Granule membranes play dice. The quantal nature of secretion

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To the memory of Bruno Bassan

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E. NITZANY, **I. HAMMEL, I. MEILIJSON, *JTB 2010***

I. HAMMEL, I. MEILIJSON, *JRSI 2012*

I. HAMMEL, I. MEILIJSON, *JRSI 2013*

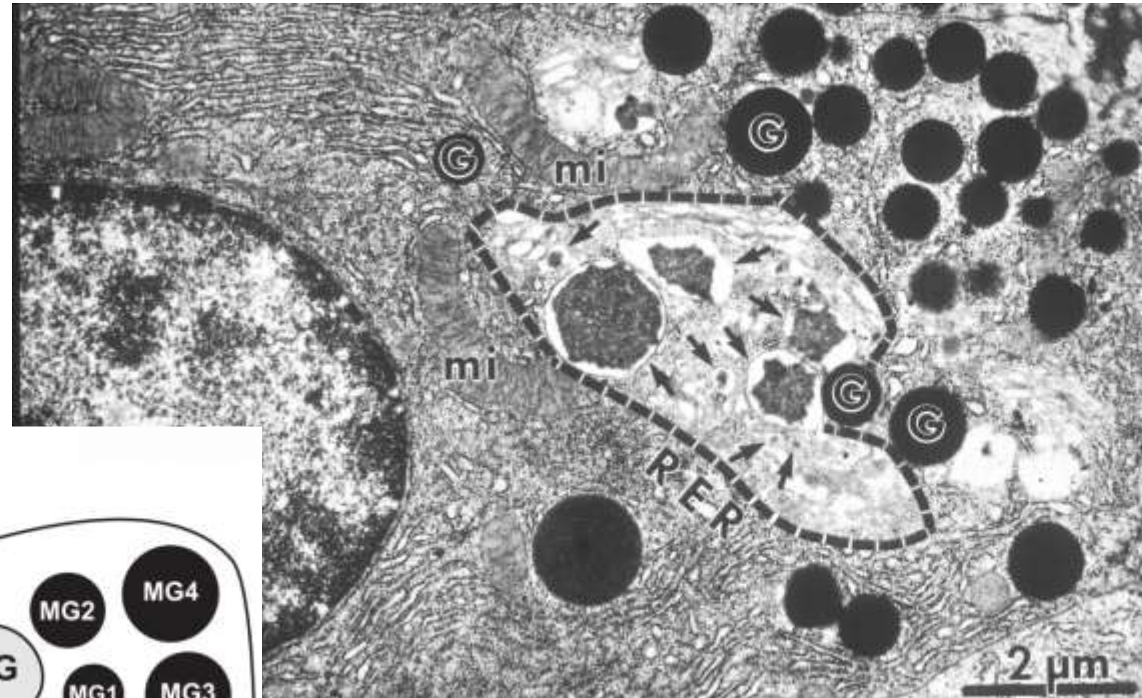
I. HAMMEL, M. KREPELOVA, I. MEILIJSON, *MRT 2014*

