Mathematical models of erythrocyte. What they give us for understanding the disorders and ageing of this cell

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In memory of Anatol Zhabotinsky

Questions:

- What is a disease from the mathematical point of view?
- Complex and simple models: how do they relate with each other?
- Why do so many cellular enzymes have excessively high activities?

Topics of this lecture:

- Red blood cell (RBC) – an overview
- Red blood cell – metabolism and viability; ageing
- A mathematical model
- Hereditary anemia due to enzyme deficiency: key and non-key enzymes
- Modeling of viability of the red blood cells with unstable mutant forms of enzymes


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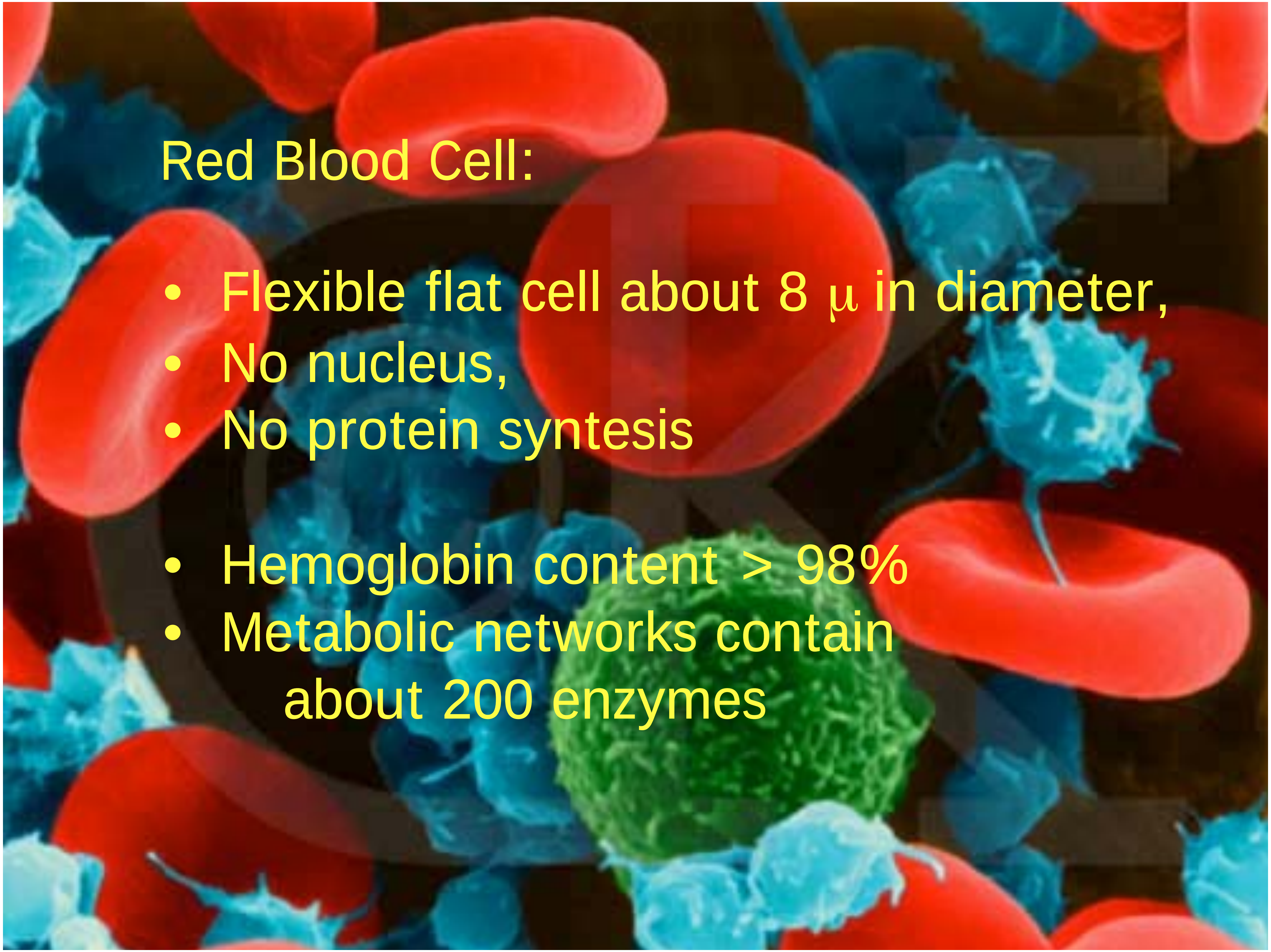
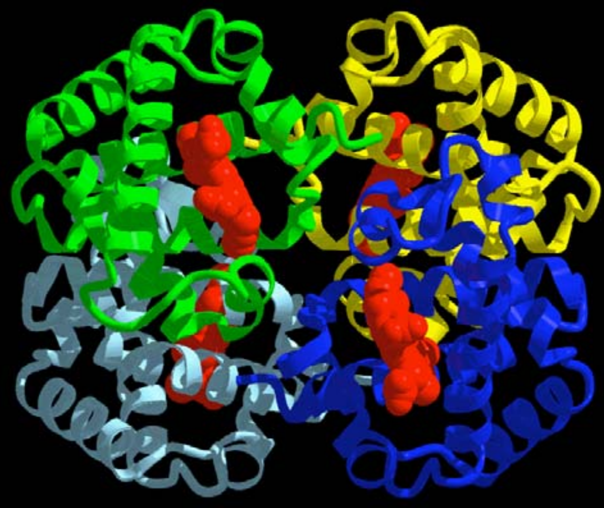


Red Blood Cell:

- Flexible flat cell about 8 μ in diameter,
- No nucleus,
- No protein syntesis

- Hemoglobin content > 98%
- Metabolic networks contain about 200 enzymes

Red Blood Cell:

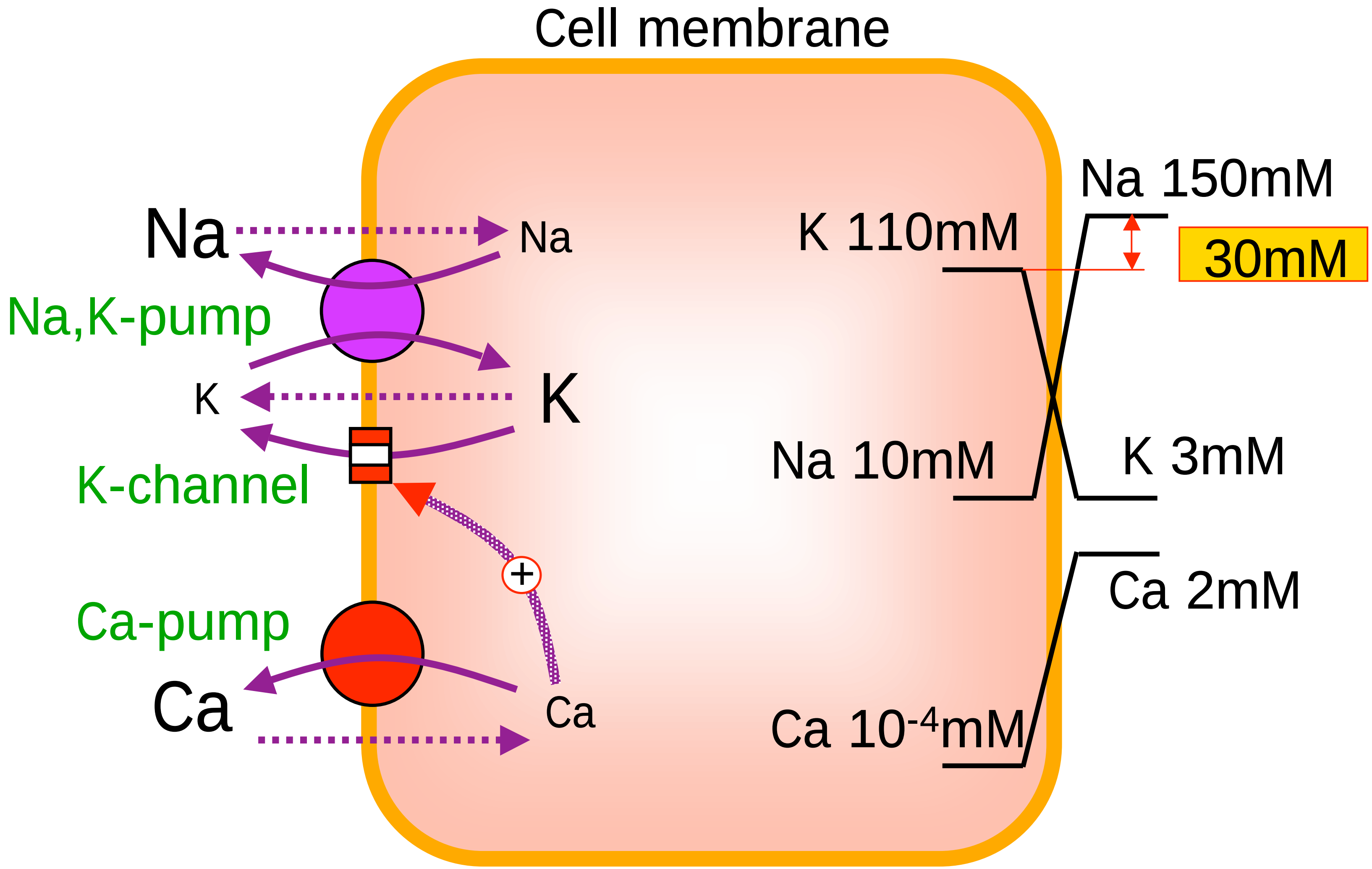


Hemoglobin content > 98%

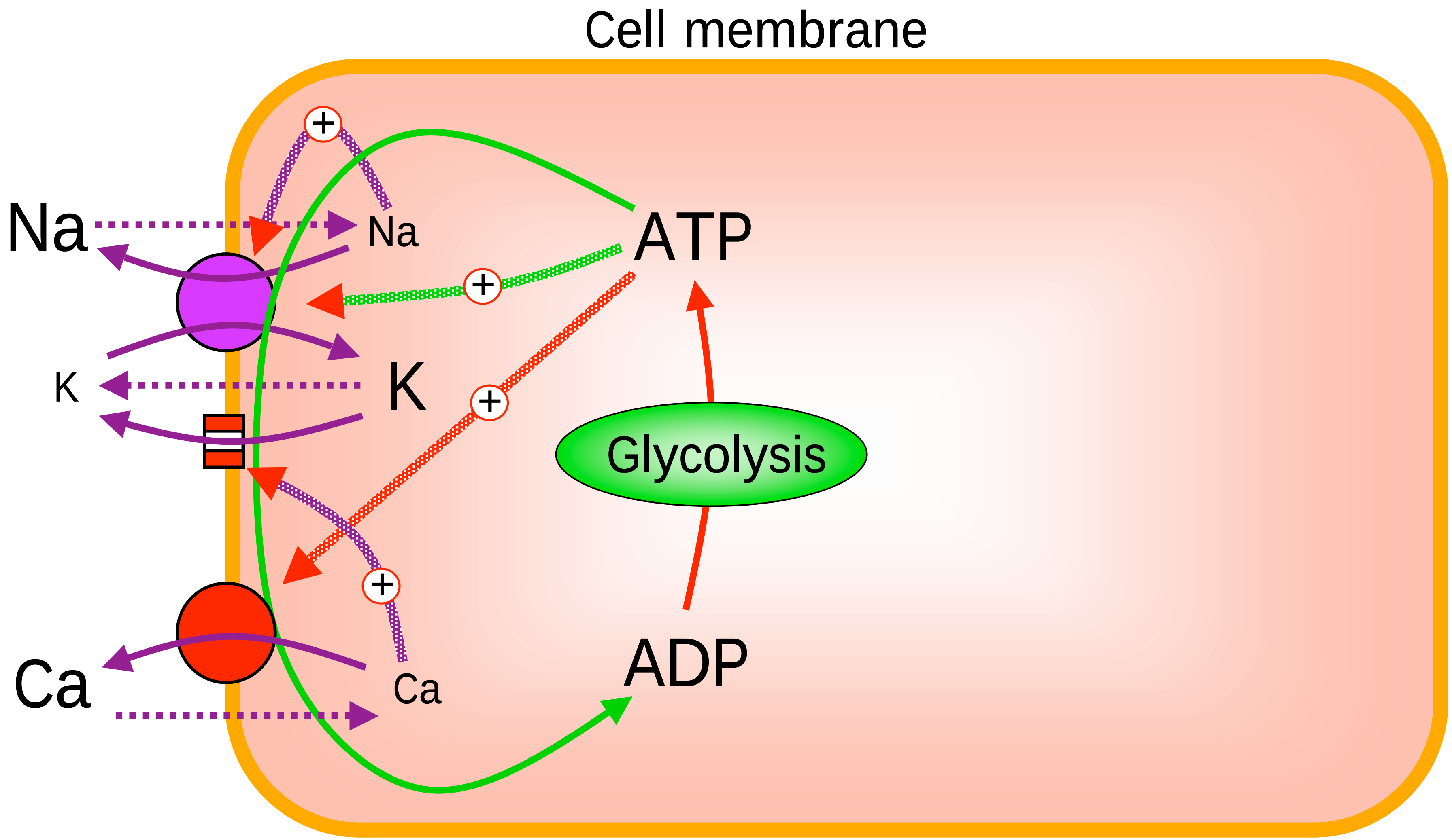
Redox control

Osmotic control -> volume stabilization

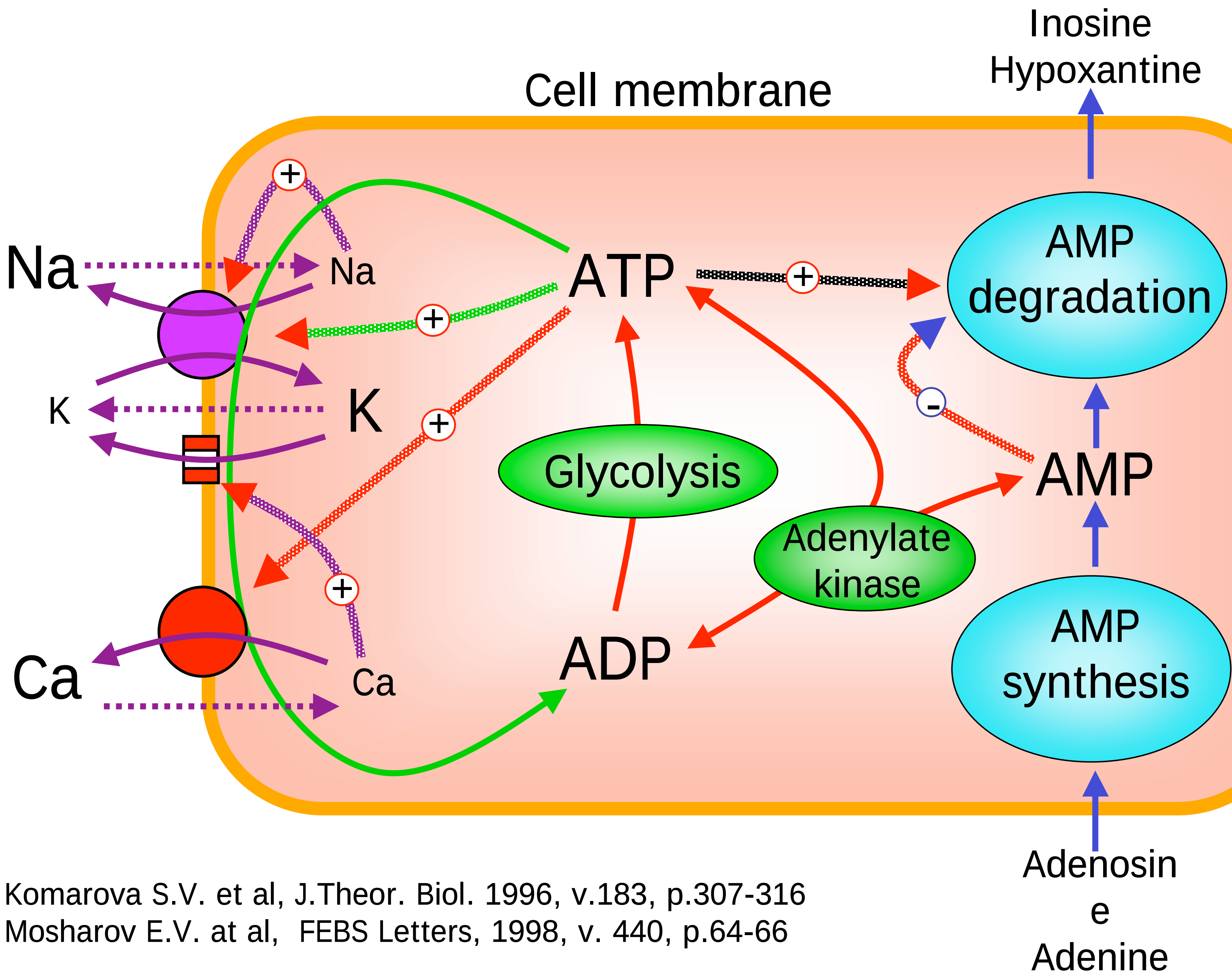
Red Blood Cell metabolism:



Red Blood Cell metabolism:

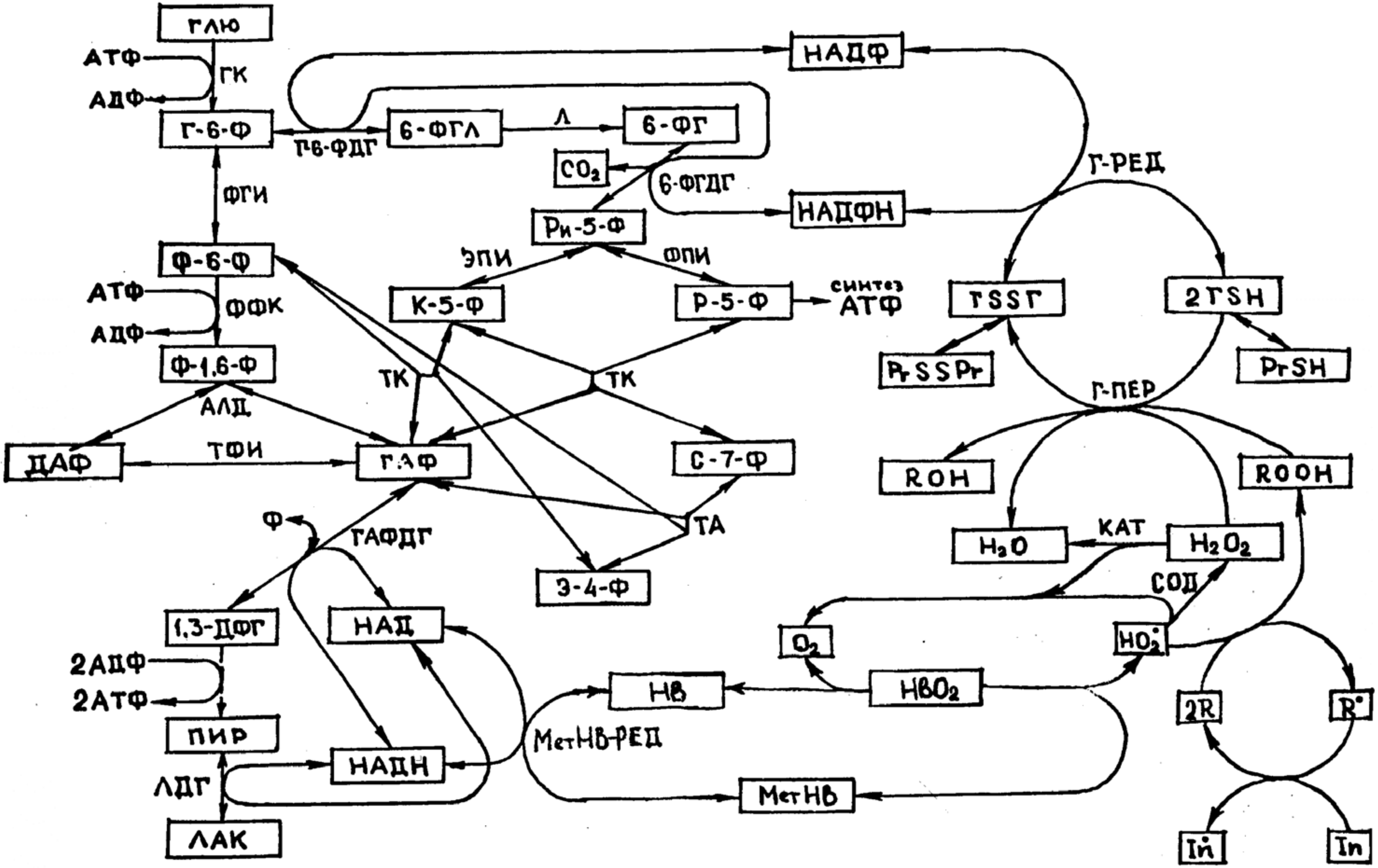


Red Blood Cell metabolism:



Komarova S.V. et al, J.Theor. Biol. 1996, v.183, p.307-316
Mosharov E.V. at al, FEBS Letters, 1998, v. 440, p.64-66

