

Logical model specification assisted with model checking: application to mammalian cell cycle

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Supplementary tables

Table S1. Comparison of the results of perturbation simulations for the original model of Fauré et al (2006) and the revised version (cf. Figure 1 and Table 1). All perturbations are encoded in the model available in the repository associated with GINsim.

Genetic perturbation	Phenotype	Original model	Revised model
Ectopic CycE Henley et al., 2012	Viable; cycle in absence of growth factor; less serum-dependent	For CycD=0, cycle or steady state (questionable)	Cycle for CycD=1, steady state for CycD=0 (questionable)
Ectopic p27 Rivard et al., 1996; Ji et al., 2006	Cell cycle arrest in presence of growth factors	OK	OK
Ectopic p27 and CycA Rivard et al., 1996; Rape et al., 2004	Cell cycle arrest in presence of growth factors	OK	OK
Ectopic p27 and CycE Rivard et al., 1996	Cell cycle arrest in presence of growth factors	OK	OK
Ectopic p27 and E2F Rivard et al., 1996	Cell cycle arrest in presence of growth factors	OK	OK
Ectopic Rb (maximum level) Ji et al., 2004; Dick & Rubin, 2013	G1 arrest + p27 accumulation	OK	OK
Cdh1 KO Yuan et al., 2014	Viable cell cycle in presence or absence of growth factors.	OK	OK
Rb KO, Loss of Rb Wong et al., 2014	Viable cell cycle in presence or absence of growth factors	OK	OK
E2F over-expression in Rb-/ E2F E1 Rb KO Ji et al., 2004	Viable cell cycle in presence or absence of growth factors	OK	OK
E2F over-expression in Rb+/ E2F E1 Rb E1 CycE [Rb@0] CycA [Rb@0] Ji et al., 2004	Targets of E2F are expressed even in absence of growth factor but the cell cycle is arrested	Cyclic attractor with CycD=0 or CycD=1 Disagreement	OK
CycE KO Geng et al., 2003; Ohtsubo et al., 1995	Cell cycle arrest before S phase	Disagreement	OK
CycE KO CycA KO Dai et al., 2013	Cell cycle arrest before S phase	Disagreement	OK
UbcH10 KO Townsend et al., 1997	Cell cycle arrested in M phase, destruction of cyclin A and B inhibited.	Disagreement	OK
Rb induction (early kinetics) Rb E1 E2F [Rb@0] CycE [Rb@0] CycA [Rb@0] Ji et al., 2004	Cell cycle arrest	Cyclic attractor with CycD=0 or CycD=1 Disagreement	OK
RbR661W E2F [Rb@0] CycE [Rb@0] CycA [Rb@0] Ji & Zhu, 2005	Cell cycle in presence of growth factors, cell cycle arrest in their absence	Cyclic attractor with CycD=0 or CycD=1 Disagreement	OK
Skp2 KO Kotshiba et al., 2014	Cell cycle arrest or endoreplication with Cyclin E and p27 accumulation	Skp2 absent from the model	OK
Skp2 KO p27 KO Kotshiba et al., 2014	Cell cycle in presence of growth factors, cycle arrest in their absence	Skp2 absent from the model	OK
Skp2 over-expression and Rb induction (early kinetics) Skp2 E1 Rb E2 E2F [Rb@0] CycE [Rb@0] CycA [Rb@0] Ji et al., 2004; Dick & Rubin, 2013	Viable cell cycle in presence or absence of growth factors	Skp2 absent from the model	OK
p27 KO (early kinetics) Rb E1 p27 KO E2F [Rb@0] CycE [Rb@0] CycA [Rb@0] Ji et al., 2004	Viable cell cycle in presence or absence of growth factors	OK	OK
p27 KO (cell dependent) Rivard et al., 1996; Kotshiba et al., 2014	Cycle in absence of growth factors	Disagreement	Disagreement

Table S2. Verification of dynamical biological properties with model-checking. The initial state G1 is represented by CycE=0 & CycA=0 & CycB=0 & Cdh1=1 in the initial model, and by CycE=0 & CycA=0 & CycB=0 & Cdh1=1 & Rb=1 & p27=1 in the updated model. The initial state G2 is represented by Rb=0 & Cdh1=0 & p27=0 & CycE=0.

