
# Qualitative Modelling of Regulated Metabolic Pathways: Application to the Tryptophan Biosynthesis in *E. coli*

Claudine Chaouiya
chaouiya@igc.gulbenkian.pr
Instituto Gulbenkian de Ciência

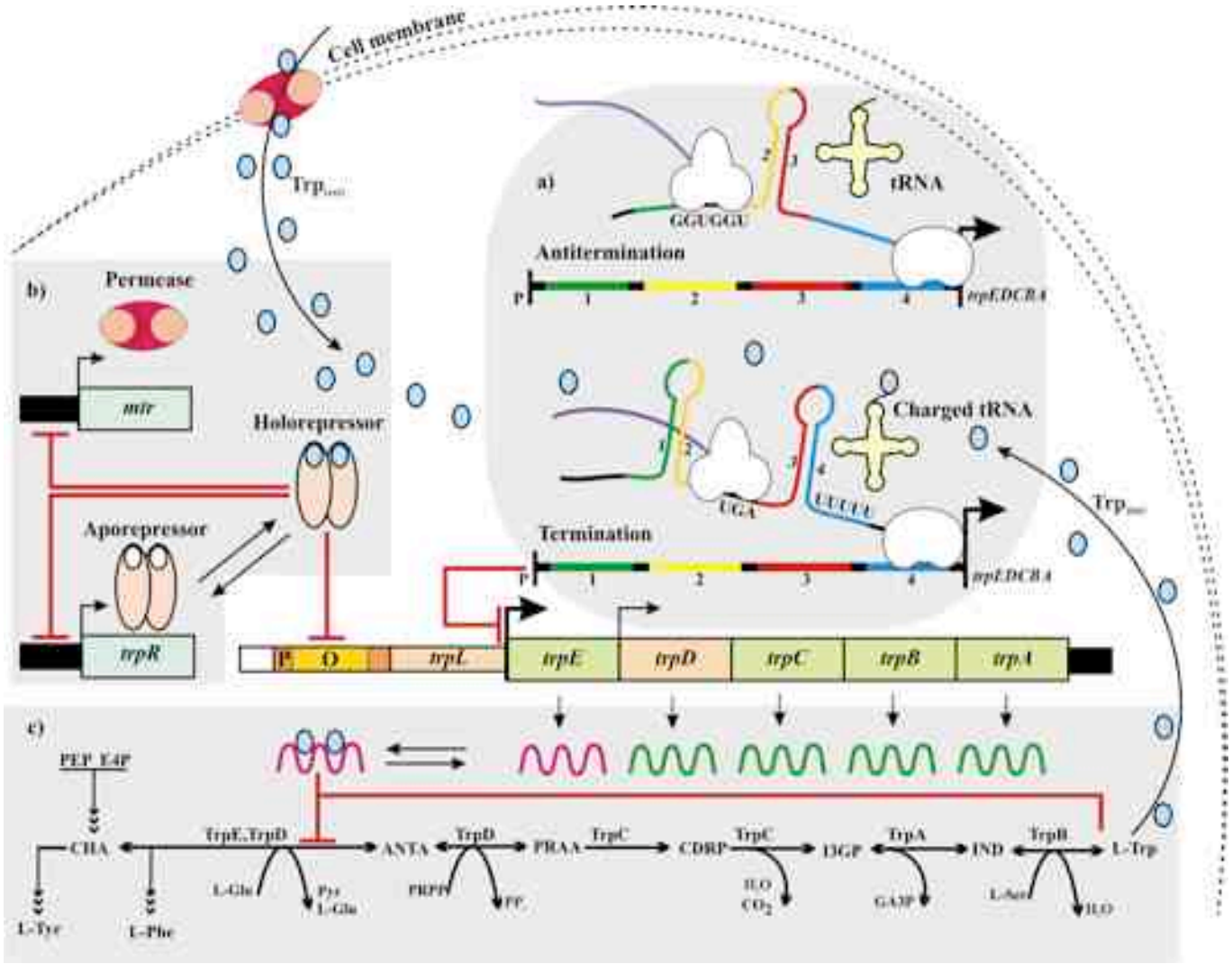
# Outline

- Motivation
- Petri nets, basics
- PN representation of *logical* regulatory networks

- Integrated modelling of the regulated Trp biosynthesis in *E. coli*

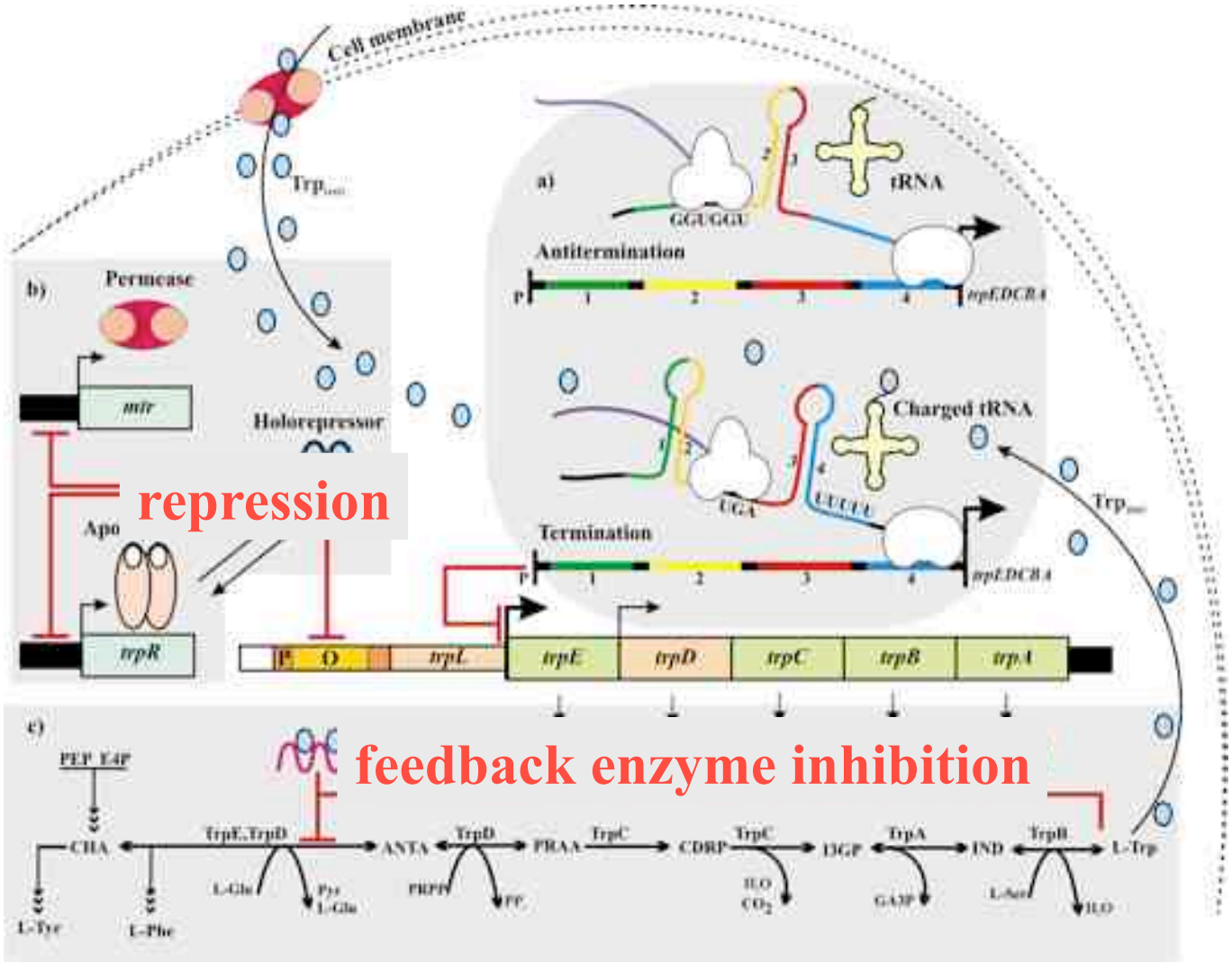
# Integrated modelling of regulated metabolic pathways

## The Tryptophan biosynthesis in *E. coli*

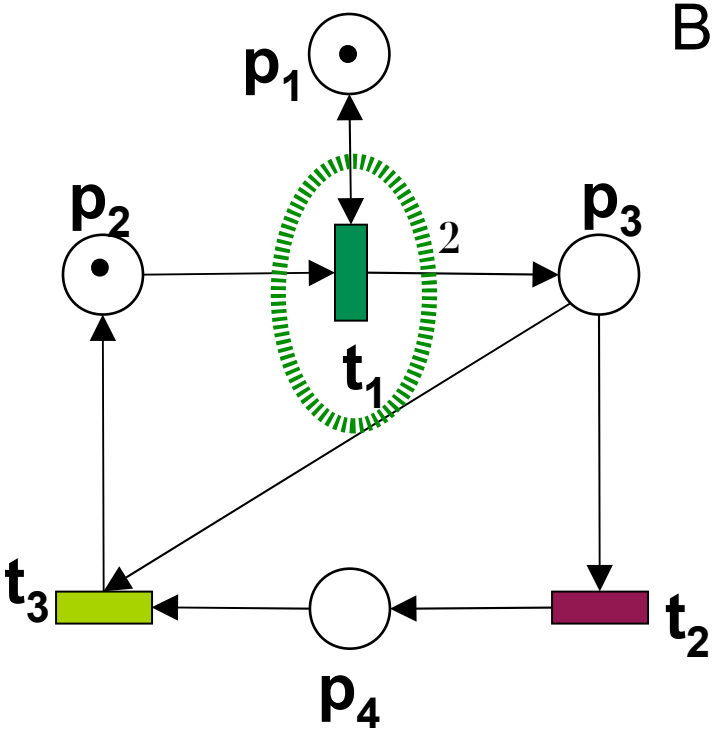


# Integrated modelling of regulated metabolic pathways

## The Tryptophan biosynthesis in *E. coli*



# Petri net basics




## Bipartite graph: places and transitions

- places resources
- marking state (number of tokens)
- transitions events
- weighted arcs connecting places ↔ transitions
- token game evolution of the system

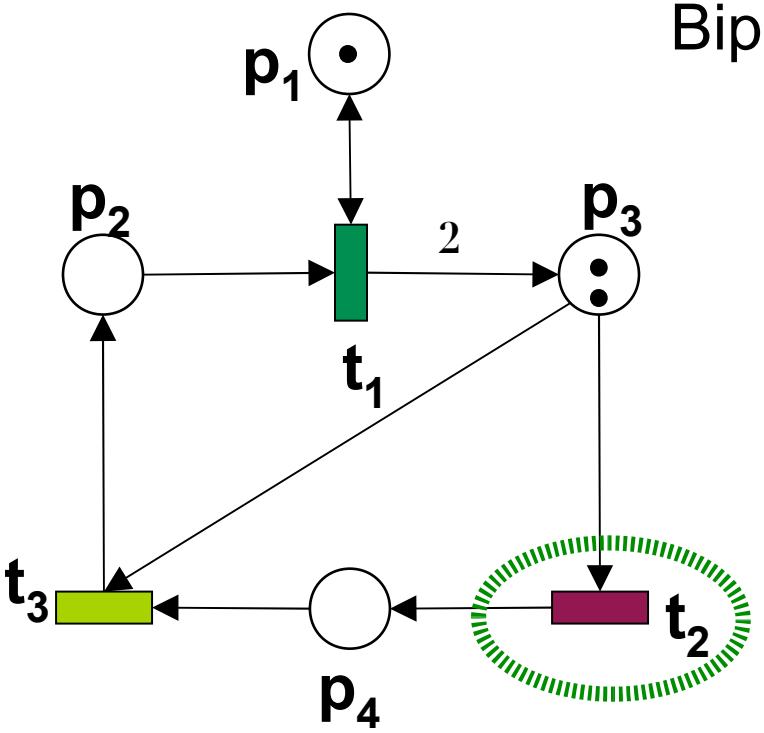
Transition t is enabled if its input places are *sufficiently* marked

The firing of t leads to the required

1/ consumption of tokens of input places

2/ production of tokens in output places

# Petri net basics




## Bipartite graph: places and transitions

- places → resources
- marking → state (number of tokens)
- transitions → events
- weighted arcs connecting places ü transitions
- token game →evolution of the system

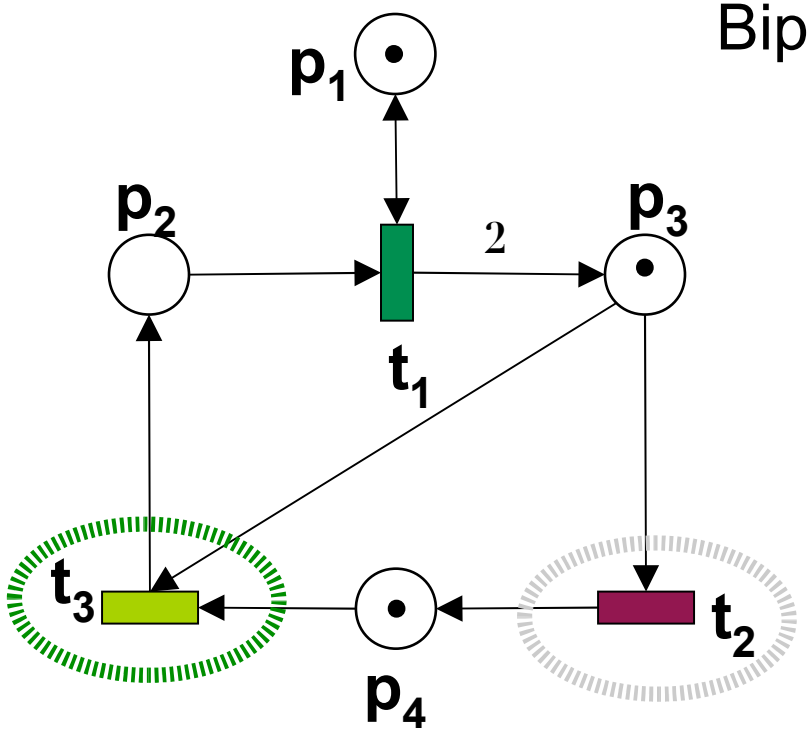
Transition t is enabled if its input places are *sufficiently* marked

The firing of t leads to the required

1/ consumption of tokens of input places

2/ production of tokens in output places

# Petri net basics




## Bipartite graph: places and transitions

- places resources
- marking state (number of tokens)
- transitions events
- weighted arcs connecting places ↔ transitions
- token game evolution of the system

