Amino Acid Metabolism
Fate of Dietary Protein

Dietary protein

Stomach: HCl, pepsin

Denatured and partially hydrolyzed protein (large polypeptides)

Small intestine: proteases

Amino acids and dipeptides

Intestinal lining: proteases

Amino acids in bloodstream
The catabolism of amino acids takes place in three stages:

1) Removal of the amino group, leaving the carbon skeleton of the amino acid.

2) Breakdown of the carbon skeletons to a glycolytic intermediate, citric acid cycle intermediate, or acetyl-S-CoA.

3) Oxidation of these intermediates to CO$_2$ and H$_2$O with the production of ATP.
Representative Pathways of Amino Acid Catabolism

**Alanine**

\[ \text{alanine} \rightarrow \text{pyruvate} \]

**Aspartate**

\[ \text{aspartate} \rightarrow \text{oxaloacetate} \]

**Glutamine**

\[ \text{glutamine} \rightarrow \text{α-ketoglutarate} \]

**Phenylalanine**

\[ \text{phenylalanine} \rightarrow \text{acetoacetate} + \text{fumarate} \]
pyruvate $\rightarrow$ acetyl-CoA $\leftrightarrow$ acetoacetyl-CoA $\rightarrow$ ketones

gluconeogenesis

citrate

oxaloacetate

malate

fumarate

succinate

succinyl-CoA

aconitate

isocitrate

oxalosuccinate

$\alpha$-ketoglutarate
Glucogenic and Ketogenic Amino Acids

The α-keto acids derived from catabolism of many amino acids are intermediates in glcolysis and the citric acid cycle.

These α-keto acids may, therefore, replenish citric acid cycle intermediates.

Amino acids that can be converted into pyruvate, α-ketoglutarate, succinyl-CoA, fumarate, and oxaloacetate can be converted into glucose by gluconeogenesis and are said to be glucogenic.

The α-keto acids derived from catabolism of some amino acids are broken down to acetyl-CoA or acetoacetyl-CoA and are oxidized by the citric acid cycle or converted to ketone bodies.

These amino acids are said to be ketogenic.
Glucogenic and Ketogenic Amino Acids

- Alanine, glycine, threonine, cysteine, serine
- Leucine, isoleucine, tryptophan, threonine
- Aspartate, asparagine
- Phenylalanine, tyrosine
- Isoleucine, valine, methionine, threonine
- Glutamate, glutamine, arginine, histidine, proline

Reactions:
- Pyruvate → acetyl-CoA
- Oxaloacetate → citrate
- Malate → isocitrate
- Fumarate → oxalosuccinate
- Succinate → α-ketoglutarate
- Succinyl-CoA → citrate
- Aconitate
- Acetyl-CoA → acetoacetyl-CoA → ketones
- Gluconeogenesis: pyruvate → acetyl-CoA → α-ketoglutarate → oxalosuccinate → fumarate → malate → isocitrate → oxaloacetate → pyruvate
The Fate of the **Amino Group** of Amino Acids

Nitrogen is present in the bloodstream in the form of ammonium ions:

\[ \text{NH}_4^+ \]

The normal concentration of \( \text{NH}_4^+ \) ion in the blood is \( 3.0 \times 10^{-5} \) to \( 6.0 \times 10^{-5} \) M.

Above these concentrations (hyperammononemia) coma may result.

The extraction of the amino group from amino acids must be done in such a way so as not to increase blood ammonium ion levels above normal values.
The Fate of the Amino Group of Amino Acids

The pathway for the extraction of amino groups from amino acids consists of three phases:

1) Conversion of amino groups from all amino acids into a single product, glutamate, by transamination.

2) Conversion of glutamate into α-ketoglutarate by oxidative deamination, releasing NH₄⁺.

3) Conversion of NH₄⁺ into urea, which is extracted from the blood by the kidneys and excreted.
\[ \text{\(\alpha\)-amino acid} \rightarrow \text{\(\alpha\)-keto acid} \rightarrow \text{\(\alpha\)-keto glutarate} \rightarrow \text{glutamate} \]

Transamination

Oxidative deamination

\[ \text{NADH, (NADPH), } \text{NH}_4^+ \rightarrow \text{CO}_2 \rightarrow \text{NAD}^+, (\text{NADP})^+, \text{H}_2\text{O} \]

Urea cycle

\[ \text{aspartate} \]

\[ \text{fumarate} \]
α-aminoc acid \rightarrow α-keto acid

α-keto glutarate \rightarrow glutamate

transamination

oxidative deamination

NADH, (NADPH), NH₄⁺

NAD⁺, (NADP)⁺, H₂O

aspartate

fumarate

Urea cycle

Urea

H₂N

NH₂

O

C

CO₂
Transamination
The first step in amino acid metabolism is the removal of the amino group by transamination followed by oxidative deamination.

A transamination reaction can be represented as:

\[
\text{Amino acid}_1 + \alpha\text{-ketoacid}_2 \Leftrightarrow \alpha\text{-ketoacid}_1 + \text{amino acid}_2
\]

Transamination reactions occur in all cells.

The enzymes responsible for transaminations are called transaminases or amino transferases.
Most transaminases are specific for $\alpha$-ketoglutarate but are less specific for the amino acid. This means that the amino groups of almost all amino acids end up on glutamic acid.
One exception to this rule is in skeletal muscle, where transaminases use pyruvate as the amino acceptor, producing alanine as the product.

\[
\text{L-amino acid} + \text{pyruvate} \Leftrightarrow \text{α-ketoacid} + \text{L-alanine}
\]

Another example of a specific transaminase reaction is aspartate transaminase:

\[
\text{L-aspartate} + \text{α-ketoglutarate} \Leftrightarrow \text{oxaloacetate} + \text{L-glutamate}
\]
The presence of alanine transaminase (glutamate:pyruvate transaminase or GPT) and aspartate transaminase (glutamate:oxaloacetate transaminase, or GOT) in the bloodstream, above a certain base level, may indicate liver damage.

