

**Ministry of Public Health of Ukraine
Poltava State Medical University**

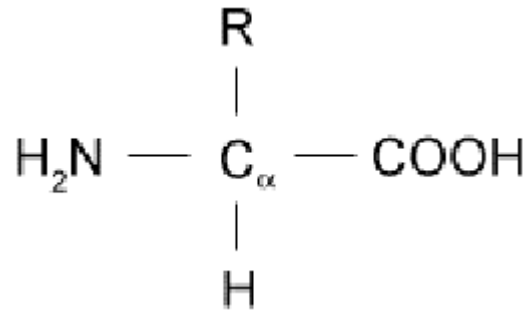
**Amino acid metabolism.
General pathways of transformation
of amino acids.
Ammonia metabolism: biosynthesis of urea
and its disorders.**

Assoc. Prof.
Bilets M.V.

Lecture plan

- The general pathways of amino acids metabolism:
 - Decarboxylation of amino acids.
 - Transamination of amino acids
 - Direct and indirect deamination of amino acids.
- Pathways of ammonia formation.
- The molecular mechanisms of ammonia toxicity.
- The molecular mechanisms of ammonia detoxification.

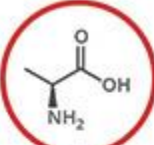
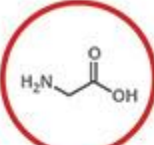
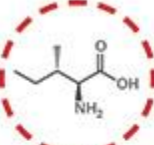
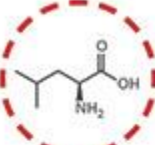
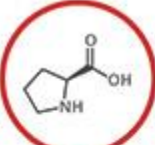
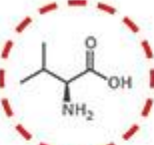
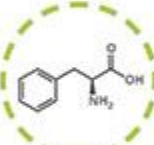
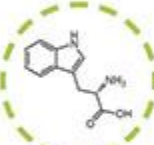
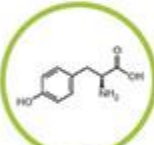
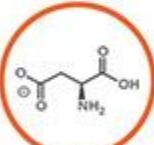
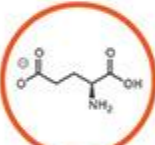
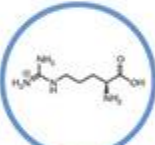
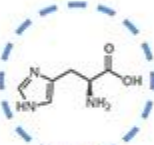
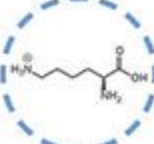
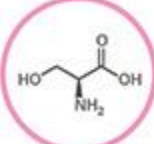
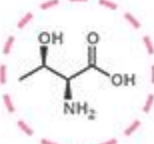
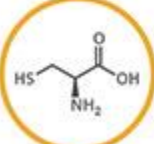
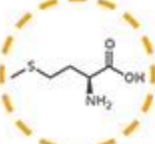
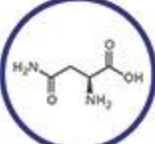
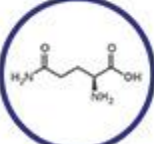
Amino acid, any of a group of organic molecules that consist of a basic amino group (—NH_2), an acidic carboxyl group (—COOH), and an organic R group (or side chain) that is unique to each amino acid. The term *amino acid* is short for α -amino [*alpha-amino*] carboxylic acid. Each molecule contains a central carbon (C) atom, called the α -carbon, to which both an amino and a carboxyl group are attached. The remaining two bonds of the α -carbon atom are generally satisfied by a hydrogen (H) atom and the R group. The formula of a general amino acid is:



- 1.The amino acids are used by various tissues to synthesize proteins and to produce nitrogen-containing compounds (e.g., purines, heme, creatine, epinephrine), or they are oxidized to produce energy.
- 2.The breakdown of both dietary and tissue proteins yields nitrogen-containing substrates and carbon skeletons.
- 3.The nitrogen-containing substrates are used in the biosynthesis of purines, pyrimidines, neurotransmitters, hormones, porphyrins, and nonessential amino acids.
- 4.The carbon skeletons are used as a fuel source in the citric acid cycle, used for gluconeogenesis, or used in fatty acid synthesis.

Amino acids classification

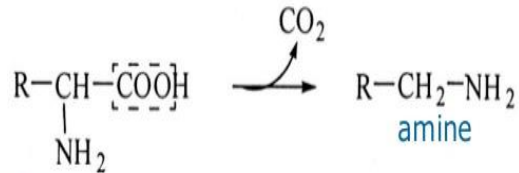
Chart Key: ● ALIPHATIC ● AROMATIC ● ACIDIC ● BASIC ● HYDROXYLIC ● SULFUR-CONTAINING ● AMIDIC ○ NON-ESSENTIAL ○ ESSENTIAL