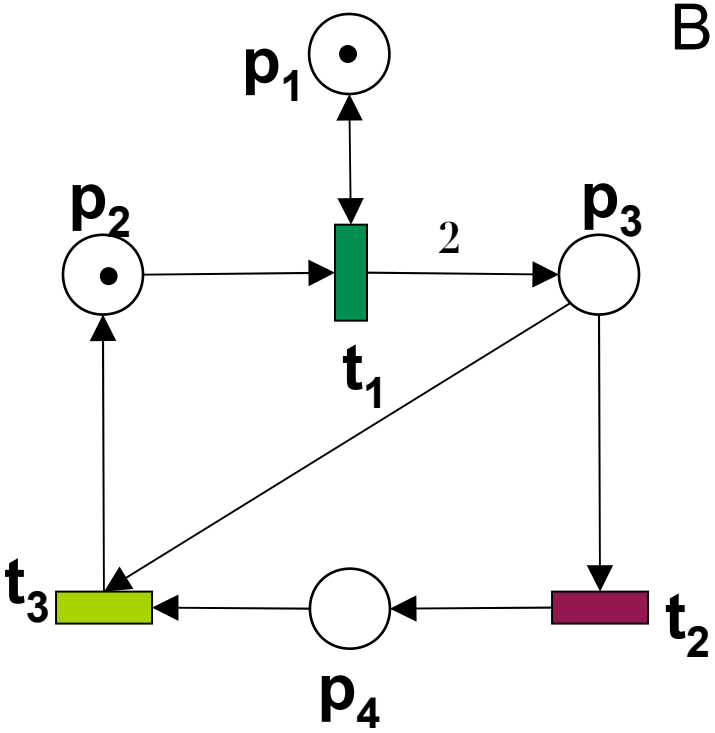
Transition t is enabled if its input places are *sufficiently* marked

The firing of t leads to the required

1/ consumption of tokens of input places

2/ production of tokens in output places

# Petri net basics




## Bipartite graph: places and transitions

- places  resources
- marking  state (number of tokens)
- transitions  events
- weighted arcs connecting places ↔ transitions
- token game  evolution of the system

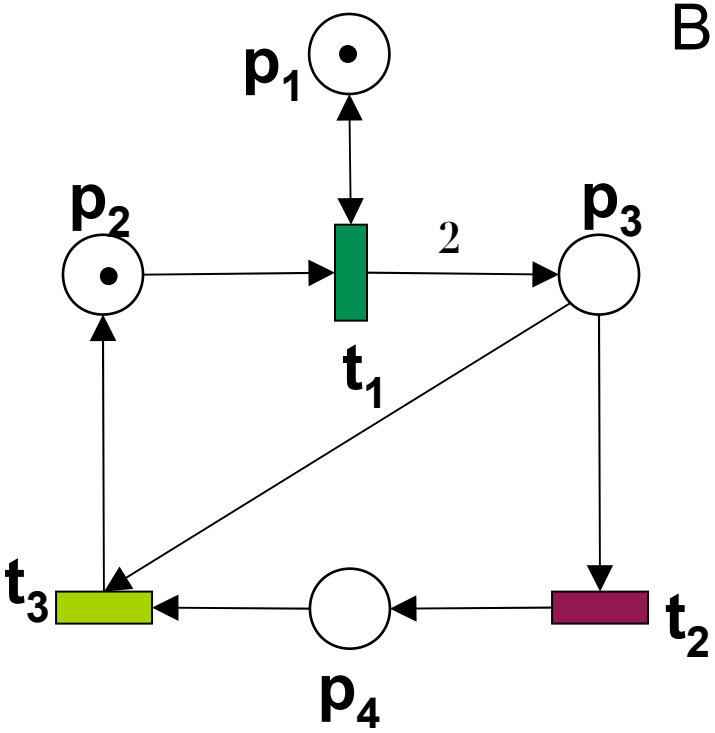
Transition t is enabled if its input places are *sufficiently* marked

The firing of t leads to the required

1/ consumption of tokens of input places

2/ production of tokens in output places

# Petri net basics




## Bipartite graph: places and transitions

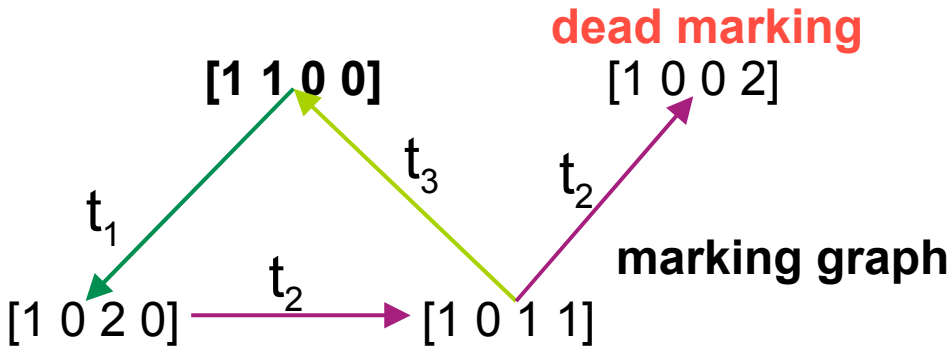
- places → resources
- marking → state (number of tokens)
- transitions → events
- weighted arcs connecting places ü transitions
- token game →evolution of the system

Transition t is enabled if its input places are *sufficiently* marked

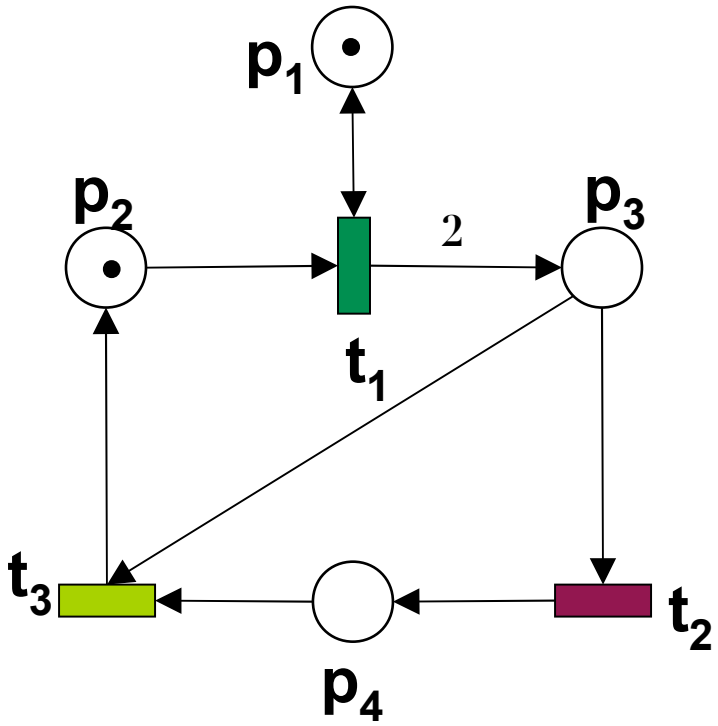
The firing of t leads to the required

1/ consumption of tokens of input places

2/ production of tokens in output places



# Petri net basics




## Mathematical representation

$$M_0 = \begin{bmatrix} 1 \\ 1 \\ 0 \\ 0 \end{bmatrix}$$

Initial marking

$$Pre = \begin{bmatrix} 1 & 0 & 0 \\ 1 & 0 & 0 \\ 0 & 1 & 1 \\ 0 & 0 & 1 \end{bmatrix}$$

PxT IN

$$Post = \begin{bmatrix} 1 & 0 & 2 & 0 \\ 0 & 0 & 0 & 1 \\ 0 & 1 & 0 & 0 \end{bmatrix}$$

TxP IN

Incidence matrix (PxT IN)

$$C = Post^{T} - Pre = \begin{bmatrix} \mathbf{0} & 0 & 0 \\ -1 & 0 & 1 \\ 2 & -1 & -1 \\ 0 & 1 & -1 \end{bmatrix}$$

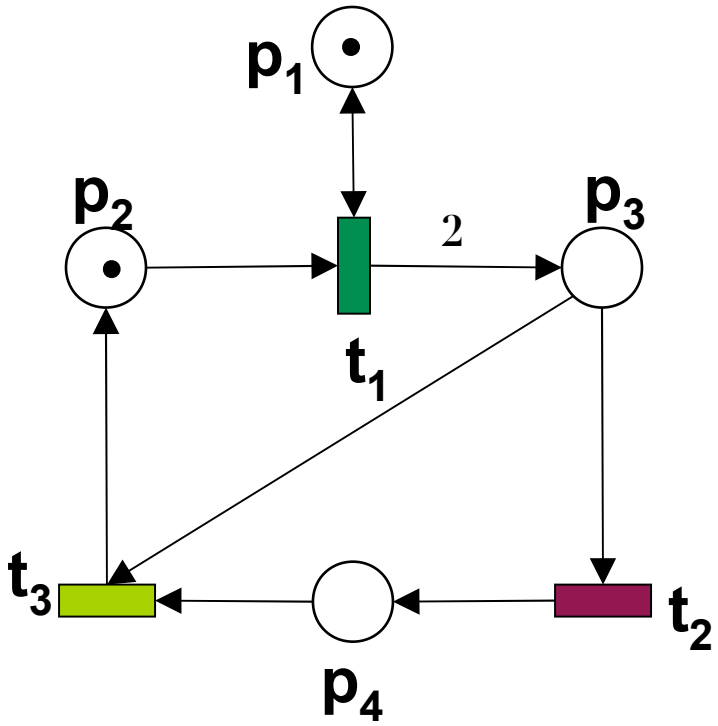
[1 1 0 0]

$t_1$

[1 0 2 0]

$$M_1 = M_0 + C \begin{bmatrix} 1 \\ 0 \\ 0 \end{bmatrix} = \begin{bmatrix} 1 \\ 0 \\ 2 \\ 0 \end{bmatrix}$$

# Petri net basics




## Mathematical representation

$$M_0 = \begin{bmatrix} 1 \\ 1 \\ 0 \\ 0 \end{bmatrix}$$

Initial marking

$$Pre = \begin{bmatrix} 1 & 0 & 0 \\ 1 & 0 & 0 \\ 0 & 1 & 1 \\ 0 & 0 & 1 \end{bmatrix}$$

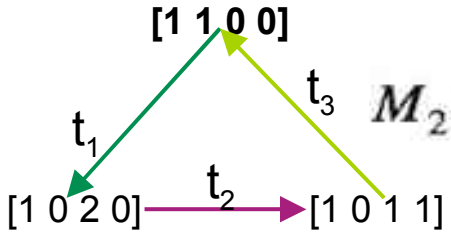
PxT IN

$$Post = \begin{bmatrix} 1 & 0 & 2 & 0 \\ 0 & 0 & 0 & 1 \\ 0 & 1 & 0 & 0 \end{bmatrix}$$

TxP IN

Incidence matrix (PxT IN)

$$C = Post^{T} - Pre = \begin{bmatrix} \mathbf{0} & 0 & 0 \\ -1 & 0 & 1 \\ 2 & -1 & -1 \\ 0 & 1 & -1 \end{bmatrix}$$




$$M_2 = M_0 + C \begin{bmatrix} 2 \\ 2 \\ 1 \end{bmatrix} = \begin{bmatrix} 1 \\ 0 \\ 1 \\ 1 \end{bmatrix}$$

# Petri net basics

- Mathematical and graphical formalism
- Representation of conflict/parallelism

**Properties**

- Structural: P-invariants (conservative components); T-invariants (repetitive components)
- Dynamical: liveness; boundness; reachability

**Tools**

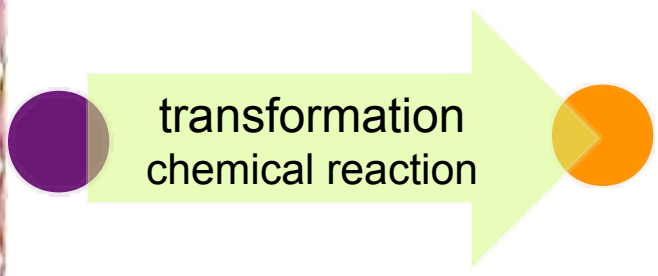
- Analytical approaches: state equations, algebraic equations, graph analysis...
- Model checking
- Simulation

a variety of analysis tools and simulation shells available

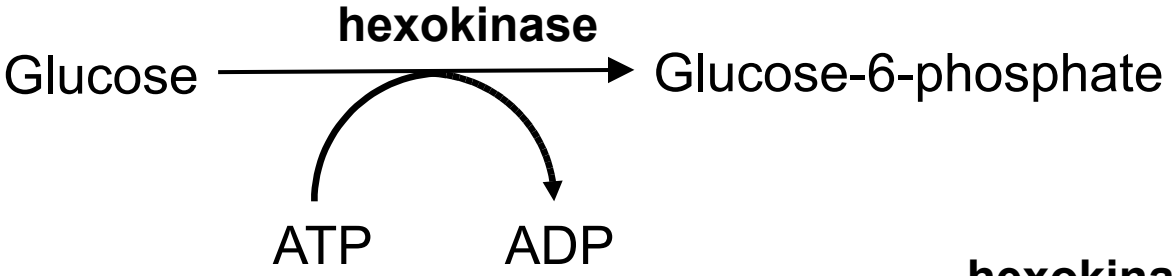
G. Balbo & M. Heiner lectures tomorrow!

