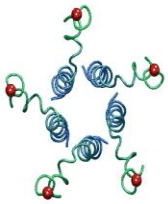
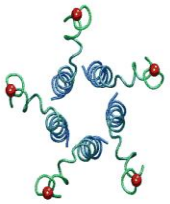


Protein metabolism

By

Dr. Anwar Almzaiel



GOALS

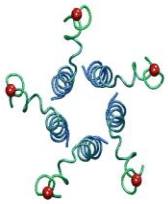
-Amino acid pool

- Protein turnover
- Nitrogen Balance
- Digestion of Dietary Proteins
- Removal of Nitrogen from Amino Acids

A. Transamination

B. oxidative deamination

- Transport of ammonia to the liver
- Urea Cycle
- Transport of ammonia in the circulation

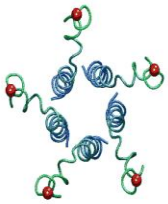


Introduction

- Proteins are degraded into amino acids.
- Protein turnover is tightly regulated.
- First step in protein degradation is the removal of the nitrogen
- Ammonium ion is converted to urea in most mammals.
- Carbon atoms are converted to other major metabolic intermediates.

Metabolic uses of amino acids

- building blocks for protein synthesis
- precursors of nucleotides and heme
- source of energy
- neurotransmitters
- precursors of neurotransmitters and hormones



The amino group nitrogen is converted to urea and excreted.

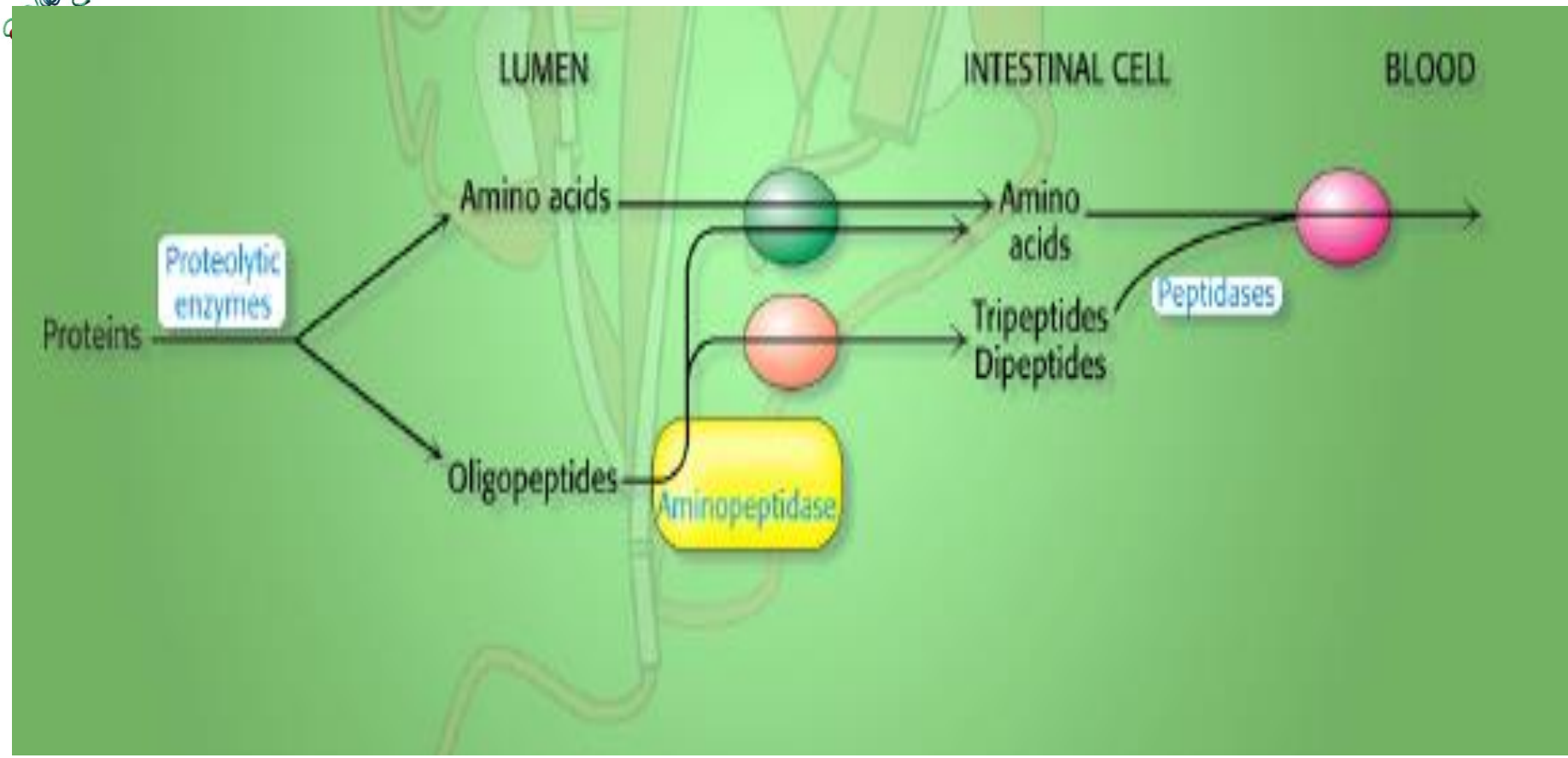
-Glucose, fatty acids and ketone bodies can be formed from amino acids.

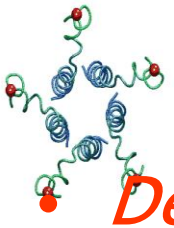
Dietary Protein Degradation

Dietary proteins are a vital source of amino acids.

Discarded cellular proteins are another source of amino acids.

Dietary proteins are hydrolyzed to amino acids and absorbed into the bloodstream.


LUMEN
INTESTINAL CELL
BLOOD
Amino acids
Amino acids
Proteolytic enzymes
Proteins
Tripeptides
Dipeptides
Peptidases
Oligopeptides
Aminopeptidase

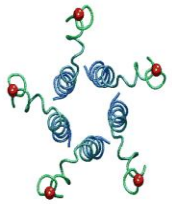


some major biological functions

* Detoxification of drugs, chemicals and metabolic by-products

* Excess dietary amino acids(AAs)are neither stored nor excreted. Rather, they are converted to common metabolic intermediates444

- The requirements of protein for the health: the minimal requirement of protein is 30~50 gram for the adult
- Advice: 80 gram/day? ? ?



Nitrogen balance

- Zero or total nitrogen balance:

  the intake = the excretion (adult)

  Amount of nitrogen intake is equal to the amount of nitrogen excreted is zero or total nitrogen balance
- Positive nitrogen balance:

  the intake > the excretion

  during pregnancy, infancy, childhood and recovery from severe illness or surgery
- Negative nitrogen balance:

  the intake < the excretion

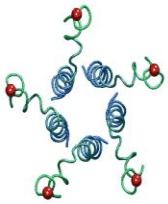
  following severe trauma, surgery or infections. Prolonged periods of negative balance are dangerous and fatal if the loss of body protein reaches about one-third of the total body protein

CHEMICAL NATURE OF PROTEINS

All proteins are polymers of amino acids. The amino acids in proteins are united through "Peptide" linkage. Sometimes proteins are also called as polypeptides because they contain many peptide bonds.

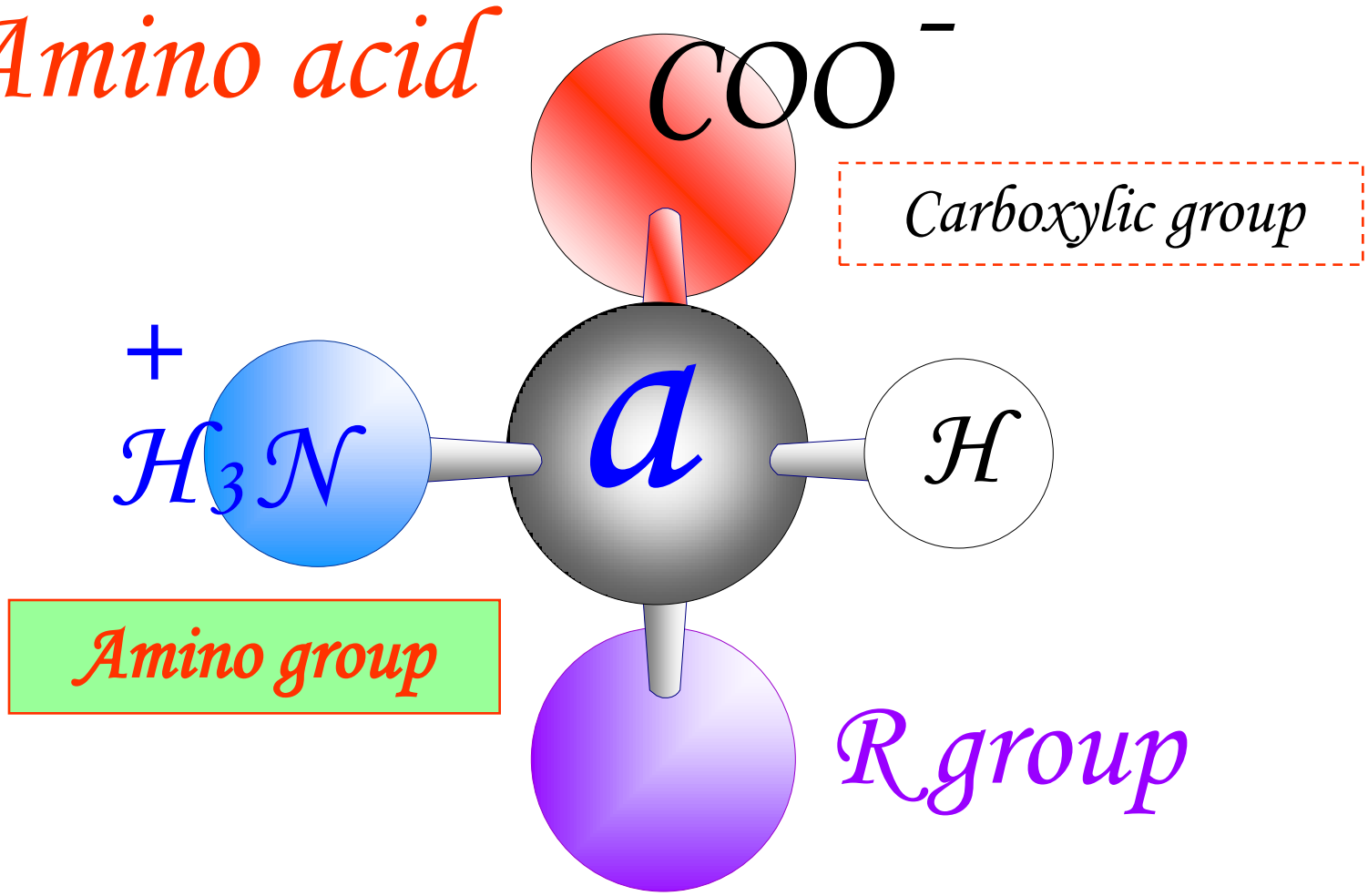
Amino acids (a.a)

-Amino acids are the fundamental units of proteins.
-Amino acids are composed of an amino group (-NH2), a carboxyl group (-COOH), a hydrogen atom
-Proteins are polymers of amino acids, with each amino acid residue joined to its neighbor by a specific type of covalent bond.
-Proteins can be broken down (hydrolyzed) to their constituent amino acids the free amino acids derived from them.