Serum GPT and GOT (SGPT and SGOT) tests measure the severity and stage of liver damage.
Vitamin B-6 as a Coenzyme

All transaminases require the coenzyme pyridoxal phosphate (derived from pyridoxine, vitamin B-6):

Vitamin preparations may contain the precursor to pyridoxal phosphate in different forms:
The Transaminase Mechanism

pyridoxal phosphate

pyridoxamine phosphate
Oxidative Deamination

This reaction is reversible and provides a mechanism for

1) generating ammonium ion for excretion as urea

2) generating α-ketoglutarate

3) assimilating ammonium ion for use in other metabolic pathways in the liver and kidneys

Glutamate dehydrogenase

Glutamate

α-Ketoglutarate

NAD+ (NADP+)

H₂O

NADH (NADPH)

NH₄⁺

Does not enter the bloodstream
Amino Group and Ammonia Transport

Amino groups collected in extrahepatic tissues in the form of glutamate must be packaged in a non-toxic form for transport through the blood to the liver.

Glutamate, itself, cannot pass through the cell membranes.

Two different transport forms are used:

1) Production of glutamine in most cell types

2) Production of alanine in muscle cells.
In almost all cell types, glutamine synthetase catalyzes the formation of glutamine from glutamate:

\[
\text{L-glutamate} + \text{NH}_4^+ + \text{ATP} \rightarrow \text{L-glutamine} + \text{H}^+ + \text{ADP} + \text{P}_i
\]

The glutamine, thus formed, is electrically neutral, nontoxic, and can pass through the cell membranes into the blood. The concentration of glutamine in the blood is higher than any other amino acid.
Amino Group and Ammonia Transport

Once in the liver, the reverse reaction takes place and glutamine is deaminated to ammonium ion and glutamate.

\[
\text{L-glutamine} \rightarrow \text{L-glutamate} + \text{NH}_4^+ + \text{H}_2\text{O}
\]

Glutamate can then be oxidatively deaminated to ammonium ion and \(\alpha\)-ketoglutarate.

\[
\text{L-glutamate} + \text{NAD}^+ + \text{H}_2\text{O} \xrightarrow{\text{glutamate dehydrogenase}} \text{\(\alpha\)-ketoglutarate} + \text{NADH} + \text{NH}_4^+
\]
In active muscle cells, large quantities of ammonium ion are produced. After two reactions, alanine is formed. Like glutamine, alanine is electrically neutral:
Like glutamine, alanine is electrically neutral and readily traverses membranes and enters the blood stream.

In the liver, the combination of transamination and oxidative deamination reaction releases ammonium ions.
Glucose/Alanine Cycle

Liver

Glucose

6 ATP

Pyruvate

Urea

4 ATP

Alanine

Muscle tissue

Glucose

2 ATP

Pyruvate

Lactate

α-amino acid

α-keto acid

Alanine

N
Overall Reaction:

$$\text{NH}_4^+ + \text{HCO}_3^- + 3 \text{ATP} + \text{aspartate}(-\text{NH}_3^+)$$

$$\text{urea} + 2 \text{ADP} + \text{AMP} + 4 \text{PO}_4^{3-} + \text{fumarate}$$
Urea Cycle

arginine → ornithine → arginosuccinate → citrulline → urea

arginosuccinate
Urea Cycle

- Ornithine
- Arginine
- Glutamate
- A-ketoglutarate
- Carbamoyl phosphate
- HCO₃⁻
- NH₄⁺
- Pi
- ATP, 2Pᵢ
- ADP, Pᵢ
- Mitochondria
- Cytosol
- Aspartate
- Fumarate
- Argininosuccinate
- Citrulline
- Urea

The Urea Cycle involves the conversion of ammonia (NH₄⁺) into urea (urea), with the help of several enzymes and molecules, including ornithine, arginine, glutamate, a-ketoglutarate, carbamoyl phosphate, and HCO₃⁻. The cycle takes place in both the mitochondria and cytosol of the cell, and it requires the energy of ATP for its operation.
Urea Cycle

\[
\text{arginine} \rightarrow \text{ornithine} \rightarrow \text{citrulline} \rightarrow \text{arginosuccinate} \rightarrow \text{fumarate} \rightarrow \text{urea} \rightarrow \text{carbamoyl phosphate} \rightarrow \text{aspartate}
\]

\[
\text{NH}_4^+ , \text{HCO}_3^- , 2\text{ATP} \rightarrow 2\text{ADP, P}_i
\]
Overall Reaction:

\[ \text{NH}_4^+ + \text{HCO}_3^- + 3 \text{ATP} + \text{aspartate} \]

\[ \text{urea} + 2 \text{ADP} + \text{AMP} + 4 \text{PO}_4^{3-} + \text{fumarate} \]
Dietary protein

Amino acid pool

pyruvate, acetyl-CoA, acetoacetate, TCA cycle intermediates

ATP, via TCA cycle

Fatty acids

Ketones

Glucose

Urea

NH₄⁺

Liver proteins

Plasma proteins

Other nitrogen-containing compounds
Nitrogen excretion products for various organisms

-NH₂ groups

Aquatic invertebrates, bony fishes, crocodiles

Mammals, sharks, some bony fishes, turtles

Birds, insects, reptiles, land gastropods

Scorpions, spiders

Water solubility

Energy needed to produce

NH₃ ammonia

H₂N C=O urea

H₂N C=O uric acid

H₂N N C=O guanine