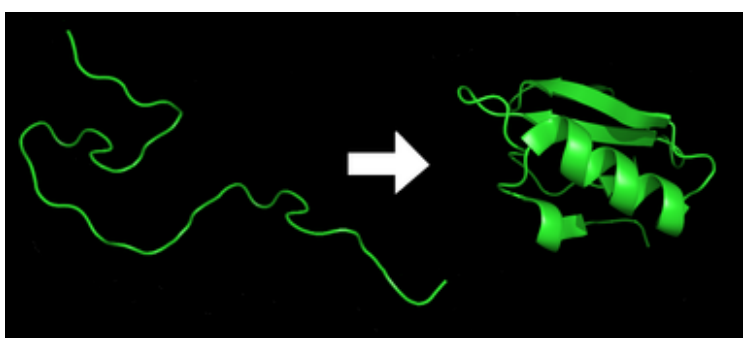
Lecture 24-25

Protein Folding

Protein Folding

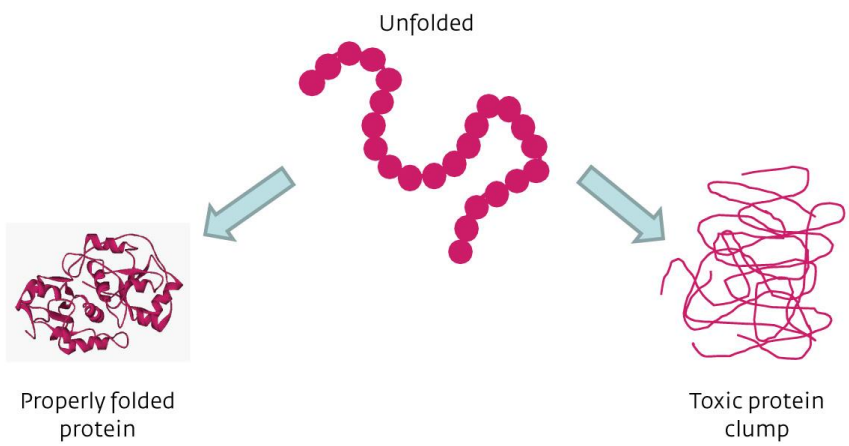
Protein folding is the process by which a protein structure assumes its functional shape or conformation. It is the physical process by which a polypeptide folds into its characteristic and functional three-dimensional structure from random coil.



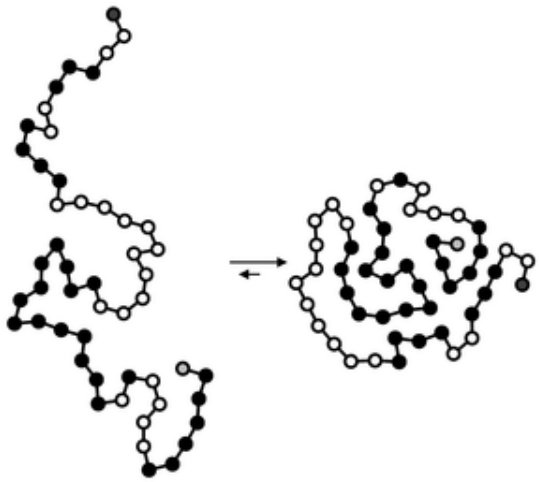
Folding, unfolding, misfolding

- The correct three-dimensional structure is essential to function, although some parts of functional proteins may remain unfolded.
- Failure to fold into native structure generally produces inactive proteins, but in some instances misfolded proteins have modified or toxic functionality.
- Several neurodegenerative and other diseases are believed to result from the accumulation of amyloid fibrils formed by misfolded proteins.
- Many allergies are caused by incorrect folding of some proteins, for the immune system does not produce antibodies for certain protein structures

