Biochemistry of Proteins and Nucleic Acids

2

Tissue Amino Acid Metabolism

Lecture #17-19

Lecturer: Alexander Koval

Introduction to Amino Acid Metabolism

- Amino Acid Catabolism
- Non Essential Amino Acid Biosynthesis

- Plants and microorganisms can synthesize all 20 amino acids required for protein synthesis whereas vertebrates can only synthesize about half this number.

Inborn Errors of Metabolism Can Disrupt Amino Acid Degradation

- Errors in amino acid metabolism served as sources of some of the first insights into the correlation between pathology and biochemistry.
- Although there are many hereditary errors of amino acid metabolism, **phenylketonuria** is the best known.
  - This condition is the result of the accumulation of high levels of phenylalanine in the body fluids.
  - By unknown mechanisms, this accumulation results in *mental retardation* unless the afflicted are placed on low phenylalanine diets immediately after birth.

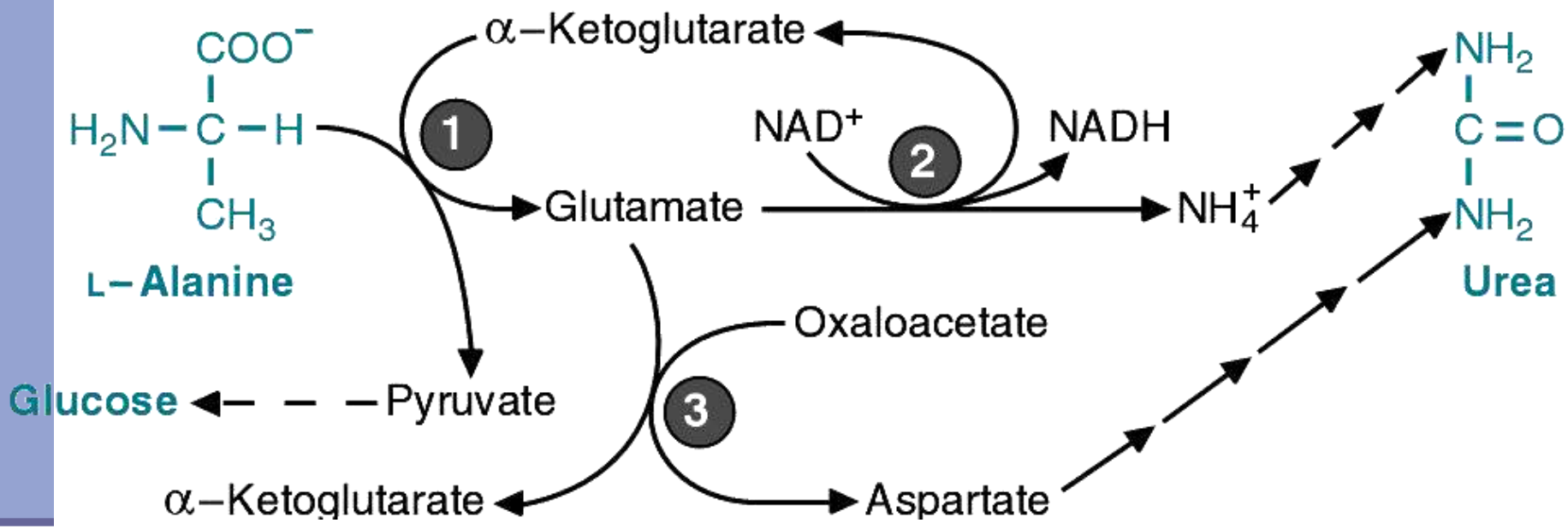
Glutamine/Glutamate and Asparagine/Aspartate Catabolism

- **Glutaminase** is an important kidney tubule enzyme involved in converting glutamine (from liver and from other tissue) to glutamate and NH^{3+}, with the NH^{3+} being excreted in the urine.
  - Glutaminase activity is present in many other tissues as well, although its activity is not nearly as prominent as in the kidney. The glutamate produced from glutamine is converted to α-ketoglutarate, making glutamine a glucogenic amino acid.
- **Asparaginase** is also widely distributed within the body, where it converts asparagine into ammonia and aspartate. Aspartate transaminates to oxaloacetate, which follows the gluconeogenic pathway to glucose.
- Glutamate and aspartate are important in collecting and eliminating amino nitrogen via **glutamine synthetase** and the **urea cycle**, respectively. The catabolic path of the carbon skeletons involves simple 1-step **aminotransferase** reactions that directly produce net quantities of a TCA cycle intermediate. The **glutamate dehydrogenase** reaction operating in the direction of α-ketoglutarate production provides a second avenue leading from glutamate to gluconeogenesis.

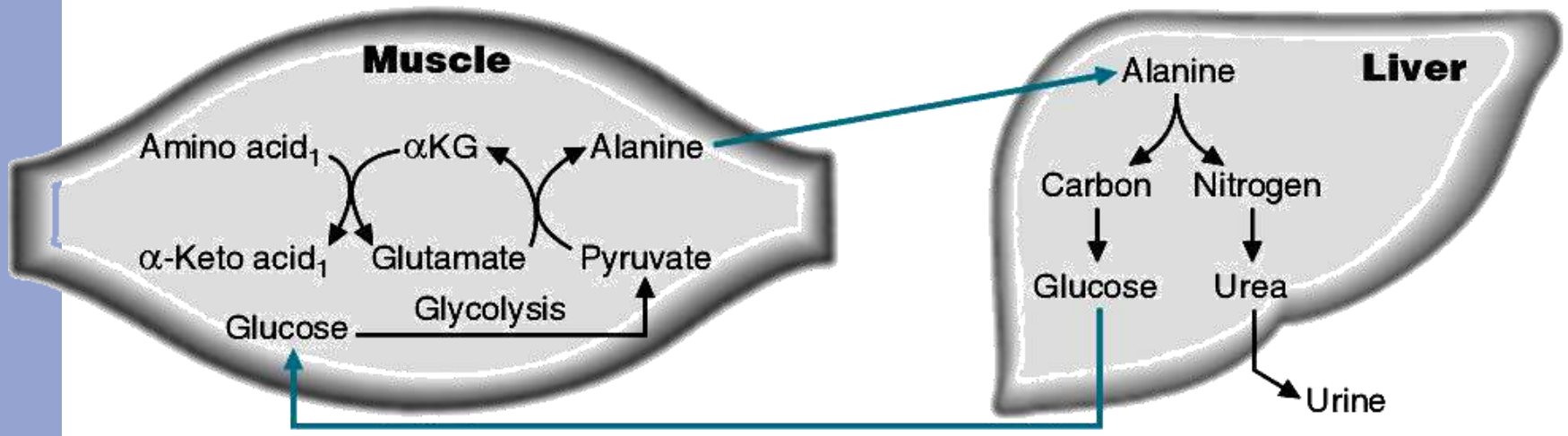
Alanine Catabolism

- **Alanine** is also important in intertissue nitrogen transport as part of the glucose-alanine cycle.
  - Alanine's catabolic pathway involves a simple **aminotransferase** reaction that directly produces pyruvate.
  - Generally pyruvate produced by this pathway will result in the formation of oxaloacetate, although when the energy charge of a cell is low the pyruvate will be oxidized to CO_2 and H_2O via the PDH complex and the TCA cycle.
  - This makes alanine a glucogenic amino acid.

Ala Conversion to Urea



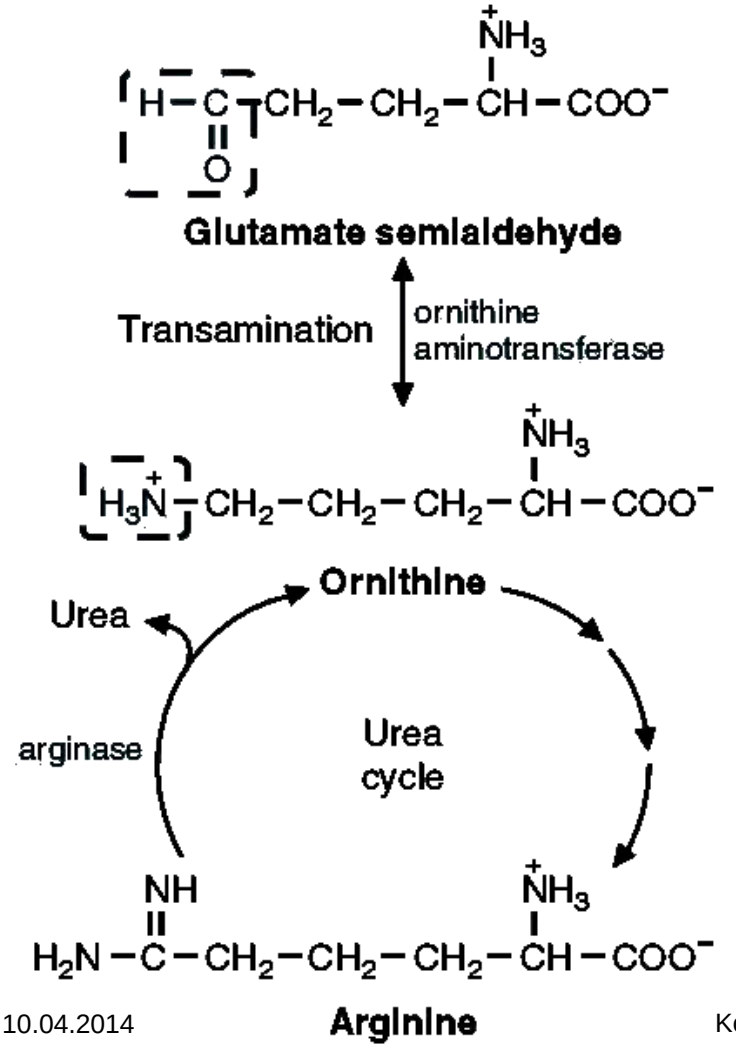
Glucose-Alanine Cycle



Arginine, Ornithine and Proline Catabolism

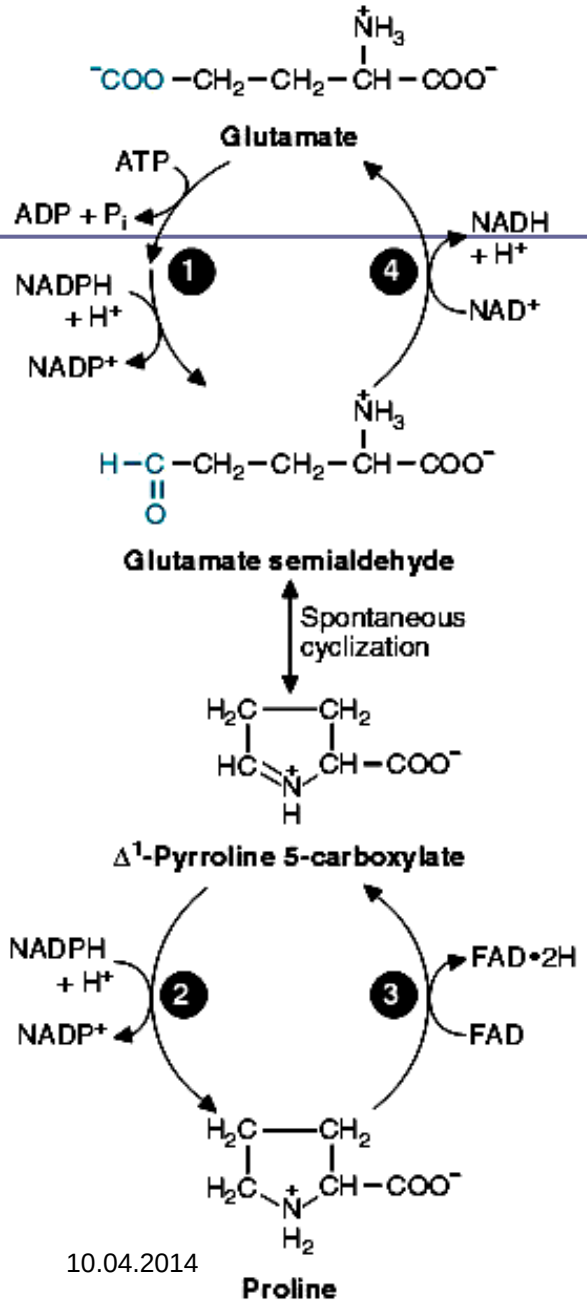
- The catabolism of **arginine** begins within the context of the **urea cycle**.
- Proline catabolism is a reversal of its synthesis process.
- Thus arginine, ornithine and proline, are glucogenic.

Arginine Metabolism



- ARG is hydrolyzed to urea and ornithine by **arginase**.
- Ornithine, in excess of urea cycle needs, is transaminated to form *glutamate semialdehyde*.
  - Glutamate semialdehyde can serve as the precursor for proline biosynthesis as described above or it can be converted to glutamate.

Prolin Metabolism

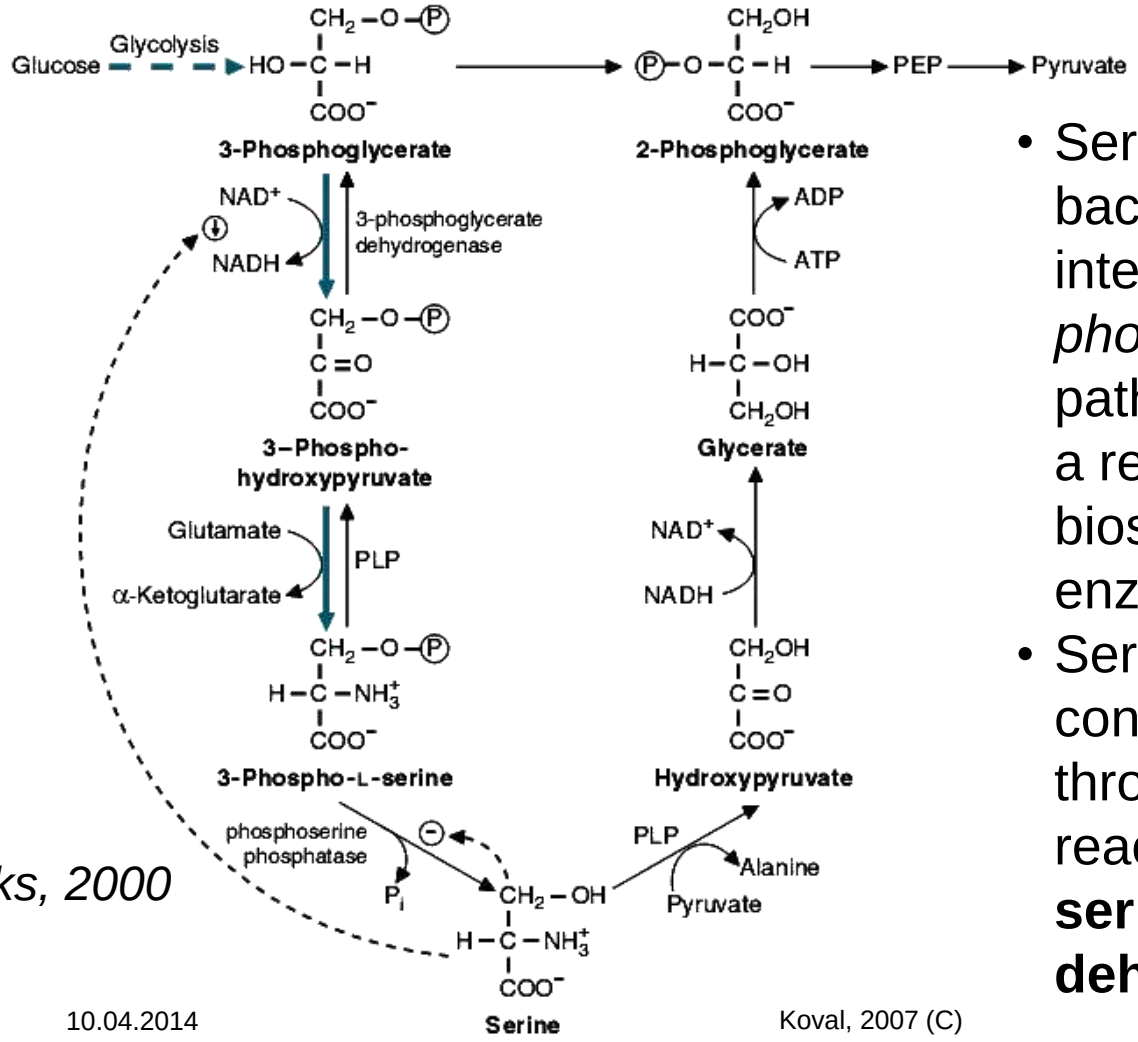


- The glutamate semialdehyde generated from ornithine and proline catabolism is oxidized to glutamate by an **ATP-independent glutamate semialdehyde dehydrogenase**. The glutamate can then be converted to α-ketoglutarate in a transamination reaction.

Serine Catabolism

- Serine can be converted to glycine and then glycine can be oxidized to CO_2 and NH_3, with the production of two equivalents of N^5,N^{10}-methylene-THF.

Serine Catabolism: 3-Phosphoglycerate and Pyruvate Formation

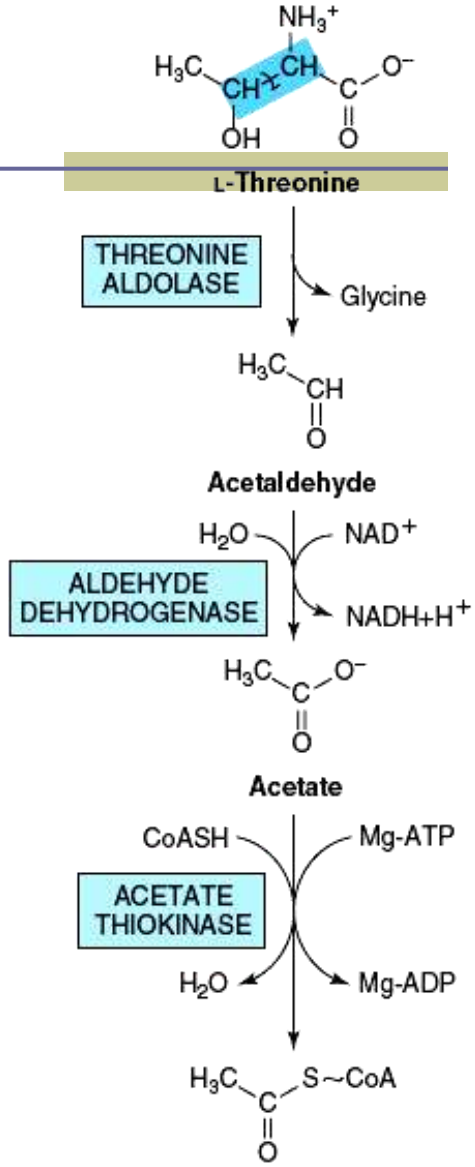


Marks, 2000

- Serine can be catabolized back to the glycolytic intermediate, *3-phosphoglycerate*, by a pathway that is essentially a reversal of serine biosynthesis. However, the enzymes are different.
- Serine can also be converted to *pyruvate* through a deamination reaction catalyzed by **serine/threonine dehydratase**.

