# An Introduction to $^{13}C$ Metabolic flux analysis

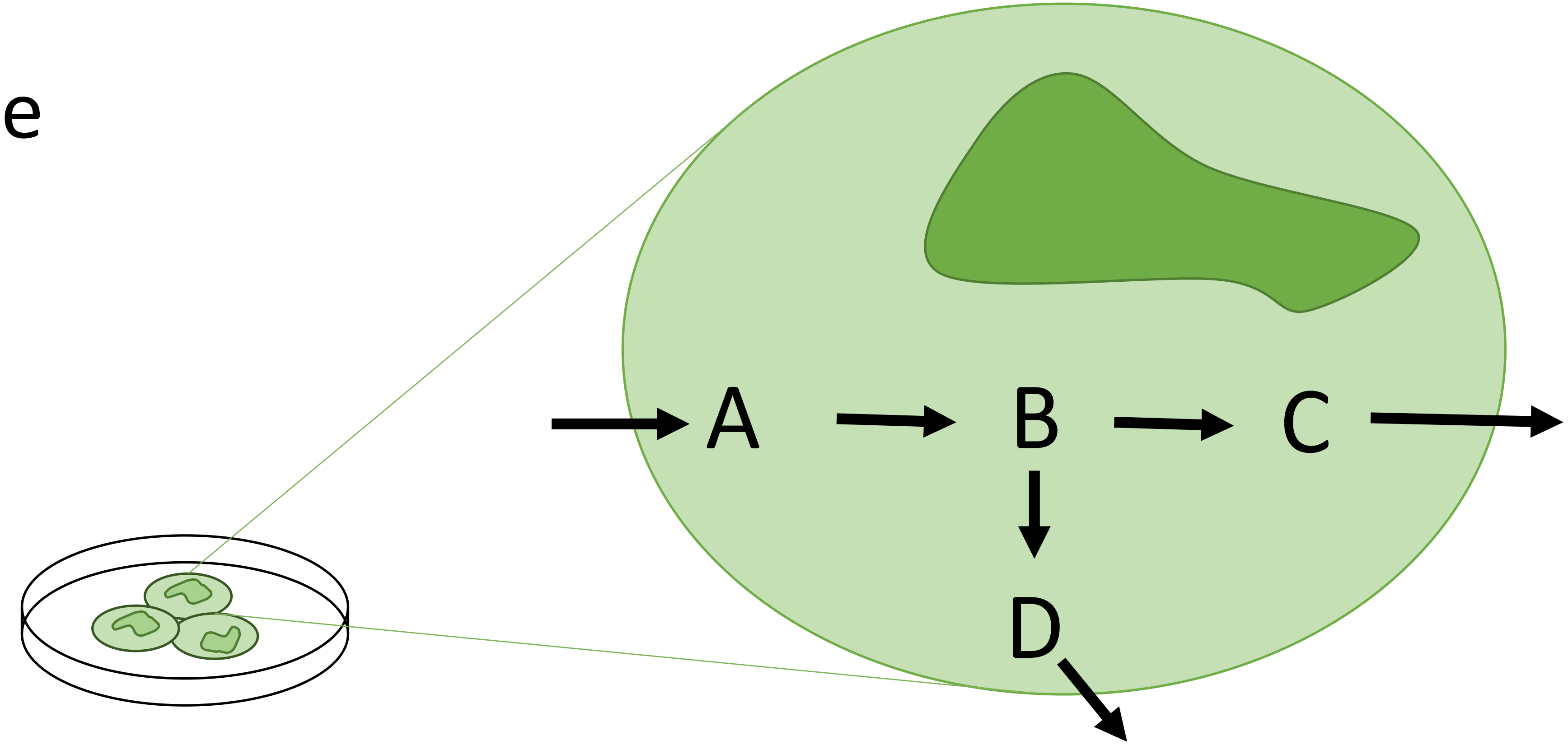
Nicolas Sundqvist

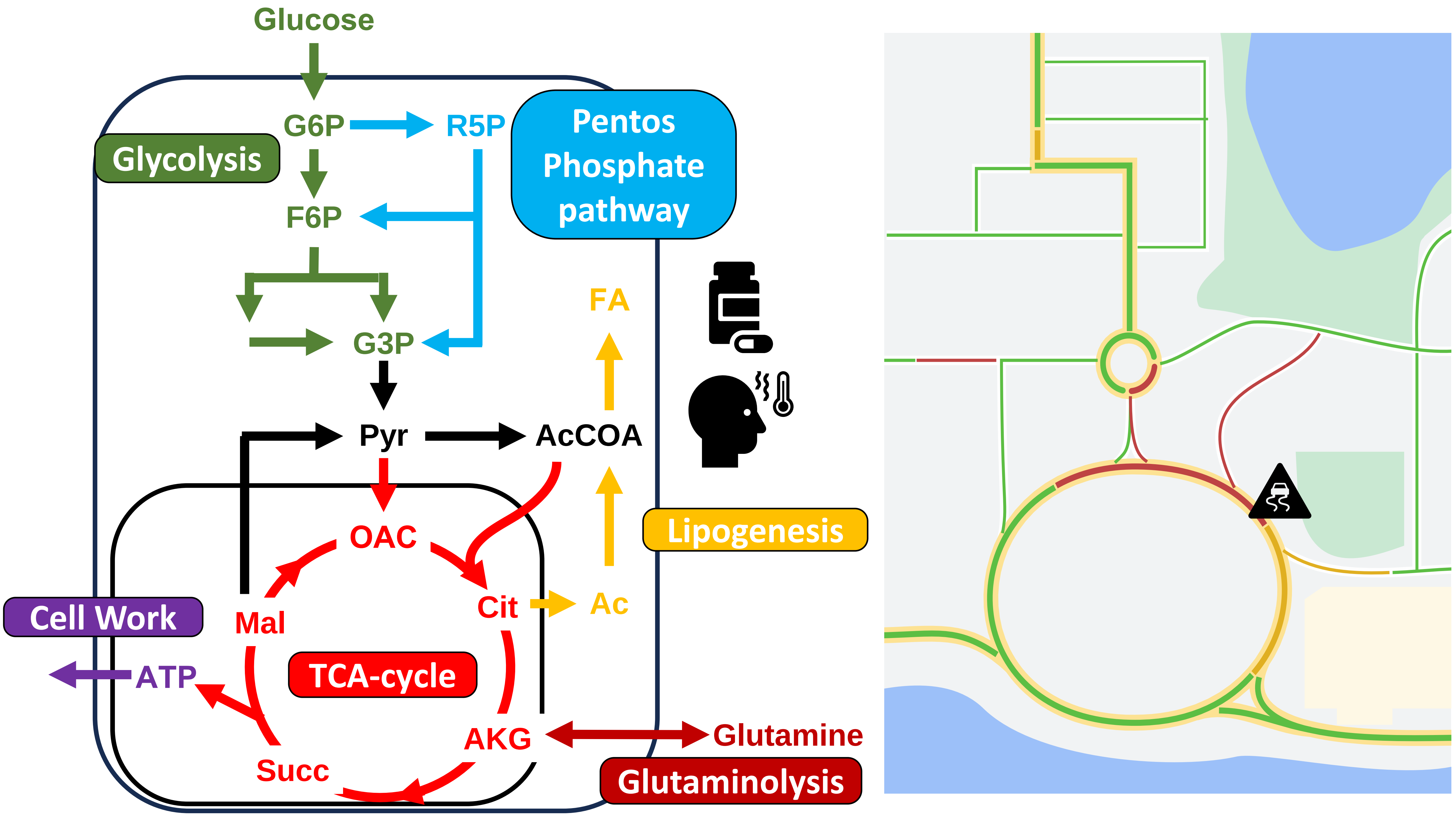
2023-10-11

# Metabolic Flux Analysis (MFA)

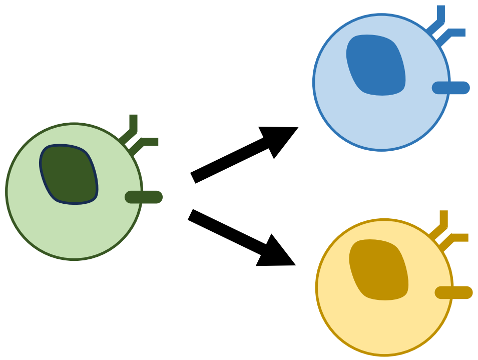
- Quantitatively determine the flux configuration

- Steady state

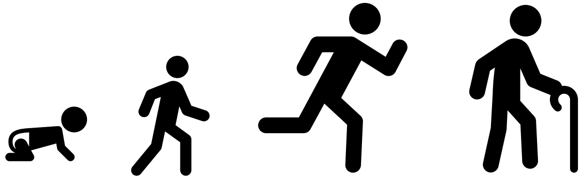



Glucose
G6P
R5P
Pentos Phosphate pathway
Glycolysis
F6P
G3P
FA
Pyr
AcCOA
OAC
Cit
Ac
Lipogenesis
Cell Work
Mal
ATP
TCA-cycle
AKG
Glutamine
Succ
Glutaminolysis

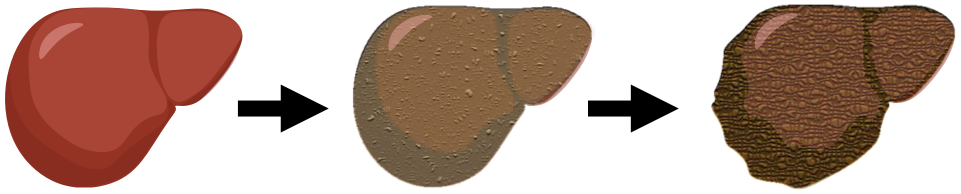
# Systematic alterations of Metabolic Fluxes



