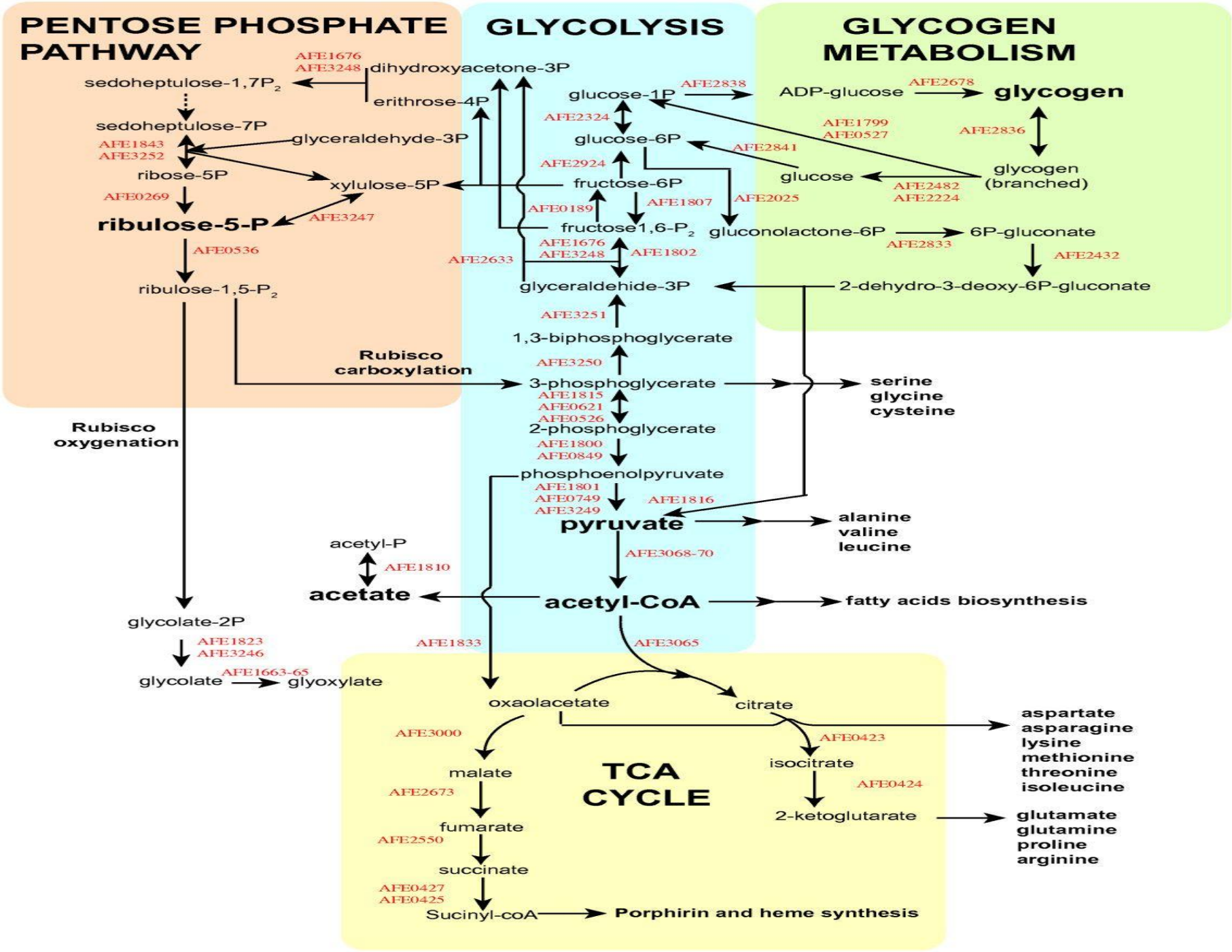
Lecture 1
Session Three
Carbohydrate Metabolism 2
Dr. Khawla A. Shemran


PENTOSE PHOSPHATE PATHWAY
GLYCOLYSIS
GLYCOGEN METABOLISM
AFE1676
AFE3248
dihydroxyacetone-3P
sedoheptulose-1,7P2
erithrose-4P
sedoheptulose-7P
AFE1843
AFE3252
glyceraldehyde-3P
ribose-5P
xylulose-5P
AFE0269
ribulose-5-P
AFE3247
AFE0536
ribulose-1,5-P2
Rubisco carboxylation
Rubisco oxygenation
AFE2838
glucose-1P
ADP-glucose
AFE2678
glycogen
AFE2324
AFE1799
AFE0527
AFE2836
glucose-6P
AFE2841
AFE2924
fructose-6P
glucose
glycogen (branched)
AFE2482
AFE2224
AFE0189
AFE1807
AFE2025
fructose1,6-P2
gluconolactone-6P
6P-gluconate
AFE2833
AFE1676
AFE3248
AFE1802
AFE2633
AFE2432
glyceraldehide-3P
2-dehydro-3-deoxy-6P-gluconate
AFE3251
1,3-biphosphoglycerate
AFE3250
3-phosphoglycerate
serine
glycine
cysteine
AFE1815
AFE0621
AFE0526
2-phosphoglycerate
AFE1800
AFE0849
phosphoenolpyruvate
AFE1801
AFE0749
AFE3249
AFE1816
pyruvate
alanine
valine
leucine
acetyl-P
AFE1810
AFE3068-70
acetate
acetyl-CoA
fatty acids biosynthesis
glycolate-2P
AFE1823
AFE3246
AFE1833
AFE3065
AFE1663-65
glycolate
glyoxylate
oxaolacetate
citrate
aspartate
asparagine
lysine
methionine
threonine
isoleucine
AFE3000
AFE0423
malate
TCA CYCLE
isocitrate
AFE0424
AFE2673
2-ketoglutarate
glutamate
glutamine
proline
arginine
fumarate
AFE2550
succinate
AFE0427
AFE0425
Sucinyl-coA
Porphirin and heme synthesis

Pentose Phosphate Pathway (PPP) or Hexose Monophosphate Shunt (HMS)

It is characterized by:

- carried out in the **cytosol**
- It is an **alternative** pathway for **glycolysis** that does ***not*** produce **energy**.
- Its enzymes (like glycolysis) are in the **cytosol** *but* the hydrogen acceptor is **$NADP^+$** instead of NAD^+.

functions

- **It has three main functions:**

1- Generates NADPH (reducing equivalent) for:

- biosynthetic/anabolic pathways such as fatty acid and steroid hormone synthesis.
- Protection against oxidants (see later)

2- Production of C5 ribose sugar for biosynthesis of nucleosides and nucleic acids. →→→→→ DNA & RNA synthesis

3- **Re- production of glucose** and other Hexoses from pentose sugar.

