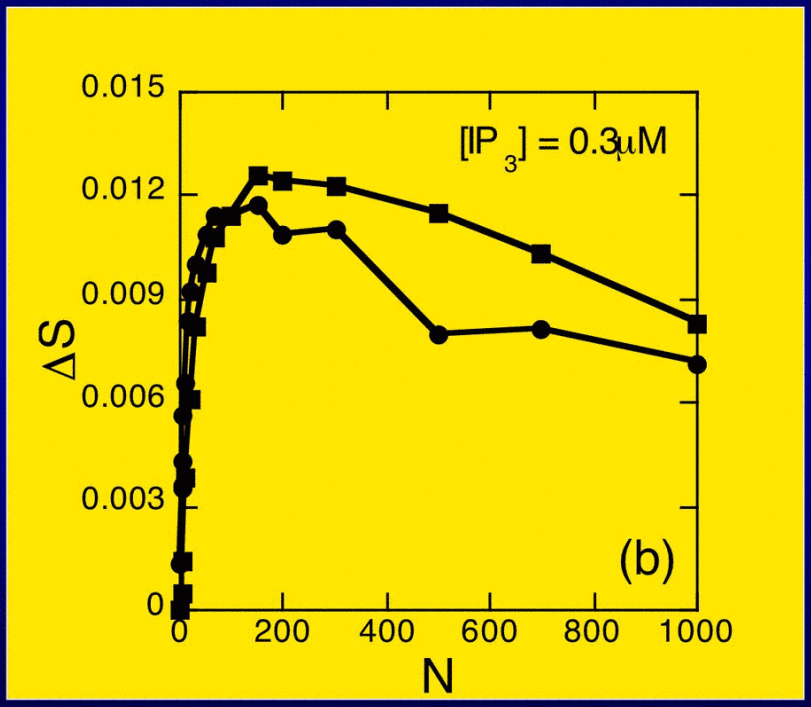



$Ca^{2+}$ signal mostly in frequency content !


Single cluster release
Li-Rinzel model N=20, [IP3]=0.3
(a)
(b)
(c)
[IP3]=0.5
[IP3]=0.8
[Ca2+] (μM)
Time (sec)
0
100
200
300
I
II
III


Frequency Content
N = 2
N = 150
N = 10000
ΔS
[IP3] = 0.3uM
S(ω)
ω

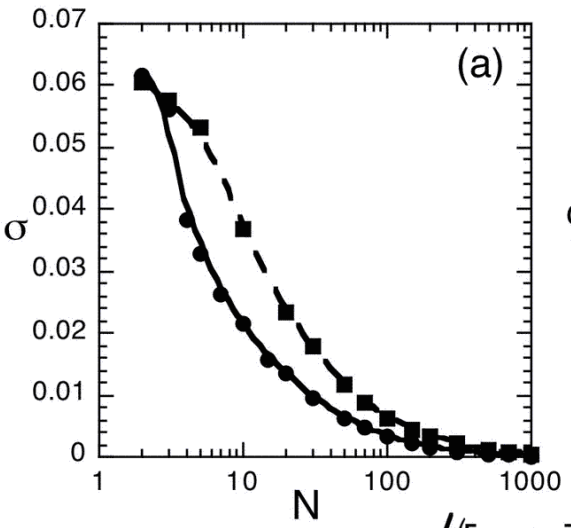
# Signaling Coherence

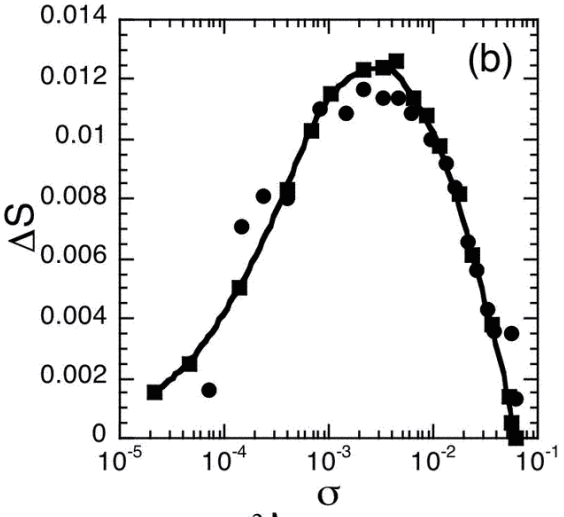



Optimal Size of cluster!

(J.W. Shuai and P.J, 2002)

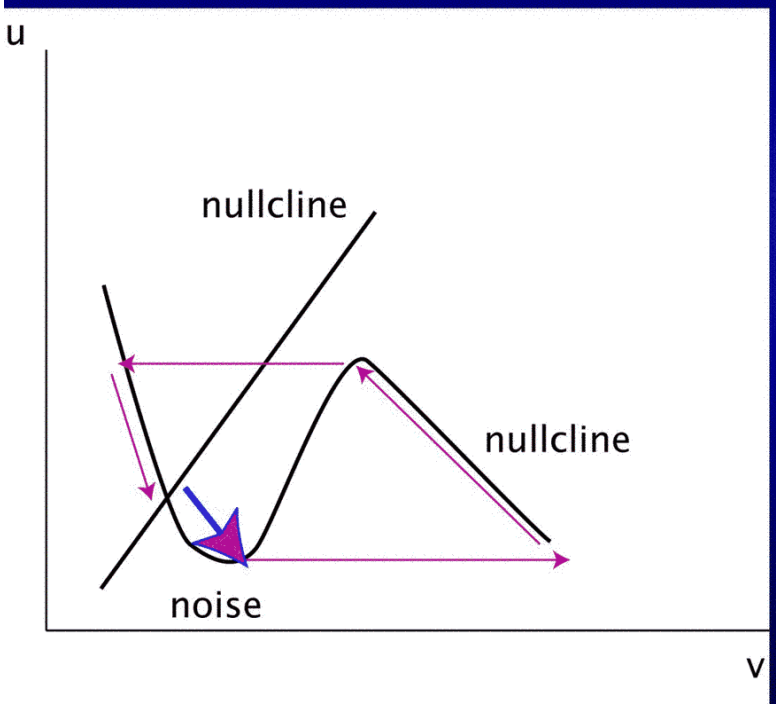
# Coherence versus Noise



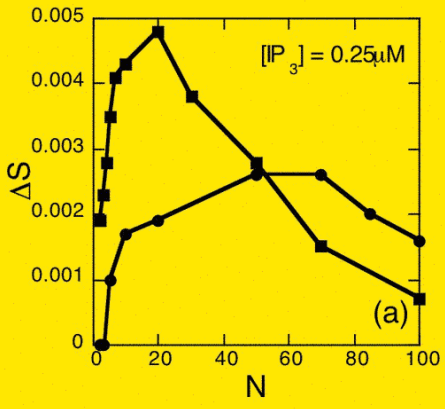




$$\sigma = \left\langle \left( \left[ Ca^{2+} \right](t) - \left\langle \left[ Ca^{2+} \right] \right\rangle \right)^2 \right\rangle$$

# Coherence Resonance




Can we predict the optimal cluster size as we decrease IP3?

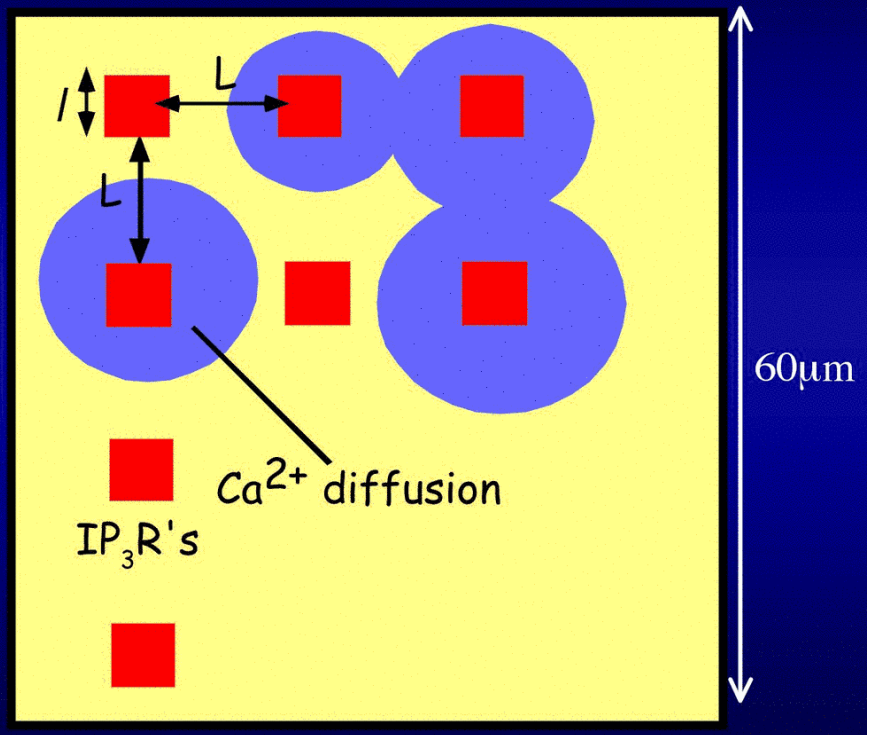



# Calcium Signals in Cells

Active clusters embedded in medium !

Pumps and leaks Homogeneously distributed.