Threonine Catabolism

- There are at least 3 pathways for threonine catabolism.
  - One involves a pathway initiated by **threonine dehydrogenase** yielding α-amino-β-ketobutyrate. The α-amino-β-ketobutyrate is either converted to acetyl-CoA and glycine or spontaneously degrades to *aminoacetone* which is converted to pyruvate.
  - The second pathway involves **serine/threonine dehydratase** yielding α-ketobutyrate which is further catabolized to *propionyl-CoA* and finally the TCA cycle intermediate, *succinyl-CoA*.
  - The third pathway utilizes **threonine aldolase**. The products of this reaction are both ketogenic (*acetyl-CoA*) and glucogenic (*pyruvate*).

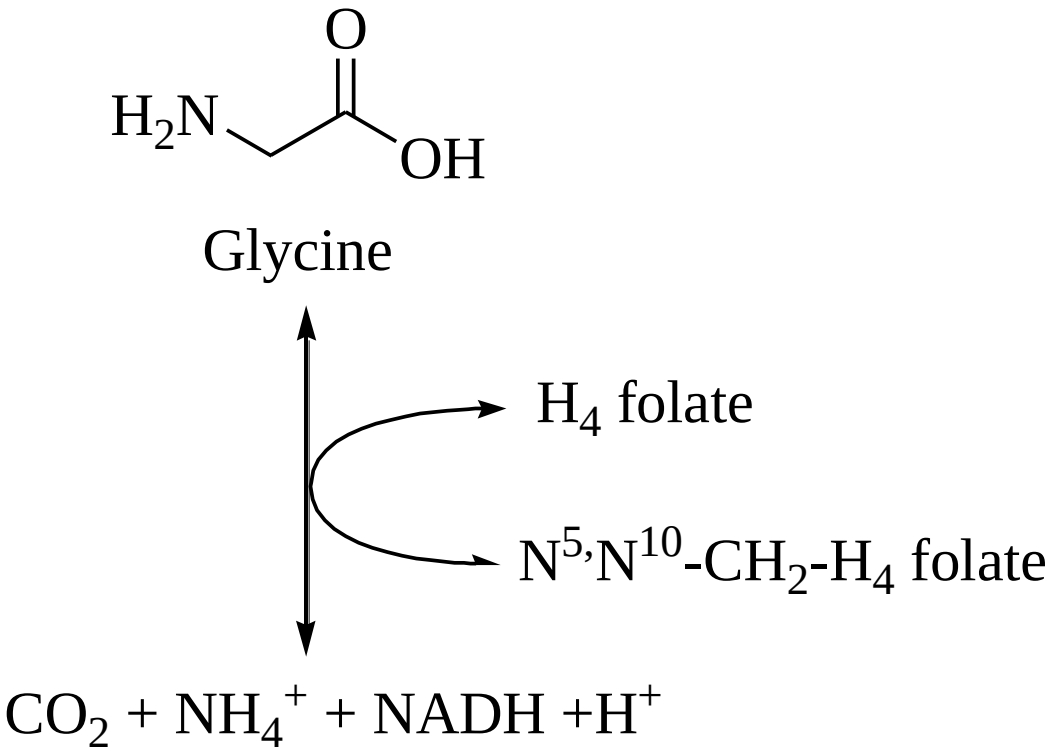


Glycine Catabolism

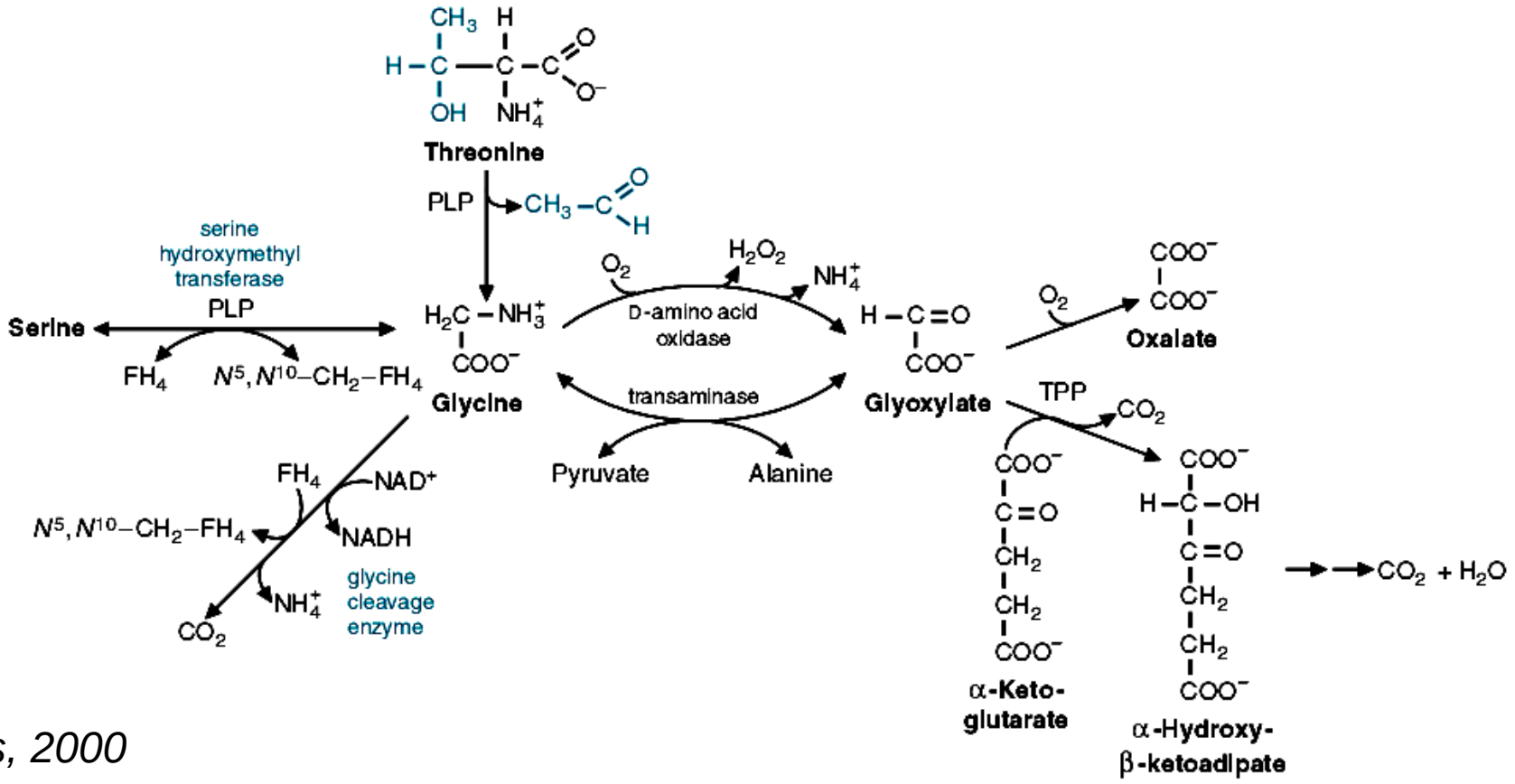
- Glycine can be converted to **serine** by **serine hydroxymethyltransferase**,
  - serine can be converted back to 3-phosphoglycerate or to pyruvate by **serine/threonine dehydratase**.
- the main glycine catabolic pathway leads to the production of CO_2, NH_4, and one equivalent of N^5,N^{10}-methylene-THF by the **mitochondrial glycine cleavage complex**.

Glycine Oxidation in Mitochondria

- Glycine produced from serine or from the diet can also be oxidized by **glycine cleavage complex, GCC**, to yield a second equivalent of N^5,N^{10}-methylene-tetrahydrofolate as well as ammonia and CO_2.
- Glycine is involved in many anabolic reactions other than protein synthesis including the **synthesis of purine nucleotides**, **heme**, **glutathione**, **creatine** and **serine**.



Glycine, Serine & Threonine Metabolism



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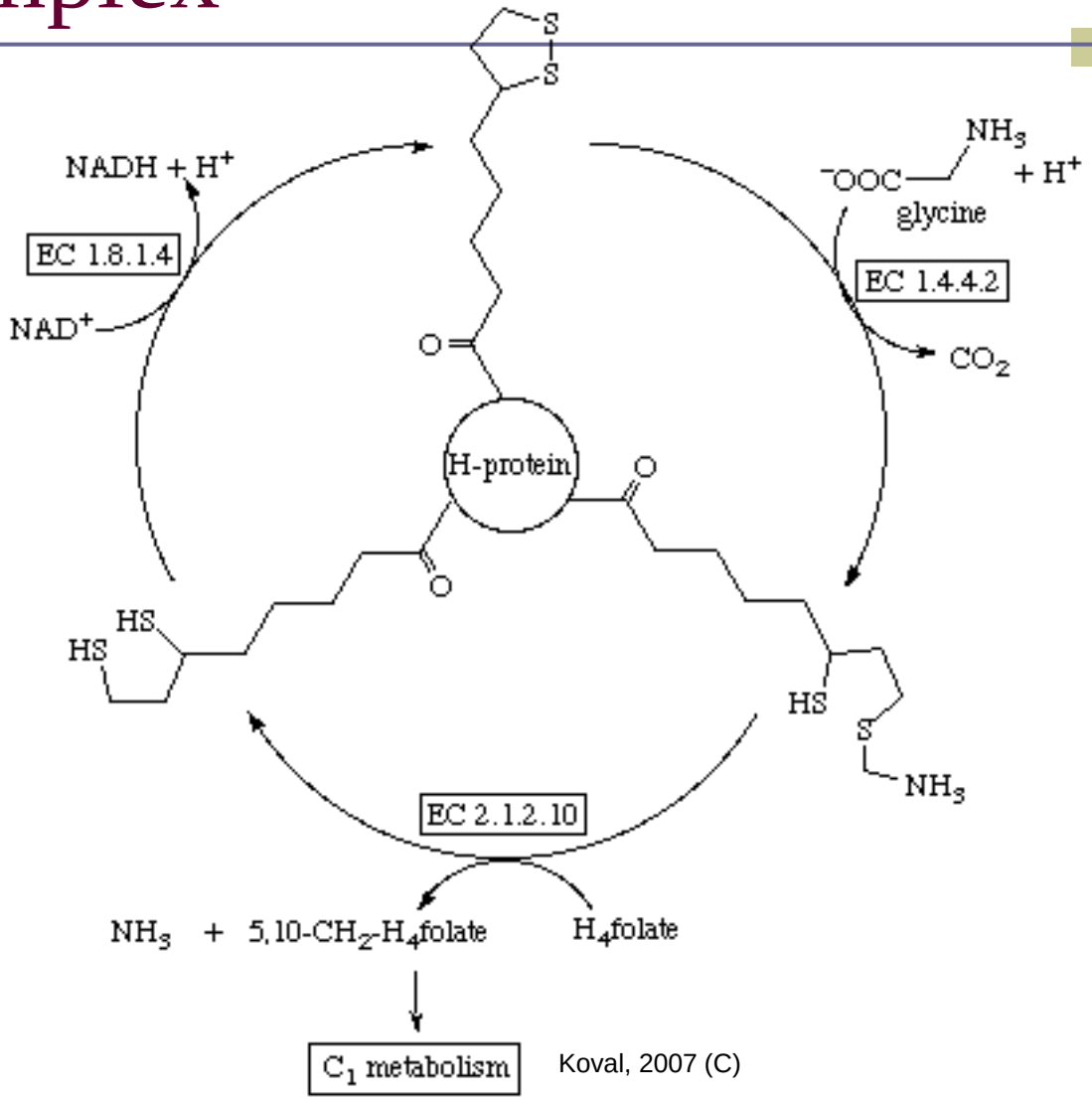
Structure of Glycine Cleavage Complex

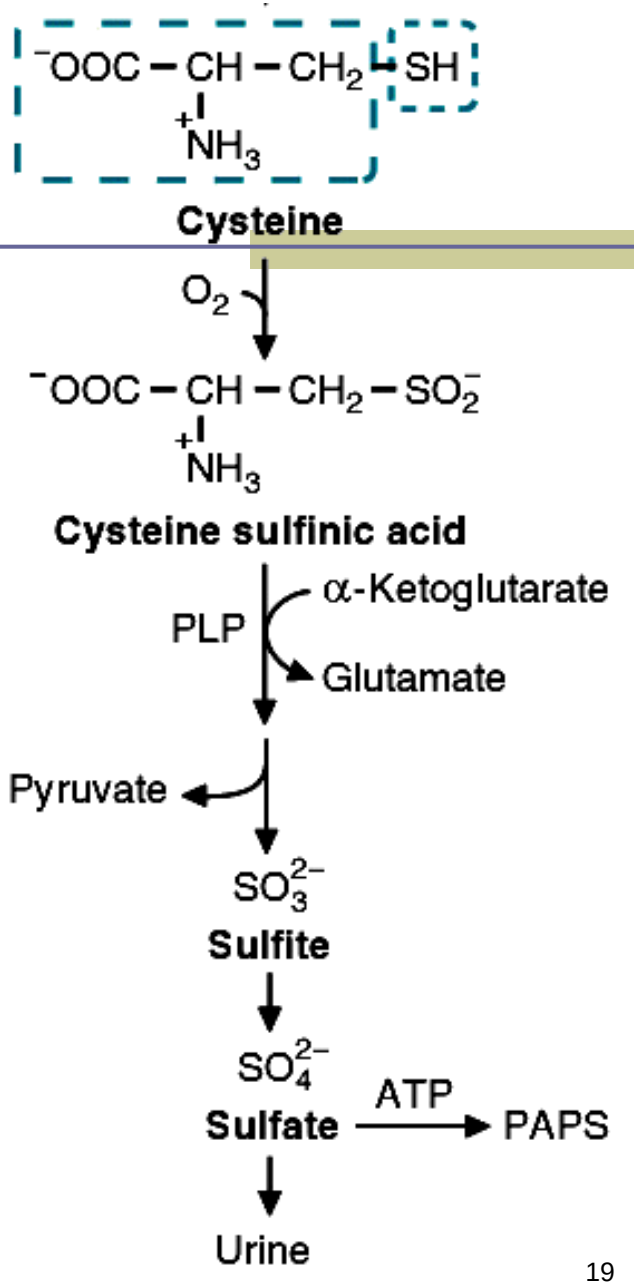


Cysteine Catabolism

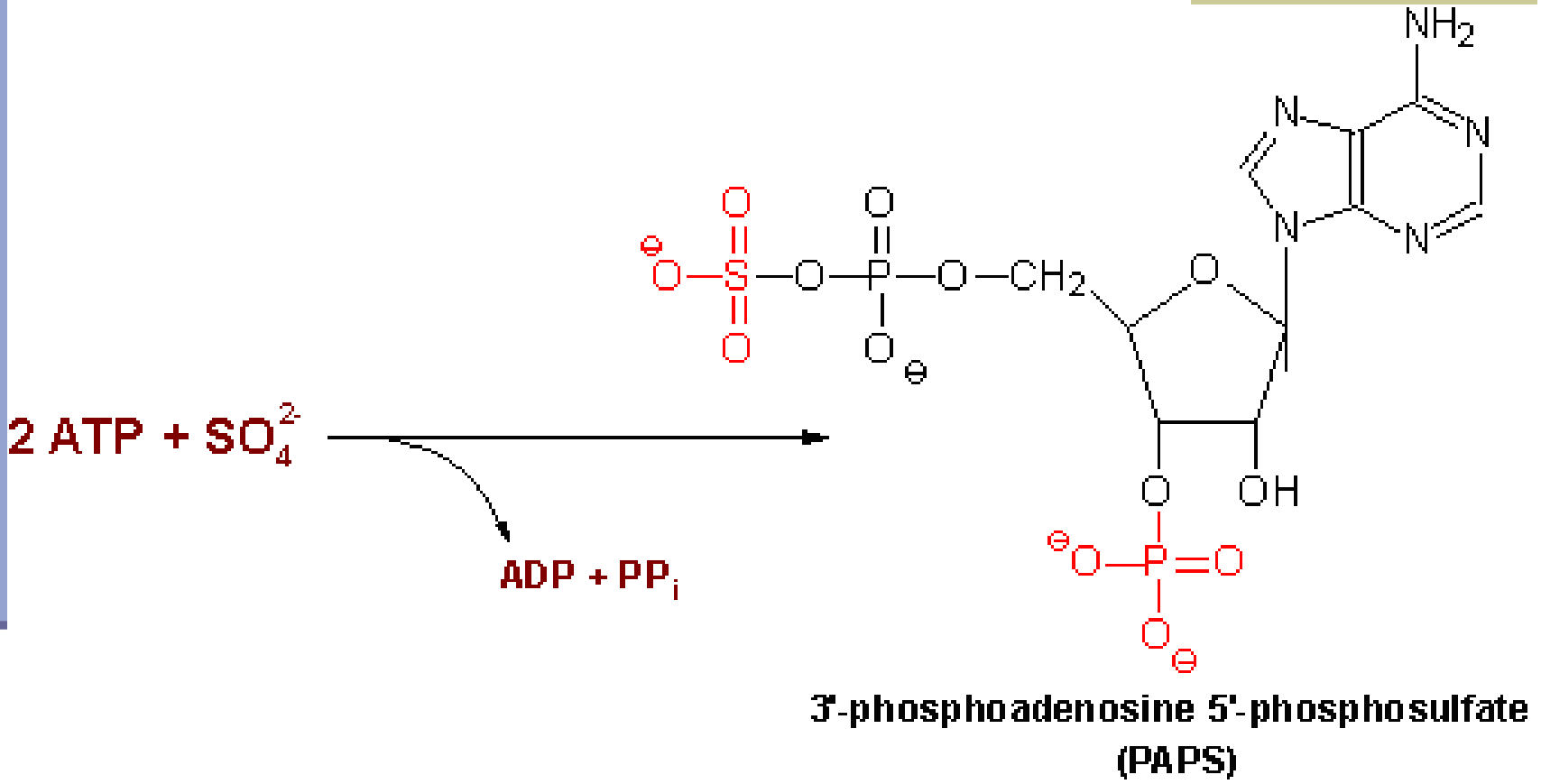
- There are several pathways for cysteine catabolism.
  - The simplest, but least important pathway is catalyzed by a liver desulfurase and produces hydrogen sulfide, (H_2S) and pyruvate.
  - The more important catabolic pathway is via a cytochrome-P_{450}-coupled enzyme, **cysteine dioxygenase** that oxidizes the cysteine sulfhydryl to sulfinate, producing the intermediate **cysteinesulfinate**.

