Electronic Supplementary Information for "The Alternate Ligand Jagged Enhances the Robustness of Notch Signaling Patterns"

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The description of the video files

Video link: click here

Video S1: Spatiotemporal patterns of Delta (D) starting from uniform initial condition with small fluctuations for $\lambda_N = 5.0$, $\lambda_D = 10.0$, $\lambda_J = 0.5$ and $L = 6$. All other parameters are standard.

Video S2: Spatiotemporal patterns of Delta (D) starting from uniform initial condition with small fluctuations for $\lambda_N = 5.0$, $\lambda_D = 10.0$, $\lambda_J = 0.5$ and $L = 50$. All other parameters are standard.

Video S3: Spatiotemporal patterns of Delta (D) starting from uniform initial condition with small fluctuations for $\lambda_N = 1.5$, $\lambda_D = 10.0$ and $\lambda_J = 0.1$. All other parameters are standard.

Video S4: Spatiotemporal patterns of Delta (D) starting from a hexagonal seed for $\lambda_N = 5.0$, $\lambda_D = 10.0$ and $\lambda_J = 0.5$. All other parameters are standard.

Video S5: Spatiotemporal patterns of Delta (D) starting from a hexagonal seed for $\lambda_N = 5.0$, $\lambda_D = 10.0$ and $\lambda_J = 0.9$. All other parameters are standard.

The reduced set of 12 ODE's

$$\dot{N}_A = \lambda_N(1 + I_A^{n_N}/(1 + I_A^{n_N})) - N_A(k_c(D_A + J_A) + 0.5k_t(D_B + D_C + J_B + J_C)) - \gamma N_A$$
$$\dot{N}_B = \lambda_N(1 + I_B^{n_N}/(1 + I_B^{n_N})) - N_B(k_c(D_B + J_B) + 0.5k_t(D_C + D_A + J_C + J_A)) - \gamma N_B$$
$$\dot{N}_C = \lambda_N(1 + I_C^{n_N}/(1 + I_C^{n_N})) - N_C(k_c(D_C + J_C) + 0.5k_t(D_A + D_B + J_A + J_B)) - \gamma N_C$$
$$\dot{D}_A = \lambda_D/(1 + I_A^{n_D}) - D_A(k_c N_A + 0.5k_t(N_B + N_C)) - \gamma D_A$$
$$\dot{D}_B = \lambda_D/(1 + I_B^{n_D}) - D_B(k_c N_B + 0.5k_t(N_C + N_A)) - \gamma D_B$$
$$\dot{D}_C = \lambda_D/(1 + I_C^{n_D}) - D_C(k_c N_C + 0.5k_t(N_A + N_B)) - \gamma D_C$$
$$\dot{J}_A = \lambda_J(1 + I_A^{n_J}/(1 + I_A^{n_J})) - J_A(k_c N_A + 0.5k_t(N_B + N_C)) - \gamma J_A$$
$$\dot{J}_B = \lambda_J(1 + I_B^{n_J}/(1 + I_B^{n_J})) - J_B(k_c N_B + 0.5k_t(N_C + N_A)) - \gamma J_B$$
$$\dot{J}_C = \lambda_J(1 + I_C^{n_J}/(1 + I_C^{n_J})) - J_C(k_c N_C + 0.5k_t(N_A + N_B)) - \gamma J_C$$
$$\dot{I}_A = 0.5k_t N_A(D_B + D_C + J_B + J_C) - \gamma I_A$$
$$\dot{I}_B = 0.5k_t N_B(D_C + D_A + J_C + J_A) - \gamma I_B$$
$$\dot{I}_C = 0.5k_t N_C(D_A + D_B + J_A + J_B) - \gamma I_C$$

Method

Linear Stability Analysis:

At first, we calculate numerically the steady state solutions (fixed points), where $\dot{N}_{(A,B,C)} = \dot{D}_{(A,B,C)} = \dot{J}_{(A,B,C)} = \dot{I}_{(A,B,C)} = 0$.

In general three kinds of solutions are possible:
(1) Uniform (U): $\Delta N = \Delta D = \Delta J = \Delta I = 0$,
(2) Hexagon (H): $\Delta N < 0$, $\Delta D > 0$, $\Delta I < 0$ and ΔJ can be positive or negative depending on the parameters and
(3) Antihexagon (A): $\Delta N > 0$, $\Delta D < 0$, $\Delta I > 0$ and ΔJ can be positive or negative depending on the parameters,
where ΔN, ΔD, ΔJ and ΔI are defined as $(N_A - N_B)$, $(D_A - D_B)$, $(J_A - J_B)$ and $(I_A - I_B)$ respectively and $(N, D, J, I)_B = (N, D, J, I)_C \neq (N, D, J, I)_A$.

