BIOMODEL ENGINEERING
- A PETRI NET PERSPECTIVE -

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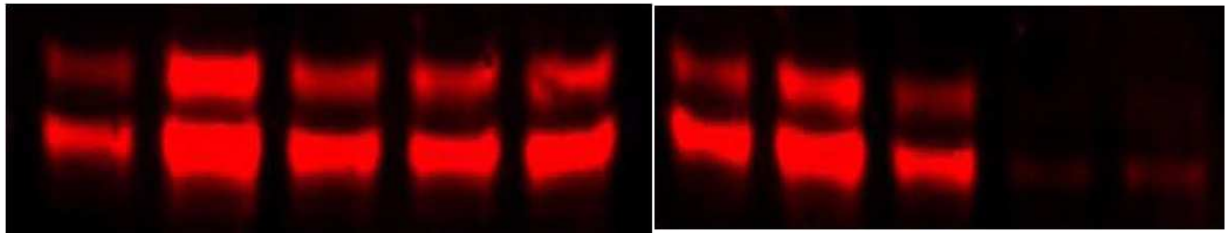
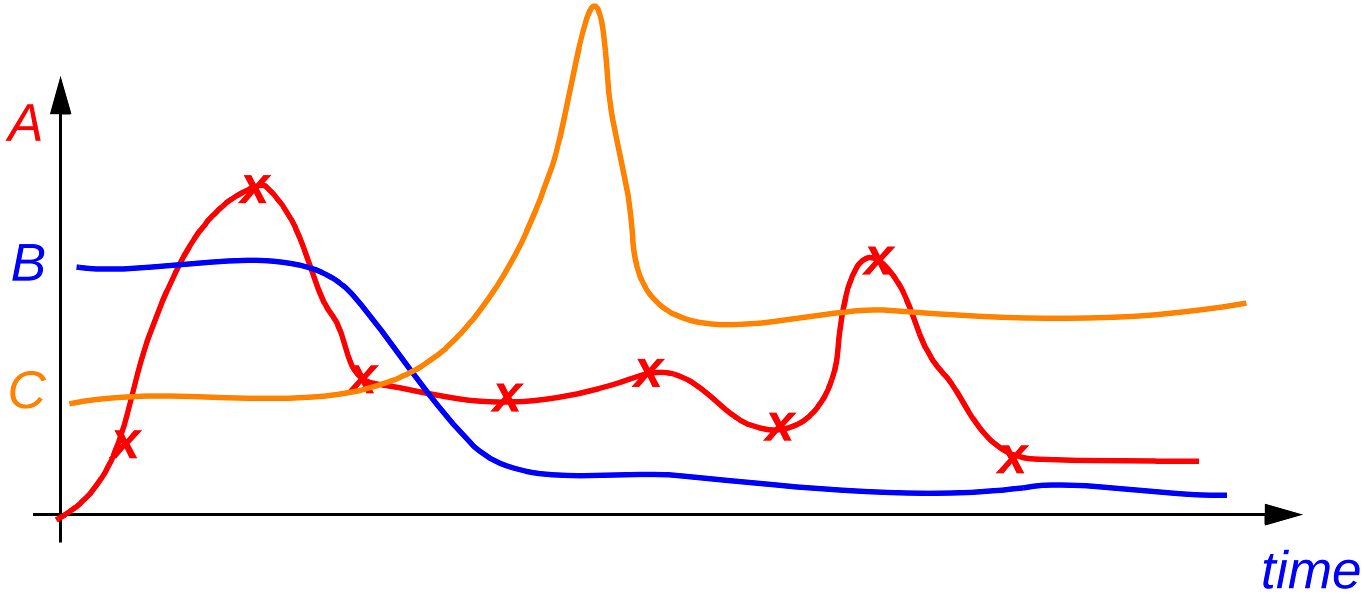



A
B
C
time

- **Qualitative**

  Protein rises then falls

- **Semi-qualitative**

  Protein rises then falls to less than 50% of its peak concentration

- **Semi-quantitative**

  Protein rises then falls to less than 50% of its peak concentration at 60 minutes

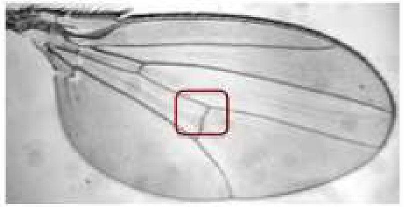
- **Quantitative**

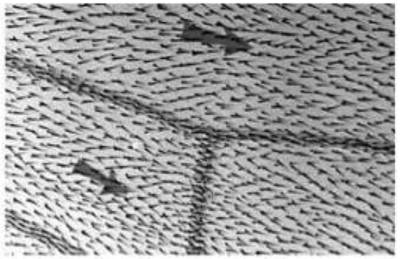
  Protein rises then falls to less than 100 microMol at 60 minutes

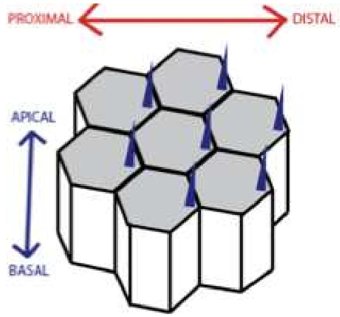
-> networks explaining the observations ?

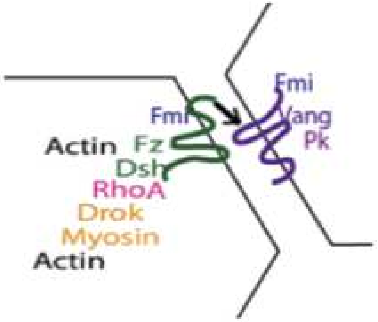
OUTLINE

- **BACKGROUND**
  - -> *modelling, what for ?*
  - -> *how many model types do we need ?*
- **FRAMEWORK**
  - -> *unifying four paradigms: QPN - SPN - CPN - HPN*
  - -> *some case studies*
  - -> *moving between paradigms*
- **COLOUR AND MUTLI-SCALE SYSTEM**
  - -> *replication*
  - -> *encoding space*
- **SUMMARY & OUTLOOK**
  - -> *open problems*
  - -> *next steps*



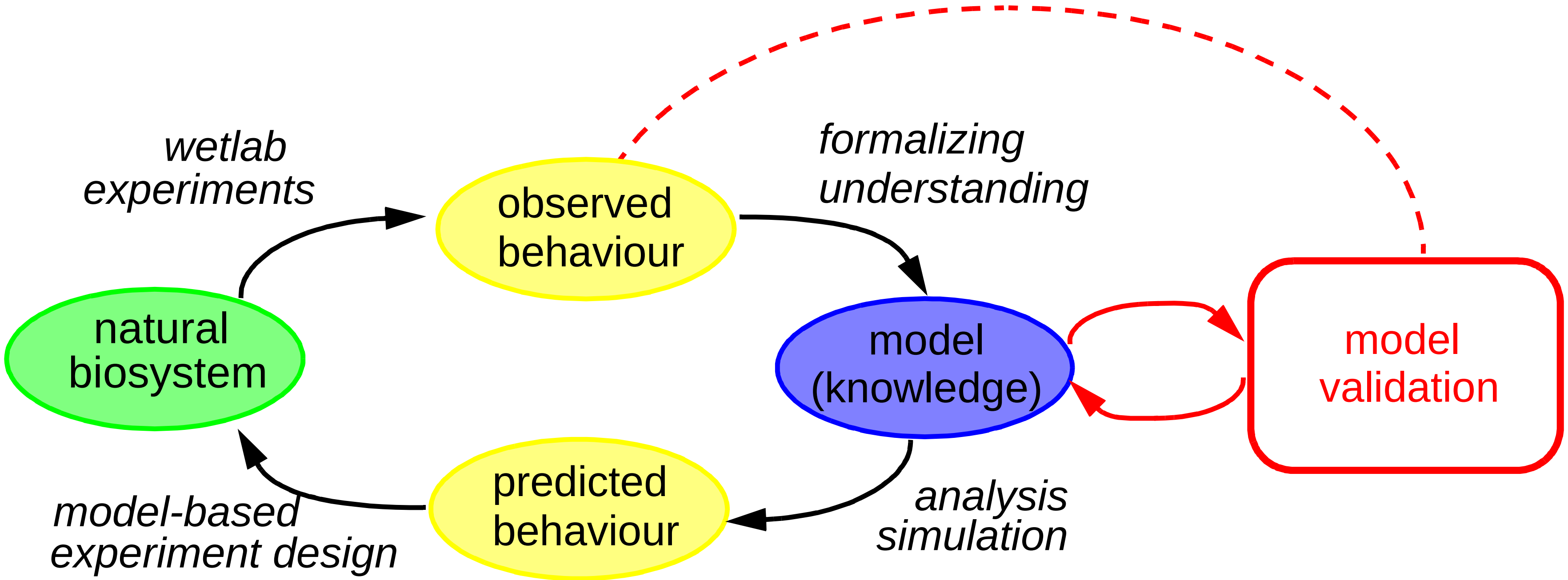






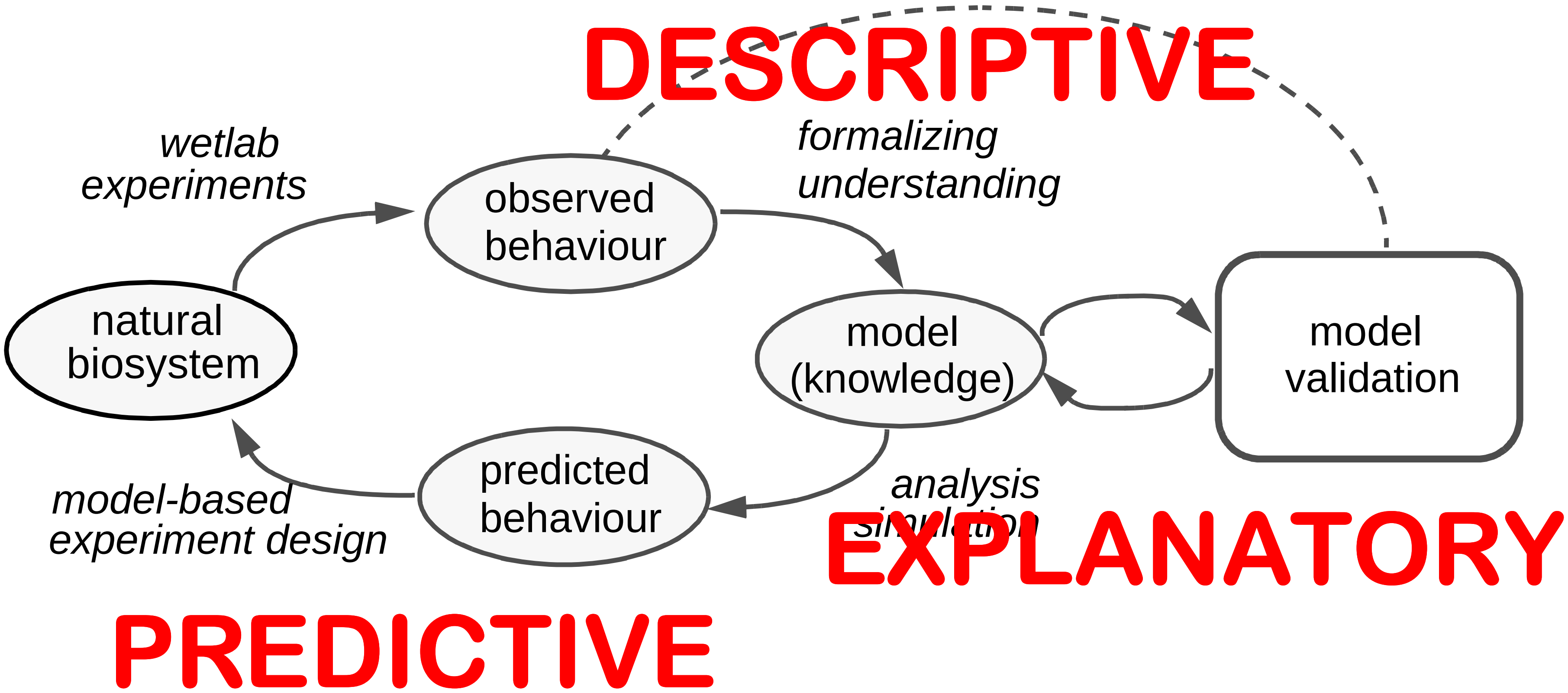
BACKGROUND

MODELLING = FORMAL KNOWLEDGE REPRESENTATION



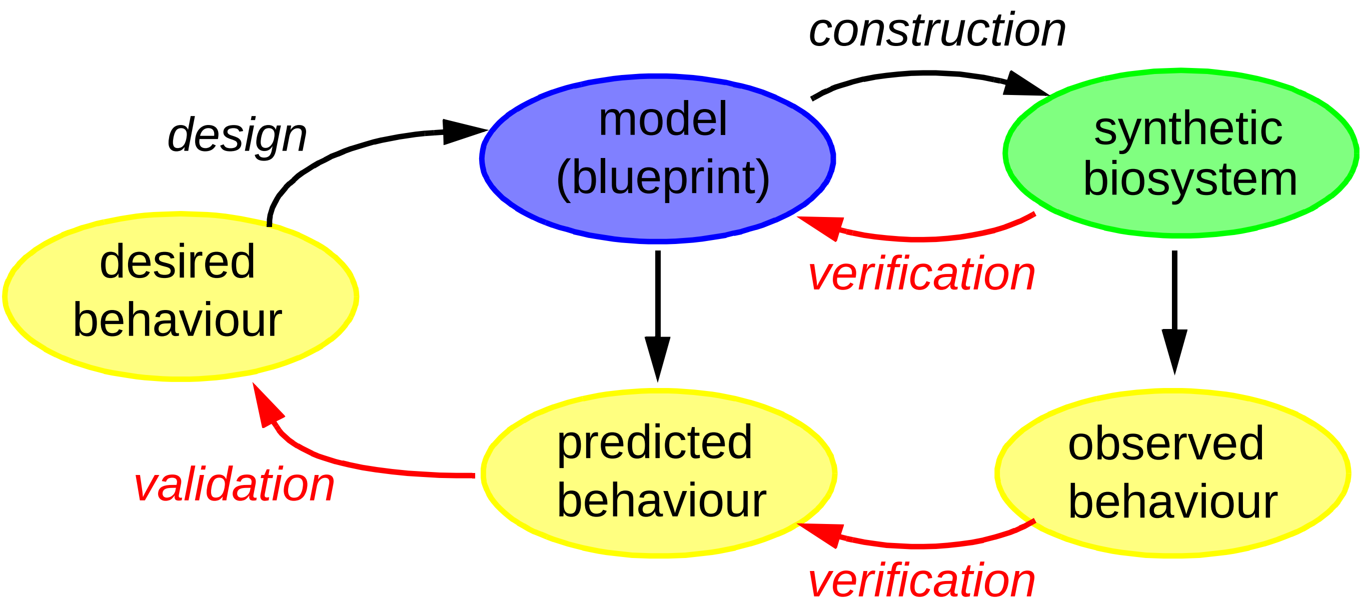
MODEL VALIDATION = CONFIDENCE INCREASE

MODELLING = FORMAL KNOWLEDGE REPRESENTATION



MODEL VALIDATION = CONFIDENCE INCREASE

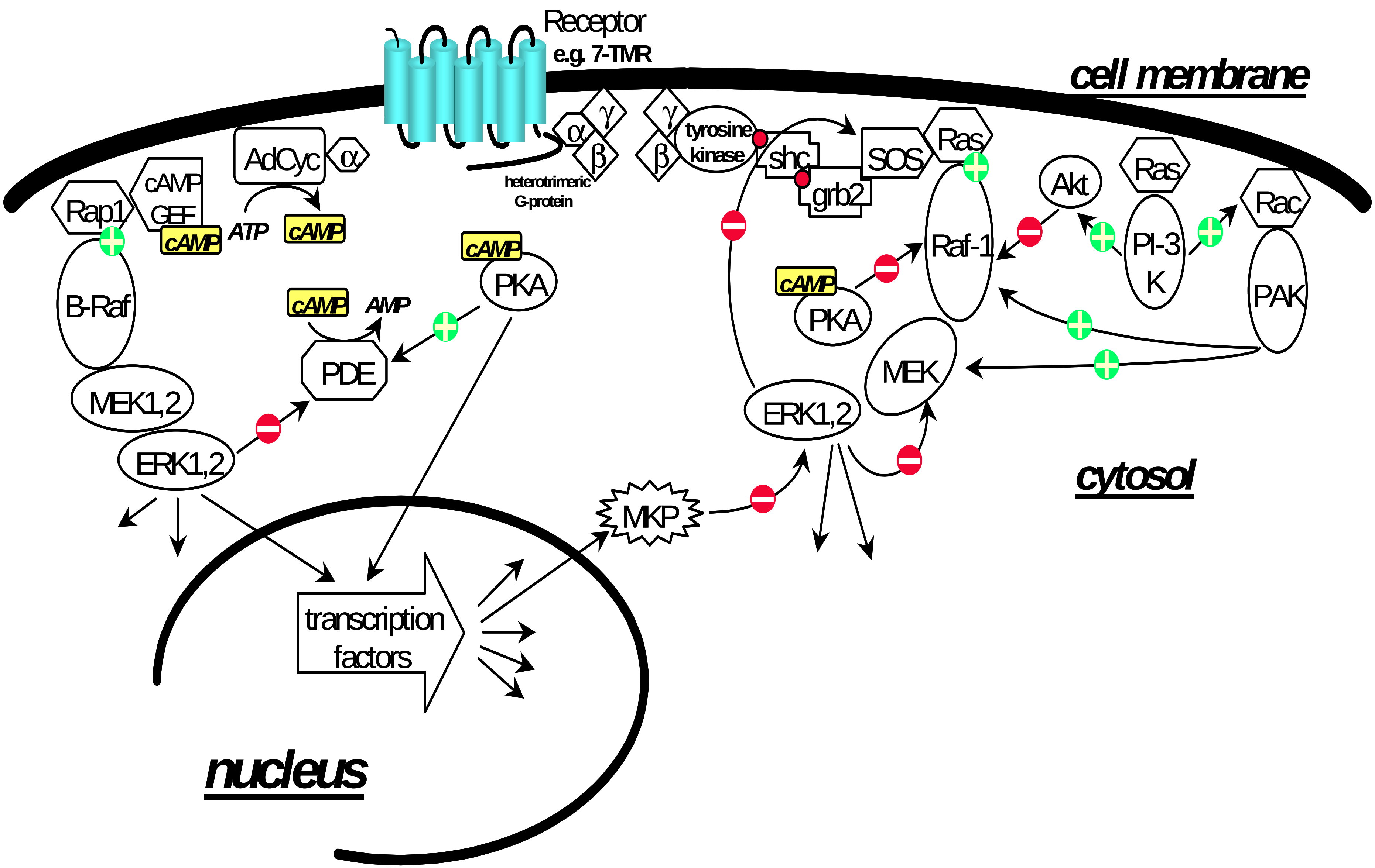
MODELLING = BLUEPRINT FOR SYSTEM CONSTRUCTION



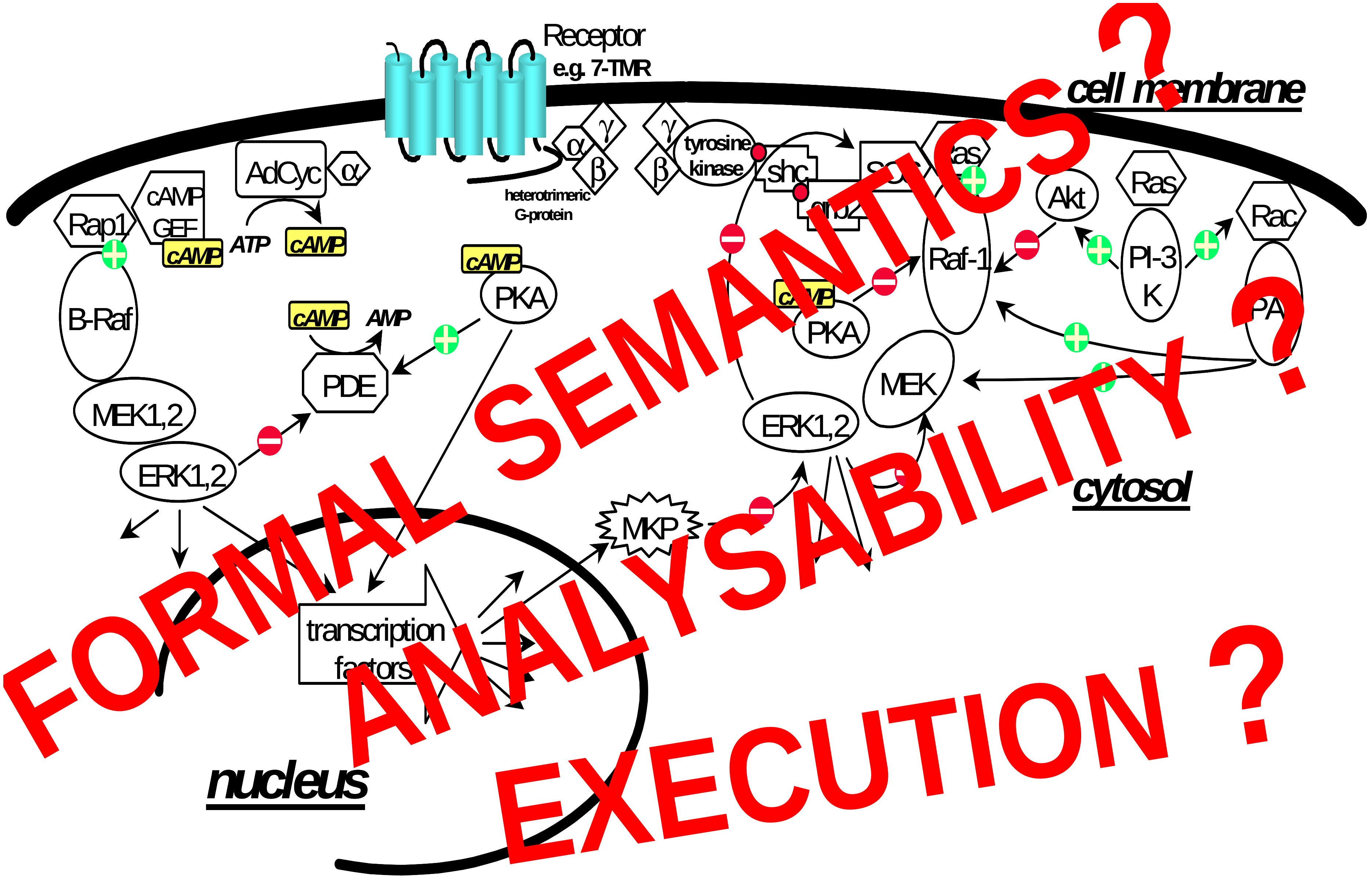
RELIABLE AND ROBUST ENGINEERING REQUIRES VERIFIED MODELS

WHAT KIND OF MODEL SHOULD BE USED?

(BIOCHEMICAL NETWORKS)



Receptor
e.g. 7-TMR
cell membrane
heterotrimeric
G-protein
α
β
γ
AdCyc
α
cAMP
GEF
Rap1
cAMP
ATP
cAMP
B-Raf
cAMP
PKA
cAMP
AMP
PDE
MEK1,2
ERK1,2
tyrosine
kinase
shc
grb2
SOS
Ras
Raf-1
Akt
Ras
PI-3
K
Rac
PAK
cAMP
PKA
MEK
ERK1,2
cytosol
MKP
transcription
factors
nucleus


Receptor
e.g. 7-TMR
cell membrane
α
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heterotrimeric
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kinase
shc
Akt
Ras
Rac
Raf-1
PI-3
K
PKA
cAMP
MEK
ERK1,2
MKP
cytosol
Rap1
cAMP
GEF
AdCyc
ATP
AMP
PDE
B-Raf
MEK1,2
transcription
factors
nucleus
FORMAL SEMANTICS ?
ANALYSABILITY ?
EXECUTION ?

$$\frac{d\alpha}{dt} = -\nu_1$$
$$\frac{dSte2}{dt} = -\nu_2 + \nu_3 - \nu_5$$
$$\frac{dSte2_{active}}{dt} = \nu_2 - \nu_3 - \nu_4$$
$$\frac{dSst2_{active}}{dt} = \nu_{46} - \nu_{47}$$
$$\frac{dG\alpha\beta\gamma}{dt} = -\nu_6 + \nu_9$$
$$\frac{dG\alpha GTP}{dt} = \nu_6 - \nu_7 - \nu_8$$
$$\frac{dG\alpha GDP}{dt} = \nu_7 + \nu_8 - \nu_9$$
$$\frac{dG\beta\gamma}{dt} = \nu_6 - \nu_9 - \nu_{10} + \nu_{11} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32} - \nu_{42} + \nu_{43}$$
$$\frac{dSte5}{dt} = -\nu_{12} + \nu_{13} + \nu_{17} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32}$$
$$\frac{dSte11}{dt} = -\nu_{12} + \nu_{13} + \nu_{17} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32}$$
$$\frac{dSte7}{dt} = -\nu_{14} + \nu_{15} + \nu_{17} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32}$$
$$\frac{dFus3}{dt} = -\nu_{14} + \nu_{15} + \nu_{17} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} - \nu_{29} + \nu_{30} + \nu_{33}$$
$$\frac{dSte20}{dt} = -\nu_{18} + \nu_{19} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32}$$

$$\nu_1 = \alpha[t] \cdot Bar1_{active}[t] \cdot k_1$$
$$\nu_2 = Ste2[t] \cdot \alpha[t] \cdot k_2$$
$$\nu_3 = Ste2_{active}[t] \cdot k_3$$
$$\nu_4 = Ste2_{active}[t] \cdot k_4$$
$$\nu_5 = Ste2[t] \cdot k_5$$
$$\nu_6 = Ste2_{active}[t] \cdot G\alpha\beta\gamma[t] \cdot k_6$$
$$\nu_7 = G\alpha GTP[t] \cdot k_7$$
$$\nu_8 = G\alpha GTP[t] \cdot Sst2_{active}[t] \cdot k_8$$
$$\nu_9 = G\alpha GDP[t] \cdot G\beta\gamma[t] \cdot k_9$$
$$\nu_{10} = G\beta\gamma[t] \cdot C[t] \cdot k_{10}$$
$$\nu_{11} = D[t] \cdot k_{11}$$
$$\nu_{12} = Ste5[t] \cdot Ste11[t] \cdot k_{12}$$
$$\nu_{13} = A[t] \cdot k_{13}$$
$$\nu_{14} = Ste7[t] \cdot Fus3[t] \cdot k_{14}$$
$$\nu_{15} = B[t] \cdot k_{15}$$
$$\nu_{16} = A[t] \cdot B[t] \cdot k_{16}$$
$$\nu_{17} = C[t] \cdot k_{17}$$
$$\nu_{18} = D[t] \cdot Ste20[t] \cdot k_{18}$$

NETWORK REPRESENTATIONS, EX2

$$\frac{d\alpha}{dt} = -\nu_1$$
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$$\frac{dG\beta\gamma}{dt} = \nu_6 - \nu_9 - \nu_{[illegible]} + \nu_{[illegible]} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32} - \nu_{42} + \nu_{43}$$
$$\frac{dSte5}{dt} = -\nu_{12} + \nu_{13} + \nu_{17} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32}$$
$$\frac{dSte11}{dt} = -\nu_{12} + \nu_{13} + \nu_{17} + \nu_{21} + \nu_{2[illegible]} + \nu_{25} + \nu_{27} + \nu_{32}$$
$$\frac{d[illegible]}{dt} = -\nu_{14} + \nu_{15} + \nu_{17} + \nu_{2[illegible]} + \nu_{23} + \nu_{25} + \nu_{27} + \nu_{32}$$
$$\frac{dFus3}{dt} = -\nu_{14} + \nu_{15} + \nu_{17} + \nu_{21} + \nu_{23} + \nu_{25} + \nu_{27} - \nu_{29} + \nu_{30} + \nu_{33}$$
$$\frac{dSte20}{dt} = -\nu_{18} + \nu_{[illegible]} - \nu_{[illegible]} + \nu_{23} + [illegible] + \nu_{27} + \nu_{32}$$

$$\nu_1 = \alpha[t] \cdot Bar1_{active}[t] \cdot k_1$$
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$$\nu_{10} = G\beta\gamma[t] \cdot C[t] \cdot k_{10}$$
$$\nu_{11} = D[t] \cdot k_{11}$$
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$$\nu_{13} = A[t] \cdot k_{13}$$
$$\nu_{14} = Ste7[t] \cdot Fus3[t] \cdot k_{14}$$
$$\nu_{15} = B[t] \cdot k_{15}$$
$$\nu_{16} = [illegible][t] \cdot E[t] \cdot k_{16}$$
$$\nu_{17} = C[t] \cdot k_{17}$$
$$\nu_{18} = D[t] \cdot Ste20[t] \cdot k_{18}$$

READABILITY ?

CAUSALITY ?

UNIQUE STRUCTURE ?

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*
  - -> *hard to read*

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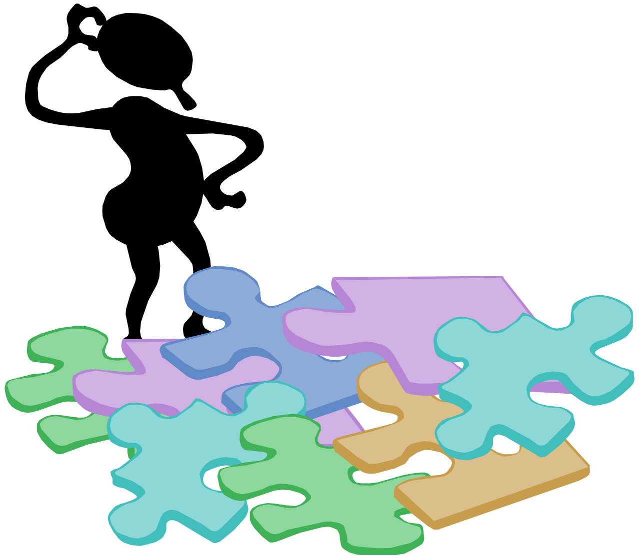
MODELS ARE PATCHWORKS FULL OF ASSUMPTIONS

PETRI NETS - THE BIG PROS



- **readable & unambigious**
  -> fault avoidant model construction
- **locality - causality - concurrency**
- **compositional, hierarchical notations**
  -> logical and macro nodes
- **executable**
  -> to experience the model, spec. causality

- **umbrella with unifying power**
  -> interpretation in qualitative / stochastic / continuous / hybrid paradigms
- **Petri net theory**
  -> P/T-invariants, ADT sets, partial order interpretation of T-invariants, conclusions CTI/CPI -> behavioural properties
  -> static analysis techniques: STP, reduction rules, . . .

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- **Petri net theory**
  -> P/T-invariants, partial order interpretation of T-invariants,
  conclusions CTI/CPI -> behavioural properties
  -> [illegible] reduction rules, . . .

THE PETRI NET FRAMEWORK

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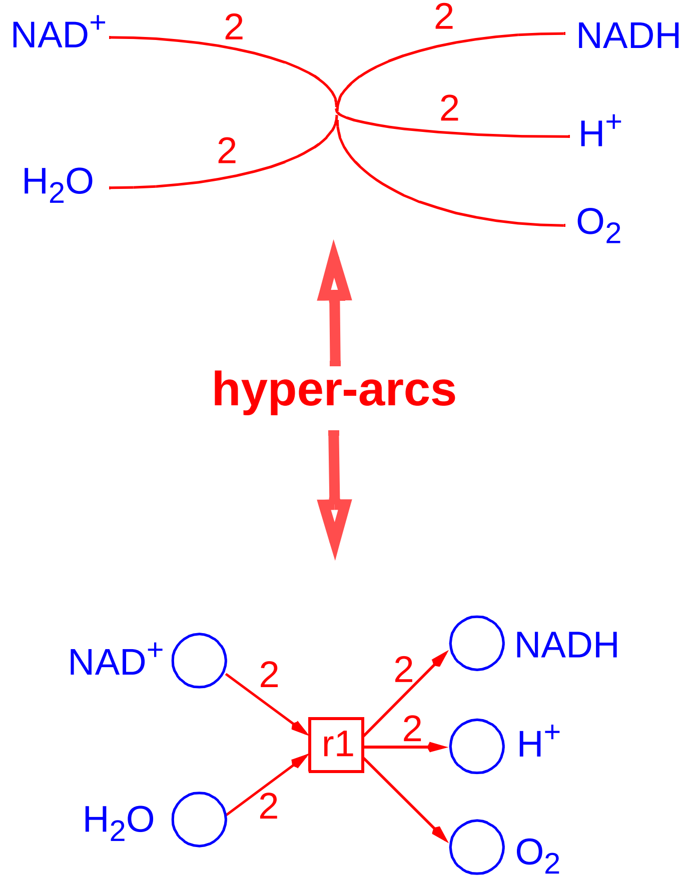
.. .

ARE NETWORKS OF BIOCHEMICAL REACTIONS

. . .

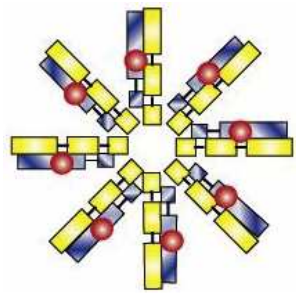
NATURALLY EXPRESSIBLE AS PETRI NETS

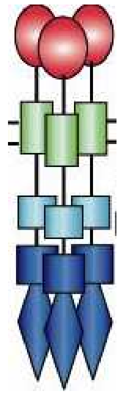
$$2\ NAD^+ + 2\ H_2O \rightarrow 2\ NADH + 2\ H^+ + O_2$$

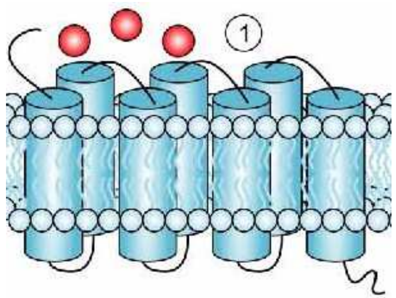


PLACES, TRANSITIONS - SOME BIO INTERPRETATIONS

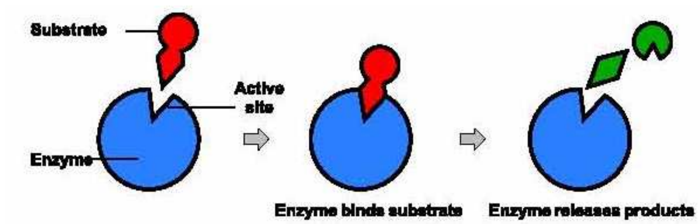
- **places -> model variables**
  - -> *(bio-) chemical compounds*
  - -> *proteins*
  - -> *protein conformations*
  - -> *complexes*
  - -> *genes, . . . , etc.*
  - *. . . in different locations*







- **transitions -> atomic events**
  - -> *(stoichiometric) chemical reaction*
  - -> *complexation / decomplexation*
  - -> *phosphorylation / dephosphorylation*
  - -> *conformational change*
  - -> *transport step, . . . , etc.*
  - *. . . in different locations*



STATE-DEPENDENT RATE FUNCTIONS

STOCHASTIC RATES

↓

LAMBDA OF EXPONENTIAL WAITING TIME

↓

CTMC

CONTINUOUS RATES

↓

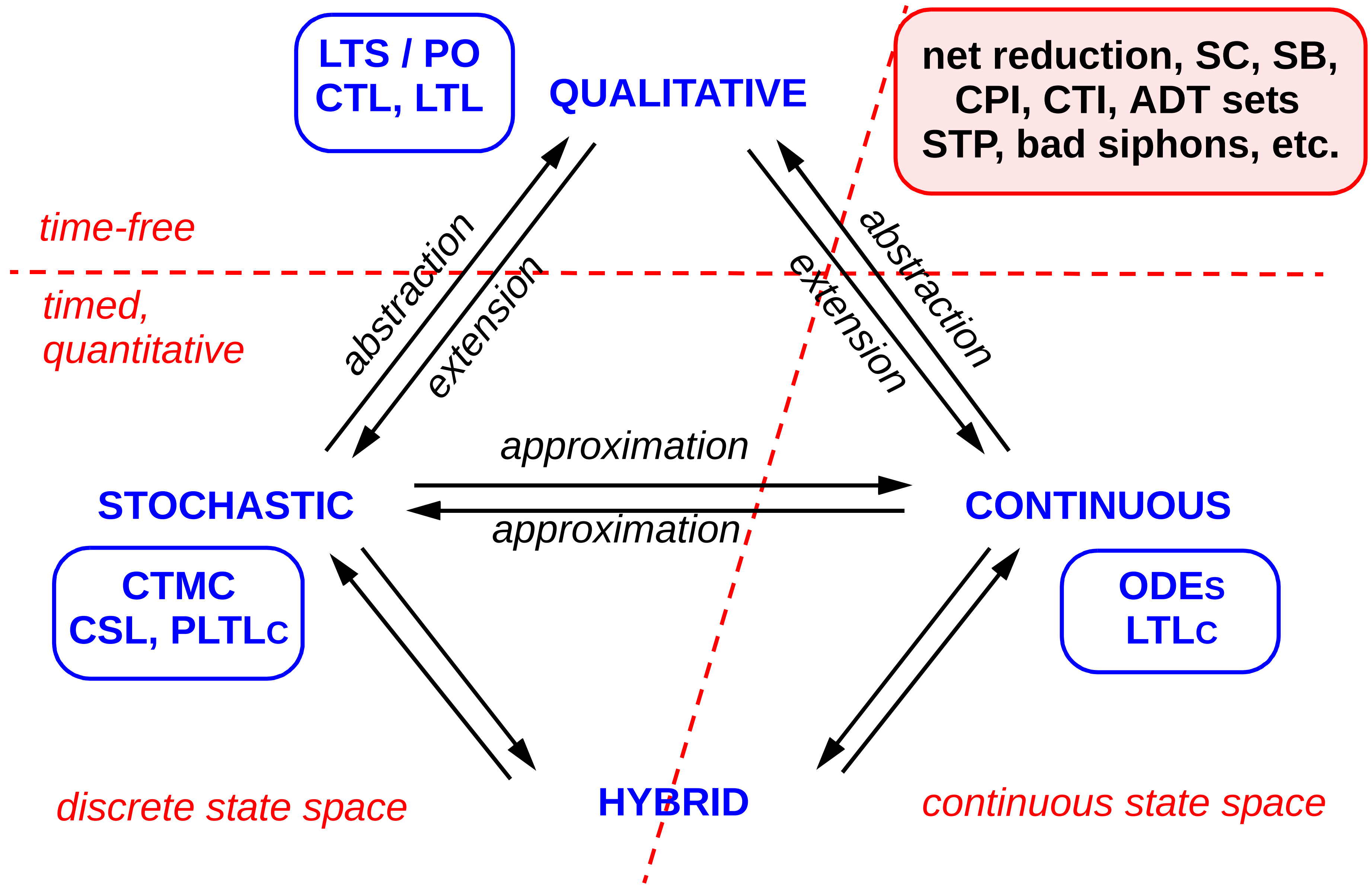
STRENGTH OF CONTINUOUS FLOW

↓

ODEs

-> supported by, e.g., COPASI, Dizzy, ..., Snoopy

FRAMEWORK 2010



4 MODELS SHARING STRUCTURE

↓ ↓ ↓

QUANTITATIVE MODEL = QUALITATIVE MODEL

\+

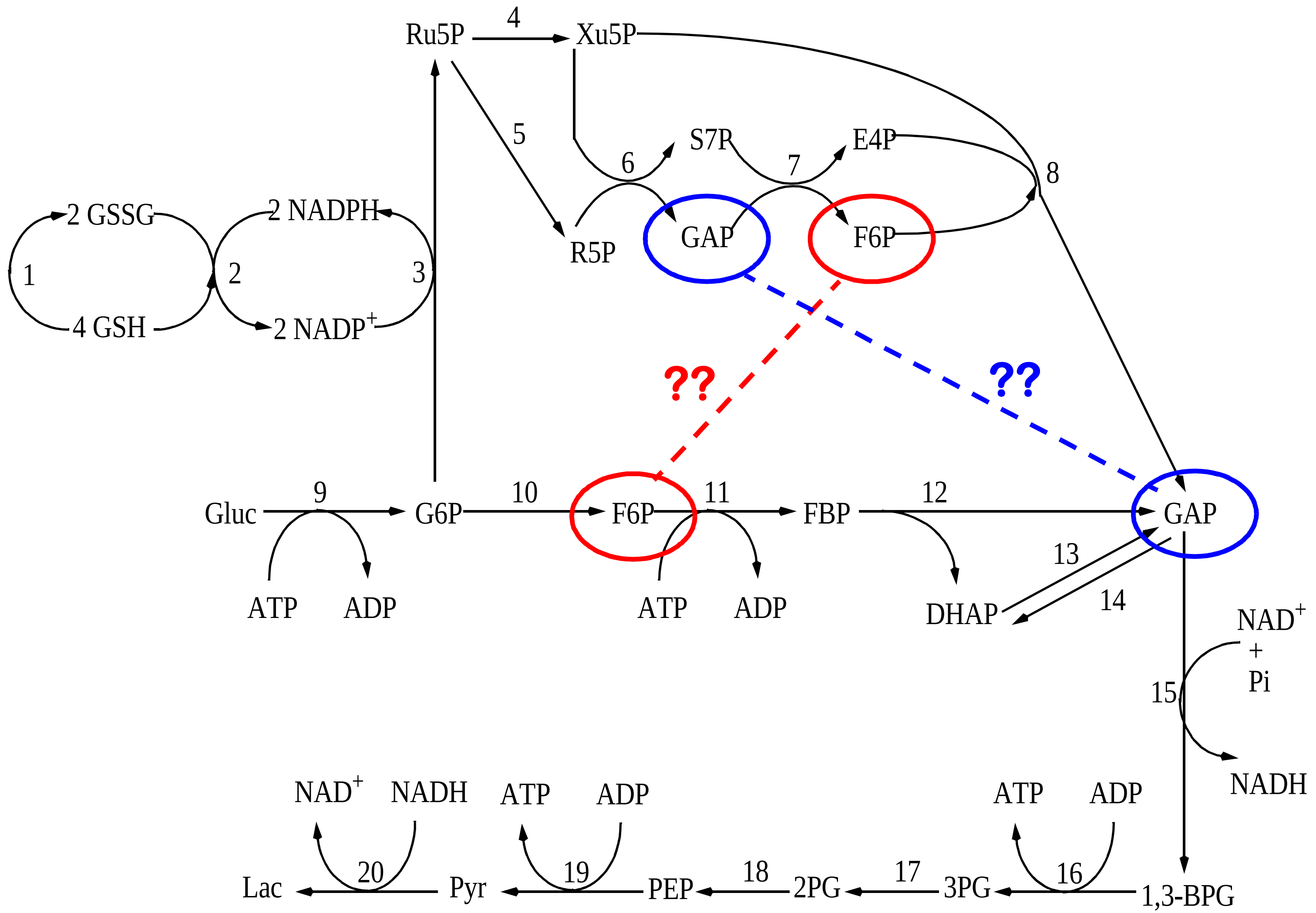
RATE FUNCTIONS

(KINETICS)

BIO PETRI NETS - SOME EXAMPLES

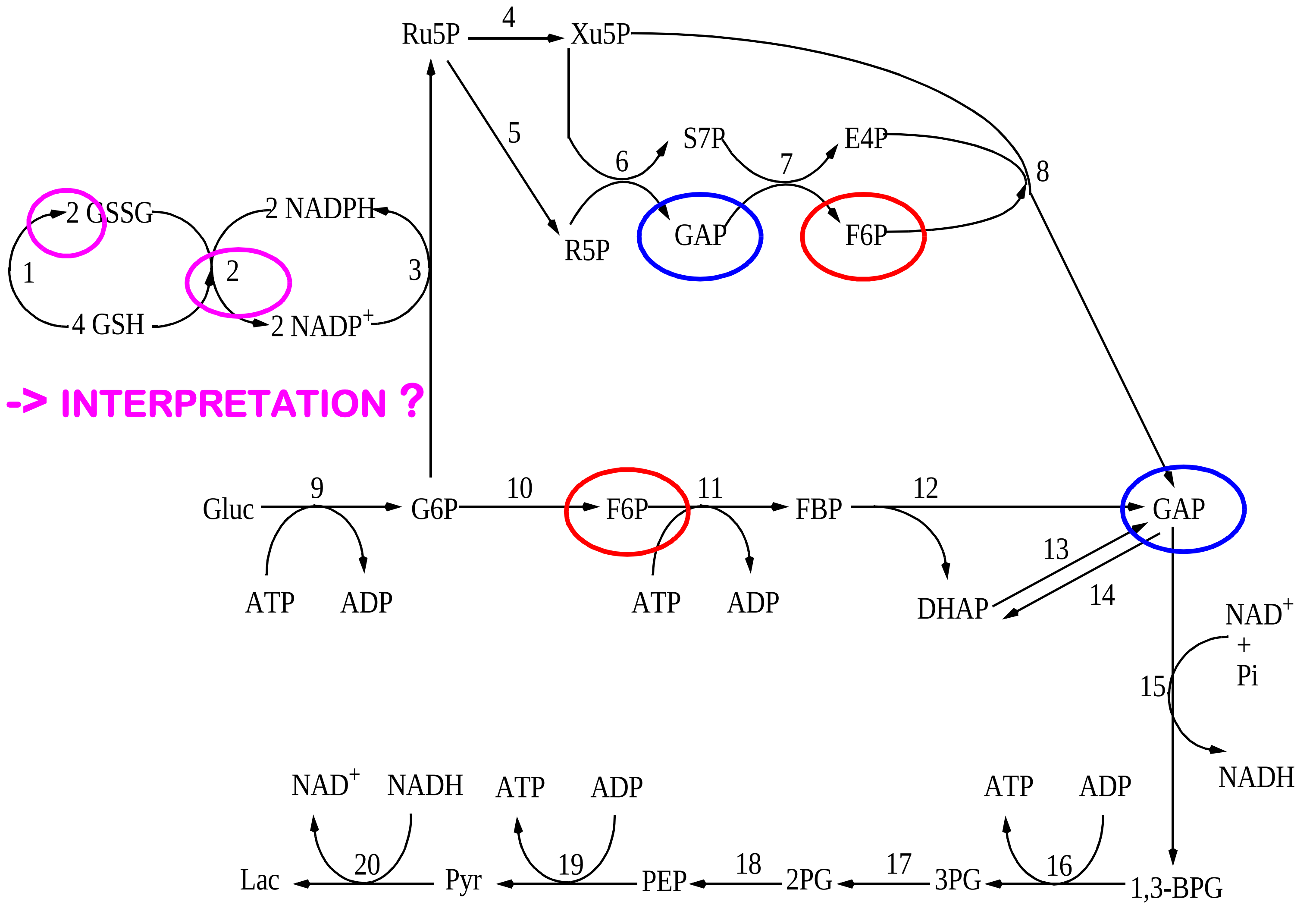
Ex1 - Glycolysis and Pentose Phosphate Pathway

[Reddy 1993]



Ex1 - Glycolysis and Pentose Phosphate Pathway

[Reddy 1993]



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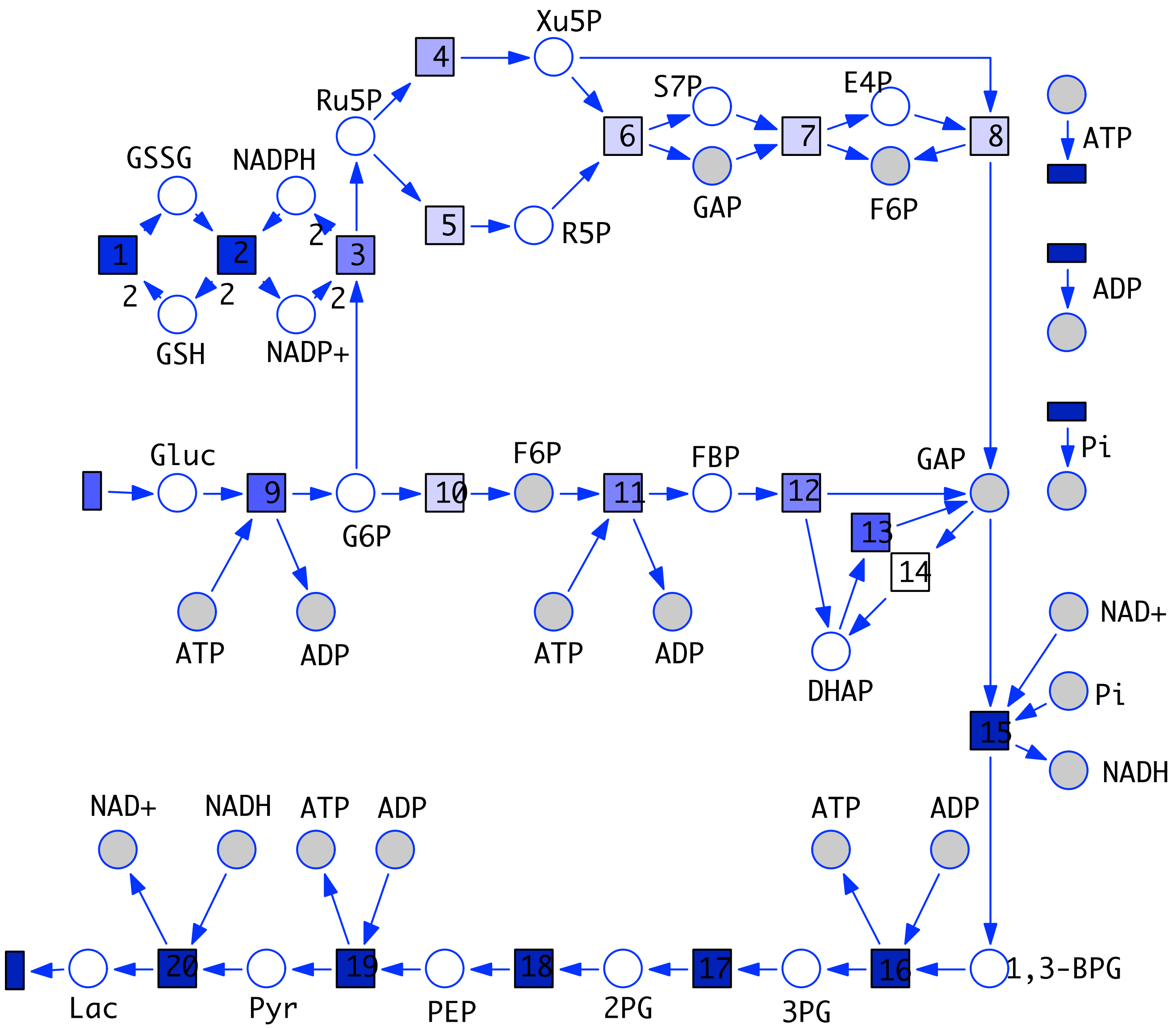


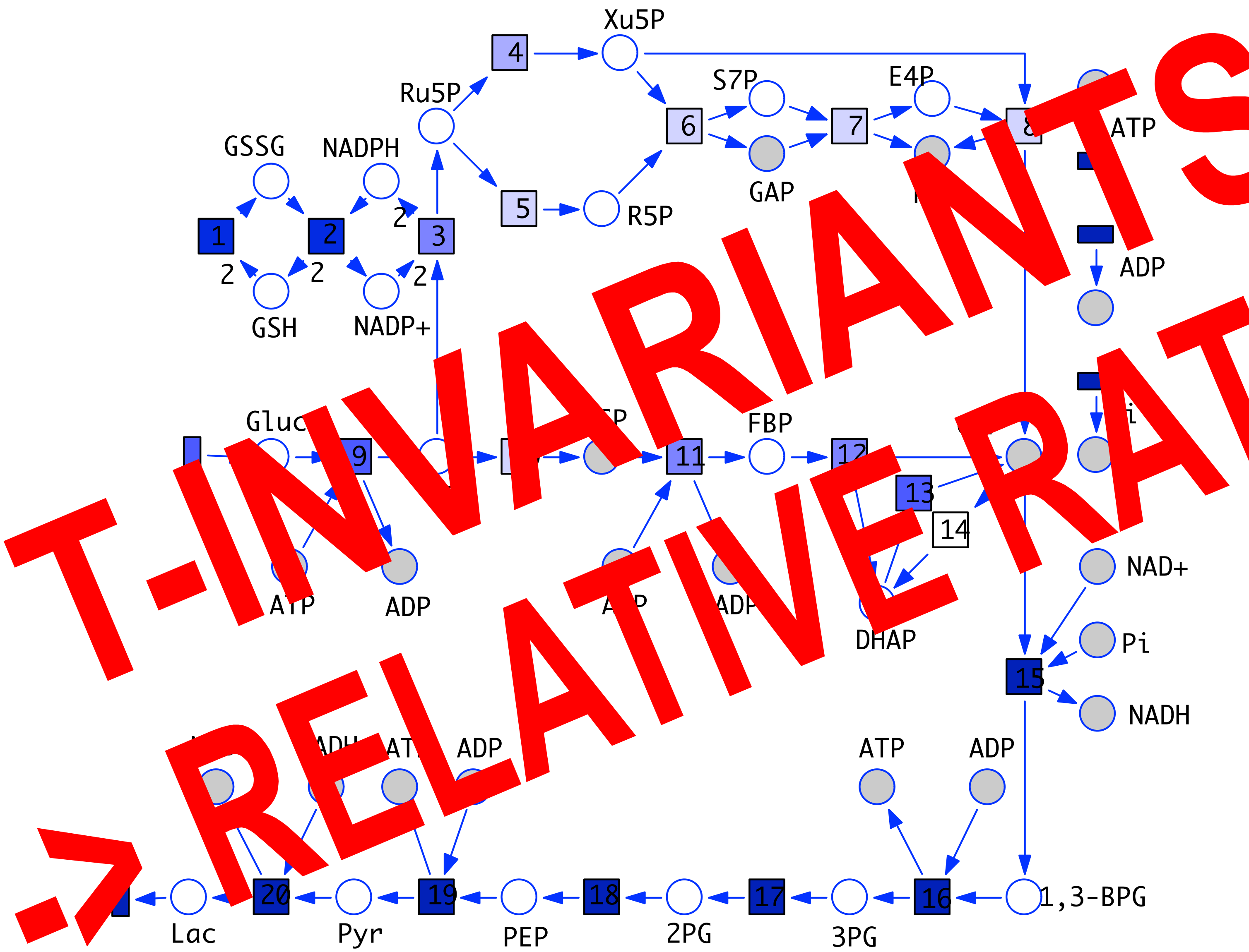
[Reddy 1993]

[Heiner 1998]

. . .

