Biochemistry

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生物大分子	糖类	脂类	蛋白质	核酸
长啥样				
干啥的				
咋来的?	外源消化和吸收（动物），糖异生；光合磷酸化（植物）	外源消化和吸收，运输，合成	aa合成	核苷酸合成
咋没的?	糖酵解，TCA，氧化磷酸化，HMP	β-氧化	aa分解	核苷酸分解

11.17	北316	代谢概述，糖酵解
11.20	北316	三羧酸循环
11.24	北316	氧化磷酸化
11.27	北316	磷酸戊糖途径、糖异生
12.1	北316	糖原合成与分解
12.4	北316	光合磷酸化
12.8	北316	脂类分解代谢（一）
12.11	北316	脂类合成代谢（二）
12.15	北316	脂类合成代谢（三）
12.18	北316	蛋白质代谢（一）
12.22	北316	蛋白质代谢（二）
12.25	北316	核酸代谢
12.29	北316	代谢调控

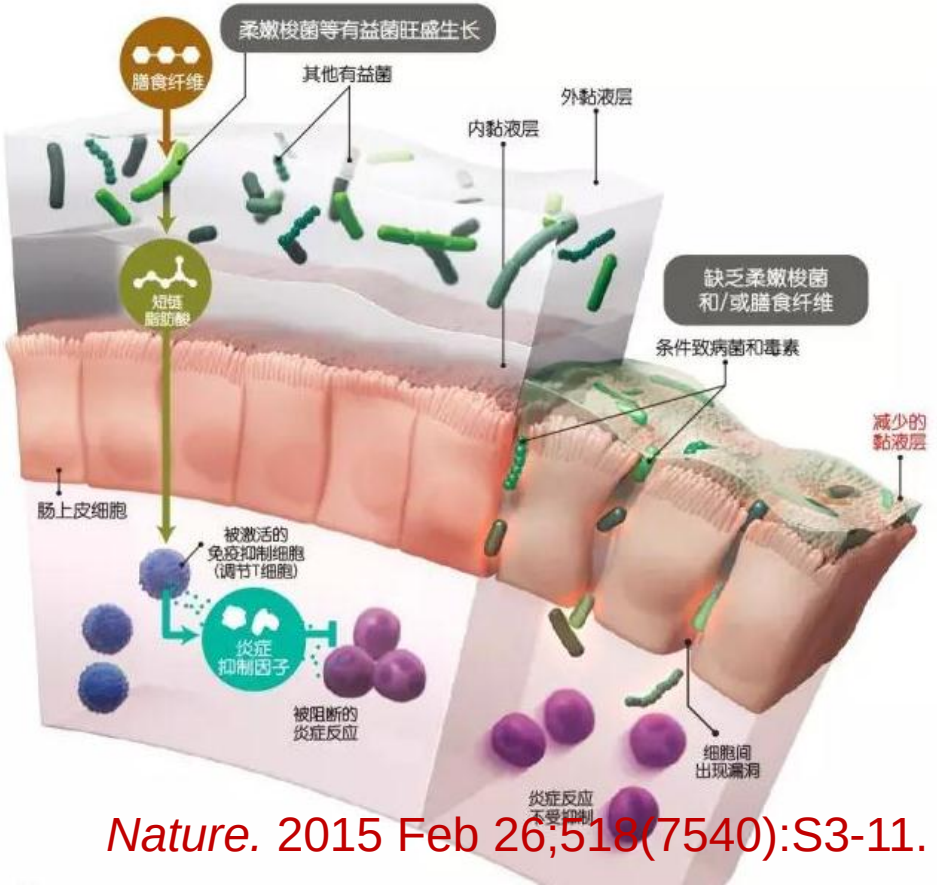
Metabolism Overview

Outline

1. Nutrition
2. Disease with metabolism-obesity, diabetes , CVD（心血管疾病）, cancer
3. Metabolism
4. Metabolic regulation
5. Experimental methods to reveal metabolic pathways

1. Nutrition

- **Carbohydrates – energy, essential components for nucleotides and nucleic acids.**
- **Protein-- source of nitrogen, essential amino acids.**
- **Lipids-- essential fatty acids.**
- **Fiber**



Nature. 2015 Feb 26;518(7540):S3-11.

2. Diseases associated with metabolism

- **Obesity** - One of the most serious **public health problems**

  ✓ Increase **heart diseases**, **type 2 diabetes**, **cancer**（**colon cancer**）

  ✓ **Dieting and physical exercise – control obesity**

No leptin Wild type

Diabetes

- **High blood glucose**

  - Type 1： pancreas does not produce insulin-- inherited

  - Type 2： cells does not respond to insulin -- lifestyle and genetics

  - Gestation（妊娠） diabetes

- **Symptoms: polyuria（多尿） polydipsia（多饮） polyphagia（多食）**
- **Treatment**

  -insulin for Type 1

  -medications Metformin（甲福明，二甲双胍） for control type 2

Metformin can reduce cancer incidence Euro. J. Cancer 2010;46:2369-2380

3. What is metabolism？

Metabolism

-the sum of all the chemical transformations taking place in a cell or organism, occurs through a series of enzyme-catalyzed reactions that constitute metabolic pathways.

Organisms vary in type

- aerobic, anaerobic

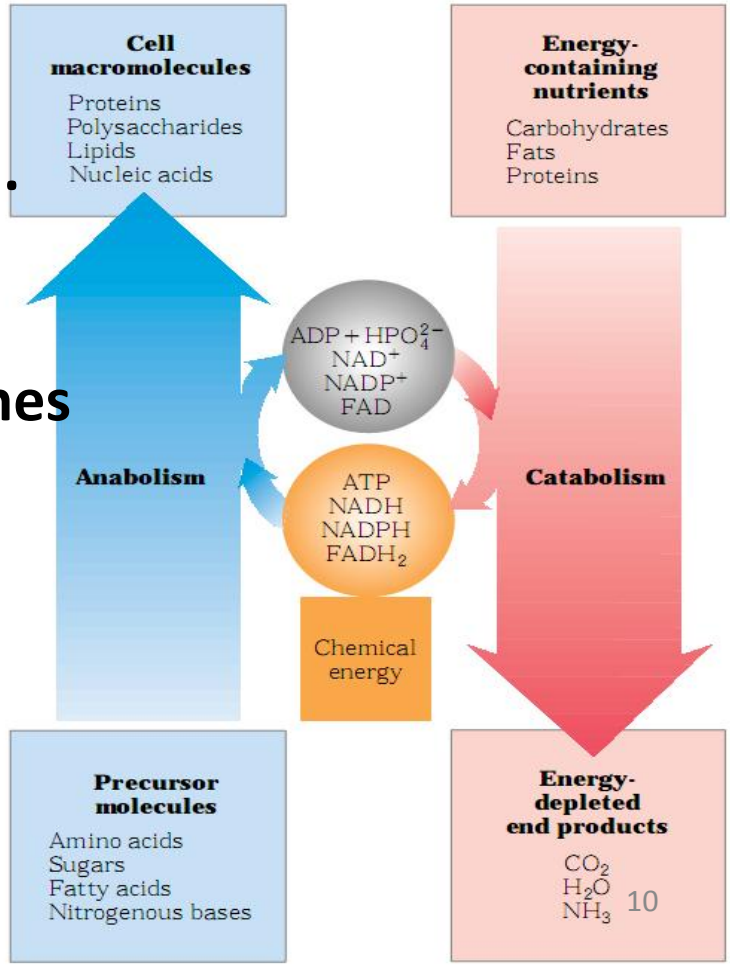
Metabolic pathways

- anabolic（合成） or catabolic（分解）
- “amphibolic”（两用代谢）： anabolic + catabolic
- Pathways vary in type:

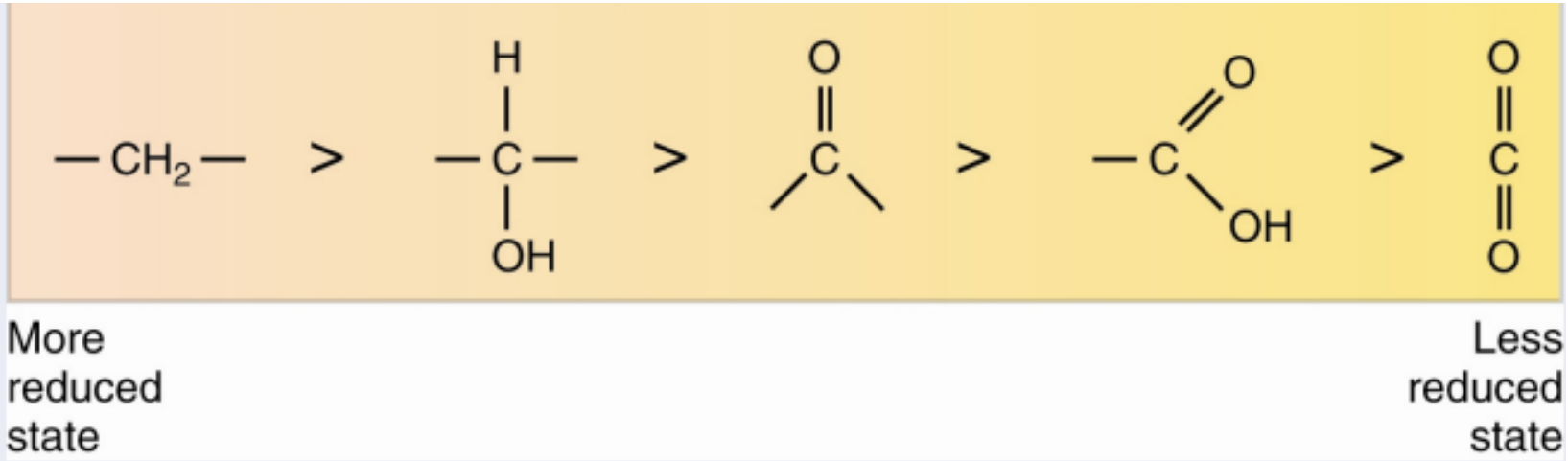
  -Linear, Cyclic, Spiral or Branched.

- Enzymes may appear as:

  - Individual, monofunctional enzymes
  - Multienzyme complexes
  - Multifunctional enzymes



The Substrates of Catabolism Contain Reduced Forms of Carbon



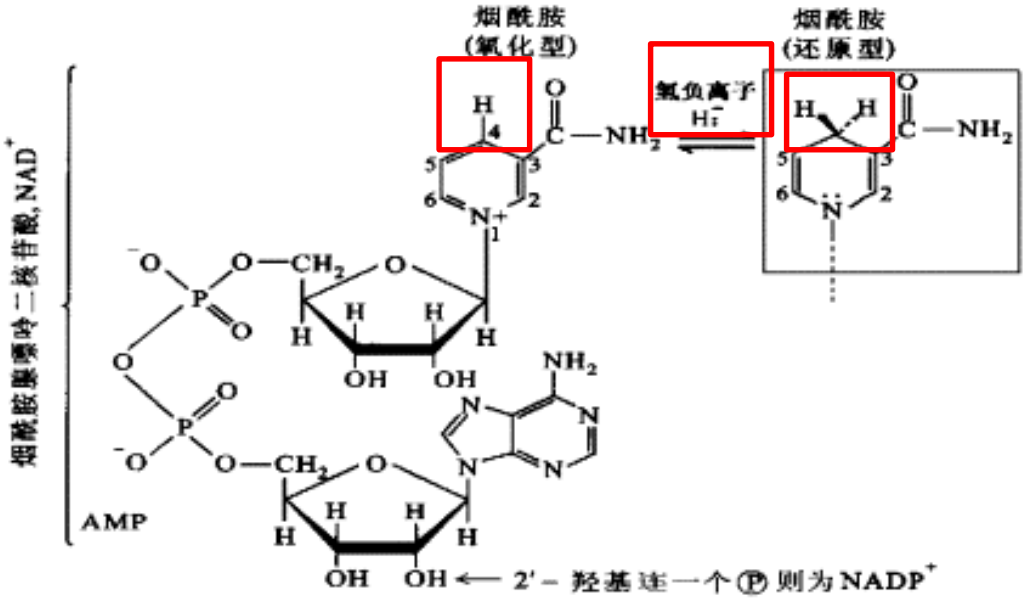
- **-CH_2- are the most form of reduced carbon.**
- **CO_2 is the most oxidized form of carbon.**
- **Oxidation is the loss of electrons.**

ATP Serves in a Cellular Energy Cycle

- **Phototrophs transform light energy into ATP.**
- **In heterotrophs, catabolism produces ATP, which drives activities of cells.**
- **Energy is also conserved as reducing equivalents.**

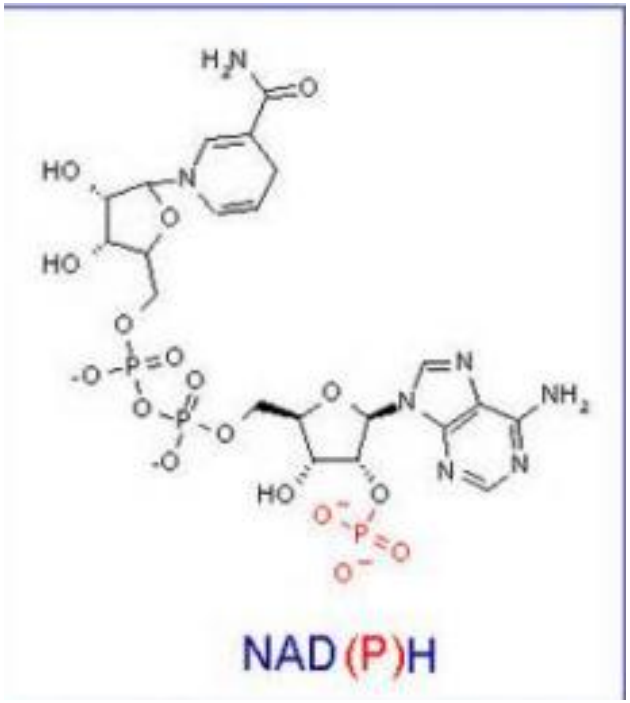
  e.g. NADH, NADPH, $FADH_2$, $FMNH_2$ and $CoQH_2$.
- **The other common energy carrier is acetylSCoA.**

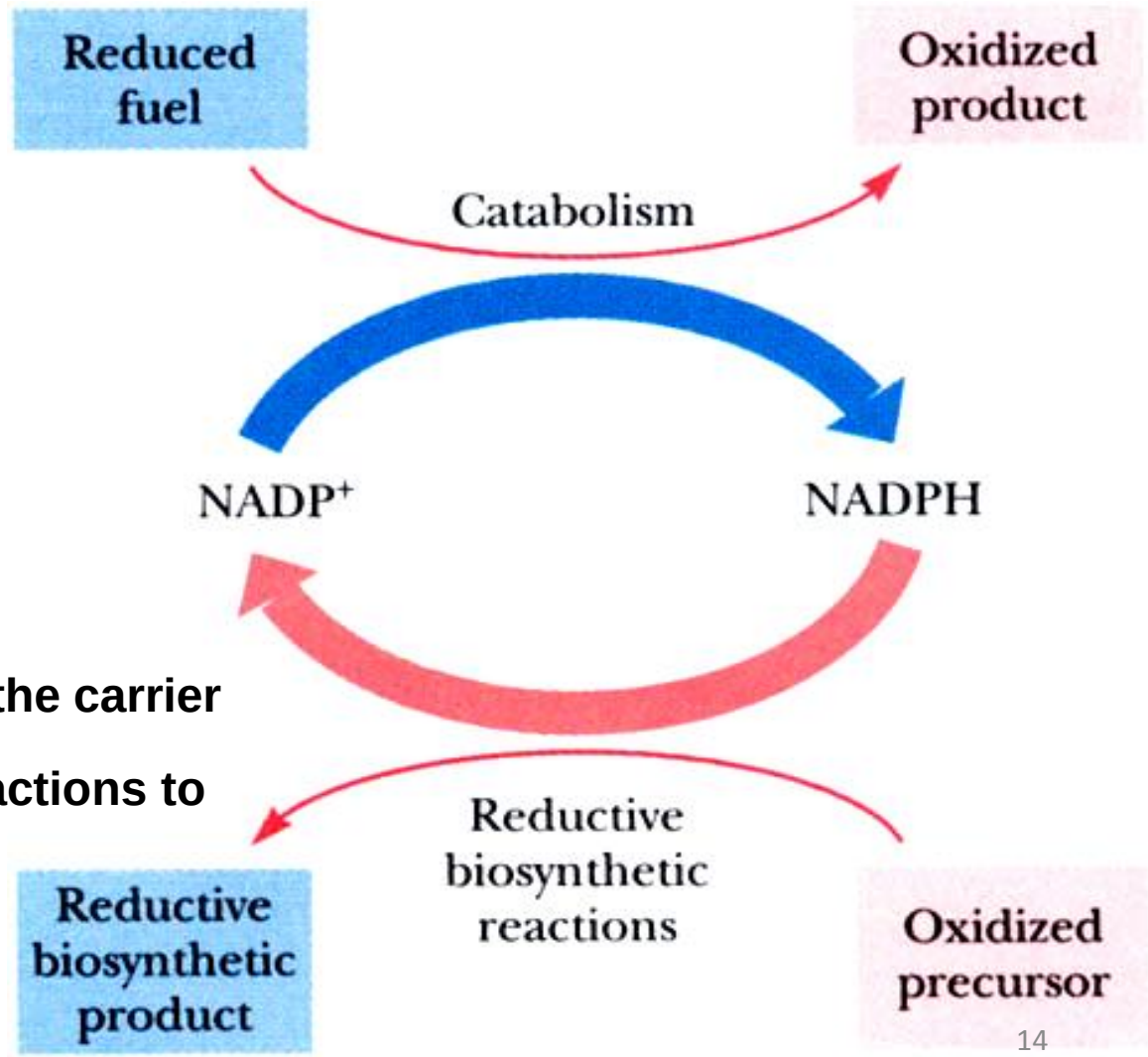
NADH and Catabolism



- **Proteins, carbohydrates, and lipids – in a reduced state.**
- **The oxidation release reducing equivalents from these substrates, often in the form of hydride ions.**
- **These hydrides are transferred to $NAD^+ \rightarrow NADH \rightarrow$ other acceptors $\rightarrow O_2$**

