modeling development

classical models of pattern formation segmentation patterns; STRIPES

Course Computational Biology 2025; Paulien Hogeweg;
Theoretical Biology and Bioinformatics Grp Utrecht University

One more level between genotype and phenotype: Modeling development (and its evolution)

Pattern formation (dependent on Shape)

Pattern formation $-----> $ Shape

Pattern formation $<----->$ Shape

TODAY: Classical models of pattern formation / segmentation

Supervised modeling
Top down modeling:
- Given observed pattern/behaviour X and assumptions A
CAN A $--->$ X (AND does it generate X++)

- Data driven models , quantitative fitting

Theme: specific and/or general mechanisms
and/or specific instantiations (?)

development: cell differentiation,pattern formation and morphogenesis ss

classically most studied: pattern formation

prepattern $--> $ cell differentiatrion $--> $ morphogenesis ss

3 most discussed general mechanisms for stationary pattern formation for development

Turing patterns (Turing 1952)
introduced term 'morphogen'

Positional information (Wolpert 1969)
morphogen gradient - coordinate system

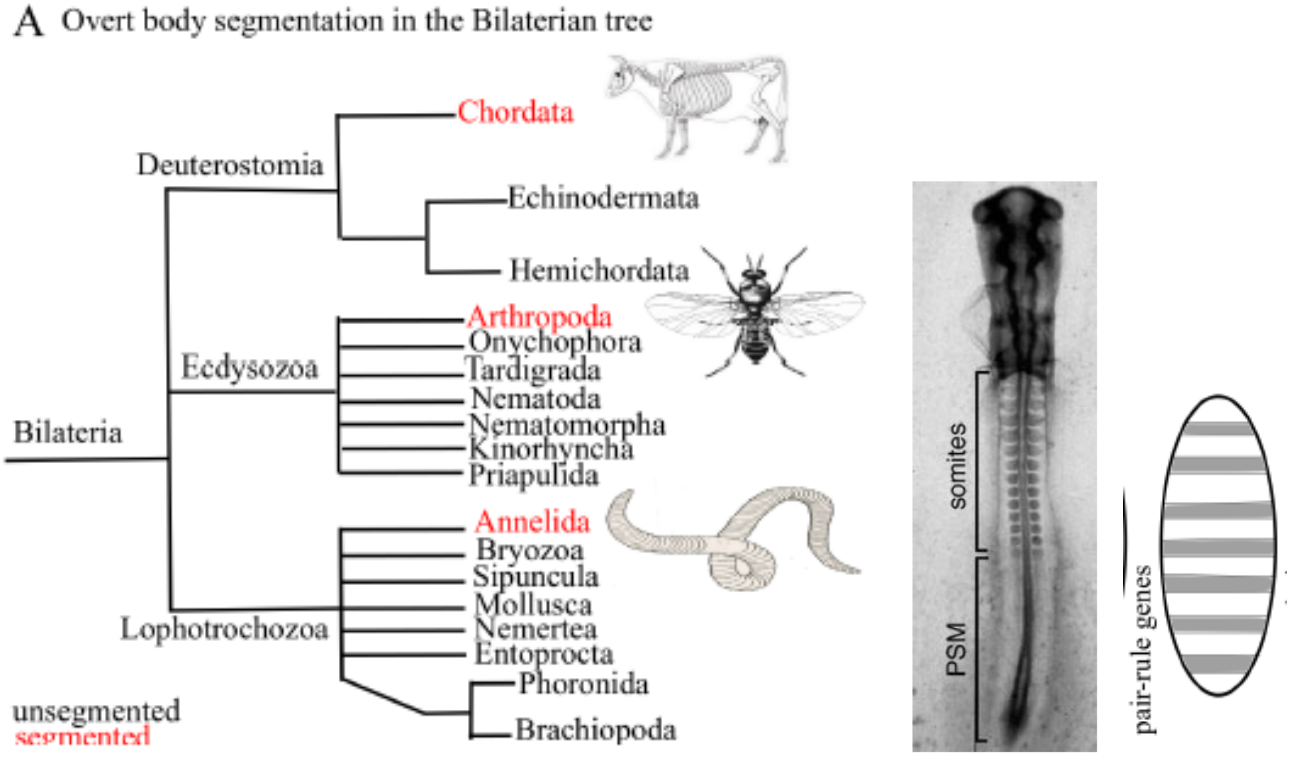
" Clock and wavefront" Cook and Zeeman 1976
temporal oscilation $--> $ spatial pattern

compare: "pattern is 'default"

however here specific positioning/orientation

in continuos medium

Segmented bodyplans



from Ten Tusscher EPJE

reinventions (?)
generic mechanism?
homologous at molecular, pathway level?

A generic regular pattern formation mechanisms
Turing Patterns

Can DIFFUSION create patterns from homogeneous state?

- 2 interacting substances
- stable homogeneous equillibrium in absence of diffusion
- unstable for spatial heterogeneous perturbations
- with diffusion: stable (+ regular) patterns

Turing patterns: formal requirements

$$\begin{cases} \frac{\partial A}{\partial t} &= D_a \Delta A + f_1(A,I) \\ \frac{\partial I}{\partial t} &= D_i \Delta I + f_2(A,I) \end{cases}$$

without diffusion stable:

$$trJ = a_{11} + a_{22} < 0$$
$$detJ = a_{11} * a_{22} - a_{21} * a_{12} > 0$$

with diffusion unstable

$$\begin{cases} a_{11} + a_{22} < 0 \\ a_{11} * a_{22} - a_{21} * a_{12} > 0 \\ D_a a_{22} + D_i a_{11} > 2\sqrt{D_a D_i * (a_{11} * a_{22} - a_{21} * a_{12})} > 0 \end{cases}$$

simplified requirements

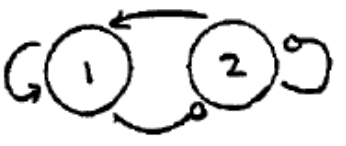
$$\begin{cases} a_{11} + a_{22} < 0 \\ a_{11} * a_{22} - a_{21} * a_{12} > 0 \\ D_a a_{22} + D_i a_{11} > 0 \end{cases}$$

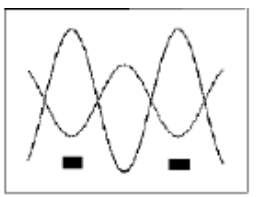
$a_{11} > 0$ and $a_{22} < 0$ $\quad \frac{D_i}{|a_{22}|} > \frac{D_a}{|a_{11}|}$

Diffusion I >> Diffusion A:

short range activation, long range inhibition

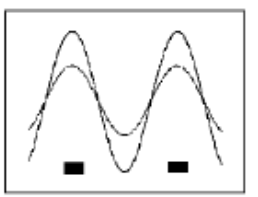
Positive feedback system : $\begin{pmatrix} + & + \\ - & - \end{pmatrix}$





Variables vary over space in phase:

Activator – inhibitor system : $\begin{pmatrix} + & - \\ + & - \end{pmatrix}$



Turing patterns

In 2D:

NB wavelength

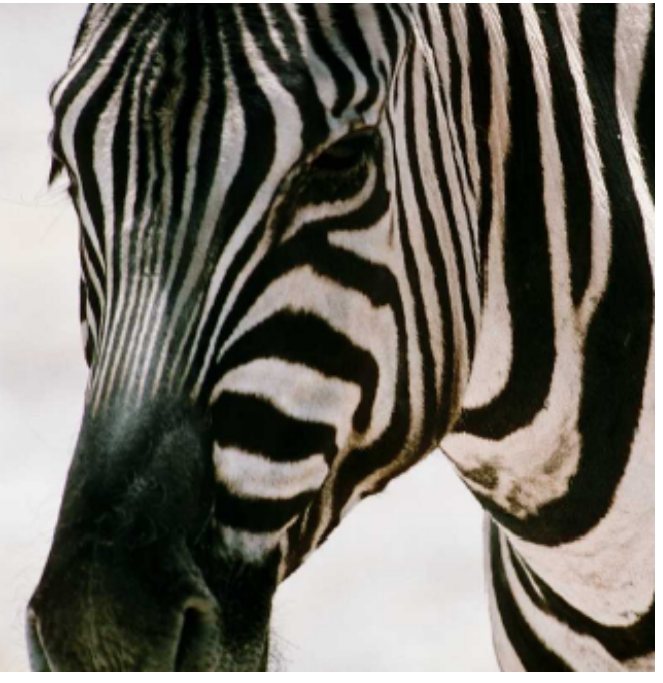
fitting in domain (selects largest eigenvalue)

regular patterns seen in e.g. coat patterns

Not only regular patterns,
but also domain dependence
shifting with irregular domains

Zebra: 'face recognition'

However sometimes "wrong"
small domain: spots;
large domain only 2 phases



"the stripes are easy, but what about the horse part?", Turing

applicable in Biology? If so HOW?

Strictly speaking:
Needs homogeneous initial state;
Needs diffusion
Needs large difference in diffusion;

HAS been sought but NOT BEEN FOUND

Less strictly speaking

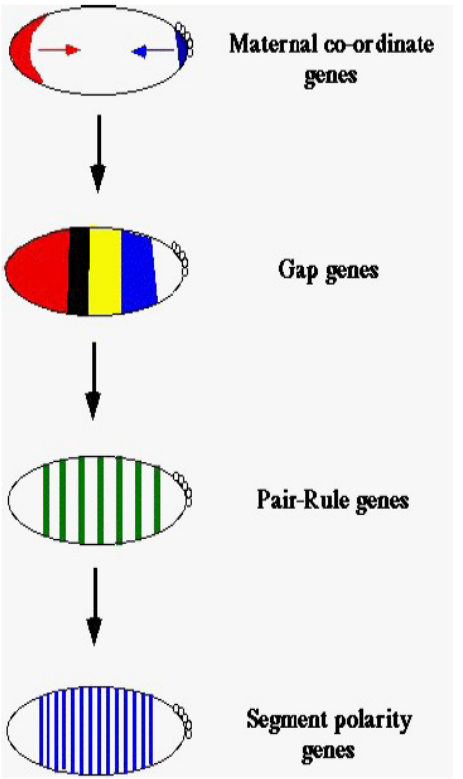
Needs SOME mechanism of
local activation / longer range inhibition

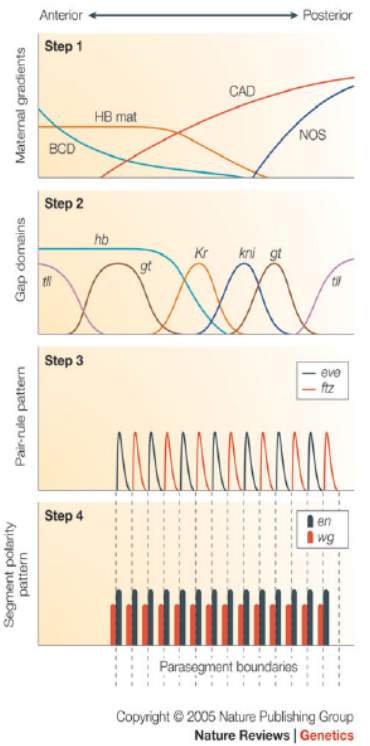
Classical Modeling Fallacy
Drosophila stripes as Turing patterns

