Logical modelling in biology through the case study of the cell cycle and its checkpoints

Déborah Boyenval iBV seminar

March 24, 2022

Logical modeling framework

Discrete modeling of the cell cycle itsel

Discrete modeling of cell cycle checkpoints

Conclusion

Pluridisciplinary current research project



- PhD student in bioinformatics
- Initial education in biology



institut Valrose **B** i o l o g i e

Team SPARKS (Gilles Bernot and Jean-Paul Comet) Team Franck Delaunay

Understanding the interactions between biological oscillating systems

Discrete modeling of the cell cycle itsel

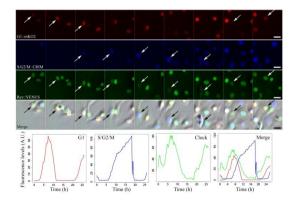
Discrete modeling of cell cycle checkpoints

Conclusion

The study of oscillating processes

Biological knowledge from experiments

- Observation of biological oscillations (circadian clock and cell cycle time series)
- Highlighting the mechanisms underlying oscillations (gain/loss of function)



Feillet et al. 2015 Frontier in Neurology

- Red: G1 phase marker
- Blue: S/G2 phase marker
- Green: Clock marker

 Discrete modeling of the cell cycle itsel

Discrete modeling of cell cycle checkpoints

Conclusion 0000

The study of oscillating processes

Logical modeling framework of biological regulatory network

- Formalizes the interactions between biological species using logical formulas
- Is an oscillatory behavior compatible with these formalized interactions?

Logical modeling framework

Discrete modeling of the cell cycle itsel

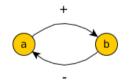
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The study of oscillating processes

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Logical modeling framework

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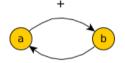
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The study of oscillating processes

Logical modeling framework of biological regulatory network

- Formalizes the interactions between biological species using logical formulas
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 $a = 0 \Leftrightarrow a$ is inactive (the same for b) $a = 1 \Leftrightarrow a$ activates $b, b = 1 \Leftrightarrow b$ inhibits a



 $a \ge 1 \Leftrightarrow a$ is a resource of b $\neg(b \ge 1) \Leftrightarrow b$ is a resource of a

A resource is the presence of an activator or the absence of an inhibitor.

Logical modeling framework

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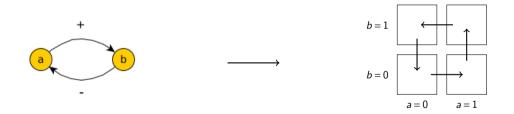
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The study of oscillating processes

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• $K_{\nu,\omega}$ formalizes the influence of all the regulators of a variable on its activity

Logical modeling framework

Discrete modeling of the cell cycle itsel

Discrete modeling of cell cycle checkpoint

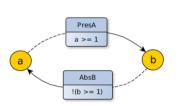
(3)

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Logical modeling framework of biological regulatory network

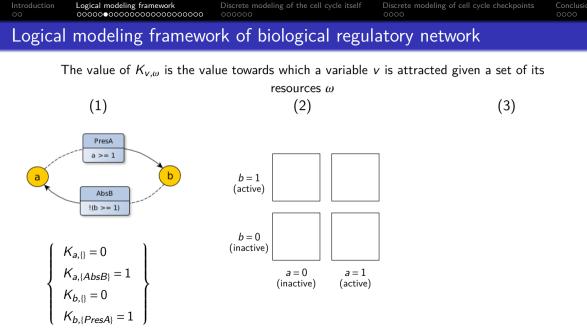
The value of $K_{v,\omega}$ is the value towards which a variable v is attracted given a set of its

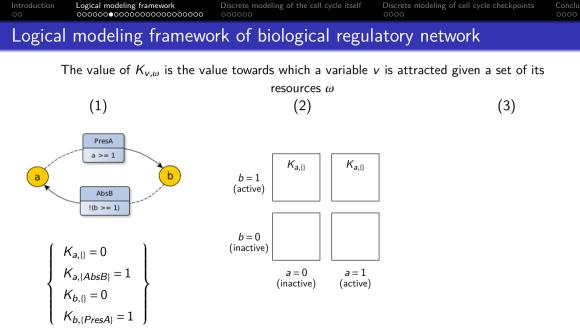
resources ω (2)