[Koch, Heiner 2010]

Ex1 - Glycolysis and Pentose Phosphate Pathway



T-INVARIANTS -> RELATIVE RATES

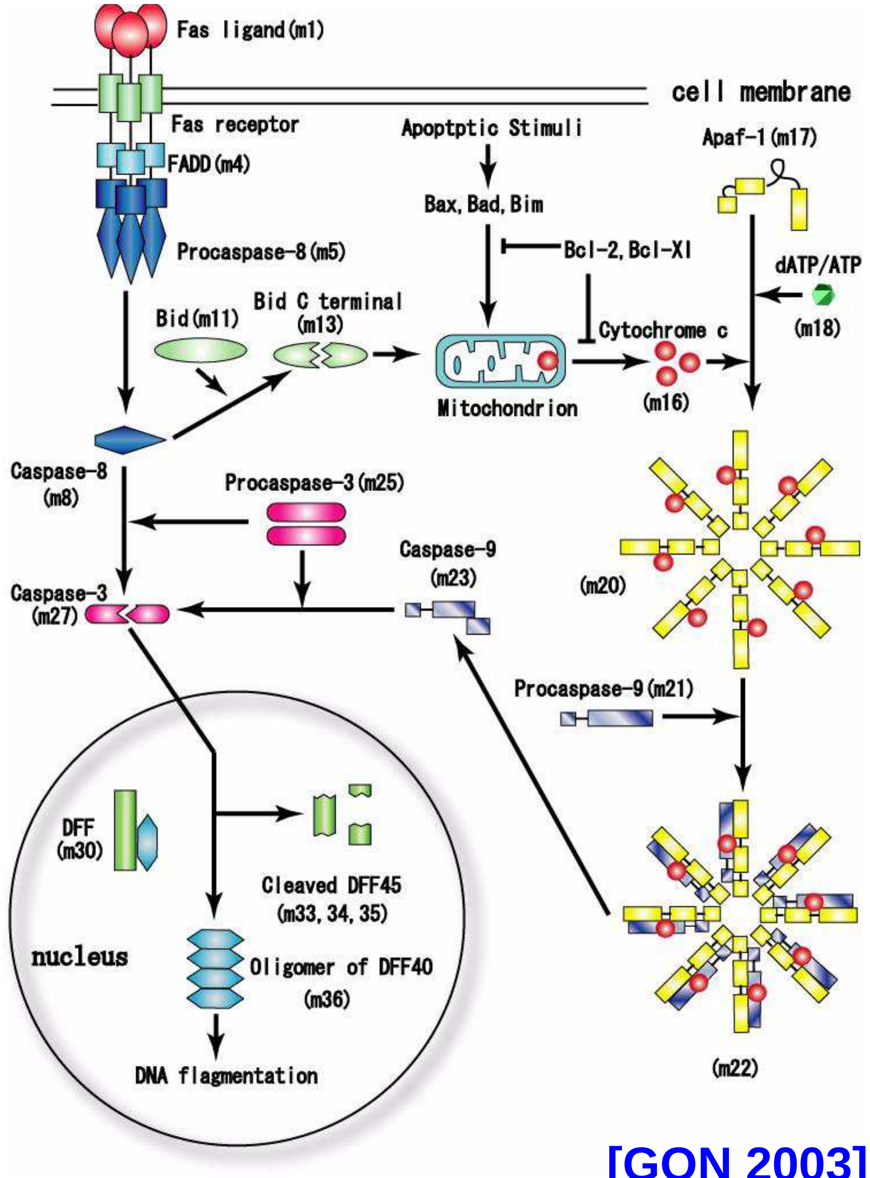
[Reddy 1993]

[Heiner 1998]

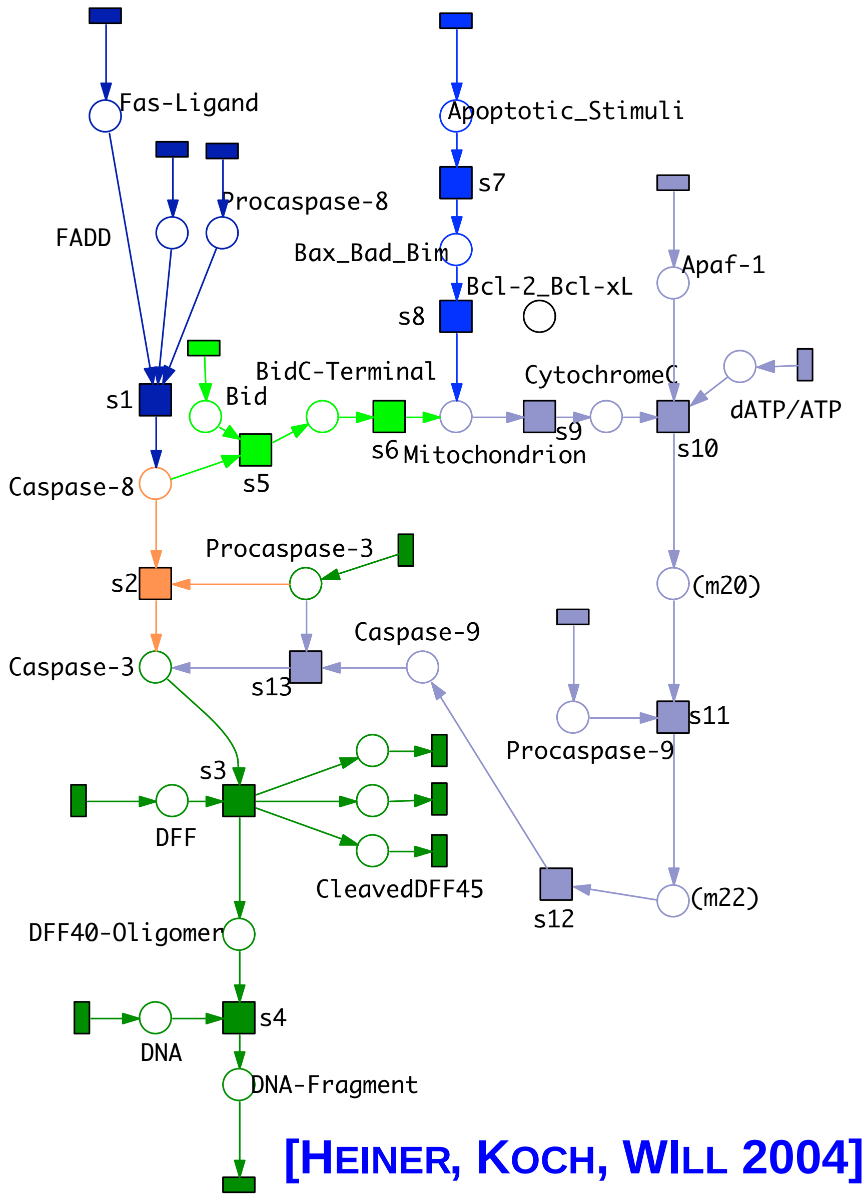
. . .

[Koch, Heiner 2010]

Ex2 - Apoptosis in Mammalian Cells

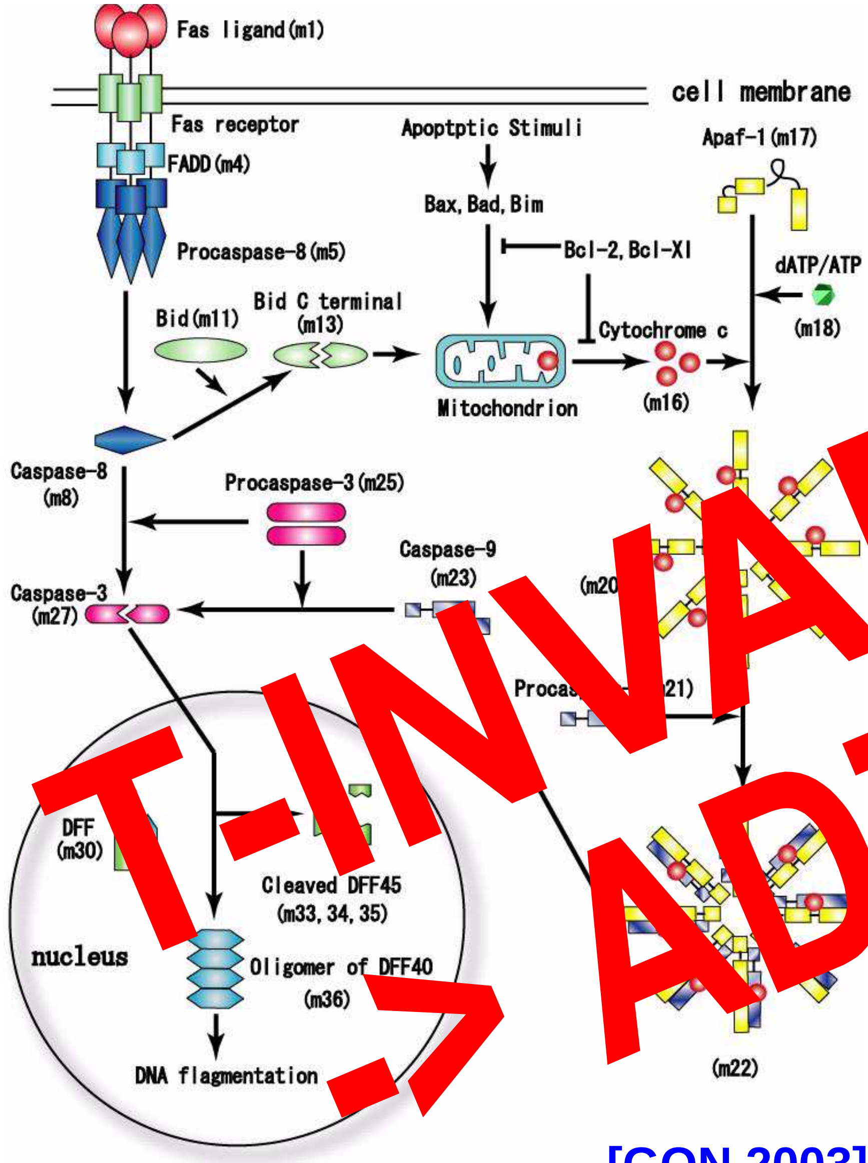


[GON 2003]

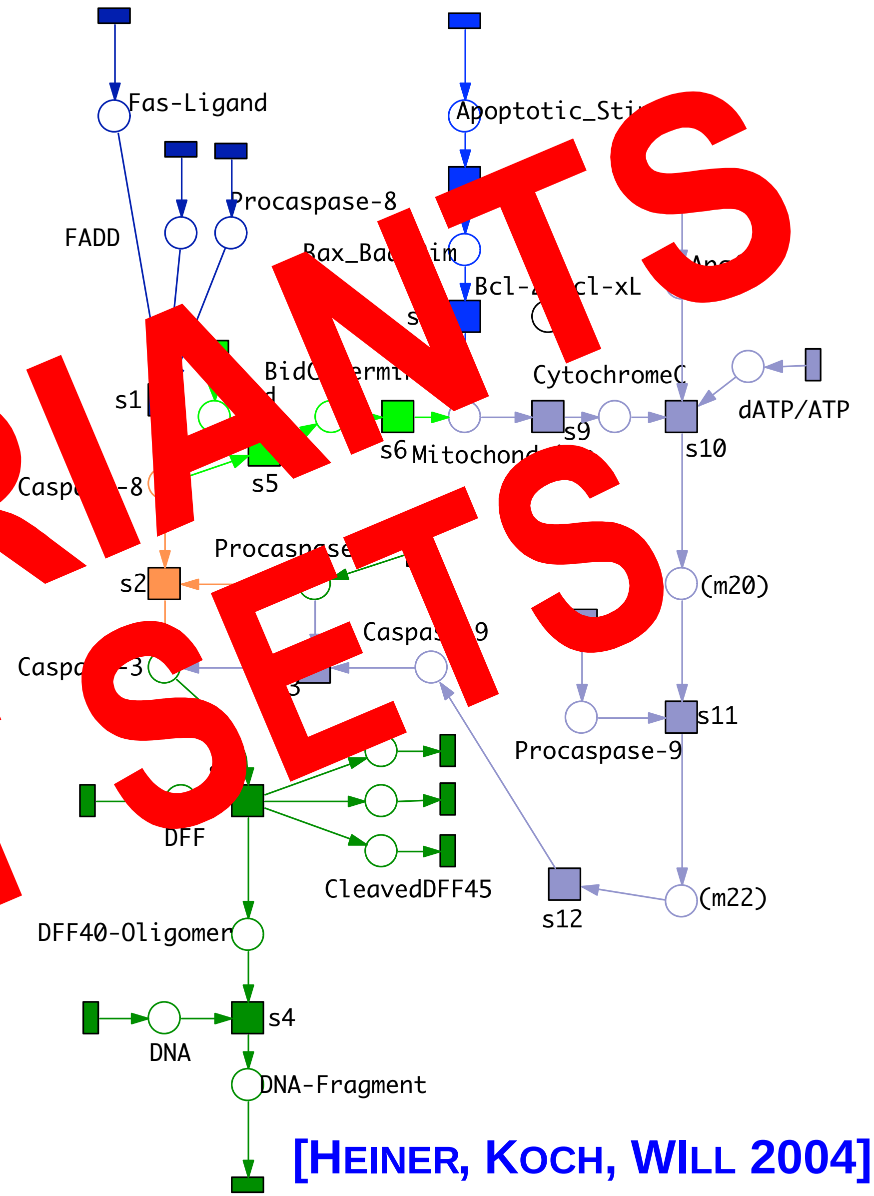


[Heiner, Koch, Will 2004]

Ex2 - Apoptosis in Mammalian Cells



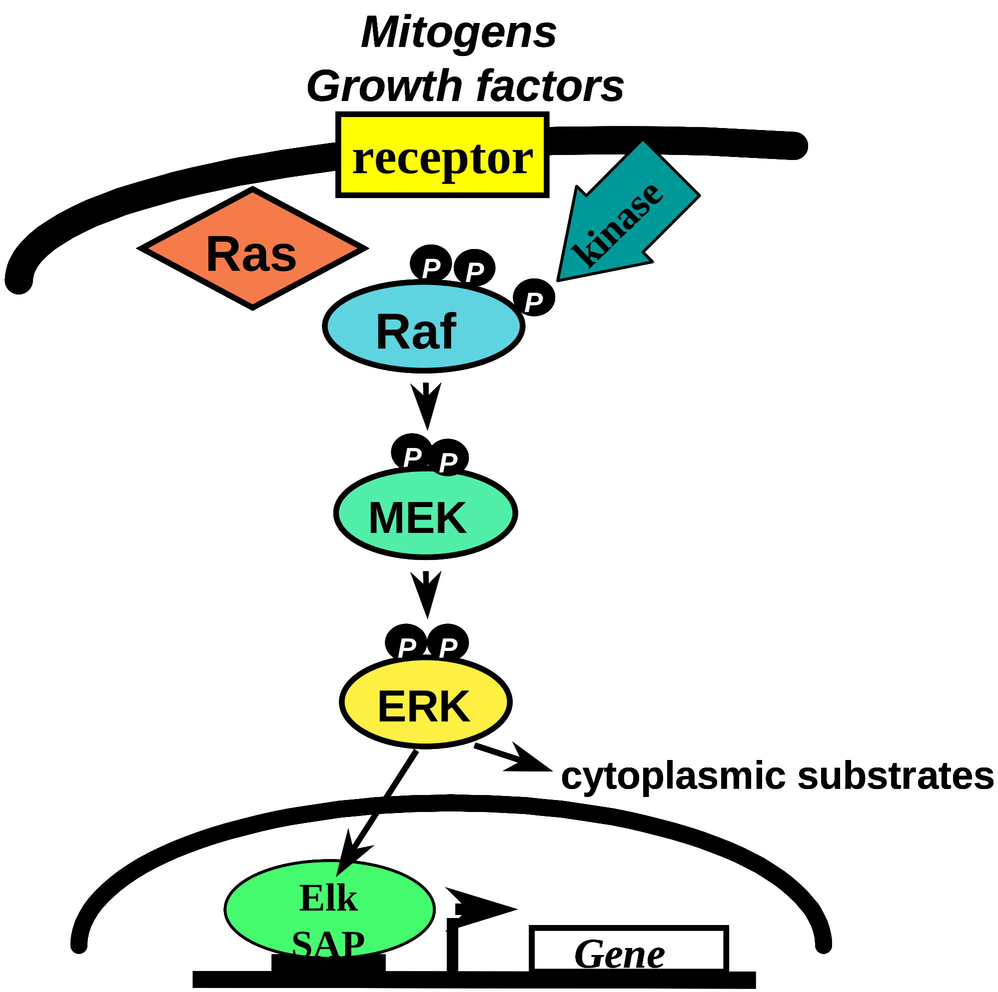
[Gon 2003]



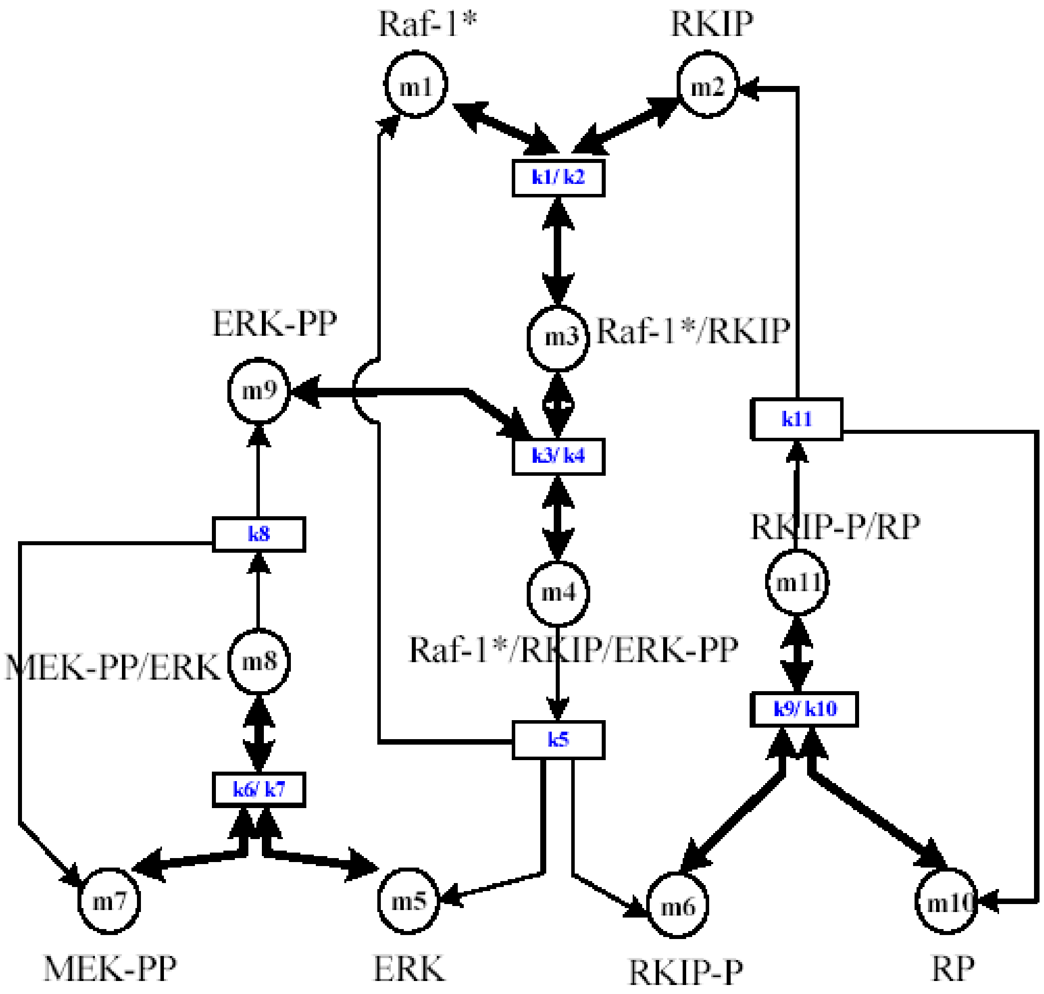
[Heiner, Koch, Will 2004]

T-INVARIANTS -> ADT SETS

…one pathway…

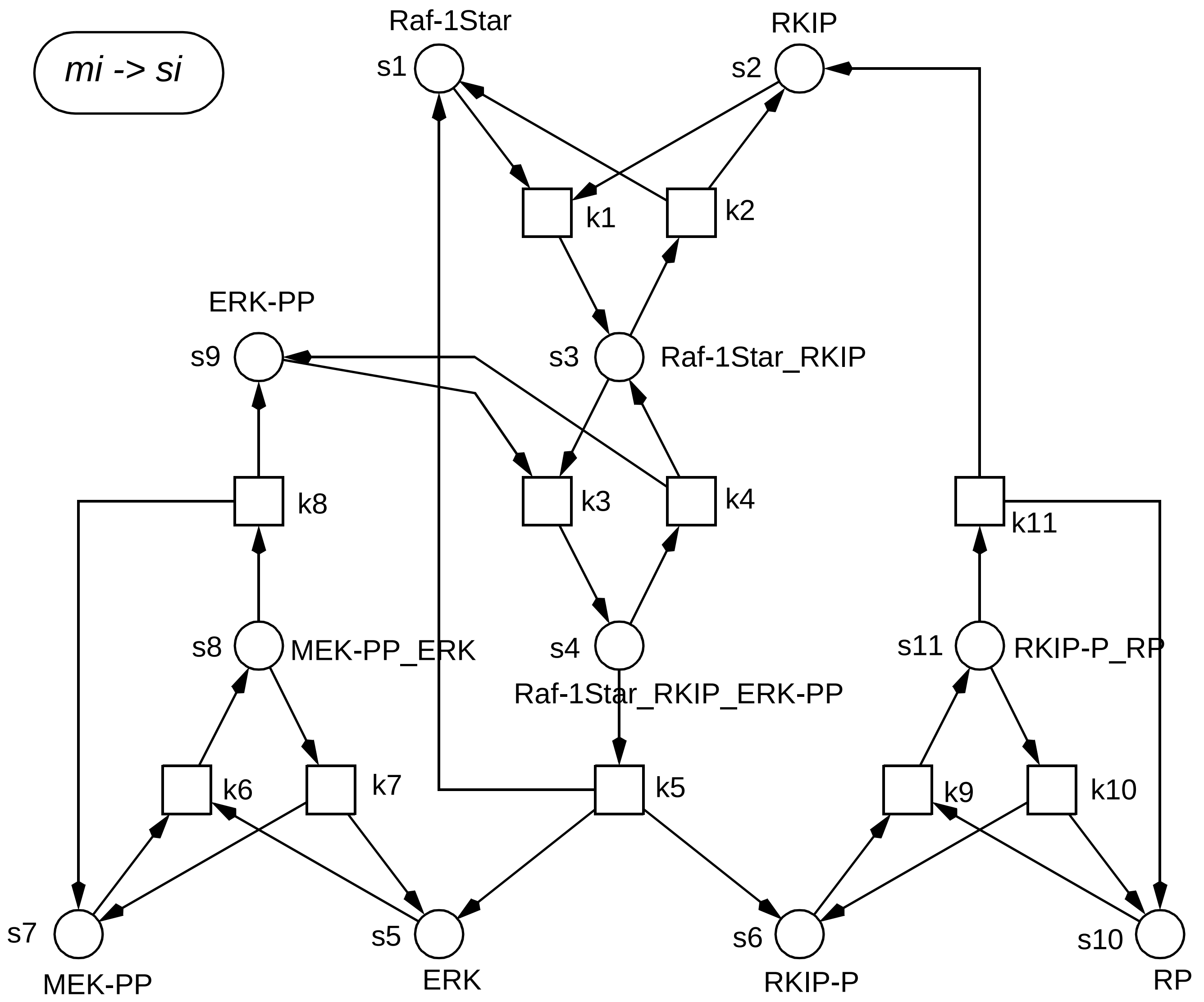

Mitogens
Growth factors
receptor
kinase
Ras
P
P
P
Raf
P
P
MEK
P
P
ERK
cytoplasmic substrates
Elk
SAP
Gene

EX3 - RKIP SIGNALLING PATHWAY



[Cho et al. 2003]

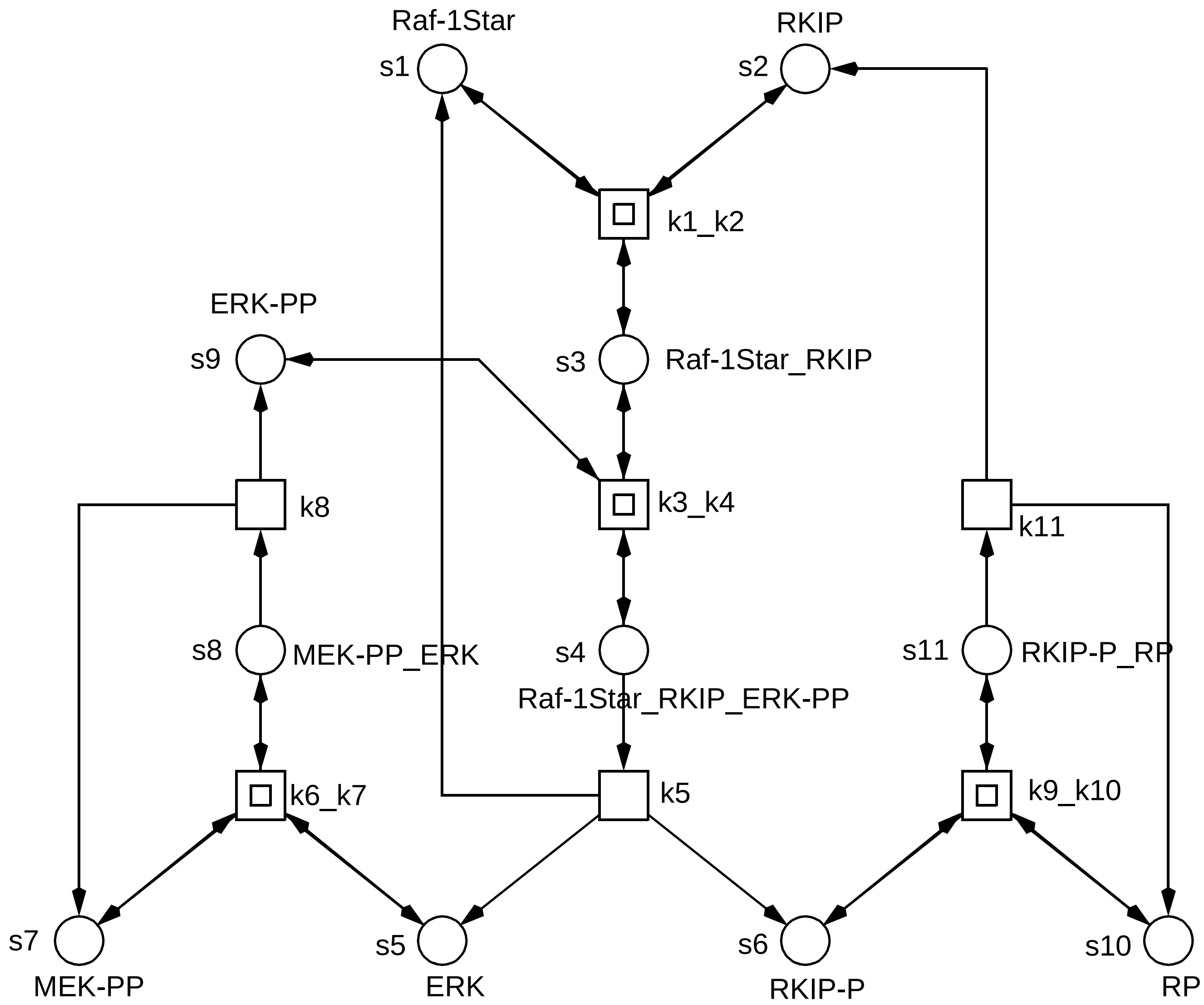
Ex3 - RKIP Signalling Pathway, Petri Net



[Heiner, Gilbert 2006]

[Heiner, Donaldson, Gilbert 2010]

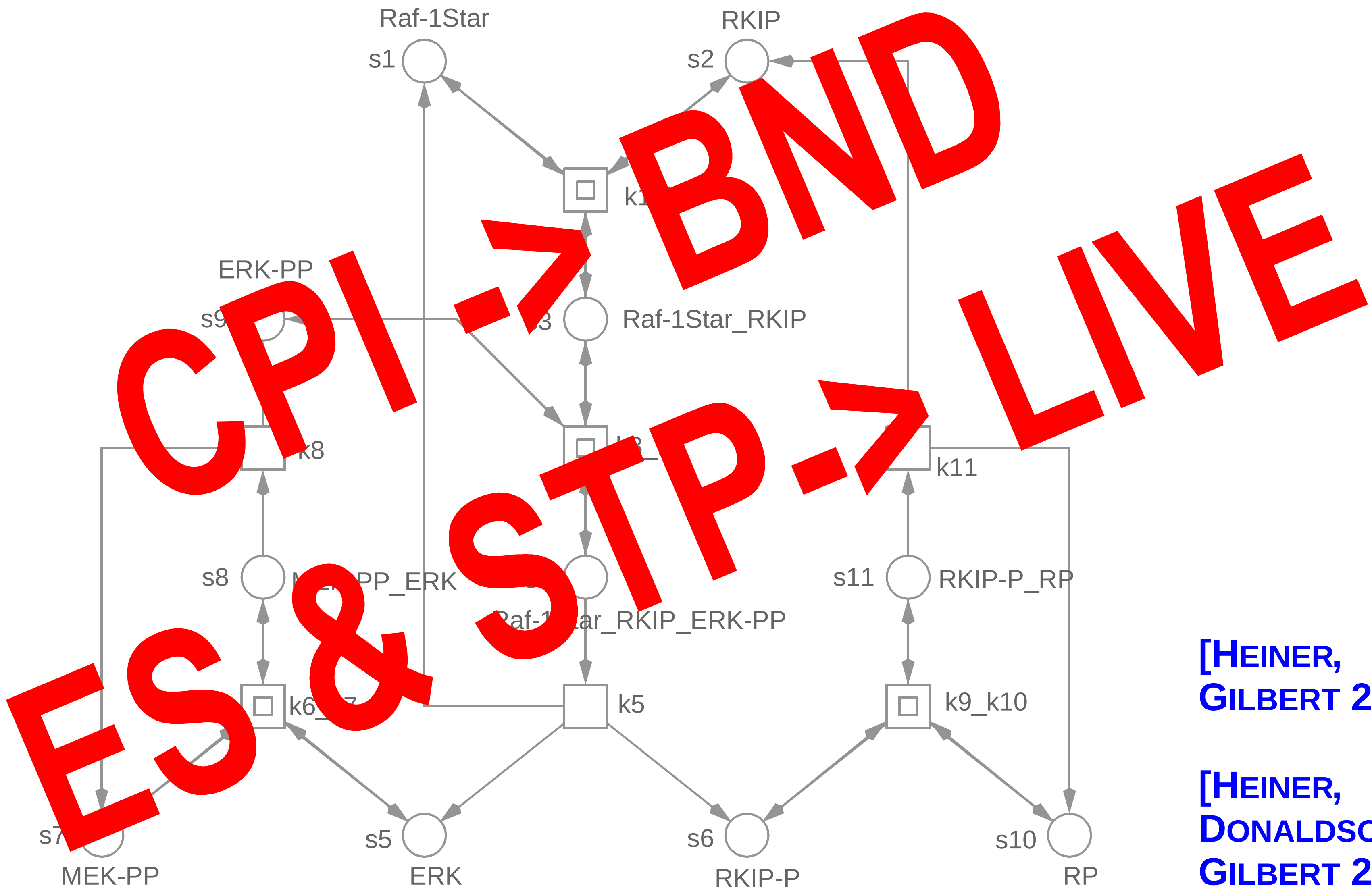
EX3 - RKIP SIGNALLING PATHWAY, HIERARCHICAL PETRI NET



[HEINER, GILBERT 2006]

[HEINER, DONALDSON, GILBERT 2010]

Ex3 - RKIP Signalling Pathway, Hierarchical Petri Net



[Heiner, Gilbert 2006]

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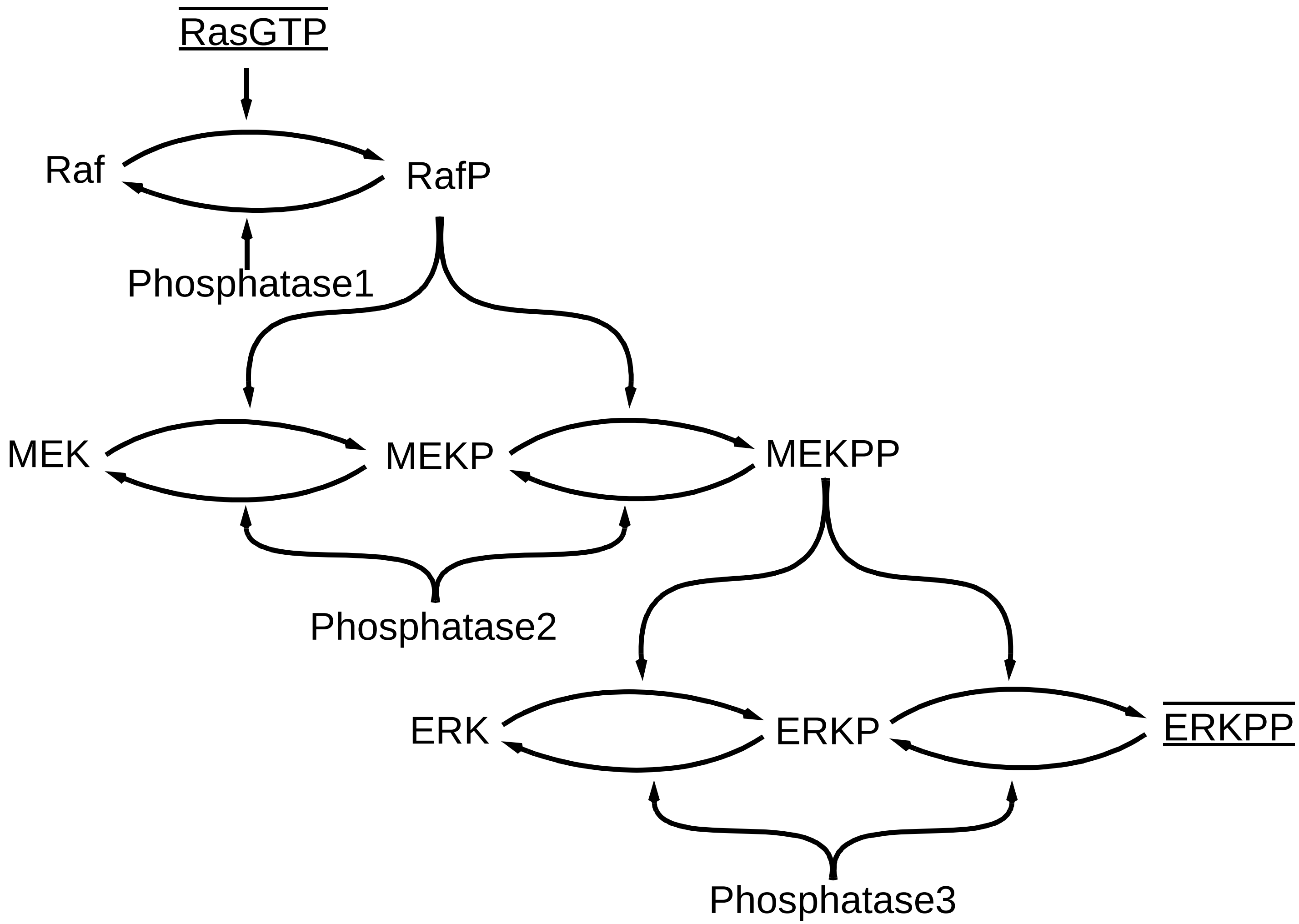
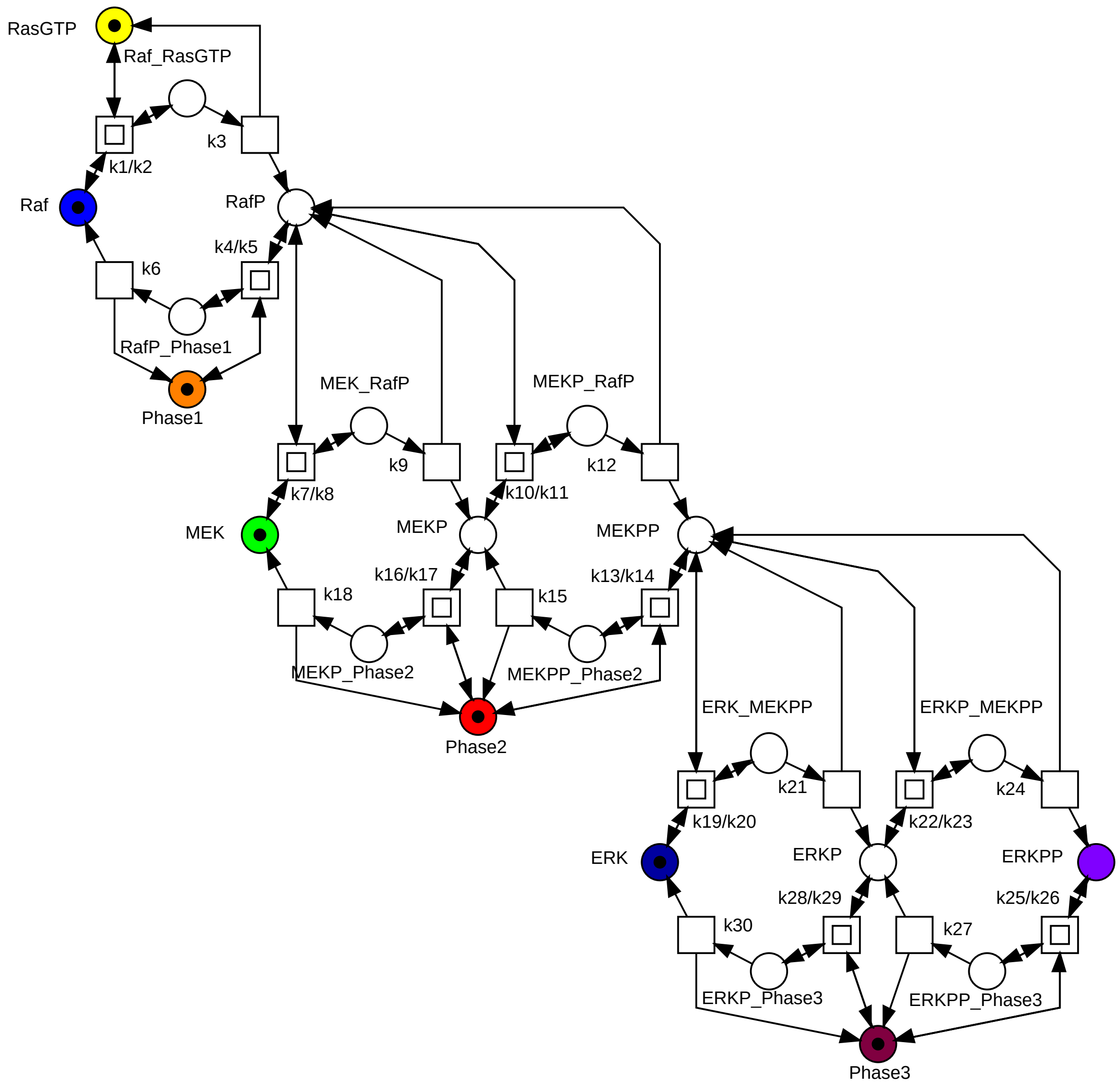
EX4 - SIGNALLING CASCADE



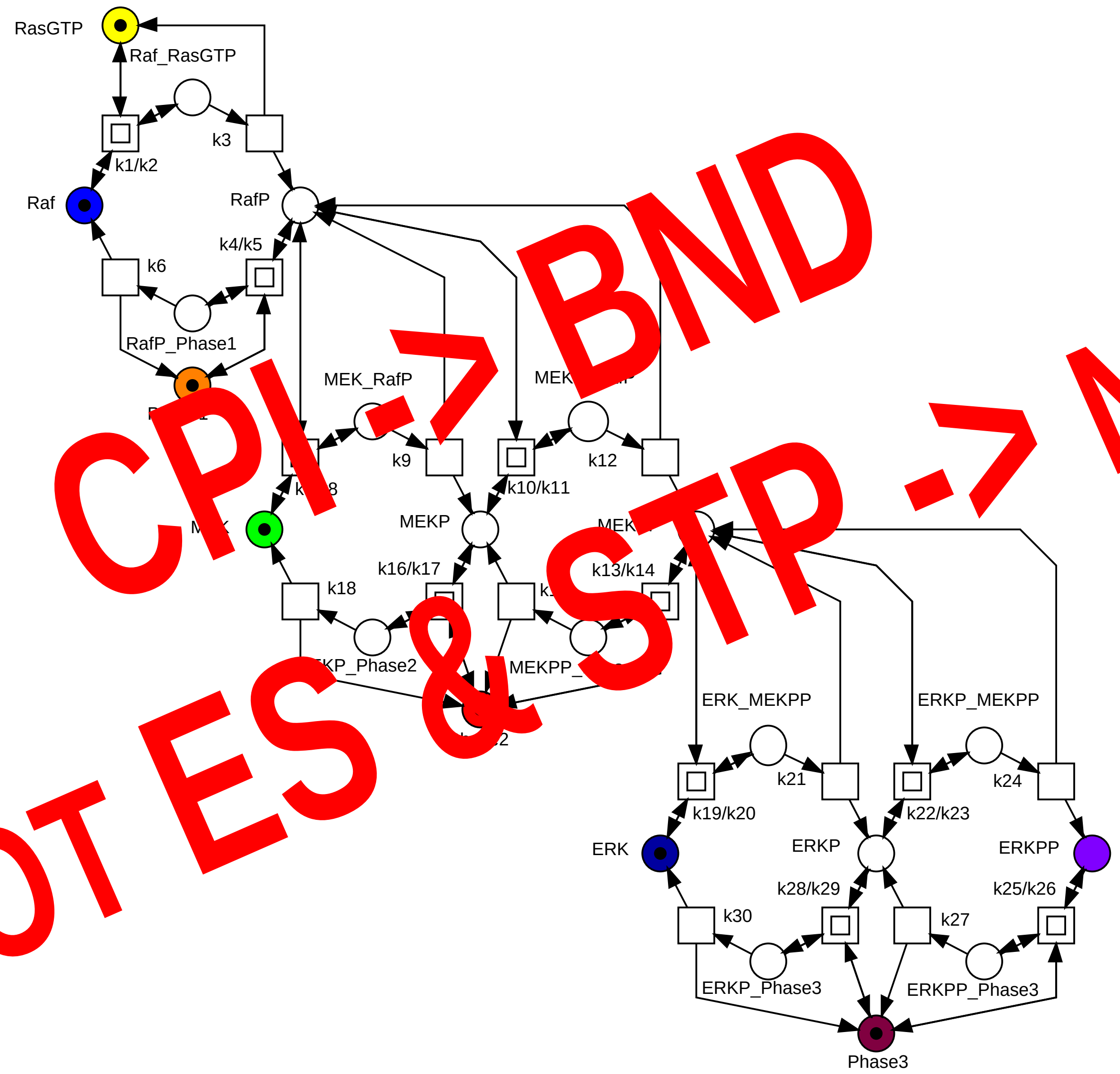
EX4 - SIGNALLING CASCADE



[GILBERT, HEINER, LEHRACK 2007]

[HEINER, GILBERT, DONALDSON 2008]

Ex4 - Signalling Cascade



CPI -> BND

NOT ES & STP -> NO DST

[Gilbert, Heiner, Lehrack 2007]

[Heiner, Gilbert, Donaldson 2008]