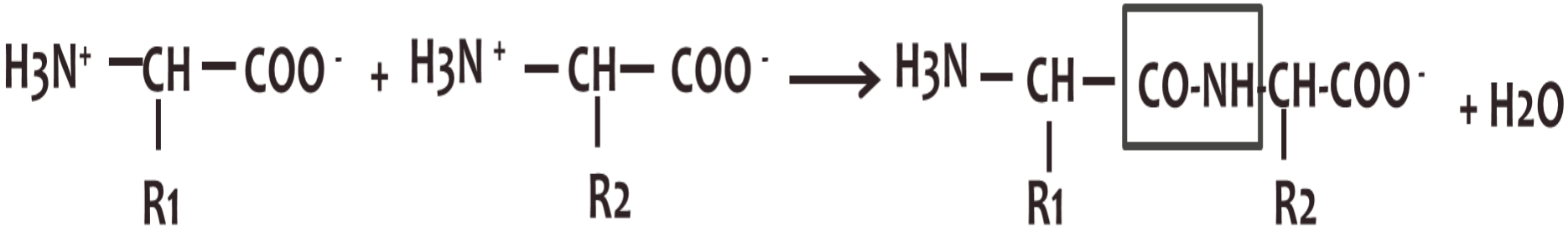
Amino Acids

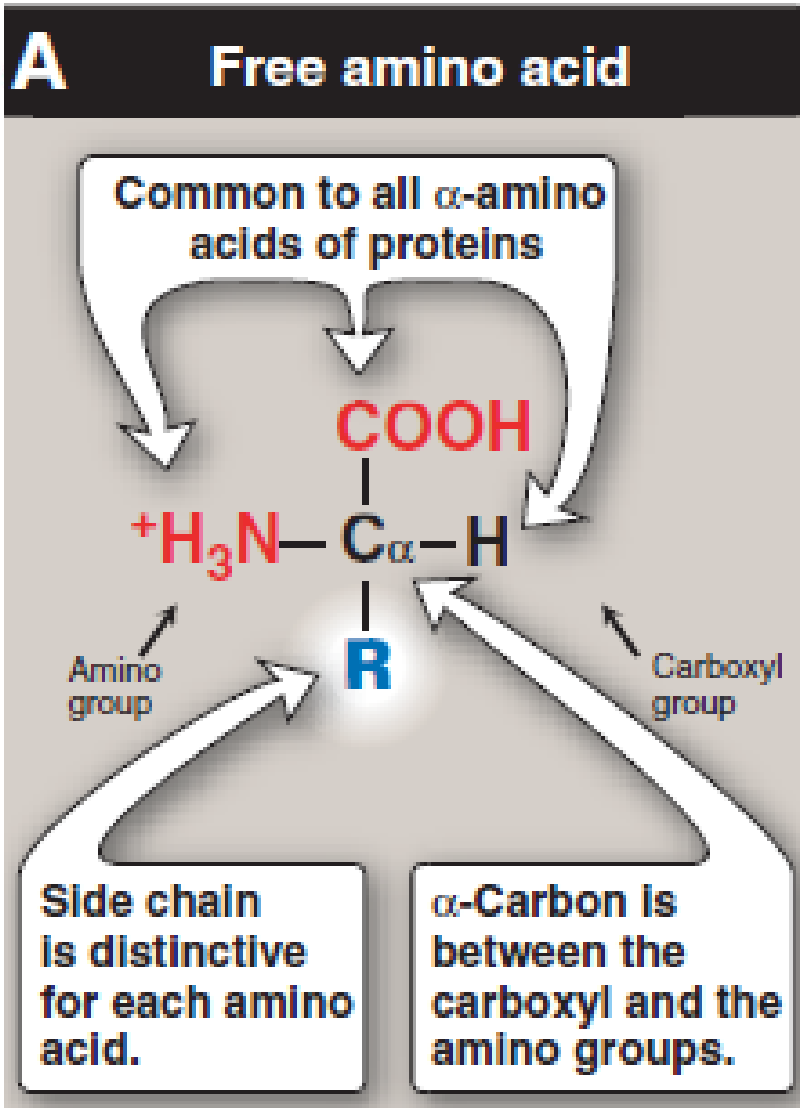
Amino acid

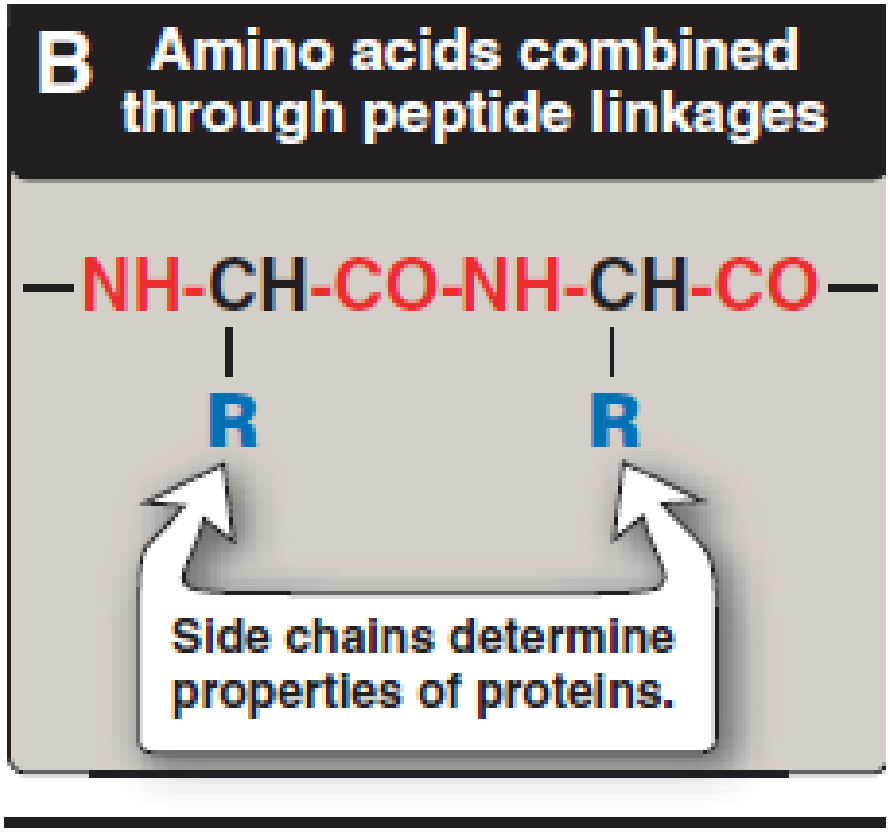


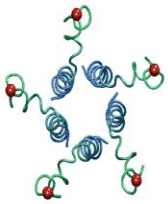
Peptides and polypeptide:

The linkage of a.a together produces peptide chains or polypeptides if many amino acids are linked. The peptide bond is the bond formed between the α-carboxyl group of one a.a and the α-amino group of another, H_2O is removed.




A
Free amino acid
Common to all α-amino acids of proteins
COOH
+H3N–Cα–H
R
Amino group
Carboxyl group
Side chain is distinctive for each amino acid.
α-Carbon is between the carboxyl and the amino groups.


B
Amino acids combined through peptide linkages
–NH-CH-CO-NH-CH-CO–
R
R
Side chains determine properties of proteins.



Classification of amino acids:

1. Chemical classification.

a. According to the **chemistry** of the side chains.

b. According to **polarity** of side chains.

3. Nutritional classification :

- Essential
- Non-essential

3. Metabolic classification :

- Glucogenic,
- Ketogenic
- Both glucogenic and ketogenic

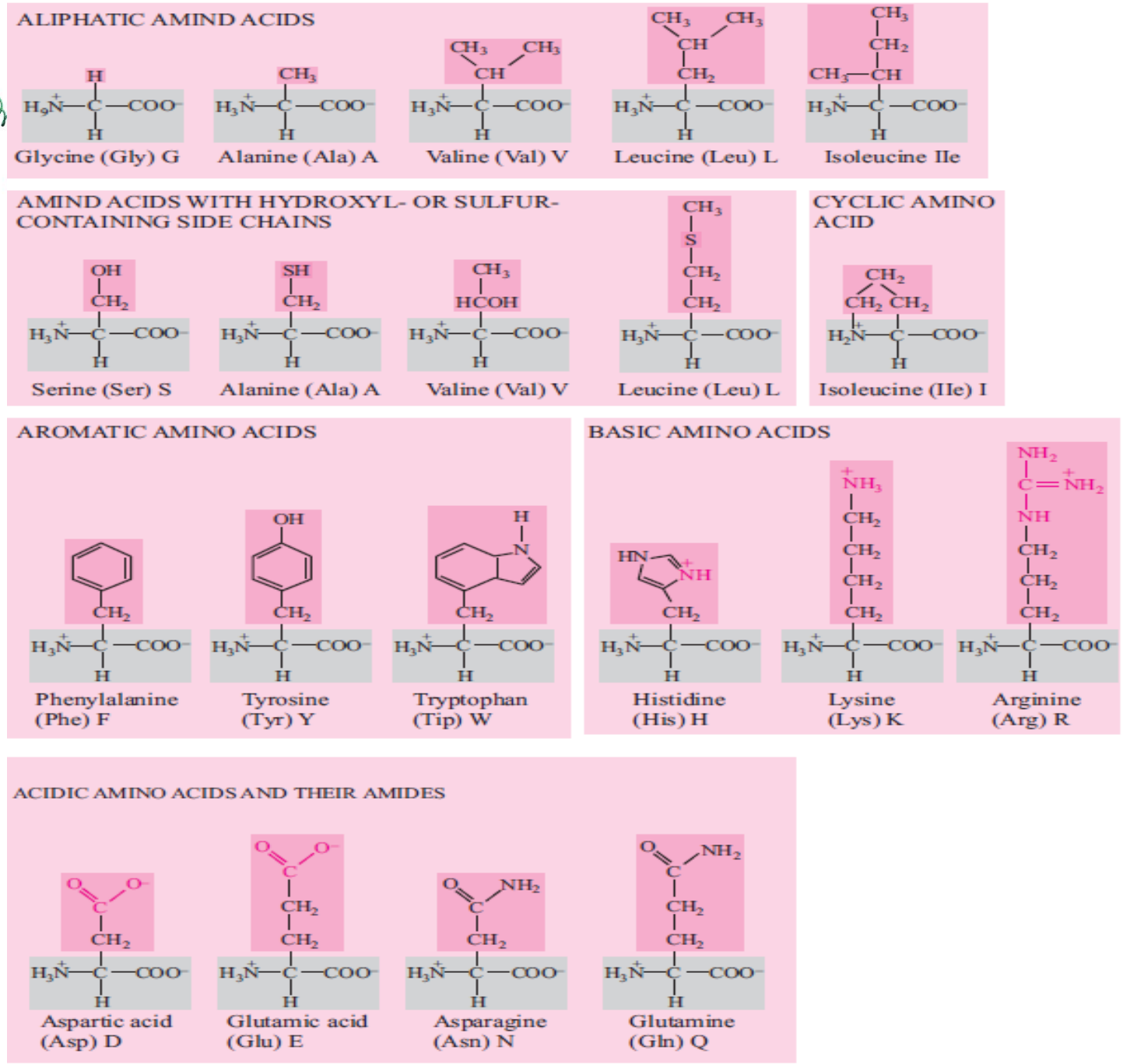
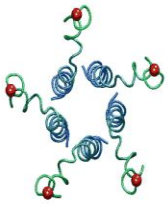


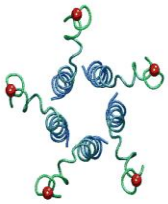
Fig. 4.5: Classification of amino acids.



Classification of amino acids

- ***non-essential amino acids***
  - *can be synthesized by an organism*
  - *usually are prepared from precursors in 1-2 steps*
- ***Essential amino acids*** ***
  - ***cannot be made endogenously***
  - ***must be supplied in diet***

    eg. Leu, Phe…..



Nonessential	Essential
Alanine	Arginine*
Asparagine	Histidine *
Aspartate	Valine
Cysteine	Lysine
Glutamate	Isoleucine
Glutamine	Leucine
Glycine	Phenylalanine
Proline	Methionine
Serine	Threonine
Tyrosine	Tyrptophan

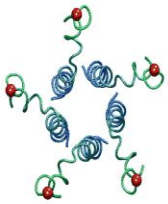
*The amino acids Arg, His are considered "conditionally essential" for reasons not directly related to lack of synthesis and they are essential for growth only

Essential and nonessential amino acids

Essential	Nonessential
Arginine	Alanine
Histidine	Aspartate
Isoleucine	Asparagine
Leucine	Cysteine
Lysine	Glutamate
Methionine	Glutamine
Phenylalanine	Glycine
Threonine	Proline
Trptophan	Serine
Valine	Tyrosine

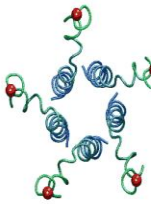
Glucogenic and ketogenic amino acids

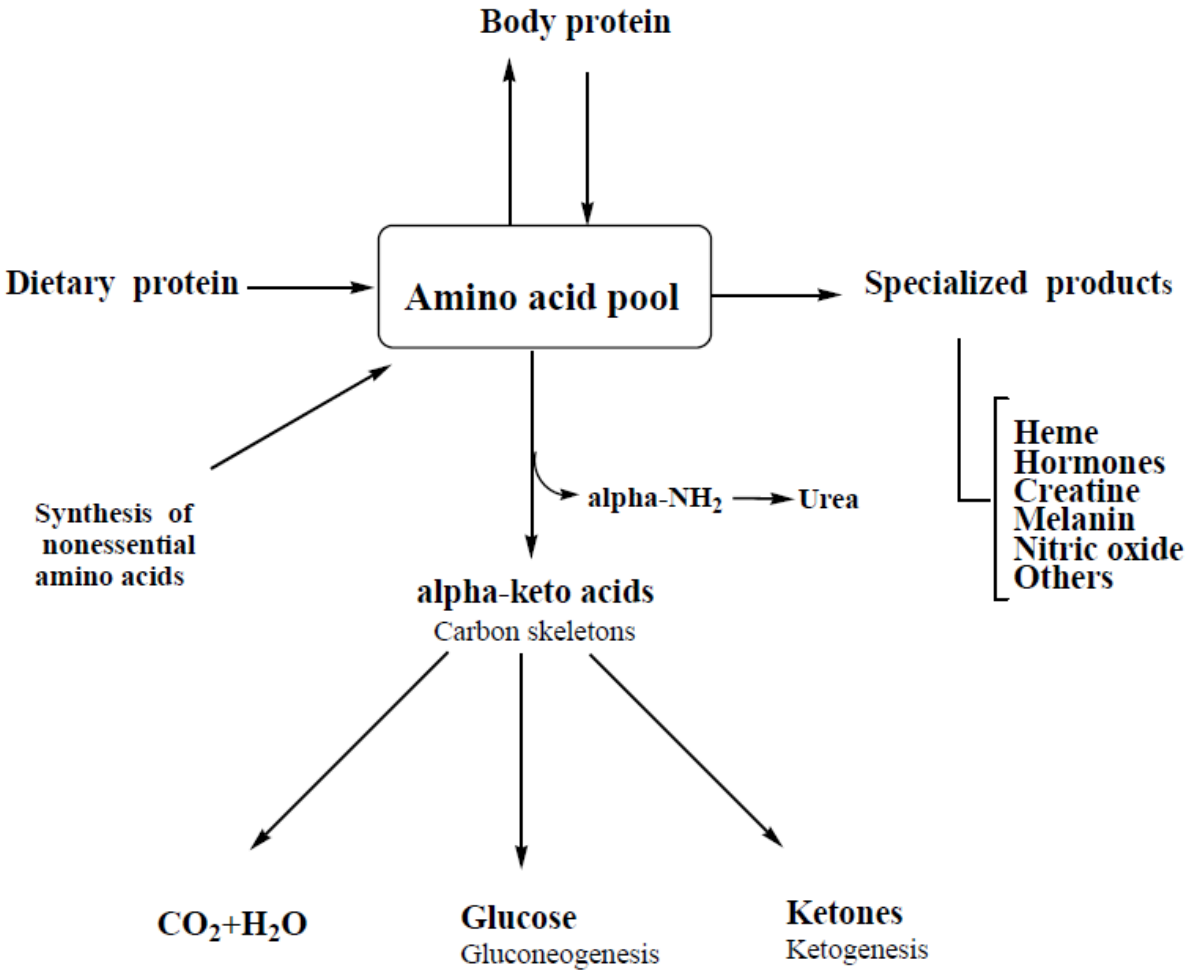
Glucogenic		Both Glucogenic and ketogenic	ketogenic
Alanine	Arginine	Isoleucine	Leucine
Asparagine	Aspartate	Phenylalanine	Lysine
Cysteine	Glutamate	Trptophan	
Glutamine	Glycine	Tyrosine	
Histidine	Methionine		
Proline	Serine		
Threonine	Valine		



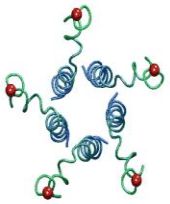
Amino acids Abbreviations

Amino acid	3-letter abbreviation	1-letter abbreviation
Alanine	Ala	A
Arginine	Arg	R
Asparagine	Asn	N
Aspartic acid	Asp	D
Cysteine	Cys	C
Glutamic acid	Glu	E
Glutamine	Gln	Q
Glycine	Gly	G
Histidine	His	H
Isoleucine	Ile	I
Leucine	Leu	L
Lysine	Lys	K
Methionine	Met	M
Phenylalanine	Phe	F
Proline	Pro	P
Serine	Ser	S
Threonine	Thr	T
Tryptophan	Trp	W
Tyrosine	Tyr	Y
Valine	Val	V



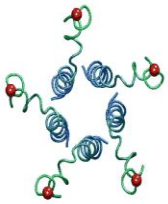


Overview of amino acid metabolism



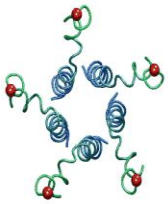
Protein catabolism
Digestive Tract of protein

- Proteins are generally too large to be absorbed by the intestine and therefore must be hydrolyzed to the amino acids
- The proteolytic enzymes responsible for hydrolysis are produced by three different organs: the stomach pancreas and small intestine (the major organ)



Stomach

- HCl (parietal cells) and Pepsinogen (chief cells)
- The pH of gastric juice is around 1.0. Food is retained in the stomach for 2-4 hrs
- HCl kills microorganisms, denatures proteins, and provides an acid environment for the action of pepsin
- Autocatalysis: pepsinogen is converted to active pepsin(*Pepsin A)* by HCl
- Pepsin coagulates milk in presence of Ca^{2+} ions

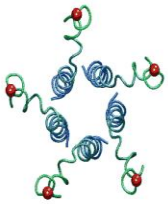


Pancreas and small intestine

- Endopeptidase (pancreas)
  - Trypsin: carbonyl of arg and lys
  - Chymotrypsin: carbonyl of Trp, Tyr, Phe, Met, Leu
  - Elastase: carbonyl of Ala, Gly, Ser

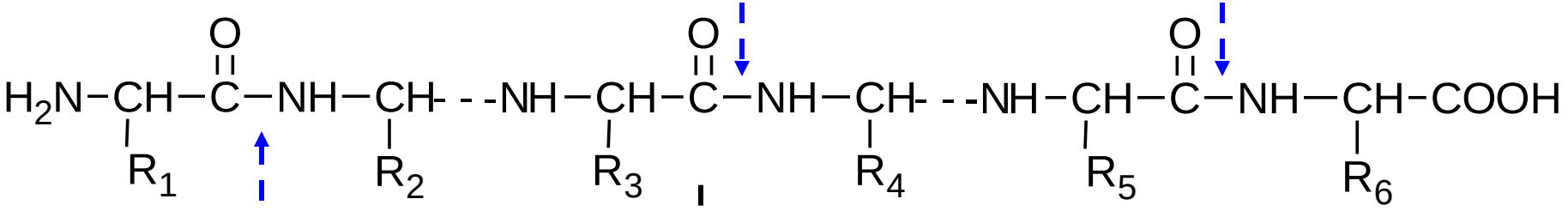
- Exopeptidase (pancreas)
  - Carboxypeptidase A:amine side of Ala, Ile, Leu, Val
  - Carboxypeptidase B: amine side of Arg, lys

