BIOCHEMICALLY INTERPRETED PETRI NETS

- SOME OPEN PROBLEMS -

Monika Heiner

Brandenburg University of Technology Cottbus

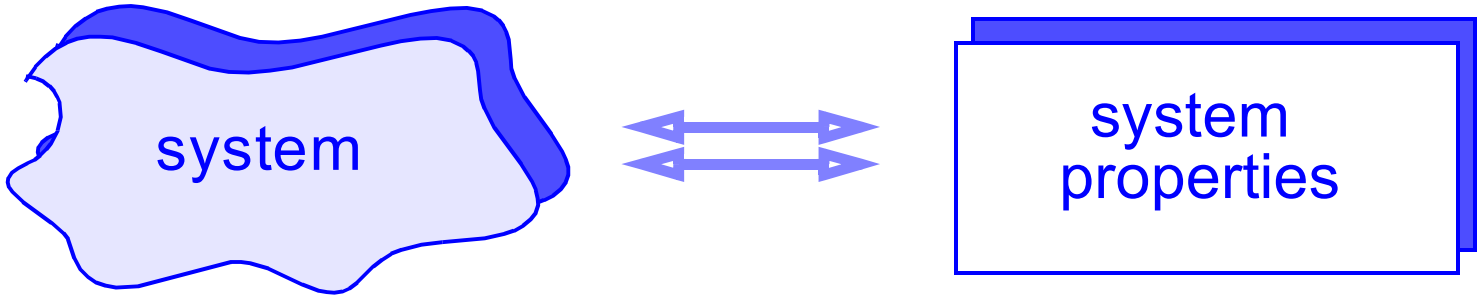
Dept. of CS

OUTLINE

- **BACKGROUND**
- **HOW STRUCTURAL ANALYSIS TECHNIQUES CAN CONTRIBUTE**
  - -> modularization approach
    connected ADT sets = flow equivalent server component ?
  - -> *identify core network*
- **LATEST NEWS**
  - -> *our tool box: Snoopy, Charlie, Marcie*
  - -> *colored framework*
  - -> *Generalized Hybrid Petri Nets*

BACKGROUND

MODEL- BASED SYSTEM ANALYSIS



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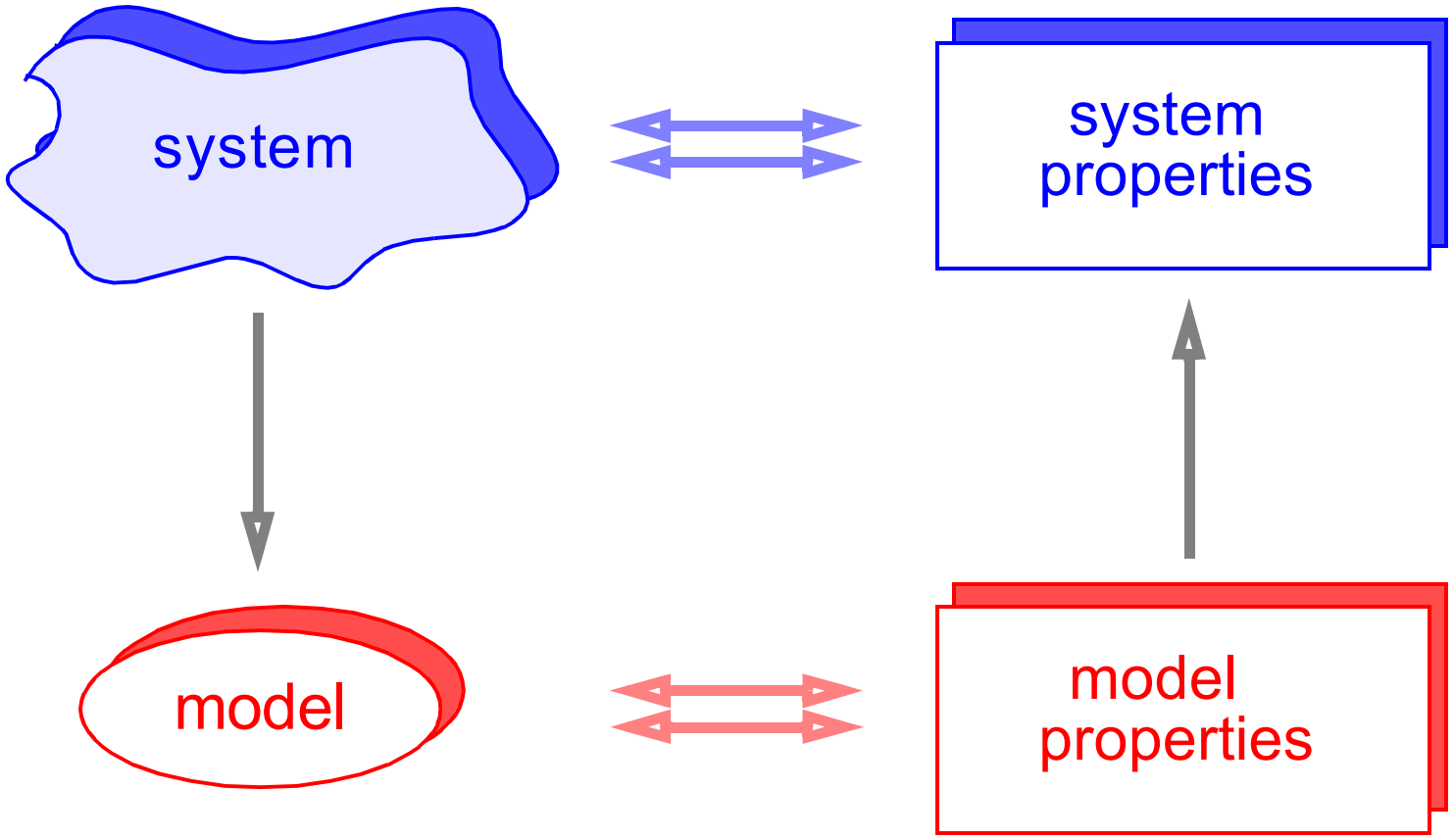

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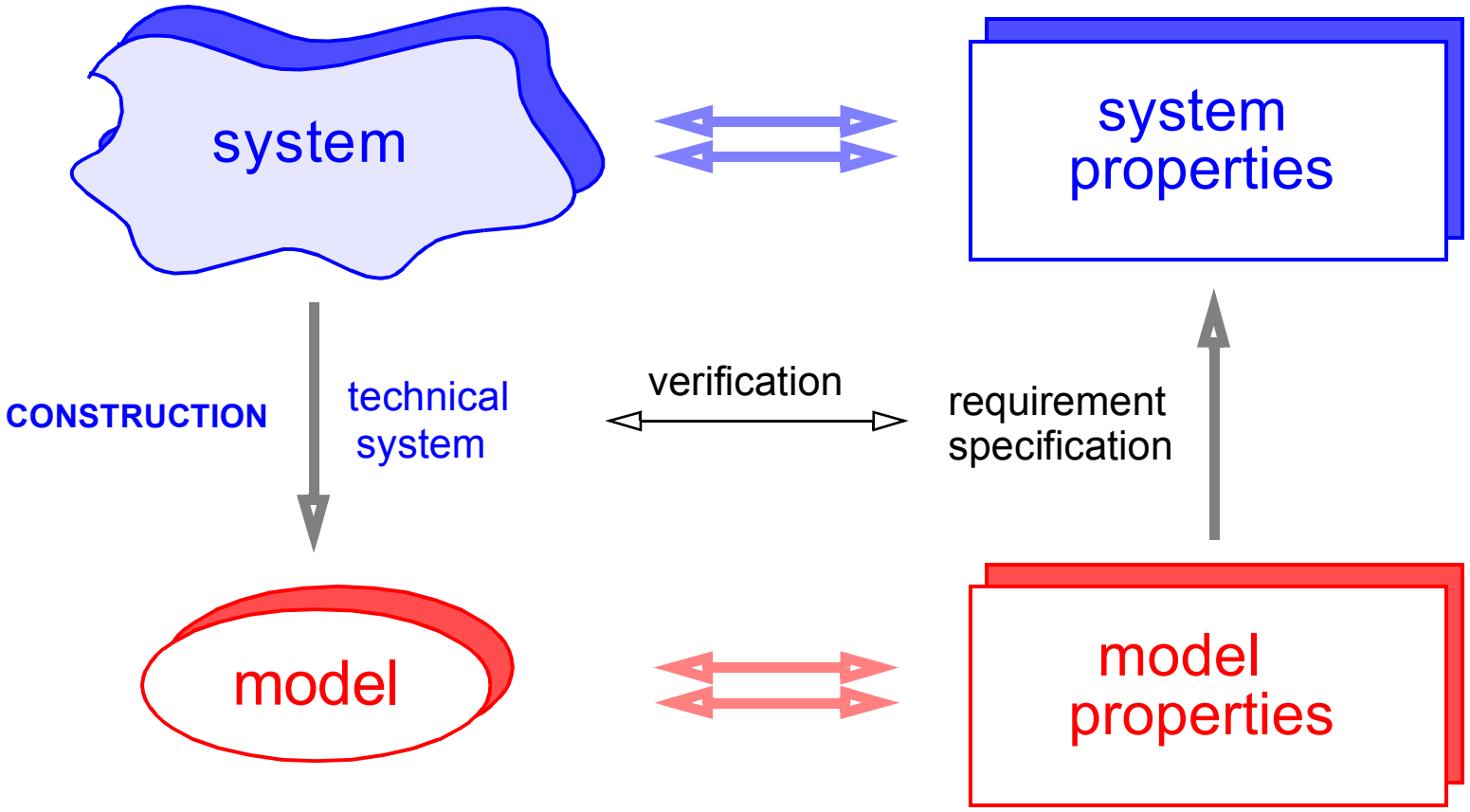

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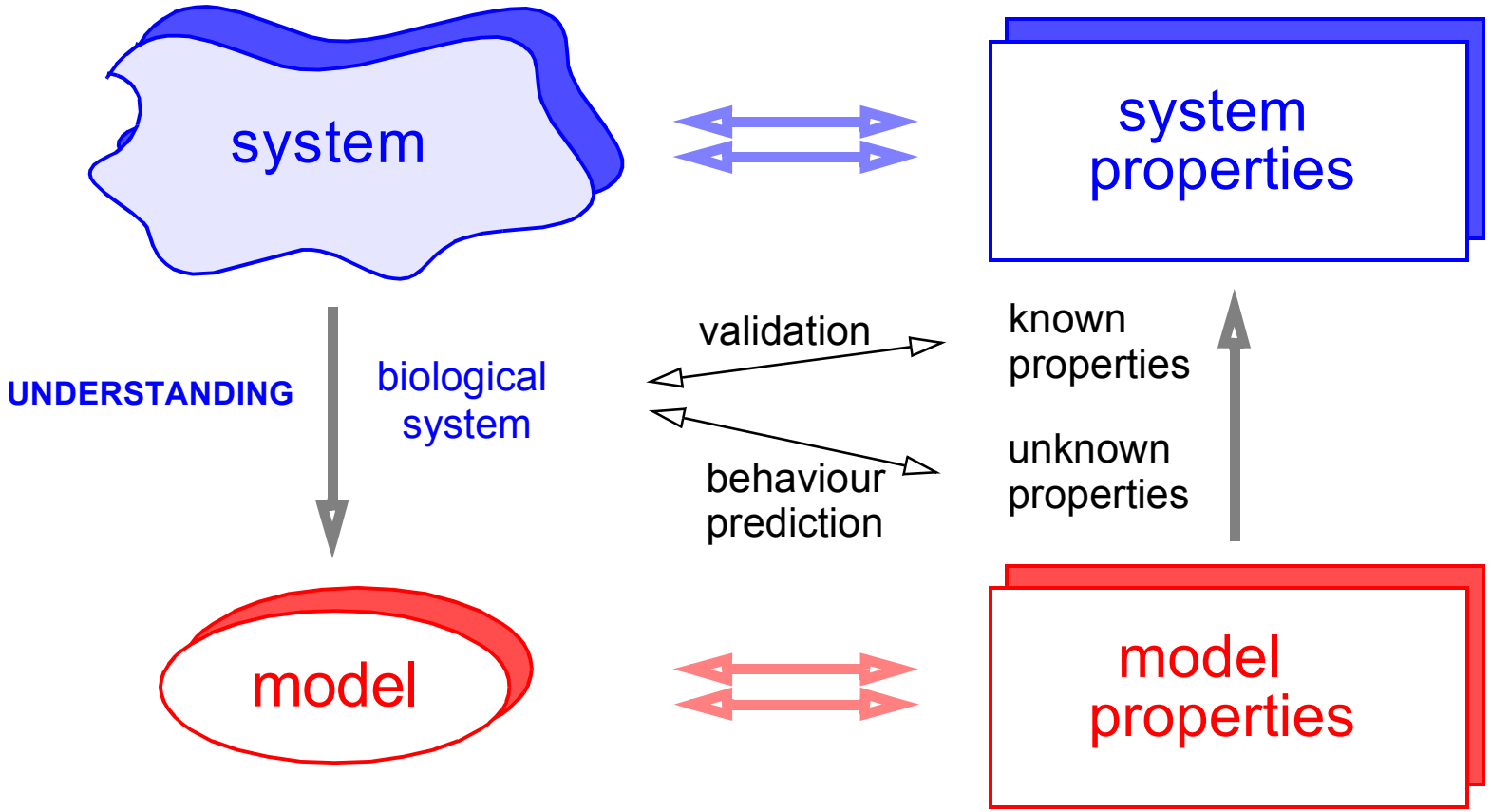
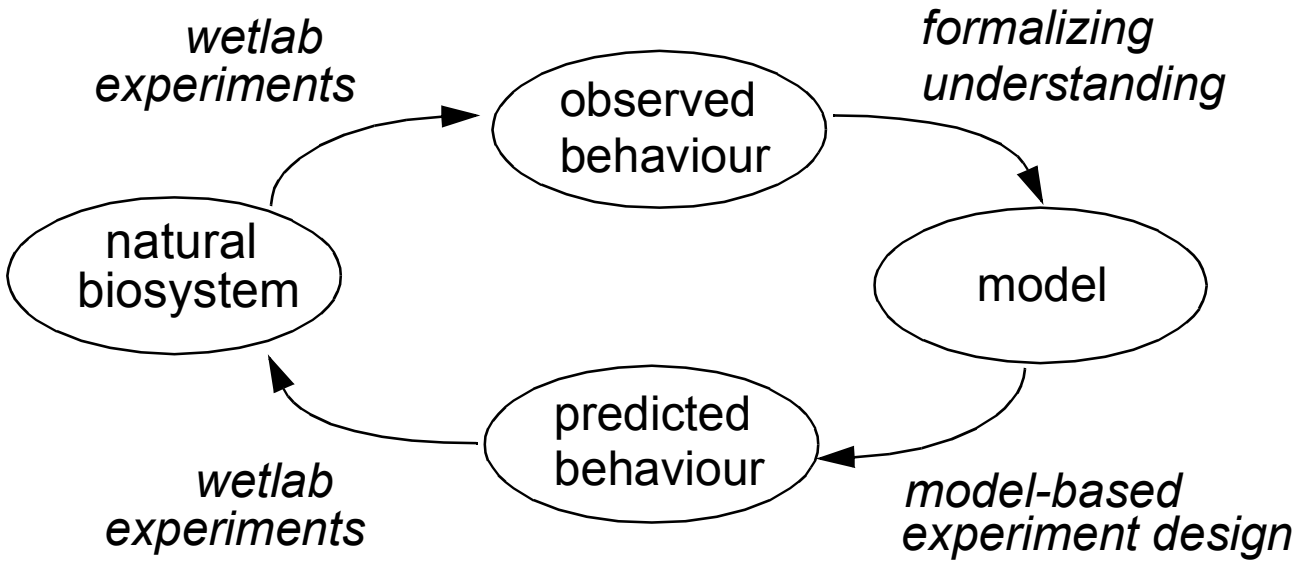


MODELS IN SYSTEMS BIOLOGY

MODELLING = FORMAL KNOWLEDGE REPRESENTATION



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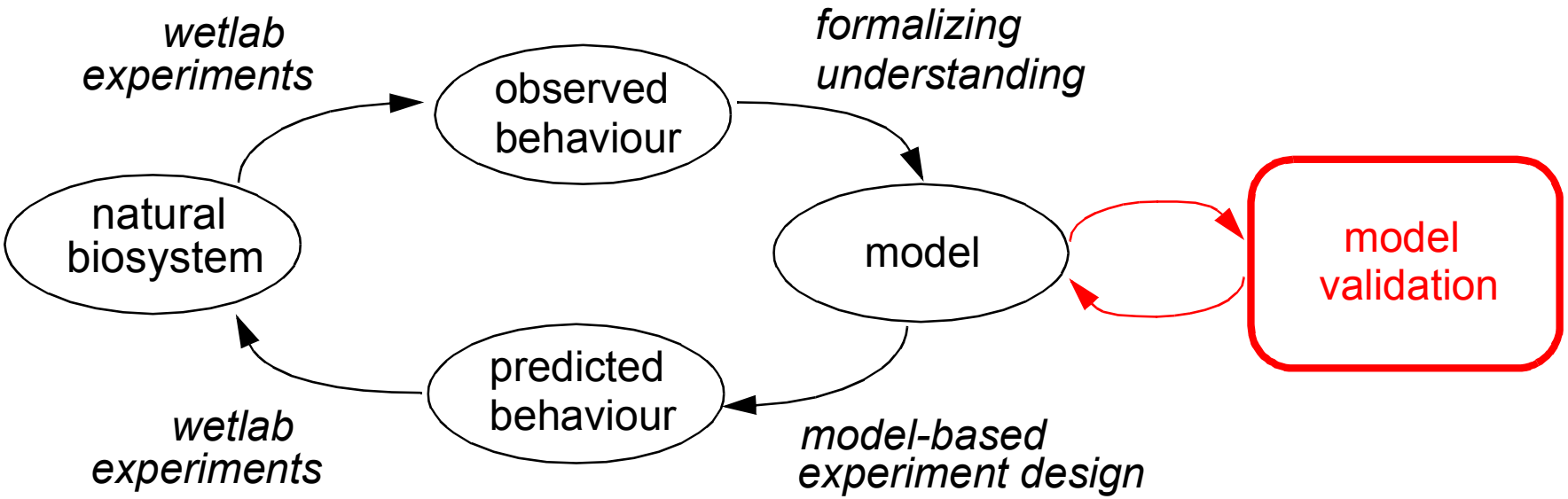
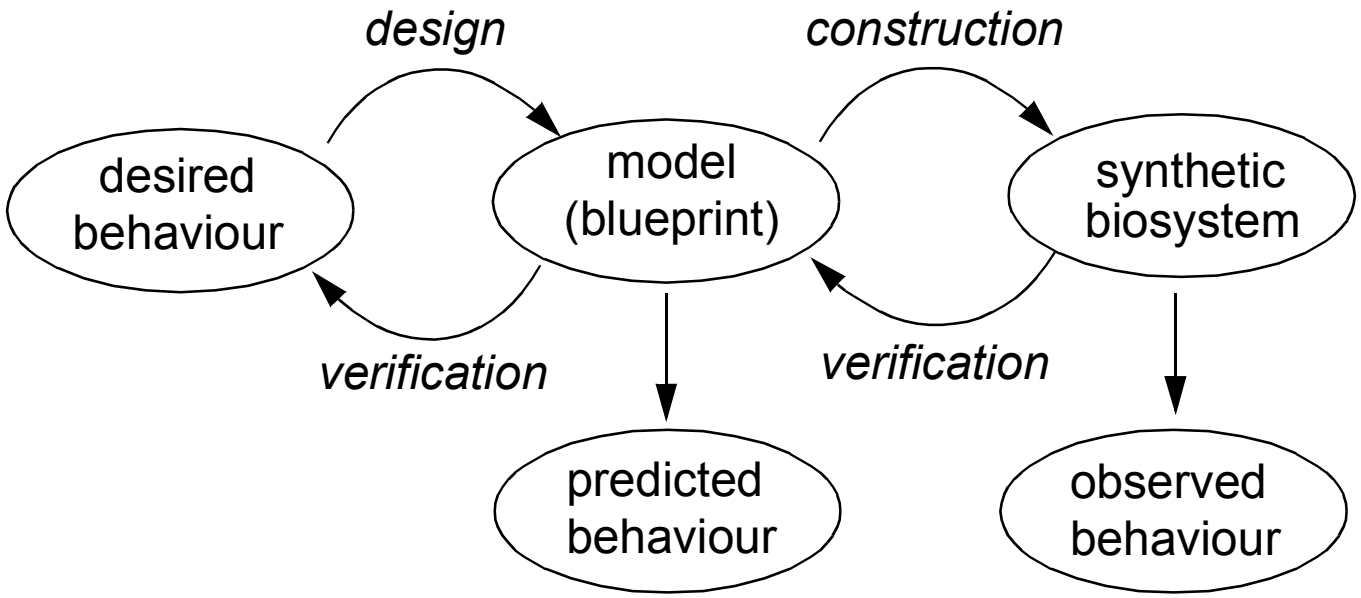


