

## Catabolism of Proteins

Proteins are essential macromolecules found in all living cells. They consist of long chains of amino acids linked by peptide bonds. Protein catabolism is a vital process for microorganisms, enabling them to degrade complex proteins into simpler molecules for energy production, biosynthesis, or metabolic transformation. Microorganisms capable of degrading proteins—known as proteolytic microorganisms—play a key role in putrefaction and nutrient recycling. Their activity is primarily due to the secretion of extracellular proteases. Major groups of proteolytic microorganisms include: Filamentous fungi (molds), Strict aerobic bacteria (e.g., *Pseudomonas*, *Bacillus*), Facultative aerobic–anaerobic bacteria (e.g., *Proteus*, *Serratia*), Strict anaerobic spore-forming bacteria (e.g., *Clostridium*)

### 1. Stages of Protein Catabolism

Protein catabolism occurs in two major steps: **extracellular proteolysis** followed by **intracellular degradation**.

#### 1.1. Extracellular Proteolysis

Many microorganisms secrete **extracellular proteases**, enzymes capable of hydrolyzing peptide bonds and breaking proteins into peptides of smaller size. These enzymes act outside the cell, enabling microorganisms to utilize complex proteins that are too large to be transported across the cell membrane. Examples of microbial protease production:

- *Clostridium histolyticum* produces four distinct types of proteases.
- *Bacillus subtilis* secretes proteases in large quantities (up to 1 g/L), widely used in industrial enzyme production.
- Fungi such as *Penicillium roquefortii* contribute to the specific flavor and texture of cheeses such as Roquefort, thanks to their proteolytic enzymes.

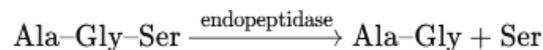
- **Gelatinase** is used in microbiology to differentiate species such as *Bacillus* and various enterobacteria.
- Some proteases cause food spoilage by degrading milk proteins (caseinases), whey proteins, or egg white.

The peptides released during extracellular proteolysis are then transported into the cell via specific membrane proteins called **permeases**.

## 1.2. Intracellular Proteolysis

Once transported inside the cell, peptides undergo further degradation by **intracellular proteases**, known as **peptidases**. These enzymes hydrolyze peptides into free amino acids that can be used for microbial metabolism. Peptidases are classified into two main types:

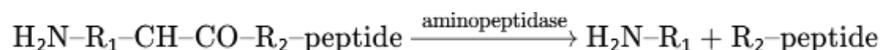
**1. Endopeptidases** : Act within the interior of the peptide chain. Break internal peptide bonds to generate shorter peptides.



**2. Exopeptidases** : Act on the terminal ends of peptides. Two major subtypes exist:

### i- Aminopeptidases

- Attack the N-terminal ( $-\text{NH}_2$ ) end of the peptide.
- Their activity often depends on metal ions (metalloenzymes).



### ii- Carboxypeptidases

- Attack the **C-terminal** ( $-\text{COOH}$ ) end of the peptide.
- Responsible for releasing single amino acids or short peptides.

The combined action of endopeptidases and exopeptidases produces **dipeptides and tripeptides**, which are then completely hydrolyzed into individual amino acids.



These amino acids may then enter various metabolic pathways, including: Protein synthesis, Deamination for energy generation, Conversion into metabolic intermediates (e.g., pyruvate,  $\alpha$ -ketoglutarate, succinyl-CoA)

## 2. Degradation of Amino Acids

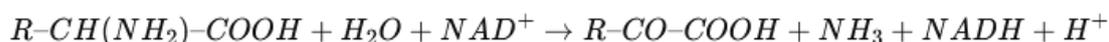
The degradation of amino acids differs from carbohydrate catabolism because it does not directly generate ATP. The energy released from breaking chemical bonds is dissipated mainly as heat. Only the degradation of the resulting carbon skeletons—converted into Krebs cycle intermediates—can yield metabolically useful energy. Amino acid degradation in microorganisms occurs primarily through two pathways: deamination and decarboxylation.

### a) Deamination

Deamination is the removal of the amino group ( $\text{NH}_2$ ) from an amino acid. The reaction releases ammonia ( $\text{NH}_3$ ) and leaves behind a carbon skeleton, generally in the form of an  $\alpha$ -keto acid. Several enzymatic mechanisms exist. Amino acid deaminases remove the amino group, most often producing ammonia. Deamination reactions can be oxidative or non-oxidative.

