Genetic Oscillations and Feed-Backs in NF-κB, p53 and Wnt Systems

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1. Four eukaryotic systems with oscillatory gene expressions:

   • *Hes1-mRNA* protein network (Hirata et al (2002))
   • NF-κB transcription factor (Nelson, White et al)
   • Wnt-Notch segmentation network (Goldbeter, Pourquie)

   → Identify the ‘simplest’ negative feed-back loop.

   → Oscillating regimes: Ultradian time period (2-3 hours)
2. NF-κB, p53, Wnt systems: Regulated by negative feed-back loops: inflammation, apoptosis, segmentation.

3. NF-κB - IκB feed-back loop:
   Reduce 26-dimensional dynamics to three/nine variables
   Spiky oscillations:
   → Saturated degradation.
   → A20 change period. Chaotic response

4. External stresses and responses in p53-Mdm2:
   DNA damage, hypoxia, nutlin, etc

5. Somite segmentation in embryos (space):
   Oscillating proteins: Wnt signaling feed-back loop

6. Coupled feed-back loops in space:
   Cell-to-cell communications.
Collaborators:

- Sandeep Krishna, Guido Tiana (Milan), Simone Pigolotti
- Kim Sneppen
- Peter B. Jensen, Lykke Pedersen, Alex Hunziker, Benedicte Mengel,


‘Typical’ Oscillating data: Hes1 - segmentation

(Hirata et al, 2002)
Simplest negative feed-back loop: Hes1

\[
\frac{d[mRNA]}{dt} = \alpha \cdot [\rho_{free}] - \frac{[mRNA(t)]}{\tau_{rRNA}}
\]

\[
\frac{d[Hes1]}{dt} = \beta \cdot [mRNA(t)] - \frac{[Hes1(t)]}{\tau_{Hes1}}
\]

Jensen et al 2003
\[
\frac{d[mRNA]}{dt} = \alpha \cdot \frac{K_M}{K_M + [Hes1(t - \tau)]^n} - \frac{[mRNA(t)]}{\tau_{rna}}
\]

\[
\frac{d[Hes1]}{dt} = \beta \cdot [mRNA(t)] - \frac{[Hes1(t)]}{\tau_{hes1}}
\]

- Dashed curve [Hes1]
- Solid curve [mRNA]

- \( \tau_{rna} = 24.1 \text{ min} \)
- \( \tau_{hes1} = 22.3 \text{ min} \)
- \( \tau = 24 \text{ min} \)
- \( \alpha = 20 [R]_0 \text{ min}^{-1} \)
- \( \beta = 1/20 \text{ min}^{-1} \)
- \( K_M = (0.1[R]_0)^n \)
- \( n = 4 \)
Negative Feedback loops:

A.

B.

(1) signal

(2) time delay $\tau$

(3) steady state

(4) damped osc

(5) sustained osc

degr. of one component
Why oscillations?

• Importance for DNA-repair and apoptosis
• Essential in segmentation
• Spiky oscillations →
  important for sharp responses, fast regulations,
  high Hill coefficients
  → hormones also come in spikes

Mathematically: The most `simple’ dynamics!
The NF-κB System in Mammalian Cells

- NF-κB family: dimeric transcription factors
- Regulates immune response, inflammation, apoptosis
- Over 150 triggering signals, over 150 targets
- Each NF-κB has a partner inhibitor IκB
- Fluorescence imaging of NF-κB and IκB in human S-type neuroblastoma cells.
  

How does the network produce oscillations?
Why does the cell need the oscillations?
‘Direct’ observations of oscillations in nucleus

Oscillations in the nuclear localization of an NF-κB transcription factor in human cells
The NF-κB feed-back network