T-Cell differentiation



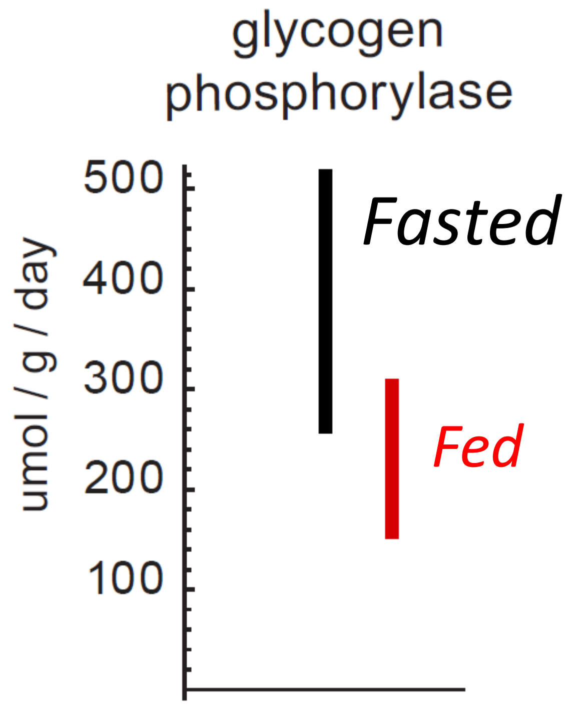
Ageing

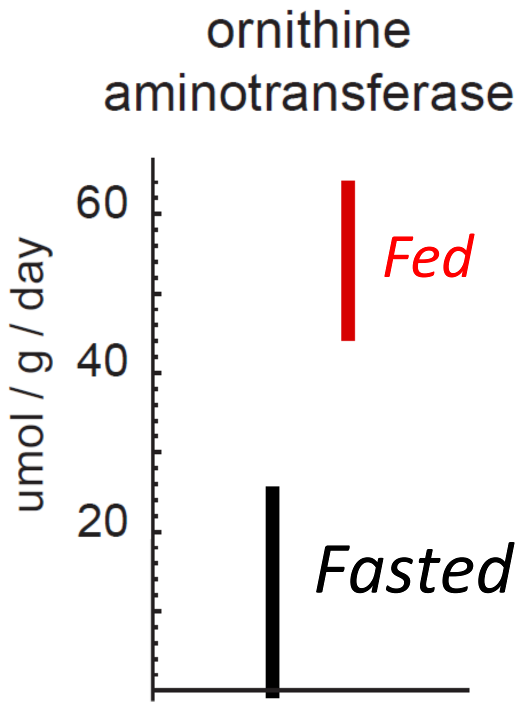


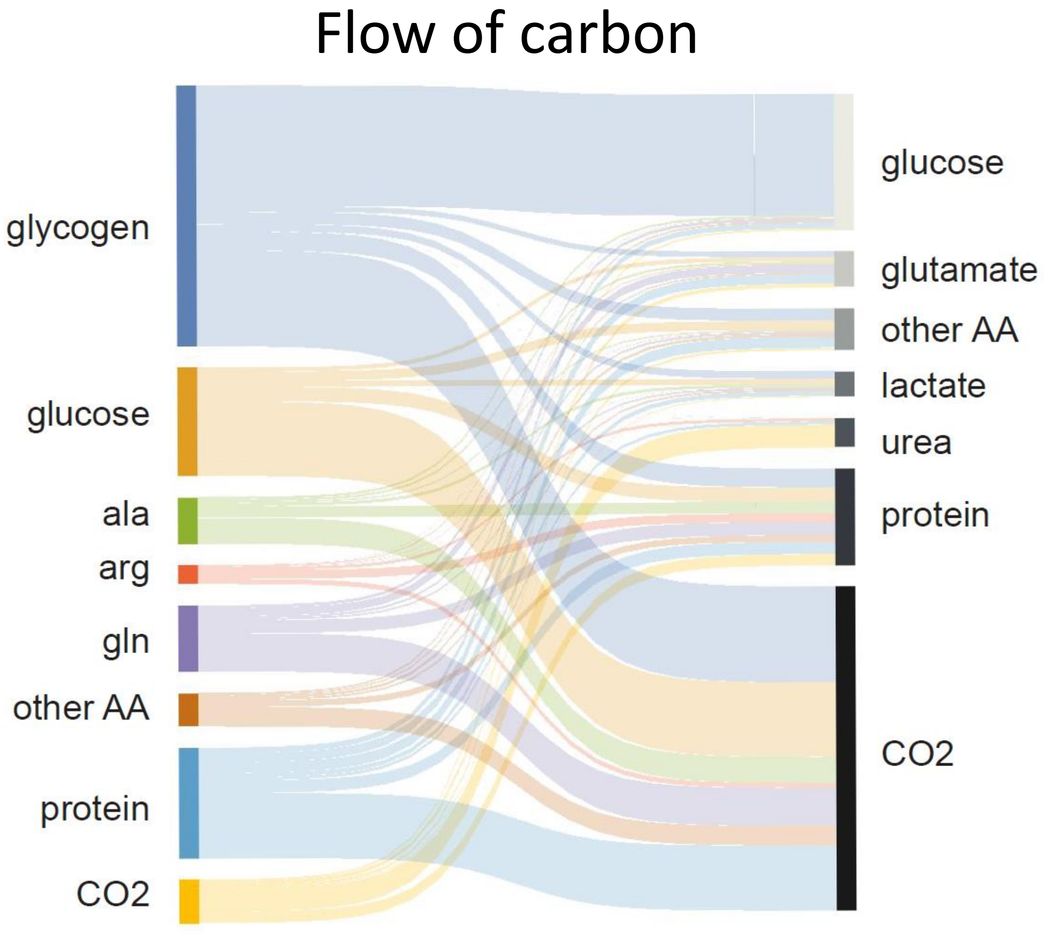
Normal NAFLD NASH

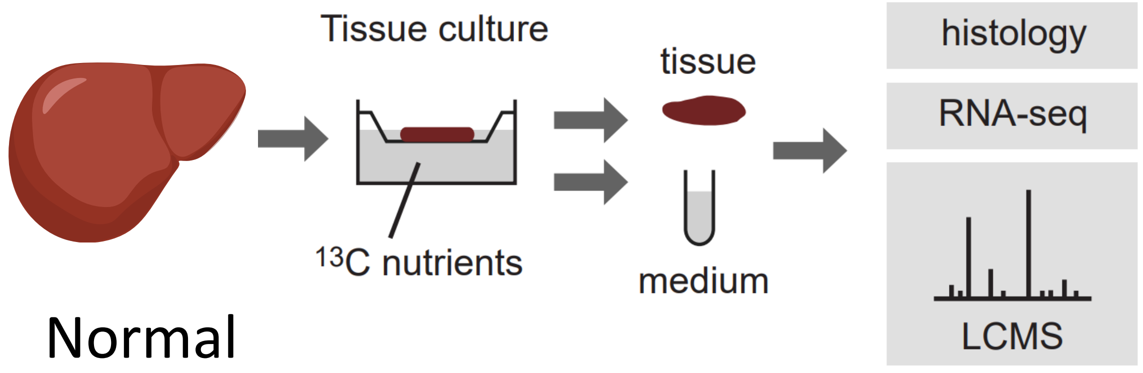
Metabolic syndrome

Neurodegenerative diseases

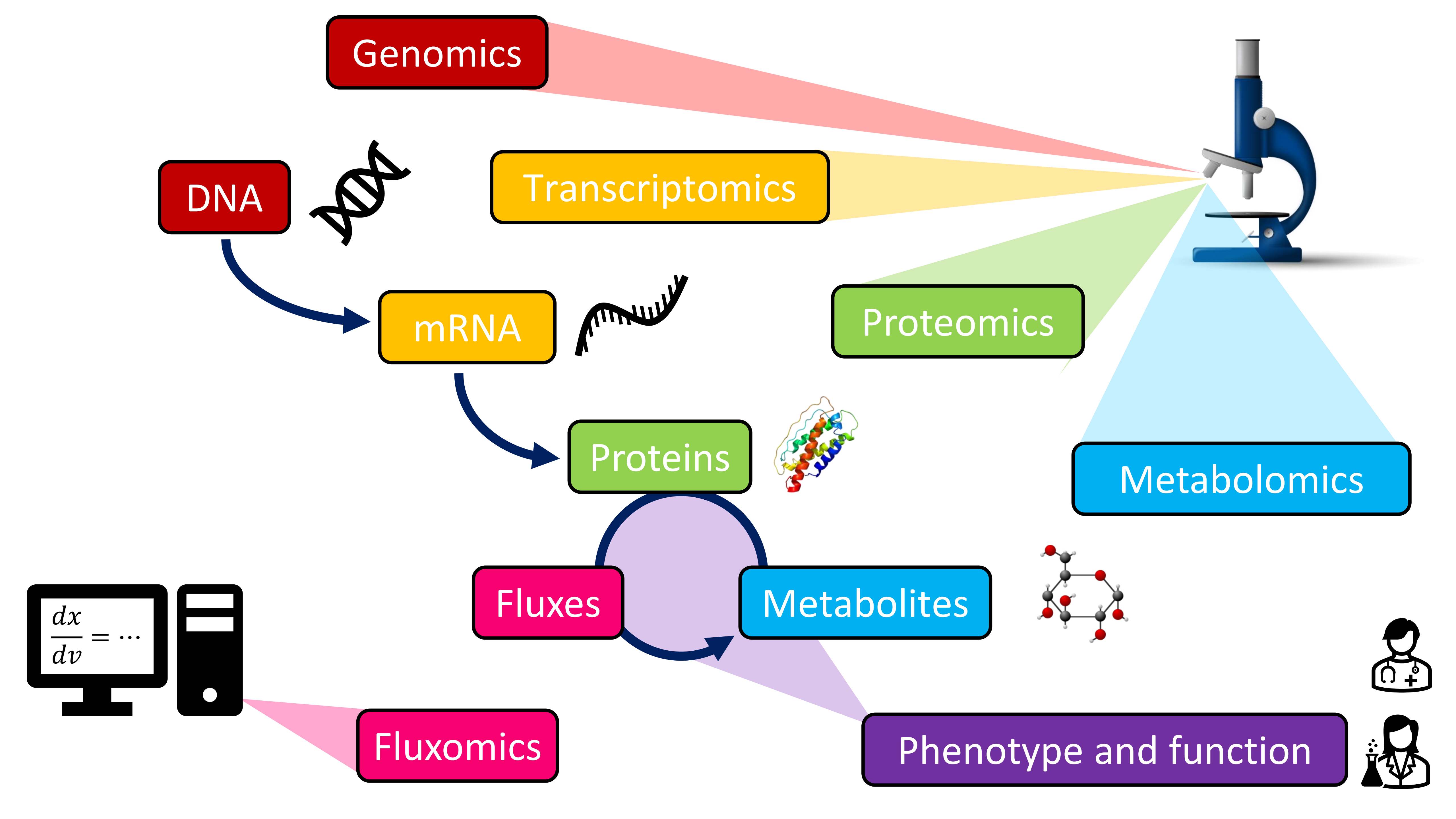


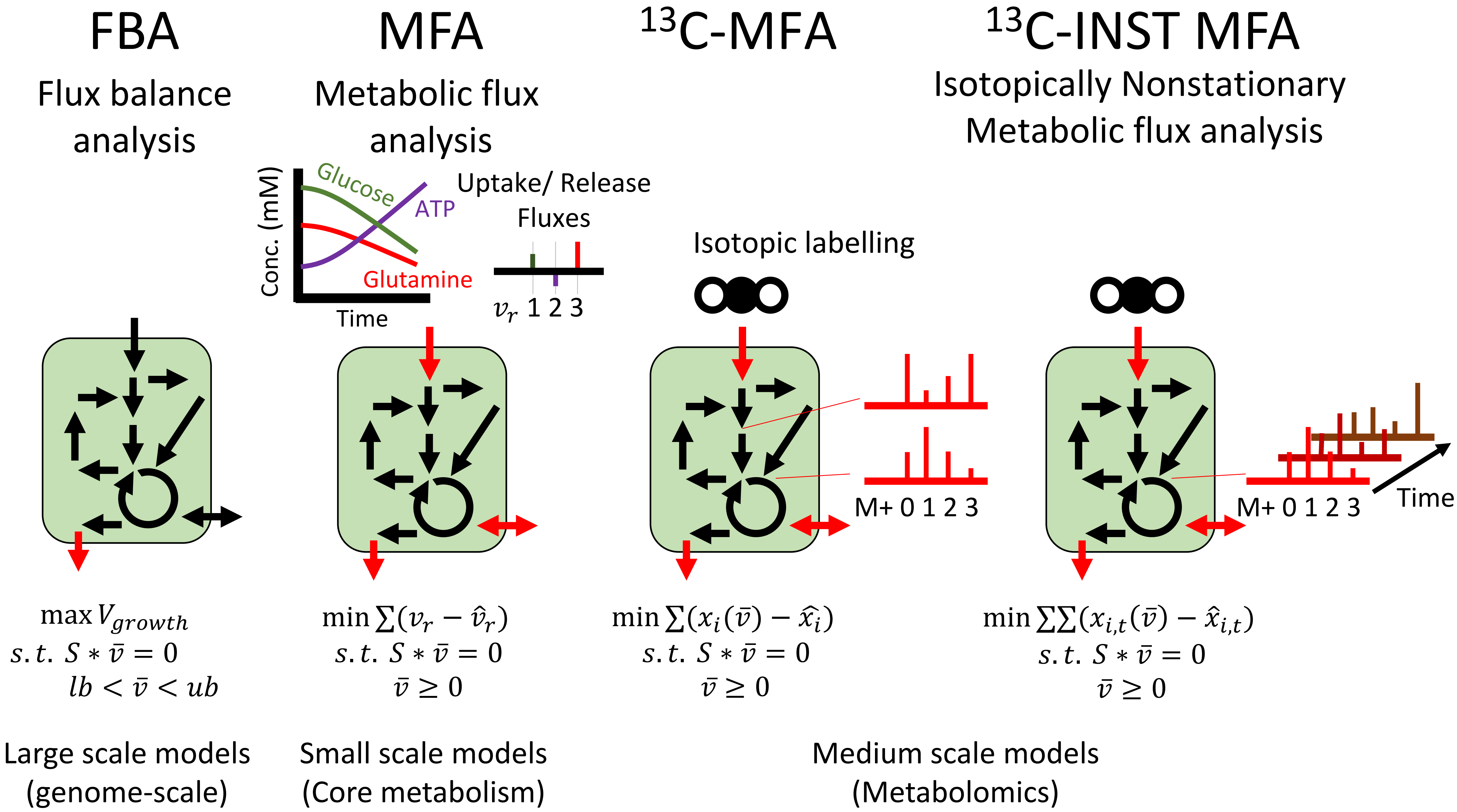



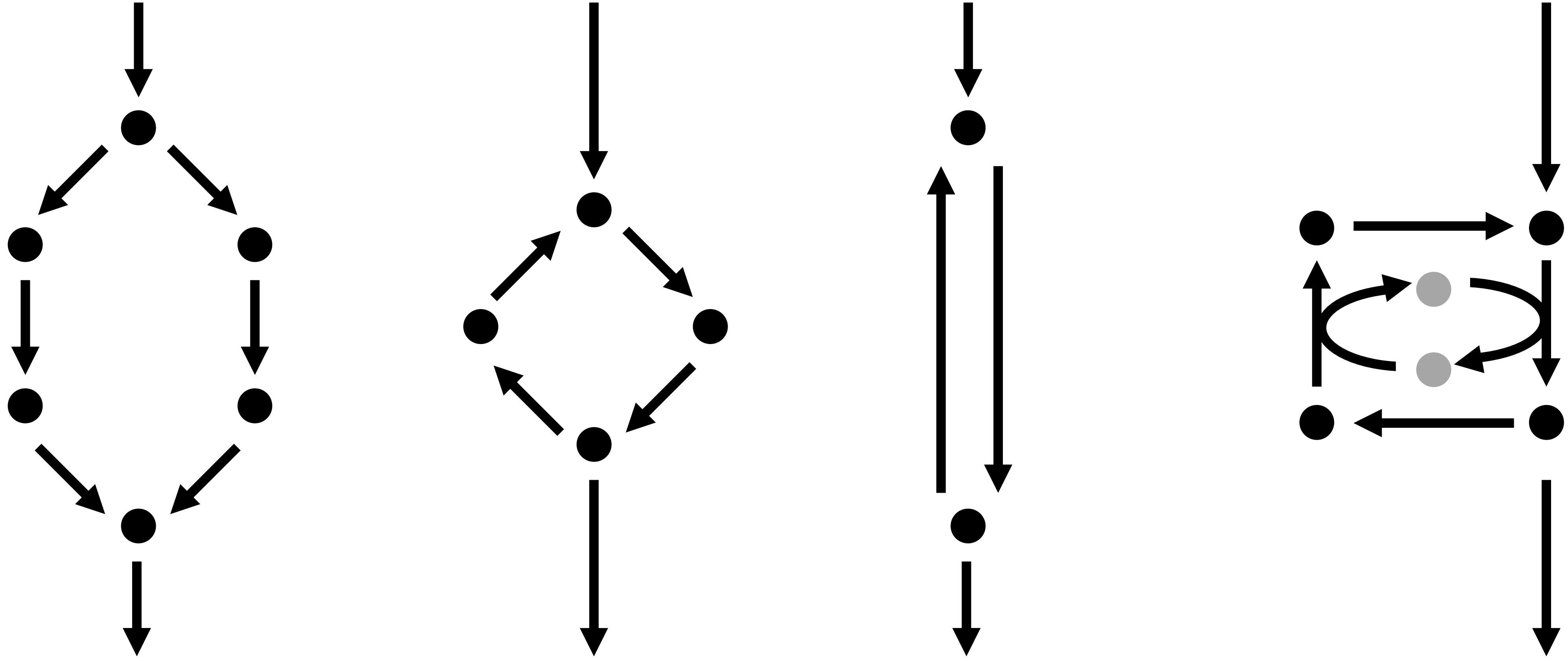

Grankvist N, *et al*, Global 13C tracing and metabolic flux analysis of intact human liver tissue ex vivo, Submitted Nat. Metab.


Genomics
DNA
Transcriptomics
mRNA
Proteomics
Proteins
Metabolomics
Fluxes
Metabolites
$\frac{dx}{dv} = \cdots$
Fluxomics
Phenotype and function



FBA
Flux balance analysis
MFA
Metabolic flux analysis
$^{13}$C-MFA
$^{13}$C-INST MFA
Isotopically Nonstationary Metabolic flux analysis
Conc. (mM)
Glucose
ATP
Glutamine
Time
Uptake/ Release Fluxes
$v_r$ 1 2 3
Isotopic labelling
M+ 0 1 2 3
M+ 0 1 2 3
Time
$\max V_{growth}$
$s.t.\ S * \bar{v} = 0$
$lb < \bar{v} < ub$
$\min \sum(v_r - \hat{v}_r)$
$s.t.\ S * \bar{v} = 0$
$\bar{v} \geq 0$
$\min \sum(x_i(\bar{v}) - \hat{x}_i)$
$s.t.\ S * \bar{v} = 0$
$\bar{v} \geq 0$
$\min \sum\sum(x_{i,t}(\bar{v}) - \hat{x}_{i,t})$
$s.t.\ S * \bar{v} = 0$
$\bar{v} \geq 0$
Large scale models (genome-scale)
Small scale models (Core metabolism)
Medium scale models (Metabolomics)

# Why isotopic labelling?



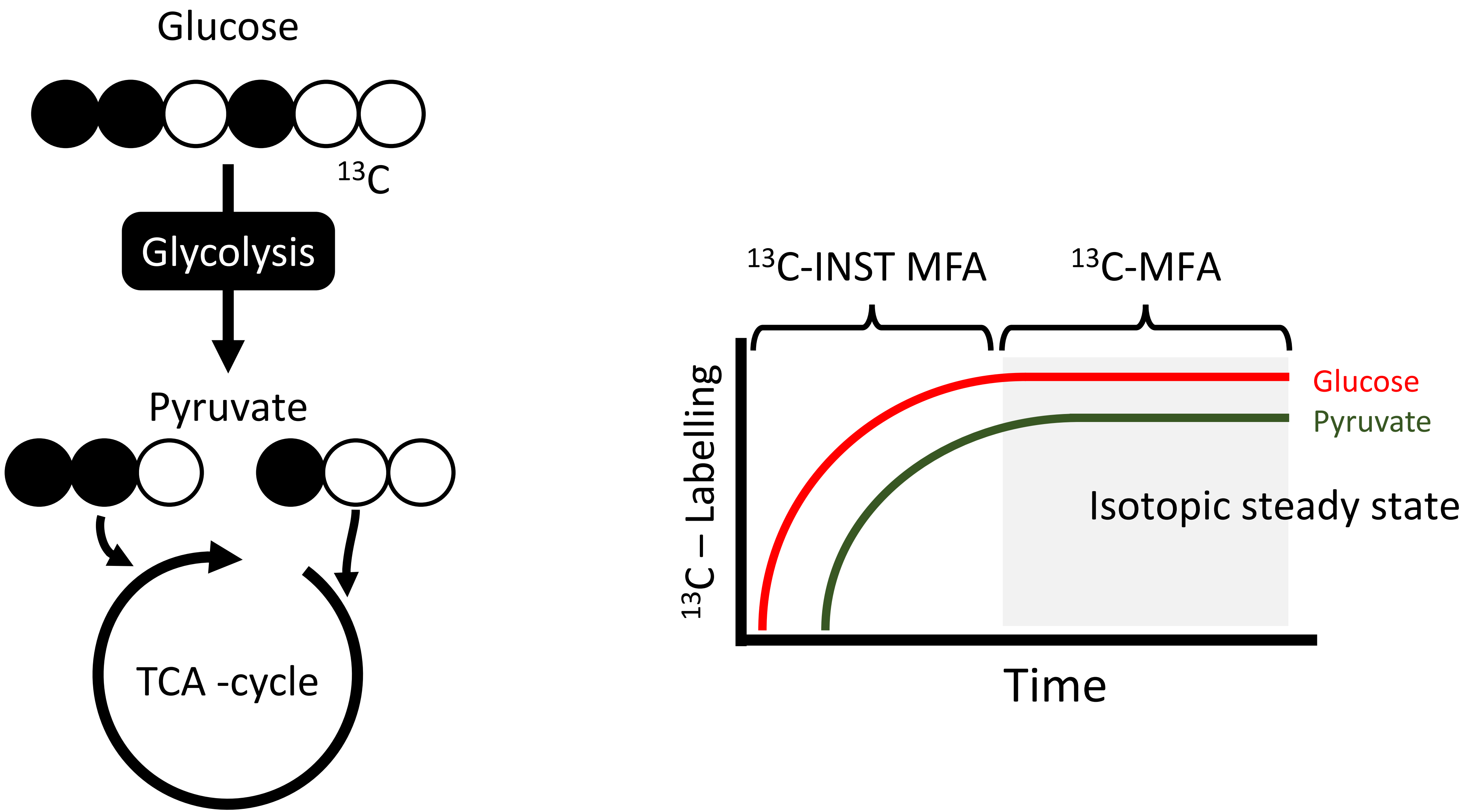
Parallel pathways

Internal cycles

Reversible reactions

Unbalances Co-metabolites

Wiechert W., 2001, Metab. Eng.


