AMINO ACID METABOLISM

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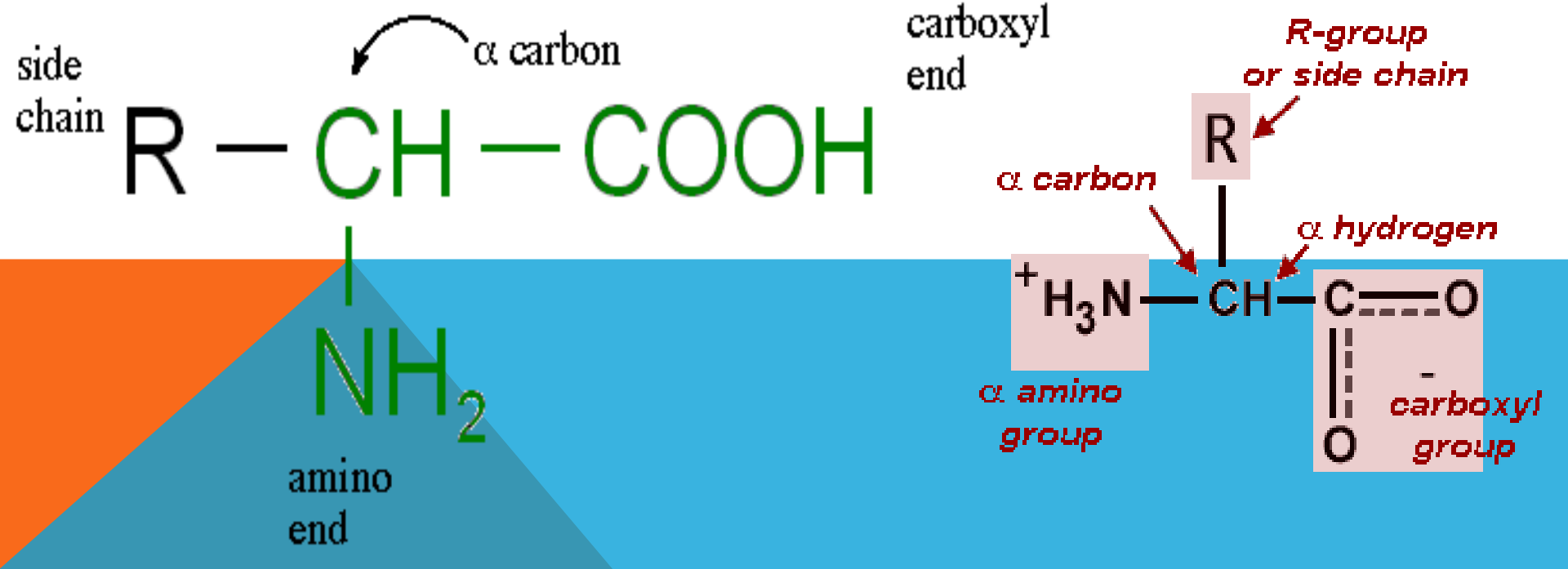
OBJECTIVES

- Digestion and absorption of proteins and amino acids
- Introduction to amino acids, structure and types
- Amino acid and nutrition
- General and individual Amino acid metabolism; and inborn errors of metabolism
- Metabolism of ammonia
- Clinical significance of amino acid and ammonia metabolism

WHAT IS AMINO ACID?

Amino acids are derivatives of carboxylic acids formed by substitution of α-hydrogen for amino functional group



WHAT DO AMINO ACIDS DO?

- **Amino acids are essential to life, have a role in metabolism, and are important in nutrition.**
- **They form short polymer chains called peptides, as well as longer chains that are called polypeptides or proteins.**
- **About 75 percent of the human body is made up of chains of amino acids, which is why they are so vital to how your system functions.**
- **All the chemical reactions that occur in the body depend on amino acids and the proteins they build.**

TYPES OF AMINO ACIDS

Amino acids are classified as

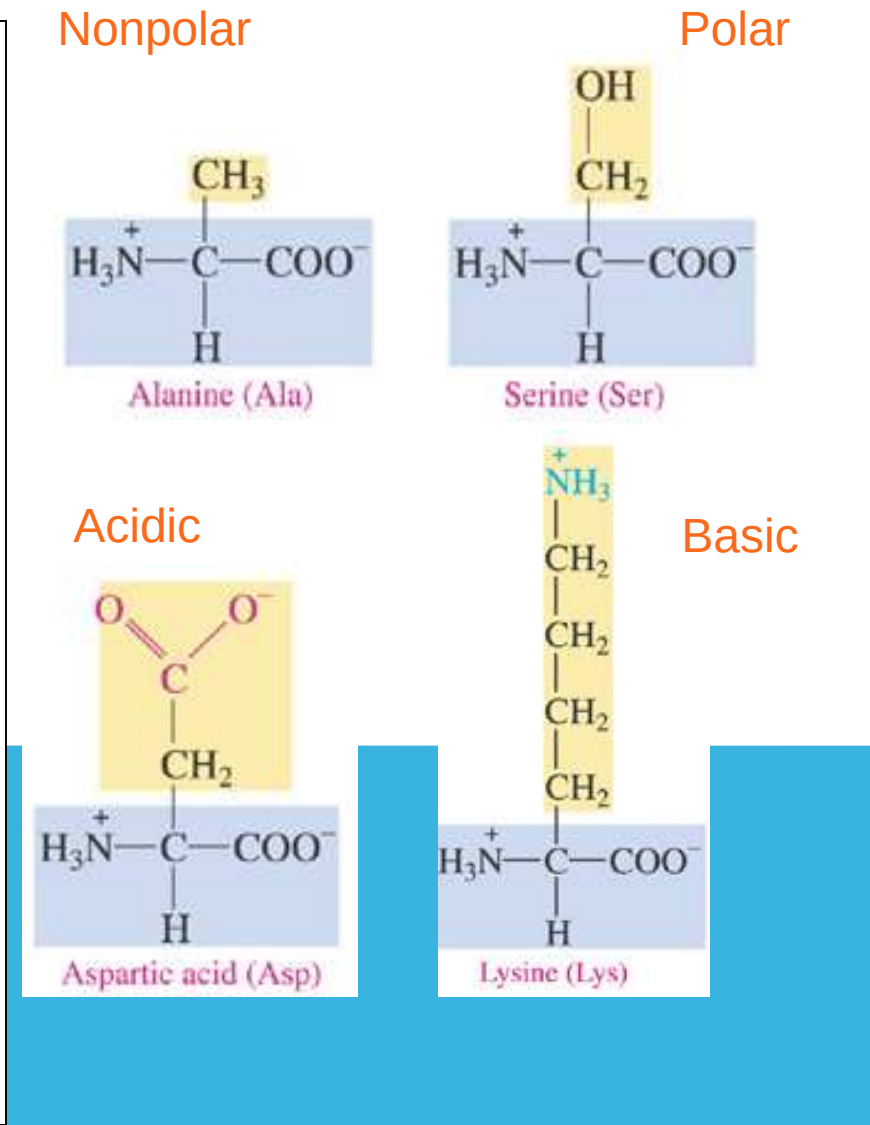
Nonpolar (hydrophobic) with hydrocarbon side chains.

Polar (hydrophilic) with polar or ionic side chains.

Acidic (hydrophilic) with acidic side chains.

Basic (hydrophilic) with

$-NH_2$ side chains.



- **non-essential amino acids**

  - can be synthesized by an organism

  - usually are prepared from precursors in 1-2 steps

- **Essential amino acids**

  - cannot be made endogenously

  - must be supplied in diet

  eg. Leu, Phe…..

Nutritionally-Essential amino acids :

Lysine, Leucine, Isoleucine, Valine, Methionine, Phenylalanine,

Threonine, Tryptophan

Nutritionally Nonessential amino acids: Alanine, glycine, aspartate , glutamate, serine, tyrosine, cysteine, proline , glutamine, aspargine

N.B. Histidine & arginine are semi essential. They are essential only for infants growth, but not for old children or adults where in adults histidine requirement is obtained by intestinal flora & arginine by urea cycle

nutritional value

- Legumes poor in Trp, but rich in Lys;

  Cereals poor in Lys, but rich in Trp

- Mutual complementation of amino acids
- Protein deficiency-kwashiorkor, generalized edema and liver enlargement, abdomen bulged
- **Suggestion: the combined-action of protein in diet**

PROTEIN DIGESTION

Digestive Tract of protein

- Proteins are generally too large to be absorbed by the intestine and therefore must be hydrolyzed to the amino acids
- The proteolytic enzymes responsible for hydrolysis are produced by three different organs: the stomach、pancreas and small intestine (the major organ)

Stomach

- HCl (parietal cells) and Pepsinogen (chief cells)
- The pH of gastric juice is around 1.0. Food is retained in the stomach for 2-4 hrs
- HCl kills microorganisms, denatures proteins, and provides an acid environment for the action of pepsin
- **Autocatalysis**: pepsinogen is converted to active pepsin(*Pepsin A)* by HCl

Pancreas and small intestine

- **Endo**peptidase (pancreas)

  Trypsin: carbonyl of arg and lys

  Chymotrypsin: carbonyl of Trp, Tyr, Phe, Met, Leu

  Elastase: carbonyl of Ala, Gly, Ser

  Exopeptidase (pancreas)

  Carboxypeptidase A:amine side of Ala, Ile, Leu, Val

  Carboxypeptidase B: amine side of Arg, lys

- Aminopeptidase (small intestine):

  cleaves N-terminal residue of oligopeptidaes

PROTEIN ABSORPTION

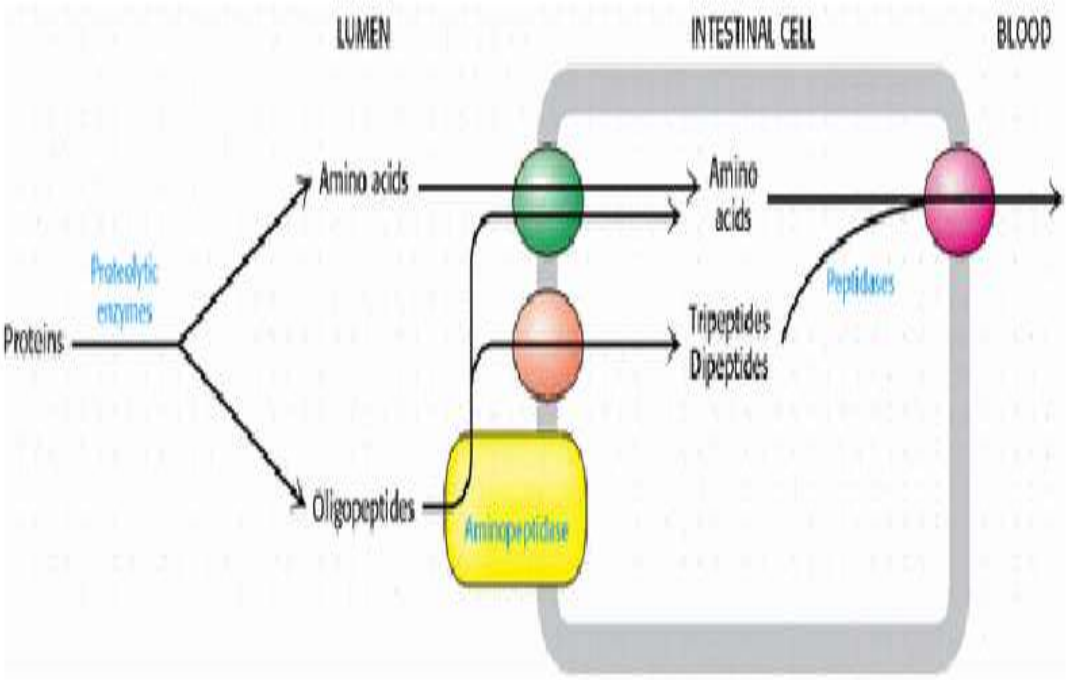
*L-amino acids are actively transported across the intestinal mucosa (need carrier, Na + pump,

Na+ ions, ATP).

Different carrier transport systems are: a) For neut amino acids.
b) For basic amino acid and cysteine.
c) For imino acids and glycine.

d) For acidic amino acids.
e) For B-amino acids (B-alanine & taurine).

*D-isomers transported by simple diffusion.



Nitrogen Balance (NB):

- **Nitrogen balance** is a **comparison between**

  Nitrogen intake (in the form of dietary protein)

  and

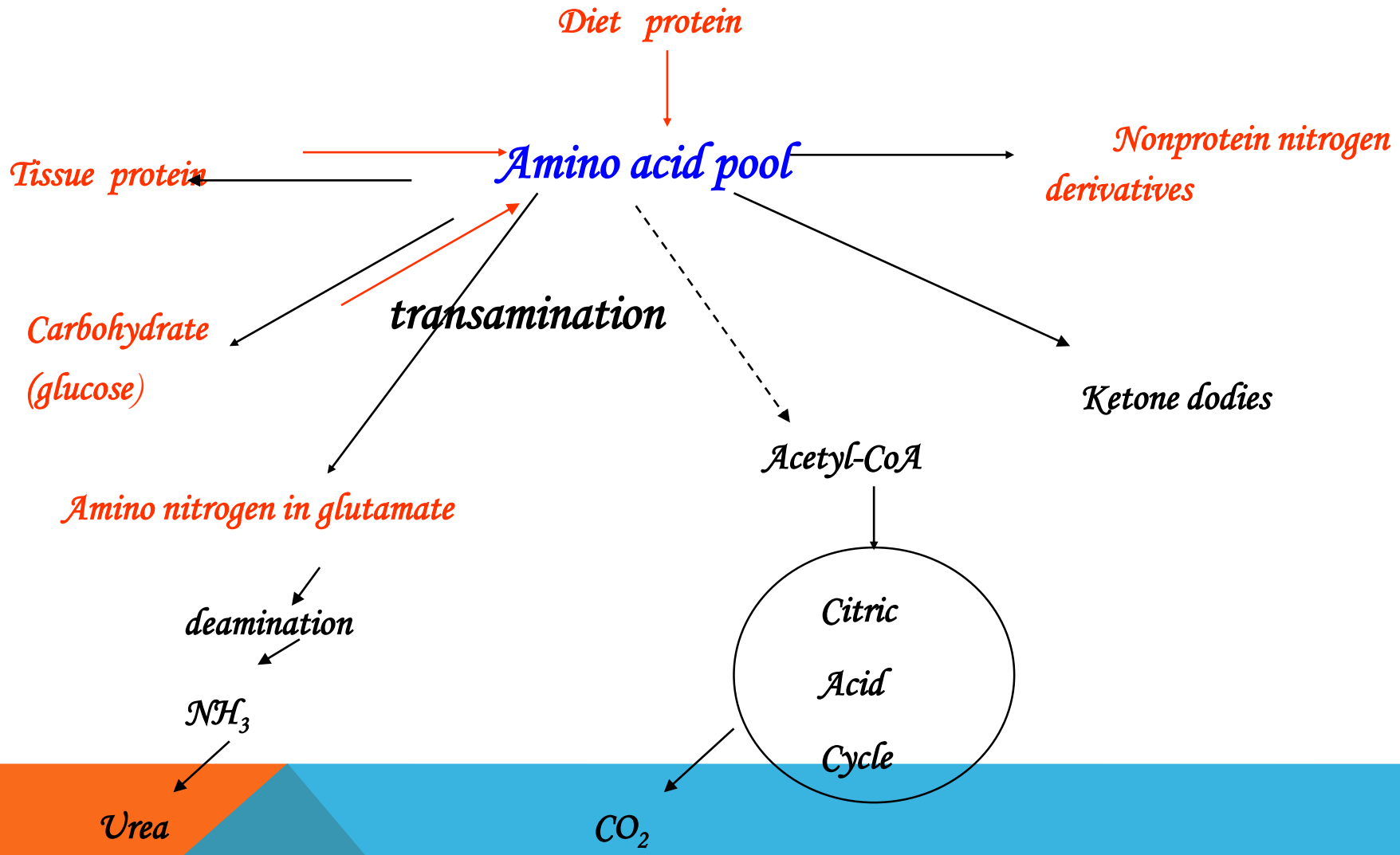
  Nitrogen loss (as **undigested protein** in feces , **NPN** as urea, ammonia, creatinine & uric acid in urine, sweat & saliva & **losses** by hair, nail, skin).

- NB is important **in** defining

  1.overall protein metabolism of an individual

  2.nutritional nitrogen requirement.

AMINO ACID METABOLISM

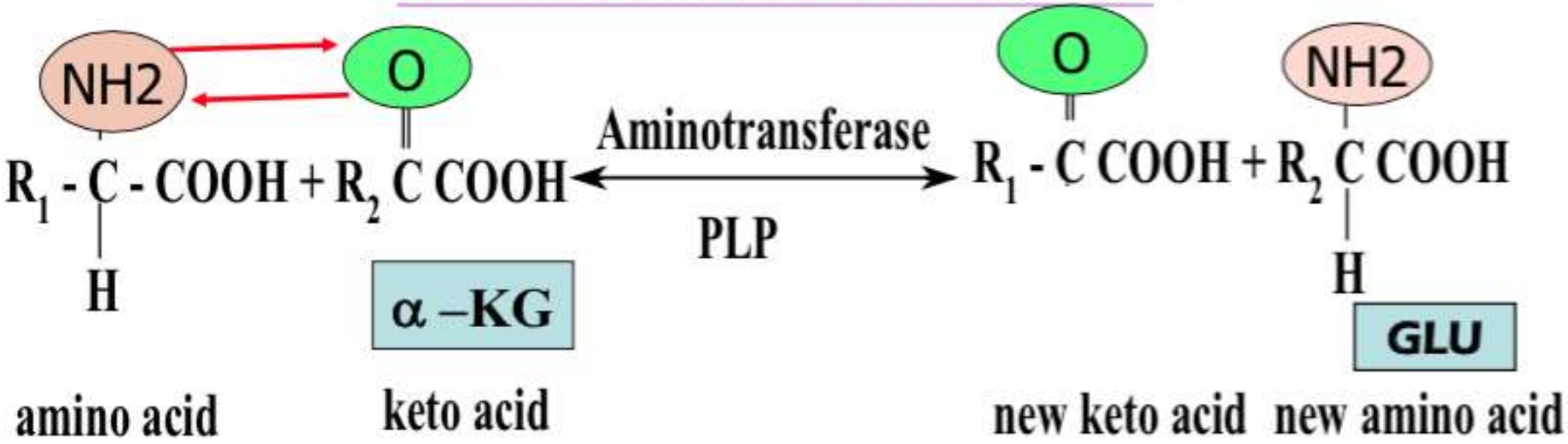


Overview of the protein metabolism

Metabolism OF AMINO ACIDS:

1. **Removal of amonia** by :
   - Deamination
     - **Oxidative deamination**
       1) glutamate dehydrogenase in mitochondria
       2) amino acid oxidase in peroxisomes
     - **Direct deamination (nonoxidative)**
       1) dea. by dehydration ($-H_2O$)
       2) dea. by desulhydration ($-H_2S$)
   - Transamination (GPT & GOT)
   - and transdeamination.
2. **Fate of carbon-skeletons of amino acids**
3. **Metabolism of ammonia**

Transamination:



Aminotransferases are **active** both in cytoplasm and mitochondria e.g.:

1. Aspartate aminotransferase (AST), Glutamate oxaloacetate transaminase **(GOT),**

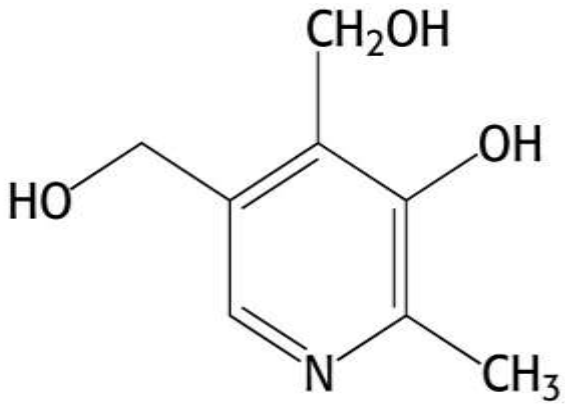
2. Alanine aminotransferase (ALT), Glutamate pyruvate transaminase, **(GPT)**

In all transamination reactions, α-ketoglutarate (α –KG) acts as amino group acceptor.