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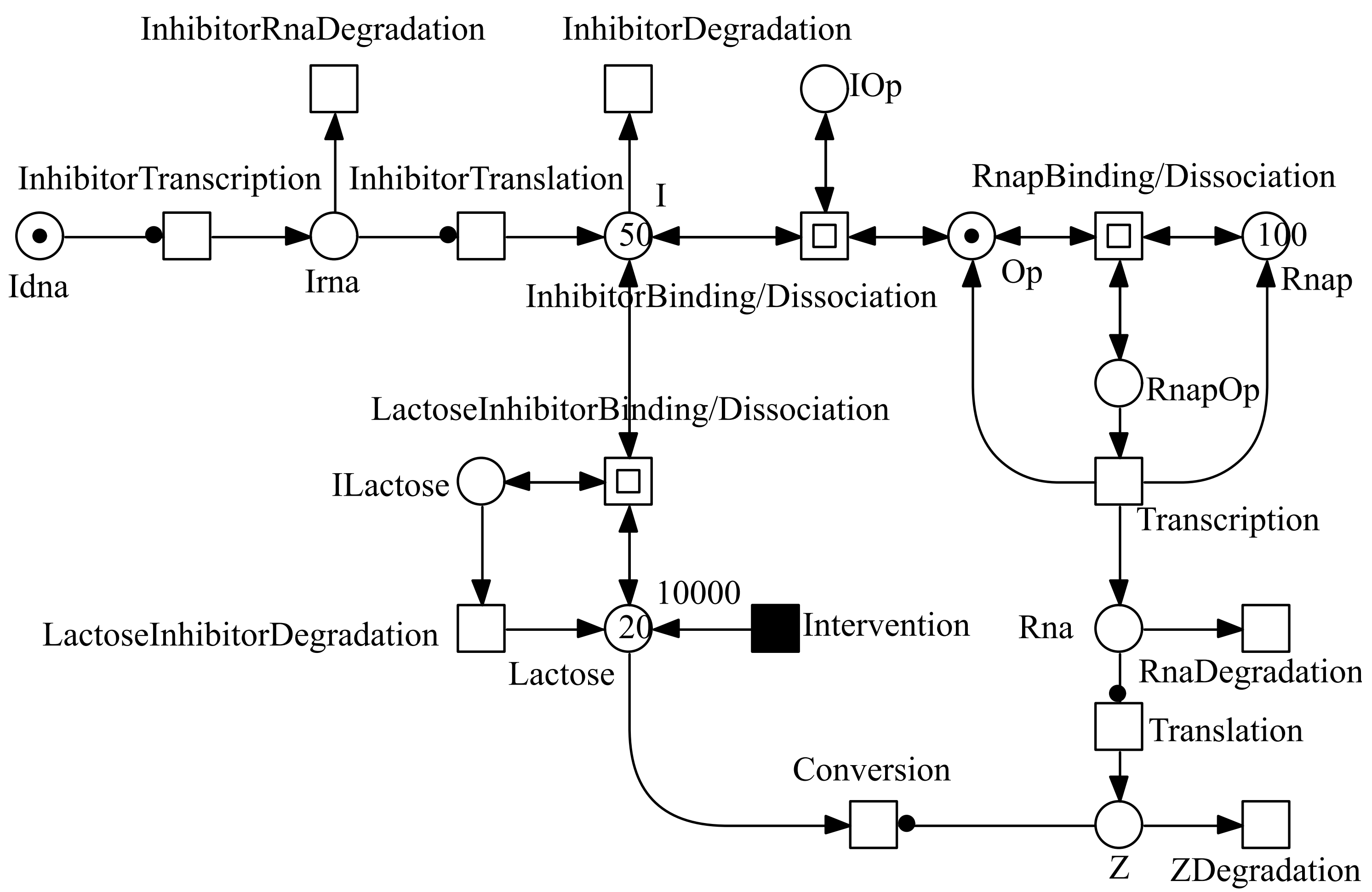
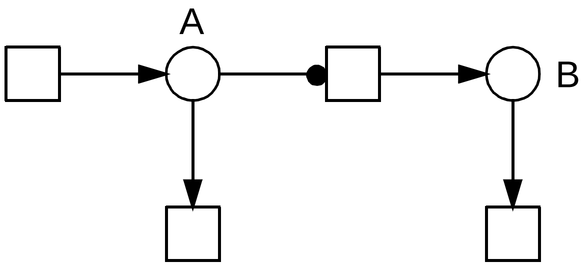
Ex5 - Lac Operon



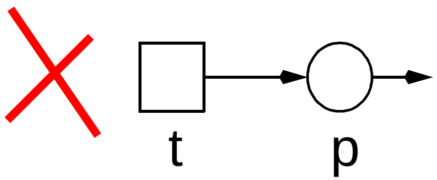
[Wilkinson 2006]

- **reduced net structure while preserving liveness & boundedness**

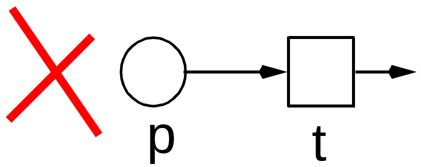


liveness becomes obvious

- **example of two simple reduction rules**

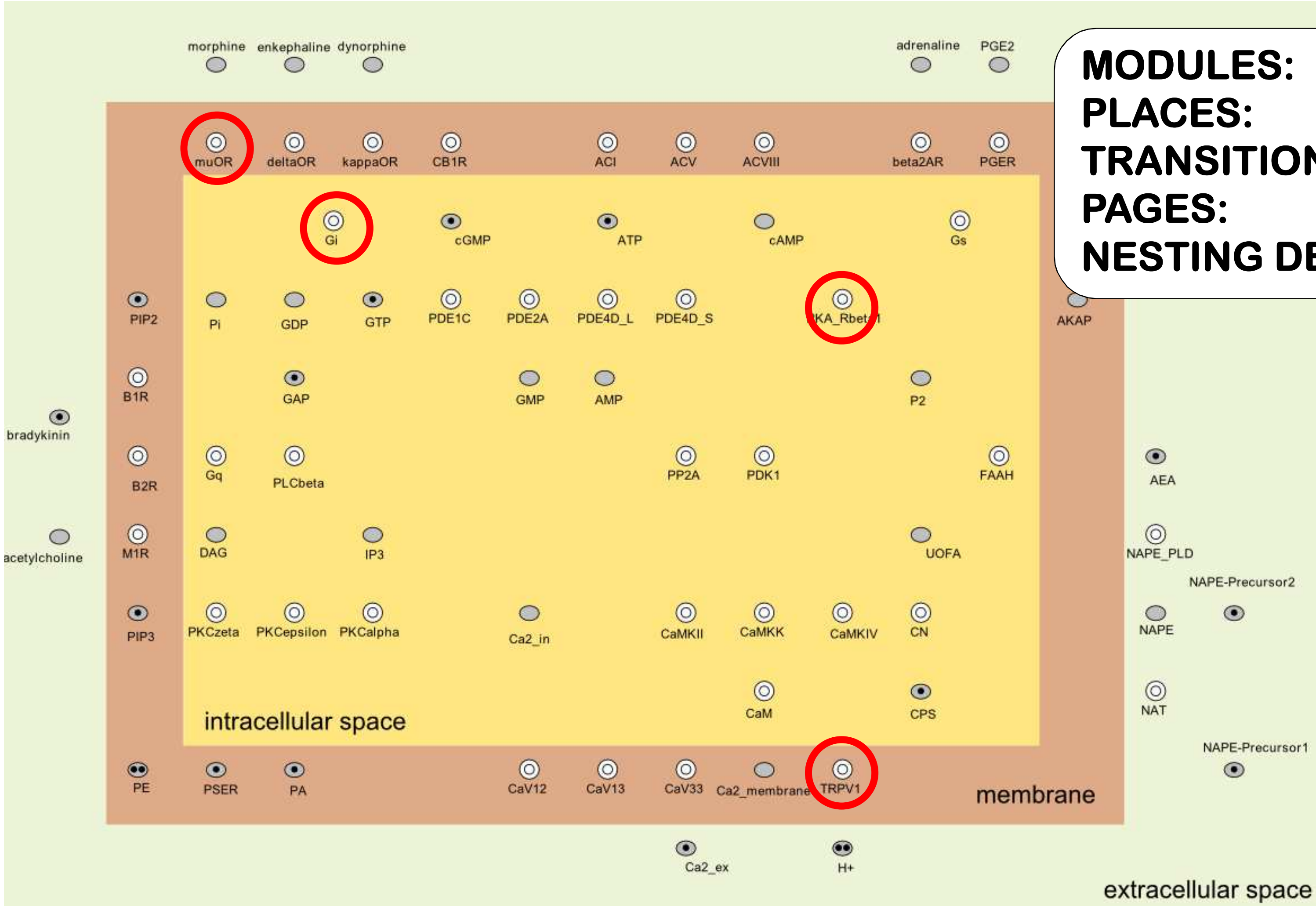


t live
p unbounded



t not live
p bounded

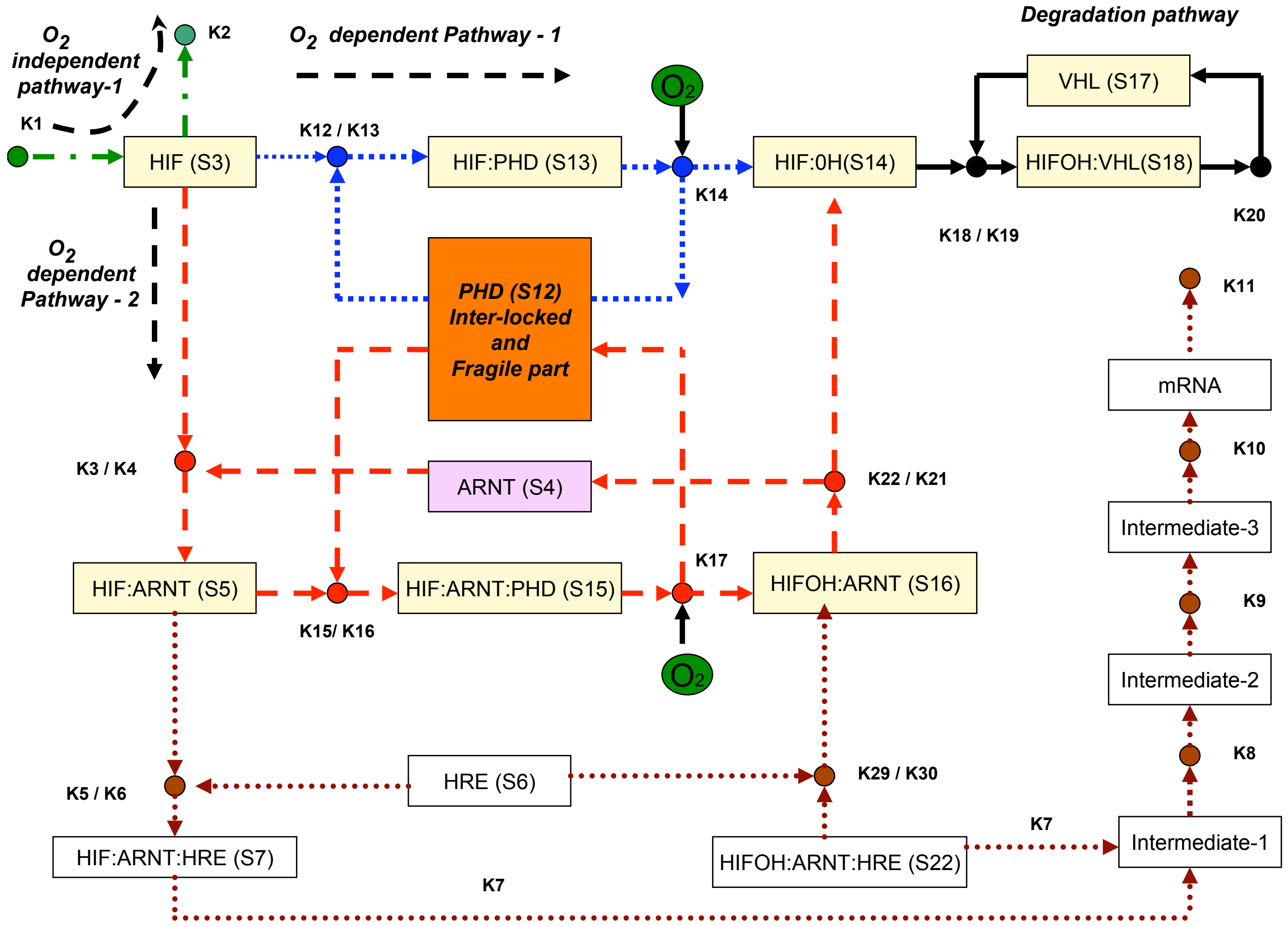
[BLÄTKE, MEYER, MARWAN 2011]
-> A PROTEIN-ORIENTED MODULAR MODELLING CONCEPT



MODULES: 38
PLACES: 713
TRANSITIONS: 775
PAGES: 325
NESTING DEPTH: 4

About the Relation Qualitative vs Continuous

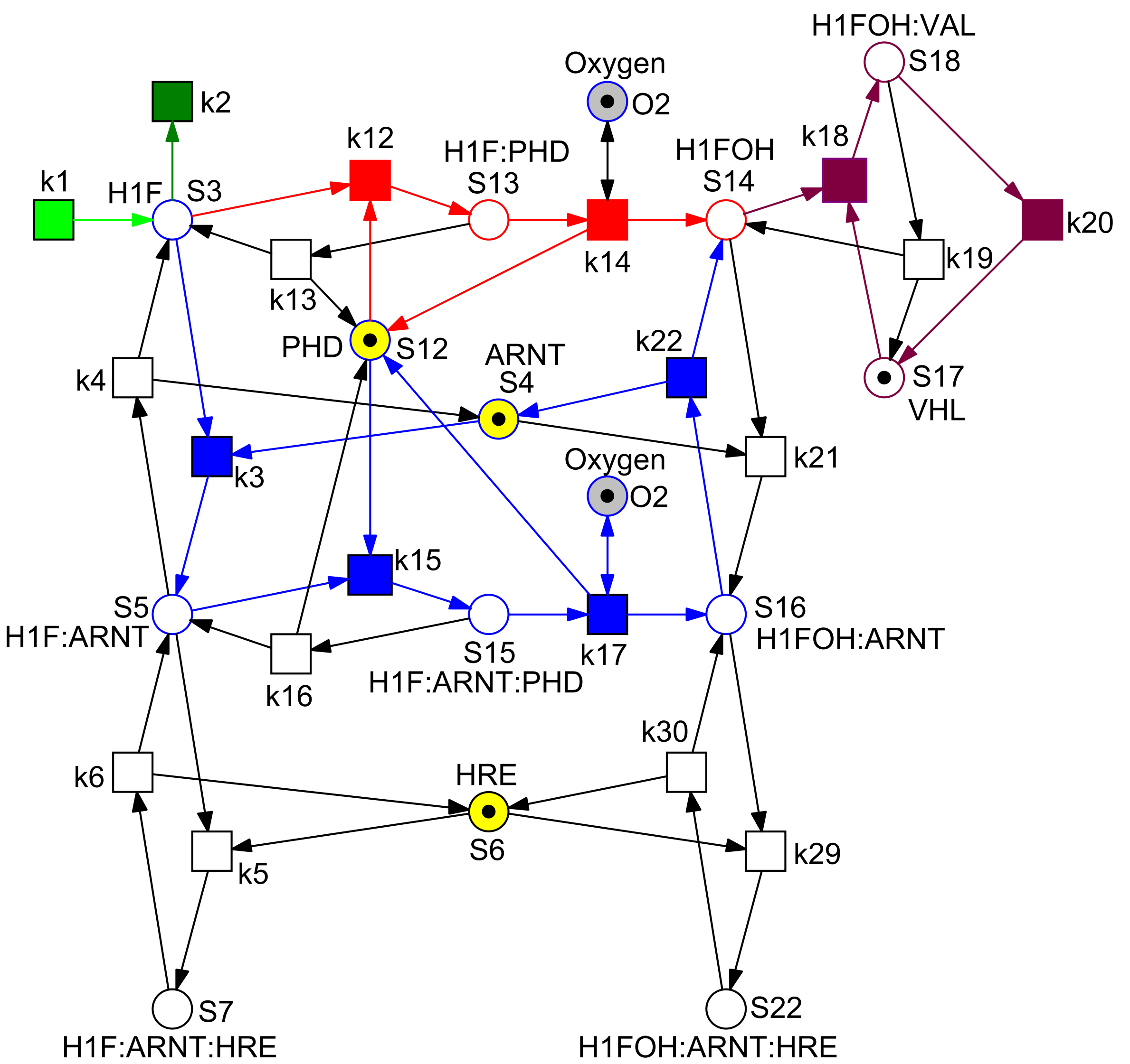
[Yu et al. 2007]

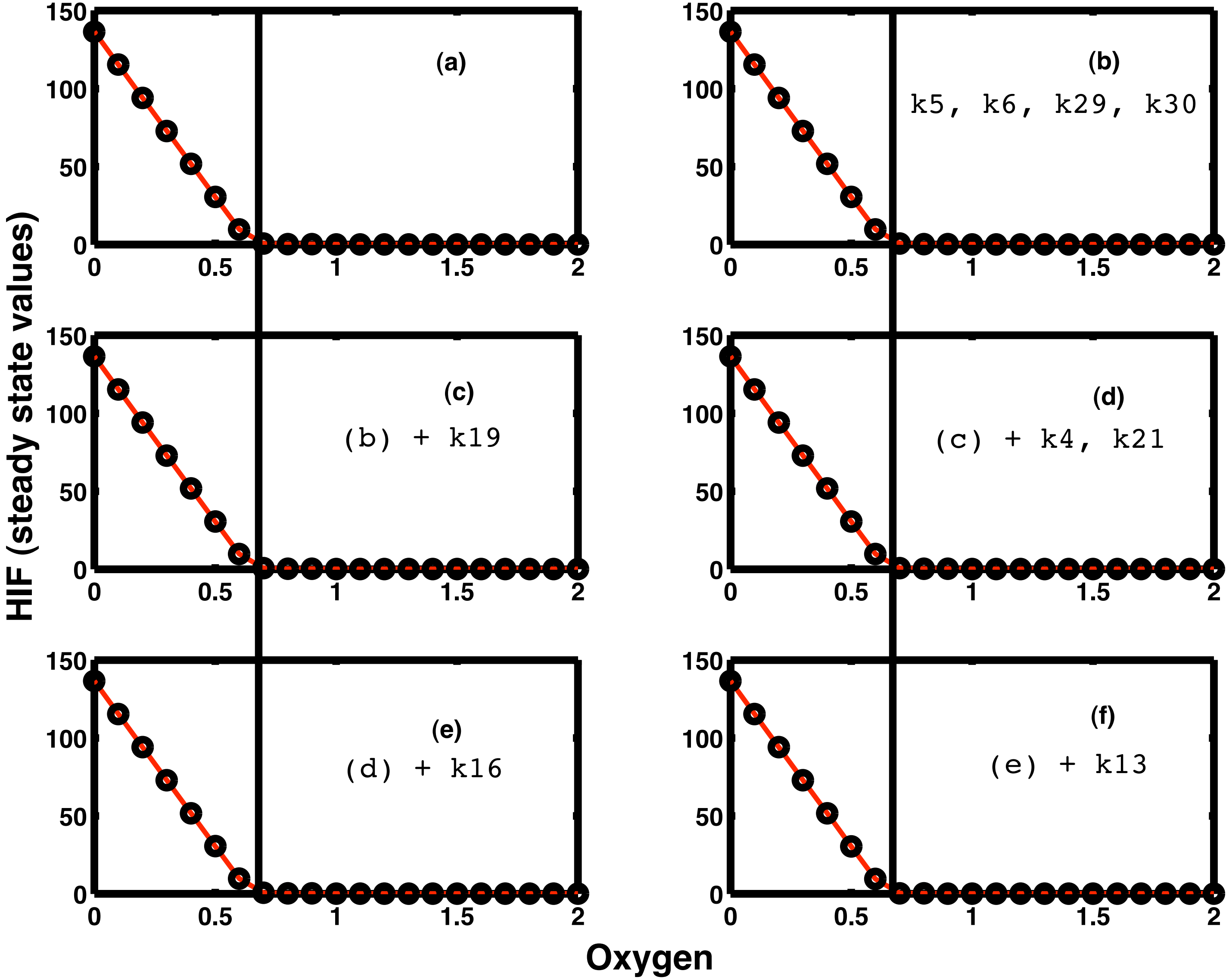


O_2 independent pathway-1
K1
K2
O_2 dependent Pathway - 1
Degradation pathway
O_2
VHL (S17)
HIF (S3)
K12 / K13
HIF:PHD (S13)
HIF:0H(S14)
HIFOH:VHL(S18)
K14
K18 / K19
K20
O_2 dependent Pathway - 2
PHD (S12) Inter-locked and Fragile part
K11
mRNA
K10
K3 / K4
ARNT (S4)
K22 / K21
Intermediate-3
K17
HIF:ARNT (S5)
HIF:ARNT:PHD (S15)
HIFOH:ARNT (S16)
K9
K15/ K16
O_2
Intermediate-2
HRE (S6)
K8
K5 / K6
K29 / K30
HIF:ARNT:HRE (S7)
K7
HIFOH:ARNT:HRE (S22)
Intermediate-1
K7

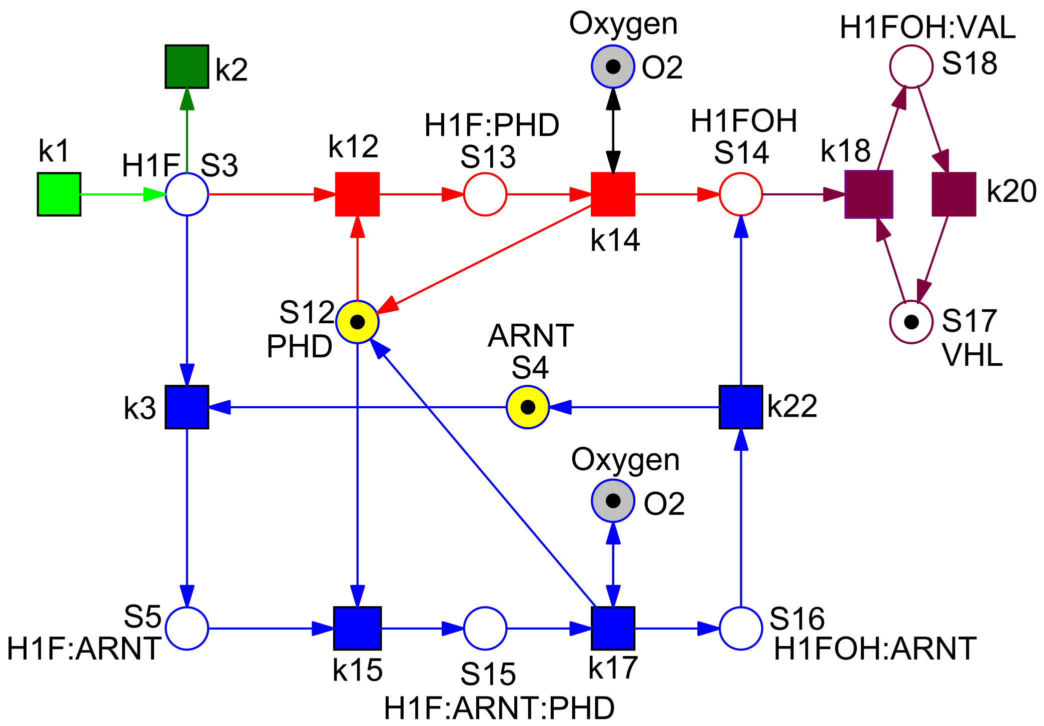
EX7 - HYPOXIA

[HEINER, SRIRAM 2010]

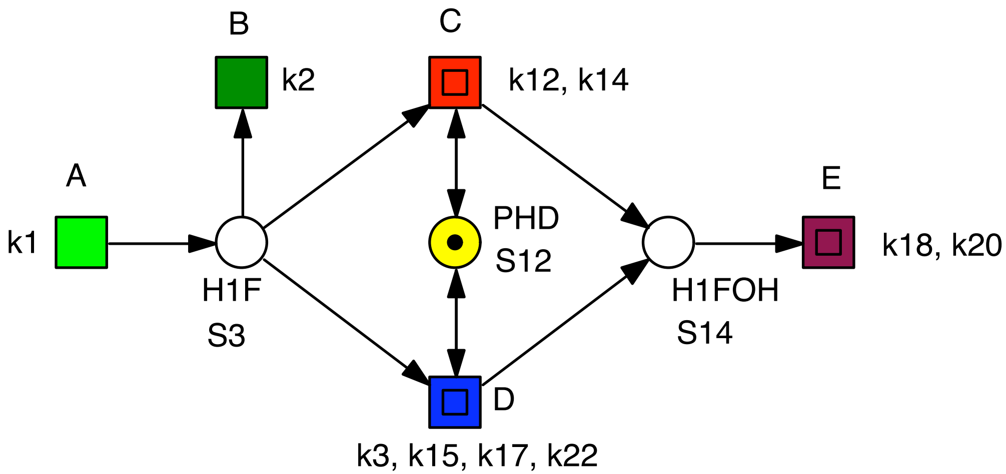



HIF (steady state values)
Oxygen
(a)
(b)
k5, k6, k29, k30
(c)
(b) + k19
(d)
(c) + k4, k21
(e)
(d) + k16
(f)
(e) + k13
0
50
100
150
0.5
1
1.5
2

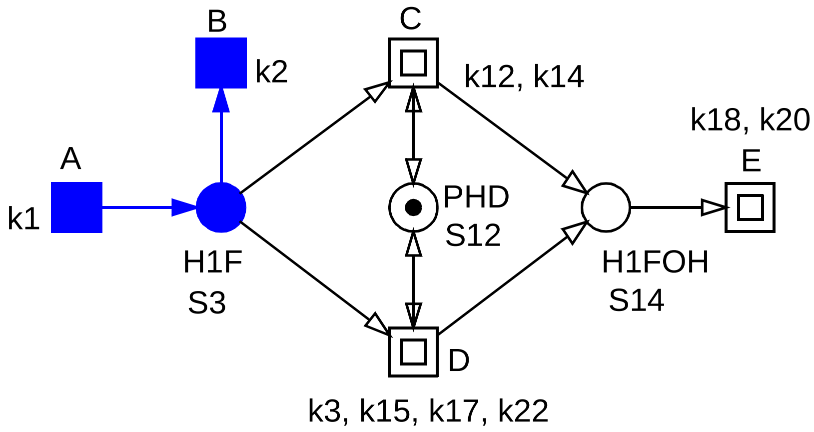
EX7 - HYPOXIA

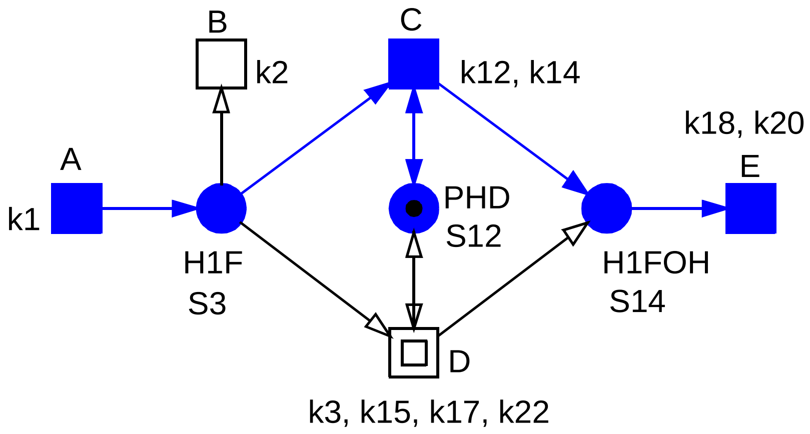

Oxygen
O2
H1FOH:VAL
S18
k2
H1F:PHD
S13
H1FOH
S14
k1
H1F
S3
k12
k18
k20
k14
S12
PHD
ARNT
S4
S17
VHL
k3
k22
Oxygen
O2
S5
H1F:ARNT
k15
S15
H1F:ARNT:PHD
k17
S16
H1FOH:ARNT

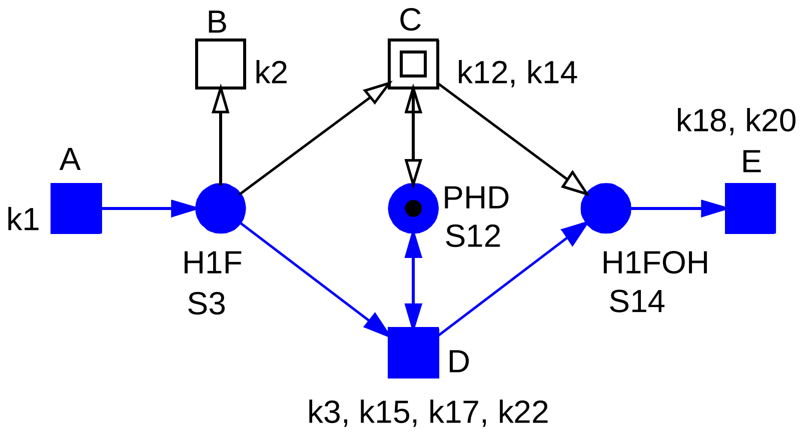
EX7 - HYPOXIA


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

Ex7 - Hypoxia

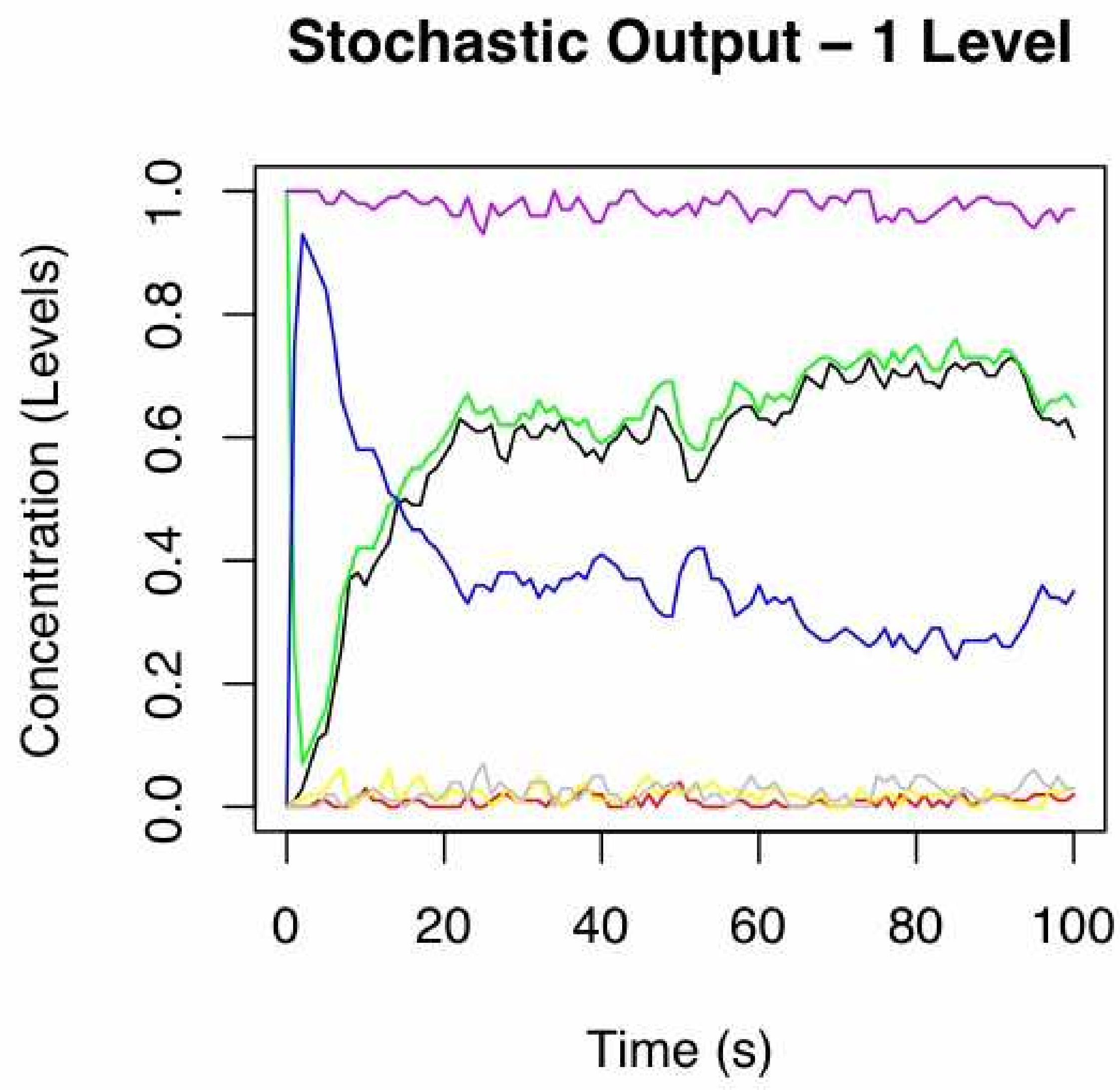

B
C
k2
k12, k14
k18, k20
E
A
k1
PHD
S12
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22

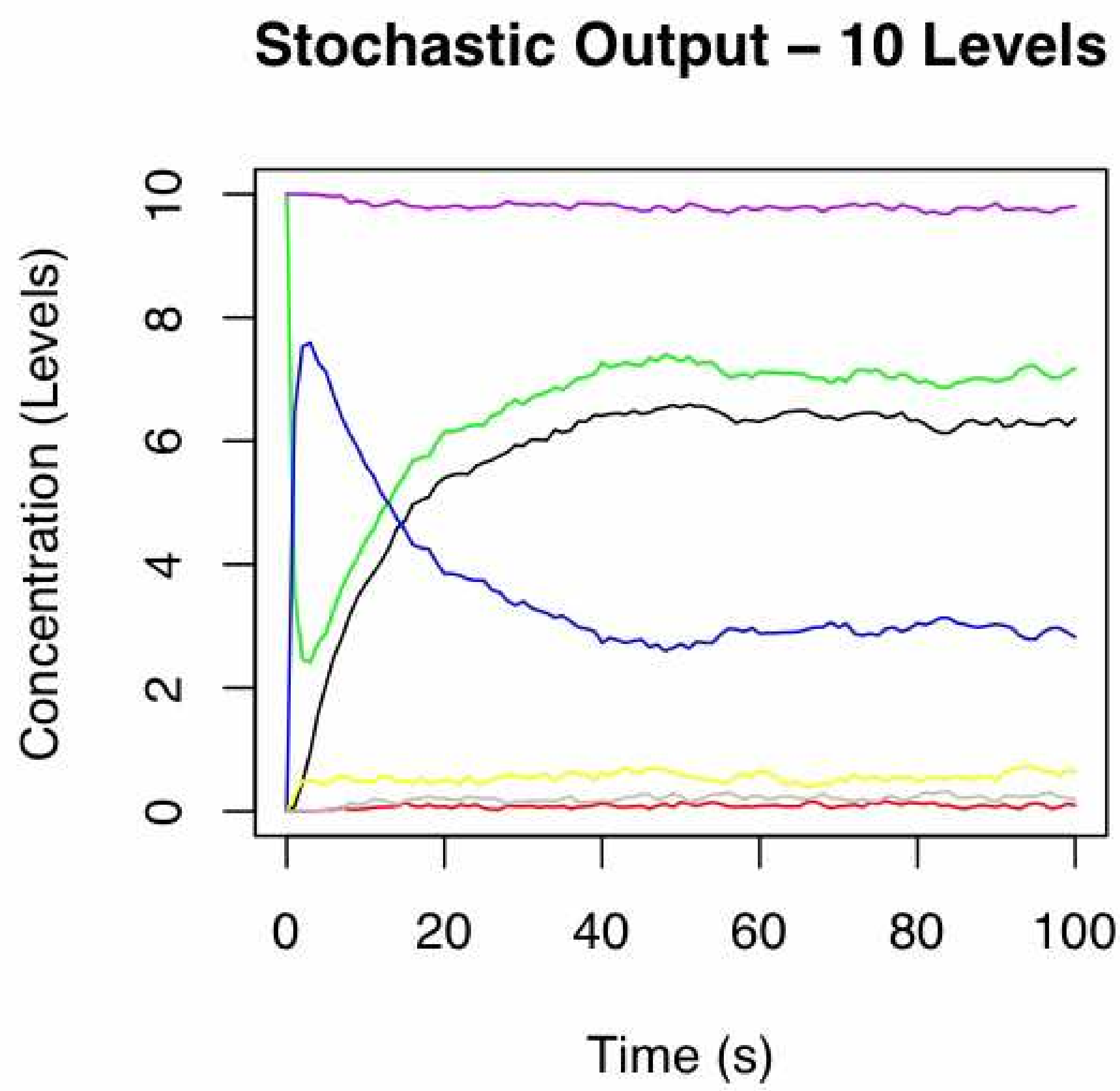

B
C
k2
k12, k14
k18, k20
E
A
k1
PHD
S12
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22

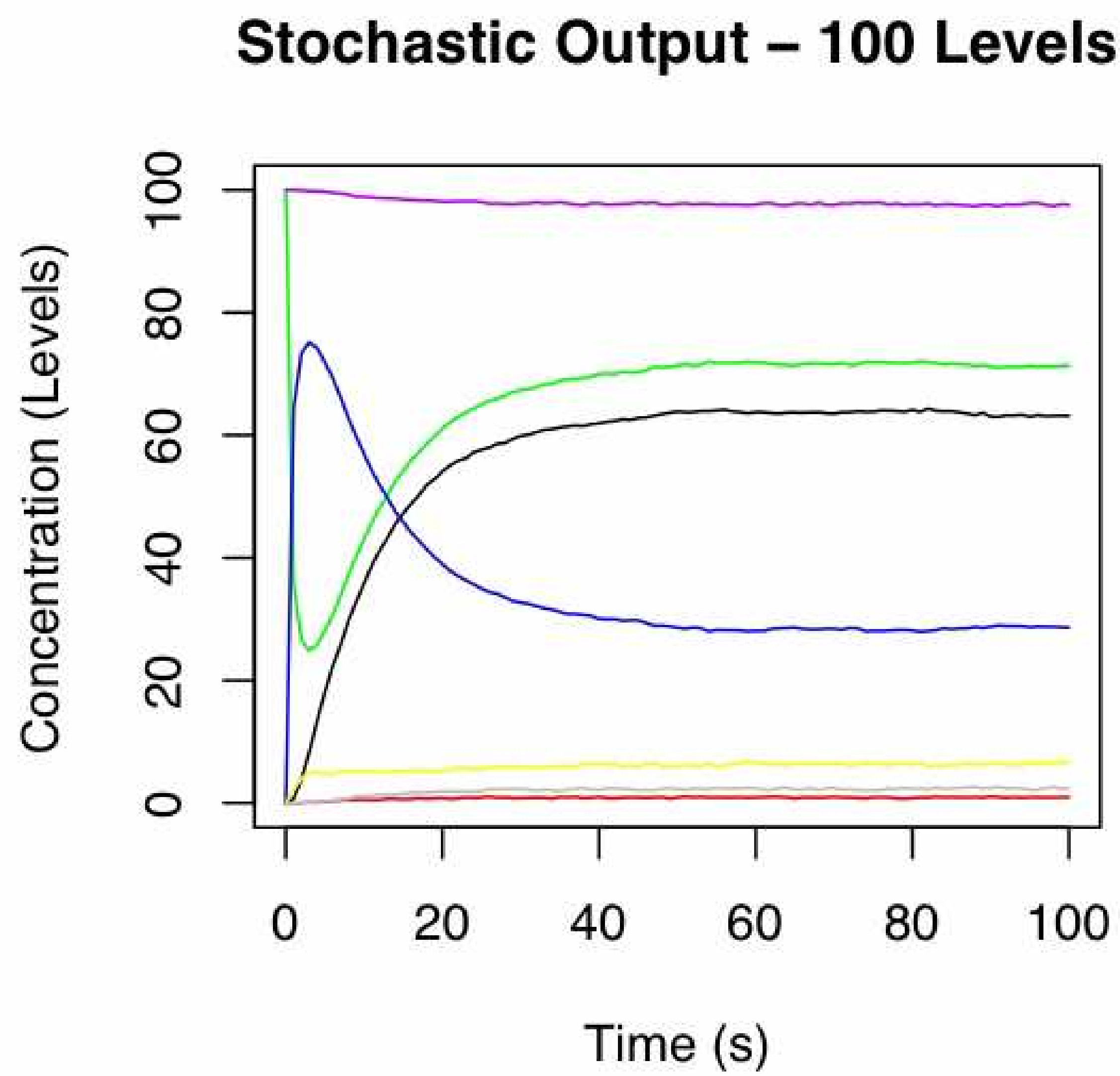

B
C
k2
k12, k14
k18, k20
E
A
k1
PHD
S12
H1F
S3
H1FOH
S14
D
k3, k15, k17, k22

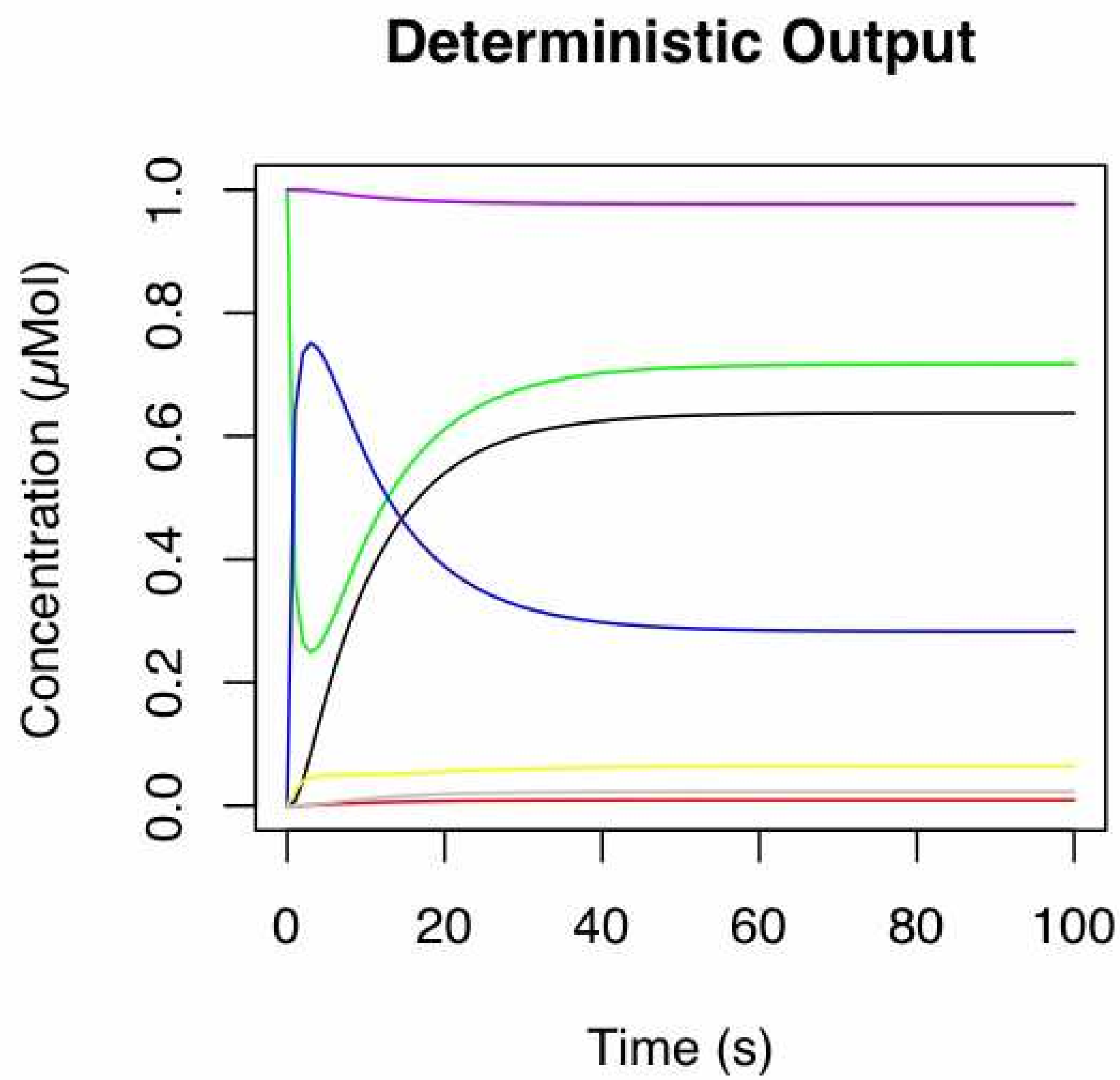
ABOUT THE RELATION
STOCHASTIC VS CONTINUOUS

monika.heiner@tu-cottbus.de


Stochastic Output – 1 Level
Concentration (Levels)
0.0
0.2
0.4
0.6
0.8
1.0
0
20
40
60
80
100
Time (s)

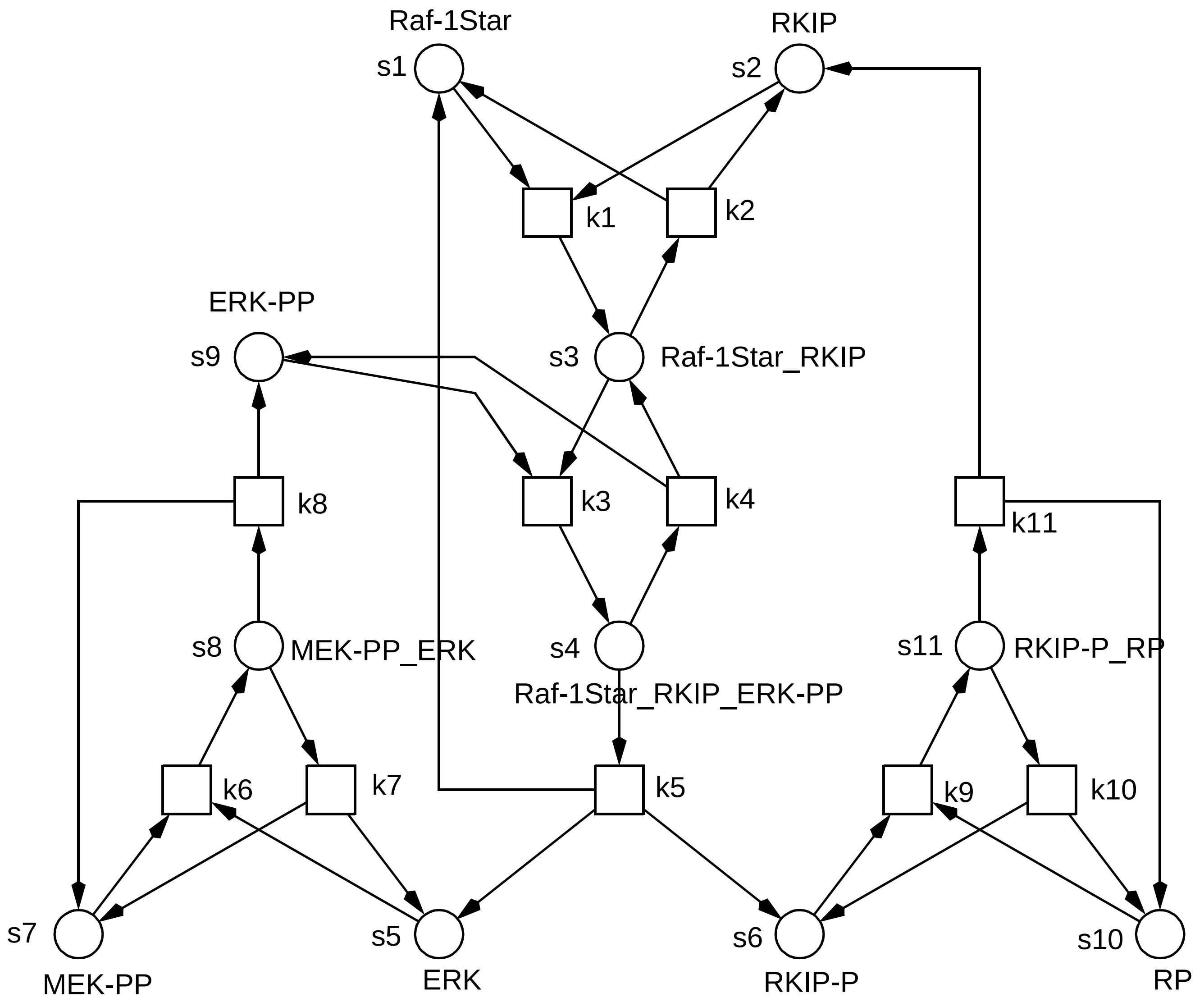

Stochastic Output – 10 Levels
Concentration (Levels)
0
2
4
6
8
10
0
20
40
60
80
100
Time (s)


Stochastic Output – 100 Levels
Concentration (Levels)
0
20
40
60
80
100
0
20
40
60
80
100
Time (s)



Deterministic Output
Concentration (μMol)
0.0
0.2
0.4
0.6
0.8
1.0
0
20
40
60
80
100
Time (s)

