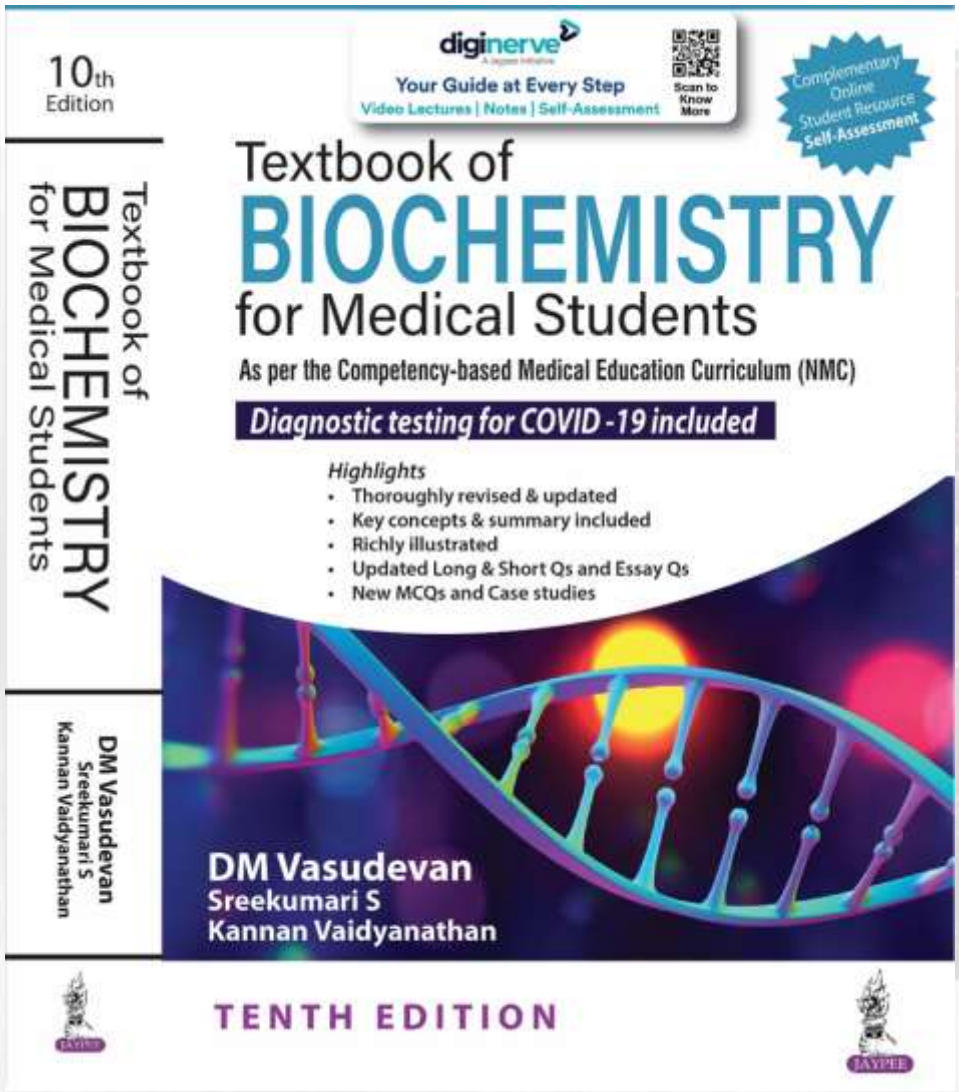


Chapter 15:

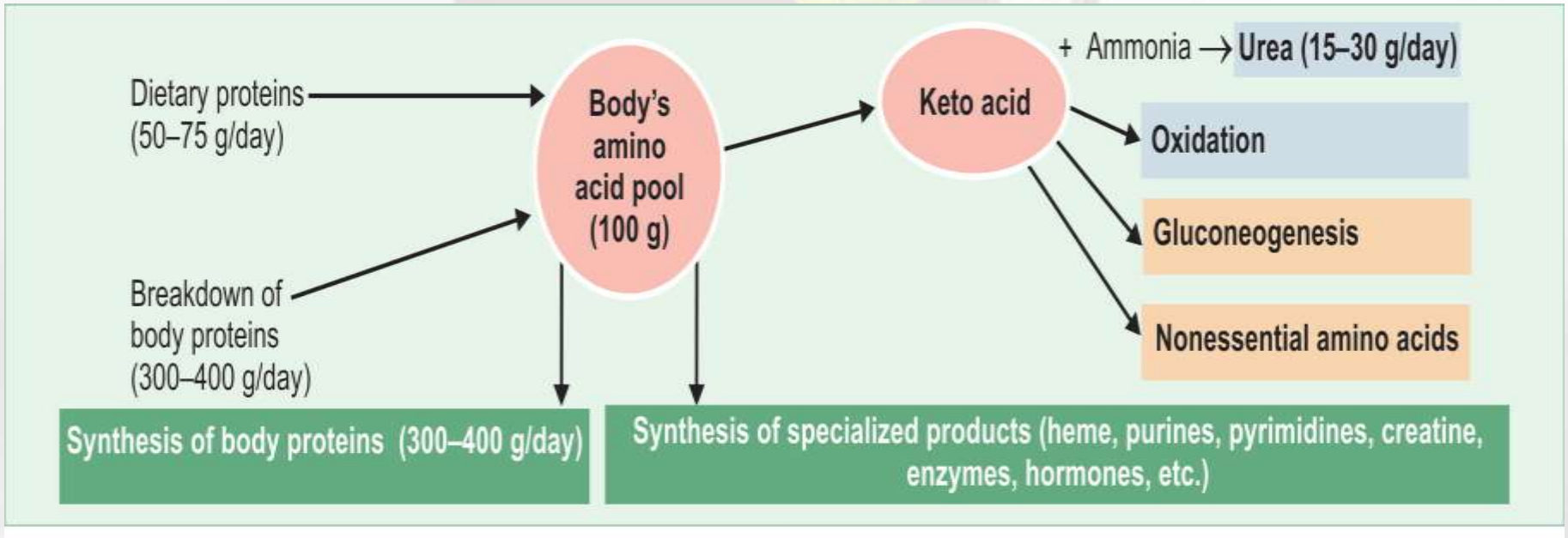
Proteins, Digestion, Absorption, General Amino Acid Metabolism: Urea Cycle, and One-Carbon Metabolism

Textbook of
BIOCHEMISTRY
for Medical Students

By DM Vasudevan, *et al.*

TENTH EDITION

- The main role of amino acids is in the **synthesis of structural and functional proteins**.
- A 70 kg man has an average protein turnover rate of 400 g per day (same amount synthesized and same amount broken down).
- The nonessential amino acids are either derived from the diet or synthesized in the body.
- The **essential amino acids are obtained from the diet**.
- Even if one amino acid is deficient, protein synthesis cannot take place.
- The body amino acid pool is always in a dynamic steady state.
- In an adult, the rate of synthesis of proteins balances the rate of degradation, so that nitrogen balance is maintained proteins are generally not used for providing energy



Overview of metabolism of amino acids

Proteolytic enzymes are secreted as inactive zymogens.

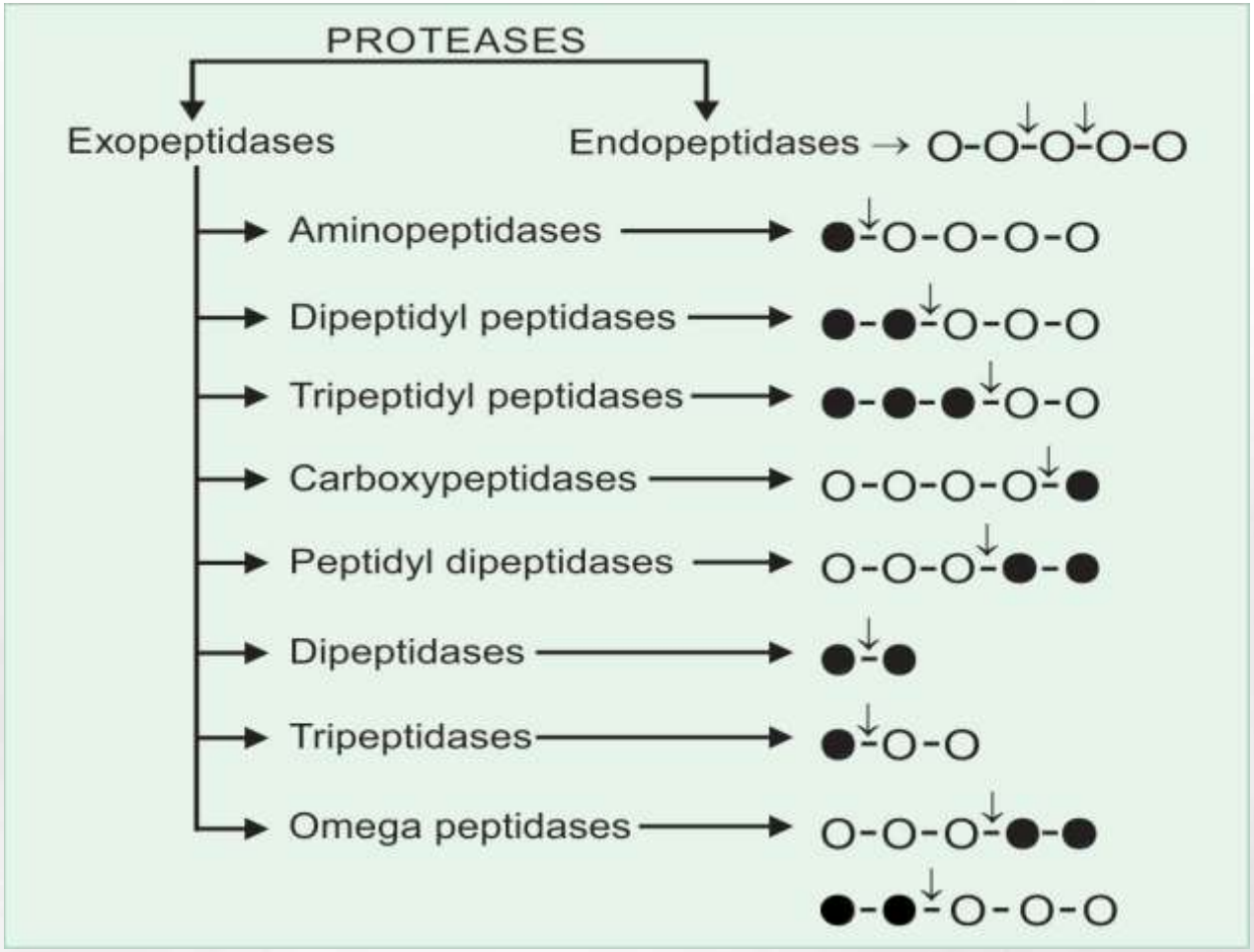
These are then converted to their active form in the intestinal lumen.

This would prevent auto-digestion of the secretory acini.

Premature activation of trypsinogen results in acute pancreatitis.

It is a life-threatening condition.

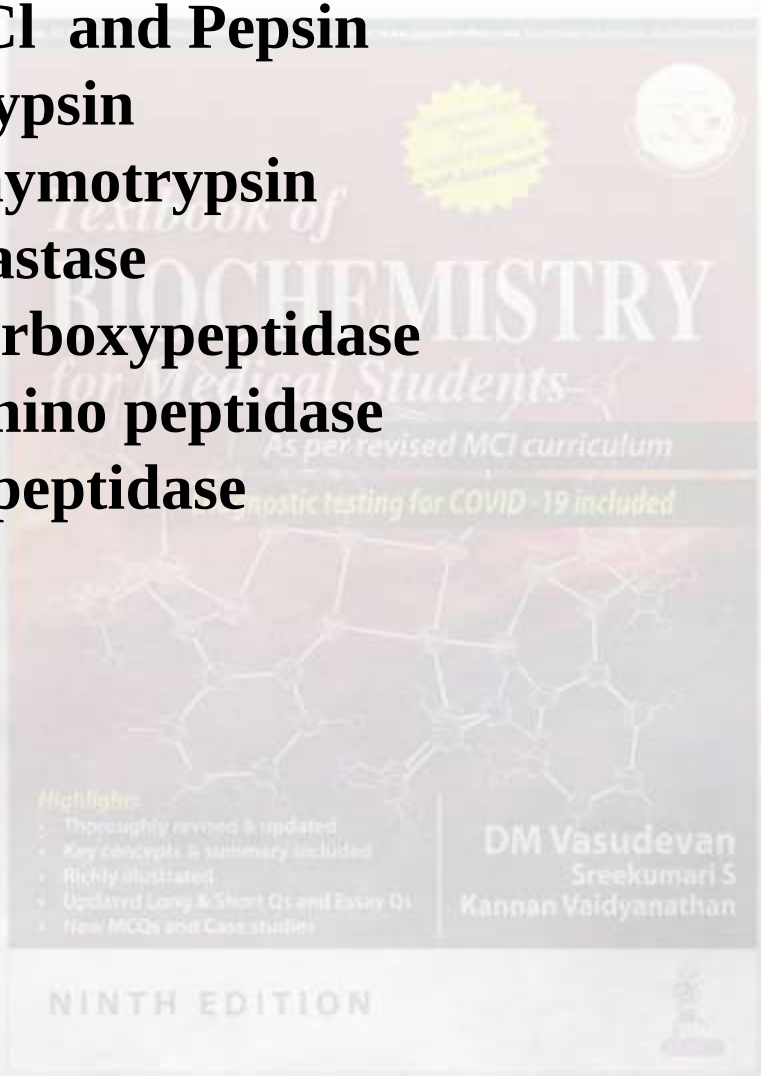
- The proteolytic enzymes include:
  1. **Endopeptidases:** They act on peptide bonds inside the protein molecule, so that the protein becomes successively smaller and smaller units.
- This group includes Pepsin, Trypsin, Chymotrypsin and Elastase.
  2. **Exopeptidases:** Which act only on the peptide bond located at the ends of the polypeptide chain.
- This group includes:
  a. **Carboxypeptidase,** which acts only on the peptide bond at the carboxy terminal end of the chain.
  b. **Aminopeptidase,** which acts only on the peptide bond at the amino terminal end of the chain.



Action of proteases. The enzyme hydrolyses the peptide bond at the site of arrow

Digestion of Proteins to Amino Acids

Stomach :	**HCl and Pepsin**
Pancreas :	**Trypsin**
	Chymotrypsin
	Elastase
Intestine:	**Carboxypeptidase**
	Amino peptidase
	Dipeptidase



Gastric Digestion of Proteins

- In the stomach, hydrochloric acid is secreted.
- It makes the pH optimum for the action of pepsin and also activates pepsin.
- The acid also denatures the proteins.

Rennin

- Rennin otherwise called **Chymosin**, is active in infants and is involved in the curdling of milk.
- It is absent in adults.
- Milk protein, casein is converted to paracasein by the action of rennin.
- This denatured protein is easily digested further by pepsin.

Pepsin

- It is secreted by the chief cells of stomach as inactive **pepsinogen**.
- The conversion of pepsinogen to pepsin is brought about by removal of 44 amino acids from the N-terminal end, by the hydrochloric acid.
- The optimum pH for activity of pepsin is **around 2**.
- Pepsin is an endopeptidase.
- Pepsin catalyzes hydrolysis of the bonds formed by carboxyl groups of Phe, Tyr, Trp and Met.
- By the action of pepsin, proteins are broken into proteoses and peptones.

Rennin is active in infants and is involved in the curdling of milk. It is absent in adults. Milk protein, casein is converted to paracasein by the action of rennin.

Rennin and Renin are different

Rennin is the proteolytic enzyme present in gastric juice.
Renin is proteolytic enzyme, secreted by kidneys. It is involved in the activation of angiotensinogen to angiotensin, a hypertensive agent.

Pancreatic Digestion of Proteins

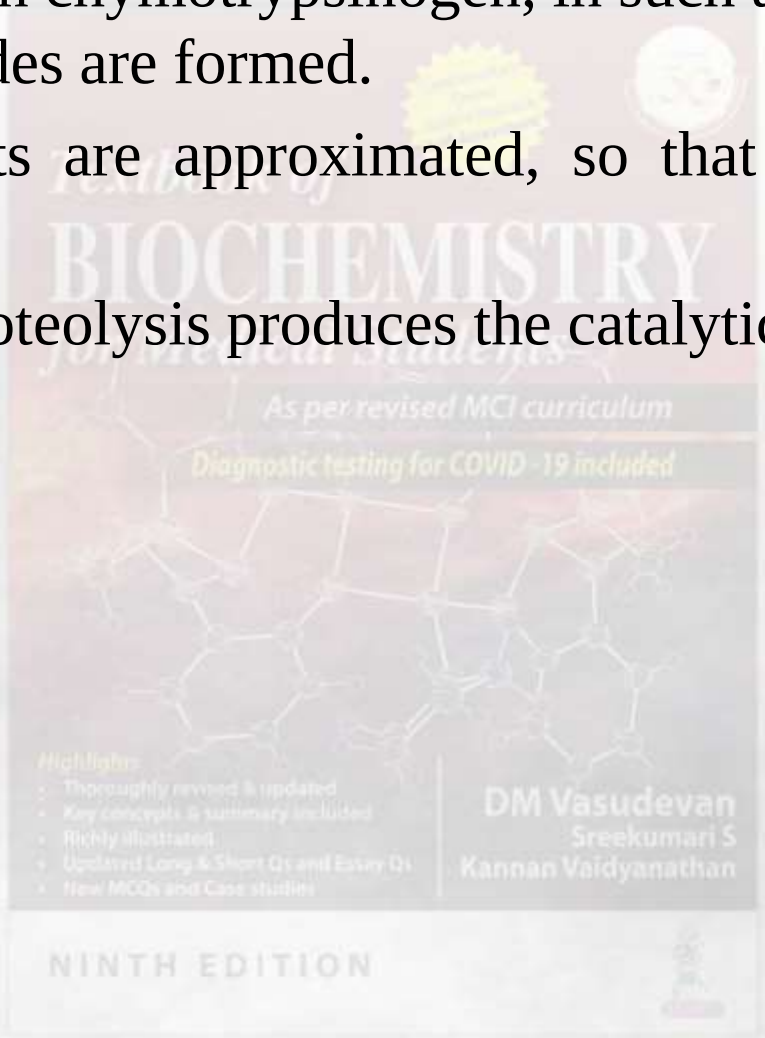
- The optimum pH for the activity of pancreatic enzymes (pH 8) is provided by the alkaline bile and pancreatic juice.
- The secretion of pancreatic juice is stimulated by the peptide hormones, **Cholecystokinin** and **Pancreozymin.**
- Pancreatic juice contains the important endopeptidases, namely **Trypsin, Chymotrypsin, Elastase** and **Carboxypeptidase.**
- These enzymes are also secreted as zymogens (trypsinogen, chymotrypsinogen and proelastase), so that the pancreatic acinar cells are not autolyzed.
- All the three are serine proteases, i.e. the active centers of these enzymes contain serine residues.

Trypsin

- Trypsinogen is activated by **enterokinase** (enteropeptidase) present on the intestinal microvillus membranes.
- Once activated, the trypsin activates other enzyme molecules.
- Trypsin is activated by the removal of a hexapeptide from N-terminal end.
- Trypsin catalyzes hydrolysis of the bonds formed by carboxyl groups of Arg and Lys.
- **Acute pancreatitis:** Premature activation of trypsinogen inside the pancreas itself, will result in the autodigestion of pancreatic cells.
- The result is acute pancreatitis.
- It is a life-threatening condition.

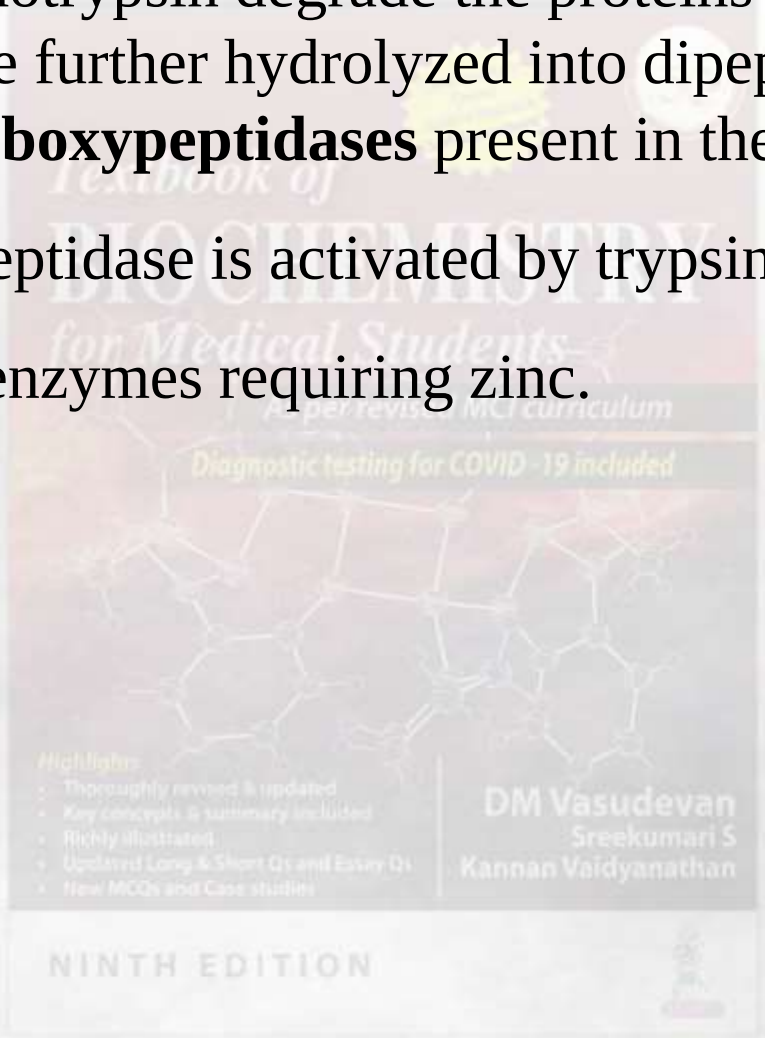
Chymotrypsin

- Trypsin will act on chymotrypsinogen, in such a manner that A, B and C peptides are formed.
- These 3 segments are approximated, so that the active site is formed.
- Thus selective proteolysis produces the catalytic site.



Carboxypeptidases

- Trypsin and chymotrypsin degrade the proteins into small peptides; these are further hydrolyzed into dipeptides and tripeptides by **carboxypeptidases** present in the pancreatic juice.
- The procarboxypeptidase is activated by trypsin.
- They are metalloenzymes requiring zinc.



Pepsin (endopeptidase)	***aromatic* (tyr,phe, trp)** ***acidic* (glu)**
Trypsin (endopeptidase)	***basic* (arg, lys)**
Chymotrypsin (endopeptidase)	***aromatic* (trp, phe, tyr,met)**
Elastase (endopeptidase)	***neutral* (gly, ser, ala)**
Carboxy-peptidase A (exopeptidase)	**tyr, phe, trp val, leu, ile**
Carboxy- peptidase B (exopeptidase)	***basic* (arg, lys)**

Intestinal Digestion of Proteins

- Complete digestion of the small peptides to the level of amino acids is brought about by enzymes present in intestinal juice (**succus entericus**).
- The luminal surface of intestinal epithelial cells contains the following enzymes:
- Aminopeptidases release the N-terminal amino acids.
- Dipeptidases and tripeptidases will complete the digestion of proteins.

