



Lecture 3: Morphogen Gradients

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MSc Computational Biology 2019/20

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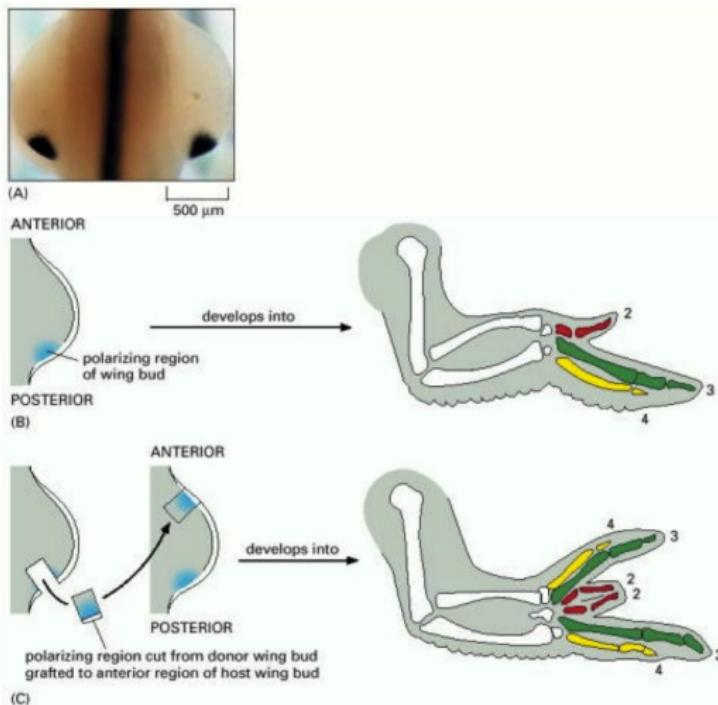
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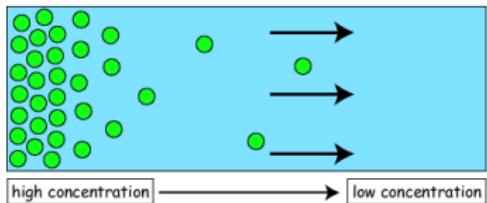
Previous Lecture

Patterning by Diffusible Reagents



Morphogen Transport by Diffusion

$$\frac{\partial c}{\partial t} = D \frac{\partial^2 c}{\partial x^2}; \quad 0 \leq x \leq L$$

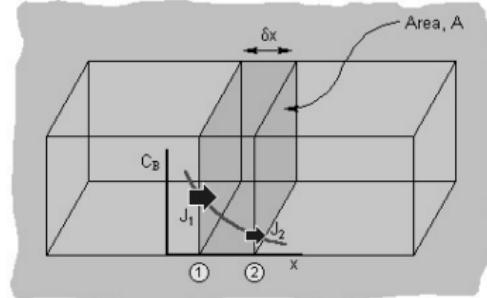


Fick's First Law

$$J = -D \frac{\partial c}{\partial x}$$

Fick's Second Law

$$\frac{\partial c}{\partial t} = -\frac{\partial J}{\partial x} = D \frac{\partial^2 c}{\partial x^2}$$



What model for Morphogen Gradients?

1D Domain : $0 \leq x \leq L$

$$PDE : \frac{\partial c}{\partial t} = D\Delta c - f(c)$$

$$IC : c(x > 0, 0) = 0$$

Degradation Term $f(c)$

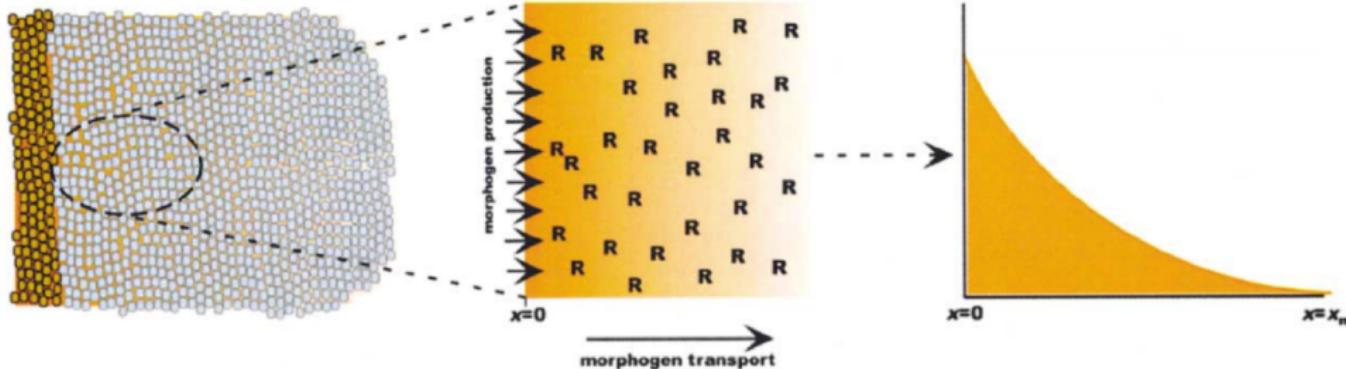
- none \Rightarrow linear gradient
- linear \Rightarrow exponential gradient
- non-linear \Rightarrow powerlaw gradient

Boundary Conditions

- Fixed concentration at both boundaries, i.e. $c(0, t) = c_0$ & $c(L) = 0$
- Fixed concentration at source, zero concentration at infinity, $c(0, t) = c_0$ & $c(x \rightarrow \infty) = 0$
- Flux boundary conditions, i.e.

$$\left. \frac{\partial c}{\partial x} \right|_{x=0} = -j \text{ & } \left. \frac{\partial c}{\partial x} \right|_{x=L} = 0$$