Red Blood Cell metabolism:



Osmotic equations:

$$\frac{[A_p^-]_e}{[A_p^-]_i} = \exp\left(-\frac{\psi F}{R\theta}\right)$$

$$[K^+]_i + [Na^+]_i - [A^-]_i + ZW = 0$$

$$[K^+]_i + [Na^+]_i + [A^-]_i + \Omega + W = [K^+]_e + [Na^+]_e + [A^-]_e$$
$$= 2L = 300 \text{ mM}$$

$P_K = 1.24\times10^{-2}$ 1/h; $P_{Na} = 1.22\times10^{-2}$ 1/h; $[K^+]_e = 5$ mM; $[Na^+]_e = 145$ mM; $[A^-]_e = 150$ mM

Osmotic equations:

$$\frac{d}{dt}\left([Na^+]_i \frac{V}{V^0}\right) = -3\upsilon_{Na,K-ATPase} + J_{Na};$$

$$J_{Na} = P_{Na}\frac{\frac{\psi F}{R\theta}}{\exp\left(\frac{\psi F}{R\theta}\right)-1}\left([Na^+]_e - [Na^+]_i \exp\left(\frac{\psi F}{R\theta}\right)\right)$$

$$\frac{d}{dt}\left([K^+]_i \frac{V}{V^0}\right) = \ldots$$

$$\frac{d}{dt}\left([Ca^{++}]_i \frac{V}{V^0}\right) = \ldots$$

Metabolic equations (examples):

$$\frac{d}{dt}\left([FDP]\frac{V}{V^0}\right) = \upsilon_{PFK} - \upsilon_{ALD}$$

$$\frac{d}{dt}\left([DAP]\frac{V}{V^0}\right) = \upsilon_{ALD} - \upsilon_{TPI}$$

$$\frac{d}{dt}\left([GAP]\frac{V}{V^0}\right) = \upsilon_{ALD} + \upsilon_{TPI} - \upsilon_{GAPDH}$$

....

Rates of enzymatic reactions (examples):

$$\upsilon_{GPI} = \alpha_{GPI} \frac{([G6P]-[F6P]K^1_{GPI})/K^2_{GPI}}{1+[G6P]/K^2_{GPI}+[F6P]/K^3_{GPI}}$$

α^0_{GPI} =360 mM/h, K^1_{GPI} =3, K^2_{GPI} =0.3 mM, K^3_{GPI} =0.2 mM.

$$\upsilon_{PFK} = \alpha_{PFK} \frac{1.1\cdot[ATP][F6P]}{\left(K^2_{PFK}+[ATP]\right)\left(K^1_{PFK}+[F6P]\right)} \frac{\left\lfloor 1/\left(1+[AMP]/K^3_{PFK}\right)+2[AMP]/(K^3_{PFK}+[AMP])\right\rfloor}{\left[1+10^8\frac{\left(1+[ATP]/K^4_{PFK}\right)^4}{\left(1+[AMP]/K^3_{PFK}\right)^4\left(1+[F6P]/K^5_{PFK}\right)^4}\right]}$$

α^0_{PFK} =380 mM/h, K^1_{PFK} =0.1 mM, K^2_{PFK} =2 mM,

K^3_{PFK} =10^{-2}mM, K^4_{PFK} =$19.5\cdot10^{-2}$mM,

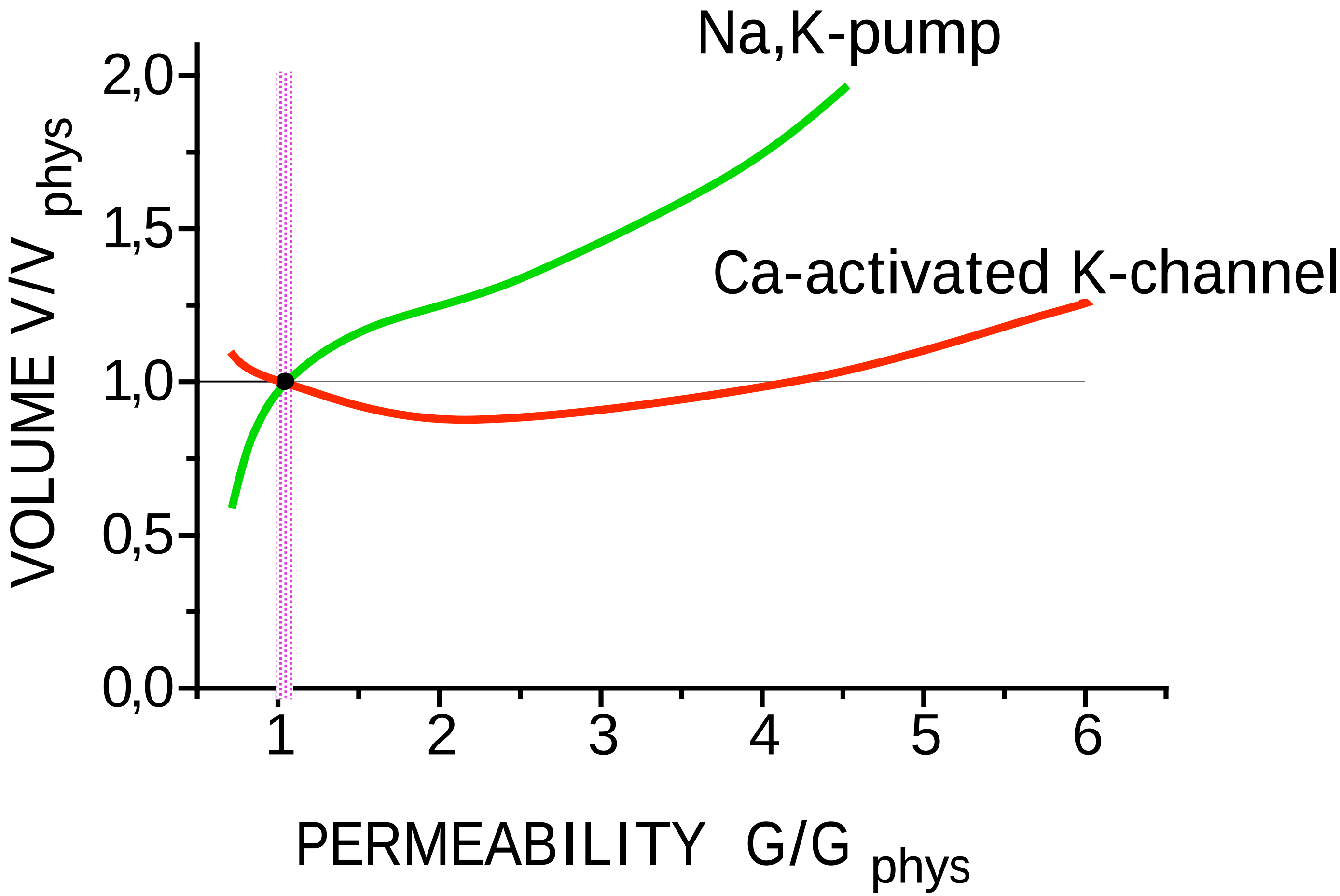
....

Anemia – low RBC content in the blood

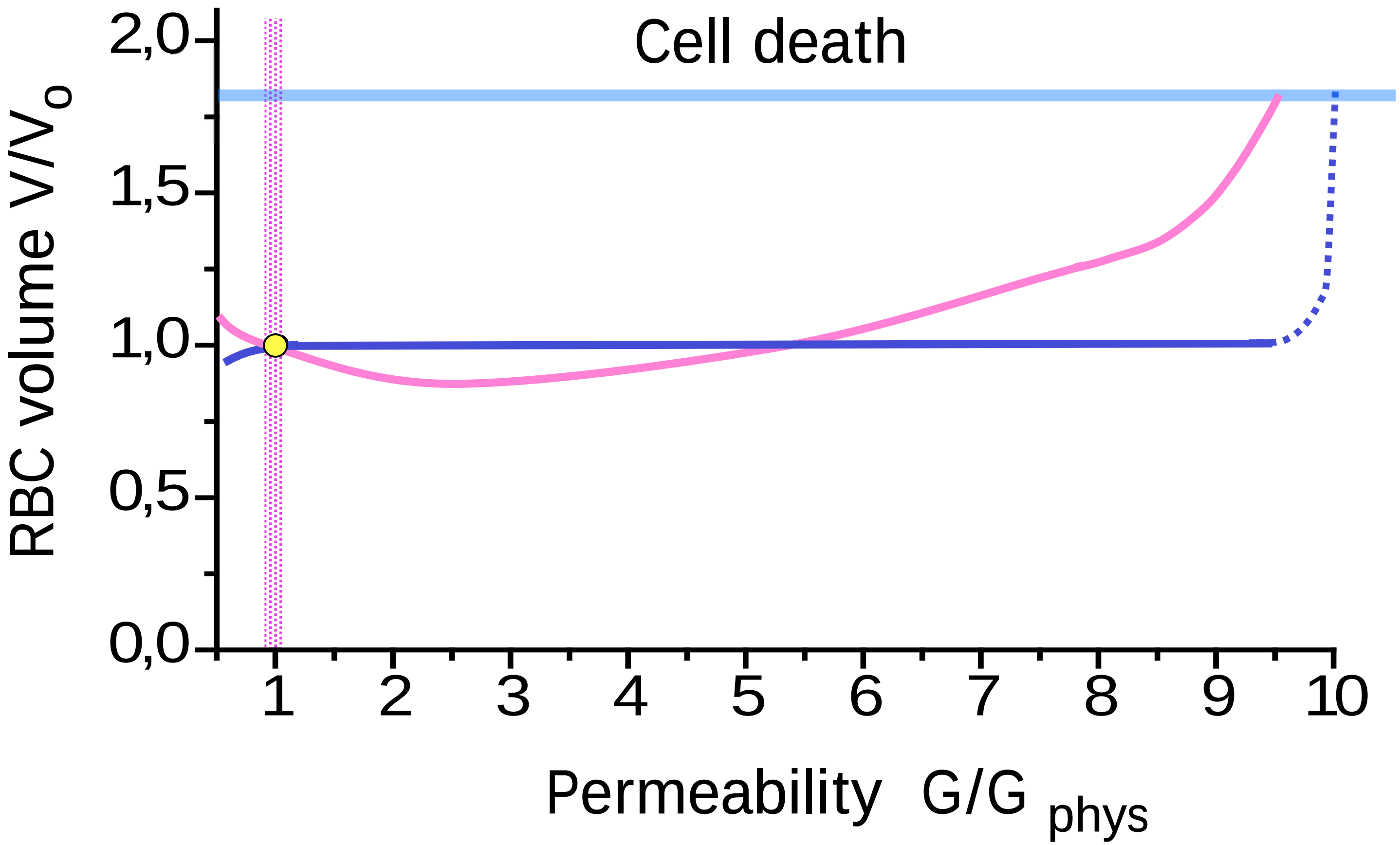
Red blood cell death caused mostly by osmotic swelling