Phases (2 phases)
oxidative & Rearrangement

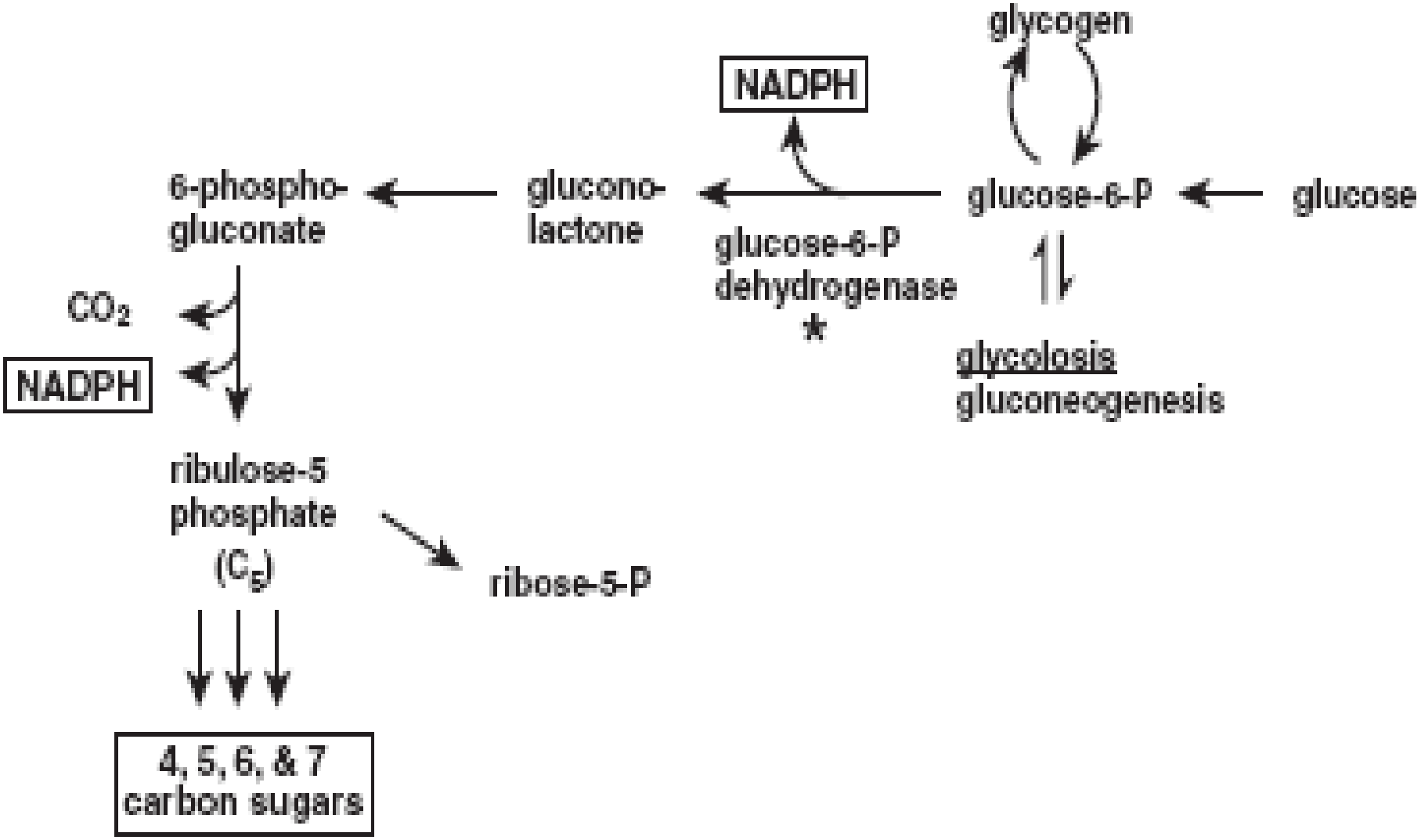
- **In phase one:**

  Glucose-6-phosphate + 2 NADP+ → → → C5-sugar phosphate + 2 NADPH + 2H+ + CO_2

- **In phase two:**

  3C5-sugar phosphate →→→→→ 2fructose-6-phosphate and glyceraldehyde 3-phosphate →→→→→ Glucose again

Two Phases


glycogen
NADPH
6-phospho-
gluconate
glucono-
lactone
glucose-6-P
glucose
glucose-6-P
dehydrogenase
*
glycolysis
gluconeogenesis
CO_2
NADPH
ribulose-5
phosphate
(C_5)
ribose-5-P
4, 5, 6, & 7
carbon sugars

Check point

What is the source of NADPH?

Which organs in human rich in this metabolic pathway?

Answers

- **The major source of NADPH is the pentose phosphate pathway.**

Lesser-known mechanisms are:

- **NADP-linked malic enzyme**
- **NADP-linked isocitrate dehydrogenase**

- **This pathway is active in:**
- **liver,**
- **adipose tissue,**
- **adrenal cortex,**
- **testes and**
- **lactating mammary glands.**

Role of NADPH in RBCs

- In RBCs, reactive oxygen species (superoxide and hydrogen peroxide) are formed normally during the process of oxygen transport as in the following:

$$Hb\text{-}Fe^{+2} \text{-} O_2 \rightarrow Hb\text{-}Fe^{+3} + O_2^{\cdot -}$$

(superoxide anion)

- This reaction is spontaneous and it takes place 1% per hour

- **Superoxide anion is very reactive and harmful to the cell due to the formation of free radicals, it inactivated by:**

$$2O_2^{\cdot -} + 2H^+ \rightarrow H_2O_2 + O_2$$

- **Hydrogen peroxide can also form free radicals, so that it must be inactivated by glutathione (GSH)**
- $2GSH + H_2O_2 \rightarrow G\text{-}S\text{-}S\text{-}G + 2H_2O$
- **Regeneration of GSH must be carried out to maintain sufficient level for normal metabolism by:**

$$G\text{-}S\text{-}S\text{-}G + 2NADPH \rightarrow 2GSH + 2NADP^+$$

- **Sufficient level of NADHP** is crucial for normal metabolism of RBC.

