Text S1

Does the potential for chaos constrain the embryonic cell-cycle oscillator?

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Cell-cycle model

The most current model of the embryonic cell cycle in *X. laevis* was derived by Pomerening *et al.* [1] In the current study, we extended this model to include a spatial component. Simulations were performed in 1D and 3D geometries, with all species able to diffuse.

At a qualitative level, oscillations in Cdk1 activity are driven by dual fast positive feedbacks and a slower negative feedback. The period of the oscillations is determined by the rate of cyclin B synthesis [2]. Cdk1 binds cyclin B (forming Cyclin B-Cdk1), and, upon activation, the active Cyclin B-Cdk1 complex phosphorylates the mitotic substrates that transition the cell from interphase to M-phase. The transition from M-phase back to interphase is driven by anaphase-promoting complex (APC)-mediated polyubiquitination of cyclin B [3, 4, 5].

In the Pomerening *et al.* [1] model, the dynamics of cyclin B and the phosphorylated/dephosphorylated states of the Cyclin B-Cdk1 complex are explicitly modeled. Phosphorylation at Tyr-15 results in inactivation of the Cyclin B-Cdk1 complex; namely, Cyclin B-Cdk1-Yp and Cyclin B-Cdk1-YpTp (also phosphorylated at Thr-14) are inactive. The complex Cyclin B-Cdk1-Tp is phosphorylated at Thr-14 and lacks phosphorylation at Tyr-15.

To achieve negative feedback, the presence of the complex Cyclin B-Cdk1-Tp results in activation of the polo-like kinase, $\text{Plx1}|_{\text{act}}$. $\text{Plx1}|_{\text{act}}$ then phosphorylates and activates the anaphase-promoting complex, $\text{APC}|_{\text{act}}$, which destroys cyclins and Cyclin B-Cdk1 complexes. Dual positive feedbacks are achieved through the Cyclin B-Cdk1-Tp-mediated activation of the Tyr-15 phosphatase, $\text{Cdc25}|_{\text{act}}$, and the Cyclin B-Cdk1-Tp-mediated inactivation of the Tyr-15 kinase $\text{Wee1}|_{\text{act}}$.

Central parameters in the model include k_{synth}, the rate of cyclin B synthesis; k_{dest}, the rate at which active APC destroys cyclins; and r, the strength of the positive feedbacks. The active forms of Cdc25 and Wee1 ($\text{Cdc25}|_{\text{act}}$ and $\text{Wee1}|_{\text{act}}$) have phosphatase and kinase activities given by k_{cdc25} and k_{wee1}, respectively. The basal activities of Cdc25 and Wee1 are k_{wee1}/r and k_{cdc25}/r, where the parameter r is the fraction of phosphatase/kinase activity in interphase versus M-phase. When $r = 1$, the model becomes negative-feedback-only.

$$
\begin{aligned}
\frac{\partial[\text{Cyclin B}]}{\partial t} &= D\nabla^2[\text{Cyclin B}] \\
&\quad + k_{\text{synth}} - k_{\text{dest}}[\text{APC}]_{\text{act}}[\text{Cyclin B}] - k_a\left([\text{Cdk1}]_{\text{tot}}\right. \\
&\quad - [\text{Cyclin B-Cdk1}] - [\text{Cyclin B-Cdk1-Yp}] \\
&\quad \left. - [\text{Cyclin B-Cdk1-YpTp}] - [\text{Cyclin B-Cdk1-Tp}]\right)[\text{Cyclin B}] \\
&\quad + k_d[\text{Cyclin B-Cdk1}] \\
\frac{\partial[\text{Cyclin B-Cdk1}]}{\partial t} &= D\nabla^2[\text{Cyclin B-Cdk1}] \\
&\quad + k_a\left([\text{Cdk1}]_{\text{tot}} - [\text{Cyclin B-Cdk1}] - [\text{Cyclin B-Cdk1-Yp}]\right. \\
&\quad \left. - [\text{Cyclin B-Cdk1-YpTp}] - [\text{Cyclin B-Cdk1-Tp}]\right)[\text{Cyclin B}] \\
&\quad - k_d[\text{Cyclin B-Cdk1}] - k_{\text{dest}}[\text{APC}]_{\text{act}}[\text{Cyclin B-Cdk1}]
\end{aligned}
$$

$$
\begin{aligned}
& -k_{\text{wee1}}[\text{Wee1}]_{\text{act}}[\text{Cyclin B-Cdk1}] \\
& -k_{\text{wee1,basal}}\left([\text{Wee1}]_{\text{tot}} - [\text{Wee1}]_{\text{act}}\right)[\text{Cyclin B-Cdk1}] \\
& +k_{\text{cdc25}}[\text{Cdc25}]_{\text{act}}[\text{Cyclin B-Cdk1-Yp}] \\
& +k_{\text{cdc25,basal}}\left([\text{Cdc25}]_{\text{tot}} - [\text{Cdc25}]_{\text{act}}\right)[\text{Cyclin B-Cdk1-Yp}]
\end{aligned}
$$