Afterwards, we analyze their stability via linear stability analysis across the parameter space by calculating the eigenvalues (eigv) of the Jacobian matrix at those fixed points:

$$\begin{bmatrix}
\frac{\partial \dot{N}_A}{\partial N_A} & \frac{\partial \dot{N}_A}{\partial N_B} & \frac{\partial \dot{N}_A}{\partial N_C} & \frac{\partial \dot{N}_A}{\partial D_A} & \frac{\partial \dot{N}_A}{\partial D_B} & \frac{\partial \dot{N}_A}{\partial D_C} & \frac{\partial \dot{N}_A}{\partial J_A} & \frac{\partial \dot{N}_A}{\partial J_B} & \frac{\partial \dot{N}_A}{\partial J_C} & \frac{\partial \dot{N}_A}{\partial I_A} & \frac{\partial \dot{N}_A}{\partial I_B} & \frac{\partial \dot{N}_A}{\partial I_C} \\
\frac{\partial \dot{N}_B}{\partial N_A} & \frac{\partial \dot{N}_B}{\partial N_B} & \frac{\partial \dot{N}_B}{\partial N_C} & \frac{\partial \dot{N}_B}{\partial D_A} & \frac{\partial \dot{N}_B}{\partial D_B} & \frac{\partial \dot{N}_B}{\partial D_C} & \frac{\partial \dot{N}_B}{\partial J_A} & \frac{\partial \dot{N}_B}{\partial J_B} & \frac{\partial \dot{N}_B}{\partial J_C} & \frac{\partial \dot{N}_B}{\partial I_A} & \frac{\partial \dot{N}_B}{\partial I_B} & \frac{\partial \dot{N}_B}{\partial I_C} \\
\frac{\partial \dot{N}_C}{\partial N_A} & \frac{\partial \dot{N}_C}{\partial N_B} & \frac{\partial \dot{N}_C}{\partial N_C} & \frac{\partial \dot{N}_C}{\partial D_A} & \frac{\partial \dot{N}_C}{\partial D_B} & \frac{\partial \dot{N}_C}{\partial D_C} & \frac{\partial \dot{N}_C}{\partial J_A} & \frac{\partial \dot{N}_C}{\partial J_B} & \frac{\partial \dot{N}_C}{\partial J_C} & \frac{\partial \dot{N}_C}{\partial I_A} & \frac{\partial \dot{N}_C}{\partial I_B} & \frac{\partial \dot{N}_C}{\partial I_C} \\
\frac{\partial \dot{D}_A}{\partial N_A} & \frac{\partial \dot{D}_A}{\partial N_B} & \frac{\partial \dot{D}_A}{\partial N_C} & \frac{\partial \dot{D}_A}{\partial D_A} & \frac{\partial \dot{D}_A}{\partial D_B} & \frac{\partial \dot{D}_A}{\partial D_C} & \frac{\partial \dot{D}_A}{\partial J_A} & \frac{\partial \dot{D}_A}{\partial J_B} & \frac{\partial \dot{D}_A}{\partial J_C} & \frac{\partial \dot{D}_A}{\partial I_A} & \frac{\partial \dot{D}_A}{\partial I_B} & \frac{\partial \dot{D}_A}{\partial I_C} \\
\frac{\partial \dot{D}_B}{\partial N_A} & \frac{\partial \dot{D}_B}{\partial N_B} & \frac{\partial \dot{D}_B}{\partial N_C} & \frac{\partial \dot{D}_B}{\partial D_A} & \frac{\partial \dot{D}_B}{\partial D_B} & \frac{\partial \dot{D}_B}{\partial D_C} & \frac{\partial \dot{D}_B}{\partial J_A} & \frac{\partial \dot{D}_B}{\partial J_B} & \frac{\partial \dot{D}_B}{\partial J_C} & \frac{\partial \dot{D}_B}{\partial I_A} & \frac{\partial \dot{D}_B}{\partial I_B} & \frac{\partial \dot{D}_B}{\partial I_C} \\
\frac{\partial \dot{D}_C}{\partial N_A} & \frac{\partial \dot{D}_C}{\partial N_B} & \frac{\partial \dot{D}_C}{\partial N_C} & \frac{\partial \dot{D}_C}{\partial D_A} & \frac{\partial \dot{D}_C}{\partial D_B} & \frac{\partial \dot{D}_C}{\partial D_C} & \frac{\partial \dot{D}_C}{\partial J_A} & \frac{\partial \dot{D}_C}{\partial J_B} & \frac{\partial \dot{D}_C}{\partial J_C} & \frac{\partial \dot{D}_C}{\partial I_A} & \frac{\partial \dot{D}_C}{\partial I_B} & \frac{\partial \dot{D}_C}{\partial I_C} \\
\frac{\partial \dot{J}_A}{\partial N_A} & \frac{\partial \dot{J}_A}{\partial N_B} & \frac{\partial \dot{J}_A}{\partial N_C} & \frac{\partial \dot{J}_A}{\partial D_A} & \frac{\partial \dot{J}_A}{\partial D_B} & \frac{\partial \dot{J}_A}{\partial D_C} & \frac{\partial \dot{J}_A}{\partial J_A} & \frac{\partial \dot{J}_A}{\partial J_B} & \frac{\partial \dot{J}_A}{\partial J_C} & \frac{\partial \dot{J}_A}{\partial I_A} & \frac{\partial \dot{J}_A}{\partial I_B} & \frac{\partial \dot{J}_A}{\partial I_C} \\
\frac{\partial \dot{J}_B}{\partial N_A} & \frac{\partial \dot{J}_B}{\partial N_B} & \frac{\partial \dot{J}_B}{\partial N_C} & \frac{\partial \dot{J}_B}{\partial D_A} & \frac{\partial \dot{J}_B}{\partial D_B} & \frac{\partial \dot{J}_B}{\partial D_C} & \frac{\partial \dot{J}_B}{\partial J_A} & \frac{\partial \dot{J}_B}{\partial J_B} & \frac{\partial \dot{J}_B}{\partial J_C} & \frac{\partial \dot{J}_B}{\partial I_A} & \frac{\partial \dot{J}_B}{\partial I_B} & \frac{\partial \dot{J}_B}{\partial I_C} \\
\frac{\partial \dot{J}_C}{\partial N_A} & \frac{\partial \dot{J}_C}{\partial N_B} & \frac{\partial \dot{J}_C}{\partial N_C} & \frac{\partial \dot{J}_C}{\partial D_A} & \frac{\partial \dot{J}_C}{\partial D_B} & \frac{\partial \dot{J}_C}{\partial D_C} & \frac{\partial \dot{J}_C}{\partial J_A} & \frac{\partial \dot{J}_C}{\partial J_B} & \frac{\partial \dot{J}_C}{\partial J_C} & \frac{\partial \dot{J}_C}{\partial I_A} & \frac{\partial \dot{J}_C}{\partial I_B} & \frac{\partial \dot{J}_C}{\partial I_C} \\
\frac{\partial \dot{I}_A}{\partial N_A} & \frac{\partial \dot{I}_A}{\partial N_B} & \frac{\partial \dot{I}_A}{\partial N_C} & \frac{\partial \dot{I}_A}{\partial D_A} & \frac{\partial \dot{I}_A}{\partial D_B} & \frac{\partial \dot{I}_A}{\partial D_C} & \frac{\partial \dot{I}_A}{\partial J_A} & \frac{\partial \dot{I}_A}{\partial J_B} & \frac{\partial \dot{I}_A}{\partial J_C} & \frac{\partial \dot{I}_A}{\partial I_A} & \frac{\partial \dot{I}_A}{\partial I_B} & \frac{\partial \dot{I}_A}{\partial I_C} \\
\frac{\partial \dot{I}_B}{\partial N_A} & \frac{\partial \dot{I}_B}{\partial N_B} & \frac{\partial \dot{I}_B}{\partial N_C} & \frac{\partial \dot{I}_B}{\partial D_A} & \frac{\partial \dot{I}_B}{\partial D_B} & \frac{\partial \dot{I}_B}{\partial D_C} & \frac{\partial \dot{I}_B}{\partial J_A} & \frac{\partial \dot{I}_B}{\partial J_B} & \frac{\partial \dot{I}_B}{\partial J_C} & \frac{\partial \dot{I}_B}{\partial I_A} & \frac{\partial \dot{I}_B}{\partial I_B} & \frac{\partial \dot{I}_B}{\partial I_C} \\
\frac{\partial \dot{I}_C}{\partial N_A} & \frac{\partial \dot{I}_C}{\partial N_B} & \frac{\partial \dot{I}_C}{\partial N_C} & \frac{\partial \dot{I}_C}{\partial D_A} & \frac{\partial \dot{I}_C}{\partial D_B} & \frac{\partial \dot{I}_C}{\partial D_C} & \frac{\partial \dot{I}_C}{\partial J_A} & \frac{\partial \dot{I}_C}{\partial J_B} & \frac{\partial \dot{I}_C}{\partial J_C} & \frac{\partial \dot{I}_C}{\partial I_A} & \frac{\partial \dot{I}_C}{\partial I_B} & \frac{\partial \dot{I}_C}{\partial I_C}
\end{bmatrix}$$

