

# Systems Biology

# Simulation of Dynamic Network States



# Lecture #12

# Building Networks

# Bernhard Ø. Palsson

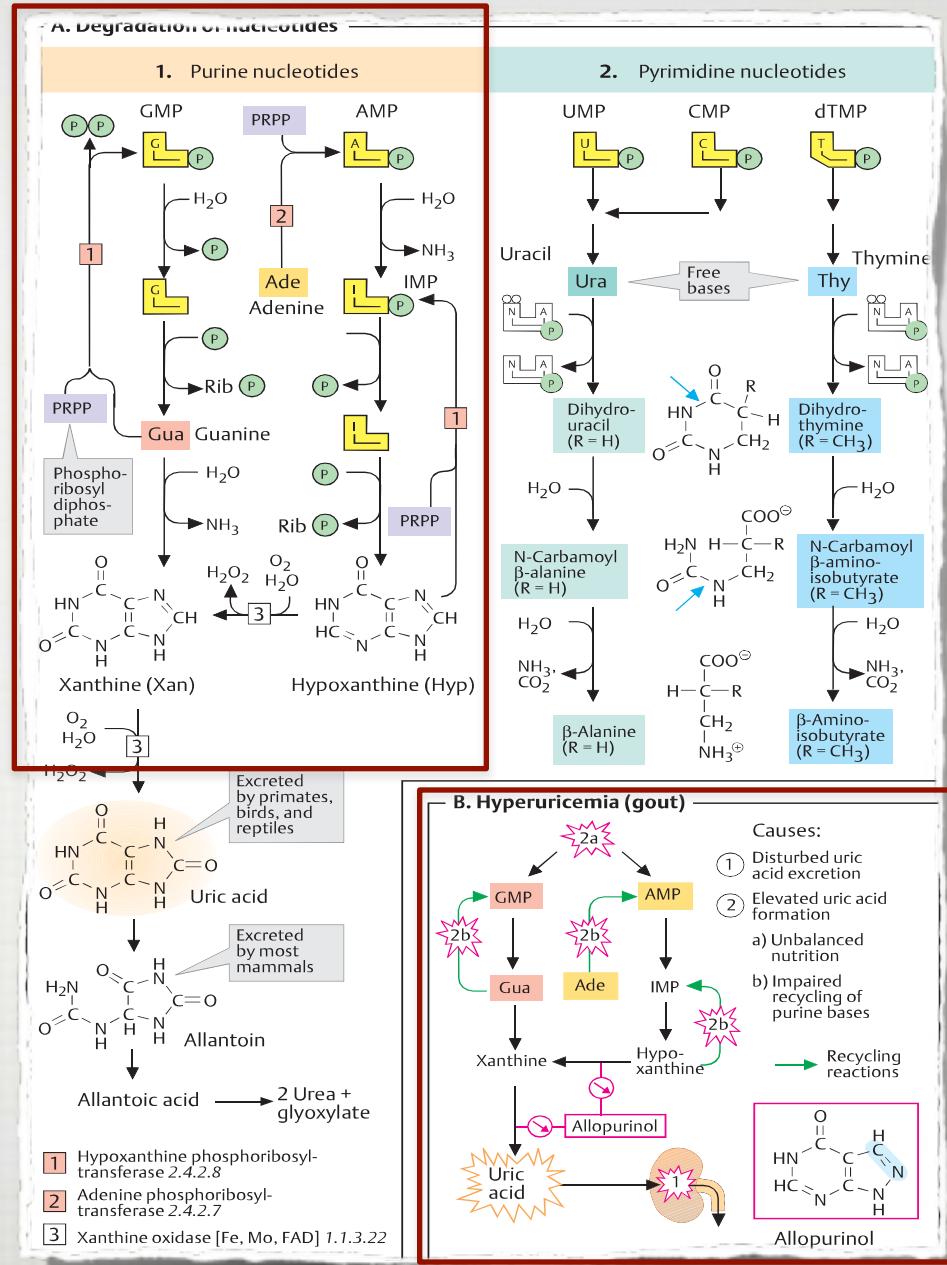
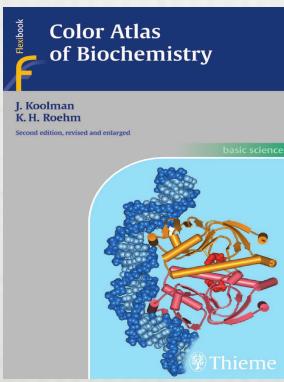
# Outline

- AMP biosynthesis and degradation
  - A dynamic balance (before the AMP input was fixed)
- Inborn errors in metabolism (IEM)
  - Quite common in this pathway
- The AMP sub-network
  - Formulation, balancing, QC/QA, simulation
- Integration with coupled pathways
  - Integration issues are many, many points of contact
- Dynamic simulation for 50% in rate of ATP use
- Path towards whole cell models

Cofactors represent low flux but important pathways

## **SOME BIOCHEMISTRY**

# Nucleotide metabolism: associated with many diseased states



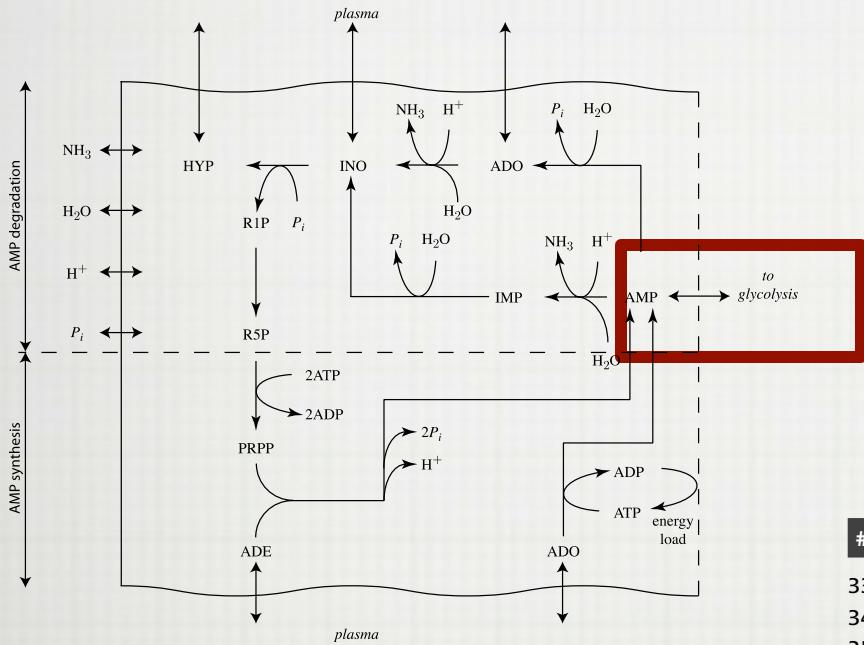
# Table of mutations & associated pathology