Observe stripes

Turing instability $->$ stripes

Hence == Turing pattern

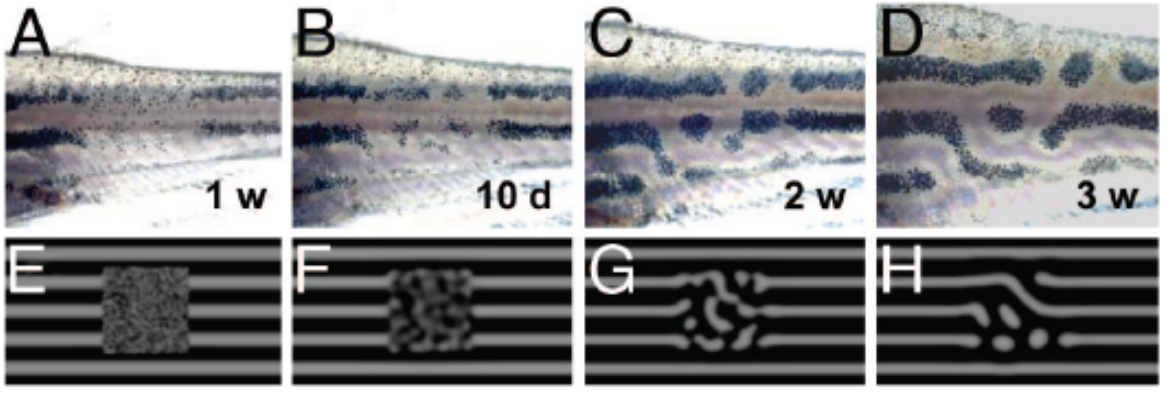


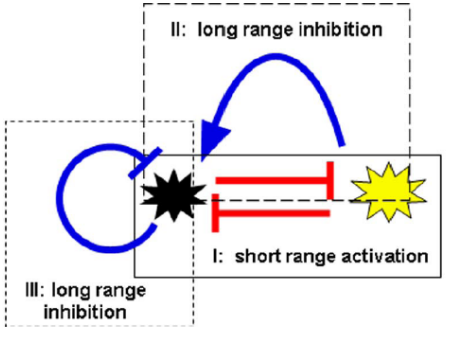


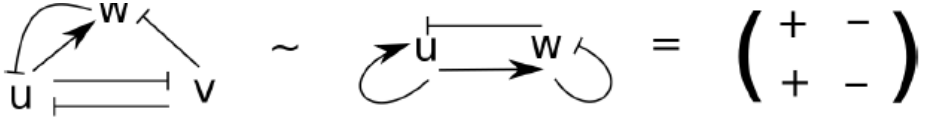
SHAME on "US" Math Biologists

Activation/inhibition scheme: fish stripes, Kondo-group

"looks like Turing patterns" (stripes)
"looks like turing patterns after ablation"
"short range activation, long range inhibition demonstrated by ablation experiments in pigment cells (no molecular interactions known)"



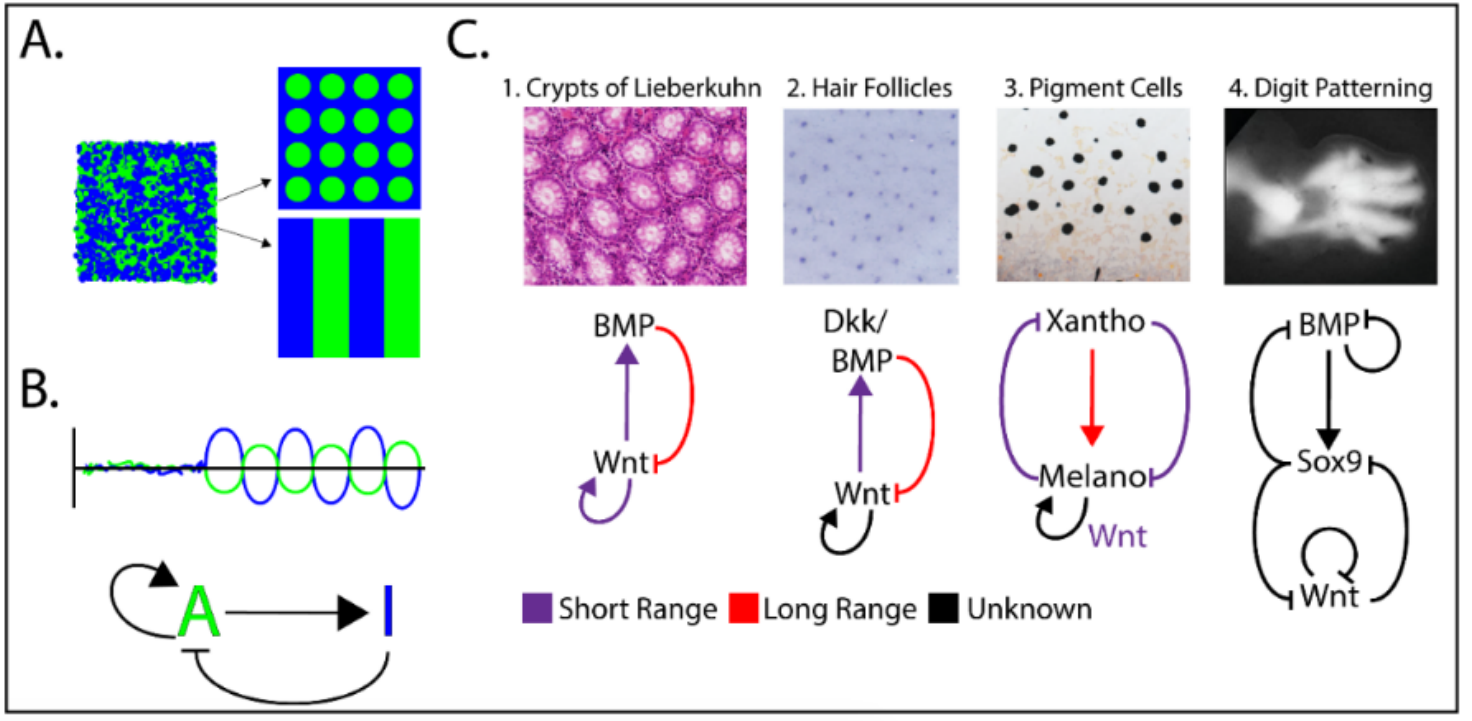




Interactions between zebrafish pigment cells responsible for the generation of Turing patterns
Nakamasua, Takahashia, Kanbea, Kondo PNAS 2008

KW Pond, K Doubrovinski, CA Thorne - Genes, 2020

Molecular mechanisms and Turing patterns in various systems



Wnt β-catenin Signaling in Tissue Self-Organization
KW Pond, K Doubrovinski, CA Thorne - Genes, 2020

Pattern formation with larger networks long range diffusion not neccessary

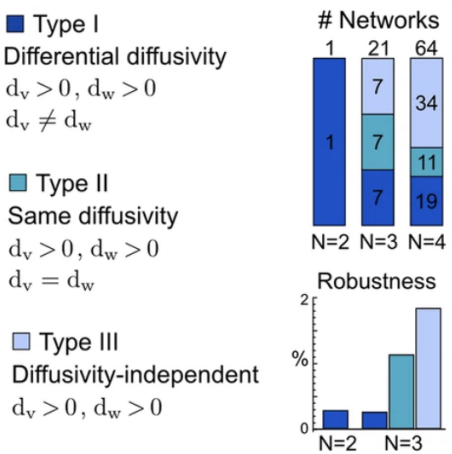
- Originally the condition of high diffusion rates of inhibitors was seen as a problem;
- However several mechanisms identified, e.g. active transport and release by mechanical forces.
- The larger experimentally determined networks reduced to 2-node networks to fit into Turing framework (see above)
- when Turing-like patterns observed, diffusion conditions assumed)

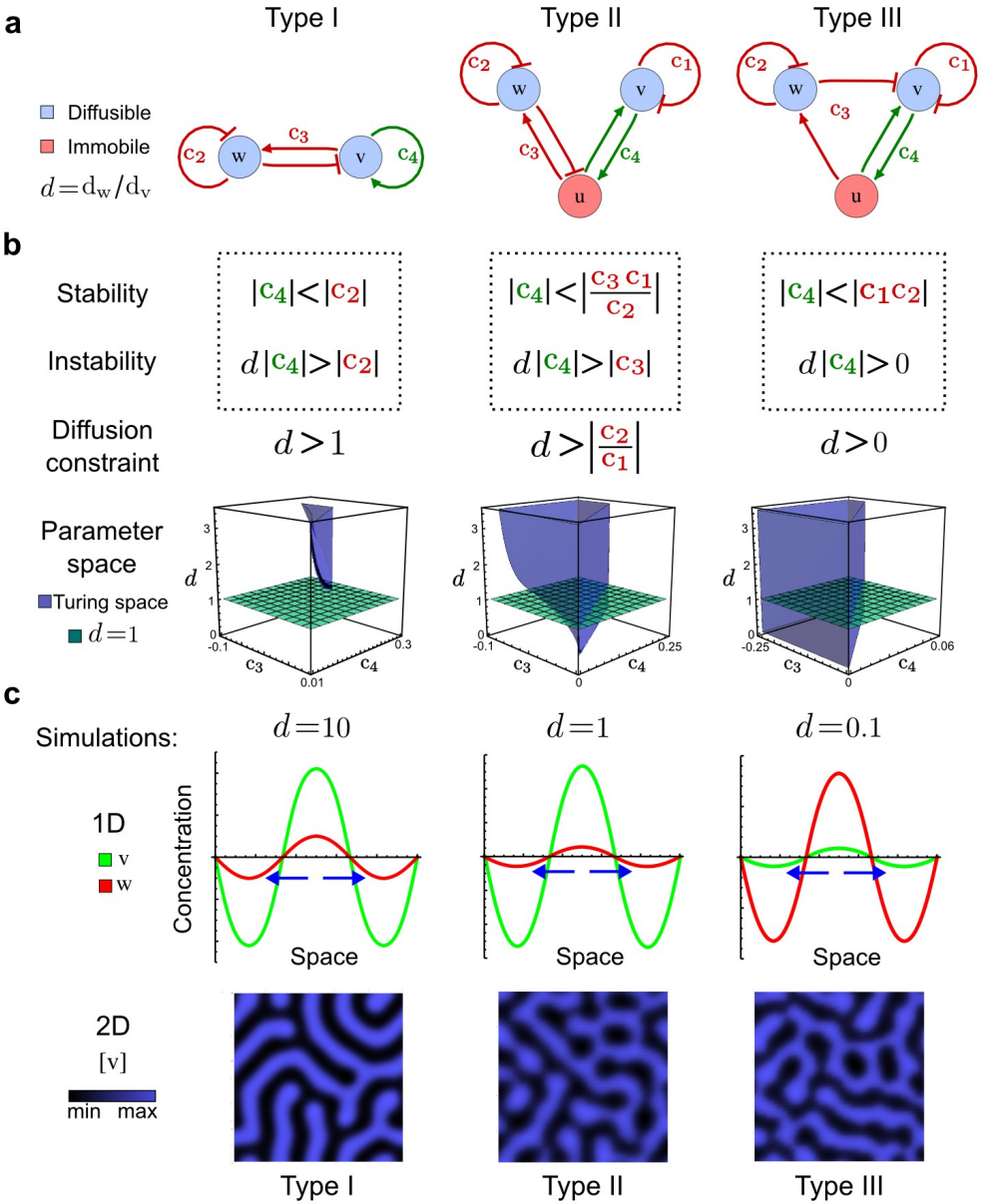
Do the same conditions hold for the larger networks??

NO...

