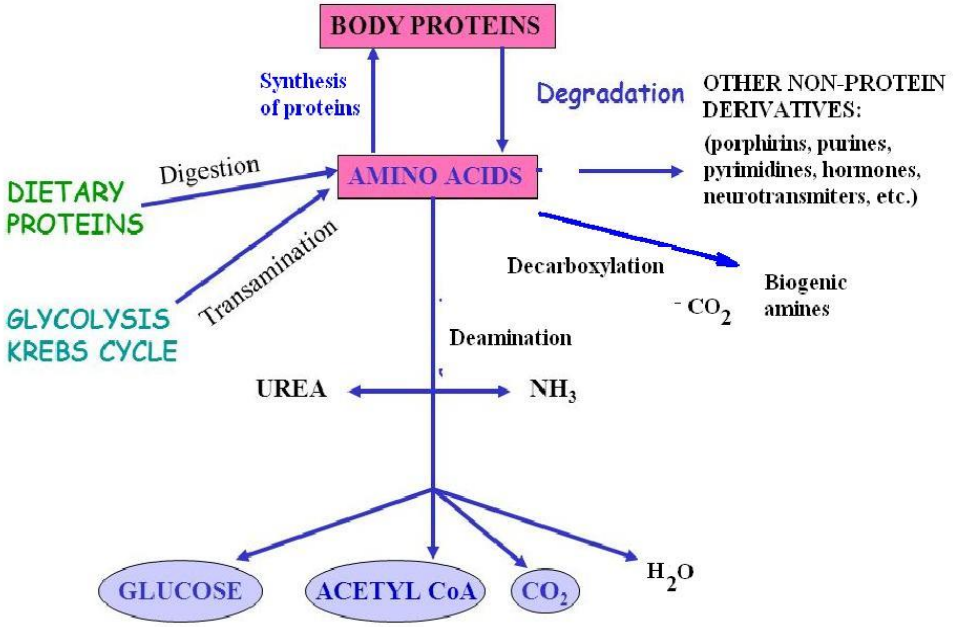
General pathways of amino acid metabolism



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QUESTIONS

1. Dynamic state of body proteins. Nitrogen balance.
2. Sources of amino acids in the body and ways of their use.
3. Digestion of proteins in the gastrointestinal tract. Absorption of amino acids.
4. Intestinal putrefaction of proteins (conversion of amino acids by intestinal bacteria).
5. General pathways of amino acid metabolism.
6. Transamination of amino acids, enzymes, biological role. Coenzyme function of vitamin B6. Mechanism of transamination. Aminotransferases, their tissue specificity and diagnostic significance.
7. Types of deamination of amino acids. Oxidative deamination and reductive amination. Biological role.
8. Transdeamination. Biological role.

Metabolism of proteins and amino acids

- **Refers to all chemical and physical transformations of proteins and amino acids in the body.**

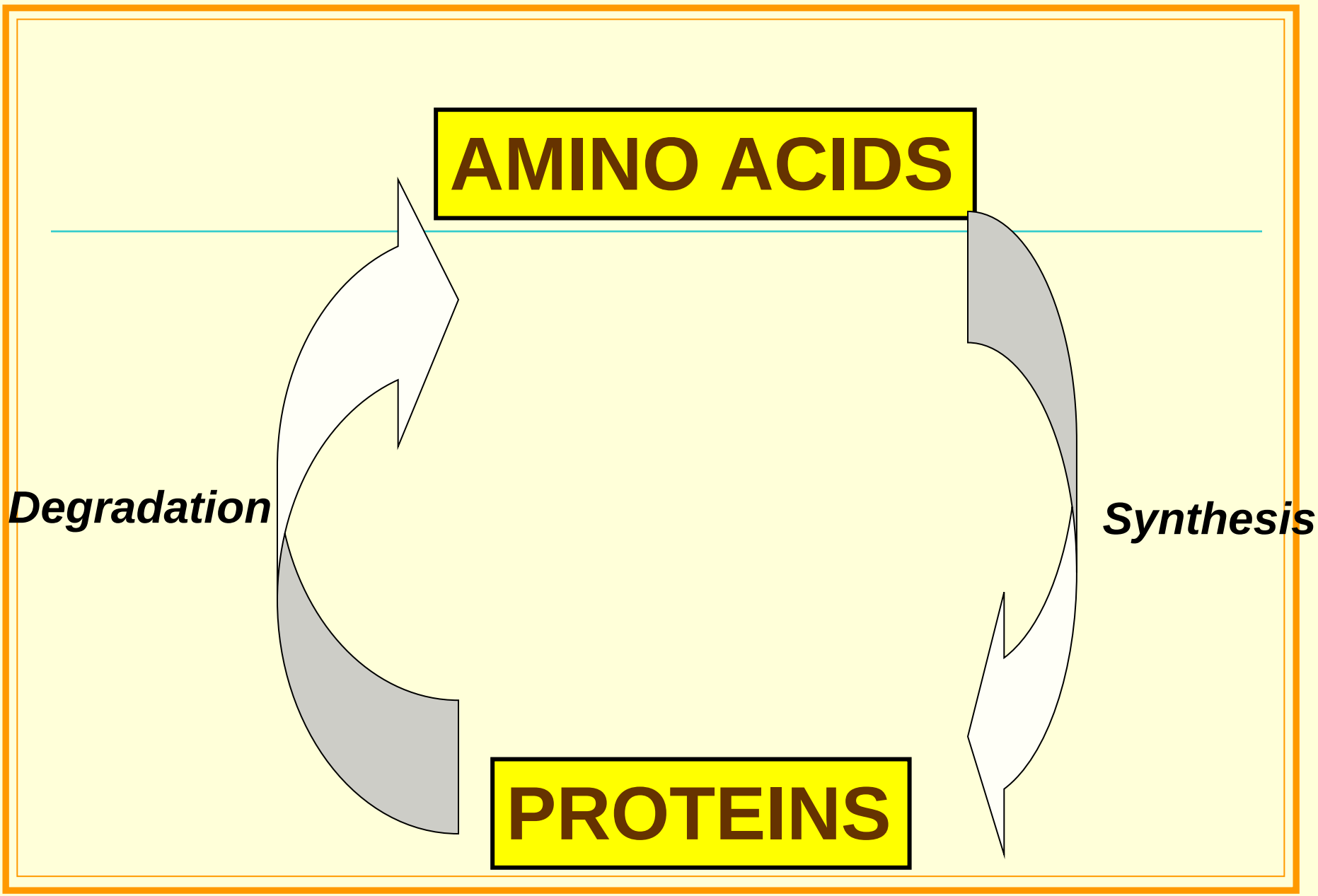
- **represents all the enzymatic reactions within the cells and gastrointestinal tract.**

"Dynamic state of body proteins"

- Once formed, proteins only exist for a certain period and are then degraded and recycled by the cell's machinery through the process of protein turnover
- A protein's lifespan is measured in terms of its half-life and covers a wide range.
- They can exist for minutes or years with an average lifespan of 1–2 days in mammalian cells.

"Dynamic state of body proteins"

- The body's high level of protein turnover is due to the fact that many proteins are relatively short-lived (hormones, enzymes).
- By contrast, structural proteins such as the histones, hemoglobin, and the components of the cytoskeleton are particularly long-lived.
- Abnormal or misfolded proteins are degraded to amino acids more rapidly either due to being targeted for destruction, or due to being unstable.


AMINO ACIDS
Degradation
Synthesis
PROTEINS

- **Nitrogen balance** is the traditional method of determining **dietary protein requirements.**

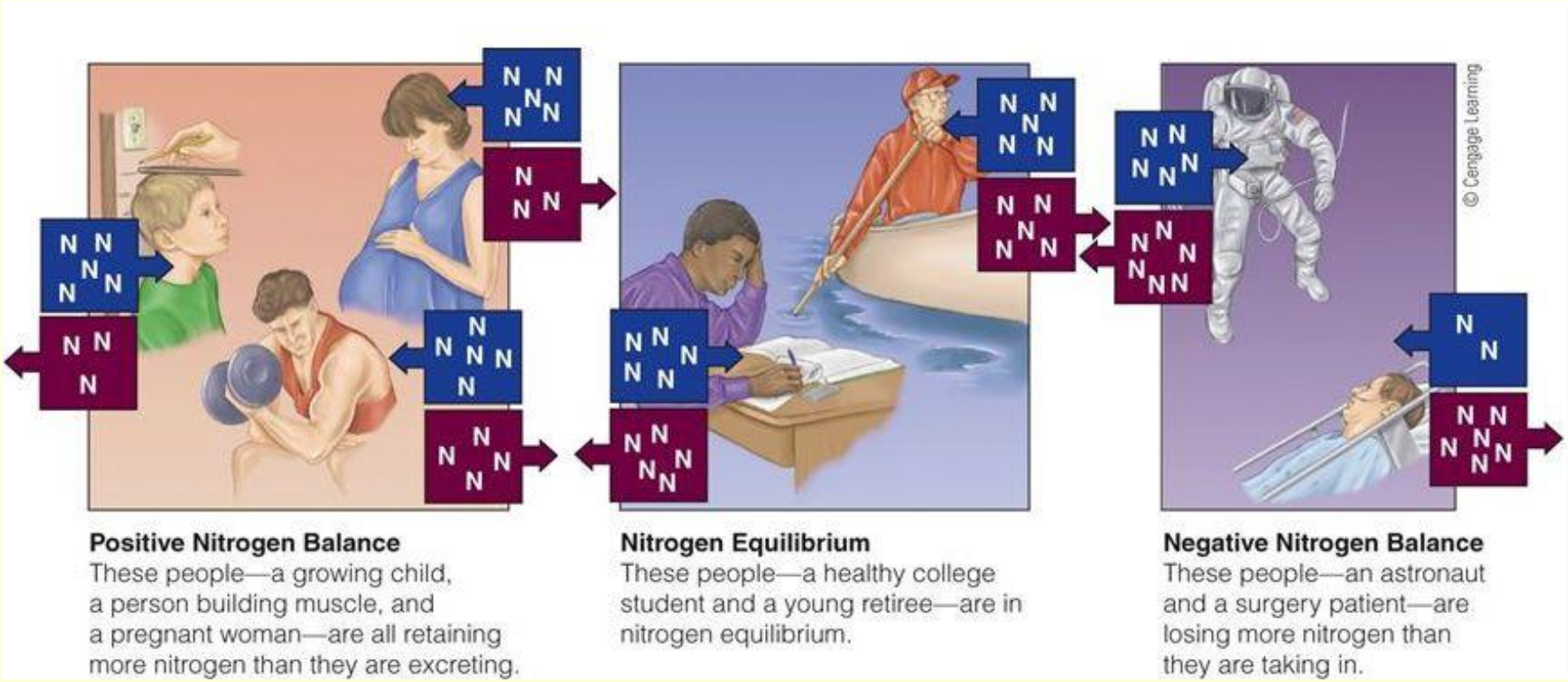
- Determining **dietary protein requirements** using nitrogen balance requires that all nitrogen inputs and losses are carefully collected, to ensure that all nitrogen exchange is accounted for.

