Simulating Gene Expression Patterns During Zebrafish Embryo Development

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June 2003 - August 2003

Projects

- Simulations
  1. Neurogenesis
  2. Somitogenesis
  3. Morphogenesis during A/P Patterning

- Model organism: Zebrafish

- Program simulations in StarLogoT/NetLogo

Why Zebrafish (*Danio rerio*)?

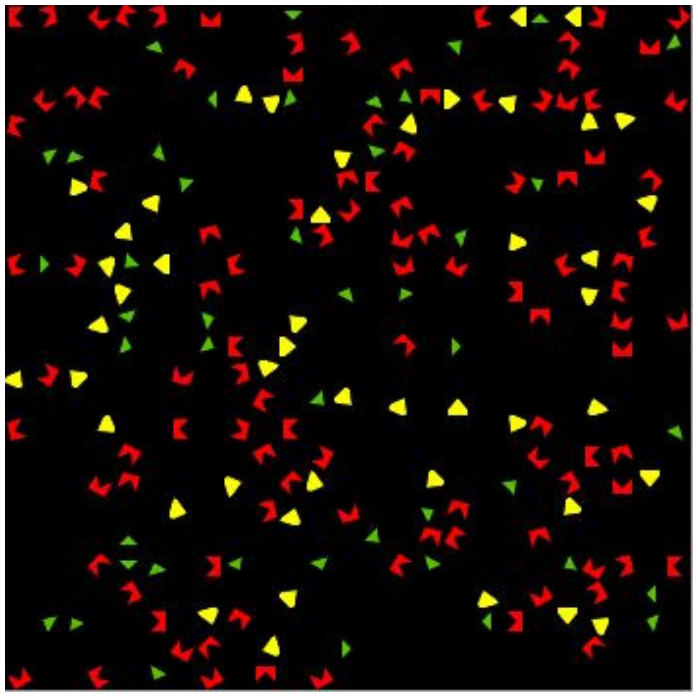
- Zebrafish are vertebrates
- Inexpensive and easy to maintain and breed
- Experimentally accessible
- Easy to manipulate the genetics and the embryos
- Transparent during early development
- Large community of resources (Zfin)

StarLogo/NetLogo

- Based on LOGO programming enviornment
  - Daniel Bobrow and Wallace Feurzeig at Bolt, Beranek and Newman, Inc., and Seymour Papert, at the MIT in the 1960's.
- StarLogo: 1989 - 90 (MIT Media Lab)
- MacStarLogo: 1994
- StarLogoT: 1997 (Center for connected Learning and Computer-Based Modeling, Uri Wilensky)
- StarLogo in Java: 1999
- NetLogo (StarLogoT in Java): 2002

What is StarLogo/NetLogo?

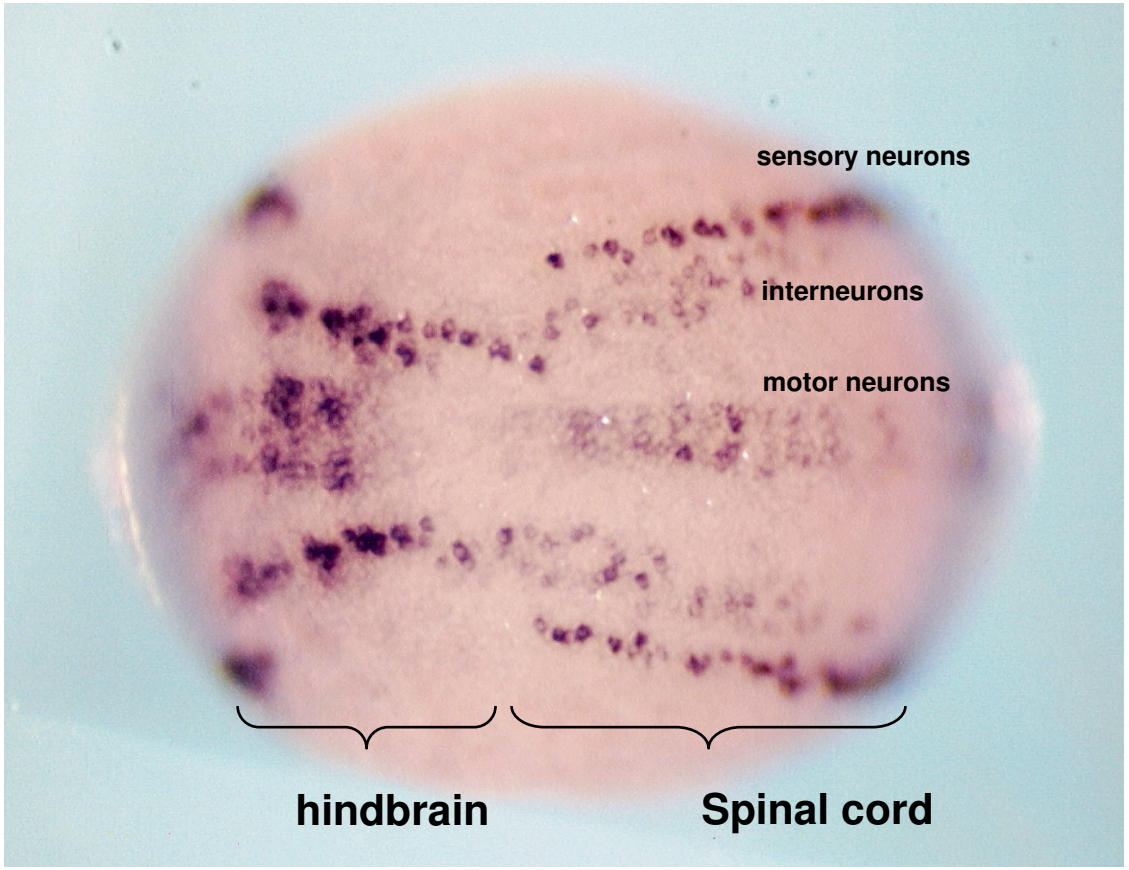
- Programmable modeling environment for exploring *emergent phenomena*.
- Logo language support agents: turtles, patches, and the observer.
- Simulate Parallelism on a one-processor computer.



Neurogenesis

- In early development, the neural plate consists of domains which give rise to primary neurons.
- Cells' fate is determined by the expression of *neuralgenin1* (*ngn1*).
- *ngn1* is modulated by lateral inhibition through Delta/Notch signaling cascade.

