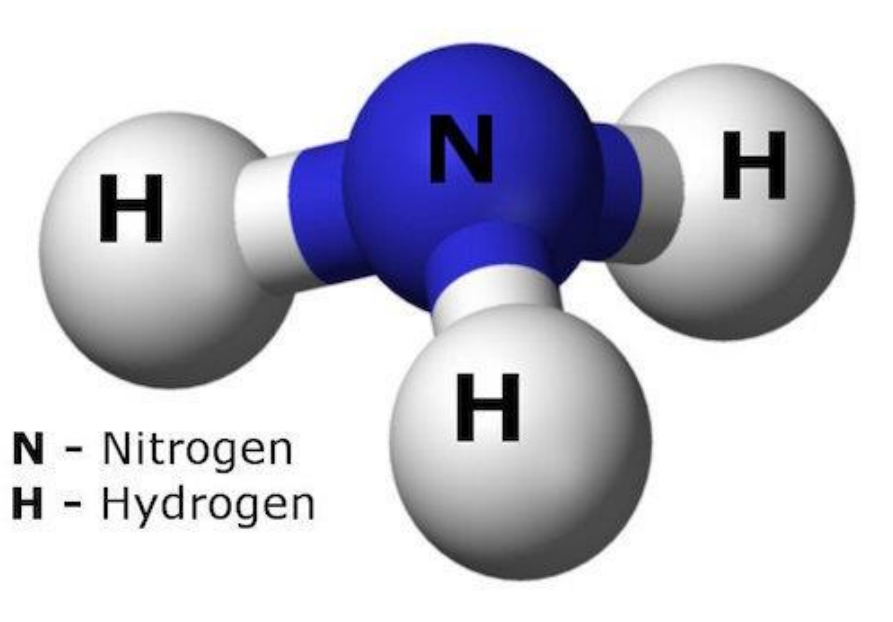
Detoxification of Ammonia



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QUESTIONS

1. Decarboxylation of amino acids. Types of decarboxylation, biological role. Biogenic amines: synthesis, their functions. Oxidation of biogenic amines.

2. Ways for the formation and detoxification of ammonia.

3. Intracellular detoxification of ammonia: reductive amination, synthesis of glutamine and asparagine. Role of glutaminase in the maintenance of acid-base balance in the body.

4. Biosynthesis of urea. Disorders of the urea synthesis and excretion.

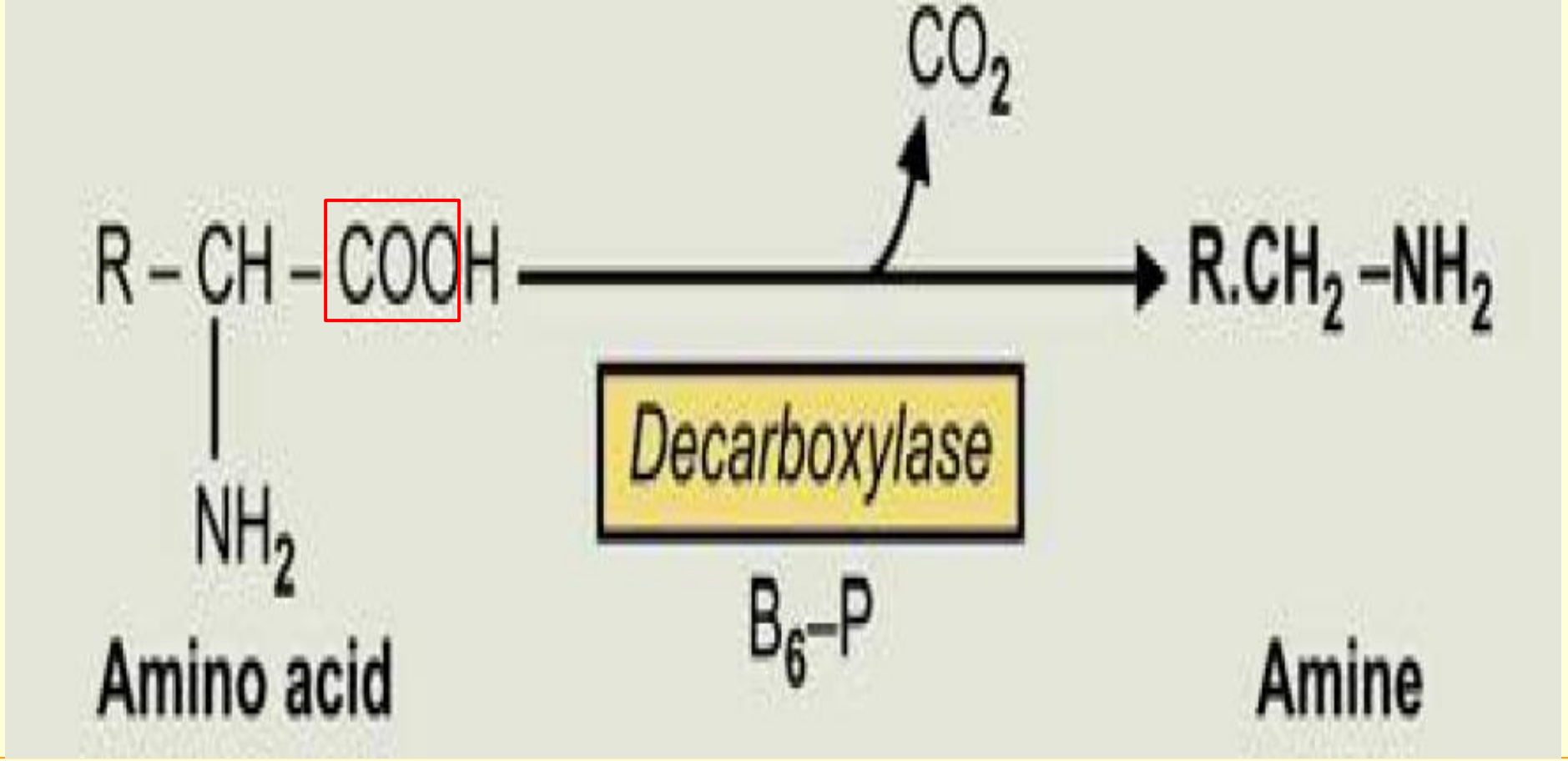
DECARBOXYLATION

- is a chemical reaction that removes a carboxyl group and releases carbon dioxide (CO_2). Usually, decarboxylation refers to a reaction of carboxylic acids, removing a carbon atom from a carbon chain.

- Enzymes that catalyze decarboxylation are called DECARBOXYLASES or, the more formal term, carboxy-lyases (EC number 4.1.1).

- Decarboxylases require prostetic group PYRIDOXYL-PHOSPHATE (PLP).

Decarboxylation of AA



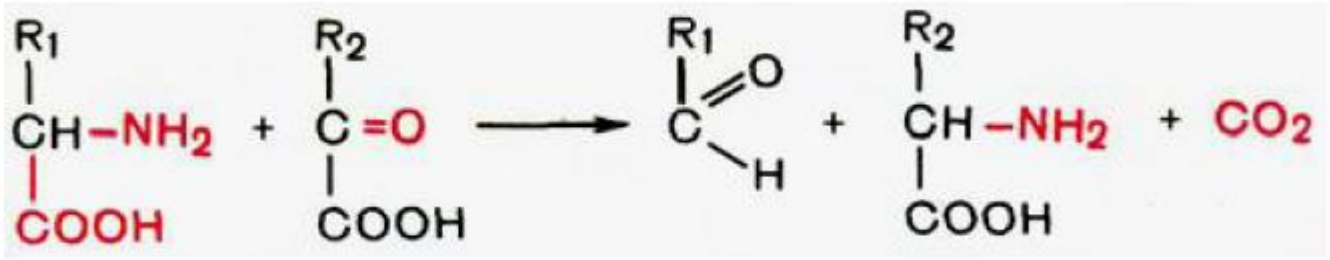
4 types of decarboxylation of amino acids

1) α-Decarboxylation (is typical of animal tissues)

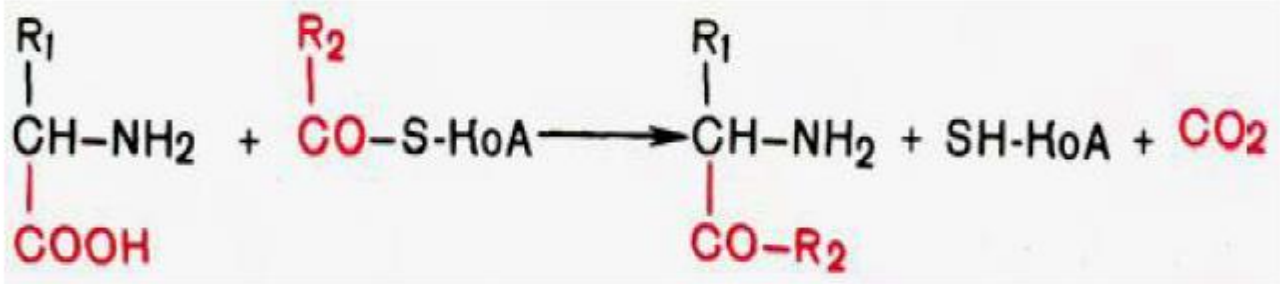
2) ω-Decarboxylation (This type of decarboxylation is typical of microorganism)

4 types of decarboxylation of amino acids

3) Decarboxylation involving a transamination reaction.



4)Decarboxylation involving condensation reaction of two molecules.



The most common amines, formed from amino acids and derivatives

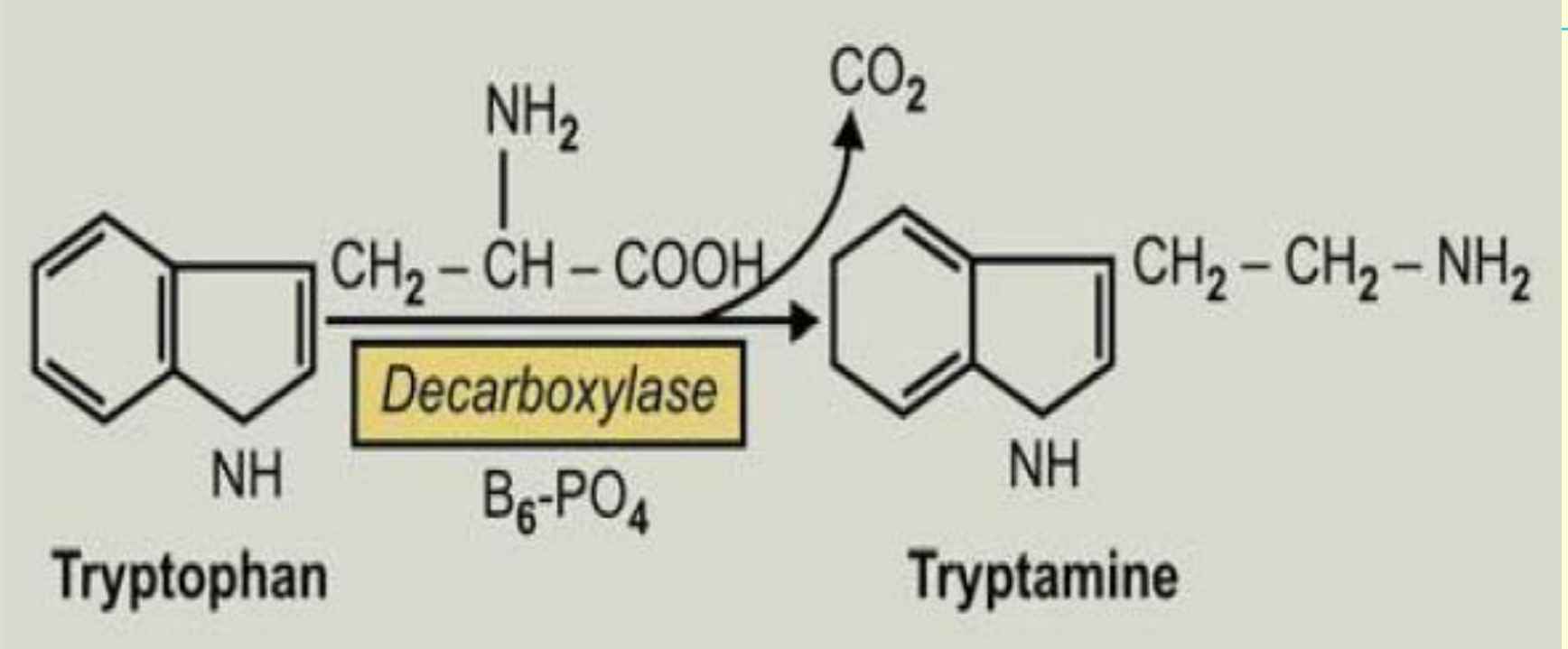
- tryptophan to tryptamine
- phenylalanine to phenylethylamine
- tyrosine to tyramine
- histidine to histamine
- glutamic acid to GABA
- lysine to cadaverine
- ornithine to putrescine
- 5-hydroxytryptophan to serotonin
- L-DOPA to dopamine