MODEL VALIDATION = CONFIDENCE INCREASE

MODELS IN SYNTHETIC BIOLOGY

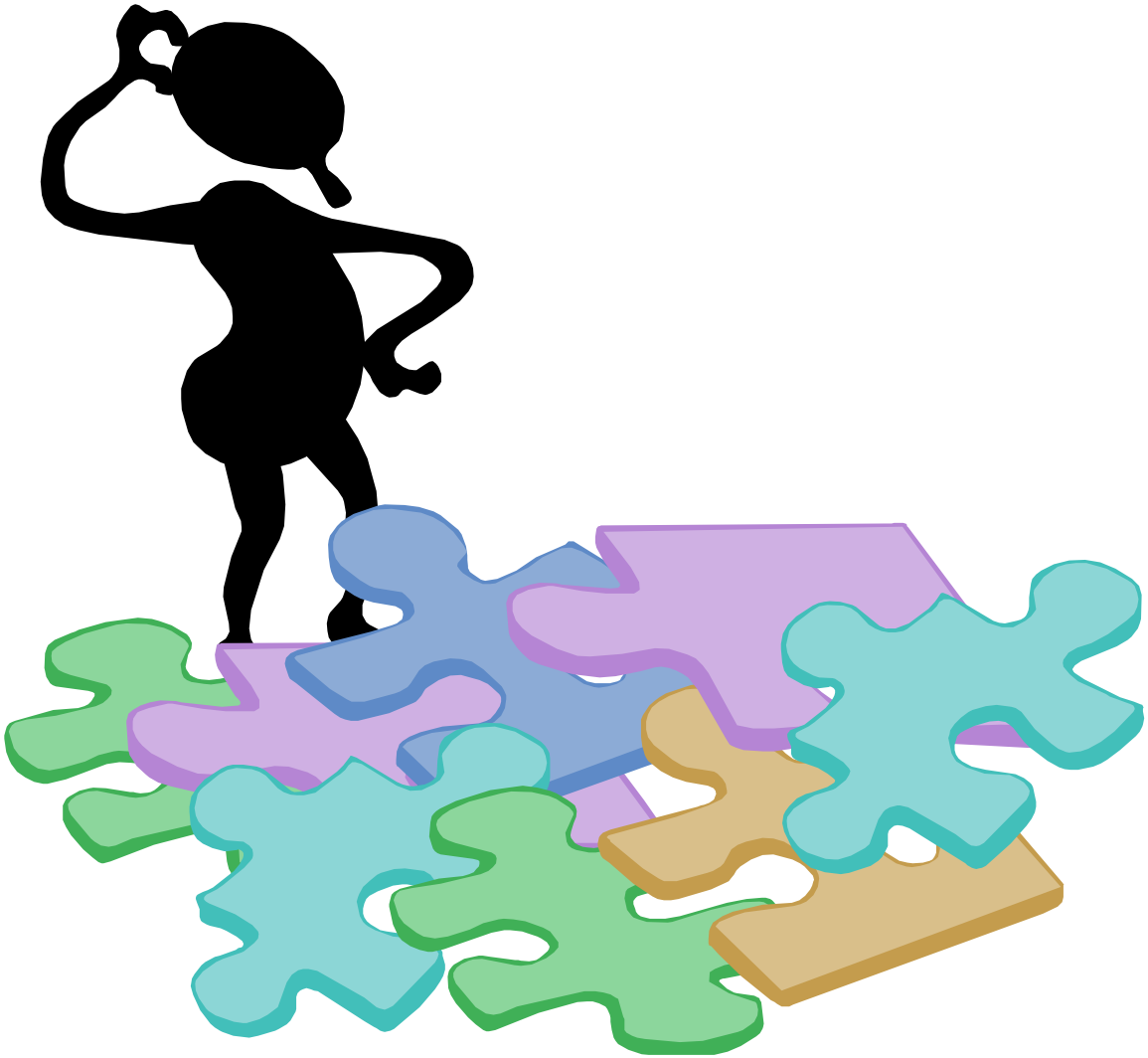
MODELLING = BLUEPRINT FOR SYSTEM CONSTRUCTION



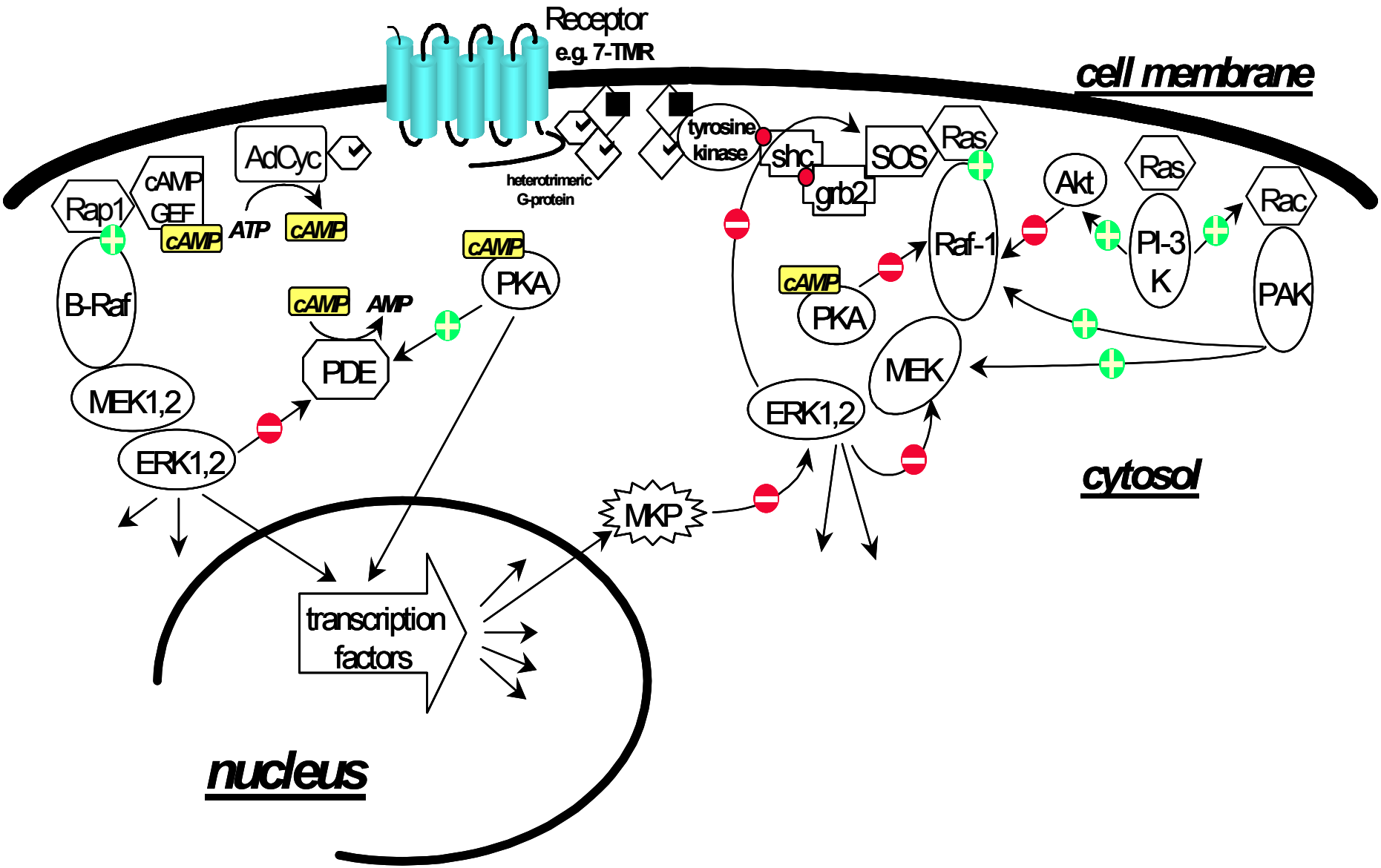
RELIABLE AND ROBUST ENGINEERING REQUIRES VERIFIED MODELS

WHAT KIND OF MODEL SHOULD BE USED?

BIO NETWORKS, SOME PROBLEMS



NETWORK REPRESENTATIONS, EX1



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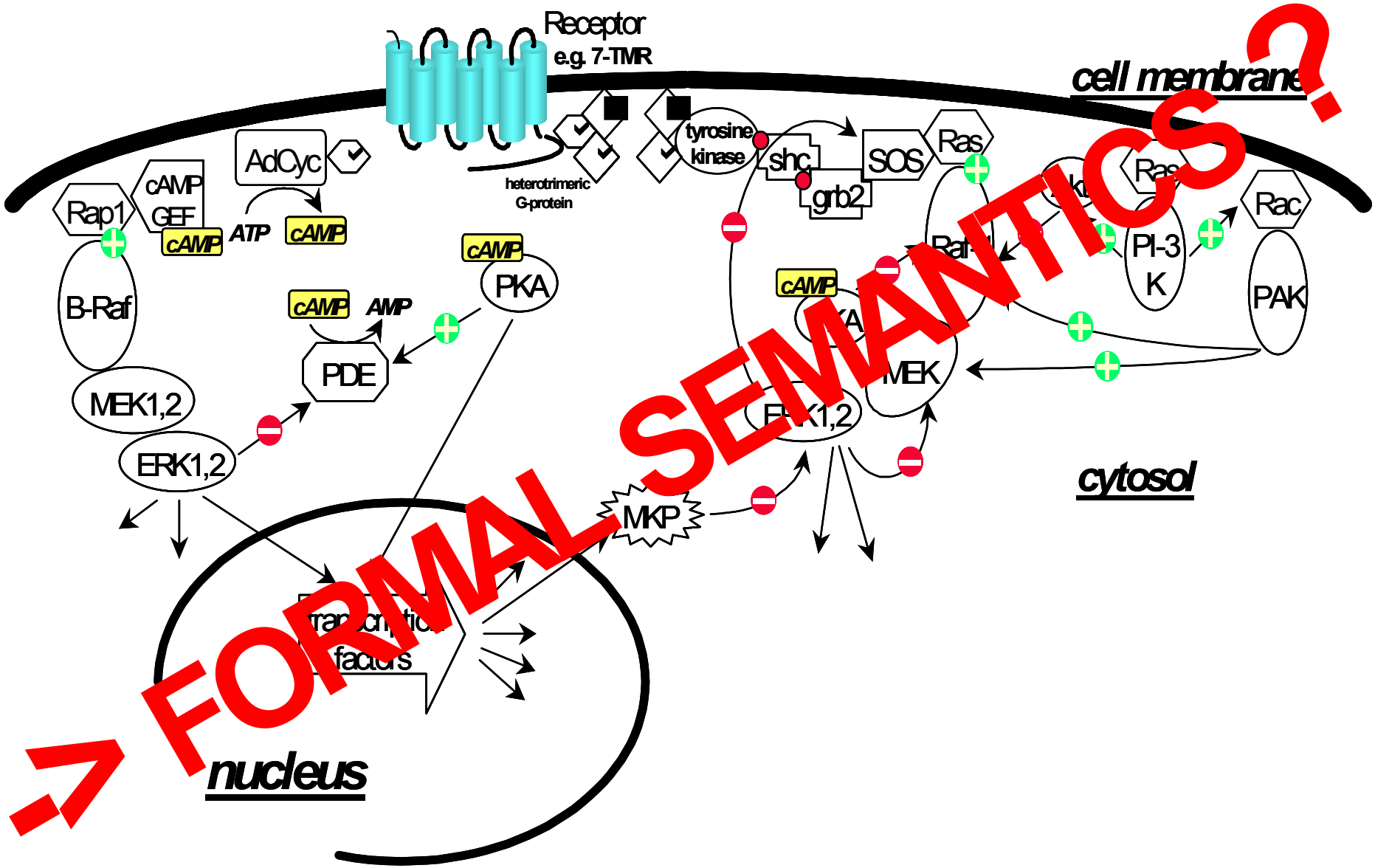

NETWORK REPRESENTATIONS, EX2

$$\frac{d\alpha}{dt} = -v_1$$
$$\frac{dSte2}{dt} = -v_2 + v_3 - v_5$$
$$\frac{dSte2_{active}}{dt} = v_2 - v_3 - v_4$$
$$\frac{dSst2_{active}}{dt} = v_{46} - v_{47}$$
$$\frac{dG\alpha\beta\gamma}{dt} = -v_6 + v_9$$
$$\frac{dG\alpha GTP}{dt} = v_6 - v_7 - v_8$$
$$\frac{dG\alpha GDP}{dt} = v_7 + v_8 - v_9$$
$$\frac{dG\beta\gamma}{dt} = v_6 - v_9 - v_{10} + v_{11} + v_{21} + v_{23} + v_{25} + v_{27} + v_{32} - v_{42} + v_{43}$$
$$\frac{dSte5}{dt} = -v_{12} + v_{13} + v_{17} + v_{21} + v_{23} + v_{25} + v_{27} + v_{32}$$
$$\frac{dSte11}{dt} = -v_{12} + v_{13} + v_{17} + v_{21} + v_{23} + v_{25} + v_{27} + v_{32}$$
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$$\frac{dFus3}{dt} = -v_{14} + v_{15} + v_{17} + v_{21} + v_{23} + v_{25} + v_{27} - v_{29} + v_{30} + v_{33}$$
$$\frac{dSte20}{dt} = -v_{18} + v_{19} + v_{21} + v_{23} + v_{25} + v_{27} + v_{32}$$

$$v_1 = \alpha[t] \cdot Bar1_{active}[t] \cdot k_1$$
$$v_2 = Ste2[t] \cdot \alpha[t] \cdot k_2$$
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$$v_7 = G\alpha GTP[t] \cdot k_7$$
$$v_8 = G\alpha GTP[t] \cdot Sst2_{active}[t] \cdot k_8$$
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BIO NETWORKS, SOME PROBLEMS

- **knowledge** ***-> PROBLEM 1***
  - -> *uncertain*
  - -> *growing, changing*
  - -> *distributed over independent data bases, papers, journals, . . .*
- **various, mostly ambiguous representations** ***-> PROBLEM 2***
  - -> *verbose descriptions*
  - -> *diverse graphical representations*
  - -> *contradictory and / or fuzzy statements*
- **network structure** ***-> PROBLEM 3***
  - -> *tend to grow fast*
  - -> *dense, apparently unstructured*
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-> MODELS ARE FULL OF ASSUMPTIONS <-

BIO NETWORK REPRESENTATIONS SHOULD BE

- **readable**
  - -> *fault avoidance*
  - -> *informal = cartoon-like representations ?*
- **analysable**
  - -> *formal = mathematical representations*
- **executable**
  - -> *to experience the model*
- **unifying power**
  - -> *high-level description for various analysis approaches*

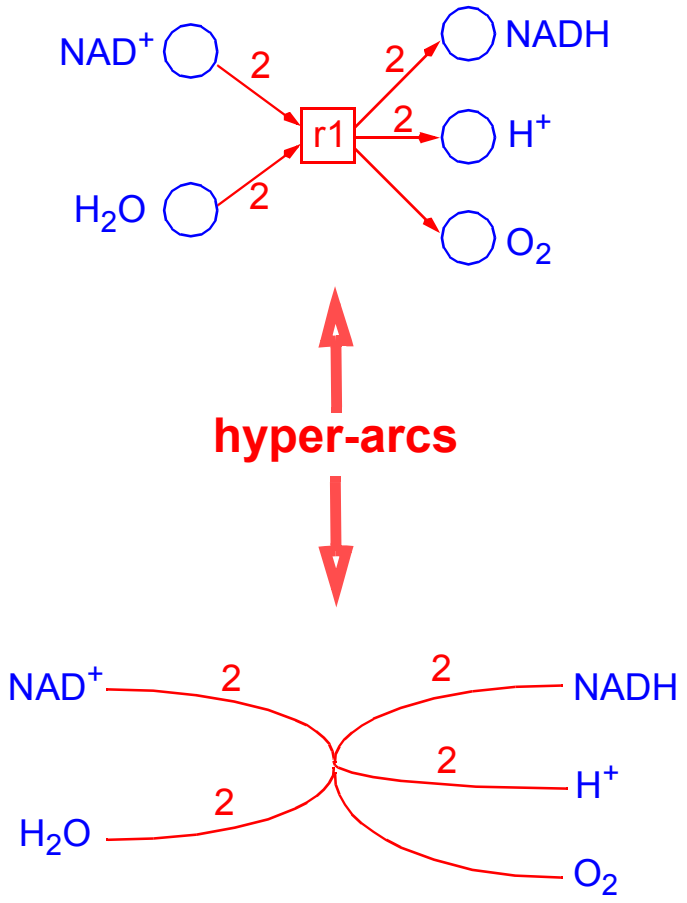
. . .

ARE NETWORKS OF BIOCHEMICAL REACTIONS

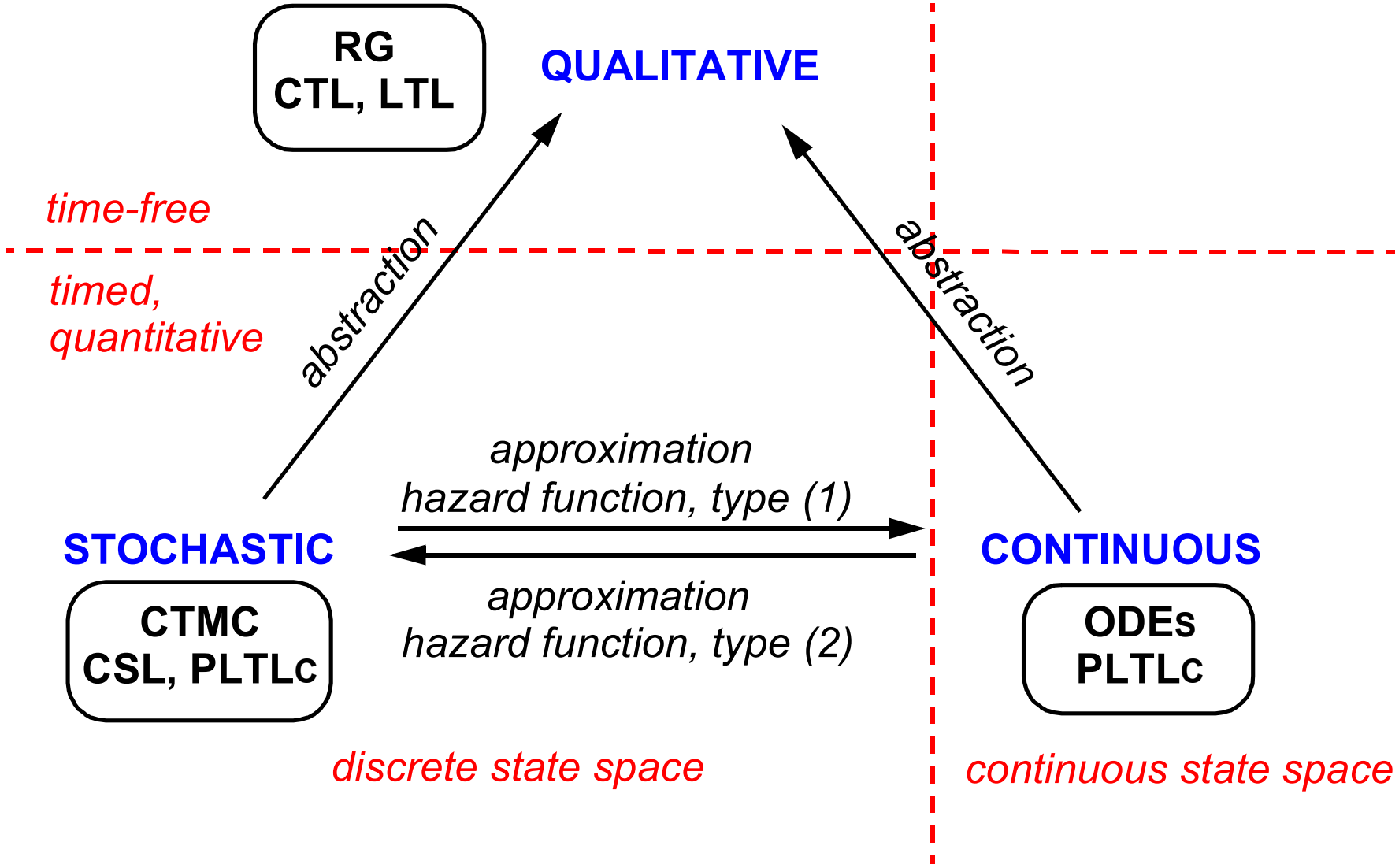
. . .