NADPH and Anabolism

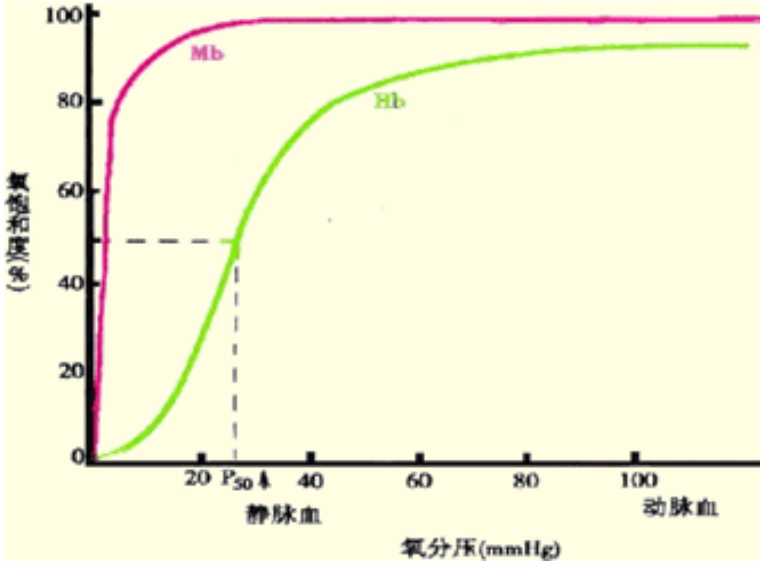


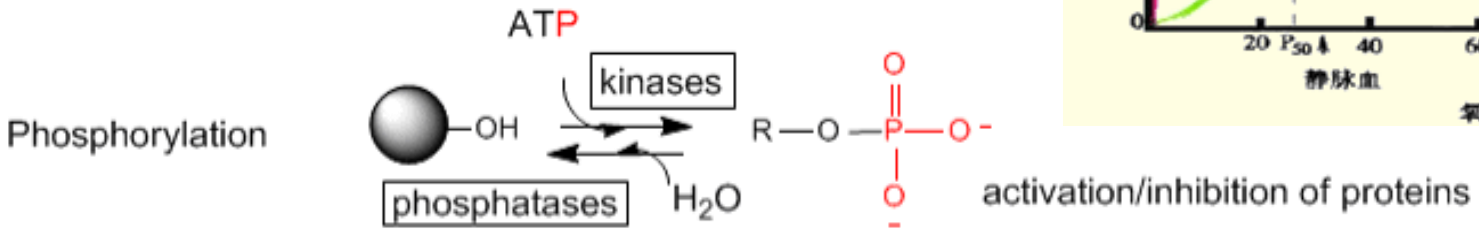


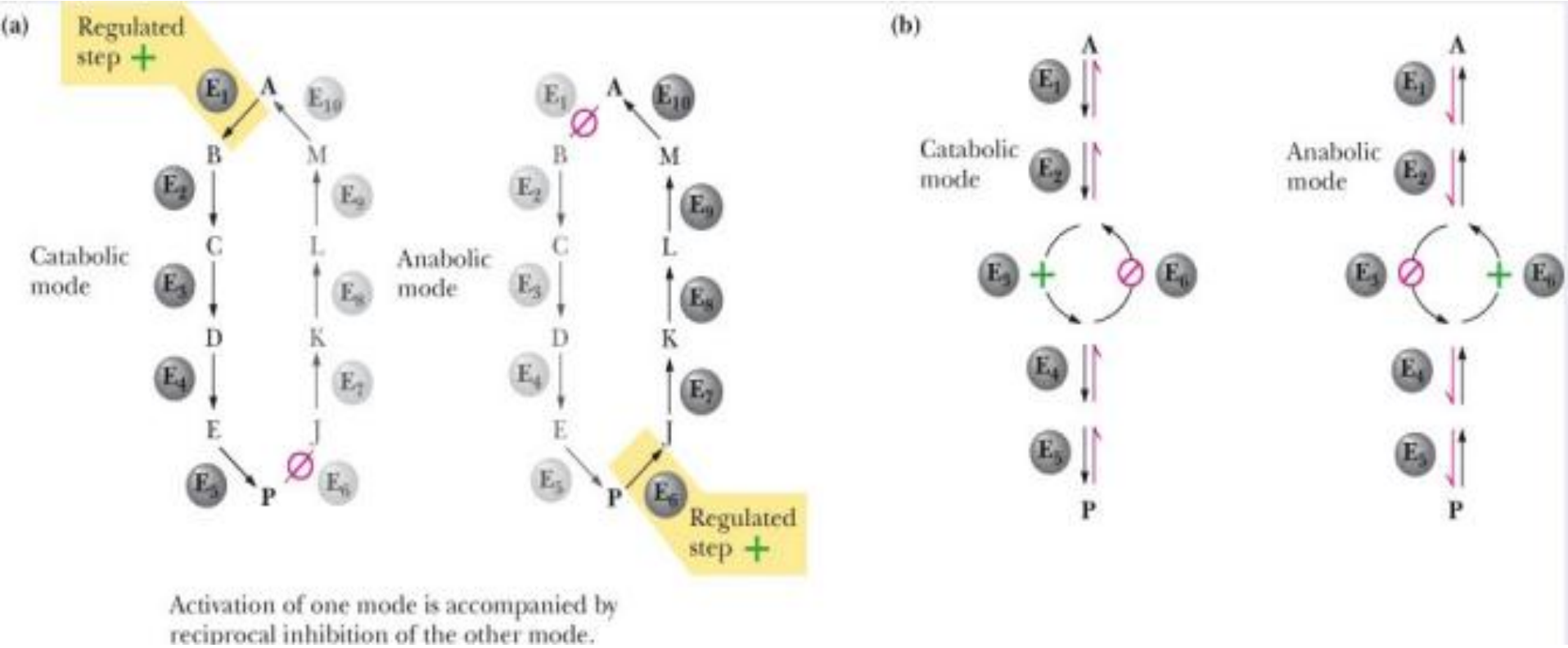
- **NADPH** can be viewed as the carrier of electrons from catabolic reactions to **anabolic** reactions.

4. Metabolic regulation

- **Molecular level:** *e.g.* enzyme
- **Cell level：** Compartmentation（分区）
- **The whole level:** hormone regulation



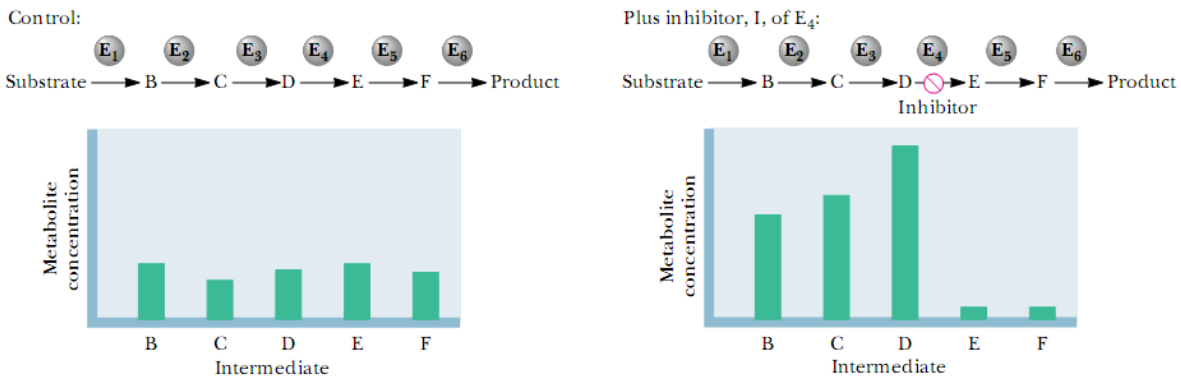




Parallel pathways of catabolism and anabolism must differ in at least one metabolic step in order that they can be regulated independently.

5. What Experiments Can Be Used to Elucidate Metabolic Pathways?

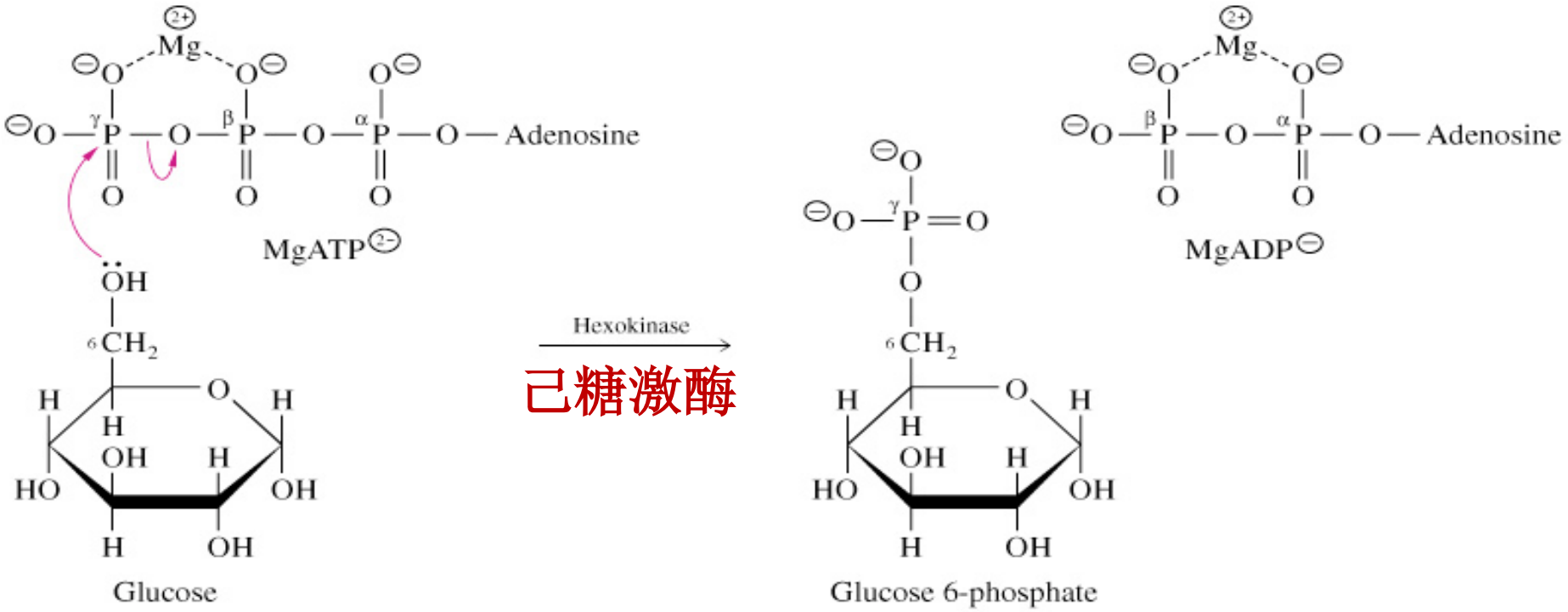
- **Metabolic inhibitors were important tools for elucidating the pathway steps.**



- **Mutations also were used to create specific metabolic blocks.**
- **Isotopic tracers （同位素示踪） as metabolic probes**
- **Mass spectrometry (MS) and nuclear magnetic resonance (NMR) are both powerful techniques for metabolomic analysis.**

- Bioenergetics（生物能学）（自学）

------P302-314



ΔGº′= –16.7 kJ/mol

Craig Venter 人造生命
http://open.163.com/movie/2008/2/G/F/MANER2A9T_MANER8LGF.html

Glycolysis
（糖酵解）

Outline

1. Introduction
2. Catabolism of carbohydrates- three stages
3. Glycolysis
4. Regulation of glycolysis
5. The fates of NADH and pyruvate
6. Other substrates used in glycolysis
7. Summary on glycolysis

1. Introduction

Some Points About Glucose

- **Glucose is very soluble source of quick and ready energy.**
- **Stable and easily transported.**
- **In mammals, the brain uses only glucose under non-starvation conditions.**
- **Glucose is the only source of energy in red blood cells.**
- **Under cellular conditions, 5% of the energy of glucose is released in glycolysis**

Central Role of Glucose

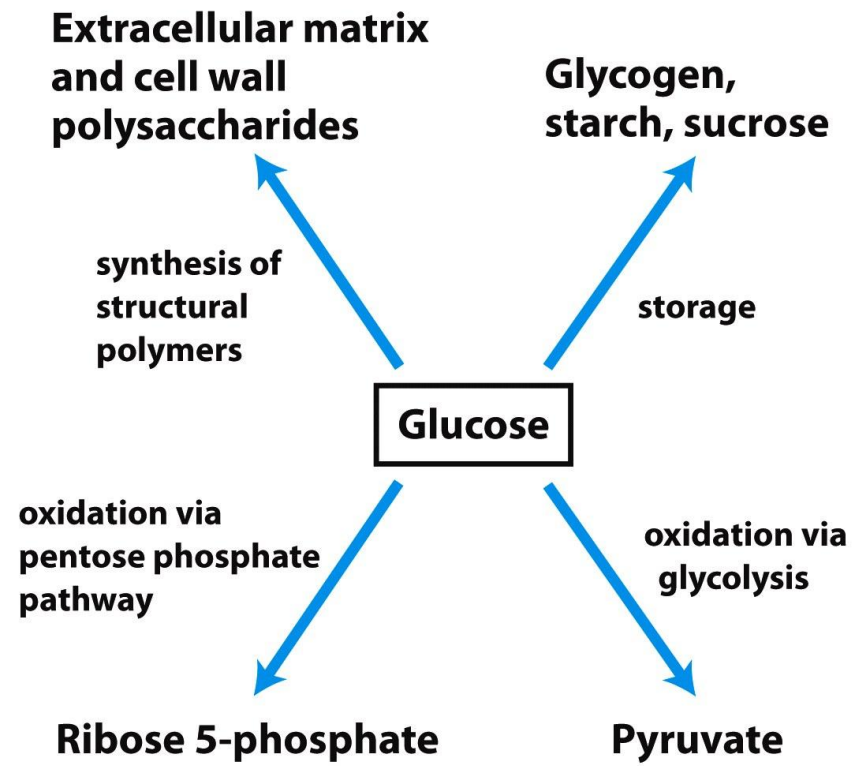


Figure 14-1
Lehninger Principles of Biochemistry, Fifth Edition

2. Catabolism of carbohydrates

➢ **Stage 1: Breakdown by enzymes in the digestive systems followed by intestinal absorption and cellular distribution**

- starch and glycogen

  * salivary and pancreatic amylase

  * maltase, sucrase and lactase → hexose

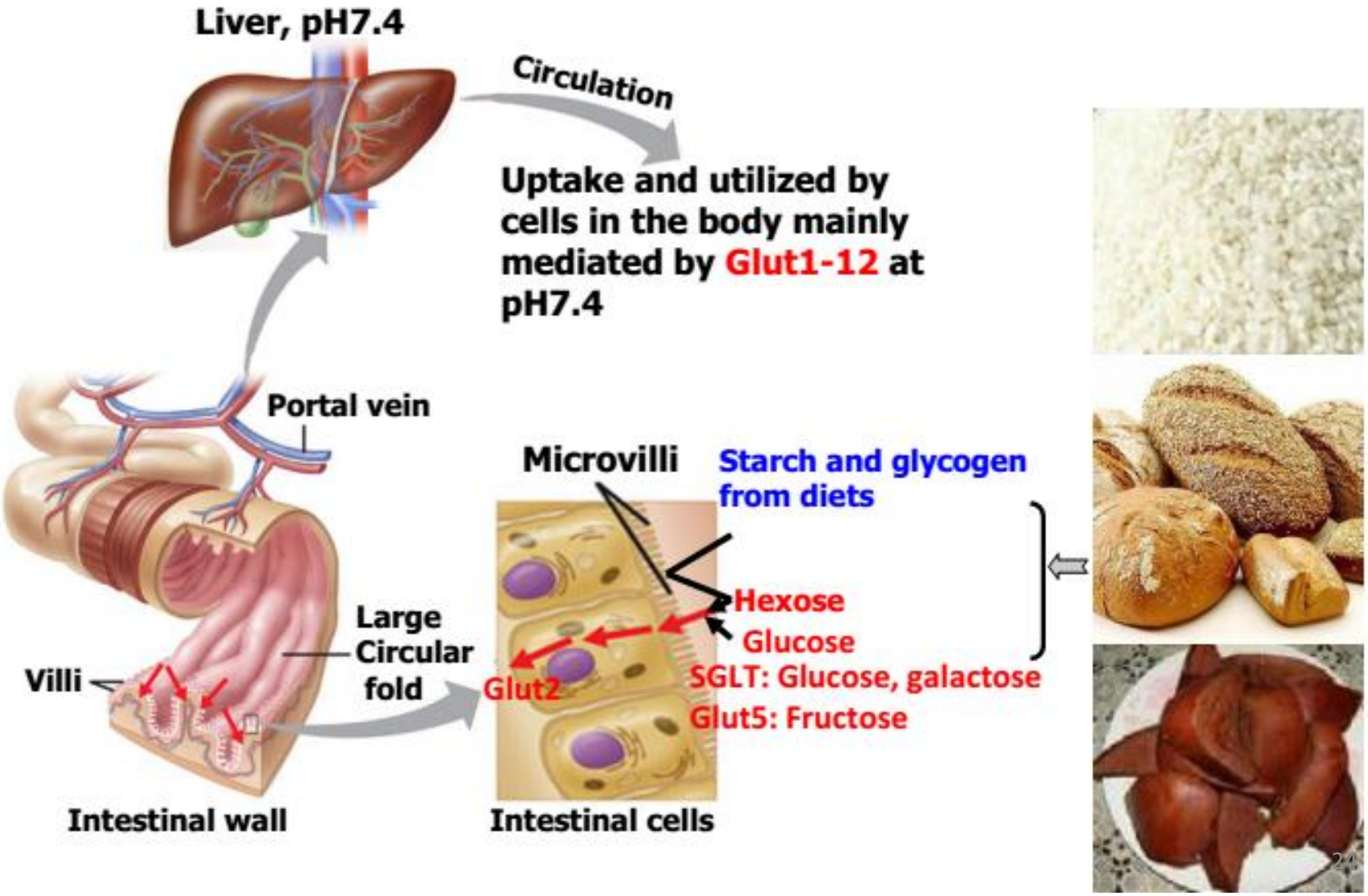
- intestinal absorption: SGLT1 and SGLT2, Glut5 （小肠单糖转运体）

➢ **Stage 2: Glycolysis**

➢ **Stage 3: TCA and oxidative phosphorylation**

Intestinal absorption and cellular distribution

Only **monosaccharides** can be absorbed in mammalian intestine



3. glycolysis : Embden Meyerhof pathway

- **1854-1864, Louis Paster, fermentation—”vital force”（活力）**
- **1897 Hans Buchner and Edward Buchner（1907 Nobel）**
  - **Yeast cell-free extracts , sucrose**
  - **First time that fermentation could occur outside cell**
- **1905 Arthur Harden and William Young**
- **1940,** Gustav **E**mbden, Otto **M**eyerhof, Carl Newberg, Jacob Parnas, Otto Warburg, **Gerty Cori** and **Carl Cori** (1947 Nobel)

