Urea Cycle and associated disorders

Department of Biochemistry

Specific Learning Objectives

- Describe factors affecting nitrogen balance in health and disease
- Explain rationale of urea cycle in ammonia excretion
- List two subcellular compartments used by urea cycle
- Describe reactions of Urea Cycle, including specific enzymes, input substrates (NH4, HCO3, ornithine, and aspartate), and energy requirements
- Describe urea cycle regulation by allosteric effectors, substrate availability, and enzyme levels
- Outline steps of Urea cycle and inherited disorders associated with urea cycle
- Identify connections and common intermediates between Urea Cycle and TCA cycle

Urea cycle and reactions that feed amino groups into cycle

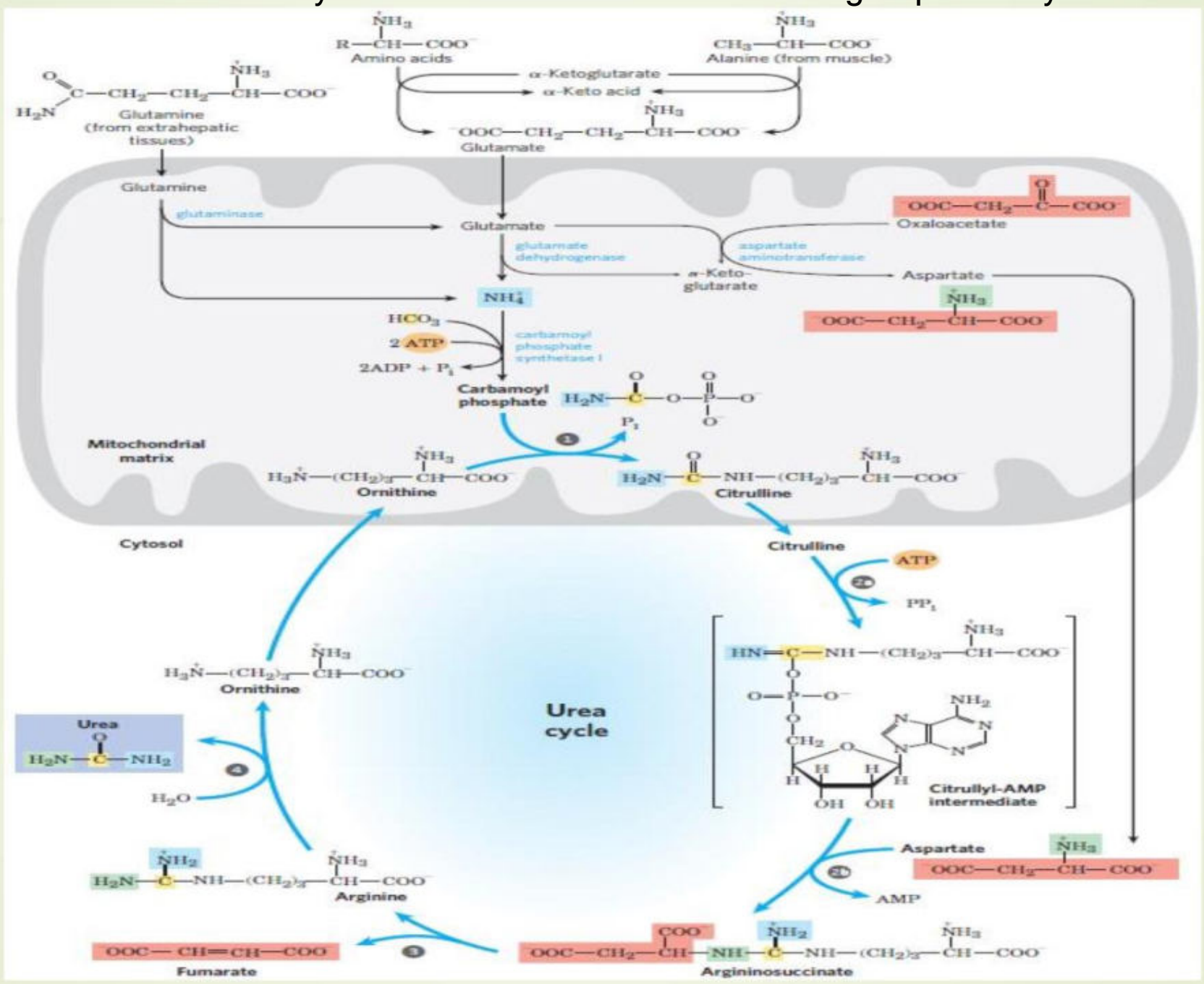
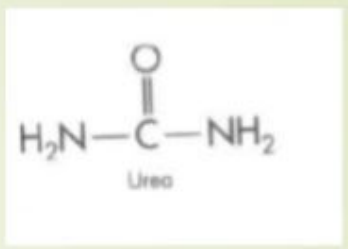