Nitrogen balance

- is defined as the difference between **Intake** and **Output**, and may be formally represented by the following equation:

NITROGEN BALANCE = **NITROGEN INTAKE** *(with dietary proteins)* – **NITROGEN LOSS** *(through urine, feaces, skin)*

3 types of Nitrogen Balance



Positive Nitrogen Balance
These people—a growing child, a person building muscle, and a pregnant woman—are all retaining more nitrogen than they are excreting.

Nitrogen Equilibrium
These people—a healthy college student and a young retiree—are in nitrogen equilibrium.

Negative Nitrogen Balance
These people—an astronaut and a surgery patient—are losing more nitrogen than they are taking in.

Positive nitrogen balance

- Intake of nitrogen into the body is greater than the loss of nitrogen from the body, so there is an increase in the total body pool of protein.
- Is observed in:
  - Growing children.
  - Pregnant women.
  - Persons building muscles.
  - Patients with hypothyroidism (low level of thyroid hormones).
  - During tissue repair.

Negative nitrogen balance

- The amount of nitrogen excreted from the body is greater than the amount of nitrogen ingested.
- Is associated with
  - burns,
  - serious tissue injuries,
  - fevers, hyperthyroidism,
  - wasting diseases, starvation, and malnutrition
  - In elderly people
  - In astronauts
  - In surgery patients

Nitrogen equilibrium

- Intake of nitrogen into the body is equal to the loss of nitrogen from the body

- Observed in healthy adults eating balanced diet

Dietary proteins

proteins which we take in our diet are either from animal source or vegetable source

- ***Principal animal sources:***

milk ,meat, fish, liver, eggs.

- ***Principal vegetable sources:***

cereals, pulses, peas, beans and nuts.

The minimum daily requirement of protein is 37 g for men and 29 g for women, but the recommended amounts **80-100 g**.
Requirements in pregnant and breastfeeding women are higher.

10 Essential amino acids (our body cannot make them!)	10 Non-essential amino acids (synthesized in the human body)
Methionine	Glycine
Tryptophan	Alanine
Threonine	Glutamate
Valine	Glutamine
Leucine,	Aspartate
Isoleucine,	Asparagine
Phenylalanine,	Proline
Lysine,	Tyrosine
Histidine*	Cysteine
Arginine*	Serine

** essential only for children*

Digestion of proteins in GIT and cells

- Takes place both in cells and gastrointestinal tract (GIT).
- Is carried out by proteolytic enzymes (proteases and peptidases) that hydrolyze proteins to small peptides or amino acids.

Proteases are of 2 types

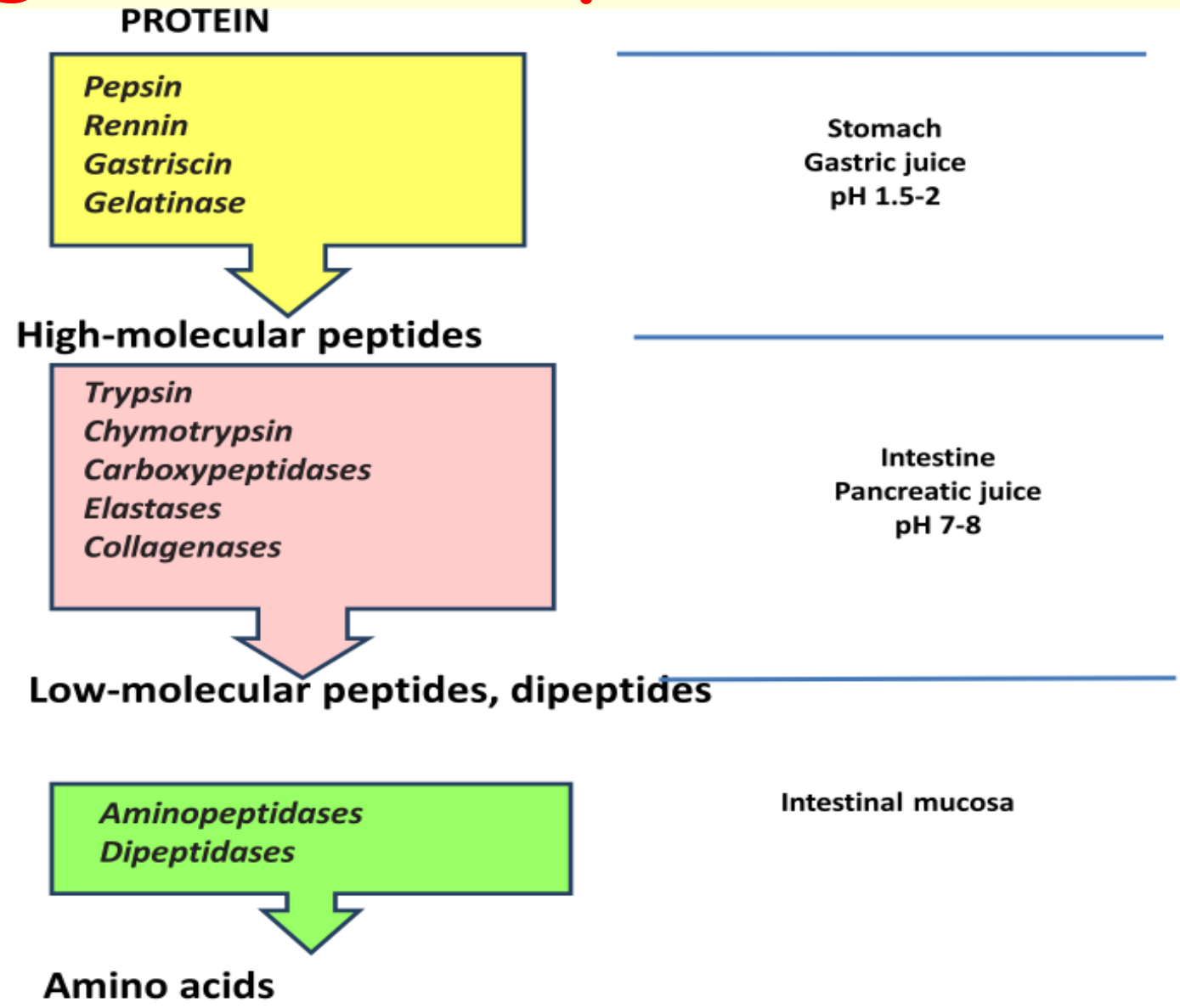
- **Endopeptidases.** Hydrolyze peptide bonds between specific AA throughout the protein molecule, yielding a number of peptide fragments.

- **Exopeptidases.** Hydrolyze peptide bonds from the ends of polypeptide chains to form mixture of AA in completed hydrolysis.

- In cells degradation of proteins is catalyzed by **lysosomal proteases.**

Endopeptidases	Exopeptidases
Gastric juice: – Pepsin, – Gastricsin, – Rennin •**Pancreatic juice:** – Trypsin, – Chemotrypsin, – Elastase	**Pancreatic juice:** – Carboxypeptidases **Intestinal mucosa:** – Aminopeptidases, – Tripeptidases, – Dipeptidases

Degradation of proteins in GIT



Activation of digestive proteases

- They are synthesized in cells as inactive zymogens secreted to GIT.
- The active site of the enzyme is masked by a small region of the peptide chain that is removed by hydrolysis.
  - Pepsinogen → Pepsin
  - Trypsinogen → Trypsin
  - Chymotrynsinogen → Chymotrypsin
  - Proelastase → Elastase
  - Procarboxypeptidase → Carboxypeptidase
  - Proaminopeptidase → Aminopeptidase

The Production and Action of Pepsin

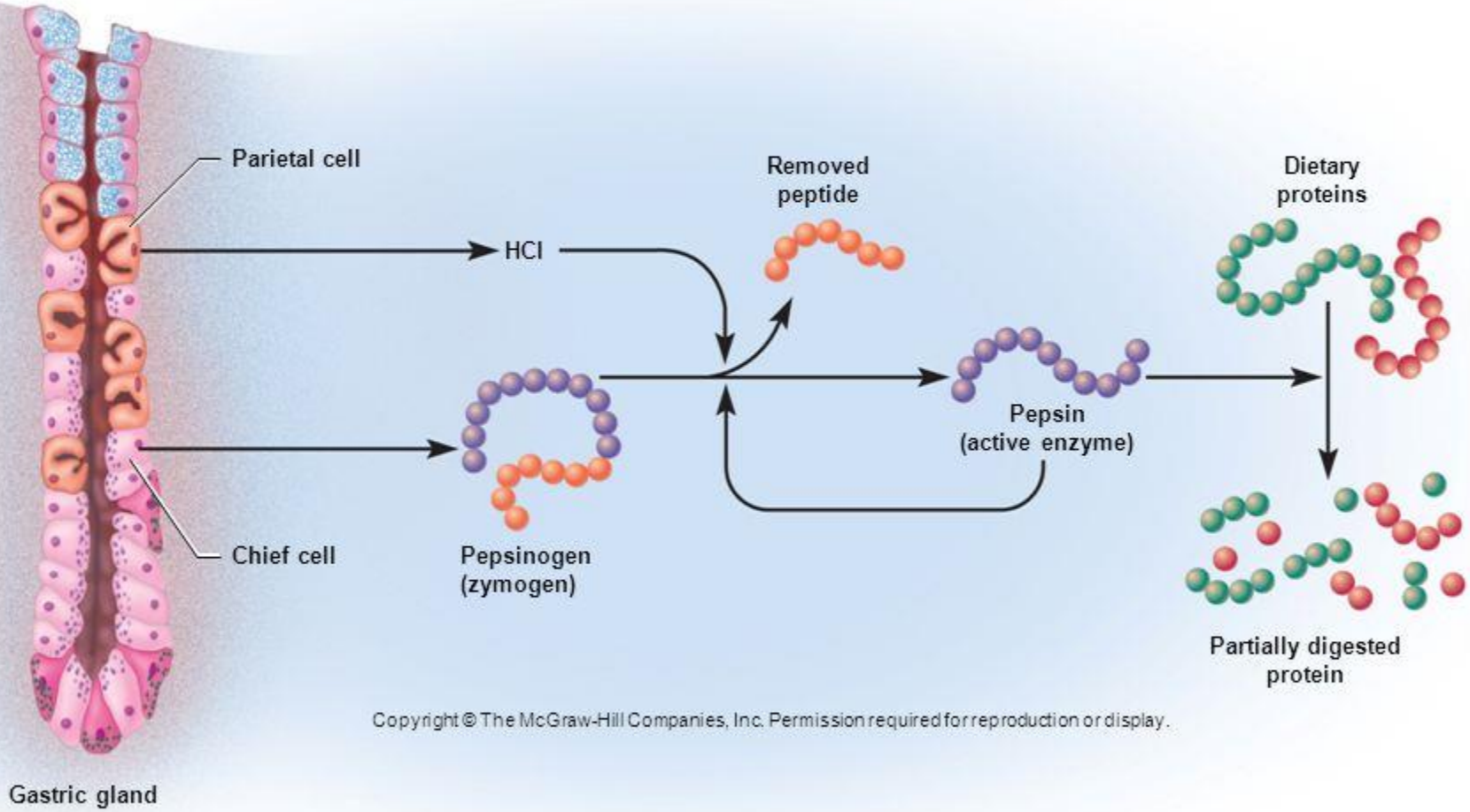


Figure 25.15

Absorption of amino acids

- In digestion proteins are degraded into a mixture of free amino acids, di- and tripeptides, and oligopeptides.