Summary of Digestion of Proteins

Enzyme	Digestivity juice	Activator	Bond specificity	Products
Pepsin Endopeptidase Aspartyl Protease	Gastric juice	HCl-Pepsinogen to Pepsin	Peptide bonds formed by carboxyl groups of Phe, Tyr, Trp & Met	Proteoses and peptones
Rennin	Gastric Juice	HCl-Curdles milk		Casien to Ca-paracasienate
Trypsin Endopeptidase Serine preotease	Pancreatic juice	Enterokinase-Trypsinogen to trypsin CCK	Peptide bonds formed by carboxyl groups of basic amino acids Arg & Lys	Proteoses and peptones and peptides

Summary of Digestion of Proteins

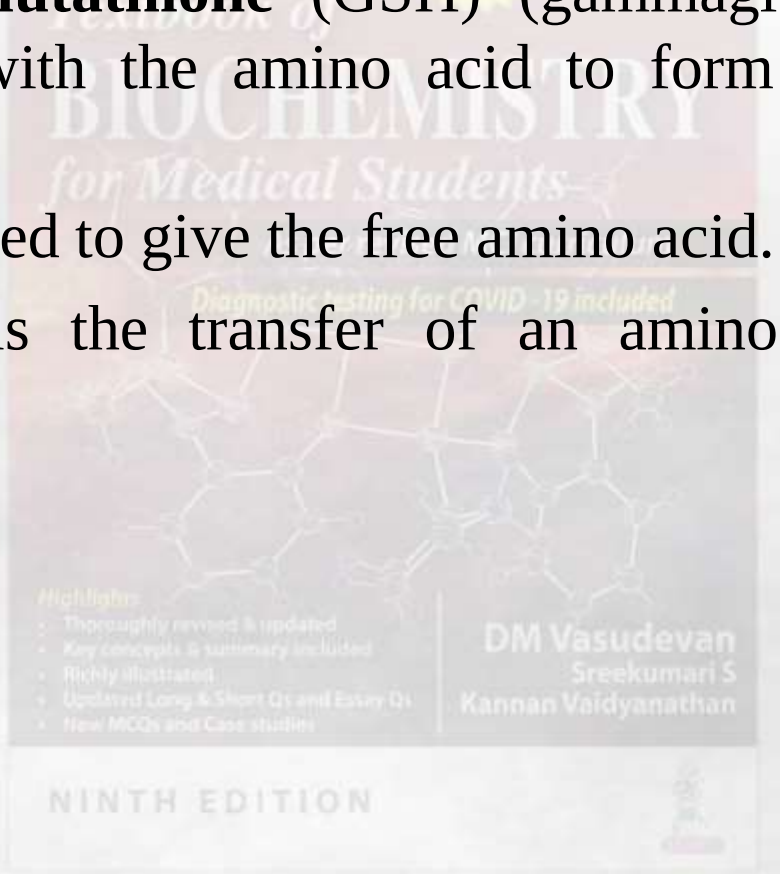
Enzyme	Digestivity juice	Activator	Bond specificity	Products
Chymotrypsin Endopeptidase Serine preotease	Pancreatic Juice	Trypsin	Peptide bonds of hydrophobic amino acids, Met, Leu,and aromatic amino acids	Proteoses and peptones and peptides
Elastase Endopeptidase Serine preotease	Pancreatic juice	Trypsin		Proteoses and peptones and peptides
Carboxy peptidase Exopeptidase	Pancreatic juice	Trypsin	C terminal amino acid	Free amino acid
Amino peptidases	Intestinal juice		N terminal amino acid	Free amino acid
Tri- and Dipeptidases	Intestinal juice		Peptide bonds of small peptides	Free amino acids

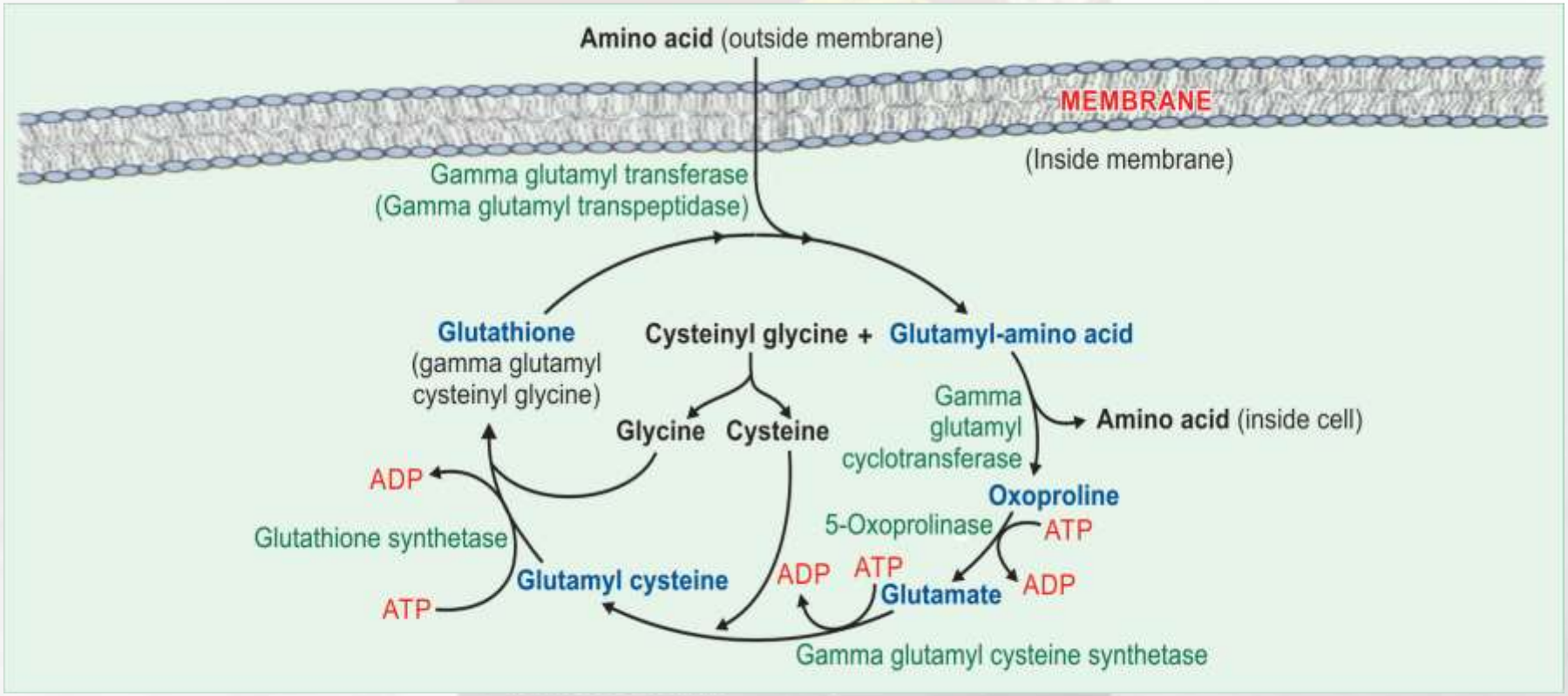
Absorption of Amino Acids

- The absorption of amino acids occurs mainly in the small intestine.
- It is an energy requiring process.
- These transport systems are carrier mediated and/or ATP sodium dependent symport systems.
- There are **5 different carriers** for amino acids:
  1. Neutral amino acids (Ala, Val, Leu, Met, Phe, Tyr, Ile)
  2. Basic amino acids (Lys, Arg) and Cysteine
  3. Imino acids and Glycine
  4. Acidic amino acids (Asp, Glu)
  5. Beta amino acids (beta alanine).

Miester Cycle (Gamma-Glutamyl Cycle)

- In intestines, kidney tubules and brain, the absorption of neutral amino acids is effected by the gamma-glutamyl cycle.
- The tripeptide **glutathione** (GSH) (gammaglutamyl- cysteinyl glycine) reacts with the amino acid to form gamma-glutamyl amino acid.
- That is then cleaved to give the free amino acid.
- The net result is the transfer of an amino acid across the membrane.

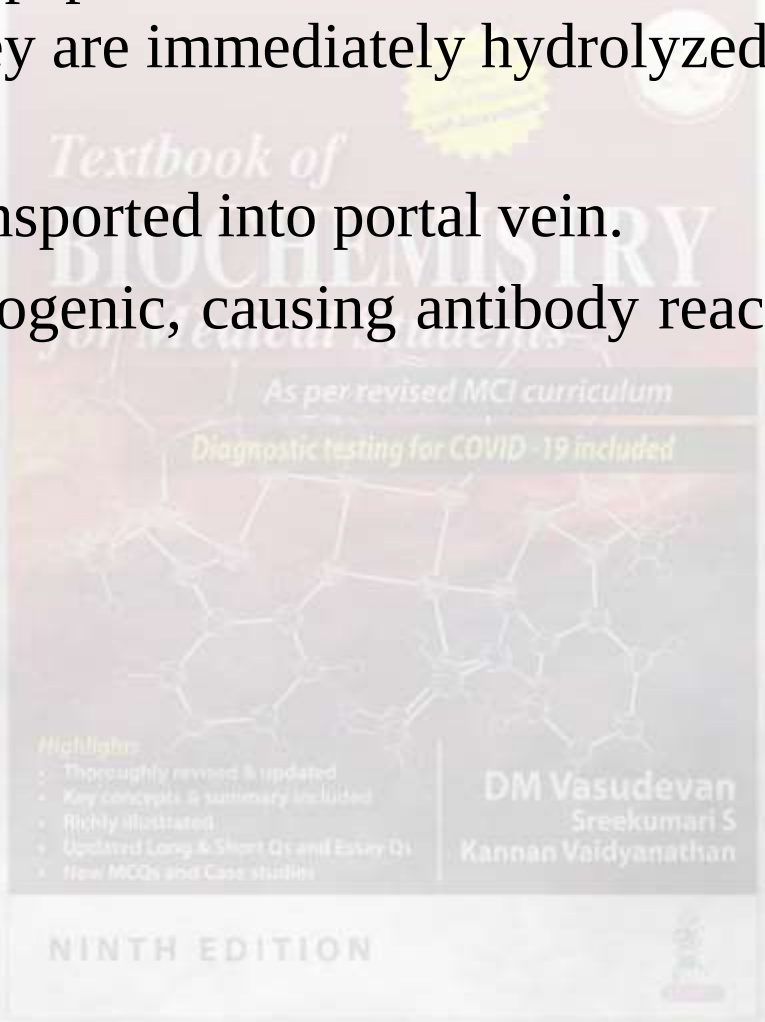




Gamma-glutamyl cycle (Meister cycle).

Food Allergy

- Dipeptides and tripeptides can enter the brush border of mucosal cells; they are immediately hydrolyzed into single amino acids.
- They are then transported into portal vein.
- These are immunogenic, causing antibody reaction, leading to food allergy.



Clinical Applications

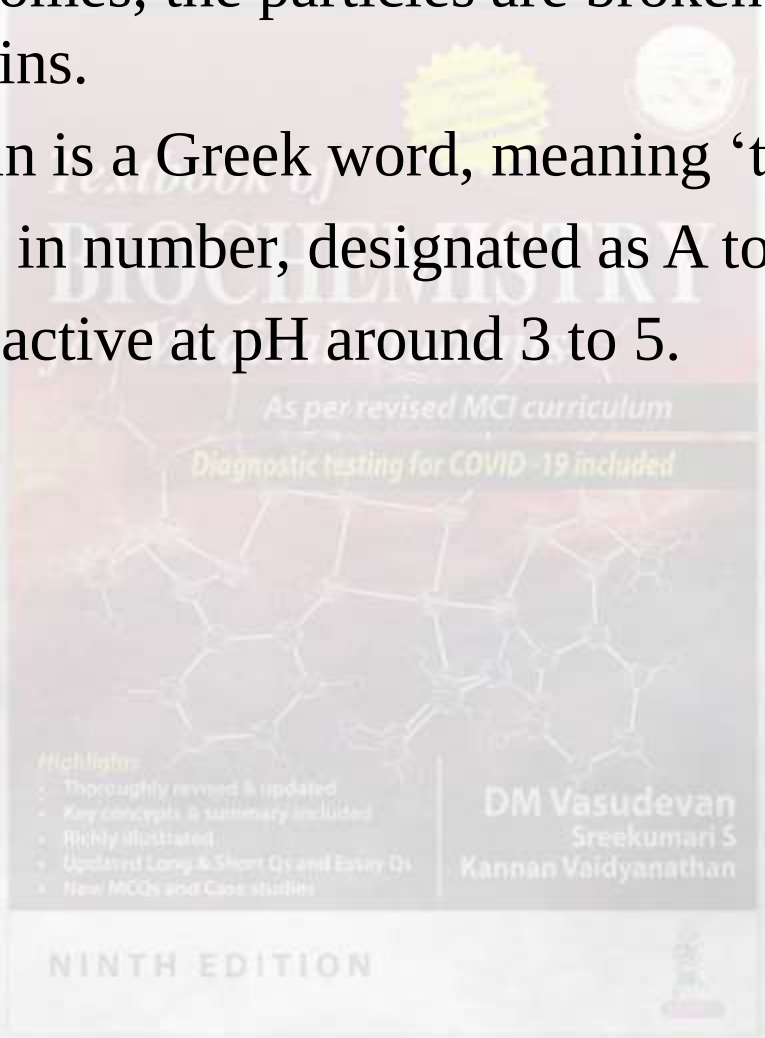
1. The deficiency of the enzyme 5-oxoprolinase leads to **oxoprolinuria** (pyroglutamic aciduria).
2. The allergy to certain food proteins (milk, fish) is believed to result from absorption of partially digested proteins.
3. Defects in the intestinal amino acid transport systems are seen in inborn errors of metabolism such as:
   a. **Hartnup disease**
   b. Imino glycinuria
   c. Cystinuria
4. Partial gastrectomy, pancreatitis, carcinoma of pancreas and cystic fibrosis may affect the digestion and absorption of proteins.
5. **Protein-losing enteropathy:** There is an excessive loss of proteins through the gastrointestinal tract.

Intracellular Protein Degradation

- All proteins in the body are constantly being degraded.
- Half-life (t *1/2*) of a protein is the time taken to lower its concentration to half of the initial value.
- General tissue proteins have half lives of few hours.
- Key enzymes have half lives of about few minutes only.
- **PEST sequence** (areas rich in proline, glutamate, serine and threonine) on a protein will give an inherent message to breakdown that protein very quickly.
- Proteins are taken by endocytosis and are fused with **lysosomes.**
- The half-life of proteins is highly variable.
- Ornithine decarboxylase has only 11 minutes.
- Half-life of hemoglobin depends on the lifespan of RBCs.
- The lens protein, Crystallin remains unchanged throughout the life of the organism.
- Damaged or defective proteins are prematurely degraded.

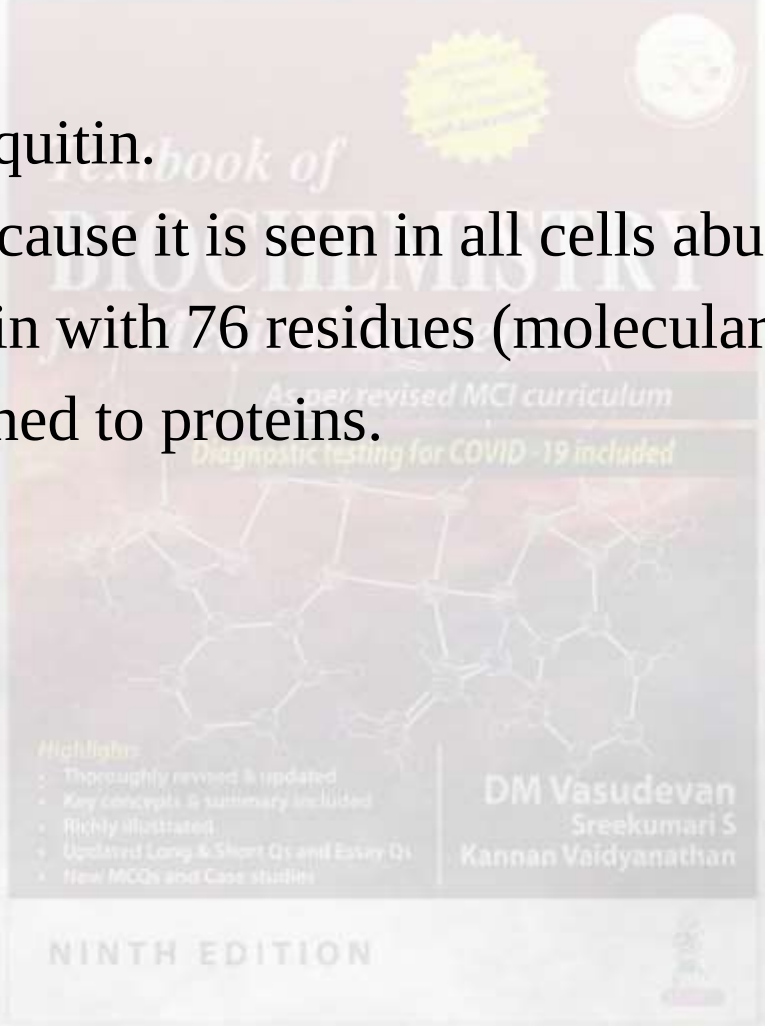
Cathepsins

- In the phagolysosomes, the particles are broken down by enzymes known as cathepsins.
- The term cathepsin is a Greek word, meaning ‘to digest'.
- Cathepsins are 18 in number, designated as A to T.
- Most of them are active at pH around 3 to 5.



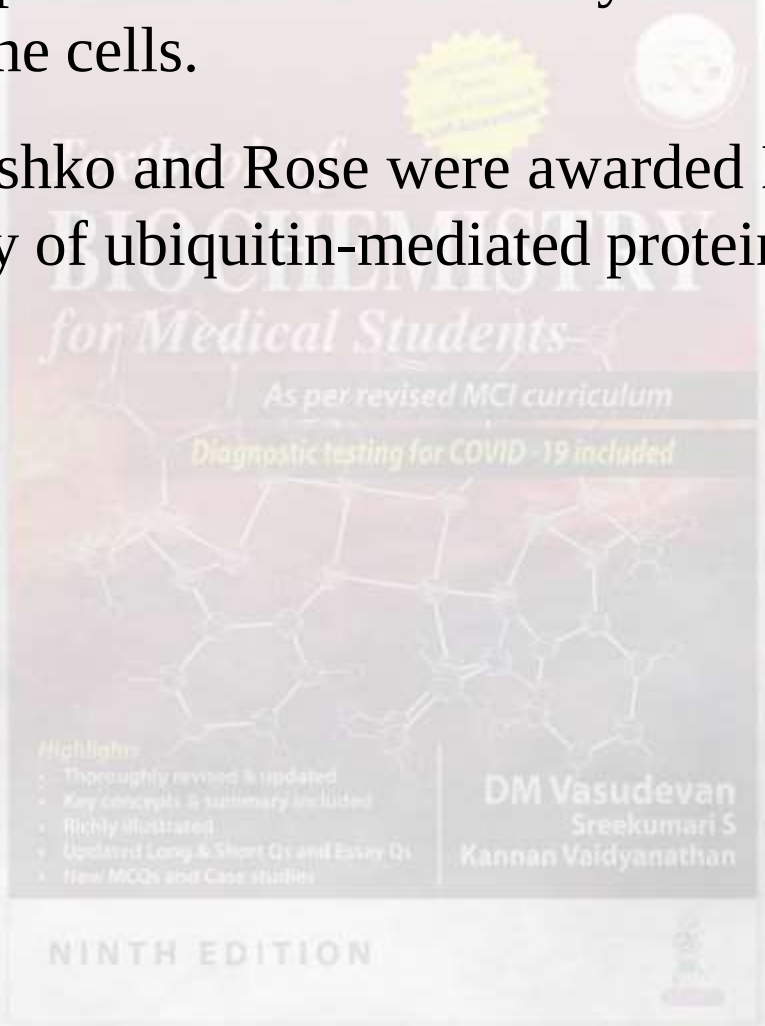
Ubiquitin

- Intracellular protein breakdown also occurs independent of lysosomes.
- This involves ubiquitin.
- It is so named, because it is seen in all cells abundantly.
- It is a small protein with 76 residues (molecular weight, 8.5 kDa).
- Ubiquitin is attached to proteins.

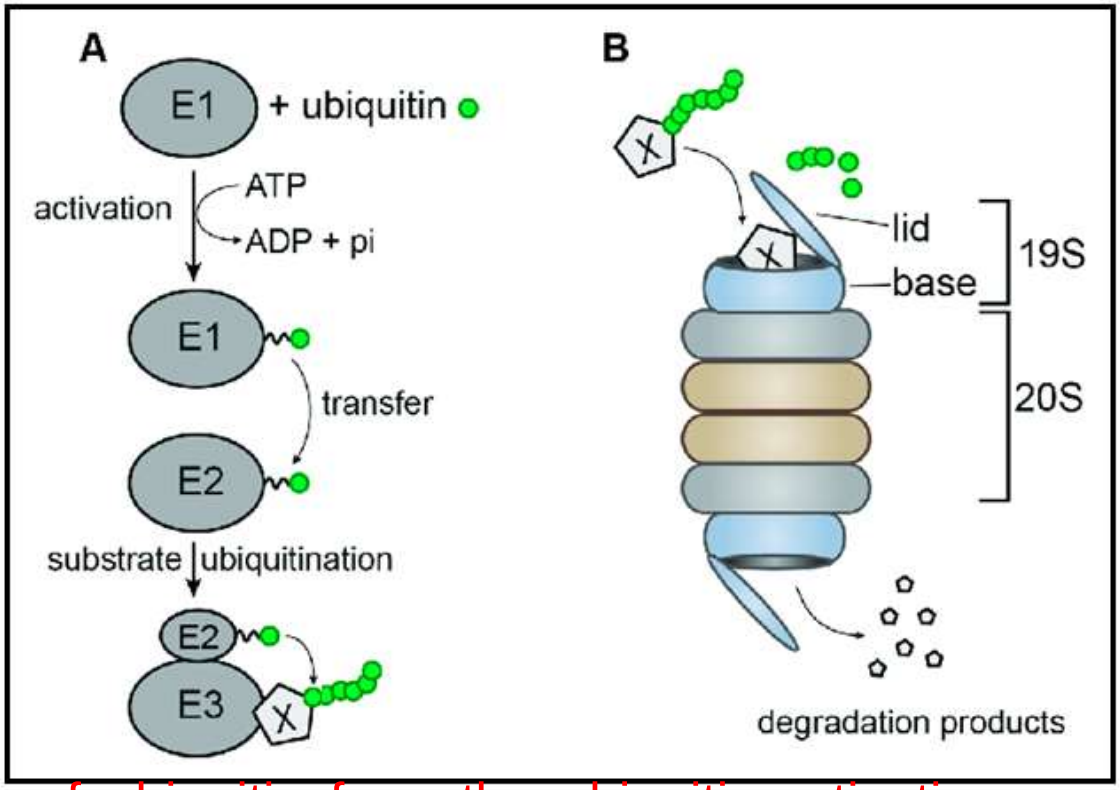


Proteasomes

- Ubiquitin tagged proteins are immediately broken down inside the **proteasomes** of the cells.
- Ciechanover, Hershko and Rose were awarded Nobel Prize in 2004 for their discovery of ubiquitin-mediated protein degradation.



