modeling development

classical models of pattern formation
segmentation patterns; STRIPES
One more level between genotype and phenotype: Modeling development (and its evolution)

Pattern formation (dependent on Shape)

Pattern formation \(-\ldots\rightarrow\) Shape

Pattern formation \(<\ldots\rightarrow\) Shape

TODAY: Classical models of pattern formation / segmentation

*Supervised modeling*

Top down modeling:
- Given observed pattern/behaviour X and assumptions A
  CAN A \(-\ldots\rightarrow\) 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 $\rightarrow$ cell differentiation $\rightarrow$ 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 oscillation $\rightarrow$ spatial pattern

compare: “pattern is 'default”

however here specific positioning/orientation in continuous 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 equilibrium in absence of diffusion
• unstable for spatial heterogeneous perturbations
• with diffusion: stable (+ regular) patterns
Turing patterns: formal requirements

\[
\begin{align*}
\frac{\partial A}{\partial t} &= D_a \Delta A + f_1(A, I) \\
\frac{\partial I}{\partial t} &= D_i \Delta I + f_2(A, I)
\end{align*}
\]

without diffusion stable:

\[
\text{tr} J = a_{11} + a_{22} < 0
\]
\[
\text{det} J = a_{11} * a_{22} - a_{21} * a_{12} > 0
\]

with diffusion unstable

\[
\begin{align*}
a_{11} + a_{22} &< 0 \\
a_{11} * a_{22} - a_{21} * a_{12} &> 0 \\
D_a a_{22} + D_i a_{11} &> 2 \sqrt{D_a D_i * (a_{11} * a_{22} - a_{21} * a_{12})} > 0
\end{align*}
\]
simplified requirements

\[
\begin{align*}
    a_{11} + a_{22} &< 0 \\
a_{11} a_{22} - a_{21} a_{12} &> 0 \\
D_a a_{22} + D_i a_{11} &> 0
\end{align*}
\]

\[a_{11} > 0 \text{ and } a_{22} < 0 \quad \frac{D_i}{|a_{22}|} > \frac{D_a}{|a_{11}|}\]

Diffusion I >> Diffusion A:
short range activation, long range inhibition

Positive feedback system: \((- + (-)

Variables vary over space in phase:

Activator – inhibitor system: \((+ - +)

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 $\Rightarrow$ stripes

Hence $\Rightarrow$ 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
Wnt β-catenin Signaling in Tissue Self-Organization
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
domain, wavelength and number of digits

On the Formation of Digits and Joints during Limb Development Hiscock et al 2017
Conclusions Turing Patterns

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

Prepattern for and influenced by tissue deformation/cell movement

Also used for vegetation patterns
“French flag”:
different morphogen concentrations → activate different genes

Alternative attractors:
maintain expression domains when morphogen gradient disappears
Source/sink/diffusion for gradient formation
'read-out' of concentration \(\implies\) cell differentiation
(stabilization by mutual inhibition)

french flag problem: 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 cellularization)

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, .... Reinititz 2009 Canalization of Gene Expression in the Drosophila Blastoderm by Gap Gene Cross Regulation, Pos Biology
J.Jaeger .. Reinitiz 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

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

T interaction between gap genes; m interaction with bicoid; E interaction of gap genes with time varying external factors; \( \lambda \) 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 variation of non-regulated regulators.

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

\[ E = \sum_{all \ a, \ i, \ t, \ and \ genotypes \ for \ which \ data \ exists} (v^a_i(t)_{model} - v^a_i(t)_{data})^2 + \text{(penalty terms)} \]

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

<table>
<thead>
<tr>
<th>Target gene (a)</th>
<th>(bcd)</th>
<th>(cad)</th>
<th>(tll)</th>
<th>(hb)</th>
<th>(Kr)</th>
<th>(gt)</th>
<th>(kni)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(hb)</td>
<td>0.025</td>
<td>0.004</td>
<td>0.003</td>
<td>0.021</td>
<td>-0.001</td>
<td>0.022</td>
<td>-0.112</td>
</tr>
<tr>
<td>(Kr)</td>
<td>0.118</td>
<td>0.021</td>
<td>-0.203</td>
<td>-0.026</td>
<td>0.035</td>
<td>-0.042</td>
<td>-0.062</td>
</tr>
<tr>
<td>(gt)</td>
<td>0.256</td>
<td>0.023</td>
<td>-0.011</td>
<td>-0.028</td>
<td>-0.202</td>
<td>0.007</td>
<td>0.003</td>
</tr>
<tr>
<td>(kni)</td>
<td>0.012</td>
<td>0.020</td>
<td>-0.187</td>
<td>-0.082</td>
<td>0.000</td>
<td>-0.017</td>
<td>0.013</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Parameter</th>
<th>(hb)</th>
<th>(Kr)</th>
<th>(gt)</th>
<th>(kni)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(R^a)</td>
<td>15.000</td>
<td>10.354</td>
<td>15.000</td>
<td>15.000</td>
</tr>
<tr>
<td>(D^a)</td>
<td>0.166</td>
<td>0.200</td>
<td>0.103</td>
<td>0.200</td>
</tr>
<tr>
<td>(t^{1/2})</td>
<td>9.529</td>
<td>15.908</td>
<td>9.438</td>
<td>13.062</td>
</tr>
</tbody>
</table>

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 variation (20\%)

without cross regulation gap-genes
model also reproduces shifts 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
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

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: with delay: longer period
reinvented or conserved, which genes oscillate?

GO terms: signalling and transcription

Krol et al Development 2011

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 in 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

Conserved 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 asymmetric somatogenesis which is different for different vertebrate species HOW/WHY??

Model in more detail to find out which difference in regulatory network may explain difference in phenotype of RA knockouts
<table>
<thead>
<tr>
<th>organism</th>
<th>pErk dynamics</th>
<th>oscillating pathways</th>
<th>left-right phenotype</th>
<th>Slower osc</th>
<th>FGF8</th>
<th>delay (somite nr)</th>
<th>somite size diff</th>
<th>return to symmetry</th>
</tr>
</thead>
<tbody>
<tr>
<td>chick</td>
<td>smoothly retracting</td>
<td>FGF, Wnt, Notch</td>
<td>right side</td>
<td>symmetric, more anterior</td>
<td>no; left somites smaller</td>
<td>yes</td>
<td>unclear</td>
<td></td>
</tr>
<tr>
<td>zebrafish</td>
<td>retracts in jumps</td>
<td>Notch</td>
<td>right side</td>
<td>right side more anterior</td>
<td>right side 2-3 somites delayed</td>
<td>no</td>
<td>yes</td>
<td></td>
</tr>
<tr>
<td>mouse</td>
<td>oscillates</td>
<td>FGF, Wnt, Notch</td>
<td>right side</td>
<td>right side more anterior</td>
<td>right side 2-3 somites delayed</td>
<td>sometimes</td>
<td>yes</td>
<td></td>
</tr>
</tbody>
</table>

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