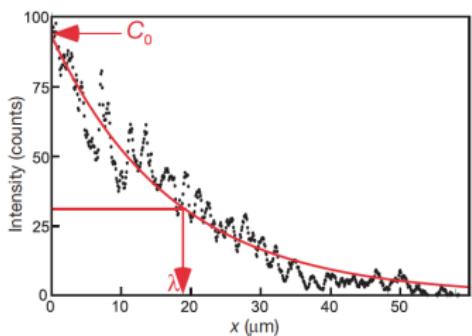
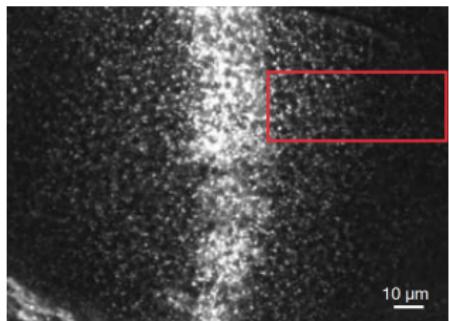
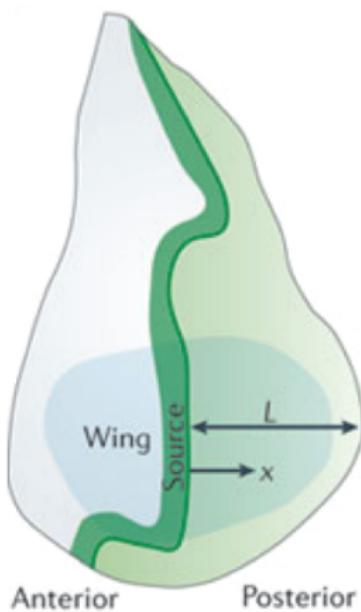
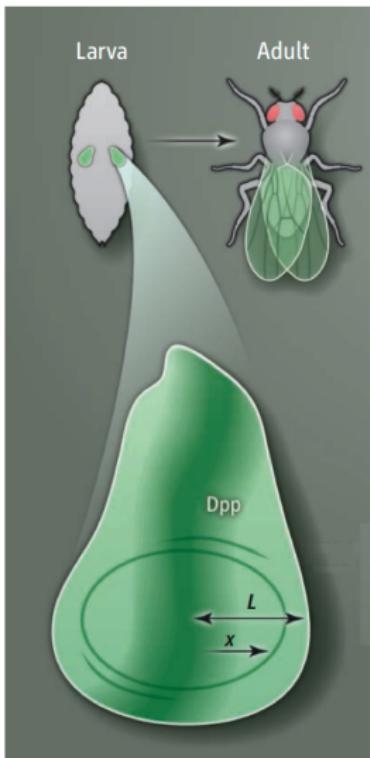
STANDARD MODEL: boundary at infinity



Steady-state solution:

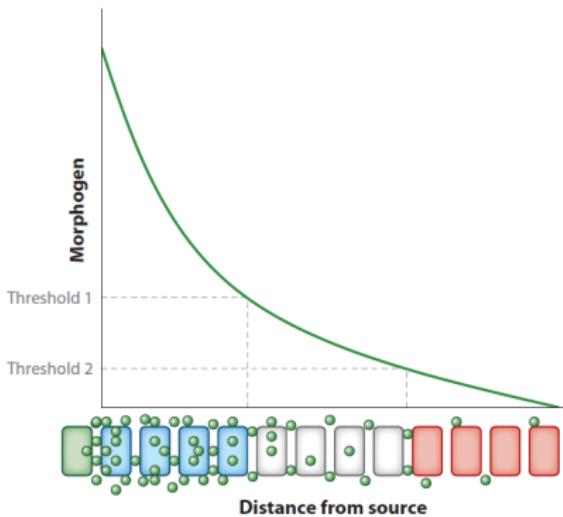
$$\begin{aligned}
 PDE : \quad & \frac{\partial c}{\partial t} = D \Delta c - kc; \quad 0 \leq x \leq L & c(x) &= c_0 \exp\left(-\frac{x}{\lambda}\right) \\
 IC : \quad & c(x, 0) = 0 & \lambda &= \sqrt{\frac{D}{k}} \\
 BC : \quad & c(0, t) = c_0 \quad c(x \rightarrow \infty) = 0
 \end{aligned}$$

The Dpp Gradient is of exponential shape



Morphogen Gradient Read-Out

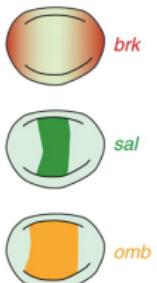
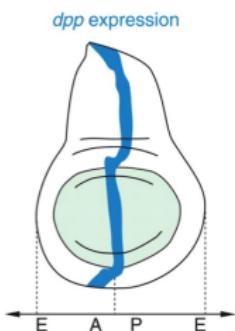
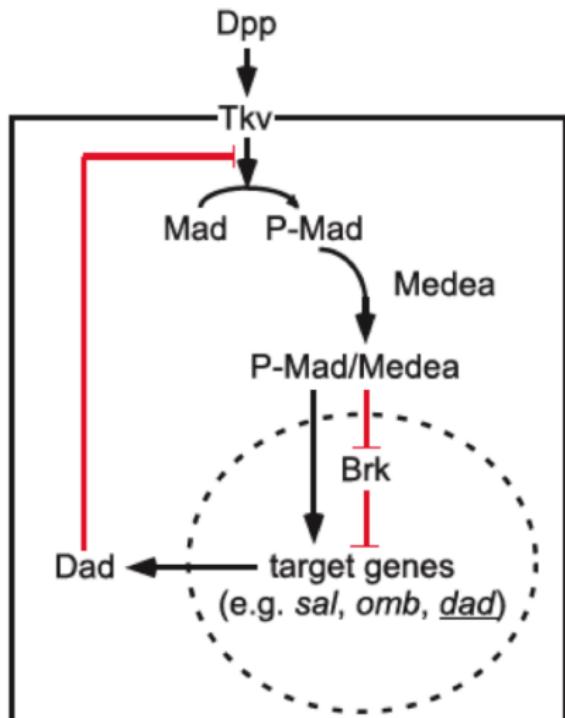
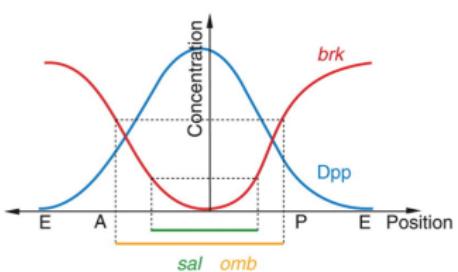
The French Flag Model (Lewis Wolpert)