Folding, unfolding, misfolding



Folding driving forces

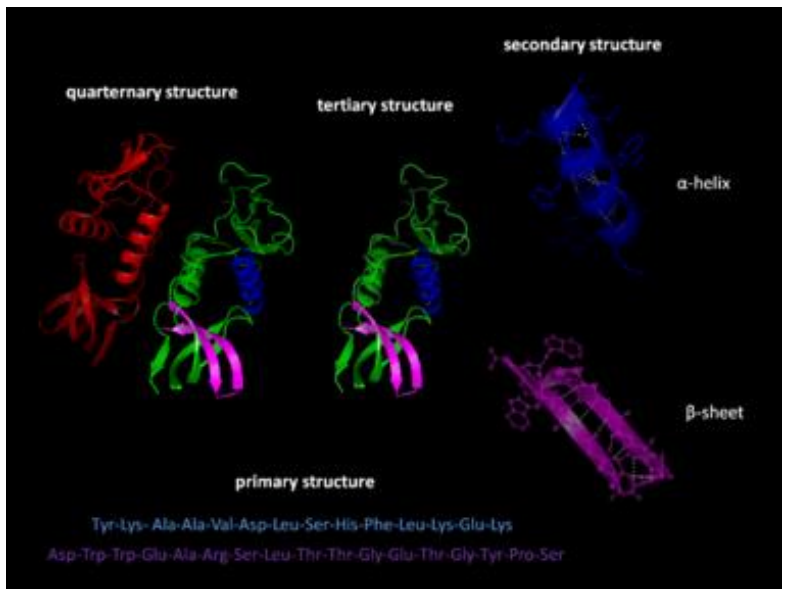


- Minimizing hydrophobic side-chains exposed to water,
- solvent (water or lipid bilayer),
- concentration of salts,
- pH,
- temperature,
- possible presence of cofactors, molecular chaperones.
- intramolecular hydrogen bonds
- van der Waals interaction
- Electrostatic interaction
- And many more …

Folding Simulation

- Video

Protein Structure



Protein Folding Models

Folding often begins co-translationally, so that the N-terminus of the protein begins to fold while the C-terminal portion of the protein is still being synthesized by the ribosome.

- The diffusion collision model, in which a nucleus is formed, then the secondary structure is formed, and finally these secondary structures are collided together and pack tightly together.
- The nucleation-condensation model, in which the secondary and tertiary structures of the protein are made at the same time.

Relationship between folding and amino acid sequence

Anfinsen's dogma

The native structure is determined only by the protein's amino acid sequence.

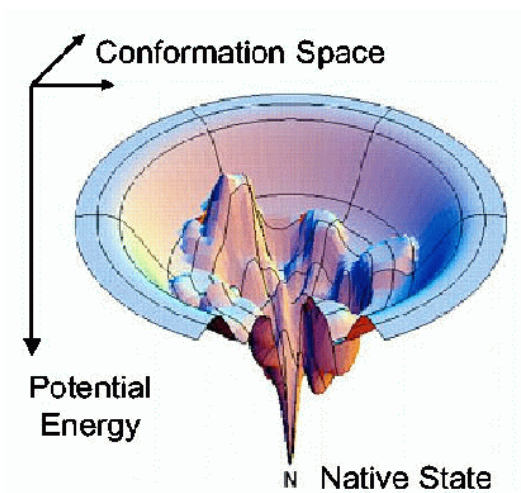
Tested the folding on ribonuclease A.

Limitations:

- Uniqueness
- Stability
- Kinetical accessibility

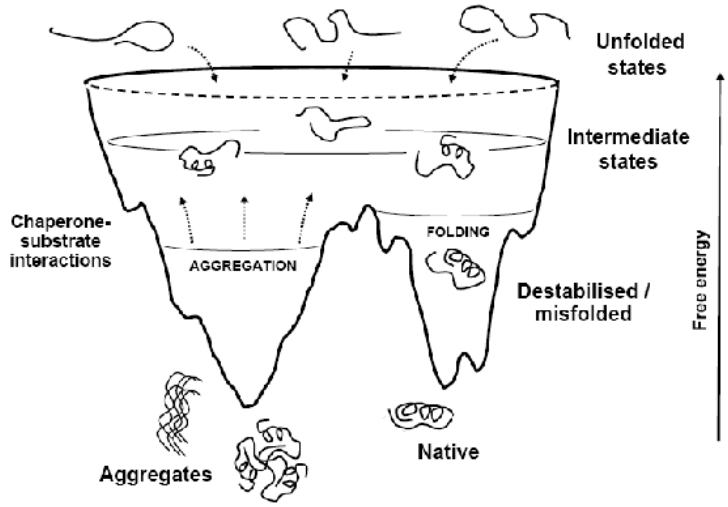
Energy landscape

In physics and biochemistry, an ***energy landscape*** is a mapping of all possible conformations of a molecular entity, or the spatial positions of interacting molecules in a system, and their corresponding energy levels, typically Gibbs free energy, on a two- or three-dimensional Cartesian coordinate system.