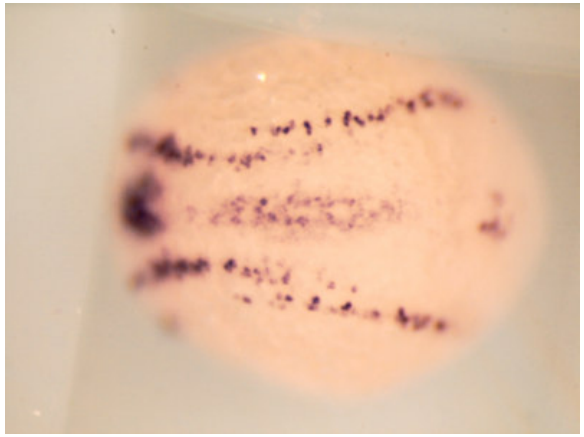
Proneuro Domains

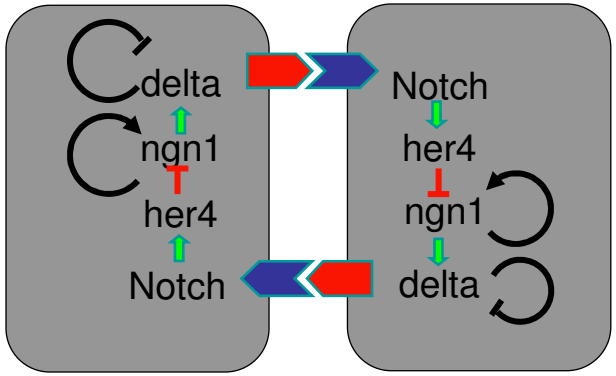


Lateral Inhibition

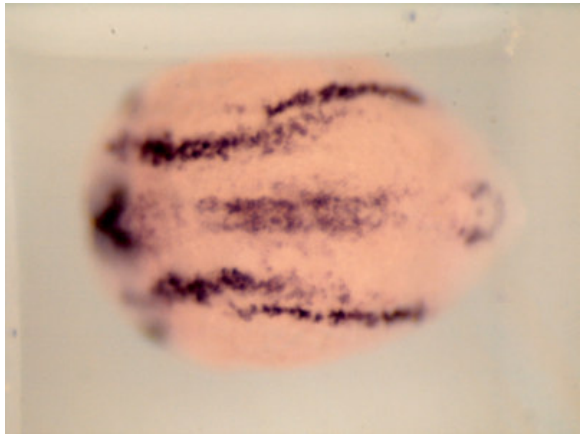
Delta/Notch Signaling

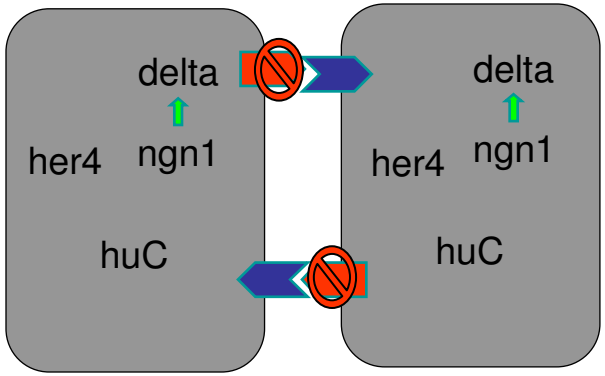
Normal



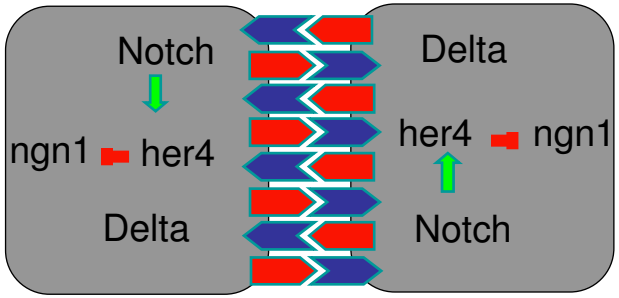


Delta MO injected





Injected with *DeltaA* RNA



Autocatalytic/Lateral Model

- Self-enhancement and long-range inhibition leads to spatially-patterned differentiation (Meinhardt and Gierer, 1974)

$$\frac{\partial a}{\partial t} = s\left(\frac{a^2}{(b_a+1)(1+S_a a^2)}\right) - r_a a + D_a \frac{\partial^2 a}{\partial x^2}$$

$$\frac{\partial b}{\partial t} = s a^2 - r_b b + D_b \frac{\partial^2 b}{\partial x^2} + b_b$$

Neurogenesis Model

$$\frac{\partial A_{ngn}}{\partial t} = P_{ngn}\left(\frac{A_{ngn}^{\ 2}}{\left(A_{her}+1\right)\left(1+S_{ngn}A_{ngn}^{\ 2}\right)}\right) - R_{ngn}A_{ngn} \ ,$$

$$\frac{\partial A_{del}}{\partial t} = P_{del}\left(\frac{A_{ngn}}{1+S_{del}A_{del}}\right) - R_{del}A_{del} \ , \text{ and}$$

$$\frac{\partial A_{her}}{\partial t} = P_{her}C_{notch}A_{tot\_del} - R_{her}A_{her} \ .$$

ngn – neurogenin *del* – delta *her* – hairy
tot_del – total delta in neighboring cells

NetLogo Model

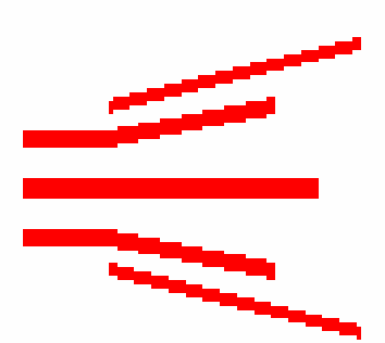
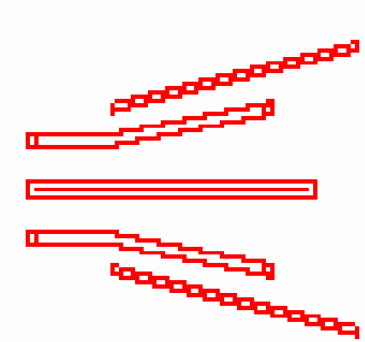
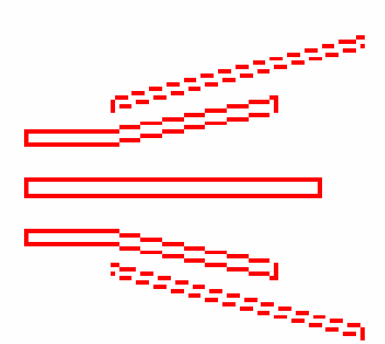
- Setup proneuro domains.
- Set $A_{ngn}[0]$ and calculate $A_{del}[0]$ and $A_{her}[0]$. Add Type 1 noise.
- Update $A_{ngn}[t]$, $A_{del}[t]$, and $A_{her}[t]$ using Euler's method with step size of 1. Add Type 2 noise.
- If A_{ngn} > threshold, the patch is fated to become neuron.

Neurogenesis Model Parameters

Parameter	Gene		
	ngn	*delta*	*her*
Rate of synthesis (*P*)	0.5	9.2	0.25
Rate of Degradation (*R*)	0.20	0.91	0.90
Rate of Saturation or Self-Inhibition (*S*)	0.06	0.30	None
Threshold	4		

Importance of Notch Signaling

C_{notch}

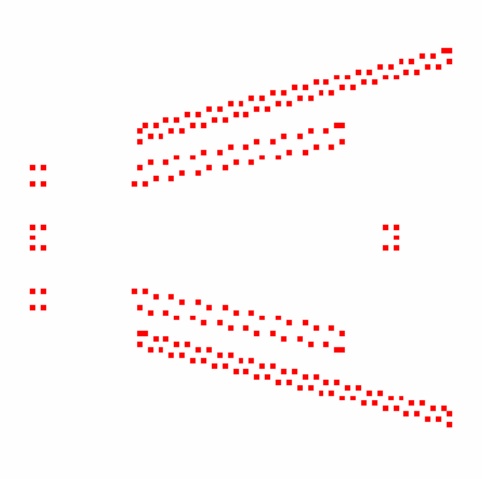
0.025	0.05	0.075	0.3
			
1171 (100%)	785 (67%)	568 (49%)	2 (< 1%)

