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Polypeptide & protein Structure - 2

Fibrous Proteins

α-keratins, bundles of α-helices

- Contain polypeptide chains organized approximately parallel along a single axis:
  - Consist of long fibers or large sheets
  - Mechanically strong
  - Insoluble
  - play an important structural role

- Examples are
  - Keratin
  - Collagen
 - fibroin

Fibroin, β-sheets,

Globular Proteins

- Folded to, a more or less, spherical shape
  - Soluble
  - Polar vs. non-polar, exterior vs. interior
  - Most of them have substantial sections of α-helix and β-sheet

Filament
(four right-hand
twisted protofilaments)
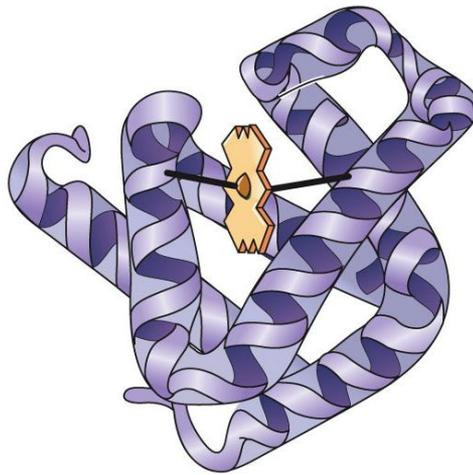
Myoglobin, a globular protein
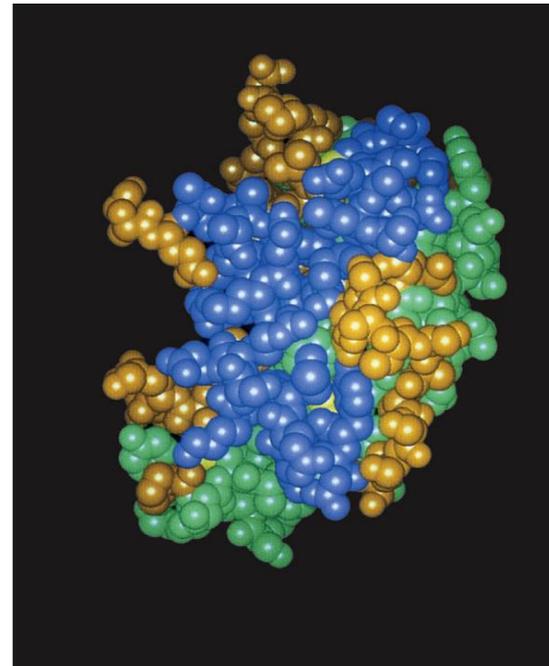
3° Structure

- The 3-dimensional arrangement of all amino acids in a protein

    - The overall conformation of a polypeptide chain

    - The spatial arrangement of amino acid residues that are far apart in the sequence

- Simple vs. conjugated

How to look at proteins…

Protein surface map
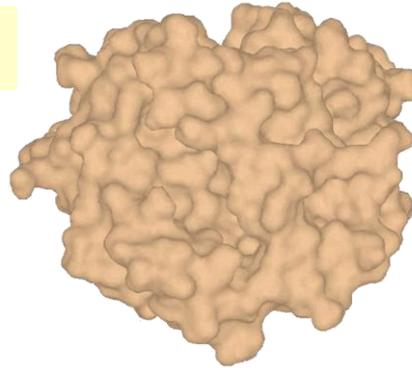
**Space filling structure**
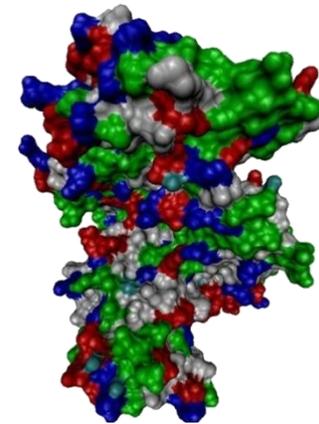
Trace structure

Ribbon structure

MLV
HIV
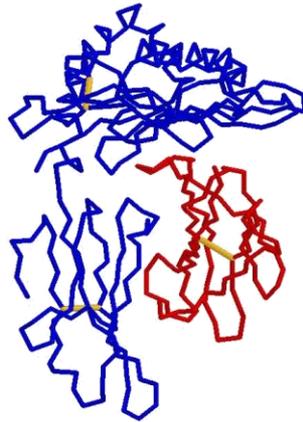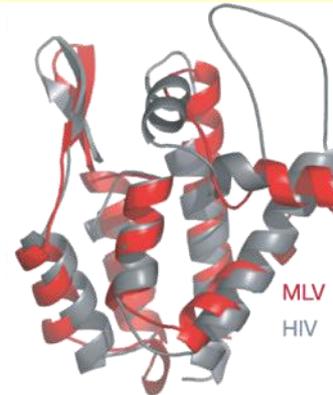
**Cylinder structure**