Smith et al., 1998
Falcke et al.,2000



# The Model Equations

$$\frac{\partial [Ca]}{\partial t} = f(x,y) I_{\text{channel}} - I_{\text{pump}} + I_{\text{leak}} + D\nabla^2 [Ca]$$

$$\frac{\partial [Ca]_{ER}}{\partial t} = -f(x,y) I_{\text{channel}} + I_{\text{pump}} - I_{\text{leak}} + D_{ER}\nabla^2 [Ca]_{ER}$$

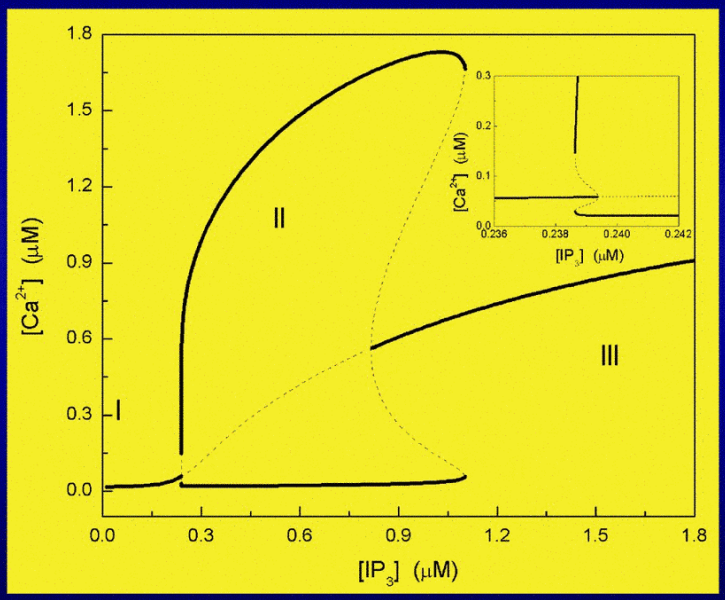
$$f(x,y) = \begin{cases} 1 & \text{in cluster} \\ 0 & \text{otherwise} \end{cases} \qquad J^{i,j}_{\text{channel}} \propto N^{i,j}_{\text{open}} \left( \left[ Ca^{2+} \right] - \left[ Ca^{2+} \right]_{ER} \right)$$

$D = 20 - 30 \mu m^2/s$ $D_{ER} < 1 \mu m^2/s$

Parameters adjusted (correct wave speed and frequency) (Shuai and Jung, PRE, 2003).

Solve PDE's together with Markov-simulations of embedded clusters (Shuai and Jung, PRE (2003), PNAS (2003)).