NATURALLY EXPRESSIBLE AS PETRI NETS

$2\ NAD^+ + 2\ H_2O \rightarrow 2\ NADH + 2\ H^+ + O_2$



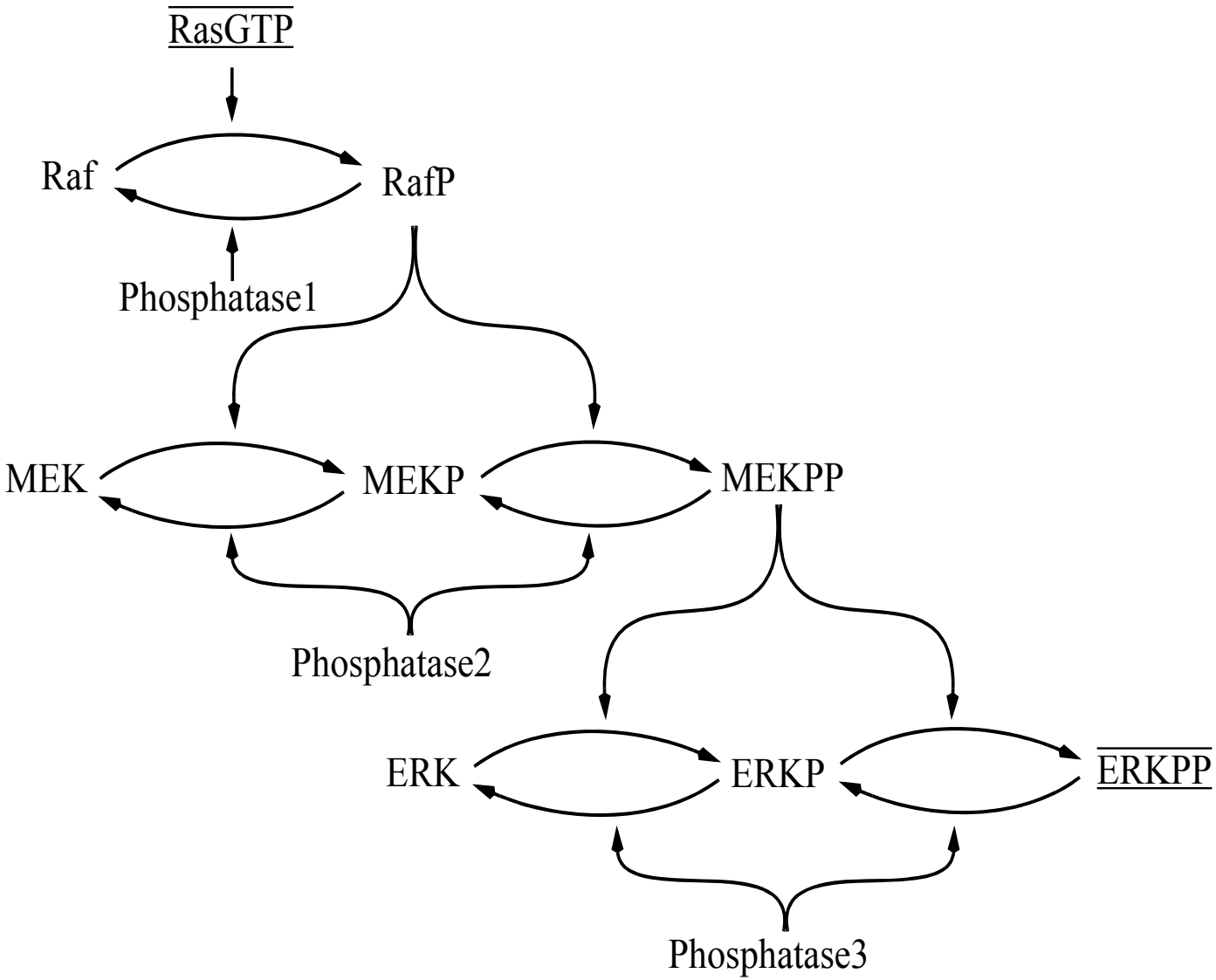
SUMMARY, FRAMEWORK



BIO PETRI NETS - SOME EXAMPLES

monika.heiner@informatik.tu-cottbus.de

EX1 - SIGNALLING CASCADE



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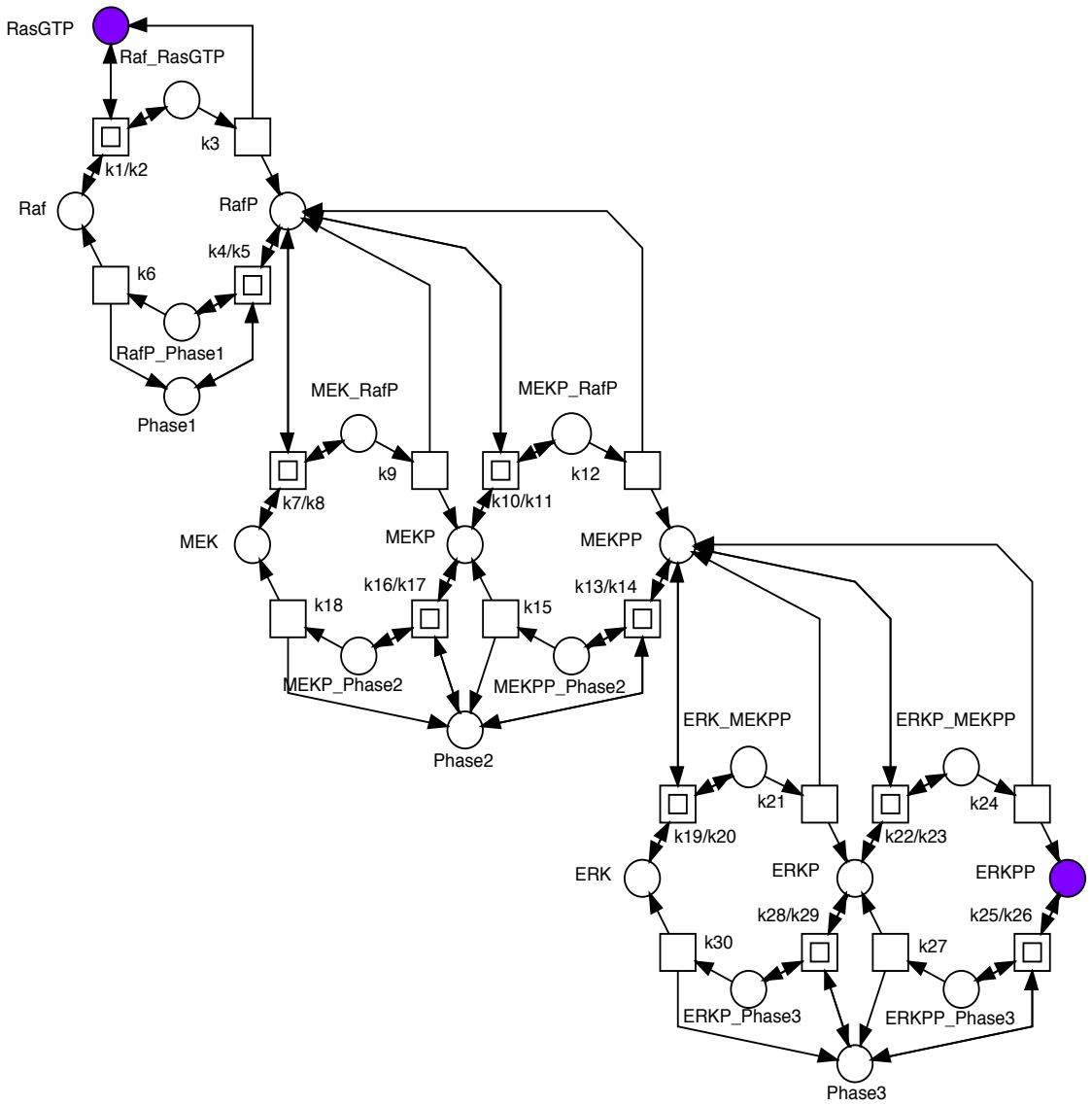
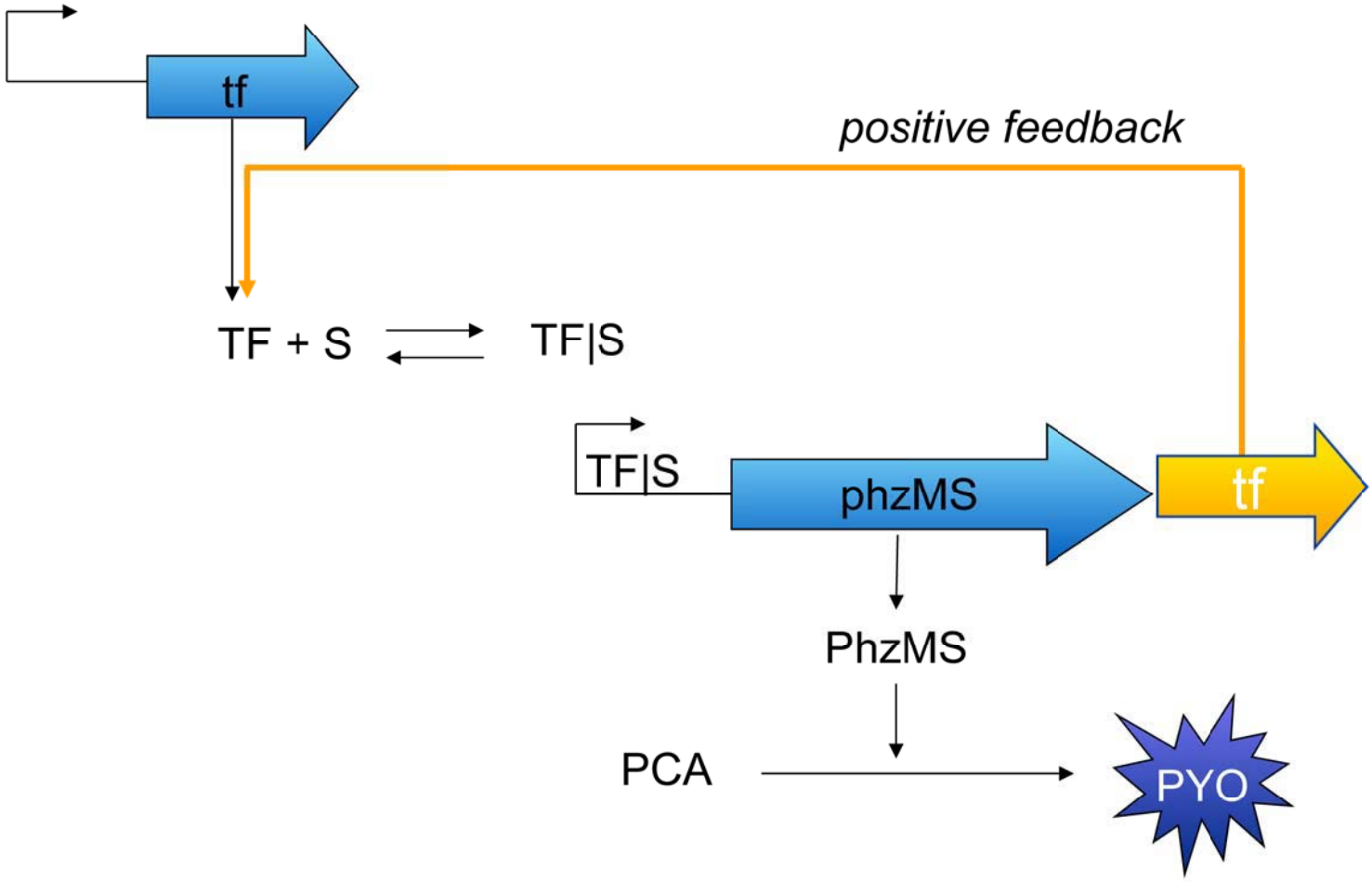


EX2 - BIOSENSOR



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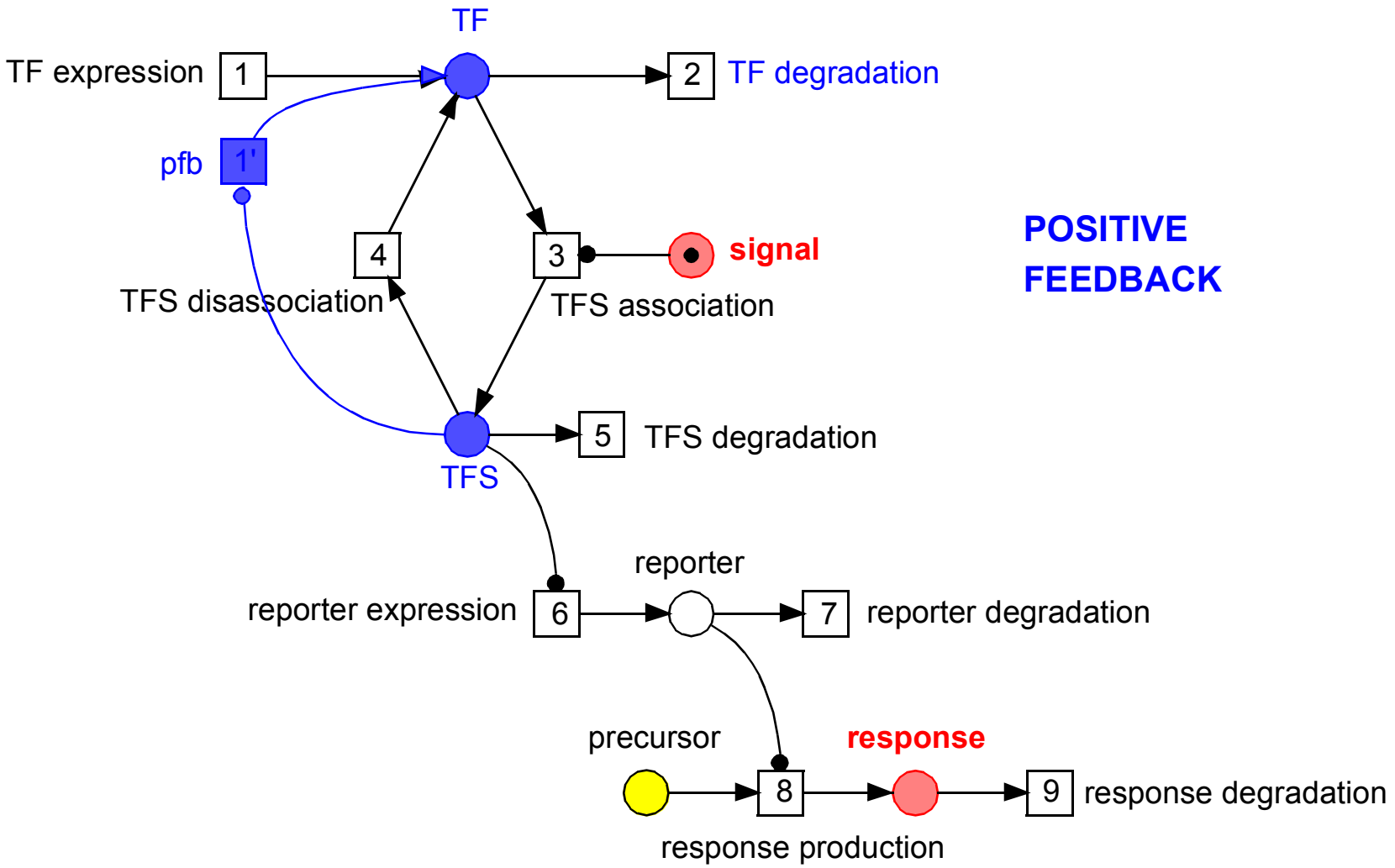
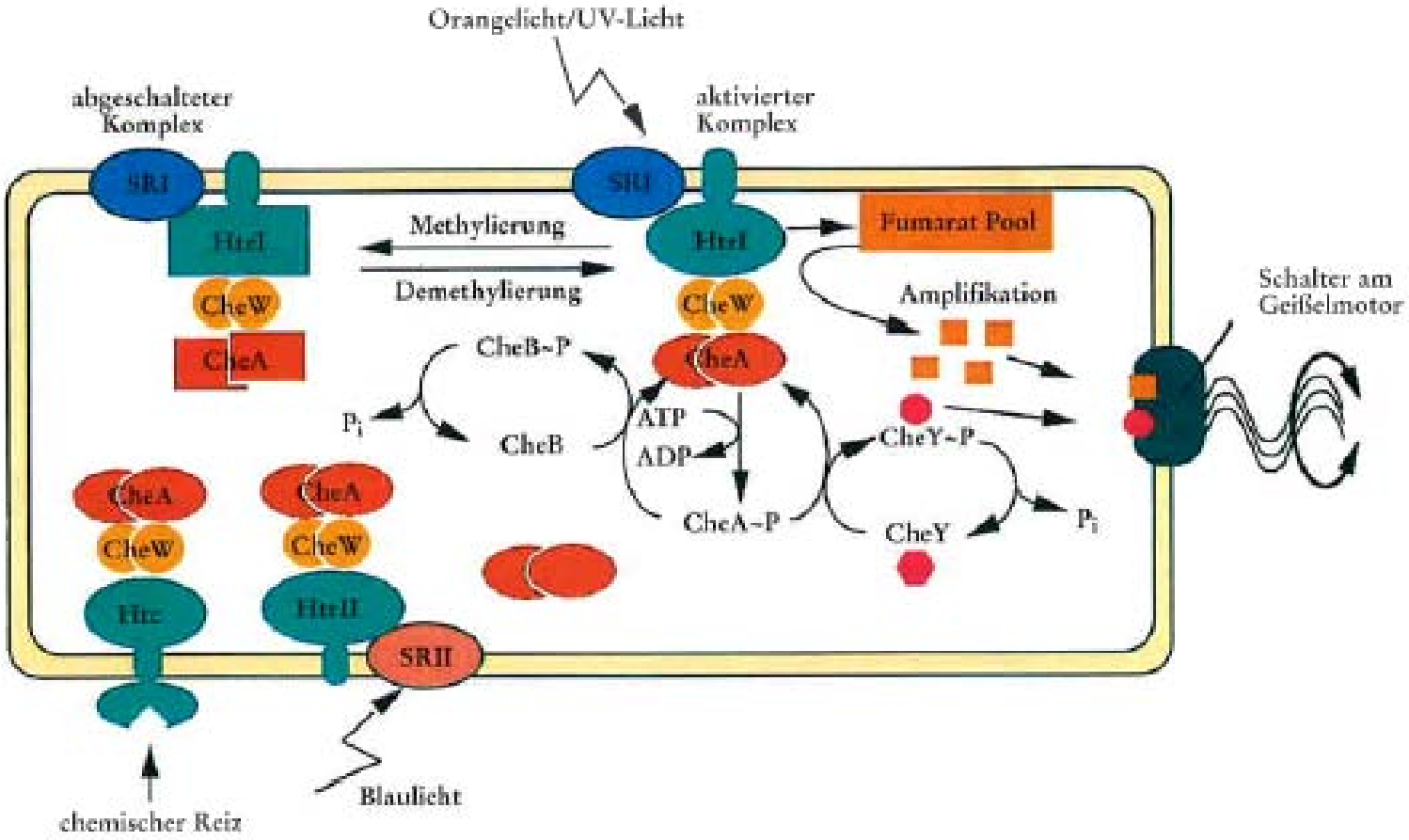


EX3 - SWITCH CYCLE HALOBACTERIUM SALINARUM



[Marwan; Oesterhelt 1999]