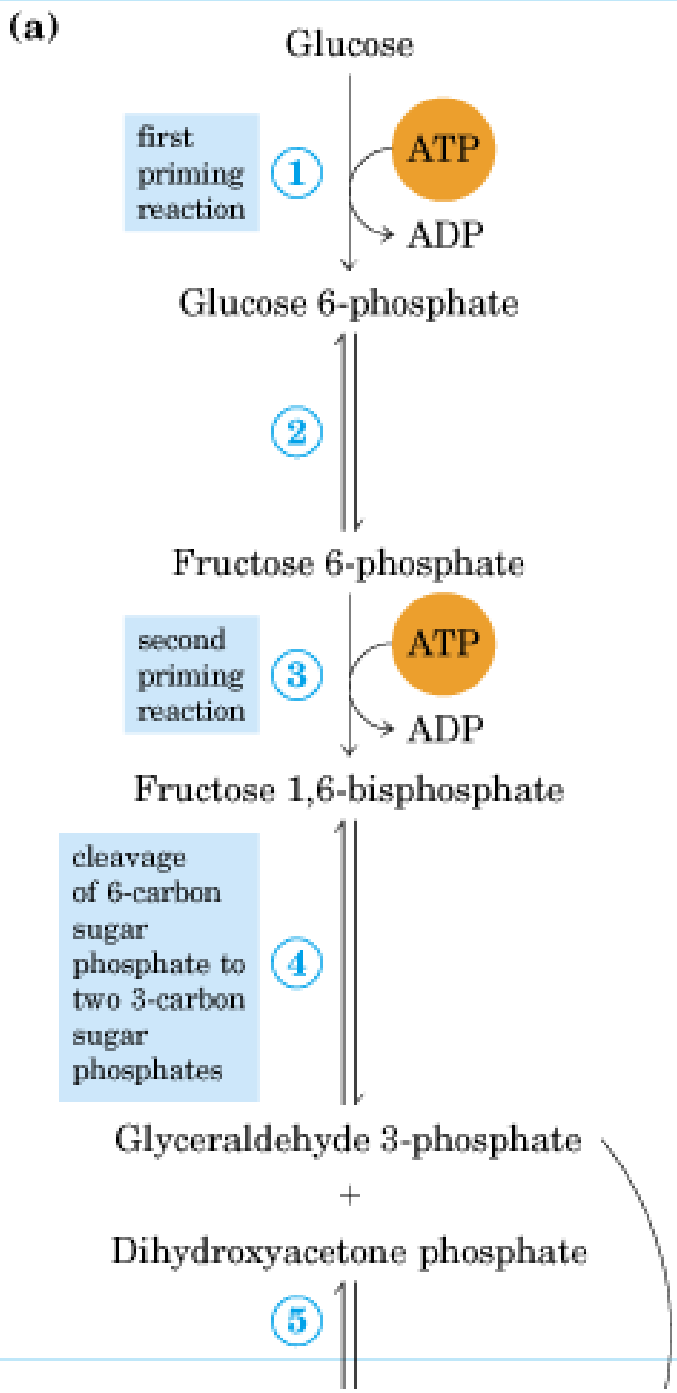
Importance of Glycolysis

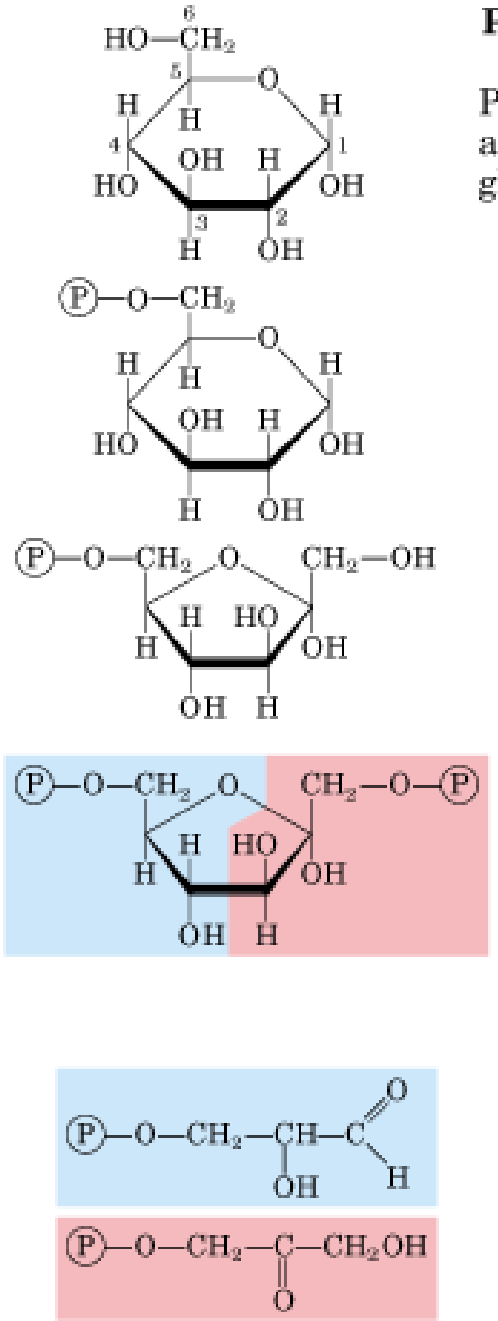
- Central energy-yielding path
- Provides precursors for many biosynthetic paths
- Some tissues (brain, kidney medulla, rapidly contracting skeletal muscles) and some cells (erythrocytes cells)---glucose is the only source of metabolic energy

- **10 steps** of glycolysis （in the **cytosol**）
  - **The starting point ——— glucose**
  - **The process ends ——— formation of two pyruvate**
  - **Aerobic or anaerobic**

- **Glycolysis is the major source of pyruvate** → **TCA** → **ATP+ NADH**

Reaction	Enzyme
1. Glucose + ATP ⟶ Glucose6-phosphate + ADP + $H^{\oplus}$	Hexokinase,glucokinase
2. Glucose6-phosphate ⇌ Fructose 6-phosphate	Glucose-6-phosphateisomerase
3. Fructose6-phosphate + ATP ⟶ Fructose1,6-*bis*phosphate + ADP + $H^{\oplus}$	Phosphofructokinase-1
4. Fructose1,6-*bis*phosphate ⇌ Dihydroxyacetone phosphate + Glyceraldehyde 3-phosphate	Aldolase
5. Dihydroxyacetone phosphate ⇌ Glyceraldehyde 3-phosphate	Triose phosphateisomerase
6. Glyceraldehyde 3-phosphate + $NAD^{\oplus}$ + iP ⇌ 1,3-*Bis*phosphoglycerate + NADH + $H^{\oplus}$	Glyceraldehyde 3-phosphatedehydrogenase
7. 1,3-*Bis*phosphoglycerate + ADP ⇌ 3-Phosphoglycerate + ATP	Phosphoglycerate kinase
8. 3-Phosphoglycerate ⇌ 2-Phosphoglycerate	Phosphoglyceratemutase
9. 2-Phosphoglycerate ⇌ Phosphoenolpyruvate + H_2O	Enolase
10. Phosphoenolpyruvate + ADP + $H^{\oplus}$ ⟶ Pyruvate + ATP	Pyruvate kinase



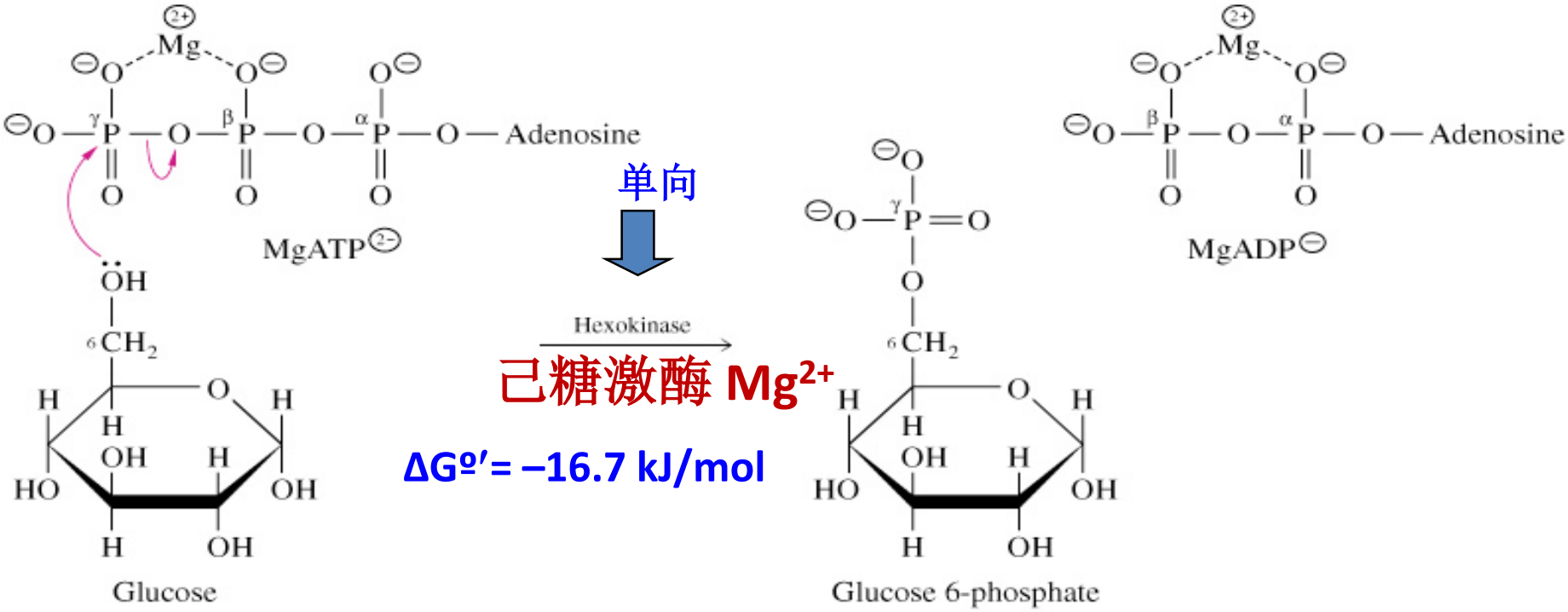


Preparatory phase

Phosphorylation of glucose and its conversion to glyceraldehyde 3-phosphate

Glycolysis 1st Stage

Step 1. Hexokinase（己糖激酶）reaction



ΔG for the 1st step is large and negative.

- **Four kinases in glycolysis: steps 1,3,7, and 10,** all of which require Mg^{2+} and have a similar mechanism.

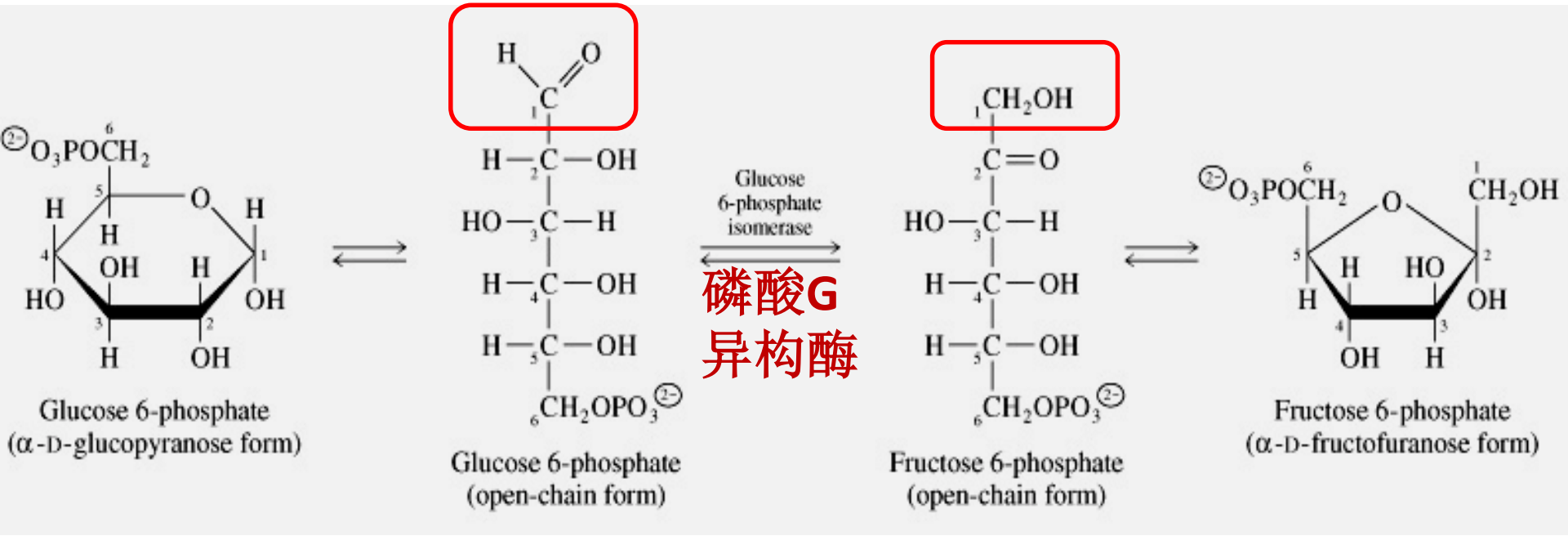
Properties of hexokinases

- **Hexokinases I, II, III** （**broad substrate specificity**）are active at normal glucose concentrations (K_m ~10^{-6} to 10^{-4}M)

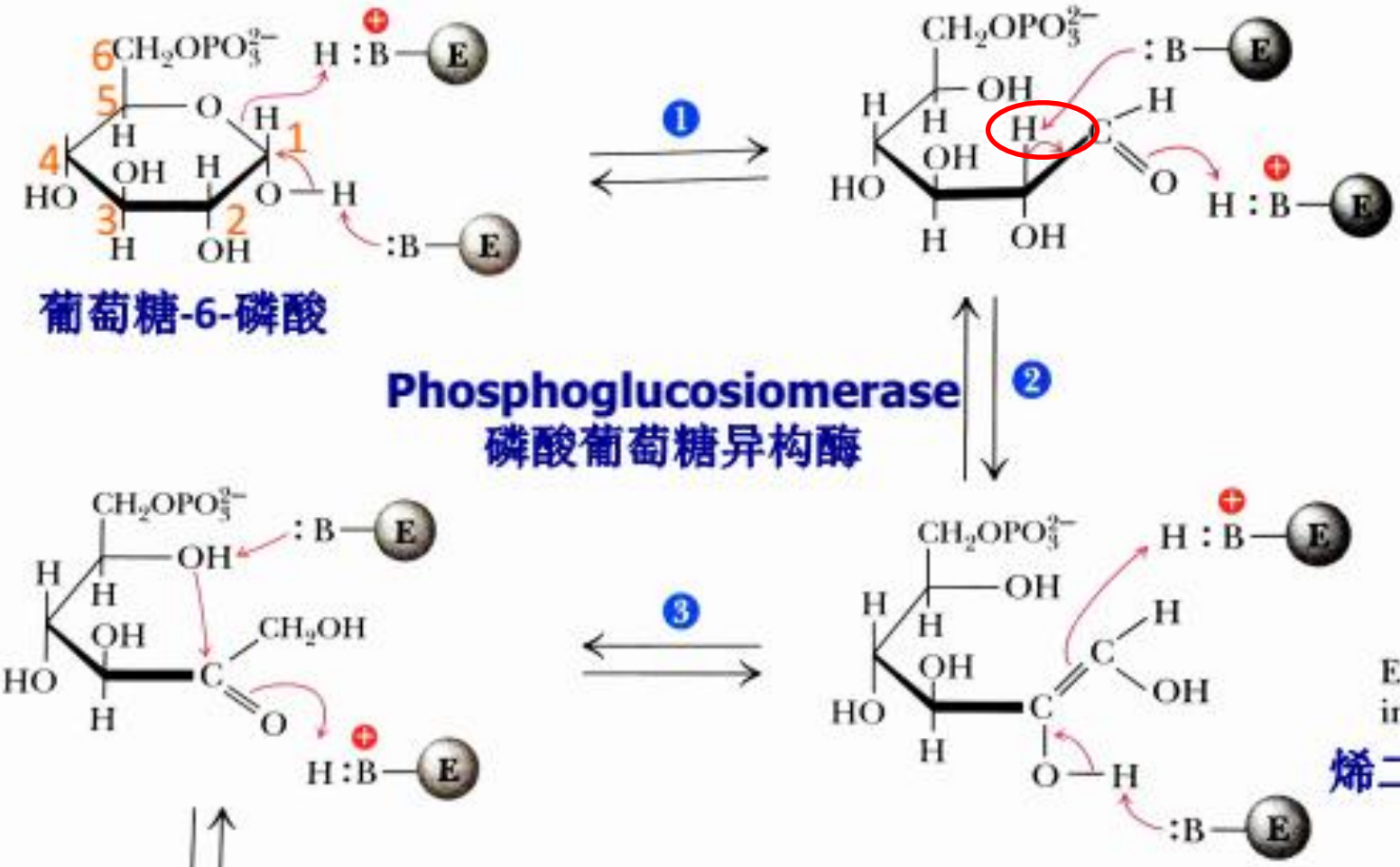
- **Hexokinase IV** (**Glucokinase**, K_m ~10^{-2}M) in the hepatocyte is active at **higher** glucose levels, allows the liver to respond to large increases in blood glucose

Glycogen（糖原）

Step 2. Conversion of G6P to F6P



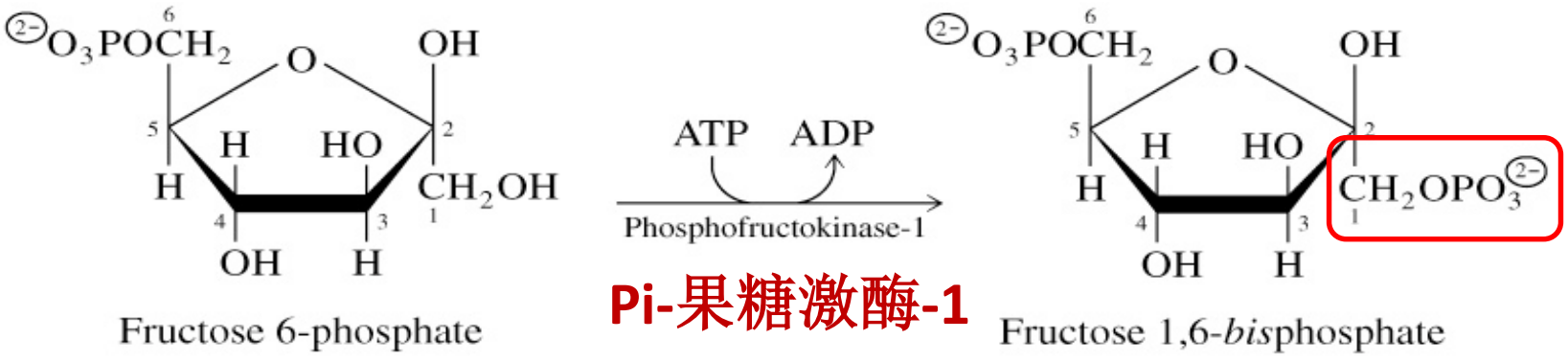
$\Delta G^{o\prime}$= 1.7 kJ/mol 可逆



- Make C1 phosphorylation easier
- Activate C-3 and thus facilitate the C3-C4 bond cleavage

critical regulatory point

Step 3. Phosphofructokinase-1 (PFK-1) Reaction



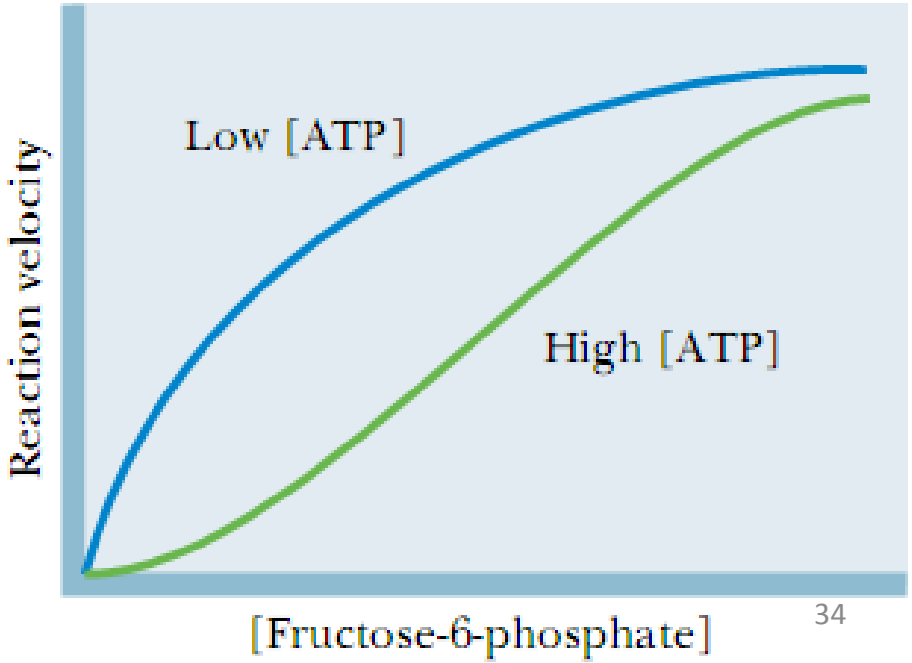
Pi-果糖激酶-1

果糖-6-磷酸　　$\Delta G^{o\prime} = -14.2$ kJ/mol　　果糖-1，6-二磷酸　　(11.3)

