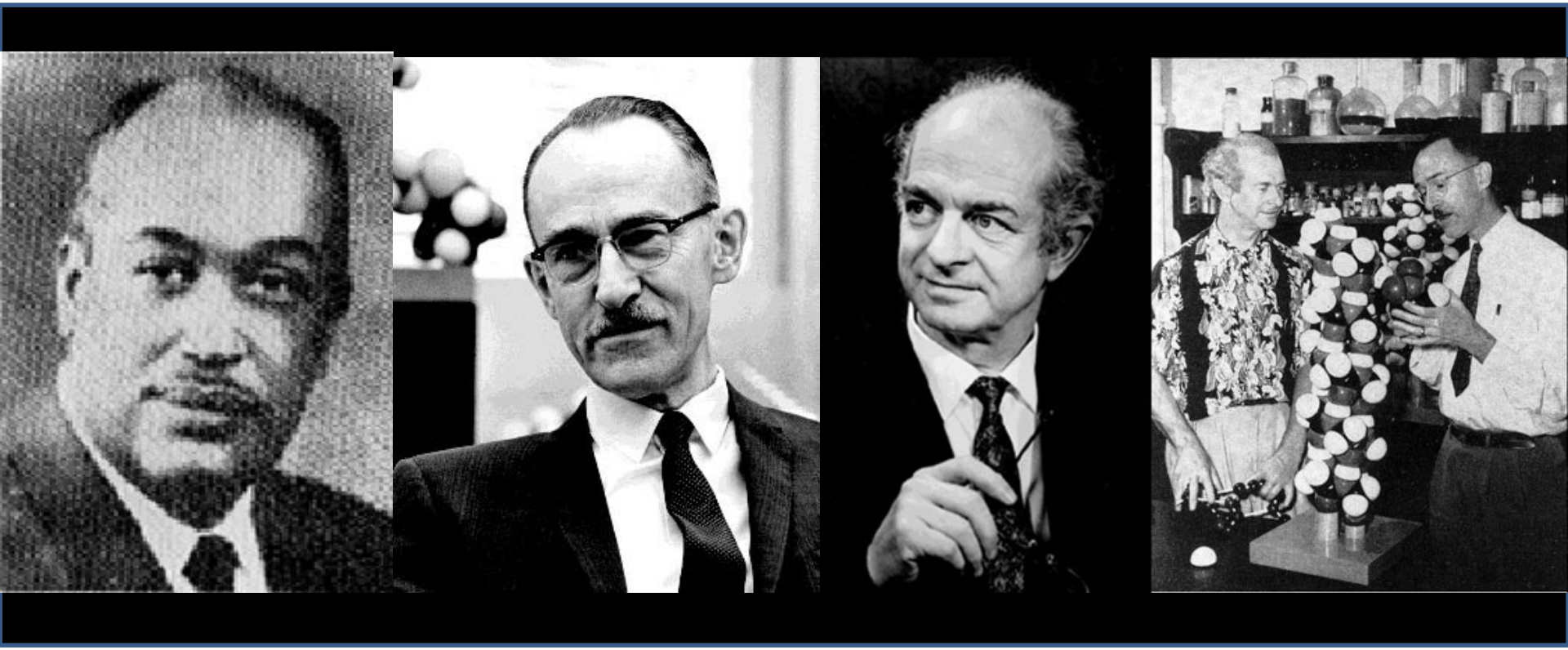
The structural organization within proteins