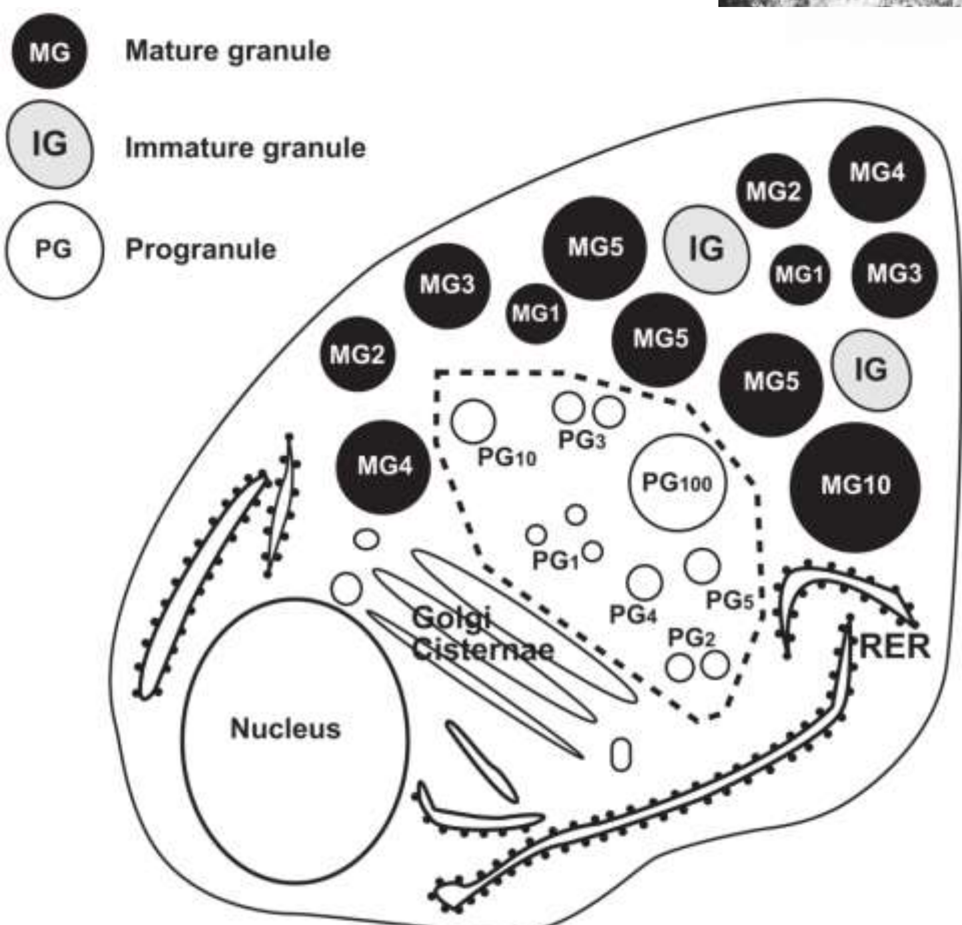
I. HAMMEL, I. MEILIJSON, *MI 2014*

I. HAMMEL, I. MEILIJSON, *NCB 2014*

I. HAMMEL, I. MEILIJSON, *D 2014*

D. AMIHAI, I. HAMMEL, I. MEILIJSON, J. TERKEL, *TO BE SUBMITTED*



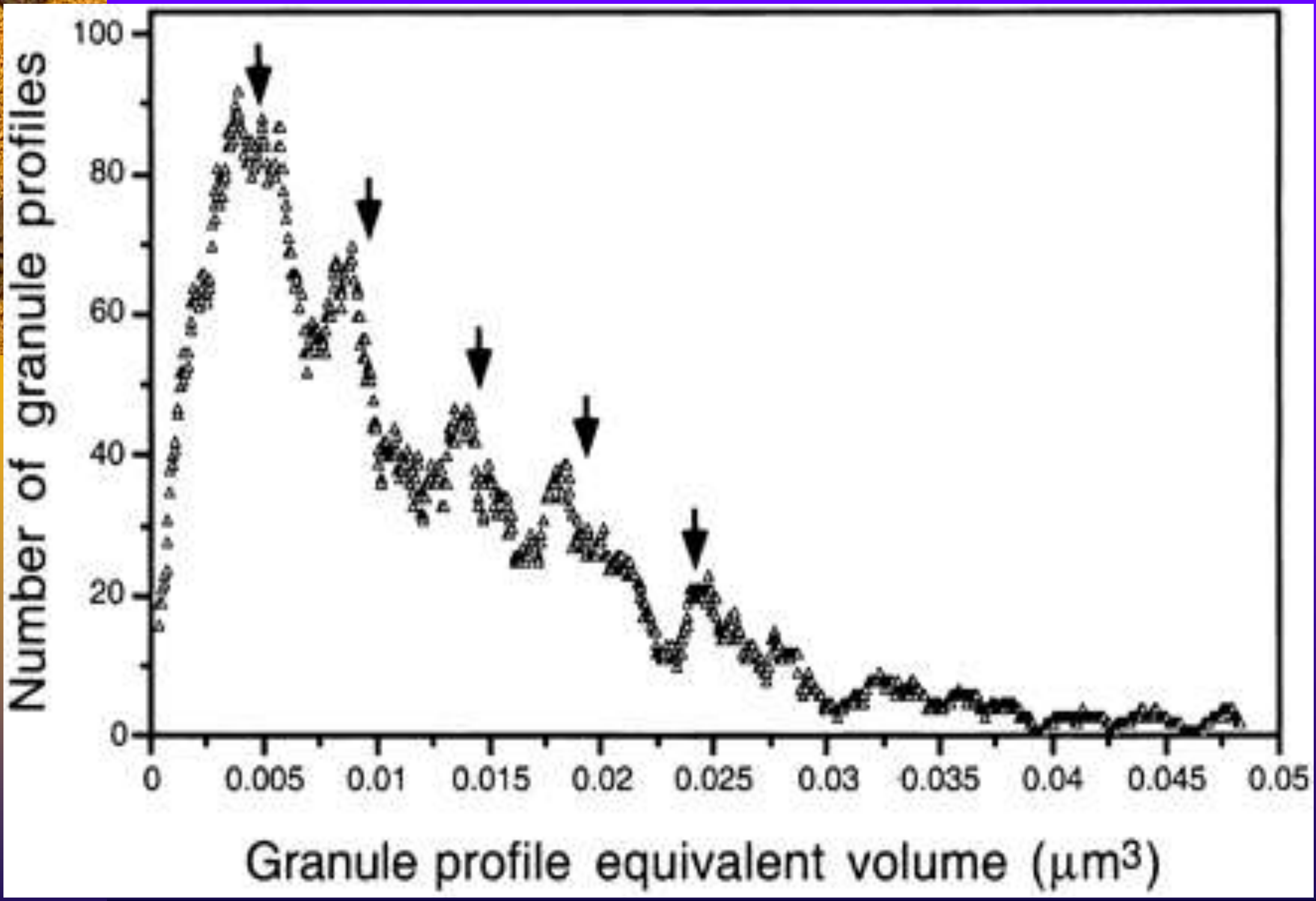


Pancreatic Acinar Cell

Two fundamental reasons for keeping granule inventory:

1. **Uncertainty of demand:** Granule stock is maintained as a buffer to meet uncertainty in demand by the extracellular environment.
2. **Lead time for production:** A source of supply during the lead time to produce granules of adaptive content.

Quantal nature of granular volume

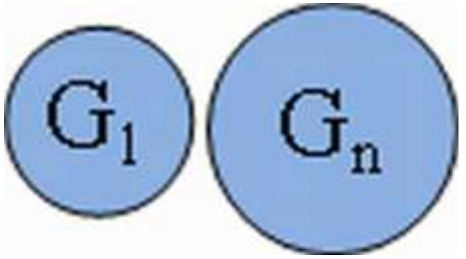


Most researchers hypothesize that granules stay as created and exit many-at-a-time

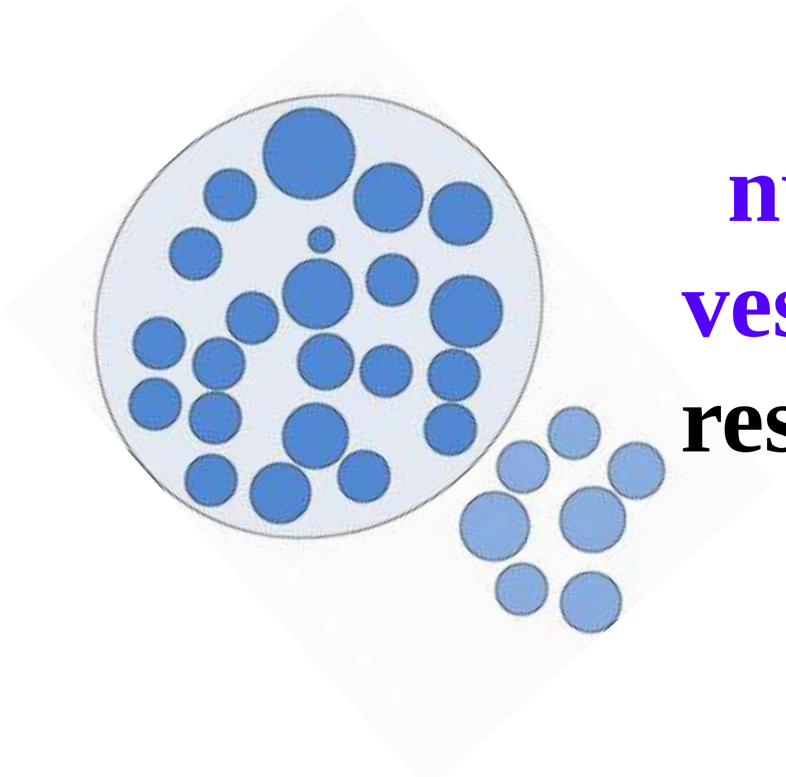
Bernard Katz (Nobel Prize 1970) group in England

Cope & Williams 1992 review

Is *Quantal size* the synaptic content of a single vesicle out of a heterogeneous pool?



$$G_n = nG_1$$

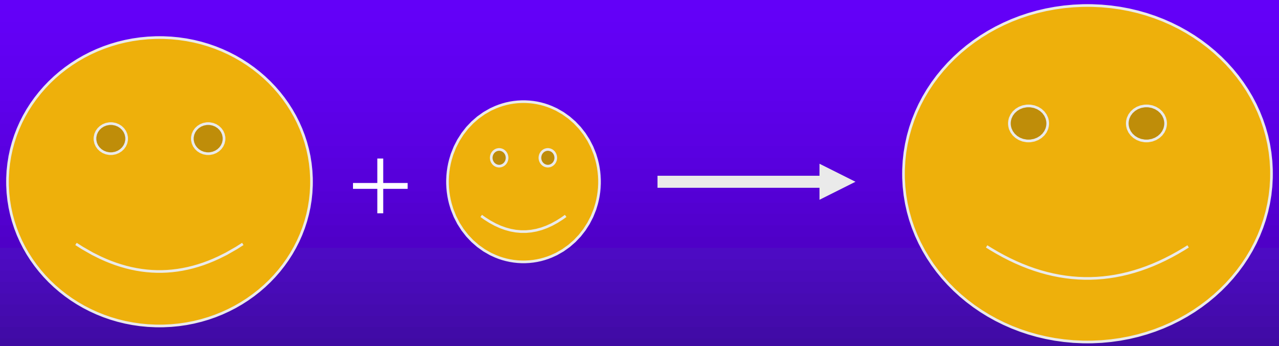


Is *Quantal content* the number of homogeneous vesicles released in a single response of cell activation?