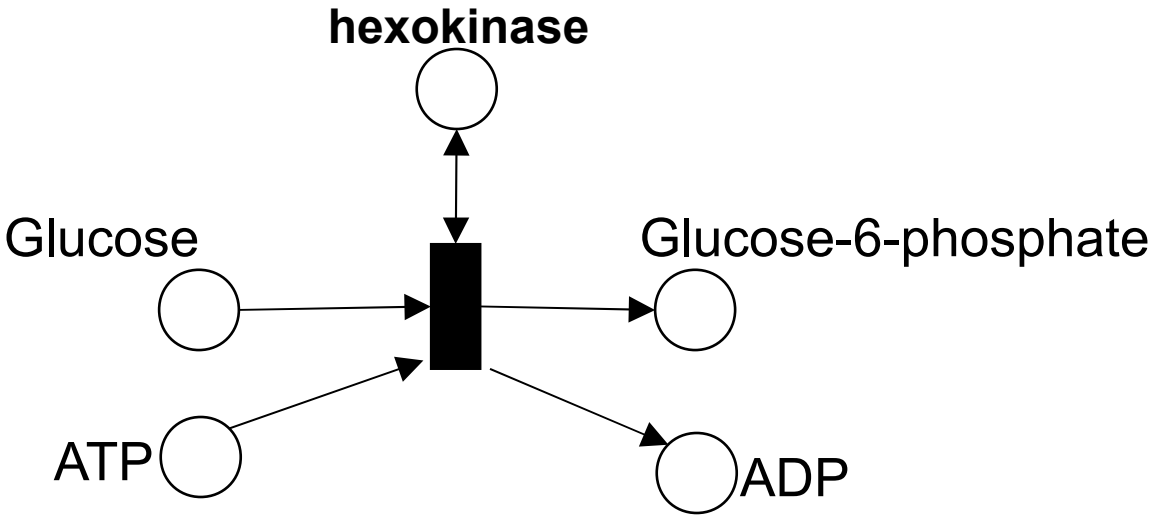
**Extensions** Stochastic PN, Coloured nets, Hybrid nets...

# PN modelling of reaction networks



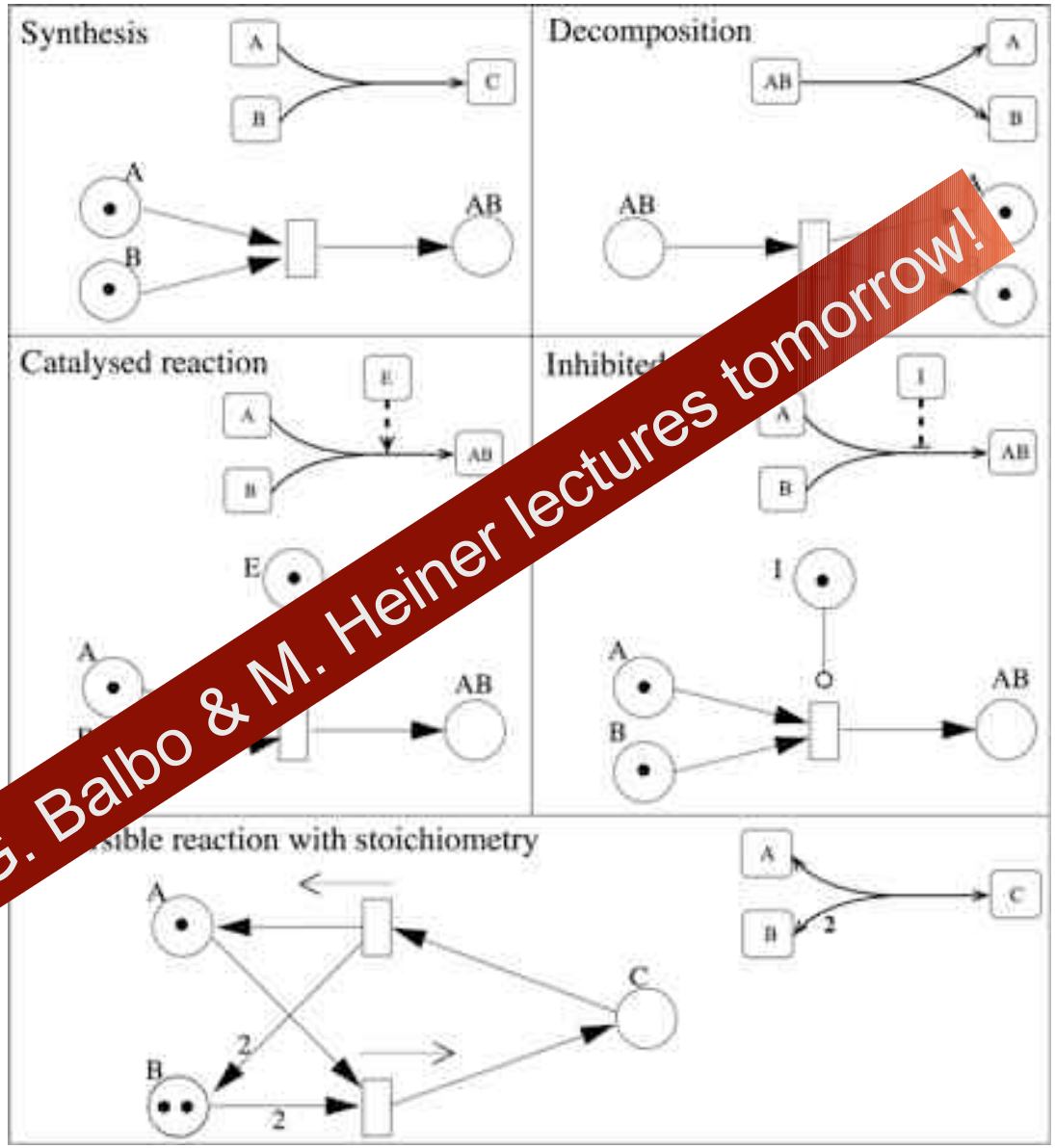

**places:** reactants, products, enzymes...
**transitions:** reactions, catalysis ...
**weighted arcs:** stoichiometry



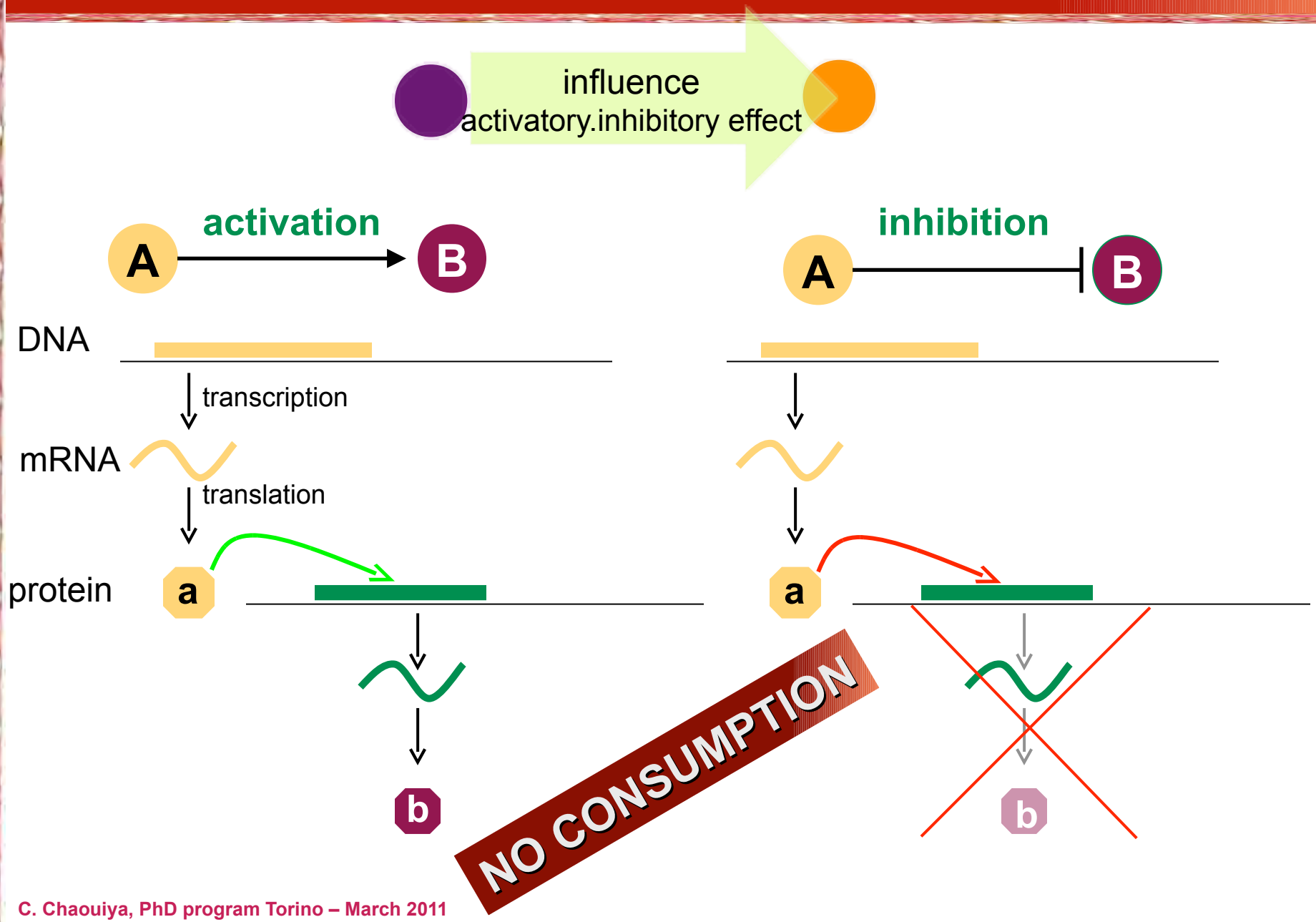
# PN modelling of reaction networks

transformation
chemical reaction




G. Balbo & M. Heiner lectures tomorrow!

# Genetic regulatory networks, a schematic view



# PN modelling of *logical* regulatory networks

## Multi-valued Regulatory Petri Nets

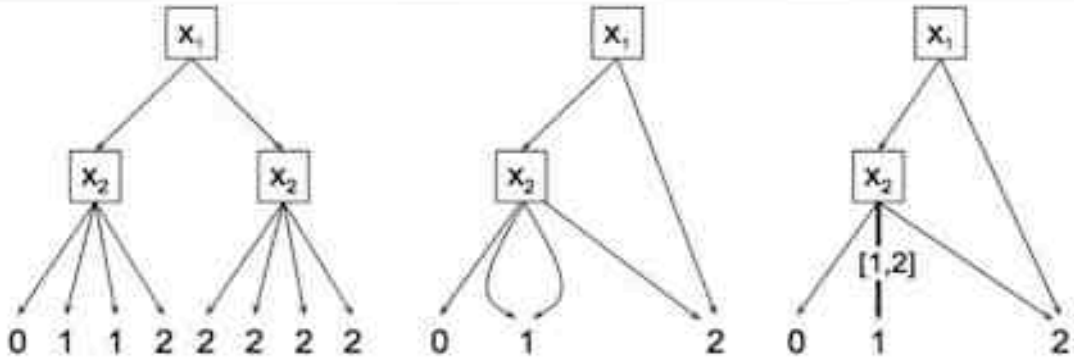

$Reg(g_1)=Reg(g_3)=\{g_1,g_2\}$, $Reg(g_2)=\emptyset$

$Max_1 = 1$, $m_{1,1} = m_{1,3} = 1$, $\theta_{1,1,1} = \theta_{1,3,1} = 1$

$Max_2 = 3$, $m_{2,1} = 1$, $m_{2,3} = 2$, $\theta_{2,1,1} = 2$, $\theta_{2,3,1} = 1$, $\theta_{2,3,2} = 3$

$Max_3 = 2$

| $x_1$ | $x_2$ | Interactions acting on $g_3$ | | $K_3$ |
|---|---|---|---|---|
| 0 | 0 | None | | 0 |
| 0 | 1 | From $g_2$ at level 1 or 2 | (2,3,1) | 1 |
| 0 | 2 | | | |
| 0 | 3 | From $g_2$ at level 3 | (2,3,2) | 2 |
| 1 | 0 | From $g_1$ | (1,3,1) | 2 |
| 1 | 1 | From $g_1$ **and** from $g_2$ at level 1 or 2 | (1,3,1) **and** (2,3,1) | 2 |
| 1 | 2 | | | |
| 1 | 3 | From $g_1$ **and** from $g_2$ at level 3 | (1,3,1) **and** (2,3,2) | 2 |



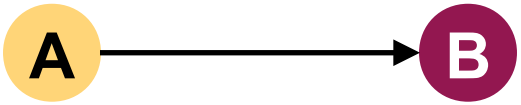
# PN modelling of *logical* regulatory networks

## Multi-valued Regulatory Petri Nets

Genetic regulatory networks described in terms of logical models

- ➔ two complementary places for each gene
- ➔ two transitions for each logical parameter (effect of interactions on a given gene)

**Example**

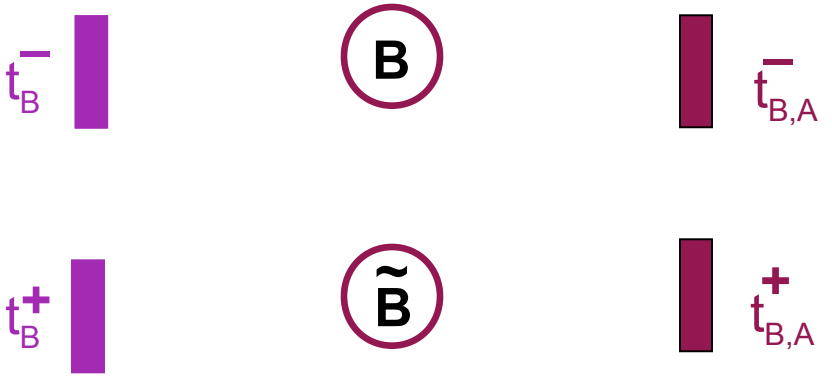



$max_A=1$ $max_B=3$

$K_B(A)=2$ $K_B(\ \ )=1$

$M(A)+M(\tilde{A})=1$

$M(B)+M(\tilde{B})=3$

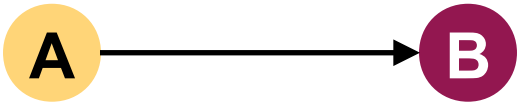
# PN modelling of *logical* regulatory networks

## Multi-valued Regulatory Petri Nets

Genetic regulatory networks described in terms of logical models

- two complementary places for each gene
- two transitions for each logical parameter (effect of interactions on a given gene)