Glucose
$^{13}C$
Glycolysis
Pyruvate
TCA -cycle
$^{13}C$-INST MFA
$^{13}C$-MFA
Glucose
Pyruvate
Isotopic steady state
$^{13}C$ – Labelling
Time

# Reactions

$v_1 \quad A \rightarrow B$

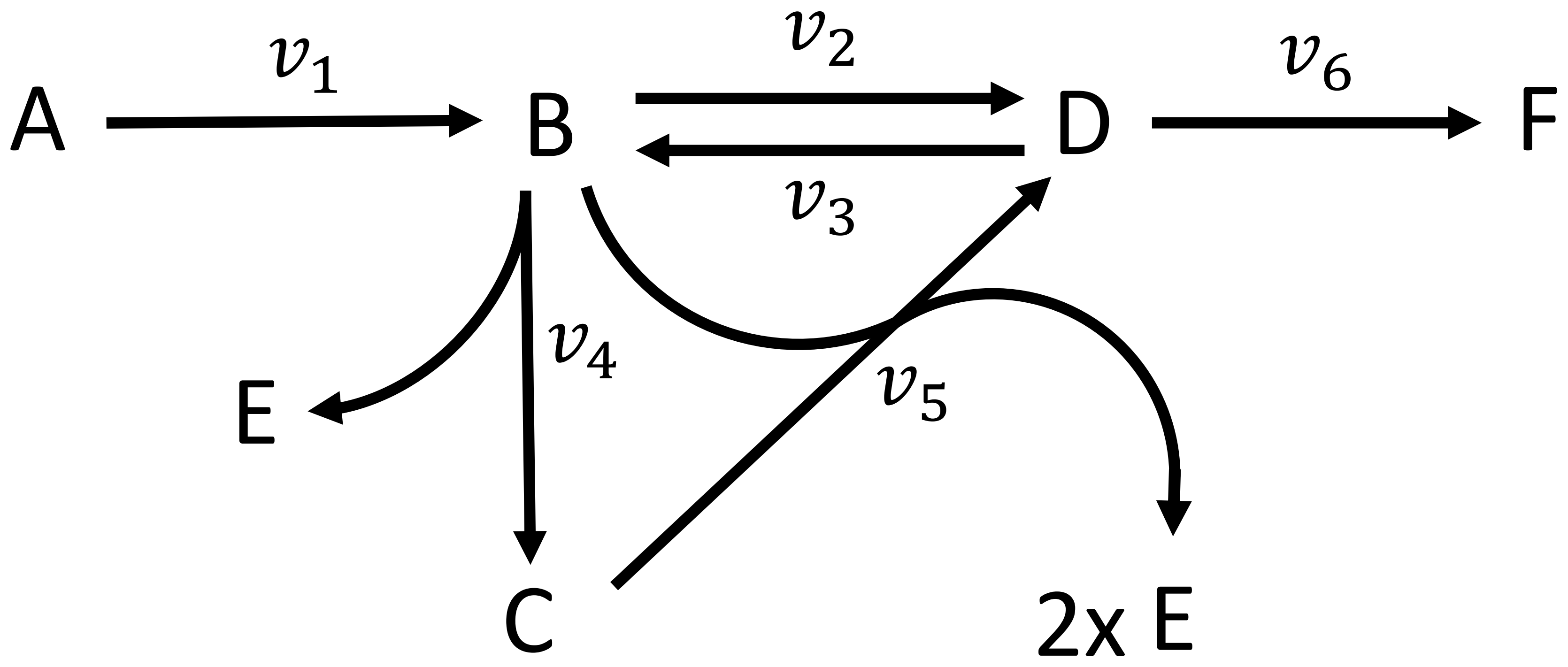
$v_2 \quad B \rightarrow D$

$v_3 \quad D \rightarrow B$

$v_4 \quad B \rightarrow C + E$

$v_5 \quad B + C \rightarrow D + 2E$

$v_6 \quad D \rightarrow F$



# Reactions

$v_1 \quad A \rightarrow B$

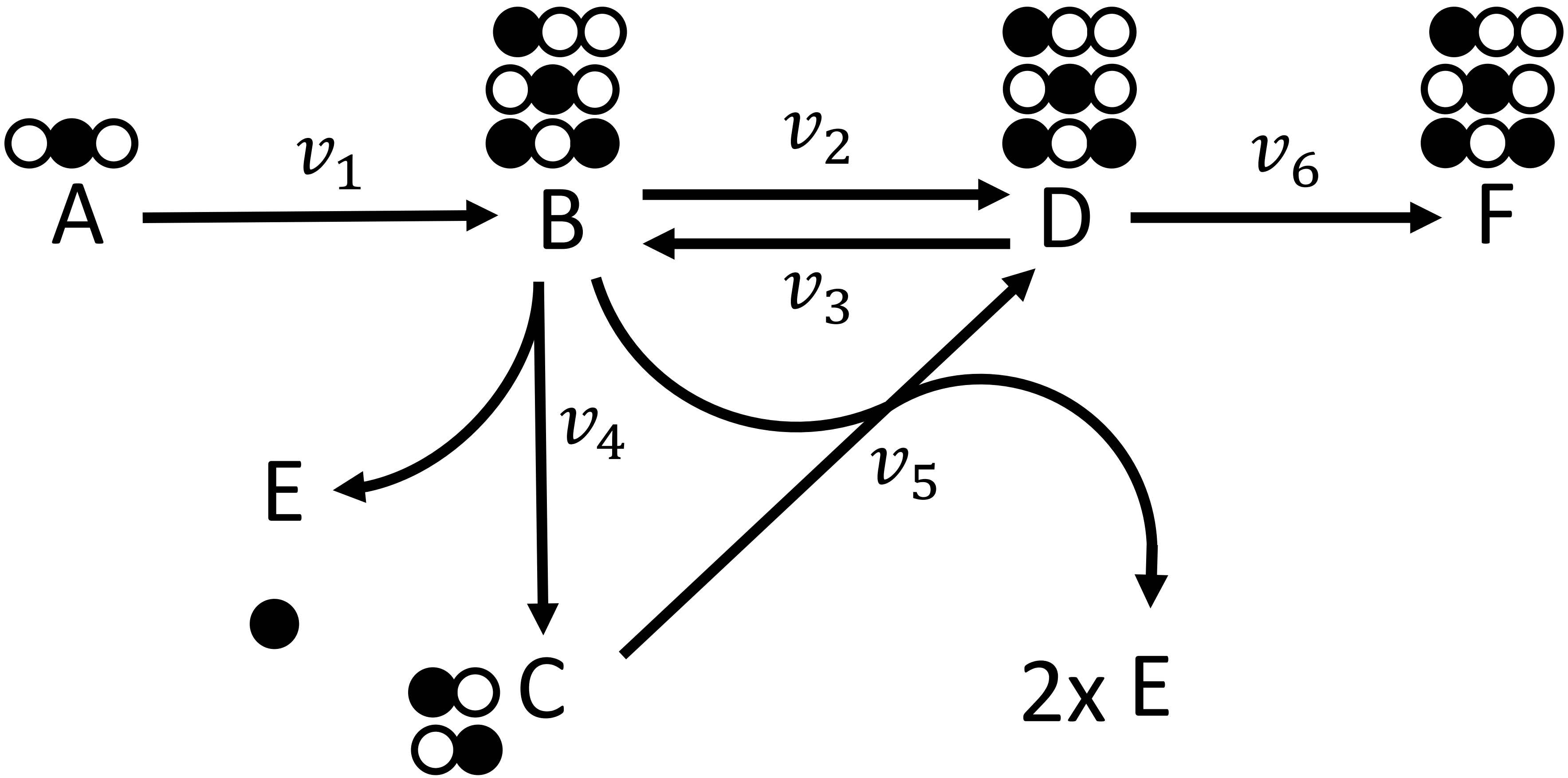
$v_2 \quad B \rightarrow D$

$v_3 \quad D \rightarrow B$

$v_4 \quad B \rightarrow C + E$

$v_5 \quad B + C \rightarrow D + 2E$

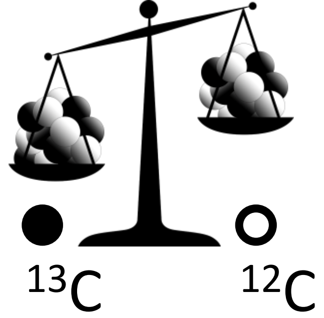
$v_6 \quad D \rightarrow F$



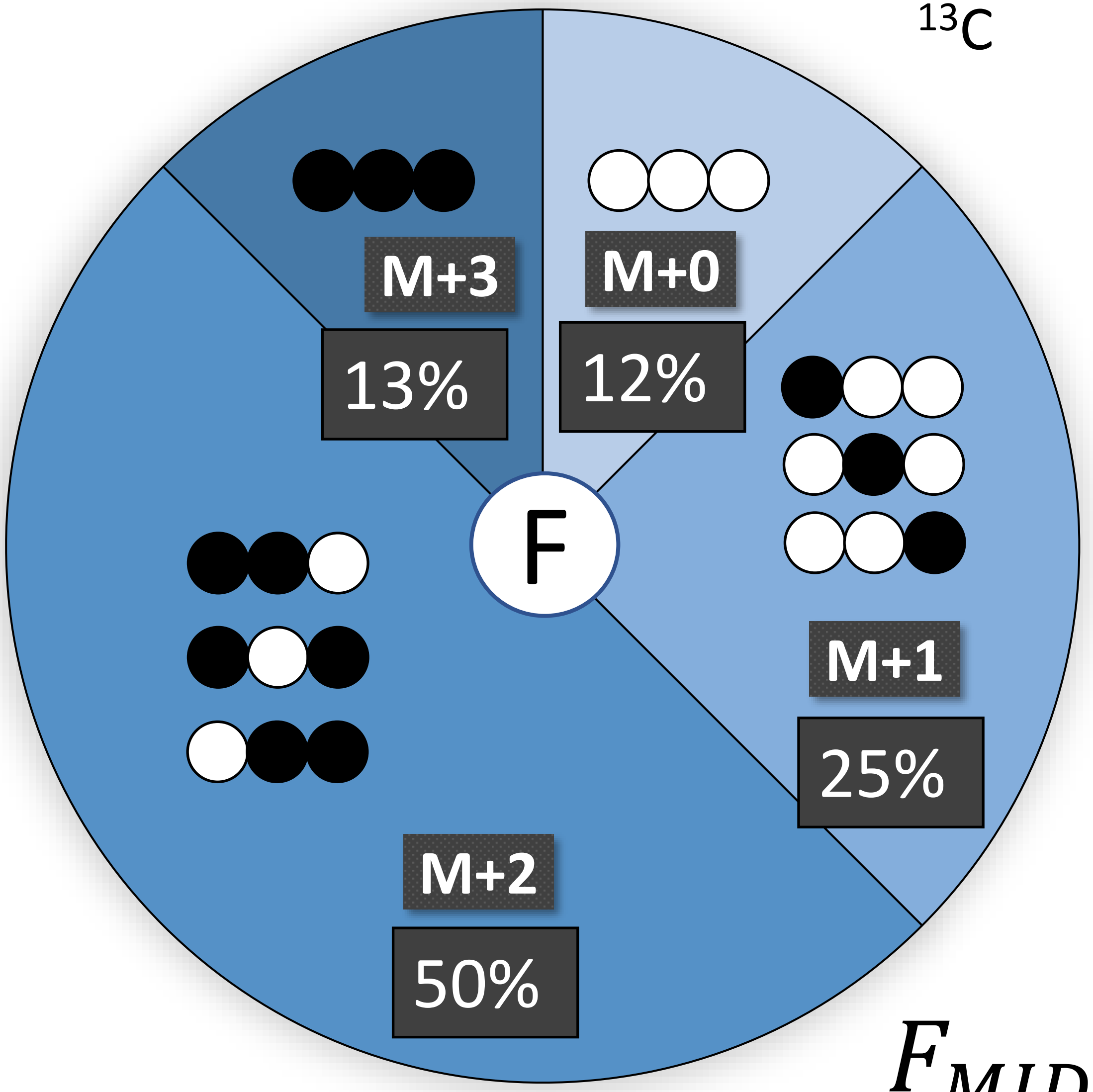
Isotope isomers

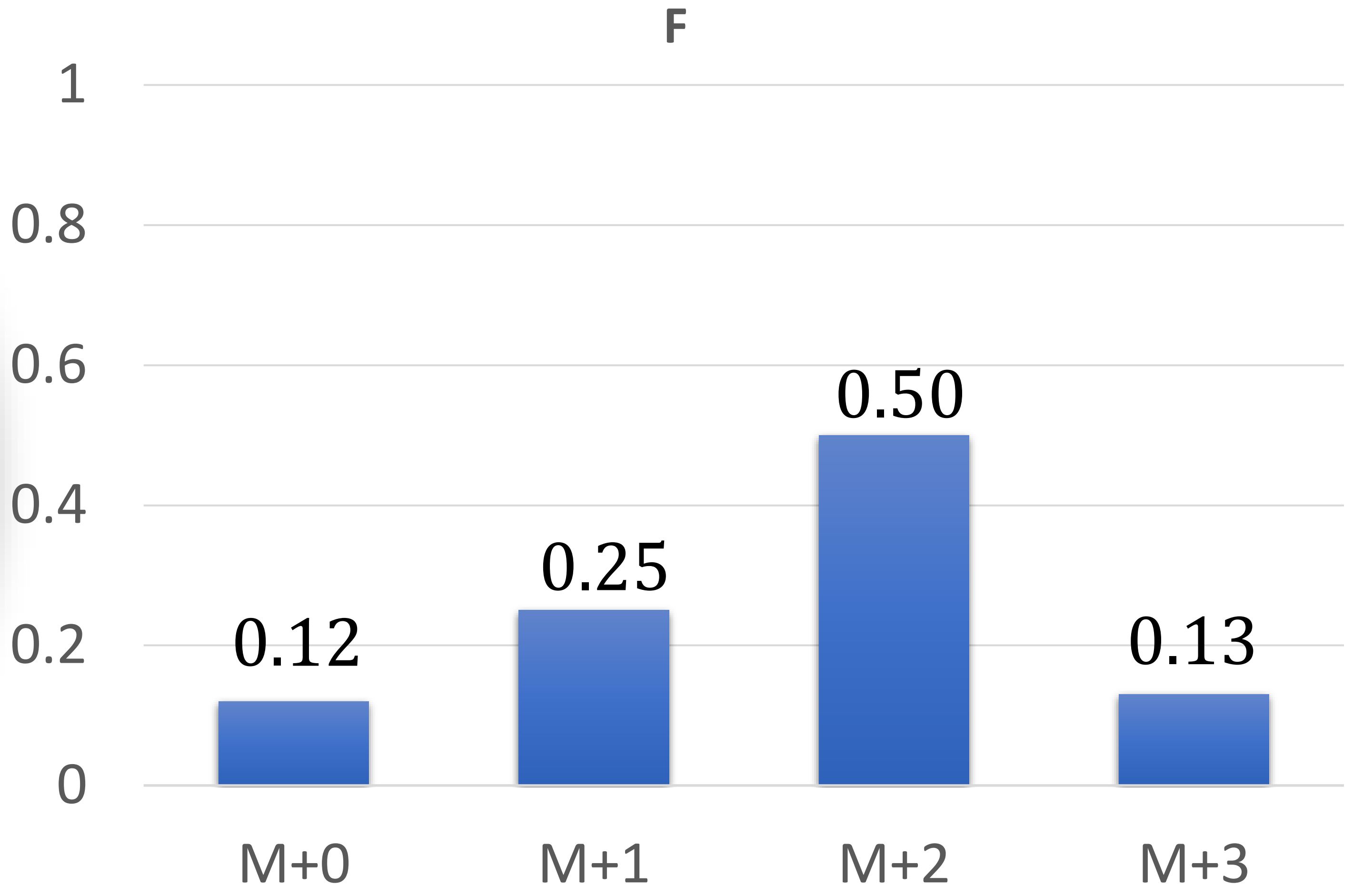
=

Isotopomers

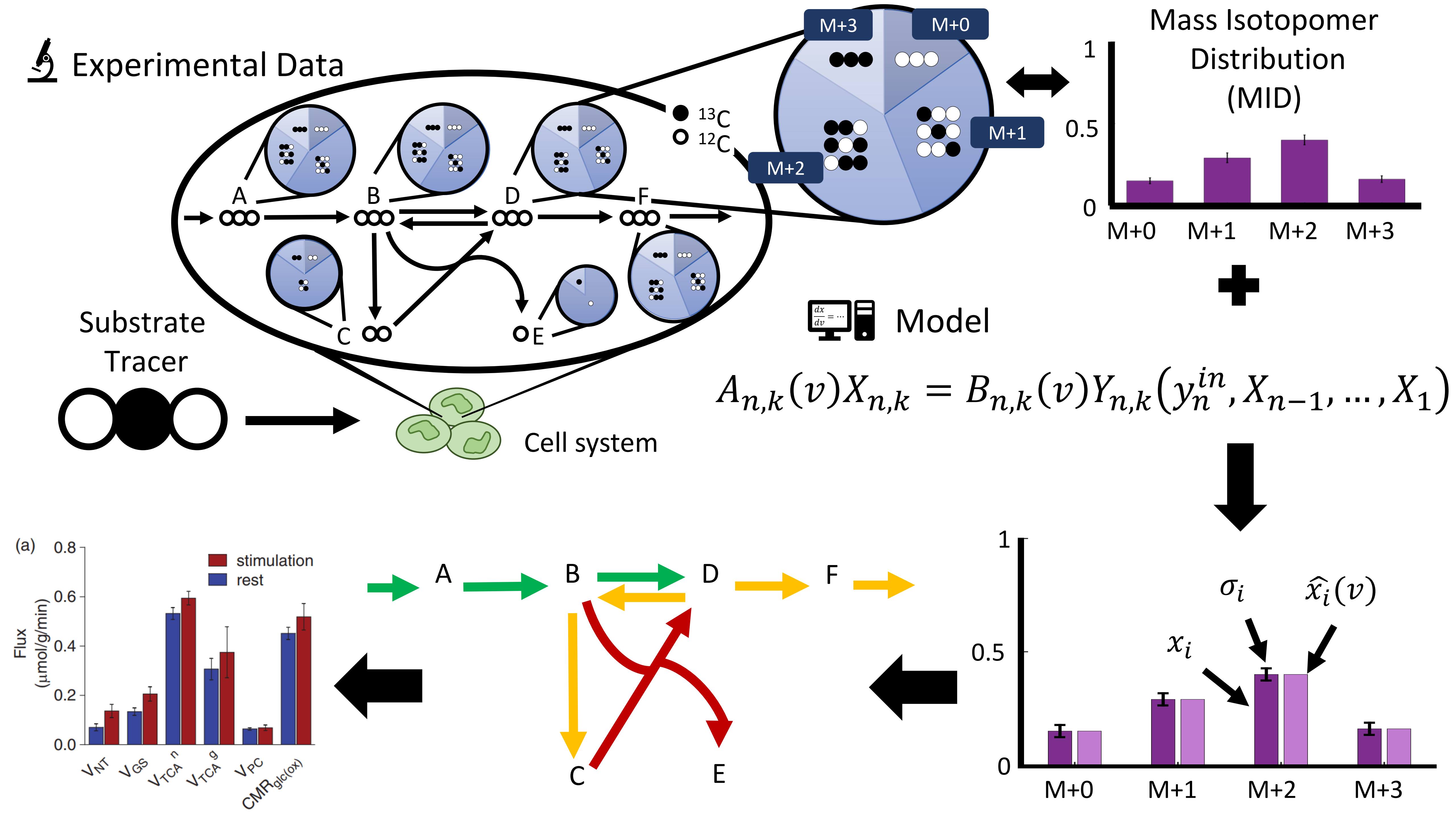



# Mass Isotopomer Distribution (MID)



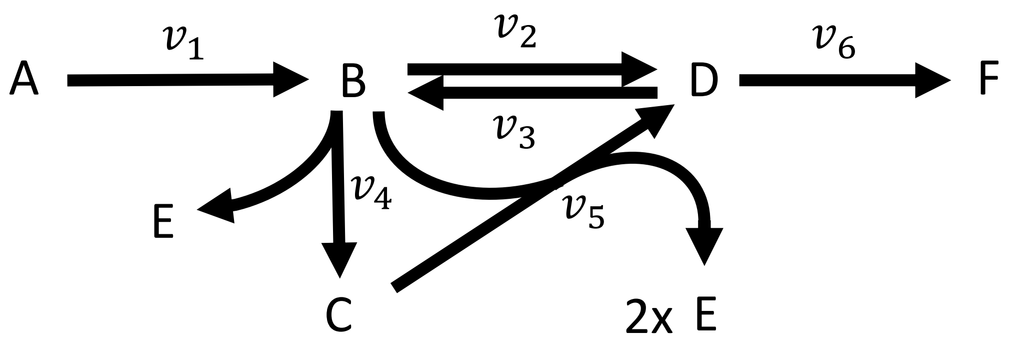




$$F_{MID} = [0.12 \quad 0.25 \quad 0.5 \quad 0.13]$$



Experimental Data
A
B
C
D
E
F
13C
12C
M+3
M+0
M+1
M+2
Mass Isotopomer Distribution (MID)
1
0.5
0
M+0
M+1
M+2
M+3
Substrate Tracer
Cell system
Model
$A_{n,k}(v)X_{n,k} = B_{n,k}(v)Y_{n,k}(y_n^{in}, X_{n-1}, \dots, X_1)$
(a)
Flux (µmol/g/min)
0.8
0.6
0.4
0.2
0.0
stimulation
rest