β2   β1
α6
α5
α2
α3
α4
α1
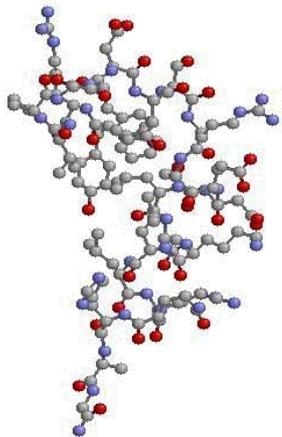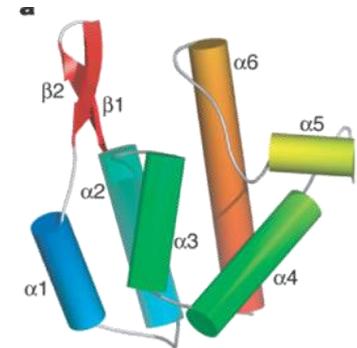
Ball & stick structure

Interactions in Protein Structure

- Backbone H-bonding
- Side chain H-bonding
- Hydrophobic interactions
- Electrostatic attraction
- Electrostatic repulsion
- Metal coordination
- Covalent bonding

Metal ion coordination

Hydrophobic interactions

Electrostatic attraction

M^{2+}

NH_3^+

COO^-

Leu
Val
Ile

CH_2
S—S
CH_2

Side chain hydrogen bonding

Disulfide bond

Helical structure **Sheet structure**

N   C
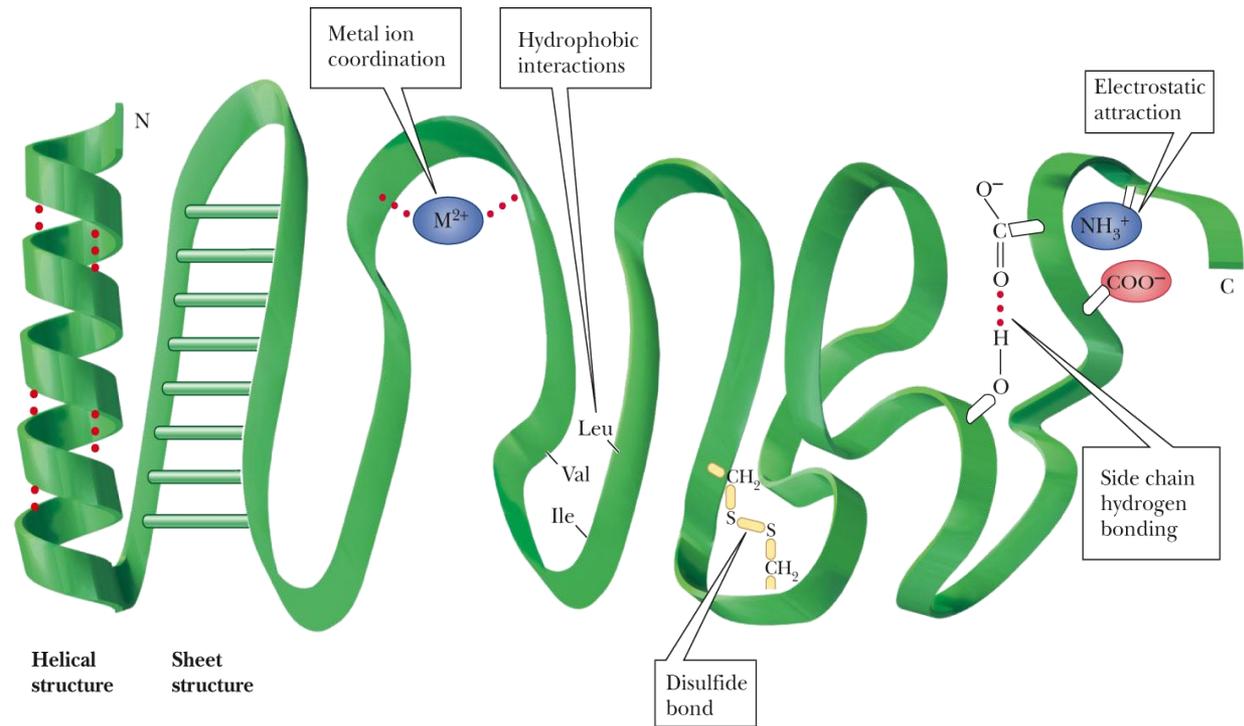
- **Not every protein have all kinds of interactions** (myoglobin & hemoglobin; no S-S) (trypsin & chymotrypsin, no metal complexes)