Most, but not all amino acids undergo transamination reaction with few exceptions **(lysine, threonine and imino acids)**

Mechanism of transamination

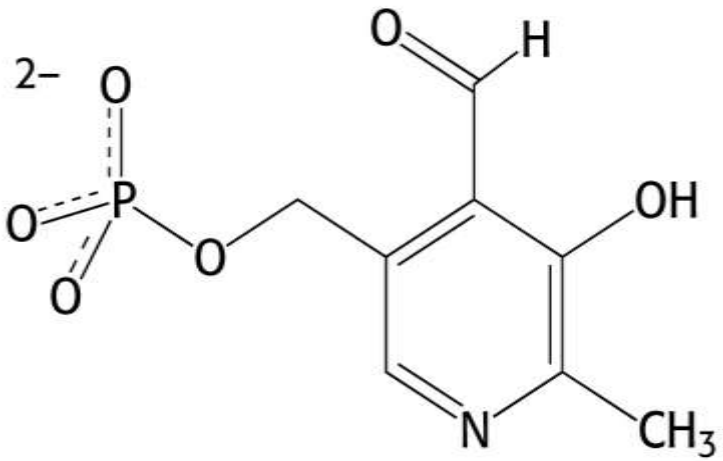
All aminotransferases require the prosthetic group ***pyridoxal phosphate (PLP),*** which is derived from ***pyridoxine (vitamin B_6).***



Pyridoxine (Vitamin B_6)

Ping-pong kinetic mechanism

First step: the amino group of amino acid is transferred to pyridoxal phosphate, forming pyridoxamine phosphate and releasing ketoacid.



Pyridoxal phosphate (PLP)

Second step: α-ketoglutarate reacts with pyridoxamine phosphate forming glutamate

B. Oxidative Deamination

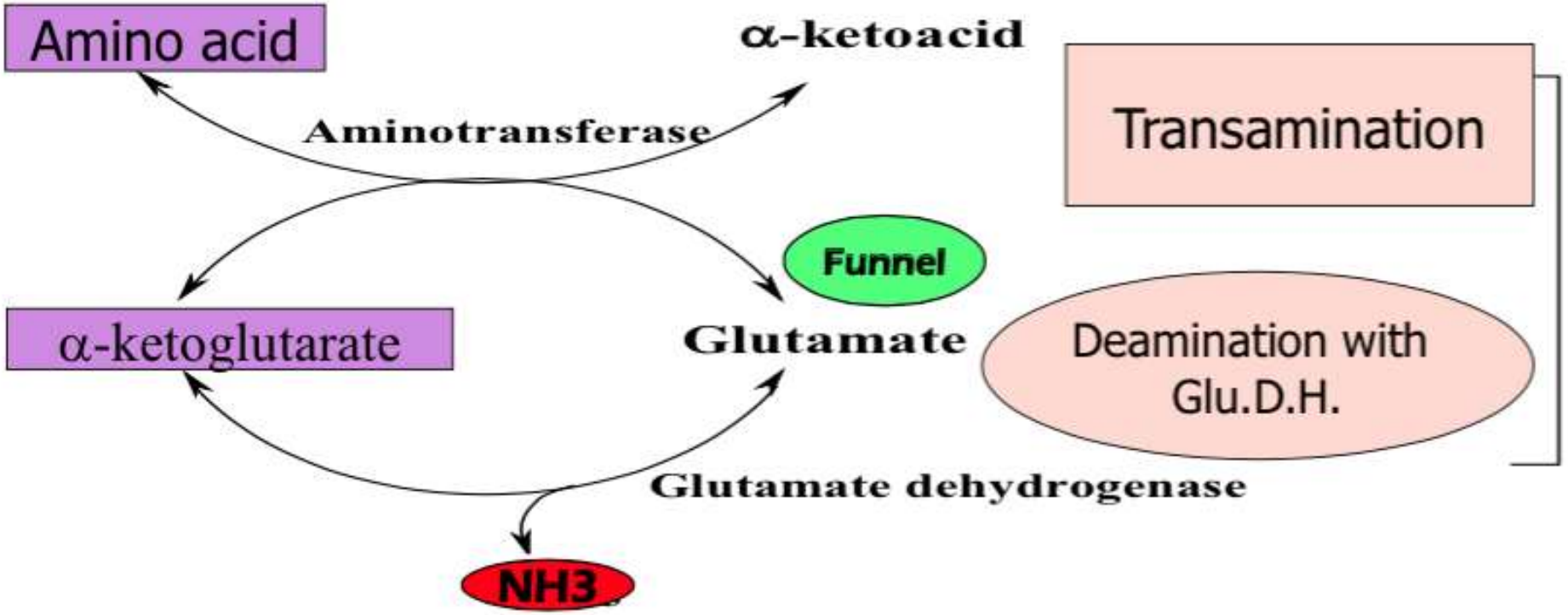
- L-glutamate dehydrogenase (in mitochondria)

Glu + NAD^+ (or $NADP^+$) + H_2O ⇆ NH_4^+ + a-ketoglutarate + NAD(P)H +H^+

Requires NAD^+ or $NADP^+$ as a cofactor

Plays a central role in AA metabolism

Transdeamination:



Due to…L-amino acid oxidases, but not **glutamate dehydrogenase,** can sluggish (decrease) the rate of deamination of the amino acids.

So… the most **important** and **rapid** way to deamination of amino acids **is first** transamination with α-ketoglutarate **followed** by deamination of glutamate.

Therefore **glutamate through transdeamination serves to a funnel ammonia from all amino acids.**

THE FATE OF CARBON-SKELETONS OF AMINO ACIDS

a) Simple degradation:

(amino acid → Common metabolic intermediate)

Alanine → Pyruvate

Glutamate → α-ketoglutarate

Aspartate → Oxaloacetate

b) Complex degradation:

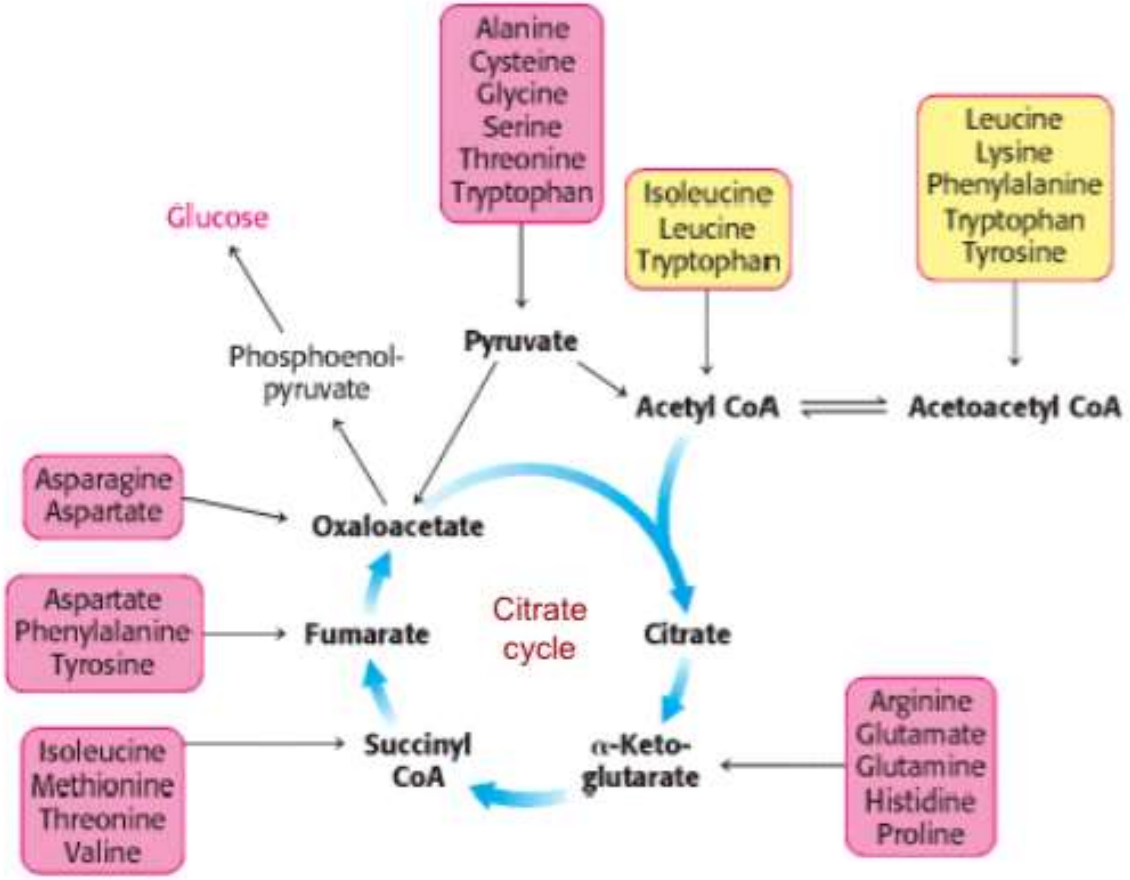
(amino acid--- Keto acid----- **complex** pathway---- Common metabolic intermediate)

Amino acids whose ketoacids are metabolized via **more complex** pathway **e.g. Tyrosine, Lysine, Tryptophan**

c) Conversion of one amino acid into another amino acid before degradation:

Phenylalanine is converted to **tyrosine** prior to its further degradation.

The common metabolic intermediates that arised from the degradations of amino acids are: acetyl CoA, pyruvate, one of the krebs cycle intermediates (α-ketoglutarate, succinyl CoA, fumarate& oxaloacetate)



Fates of the Carbon Skeletons of Amino Acids. Glucogenic amino acids are shaded red, and ketogenic amino acids are shaded yellow. Most amino acids are both glucogenic and ketogenic.

Metabolism of the Common Intermediates

1. ***Oxidation:*** all amino acids can be oxidized in **TCA** cycle with **energy** production
2. ***Fatty acids synthesis:*** some amino acids provide **acetyl CoA** e.g. leucine and lysine (ketogenic amino acids).
3. ***Gluconeogenesis:*** ketoacids derived from amino acids are used for synthesis of **glucose** (is important in starvation).

Glucogenic	***Ketogenic***	***Glucogenic&Ketogenic***
Ala, Ser, Gly, Cys, Arg, His, Pro, Glu, Gln, Val, Met, Asp, Asn.	Leu , Lys	Phe,Tyr,Trp,Ile,Thr

METABOLISM OF AMMONIA

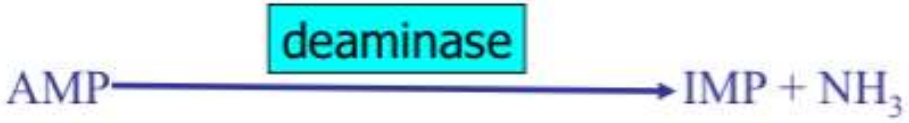
Ammonia is formed in body from:

a) From amino acids: 1.Transdeamination in liver (NOT T.A.)

2.amino acid oxidases and amino acid deaminases in liver and kidney.

b) Deamination of physiological amines: by monoamine oxidase.

c) Deamination of purine nucleotides: especially adenine nucleotides



d) Pyrimidine catabolism.

e) From bacterial action in the intestine **on dietary protein & on urea in the gut.**

NH3 is also produced by glutaminase on glutamine .

Excretion of excess AA (amino group)

MOST MAMMALS CONVERT AMINO-ACID NITROGEN TO UREA FOR EXCRETION

amino acids

NH_4^+

most terrestrial vertebrates

fish & other aquatic vertebrates

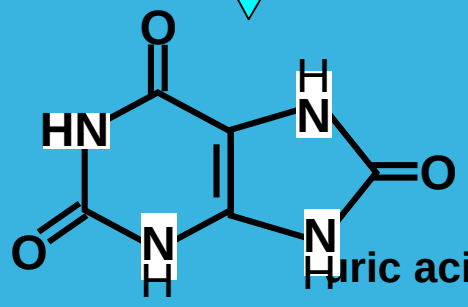
birds & reptiles

$H_2N\text{-}C(=O)\text{-}NH_2$
urea

(ureotelic)

NH_4^+
ammonium ion

(ammonotelic)



uric acid

(uricotelic)[2]

TRANSPORT OF AMMONIA TO THE LIVER

Two mechanism are available for the transport of ammonia from peripheral cells to liver for detoxification

The first uses glutamine synthetase to combine glutamate with ammonia

The second , used primarily by muscle, involves transamination of pyruvate to Alanine

GLUTAMATE AND GLUTAMINE RELATIONSHIP

Ammonia Nitrogen can be transported as glutamine.

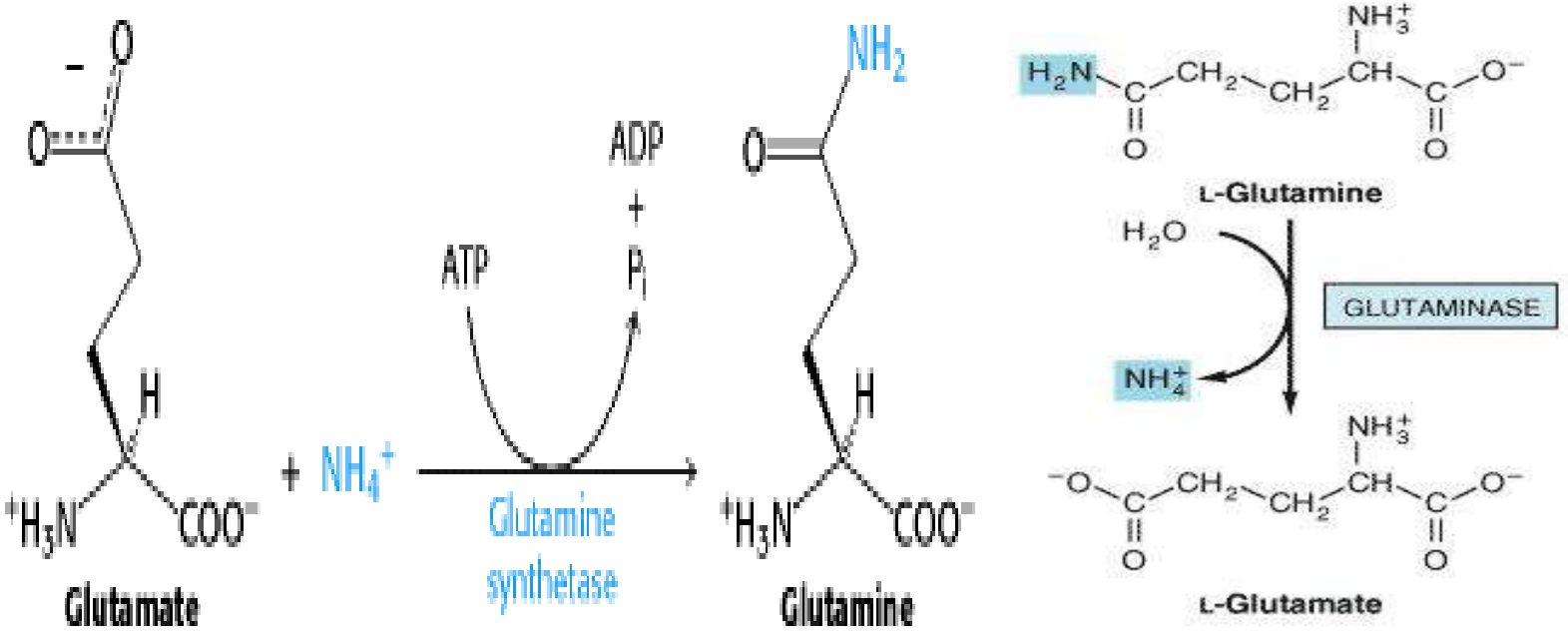
This is the first line of defense in brain cells.

Glutamine synthetase catalyzes the synthesis of glutamine from glutamate and NH4 + in an ATP-dependent reaction

The nitrogen of glutamine can be converted to urea in liver by the action of glutaminase in liver

Hydrolytic release of the amide nitrogen of glutamine as ammonia, catalyzed by glutaminase favors glutamate formation.

GLUTAMATE AND GLUTAMINE RELATIONSHIP



The concerted action of glutamine synthase and glutaminase thus catalyzes the interconversion of free ammonium ion and glutamine

GLUCOSE ALANINE CYCLE AND ROLE OF GLUTAMATE

The transport of amino group of amino acids also takes place in the form of Alanine.

Nitrogen is transported from muscle to the liver in two principal transport forms.