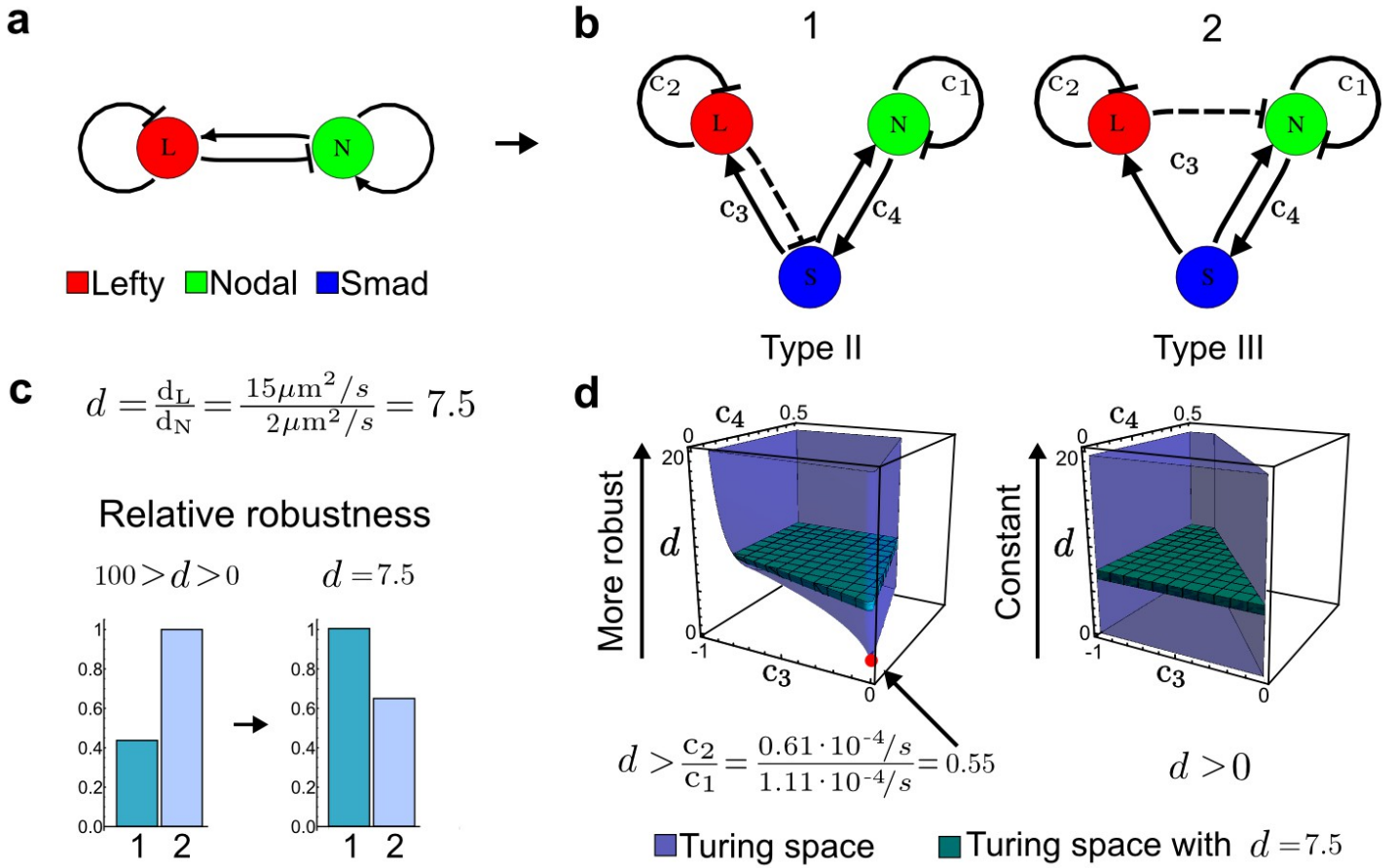
High-throughput mathematical analysis identifies Turing networks for patterning with equally diffusing signals L Marcon, X Diego, J Sharpe, P Müller Elife, 2016

Networks analysed as feedback loop positive/negative conditions for stability/instability

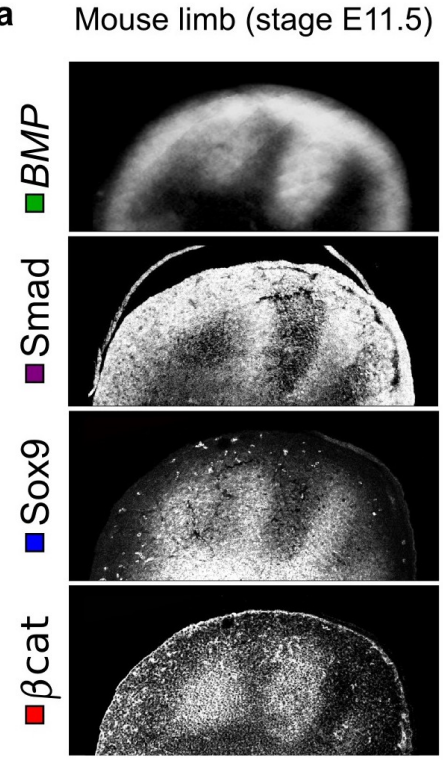


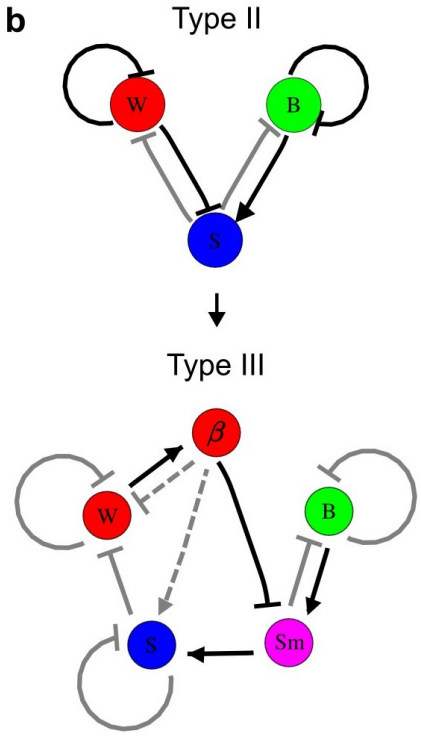


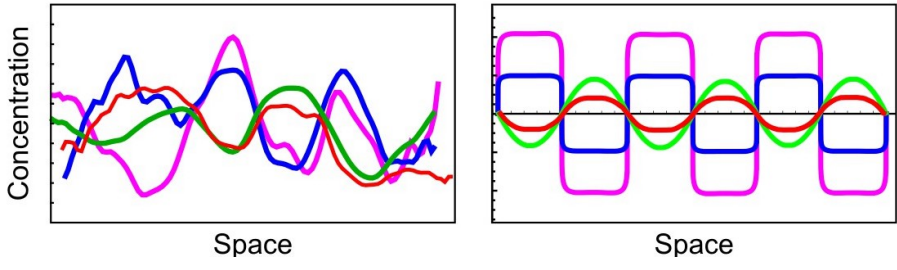
Example Nodal/Lefty network (germlayers & left/right in vertibrates)



Example: Mouse digit formation: hypothesise interactions







note antiphase B, SM

From squeezing known interaction into 2D Turing model to

Find network including known interactions which allows

Turing instabilitay

Conclusions Turing Patterns
how to use (or misuse) general mechanism to elucidate specific pattern formation mechanisms

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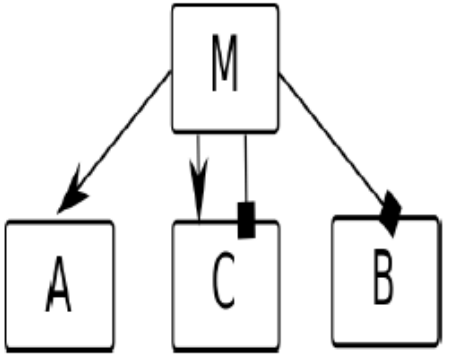
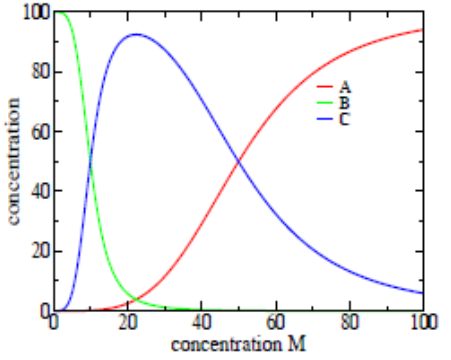
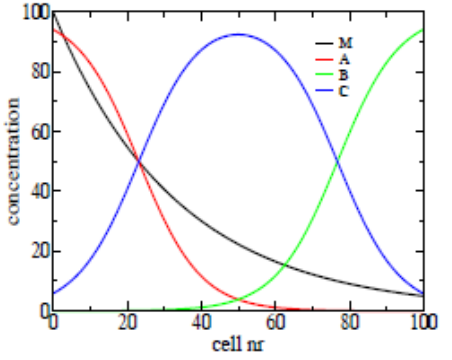
- Elegant, very general
- beyond Original diffusion $->$ pattern
- However Stripes: too degenerate pattern to infer anything (needs $++$)
- Domain / disturbance variations more informative
- However random positioning - but may be tweaked
- Often invoked, eg. limb$->$digitis including special conditions (cd possitional information, see below)
- Molecular mechanisms elucidated: similarities/differences
- Conditions on diffusion relaxed for larger networks
- Prepattern for and influenced by tissue deformation/cell movement
- Also used for vegetation patterns

Positional information/ french flag model
Wolpert 1969

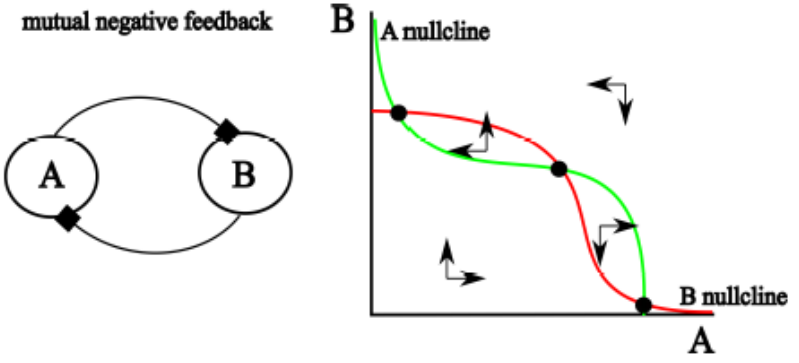
"French flag":
different morphogen concentrations → activate different genes







Alternative attractors:
maintain expression domains when morphogen gradient disappears



Positional information/ french flag problem
Wolpert 1969

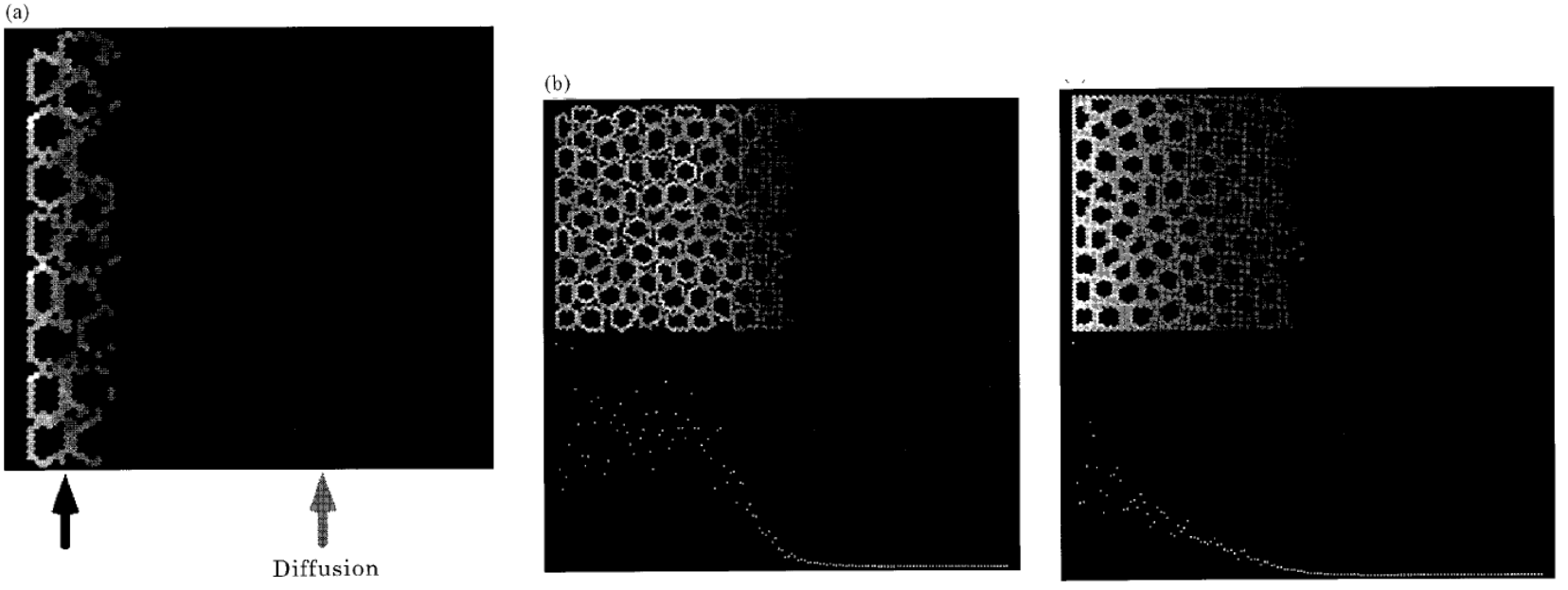
Source/sink/diffusion for gradient formation
'read-out' of concentration $->$ cell differentiation
(stabilization by mutual inhibition)

french flagproblem: how to be scale invariant?