- Aminopeptidase (small intestine):
  - cleaves N-terminal residue of oligopeptidaes
  - Dipeptidase (small intestine)



endopeptidase

carboxypepidase

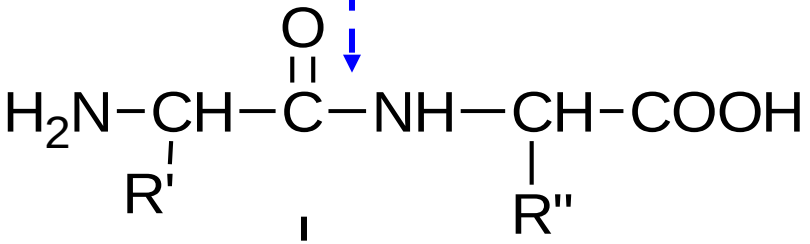


aminopeptidase

dipeptidase

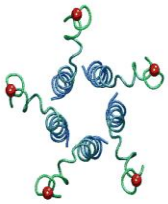
Amino acids +

1/3



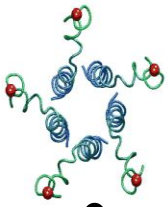
Amino acids

95%



absorption

- There is little absorption from the stomach apart from short- and medium- chain fatty acids and ethanol
- Under normal circumstances, the dietary proteins are almost completely digested to their constituent amino acids, and these end products of protein digestion are rapidly absorbed from the intestine into the portal blood



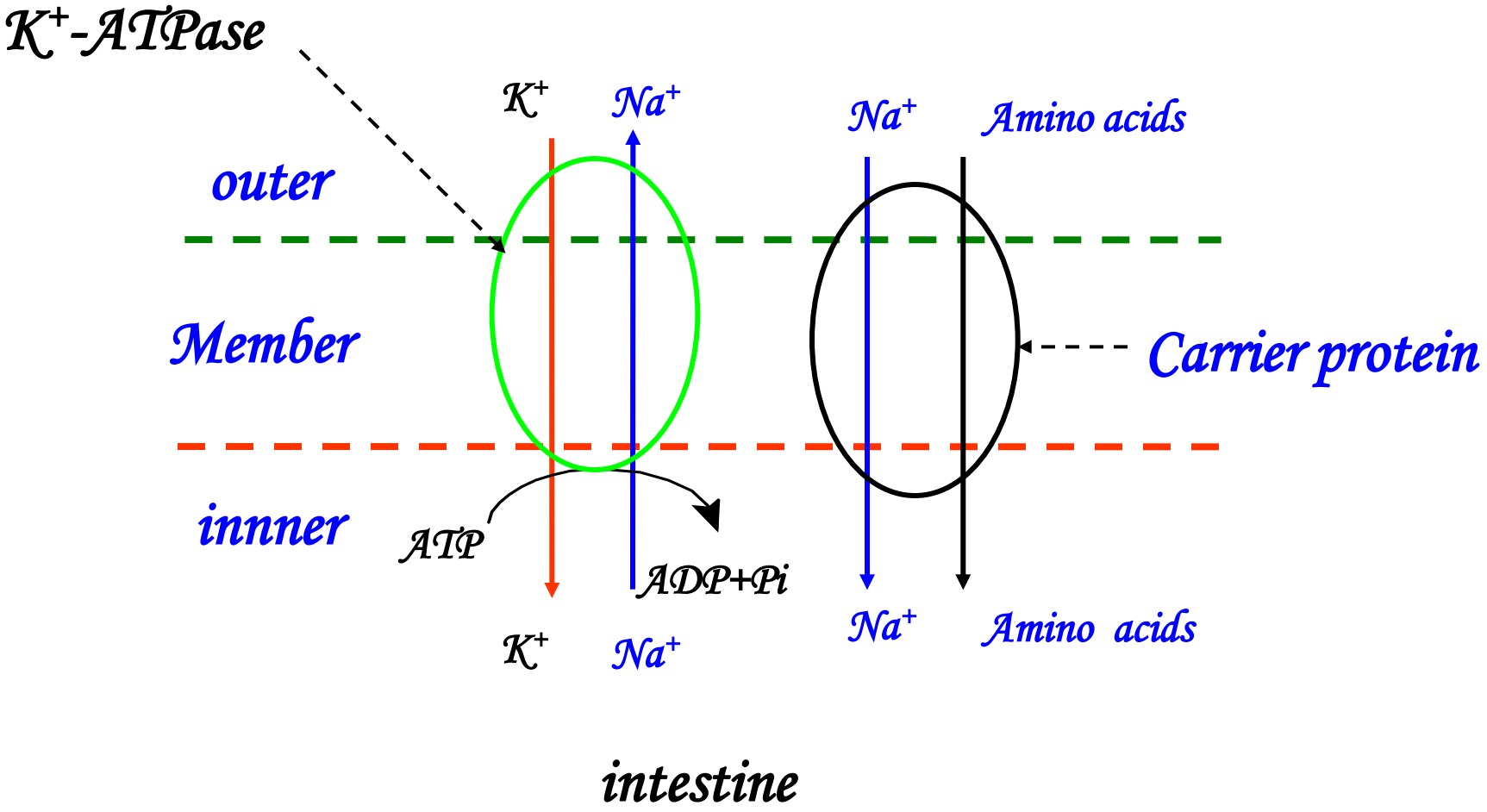
- Amino acids are transported through the brush border by the carrier protein and it is an active transport

1. The classification of carrier protein:

   aciditic; basic; neutral and gly-carrier

2. γ-glutamyl cycle
3. The bi-and tri- peptidase carrier system in the intestinal mucosa cell

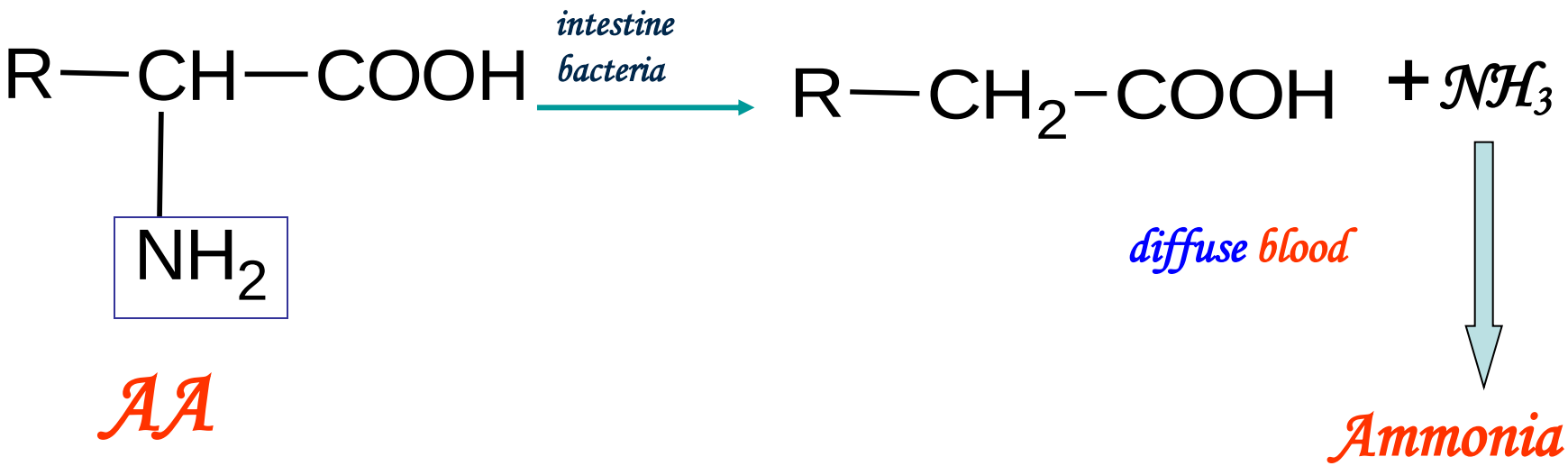
The mechanism of AA's absorption



2-Ammonia

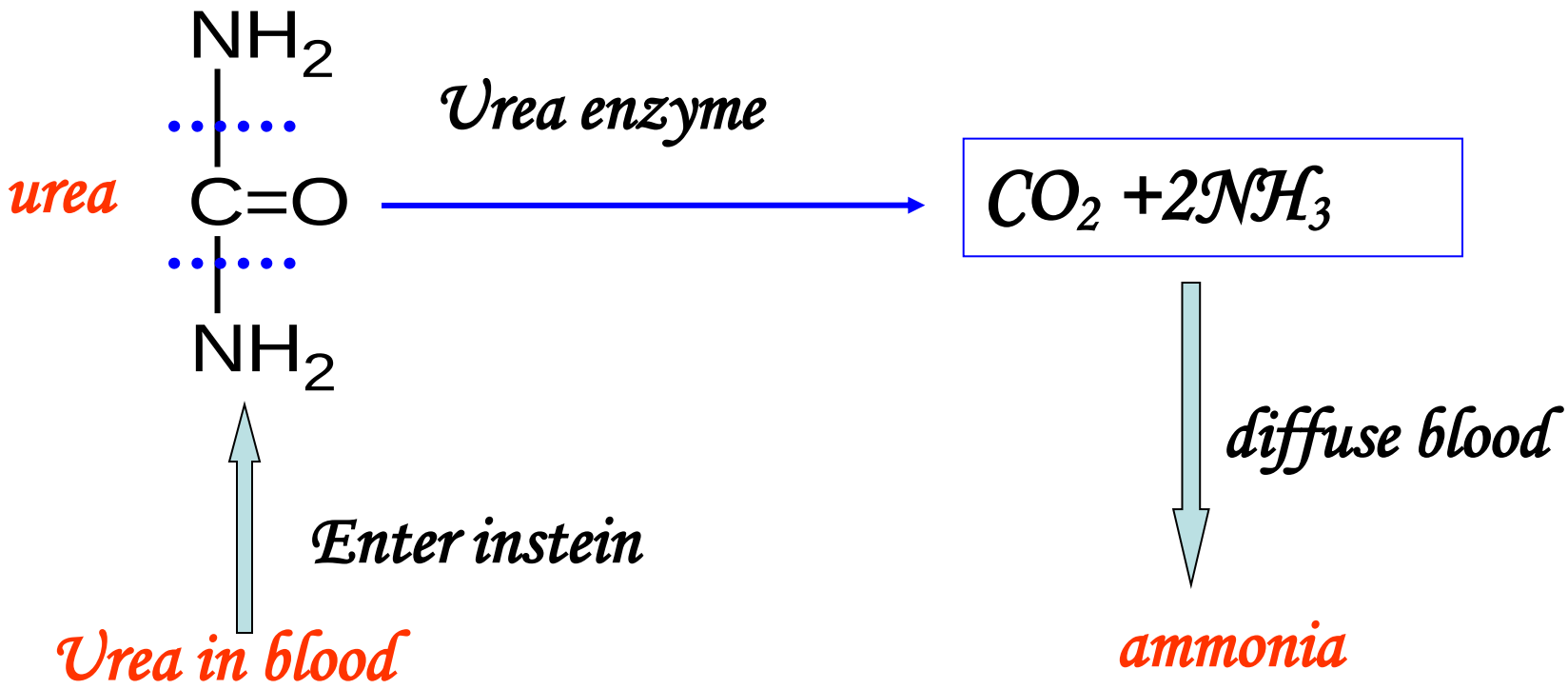
Source of ammonia

A. some amino acids are degraded by the in the intestine bacteria

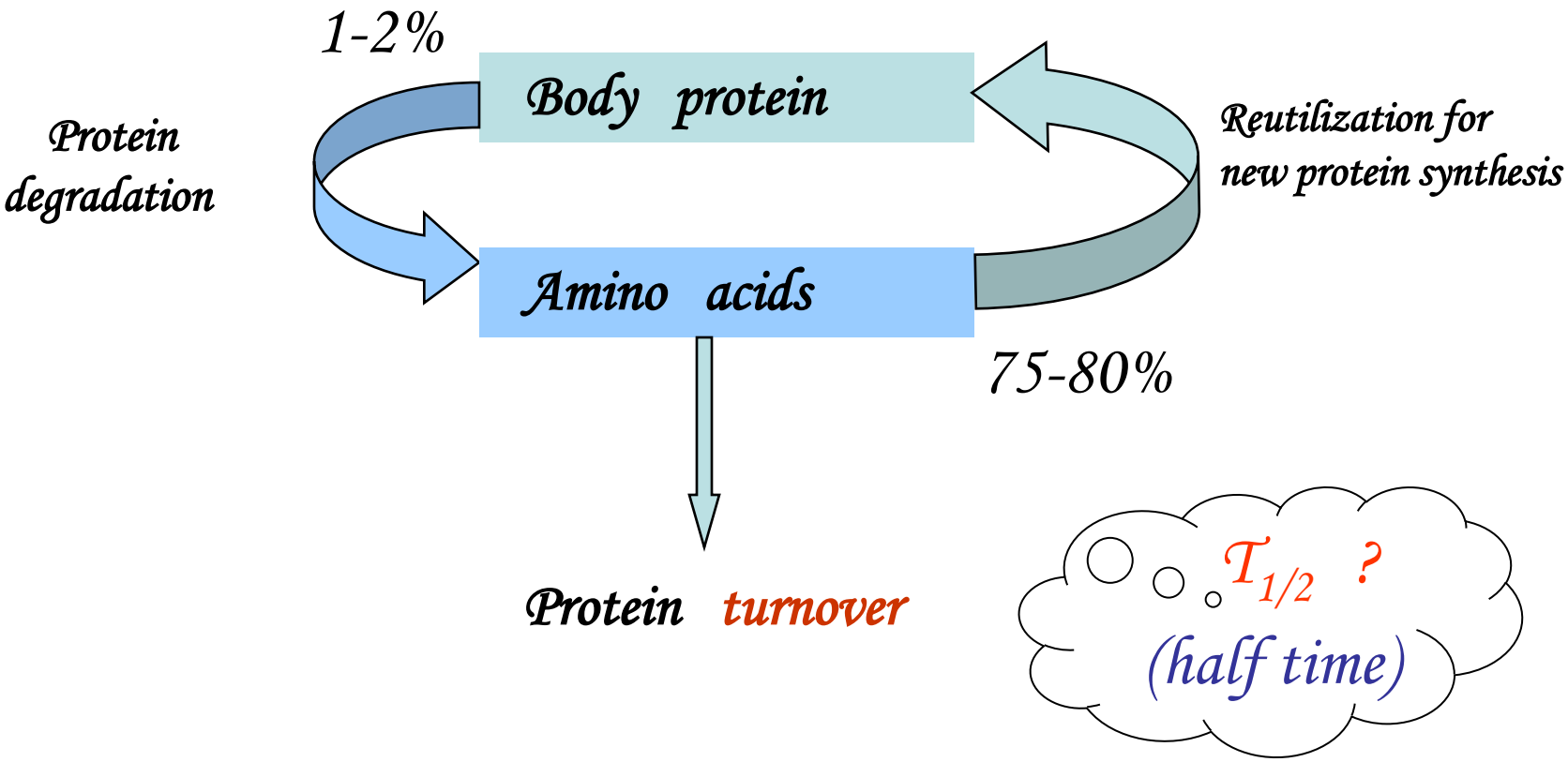


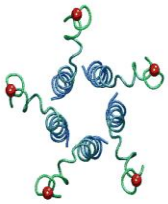
2. Ammonia

B. urea from the blood to the intestine with resultant increased diffusion of NH_3 into the intestinal



Protein and amino acid turnover





Diet protein

Tissue protein

Amino acid pool

Nonprotein nitrogen derivatives

Carbohydrate (glucose)

