Chapt. 37, 38, 39 Nitrogen metabolism, Amino acids, Proteins

Student Learning Outcomes:
- Describe digestion of proteins, absorption of amino acids in intestine and transport of through blood
- Describe some compounds made from amino acids
- Describe role of intracellular proteases, proteasome in recycling proteins
- Explain the essentials of the urea cycle for elimination of nitrogen – fed vs. fasting state
- Describe synthesis of nonessential aa and pathways for degradation
- Describe some genetic errors of aa metabolism

VII. Nitrogen metabolism overview

- Dietary proteins digested to aa
- Transported through blood to cells
- Amino acids absorbed, used to make proteins, other N-containing (hormones, purines, creatine, heme)
- C skeleton used for energy; enters TCA, fatty acids, stored glycogen
- NH$_3$ is toxic;
- Excess N to urea (liver), excreted
- Recall Ch. 2 fed vs. fasting

Fig. VII.1 amino acid metabolism

Major nitrogenous urinary excretory products

<table>
<thead>
<tr>
<th>Table VII.1 Amount N excreted urine per day</th>
</tr>
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<tbody>
<tr>
<td>• Urea 12-20 g urea N (12,000-20,000 mg)</td>
</tr>
<tr>
<td>• NH$_3$+ 140-1500 mg ammonia N</td>
</tr>
<tr>
<td>• Creatinine men 14-26 mg/kg; women 11-20 mg/kg</td>
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<tr>
<td>(Creatine PO$_4$ is energy storage in muscle cells)</td>
</tr>
<tr>
<td>• Uric acid 250-750 mg</td>
</tr>
<tr>
<td>(from breakdown purines – Fig. 24.20)</td>
</tr>
</tbody>
</table>

Essential amino acids and Synthesized

<table>
<thead>
<tr>
<th>Essential Amino acids</th>
<th>Synthesized in body</th>
</tr>
</thead>
<tbody>
<tr>
<td>Histidine</td>
<td>Alanine</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>Arginine</td>
</tr>
<tr>
<td>Leucine</td>
<td>Asparagine</td>
</tr>
<tr>
<td>Lysine</td>
<td>Aspartate</td>
</tr>
<tr>
<td>Methionine</td>
<td>Cysteine</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>Glutamate</td>
</tr>
<tr>
<td>Threonine</td>
<td>Glutamine</td>
</tr>
<tr>
<td>Tryptophan</td>
<td>Glycine</td>
</tr>
<tr>
<td>Valine</td>
<td>Proline</td>
</tr>
<tr>
<td>Arginine (not for adult)</td>
<td>Serine</td>
</tr>
<tr>
<td>Tyrosine (from Phe)</td>
<td></td>
</tr>
</tbody>
</table>
Overview nitrogen metabolism

VII. Metabolism Glucose, Fatty acids, amino acids:
• Removal of $\text{--NH}_2$ takes Carbon skeleton into other paths

VII.2 Overview Nitrogen metabolism:
(no cell has all these pathways)

Ch. 37 Protein digestion

37. Overview protein digestion: amino acid absorption
• Diverse proteases –zymogens
• Activated by cleavage in GI
  {pro- and –ogen}
  Pepsin from stomach
  – acid denatures target proteins
  Pancreatic enzymes, buffer ↑ pH
  *Trypsin is key
• Amino acids transported across intestinal cell membranes

Activation of gastric, pancreatic zymogens

Activation of gastric, pancreatic zymogens:
• Pepsinogen self-cleavage in stomach;
• Trypsinogen cleaved in intestine to trypsin
• Trypsin cleaves other pancreatic
• They can digest each other
• Anti-trypsin inhibitor in pancreatic cells