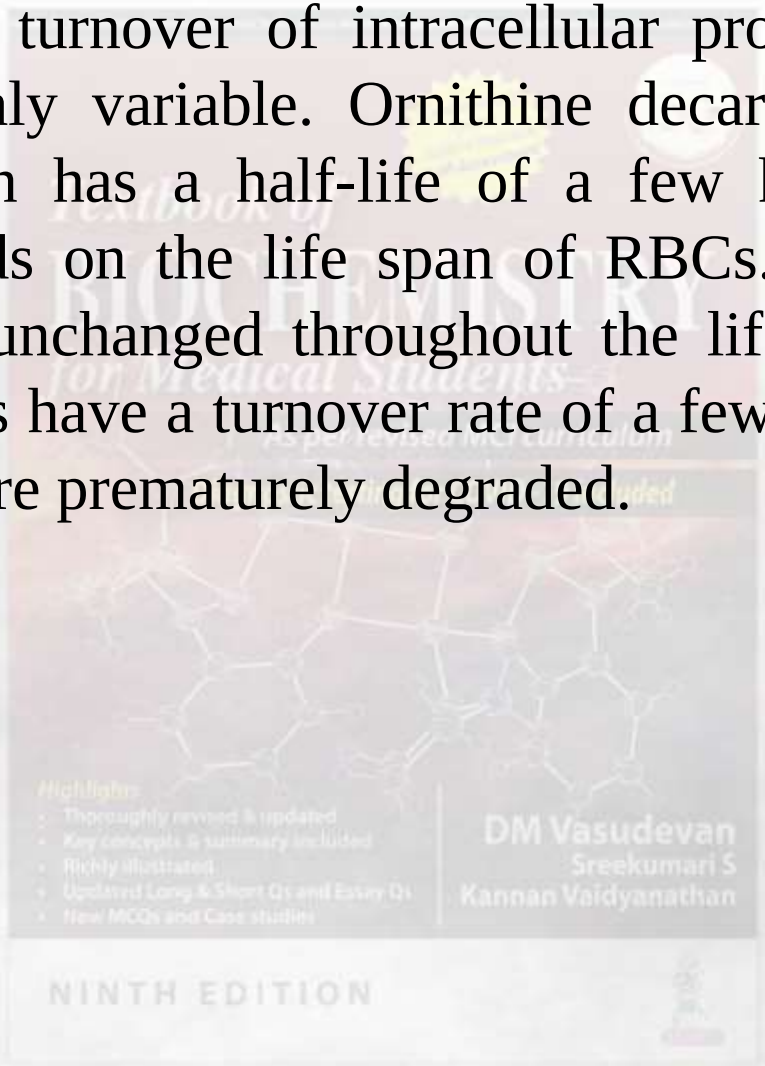
Ubiquitin-Proteasome System

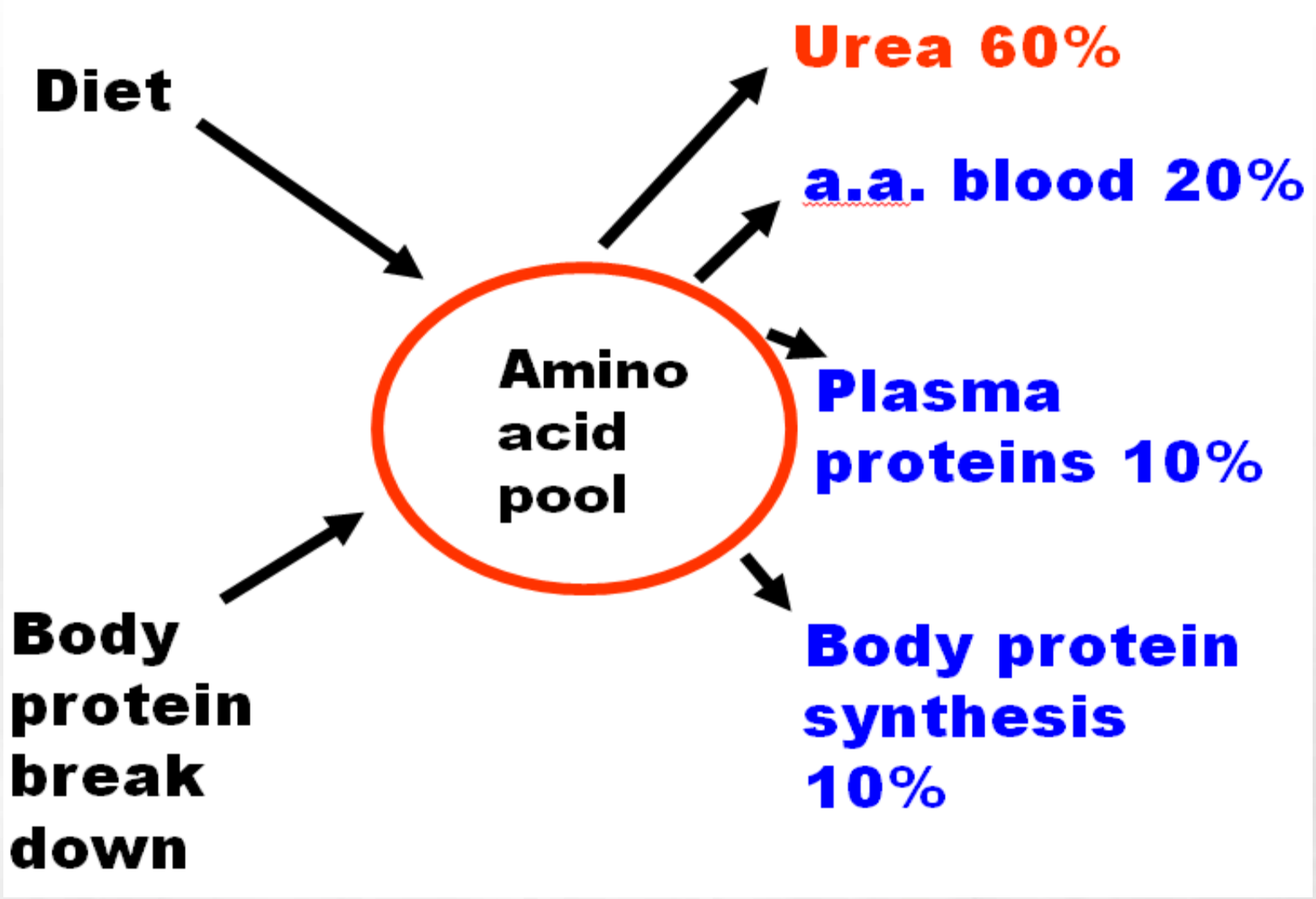


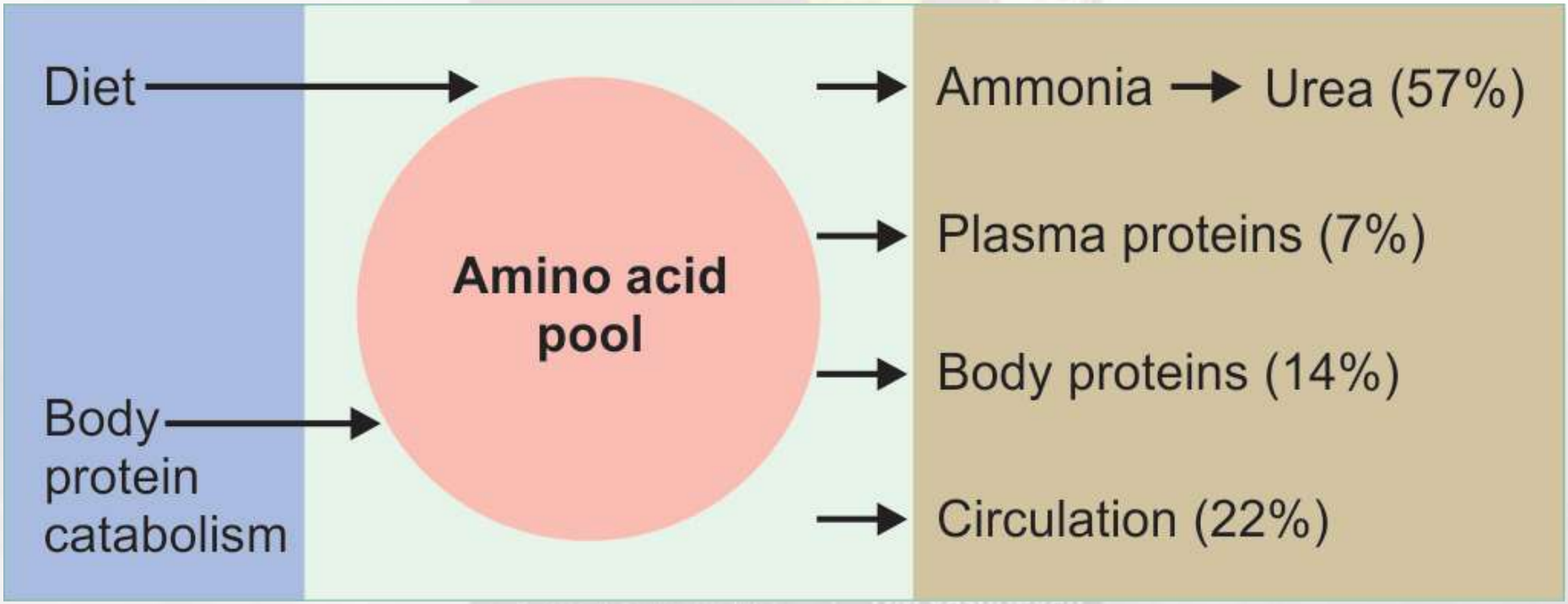
(A) Transfer of ubiquitin from the ubiquitin-activating enzyme E1 to the ubiquitin-conjugating enzyme E2 followed by its transfer onto the target protein X by the ubiquitin ligase E3. (B) The proteasome is composed of one barrel and two lids. The ubiquitinated target protein is taken into the proteasome and degraded products come out. Green dots are UB molecules. X is the protein to be degraded.

Life Span of Proteins

There is a regular turnover of intracellular proteins. The halflife of proteins is highly variable. Ornithine decarboxylase has only 11 minutes. Insulin has a half-life of a few hours. Half-life of hemoglobin depends on the life span of RBCs. The lens protein, crystallin remains unchanged throughout the life of the organism. Majority of proteins have a turnover rate of a few days. Damaged or defective proteins are prematurely degraded.



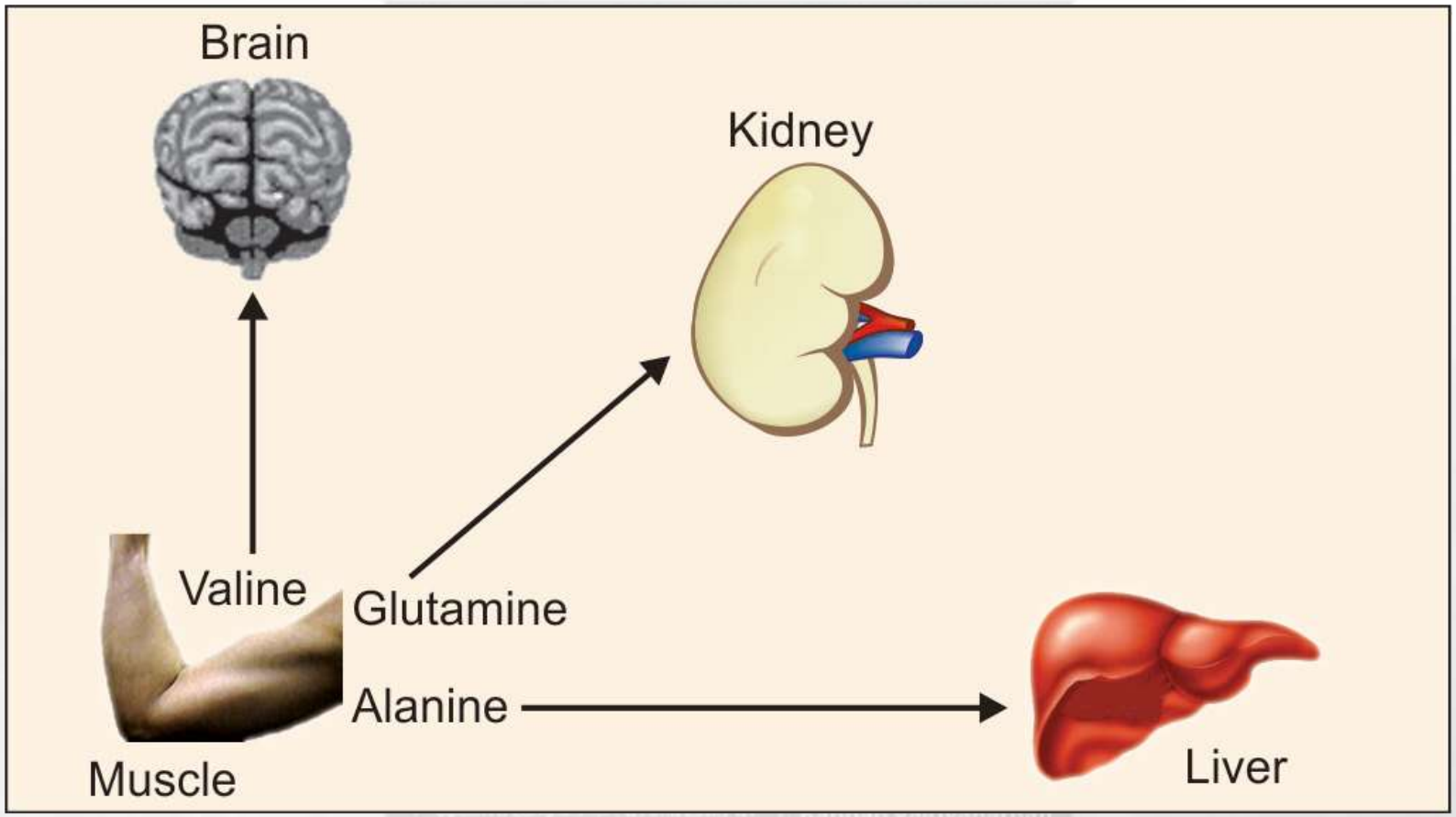

Diet
Body protein break down
Amino acid pool
Urea 60%
a.a. blood 20%
Plasma proteins 10%
Body protein synthesis 10%



Amino acid pool.

Inter-Organ Transport of Amino Acids

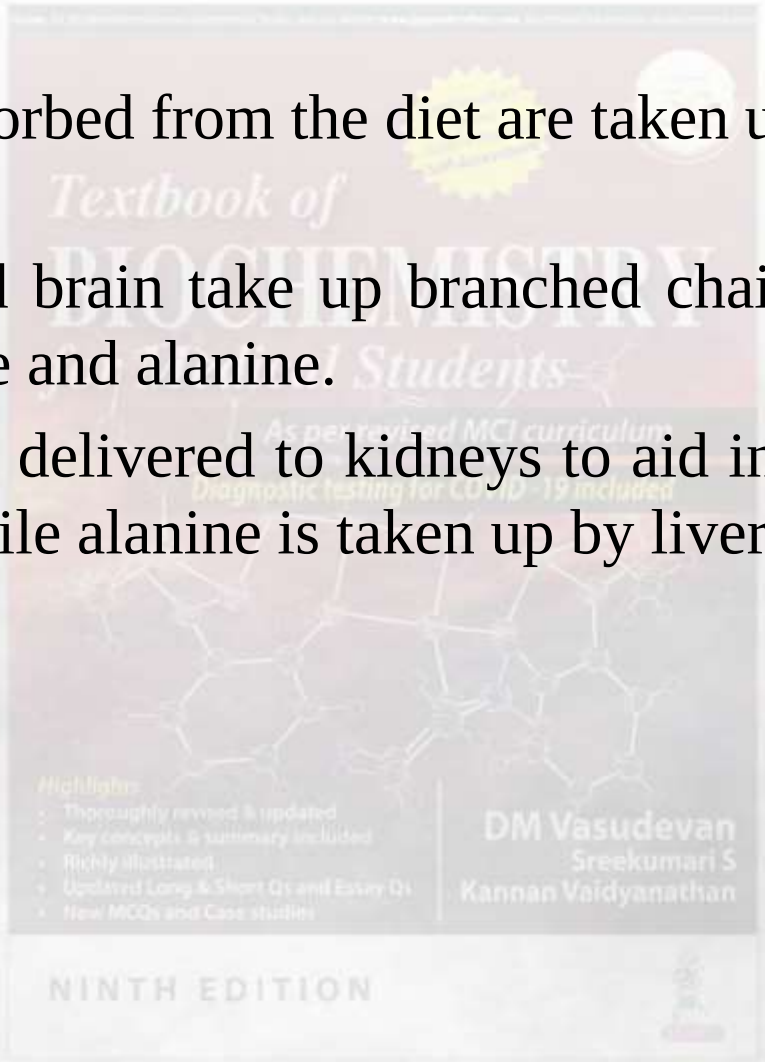
- Breakdown of muscle protein is the source of amino acids for tissues while liver is the site of disposal.
- **In Fasting State**
- The muscle releases mainly alanine and glutamine of which **alanine is taken up by liver** and glutamine by kidneys.
- Liver removes the amino group and converts it to urea and the carbon skeleton is used for **gluconeogenesis**.
- Students should also see glucose-alanine cycle, under gluconeogenesis.
- The brain predominantly takes up branched chain amino acids.

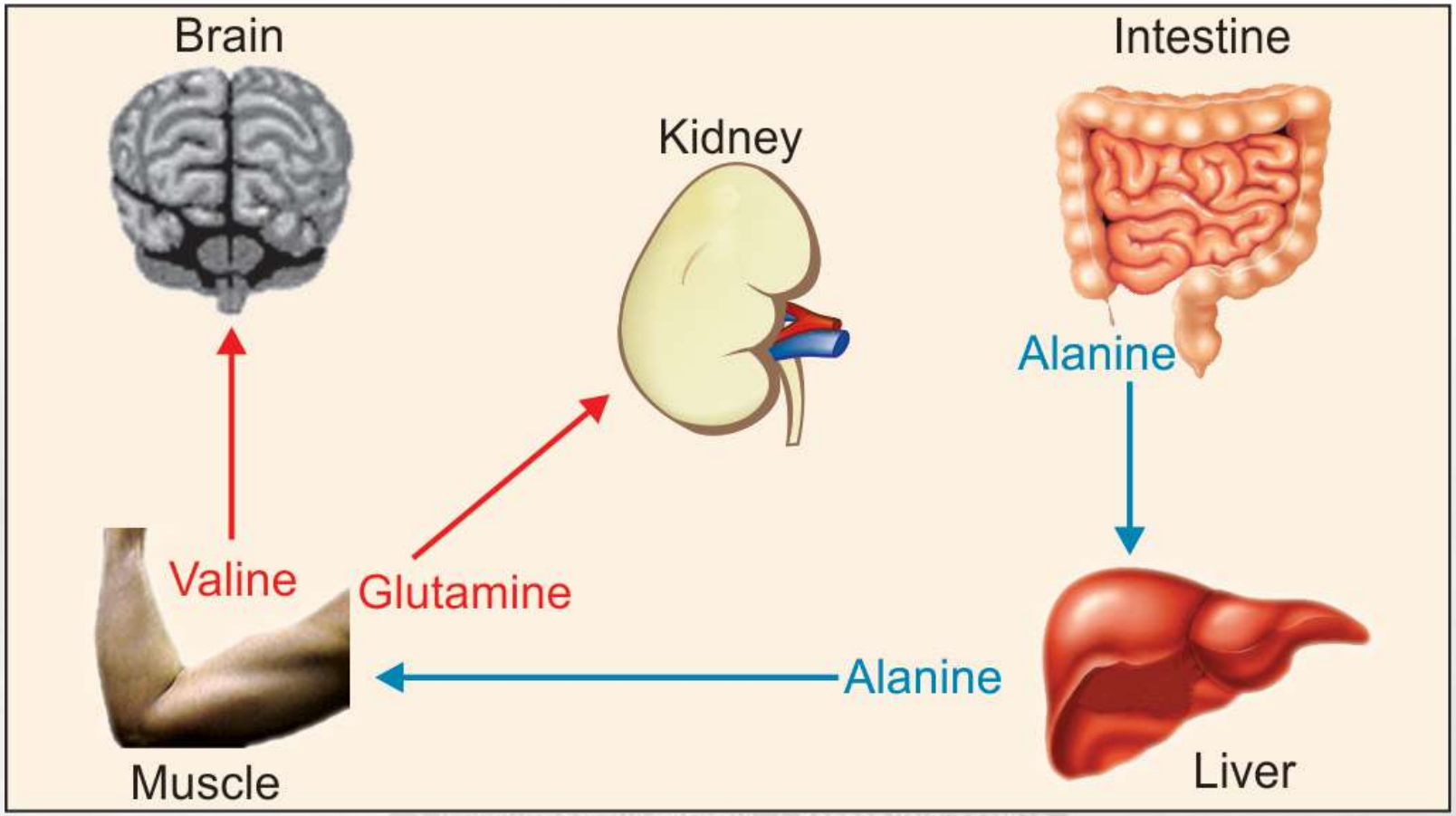


Inter-organ transport of amino acids during fasting conditions

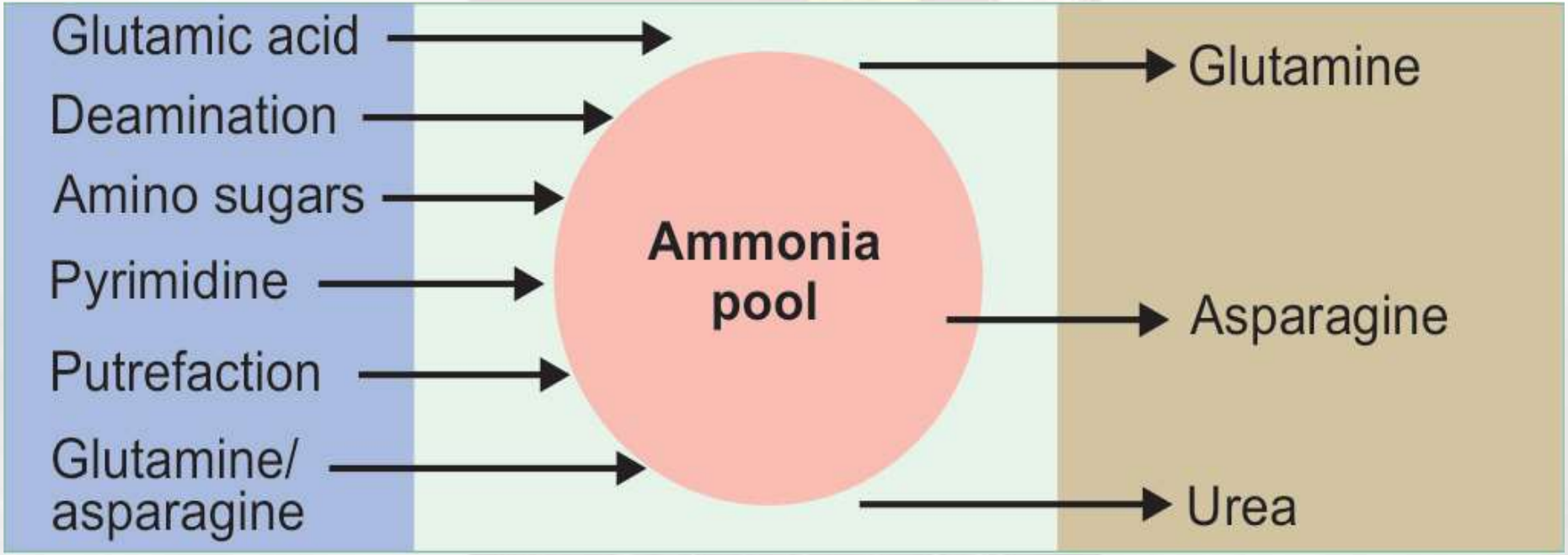
In the Fed State

- Amino acids absorbed from the diet are taken up by different tissues.
- Both muscle and brain take up branched chain amino acids, and release glutamine and alanine.
- The glutamine is delivered to kidneys to aid in regulation of acid-base balance, while alanine is taken up by liver.

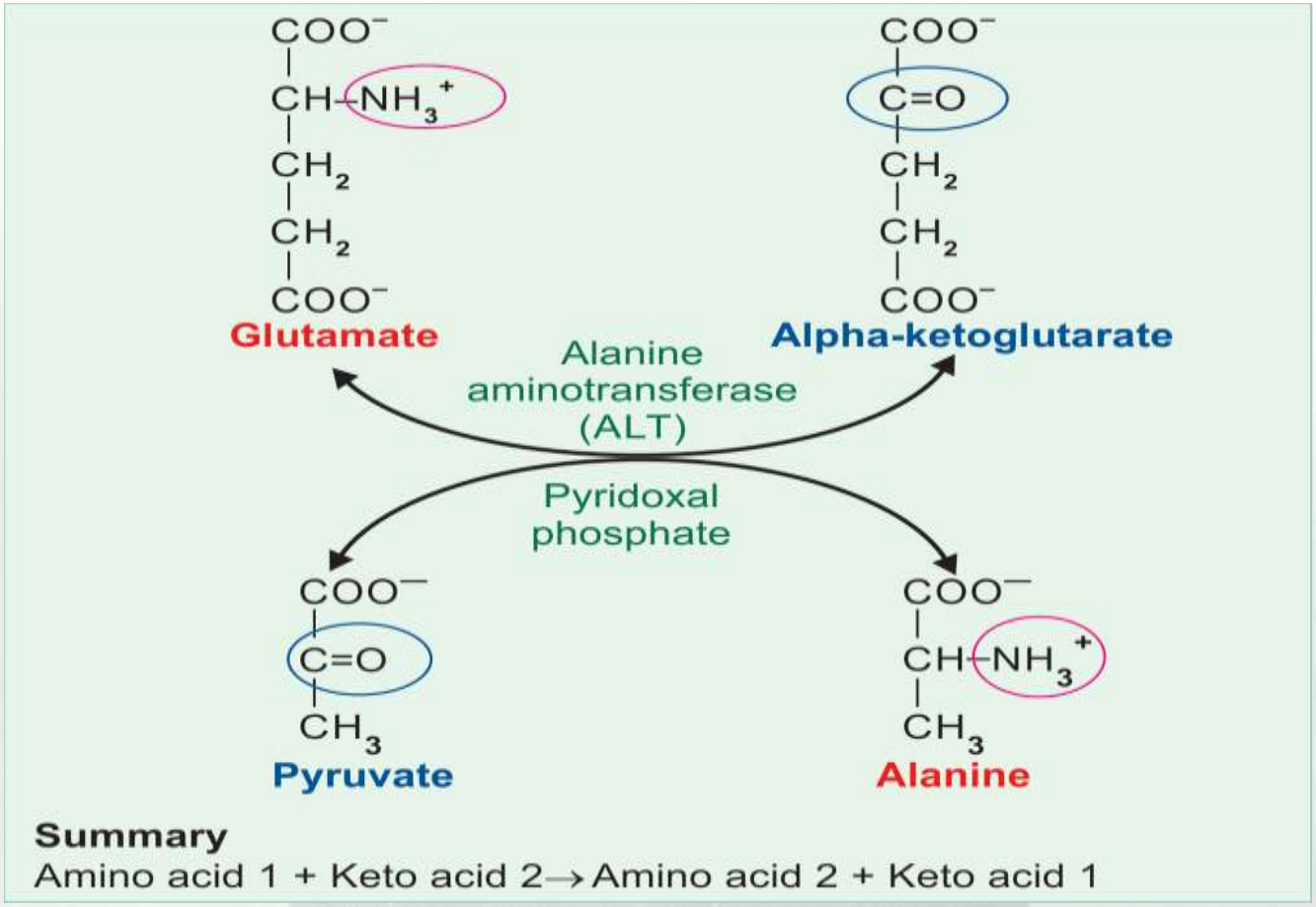




Inter-organ transport of amino acids after taking food (post-prandial condition)



Sources and fate of ammonia



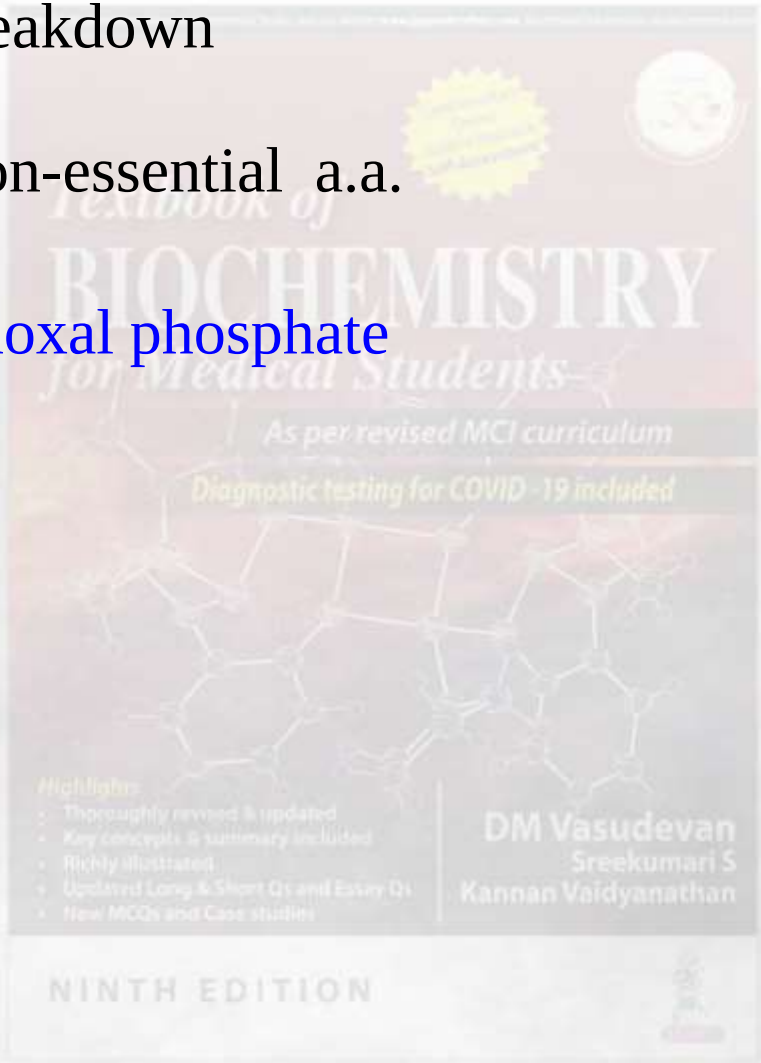
Transamination reaction. In this example, the enzyme is alanine aminotransferase (ALT) and the coenzyme is pyridoxal phosphate. The reaction is readily reversible.