Number of Neurons

Initial Condition

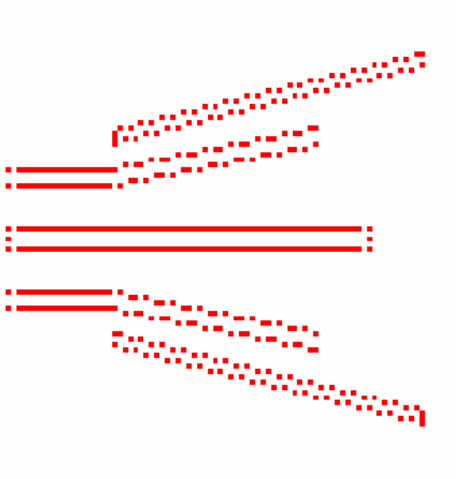
$P_{ngn}[0]$

0.52



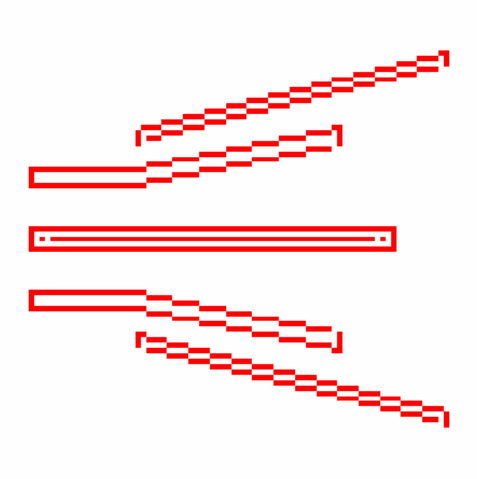
208

0.53



430

0.63

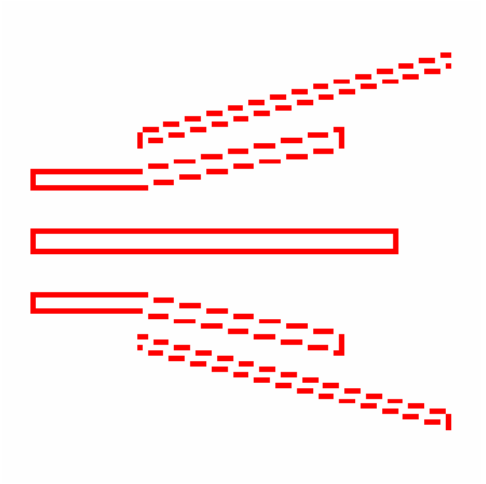


685

Number of Neurons

Noise

No Noise

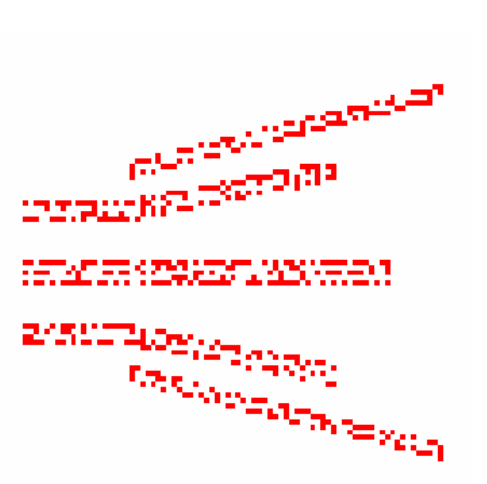


536

Type 1 Noise
var = 0.04

532

Type 2 Noise
var = 0.04

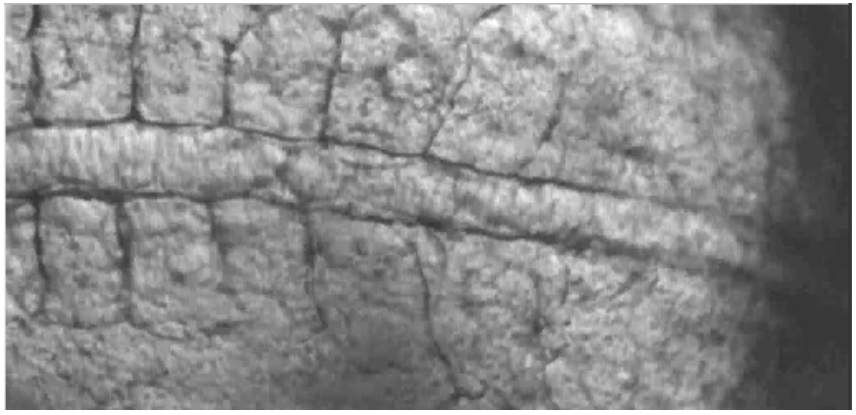


539

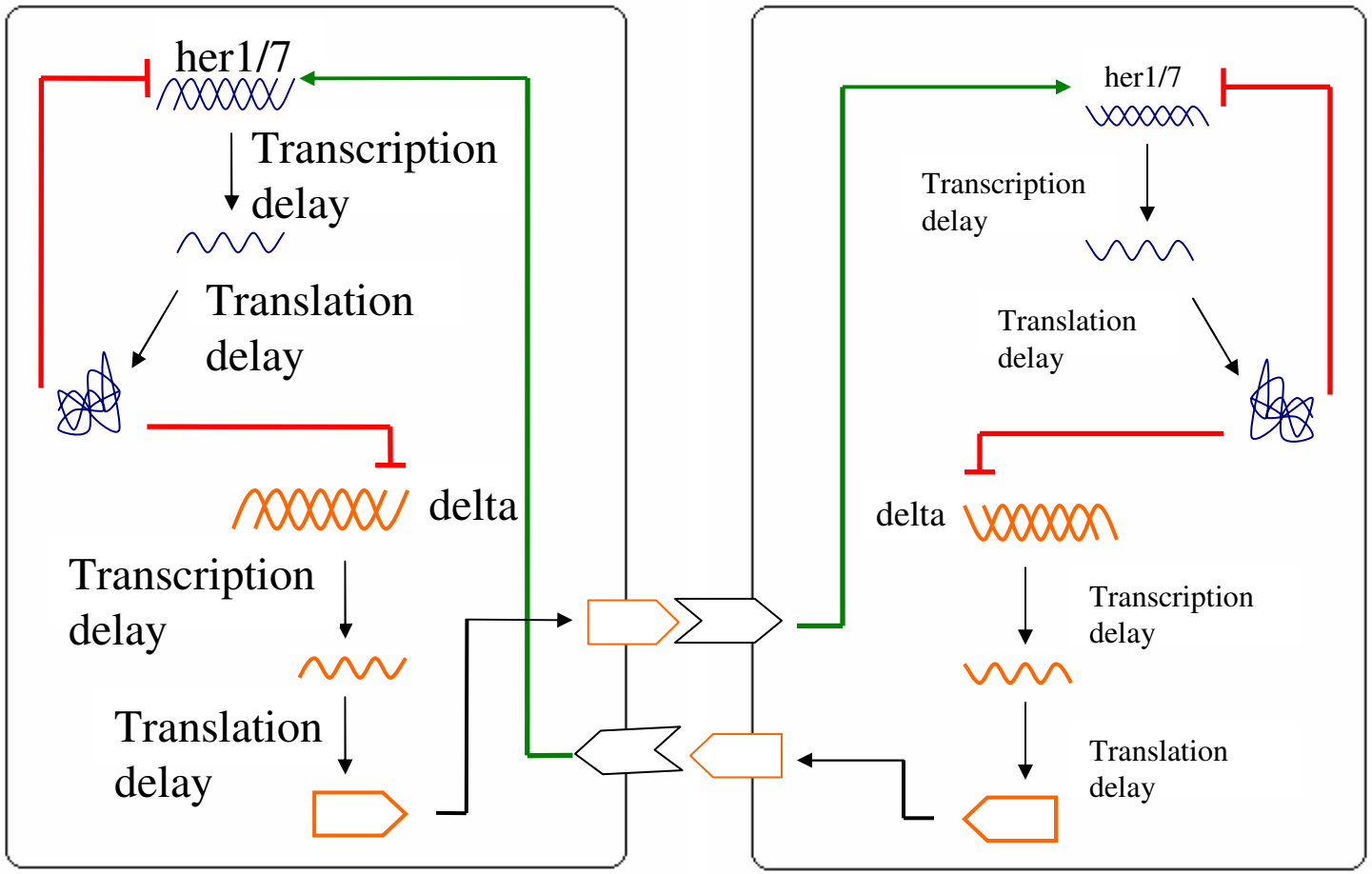
Number of Neurons

Somitogenesis

- Somites are formed from the presomitic mesoderm (PSM)
- Segments are formed head to tail in periodic succession
- *her1/her7* oscillate and regulated by Notch pathway



Zebrafish


her1/7
Transcription
delay
Translation
delay
delta
Transcription
delay
Translation
delay
her1/7
Transcription
delay
Translation
delay
delta
Transcription
delay
Translation
delay

Lewis Model
Two-cell *her1/her7* Oscillator

$$\frac{dp_{her1}}{dt} = a_{her1}\left[m_{her1}\right]_{t-T_{pher1}} - b_{her1}\left[p_{her1}\right]_t$$

$$\frac{dp_{her7}}{dt} = a_{her7}\left[m_{her7}\right]_{t-T_{pher7}} - b_{her7}\left[p_{her7}\right]_t$$

$$\frac{dp_{del}}{dt} = a_{del}\left[m_{del}\right]_{t-T_{pdel}} - b_{del}\left[p_{del}\right]_t$$

Lewis Model
Two-cell *her1/her7* Oscillator

$$\frac{dm_{her1}}{dt} = \left[f_{her1}\left(p_{her1}, p_{her7}, p_{n\_del}\right) \right]_{t-T_{mher1}} - c_{her1}\left[m_{her1}\right]_t$$

$$\frac{dm_{her7}}{dt} = \left[f_{her7}\left(p_{her1}, p_{her7}, p_{n\_del}\right) \right]_{t-T_{mher7}} - c_{her7}\left[m_{her7}\right]_t$$

$$\frac{dm_{del}}{dt} = \left[f_{del}\left(p_{her1}, p_{her7}, p_{n\_del}\right) \right]_{t-T_{mdel}} - c_{del}\left[m_{del}\right]_t$$

Lewis Model
Two-cell *her1/her7* Oscillator

$$f_{her1} = k_{her1}\left(r_0 + r_d \frac{\phi_{n\_del}}{1+\phi_{n\_del}} + r_h \frac{1}{1+\phi_{her1}\phi_{her7}} + r_{hd} \frac{\phi_{n\_del}}{\left(1+\phi_{n\_del}\right)} \frac{1}{\left(1+\phi_{her1}\phi_{her7}\right)} \right)$$

$$f_{her7} = k_{her7}\left(r_0 + r_d \frac{\phi_{n\_del}}{1+\phi_{n\_del}} + r_h \frac{1}{1+\phi_{her1}\phi_{her7}} + r_{hd} \frac{\phi_{n\_del}}{\left(1+\phi_{n\_del}\right)} \frac{1}{\left(1+\phi_{her1}\phi_{her7}\right)} \right)$$

$$f_{del} = k_{del}\left(s_0 + s_d \frac{\phi_{n\_del}}{1+\phi_{n\_del}} + s_h \frac{1}{1+\phi_{her1}\phi_{her7}} + s_{hd} \frac{\phi_{n\_del}}{\left(1+\phi_{n\_del}\right)} \frac{1}{\left(1+\phi_{her1}\phi_{her7}\right)} \right)$$