Ex3 - RKIP Signalling Pathway, Petri Net

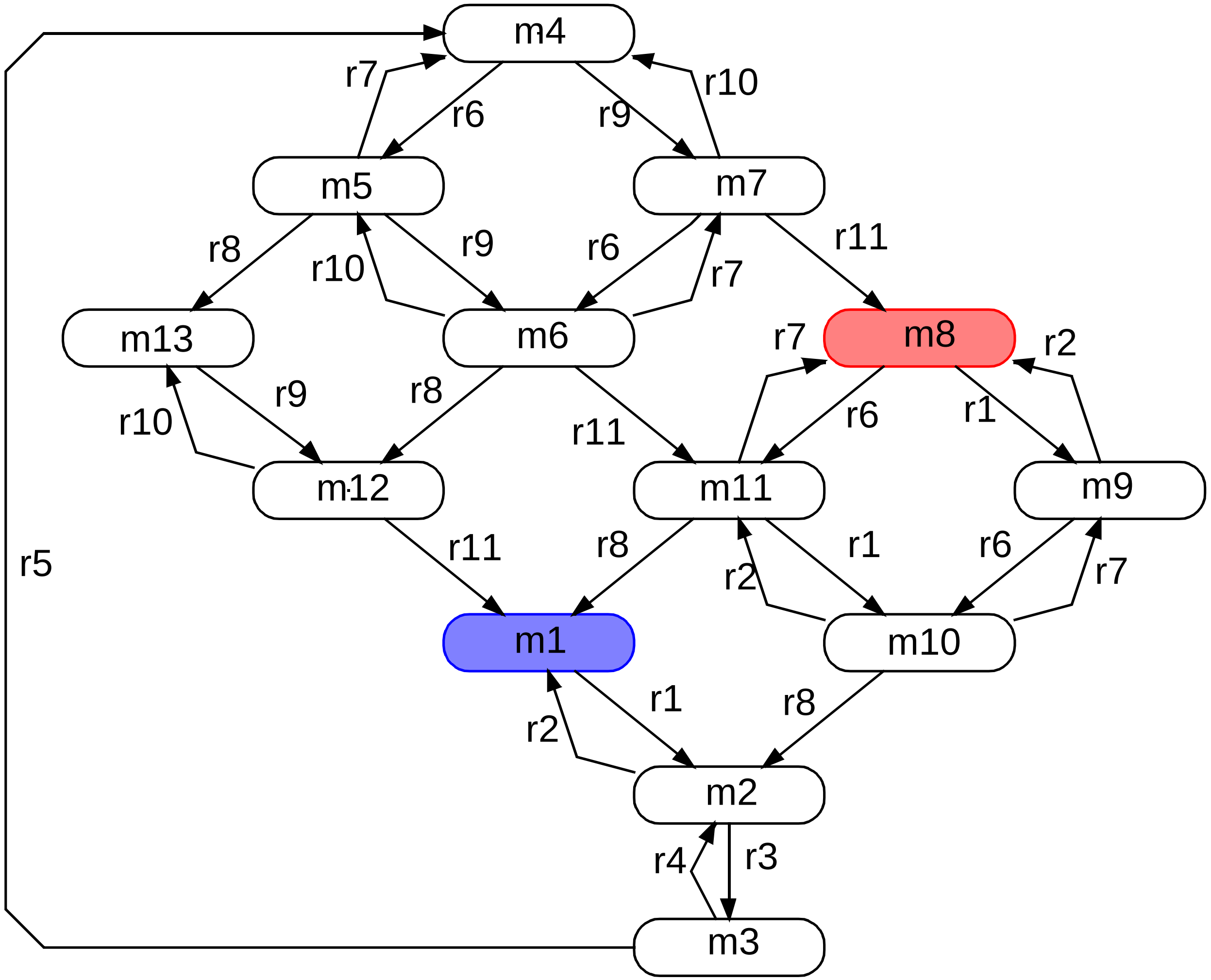


[Heiner, Gilbert 2006]

[Heiner, Donaldson, Gilbert 2010]

EX3 - RKIP, REACHABILITY GRAPH (STS)

- simple algorithm
- nodes : system states
- arcs : the (single) firing transition
- single step firing rule



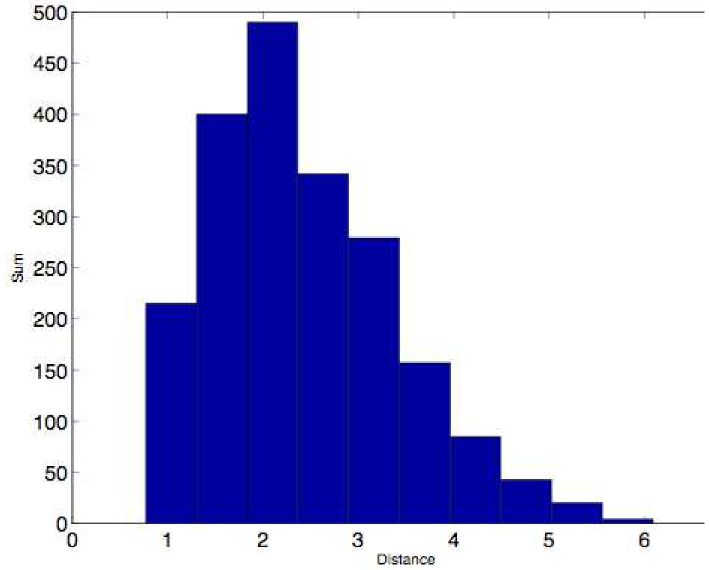
Ex3 - RKIP, Quantitative Analysis

Species	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	S11	S12	S13
Raf-1*	1	0	0	1	1	1	1	1	0	0	1	1	1
RKIP	1	0	0	0	0	0	0	1	0	0	1	0	0
Raf-1*_RKIP	0	1	0	0	0	0	0	0	1	1	0	0	0
Raf-1*_RKIP_ERK-PP	0	0	1	0	0	0	0	0	0	0	0	0	0
ERK	0	0	0	1	0	0	1	1	1	0	0	0	0
RKIP-P	0	0	0	1	1	0	0	0	0	0	0	0	1
MEK-PP	1	1	1	1	0	0	1	1	1	0	0	1	1
MEK-PP_ERK	0	0	0	0	1	1	0	0	0	1	1	0	0
ERK-PP	1	1	0	0	0	0	0	0	0	0	0	1	1
RP	1	1	1	1	1	0	0	1	1	1	1	0	1
RKIP-P_RP	0	0	0	0	0	1	1	0	0	0	0	1	0

Cho et al

Biochemist

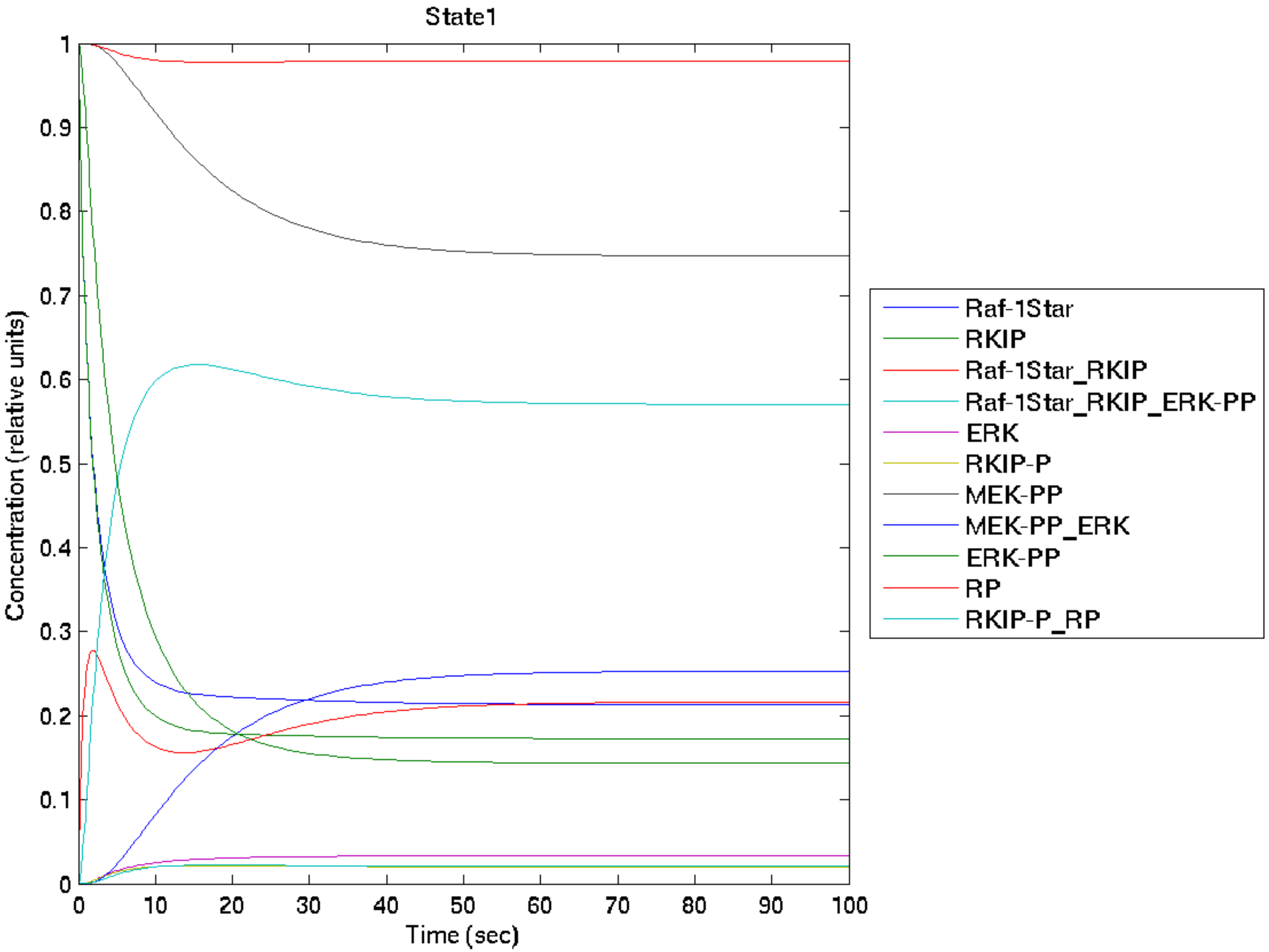
13 good state configurations



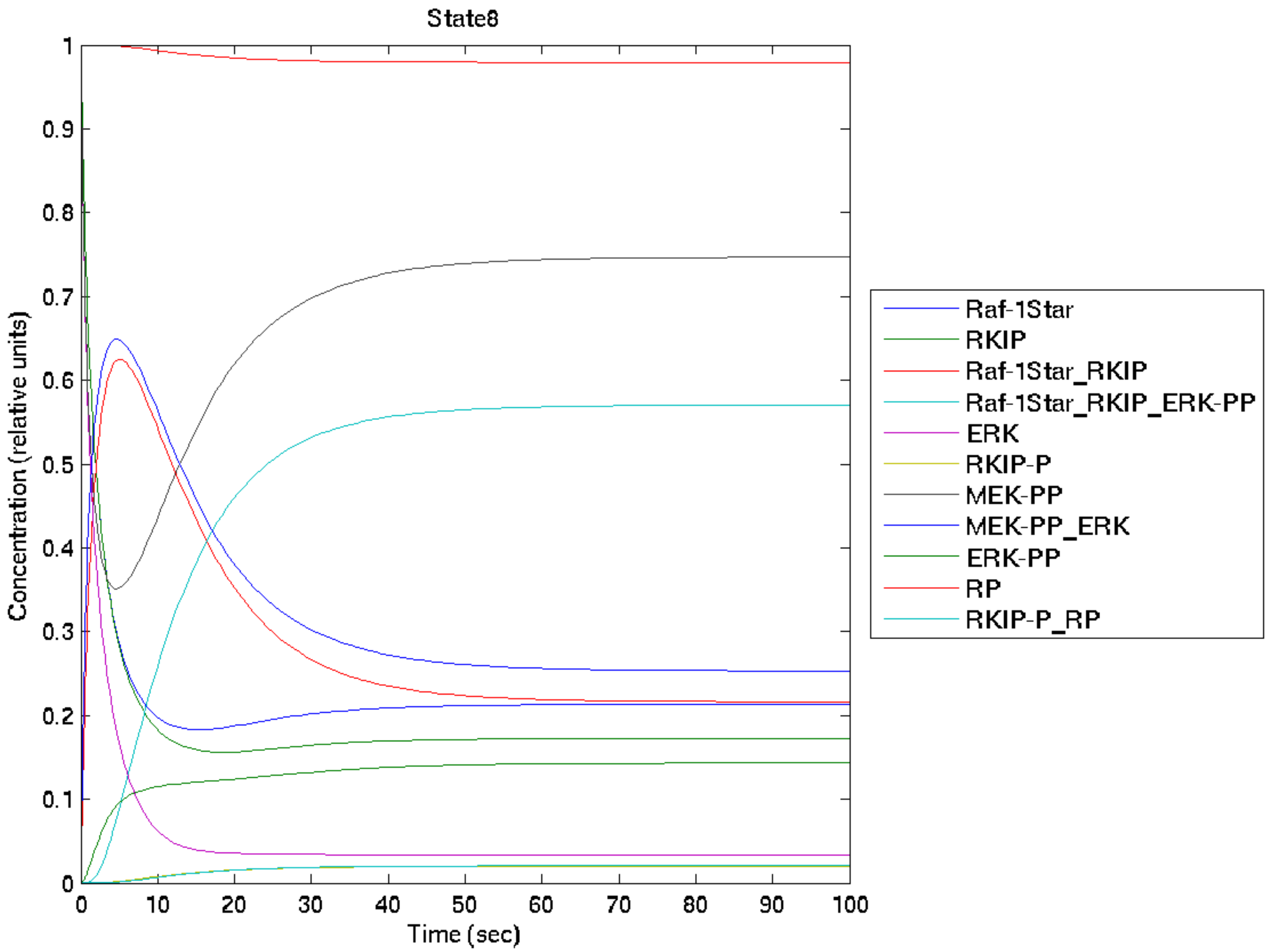
Distribution of `bad' steady states as euclidean distances from the `good' final steady state

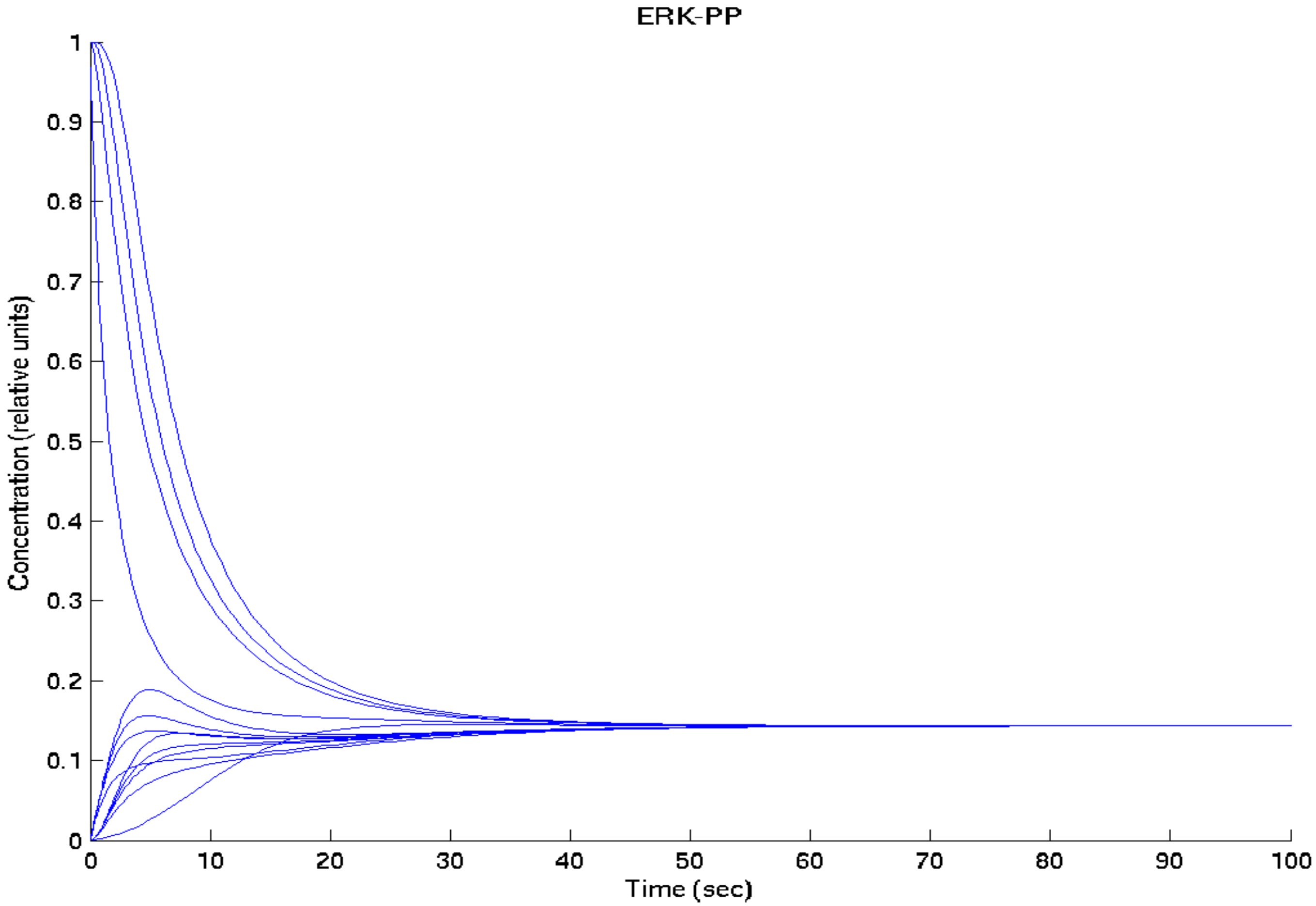
the bad ones

Ex3 - RKIP, Quantitative Analysis



Ex3 - RKIP, Quantitative Analysis




ERK-PP
Concentration (relative units)
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
0
10
20
30
40
50
60
70
80
90
100
Time (sec)

BUT,

TRANSITION SPN -> CPN

MAY COME WITH

COUNTERINTUITIVE EFFECTS.

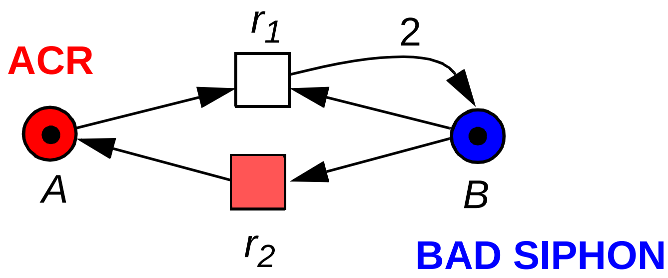
EX8 - ABSOLUT CONCENTRATION ROBUSTNESS (ACR)

- **ACR: steady state value of variable (place) does not depend on total mass, only on kinetic constants** -> [SHINAR, FEINBERG 2010]

- **simple example** **mass-action kinetics**

  r_1: $A + B \rightarrow 2B$ $v_1(r_1) = k_1 AB$

  r_2: $B \rightarrow A$ $v_2(r_2) = k_2 B$



- **ODEs**

  $dA/dt = v_2 - v_1 = k_2 B - k_1 AB$

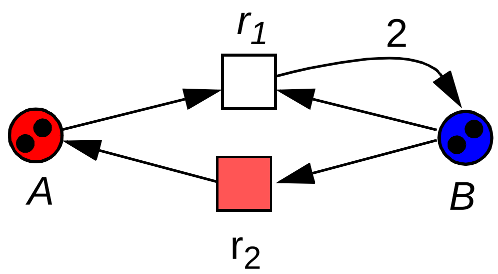
  $dB/dt = v_1 - v_2 = k_1 AB - k_2 B$

CPI: $m_0(A) + m_0(B) = total$

- **steady state**

  $dA/dt = k_2 B - k_1 AB = 0$

  $dB/dt = k_1 AB - k_2 B = 0$



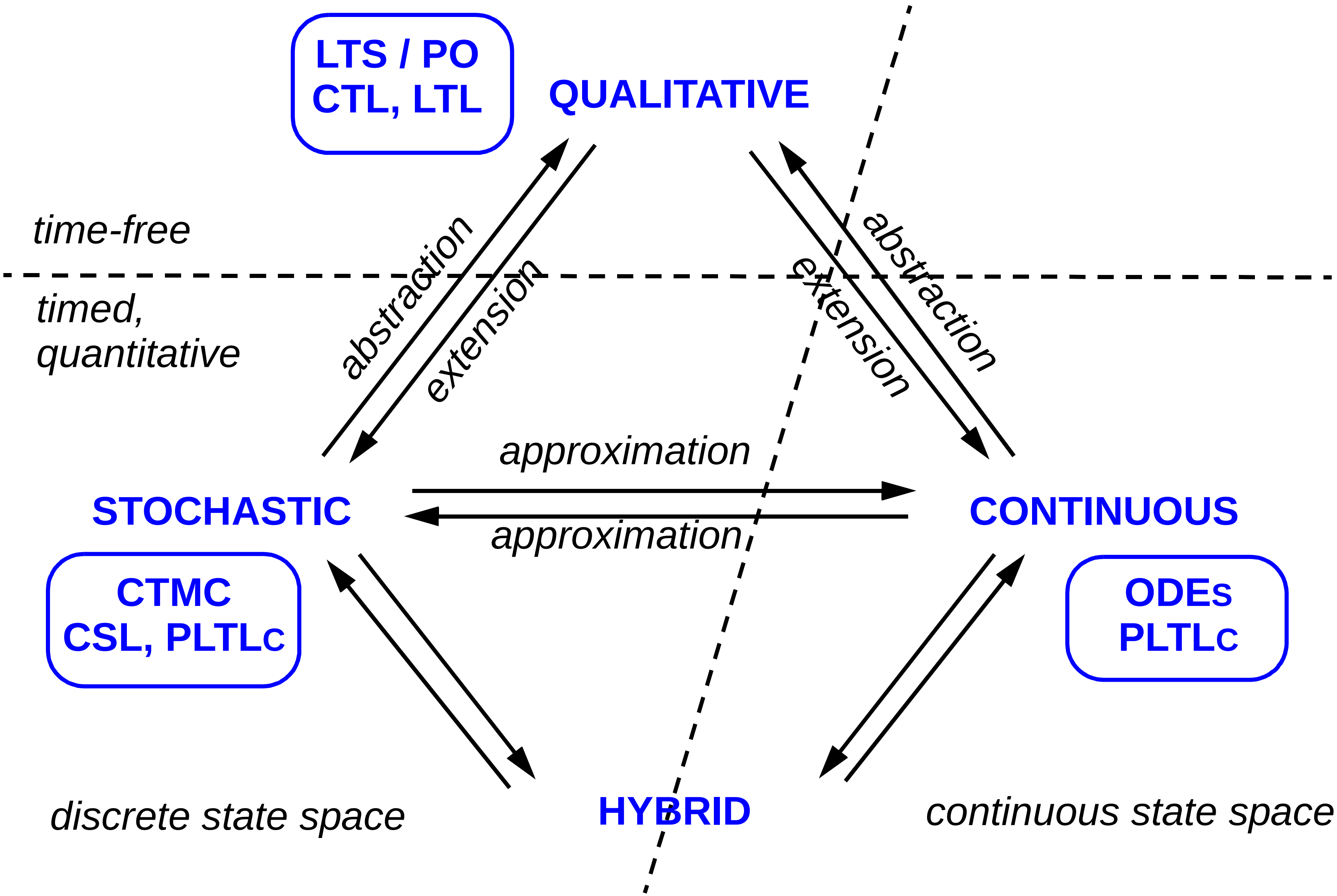
-> $steady\_state(A) = k_2/k_1$

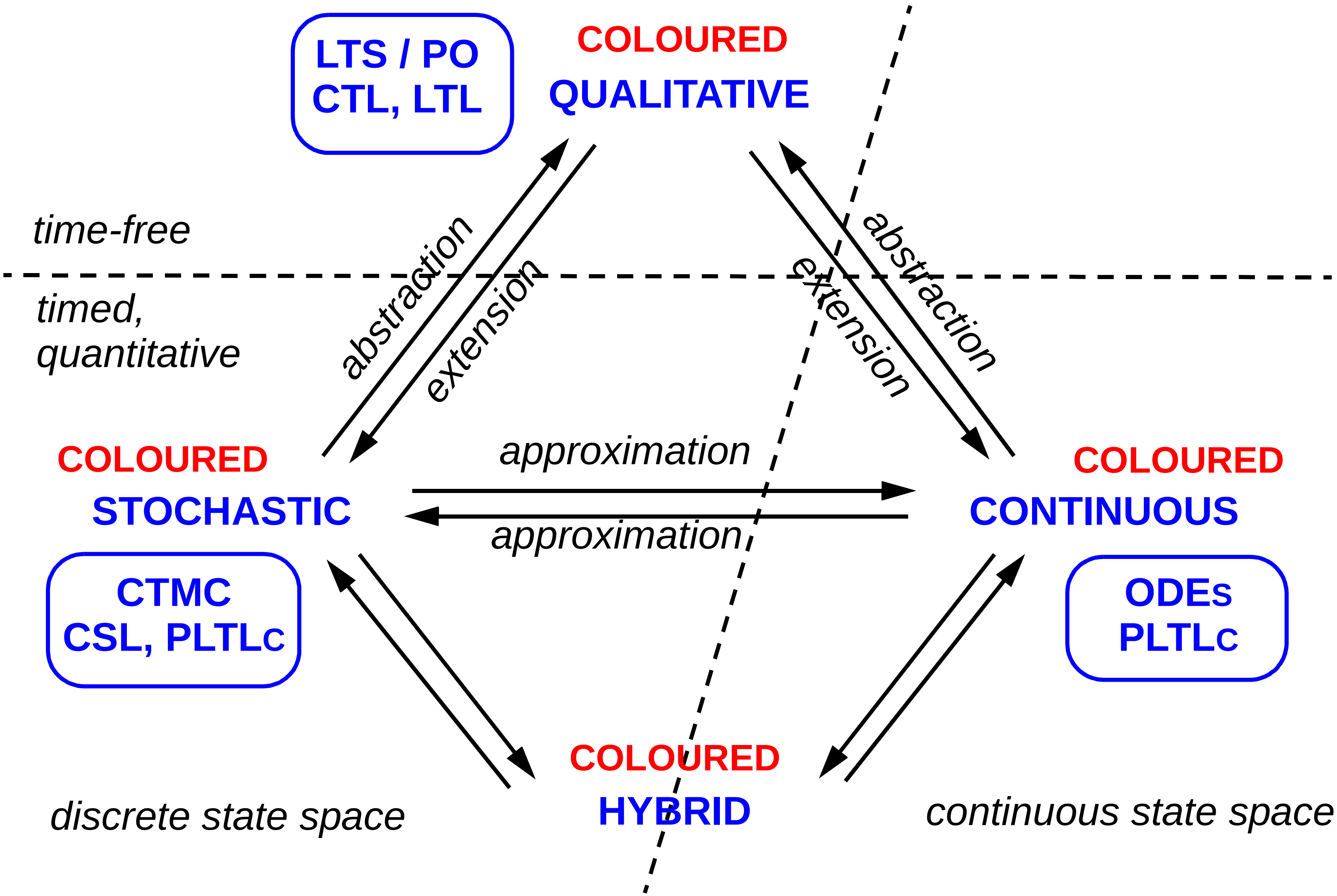
$steady\_state(B) = total - k_2/k_1$

. . . AND THEN THERE WAS COLOUR

Kew Gardens, 24/04/2011

FRAMEWORK 2010



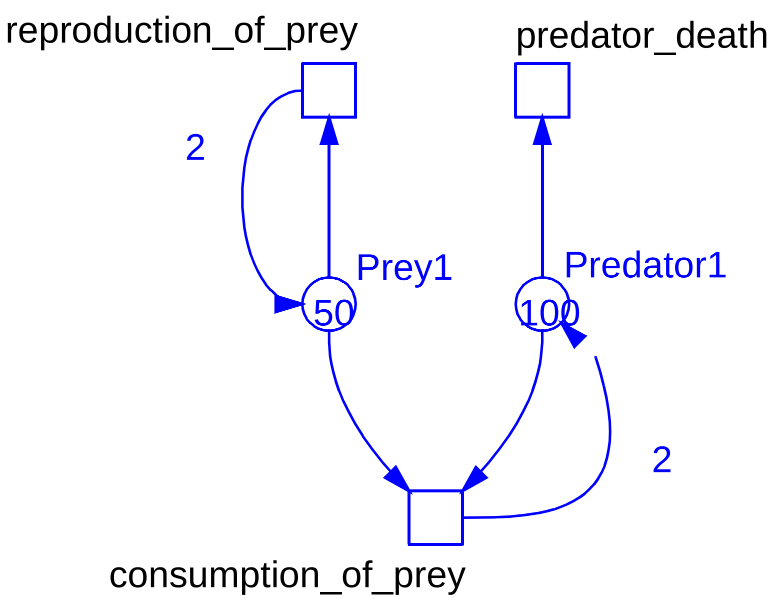

LTS / PO
CTL, LTL
COLOURED
QUALITATIVE
time-free
timed,
quantitative
abstraction
extension
abstraction
extension
COLOURED
STOCHASTIC
approximation
approximation
COLOURED
CONTINUOUS
CTMC
CSL, PLTLc
ODEs
PLTLc
COLOURED
HYBRID
discrete state space
continuous state space

COLOUR -

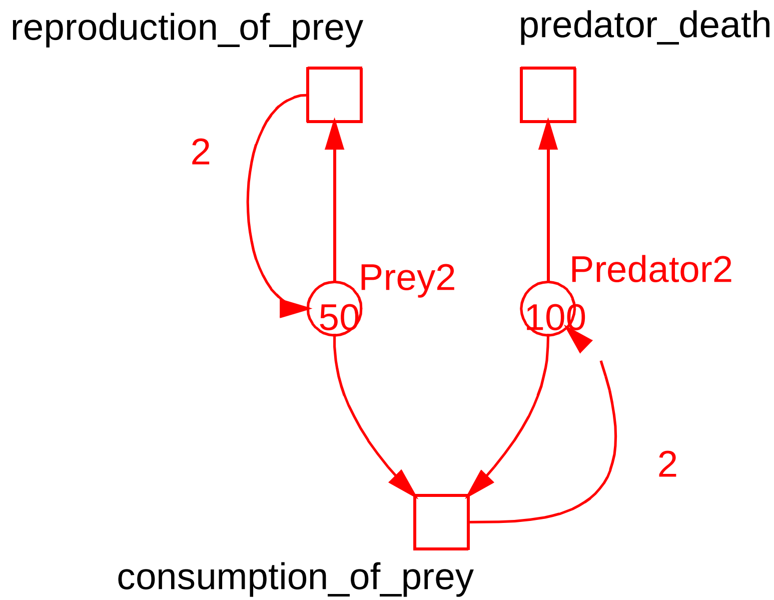
WHAT FOR ?

Ex1: Prey - Predator

sub-system1



sub-system2



EX1: PREY - PREDATOR

- **definitions**

  colourset *CS = 1-2;*

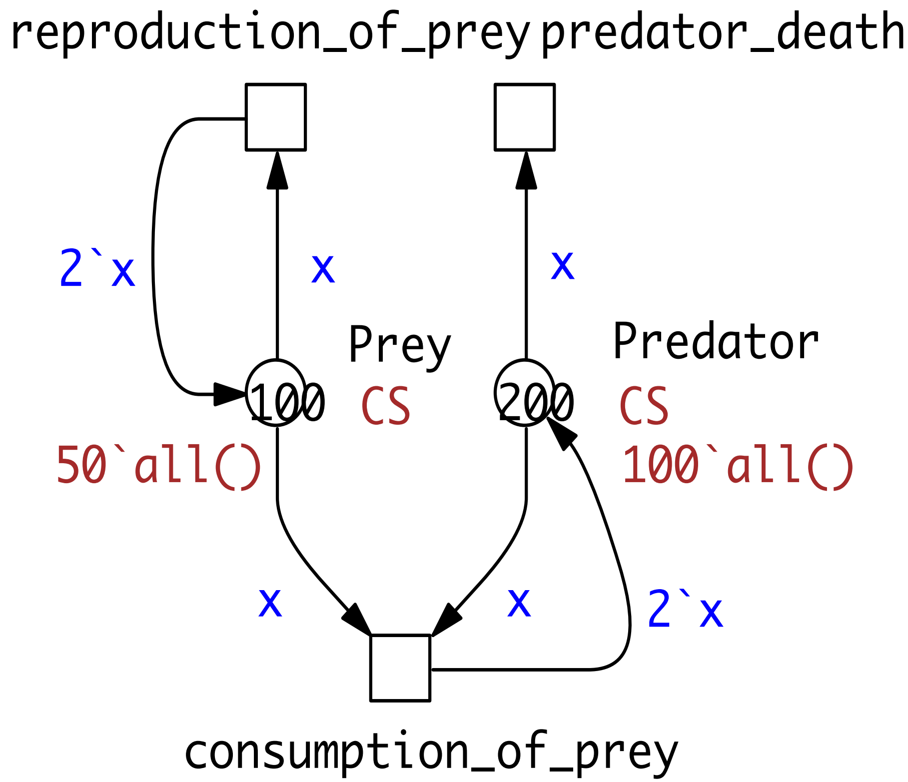
  var *x : CS;*

- **better:**

  const *SIZE = 2;*

  colourset *CS = 1-SIZE;*

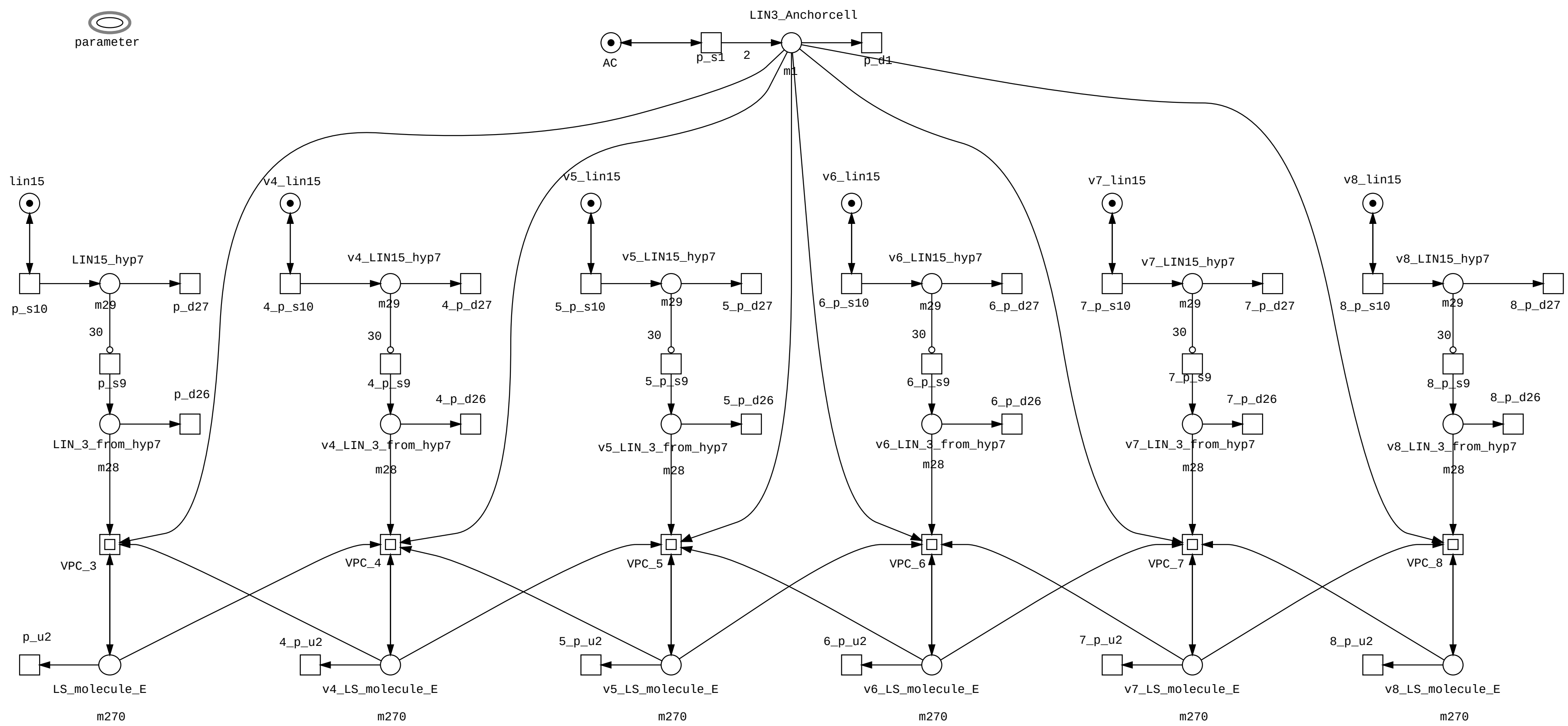
  var *x : CS;*



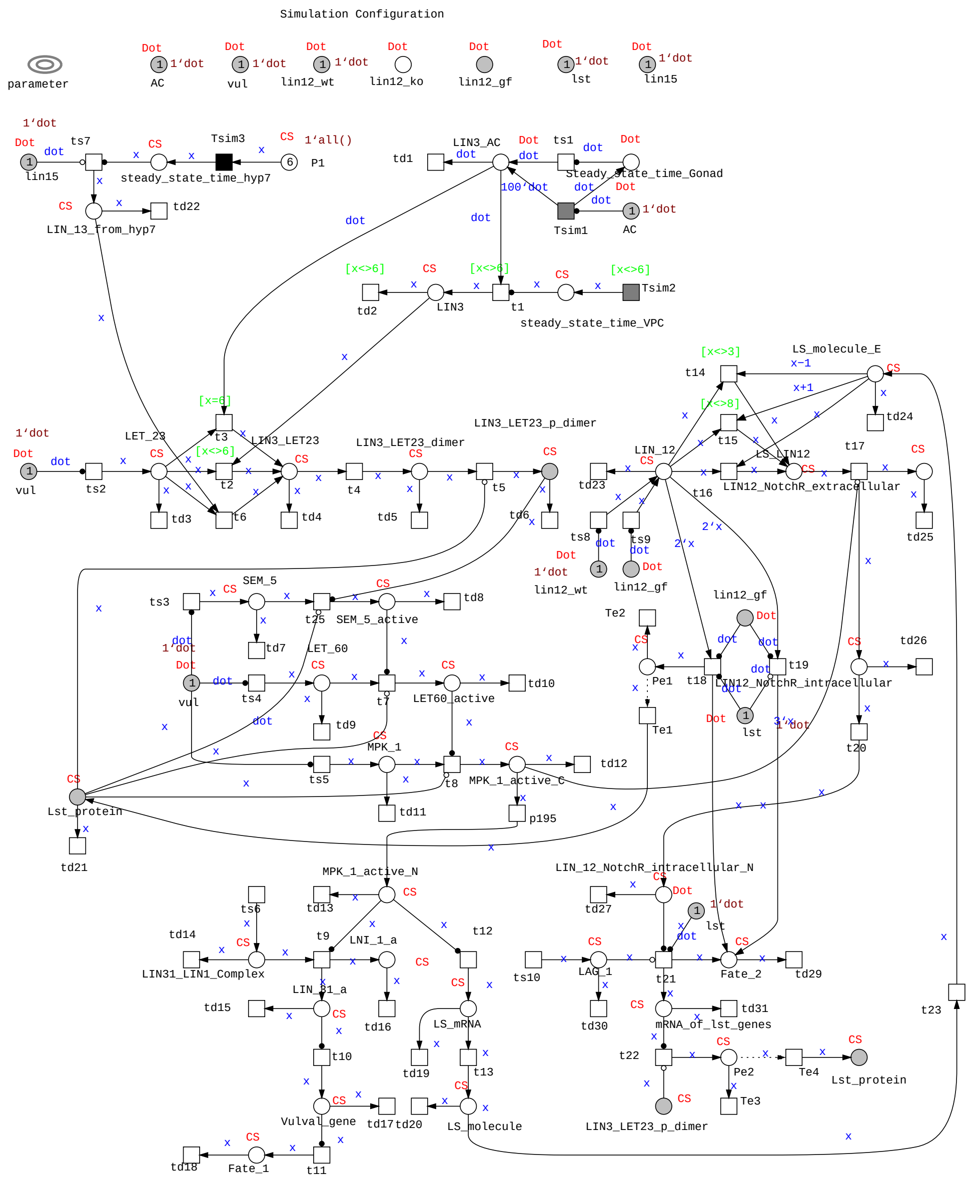
- **changing SIZE adapts the model to various scenarious**

[Li et al. 2009]
[Bonzanni et al. 2009]

PLACES: 206
TRANSITIONS: 366

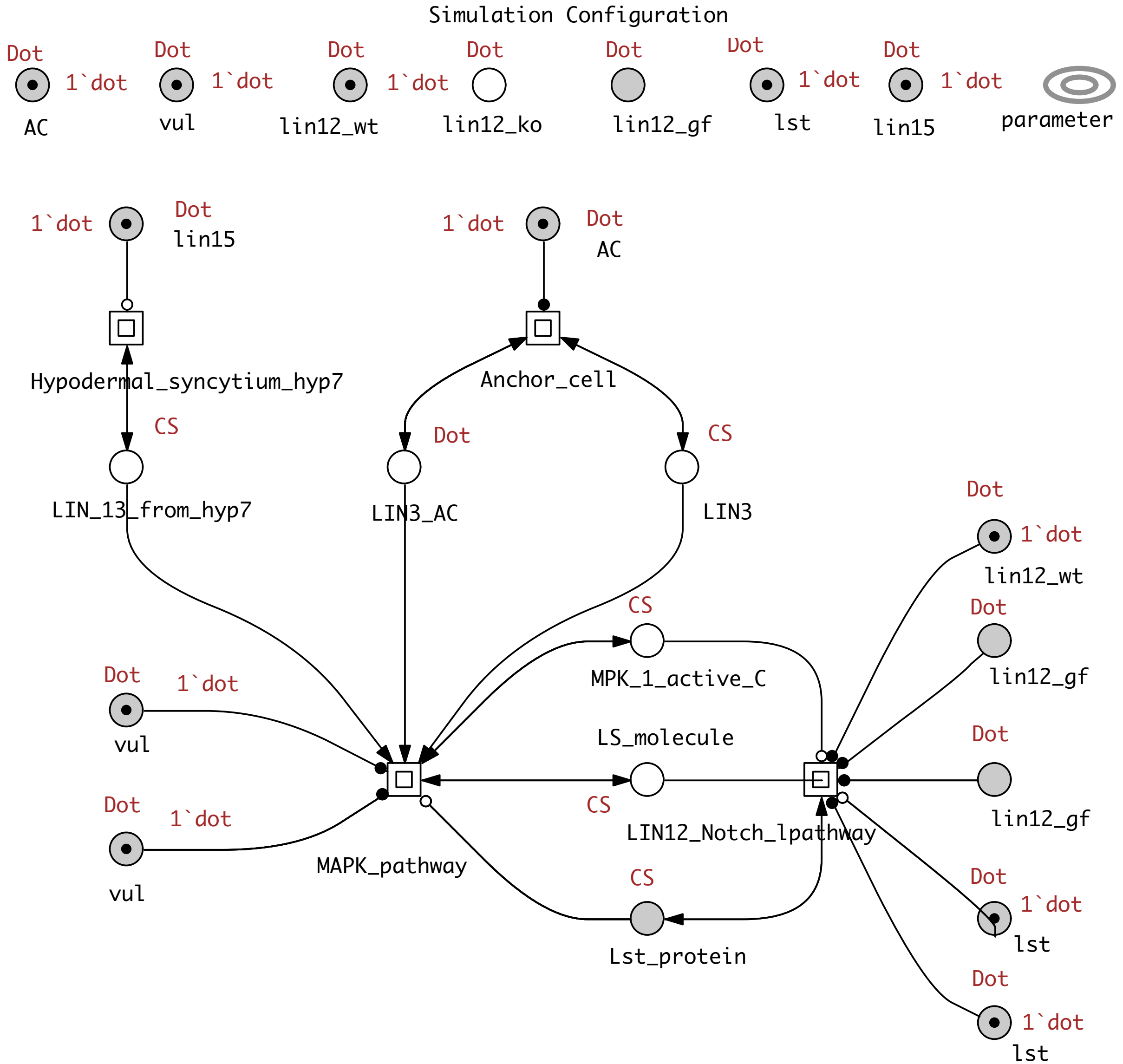


Ex2: C. Elegans, Coloured



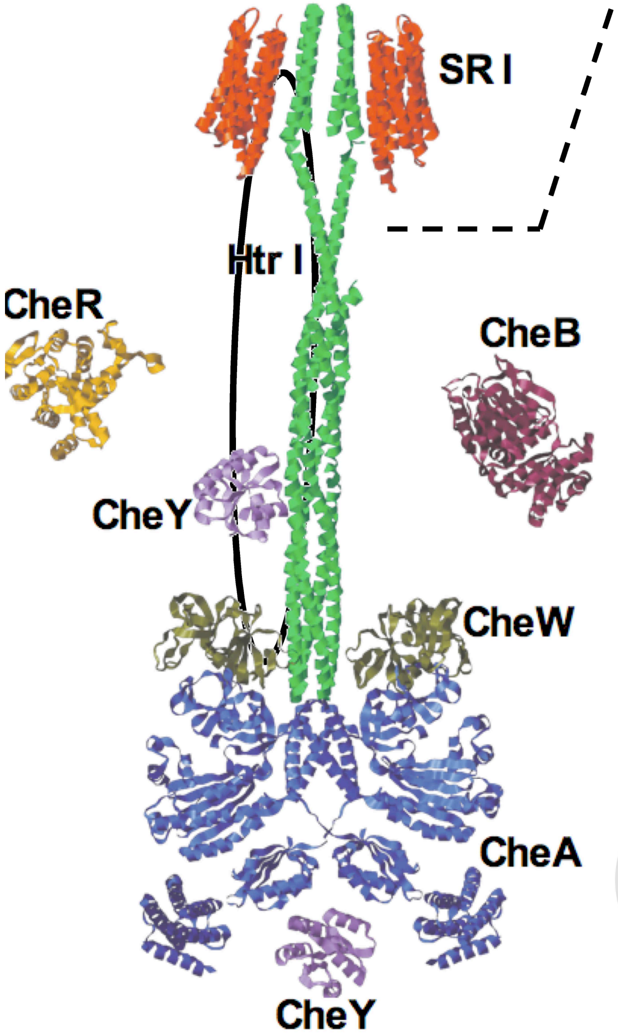
PLACES: 44
TRANSITIONS: 72

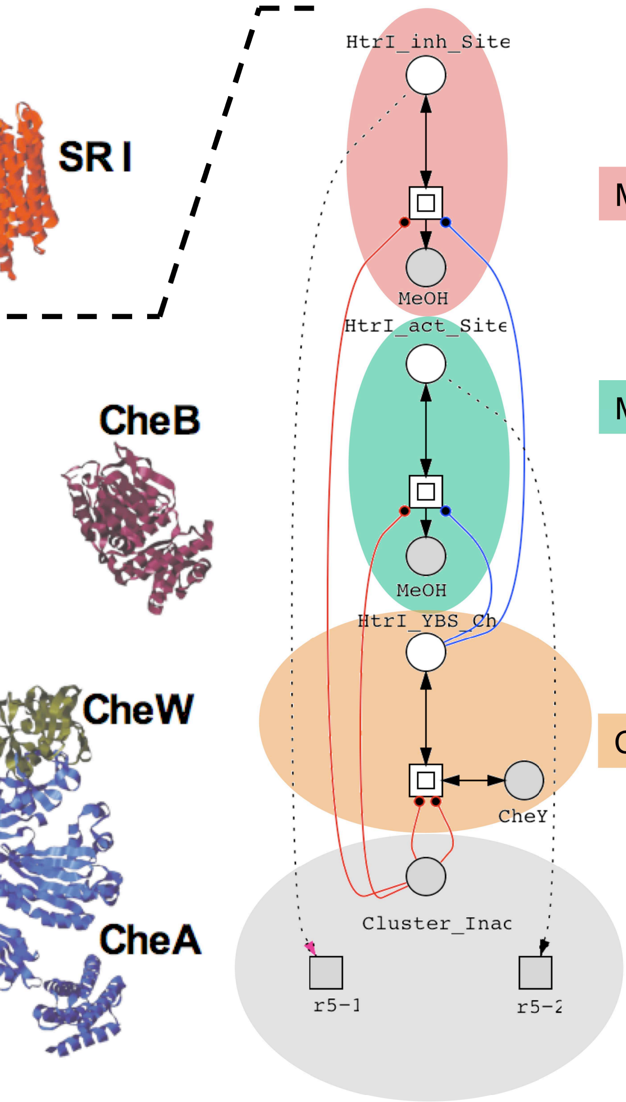
Ex2: C. Elegans, Coloured & Hierarchies

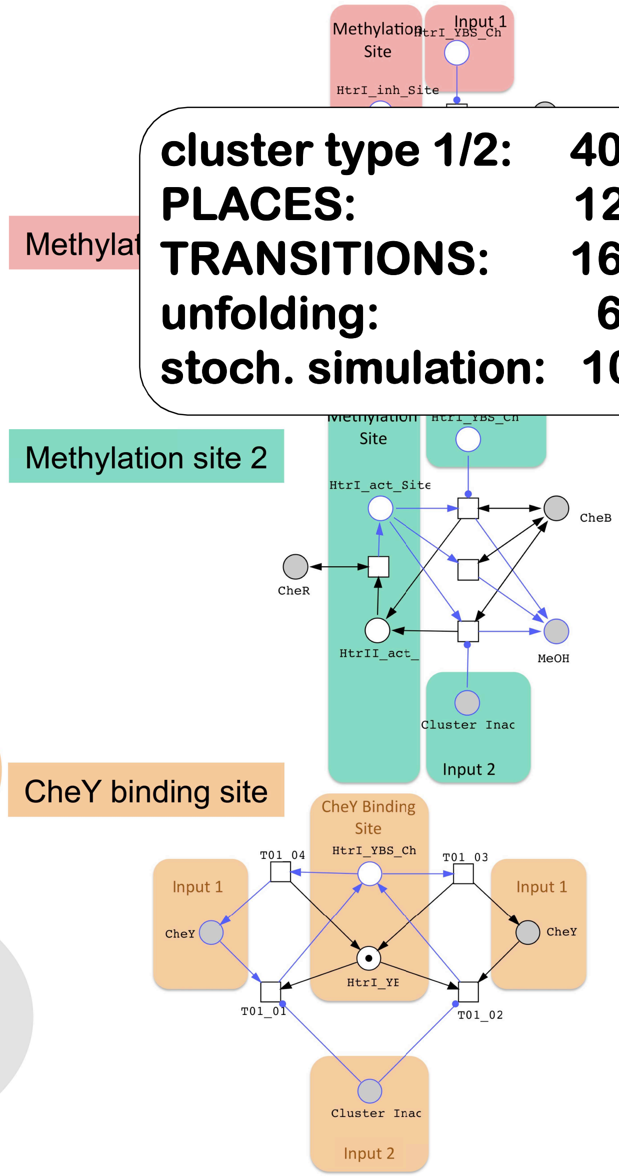


Ex3 - Halobacterium Salinarum

[Marwan 2010]