Osmotic swelling caused by decrease of the enzyme activity

Hereditary anemia due to enzyme deficiency –>
caused by increased rate of a cell death

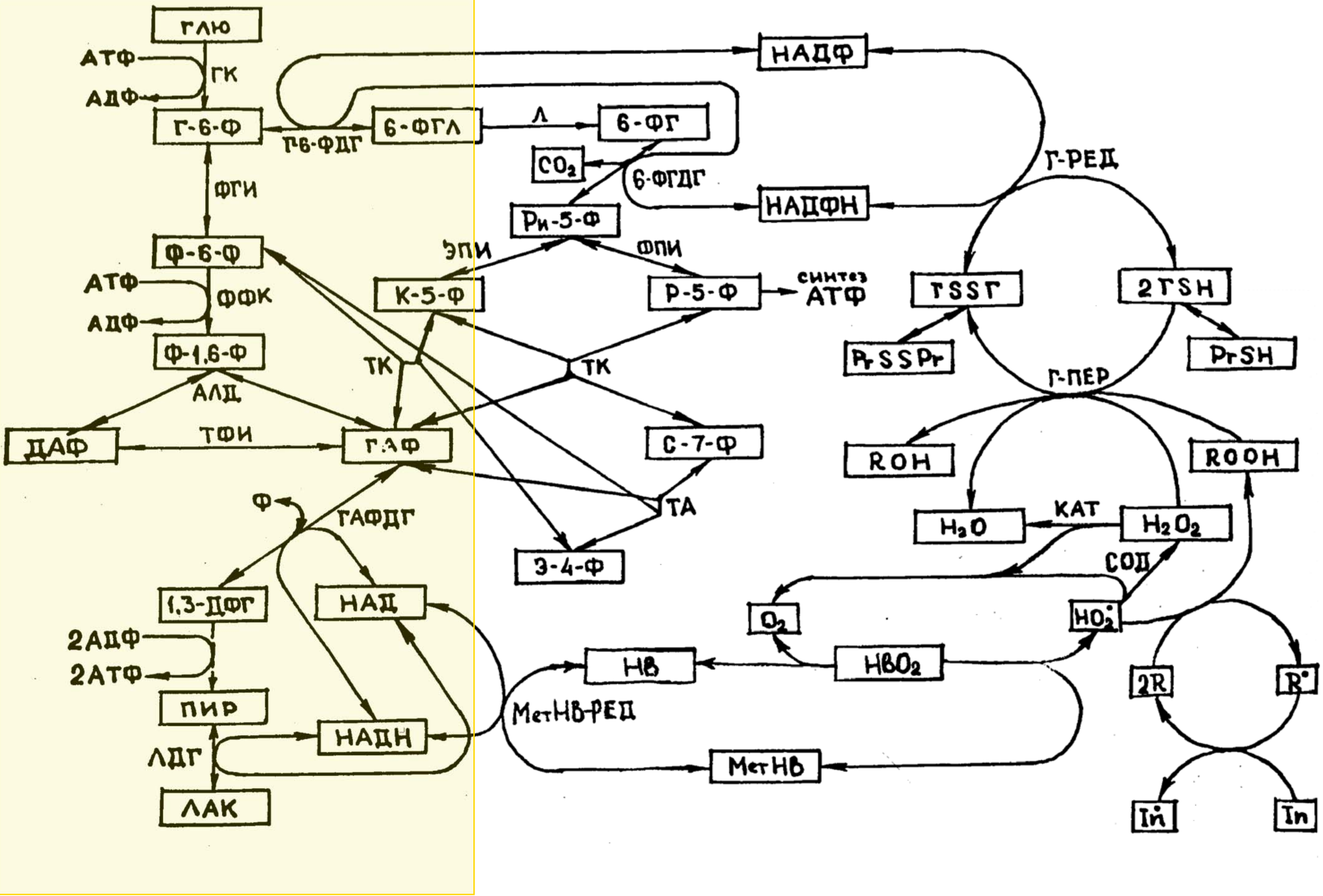


Martinov M. et al. Biophys Chem, 1999, v.80, p.199-215



Cell death
RBC volume V/V$_o$
2,0
1,5
1,0
0,5
0,0
1
2
3
4
5
6
7
8
9
10
Permeability G/G $_{phys}$

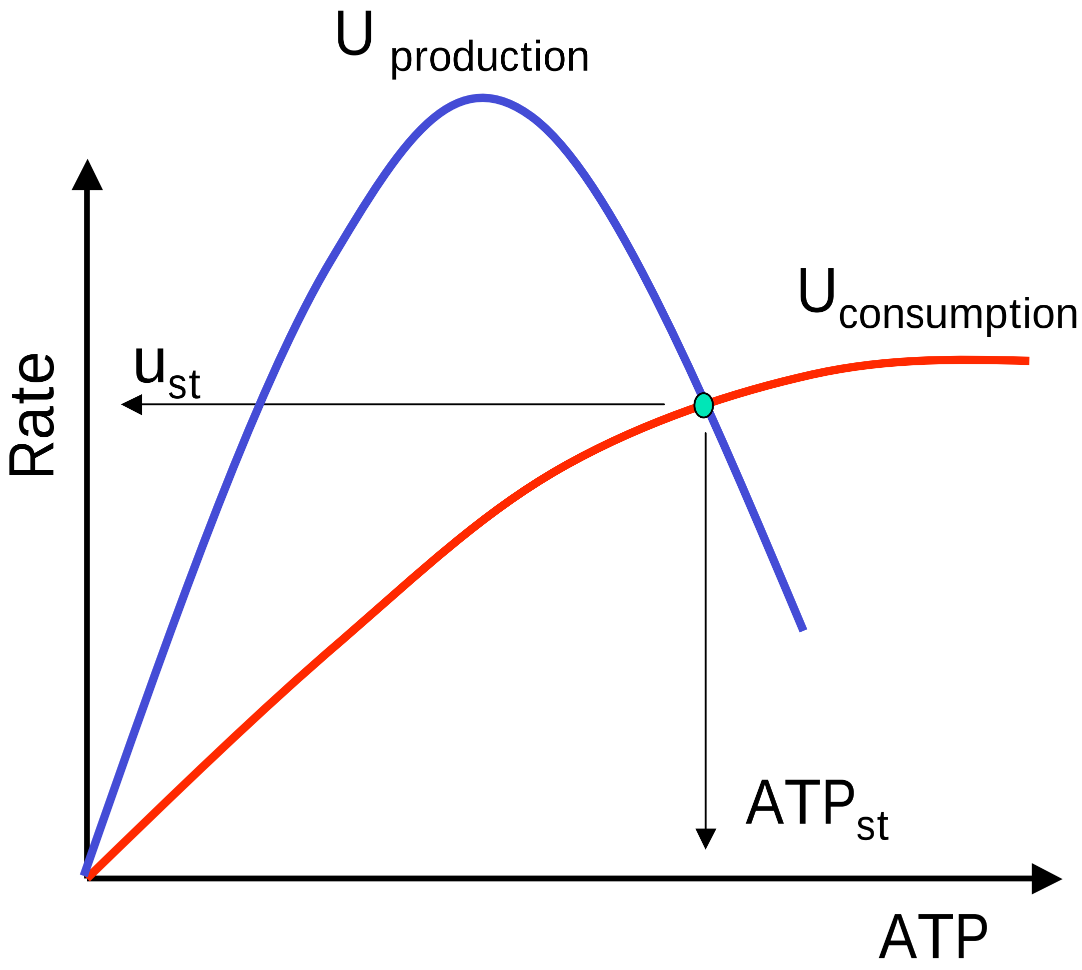
Red Blood Cell metabolism:

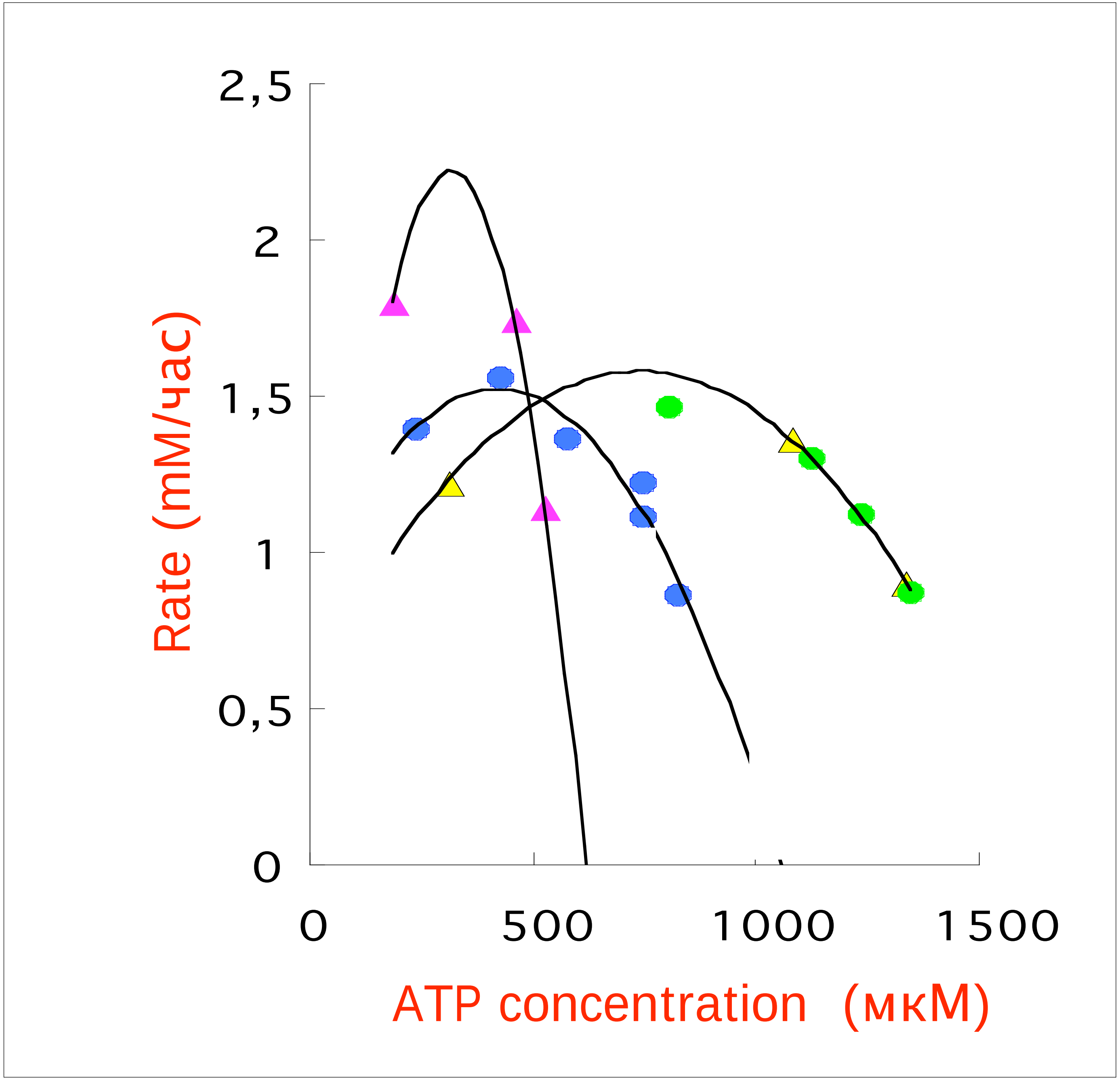


Steady-state fluxes should be equal

$$2u_1 = 2u_3 = u_7 = u_{10}$$

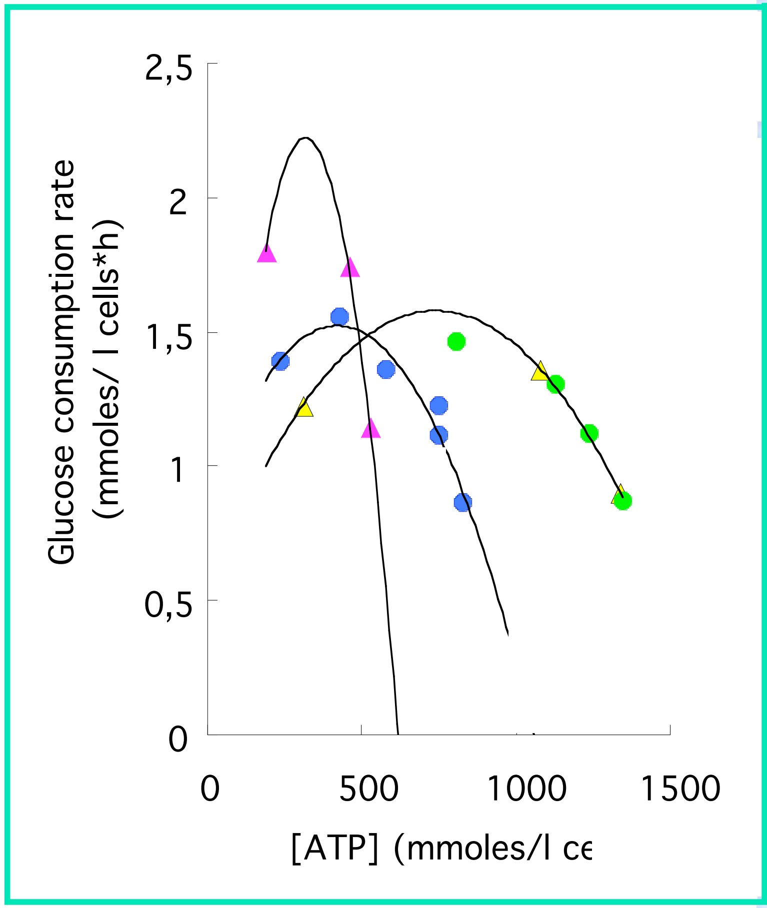
$$dATP/dt = 2u_1 - u_{consumption}$$


U production
U consumption
Rate
u st
ATP st
ATP

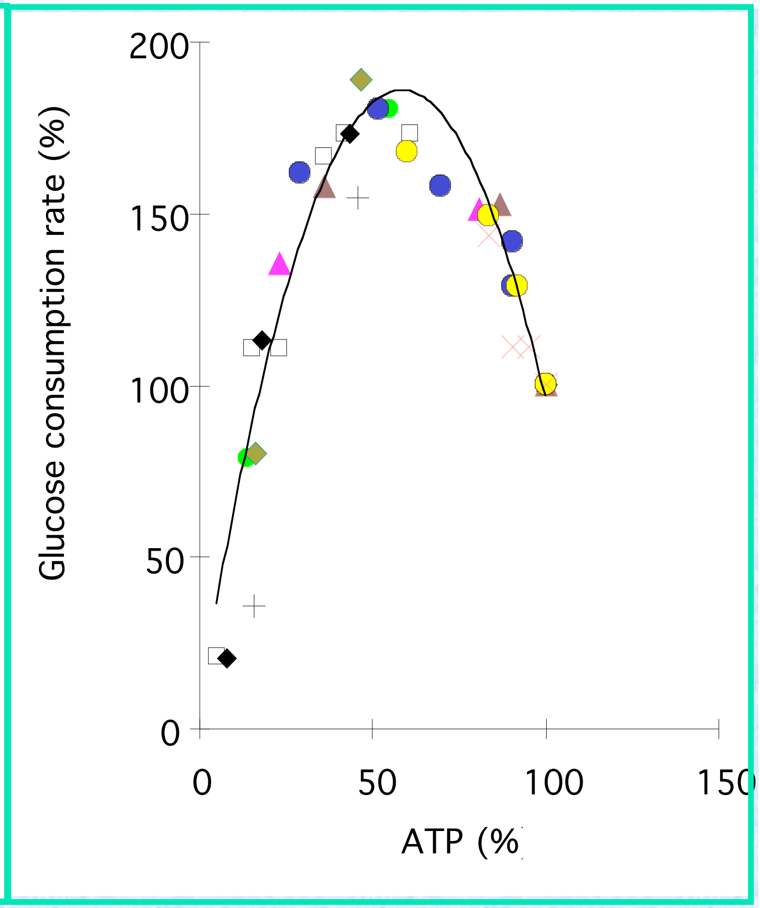


2,5
2
1,5
1
0,5
0
0
500
1000
1500
Rate (mM/час)
ATP concentration (мкМ)