$V_{NT}$
$V_{GS}$
$V_{TCA}^n$
$V_{TCA}^g$
$V_{PC}$
$CMR_{glc(ox)}$
A
B
D
F
C
E
1
0.5
0
$\sigma_i$
$\widehat{x_i}(v)$
$x_i$
M+0
M+1
M+2
M+3

# Stoichiometry




$$\Rightarrow \begin{cases} v_1 + v_3 = v_2 + v_4 + v_5 \\ v_4 = v_5 \\ v_5 + v_2 = v_3 + v_6 \end{cases}$$

| | $v_1$ | $v_2$ | $v_3$ | $v_4$ | $v_5$ | $v_6$ |
|---|---|---|---|---|---|---|
| **A** | -1 | 0 | 0 | 0 | 0 | 0 |
| **B** | 1 | -1 | 1 | -1 | -1 | 0 |
| **C** | 0 | 0 | 0 | 1 | -1 | 0 |
| **D** | 0 | 1 | -1 | 0 | 1 | -1 |
| **E** | 0 | 0 | 0 | 1 | 2 | 0 |
| **F** | 0 | 0 | 0 | 0 | 0 | 1 |

$$S = \begin{bmatrix} 1 & -1 & 1 & -1 & -1 & 0 \\ 0 & 0 & 0 & 1 & -1 & 0 \\ 0 & 1 & -1 & 0 & 1 & -1 \end{bmatrix} \quad \bar{v} = \begin{bmatrix} v_1 \\ v_2 \\ v_3 \\ v_4 \\ v_5 \\ v_6 \end{bmatrix}$$

$$\bar{u} = \begin{bmatrix} v_1 \\ v_3 \\ v_4 \end{bmatrix}$$

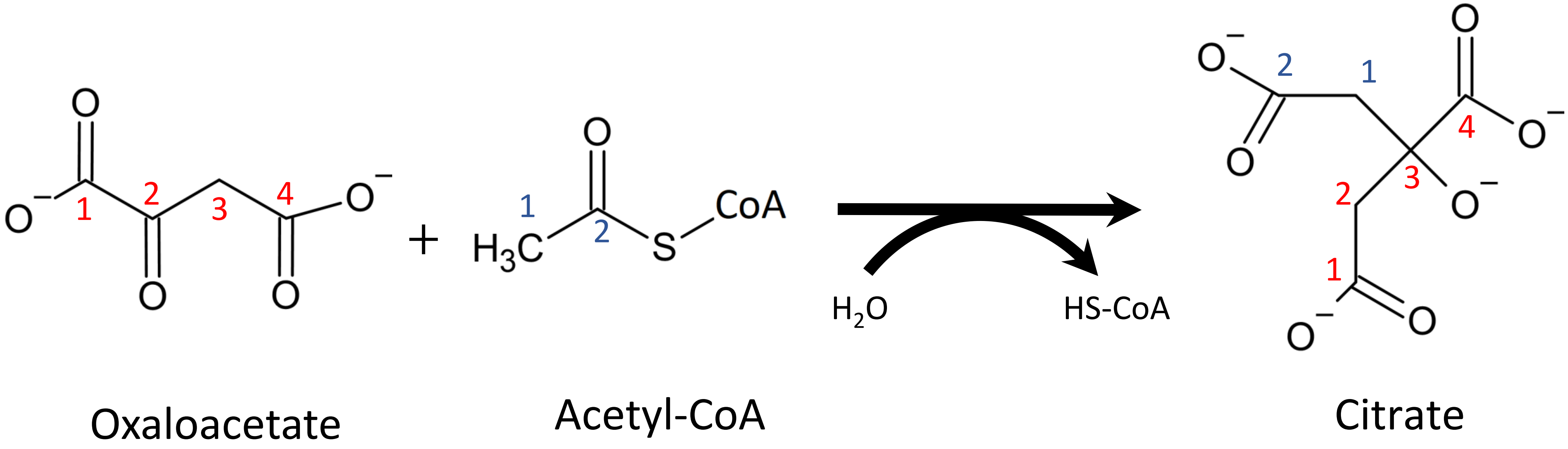
Steady state condition

Independent fluxes

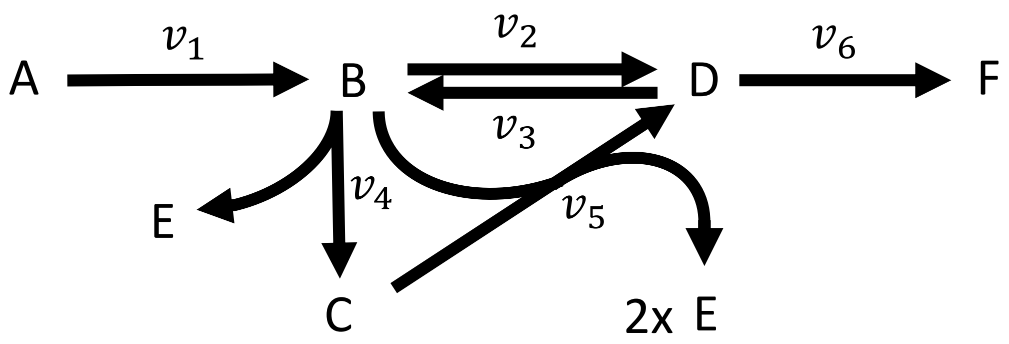
$$S * \bar{v} = 0 \Leftrightarrow \begin{aligned} v_1 - v_2 + v_3 - v_4 - v_5 &= 0 \\ v_4 - v_5 &= 0 \\ v_2 - v_3 + v_5 - v_6 &= 0 \end{aligned} \Rightarrow \begin{aligned} v_2 &= \overbrace{v_1 + v_3 - 2 * v_4} \\ v_5 &= v_4 \\ v_6 &= v_1 - v_4 \end{aligned} \Rightarrow \bar{v} = N * \bar{u}$$

