Amino acid Metabolism (Biosynthesis and Catabolism)

All tissues have some capability for synthesis of the non-essential amino acids, amino acid remodelling, and conversion of non-amino acid carbon skeletons into amino acids and other derivatives that contain nitrogen. However, the liver is the major site of nitrogen metabolism in the body. In times of dietary surplus, the potentially toxic nitrogen of amino acids is eliminated via **trans-amination, deamination, and urea formation;** the carbon skeletons are generally conserved as carbohydrate, via gluconeogenesis, or as fatty acid via fatty acid synthesis pathways. In this respect amino acids fall into three categories: **glucogenic**, **ketogenic**, or both **glucogenic and ketogenic**. Glucogenic amino acids are those that give rise to a net production of pyruvate or TCA cycle intermediates, such as α-ketoglutarate or oxaloacetate, all of which are precursors to glucose via gluconeogenesis. All amino acids except lysine and leucine are at least partly glucogenic. Lysine and leucine are the only amino acids that are solely ketogenic, giving rise only to acetylCoA or acetoacetylCoA, neither of which can bring about net glucose production.

A small group of amino acids comprised of isoleucine, phenylalanine, threonine, tryptophan, and tyrosine give rise to both glucose and fatty acid precursors and are thus characterized as being glucogenic and ketogenic. Finally, it should be recognized that amino acids have a third possible fate. During times of starvation the reduced carbon skeleton is used for energy production, with the result that it is oxidized to CO_2 and H_2O.

Overview of amino acids metabolism:

a) Amino acids serve as substrates for the synthesis of protein,
b) Amino acids provide nitrogen for the synthesis of other nitrogencontainingcompounds,
c) Amino acids are catabolized as fuels.

Classification of amino acids:

1. Chemical classification,

a. According to the **chemistry** of the side chains.
b. According to **polarity** of side chains.

2. Nutritional classification,
• Essential
• Non-essential

3. Metabolic classification,
• Glucogenic,
• Ketogenic
• Both glucogenic and ketogenic

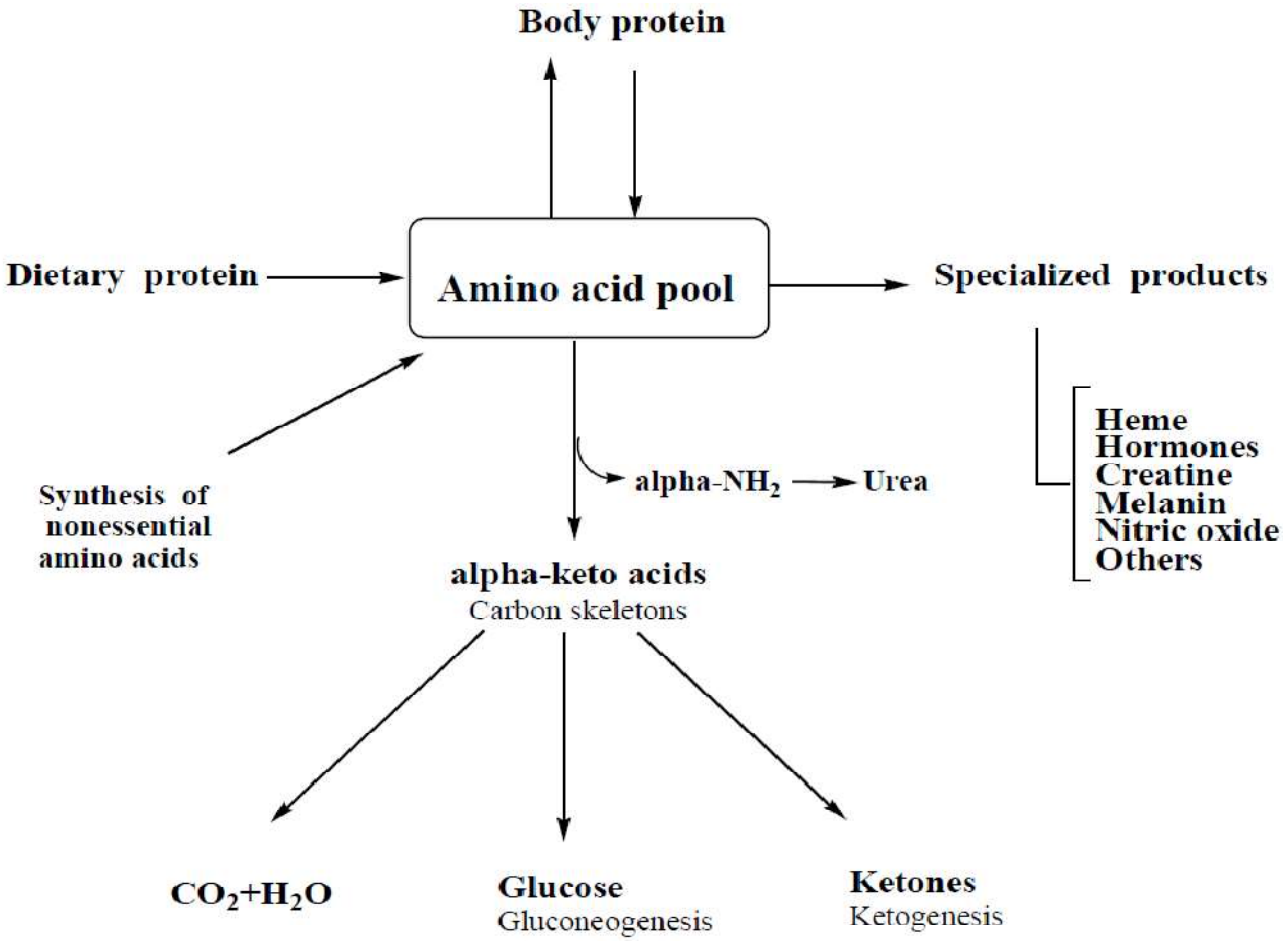


Figure: Overview of amino acids metabolism

Essential & Nonessential Amino Acids

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

*The amino acids **arginine**, **methionine** and **phenylalanine** are considered essential for reasons not directly related to lack of synthesis. Arginine is synthesized by mammalian cells but at a rate that is insufficient to meet the growth needs of the body and the majority that is synthesized is cleaved to form urea. Methionine is required in large amounts to produce cysteine if the latter amino acid is not adequately supplied in the diet. Similarly, phenyalanine is needed in large amounts to form tyrosine if the latter is not adequately supplied in the diet.