# Bifurcation Diagram

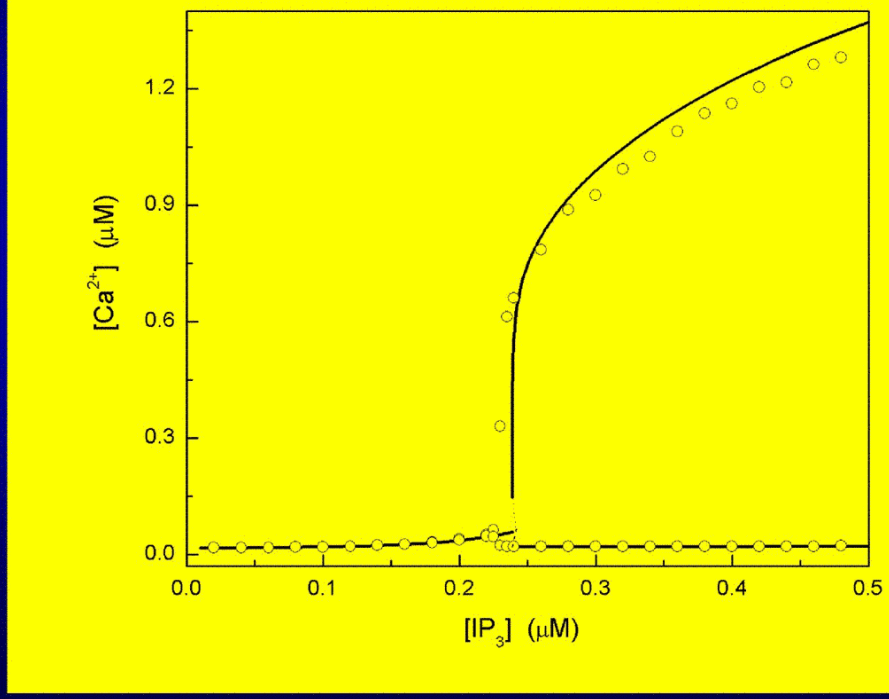



I: no coding

II: coding

III: no coding

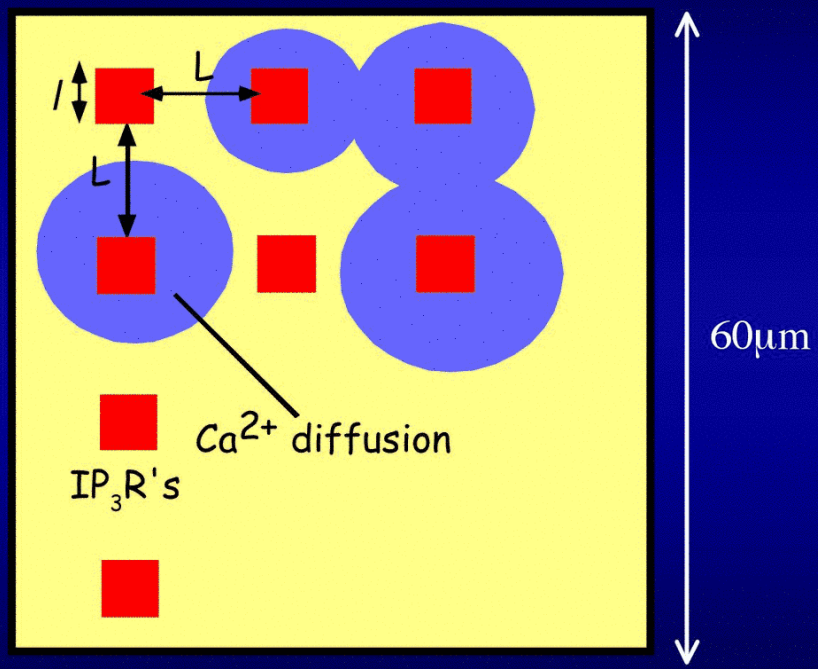
# Large Ca2+ diffusion



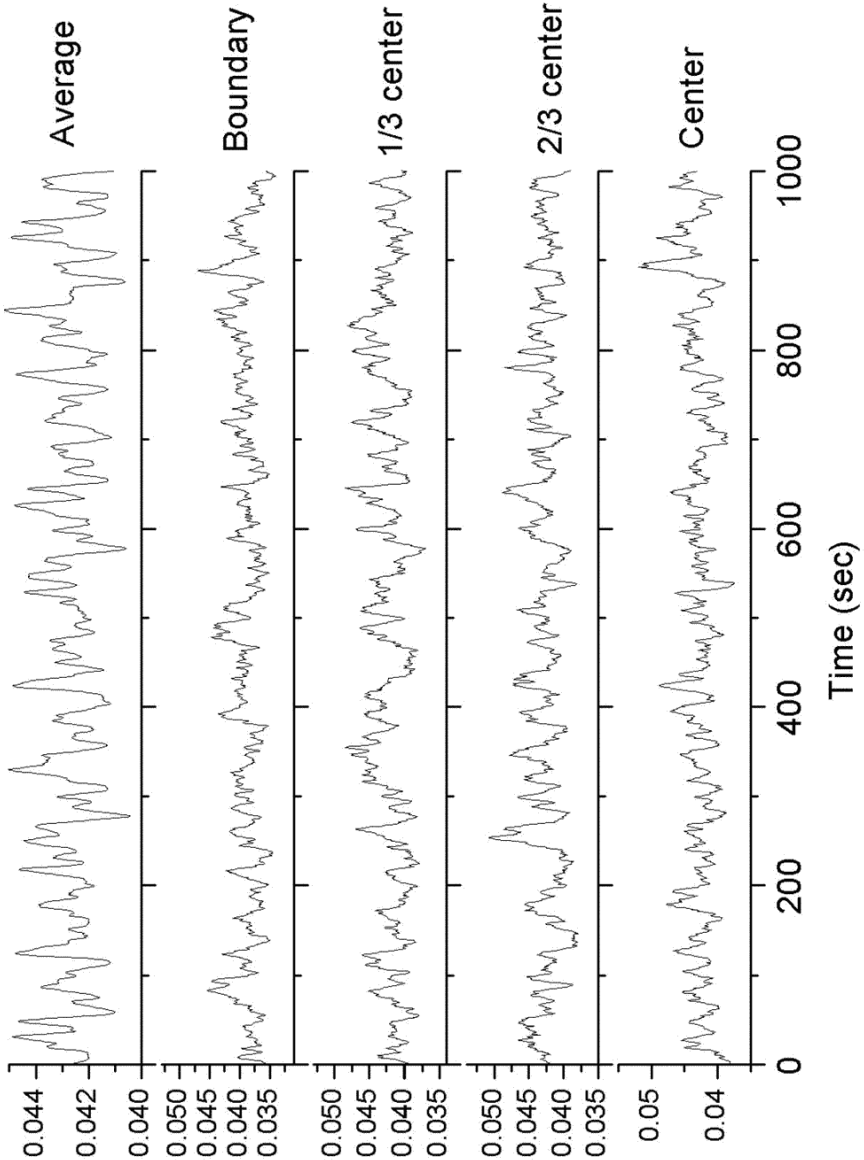

# Effects of Channel Clustering

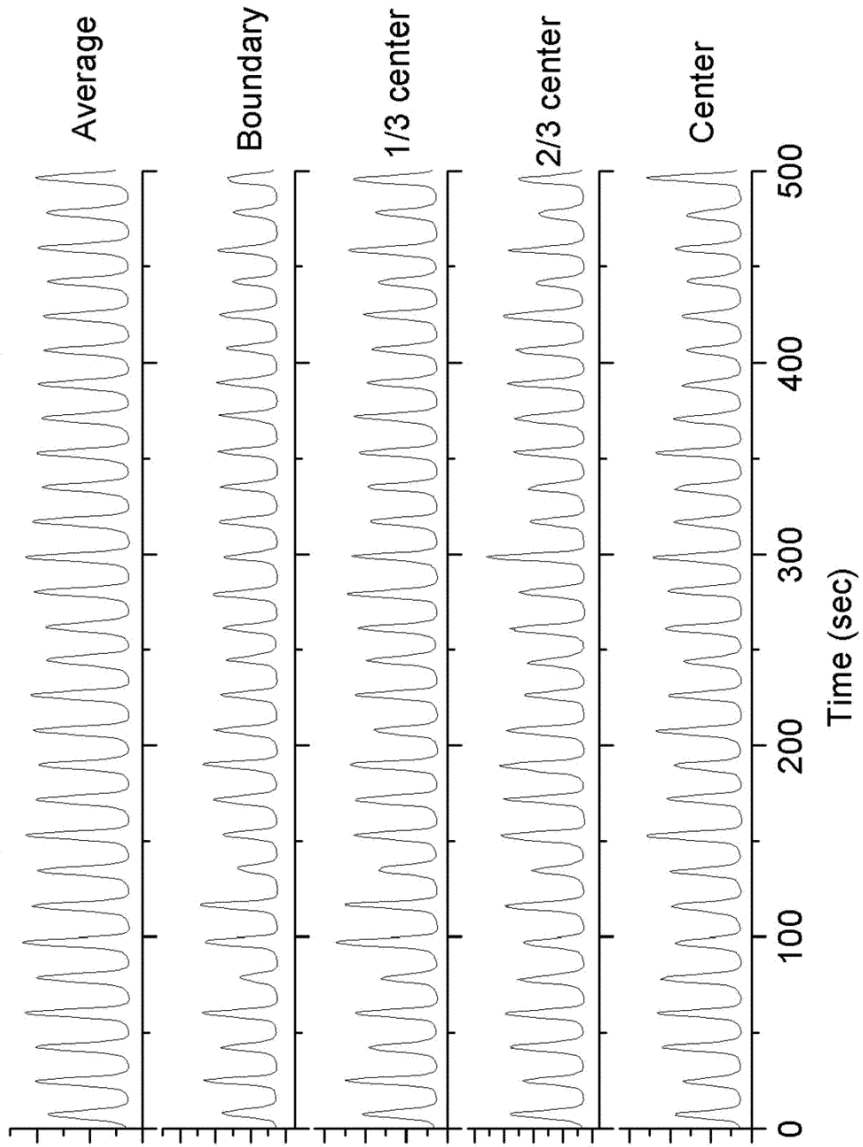
Total: 14,400 channels
0.5μm < L < 6 μm
1 < N < 144

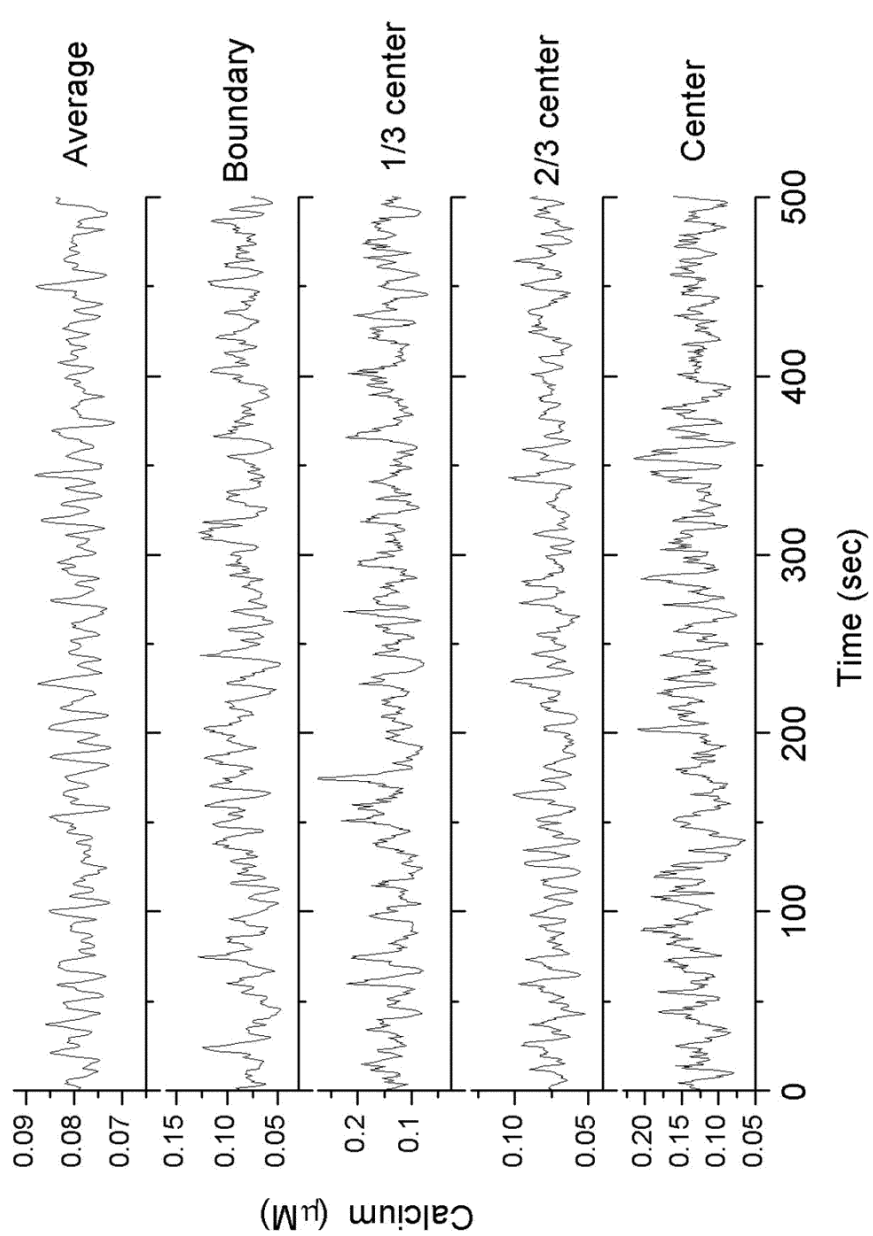
Clusters on regular grid as point sources