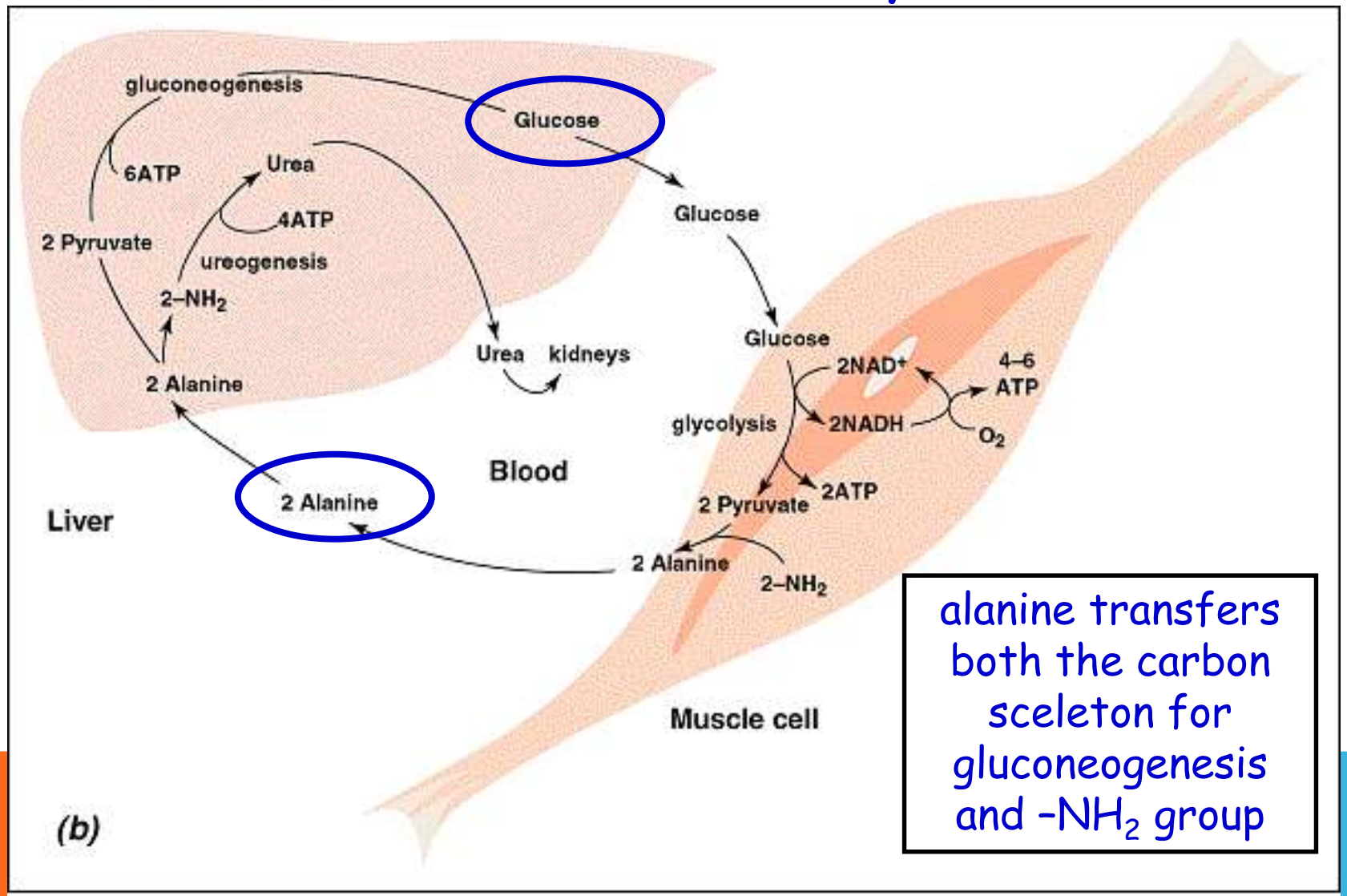
Glutamate is formed by transamination reactions, but the nitrogen is then transferred to pyruvate to form alanine, which is released into the blood.

The liver takes up the alanine and converts it back into pyruvate by transamination.

The pyruvate can be used for gluconeogenesis and the amino group eventually appears as urea.

This transport is referred to as the *alanine cycle.*

Glucose-alanine cycle

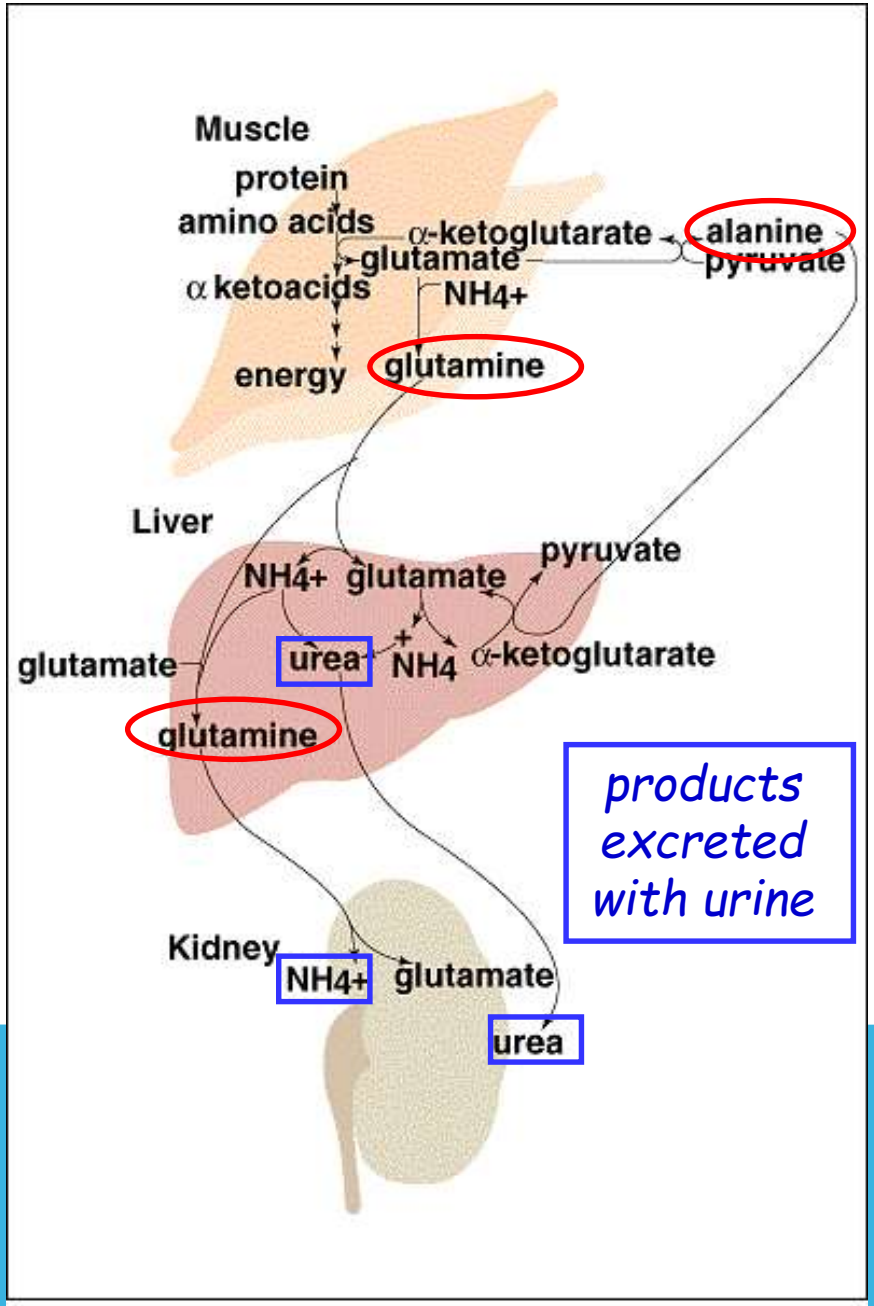


alanine transfers both the carbon sceleton for gluconeogenesis and $-NH_2$ group

The figure was adopted from Devlin, T. M. (editor): Textbook of Biochemistry with Clinical Correlations, 4th ed. Wiley-Liss, Inc., New York, 1997. ISBN 0-471-15451-2

Transport of amino nitrogen

from degraded muscle proteins



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AMMONIA INTOXICATION

The ammonia produced by enteric bacteria and absorbed into portal venous blood and the ammonia produced by tissues are rapidly removed from circulation by the liver and converted to urea.

Thus, only traces (10–20 g/dL) normally are present in peripheral blood.

This is essential, since ammonia is toxic to the central nervous system.

Should portal blood bypass the liver, systemic blood ammonia levels may rise to toxic levels.

This occurs in severely impaired hepatic function or the development of collateral links between the portal and systemic veins in cirrhosis.

AMMONIA INTOXICATION

Excess of ammonia depletes glutamate and hence GABA level in brain

To compensate for glutamate, alpha keto glutarate is used , the decrease concentration of which subsequently depresses TCA and thus deprives brain cells of energy.

Excess Glutamine is exchanged with Tryptophan , a precursor of Serotonin , resulting in hyper excitation.

Symptoms of ammonia intoxication include tremor, slurred speech, blurred vision, coma, and ultimately death.

UREA CYCLE

UREA (ORNITHINE) CYCLE

detoxification pathway (NH_3 is toxic for brain)

proceeds only in the liver

localized in mitochondria /cytoplasm

carbamoyl phosphate synthetase I (= mitoch.)

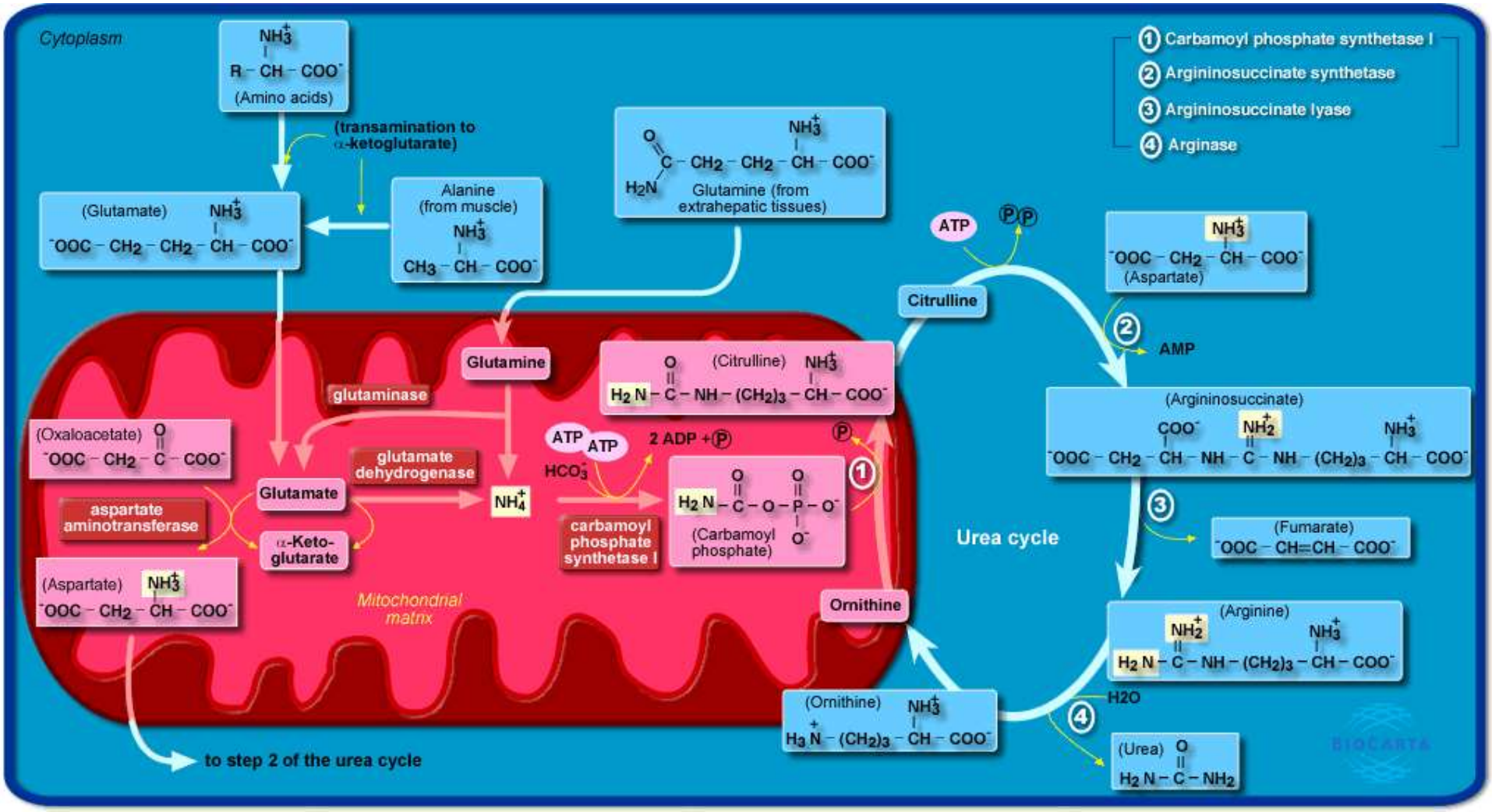
can acidify an organism (consumes HCO_3^-)

needs energy (3 ATP, but 4 energy rich bonds)

connected with citrate cycle through fumarate

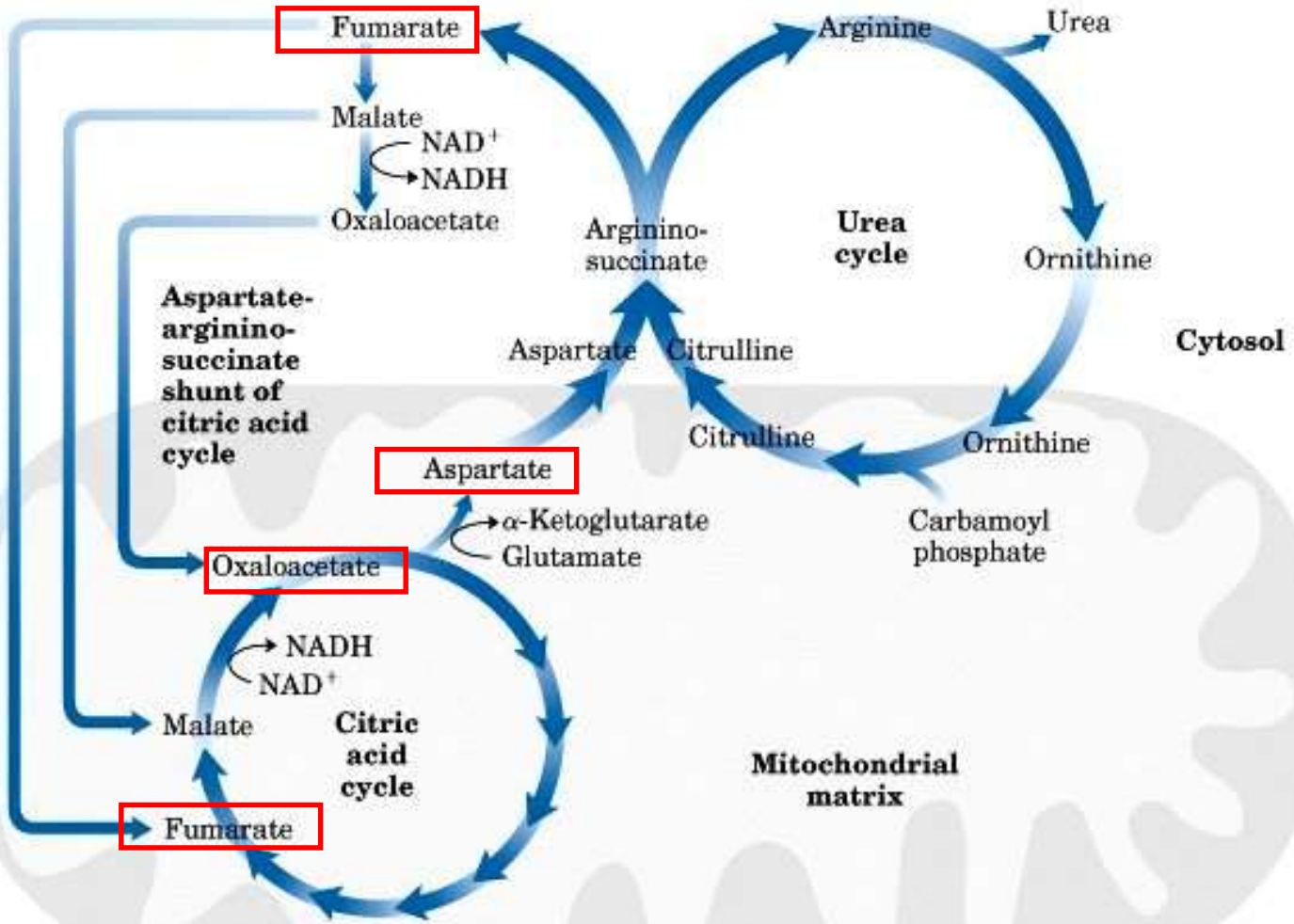
urea is end product of $-NH_2$ metabolism ($\rightarrow$ urine)

Detoxication of ammonia in the liver



The figure is from http://www.biocarta.com/pathfiles/ureacyclePathway.asp (Jan 2007)

Interconnection of the urea cycle with the citrate cycle



The figure is from http://courses.cm.utexas.edu/archive/Spring2002/CH339K/Robertus/overheads-3/ch18_TCA-Urea_link.jpg *(Jan 2007)*

Regulation of urea cycle

1. Mitochondrial carbamoyl phosphate synthetase I (CPS I)

CPS I catalyzes the first committed step of the urea cycle

CPS I is also an allosteric enzyme sensitive to activation by N-acetylglutamate（AGA） which is derived from glutamate and acetyl-CoA

UREA CYCLE DEFECTS AND HYPERAMMONEMIA

NB- complete loss of a urea cycle enzyme causes death shortly after birth,

Urea Cycle Defects and Hyperammonemia—

(1) Hereditary Hyperammonemia (genetic deficiencies of Urea cycle enzymes)

- Ornithine carbamyl transferase (OTC) deficiency (X linked)
- Carbamyl phosphate synthetase I (CPS I) deficiency
- Citrullinemia (enzyme defect?)
- Arginosuccinic Aciduria (enzyme defect?)
- Argininemia **(not severe why?)**(enzyme defect?)

- N-acetylGlu synthase deficiency

Urea Cycle Defects and Hyperammonemia

(2) **Acquired Hyperammonemia-**------

a) Liver disease---- (cirrhosis , hepatitis)
b) High protein diet

Clinical significance of blood urea:

- Elevated in renal insufficiency.
- Decreased in hepatic failure.

Principles of treatment of urea cycle defects

- Alternative pathway stimulation; oral drugs that cause an increase in the excretion of glycine thereby depleting ammonia by stimulating the replacement synthesis of glycine

Most commonly:

- Benzoate

Can also involve using:

- Phenylbutyrate
- Phenylacetate

(See Treatment and Monitoring Module)

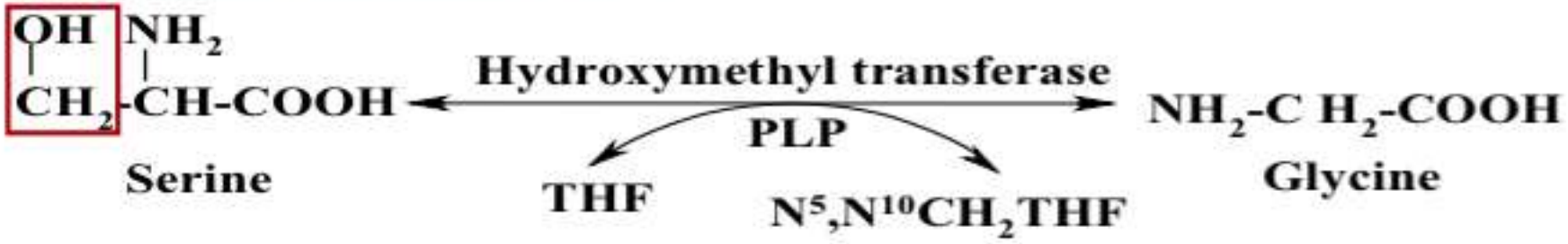
- Haemodialysis, in cases of acute, extreme hyperammonaemia
- Stimulation of CPS by a synthetic co-factor
- A low protein diet is a very common strategy to control the chronic hyperammonaemia
- Arginine supplementation, in relevant disorders

METABOLISM OF INDIVIDUAL AMINO ACIDS

1. Metabolism of Glycine: nonessential, glucogenic.

Biosynthesis of glycine:

1



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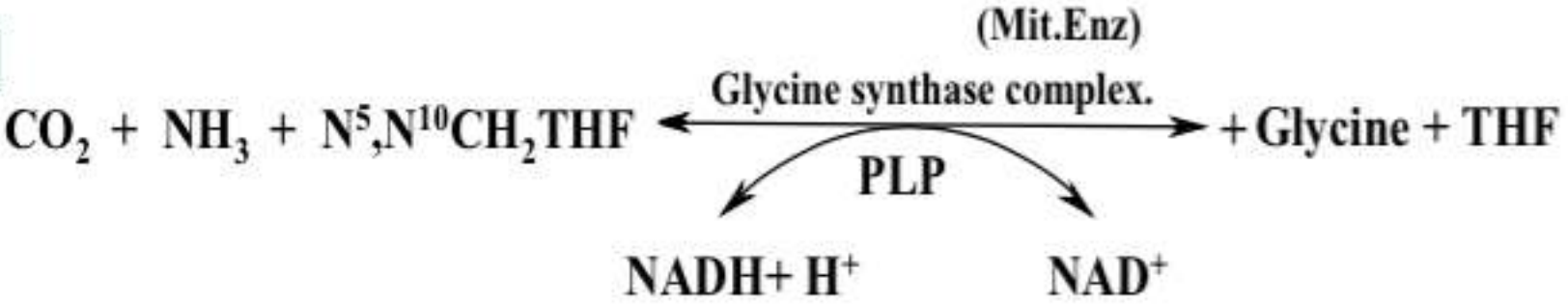


Special Functions of Glycine:

a-Protein, Hormones & enzymes.