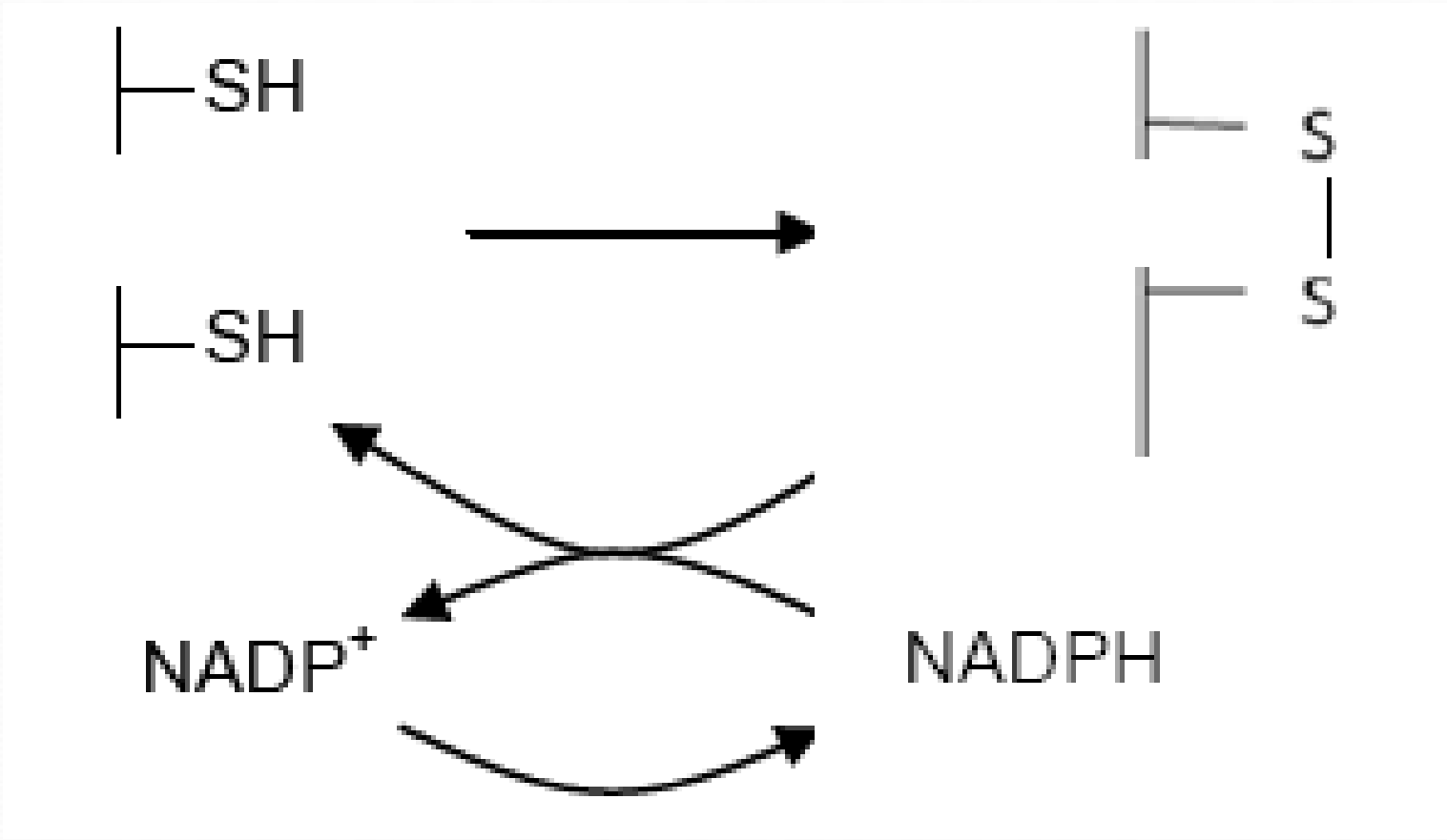
Which control G6PD synthesis

- The gene encodes G6PD is found on the long arm of the X chromosome .
- In some individuals this gene is mutated
- So it is X-linked recessive hereditary disease

What happens when G6PD gene is mutated?

- G6PD is low
- H_2O_2 accumulates in RBCs
- Leads to oxidation of hemoglobin and other proteins.
- Hemoglobin becomes cross-linked by disulphide bonds to form insoluble aggregates called **Heinz bodies** as in the following

Oxidation of hemoglobin



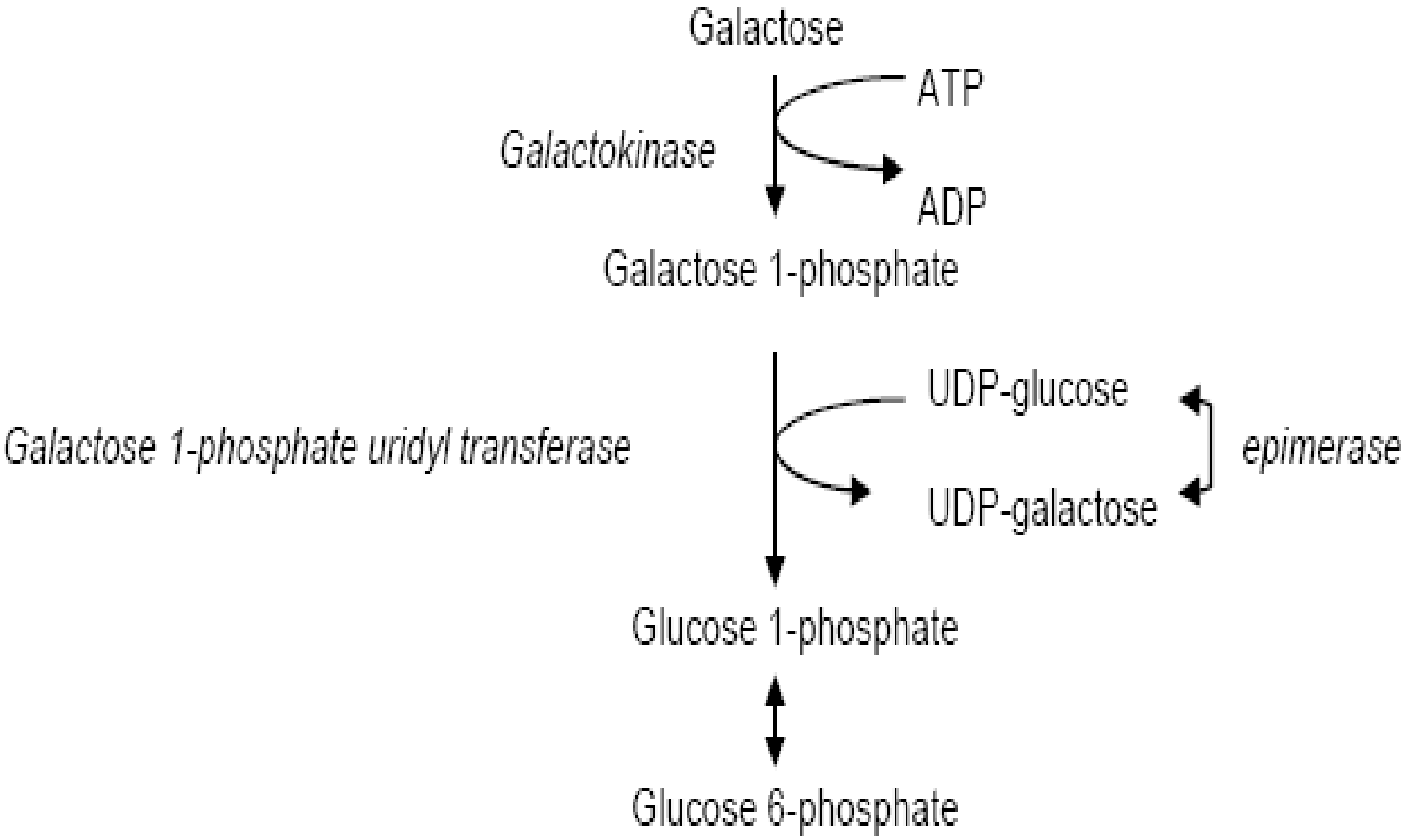
What happens when G6PD gene is mutated?