Somitogenisis Simulation

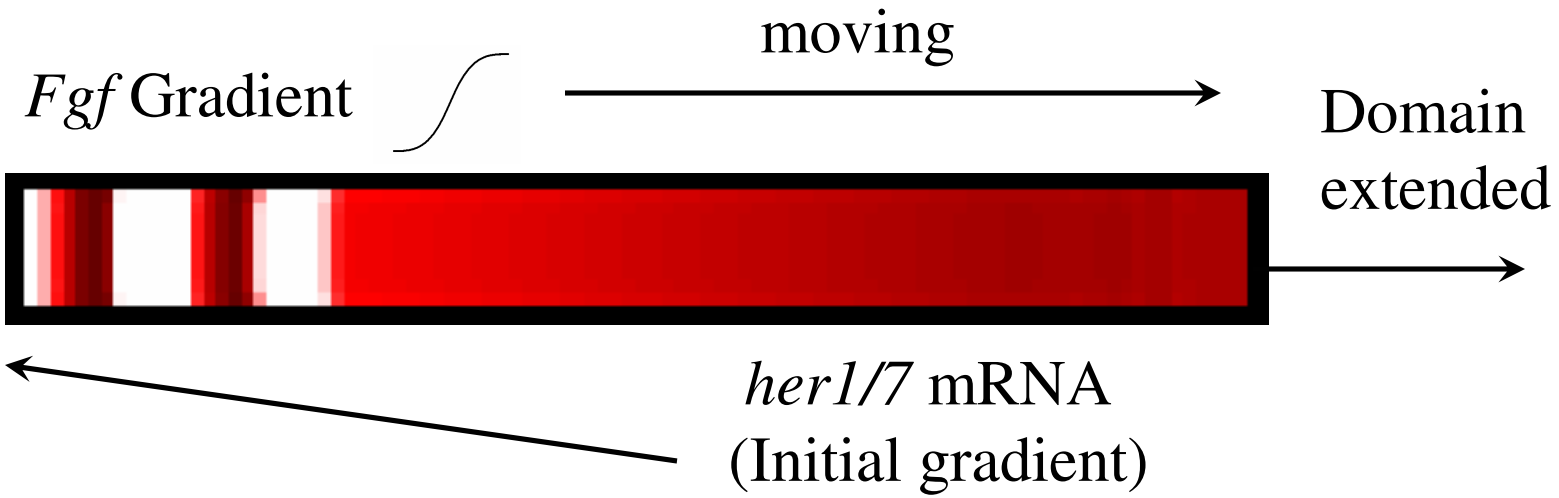
- Initial setup
  1. mRNA gradients for *her1* and *her7* (noise can be added)
  2. *Fgf* gradient

- Update *p* and *m* for her1, her7, and delta using Euler's method with step size of 1.

- *Fgf* gradient moves and the domain is extended w/ time.

- Oscillation ceases when *Fgf* is below critical point

Somitogenisis Simulation



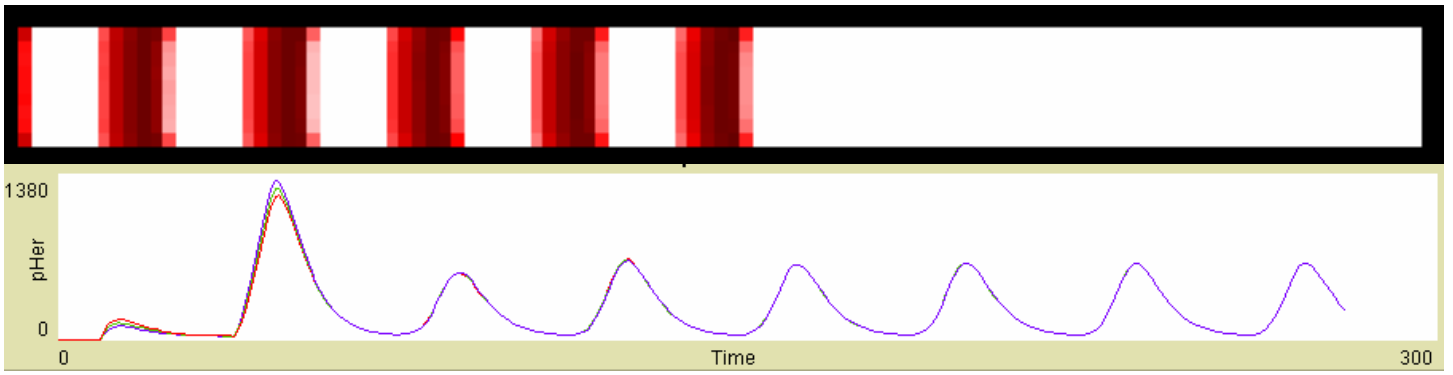
Somitogenesis Simulation Parameters

Parameter	Gene		
	her1	*her7*	*delta*
Rate of protein synthesis (*a*)	4.5	5	4.5
Rate of protein degradation (*b*)	0.23	0.23	0.23
Transcription delay (*Tm*)	10	6	26
Rate of mRNA synthesis (*k*)	33	3	33
Rate of mRNA gradation (*c*)	0.23	0.23	0.23
Translation delay (*Tp*)	3	2	6
Critical protein level (*Pcrit*)	5	40	1000
Threshold	200		

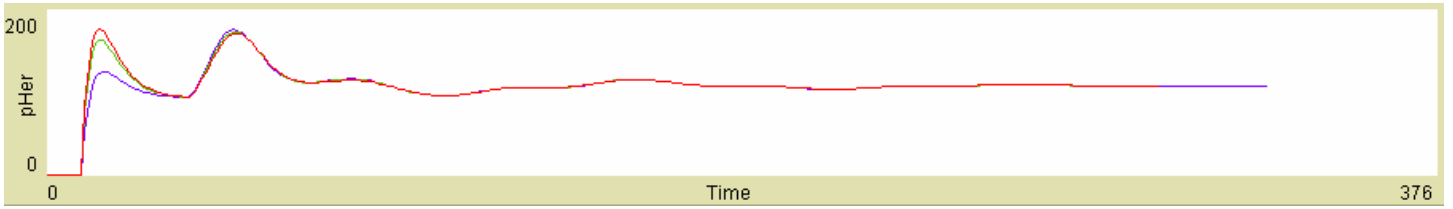
Synchronization & Oscillating Pattern

r_{hd} =

0.99



0.97



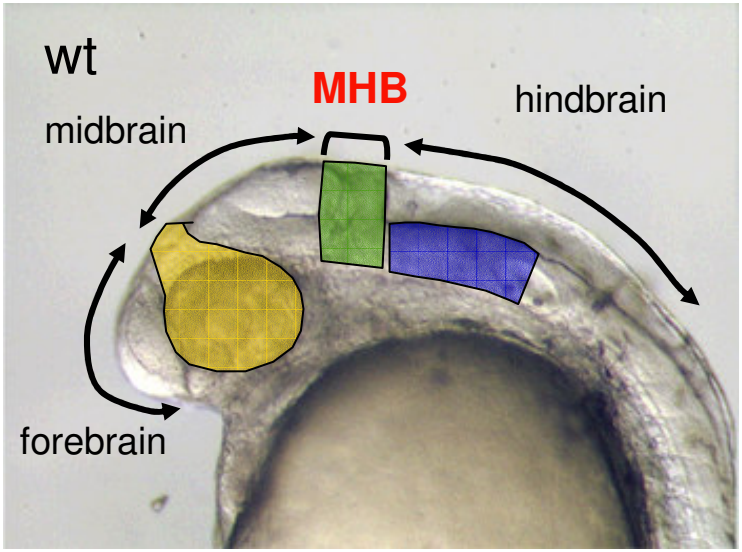
0.70

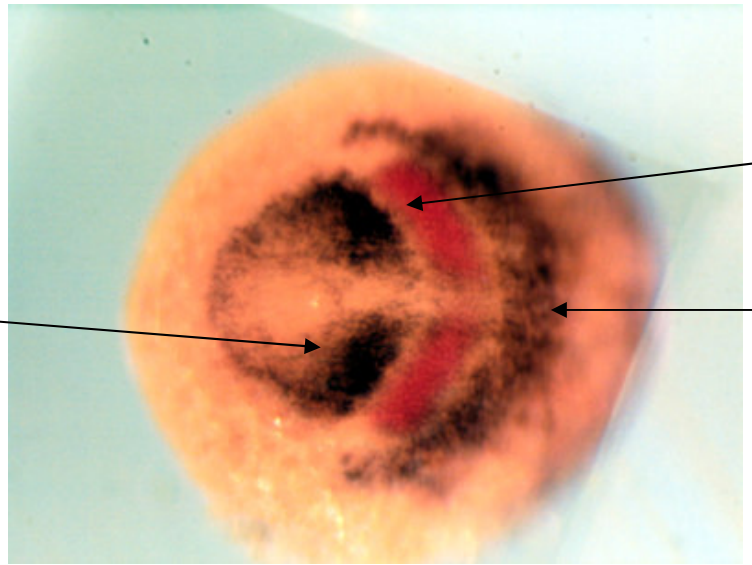
Zebrafish A/P Patterning

- Rostral-caudal axis of the developing vertebrate nervous system differentially express genes.

