Supplemetary Material to

Reaction wavefront theory of notochord segment patterning

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Homogeneous system

The condition for a reaction-diffusion system to produce a pattern is to have a linearly stable state in the absence of diffusion, that becomes unstable when diffusion is introduced [1]. Below we analyze the conditions to form a pattern for the system considered in this work. We first consider the homogeneous system $\partial_{xx} u = \partial_{xx} v = 0$, with a vanishing sink profile strength $s_0 = 0$. Equations for the local reactions are

$$\frac{\partial u}{\partial t} = \gamma \left(u - u^3 - \kappa_4 v \right) \tag{1}$$

$$\frac{\partial v}{\partial t} = \gamma \left(\kappa_5 u - \kappa_6 v \right) . \tag{2}$$

Introducing the functions

$$f(u,v) = u - u^3 - \kappa_4 v \tag{3}$$

$$g(u,v) = \kappa_5 u - \kappa_6 v \,, \tag{4}$$

the nullclines, defined by $f(u,v) = 0$ y $g(u,v) = 0$, are the curves in the (u,v) plane

$$v = \frac{1}{\kappa_4}(u - u^3) \tag{5}$$

$$v = \frac{\kappa_5}{\kappa_6} u \,. \tag{6}$$

The first one is a cubic function that crosses the origin and the second one is a linear function through the origin with slope κ_5/κ_6. The fixed points of the dynamical system are given by the nullclines intersection, Fig. 1, and determined as the points that simultaneously verify $f(u,v) = 0$ and $g(u,v) = 0$,

$$u \left(\left(1 - \frac{\kappa_4 \kappa_5}{\kappa_6} \right) - u^2 \right) = 0 \,. \tag{7}$$

Regardless of parameter values, there is always a solution $(u_0, v_0) = (0,0)$. For $(\kappa_4\kappa_5)/\kappa_6 < 1$ there are two additional solutions

$$u_\pm = \pm\sqrt{1 - \frac{\kappa_4 \kappa_5}{\kappa_6}} \,. \tag{8}$$

For a non-vanishing sink profile $s_0 > 0$ we have

$$g(u,v) = \kappa_5 u - \kappa_6 v - s_0 s(x) v \,, \tag{9}$$

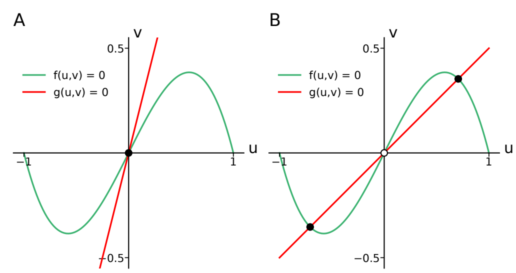

Figure 1. Nullclines for the homogeneous system Eqs. (5) and (6). Depending on parameter values for the local reactions, the system displays (A) a single stable fixed point at $(0,0)$ (solid black dot), or (B) two stable fixed points (solid black dots) and an unstable fixed point (open dot) at $(0,0)$.

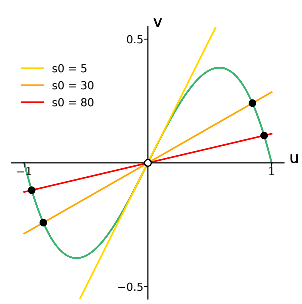

Figure 2. Effective nullclines for the system with sinks Eqs. (5) and (10), for different values of the dimensionless sink strength s_0 as indicated.

and the second nullcline becomes

$$v = \frac{\kappa_5}{\kappa_6 + s_0 s(x)} u \,. \tag{10}$$

Thus, in the presence of sinks the slope of the linear nullcline depends on position x and the sink strength s_0. The effect of sinks on the nullcline can be thought of as an effective κ_6, Fig. 2.

The stability of fixed points can be determined by means of small perturbations and linearization around the equilibrium points. We follow the standard procedure and consider a perturbation $\boldsymbol{w}$ with respect to the fixed point (u_0, v_0)

$$\boldsymbol{w} = \begin{pmatrix} u - u_0 \\ v - v_0 \end{pmatrix} . \tag{11}$$

For a small perturbation, dynamics is given by

$$\frac{d\boldsymbol{w}}{dt} = \gamma A \boldsymbol{w} \quad , \quad A = \begin{pmatrix} f_u & f_v \\ g_u & g_v \end{pmatrix} \tag{12}$$

where

$$\begin{aligned} f_u = \partial_u f(u,v) \,, \quad f_v = \partial_v f(u,v) \,, \\ g_u = \partial_u g(u,v) \,, \quad g_v = \partial_v g(u,v) \end{aligned} \tag{13}$$

and derivatives are to be evaluated at the fixed point (u_0, v_0). Solutions to Eq. (12) are

$$\boldsymbol{w} \propto e^{\Lambda t} \tag{14}$$

with an eigenvalue Λ. Eigenvalues are obtained as the roots of the characteristic polynomial, which is given by the condition

$$|\Lambda I - \gamma A| = 0 \,, \tag{15}$$

where I is the identity matrix. The resulting eigenvalues are

$$\Lambda_{1,2} = \frac{1}{2}\gamma \left[(f_u + g_v) \pm \{ (f_+ g_v)^2 - 4(f_u g_v - f_v g_u) \}^{1/2} \right] . \tag{16}$$