transamination

Ketone dodies

Acetyl-CoA

Amino nitrogen in glutamate

deamination

NH_3

Urea

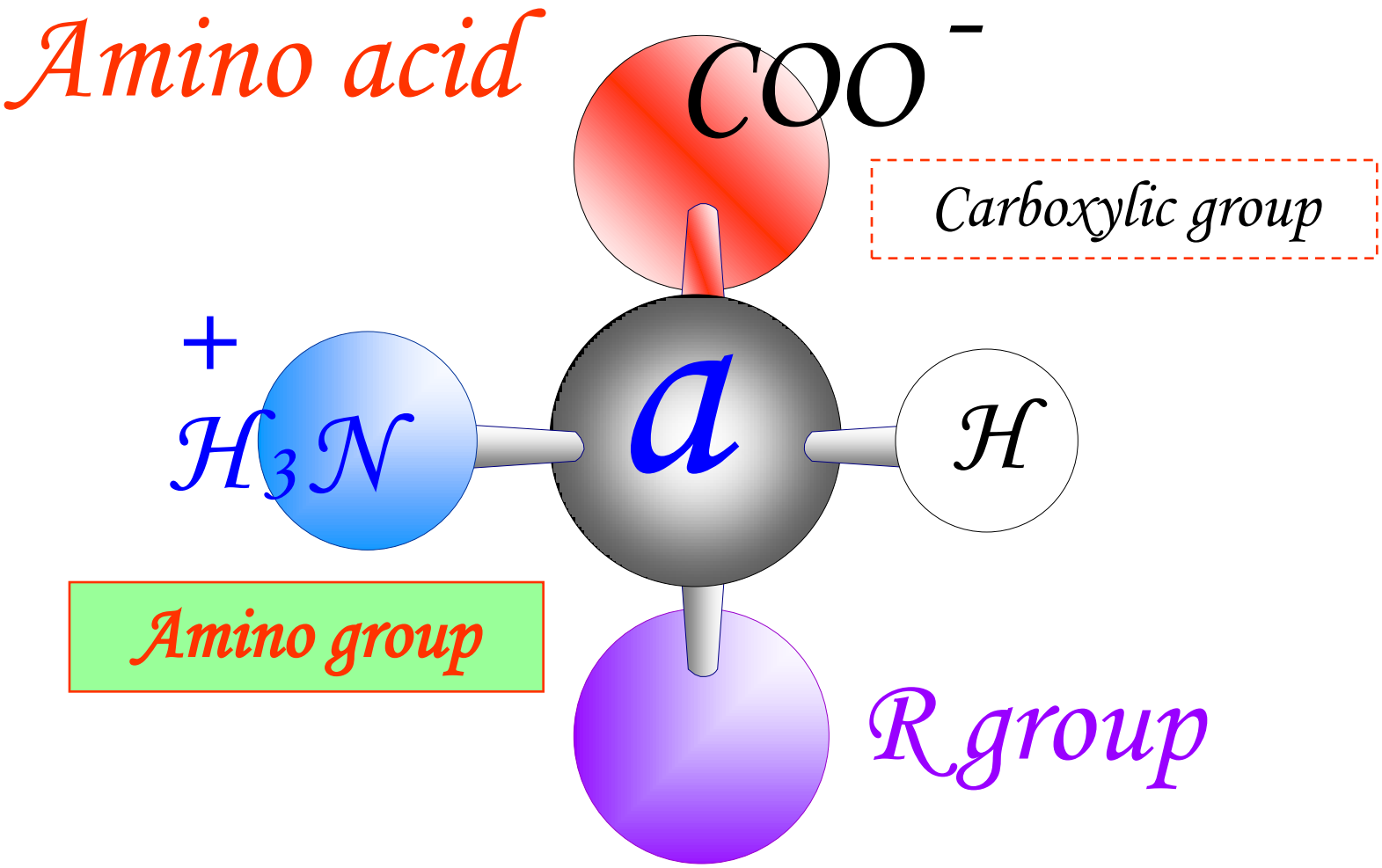
Citric Acid Cycle

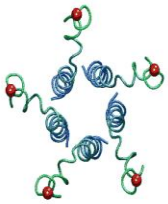
CO_2

Overview of the protein metabolism

Degradation of Amino Acids

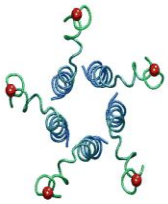
– Reactions in amino acid metabolism





introduction

- Free amino acids are metabolized in identical ways, regardless of whether they are released from dietary or intracellular proteins
- The metabolism of the resulting amino group and nitrogen excretion are a central part of nitrogen metabolism



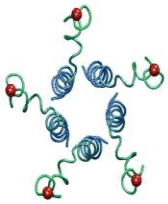
FATE OF AMINO GROUP

DEAMINATION

A. Transamination

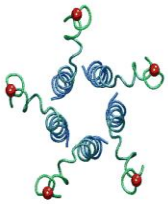
B. Oxidative deamination

C. purine nucleotide cycle



A. Transamination

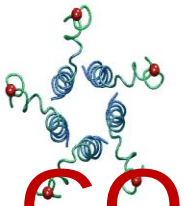
- Transamination by Aminotransferase (transaminase)
- always involve PLP coenzyme (pyridoxal phosphate)
- reaction goes via a Schiff's base intermediate
- all transaminase reactions are reversible



Aminotransferases

- Aminotransferases can have specificity for the alpha-keto acid or the amino acid
- Aminotransferases exist for all amino acids except proline and lysine
- The most common compounds involved as a donor/acceptor pair in transamination reactions are glutamate and a-ketoglutarate, which participate in reactions with many different aminotransferases

to an alpha-keto acid → alpha-amino acid



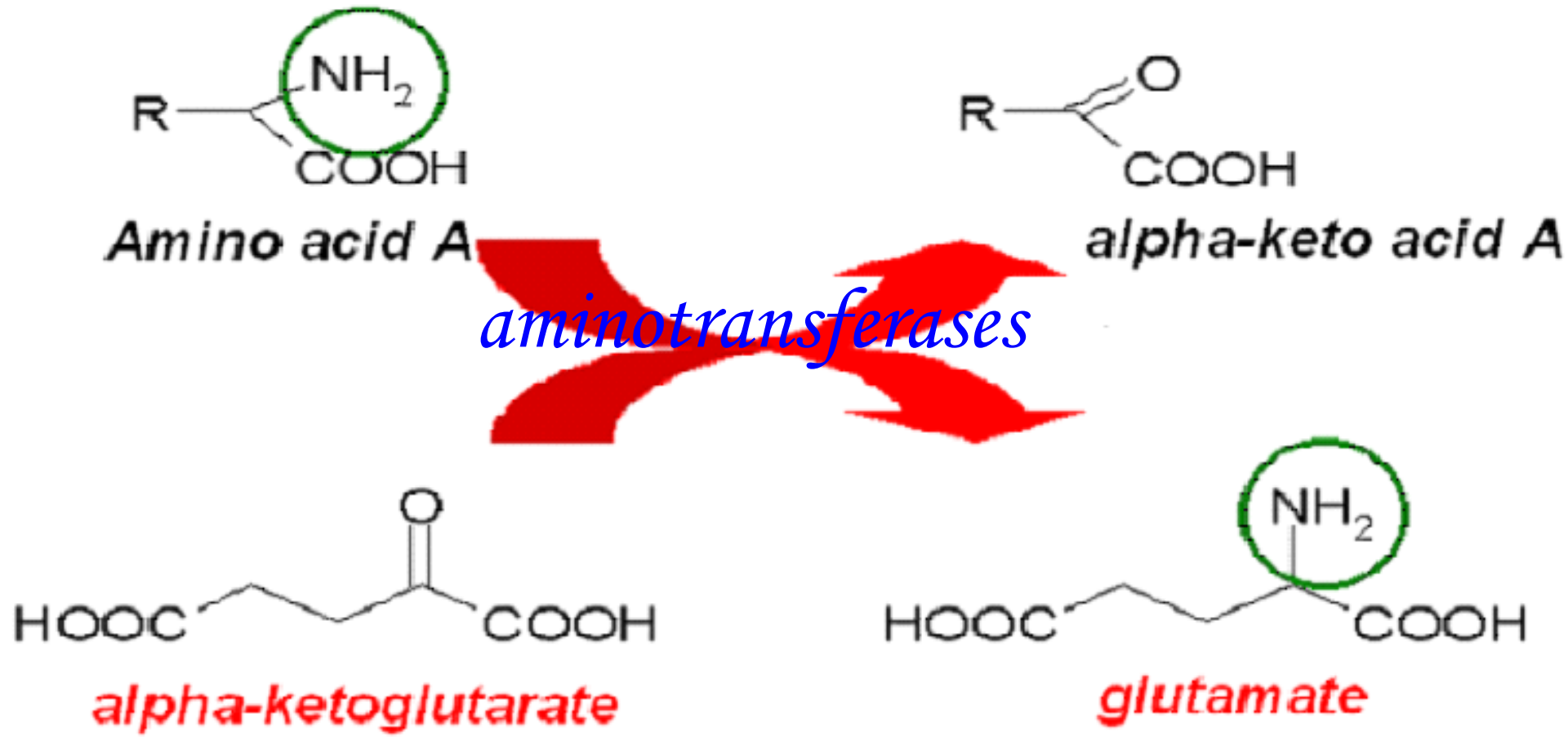
GOT and GPT

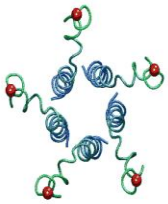
- **The most important transaminases identified are glutamate-oxaloacetate transaminase (GOT) glutamate- pyruvate transaminase (GPT).**

One of the thousands kinds of liver enzymes, and a kind of transferase

Large amount of transaminase is released into blood mostly on liver cell damages.
Thus, detection of serum level tells the existence of liver cell damage.

Transamination





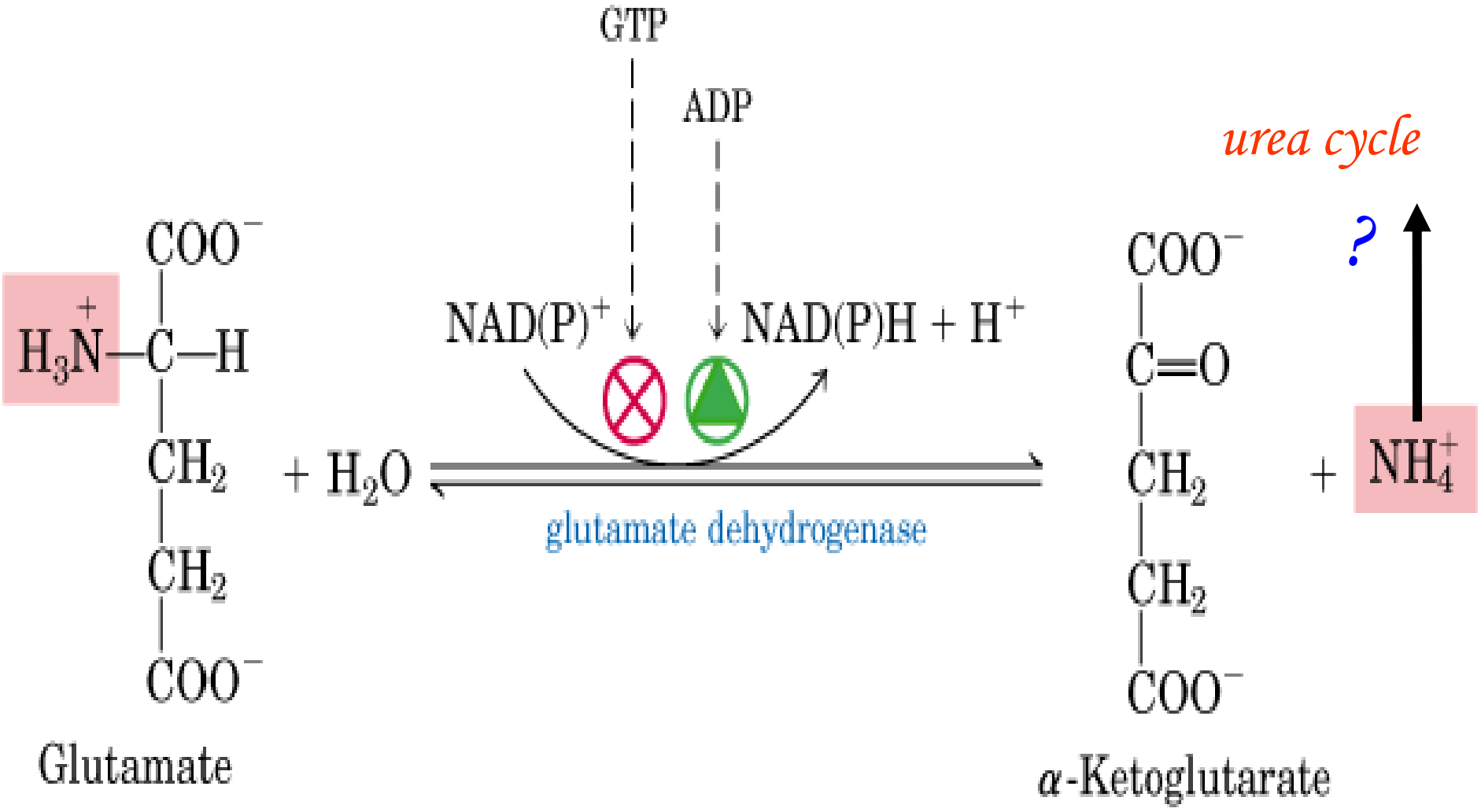
B. Oxidative Deamination

- L-glutamate dehydrogenase (in mitochondria)

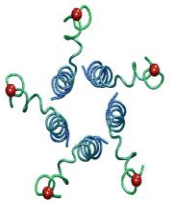
Glu + NAD^+ (or $NADP^+$) + H_2O ⇆ NH_4^+ + a-ketoglutarate + NAD(P)H +H^+