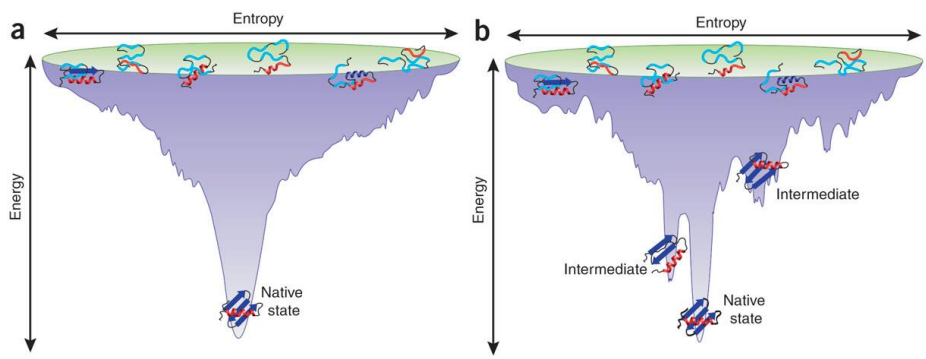
Dill, K.A. and Chan, S. (1997) *Nature Structural. Biology*, 4:10–19

Energy landscape



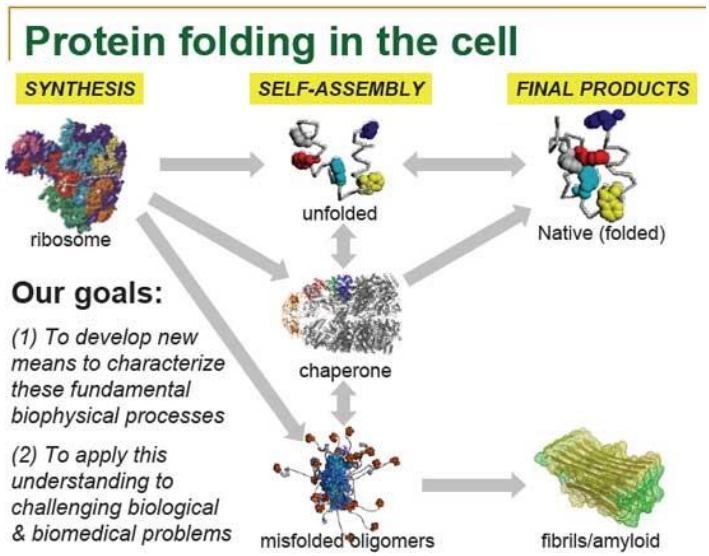
Leandro, Paula & Gomes, Cláudio. (2008). Mini reviews in medicinal chemistry. 8. 901-11. 10.2174/138955708785132783.

Folding funnel

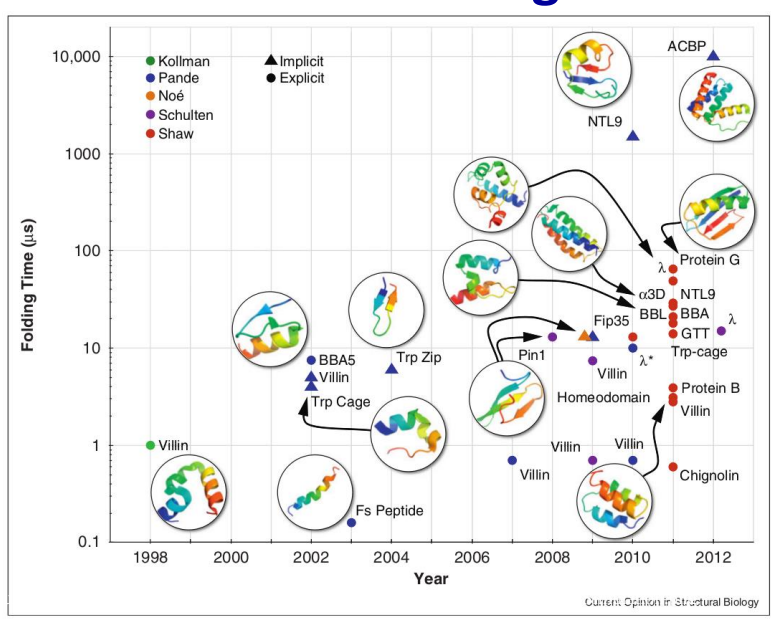


Alice I Bartlett & Sheena E Radford (2009). *Nature Structural & Molecular Biology* 16:582-588