It is essential to note that the stability matrix is derived from the full 12 variable dynamical system even though the base states all have the same concentration values at the B and C sites. Demanding that the perturbation obey this symmetry and reducing the full system to 8 equations misses the instability which restricts the allowed range of anti-hexagon patterns.

To calculate the bifurcation diagrams we note the following:
(1) If Re(eigv)< 0, the fixed points are linearly stable and
(2) If Re(eigv)> 0, the number of positive eigenvalues defines the number of unstable modes of the corresponding fixed points.

Initial conditions

To generate the patterns on the lattice of size L, as shown in Fig. 4 in the main text, we integrate the the system of $4 * L^2$ equations (Eq. 1 in the main text) by Euler method with time step $= 0.1$ using periodic boundary condition. We use the following initial conditions:

$$\begin{aligned}
&N_i(t=0) = \lambda_N \\
&D_i(t=0) = \epsilon\lambda_D(1+\sigma U_i) \\
&J_i(t=0) = \epsilon\lambda_J(1+\sigma U_i) \\
&I_i(t=0) = 0
\end{aligned}$$

where, ϵ is a small number ($\epsilon = 10^{-5}$), σ is the amplitude of the noise ($\sigma = 0.5$) and U_i is a uniform random number between -0.5 and 0.5.

For the pattern formation in the bistable region (as shown in Fig. 7 in the main text), we use a hexagonal seed (hexagon solution where $\Delta N < 0$, $\Delta D > 0$ and $\Delta I < 0$ with some noise on 6 cells) in the center of the lattice and the previous initial condition on the rest of the lattice.

Parameters

Typically considering the number of proteins in the membrane up to few thousand per cell, we scale those values by 10^3. All the parameters and their values and unit are given in the Tables below:

Parameters					
Figures	λ_N	λ_D	λ_J	k_c	k_t
Figure 2	1.822, 0.9, 5.0	$[1.0-4.0]$	0.1	0.1	0.04
Figure 3	$[0.5-3.0]$	$[0.01-40.0]$	$[0.01-3.5]$	0.1	0.04
Figure 4	5.0	10.0, 20.0	0.001, 0.5, 0.65	0.1	0.04
Figure 5	5.0	5.0, 10.0, 20.0, 40.0	$[0.001-10]$	0.1	0.04
Figure 6	5.0	20.0	$[0.001-4.5]$	$[0.001-0.12]$	$[0.01-0.09]$
Figure 7	5.0	10.0	0.5, 0.9	0.1	0.04
Figure S1	5.0	10.0	0.001, 0.5	0.1	0.04
Figure S2	5.0	5.0, 10.0, 20.0	0.001, 0.5	0.1	0.04
Figure S3	$[1.5-3.0]$	10.0	$[0.1-1.3]$	0.1	0.04
Figure S4	$[1.5-5.0]$	$[5.0-40.0]$	$[0.001-10]$	0.1	0.04
Figure S5	$[1.0-5.0]$	$[0.01-50.0]$	0.001, 0.1	$[0.001-0.12]$	0.04
Figure S6	2.5, 5.0, 10.0	$[0.01-40.0]$	$[0.01-7]$	0.05, 0.1, 0.2	0.02, 0.04, 0.08
Other parameters for all the Figures: $\gamma = 0.1$, $\gamma_I = 0.5$, $n_N = n_D = 2$ and $n_J = 5$					

TABLE S1: **Parameters for all the Figures described in the manuscript.**

Unit of the Parameters	
Parameters	Unit
λ_N, λ_D, λ_J	$(10^{-3}$ molecules per cell$)\text{h}^{-1}$
k_c, k_t, γ, γ_I	time^{-1} (h^{-1})
n_N, n_D, n_J	Dimensionless

TABLE S2: **Unit of all the parameters described in the manuscript.**

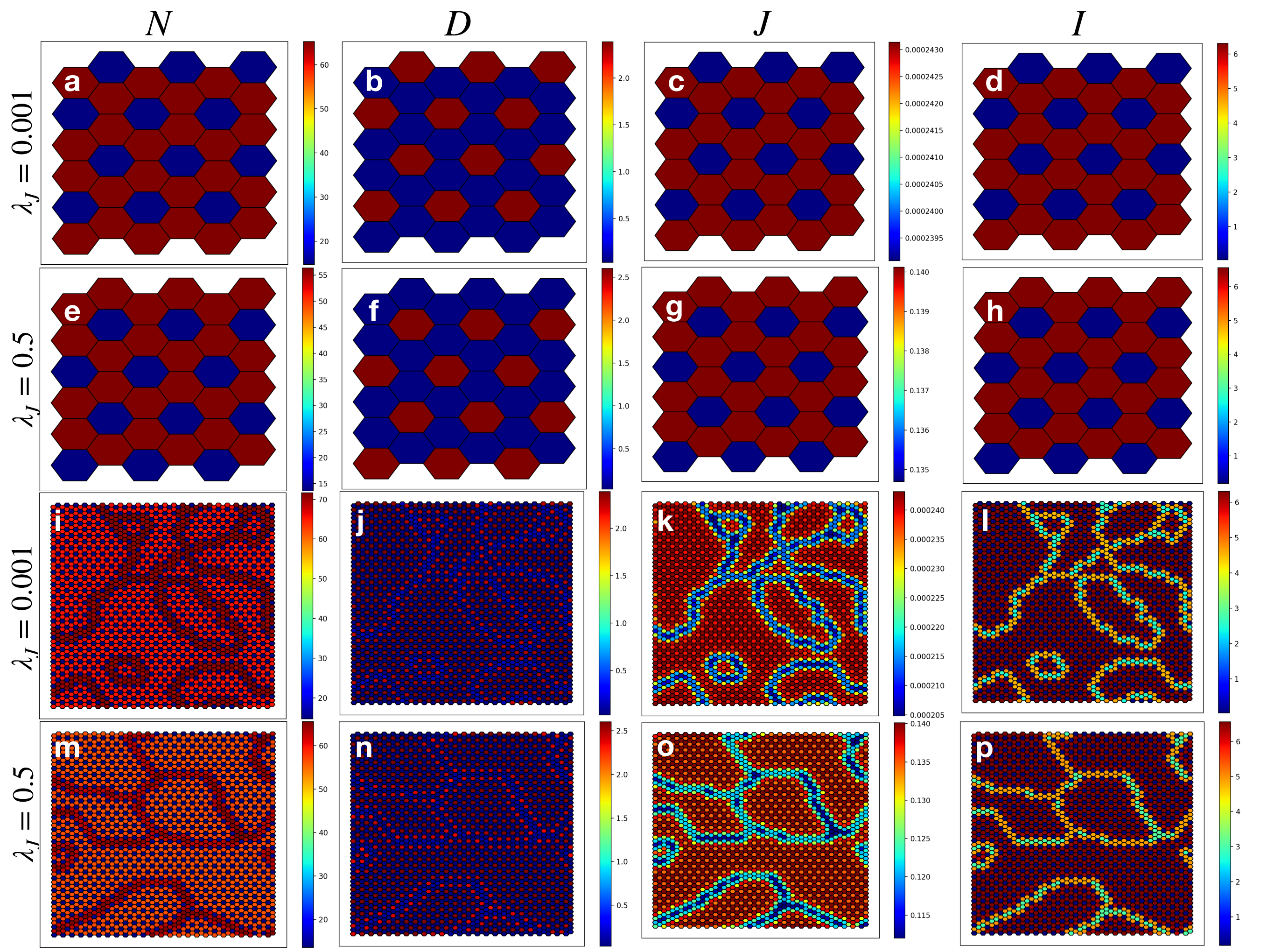

FIG. S1: **Steady state patterns of Notch (N), Delta (D), Jagged (J) and NICD (I) on a hexagonal lattice.** The steady states of Notch (N), Delta (D), Jagged (J) and NICD (I) at $\lambda_N = 5.0$, $\lambda_D = 10.0$ and $\lambda_J = 0.001$ and 0.5 on a hexagonal lattice of size (a–h) $L = 6$ and (i–p) $L = 50$. All other parameters are standard.