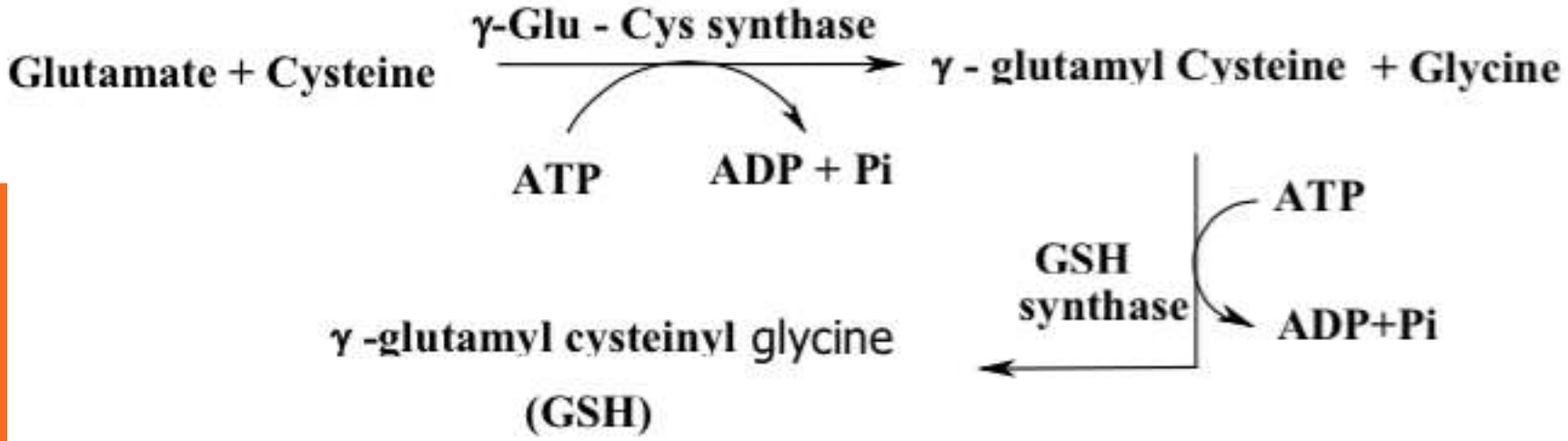
b- Heme **c- Purines (C_4,C_5,N_7)** **d- Creatine**

e- Glutathione

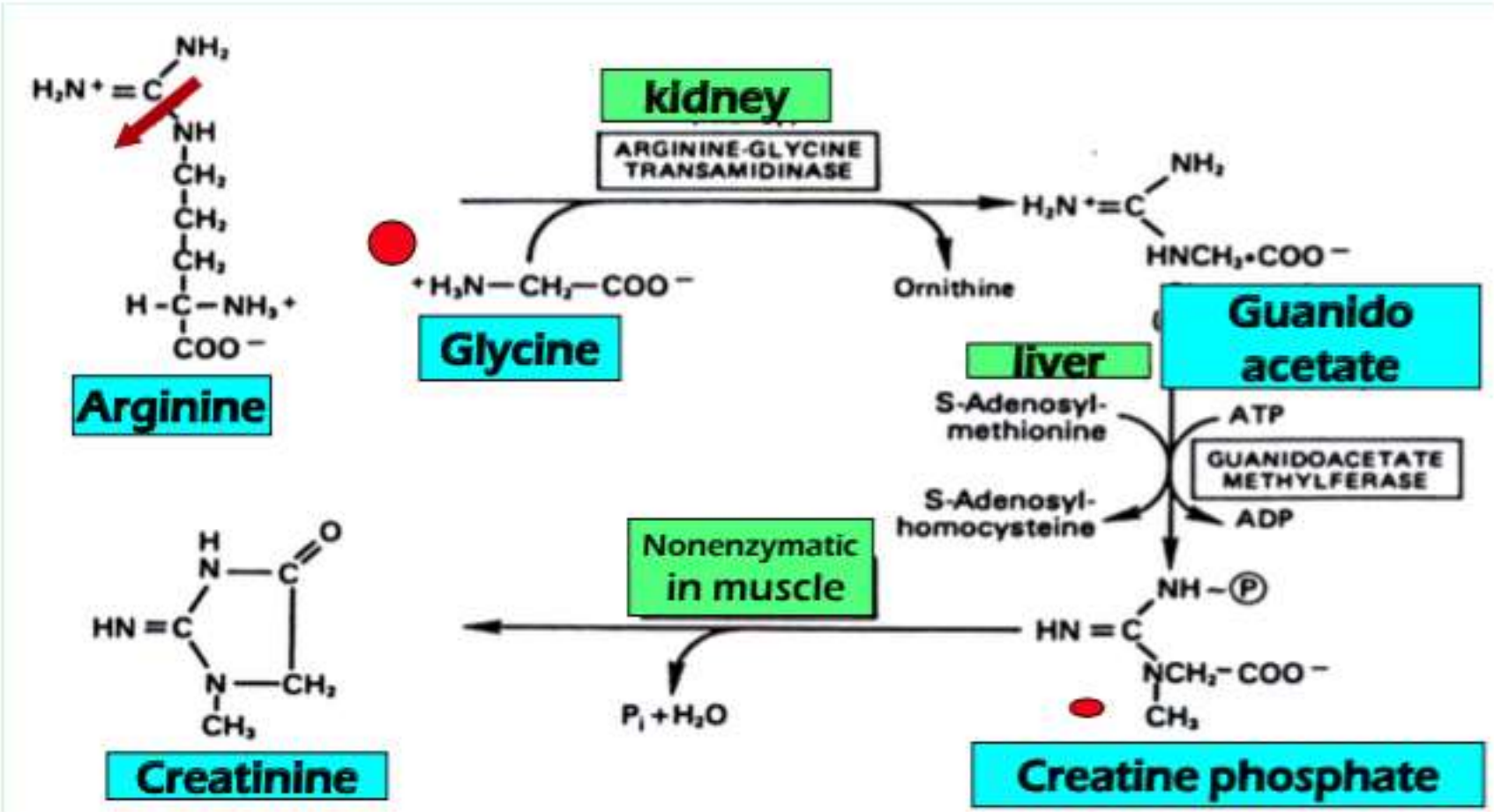
f- Conjugating reactions:

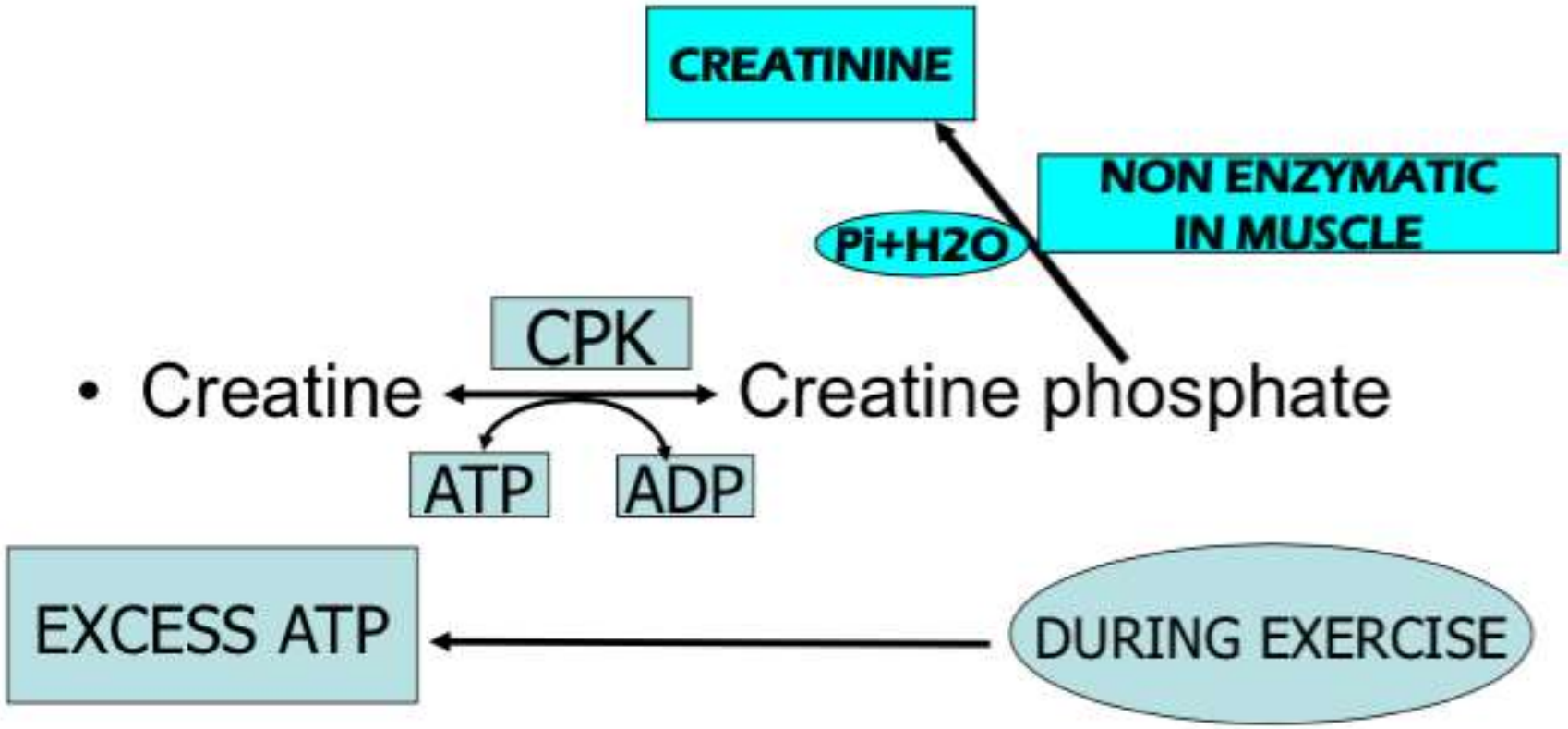
- Glycine + Cholic acid → glycocholate.
- Glycine + Benzoic acid → Hippuric acid

1.Formation of Glutathione (GSH) Dest.FR & Peroxides



2. Formation of creatine (Methyl guanidoacetate)

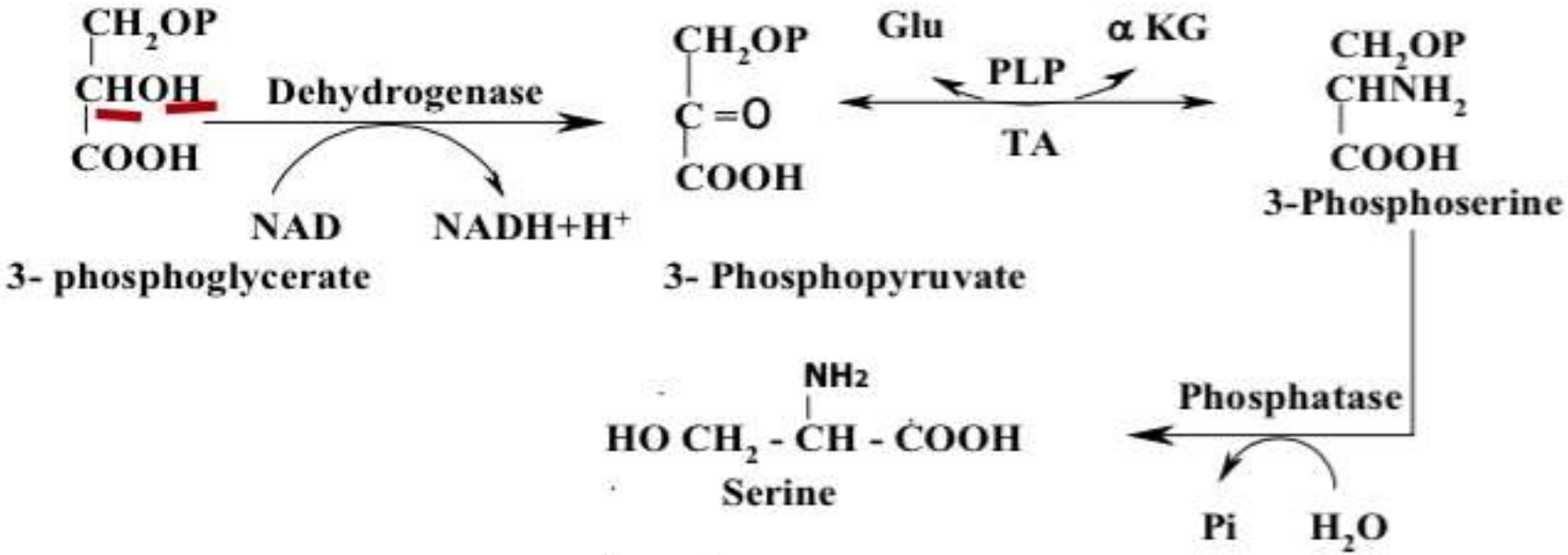




- Cr-P is the storage form of high energy phosphate in muscle
- Creatinine is excreted in urine & increases on kidney failure due to its filteration is decreased.
  Its level is constant per 24 hrs
  & is proportional to muscle mass in human.

2. Metabolism of Serine: nonessential & glucogenic

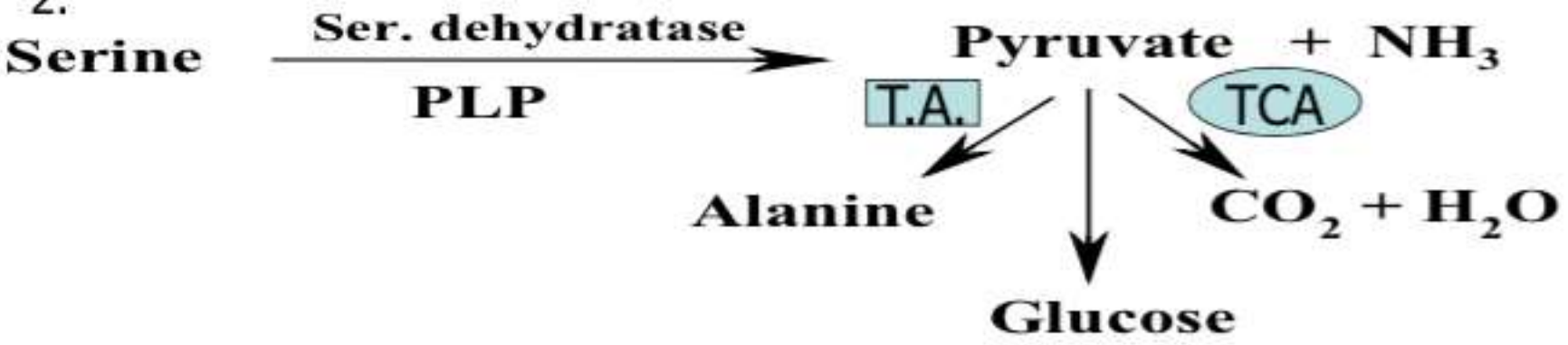
- It is synthesize from glycine or
- intermediate of glycolysis,
- all enzymes are activated by testosterone in liver, kidney & prostate.



Degradative Pathways of Serine:

1. Serine ⟷ Glycine ⟷ CO2+NH3 (major)

2.



Serine is important in synthesis of:

a. Phosphoprotein
b. Purines & pyrimidine
c. Sphingosine
d. Choline
e. Cysteine

3. Metabolism of Sulfur-Containing amino acids

(Methionine, cyteine & Cystine):

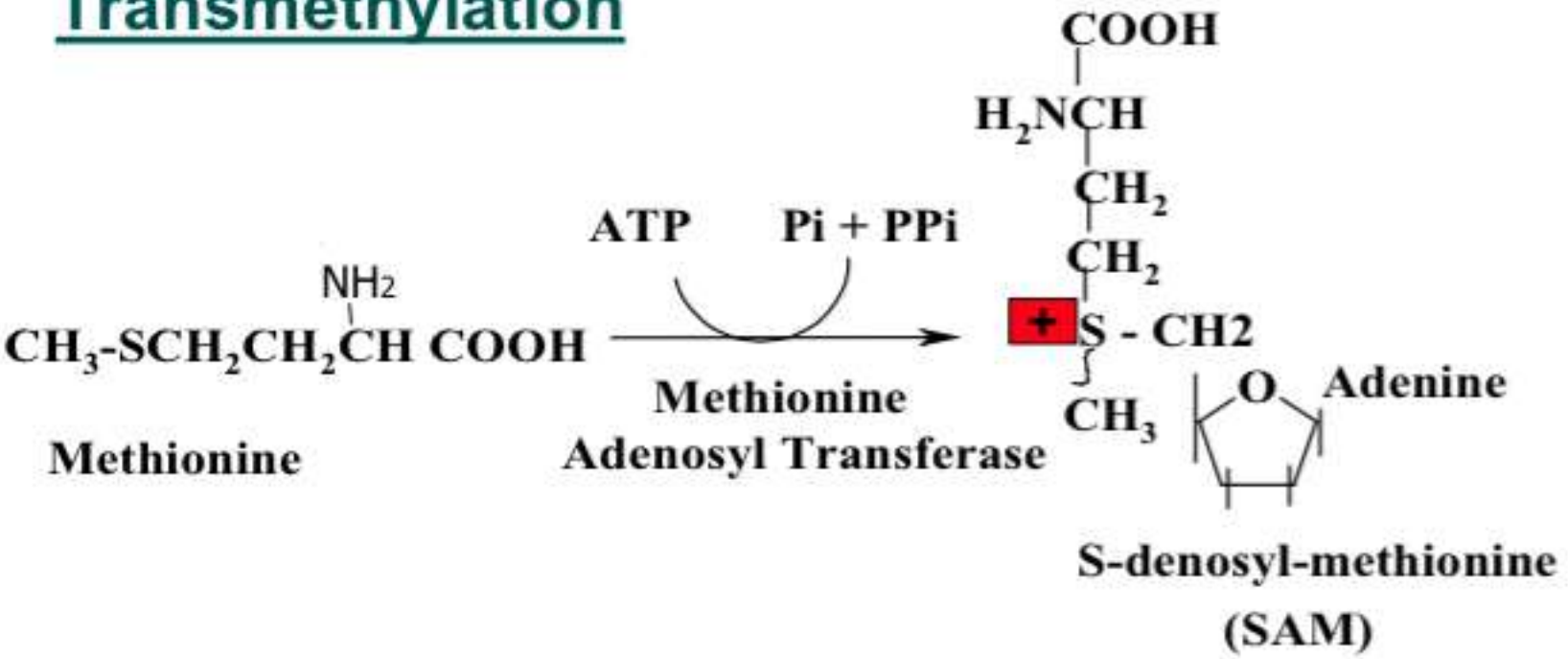
a) Metabolism of methionine: (essential)

Met. ⟶ Cysteine (diet.pr.)