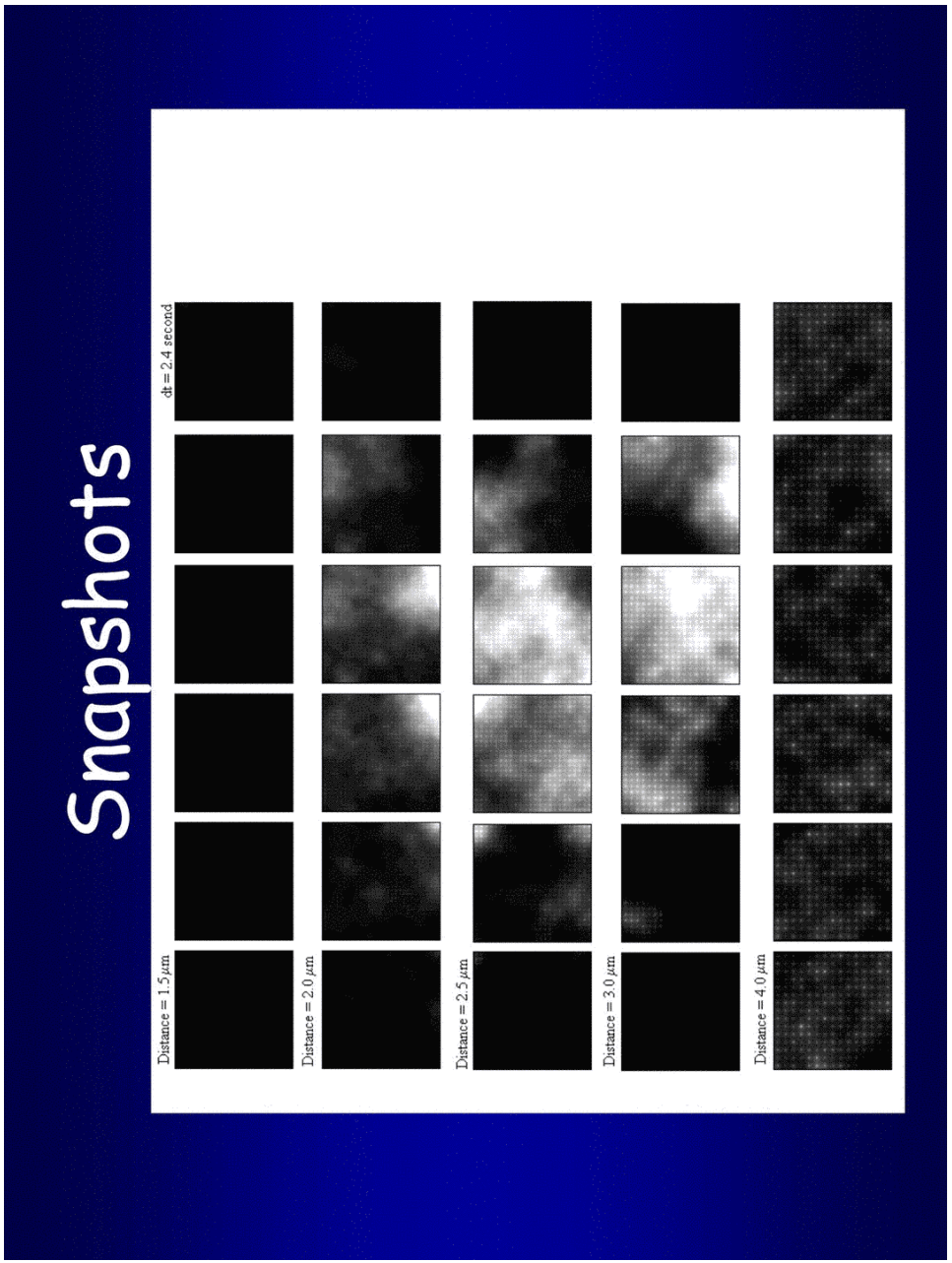


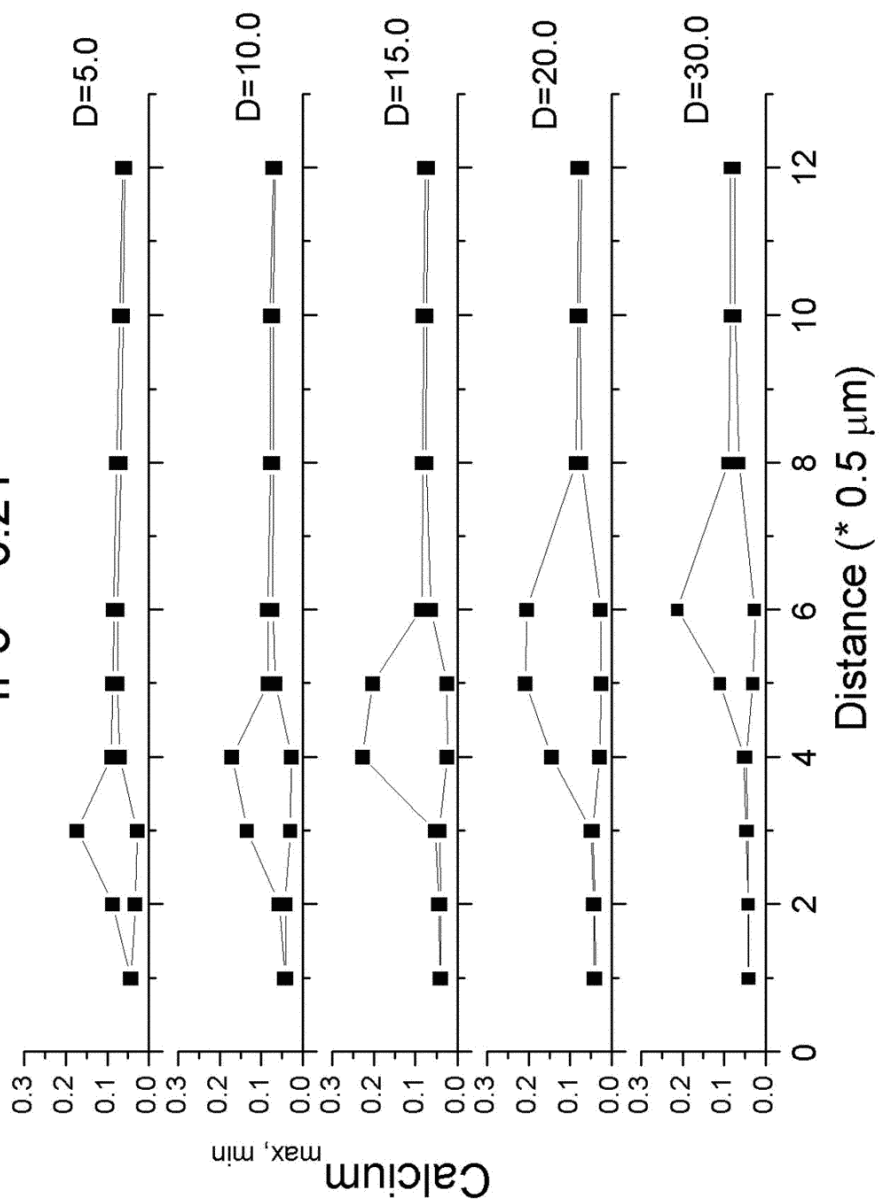

(Shuai and Jung, PNAS, 2003)

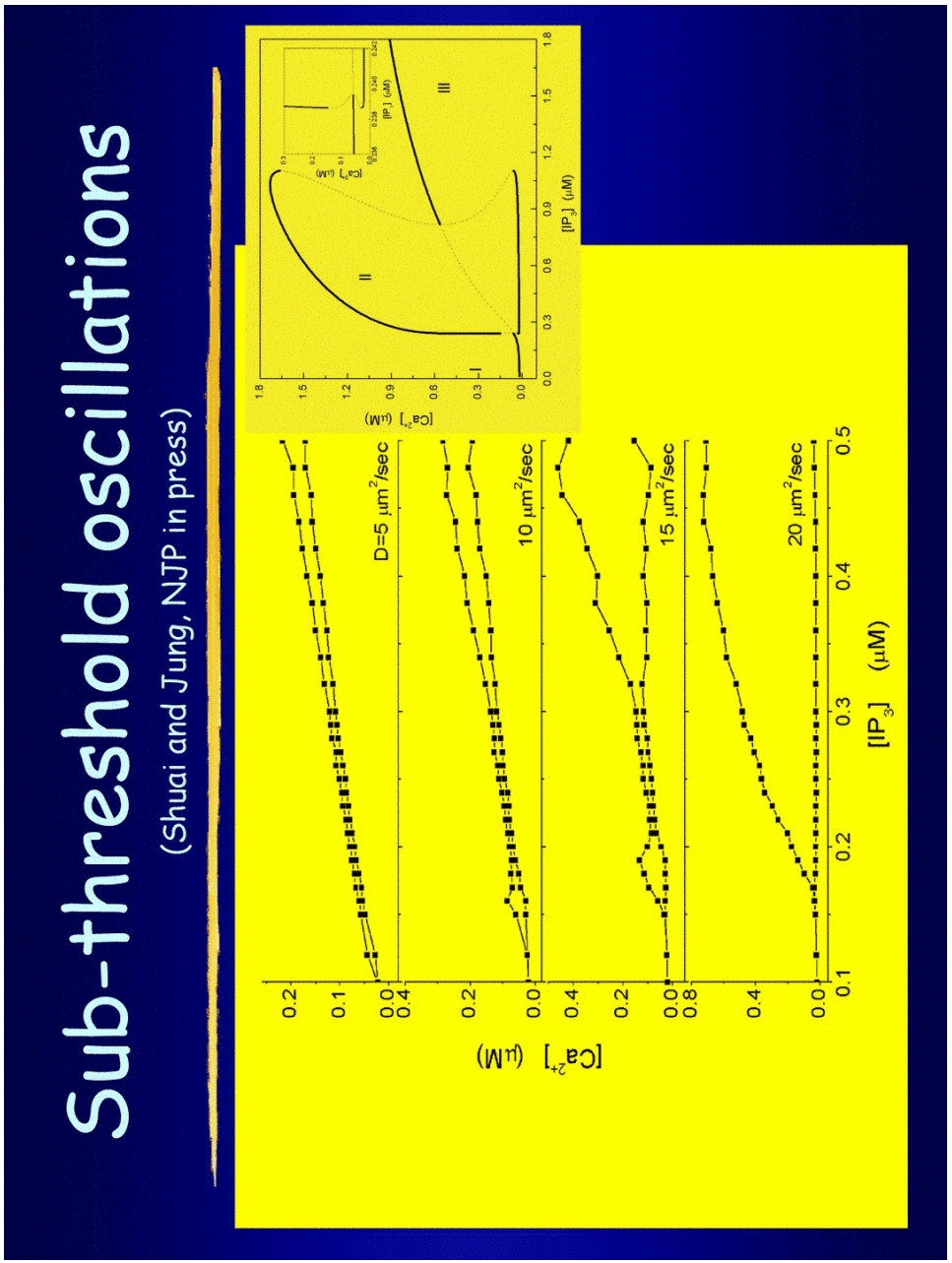

D=20 , IP3=0.21, Cluster Distance = 1 μm
Average
Boundary
1/3 center
2/3 center
Center
Calcium (μM)
Time (sec)