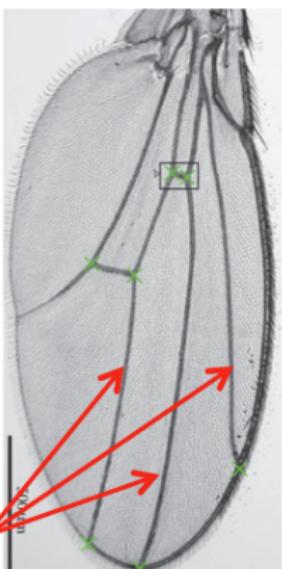
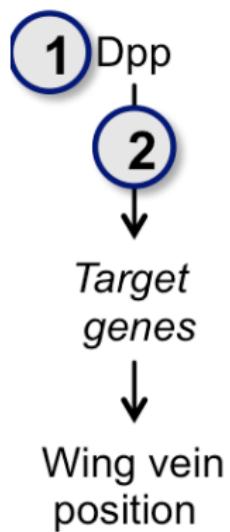
The French Flag model can still be used also with an exponential gradient:

$$c = c_0 \exp(-x/\lambda)$$

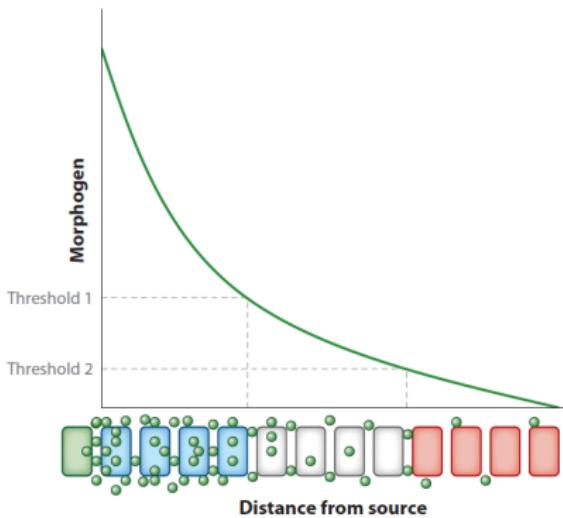
Read-Out of the Dpp Gradient

A Wing imaginal disc*dpp expression***B** Dpp gradient

Dpp Gradient determines Vein Positions



How accurate do gradients need to be?



To sense a threshold signal, two neighbour cells must be able to respond differently to the difference in input signal.

Cells average the signal over the entire cell surface, and over a time interval, Δt .

Challenges to Read-Out Precision

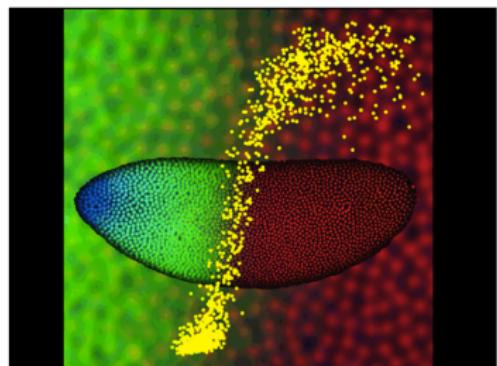
Challenges to Read-Out Precision

I. Noise

- 1 Thermal Ligand Fluctuations
- 2 Ligand-Receptor Binding Noise
- 3 Gradient Variability (source, transport, turn-over)

II. Changes in the Gradient Shape

III. Tissue Growth / Size



Changes in the Source

Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D\Delta c - kc$$

$$IC : c(x, 0) = 0; \quad x > 0$$

$$BC : c(0, t) = c_0$$

$$c(x \rightarrow \infty) = 0$$

Changes in the Source

Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D \Delta c - kc$$

$$IC : c(x, 0) = 0; \quad x > 0$$

$$BC : c(0, t) = c_0$$

$$c(x \rightarrow \infty) = 0$$

Exp Steady-state solution:

$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right)$$

$$\lambda = \sqrt{\frac{D}{k}}$$

Changes in the Source

Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D \Delta c - kc$$

$$IC : c(x, 0) = 0; \quad x > 0$$

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Exp Steady-state solution:

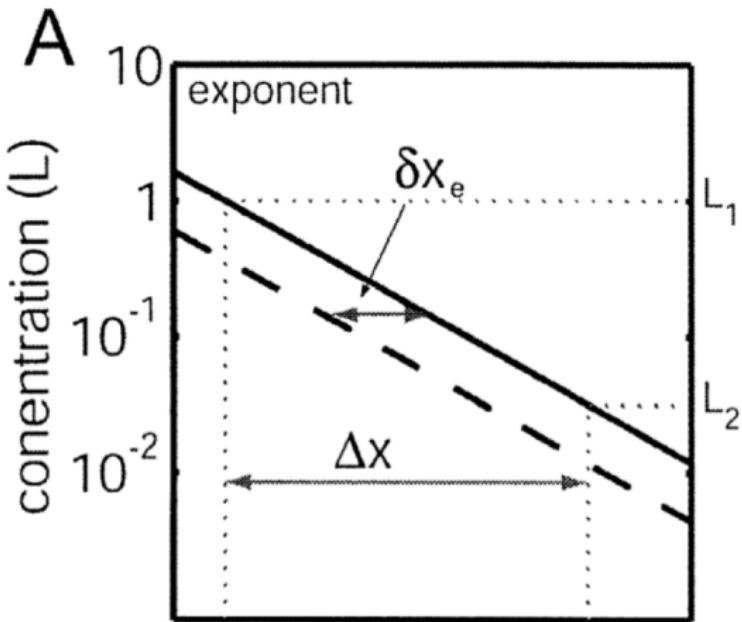
$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right)$$

$$\lambda = \sqrt{\frac{D}{k}}$$

Read-Out Shift:

Now consider a change in the source from c_0 to c_0^* . How large is the shift in the read-out position?

Impact of a Change in the Ligand Source



$$\frac{\partial L}{\partial t} = D \nabla^2 L - \alpha L$$

$$L = L_0 \exp(-x/\Delta_d)$$

$$\Delta_d = (D/\alpha)^{1/2}$$

Uniform decay (UD) does not provide robustness to fluctuations in the source L_0 .

Impact of a Change in the Ligand Source

We now want to calculate the distance

$$\Delta x = x_\theta^* - x_\theta$$

between the positions where the gradient reaches the threshold concentration c_θ with either c_0 or c_0^* :

$$\begin{aligned} c_\theta &= c_0 \exp\left(-\frac{x_\theta}{\lambda}\right) \\ c_\theta &= c_0^* \exp\left(-\frac{x_\theta^*}{\lambda}\right). \end{aligned}$$

