Modeling and Evaluating the Cdc2 and Cyclin Interactions in the Cell Divison Cycle with a Time Dependent Petri Net – A Case Study

Louchka Popova-Zeugmann
Department of Computer Science, Humboldt University, Berlin, Germany
popova@informatik.hu-berlin.de

Extended Abstract

In this study we consider a model of the relationship between cdc2 and cyclin in the cell cycle, considered by [Tys91]. It is a "first approximation" of the cell cycle as a hypergraph:

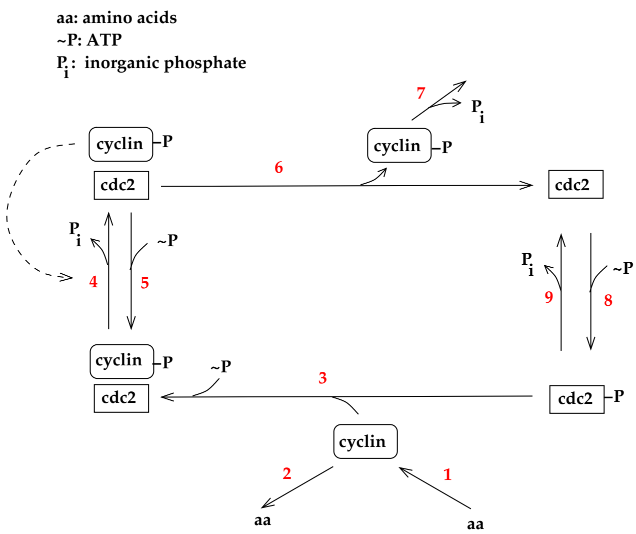

Figure 1: In step 1, cyclin is synthesized *de novo*. Newly synthesized cyclin may be unstable (step 2). Cyclin combines with cdc2-P (step 3) to form pre-maturation promoting factor (preMPF). At some point after heterodimer formation, the cyclin subunit is phosphorylated. ... The cdc2 subunit is then dephosphorylated (step 4) to form active MPF. In principle, the activation of MPF may be opposed by protein kinase (step 5). Assuming that active MPF enhances the catalytic activity of the phosphatase, I arrange that MPF activation is switched on in an autocatalytic fashion. Nuclear division is triggered when a sufficient quantity of MPF has been activated, but concurrently active MPF is destroyed by step 6. Breakdown of the MPF complex releases phosphorylated cyclin, which is subject to rapid proteolysis (step 7). Finally, the cdc2 subunit is phosphorylated (step 8, possibly reversed by step 9), and the cycle repeats itself. (Tyson, J.J.)

In Fig. 3 we describe the model, given by Tyson as a hypergraph, as a DI-PN. The table in Fig. 2 shows the derivation of the minimal and the maximal durations for each transition for the initial p-marking m_0 with: $m_0(aa) = 12$, $m_0(C2) = 12$, and the remaining places do not contain tokens in m_0. This initial marking was chosen so that the skeleton is live and bounded.

t_i	k_i	min_rate	max_rate	min_dur.	max_dur.	[⌈min_dur.⌉, ⌈max_dur.⌉]
r_1	0.015	0.015	0.18	$\frac{100}{18}$	$\frac{200}{3}$	$[6, 67]$
r_3	200	200	28800	$\frac{1}{28800}$	$\frac{1}{200}$	$[0, 0]$[1]
r_4	10	$\frac{5}{18}$	$\frac{2560}{144}$	$\frac{144}{2560}$	$\frac{18}{5}$	$[0, 4]$[2]
r'_4	0.018	$\frac{9}{500}$	$\frac{27}{125}$	$\frac{125}{27}$	$\frac{500}{9}$	$[5, 56]$
r_6	0.1	0.1	1.2	$\frac{5}{6}$	10	$[1, 10]$
r_7	0.6	0.6	7.2	$\frac{5}{36}$	$\frac{5}{3}$	$[1, 2]$
r_8	10	10	1200	$\frac{1}{120}$	$\frac{1}{10}$	$[0^1, 1]$
r_9	0.1	0.1	1.2	$\frac{5}{6}$	10	$[1, 10]$

Figure 2: Table: Evaluation of the lower and the upper bound for the duration of each transition with the initial p-marking m_0 with: $m_0(aa) = 12$, $m_0(C2) = 12$ in the DI-PN given in Fig. 3

