Lecture 11 - Biosynthesis of Amino Acids

Chem 454: Regulatory Mechanisms in Biochemistry
University of Wisconsin-Eau Claire

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Introduction

- Biosynthetic pathways for amino acids, nucleotides and lipids are very old
- Biosynthetic (anabolic) pathways share common intermediates with the degradative (catabolic) pathways.
- The amino acids are the building blocks for proteins and other nitrogen-containing compounds

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Introduction

- Nitrogen Fixation
  - Reducing atmospheric N_2 to NH_3
- Amino acid biosynthesis pathways
- Regulation of amino acid biosynthesis.
- Amino acids as precursors to other biological molecules.
  - *e.g.*, Nucleotides and porphoryns

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Introduction

- Nitrogen fixation is carried out by a few select anaerobic micororganisms
- The carbon backbones for amino acids come from glycolysis, the citric acid cycle and the pentose phosphate pathway.
- The L-stereochemistry is enforced by transamination of α-keto acids

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1. Nitrogen Fixation

- Microorganisms use ATP and ferredoxin to reduce atmospheric nitrogen to ammonia.
  - 60% of nitrogen fixation is done by these microorganisms
  - 15% of nitrogen fixation is done by lighting and UV radiation.
  - 25% by industrial processes
    - Fritz Habers (500°C, 300 atm)

$$N_2 + 3\,H_2 \longrightarrow 2\,N_2$$

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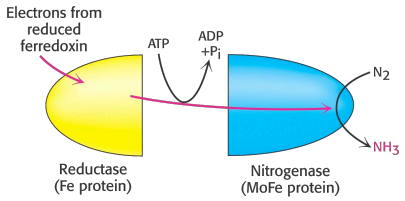
1. Nitrogen Fixation

Enzyme has both a reductase and a nitrogenase activity.



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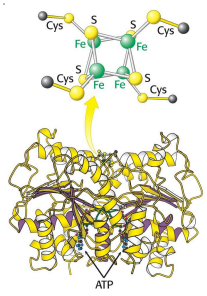
1.1 The Reductase (Fe protein)

Contains a 4Fe-4S center

- Hydrolysis of ATP causes a conformational change that aids the transfer of the electrons to the nitrogenase domain (MoFe protein)



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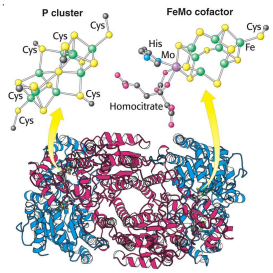
1.1 The Nitrogenase (MoFe Protein)

The nitrogenase component is an $\alpha_2\beta_2$ tetramer (240 kD)

- Electrons enter the *P-cluster*



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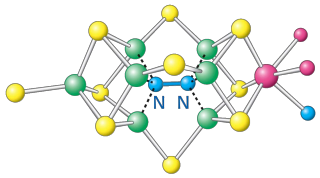
1.1 The Nitrogenase (MoFe Protein)

An Iron-Molybdenum cofactor for the nitrogenase binds and reduces the atmospheric nitrogen.



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1.2 Assimilation of Ammonium Ion

The ammonium ion is assimilated into an amino acid through glutamate and glutamine

- Most amino acids obtain their α-amino group from glutamate by transamination.
- The sidechain nitrogen of glutamine is the nitrogen source for the sidechain nitrogens of tryptophan and histidine.

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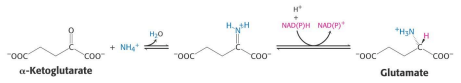
1.2 Assimilation of Ammonium Ion

Glutamate dehydrogenase

NH_4^+ + □-ketoglutarate + NADPH + H^+ ⟶ glutamate + $NADP^+$ + H_2O



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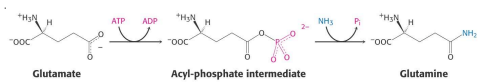
1.2 Assimilation of Ammonium Ion

Glutamine synthetase

NH_4^+ + glutamate + ATP ⟶ glutamine + ADP + P_i



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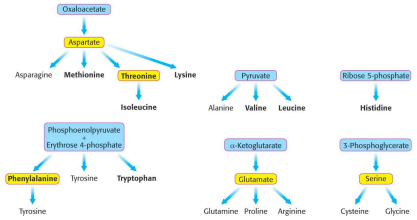
2. Amino Acid Biosynthesis