<p><i>Chemical Structure</i> single letter code</p> <p>NAME A three letter code DNA codons</p>	 <p>ALANINE A <i>Ala</i> GCT, GCC, GCA, GCG</p>	 <p>GLYCINE G <i>Gly</i> GGT, GGC, GGA, GGG</p>	 <p>ISOLEUCINE I <i>Ile</i> ATT, ATC, ATA</p>	 <p>LEUCINE L <i>Leu</i> CTT, CTC, CTA, CTG, TTA, TTG</p>	 <p>PROLINE P <i>Pro</i> CCT, CCC, CCA, CCG</p>	 <p>VALINE V <i>Val</i> GTT, GTC, GTA, GTG</p>
 <p>PHENYLALANINE F <i>Phe</i> TTT, TTC</p>	 <p>TRYPTOPHAN W <i>Trp</i> TGG</p>	 <p>TYROSINE Y <i>Tyr</i> TAT, TAC</p>	 <p>ASPARTIC ACID D <i>Asp</i> GAT, GAC</p>	 <p>GLUTAMIC ACID E <i>Glu</i> GAA, GAG</p>	 <p>ARGININE R <i>Arg</i> CGT, CGC, CGA, CGG, AGA, AGG</p>	 <p>HISTIDINE H <i>His</i> CAT, CAC</p>
 <p>LYSINE K <i>Lys</i> AAA, AAG</p>	 <p>SERINE S <i>Ser</i> TCT, TCC, TCA, TCG, AGT, AGC</p>	 <p>THREONINE T <i>Thr</i> ACT, ACC, ACA, ACG</p>	 <p>CYSTEINE C <i>Cys</i> TGT, TGC</p>	 <p>METHIONINE M <i>Met</i> ATG</p>	 <p>ASPARAGINE N <i>Asn</i> AAT, AAC</p>	 <p>GLUTAMINE Q <i>Gln</i> CAA, CAG</p>

Decarboxylation of amino acids

Decarboxylation of amino acids

Decarboxylation – removal of carbon dioxide from amino acid with formation of amines.

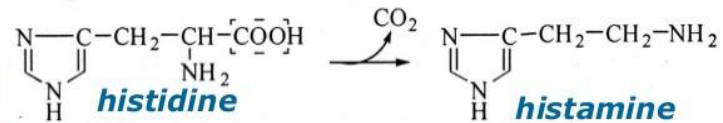
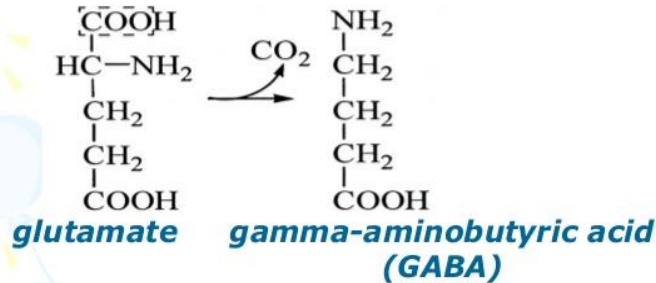


Usually amines have high physiological activity (hormones, neurotransmitters etc).

Enzyme: *decarboxylases*
Coenzyme - *pyridoxalphosphate*

Significance of amino acid decarboxylation

1. Formation of physiologically active compounds



1) A lot of histamine is formed in inflammatory place;
It has vasodilator action;
Mediator of inflammation, mediator of pain;
Responsible for the allergy development;
Stimulate HCl secretion in stomach.

-CO₂

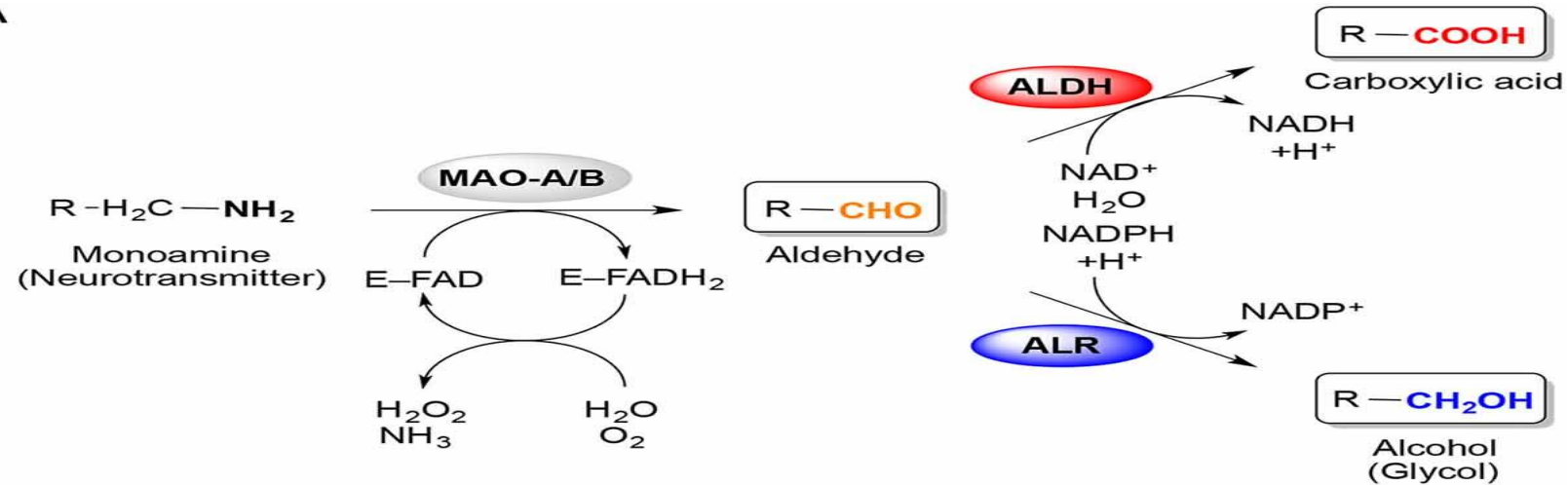
2) Tryptophan → Serotonin
Vasokonstrictor
Takes part in regulation of arterial pressure, body temperature, respiration, kidney filtration, mediator of nervous system

3) Tyrosine → Dopamine
It is precursor of epinephrine and norepinephrine.
mediator of central nervous system