- Free AA are absorbed across the intestinal mucosa by Na-dependent active transport.

- Small peptides (tri- and dipeptides) are absorbed by enterocytes, where they are cleaved to AA which are than transported to the blood capillaries.

2 systems of absorption of amino acids

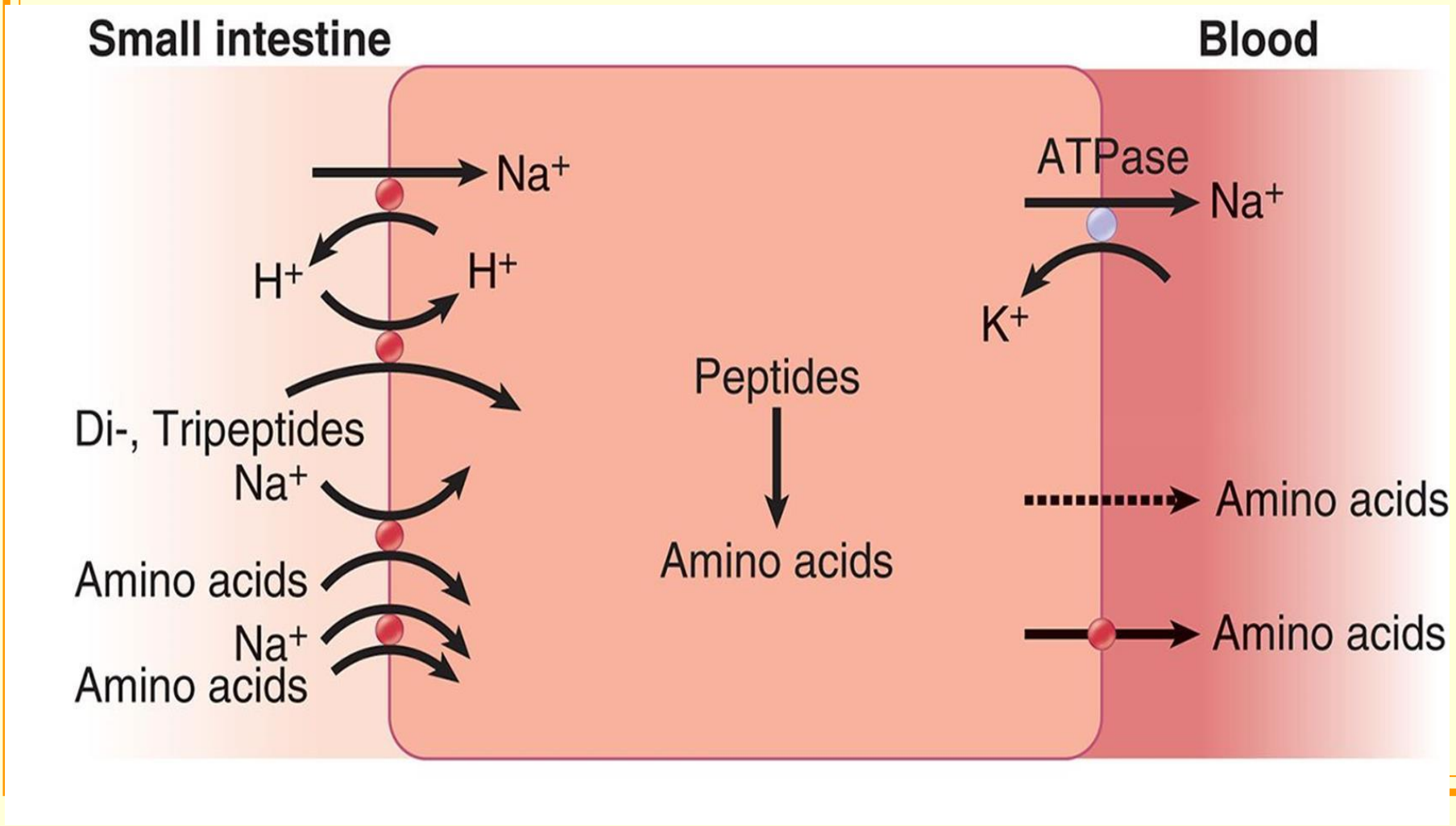
I. Carrier protein transport system (in GIT and cells)

II. Glutathione transport system or γ-Glutamyl cycle (in cells)

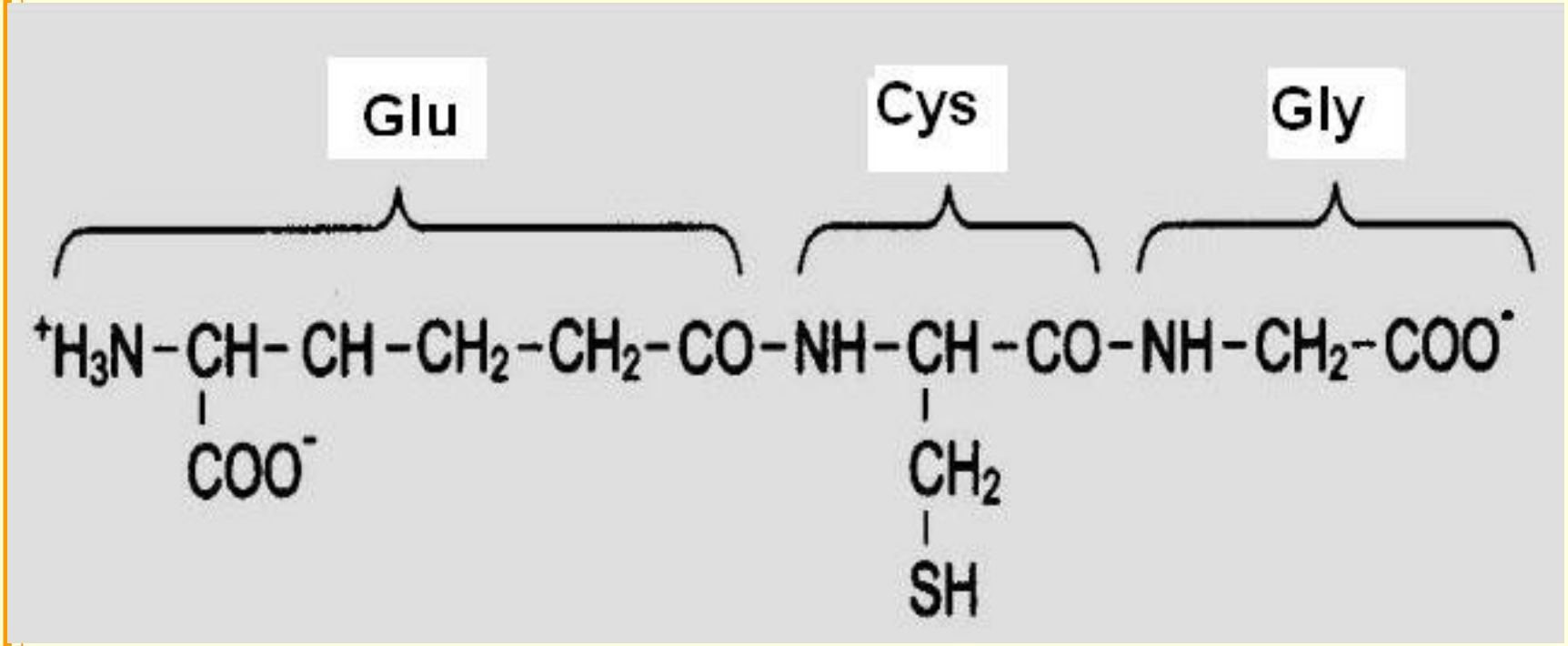
I. Carrier protein transport system for absorption of AA

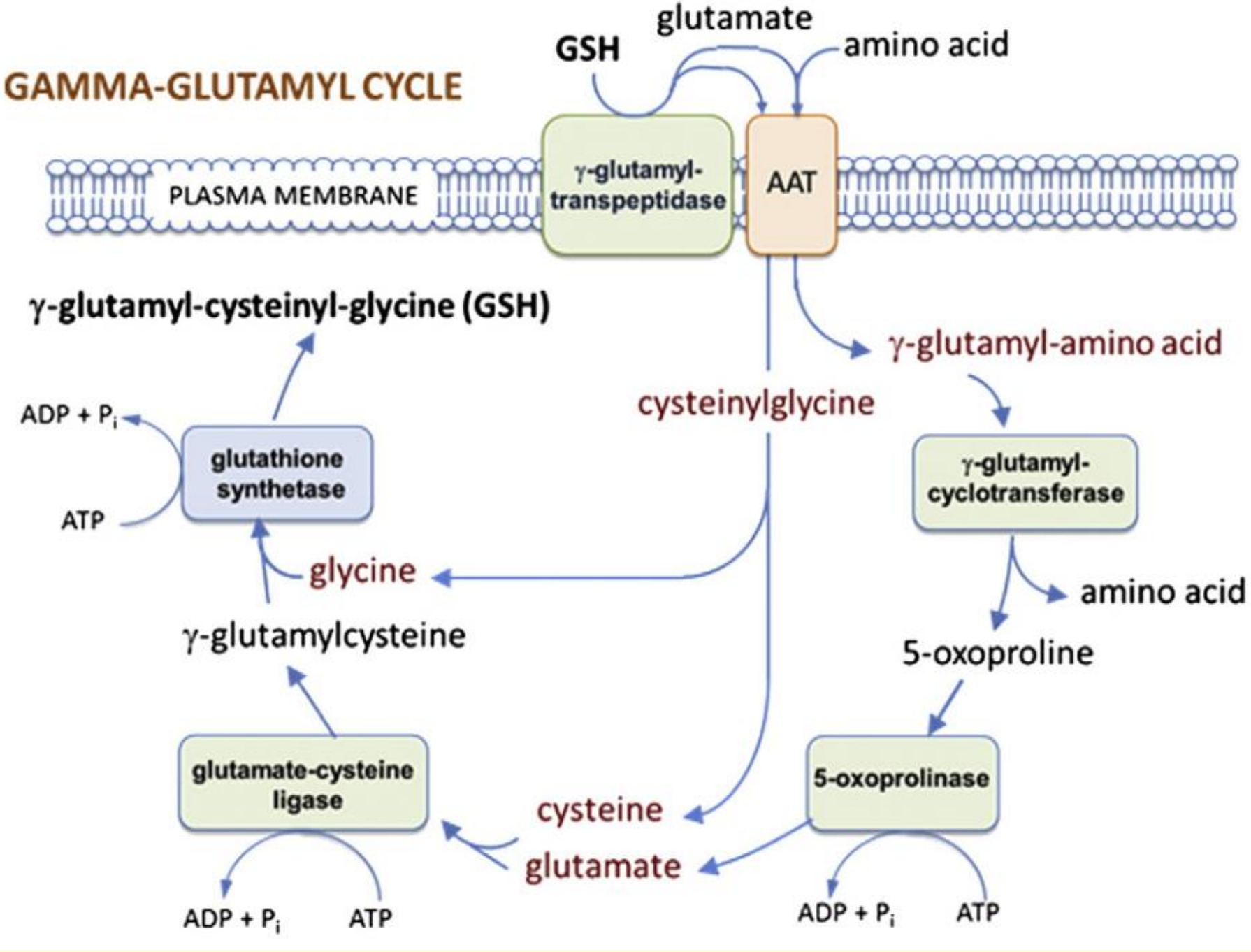
- Is an energy requiring process.
- Transport systems are carrier-mediated or/and ATP-Na-dependent symport systems.
- 5 different carriers for AA:
  1. Neutral AA (Ala, Val, Leu, Met, Phe, Ile)
  2. Basic AA (Arg, Lys) and Cys.
  3. Imino acids (Pro) and Gly.
  4. Acidic AA (Glu, Asp)
  5. Beta amino acids (β-alanine).