$$N = null(S)$$

# Atom mapping



# Atom mapping




$v_1$ $A(abc) \rightarrow B(abc)$

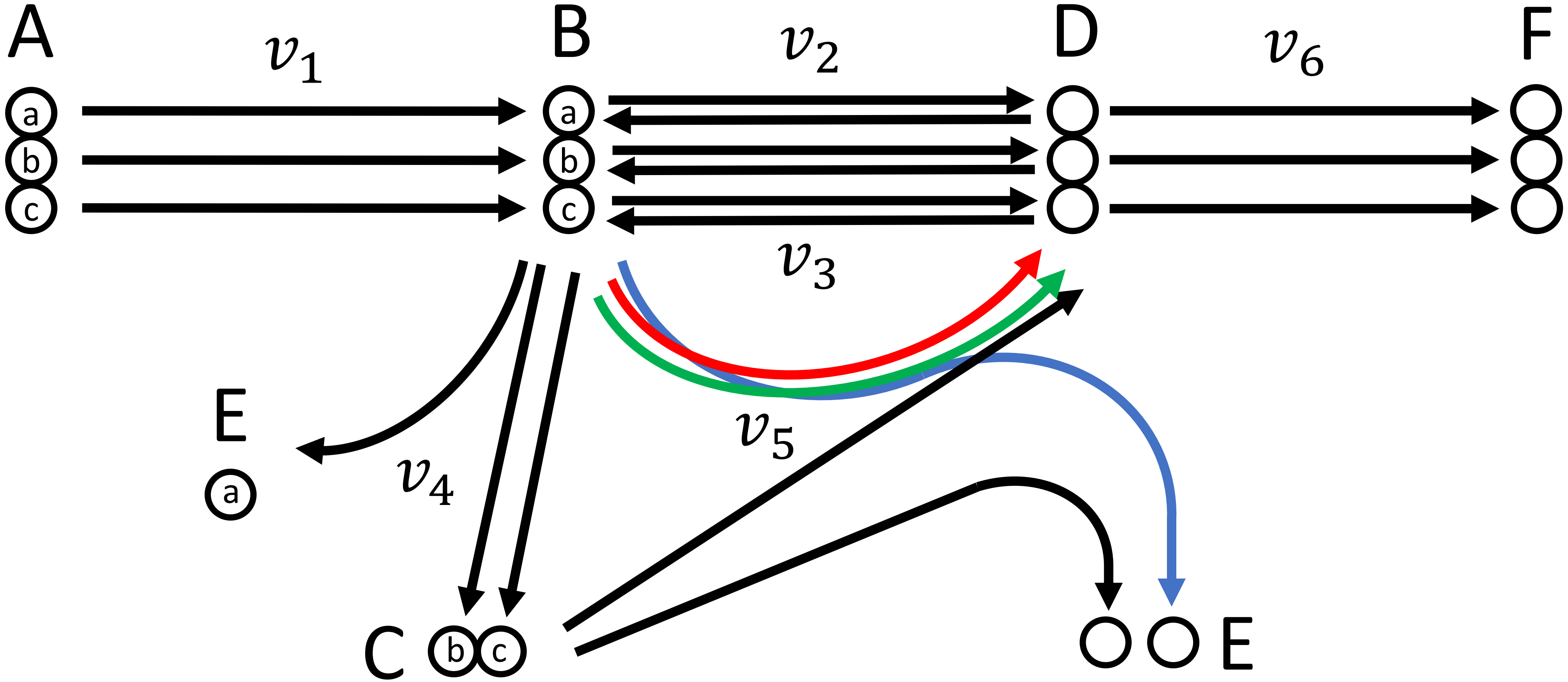
$v_2$ $B(abc) \rightarrow D(abc)$

$v_3$ $D(abc) \rightarrow B(abc)$

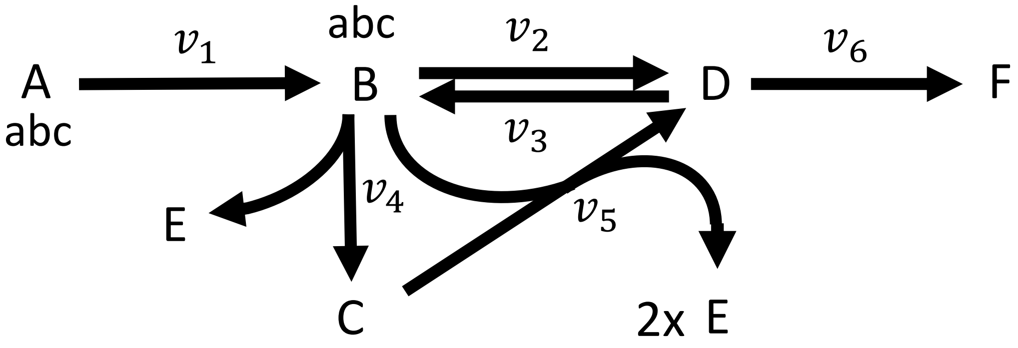
$v_4$ $B(abc) \rightarrow C(bc) + E(a)$

$v_5$ $B(abc) + C(de) \rightarrow D(bcd) + E(a) + E(e)$

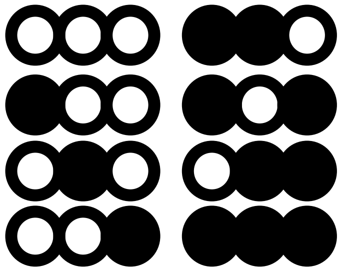
$v_6$ $D(abc) \rightarrow F(abc)$



# ODE equations




$$\frac{dM_{ij}}{dt} = \sum_{k \in In(i)} v_k \sum_{\theta \in Gen(k,i,j)} \prod_{(l,m) \in \theta} r_{lm} - \left( \sum_{k \in Out(i)} v_k \right) r_{ij} = 0$$



$$\frac{d}{dt} A_a = -v_1 A_a \qquad \frac{d}{dt} B_a = v_1 A_a - v_2 B_a + v_3 D_a - v_4 B_a$$

$$\frac{d}{dt} A_b = -v_1 A_b \qquad \frac{d}{dt} B_b = v_1 A_b - v_2 B_b + v_3 D_b - v_4 B_b$$

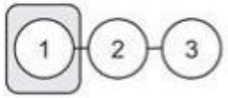
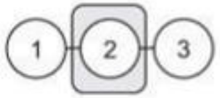
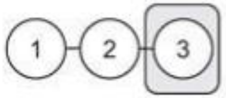
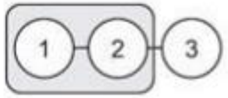
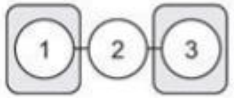
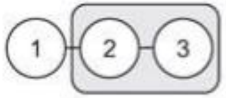
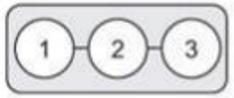
$$\frac{d}{dt} A_c = -v_1 A_c \qquad \frac{d}{dt} B_c = v_1 A_c - v_2 B_c + v_3 D_c - v_4 B_c$$

$$\frac{d}{dt} C_a = v_4 B_b - v_5 C_a \quad \frac{d}{dt} C_b = v_4 B_c - v_5 C_b \qquad \ldots$$

- Number of C-atoms: $N$
- Number of states: $N^2$

- Example: 225 states

# Elementary Metabolite Units

| Elementary metabolite unit | Atoms included in the EMU | EMU size |
|---|---|---|
| $A_1$ |  | 1 |
| $A_2$ |  | 1 |
| $A_3$ |  | 1 |
| $A_{12}$ |  | 2 |
| $A_{13}$ |  | 2 |
| $A_{23}$ |  | 2 |
| $A_{123}$ |  | 3 |

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Metabolic Engineering 9 (2007) 68–86
METABOLIC ENGINEERING
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## Elementary metabolite units (EMU): A novel framework for modeling isotopic distributions

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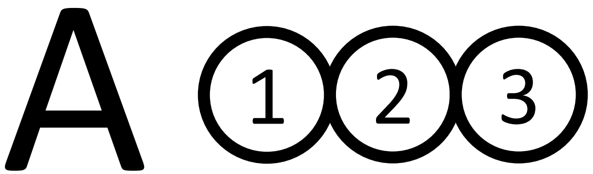
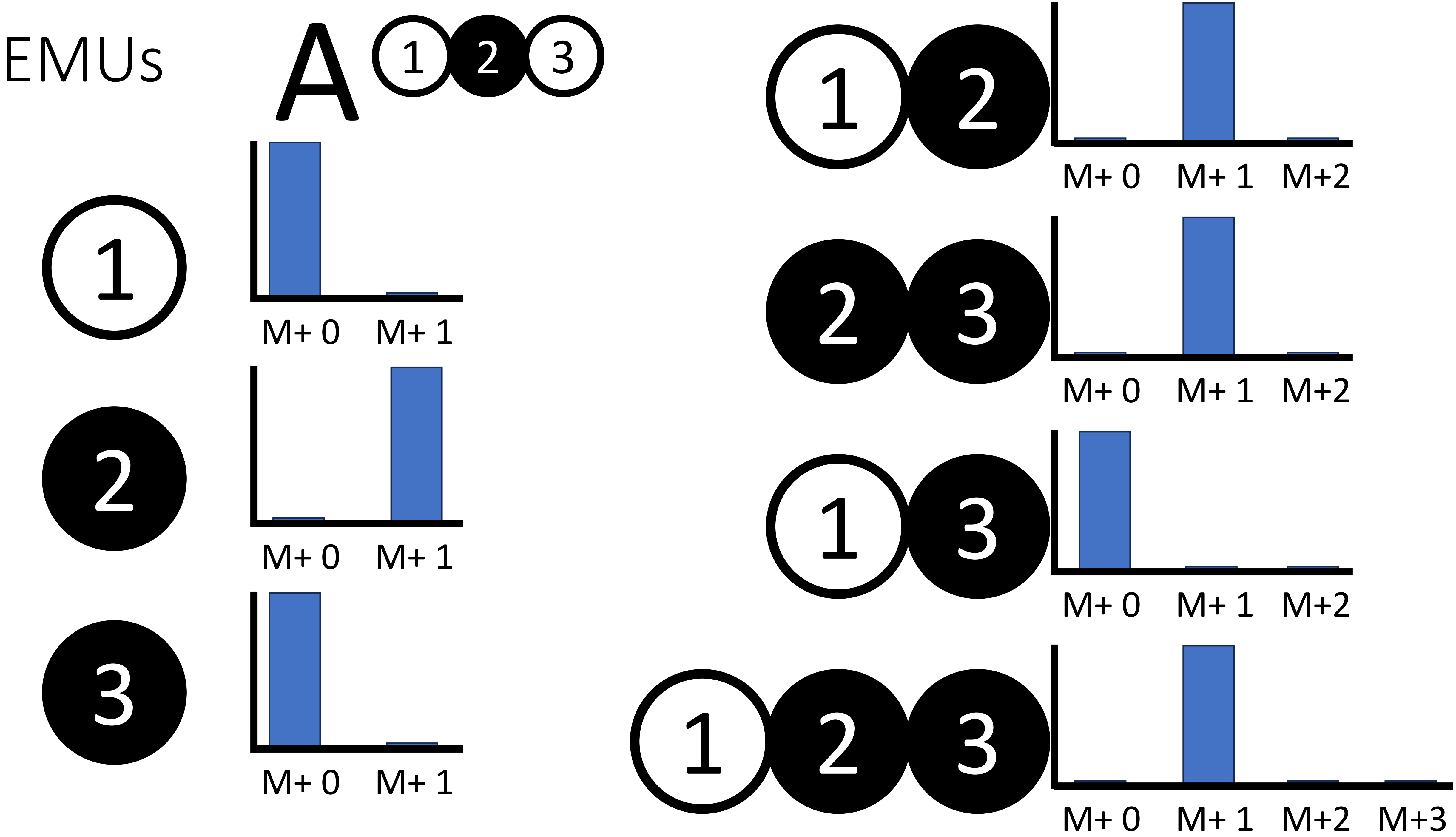
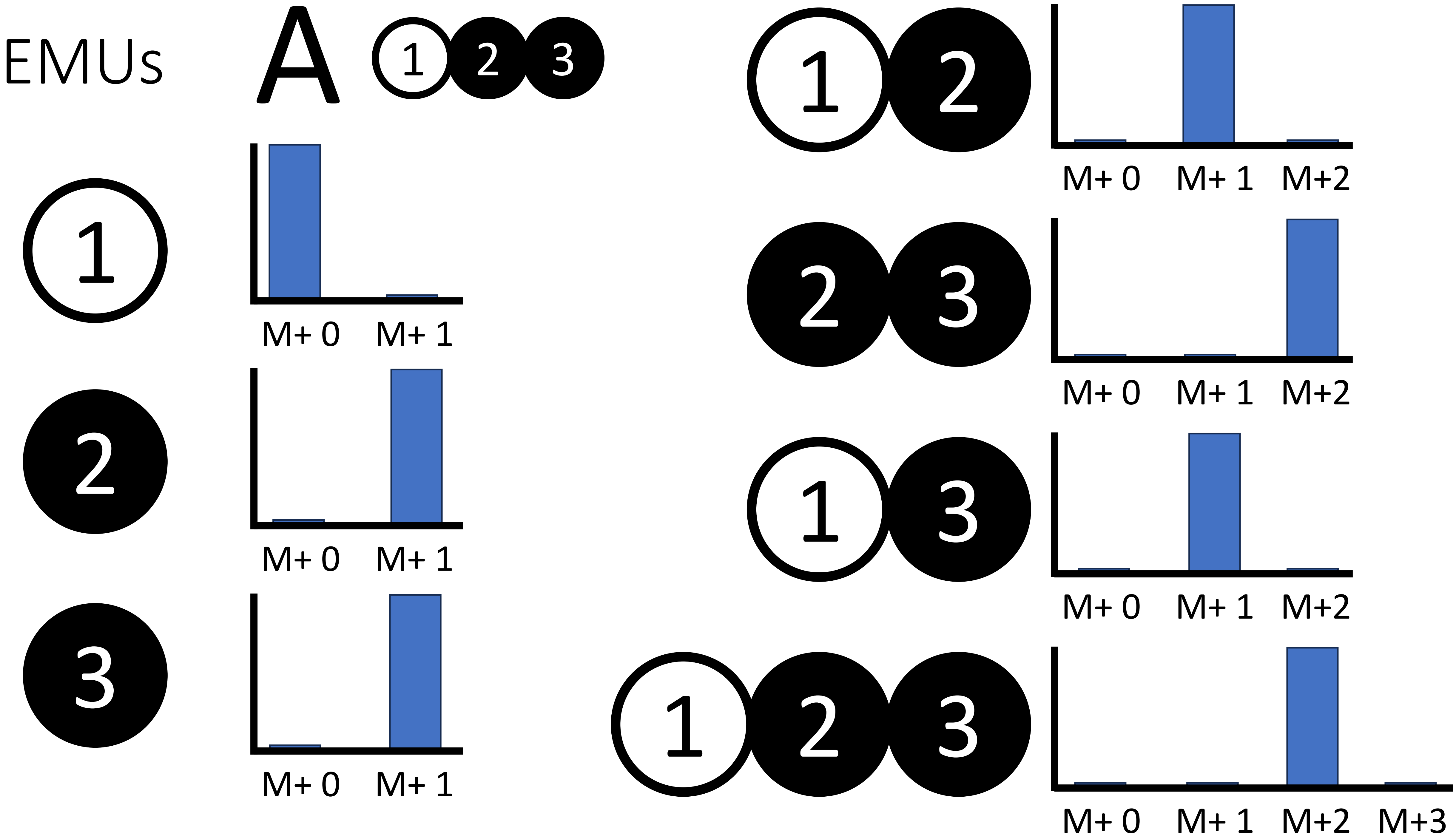
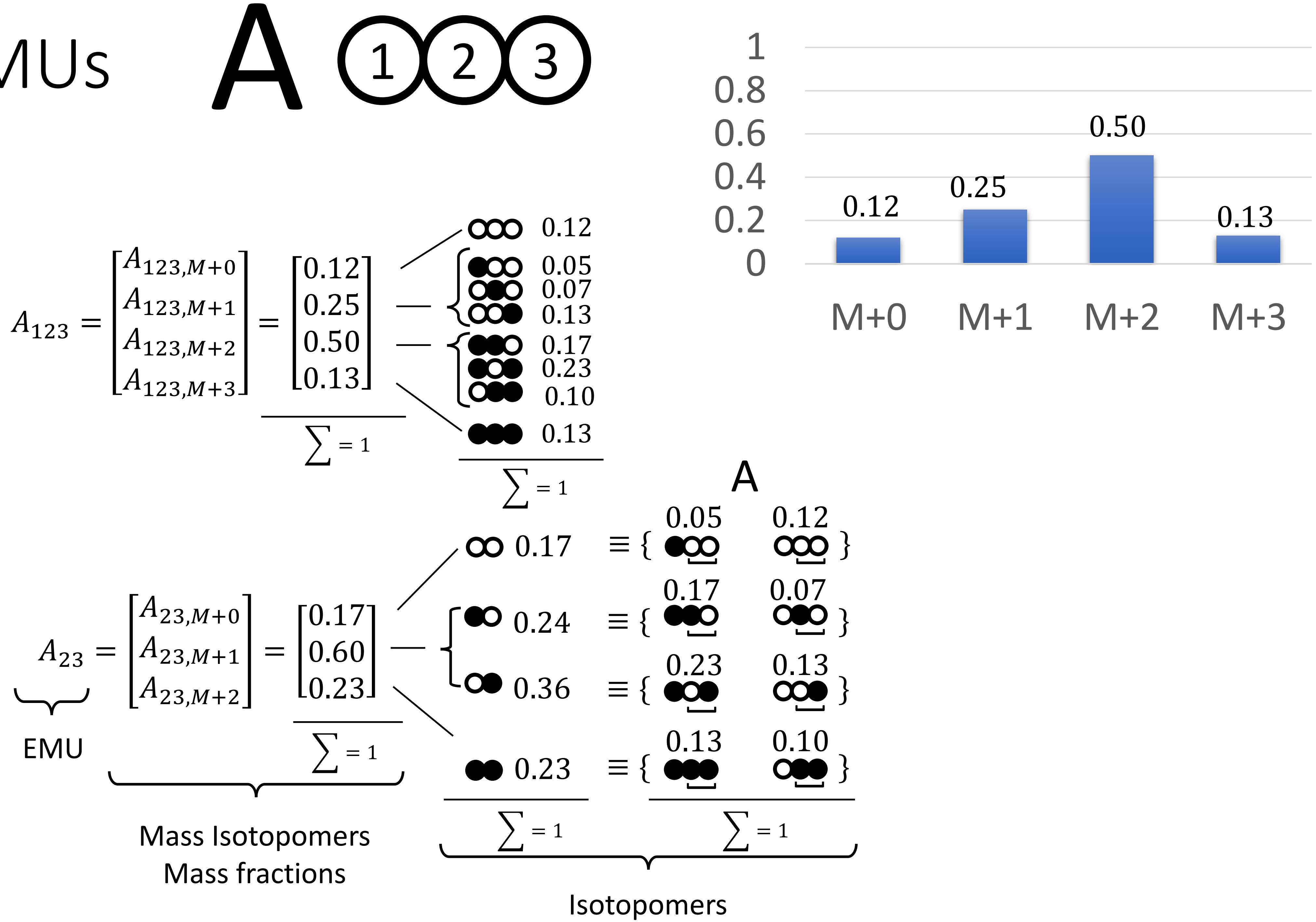
Available online 17 September 2006