Enzyme	Pathology from Abnormal Enzyme Activity
Adenosine Deaminase	Deficiency results in immune deficiency disease
Adenine	Deficiency results in renal failure due to production of toxic 2,8-dihydroxyadenine
Phosphoribosyltransferase	
Hypoxanthine-guanine Phosphoribosyl Transferase	Deficiency results in Lesch-Nyhan Syndrome
Prpp synthase	Overactivity results in hyperuricemia
AMP Deaminase	Deficiency in erythrocytes is asymptomatic
AMPase/IMPase	Deficiency results in decreased red cell survival
Purine Nucleoside Phosphorylase	Deficiency results in defective T-cell immunity

Forming a sub-network

# **AMP METABOLISM**

# AMP Salvage Network



	AMP	ADP	ATP	P <sub>i</sub>	H	H <sub>2</sub> O	NH <sub>3</sub>	ADO	ADE	IMP	INO	HYP	R1P	R5P	PRPP
C	10	10	10	0	0	0	0	10	5	10	10	5	5	5	5
H	13	13	13	1	1	2	3	13	5	12	12	4	9	9	8
O	7	10	13	4	0	1	0	4	0	8	5	1	8	8	14
P	1	2	3	1	0	0	0	0	0	1	0	0	1	1	3
N	5	5	5	0	0	0	1	5	5	4	4	4	0	0	0

#	Abbreviation	Intermediates	Concentration (mM)
33	ADO	Adenosine	0.0012
34	ADE	Adenine	0.001
35	IMP	Inosine monophosphate	0.01
36	INO	Inosine	0.001
37	HYP	Hypoxanthine	0.002
38	R1P	Ribose 1-phosphate	0.06
39	PRPP	Phosphoribosyl diphosphate	0.005

#	Abbrev.	Enzymes/transporter/load	Elementally balanced reaction
33	AK	Adenosine kinase	ATP + ADO → AMP + ATP
34	AMPase	AMP phosphohydrolase	AMP + H <sub>2</sub> O → ADO + P <sub>i</sub> + H
35	AMPDA	AMP deaminase	AMP + H <sub>2</sub> O → IMP + NH <sub>3</sub>
36	IMPase	IMP phosphohydrolase	IMP + H <sub>2</sub> O → INO + P <sub>i</sub> + H
37	ADA	Adenosine deaminase	ADO + H <sub>2</sub> O → INO + NH <sub>3</sub>
38	PNPase	Purine nucleoside phosphorylase	INO + P <sub>i</sub> → HYP + R1P
39	PRM	Phosphoribomutase	R1P → R5P
40	PRPPsyn	PRPP synthase	R5P + 2 ATP → PRPP + 2 ADP + H
41	ADPRT	Adenine phosphoribosyl transferase	PRPP + ADE + H <sub>2</sub> O → AMP + 2 P <sub>i</sub>
42	ADO	Adenosine exchange	Transport
43	ADE	Adenine exchange	Transport
44	INO	Inosine exchange	Transport
45	HYP	Hypoxanthine exchange	Transport
46	NH <sub>3</sub>	Ammonium exchange	Transport

# AMP Salvage: S Matrix

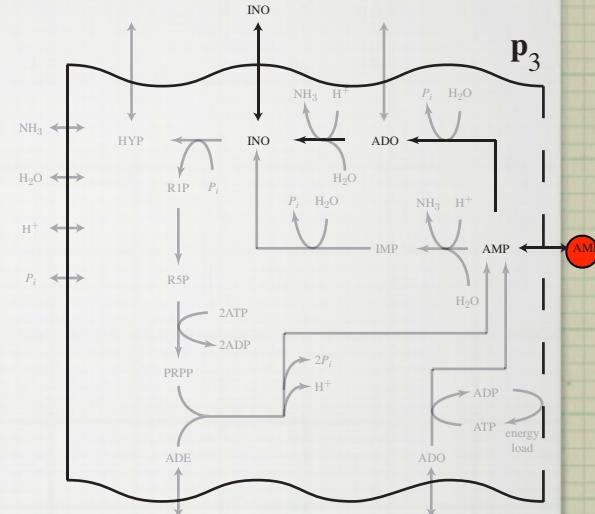
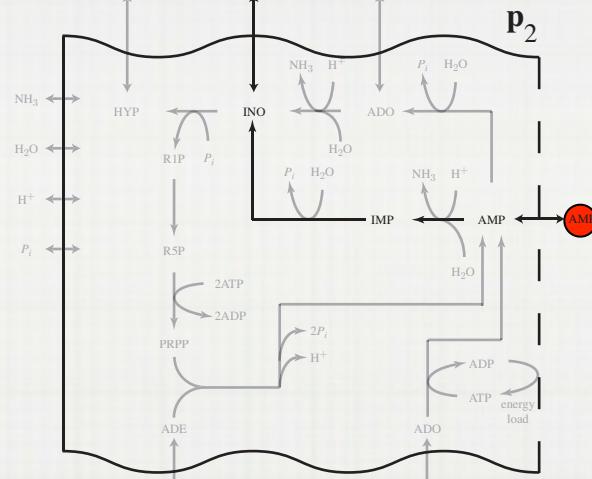
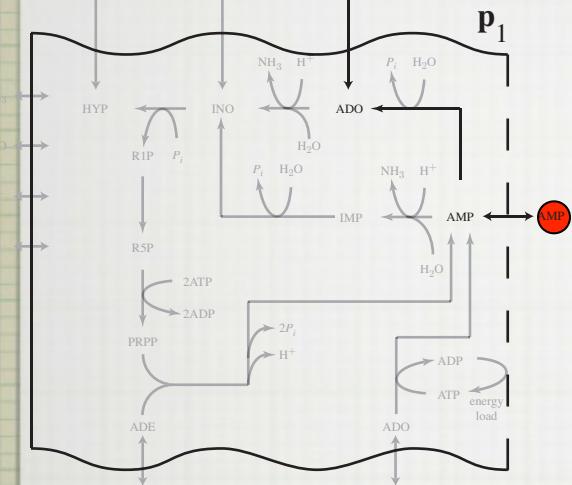
internal