Cysteine Metabolism-1

- Cysteinesulfinate can serve as a biosynthetic intermediate undergoing decarboxylation and oxidation to produce **taurine.**
- Catabolism of cysteinesulfinate proceeds through transamination to β-sulfinylpyruvate which is in undergoes desulfuration yielding bisulfite, (HSO_3^-) and the glucogenic product, pyruvate.
- The enzyme **sulfite oxidase** uses O_2 and H_2O to convert HSO_3^- to sulfate, (SO_4^-) and H_2O_2.
- The resultant sulfate is used as a precursor for the formation of *3'-phosphoadenosine-5'-phosphosulfate (PAPS)*.



Formation of 3'-phosphoadenosine-5'-phosphosulfate (PAPS)



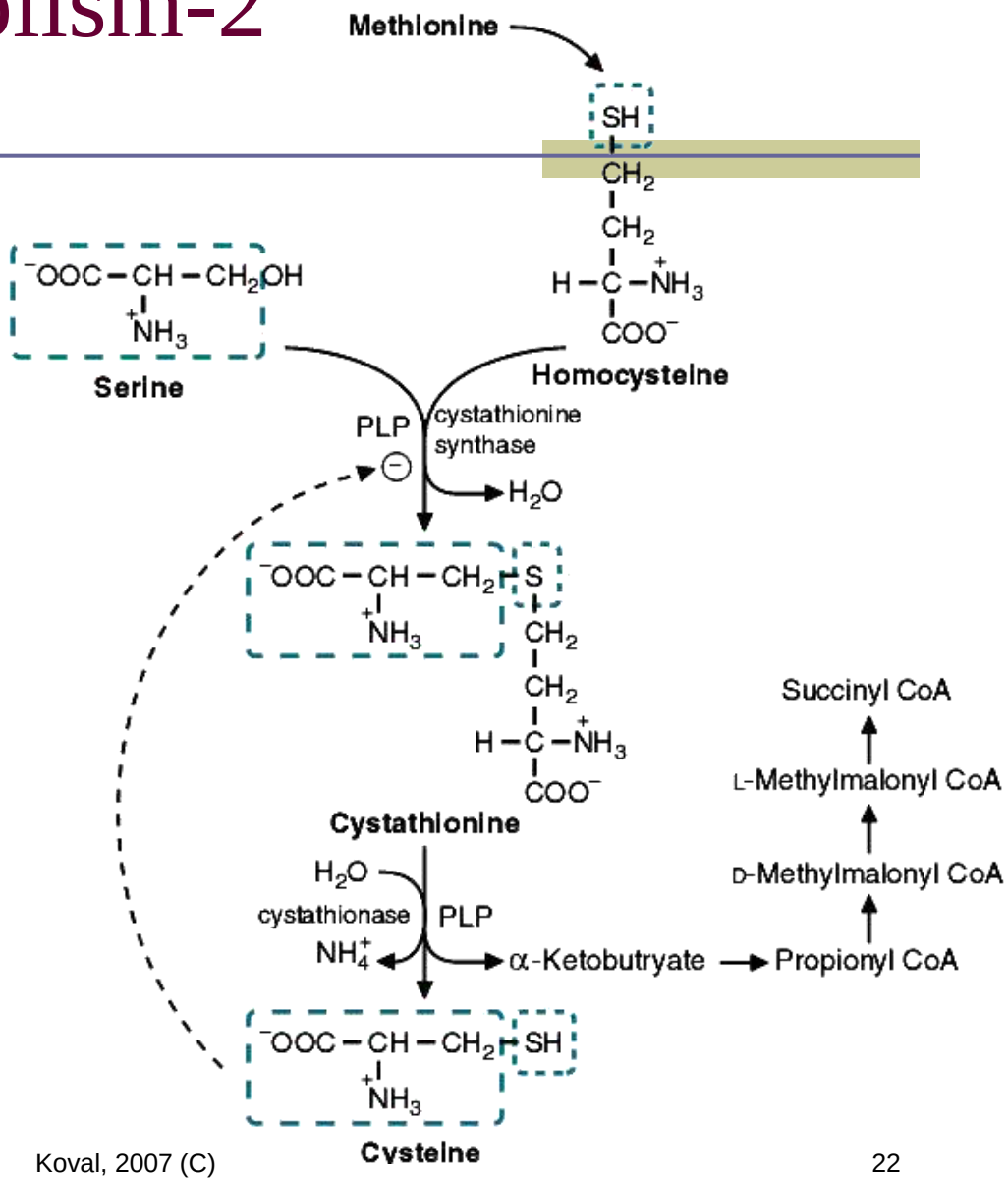
PAPS is used for the transfer of sulfate to biological molecules such as the sugars of the glycosphingolipids.

Other Cysteine Catabolism Pathways

- Other than protein, the most important product of cysteine metabolism is the bile salt precursor **taurine,** which is used to form the bile acid conjugates **taurocholate** and **taurochenodeoxycholate.**

Cysteine Metabolism-2

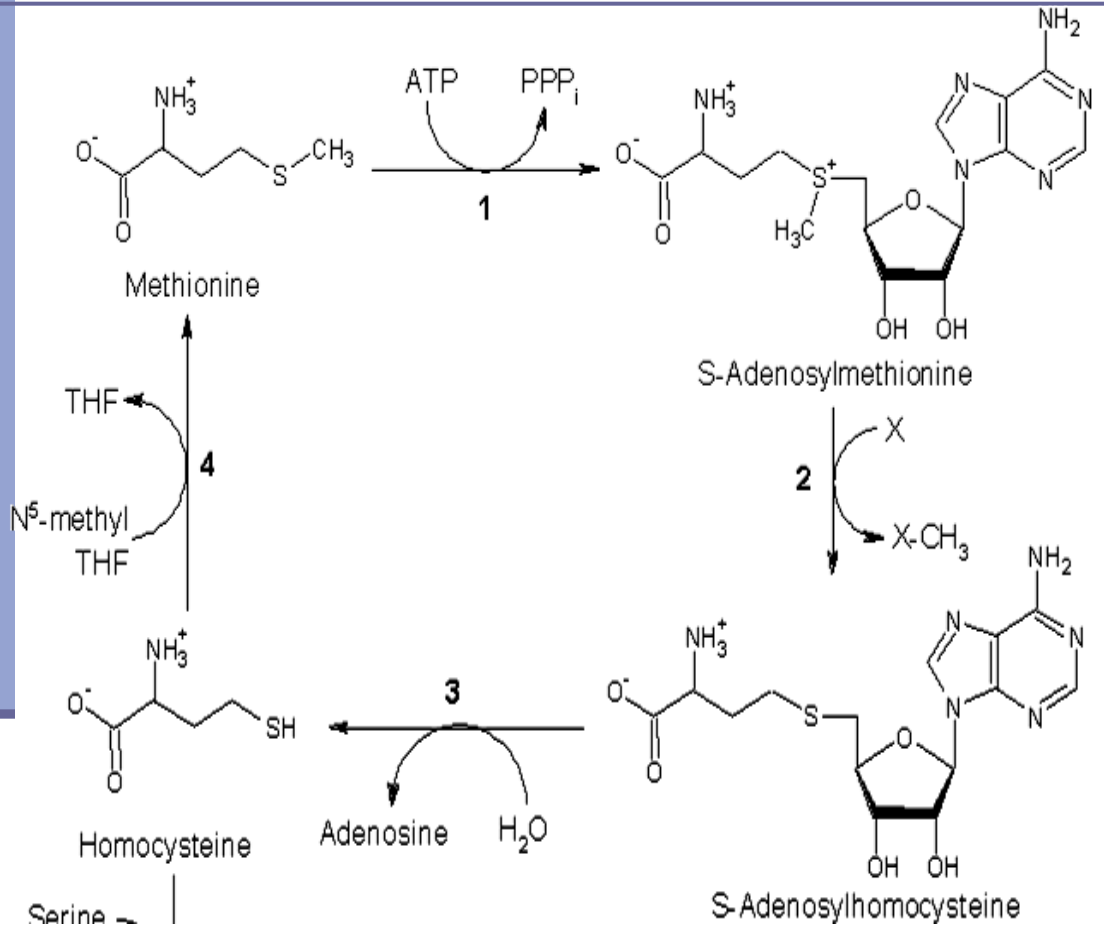
- The enzyme **cystathionase** can also transfer the sulfur from one cysteine to another generating thiocysteine and pyruvate.
- Transamination of cysteine yields β-mercaptopyruvate which then reacts with sulfite, (SO_3^{2-}), to produce thiosulfate, ($S_2O_3^{2-}$) and pyruvate.
  - Both thiocysteine and thiosulfate can be used by the enzyme **rhodanese** to incorporate sulfur into cyanide, (CN^-), thereby detoxifying the cyanide to thiocyanate.



Methionine Catabolism

- The principal fates of the essential amino acid methionine are incorporation into polypeptide chains, and use in the production of α-ketobutyrate and cysteine via SAM as described above.
  - The transulfuration reactions that produce cysteine from homocysteine and serine also produce α-ketobutyrate, the latter being converted to succinyl-CoA.
- Regulation of the methionine metabolic pathway is based on the availability of methionine and cysteine. If both amino acids are present in adequate quantities, SAM accumulates and is a positive effector on cystathionine synthase, encouraging the production of cysteine and α-ketobutyrate (both of which are glucogenic).
  - However, if methionine is scarce, SAM will form only in small quantities, thus limiting cystathionine synthase activity. Under these conditions accumulated homocysteine is remethylated to methionine, using N^5-methyl-THF and other compounds as methyl donors.

Methionine Cycle



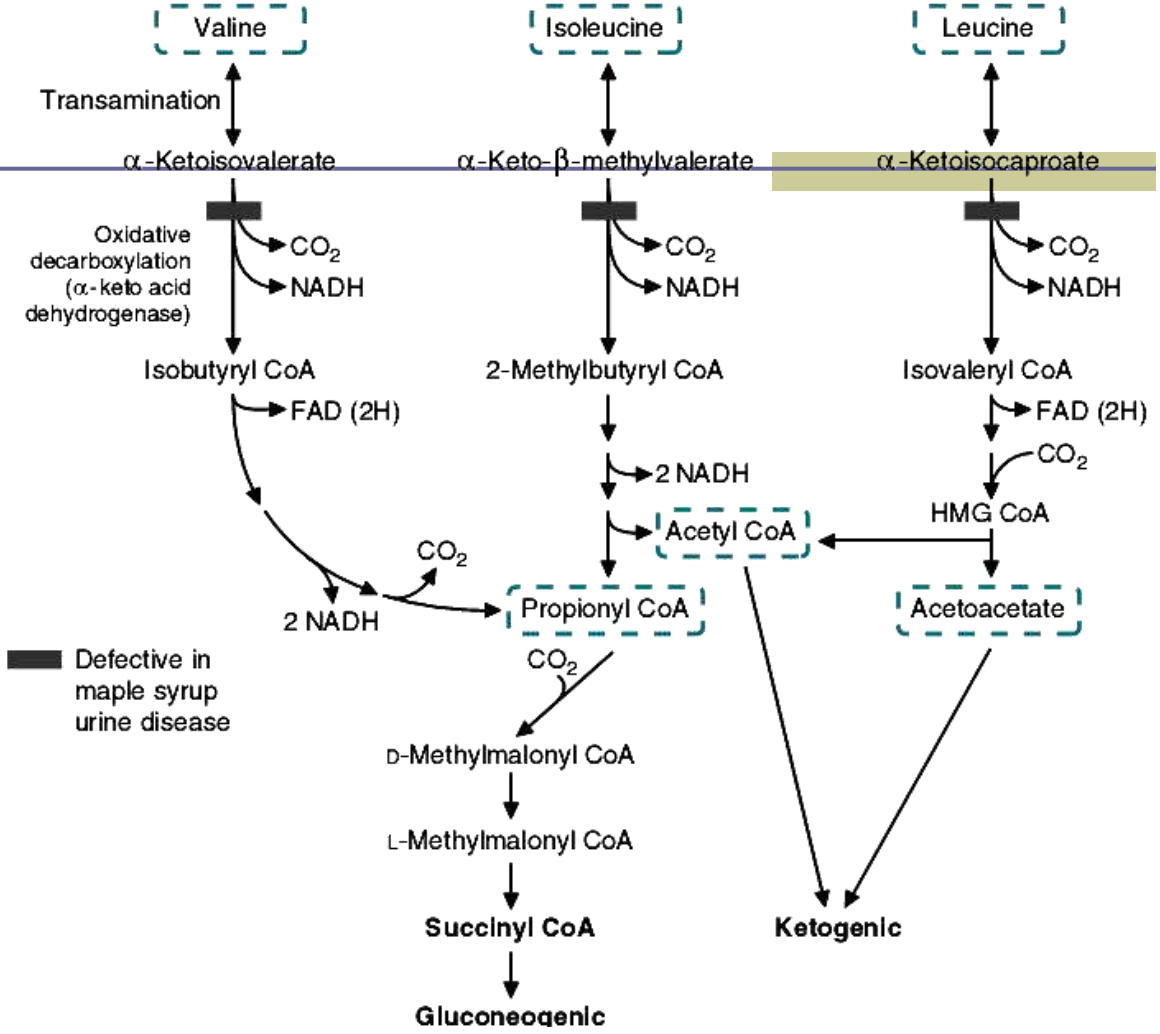
- Methionine is converted to S-adenosylmethionine (SAM) by (1) *methionine adenosyltransferase*.
- SAM serves as a methyl-donor in many (2) *methyltransferase* reactions and is conveted to S-adenosylhomocysteine (SAH).
- (3) *adenosylhomocysteinase* converts SAH to homocysteine.
- methionine can be regenerated from homocysteine via (4) *methionine synthase.*

Valine, Leucine and Isoleucine Catabolism

- This group of essential amino acids are identified as the **branched-chain amino acids, BCAAs**.
  - Because this arrangement of carbon atoms cannot be made by humans, these amino acids are an essential element in the diet.
- The catabolism of all three compounds initiates in muscle and yields NADH and $FADH_2$ which can be utilized for ATP generation.
- The catabolism of all three of these amino acids uses the same enzymes in the first two steps.

BCAA Catabolism-1

- The first step in each case is a transamination using a single BCAA aminotransferase, with α-ketoglutarate as amine acceptor. As a result, three different α-keto acids are produced and are oxidized using a common branched-chain α-keto acid dehydrogenase, yielding the three different CoA derivatives.
- Subsequently the metabolic pathways diverge, producing many intermediates.



BCAA Catabolism (2)

- The principal product from valine is propionylCoA, the glucogenic precursor of succinyl-CoA.
- Isoleucine catabolism terminates with production of acetylCoA and propionylCoA; thus isoleucine is both glucogenic and ketogenic.
- Leucine gives rise to acetylCoA and acetoacetylCoA, and is thus classified as strictly ketogenic.

Maple Syrup Urine Disease, MSUD

- There are a number of genetic diseases associated with faulty catabolism of the BCAAs.
  - The most common defect is in the branched-chain α-keto acid dehydrogenase.
    - all three α-keto acids accumulate and are excreted in the urine.
  - Maple syrup urine disease because of the characteristic odor of the urine in afflicted individuals.
    - Mental retardation in these cases is extensive.
    - The life of afflicted individuals is short and development is abnormal.
    - The main neurological problems are due to poor formation of myelin in the CNS.

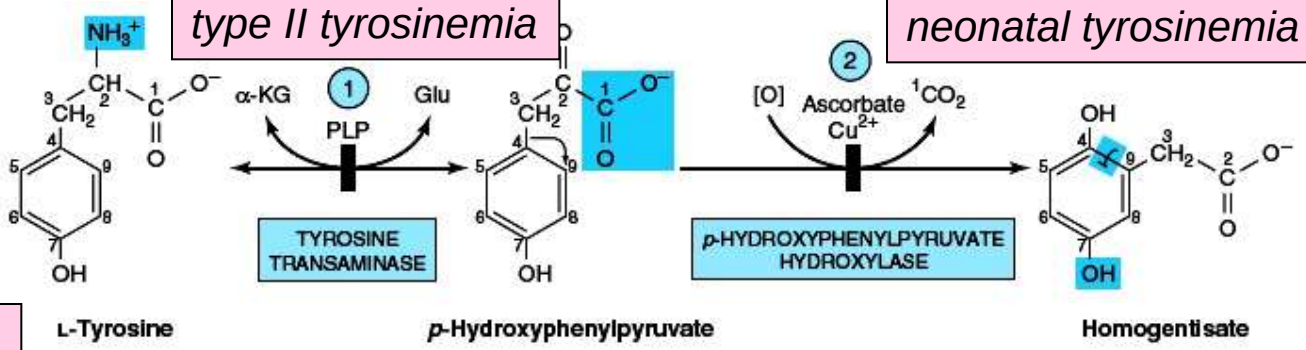
Phenylalanine and Tyrosine Catabolism

1. Incorporated into polypeptide chains
2. Production of tyrosine from phenylalanine.
   - Phe and Tyr degradation – conversion to fumarate and acetoacetate: both glucogenic and ketogenic AA.
3. Biosynthesis of physiologically important metabolites e.g.
   - Dopamine
   - Norepinephrine
   - Epinephrine.