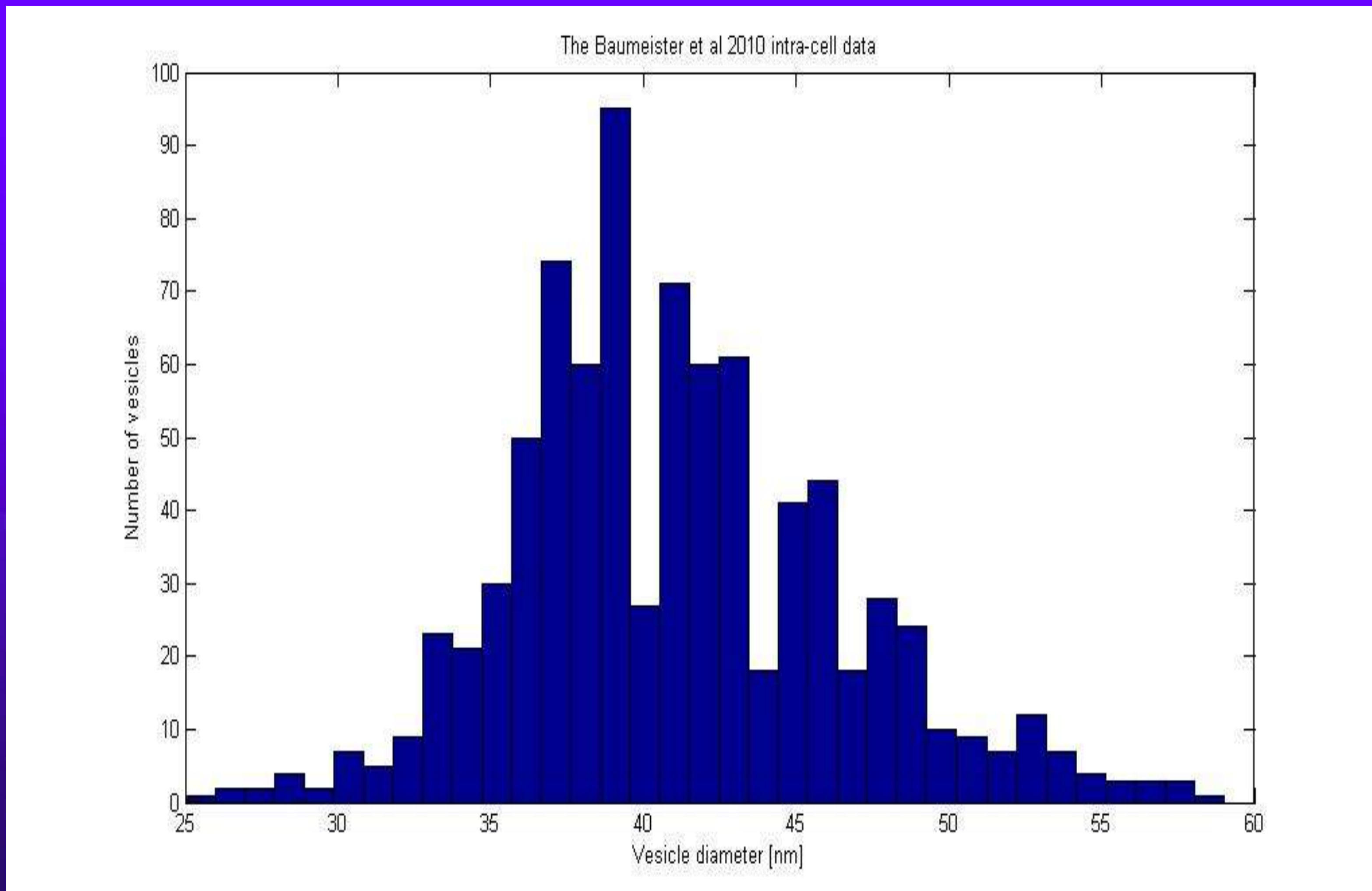
Granule Quantal Growth
addition of unit granule