Digestive proteases

Action of digestive proteases:
• Serine proteases
• Cleave different specificities
• Endopeptidases internal
• Exopeptidases from end
• Enzymes will digest each other
Transepithelial transport of amino acids:
- **Na\(^+\)**-dependent cotransport into cells
  - Semi-specific for categories of amino acids
- **Na\(^+\)/K\(^+\)** ATP active transport pump removes **Na\(^+\)**
- Facilitated transporter into blood
- Facilitated transporters into cells of different tissues
- Liver, muscle have Na\(^+\)-dependent transporters into cells also

Protein turnover, replenishment:
- Proteins have variable half-lives – minutes to days in cells
- Enzymes in cytoplasm, lysosome recycle to amino acids
- Cells of digestive system turn over rapidly
- Hemoglobin from old RBCs recycled in macrophages
- Muscle protein degraded during fasting

**Some proteases:**
- Cathepsins  Cysteine protease   lysosomal enzymes
- Caspases  cysteine protease   apoptosis (ch. 18)
- Serine proteases   digestion, blood clotting
- Proteasome large complex   Ub-protein turnover

Proteases degrade proteins:
- Ubiquitin is 76-aa protein; ligases attach it to targets
- Ubiquitination (poly-) signals destruction
- Proteasome large complex:
  - ATP unfolds target
  - Ub is recycled

Review question Ch. 37:  (see p. 706)
Kwashiorkor can result from which of the following?
A. Consuming calorie-deficient diet that is also deficient in protein
B. Consuming calorie-adequate diet that is deficient in carbohydrates
C. Consuming calorie-adequate diet that is deficient in fatty acids
D. Consuming calorie-adequate diet that is deficient in proteins
E. Consuming calorie-deficient diet that is primarily proteins
Key concepts

**Key concepts Ch. 37-38:**
- Humans can synthesize 11 of 20 amino acids
  - Others are essential in the diet
- Amino acid metabolism uses cofactors PLP, others
- Dietary nonessential aa made from glycolytic intermediates or from existing aa
- Amino acids are degraded to urea; Carbon skeleton is glucogenic or ketogenic
- Defects in aa degradation cause disease
- Defects Phe or Tyr metabolism → PKU, alkaptonuria, albinism

Chapt. 38

**Ch. 38 Nitrogen metabolism, urea cycle**

**Student Learning Outcomes:**
- Describe complex metabolism of amino acids:
  - Nitrogen balance is critical; NH$_3$ is toxic;
  - Normal diet: most aa for synthesis of proteins,
    - extra amino acids are used for energy, gluconeogenesis
  - Fasting: recycle muscle protein to aa
- Explain importance of enzymes that interconvert amino acids:
  - Transaminases, dehydratases, glutaminase, glutamate dehydrogenase

**Fate of amino acid Carbon, Nitrogen: urea cycle**
- C for energy storage (glycogen, fatty acids)
- C for energy (TCA, e-transport)
- N goes to liver for urea:
  - 1 from NH$_4$$^+$
  - 1 from Aspartate

**Amino acid metabolism:**

**Fed:**
- Liver makes blood proteins
- Excess amino acids → glucose, fatty acids, TAG
- Amino acids to other cells

**Fasting:**
- Muscle release aa
- N transported in Gln,
- N transported in Ala (gluconeogenic)
Fate of amino acid Nitrogen

Fate of amino acid nitrogen: Transamination removes N:
Reversible reactions:
PLP = pyridoxal PO_4 cofactor

Used in synthesis, degradation of amino acids
All aa except Lys, Thr do this
Glutamate and Asp common

Fig. 38.3,4
Transamination
PLP derived from vitamin B_6

B. Removal of aa N as ammonia

B. Removal of aa Nitrogen as ammonia:
• Sources of NH_4^+ for urea cycle:
  • Deaminations, deamidations
  • Many aa release NH_4^+
• Reactions irreversible:
  • Except GDH reversible:
• Liver makes urea

\[ \text{NH}_4^+ \rightarrow \text{NH}_3 + H^+ \]
\[ pK_a = 9.3 \]