Fig18.10: Lehninger Principles of Biochemistry by David L Nelson, 6th Ed.

Introduction



- Urea is major disposal form of amino groups derived from aa.
- It accounts for about 86-90% of the nitrogen containing components of urine
- One nitrogen group of urea is supplied by free NH_3 and other nitrogen from aspartate

Cont--

- Glutamate is immediate precursor of both ammonia groups by following reactions:

Transamination: transfer of ammonia to oxaloacetate to form aspartate transaminase (AST)

Oxidative deamination: removal of free ammonia, glutamate dehydrogenase

Cont--

- Carbon and oxygen of urea are derived from CO2
- Urea is produced by Liver and then transported in blood to kidneys for excretion in kidneys
- Blood urea level is measured as blood urea nitrogen (BUN). Levels are 8-20 mg/dl
- BUN is low in liver failure and is very high in patients with renal failure in uremia.

Nitrogen Balance

Nitrogen balance occurs when synthesis of body protein equals degradation.

- Amount of nitrogen excreted in urine each day equals amount of nitrogen ingested each day

Positive nitrogen balance occurs when synthesis of body protein excess compare to degradation.

- Less nitrogen is excreted than ingested (growth, e.g. growing infants and children, pregnant women, tissue repair)

Cont--

Negative nitrogen balance occurs when synthesis of body protein lesser compare to degradation.

- More nitrogen is excreted than ingested (malnutrition, absence of one or more essential aa in diet)
- It occurs in injury, stress response, malnutrition of essential aa

Urea Biosynthesis

- Synthesis of 1 mol of urea requires 3 mol of ATP, 1 mol each of NH_4+ and of aspartate, and five enzymes

- Of six participating aa, N-acetylglutamate as an enzyme activator, others serve as carriers of atoms that become urea