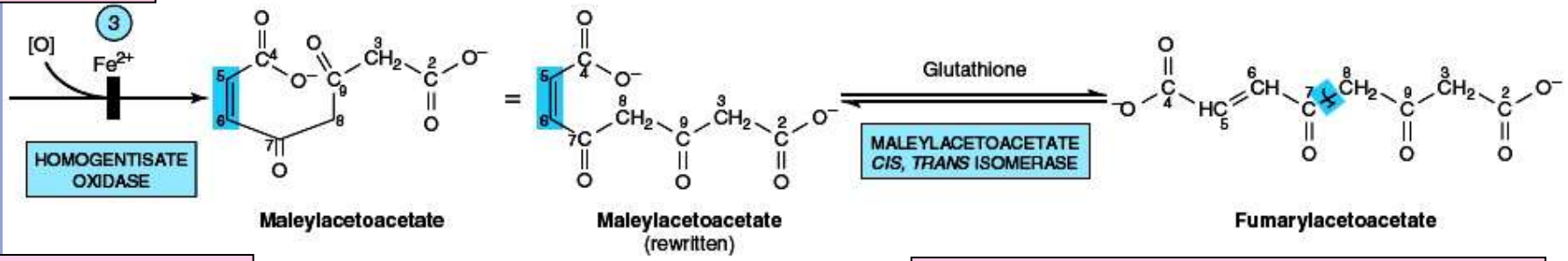
Aromatic Amino Acid Catabolism

type II tyrosinemia

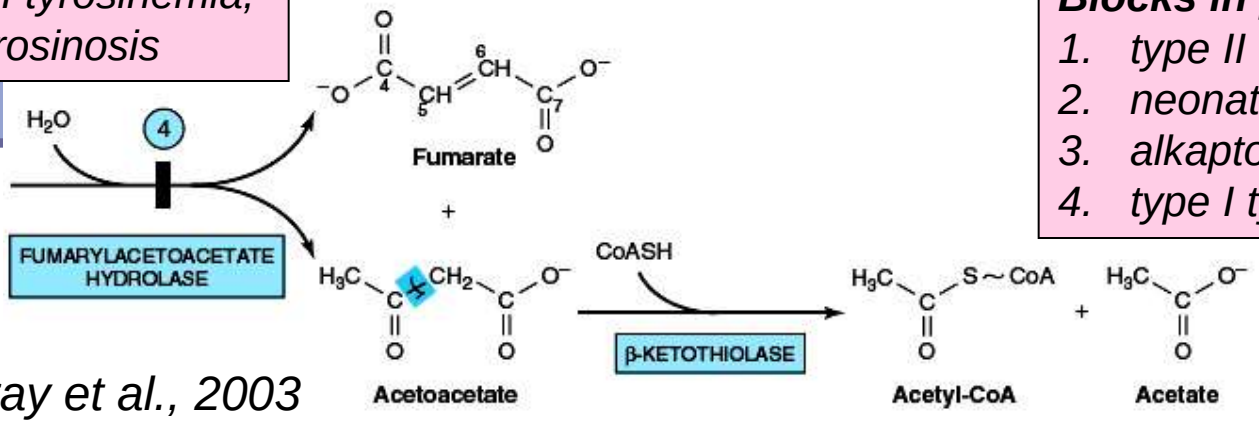
neonatal tyrosinemia



alkaptonuria



type I tyrosinemia, or tyrosinosis



Blocks in pathology

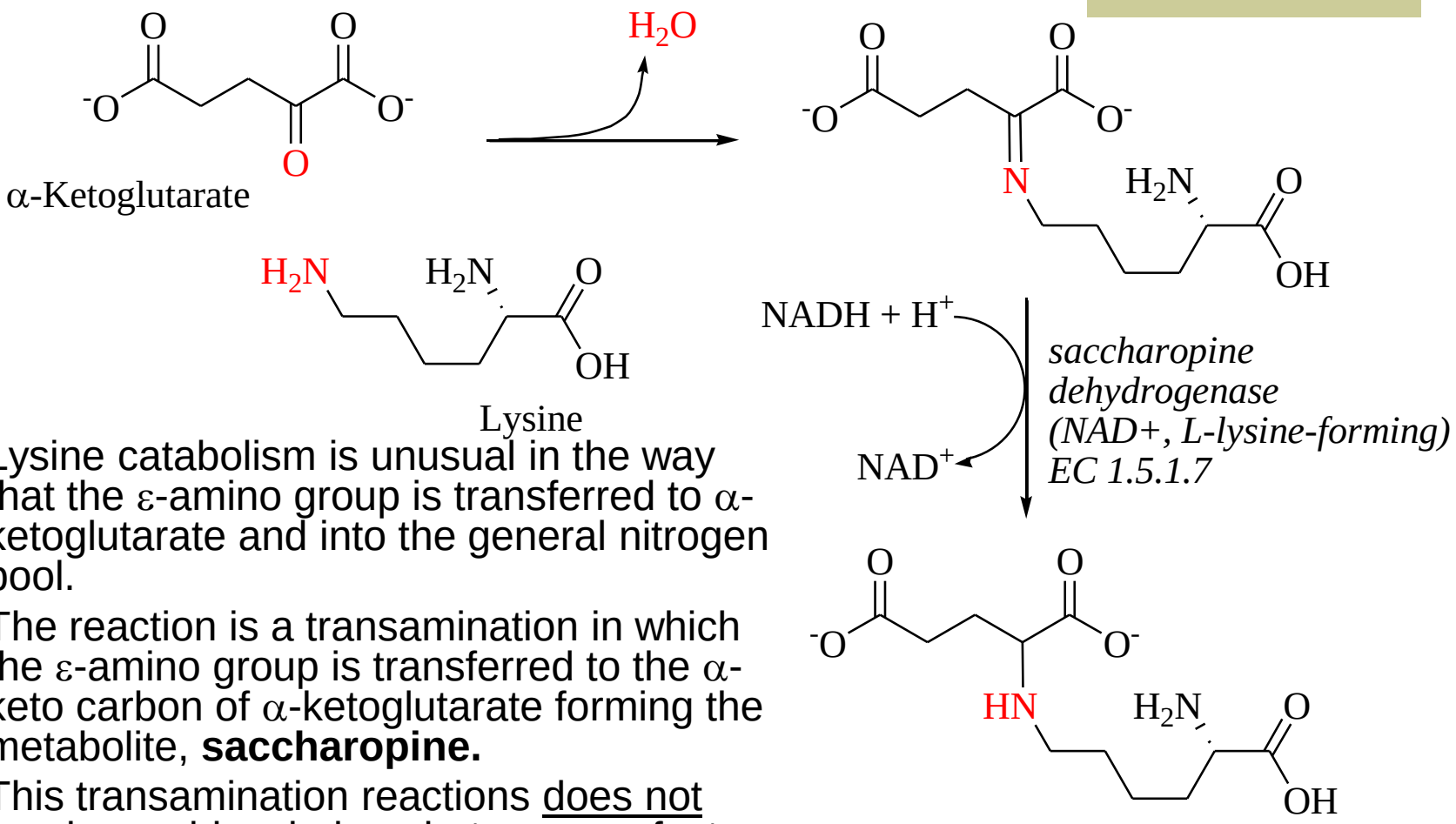
1. *type II tyrosinemia;*
2. *neonatal tyrosinemia;*
3. *alkaptonuria;*
4. *type I tyrosinemia, or tyrosinosis*

Murray et al., 2003

Defective Aromatic AA Catabolism

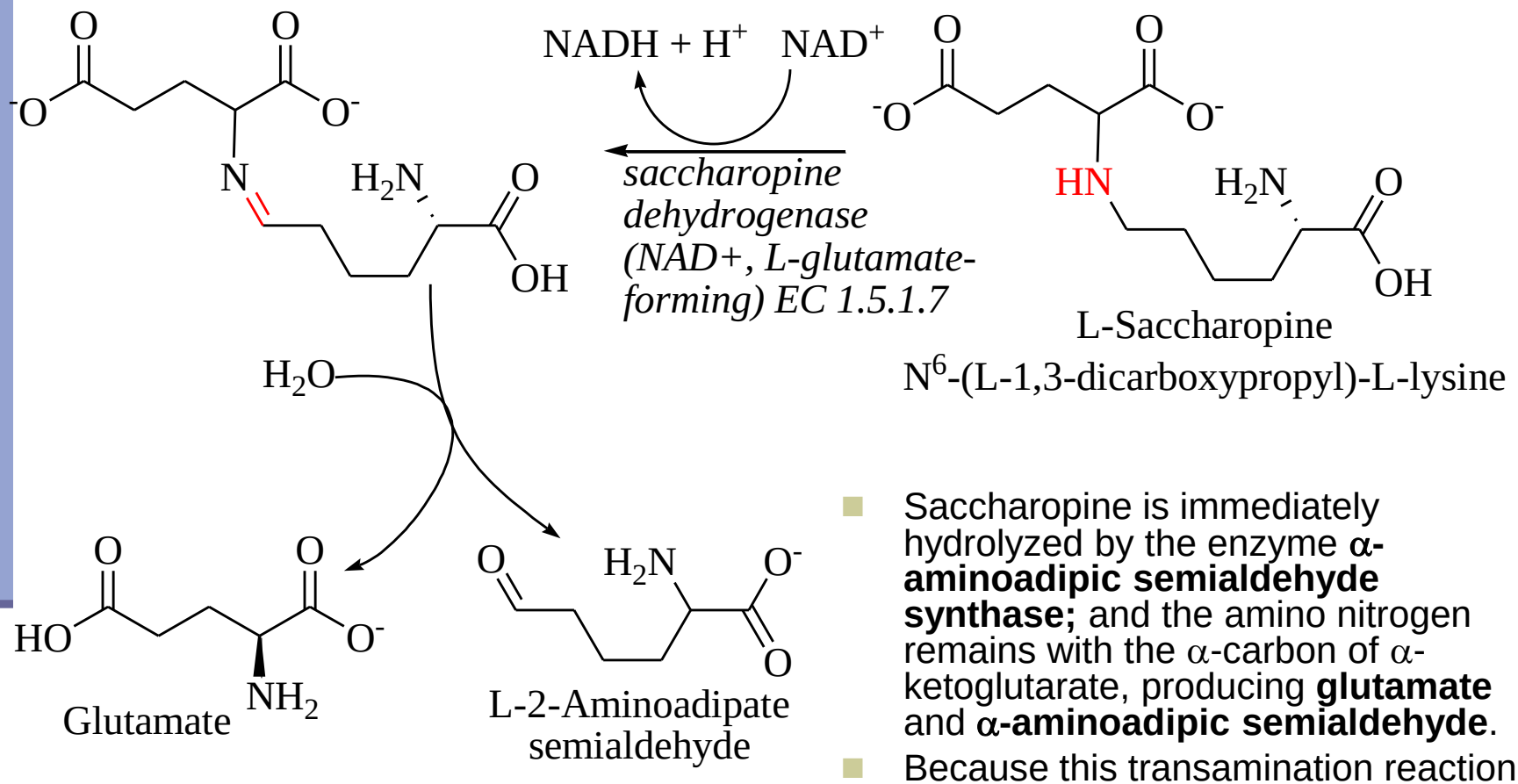
- Deficiency of **tyrosine aminotransferase** (TAT) leads to:
  - Hypertyrosinemia
  - Urinary excretion of tyrosine and the catabolic intermediates
  - The neurological symptoms are similar for both deficiencies.
    - Hypertyrosinemia leads to painful corneal eruptions and photophobia.
- **Alkaptonuria:** defective **homogentisic acid oxidase**.
  - Homogentisic acid accumulation is relatively innocuous, causing urine to darken on exposure to air, but no life-threatening effects accompany the disease.

Lysine Catabolism



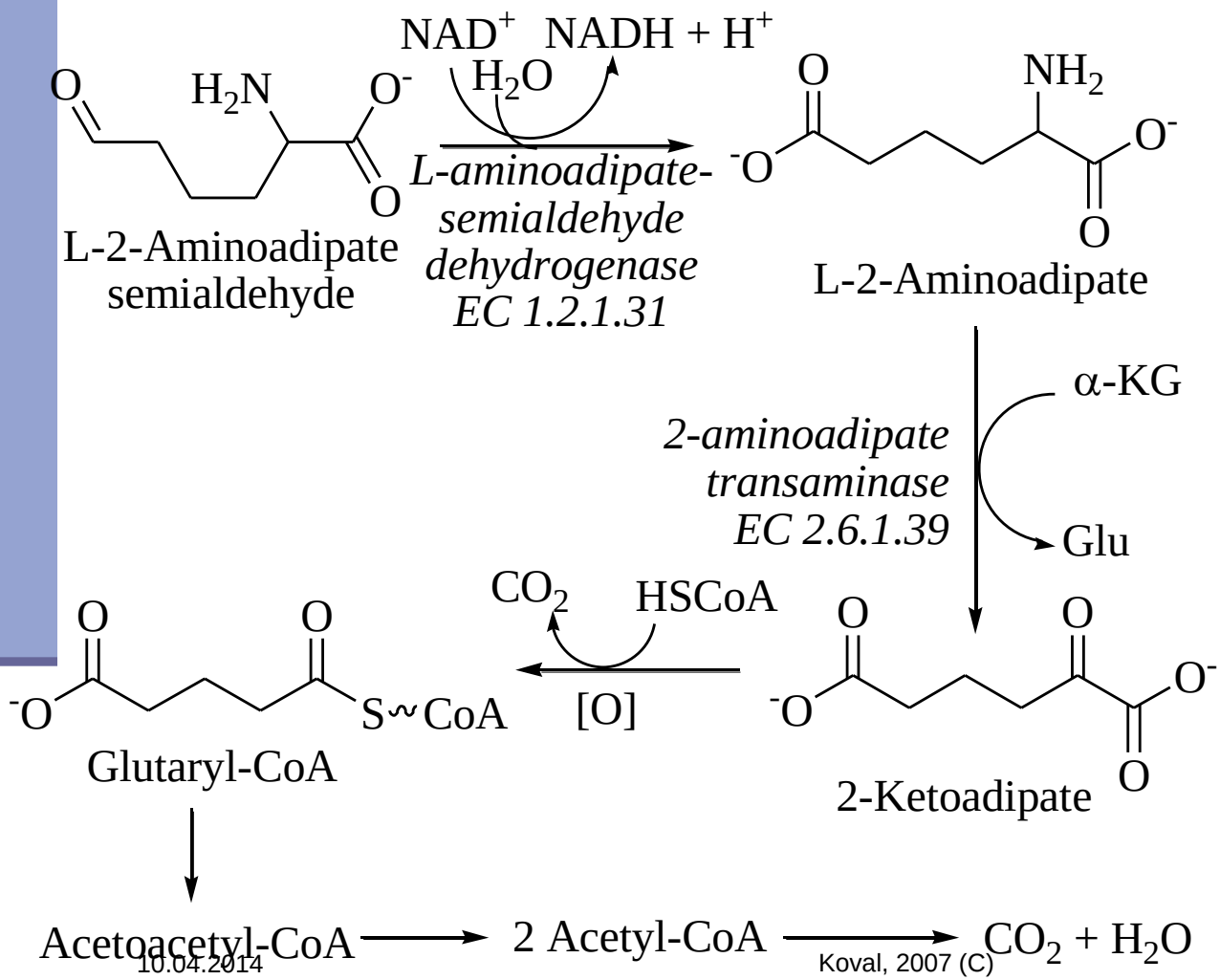
- Lysine catabolism is unusual in the way that the ε-amino group is transferred to α-ketoglutarate and into the general nitrogen pool.
- The reaction is a transamination in which the ε-amino group is transferred to the α-keto carbon of α-ketoglutarate forming the metabolite, **saccharopine.**
- This transamination reactions does not employ pyridoxal phosphate as a cofactor.

Lysine Catabolism (cont'd)



- Saccharopine is immediately hydrolyzed by the enzyme **α-aminoadipic semialdehyde synthase;** and the amino nitrogen remains with the α-carbon of α-ketoglutarate, producing **glutamate** and **α-aminoadipic semialdehyde**.
- Because this transamination reaction is not reversible, lysine is an essential amino acid.

Lysine Catabolism (final of pathway)



- The ultimate end-product of lysine catabolism is **acetoacetyl-CoA.**
  - This one catabolizes by β-oxidation pathway to 2 acetyl-CoA
  - CO_2 and H_2O are produced in TCA.

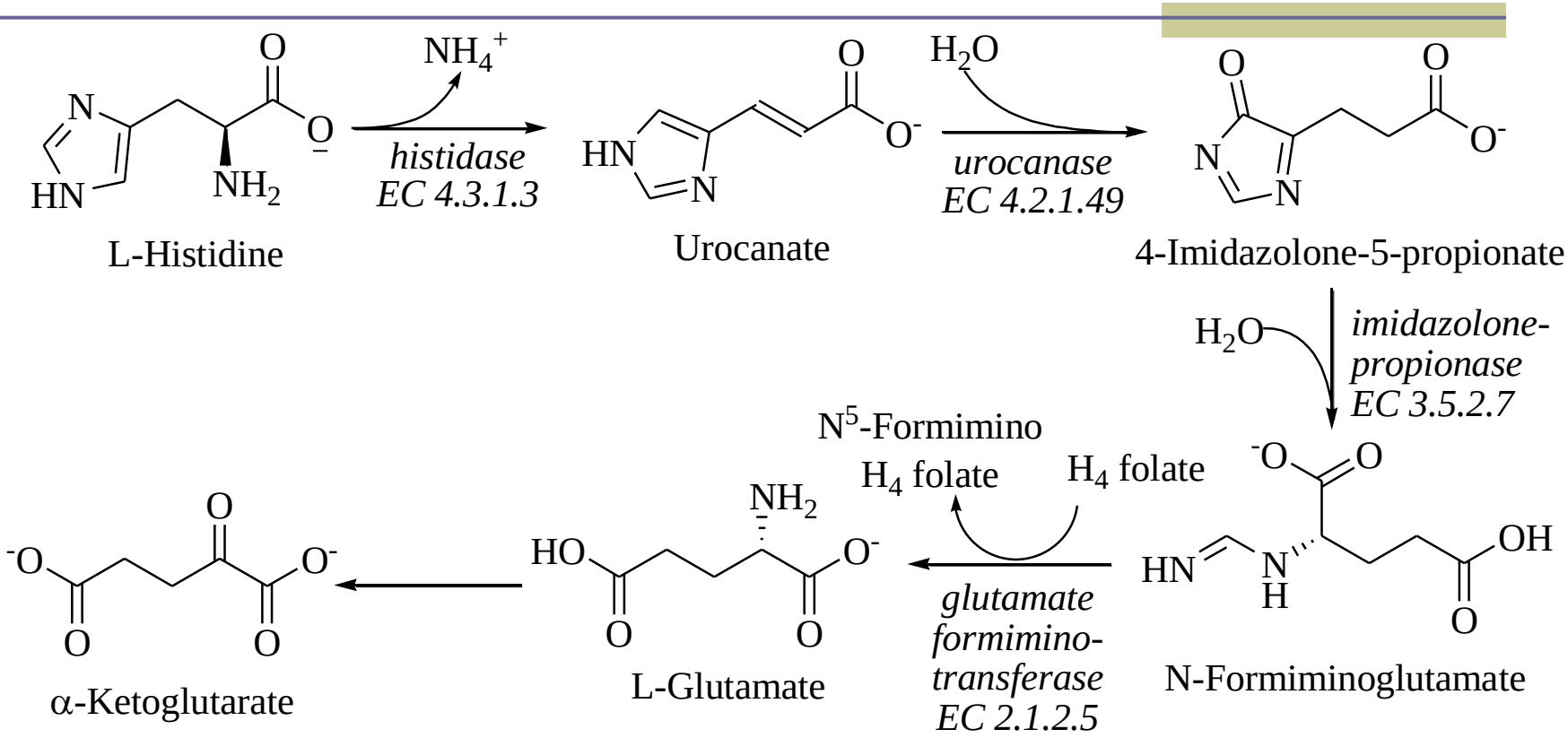
Pathology of Lysine Catabolism

- Genetic deficiencies in the enzyme **α-aminoadipic semialdehyde synthase** have been observed in individuals who excrete large quantities of urinary lysine and some saccharopine.
  - The **lysinemia** and associated **lysinuria** are benign.
- Other serious disorders associated with lysine metabolism are due to failure of the transport system for lysine and the other **dibasic** amino acids across the intestinal wall.
- Lysine is essential for protein synthesis;
  - a deficiencies of its transport into the body can cause seriously diminished levels of protein synthesis.
  - Probably more significant however, is the fact that arginine is transported on the same dibasic amino acid carrier, and resulting arginine deficiencies limit the quantity of ornithine available for the urea cycle.
  - The result is severe **hyperammonemia** after a meal rich in protein.
  - The addition of **citrulline** to the diet prevents the hyperammonemia.