Tryptamine

- is found in plants, fundi, animals, and in trace amounts in brains of mammals

- is believed to play a role as a **neuromodulator** *(regulator of metabolism in nervous cells)* or **neurotransmitter** *(a chemical released by neurons to send signals to other neurons).*

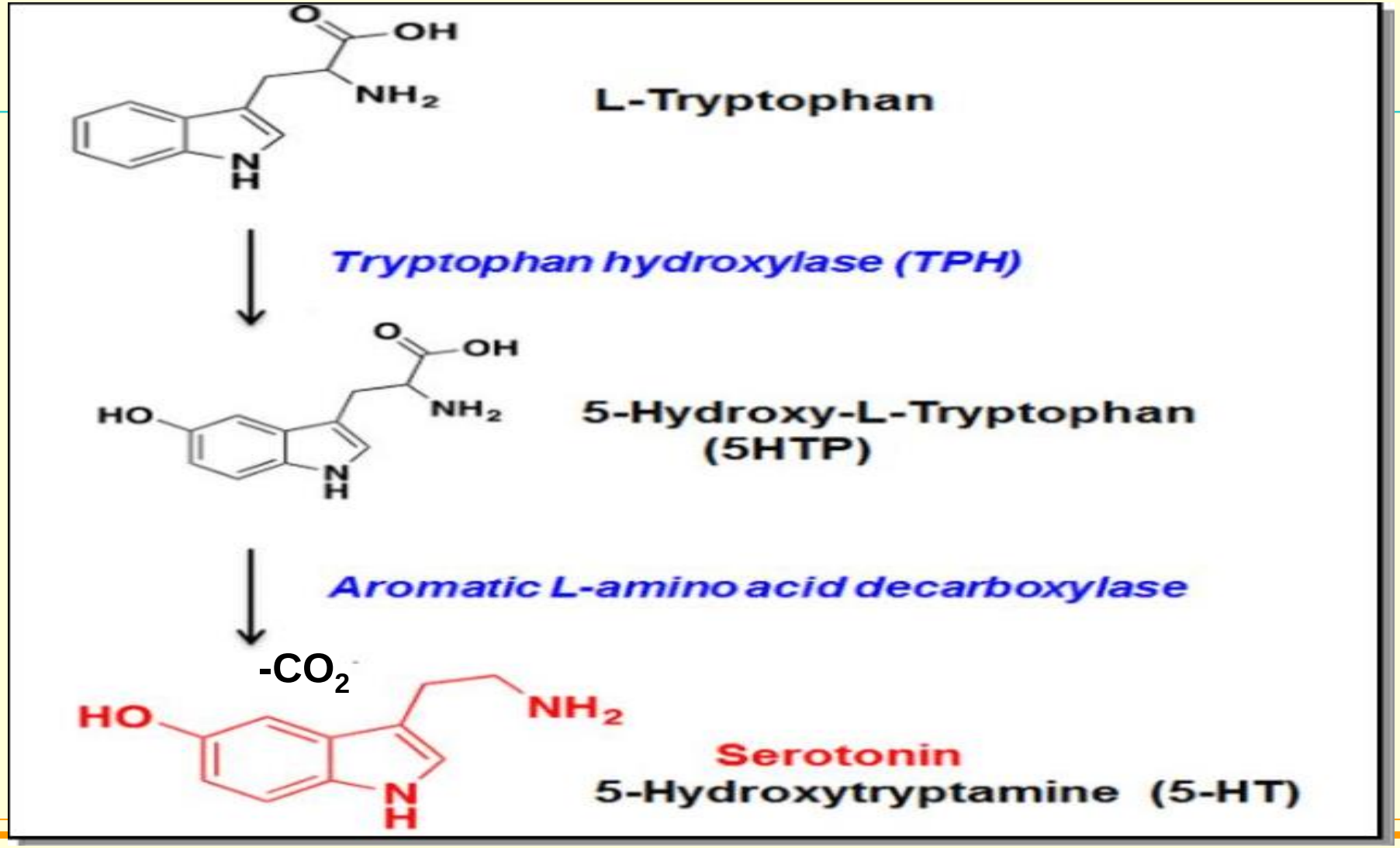
Synthesis of tryptamine from L-tryptophan



Serotonin (5-hydroxytryptamine, 5-HT)

- is primarily found in the central nervous system (CNS).
- Outside the CNS is found in the enteric nervous system located in the GIT.
- Additionally, serotonin is stored in blood platelets.

Synthesis of serotonin from L-tryptophan



Biological functions of serotonin

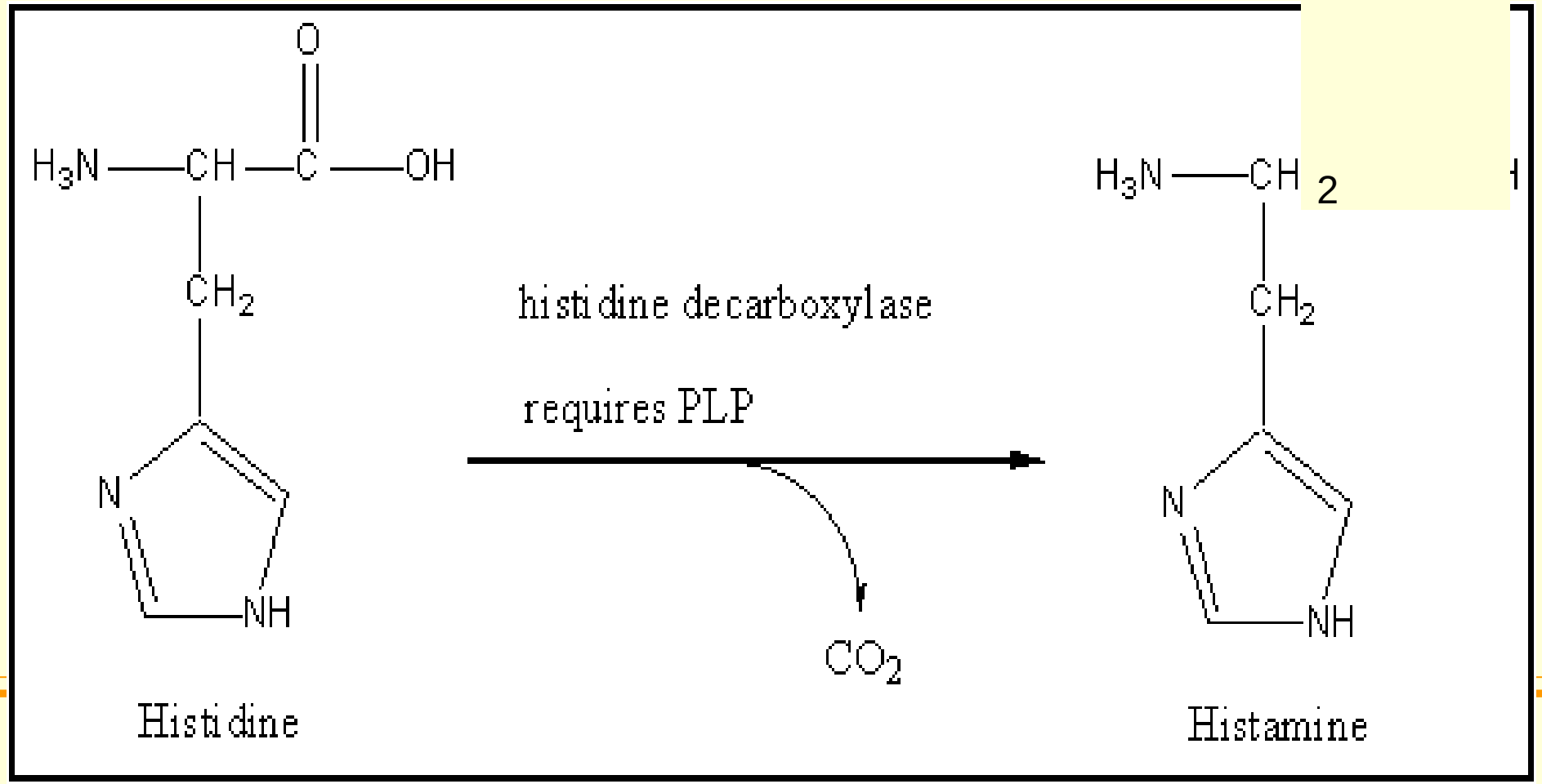
- Monoamine neurotransmitter in the CNS.
- Modulation of mood, appetite, sleep.
- Role in cognition, reward, learning, memory.
- Regulation of vomiting (in the GIT).
- Role in vasoconstriction of blood vessels (in platelets).
- Several classes of drugs target the 5-HT system, including some antidepressants, antipsychotics, anxiolytics, and antimigraine drugs, as well as, the psychedelic drugs.

Histamine

- is derived from the decarboxylation of the histidine, a reaction catalyzed by the enzyme L-histidine decarboxylase.

- Places of synthesis: mast cells, basophils, histaminergic neurons in the CNS and enterochromaffin-like cells (ECL) in the stomach.

Synthesis of histamine from Histidine



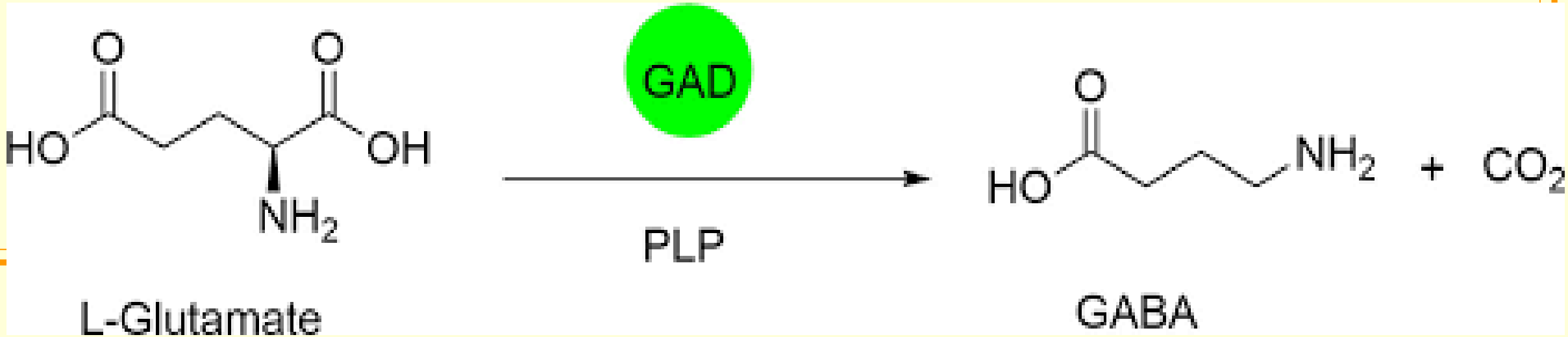
Histamine function

- CNS: Regulates Sleep-wake cycle (promotes wakefulness), body temperature, nociception, endocrine homeostasis, appetite, is involved in cognition

- Outside CNS: Causes bronchoconstriction (bronchial smooth muscle contraction), urinary bladder contractions, vasodilation, promotes hypernociception (visceral hypersensitivity), is involved in itch perception and urticaria.