- Interactions between side chains also play a role

# Shape determining forces

- Non-covalent interactions
  - Hydrophobic interactions
  - Hydrogen bonds: amino acids, aqueous medium
  - Charge-charge interactions (salt bridges)
  - Charge-dipole interactions: charged R groups with partial charges of water
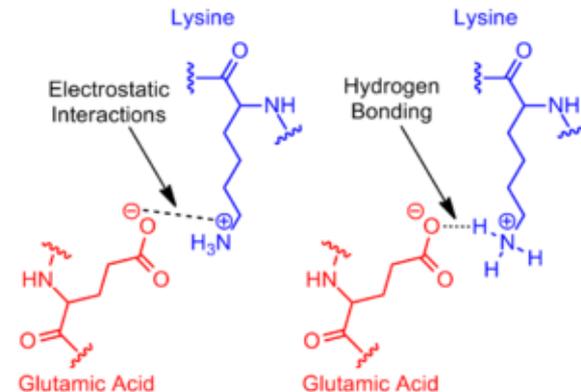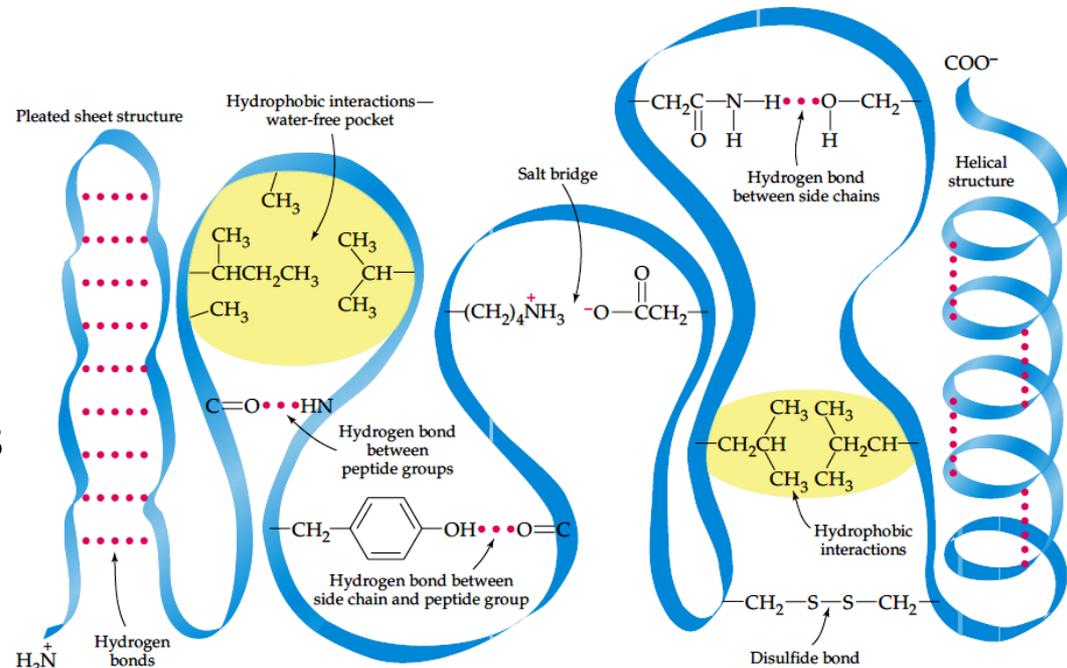
Hydrophobic interactions

- A system is more thermodynamically (energetically) stable when hydrophobic groups are clustered together rather than extended into the aqueous surroundings
- Can polar amino acids be found in the interior?
  - H-bonds to other amino acids (side chain or backbone)
  - Play important roles in the function of proteins

polar side chains   nonpolar side chains

unfolded polypeptide

hydrophobic core region contains nonpolar side chains

polar side chains on the outside of the molecule can form hydrogen bonds to water