exchange

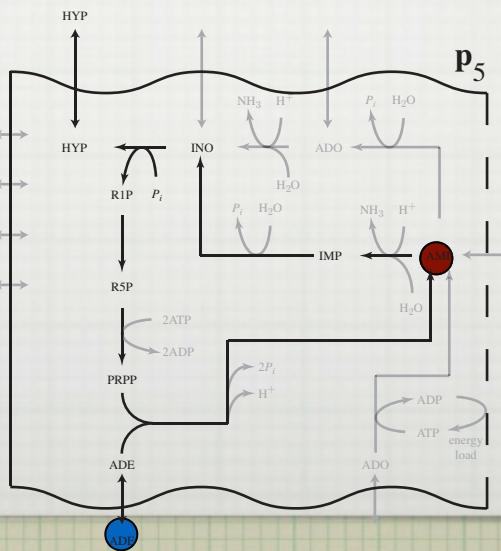
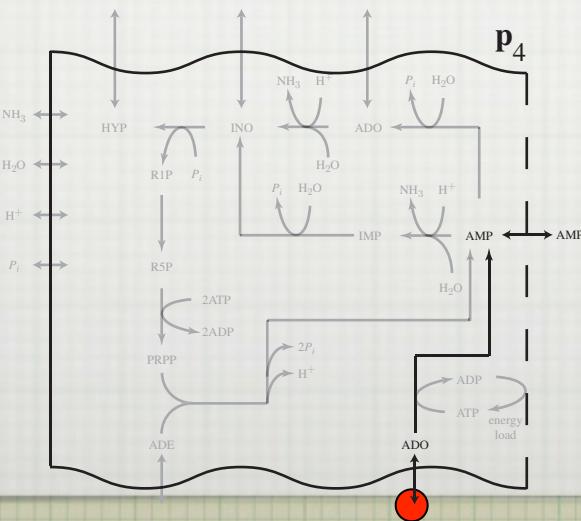
	Nucleotide metabolism								Exchange fluxes										
	$V_{AK}$	$V_{AMPase}$	$V_{AMPDA}$	$V_{IMPase}$	$V_{ADA}$	$V_{PNPase}$	$V_{PRM}$	$V_{ATPgen}$	$V_{PRPPsyn}$	$V_{ADPRT}$	$V_{ADO}$	$V_{ADE}$	$V_{INO}$	$V_{HYP}$	$V_{AMP}$	$V_H$	$V_{H_2O}$	$V_P$	$V_{NH_3}$
AMP	1	1	1	2	2	2	2	2	2	1	0	0	0	0	-1	0	0	0	0
A	$p_1 + p_4$ give a net AMP synthesis of 0.01 mM/hr																		
A	so $p_2 + p_3$ drain AMP by 0.01 mM/hr																		
P	set $V_{ADA}$ to 0.01 mM/hr and we set																		
H	the weight on $p_2$ to be 0 since it overlaps with $p_5$ ,																		
F	and thus the weight on $p_3$ is 0.01 mM/hr																		
N																			
A																			
II																			
NO	0	0	0	1	1	-1	0	0	0	0	0	0	0	0	-1	0	0	0	
HYP	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	
R1P							1	-1	0	0	0	0	0	0	0	0	0	0	
R5P							0	1	0	-1	0	0	0	0	0	0	0	0	
PRPP							0	0	0	1	-1	0	0	0	0	0	0	0	
CO							0	0	0	0	0	-1	-5	-10	-5	-10	0	0	
H							0	0	0	0	0	-1	-5	-12	-4	-13	-1	-2	
C							0	0	0	0	0	-1	0	-5	-1	-7	0	-1	
F							0	0	0	0	0	0	0	0	-1	0	0	-1	
N							0	0	0	0	0	0	-1	-4	-4	-5	0	0	
R1	1	0	0	0	0	0	0	0	0	0	-1	1	0	0	0	-1	1	0	
$p_2$	0	1	1	0	0	0	0	0	0	0	0	0	0	1	0	1	-2	1	
$p_3$	1	0	0	1	0	0	0	0	0	0	0	0	0	1	0	-1	1	1	
$p_4$	0	0	0	0	0	0	1	0	0	0	-1	0	0	0	1	-1	1	0	
$p_5$	0	0	1	1	0	1	1	2	1	1	0	-1	0	1	0	0	-1	0	
$v_{stst}$	0.12	0.12	0.014	0.014	0.01	0.014	0.014	0.15	0.014	0.014	-0.01	-0.014	0.01	0.014	0.0	0.0	-0.024	0.0	0.024