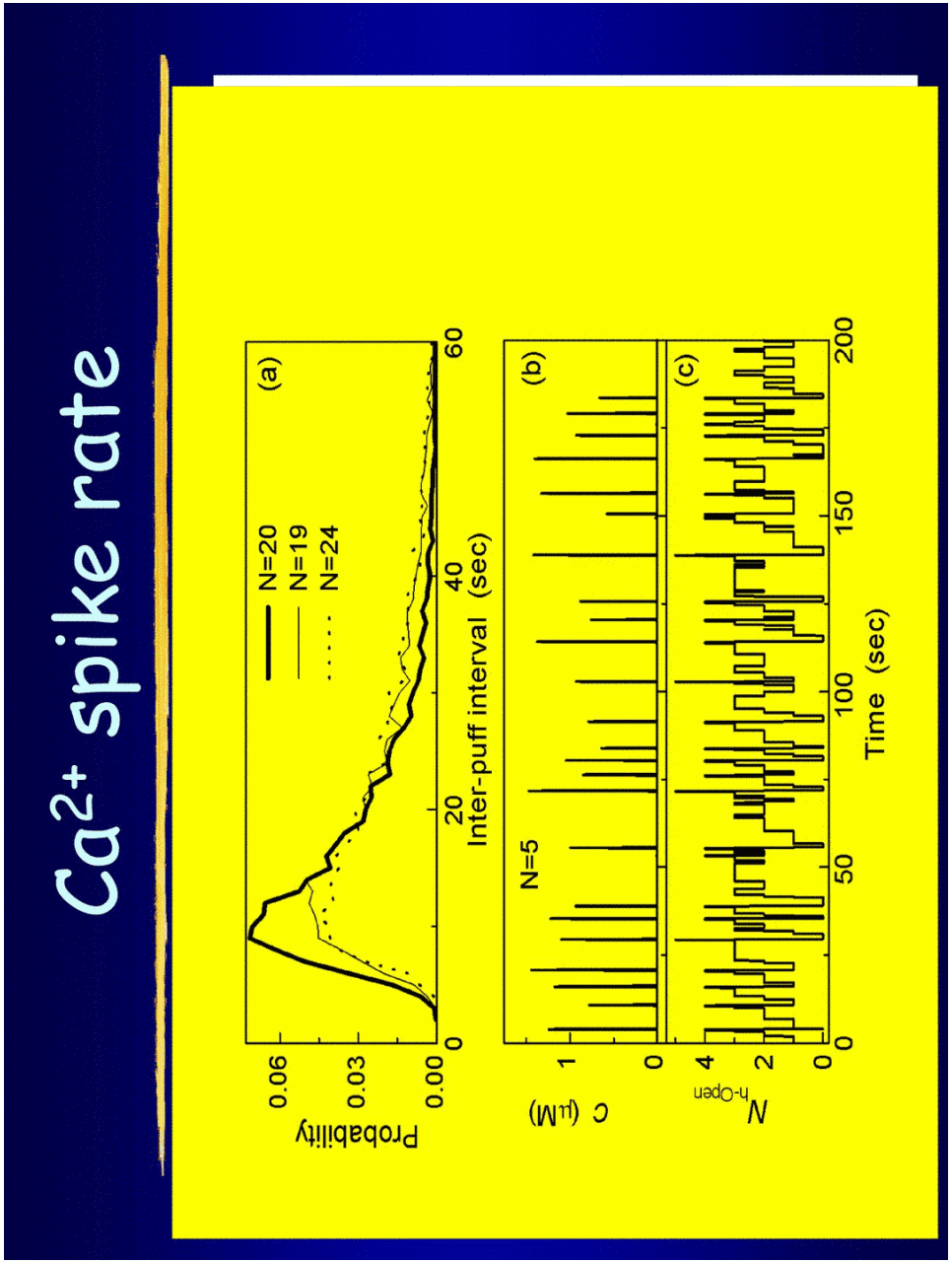

D=20, IP3=0.21, Cluster Distance = 3 μm
Average
Boundary
1/3 center
2/3 center
Center
Calcium (μM)
Time (sec)

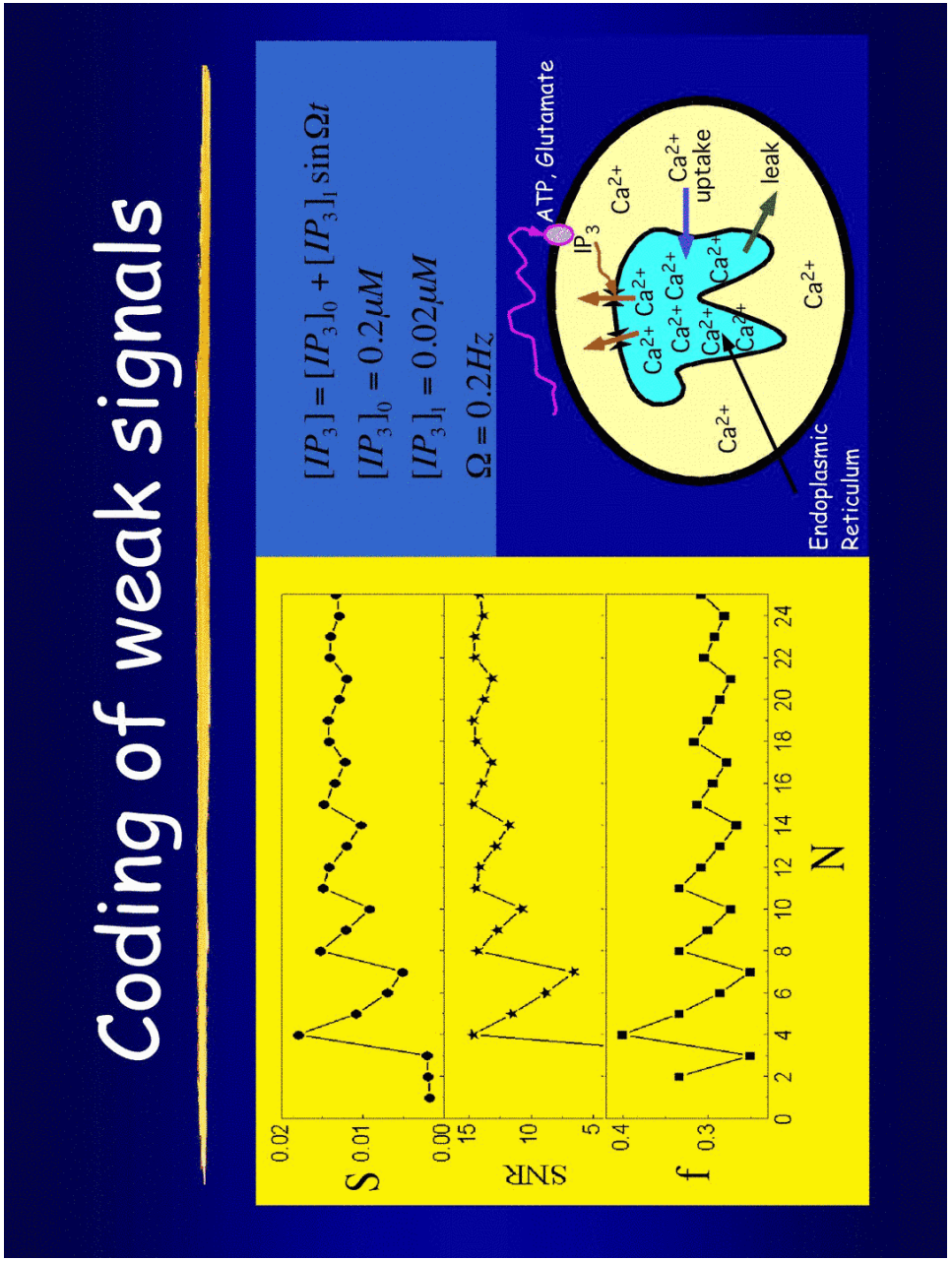

D=20, IP3=0.21, Cluster Distance = 4 μm
Average
Boundary
1/3 center
2/3 center
Center
Calcium (μM)
Time (sec)
0.09
0.08
0.07
0.15
0.10
0.05
0.2
0.1
0.10
0.05
0.20
0.15
0.10
0.05
0
100
200
300
400
500


Snapshots
dt = 2.4 second
Distance = 1.5 μm
Distance = 2.0 μm
Distance = 2.5 μm
Distance = 3.0 μm
Distance = 4.0 μm


IP3 = 0.21
D=5.0
D=10.0
D=15.0
D=20.0
D=30.0
Calcium max, min
Distance (* 0.5 μm)


Sub-threshold oscillations
(Shuai and Jung, NJP in press)
D=5 μm²/sec
10 μm²/sec
15 μm²/sec
20 μm²/sec
[Ca²⁺] (μM)
[IP₃] (μM)


Ca2+ spike rate
(a)
N=20
N=19
N=24
Probability
0.06
0.03
0.00
0
20
40
60
Inter-puff interval (sec)
(b)
N=5
c (μM)
1
0
(c)
N_open
4
2
0
0
50
100
150
200
Time (sec)


Coding of weak signals
$[IP_3] = [IP_3]_0 + [IP_3]_1 \sin \Omega t$
$[IP_3]_0 = 0.2\mu M$
$[IP_3]_1 = 0.02\mu M$
$\Omega = 0.2Hz$
ATP, Glutamate
IP3
Ca2+
Ca2+ uptake
leak
Endoplasmic Reticulum
S
0.02
0.01
SNR
15
10
5
f
0.4
0.3
0
2
4
6
8
10
12
14
16
18
20
22
24
N

# Summary and Conclusions

- Channel clustering enhances cellular $Ca^{2+}$ signal to small stimuli. Optimal Clustering (few molecules binding)
- System-size fluctuations are an important player for this phenomenon.
- Large signal $Ca^{2+}$ signals not affected by clustering