Rate of glycolysis



ATP concentration

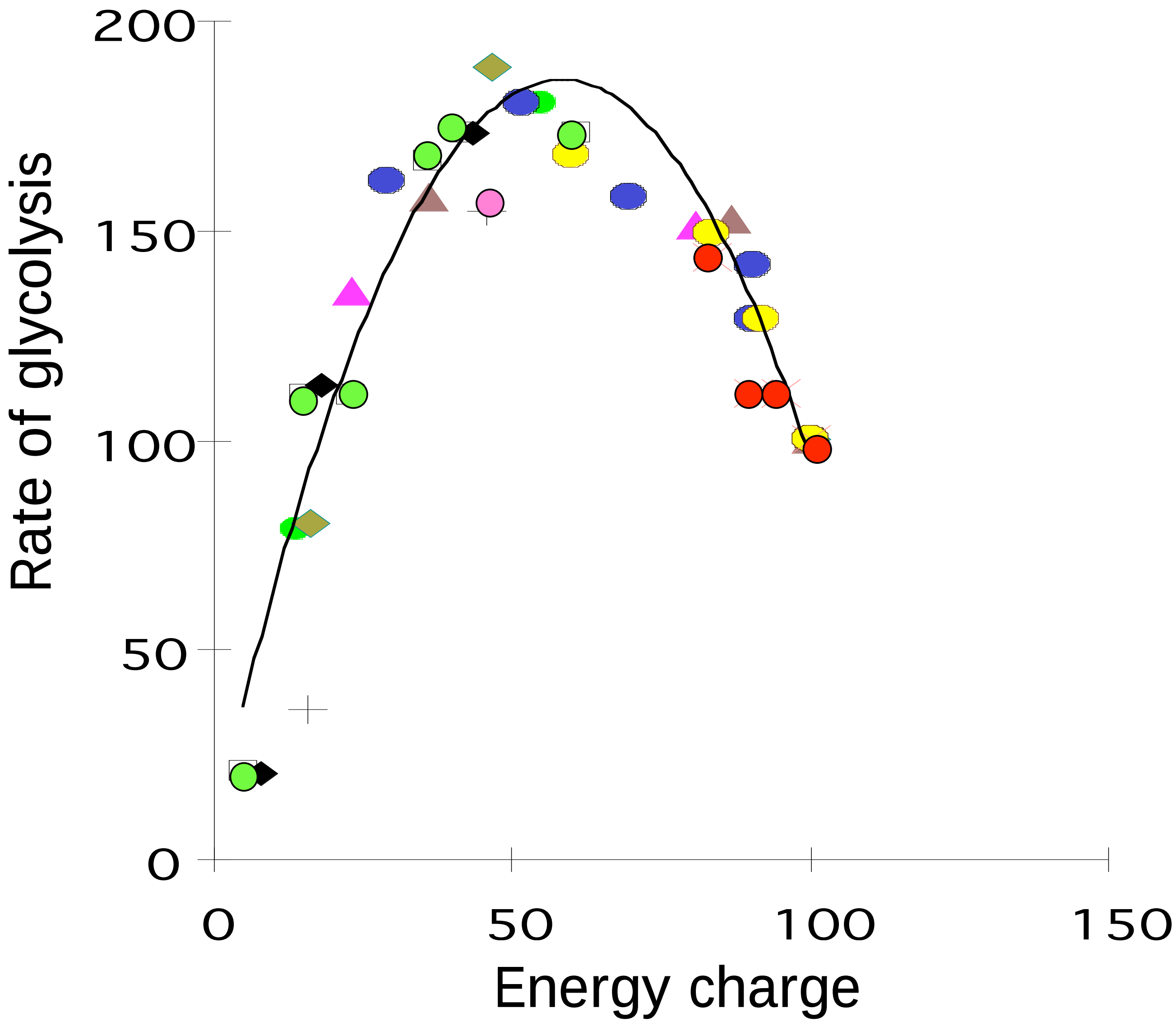


energy charge φ

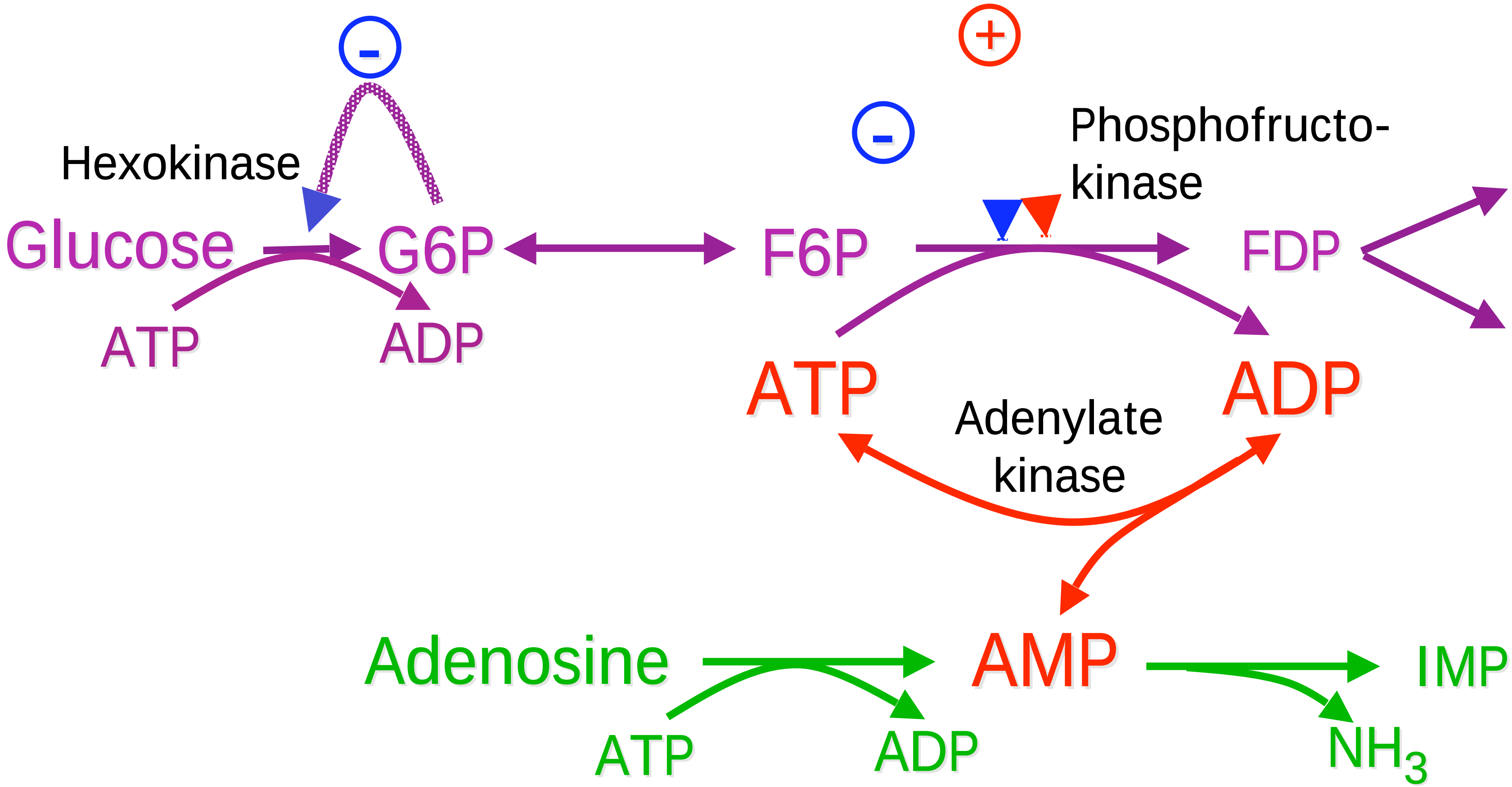
Ataullakhanov F. et al. Eur J Biochem., 1981, v.115, p.359-365

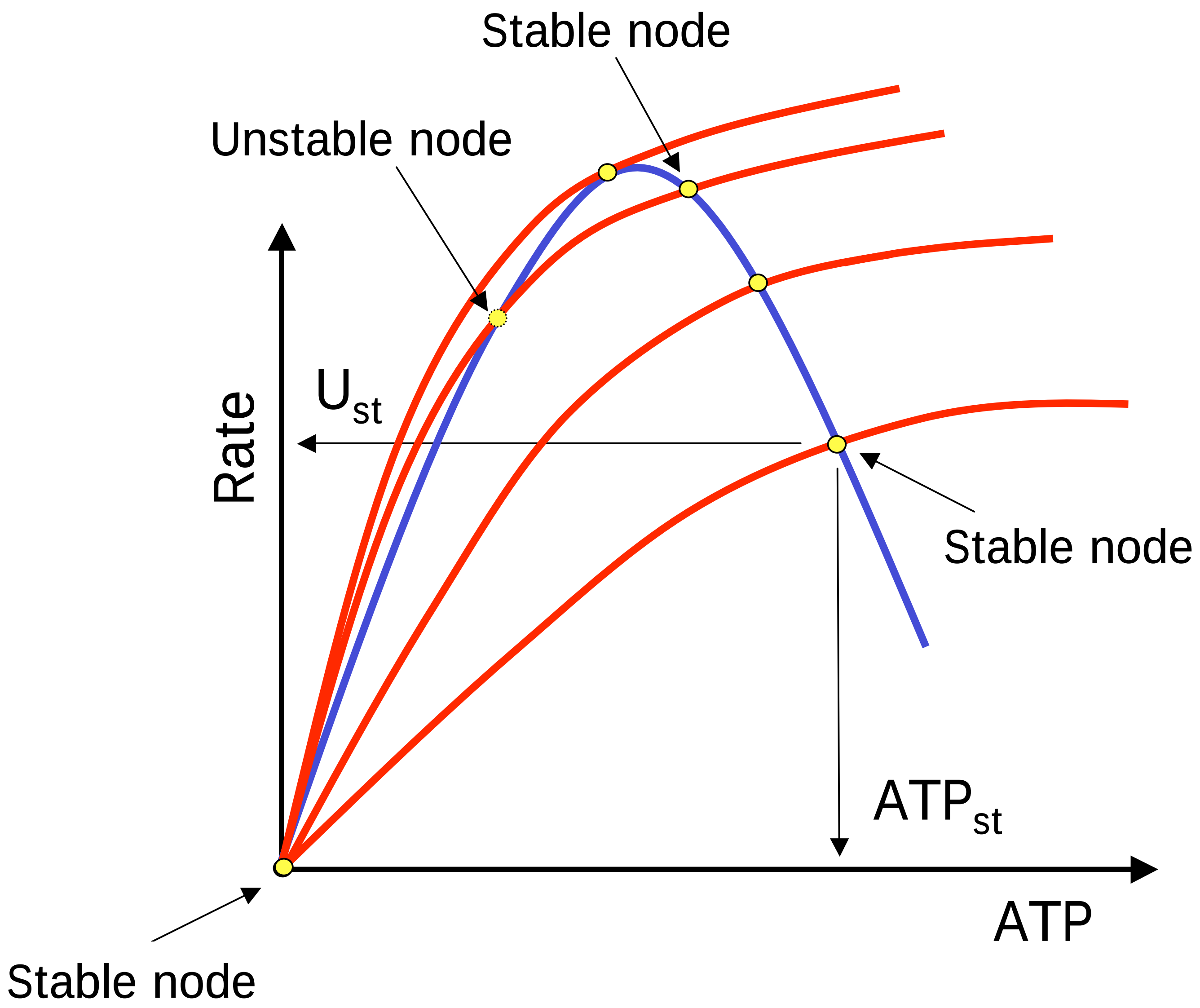
Energy charge is one of the few essential variables:

$$\varphi = \frac{\text{ATP} + 0.5\ \text{ADP}}{\text{ATP} + \text{ADP} + \text{AMP}}$$

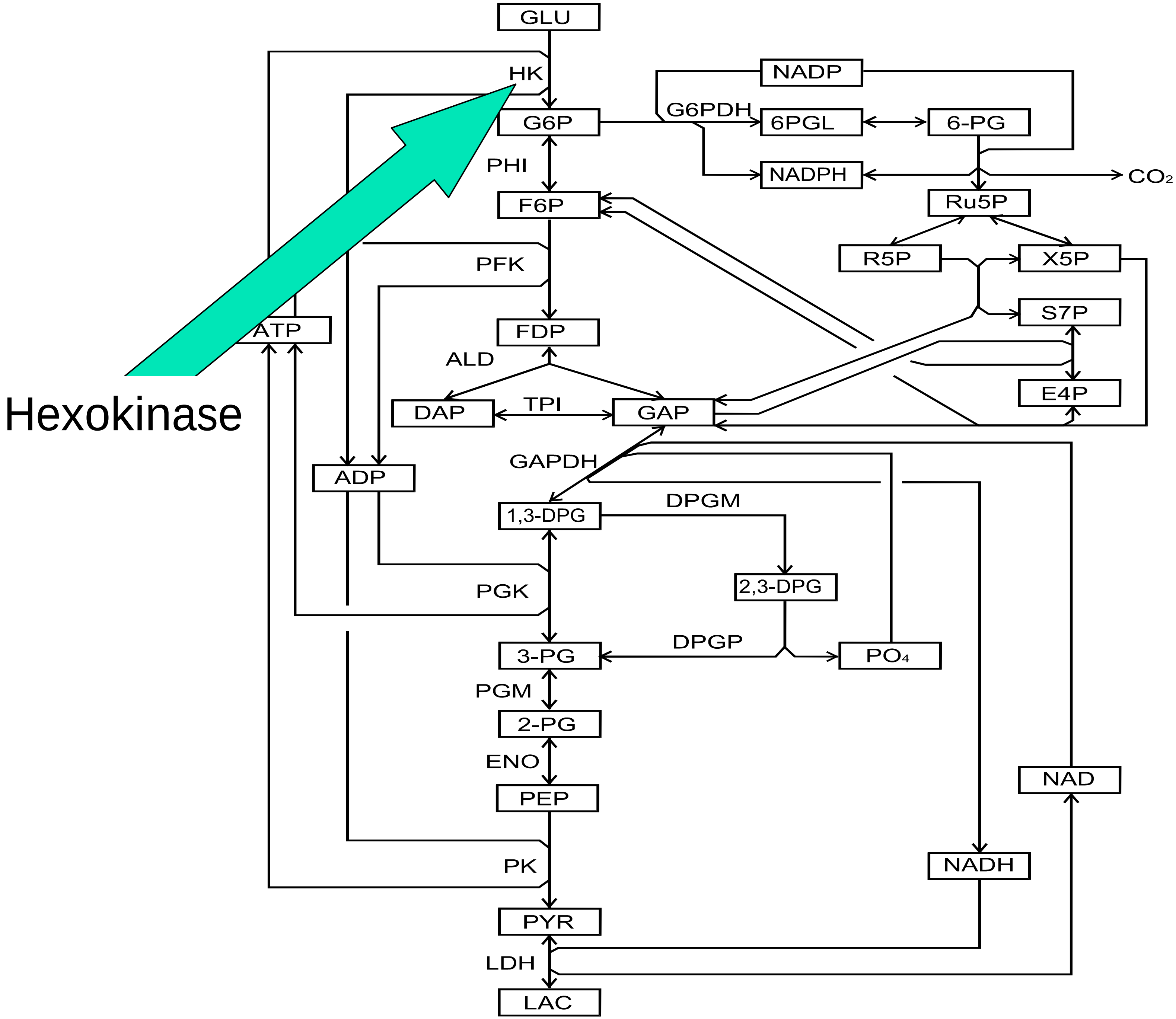


Ataullakhanov F. et al. Eur J Biochem., 1981, v.115, p.359-365


-
Hexokinase
Glucose
G6P
ATP
ADP
F6P
+
-
Phosphofructo-
kinase
FDP
ATP
ADP
Adenylate
kinase
Adenosine
AMP
IMP
ATP
ADP
NH3



Stable node
Unstable node
Rate
U_st
Stable node
ATP_st
ATP
Stable node


GLU
HK
G6P
NADP
G6PDH
6PGL
6-PG
NADPH
CO_2
PHI
F6P
Ru5P
R5P
X5P
PFK
S7P
ATP
FDP
ALD
E4P
DAP
TPI
GAP
GAPDH
ADP
1,3-DPG
DPGM
2,3-DPG
PGK
3-PG
DPGP
PO_4
PGM
2-PG
ENO
PEP
NAD
PK
NADH
PYR
LDH
LAC
Hexokinase

Panel a:

(1) G6P,
(2) 2,3-DPG,
(3) ATP.