# AMP Salvage:

## pathway vectors

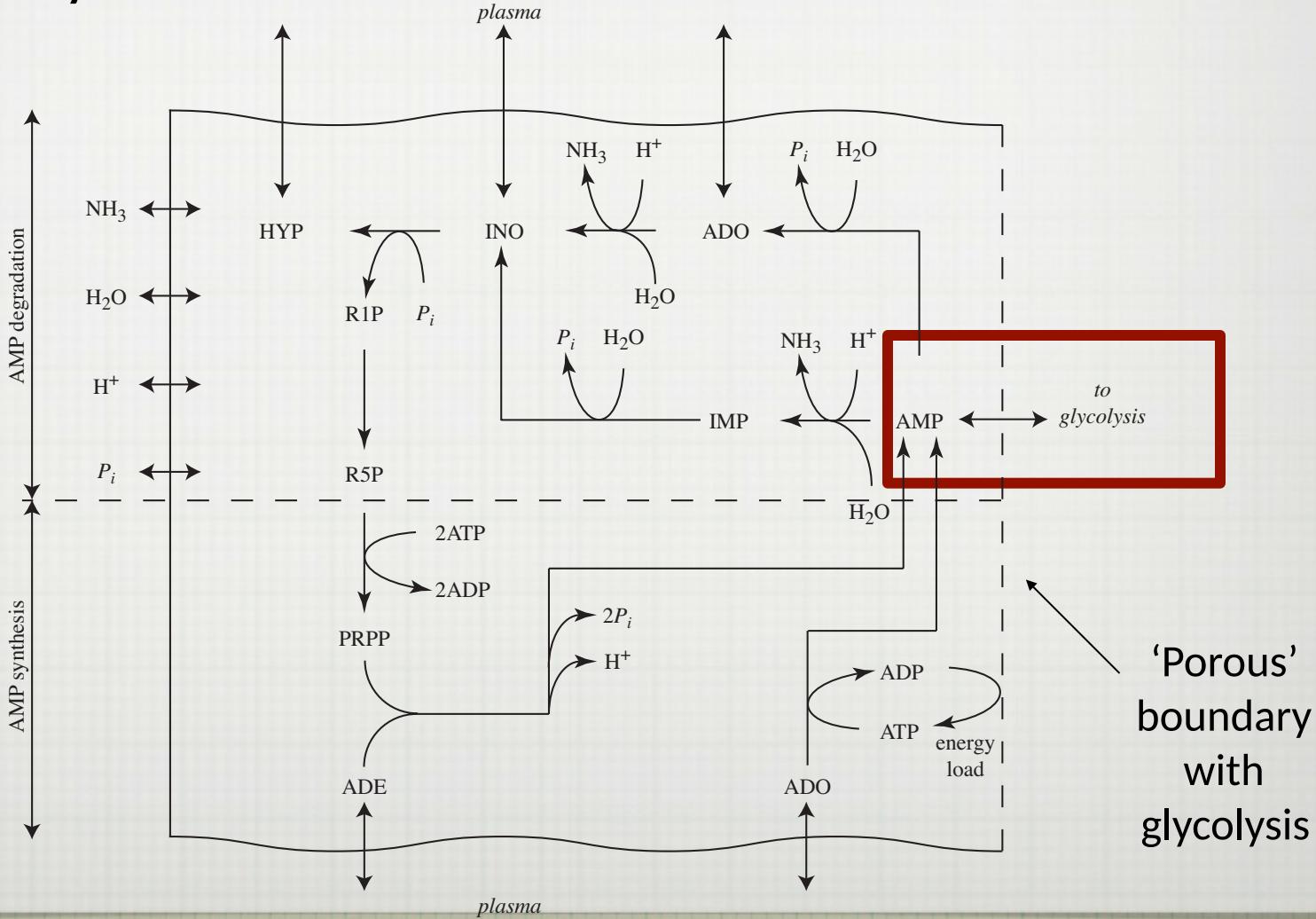


*Degradation*  
*Biosynthesis*



# AMP Salvage Network:

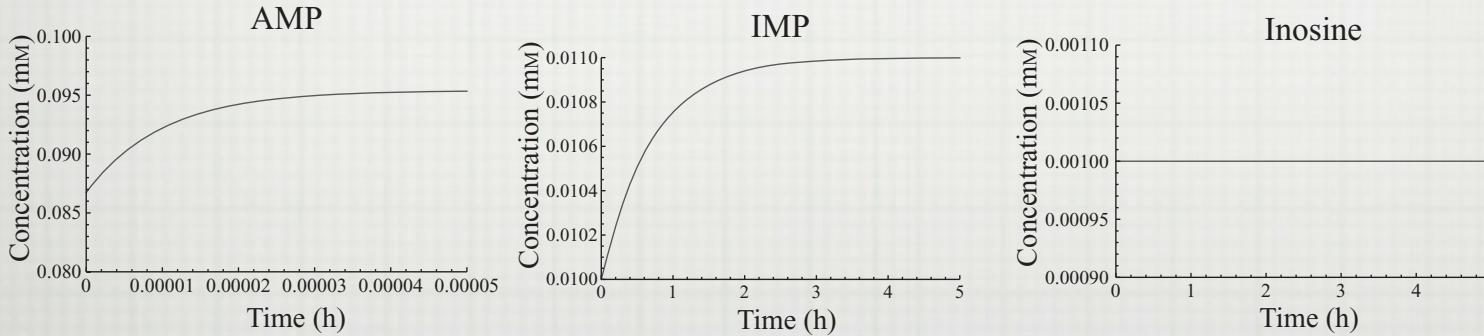
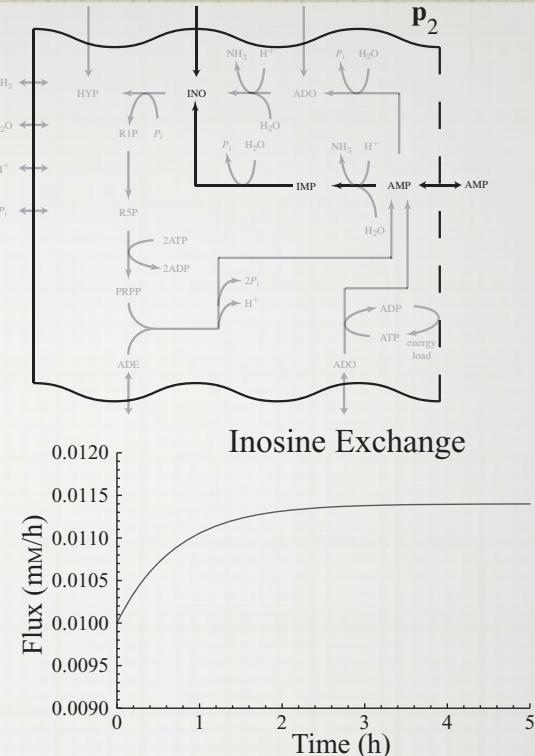
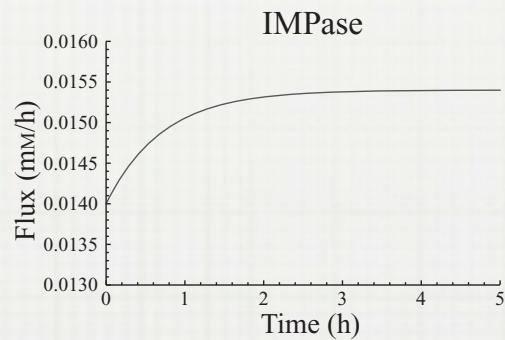
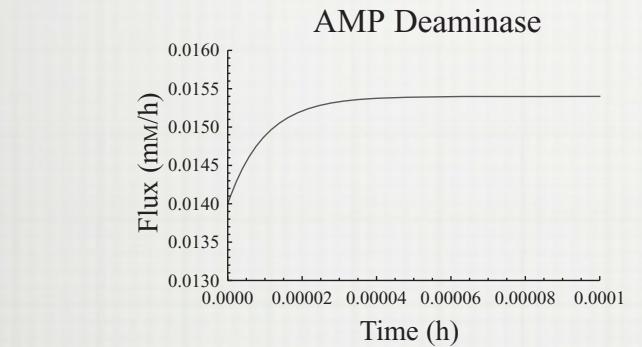
PERCs computed from the steady state data  
dynamic simulation to AMP increase