EMUs
A
1
2
3
M+ 0
M+ 1
M+ 2
M+ 3


EMUs
A
1
2
3
1 2
M+ 0
M+ 1
M+2
2 3
1 3
1 2 3
M+3

# EMUs

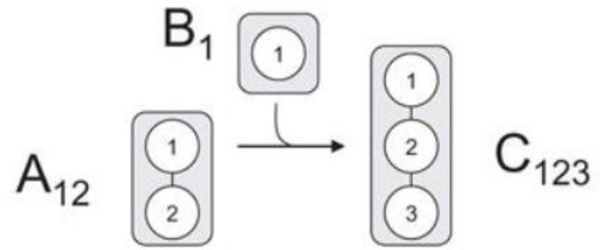


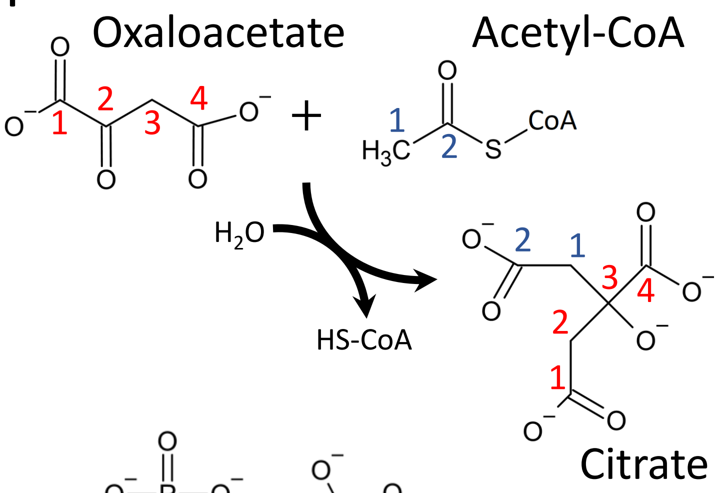
A
1
2
3
1
0.8
0.6
0.4
0.2
0
0.12
0.25
0.50
0.13
M+0
M+1
M+2
M+3
$A_{123} = \begin{bmatrix} A_{123,M+0} \\ A_{123,M+1} \\ A_{123,M+2} \\ A_{123,M+3} \end{bmatrix} = \begin{bmatrix} 0.12 \\ 0.25 \\ 0.50 \\ 0.13 \end{bmatrix}$
$\sum = 1$
0.12
0.05
0.07
0.13
0.17
0.23
0.10
0.13
$\sum = 1$
A
$A_{23} = \begin{bmatrix} A_{23,M+0} \\ A_{23,M+1} \\ A_{23,M+2} \end{bmatrix} = \begin{bmatrix} 0.17 \\ 0.60 \\ 0.23 \end{bmatrix}$
EMU
$\sum = 1$
0.17 ≡ { 0.05 0.12 }
0.24 ≡ { 0.17 0.07 }
0.36 ≡ { 0.23 0.13 }
0.23 ≡ { 0.13 0.10 }
$\sum = 1$
$\sum = 1$
Mass Isotopomers
Mass fractions
Isotopomers

Antoniewicz *et al.*,*Metab. Eng.*, *2006*

# EMUs

## A) Condensation reaction

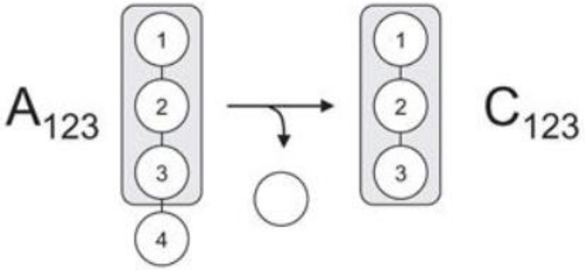


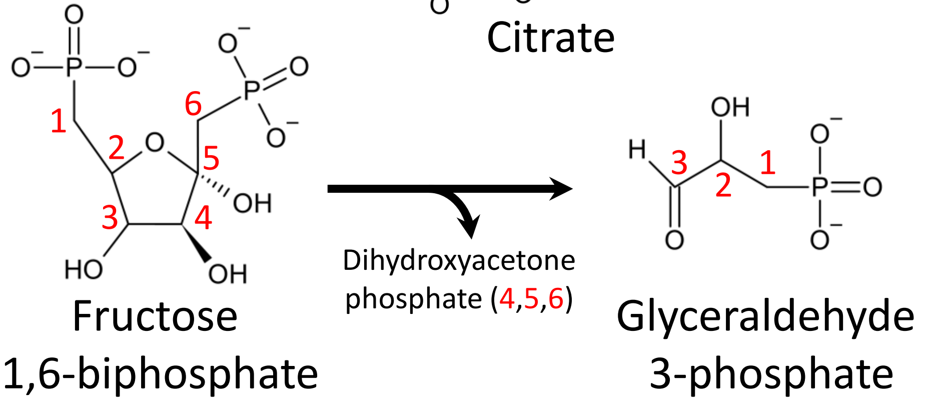




$$C_{123} = A_{12} \times B_1$$

$$\begin{bmatrix} C_{123,M+0} \\ C_{123,M+1} \\ C_{123,M+2} \\ C_{123,M+3} \end{bmatrix} = \begin{bmatrix} A_{12,M+0} * B_{1,M+0} \\ A_{12,M+0} * B_{1,M+1} + A_{12,M+1} * B_{1,M+0} \\ A_{12,M+1} * B_{1,M+1} + A_{12,M+2} * B_{1,M+0} \\ A_{12,M+2} * B_{1,M+1} \end{bmatrix}$$

## B) Cleavage reaction

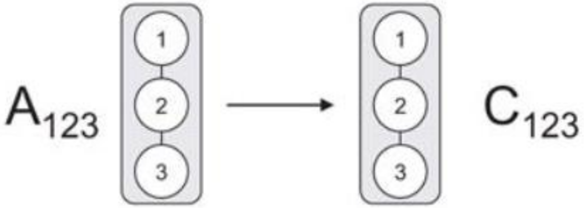


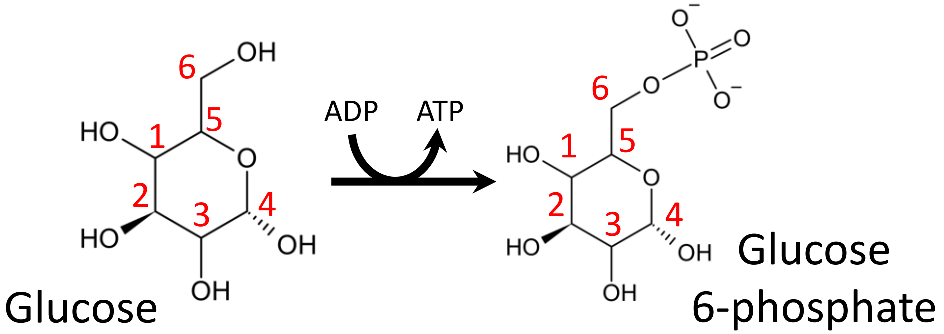




$$C_{123} = A_{123}$$

$$\begin{bmatrix} C_{123,M+0} \\ C_{123,M+1} \\ C_{123,M+2} \\ C_{123,M+3} \end{bmatrix} = \begin{bmatrix} A_{123,M+0} \\ A_{123,M+1} \\ A_{123,M+2} \\ A_{123,M+3} \end{bmatrix}$$

## C) Unimolecular reaction

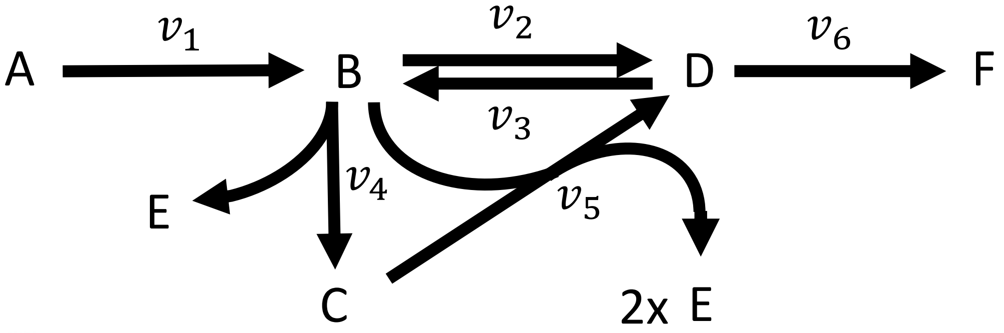



$$C_{123} = A_{123}$$

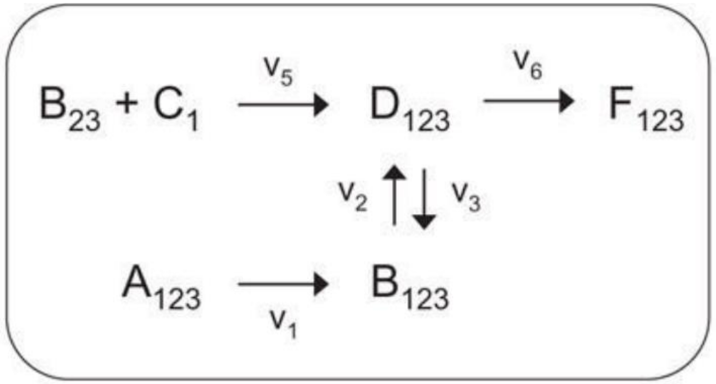
$$\begin{bmatrix} C_{123,M+0} \\ C_{123,M+1} \\ C_{123,M+2} \\ C_{123,M+3} \end{bmatrix} = \begin{bmatrix} A_{123,M+0} \\ A_{123,M+1} \\ A_{123,M+2} \\ A_{123,M+3} \end{bmatrix}$$

# EMU Decomposition



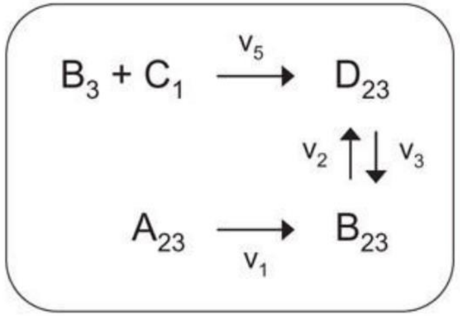

EMU size 3

$D_{123} \rightarrow F_{123}$
$B_{123} \rightarrow D_{123}$
$B_{23} + C_1 \rightarrow D_{123}$
$A_{123} \rightarrow B_{123}$
$D_{123} \rightarrow B_{123}$




EMU size 2

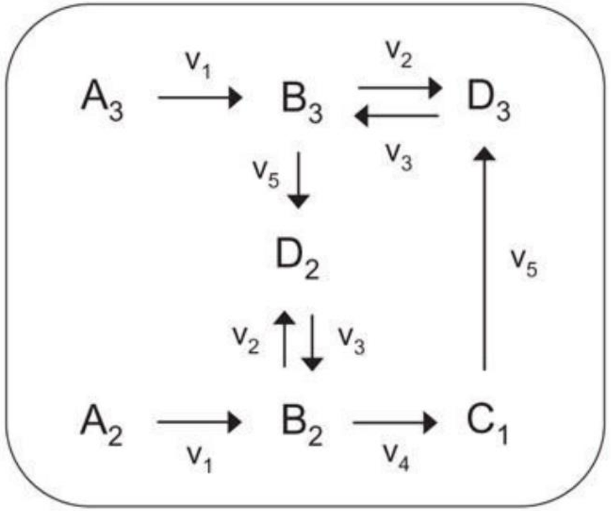
$A_{23} \rightarrow B_{23}$
$D_{23} \rightarrow B_{23}$
$B_{23} \rightarrow D_{23}$
$B_3 + C_1 \rightarrow D_{23}$



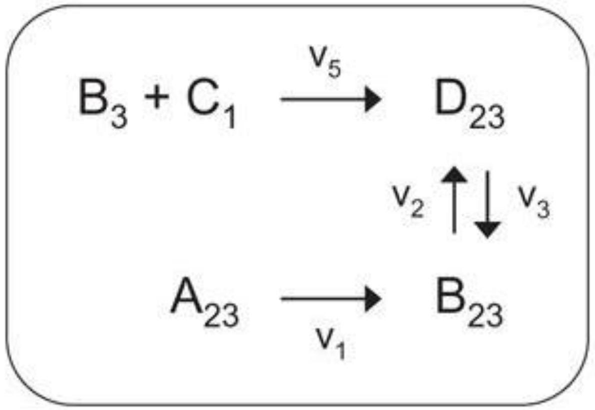

EMU size 1

$B_2 \rightarrow C_1$
$A_2 \rightarrow B_2$
$D_2 \rightarrow B_2$
$B_2 \rightarrow D_2$
$B_3 \rightarrow D_2$

$A_3 \rightarrow B_3$
$D_3 \rightarrow B_3$
$B_3 - D_3$
$C_1 \rightarrow D_3$



Reaction network for EMU size 2




$$\frac{d}{dt}D_{23} = v_5 \cdot (B_3 \times C_1) - v_3 \cdot D_{23} + v_2 \cdot B_{23} = 0$$

$$\frac{d}{dt}B_{23} = v_1 \cdot A_{23} \qquad + v_3 \cdot D_{23} - v_2 \cdot B_{23} = 0$$

$$- v_3 \cdot D_{23} + v_2 \cdot B_{23} = -v_5 \cdot (B_3 \times C_1)$$

$$+ v_3 \cdot D_{23} - v_2 \cdot B_{23} = -v_1 \cdot A_{23}$$

$$\begin{bmatrix} -v_3 & \overbrace{v_2}^{v_2 = v_1 + v_3 + 2v_4} \\ v_3 & -v_2 \end{bmatrix} \underbrace{\begin{bmatrix} D_{23} \\ B_{23} \end{bmatrix}}_{\text{Unknown Components}} = \begin{bmatrix} -v_5 & 0 \\ 0 & -v_1 \end{bmatrix} \underbrace{\begin{bmatrix} (B_3 \times C_1) \\ A_{23} \end{bmatrix}}_{\text{Known Components}}$$

$$(B_3 \times C_1) = [B_{3,M+0} \quad B_{3,M+1}] \times [C_{1,M+0} \quad C_{1,M+1}]$$
$$= [\underbrace{B_{3,M+0} \cdot C_{1,M+0}}_{\text{New M+0 term}} \quad \underbrace{(B_{3,M+0} \cdot C_{1,M+1} + B_{3,M+1} \cdot C_{1,M+0})}_{\text{New M+1 term}} \quad \underbrace{B_{3,M+1} \cdot C_{1,M+1}}_{\text{New M+2 term}}]$$