Gamma-aminobutyric acid (GABA)

- Non-proteinogenic amino acid produced by glutamate decarboxylase (GAD) from L-glutamate.
- Present in high concentration in the brain.
- Most abundant inhibitory neurotransmitter widely distributed throughout the brain.
- Less distributed outside the CNS.

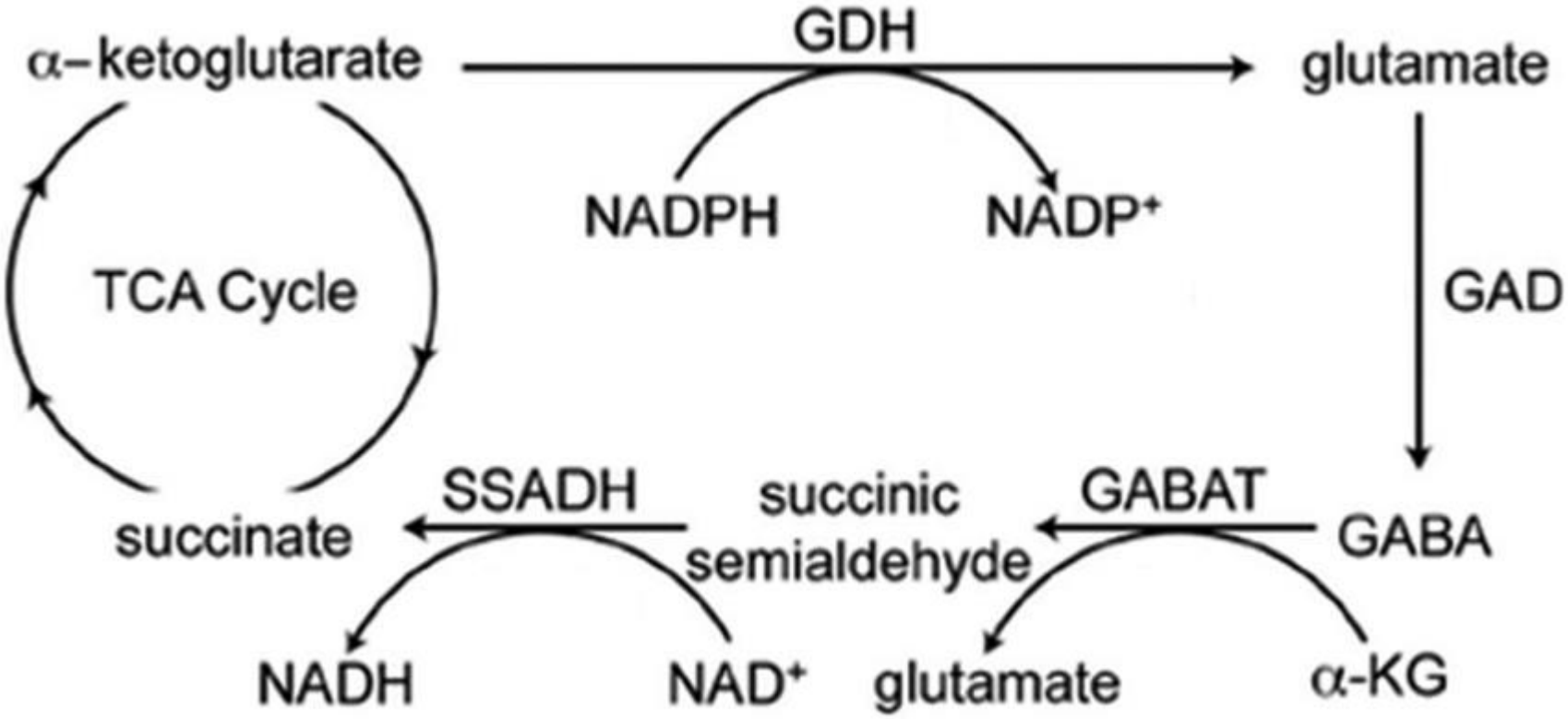


GABA function

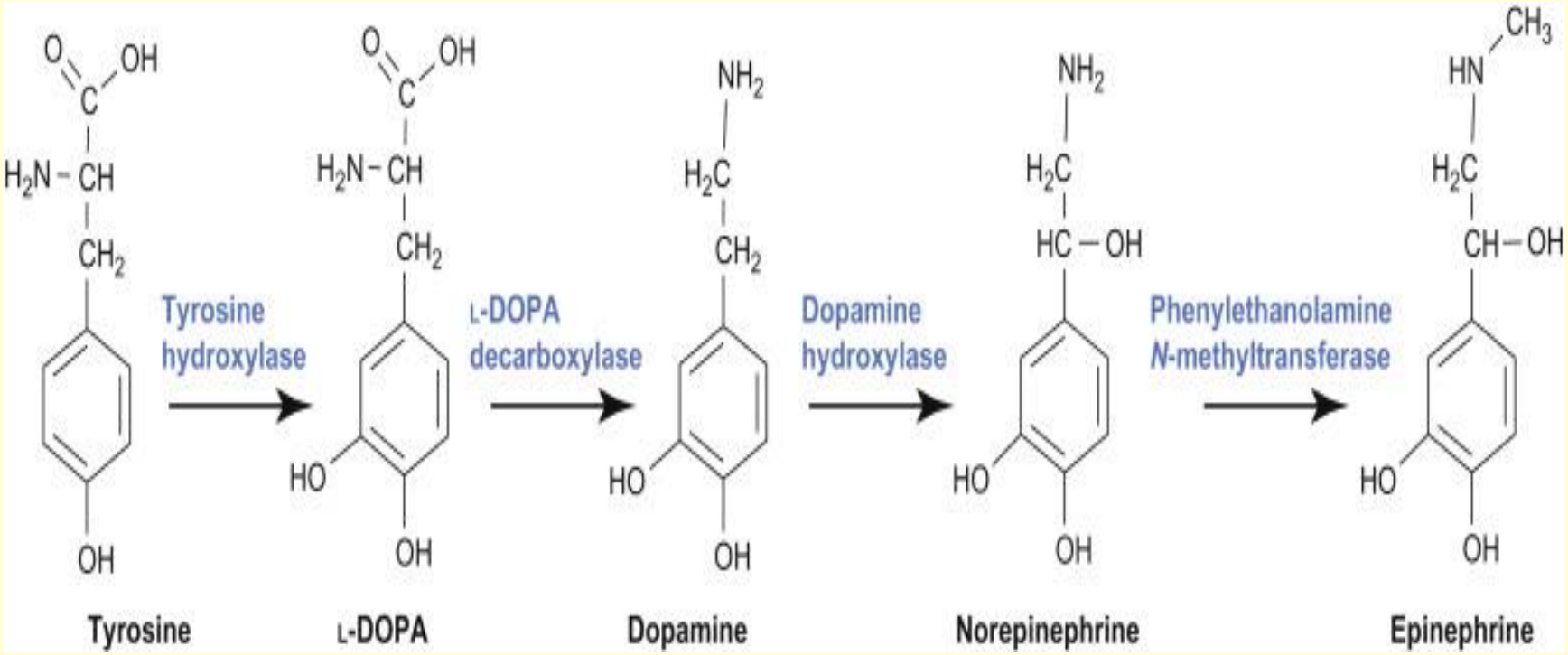
- Reduces activity of excitatory neurons.
- Produce a calming effect, control the fear or anxiety experienced when neurons are overexcited.
- Improving mood.
- Role in onset of epilepsy, seizures, alcohol and drug addiction.
- Relieving premenstrual syndrome.
- Stabilize blood pressure.
- Regulating the release of sex hormones.

GABA metabolism (or GABA shunt)…

- Regulates production of energy in neural cells.
- In the CNS GABA shunt acts as an additional source of intermediate substrate succinate to the tricarboxylic acid cycle (TCA cycle).



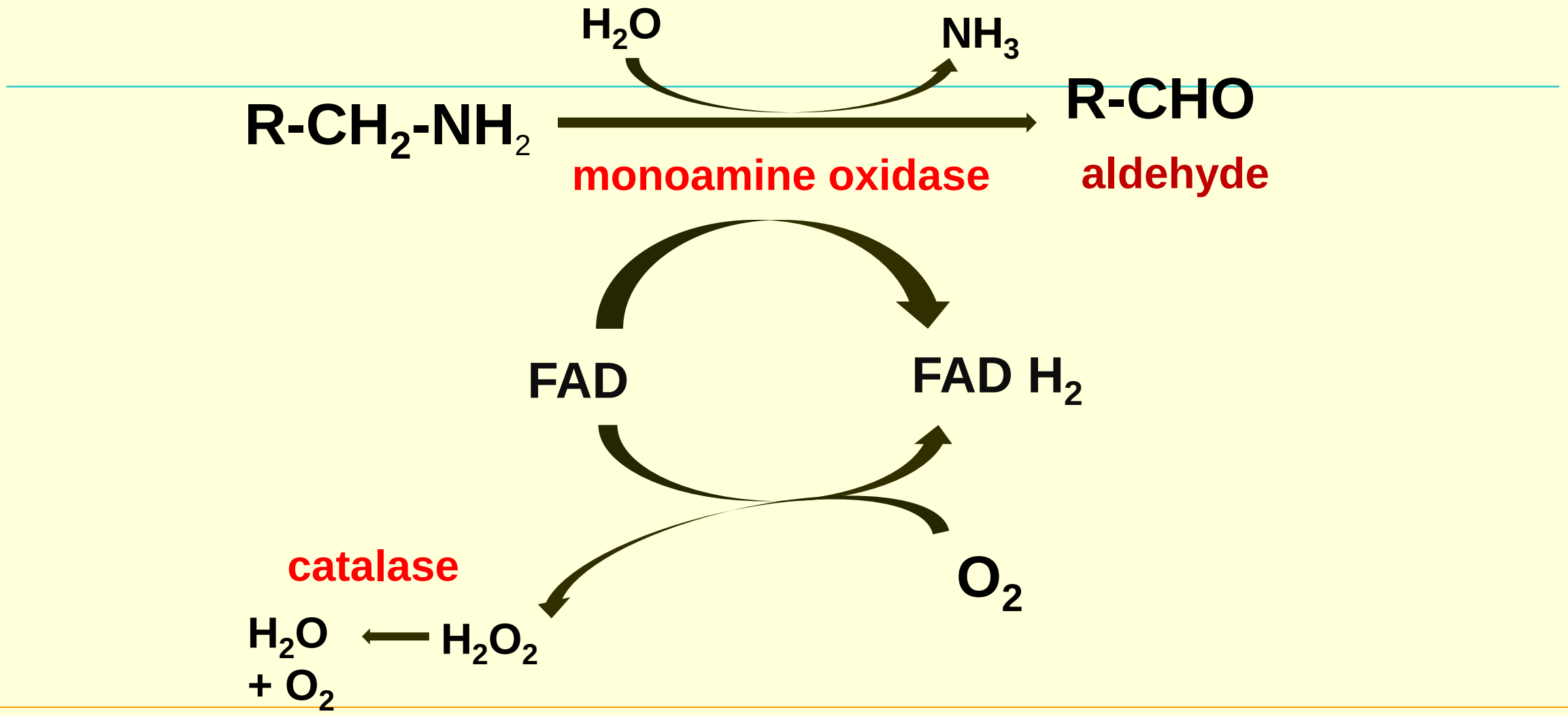
Catecholamines (dopamine, norepinephrine, epinephrine)