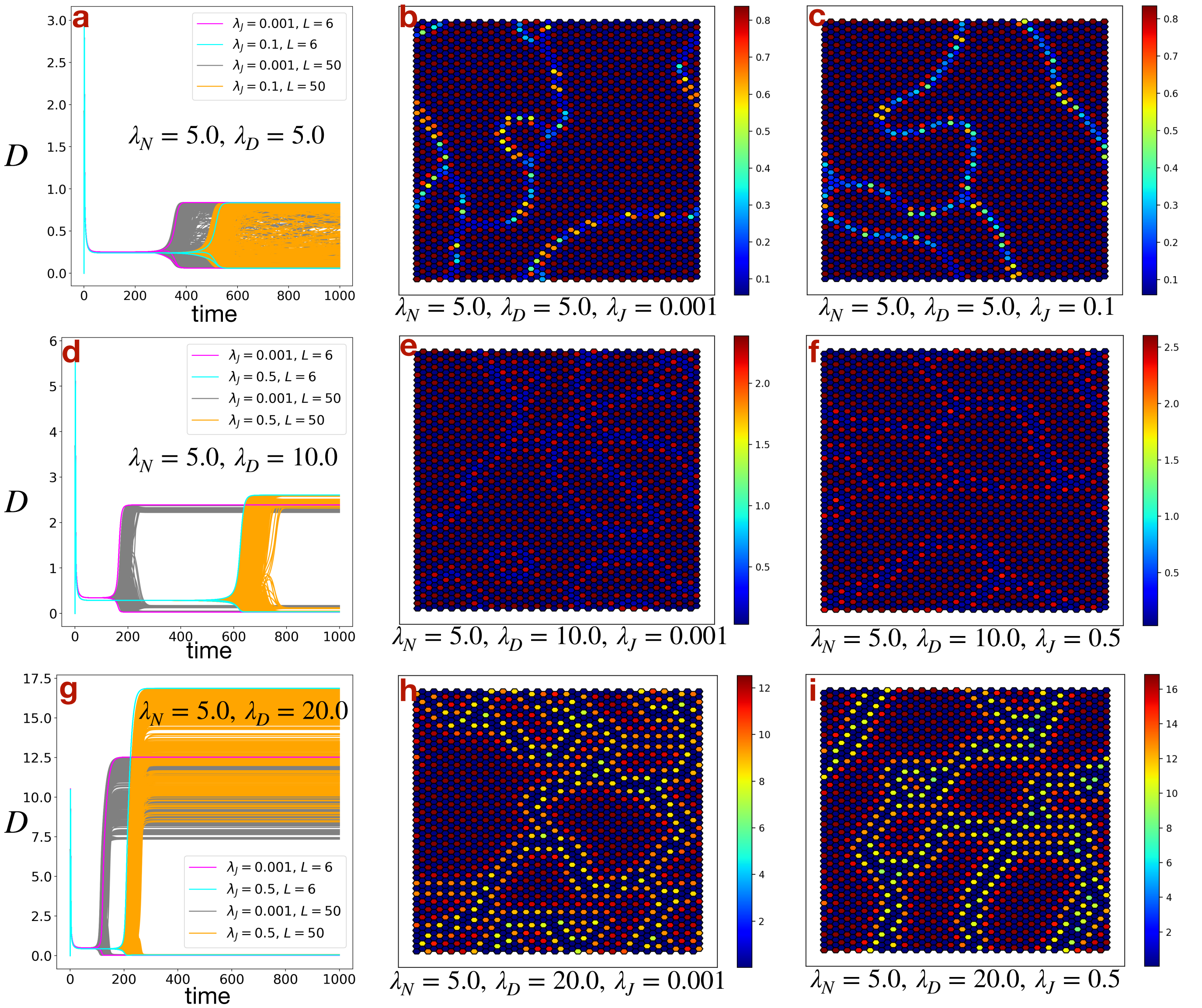

FIG. S2: **Effect of production rate of Delta (λ_D).** Dynamics of Delta (D) for all the cells on a hexagonal lattice of size $L = 6$ and $L = 50$ at $\lambda_N = 5.0$, $\lambda_J = 0.001$ and 0.5 (0.1 instead of 0.5 for $\lambda_D = 5.0$, because at $\lambda_J = 0.5$, $\lambda_D = 5.0$ and $\lambda_N = 5.0$, the steady states become uniform (U)) for (a) $\lambda_D = 5.0$, (d) $\lambda_D = 10.0$ and (g) $\lambda_D = 20.0$. (b), (c), (e), (f), (h), (i) Corresponding patterns of Delta (D) at steady states for $L = 50$ for different values of λ_D and λ_J. All other parameters are standard.