- Morphogen, such as Wnt plays a critical role.

Anterior Posterior Patterning



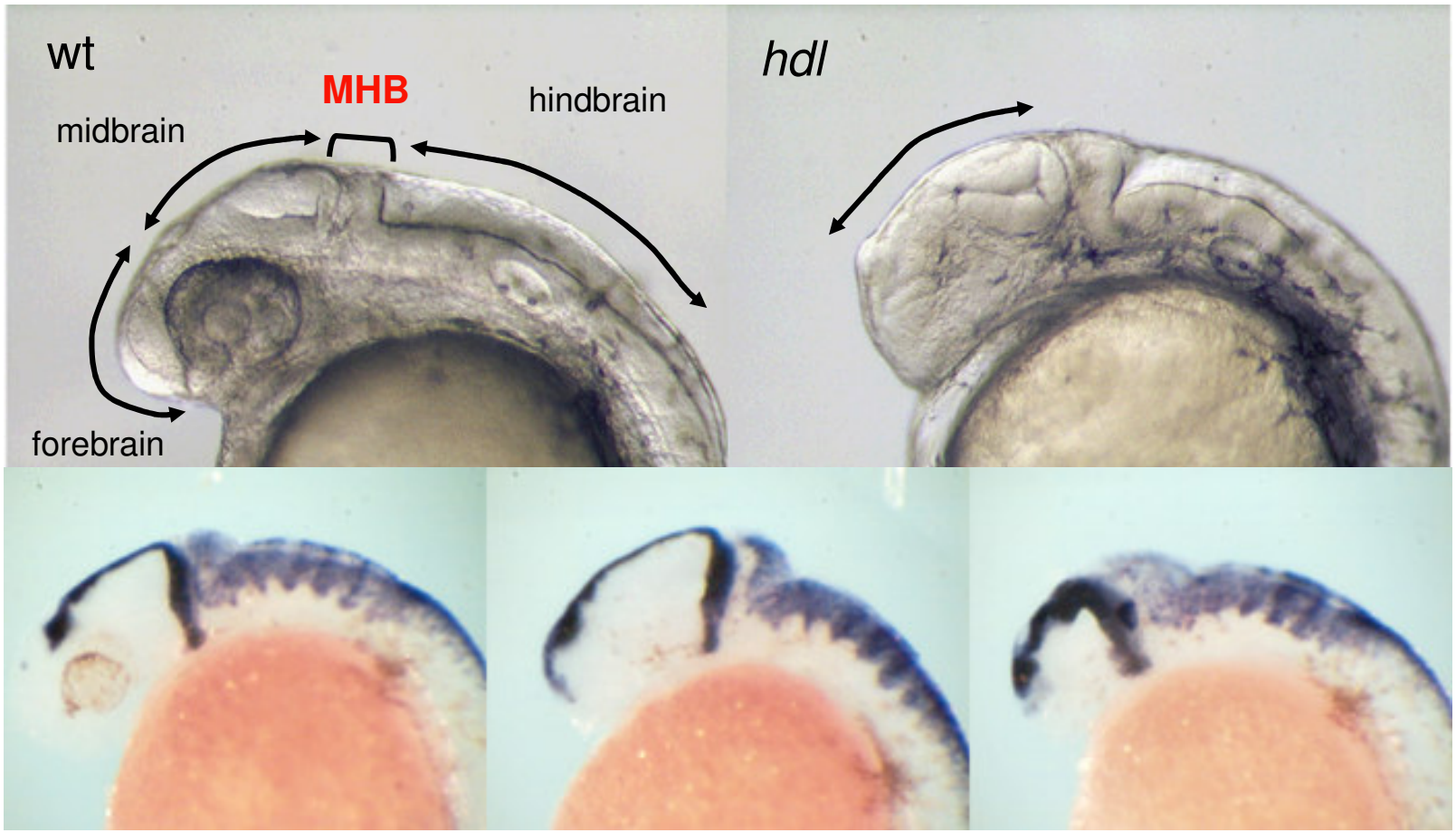


pax6

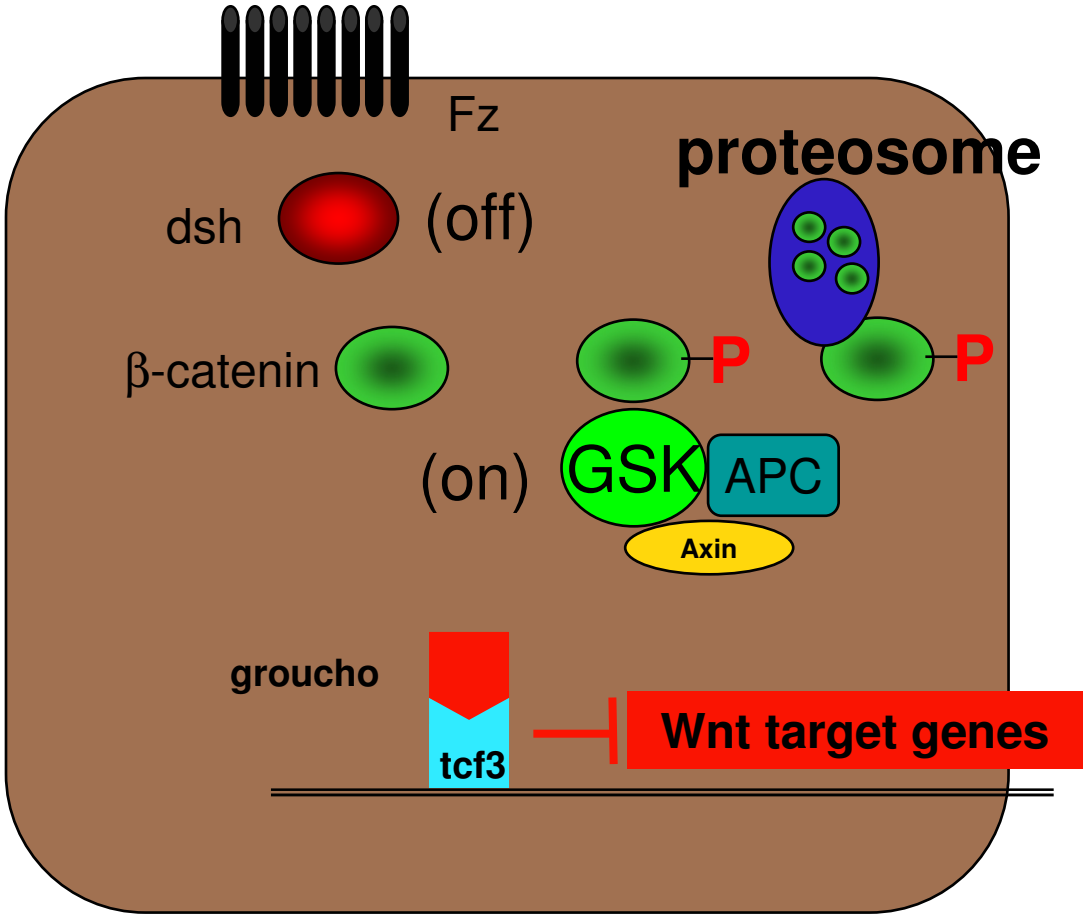
pax2.1

gbx1

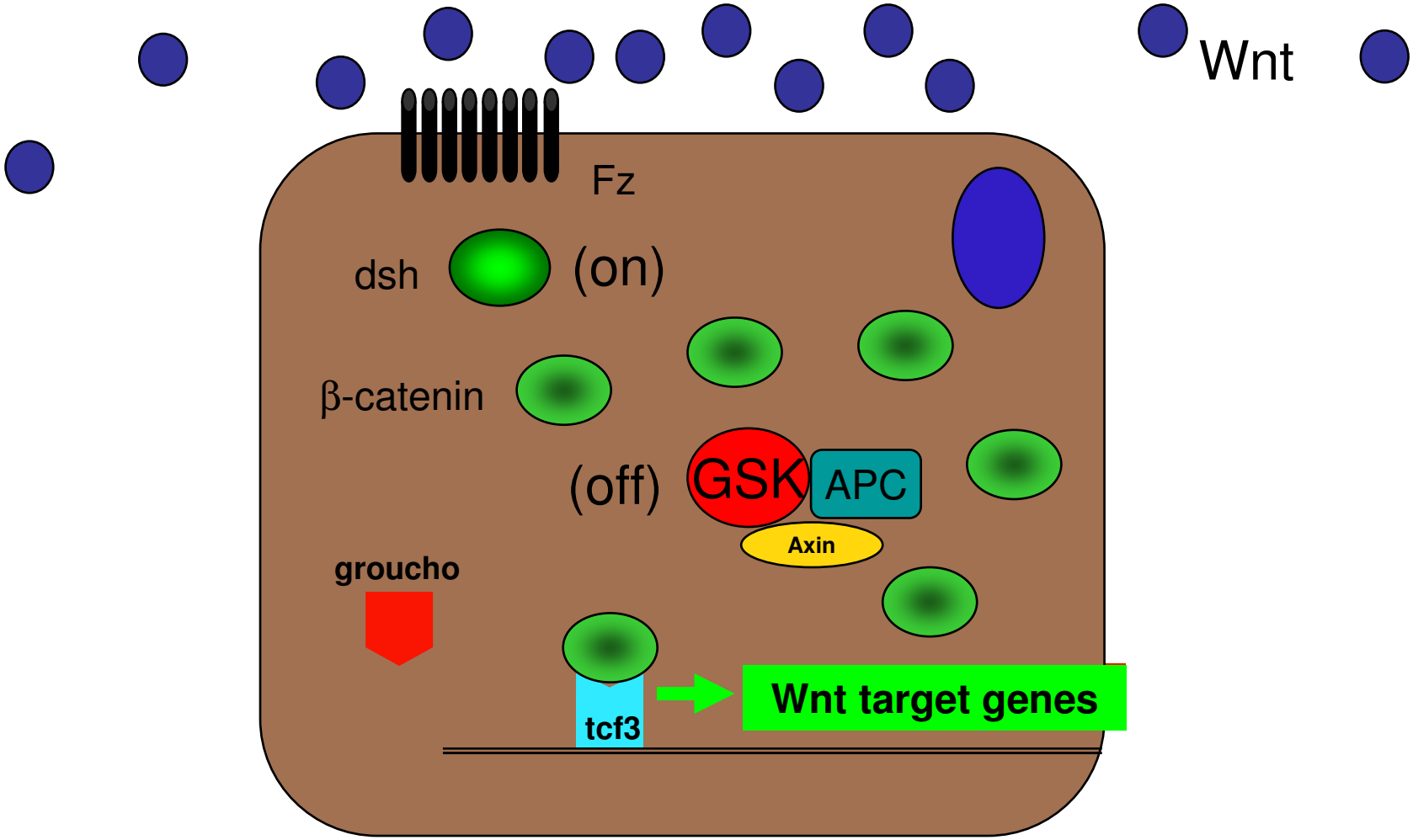
Headless Mutants



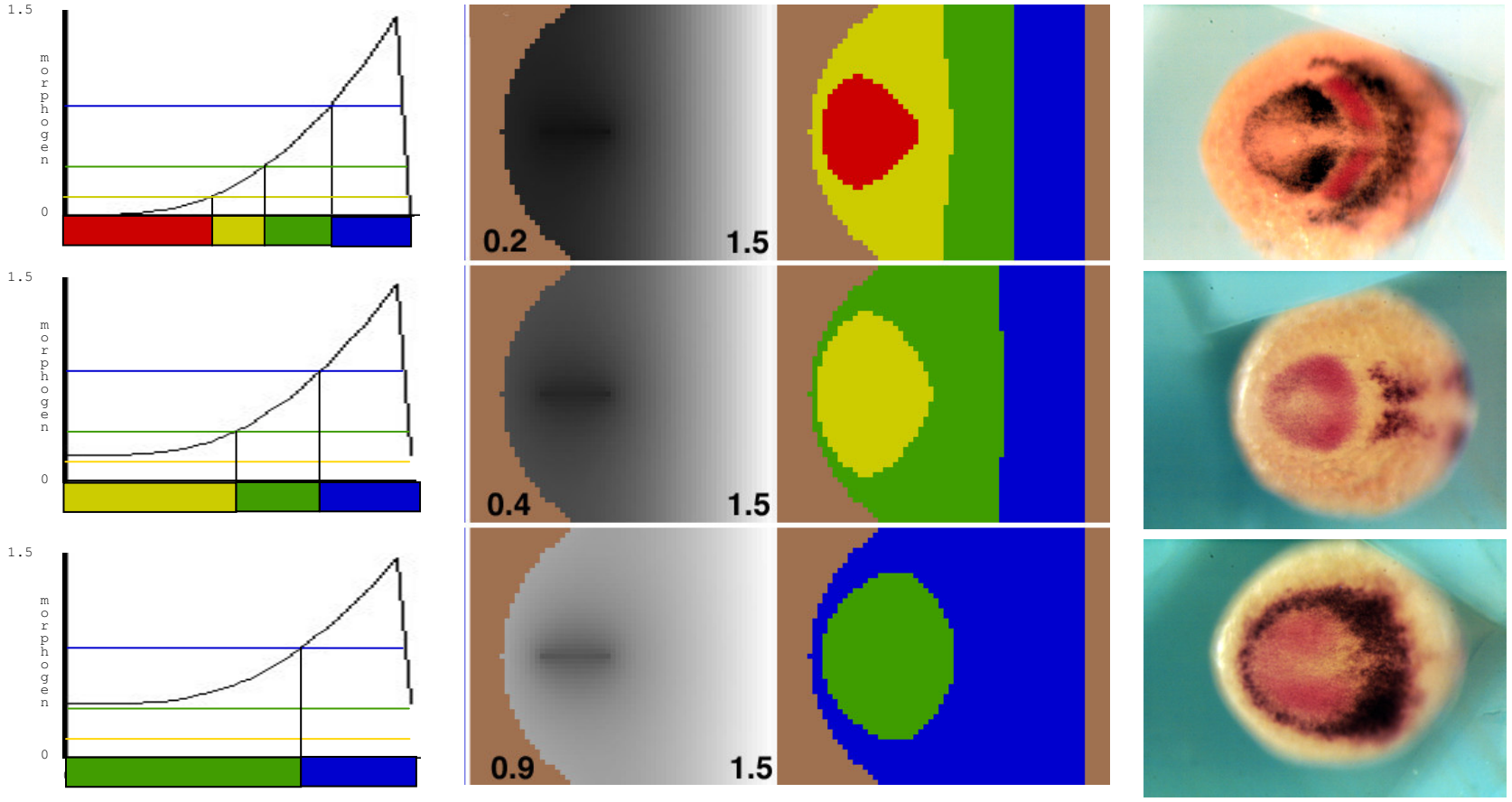
Repressor tcf3



Wnt Target Genes Activated



Neural Patterning and Morphogen Gradient



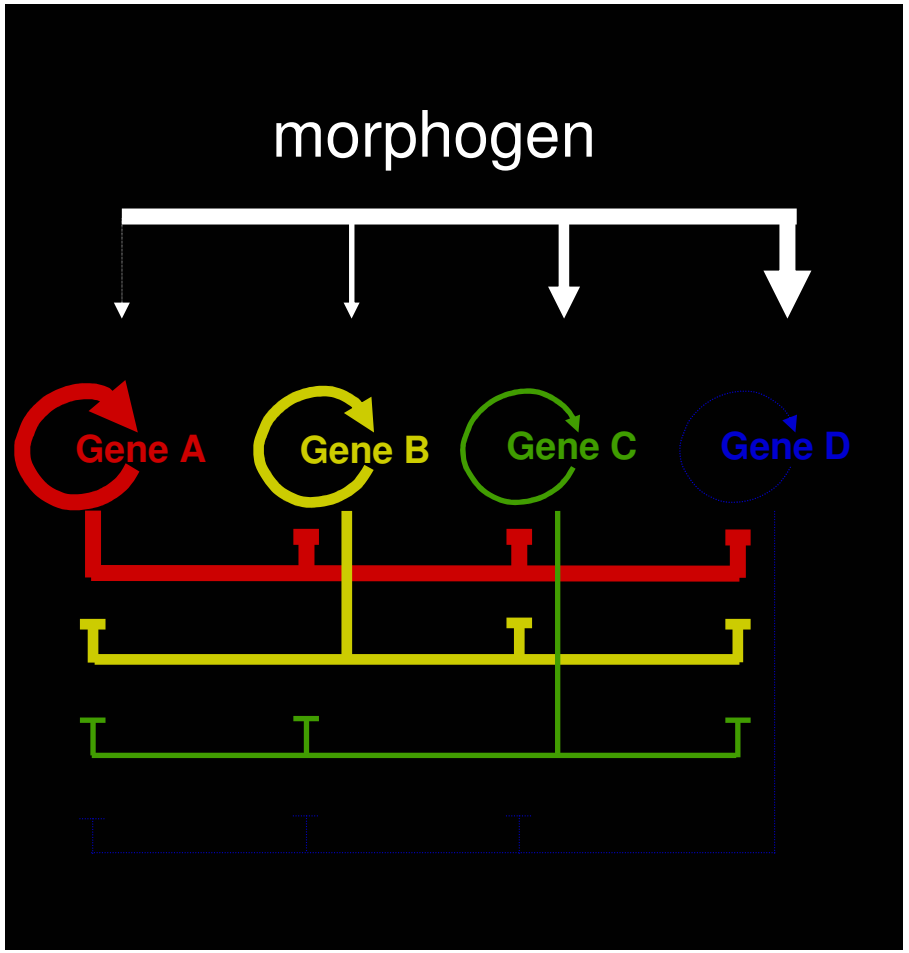
Meinhardt Model:
Spatial Sequences/Morphogen Gradient

$$\frac{dG_a}{dt} = \frac{S_a(G_a^{\,2} + M_t M_a)}{G_a^{\,2} + G_b^{\,2} + G_c^{\,2} + G_d^{\,2} + 1} - R_a G_a$$

$$\frac{dG_b}{dt} = \frac{S_b(G_b^{\,2} + M_t M_b G_a)}{G_a^{\,2} + G_b^{\,2} + G_c^{\,2} + G_d^{\,2} + 1} - R_b G_b$$