- **2 principal metabolic pathways:**

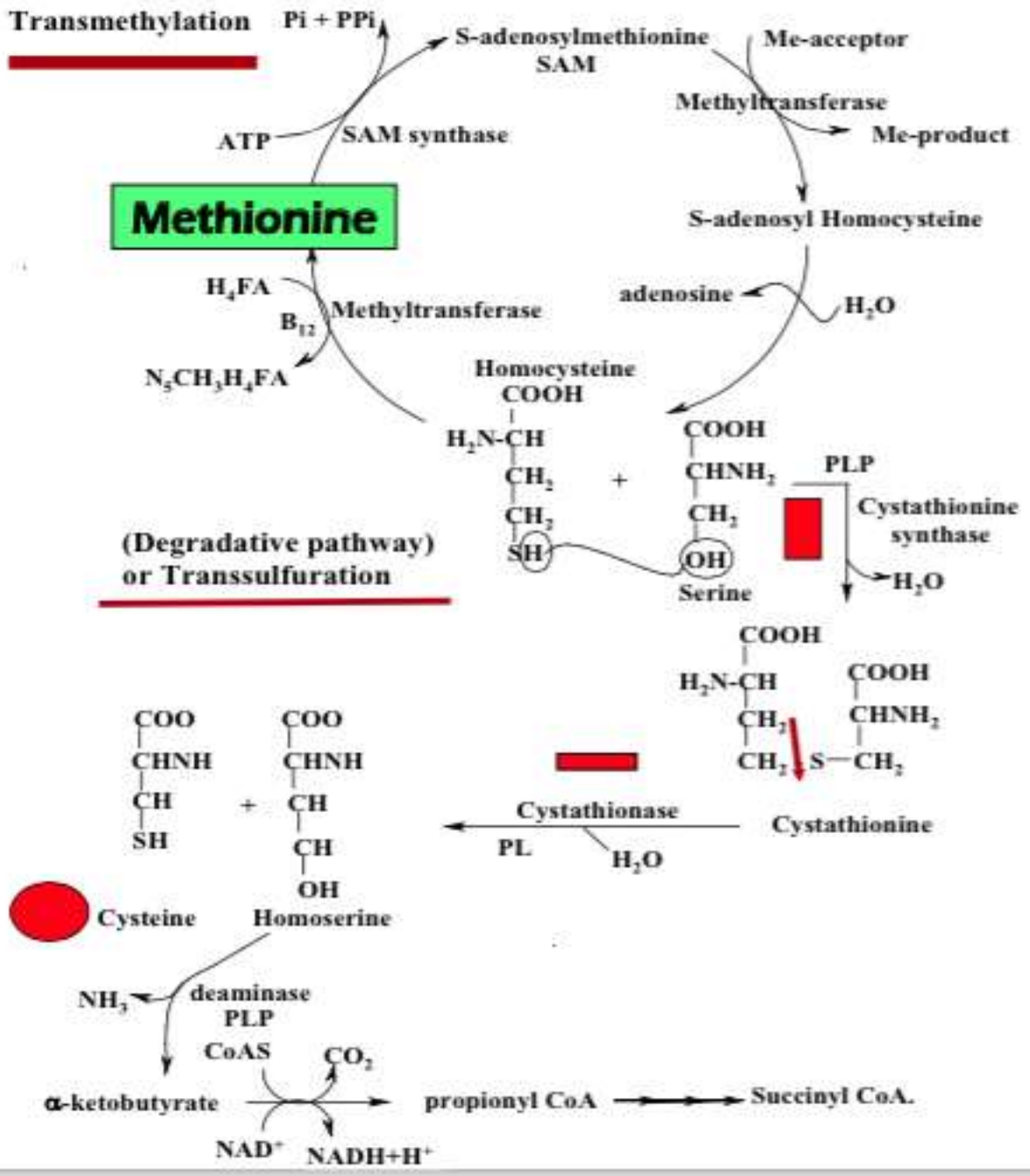
  Transmethylation and transsulfuration

- **Transmethylation**



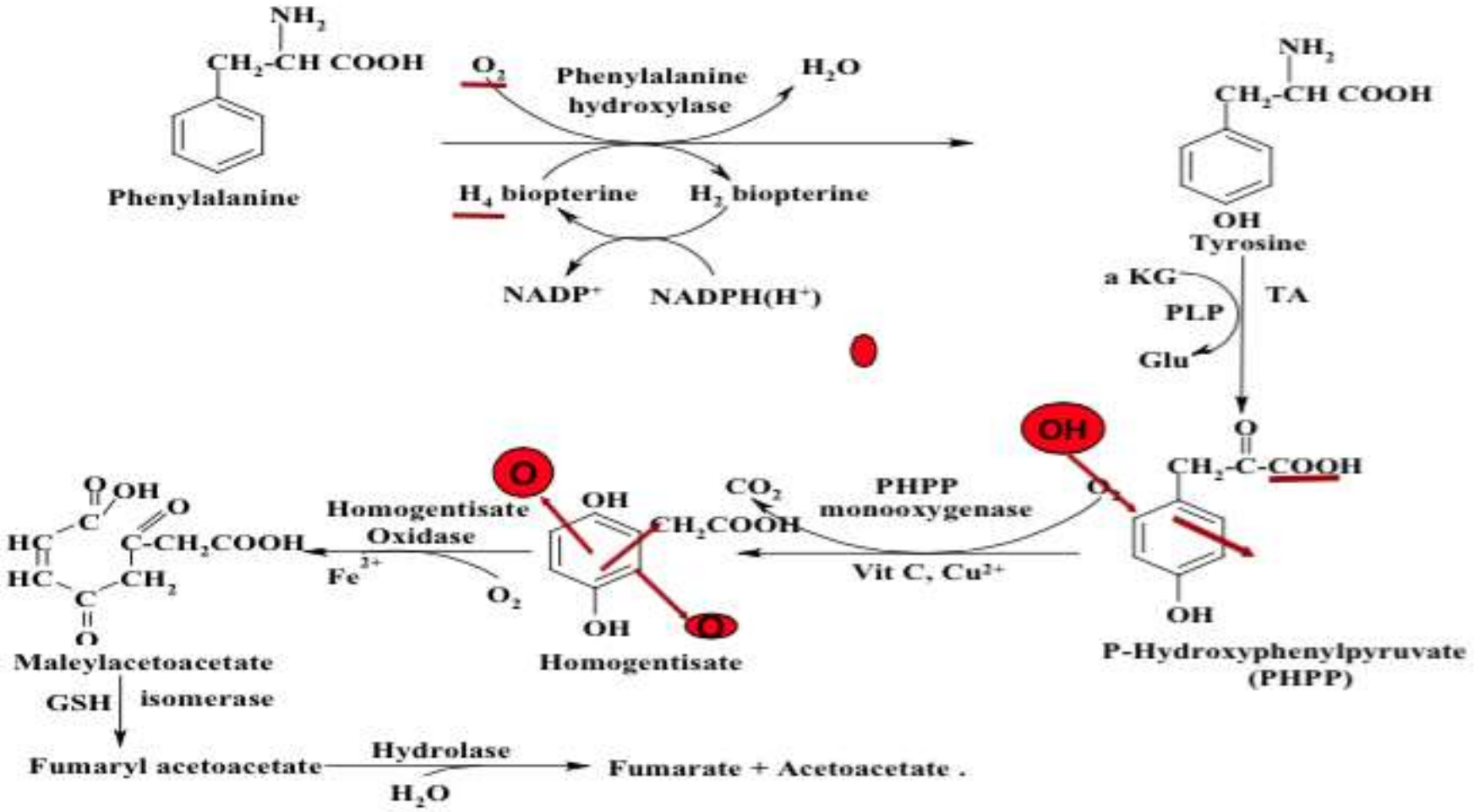
Homocystinuria
Lack of
Cystathionine
synthase

C-skeleton of cysteine
From serine &
S from methionine



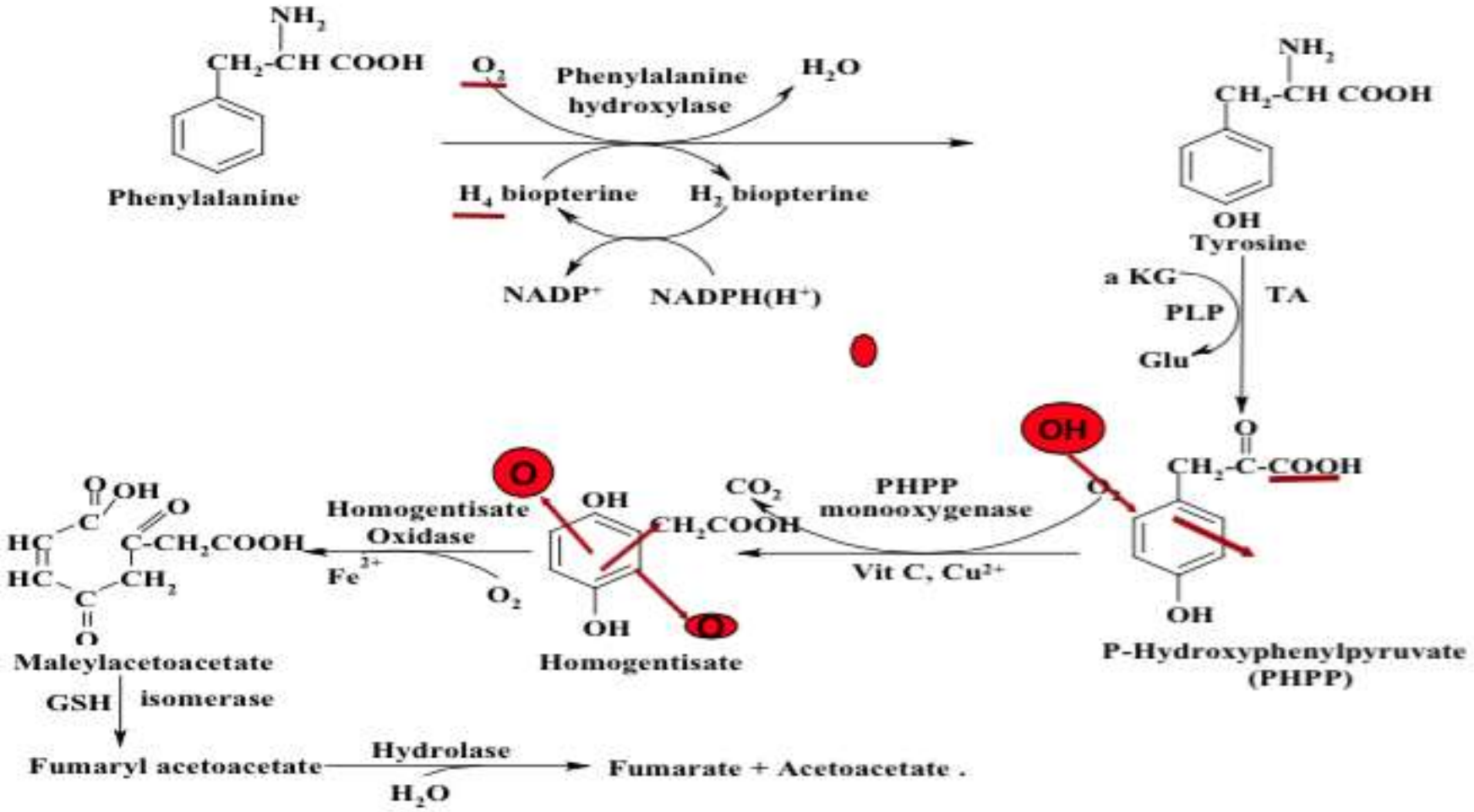
4. Aromatic amino acids

a) Metabolism of Phenylalanine (glucogenic & ketogenic)



4. Aromatic amino acids

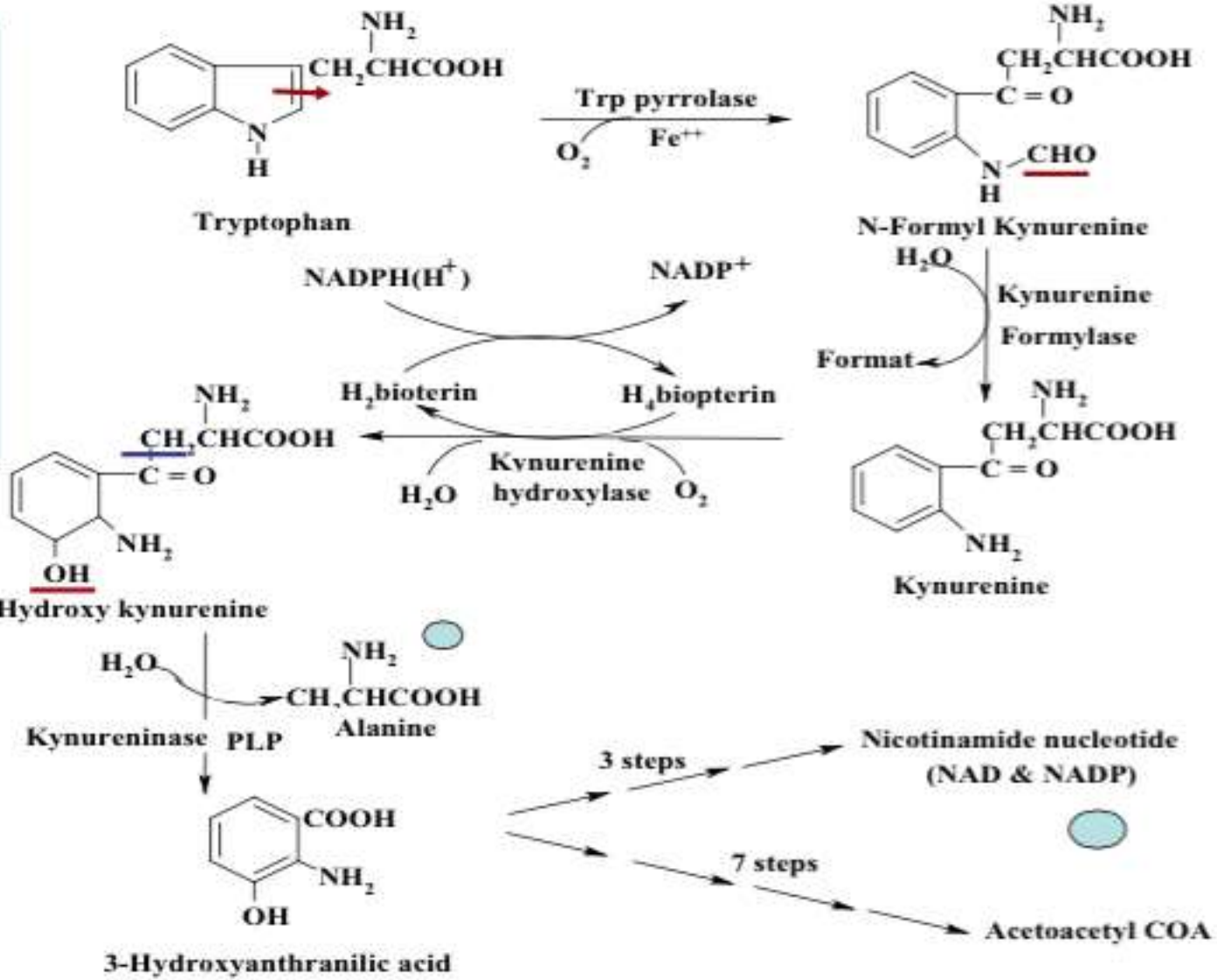
a) Metabolism of Phenylalanine (glucogenic & ketogenic)



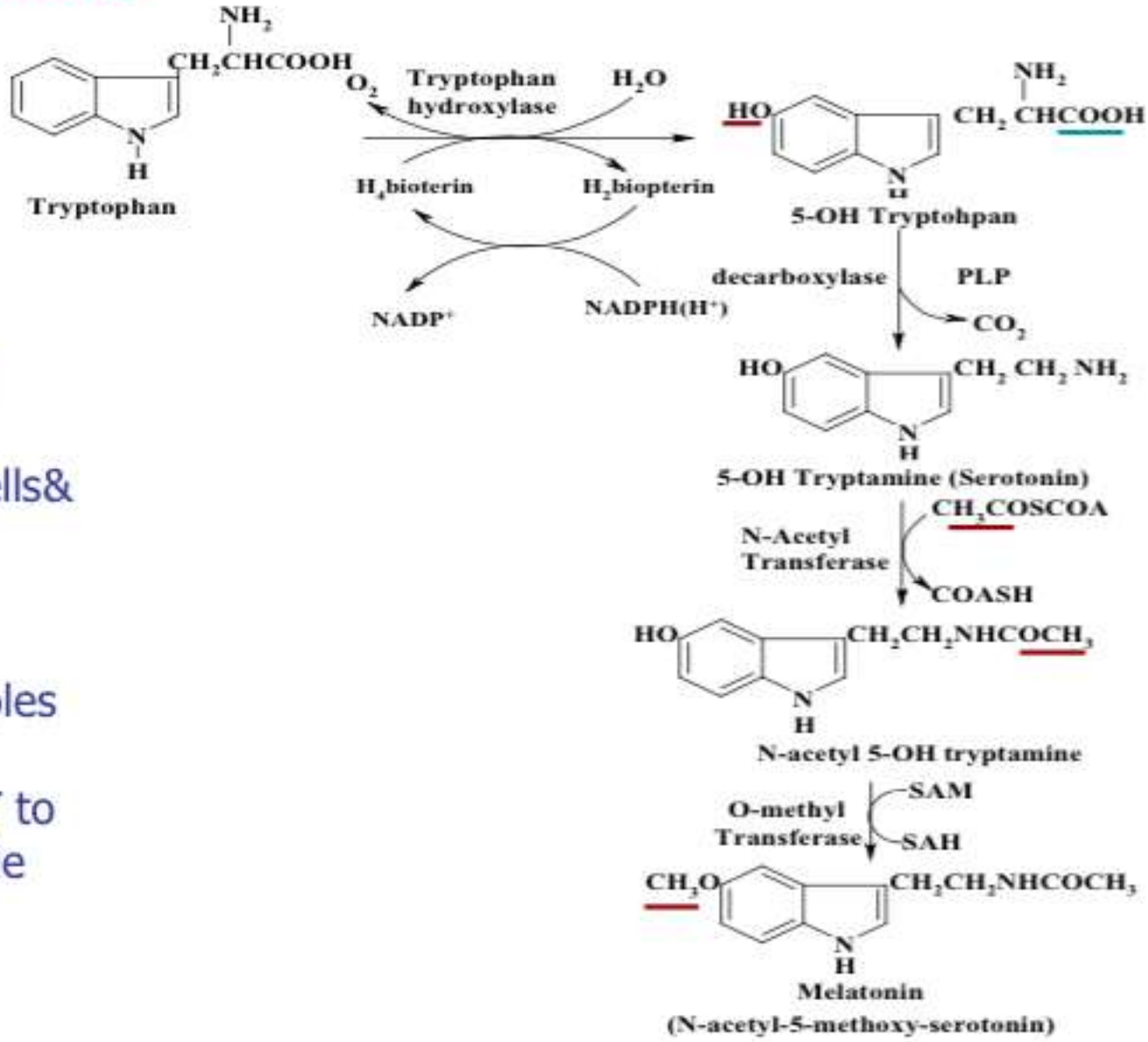
c) Tryptophan (essential,glucogenic&ketogenic)

I] 3-hydroxyanthranilic acid pathway:

- Trp pyrrolase Inc.by Cortico. & tryptophan & Dec.by Niacin, NAD & NADP



III] Serotonin Pathway:



* Neurotransmitter
* Founds in mast cells& platelets.
* Vasoconstrictor for B.V.& bronchioles
* Transmitter in GIT to release the peptide hormones.