All agree on Granule-Membrane fusion

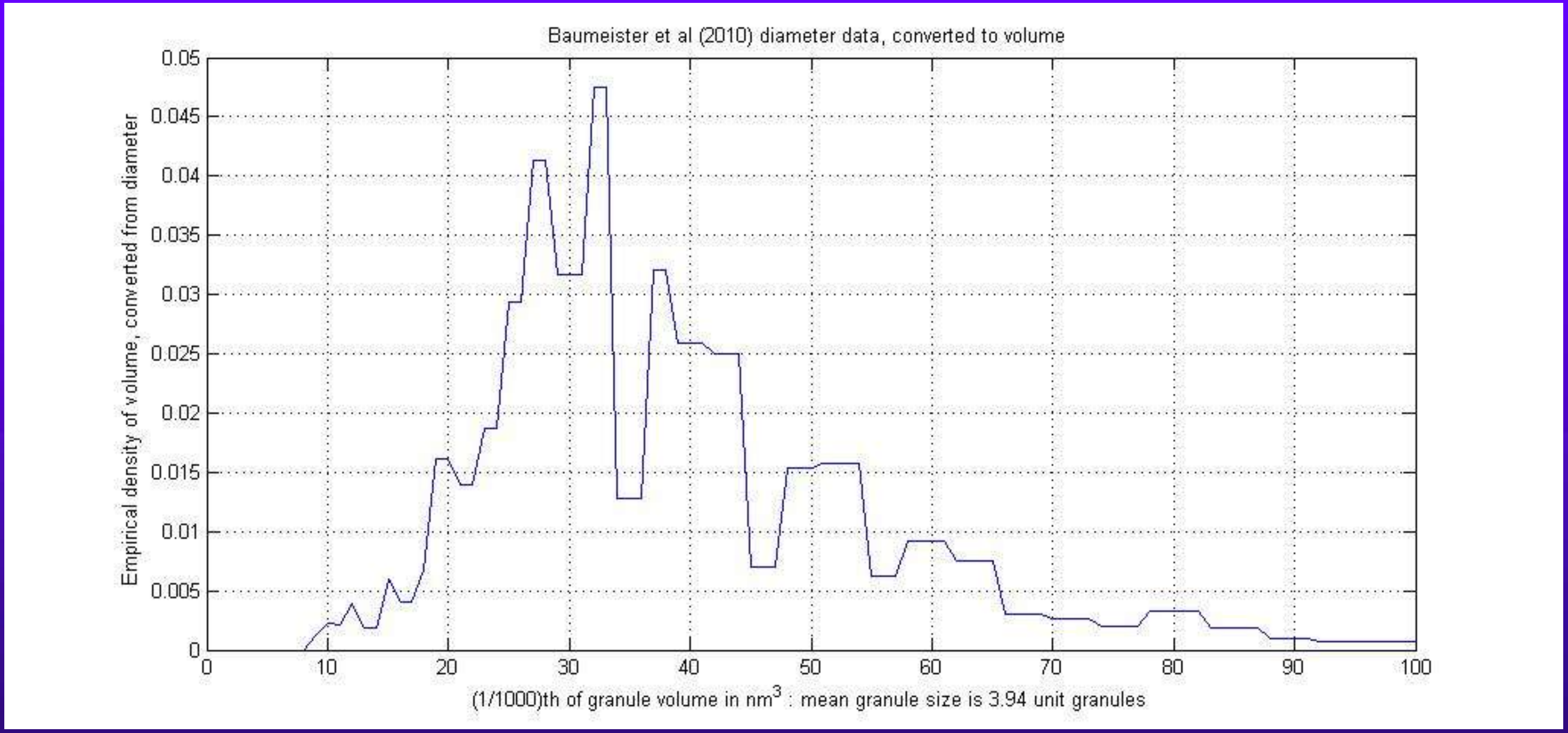
But not on Granule - Granule fusion



Baumeister et all 2010 diameter data



Histogram of volume data



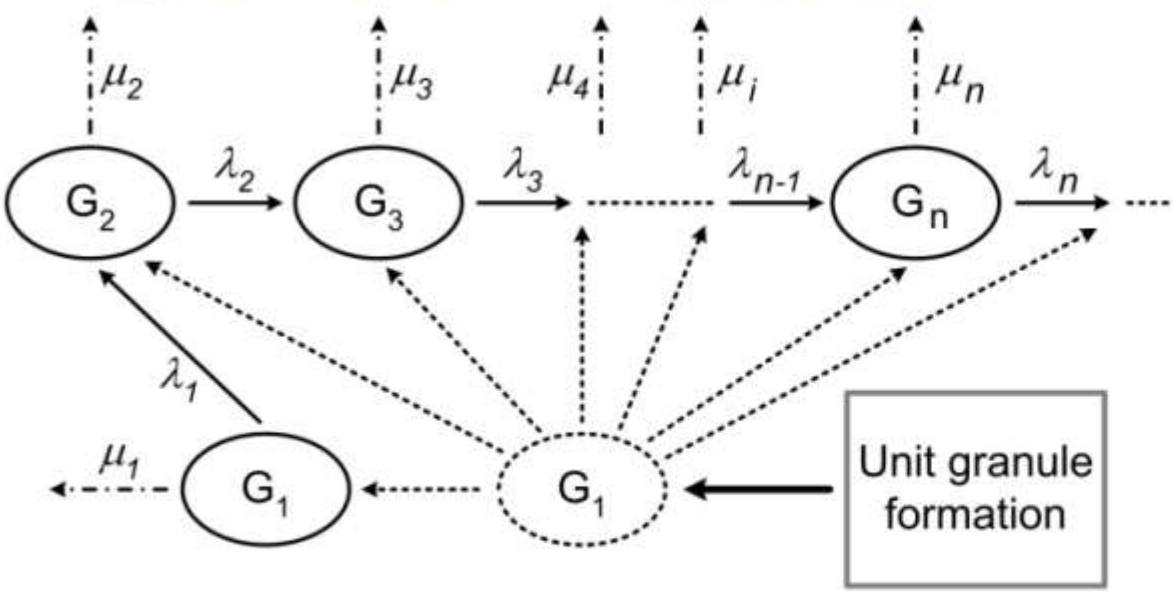
Markov Stationary Model

Markov process: The distribution of Future given History is summarized by Present state.

Granule Growth & Elimination : "state" is granule size

Probability of advancing from size n to size $n+1$ = $\frac{\lambda_n}{(\lambda_n + \mu_n)}$

$\lambda_n = \lambda n^{\beta} = \lambda n^{-(2/3)(K_\beta - 1)}$ $\mu_n = \mu n^{\gamma} = \mu n^{-(2/3)(K_\gamma - 1)}$

