

BE 150: Design Principles of Genetic Circuits

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21 Turing patterns

In recent lectures, we have been talking about signaling *between* cells, whether via cytokines regulating homeostasis, via Delta-Notch signaling, or via BMP signaling. The latter two signaling pathways are important in development of an organism, as we saw in lecture. They lead to spatial patterning of cells of different types.

As we saw in the discussion of BMP, the expression of other genes in the cells of interest depend on the concentration of the ligands and receptors involved in signaling. It seems that in many contexts, biochemical cues that influence cell fate do so in a *concentration-dependent manner*; higher concentrations result in stronger signals than lower concentrations. Such chemical species, which determine cell fate in a developmental context in a concentration-dependent way, are called **morphogens**.

For this lecture, we will discuss **reaction-diffusion** mechanisms for generating spatial distributions of morphogens. This is important both practically, and historically.

21.1 Turing's thoughts on reaction-diffusion mechanisms for morphogenesis

In my favorite paper of all time, Alan Turing laid out a prescription for morphogenesis. He described what should be considered when studying the “changes of state” of a developing organism. Turing said,

In determining the changes of state one should take into account:

- (i) the changes of position and velocity as given by Newton's laws of motion;
- (ii) the stresses as given by the elasticities and motions, also taking into account the osmotic pressures as given from the chemical data;
- (iii) the chemical reactions;
- (iv) the diffusion of the chemical substances (the region in which this diffusion is possible is given from the mechanical data).

This makes perfect sense; if we are shaping an organism, we have to think about what changes the shape and also how the little molecules that govern the shape move about and react. That said, a few lines later in the paper, Turing wrote, “The interdependence of the chemical and mechanical data adds enormously to the difficulty, and attention will therefore be confined, so far as is possible, to cases where these can be separated.”

Turing went on to consider only the chemical reactions coupled with diffusion. His claim, which we will explore mathematically in this lecture, was that when diffusion was considered, spatial patterns in chemical species could form. Stop and think about that for a moment. Diffusion, a *dissipative* process, can help give spatial patterns in chemical systems that otherwise have no spatial information. In all of your life experience, I bet diffusion has served to flatten out patterns, like when you put food coloring in a glass of water. Let's explore Turing's counterintuitive proposal.

We will specifically explore a type of patterning that now bears the name “Turing patterns.” A Turing pattern is formed *spontaneously* due to diffusion from a system of two chemical species that have a stable fixed point in the absence of diffusion.

21.2 Reaction-diffusion equations for two components

Consider two chemical components, U and V, that can react with each other. The dynamical equations we have considered thus far are of the form

$$\frac{du}{dt} = f(u, v) \quad (21.1)$$

$$\frac{dv}{dt} = g(u, v). \quad (21.2)$$

If the species can undergo diffusion, we need to include diffusive terms. We use Fick's second law, which says that the rate of change of concentration of a diffusing species is

$$\frac{\partial c}{\partial t} = D\nabla^2 c, \quad (21.3)$$

to update our dynamical equations.

$$\frac{\partial u}{\partial t} = D_u \nabla^2 u + f(u, v) \quad (21.4)$$

$$\frac{\partial v}{\partial t} = D_v \nabla^2 v + g(u, v). \quad (21.5)$$

Our goal is to determine what type of regulation we need in the chemical circuit (i.e., to decide what type of arrowheads, activating or repressing, to use in Fig. 19) and what values of D_u and D_v are necessary to give spontaneous patterning of U and V.

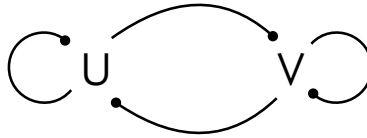


Figure 19: Unspecified interactions of a circuit that can for Turing patterns with diffusion. Our goal is to figure out the arrowheads.

21.2.1 Linear stability analysis of a PDE

Earlier in the class, we learned how to do linear stability analysis of a fixed point of a system of ordinary differential equations. For this system, we also have spatial derivatives, so we need to perform linear stability analysis on a system of *partial* differential equations. Specifically, we will consider a chemical reaction system that has a stable fixed point in the absence of diffusion. The idea is that the chemical reaction system itself will not be oscillatory and run away to some other state. We will test to see if the inclusion of spatial information, by explicitly considering diffusion, can make the entire system unstable, thereby giving rise to patterns.

