

Denaturation of Proteins

Revision:

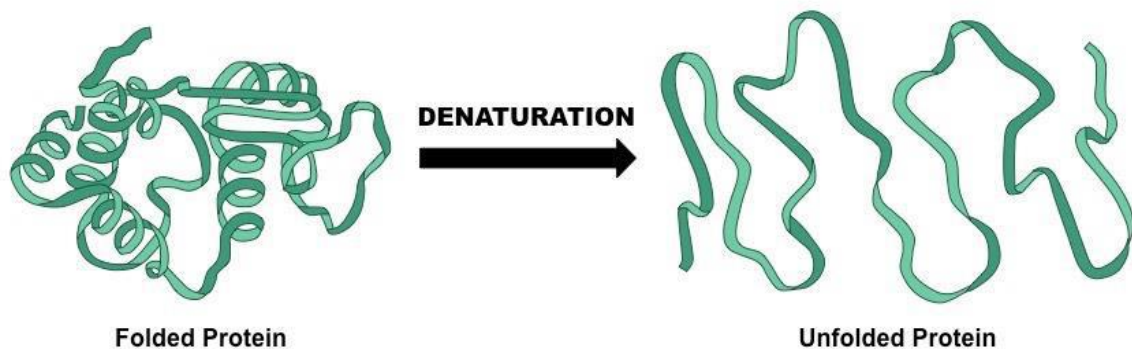
Proteins are polypeptides consist of many amino acids bound together via peptide bonds.

Amino acids interact with each other via different forces to build up the final three dimensional structure of the protein.

These forces are responsible for the stability and rigidity of the protein.

Denaturation of proteins:

Denaturation : is defined as any change that occurs in the structure of a protein, and causes it to lose its biological properties.



The disruption of protein structure is a gradual process.

The 1steady addition of precipitating agent along with 2continuous mixing causes the contents to collide.

When these molecules react with each other, a complex will form, the bonds will be disrupted, and a precipitate may be observed.

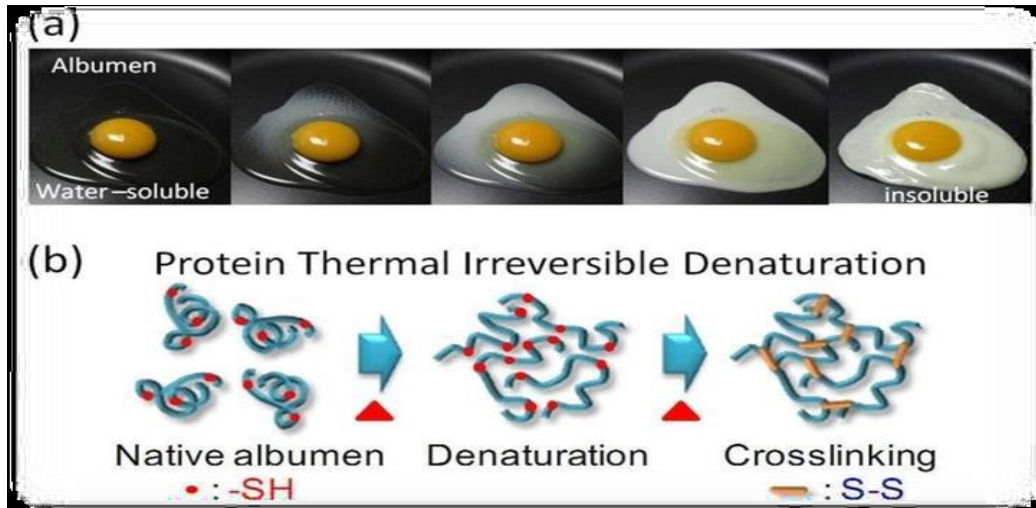
1. Heat:

Temperature and pH changes are usually the main causes of denaturation and precipitation of proteins, because they destroy the weak bonds and affect the tertiary and secondary structures.

However, the primary structure remains unaffected.

Method of denaturation by Heat:

- In a water bath, boil (2 ml) of protein solution (Albumin), for \approx 5 minutes.
- A noticeable change in color and texture is observed.

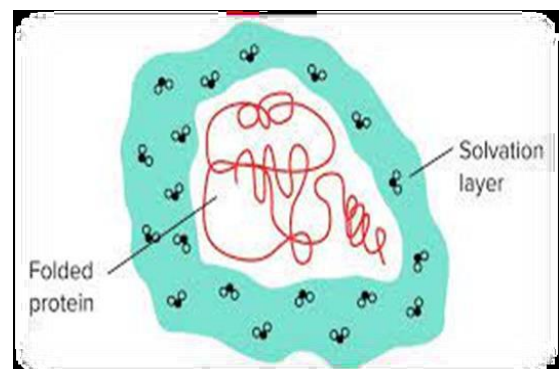
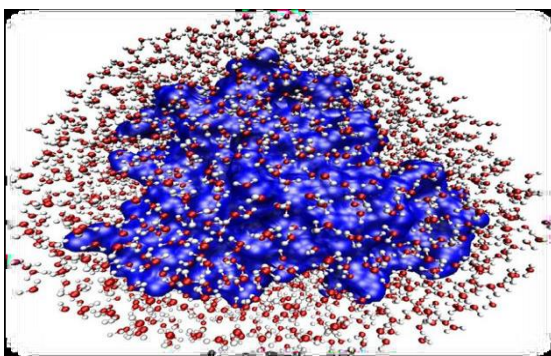


2. Neutral Salts:

Commonly known as **salting-out method**.

Addition of neutral salt (Ammonium sulfate) causes water ions to associate with the added ions; hence, less water will be available for the solvation layer around proteins.

This exposes the hydrophobic parts of proteins to the surface, causing them to precipitate away from water.



Method of denaturation by neutral salts:

- Take (1 ml) of protein solution (Albumin).
- Add (1 ml) of saturated ammonium sulfate solution.
- Mix well.
- Observe the formation of a precipitate in the sample.

3. Heavy Metal Salts:

Metal salts usually contain ions with high atomic weight (Hg^{2+} , Pb^{2+} , Ag^{1+}). Since these ions are positively charged, they react with the negatively charged carboxyl group, forming an insoluble metal protein salt, and disrupt the structure of proteins.

Method of denaturation by heavy metal salts:

- Take (1 ml) of protein solution (Albumin).
- Add (0.5 ml) of mercuric nitrate solution.
- Mix well.
- Observe the formation of a precipitate in the sample.

4. Change in pH (Acidic, Alkaloid):

Changing the pH of the environment of a protein results in a change in the charge of its amino acids.

Increasing the acidity or alkalinity of a protein solution will cause the appearance of a precipitate.

a. Method of denaturation by Acidic solution:

- Take (1 ml) of protein solution (Albumin).
- Add (1 ml) of HCL solution.
- Mix well, add more drops of acid if needed.

- Observe the formation of a precipitate in the sample.

b. Method of denaturation by Alkaloid solution:

- Take (1 ml) of protein solution (Albumin).
- Add (1 ml) of conc. NaOH solution.
- Mix well.
- Observe the formation of a precipitate in the sample.

5. Alcohol:

Ethanol disrupts the hydrogen bonding within the protein structure by forming intra hydrogen-bonds.

Hence, 70% alcohol is used in sterilization, because it effectively penetrates the bacterial cell wall. Whereas, 95% alcohol forms a crust, preventing the penetration of alcohol into the cell wall.

Method of denaturation by alcohol:

- Take (1 ml) of protein solution (Albumin).
- Add (1 ml) of 70% alcohol solution.
- Mix well, add more drops of alcohol if needed.
- Observe the formation of a precipitate in the sample.