source/sink diffusion is scale invariant!
(but not a likely solution...)

problems: spatial/temporal scaling of diffusion
in tissue: cell boundaries may not allow gradients
how to have precise quantitative readout?
"simple mechanism may not be simple"
noise
"pathways which produce and use positional information"

receptors disturb gradient cf Kerzberg and Wolpert 1998



several potential solutions proposed

early patterning in Drosophila
Model 1: gap gene expression in Drosophila (pre-gastrulation / pre cellularizatrion)

paradigm system for positional information

Maternal gradient (Bicoid) (measured)
In syncytium stage (no cell walls to pass)

paradigm system for data driven quantitative modeling

Very precise description of pattern in space/time available

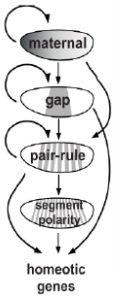
Much experimental knowledge about genes involved and their interactin

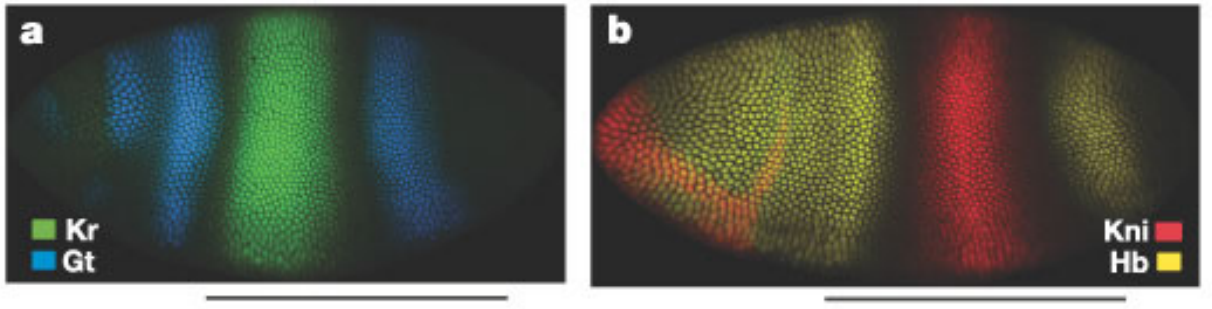
many papers main authors J. Reinitz anf J. Jaeger; here used:

Manu, ... Reinitz 2009 Canalization of Gene Expression in the Drosophila Blastoderm by Gap Gene Cross Regulation, Pos Biology

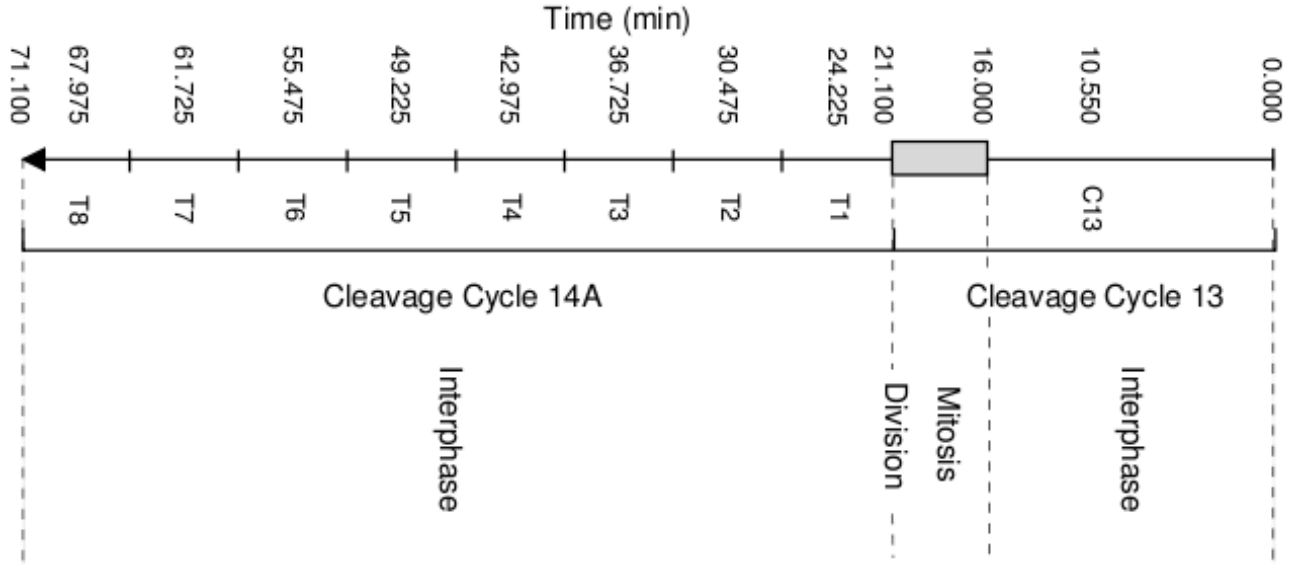
J.Jaeger .. Reinitz 2004.Dynamic control of positional information in the early Drosophila embryo Nature

modelled space-time frame





gap gene expression in late stage: black line: modeled area



modeling gene regulation: ODE for each nucleus

$$\frac{dv_i^a}{dt} = R^a g\left(\sum_{b=1}^{N} T^{ab} v_i^b + m^a v_i^{\mathrm{Bcd}} + \sum_{\beta=1}^{N_e} E^{a\beta} v_i^\beta(t) + h^a\right) + D^a(n)\left[(v_{i-1}^a - v_i^a) + (v_{i+1}^a - v_i^a)\right] - \lambda^a v_i^a.$$

T interaction between gap genes; m interaction with bicoid;
E interaction of gap genes with time varying external factors; λ decay; D diffusion

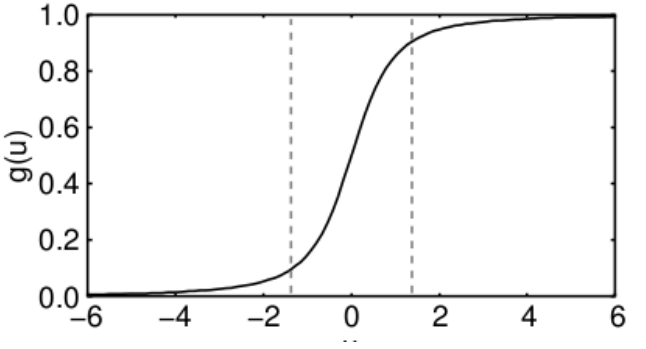
interphase: production, diffusion and decay;

mitosis: only diffusion and decay

division: nuclei divide, inherit state,

distance between them halved

transcription: $g(u^a) = \frac{1}{2}\left[\left(u^a/\sqrt{(u^a)^2+1}\right)+1\right]$



"data driven modeling": massive fitting using simulated annealing

use: 'known genes", initial conditions, spatial/temporal variaton of non-regulated regulators.

Fit model output in all M nuclei, for all genes, at all N timepoints for which data are available.

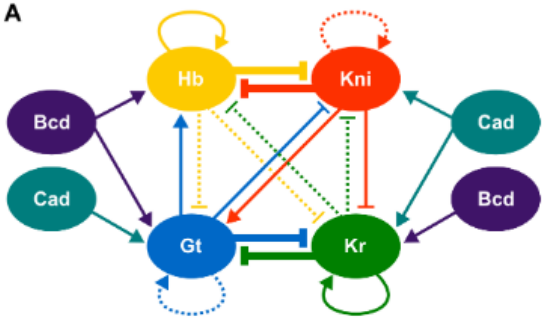
$$E = \sum_{\substack{\text{all } a, i, t, \text{ and} \\ \text{genotypes for} \\ \text{which data exists}}} \left(v_i^a(t)_{\text{model}} - v_i^a(t)_{\text{data}}\right)^2 + (\text{penalty terms})$$

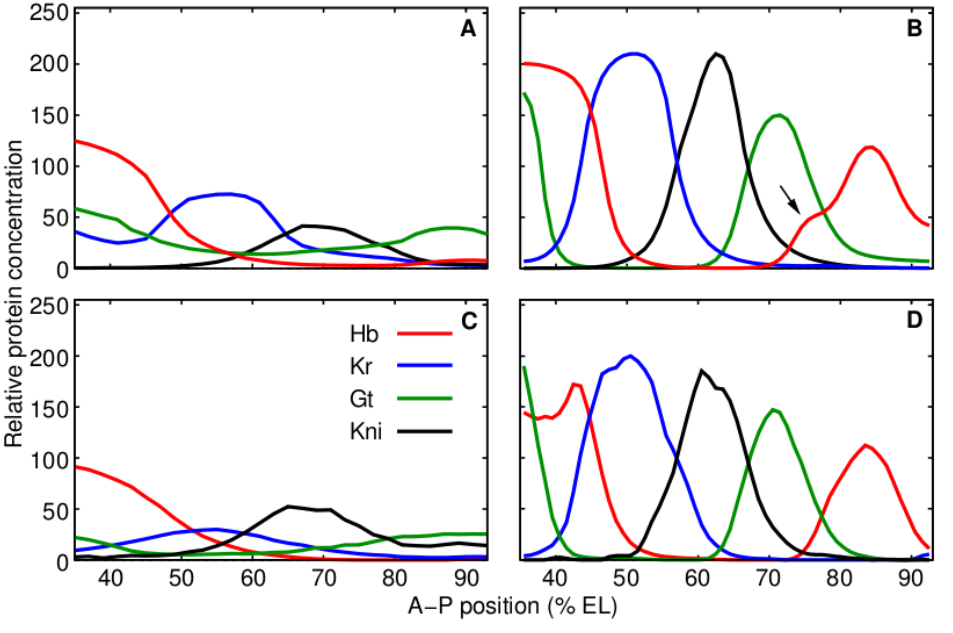
Do this Z=65 times gives Z different outcomes; and select good fits, no major patterning defects, no known regulatory mistakes (23/65) similar networks

used example of 'good' network

Target gene a	Regulator gene b						
	bcd	*cad*	*tll*	*hb*	*Kr*	*gt*	*kni*
hb	0.025	0.004	0.003	0.021	–0.001	0.022	–0.112
Kr	0.118	0.021	–0.203	–0.026	0.035	–0.042	–0.062
gt	0.256	0.023	–0.011	–0.028	–0.202	0.007	0.003
kni	0.012	0.020	–0.187	–0.082	0.000	–0.017	0.013

Parameter	Gene a			
	hb	*Kr*	*gt*	*kni*
R^a	15.000	10.354	15.000	15.000
D^a	0.166	0.200	0.103	0.200
$t^a_{1/2}$	9.529	15.908	9.438	13.062