Let us assume that (u_0, v_0) is a stable fixed point of the dynamical system in the absence of diffusion. That is, $f(u_0, v_0) = g(u_0, v_0) = 0$. Therefore, we can construct a *homogeneous steady state* where $a(\mathbf{x}) = u_0$ and $b(\mathbf{x}) = v_0$, where \mathbf{x} denotes spatial variables. This is to say that the concentrations of

species u and v are everywhere equal to their value at the stable fixed point. This is the unpatterned state, which is stable in the absence of diffusion.

The question now is *is the homogeneous steady state stable in the presence of diffusion?* To answer this question, we will perform a linear stability analysis. We linearize $f(u, v)$ and $g(u, v)$ about (u_0, v_0) .

$$f(u, v) \approx f(u_0, v_0) + f_u \delta u + f_v \delta v, \quad (21.6)$$

$$g(u, v) \approx g(u_0, v_0) + g_u \delta u + g_v \delta v, \quad (21.7)$$

where

$$f_u = \left. \frac{\partial f}{\partial u} \right|_{(u_0, v_0)}, \quad f_v = \left. \frac{\partial f}{\partial v} \right|_{(u_0, v_0)}, \quad (21.8)$$

$$g_u = \left. \frac{\partial g}{\partial u} \right|_{(u_0, v_0)}, \quad g_v = \left. \frac{\partial g}{\partial v} \right|_{(u_0, v_0)}, \quad (21.9)$$

and $\delta u = u - u_0$ and $\delta v = v - v_0$ represent the perturbation from the homogeneous steady state. Note that δu and δv are functions of position \mathbf{x} and time t . Thus, our linearized system is

$$\frac{\partial \delta u}{\partial t} = D_u \nabla^2 \delta u + f_u \delta u + f_v \delta v, \quad (21.10)$$

$$\frac{\partial \delta v}{\partial t} = D_v \nabla^2 \delta v + g_u \delta u + g_v \delta v. \quad (21.11)$$

Note that the terms f_u and g_v describe the nature of the autoregulation of the genetic circuit

Taking the Fourier transform of the equations gives

$$\frac{\partial \widehat{\delta u}}{\partial t} = -k^2 D_u \widehat{\delta u} + f_u \widehat{\delta u} + f_v \widehat{\delta v} \quad (21.12)$$

$$\frac{\partial \widehat{\delta v}}{\partial t} = -k^2 D_v \widehat{\delta v} + g_u \widehat{\delta u} + g_v \widehat{\delta v}, \quad (21.13)$$

where \mathbf{k} is the vector of wave numbers (e.g., $\mathbf{k} = (k_x, k_y)$ for a two-dimensional system) and $k^2 \equiv \mathbf{k} \cdot \mathbf{k}$. This is now a system of linear *ordinary* differential equations in $\widehat{\delta u}$ and $\widehat{\delta v}$, which we can approach in our usual way of doing linear stability analysis.

To avoid the cumbersome hats on the Fourier-transformed variables, we note that solving the linear system above is the same as searching for harmonic solutions to equations (21.10) and (21.11) of the form

$$\delta u = \delta u_0 e^{-i\mathbf{k} \cdot \mathbf{x} + \lambda t}, \quad (21.14)$$

$$\delta v = \delta v_0 e^{-i\mathbf{k} \cdot \mathbf{x} + \lambda t}. \quad (21.15)$$

We can then write the resulting system of linear ODEs as

$$\lambda \begin{pmatrix} \delta u \\ \delta v \end{pmatrix} = \mathbf{A} \begin{pmatrix} \delta u \\ \delta v \end{pmatrix}, \quad (21.16)$$

where the linear stability matrix is given by

$$\mathbf{A} = \begin{pmatrix} -k^2 D_u + f_u & f_v \\ g_u & -k^2 D_v + g_v \end{pmatrix}. \quad (21.17)$$

As before, our goal is to compute the eigenvalues λ of this matrix. For a 2×2 matrix, the eigenvalues are

$$A = \frac{1}{2} \left(\text{tr}(A) \pm \sqrt{\text{tr}^2(A) - 4\det(A)} \right), \quad (21.18)$$

where $\text{tr}(A)$ is the trace of A and $\det(A)$ is its determinant.

In the absence of diffusion, $\text{tr}(A) = f_u + g_v$. Because we have specified that in the absence of diffusion, the chemical reaction system has a stable fixed point, at least one of f_u or g_v must be negative. Interestingly, the trace of the linear stability matrix with diffusion included is maximal for the zeroth mode ($k = 0$), which means that an instability arising from the trace becoming positive has the zeroth mode as its fastest growing. If the determinant is positive at the onset of the instability (when the trace crosses zero), the eigenvalues are imaginary, which means that the zeroth mode is oscillatory. This is called a Hopf bifurcation.