folded conformation in aqueous environment
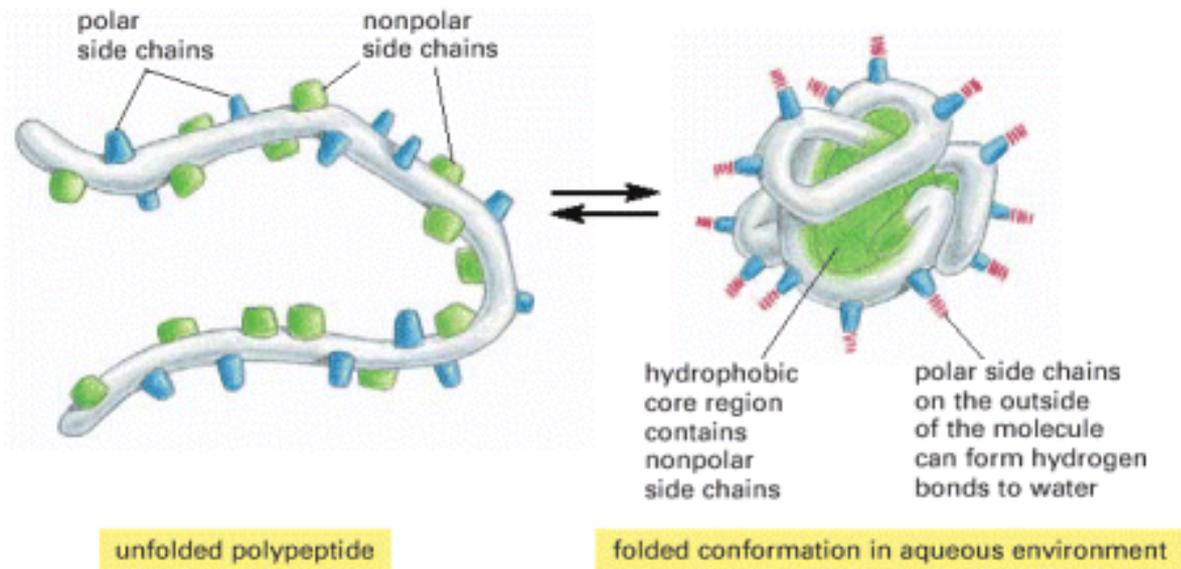
Factors that stabilize protein structures
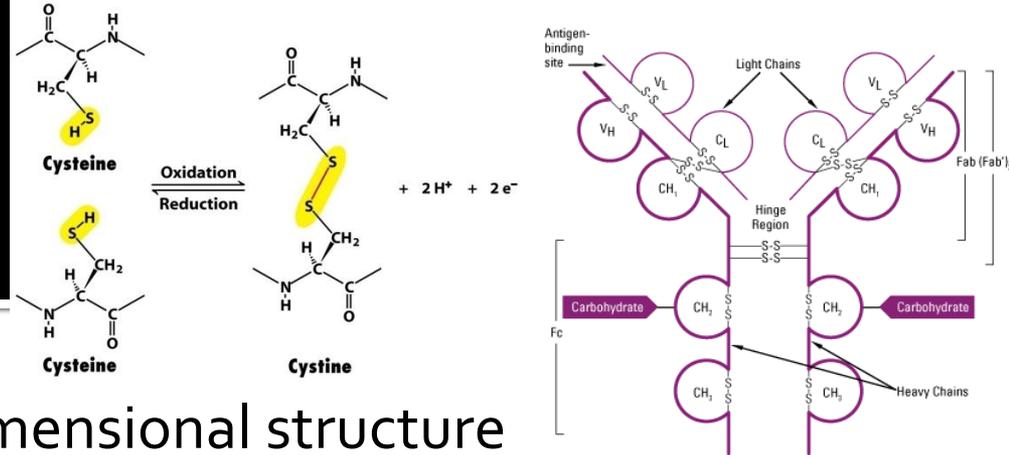
- Do not determine the 3-dimensional structure but stabilizes it:

  - Disulfide bonds (redox)

  - Metal ions

    - Covalent interaction (myoglobin)

    - Salt bridges (carbonic anhydrase)
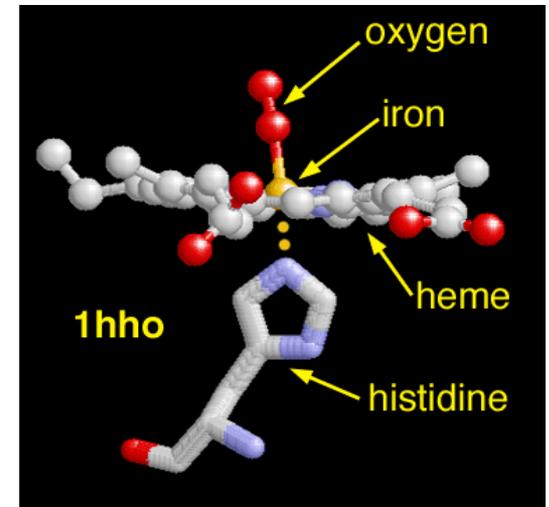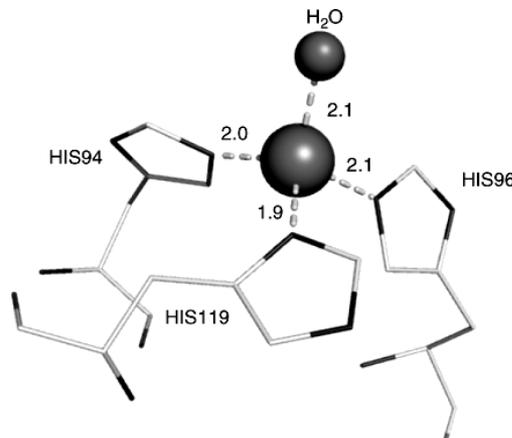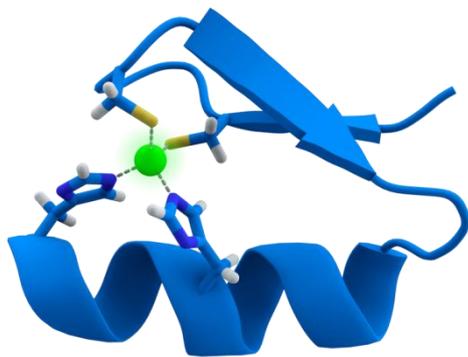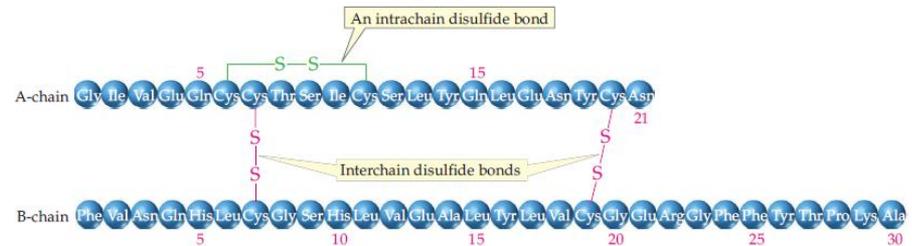
3°& 4°Structure
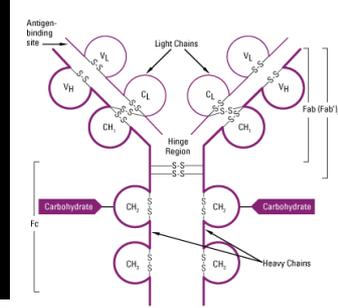
- **Tertiary (3°) structure**: the arrangement in space of all atoms in a polypeptide chain
  - **It is not always possible to draw a clear distinction between 2°and 3°structure**
- **Quaternary (4°) structure: the association of polypeptide chains into aggregations called "subunits" (dimers, trimers, tetramers,…etc).**
  - Simple or conjugated (holo vs. apo)
  - Homo vs. hetero
  - Interactions:
    - Mainly: Non-covalent
    - Sometimes: covalent (S-S)
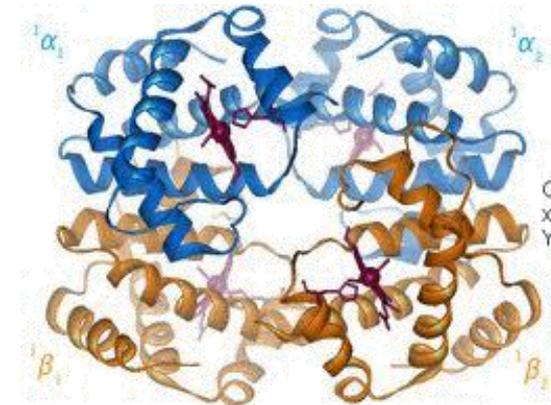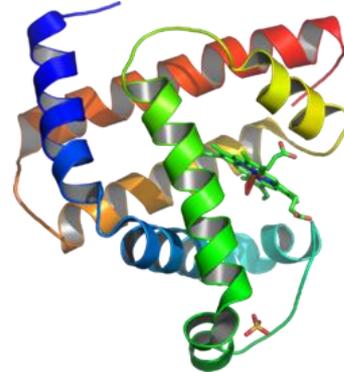
Complex Protein Structures