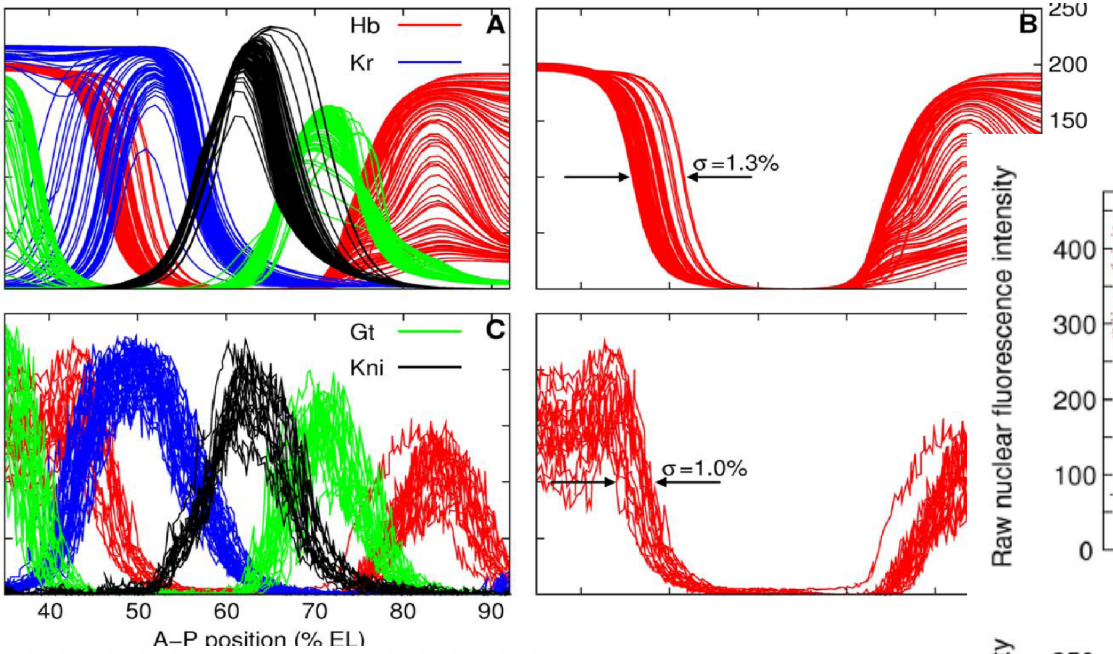


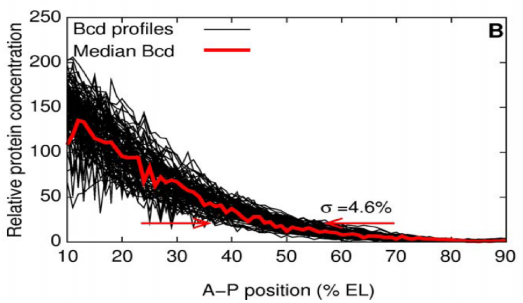
above: model: early - late; below av. exp. early-late

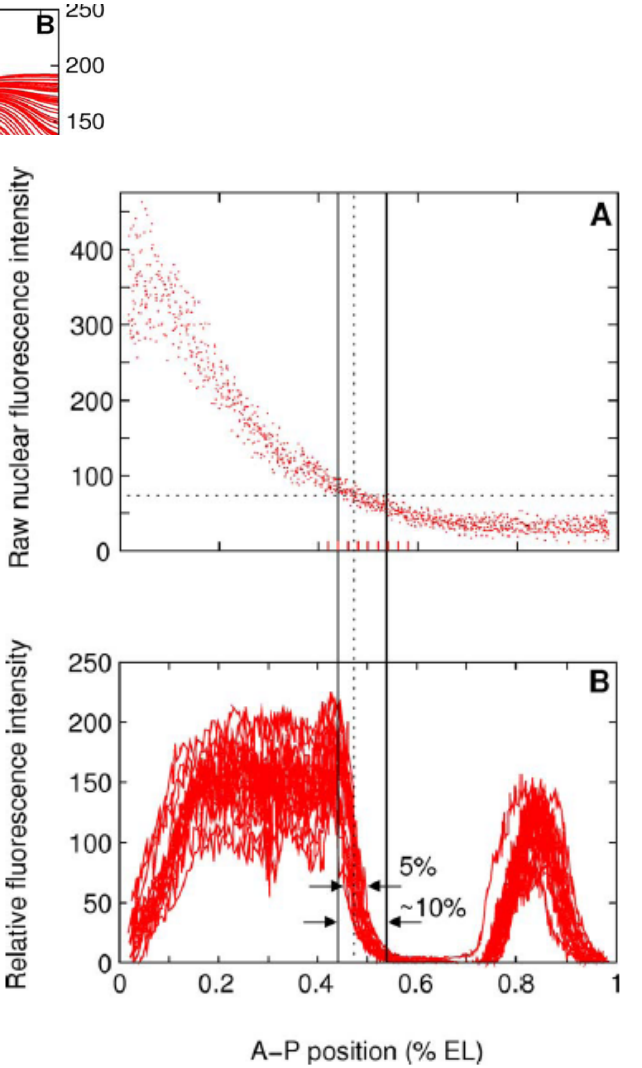
classical question
developmental patterning very precise, despite differences in
e.g. size of embryo or gradient noise

Manu et al 2009: is due to regulatory circuit.

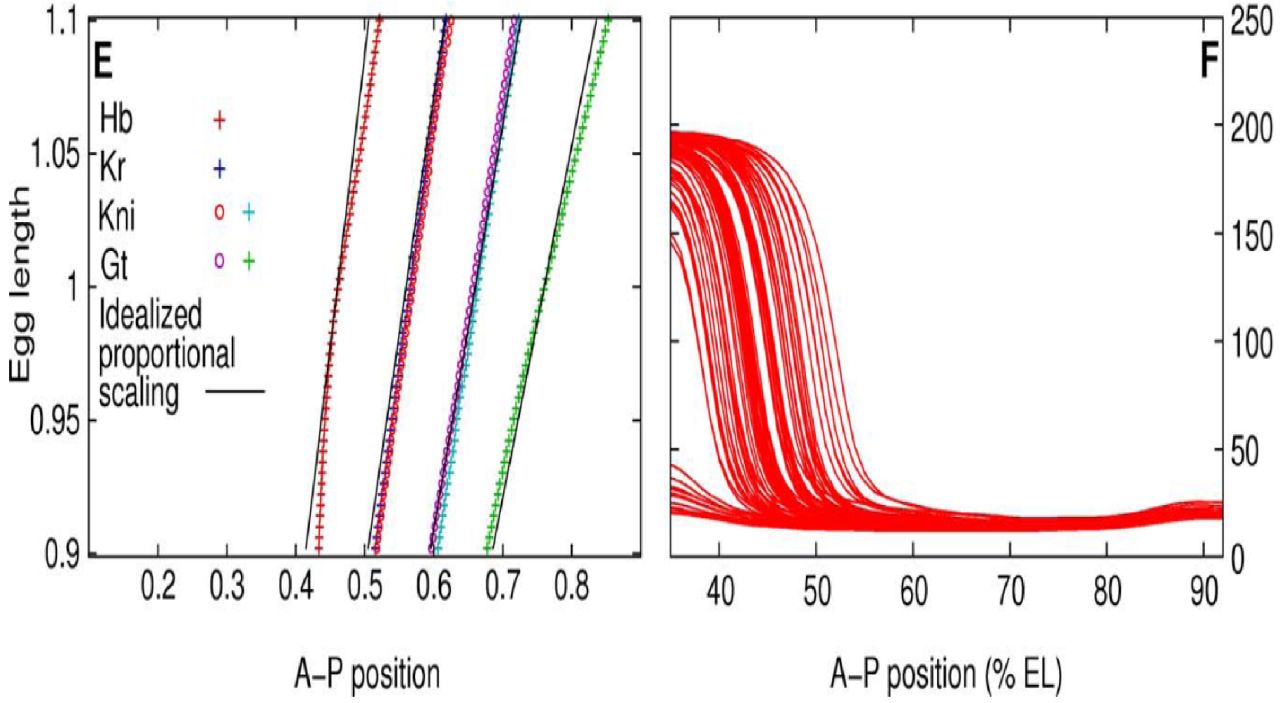
robustness to variation in bicoid gradient





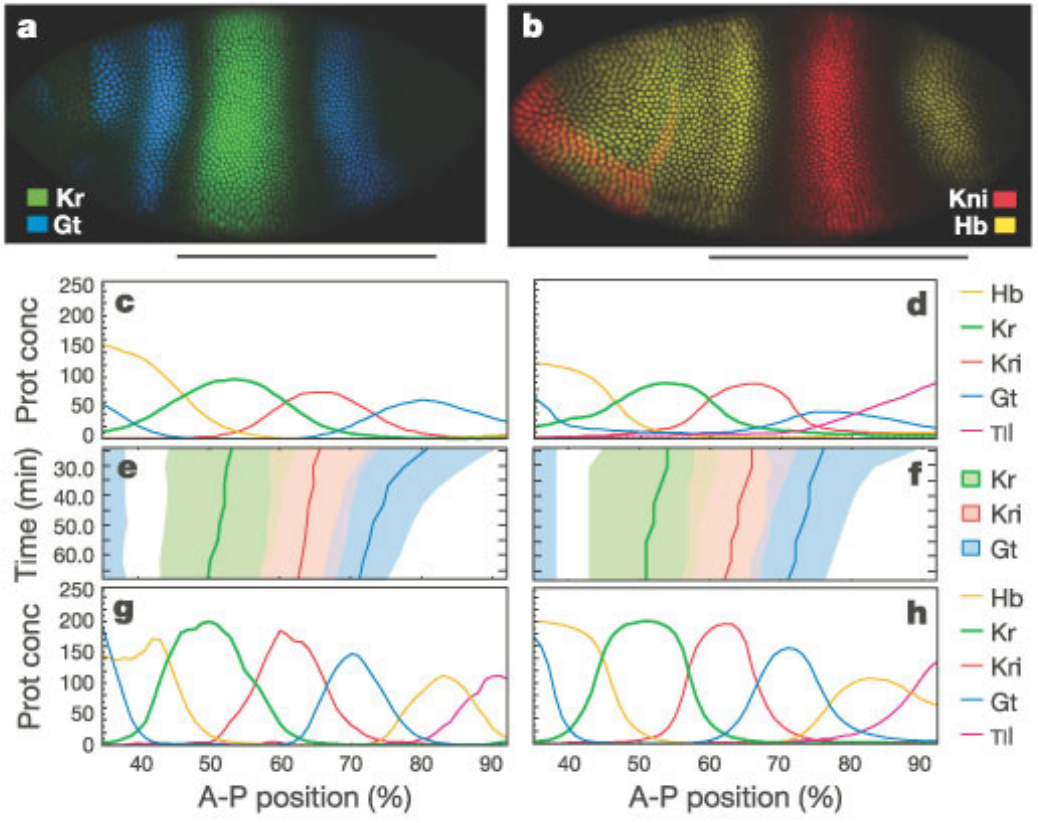


robustnes to size varation (20%)