Discrete Convolution (Cauchy product )

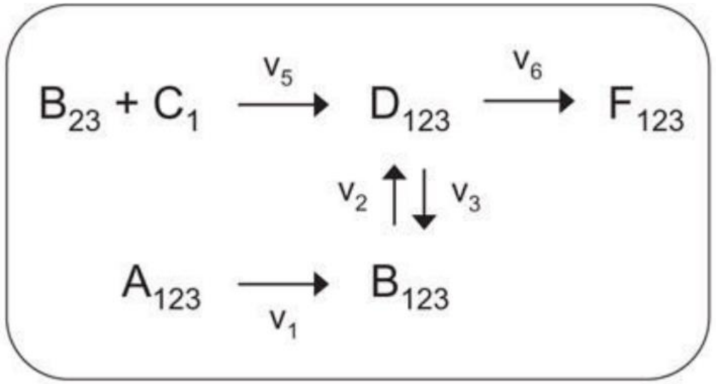
$$\begin{bmatrix} D_{23} \\ B_{23} \end{bmatrix} = \begin{bmatrix} D_{23,M+0} & D_{23,M+1} & D_{23,M+2} \\ B_{23,M+0} & B_{23,M+1} & B_{23,M+2} \end{bmatrix}$$

$$\begin{bmatrix} -v_3 & v_1 + v_3 \\ v_3 & -v_1 - v_3 \end{bmatrix} \begin{bmatrix} D_{23} \\ B_{23} \end{bmatrix} = \begin{bmatrix} -v_5 & 0 \\ 0 & -v_1 \end{bmatrix} \begin{bmatrix} (B_3 \times C_1) \\ A_{23} \end{bmatrix}$$

$$\frac{d}{dt}F_{123} = 0 \cdot (B_{23} \times C_1) + 0 \cdot B_{123} + v_6 \cdot D_{123} = 0$$

$$\frac{d}{dt}D_{123} = v_5 \cdot (B_{23} \times C_1) + v_2 \cdot B_{123} - (v_3 + v_6) \cdot D_{123} = 0$$

$$\frac{d}{dt}B_{123} = v_1 \cdot A_{123} - v_2 \cdot B_{123} + v_3 \cdot D_{123} = 0$$




$$0 \cdot B_{123} + v_6 \cdot D_{123} = 0 \cdot (B_{23} \times C_1)$$

$$v_2 \cdot B_{123} - (v_3 + v_6) \cdot D_{123} = -v_5 \cdot (B_{23} \times C_1)$$

$$v_2 \cdot B_{123} + v_3 \cdot D_{123} = -v_1 \cdot A_{123}$$

$$\begin{bmatrix} 0 & v_6 & 0 \\ 0 & -v_3 - v_6 & v_2 \\ 0 & v_3 & v_2 \end{bmatrix} \begin{bmatrix} F_{123} \\ D_{123} \\ B_{123} \end{bmatrix} = \begin{bmatrix} 0 & 0 \\ -v_5 & 0 \\ 0 & -v_1 \end{bmatrix} \begin{bmatrix} (B_{23} \times C_1) \\ A_{123} \end{bmatrix}$$

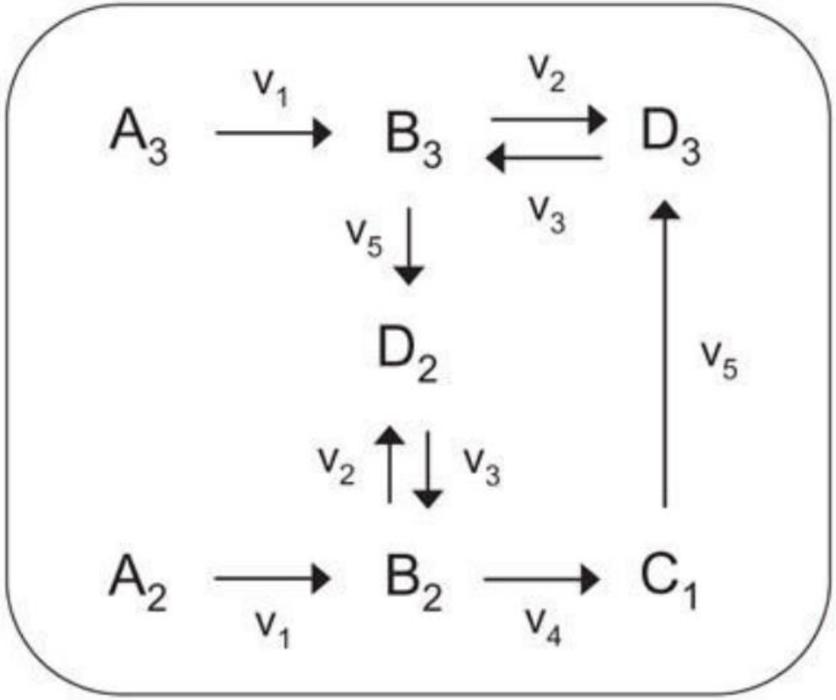
$$\frac{d}{dt}C_1 = v_4 \cdot B_2 - v_5 \cdot C_1 = 0$$

$$\frac{d}{dt}B_2 = v_1 \cdot A_2 + v_3 \cdot D_2 - (v_2 + v_4) \cdot B_2 = 0$$

$$\frac{d}{dt}D_2 = v_5 \cdot B_3 + v_2 \cdot B_2 - v_3 \cdot D_2 = 0$$

$$\frac{d}{dt}B_3 = v_1 \cdot A_3 + v_3 \cdot D_3 - (v_2 + v_5) \cdot B_3 = 0$$

$$\frac{d}{dt}D_3 = v_2 \cdot B_3 + v_5 \cdot C_1 - v_2 \cdot D_3 = 0$$




$$\begin{bmatrix} -v_5 & v_4 & 0 & 0 & 0 \\ 0 & -v_2 - v_4 & v_3 & 0 & 0 \\ 0 & v_2 & -v_3 & v_5 & 0 \\ 0 & 0 & 0 & -v_2 - v_5 & v_3 \\ v_5 & 0 & 0 & v_2 & -v_2 \end{bmatrix} \begin{bmatrix} C_1 \\ B_2 \\ D_2 \\ B_3 \\ D_3 \end{bmatrix} = \begin{bmatrix} 0 & 0 \\ -v_1 & 0 \\ 0 & 0 \\ 0 & -v_1 \\ 0 & 0 \end{bmatrix} \begin{bmatrix} A_2 \\ A_3 \end{bmatrix}$$

EMU size 1

$$\begin{bmatrix} -v_5 & v_4 & 0 & 0 & 0 \\ 0 & -v_2 - v_4 & v_3 & 0 & 0 \\ 0 & v_2 & -v_3 & v_5 & 0 \\ 0 & 0 & 0 & -v_2 - v_5 & v_3 \\ v_5 & 0 & 0 & v_2 & -v_2 \end{bmatrix} \begin{bmatrix} C_1 \\ B_2 \\ D_2 \\ B_3 \\ D_3 \end{bmatrix} = \begin{bmatrix} 0 & 0 \\ -v_1 & 0 \\ 0 & 0 \\ 0 & -v_1 \\ 0 & 0 \end{bmatrix} \begin{bmatrix} A_2 \\ A_3 \end{bmatrix}$$

$$A_{1,1}(v) \quad X_{1,1} = B_{1,1}(v) \quad Y_{1,1}(y_1^{in})$$

EMU size 2

$$\begin{bmatrix} -v_5 - v_2 & v_2 \\ v_3 & -v_1 - v_3 \end{bmatrix} \begin{bmatrix} D_{23} \\ B_{23} \end{bmatrix} = \begin{bmatrix} -v_5 & 0 \\ 0 & -v_1 \end{bmatrix} \begin{bmatrix} (B_3 \times C_1) \\ A_{23} \end{bmatrix}$$

$$A_{2,1}(v) \quad X_{2,1} = B_{2,1}(v) \quad Y_{2,1}(y_2^{in}, X_{1,1})$$

EMU size 3

$$\begin{bmatrix} 0 & v_6 & 0 \\ 0 & -v_3 - v_6 & v_2 \\ 0 & v_3 & v_2 \end{bmatrix} \begin{bmatrix} F_{123} \\ D_{123} \\ B_{123} \end{bmatrix} = \begin{bmatrix} 0 & 0 \\ -v_5 & 0 \\ 0 & -v_1 \end{bmatrix} \begin{bmatrix} (B_{23} \times C_1) \\ A_{123} \end{bmatrix}$$

$$A_{3,1}(v) \quad X_{3,1} = B_{3,1}(v) \quad Y_{3,1}(y_3^{in}, X_{2,1}, X_{1,1})$$

$$A_{n,k}(v) X_{n,k} = B_{n,k}(v) Y_{n,k}\left(y_n^{in}, X_{n-1}, \dots, X_1\right) \rightarrow X_i = A_i^{-1} B_i Y_i \qquad \forall\, i = 1 \dots n, \; k = 1$$

# FBA

Flux balance analysis

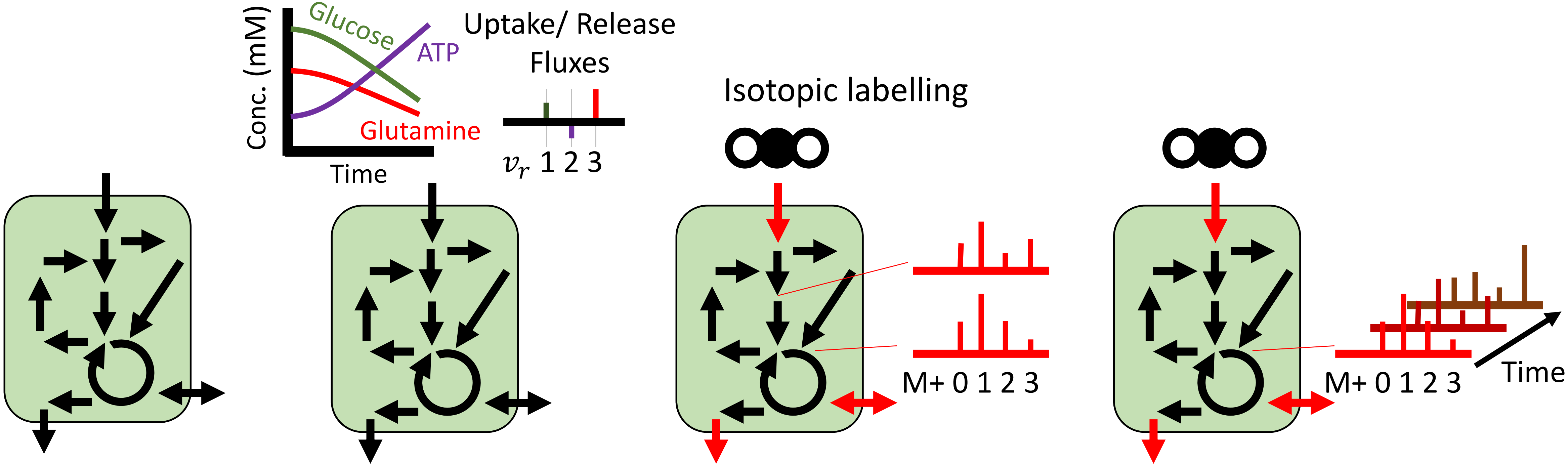
# MFA

Metabolic flux analysis

# $^{13}$C-MFA

# $^{13}$C-INST MFA

Isotopically Nonstationary Metabolic flux analysis




**FBA:**

$$\max V_{growth}$$
$$s.t.\ S * \bar{v} = 0$$
$$lb < \bar{v} < ub$$

**MFA:**

$$\min \sum (v_r - \hat{v}_r)$$
$$s.t.\ S * \bar{v} = 0$$
$$\bar{v} \geq 0$$

**$^{13}$C-MFA:**

$$\min \sum (x_i(\bar{v}) - \hat{x}_i)$$
$$s.t.\ S * \bar{v} = 0$$
$$\bar{v} \geq 0$$

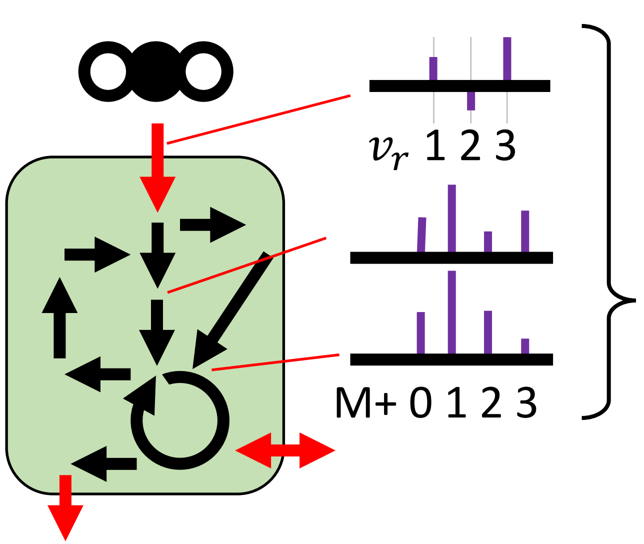
**$^{13}$C-INST MFA:**

$$\min \sum\sum (x_{i,t}(\bar{v}) - \hat{x}_{i,t})$$
$$s.t.\ S * \bar{v} = 0$$
$$\bar{v} \geq 0$$

Large scale models (genome-scale)

Small scale models (Core metabolism)

Medium scale models (Metabolomics)




Experimental Data

$$\hat{x} = \begin{bmatrix} vec(\hat{X}_{1,1}^T) \\ \vdots \\ vec(\hat{X}_{n,k}^T) \\ \hat{v}_r \end{bmatrix}$$