Linear stability of the fixed point requires the real part of these eigenvalues to be negative, $\mathrm{Re}\Lambda < 0$. This translates into the two conditions

$$\det A = f_u g_v - f_v g_u > 0 \,, \tag{17}$$

$$\mathrm{tr}\, A = f_u + g_v < 0 \,. \tag{18}$$

In particular, for the fixed point $(u_0, v_0) = (0, 0)$ we have

$$A = \begin{pmatrix} 1 & -\kappa_4 \\ \kappa_5 & -\kappa_6 \end{pmatrix} \tag{19}$$

and the conditions on the determinant (17) and the trace (18) become

$$\frac{\kappa_4 \kappa_5}{\kappa_6} > 1 \,, \tag{20}$$

$$\kappa_6 > 1 \,. \tag{21}$$

Under these two conditions, the fixed point $(u_0, v_0) = (0, 0)$ is linearly stable in the homogeneous case [2]. Breaking the first condition Eq. (20), the fixed point loses stability to the two fixed points Eq. (8). Breaking the second condition Eq. (21), the fixed point loses stability to an oscillatory state.

Diffusion-driven instability

Above we considered the conditions for the homogeneous system to be stable. Next, we obtain conditions for the homogeneous steady state to become unstable in the presence of diffusion. We follow the standard derivation, as in [2]. Considering now the full system, the linear approximation around the steady state is

$$\frac{\partial \boldsymbol{w}}{\partial t} = \gamma A \boldsymbol{w} + D \nabla^2 \boldsymbol{w} \,, \text{ with } \quad D = \begin{pmatrix} 1 & 0 \\ 0 & \delta \end{pmatrix} . \tag{22}$$

We introduce the time independent solution $\boldsymbol{W}(x)$ for the spatially extended problem

$$\nabla^2 \boldsymbol{W} + k^2 \boldsymbol{W} = 0 \,. \tag{23}$$

With no flux boundary conditions $(\boldsymbol{n} \cdot \nabla)\boldsymbol{W} = 0$, we obtain solutions

$$\boldsymbol{W}(x) \propto \cos(n\pi x / L) \,, \tag{24}$$

where L denotes dimensionless system size and the wave number $k = n\pi/L$ is the eigenvalue of the time independent solution. For the complete system, solutions take the form

$$\boldsymbol{w}(x,t) = \sum_k c_k e^{\Lambda t} \boldsymbol{W}_{\boldsymbol{k}}(x) \tag{25}$$

where the coefficients c_k can be determined from a Fourier expansion of the initial conditions in terms of $\boldsymbol{W}_{\boldsymbol{k}}(x)$. Substituting $\boldsymbol{w}(x,t)$ in the complete system Eq. (22) and dropping the exponentials $e^{\Lambda t}$ we arrive at

$$\Lambda \boldsymbol{W}_{\boldsymbol{k}}(\boldsymbol{x}) = \gamma A \boldsymbol{W}_{\boldsymbol{k}} - D k^2 \boldsymbol{W}_{\boldsymbol{k}} \,. \tag{26}$$

Non-trivial solutions require a vanishing determinant

$$|\Lambda I - \gamma A + D k^2| = 0 \,. \tag{27}$$

We evaluate this determinant and obtain eigenvalues $\Lambda(k)$ as the root of

$$\Lambda^2 + \Lambda[k^2(1+\delta) - \gamma(f_u + g_v)] + h(k^2) = 0 \tag{28}$$

with

$$h(k^2) = \delta k^4 - \gamma(\delta f_u + g_v)k^2 + \gamma^2 |A| \,. \tag{29}$$

The characteristic polynomial for the homogeneous system is recovered for $k = 0$. For a diffusion driven instability to occur, at least one eigenvalue must have a positive real part. This requires that $h(k^2) < 0$ for some $k \neq 0$, which leads to the condition

$$\delta f_u + g_v > 0\,. \tag{30}$$

Together with the condition $f_u + g_v < 0$ of Eq. (18) this implies $\delta \neq 1$ and that f_u and g_v must be of opposite signs. Furthermore, given the signs of the partial derivatives $f_u > 0$ y $g_v < 0$, this imposes a condition on δ, that results in the relation between the diffusion coefficients

$$\delta = \frac{D_V}{D_U} > 1\,. \tag{31}$$

This means that for a diffusion driven instability to occur, the inhibitor must diffuse faster than the activator, as it is well known [3]. For the solution $(u_0, v_0) = (0, 0)$ this further implies

$$\delta - \kappa_6 > 0\,. \tag{32}$$

This condition is necessary but not sufficient to ensure that $h(k^2)$ is negative for some nonzero k. Additionally, we require the minimum of h to be negative, which results in

$$\frac{(\delta f_u + g_v)^2}{4\delta} > |A|\,. \tag{33}$$

For the vanishing state $(u_0, v_0) = (0, 0)$ this becomes

$$\frac{(\delta - \kappa_6)^2}{4\delta} > -\kappa_6 + \kappa_4\kappa_5\,. \tag{34}$$