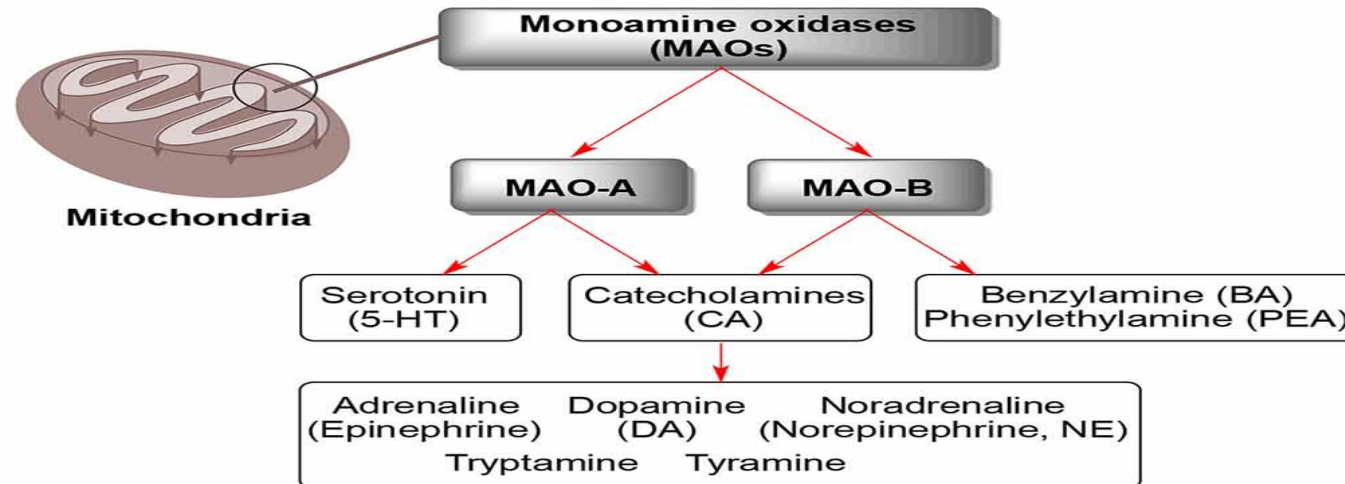
4) Glutamate → γ-aminobutyrate (GABA)
Is inhibitory mediator of central nervous system. In medicine we use with anticonvulsion purpose (action).

Degradation of biogenic amines

A

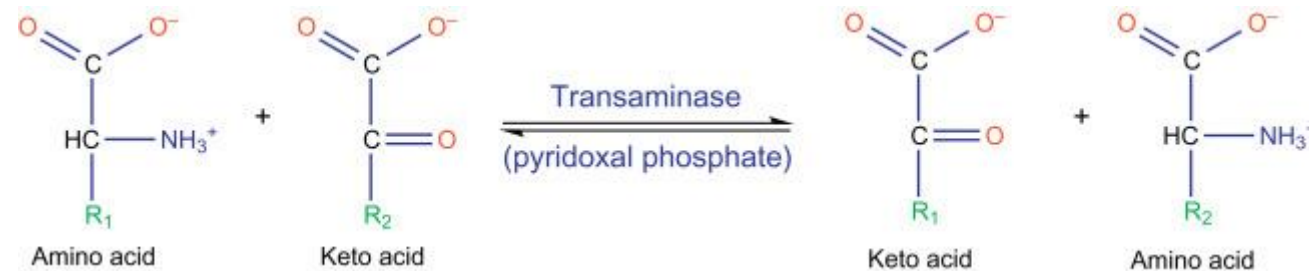


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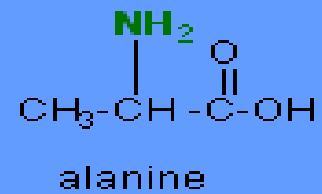


Transamination of amino acids

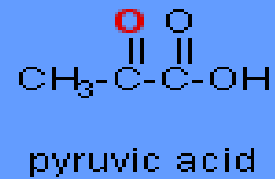
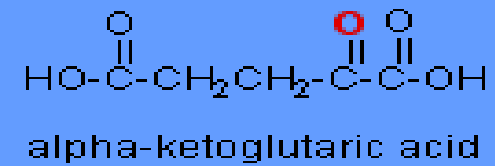
- **Transamination**, a chemical reaction that transfers an amino group to a ketoacid to form new amino acids. This reaction is catalyzed by a family of enzymes called transaminases. Actually, the transamination reaction results in the exchange of an amine group on one acid with a ketone group on another acid. It is analogous to a double replacement reaction.
- The most usual and major ketoacid involved with transamination reactions is α -ketoglutaric acid, an intermediate in the citric acid cycle. A specific example is the transamination of alanine to make pyruvic acid and glutamic acid.



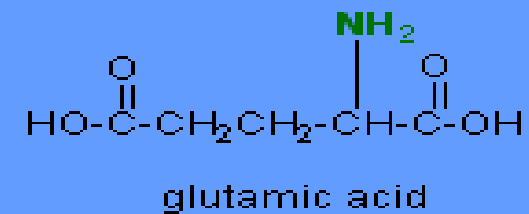
Transamination Reaction



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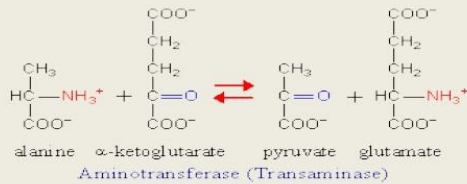
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C. Ophardt, c. 2003

ALT- Serum glutamate Alanine transferase

- ALT is found primarily in the liver.
- The normal serum activity ranges between 0-45 IU/L
- Reaction catalyzed can be represented as follows-



Clinical significance of Transaminases

Serum aminotransferases such as *serum glutamate-oxaloacetate-aminotransferase (SGOT)* (also called *Aspartate aminotransferase, AST*) and *serum glutamate-pyruvate aminotransferase (SGPT)* (also called *alanine transaminase, ALT*) have been used as clinical markers of tissue damage, with increasing serum levels indicating an increased extent of damage.