NF-κB oscillations

IκBβ/ε knocked out

The NF-κB System in Mammalian Cells

Reduction of the NF-κB system

7-variable model

 IkB mRNA → IkB → IkB mRNA

 NFκB → IkB mRNA → IkB

 IkB mRNA → IkB mRNA → IkB

 IkB mRNA → IkB mRNA → IkB

 IkB mRNA → IkB mRNA → IkB

 3-variable model

 IkB → IkB mRNA → NFκB

 IkB → IkB mRNA → NFκB

 IkB → IkB mRNA → NFκB

 IkB → IkB mRNA → NFκB

 IkB dependent degradation of IkB

 Remove very slow transport reactions
 Assume complexes are in equilibrium

 Assume certain concentrations ratios are constant

 complex formation/dissociation
 transport into/out of nucleus
 transcription & translation
Simple Model for Protein Oscillations

\[
\frac{dN_n}{dt} = A \frac{(1 - N_n)}{\epsilon + I} - B \frac{IN_n}{\delta + N_n},
\]

\[
\frac{dI_m}{dt} = N_n^2 - I_m,
\]

\[
\frac{dI}{dt} = I_m - C \frac{(1 - N_n)I}{\epsilon + I}.
\]

\[A = 0.007, \quad B = 954.5, \quad C = 0.035,\]

\[\delta = 0.029, \quad \epsilon = 2 \times 10^{-5}\]
Hopf bifurcation

A

unstable

$\varepsilon = 5 \times 10^{-4}$

stable

$\varepsilon = 1.5 \times 10^{-3}$

B

C

$\varepsilon = 5 \times 10^{-4}$

$\varepsilon = 1.5 \times 10^{-3}$
Robust Spiky Oscillations

Spiky Oscillations, $Z > 2$

$Z = \frac{\text{max} - \text{min}}{\text{avg}}$
Nested feed-back loops

A) Extracellular triggering signal (TNF)

- IKK (neutral)
- IKK (inactive)
- IκBα
- IκBε
- NFκB

B) Extracellular triggering signal (TNF)

- IKK
- IKK inactivation rate = $[K]^2$
- IKK inactive to neutral rate = $\frac{\text{inactive}K}{1+[A]^2}$
- IκBα degradation rate = $[K][N_I]\alpha$
- IκBε degradation rate = $[K][N_I]\varepsilon$

\[
[N:I_a] = \frac{I_a[N_e]}{1+I_a[I_e]} \quad (1)
\]
\[
[N:I_e] = \frac{I_e[N_a]}{1+I_a[I_e]} \quad (2)
\]

Cytoplasmic NFκB $[N_c]$

Nuclear NFκB $[N_n]$

Production rates

- fast
- slow

- Effective negative regulations
- (…) Concentrations

<table>
<thead>
<tr>
<th>NFκB regulated promoters</th>
<th>Interactions</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>activating</td>
</tr>
<tr>
<td></td>
<td>inhibiting</td>
</tr>
</tbody>
</table>

Legend: 
- Green: Activating 
- Red: Inhibiting 
- Yellow: Negative Regulation 
- Grey: Positive Regulation 
- Blue: Triggering Signal 
- Black: Others
A20 regulates period of NF-κB oscillations

Single cells!
IKK has been observed to oscillate itself

\[
\frac{dN_n}{dt} = A\frac{(1 - N_n)}{\epsilon + I} - B\frac{IN_n}{\delta + N_n},
\]

\[
\frac{dI_m}{dt} = N_n^2 - I_m,
\]

\[
\frac{dI}{dt} = I_m - C\frac{(1 - N_n)I}{\epsilon + I}.
\]

\[C' \rightarrow C(1 + \sin 2\pi \omega t)\]

(Kristian Rud, Jesper Fonslet)
Strange attractor of periodically ‘forced’ NF-κB system

\[
\begin{align*}
\frac{dN_n}{dt} &= A \frac{(1 - N_n)}{\epsilon + I} - B \frac{IN_n}{\delta + N_n}, \\
\frac{dI_m}{dt} &= N_n^2 - I_m, \\
\frac{dI}{dt} &= I_m - C \frac{(1 - N_n)I}{\epsilon + I}.
\end{align*}
\]

\[C \rightarrow C(1 + \sin2\pi\omega t)\]

\[A = 0.007, \ B = 954.5, \ C = 0.035, \ \\
\delta = 0.029 \text{ and } \epsilon = 2 \times 10^{-5}\] (2-3 hour period)
Often time series are very noisy! → Then what?