Absorption of AA is an active process and requires energy of ATP



II. Glutathione transport system requires Glutathione (γ-Glutamyl-cysteinyl-glycine)




GAMMA-GLUTAMYL CYCLE
GSH
glutamate
amino acid
PLASMA MEMBRANE
γ-glutamyl-transpeptidase
AAT
γ-glutamyl-cysteinyl-glycine (GSH)
γ-glutamyl-amino acid
cysteinylglycine
ADP + Pi
ATP
glutathione synthetase
γ-glutamyl-cyclotransferase
glycine
amino acid
γ-glutamylcysteine
5-oxoproline
glutamate-cysteine ligase
5-oxoprolinase
cysteine
glutamate
ADP + Pi
ATP
ADP + Pi
ATP

Conversion of amino acids under the action of intestinal microflora

- Gut flora consist of a complex microorganism species that live in the digestive tract of animals (Microbiota).

- These bacteria make up most of the flora in the colon, and the number of m/o there 10 time greater than the total number of human cells in the body.

- In the colon undigested and unabsorbed peptides and amino acids are fermented by bacterial enzymes into toxic and neutral compounds.

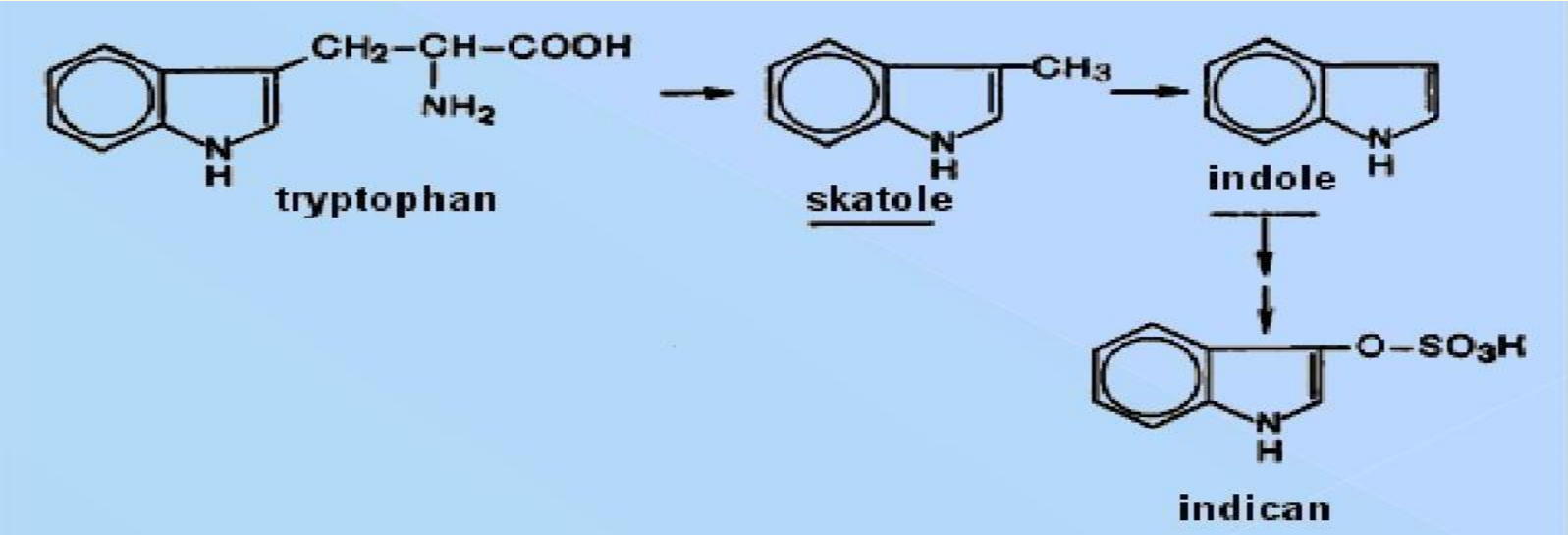
- Toxic products of amino acid metabolism are than detoxified in the liver.

Conversion of amino acids under the action of intestinal microflora

- Conversion of sulfur-containing amino acids to hydrogen sulfide (H_2S) and methylmercaptan (CH_3–SH), the products which are removed from the intestine with intestinal gas.

- Putrefaction of diaminomonocarboxylic acids to amines:
  - Ornithine → Putrescine
  - Lysine → Cadaverine
  - Putrescine and Cadaverine are detoxified in enterocytes by diaminoxidases.

Conversion of aromatic amino acids by microbiota

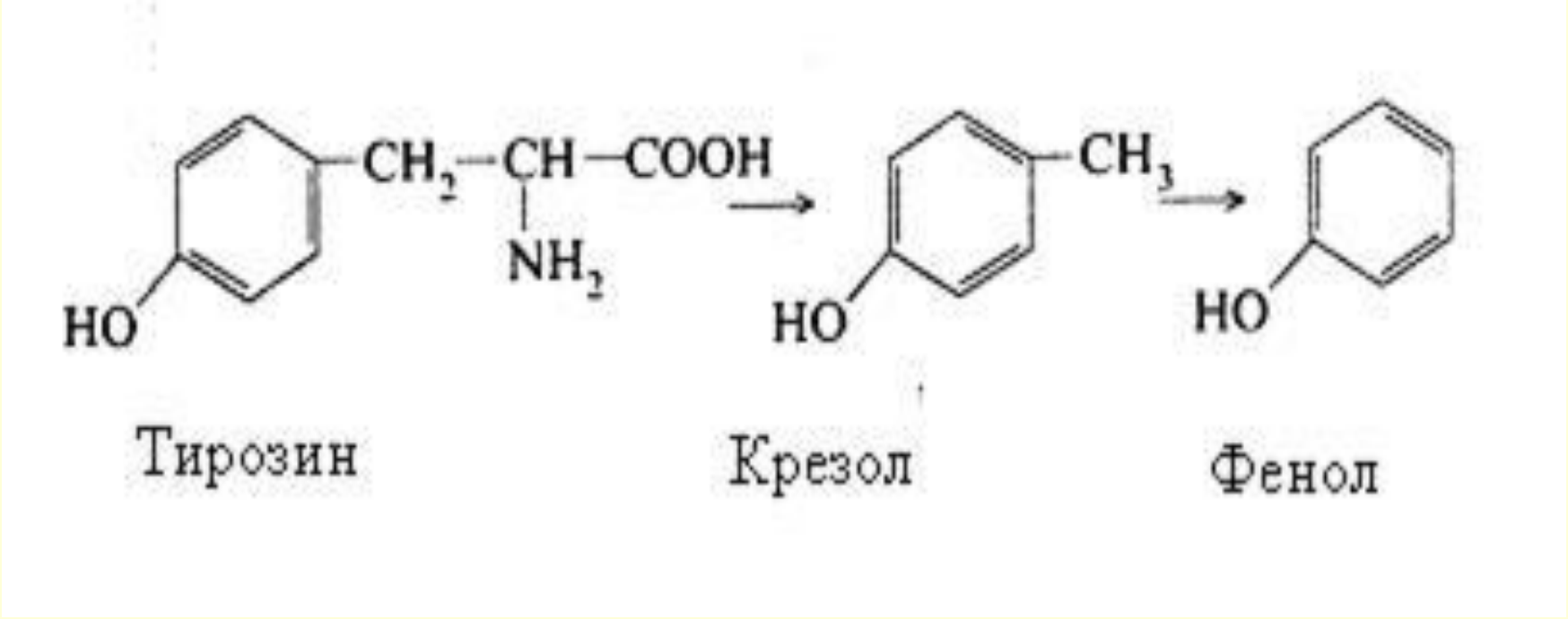


Increased level of indican in the blood may points to renal failure because of decreased urination.