$$\frac{dG_c}{dt} = \frac{S_c(G_c^{\,2} + M_t M_c G_b)}{G_a^{\,2} + G_b^{\,2} + G_c^{\,2} + G_d^{\,2} + 1} - R_c G_c$$

$$\frac{dG_d}{dt} = \frac{S_d(G_d^{\,2} + M_t M_d G_c)}{G_a^{\,2} + G_b^{\,2} + G_c^{\,2} + G_d^{\,2} + 1} - R_d G_d$$



Morphogensis Simulation

- Two regions:
  - Morphogen gradient
  - Time exposure to morphogen.

- Numerical method: Euler's method with step size of 1.

Morphogenesis Simulation Parameters

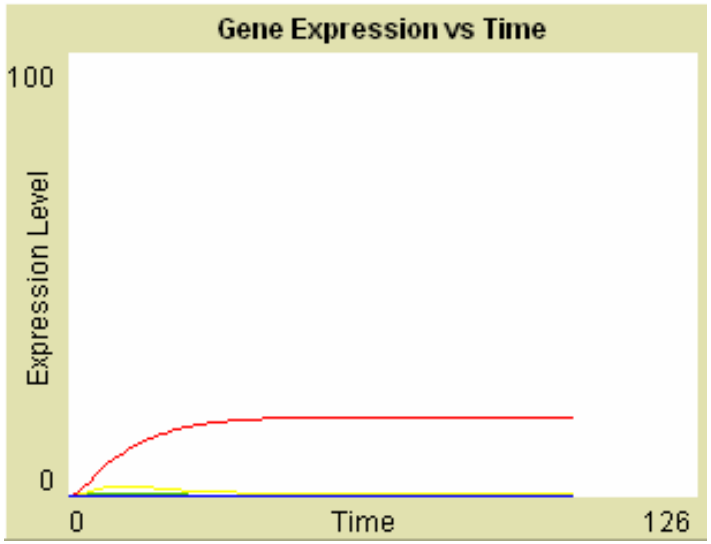
Parameter	$S_a > S_b > S_c > S_d$ and $M_a < M_b < M_c < M_d$				$S_a < S_b < S_c < S_d$ and $M_a > M_b > M_c > M_d$			
	A	B	C	D	A	B	C	D
Rate synthesis (*S*)	2.0	1.9	1.8	1.7	1.8	2.8	4.4	6.7
