Protein Metabolism

Digestion And Absorption Of Proteins

- Proteolytic enzymes break down dietary proteins into their constituent amino acids.
- These enzymes are produced by:

  stomach

  pancreas

  small intestine.

- There is no digestion of protein in mouth.

Digestion in Stomach

When protein enters the stomach, *gastrin* stimulates the release of :

- Hydrochloric acid
- Pepsinogen
- Rennin in infants.

Hydrochloric acid

- Denatures proteins making their internal peptide bonds more accessible to subsequent hydrolysis by proteoses

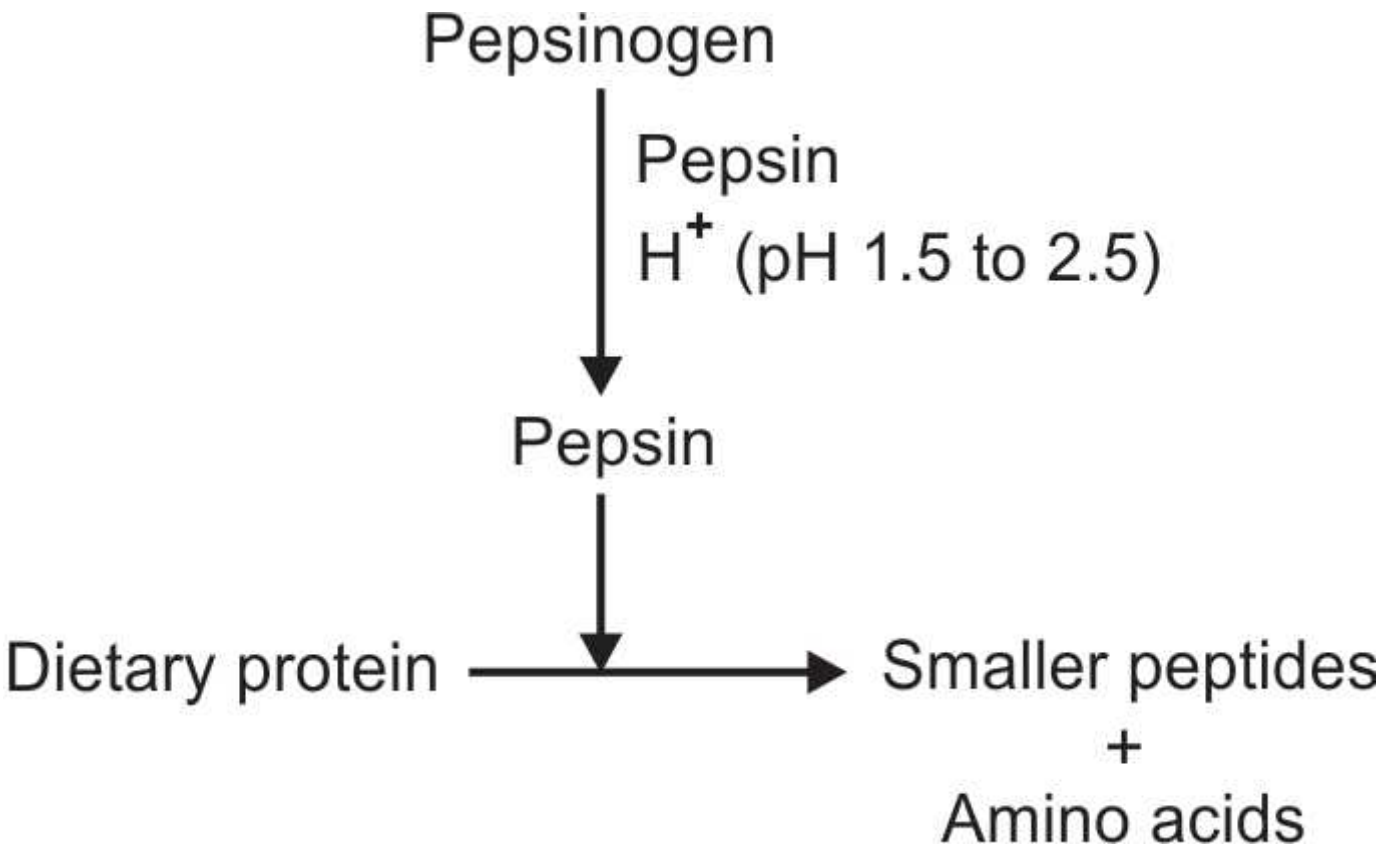
- Provides an acid environment for the action of pepsin.

Pepsin

- It is secreted as the *pepsinogen* an inactive form.

- Pepsin cleaves those peptide bonds of protein involving :
  - *Aromatic amino acids* (phenylalanine, tyrosine and tryptophan)
  - *Acidic amino acids* (aspartic acid and glutamic acid).

- Pepsin cleaves long polypeptide chains into a mixture of smaller peptides and some free amino acids.



Activation of pepsinogen and action of pepsin

Rennin

- Absent in adults.
- Also called chymosin or rennet.
- Action of rennin is to clot milk. The purpose of this reaction is to convert milk into a more solid form to prevent the rapid passage of milk from the stomach of infants.

Rennin and renin are different. Renin is secreted by kidney and is involved in regulation of water and electrolyte balance and blood pressure.

Digestion in Intestine by Pancreatic Enzymes

There are two types of peptidase enzymes secreted by pancreas.

1. Endopeptidase
2. Exopeptidase

- Endopeptidases cleave internal peptide bonds.

- Exopeptidase hydrolyse the peptide bonds of terminal amino acids.

- Endopeptidases secreted by pancreas are:

  - *Trypsin*
  - *Chymotrypsin*
  - *Elastase*.

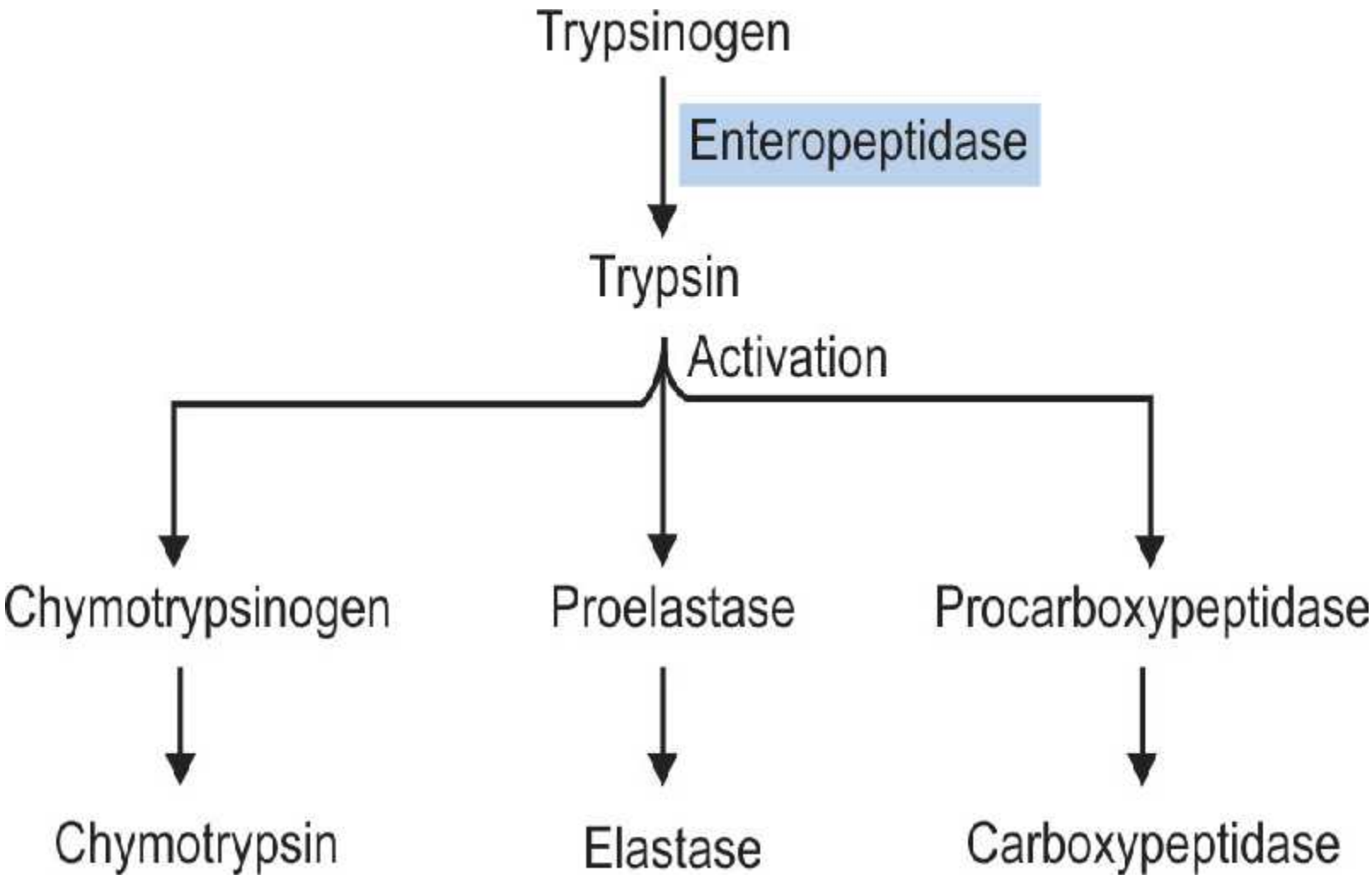
- Endopeptidase are secreted in proenzyme (inactive) forms,

  Trypsinogen

  Chymotrypsinogen

  Proelastase.

- Exopeptidase are of two types:
  - Carboxypeptidase secreted by pancreas act on C-terminal amino acid
  - Aminopeptidases secreted by mucosal cell act on N-terminal amino acid.



Activation of pancreatic proenzymes.

Specificity of proteolytic enzymes.		
Enzymes	***Site of action***	***Cleavage points of peptide bond***
Pepsin	Stomach	Phe, Trp, Tyr, and Leu (N)
Trypsin	Intestine	Lys, Arg (C)
Chymotrypsin	Intestine	Phe, Trp, and Tyr (C)
Elastase	Intestine	Small nonpolar amino acids like alanine, serine, and glycine
Carboxypeptidase	Intestine	Successive C-terminal amino acids
Aminopeptidase	Intestinal mucosa	Successive N-terminal amino acids
Peptide bond cleavage occurs on either the carbonyl (C) or the amino (N) side of the indicated amino acid residues.		

Digestion in Intestine by Intestinal Proteases

- Enzymes secreted by the mucosa of the small intestine:

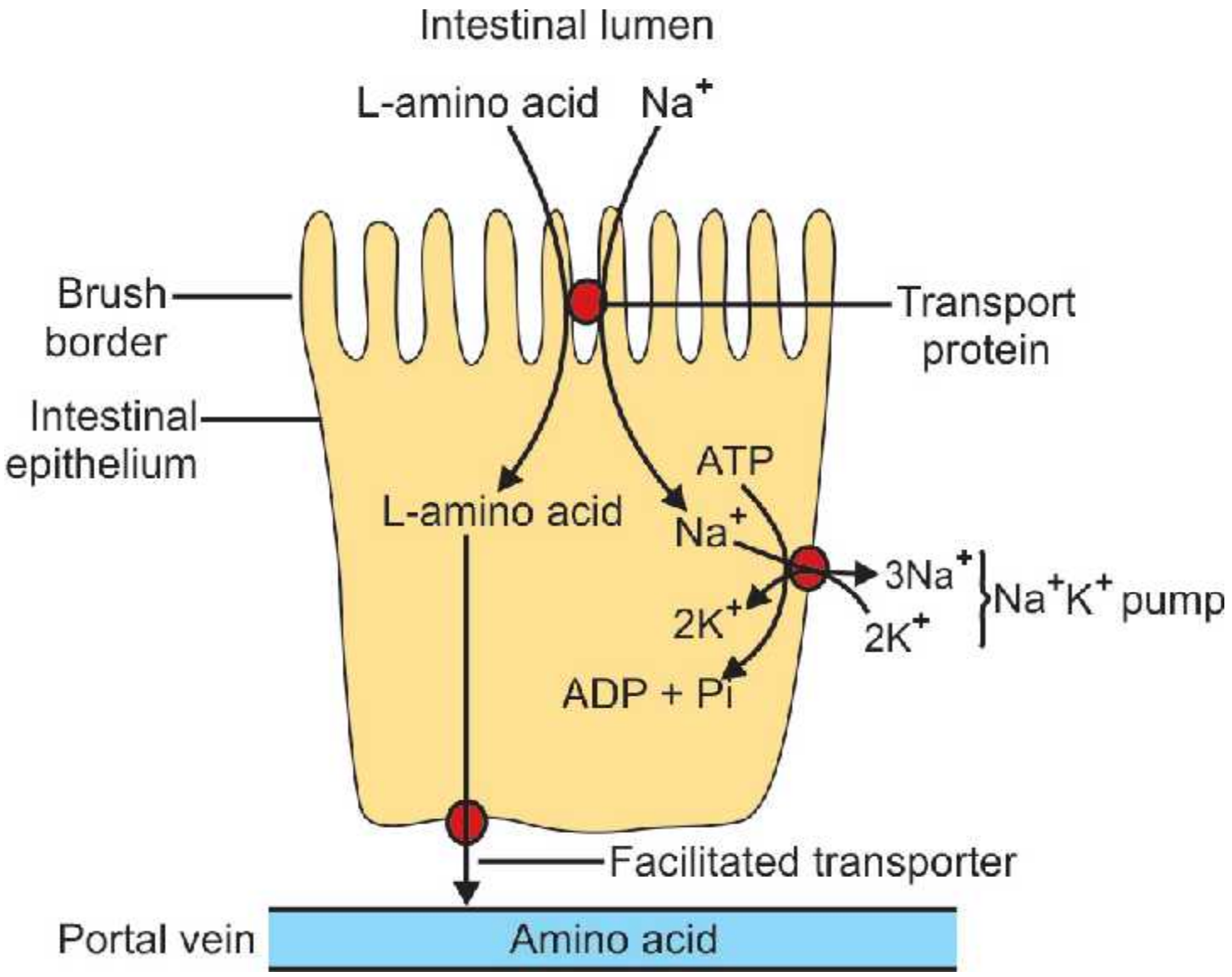
  Aminopeptidases

  Dipeptidases.

- Aminopeptidase is an exopeptidase, hydrolyze peptide bonds next to N-terminal amino acids of the short peptides.
- The dipeptidases complete digestion of dipeptides to free amino acids.

Absorption of Amino Acid

- The absorption of most amino acids involves an *active transport mechanism*, requiring ATP and specific transport proteins
- Many transporters have Na+ dependent mechanisms, coupled with Na+ K+ pump .
- All are specific for only L-amino acid.
- D-amino acids are transported by passive diffusion.



Stereospecific transport of L-amino acid across the intestinal epithelium.

Alton Meister proposed that glutathione (γ-glutamyl cysteinylglycine) participates in absorption of amino acids in intestine, kidneys and brain and the cycle is called gammaglutamyl cycle or Meister cycle.