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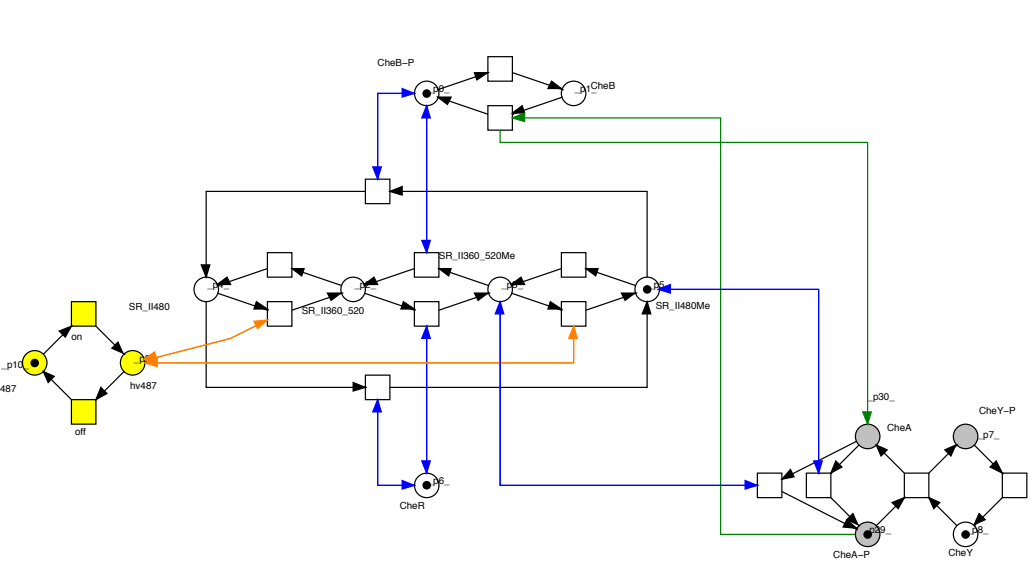
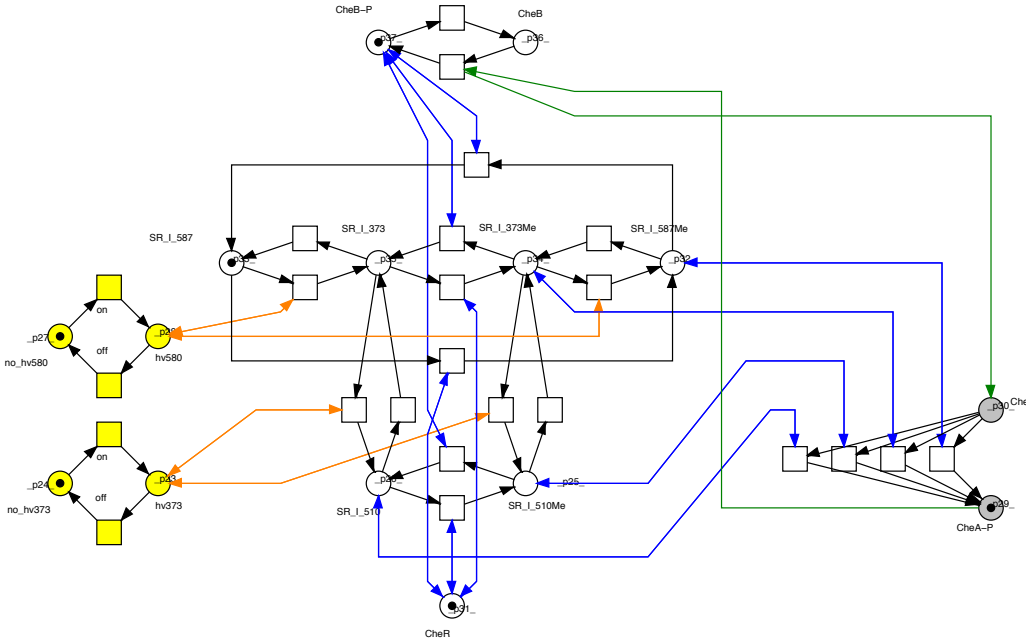
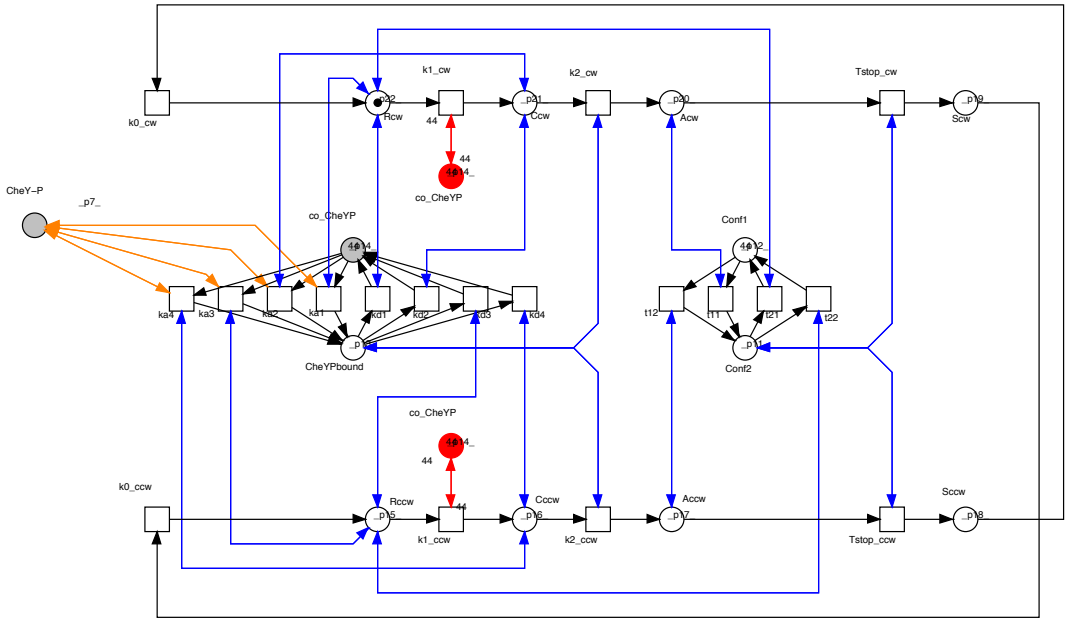
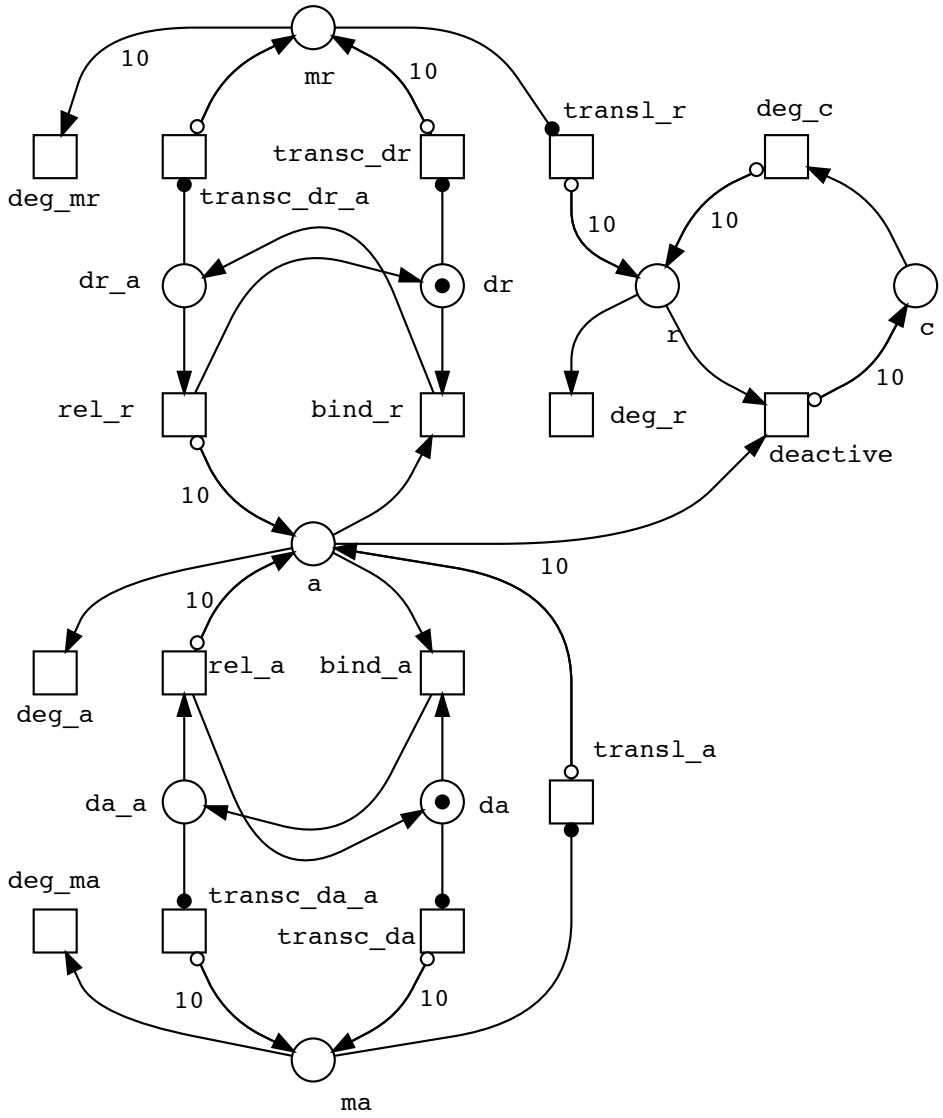






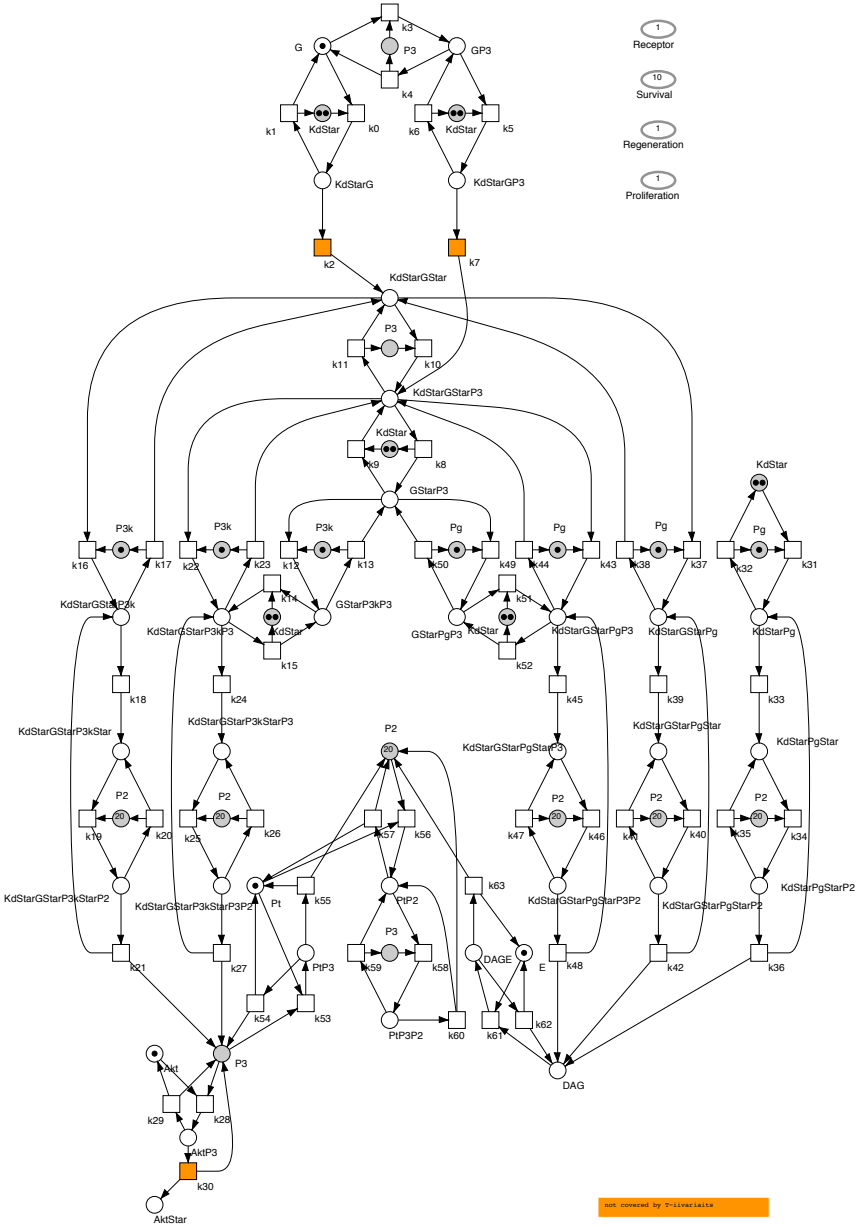
EX4 - CIRCADIAN CLOCK

[PRISM website]



EX5 - ANGIOGENESIS

[... Balbo ... 2009]



MOULARIZATION BY T-INVARIANTS

monika.heiner@informatik.tu-cottbus.de

T-INVARIANTS, EX1

r1: A -> 2 B
r2: 2 A -> 3 C

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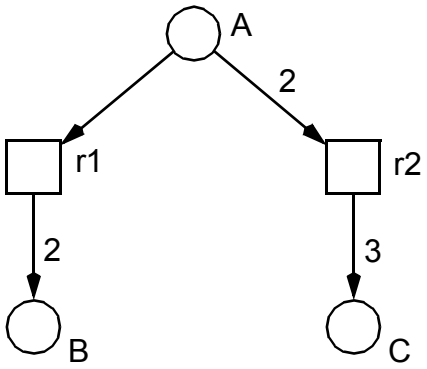

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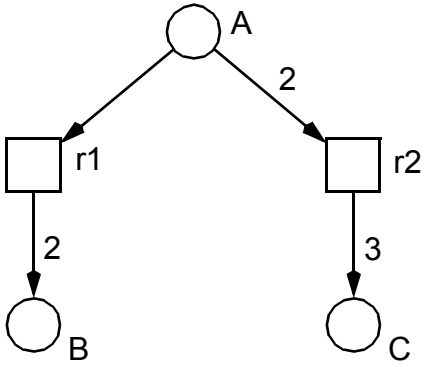
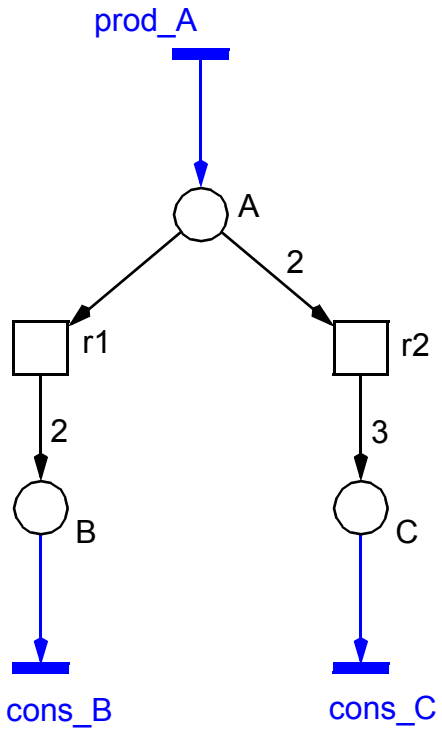
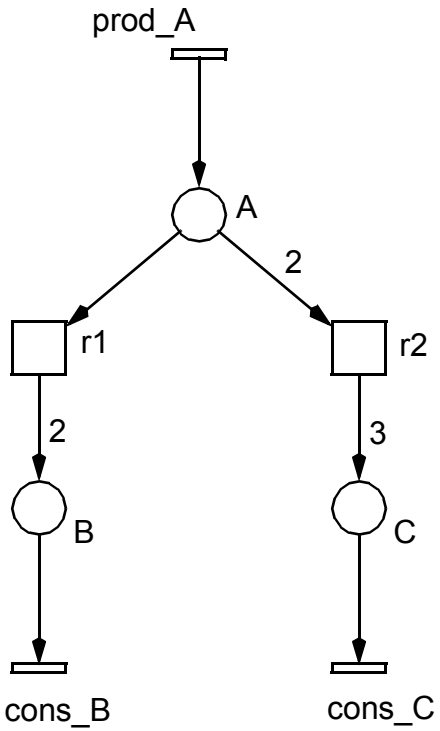


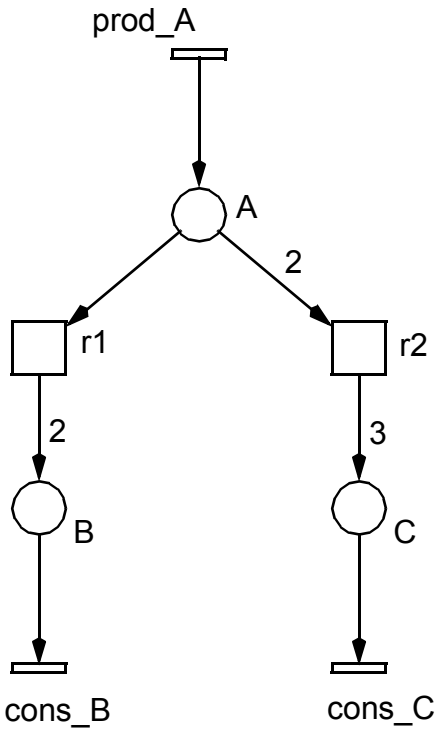


INCIDENCE MATRIX C, EX1



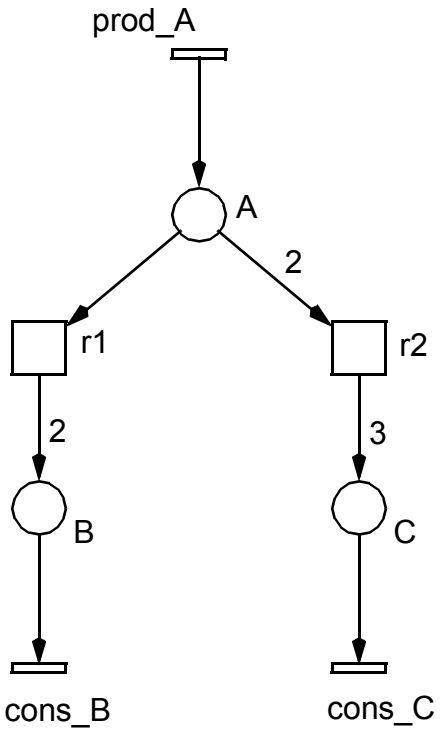
P \ T	r1	r2	prod_A	cons_B	cons_C
A					
B					
C					

INCIDENCE MATRIX C, EX1



P \ T	r1	r2	prod_A	cons_B	cons_C
A	-1	-2	1		
B	2			-1	
C		3			-1

INCIDENCE MATRIX C, EX1



P \ T	r1	r2	prod_A	cons_B	cons_C
A	-1	-2	1		
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C		3			-1

1 **1** **2**

T-INVARIANTS, BASICS

- **Lautenbach, 1973** -> **Schuster, 1993**
- **T-invariants** -> *multisets of transitions*
  - -> *integer solutions x*

$$Cx = 0, x \neq 0, x \geq 0$$

- **minimal T-invariants**
  - -> *there is no T-invariant with a smaller support* -> *sets of transitions*
  - -> *gcD of all non-zero entries is 1*
- **any T-invariant is a non-negative linear combination of minimal ones**
  - -> *multiplication with a positive integer*
  - -> *addition*
  - -> *division by a common divisor*

$$kx = \sum_i a_i x_i$$

- **Covered by T-Invariants (CTI)**
  - -> *each transition belongs to a T-invariant*

T-INVARIANTS, INTERPRETATIONS

- **T-invariants = (multi-) sets of transitions = Parikh vector**
  - -> *zero effect on marking*
  - -> *reproducing a marking / system state*

- **two interpretations**
  1. *partially ordered transition sequence* **-> behaviour understanding**
     of transitions occuring one after the other
     -> *substance / signal flow*
  2. *relative transition firing rates* **-> steady state behaviour**
     of transitions occuring permanently & concurrently
     -> *steady state behaviour*

- **a minimal T-invariant defines a connected subnet**
  - -> *the T-invariant's transitions (the support),*
    + all their pre- and post-places
    + the arcs in between
  - -> *pre-set of support = post-set of support*

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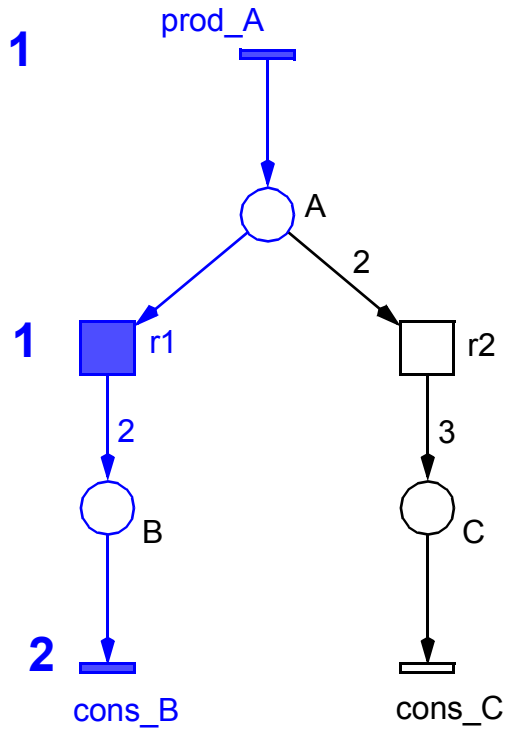

T-INVARIANT 1

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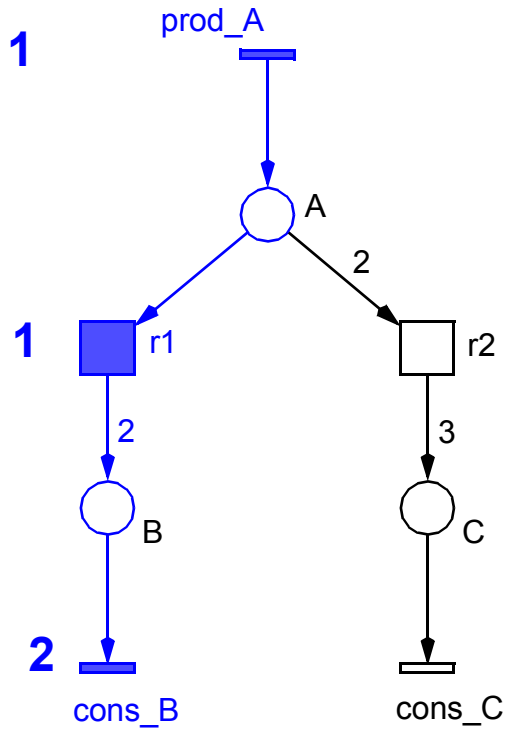

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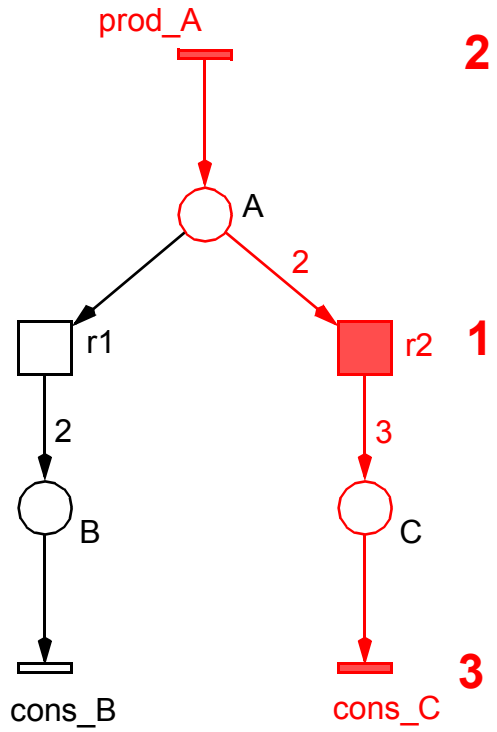
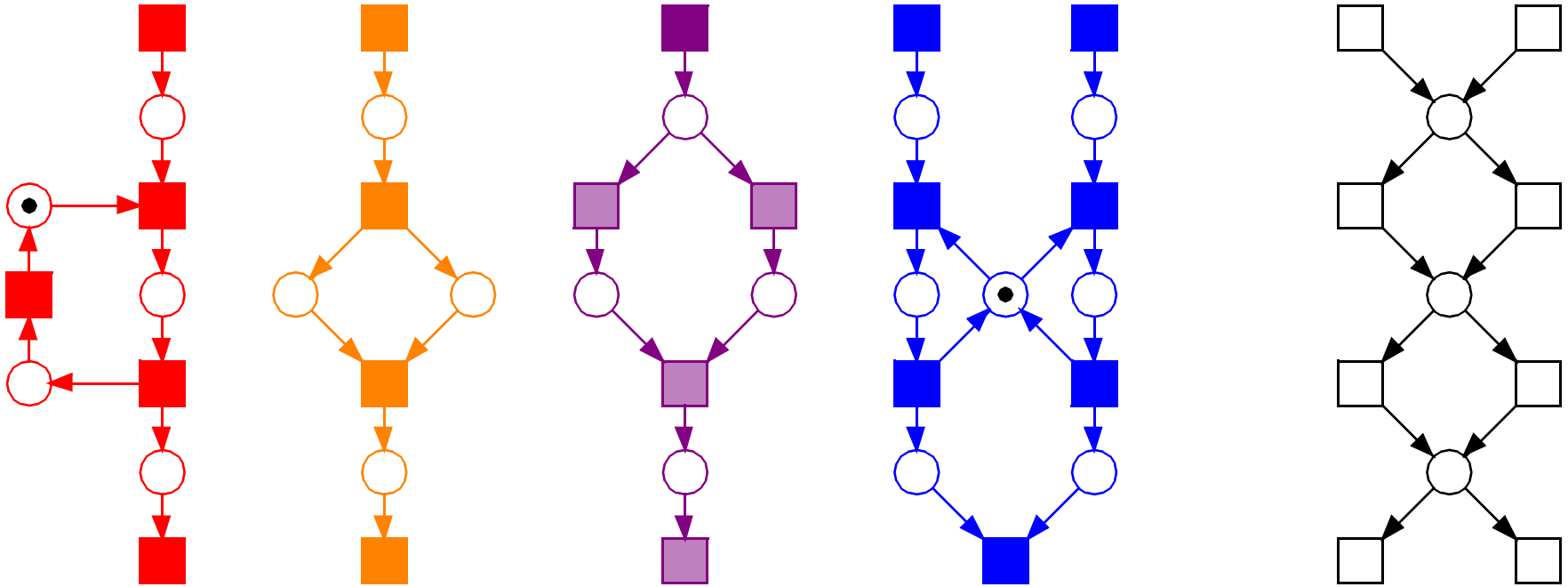


T-INVARIANT 2

T-INVARIANTS, OBSERVATIONS

- **T-invariants may contain any structure**



- **T-invariants generally overlap**

  -> combinatorial effect brings explosion in the number of min. T-invariants **(2^4)**

ABSTRACT DEPENDENT TRANSITION SETS (ADT-SETS)

- Let X denote a set of (all / non-trivial) minimal T-invariants x of a given PN.

