L5.3: Cells as Biosensors II

Prof. Rickus
In this lecture

• Sensitivity

• Sensitivity Amplification

• Ultrasensitivity
What is a biosensor?
Sensitivity

Sensitivity: The differential change in $R$ as a ratio to the differential change in $S$

Plot the magnitude of the response magnitude as a function of the signal magnitude
Sensitivity

Increase the sensitivity of the sensor response

\[ R = mS + b \]

- More sensitive
- Less sensitive

slope = \( \frac{\text{unit of response}}{\text{unit of signal}} \)

background

sensitivity
Dynamic Range

Concentration Range for which Response is Predictably Dependent Upon the Signal Level

max – usually some saturation of the response (e.g. saturated binding)
min – signal lost in the noise (limit of detection)
Classic Example of Magnitude Amplification - Phototransduction

1 photon event results in the hydrolysis of $10^5$ cGMPs

1) 1 metarhodopsin catalyzes many GDP/GTP exchanges
2) Each phosphodiesterase can hydrolyze many molecules of cGMP

From Neuron to Brain. 4th ed. Nicholls, Martin, Wallace, Fuchs, chapter 19
Whole Cell Biosensor

commonly genetically encoded

Whole Cell Biosensor from Simple Activation

Response of the Sensor Circuit at Zero Signal
Some circuits can be “leaky”. E.g. still get basal transcription with 0 activator.

Simple activation model

\[
\frac{dY}{dt} = \beta_o + \beta_1 \frac{X}{X + K} - \alpha_Y Y
\]

Maximum
\(\beta_1\)

Transcription rate of \(Y\)

Background
\(\beta_o\)

Graph showing response of the sensor circuit as a function of signal.
Classic Hyperbolic Response (e.g. Calibration) Curve

Normalize the response and stimulus parameters

\[ R = \frac{R_{\text{max}}S}{S + S_{0.5}} \]

\[ r = \frac{R}{R_{\text{max}}} \]

\[ s = \frac{S}{S_{0.5}} \]

\[ r = \frac{s}{s + 1} \]

\[ dr = \frac{1}{(s + 1)^2} \]

\[ S_{0.5} = K_{XY} = \text{level of } X \text{ signal that results in a half maximum response (transcription rate of } Y) \]
Sensitivity Amplification

An increase in the % change in response, \( R \), relative to the % change in stimulus, \( S \).

We define the sensitivity amplification factor, \( A_s \).

\[
A_s = \frac{\Delta R}{\Delta S} = \frac{(R_f - R_i)}{R_i} \frac{R_i}{(S_f - S_i)} \frac{(S_f - S_i)R_i}{(S_f - S_i)S_i} = \frac{(R_f - R_i)S_i}{(S_f - S_i)R_i}
\]
**Ultrasensitivity**

$A_s > 1$ for a range of $S$

**Hyperbolic sensitivity**

Classic hyperbolic calibration

**Sub-sensitivity**

The $A_s$ is never greater than 1.

Note the trade-off between dynamic range and sensitivity.

Koshland, Goldbeter, Stock Science 1982
Biological Design Approaches to Achieve Ultra-sensitivity

1. Cooperativity
2. Cascades
3. Zero-Order Ultra-sensitivity
Ultra-sensitivity – Cooperativity

Hill function model of gene activation

<table>
<thead>
<tr>
<th>[X]</th>
<th>prob.</th>
<th>rate</th>
<th>gene</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\frac{x^n}{x^n + K^n}$</td>
<td>$\frac{x^n}{x^n + K^n}$</td>
<td>$\beta \frac{x^n}{x^n + K^n}$</td>
<td></td>
</tr>
<tr>
<td>$x \gg K$</td>
<td>$\rightarrow 1$</td>
<td>$\rightarrow \beta$</td>
<td>ON high</td>
</tr>
<tr>
<td>$x = K$</td>
<td>$0.5$</td>
<td>$\beta / 2$</td>
<td>ON mod</td>
</tr>
<tr>
<td>$x \ll K$</td>
<td>$\rightarrow 0$</td>
<td>$\rightarrow 0$</td>
<td>OFF</td>
</tr>
</tbody>
</table>

K:  - units conc.
    - defines functional concentration range of X
    - may correlate with (but is not formally) the binding affinity of X to the DNA
    - other factors contribute to K

n:  - Hill coefficient
    - increases nonlinearity of function
    - increases steepness of sigmoidal
    - greater n, more on/off switch-like

From Lecture on Simple Models of Gene Expression
Ultra-sensitivity: Multi-stage Cascades

A

aTc

Circuit 1

\[
\text{TetR} \rightarrow \text{tetR} \rightarrow \text{eyfp} \\
P_{\text{lacq}} \rightarrow P_{\text{Ltet-O1}}
\]

B

Circuit 1

Mean fluorescence (MEFL)

C

Circuit 3

aTc

\[
\text{TetR} \rightarrow \text{LacI} \rightarrow \text{Cl} \rightarrow \text{eyfp} \\
P_{\text{lacq}} \rightarrow P_{\text{Ltet-O1}} \rightarrow P_{\text{lac}} \rightarrow \lambda_{P(R-O12)}
\]

Sara Hooshangi et al. PNAS 2005;102:3581-3586
Ultra-sensitivity: Zero Order Sensitivity

S is an activator of Enzyme, E1

E1 and E2 are enzymes that follow classic Michaelis Menton Kinetics

R = \( \frac{W^*}{W_{\text{total}}} \)

W is some Response Signal Protein that can exist in 2 states
Structure is used widely by biology

Kinases make up 1.5 – 2.5% of all proteins in most genomes examined.

Table 1
Abundance of kinases in various organisms: taken from Manning et al. (2000)

<table>
<thead>
<tr>
<th>Organism</th>
<th>Number of putative kinases</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli</em></td>
<td>29</td>
</tr>
<tr>
<td><em>Saccharomyces cerevisiae</em></td>
<td>130</td>
</tr>
<tr>
<td><em>Drosophila melanogaster</em></td>
<td>239</td>
</tr>
<tr>
<td><em>Caenorhabditis elegans</em></td>
<td>454</td>
</tr>
<tr>
<td>Human</td>
<td>518</td>
</tr>
</tbody>
</table>

0° Ultrasensitivity Depends on Certain Conditions:

**Zero Order Regime:** The total amount of $W_T = W + W^*$ must be very high relative to the $K_M$'s of the reactions catalyzed by E1 and E2.

- $K_{M,1} / W_T = K_{M,1} / W_T = 0.1$
- Equivalent Hill coefficient = 2.9
- $K_{M,1} / W_T = K_{M,1} / W_T = 0.01$
- Equivalent Hill coefficient = 13
Cascades to Enhance $0^\circ$ Ultra-sensitivity:

Adding the 2nd cycle increases the ultrasensitivity response Equivalent to hill coeff. of

\[ R_1 = \frac{W^*}{W_T} \quad 1 \text{ cycle} \quad 3.6 \]
\[ R_2 = \frac{Z^*}{Z_T} \quad 2 \text{ cycle} \quad 7.5 \]

ALBERT GOLDBETER AND DANIEL E. KOSHLAND, JR. PNAS 1981
Engineering Design: Analogy to the Transistor

View these systems as sensors, amplifiers, or switches

Fig. 4. Comparison of transistor to cascade response.

Herbert M. Sauroa, Boris N. Kholodenkoc
Coming up …

• Move on to Big Picture Thinking
  • Future Technologies – Synthetic Life
  • Ethics, Science & Society