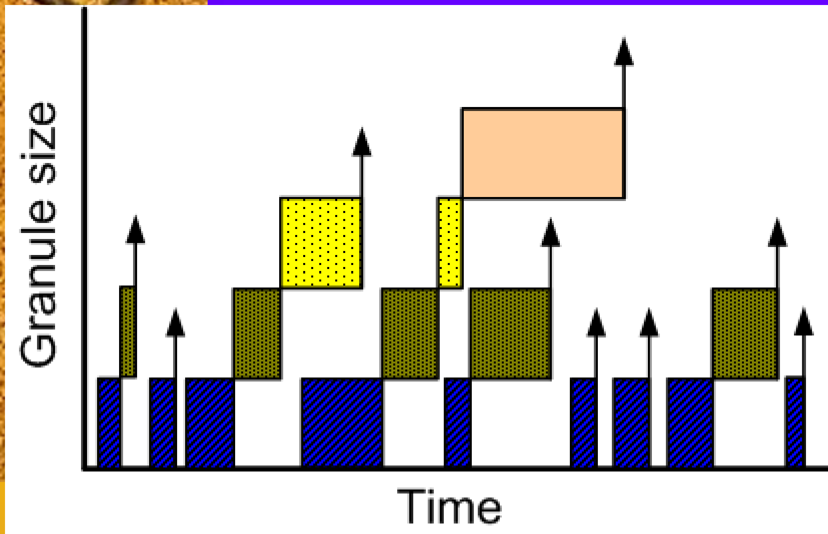


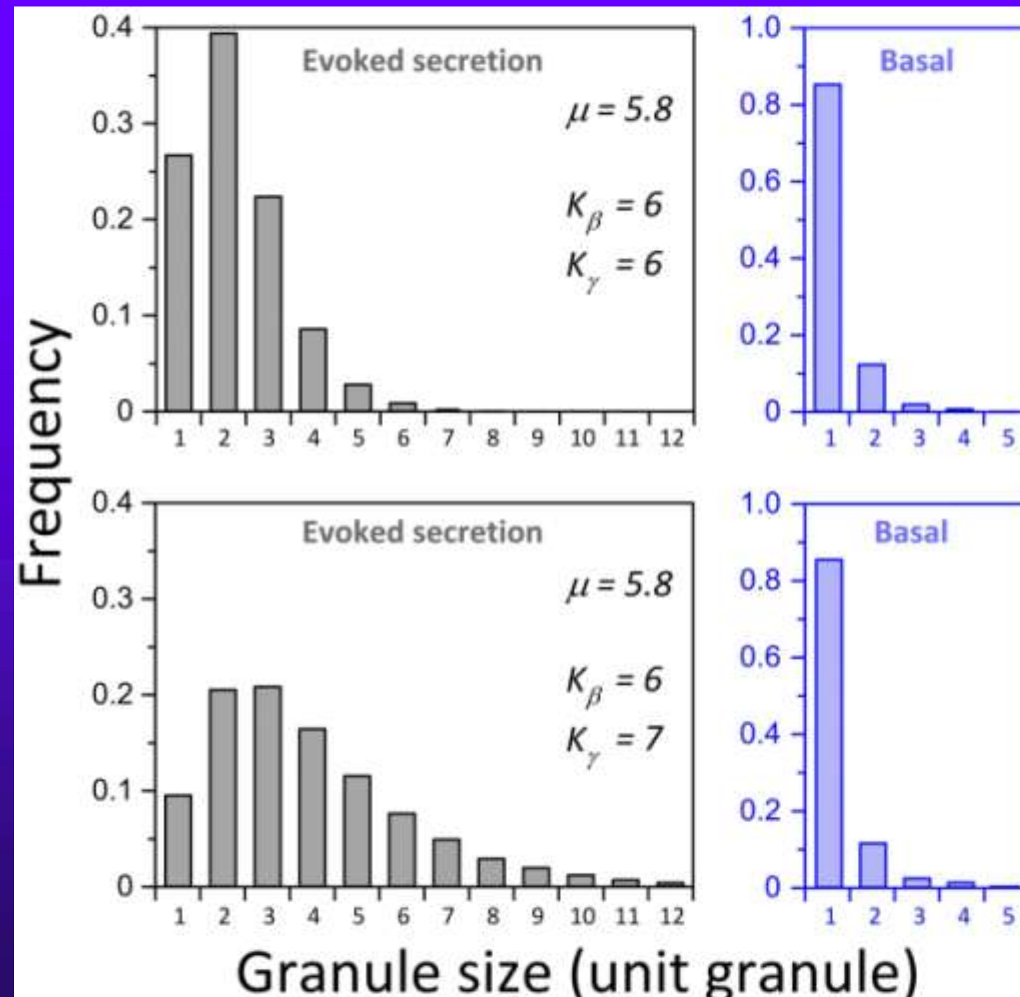
λ = addition

μ = elimination

Granule steady-state model

Stationary distribution ≠ Exit distribution



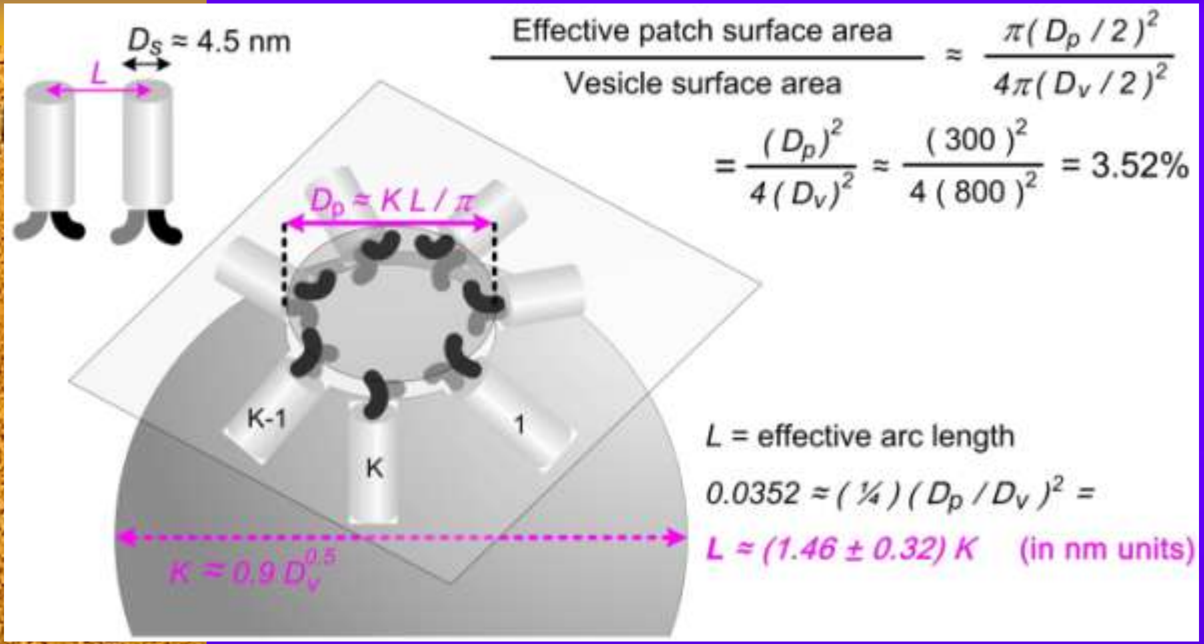


Ryan et al. (1997) used optical microscopy and fluorescent dyes to track the spontaneous and evoked vesicle release.