AST-Serum glutamate-oxaloacetate-aminotransferase (SGOT)

- **AST** is found in the liver, cardiac muscle, skeletal muscle, kidneys, brain, pancreas, lungs, leukocytes, and erythrocytes
- Normal serum activity is 0-41 IU/L. The concentration of the enzyme is very high in myocardium.
- The enzyme is both cytoplasmic as well as mitochondrial in nature

Diagnostic significance of amino transferases

1) Liver Diseases

- The aminotransferases are normally present in the serum in low concentrations.
- These enzymes are released into the blood in greater amounts when there is damage to the liver cell membrane resulting in increased permeability.
- These are sensitive indicators of liver cell injury and are most helpful in recognizing acute hepatocellular diseases such as hepatitis.
- Any type of liver cell injury can cause modest elevations in the serum aminotransferases

Diagnostic significance of amino transferases

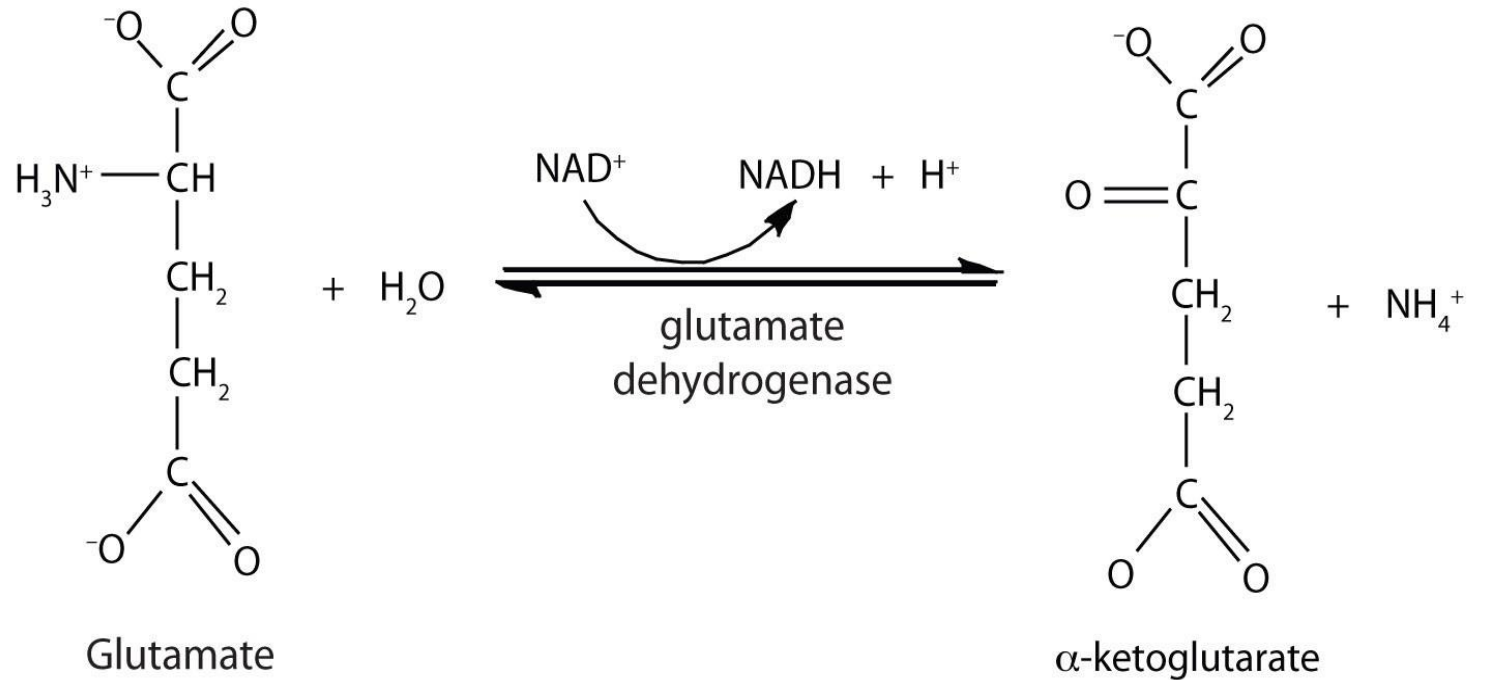
2) Acute myocardial infarction- In acute MI the serum activity rises sharply within the first 12 hours, with a peak level of 24 hours or over and returns to normal within 3 to 5 days.

3) Extra cardiac and extra hepatic conditions

- Elevation of AST can also be seen in Muscle disorders like muscular dystrophies- myositis etc.
- Increase activity of AST is also observed in acute pancreatitis, leukemias and acute hemolytic anemias
- In normal health slight rise of AST level can be observed after prolonged exercise

Oxidative deamination

This reaction occurs primarily in liver mitochondria. Most of the NH_4^+ ion formed by oxidative deamination of glutamate is converted to urea and excreted in the urine in a series of reactions known as the **urea cycle**.