$$
\begin{aligned}
\frac{\partial[\text{Cyclin B-Cdk1-Yp}]}{\partial t} &= D\nabla^2[\text{Cyclin B-Cdk1-Yp}] \\
& +k_{\text{wee1}}[\text{Wee1}]_{\text{act}}[\text{Cyclin B-Cdk1}] \\
& +k_{\text{wee1,basal}}\left([\text{Wee1}]_{\text{tot}} - [\text{Wee1}]_{\text{act}}\right)[\text{Cyclin B-Cdk1}] \\
& -k_{\text{cdc25}}[\text{Cdc25}]_{\text{act}}[\text{Cyclin B-Cdk1-Yp}] \\
& -k_{\text{cdc25,basal}}\left([\text{Cdc25}]_{\text{tot}} - [\text{Cdc25}]_{\text{act}}\right)[\text{Cyclin B-Cdk1-Yp}] \\
& -k_{\text{cak}}[\text{Cyclin B-Cdk1-Yp}] + k_{\text{pp2c}}[\text{Cyclin B-Cdk1-YpTp}] \\
& -k_{\text{dest}}[\text{APC}]_{\text{act}}[\text{Cyclin B-Cdk1-Yp}]
\end{aligned}
$$

$$
\begin{aligned}
\frac{\partial[\text{Cyclin B-Cdk1-YpTp}]}{\partial t} &= D\nabla^2[\text{Cyclin B-Cdk1-YpTp}] \\
& +k_{\text{cak}}[\text{Cyclin B-Cdk1-Yp}] - k_{\text{pp2c}}[\text{Cyclin B-Cdk1-YpTp}] \\
& -k_{\text{cdc25}}[\text{Cdc25}]_{\text{act}}[\text{Cyclin B-Cdk1-YpTp}] \\
& -k_{\text{cdc25,basal}}\left([\text{Cdc25}]_{\text{tot}} - [\text{Cdc25}]_{\text{act}}\right)[\text{Cyclin B-Cdk1-YpTp}] \\
& +k_{\text{wee1}}[\text{Wee1}]_{\text{act}}[\text{Cyclin B-Cdk1-Tp}] \\
& +k_{\text{wee1,basal}}\left([\text{Wee1}]_{\text{tot}} - [\text{Wee1}]_{\text{act}}\right)[\text{Cyclin B-Cdk1-Tp}] \\
& -k_{\text{dest}}[\text{APC}]_{\text{act}}[\text{Cyclin B-Cdk1-YpTp}]
\end{aligned}
$$

$$
\begin{aligned}
\frac{\partial[\text{Cyclin B-Cdk1-Tp}]}{\partial t} &= D\nabla^2[\text{Cyclin B-Cdk1-Tp}] \\
& +k_{\text{cdc25}}[\text{Cdc25}]_{\text{act}}[\text{Cyclin B-Cdk1-YpTp}] \\
& +k_{\text{cdc25,basal}}\left([\text{Cdc25}]_{\text{tot}} - [\text{Cdc25}]_{\text{act}}\right)[\text{Cyclin B-Cdk1-YpTp}] \\
& -k_{\text{wee1}}[\text{Wee1}]_{\text{act}}[\text{Cyclin B-Cdk1-Tp}] \\
& -k_{\text{wee1,basal}}\left([\text{Wee1}]_{\text{tot}} - [\text{Wee1}]_{\text{act}}\right)[\text{Cyclin B-Cdk1-Tp}] \\
& -k_{\text{dest}}[\text{APC}]_{\text{act}}[\text{Cyclin B-Cdk1-Tp}]
\end{aligned}
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\begin{aligned}
\frac{\partial[\text{Cdc25}]_{\text{act}}}{\partial t} &= D\nabla^2[\text{Cdc25}]_{\text{act}} \\
& +k_{\text{cdc25,on}}\left(\frac{[\text{Cyclin B-Cdk1-Tp}]^{n_{\text{cdc25}}}}{(EC50)_{\text{cdc25}}^{n_{\text{cdc25}}} + [\text{Cyclin B-Cdk1-Tp}]^{n_{\text{cdc25}}}}\right) \\
& \times\left([\text{Cdc25}]_{\text{tot}} - [\text{Cdc25}]_{\text{act}}\right) - k_{\text{cdc25,off}}[\text{Cdc25}]_{\text{act}}
\end{aligned}
$$

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\begin{aligned}
\frac{\partial[\text{Wee1}]_{\text{act}}}{\partial t} &= D\nabla^2[\text{Wee1}]_{\text{act}} \\
& -k_{\text{wee1,off}}\left(\frac{[\text{Cyclin B-Cdk1-Tp}]^{n_{\text{wee1}}}}{(EC50)_{\text{wee1}}^{n_{\text{wee1}}} + [\text{Cyclin B-Cdk1-Tp}]^{n_{\text{wee1}}}}\right) \\
& \times[\text{Wee1}]_{\text{act}} + k_{\text{wee1,on}}\left([\text{Wee1}]_{\text{tot}} - [\text{Wee1}]_{\text{act}}\right)
\end{aligned}
$$