Glucogenic and ketogenic amino acids:

Glucogenic	**Ketogenic**	**Both Gluco and Ketogenic**
Alanine Asparagine Cysteine Glutamine Histidine Proline Threonine Arginine Aspartate Glutamate Glycine Methionine Serine Valine	Leucine Lysine	Isoleucine Phenylalanine Trptophan Tyrosine

Biosynthesis of Nonessential Amino Acids:

Humans do not have the ability to synthesize 10 of the necessary 20amino acids and must obtain them from the diet. These 10 are termed thenutritionally essential amino acids. The 10 nonessential amino acids areformed by 3 general mechanisms:

a) Transamination:
• **Alanine,** can be synthesized by transamination of thecorresponding α -ketoacid,pyruvate.

• **glutamate,** can be synthesized by transamination of thecorresponding α -keto acid, α -ketoglutarate.

• **aspartate**can be synthesized by transamination of thecorresponding α -keto acid, oxaloacetate

• **Serine** is synthesized by the transamination and dephosphorylationof 3-phosphogylcerate, an intermediate of glycolysis.

b) Assimilation of free ammonia:

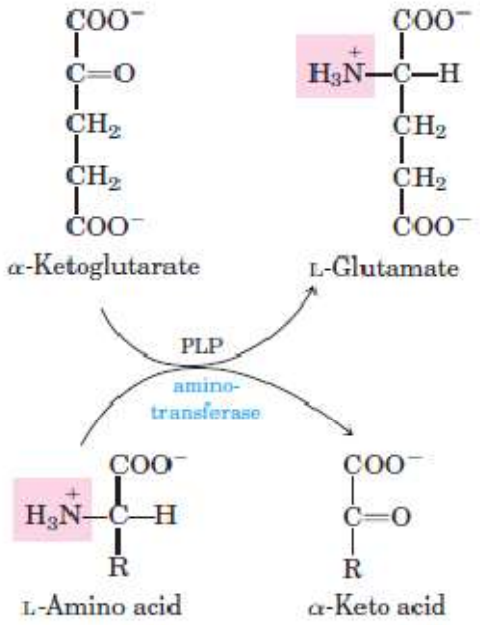
• **Glutamate:** Formation of glutamate from ammonia and α-ketoglutarate is catalyzed by glutamate dehydrogenase. Thisreaction is reversible and plays a role in both synthesis and breakdown of glutamate. Both NADPH and NADH can serve asthe source of reducing equivalents used in this reaction.

• **Glutamine:** Glutamine synthetasecatalyzes the ATP-dependentformation of glutamine, using glutamate and ammonia assubstrates.

c) Modification of the carbon skeletons of existing amino acids.

• **Cysteine**: Cysteine contains atoms donated by both methionine andserine.
• **Glycine:** Serine is also converted to glycine by the removal of itshydroxymethyl group**.**
• **Tyrosine:** Phenylalanine is hydroxylated to form tyrosine.
• **Proline:** Glutamate is reduced and cyclized to form proline.
• **Asparagine:** Asparagine is synthesized by the transfer to the amidegroup of glutamine to the ß-carboxyl group of aspartate

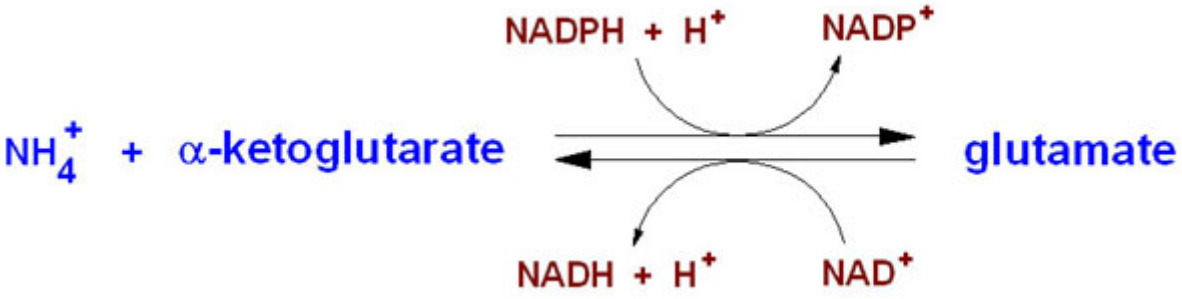
Pyridoxal Phosphate (PLP):
In a transamination reaction, i.e. reactions involving the transfer ofthe α -amino group of an amino acid to the α -carbon of a keto acid,thereby forming a new amino acid and a new keto acid.Pyridoxal phosphate (**vitamin B6**) acts as an intermediate carrier ofthe amino group that is being transferred.