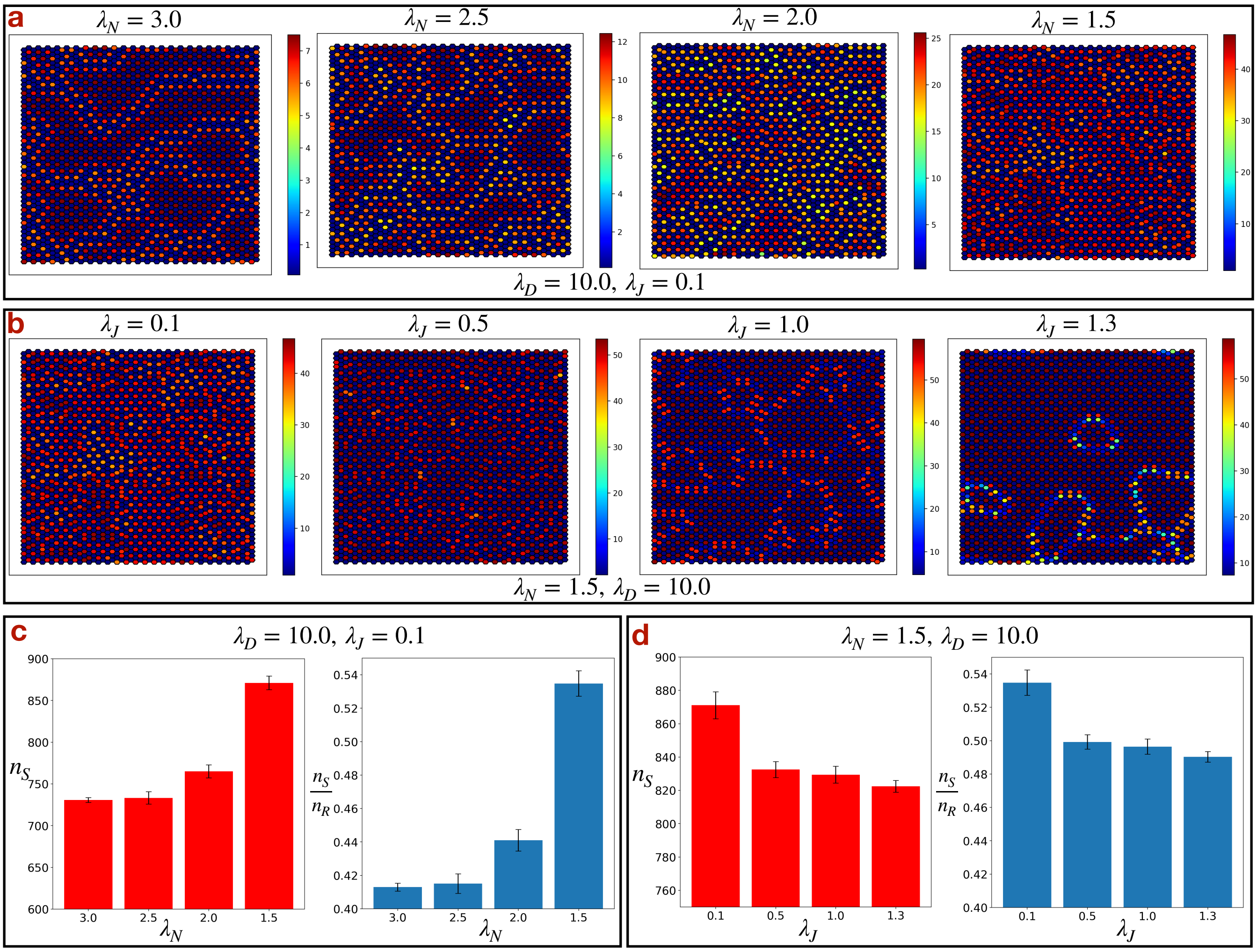

FIG. S3: **Effect of production rate of Notch (λ_N).** The steady state patterns of Delta (D) on a hexagonal lattice of size $L = 50$ (a) for varying λ_N at $\lambda_D = 10.0$, $\lambda_J = 0.1$ and (b) for varying λ_J at $\lambda_N = 1.5$, $\lambda_D = 10.0$. (c), (d) The corresponding number of Sender (S) states (n_S) and ratio of number of Sender (S) and Receiver (R) states ($\frac{n_S}{n_R}$). All other parameters are standard.