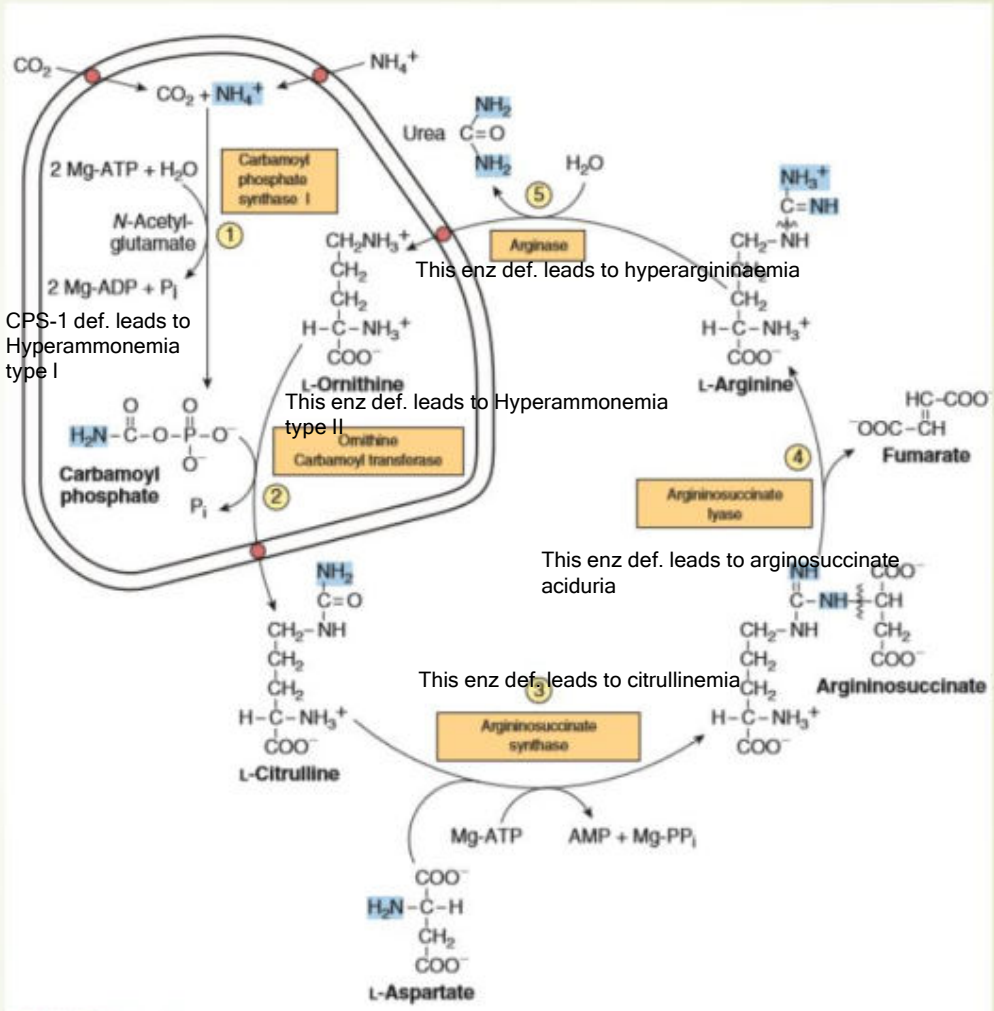


FIGURE 28–16 **Reactions and intermediates of urea biosynthesis.** The nitrogen-containing groups that contribute to the formation of urea are shaded. Reactions ① and ② occur in the matrix of liver mitochondria and reactions ③, ④, and ⑤ in liver cytosol. CO_2 (as bicarbonate), ammonium ion, ornithine, and citrulline enter the mitochondrial matrix via specific carriers (see red dots) present in the inner membrane of liver mitochondria.

Fig 28.16. Harper's Illustrated Biochemistry 30th Edition

Cont--

- While ammonium ion, CO2, ATP, and aspartate are consumed, ornithine consumed in reaction 2 is regenerated in reaction 5

- Thus is no net loss or gain of ornithine, citrulline, argininosuccinate, or arginine

Cont--

- Ammonia, which is very toxic in humans, is converted to urea, which is nontoxic, very soluble, and readily excreted by kidneys
- Enzymes of urea cycle are induced if a high-protein diet is consumed for several days
- When nitrogen of aa is converted to urea in liver, their carbon skeletons are converted either to glucose (in fasting state) or to fatty acids (in fed state)

Regulation of urea cycle

- Carbamoyl phosphate synthetase I (CPS-I), is allosterically activated by N-Acetylglutamate
- Steady-state levels of N-acetylglutamate are determined by concentrations of glutamate and acetyl-CoA and arginine
- Arginine is an activator of N-acetylglutamate synthase, and thus an activator of urea cycle