Biological Significance of Transamination Reactions

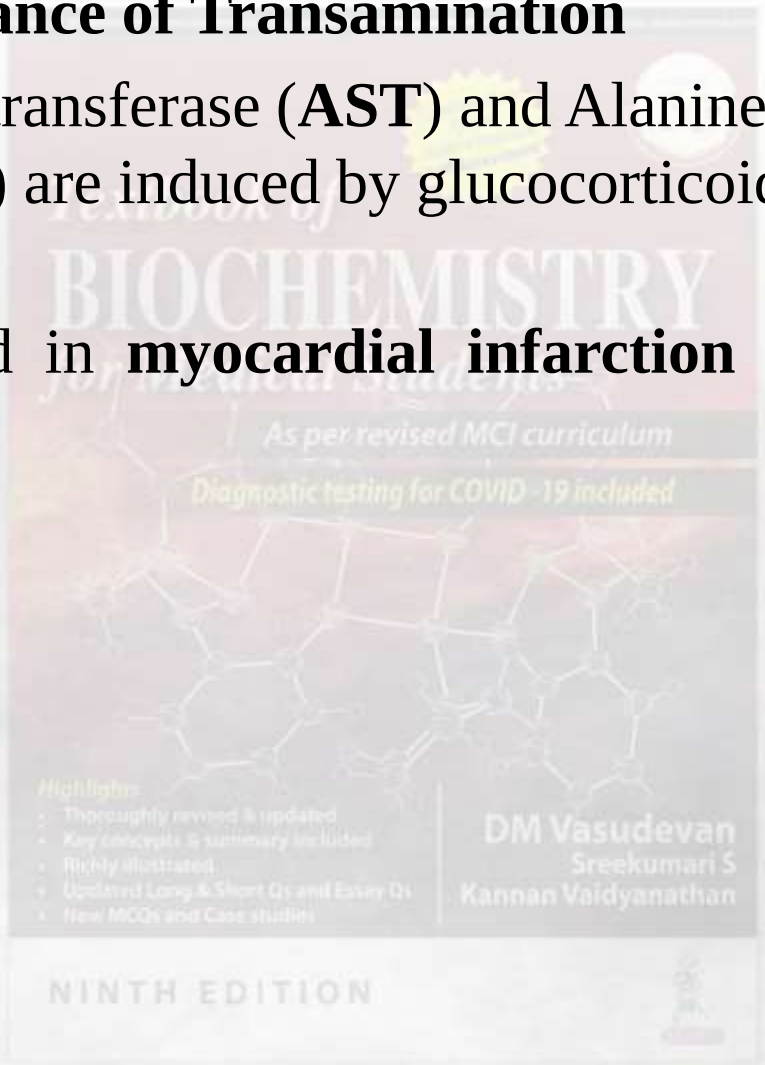
1. Amino acid breakdown

2. Synthesis of non-essential a.a.

Co-enzyme, Pyridoxal phosphate



- **Clinical Significance of Transamination**
- Aspartate amino transferase (**AST**) and Alanine amino transferase (**ALT**) are induced by glucocorticoids, which favor gluconeogenesis.
- AST is increased in **myocardial infarction** and ALT in **liver** diseases.



Trans Deamination

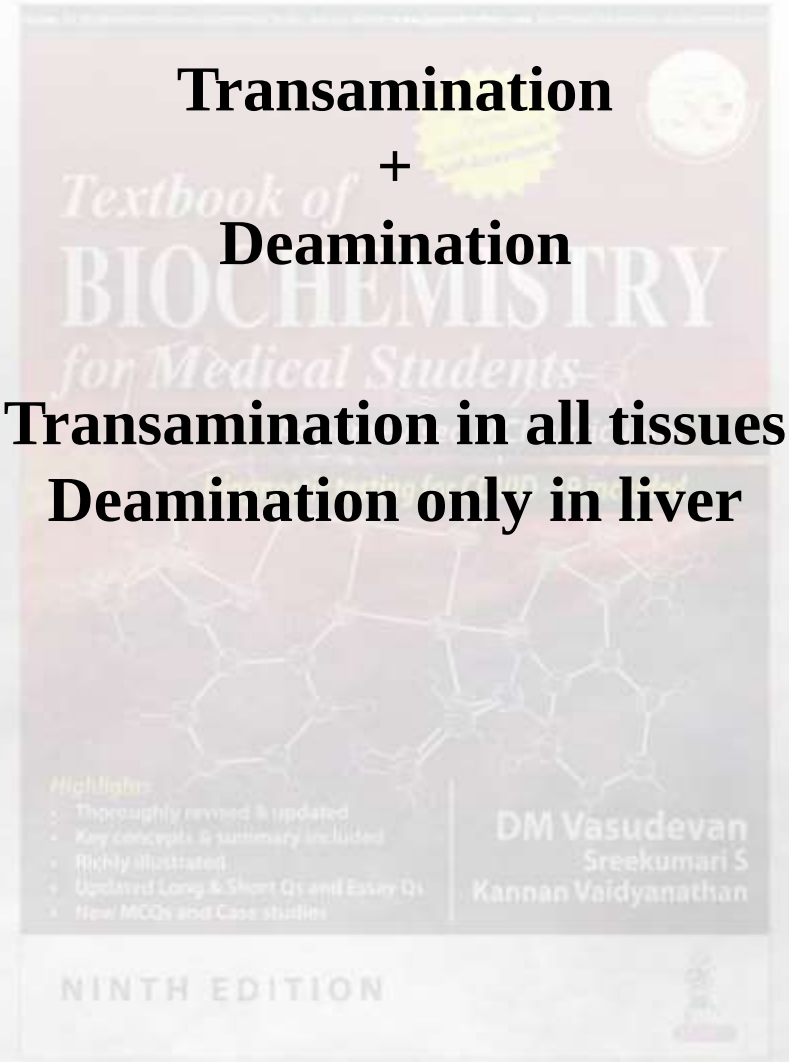
Transamination

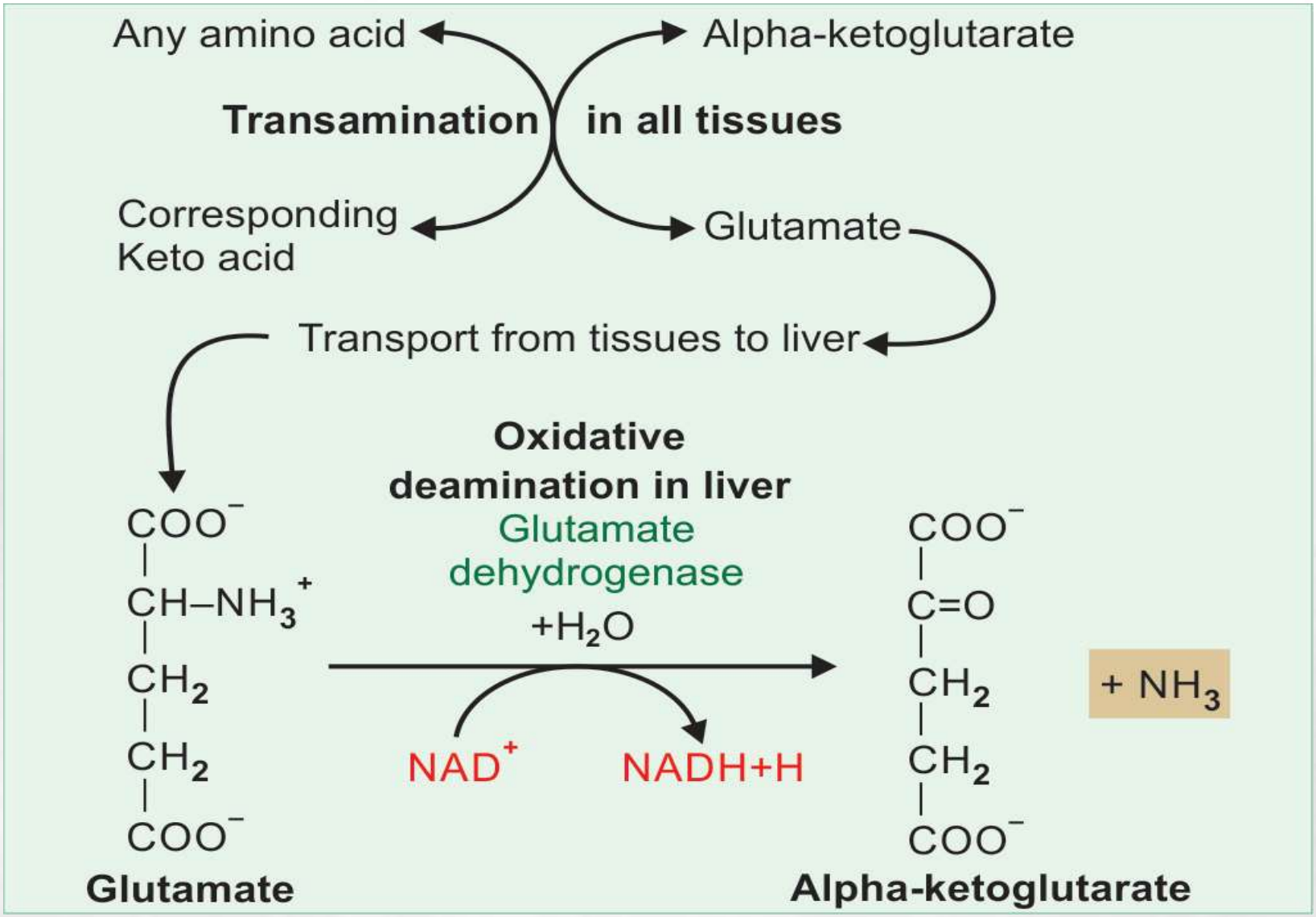
+

Deamination

Transamination in all tissues

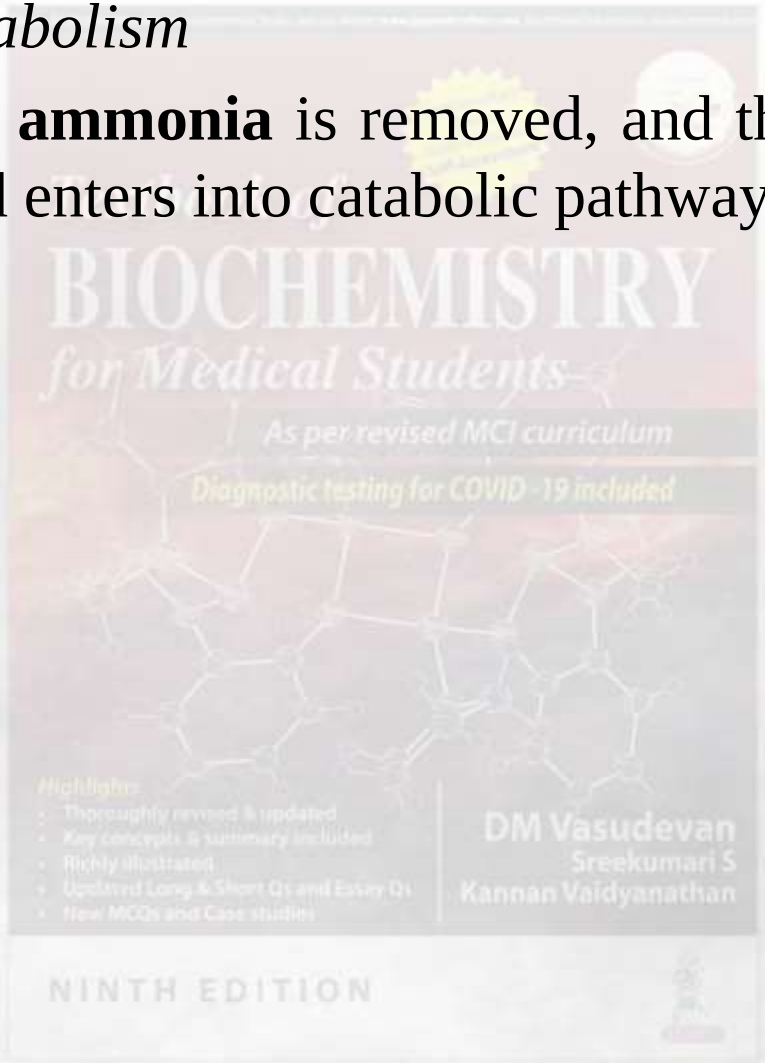
Deamination only in liver




Any amino acid
Alpha-ketoglutarate
Transamination
in all tissues
Corresponding
Keto acid
Glutamate
Transport from tissues to liver
Oxidative
deamination in liver
Glutamate
dehydrogenase
+H2O
NAD+
NADH+H
+ NH3
Glutamate
Alpha-ketoglutarate

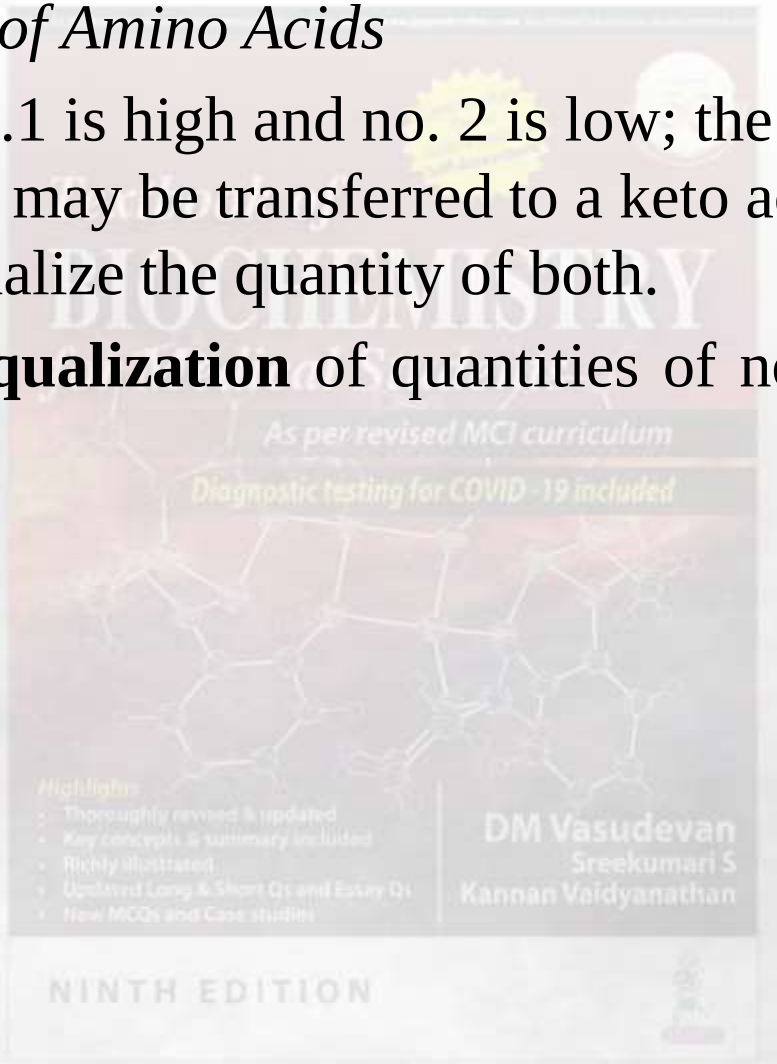
Biological Significance of Transamination

- *First Step of Catabolism*
- In this first step, **ammonia** is removed, and the carbon skeleton of the amino acid enters into catabolic pathway.



- *Synthesis of Non-essential Amino Acids*
- By means of transamination, all nonessential amino acids can be synthesized by the body from keto acids available from other sources.
- For example, **pyruvate** can be transaminated to synthesize **alanine**.
- Similarly oxaloacetate produces aspartic acid.
- Alpha ketoglutarate is transaminated to form glutamic acid.
- Those amino acids, which cannot be synthesized in this manner, are therefore essential; they should be made available in the food.

- *Interconversion of Amino Acids*
- If amino acid no.1 is high and no. 2 is low; the amino group from no.1 may be transferred to a keto acid to give amino acid no. 2 to equalize the quantity of both.
- This is called **equalization** of quantities of non-essential amino acids.

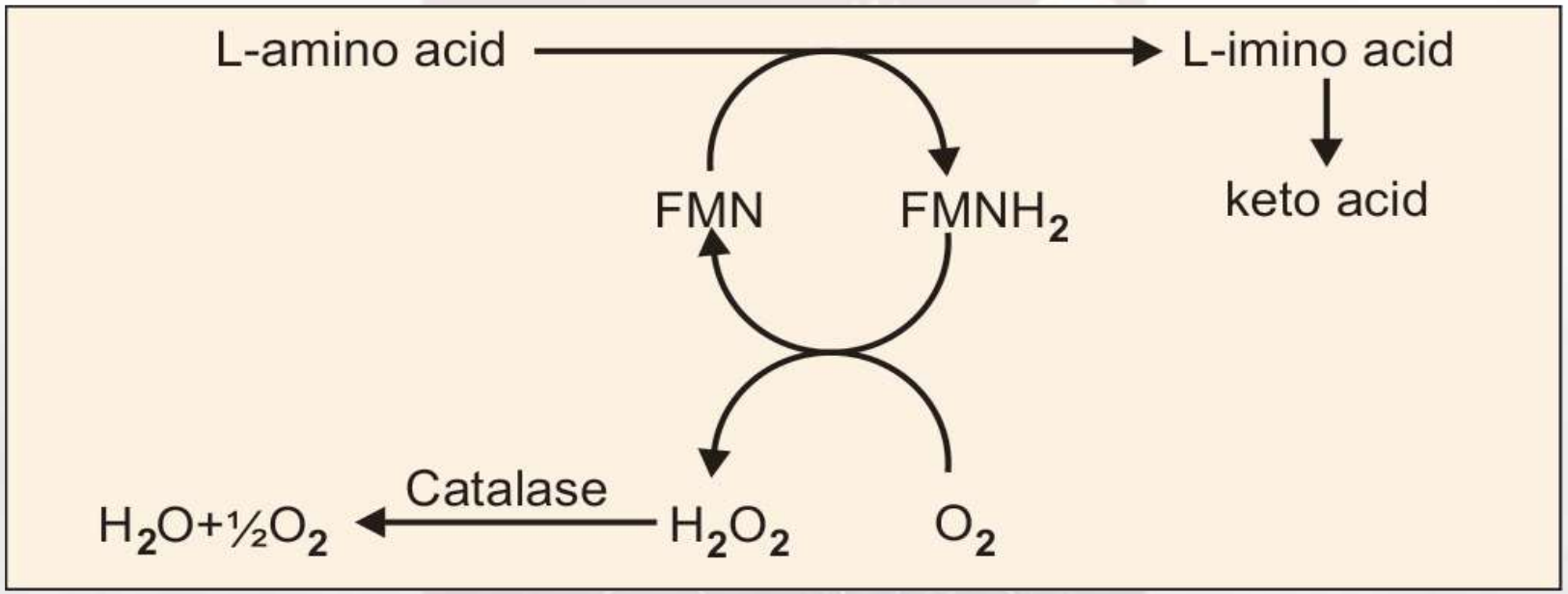


Oxidative Deamination of Glutamate

- Only **liver** mitochondria contain **glutamate dehydrogenase (GDH)** which deaminates glutamate to alphaketoglutarate plus ammonia.
- So, all amino acids are first transaminated to glutamate, which is then finally deaminated **(transdeamination)**. Amino acids are deaminated at the rate of about 50–70 gram per day.
- During the transamination reaction the amino group of all other amino acids is funneled into glutamate.
- Hence, the glutamate dehydrogenase reaction is the final reaction, which removes the amino group of all amino acids. It needs NAD+ as coenzyme.
- It is an allosteric enzyme; it is activated by ADP and inhibited by GTP.
- The hydrolysis of glutamine also yields NH3 but this occurs mainly in the kidney where the NH4+ excretion is required for acid-base regulation.

Minor Pathways of Deamination

1. L-amino acid oxidase can act on all amino acids except hydroxy amino acids and dicarboxylic amino acids. It uses FMN as coenzyme. The peroxide formed in this reaction is decomposed by catalase in the peroxisomes.
2. D-amino acid oxidase can oxidize glycine and any D amino acid that may be formed by bacterial metabolism. It uses FAD as coenzyme.
3. Small quantities of ammonia may be formed in the body through minor reactions like oxidation of monoamines by MAO (mono amine oxidase)



L-amino acid oxidase

Non-Oxidative Deaminations

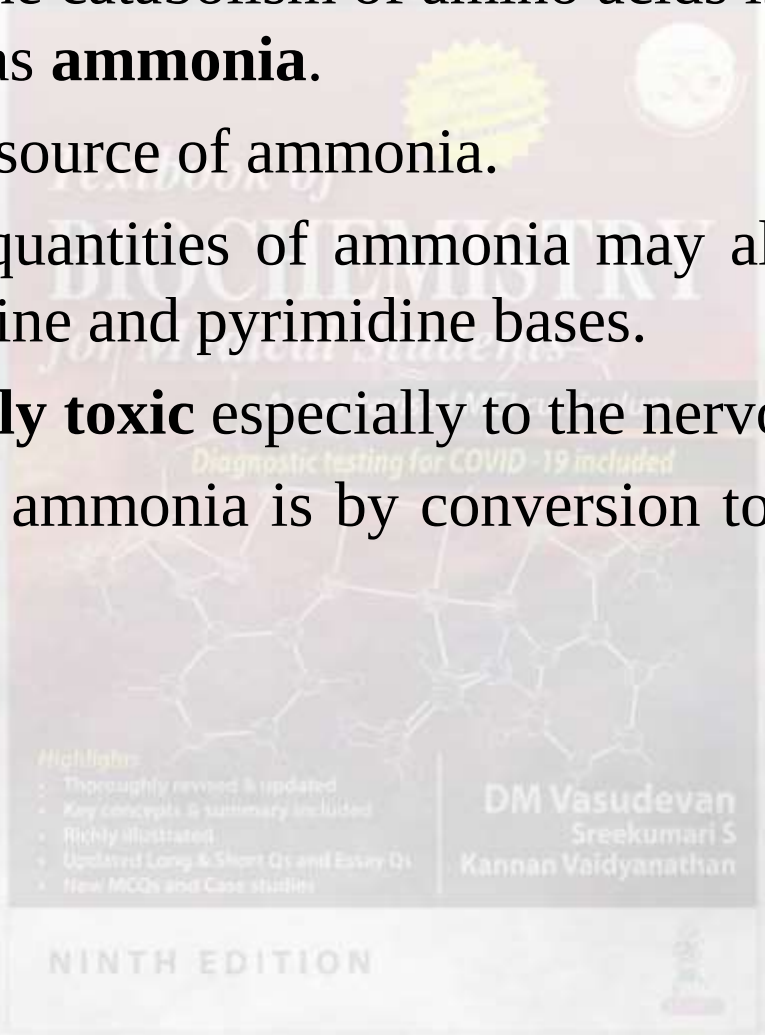
- **Dehydratases** act on hydroxy amino acids to remove ammonia from the following amino acids:
  a. **Serine** will give rise to pyruvate
  b. **Threonine** is converted to alpha keto butyric acid.
- **Desulfhydrase: Cysteine** undergoes deamination and simultaneous transsulfuration to form pyruvate.
- **Histidine** also undergoes non-oxidative deamination to form urocanic acid; catalyzed by histidase.
- Ammonia may also be produced by degradation of purines and pyrimidines due to bacterial putrefaction in the gastrointestinal tract.