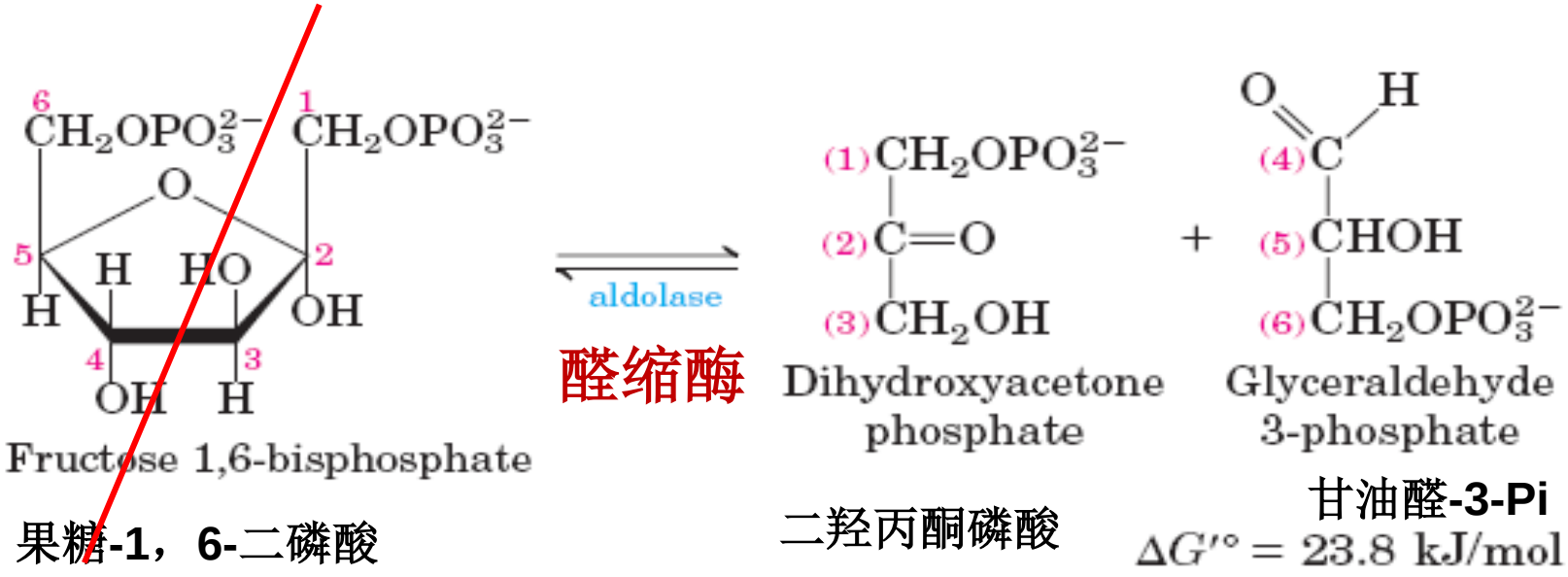
- **PFK-1** ---- **irreversible** and **critical regulatory point**
- **PFK-2** ---- **fructose 2,6-*bis*phosphate (F2,6BP)**

Phosphofructokinase （PFK）

- MW 360,000
- **Rate-limiting step in glycolysis**
- **Major** control point: allosteric regulation
  - High ATP inhibits
  - High AMP, ADP stimulates
  - Other “fuels” alter activity
  - Fru-2,6-bisP
  - hormonal signal



Step 4. Aldolase Reaction



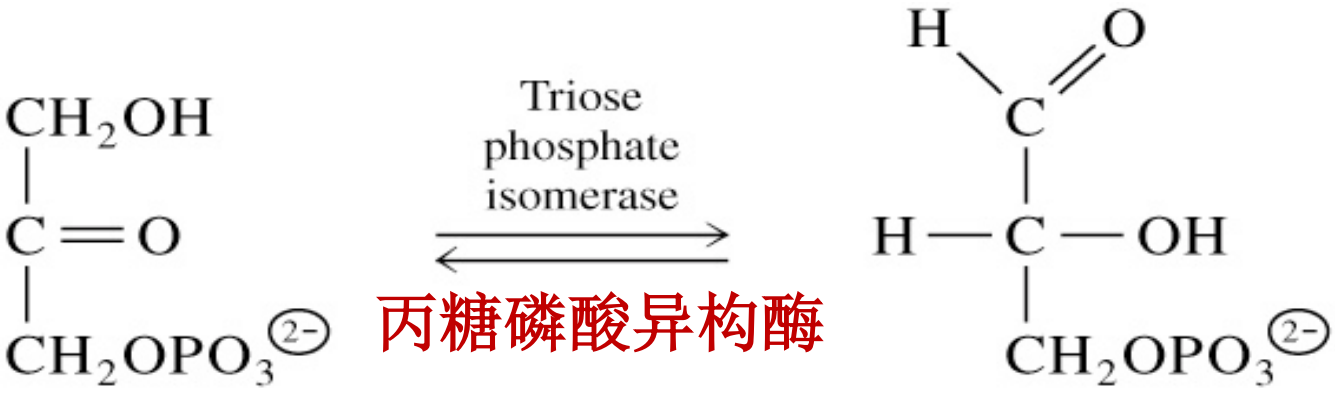
果糖-1，6-二磷酸

二羟丙酮磷酸

甘油醛-3-Pi

$\Delta G'° = 23.8$ kJ/mol

Step 5. Reaction of Triose phosphate isomerase



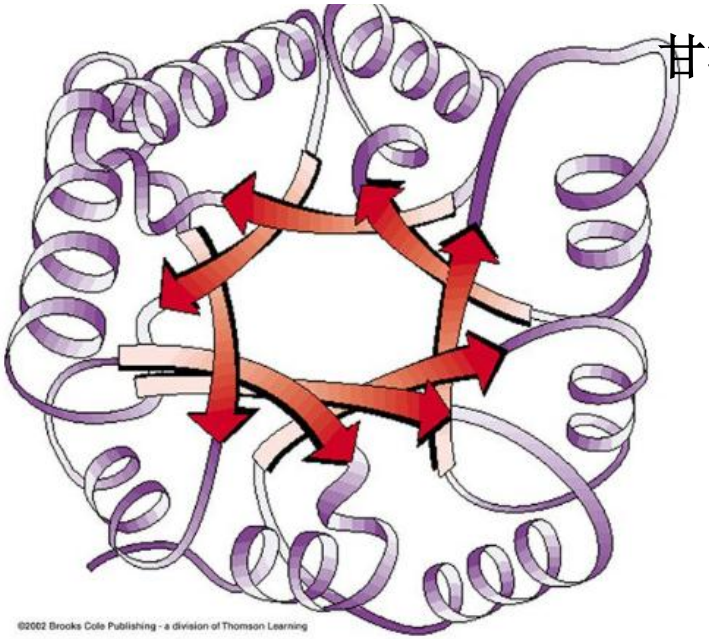
丙糖磷酸异构酶

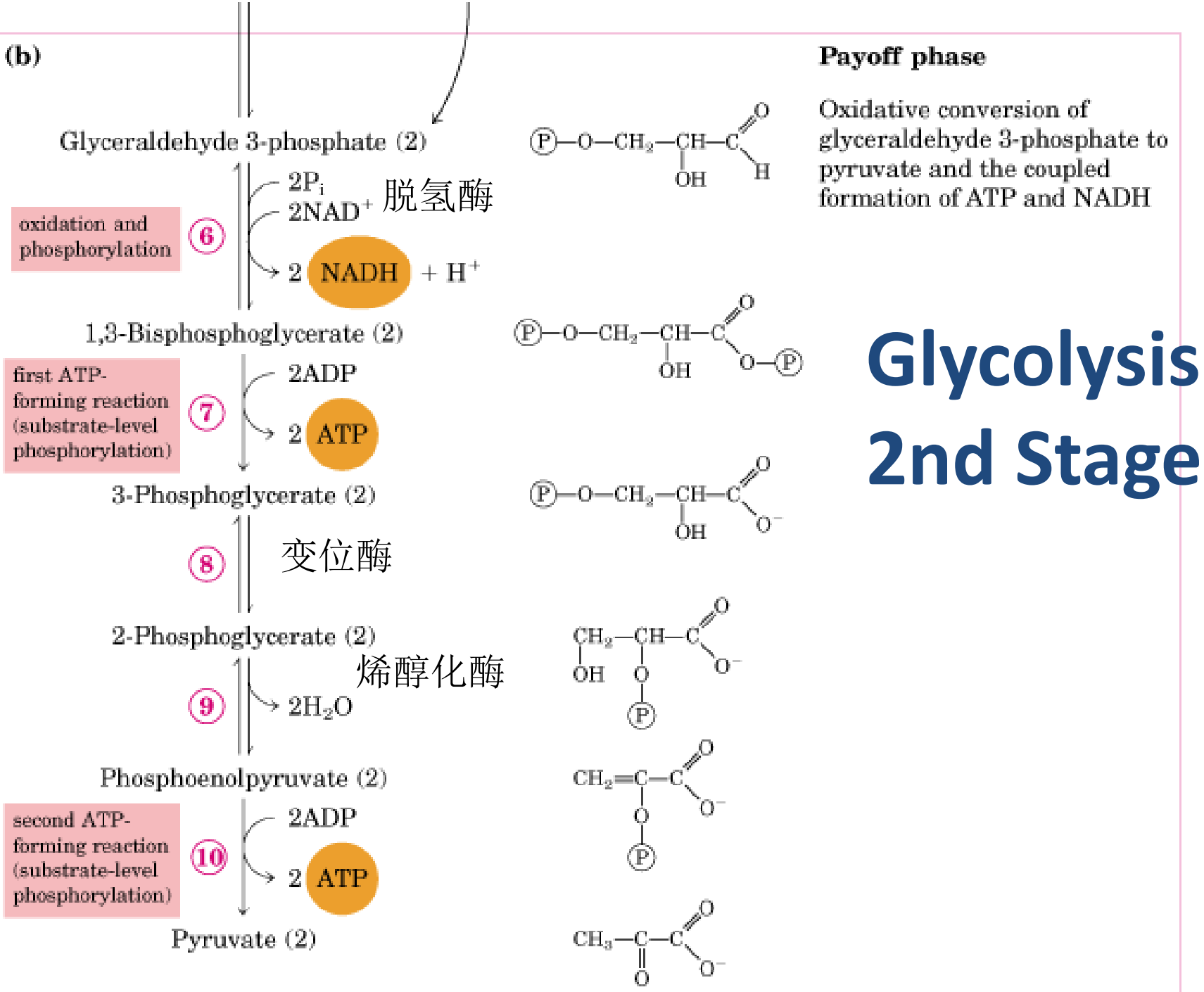
Dihydroxyacetone phosphate

二羟丙酮磷酸

Glyceraldehyde 3-phosphate

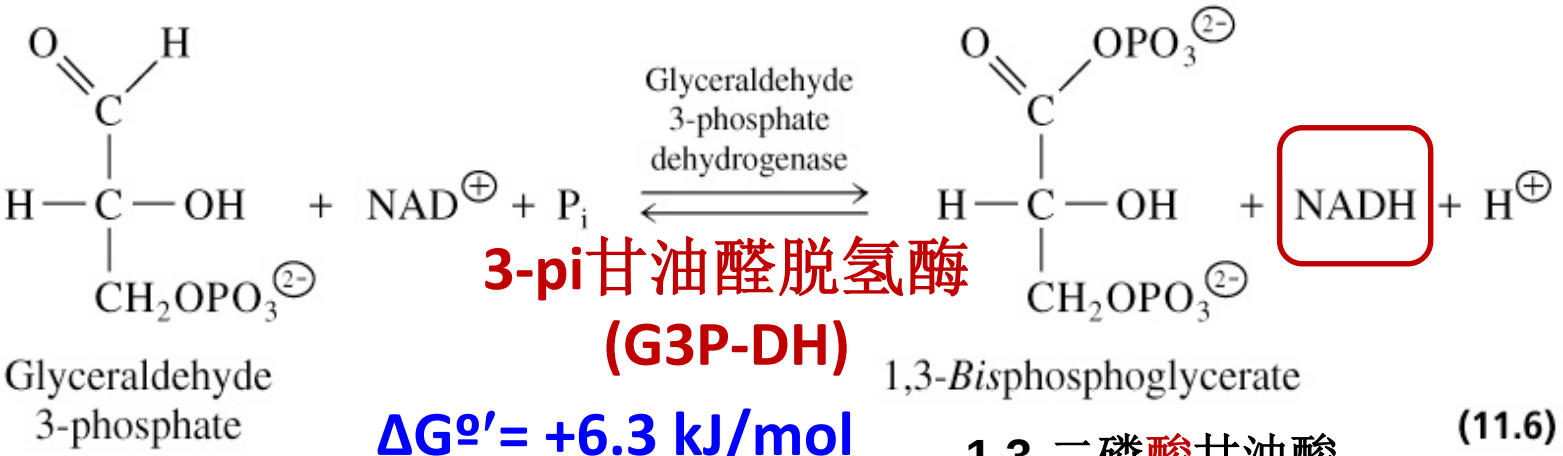
甘油醛-3-Pi





Glycolysis 2nd Stage

Step 6. Reaction of Glyceraldehyde 3-Phosphate Dehydrogenase (GAPDH)



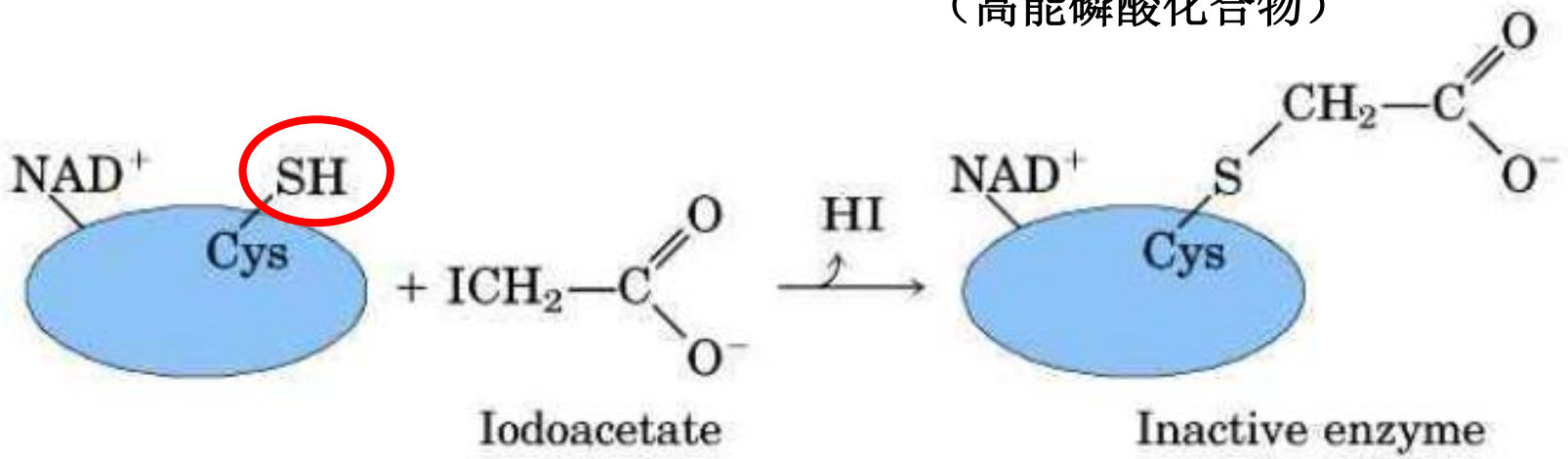
3-pi甘油醛脱氢酶 (G3P-DH)