- **dependency relation:**
  Two transitions i, j depend on each other,
  if they always appear together in all minimal T-invariants x, i.e.

$$\forall x \in X: \; i \in supp(x) \Leftrightarrow j \in supp(x)$$

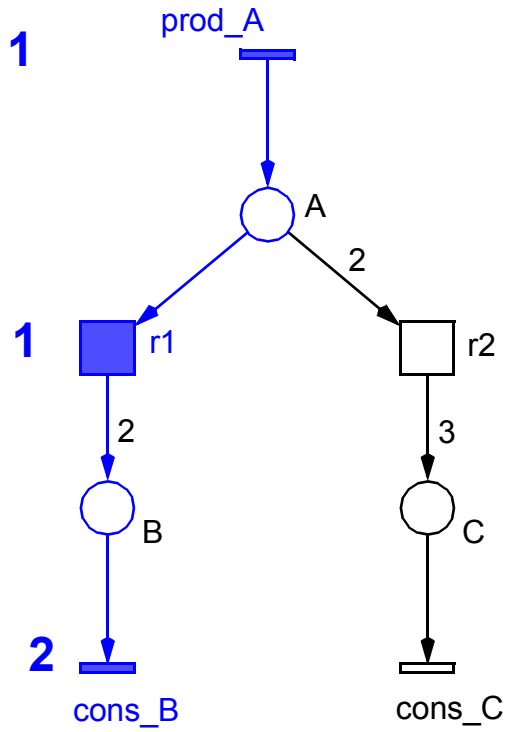
- **equivalence relation in the transition set, leading to a partition of T**
  - -> *reflexive*
  - -> *symmetric*
  - -> *transitive*

- **the equivalence classes A represent maximal ADT-sets**

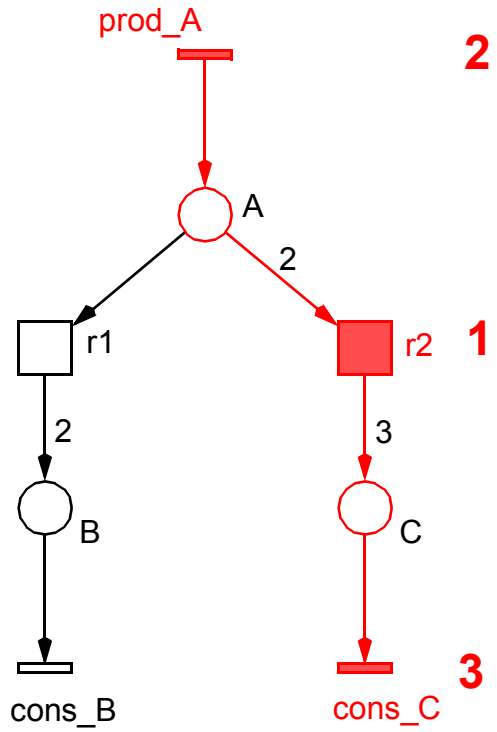
$$\forall x \in X: A \subseteq supp(x) \vee A \cap supp(x) = \varnothing$$

ADT-SETS, Ex1

r1: A -> 2 B
r2: 2 A -> 3 C



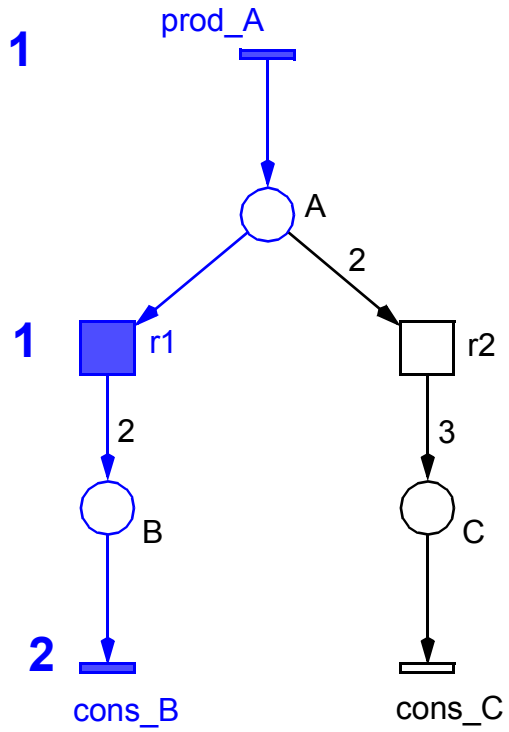
T-INVARIANT 1



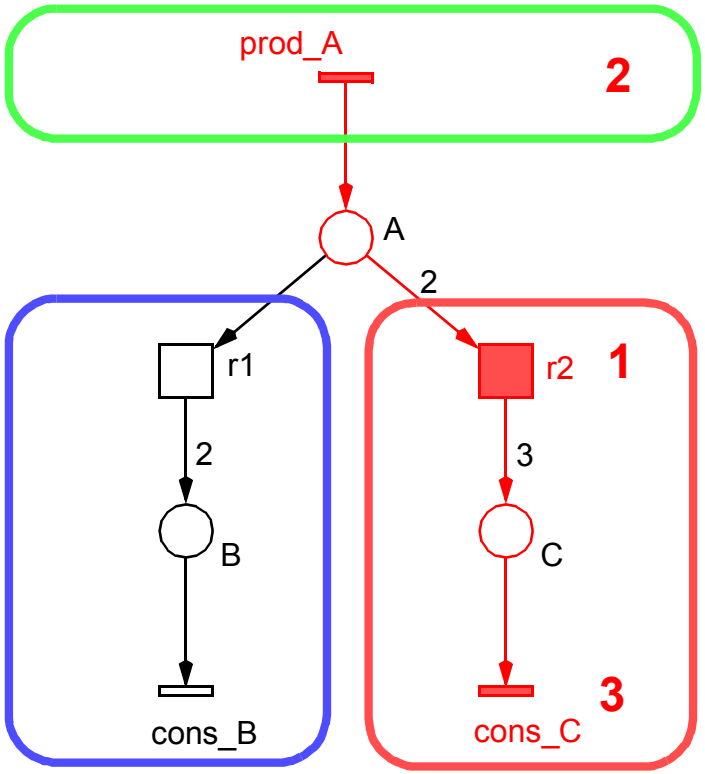
T-INVARIANT 2

ADT-SETS, Ex1

r1: A -> 2 B
r2: 2 A -> 3 C



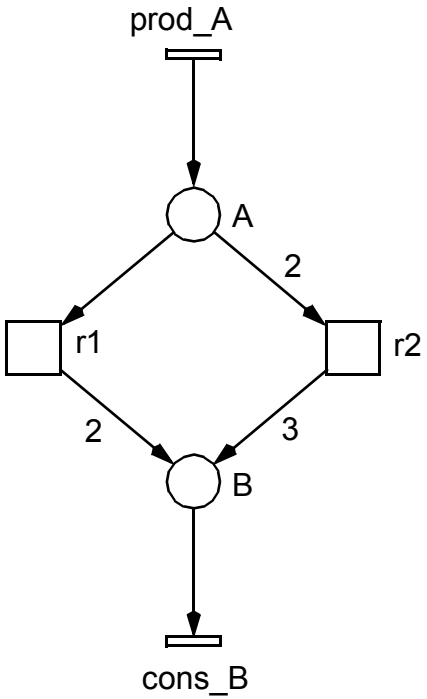
T-INVARIANT 1



T-INVARIANT 2

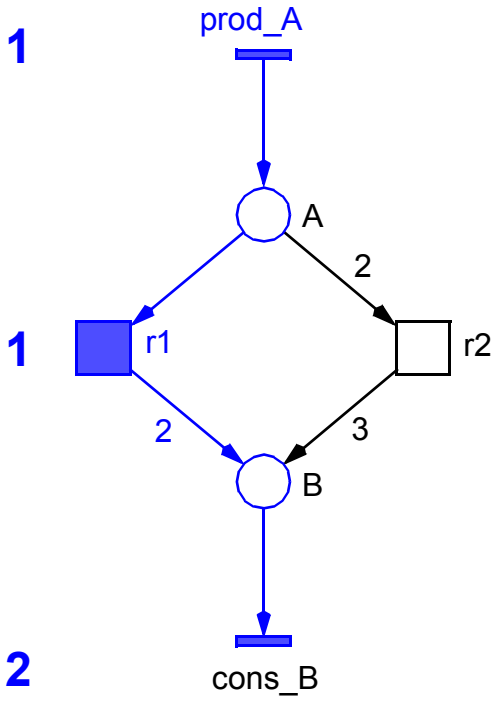
ADT-SETS, Ex2

r1: A -> 2 B
r2: 2 A -> 3 B

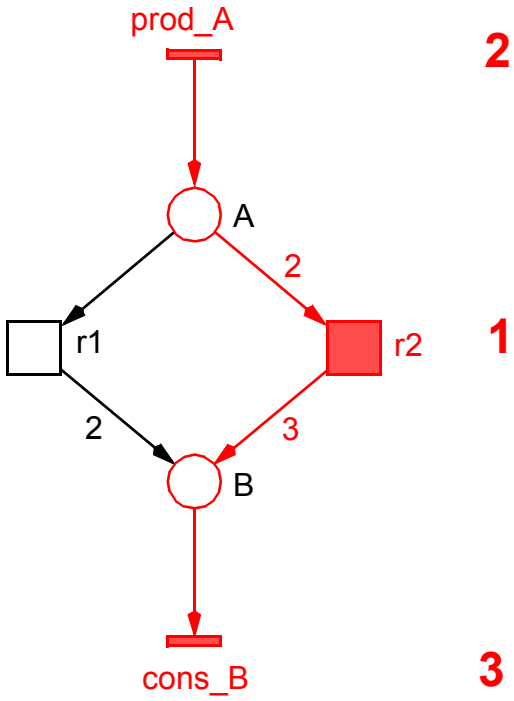


ADT-SETS, Ex2

r1: A -> 2 B
r2: 2 A -> 3 B



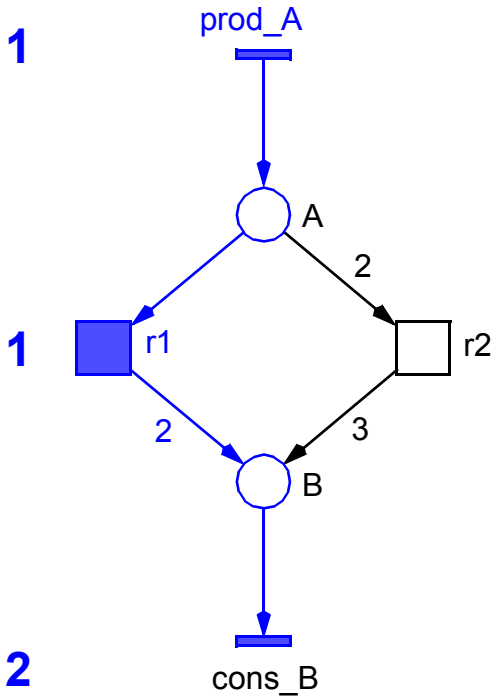
T-INVARIANT 1



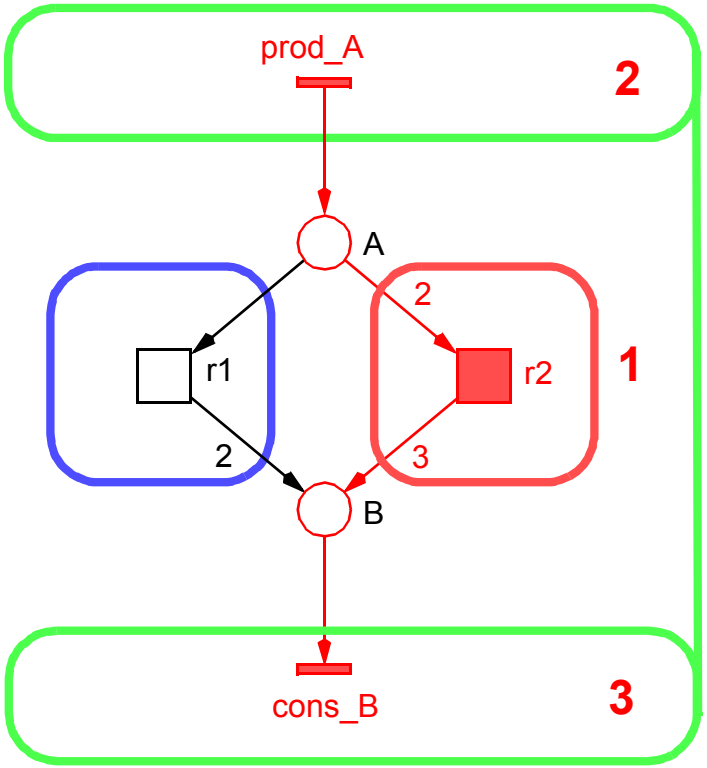
T-INVARIANT 2

ADT-SETS, Ex2

r1: A -> 2 B
r2: 2 A -> 3 B



T-INVARIANT 1



T-INVARIANT 2

ADT-SETS, INTERPRETATION

- **maximal ADT-sets**
  - -> *disjunctive subnets*
  - -> *not necessarily connected*

minimal T-invariants
- -> *overlapping subnets*
- -> *connected*

- **interpretation**
  - -> *structural decomposition into rather small subnets*
  - -> *smallest biologically meaningful functional units*
  - -> *building blocks*

- **variations**
  - -> *with / without trivial T-invariants*
  - -> *whole / partial set of T-invariants*

} *not necessarily maximal ADT-sets*

- **classification of all transitions based on the T-invariants' support**

ADT-SETS, APPLICATIONS

- **maximal ADT-sets**
  -> *not necessarily connected*

- **further decomposition into connected ADT-sets**
  -> *possibly according to primary compounds, only, i.e. neglecting connections by auxiliary compounds*
  -> *non-maximal ADT-sets*

- **coarse network structure, definition**
  -> *macro transitions* - *abstract from connected ADT-sets*
  -> *places* - *interface between functional units*

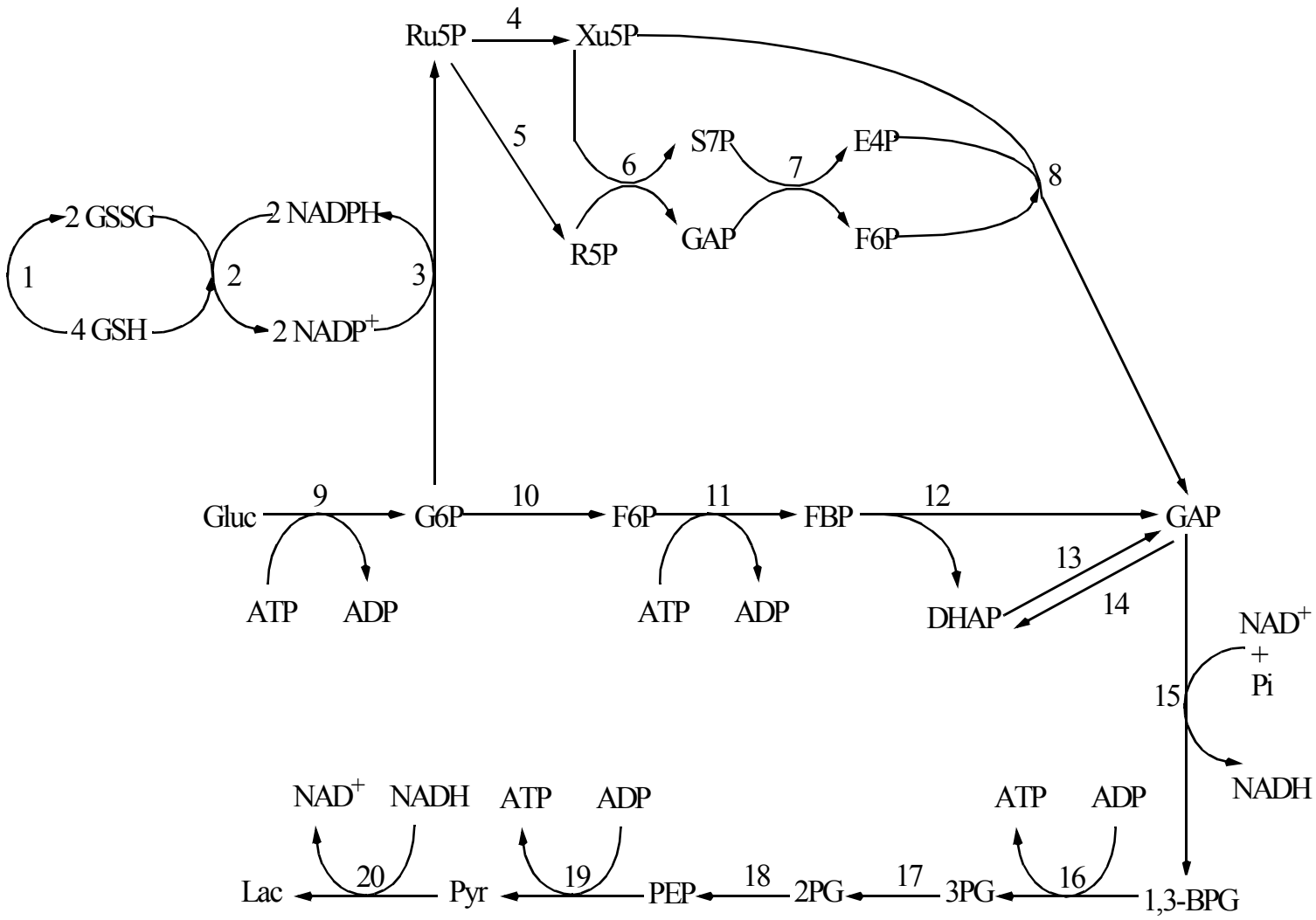
- **coarse network structure, what for?**
  -> *set of T-invariants gets structured*
  -> *better understanding of the net behaviour*

- **flow equivalent server component ?**

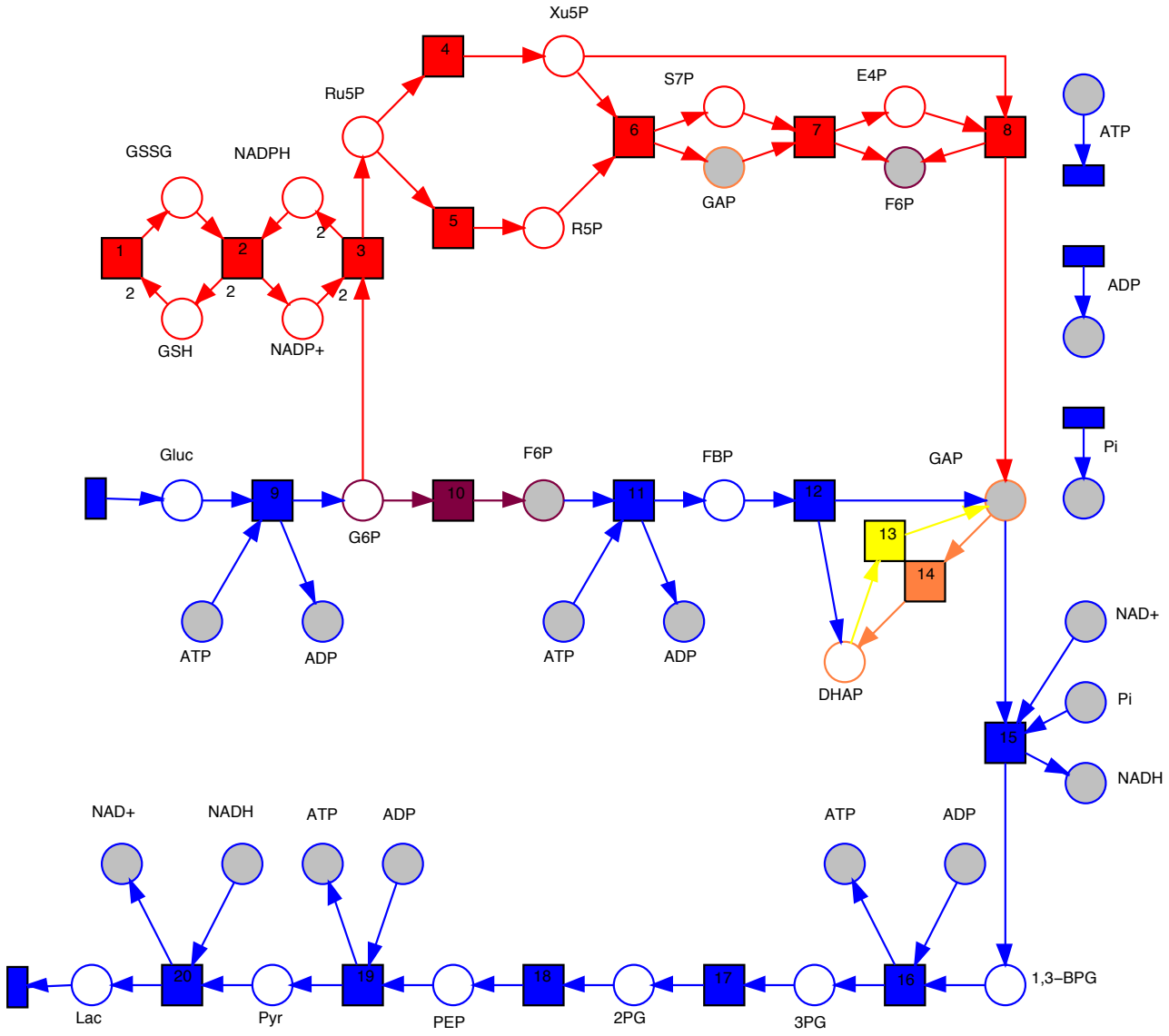
BIO PETRI NETS, SOME EXAMPLES

Ex1 - Glycolysis and Pentose Phosphate Pathway

[Reddy 1993]