- Oxidation of Hb leads to premature destruction of RBCs (fragile) and causes haemolysis (**Acute haemolytic anemia)**
- It is **precipitated** by chemicals that increase the oxidative stress (e.g. sulphonamides, aspirin, NSAIDs and antimalarials),
- It is also **precipitated** when an individual have eaten fava beans which contain glycosides .
- For this reason it is called **favism**

Consequences of G6PD deficiency

- G6PD gene mutation
- No or Low NADPH
- GSSG not return back to GSH
- RBC membrane rigid and fragile
- Mechanical destruction of RBCs leads
- Low Hb----- anemia
- High bilirubin------- Jaundice

Galactose Metabolism

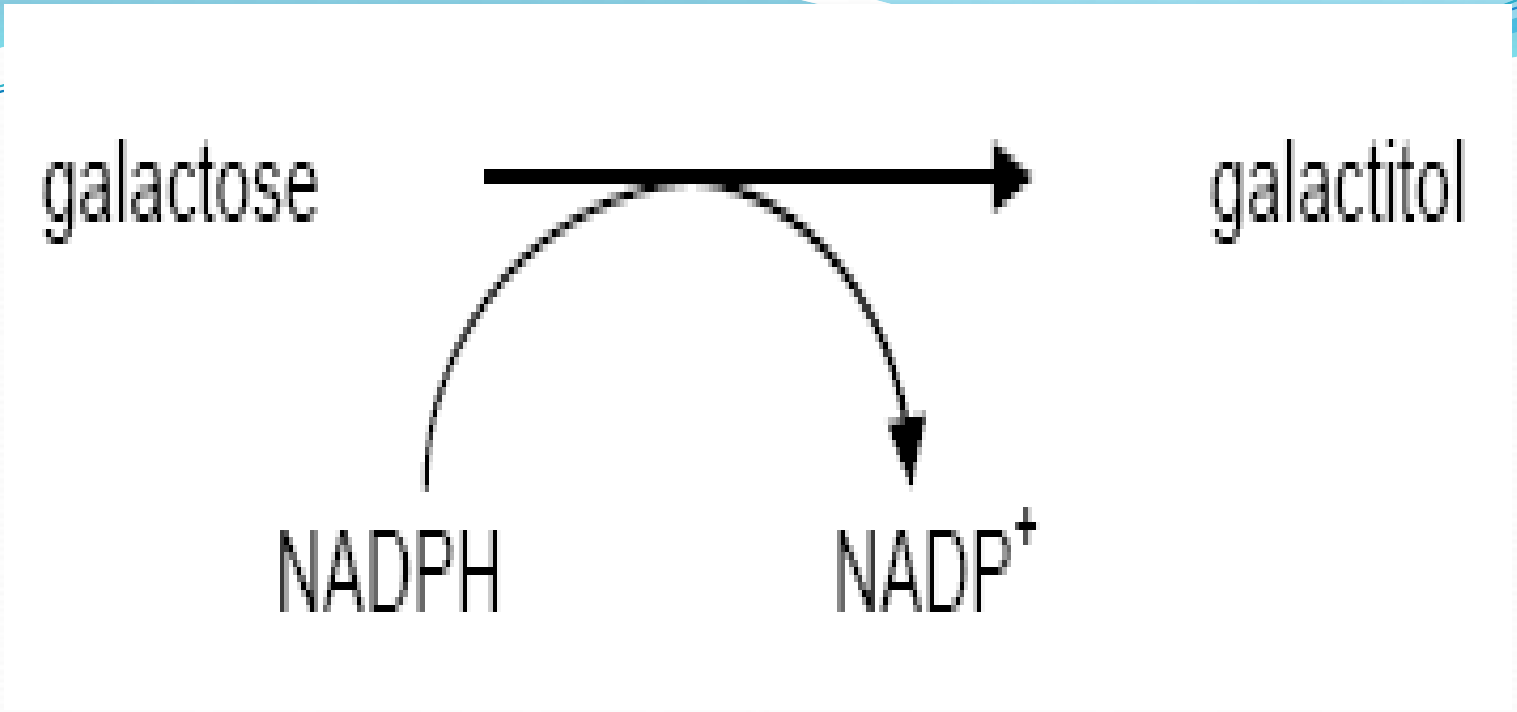


Galactosemia

- is a rare **genetic** metabolic disorder that affects an individual's ability to metabolize the diet sugar **galactose**
- **Lactose** in food (such as dairy products) is broken down by the enzyme lactase into Glucose and Galactose.
- In individuals with **galactosemia**, the enzymes needed for further metabolism of galactose (*kinase* or *transferase)* are severely diminished or missing entirely,
- Leading to accumulation of toxic levels galactose and /or Gal-1-P

Galactosemia

- The absence of the **kinase** is relatively rare and is characterized by accumulation of **galactose only** in tissues.
- The absence of the **transferase** is more common and more serious as **both** galactose and galactose 1-phosphate accumulate in tissues.
- Accumulation of G1P in tissues leads to its reduction to **galactitol** (aldehyde group reduced to alcohol group) by the activity of the enzyme *aldose reductase*:



- Galactitol is not a substrate for next enzyme the in the polyol pathway, polyol dehydrogenase ---increase osmolarity---- cells rupture (damage)

- **This reaction depletes some tissues of NADPH.---no reducing equivalent---protein cross linking (damage)**