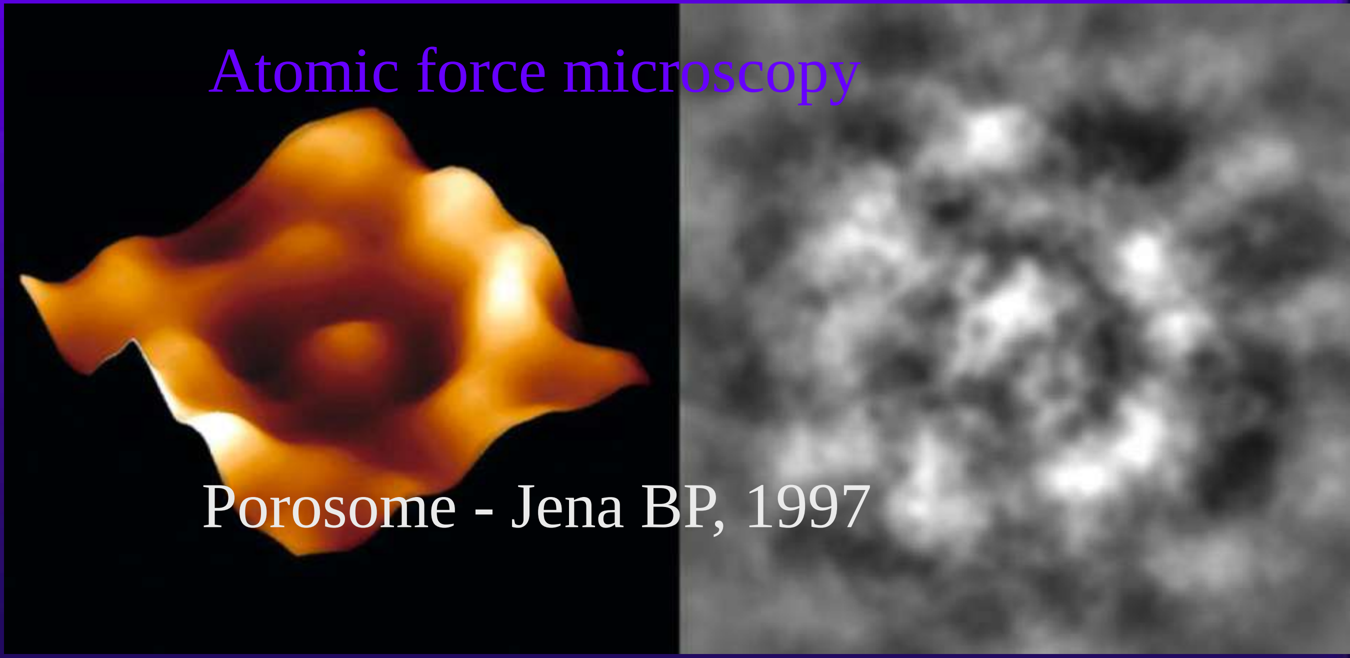
Three-parameter model

μ/λ , γ , β

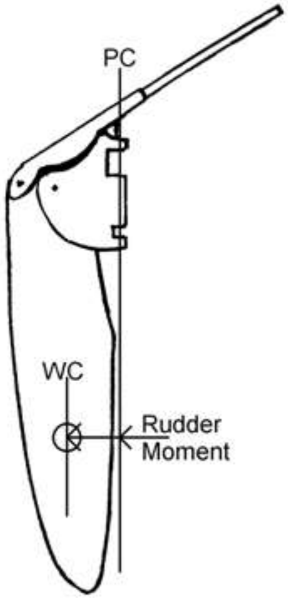
- Exit rate from n: $\mu * n^{\gamma}$
- Transition rate from n to n+1: $\lambda * n^{\beta}$
- Markov: independent exponentials, earliest wins
- Mean sojourn in n: $1/(\mu * n^{\gamma} + \lambda * n^{\beta})$
- Probability of exit $\mu * n^{\gamma} / (\mu * n^{\gamma} + \lambda * n^{\beta})$

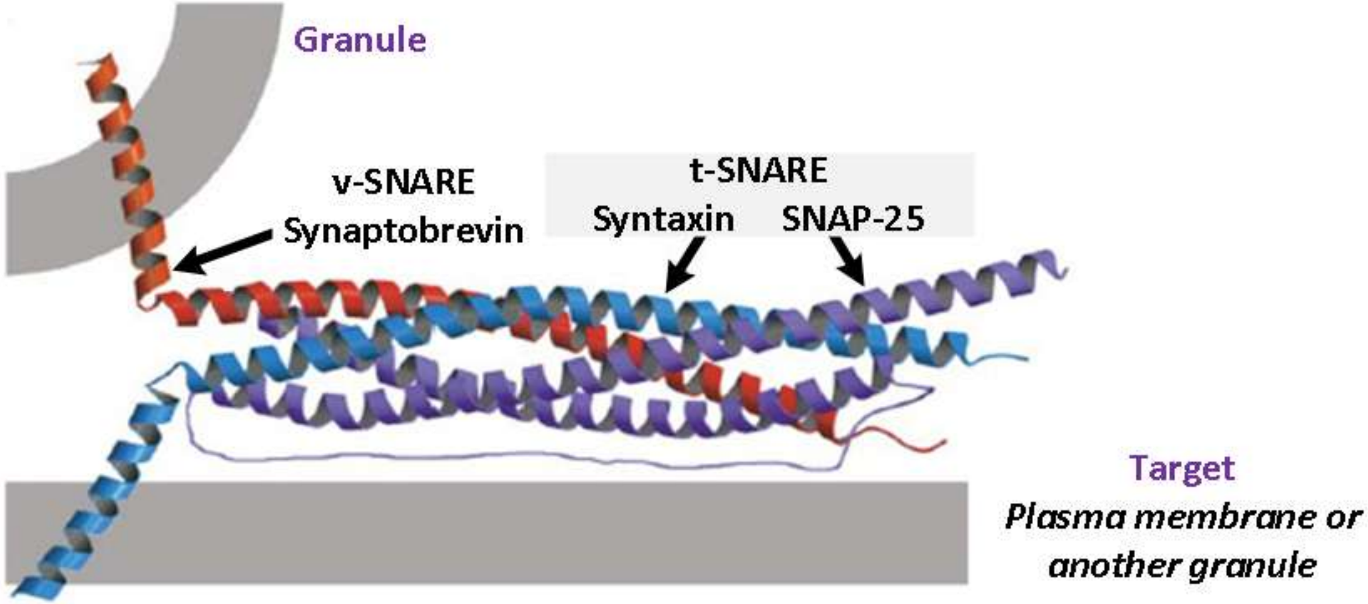


SNARE, ROSETTE, POROSOME



The rudder: The SNARE SYSTEM





Human subtlety will never devise an invention ***more beautiful, more simple or more direct*** *than does nature because in her inventions nothing is lacking, and nothing is superfluous.*

—Leonardo da Vinci

(http://www.brainyquote.com/quotes/authors/l/leonardo_da_vinci.html)

Rationale for rate shape SNARE: $\mu * n^{\gamma}$ and $\lambda * n^{\beta}$

N "rings" diffuse randomly on surface (area $n^{2/3}$) of granule, of which K must be close to each other and to K "hooks" in unit granule or membrane. Probability is of order

$(const/area)^{K-1} = const * n^{-(2/3)(K-1)} = \mu * n^{\gamma}$ or $\lambda * n^{\beta}$

Similar to Hua & Scheller PNAS 2001

Steady-state condition: flow into n+1 equals flow out of n+1

$$\mathbf{STAT(n)^{*}\ \lambda^{*}n^{\beta} = STAT(n+1)^{*}(\mu^{*}(n+1)^{\gamma} + \lambda^{*}(n+1)^{\beta})}$$