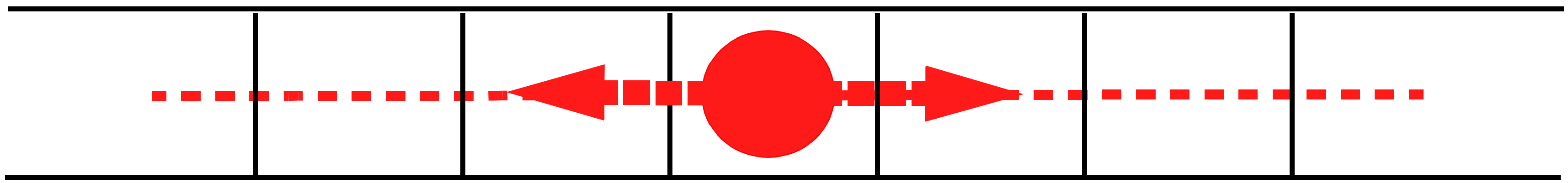


cluster type 1/2:	400/850
PLACES:	12,426
TRANSITIONS:	16,577
unfolding:	6 sec
stoch. simulation:	10-15h

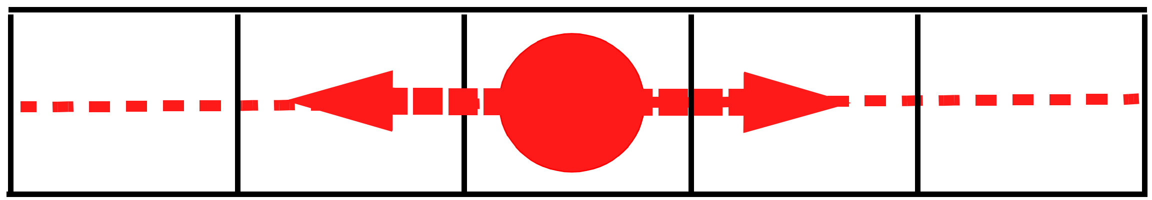
EXAMPLE: DIFFUSION IN SPACE

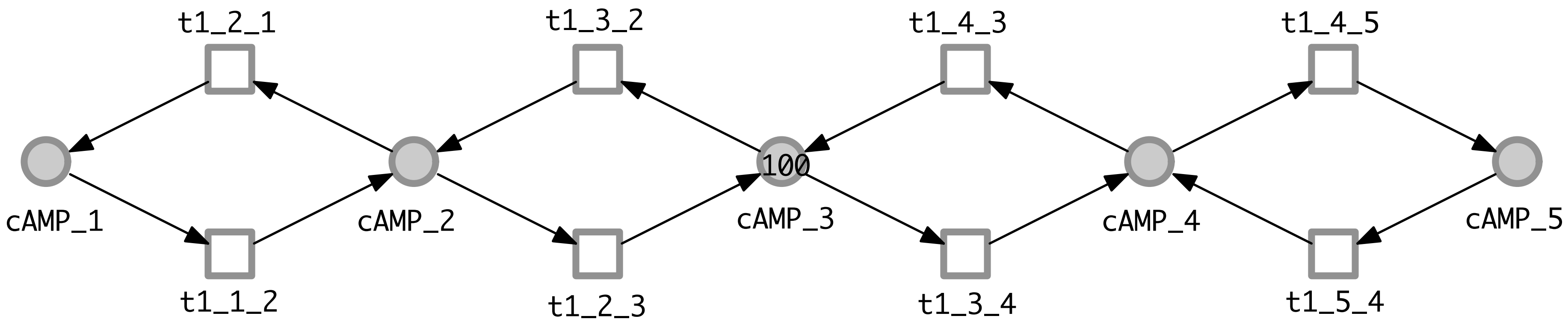
Richmond, 13/09/2011

EX4: DIFFUSION - 1D



EX4: DIFFUSION - 1D





EX4: DIFFUSION - 1D

- **definitions**

const *D1 = 5;* *// grid size*

const *MIDDLE = D1/2;*

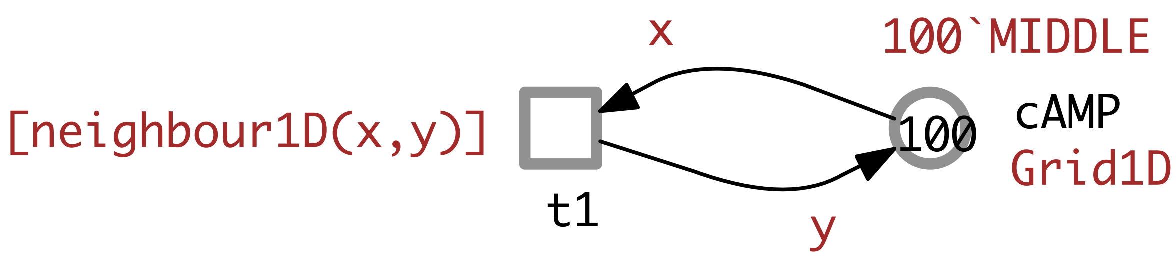
colorset *CS = 1-D1;* *// grid positions*

var *x,y : CS;*

function *neighbour1D (CS x,a)* ***bool****:*

// a is neighbour of x

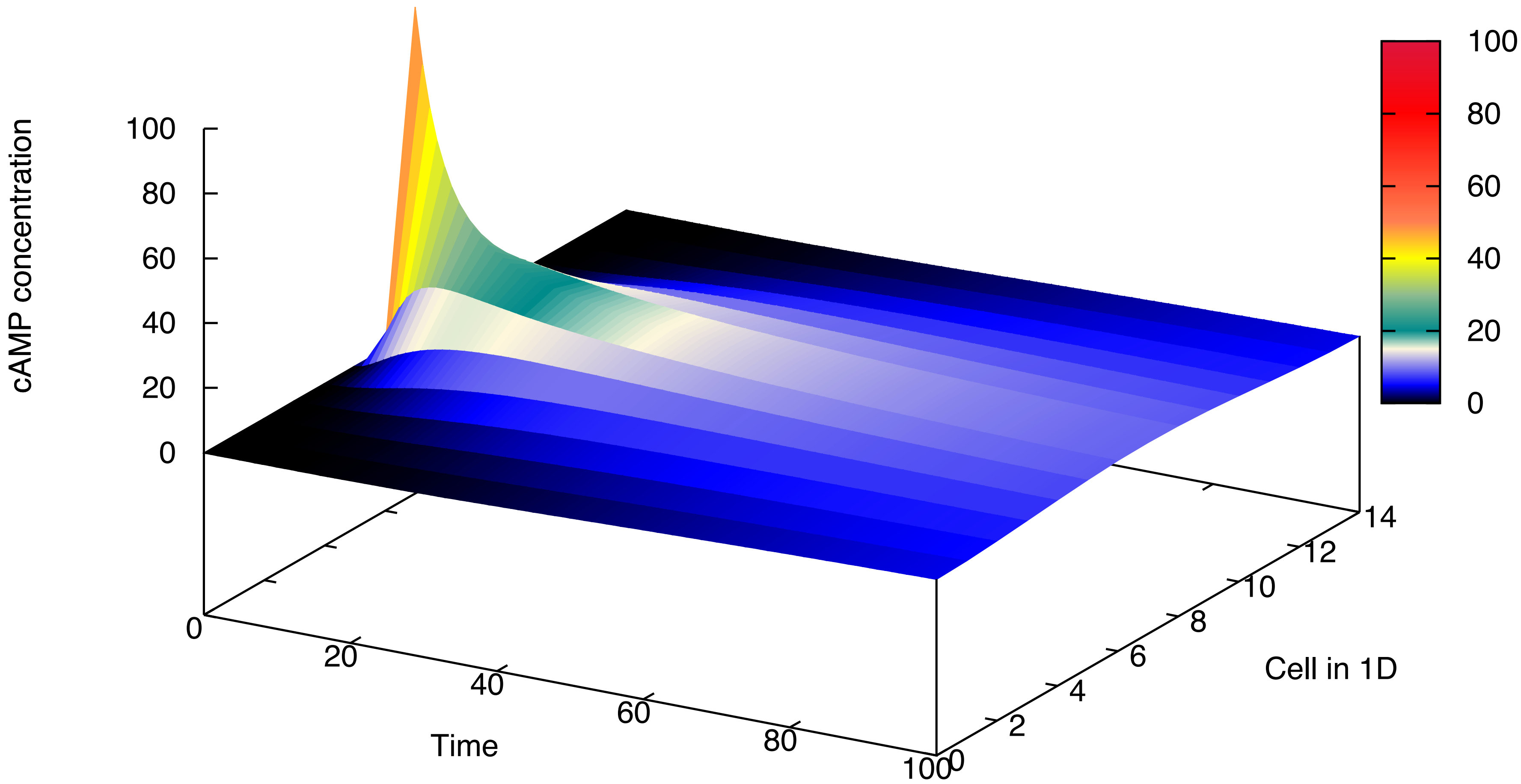
(a=x-1 | a=x+1) & (1<=a) & (a<=D1);



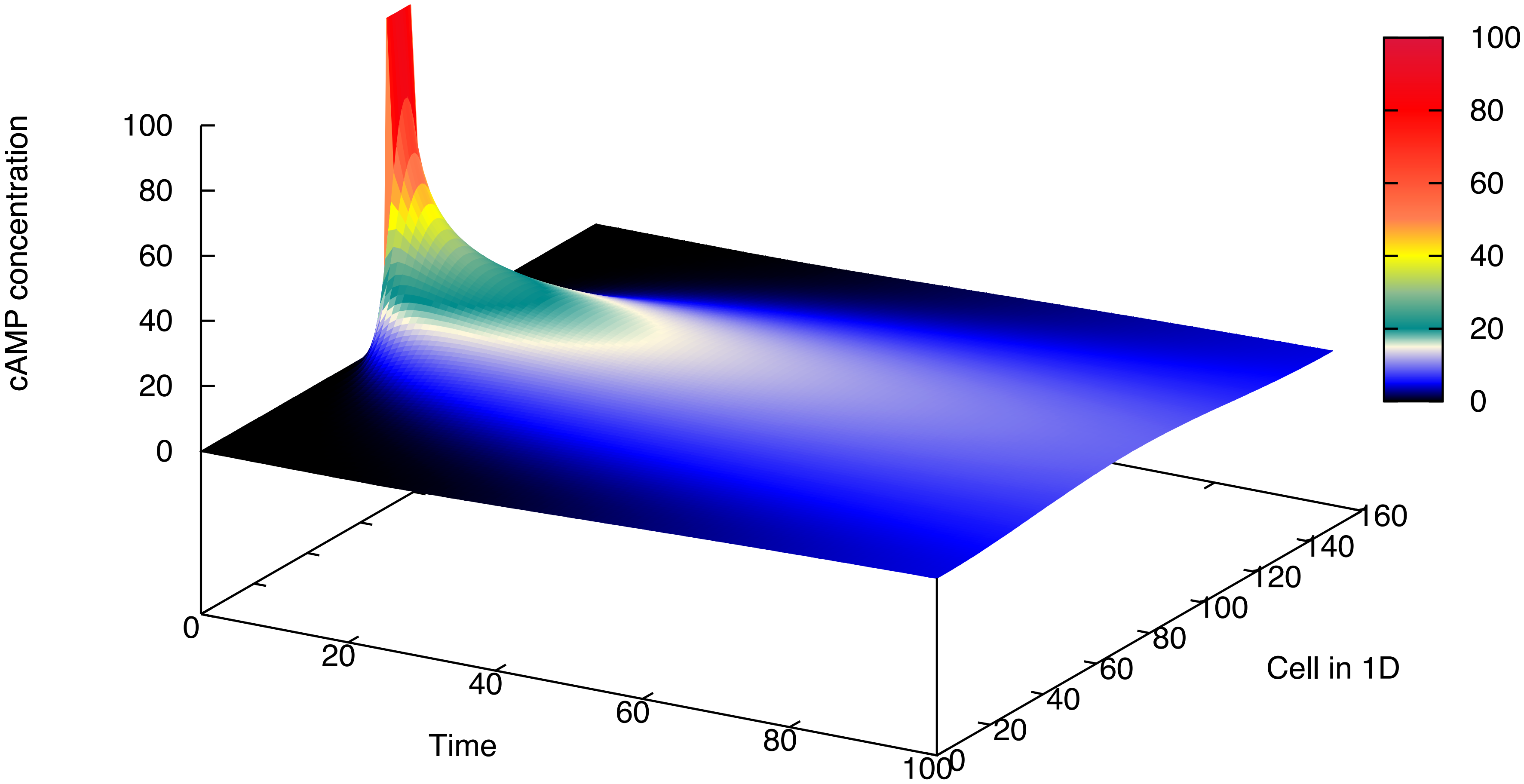
- **movement = changing colour**

$$
\begin{aligned}
\frac{dc_1}{dt} &= k \cdot c_2 - k \cdot c_1 \\
\frac{dc_2}{dt} &= k \cdot c_1 + k \cdot c_3 - 2 \cdot k \cdot c_2 \\
\frac{dc_3}{dt} &= k \cdot c_2 + k \cdot c_4 - 2 \cdot k \cdot c_3 \\
\frac{dc_4}{dt} &= k \cdot c_3 + k \cdot c_5 - 2 \cdot k \cdot c_4 \\
\frac{dc_5}{dt} &= k \cdot c_4 - k \cdot c_5
\end{aligned}
$$

Ex4: Diffusion - 1D



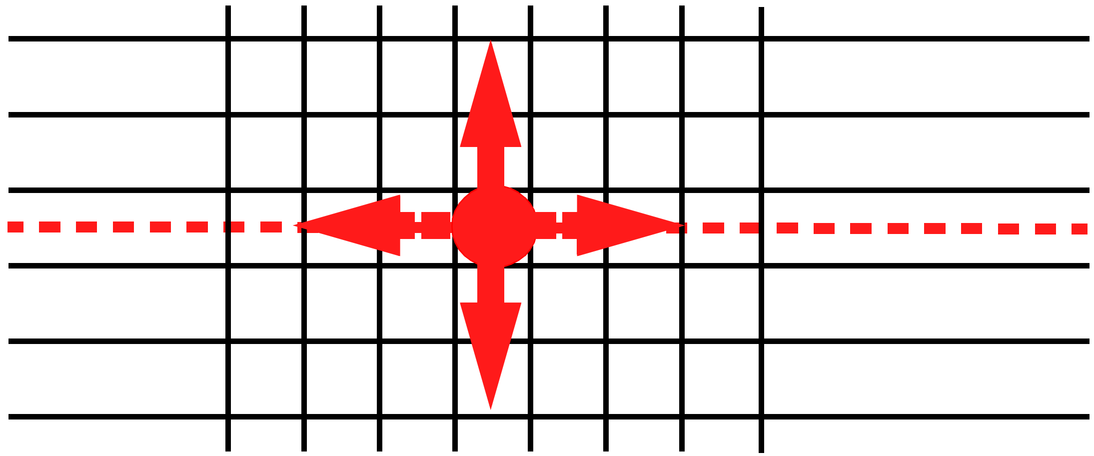
15 Grid Positions



150 GRID POSITIONS, SCALING OF INITIAL MARKING AND RATES

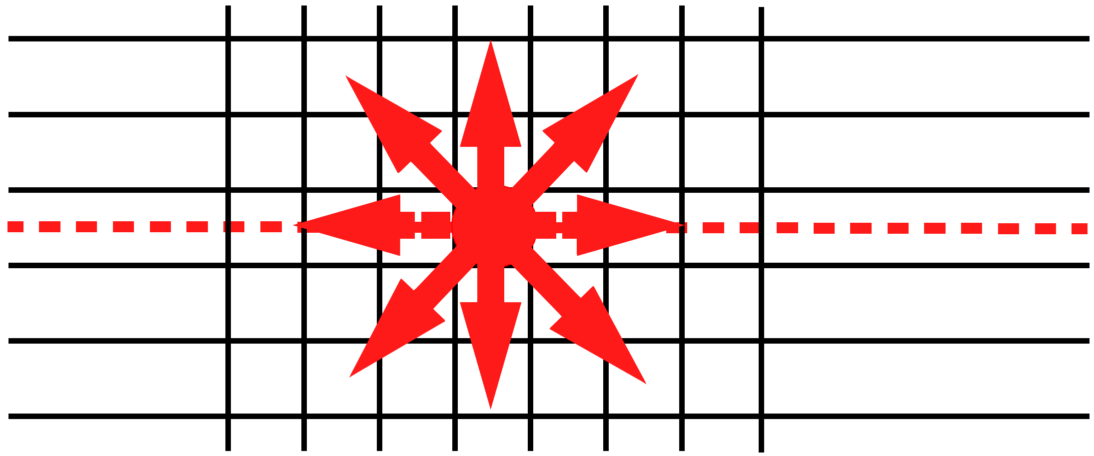
EX4: DIFFUSION - 2D

❑ SCHEME

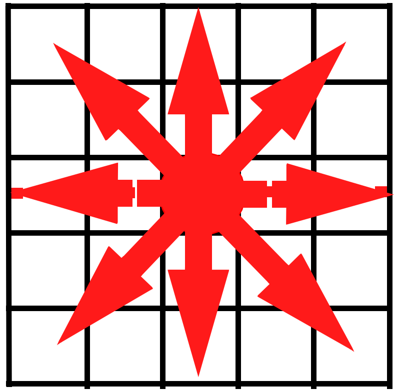


EX4: DIFFUSION - 2D

- SCHEME



SCHEME



definitions

```
const D1 = 5;                         // grid size first dimension
const D2 = 5;                         // grid size second dimension
const MIDDLE = D1/2;

colorset CD1 = 1-D1;                  // row index
colorset CD2 = 1-D2;                  // column index
colorset Grid2D = CD1 x CD2;          // 2D grid

var x, a : CD1;
var y, b : CD2;
```

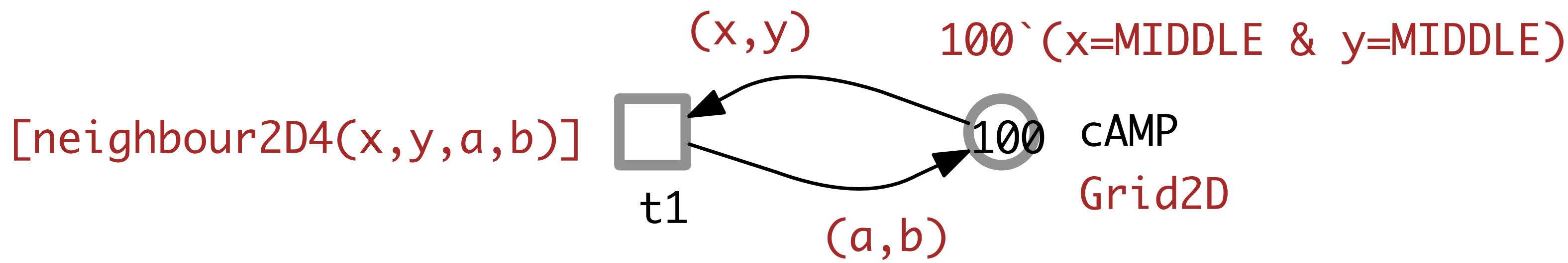
four neighbours

function *neighbour2D4 (CD1 x, CD2 y, CD1 a, CD2 b)* ***bool****:*

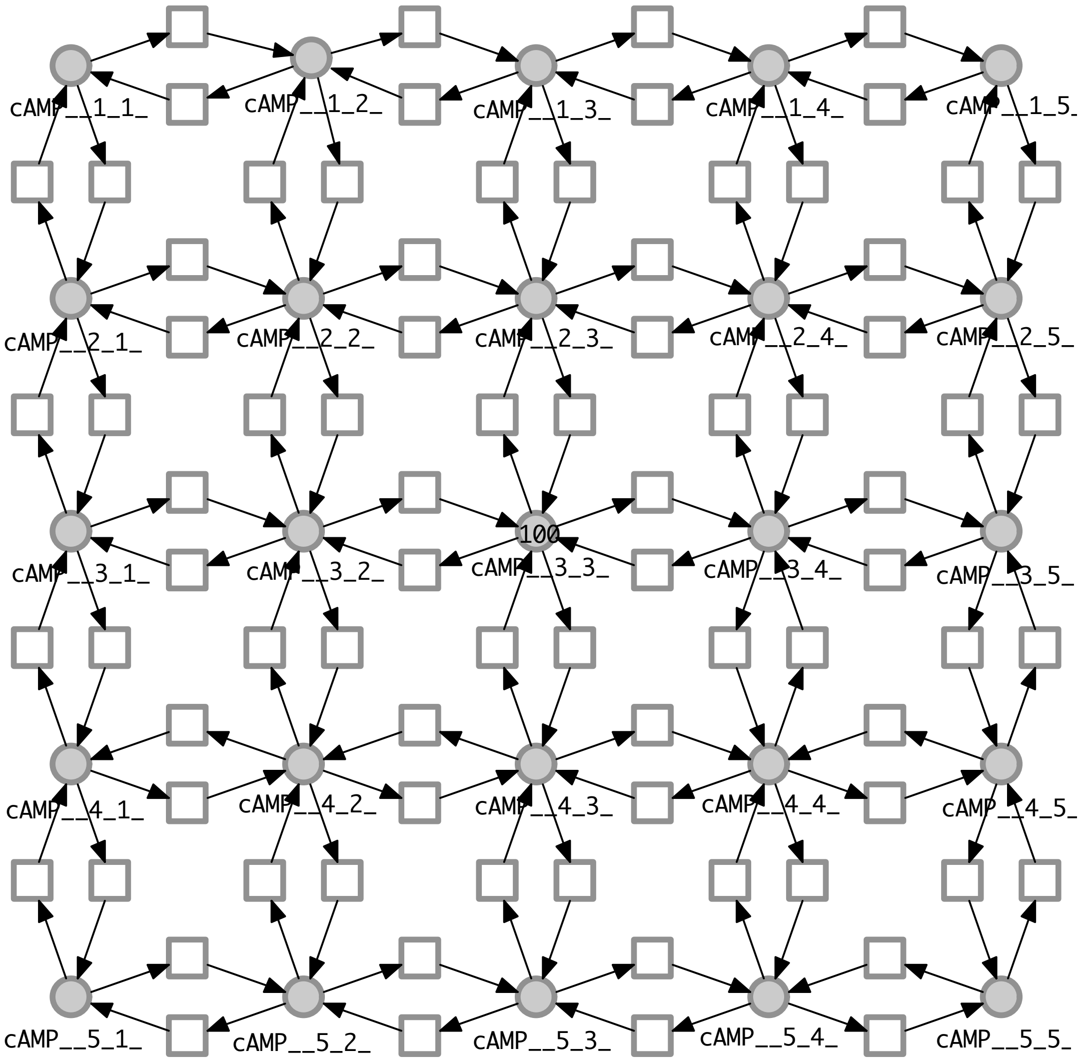
// (a,b) is one of the up to four neighbours of (x,y)

(a=x & b=y-1) | (a=x & b=y+1)

| (b=y & a=x-1) | (b=y & a=x+1);



EX4: DIFFUSION - 2D4 NEIGHBOURHOOD



EX4: DIFFUSION - 2D8 NEIGHBOURHOOD

- **eight neighbours**

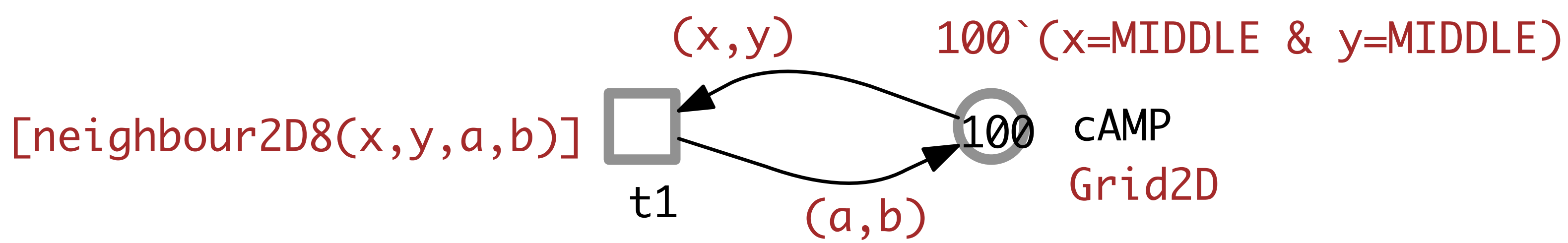
  function *neighbour2D8 (CD1 x, CD2 y, CD1 a, CD2 b)* ***bool****:*

  // (a,b) is one of the up to eight neighbours of (x,y)

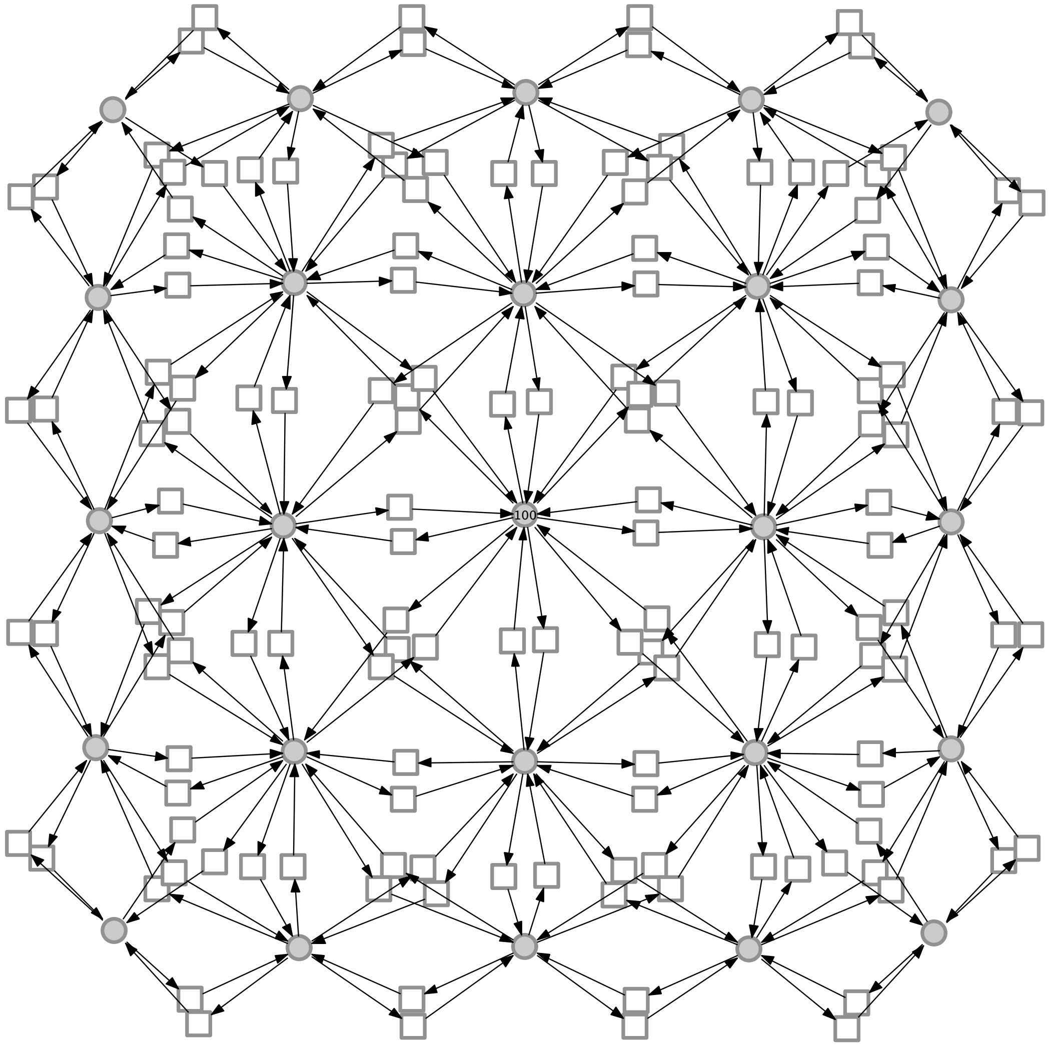
  (a=x-1 | a=x | a=x+1) & (b = y-1 | b=y | b=y+1)

  & (!(a=x & b=y))

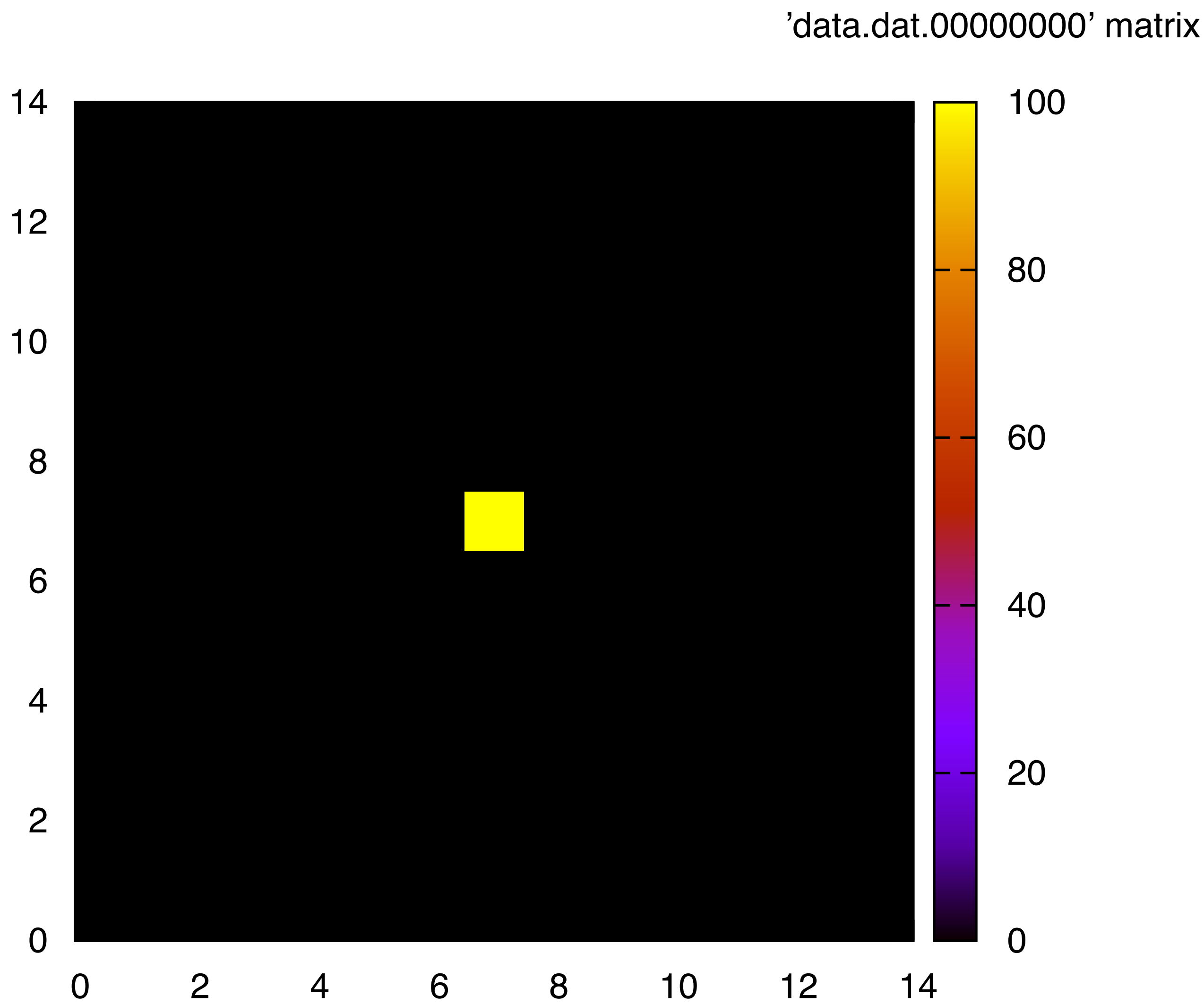
  & (1<=a & a<=D1) & (1<=b & b<=D2);



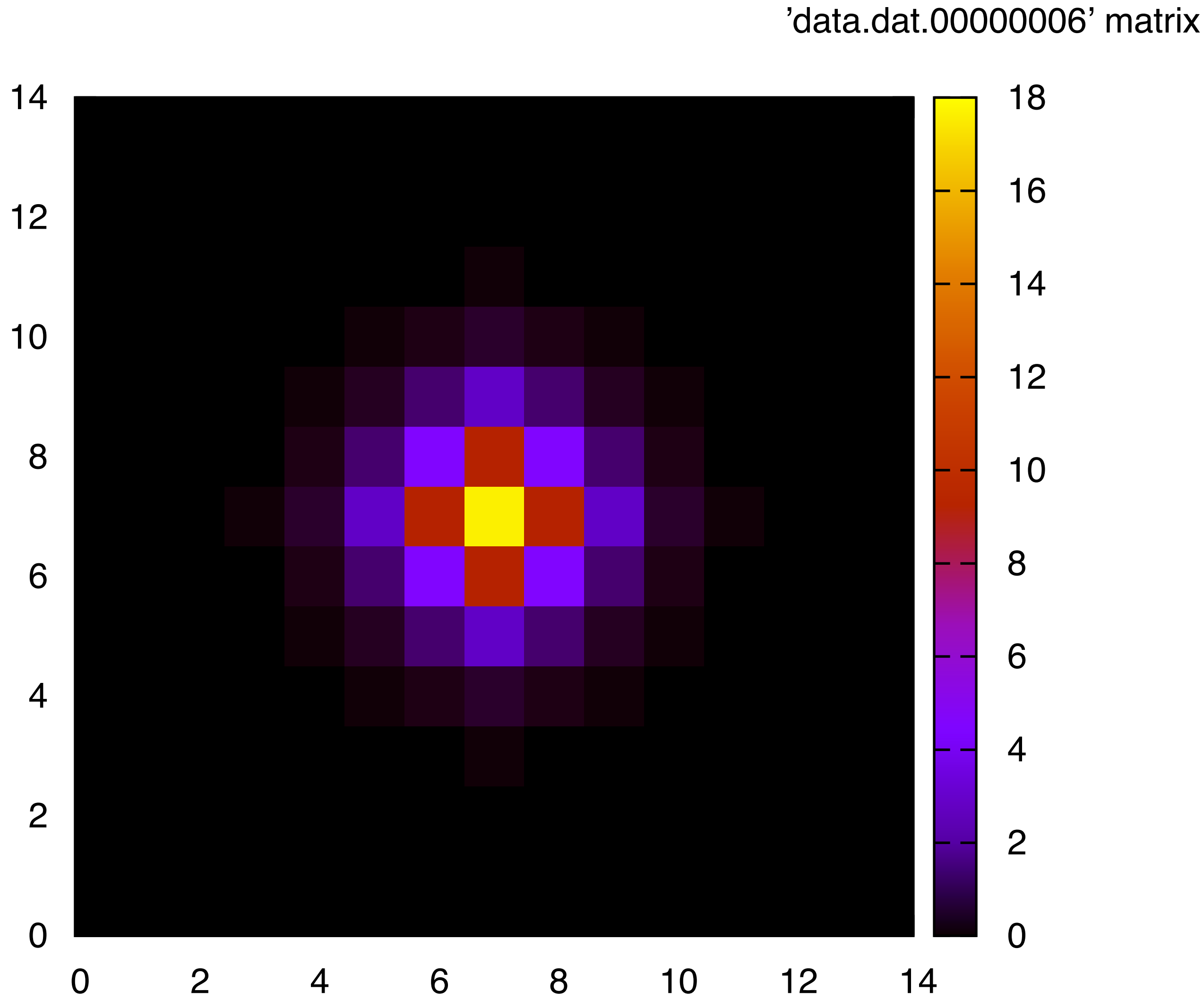
EX4: DIFFUSION - 2D8 NEIGHBOURHOOD



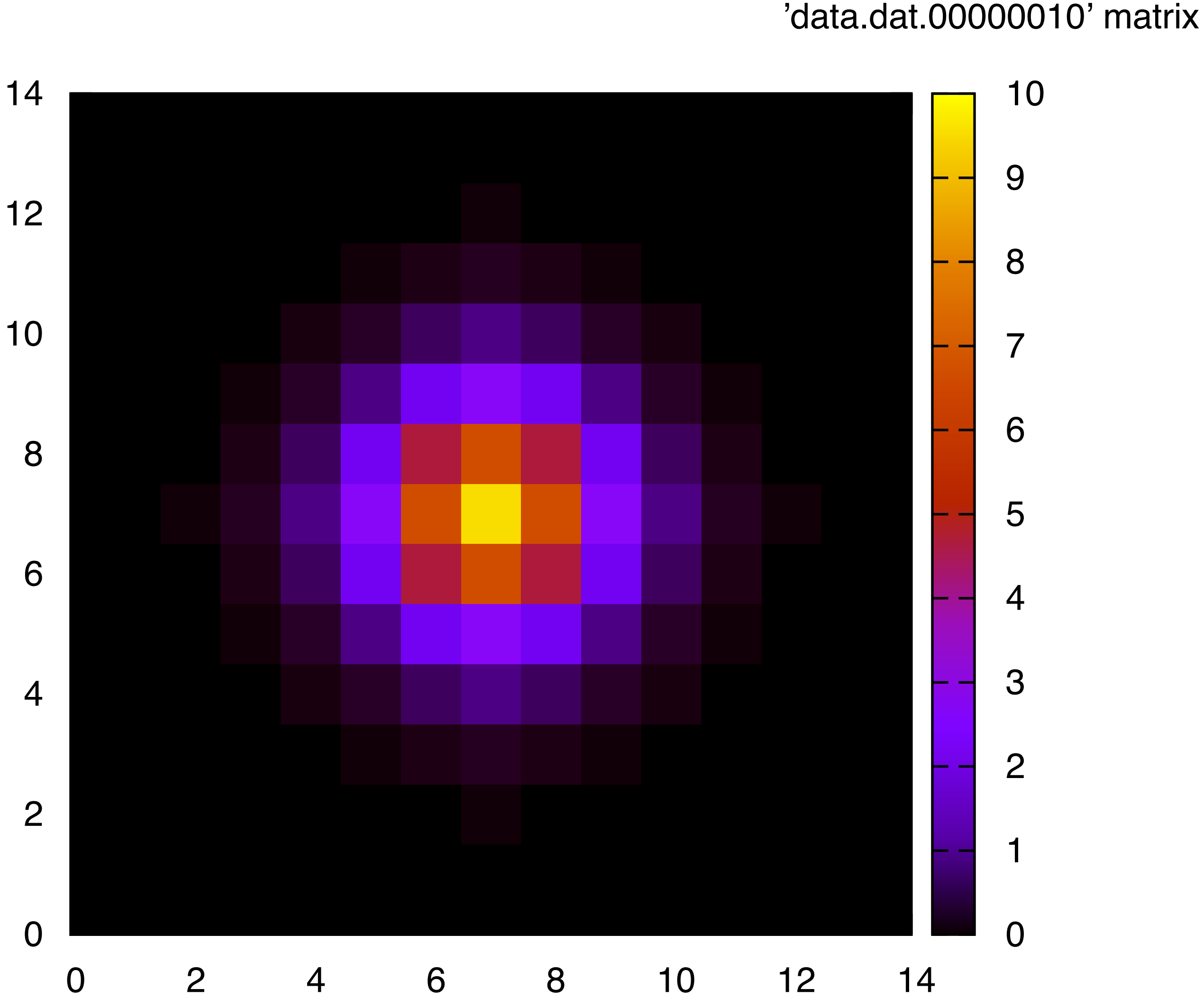
EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 15X15



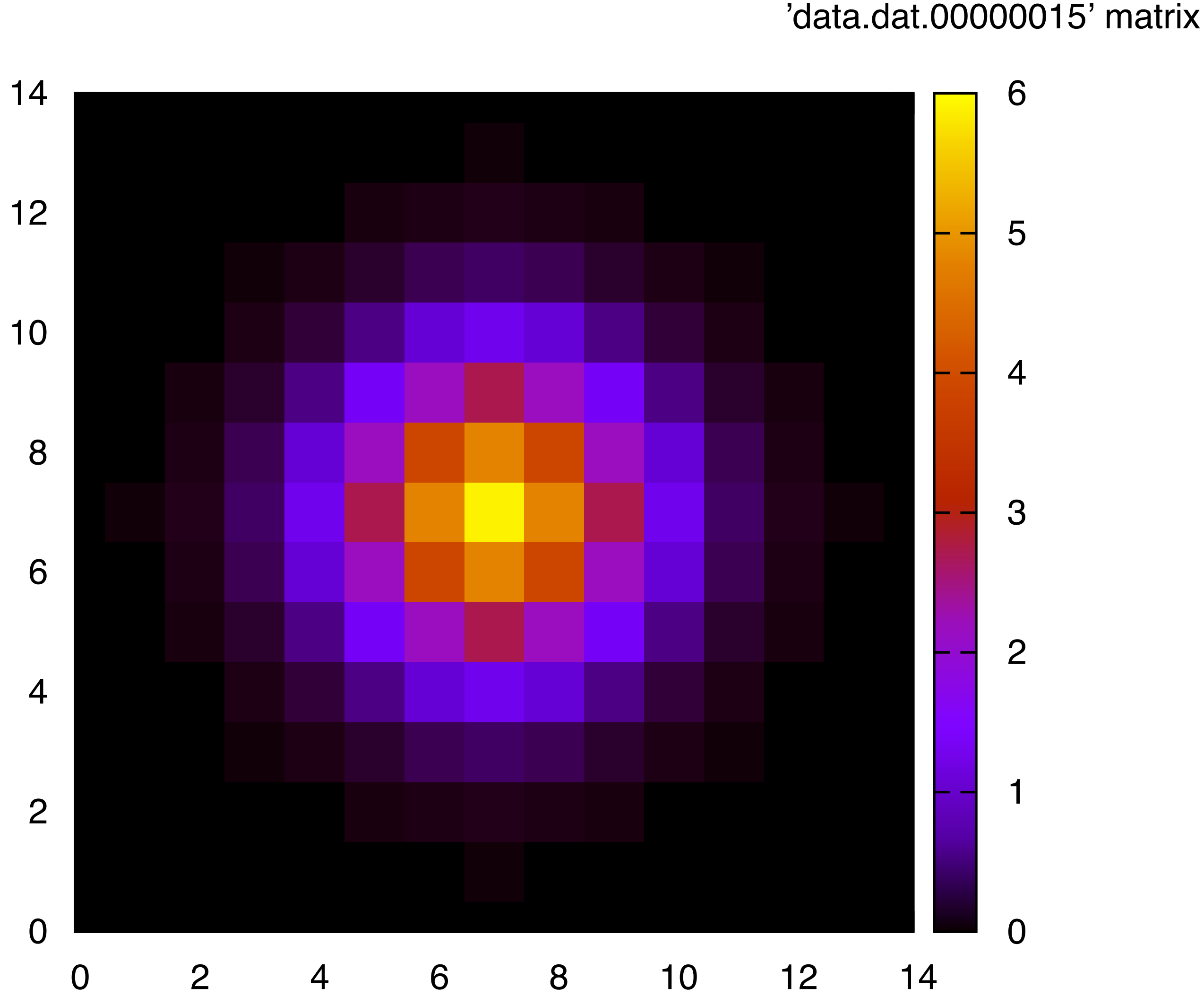
Ex4: Diffusion - 2D4 Neighbourhood, 15x15



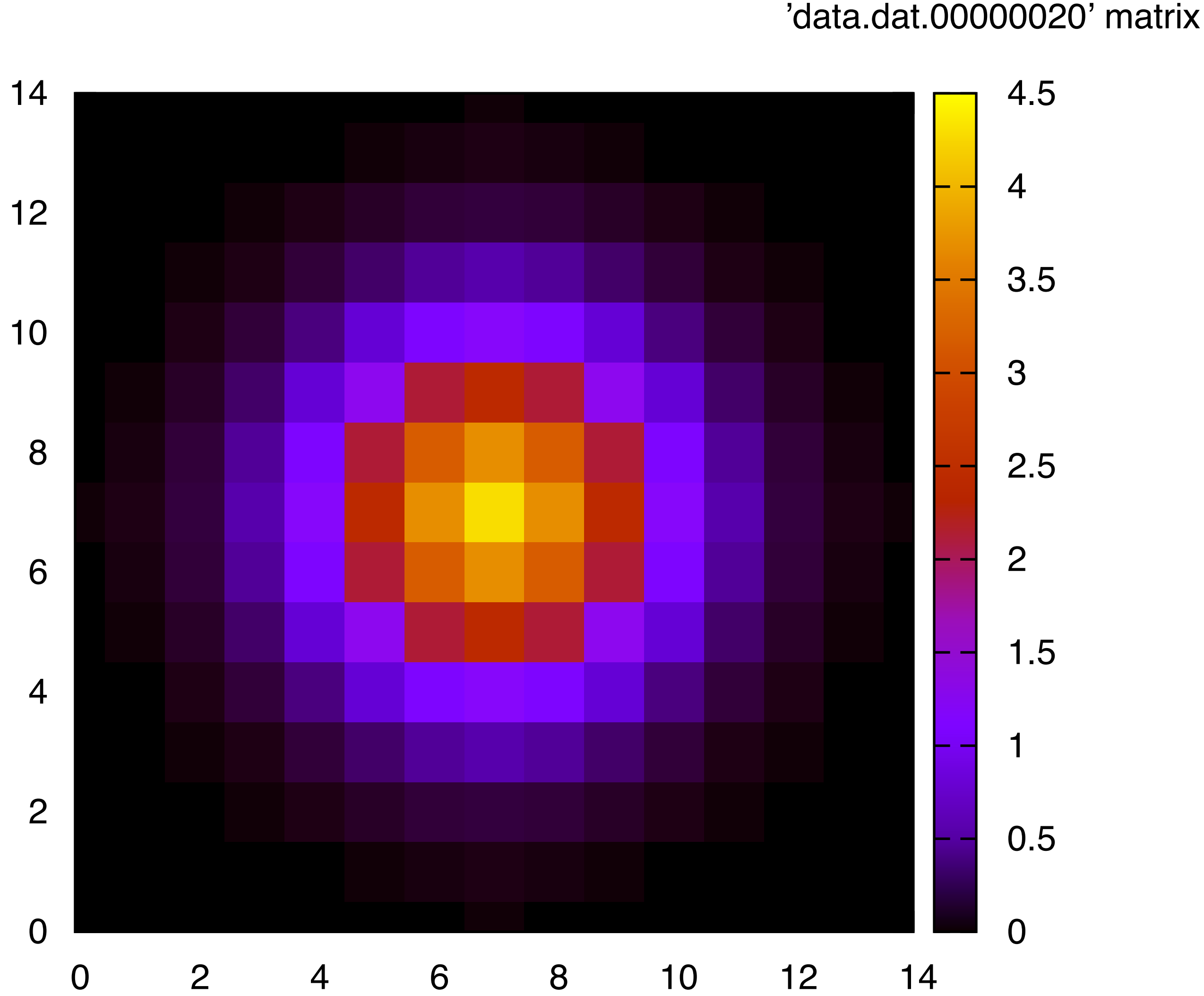
EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 15X15