Response to irradiation in single cells

Figure 1  Prolonged oscillations in the nuclear levels of fluorescently tagged p53 and Mdm2 in individual MCF7, U280, cells following gamma irradiation. (A) Time-lapse fluorescence images of one cell over 29 h after 6 Gy of gamma irradiation. Nuclear p53-CFP and Mdm2-YFP are imaged in green and red, respectively. Time is indicated in hours. (B) Normalized nuclear fluorescence levels of p53-CFP (green) and Mdm2-YFP (red) following gamma irradiation. Top left: the cell shown in panel A. Other panels: five cells from one field of view, after exposure to 2.5 Gy gamma irradiation.

Apoptosis


Lahav, Alon, Levine,..
Mdm2 regulates both activity and stability.

\[
\frac{dp}{dt} = \sigma - \alpha p - k_f p m + k_b c + \gamma c
\]

\[
\frac{d{m_m}}{dt} = k_t p^2 - \beta {m_m}
\]

\[
\frac{dm}{dt} = k_{tl} m_m - k_f p m + k_b c + \delta c - \gamma m
\]

\[
\frac{dc}{dt} = k_f p m - k_b c - \delta c - \gamma c
\]

tions: nuclear-p53, \( p \); Mdm2, \( m \); Mdm2 mRNA, \( m_m \); and the p53-Mdm2 complex, \( c \). The tempo-
Four different stresses

- DNA damage: Irradiation triggers oscillations: increase auto-ubiq. of Mdm2, decrease ubiq. of p53 by Mdm2, weak p53-Mdm2 binding
- Hypoxia: Deprive oxygen, apoptosis: decrease transactivation, prevents degrad. of p53
- Nutlin (chemical), cell-cycle arrest (not apoptosis): reduces p53-Mdm2 binding
Stress variations in parameters

A

Free p53 [nM]

0 5 10 15

Time [h]

B

Oncogene

DNA damage

Hypoxia

C

Spikyness ➔

D

p53 concentrations (peak)
Responses:

\( \delta \): DNA damage, hypoxia, oncogenes
Mdm2 SNP309 allele

Weaker response for GG than for TT
Stochastic simulations (for p53!!)

![Graph showing free p53 molecules over time](image-url)
The presomitic mesoderm (PSM) segments anterior-posterior as somites bud off from the anterior end.

Dividing stem cells in the tailbud supply cells to posterior PSM and elongates the embryo.

PSM cells have locally synchronized oscillating expression patterns with periods matching somite formation (90 min in chick) – **Clock**

A morphogen gradient (**Wavefront**) determines onset of segmentation program.

**Clock** determines susceptibility to **wavefront**, which ensures groupwise incorporation into somites.

(Cooke and Zeeman 1976)

Master thesis work by Peter B. Jensen
Several signaling pathways are involved

Oscillating transcripts are mainly targets of Notch and Wnt pathways

Notch and Wnt targets have same period but are 180 out of phase

Crosstalk & possible hierarchical relationship

Focus is on *Hes1/7, Lfng* and *Axin2* that have all been associated with feedback loops

One crosstalk candidate (out of many possible) is GSK3β, which can bind and phosphorylate both β-catenin and Notch

Dequèant et al. 2006
The Wnt systems

Equations for the Wnt system

Destruction complex
\[
\frac{dC}{dt} = c_{tC}B[GA] - c_{bC}C - \alpha C, \quad (1)
\]

GA-complex
\[
\frac{d[GA]}{dt} = c_{f[GA]}GA - c_{b[GA]}[GA] - c_{fC}B[GA] + c_{bC}C + \alpha C, \quad (2)
\]

\(\beta\)-catenin
\[
\frac{dB}{dt} = S - c_{tC}B[GA] + c_{bC}C, \quad (3)
\]

GSK
\[
\frac{dG}{dt} = -c_{f[GA]}GA + c_{b[GA]}[GA], \quad (4)
\]