Requires NAD^+ or $NADP^+$ as a cofactor

Plays a central role in AA metabolism *?*

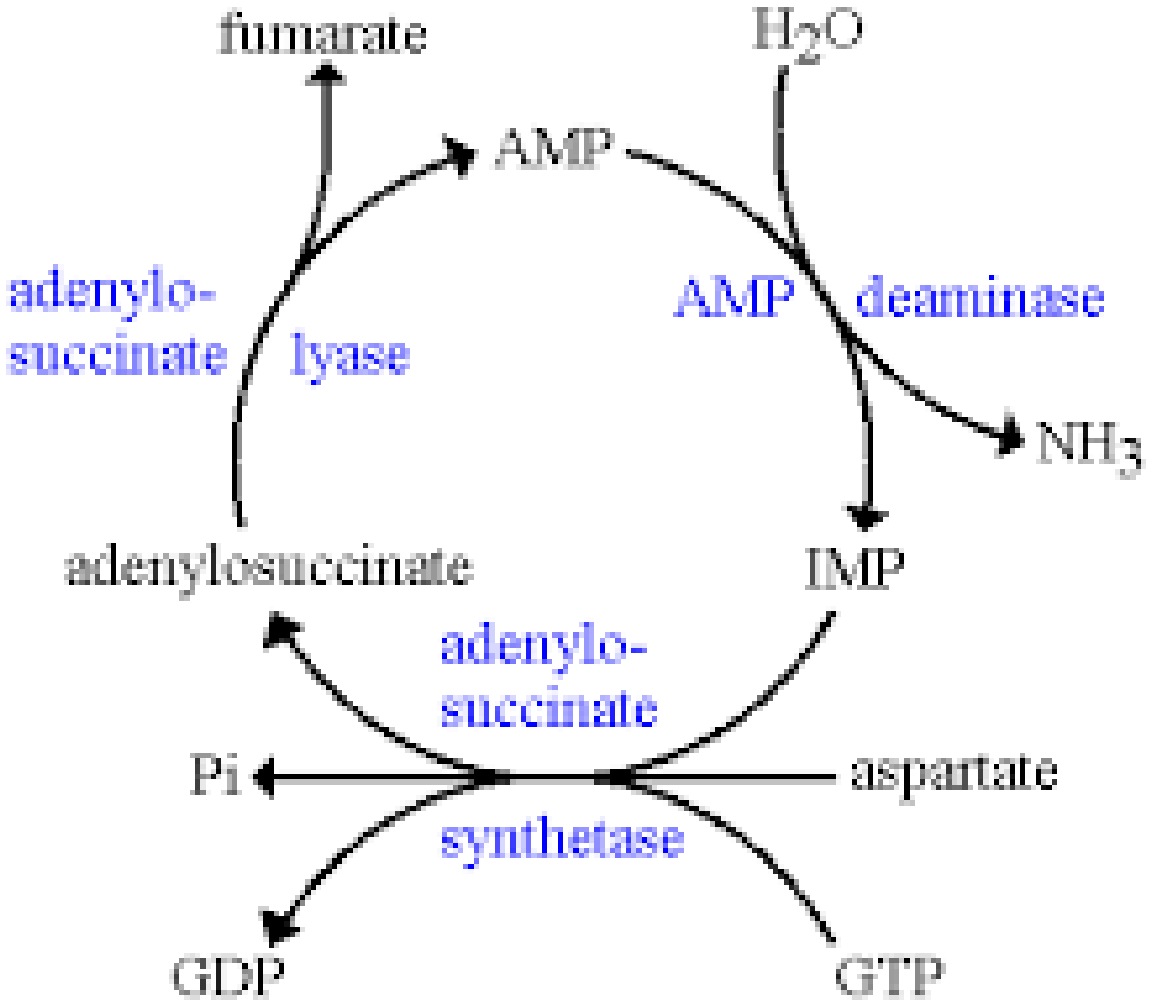


It is inhibited by GTP and ATP, and activated by GDP and ADP

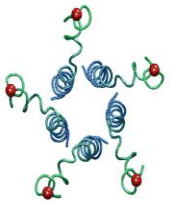


Glutamate Dehydrogenase

This enzyme is found in many tissues, where it catalyzes the reversible oxidative deamination of the amino acid glutamate. It produces the citric acid cycle intermediate α-ketoglutarate, which serves as an entry point to the cycle for a group of glucogenic amino acids. Its role in urea synthesis and nitrogen removal is still controversial, but has been included in Figure I-17-1.



C. purine nucleotide cycle



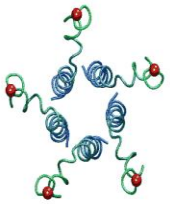
The metabolism of α-ketoacid

- Biosynthesis of nonessential amino acids

  TCA cycle member + amino acid

  →α-keto acid + nonessential amino acid
- A source of energy (10%)

  (CO_2+H_2O)
- Glucogenesis and ketogenesis

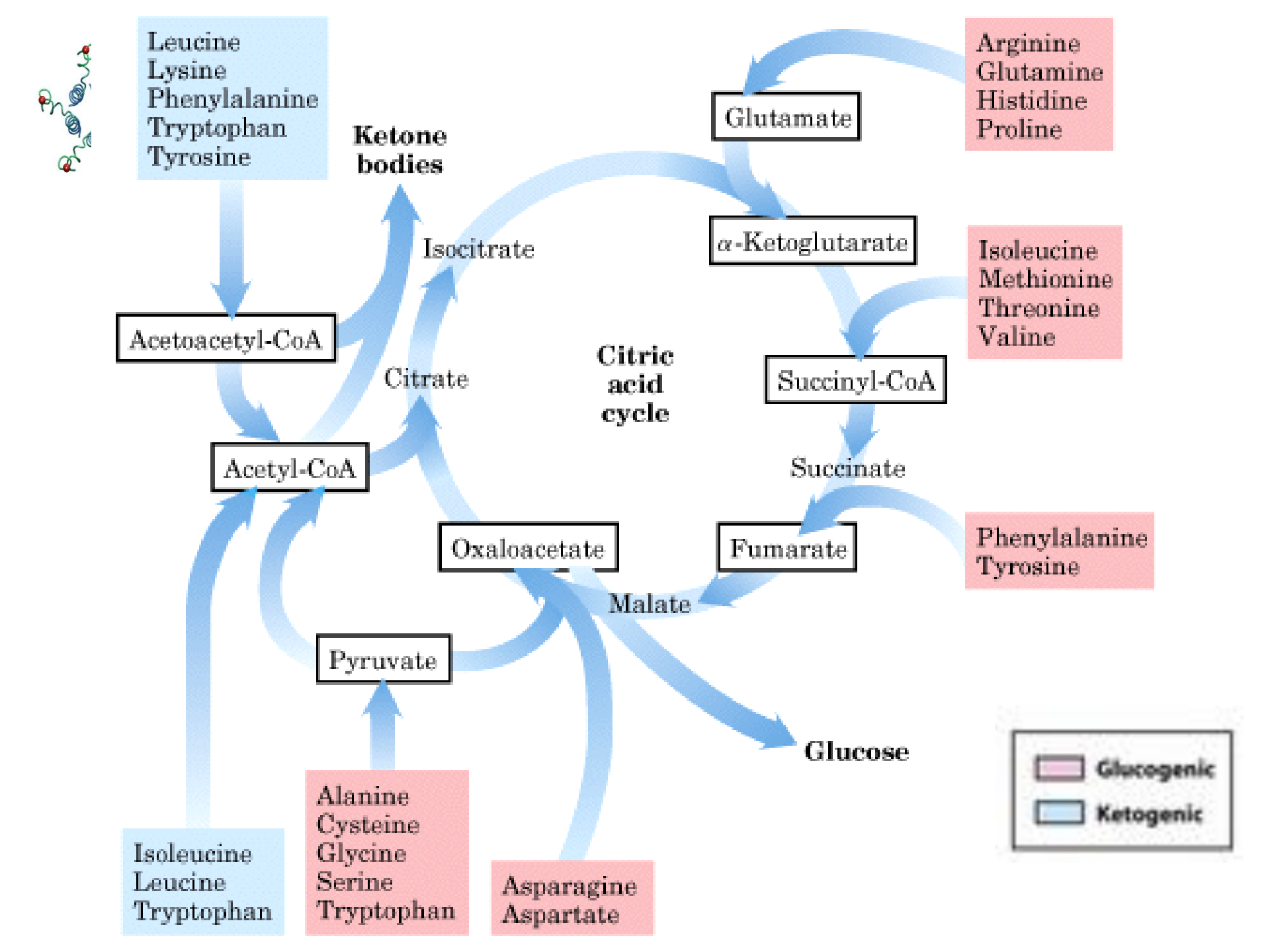


✯ *Classification of amino acids*

* glucogenic amino acid : are converted into either pyruvate or one of the citric acid cycle intermediates (a-ketoglutarate, succinyl CoA, fumarate or malate)

- ketogenic amino acid: will be deaminated via Acetylc-CoA and thus can be made into a ketone body. such as: Leucine and lysine

* glucogenic and ketogenic amino acid: isoleucine, phenylalanine, tryptophan and tyrosine, threonine

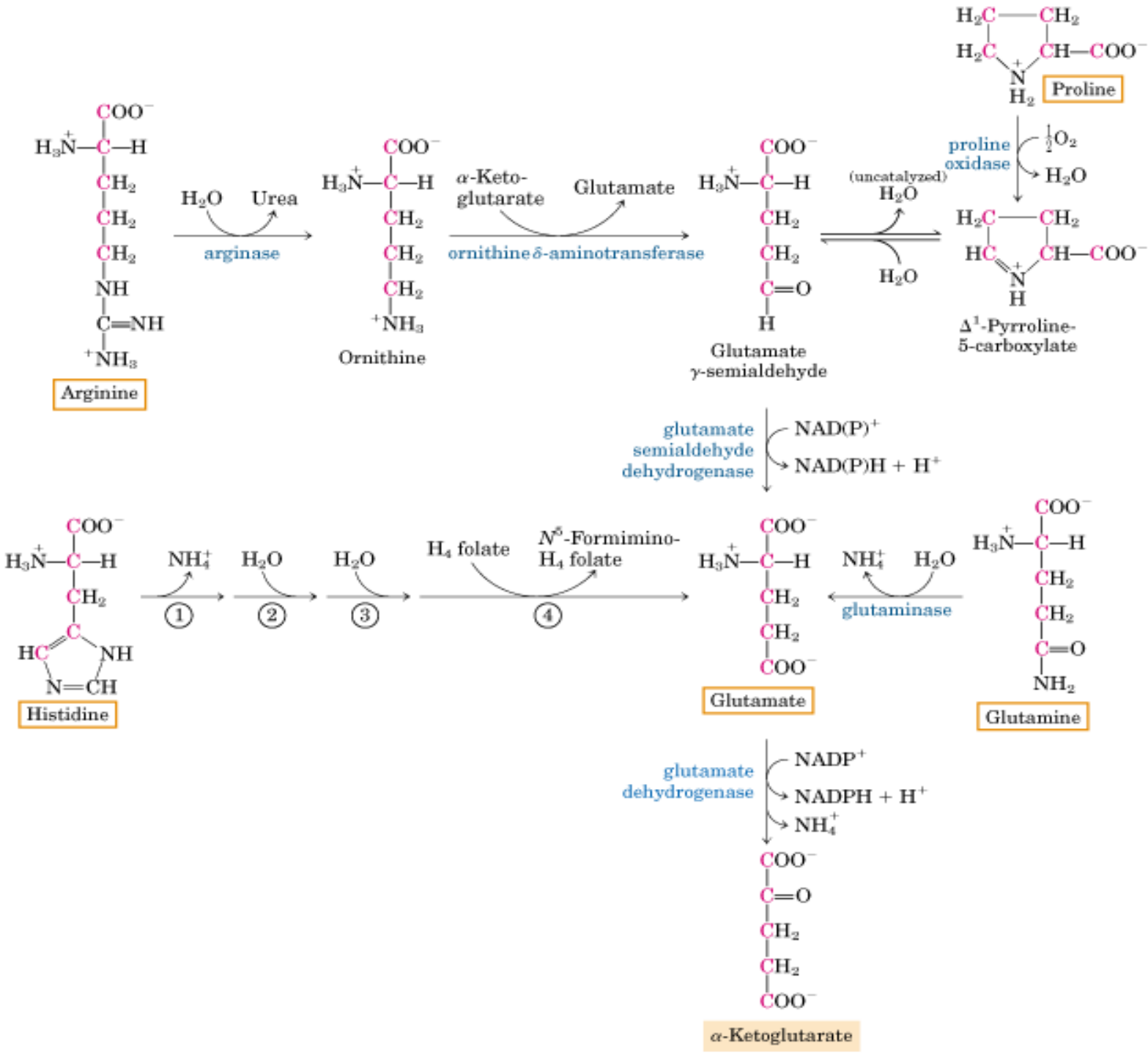

Leucine
Lysine
Phenylalanine
Tryptophan
Tyrosine
Ketone bodies
Glutamate
Arginine
Glutamine
Histidine
Proline
Isocitrate
α-Ketoglutarate
Isoleucine
Methionine
Threonine
Valine
Acetoacetyl-CoA
Citrate
Citric acid cycle
Succinyl-CoA
Acetyl-CoA
Succinate
Oxaloacetate
Fumarate
Phenylalanine
Tyrosine
Malate
Pyruvate
Glucose
Alanine
Cysteine
Glycine
Serine
Tryptophan
Isoleucine
Leucine
Tryptophan
Asparagine
Aspartate
Glucogenic
Ketogenic

Degradation of amino acids

- **Amino acid breakdown can yield:**
  - **Acetyl-CoA**
  - **α-KG(alpha keto glutarate**
  - **Succinyl-CoA**
  - **OAA(oxalacetate)**
  - **fumarate**

α-KG is generated from five amino acids

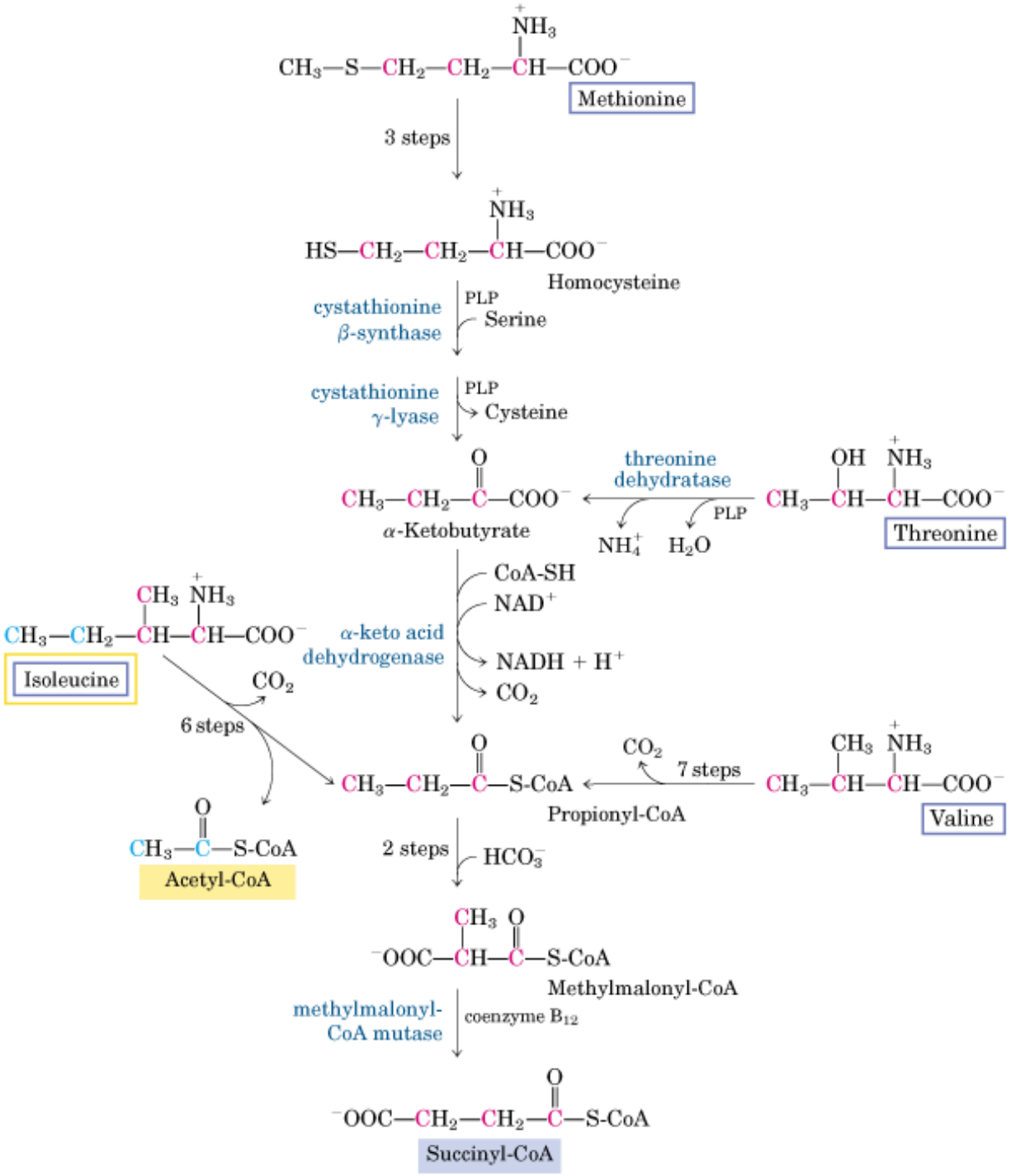
- Proline
- Glutamate
- Glutamine
- Arginine
- Histidine