In vivo folding



In-silico Folding



Post Translation Modifications

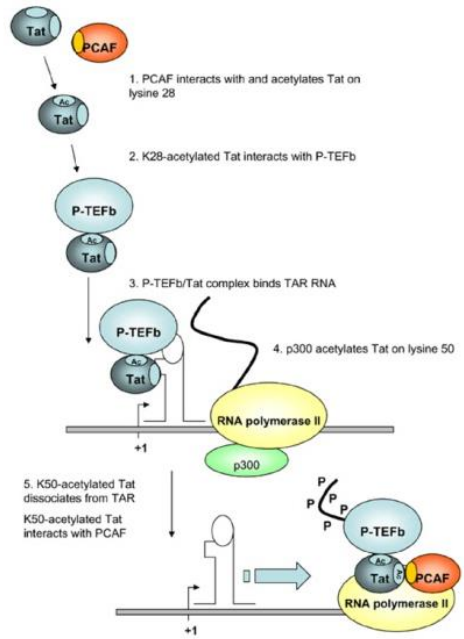
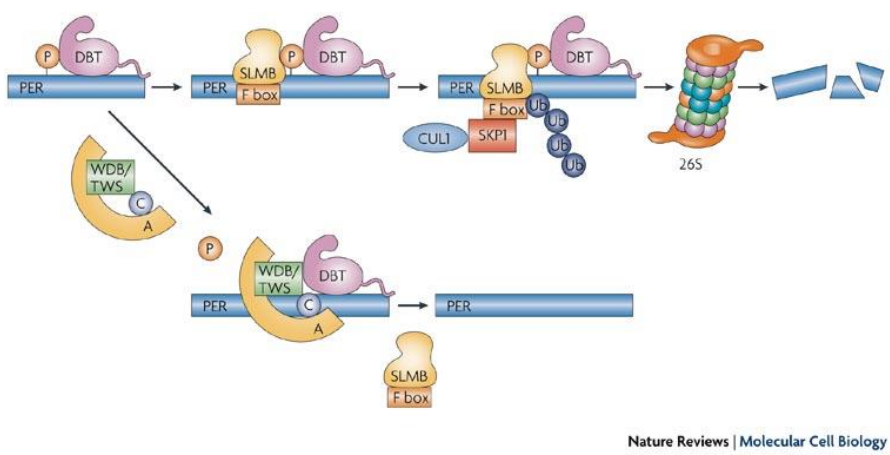


Fig.2: Regulation of the viral transactivator Tat transcriptional activity by post translational modifications