Impact of a Change in the Ligand Source

$$c_\theta = c_0 \exp\left(-\frac{x_\theta}{\lambda}\right) = c_0^* \exp\left(-\frac{x_\theta^*}{\lambda}\right)$$

$$\frac{c_0^*}{c_0} = \exp\left(-\frac{x_\theta}{\lambda} + \frac{x_\theta^*}{\lambda}\right)$$

$$\Delta x = x_\theta^* - x_\theta = \lambda \ln\left(\frac{c_0^*}{c_0}\right)$$

Impact of a Change in the Ligand Source

$$c_\theta = c_0 \exp\left(-\frac{x_\theta}{\lambda}\right) = c_0^* \exp\left(-\frac{x_\theta^*}{\lambda}\right)$$

$$\frac{c_0^*}{c_0} = \exp\left(-\frac{x_\theta}{\lambda} + \frac{x_\theta^*}{\lambda}\right)$$

$$\Delta x = x_\theta^* - x_\theta = \lambda \ln\left(\frac{c_0^*}{c_0}\right)$$

Read-Out Shift:

$$\Delta x = \lambda \ln \frac{c_0^*}{c_0}$$

Summary Changes in the Source

Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D \Delta c - kc$$

$$IC : c(x, 0) = 0; \quad x > 0$$

$$BC : c(0, t) = c_0$$

$$c(x \rightarrow \infty) = 0$$

Exp Steady-state solution:

$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right)$$

$$\lambda = \sqrt{\frac{D}{k}}$$

Read-Out Shift:

$$\Delta x = x^* - x = \lambda \ln \frac{c_0^*}{c_0}$$

Impact of non-linear decay

Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D\Delta c - kc$$

Non-Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D\Delta c - kc^{\textcolor{red}{n}}$$

$$IC : c(x, 0) = 0; \quad x > 0$$

$$BC : c(0, t) = c_0 \quad c(x \rightarrow \infty) = 0$$

Impact of non-linear decay

Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D\Delta c - kc$$

Non-Linear Degradation

$$PDE : \frac{\partial c}{\partial t} = D\Delta c - kc^{\textcolor{red}{n}}$$

$$IC : c(x, 0) = 0; \quad x > 0$$

$$BC : c(0, t) = c_0 \quad c(x \rightarrow \infty) = 0$$

Exp Steady-state solution:

$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right); \quad \lambda = \sqrt{\frac{D}{k}}$$

Impact of non-linear decay

Linear Degradation

$$PDE : \quad \frac{\partial c}{\partial t} = D\Delta c - kc$$

Non-Linear Degradation

$$PDE : \quad \frac{\partial c}{\partial t} = D\Delta c - kc^{\textcolor{red}{n}}$$

$$IC : \quad c(x, 0) = 0; \quad x > 0$$

$$BC : \quad c(0, t) = c_0 \quad c(x \rightarrow \infty) = 0$$

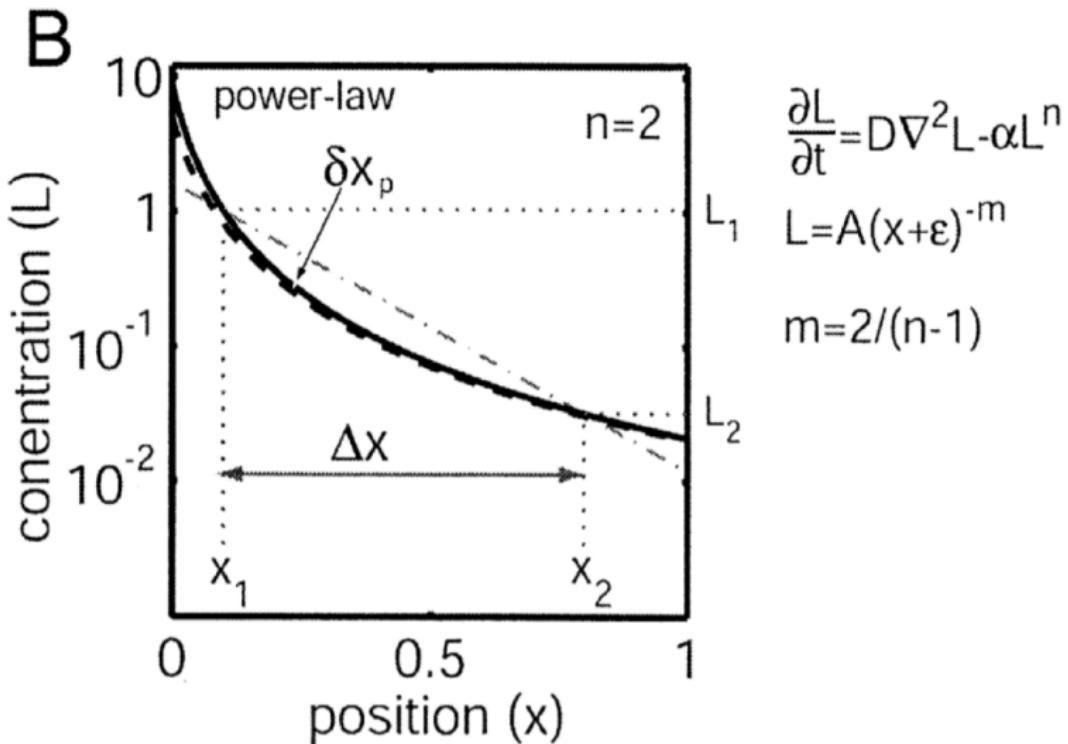
Exp Steady-state solution:

$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right); \quad \lambda = \sqrt{\frac{D}{k}}$$

Power-law Steady-state solution:

$$c(x) = A \frac{1}{(x + \epsilon)^m}; \quad m = \frac{2}{n - 1}$$

Self-enhanced decay (SEC) of the morphogen



Gradient Shift

Exp Steady-state solution:

$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right); \quad \lambda = \sqrt{\frac{D}{k}}$$

Gradient Shift

$$\Delta x = x^* - x = \lambda \ln \frac{c_0^*}{c_0}$$

Power-law Steady-state solution:

$$c(x) = A \frac{1}{(x + \epsilon)^m}; \quad m = \frac{2}{n - 1}$$

Gradient Shift

Consider simplified power-law equation

$$\begin{aligned}
 c_\theta(x) &= A \cdot (x_\theta + \epsilon)^{-m} = A^* \cdot (x_{\theta^*} + \epsilon^*)^{-m} \\
 \frac{A^*}{A} &= \left(\frac{x_\theta + \epsilon}{x_{\theta^*} + \epsilon^*} \right)^{-m} \\
 \ln \left(\frac{x_{\theta^*} + \epsilon^*}{x_\theta + \epsilon} \right) &= \frac{1}{m} \ln \left(\frac{A^*}{A} \right) \\
 \ln (x_{\theta^*} + \epsilon^*) - \ln (x_\theta + \epsilon) &= \frac{1}{m} \ln \left(\frac{A^*}{A} \right)
 \end{aligned}$$

compared to

$$\Delta x = x_{\theta^*}^* - x_\theta = \lambda \ln \left(\frac{c_0^*}{c_0} \right)$$