Arginine
H2O
Urea
arginase
Ornithine
α-Keto-glutarate
Glutamate
ornithine δ-aminotransferase
Glutamate γ-semialdehyde
(uncatalyzed)
H2O
H2O
Δ1-Pyrroline-5-carboxylate
proline oxidase
½O2
H2O
Proline
glutamate semialdehyde dehydrogenase
NAD(P)+
NAD(P)H + H+
Histidine
NH4+
H2O
H2O
H4 folate
N5-Formimino-H4 folate
1
2
3
4
Glutamate
NH4+
H2O
glutaminase
Glutamine
glutamate dehydrogenase
NADP+
NADPH + H+
NH4+
α-Ketoglutarate

Four amino acids are converted to Succinyl-CoA

- Methionine
  - Converted to homocysteine through methyl group transfer, generates cysteine as converted to α-ketobutyrate
- Isoleucine
  - Transamination, oxidative decarboxylation to acetyl-CoA and propionyl CoA
- Valine
  - Transamination, decarboxylation to propionyl CoA
- Threonine
  - α-ketobutyrate generated and converted to propionyl CoA

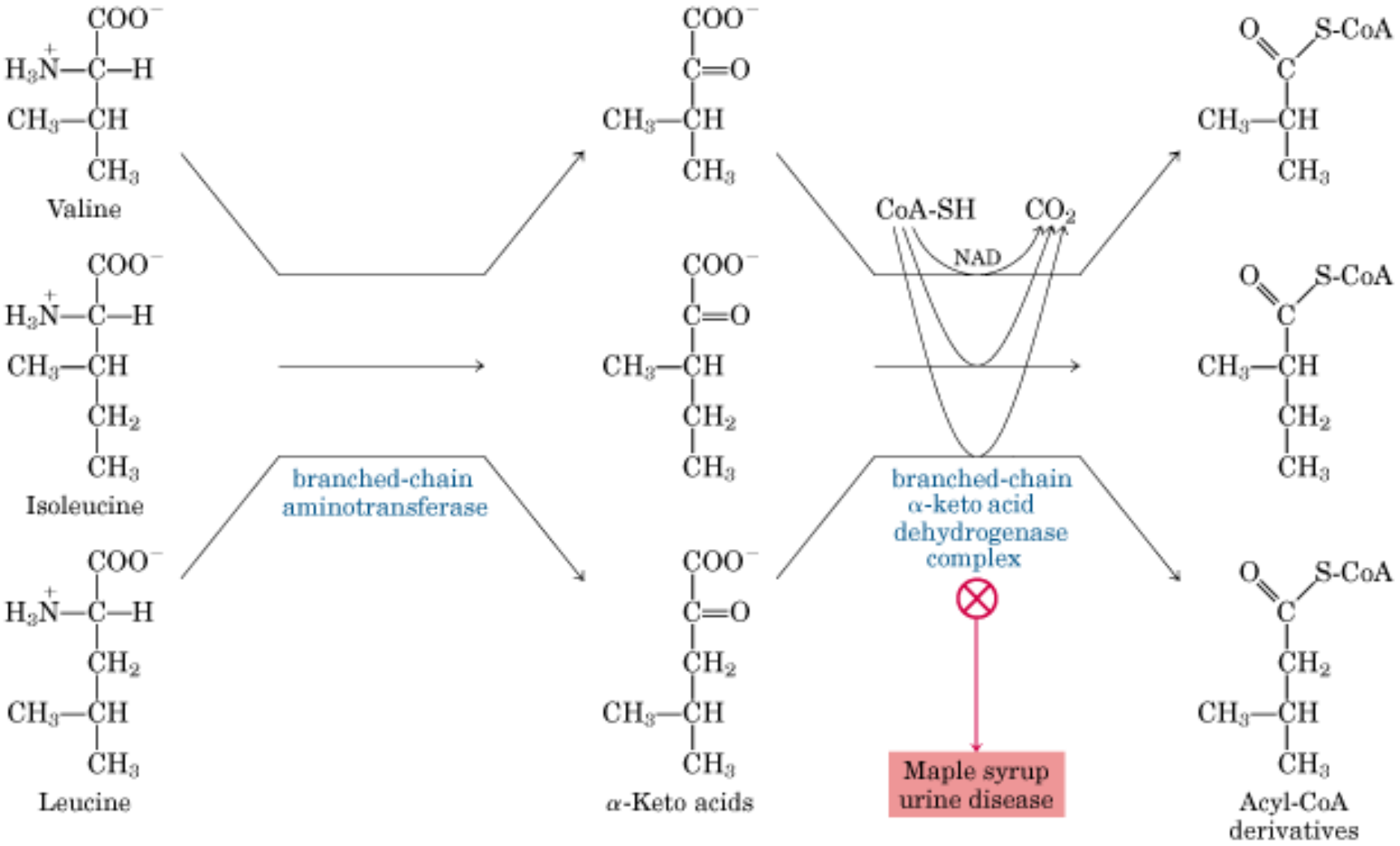
Propionyl-CoA is a common intermediat for amino acids → succinyl-CoA



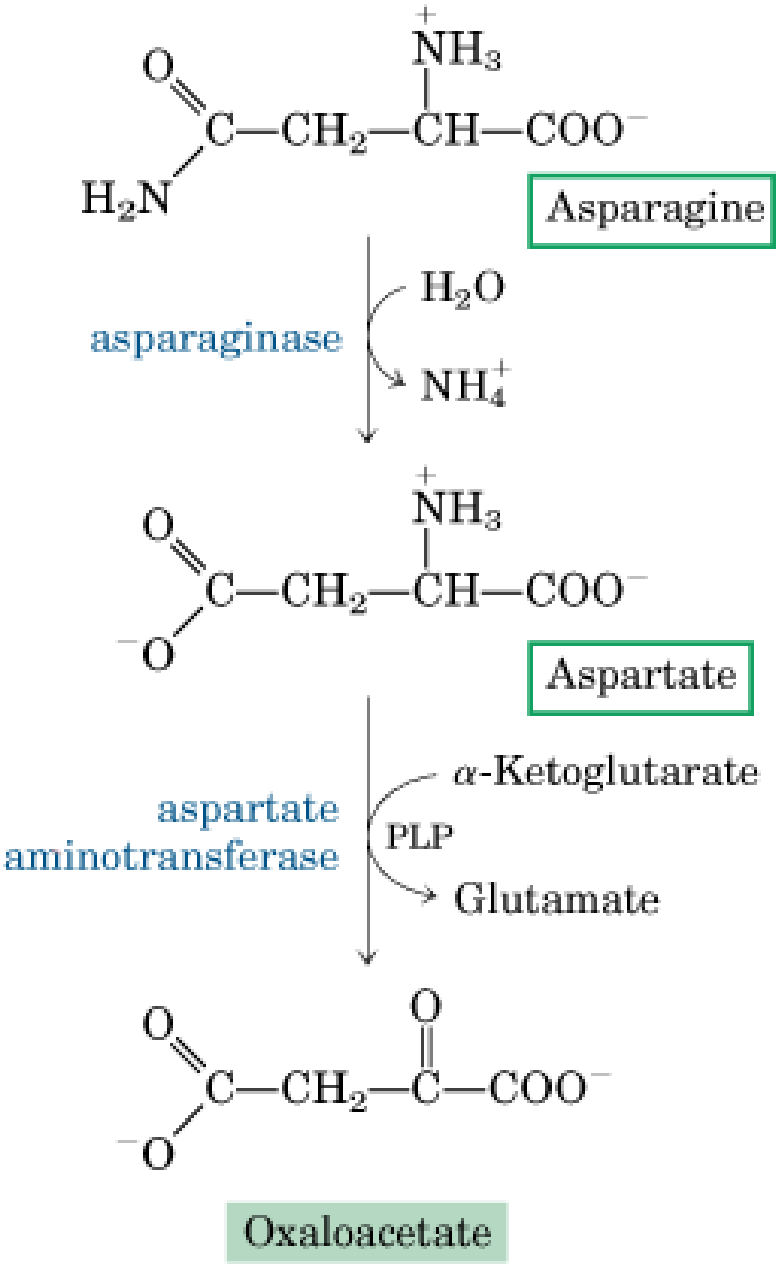
Branched-chain α-keto acid dehydrogenase complex

- In certain body tissues, this enzyme catalyzes the oxidative decarboxylation of valine, isoleucine, and leucine yielding CO_2, and acyl-CoA derivatives.
- Shares ancestry with pyruvate dehydrogenase complex, α-KG dehydrogenase complex – another example of gene duplication

Branched-chain …complex



Asparagine and aspartate are degraded to OAA

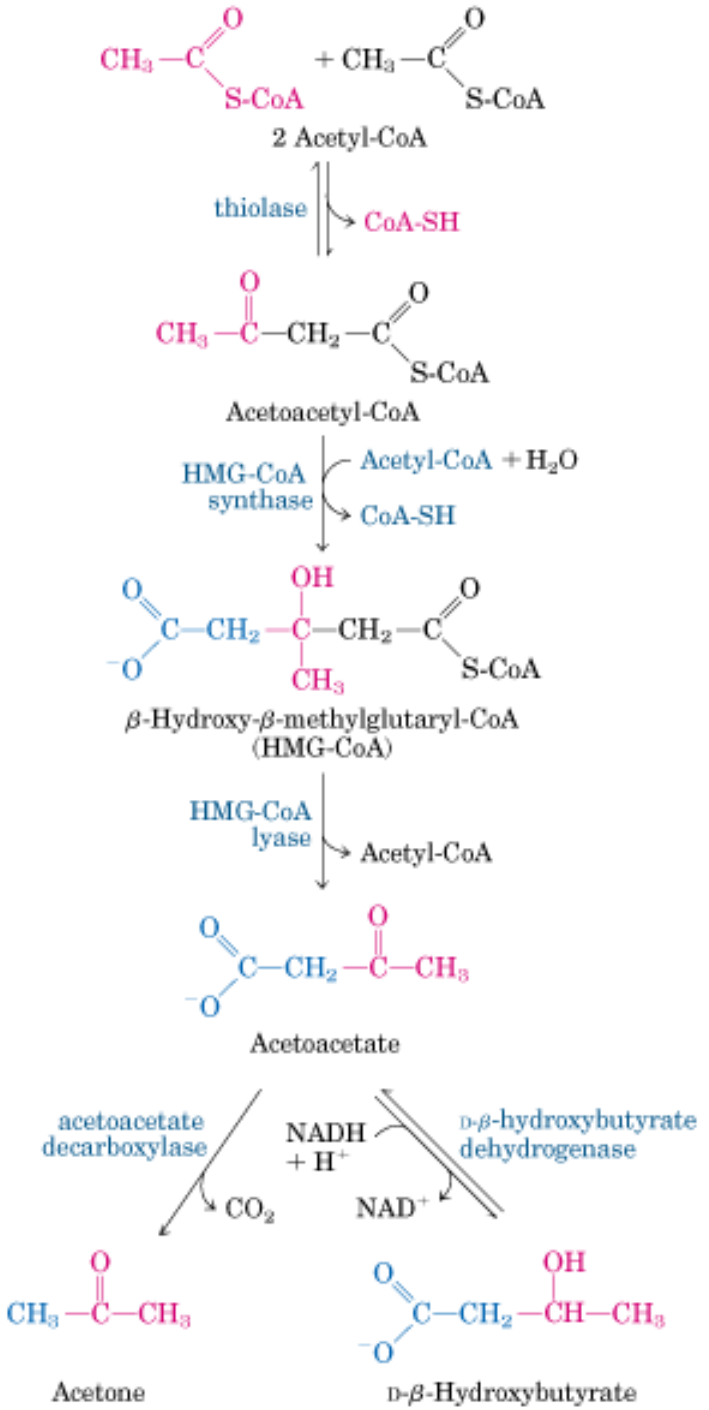


Fate of metabolites derived from amino acids

- In addition to feeding the citric acid cycle, amino acids can result in ketone bodies, while others are gluconeogenic

Ketone bodies

- The six amino acids that are degraded to acetoacetyl-CoA and/or acetyl-CoA) can be converted to acetoacetate and β-hydroxybutyrate



Glucogenic amino acids

- Amino acids that are degraded to pyruvate, α-KG, succinyl-CoA fumarate, and/or OAA can be converted to glucose

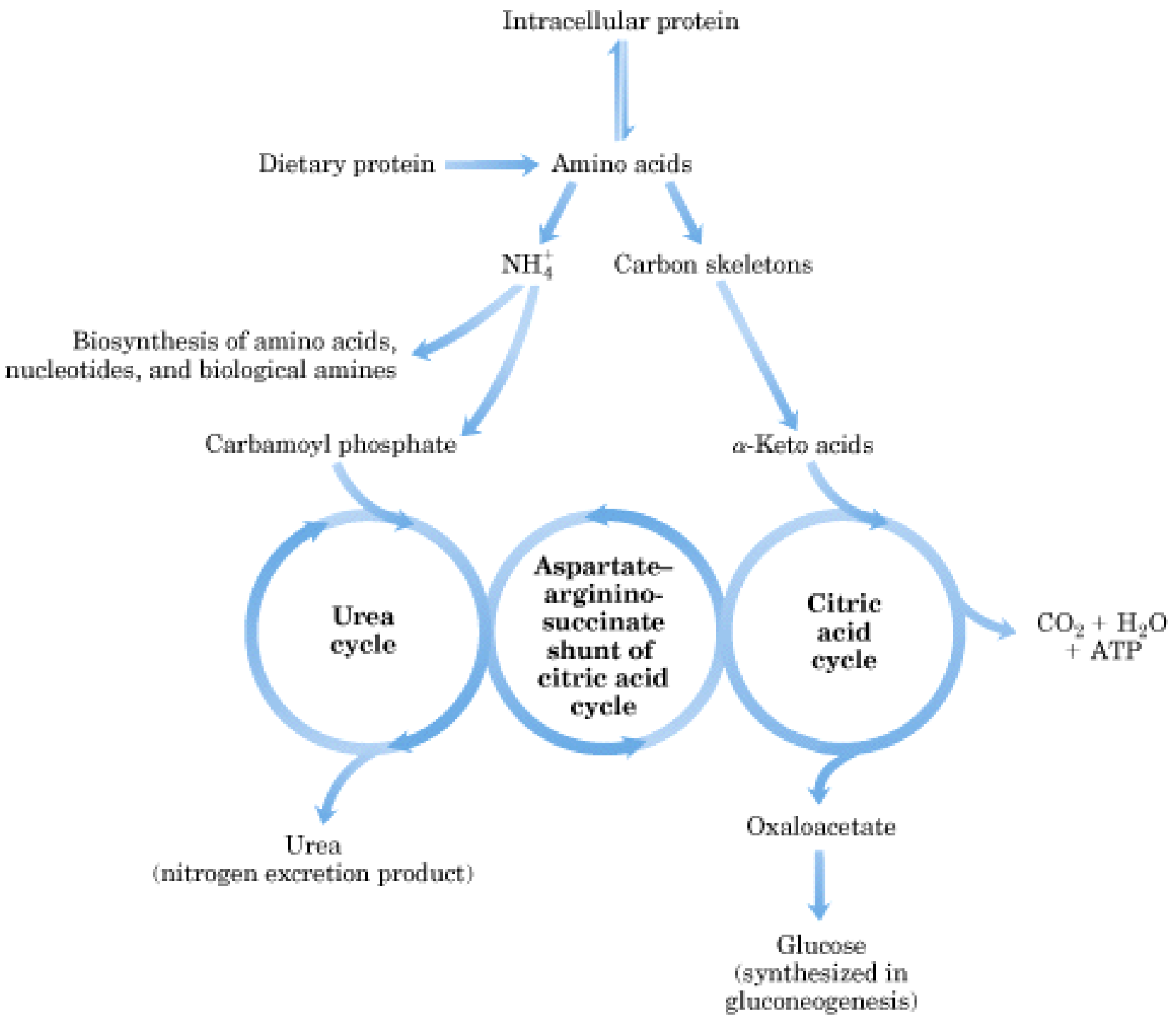
table 20–3

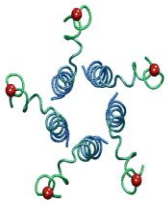
Glucogenic Amino Acids, Grouped by Site of Entry*