- Carbohydrates (glycoproteins):
  - Covalent conjugation
    - *N*-linked (-N of Asn)
    - *O*-linked (-OH of Ser or Thr) & occasionally to –OH of hydroxy-lysine
- Lipids (lipoproteins):
  - Non-covalent
  - Store & transport lipids & cholesterol
- Phosphates (Phosphoproteins):
  - Covalent
  - Esterified to Ser, Thr, or Tyr
 - Usually regulates protein function
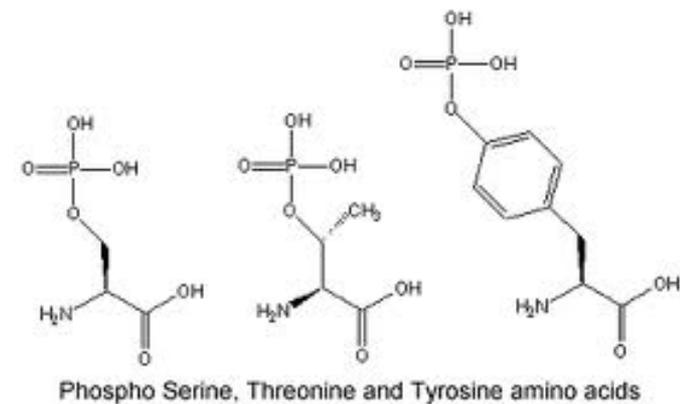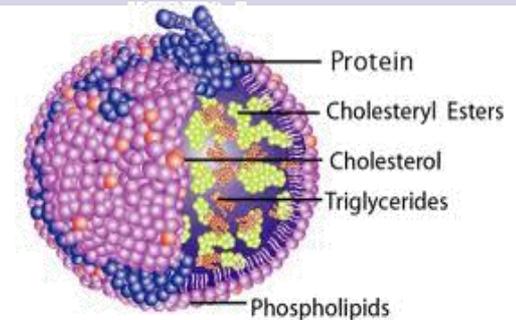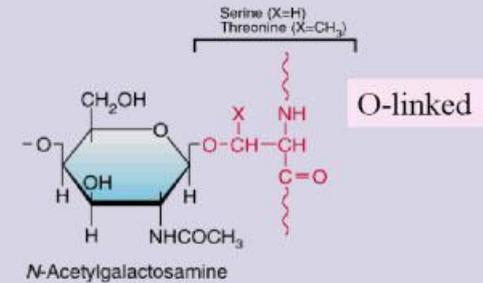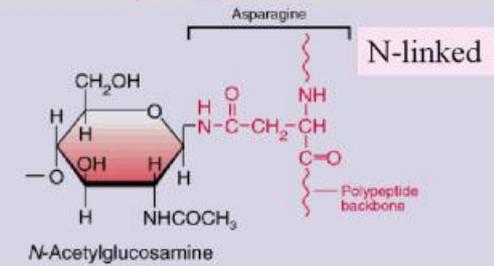
Chemical Properties of Proteins