Gradient Shift

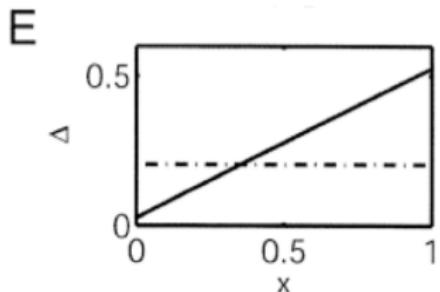
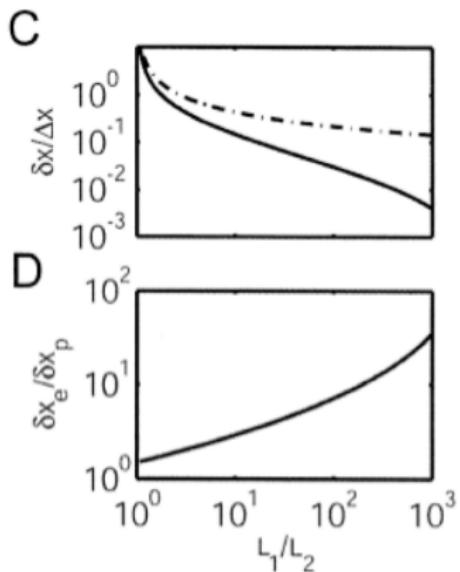
We can rewrite the power-law equation also as

$$c(x) = c_0 \cdot x^{-m} = c_0 \cdot \exp(-m \ln(x)) = c_0 \cdot \exp(-x/\lambda)$$

with spatially varying

$$\lambda = \frac{x}{-m \ln(x)}.$$

Self-enhanced decay (SEC) enhances robustness to noise in source



SEC provides for greater robustness compared to UD because the length scale close to the source is smaller.

Sensitivity coefficient

Sensitivity coefficient

Measure of the fold-change in system output with respect to any fold-change in input

$$S_{a,b} = \frac{d \ln a}{d \ln b} = \frac{da/a}{db/b}. \quad (1)$$

Sensitivity coefficients are unitless, facilitating comparisons among models and mechanisms.

Sensitivity coefficient of Dpp gradient

$$c(x) = c_0 \exp\left(-\frac{x}{\lambda}\right); \quad \lambda = \sqrt{\frac{D}{k}}$$

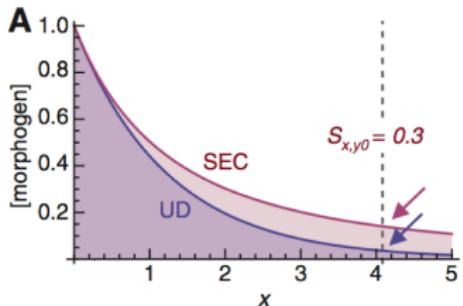
$$x_\theta = \lambda (-\ln c_\theta + \ln c_0)$$

$$\frac{dx_\theta}{dc_0} = \lambda \frac{d \ln c_0}{dc_0} = \lambda \frac{1}{c_0}$$

The sensitivity, S_{x_θ, c_0} , of any threshold position $x_\theta > 0$ to the level of morphogen, $c_0 = c(x=0)$, is then

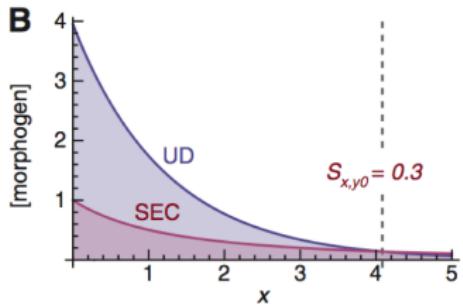
$$S_{x_\theta, c_0} = \frac{dx_\theta/x_\theta}{dc_0/c_0} = \frac{\lambda(x=0)}{x_\theta}. \quad (2)$$

Same sensitivity and ligand concentration possible at read-out point with UD and SEC



Could achieve same positional robustness by choosing a smaller length scale in the UD model

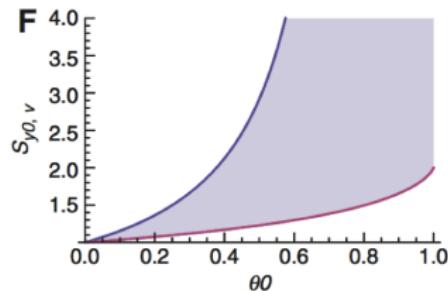
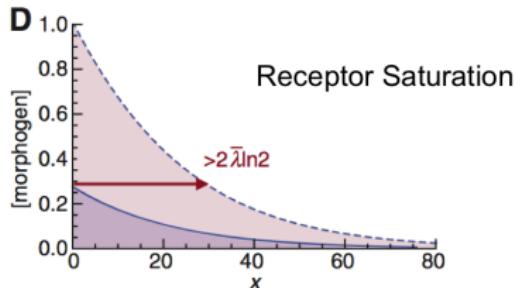
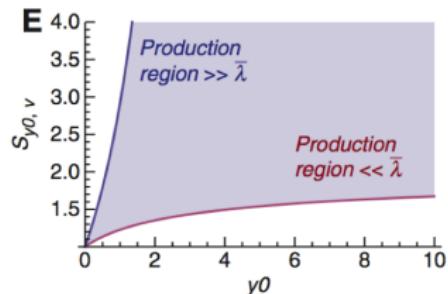
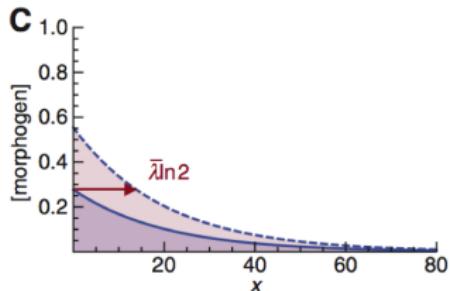
=> however, then much stronger decay



=> higher starting value required to have same morphogen concentration at the threshold

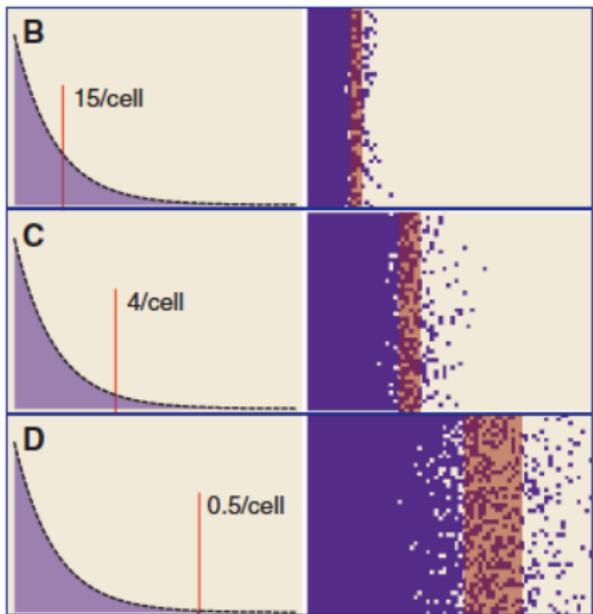
Lander, A.D., W.C. Lo, Q. Nie, and F.Y.M. Wan, The Measure of Success: Constraints, Objectives, and Tradeoffs in Morphogen-mediated Patterning, Cold Spring Harbor Perspectives in Biology, 1: p. a002022-a002022. (2009)

What are the limits on the maximum of the gradient?