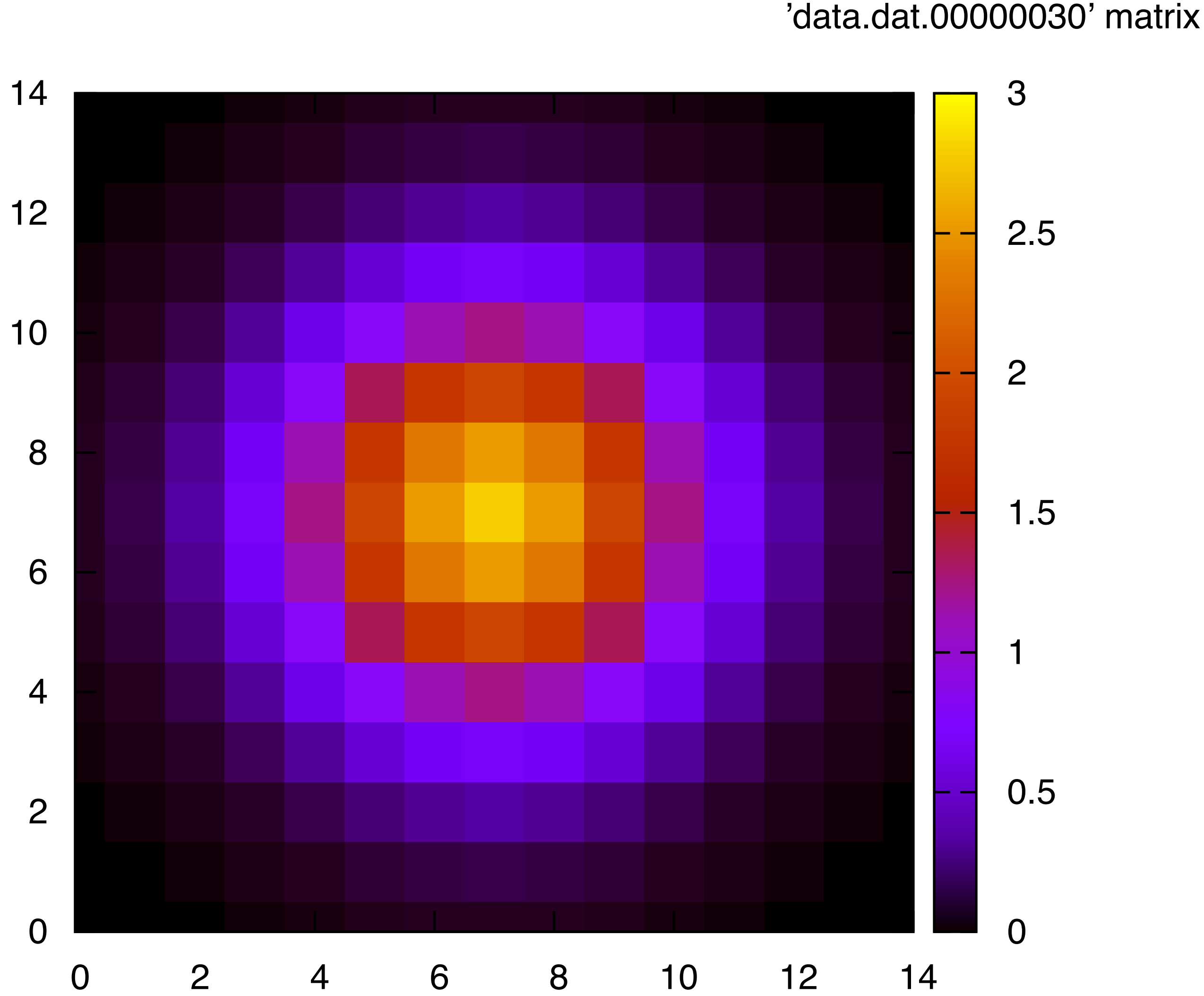
Ex4: Diffusion - 2D4 Neighbourhood, 15x15



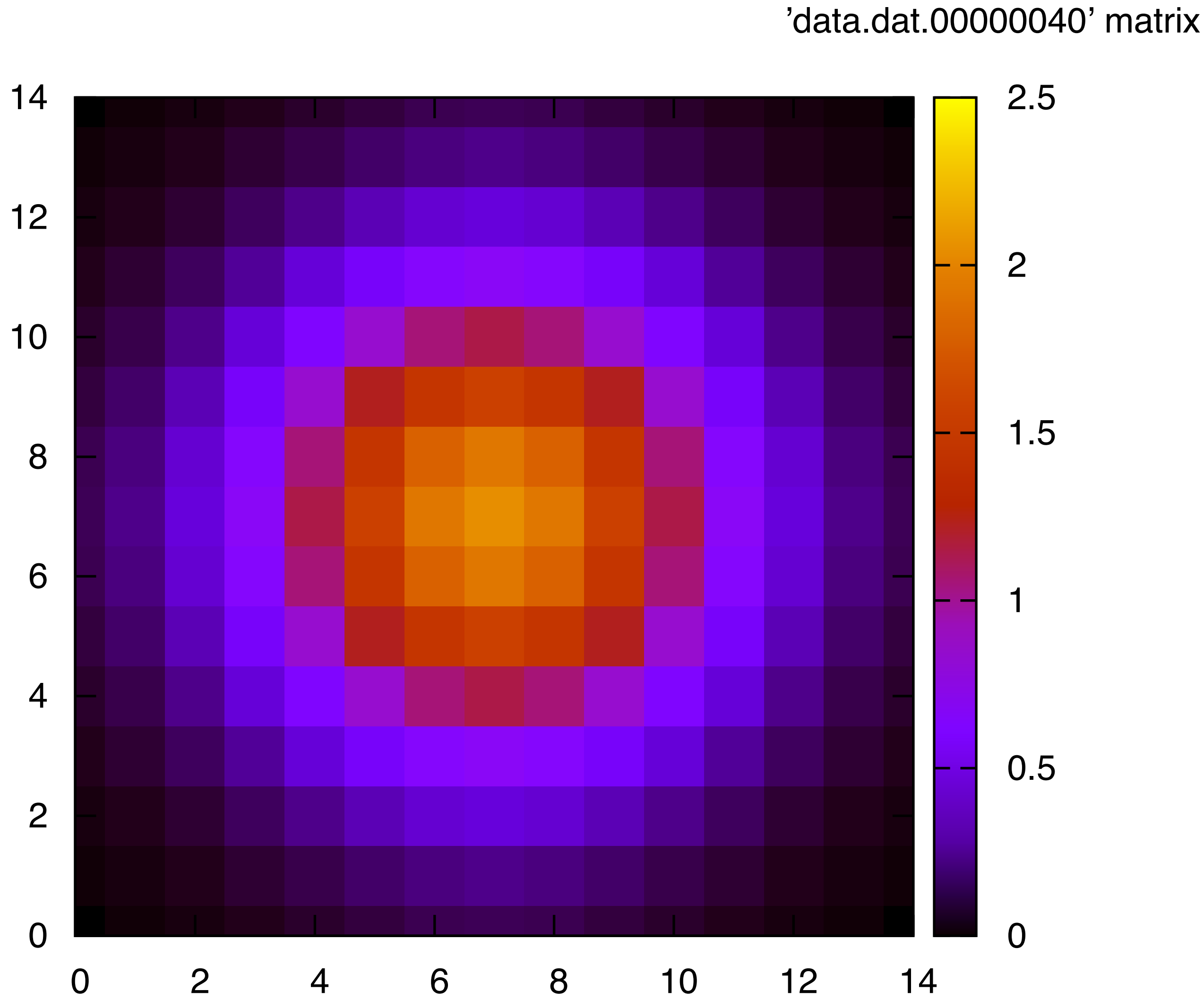
EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 15X15



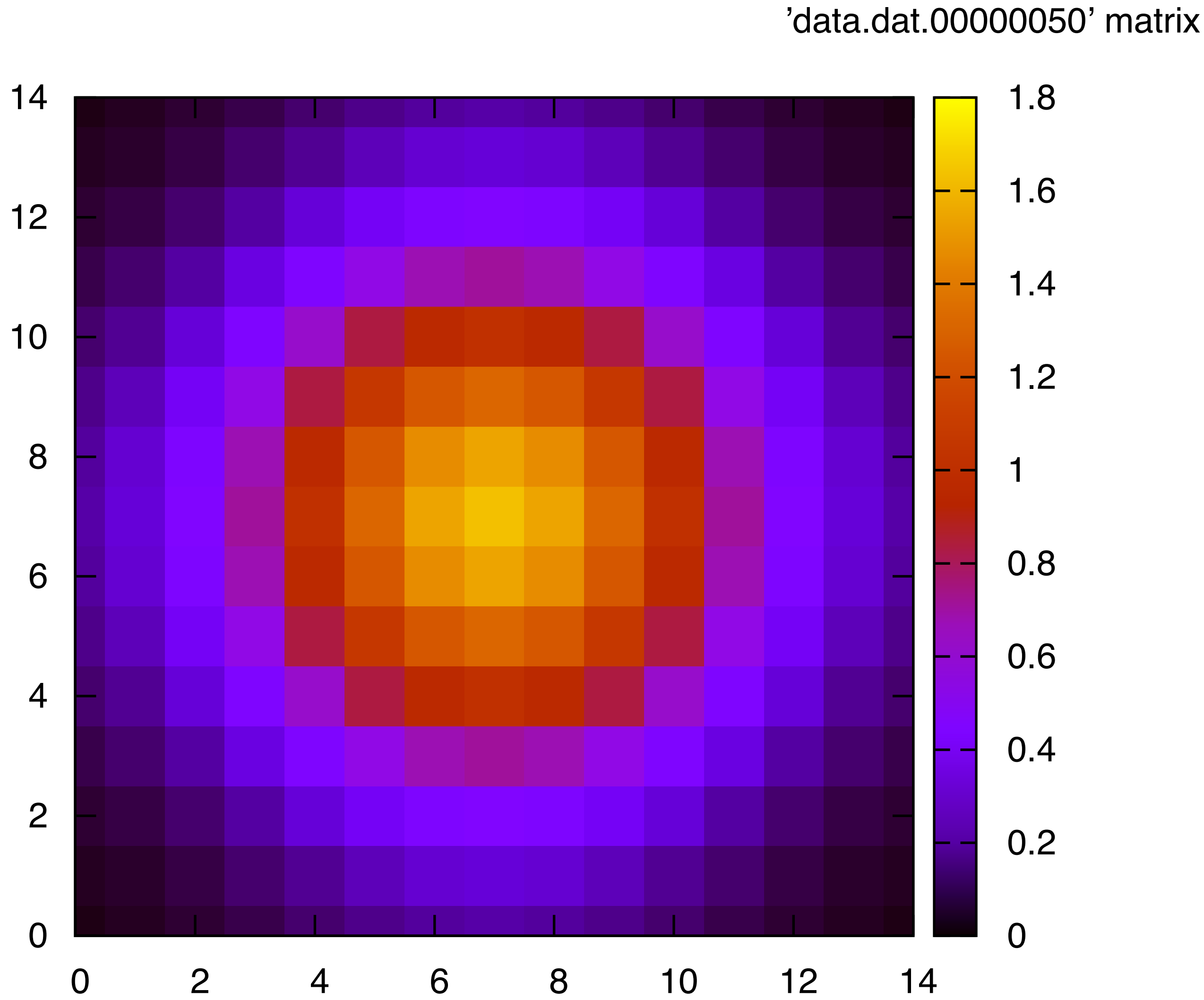
Ex4: Diffusion - 2D4 Neighbourhood, 15x15



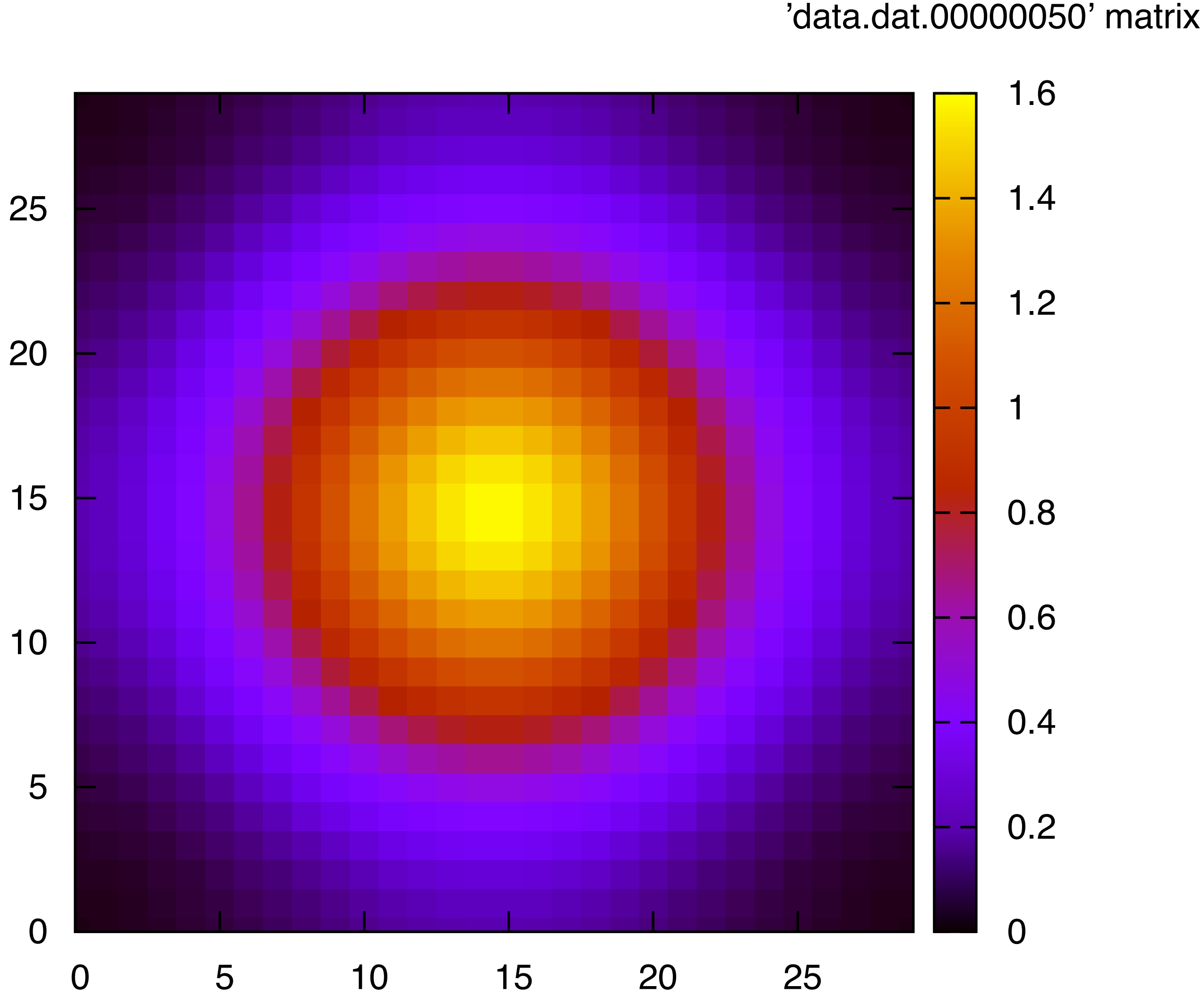
EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 15X15



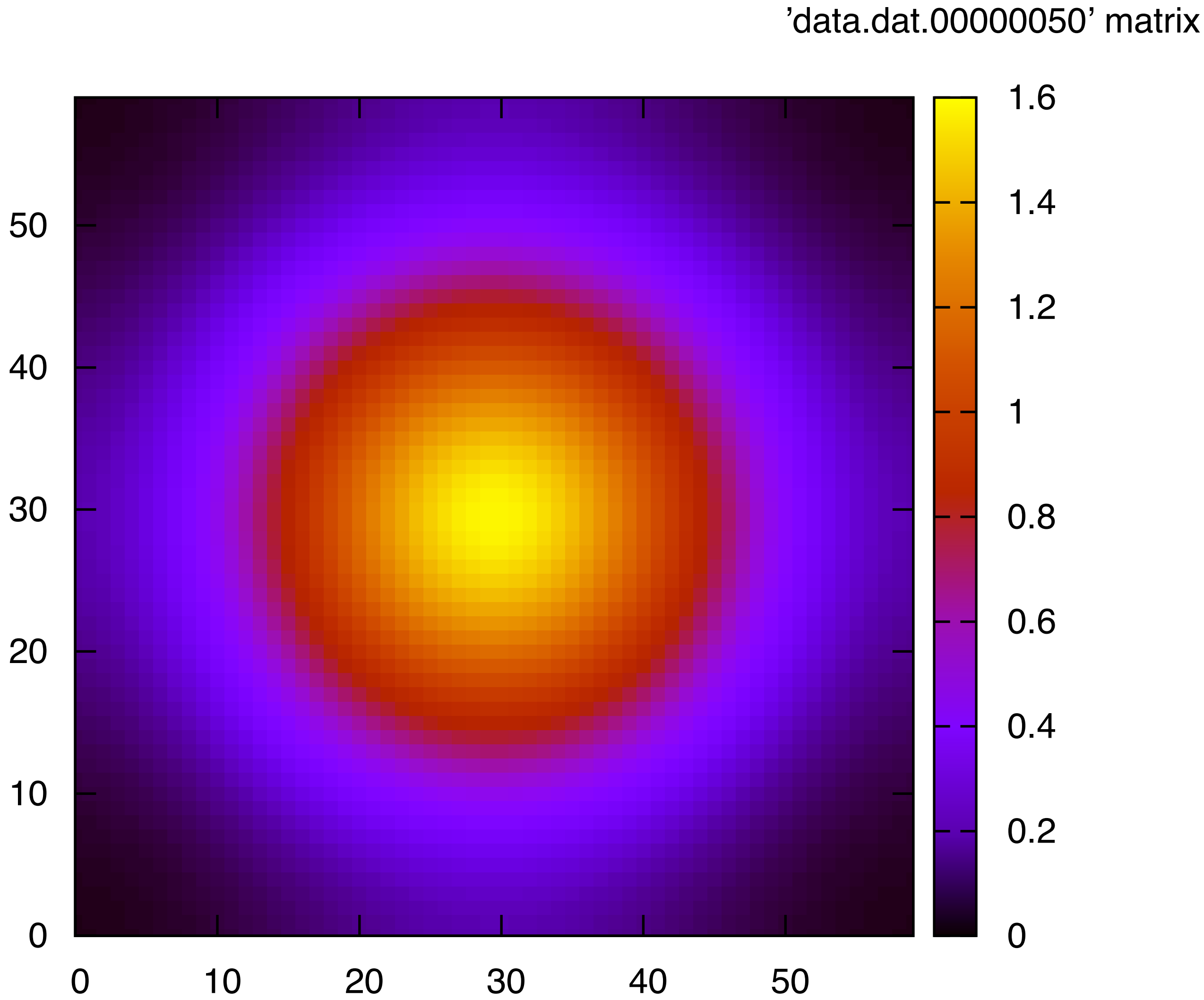
EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 15X15



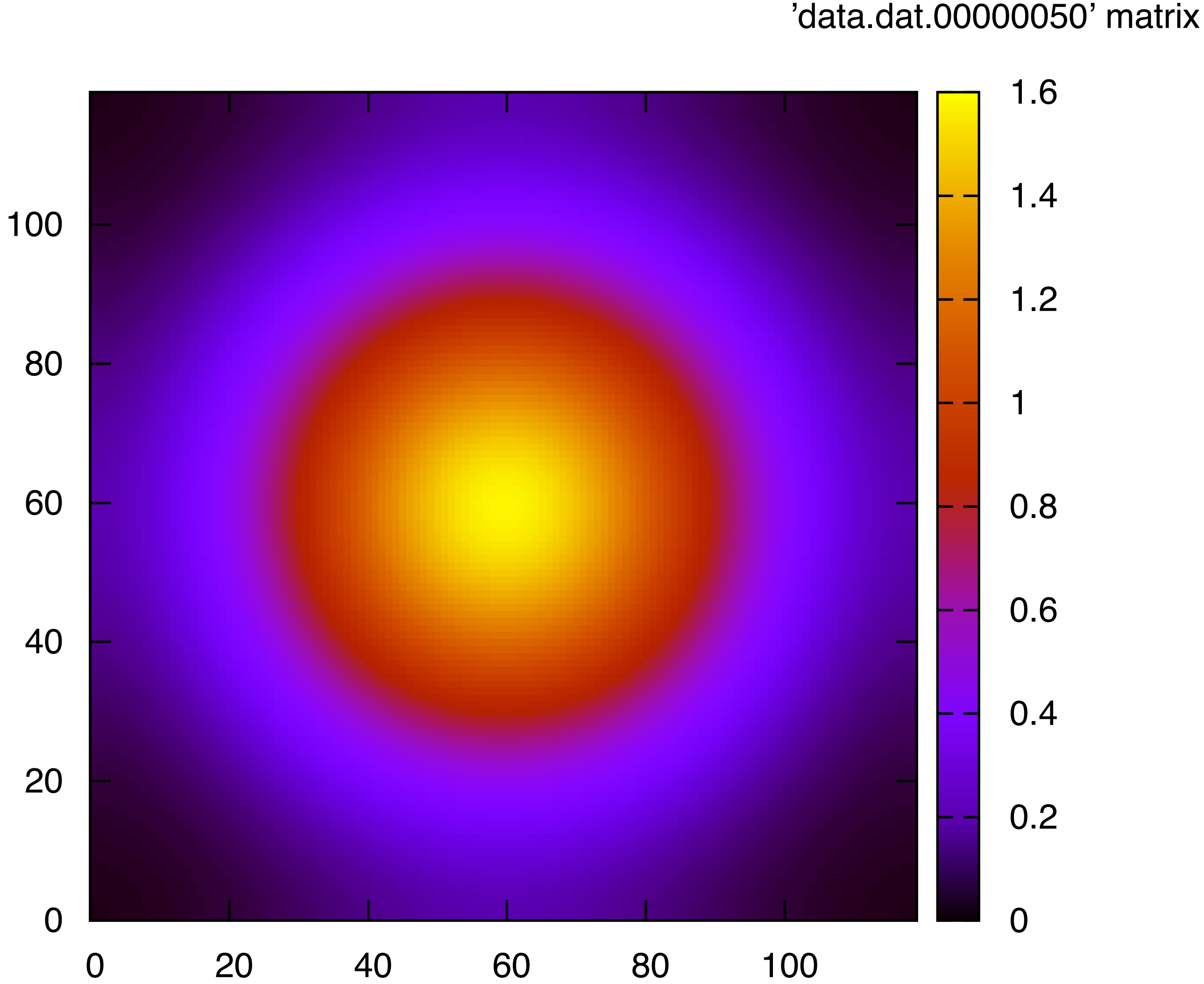
EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 30X30



EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 60X60



EX4: DIFFUSION - 2D4 NEIGHBOURHOOD, 120X120



EXAMPLE:

PHASE VARIATION IN

MULTISTRAIN CELL COLONIES

- **method for dealing with rapidly varying environments without requiring random mutations**

- **contingency gene**

  -> *populations include variants adapted to "foreseeable" frequently encountered environmental or selective conditons*

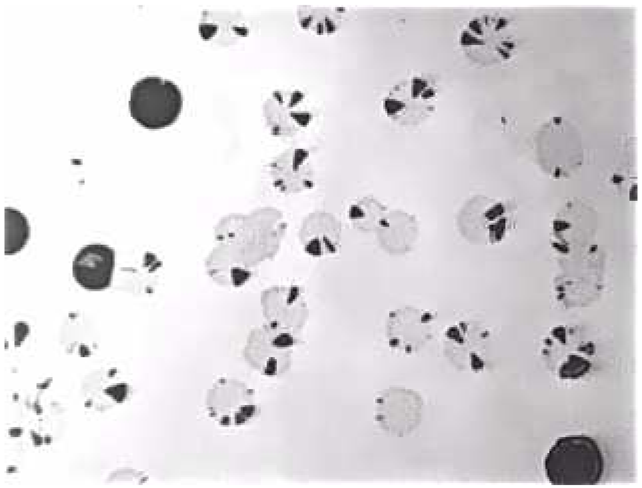
- **stochastic gene switching process**

  -> *controlled by reversible gene mutations, inversions, or epigenetic modification*

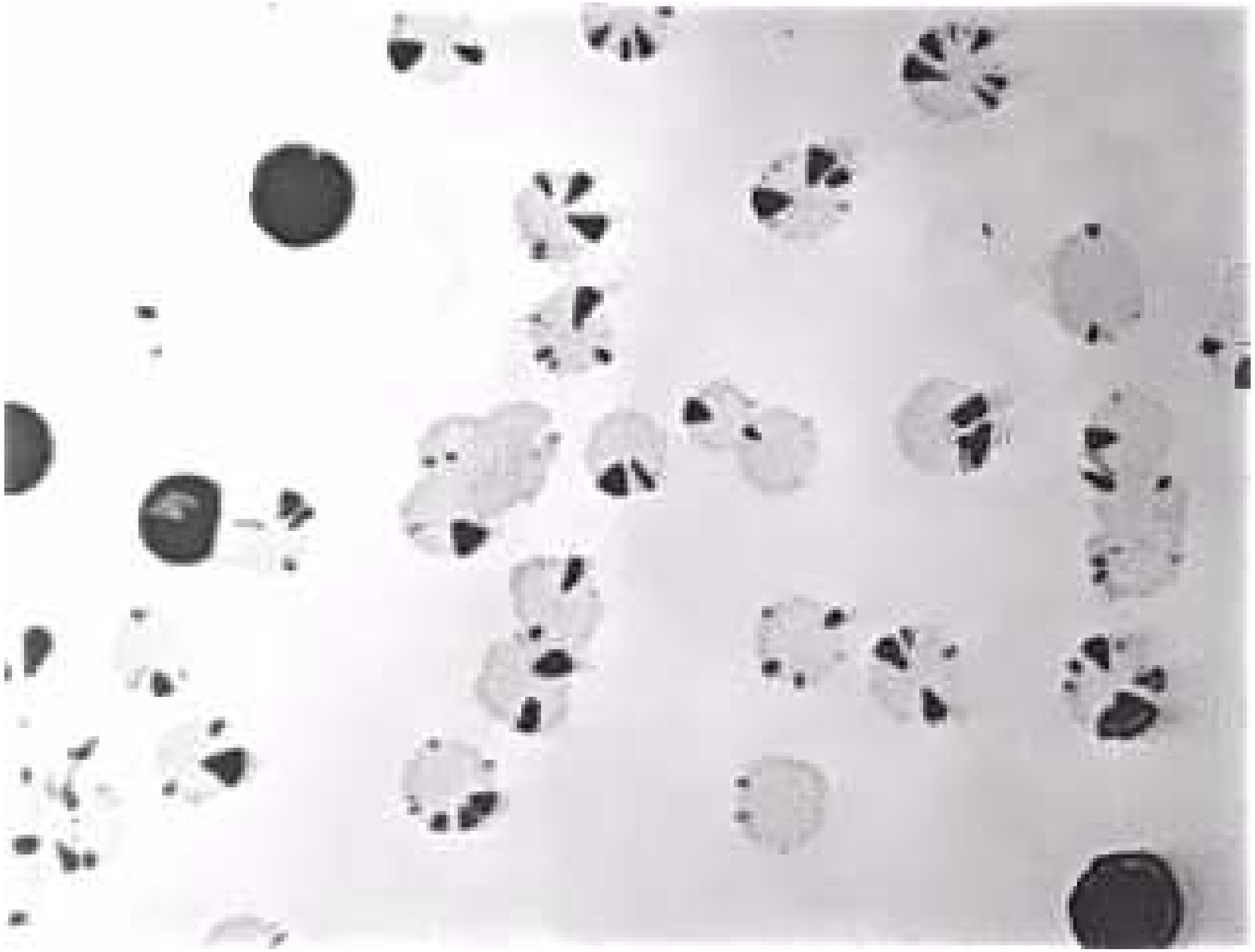
  -> *e.g. switch between two phenotypes A, B*

- **colonial sectoring**

  -> *observable effect in cultures grown in vitro*



EX5: CELL COLONIES, WETLAB OBSERVATIONS



(courtesy of N Saunders)

Microbiology (2003), **149**, 485–495 DOI 10.1099/mic.0.25807-0

Mutation rates: estimating phase variation rates when fitness differences are present and their impact on population structure

Nigel J. Saunders,[1]† E. Richard Moxon[1] and Mike B. Gravenor[2]

[1]Molecular Infectious Diseases Group, Institute of Molecular Medicine, University of Oxford, Headington, Oxford OX3 9DS, UK

[2]Institute for Animal Health, Compton, Berkshire RG20 7NN, UK

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Nigel J. Saunders
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Phase variation is a mechanism of ON–OFF switching that is widely utilized by bacterial pathogens. There is currently no standardization to how the rate of phase variation is determined experimentally.

Microbiology (2003), **149**, 485–495

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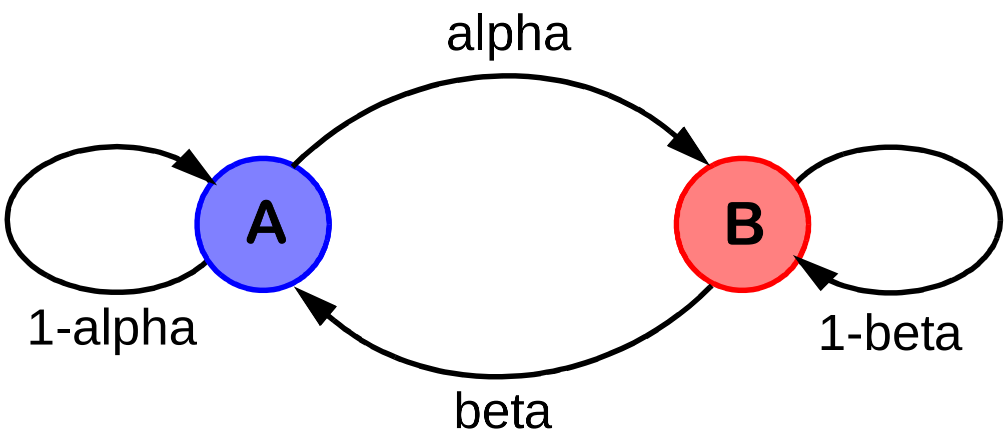
Phase variation is a mechanism of ON–OFF switching that is widely utilized by bacterial pathogens. There is currently no standardization to how the rate of phase variation is determined experimentally.

NO SPACE

- **two cell types: phenotype A and B**

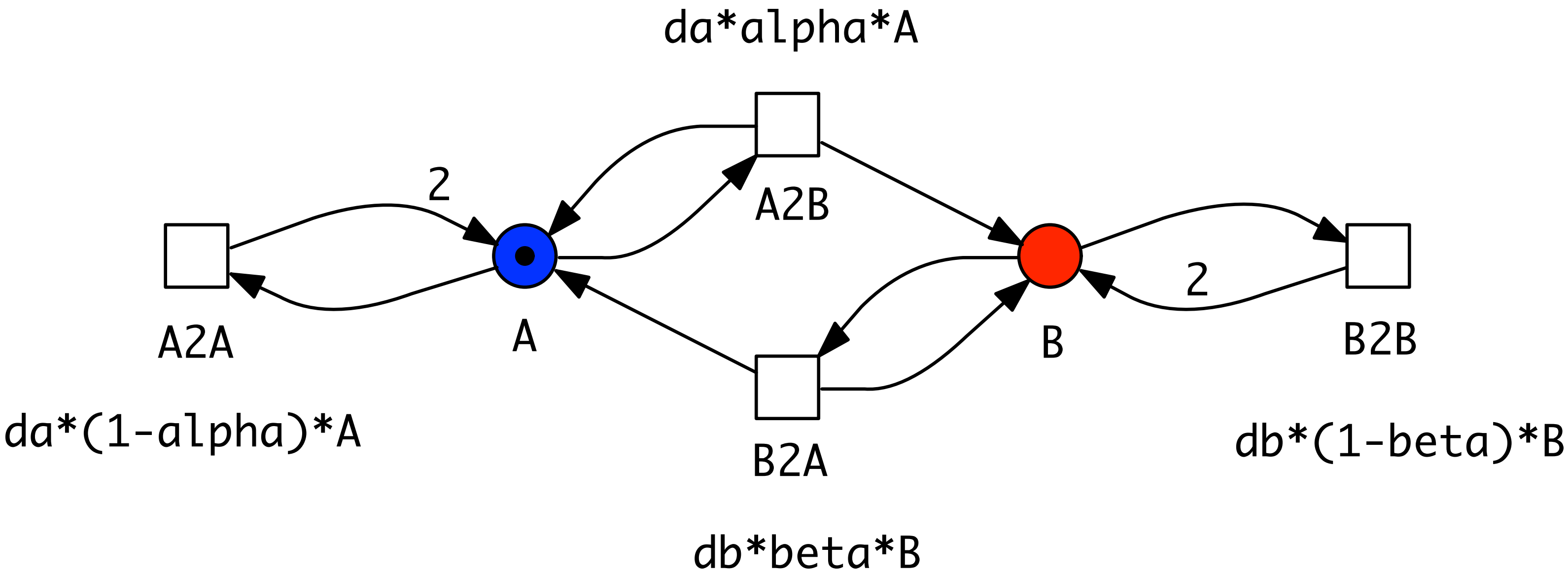
- **cell divide**
  - -> *cell division may involve mutation of the offspring*
  - -> *parent cell keeps its phenotype*



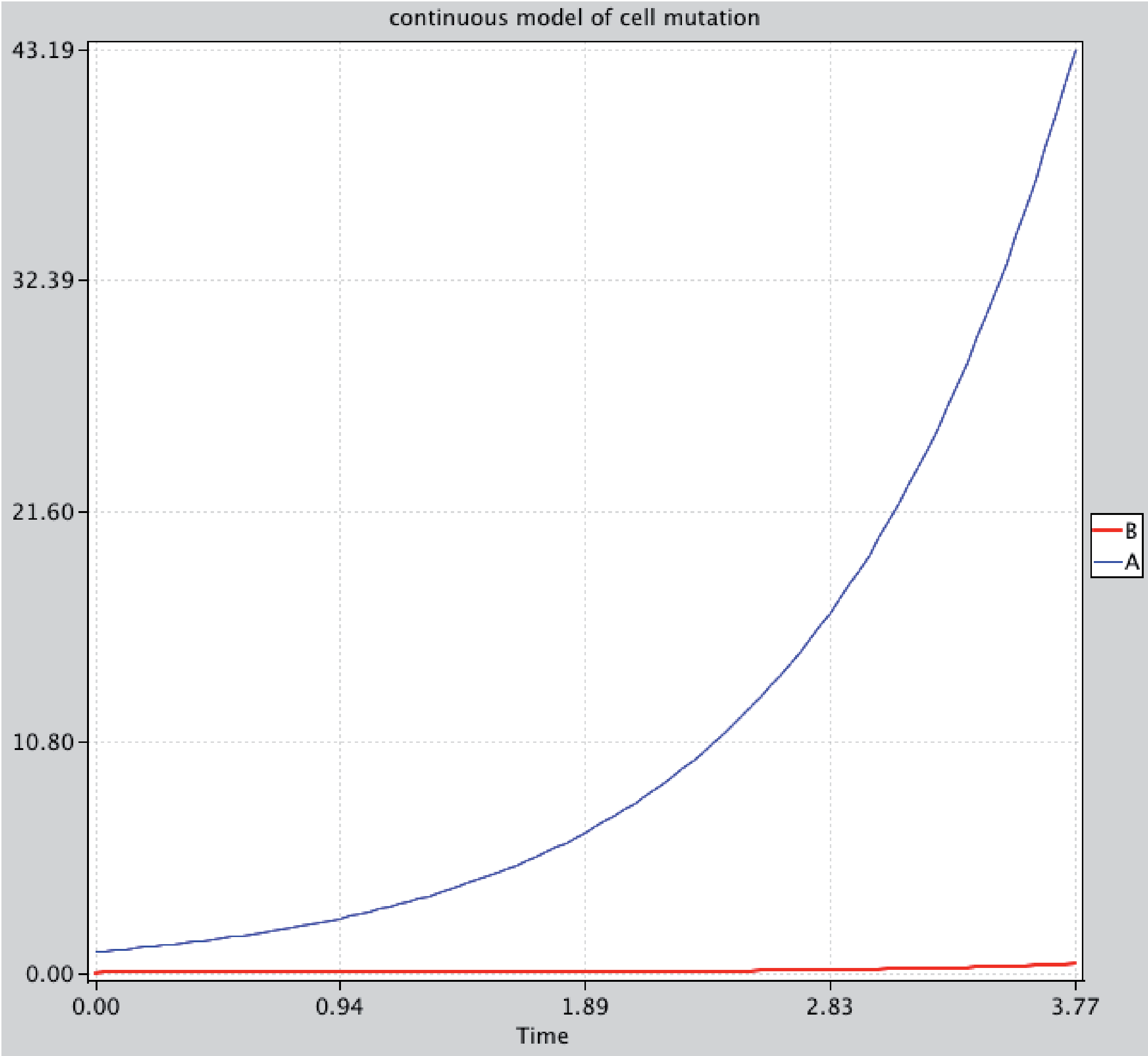
- **model parameters**
  - -> *alpha = beta - mutation rates*
  - -> *da, db - fitness of A, B*
  - -> *da/db - relative fitness*

- **output**
  - -> *total number of cells*
  - -> *proportion of A = A / (A + B)*
  - -> *proportion of B = B / (A + B)*

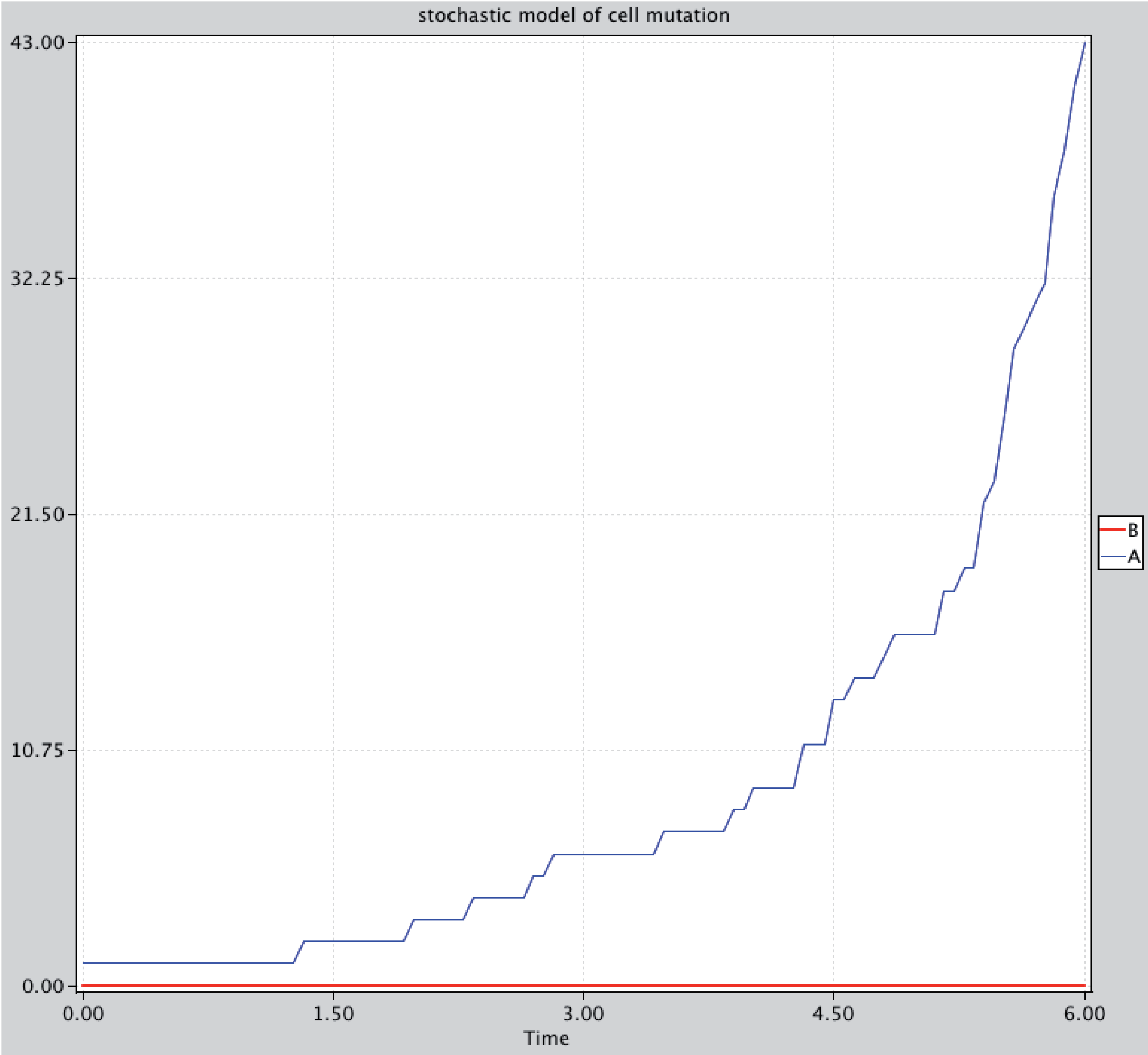
EX5: CELL COLONIES, PETRI NET



EX5: CELL COLONIES, CONTINUOUS PLOT

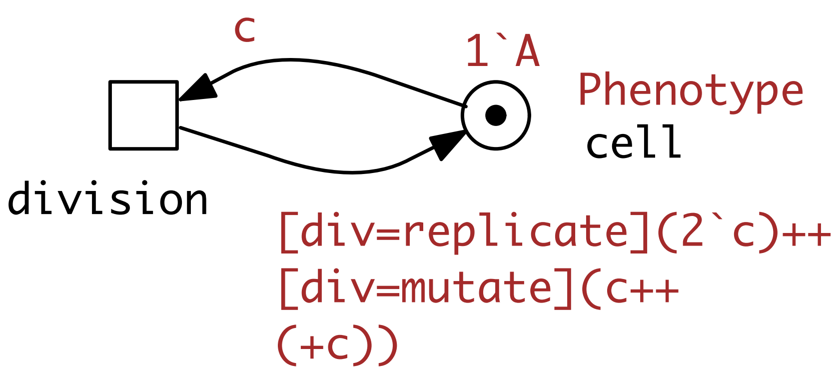


EX5: CELL COLONIES, STOCHASTIC PLOT



EX5: CELL COLONIES, BASIC MODEL

colorset Phenotype = **enum with** A, B;
colorset DivisionType = **enum with** replicate , mutate ;




```
(c=A) & (div=replicate) : cell*da*(1-alpha)
(c=A) & (div=mutate) : cell*(da*alpha)
(c=B) & (div=replicate) : cell*(db*(1-beta))
(c=B) & (div=mutate) : cell*(db*beta)
```

colorset Phenotype = **enum with** A, B;
colorset DivisionType = **enum with** replicate , mutate ;

ADDING SPACE

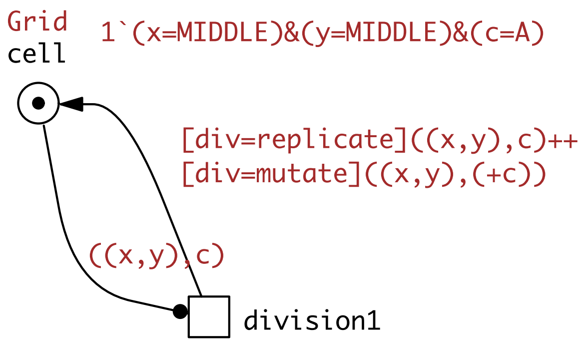
CONTROLLING COLONY SPREADING

CONTROLLING THICKNESS

(c=A) & (div=replicate) : cell*(da*(1-alpha))
(c=A) & (div=mutate) : cell*(da*alpha)
(c=B) & (div=replicate) : cell*(db*(1-beta))
(c=B) & (div=mutate) : cell*(db*beta)

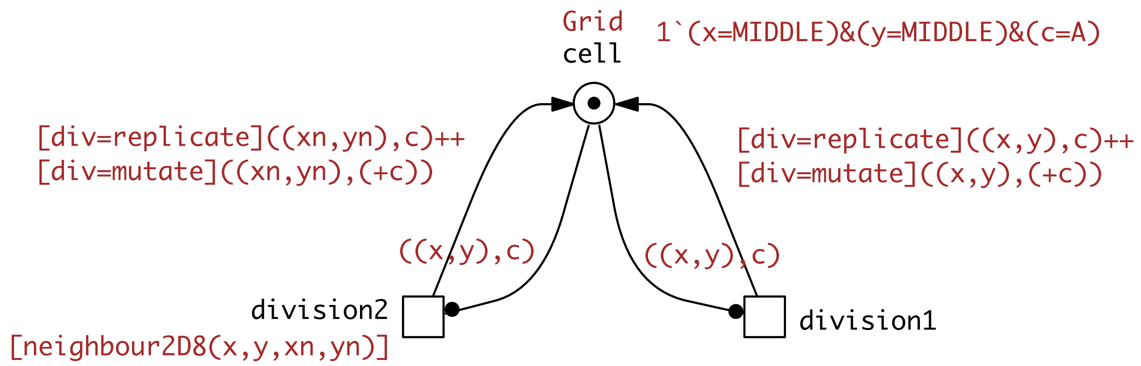
CONTROLLING COLONY SIZE

colorset Grid = **product with** Grid2D x Phenotype;

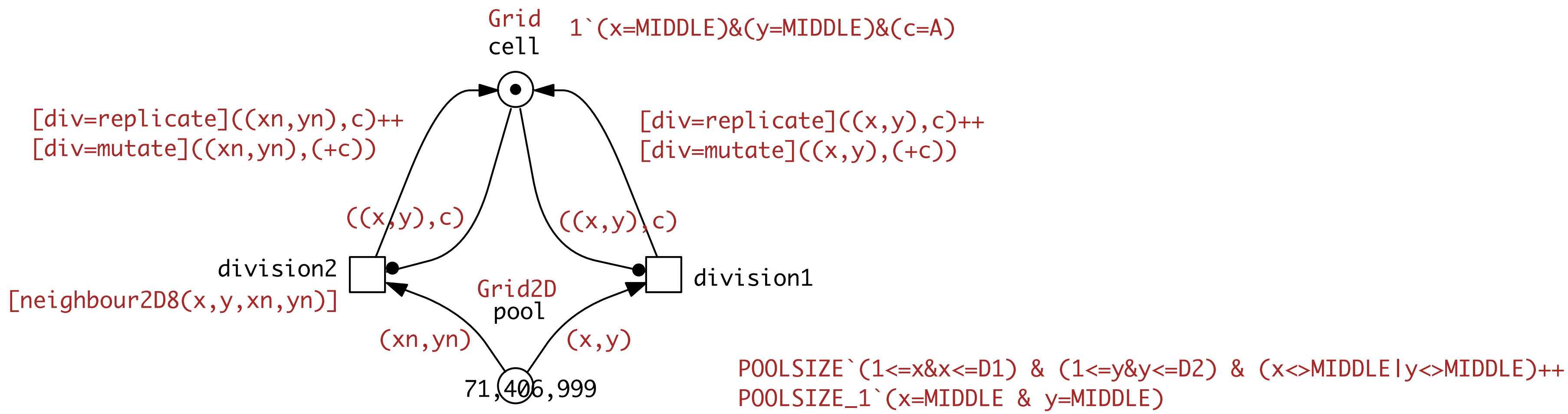


EX5: CELL COLONIES, CONTROLLING COLONY SPREADING

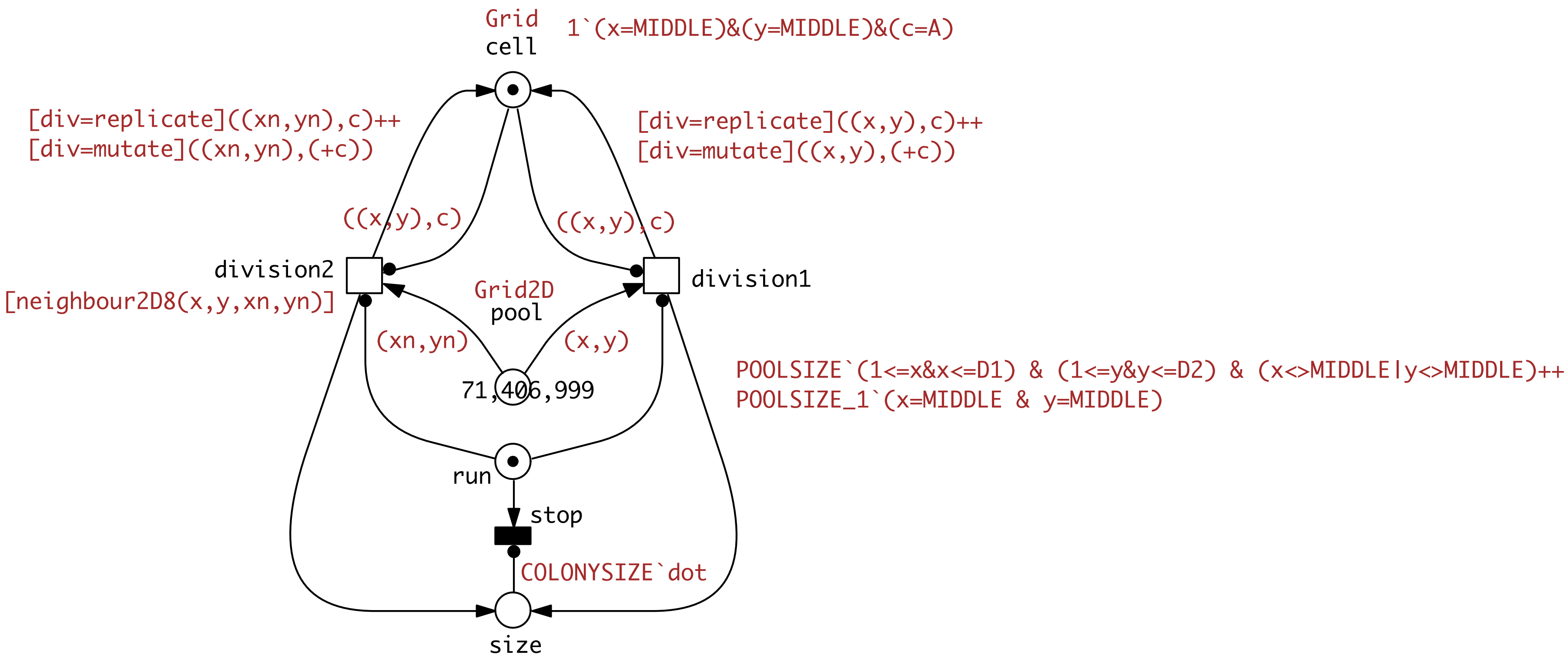
colorset Grid = **product with** Grid2D x Phenotype;



colorset Grid = **product with** Grid2D x Phenotype;



colorset Grid = **product with** Grid2D x Phenotype;