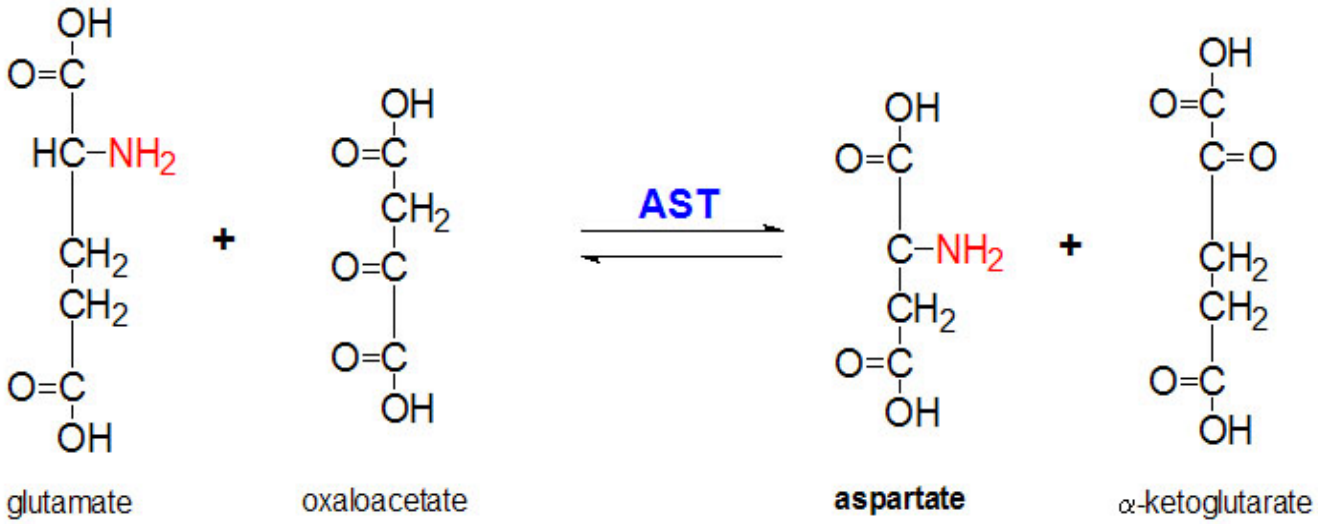
Enzyme-catalyzedtransaminations. In many aminotransferasereactions,α-ketoglutarate is the amino group acceptor. Allaminotransferases have pyridoxal phosphate (PLP) as cofactor. Althoughthe reaction is shown here in the direction of transfer of theamino group to α-ketoglutarate, it is readily reversible.

Glutamate and Aspartate biosynthesis

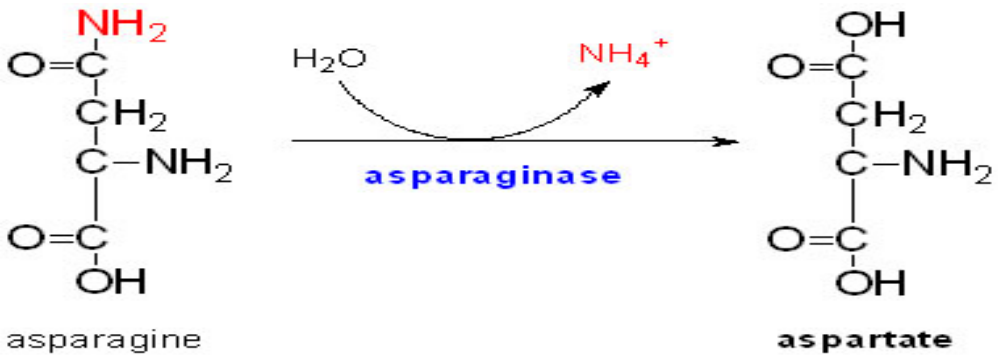
Glutamate is synthesized from its' widely distributed α-keto acid precursor by a simple 1-step transamination reaction catalyzed by glutamate dehydrogenase.Glutamate dehydrogenase reaction plays a central role in overall nitrogen homeostasis.



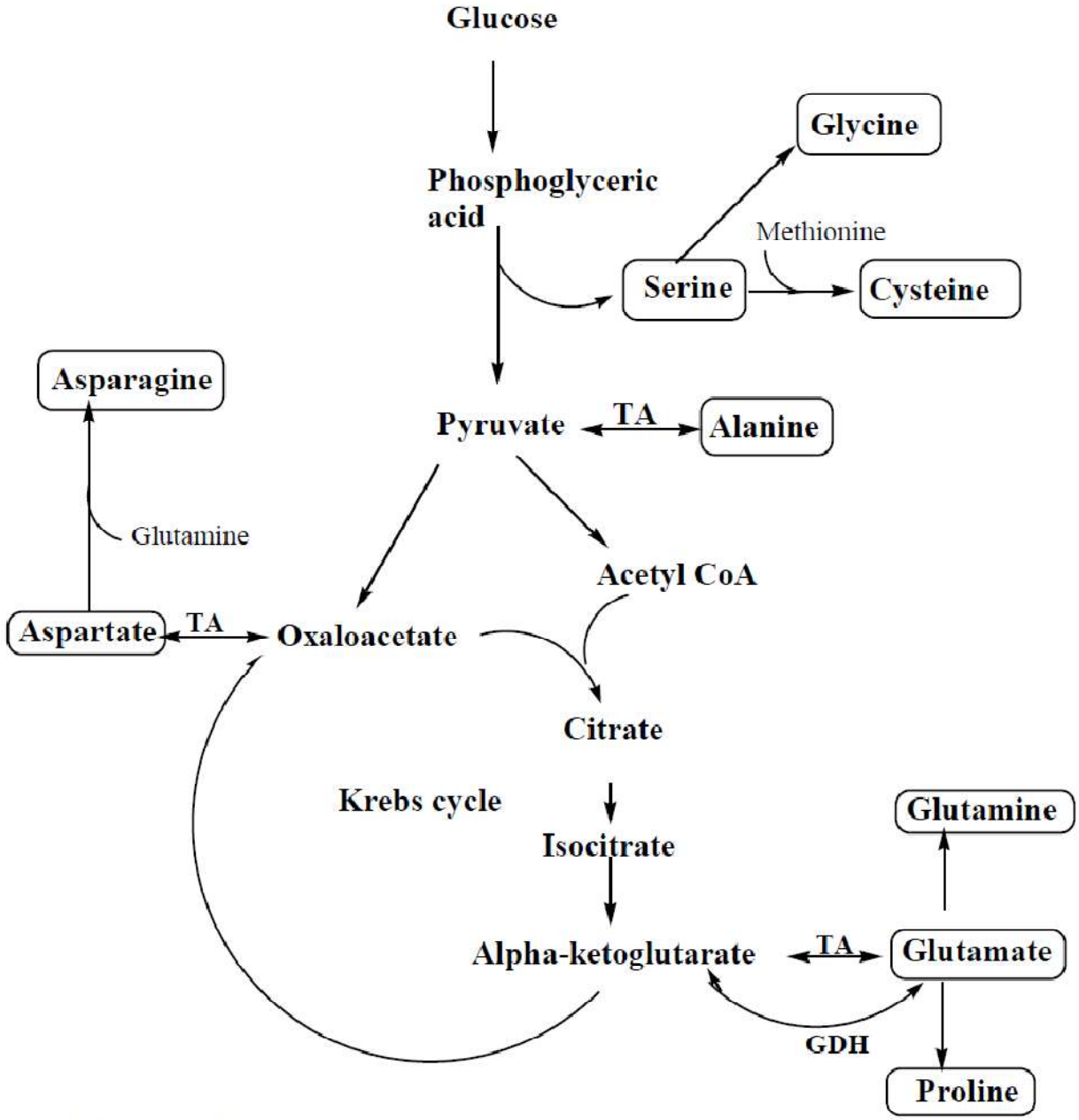
Like glutamate, aspartate is synthesized by a simple 1-step transamination reaction catalyzed by aspartate aminotransferase, AST (formerly referred to as serum glutamate-oxalate transaminase, SGOT).