Conclusion: matching both robustness and morphogen levels with UD is impossible.

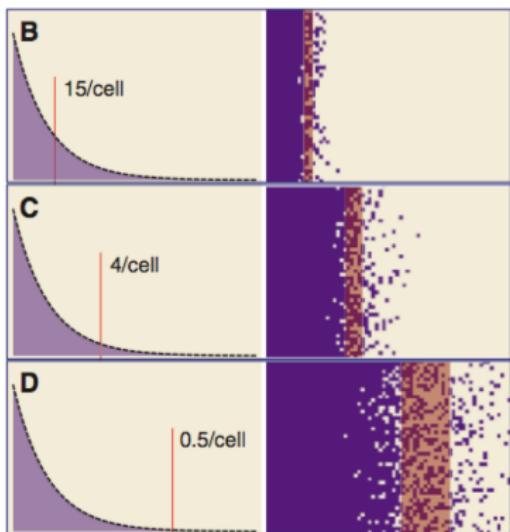
Effect of binding noise on patterning



Field of 50×70 cells exposed to an exponentially declining morphogen gradient with length scale of 10 cell diameters. The initial morphogen concentration is sufficient to occupy 50 receptors per cell. Thresholds for activating gene expression (represented by a colour change from light to dark) occur at occupancy levels of 15 (B), 4 (C), or 0.5 (D) receptors per cell.

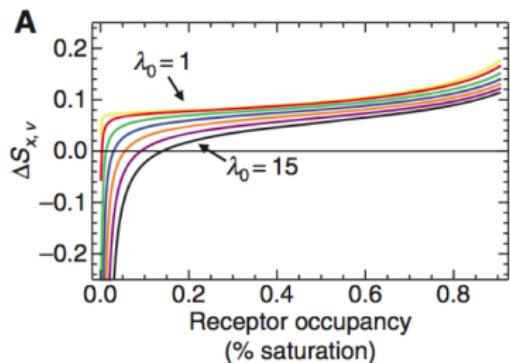
The width of the variegated response region increases with lower occupancy thresholds; this is quantified by overlaid pink boxes, which mark the regions within which cells have more than a 15% chance of responding inappropriately for their position.

Receptor Fluctuations increase with lambda



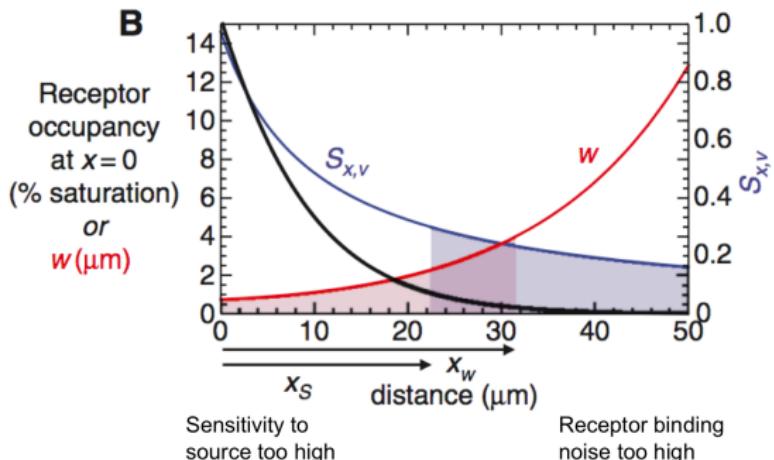
Transition zone:

2 x coefficient of variation x lambda



UD has an advantage here because lambda does not increase with distance

"Useful" Region for Gradient Read-out

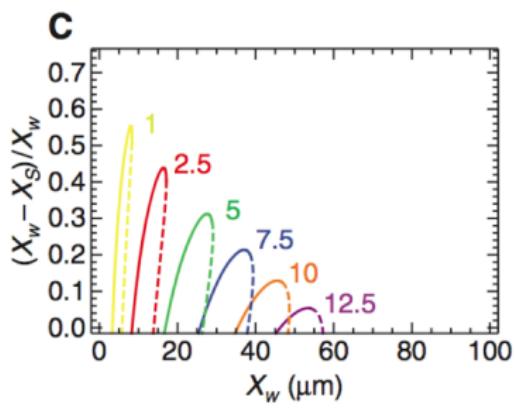


$$\begin{aligned} S_{x_\theta, c_0} &= \frac{dx_\theta/x_\theta}{dc_0/c_0} \\ &= \frac{\lambda(x=0)}{x_\theta} \end{aligned}$$

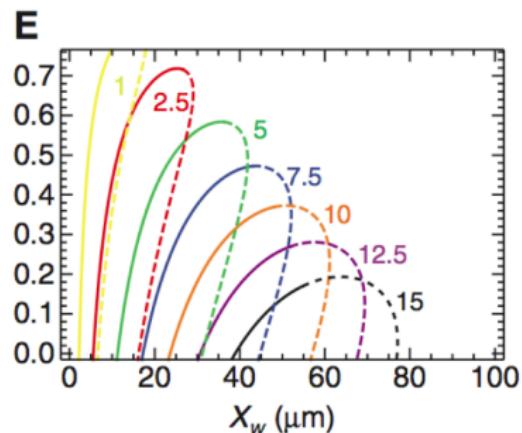
Transition zone
 $w = 2 \times$ coefficient of variation $\times \lambda$