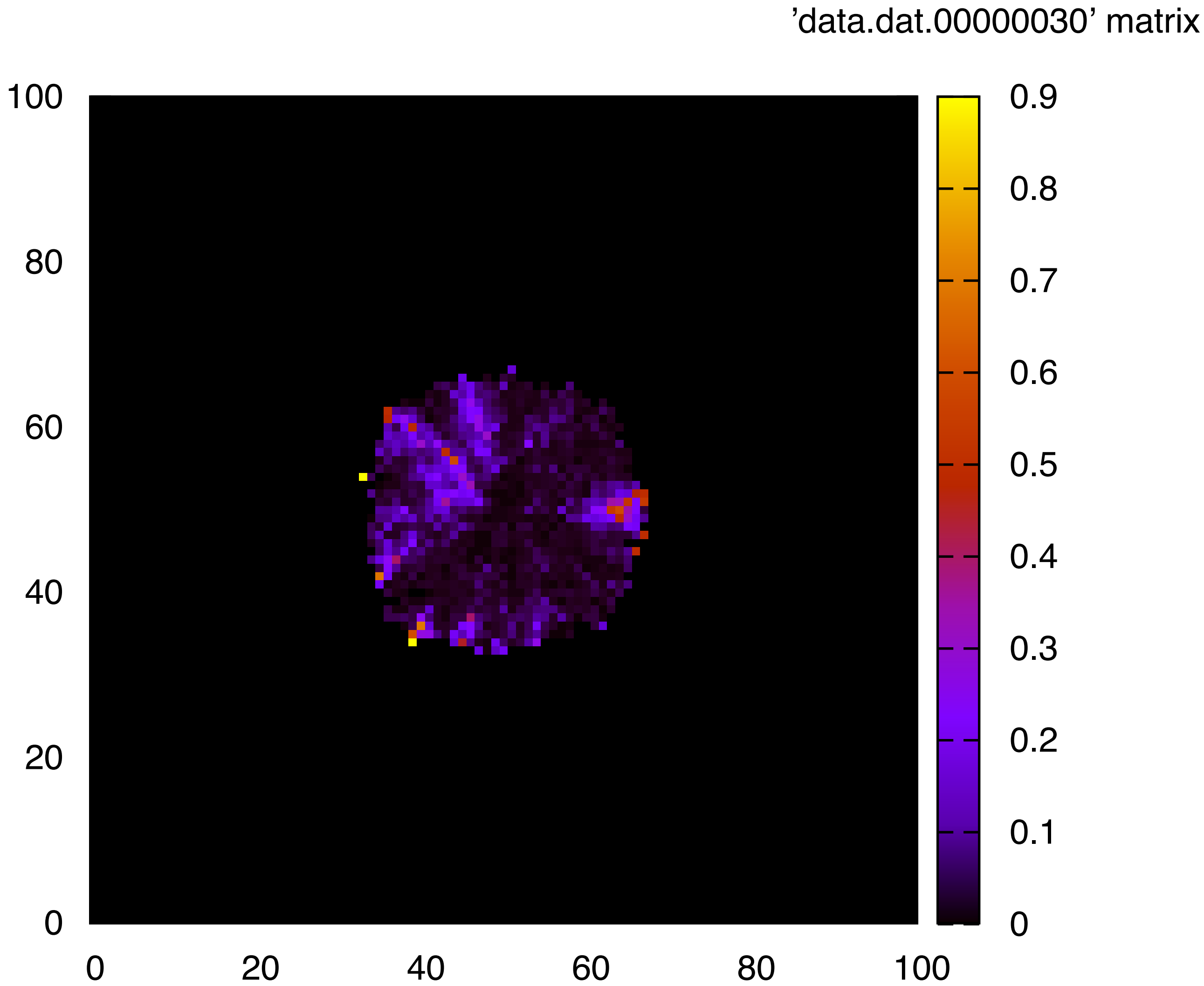


EX5: CELL COLONIES, SOME DETAILS

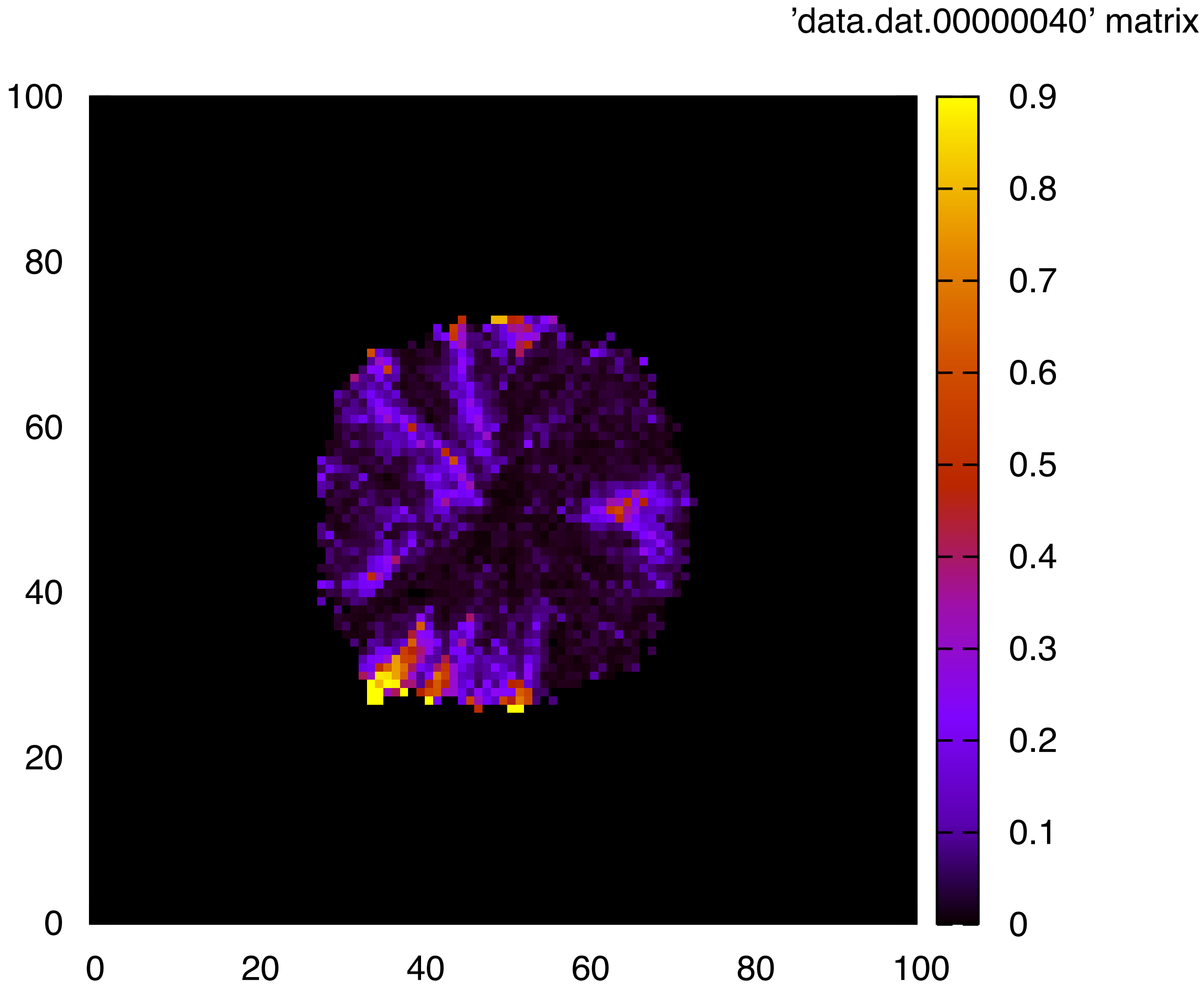
- **model assumptions**
  - -> *"If phase variation occurs, the progeny consists of one A and one B"*
    (Saunders 2003)
  - -> *It is always the mutant who goes to a neighbouring position, if any.*
  - -> *constant biofilm thickness (so far)*
- **colony size - 24 h**
  - -> *25 generations: 33.5 E+06*
  - -> *26 generations: 67 E+06*
  - -> *COLONYSIZE = 70,000,000*
- **grid size**
  - -> *61 x 61 grid: 11,163 P / 131,044 T; unfolding: 152 sec;*
  - -> *101 x 101 grid: 30,603 P / 362,404 T; unfolding: 9 min;*
    - -> *runtime 1 stoch. simulation: 35-40 minutes*

. . . SOME EXPERIMENTS

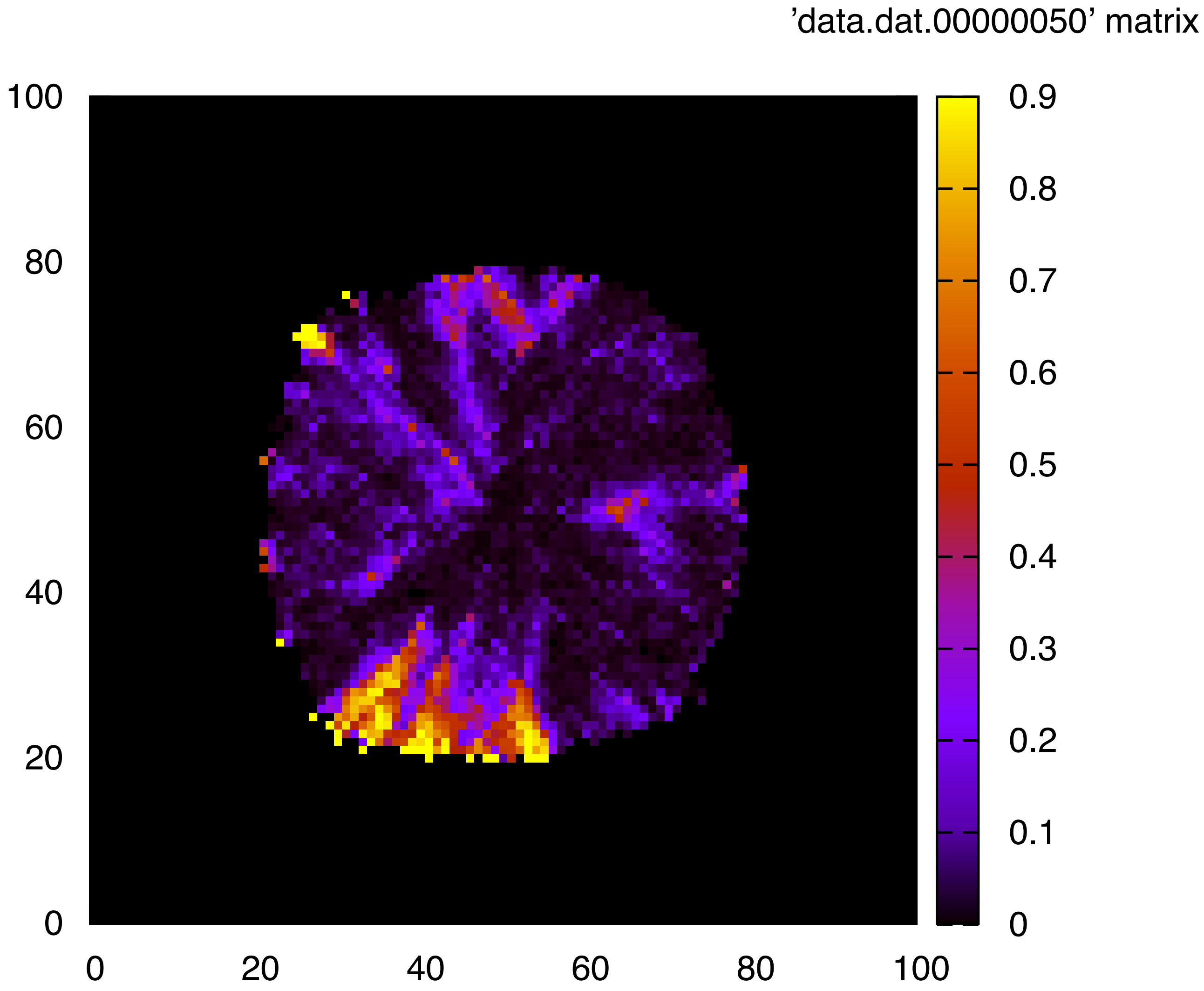
Ex5: 2D - Trace 1 (High, F=1)



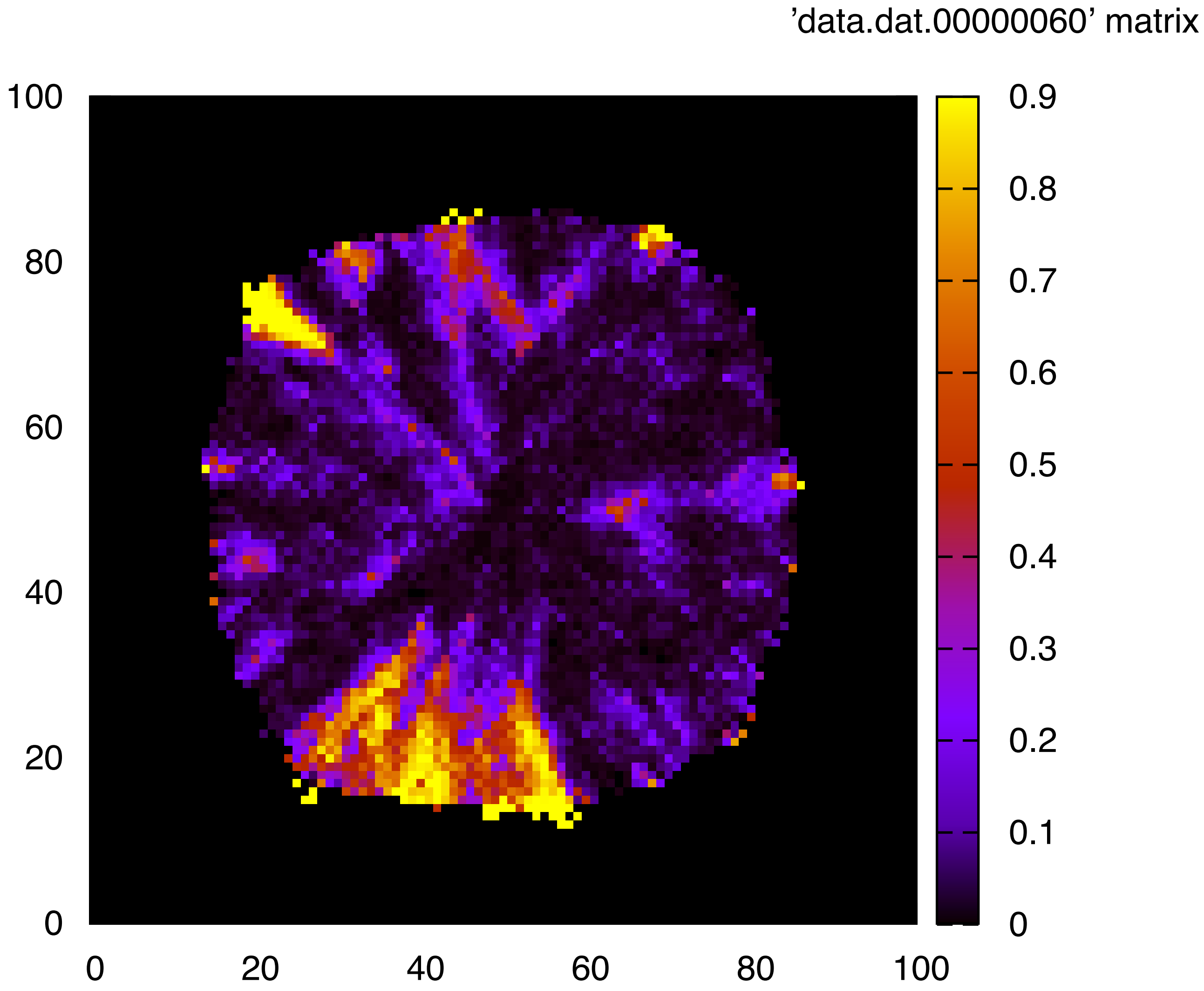
Ex5: 2D - Trace 1 (High, F=1)



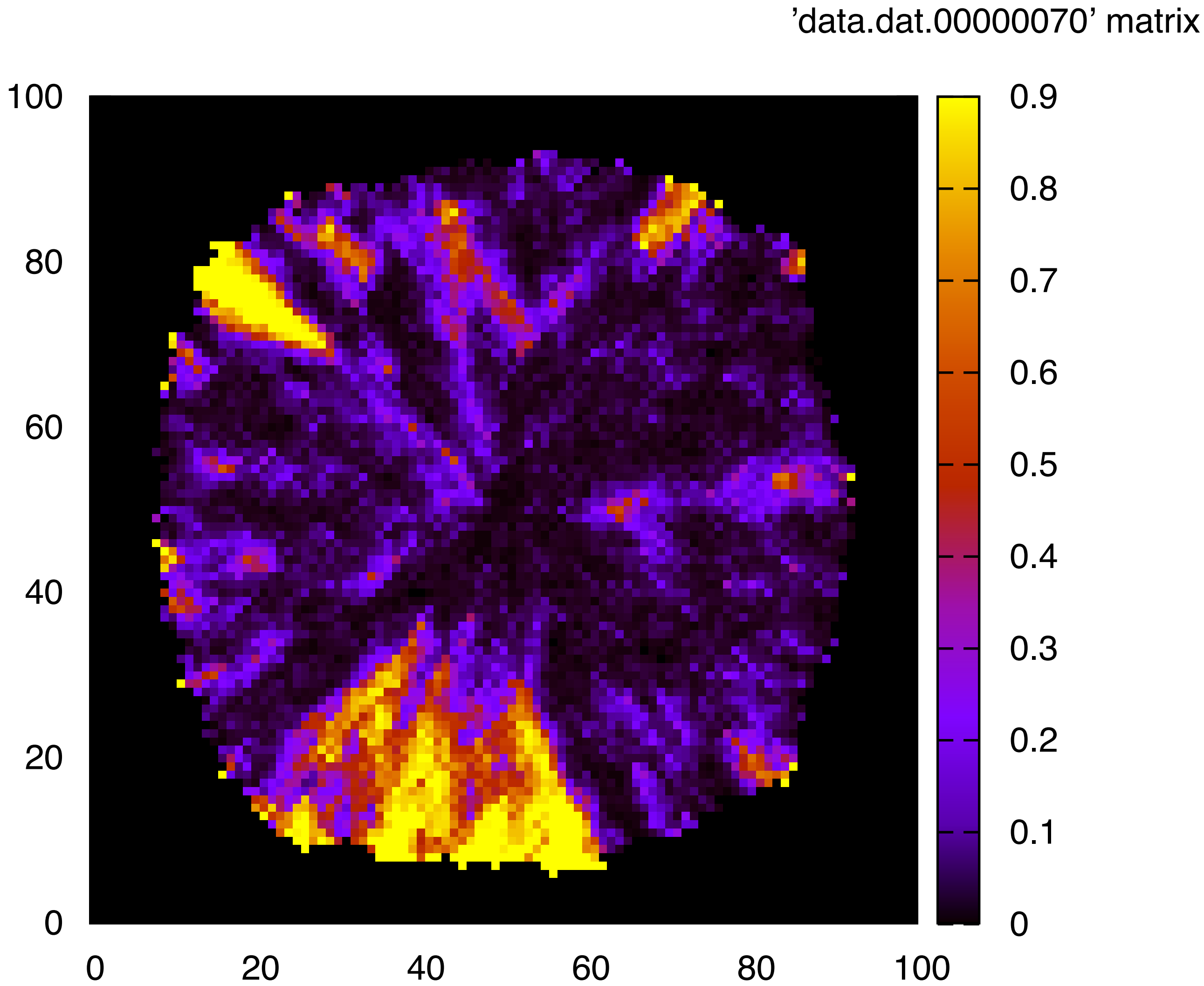
EX5: 2D - TRACE 1 (HIGH, F=1)



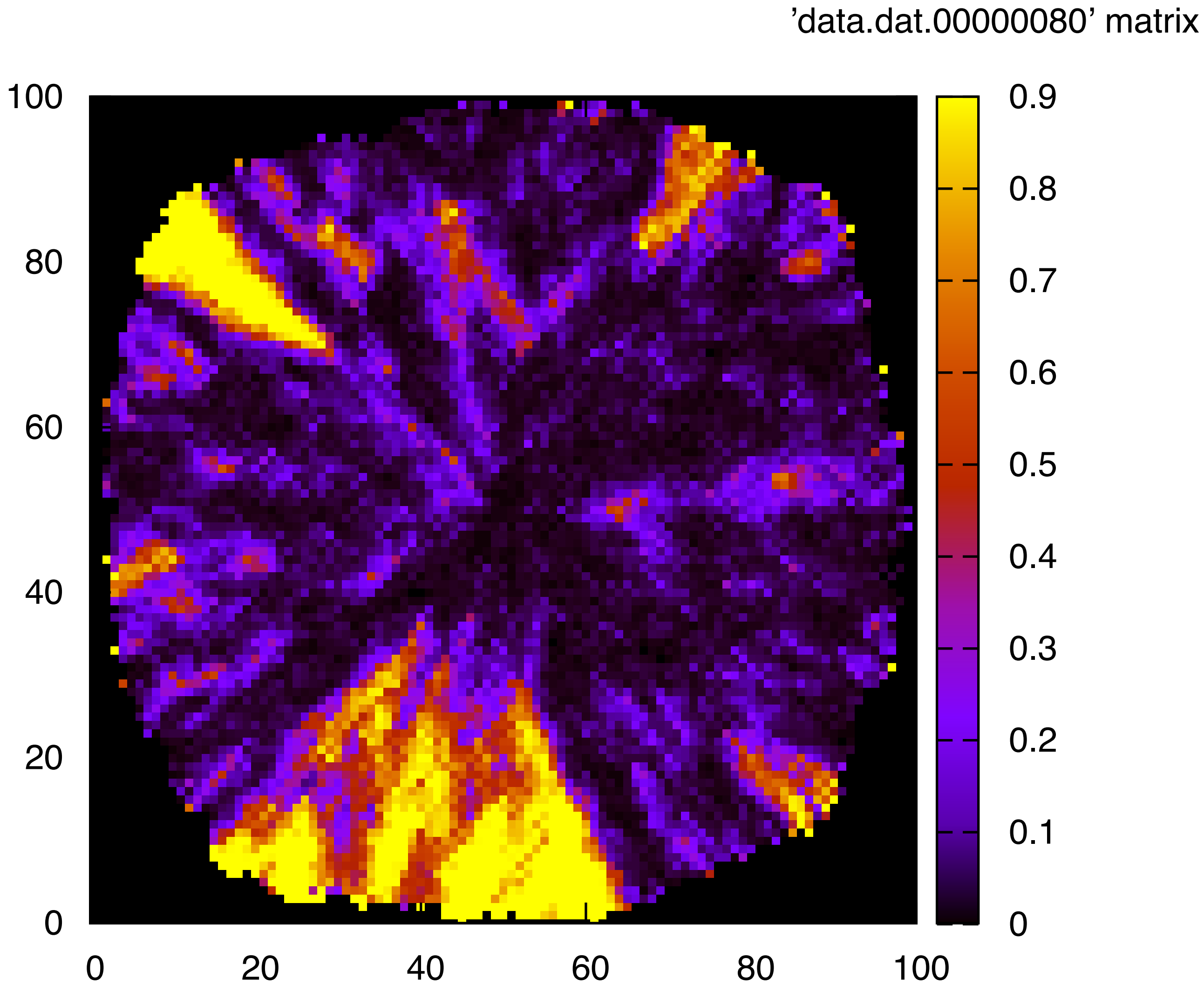
Ex5: 2D - Trace 1 (High, F=1)



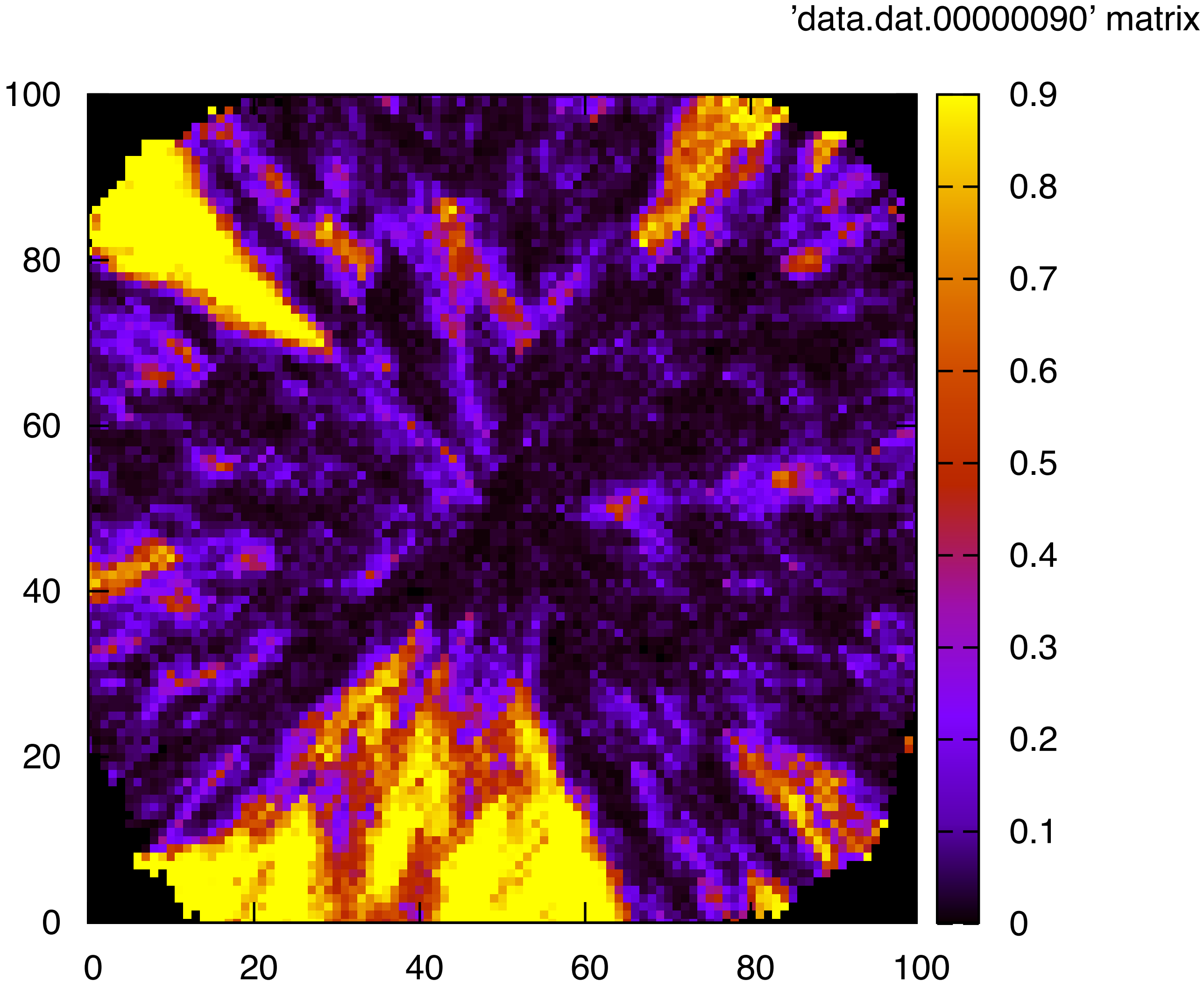
EX5: 2D - TRACE 1 (HIGH, F=1)



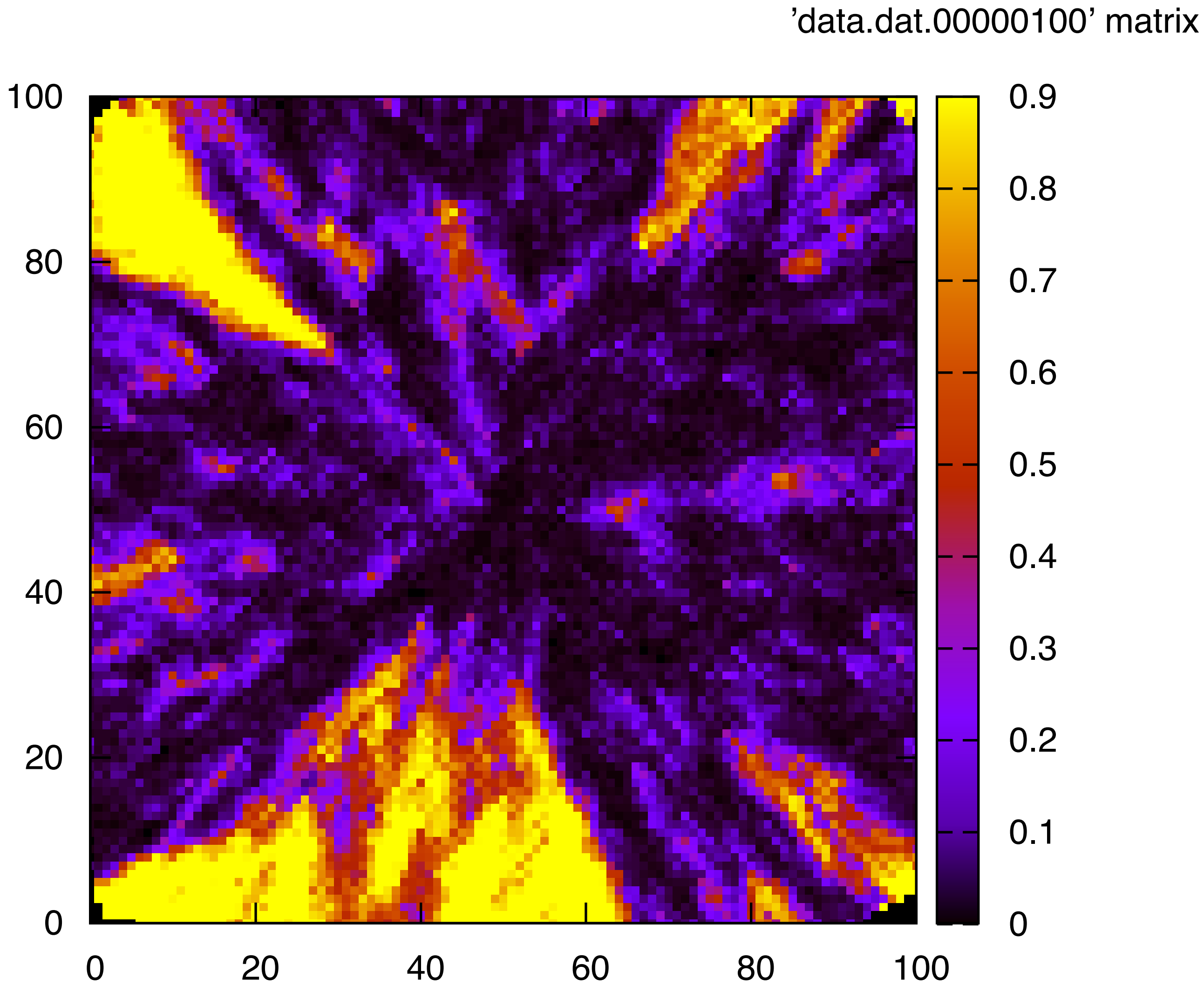
Ex5: 2D - Trace 1 (High, F=1)



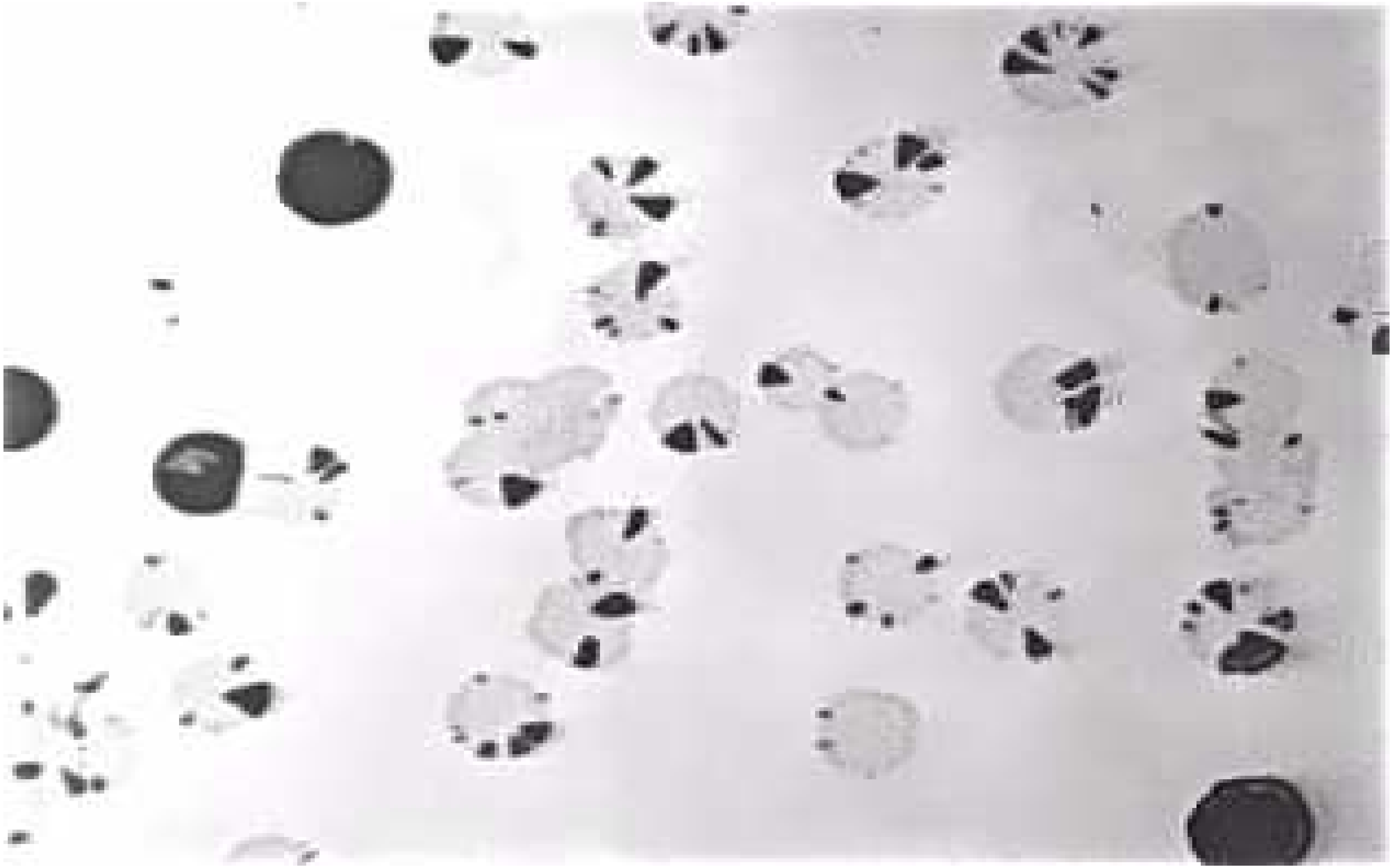
EX5: 2D - TRACE 1 (HIGH, F=1)



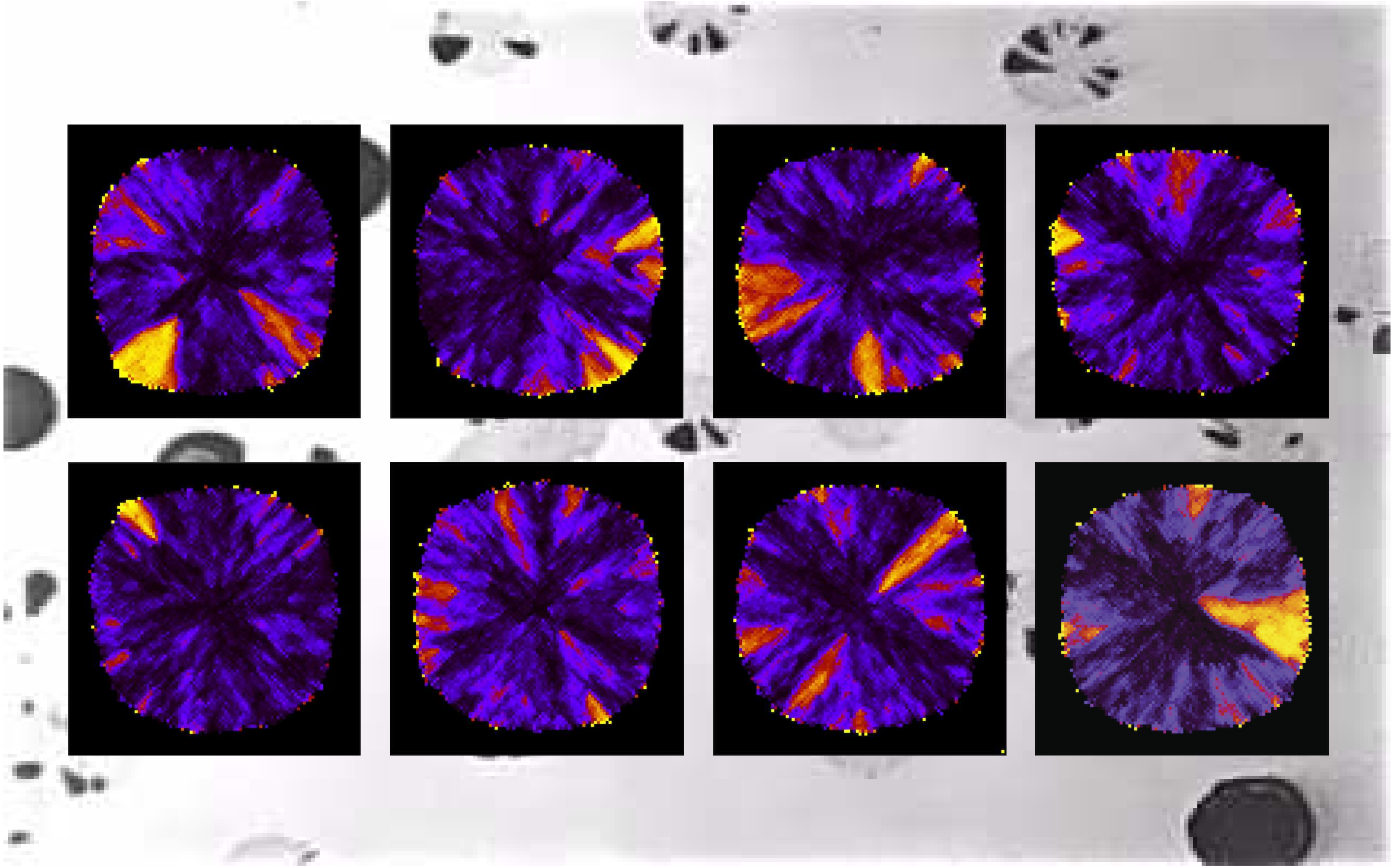
EX5: 2D - TRACE 1 (HIGH, F=1)



EX5: SOME FINAL STATES (HIGH, F=1)



EX5: SOME FINAL STATES (HIGH, F=1)

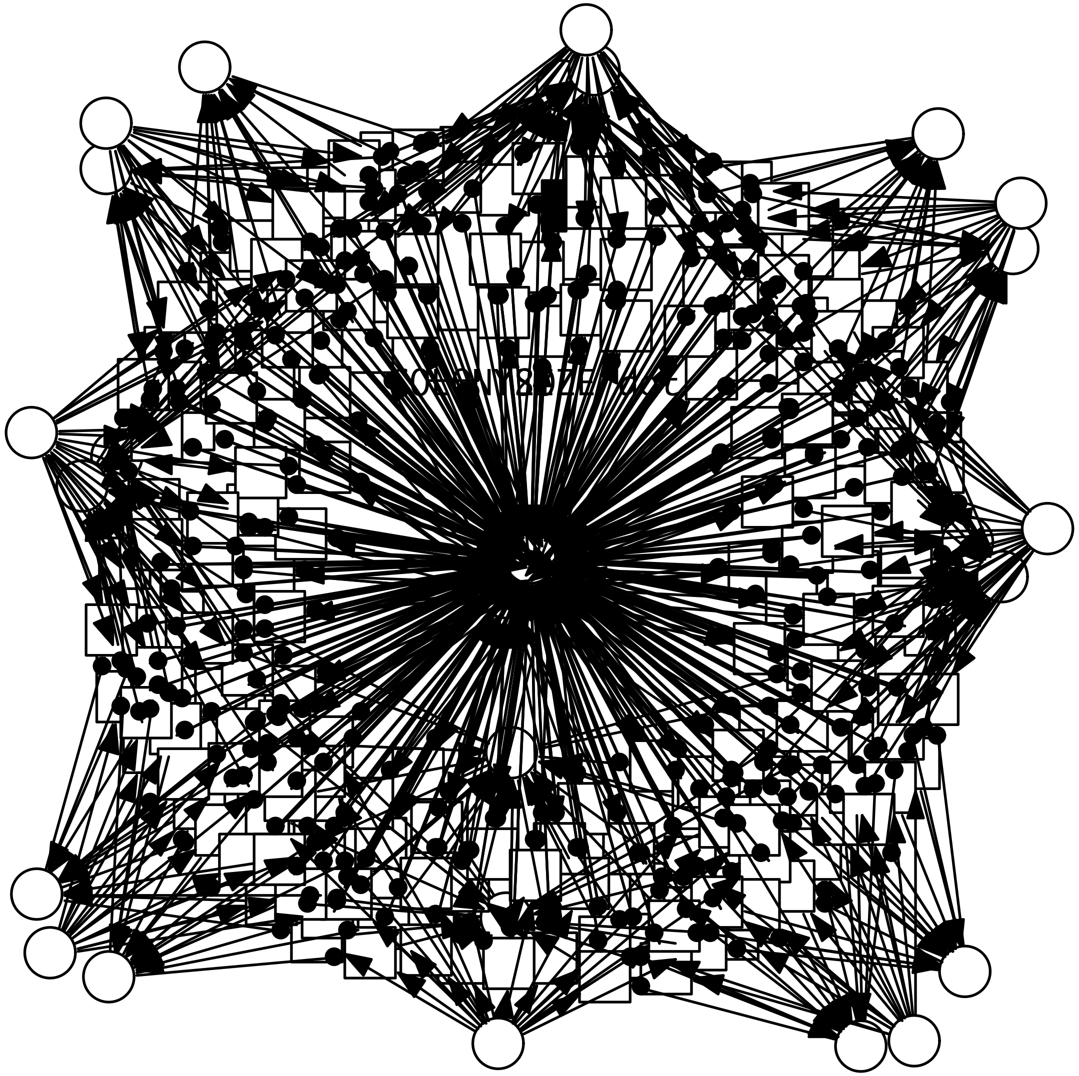


ADVANTAGES OF OUR APPROACH

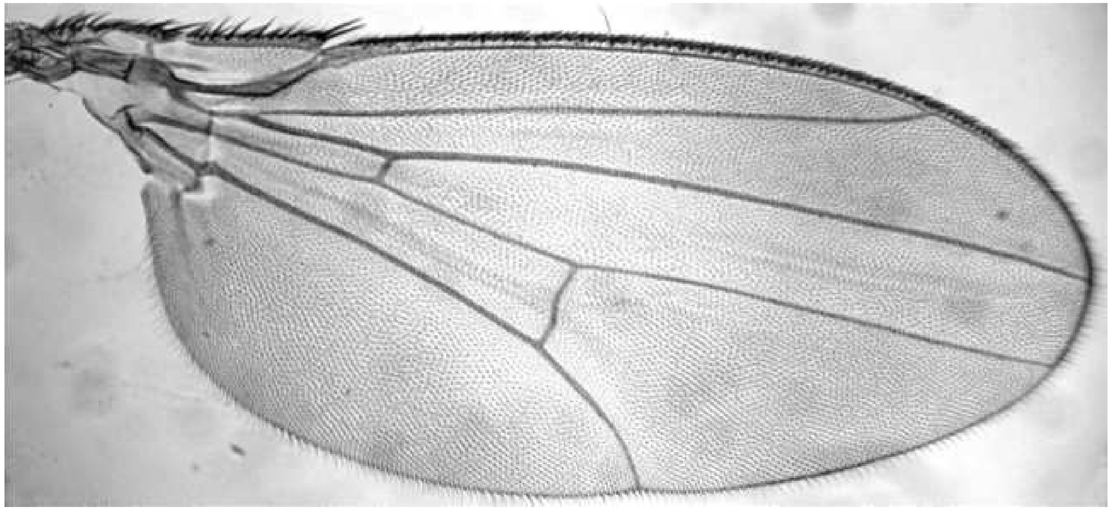
- **the spatial modelling principle can be equally applied to all paradigms**
  - -> *qualitative, stochastic, continuous, and hybrid*
  - -> *model transformations preserve all spatial attributes*
- **all space-related information is encoded in colour**
  - -> *reuse in other models*
- **changing the notion of space**
  - -> *adapt colour-related definitions*
  - -> *net structure itself needs not to be touched.*
- **use of a priori finitely discretised space preserves model analysibility**

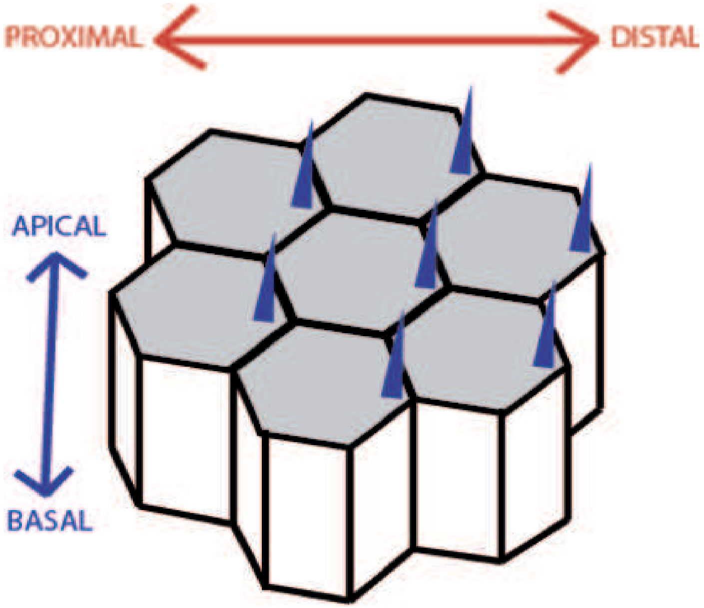
- **automatic unfolding**
  - -> *resue of all analysis and simulation techniques of uncoloured Petri nets*

PHASE VARIATION, PLAIN MODEL (3X3)