Post Translation Modifications



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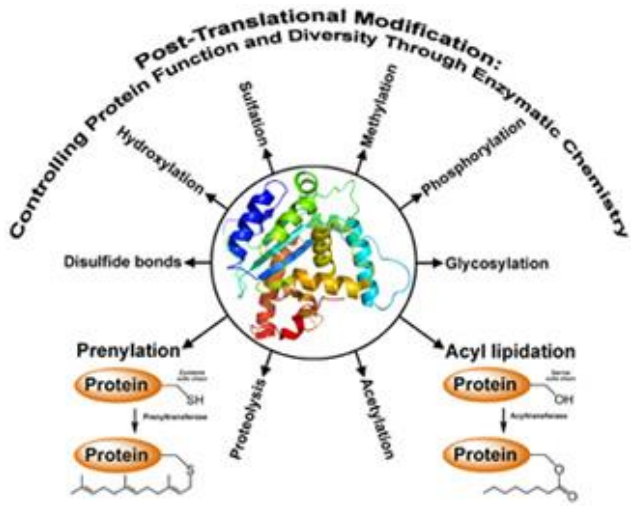
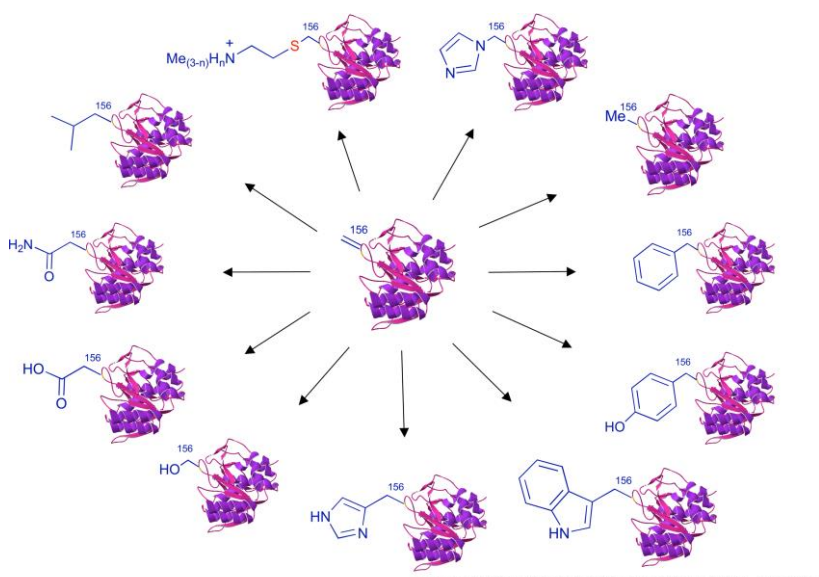
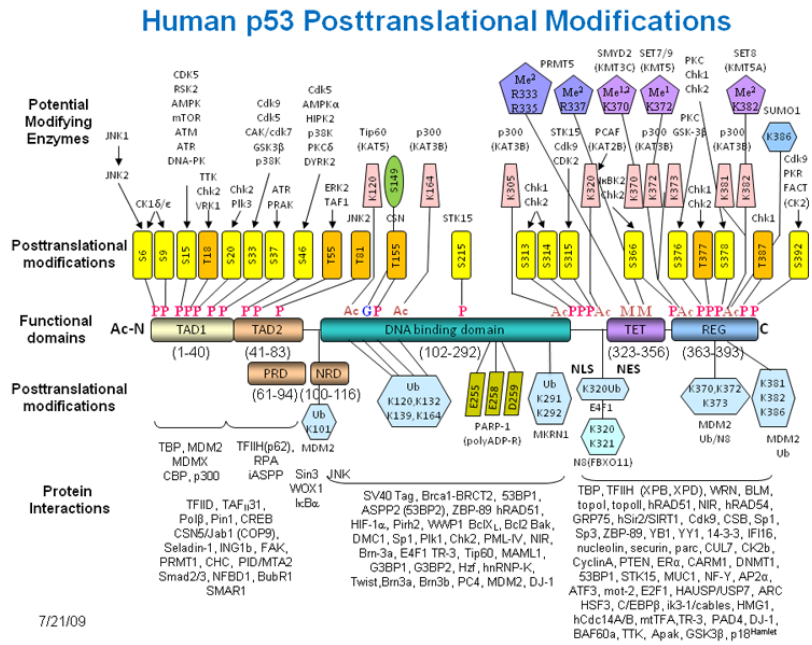
Post Translation Modifications



Post Translation Modifications



Koshland *et al*, *Proc Natl Acad Sci USA* **1966**, *56*, 1606

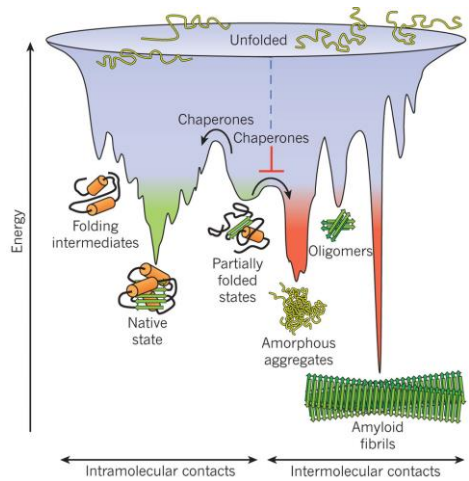


External factors on protein trajectories

Modification of the local minima by external factors can also induce modifications of the folding trajectory.