Lysine is Important for Carnitine Synthesis

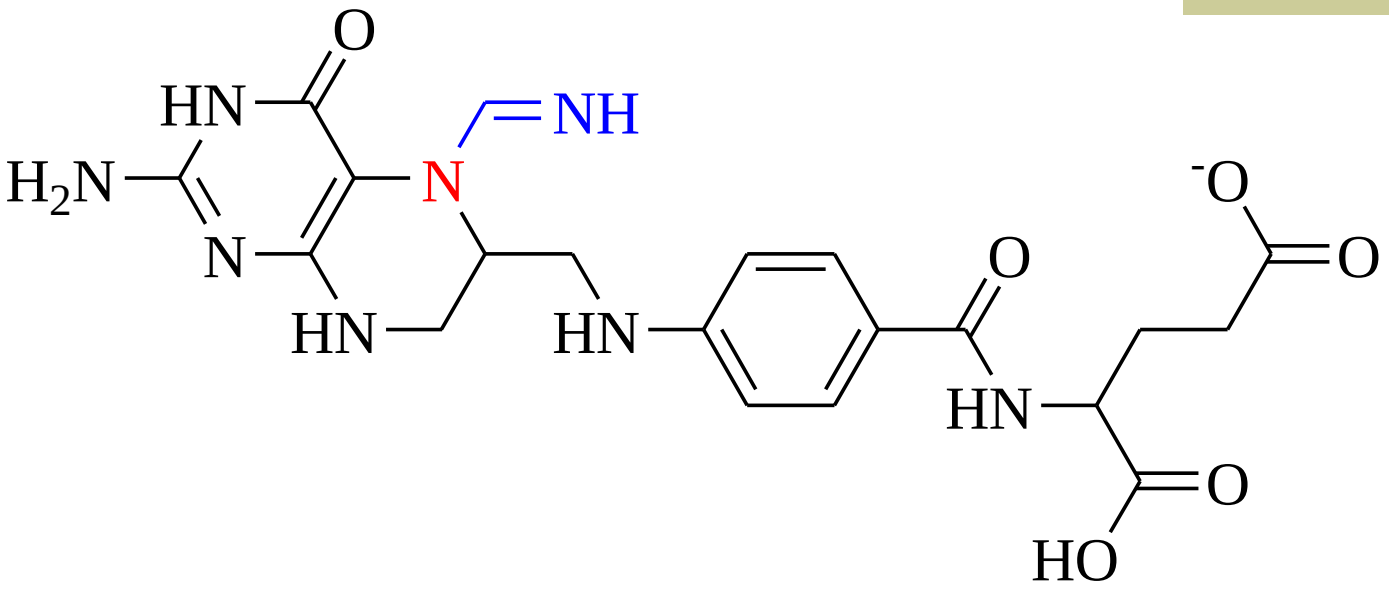
- Lysine is also important as a precursor for the synthesis of **carnitine**, required for the transport of fatty acids into the mitochondria for oxidation.
  - Free lysine does not serve as the precursor for this reaction, rather the modified lysine found in certain proteins.
  - Some proteins modify lysine to **trimethyllysine** using **SAM** as the methyl donor to transfer methyl groups to the ε-amino of the lysine side chain.
  - Hydrolysis of proteins containing trimethyllysine provide the substrate for the subsequent conversion to **carnitine**.

Histidine Catabolism



- **Histidine catabolism** begins with release of the α-amino group catalyzed by *histidase*.
- The deaminated product, **urocanate**, is not the usual α-keto acid.
- The end product of histidine catabolism is glutamate (histidine is glucogenic amino acids).

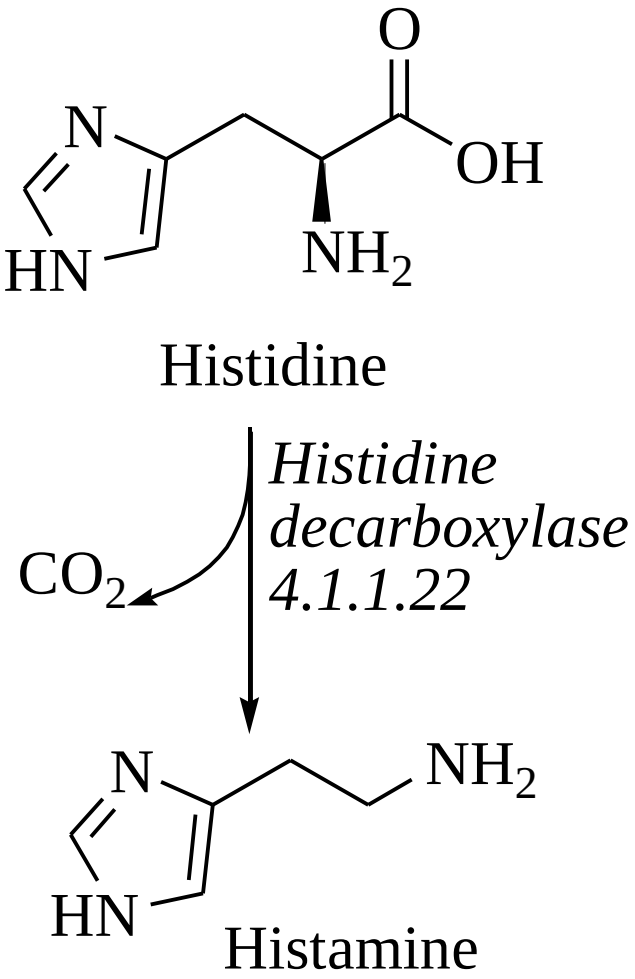
Histidine is a Source of N^5-formiminoTHF



N^5-formimidoyl tetrahydrofolate

- Another key feature of histidine catabolism is that it serves as a source of ring nitrogen to combine with tetrahydrofolate (THF), producing the 1-carbon THF intermediate known as ***N*^{5}-formiminoTHF** (or N^5-formimidoyl tetrahydrofolate).
- The latter reaction is one of two routes to *N*^{5}-formiminoTHF.

Decarboxylation of Histidine

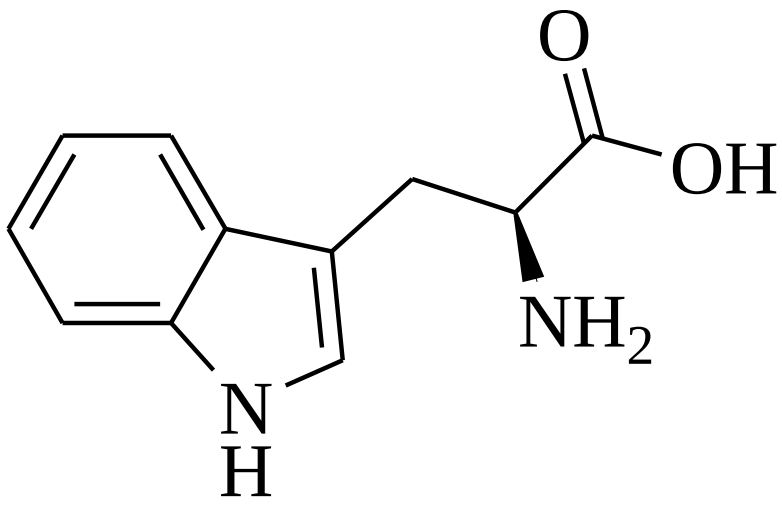


- Decarboxylation of histidine in the intestine by bacteria gives rise to **histamine.**
- Similarly, histamine arises in many tissues by the decarboxylation of histidine, which in excess causes constriction or dilation of various blood vessels.
- The general symptoms are those of **asthma** and various **allergic reactions**.

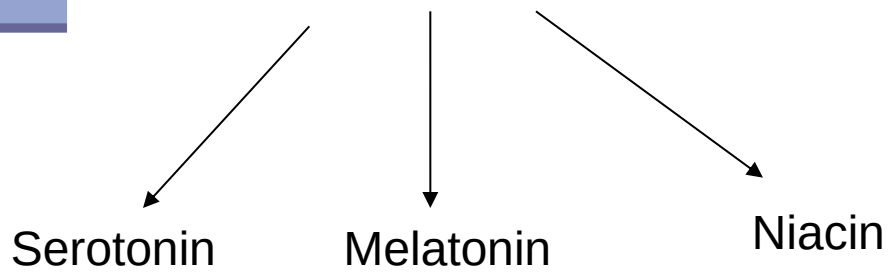
Histidine Catabolism Pathology

- The principal genetic deficiency associated with histidine metabolism is absence or deficiency of the first enzyme of the pathway, **histidase**.
- The resultant **histidinemia** is relatively benign.
  - The disease, which is of relatively high incidence (1 in 10,000), is most easily detected by the absence of urocanate from skin and sweat, where it is normally found in relative abundance.

Tryptophan

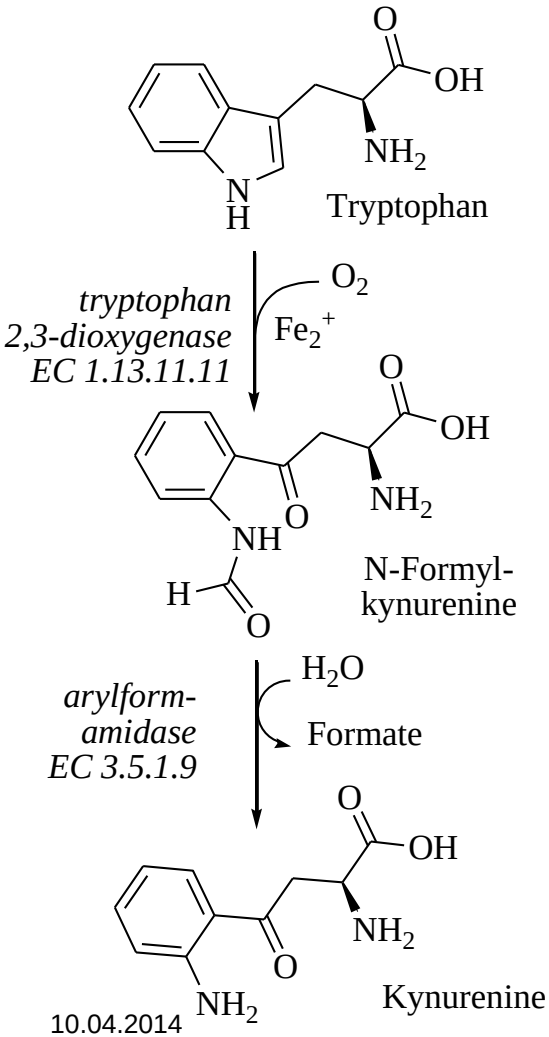


Tryptophan



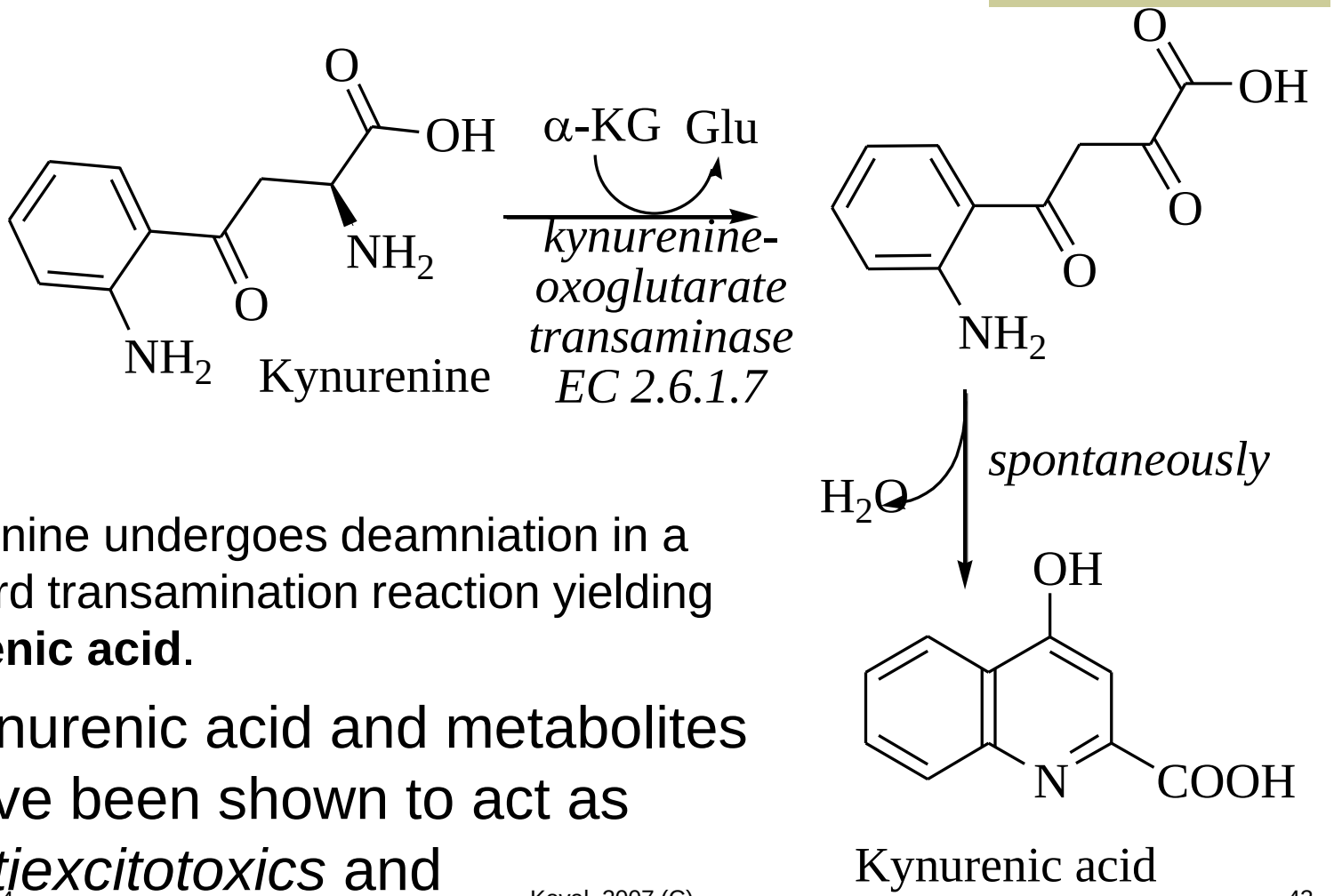
- The main function of tryptophan is as a building block in **protein synthesis**.
- Tryptophan is a precursor for **serotonin** (a neurotransmitter), **melatonin** (a neurohormone), and **niacin**.
  - The functional group of trytophan is **indole**.
- Tryptophan has been implicated as a possible cause of **schizophrenia** in people who cannot metabolize it properly.
  - When improperly metabolized it creates a waste product in the brain which is toxic and causes hallucinations and delusions.
  - Tryptophan has also been indicated as an aid for schizophrenic patients.

Tryptophan Catabolism: Kynurenic Acid Formation (1)



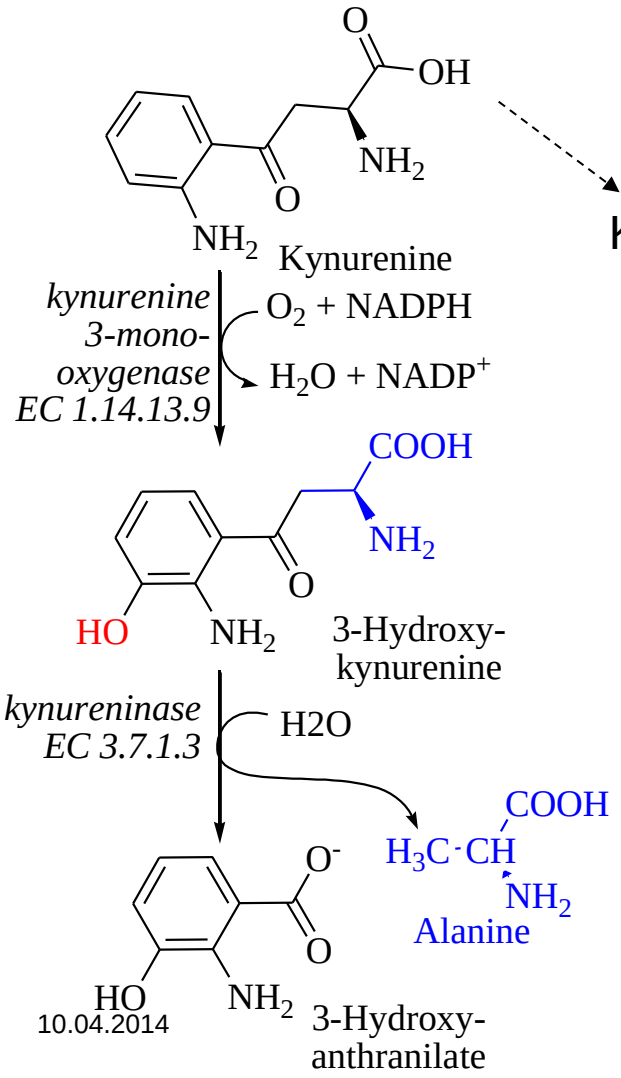
- A number of important side reactions occur during the catabolism of tryptophan on the pathway to **acetoacetate**.
  - The first enzyme of the catabolic pathway is an iron **tryptophan 2,3-dioxygenase** that opens the indole ring.
  - The latter enzyme is highly inducible, its concentration rising almost 10-fold on a diet high in tryptophan.
- **Kynurenine** is the first key branch point intermediate in the pathway.

Kynurenic Acid Formation (2)



- Kynurenine undergoes deamniation in a standard transamination reaction yielding **kynurenic acid**.
  - Kynurenic acid and metabolites have been shown to act as *antiexcitotoxics* and *anticonvulsives*.