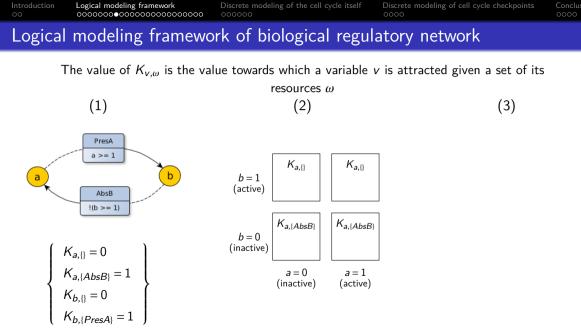


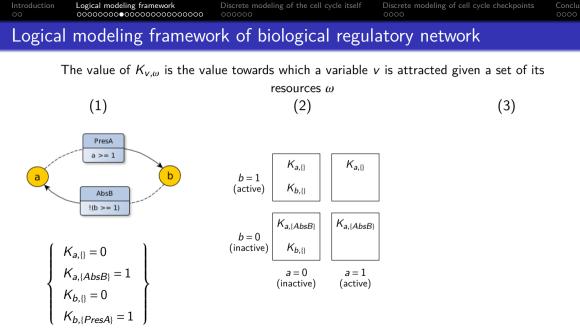
(1)

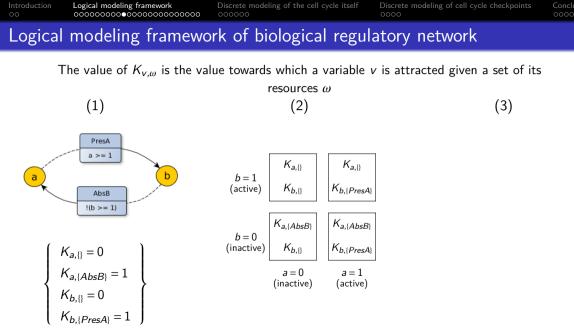
$$\left\{ \begin{array}{l} \mathcal{K}_{a,\{\}} = 0 \\ \mathcal{K}_{a,\{AbsB\}} = 1 \\ \mathcal{K}_{b,\{\}} = 0 \\ \mathcal{K}_{b,\{PresA\}} = 1 \end{array} \right\}$$