1. Protein Hydrolysis

- The reverse of protein synthesis
- Digestion of proteins is hydrolyzing peptide bonds
- Takes place in the stomach and small intestine
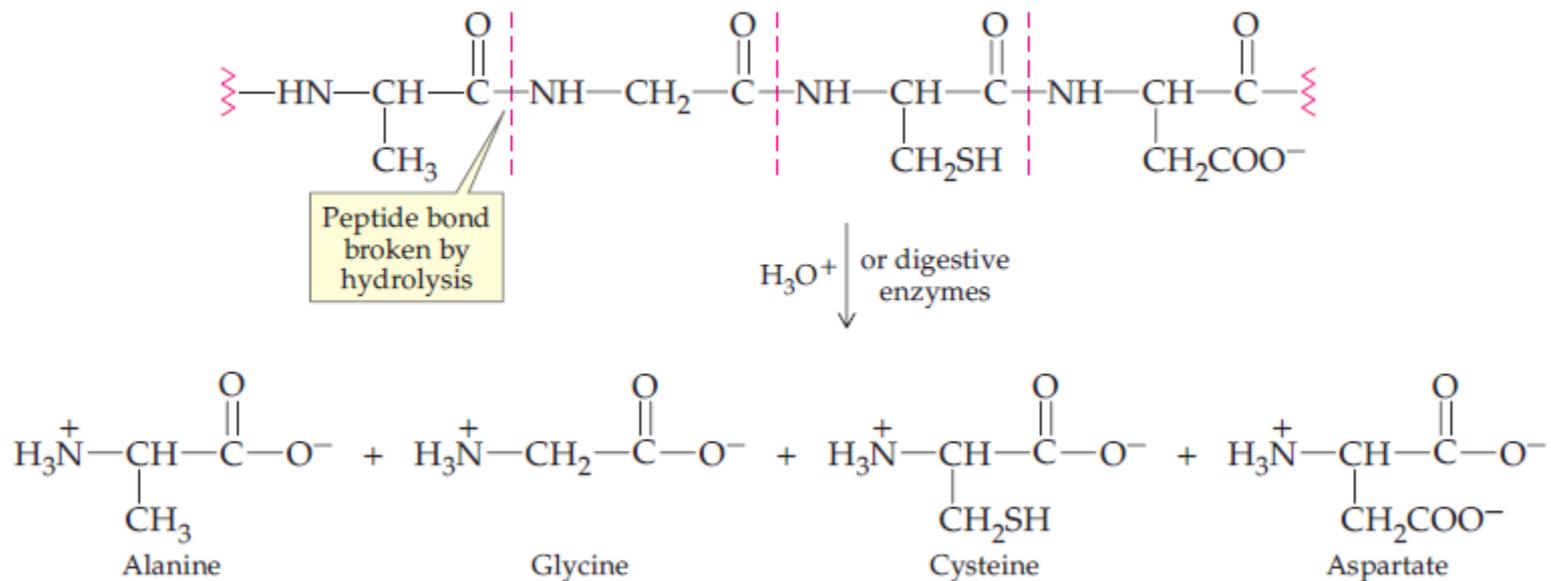
2. Protein Denaturation

- How the protein preserve its shape?
- What is denaturation? It affects physical, chemical, and biological properties, such as enzymes
- Solubility decreased
- Causes:
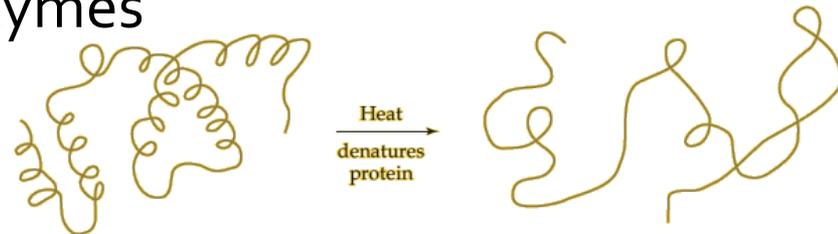
1) Heat (≈≥50 °C): low-energy van der Waals forces & H-bonding

2) Mechanical agitation

3) Detergents: hydrophobic forces
    - Triton X-100 (nonionic, uncharged)
    - Sodium dodecyl sulfate (SDS, anionic, charged) - also electrostatic interactions

2. Protein Denaturation

- Causes:

  4) Organic compounds: acetone, ethanol, bacterial proteins

  5) pH change: disrupt salt bridges & H-bonding
     - Urea and guanidine hydrochloride

  6) Reducing agents: disulfide bonds
     - β-mercaptoethanol (βME) and dithiothreitol (DTT)

72
58 65
110   84 26
95
40
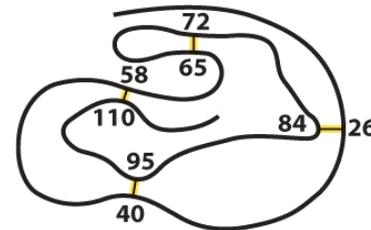
Native state; catalytically active.