Pyruvate	**Succinyl-CoA**
Alanine	Isoleucine†
Cysteine	Methionine
Glycine	Threonine
Serine	Valine
Tryptophan†	
α-Ketoglutarate	**Fumarate**
Arginine	Phenylalanine†
Glutamate	Tyrosine†
Glutamine	**Oxaloacetate**
Histidine	Asparagine
Proline	Aspartate

*These amino acids are precursors of blood glucose or liver glycogen because they can be converted to pyruvate or citric acid cycle intermediates. Only leucine and lysine are unable to furnish carbon for net glucose synthesis.

†These amino acids are also ketogenic (see Fig. 18–19).


Intracellular protein
Dietary protein
Amino acids
NH_4^+
Carbon skeletons
Biosynthesis of amino acids, nucleotides, and biological amines
Carbamoyl phosphate
α-Keto acids
Urea cycle
Aspartate–arginino-succinate shunt of citric acid cycle
Citric acid cycle
$CO_2 + H_2O$ + ATP
Urea
(nitrogen excretion product)
Oxaloacetate
Glucose
(synthesized in gluconeogenesis)



Ammonia is toxic, so cells need to get rid of it…..

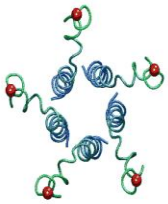
Sources:

1. *amino acids degradation*

$$RCCO_2H_2NH_2 \xrightarrow{MAO} RCHO + NH_3$$

Monoamine oxidase

2. *glutamine (glutaminase, kidney)*
3. *catabolism from bacteria in intestine (two)*
4. *purine and pyrimidine catabolism*



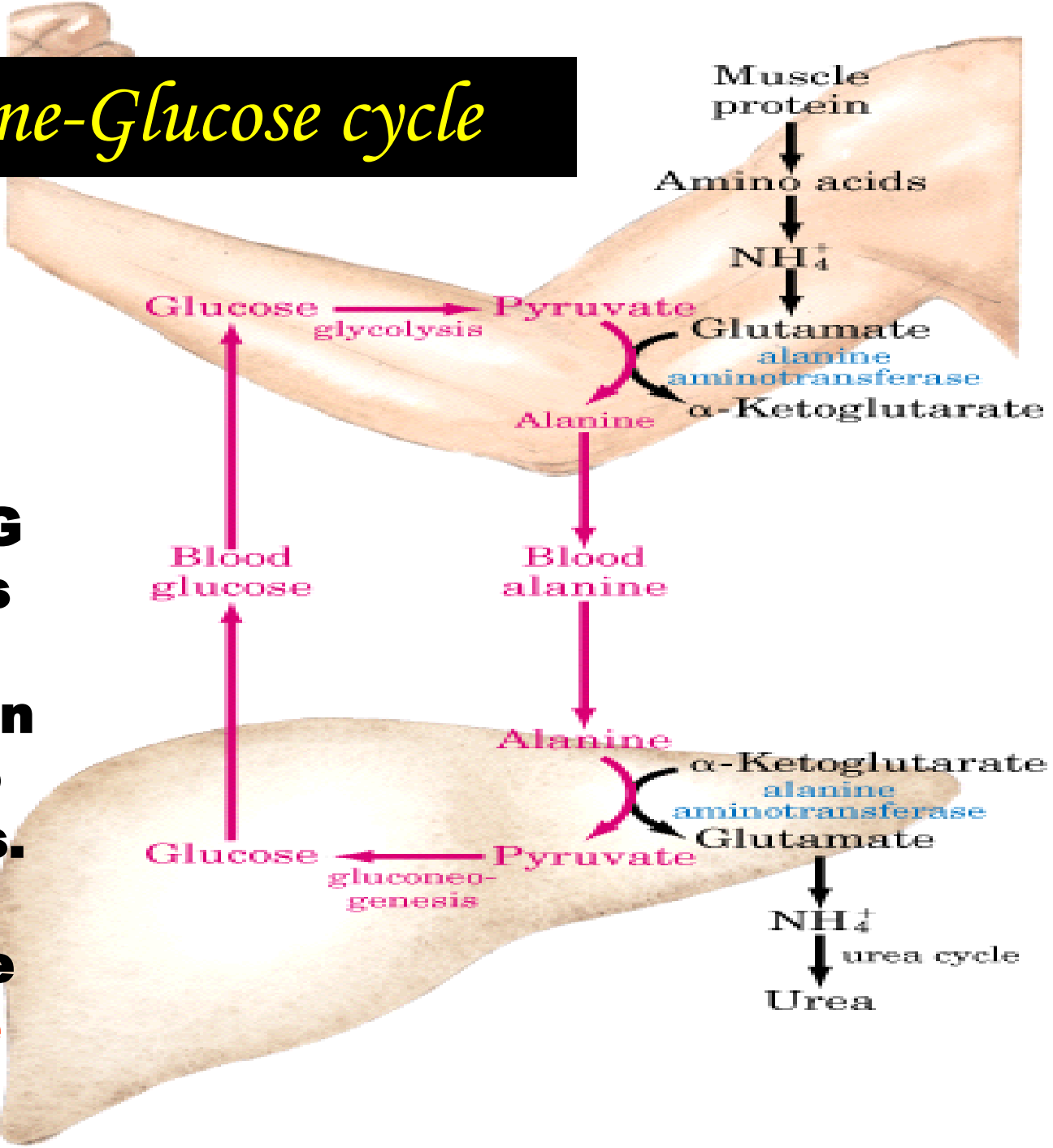
Metabolism of ammonia

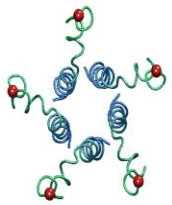
- Fix ammonia onto glutamate to form glutamine(Gln) and use as a transport mechanism
- Transport ammonia by alanine-glucose cycle and Gln regeneration
- Excrete nitrogenous waste through urea cycle

- *Transport of ammonia*
- *alaninie - glucose cycle* ✭
- *regenerate Gln*

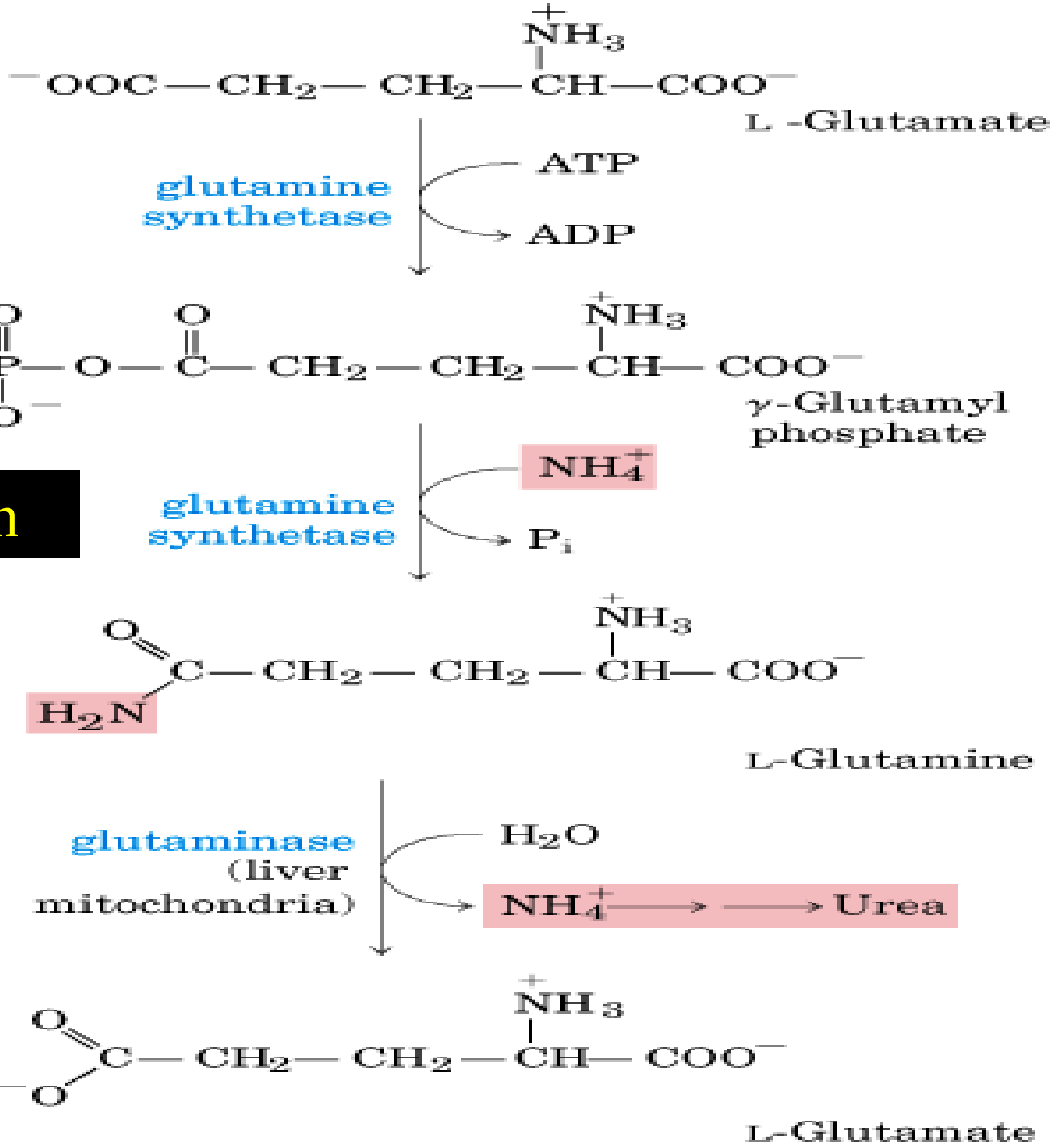
Alanine-Glucose cycle

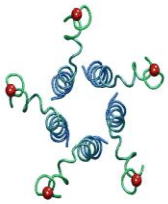
- **In the liver *alanine* transaminase tranfers the ammonia to α-KG and regenerates pyruvate. The pyruvate can then be diverted into gluconeogenesis. This process is refered to as the glucose-alanine cycle**





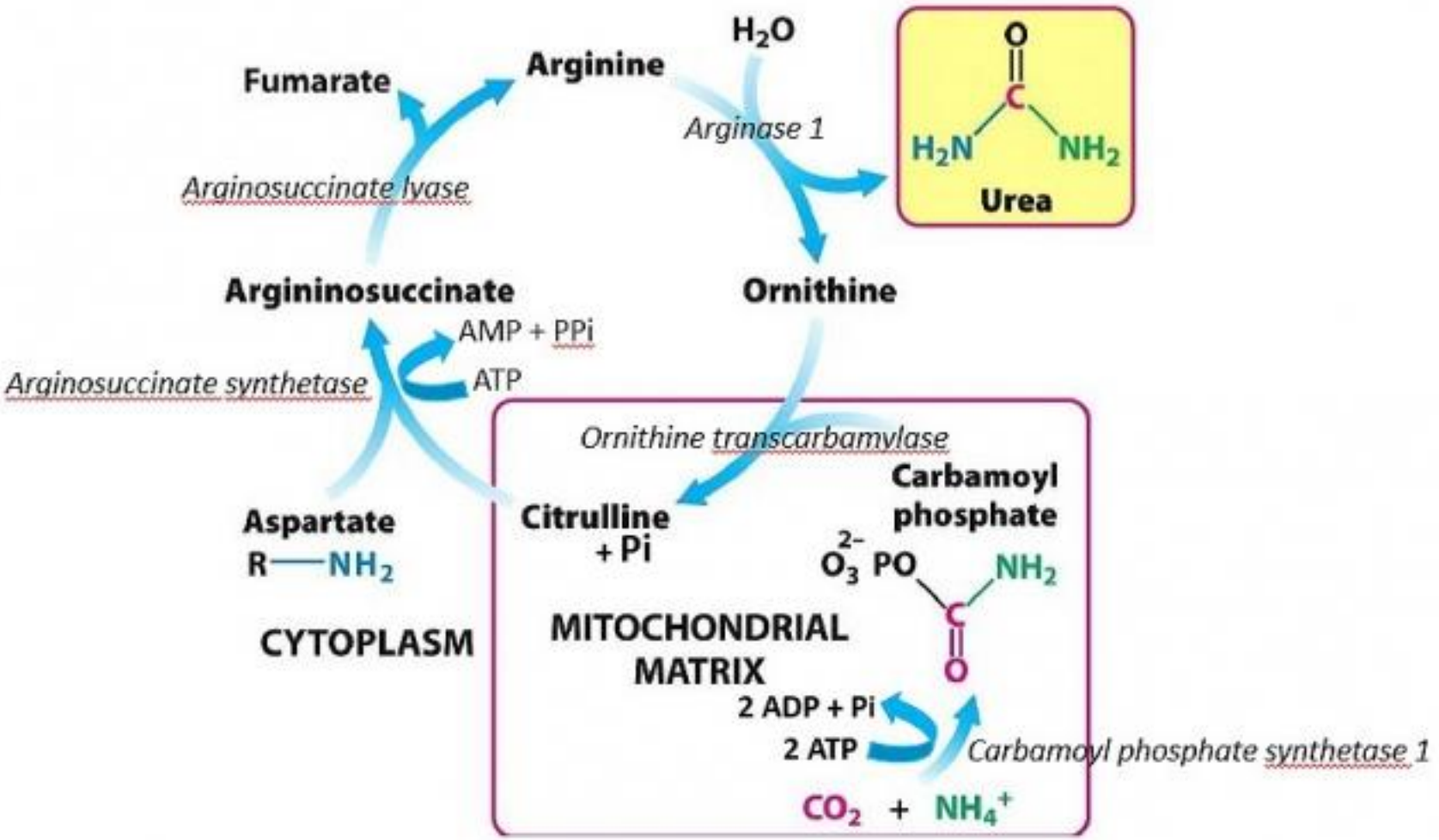
Gln regeneration





★★★★ Urea synthesis

- *Synthesis in liver (Mitochondria and cytosol)*
- *Excretion via kidney*
- *To convert ammonia to urea for final excretion*