We will stipulate that $g_v < 0$, leaving for now the possibility that f_u may be negative as well. In the presence of diffusion, the trace is

$$\text{tr}(A) = f_u + g_v - (D_u + D_v) k^2. \quad (21.19)$$

Since the second term is always negative, and we have decreed that $f_u + g_v < 0$ by virtue of the chemical reaction system being stable in the absence of diffusion, the trace is always negative. As such, we can only get an eigenvalue with a positive real part (an instability) if the determinant is negative.

The determinant of the linear stability matrix is

$$\det(A) = D_u D_v k^4 - (D_v f_u + D_u g_v) k^2 + f_u g_v - f_v g_u. \quad (21.20)$$

This is quadratic in k^2 , and since $D_u D_v > 0$, this is a convex parabola. Therefore, we can calculate the wave number where the parabola is a minimum.

$$\frac{\partial}{\partial k^2} \det(A) = 2D_u D_v k^2 - D_v f_u - D_u g_v = 0. \quad (21.21)$$

Thus, we have

$$k_{\min}^2 = \frac{D_u g_v + D_v f_u}{2D_u D_v}. \quad (21.22)$$

Inserting this expression into the expression for the determinant gives the minimal value of the determinant.

$$\det(A(k_{\min})) = f_u g_v - f_v g_u - \frac{(D_u g_v + D_v f_u)^2}{4D_u D_v}. \quad (21.23)$$

Therefore, the determinant is negative when

$$D_u g_v + D_v f_u > 2\sqrt{D_u D_v (f_u g_v - f_v g_u)}. \quad (21.24)$$

Because we have decreed that the chemical system in the absence of diffusion is stable, the determinant of the linear stability matrix of that system, $f_u g_v - f_v g_u$, must be positive, so the term in parentheses in the square root must be positive. Thus, we must have

$$D_u g_v + D_v f_u > 0 \quad (21.25)$$

in order to have a negative determinant and therefore an instability. Since we already showed that at least one of f_u or g_v must be negative, we must have *exactly* one be negative. We will take $f_u > 0$,

as we already decreed that $g_v < 0$. Note that having $g_v < 0$ does not necessarily mean that we have autoinhibition, since we could also have $g_v < 0$ by degradation or dilution. However, the signs of f_u and g_v indicate that U is autoactivating and is in a negative feedback loop with V.

Finally, let's look again at our expression for k_{\min}^2 , equation (21.22). For k_{\min} to be real, we must have

$$D_v f_u + D_u g_v > 0. \quad (21.26)$$

We also have $f_u + g_v < 0$ from the condition that the trace of the linear stability matrix is negative. So, we have four inequalities,

$$f_u > 0, \quad (21.27)$$

$$g_v < 0, \quad (21.28)$$

$$f_u + g_v < 0, \quad (21.29)$$

$$\frac{D_v}{D_u} f_u + g_v > 0. \quad (21.30)$$

A requirement for all of these inequalities can only hold is that $D_v > D_u$. Thus, we have arrived at our conditions for formation of Turing patterns.

- 1) We need an autocatalytic species (an “activator”).
- 2) The activator must have a negative feedback loop with the other species, the “inhibitor.”
- 3) The inhibitor must diffuse more rapidly than the activator.

The intuition here is that the activator starts producing more of itself locally. The local peak starts to spread, but the inhibitor diffuses more quickly so that it pins the peak of activator in so that it cannot spread. This gives a set wavelength of the pattern of peaks.



Figure 20: Circuits that can form Turing patterns. The dashed line indicates that auto-repression is not necessary, only that $g_v < 0$.

21.2.2 Wavelength of a Turing pattern

The wavelength of a Turing pattern is set, at least as the instability begins to grow, by the *fastest growing mode*, k . That is, for the value of k for which the eigenvalue λ is largest. We have already shown that k_{\min} , the k for which the determinant of the linear stability matrix is minimal, is the k corresponding to the fastest growing mode.

21.2.3 Turing patterns do not scale

Turing patterns do not scale with the size of the organism because the wavelength of the pattern, given by the fastest growing mode, k , is independent of system size. So, if a system is twice as large,

it would have twice as many peaks and valleys in the pattern. In the next lecture, we will address this problem of scaling and talk about reaction-diffusion systems that can give patterns that scale with organism size.