Axin2
\[
\frac{dA}{dt} = -c_{f[GA]}GA + c_{b[GA]}[GA] + c_{tIA}A_m - c_{f[AL]}AL \\
+ c_{b[AL]}[AL], \quad (5)
\]

\[
\frac{dA_m}{dt} = c_{tsA}B^2 - \frac{A_m}{\tau_{Am}}, \quad (6)
\]

\[
\frac{d[AL]}{dt} = c_{f[AL]}AL - c_{b[AL]}[AL] - \nu[AL], \quad (7)
\]

Axin2-Wnt
\[
\frac{dL}{dt} = -c_{f[AL]}AL + c_{b[AL]}[AL] + \nu[AL], \quad (8)
\]
<table>
<thead>
<tr>
<th>Parameter</th>
<th>Process</th>
<th>Default value</th>
</tr>
</thead>
<tbody>
<tr>
<td>$c_{fC}$</td>
<td>Binding of B to [GA] to form destruction complex C</td>
<td>$0.1 \text{ nM}^{-1} \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$c_{bC}$</td>
<td>Dissociation of C into B and [GA]</td>
<td>$7 \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$\alpha$</td>
<td>Dissociation of C due to destruction of $\beta$-catenin</td>
<td>$200 \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$c_{f[GA]}$</td>
<td>Binding of G to A to form [GA]</td>
<td>$0.2 \text{ nM}^{-1} \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$c_{b[GA]}$</td>
<td>Dissociation of [GA] into G and A</td>
<td>$1.2 \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$S$</td>
<td>Constant source of $\beta$-catenin</td>
<td>$0.4 \text{ nM min}^{-1}$</td>
</tr>
<tr>
<td>$c_{f[AL]}$</td>
<td>Binding of A to L</td>
<td>$10 \text{ nM}^{-1} \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$c_{b[AL]}$</td>
<td>Dissociation of [AL] into A and L</td>
<td>$0.08 \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$c_{tsA}$</td>
<td>Transcription of $axin2$ gene</td>
<td>$0.7 \text{ nM}^{-1} \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$c_{tLA}$</td>
<td>Translation of Axin2 mRNA</td>
<td>$0.7 \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$\tau_{Am}$</td>
<td>Average lifetime of Axin2 mRNA</td>
<td>40 $\text{ min}$</td>
</tr>
<tr>
<td>$\nu$</td>
<td>Degradation of Axin2 in [AL] complex</td>
<td>$0.1 \text{ min}^{-1}$</td>
</tr>
<tr>
<td>$GSK3\beta_{tot}$</td>
<td>Total G level</td>
<td>$50 \text{ nM}$</td>
</tr>
<tr>
<td>$L_{tot}$</td>
<td>Total L level</td>
<td>$70 \text{ nM}$</td>
</tr>
</tbody>
</table>
The Wnt reference state
\[ D_C = \frac{C_{bC}}{C_{fC}} \]

\[ D_{[GA]} = \frac{C_{b[GA]}}{C_{f[GA]}} \]

FIGURE 3 The \( D_C \) and \( D_{[GA]} \) parameter plane. (Left panel) The amplitude of the Axin2 oscillations. (Right panel) Oscillation period of Axin2. The shaded borderline indicates the boundary of sustained oscillations, derived from a stability analysis of the system. The reference state (see Fig. 2) is indicated by the dot labeled “ref state”. Timeseries corresponding to the four numbered dots are shown in Fig. 4.
Spatial gradient of Wnt:

FIGURE 6 Oscillations of the Wnt variables as $\nu$ is linearly decreased from 0.1 to 0.03 min$^{-1}$ in 1200 min. Note that the oscillations cease when the Wnt signal falls below a certain threshold level.
Three node motifs: Oscillations and Switches

a) Repressilator: oscillations

\[
\frac{dx}{dt} = c - \gamma x + \alpha \frac{1}{1 + \left(\frac{\tilde{x}}{K}\right)^h}
\]
A model for cell-to-cell communications:

\[
\frac{dx_{m,n}}{dt} = c - \gamma x_{m,n} + \alpha F_{\text{int}}
\]
Dynamical equation for site \((m,n)\):