Ex1 - Glycolysis and Pentose Phosphate Pathway



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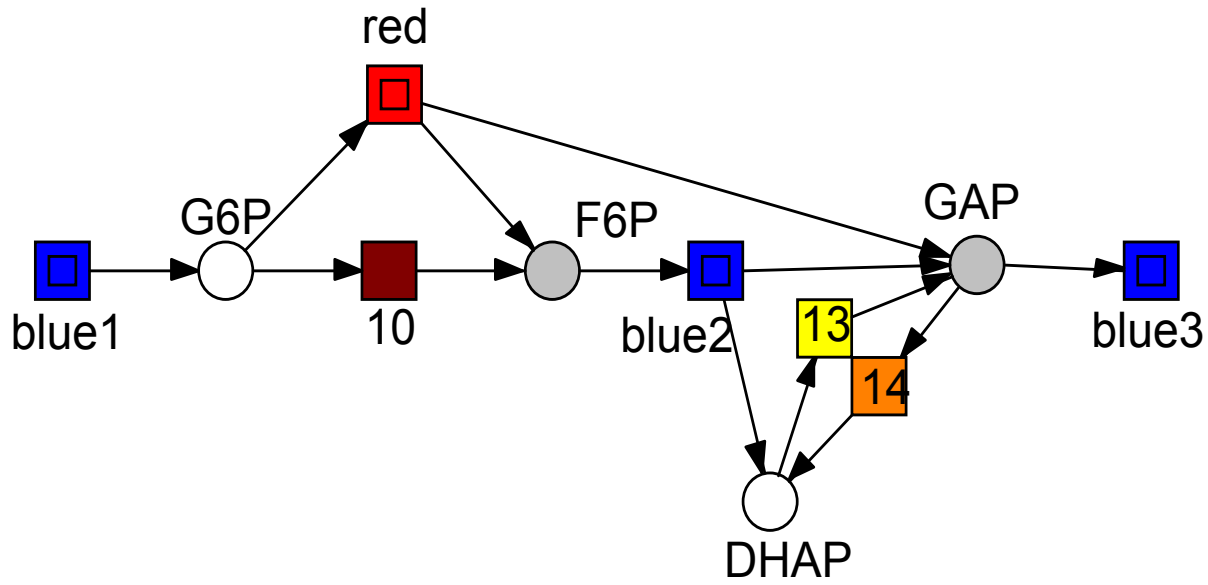
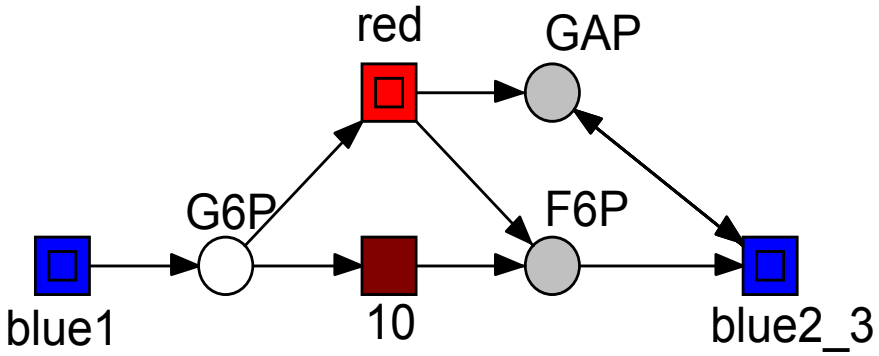
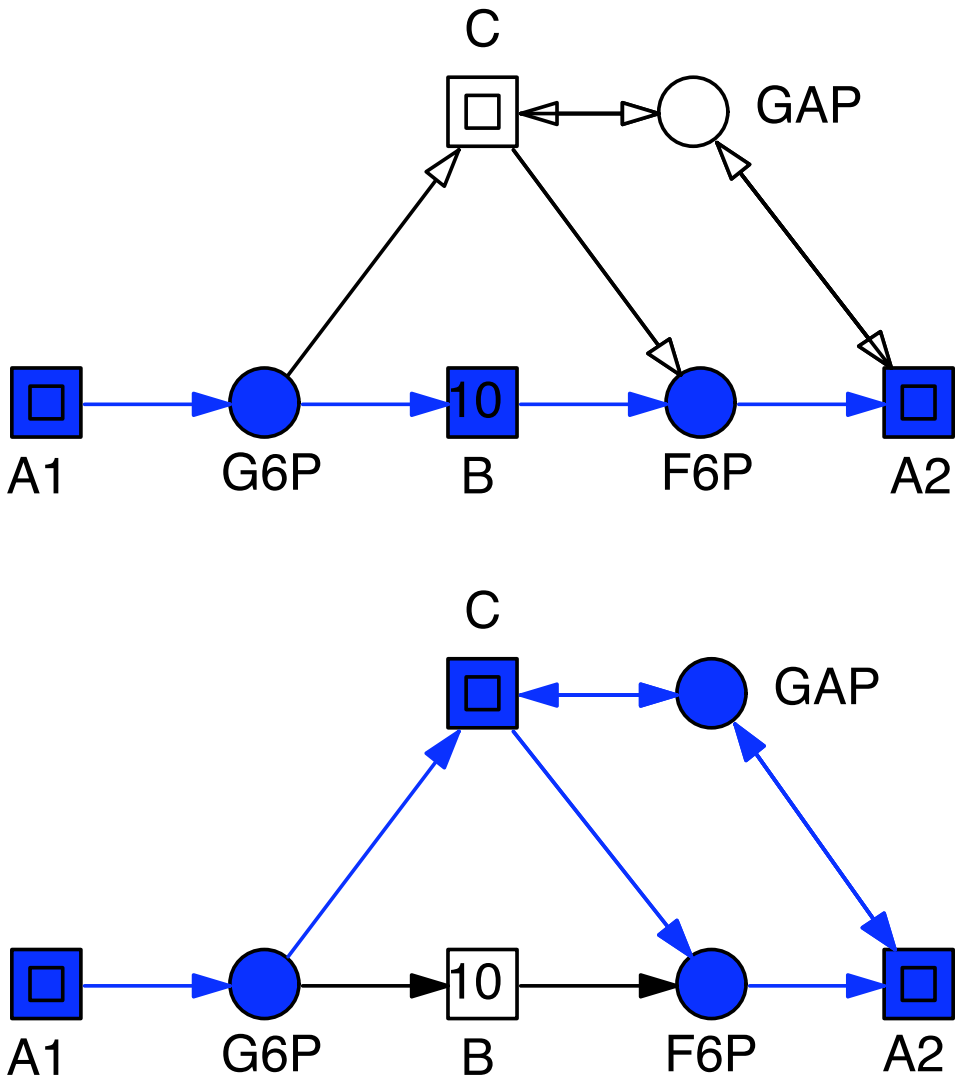
Ex1 - Glycolysis and Pentose Phosphate Pathway





Ex1 - Glycolysis and Pentose Phosphate Pathway



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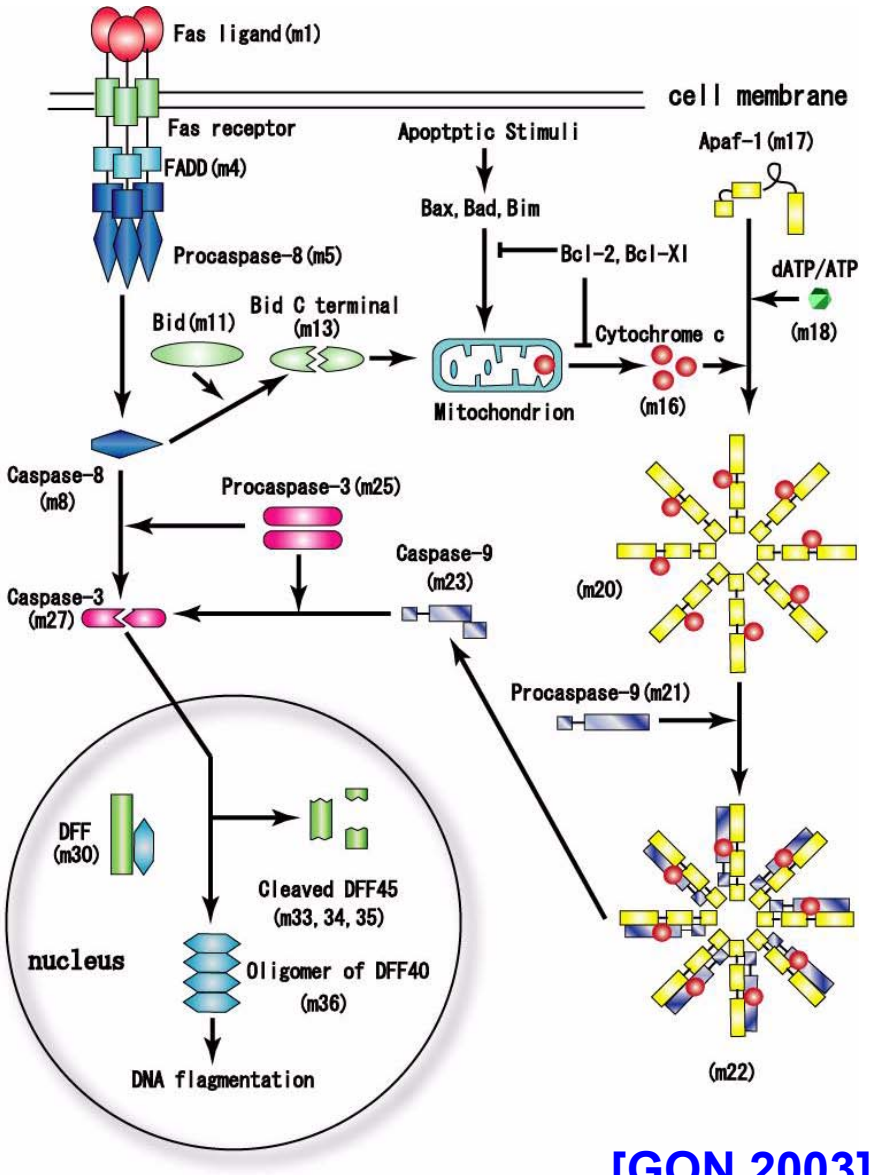
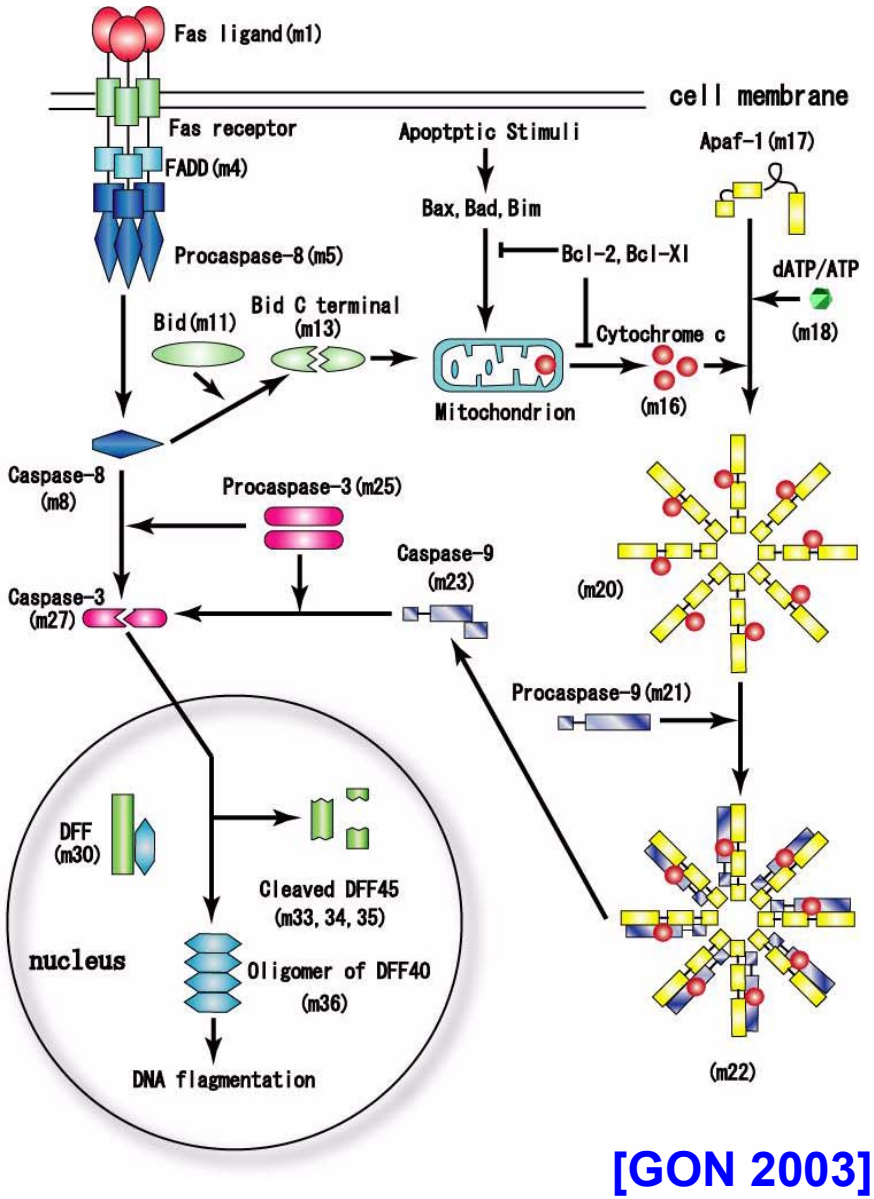
EX2: APOPTOSIS IN MAMMALIAN CELLS

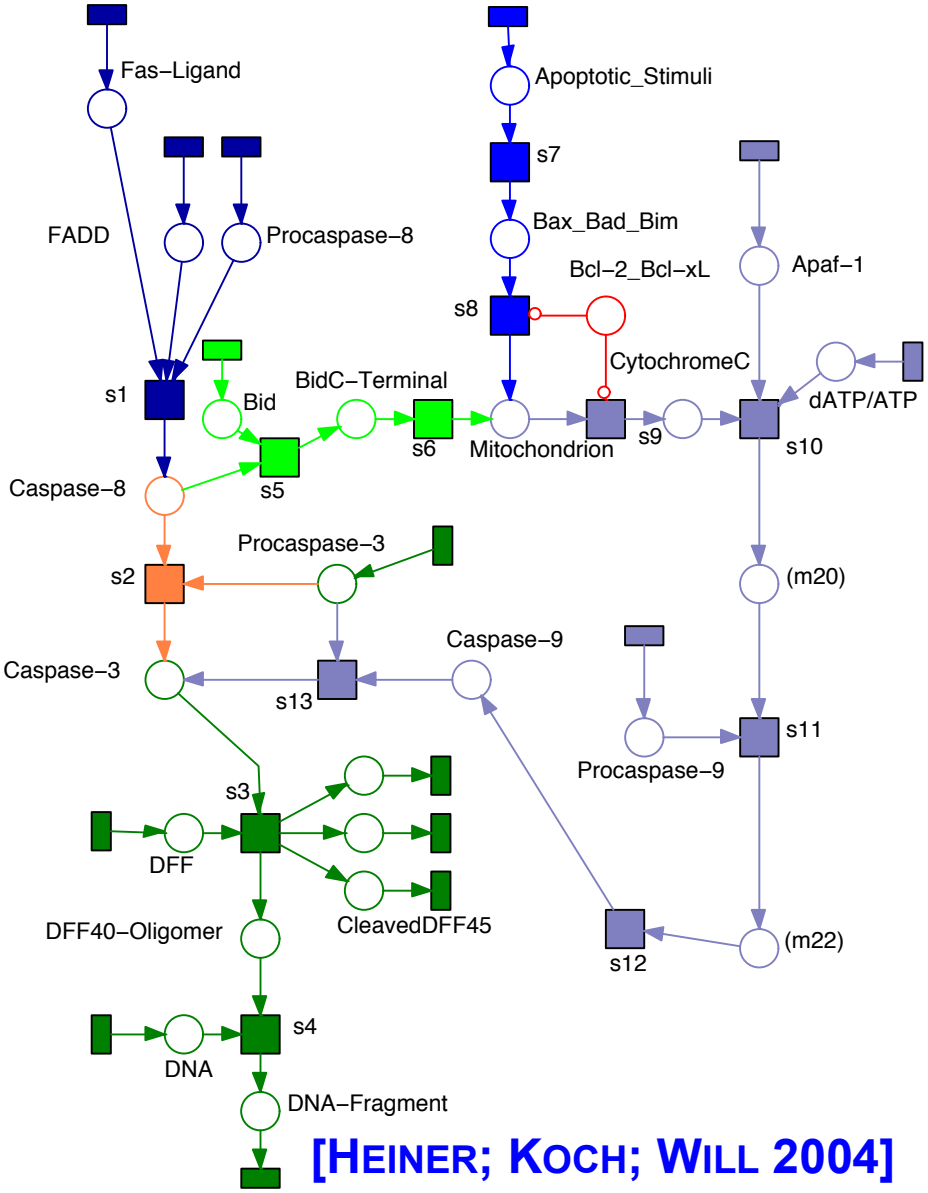


[GON 2003]

EX2: APOPTOSIS IN MAMMALIAN CELLS

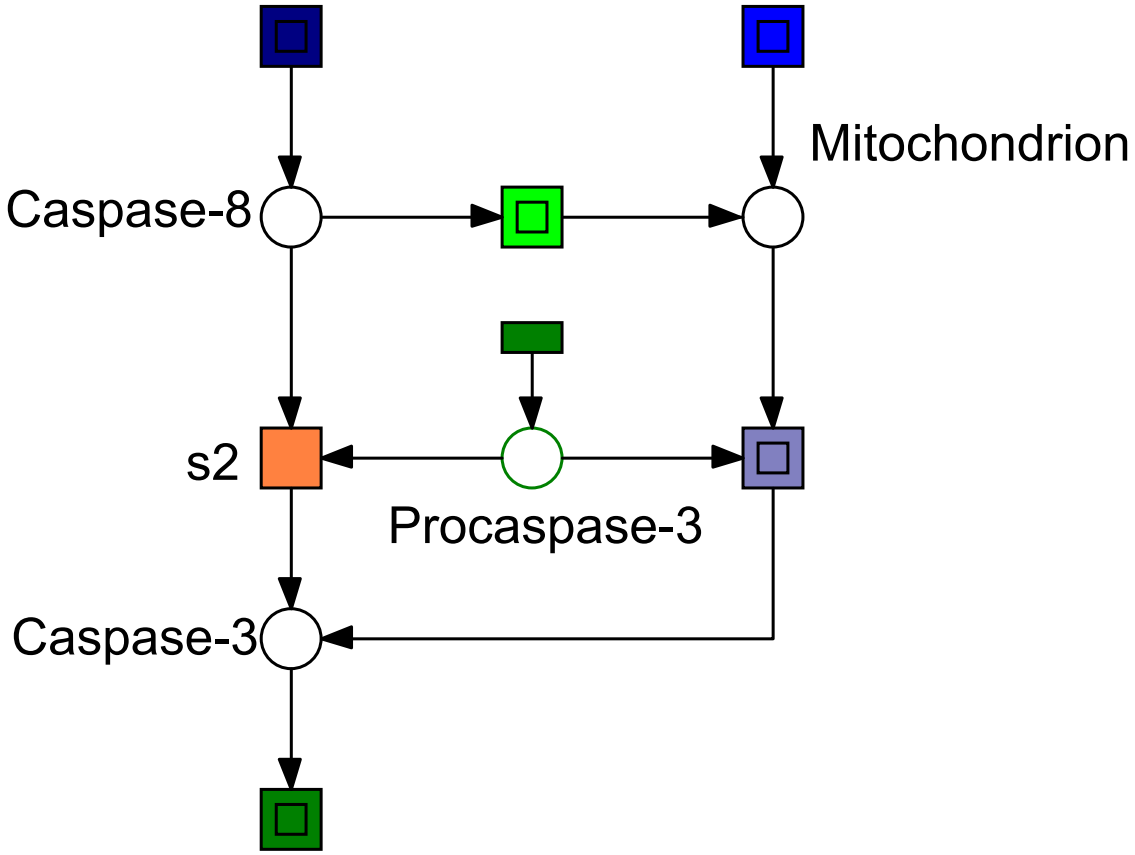


[GON 2003]