Absorption of Intact Protein

- Small intestinal cells of fetal and new-born infants are able to absorb intact proteins, e.g. immunoglobulin
- IgA from colostrum of maternal milk are absorbed intact without loss of biologic activity, so that they provide passive immunity to the infant.
- The intact proteins are not absorbed by adult intestine

Disorders Associated with Amino Acid Transport

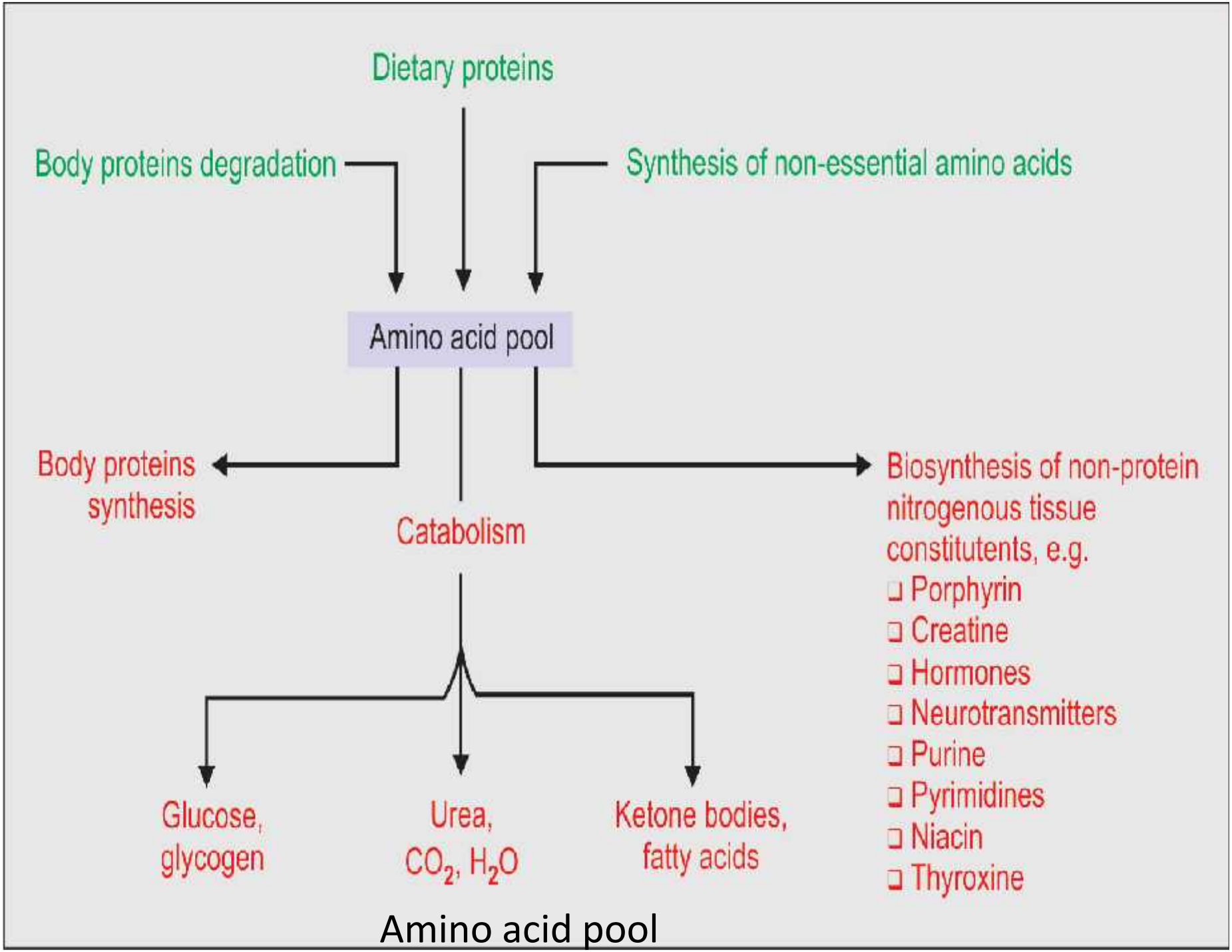
Genetic diseases arising from defects in amino acid transporters have been reported

Amino acid transport systems and disorders.		
Transport systems	***Amino acid transported***	***Disorders associated with defective transport***
Small neutral amino acids	Alanine, serine, and threonine	Hartnup disease
Large neutral amino acids	Isoleucine, leucine, valine, tyrosine, tryptophan, and phenylalanine	
Basic amino acids and cystine	Arginine, lysine, and cystine	Cystinuria
Acidic amino acid	Glutamic acid and aspartic acid	
Imino acid and glycine	Proline, hydroxyproline, and glycine	Glycinuria

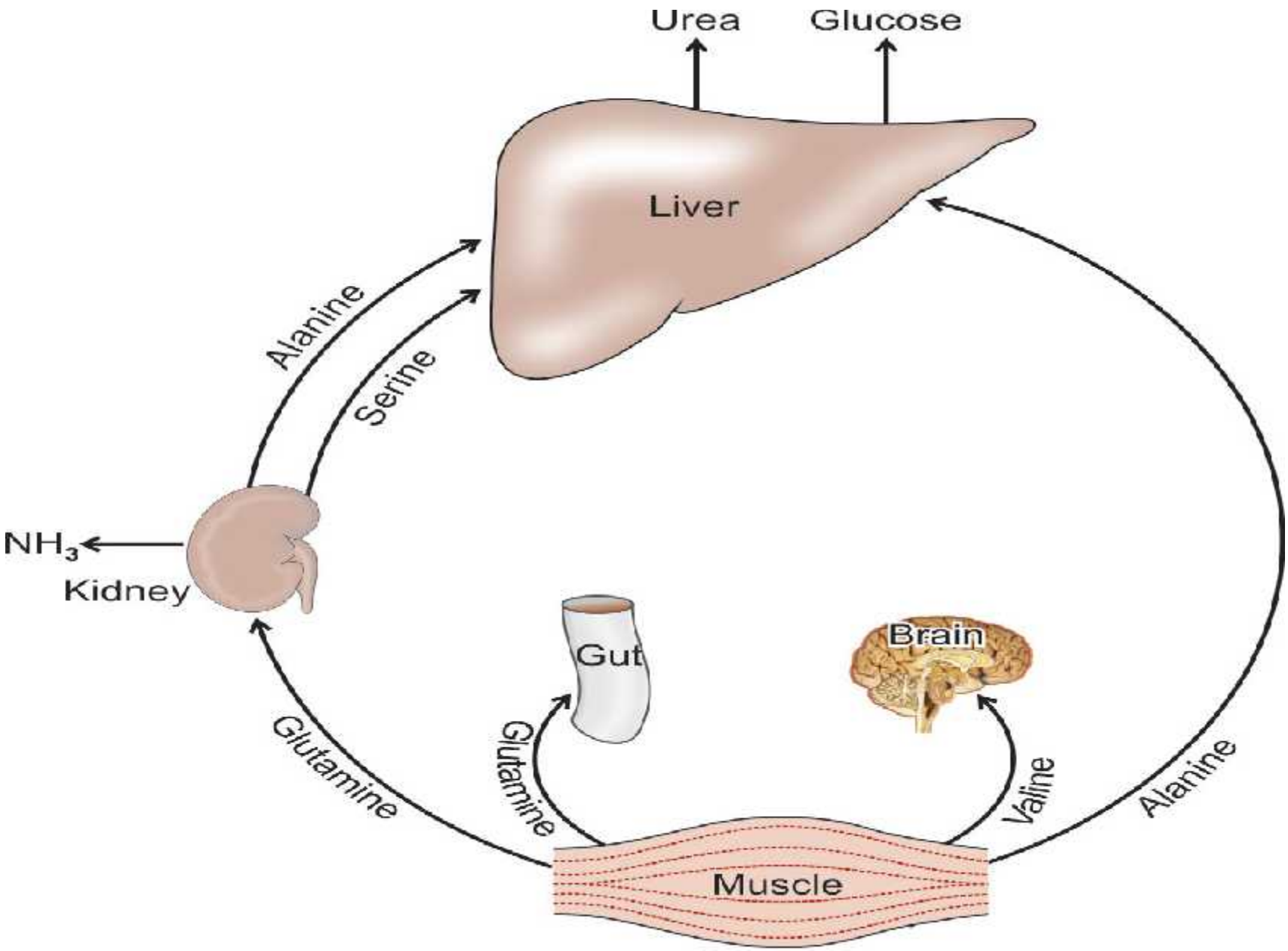
Amino Acid Pool

Amino acids, released by hydrolysis of dietary protein, and tissue proteins together constitute the *amino acid pool*.

- The primary role of amino acids is to serve as building blocks of synthesis of tissue protein and other nitrogen containing compounds
- Amino acids, in excess of those needed for the synthesis of proteins and other biomolecules cannot be stored nor are they excreted. Surplus amino acids are oxidized for energy.



Amino acid pool



Amino acid exchanges between organs and key role of alanine.

STAGES OF CATABOLISM OF AMINO ACIDS

- In the catabolism, amino acids lose their **amino groups** in the form of **ammonia**, which is excreted in the form of **urea** in the liver by reactions of the **urea cycle**.
- The remaining **carbon skeleton** of amino acids, the α-keto acids undergo oxidation to carbon dioxide and water by reactions of **citric acid cycle**.

The complete catabolism of amino acids includes:

1. The removal of α-amino group in the form of ammonia by following reactions:

- Transamination by the enzyme aminotransferase, also called transaminase.
- Deamination may be oxidative or non-oxidative.

  a. Oxidative deamination is by glutamate dehydrogenase or amino acid oxidase.

  b. Non-oxidative deamination by amino acid dehydratase.

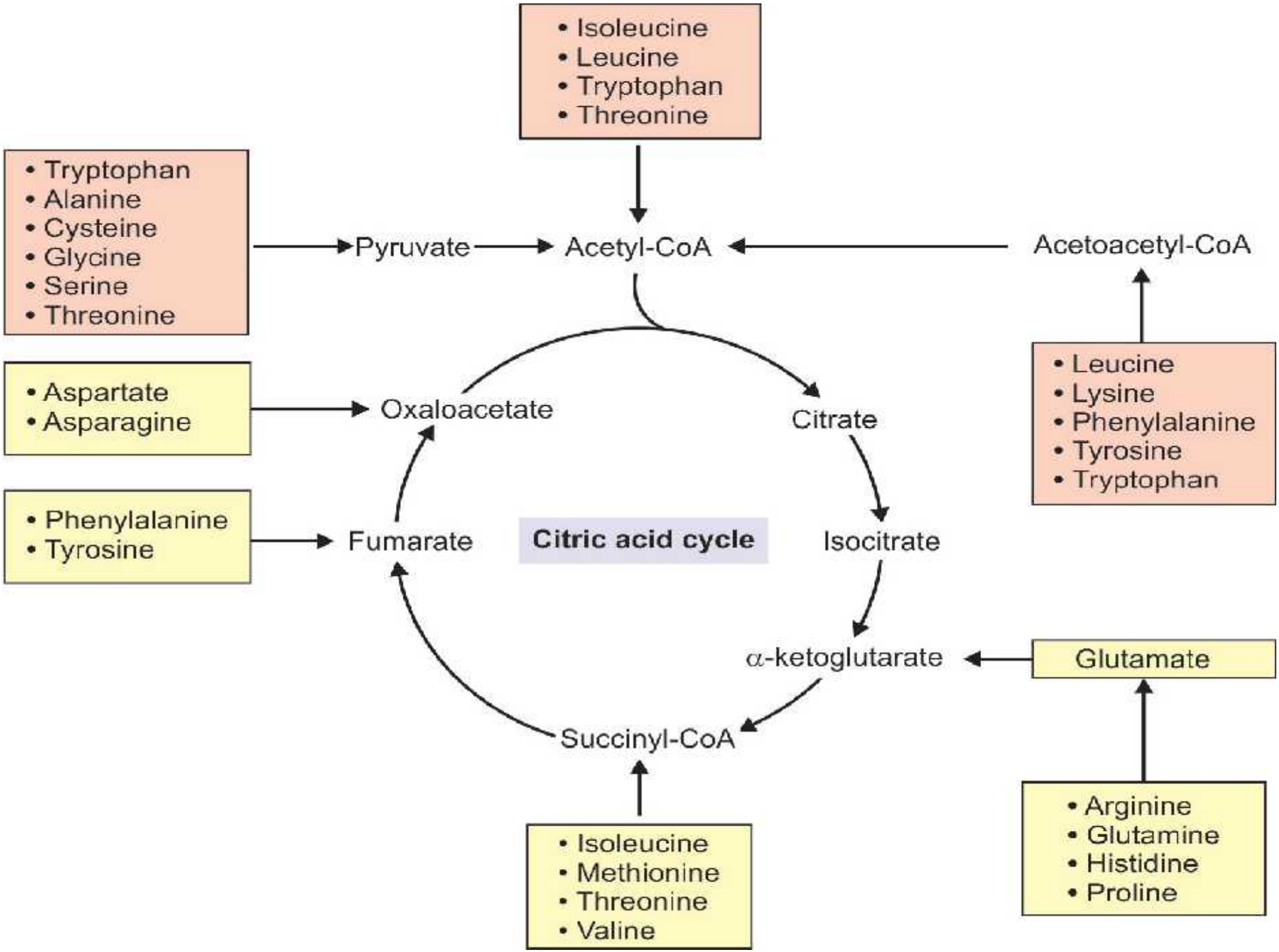
2. Transport of ammonia.

3. Disposal of ammonia in the form of urea in the liver by reactions of the urea cycle.

4. Disposal (catabolism) of carbon skeleton of amino acids after the removal of α-amino group. The carbon skeletons of 20 amino acids converged to form only seven major products. These are:

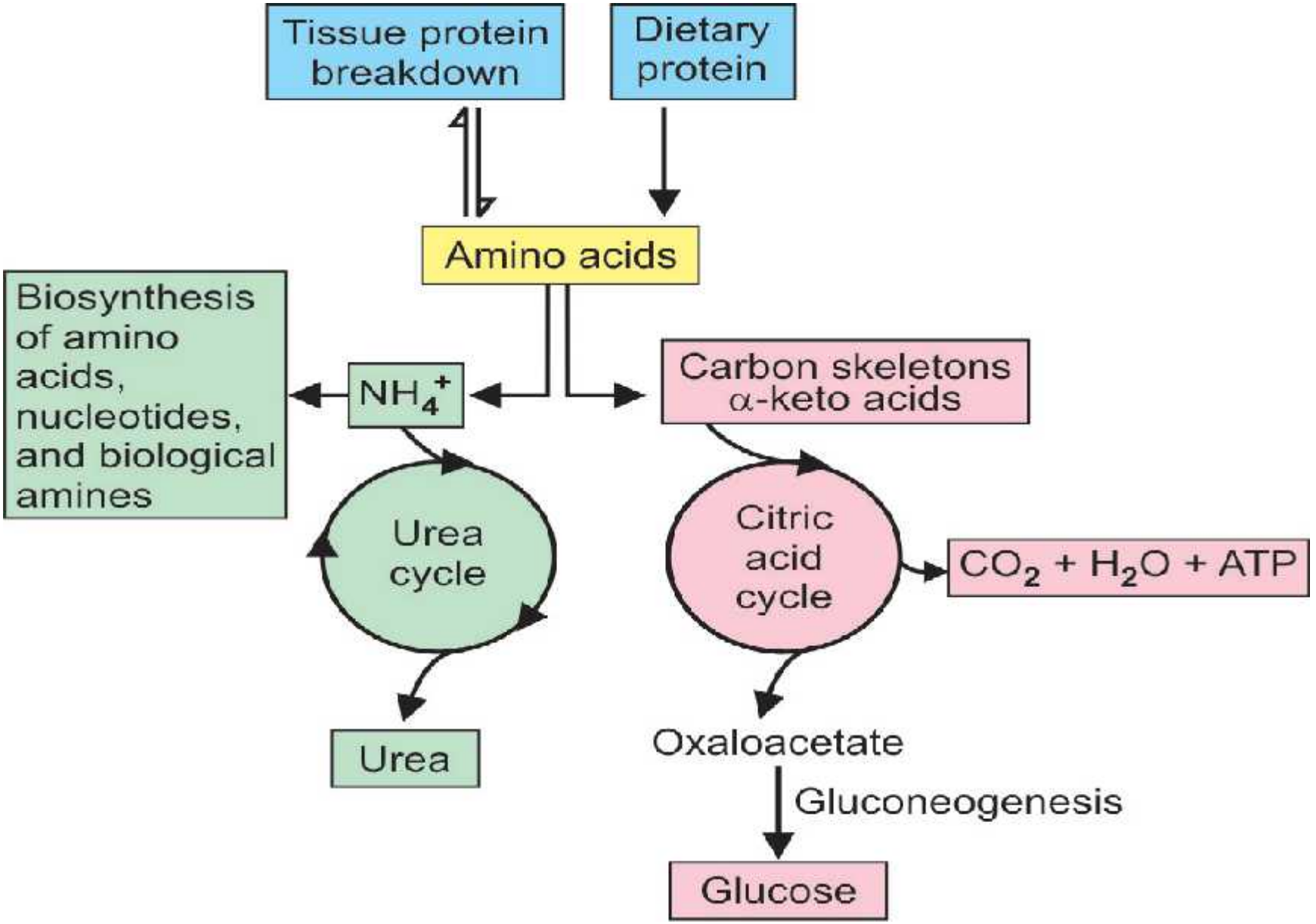
1. Pyruvate
2. Acetyl-CoA
3. Acetoacetyl-CoA
4. α-ketoglutarate
5. Succinyl-CoA
6. Fumarate
7. Oxaloacetate

- All these enter the citric acid cycle
- From here the carbon skeletons are diverted to gluconeogenesis or ketogenesis or are completely oxidized to CO_2 and H_2O.