Size of useful region for UD and SEC

UD



SEC



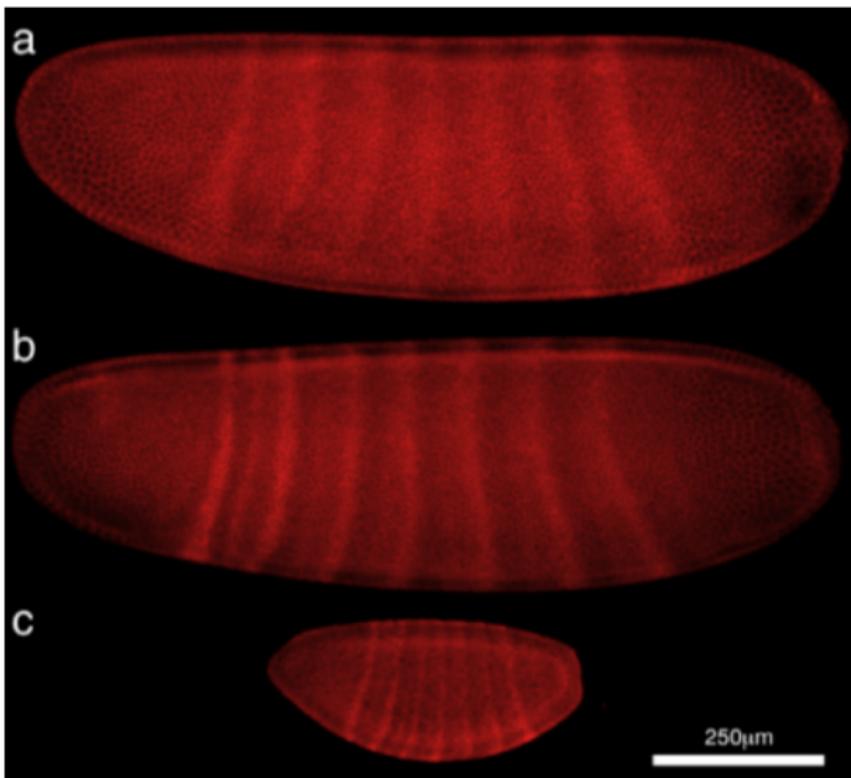
Dpp creates gene expression boundaries in a small range (omb / sal) \rightarrow UD

Wg creates gene expression boundaries in wide range \rightarrow SEC

Morphogen Gradient Scaling

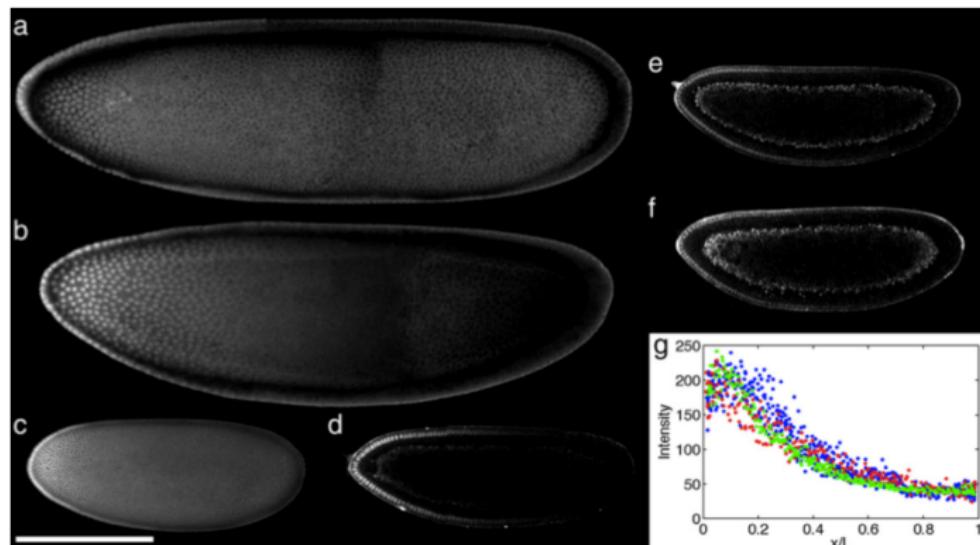
The Bicoid Gradient

Patterning mechanisms are conserved across scales



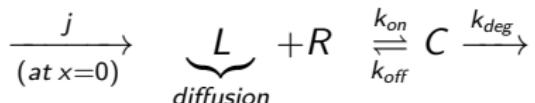
The Bicoid AP-gradient scales with the size of the embryo

Hypothesis: nuclei numbers independent of fly size



Gregor et al. Cell (2007) 130, 141-52

The standard model for morphogen gradients



The total number of receptors is conserved, i.e. $R_T = R + C = \text{const.}$

$$\text{Ligand} \quad \frac{\partial L}{\partial t} = D \frac{\partial^2 L}{\partial x^2} - k_{on}(R_T - C)L + k_{off}C$$

$$\text{Complex} \quad \frac{\partial C}{\partial t} = k_{on}(R_{tot} - C)L - k_{off}C - k_{deg}C$$

$$BC : \quad D \frac{\partial L}{\partial x} \Big|_{x=0} = -j \quad \frac{\partial L}{\partial x} \Big|_{x=\Lambda} = 0$$

Long-time approximation

Assuming that the reaction kinetics are fast, in the limits of long times we can make a quasi steady-state approximation, i.e.

$$\frac{\partial C}{\partial t} = k_{on}(R_T - C)L - k_{off}C - k_{deg}C = 0.$$

such that

$$C_{eq} = \frac{k_{on}R_T L}{k_{on}L + k_{off} + k_{deg}} \approx \frac{k_{on}R_T L}{k_{off} + k_{deg}} \quad \text{for } C \ll R_{tot}.$$

The ligand dynamics then follow as

$$\frac{\partial L}{\partial t} = D \frac{\partial^2 L}{\partial x^2} - k_{deg} \frac{k_{on}R_T}{k_{off} + k_{deg}} L + IC + BC.$$

Scaling with domain size

$$\frac{\partial L}{\partial t} = D \frac{\partial^2 L}{\partial x^2} - k_{deg} \frac{k_{on} R_T}{k_{off} + k_{deg}} L + IC + BC$$

Re-scaling position as $\zeta = x/\Lambda$, $0 < \zeta < 1$, yields

$$\frac{\partial L}{\partial t} = D^* \frac{\partial^2 L}{\partial \zeta^2} - kL + IC + BC$$

with $D^* = \frac{D}{\Lambda^2}$ and $k = k_{deg} \frac{R_T}{K_m}$, where $K_m = \frac{k_{off} + k_{deg}}{k_{on}}$.