Aspartate can also be derived from asparagine (whose synthesis is outlined below) through the action of asparaginase. The importance of aspartate as a precursor of ornithine for the urea cycle.



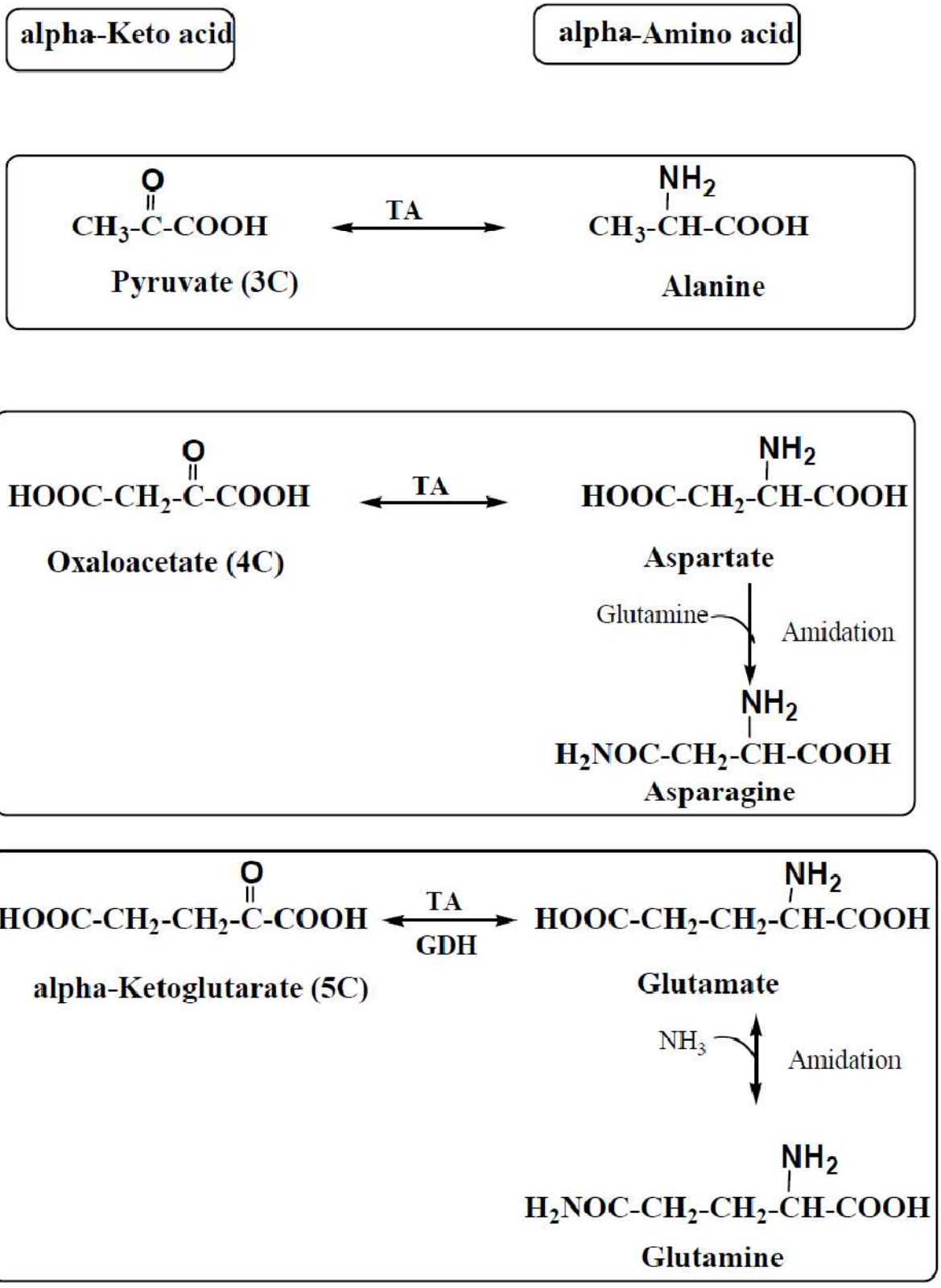
General pathways involved in biosynthesis of non-essential amino acids:



TA= transamination
GDH=glutamate dehdrogenase

Phenylalanine (essential) —hydroxylation→ **Tyrosine**

Some examples of Transamination reactions:



Amino Acid Catabolism (Mainly Urea Cycle):

Fate of amino groups:

Step 1: transamination with α -ketoglutarate to form glutamate andnew α -keto acid.
Step 2: glutamate is deaminated through oxidative process.
Step 3: formation of urea through urea cycle.