Tryptophan Catabolism: Kynurenine Degradation



- A second side branch reaction produces **anthranilic acid** plus **alanine**.
  - Another equivalent of alanine is produced further along the main catabolic pathway, and it is the production of these alanine residues that allows tryptophan to be classified among the glucogenic and ketogenic amino acids.

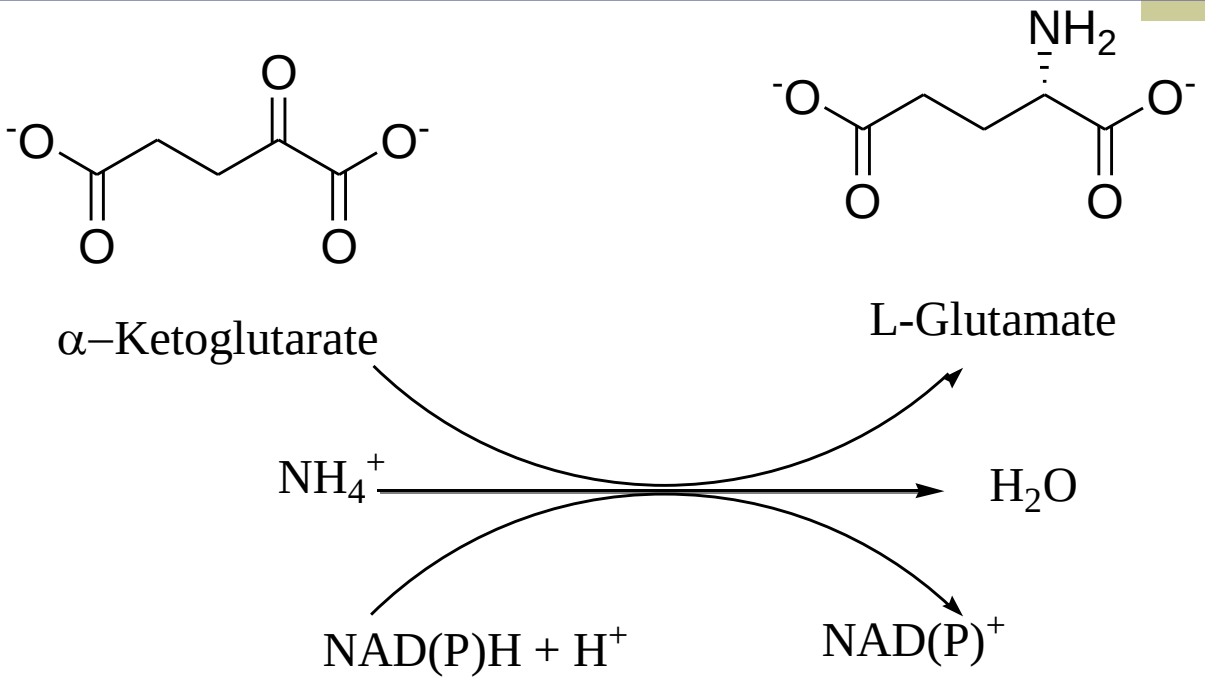
Further Tryptophan Catabolism

- The second important branch point converts kynurenine into 2-amino-3-carboxymuconic semialdehyde, which has two fates.
  - The main flow of carbon – to **glutarate**.
  - Important side reaction in liver – transamination, → nicotinic acid → small amount of NAD^+ and $NADP^+$
- Also Trp roles as AA in protein biosynthesis, precursor of **serotonin** and **melatonin**.

Non Essential Amino Acid Biosynthesis

- Glutamate and Aspartate
- Alanine and the Glucose-Alanine Cycle
- Cysteine
- Tyrosine
- Ornithine and Proline
- Serine
- Glycine
- Aspartate/Asparagine and Glutamate/Glutamine

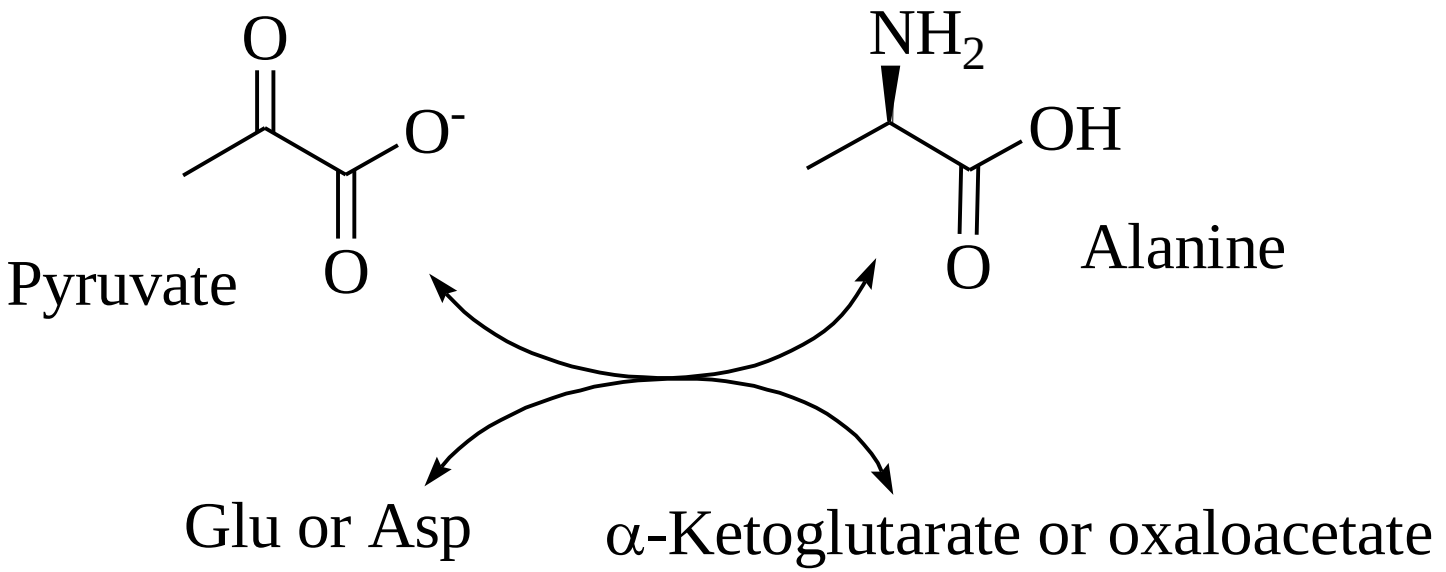
Glutamate and Aspartate



The glutamate dehydrogenase

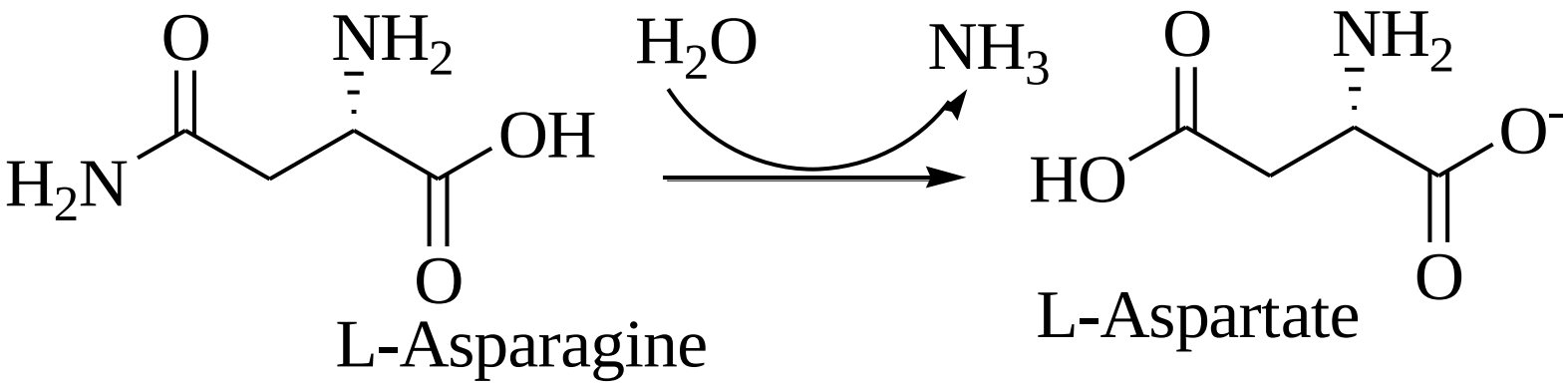
- Glutamate and aspartate are synthesized from their widely distributed α-keto acid precursors by simple 1-step transamination reactions. The former catalyzed by glutamate dehydrogenase and the latter by aspartate aminotransferase, AST.

Transaminase reactions



- **Transamination** is the most widespread form of nitrogen transfer.

Asparaginase



- Aspartate is also derived from asparagine through the action of **asparaginase**.

Alanine and the Glucose-Alanine Cycle

- Aside from its role in protein synthesis, **alanine** is second only to glutamine in prominence as a circulating amino acid.
  - In this capacity it serves a unique role in the transfer of nitrogen from peripheral tissue to the liver.
  - Alanine is transferred to the circulation by many tissues, but mainly by muscle, in which alanine is formed from pyruvate at a rate proportional to intracellular pyruvate levels.

Glucose-Alanine Cycle

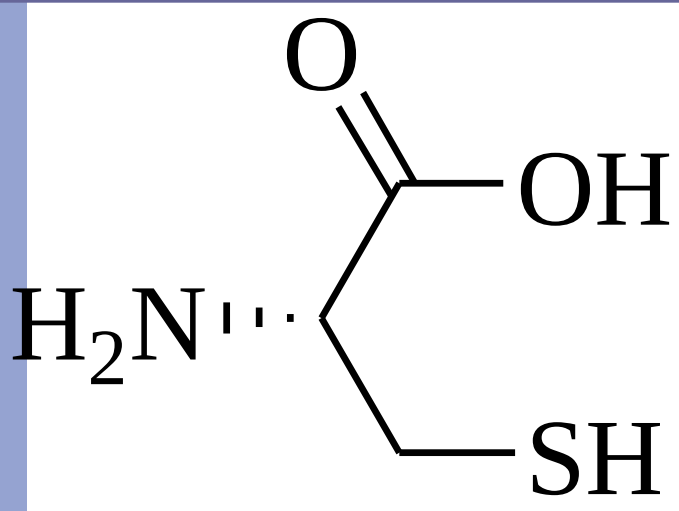
- Liver accumulates plasma alanine, reverses the transamination that occurs in muscle, and proportionately increases urea production.
- The pyruvate is either oxidized or converted to glucose via gluconeogenesis.
  - When alanine transfer from muscle to liver is coupled with glucose transport from liver back to muscle, the process is known as the **glucose-alanine cycle**.
  - The key feature of the cycle is that in 1 molecule, alanine, peripheral tissue exports pyruvate and ammonia (which are potentially rate-limiting for metabolism) to the liver, where the carbon skeleton is recycled and most nitrogen eliminated.

2 Main Pathways of Muscle Alanine Production

- There are 2 main pathways to production of muscle alanine:
  - directly from protein degradation,
  - and via the transamination of pyruvate by **glutamate-pyruvate aminotransferase** (also called *alanine transaminase,* ***ALT***).

glutamate + pyruvate ↔ α-KG + alanine

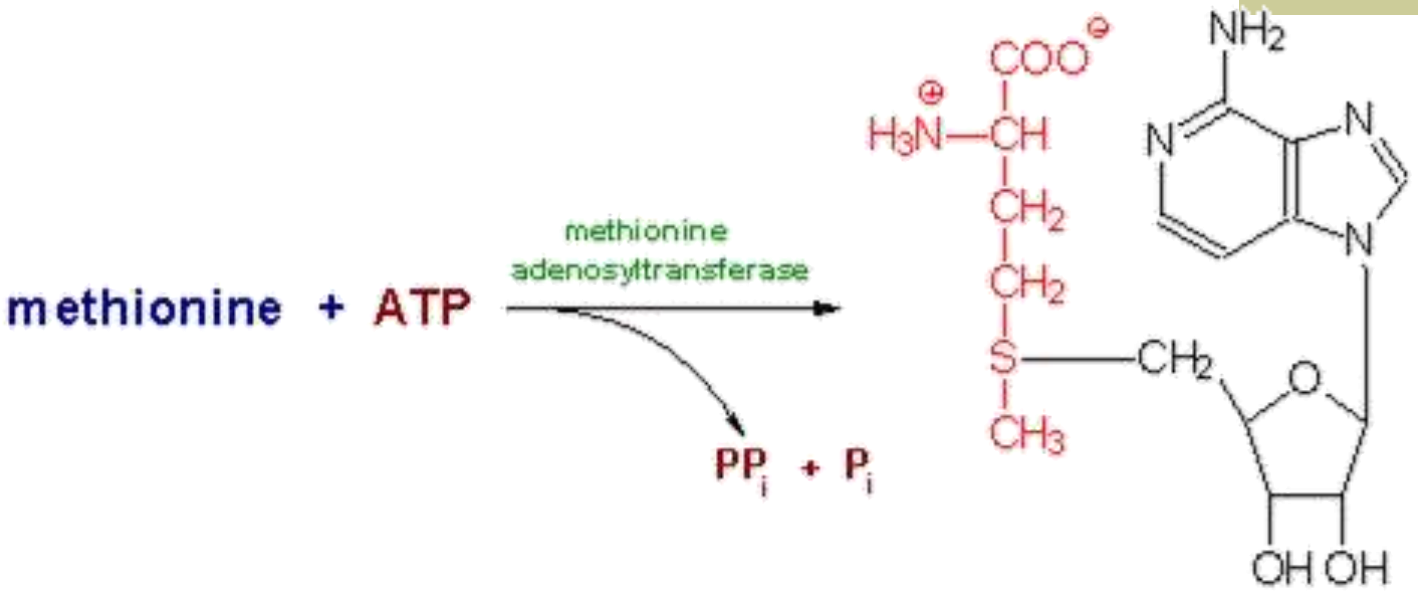
Cysteine metabolism



cysteine

- Synthesis
  - Cysteine derives its carbon and nitrogen from **serine**.
  - The essential amino acid methionine supplies the sulfur.
- Degradation
  - Forms pyruvate. Its sulfur is converted to H_2SO_4.

Cysteine Biosynthesis

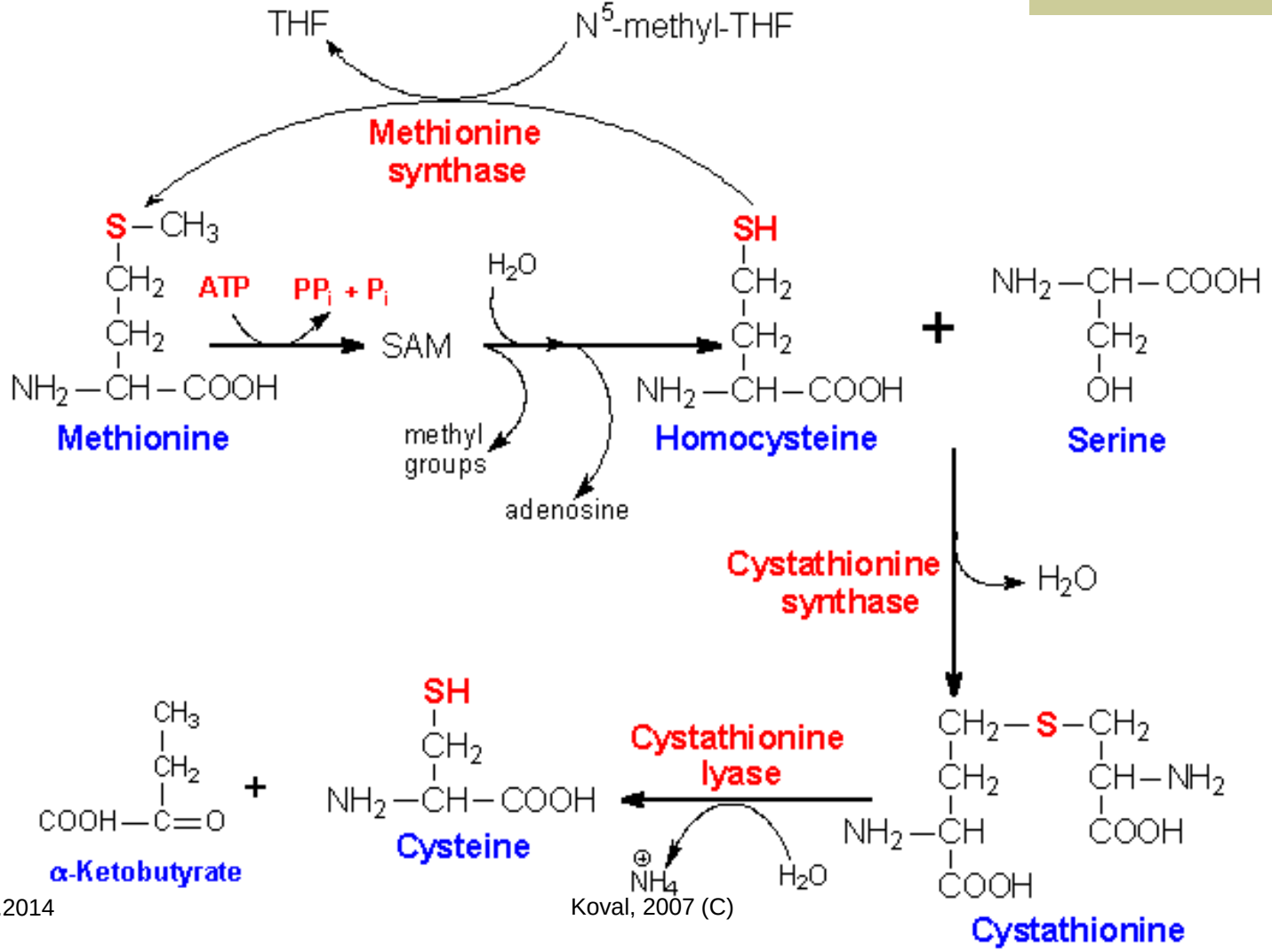


- The sulfur for cysteine synthesis comes from the essential amino acid methionine.
  - A condensation of ATP and methionine catalyzed by methionine adenosyltransferase yields S-adenosylmethionine (**SAM or AdoMet**).

Transmethylation Reactions

- SAM serves as a precurosor for numerous methyl transfer reactions (e.g. the conversion of norepinephrine to epinenephrine).
- The result of methyl transfer is the conversion of **SAM** to **S-adenosylhomocysteine**.
  - S-adenosylhomocysteine is then cleaved by adenosylhomocyteinase to yield homocysteine and adenosine.
  - Homocysteine can be converted back to methionine by methionine synthase, a reaction that occurs under methionine-sparing conditions and requires ***N*5-methyl-tetrahydrofolate** as methyl donor.