• Isoleucine
• Leucine
• Tryptophan
• Threonine
• Tryptophan
• Alanine
• Cysteine
• Glycine
• Serine
• Threonine
Pyruvate
Acetyl-CoA
Acetoacetyl-CoA
• Leucine
• Lysine
• Phenylalanine
• Tyrosine
• Tryptophan
• Aspartate
• Asparagine
Oxaloacetate
Citrate
• Phenylalanine
• Tyrosine
Fumarate
Citric acid cycle
Isocitrate
α-ketoglutarate
Glutamate
Succinyl-CoA
• Isoleucine
• Methionine
• Threonine
• Valine
• Arginine
• Glutamine
• Histidine
• Proline

Metabolic fate of carbon skeleton of amino acids.

- Yellow shade: Glucogenic amino acids;
- Red shade: Ketogenic amino acid.
- Several amino acids are both glucogenic and ketogenic.
- Only two amino acids, leucine and lysine, are exclusively ketogenic.

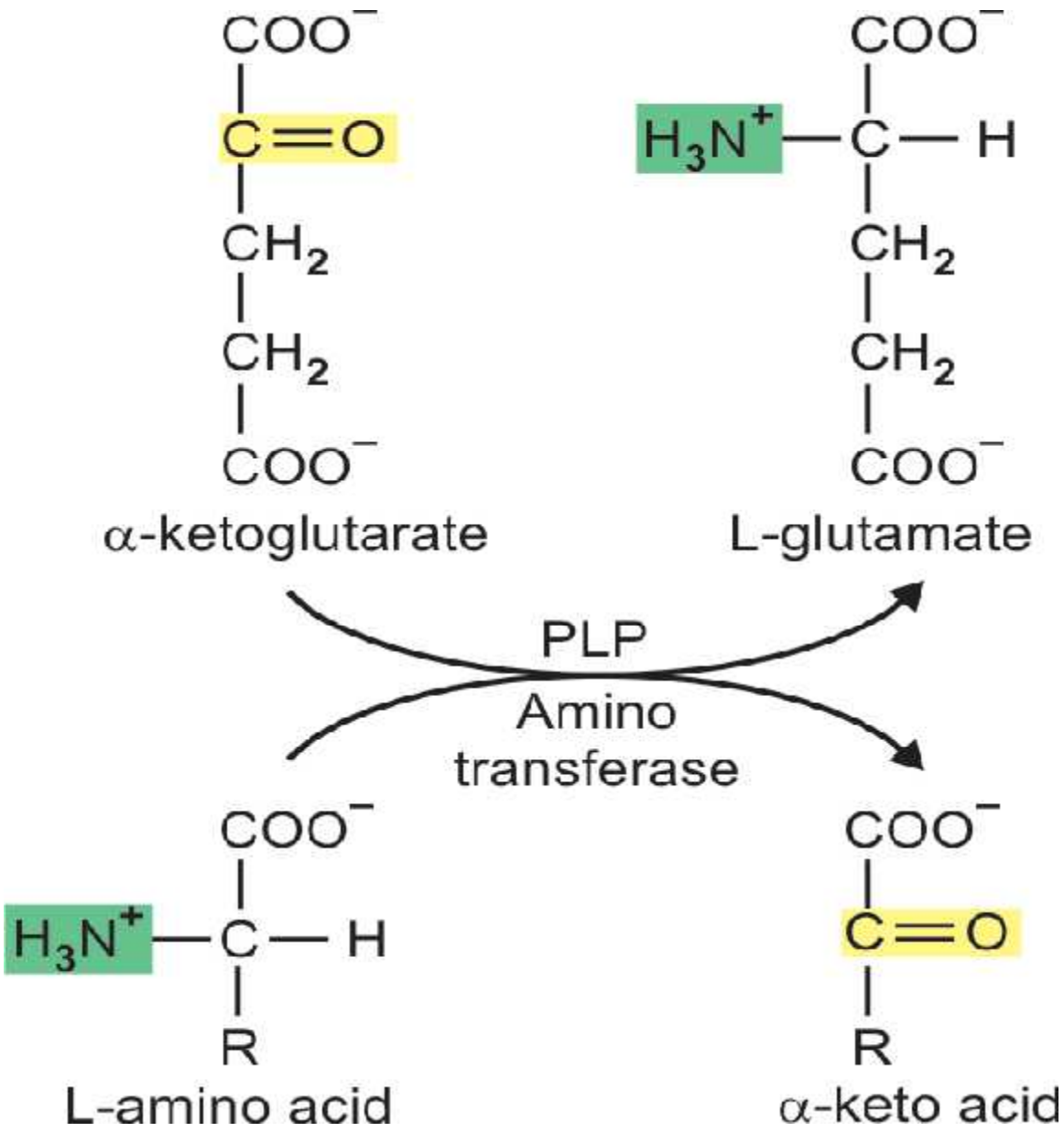


Amino acid catabolism overview.

1. The removal of α-amino group in the form of ammonia

Transamination

- Transamination involves the transfer of α-amino group of α-amino acid to an α-keto acid to form new amino acid and a new keto acid.
- The enzymes that catalyze these reactions are called *aminotransferases* or *transaminases*
- All transaminases require pyridoxal phosphate (PLP) as a coenzyme.



Transamination reaction.

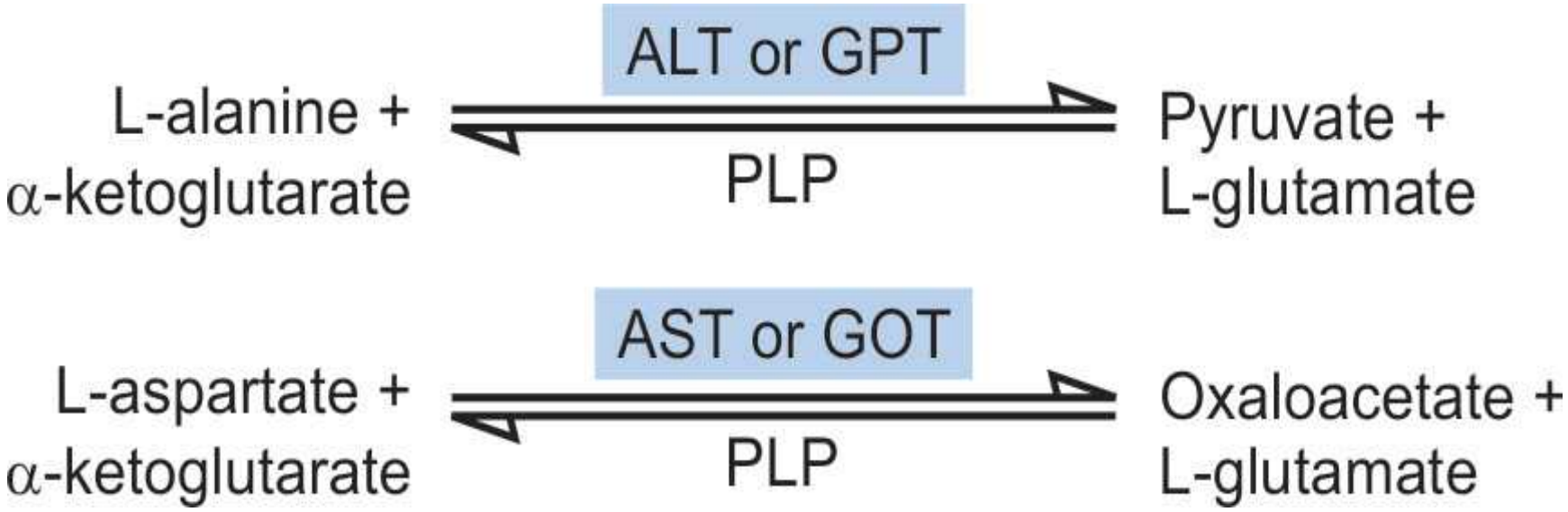
- Some of the most important transaminases are :

  - Alanine transaminase (ALT) or

    Glutamate pyruvate transaminase (GPT)

  - Aspartate transaminase (AST) or

    Glutamate oxaloacetate transaminase (GOT)



Reactions catalyzed by alanine transaminase and aspartate transaminase.

- Except *lysine*, *threonine*, proline and *hydroxyproline* all amino acids undergo transamination reaction

- There is no net loss of amino groups in transamination reactions.

Metabolic significance of transamination reactions

- This reaction provides a mechanism for collecting the amino groups from many different amino acids into one common product L-glutamate.
- L-glutamate, produced by transamination, can be used as an amino group donor in the synthesis of non-essential amino acids.

Clinical significance of transaminase enzyme

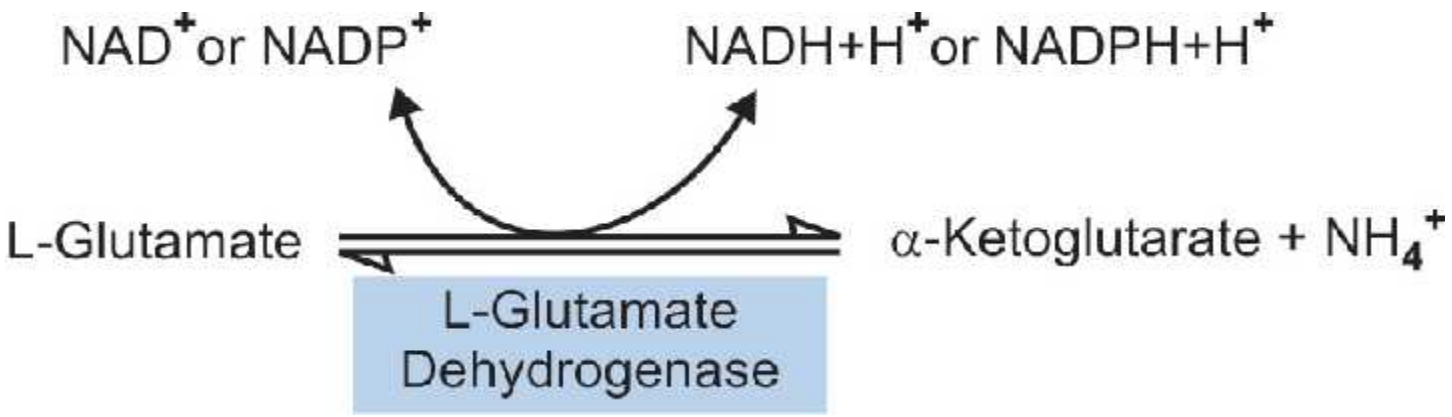
ALT and AST are important in the diagnosis of liver and heart damage

Deamination

Deamination may be oxidative or non-oxidative

- Oxidative deamination is by glutamate dehydrogenase or amino acid oxidase.
- Nonoxidative deamination is by amino acid dehydratase.

Oxidative deamination by glutamate dehydrogenase



The net removal of α-amino groups to ammonia, requires the combined action of glutamate transaminase and glutamate dehydrogenase.

Metabolic significance

- This reaction functions both in amino acid catabolism and biosynthesis.
- Catabolically it channels nitrogen from glutamate to ammonia
- Anabolically it catalyzes amination of α-ketoglutarate to glutamate.

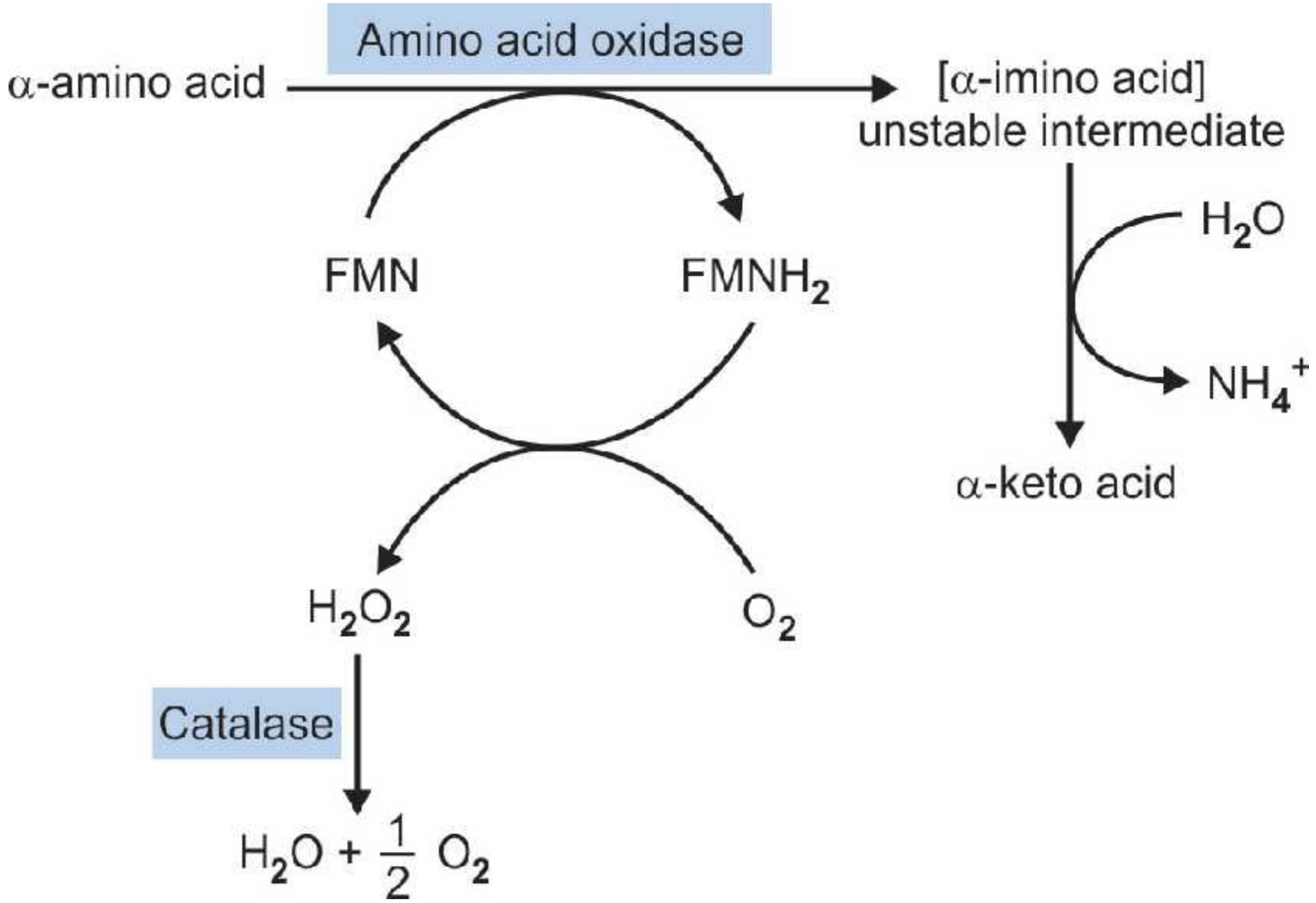
Clinical significance of glutamate dehydrogenase (GLD)

GLD is present in normal serum in trace amount only, but increased activities are observed in cases of liver disease.