ΔGº′= +6.3 kJ/mol

甘油醛-3-Pi

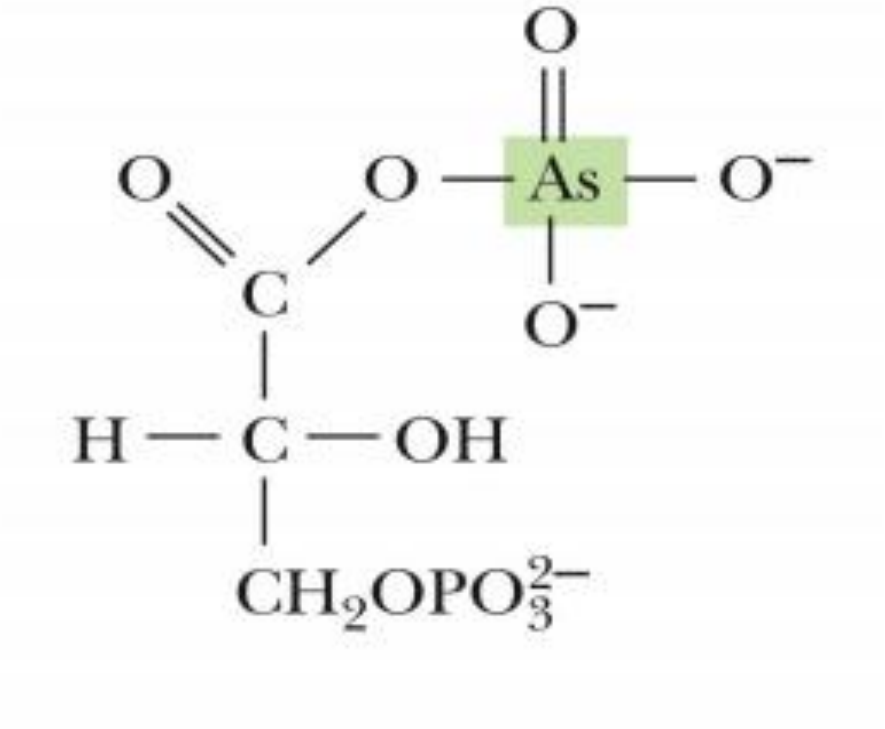
1,3-二磷酸甘油酸

（高能磷酸化合物）

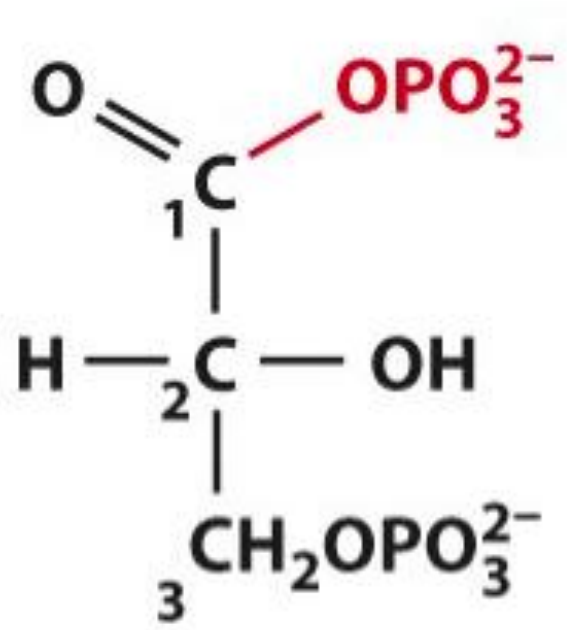


碘乙酸

G3P-DH is the site of action of arsenate (砷酸盐)

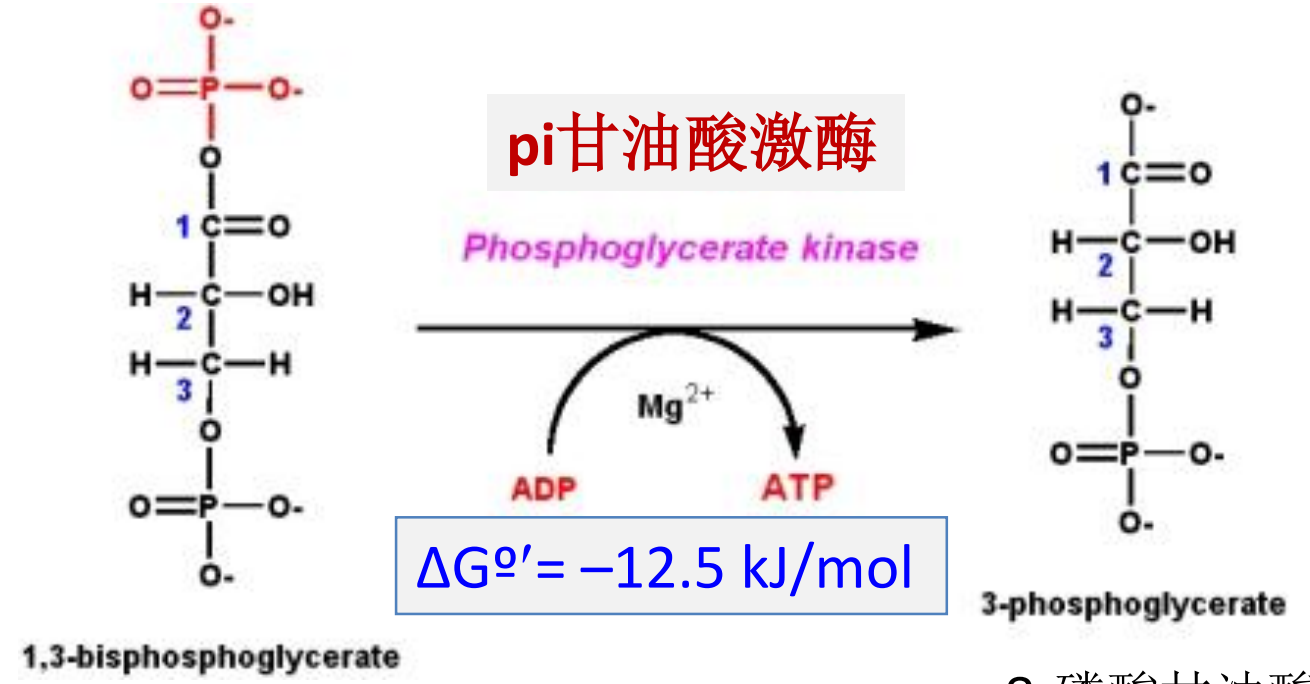


1-Arseno-3-phosphoglycerate



1,3-Bisphosphoglycerate

Step 7. Phosphoglycerate kinase reaction

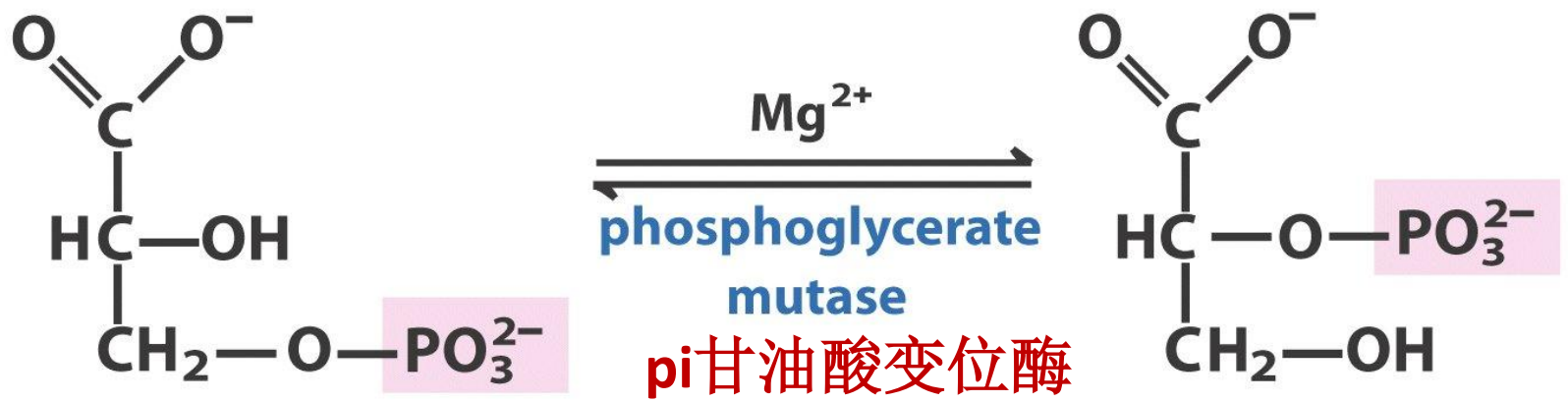


1,3-二磷酸甘油酸

3-磷酸甘油酸

➢ **Substrate-level phosphorylation --- ATP**

Step 8. Phosphoglycerate mutase 变位酶reaction



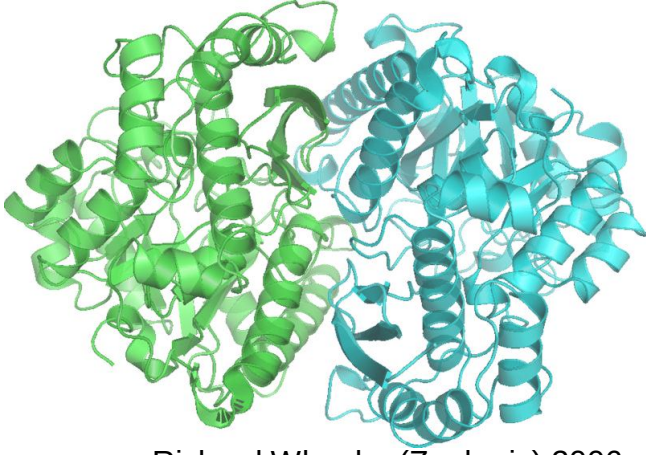
pi甘油酸变位酶

3-Phosphoglycerate

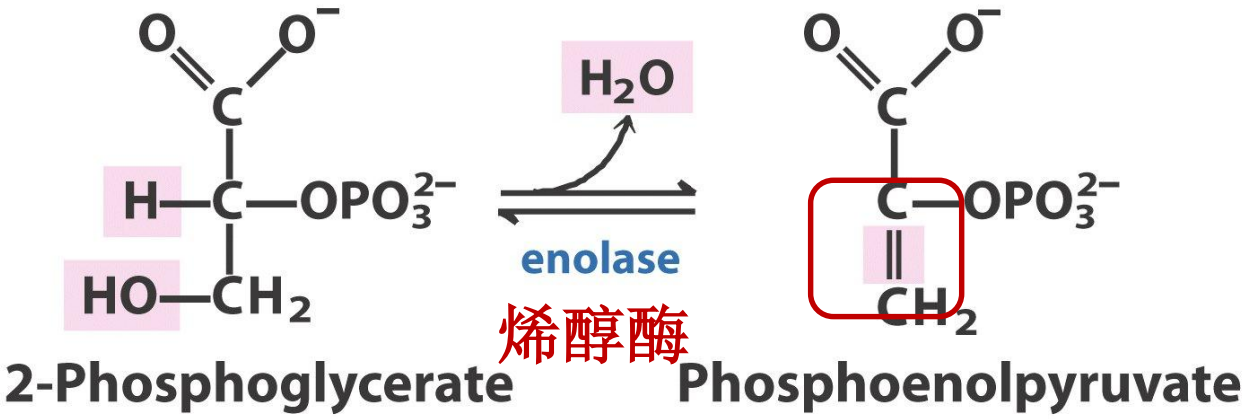
2-Phosphoglycerate

$\Delta G'^{\circ} = 4.4$ kJ/mol

Step 9. Enolase 烯醇酶 reaction

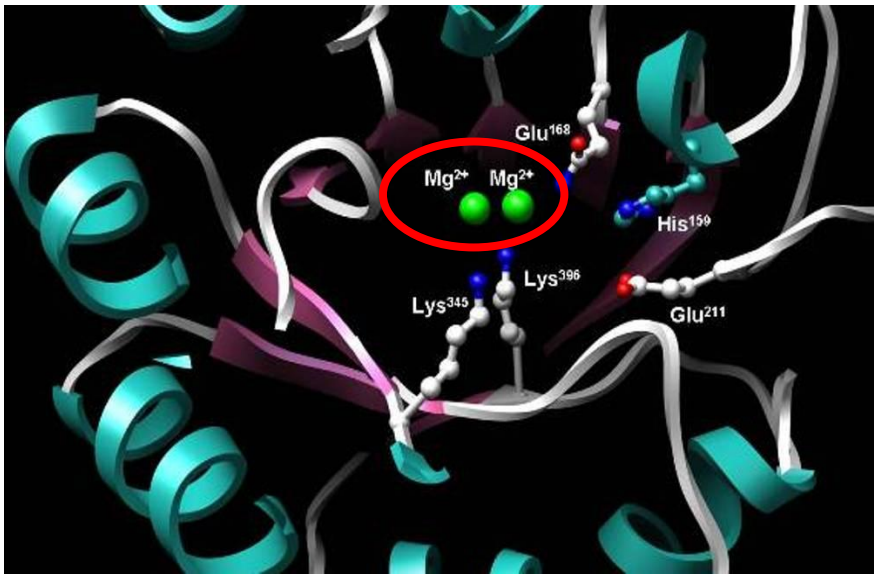


Richard Wheeler (Zephyris) 2006

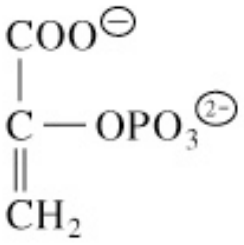


$\Delta G'^{\circ} = 7.5$ kJ/mol

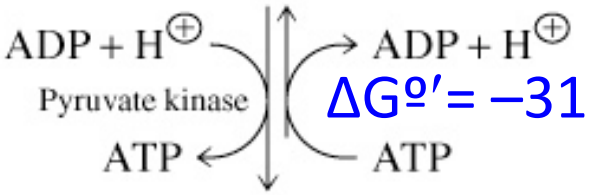
- Fluoride ion（氟化物） inhibits enolase



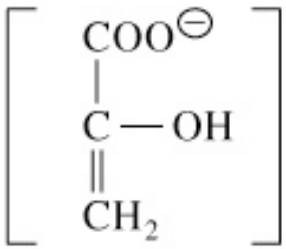
Step 10. Pyruvate kinase reaction



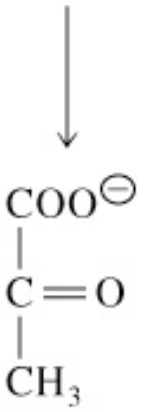
Phosphoenolpyruvate



ΔG°′ = –31.4 kJ/mol



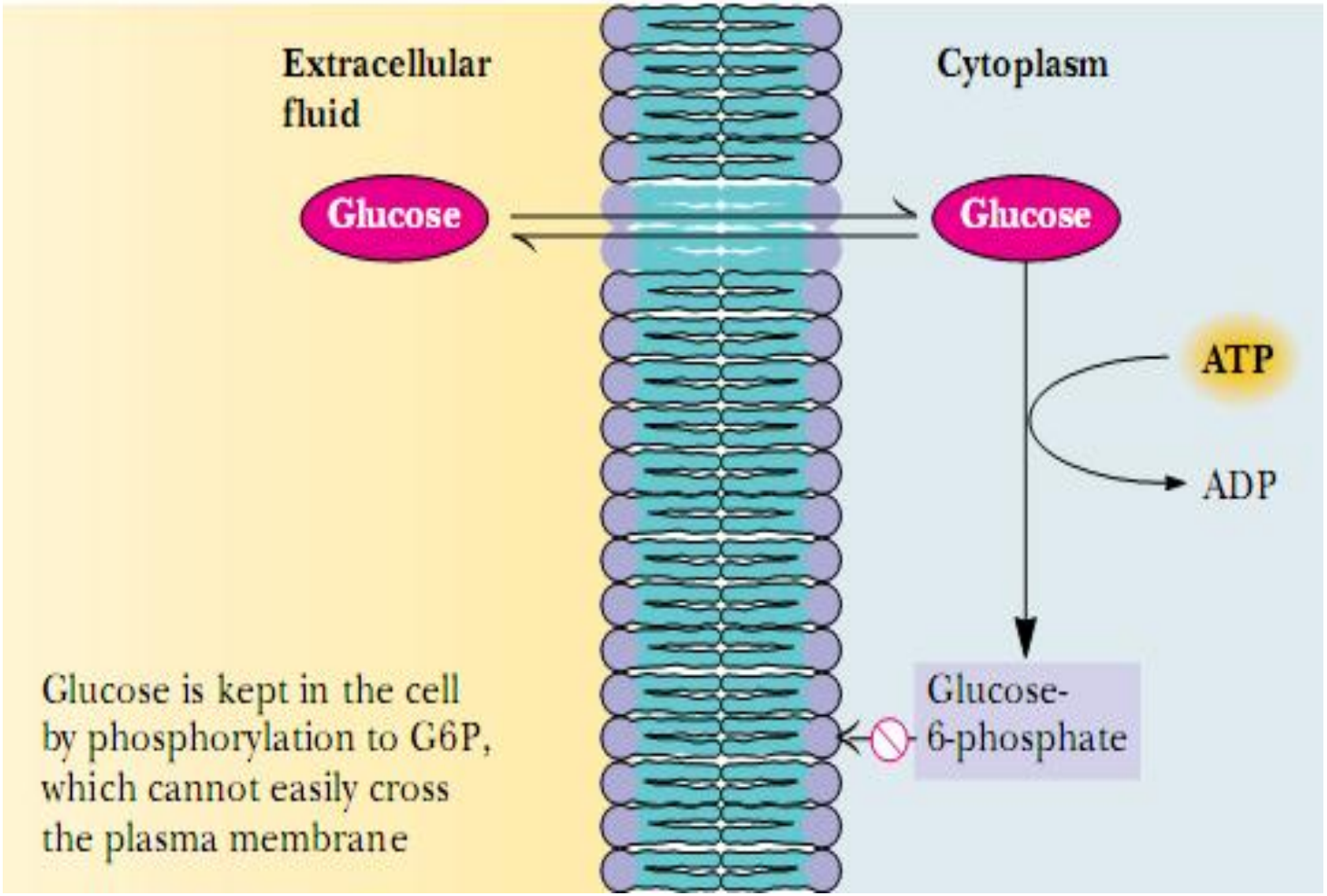
Enolpyruvate



Pyruvate

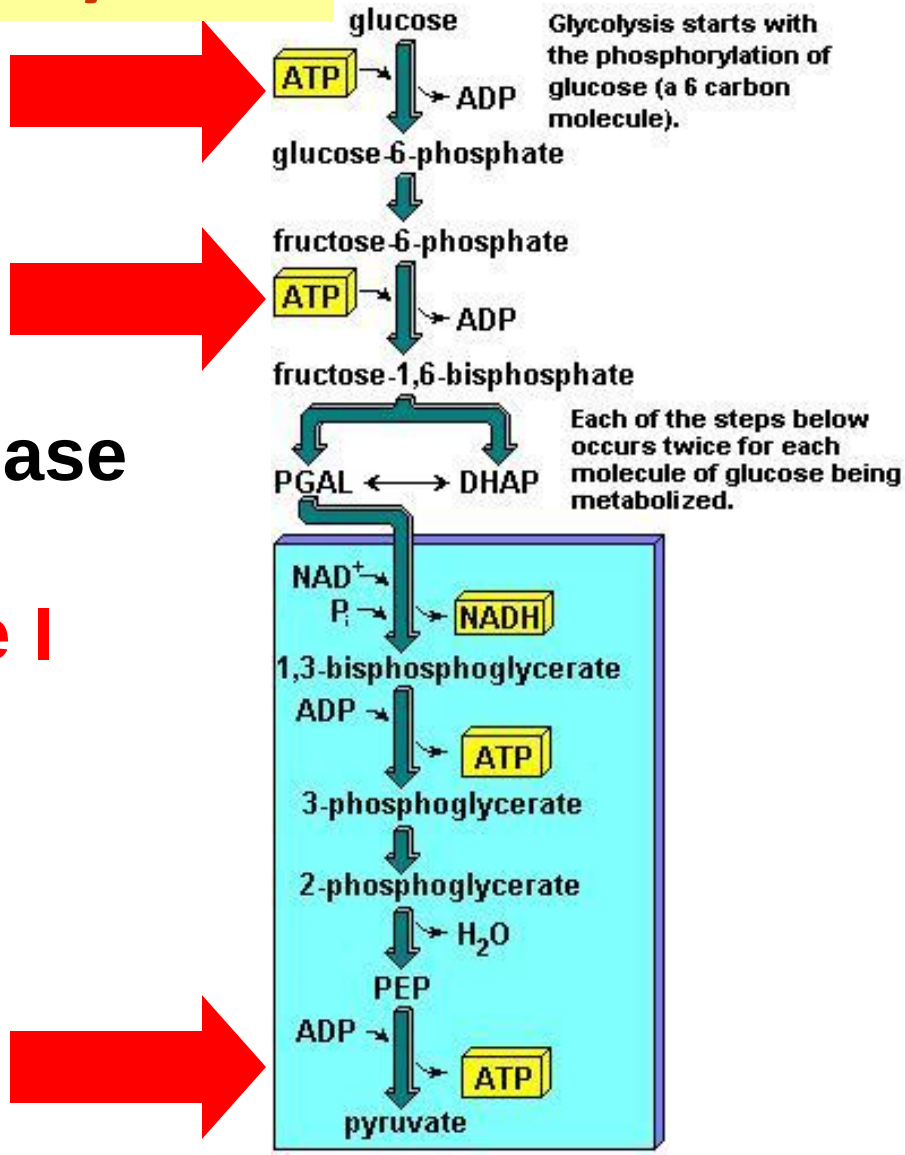
- **substrate-level phosphorylation**
- irreversible reaction
- allosteric modulators and covalent modification
- Pyruvate kinase gene is regulated by various hormones and nutrients

