LECTURE 5 - Pentose Phosphate Pathway

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9:41 AM

Metabolism Lecture 5 — PENTOSE PHOSPHATE PATHWAY — Restricted for students enrolled in MCB102, UC Berkeley, Spring 2008 ONLY

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Reading: Ch. 14 of Principles of Biochemistry, “Glycolysis, Gluconeogenesis, & Pentose Phosphate Pathway.”

PENTOSE PHOSPHATE PATHWAY

This pathway produces ribose from glucose, and it also generates 2 NADPH.


Glucose 6-Phosphate + 2 NADP⁺ + H₂O ⇌ Ribose 5-Phosphate + 2 NADPH + CO₂ + 2H⁺

- What are pentoses? Why do we need them?
  - DNA & RNA
  - Cofactors in enzymes
- Where do we get them? Diet and from glucose (and other sugars) via the Pentose Phosphate Pathway.
- Is the Pentose Phosphate Pathway just about making ribose sugars from glucose? (1) Important for biosynthetic pathways using NADPH, and (2) a high cytosolic reducing potential from NADPH is sometimes required to avert oxidative damage by radicals, e.g.,

\[ \cdot O₂⁻ \text{ and } H⁻O⁻ \]
Two Phases of the Pentose Pathway

Nonoxidative phase

Glucose 6-phosphate

6-Phosphogluconate

Ribulose 5-phosphate

Ribose 5-phosphate

Nucleotides, coenzymes, DNA, RNA

Oxidative phase

NADP* → 2 GSH glutathione reductase GSSG

NADPH

CO2

Fatty acids, sterols, etc. reductive biosynthesis Precursors

transketolase, transaldolase
NADPH vs. NADH
**Oxidative Phase: Glucose-6-P → Ribose-5-P**

**Glucose 6-phosphate dehydrogenase.** First enzymatic step in oxidative phase, converting NADP⁺ to NADPH.

\[
\text{Glucose 6-phosphate + NADP}^+ \rightleftharpoons 6\text{-Phosphoglucono-5-lactone + NADPH + H}^+ 
\]

**Mechanism.** Oxidation reaction of C1 position. Hydride transfer to the NADP⁺, forming a lactone, which is an intra-molecular ester.

**Energetics.** Very favorable reaction in the forward direction for producing NADPH.
**Lactonase.** A specific enzyme that targets 6-Phosphoglucono-δ-lactone for hydrolysis.

\[ 6\text{-Phosphoglucono-δ-lactone} \leftrightarrow 6\text{-Phosphogluconate} \]

**Mechanism.** The lactone is opened by hydrolysis, the addition of water to cleave a bond, usually a type of amide or ester. In this case, since the lactone (by definition) is intramolecular, then 6-phosphoglucono-δ-lactone is opened up to the acid form, gluconate.

**Energetics.** Often these types of hydrolysis reactions are very favorable.
**6-Phosphogluconate dehydrogenase.** Performs oxidative decarboxylation converting the 6-carbon skeleton to a pentose.

\[
6\text{-Phosphogluconate} + \text{NADP}^+ \leftrightarrow \text{D-Ribulose 5-phosphate} + \text{NADPH} + \text{H}^+ + \text{CO}_2
\]

**Mechanism.** The C1 carboxylate is removed as the C3 position is oxidized to a ketone, making 5-carbon ketose, ribulose.
**Phosphopentose Isomerase.**

D-Ribulose 5-phosphate $\leftrightarrow$ D-Ribose 5-phosphate

**Mechanism.** Typical ketose to aldose conversion. Another example of a general acid-base catalyzed reaction.
Non-oxidative Phase: Recycling Pentose Phosphates to Glucose 6-Phosphate

*What if the cell needs much more NADPH than it needs pentose?* D-ribose 5-phosphate has to be converted back to glucose 6-phosphate in multiple enzyme catalyzed steps.

The recycling of 5-carbon skeletons as expected for the stoichiometry below is complex:

\[
\text{6 Pentose} \leftrightarrow \text{5 Hexose}
\]
**Ribulose 5-Phosphate Epimerase.**

Ribulose 5-phosphate $\leftrightarrow$ Xylulose 5-phosphate

**Mechanism.** *Epimerization* reaction is the flipping of a stereo center in the substrate. An *epimer* of a compound occurs when only *one* stereo center is flipped.
Transketolase & Transaldolase

A myriad of steps are required to transform pentoses back to glucose, but besides the epimerase, only two other new enzymes are needed: [1] transketolase and [2] transaldolase.

**Transketolase.** This enzyme uses a cofactor, called thiamine pyrophosphate (TPP). TPP is used to stabilize a 2-carbon carbanion intermediate.

**Transaldolase.** This enzyme forms a protonated Schiff base intermediate with a ketose, stabilizing a 3-carbon carbanion intermediate, allowing an aldehyde based sugar to react with the enzyme-linked ketose. The mechanism is similar to aldolase.
[1] Transketolase + TPP

\[ \overset{5}{\text{Ribulose 5-phosphate + Xylulose 5-phosphate}} \leftrightarrow \overset{7}{\text{Sedoheptulose 7-phosphate}} \overset{10}{\leftrightarrow \text{Glyceraldehyde 3-phosphate (G3P)}} \]

[2] Transaldolase

\[ 7 + 3 \]
\[ \overset{7}{\text{Sedoheptulose 7-phosphate + Glyceraldehyde 3-phosphate (G3P)}} \leftrightarrow \overset{10}{\text{Fructose 6-phosphate + Erythrose 4-phosphate}} \]

[3] Transketolase + TPP

\[ \overset{6}{\text{Erythrose 4-phosphate + Xylulose 5-phosphate}} \leftrightarrow \overset{9}{\text{Glyceraldehyde 3-phosphate (G3P) + Fructose 6-phosphate}} \]


\[ \text{G3P} \leftrightarrow \text{DHAP + G3P} \leftrightarrow \text{Fructose 1,6-bisphosphate} \rightarrow \text{Fructose 6-phosphate} \]
Reactions Required to Convert 6 Pentoses to 5 Hexoses
Protection from Radical Damage

Mitochondrial respiration, ionizing radiation, sulfa drugs, herbicides, antimalarials, divicine

Oxidative damage to lipids, proteins, DNA

Glucose 6-phosphate → glucose 6-phosphate dehydrogenase (G6PD) → 6-Phospho-glucono-δ-lactone
Regulation of the Pentose Phosphate Pathway.

How is this pentose phosphate pathway regulated? NADPH which is generated in the oxidative branch of the pathway can feed back and inhibit the pathway. Glucose 6-phosphate can still be utilized by glycolysis.