Why we did chose this initial place-marking? In order to keep the net live adding time each transition has to be live. Transition r_4 can fire if the place M contains at least 2 tokens. For that it is necessarily that holds:

$$2 \cdot min\_dur.(r'_4) \leq max\_dur.(r_6).$$

From this it follows that

$$2 \cdot \big(1/max\_rate(r'_4)\big) \leq 1/min\_rate(r_6)$$

. Hence, it have to be true

$$2 \cdot min\_rate(r_6) \leq max\_rate(r'_4).$$

Consequently, the minimal number of tokens $[M]_{min}$ of the place M and the maximal number of tokens $[pM]_{max}$ of place pM have to fulfill the inequation

$$2 \cdot k_6 \cdot [M]_{min} \leq k'_4[pM]_{max},$$

i.e.

$$2 \cdot 0.1 \cdot 1 \leq 0.018 \cdot [pM]_{max}.$$

At least, we obtain $[pM]_{max} \leq 11.111$, i.e. if $[pM]_{max} \geq 12$ then the transition r_4 cannot fire and the net is not live. Thus, an initial marking m_0 with $m_0(aa) = 12$, $m_0(C2) = 12$ is a minimal one so that the derived DI-PN is also live.

Furthermore, there are two P-invariants, covering the skeleton. Thus the skeleton is bounded. The total token sum of both P-invariants is 12 ; and we consider all places to be 12-bounded.

According to the transformation rule, introduced in [Pop07] we transform the Duration-Interval-Petri net (short: DI-PN) into a Time Petri net (short: TPN). This TPN can be reduced. (1) The transitions r_2 and r_5 can be removed, they will never fire. (2) The transformation can be simplified for the transitions r_1, r_7 and r_8, because none of them is involved in a conflict, and they all have minimal duration greater than zero (for more cf. [Pop07]). (3) Finally, the transformation for t_3 can be simplified because the maximal bound for its duration in the D-TPN is zero. Thus, we obtain the TPN given in Fig. 4, which models the cell division cycle described in [Tys91].

The skeleton of this net is bounded (the state space contains 101,840 markings) and live. Hence, the TPN is also bounded. However, the state space contains more than 20 millions essential states.

[1]These values are very small in relation to the rest. Therefore they are rounded to zero.

[2]These values are obtained using the mass-action equation, given in [Tys91]: $F([M]) = k'_4 + k_4 \cdot ([M]/[CT])^2$, where $[CT] = [pM] + [M] + [C2] + [CP]$ (a P-invariance).Than the rate equation is: $k_4 \cdot [pM]([M]/[CT])^2$. Please, notice that the notation $[X]$ used by Tyson means a p-marking of a place X. We consider two p-markings – one with minimal number of tokens and one with maximal number such that the transition is enabled. These are $[pM] = 1$, $[M] = 2$ and $[CT] = 12$ in the minimal p-marking and $[pM] = 4$, $[M] = 8$ and $[CT] = 12$ in the maximal p-marking.

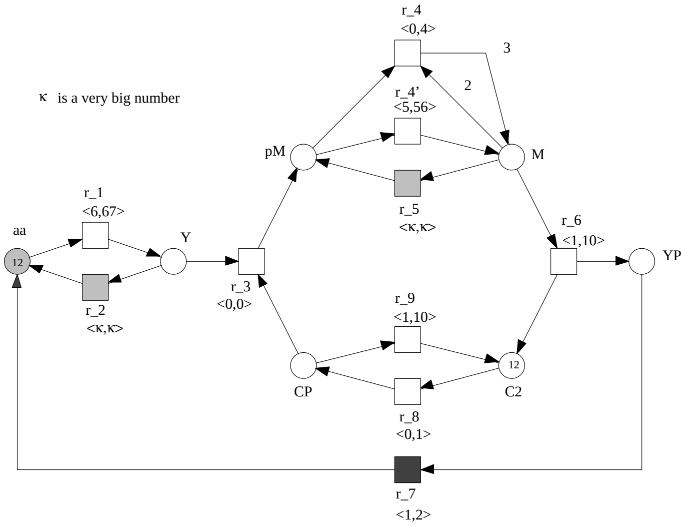

Figure 3: Modeling the cell division cycle: cdc2 and cyclin interactions (Tyson, 1991, 6 variables). $r_1 : aa \rightarrow Y, r_2 : Y \rightarrow aa, r_3 : CP + Y \rightarrow pM, r_4' : pM \rightarrow M, r_4 : pM + 2M \rightarrow 3M, r_5 : M \rightarrow pM, r_6 : M \rightarrow C2 + YP, r_7 : C2 \rightarrow CP, r_8 : CP \rightarrow C2, r_9 : YP \rightarrow aa$. The rates are: $k_1 = 0.015, k_2 = 0, k_3 = 200, k_4 = 10 - 1000$ (adjustable), $k_4' = 0.018, k_5 = 0, k_6 = 0.1 - 10$ (adjustable), $k_7 = 0.6, k_8 >> k_9, k_9 >> k_6$. In this Petri net model $k_4 >> k_4', k_4$ models autocatalysis.