We consider two types of interaction terms—either an additive repression (an “OR gate”),

\[
F_{\text{int}} = \frac{1}{1 + \left(\frac{x_{m+1,n}}{K}\right)h} + \frac{1}{1 + \left(\frac{x_{m,n-1}}{K}\right)h} + \frac{1}{1 + \left(\frac{x_{m-1,n+1}}{K}\right)h}, \tag{2}
\]

or a multiplicative repression (an “AND gate”),

\[
F_{\text{int}} = \frac{1}{1 + \left(\frac{x_{m+1,n}}{K}\right)h} \cdot \frac{1}{1 + \left(\frac{x_{m,n-1}}{K}\right)h} \cdot \frac{1}{1 + \left(\frac{x_{m-1,n+1}}{K}\right)h}. \tag{3}
\]
Coupled Repressilators: A Repressor-Lattice

‘Natural’ phases: No frustration!

Repressing genes/proteins: in a cell or between cells!
Unit cells:

3x3

4x4
Dynamical solutions

3x3

4x4
5x5

Solutions from rotational symmetry: Group theory!
Dynamical frustrations: Chaotic solutions
Non-balanced repressor-lattice

One activator

frustration!
Frustrated solutions:

3 x 3 lattice

Phases are fixed!
Cell-to-cell communication:

- Cell touches: gap junctions → Directed (or bidirected?) interactions! Oscillations!
- Cells do not touch: Send out signalling molecules → bi-directed interactions:
  ‘Local’ switches → High or low state!

Relevant for ordered tissues: fat tissue, onion skin, human skin, etc.
Bi-directional interactions

No oscillations: hi or low states. Mutations!
Cell-to-cell communication in space:
Fatty tissues: organized on a “lattice”
Negative feed-back loops

• Guess the feed-back loop from an experimental time series: Algorithm
• Derive symbolic dynamics
• From fixed points to Hopf bifurcations

with Simone Pigolotti and Sandeep Krishna

Negative Feed-Back Loops:

- Always odd number of repressor links
- A ‘closed’ loop: No cross-links
- Node: concentration, expression level, etc
Determined by the following equation:

\[
\frac{dx_i}{dt} = g_i^{(A,R)}(x_i, x_{i-1})
\]

Assume \( g_i^{(A;R)} \) are monotonic

Example:

\[
\frac{dx_i}{dt} = c - \gamma x_i + \alpha \frac{1}{1 + (x_{i-1}/K)^h}
\]

- **c**: basal production
- **\( \gamma \)**: degradation → linear
- **\( \alpha \)**: production rate: activation↔repression
Often time series are very noisy! → Then what?
Experimental time series: (transient is OK !)


Symbolic Dynamics:
Stationary point:

\[ g_i^{(A,R)}(x_i^*, x_{i-1}^*) = 0 \implies x_i^* = f_i^{(A,R)}(x_{i-1}^*) \]

Notice:

\[ f_i^{(A,R)} \rightarrow \text{same monotonocity as } g_i^{(A,R)} \]
\[ (\text{when } g_i^{(A,R)} \text{ is decreasing in } x_i) \]

Fixed point equation:

\[ x_i^* = f_i(x_{i-1}^*) = f_i(f_{i-1}(x_{i-2}^*)) = \ldots = \]
\[ = f_i \circ f_{i-1} \circ f_{i-2} \circ \ldots \circ f_{i+1}(x_i^*) \equiv F_i(x_i^*) \]

\[ \text{i.e. measures how species ‘i’ interacts with itself through the loop} \]
Using chain rule: 

\[ F'_i(x) = \prod_j f'_j(x_j)|_{x_i=x} \]

Therefore: If even number of repressors → positive → multiple fixed points (bistability)

If odd number of repressors → negative → only one fixed point!