5. Branched Chain Amino Acids:

- **Leucine, isoleucine and valine** are taken up by striated muscles after protein meal and oxidized in sk. muscle.
- They are used by **the brain**.
- ***Summary of their degredation:***

Nitrogen : Transferred from all of them forming glutamate

Carbons :

Leucine	**Acetyl CoA** & acetoacetate
Isoleucine	**Succinyl CoA** & Acetyl COA
Valine	**Succinyl CoA** & CO_2

6. Basic Amino Acids:

1) Histidine (glucogenic amino acid):

a) Together with B-alanine , It forms **carnosine** (B-alanyl histidine) and **anserine** (methyl carnosine):

1. They are **buffer the pH of anerobically contracting skeletal muscle**
2. They **activate myosin ATP-ase**
3. They **chelate copper and enhance Cu^{2+} uptake**.

b) Histidine is a source of one-carbon atom.

c) Histidine —(decarboxylase)→ Histamine

Histamine is a chemical messenger that mediates **allergic and inflammatory reactions, gastric acid secretion and neurotransmission in the brain.**

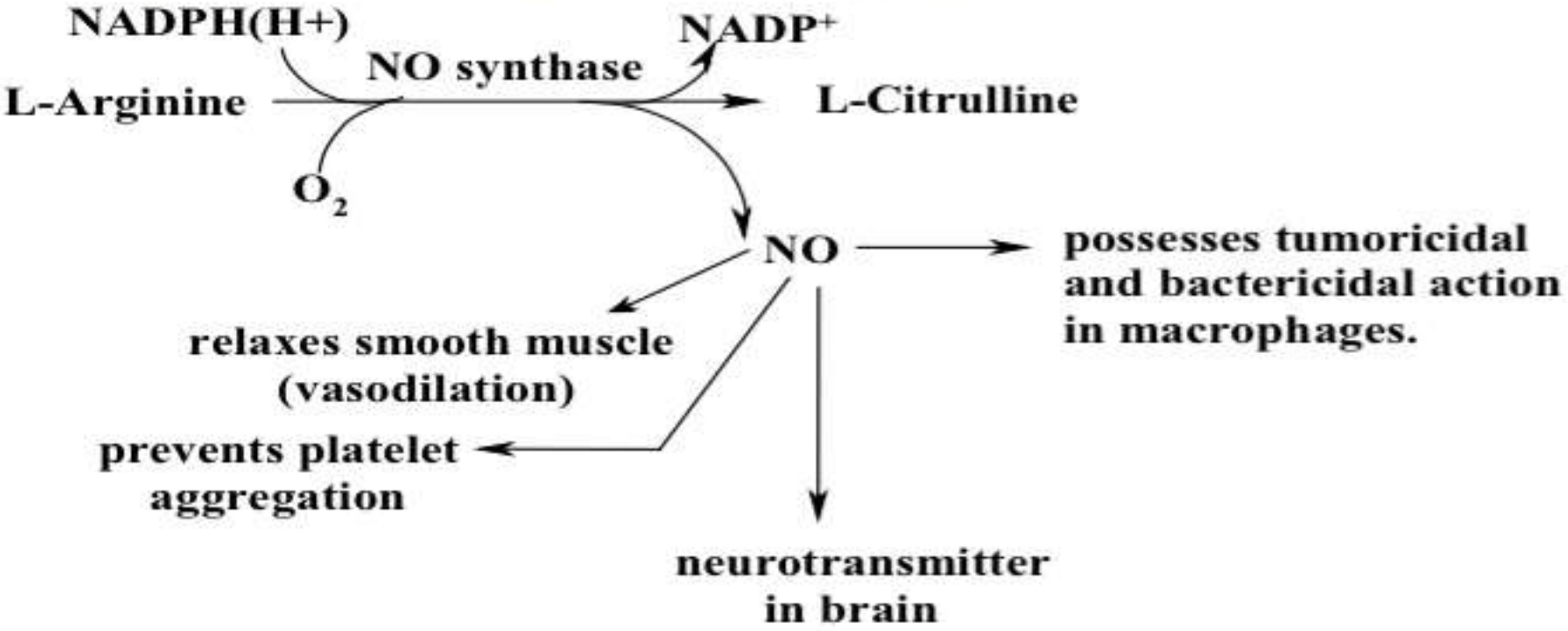
(2) Arginine: (nonessential & glucogenic amino acid):

It participates in formation of:

a)Creatine

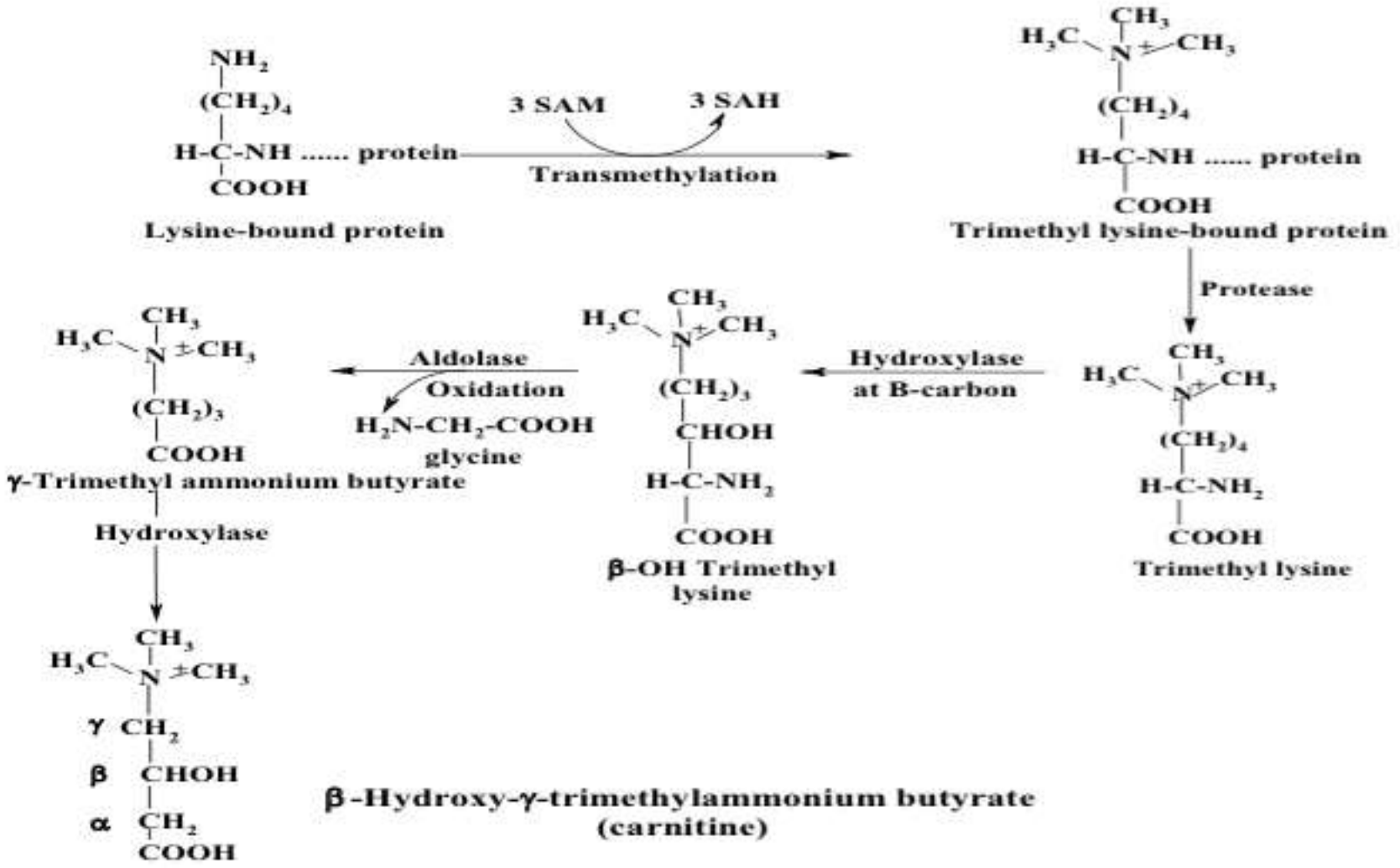
b)Polyamines

C)Nitric oxide NO (Free radical gas).



3) Lysine: (essential, ketogenic)

it is involved in the formation of histone, hydroxylysine & carnitine:



7. Acidic Amino Acids :

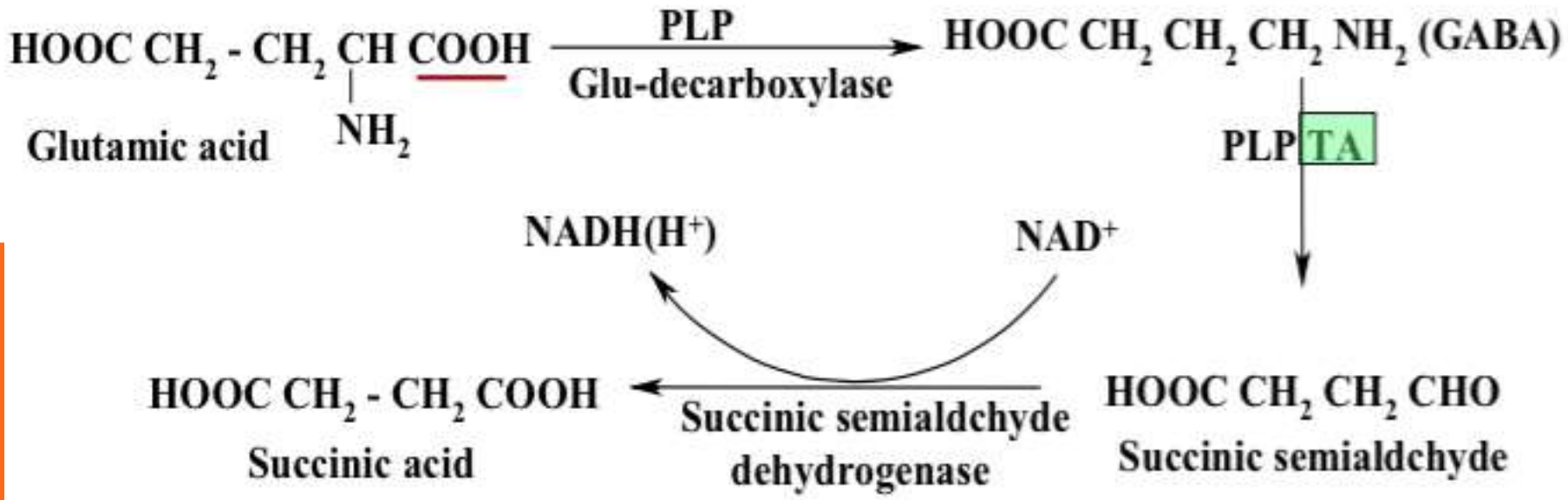
1. Glutamic acid : (nonessential & glucogenic amino acid).

It participates in formation of:

1- GSH. 2- Proline

3- Glutamine: as storage and transporter form of ammonia

4- GABA (δ-aminobutyric acid) neurotransmitter in brain.



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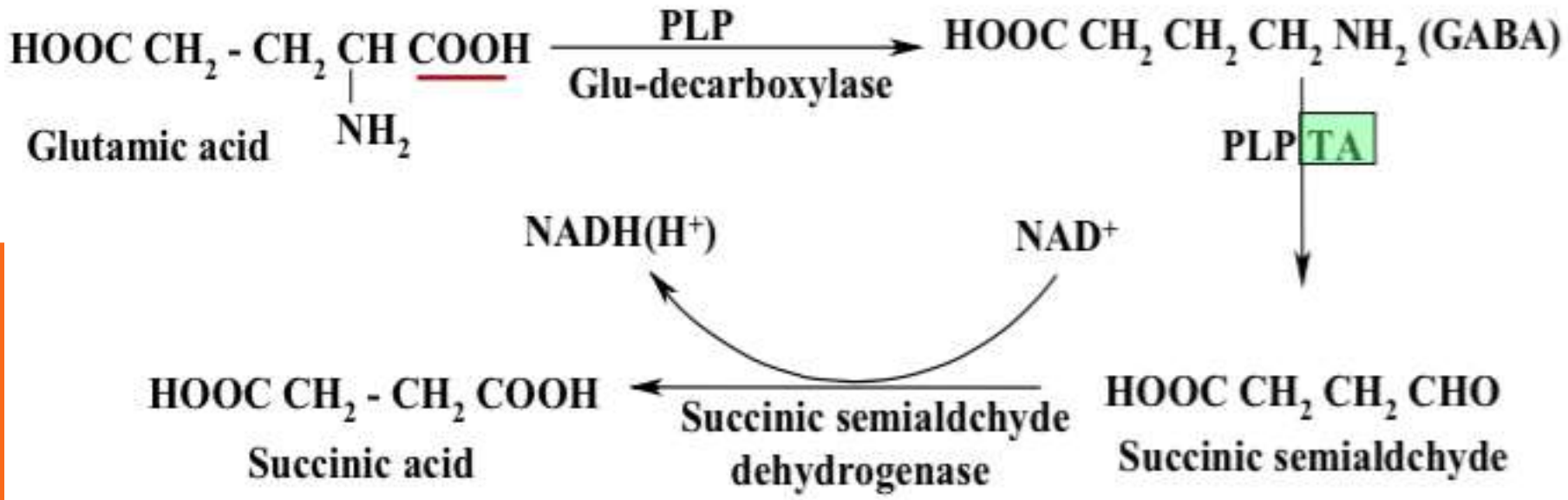


2. Aspartic acid

- Acidic, non essential & glucogenic
- It is important in formation of:

1. Asparagine with NH3.
2. Purine&pyrimidine.
3. Arginosuccinate in urea cycle.
4. Alanine by decarboxylation.
5. Oxalate & glucose by T.A.

Amino acids as precursors of neurotransmitters

1. Serine Choline --- Acetyl choline.
2. Arginine ----------NO
3. Tryptophan----------Serotonin
4. Histidine----------Histamine
5. Phenyl alanine------dopa,dopamine, NE&E
6. Glutamic acid--------GABA