Oxidative deamination by amino acid oxidases

- Both L- and D-amino acid oxidases occur in the kidneys and the liver.

- Amino acid oxidases use auto-oxidizable FMN or FAD as coenzyme

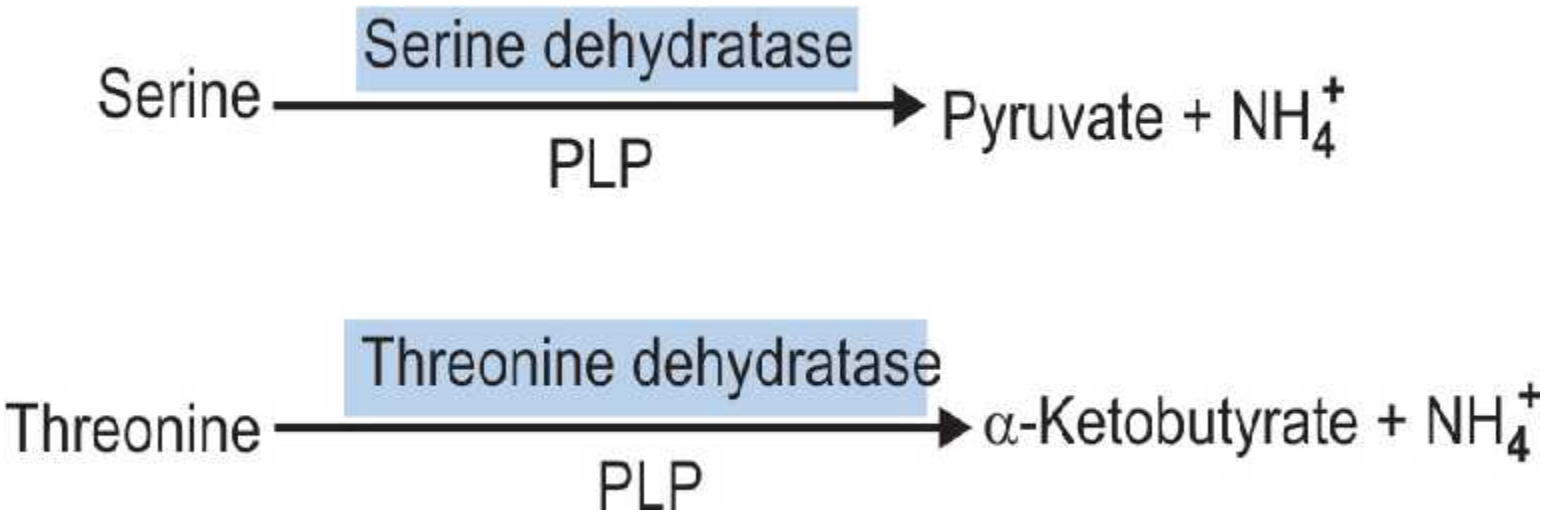


Oxidative deamination by amino acid oxidase.

Metabolic significance

D-amino acids present in the diet are metabolized by D-amino acid oxidase in the liver

Nonoxidative deamination by amino acid dehydratase



2. Transport of ammonia.

Disposal Of Ammonia Or Metabolic Fate Of Ammonia

Transport Ammonia from Peripheral Tissues to the Liver

- In many tissues, including the brain, some processes such as nucleotide degradation generate free ammonia.

- Since ammonia is extremely toxic, much of the free ammonia is immediately converted to nontoxic metabolites before export from the extrahepatic tissues into the blood and transport to the liver or kidneys.

- From extrahepatic tissues and muscles, free ammonia is transported to the liver in two principal transport forms:

    1. Glutamine, and

    2. Alanine.

Transport of Ammonia in the Form of Glutamine

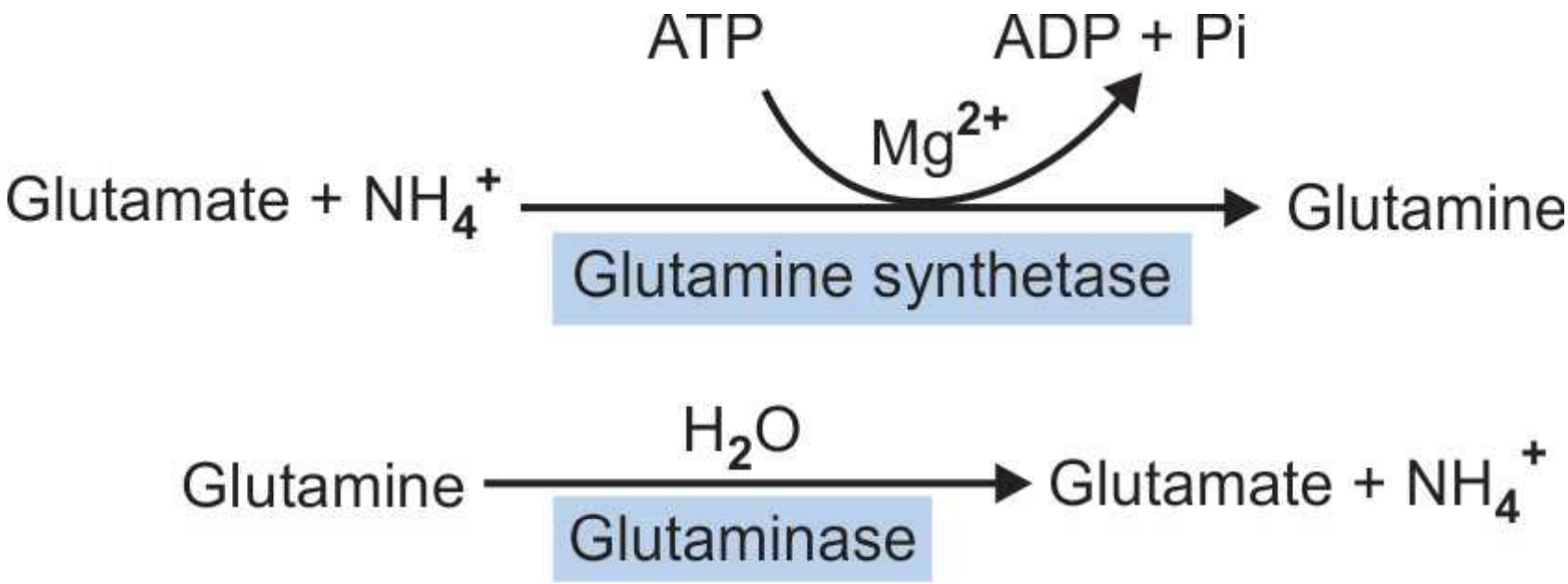
- In many tissues (liver, kidney, and brain), free ammonia is combined with **glutamate** to yield **glutamine** by the action of **glutamine synthetase**.

- The glutamine, so formed, is a neutral nontoxic compound, which can readily pass through cell membrane, whereas **glutamate cannot**.

- Glutamine is normally present in blood in much higher concentrations than other amino acids.

- Glutamine also serves as a source of amino groups in a variety of biosynthetic reactions.

- The glutamine is carried via blood to the liver, where it can be acted upon by glutaminase to yield glutamate and ammonia

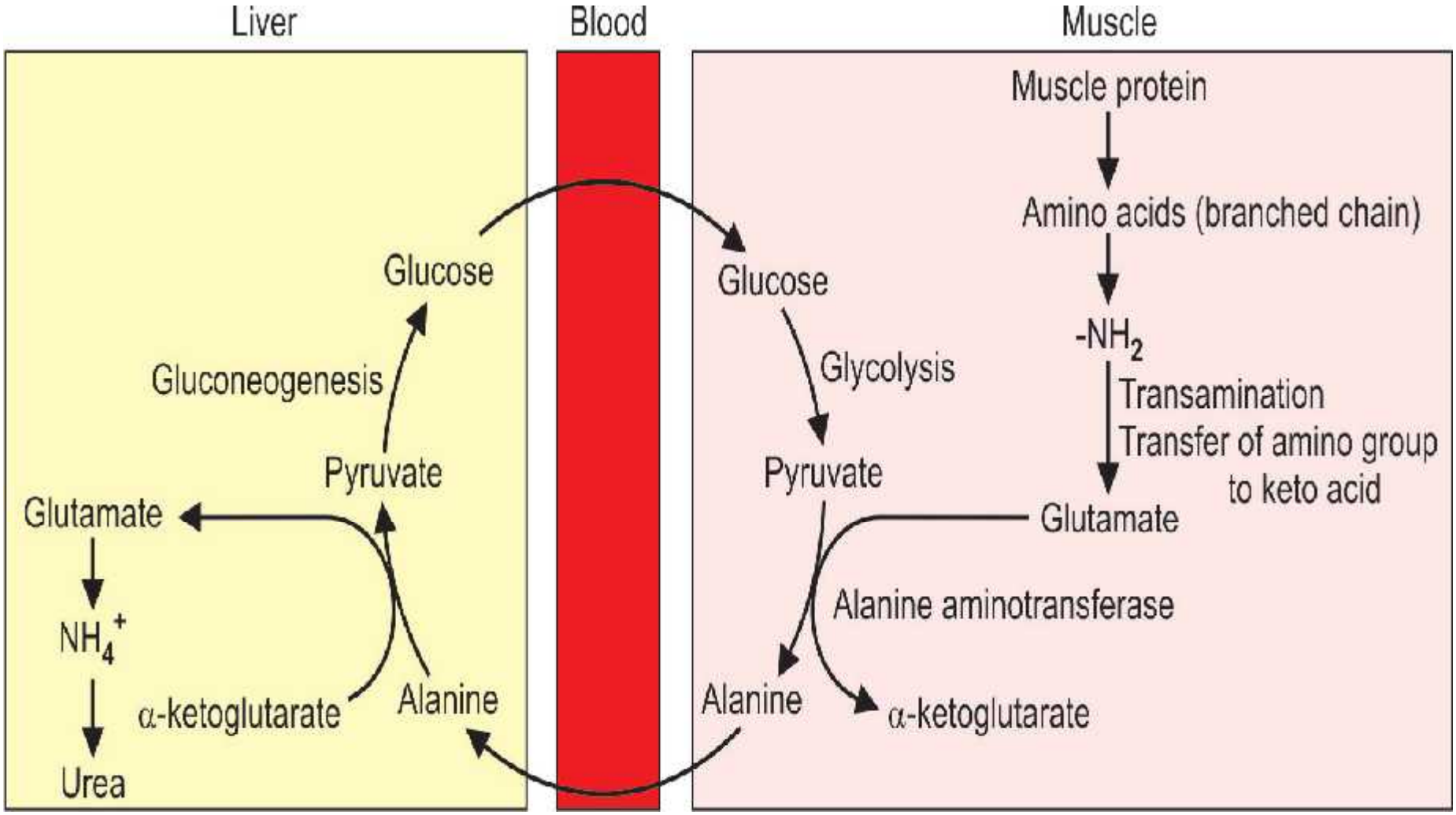


Formation and breakdown of glutamine.

Glutamine is a major transport and temporary storage form of ammonia in the body.

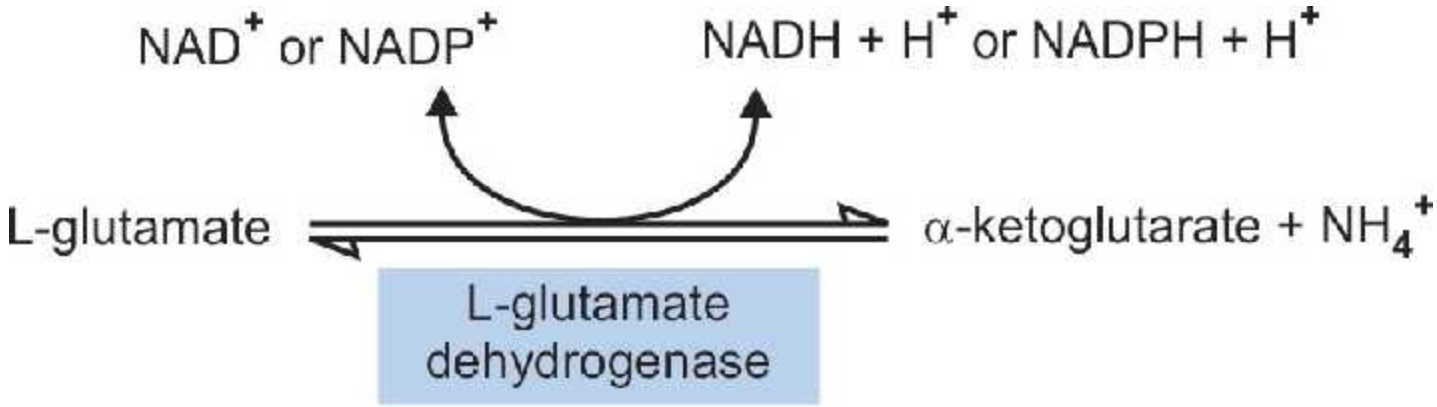
Transport of Ammonia in the Form of Alanine

- Alanine transports ammonia to the liver in nontoxic form via a pathway called the **glucose alanine cycle.**
- In muscle, amino groups from amino acids are collected in the form of **glutamate** by transamination.
- Glutamate transfer its α-amino group to **pyruvate** by the action of **alanine aminotransferase (ALT)** to form **alanine**. Alanine so formed passes into the blood and travels to the liver.