21.3 Example: The Activator-Substrate Depletion Model

As an example of a system that undergoes a Turing pattern, let us consider now the activator-substrate depletion model, or ASDM. This is a simple model in which there are two species, the activator and substrate. The activator is autocatalytic, but consumes substrate in the process. It also has our standard degradation. The substrate is constitutively produced, and for the sake of simplicity, we assume it is very stable such that its degradation is all by consumption by the activator. This corresponds to the right circuit in Fig. 20. The dynamical equations are

$$\frac{\partial a}{\partial t} = D_a \nabla^2 a + \rho a^2 s - \gamma a, \quad (21.31)$$

$$\frac{\partial s}{\partial t} = D_s \nabla^2 s + \beta - \rho a^2 s. \quad (21.32)$$

We nondimensionalize by choosing $t \leftarrow t\gamma$, $(x, y, z) \leftarrow \sqrt{D_s/\gamma}(x, y, z)$, $a \leftarrow \gamma a/\beta$, $s \leftarrow \beta \rho s/\gamma^2$, $d = D_a/D_s$ and $\mu = \beta^2 \rho/\gamma^3$. Then, the dimensionless equations are

$$\frac{\partial a}{\partial t} = d \nabla^2 a + a^2 s - a, \quad (21.33)$$

$$\frac{\partial s}{\partial t} = \nabla^2 s + \mu(1 - a^2 s). \quad (21.34)$$

This system has a unique homogeneous steady state of $a_0 = s_0 = 1$. In the language of our general Turing system, here we have $f_a = f_s = 1$, $g_a = -2\mu$, and $g_s = -\mu$, so we have a linear stability matrix

$$A = \begin{pmatrix} 1 - dk^2 & 1 \\ -2\mu & -\mu - k^2 \end{pmatrix}. \quad (21.35)$$

In order to avoid oscillatory instabilities, we must have $\mu > 1$, since we must have a negative trace of the linear stability matrix in the absence of diffusion. The fastest growing mode is

$$k_{\min}^2 = \frac{1 - d\mu}{2d}. \quad (21.36)$$

For k_{\min} to be real, we must have $d/\mu < 1$, so $d < 1$ since $\mu > 1$.

We can compute the eigenvalues of the linear stability matrix and make a linear stability diagram, shown in Fig. 21. We see that we must have a fast diffusing substrate to get a Turing pattern. An example pattern is given in Fig. 22.

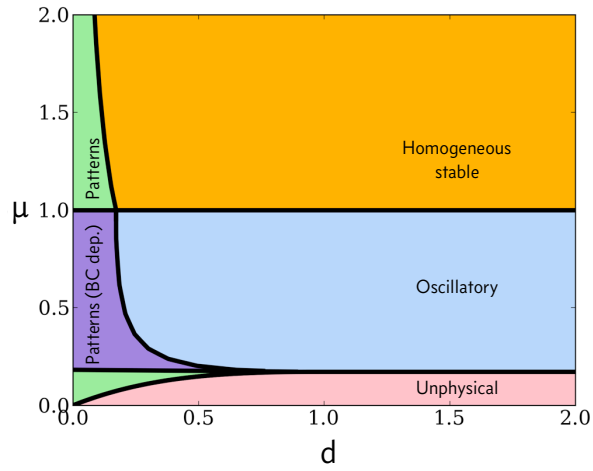


Figure 21: Linear stability diagram for the ASDM.

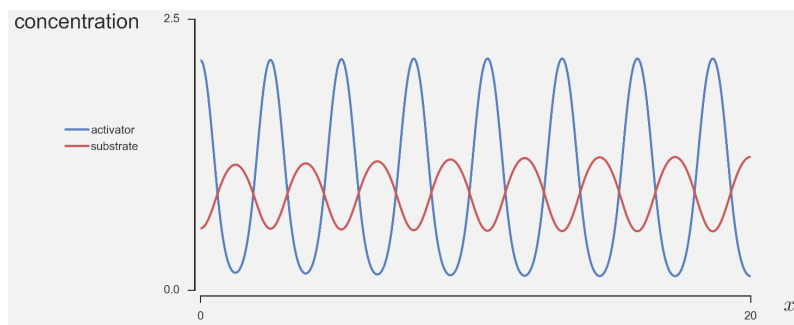


Figure 22: A pattern arising from a one-dimensional activator-substrate depletion model.