Blue diaper syndrome

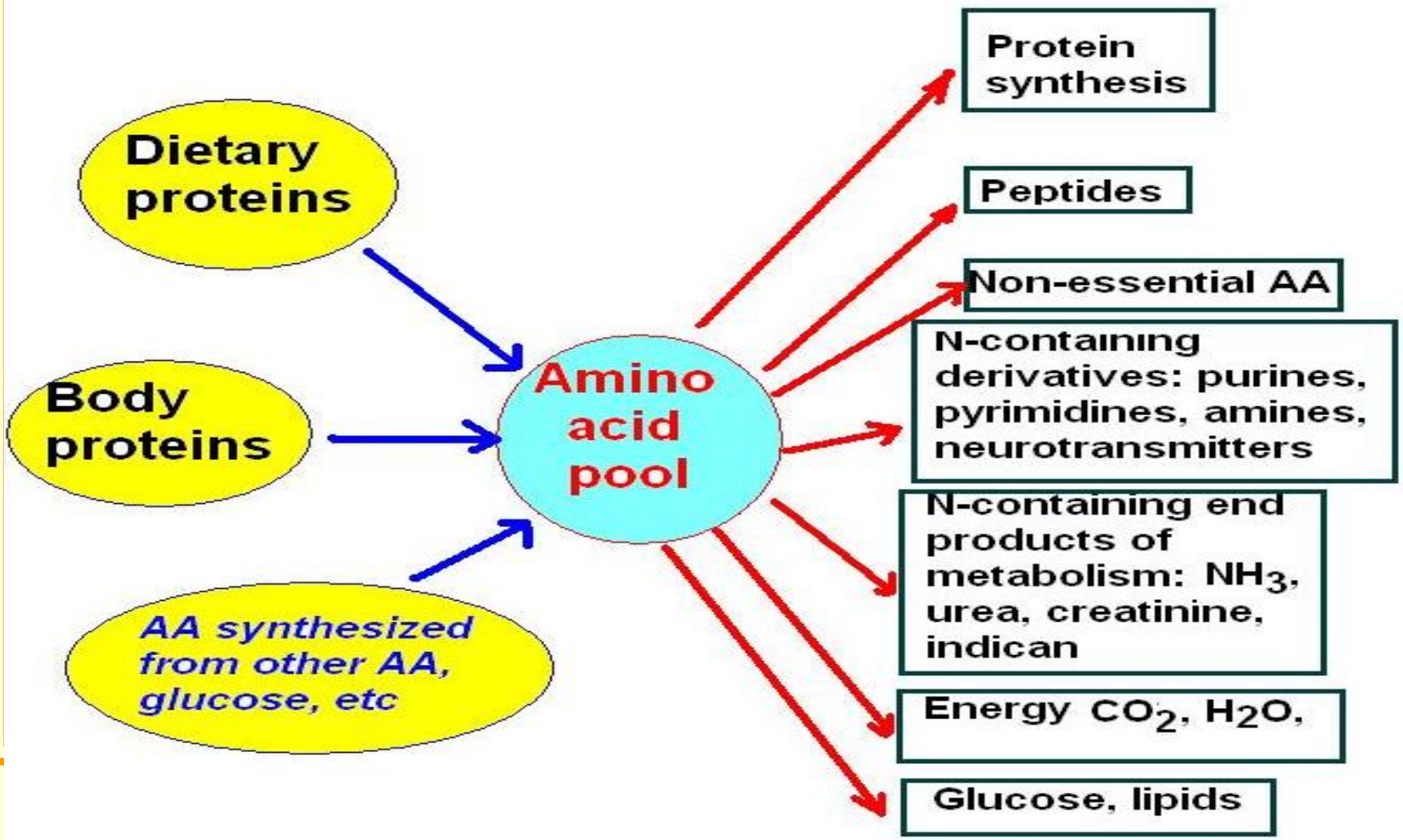
- is a rare, autosomal recessive metabolic disorder resulted from a defect in tryptophan metabolism.
- is characterized in infants by [illegible] urine-stained diapers.
- Indican is excreted into the urine and from there into the diaper where, upon exposure to air, it is converted to indigo blue dye due to oxidation by atmospheric oxygen.

Tyrosine ⟶ Cresol ⟶ Phenol



- **Cresol, phenol, and other toxic products are than detoxified in the liver**

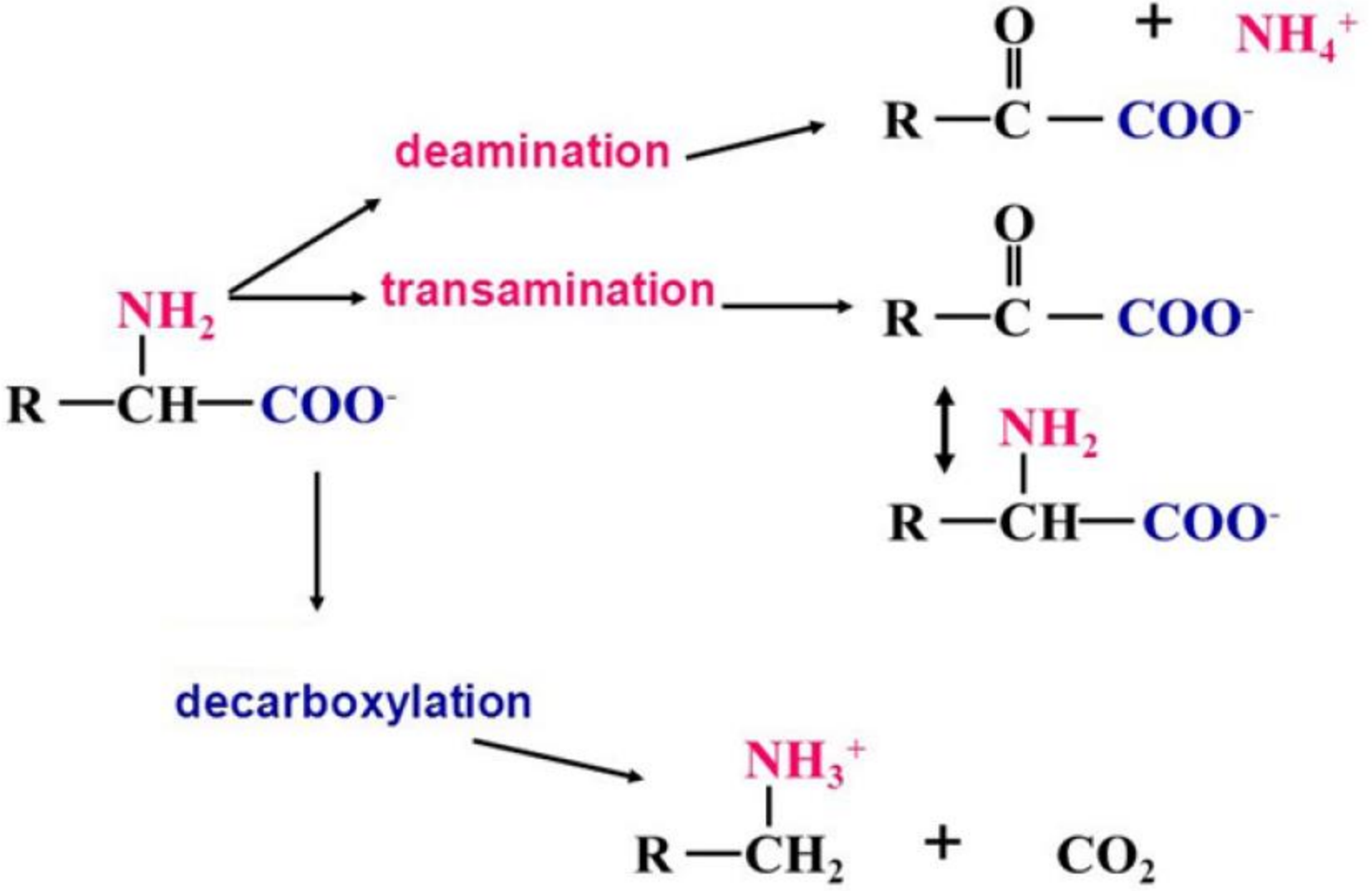
Sources of amino acids in the body and their use



General pathways of amino acid metabolism

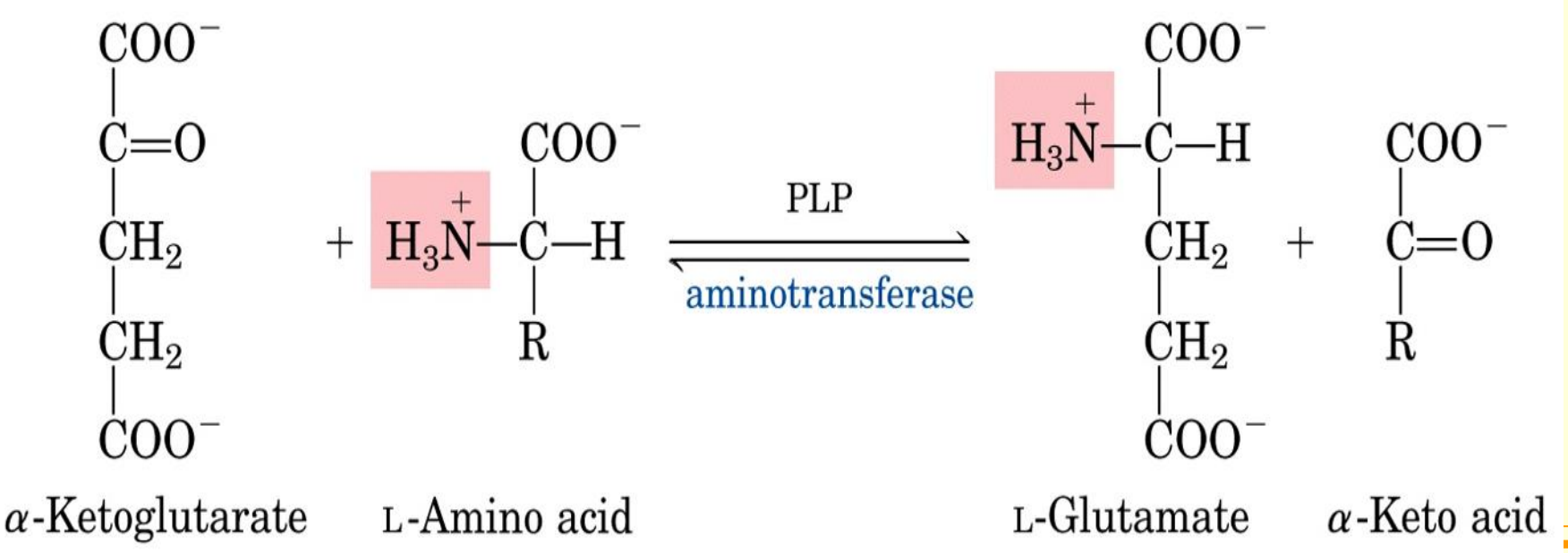
- 20 proteinogenic amino acids are converted by their own metabolic pathways.
- Most of AA can be metabolized through following general catabolic pathways:
  1. Transamination
  2. Deamination
  3. Decarboxylation
  4. Polymerization (protein synthesis)
  5. Synthesis of non-essential AA by modification of their side chains.