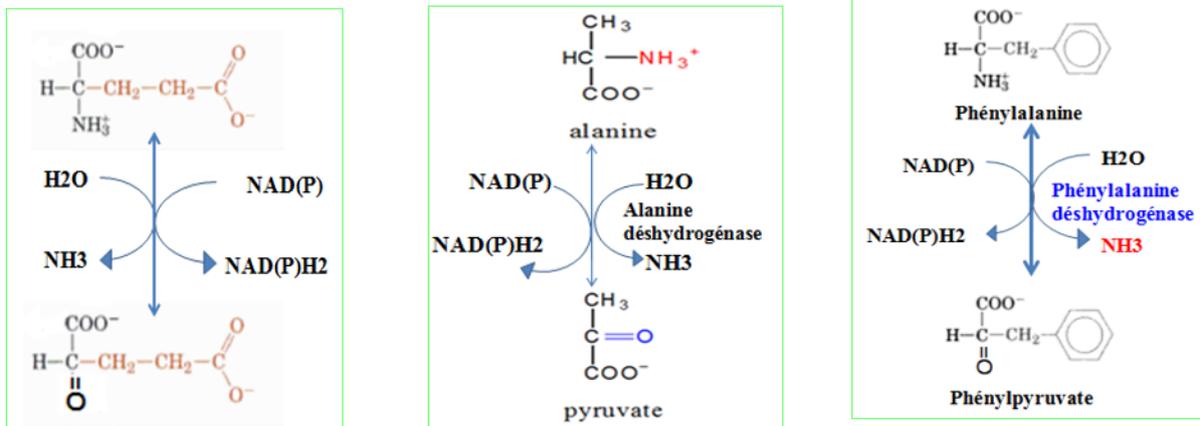
Oxidative deamination is a biochemical reaction in which the amino group of an amino acid is removed and oxidized, producing **ammonia ( $\text{NH}_3$ )** and a corresponding  **$\alpha$ -keto acid**. During this process, electrons are transferred to a coenzyme such as  **$\text{NAD}^+$**  or  **$\text{FMN}$** , which is reduced to  **$\text{NADH}$**  or  **$\text{FMNH}_2$** . This reaction is an important step in amino acid catabolism because the  $\alpha$ -keto acid can enter the **Krebs cycle** to generate energy, while the released ammonia can be excreted or used in nitrogen metabolism.

A general equation for oxidative deamination is:



Examples for specific amino acids include:

- Glutamate + H<sub>2</sub>O + NAD<sup>+</sup> → α-ketoglutarate + NH<sub>3</sub> + NADH + H<sup>+</sup> (catalyzed by glutamate dehydrogenase)
- Alanine + H<sub>2</sub>O + NAD<sup>+</sup> → Pyruvate + NH<sub>3</sub> + NADH + H<sup>+</sup> (catalyzed by alanine dehydrogenase)
- Phenylalanine + H<sub>2</sub>O + NAD<sup>+</sup> → Phenylpyruvate + NH<sub>3</sub> + NADH + H<sup>+</sup> (catalyzed by phenylalanine deaminase)



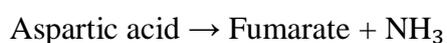
## 2. Non-oxidative deamination

Non-oxidative deamination is a biochemical process in which the amino group of an amino acid is removed without the involvement of an electron-transport cofactor such as NAD<sup>+</sup> or FMN. This type of deamination occurs through three main mechanisms:

### i. Desaturating deamination

In this mechanism, the amino group is removed from the amino acid, producing ammonia (NH<sub>3</sub>) and an unsaturated organic acid. This reaction is catalyzed by specific enzymes such as aspartase in microorganisms like *E. coli* and *Proteus*.