Utilization of Methionine in the Synthesis Of Cysteine



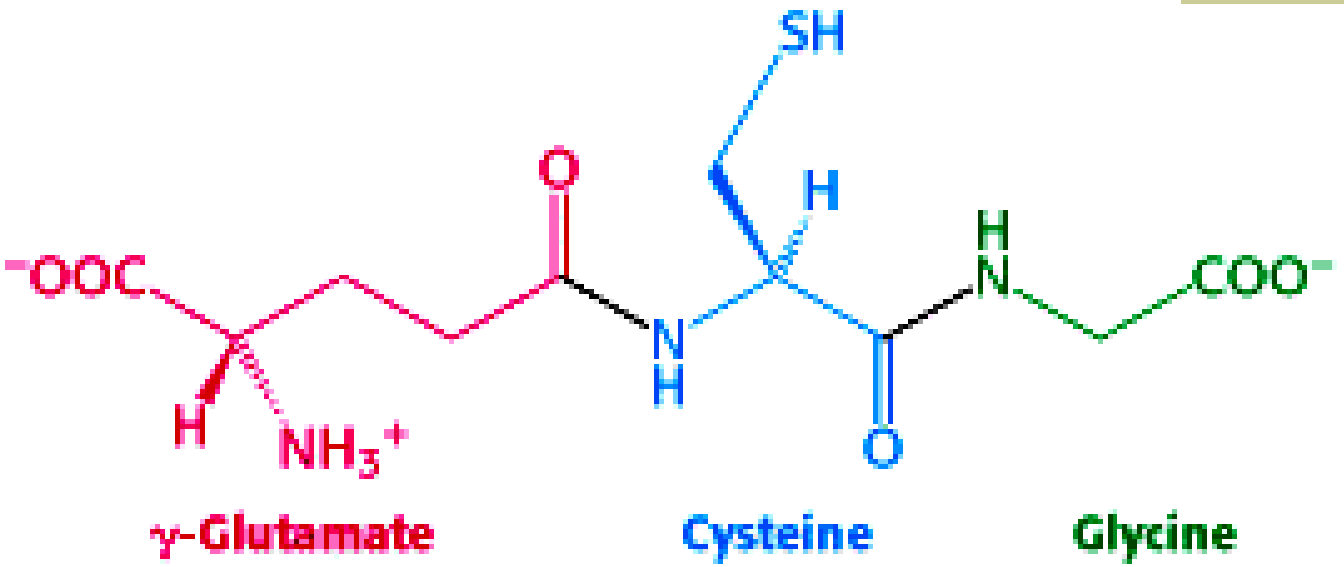
Transmethylation Reactions (cont'd)

- **Transmethylation** reactions employing SAM are extremely important, but in this case the role of S-adenosylmethionine in transmethylation is secondary to the production of homocysteine (essentially a by-product of transmethylase activity).
- In the production of SAM all phosphates of an ATP are lost: one as Pi and two as PPi.
- It is adenosine which is transferred to methionine and not AMP.

Cysteine Formation

- In cysteine synthesis, homocysteine condenses with serine to produce cystathionine, which is subsequently cleaved by cystathionase to produce cysteine and α-ketobutyrate.
- The sum of the latter two reactions is known as **trans-sulfuration**.
- Cysteine is used for protein synthesis and other body needs, while the α-ketobutyrate is decarboxylated and converted to propionyl-CoA.

Glutathione



- Cysteine readily oxidizes in air to form the disulfide cystine.
  - cells contain little free cystine because **glutathione** reverses the formation of cystine by a non-enzymatic reduction reaction.

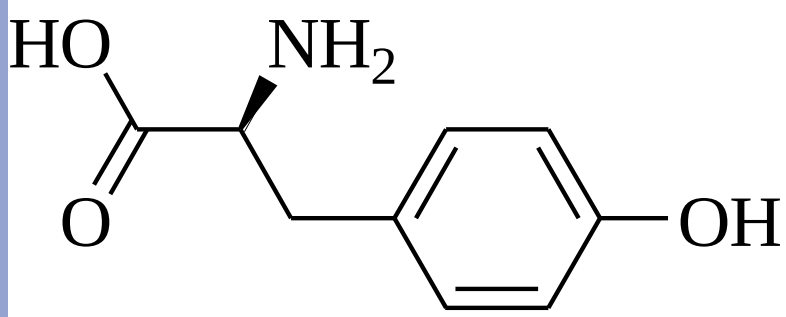
Homocystinuria

- The 2 key enzymes of this pathway, **cystathionine synthase** and **cystathionase (cystathionine lyase)**, both use *pyridoxal phosphate* as a cofactor, and both are under regulatory control.
- Cystathionase is under negative allosteric control by cysteine, as well, cysteine inhibits the expression of the cystathionine synthase gene.

Homocystinyria (cont'd)

- Genetic defects are known for both the synthase and the lyase. Missing or impaired cystathionine synthase leads to ***homocystinuria*** and is often associated with mental retardation, although the complete syndrome is multifaceted and many individuals with this disease are mentally normal.
- Some instances of genetic homocystinuria respond favorably to pyridoxine therapy, suggesting that in these cases the defect in cystathionine synthase is a decreased affinity for the cofactor.
- Missing or impaired cystathionase leads to excretion of cystathionine in the urine but does not have any other untoward effects.
- Rare cases are known in which cystathionase is defective and operates at a low level.
  - This genetic disease leads to methioninuria with no other consequences.

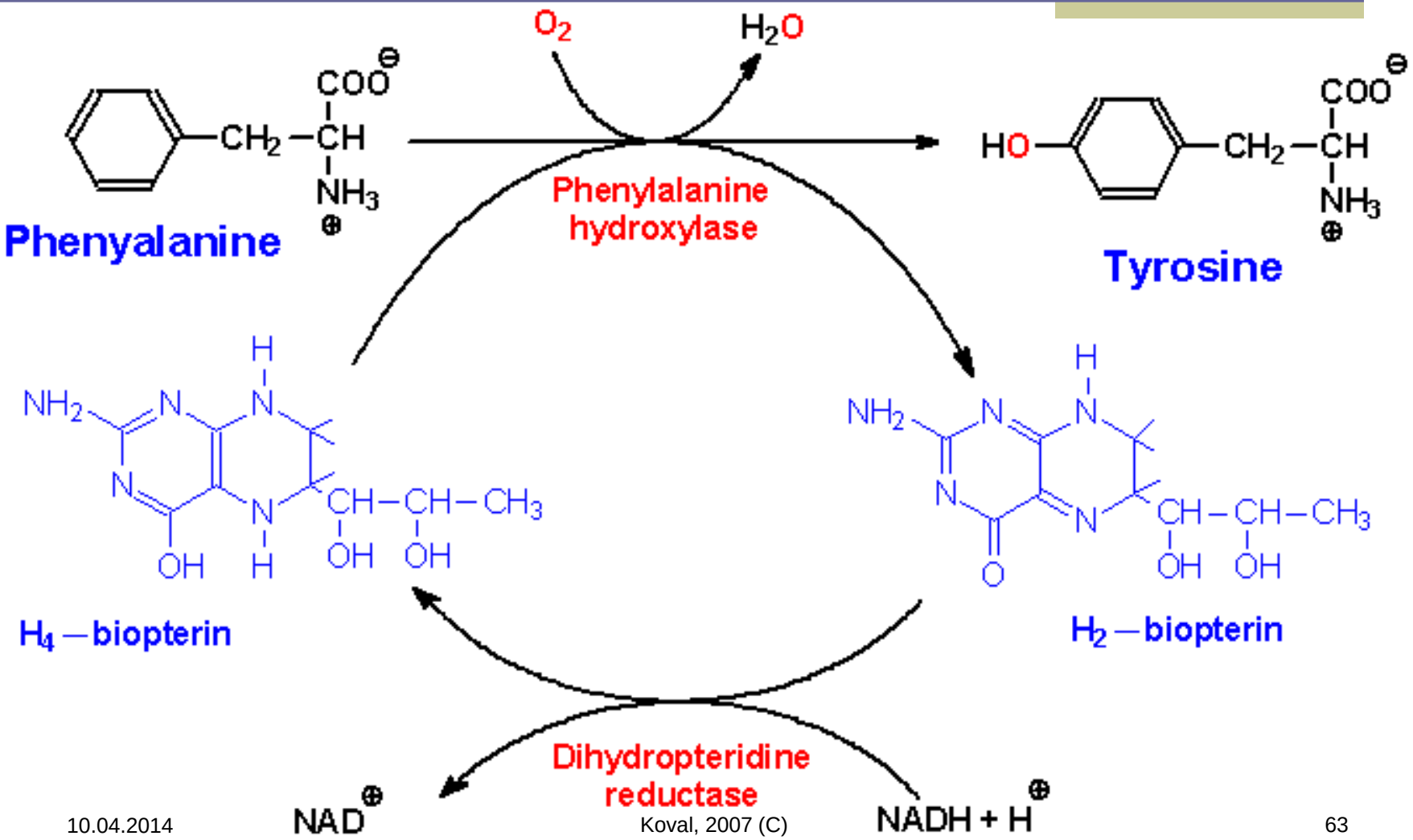
Tyrosine Biosynthesis



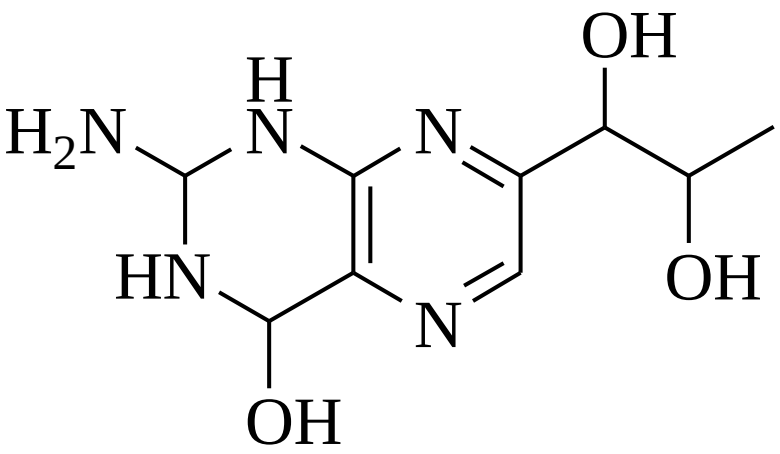
tyrosine

- Tyrosine is produced in cells by hydroxylating the essential amino acid **phenylalanine**.
  - This relationship is much like that between cysteine and methionine.
- Half of the phenylalanine required goes into the production of tyrosine;
  - if the diet is rich in tyrosine itself, the requirements for phenylalanine are reduced by about 50%.

Biosynthesis of tyrosine from phenylalanine



Phenylalanine Hydroxylase Functions



Tetrahydrobiopterin

- Phenylalanine hydroxylase is a **mixed-function oxygenase**: one atom of oxygen is incorporated into water and the other into the hydroxyl of tyrosine.
- The reductant is the tetrahydrofolate-related cofactor **tetrahydrobiopterin**, which is maintained in the reduced state by the NADH-dependent enzyme **dihydropteridine reductase (DHPR)**.

Disorders of phenylalanine metabolism

- Missing or deficient phenylalanine hydroxylase results in **hyperphenylalaninemia**.
  - **Hyperphenylalaninemia** is defined as a plasma phenylalanine concentration greater than 2 mg/dL (120 mM).
    - The most widely recognized hyperphenylalaninemia (and most severe) is the genetic disease known as **phenlyketonuria** (PKU).
    - Patients suffering from PKU have plasma phenylalanine levels >1000mM, whereas the non-PKU hyperphenylalaninemias exhibit levels of plasma phenylalanine <1000mM.

Untreated PKU

- Untreated PKU leads to ***severe mental retardation***.
  - The mental retardation is caused by the accumulation of phenylalanine, which becomes a major donor of amino groups in aminotransferase activity and depletes neural tissue of α-ketoglutarate.
  - This absence of α-ketoglutarate in the brain shuts down the TCA cycle and the associated production of aerobic energy, which is essential to normal brain development.

PKU Diagnostics

- The product of phenylalanine transamination, **phenylpyruvic acid**, is reduced to **phenylacetate** and **phenyllactate**, and all 3 compounds appear in the urine.
  - The presence of phenylacetate in the urine imparts a "mousy" odor.
  - If the problem is diagnosed early, the addition of tyrosine and restriction of phenylalanine from the diet can minimize the extent of mental retardation.

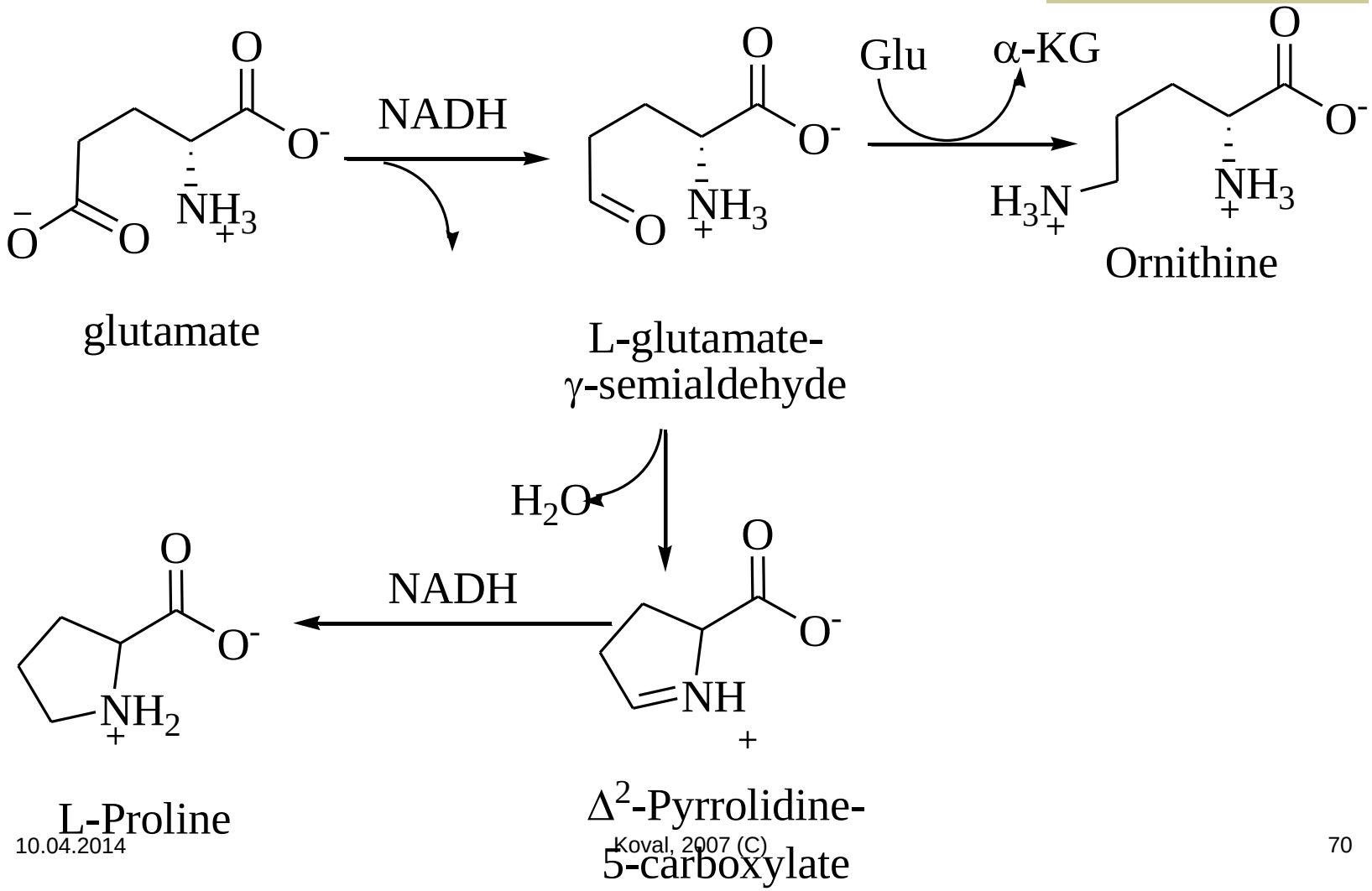
Tetrahydrobiopterin and the Effects of Missing or Defective DHPR

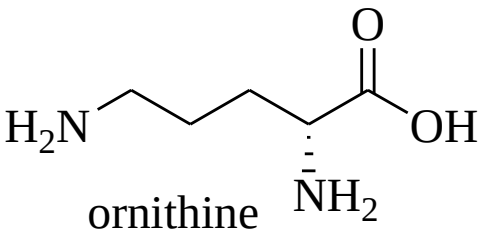
- Because of the requirement for tetrahydrobiopterin in the function of phenylalanine hydroxylase, deficiencies in DHPR can manifest with **hyperphenylalaninemia**.
  - Tetrahydrobiopterin is a cofactor in several other reactions (e.g. in the synthesis of the neurotransmitters as well as nitric oxide),
  - the effects of missing or defective DHPR cause even more severe neurological difficulties than those usually associated with PKU caused by deficient phenylalanine hydroxylase activity.