Dopamine, norepinephrine, and epinephrine

- **DOPAMINE Is produced from DOPA (dihydroxy-phenylalanine) by L-DOPA decarboxylase. It is an abundant excitatory neurotransmitter in the CNC.**
- **DOPAMINE plays the role in motor function, reward-motivated behavior, including development of addictive types of behavior.**
- **Outside the CNS DOPAMINE acts as the precursor for NOREPINEPHRINE and EPINEPHRINE.**
- **Noreninephrine (noradrenaline) and epinephrine (adrenaline) are hormones and synthesized in the sympathetic nervous tissue and adrenal glands.**

Degradation of biogenic amines

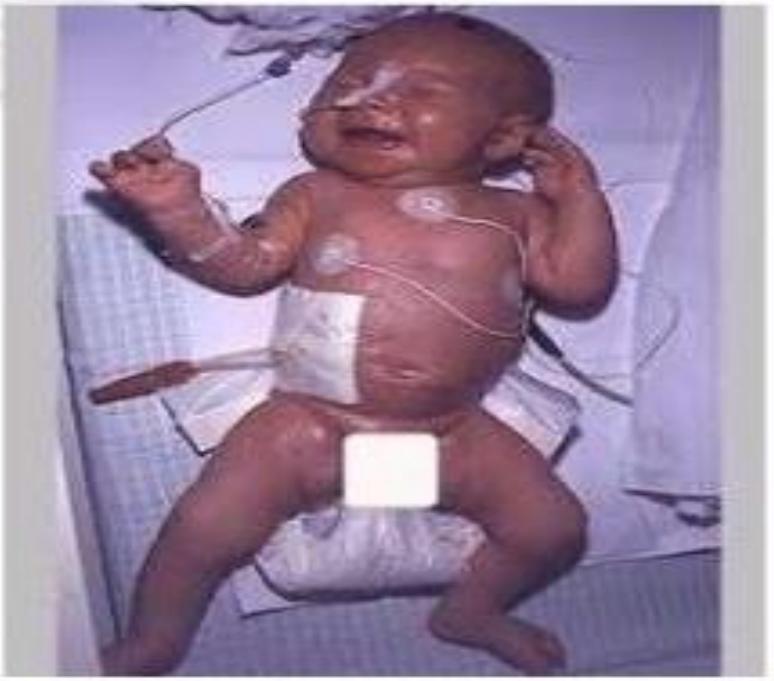


Formation and detoxification of ammonia

- Ammonia (NH_3) or ammonium ion (NH_4^+) is produced in various reactions.
- NH_3 is very toxic in humans and must be converted to non-toxic compounds.
- Normal level of NH_3 in the blood must not exceed ~60 mkmol/l
- Hyperammoniemia is a dangerous condition that may lead to brain injury and death.

Signs and symptoms

- Signs and symptoms of early-onset hyperammonemia (neonates) may include the following:
- Lethargy
- Irritability
- Poor feeding
- Vomiting
- Seizures
- Hyperventilation, grunting respiration



Symptoms of

Hyperammonemia

General
- Growth retardation
- Hypothermia

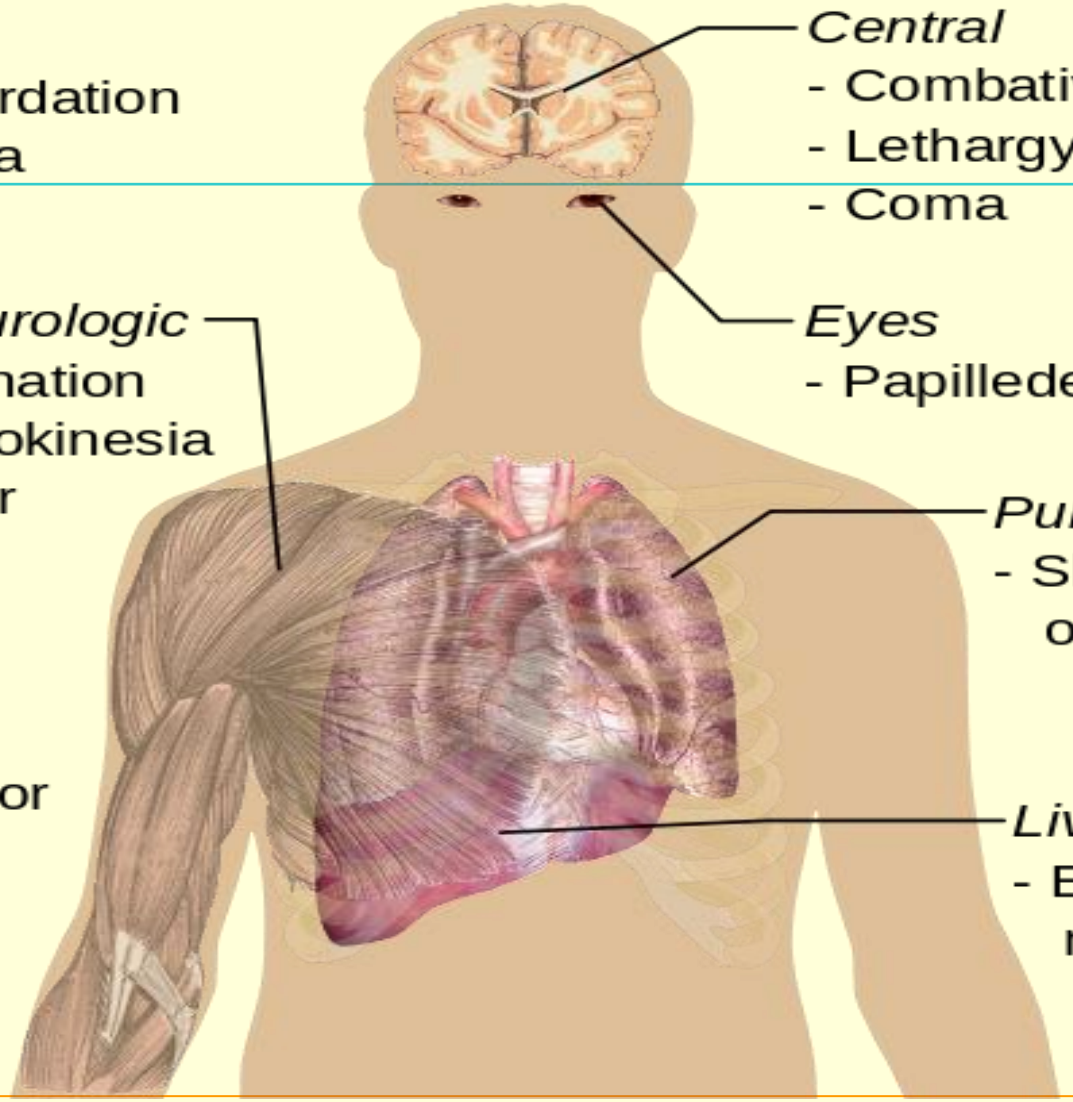
Central
- Combativeness
- Lethargy
- Coma

Eyes
- Papilledema

Muscular/Neurologic
- Poor coordination
- Dysdiadochokinesia
- Hypotonia or hypertonia
- Ataxia
- Tremor
- Seizures
- Decorticate or decerebrate posturing

Pulmonary
- Shortness of breath

Liver
- Enlarge-ment



Ammonia is produced in….

- Oxidative deamination of Glutamate.
- Transdeamination.
- Deamination of Glutamine and Asparagine to Glutamate and Aspartate.
- Deamination of purine and pyrimidine nucleotides.
- Degradation of amines.

Ammonia is also produced in….

the gut by intestinal bacterial flora, both

- From dietary proteins,
- From urea present in fluids secreted into the GIT.

Detoxification of ammonia

- Ammonia produced in the liver is detoxified in the Urea cycle (~60% of all ammonia in the body).
- Ammonia formed in the brain is used for synthesis of glutamine and asparagine from glutamate and aspartate.
- In the kidney ammonia is excreted in form of ammonium salts.

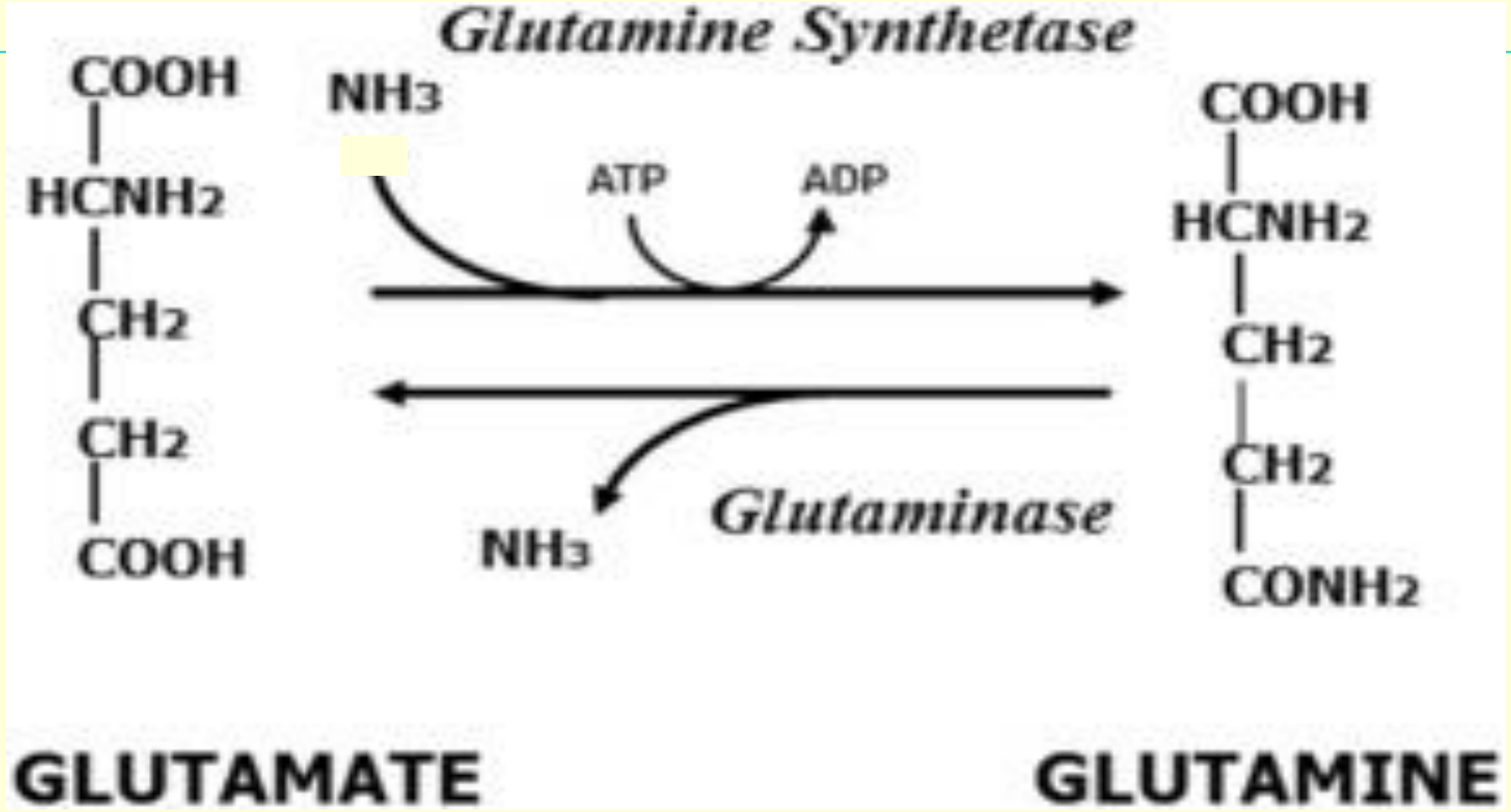
Another reactions of detoxification are...