Eigenvalue equation:

\[ \prod_{i=1}^{N} \left( \frac{\lambda}{h_i} + 1 \right) = F'(x^*) \]

where \( h_i = -\partial_x g_i(x_i, x_i=1)|_{x^*} \) (degradation rates at fixed point)
\[ \prod_{i=1}^{N} \left( \frac{\lambda}{h_i} + 1 \right) = F'(x^*) \]

Right hand side: Negative
Left hand side: Positive coefficients

: Hopf bifurcation
Example:

\[ \alpha = 3.0, \ c = 0.1, \ K = 1, \ \gamma = 1, \ h = 2 \]
\[ \text{Not OK: } h = 4 \]

Three repressors:

\[
\frac{dx_i}{dt} = c - \gamma x_i + \alpha \frac{1}{1 + (x_{i-1}/K_i)^h}
\]

\[ i = 1 \ldots 3. \]

We denote by \( x^* \) the solution to the equation \( \gamma x = c + \alpha/(1 + (x/K)^h) \). Then the characteristic polynomial is simply:

\[
(\lambda + \gamma)^3 = -\left( \frac{\alpha}{1 + (x^*/K_i)^h} \right)^3
\]

Stability condition

\[
\Rightarrow \left( \frac{\lambda}{\gamma} + 1 \right)^3 = F'(x^*) \Rightarrow |F'(x^*)| \cos(\pi/3) < \gamma
\]

\{ \text{OK: } h = 2 \}
\{ \text{Not OK: } h = 4 \}
Circadian rhythms of kai genes in cyanobacteria

Binding of four proteins to pS2 promoter

Adding estradiol:
ER: estradiol receptor, initiates transcription
Pol: RNA polymerase

Now with possible cross-links!
‘Normal’ symbolic dynamics

Kick!
FIG. 2: Network of two coupled two-species oscillators, or a three trophic level ecosystem. (a) Structure of the network. (b) The transition network for this 3-node system. Black arrows indicate all the allowed transitions. Blue arrows are the transitions actually observed in the HP system and red arrows are the transitions observed in the BHS model (see text). In both cases, dashed arrows indicate “kicks”, i.e., transitions
Two possible dynamical systems for system

\[
\begin{align*}
\dot{x}_1 &= r x_1 (1 - k x_1) - \alpha_1 \frac{x_1 x_2}{1 + b_1 x_1} \\
\dot{x}_2 &= -d_1 x_2 + \alpha_1 \frac{x_1 x_2}{1 + b_1 x_1} - \alpha_2 \frac{x_2 x_3}{1 + b_2 x_2} \\
\dot{x}_3 &= -d_2 x_3 + \alpha_2 \frac{x_2 x_3}{1 + b_2 x_2}
\end{align*}
\]

Hastings-Powell

\[
\begin{align*}
\dot{x}_1 &= x_1 - \alpha_1 \frac{x_1 x_2}{1 + k x_1} \\
\dot{x}_2 &= -d x_2 + \alpha_1 \frac{x_1 x_2}{1 + k x_1} - \alpha_2 x_2 x_3 \\
\dot{x}_3 &= c (x_3^* - x_3) + \alpha_2 x_2 x_3
\end{align*}
\]

Blasius et al
Attractors of the dynamics of HP model
Bifurcation diagrams for HP and Blasius
Reduction of the NF-κB System

Sandeep Krishna

7 variable model

Equilibrium of complexes

Small terms deleted

3 variable model

where

\[ A = \frac{k_{Nf}I_mN_{tot}^2}{k_{lf}k_{il}N_{tot}} \]
\[ B = \frac{k_{lf}I_mN_{tot}^2}{k_{il}} \]
\[ C = \frac{\gamma_m}{k_{lf}k_{il}N_{tot}} \]
\[ \epsilon = \frac{k_{lf}I_mN_{tot}^2}{k_{il}} \]
\[ \delta = \frac{k_{lf}N_{tot}}{k_{il}N_{tot}} \]
Two oscillators: Wnt and Notch: out of phase