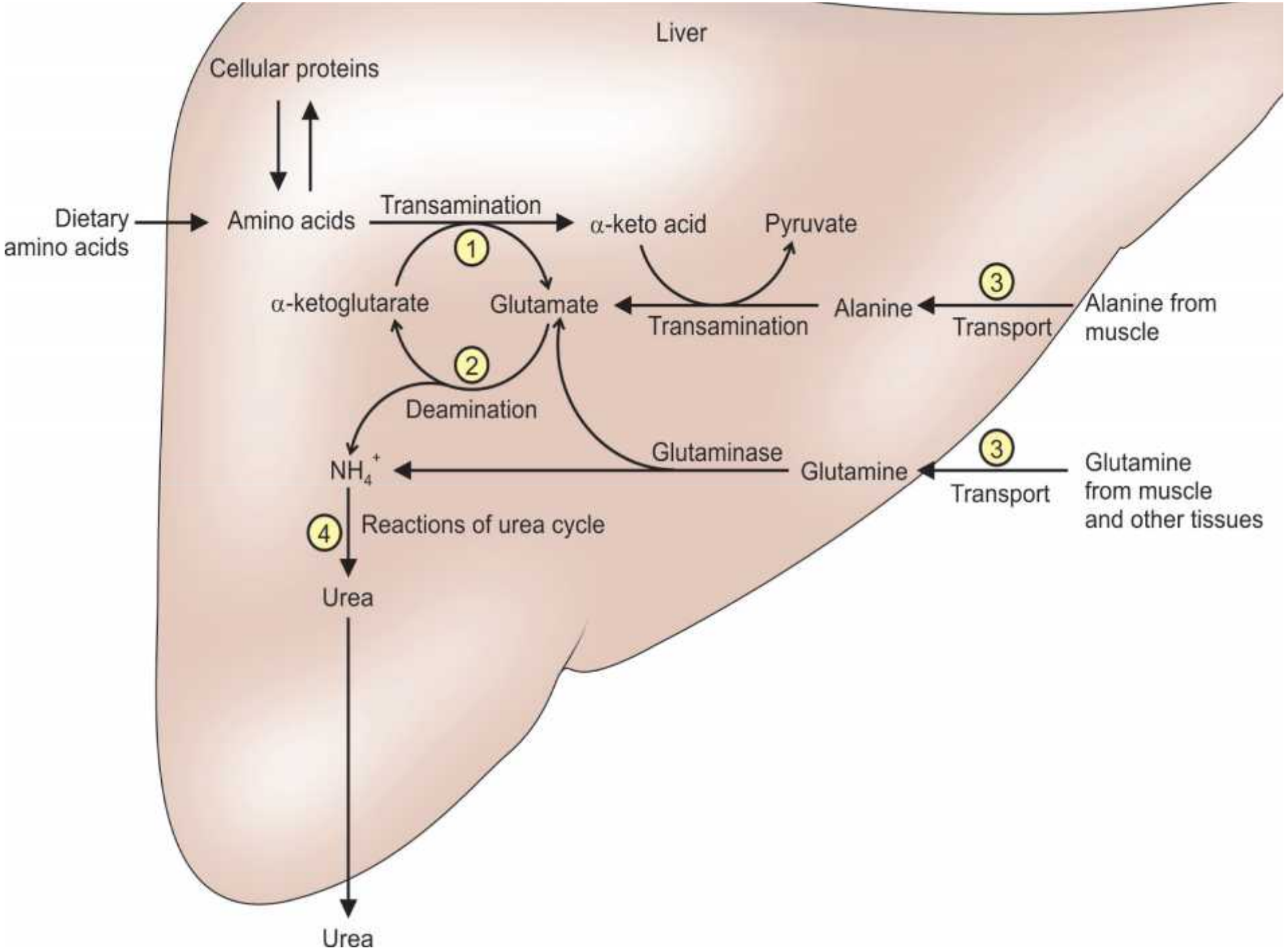


Transport of ammonia from muscle to liver by glucose-alanine cycle.

- In the liver by reverse reaction, ALT transfers the amino group from alanine to α-ketoglutarate, forming pyruvate and glutamate.
- Glutamate can then enter mitochondria where **glutamate dehydrogenase** reaction release ammonia.

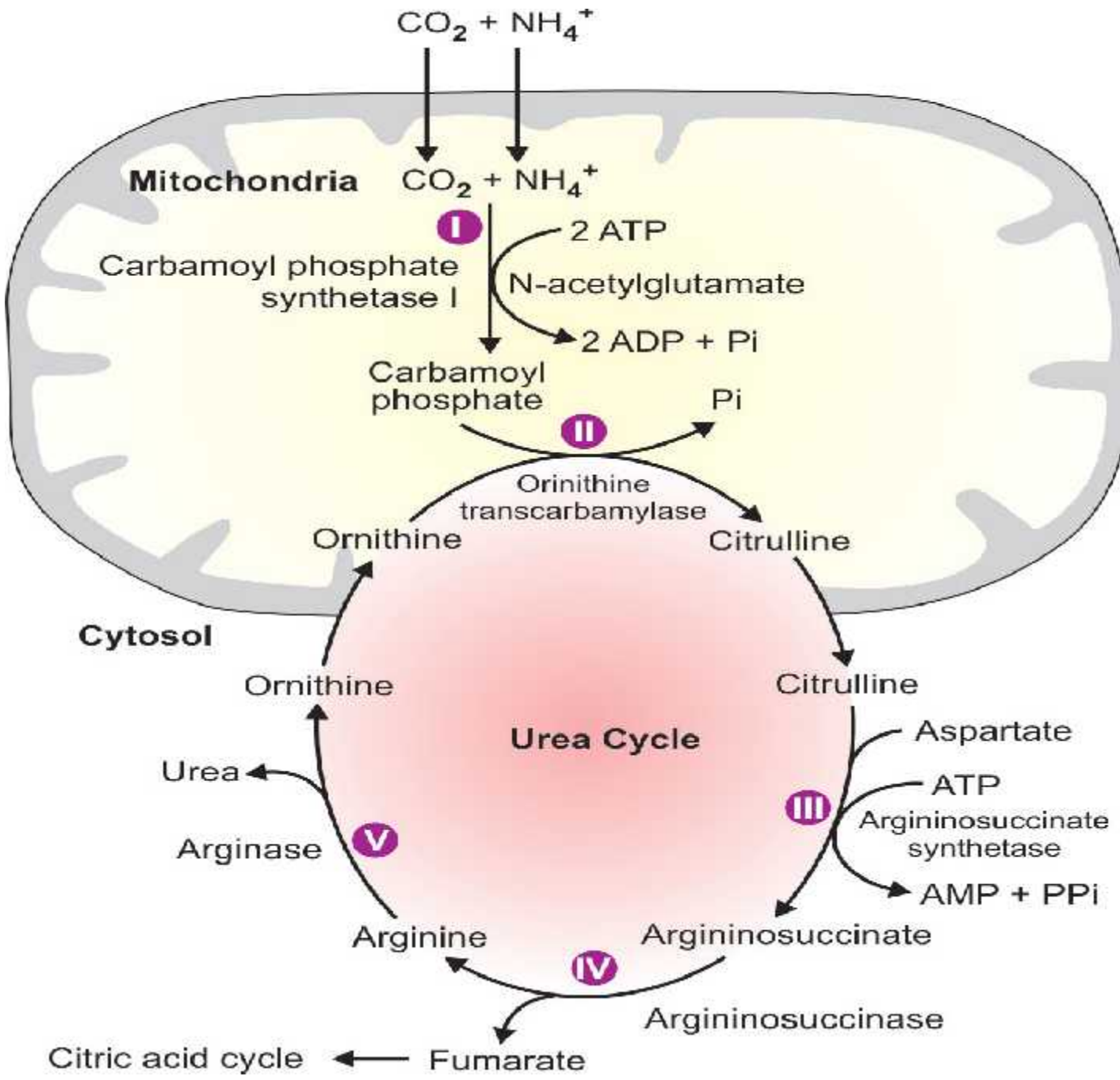


The overview of catabolism of amino groups


Liver
Cellular proteins
Dietary amino acids
Amino acids
Transamination
α-keto acid
Pyruvate
1
α-ketoglutarate
Glutamate
Transamination
Alanine
3
Transport
Alanine from muscle
2
Deamination
Glutaminase
NH_4^+
Glutamine
3
Transport
Glutamine from muscle and other tissues
4
Reactions of urea cycle
Urea
Urea

3. Disposal of ammonia in the form of urea in the liver by reactions of the urea cycle.

- Ammonia formed in the breakdown of amino acids if not reused for the synthesis of new amino acids or other nitrogenous compounds, the excess ammonia is converted into urea in the urea cycle and excreted into the urine.
- Formation of urea by *“Kreb’s Henseleit urea cycle "is* an ultimate route for the metabolic disposal of ammonia.
- Urea is produced exclusively by the liver.
- Enzymes catalyzing the urea cycle reactions are distributed between the *mitochondria* and the *cytosol* of the liver


$CO_2 + NH_4^+$
Mitochondria
$CO_2 + NH_4^+$
I
2 ATP
Carbamoyl phosphate synthetase I
N-acetylglutamate
2 ADP + Pi
Carbamoyl phosphate
Pi
II
Orinithine transcarbamylase
Ornithine
Citrulline
Cytosol
Ornithine
Citrulline
Urea Cycle
Aspartate
Urea
ATP
III
Arginase
V
Argininosuccinate synthetase
AMP + PPi
Arginine
Argininosuccinate
IV
Argininosuccinase
Citric acid cycle
Fumarate

- *Carbamoyl phosphate synthase-l* is a hepatic mitochondrial enzyme functions in urea synthesis.

- *Carbamoyl phosphate synthase-ll* a cytosolic enzyme that uses glutamine rather than ammonia as a nitrogen source and functions in pyrimidine nucleotide biosynthesis

The Energy Cost of Urea Cycle

Four ATPs are consumed in the synthesis of each molecule of urea as follows:

- Two ATP are needed to make carbamoyl phosphate.
  - -- One ATP serves as a source of phosphate
  - -- Second ATP is converted to AMP + PPi.
- One ATP is required to make arginosuccinate.
- One ATP is required to restore AMP to ATP.

Significance of Urea Cycle

- The toxic ammonia is converted into the harmless nontoxic urea.
- It disposes off two waste products, **ammonia and carbon dioxide**.
- It participates in the regulation of blood pH, which depends upon the ratio of dissolved CO_2, i.e. H_2CO_3 to HCO_3^-.

- It forms semi-essential amino acid, arginine. Arginine also serves as the precursor of the potent muscle relaxants nitric oxide (NO).

- Ornithine, which is formed in urea cycle, is a precursor for the formation of polyamines like putrescine, spermidine, and spermine and can also be involved in formation of nonessential amino acid **proline**

Regulation of Urea Cycle

Carbamoyl phosphate synthetase-I is an allosteric regulatory enzyme of urea cycle, which is allosterically activated by *N-acetylglutamate (NAG).*

Metabolic Inborn Errors of Urea Cycle

- Five disorders associated with each of the five enzymes of urea cycle have been reported.
- Since urea synthesis converts toxic ammonia to non-toxic urea, all defects in urea synthesis result in *hyperammonemia* and *ammonia intoxication*.

- This intoxication is more severe when the metabolic block occurs at reaction I or II, since it accumulates ammonia itself.
- Deficiency of later enzymes result in the accumulation of other intermediates of the urea cycle, which are less toxic and therefore severity of symptoms is less.

Symptoms

Clinical symptoms, common to all urea cycle, disorders include:

- Ammonia intoxication
- Protein-induced vomiting
- Intermittent ataxia
- Irritability
- Lethargy
- Mental retardation.

[redacted]: Disorders caused by genetic defects of urea cycle enzymes.

Disorders	*Defective enzymes*	*Products accumulated*
Hyperammonemia type-I	Carbamoyl phosphate synthetase-I	Ammonia
Hyperammonemia type-II	Ornithine transcarbamylase	Ammonia
Citrullinemia	Argininosuccinate synthetase	Citrulline
Argininosuccinic aciduria	Argininosuccinase	Argininosuccinate
Argininemia	Arginase	Arginine

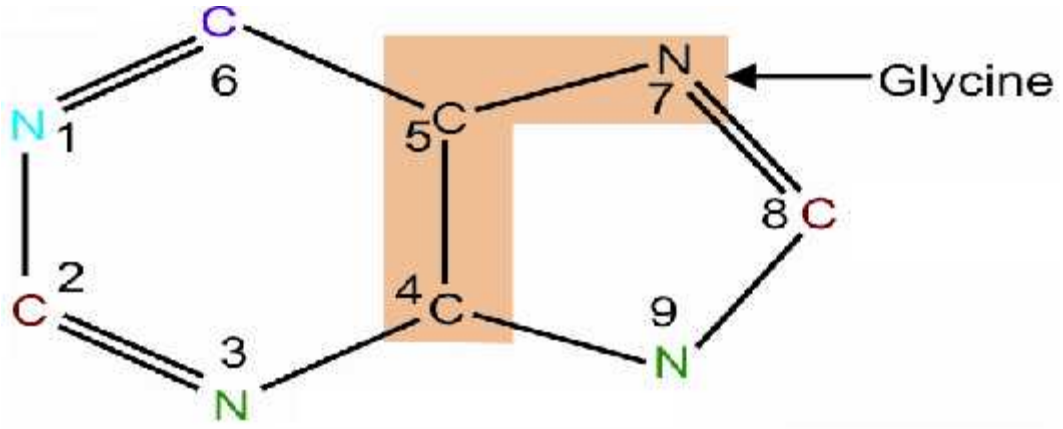
Treatment

Low protein diet, food intake should be in frequent small meals to avoid sudden increase in blood ammonia levels.

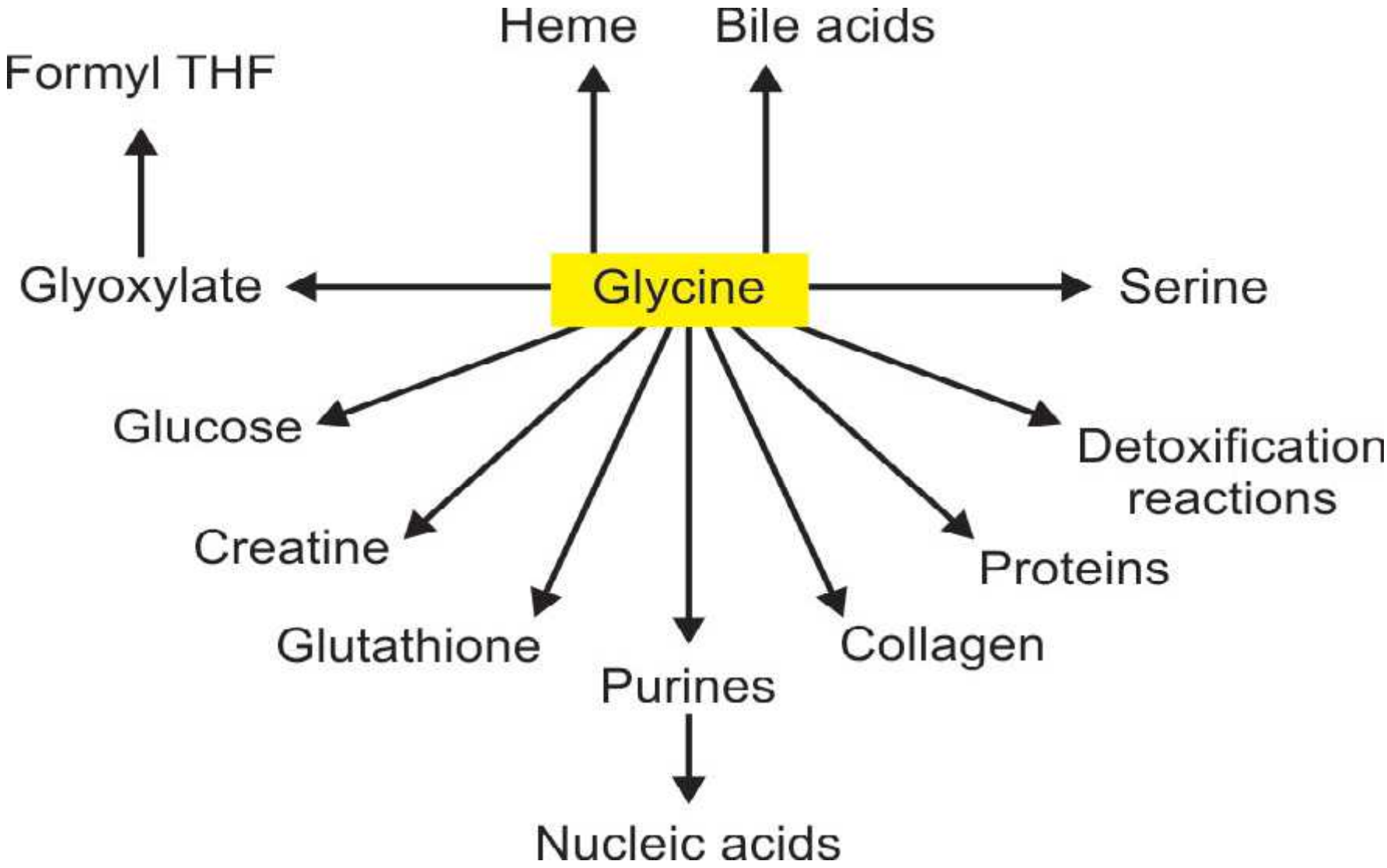
The Metabolic Pathways For Aliphatic Side Chain Containing Amino Acids (Glycine, Alanine, Valine, Isoleucine And Leucine)

Metabolic Importance of Glycine

- Synthesis of heme
- Synthesis of glutathione (γ-glutamyl-cysteinyl-glycine).
- Formation of purine ring



- Formation of bile acids: glycocholic & glyco- chenodeoxy cholic acid.
- Glycine is involved in detoxification reactions :e.g. benzoic acid is detoxicated by conjugating with glycine to hippuric acid.
- Synthesis of creatin: Glycine +Arginine +Methionine
- Collagen formation
- Synthesis of glucose.
- Constituent of various tissue proteins, hormones ,enzymes.