**Example**

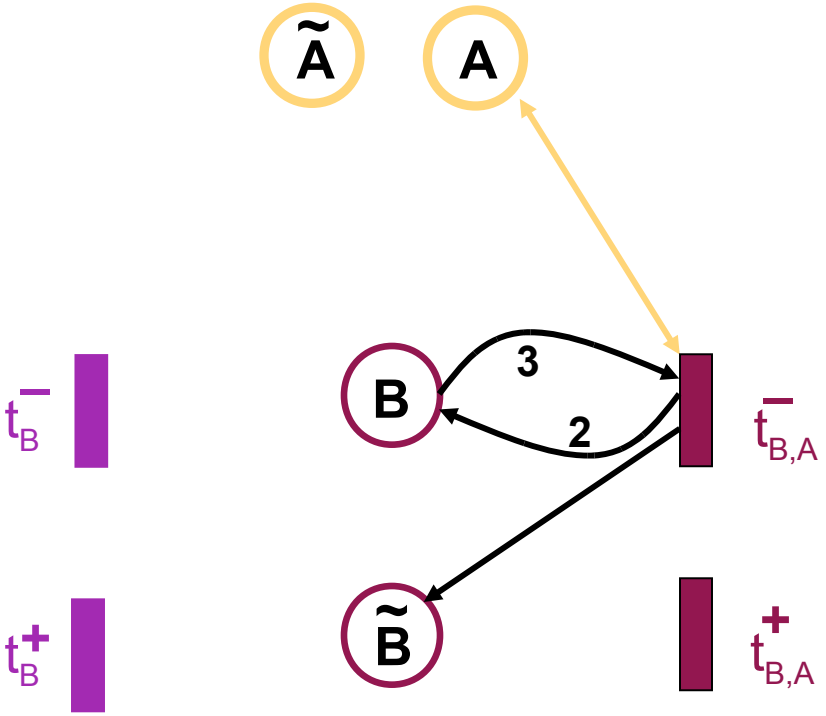



$max_A=1$ $max_B=3$

$K_B(A)=2$ $K_B(\ \ )=1$

$M(A)+M(\tilde{A})=1$

$M(B)+M(\tilde{B})=3$



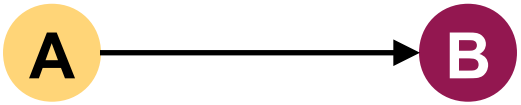
# PN modelling of *logical* regulatory networks

## Multi-valued Regulatory Petri Nets

Genetic regulatory networks described in terms of logical models

- two complementary places for each gene
- two transitions for each logical parameter (effect of interactions on a given gene)

**Example**

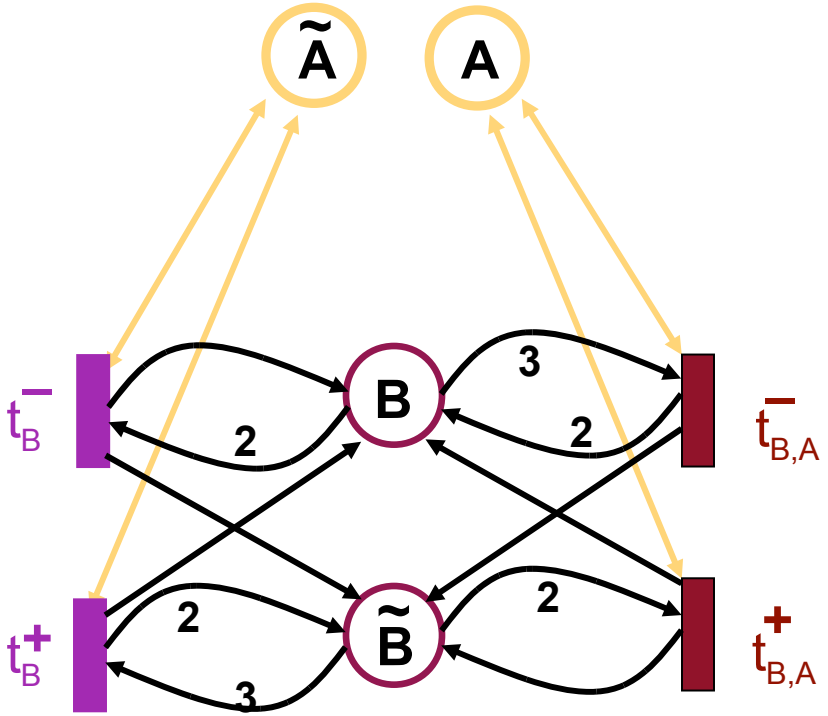



$max_A=1$ $max_B=3$

$K_B(A)=2$ $K_B(\ \ )=1$

$M(A)+M(\tilde{A})=1$

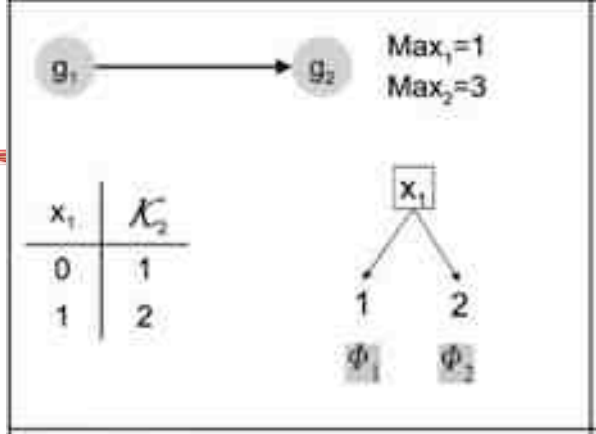
$M(B)+M(\tilde{B})=3$

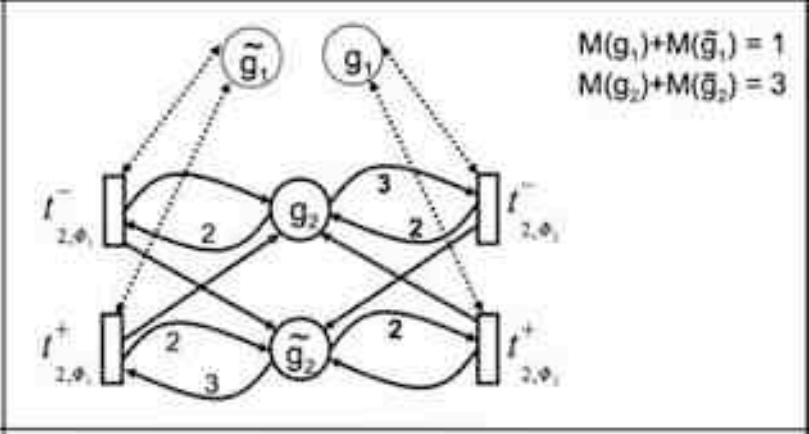


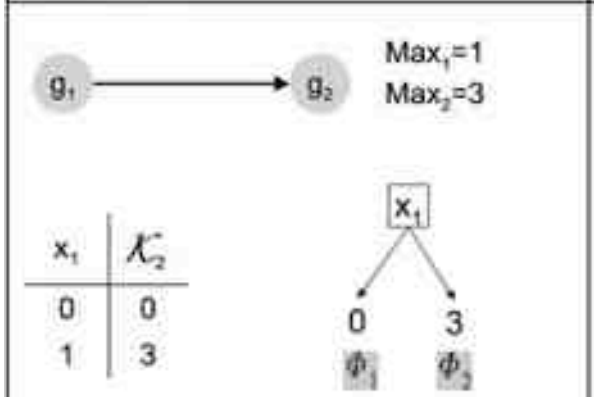
## Definition

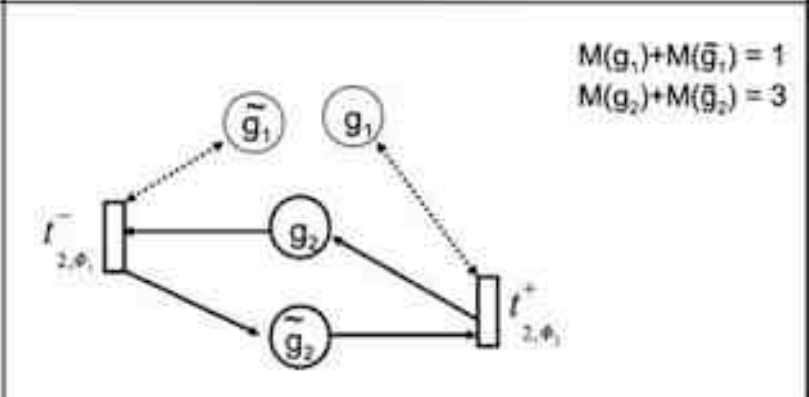
Given a LRG $\mathcal{R} = (\mathcal{G}, \mathcal{M}ax, \mathcal{E}, \Theta, \mathcal{K})$, the corresponding *Multi-valued Regulatory Petri Net* (MRPN) is defined by:

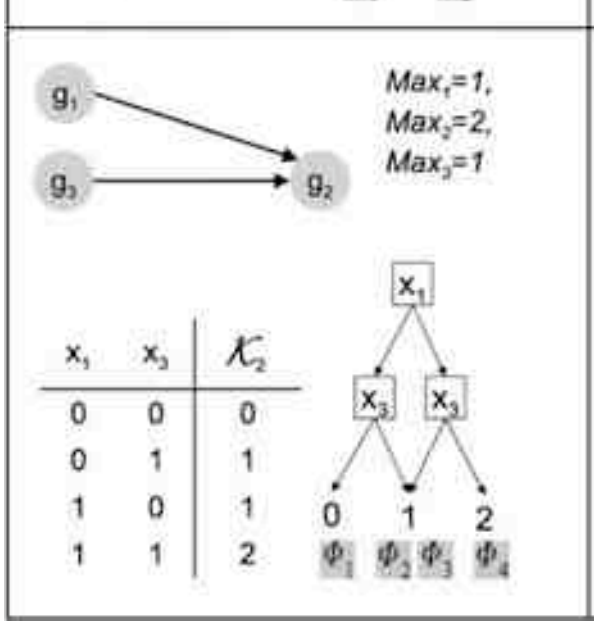
- $\forall g_i \in \mathcal{G}$, two places $g_i$ and $\widetilde{g_i}$ s.t $\forall M$, $M(g_i) + M(\widetilde{g_i}) = Max_i$.
- $\forall g_i \in \mathcal{G}$, for each path $\Phi$ of the MDD $\mathcal{K}_i$, at most two transitions: $t^+_{i,\Phi}$ and $t^-_{i,\Phi}$. Recall that $\Phi$ defines assignment intervals of the levels of $g_j$ in $Reg(i)$: $x_j \in [\phi_j, \phi'_j]$, where $\phi_j, \phi'_j \in [0, Max_j]$ and $\phi_j \leq \phi'_j$.
- Transitions $t^+_{i,\Phi}$ and $t^-_{i,\Phi}$ are connected to:
  place $g_j$, $j \in Reg(i)$, with a test arc weighted $\phi_j$,
  place $\widetilde{g_j}$, $j \in Reg(i)$, with a test arc weighted $Max_j - \phi'_j$.
- Transition $t^+_{i,\Phi}$ is further connected to:
  place $g_i$, with an outgoing arc (increasing the level of $g_i$),
  place $\widetilde{g_i}$, with an incoming arc weighted $Max_i - v_\Phi + 1$ and an outgoing arc weighted $Max_i - v_\Phi$.
- Symmetrically, transition $t^-_{i,\Phi}$ is further connected to:
  place $\widetilde{g_i}$, with an outgoing arc (decreasing the level of $g_i$),
  place $g_i$, with an incoming arc weighted $v_\Phi + 1$ and an outgoing arc

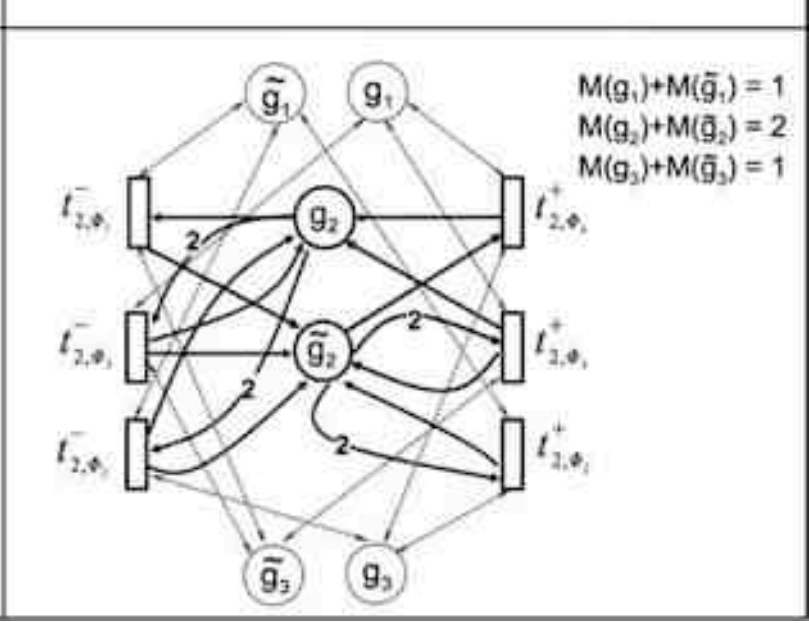

$g_1$
$g_2$
$Max_1=1$
$Max_2=3$
$x_1$ | $\mathcal{K}_2$
0 | 1
1 | 2
$x_1$
1
2
$\Phi_1$
$\Phi_2$


$M(g_1)+M(\bar{g}_1) = 1$
$M(g_2)+M(\bar{g}_2) = 3$
$\bar{g}_1$
$g_1$
$g_2$
$\bar{g}_2$
2
3
2
2
2
3


$g_1$
$g_2$
$Max_1=1$
$Max_2=3$
$x_1$ | $\mathcal{K}_2$
0 | 0
1 | 3
$x_1$
0
3
$\Phi_1$
$\Phi_2$


$M(g_1)+M(\bar{g}_1) = 1$
$M(g_2)+M(\bar{g}_2) = 3$
$\bar{g}_1$
$g_1$
$g_2$
$\bar{g}_2$


$g_1$
$g_3$
$g_2$
$Max_1=1,$
$Max_2=2,$
$Max_3=1$
$x_1$ | $x_3$ | $\mathcal{K}_2$
0 | 0 | 0
0 | 1 | 1
1 | 0 | 1
1 | 1 | 2
$x_1$
$x_3$
$x_3$
0
1
2
$\Phi_1$
$\Phi_2$
$\Phi_3$
$\Phi_4$


$M(g_1)+M(\bar{g}_1) = 1$
$M(g_2)+M(\bar{g}_2) = 2$
$M(g_3)+M(\bar{g}_3) = 1$
$\bar{g}_1$
$g_1$
$g_2$
$\bar{g}_2$
$\bar{g}_3$
$g_3$
2
2
2
2