General Metabolism of Amino Acids

1. The **anabolic reactions** where proteins are synthesized.
2. **Synthesis** of specialized products such as heme, creatine, purines and pyrimidines.
3. The **catabolic reactions** where dietary proteins and body proteins are broken down to amino acids.
4. **Transamination**: Amino group is removed to produce the carbon skeleton (keto acid). The amino group is excreted as **urea**.
5. The carbon skeleton is used for synthesis of **nonessential** amino acids.
6. It is also used for **gluconeogenesis** or for complete oxidation.
7. Amino acids are used for minor metabolic functions like conjugation, methylation, amidation, etc.

Formation of Ammonia

- The first step in the catabolism of amino acids is to remove the amino group as **ammonia**.
- This is the major source of ammonia.
- However, small quantities of ammonia may also be formed from catabolism of purine and pyrimidine bases.
- Ammonia is **highly toxic** especially to the nervous system.
- Detoxification of ammonia is by conversion to urea and excretion through urine.

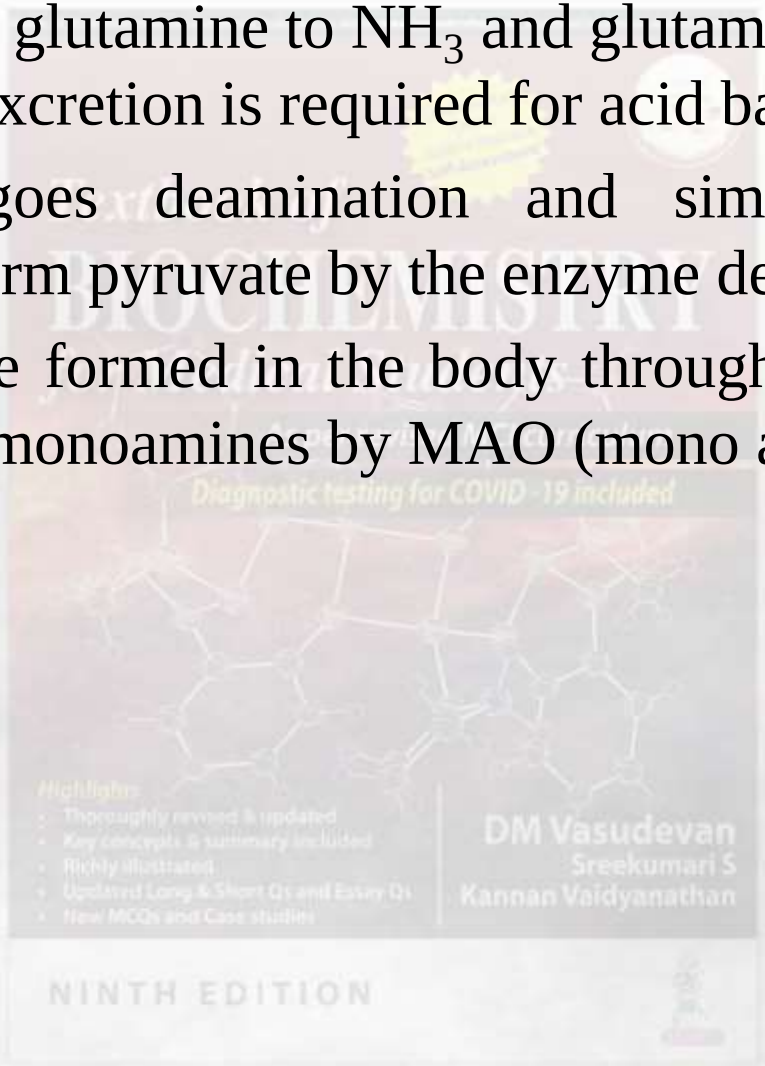


End product of Protein metabolism

Minor Pathways Producing Ammonia

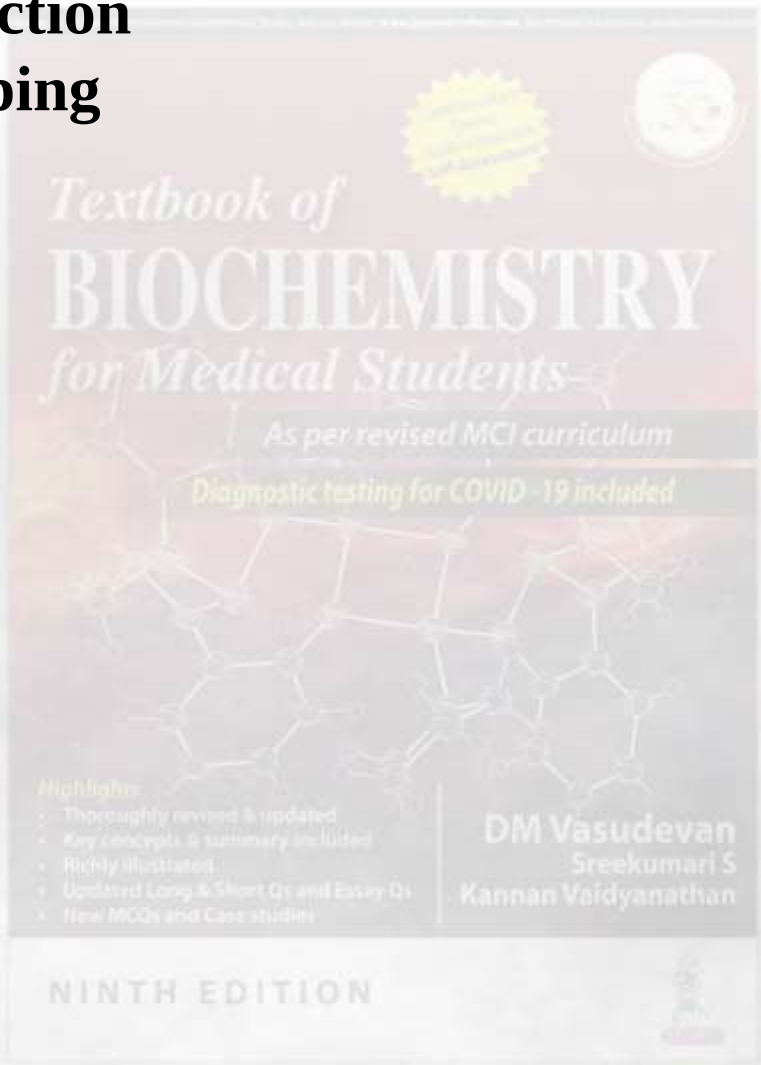
- Histidine to urocanic acid and NH_3 by histidase
- Asparagine to aspartate and NH_3 by asparaginase
- Serine and threonine to pyruvate and alpha ketobutyrate respectively by dehydratases
- Ammonia is also released during the purine nucleotide cycle (adenylosuccinase), purine catabolism (ADA and guanase) and pyrimidine catabolism.
- In the gut NH_3 is produced by bacterial metabolism which reaches the liver through portal vein.
- L amino acid oxidase a flavoprotein also releases ammonia from amino acids.
- Ammonia may also be produced in the gastro-intestinal tract by bacterial putrefaction

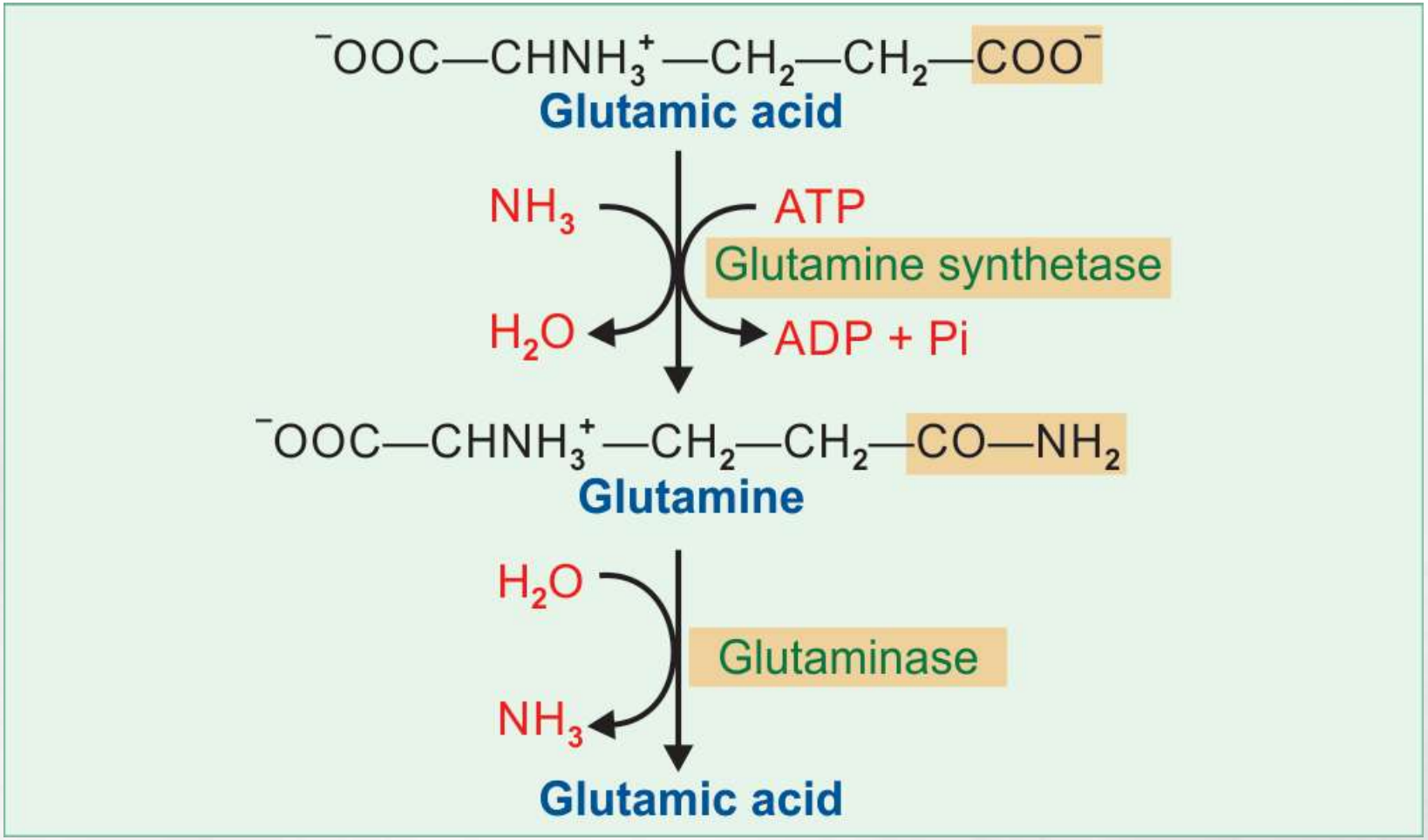
- The hydrolysis of glutamine to NH_3 and glutamate in the kidney where the NH_4^+ excretion is required for acid base regulation.
- **Cysteine** undergoes deamination and simultaneous trans-sulphuration to form pyruvate by the enzyme desulfhydrase.
- Ammonia may be formed in the body through minor reactions like oxidation of monoamines by MAO (mono amine oxidase)



Detoxification of Ammonia

1. **Reduced production**
2. **Ammonia trapping**
3. **Urea synthesis**

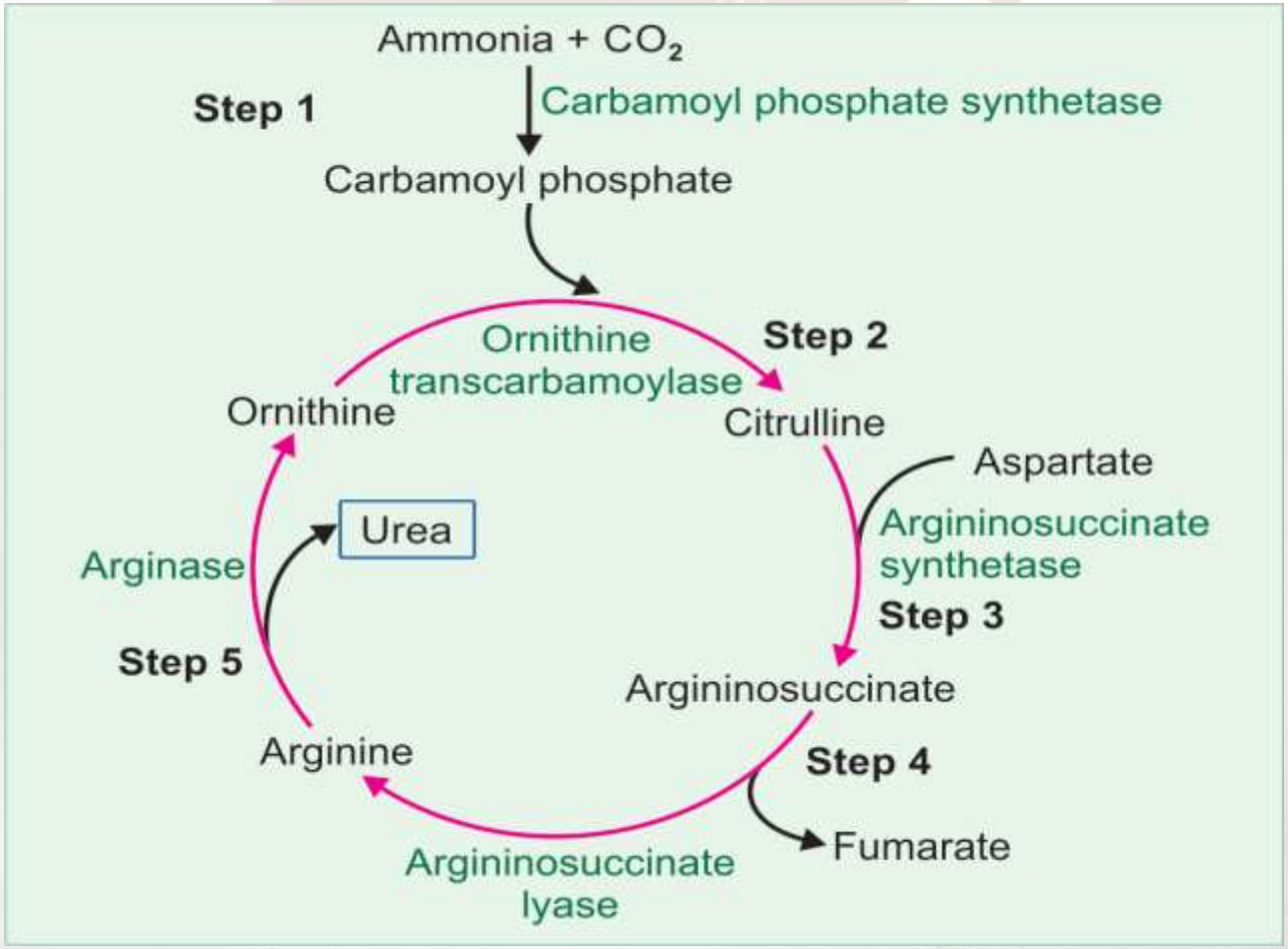




Ammonia trapping as glutamine.

Urea Cycle Ornithine Cycle Krebs-Henseleit Cycle

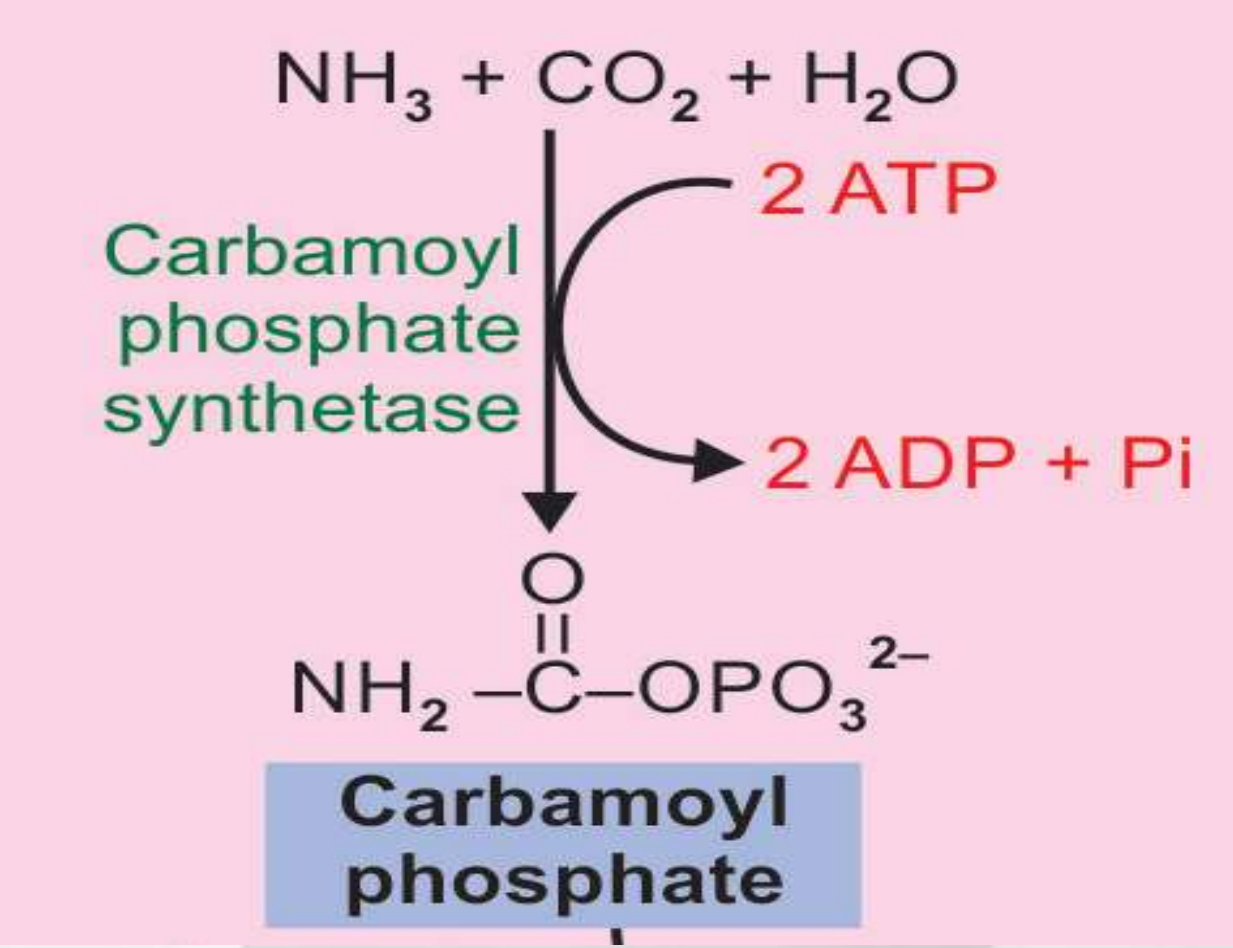
End product of Protein metabolism



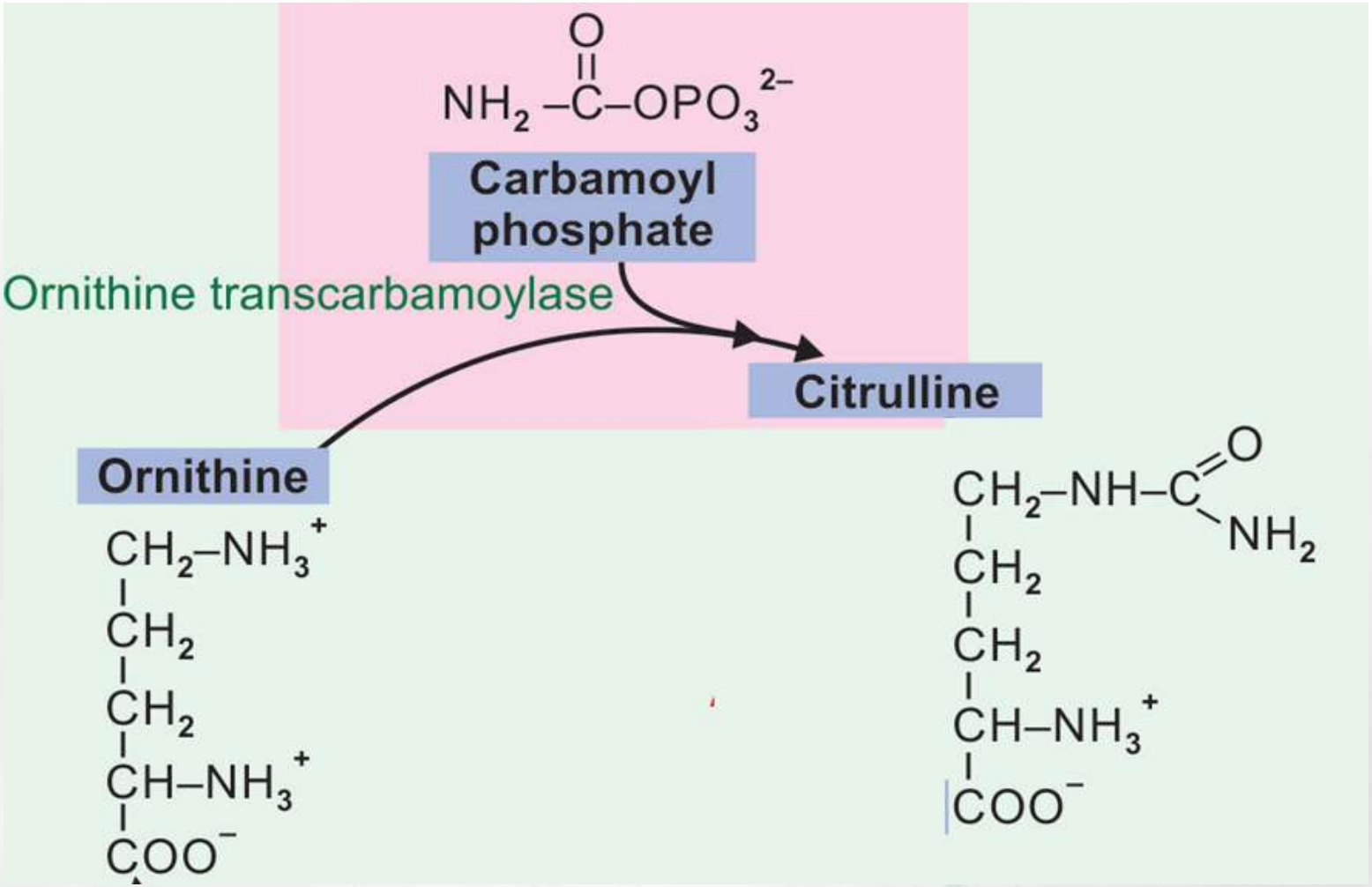
Summary of urea cycle

Comparison of CPS I and II enzymes		
	CPS-I	**CPS-II**
1. Site	Mitochondria	Cytosol
2. Pathway of	Urea	Pyrimidine
3. Positive effector	NAG	Nil
4. Source for N	Ammonia	Glutamine
5. Inhibitor	Nil	CTP