Rate gradation (*R*)	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Importance of morphogen (*M*)	0.50	1.75	3.25	4.5	1.10	1.20	1.30	1.40
Morphogen	Region 1		Region 2		Region 1		Region 2	
Max Morphgen (*Mmax*)	1.00		1.00		1.00		1.00	
C1	0.01		0.01		0.01		0.01	
Decay Rate (*D*)	-		0.95		-		0.30	
C2	-		0.40		-		1.00	

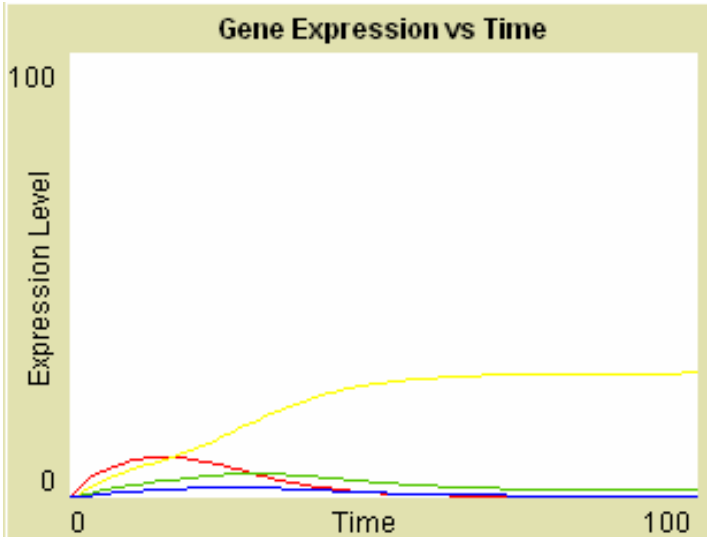
Morphogenesis Simulation

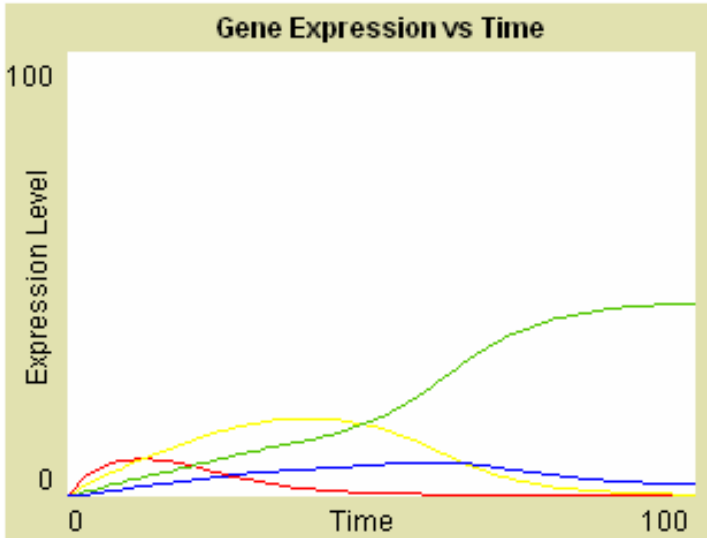
- Spatial differential expression
  1. $S_a > S_b > S_c > S_d$ and $M_a < M_b < M_c < M_d$
  2. $S_a < S_b < S_c < S_d$ and $M_a > M_b > M_c > M_d$

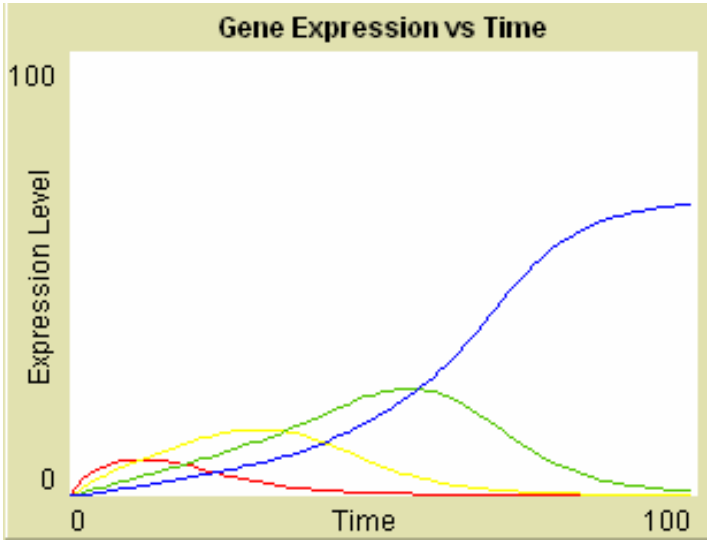
- Similar patterns can be obtained from gradient and time-exposure.