Clinical Effects

- **Eye Damage:**

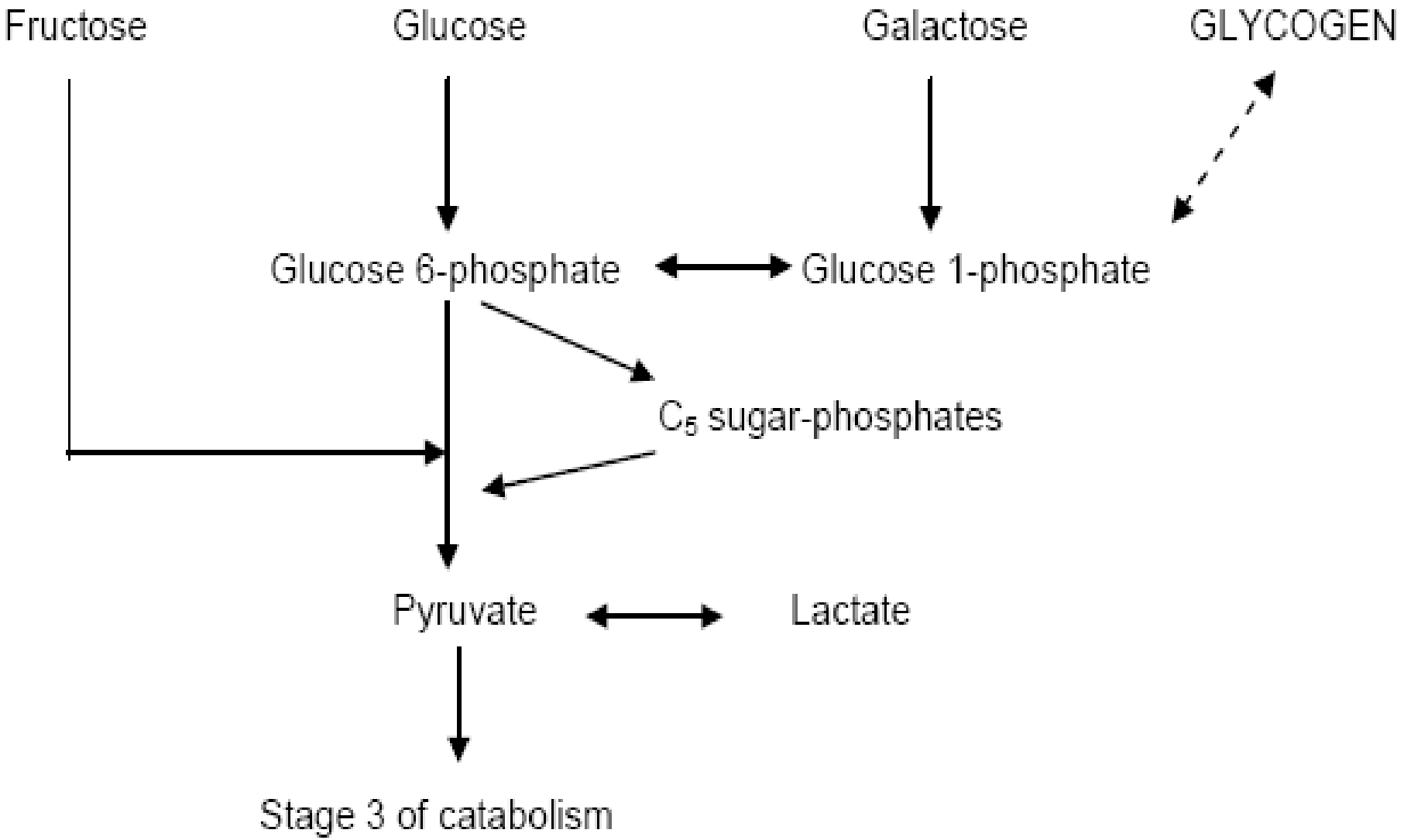
1. **Cataract**: due to-- cross linking of lens proteins by –S-S- bond formation and nonenzymatic glycosylation of the lens proteins because of the high concentration of galactose
2. **Glucoma**; Accumulation of galactose and galactitol in the eye may lead to raised intra-ocular pressure (glaucoma) which if untreated may cause **blindness**

- **Other tissues:**
- Accumulation of galactose 1-phosphate resulting in hepatomegaly (an enlarged liver), cirrhosis, renal failure, cataract, brain damage, and ovarian failure.
- This related to the **sequestration** of Pi making it unavailable for ATP synthesis.

Metabolism of Pyruvate

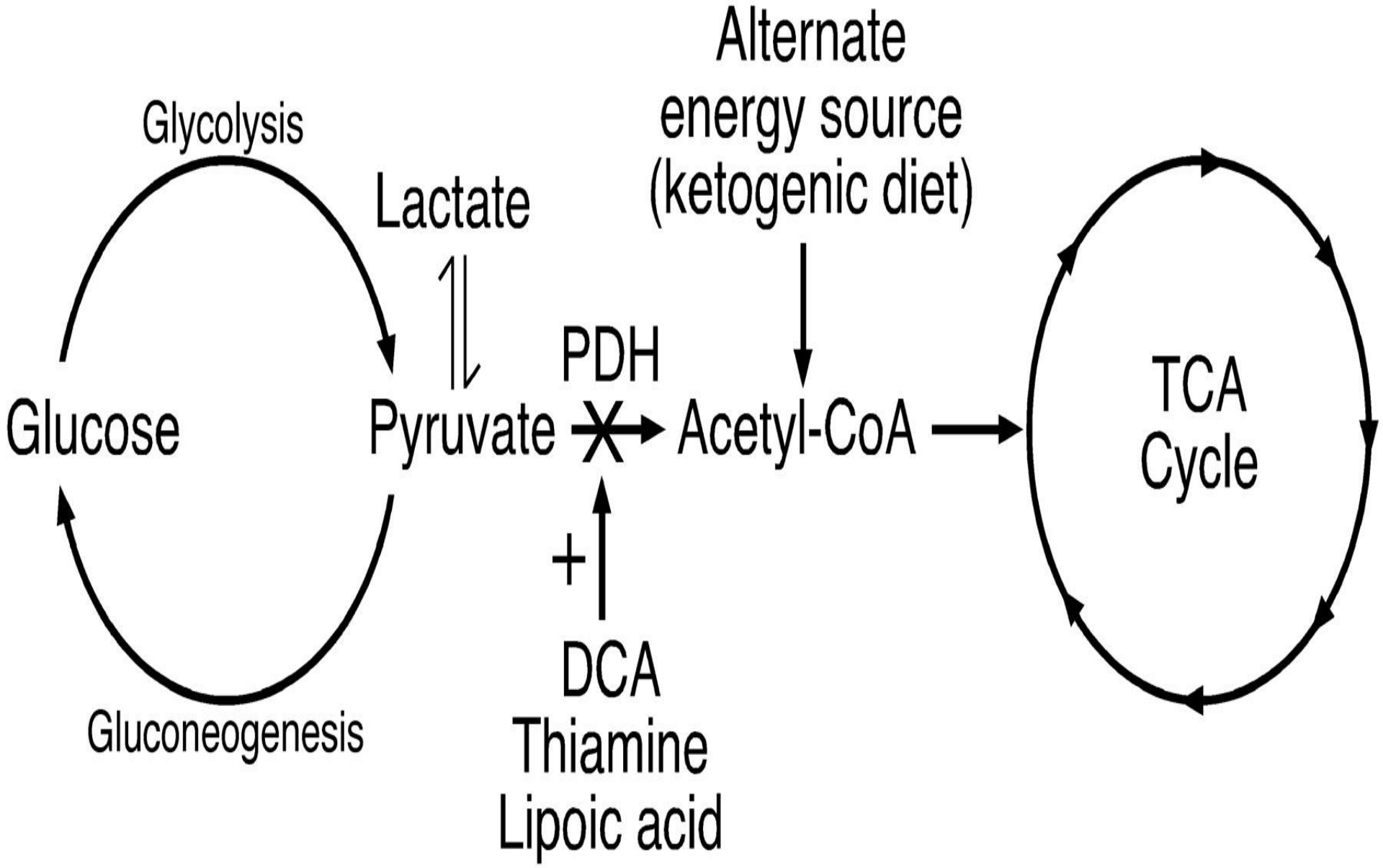
- To enter stage 3 of catabolism, Pyruvate is first converted to acetyl~CoA by the enzyme ***pyruvate dehydrogenase (PDH).***
- PDH is a multi-enzyme complex that catalyses the overall reaction:
- Pyruvate + CoA + NAD+ → Acetyl~CoA + CO_2 + NADH + H+
- This reaction is **irreversible**
- **Is PDH sensitive to vitamin B deficiency?**