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\begin{aligned}
\frac{\partial[\text{Plx1}]_{\text{act}}}{\partial t} &= D\nabla^2[\text{Plx1}]_{\text{act}} \\
&\quad + k_{\text{plx1,on}}\left(\frac{[\text{Cyclin B-Cdk1-Tp}]^{n_{\text{plx1}}}}{(EC50)_{\text{plx1}}^{n_{\text{plx1}}} + [\text{Cyclin B-Cdk1-Tp}]^{n_{\text{plx1}}}}\right) \\
&\quad \times ([\text{Plx1}]_{\text{tot}} - [\text{Plx1}]_{\text{act}}) - k_{\text{plx1,off}}[\text{Plx1}]_{\text{act}} \\
\frac{\partial[\text{APC}]_{\text{act}}}{\partial t} &= D\nabla^2[\text{APC}]_{\text{act}} \\
&\quad + k_{\text{APC,on}}\left(\frac{[\text{Plx1}]_{\text{act}}^{n_{\text{APC}}}}{(EC50)_{\text{plx1}}^{n_{\text{APC}}} + [\text{Plx1}]_{\text{act}}^{n_{\text{APC}}}}\right) \\
&\quad \times ([\text{APC}]_{\text{tot}} - [\text{APC}]_{\text{act}}) - k_{\text{APC,off}}[\text{APC}]_{\text{act}}
\end{aligned}
$$

In our simulations, each of the densities ([Cyclin B], [Cyclin B-Cdk1], [Cyclin B-Cdk1-Yp], [Cyclin B-Cdk1-YpTp], [Cyclin B-Cdk1-Tp], $[\text{Cdc25}]_{\text{act}}$, $[\text{Wee1}]_{\text{act}}$, $[\text{Plx1}]_{\text{act}}$, $[\text{APC}]_{\text{act}}$) are functions of both space and time. For the parameters, all concentrations are in arbitrary units (au), all times are in sec, and all rates are either in $(\text{sec})^{-1}$ or $(\text{au} \times \text{sec})^{-1}$. Except where noted, the parameters are as follows:

$$
\begin{aligned}
k_{\text{synth}} &= 0.02 \\
r &= 10 \\
D &= 0.001 \\
k_{\text{dest}} &= 0.01 \\
k_a &= 0.1 \\
k_d &= 0.001 \\
k_{\text{wee1}} &= 0.05 \\
k_{\text{wee1,basal}} &= k_{\text{wee1}}/r \\
k_{\text{cdc25}} &= 0.1 \\
k_{\text{cdc25,basal}} &= k_{\text{cdc25}}/r \\
[\text{Cyclin}]_{\text{tot}} &= 230 \\
[\text{Cdc25}]_{\text{tot}} &= 15 \\
[\text{Wee1}]_{\text{tot}} &= 15 \\
[\text{APC}]_{\text{tot}} &= 50 \\
[\text{Plx1}]_{\text{tot}} &= 50 \\
n_{\text{wee1}} &= 4 \\
n_{\text{cdc25}} &= 4 \\
n_{\text{APC}} &= 4 \\
n_{\text{plx1}} &= 4 \\
(EC50)_{\text{plx1}} &= 40
\end{aligned}
$$

$$
\begin{aligned}
(EC50)_{\text{wee1}} &= 40 \\
(EC50)_{\text{cdc25}} &= 40 \\
\text{APC} &= 40 \\
k_{\text{cdc25,on}} &= 1.75 \\
k_{\text{cdc25,off}} &= 0.2 \\
k_{\text{APC,on}} &= 1 \\
k_{\text{APC,off}} &= 0.15 \\
k_{\text{plx1,on}} &= 1 \\
k_{\text{plx1,off}} &= 0.15 \\
k_{\text{wee1,on}} &= 0.2 \\
k_{\text{wee1,off}} &= 1.75 \\
k_{\text{cak}} &= 0.8 \\
k_{\text{pp2c}} &= 0.008
\end{aligned}
$$

Besides APC, the cell-cycle proteins have very similar molecular weights (listed below). Treating proteins as spheres, the radius of the protein r scales as $M^{1/3}$, where M is the mass of the protein. From the Stokes-Einstein relation, the diffusion constant (D) in low Reynolds number liquids scales as r^{-1}. Based on the Stokes-Einstein relation, a two-fold increase in the mass of the protein reduces D by 20%, and a five-fold increase in the protein mass reduces D by 40%. Thus, we set all diffusion constants to be equal and within the range of *in vivo* measurements for our simulations. We also note that D can be affected by specific or non-specific *in vivo* interactions.

Table S1: Molecular weights of cell-cycle proteins

Species	MW (kDa)
Cyclin B-Cdk1	79.4
Wee1	67
Cdc25	60.3
Plx1	68.13
Cyclin B	44.92
Cdk1	34.52
APC	300

References

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