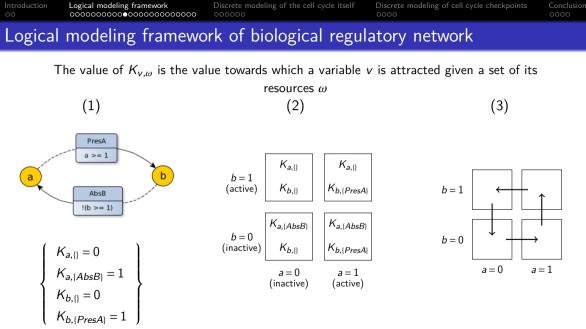


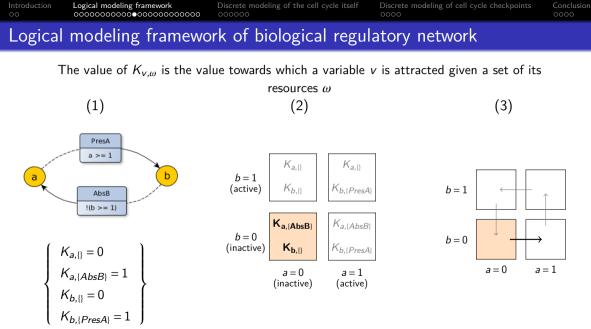


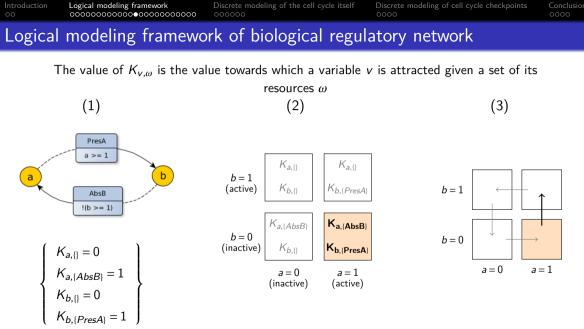


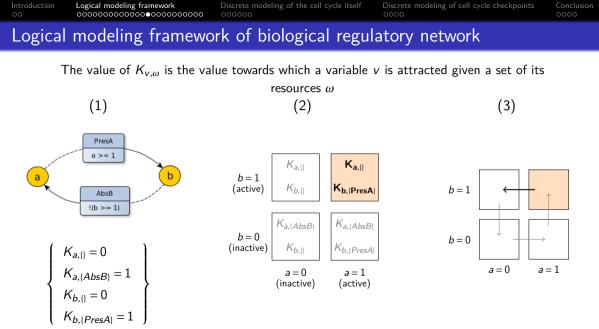


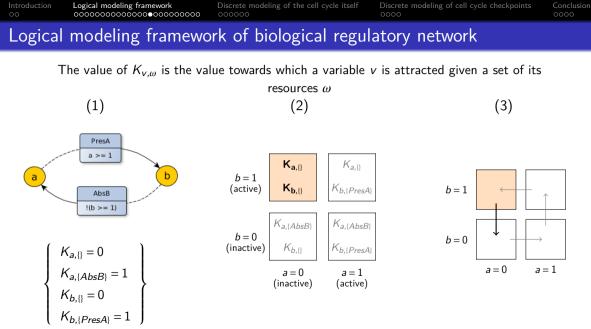












Logical modeling framework

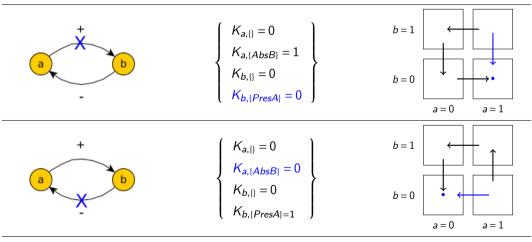
Discrete modeling of the cell cycle itse

Discrete modeling of cell cycle checkpoints

Conclusion

What is a logical model of a biological system?





These two models with a non-functional interaction (crossed edge) \Longrightarrow No oscillation

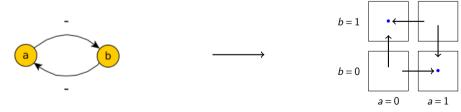
What are the necessary conditions to observe certain behaviors?

What are the models that produce oscillations?

- Odd number of inhibiting edges (i.e. a functional negative cycle)
- There is a mathematical proof! So it is generalized to any model!

What are the models that produce multiple steady states?

• Even number of inhibiting edges (*i.e.* a functional positive cycle)



This interaction graph will never produce oscillations, but can produce bistability !

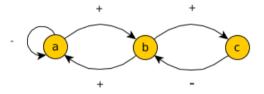
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Logical reasoning on higher dimensional model



Let's try to answer these questions:

- Does this biological interaction network produce oscillations?
- What are the key interactions to obtain an oscillatory behaviour?

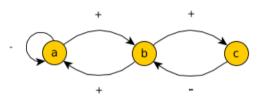
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Logical reasoning on higher dimensional model



Let's try to answer these questions:

- Does this biological interaction network produce oscillations?
- What are the key interactions to obtain an oscillatory behaviour?

This is clearly non trivial ! It depends on the functionality of negative cycles i.e. the parameterizations

Logical modeling framework

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Logical reasoning on higher dimensional model

Automation of the logical reasonning (René Thomas' approach)

• Application of the logical modeling formalism manually: certainly not!

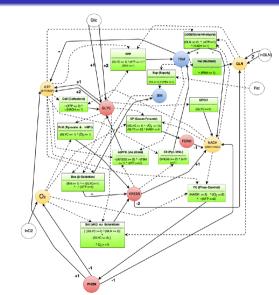
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Discrete modeling of the cell cycle itself

Discrete modeling of cell cycle checkpoints $_{\rm OOOO}$

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A very complex interaction graph of the energy metabolism!



- 14 variables and 100 parameters
- More parametrizations than stars in the universe
- Khoodeeram et al. 2017 Advances in Systems and Synthetic Biology

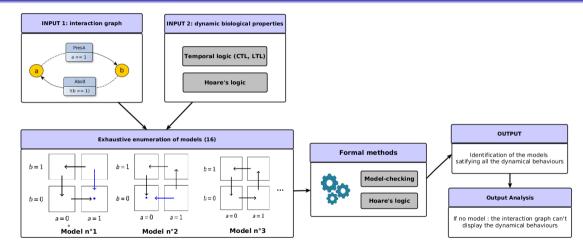
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Learning of parameterizations verifying given biological behaviors



The TotemBioNet tool Boyenval *et al.* 2020 - CMSB

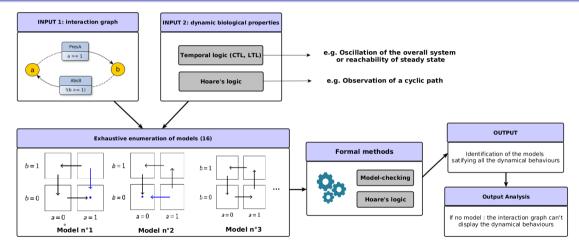
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Learning parameterizations verifying given biological behaviors



The TotemBioNet tool Boyenval *et al.* 2020 - CMSB Discrete modeling of the cell cycle itsel