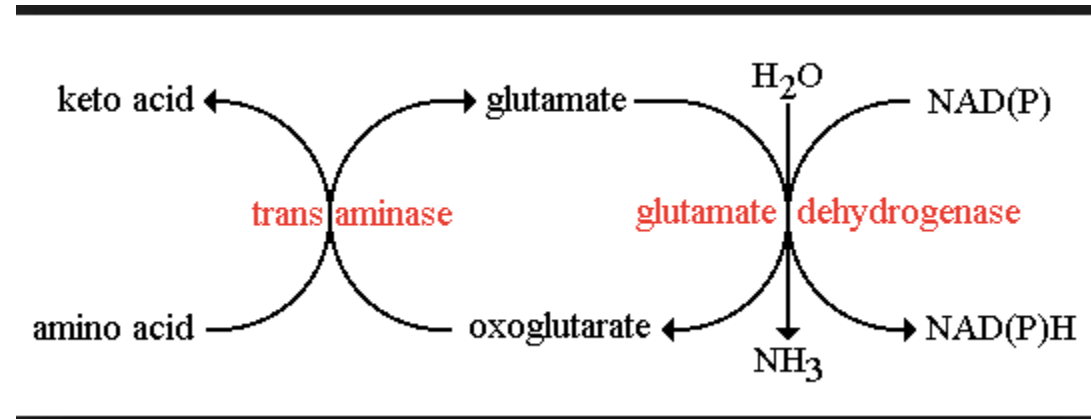
Indirect oxidative deamination (transdeamination)

Indirect oxidative deamination includes 2 stages.

The first stage consists in the reversible transfer of the NH_2 group from an amino acid to a ketoacid with the formation of a new amino acid and a new ketoacid with the participation of aminotransferase enzymes. It is reaction of transamination. the final acceptor keto acid, usually uses α -ketoglutaric acid, which is converted to glutamate.

The second stage consists in cleavage of the amino group from amino acid - deamination. Because in the body, the collector of all amino acid amino groups is glutamic acid, then only it undergoes oxidative deamination with the formation of ammonia and α -ketoglutaric acid. This stage is catalyzed by glutamate dehydrogenase.

Coupled reactions of transamination and deamination create a flow of excess amine nitrogen from peripheral cells to the liver for the synthesis of urea and to the kidneys for the synthesis of ammonium salts.



TRANSAMINATION VERSUS DEAMINATION

TRANSAMINATION

The transfer of an amino group from one molecule to another, especially from an amino acid to a keto acid

Involves in the synthesis of nonessential amino acids

Occurs in all cell of the body

Transaminases or aminotransferases catalyze transamination

Results in an exchange of an amine group with a keto group

Glutamic acid is the main form of amino acid produced in transamination reactions

Reversible

DEAMINATION

The removal of an amino group from an amino acid or other compounds

Involves in the breakdown of excess proteins

Occurs in the liver

Deaminases catalyze deamination

Results in the elimination of ammonia

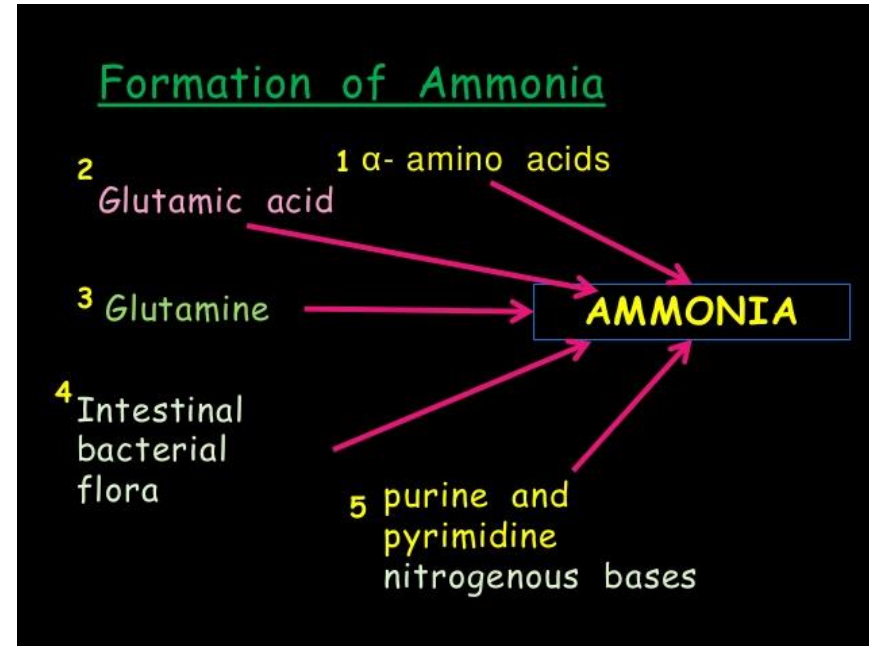
Glutamic acid is the primary form of amino acid, which undergo deamination

Irreversible

Visit www.PEDIAA.com

Ammonia sources:

- Deamination of:
amino acids;
nitrogenous bases;
biogenic amines.



Ammonia blood concentration: 25-40 $\mu\text{mol/l}$

- **Hyperammonemia** is a metabolic disturbance characterised by an excess of ammonia in the blood. It may be primary (congenital) or secondary (acquired).