'Porous'  
boundary  
with  
glycolysis

# AMP Salvage:

## Dynamic Simulation; 10% increase in AMP concentration

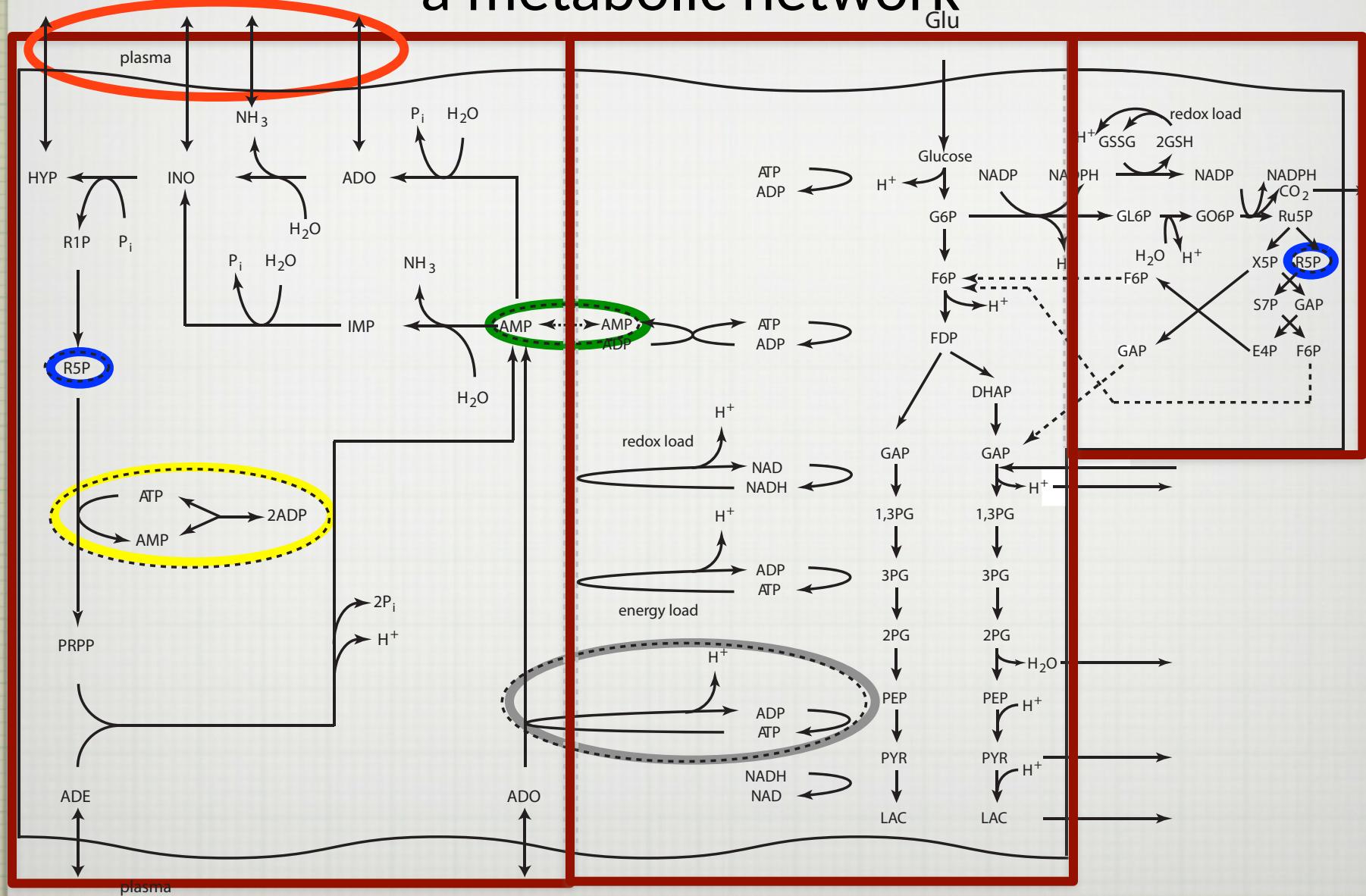


One of the degradation routes is activated

Adding AMP Salvage:

## **FORMING INTEGRATED NETWORKS**

# Glycolysis, PPP, & AMP: a metabolic network



- The AMP molecule in the two networks connects the two. These two nodes need to be merged into one.
- The R5P molecule appears in both the AMP metabolic subnetwork and the pentose pathway, so these two nodes also need to be merged.
- In the sub-network described above, the stoichiometry of the PRPP synthase reaction is



but in actuality it is



this difference disappears since the ApK reaction is in the combined glycolytic and pentose pathway network :



- The ATP cost of driving the biosynthetic pathways to AMP is now a part of the ATP load in the integrated model.
- The AMP exchange reaction disappears, and instead we now have exchange reactions for ADO, ADE, INO, and HYP, and the deamination reactions create an exchange flux for NH<sub>3</sub>.