Fig. 38.5 Mostly NH_4^+ physiological pH
NH_3 toxic, diffuses across membranes; NH_4^+ not cross membranes

Reversible Glutamate dehydrogenase

Reversible Glutamate dehydrogenase
• Mitochondria
• Glu collects N by transamination with other aa
• Glu releases NH_4^+ with GDH
• GDH can ‘fix’ NH_4^+ into organic molecules

C. Glutamate is key

C. Glutamate is key to metabolism of amino acid nitrogen:
• For synthesis, degradation
  • Glu formed from α-kG:
    • By transamination
    • By GDH with NADPH, NH_4^+

Fig. 38.8
PLP is pyridoxal PO_4
**Glutamate is Key for urea production**

Glutamate collects N from other amino acids

Glutamate releases NH$_4^+$ by GDH

Aspartate also critical for urea production

Asp can get its N from Glu

**D. Ala and Gln transport aa in blood**

D. Alanine and Glutamine transport aa in blood:

Glucose/alanine cycle

• Moves C and N between muscle/liver

• Pyruvate + NH$_4^+$ ↔ Alanine; pyruvate for gluconeogenesis

**II. Urea Cycle**

Urea cycle in liver excretes toxic NH$_4^+$ as nontoxic

N enter as NH$_4$, Asp; costs 3 ATP

1. Formation carbamoyl PO$_4$

2. Orn → citrulline (OTC)

3. Cit + Asp → Arg-Suc

4. Arg-Suc → Arg + Fum

5. Arg → Urea + Orn

Fum can do Malate for gluconeogenesis

Fum can do OAA → Asp

**Krebs bi-cycle**

Krebs bi-cycle: TCA cycle and urea cycle

• Interrelationship of TCA and urea cycles

• Urea cycle reactions in cytoplasm except citrulline

• Ornithine is not incorporated in proteins (no codon)
**Regulation of urea cycle:**
- Liver makes urea to prevent NH$_3$ poisoning
- Regulated by substrate availability (NH$_3$)
- High protein diets (or fasting) stimulate synthesis of enz:
  - Convert excess C to glucose, N to urea
- Allosteric regulation:
  - Arg stimulates more ornithine production
  - Arg stimulate formation N-Acetyl glutamate (NAG)
  - NAG stimulates CPSI

**Fig. 38.15**

**Urea cycle in fasting:** liver maintains blood glucose
- Uses muscle protein (aa) for gluconeogenesis
- Nitrogen excretion
- Ala $\rightarrow$ glucose + urea
- 2 Ala $\rightarrow$ 1 glucose, 1 urea

**Disorders of Urea cycle:**
- Dangerous since NH$_3$ toxic
- Need to fix NH$_3$ to Glu or Gln
- See ↑NH$_3$ in blood; also Gln
- Most common is OTC deficiency
- X-linked recessive

**Disorders of urea cycle:**
- Dangerous since NH$_3$ toxic
- Use compounds to absorb N
- Benzoate, phenylbutyrate

**Disorders of Urea cycle**: Fig. 38.18 excretion excess NH$_3$
A. benzoic acid absorbs NH$_3$
B. Phenylbutyrate absorbs NH$_3$
### Key concepts

**Key concepts:**
- Aa catabolism generates urea, nontoxic carrier of N atoms
- Urea synthesis in liver
  - (Ala and Gin carry from tissues)
- Key enzymes are transaminases, glu dehydrogenase (GDH), glutaminase
- Urea cycle has 4 steps: 1 N from NH$_4^+$, 1 from Asp
- Disorders of urea cycle → hyperammonia

### Review question

**Review question:**

2. The nitrogens in urea are derived directly from which of the following compounds?
   - a. Ornithine and carbamoyl phosphate
   - b. Ornithine and aspartate
   - c. Ornithine and glutamate
   - d. Carbamoyl phosphate and aspartate.
   - e. Carbamoyl phosphate and glutamine
   - f. Aspartate and glutamine