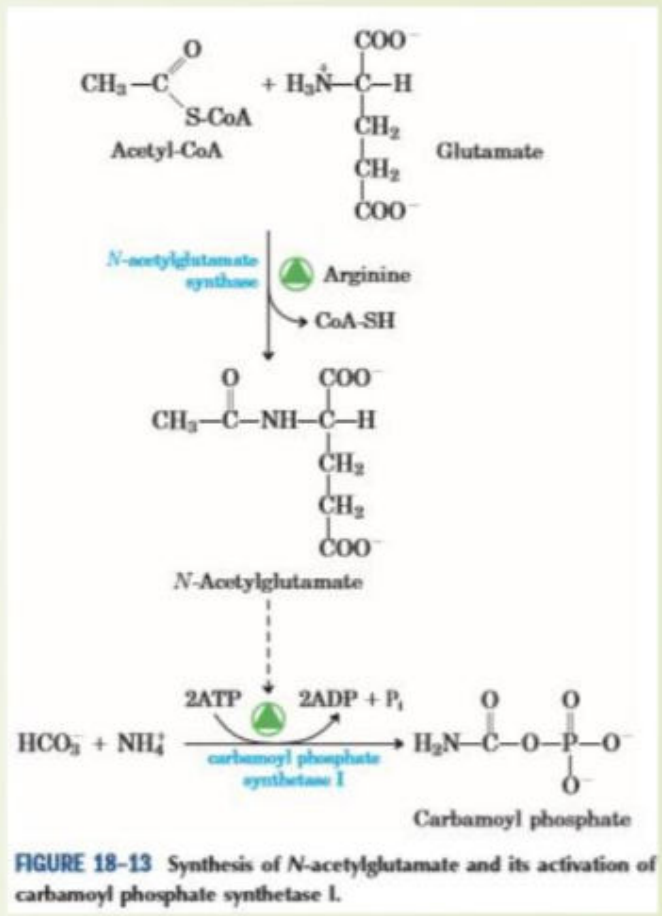


FIGURE 18–13 Synthesis of *N*-acetylglutamate and its activation of carbamoyl phosphate synthetase I.

Fig18.13: Lehninger Principles of Biochemistry by David L Nelson, 6th Ed.

Disorders of Urea cycle

- Urea cycle disorders are characterized by hyperammonemia, encephalopathy, and respiratory alkalosis
- Deficiencies of CPS-1, ornithine carbamoyl transferase, argininosuccinate synthase, and argininosuccinate lyase, result in accumulation of precursors of urea, principally ammonia and glutamine
- Ammonia intoxication is most severe when metabolic block occurs at reactions 1 or 2 of urea cycle
- Leads to feeding difficulties, vomiting ataxia, lethargy, irritability, poor intellectual development

Cont--

- Hyperammonemia Type I: Deficiency of CPS-1, infants die in neonatal period
- Hyperammonemia Type II: Deficiency of ornithine transcarbamoylase, Levels of glutamine are elevated in blood, cerebrospinal fluid, and urine, result of enhanced glutamine synthesis in response to elevated levels of tissue ammonia

Cont--

Citrullinemia: Patients who lack detectable argininosuccinate synthase activity, citrulline levels elevated

- Plasma and cerebrospinal fluid citrulline levels are elevated, and 1 to 2 g of citrulline are excreted daily.

Cont--

Argininosuccinic aciduria: Metabolic defect is in argininosuccinate lyase

- Elevated levels of argininosuccinate in blood, CSF, and urine, is associated with friable, tufted hair.

- Diagnosis by measurement of erythrocyte argininosuccinate lyase activity can be performed on umbilical cord blood or amniotic fluid cells

Hyperargininemia: is an autosomal recessive defect in gene for arginase

- Blood and CSF levels of arginine are elevated
- Urinary aa pattern, which resembles that of lysine-cystinuria may reflect competition by arginine with lysine and cysteine for reabsorption in renal tubule

Gene Therapy for Correcting Defects in Urea Biosynthesis:

- Gene therapy for rectification of defects in enzymes of urea cycle is an area of active investigation
- Animal models using an adenoviral vector to treat citrullinemia