Aulehla and Herrmann

Goldbeter, Pourquie

Wnt gradient and Clock are coupled
The full systems with Wnt and Noctch and cross-talk
Equations of the Wnt-Notch network

\[
\begin{align*}
\frac{dC}{dt} & = c_{fC} B[G] - c_{bC} C - \alpha_1 C \\
\frac{dG}{dt} & = -c_{f[G]} AG + c_{b[G]} [GA] - c_{f[GN]} GN + c_{b[GN]} [GN] + \alpha_2 [GN] \\
\frac{dB}{dt} & = S - c_{fC} B[G] + c_{bC} C - \frac{B}{\tau_B} \\
\frac{dA}{dt} & = -c_{f[G]} AG + c_{b[G]} [GA] + c_{lA} A_m - c_A \frac{A}{k_A + A} \\
\frac{dA_m}{dt} & = c_{tsA} B^h - \frac{A_m}{\tau_{Am}} \\
\frac{d[G]}{dt} & = c_{f[G]} AG - c_{b[G]} [GA] - c_{fC} B[G] + c_{bC} C + \alpha_1 C \\
\frac{dN}{dt} & = c_s \frac{k_s}{k_s + L} - \frac{N}{\tau_N} - c_{f[GN]} GN + c_{b[GN]} [GN] \\
\frac{dH_m}{dt} & = c_{tsH} \left( \frac{N^{h_{NH}}}{k_{NH} + N^{h_{NH}}} \cdot \frac{k_{HH}}{k_{HH} + H^{h_{HH}}} \right) - \frac{H}{\tau_{Hm}} \\
\frac{dH}{dt} & = c_{tIH} H_m - \frac{H}{\tau_H} \\
\frac{dL_m}{dt} & = c_{tsL} \left( \frac{N^{h_{NL}}}{k_{NL} + N^{h_{NL}}} \cdot \frac{k_{HL}}{k_{HL} + H^{h_{HL}}} \right) - \frac{L_m}{\tau_{Lm}} \\
\frac{dL}{dt} & = c_{tLL} L_m - \frac{L}{\tau_L} \\
\frac{d[NG]}{dt} & = c_{f[NG]} NG - c_{b[NG]} [NG] - \alpha_2 [NG]
\end{align*}
\]

A total of approx. 27 constants – Some are poorly defined
The full Wnt-Notch system

Transcription factors
Figure 7.2: Timeseries of the 12 equation system with Lfng negative feedback
Spatial gradient of Wnt:

Gaussian profiles

Oscillations stop when Wnt under a threshold
Simple Model for Protein Oscillations

Simple model qualitatively reproduces several features of NF-κB oscillations:

- fact of oscillations
- time period
- shape of oscillations
- phase relationships
- no osc. in the absence of feedback
- increased transcription → lower freq.

\[
A = 0.007, \quad B = 954.5, \quad C = 0.035, \\
\delta = 0.029, \quad \epsilon = 2 \times 10^{-5}
\]
Coupling NF-κB to a Downstream Gene