addition of urea and mercapto-ethanol

SH
65
HS
72
HS 40 58
SH   SH
HS 26   84
95
HS
HS 110
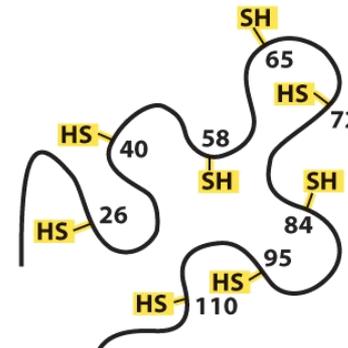
Unfolded state; inactive. Disulfide cross-links reduced to yield Cys residues.

Heat denatures protein
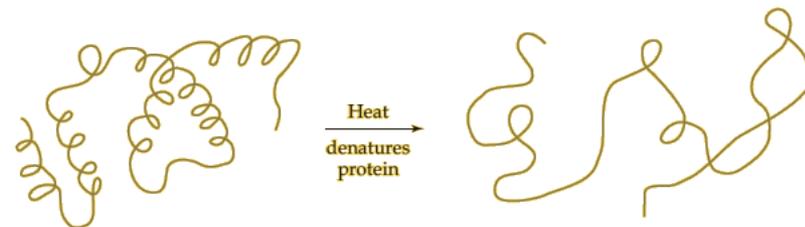
Factors that determine the 3D-structure of a protein

– The least amount of energy needed to stabilize the protein. This is determined by:

  – The amino acid sequence (the primary structure), mainly the internal residues - hydrophobic

 – Non-protein molecules

  – Chaperones

The problem of misfolding

- Hydrophobic interactions are spontaneous
- When proteins do not fold correctly, their internal hydrophobic regions become exposed and interact with other hydrophobic regions on other molecules, and form aggregates
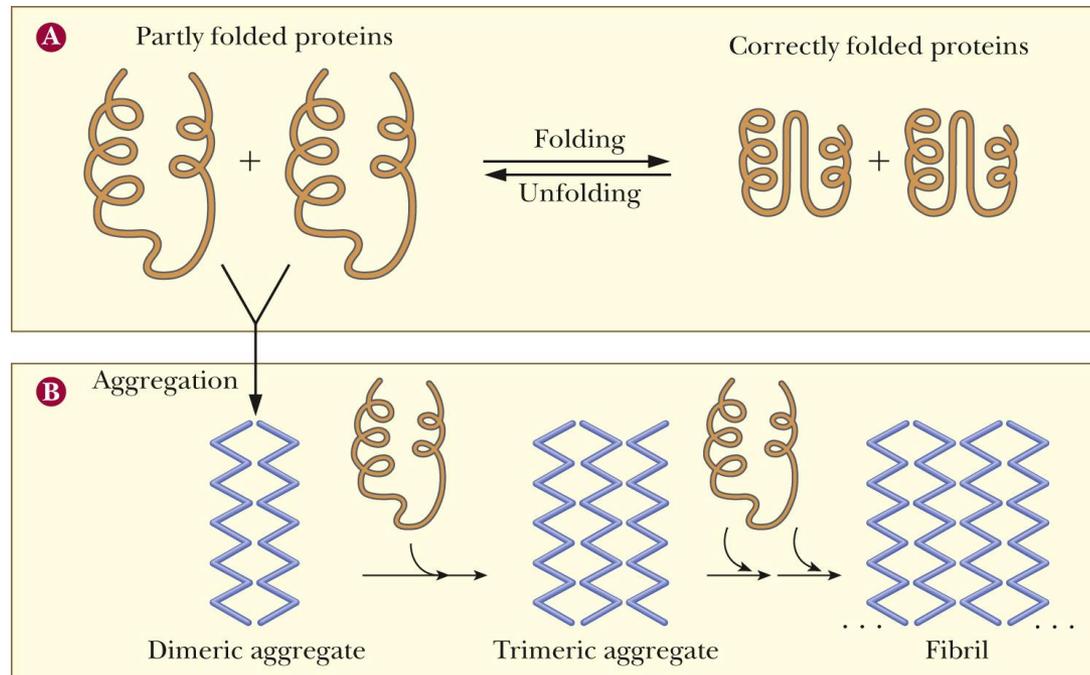
Problem solvers: chaperones

- Aid in correct & timely folding of many proteins
- Exist in organisms from prokaryotes to humans
- hsp70 were the first chaperone proteins discovered
- Function:
  - Help them fold with the most energetically favorable folding pathway
  - Prevent the hydrophobic regions in newly synthesized protein chains from associating with each other to form protein aggregates