**Property**

*In the state transition graph* $(\mathcal{S}, \mathcal{T})$ *of a LRG* $\mathcal{R} = (\mathcal{G}, \mathcal{M}ax, \mathcal{E}, \Theta, \mathcal{K})$, *there exists a transition between two states* $x$ *and* $x'$ *iff there exists an enabled transition* $t$ *in the associated MRPN such that* $M[t\rangle M'$ *(*$t$ *is enabled by the marking* $M$ *and its firing leads to the new marking* $M'$*) with, for all* $k = 1, \ldots n$*:*

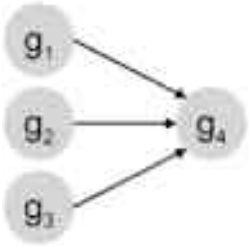
$$M(g_k) = x_k \qquad M(\widetilde{g_k}) = Max_k - x_k\,,$$
$$M'(g_k) = x'_k \qquad M'(\widetilde{g_k}) = Max_k - x'_k\,.$$

# PN modelling of *logical* regulatory networks

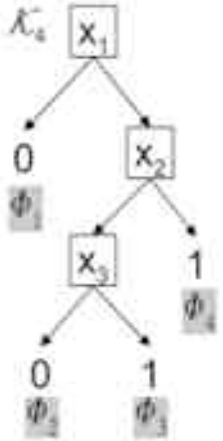
## Multi-valued Regulatory Petri Nets

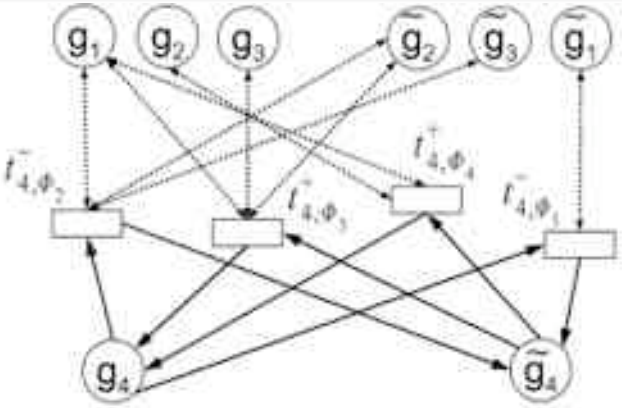
### Property

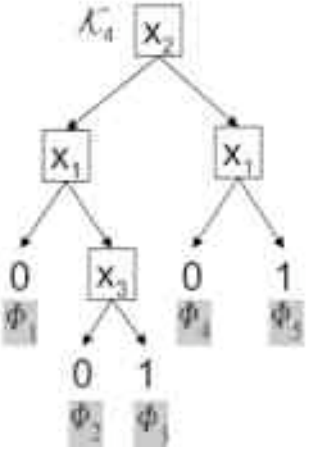
*Given a LRG $\mathcal{R}$, two different orderings of the regulatory nodes can lead to different MRPNs, which have the same dynamical behaviour (i.e. their marking graphs are isomorph for a given initial state x).*

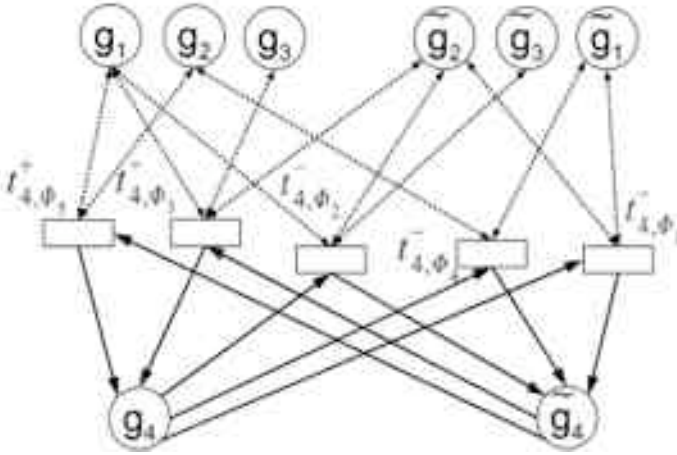



| $x_1$ | $x_2$ | $x_3$ | $\mathcal{K}_4$ |
|---|---|---|---|
| 0 | 0 | 0 | 0 |
| 0 | 0 | 1 | 0 |
| 0 | 1 | 0 | 0 |
| 0 | 1 | 1 | 0 |
| 1 | 0 | 0 | 0 |
| 1 | 0 | 1 | 1 |
| 1 | 1 | 0 | 1 |
| 1 | 1 | 1 | 1 |



# PN modelling of *logical* regulatory networks

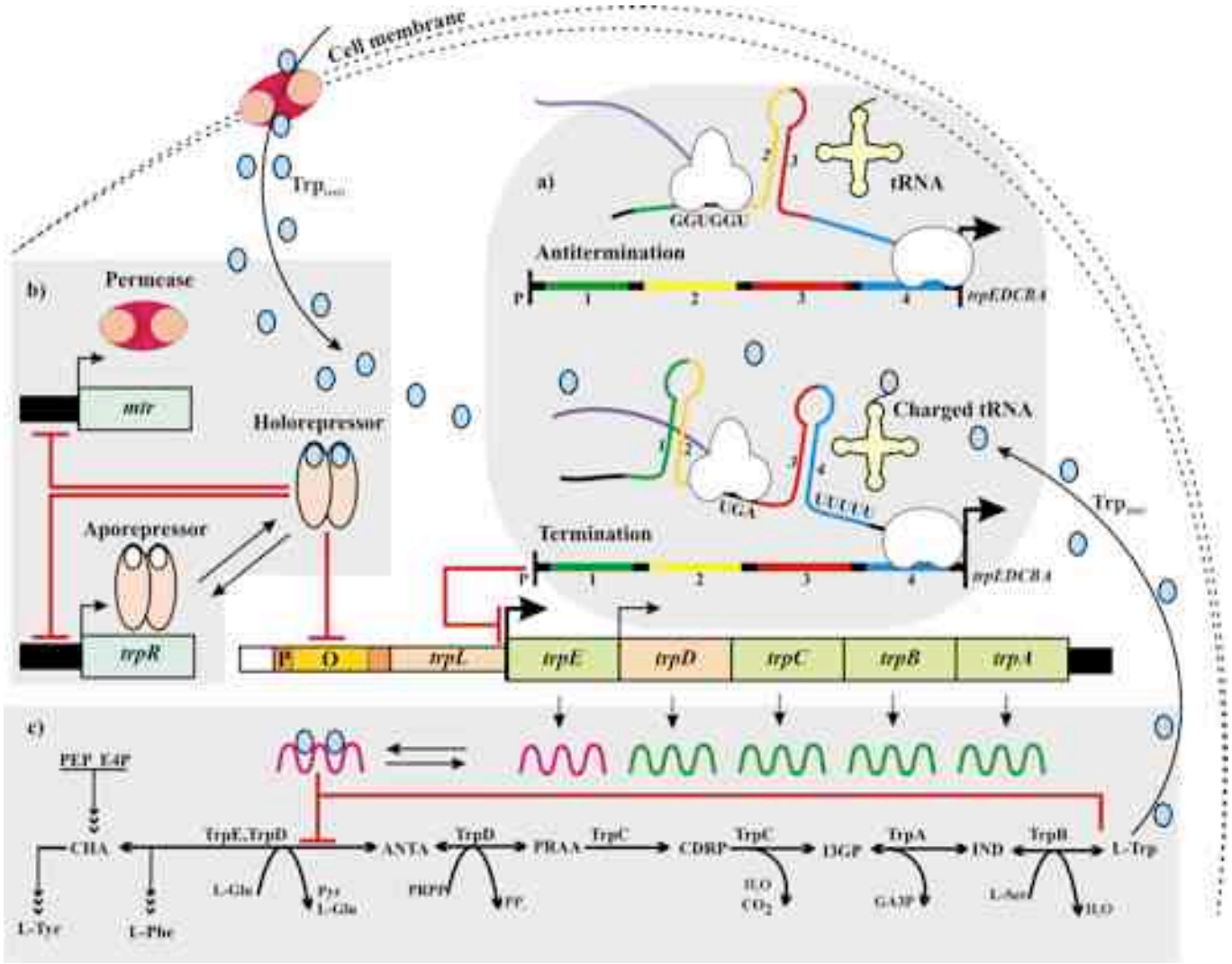
## Multi-valued Regulatory Petri Nets

- Export of logical models in several PN formats
- Use of exiting tools for PN analysis
- Consideration of PN extensions
  - Coloured PN for compositional modelling
  - Timed or stochastic PN to sort out relevant dynamics

- Integrated modelling of metabolic pathways and their regulation

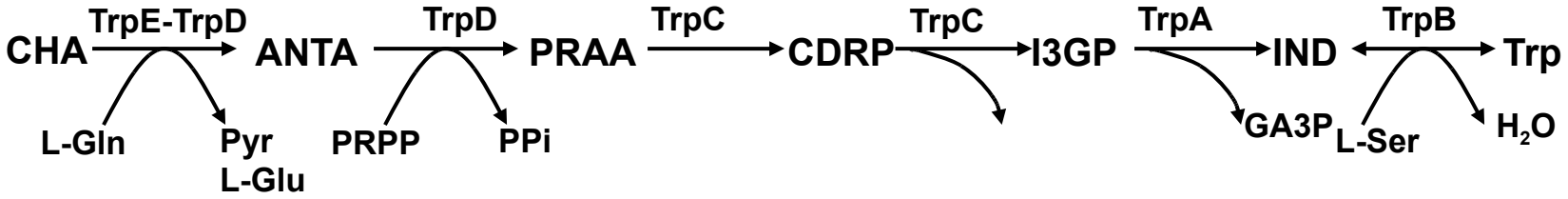
# Integrated modelling of regulated metabolic pathways

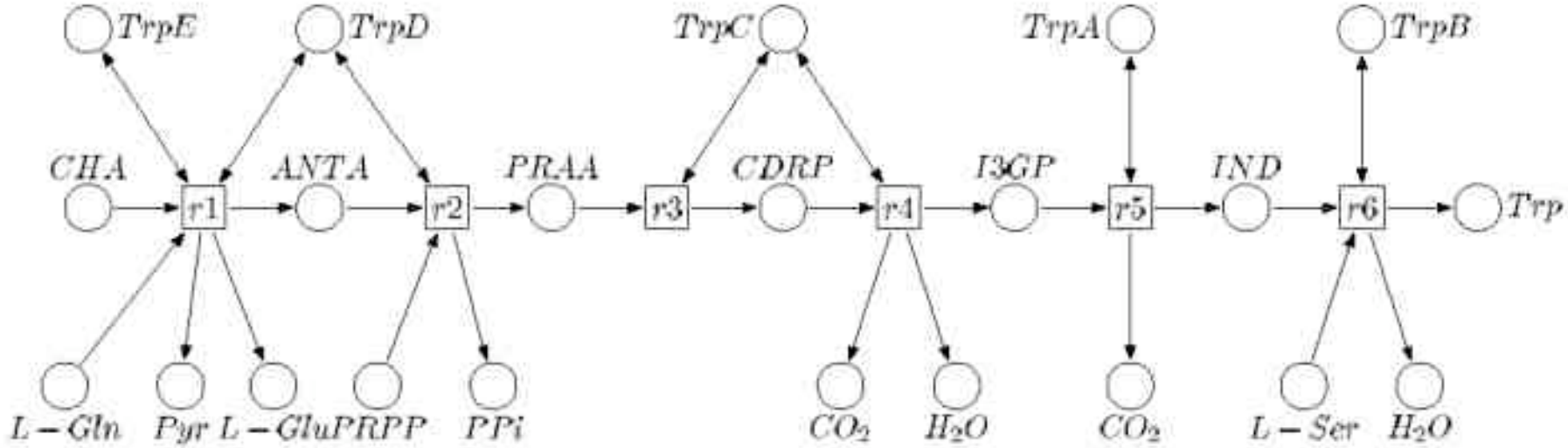
## The Tryptophan biosynthesis in *E. coli*



# Integrated modelling of regulated metabolic pathways

## The Tryptophan biosynthesis in *E. coli*

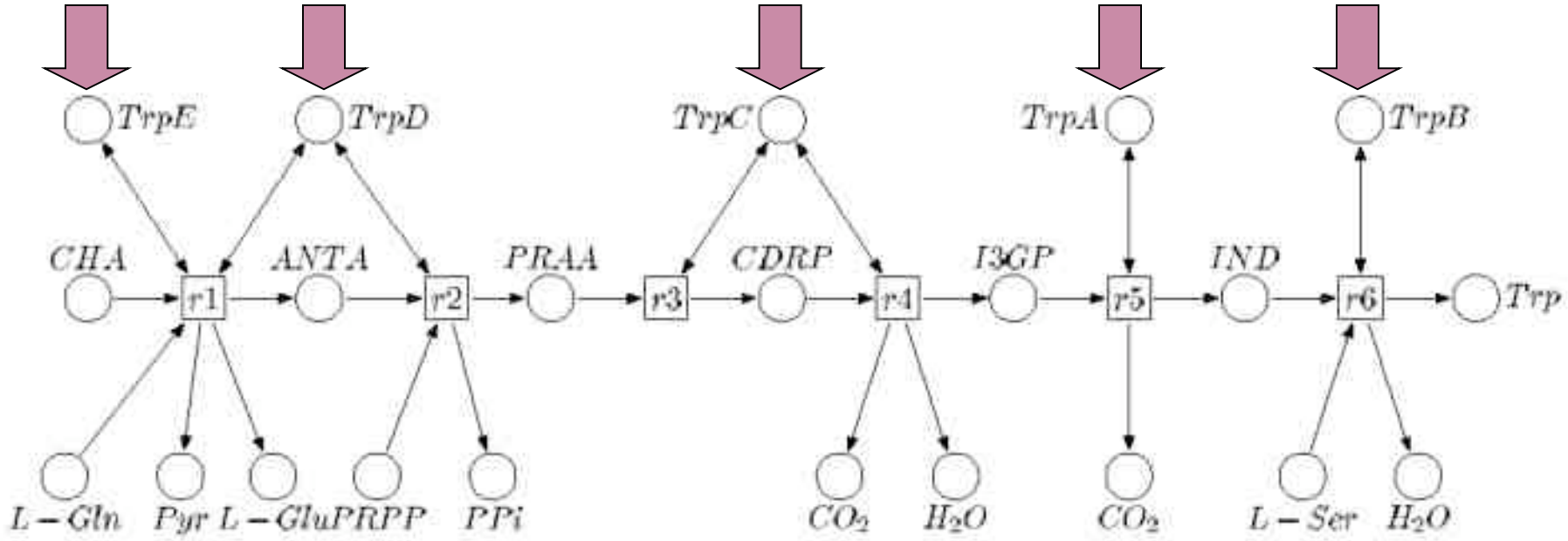


# Integrated modelling of regulated metabolic pathways

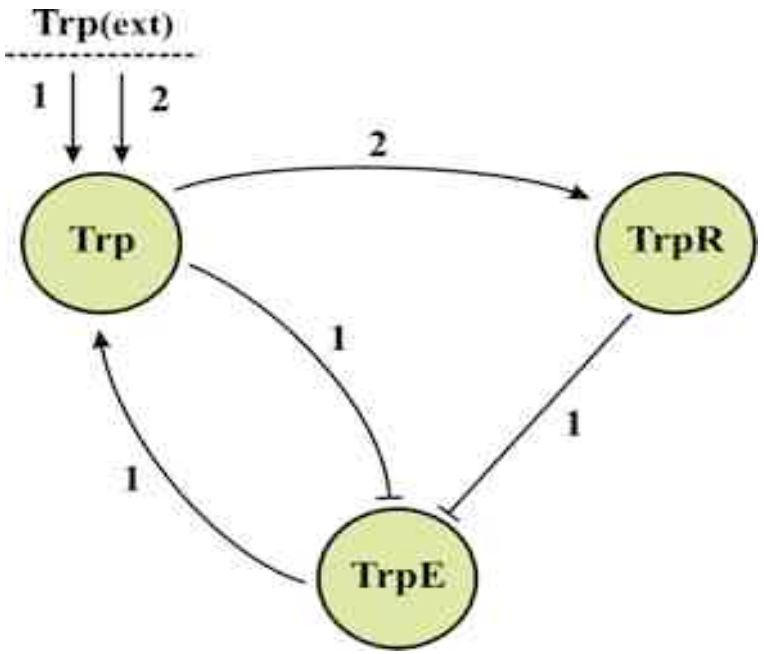
## The Tryptophan biosynthesis in *E. coli*