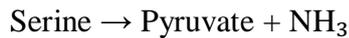
Example:



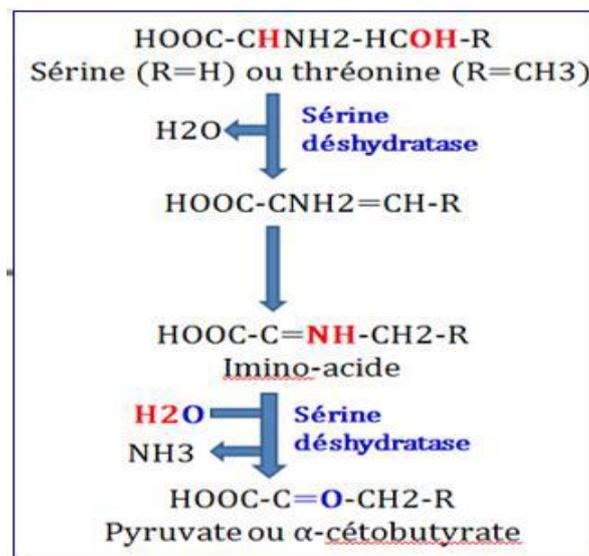
## ii. Deamination by dehydration

This mechanism is specific to hydroxylated amino acids, such as serine and threonine, and occurs exclusively in microorganisms. It involves the removal of the amino group along with a water molecule, producing ammonia ( $\text{NH}_3$ ) and a corresponding  $\alpha$ -keto acid.

Examples:



These non-oxidative deamination reactions allow microorganisms to degrade amino acids under conditions where oxidative deamination is not possible, contributing to nitrogen metabolism and providing carbon skeletons that can enter central metabolic pathways.



## iii. Reductive deamination

Reductive deamination occurs under anaerobic conditions, where the amino acid is reduced rather than oxidized. This reaction produces ammonia ( $\text{NH}_3$ ) and saturated fatty acids. It is a characteristic metabolic pathway of Clostridium species, which use this process to obtain energy and generate reduced carbon compounds in the absence of oxygen.

**Example :**

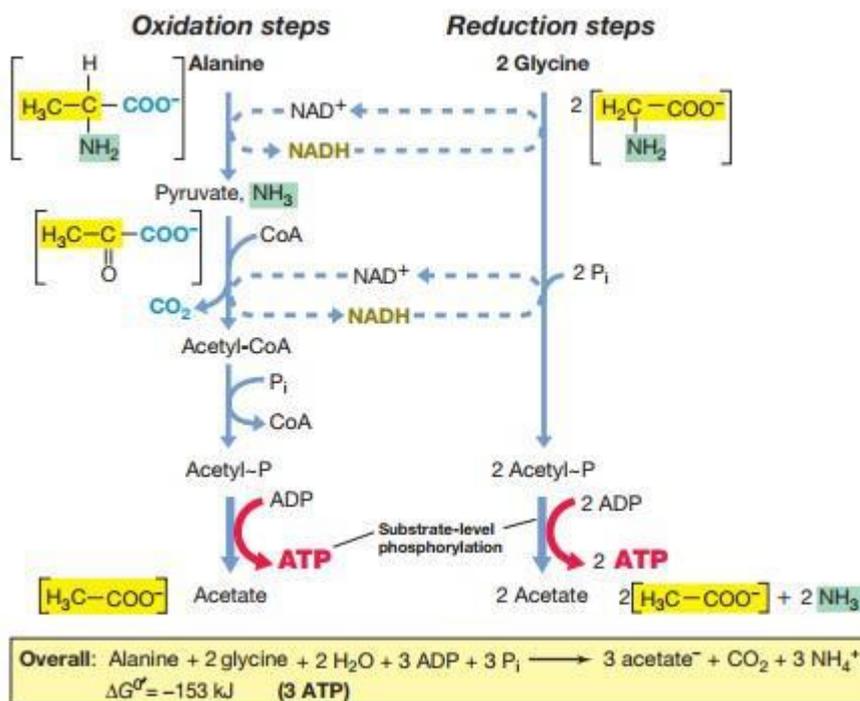


### 3. Coupled deamination (Stickland reaction)

The Stickland reaction is a paired oxidation-reduction (redox) process in which two amino acids are metabolized simultaneously: one amino acid acts as a hydrogen donor, while the other serves as a hydrogen acceptor. This reaction is characteristic of strictly anaerobic, spore-forming bacteria, especially *Clostridium* species, and requires NAD as a coenzyme.

Amino acids that participate in this reaction are degraded in a way that conserves energy for the microorganism. In species that do not perform the Stickland reaction, amino acids are metabolized primarily through transamination pathways, similar to those observed in higher animals, producing  $\alpha$ -keto acids that can enter central metabolic pathways.