Unfolded polypeptide   +   Chaperone n ATP n ADP + n P$_i$ +   Folded polypeptide
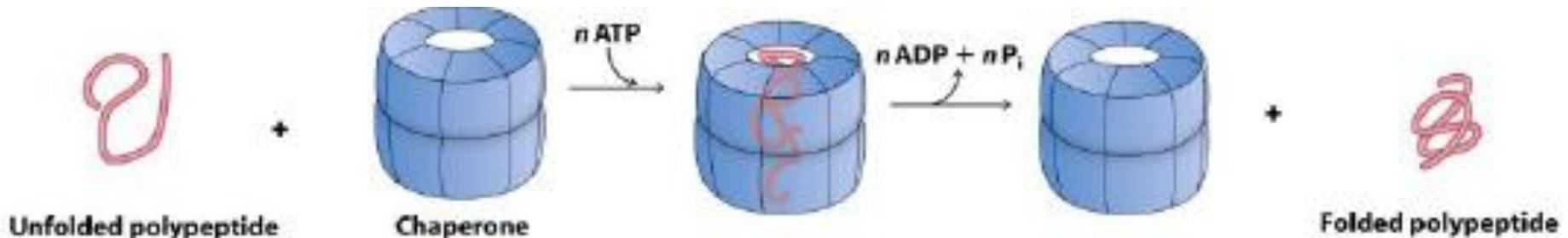
Folding & Diseases

Outcome of protein misfolding

- Partly folded or misfolded polypeptides or fragments may associate with similar chains to **form aggregates**

- Aggregates vary in size from soluble dimers and trimers up to insoluble fibrillar structures (amyloid)

- Both soluble and insoluble aggregates **can be toxic to cells**

Prion disease

- Prion diseases:
  - Creutzfeldt-Jacob disease (in humans)
  - Mad cow disease (in cows)
  - Scrapie (in sheep) - قعاص الغنم أو الراعوش
- Prion protein (PrP, 28 kDa) is misfolded into an incorrect form called PrP^{sc} - (Met$_{129}$)
- PrP has a lot of α-helical conformation, but PrP^{sc} has more β strands forming aggregates
- Abnormal protein can be acquired by:
  - Infection
  - Inheritance
  - Spontaneously
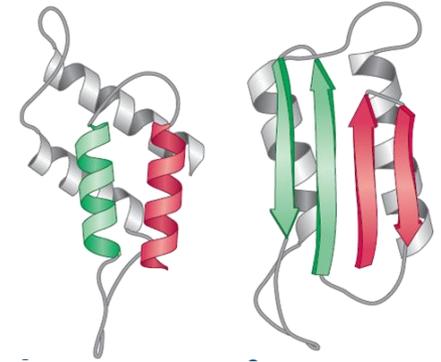
(A) very rare conformational change

(B) infectious seeding of amyloid fiber formation

heterodimer   homodimer   amyloid
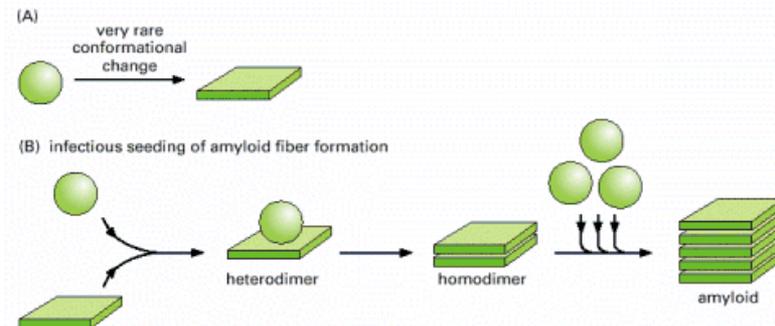
Alzheimer's Disease

- Not transmissible between individuals

- **Aβ** (≈40 a.a) is a short peptide derived from a larger protein (amyloid precursor protein, **APP**)
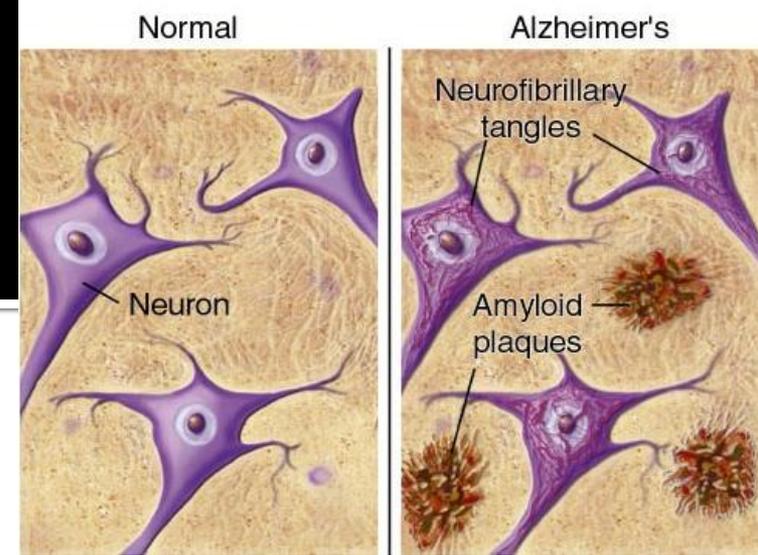
- Extracellular plaques: amyloid (**Aβ**)
- Intracellular tangles: *tau*