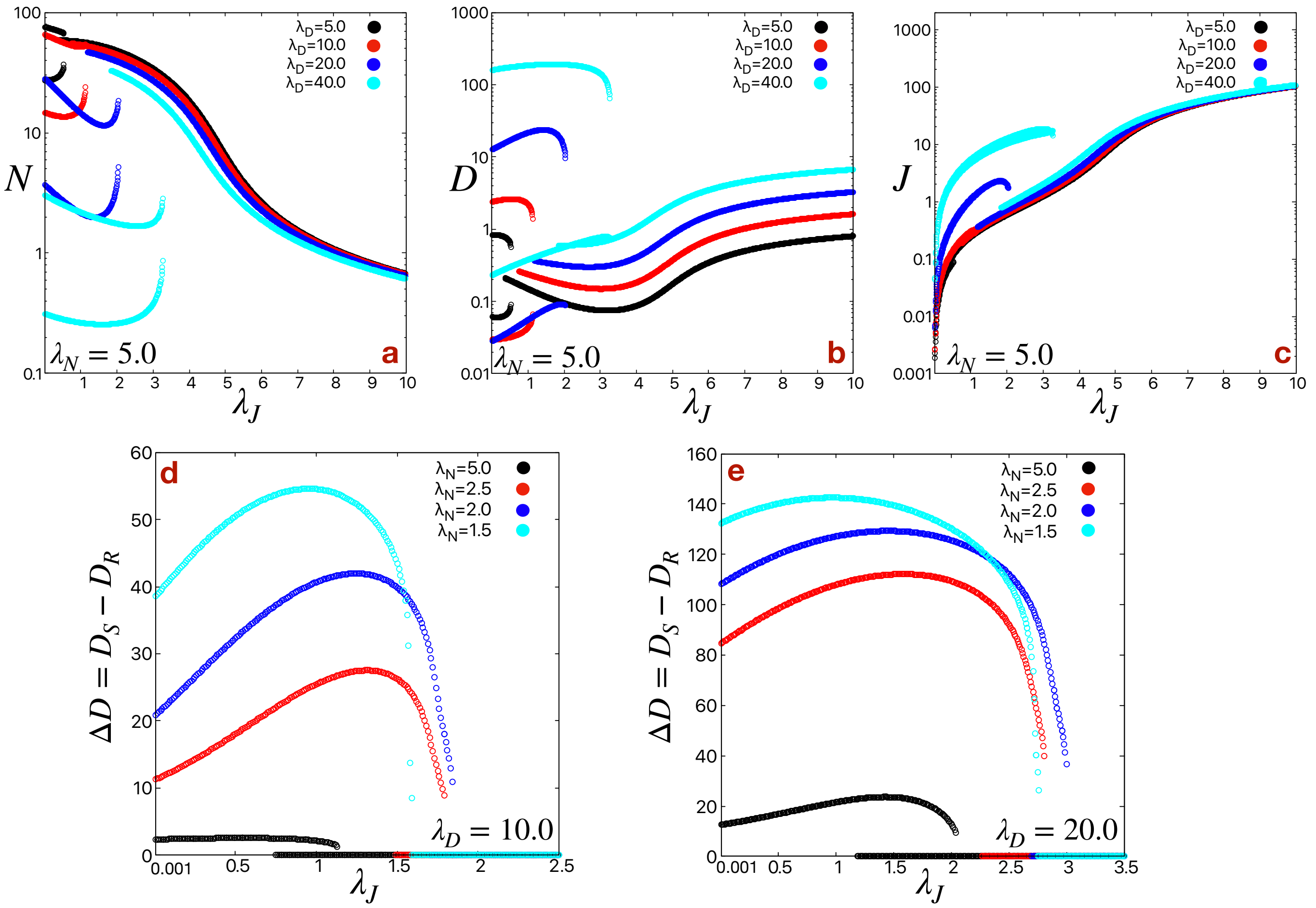

FIG. S4: **Steady state values of Notch (N), Delta (D) and Jagged (J) as a function of λ_J.** Steady state values of (a) Notch (N), (b) Delta (D), (c) Jagged (J) as a function of λ_J for different values of λ_D at $\lambda_N = 5.0$. The y-axes (N, D and J-axes) are in log-scale. The difference in Delta (ΔD) between the Sender (S) and Receiver (R) states ($D_S - D_R$) as a function of λ_J for different values of λ_N at (d) $\lambda_D = 10.0$ and (e) $\lambda_D = 20.0$. All other parameters are standard.

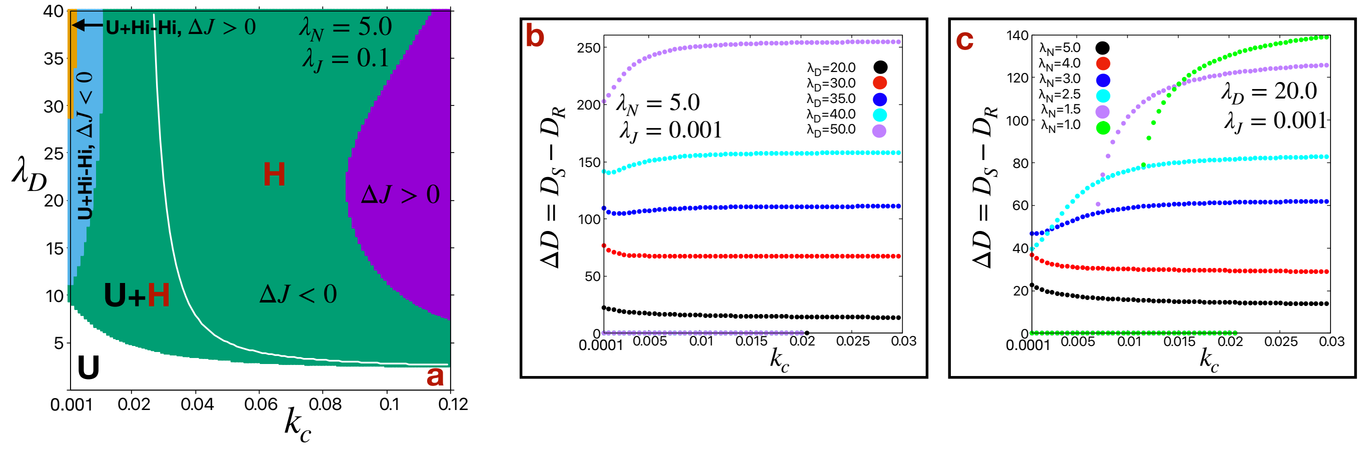

FIG. S5: **Effect of rate of cis-inhibition (k_c).** (a) Phase diagrams in $\lambda_D - k_c$ plane for $\lambda_N = 5.0$, $\lambda_J = 0.1$. The white and colored (green: $\Delta J < 0$) and purple: $\Delta J > 0$)) regions represent uniform (U: $\Delta N = \Delta D = 0$) and hexagon (H: $\Delta N < 0$, $\Delta D > 0$) phases respectively. The cyan ($\Delta J < 0$) and orange ($\Delta J > 0$) colored region represents the bistability of uniform (U) and High-High (Hi-Hi: $\Delta N > 0$, $\Delta D > 0$) phases. The white line represents the boundary of U region. ΔN, ΔD and ΔJ are defined as $(N_S - N_R)$, $(D_S - D_R)$ and $(J_S - J_R)$ respectively, where S and R represent the Sender (high D, low N) and Receiver (low D, high N) states respectively. The difference in Delta (ΔD) between the Sender (S) and Receiver (R) states $(D_S - D_R)$ as a function of k_c (b) at $\lambda_N = 5.0$, $\lambda_J = 0.001$ for different values of λ_D and (c) at $\lambda_D = 20.0$, $\lambda_J = 0.001$ for different values of λ_N. All other parameters are standard.