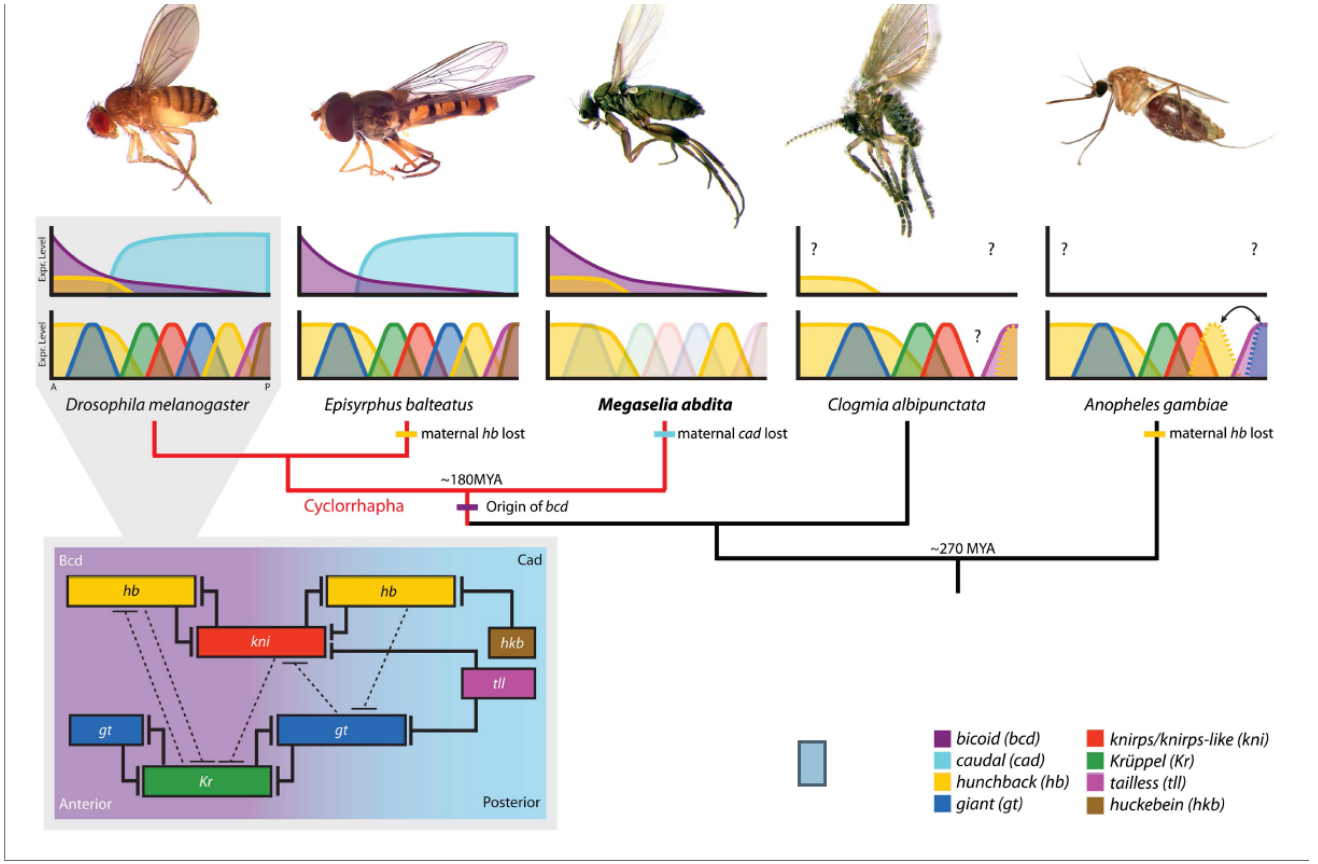


without cross regulation gap-genes

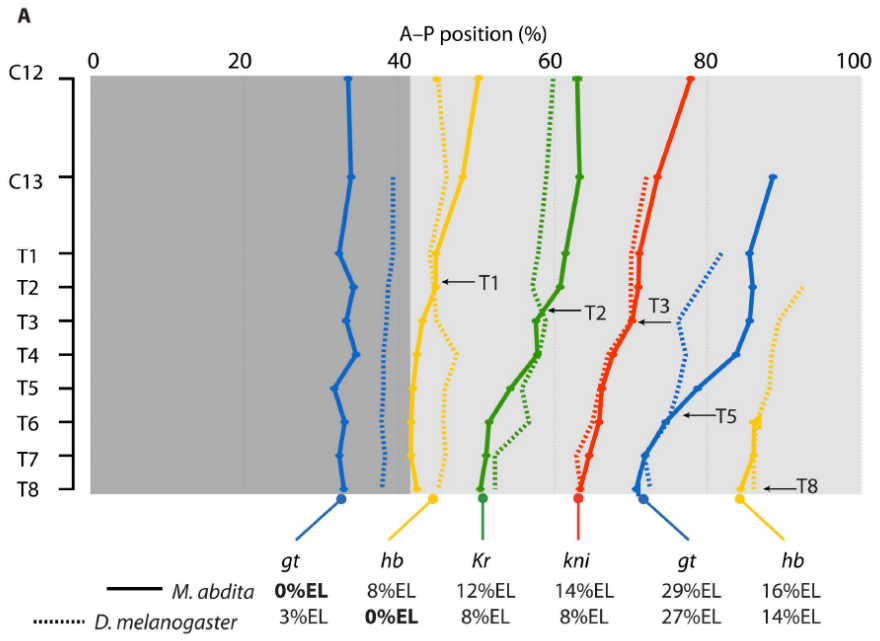
model also reproduces shfts in expression patterns over time
Jaeger et al 2004 op.cit

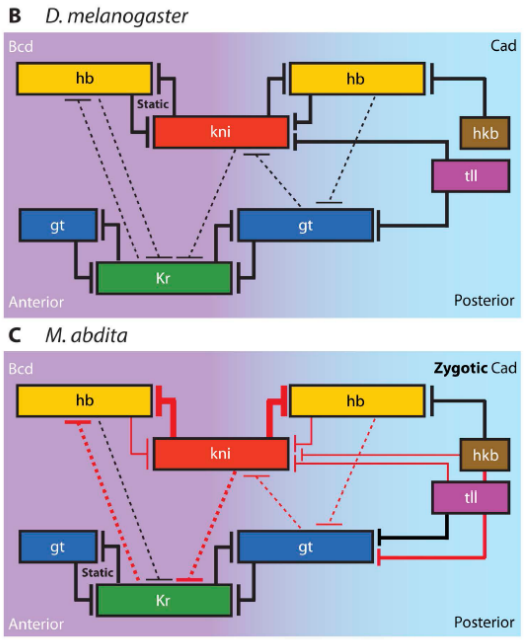


"Quantitative system drift compensates for altered maternal inputs to the gap gene network of the scuttle fly Megaselia abdita" Wotton et al eLife 2015



Some, but only tiny differences in expression patterns





discussion/conclusions

Fitting not very robust:
alternative "as good" fits with even opposite signs of interaction
(filtered to agree with experimental knowledge)

because of shifting "better" fitting because less degenerate

supervised models: Fits

++ = scaling property and noise reduction

++ insight in evolutionary drift / compensation in conserved patterning

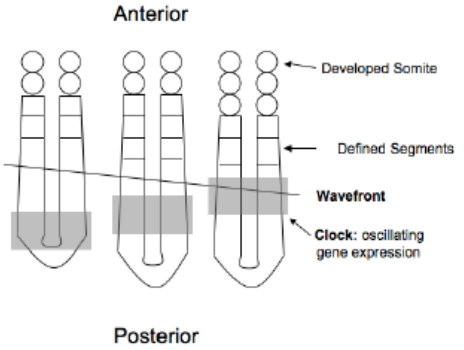
Positional information (?):

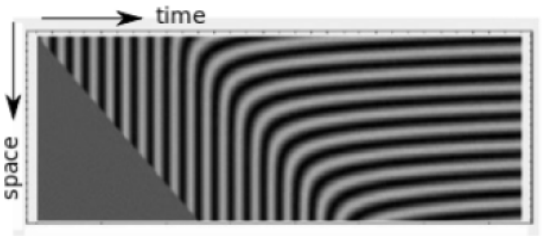
yes - gradient given and provides "coordinate system"

no - not simple concentration readout
readout itself 'makes the pattern'

scale invariant (tolerant) because of regulation / not invariant bicoid gradient

a common mechanism in segmentation development in many organisms
clock and wavefront mechanisms
from temporal to spatial pattern
Cooke and Zeeman 1976

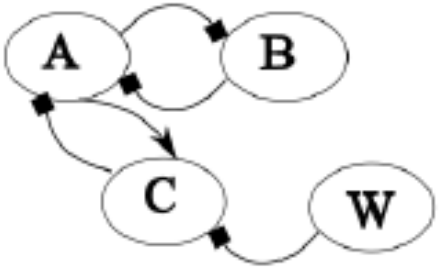


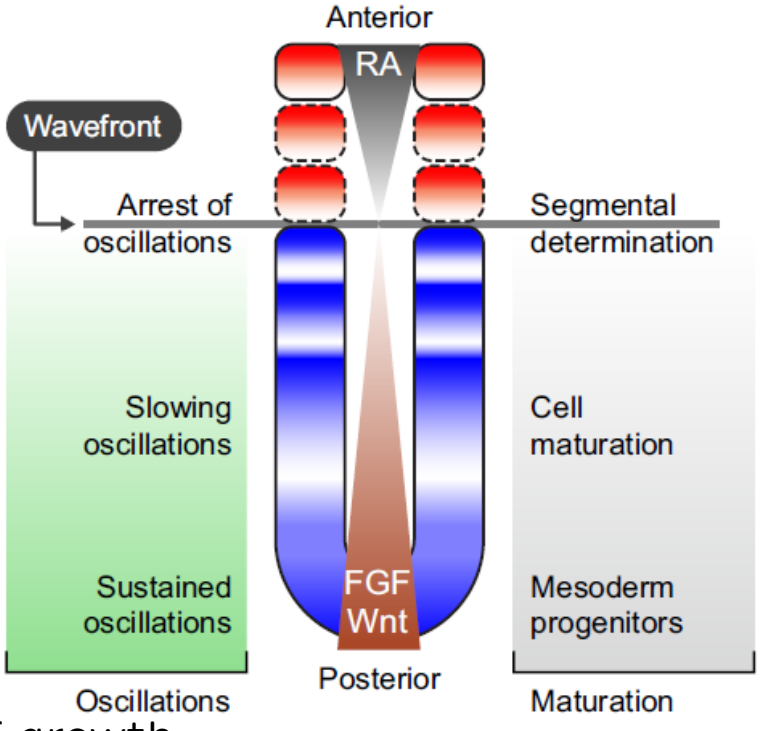


clock:
internal cellular oscillations, phase synchronized between cells
wavefront:
competence wave moving from anterior to posterior at constant speed

gradients which appear to play a role

"arrest" can be autonomous
(Hopf or other bifurcation
or extern
because of bistability
Goldbeter 20..



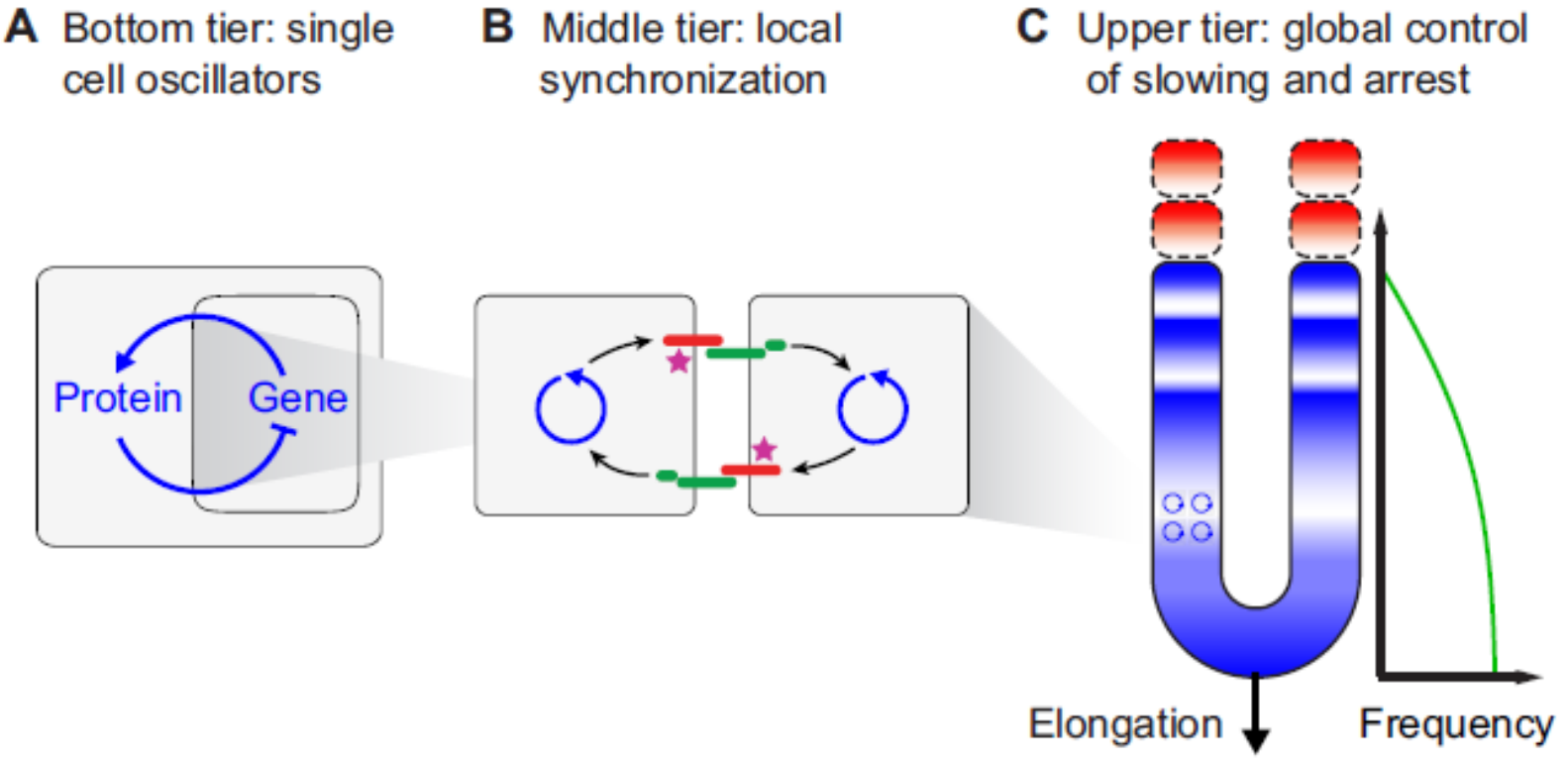


distance governed by posterior rate of growth

similar result .