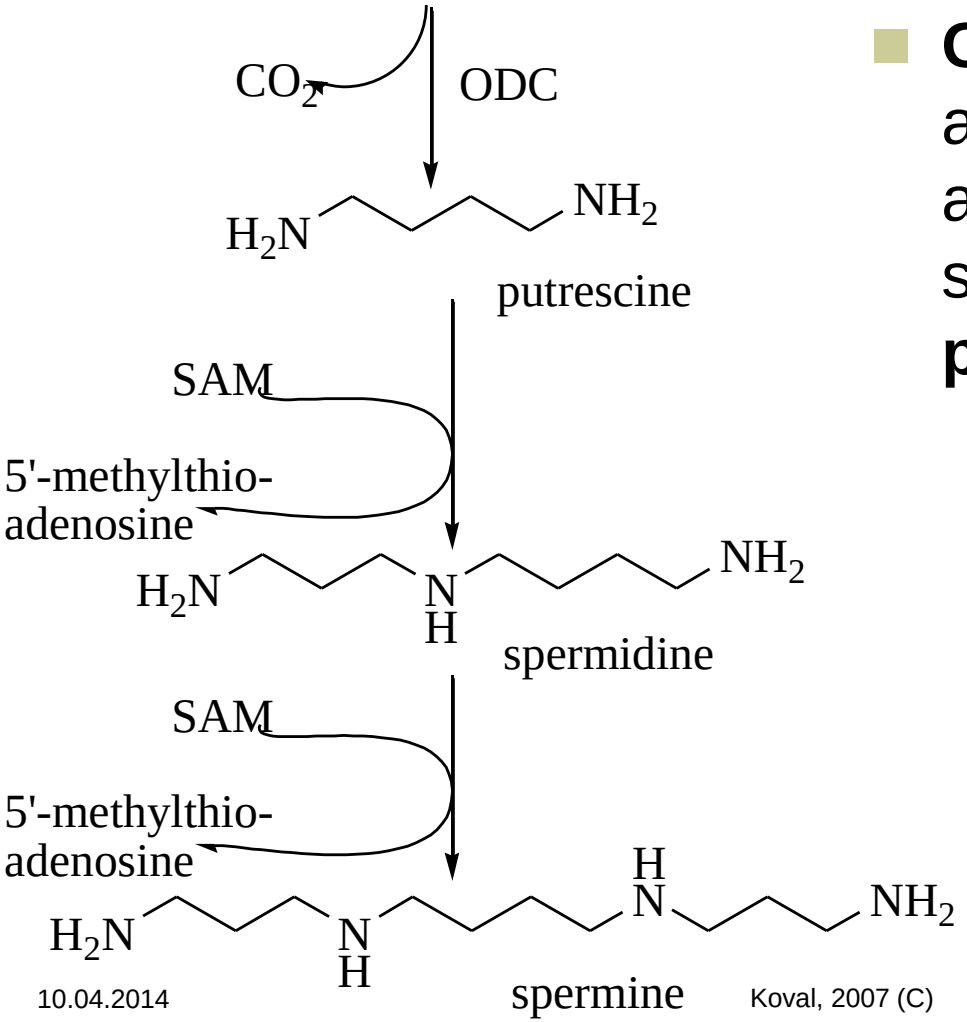
Ornithine and Proline Biosynthesis

- **Glutamate** is the precursor of both **proline** and **ornithine**, with **glutamate semialdehyde** being a branch point intermediate leading to one or the other of these 2 products.
- While **ornithine** is not one of the 20 amino acids used in protein synthesis, it plays a significant role as the acceptor of carbamoyl phosphate in the **urea cycle**.

Ornithine and Proline Biosynthesis (cont'd)





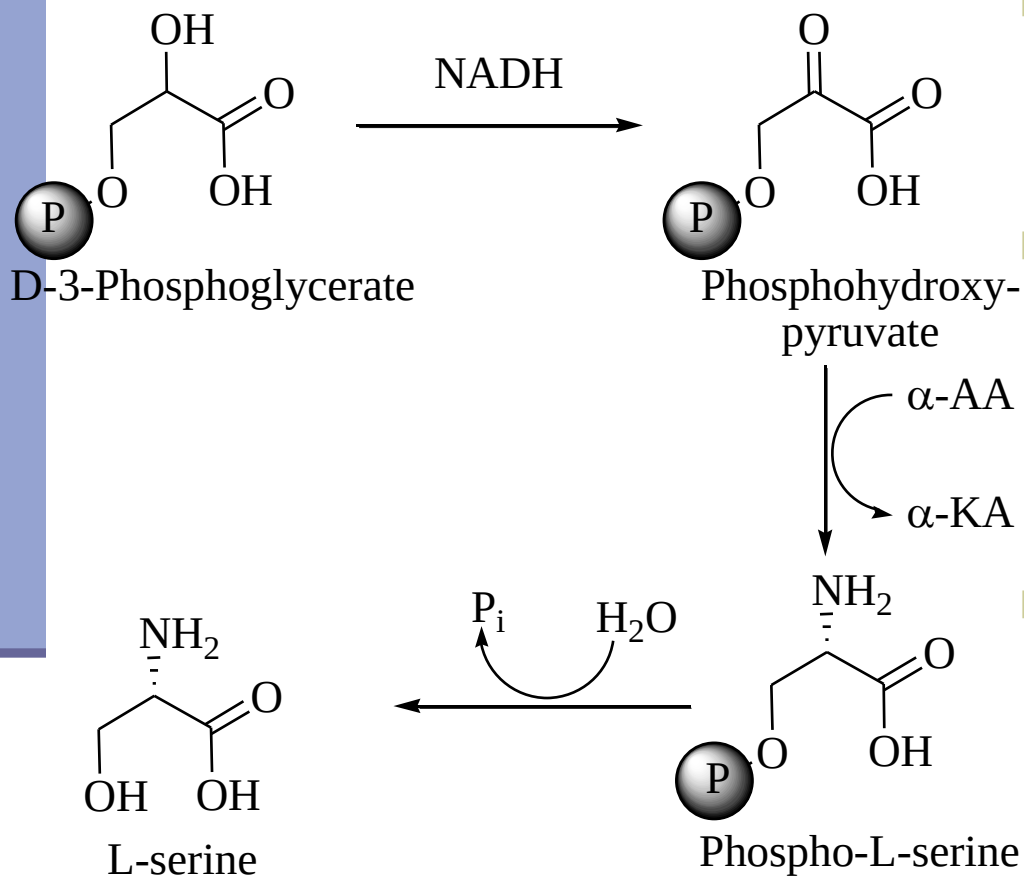


- **Ornithine** serves an additional important role as the precursor for the synthesis of the **polyamines**.
  - The production of ornithine from glutamate is important when dietary arginine, the other principal source of ornithine, is limited.

The Fate of Glutamate Semialdehyde

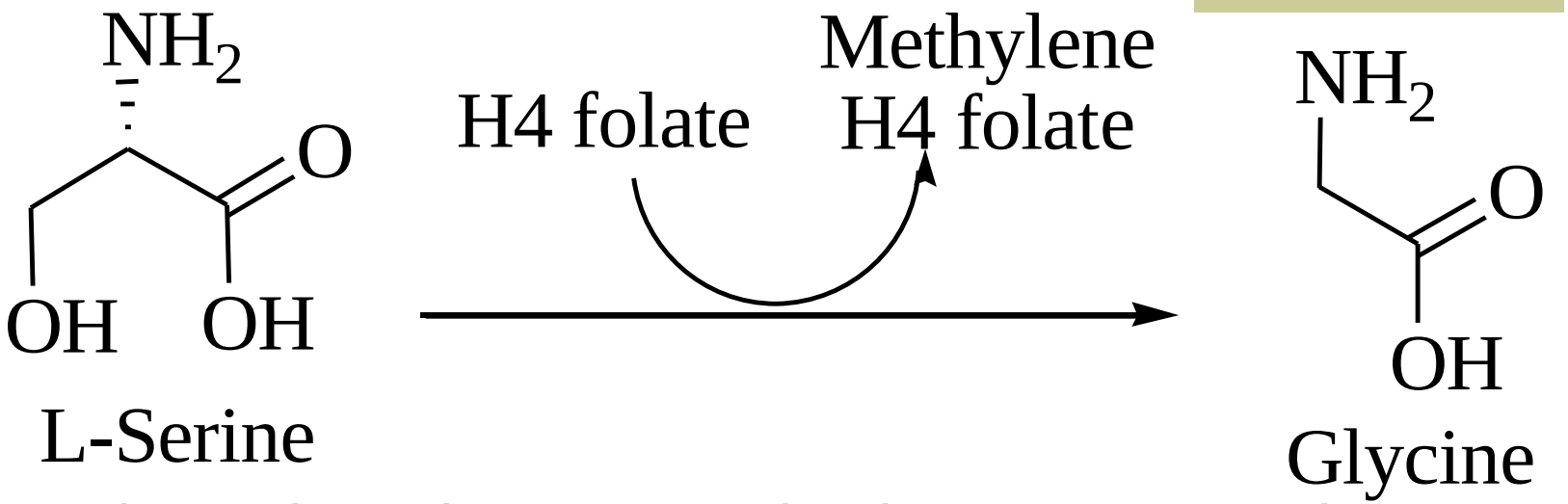
- The fate of **glutamate semialdehyde** depends on prevailing cellular conditions.
  - **Ornithine production** occurs from the semialdehyde via a simple glutamate-dependent transamination, producing **ornithine**.
  - At high arginine concentrations the ornithine contributed from the **urea cycle** plus that from glutamate semialdehyde inhibit the aminotransferase reaction. Semialdehyde is accumulated.
- The semialdehyde cyclizes spontaneously to Δ^2-pyrrolidine-5-carboxylate which is then reduced to **proline** by an **NADPH-dependent reductase**.

Serine Biosynthesis



- The main pathway to serine starts with the glycolytic intermediate 3-phosphoglycerate.
- An NADH-linked dehydrogenase converts 3-phosphoglycerate into a keto acid, 3-phosphopyruvate, suitable for subsequent transamination.
- Aminotransferase activity with glutamate as a donor produces 3-phosphoserine, which is converted to serine by phosphoserine phosphatase.

Glycine Biosynthesis

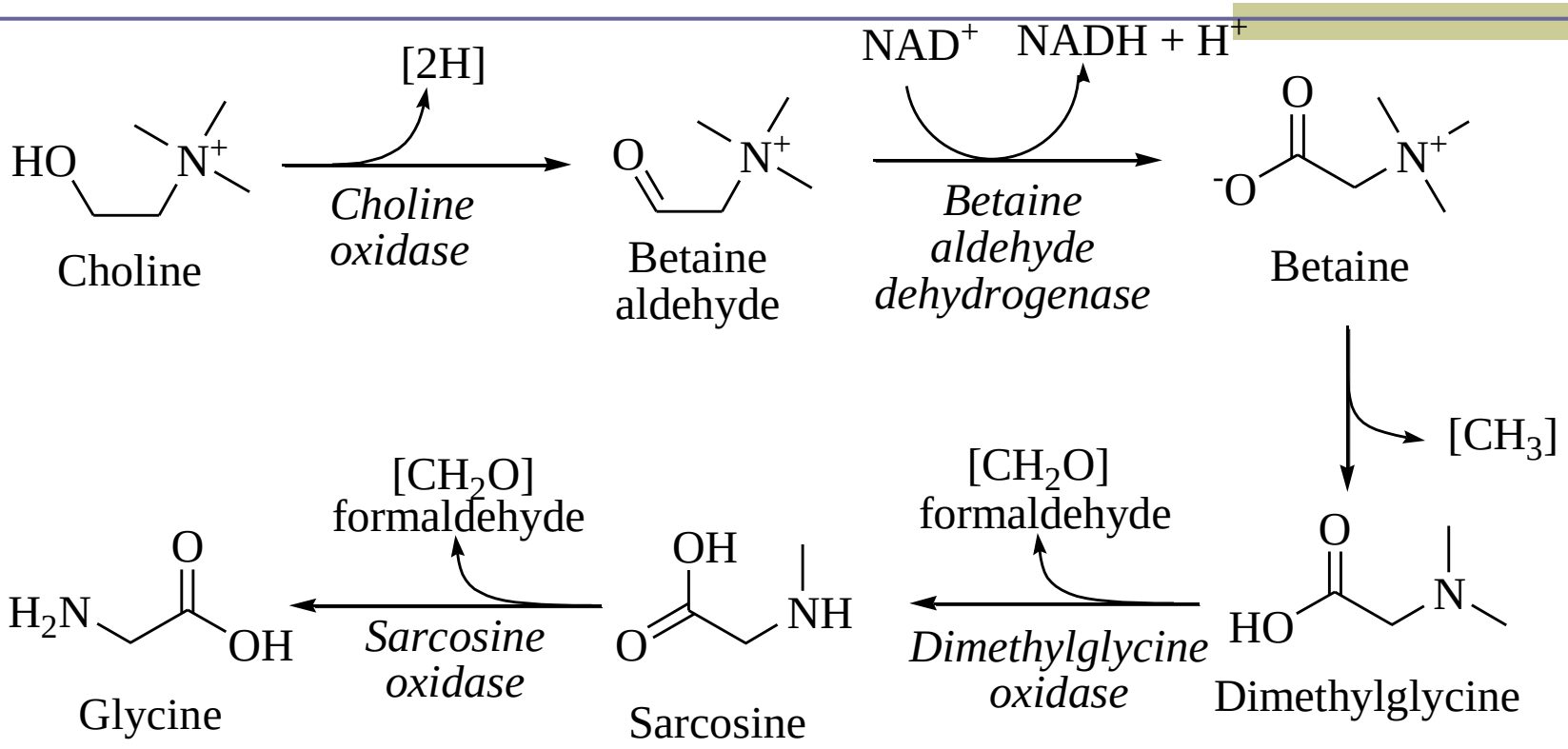


L-Serine

Glycine

- The main pathway to glycine is a 1-step reaction catalyzed by **serine hydroxymethyl-transferase**.
- This reaction involves the transfer of the hydroxymethyl group from serine to the cofactor tetrahydrofolate (THF), producing glycine and N^5,N^{10}-methylene-THF.

Glycine Formation from Choline



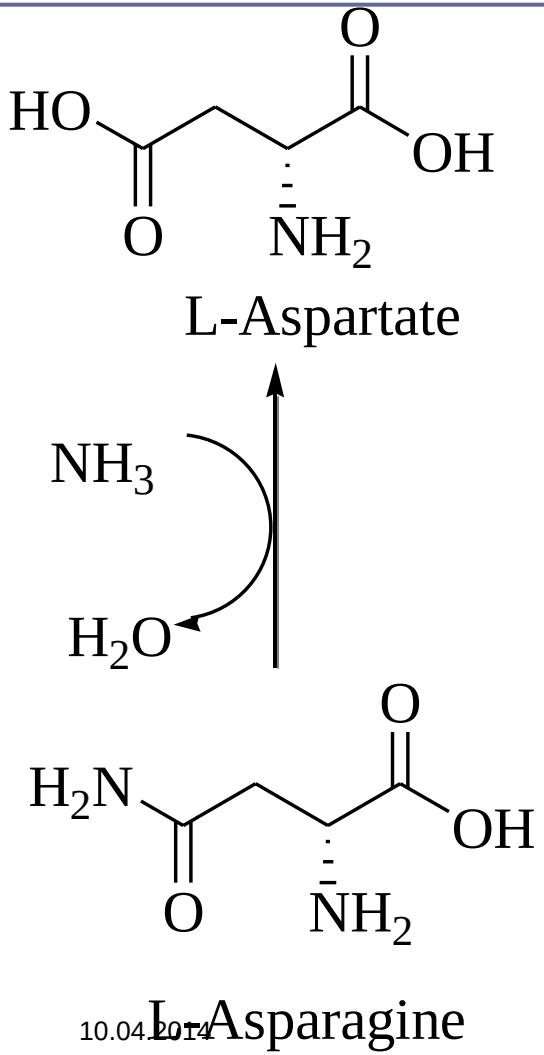
Aspartate/Asparagine and Glutamate/Glutamine Biosynthesis

- Glutamate is synthesized by the **reductive amination** of α-ketoglutarate catalyzed by **glutamate dehydrogenase**;

$$\alpha\text{-KG} + NH_3 + NADPH{+}H^+ \rightarrow Glu + NADP^+ + H_2O$$

  - it is thus a nitrogen-fixing reaction.
- In addition, glutamate arises by **aminotransferase reactions**, with the amino nitrogen being donated by a number of different amino acids.
- Thus, **glutamate is a general collector of amino nitrogen**.

Aspartate/Asparagine and Glutamate/Glutamine Biosynthesis (cont'd)

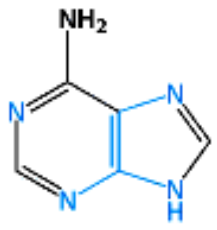


- Aspartate is formed in a transamintion reaction catalyzed by **aspartate transaminase, AST**.
  - This reaction uses the aspartate α-keto acid analog, **oxaloacetate**, and **glutamate** as the amino donor.
- Aspartate can also be formed by deamination of asparagine catalyzed by **asparaginase**.

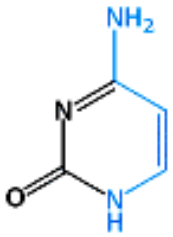
Aspartate/Asparagine and Glutamate/Glutamine Biosynthesis (cont'd)

- **Asparagine synthetase** and **glutamine synthetase**, catalyze the production of **asparagine** and **glutamine** from their respective α-amino acids.
- Glutamine is produced from glutamate by the direct incorporation of ammonia; and this can be considered **another nitrogen fixing reaction**.
  - Asparagine, however, is formed by an **amidotransferase** reaction.

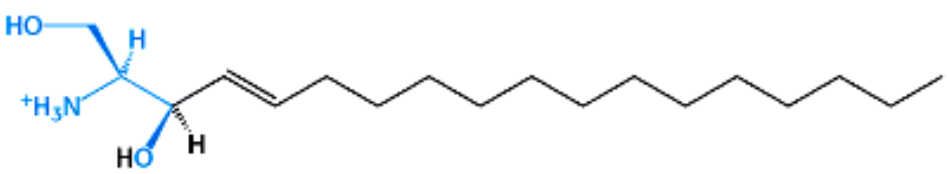
Selected Biomolecules Derived from Amino Acids



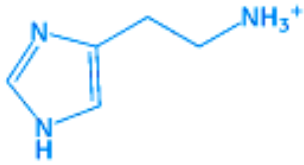
Adenine



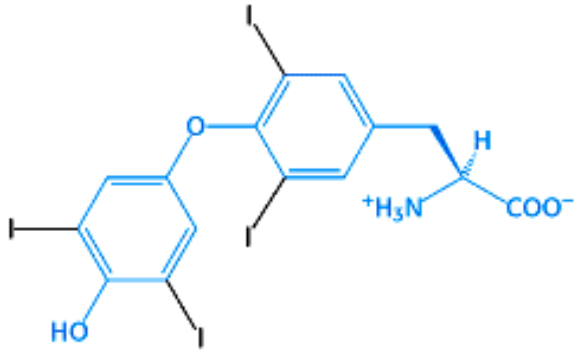
Cytosine



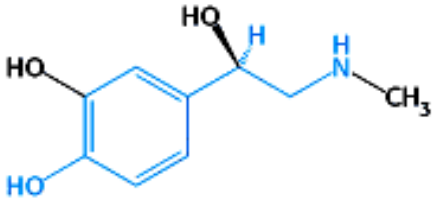
Sphingosine



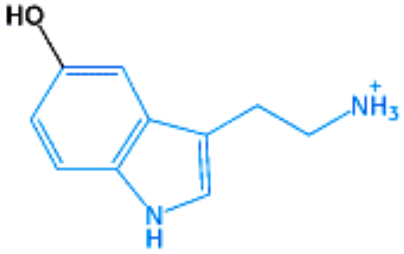
Histamine



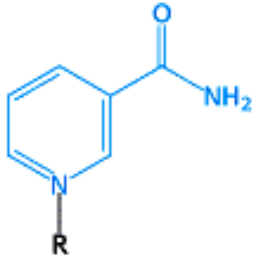
Thyroxine
(Tetraiodothyronine)



Epinephrine



Serotonin



Nicotinamide unit of NAD^+


Thank you
for your attention