Errors Of Amino Acid Metabolism And Clinical Significance--

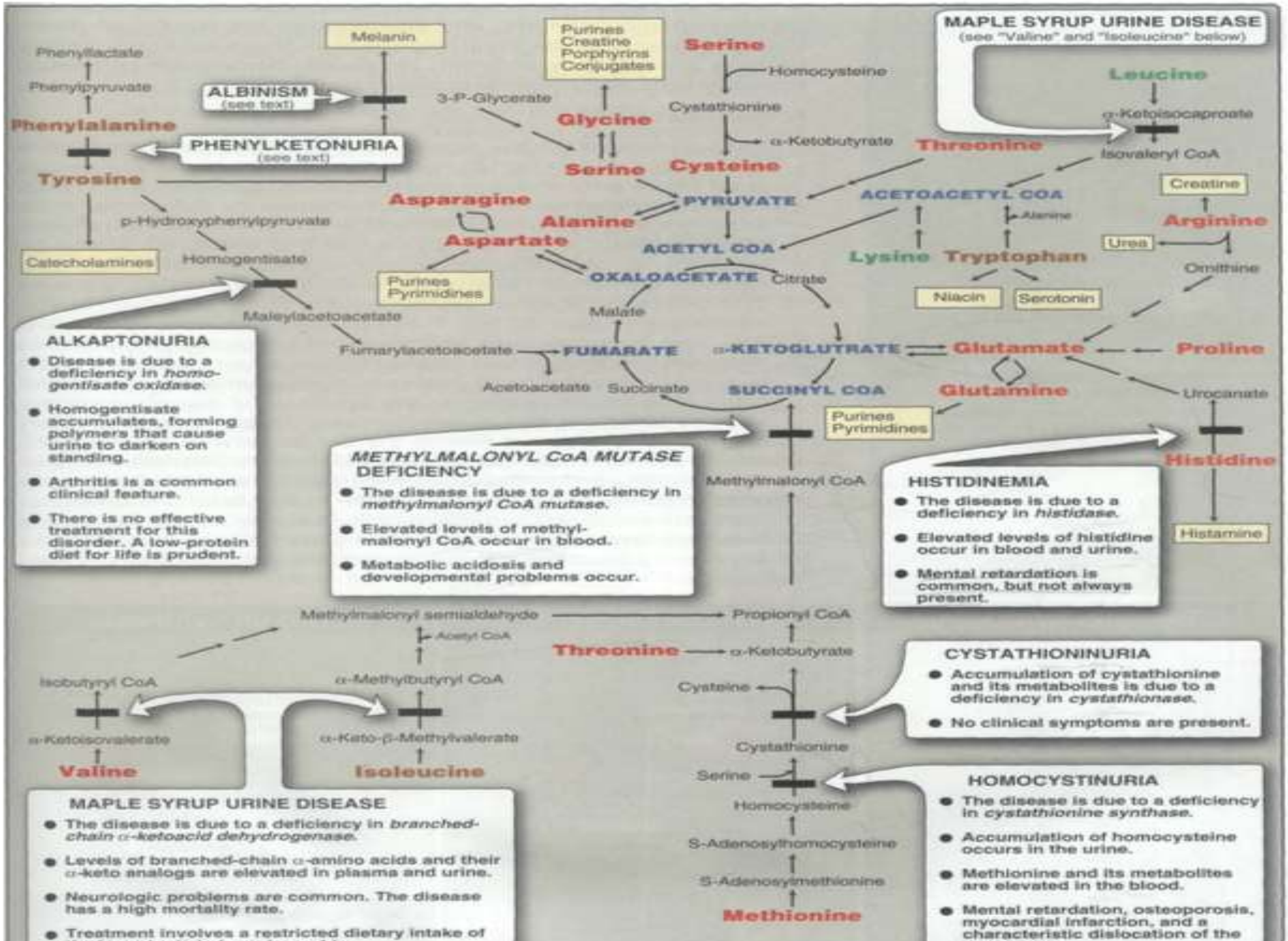

MAPLE SYRUP URINE DISEASE
(see "Valine" and "Isoleucine" below)
Leucine
α-Ketoisocaproate
Isovaleryl CoA
Threonine
Phenyllactate
Phenylpyruvate
Phenylalanine
Tyrosine
Melanin
ALBINISM
(see text)
PHENYLKETONURIA
(see text)
Purines
Creatine
Porphyrins
Conjugates
Serine
Homocysteine
Cystathionine
α-Ketobutyrate
Cysteine
3-P-Glycerate
Glycine
Serine
Asparagine
Alanine
Aspartate
PYRUVATE
ACETOACETYL COA
Alanine
ACETYL COA
Creatine
Arginine
Urea
Ornithine
Lysine
Tryptophan
p-Hydroxyphenylpyruvate
Catecholamines
Homogentisate
Purines
Pyrimidines
OXALOACETATE
Citrate
Niacin
Serotonin
Malate
Maleylacetoacetate
ALKAPTONURIA
Disease is due to a deficiency in homogentisate oxidase.
Homogentisate accumulates, forming polymers that cause urine to darken on standing.
Arthritis is a common clinical feature.
There is no effective treatment for this disorder. A low-protein diet for life is prudent.
Fumarylacetoacetate
FUMARATE
α-KETOGLUTRATE
Glutamate
Proline
Acetoacetate
Succinate
SUCCINYL COA
Glutamine
Urocanate
Purines
Pyrimidines
Histidine
METHYLMALONYL CoA MUTASE DEFICIENCY
The disease is due to a deficiency in methylmalonyl CoA mutase.
Elevated levels of methylmalonyl CoA occur in blood.
Metabolic acidosis and developmental problems occur.
Methylmalonyl CoA
HISTIDINEMIA
The disease is due to a deficiency in histidase.
Elevated levels of histidine occur in blood and urine.
Mental retardation is common, but not always present.
Histamine
Methylmalonyl semialdehyde
Acetyl CoA
Propionyl CoA
Threonine
α-Ketobutyrate
CYSTATHIONINURIA
Accumulation of cystathionine and its metabolites is due to a deficiency in cystathionase.
No clinical symptoms are present.
Isobutyryl CoA
α-Methylbutyryl CoA
Cysteine
α-Ketoisovalerate
α-Keto-β-Methylvalerate
Cystathionine
Valine
Isoleucine
Serine
HOMOCYSTINURIA
The disease is due to a deficiency in cystathionine synthase.
Accumulation of homocysteine occurs in the urine.
Methionine and its metabolites are elevated in the blood.
Mental retardation, osteoporosis, myocardial infarction, and a characteristic dislocation of the
Homocysteine
S-Adenosylhomocysteine
S-Adenosylmethionine
Methionine
MAPLE SYRUP URINE DISEASE
The disease is due to a deficiency in branched-chain α-ketoacid dehydrogenase.
Levels of branched-chain α-amino acids and their α-keto analogs are elevated in plasma and urine.
Neurologic problems are common. The disease has a high mortality rate.
Treatment involves a restricted dietary intake of

TABLE 18–2 Some Human Genetic Disorders Affecting Amino Acid Catabolism

Medical condition	*Approximate incidence (per 100,000 births)*	*Defective process*	*Defective enzyme*	*Symptoms and effects*
Albinism	<3	Melanin synthesis from tyrosine	Tyrosine 3-monooxygenase (tyrosinase)	Lack of pigmentation: white hair, pink skin
Alkaptonuria	<0.4	Tyrosine degradation	Homogentisate 1,2-dioxygenase	Dark pigment in urine; late-developing arthritis
Argininemia	<0.5	Urea synthesis	Arginase	Mental retardation
Argininosuccinic acidemia	<1.5	Urea synthesis	Argininosuccinase	Vomiting; convulsions
Carbamoyl phosphate synthetase I deficiency	<0.5	Urea synthesis	Carbamoyl phosphate synthetase I	Lethargy; convulsions; early death
Homocystinuria	<0.5	Methionine degradation	Cystathionine β-synthase	Faulty bone development; mental retardation
Maple syrup urine disease (branched-chain ketoaciduria)	<0.4	Isoleucine, leucine, and valine degradation	Branched-chain α-keto acid dehydrogenase complex	Vomiting; convulsions; mental retardation; early death
Methylmalonic acidemia	<0.5	Conversion of propionyl-CoA to succinyl-CoA	Methylmalonyl-CoA mutase	Vomiting; convulsions; mental retardation; early death
Phenylketonuria	<8	Conversion of phenylalanine to tyrosine	Phenylalanine hydroxylase	Neonatal vomiting; mental retardation

Beta-Oxidation of Fatty acids

Definition

- **Beta-Oxidation may be defined as the oxidation of fatty acids on the β-carbon atom.**
- **This results in the sequential removal of a two carbon fragment, acetyl CoA.**

Stages and tissues

- Three stages
- Activation of fatty acids - in the cytosol
- Transport of fatty acids into mitochondria
- Beta-Oxidation proper in the mitochondrial matrix
- Fatty acids are oxidized by most of the tissues in the body.
- Brain, erythrocytes & adrenal medulla cannot utilize fatty acids for energy requirement.

Fatty acid activation

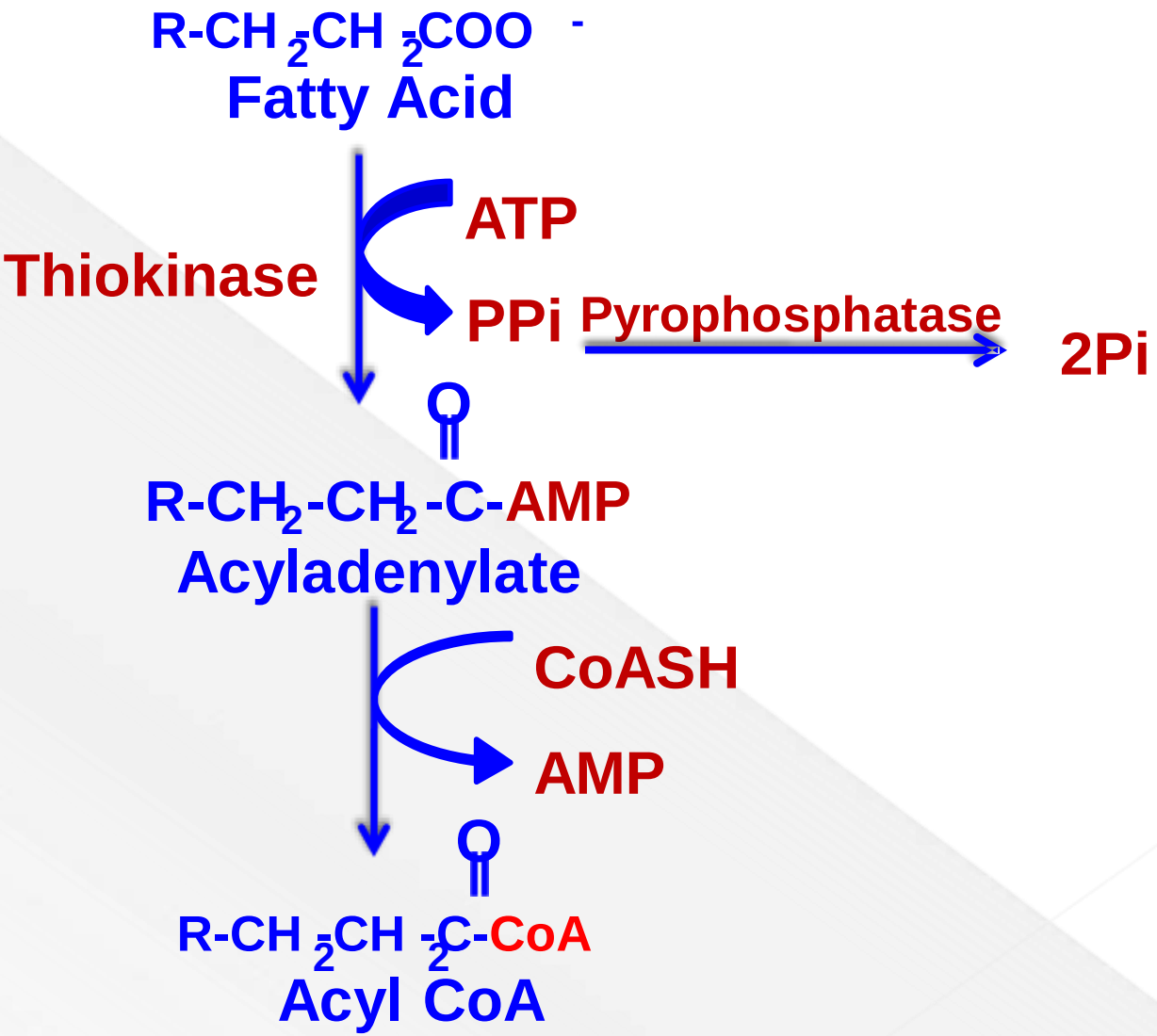
- Fatty acid activation taking place in cytoplasm.
- Fatty acids are activated to acyl CoA by thiokinases or acyl CoA synthetases.
- The reaction occurs in two steps & requires ATP, coenzyme A and Mg^{2+}
- Fatty acid reacts with ATP to form acyladenylate which then combines with coenzyme A to produce acyl CoA.

- Two high energy phosphates are utilized, since ATP is converted to pyrophosphate (PPi).
- The enzyme inorganic pyrophosphatase hydrolyses PPi to phosphate.
- The immediate elimination of PPi makes this reaction totally irreversible.

Thiokinases

- Three different enzymes, one each for short chain, medium chain & long chain fatty acids.
- Small chain fatty acids may also be activated by thiophorase enzyme, using succinyl CoA.

Activation of fatty acid to Acyl CoA



Transport of Acyl CoA into Mitochondria

- The inner mitochondrial membrane is impermeable to fatty acids.
- A specialized carnitine carrier system (carnitine shuttle) operates to transport activated fatty acids from cytosol to the mitochondria.
- Carnitine is β-hydroxy γ-trimethyl aminobutyrate, synthesized by lysine & methionine in liver & kidney.

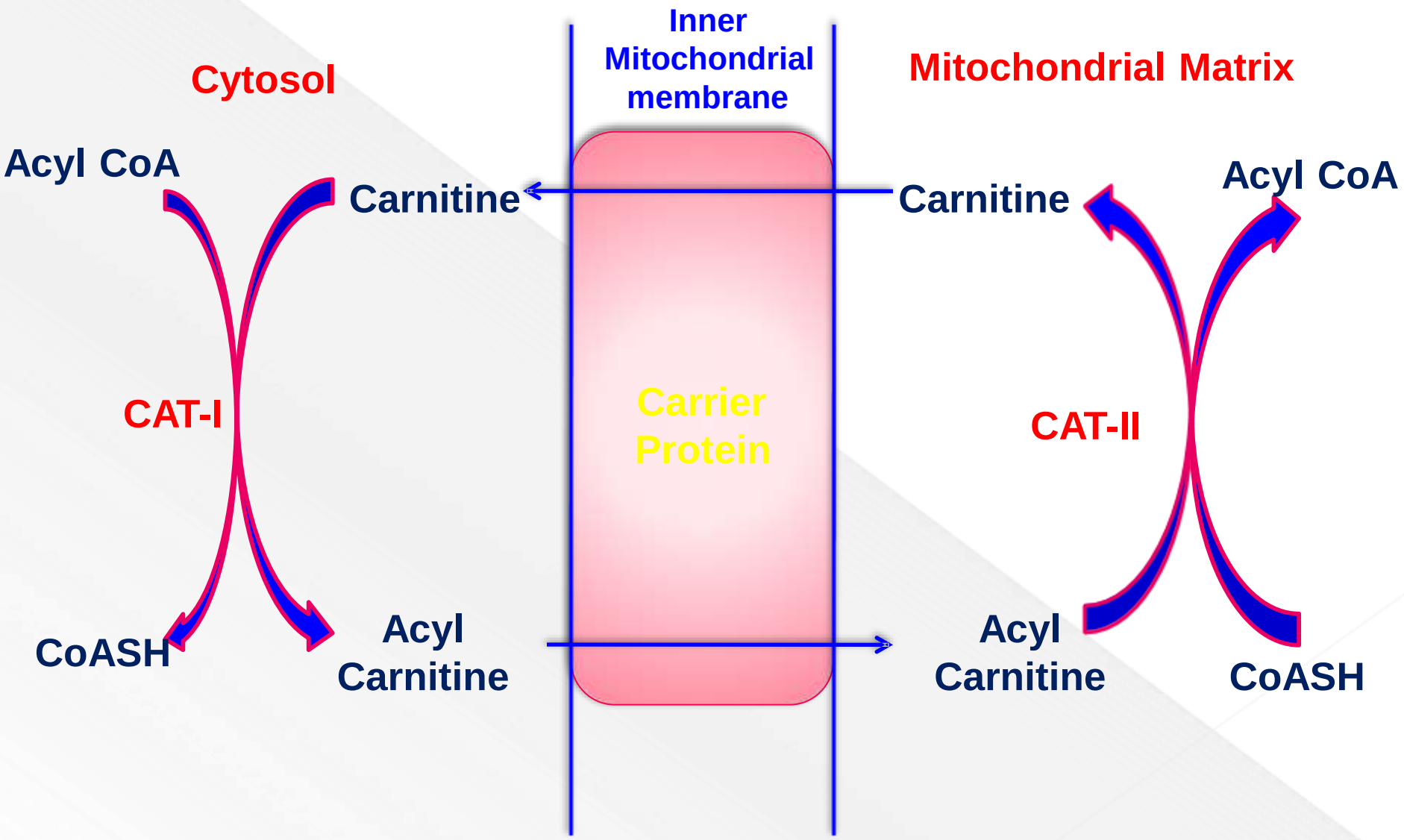
Stages

- It occur in four stages.

1. Acyl group of acyl CoA is transferred to carnitine catalyzed by carnitine acyltransferase I (CAT-I) (present on the outer surface of inner mitochondrial membrane).
2. The acyl-carnitine is transported across the membrane to mitochondrial matrix by a specific carrier protein (Translocase).

3. **Carnitine acyltransferase ll (CAT-II)** (found on the inner surface of inner mitochondrial membrane) converts acyl-carnitine to acyl CoA.
4. The carnitine released returns to cytosol for reuse by translocase.

Carnitine transport system



β-Oxidation Proper

- **Each cycle of β –oxidation, liberating a two carbon unit-acetyl CoA, occurs in a sequence of four reactions**

  1. Oxidation

  2. Hydration

  3. Oxidation

  4. Cleavage

1.Oxidation

- Acyl CoA undergoes dehydrogenation by an FAD-dependent flavoenzyme, acyl CoA dehydrogenase.
- A double bond is formed between α & β carbons (i.e., 2 & 3 carbons)

2. Hydration:

- Enoyl CoA hydratase brings about the hydration of the double bond to form β -hydroxyacyl CoA.

3. Oxidation

- β-Hydroxyacyl CoA dehydrogenase

  catalyses the second oxidation & generates NADH.

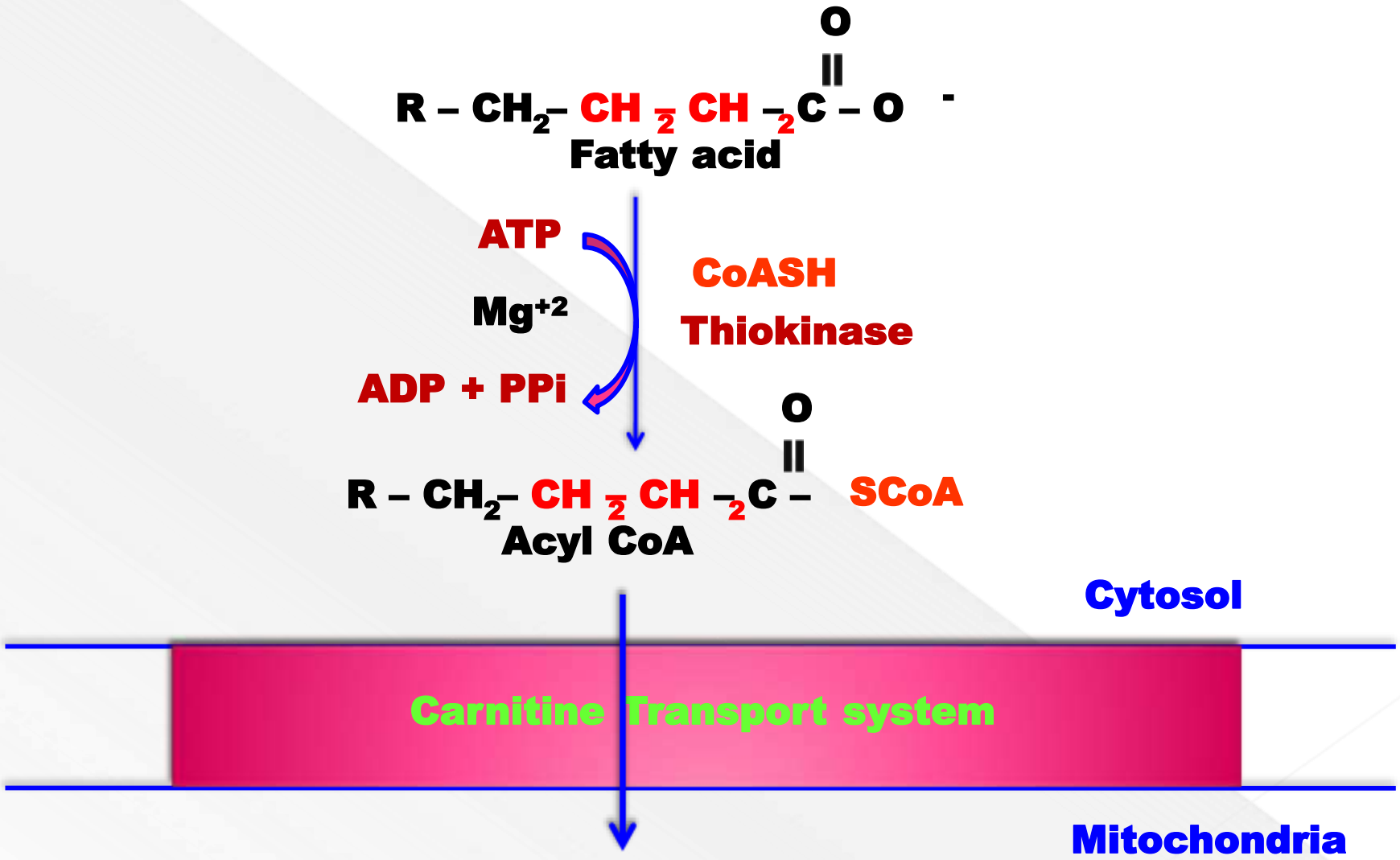
- The product formed is β-ketoacyl CoA.