# Integration Issues

# Glycolysis, PPP, & AMP: S matrix

# Glycolysis, PPP, & AMP: pathway vectors

Glycolysis

Integrated PPP

AMP  
degradation

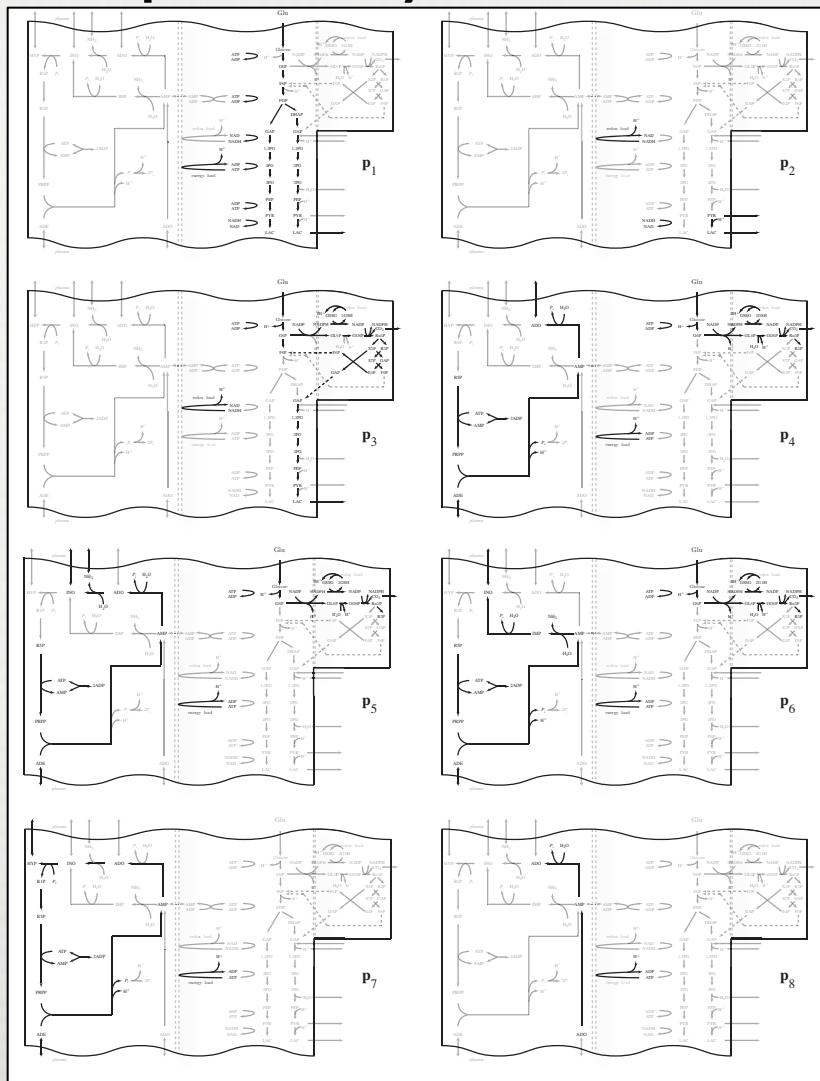
Salvage pathway

Pyr/Lac exchange

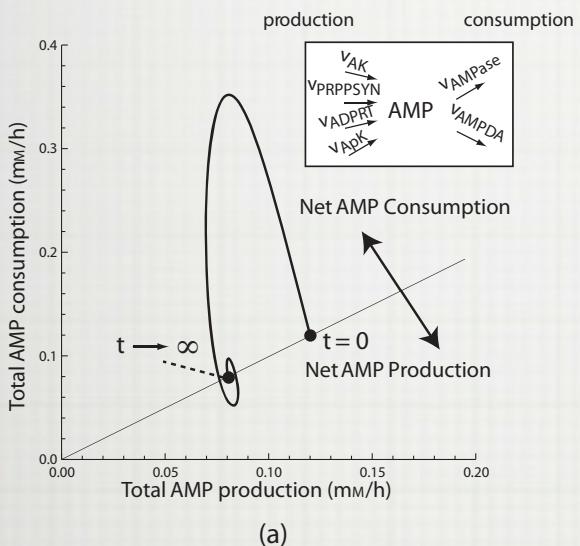
AMP  
degradation

AMP  
degradation

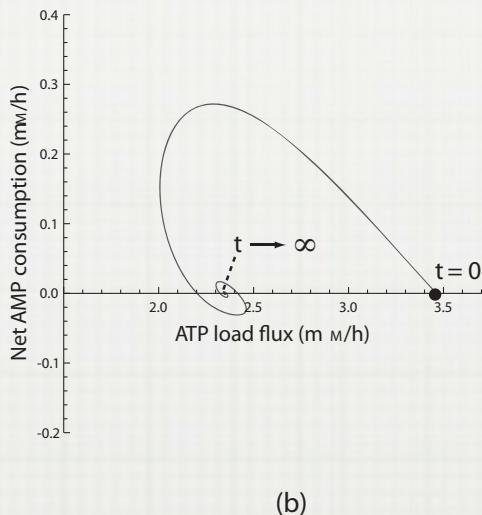
Futile cycle\*



# Integrated Model: Simulation

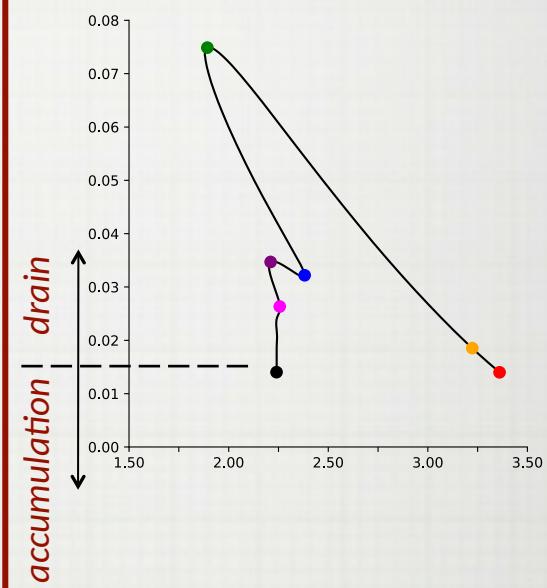


(a)



(b)

**Figure 12.6** Dynamic response of the combined glycolytic, pentose pathway, and AMP metabolism network to a 50% increase in the rate of ATP use. (a) The phase portrait of total AMP consumption and production fluxes. The  $45^\circ$  line is the steady state, above which there is a net consumption (i.e., efflux) from the AMP node and below which there is a net production (i.e., import) of AMP. (b) The phase portrait of ATP load and net AMP consumption fluxes. See Figures 10.5a and 11.6a for comparison.



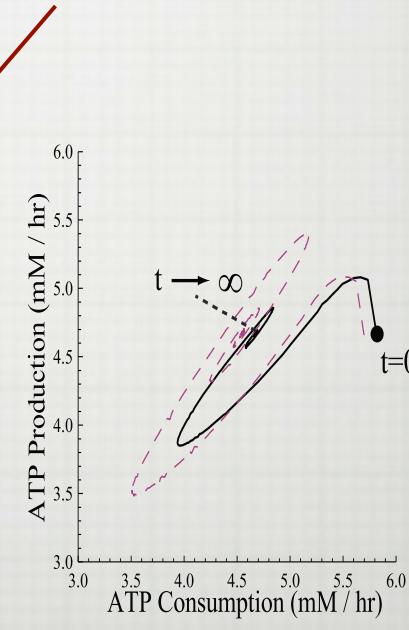
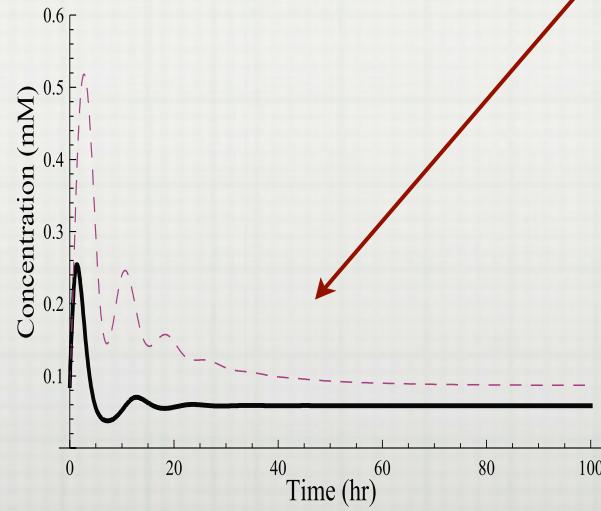
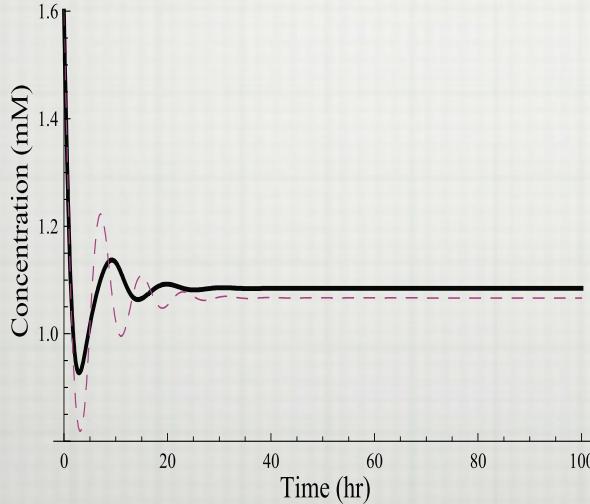
Fixed AMP  
input at 0.014  
from ch 10