All ten intermediates are phosphorylated， why？



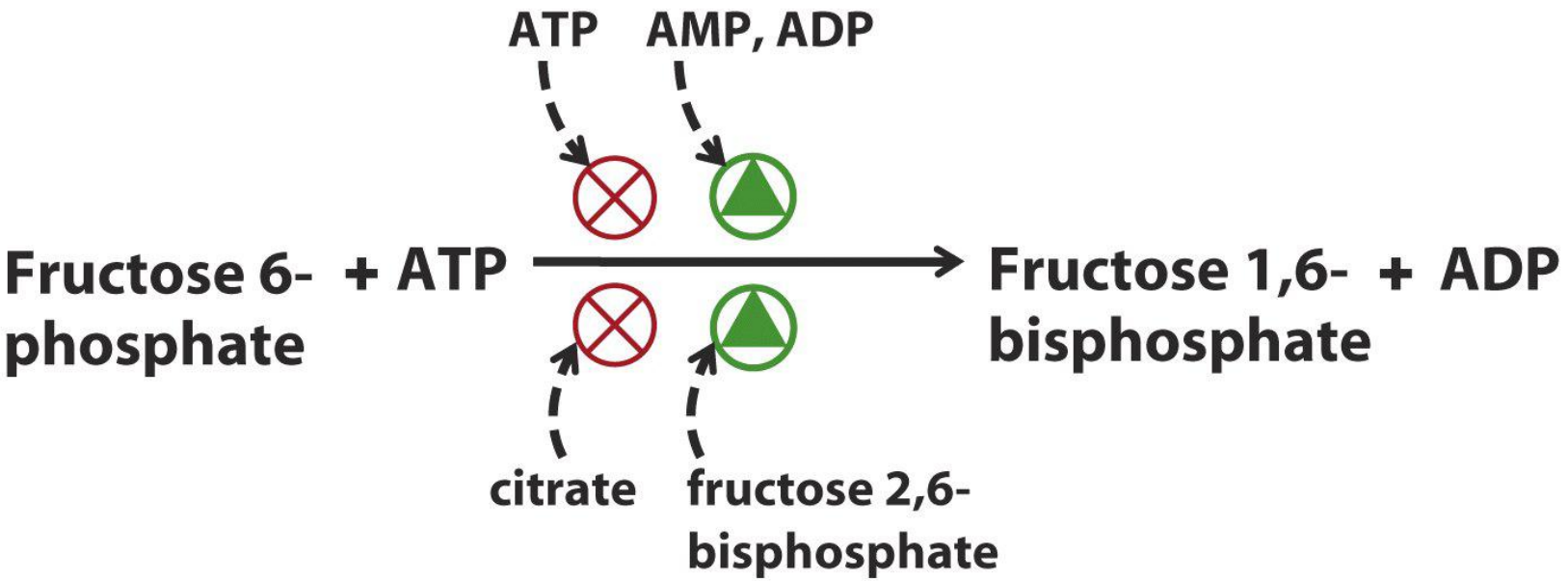
4. Regulation of glycolysis

- Hexokinase/Glucokinase
- Phosphofructokinase I
- Pyruvate Kinase



Control of PFK-1（磷酸果糖激酶-1）

- **Many allosteric effectors**



critical regulatory point

- ATP is an allosteric inhibitor of PFK-1.
- Two binding sites: substrate and allosteric site.

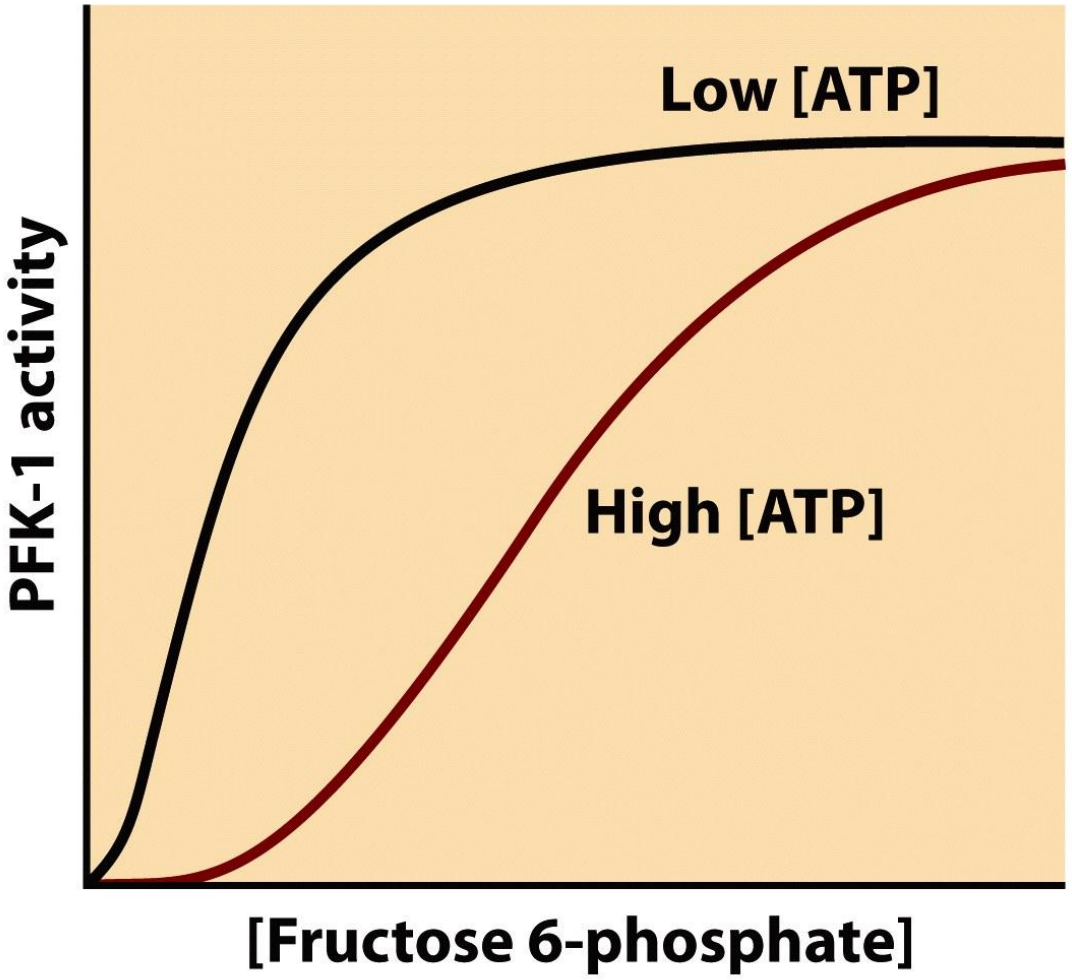
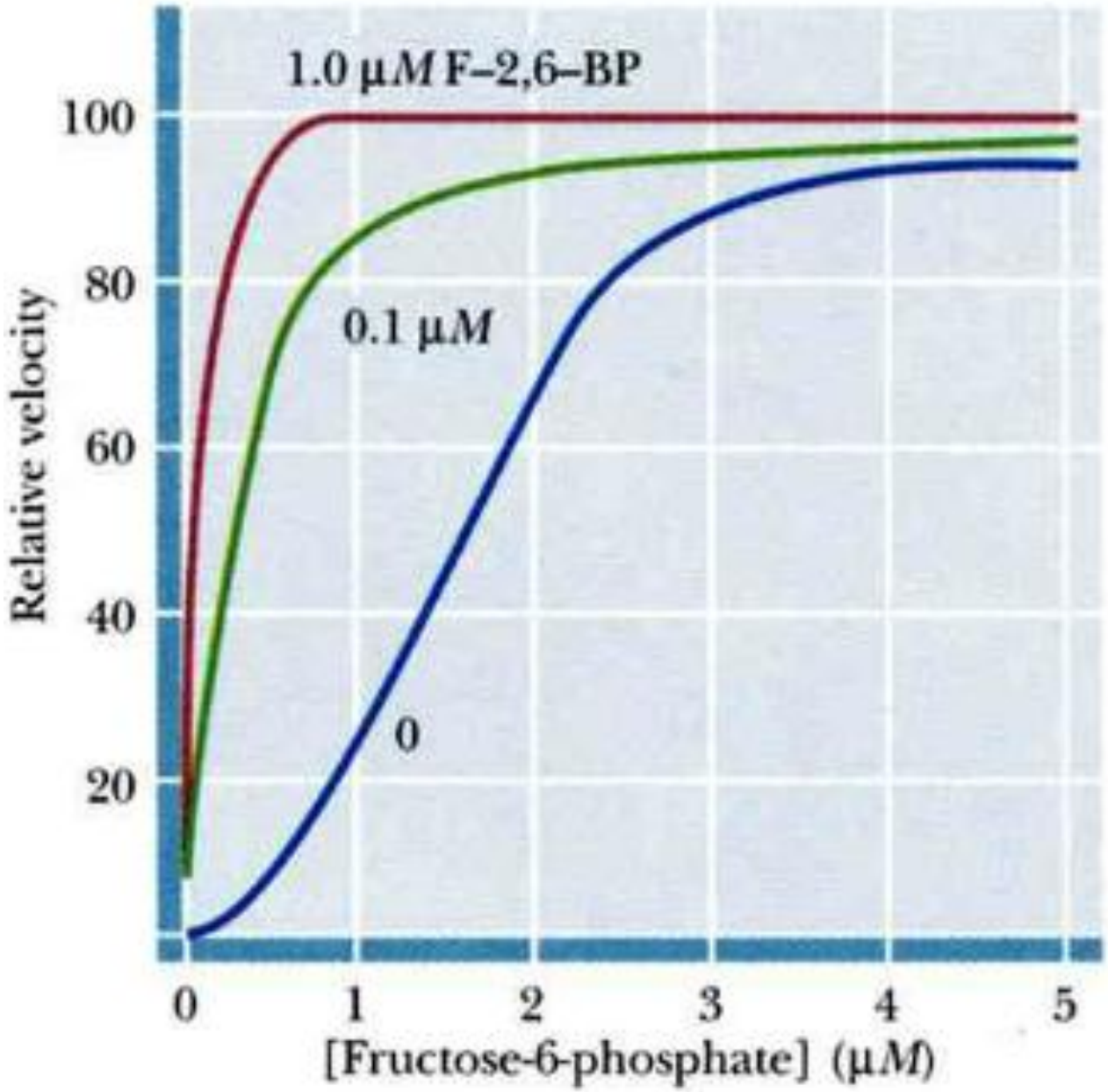
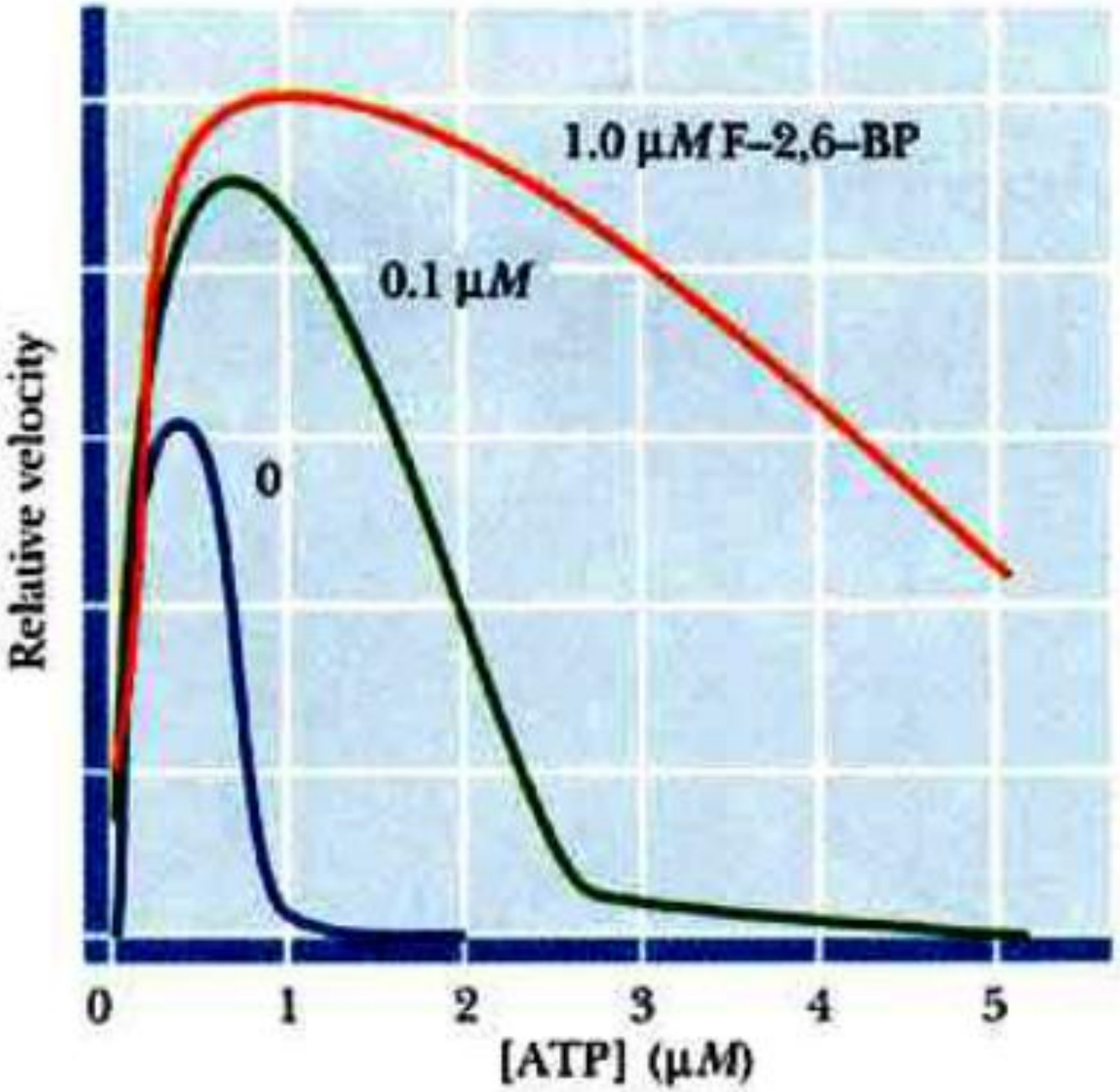
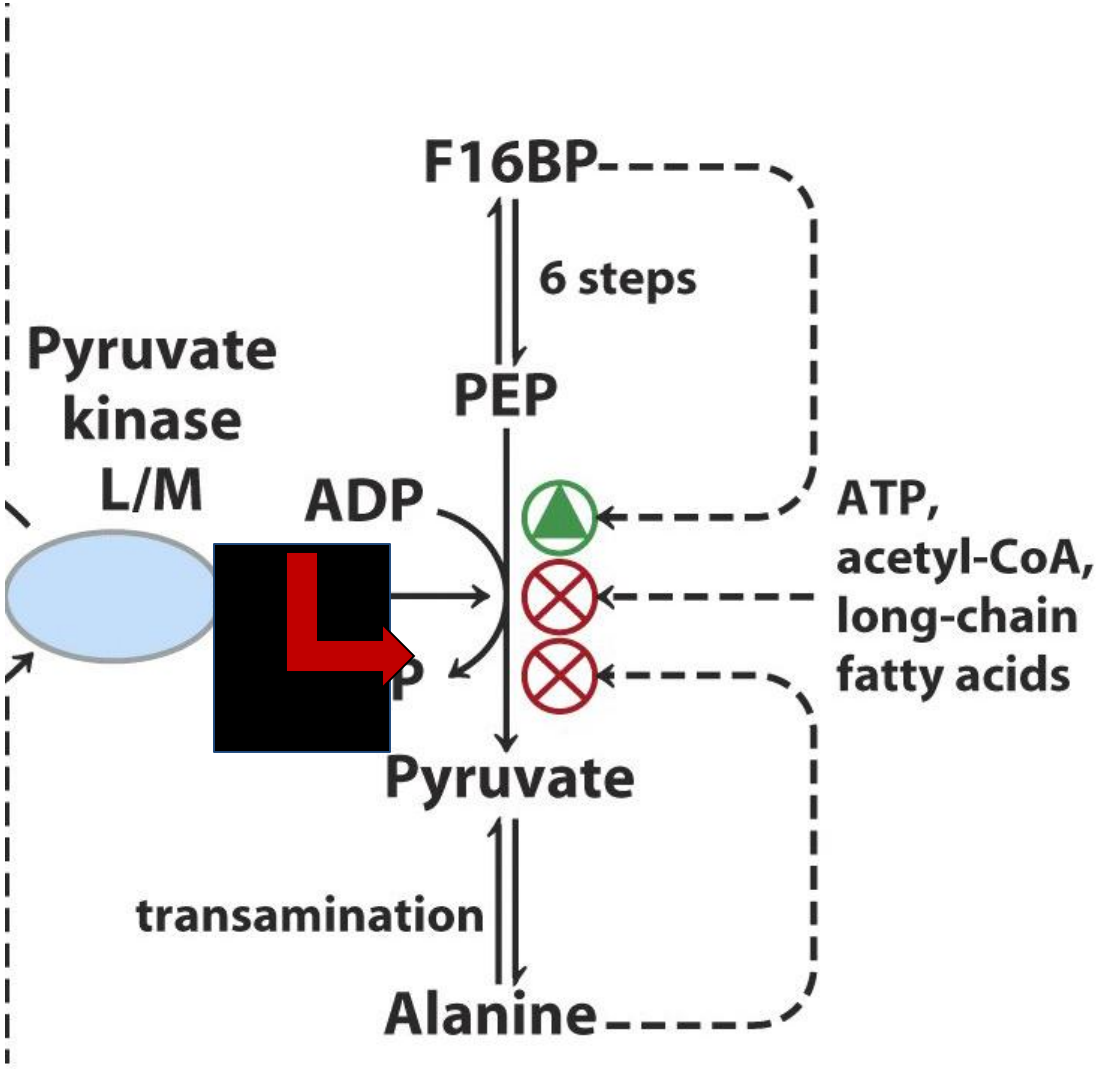