Summary of stage 2 catabolism of sugars



Control mechanisms of PDH reaction

- The acetyl~CoA **inhibits PDH** so only Acetyl~CoA from the β-oxidation of fatty acids rather than from glucose is used in stage 3 of catabolism.
- The reaction is sensitive to the energy status of the cell (**ATP and NADH inhibit** and **ADP activates** the enzyme)
- The enzyme is activated when there is plenty of glucose to be catabolised (**insulin activates** the enzyme by promoting its dephosphorylation).


Alternate
energy source
(ketogenic diet)
Glycolysis
Lactate
PDH
Glucose
Pyruvate
Acetyl-CoA
TCA
Cycle
+
DCA
Thiamine
Lipoic acid
Gluconeogenesis

Lecture 2
Session 3
Tricarboxylic Acid Cycle & Gluconeogenesis

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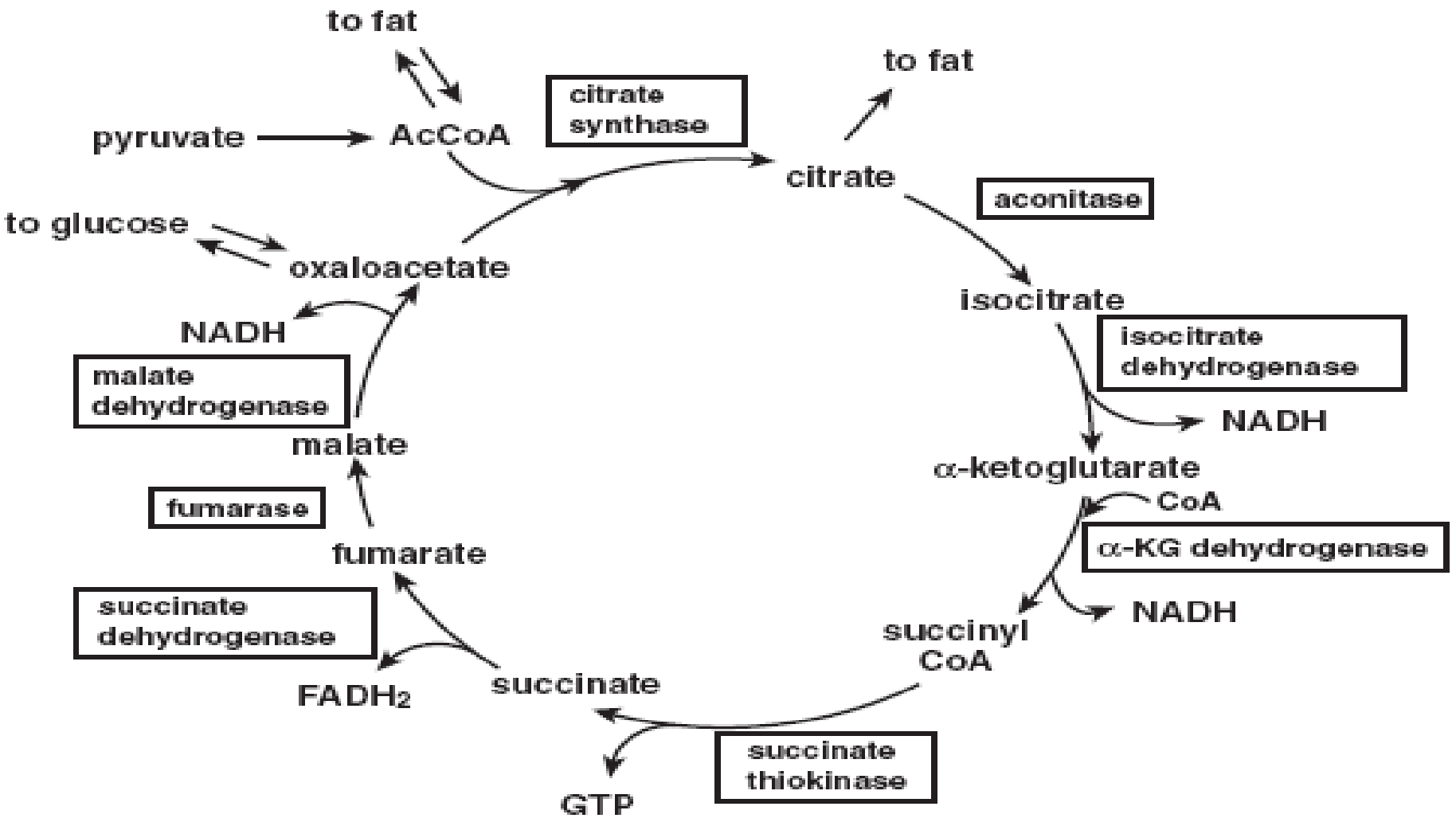
Tricarboxylic Acid Cycle (TCA)

- **This is the stage 3 of CHO metabolism. It is an** oxidative pathway that occurs in **mitochondria**
  The overall equation for the pathway is:



Functions of TCA

Catabolism: of sugars, fatty acids, ketone bodies, alcohol and amino acids leads to *formation* of acetyle CoA and CO_2. (Main function)

- The H+ and e- removed from **acetate** are transferred to NAD+ and FAD (stage 4 **energy production**) .