In order to have a TPN with a smaller state space we use a new initial p-marking: m_0 with $m_0(aa) = 4$, $m_0(C2) = 4$ and we modify some intervals slightly:

	r_1	r_3	r_4	r_4'	r_6	r_7	r_8	r_9
[min_dur. , max_dur.]	$[17, 67]$	$[0, 0]$	$[2, 30]$	$[19, 56]$	$[34, 60]$	$[1, 2]$	$[1, 1]$	$[4, 10]$

The skeleton of this net is bounded (the state space contains 477 markings) and live. The TPN is also bounded and live and the state space contains 1,053,509 essential states and 8,571,845 arcs although there are 303 p-markings [3] only. In the following we call this TPN Tyson net for short.

The minimal time for starting the dephosphorylation of the cdc2 in order to form active MPF modelled with r_4 is not less than 74 minutes.

Please note, that decreasing the number of tokens in the place $C2$ to 3 in the initial state leads to a difference in the liveness behavior of the net and its skeleton: the skeleton is live, but the net has a dead transition.

Please note, our time models provide also immediate transitions, although in reality nothing happens without consuming time. However, immediate transitions help to keep interval boundaries small, as we have just seen. Often, system activities may be classified into activities with significant time consumption and those with non-significant (much less) time consumption. Without immediate transitions, such a difference had to be modelled by an appropriate absolute difference of time values. With immediate transitions, all time values can be scaled down relatively to a suitable time axis. Moreover, immediate transitions allow a straightforward incorporation of the working time concept.

Finally, we would like to mention another approach to obtain a time-dependent Petri net from a timeless one which is a model of a considered biochemical network. It based on the relationship between steady states in the network and the T-invariants in the model. However, the derived time constraints are relative time values, and thus not suitable for the computation of absolute time bounds as considered in this paper. For more see [Pop05].

[3] The reachability graph was computed with INA (cf. [Sta03])

Using the transformation introduced in [Pop07] we obtain the following TPN:

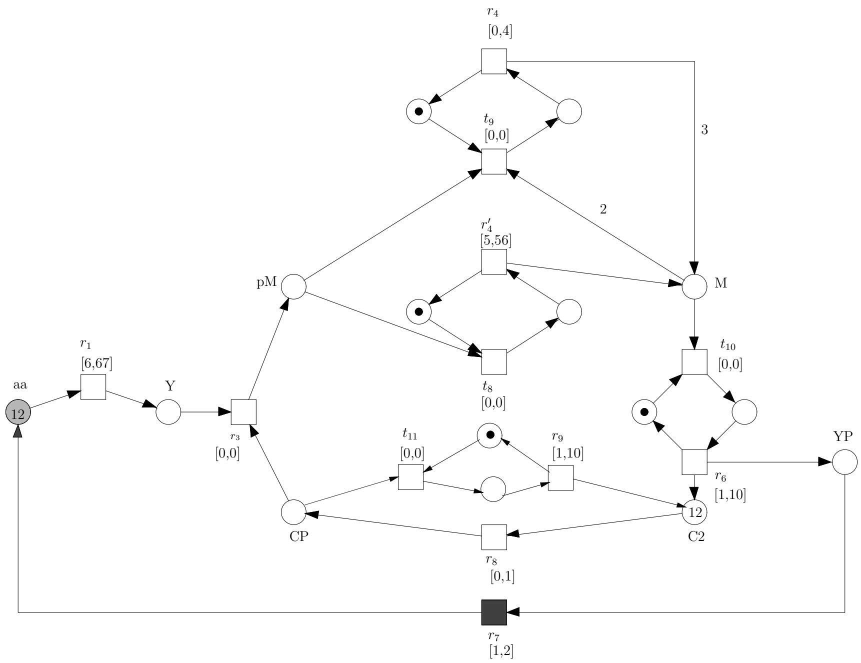

Figure 4: TPN-model for the cell division cycle: cdc2 and cyclin interactions. The interval bounds (minimal and maximal durations) are rounded up.

References

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