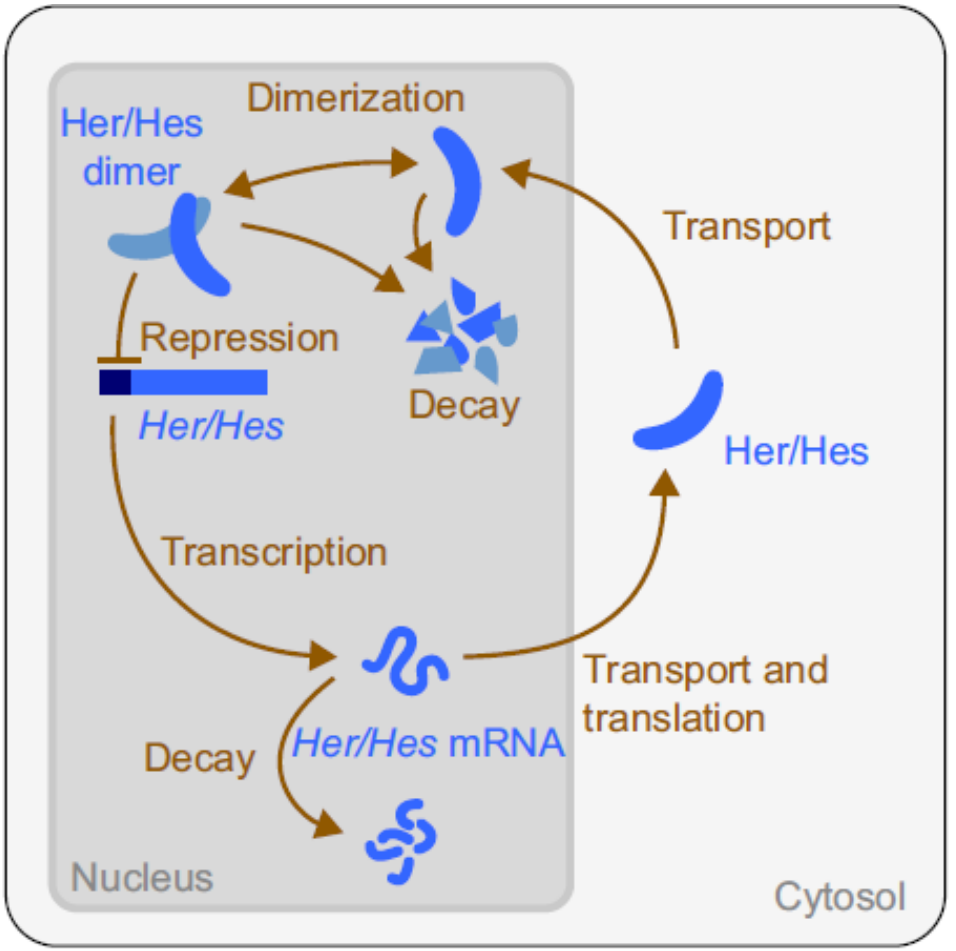
resistant to noise

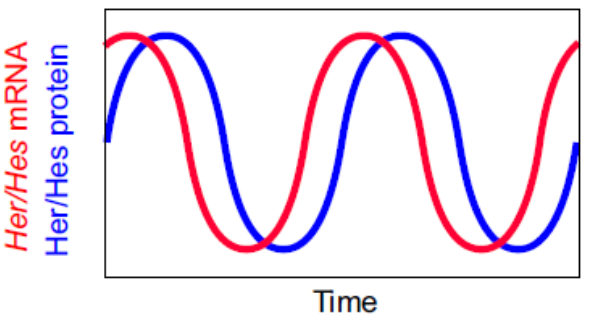
proposed "implementation" as 3 tier mechanism in somitogenesis



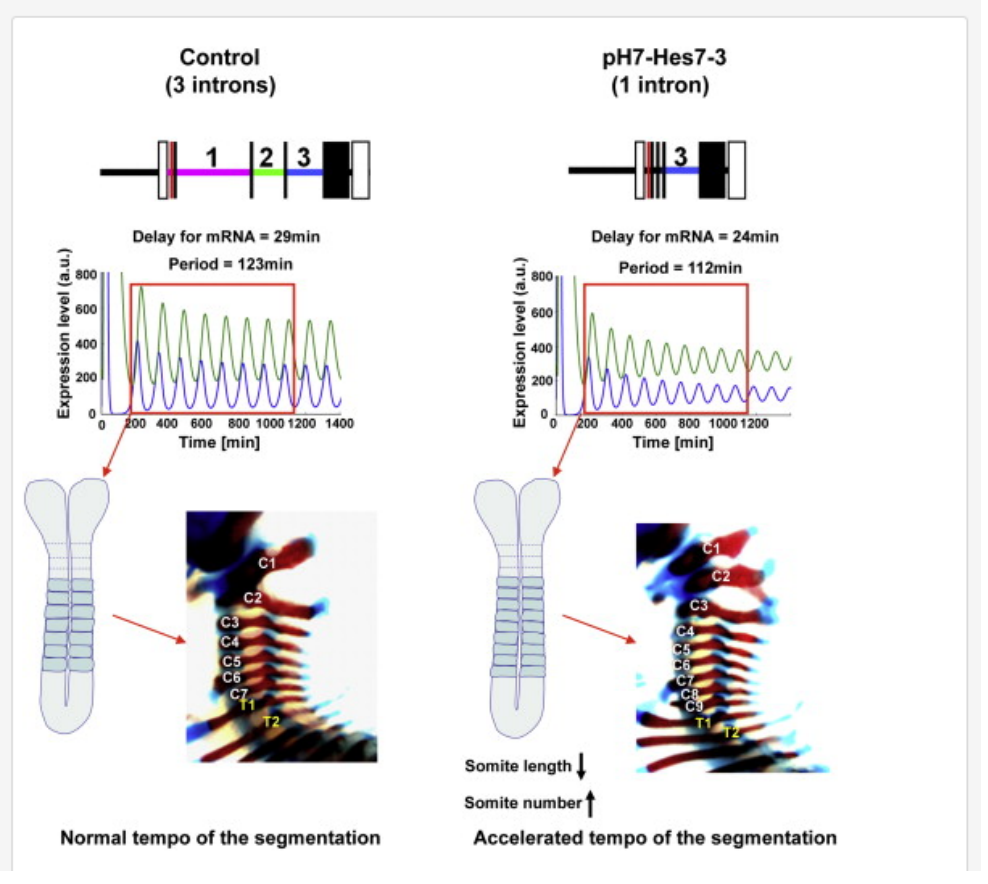
l cf Patterning embryos with oscillations: structure, function and dynamics of the vertebrate segmentation clock AC Oates, LG Morelli, S Ares Development, 2012

single cell oscillator: delayed auto-feedback systems



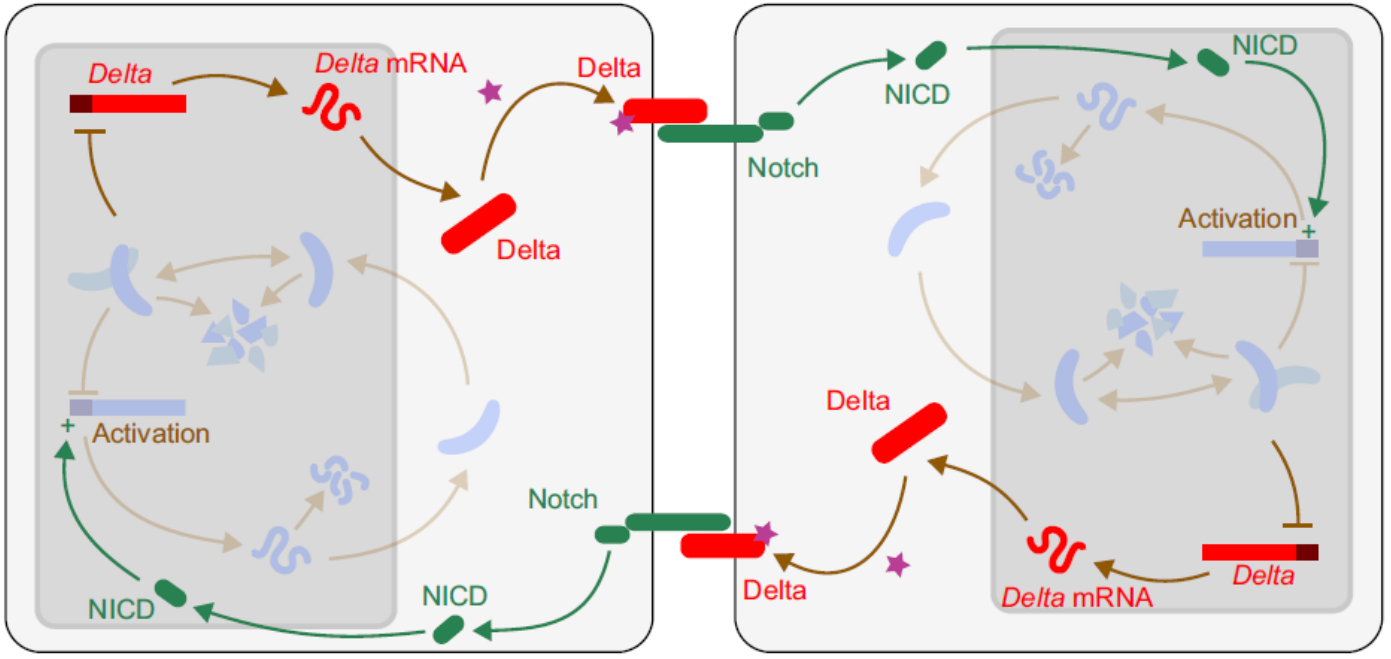


delay detemines number of segments
indeed: intron deletion speeds up the clock
Harima et al Cell 2012