Links between urea cycle and citric acid cycle

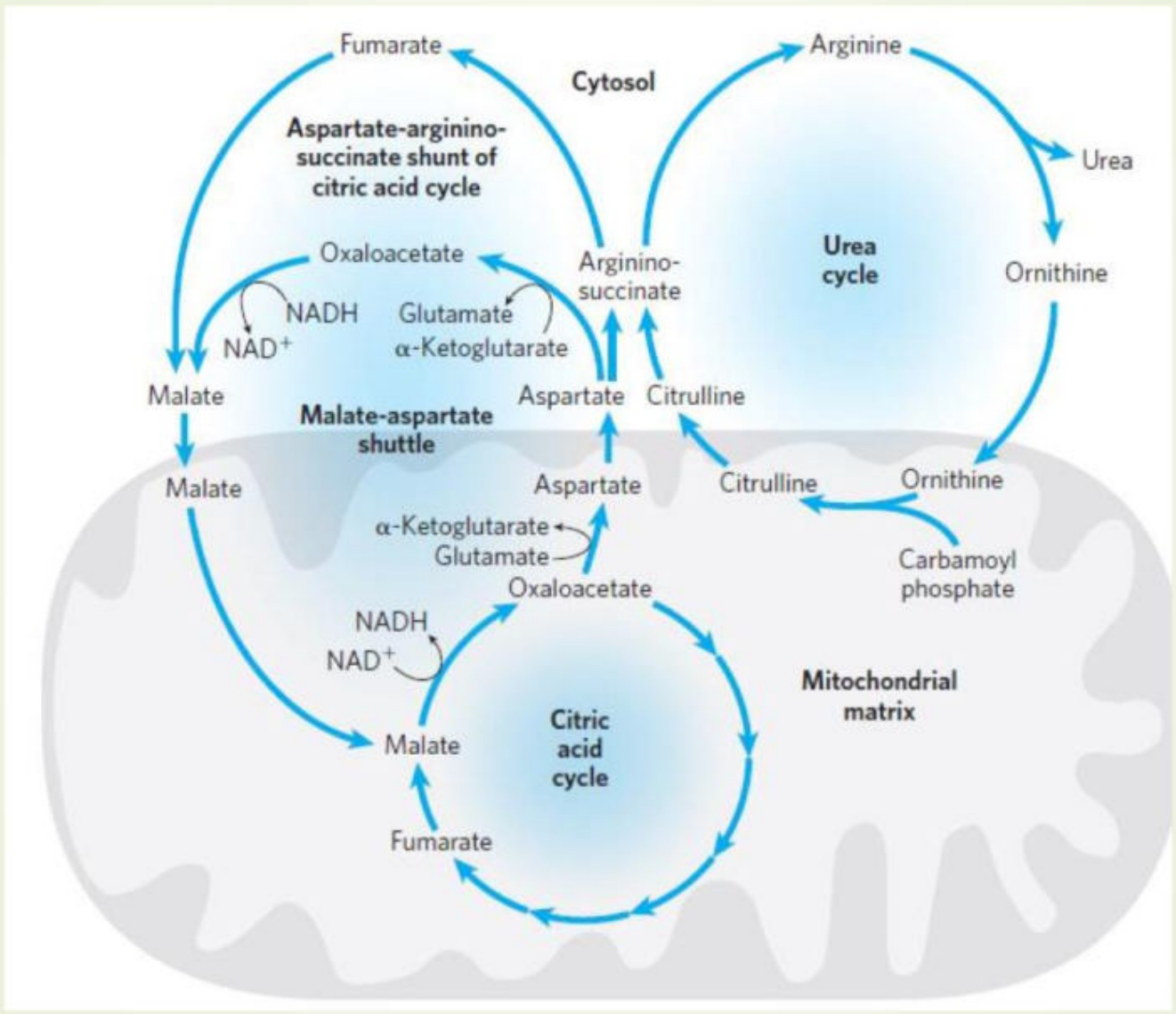


Fig18.12: Lehninger Principles of Biochemistry by David L Nelson, 6th Ed.

Reference Books

1) Text Book of Medical Biochemistry by Chatterjee & Rana Shinde, 8th Ed
2) Biochemistry, Lippincott's Illustrated Reviews, 6th Ed
3) Harper's Illustrated Biochemistry-30th Ed
4) Lehninger Principles of Biochemistry-6th Ed

Two Clinical-cases discussed

Group Discussion

- Subtopics of previous and today's class discussed in group discussion.

Thank you