General reactions of amino acids metabolism



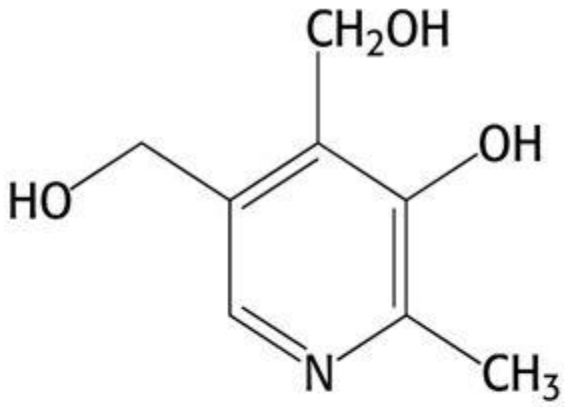
TRANSAMINATION

- Transfer of $-NH_2$ of an Amino Acid to an α-Keto Acid (usually α-ketoglutaric acid).
- Enzymes: aminotransferases (transaminases)



Mechanism of transamination

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

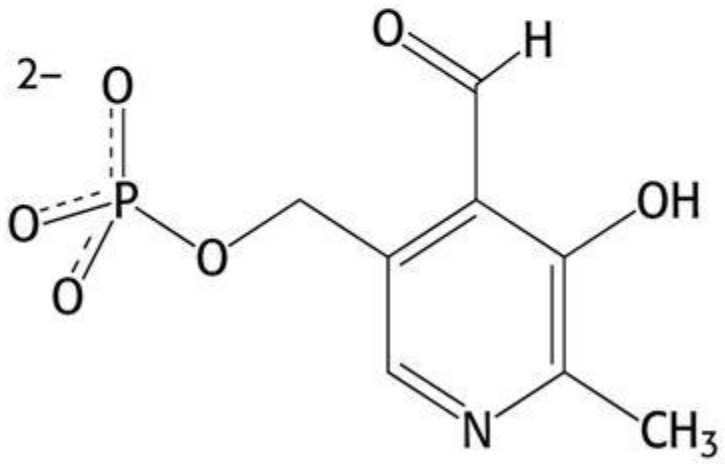


Pyridoxine (Vitamin B_6)

Ping-pong kinetic mechanism

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

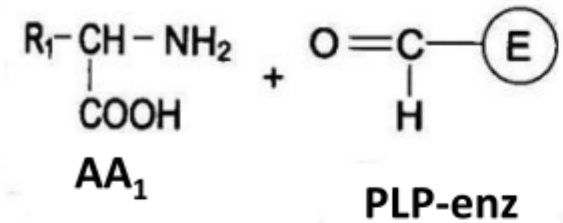
Second step: α-ketoglutarate reacts with pyridoxamine phosphate forming glutamate



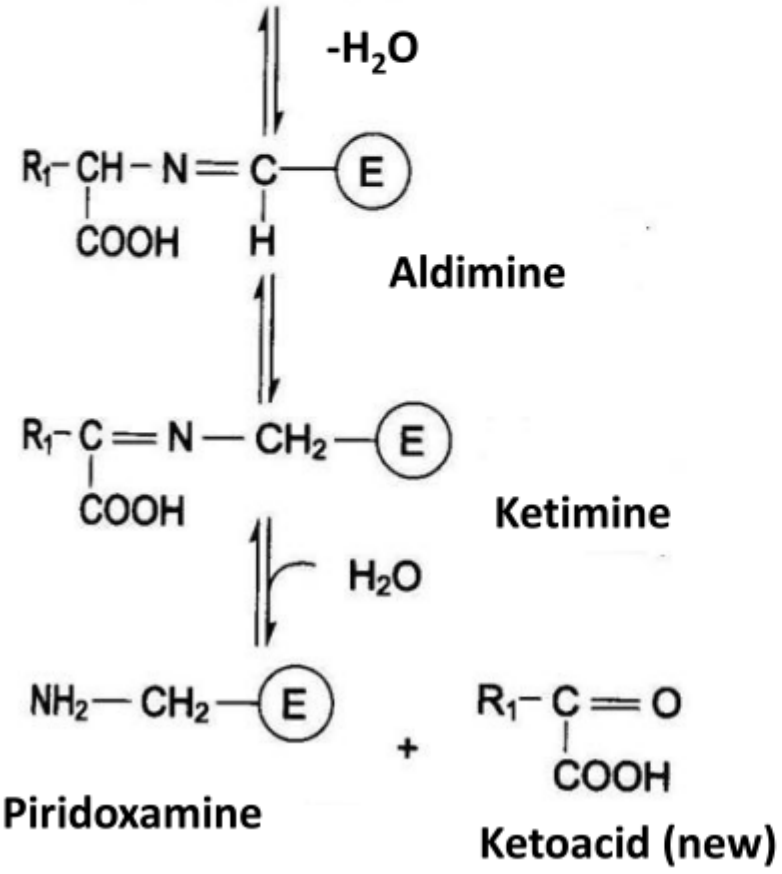
Pyridoxal phosphate (PLP)

Mechanism of transamination

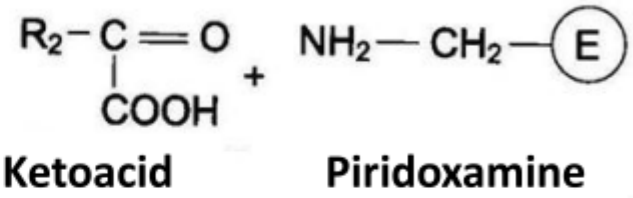
1st phase



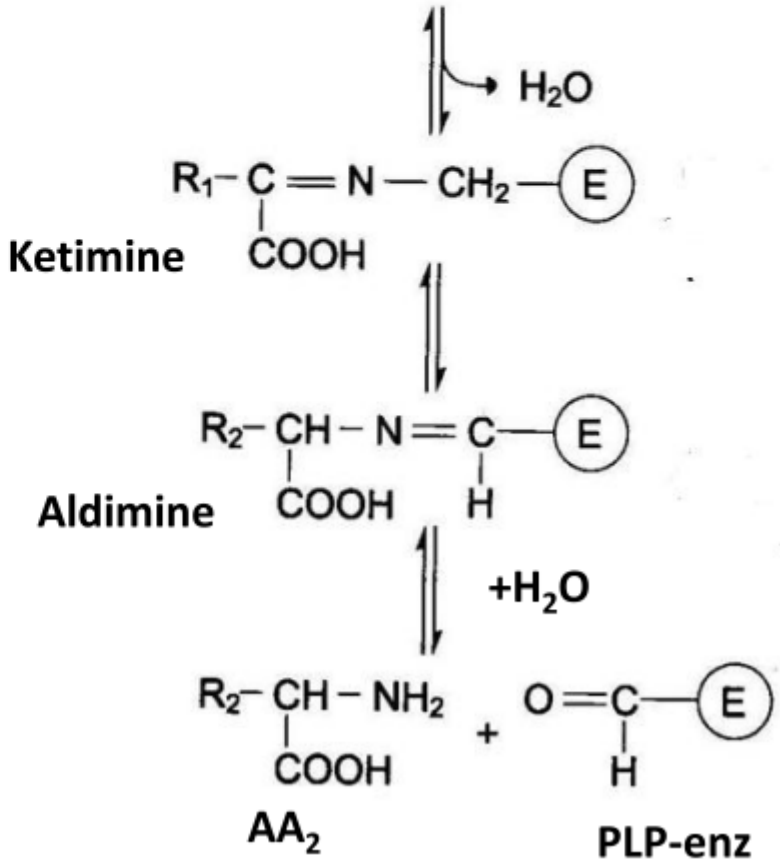
AA_1 PLP-enz



2nd phase



Ketoacid Piridoxamine



AA_2 PLP-enz

Biological role of transamination

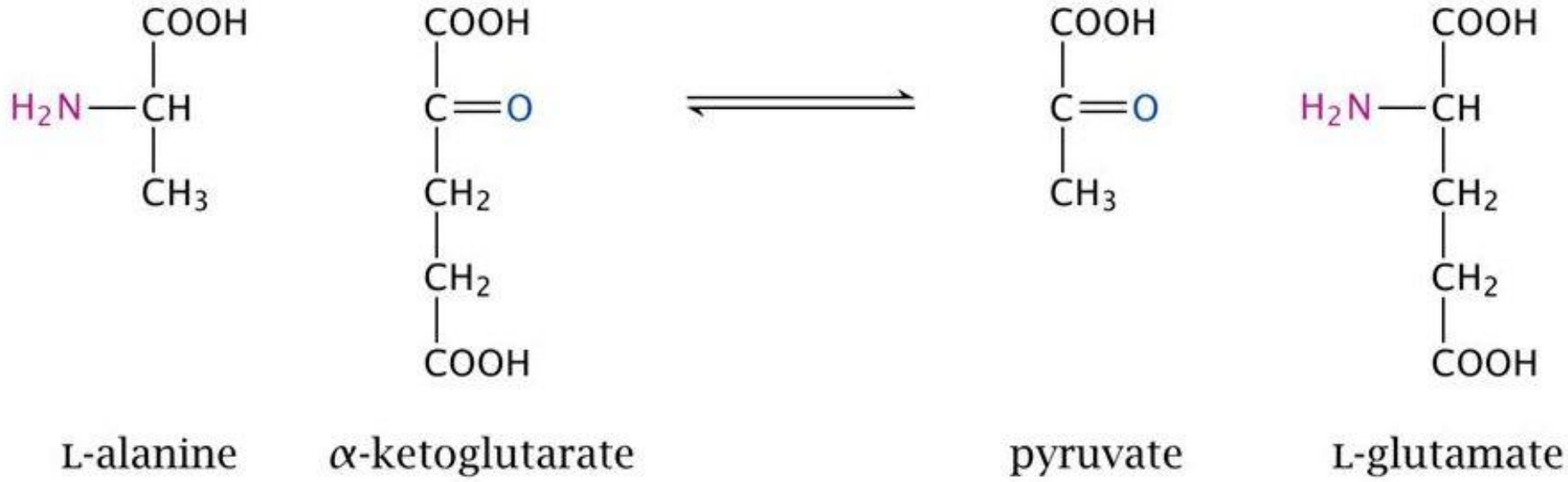
- All amino acids (except LYSINE and THREONINE) can be transaminated.
- Each aminotransferase is specific to one or few amino acids.
- Production of non-essential amino acids.
- Resulted α-Keto acids can be completely oxidized to CO_2 and H_2O with release of energy, or converted to glucose, fats, ketone bodies.
- Transamination reactions are active in many tissues, including the liver, kindey, brain, pancreas, red blood cells, etc.

Clinical significance of some transaminases

- Activity of aminotransferases is high in tissues, but low in the blood serum.
- In cell destruction or increased cell membrane permeability, transaminases are released from the tissue into the blood plasma.
- Clinical determination of alanine amino transferase (AlAT) and aspartate aminotransferase (AsAT) activity in the blood serum is used for diagnostics of certain diseases.