We then have as steady-state solution

$$L = L_0 \exp(-\zeta/\lambda^*) \quad \text{with} \quad \lambda^* = \sqrt{\frac{D^*}{k}} = \sqrt{\frac{DK_m}{k_{deg} R_T} \frac{1}{\Lambda^2}}.$$

Summary: Scaling with domain size

Re-scaling position as $\zeta = x/\Lambda$, $0 < \zeta < 1$, yields for the spatial steady state concentration profile of the morphogen ligand L :

$$L = L_0 \exp(-\zeta/\lambda^*) \quad \text{with} \quad \lambda^* = \sqrt{\frac{D^*}{k}} = \sqrt{\frac{DK_m}{k_{deg} R_T \Lambda^2}}$$

To achieve scaling require $\frac{DK_m}{k_{deg} R_T} \propto \Lambda^2$.

Diffusion constant does not differ in embryos of different size

Table 1. Effective diffusion constants, D of dextran molecules of different sizes in *D. melanogaster*

Molecular mass, kDa	r_s , nm	N	D , $\mu\text{m}^2/\text{s}$
10	2.3	11	29.1 ± 4.2
40	4.5	20	17.6 ± 1.8
70	5.9	8	15.3 ± 1.4
150	9.0	5	12.9 ± 3.4

The sample size N refers to the number of diffusion experiments analyzed.

Table 2. Effective diffusion constants of 40-kDa dextran molecules in dipteran species

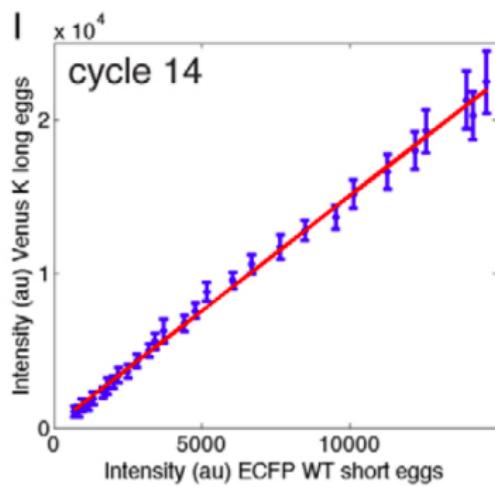
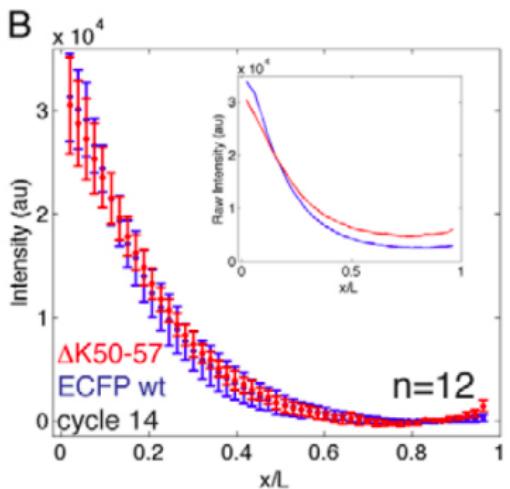
Species (mean egg length)	N	D , $\mu\text{m}^2/\text{s}$
<i>D. busckii</i> (344 μm)	8	14.5 ± 3.8
<i>D. melanogaster</i> (485 μm)	20	17.6 ± 1.8
<i>L. sericata</i> (1,170 μm)	6	22.8 ± 1.5
<i>C. vicina</i> (1,420 μm)	4	20.3 ± 1.3

Conditions for scale invariance

- D appears not to change between species (unless specific processes for Bcd)
- \Rightarrow Assume that D , k_{deg} , K_m do not change between species
- We then require $R_T \sim 1/\Lambda^2$
- Could be achieved if $R_T = \sigma N_T / \Lambda^2$, and total receptor number N_T conserved.

$\lambda^* = \sqrt{(D^*/k^*)}$ and k^* determines time-scale on which gradients reach steady state.

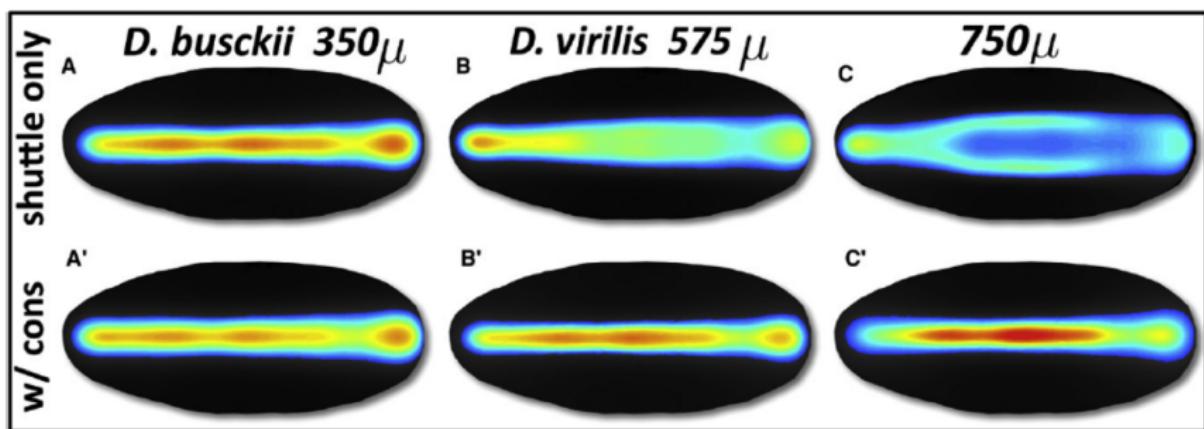
Nuclei shuttling dynamics do not affect gradient shape



Grimm and Wieschaus (2010)

The Dpp Gradient in DV patterning

The Dpp DV-gradient scales with the size of the embryo



Natural size variations

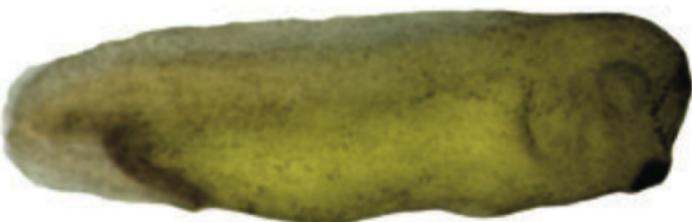
Blastula

4 h post-fertilization



Tadpole

33 h post-fertilization



Barkai & Ben-Zvi, FEBS Lett, 2009

Thanks!!

Thanks for your attention!

Slides for this talk will be available at:

<http://www.bsse.ethz.ch/cobi/education>