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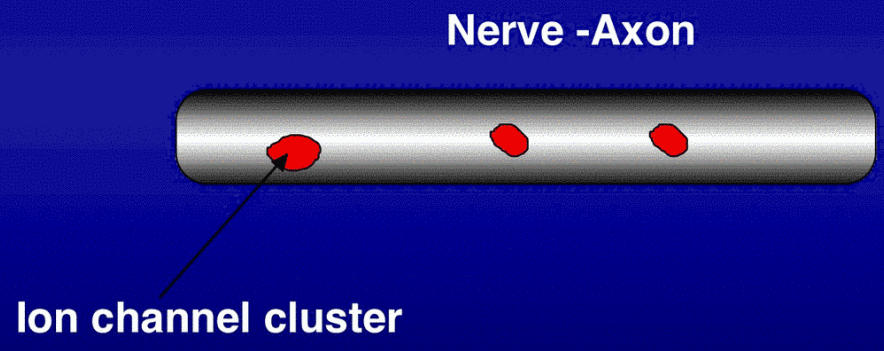
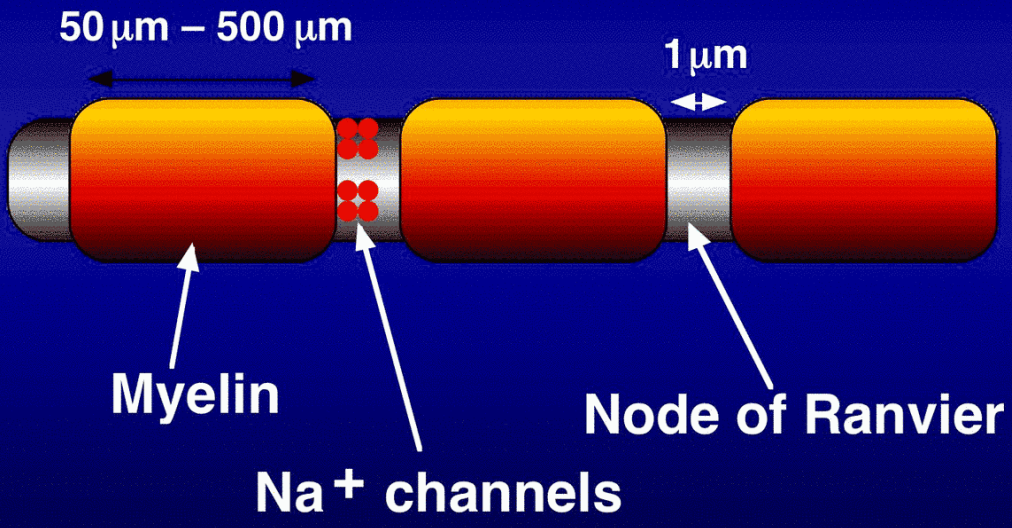
# Ion Channels in non-myelinated Axons

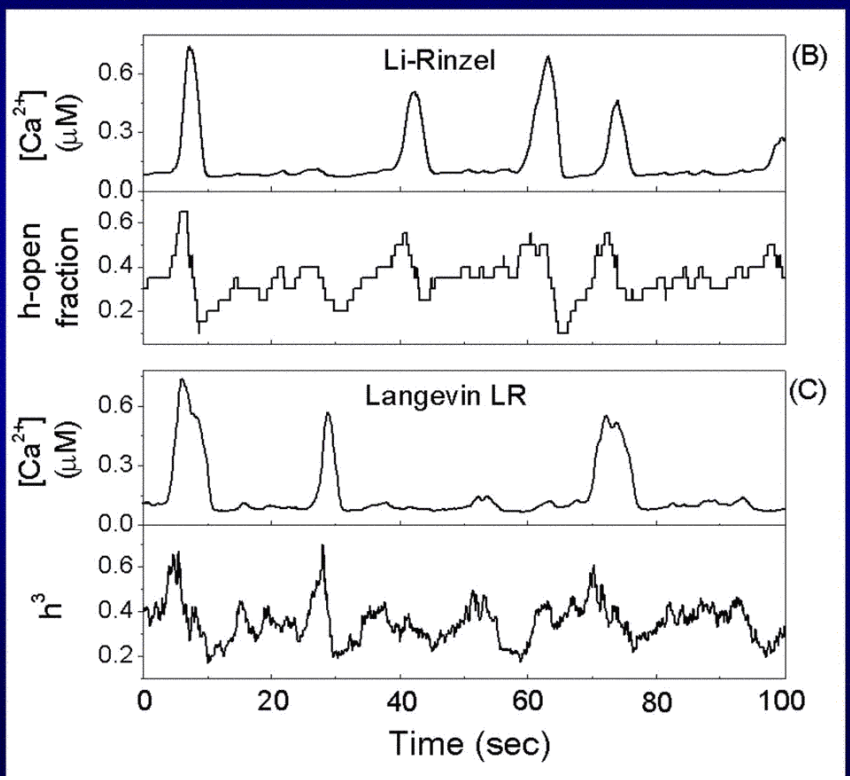



Rat - retinal nerve fiber layer (Hildebrand and Waxman, '83)
Squid giant axon ($K^+$ channels) (Clay et al. 2001)
Optimal cluster sizes (theory): Shuai and Jung, 2001

# Ion Channels in Myelinated Axons

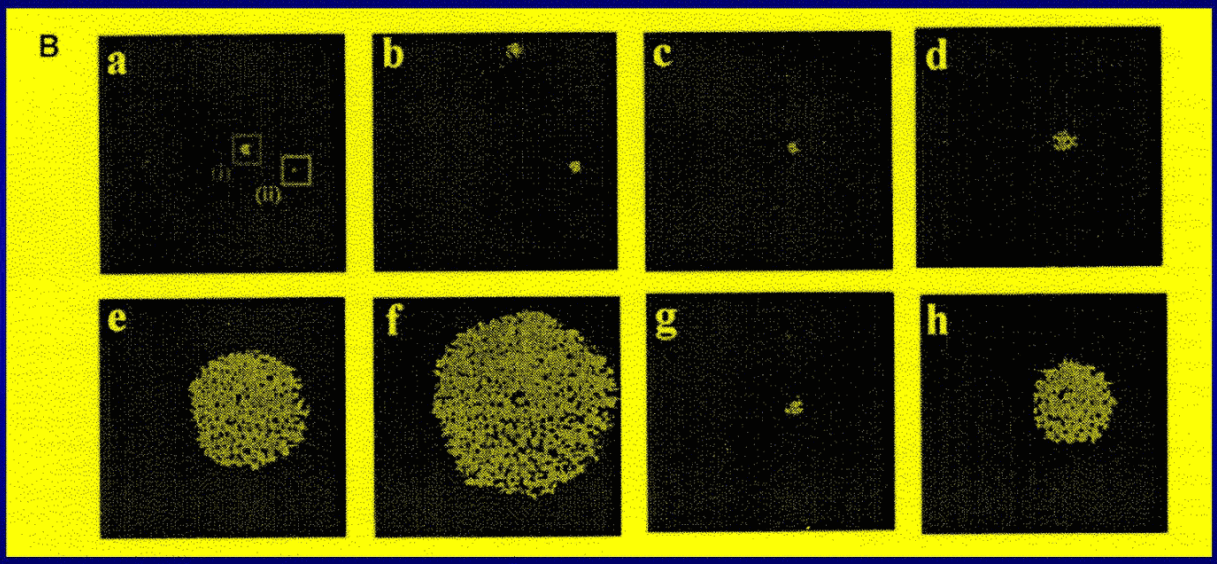


# Sample Trajectories




Cluster of 20 $IP_3Rs$ at $[IP_3]=0.3\mu m$

# Channels are clustered




Calcium Puffs: Snapshots of fluorescent images 8μm x 8μm (*Xenopus* oocyte, Marchant and Parker, EMBO (2001))

# $Ca^{2+}$ flux through Receptor channels (Li-Rinzel 1994)

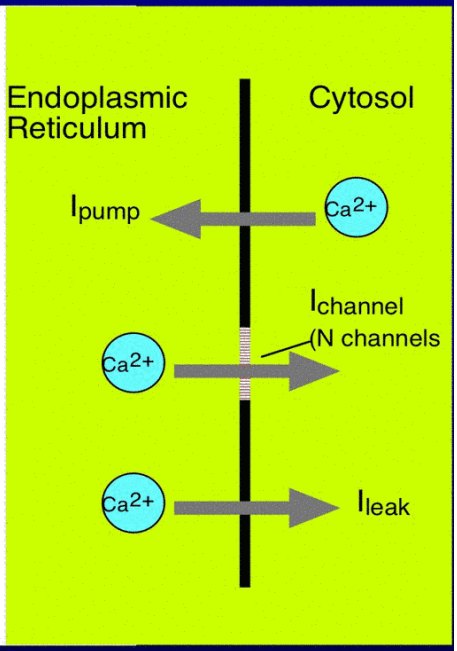
$$\frac{d[Ca^{2+}]}{dt} = I_{\text{channel}} + I_{\text{Pump}} + I_{\text{Leak}}$$

$$I_{\text{channel}} = -c_1 v_1 n_\infty^3 m_\infty^3 h^3 \left( [Ca^{2+}] - [Ca^{2+}]_{ER} \right)$$