Synthesis of urea: (urea cycle) (Refer the class notes, [(01-11-11)]
The amino groups for urea synthesis are collected in the form of**aspartate** and **ammonia**.

Site: Partly in mitochondria and partly in cytosol (Liver).
Reactions:
1) Ammonia enters the cycle by combining with CO2 and ATP to formcarbamoyl phosphate
2) Carbamoyl phosphate combines with ornithine to produce citrulline.
3) Aspartate, carrying the second nitrogen atom of urea, enters the cycleby condensing with citrulline to form argininosuccinate.
4) Argininosuccinate is cleaved to fumarate and arginine.
5) Arginine is further hydrolyzed to yield urea and regenerate theornithine needed for the next round of the cycle.

Regulation of urea cycle:
a) Glutamate dehydrogenase, which provides the bulk of the ammoniafor urea synthesis, is activated by ADP and GDP andinhibited by ATP and GTP.
b) The activity of carbamoyl phosphate synthase I is activated by Nacetylglutamate.

Energy of urea Cycle:
3 ATPs + Ammonia + Aspartate + Co2 →urea + fumurate + 2ADP + 2 Pi + AMP + PPi.

Clinical significance of blood urea:
• Elevated in renal insufficiency.
• Decreased in hepatic failure.

Why is ammonia toxic?

α-ketoglutarate + NH3 + NADPH $\overset{GDH}{\rightleftharpoons}$ glutamate + NADP+

High ammonia depletes the TCA cycle of α-ketoglutarate→ low ATP →COMA (a symptom of high ammonia levels).

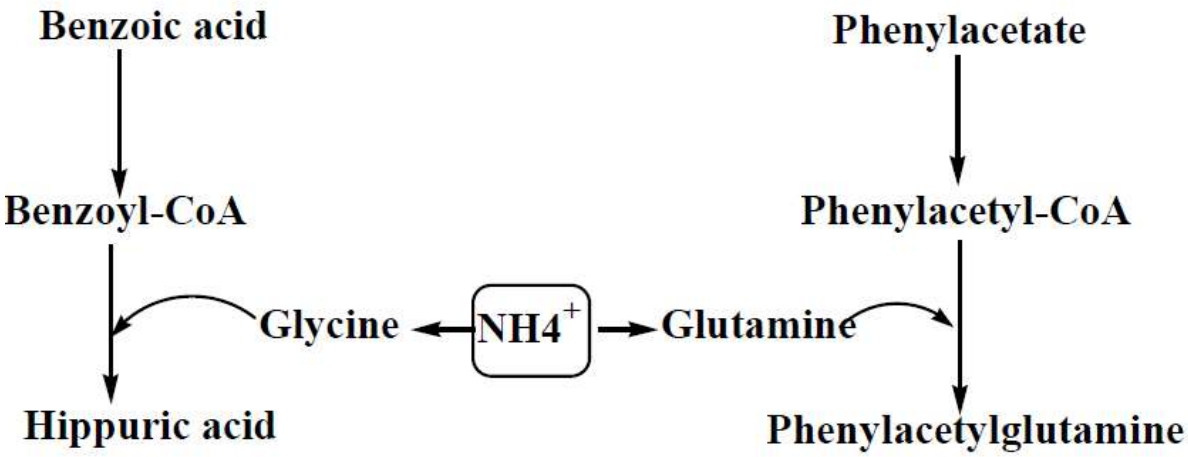
Hyperammonemia:

1. Acquired Hyperammonemia. e.g. liver disease.
2. Hereditary Hyperammonemia. e.g. genetic deficiencies of Ureacycle enzymes.e.g.

a. Congenital hyperammonmia Type I:carbamoyl phosphate synthetase deficiency
b. Congenital hyperammonmia Type II:Ornithine transcarbamoylase deficiency

Treatment:
1. Limit protein intake.
2. Give carbon skeletons to provide the essential amino acids.

3. Use a trapping molecule (sodium benzoate, sodium phenylacetate)that bind covalently to amino acids and produce nitrogencontaining molecules that are excreted.



Fate of carbon skeletons:
Following removal of their amino groups, the **carbon skeletons** ofall amino acids aredegraded to intermediates already encountered in fuelmetabolism:
1. Acetyl-CoA.
2. Pyruvate.
3. Intermediates of the citric acid cycle:
• α -ketoglutarate.
• Oxaloacetate.
• Succinyl-CoA.
• Fumarate.

Genetic Defects of Amino Acid Metabolism:

Phenylketonuria: (PKU)
.
• Deficiency of **phenylalanine hydroxylase**.
• Tyrosine is an essential amino acid
• Phenylpyruvate, phenylacetate, and phenyllactate are produced ingreater amounts and are spilled in the urine.
• The major manifestation of the disease is mental retardation.

Homocystinuria:

• Deficiency of **cystathionine synthase**, an enzyme that forms partof the pathway for cysteine synthesis, results in a disorder knownas homocystinuria.