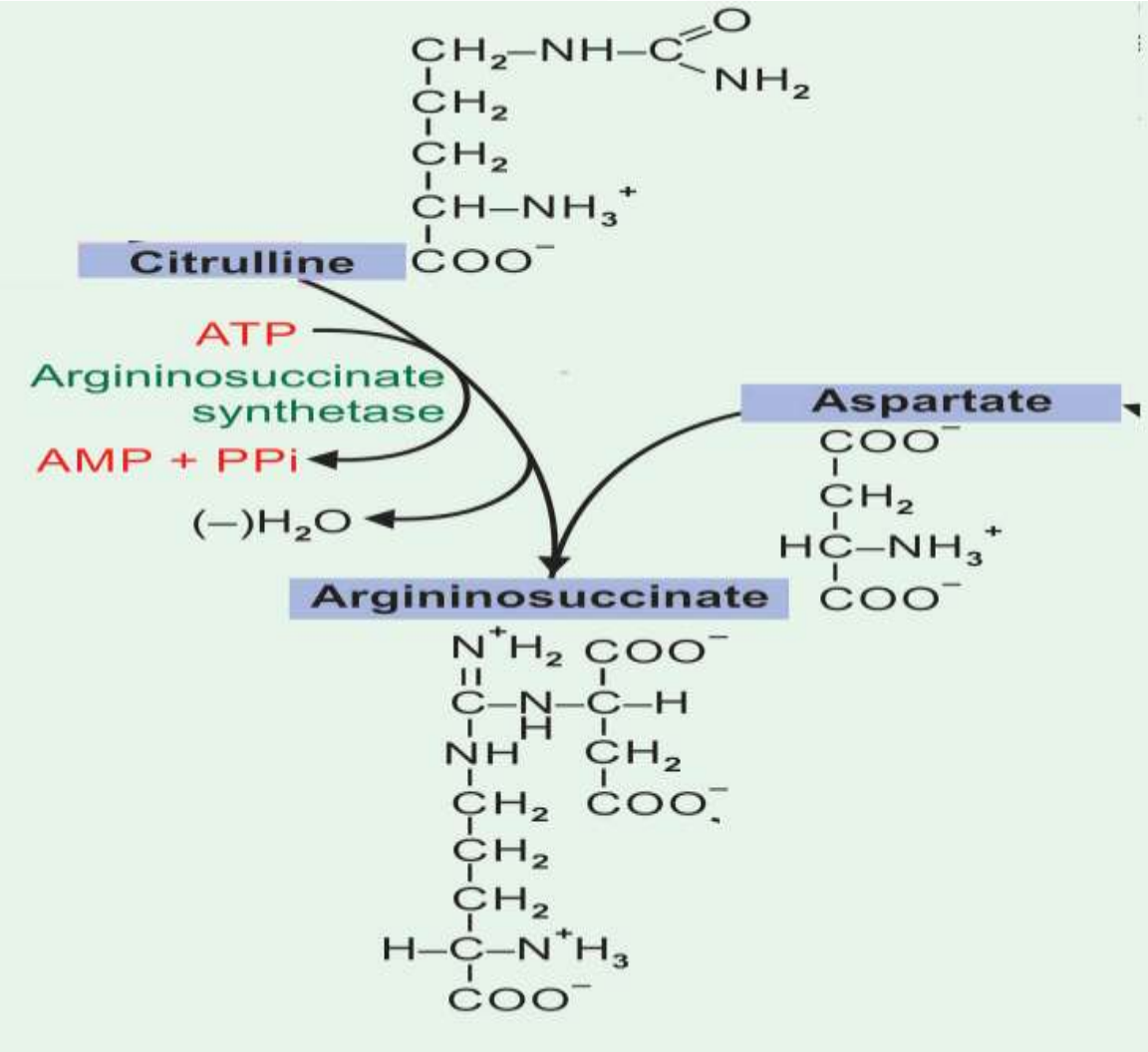
Urea Cycle



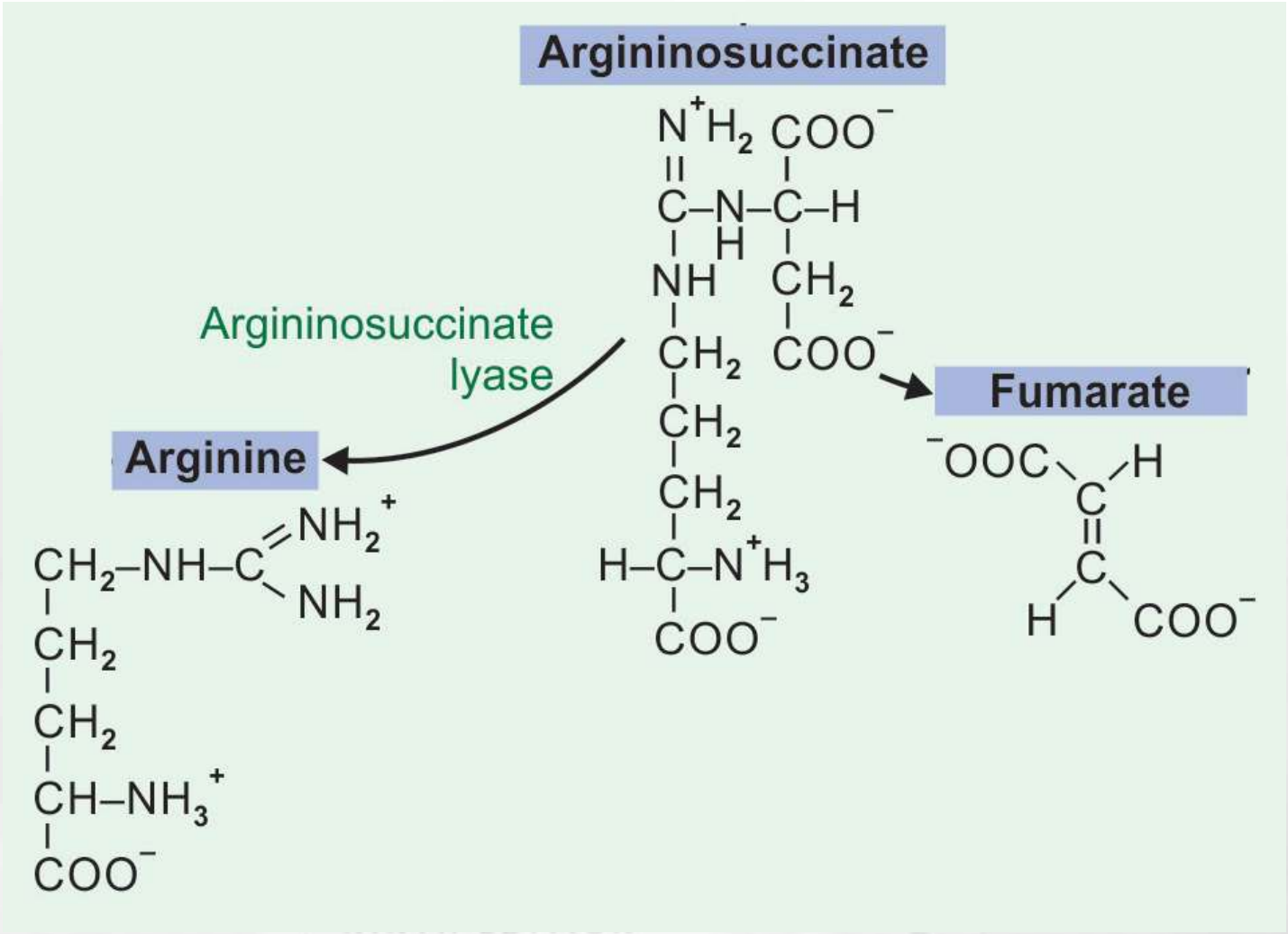
Step 1


Carbamoyl phosphate
Ornithine transcarbamoylase
Citrulline
Ornithine

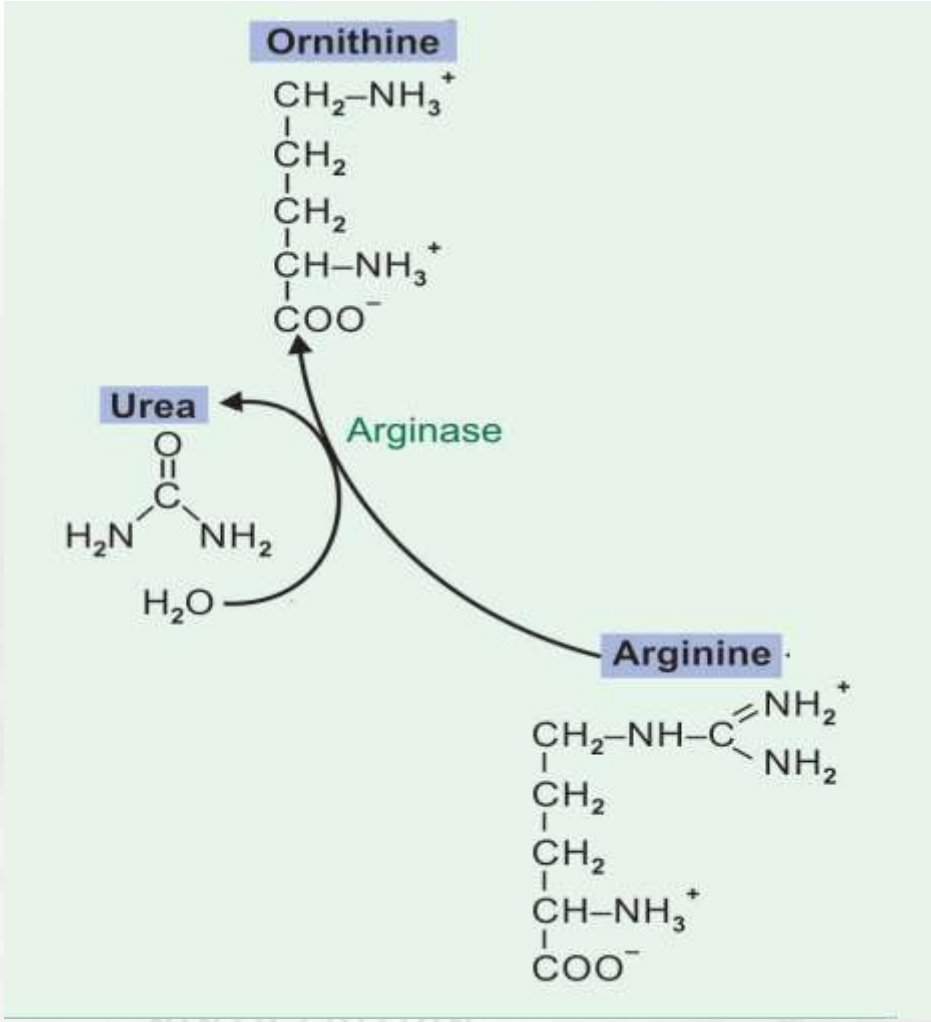
Step 2


Citrulline
ATP
Argininosuccinate synthetase
AMP + PPi
(–)H2O
Aspartate
Argininosuccinate

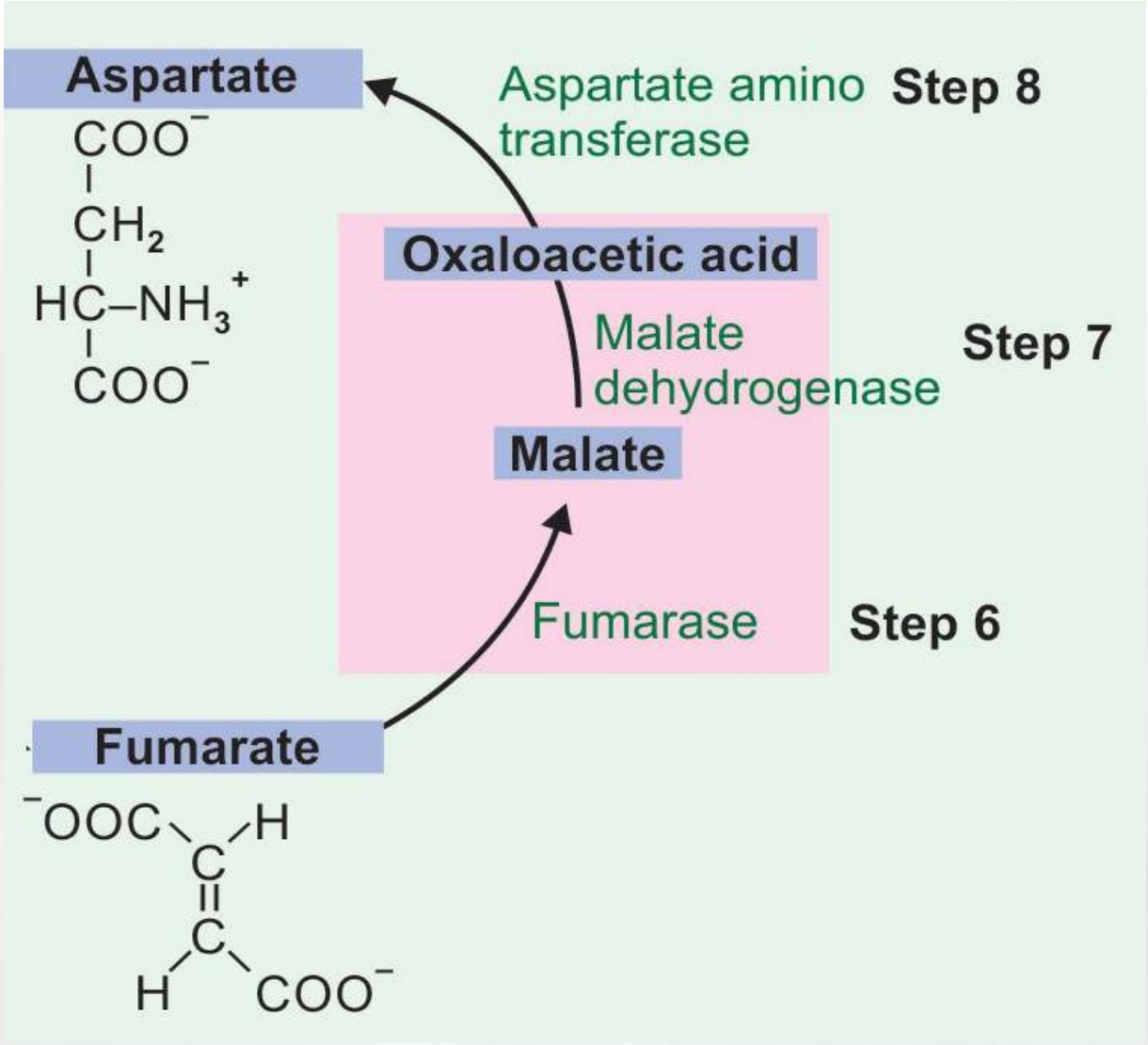
Step 3

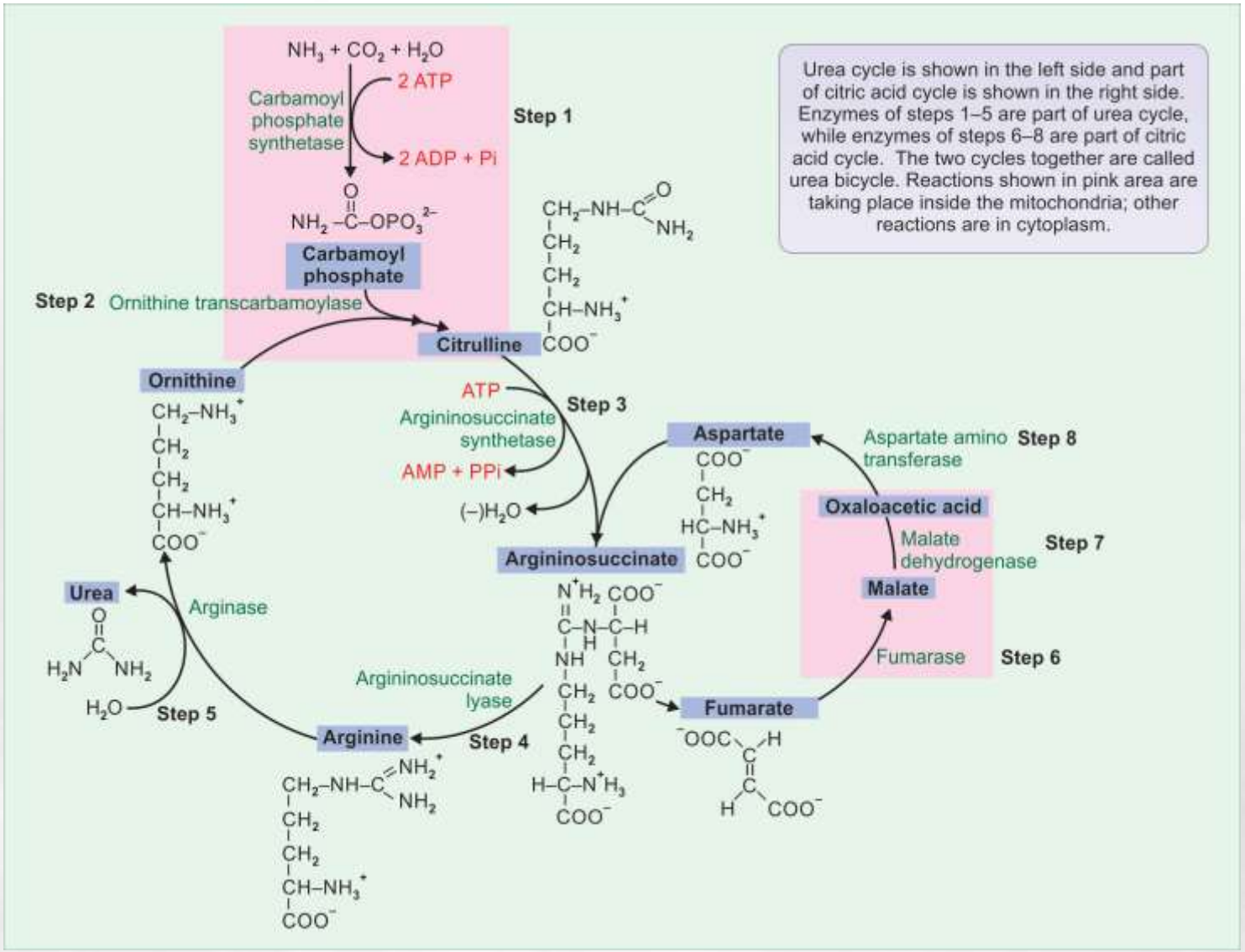

Argininosuccinate
Argininosuccinate lyase
Arginine
Fumarate

Step 4


Ornithine
CH2–NH3+
CH2
CH2
CH–NH3+
COO−
Urea
O
C
H2N
NH2
H2O
Arginase
Arginine
NH2+
CH2–NH–C
NH2
CH2
CH2
CH–NH3+
COO−

Step 5


Aspartate
COO⁻
CH2
HC–NH3+
COO⁻
Aspartate amino transferase
Step 8
Oxaloacetic acid
Malate dehydrogenase
Step 7
Malate
Fumarase
Step 6
Fumarate
⁻OOC
H
C
C
H
COO⁻


Urea cycle is shown in the left side and part of citric acid cycle is shown in the right side. Enzymes of steps 1–5 are part of urea cycle, while enzymes of steps 6–8 are part of citric acid cycle. The two cycles together are called urea bicycle. Reactions shown in pink area are taking place inside the mitochondria; other reactions are in cytoplasm.
NH3 + CO2 + H2O
2 ATP
Carbamoyl phosphate synthetase
2 ADP + Pi
Step 1
Carbamoyl phosphate
Step 2 Ornithine transcarbamoylase
Citrulline
Ornithine
ATP
Argininosuccinate synthetase
AMP + PPi
(–)H2O
Step 3
Aspartate
Aspartate amino transferase
Step 8
Oxaloacetic acid
Malate dehydrogenase
Step 7
Malate
Fumarase
Step 6
Argininosuccinate
Argininosuccinate lyase
Fumarate
Step 4
Arginine
Arginase
Urea
H2O
Step 5

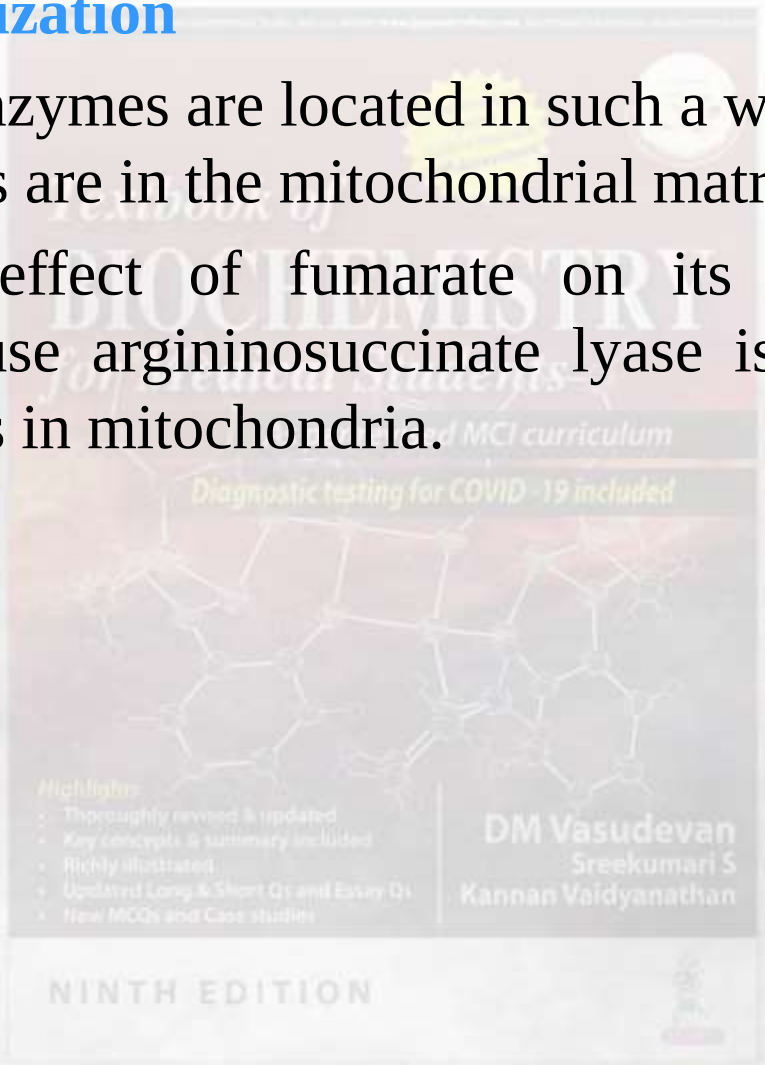
Energetics of Urea Cycle

- The overall reaction may be summarized as:
- $NH_3 + CO_2 + Aspartate \rightarrow Urea + Fumarate$
- In the urea cycle 2 ATPs are used in the first reaction.
- Another ATP is converted to AMP and PPi,which is equivalent to 2 ATPs.
- The urea cycle consumes **4 high energy phosphate bonds.**
- However, fumarate formed in the 4th step may be converted to malate.
- Malate when oxidized to oxaloacetate produces 1 NADH equivalent to 2.5 ATP.
- So, **net energy expenditure is only 1.5 high energy phosphates**.
- The urea cycle and TCA cycle are interlinked, and so, it is called as "**urea bicycle**".

Regulation of the Urea Cycle

- *Coarse Regulation*
- During starvation, the activity of urea cycle enzymes is elevated to meet the increased rate of protein catabolism.
- *Fine Regulation*
- The major regulatory enzyme is CPS-I. **N-acetyl glutamate** (NAG) will stimulate this reaction.
- It is formed from glutamate and acetyl CoA.
- Arginine is an activator of NAG synthase.

- **Compartmentalization**
- The urea cycle enzymes are located in such a way that the first two enzymes are in the mitochondrial matrix.
- The inhibitory effect of fumarate on its own formation is minimized because argininosuccinate lyase is in the cytoplasm, while fumarase is in mitochondria.