# Plotting multiple solutions

Comparing responses from two models,  
(glycolysis + PPP) +/- AMP metabolism

- The AMP I/O behavior
- More damped than before

Solid line: full network  
Dashed line: glyc+ppp



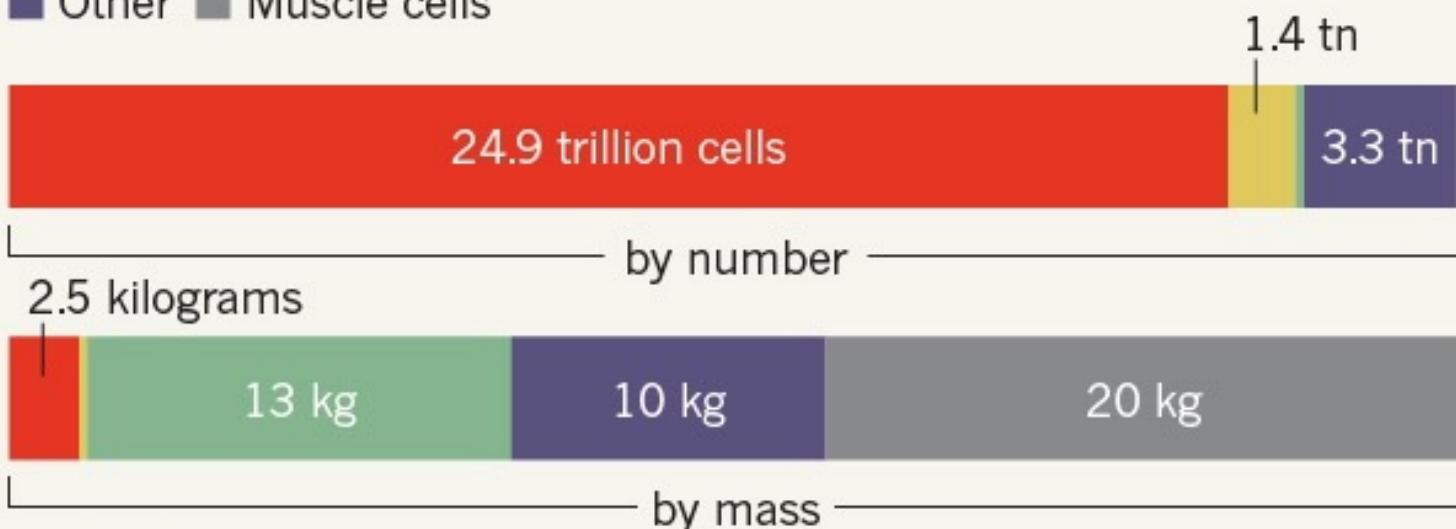
# **TOWARDS A WHOLE CELL MODEL**



## COUNTING HUMAN CELLS

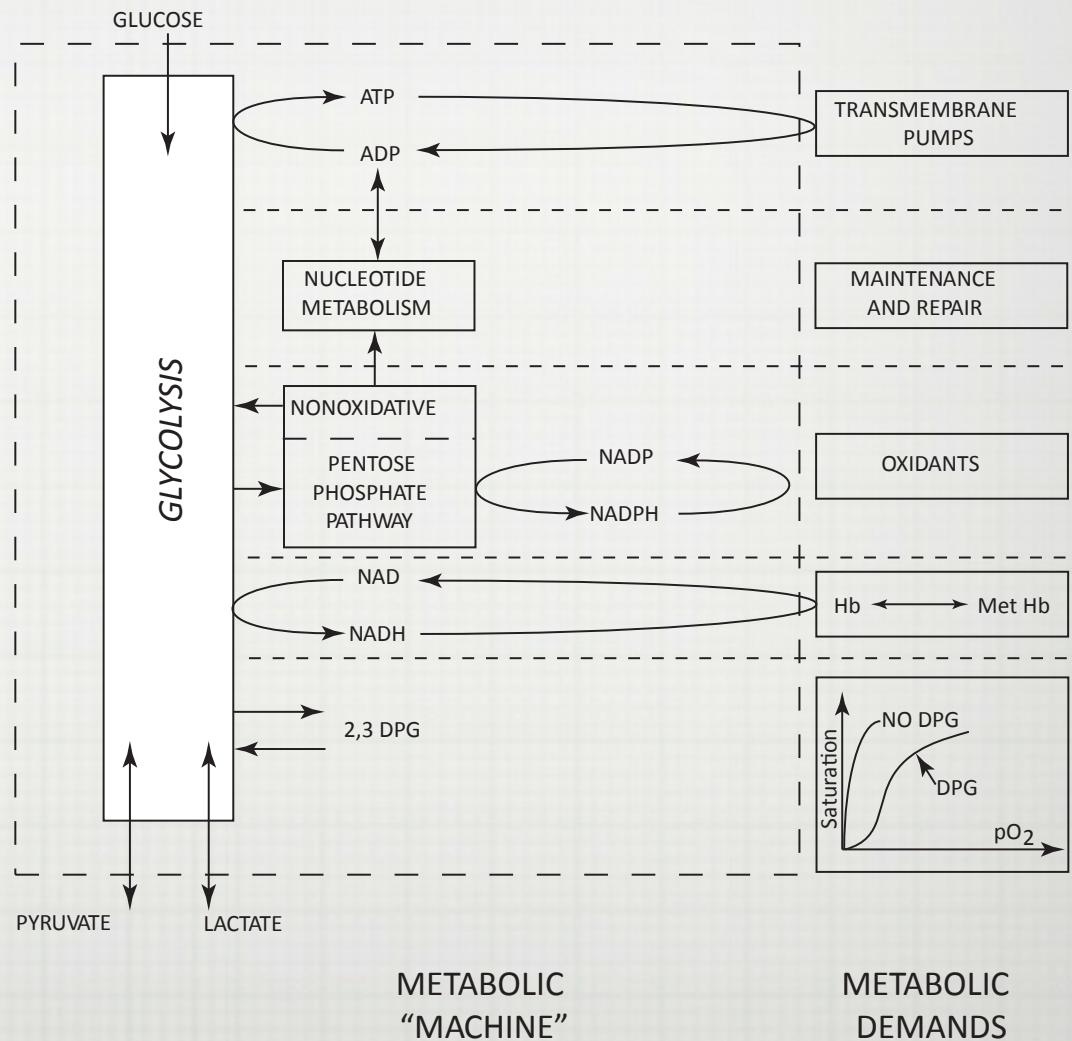
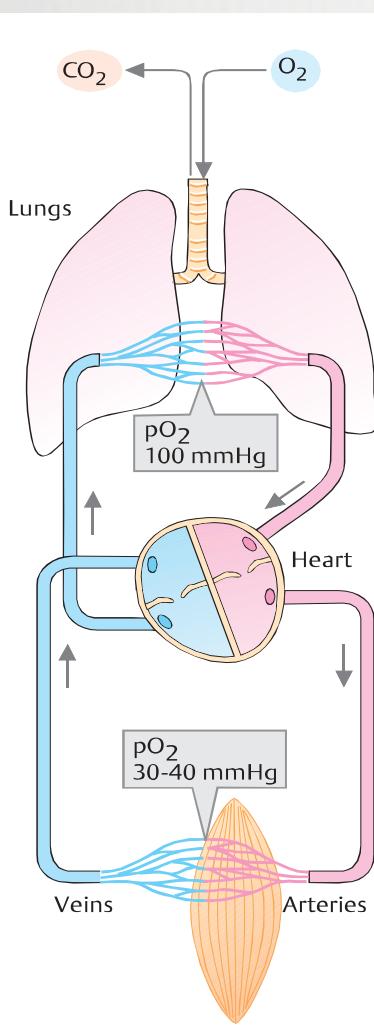
Most of our body's cells are small red blood cells, although fat cells and muscle cells make up the majority by mass.