Metabolic importance of glycine.

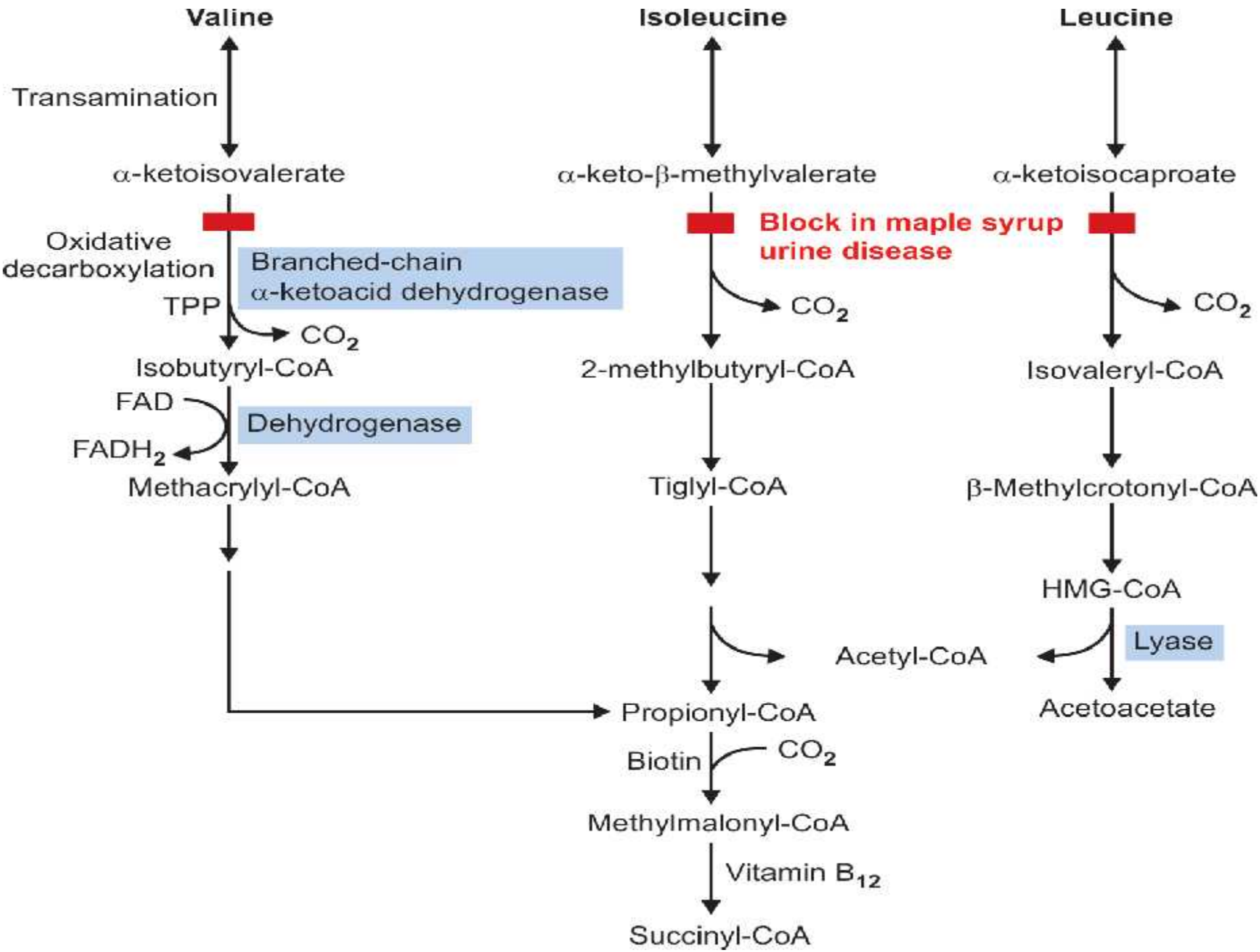
Glycinuria

- It is an inborn error characterized by increased excretion of glycine through urine, despite plasma concentration of glycine is normal.
- Since plasma levels are normal, glycinuria occurs probably due to a *defect in renal tubular reabsorption of glycine.*
- Glycinuria is characterized by increased tendency for the formation of oxalate renal stones.

Metabolic Disorder of Branched Chain Amino Acids

Maple syrup urine disease (MSUD) or branched chain keto aciduria

- Inherited defect in the branched chain α-keto acid dehydrogenase.
- Branched chain amino acids, leucine, isoleucine and valine, and their α -keto acids accumulate in blood, urine and CSF.
- α -keto acids impart a characteristic sweet odour to the urine of the affected individuals which resembles with maple syrup or burnt sugar hence the name.


Valine
Isoleucine
Leucine
Transamination
α-ketoisovalerate
α-keto-β-methylvalerate
α-ketoisocaproate
Block in maple syrup urine disease
Oxidative decarboxylation
Branched-chain α-ketoacid dehydrogenase
TPP
CO2
CO2
CO2
Isobutyryl-CoA
2-methylbutyryl-CoA
Isovaleryl-CoA
FAD
FADH2
Dehydrogenase
Methacrylyl-CoA
Tiglyl-CoA
β-Methylcrotonyl-CoA
HMG-CoA
Lyase
Acetyl-CoA
Acetoacetate
Propionyl-CoA
Biotin
CO2
Methylmalonyl-CoA
Vitamin B12
Succinyl-CoA

Symptoms

- Vomiting
- dehydration
- metabolic acidosis
- maple syrup odour to the urine.
- If untreated, it leads to mental retardation, coma and even death within one year after birth.

Diagnosis

- Many infants with MSUD are identified through newborn screening programs.
- **Tandem mass spectrometry**, an advanced newborn screening test that screens for more than 30 different disorders through one blood sample, has aided in the diagnosis of MSUD.

Treatment

- Treatment involves replacing dietary protein by mixture of amino acids that contain low or no leucine, isoleucine, and valine.
- To monitor the effectiveness of the dietary treatment, plasma and urinary levels of branched-chain amino acids with dinitrophenylhydrazine (DNPH) should be measured constantly.

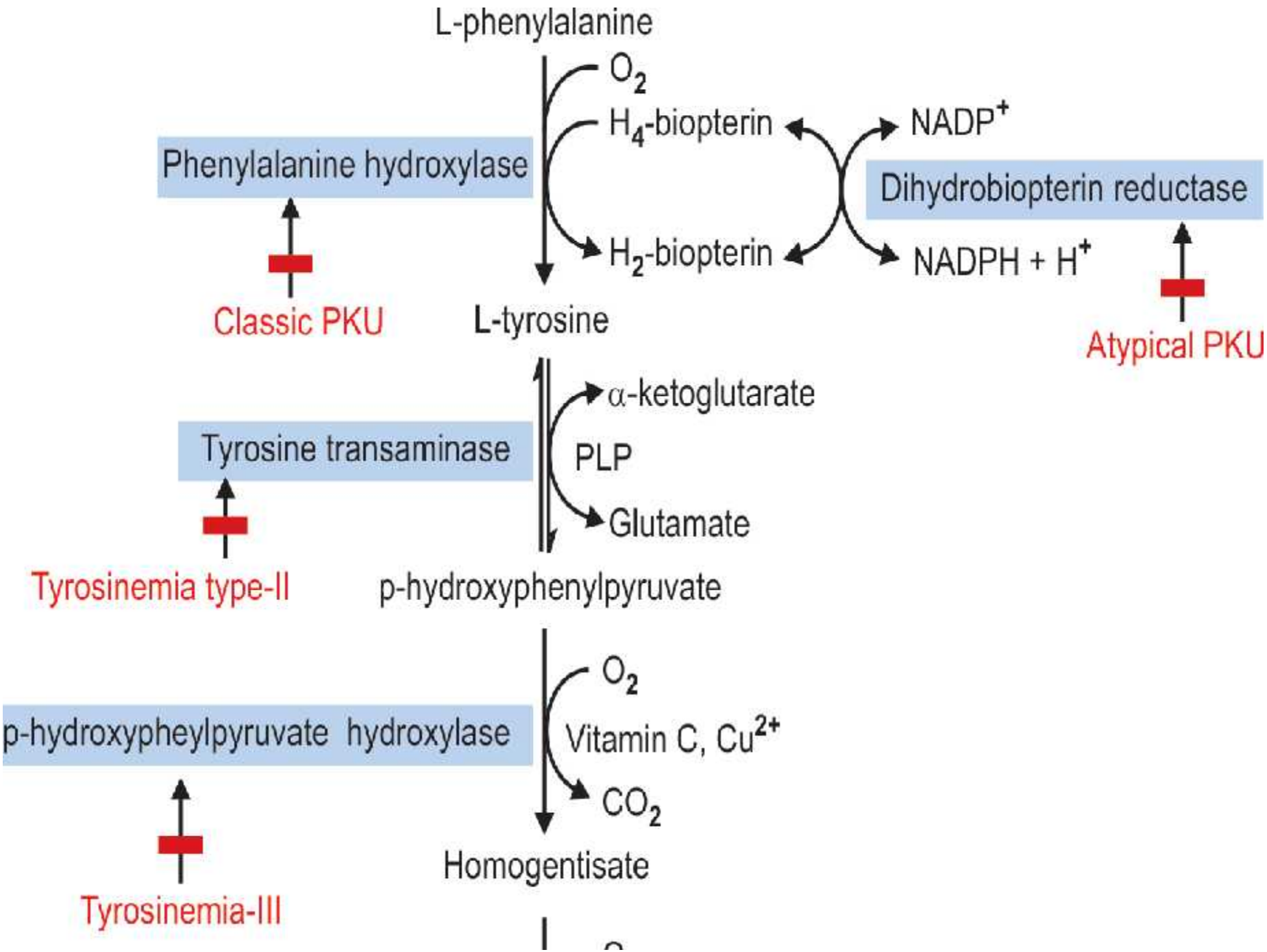
Metabolism of Phenylalanine &Tyrosine

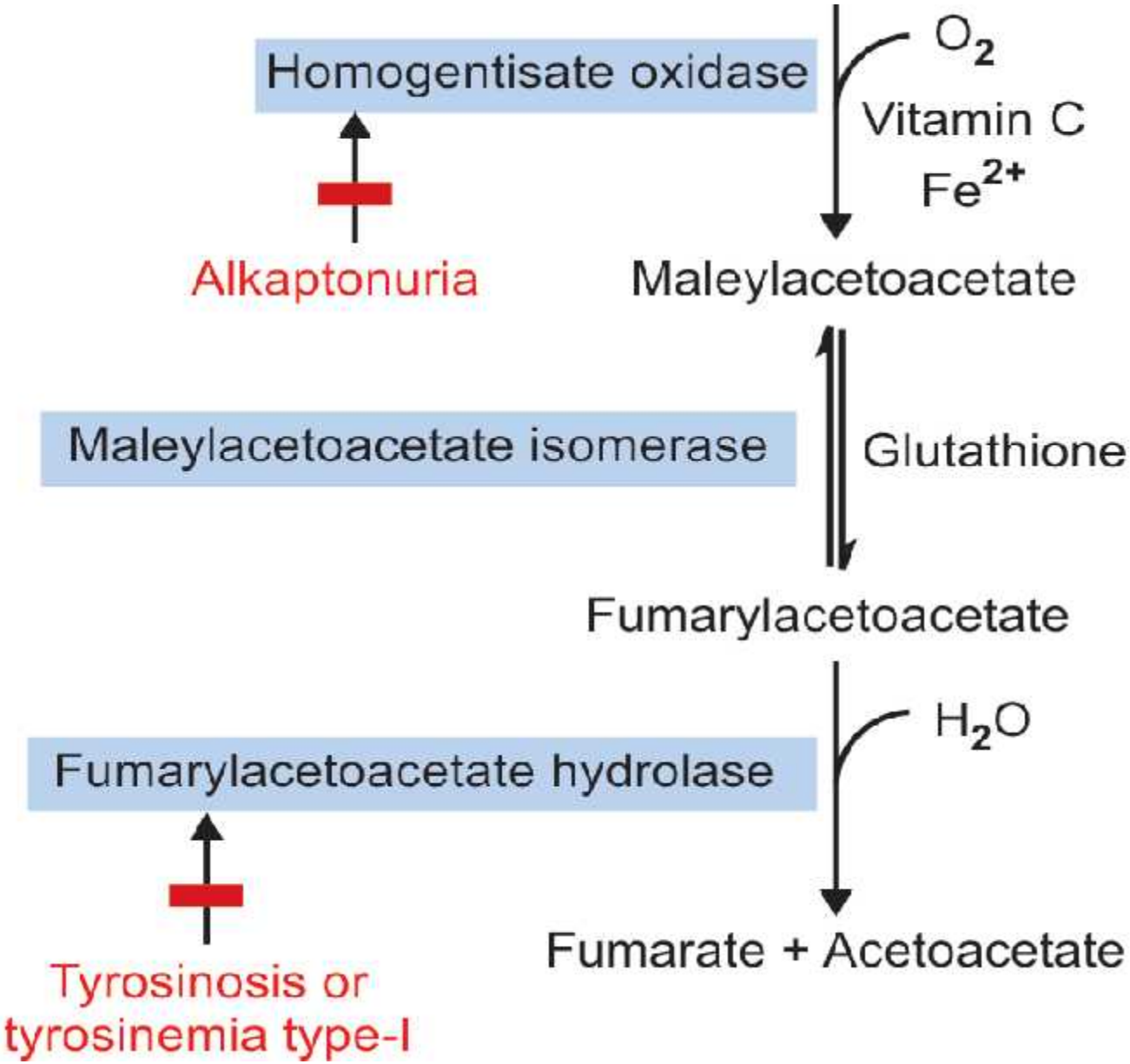
- Phenylalanine and tyrosine are the aromatic amino acids.

- Phenylalanine is nutritionally essential amino acids but tyrosine is not as it can be synthesized from phenylalanine.

Catabolism of Phenylalanine and Tyrosine

- Phenylalanine metabolism is initiated by its oxidation to tyrosine which then undergoes oxidative degradation.
- Thus, catabolic pathway for phenylalanine and tyrosine is same.