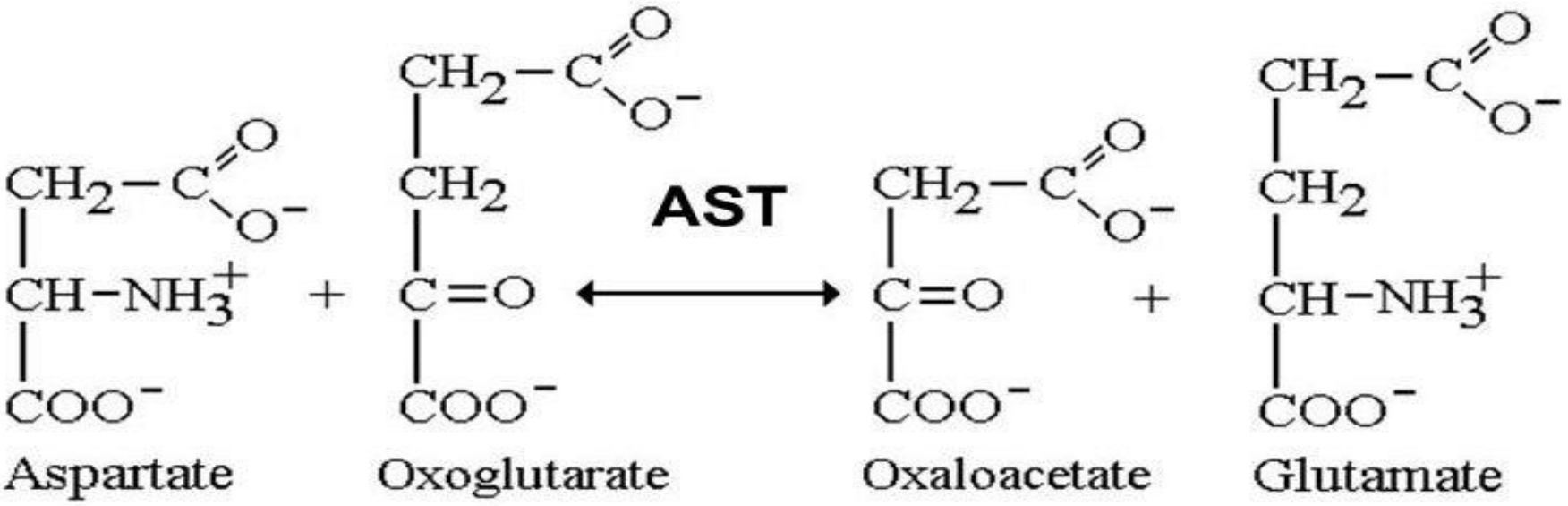
Alanine aminotransferase (AlAT or ALT)

- Normal values in the blood: 0.1-0.68 mmol/l/h
- Increased level y in the blood is commonly associated with hepatocellular injury, as the liver contains most of AlAT.



Aspartate aminotransferase (AsAT or AST)

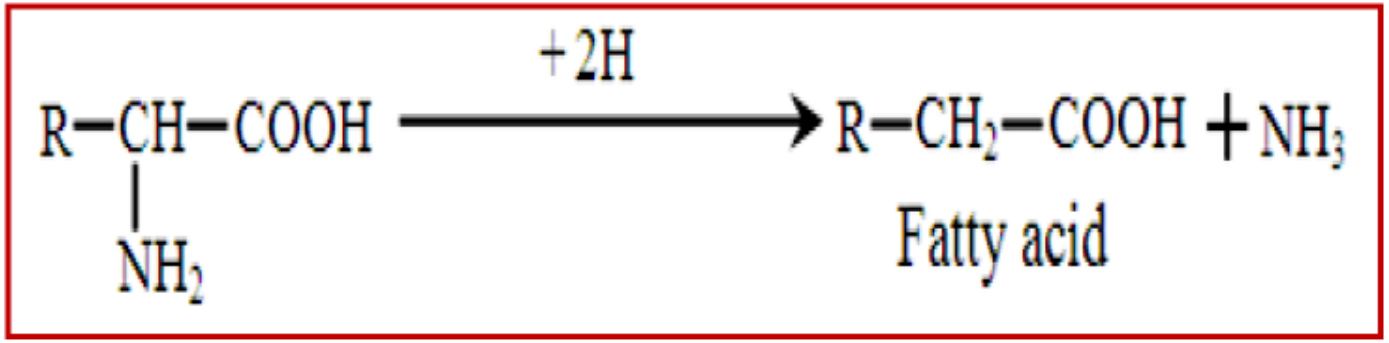
- Reference range in the blood: 0.1-0.45 mmol/l/h
- AsAT is found in the liver, heard, skeletal muscles, kidney, brain, and red blood cells.
- Increased AsAT is indicator of liver diseases, but also elevates in diseases affecting other organs, such as myocardial infarction, acute pancreatitis, severe burns, musculosceletal diseases.



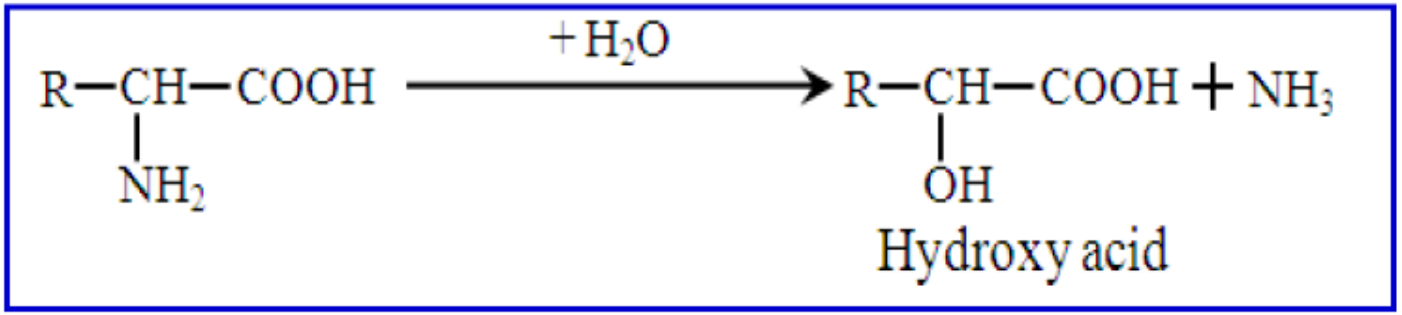
DEAMINATION

- Is the process by which amino group from AA is removed as ammonia (NH_3).
- There are 4 types of deamination.