- Reductive reamination of a-ketoglutarate to glutamate.

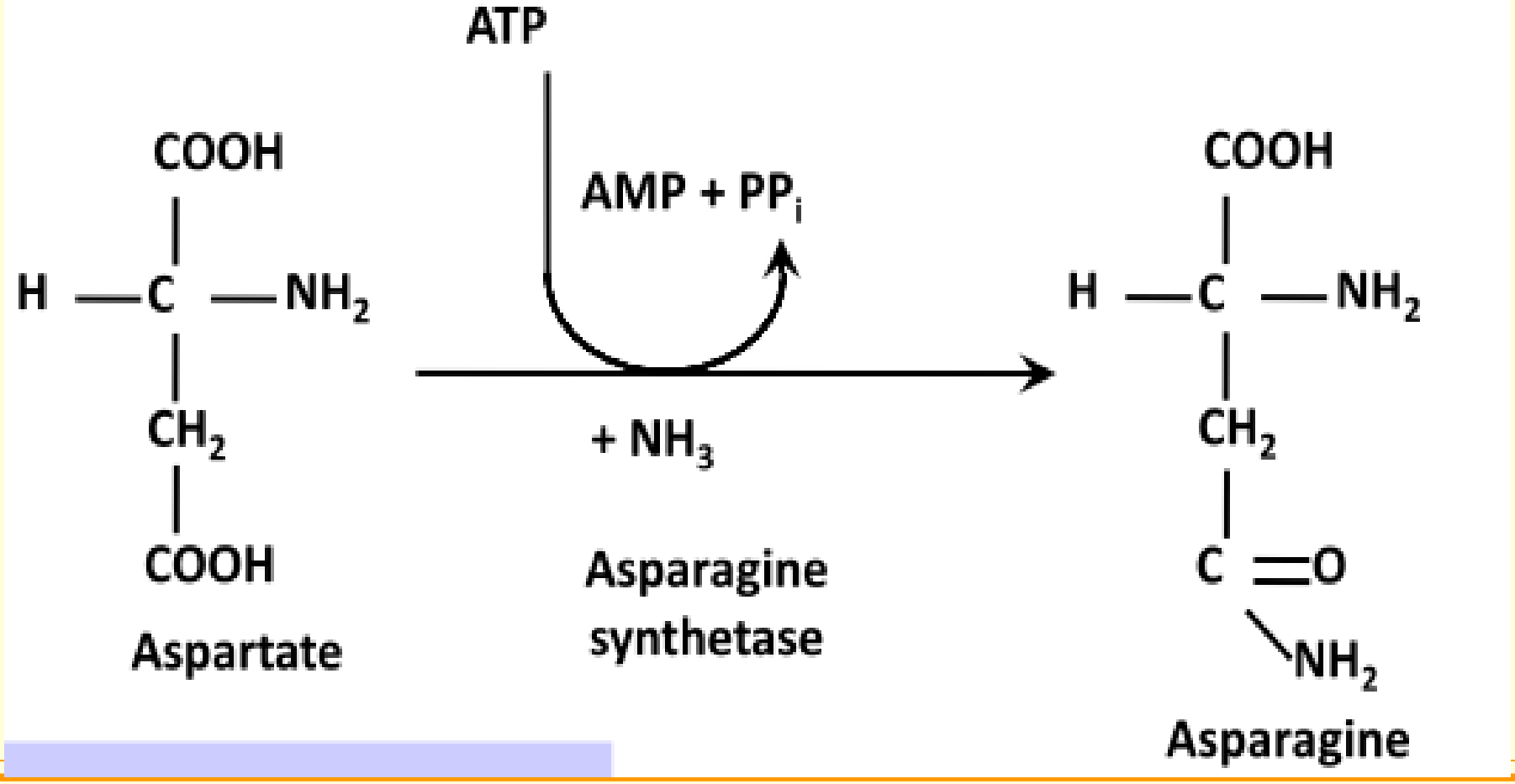
- Transreamination.

(see the previous lecture)

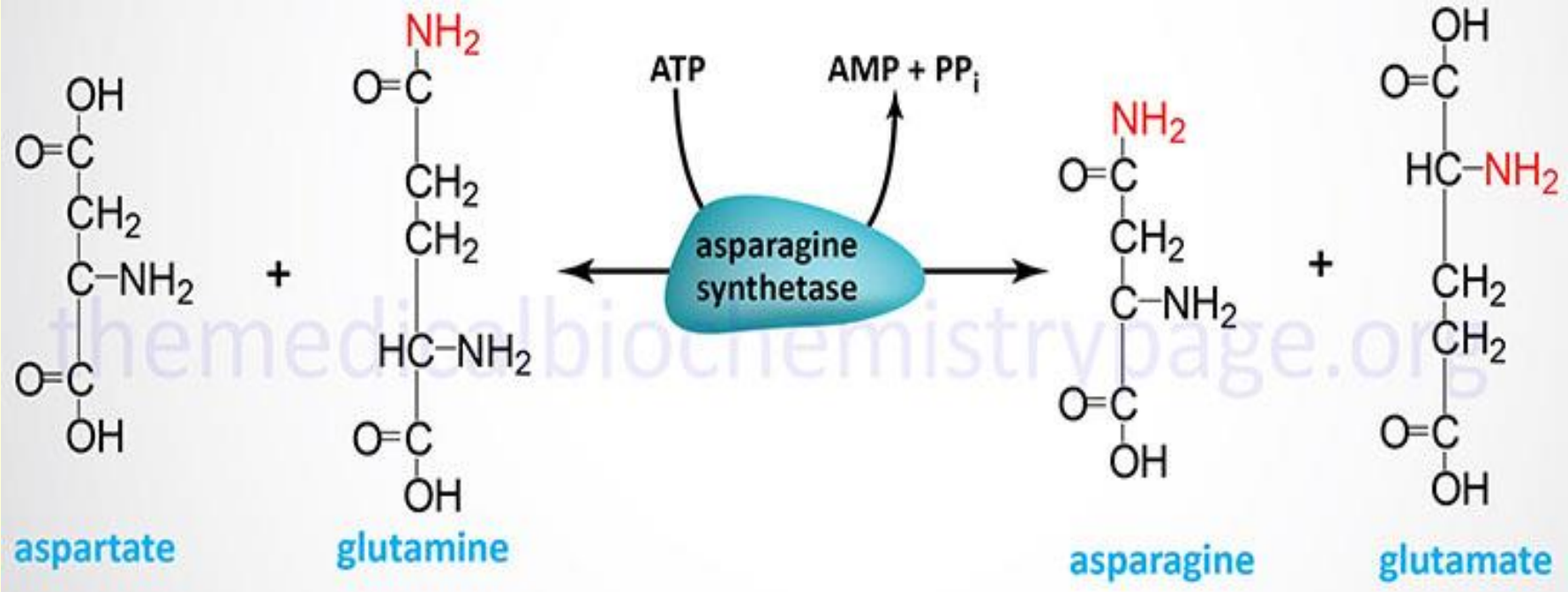
Synthesis and deamination of glutamine



Synthesis of asparagine from aspartate and ammonia

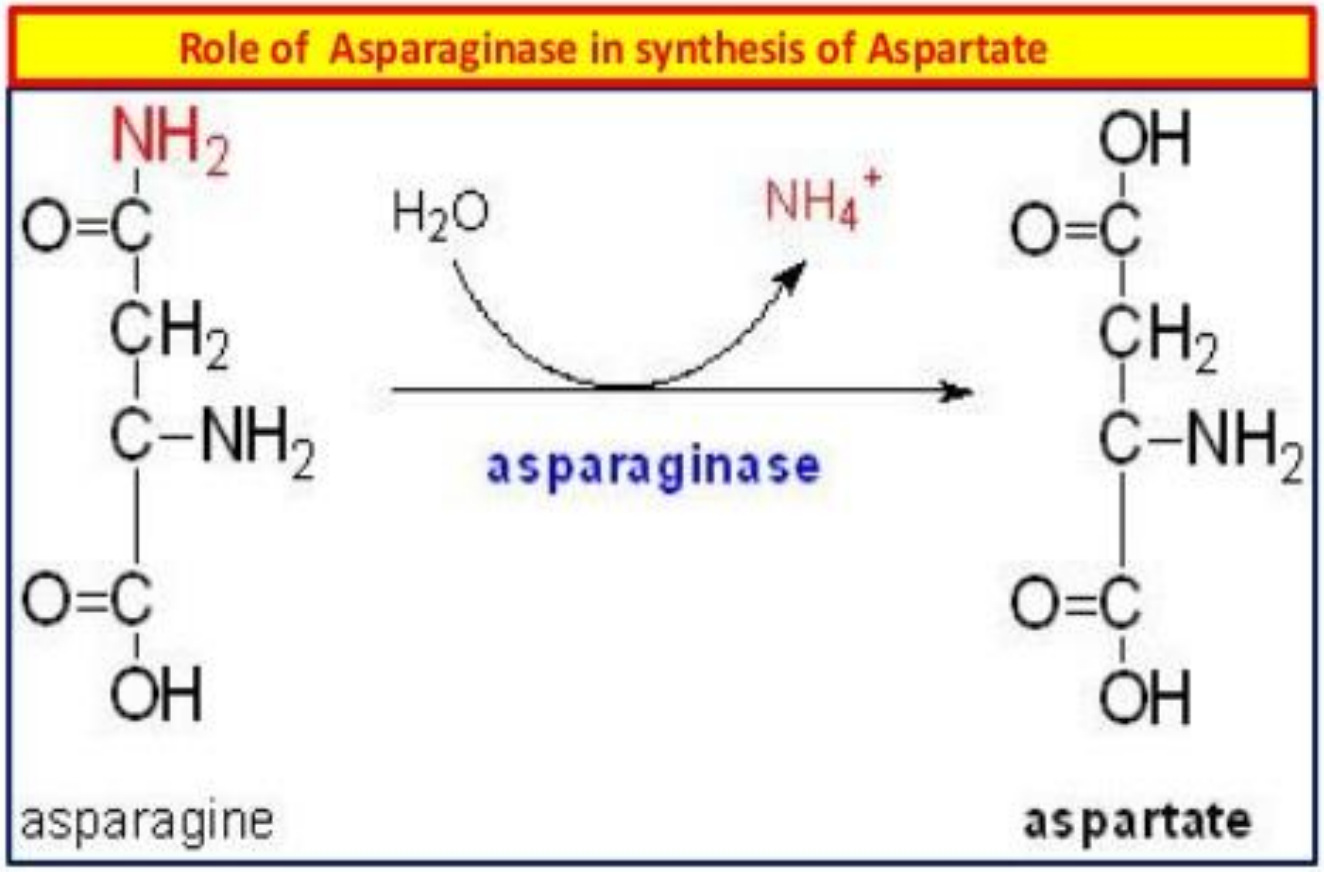


Synthesis of asparagine from aspartate and glutamine



Glutamine and asparagine are the major transport forms of ammonia from brain to the liver.