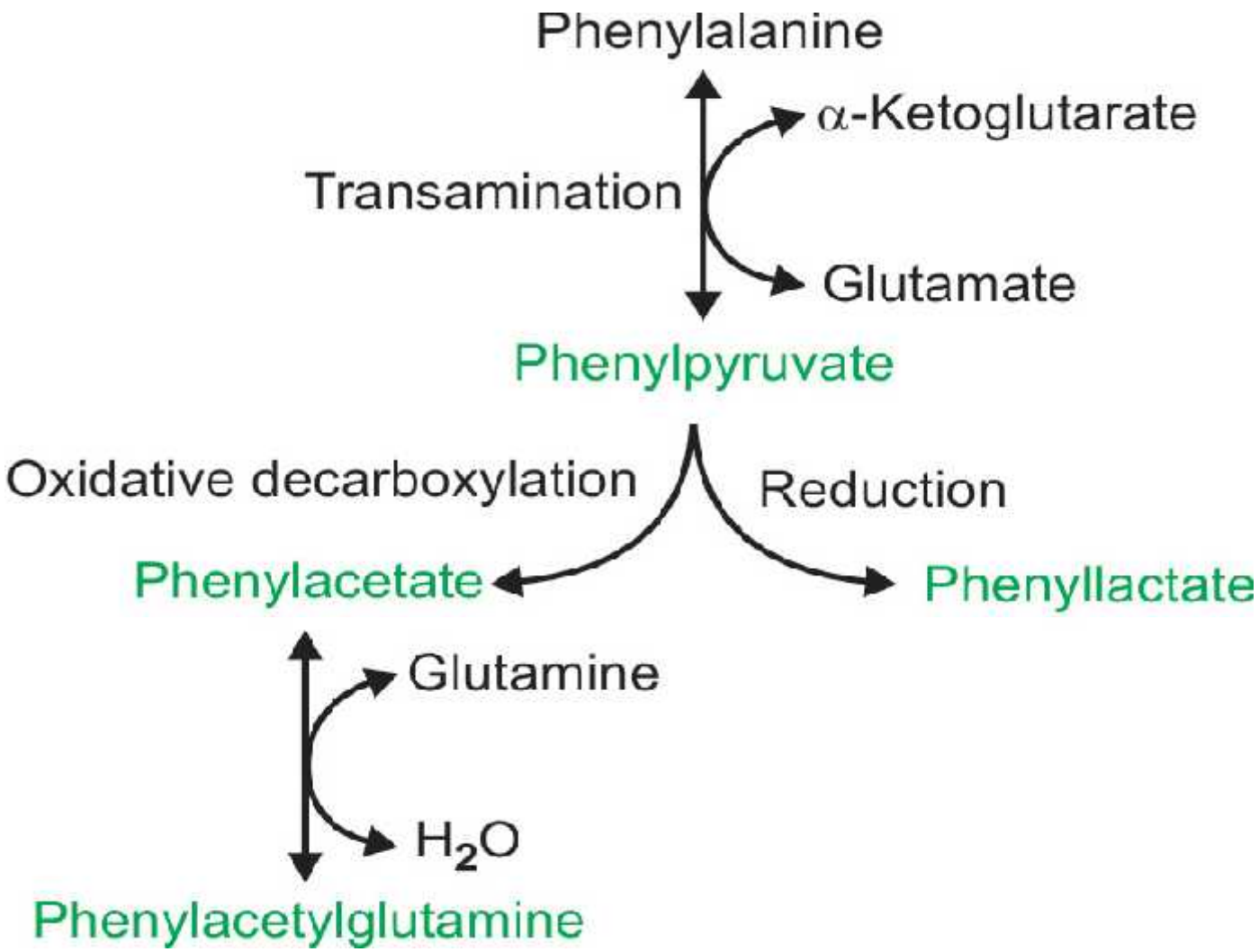

L-phenylalanine
O_2
H_4-biopterin
$NADP^+$
Phenylalanine hydroxylase
Dihydrobiopterin reductase
H_2-biopterin
$NADPH + H^+$
Classic PKU
L-tyrosine
Atypical PKU
α-ketoglutarate
Tyrosine transaminase
PLP
Glutamate
Tyrosinemia type-II
p-hydroxyphenylpyruvate
O_2
p-hydroxypheylpyruvate hydroxylase
Vitamin C, Cu^{2+}
CO_2
Homogentisate
Tyrosinemia-III


Homogentisate oxidase
O_2
Vitamin C
Fe^{2+}
Alkaptonuria
Maleylacetoacetate
Maleylacetoacetate isomerase
Glutathione
Fumarylacetoacetate
H_2O
Fumarylacetoacetate hydrolase
Fumarate + Acetoacetate
Tyrosinosis or
tyrosinemia type-I

Metabolic Disorders of Phenylalanine and Tyrosine

Phenylketonuria (PKU)

- Inborn error of phenylalanine metabolism, associated with the inability to convert phenylalanine to tyrosine.
- Result in accumulation of phenylalanine in tissues and blood and its increased excretion in urine.
- Accumulation of toxic metabolites of phenylalanine such as, *phenylpyruvate, phenylacetate, phenyllactate* and *phenylacetyl glutamine* occurs.



Alternative pathways of phenylalanine catabolism in phenylketonurics.

Characteristics of PKU

- In tissues, plasma and urine increased level of:

  Phenylalanine,

  phenylacetate,

  phenyl-lactate,

  phenylpyruvate

  phenylacetylglutamine,.

- Phenylacetate gives the urine a mousy odour.

- Neurological symptoms: Mental retardation, failure to walk, to talk, seizures, psychoses, tremor and failure to grow.
- Hypopigmentation: Phenylketonurics have a lighter skin colour, fair hair and blue eyes due to deficiency of pigment melanin.
- The hydroxylation of tyrosine by *tyrosinase* is the first step in the formation of the pigment melanin is competitively inhibited by the high levels of phenylalanine in PKU

Treatment of PKU

Treatment due to Defect in Phenylalanine Hydroxylase

- Low phenylalanine diet supplemented with tyrosine, because tyrosine is normally synthesized from phenylalanine.
- The aim is to provide just enough phenylalanine to meet the needs for growth and replacement.

- Proteins that have a low content of phenylalanine, such as casein from milk are hydrolyzed and phenylalanine is removed by adsorption.
- A low phenylalanine diet must be started very soon after birth to prevent irreversible brain damage.

Diagnostic Tests for PKU

- In the past years, the urine of newborns was assayed by the addition of $FeCl_3$ which gives an olive color in the presence of phenylpyruvate.
- The phenylalanine level in blood is detected by screening by using Guthrie test; a bacterial assay for phenylalanine is more reliable diagnostic test.
- The gene for human phenylalanine hydroxylase has been cloned, so that prenatal diagnosis of PKU is now possible with DNA probes.

Tyrosinemia type-I

- Tyrosinemia type-I, also called tyrosinosis

- Caused by a genetic deficiency of fumarylacetoacetate hydroxylase.

- Results in accumulation and excretion of tyrosine and its metabolites:

  P- hydroxyphenyl-pyruvate,
  P- hydroxyphenyl-lactate
  P-hydroxyphenyl-acetate,
  N- acetyltyrosin
  Tyramine.

- The deficiency of enzyme fumarylacetoacetate hydroxylase causes liver failure, kidney dysfunction, polyneuropathy, and vitamin D-resistant rickets.

Clinical Features

- In acute tyrosinosis, the infant exhibits diarrhea, vomiting, and cabbage-like odor to skin and urine due to **succinylacetone** an abnormal metabolite, derived from fumarylacetoacetate.
- Death may occur in infancy due to acute liver failure (within first year of life).
- Whereas in chronic tyrosinosis, in later life, it develops liver cirrhosis and death that occurs by age 10 years

Diagnosis

- A diagnosis of tyrosinemia type I may be suspected in infants who display failure to thrive and an enlarged liver (hepatomegaly) during the first three months of life.
- The diagnosis is expected when tyrosine metabolites and succinylacetone are detected in the urine.
- It is also possible to make the diagnosis based on

decreased activity of **fumarylacetoacetate hydroxylase** (FAH) in liver tissue but this test is not readily available.

- Molecular genetic testing for *FAH* gene mutations is available to confirm the diagnosis.
- Tyrosinemia type-I may also be diagnosed through newborn screening programs. Succinylacetone can be measured on the newborn blood spot by tandem mass spectroscopy.
- Carrier testing and prenatal diagnosis by DNA analysis are available if the specific gene-causing mutation has been identified in the family. Prenatal diagnosis is also possible by detection of succinylacetone in amniotic fluid.

Treatment

The patient should be kept on diet low in phenylalanine and tyrosine.

Tyrosinemia Type-II (Richner-Hanhart Syndrome)

Cause

Tyrosinemia type-II is caused by genetic deficiency of hepatic enzyme tyrosine aminotransferase (tyrosine transaminase).

Clinical features

- tyrosine and its toxic metabolites accumulates in blood and tissues and appears in urine.
- The accumulation of tyrosine produces lesions in eye and skin and causes mental retardation

Treatment

- Diet low in tyrosine and phenylalanine is recommended.
- Diet with vitamin C may benefit the corneal and skin lesions of tyrosine aminotransferase deficiency, but not the mental retardation.

Tyrosinemia Type-III (Neonatal Tyrosinemia)

- Caused by absence of the enzyme P- hydroxyphenyl-pyruvate hydroxylase
- Serum tyrosine levels are high in premature infants resulting from an immature liver and its limited ability to synthesize the enzyme, p-hydroxyphenyl-pyruvate hydroxylase.

- As the liver matures, the accumulated tyrosine is metabolized and serum levels decrease within 4 to 8 weeks of age.

- It is benign condition and responds well to ascorbic acid.

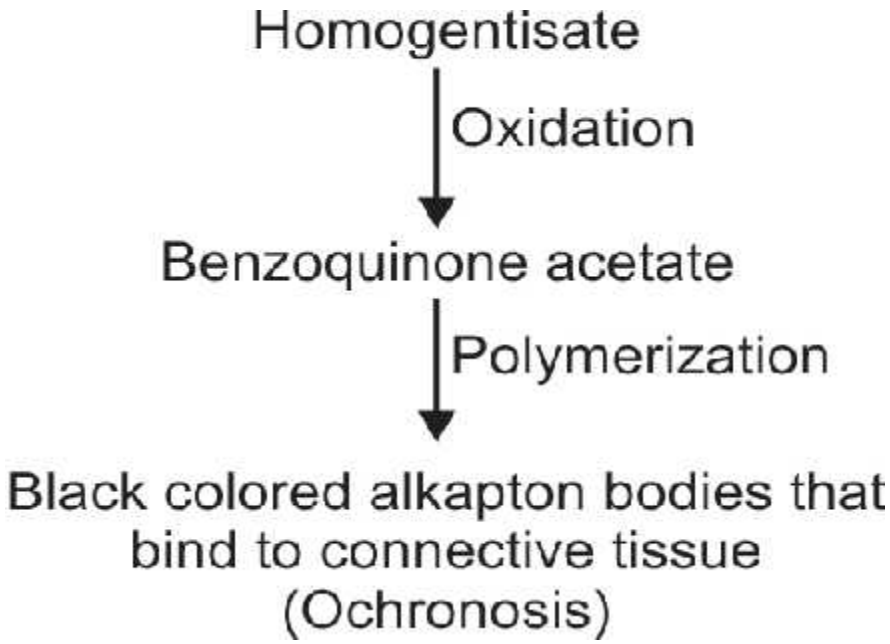
Alkaptonuria

Cause

- Defect in the enzyme *homogentisate* oxidase, that catalyzes oxidation of homogentisate
- Homogentisate accumulates in blood and body tissues and is excreted in large amounts in urine.

Clinical features

The urine of alkaptonuric patients becomes dark after being exposed to air



Formation of alkapton bodies

- The alkapton imparts a characteristic black-brown color to urine.

- Alkaptonuria is a harmless condition.

- Later in life deposition of dark colored alkapton pigments in connective tissues and bones occur.

- This results in black pigmentation of the sclera, ear, nose and cheeks and the clinical condition is known as ochronosis (because ochre color of the deposit).

- Ochronosis leads to tissue damage and may develop joint pain, arthritis and backache.

Treatment

Since alkaptonuria is not considered life threatening, this condition is not treated. Later in life, the symptoms of arthritis may be treated but the condition itself is not.

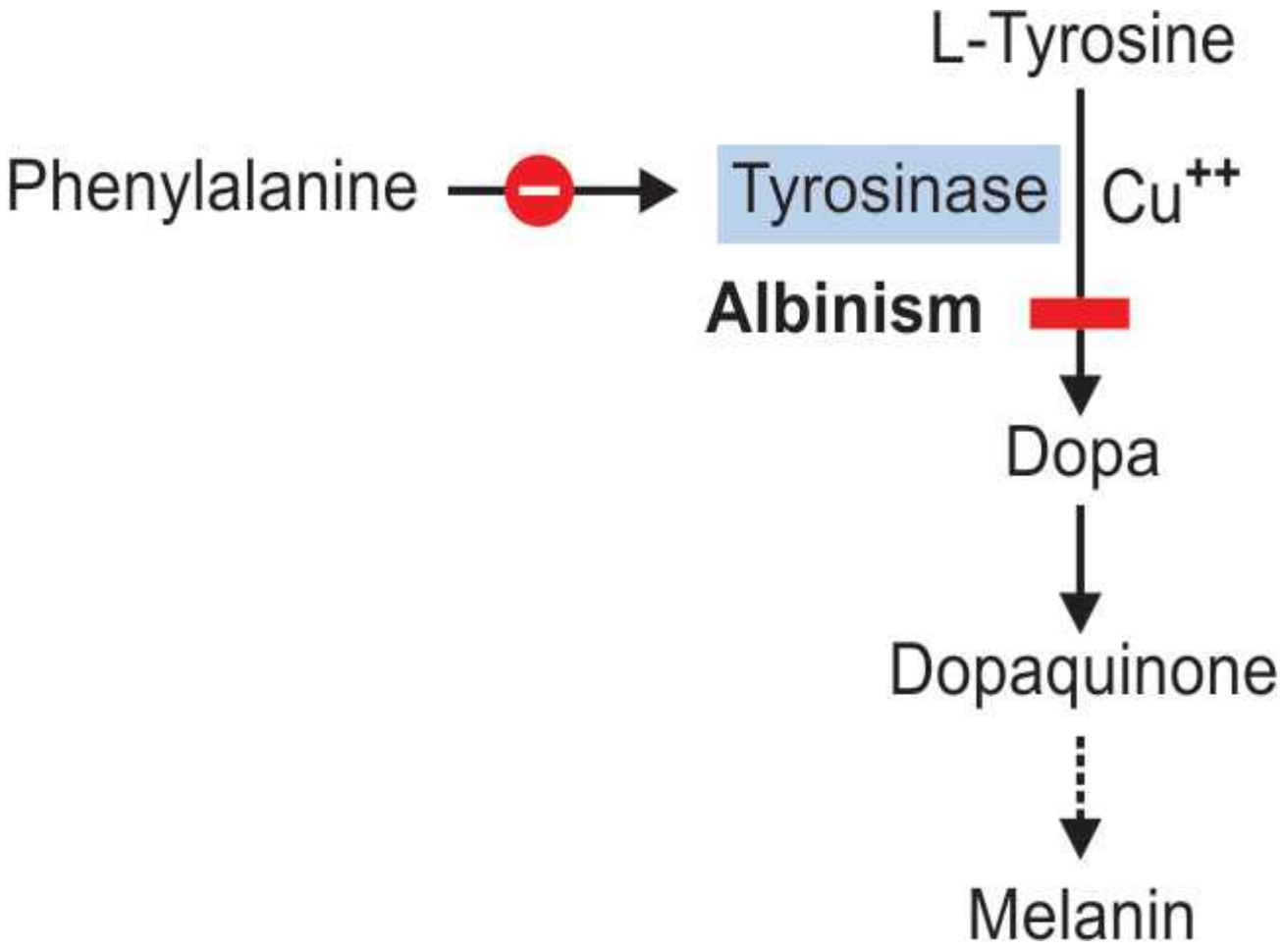
Diagnosis

The urine sample of patients of alkaptonuria turns dark on standing in air. The urine gives positive test with ferric chloride and silver nitrate due to reducing activity of homogentisat**e.**

Albinism

Cause

- Inborn error of tyrosine catabolism.
- It is due to inherited deficiency of enzyme tyrosinase which impairs the synthesis of melanin from tyrosine.
- Melanin protects the body from harmful radiation of sunlight.



Biosynthesis of melanin.

Clinical features

- Impaired synthesis of melanin results in either hypopigmentation or no pigmentation of hair, skin and eyes and leads to white hair and skin.
- Albinos are highly sensitive to sunlight and can lead to skin cancer.