Figure 15-14b
Lehninger Principles of Biochemistry, Fifth Edition

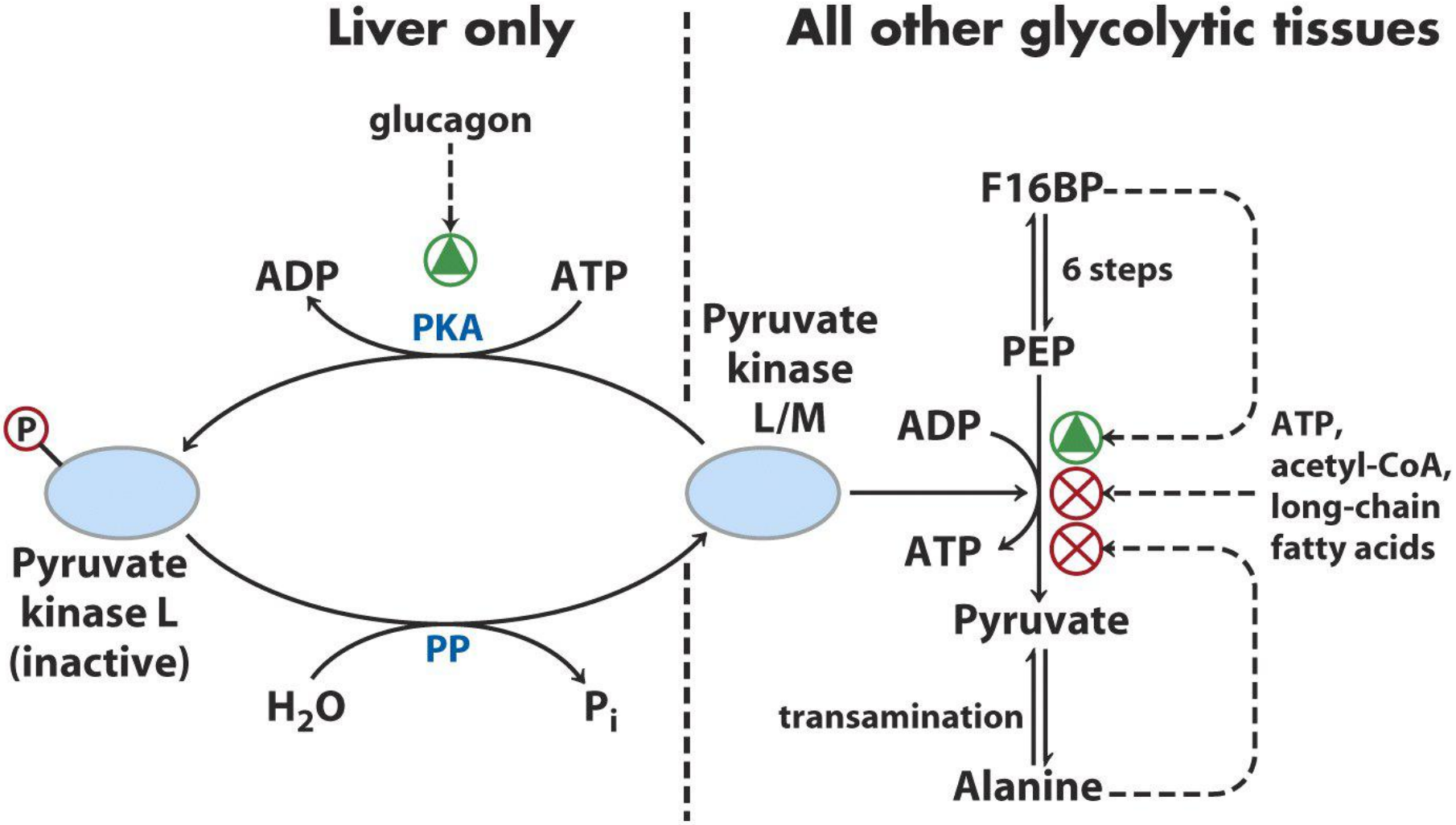

1.0 μM F–2,6–BP
0.1 μM
0
Relative velocity
100
80
60
40
20
0
1
2
3
4
5
[Fructose-6-phosphate] (μM)


1.0 μM F-2,6-BP
0.1 μM
0
Relative velocity
0
1
2
3
4
5
[ATP] (μM)

Control of pyruvate kinase



Control of pyruvate kinase



Note: all in cytosol

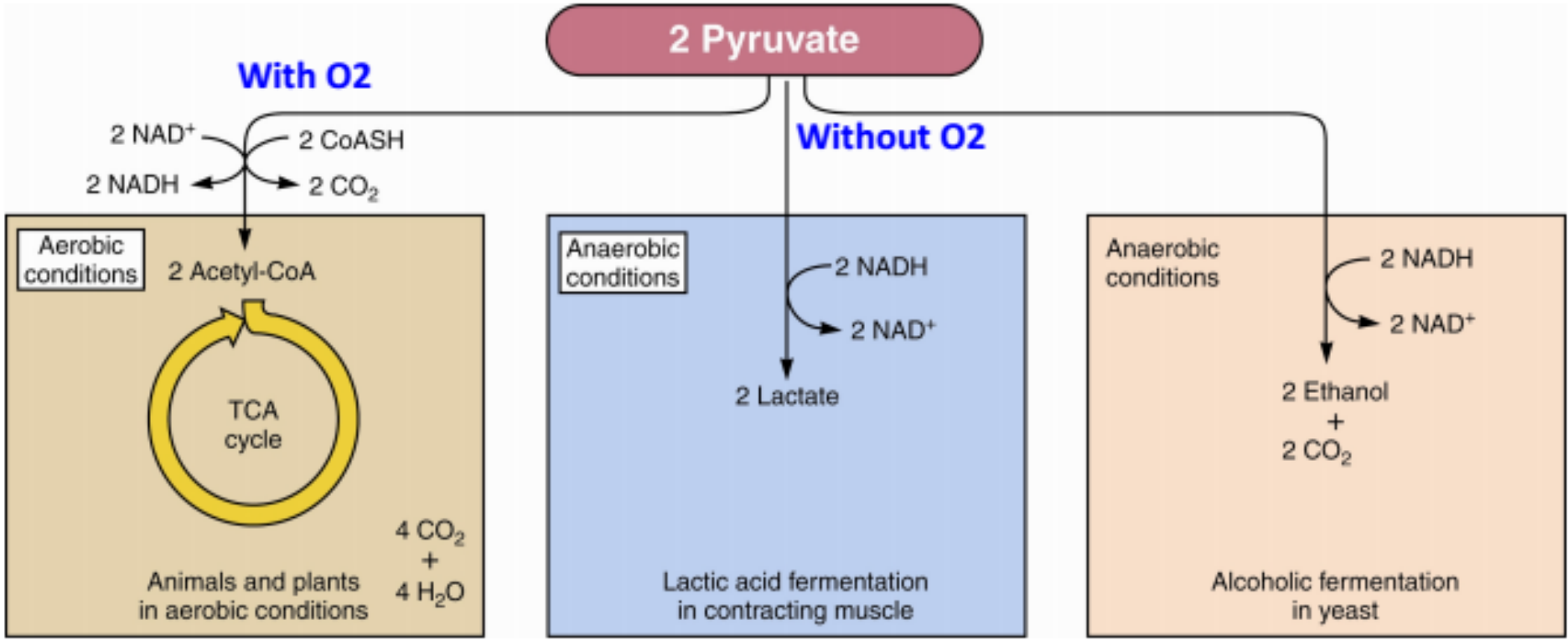
Net reaction of glycolysis

$$\text{Glucose} + 2\ \text{ADP} + 2\ \text{NAD}^+ + 2\ \text{P}_i \longrightarrow$$

$$2\ \text{Pyruvate} + 2\ \text{ATP} + 2\ \text{NADH} + 2\ \text{H}^+ + 2\ \text{H}_2\text{O}$$

那么生成的丙酮酸有哪些去路？

5. The fates of NADH and pyruvate produced in glycolysis



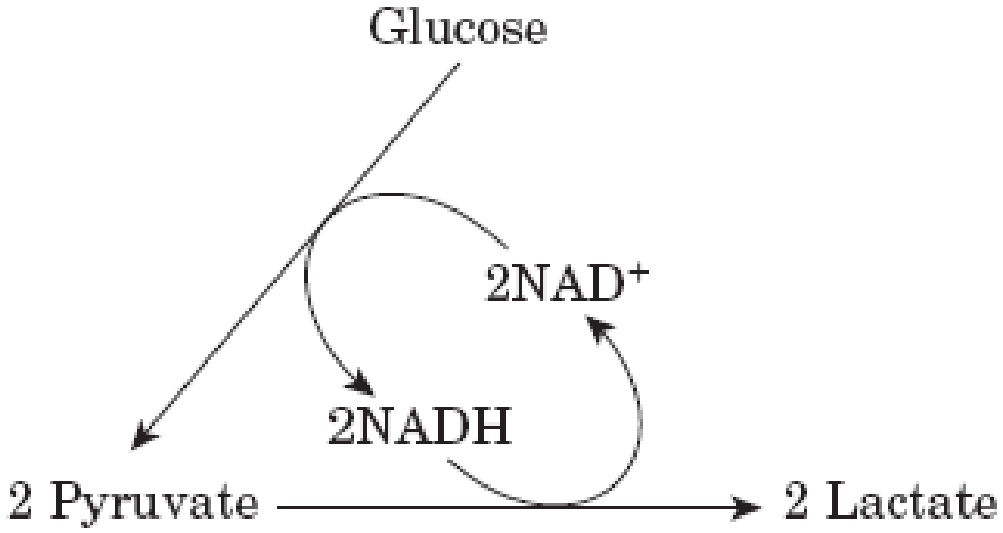
- Two acetyl-CoA(乙酰辅酶A)
- Complete degraded in the citric acid cycle
- Generate 4 CO_2 and 4 H_2O
- Large energy production

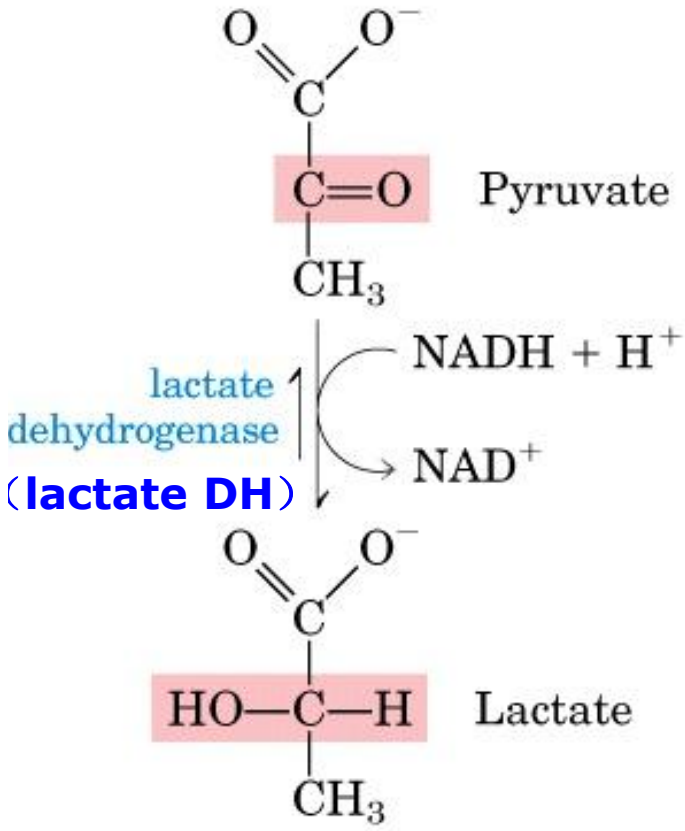
- Two lactates
- Consumption of 2 NADH
- Mechanism for the production of yogurt

- Consumption of 2 NADH
- Generate 2 ethanol and 2 CO_2
- Mechanism for the production of ethanol beverages (wine, beer, *etc*)

1）Under **anaerobic** conditions

In Exercising muscle， pyruvate →lactate

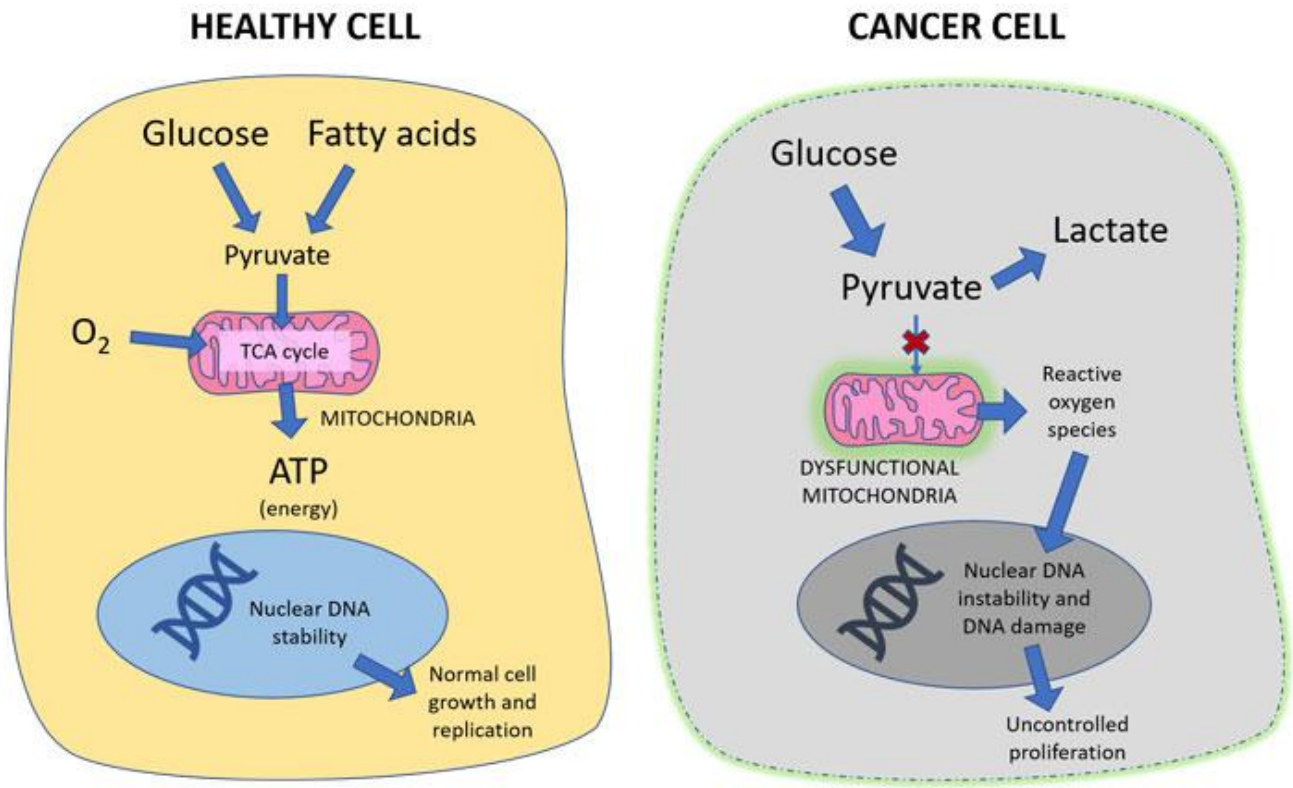




$\Delta G'^{\circ} = -25.1$ kJ/mol

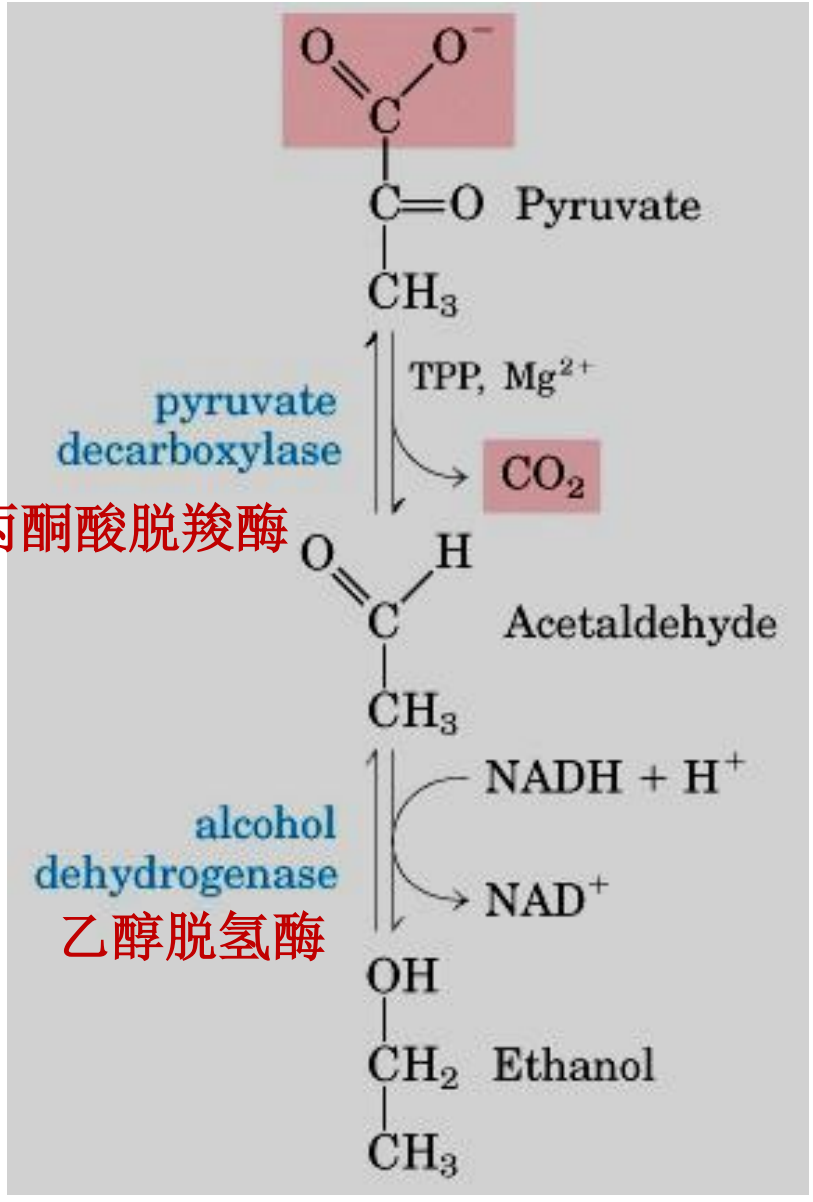
Glucose + 2 P_i +2 ADP → 2 lactate + 2 ATP + 2 H_2O