- Red blood cells (erythrocytes)
- Platelets
- Fat cells (adipocytes)
- Other
- Muscle cells

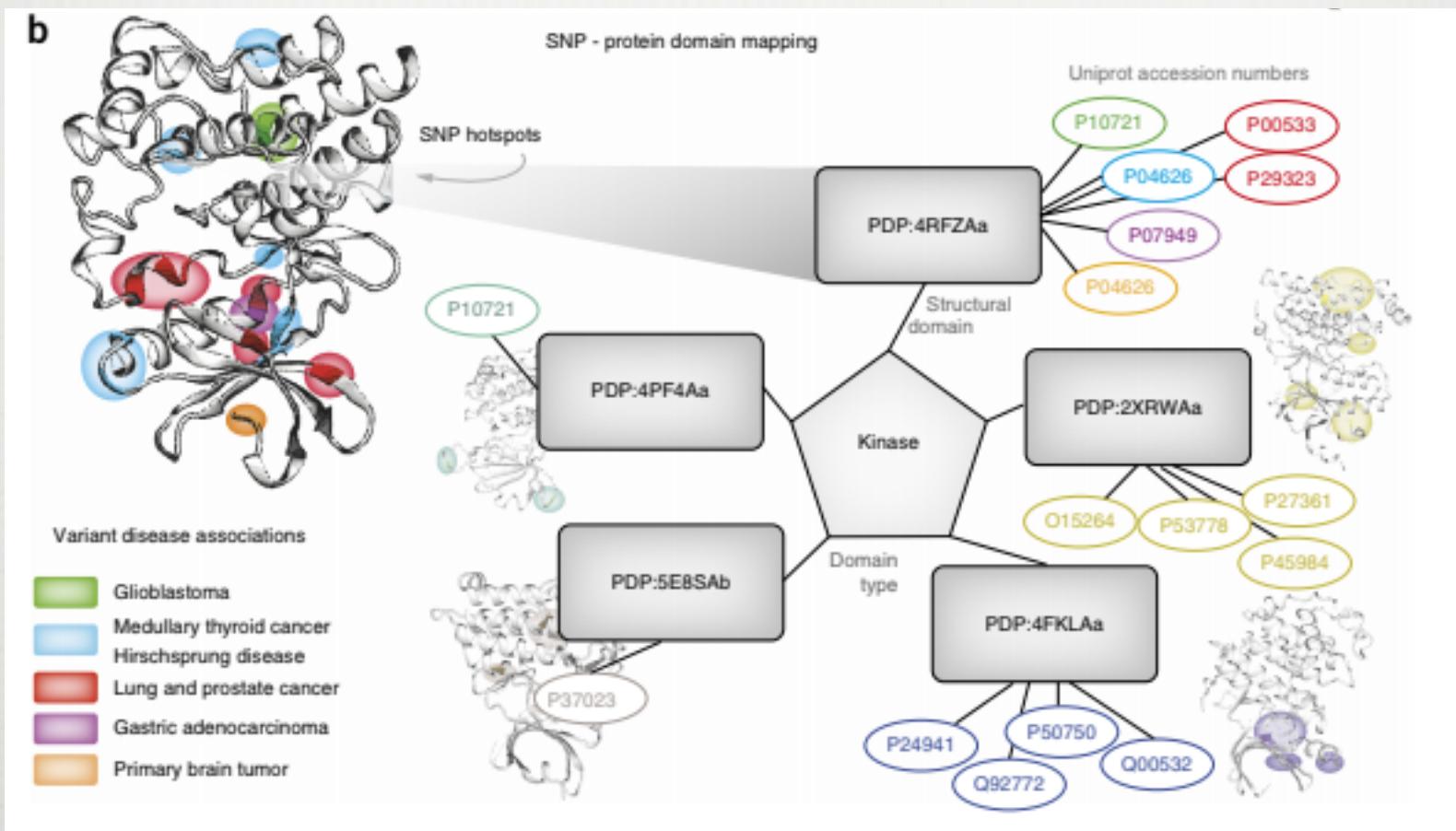


# Toward a whole cell simulation:

## Metabolic demands and the ‘machine’ that meets them

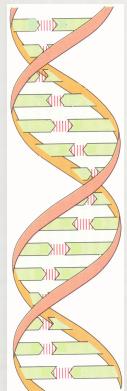


# Models can be personalized based on sequence variations



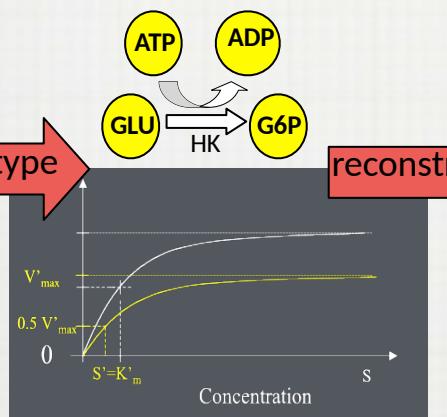
# Studying Genetic Defects

Variation  
(SNP) in DNA  
sequence



genotype

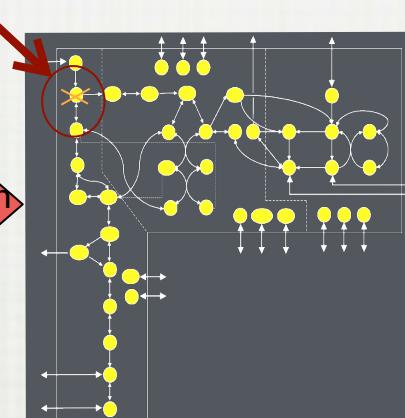
Change in  
enzyme  
kinetic  
properties



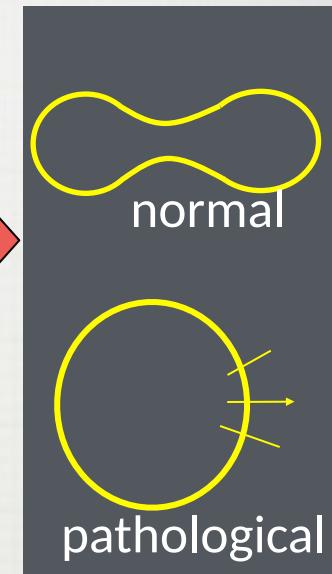
Hexokinase:  
Chromosome 10  
p11.2 (1667 T -> C)

$V_{max}$  and  $K_m$   
values  
altered by  
SNP

Affects systemic  
functioning of  
cell



Phenotypic  
expression of SNP



Decrease in rate of  
glycolysis and ATP  
production

Unable to maintain  
osmotic balance  
under stringent  
ATP loads -> cells  
lyse

# Summary

- Purine nucleotide metabolism is complicated and has many pathological states associated with it
- Nucleotides are synthesized and degraded to be in a steady state that is dynamic and can respond to perturbations
- A sub-network for AMP metabolism can be built and synthesized, and its responses simulated
- It can be integrated with the coupled glycolysis+PP pathways to form a network model
- Several integration issues show up
- The number of pathways characterizing the null space grows
- The model can be simulated and the dampening effect of the response to increased ATP rate of utilization demonstrated
- This network model can be expanded to a whole cell model

**THE END  
of part 3**