- The lack of melanin pigment in eyes is responsible for photophobia (intolerance to light) and nysta**gmus (rapid involuntary oscillation of the eyeballs)**

Biologically Important Compounds Derived from Tyrosine

Tyrosine serves as a precursor for following several biologically important compounds :

1. Catecholamine
   - Dopamine
   - Norepinephrine
   - Epinephrine
2. Melanin pigment
3. Thyroxine.

METABOLISM OF TRYPTOPHAN

- **Tryptophan is an essential amino acid, containing *indol ring*.**

- **Tryptophan is a precursor for the synthesis of:**

  - Vitamin niacin (vitamin B_3)

  - Neurotransmitter serotonin

  - Hormone melatonin.

For every 60 mg of tryptophan, 1 mg equivalent of niacin can be generated.

Functions of serotonin

- Serotonin is a neurotransmitter and stimulates cerebral activity.
- Serotonin deficiency causes a decrease in cerebral (brain) activity, which leads to depression.
- Serotonin is involved in a variety of behavioural patterns, including sleep, body temperature and blood pressure.

- Serotonin produced in intestinal cells stimulates the release of gastrointestinal peptide hormones.
- Serotonin serves as precursor of melatonin in the pineal gland.
- Serotonin is also a powerful vasoconstrictor and stimulator of smooth muscle contraction.

Melatonin

- Melatonin is a hormone produced from serotonin by the pineal gland
- Synthesis of melatonin is regulated by **light-dark cycle**.
- It is synthesized mostly at night.
- It is an inhibitor of **melanocyte-stimulating hormone (MSH)** and **adrenocorticotropic hormone (ACTH)**.

- Melatonin is a sleep-inducing substance and is involved in regulation of circadian rhythm of body. It may also be involved in regulating **reproductive functions**.

Metabolic Disorder of Tryptophan

Hartnup's disease

- Inherited disorder of tryptophan metabolism.
- This disorder was first of all reported in the family of Hartnup
- Defect in the intestinal and renal transport of tryptophan and other neutral amino acids and leads to tryptophan deficiency.

- Tryptophan deficiency leads to decreased synthesis of vitamin niacin and serotonin.
- Decreased synthesis of niacin leads to pellagra like symptoms
- and decreased serotonin synthesis is responsible for neurological symptoms
- There is amino aciduria due to failure of transport of amino acids from kidney.
- This causes increased urinary loss of alanine, serine, threonine, asparagine, glutamine, leucine, isoleucine, phenylalanine, tyrosine, tryptophan, histidine, glycine, and citrulline.

- Unabsorbed tryptophan remaining in the intestine is metabolized by intestinal bacteria to indolacetic acid and indolpyruvic acid which are subsequently excreted in urine.

Clinical Features

Hartnup disease causes:

- An intermittent photosensitive red scaly rash resembling the rash of pellagra
- Severe but reversible cerebellar ataxia
- Headache
- Muscle discomfort and
- Occasionally psychological disturbances
- Generalized neutral aminoaciduria

Diagnosis

- Due to the variability of symptoms, unambiguous diagnosis can only be made through urine analysis.
- The test is based on the detection of elevated amino acids in the urine by chromatography.
- There is increased urinary excretion of **indolacetic acid** and **indolpyruvic acid** and other indole derivatives.
- Molecular genetic testing can confirm a diagnosis of Hartnup disease in some cases. Molecular genetic testing can detect genetic alterations in the *SLC19A6*gene known to cause the disorder, but usually is not necessary to obtain a diagnosis.

Therapy

Oral nicotinic acid supplementation will permit adequate synthesis of NAD^+ in patients with Hartnup disease. This corrects pellagra like symptoms of the disorder. The aminoaciduria remains unaltered.

Metabolism Of Sulfur Containing amino Acids

Three sulphur containing amino acids are:

Cystine

Cysteine

Methionine

- Methionine is an essential whereas cysteine and cystine are nonessential amino acids.
- Cysteine and cystine are synthesized from two amino acids, methionine, and serine
- Cystine and cysteine are readily inter-convertible in the body.

Metabolism Of Methionine

Methionine is metabolized by:

1. Transfer of methyl group of methionine by

   Transmethylation reactions

2. Conversion of demethylated portion of the methionine

   to cysteine and cystine.

Transfer of Methyl Group of Methionine

(Transmethylation reactions)

- Transfer of methyl group (-CH_3) from methionine to an acceptor molecule is termed as transmethylation.
- The methyl group of methionine becomes available for transmethylation only in an active form of methionine, S- adenosylmethionine (SAM).

Some important transmethylation reactions are:

- Norepinephrine to epinephrine
- Phosphatidylethanolamine to phosphatidylcholine
- Gunaidoacetoacetate to creatine
- Ethanolamine to choline
- Acetyl serotonin to melatonin
- Nucleotides to methylated nucleotides

Conversion of demethylated portion of the methionine to cysteine and cystine

Importance of cysteine

- Glucogenic amino acid involved in formation of glucose.
- Cysteine is most important dietary source of sulphur.
- Physiologically important sulphur containing compounds derived from cysteine are:
  - Taurine
  - *Insulin*
  - *Coenzyme-A*
  - *Glutathione*
  - *Vasopressin*
- It is also involved in detoxification mechanisms.

Metabolic Disorders of Sulphur Containing Amino Acids

Cystinuria (Cystin-lysinuria)

- Cystinuria is the most common inborn error of amino acid transport.
- Cystinuria is an inherited disorder in which kidney tubules fail to reabsorb the amino acids cystine, ornithine, arginine and lysine (the mnemonic is COAL).
- This is characterized by massive urinary excretion of cystine, ornithine, arginine and lysine.

Cause

- Normally, these amino acids are filtered by the glomerulus and reabsorbed in the proximal renal tubule by specific carrier proteins.
- In cystinuria defect in this carrier system leads to the excretion of all these four amino acids.

Clinical features

- Since cystine is the least soluble its over excretion often leads to precipitation and formation of cystine calculi (stones) in the renal tubules and leads to obstruction, infection and renal insufficiency in cystinuric patient.

Treatment

- Treatment involves ingestion of large amounts of water, which increases cystine solubility through maintenance of alkaline urine

Cystinosis (cystine storage disease)

Cystinosis is a rare but serious lysosomal disorder.

Cause

- Caused by a defective carrier that transports cystine across the lysosomal membrane from lysosome to the cytosol.

Clinical features

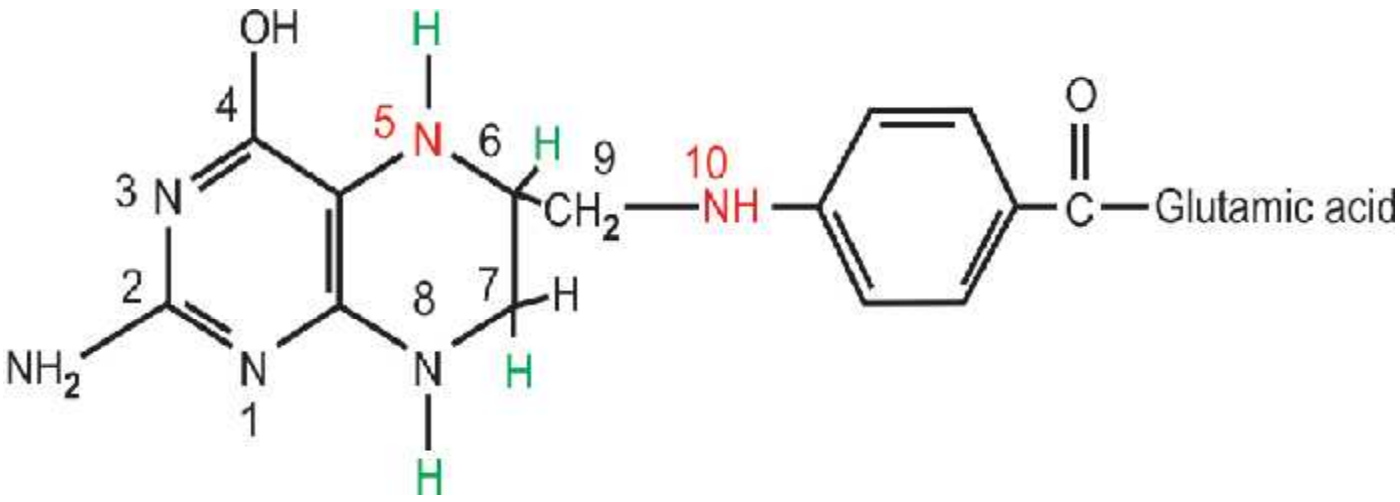
- Cystine accumulates in the lysosomes in many tissues and forms crystals impairing their function.
- Cystinosis is usually accompanied by a generalized aminoaciduria.
- Patients usually die within 10 years due to acute renal failure

ONE CARBON METABOLISM

- Groups, containing a single carbon atoms are called one carbon groups. One carbon groups are formed from several amino acids during their metabolism.

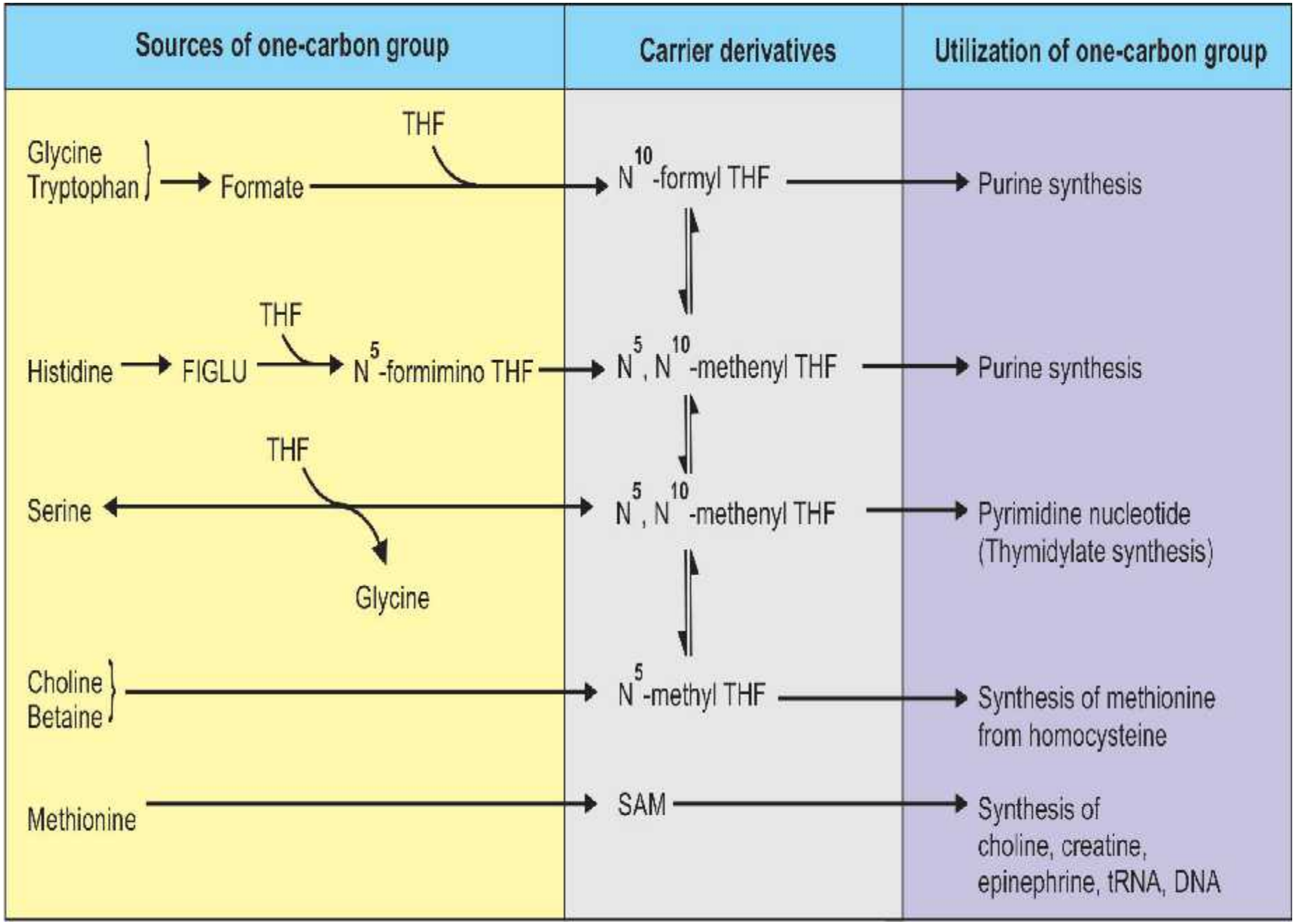
- These include serine, glycine, histidine and tryptophan.

- One carbon groups formed during metabolism are:
  - Methyl (CH_3)
  - Methylene (CH_2)
  - Methenyl (CH)
  - Formyl (CHO)
  - Formimino (CH=NH)
- These one carbon groups are transferred by way of tetrahydrofolate (THF).



Structure of tetrahydrofolate

- One carbon groups carried by THF are attached either to nitrogen N^5 or N^{10} or to both N^5 and N^{10}
- The different one carbon derivatives of THF are
  - N^5-methyl THF
  - N^5, N^{10} -methylene THF
  - N^5, N^{10} -methenyl THF
  - N^5-formyl THF
  - N^5-formimino THF.
- These different derivatives of THF are interconvertible.


Sources of one-carbon group
Carrier derivatives
Utilization of one-carbon group
Glycine
Tryptophan
Formate
THF
N10-formyl THF
Purine synthesis
Histidine
FIGLU
THF
N5-formimino THF
N5, N10-methenyl THF
Purine synthesis
Serine
THF
Glycine
N5, N10-methenyl THF
Pyrimidine nucleotide
(Thymidylate synthesis)
Choline
Betaine
N5-methyl THF
Synthesis of methionine
from homocysteine
Methionine
SAM
Synthesis of
choline, creatine,
epinephrine, tRNA, DNA

Importance of one carbon group metabolism

One carbon groups at different levels of oxidation are transferred and made available by way of the tetrahydrofolate and vitamin B_{12} coenzymes for use in a wide variety of vital anabolic processes.

BIOGENIC AMINES

- Decarboxylation of amino acids results in the formation of amines. These amines are called biogenic amines. They have diverse biological function.
- Decarboxylation reactions are catalyzed by PLP dependent decarboxy- lases.

Some important biogenic amines and their functions.		
Amines	***Amino acid precursors***	***Functions***
Dopamine	Tyrosine	Neurotransmitter
Norepinephrine	Tyrosine	Neurotransmitter
Epinephrine	Tyrosine	Hormone
Tyramine	Tyrosine	Vasoconstrictor
Serotonin	Tryptophan	Vasoconstrictor
Melatonin	Tryptophan	Vasoconstrictor
GABA	Glutamate	Neurotransmitter
Histamine	Histidine	Vasodilator
Taurine	Cysteine	Neurotransmitter
Spermine	Ornithine and methionine	Growth factor, regulator of transcription and translation

Polyamines

- Biological amines made up of multiple amino acids called polyamines, e.g.
  - Putrescine
  - Spermidine
  - Spermine.
- Polyamines are positively charged at physiological pH and associate with negatively charged nuclear DNA.
- These are present in high concentration in semen. The concentration of polyamines in brain is about 2 mM.

- *Putrescine*, *spermi*dine and spermine are derived from ornithine and methionine.
- It is presumed that the 15% of methionine is used for polyamine synthesis.
- Polyamines are involved in regulation of transcription and translation.

- They act as a growth factor and function in cell proliferation and growth.
- Polyamines are involved in stabilization of intact cells, subcellular organelles and membranes.

Clinical significance of polyamines

- Polyamines and their derivatives have application in diagnosis and treatment of cancer.
- Their levels have been shown to increase in response to cell growth and differentiation.
- Their concentration is elevated in body fluids of cancer patients.
- Assays of urinary and blood polyamines have been used to detect cancer and to determine the success of therapy (diagnostic indicator).

THANK YOU