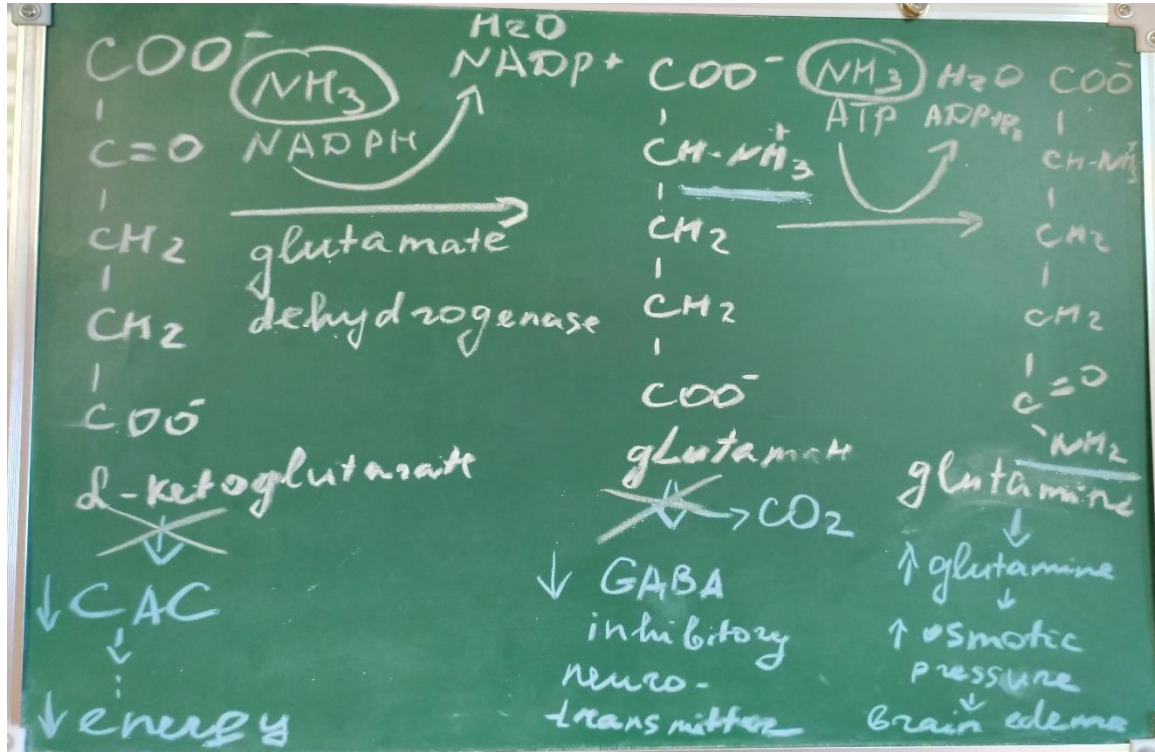
Primary: absence of the urea synthesis enzyme:

- type I hyperammonemia is hereditary - lack of carbamoyl phosphate synthetase;
- type II hyperammonemia - of ornithinecarbamoyltransferase;
- citrullinemia - lack or absence of argininosuccinate synthetase, urinary excretion of citrulline;
- argininosuccinate aciduria - absence of argininosuccinate lyase, increased levels of argininosuccinate in the blood, cerebrospinal fluid.
- hyperargininemia - low activity of arginase in erythrocytes, increased levels of arginine in the blood, cerebrospinal fluid, a lot of lysine and cysteine in the urine.

Secondary:

- liver diseases (portal hypertension, mechanical damage);
- increased putrefaction of proteins in the intestine;

Ammonia toxicity



Why ammonia is toxic ?

- Affects central nervous system
1. Alkalization of intracellular compartment
 2. Disrupts oxidative phosphorylation \rightarrow ATP depletion
 3. Increased glutamate in Brain
 4. Decreased Neurotransmitters – GABA – convulsions
 5. Cerebral edema

Ammonia toxicity

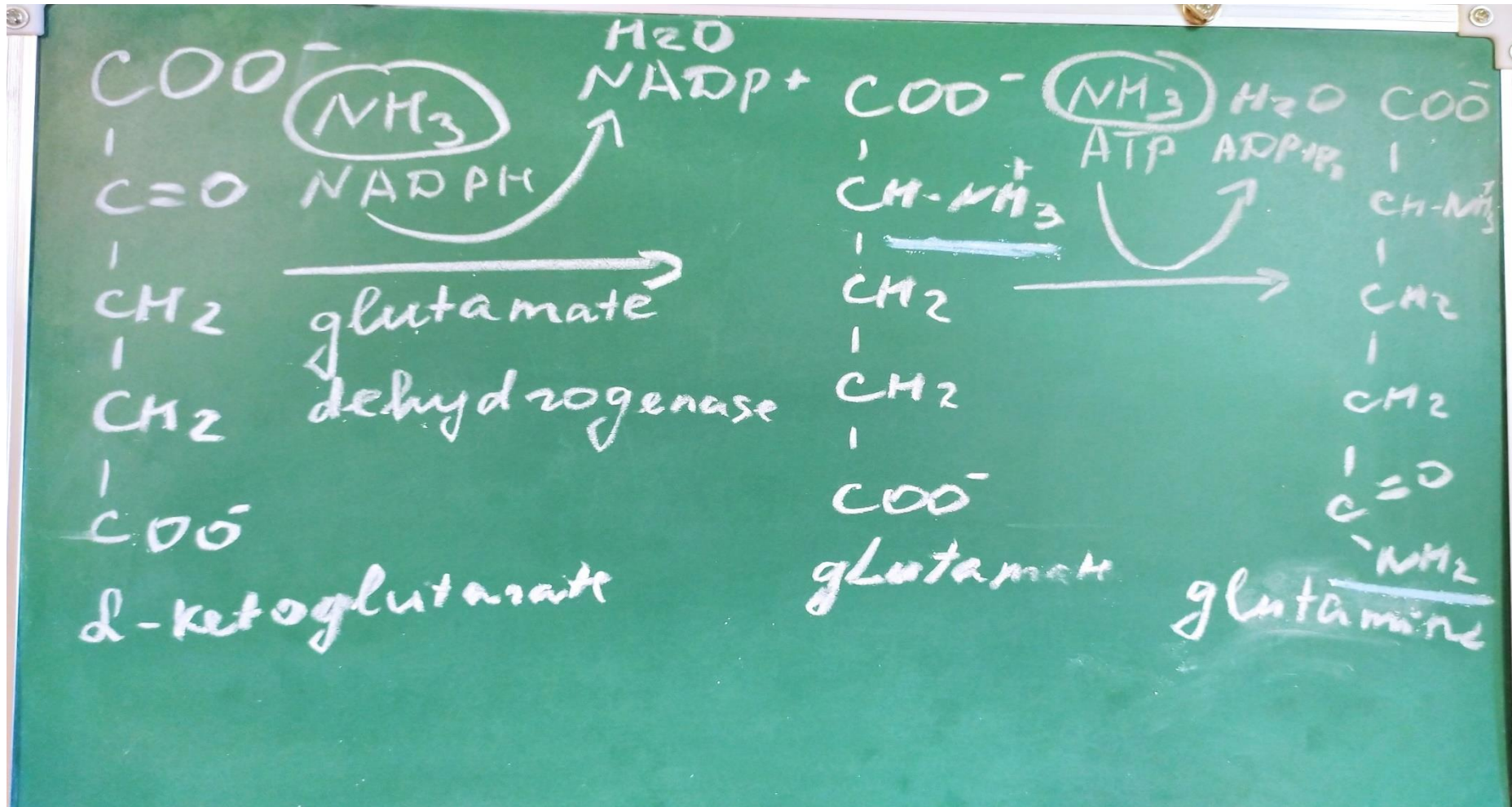
The toxicity of ammonia is associated with its effect on the central nervous system:

- ammonia passes through membranes and enters the brain cells, ammonia
- interacts with β -ketoglutarate, which leads to a decrease in the rate of glucose oxidation, deficiency of energy;
- ammonia increases the synthesis of glutamine in nervous tissue, as a result, osmotic pressure rises, cerebral edema develops;
- ammonia decreases glutamate concentration, that leads to a disruption in the metabolism of neurotransmitters (GABA), this disrupts the conduction of a nerve impulse and causes convulsions,
- ammonia in the blood and cytosol forms the NH_4^+ ion, the accumulation of which disrupts the transmembrane transport of sodium and potassium ions, which affects the conduction of nerve impulses.

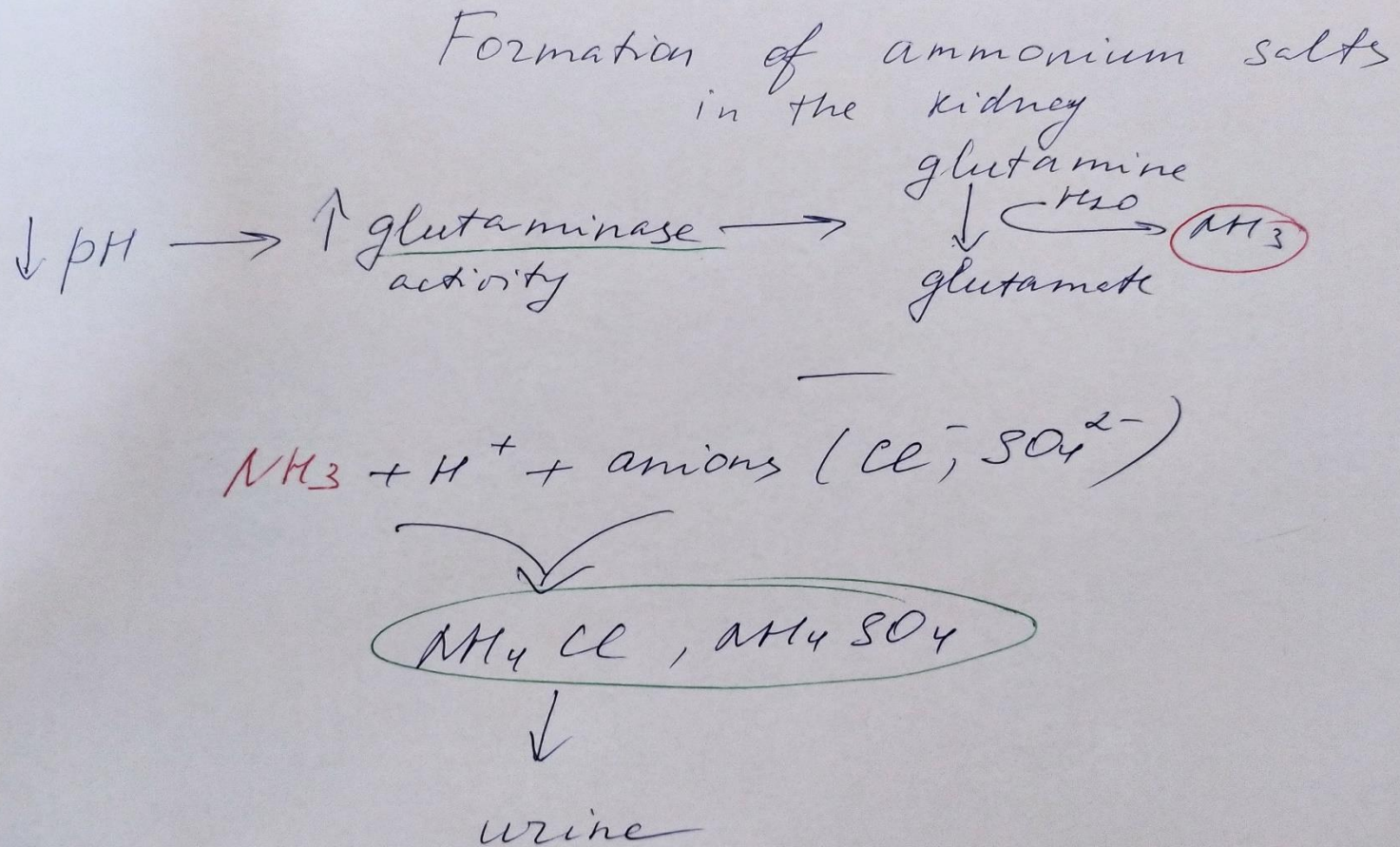
Ammonia detoxification

- Reductive amination of keto acids, formation of glutamine, asparagine (brain)
- Formation of ammonium salts (kidney)
- Synthesis of carbamoyl phosphate (liver)
- Synthesis of urea (liver)