Disorders of Urea Cycle

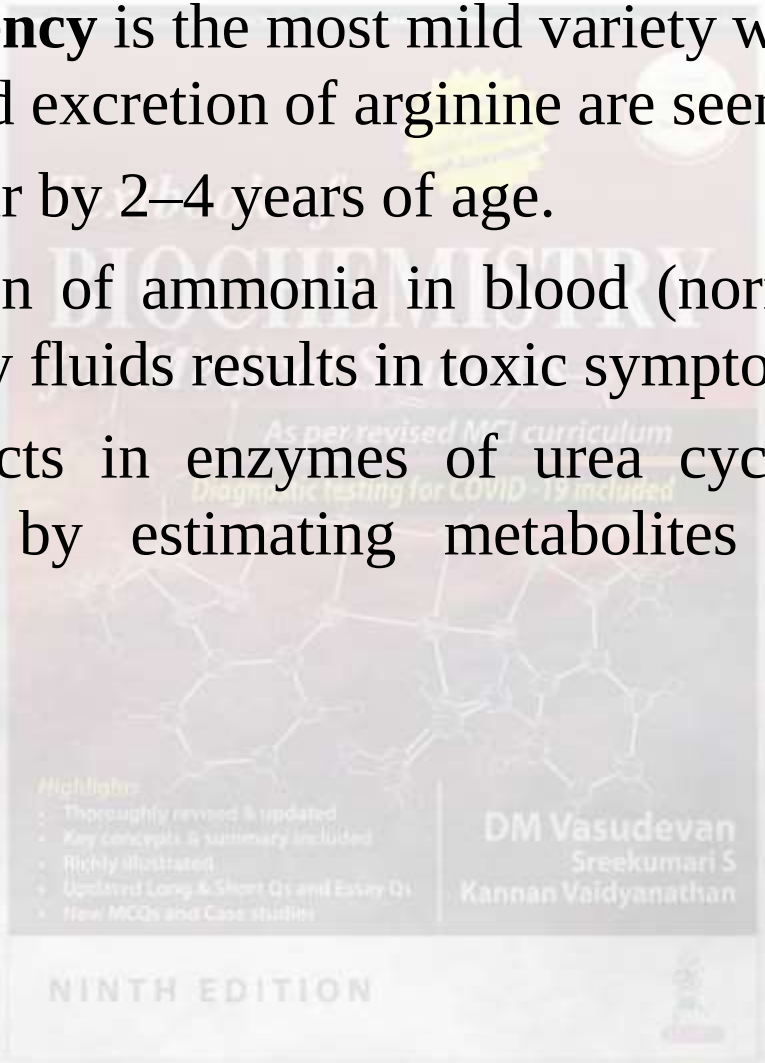
- Deficiency of any of the urea cycle enzymes would result in **hyperammonemia**.
- When the block is in one of the earlier steps, the condition is more severe, since ammonia itself accumulates.
- Deficiencies of later enzymes result in the accumulation of other intermediates, which are less toxic and hence symptoms are less.
- As a general description, disorders of urea cycle are characterized by hyperammonemia, encephalopathy and respiratory alkalosis.
- Clinical symptoms include vomiting, irritability, lethargy and severe mental retardation.
- Infants appear normal at birth, but within days progressive lethargy sets in.

- Treatment is mainly symptomatic.
- Low protein diet with sufficient arginine and energy by frequent feeding can minimize brain damage since ammonia levels do not increase very high.
- Ornithine transporter deficiency is characterized by hyperornithinemia, hyperammonemia and homocitrullinuria (HHH syndrome).
- Since ornithine is not available in the mitochondria, lysine is carbamylated to form homocitrulline.

- **Brain is very sensitive to ammonia.**
- Child may be put on a low protein diet and frequent small feeds are given.
- Attempts may be made to eliminate the amino nitrogen in other forms, e.g. as hippuric acid (Benzoyl conjugate of glycine) or phenyl acetyl glutamine.
- Since **Citrulline** is present in significant quantities in milk, breast milk is to be avoided in citrullinemia.

- Ornithine transcarbamoylase deficiency is the only urea cycle disorder, which is inherited as an X-linked trait.
- Elevated levels of ammonia are associated with high glutamine levels in CSF and blood.
- **Argininosuccinate lyase deficiency** leads to argininosuccinic acidemia and therefore metabolic acidosis.
- Hyperammonemia is less severe and argininosuccinate is elevated in CSF and excreted in urine.
- A typical clinical feature is friable tufted hair (trichorrhexis nodosa).

- **Arginase deficiency** is the most mild variety with accumulation and excretion of arginine are seen.
- Symptoms appear by 2–4 years of age.
- The accumulation of ammonia in blood (normally less than 50 mcg/dl) and body fluids results in toxic symptoms.
- Nowadays, defects in enzymes of urea cycle are detected in neonatal blood by estimating metabolites by tandem mass spectrometry.



Urea cycle disorders

Diseases	Enzyme deficit	Features
Hyperammonemia type I	CPS-I	Very high NH3 levels in blood. Autosomal recessive. Mental retardation. Incidence is 1 in 100,000.
Hyperammonemia type II	(OTC) Ornithine transcar-bamoy-lase	Ammonia level high in blood. Increased glutamine in blood, CSF and urine. Orotic aciduria due to channelling of carbamoyl phosphate into Pyrimidine synthesis. X-linked.
Hyperornithinemia	Defective ornithine trans-porter protein	Failure to import ornithine from cytoplasm to mitochondria. Defect in ORNT1 gene. Hyperornithinemia, hyperammonemia and homocitrullinuria is seen (HHH syndrome). Decreased urea in blood. Autosomal recessive condition.

Urea cycle disorders

Diseases	Enzyme deficit	Features
Citrullinemia	Argininosuccinate synthe-tase	Autosomal recessive inheritance. High blood levels of ammonia and citrulline. Citrullinuria (1-2 g/day).
Argininosuccinic aciduria	Arginino-succinate lyase	Argininosuccinate in blood and urine. Friable brittle tufted hair (Trichorrhexis nodosa). Incidence 3/200,000
Hyperargininemia	Arginase	Arginine increased in blood and CSF. Instead of arginine, cysteine and lysine are lost in urine. Incidence 1 in 100,000

BLOOD UREA : 20 - 40 mg / dl

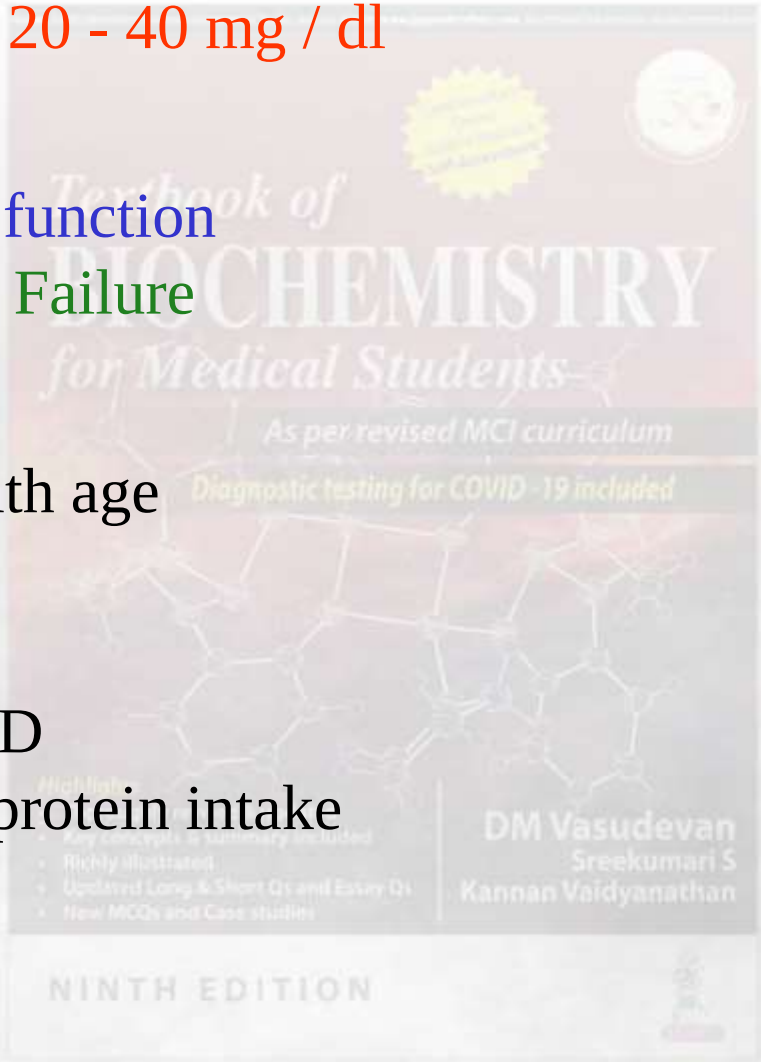
Indicator of Renal function

Increased in Renal Failure

Increased along with age

NOT INCREASED

in protein intake

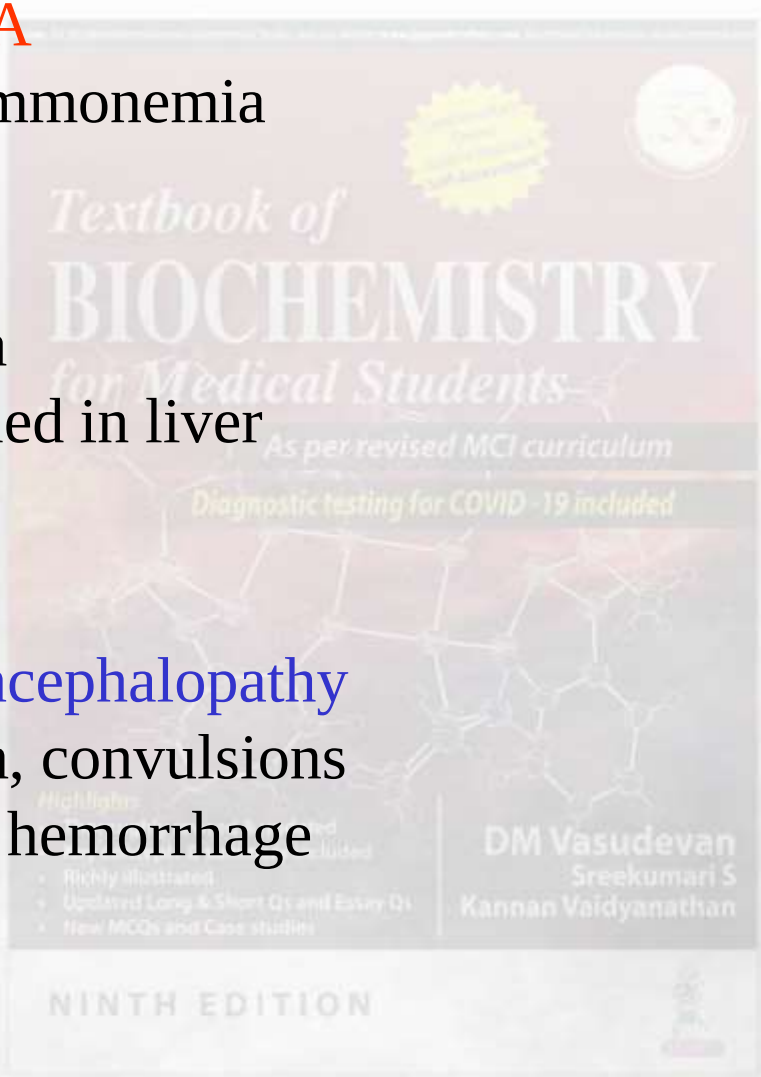


HEPATIC COMA

Acquired hyperammonemia

Intestinal bacteria
ammonia detoxified in liver

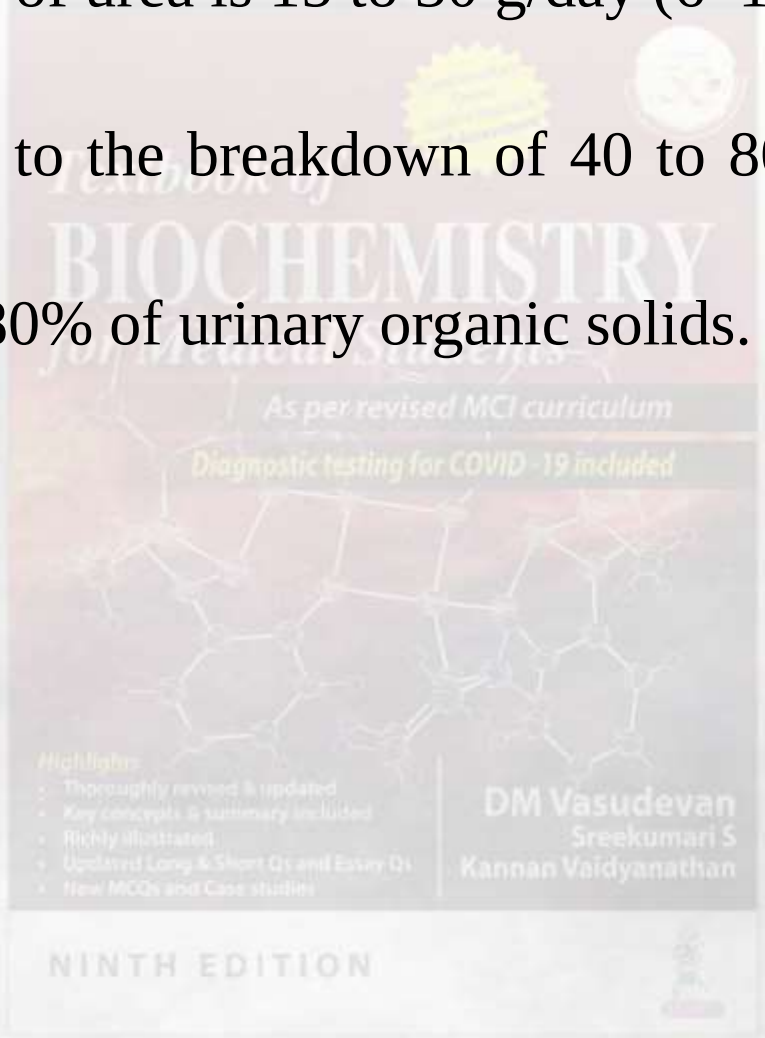
Portal systemic encephalopathy
Altered sensorium, convulsions
Ascites, jaundice, hemorrhage



Hepatic Coma (Acquired Hyperammonemia)

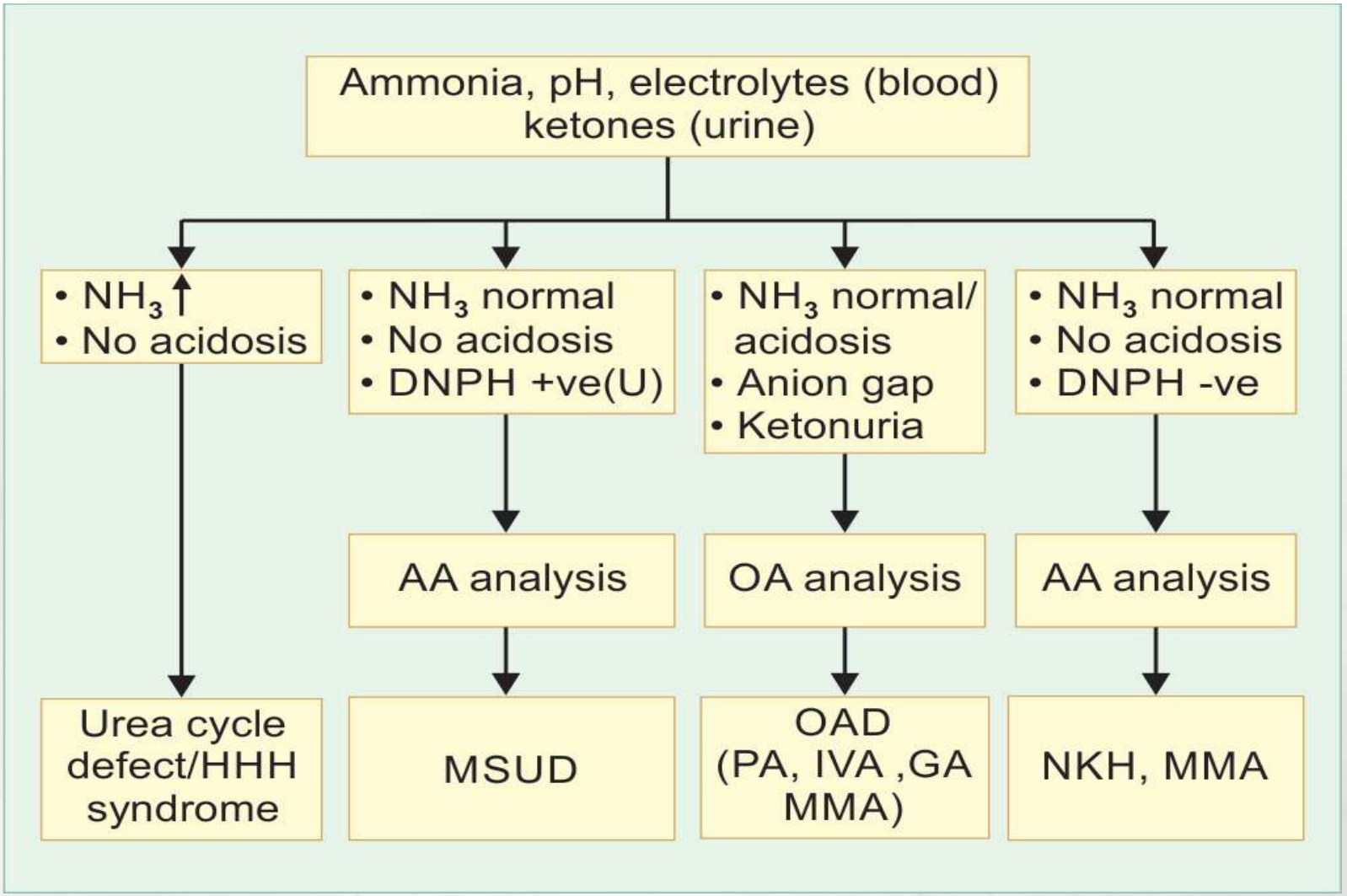
- The signs and symptoms are mainly pertaining to CNS dysfunction (altered sensorium, convulsions), or manifestations of failure of liver function (ascites, jaundice, hepatomegaly, edema, hemorrhage, spider nevi).
- The management of the condition is difficult.
- The main lines of treatment are
  1. Low protein diet.
  2. Bowel disinfection using antibiotics and clearance by lactulose.
  3. Avoid hepatotoxic drugs.
  4. Maintenance of electrolyte and acid base balance.

- Urinary excretion of urea is 15 to 30 g/day (6–15 grams nitrogen/day).
- This corresponds to the breakdown of 40 to 80 grams of proteins per day.
- Urea constitutes 80% of urinary organic solids.



Hyperammonemia

- Measurement of ammonia levels is a very critical parameter for the assessment of a variety of genetic and acquired conditions.
- The most important genetic causes of hyperammonemia are urea cycle disorders and organic acidurias.
- Organic acidurias usually present with life-threatening metabolic acidosis and hyperammonemia; whereas urea cycle disorders often have metabolic alkalosis along with hyperammonemia.
- Ammonia levels may be elevated in hepatic coma (hepatic encephalopathy). In this condition liver function tests will also often be grossly abnormal.
- Other conditions where ammonia is elevated include cor pulmonale, pulmonary emphysema and renal failure.

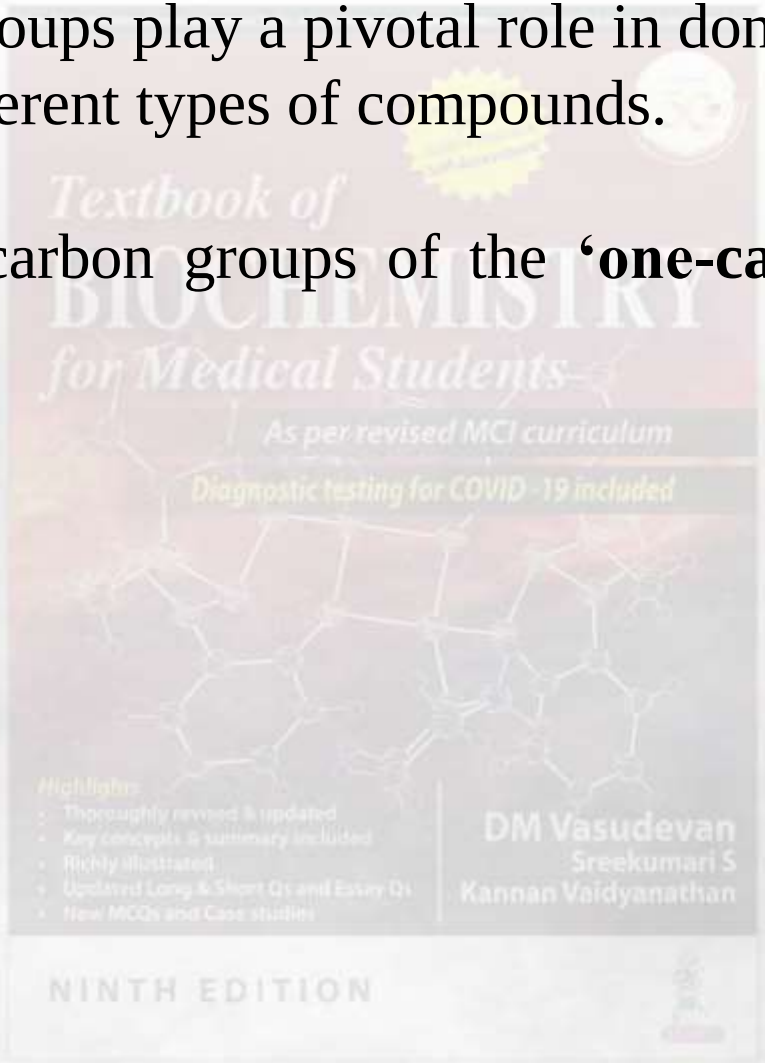


Flowchart for differential diagnosis of hyperammonemia.

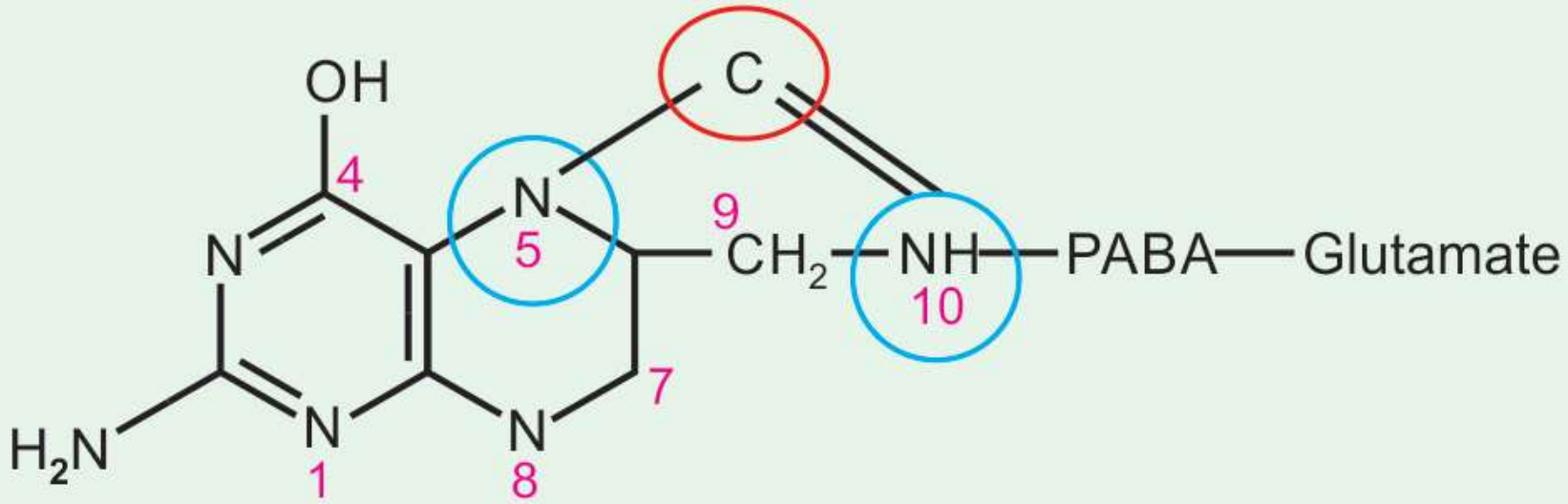
One-Carbon Metabolism

One-carbon (1C) groups play a pivotal role in donating carbon atoms for synthesis of different types of compounds.

The different one-carbon groups of the **'one-carbon pool'** of the body are



One-carbon compounds		
Group	**Structure**	**Carried by**
Formyl	–CHO	N^5–formyl–THFA and N^{10}–formyl–THFA
Formimino	–CH=NH	N^5–formimino–THFA
Methenyl	=CH–	N^5,N^{10}–methenyl–THFA
Hydroxymethyl	$–CH_2OH$	N^{10}–hydroxymethyl THFA
Methylene	$–CH_2–$	N^5,N^{10}–methylene–THFA
Methyl	$–CH_3$	N^5–methyl–THFA and methyl cobalamin



Tetrahydrofolic acid (THFA). Methenyl group is attached to N-5 and N-10.