Fumarate
Arginine
H_2O
Arginase 1
O
C
H_2N
NH_2
Urea
Argininosuccinate lyase
Argininosuccinate
Ornithine
AMP + PPi
ATP
Argininosuccinate synthetase
Ornithine transcarbamylase
Carbamoyl phosphate
Citrulline + Pi
O_3PO^{2-}
NH_2
Aspartate
R—NH_2
CYTOPLASM
MITOCHONDRIAL MATRIX
2 ADP + Pi
2 ATP
Carbamoyl phosphate synthetase 1
CO_2 + NH_4^+

urea cycle

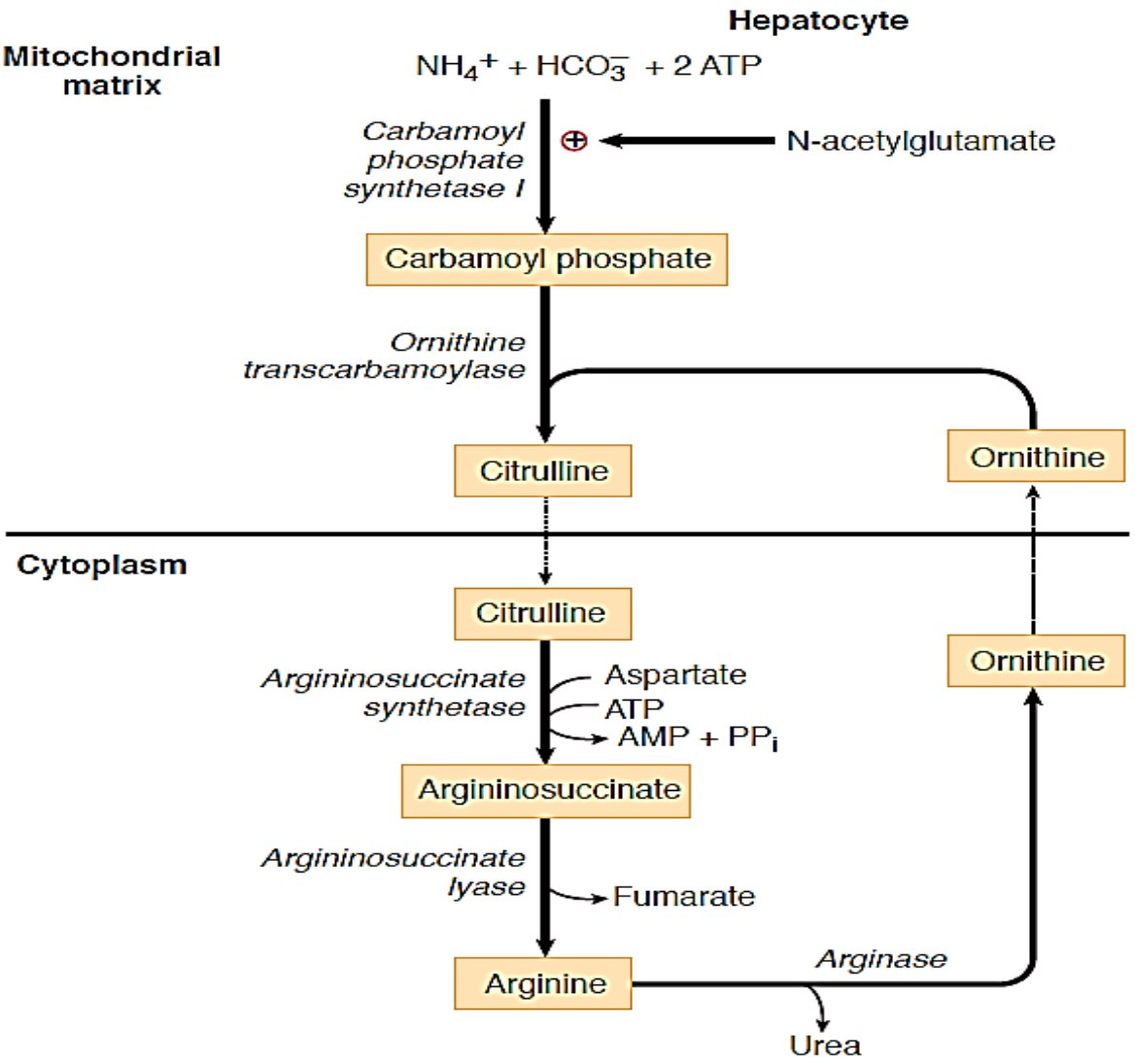
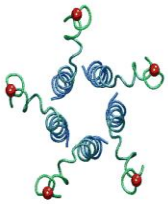


Figure I-17-2. The Urea Cycle in the Liver



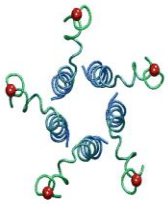
UREA CYCLE (liver)

1. Overall Reaction:

NH_3 + HCO_3^- + aspartate + 3 ATP + H_2O → urea + fumarate + 2 ADP + 2 Pi + AMP + ppi

2. Requires 5 enzymes:

2 from mitochondria and 3 from cytosol



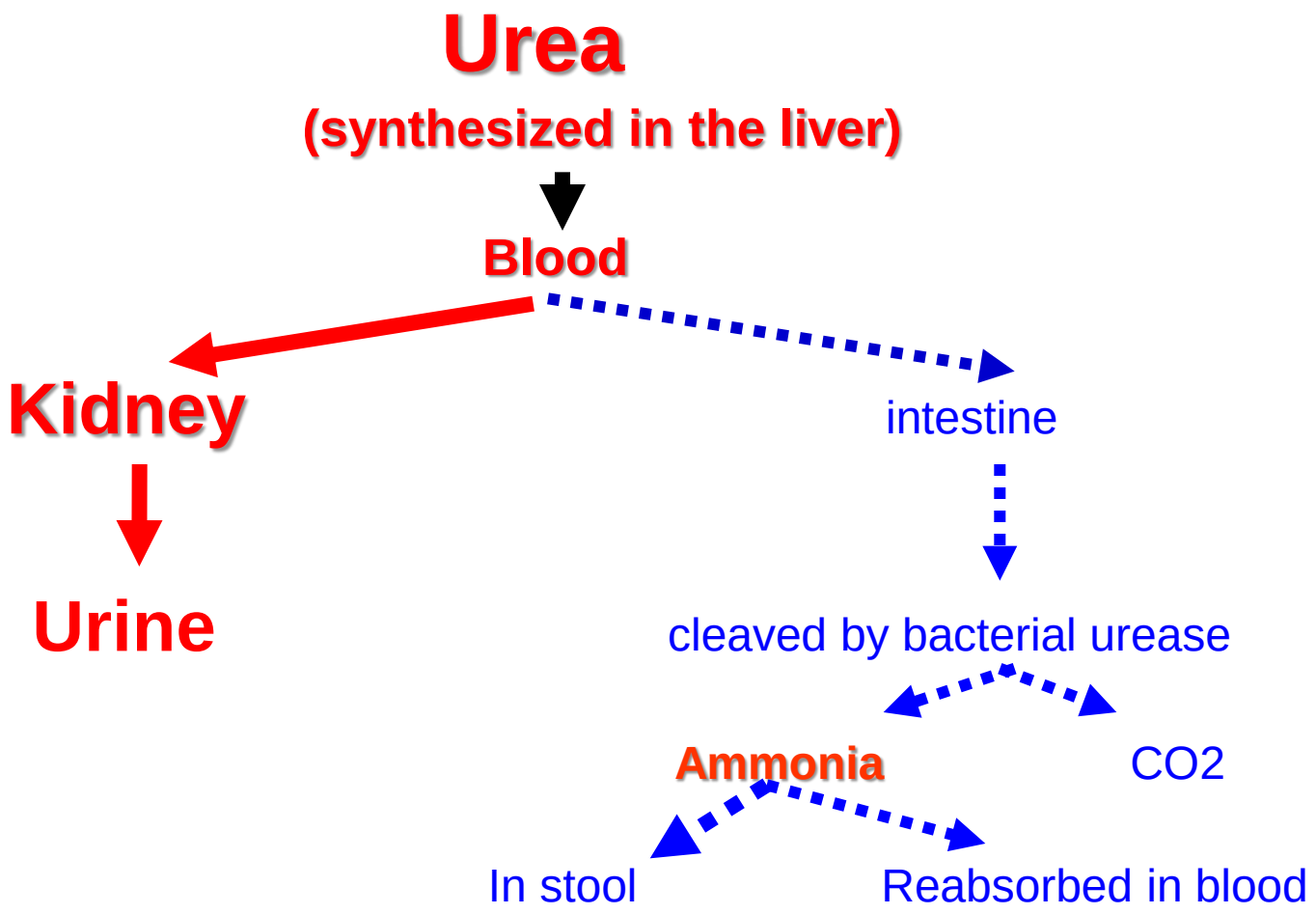
Regulation of urea cycle

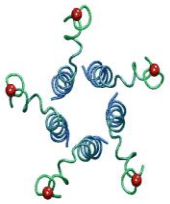
1.Mitochondrial carbamoyl phosphate synthetase I (CPS I)

CPS I catalyzes the first committed step of the urea cycle

CPS I is also an allosteric enzyme sensitive to activation by N-acetylglutamate（AGA） which is derived from glutamate and acetyl-CoA

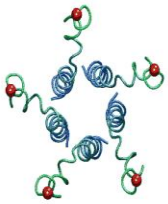
Fate of Urea


Urea
(synthesized in the liver)
Blood
Kidney
Urine
intestine
cleaved by bacterial urease
Ammonia
CO2
In stool
Reabsorbed in blood



Increased rate of AA degradation requires higher rate of urea synthesis

↑ AA degradation → ↑ glutamate concentration → ↑ synthesis of N-acetylglutamate → ↑ CPS I activity → ↑ urea cycle efficiency

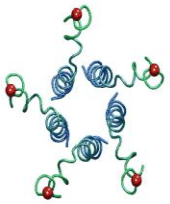


2. *All other urea cycle enzymes are controlled by the concentrations of their substrates*

Deficiency in an E → ↑(substrate) →↑rate of the deficient E

3. *The intake of the protein in food*

the intake ↑ →↑urea synthesis



Hyper-ammonemia and the toxic of the ammonia

Why is ammonia toxic?

$$\alpha\text{-ketoglutarate} + NH_3 + NADPH \xleftrightarrow{GDH} \text{glutamate} + NADP^+$$

High ammonia depletes the TCA cycle of α-ketoglutarate → low ATP →

COMA (a symptom of high ammonia levels).

- *Hyperammononemia: ammonia intoxication - tremors, slurring of speech, and blurring of vision, coma/death*
- *Cause by cirrhosis of the liver or genetic deficiencies*

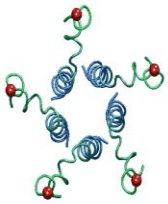


Table I-17-1. Genetic Deficiencies of Urea Synthesis

Carbamoyl Phosphate Synthetase	Ornithine Transcarbamoylase
↑ $[NH_4^+]$; hyperammonemia	↑ $[NH_4^+]$; hyperammonemia
Blood glutamine is increased	Blood glutamine is increased
BUN is decreased	BUN is decreased
No orotic aciduria Autosomal recessive	Orotic aciduria X-linked recessive
Cerebral edema	Cerebral edema
Lethargy, convulsions, coma, death	Lethargy, convulsions, coma, death

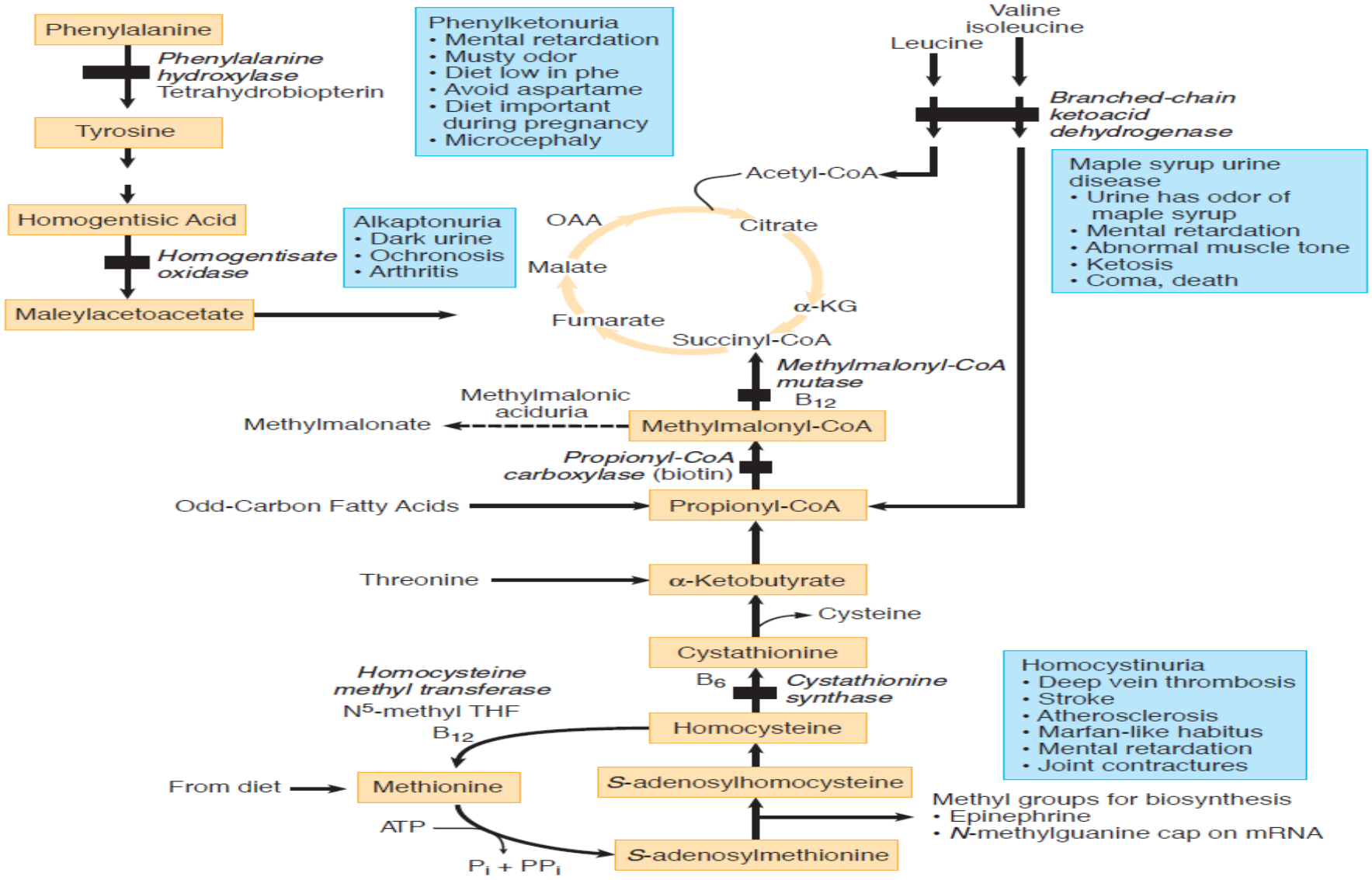


Figure I-17-3. Genetic Deficiencies of Amino Acid Metabolism