Panel b:

(1) intracellular Na,
(2) erythrocyte volume,
(3) the total concentration of osmotically active metabolites

Martinov M. et al. BBA, 2000, v.1474, p.75-87


[ATP]/ [ATPo]

Table 1. Decrease in enzyme activity in the blood of patients with hereditary anemia

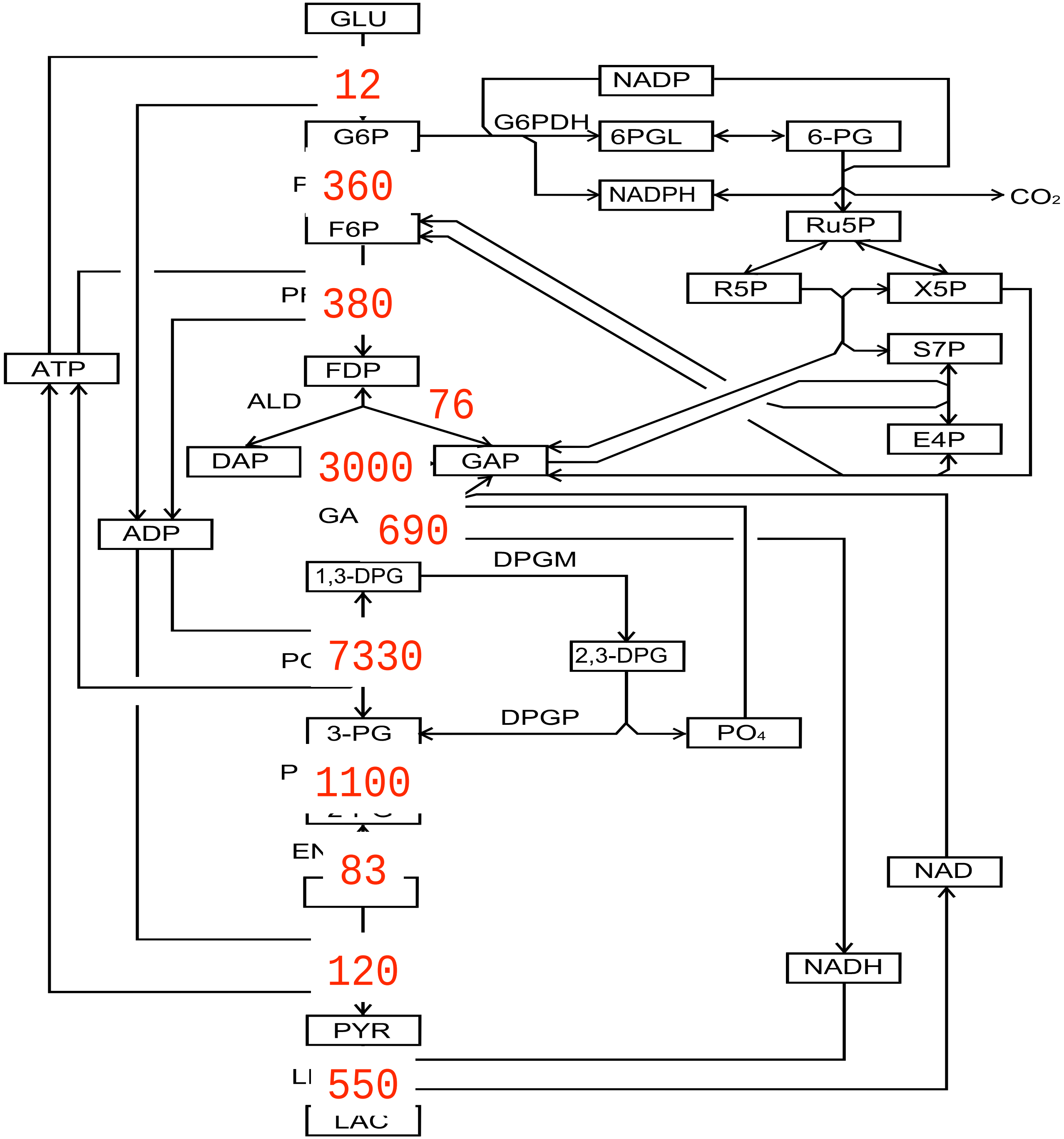
	(α/α_0)
HK	0.24–0.89
GPI	0.05–0.25
PFK	0.08–0.60
ALD	0.04–0.16
TPI	0.016–0.30
GAPDH	0.20–0.50
PGK	0.01–0.30
DPGP	—
PGM	—
ENO	0.06–0.50
PK	0.05–0.40
LDH	—
Na,K–ATPase	0.20–0.60

- Similar decrease of enzyme activity (5-20%) connected with hereditary anemia for almost all mutant enzyme

- No correlation between decrease of activity and severity of the anemia

Table 2. Comparison with experimental data

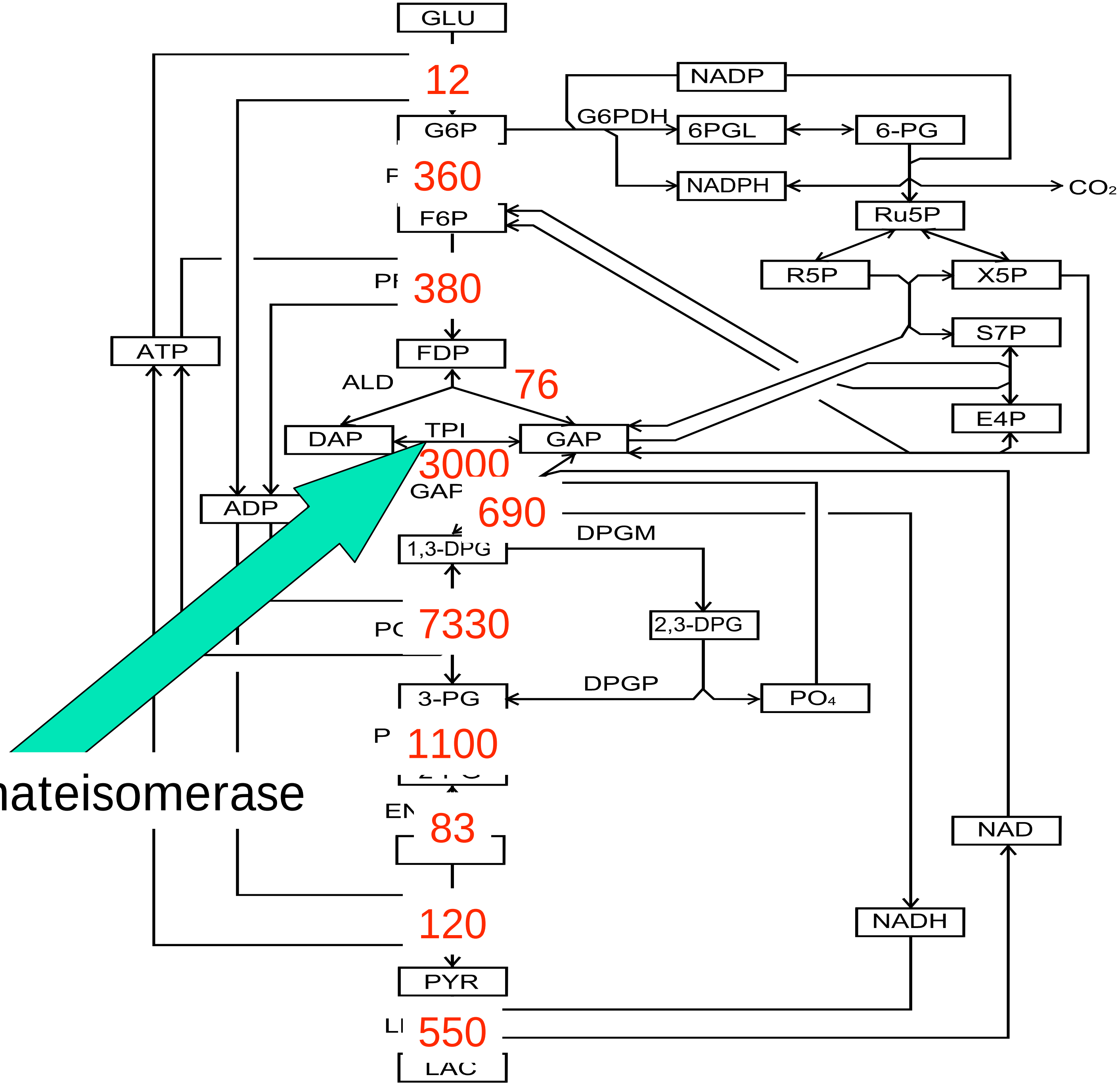
	Calculated activity	Experimental data
	(α^{cr}/α^{0})	(α/α^{0})
HK	0.39	0.24–0.89
GPI	0.015	0.05–0.25
PFK	0.011	0.08–0.60
ALD	0.03	0.04–0.16
TPI	0.0004	0.016–0.30
GAPDH	0.13	0.20–0.50
PGK	0.0033	0.01–0.30
DPGP	0.11	—
PGM	0.0074	—
ENO	0.20	0.06–0.50
PK	0.22	0.05–0.40
LDH	0.015	—
Na,K-ATPase	0.11	0.20–0.60