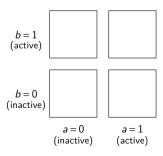
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Logical reasoning on higher dimensional model

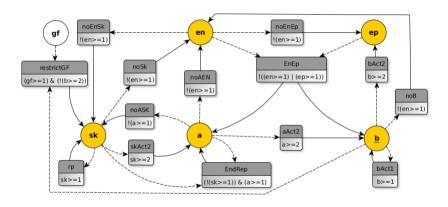
Qualitative dynamics (René Thomas' approach)

- Concentration space discretization
- Opens the door to formal methods in theoretical computer science
- No blind spot: all parameterizations (interaction graph dynamics) are calculated
- Unlike ODEs system for example



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The cell cycle study case



- *sk* (*starting kinases*) : CycE/Cdk2
- *a* : CycA/Cdk1 et CycA/Cdk2
- **b** : CycB/Cdk1
- en (enemies) : APC-cdh1, Wee1, p21, p27, PP1 et PP2
- *ep* (*exit protein*) : APC-cdc20
- gf (growth factors) : EGF, FGF, PDGF, ...

Interaction graph of the mammalian cell cycle progression (revised from Behaegel *et al.* 2015)

The cell cycle itself: Observation of a canonical cyclic path (Hoare's logic)

• {EARLYG1}

$$\frac{sk+;en-;sk+;a+;sk-;sk-;a+;en+;b+;en-;b+;ep+;a-;a-;b-;ep-;en+}{G1}$$

$$\frac{Sk+;en-;sk+;a+;sk-;sk-;a+;en+;b+;en-;b+;ep+;a-;a-;b-;ep-;en+}{M}$$

$$\frac{Sk+;en-;sk+;a+;sk-;sk-;a+;en+;b+;en-;b+;ep+;a-;a-;b-;ep-;en+}{M}$$

• EARLYG1 (one initial state) specified by $sk = 0 \land a = 0 \land b = 0 \land ep = 0 \land en = 1 \land gf = 1$

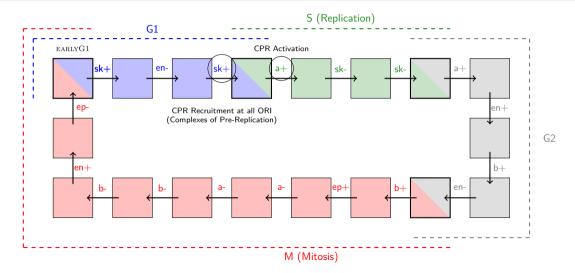
• Never far from biological data: significant bibliographical work (variable activities)

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The cell cycle itself: Observation of a canonical cyclic path



cycle checkpoints Conclu 0000

What are the models that verify the major properties of the cell cycle?

The quiescence phase blocking in absence of growth factors (Temporal logics - CTL)

- G0 (another initial state) specified by $sk = 0 \land a = 0 \land b = 0 \land ep = 0 \land en = 1 \land gf = 0$
- $AG(gf = 0) \Longrightarrow AG(G0)$

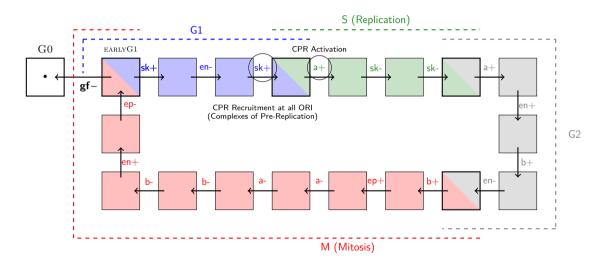
Logical modeling framework

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Discrete modeling of cell cycle checkpoints

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The quiescence phase blocking in absence of growth factors



What are the models that verify the major properties of the cell cycle?

Results using our tool TotemBioNet

- $\bullet\,$ Among the 7×10^{18} possible models, only 32 satisfy the cell cycle major properties.
- The canonical cell cycle (oscillation) and the quiescence steady state
- Mathematically proven by deduction

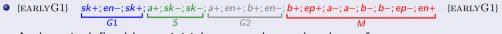
"It is through deduction that we descend from the general principle (axioms) to the particular case (theorem), without any additional biological experience"

Claude Bernard - Principes de médecine expérimentale

Discrete modeling of cell cycle checkpoints •000 Conclusion

Checkpoints are intrisically discrete mechanisms





• A phase is defined by an initial state and *unordered* set of events.