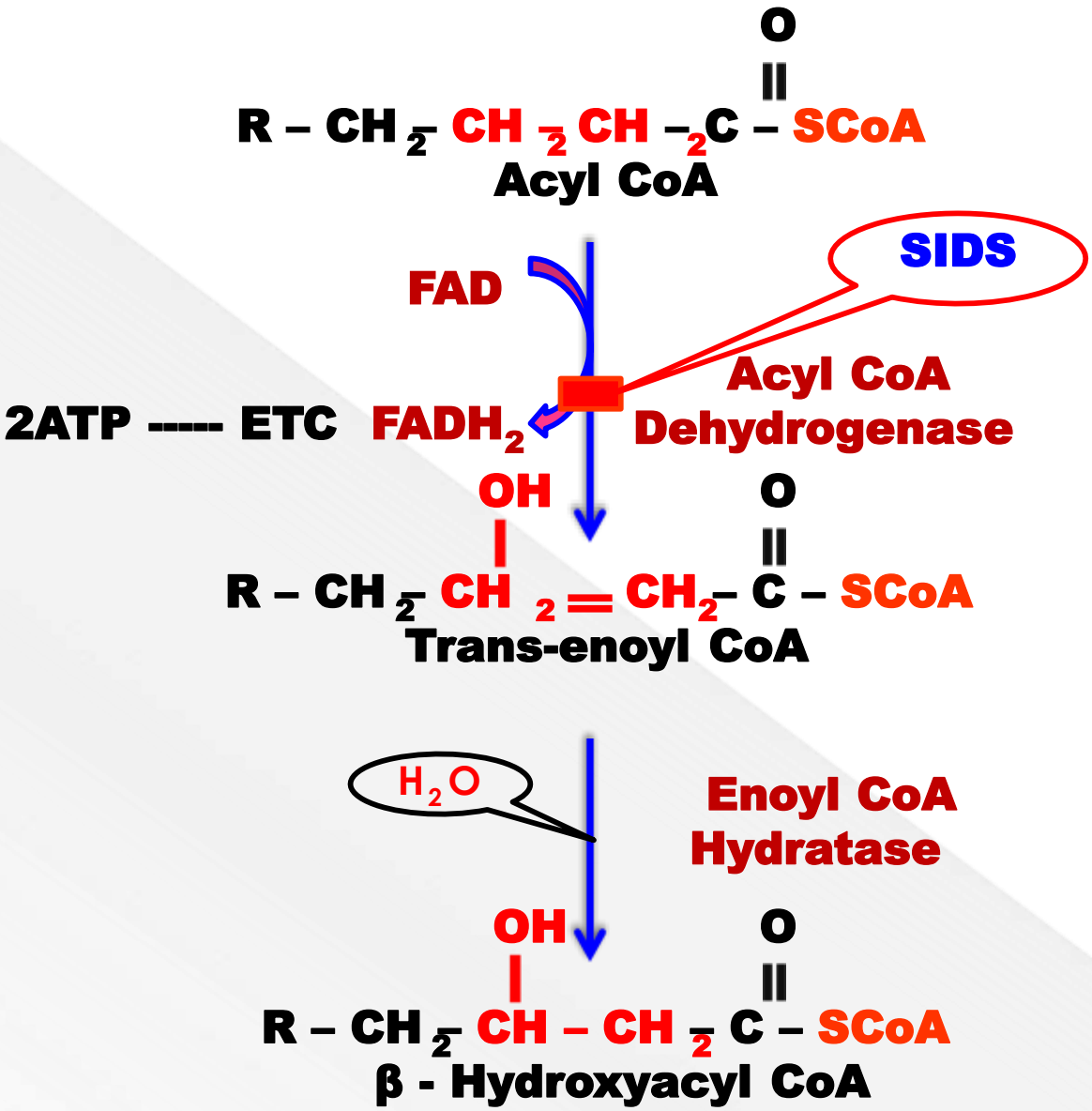
4. Cleavage

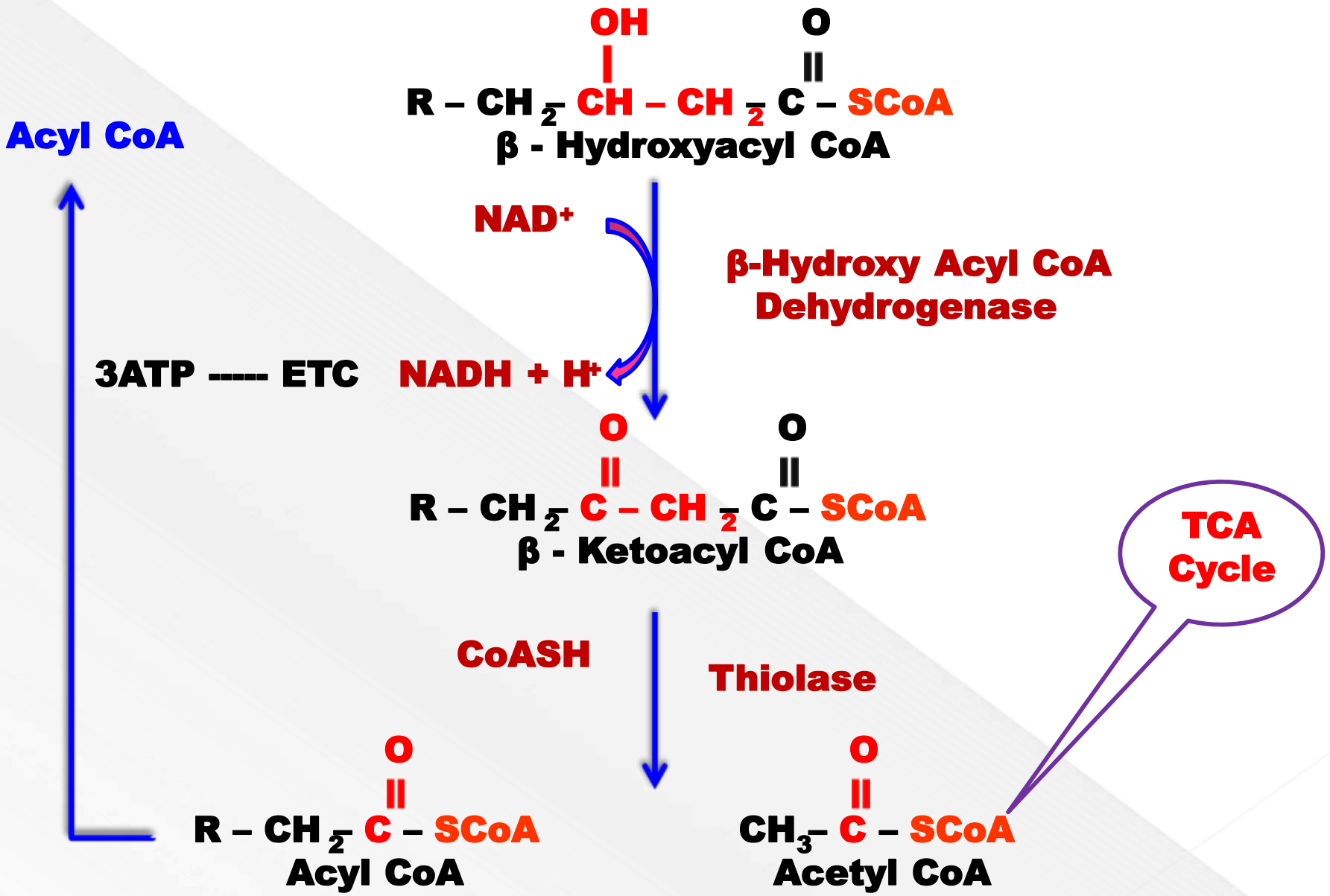
- The final reaction in β -oxidation is the liberation of a 2 carbon fragment, acetyl CoA from acyl CoA.
- This occurs by a thiolytic cleavage catalysed by β-ketoacyl CoA thiolase (or thiolase).

- **The new acyl CoA, containing two carbons less than the original, reenters the β-oxidation cycle.**
- **The process continues till the fatty acid is completely oxidized.**

β-Oxidation of fatty acids




O
R – CH2 – CH2 – CH2 – C – SCoA
Acyl CoA
SIDS
FAD
Acyl CoA
Dehydrogenase
2ATP ----- ETC
FADH2
OH
O
R – CH2 – CH = CH – C – SCoA
Trans-enoyl CoA
H2O
Enoyl CoA
Hydratase
OH
O
R – CH2 – CH – CH2 – C – SCoA
β - Hydroxyacyl CoA


OH
O
R – CH2 – CH – CH2 – C – SCoA
β - Hydroxyacyl CoA
Acyl CoA
NAD+
β-Hydroxy Acyl CoA
Dehydrogenase
3ATP ----- ETC
NADH + H+
O
O
R – CH2 – C – CH2 – C – SCoA
β - Ketoacyl CoA
TCA
Cycle
CoASH
Thiolase
O
O
R – CH2 – C – SCoA
Acyl CoA
CH3 – C – SCoA
Acetyl CoA

Oxidation of palmitoyl CoA

- Palmitoyl CoA + 7 CoASH + 7 FAD^+ 7 NAD^+ + 7 H_2O → 8 Acetyl CoA + 7 $FADH_2$ + 7 NADH + 7H^+
- Palmitoyl CoA undergoes 7 cycles of β - oxidation to yield 8 acetyl CoA.
- Acetyl CoA can enter citric acid cycle & get completely oxidized to CO_2 & H_2O.

Energetics of β -oxidation

Mechanism	ATP yield
I. β- 0xidation 7 cycles 7 FADH2 [Oxidized by Electron Transport Chain (ETC) each FADH2 gives 2 ATP] 7 NADH (Oxidized by ETC, each NADH Liberate 3ATP)	14 21
II. From 8 Acetyl CoA Oxidized by citric acid cycle, each acetyl CoA provides 12 ATP	96
Total energy from one molecule of palmitoyl CoA Energy utilized for activation (Formation of palmitoyl Co A)	131 -2
Net yield of oxidation of one molecule of palmitate	**=129**

Regulation of β-oxidation

- The availability of free fatty acid (FFA) regulates the net utilisation through β-oxidation.
- The level of FFA is controlled by glucagon:insulin ratio.
- Glucagon increases FFA level & insulin has the opposite effect.

- CAT-I is the regulator of entry of fatty acid into mitochondria.
- Malonyl CoA inhibits CAT-I activity.
- Thus during de novo synthesis of fatty acid, the beta oxidation is inhibited.

Sudden infant death syndrome (SIDS)

- **Unexpected death of healthy infants, usually overnight**
- **Due to deficiency of medium chain acyl CoA dehydrogenase.**
- **Glucose is the principal source of energy, soon after eating or feeding babies.**

- **After a few hours, the glucose level & its utilization decrease & the rate of fatty acid oxidation must simultaneously increase to meet the energy needs.**
- **The sudden death in infants is due to a blockade in β -oxidation caused by a deficiency in medium chain acyl CoA dehydrogenase (MCAD)**

Jamaican vomiting sickness

- Characterized by severe hypoglycemia, vomiting, convulsions, coma & death.
- It is caused by eating unriped ackee fruit- contains an unusual toxic amino acid, hypoglycin A.
- This inhibits the enzyme acyl CoA dehydrogenase & β -oxidation of fatty acids is blocked, leading to various complications

- Abnormalities in transport of fatty acids into mitochondria & defects in oxidation leads to deficient energy production by oxidation of long chain fatty acids.
- Features:
- Hypoglycemia, hyperammonemia, weakness & liver diseases.
- Acyl carnitine accumulates when the transferases or translocase is deficient.
- Dietary supplementation of carnitine improve the condition.

Oxidation of odd chain fatty acids

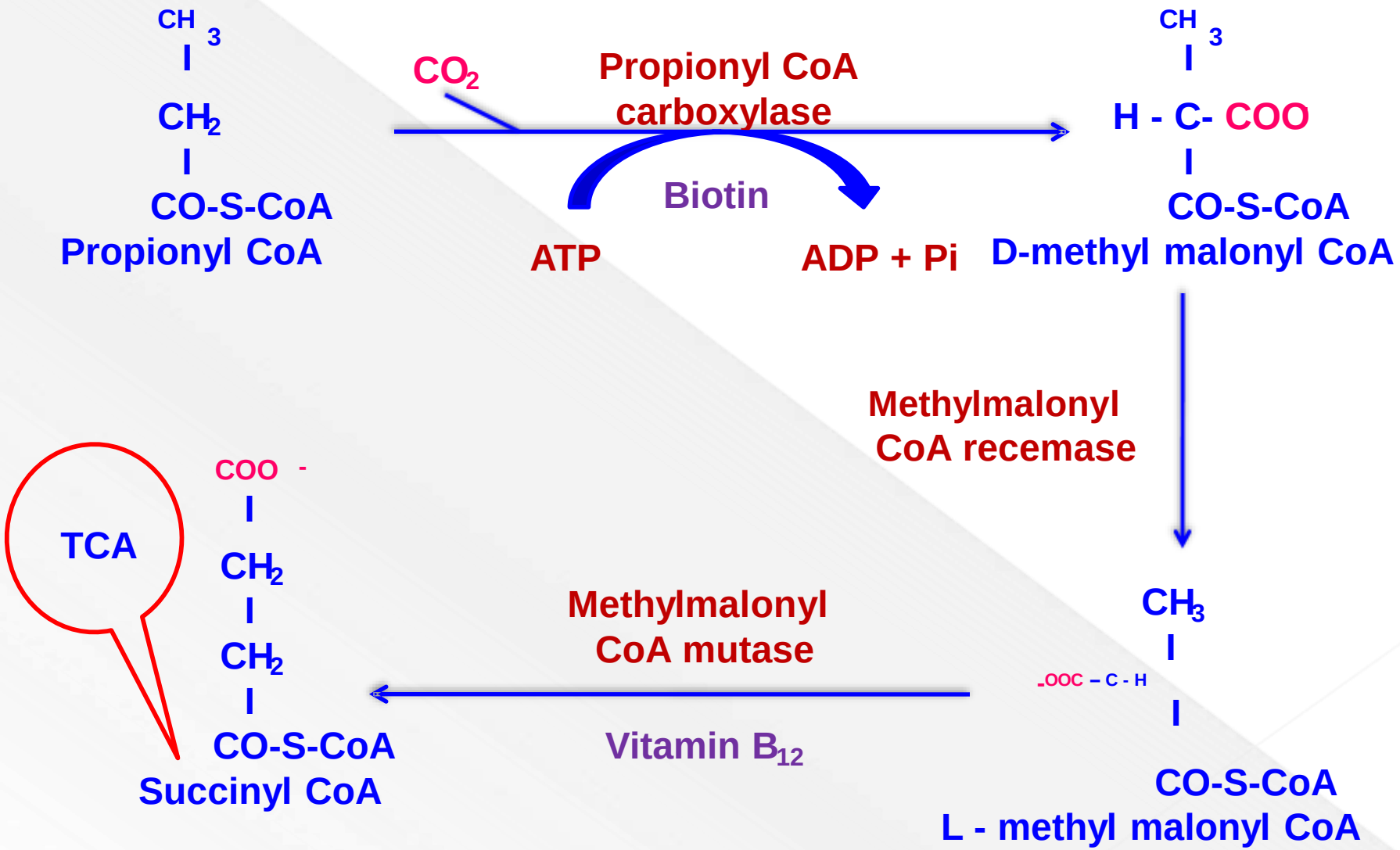
- Oxidation of odd chain fatty acids is similar to that of even chain fatty acids.
- At the end 3 carbon unit, propionyl CoA is produced.
- Propionyl CoA is converted into succinyl CoA.
- Succinyl CoA is an intermediate in TCA cycle
- Propionyl CoA is gluconeogenic.

Conversion of propionyl CoA to succinyl CoA

- **Propionyl CoA is carboxylated to D-methyl malonyl CoA by a biotin dependent carboxylase.**
- **Biotin & ATP is utilized in this step.**
- **Methylmelonyl CoA Recemase:**
- **Recemase acts upon D-methyl malonyl CoA to give L-methyl malonyl CoA.**
- **This reaction is essential for the entry of this compound into metabolic reactions of body.**

- **Methylmalonyl CoA Mutase:**
- **Mutase catalyzes the conversion of L-methyl malonyl CoA (a branched chain compound) to succinyl CoA (a straight chain compound).**
- **Mutase is an vitamin B_{12} dependent enzyme.**
- **Succinyl CoA enters the TCA cycle, & converted into oxaloacetate, it is used for gluconeogenesis.**
- **Propionyl CoA is also derived from metabolism of valine & isoleucine.**

Conversion of succinyl CoA to propionyl CoA



Inborn errors of propionate metabolism

- **Propionyl CoA carboxylase deficiency:**
- **Characterized by propionic acidemia, ketoacidosis & developmental abnormalities.**
- **Methyl malonic aciduria:**
- **Two types of methyl malonic acidemias**
- **Due to deficiency of vitamin B_{12}**
- **Due to defect in the enzyme methyl malonyl CoA mutase or recemase.**

- Accumulation of methyl malonic acid in body.
- Methyl malonic acid is excreted into urine.
- Symptoms:
- Severe metabolic acidosis, damages the central nervous system & growth retardation.
- Fatal in early years of life.
- Treatment:
- Some patients respond to treatment with pharmacological doses of B_{12}.

α-oxidation

- Oxidation of fatty acids on α-carbon atom is known as α-oxidation.
- In this, removal of one carbon unit from the carboxyl end.
- Energy is not produced.
- No need of fatty acid activation & coenzyme A
- Hydroxylation occurs at α-carbon atom.

- It is then oxidized to α-keto acid.
- This, keto acid undergoes decarboxylation, yielding a molecule of CO_2 & FA with one carbon atom less.
- Occurs in endoplasmic reticulum.
- Some FA undergo α - oxidation in peroxisomes.

- α- oxidation is mainly used for fatty acids that have a methyl group at the beta-carbon, which blocks beta- oxidation.
- Major dietary methylated fatty acid is phytanic acid.
- It is derived from phytol present in chlorophyll, milk & animal fats.

Refsum's disease

- Due to deficiency of the enzyme α-hydroxylase (phytanic acid oxidase)
- α – oxidation does not occur.
- Phytanic acid does not converted into compound that can be degraded by beta – oxidation.
- Phytanic acid accumulates in tissues.

Symptoms

- Severe neurological symptoms, polyneuropathy, retinitis pigmentosa, nerve deafness & cerebellar ataxia.
- Restricted dietary intake of phytanic acid (including milk-is a good source of phytanic acid)

Omega- oxidation

- Minor pathway, takes place in microsomes.
- Catalyzed by hydroxylase enzymes involving NADPH & cytochrome P-450.
- Methyl (CH_3) group is hydroxylated to CH_2OH & subsequently oxidized with the help of NAD^+ to COOH group to produce dicarboxylic acids.
- When β-oxidation is defective & dicarboxylic acids are excreted in urine causing dicarboxylic aciduria.

Zellweger syndrome

- It is a rare disorder.
- It is characterized by absence of peroxisomes in almost all the tissues.
- Long chain fatty acids are not oxidized.
- Long chain fatty acids are accumulated in tissues-mainly brain, liver & kidney.
- It also known as cerebrohepatorenal syndrome.

References

- Textbook of Biochemistry-U Satyanarayana
- Textbook of Biochemistry-DM Vasudevan