Anabolic (biosynthetic) of the intermediates include:

- α-ketoglutarate, succinate, fumarate and malate used for the synthesis of nonessential **amino acids**.
- Succinate and oxaloacetate used in synthesis of **heme and glucose**
- Citrate used in synthesis of **fatty acids.**

Regulation of the TCA cycle

- **Two major signals feed information on the rate of utilization of ATP to the TCA cycle:**
  - ATP/ADP ratio
  - NADH/NAD+ ratio
- **One of the early irreversible steps of the TCA cycle (catalysed by *isocitrate dehydrogenase*) is**
- inhibited by the high-energy signal NADH and
- activated by the low-energy signal ADP.

Gluconeogenesis (GNG)

- Is a metabolic pathway that results in the generation of glucose from non- carbohydrate carbon substrates

such as:

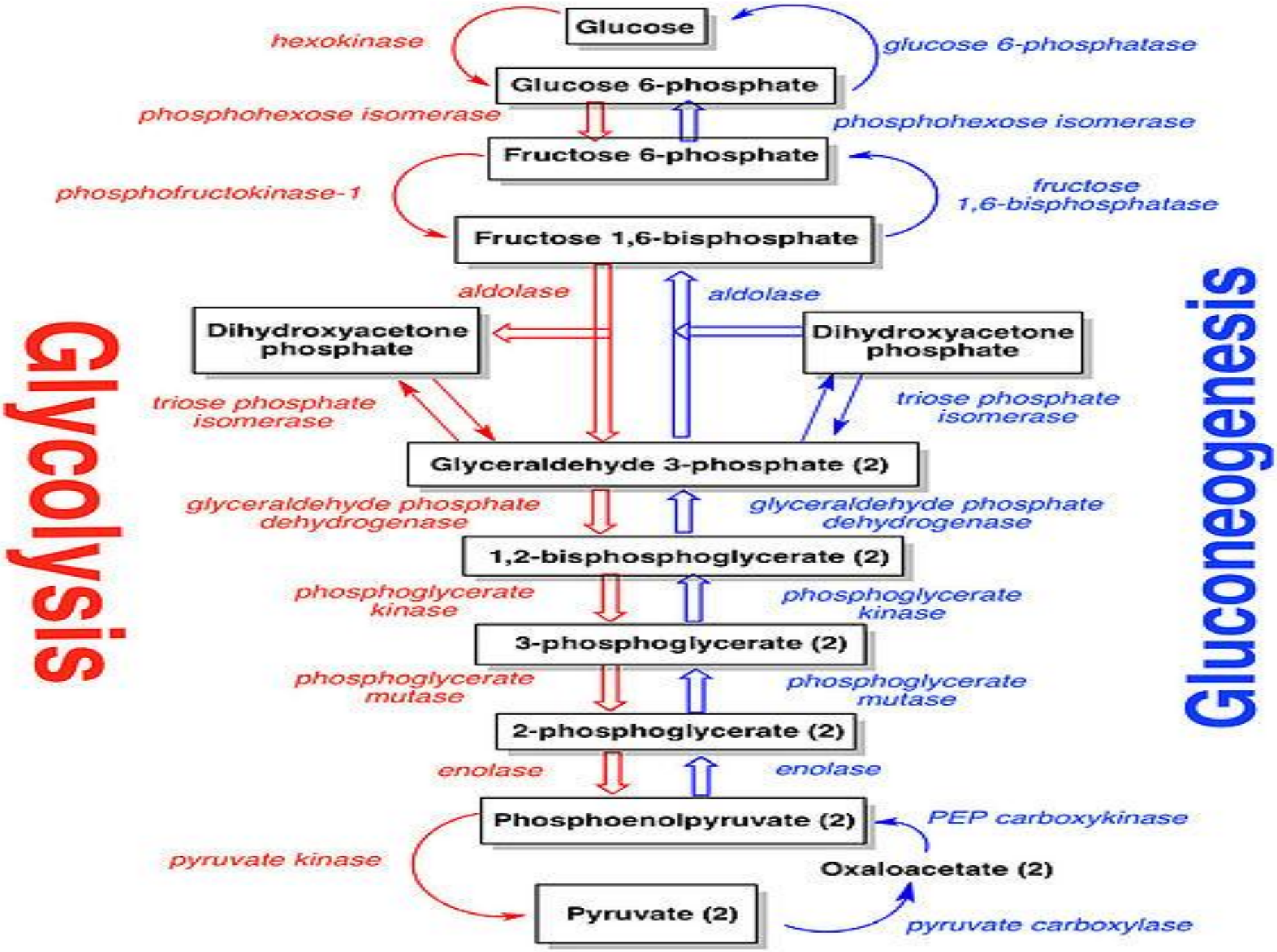
- pyruvate,
- lactate,
- Glycerol,
- glucogenic amino acids and
- Odd chain fatty acids (propionate)

Gluconeogenesis (GNG)

- It is one of the two main mechanisms humans use to keep blood glucose levels from dropping too low (hypoglycemia).
- The other means of maintaining blood glucose levels is through the degradation of glycogen (glycogenolysis).

Gluconeogenesis (GNG)

- In vertebrates, gluconeogenesis takes place mainly in the **liver** and, to a lesser extent, in the cortex of **kidneys**.
- The process occurs during periods of **fasting**, **starvation**, **low** – carbohydrate diet or severe **exercise**.
- Gluconeogenesis is also a target of therapy for type II diabetes, such as **metformin**, which inhibits glucose formation and stimulates glucose uptake by cells.


Glycolysis
Gluconeogenesis
Glucose
Glucose 6-phosphate
Fructose 6-phosphate
Fructose 1,6-bisphosphate
Dihydroxyacetone phosphate
Dihydroxyacetone phosphate
Glyceraldehyde 3-phosphate (2)
1,2-bisphosphoglycerate (2)
3-phosphoglycerate (2)
2-phosphoglycerate (2)
Phosphoenolpyruvate (2)
Pyruvate (2)
Oxaloacetate (2)
hexokinase
glucose 6-phosphatase
phosphohexose isomerase
phosphohexose isomerase
phosphofructokinase-1
fructose 1,6-bisphosphatase
aldolase
aldolase
triose phosphate isomerase
triose phosphate isomerase
glyceraldehyde phosphate dehydrogenase
glyceraldehyde phosphate dehydrogenase
phosphoglycerate kinase
phosphoglycerate kinase
phosphoglycerate mutase
phosphoglycerate mutase
enolase
enolase
PEP carboxykinase
pyruvate kinase
pyruvate carboxylase

Why Gluconeogenesis is important?