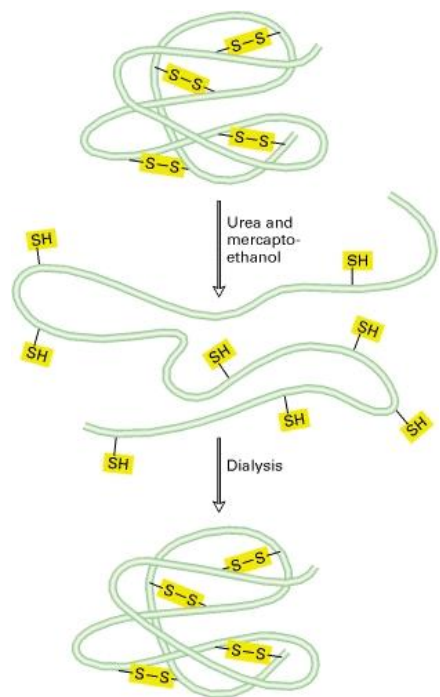
- Temperature,
- Electric, and/or magnetic fields,
- Molecular crowding
- Space constraints.

Folding in funnel



In vitro denaturation and renaturation of proteins

Treatment with an 8 M urea solution containing mercaptoethanol ($HSCH_2CH_2OH$) completely denatures most proteins.



Disruption of the native state

Native state or biochemically functional forms may be disrupted for

- Thermal instability: Temperatures above or below the admissible range
- High concentrations of solutes
- Inadmissible pH
- Presence of chemical denaturants can do the same.

Denature, Refolding, Aggregates

- A fully denatured protein lacks both tertiary and secondary structure, and exists as a so-called random coil.
- Mostly denaturation is irreversible.
- Chaperones or heat shock proteins protect against the denaturing.
- In some situations some misfolded proteins are unfold, for a second chance to refold properly. This function is crucial to prevent the risk of precipitation into insoluble amorphous aggregates.

Incorrect fold and neurodegenerative disease

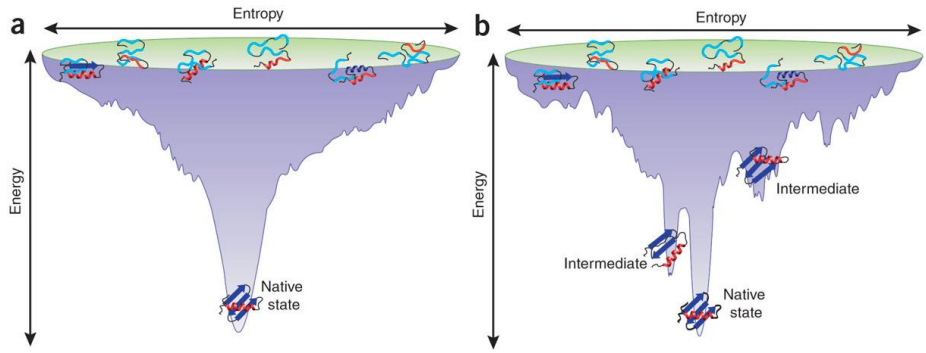
Aggregated/Misfolded proteins accompany illnesses:

- Creutzfeldt-Jakob disease,
- Bovine spongiform encephalopathy (mad cow disease),
- Amyloid-related illnesses such as Alzheimer's disease
- Familial amyloid cardiomyopathy or polyneuropathy,
- Intracytoplasmic aggregation diseases such as Huntington's and Parkinson's disease.
- Antitrypsin-associated emphysema,
- Cystic fibrosis
- Lysosomal storage diseases,

Experimental techniques

- Protein nuclear magnetic resonance spectroscopy
- Circular dichroism
- Dual polarisation interferometry
- Vibrational circular dichroism of proteins
- Studies of folding with high time resolution
- Proteolysis
- Optical tweezers

Folding funnel



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Levinthal Paradox

Levinthal proposed that a random conformational search does not occur, and the protein must, therefore, fold through a series of meta-stable intermediate states.