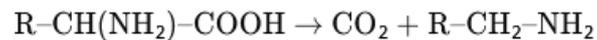
#### General representation:



## b) Decarboxylation

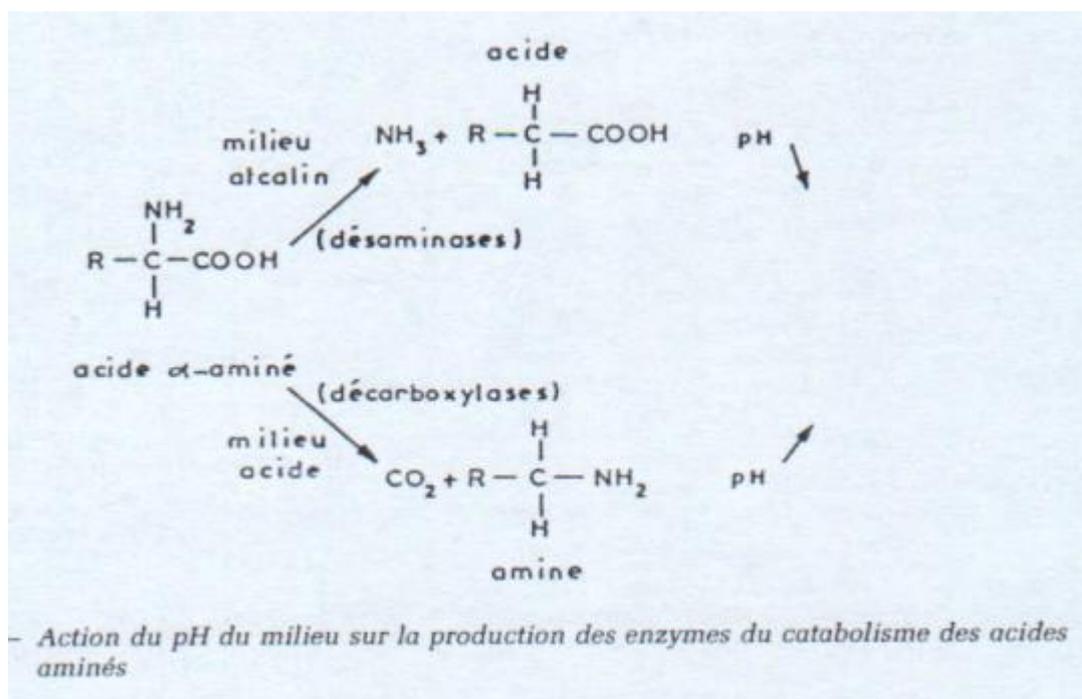
Decarboxylation is another major pathway for amino acid degradation in microorganisms. In this reaction, the carboxyl group ( $-\text{COOH}$ ) is removed from the amino acid, producing carbon dioxide ( $\text{CO}_2$ ) and a corresponding amine.

### General reaction:



This pathway is widely used by both proteolytic and non-proteolytic microorganisms. The amines formed are often strongly odorous and may be toxic; for example, histamine is produced from histidine.

The regulation of amino acid degradation depends strongly on the environmental pH: acidic conditions favor the production of decarboxylases, enhancing decarboxylation, whereas alkaline conditions promote deaminase activity. This allows microorganisms to adjust their metabolism according to environmental conditions.



### **Metabolic pathways of amino acids**

Once released, amino acids can follow different metabolic pathways:

- **Synthesis of new proteins:** Amino acids are incorporated into ribosomes for the synthesis of new proteins, which are essential for cell growth and renewal.
- **Energy production:** Amino acids can be degraded to produce energy. The amino group is removed by deamination, while the carbon skeleton is converted into intermediates of the Krebs cycle, a central metabolic pathway for energy production.
- **Synthesis of other molecules:** Some amino acids can serve as precursors for the synthesis of other molecules, such as nucleotides or coenzymes.

#### **IV.1.3. Importance of protein catabolism**

Protein catabolism is essential for microorganisms for several reasons:

- **Nutrition:** It allows microorganisms to use proteins as a source of nitrogen and carbon, which are essential for their growth.
- **Adaptation to the environment:** Microorganisms can adapt to protein-rich environments by producing specific proteases.
- **Pathogenicity:** Some pathogenic bacteria use protein catabolism to invade host tissues and cause disease.