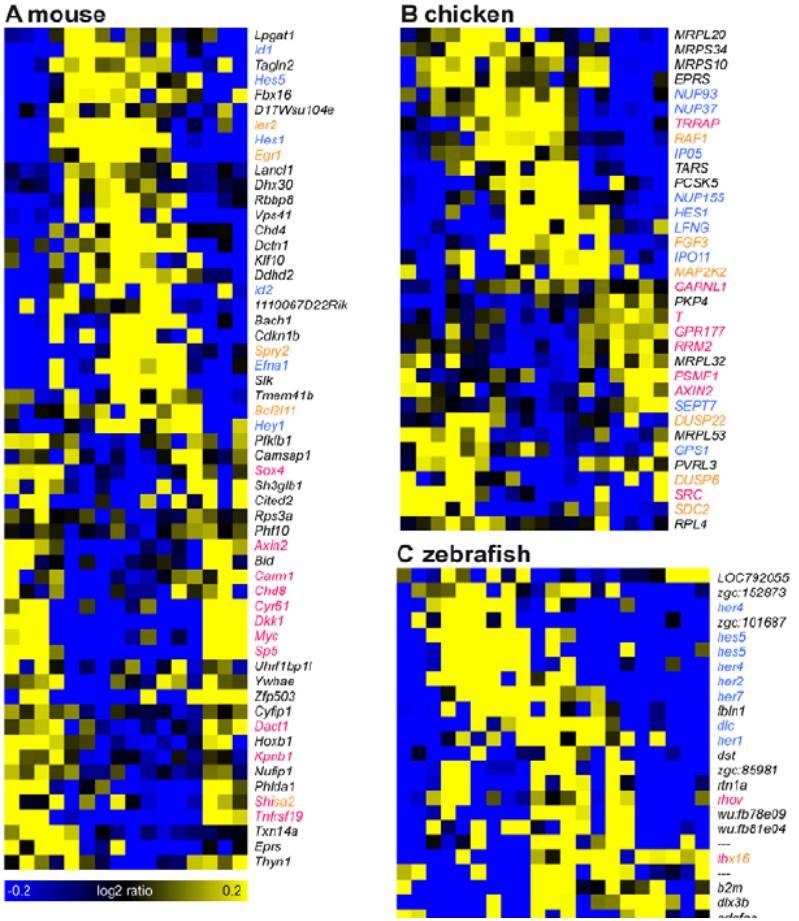


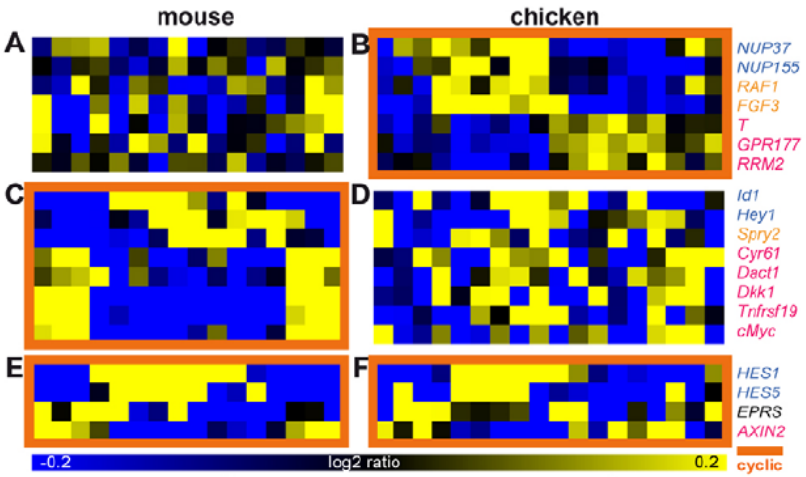
neighbour synchronization: Period is tissue-wide property

can shorten of lengthen period of single cells



reinvented or conserved, which genes oscillate?
GO terms: signalling and transcription





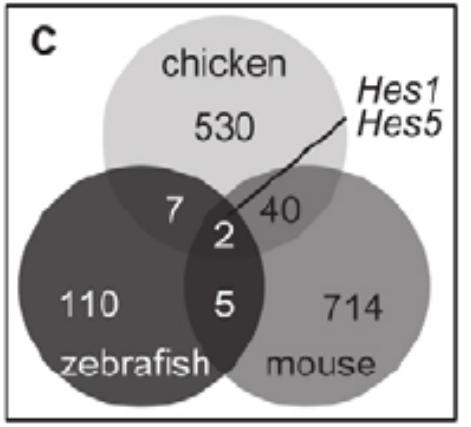
Krol et al Development 2011

orthologs

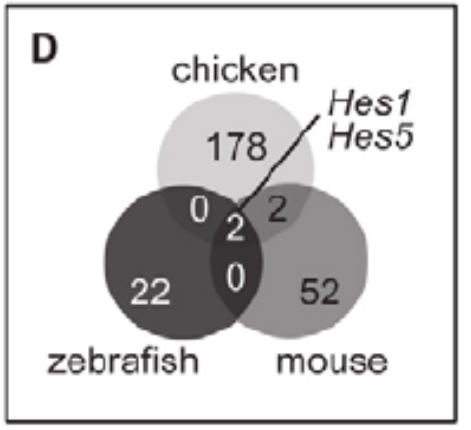
Evolutionary plasticity of segmentation clock networks

Only 2 overlapping orthologs involved in segmentation clock

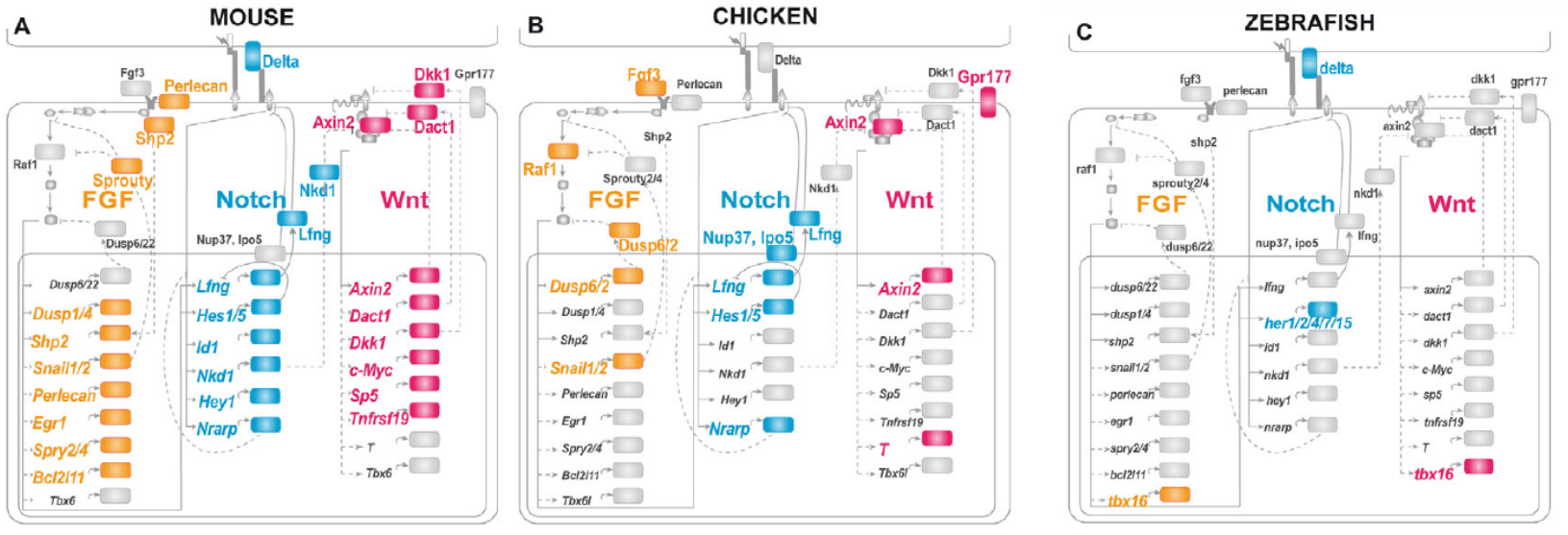
first estimate:



after filtering:



Only 2 orthologs: but members of 3 pathways in all



(this analysis first to find member WNT pathway n zebra fish)

conclusion: very high plasticity!

Only small subset of the 3 pathways oscillate:
enough for functional oscillations?
"just in time assembly")

Similar (non) conservation pattern in cell cycle mechanisms yeast and pombe

Conservered HER/HES delayed oscillator also in
medaka, Xenopus, and invertebrates (e.g.cockroach)!!

Segmentation lost? reinvented?

Is segmentation "the same" in the different organisms??

RA knockout leads to asymetric somatogenesis
which is different for different vertebrate species
HOW/WHY??
Model in more detail to find out which difference in regulatory network may explain differnce in phenotype of RA knockouts

R.M.A. Vroomans, K.H.W.J. ten Tusscher

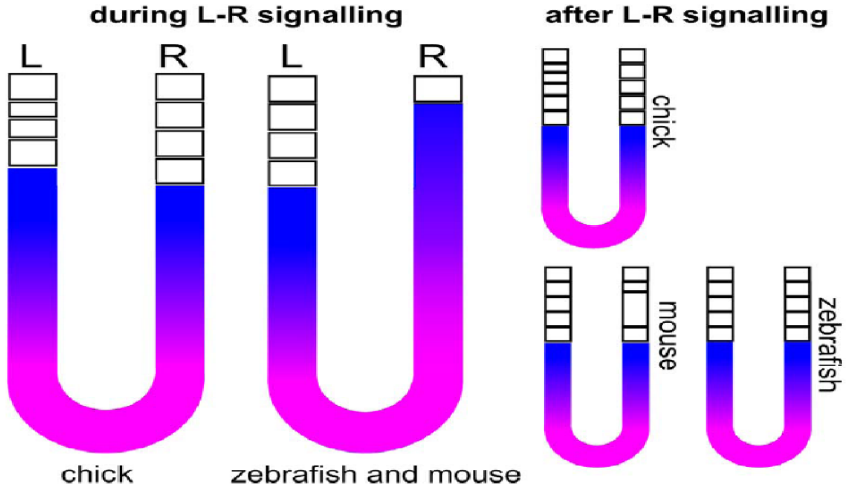
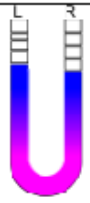
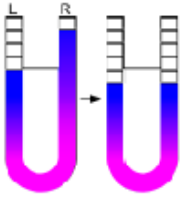
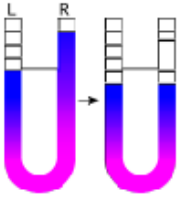


Table 3.1. phenotypes of model organisms during somitogenesis

genetic properties			left-right asymmetry in absence of RA					
organism	pErk dynamics	oscillating pathways	left-right phenotype	Slower osc	FGF8	delay (somite nr)	somite size diff	return to symmetry
chick	smoothly retracting	FGF, Wnt, Notch		right side	symmetric, more anterior	no; left somites smaller	yes	unclear
zebrafish	retracts in jumps	Notch		right side	right side more anterior	right side 2-3 somites delayed	no	yes
mouse	oscillates	FGF, Wnt, Notch		right side	right side more anterior	right side 2-3 somites delayed	sometimes	yes

Vroomans & ten Tusscher 2017,Modelling asymmetric somitogenesis: Deciphering the mechanisms behind. species differences

Generic mechanism vs species specific differences neutral drift or functional significant???

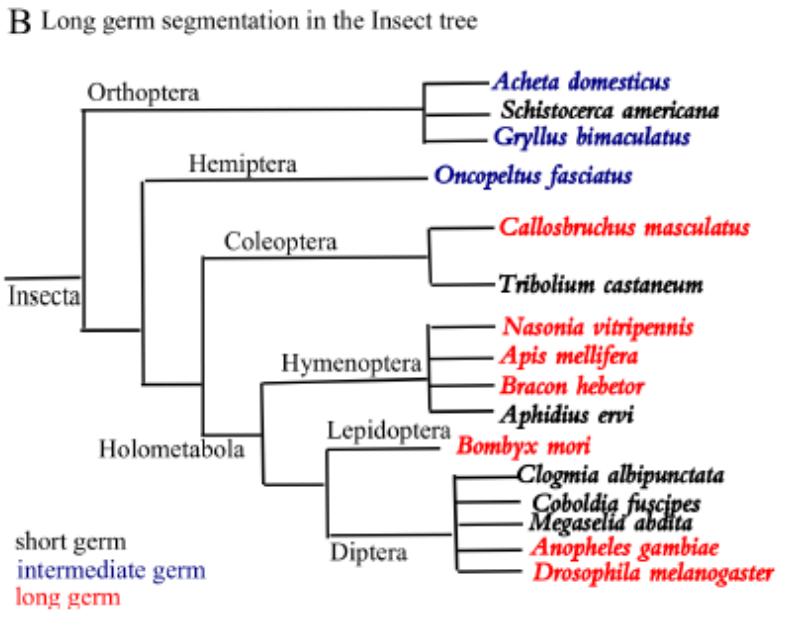
Vroomans & ten Tusscher 2017:

Indeed, our results suggest that rather than focussing on a catch-all mechanism in all vertebrate species and assuming that species differences merely reflect neutral developmental systems drift, we should keep an open mind for the possibility of functionally significant species differences.

OR

Side-efects of neutral drift

But what about Drosophila?
2 (3) mechanisms in insects short vs long germband (+intermediate)



clock-wavefront (sequential) mechanism might be ancestral - reinvention of simultaneous mechanism long germband??

conclusions: themes

General mechanisms of pattern formation

vs multiple realizations

evolutionary drift

Divergent vs convergent evolution ???

Supervised modeling:

Parameter fitting vs model search

Minimodels - and assumed neccesary conditions

results ++