Expression of Different Region

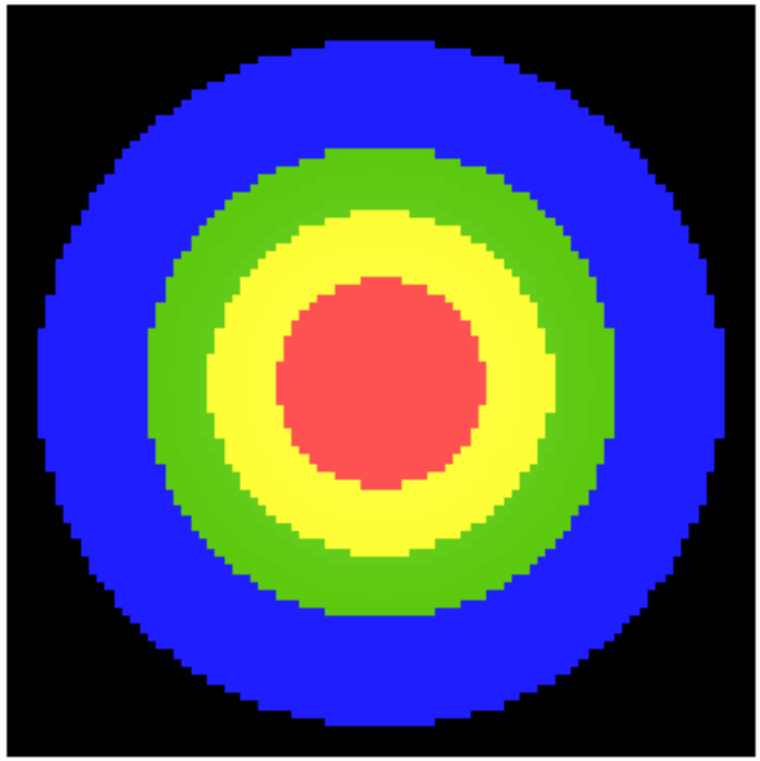




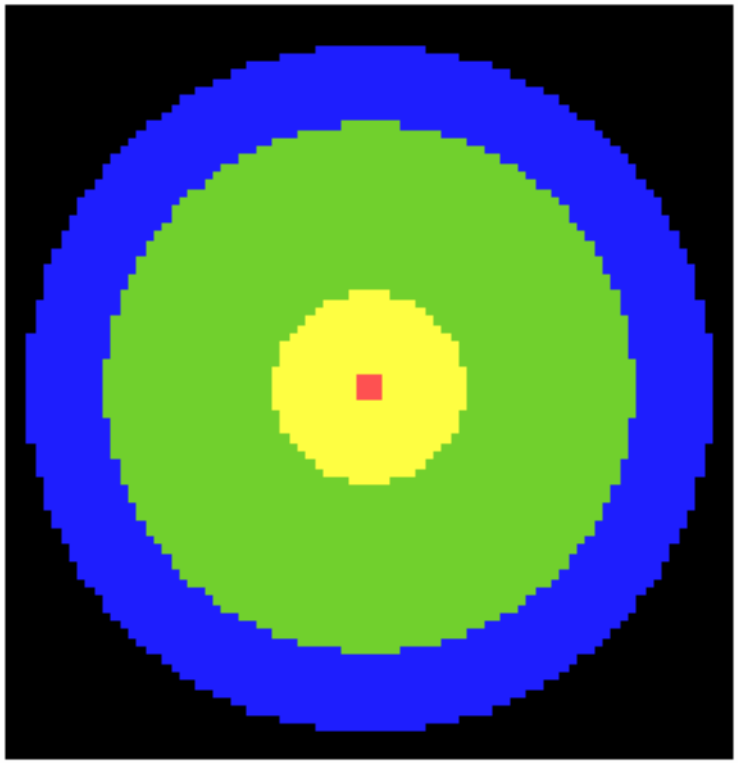




Spatial vs Time

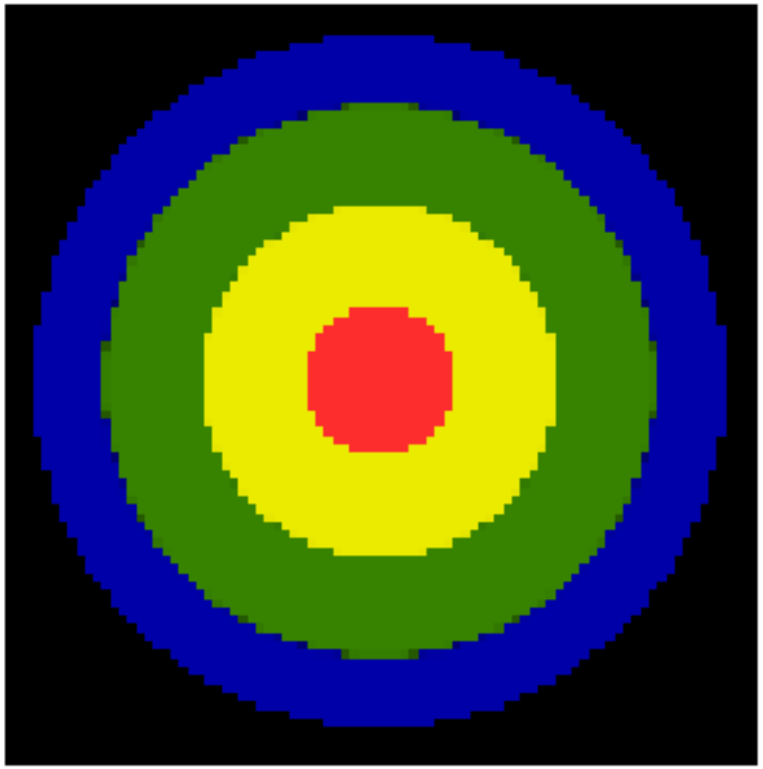


Stable-Morphogen Conc

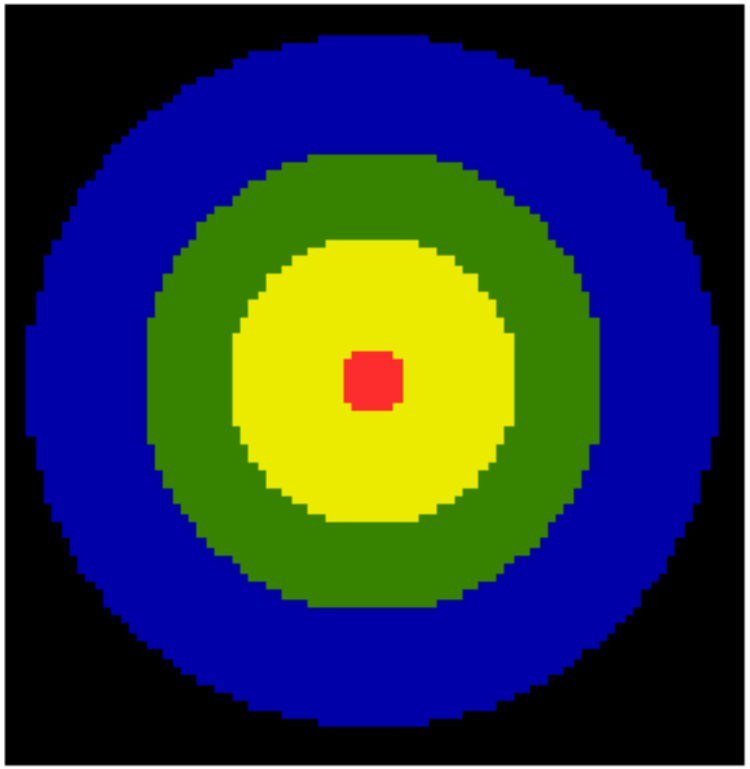


Morphogen Time-Exposure

Spatial vs Time



Stable-Morphogen Conc



Morphogen Time-Exposure

Future Work

- Study to see how much of the patterning is due to bias in the numerical approach.
  - Runge-Kutta method

- Add more aspects to the model
  - Morphogensis: add antogonist, *tcf3*

Many Thanks!

Ajay Chitnis

Moloy Goswami

Motoyuki Itoh

Michael Keller

Gregory Palardy

Sang Yeob Yeo

Darcy Hampton