Discrete definition of cell cycle checkpoints

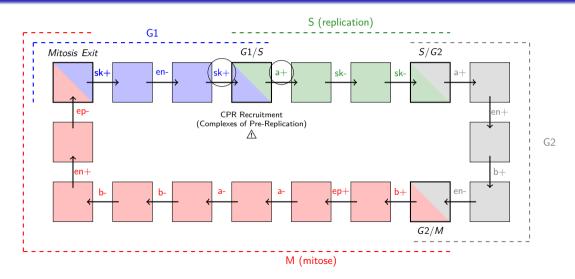
A completion requirement of events of a given **phase** before the onset of the first event of next **phase**. If each checkpoint is satisfied then the cell ensures the transmission of intact DNA to its daughter cell.

Hypothesis

- A checkpoint does not work on the basis of a regulation delay but on the basis of a chronology of events
- No chronometric modeling study (EDO systems or hybrid formalisms)

Discrete modeling of cell cycle checkpoints 0000

Checkpoints are intrisically discrete mechanisms



Discrete modeling of the cell cycle itsel

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Conclusion

Checkpoint formalization in 1st order logic

The generic predicate checkpoint

$$\underline{\exists \sigma} \mid \underline{\forall \pi_i} \in [G1, S, G2, M], \quad \forall X, \forall Y \text{ (two events of the form } v + | v -), \\ \mathbf{canEnd}_{\sigma}(X, \pi_i) \land \mathbf{canStart}_{\sigma}(Y, \pi_{i+1}) \Longrightarrow \mathbf{isRequired}_{\sigma}(X, Y)$$

The predicates *canEnd* and *canStart*

 $canEnd_{\sigma}(X, \pi_i) \iff \exists p' \in permutations(p) \mid (\sigma(wp(\{init\}p'\{final\})) \land X = last(p'))$

 $canStart_{\sigma}(\mathbf{Y}, \pi_i) \Longleftrightarrow \exists \ p' \in permutations(p) \mid (\sigma(wp(\{init\}p'\{final\})) \land \mathbf{Y} = first(p'))$

The predicate *isRequired*

•
$$X = x + or x - , Y = y + or y -$$

• $isRequired(X, Y) \iff (\sigma(K_{y, \omega_{before}}) - \eta_{before}) \times (\sigma(K_{y, \omega_{after}}) - \eta_{after}) \le 0$

What are the models that verify the 4 cell cycle checkpoints?

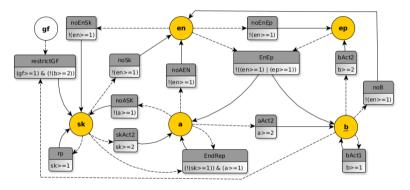
Results after implementation in Prolog

- Conclusion: 3 of the 4 modelled checkpoints are satisfied.
- Not the mitosis exit checkpoint !
- Identification of a biological counter-example ... of the generic definition
- Towards a new definition of the mitosis exit checkpoint (work in progress)

Checkpoint Eval		Output (<i>models</i> = <i>checkpoint</i>)	Computation time	
G1/S	True	24/32	1.9s	
S/G2	True	32/32	11s	
G2/M	True	32/32	1h10min	
Mitosis Exit	False	0/32	1h12min	

Introduction	Logical modeling framework	Discrete modeling of the cell cycle itself	Discrete modeling of cell cycle checkpoints	Conclusion
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Modelling conclusions



- The modeled interactions are consistent with the cell cycle and 3 of its checkpoints.
- Spoil: after revision of the definition, the mitosis exit checkpoint is also verified.

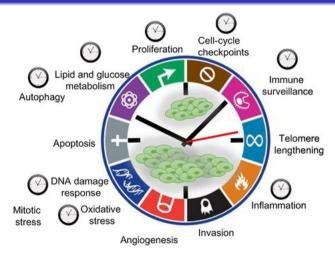
Logical modeling framework

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Cell cycle checkpoints and integration of oscillating processes



- Cellular processes are intrinsically interdependant
- Circadian oscillator is linked to many cellular processes
- Oscillators coupling understanding
- Anticancer chronotherapy

Sulli et al. 2019 - Trends Cancer

At the interface between biology, mathematics and computer science

- Biology : study of the complexity of life through experimentations (abductive science)
- Mathematics : formalization of knowledge on which rules of reasoning are applied (*e.g.* deductive science)
- Computer science : automation of mathematical calculations (the human being is the worst calculator)

Towards theoretical biology?

Our logical approach: encoding biological knowledge in a mathematical language to automatically deduce conclusions for designing new biological experiments.

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Acknowledgements



- Thank you for listening !
- All the member of my two teams (iBV and I3S)
- A special thanks to Hélène Collavizza (TotemBioNet tool and mathematical formalization)