The biosynthetic pathways can be grouped into families:



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2.1 Essential Amino Acids

TABLE 24.1 Basic set of 20 amino acids

Nonessential	Essential
Alanine	Histidine
Arginine	Isoleucine
Asparagine	Leucine
Aspartate	Lysine
Cysteine	Methionine
Glutamate	Phenylalanine
Glutamine	Threonine
Glycine	Tryptophan
Proline	Valine
Serine	
Tyrosine	

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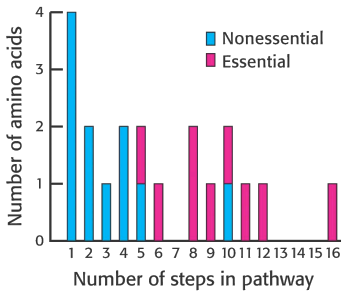
2.1 Essential Amino Acids



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2.2 Aspartate and Alanine

Transaminations:

Oxaloacetate + glutamate ⇌ aspartate + □-ketoglutarate

Pyruvate + glutamate ⇌ alanine + □-ketoglutarate

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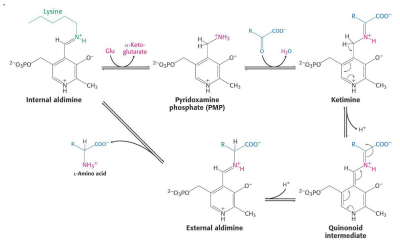
2.2 Aspartate and Alanine

Transaminations:



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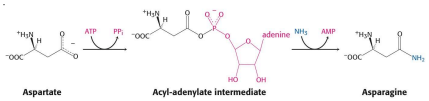
2.3 Asparagine

Amidation of aspartate



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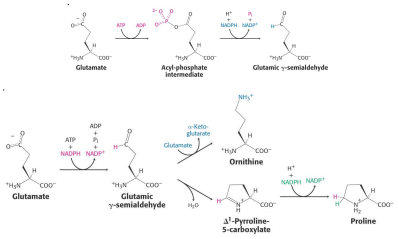
2.4 Proline and Arginine

Reduction of Glutamate



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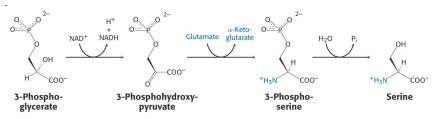
2.5 Serine and Glycine

Oxidation of 3-phosphoglycerate



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2.5 Serine and Glycine

Serine transhydroxymethylase produces glycine from serine

Serine + tetrahydrofolate ——— glycine + methylenetetrahydrofolate + H_2O

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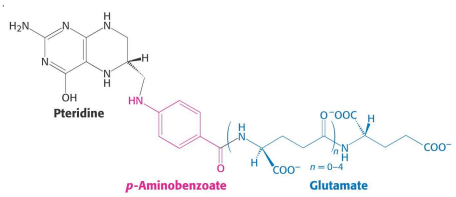
2.6 Tetrahydrofolate



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2.5 Tetrahydrofolate

TABLE 24.2 One-carbon groups carried by tetrahydrofolate

Oxidation state	Group	
Most reduced (= methanol)	$-CH_3$	Methyl
Intermediate (= formaldehyde)	$-CH_2-$	Methylene
Most oxidized (= formic acid)	–CHO –CHNH –CH=	Formyl Formimino Methenyl

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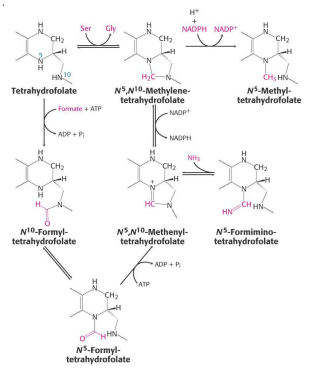
2.5 Tetrahydro-folate



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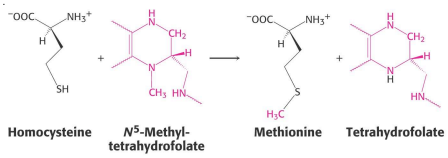
2.6 Methionine