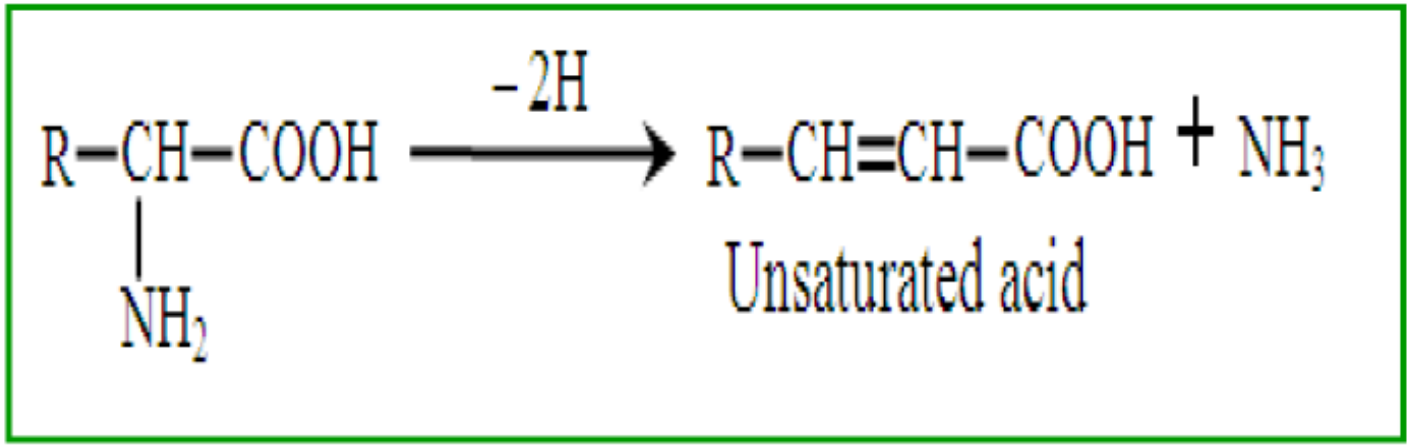
1. Reductive deamination



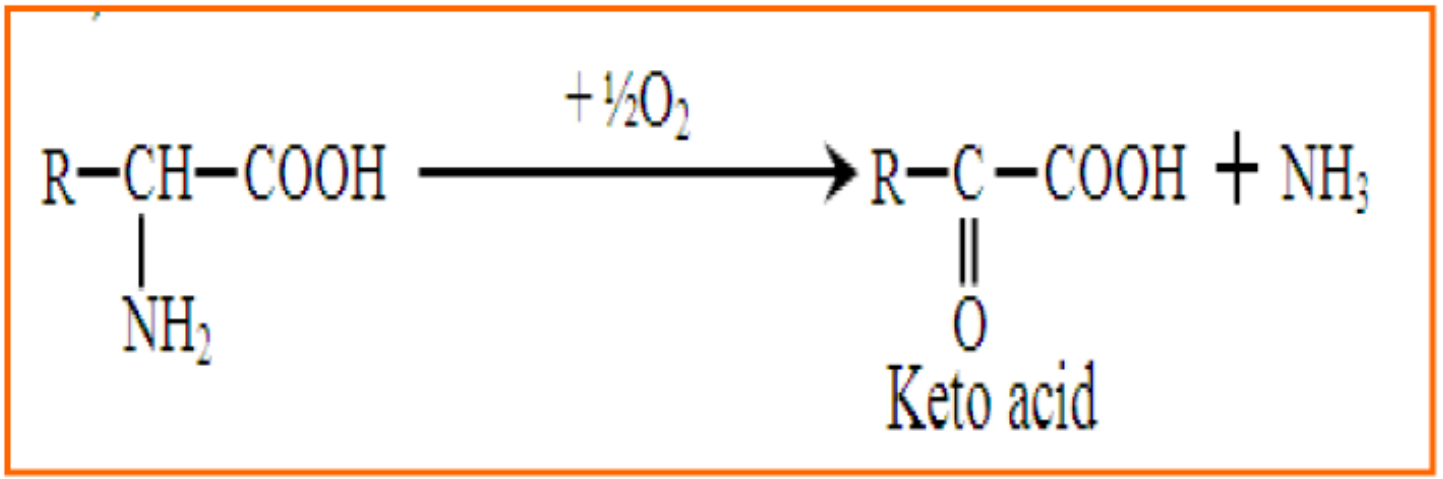
2. Hydrolytic deamination



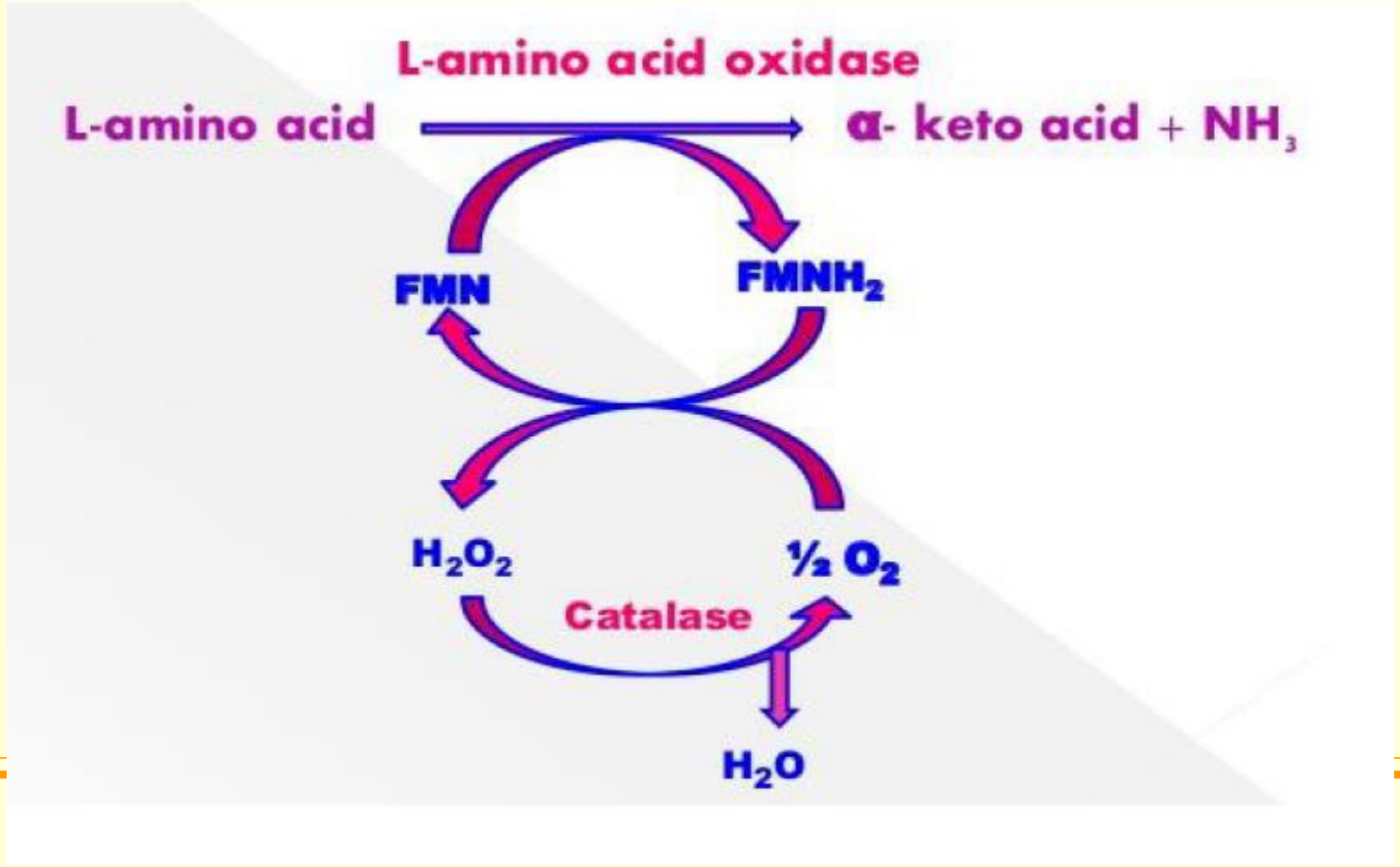
3. Intramolecular deamination



4. Oxidative deamination

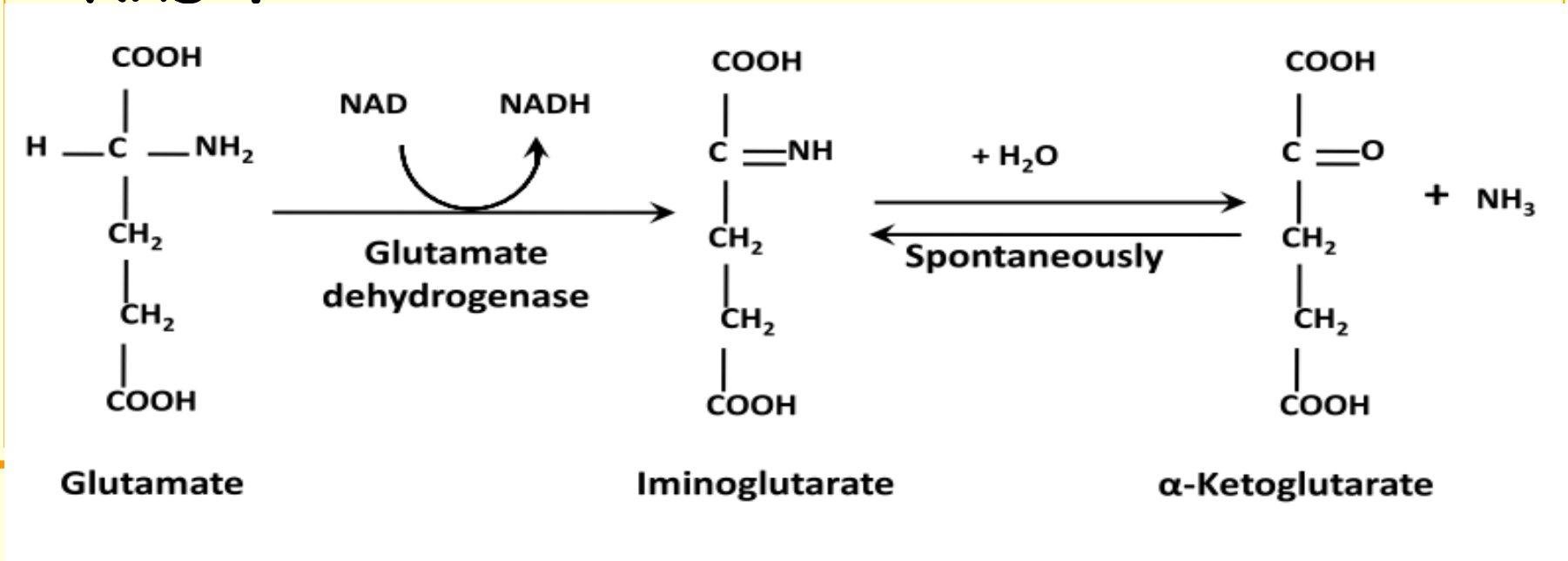


Oxidative deamination of amino acids in humans is catalyzed by oxidases of L-amino acids and D-amino acids. These enzymes are not active in tissues.



Oxidative deamination by Glutamate Dehydrogenase (GDH)

- GDH is the only active enzyme in humans involved in oxidative deamination of AA.
- Glutamate is the only substrate specific to GDH.
- The reaction is reversible. The opposite interaction of ammonia with α-ketoglutarate is called REDUCTIVE AMINATION because of reduction of NADH to NAD+.



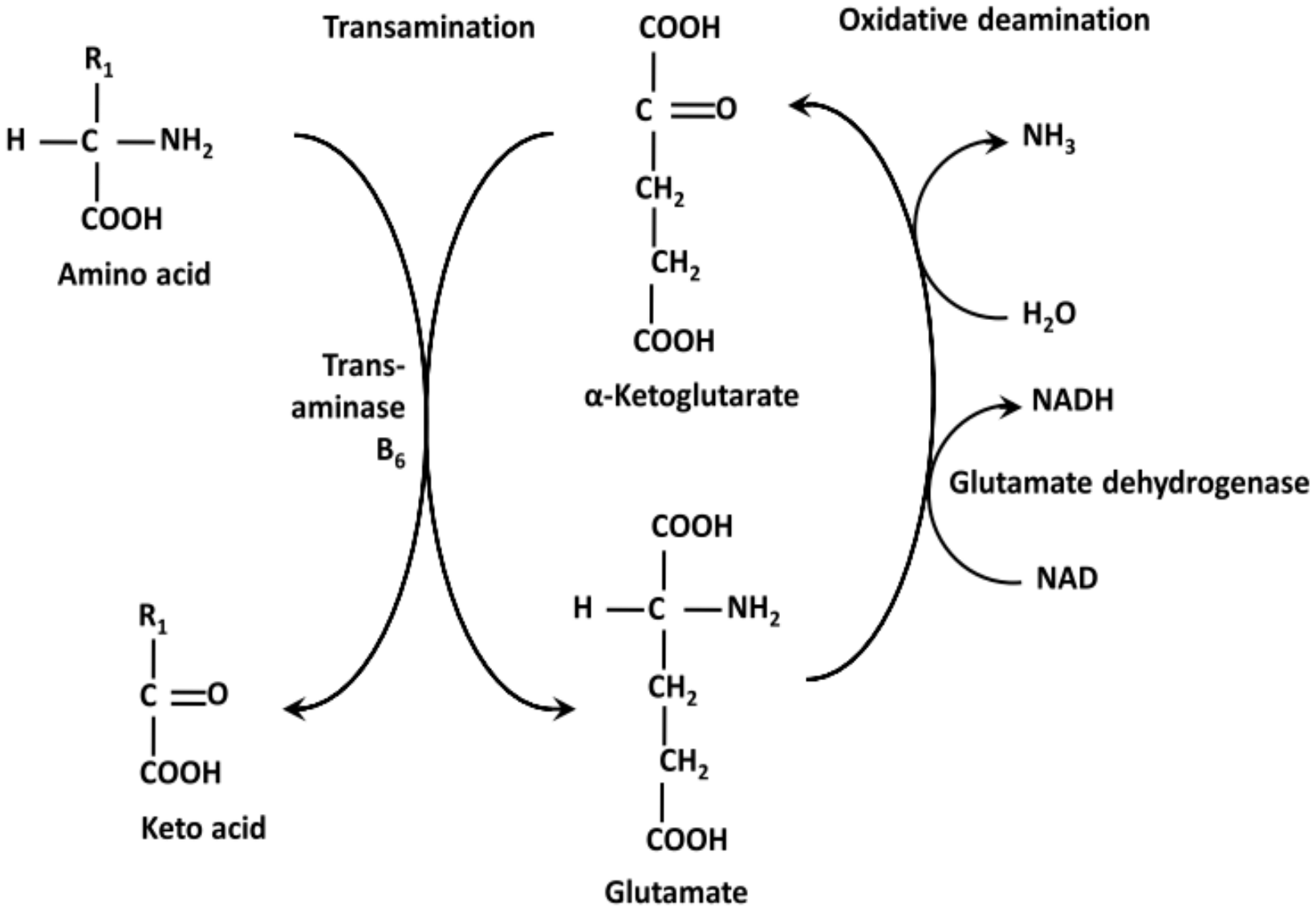
Biological role of Oxidative Deamination and Reductive Amination

- OD of glutamate remove excess of this amino acid from tissues with release of ammonia.
- OD provides indirect deamination of other amino acids in the process called TRANSDEAMINATION.
- RA of α-ketoglutarate is one of the ways in detoxification of ammonia. It also produces glutamate for cell needs.

TransDEamination (or indirect deamination)

- Is indirect removal of ammonia from amino acids (other than glutamate) using coupled action of aminotransferases and glutamate dehydrogenase.
- Biological role: deamination of all amino acids, except lysine and threonine!

Scheme of transdeamination



TransREamination

- Is an reverse process of transdeamination

1. NH_3 + H_2O +α-ketoglutarate + NADH = glutamate + NAD^+

2. glutamate + α-Keto Acid = α-ketoglutarate + AMINO ACID

Biological role of transreamination

- One of the ways of detoxification of ammonia in cells.
- Synthesis of non-essential amino acids.
- In humans synthesis of essential amino acids is limited by the absence of corresponding α-keto acids !