Reductive amination of α -ketoglutarate and formation of glutamine



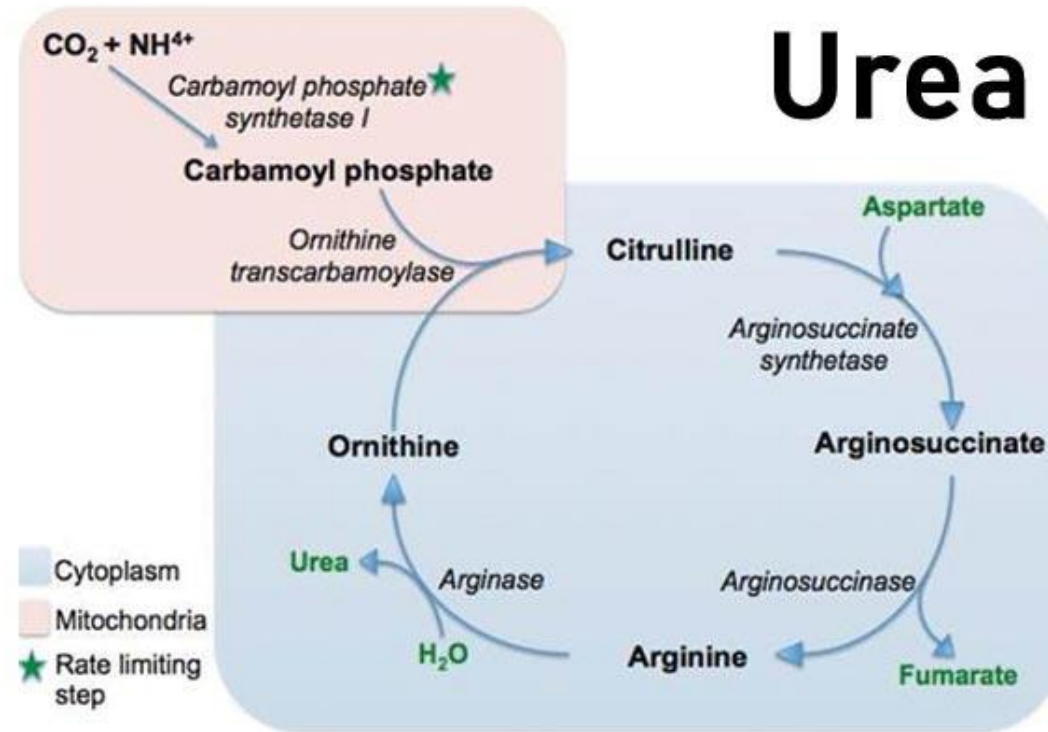
Formation of ammonium salts (kidney)



The ornithine cycle (Urea cycle) is the main pathway for ammonia detoxification

Ornithine is regenerated at each turn of the urea cycle. In urea, one amino group enters the cycle in the mitochondria during oxidative deamination of glutamate, the second amino group is supplied by aspartate from the cytosol. The urea cycle is involved in the regulation of blood pH. In the ornithine cycle, 4 high-energy bonds of three ATP molecules are consumed for each revolution of the cycle.

The process itself provides energy during the regeneration of aspartate from fumarate, the NADH₂ molecule is formed, which gives 3 ATP, during the oxidative deamination of glutamate, 3ATP is formed.



Urea Cycle

Enzymes & Steps

<https://microbenotes.com/urea-cycle-enzymes-and-steps/>

Carbamoyl phosphate synnythesis and reactions of the urea cycle

Step	Reactants	Products	Catalyzed by	Location
1	$\text{NH}_4^+ + \text{HCO}_3^- + 2\text{ATP}$	Carbamoyl phosphate + $2\text{ADP} + \text{P}_i$	Carbamoyl phosphate synthetase I	mitochondria
2	Carbamoyl phosphate + Ornithine	Citrulline + P_i	Ornithine transcarbamoylase	mitochondria
3	Citrulline + Aspartate + ATP	Argininosuccinate + AMP + pyrophosphate	Argininosuccinate synthetase	cytosol
4	Argininosuccinate	Arginine + Fumarate	Argininosuccinase	cytosol
5	Arg + H_2O	Ornithine + Urea	Arginase	cytosol

Urea cycle regulation

Urea cycle is regulated by the rate limiting enzyme carbamoyl phosphate synthase I, the first enzyme of the ammonia detoxification pathway. It is only active in presence of its allosteric activator N-methyl-glutamate amino acid. It catalyses the condensation of ammonium ions NH_4^+ , CO_2 and ATP to form carbamoyl phosphate, a product that will condense with L-ornithine in order to initiate the urea cycle.

Blood urea concentration: 2.5-8.3 mmol/l

Causes of urea decreasing: liver failure, low protein diet, malabsorption, nephrotic syndrome

Causes of urea increasing: kidney disease (pyelonephritis, glomerulonephritis), heart failure, increased protein catabolism

Urine urea excretion: 20-35 g/day

Increased excretion: High protein diet, increased catabolism of proteins, hyperthyroidism, fever.

Decreased excretion: Low protein diet and high in carbohydrates, convalescence period, Liver diseases, kidney disease and renal failure, disorders of urea formation.

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