**REGULATION ???**

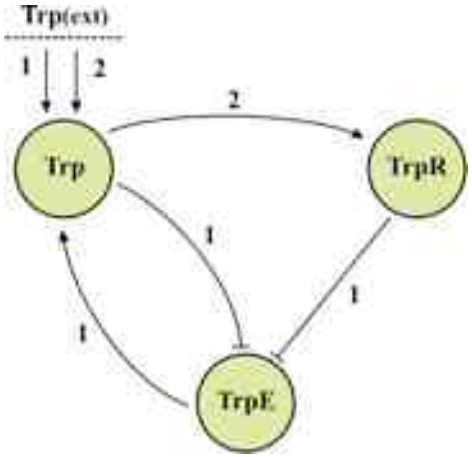


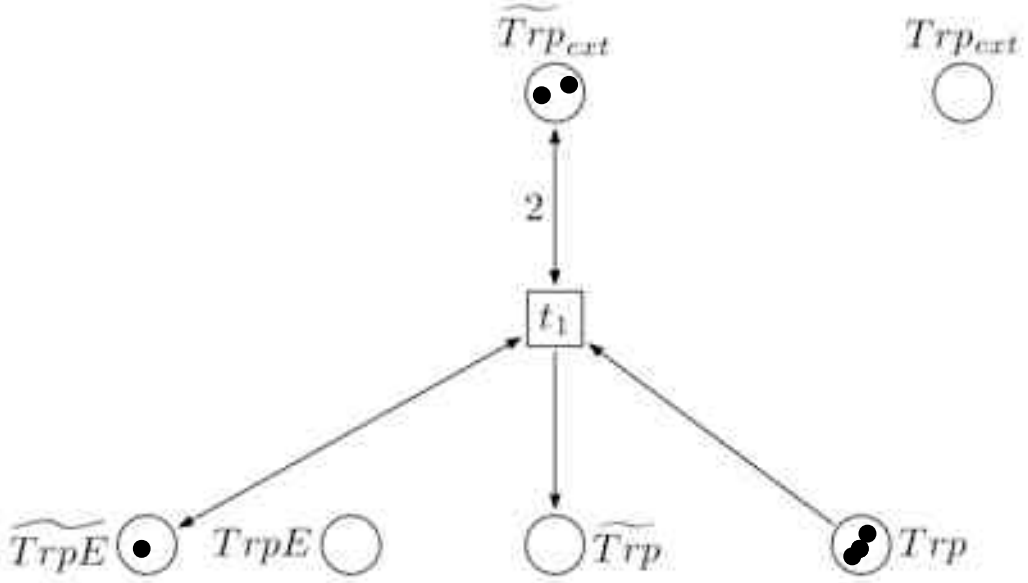
# A logical model of the regulation of the Tryptophan biosynthesis

| Context (input genes) | Parameter value |
|---|---|
| *TrpE* | |
| $TrpR = 0$ and $Trp = 0$ | 1 |
| $TrpR = 1$ and $Trp = 0$ | 0 |
| $TrpR = 0$ and $Trp \geq 1$ | 0 |
| $TrpR = 1$ and $Trp \geq 1$ | 0 |
| *TrpR* | |
| $Trp \leq 1$ | 0 |
| $Trp = 2$ | 1 |
| *Trp* | |
| $Trp_{ext} = 0$ and $TrpE = 0$ | 0 |
| $Trp_{ext} = 0$ and $TrpE = 1$ | 1 |
| $Trp_{ext} = 1$ and $TrpE = 0$ | 1 |
| $Trp_{ext} = 1$ and $TrpE = 1$ | 1 |
| $Trp_{ext} = 2$ and $TrpE = 0$ | 2 |
| $Trp_{ext} = 2$ and $TrpE = 1$ | 2 |



A logical model of the regulation of the Tryptophan biosynthesis
PN representation

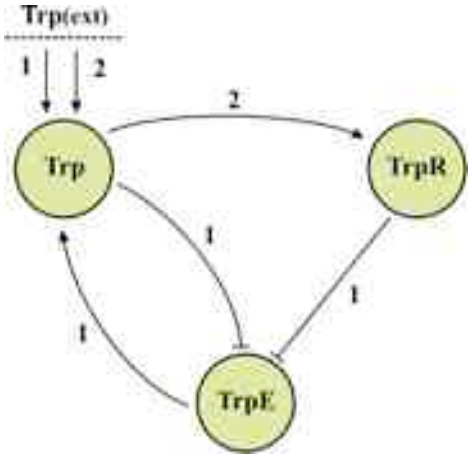


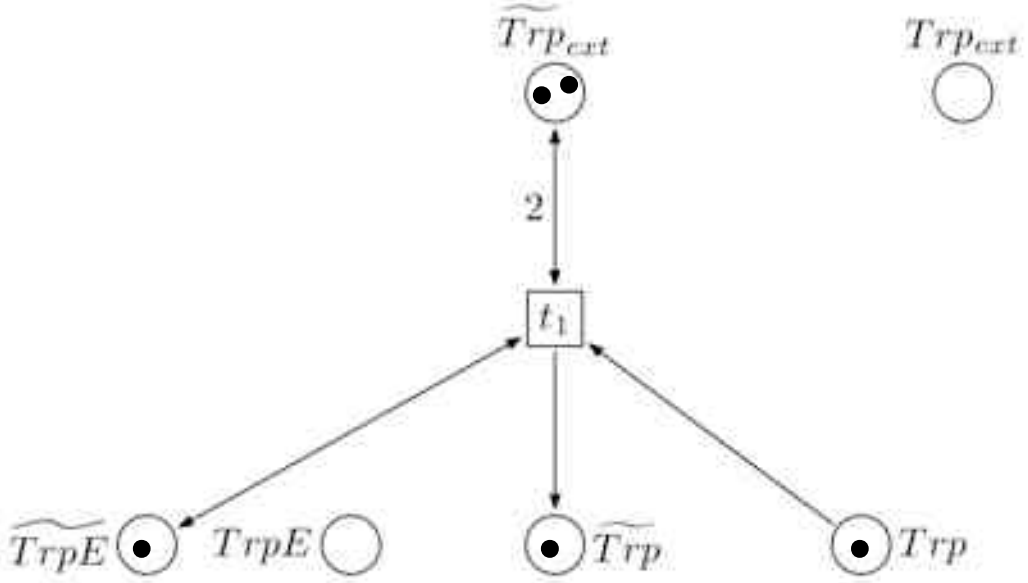




when $Trp_{ext}$=0 and TrpE=0, Trp 0

gene or regulatory product ... → 2 complementary places
combination of incoming interactions →1 or 2 transitions

A logical model of the regulation of the Tryptophan biosynthesis
PN representation

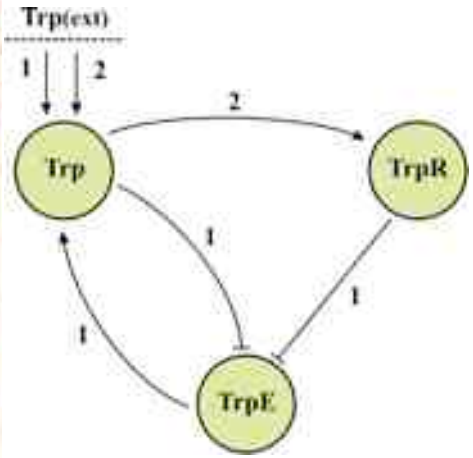


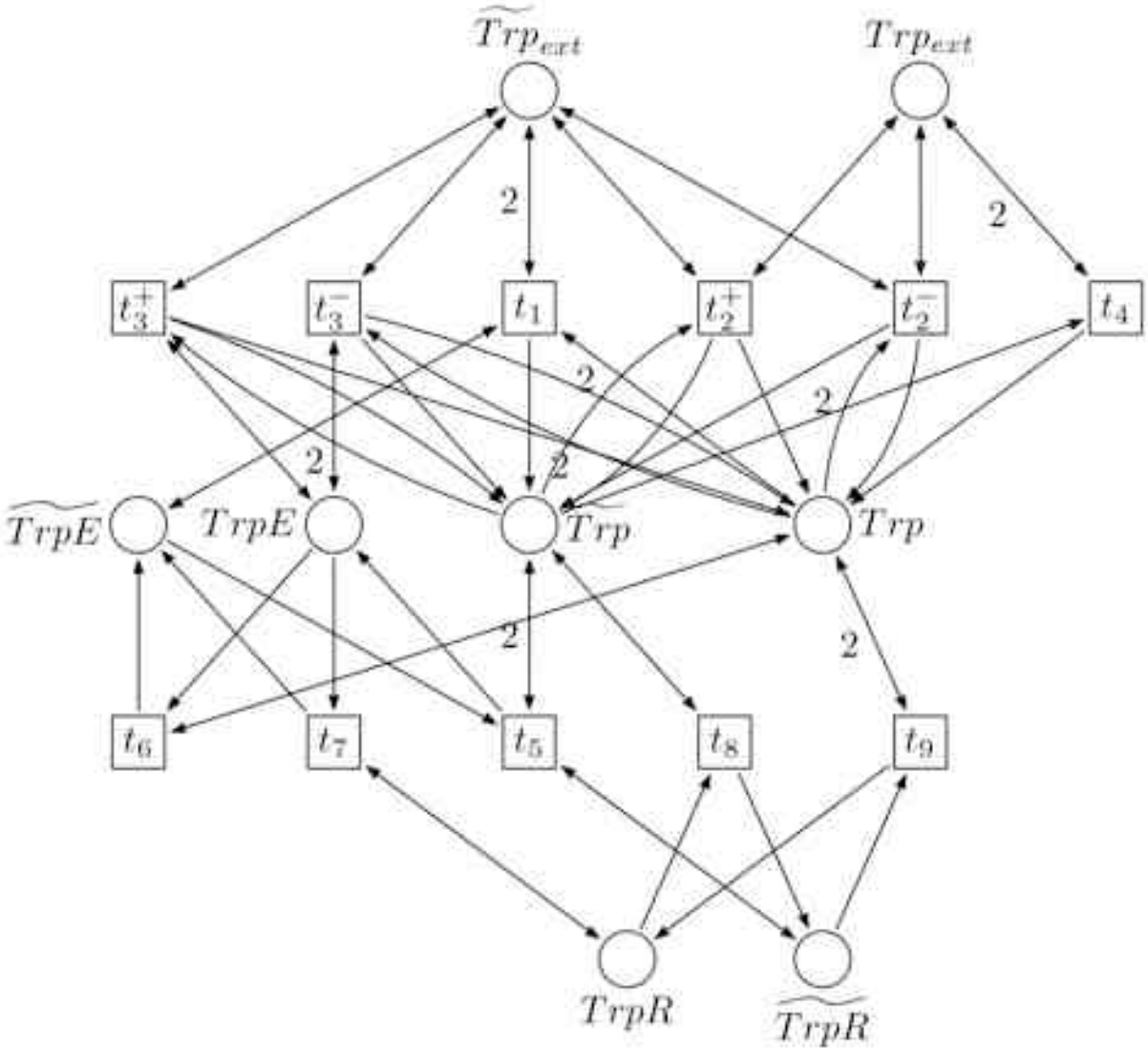

when $Trp_{ext}$=0 and TrpE=0, Trp 0

gene or regulatory product ... → 2 complementary places
combination of incoming interactions →1 or 2 transitions

# A logical model of the regulation of the Tryptophan biosynthesis PN representation



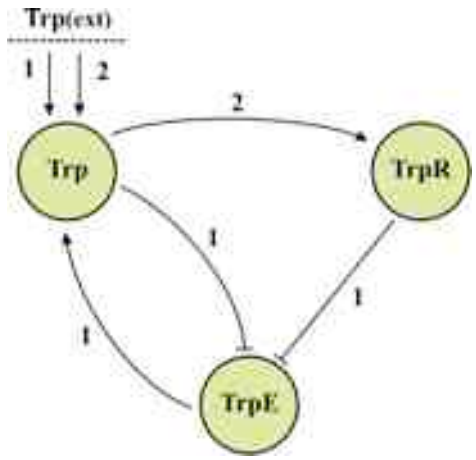

| Context (input genes) | Parameter value | Corresponding PN transitions |
|---|---|---|
| *TrpE* | | |
| $TrpR = 0$ and $Trp = 0$ | 1 | $t_1$ |
| $TrpR = 1$ and $Trp = 0$ | 0 | $t_6, t_7$ |
| $TrpR = 0$ and $Trp \geq 1$ | 0 | |
| $TrpR = 1$ and $Trp \geq 1$ | 0 | |
| *TrpR* | | |
| $Trp \leq 1$ | 0 | $t_8$ |
| $Trp = 2$ | 1 | $t_9$ |
| *Trp* | | |
| $Trp_{ext} = 0$ and $TrpE = 0$ | 0 | $t_5$ |
| $Trp_{ext} = 0$ and $TrpE = 1$ | 1 | $t_2^+, t_3^+$ |
| $Trp_{ext} = 1$ and $TrpE = 0$ | 1 | |
| $Trp_{ext} = 1$ and $TrpE = 1$ | 1 | |
| $Trp_{ext} = 2$ and $TrpE = 0$ | 2 | $t_4$ |
| $Trp_{ext} = 2$ and $TrpE = 1$ | 2 | |