Cancer cells tend to convert G→lactate even in the presence of O_2

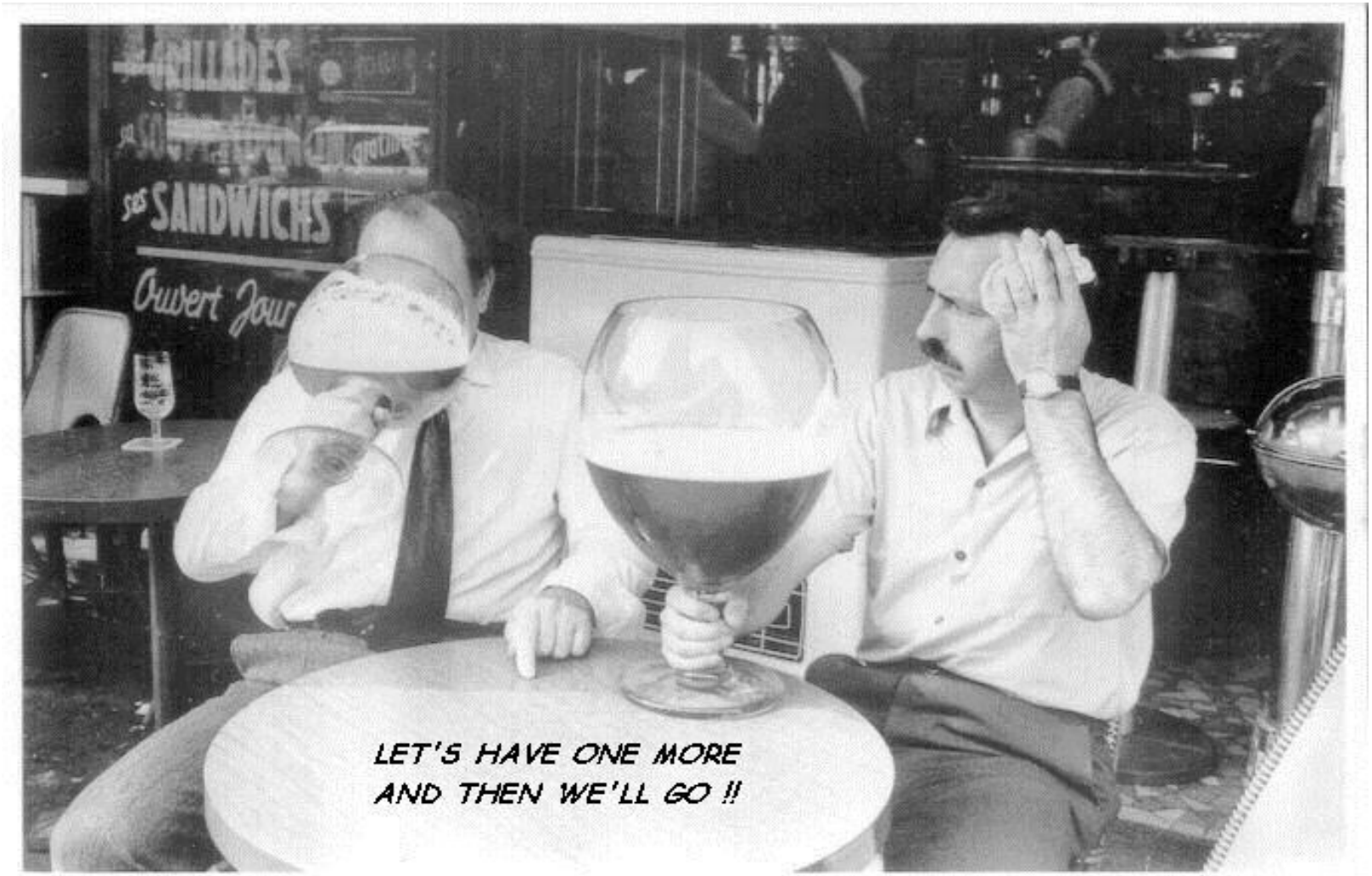


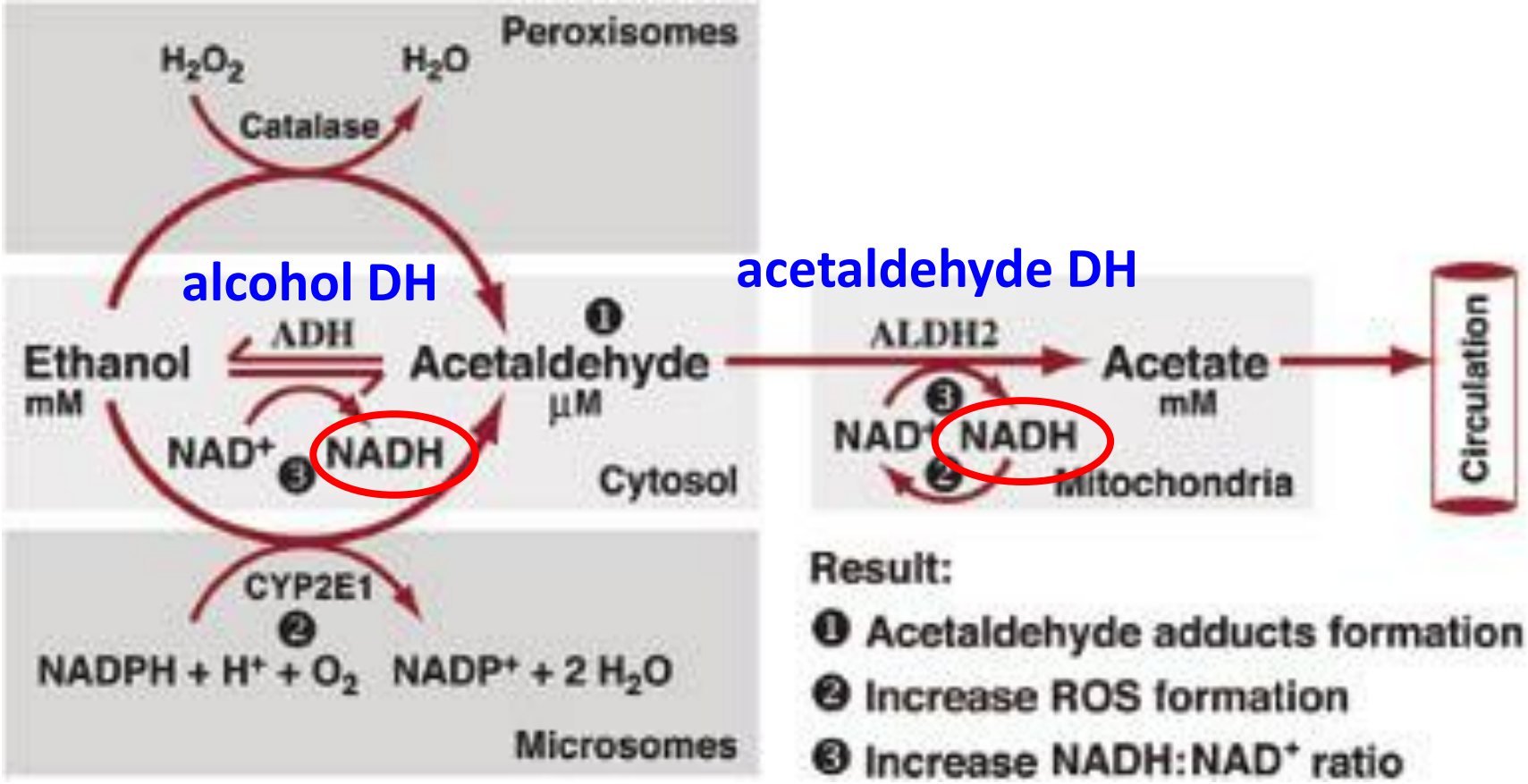
tto Warburg
931 Nobel Prize of
hysiology or Medicine

2）In anaerobic **yeast**, pyruvate→**ethanol**



What happens when we ingest what the yeast excretes.





- **high NADH----- inhibits TCA**
- **increase the synthesis of fatty acid and glycerol 3-phosphate → accumulation triacylglycerols → fatty liver.**

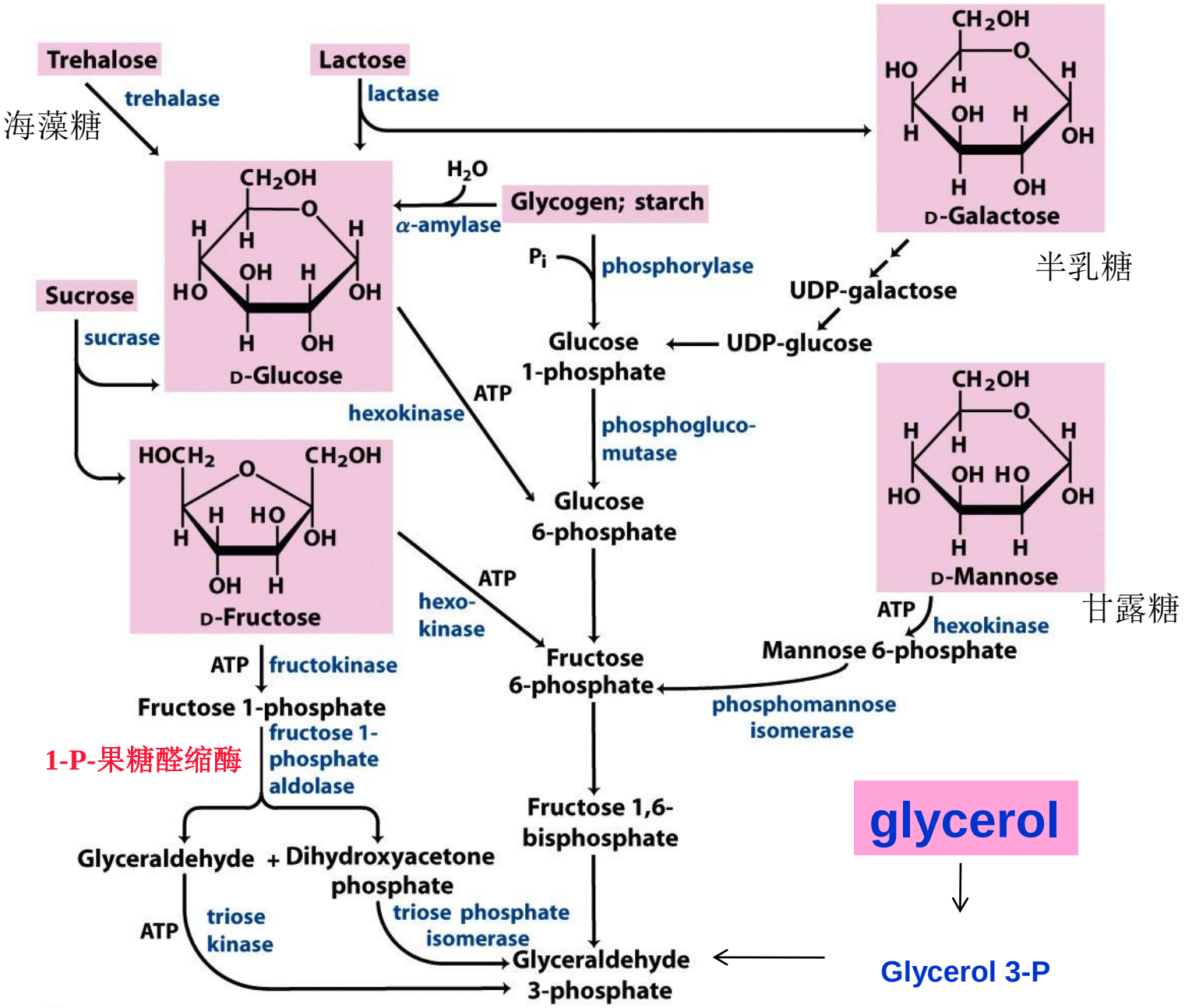
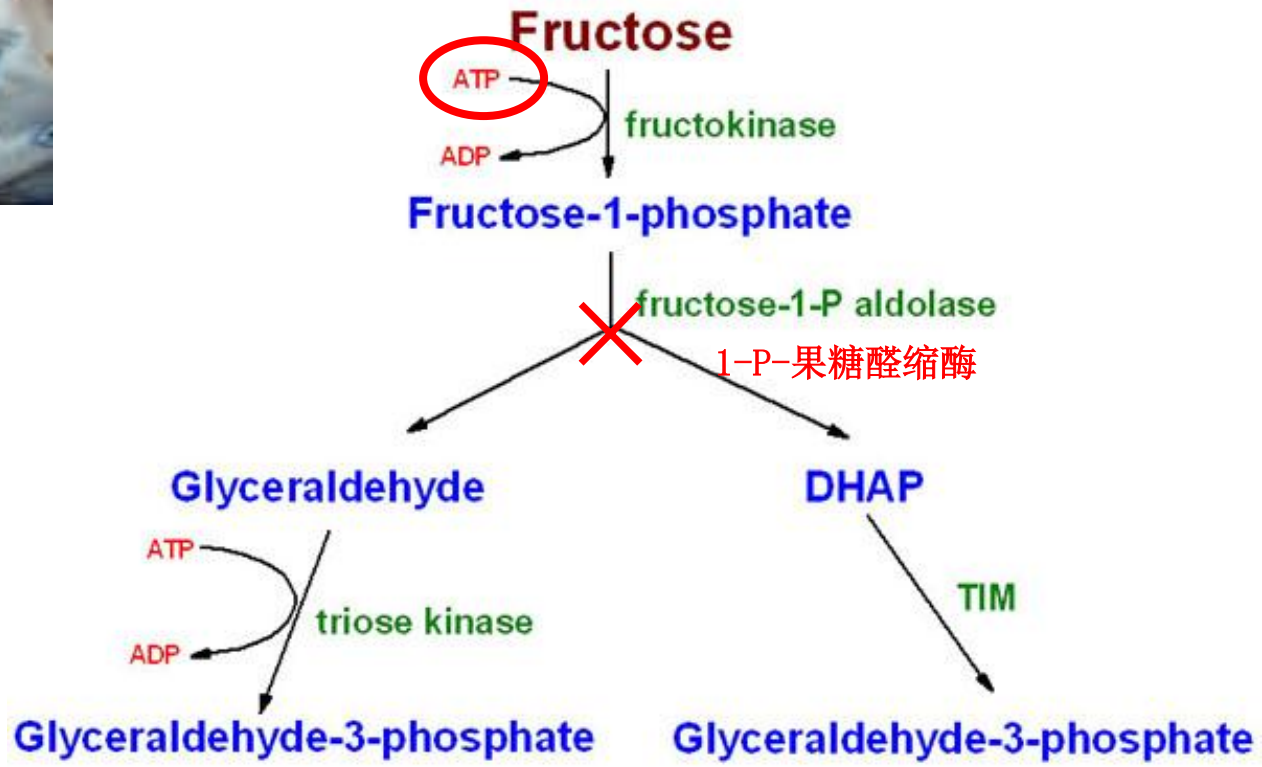


Figure 14-10
Lehninger Principles of Biochemistry, Fifth Edition

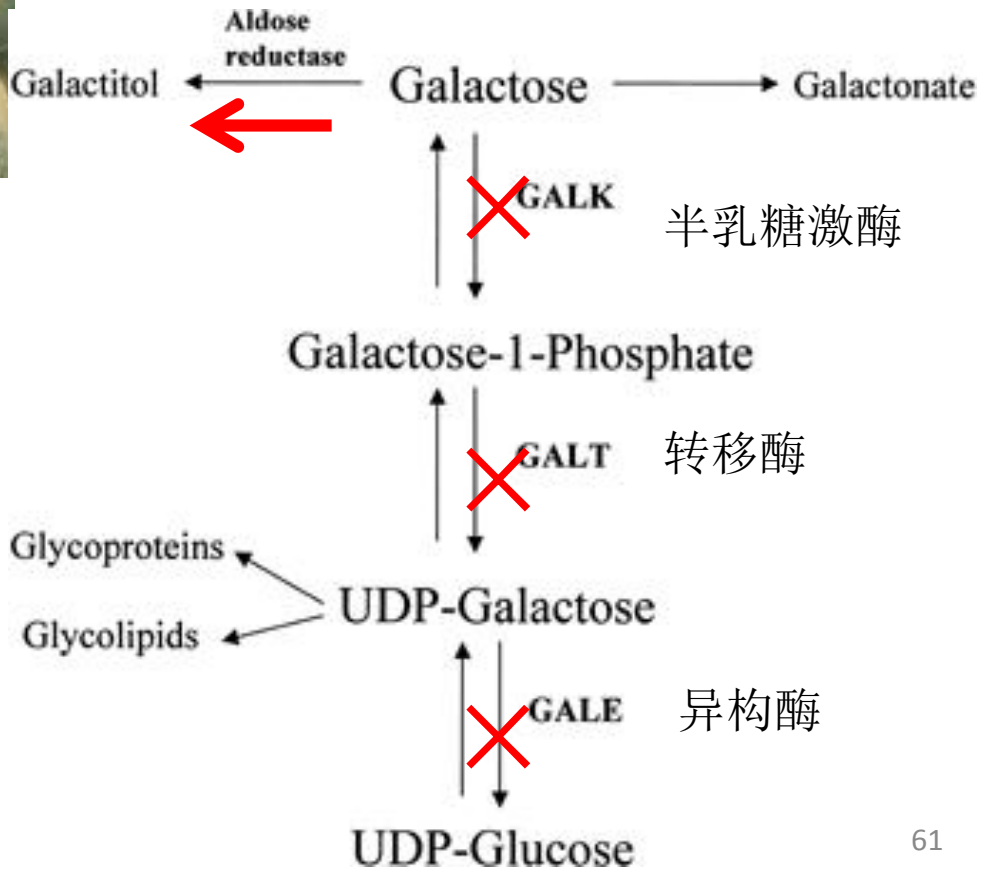
6. Other substrates used in glycolysis

Fructose intolerance（果糖不耐症）

A 9-month-old girl: bouts of sweating and vomiting (呕吐), hypoglycemia (低血糖), lactic acidosis（乳酸血症) after consumption of milk formula or fruit.



Galactosemia（半乳糖血症）



7. Summary on glycolysis

- Nearly living cell carries out glycolysis
- Glycolysis is a paradigm of metabolic pathways for the stepwise degradation of glucose which localized in the cytosol and is an anerobic process with no requirement for O_2
- Glycolysis consists of two phases each with 5 reactions
- Fates of pyruate
- Regulation of glycolysis: hexokinase, phosphofructokinase and pyruvate kinase
- Other substrates of glycolysis: frutose, galactose and mannose

思考题

1. NADH和NADPH的生物学功能是什么？
2. 常见的代谢途径类型都有哪些？分别举例。
3. 什么是糖酵解？详述糖酵解过程。
4. 丙酮酸在体内的去向。
5. 糖酵解两次底物水平磷酸化是哪两步？
6. 糖酵解关键步骤是哪一步？机体是如何调控该限速酶的？
7. 酒精诱发脂肪肝产生机制是什么？
8. 果糖不耐症和半乳糖血症产生原因和症状。