EXIT distribution

EXIT(n+) = GROW(1)*GROW(2)*...*GROW(n-1)

and

$$GROW(m) = \lambda * m^{\beta} / (\mu * m^{\gamma} + \lambda * m^{\beta})$$

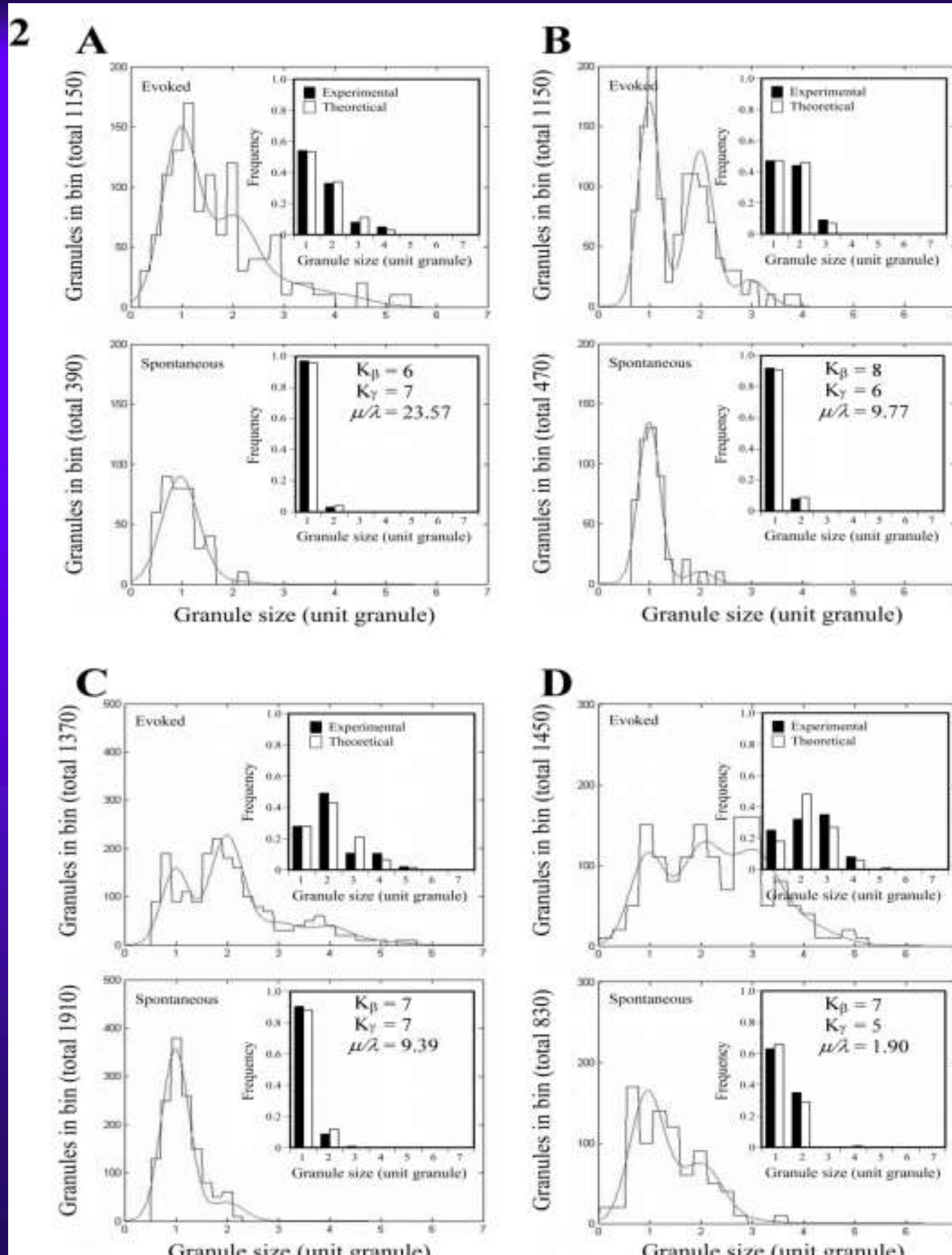
EXIT(n) = EXIT(n+) - EXIT((n+1)+)

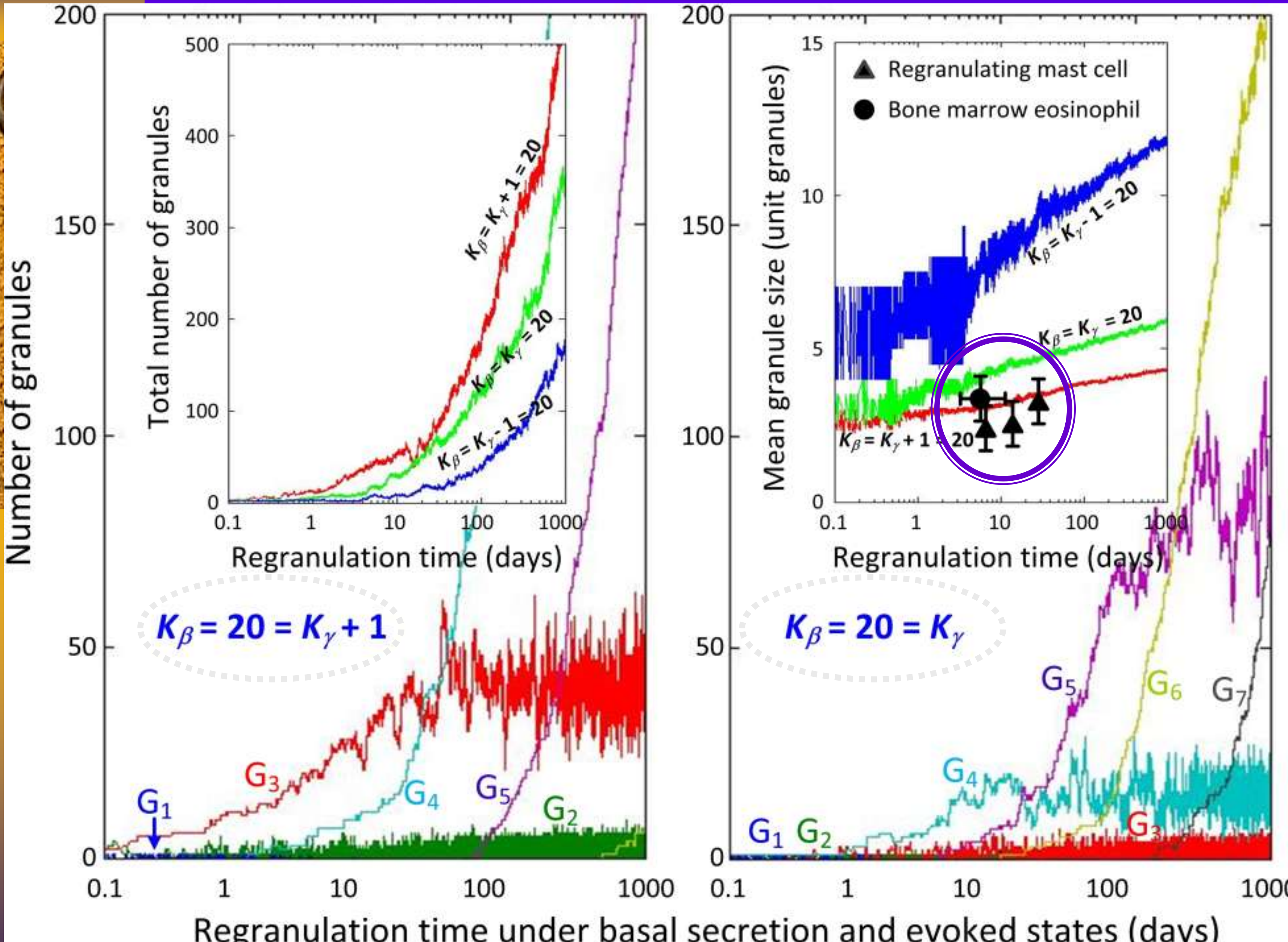
The role of γ

- STAT(n)/EXIT(n) = exp{-γ***log(n)-b(γ)}**
- **STAT** is exponential-type family with **EXIT** as case **γ=0**
- Perhaps evoked case needs one hook-ring pair!
- Reminder: **γ = -(2/3)(K-1)**