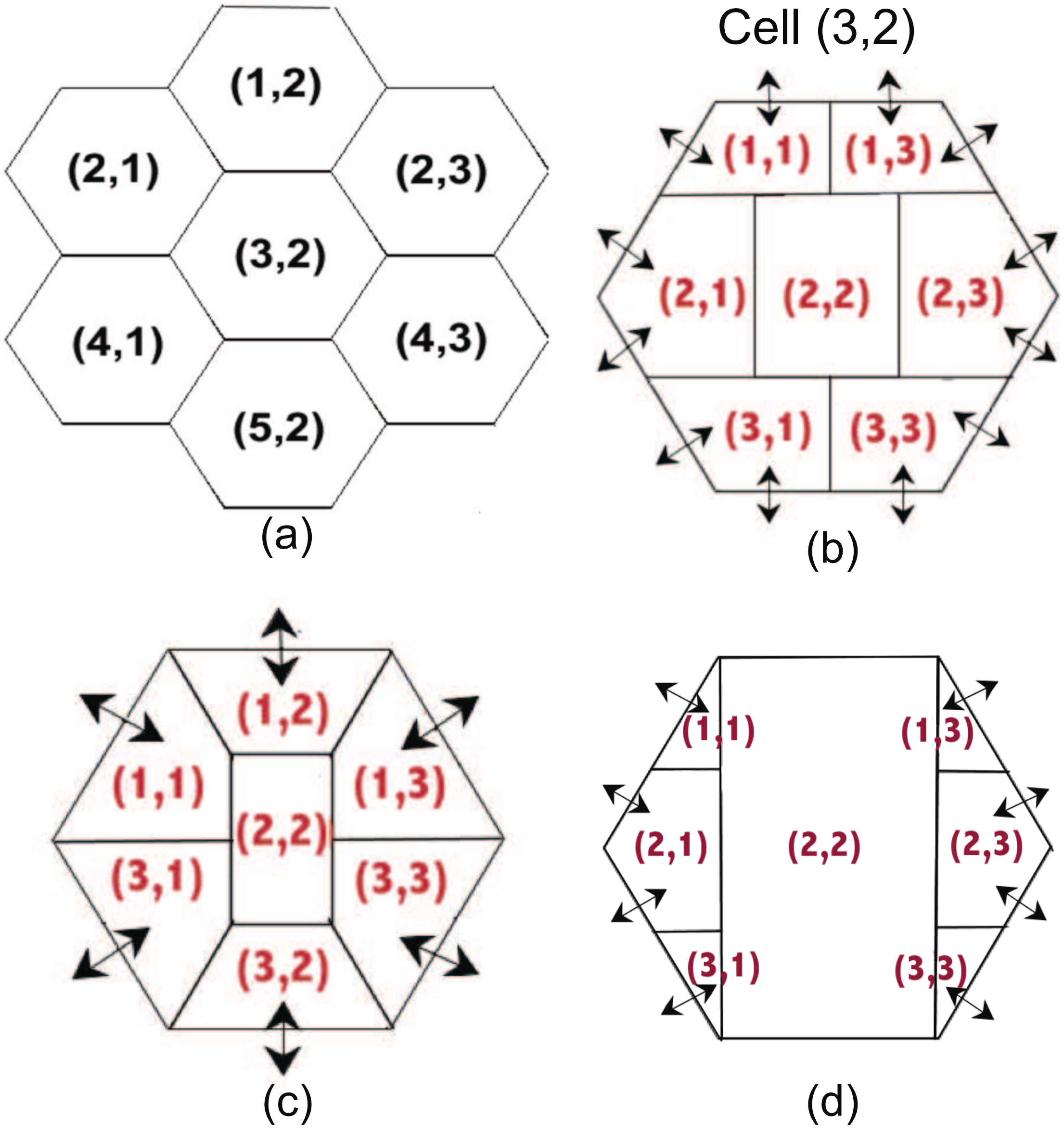
EX6 - PLANAR CELL POLARITY



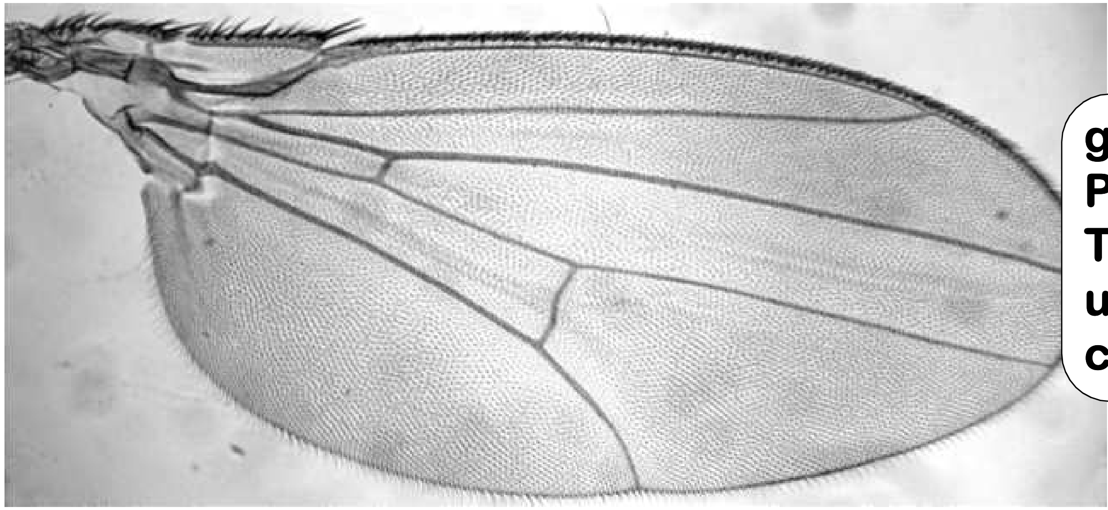


[BIOPPN 2011]
[CMSB 2011]

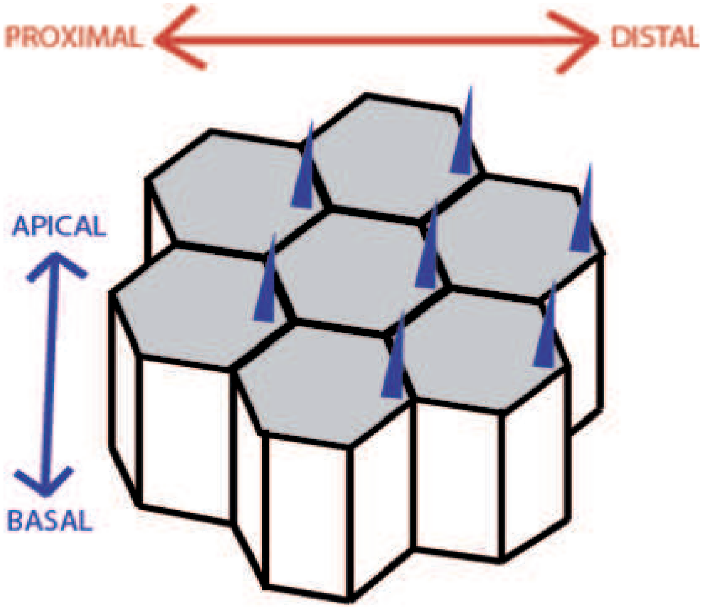
Ex6 - Planar Cell Polarity



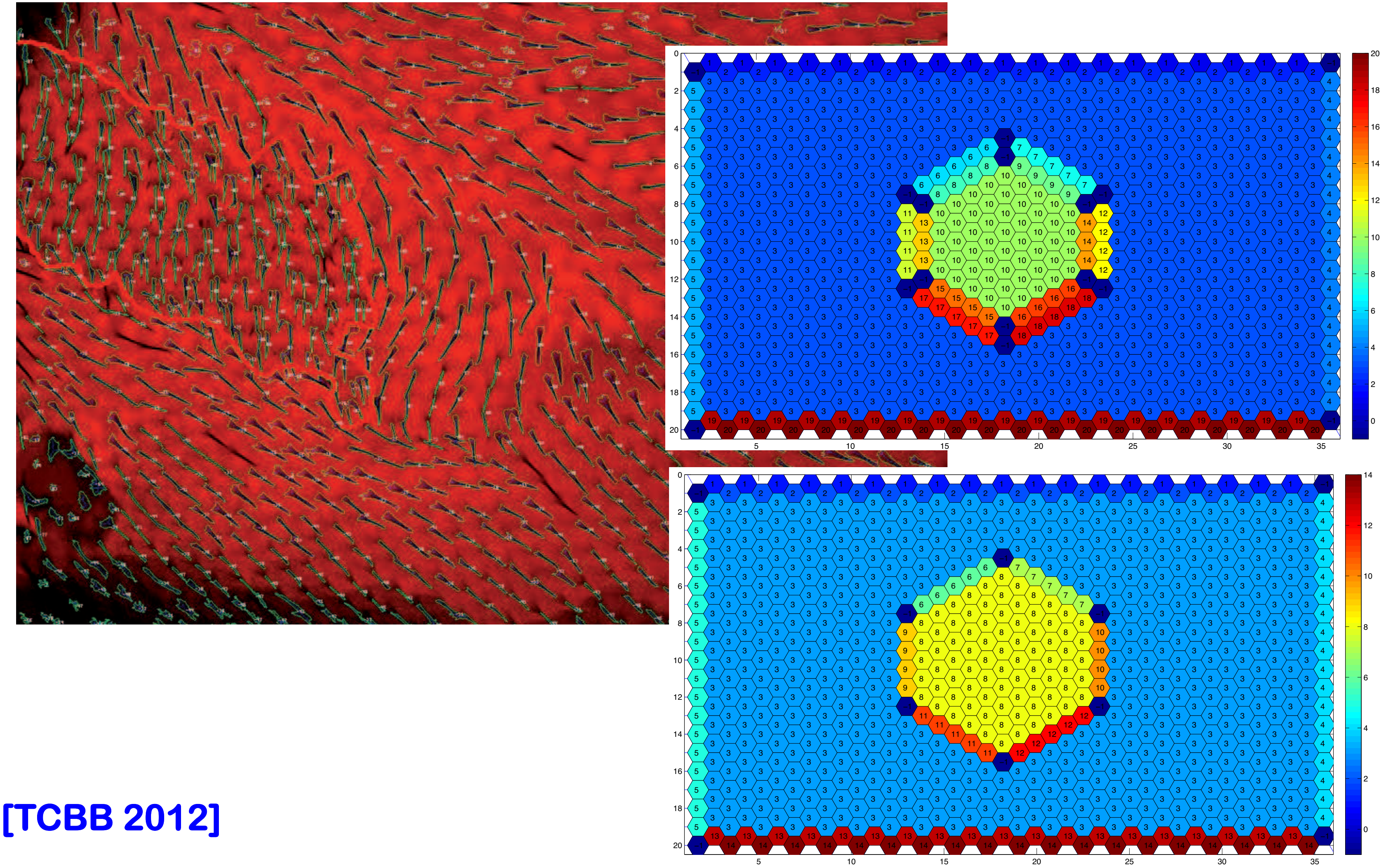
Ex6 - Planar Cell Polarity



grid size:	40 x 40
PLACES:	164,000
TRANSITIONS:	229,686
unfolding:	2 min
cont. simulation:	2 h



[BioPPN 2011]
[CMSB 2011]



[TCBB 2012]

SUMMARY & OUTLOOK

OUR TOOL BOX

- **SNOOPY**
  - -> *modelling and animation/simulation of hierarchical graphs,*
    e.g. (extended) fault trees,
    various Petri net classes, e.g. QPN, XQPN, SPN, XSPN, CPN, TPN,
    . . . ,
    free style graphs
- **CHARLIE**
  - -> *QPN, XQPN, Time/Timed Petri nets (TPN)*
  - -> *mostly standard analysis techniques of Petri net theory*
- **MARCIE**
  - -> *XQPN, SPN, XSPN, SRN*
  - -> *symbolic and simulative model checking*
- **Patty**
  - -> *animation via web browser*

- **SNOOPY**
  - -> *modelling and animation/simulation of hierarchical graphs,*
    e.g. (extended) fault trees,
    various Petri net classes, e.g. QPN, XQPN, SPN, XSPN, CPN, TPN,
    . . . ,
    tree style graphs

+

SBML import/export

- **CHARLIE**
  - -> *QPN, XQPN, Time/Timed Petri nets (TPN)*
  - -> *mostly standard analysis techniques of Petri net theory*

EXPORT TO MATAB AND

- **MARCIE**
  - -> *XQPN, SPN, XSPN, SRN*
  - -> *symbolic and simulative model checking*

MANY OTHER TOOLS

- **Patty**
  - -> *animation via web browser*

SUMMARY

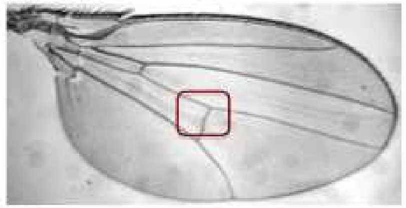
- **representation of bio networks by Petri nets**
  - -> *partial order representation* -> *better comprehension*
  - -> *formal semantics* -> *sound analysis techniques*
  - -> *unifying view*
- **purposes**
  - -> *animation* -> *to experience the model*
  - -> *model validation against consistency criteria* -> *to increase confidence*
  - -> *qualitative / quantitative behaviour prediction* -> *experiment design, new insights*
- **step-wise model development**
  - -> *qualitative model* -> *discrete Petri nets*
  - -> *discrete quantitative model* -> *stochastic Petri nets*
  - -> *continuous quantitative model* -> *continuous Petri nets = ODEs, hybrid models*
  - -> *locality and space* -> *coloured Petri nets*

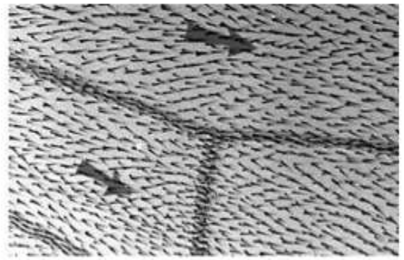
ABSTRACTION

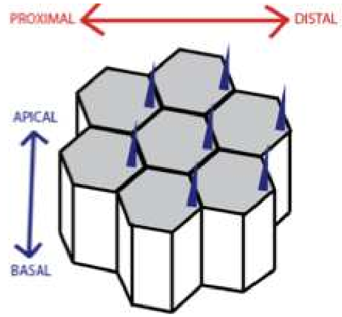
- **hierarchical organisation of components -> model variables**
  genes, molecules, organelles, cells, tissues, organs, organisms
- **functionality of atomic events**
  chemical reactions with/out stoichiometry, conformational change, transport, . . .
- **time**
  qualitative versus quantitative models
- **individual vs population behaviour** **-> hybrid**
- **(hierarchical) space**
- **observables**
- **shape and volume of components**
- **biosystem development**

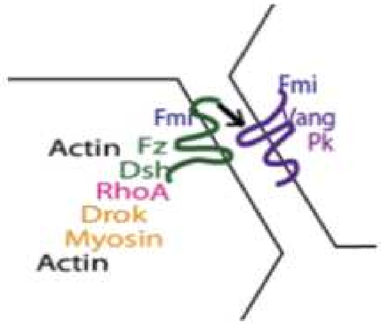
- repetition of components
- variation of components
- organisation of components
- communication between components
- mobility / motility
- replication / deletion of components
- hierarchical organisation of components

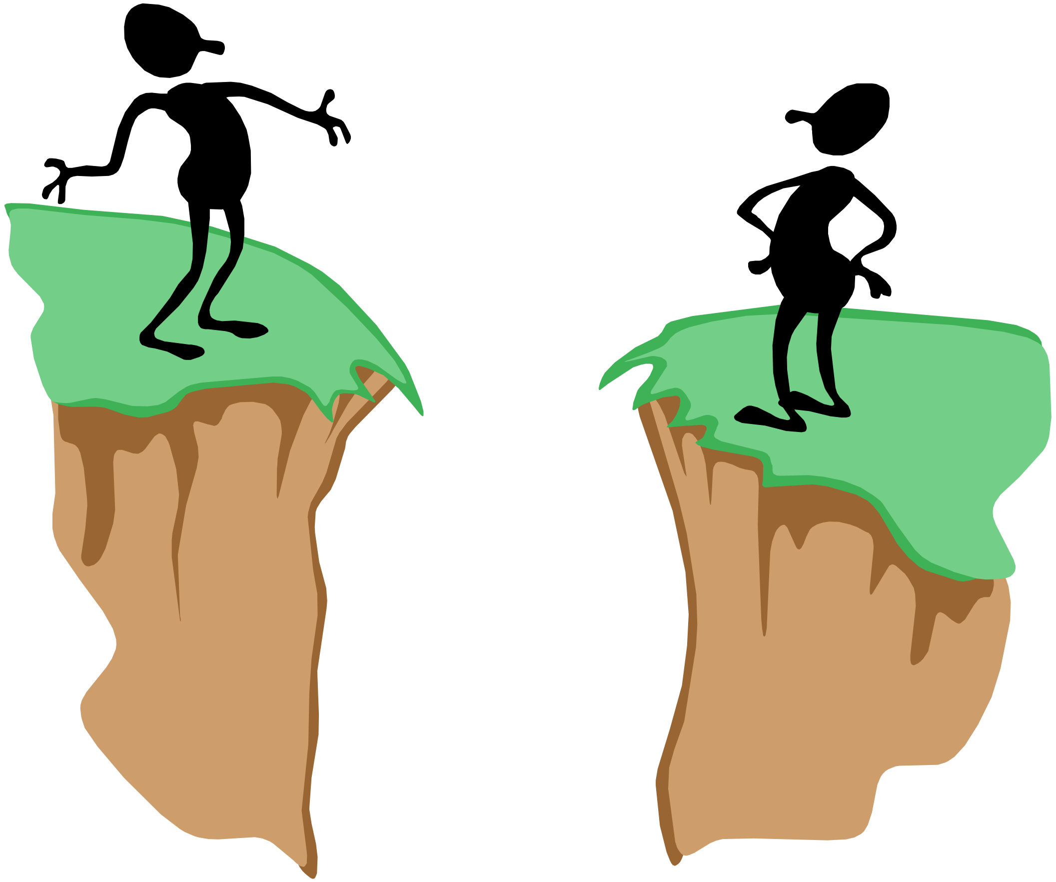
- dynamic grid size
- irregular / semi-regular organisation of components











THANKS !

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

APPENDIX

C. A. PETRI, NOVEMBER 2006

A BIT OF HISTORY

C. A. PETRI
INTERPRETATIONS OF NET THEORY
GMD, INTERNAL REPORT 75-07, 2ND IMPROVED EDITION 1976

places	transitions
state elements	transitional elements
conditions	events/facts
statements	dependencies
model domains	specifications
chemical compounds	chemical reactions
open one-point sets	closed one-point sets
channels	offices
languages	translators
products	poduction activities

[Peterson 1981]

74 *Chap. 3 Modeling with Petri Nets*

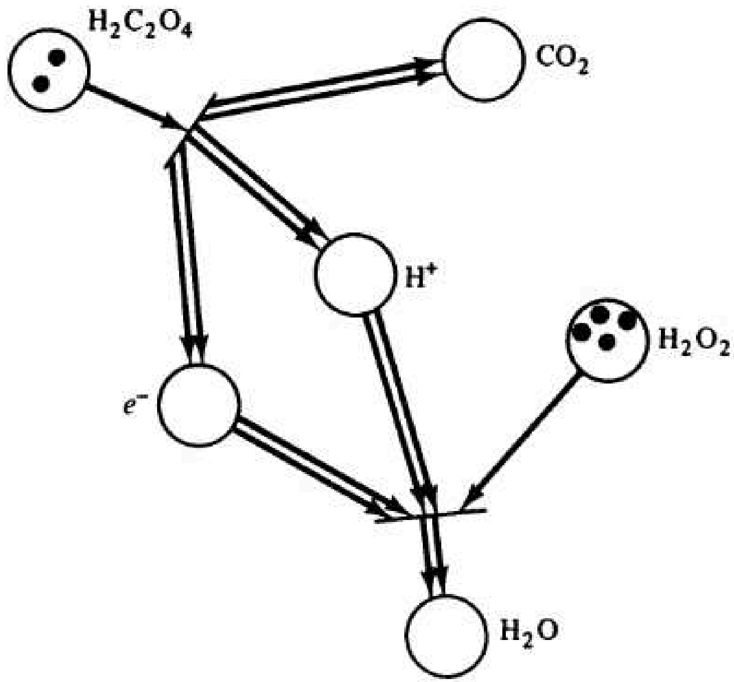


Figure 3.38 A Petri net representing the oxidation-reduction of oxalic acid and hydrogen peroxide into carbon dioxide and water.

$$H_2C_2O_4 \longrightarrow 2CO_2 + 2H^+ + 2e^-$$

$$2\,e^- + 2H^+ + H_2O_2 \longrightarrow 2H_2O$$

MORE HISTORY 2

[Murata 1989]

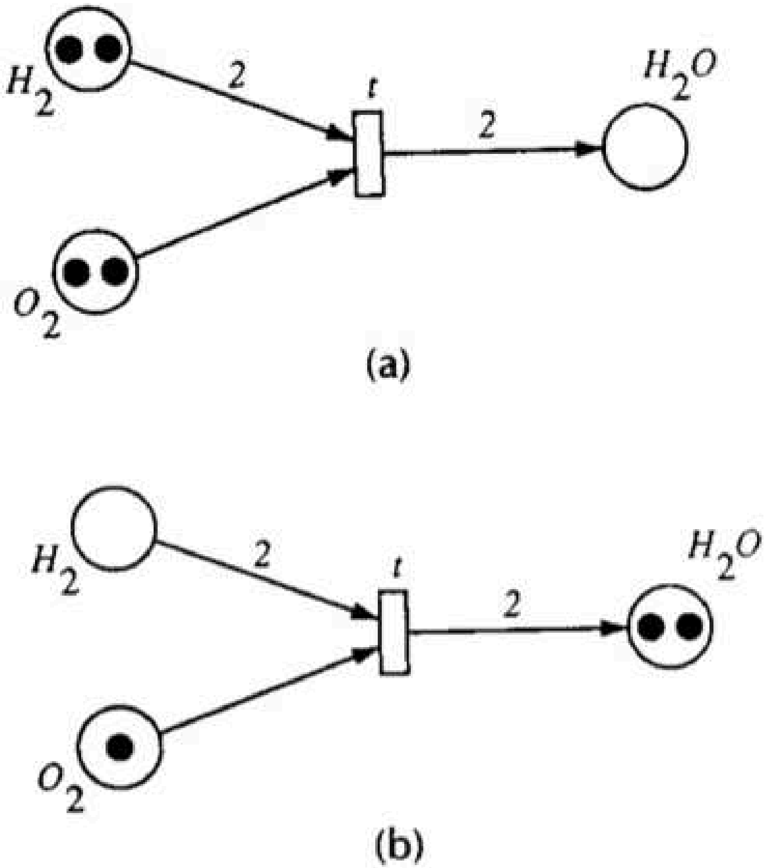


Fig. 1. Example 1: An illustration of a transition (firing) rule: (a) The marking before firing the enabled transition *t*. (b) The marking after firing *t*, where *t* is disabled.

STOCHASTIC PETRI NETS, BASICS

- **transitions r_i get a stochastic waiting time**

  -> *exponential distribution with parameter lambda*

- **state-dependent lambda defined by rate function $v_i(r_i)$**

  -> *any arithmetic function including*
  the transition's pre-places as integer variables and
  user-defined real-valued parameters

  -> *modifier arcs*

  -> *popular kinetics:*
  mass-action semantics, level semantics

- **semantics: Continuous Time Markov Chain (CTMC)**

  -> *reachability graph + state transition rates*

- **analysis**

  -> *standard Markov analysis techniques: transient, steady state*

  -> *stochastic simulation algorithms (SSA), e.g. Gillespie's SSA*

RATE FUNCTIONS

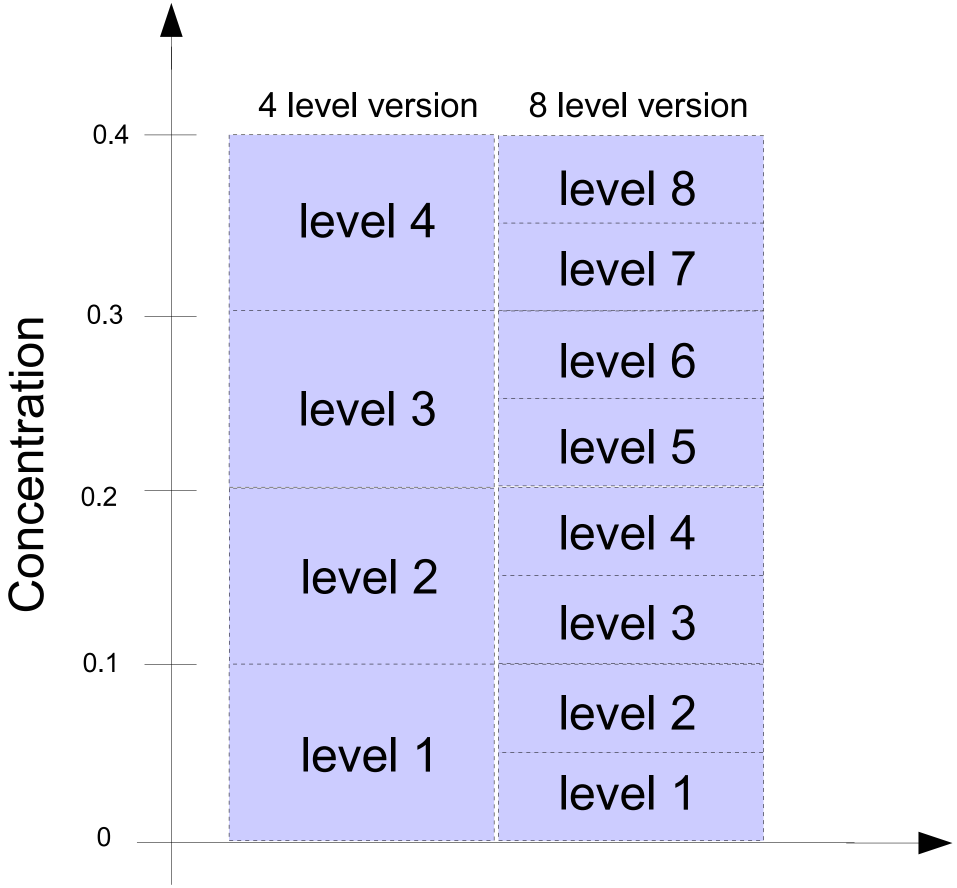
- **mass-action semantics**

$$h_t := c_t \cdot \prod_{p \in {}^{\bullet}t} \binom{m(p)}{f(p,t)}$$

- **level semantics**

$$h_t := k_t \cdot N \cdot \prod_{p \in {}^{\bullet}t} \left(\frac{m(p)}{N}\right)$$

LEVEL CONCEPT



CONTINUOUS PETRI NETS, BASICS

- **transitions r_i fire continuously**

- **rate functions $v_i(r_i)$**

  -> *any arithmetic function including*
  the transition's pre-places as real-valued variables and
  user-defined real-valued parameters

- **real-valued tokens**

  -> *concentrations*

- **semantics: set of Ordinary Differential Equations (ODEs)**

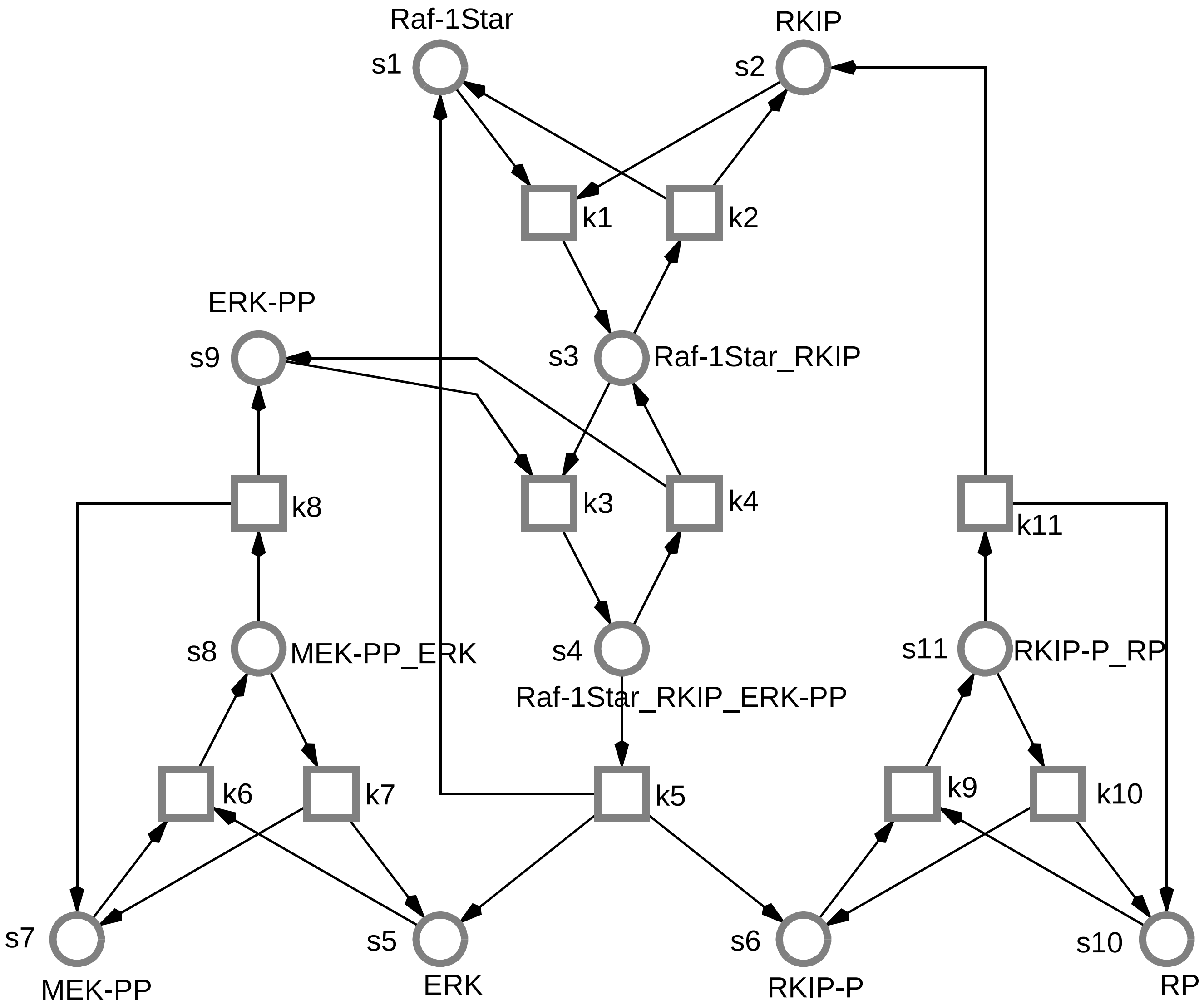
  -> *uniquely defined, but not vice versa* -> [SOLIMAN, HEINER 2010]

  -> *typically non-linear*

- **simulation (numerical integration)**

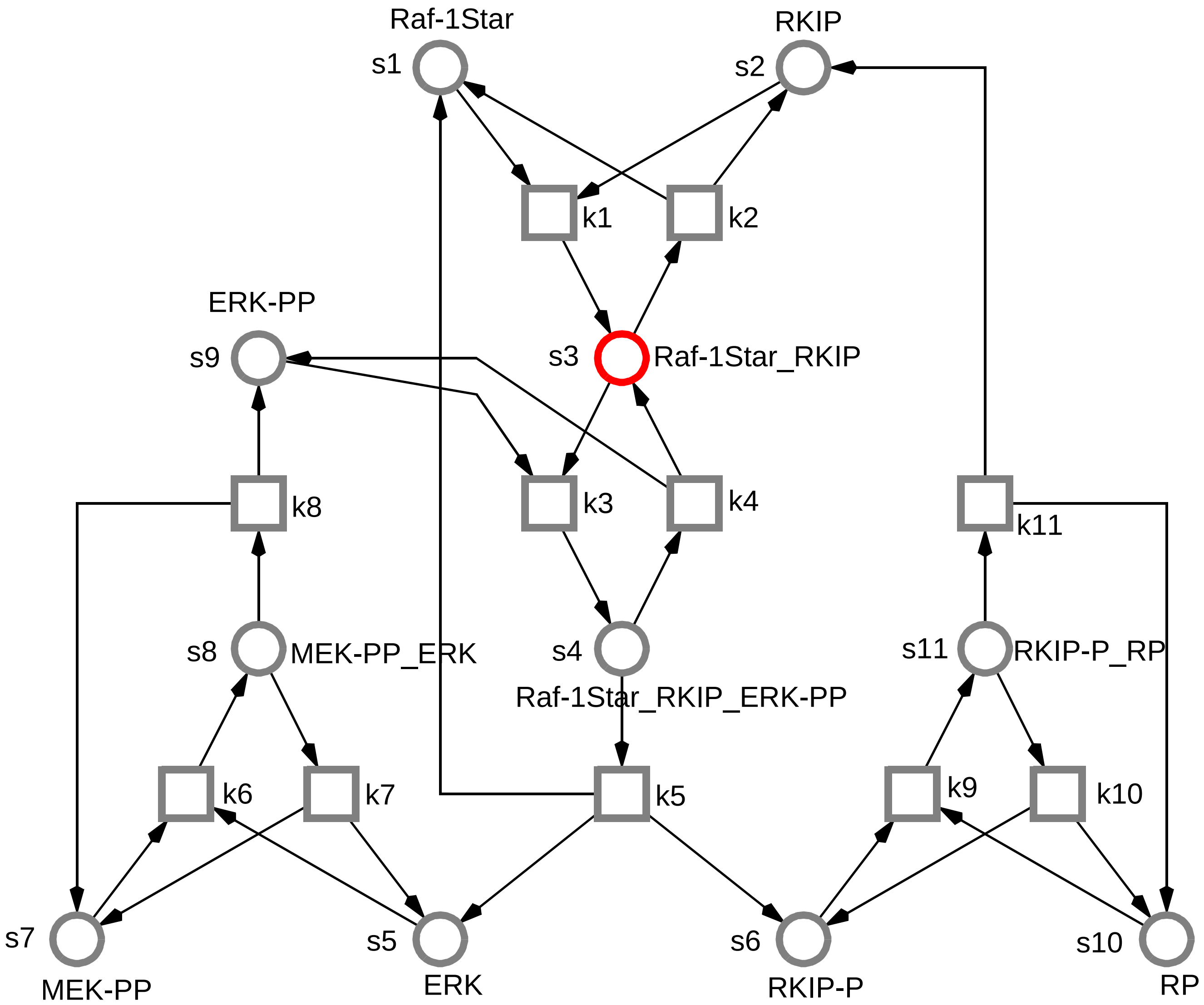
  -> *stiff / unstiff solvers*

Continuous Petri Net Defines ODEs



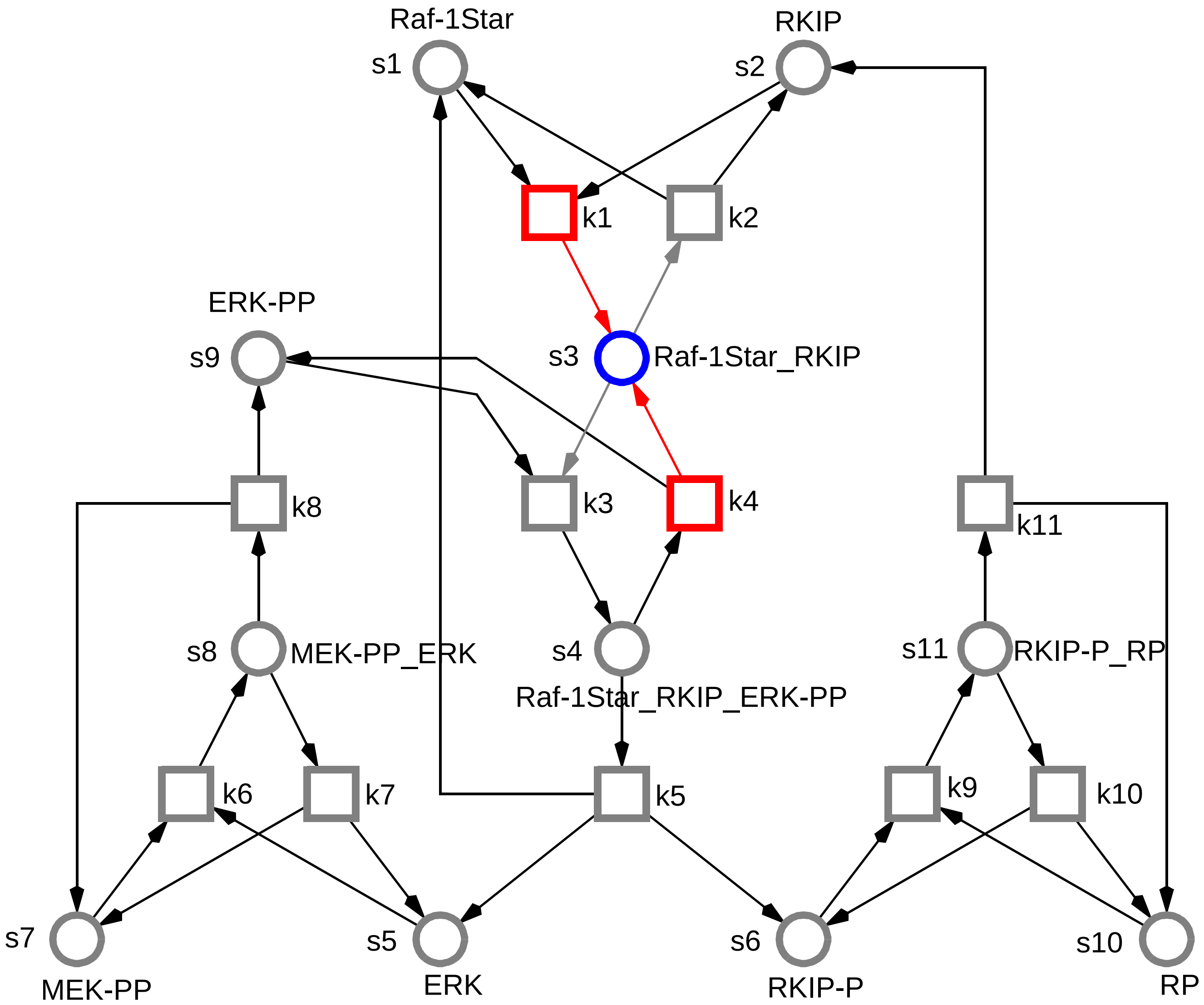
CONTINUOUS PETRI NET DEFINES ODEs

$$\frac{ds3}{dt} =$$



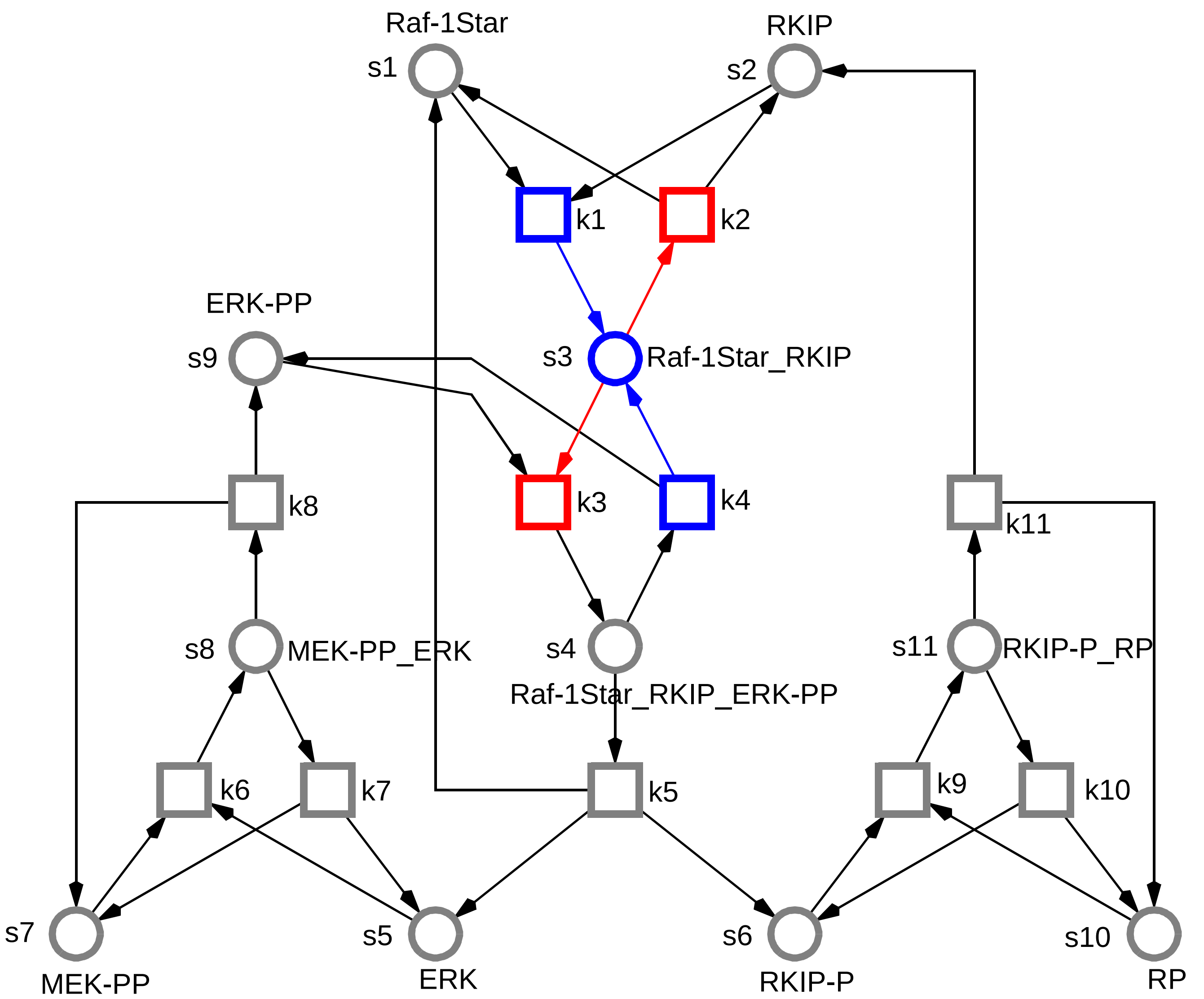
CONTINUOUS PETRI NET DEFINES ODES

$$\frac{ds3}{dt} = + v1 + v4$$



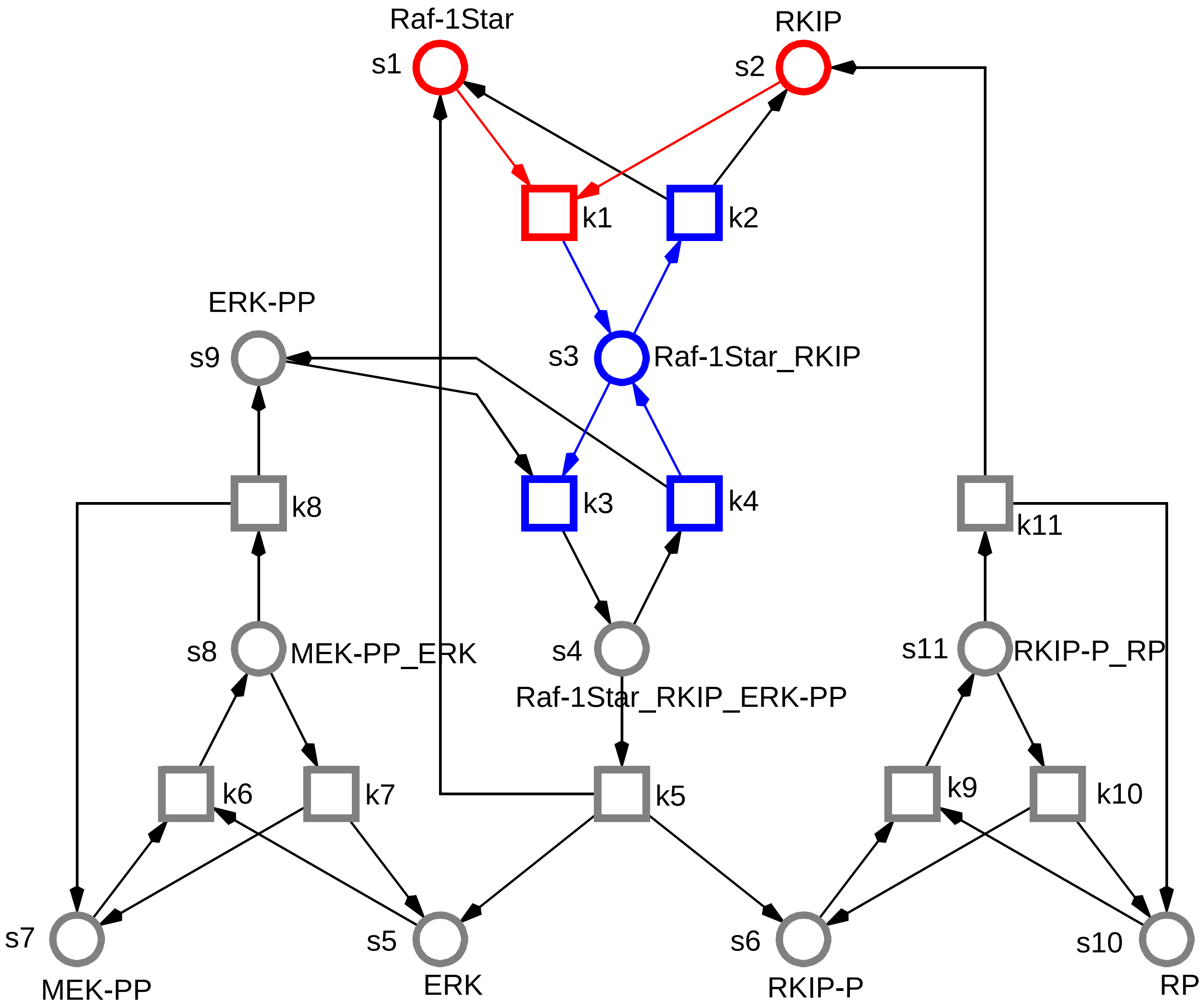
Continuous Petri Net Defines ODEs

$$\frac{ds3}{dt} = + v1 + v4 - v2 - v3$$



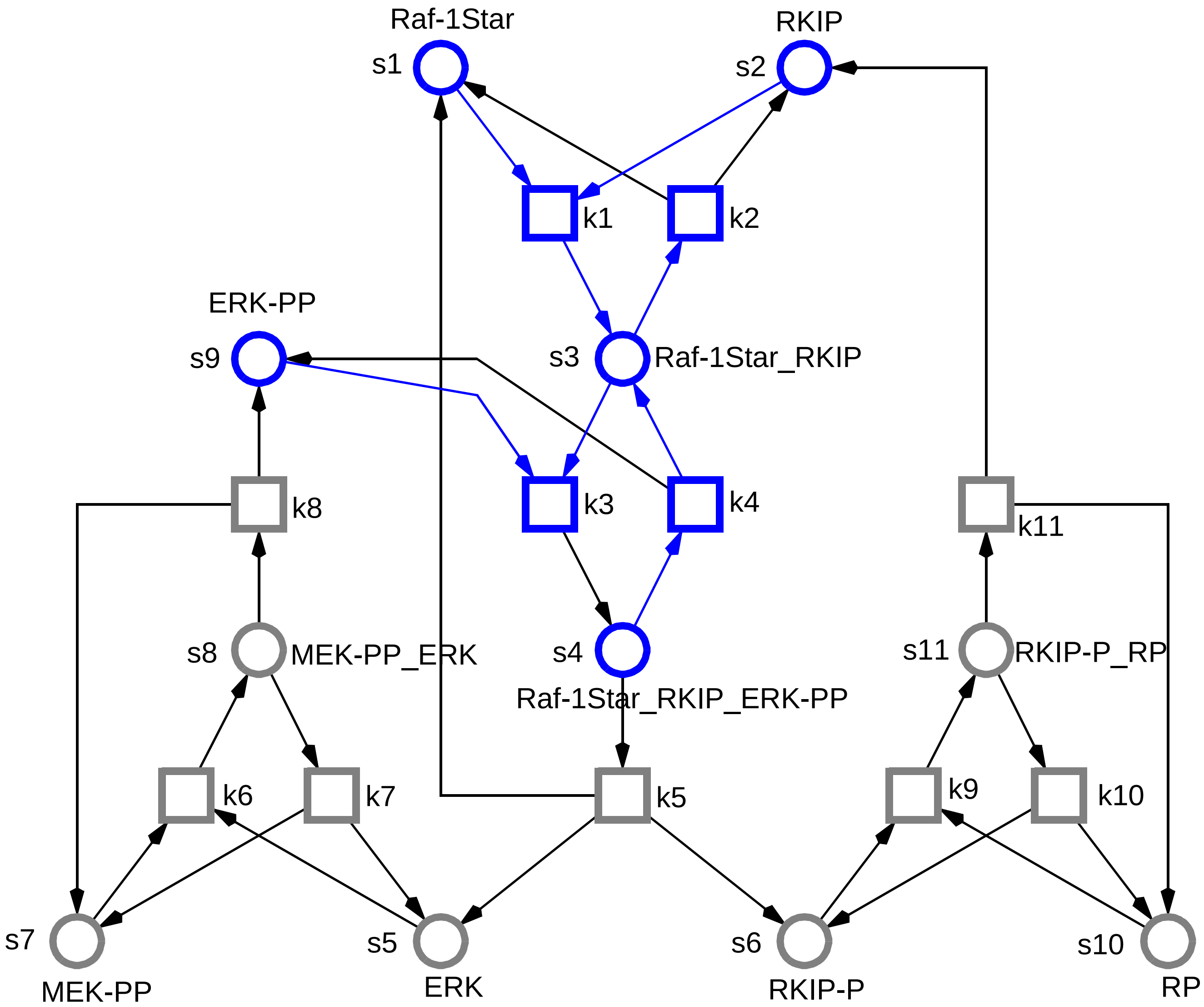
Continuous Petri Net Defines ODEs

$$\frac{ds3}{dt} = + k1 * s1 * s2 + v4 - v2 - v3$$



Continuous Petri Net Defines ODEs

$$\frac{ds3}{dt} = + k1 * s1 * s2 + k4 * s4 - k2 * s3 - k3 * s3 * s9$$



- **GHPN = XSPN + CPN** **[HERAJY, HEINER 2010]**

- **XSPN - Extended Generalized Stochastic Petri Nets**
  - -> *discrete places*
  - -> *discrete transitions: stochastic, immediate, deterministically delayed, scheduled*
  - -> *special arcs: read, inhibitor, equal, reset*

- **CPN - Continuous Petri Nets**
  - -> *continuous places*
  - -> *continuous transitions*
  - -> *special arcs: read, inhibitor*

- **hybrid simulation engine**
  - -> *static partitioning*
  - -> *dynamic partitioning*