Biological property	Sequences of events verified by model-checking	Original model	Updated model
Observed sequence for cyclins only	1. Correct sequence starting from G1: CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=1 -> CycE=0 & CycA=0 & CycB=1 -> CycE=0 & CycA=0 & CycB=0	OK	OK
Observed sequence for all species	2. Correct sequence defined in Figure 2 (with p27, Rb and Skp2 OFF) and Figure 3.	OK but with p27, Rb and Skp2 OFF	OK
Irreversibility of CycE inactivation	3. Incorrect sequence starting from G1: CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=0 -> CycE=1 & CycA=1 & CycB=0	OK	OK
Irreversibility of CycA inactivation	4. Incorrect sequence starting from G1: CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=1 -> CycE=0 & CycA=0 & CycB=1 -> CycE=0 & CycA=1 & CycB=1	OK	OK
Irreversibility of CycB inactivation	5. Incorrect sequence starting from G1: CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=1 -> CycE=0 & CycA=0 & CycB=1 -> CycE=0 & CycA=0 & CycB=0 -> CycE=0 & CycA=0 & CycB=1	No	No
Sequential activation of CycE and CycA	6. Incorrect sequence starting from G1 (activation of CycA before CycE): CycE=0 & CycA=0 -> CycE=0 & CycA=1 -> CycE=1 & CycA=1	No	OK
Degradation of CycE in S phase, activation of CycB in G2 phase	7. Incorrect sequence starting from G1 (CycB activation before CycE inactivation): CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=1 & CycA=1 & CycB=1 -> CycE=0 & CycA=1 & CycB=1 -> CycE=0 & CycA=0 & CycB=1 -> CycE=0 & CycA=0 & CycB=0	No	No
Synthesis of CycA in early S phase, activation of CycB in G2 phase	8. Incorrect sequence starting from G1 (CycB activation before CycA activation): CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=1 -> CycE=1 & CycA=1 & CycB=1	OK	OK
Degradation of CycA in early mitosis, degradation of CycB in late mitosis	9. Incorrect sequence starting from G1 (CycB inactivation before CycA inactivation): CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=1 -> CycE=0 & CycA=1 & CycB=0 -> CycE=0 & CycA=0 & CycB=0	No	No
Complete removal of CycB is essential to prevent spindle checkpoint after sister chromatid disjunction	10. Incorrect sequence starting from G1 (CycE activation before CycB inactivation): CycE=0 & CycA=0 & CycB=0 -> CycE=1 & CycA=0 & CycB=0 -> CycE=1 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=0 -> CycE=0 & CycA=1 & CycB=1 -> CycE=0 & CycA=0 & CycB=1 -> CycE=1 & CycA=0 & CycB=1	OK	OK
Feedforward loop that ensures that cells advance to S-phase irreversibly: phosphorylation of Rb by CycE to release E2F and increase the synthesis of CycE, along with the phosphorylation of p27	11. Correct sequence starting from G1: CycE=0 & CycA=0 & p27=1 -> CycE=1 & CycA=0 & p27=1 -> CycE=1 & CycA=0 & p27=0 -> CycE=1 & CycA=1 & p27=0 -> CycE=0 & CycA=1 & p27=0	No	OK
	12. Incorrect sequence starting from G1 (p27 inhibited without CycE): CycE=0 & CycA=0 & p27=1 -> CycE=0 & CycA=0 & p27=0 -> CycE=1 & CycA=0 & p27=0 -> CycE=1 & CycA=1 & p27=0 -> CycE=0 & CycA=1 & p27=0	No	OK
	13. Incorrect sequence starting from G1 (CycA activated before p27 inhibition): CycE=0 & CycA=0 & p27=1 -> CycE=1 & CycA=0 & p27=1 -> CycE=1 & CycA=1 & p27=1	No	OK
Degradation of B cyclins by the APC/C complex is required for mitosis to take place	14. Correct sequence from G2: CycA=1 & CycB=0 & Cdc20=0 -> CycA=1 & CycB=1 & Cdc20=0 -> CycA=1 & CycB=1 & Cdc20=1 -> CycA=0 & CycB=1 & Cdc20=1 -> CycA=0 & CycB=0 & Cdc20=1 -> CycA=0 & CycB=0 & Cdc20=0	OK	OK
	15. Incorrect sequence from G2 (inactivation of CycB without Cdc20): CycA=1 & CycB=0 & Cdc20=0 -> CycA=1 & CycB=1 & Cdc20=0 -> CycA=1 & CycB=1 & Cdc20=1 -> CycA=0 & CycB=1 & Cdc20=1 -> CycA=0 & CycB=1 & Cdc20=0 -> CycA=0 & CycB=0 & Cdc20=0		
	16. Incorrect sequence from G2 (inactivation of CycA without Cdc20): CycA=1 & CycB=0 & Cdc20=0 -> CycA=1 & CycB=1 & Cdc20=0 -> CycA=0 & CycB=1 & Cdc20=0 -> CycA=0 & CycB=0 & Cdc20=0		
Cdh1 is not required for mitotic exit	17. Correct sequence from G2 (mitosis exit without Cdh1): CycA=1 & CycB=0 & Cdc20=0 & Cdh1=0 -> CycA=1 & CycB=1 & Cdc20=0 & Cdh1=0 -> CycA=1 & CycB=1 & Cdc20=1 & Cdh1=0 -> CycA=0 & CycB=1 & Cdc20=1 & Cdh1=0 -> CycA=0 & CycB=0 & Cdc20=1 & Cdh1=0 -> CycA=0 & CycB=0 & Cdc20=0 & Cdh1=0	OK	OK
UbcH10 is required for the destruction of mitotic cyclins and other mitosis-related substrates	18. Incorrect sequence from G2 (mitosis exit without UbcH10): CycA=1 & CycB=0 & Cdc20=0 & UbcH10=0 -> CycA=1 & CycB=1 & Cdc20=0 & UbcH10=0 -> CycA=1 & CycB=1 & Cdc20=1 & UbcH10=0 -> CycA=0 & CycB=1 & Cdc20=1 & UbcH10=0 -> CycA=0 & CycB=0 & Cdc20=1 & UbcH10=0 -> CycA=0 & CycB=0 & Cdc20=0 & UbcH10=0	No	OK