Statistical tools

- EM algorithm for Gaussian mixture with equally spaced means and variance
- MLE of **μ/λ , γ , β** based on evoked and spontaneous data
- Omnibus program for five parameters
- CUSUM detection of change-point
- Simulation study of transient behavior
- Euclidean Geometry considerations


2
A
Evoked
Experimental
Theoretical
Frequency
Granule size (unit granule)
Granules in bin (total 1150)
Spontaneous
Kβ = 6
Kγ = 7
μ/λ = 23.57
Granules in bin (total 390)
B
Evoked
Experimental
Theoretical
Granules in bin (total 1150)
Spontaneous
Kβ = 8
Kγ = 6
μ/λ = 9.77
Granules in bin (total 470)
C
Evoked
Granules in bin (total 1370)
Spontaneous
Kβ = 7
Kγ = 7
μ/λ = 9.39
Granules in bin (total 1910)
D
Evoked
Granules in bin (total 1450)
Spontaneous
Kβ = 7
Kγ = 5
μ/λ = 1.90
Granules in bin (total 830)
Granule size (unit granule)



Number of granules
Total number of granules
Regranulation time (days)
$K_\beta = K_\gamma + 1 = 20$
$K_\beta = K_\gamma = 20$
$K_\beta = K_\gamma - 1 = 20$
$K_\beta = 20 = K_\gamma + 1$
G_1
G_2
G_3
G_4
G_5
Mean granule size (unit granules)
Regranulating mast cell
Bone marrow eosinophil
$K_\beta = 20 = K_\gamma$
G_6
G_7
Regranulation time under basal secretion and evoked states (days)

Detecting that spontaneous secretion turned evoked secretion

- CUSUM - Statistical tool for detecting change-point in distribution
- Performance measured by Kullback-Leibler divergence KLD
- CUSUM calculations may use common action potential monitoring.

CUSUM of Page (1953)

- Declare change when log likelihood
- $S_n = \Sigma \log g(X_i)/f(X_i)$ exhibits draw-up
- $$S_n - \min_{m \le n} S_m$$
- of at least a pre-assigned size.

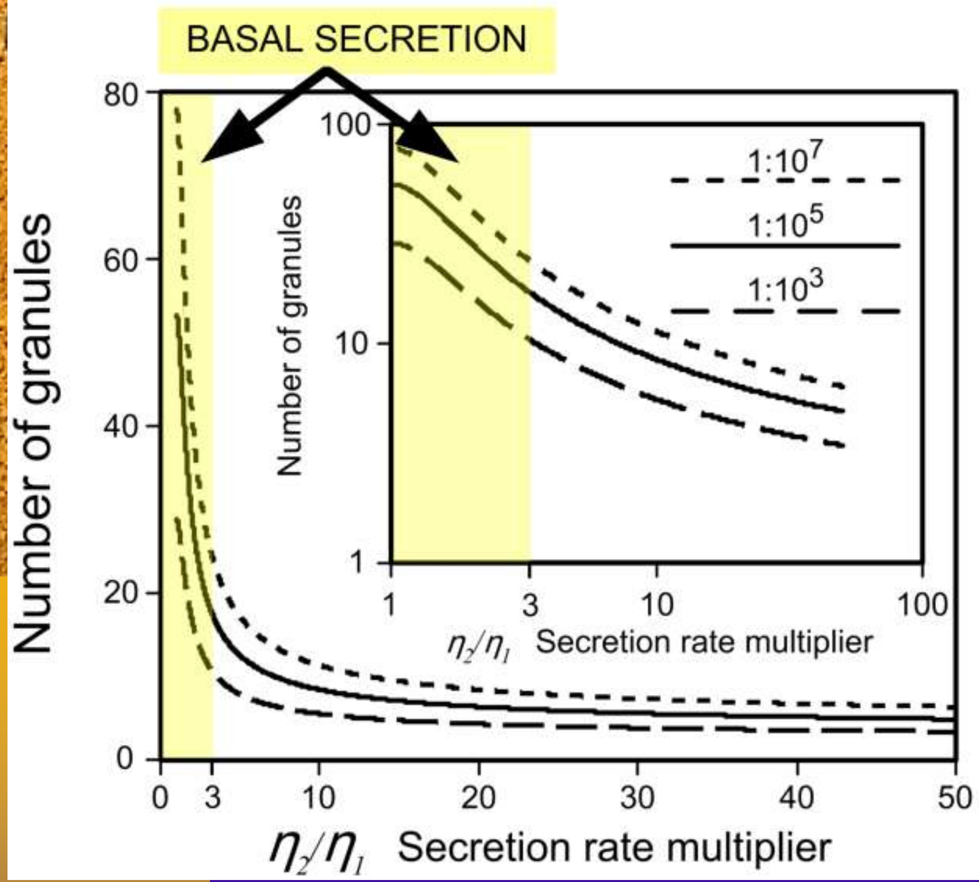
- $E_g[\log g(X)/f(X)] > 0$ *is KLD(g,f)*
- $E_f[\log g(X)/f(X)] < 0$ *is KLD(f,g)*

SUMMARY, FORMULA

- For log likelihoods $\rho=1$.
- *Putting all together, Mean Granules to Detection is*

$$\frac{\ln[MGFA]+\ln[KLD(E,S)+\eta_2/\eta_1-1-\ln(\eta_2/\eta_1)]}{KLD(S,E)+\eta_1/\eta_2-1-\ln(\eta_1/\eta_2)}$$

One word = 1 bit = 8-10 vesicles



If spontaneous secretion is steady at (the common rate of neurons) 1Hz, a false recognition alarm will be declared once every 15 minutes (10^3), once a day (10^5) or every four months (10^7).

Cellular communication: do we really need granule growth?

If the rate of secretion is increased

- by a small factor (≈2), granule size distribution plays a critical role
- by significant bursts (rate of increase = $\eta_2 / \eta_1 > 10$) the role of granule size distribution is minor.

The G&E model suggests that *granule polymerization has an advantage* for the *information gain* it achieves under *limited exocytosis* of a *small number of granules*.