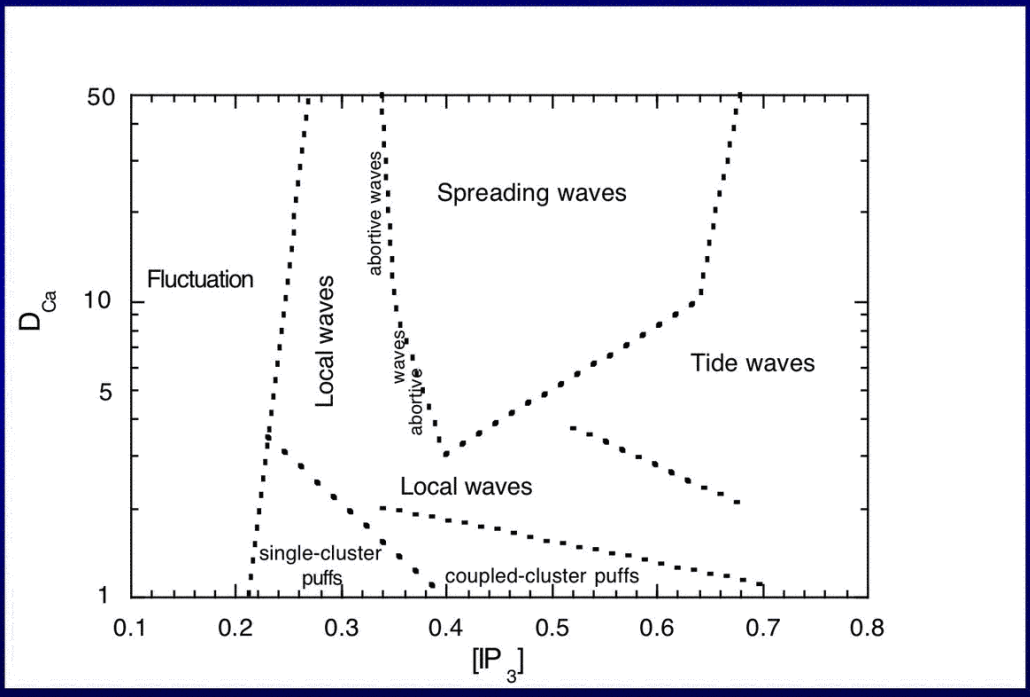
$$I_{\text{Pump}} = -\frac{v_3 [Ca^{2+}]^2}{k_3^2 + [Ca^{2+}]^2}$$

$$I_{\text{Leak}} = -c_1 v_2 \left( [Ca^{2+}] - [Ca^{2+}]_{ER} \right)$$

$$\frac{dh}{dt} = \alpha_h([IP_3])(1-h) - \beta_h([Ca^{2+}])h$$

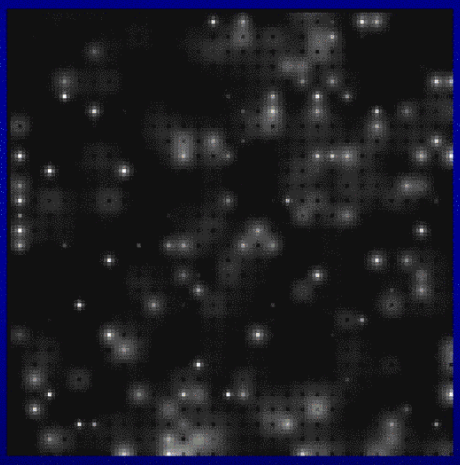



# Pattern Selection



# Calcium Patterns

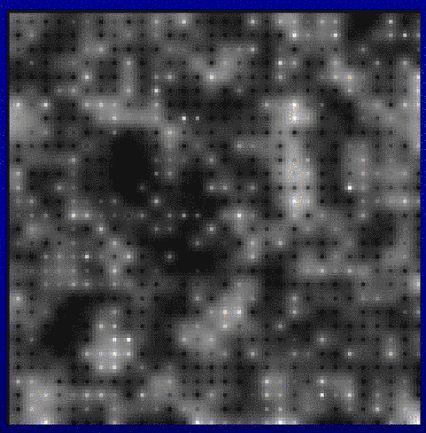
- Calcium Puffs



selection

# Calcium Patterns

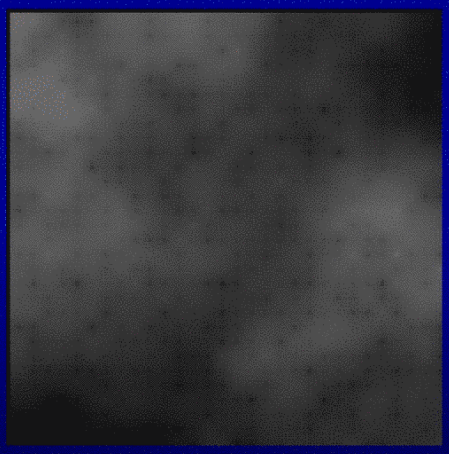
Local Waves



selection

# Calcium Patterns

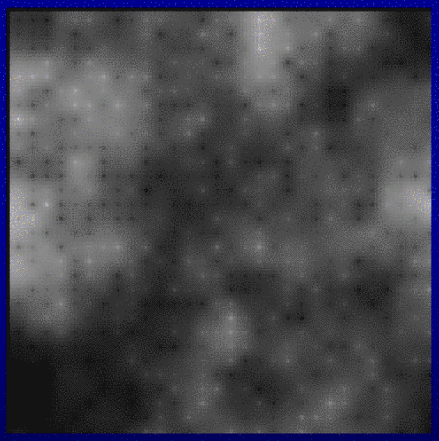
## Spreading Waves



selection

# Calcium Patterns

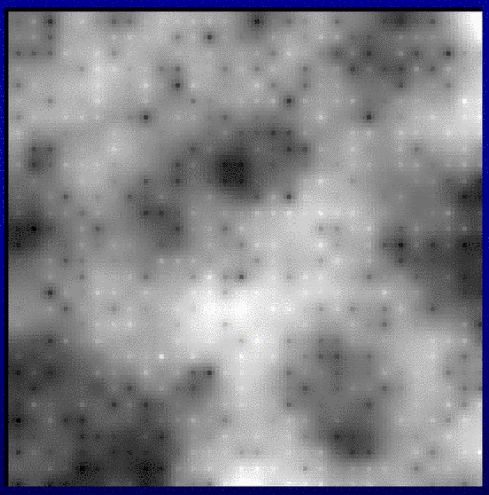
## Abortive Waves



selection

# Calcium Patterns

Tide Waves

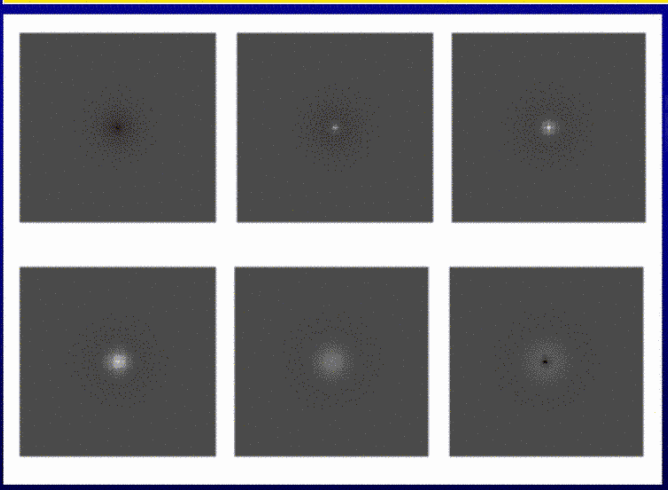


selection

# Additional stuff

# Single Cluster with $Ca^{2+}$ Diffusion

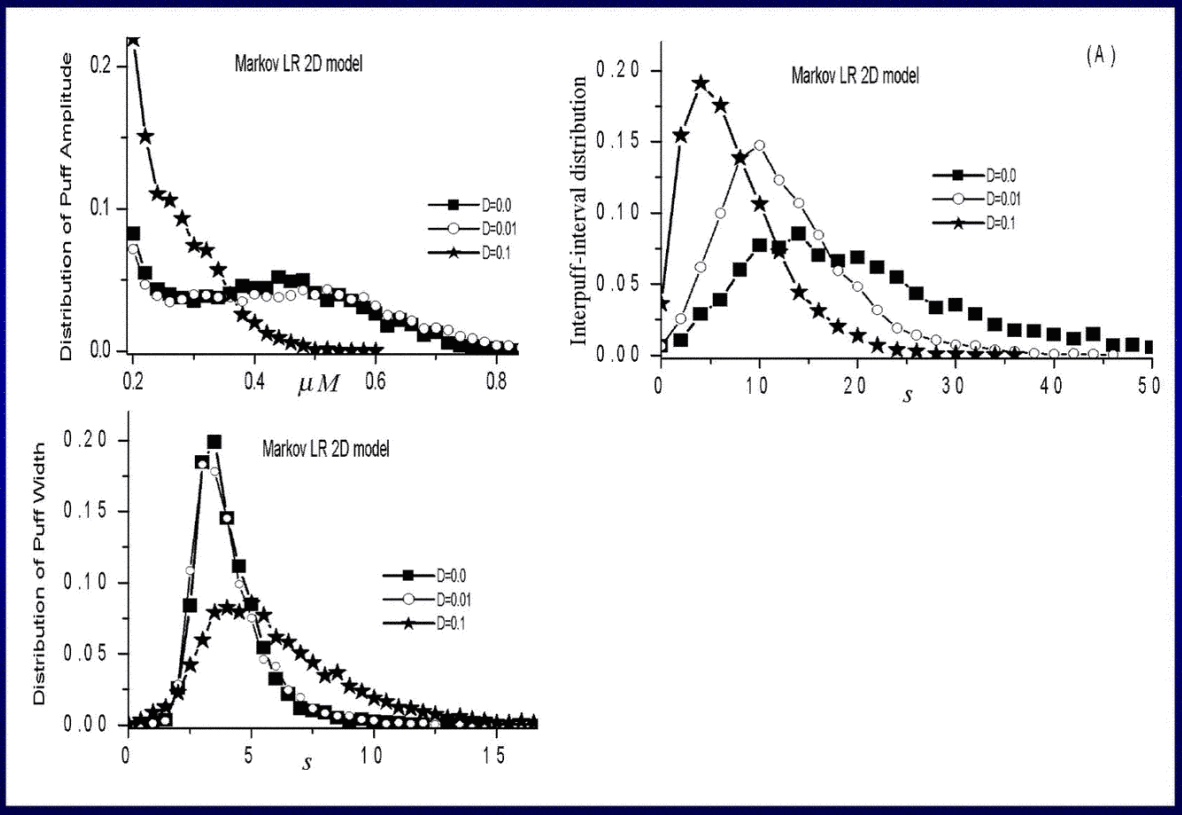
$$\frac{\partial c}{\partial t} = \left( c_1 m_\infty^3 n_\infty^3 \frac{N_{h-open}}{N} (c - c_{ER}) - c_1 v_2 (c - c_{ER}) - \frac{v_3 c^2}{k_3^2 + c^2} \right) \Xi(x)\Xi(y) + D\nabla^2 c$$