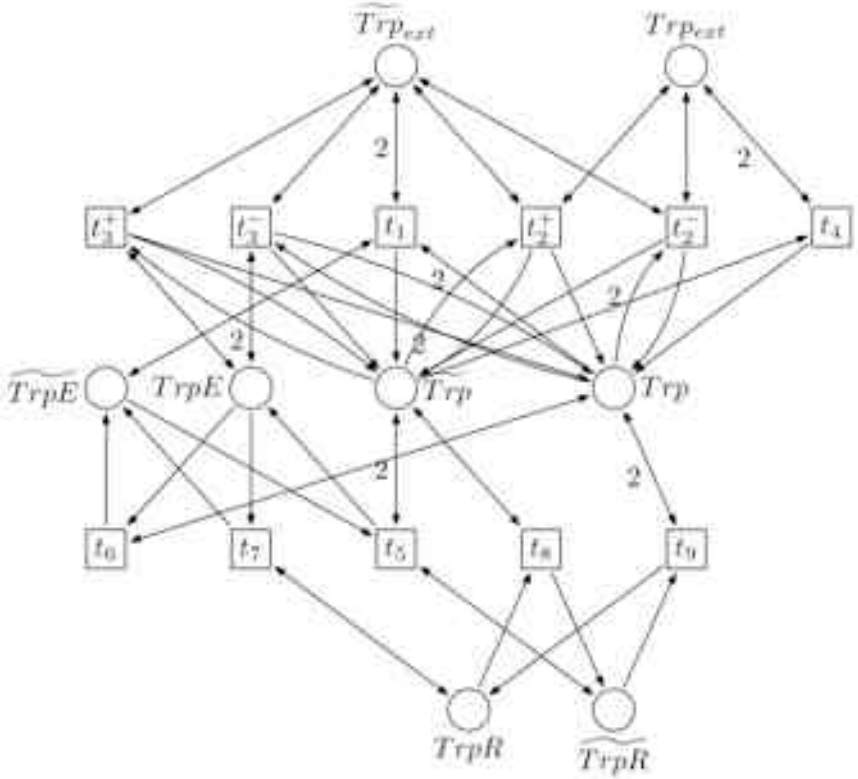


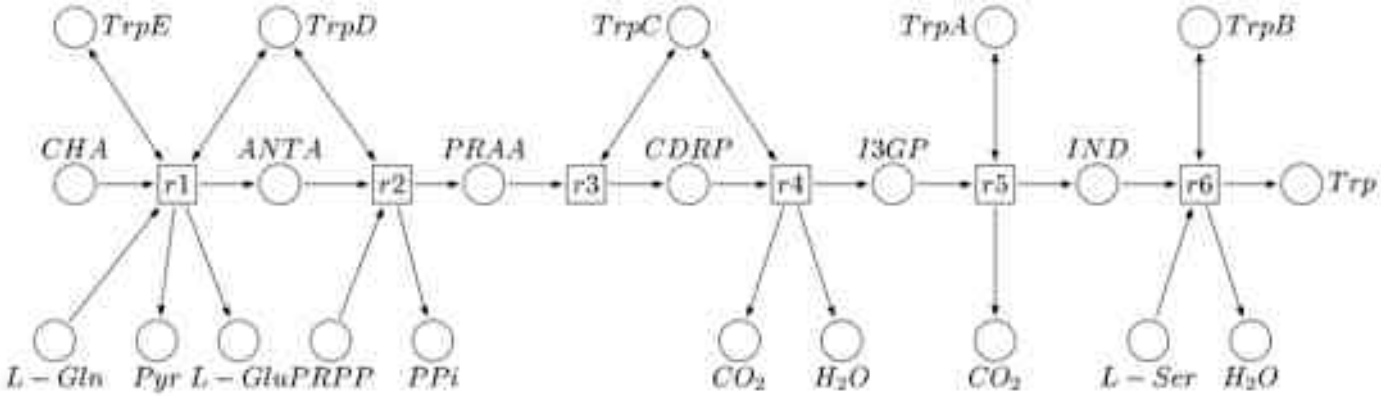
# Integrated modelling of regulated metabolic pathways

## The Tryptophan biosynthesis in *E. coli*

## Integrated PN

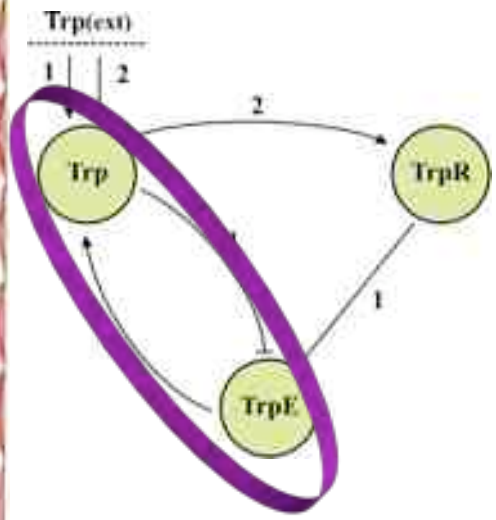


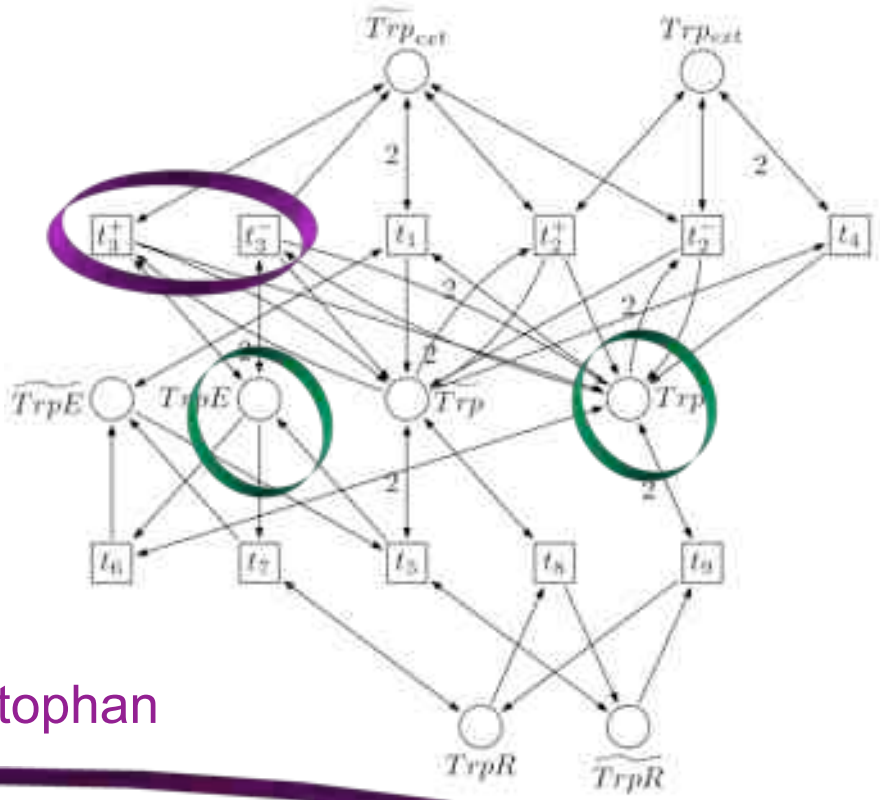



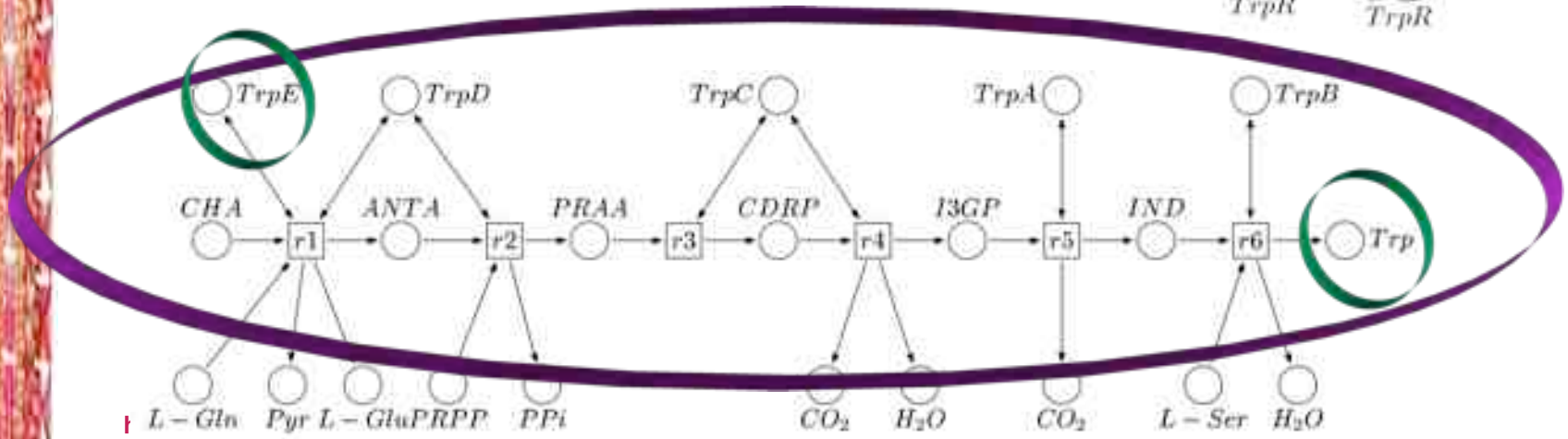
# Integrated modelling of regulated metabolic pathways
## The Tryptophan biosynthesis in *E. coli*

Integrated PN



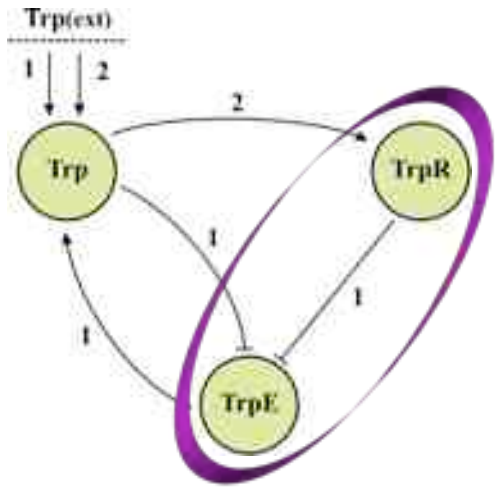



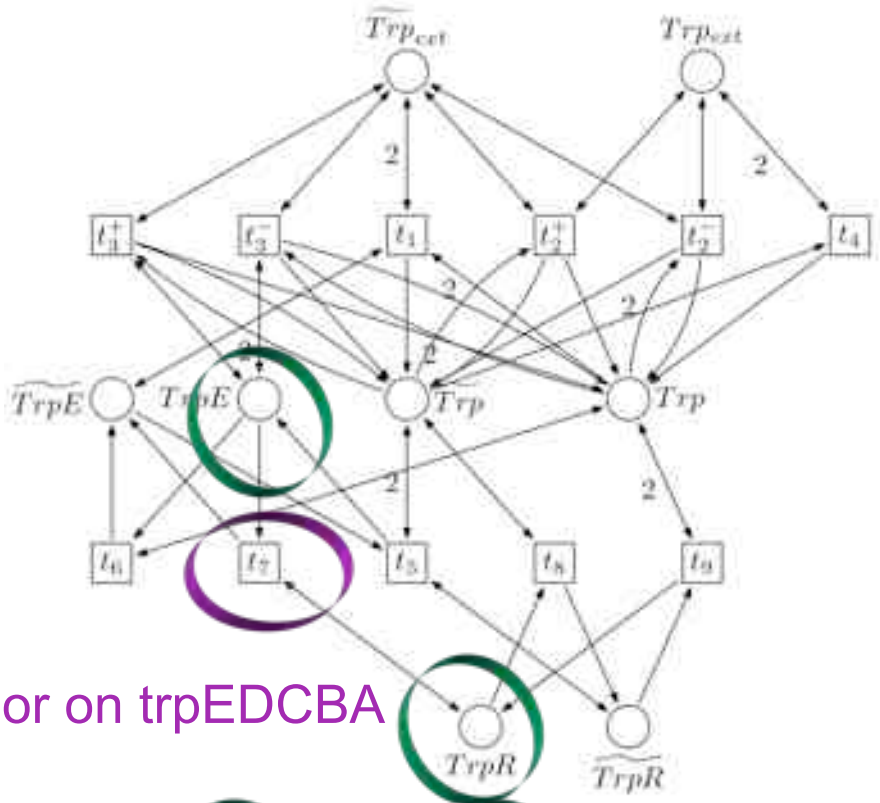

conversion of chorismate into tryptophan



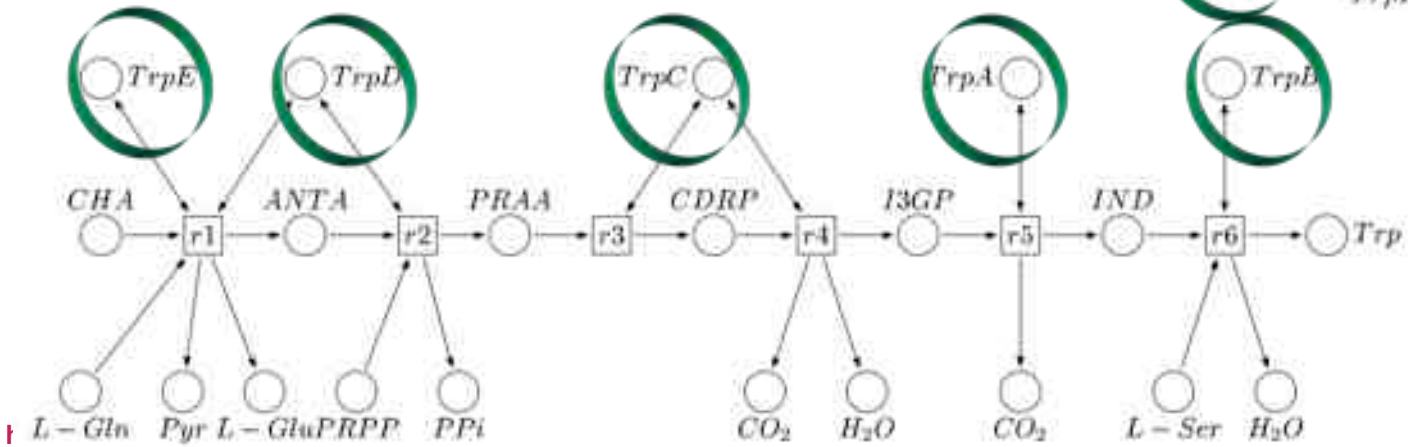
# Integrated modelling of regulated metabolic pathways
## The Tryptophan biosynthesis in *E. coli*