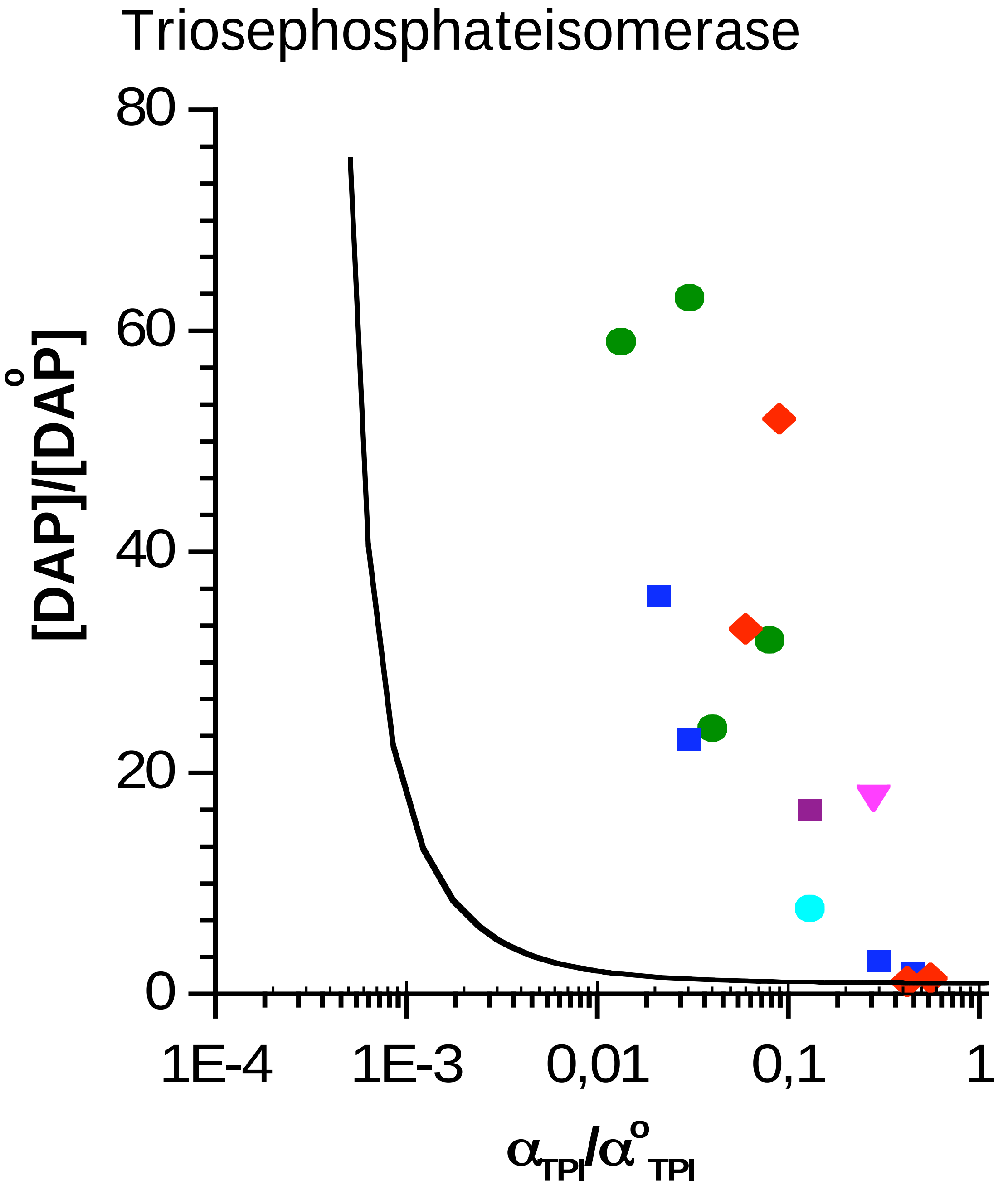
GLU
12
NADP
G6PDH
G6P
6PGL
6-PG
360
NADPH
CO2
F6P
Ru5P
R5P
X5P
380
S7P
ATP
FDP
ALD
76
E4P
DAP
3000
GAP
GA
690
ADP
DPGM
1,3-DPG
7330
2,3-DPG
DPGP
3-PG
PO4
1100
83
NAD
120
NADH
PYR
550
LAC

Dibrov B. et al., have shown that the range of dynamic stability can be widened greatly, if the pathway contains one or two reactions (but not more) with relatively small effective rate constants.

Dibrov B. et al., J. Math. Biology (1982) v.15, p.51-63



Triosephosphateisomerase



Triosephosphateisomerase
80
60
40
20
0
[DAP]/[DAP]°
1E-4
1E-3
0,01
0,1
1
αTPI/α°TPI

Hypothesis:

Mutant form of an enzyme is unstable and decays exponentially:

$$\alpha(t) = \alpha^{o}\exp(-t/\tau)$$

Erythrocyte dies when activity of the mutant enzyme decreases down to

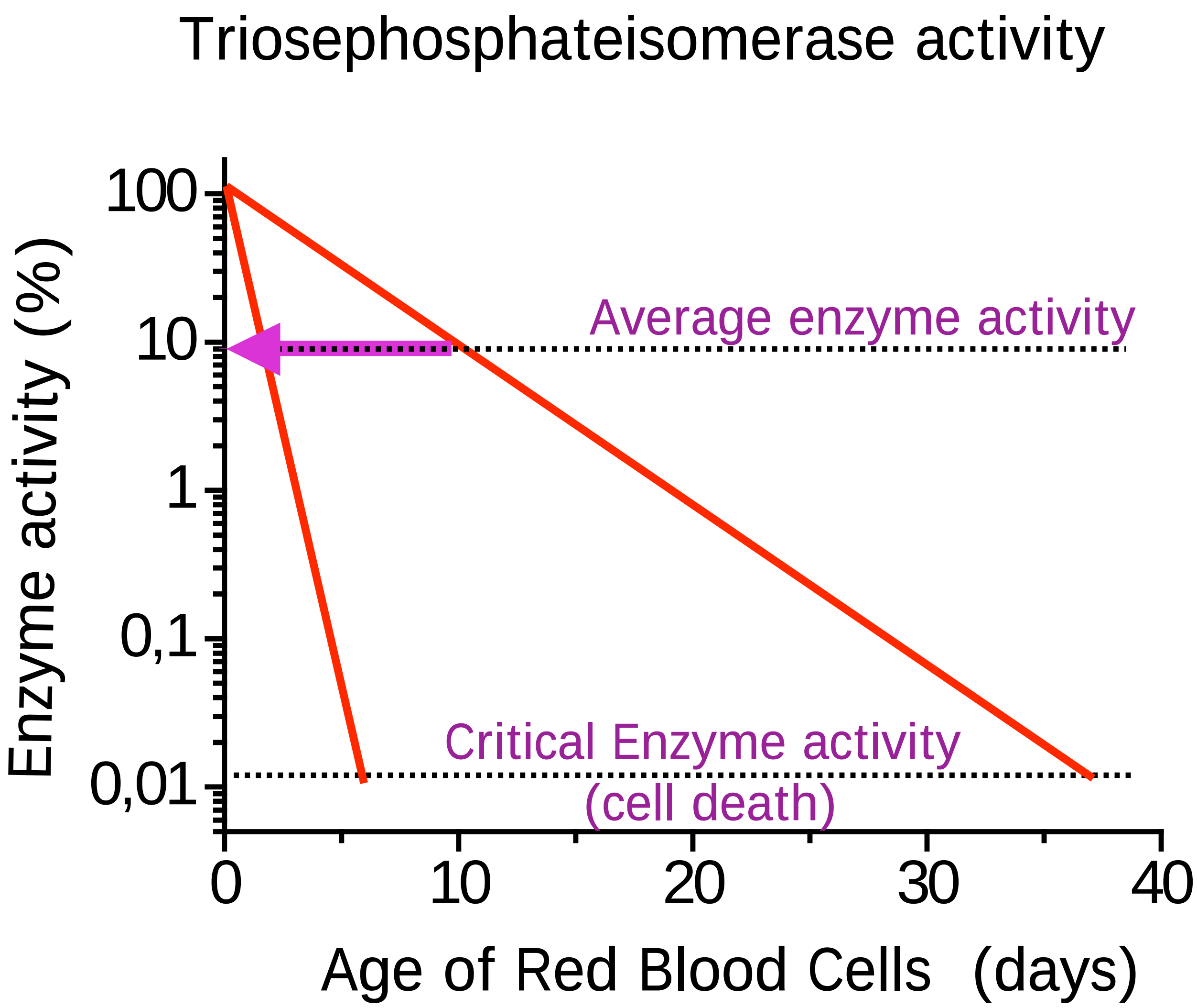
$$\alpha^{cr} = \alpha^{o}\exp(-T/t),$$

where T is an RBC's lifespan in circulation

So $T = t\ln(\alpha^{o}/\alpha^{cr})$,

and the mean value of enzyme activity in the blood is

$$\alpha^{m} = \frac{1}{T}\int_{0}^{T}\alpha dt = \frac{1}{T}\int_{0}^{T}\alpha^{o}\exp(-t/\tau)dt = (\alpha^{o}-\alpha^{cr})/\ln(\alpha^{o}/\alpha^{cr})$$


Triosephosphateisomerase activity
Enzyme activity (%)
100
10
1
0,1
0,01
Average enzyme activity
Critical Enzyme activity
(cell death)
0
10
20
30
40
Age of Red Blood Cells (days)

Hypothesis:

Mutant form of an enzyme is unstable and decays exponentially.

Predictions:

- Mean level of enzyme activity in the blood is much higher than critical and falls in a diapason of 5-20%.

- Severity of anemia should correlate with the rate of enzyme degradation in the cell but not with the mean enzyme activity.

Table 2. Comparison with experimental data

	Stable enzyme	Unstable enzyme	Experimental data
	(α^{cr}/α^{0})	(α^{m}/α^{0})	(α/α^{0})
HK	0.39	0.65	0.24–0.89
GPI	0.015	0.23	0.05–0.25
PFK	0.011	0.22	0.08–0.60
ALD	0.03	0.28	0.04–0.16
TPI	0.0004	0.13	0.016–0.30
GAPDH	0.13	0.43	0.20–0.50
PGK	0.0033	0.17	0.01–0.30
DPGP	0.11	0.40	—
PGM	0.0074	0.20	—
ENO	0.20	0.50	0.06–0.50
PK	0.22	0.52	0.05–0.40
LDH	0.015	0.23	—
Na,K-ATPase	0.11	0.40	0.20–0.60

Conclusions:

- What is a disease from the mathematical point of view?

  Existence in the vicinity of a bifurcation.

- Complex and simple models: how do they relate with each other?

  Simple model helps to understand a nature of bifurcation, thereby helping to interpret a more complete, complex quantitative model.

- Why do so many cellular enzymes have excessively high activities?

  To allow a high degree of stabilization control.

Contributors:

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A. Zhabotinsky