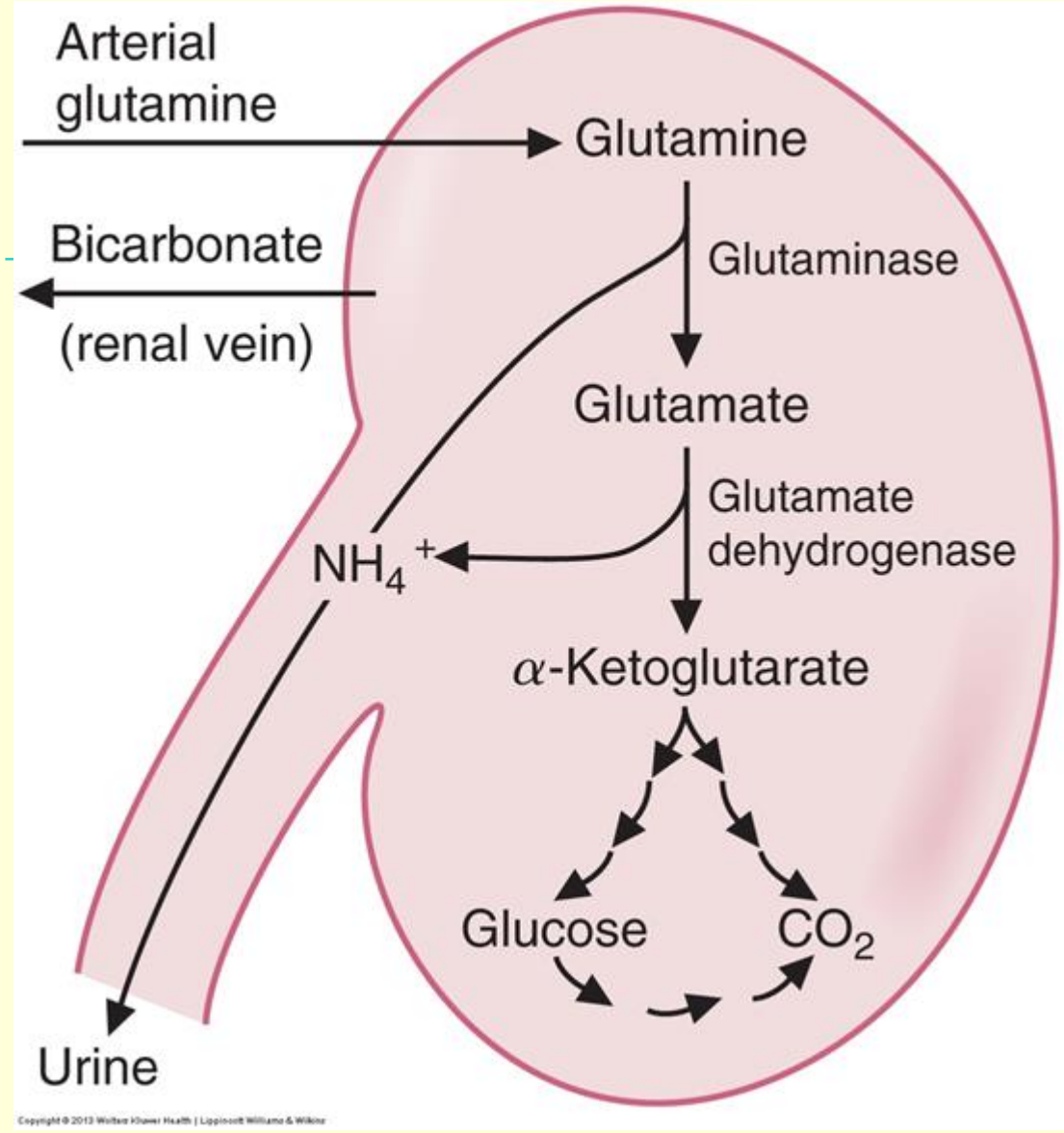
Deamination of asparagine



Role of glutaminase in the maintenance of acid-base balance in the body.

- **ACIDOSIS is a process causing increased acidity in the blood and other body tissues.**

- **One of the most powerful mechanisms of controlling the blood acid-base balance is removal of excess H^+ from the blood with ammonium salts in the urine.**



1. In acidosis epithelial cells of the renal tubules increase uptake of glutamine and H+ from the blood.

2. Acidosis stimulates activity of the kidney glutaminase and glutamate dehydrogenase, the enzymes producing NH_3 from glutamine and glutamate.

3. Non-ionized NH_3 can easily diffuse through cell membrane into the tubular lumen. In turn H^+ diffuse into the lumen in exchange for Na^+.

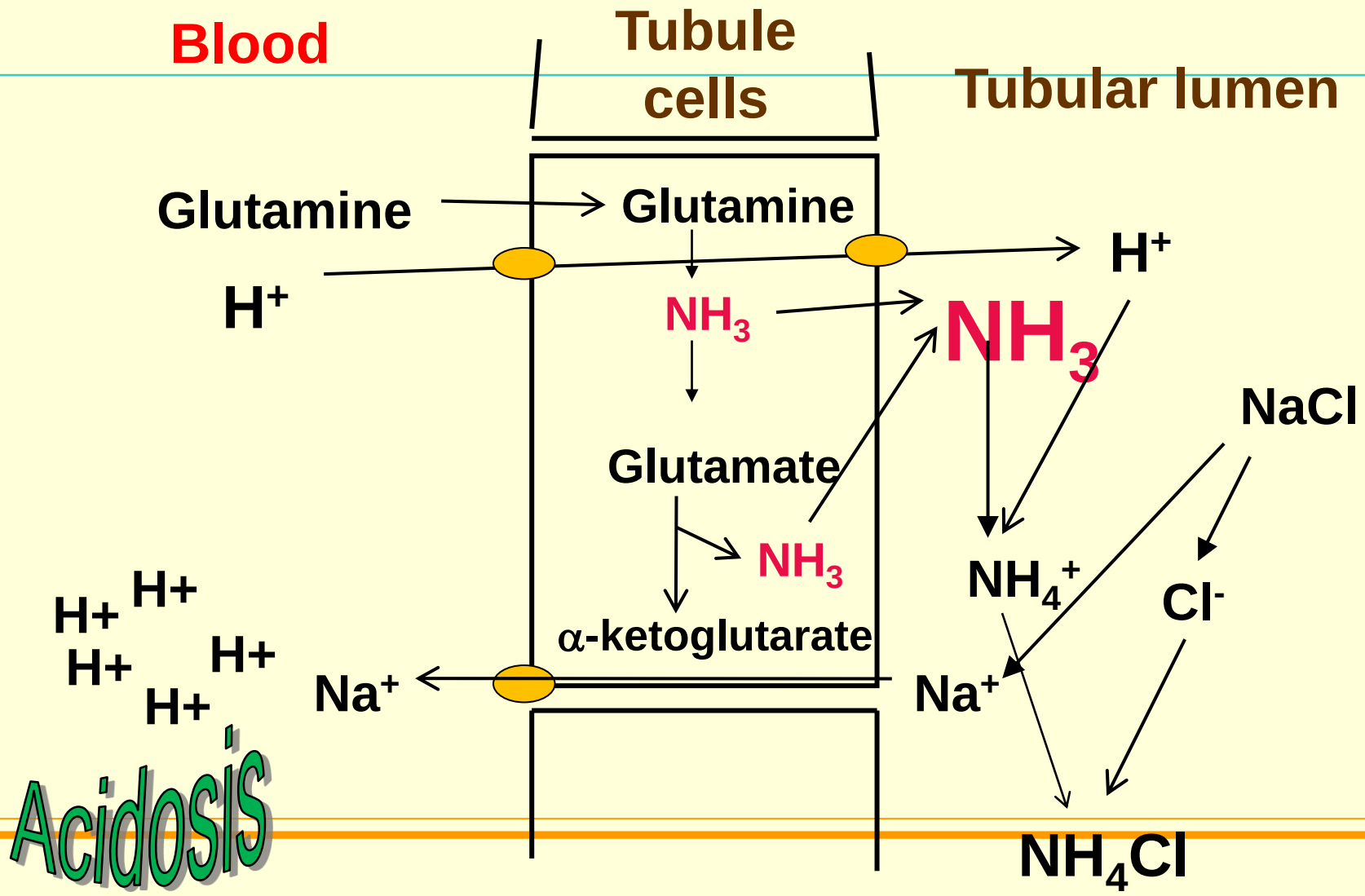
In the tubular lumen

$$NH_3 + H^+ = NH_4^+$$

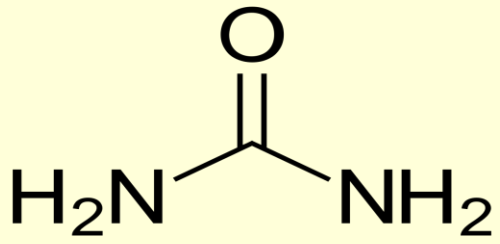
$$NH_4^+ + Cl^- = NH_4Cl$$

4. Resulted AMMONIUM CHLORIDE is excreted through urine from the body.

Role of glutaminase in the maintenance of acid-base balance in the body.



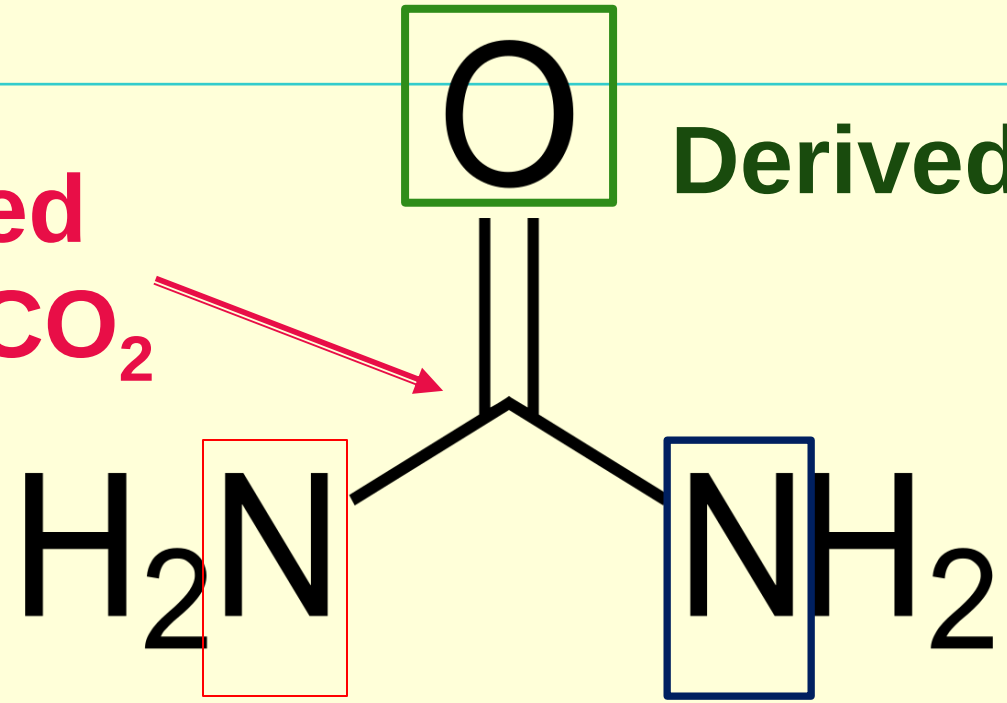
Urea cycle
(or the ornithine cycle)



- The urea cycle is a cycle of **biochemical** reactions that produces **UREA** from highly toxic NH_3 for excretion.
- The organisms producing urea from ammonia are called **UREOTELIC.**
- This cycle was the first metabolic cycle to be discovered by **Hans Krebs** and **Kurt Henseleit** in 1932.
- The urea cycle takes place primarily in **THE LIVER** and, to a lesser extent, in the **kidneys**.