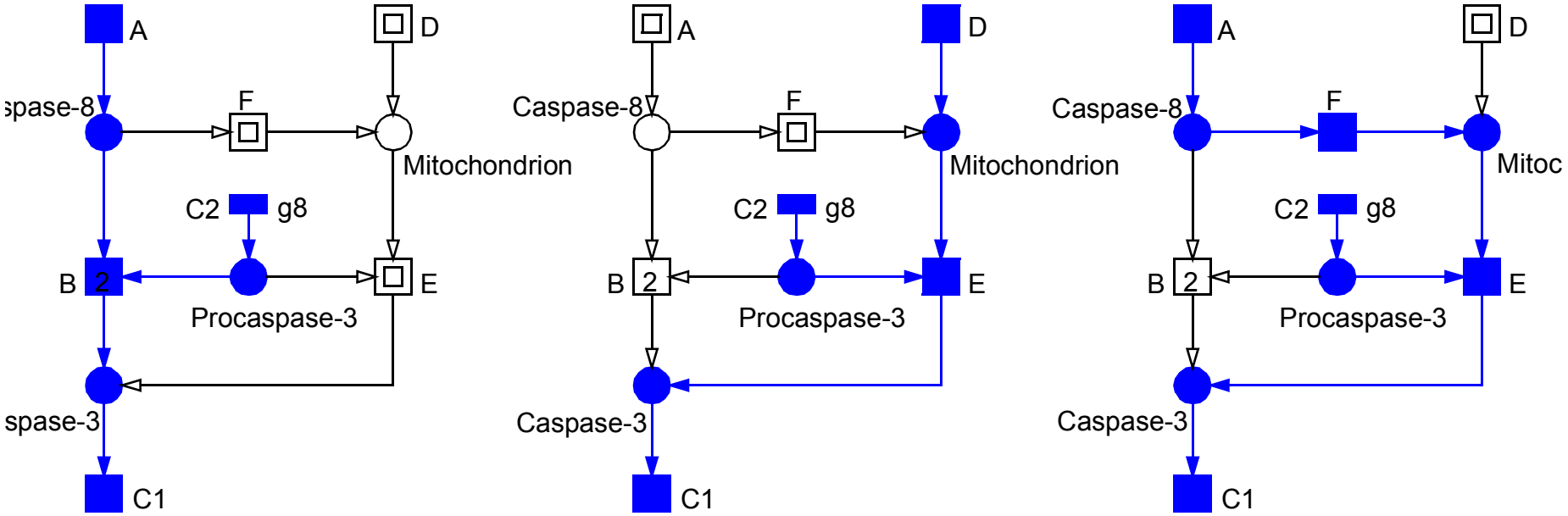


[HEINER; KOCH; WILL 2004]

EX2: APOPTOSIS IN MAMMALIAN CELLS



EX2: APOPTOSIS IN MAMMALIAN CELLS



Ex3 - Carbon Metabolism in Potato Tuber

[KOCH; JUNKER; HEINER 2005]

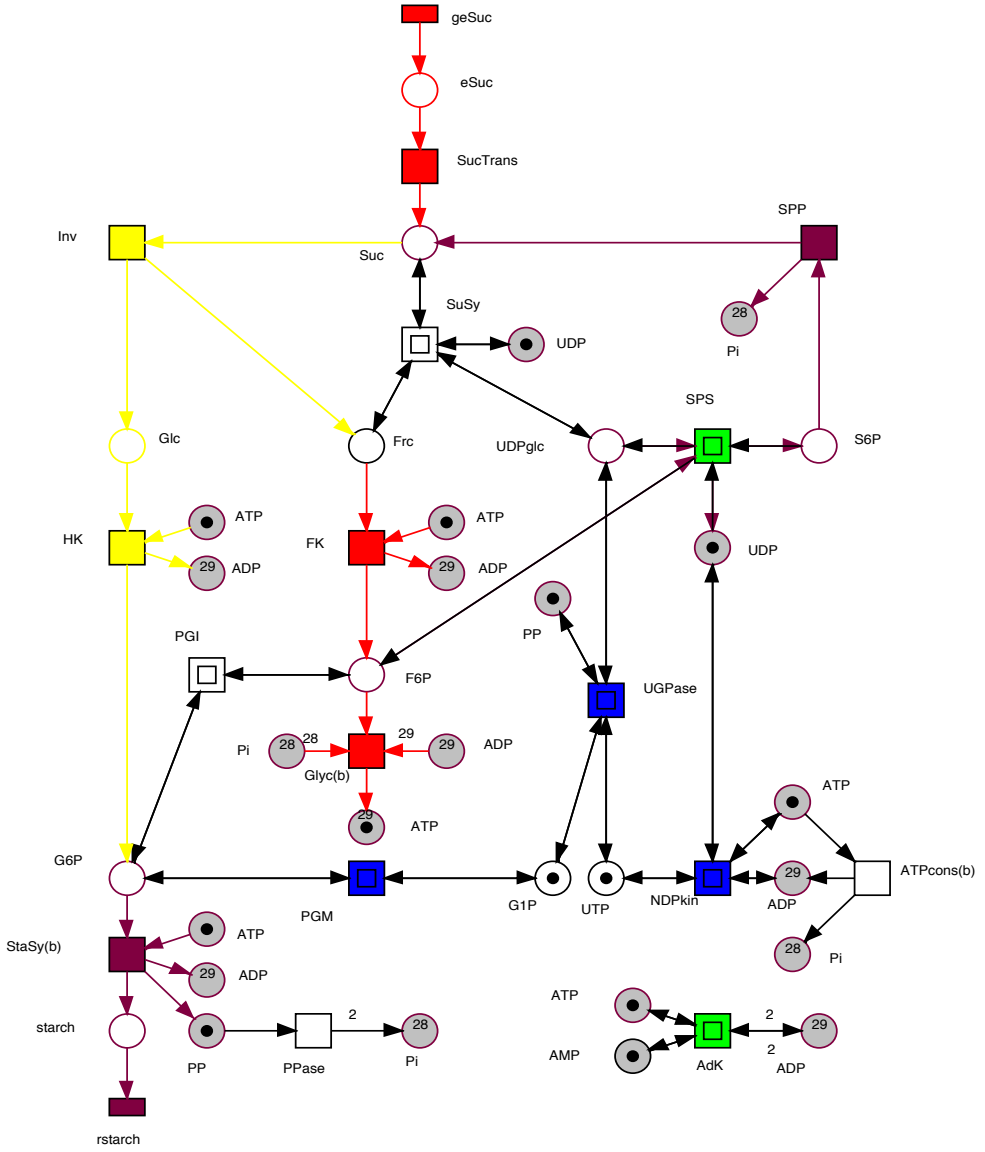
ADT-sets without trivial T-invariants

monika.heiner@informatik.tu-cottbus.de

Ex3 - Carbon Metabolism in Potato Tuber

[KOCH; JUNKER; HEINER 2005]

ADT-sets without trivial T-invariants



ADT-SETS, Interpretations

- **"promote hierarchical thinking & unbiased modularization"**
- **structured representation of invariants**
  -> *may contribute to a better understandability*
- **coarse network structure identifies sensitive net parts**
  -> *the knock-off of interface places affects several ADT-sets*
- **efficient design of wetlab experiments**
  -> *minimal sets of observation points providing coverage of the whole network (one for each ADT-set)*
- **support of dedicated layout algorithms**

"can include non-obvious groups of reactions and differ from groupings of reactions based on a visual inspection of the network topology"
(Papin, Reed, Palsson 2004)

ADT-SETS, SUMMARY

- **PROS**
  - -> *algorithmically defined*
  - -> *static analysis technique (state space not constructed), works also for unbounded models*

- **CONS**
  - -> *may be computational expensive*
  - -> *to avoid computation of all (T-) invariants:*

$$Cx = 0, x \neq 0, x \geq 0, \quad x(i) = 0, x(j) \neq 0, \forall i, j \in T$$

-- especially helpful for analyzing bio Petri nets --

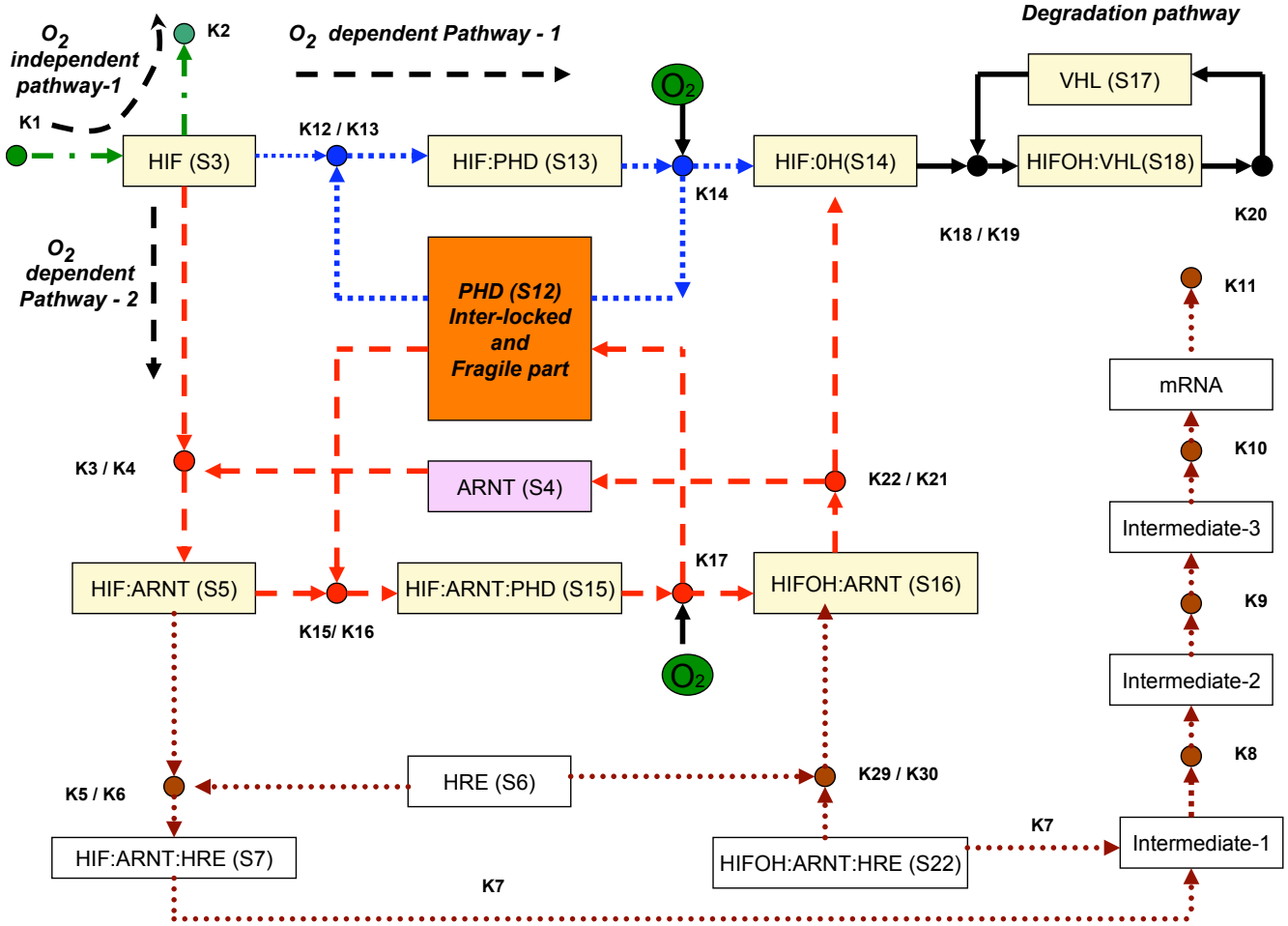
- **related work (T-invariants)**
  - -> *MCT-sets (Sackmann, Heiner, Koch 2006)*
  - -> *(A)DT-sets (Winder 2006)*
  - -> *partially correlated reaction sets (Papin, Reed, Palsson 2004)*
  - -> *Flux coupling analysis (Burgard 2004)*

CORE NETWORK IDENTIFICATION

monika.heiner@informatik.tu-cottbus.de

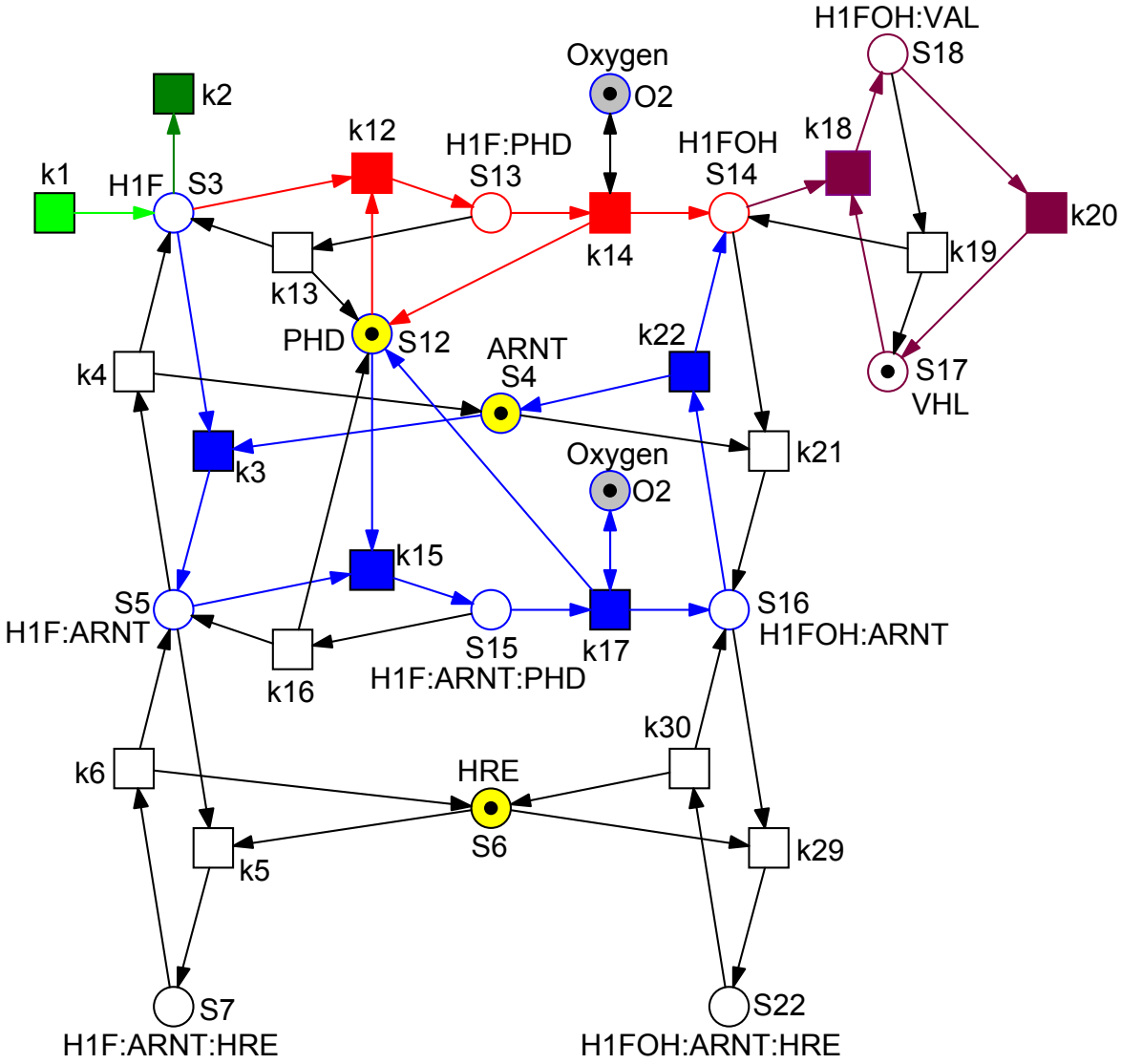
EX - HYPOXIA

[YU ET AL. 2007]



EX - HYPOXIA

[HEINER; SRIRAM 2010]



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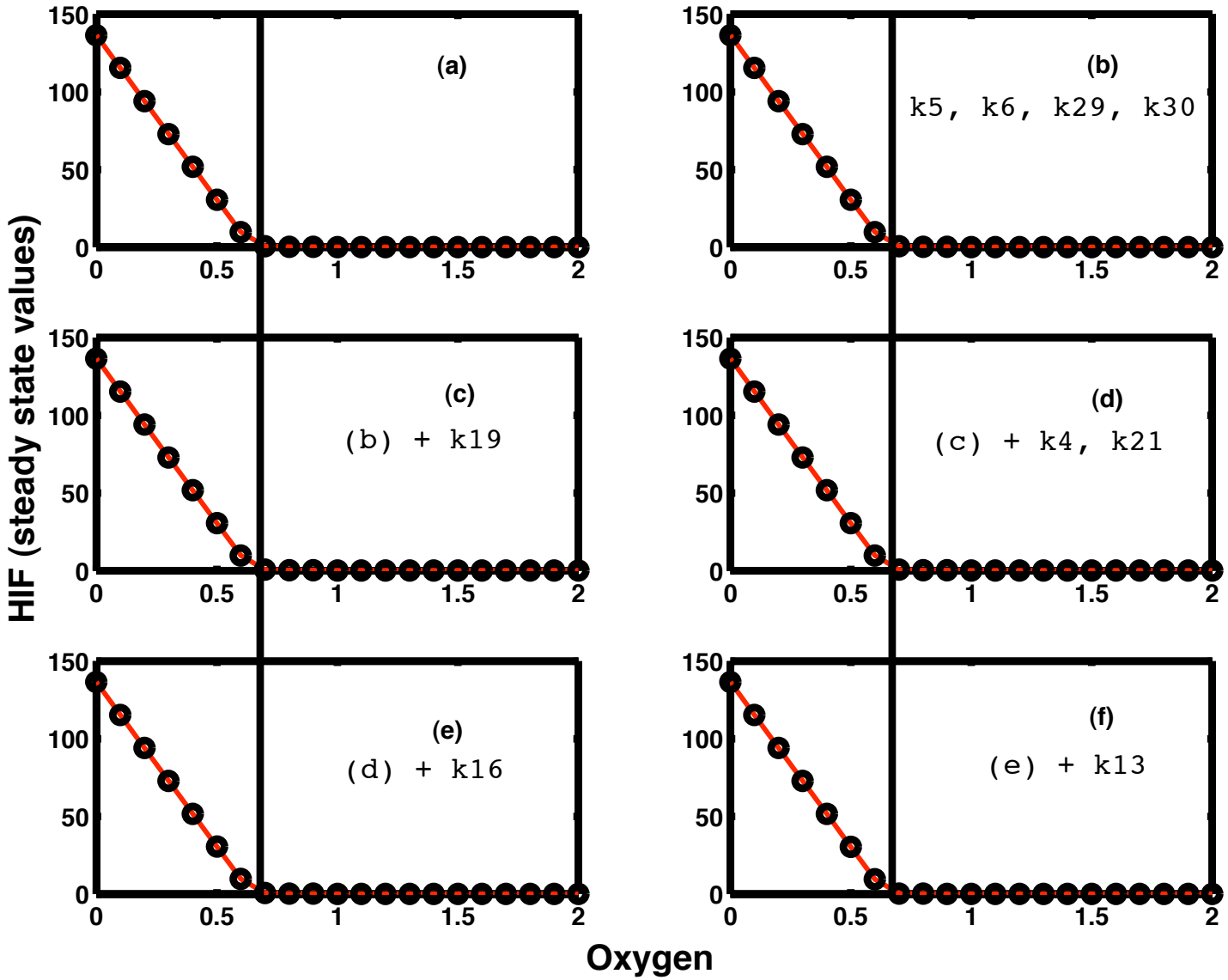
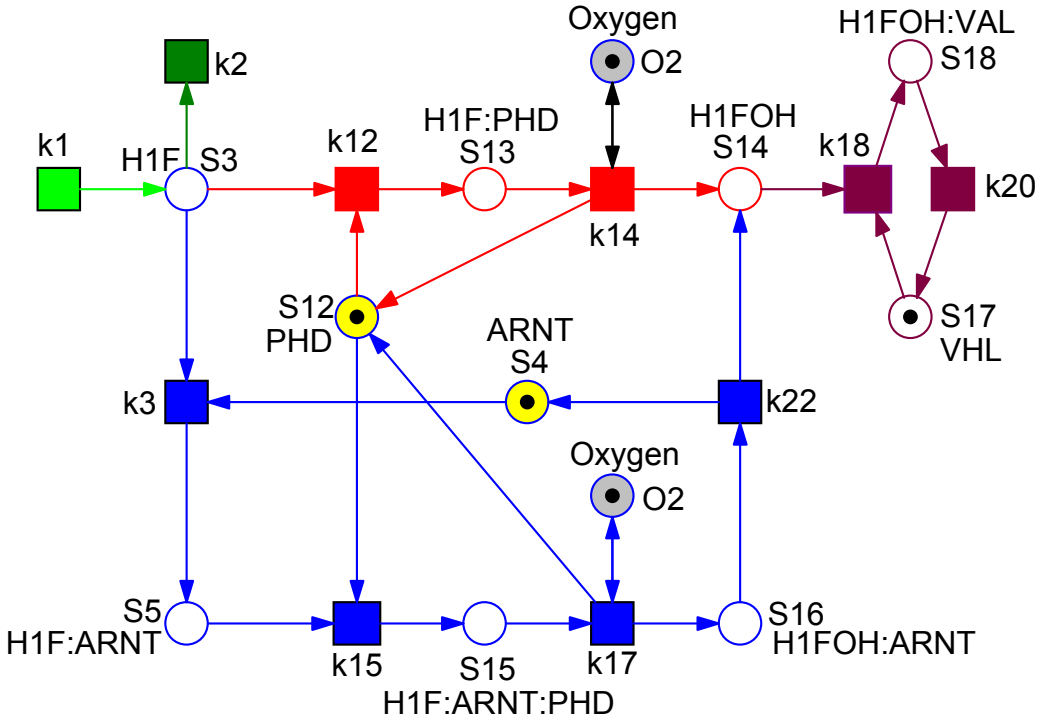
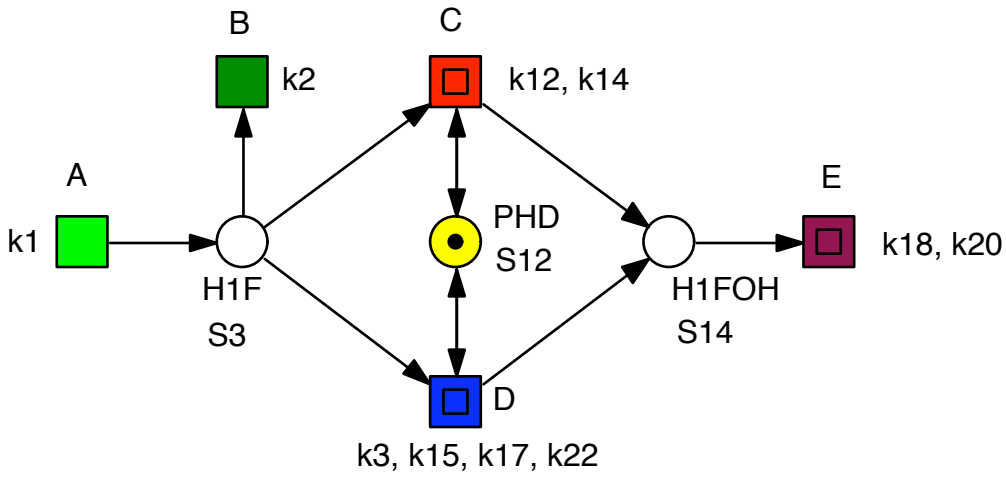
EX - HYPOXIA