Folic acid

NADPH + H+

Folate reductase

NADP+

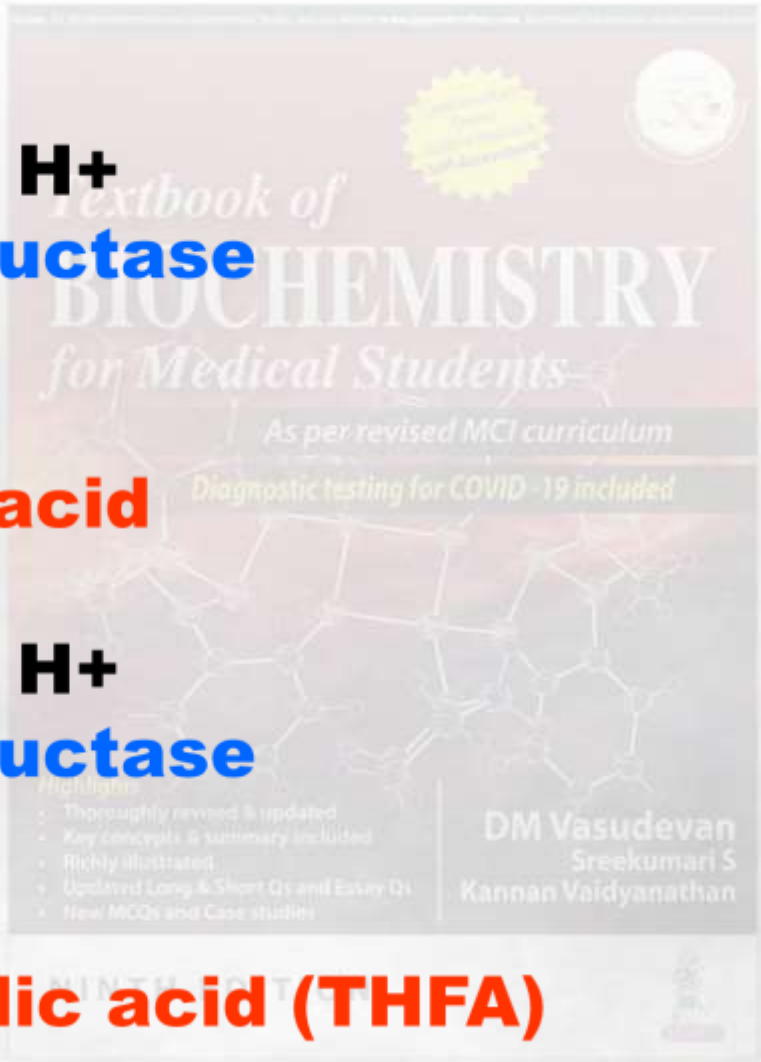
dihydro folic acid

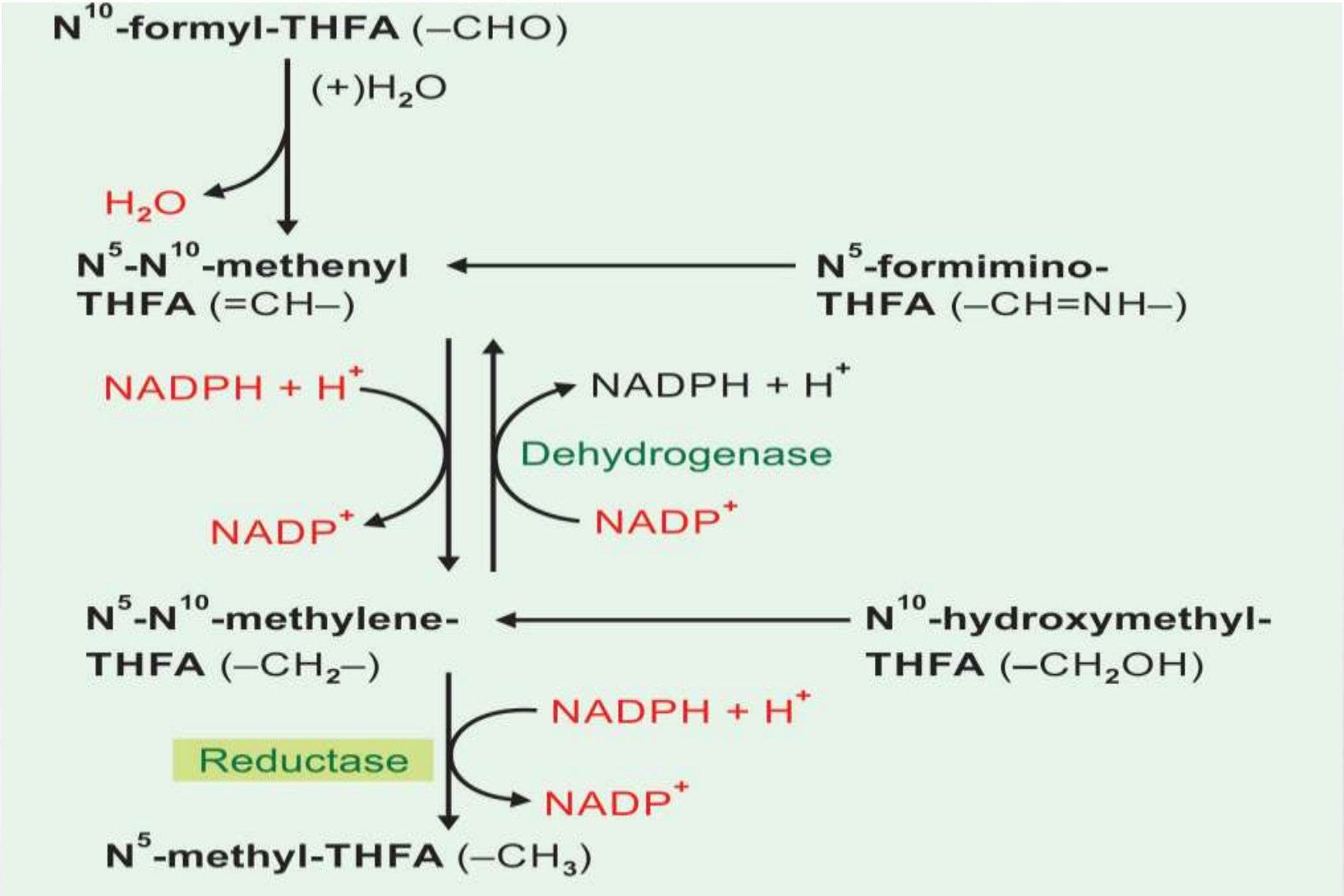
NADPH + H+

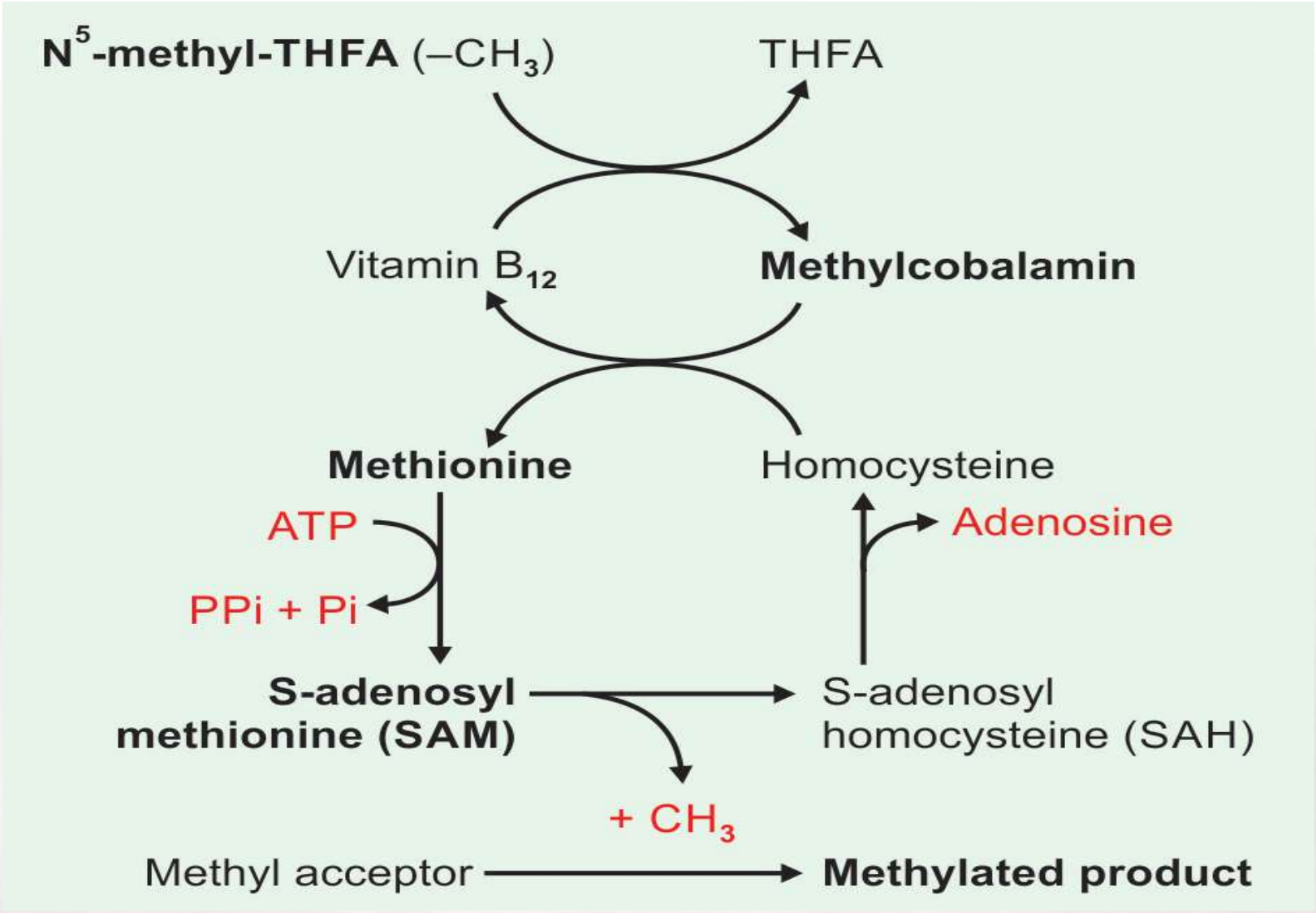
Folate reductase

NADP+

tetrahydro folic acid (THFA)



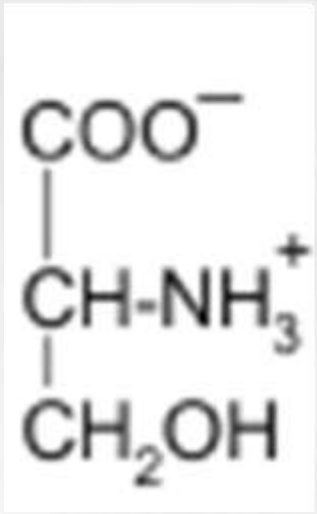

N10-formyl-THFA (–CHO)
(+)H2O
H2O
N5-N10-methenyl THFA (=CH–)
N5-formimino-THFA (–CH=NH–)
NADPH + H+
NADPH + H+
Dehydrogenase
NADP+
NADP+
N5-N10-methylene-THFA (–CH2–)
N10-hydroxymethyl-THFA (–CH2OH)
NADPH + H+
Reductase
NADP+
N5-methyl-THFA (–CH3)


N5-methyl-THFA (–CH3)
THFA
Vitamin B12
Methylcobalamin
Methionine
Homocysteine
ATP
Adenosine
PPi + Pi
S-adenosyl methionine (SAM)
S-adenosyl homocysteine (SAH)
+ CH3
Methyl acceptor
Methylated product

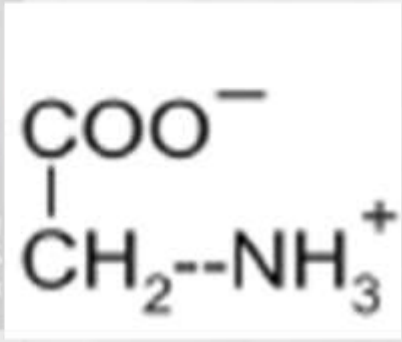
1. **Serine** to glycine is the primary contributor for methylene THFA.

Serine hydroxy methyl transferase

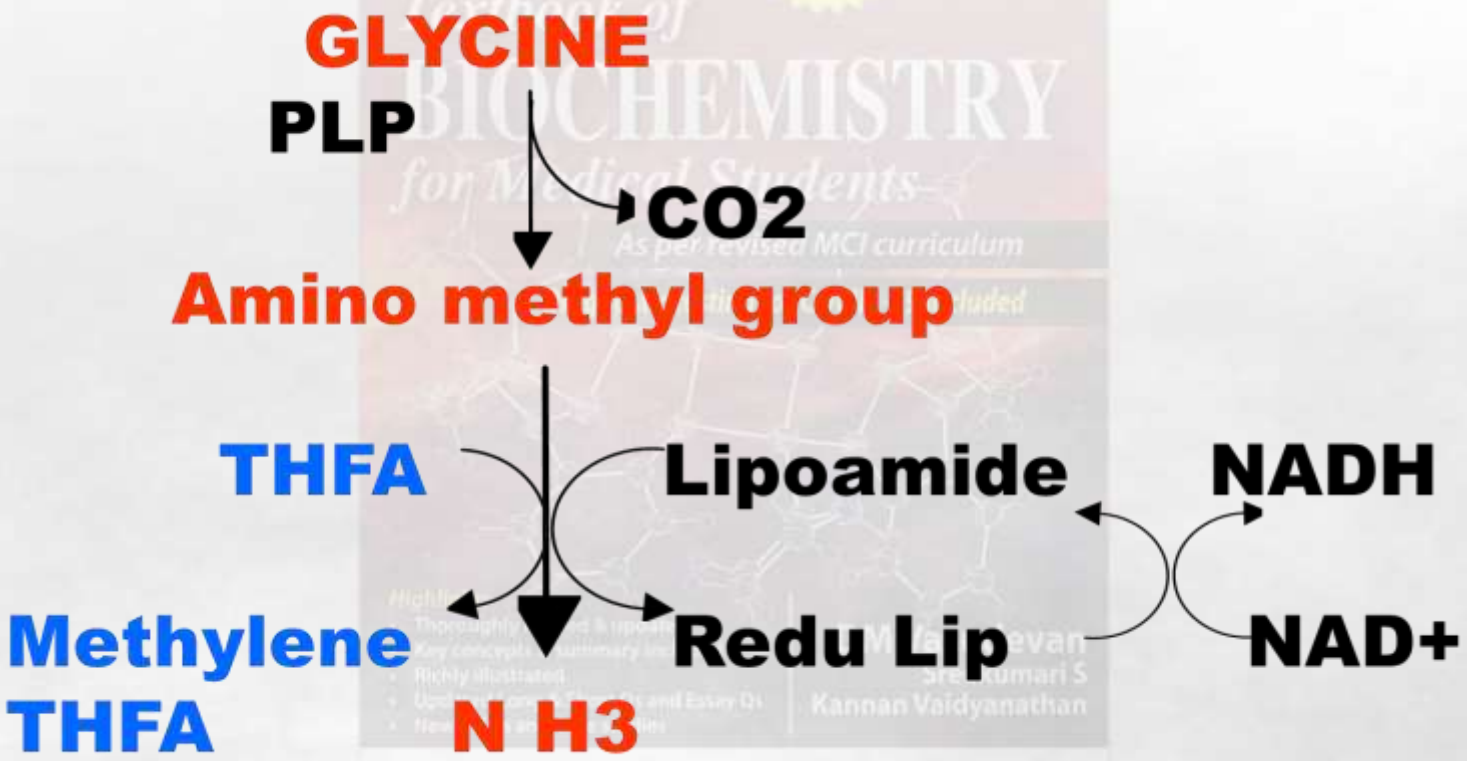
Serine → **Glycine**



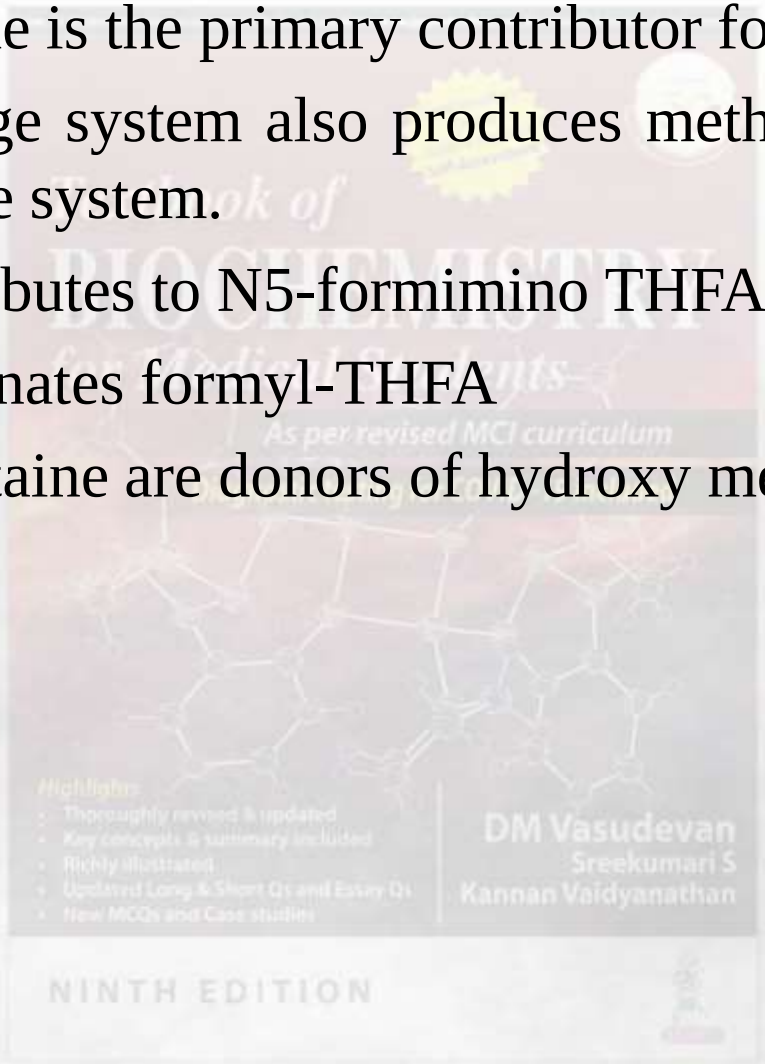
THFA → **N5,N10-Methylene-THFA**

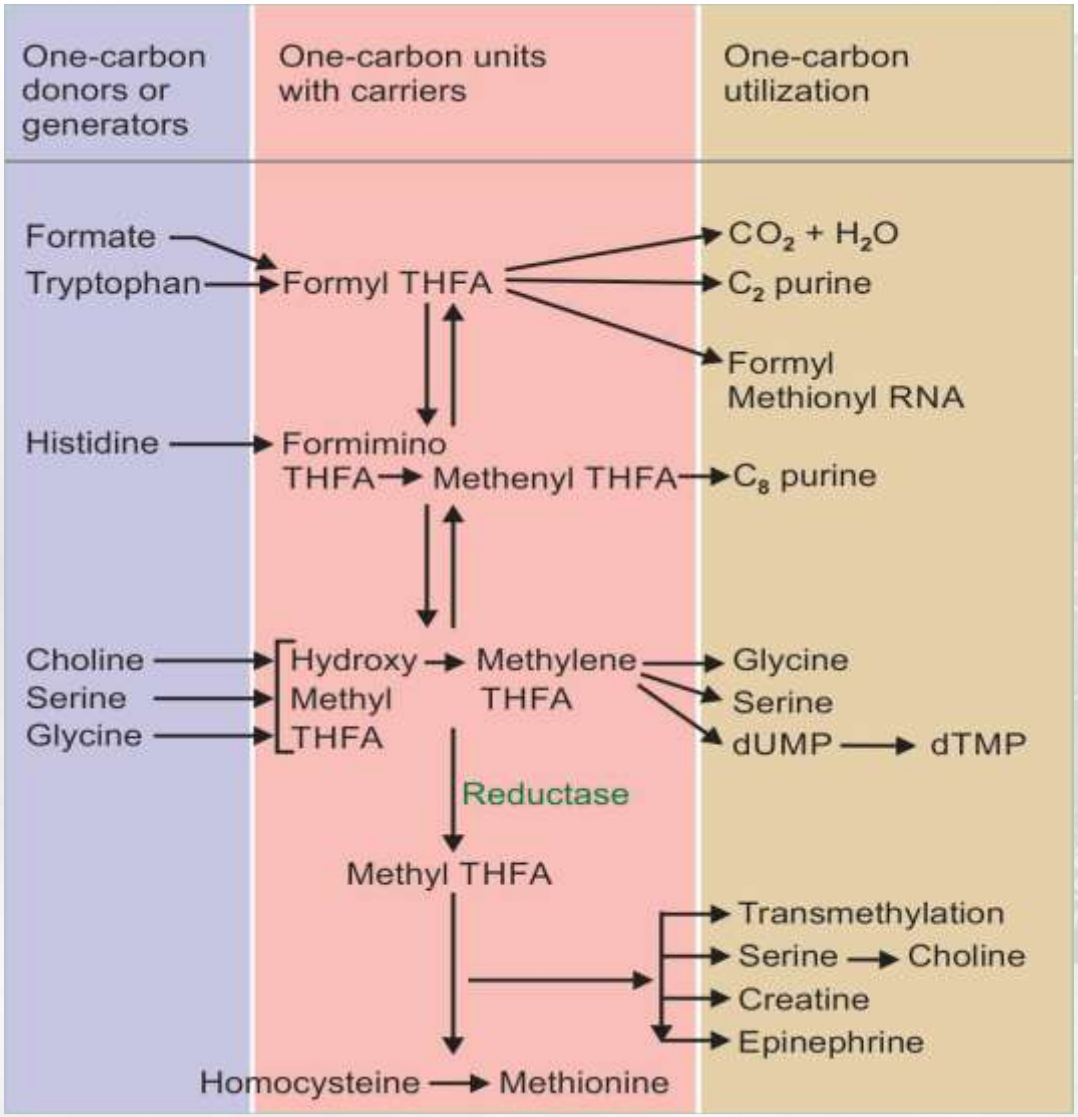


2. **Glycine** cleavage system also produces methylene groups is the glycine cleavage system



1. **Serine** to glycine is the primary contributor for methylene THFA.
2. **Glycine** cleavage system also produces methylene groups is the glycine cleavage system.
3. **Histidine** contributes to N5-formimino THFA through FIGLU
4. **Tryptophan** donates formyl-THFA
5. **Choline** and betaine are donors of hydroxy methyl groups



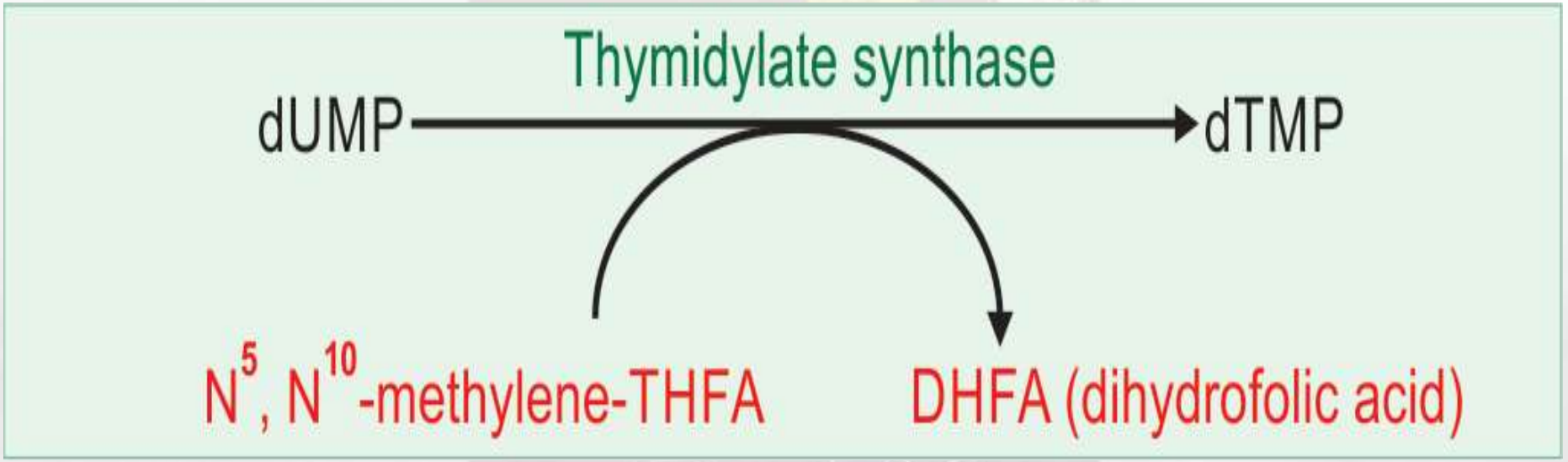


One-carbon generation and utilization

Utilization of One-Carbon Groups

The one-carbon units are used for synthesis of the following compounds:

1. C2 of purine
2. Formylation of methionyl tRNA
3. C8 of purine
4. **Glycine**
5. Serine
6. **Choline**
7. **Deoxy TMP**
8. **Transmethylation reactions** including creatine, choline and epinephrine synthesis
9. Eliminated as carbon dioxide.



Synthesis of deoxy thymidylic acid.

Transmethylation Reactions

Methyl acceptor	Methylated product
Guanido acetic acid	Creatine
Nicotinamide	N-methyl nicotinamide
Norepinephrine	Epinephrine
Epinephrine	Metanephrine
Norepinephrine	Normetanephrine
Ethanolamine	Choline
Carnosine	Anserine
Acetyl serotonin	Melatonin
Serine	Choline
Histidine	Methyl histidine
Lysine	Methyl lysine
tRNA	Methylated tRNA