- In contrast to ammonia, urea is neutral and therefore relatively non-toxic.
- As a small, uncharged molecule, urea is able to cross biological membranes easily.
- In addition, it is easily transported in the blood and excreted in the urine.

Origin of atoms in urea



Derived from H_2O

Derived from CO_2

Derived from NH_2- of Asp

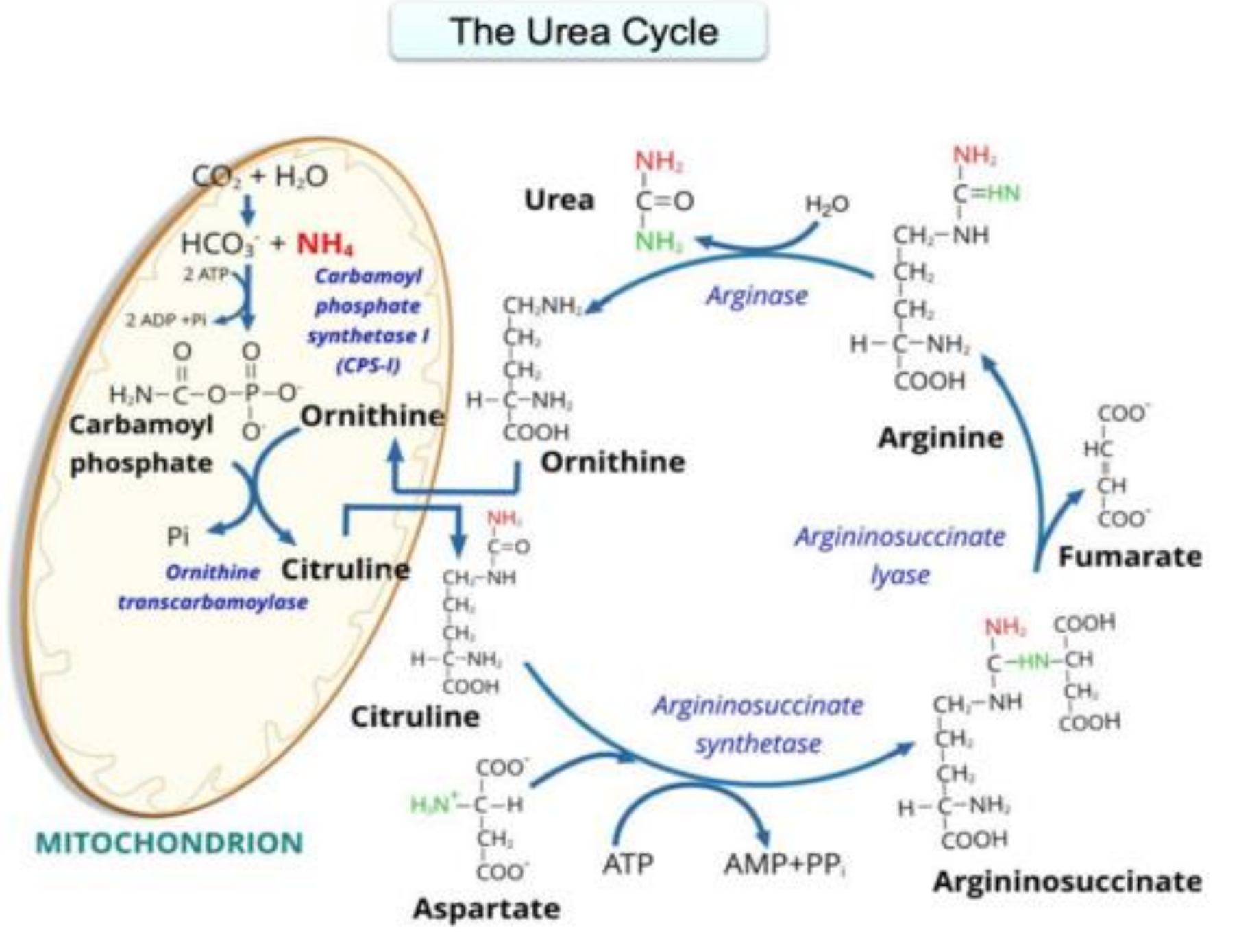
Derived from ammonia

Urea cycle

- Consists of 5 reactions:

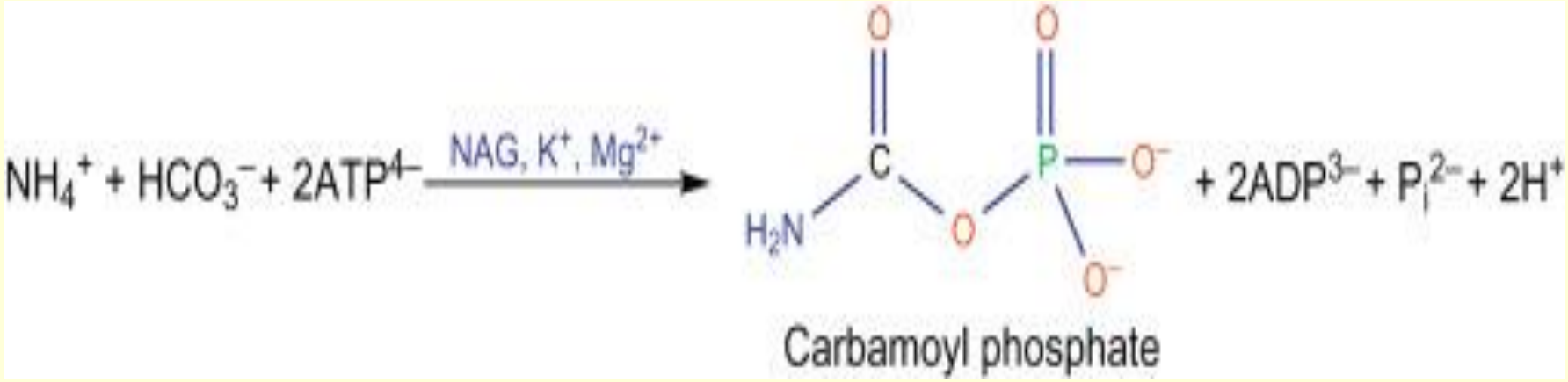
2 in the mitochondria and 3 in cytosol

The Urea Cycle

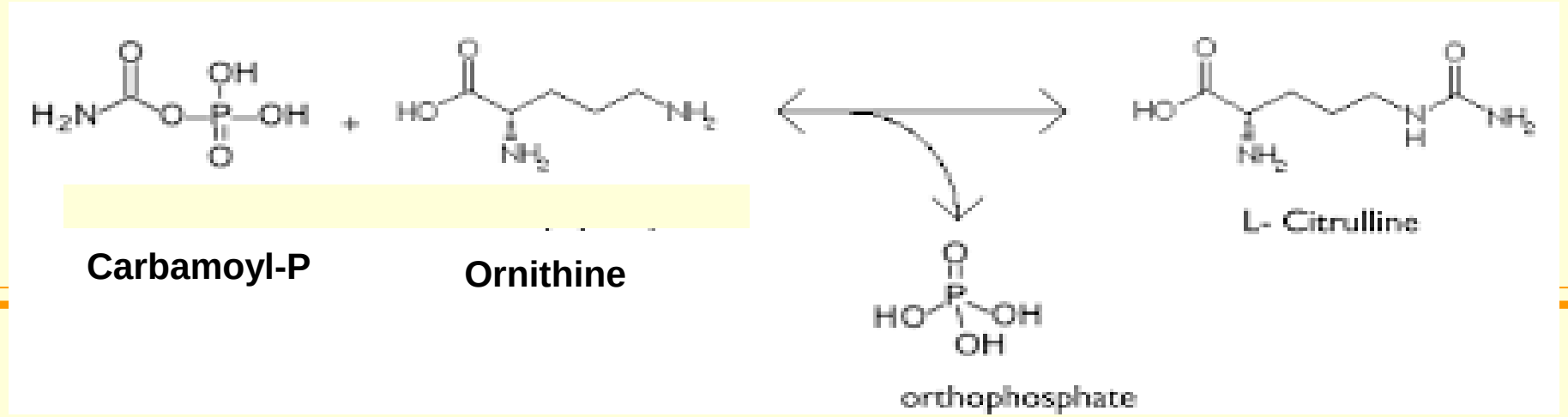


Mitochondrial reactions

Step 1. Carbamoyl-phosphate synthetase-1

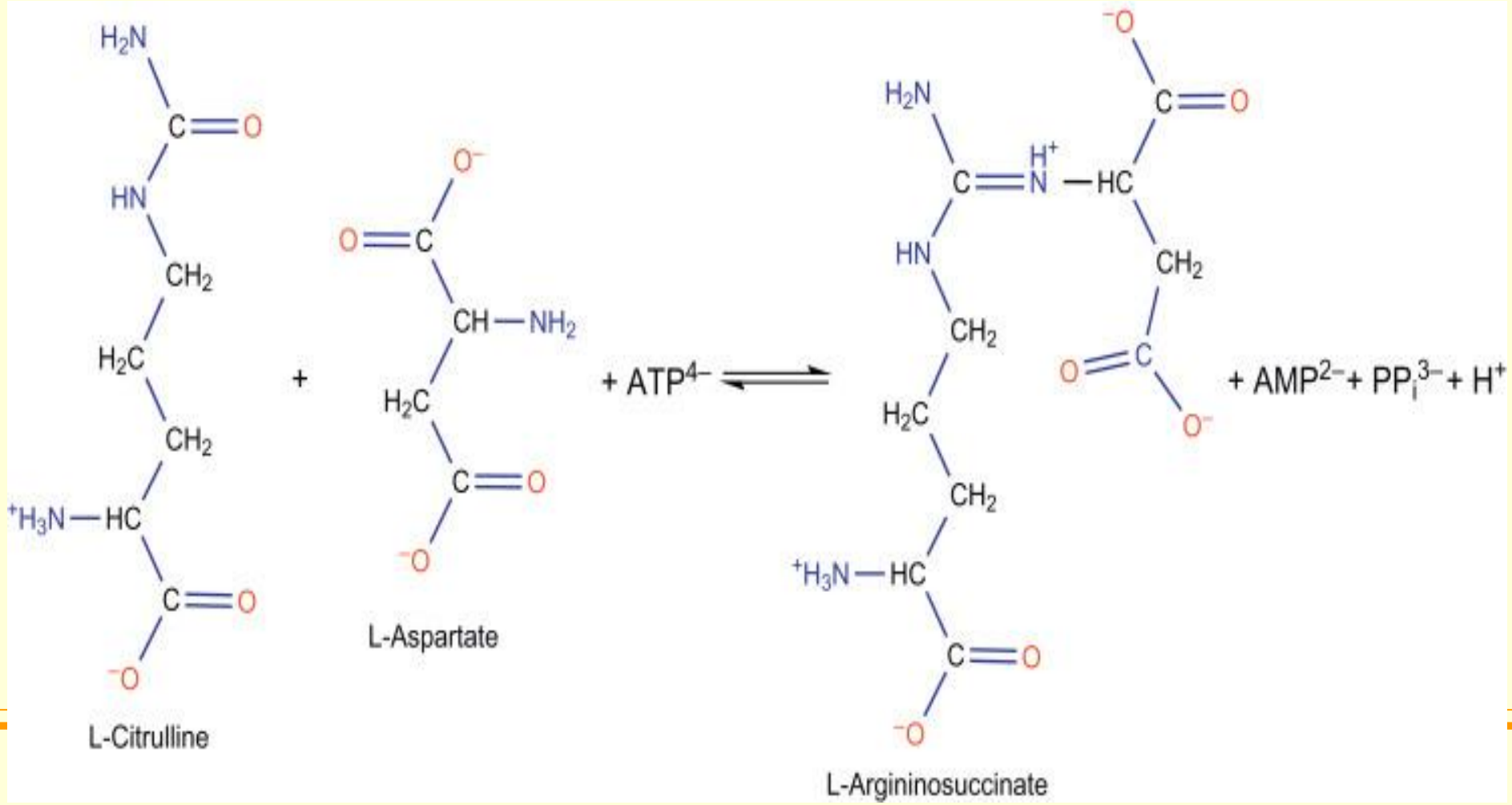


Step 2. Ornithine transcarbamylase

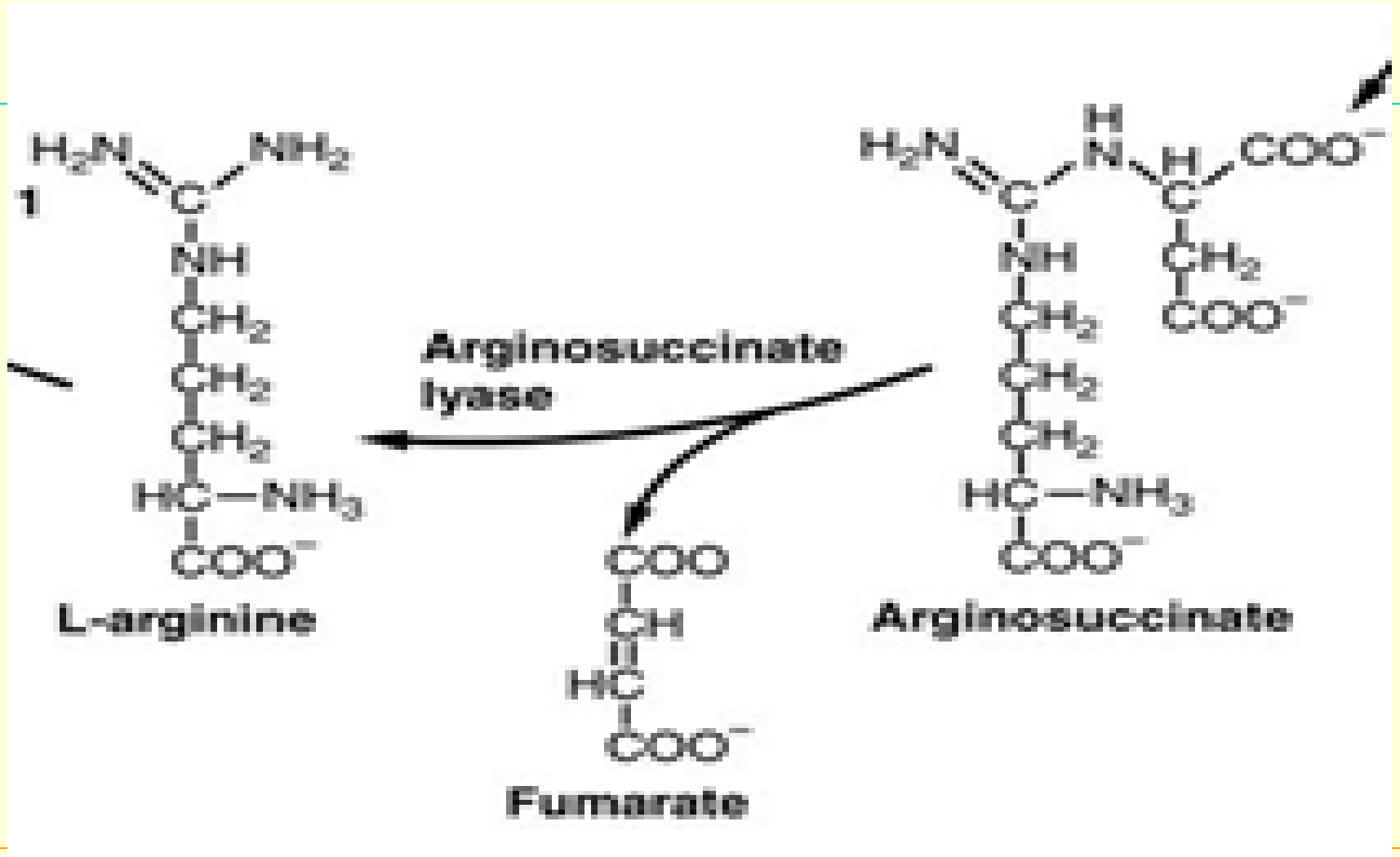


Reactions in cytosol

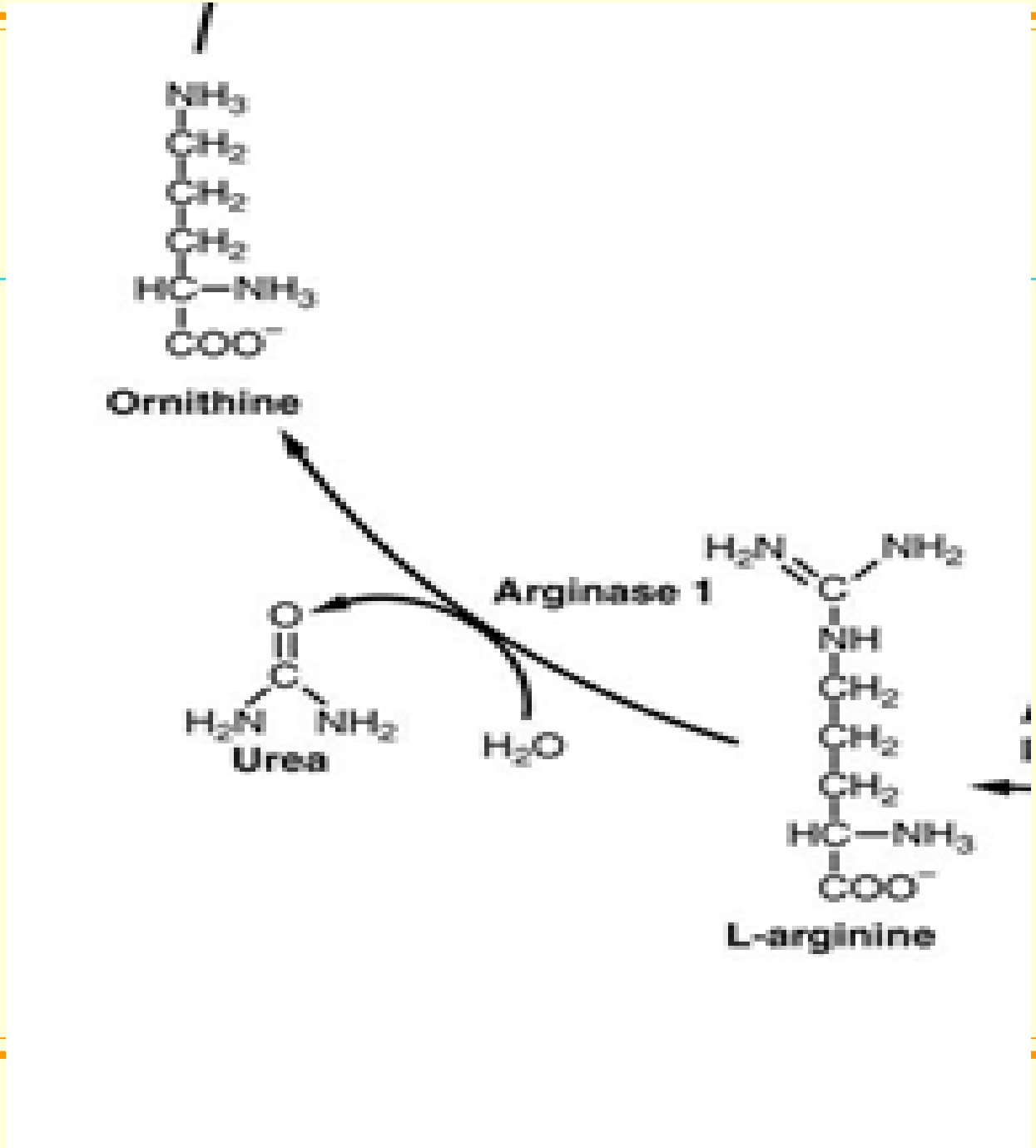
Step 3. Arginino-succinate synthase



Step 4. Argininosuccinate lyase



Step 5.
Arginase



Disorders of the urea synthesis and excretion.

Blood Urea Nitrogen (BUN) and urea in the unine

- Normal urea levels:

  Plasma: 2,5 – 8,33 mmol/l

  Urine: 333 – 583 mmol/day

- Serum/plasma urea concentration reflects the balance between urea production in the liver and urea elimination by the kidneys in urine.

- The BUN test is conducted to determine how much urea nitrogen is still present in the blood, and if the kidneys renal functions are working as they should!

Causes of High Blood Urea Nitrogen

- High protein diet.
- Heart failure and dehydration.
- Kidney stones and tumors that block the urinary tract.
- Gastrointestinal hemmorhage,
- Fever,
- Increased degradation of proteins in wasting diseases

Causes of Low Blood Urea Nitrogen

- Decreases in blood urea levels are rare.
- Present in liver damage in hepatitis, or cirrhosis,
- In genetic disorders of urea cycle.
- Blood urea may be seen to be lower in pregnancy than in normal nonpregnant women.

Urea Cycle Disorders

- Since the urea cycle is the only pathway by which we rid the body from ammonia any inborn defects in any of enzymes of the urea cycle will lead to hyperammonemia or the buildup of a cycle intermediate.

- Most urea cycle disorders are associated with elevated NH_3 and decreased Urea in the blood.

Deficient enzyme	Disorder	Clinical feature
N-acetylglutamate synthase	Hyperammonemia high plasma glutamate, alanine	Lethargy, vomiting, enlarged liver, seizures
Carbomoyl phosphate synthase	Hyperammonemia citrullinemia	Lethargy, coma, vomiting, enlarged liver, poor feeding, seizures
Ornithine transcarbamoylase	Hyperammonemia orotic acid in the urine	Seizures, vomiting, enlarged liver
Argininosuccunate synthase	Citrullinemia	Lethargy, coma, seizures, vomiting

Deficient enzyme	Disorder	Clinical feature
Argininosuccunate lyase (argininosuccinic aciduria)	Hyperammonemia argininosuccinic acid in the urine	Lethargy, vomiting, enlarged liver, seizures
Arginase	Markedly elevated arginine in the blood, moderate hyperammonemia	Delayed development, protein intolerance, spasticity, loss of muscle control, seizures

The most common clinical manifestations in Urea Cycle Disorders

- Cerebral edema,
- lethargy,
- Anorexia,
- Hyper- or hypoventilation,
- Slurring of the speech,
- Blurring vision,
- Seizures,
- Neurological posturing, coma, etc.