Methylation of homocysteine



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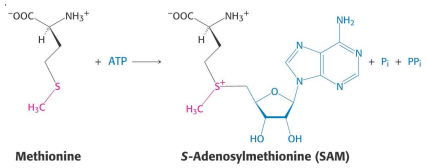
2.7 S-Adenosylmethionine (SAM)

Tetrahydrofolate does not have sufficient methyl transfer potential for many biosynthetic methylation reactions



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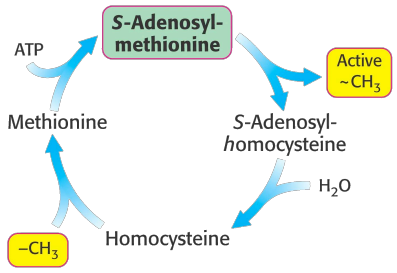
2.7 Activated Methyl Cycle



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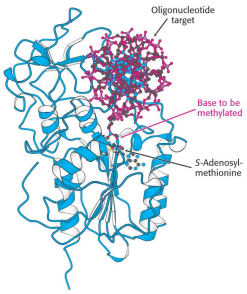
2.7 S-Adenosylmethionine

DNA methylation



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2.8 and 2.9 (skip)

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2.10 Aromatic Amino Acids

Example of essential amino acid synthesis

- Involve Shikimate and Chorismate intermediates

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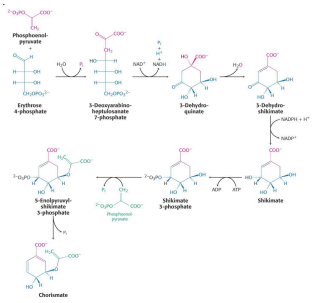
2.10 Aromatic Amino Acids

Chorismate:



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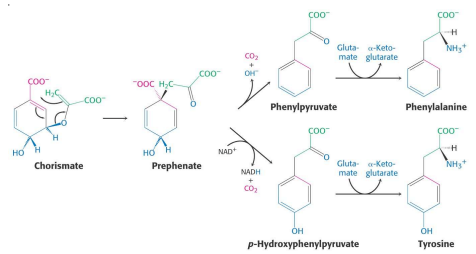
2.10 Tyrosine and Phenylalanine



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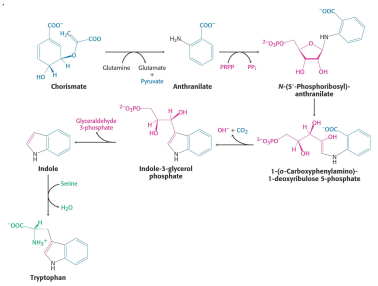
2.10 Tryptophan



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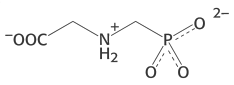
2.10 Roundup

Glycophate inhibites the enzyme that converts 5-Enolpyruvylshikimate 3-phosphate to chorismate.



Glyphosate
(Roundup)

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2.11 Substrate Channeling (skip)

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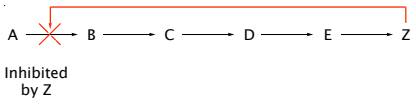
3. Regulation of Amino Acid Biosynthesis

Amino acid biosynthesis is regulated by feedback inhibition.

- The first committed step in a biosynthetic pathway is usually to the one that is regulated.



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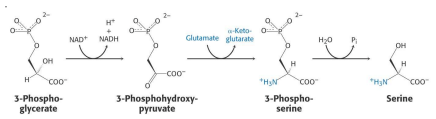
3. Regulation of Amino Acid Biosynthesis

Example: Serine biosynthesis

- 3-Phosphoglycerate dehydrogenase is inhibited by serine.



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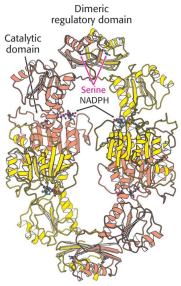
3. Regulation of Amino Acid Biosynthesis

Example: Serine biosynthesis

- 3-Phosphoglycerate dehydrogenase



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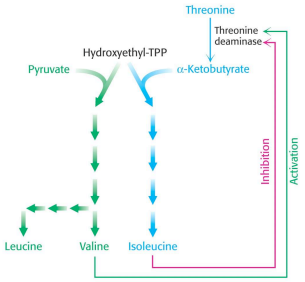
3.1 Regulation of Branched Pathways

Combination of feedback inhibition and activation



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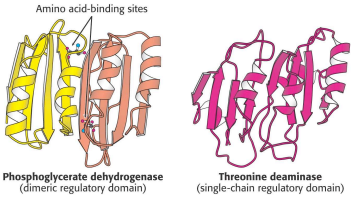
3.1 Regulation of Branched Pathways

The regulatory binding domain for threonine deaminase is similar to that found in 3-phosphoglycerate dehydrogenase.