HIF (steady state values)
Oxygen
(a)
(b)
k5, k6, k29, k30
(c)
(b) + k19
(d)
(c) + k4, k21
(e)
(d) + k16
(f)
(e) + k13
150
100
50
0
0
0.5
1
1.5
2

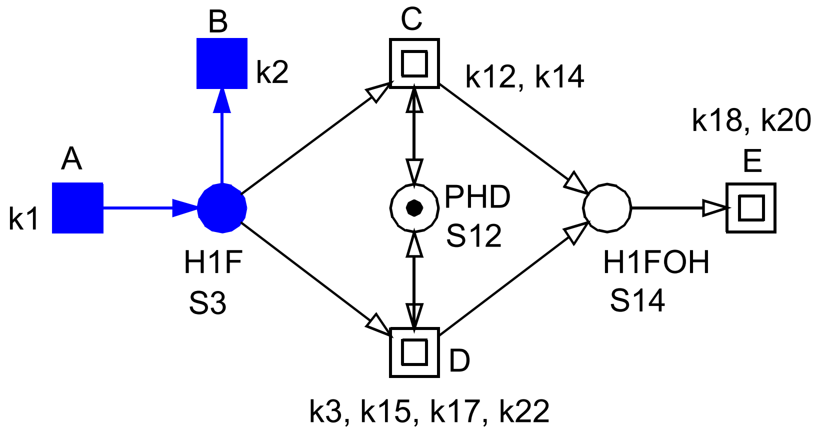
EX - HYPOXIA

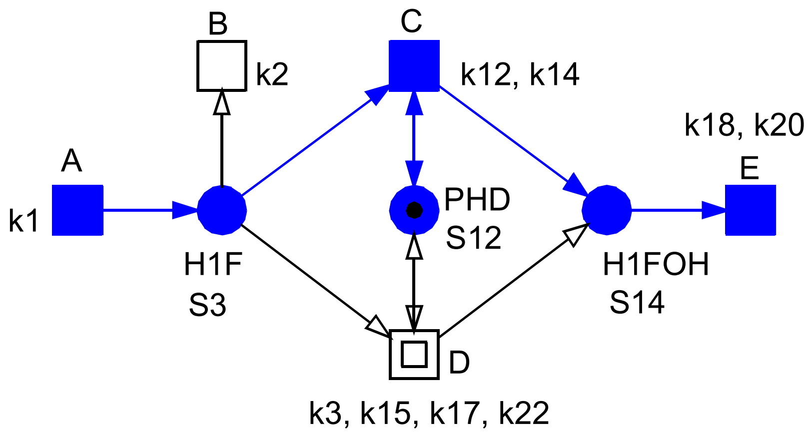

Oxygen
O2
H1FOH:VAL
S18
k2
k1
H1F
S3
k12
H1F:PHD
S13
H1FOH
S14
k18
k20
k14
S12
PHD
S17
VHL
ARNT
S4
k3
k22
Oxygen
O2
S5
H1F:ARNT
S16
H1FOH:ARNT
k15
S15
H1F:ARNT:PHD
k17

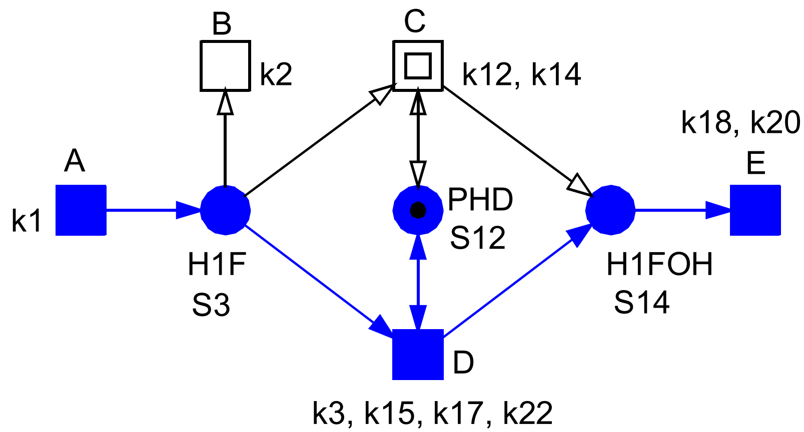
EX - HYPOXIA


B
C
k2
k12, k14
A
E
k1
PHD
S12
k18, k20
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22

EX - HYPOXIA


B
C
k2
k12, k14
k18, k20
A
E
k1
PHD
S12
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22


B
C
k2
k12, k14
k18, k20
A
E
k1
PHD
S12
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22


B
C
k2
k12, k14
k18, k20
A
E
k1
PHD
S12
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22

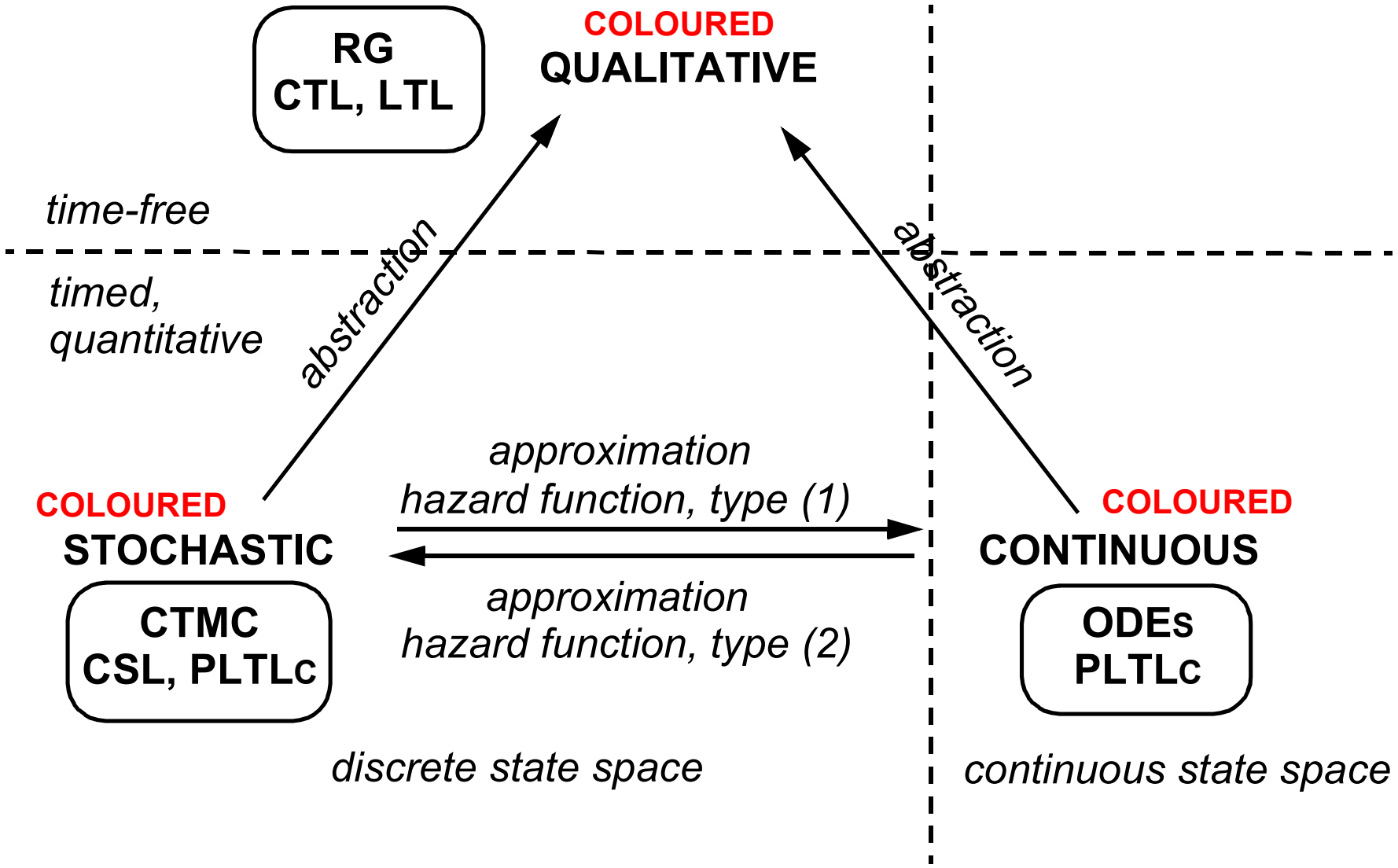
HOW TO GENERALIZE ?

LATEST NEWS

OUR TOOL BOX

- **Snoopy**
  - -> *in collaboration with Wolfgang Marwan,Magdeburg*
- **Charlie**
  - -> *inspired by INA*
  - -> *analysis tasks done by threads*
- **MARCIE**
  - -> ***M**odel checking **A**nd **R**eachability analysis done effi**CIE**ntly of qualitative and stochastic models*
  - -> *Interval Decision Diagrams*
  - -> *CTL model checking*
  - -> *"matrix free" transient and steady state analysis*
  - -> *full CSL model checking + rewards*
  - -> *distributed simulative PLTL model checking (pre-beta-version)*

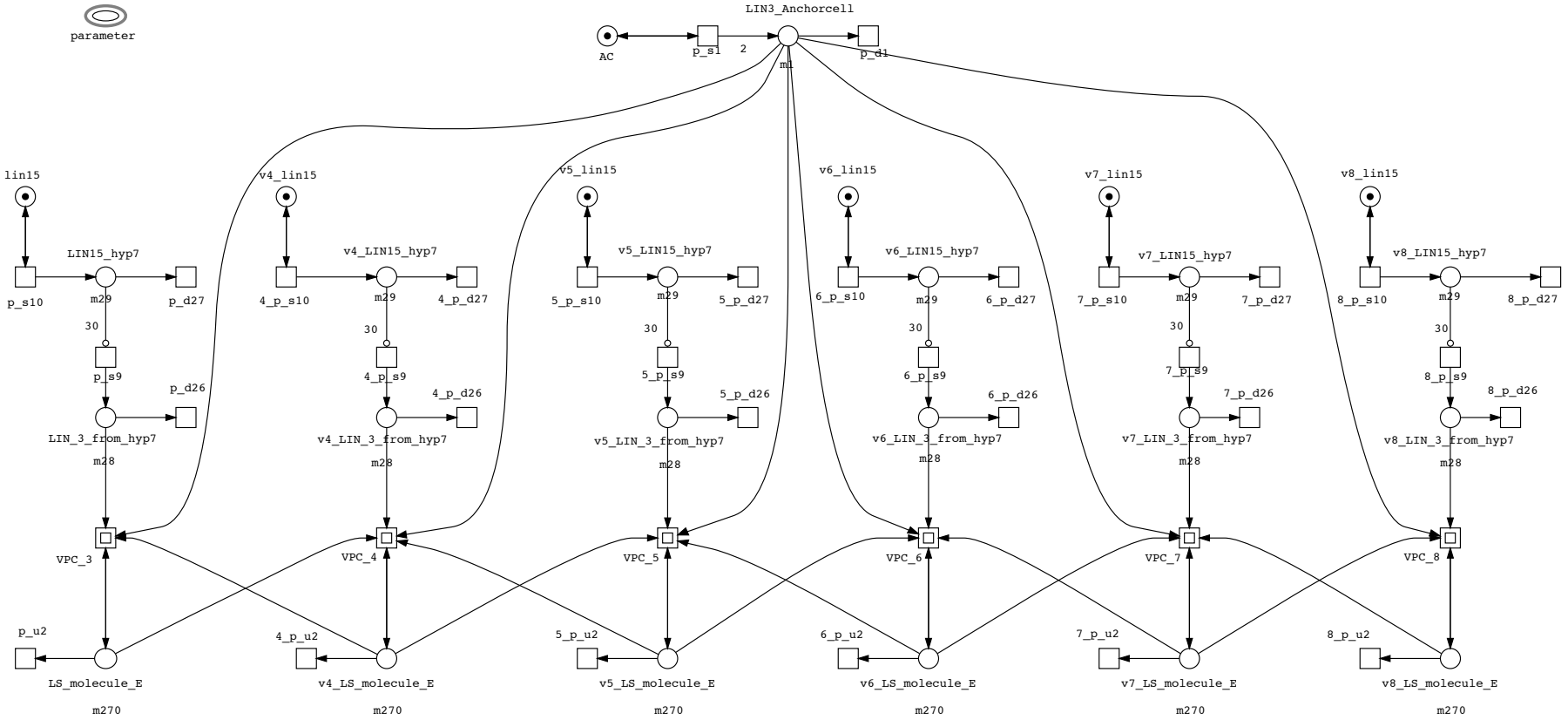
COLOURED FRAMEWORK



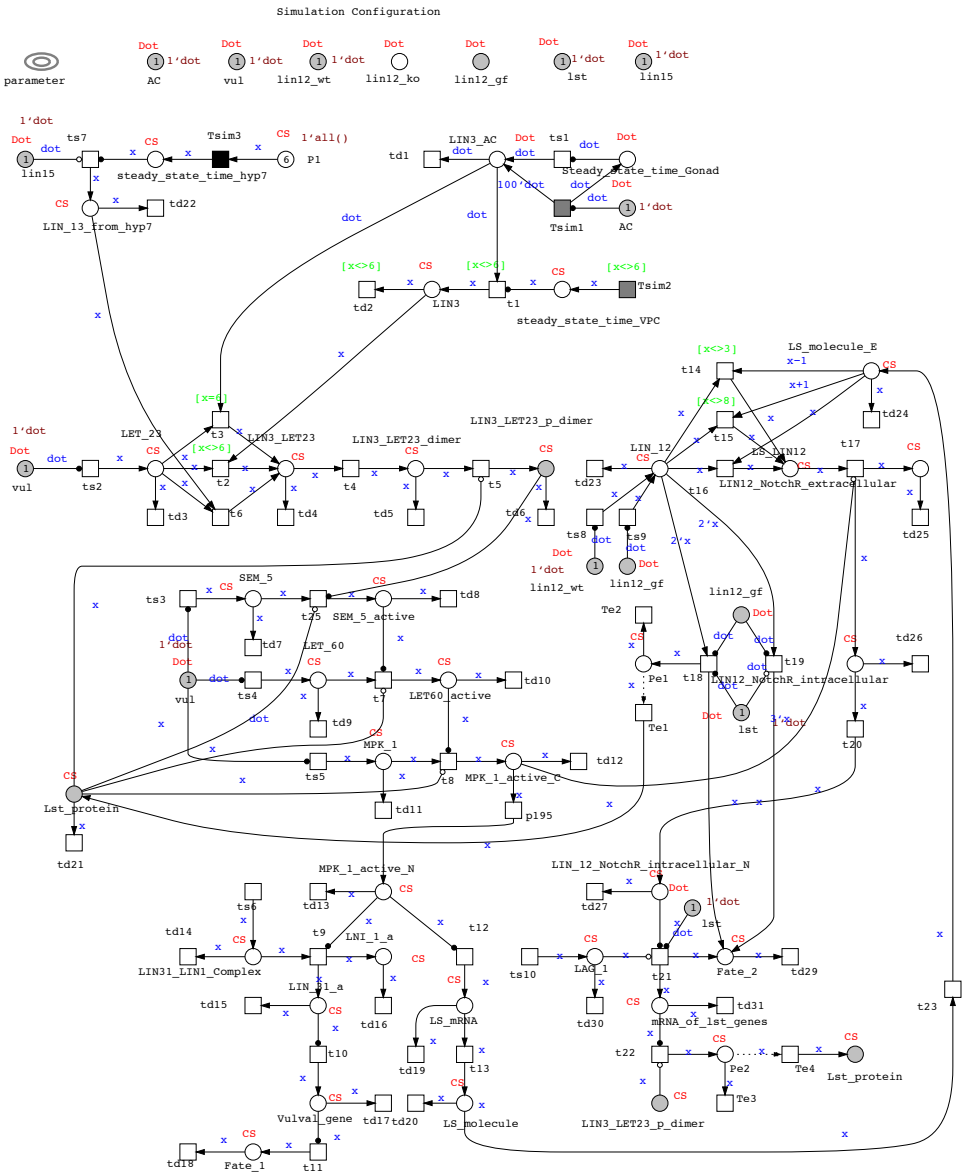
- **get multiple copies of patterns**
  - -> *Halo model, new order of net sizes*
- **differentiate between submodels within a master net**
  - -> *T-invariants*
  - -> *generated models in conformance with wet-lab data*
  - -> *mutants*
  - -> *algorithmic folding*
- **encode locality**
  - -> *Ca channel models*
  - -> *agents on a grid*
  - -> *. . .*
- **dynamic membrane systems**
- **. . .**

EX - C. ELEGANCE

[LI ET AL. 2009]
[BONZANNI ET AL. 2009]



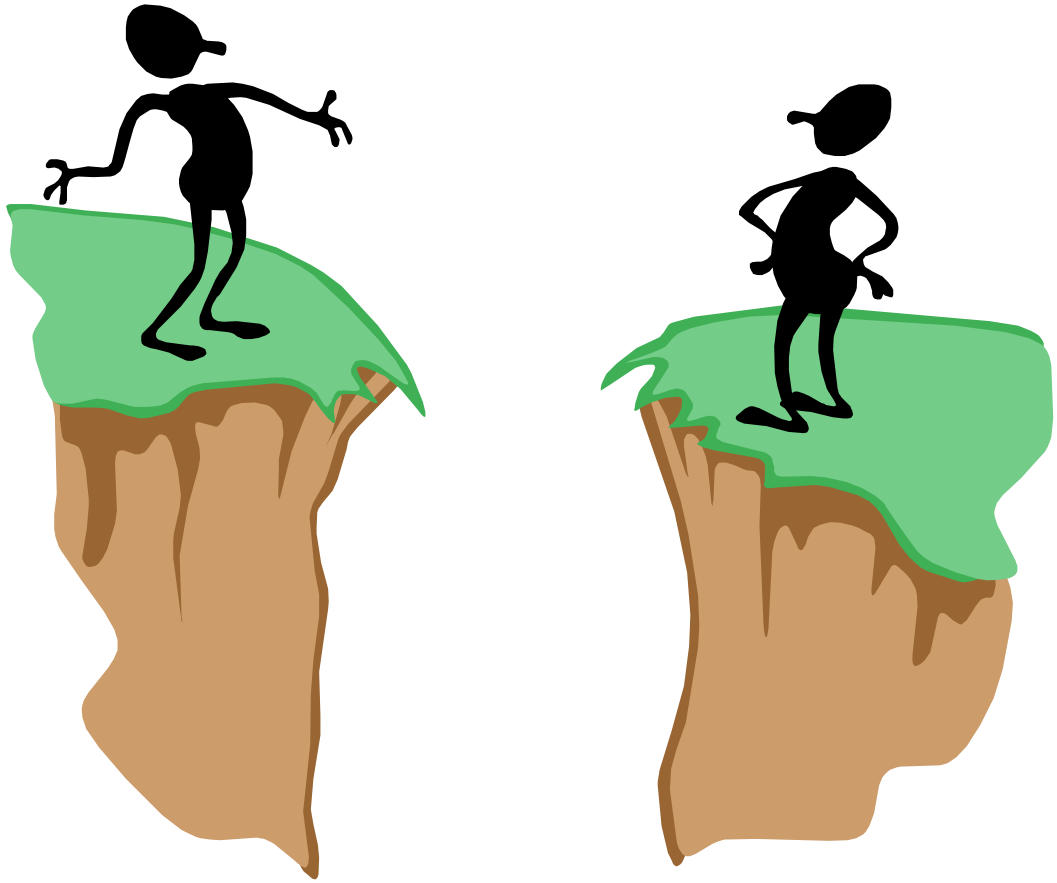
EX - C. ELEGANCE



GENERALIZED HYBRID PETRI NETS (GHPN)

- **Extended Generalized Stochastic Petri Nets (XSPN)**
  - -> *discrete places*
  - -> *discrete transitions: stochastic, immediate, deterministically delayed, scheduled*
  - -> *special arcs: read, inhibitor, equal, reset*
- **Continuous Petri Nets (CPN)**
  - -> *continuous places*
  - -> *continuous transitions*
  - -> *special arcs: read, inhibitor*
- **GHPN = XSPN + CPN**
- **hybrid simulation engine**
- **dynamic partitioning**

OUTLOOK



THANKS !

HHTP://WWW-DSSZ.INFORMATIK.TU-COTTBUS.DE