Integrated PN

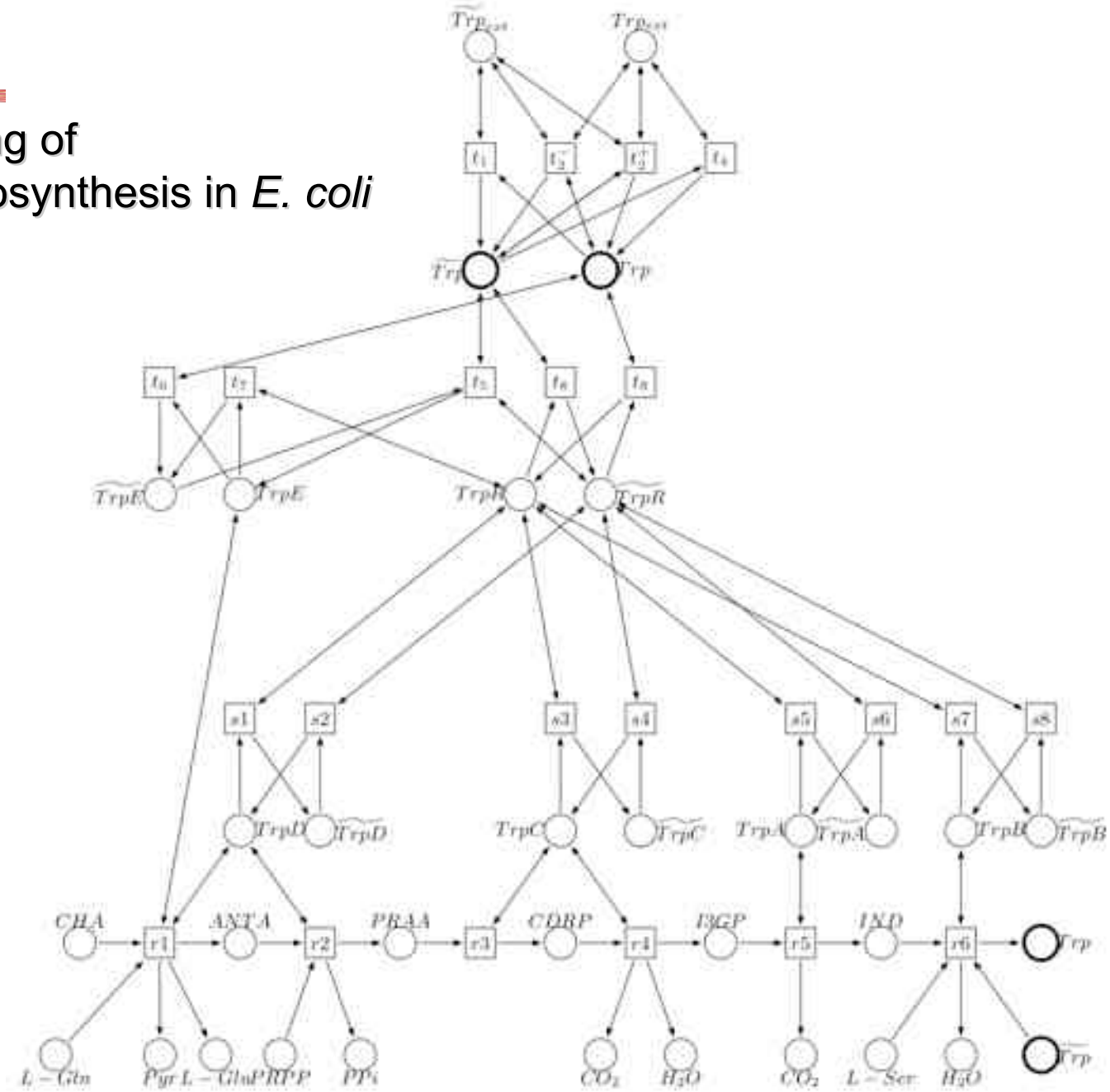



inhibitory effect of the holorepressor on trpEDCBA

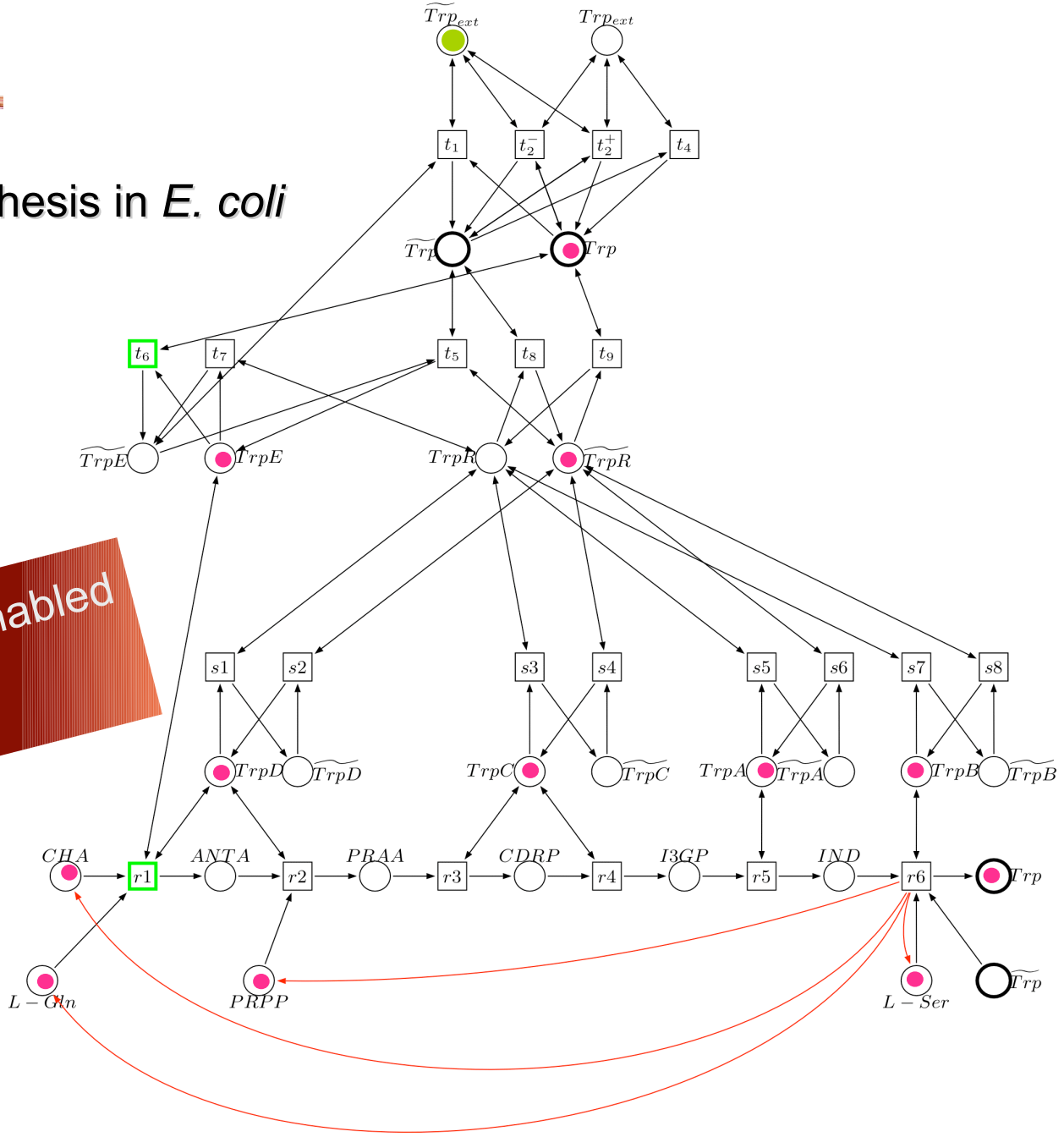


# Integrated modelling of The Tryptophan biosynthesis in *E. coli*



# Integrated modelling of The Tryptophan biosynthesis in *E. coli*

When $t_6$ and $r_1$ are both enabled $t_6$ should fire first



# Integrated modelling of regulated metabolic pathways
## The Tryptophan biosynthesis in *E. coli*

Initial state No external tryptophan, all input compounds present, all enzymes active, no internal tryptophan, no holorepressor

Attractor One cyclic attractor denoting homeostatic levels of internal tryptophan and TrpE activity

| | CHA | ANTA | PRAA | CRDP | I3GP | IND | TrpE | TrpD | TrpC | TrpB | TrpA | Trp | TrpR | $Trp_{ext}$ |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **1** | **0** | **0** | **0** | **0** | **0** | **1** | **1** | **1** | **1** | **1** | **0** | **0** | **0** |
| **r1** | **0** | **1** | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0 |
| **r2** | 0 | **0** | **1** | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0 |
| **r3** | 0 | 0 | **0** | **1** | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0 |
| **t5** **r4** | 0 | 0 | 0 | **0** | **1** | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0 |
| **r5** | 0 | 0 | 0 | 0 | **0** | **1** | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 0 |
| **r6** | **1** | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | **1** | 0 | 0 |
| **t6** | 1 | 0 | 0 | 0 | 0 | 0 | **0** | 1 | 1 | 1 | 1 | 1 | 0 | 0 |
| **t1** | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | **0** | 0 | 0 |

# Integrated modelling of regulated metabolic pathways
## The Tryptophan biosynthesis in *E. coli*

Initial state Low external tryptophan, all input compounds present, all enzymes active, no internal tryptophan, no holorepressor

Attractor A unique reachable dead marking with a moderate level of internal tryptophan ; both repressor and TrpE inactive

| | CHA | ANTA | PRAA | CRDP | I3GP | IND | TrpE | TrpD | TrpC | TrpB | TrpA | Trp | TrpR | $Trp_{ext}$ |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 1 |
| | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Δ | 1 | 0 | 0 | 0 | 0 | 0 | **0** | 1 | 1 | 1 | 1 | **1** | 0 | 1 |

# Integrated modelling of regulated metabolic pathways
## The Tryptophan biosynthesis in *E. coli*

Initial state Low external tryptophan, all input compounds present, all enzymes active, no internal tryptophan, no holorepressor

Attractor A unique reachable dead marking with a moderate level of internal tryptophan ; both repressor and TrpE inactive

```
Computation of the reachability graph
.............................Omit boundedness test? Y/N N
States generated:          624
........Write the state numbers of the dead states? Y/N Y
The net has dead reachable states.
The net is not live.
The net is not reversible (resetable).
The net is bounded.
The net has dead transitions at the initial marking.
ORD HOM NBM PUR CSV SCF CON SC  FtO tFO FpO pFO MG  SM  FC  EFC ES
 N   N   N   N   N   N   Y   Y   N   N   N   N   N   N   N   N   N
DTP CPI CTI  B  SB  REV DSt BSt DTr DCF  L   LV L&S
 ?   ?   ?  _Y   ?   N   Y   ?   Y   ?   N   ?   N

Reachable scc's: 316,
Component nr. 316: 22, term.

State nr.   22
P.nr:  0  1  2  3  4  5  6  7  8  9 10 11 12 13 14 15 16 17 18 19 20 21 22
     : 23 24
toks:  1  0  0  0  0  0  1  1  1  1  1  0  1  1  1  1  1  1  1  0  1  0  0
     :  0  0
```

Analysis performed using INA

# Integrated modelling of regulated metabolic pathways
## The Tryptophan biosynthesis in *E. coli*

Initial state external tryptophan, all input compounds present, all enzymes active, no internal tryptophan, no holorepressor

Attractor Six reachable dead markings with a high level of internal tryptophan, the six enzymes inactive, the repressor active

| CHA | ANTA | PRAA | CRDP | I3GP | IND | TrpE | TrpD | TrpC | TrpB | TrpA | Trp | TrpR | $Trp_{ext}$ |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 2 |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **1** | 0 | 0 | 0 | 0 | 0 | **0** | **0** | **0** | **0** | **0** | **2** | **1** | **2** |
| 0 | **1** | 0 | 0 | 0 | 0 | **0** | **0** | **0** | **0** | **0** | **2** | **1** | **2** |
| 0 | 0 | **1** | 0 | 0 | 0 | **0** | **0** | **0** | **0** | **0** | **2** | **1** | **2** |
| 0 | 0 | 0 | **1** | 0 | 0 | **0** | **0** | **0** | **0** | **0** | **2** | **1** | **2** |
| 0 | 0 | 0 | 0 | **1** | 0 | **0** | **0** | **0** | **0** | **0** | **2** | **1** | **2** |
| 0 | 0 | 0 | 0 | 0 | **1** | **0** | **0** | **0** | **0** | **0** | **2** | **1** | **2** |

# Conclusions & prospects

- A systematic translation of logical regulatory graphs in terms of PN (available in GINsim)
- Use of PN tools (INA, invariant analyses, model checking)
- A generic approach based on the combination of PN representations of metabolic pathways and logical models of regulatory networks
- Use of PN extensions (coloured, stochastic,...)

- Tryptophan biosynthesis pathway
  - a faithfull qualitative reproduction of the main behaviours for different influxes of external tryptophan
  - extension of the model to cover the other regulatory mechanisms

- Automatise this approach?