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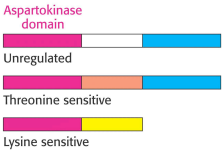
3.1 Regulation of Branched Pathways

Enzyme multiplicity

- Example: Aspartokinase
  - Threonine
  - Methionine
  - Lysine



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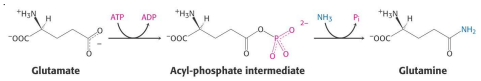
3.1 Regulation of Branched Pathways

Cumulative feedback inhibition

- Example: Glutamine Synthetase



- Glutamine is the sources for nitrogen in the synthesis of
  - tryptophan histidine
  - carbamoyl phsphate
  - glucosamine 6-phosphate
  - cytidine triphosphate
  - adenosine monophosphate

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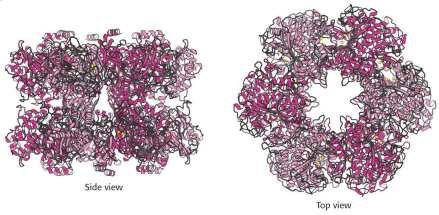
3.1 Regulation of Branched Pathways

Cumulative feedback inhibition

- Example: Glutamine Synthetase



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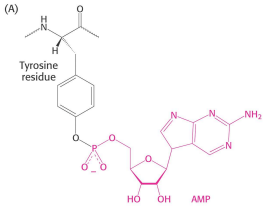
3.1 Regulation of Branched Pathways

Cumulative feedback inhibition

- Glutamine Synthetase activity is also modulated by and enzymatic cascade



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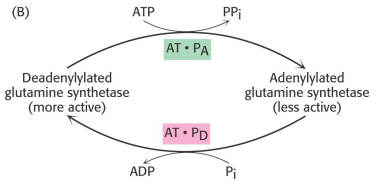
3.1 Regulation of Branched Pathways

Cumulative feedback inhibition

- Glutamine Synthetase activity is also modulated by and enzymatic cascade



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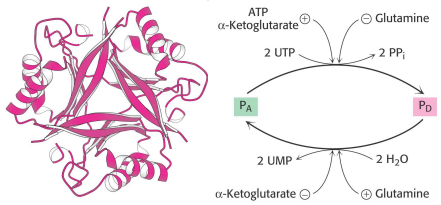
3.1 Regulation of Branched Pathways

Cumulative feedback inhibition

- The regulatory protein P (P_A or P_D)



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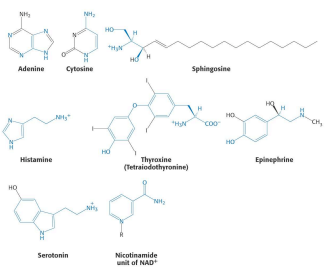
4. Amino Acid Derivatives

Amino acids are precursors for many biomolecules



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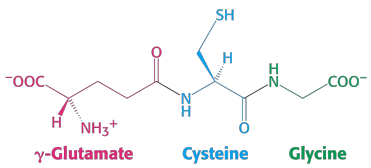
4.1 Glutathione

Glutathione

- Sulfhydryl buffer and antioxiidant



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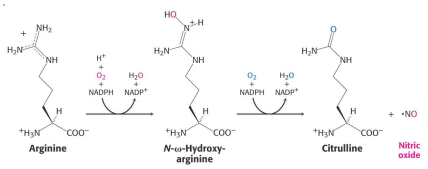
4.2 Nitric Oxide

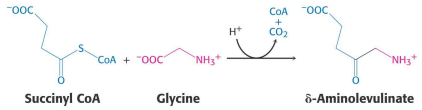
Nitric oxide is a short-lived signal molecule.

- Formed from arginine



4.3 Porphyrins

Porphyrins are synthesized from glycine an succinyl coenzyme A



4.3 Porphyrins