Equilibrium binding, \(1/k_{off}\) \(\ll\) time period

Non-equilibrium binding, \(1/k_{off}\) \(>\) time period

\[ G + 2N \xrightleftharpoons[k_{off}]{k_{on}} G^* \]
\[ G^* = \frac{N_n^2}{k_{off}/k_{on} + N_n^2} \]
<table>
<thead>
<tr>
<th>Variable</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>$N_n$</td>
<td>nuclear NF-κB</td>
</tr>
<tr>
<td>$I_{α/ε}$</td>
<td>free IκB</td>
</tr>
<tr>
<td>$I_{mα/ε}$</td>
<td>IκB mRNA</td>
</tr>
<tr>
<td>$A_m$</td>
<td>A20 mRNA</td>
</tr>
<tr>
<td>$K$</td>
<td>active IKK</td>
</tr>
<tr>
<td>$K_{i}$</td>
<td>inactive IKK</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Description</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>$T$</td>
<td>TNF stimulation</td>
<td>1</td>
</tr>
<tr>
<td>$B$</td>
<td>proportionality factor of the export of nuclear NF-κB</td>
<td>102.6</td>
</tr>
<tr>
<td>$A$</td>
<td>proportionality factor of the import of NF-κB</td>
<td>0.004</td>
</tr>
<tr>
<td>$η$</td>
<td></td>
<td>0.092</td>
</tr>
<tr>
<td>$K_{I}$</td>
<td>IκB - NF-κB complex dissociation factor</td>
<td>$1.26 \times 10^{-5} \ \mu M$</td>
</tr>
<tr>
<td>$δ$</td>
<td>concentration at which half of the $IκBα/ε$ is bound in complex with NF-κB</td>
<td>$0.0414 \ \mu M$</td>
</tr>
<tr>
<td>$p$</td>
<td>NF-κB independent transcription rate of $IκBα$ mRNA</td>
<td>$3.36 \times 10^{-5} \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$t_a$</td>
<td>NF-κB dependent transcription rate of $IκBα$ mRNA</td>
<td>$0.0042 \ \mu M \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$t_e$</td>
<td>NF-κB dependent transcription rate of $IκBε$ mRNA</td>
<td>$0.084 \ \mu M \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$t_A$</td>
<td>NF-κB dependent A20 transcription rate</td>
<td>$0.0168 \ \mu M^{-1} \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$t_{la}$</td>
<td>translation rate of $IκBα$</td>
<td>$0.0672 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$t_{le}$</td>
<td>translation rate of $IκBε$</td>
<td>$1.2 \times 10^{-5} \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$t_{la}$</td>
<td>translation rate of A20</td>
<td>$0.3024 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$γ_{lα}$</td>
<td>half-life of $IκBα$ mRNA</td>
<td>$0.0168 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$γ_{lε}$</td>
<td>half-life of $IκBε$ mRNA</td>
<td>$0.00168 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$γ_{lα/ε}$</td>
<td>half-life of the IκB’s</td>
<td>$0.005 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$γ_{A20m}$</td>
<td>half-life of the A20 mRNA</td>
<td>$0.0168 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$γ_{A20}$</td>
<td>half-life of the A20</td>
<td>$0.001 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$α_{α}$</td>
<td>IKK dependent degradation of $IκBα$</td>
<td>$0.00025 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$α_{ε}$</td>
<td>IKK dependent degradation of $IκBε$</td>
<td>$7.6 \times 10^{-6} \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$μ$</td>
<td>rate of IKK self-inactivation</td>
<td>$0.063 \ \text{min}^{-1}$</td>
</tr>
<tr>
<td>$σ$</td>
<td>strength of A20 negative feedback</td>
<td>0.25</td>
</tr>
<tr>
<td>$β$</td>
<td>proportionality factor of A20 on IKK</td>
<td>1.25</td>
</tr>
</tbody>
</table>
When oscillations in feed-back loops?

- Process that takes a finite (minimum) time
  \[ \text{evt. time delay } \tau: \quad \frac{dx}{dt} \sim P(t-\tau) \]
- Many intermediate steps
  binding, complex, steps on DNA, etc
  several components: repressilator
- Sharp response: high Hill coefficient
  \[ \frac{dm}{dt} \sim \frac{p^n}{k^n + p^n} \]
- Saturated degradation: depends on level
  \[ \frac{dI}{dt} \sim I_m - \frac{cI}{\varepsilon + I} \]
- Autocatalysis:
  \[ \frac{dx}{dt} \sim \frac{x^n}{k^n + x^n} \]
\( k_g \) : dissociation constant between p53 and DNA (O-operator site)

\[
[p] = [p_f] + [pm] \quad [O] = [O_f] + [pO] \quad ([O] = 1)
\]

\[
k_g = \frac{[p_f][O_f]}{[pO]} \Rightarrow [pO] = \frac{[p_f][O]}{k_g + [p_f]} = \frac{[p] - [pm]}{k_g + [p] - [pm]}
\]

(can add a Hill coefficient)

\([pO]\): “Equilibrium” probability that p53 is bound to DNA

\( k \) : dissociation constant between p53 and mdm2

\[
k = \frac{[p_f][m_f]}{[pm]} = \frac{([p] - [pm])([m] - [pm])}{[pm]} \Rightarrow
\]

\[
[pm]^2 - ([p] + [m] + k)[pm] + [p][m] = 0 \Rightarrow
\]

\[
[pm] = \frac{1}{2}([p] + [m] + k) - \sqrt{([p] + [m] + k)^2 - 4[p][m]}
\]
Why oscillations?

- Importance for apoptosis
- Essential in segmentation
- Spiky oscillations →
  - important for sharp responses,
  - fast regulations,
  - high Hill coefficients
  → hormones also come in spikes

Mathematically: The most `simple' dynamics!