Putting these results together, the conditions on parameter values that allow for pattern formation through a diffusion driven instability are

$$\begin{gathered} \kappa_6 > 1\,, \quad \frac{\kappa_4\kappa_5}{\kappa_6} > 1\,, \quad \delta > \kappa_6\,, \\ (\delta - \kappa_6)^2 > 4\delta(-\kappa_6 + \kappa_4\kappa_5) \end{gathered} \tag{35}$$

Together, these conditions ensure that there is at least one wave number k such that the corresponding eigenvalue $\Lambda(k^2)$ has a positive real part. Depending on parameter values, there may be more than one mode with an eigenvalue that has a positive real part. For long times, the terms in the solution that correspond to negative eigenvalues decay, while terms with positive eigenvalues contribute to the final state. Thus, it is interesting to consider the range of wavenumbers k with eigenvalues such that $\mathrm{Re}\Lambda(\mathrm{k}^2) > 0$. These are k^2 values that result in $h(k^2) < 0$, so we look for the roots of $h(k^2)$ to find the bounds of this range $k_1^2 < k^2 < k_2^2$,

$$\begin{aligned} k_1^2 &= \frac{\gamma}{2\delta}\left[(\delta f_u + g_v) - \left((\delta f_u + g_v)^2 - 4\delta|A|\right)^{1/2}\right] < k^2 \\ &< \frac{\gamma}{2\delta}\left[(\delta f_u + g_v) + \left((\delta f_u + g_v)^2 - 4\delta|A|\right)^{1/2}\right] = k_2^2 \end{aligned} \tag{36}$$

In particular, for the state $(u_0, v_0) = (0, 0)$ of our system we obtain

$$\begin{aligned} k_1^2 &= \frac{\gamma}{2\delta}\left[(\delta - \kappa_6) - \left((\delta - \kappa_6)^2 - 4\delta\left(-\kappa_6 + \kappa_4\kappa_5\right)\right)^{1/2}\right] < k^2 \\ &< \frac{\gamma}{2\delta}\left[(\delta - \kappa_6) + \left((\delta - \kappa_6)^2 - 4\delta\left(-\kappa_6 + \kappa_4\kappa_5\right)\right)^{1/2}\right] = k_2^2\,. \end{aligned} \tag{37}$$

Considering the decay of terms with negative eigenvalues for long times, the solution in this regime is approximated by

$$\boldsymbol{w}(x, t) \approx \sum_{k_1}^{k_2} c_k e^{\Lambda(k^2)t}\boldsymbol{W_k}(x)\,. \tag{38}$$

Among the modes with positive eigenvalues, the fastest growing is the one that maximizes $\mathrm{Re}\Lambda(\mathrm{k}^2) > 0$. The corresponding wave number is

$$k_0^2 = \frac{\gamma}{\delta - 1}\left((\delta + 1)\left[-\frac{f_v g_u}{\delta}\right]^{1/2} - f_u + g_v\right)\,, \tag{39}$$

which for the stable state $(u_0, v_0) = (0, 0)$ results in

$$k_0^2 = \frac{\gamma}{\delta - 1}\left((\delta + 1)\left[\frac{\kappa_4\kappa_5}{\delta}\right]^{1/2} - 1 - \kappa_6\right)\,. \tag{40}$$

This mode will have the characteristic wavelength

$$\lambda_0 = \frac{2\pi}{k_0} \tag{41}$$

Thus, in the absence of any external stimulus, with random initial conditions, the mode that grows faster corresponds to this wavelength λ_0. Both the range of wavelengths that are compatible with pattern formation and the natural wavelength λ_0 depend on parameter values. For example, the parameter δ can control the range of wave numbers compatible with pattern formation, which expands for larger δ. Decreasing the value of γ reduces the wavenumber k_0 and therefor results in a larger selected wavelength λ_0. The signs of partial derivatives $f(u, v)$ and $g(u, v)$ determine that both species co-localize instead of alternating in space [2].

In this work, we used the conditions we have obtained in these supplementary notes to choose dimensionless parameter values that ensure the possibility of pattern formation through a diffusion driven instability,

$$\gamma = 1000, \quad \delta = 100, \quad \kappa_4 = 1, \quad \kappa_5 = 10, \quad \kappa_6 = 5\,, \tag{42}$$

verifying all inequalities in Eq. (35). With this choice, the range of wavelengths supported is $0.210 < \lambda < 0.840$ and the natural wavelength is $\lambda_0 = 0.388$.

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