Model

$$x = \begin{bmatrix} vec(X_{1,1}^T) \\ \vdots \\ vec(X_{1,1}^T) \\ v_r \end{bmatrix}$$

$$X_{n,k} = A_{n,k}^{-1}(\bar{v}) B_{n,k}(\bar{v}) Y_{n,k}$$

$$\bar{v} = N * \bar{u}$$

$$if \;\; X = \begin{bmatrix} a & b \\ c & d \end{bmatrix} \; then \; vec(X) = \begin{bmatrix} a \\ c \\ b \\ d \end{bmatrix}$$

$$\min \sum (x_i(\bar{v}) - \hat{x}_i)$$
$$s.t. \; S * \bar{v} = 0$$
$$\bar{v} \geq 0$$

$$\arg\min_{\bar{u}} \sum (x_i(\bar{u}) - \hat{x}_i)$$
$$s.t. \; \bar{v} \geq 0$$
$$c(x, \bar{v}) = 0$$

$$A_{n,k}(v) X_{n,k} - B_{n,k}(v) Y_{n,k}(y_n^{in}, X_{n-1}, \ldots, X_1) = 0$$ Isotopic steady state

$$S * \bar{v} = 0$$ Metabolic steady state

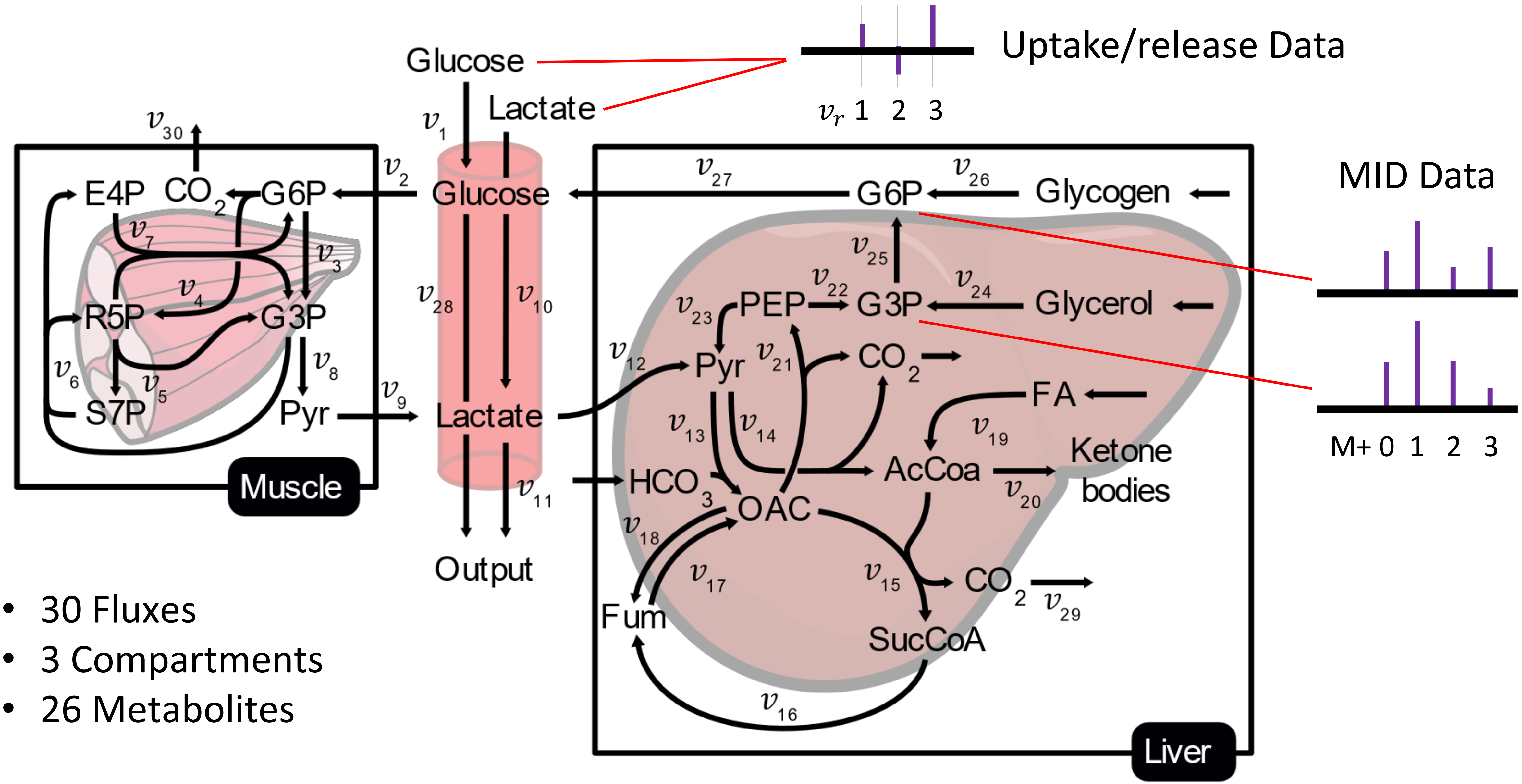
$\bar{v}$ – *flux vector*

$\bar{u}$ – *Independent flux vector*

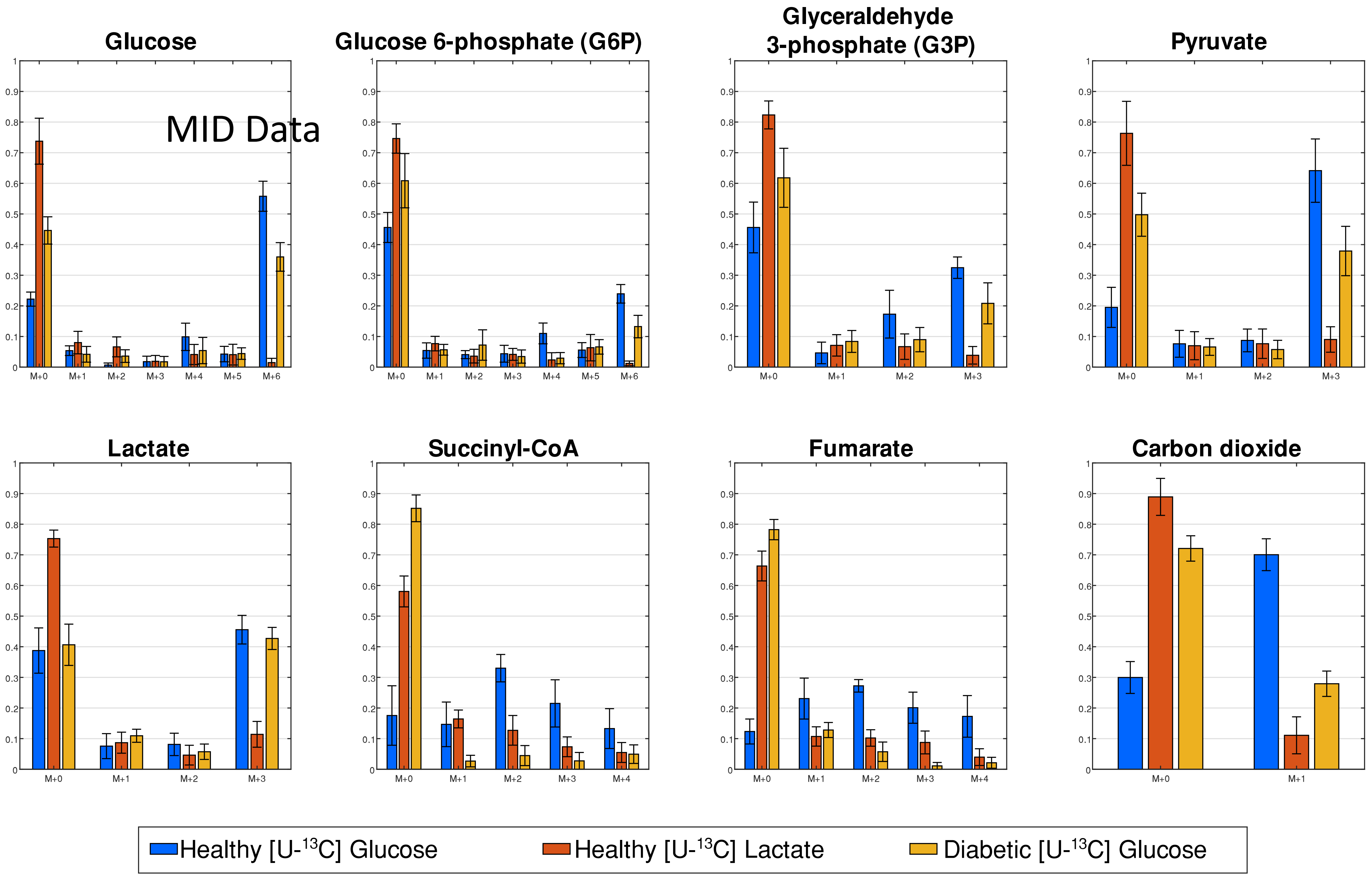
$S$ – *Stoichiometric matrix*

$N$ – *Null space matrix*

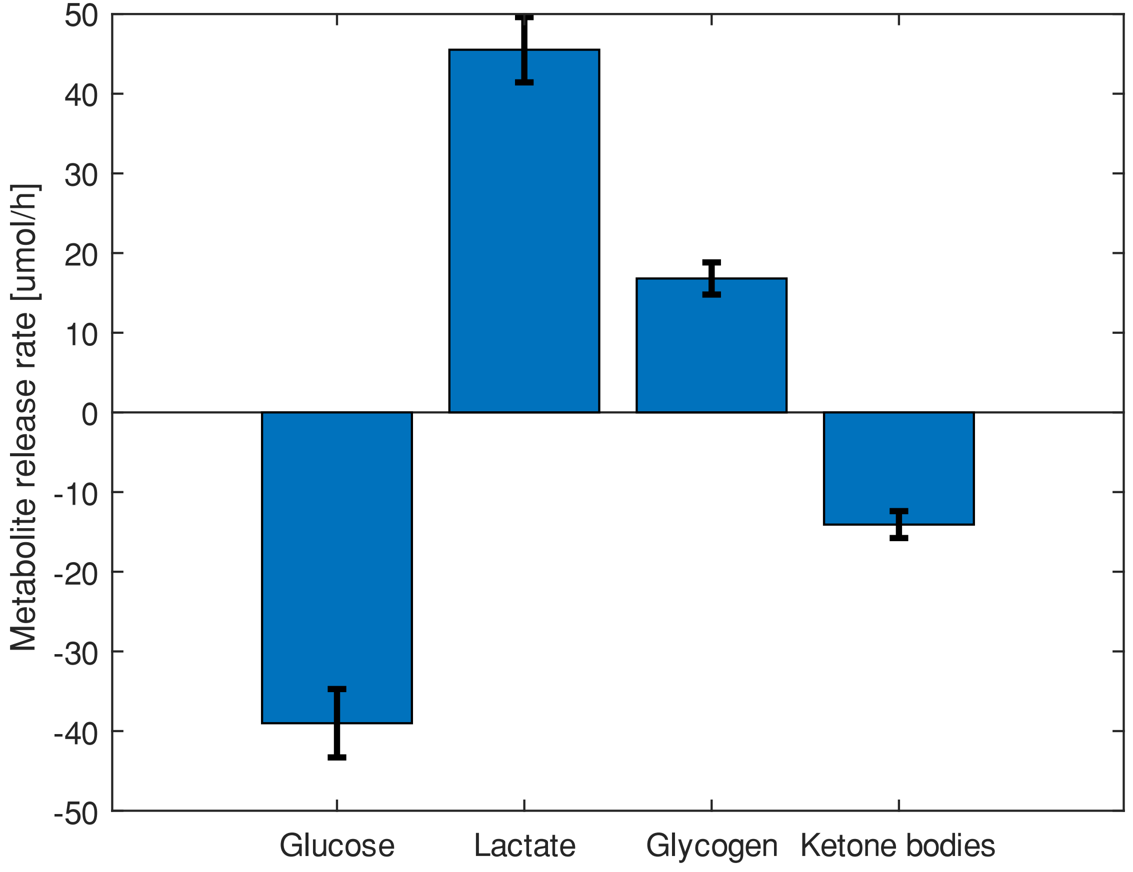
# Glucose metabolism Model

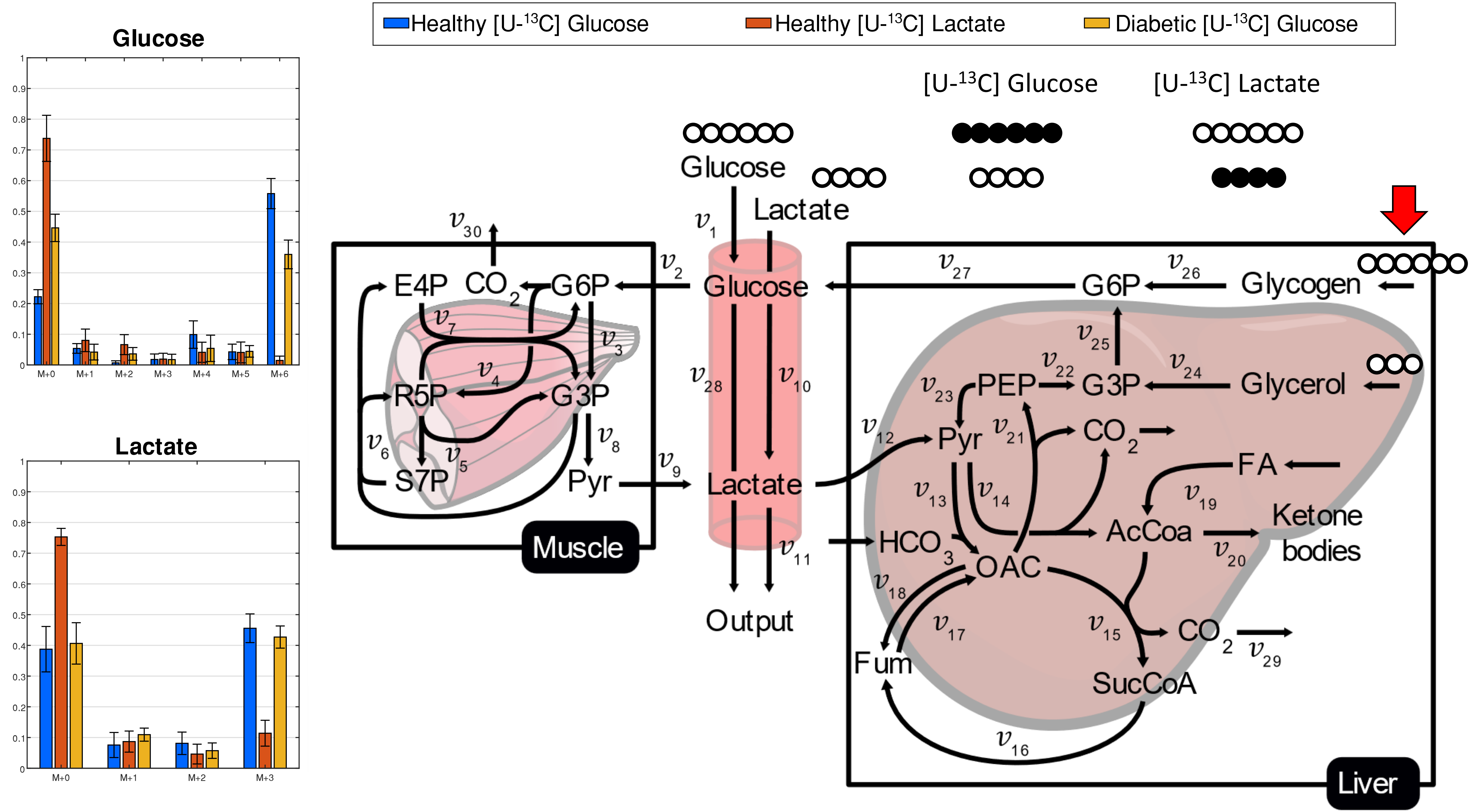



- 30 Fluxes
- 3 Compartments
- 26 Metabolites


MID Data
Glucose
M+0 M+1 M+2 M+3 M+4 M+5 M+6
Glucose 6-phosphate (G6P)
M+0 M+1 M+2 M+3 M+4 M+5 M+6
Glyceraldehyde 3-phosphate (G3P)
M+0 M+1 M+2 M+3
Pyruvate
M+0 M+1 M+2 M+3
Lactate
M+0 M+1 M+2 M+3
Succinyl-CoA
M+0 M+1 M+2 M+3 M+4
Fumarate
M+0 M+1 M+2 M+3 M+4
Carbon dioxide
M+0 M+1
0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9 1
Healthy [U-13C] Glucose
Healthy [U-13C] Lactate
Diabetic [U-13C] Glucose

# Uptake/release Data




Healthy [U-13C] Glucose
Healthy [U-13C] Lactate
Diabetic [U-13C] Glucose
Glucose
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1
M+0
M+1
M+2
M+3
M+4
M+5
M+6
Lactate
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1
M+0
M+1
M+2
M+3
[U-13C] Glucose
[U-13C] Lactate
Glucose
Lactate
Output
Muscle
E4P
CO2
G6P
R5P
G3P
S7P
Pyr
Liver
G6P
Glycogen
PEP
G3P
Glycerol
Pyr
CO2
FA
HCO3
OAC
AcCoa
Ketone bodies
CO2
Fum
SucCoA
v1
v2
v3
v4
v5
v6
v7
v8
v9
v10
v11
v12
v13
v14
v15
v16
v17
v18
v19
v20
v21
v22
v23
v24
v25
v26
v27
v28
v29
v30

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