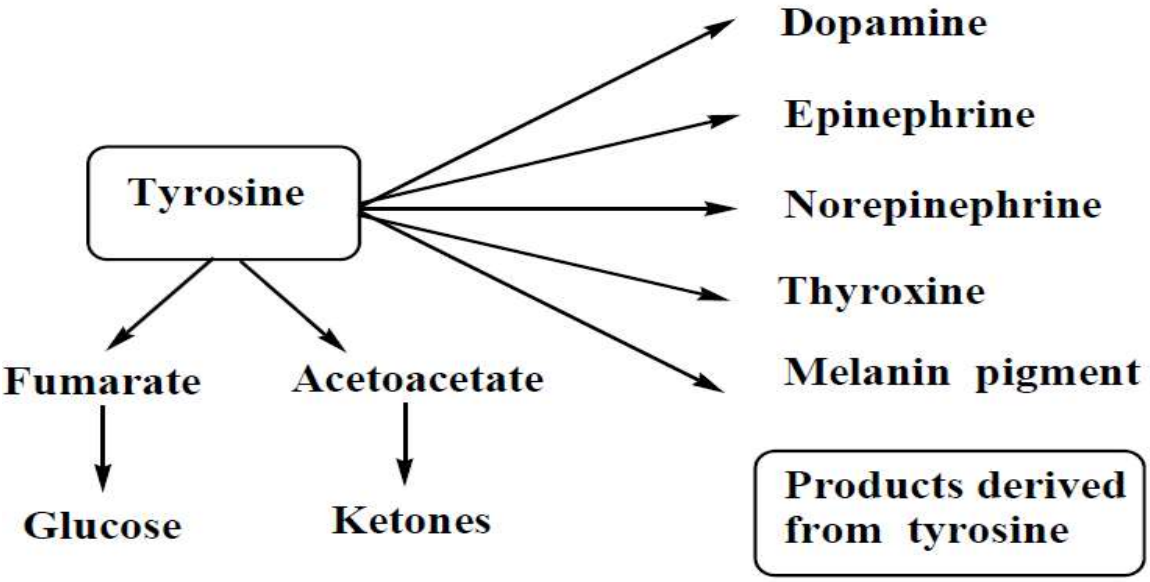
• High urinary levels of homocysteine, a substrate of the impairedenzyme.
• Two forms have been described, one of which can be treated byhigh doses of vitamin B6. This form of the disorder is due to thereduced affinity ofcystathionine synthase for its coenzyme,pyridoxal phosphate.
• The other form is treated by limiting intake of methionine and byproviding cysteine in the diet.

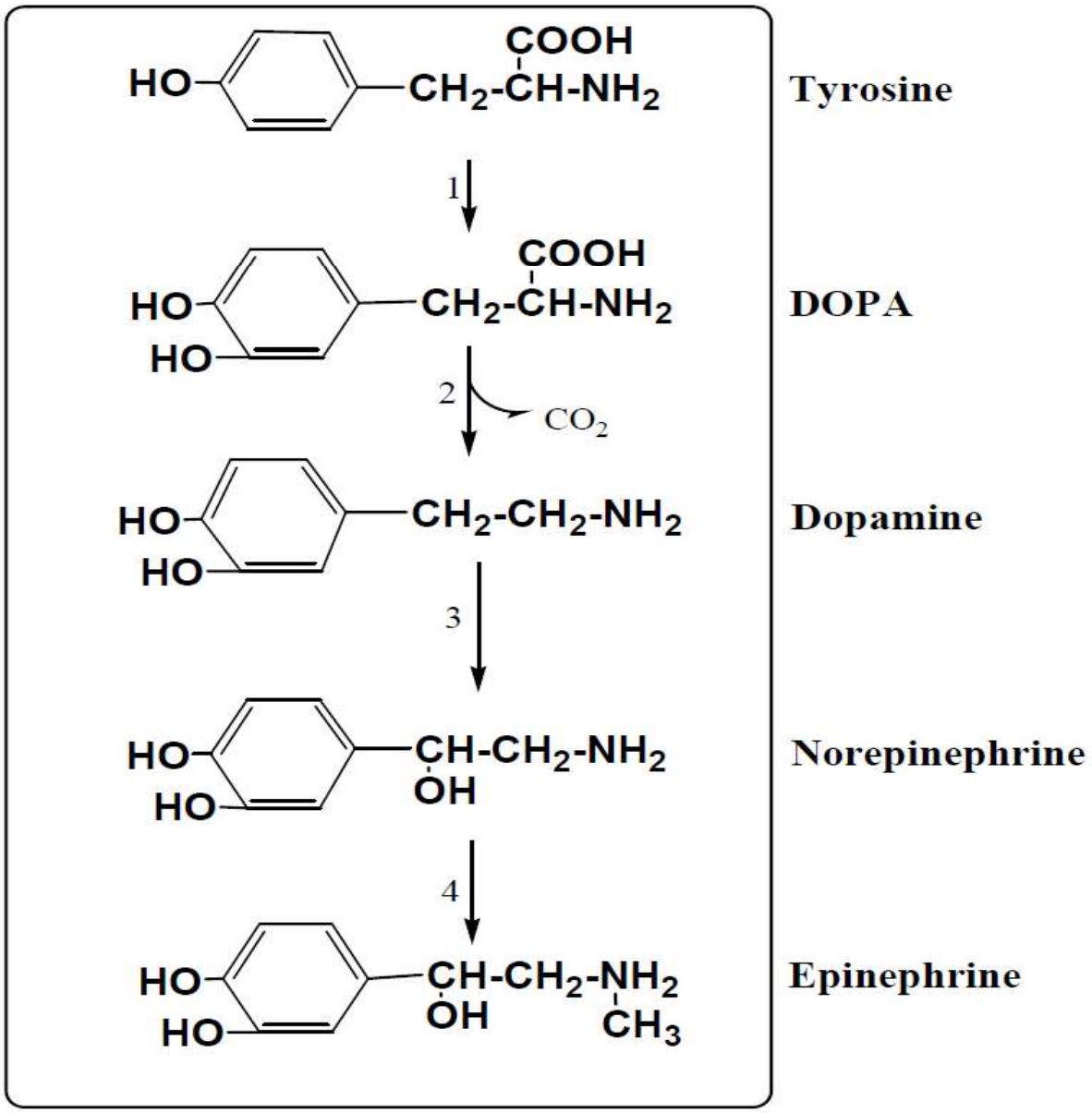
Branched-chain ketonuria(maple syrup urine disease):

• Defect in **α keto acid decarboxylase(α keto acid dehydrogenase)** an enzyme involved in thecatabolism of leucine, valine, and isoleucine.
• The disease is manifested by severe brain damage.
• Urine smells like maple syrup.
• Few infants survive beyond the first year of life.