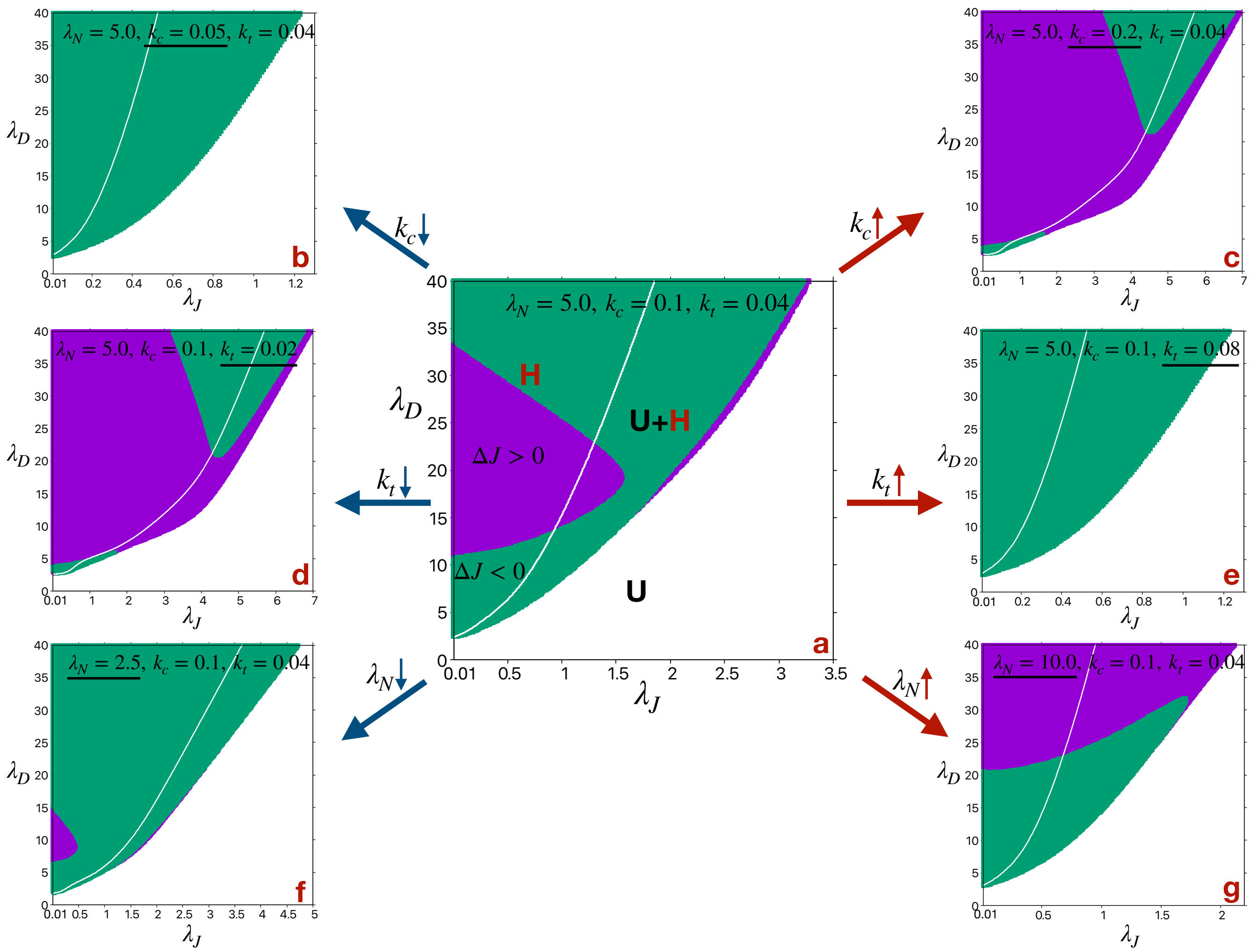

FIG. S6: **Change in phase diagram in $\lambda_D - \lambda_J$ plane.** The standard phase diagram (a) in $\lambda_D - \lambda_J$ plane at $\lambda_N = 5.0$, $k_c = 0.1$, $k_t = 0.04$ changes, especially the region of bistability (where both the uniform (U: $\Delta N = \Delta D = 0$) and hexagon (H: $\Delta N < 0$, $\Delta D > 0$) phases are stable), with the decrease and increase in (b–c) k_c, (d–e) k_t and (f–g) λ_N. The white and colored (green: $\Delta J < 0$) and purple: $\Delta J > 0$)) regions represent uniform (U) and hexagon (H) phases respectively. The white lines represent the boundary of U region. ΔN, ΔD and ΔJ are defined as $(N_S - N_R)$, $(D_S - D_R)$ and $(J_S - J_R)$ respectively, where S and R represent the Sender (high D, low N) and Receiver (low D, high N) states respectively. All other parameters are standard.