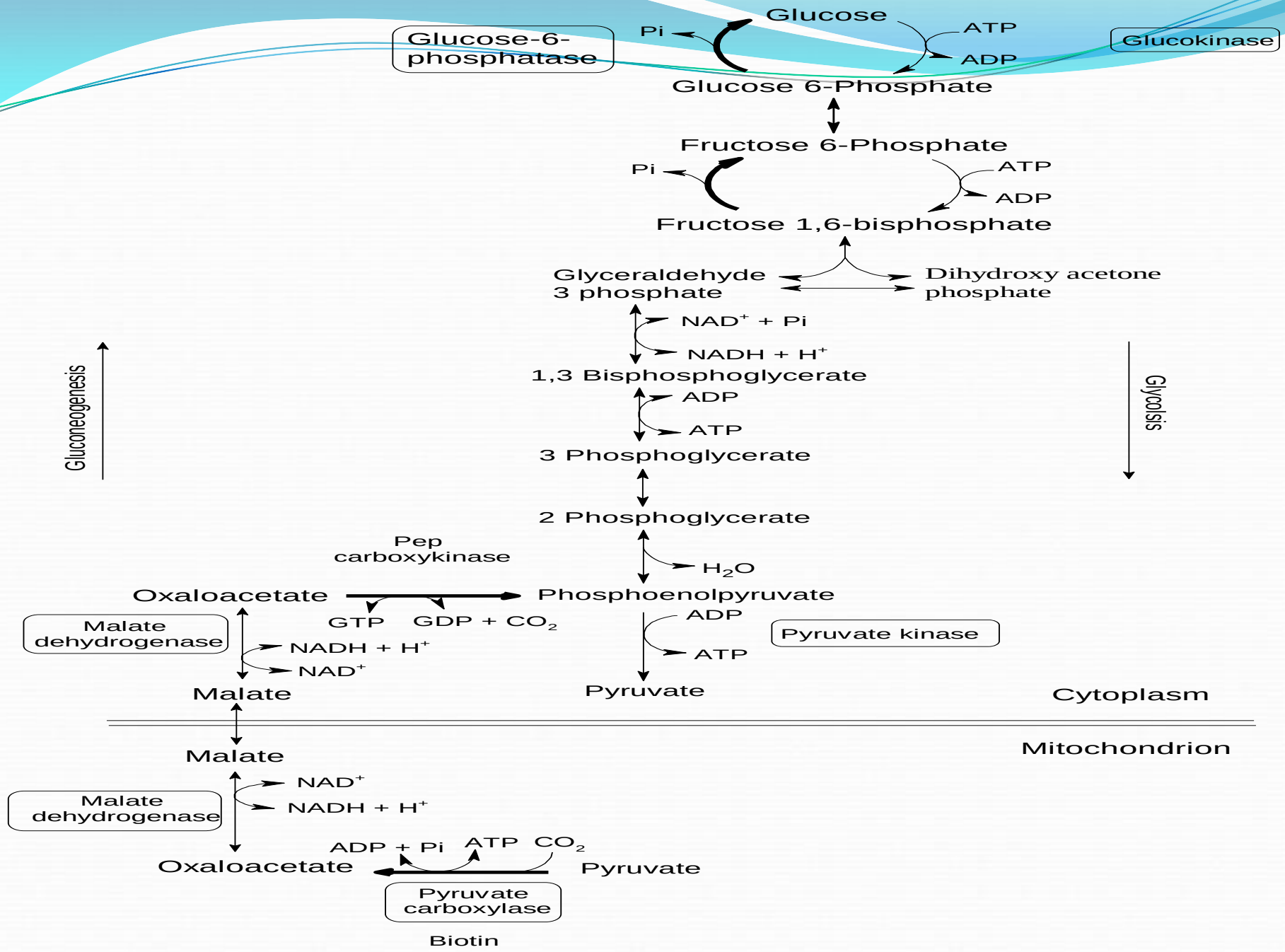
1. Glucose is necessary as a **source of energy**, especially for the brain and RBCs.
- Below the critical glucose level (＜45mg/dL) there is brain dysfunction which can lead to coma and death.

2. **Even under conditions where fat may be supplying most of the calorie requirements, there is, always basal requirements for glucose for example:**

- glucose is the **only** fuel that supplies energy to the skeletal muscles under anaerobic conditions.
- Also, glucose is the precursor of **milk sugar** (lactose) in lactating mammary glands.

3. **Gluconeogenesis is used to clear the products of metabolism of other tissues from the blood e.g.**

- **lactate** (produced by muscles and RBCs) and
- **glycerol** (produced by adipose tissue in glycolysis).


Glucose
Pi
ATP
ADP
Glucose-6-phosphatase
Glucokinase
Glucose 6-Phosphate
Fructose 6-Phosphate
Pi
ATP
ADP
Fructose 1,6-bisphosphate
Glyceraldehyde 3 phosphate
Dihydroxy acetone phosphate
NAD+ + Pi
NADH + H+
1,3 Bisphosphoglycerate
ADP
ATP
3 Phosphoglycerate
2 Phosphoglycerate
H2O
Gluconeogenesis
Glycolysis
Pep carboxykinase
Oxaloacetate
GTP
GDP + CO2
Phosphoenolpyruvate
ADP
ATP
Pyruvate kinase
Malate dehydrogenase
NADH + H+
NAD+
Malate
Pyruvate
Cytoplasm
Mitochondrion
Malate
NAD+
NADH + H+
Malate dehydrogenase
ADP + Pi
ATP
CO2
Oxaloacetate
Pyruvate
Pyruvate carboxylase
Biotin

Regulation of gluconeogenesis

- Gluconeogenesis occurs as part of the response to stress situations (e.g. fasting, starvation, prolonged exercise) and is largely under hormonal control.
- The major control sites are ***PEPCK*** and ***Fructose 1,6-bisphosphatase***. The activity of *PEPCK* is **increased** by ***glucagon and cortisol*** and **decreased** by ***insulin***
- The activity of *Fructose 1,6- bisphosphatase* is also **increased** by glucagon and **decreased** by insulin
- The ***insulin/glucagon*** ratio plays a major role in determining the rate of gluconeogenesis.
- In the absence of adequate levels of biologically effective insulin, such as occurs in **diabetes**, increased rates of gluconeogenesis contribute significantly to the hyperglycaemia.