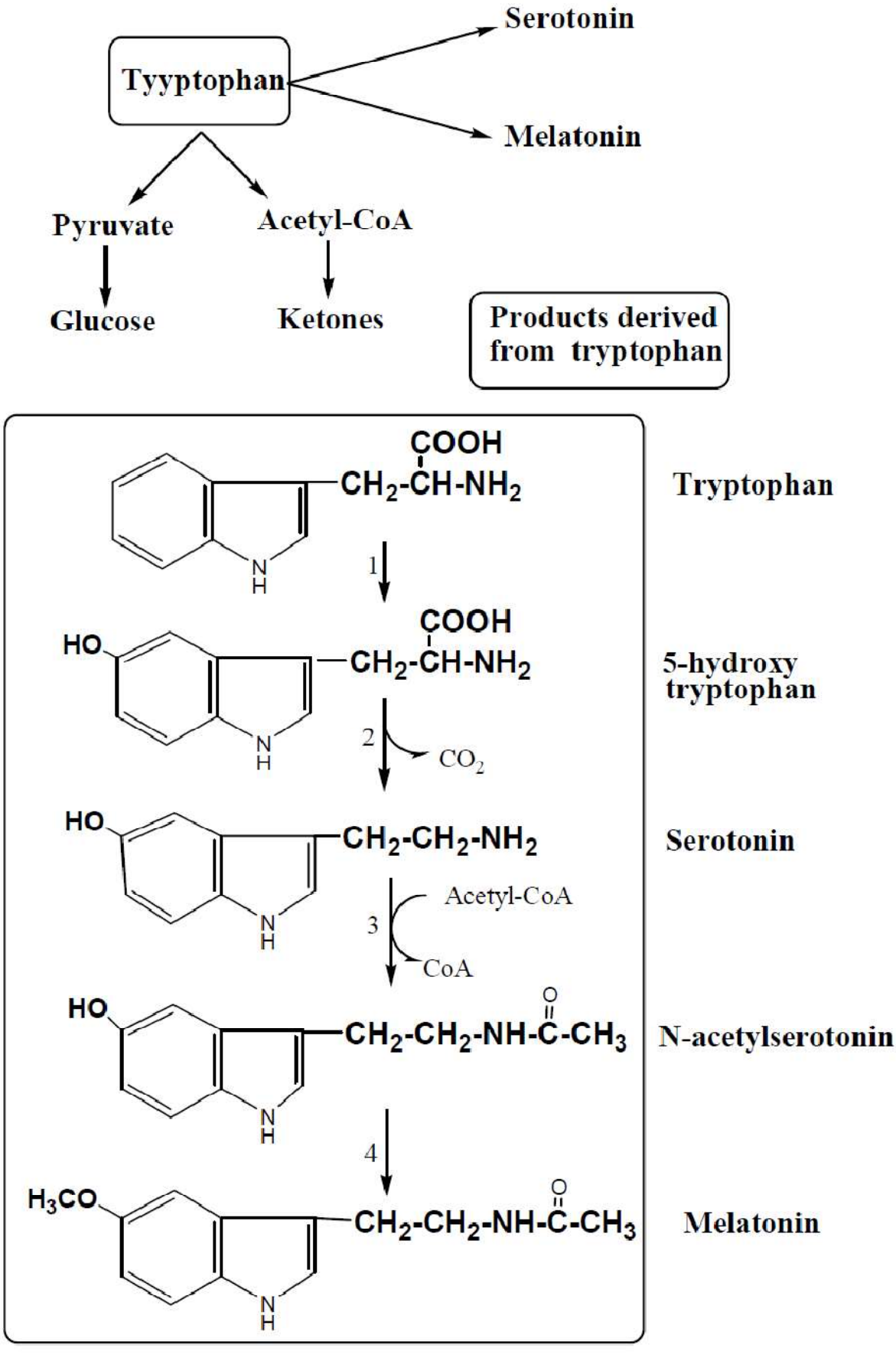
Biosynthesis of some important molecules:
• Catecholamines.
• Serotonin and melatonin.
• Histamine.
• Gamma-aminobutyric acid (GABA).
• Nitric oxide.
• Glutathione.

Biosynthesis of Catecholamines:

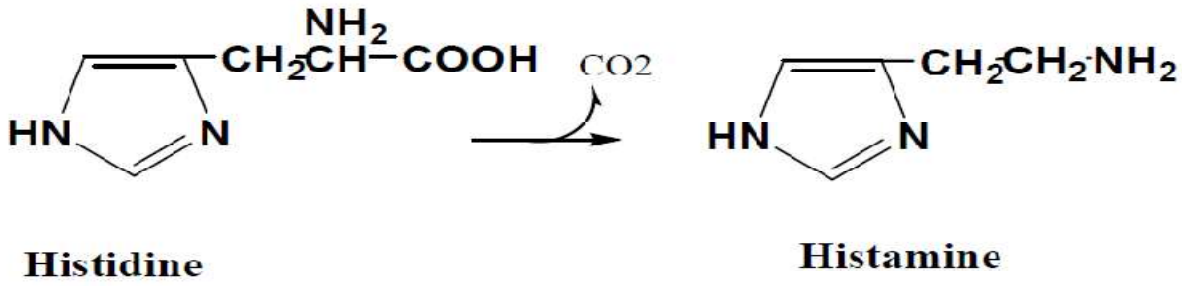

Tyrosine
Dopamine
Epinephrine
Norepinephrine
Thyroxine
Melanin pigment
Fumarate
Acetoacetate
Glucose
Ketones
Products derived from tyrosine


Tyrosine
DOPA
Dopamine
Norepinephrine
Epinephrine
1
2
3
4
CO2

Biosynthesis of Serotonin and Melatonin:



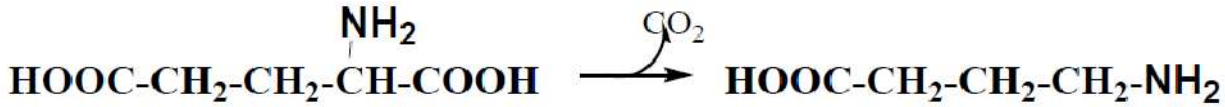
Biosynthesis of Histamine:



Histidine **Histamine**

• Synthesized and released by mast cells
• Mediator of allergic response: (H1 receptors)
H1 blockers: Diphenhydramine (Benadryl)
Loratidine (Claritin)
• Stimulates secretion of gastric acid (H2 receptors)
H2 blockers: Cimetidine (Tagamet)
Ranitidine (Zantac)

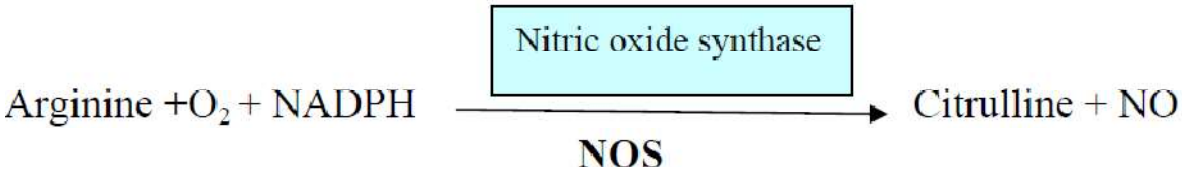
Biosynthesis of γ-aminobutyric acid:



Glutamate **GABA**

• GABA is an important inhibitory neurotransmitter in the brain

Nitric Oxide (NO)

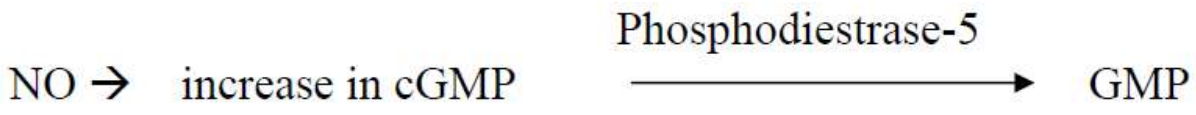


Functions of NO:
• Neurotranmitter
• Prevents platelet aggregation.
• Bactericidal.
• Relaxes smooth musle by activation of guanylylcyclase → cGMP→relaxation.

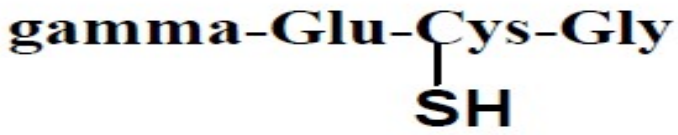
• Nitroglycerin →Glycerin + NO

• Sildenafil (Viagra): inhibits phosphodiestrase-5 invascular smooth muscle →increase in cGMP.

Phosphodiestrase-5



Glutathione (GSH)

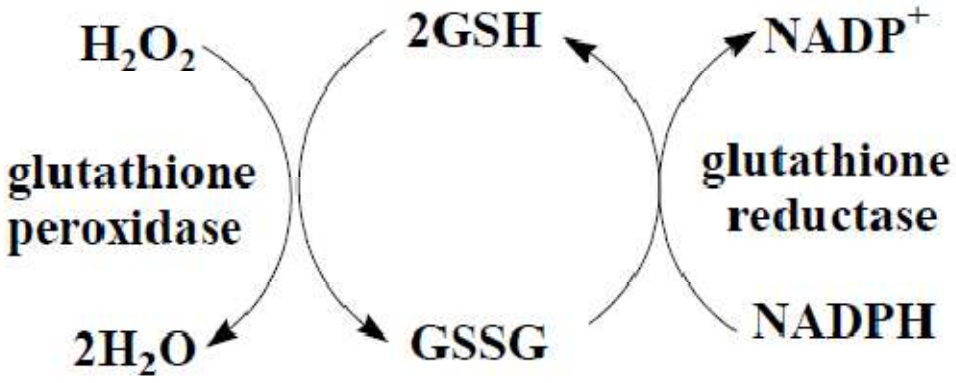


• It is three amino acids together. γ-Glutamylcysteinylglycine
• γ-glutamate – attached via the γ-carbon instead of the α-carbon
• The active part is the -SH group of cysteine (sulfhydryl group)
• The –SH is the reduced form. Two molecules can be bridged bya disulfide bond; which produces the oxidized form (GS-SG).
• These two reactions are catalyzed by **glutathione peroxidase** and**glutathione reductase**

Functions of GSH:
1. **RBCs** – GSH peroxidase and GSH reductase **scavenge for peroxide (free radicals).**

2. **Conjugation** : Lipophilic drugs can be converted to hydrophilic molecules for excretion by attaching it to glutathione

3. **Transport of amino acids** especially in the renal epithelium.



Integration of Metabolism (General information)

The average 70 kg man has fuel stores consisting of:
• 15 kg of triglyceride,
• 6 kg of protein and
• 0.2 kg of glycogen

FED STATE

Signals:
Insulin: Up
Glucagon: Down

Storage forms:
Glycogen: Up
Fat: Up
Protein: Up

FASTING STATE

Signals:
Insulin: Down
Glucagon: Up

Storage forms:
Glycogen: Down
Fat: Down
Protein: Down

STARVATION

Signals:
Insulin: Down
Glucagon: Up

Storage forms:
Glycogen: Exhausted
Fat: Down
Protein: Down

INSULIN:
• It is a signal for high blood glucose levels.
• It stimulates synthesis of glycogen,fat,and protein.
• It inhibits breakdown of glycogen, fat,and protein.
• It increases glucose transport into cells (muscle and adiposetissue).

GLUCAGON:
• It is a signal for low blood glucose levels.
• It stimulates breakdown of glycogen, fat and protein.
• It inhibits synthesis of glycogen, fat and protein.
• It increases protein phosphorylation.
It activates cAMP-dependent protein kinase.

BOOKS RECOMMENDED

1. Berg et al: Biochemistry (5th ed 2001, Freeman)
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3. Mathews et al: Biochemistry (3rd ed 1990, Benjamin/Cummings)
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6. Stryer L.: Biochemistry (5th ed 2002, Freeman)