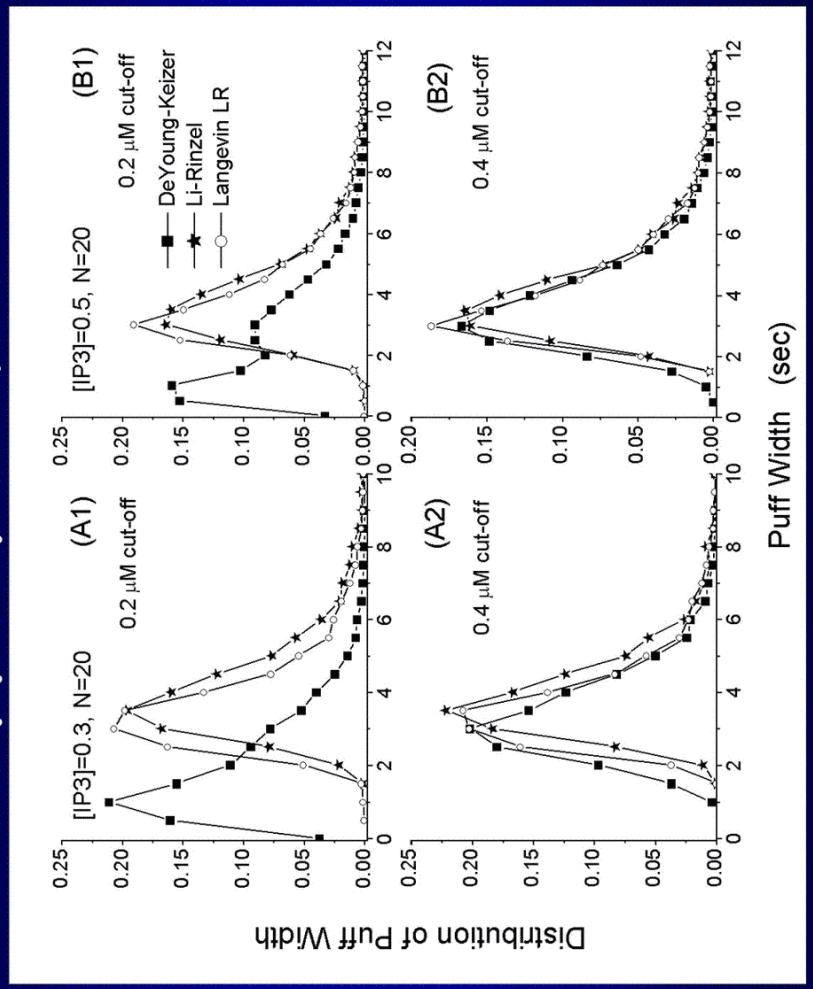
$$\Xi(x) = \begin{cases} 1 & \text{for} \quad -L/2 < x < L/2 \\ 0 & \text{for} \quad |x| > L/2 \end{cases}$$

N=20, $[IP_3]$=0.3μM
Size: 5μm x 5μm
L=0.1μm

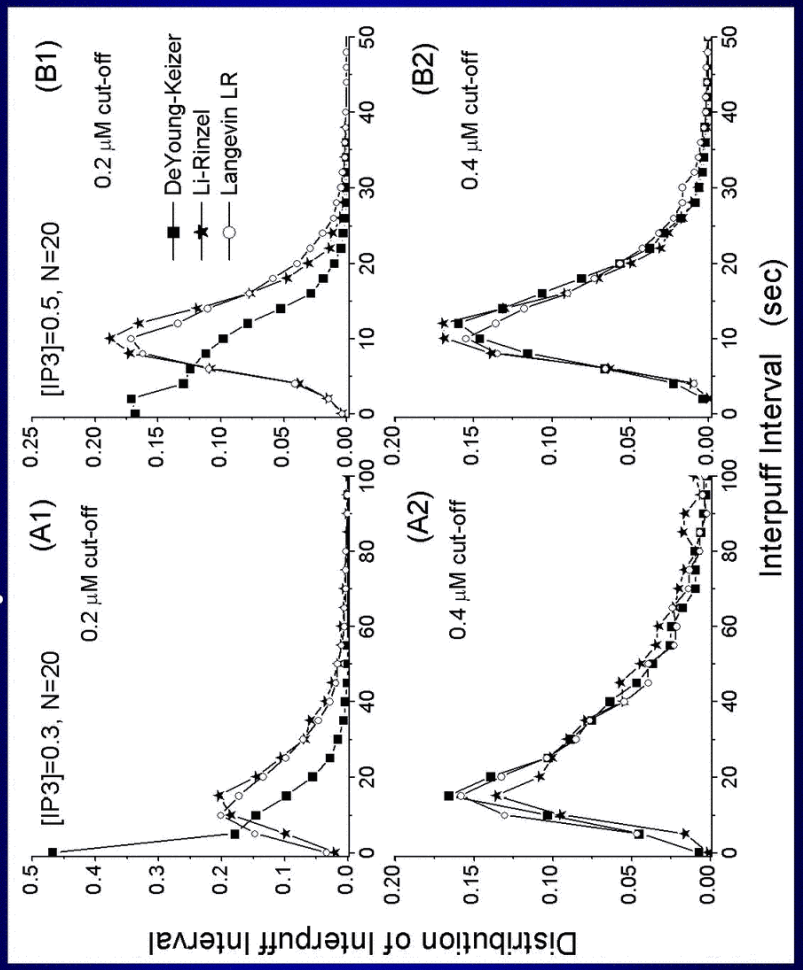
# Puff Characteristics



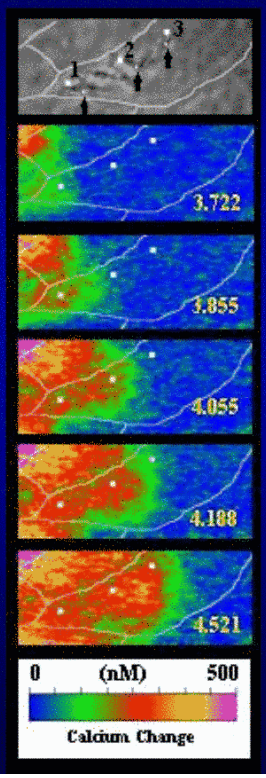
# Puff Lifetimes

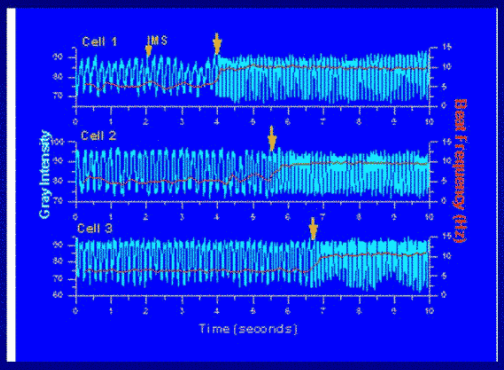



# Inter-puff Intervals



# Intra and Intercellular Waves

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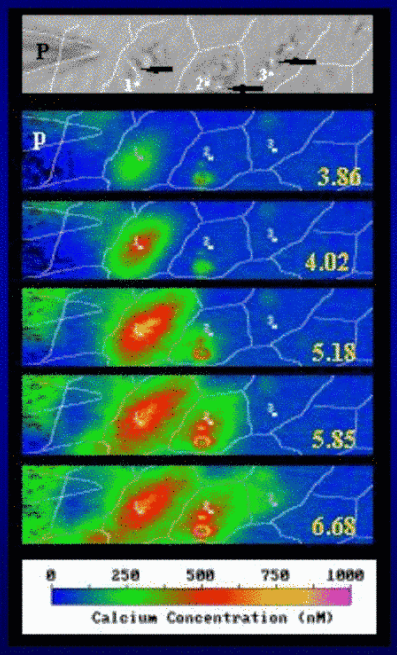


# Open Problems

- There are hot-spots (Xenopus oocyte, type-1 astrocytes).
- Mitochondria $Ca^{2+}$ handling.
- $Ca^{2+}$ is heavily buffered. Slow buffers require more complex model. (see also work by M. Falcke et al.)
- $Ca^{2+}$ dyes are $Ca^{2+}$ buffers!