Kevin Slep

From Linear Peptide to a 3D Fold

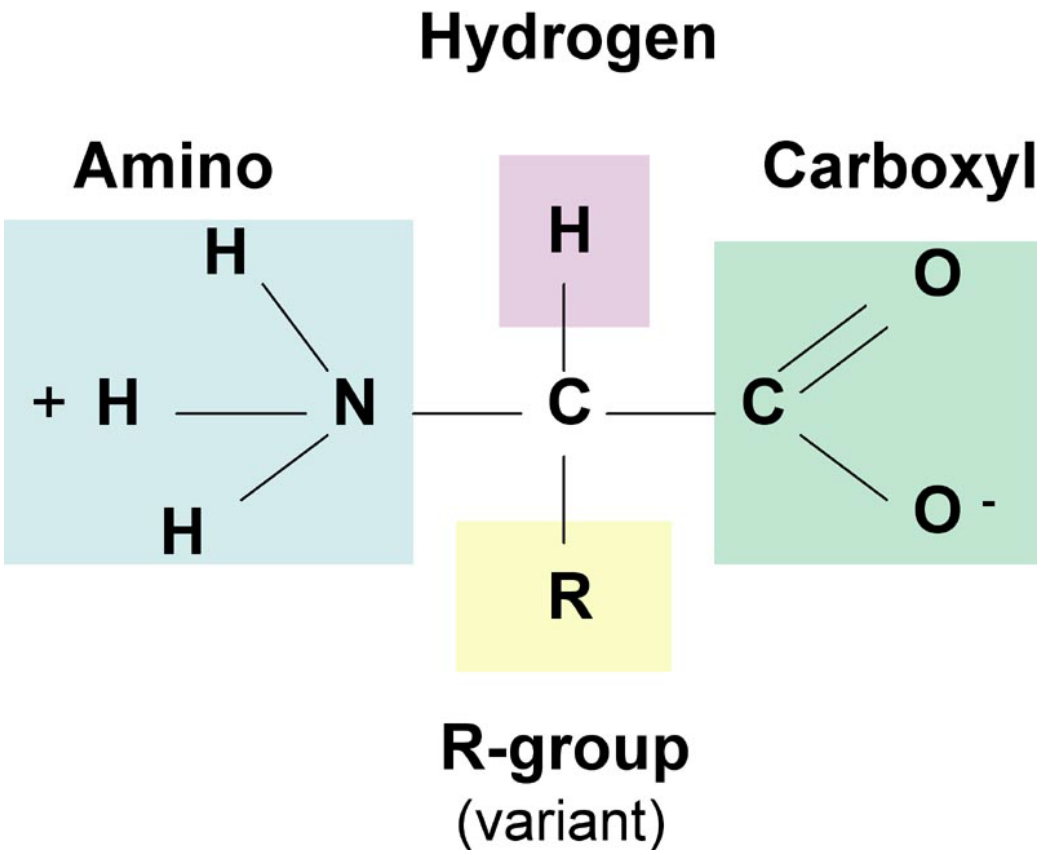
1951



Herman Branson Robert Corey Linus Pauling

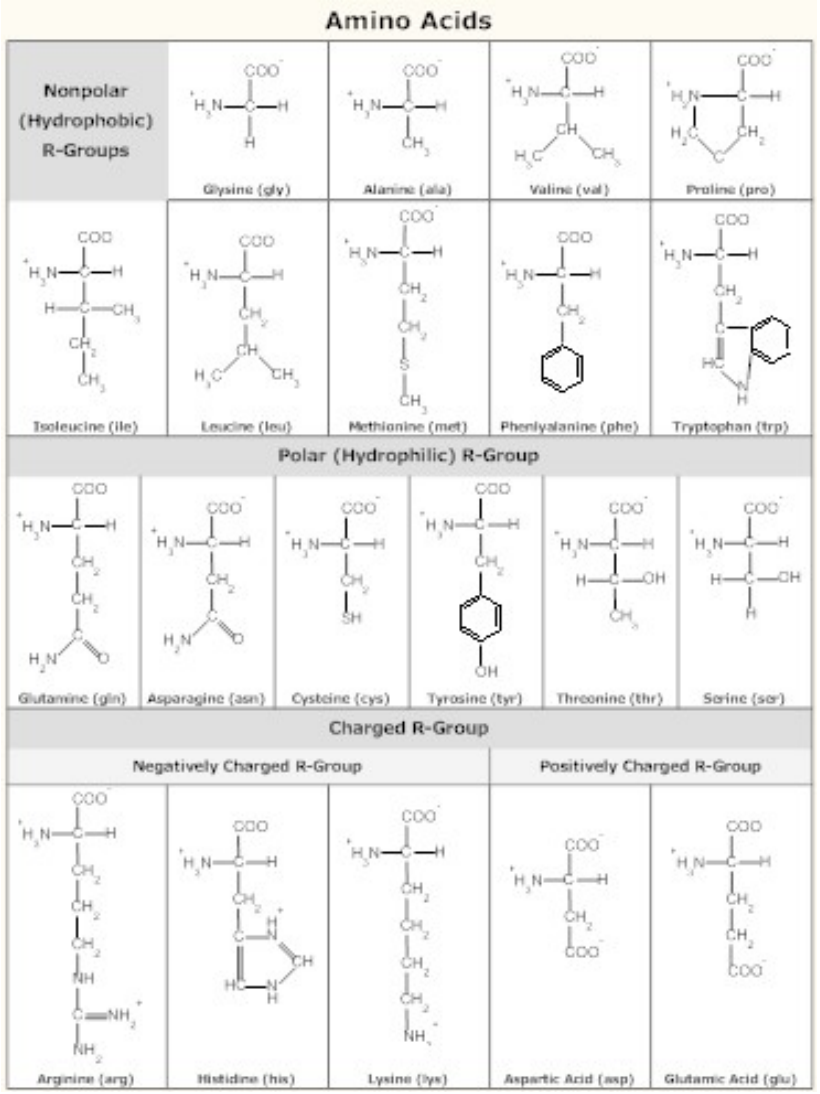
The Amino Acid

The Building Blocks of Proteins



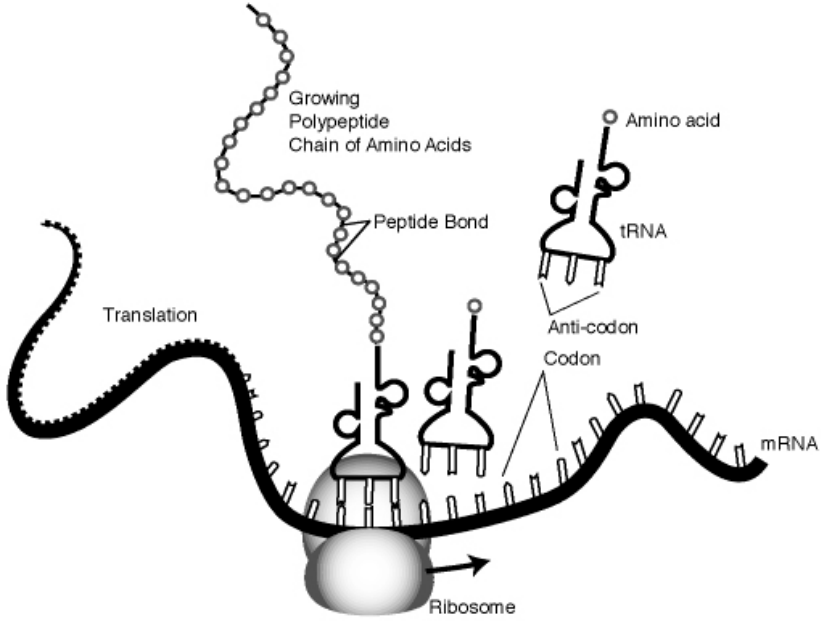
The 20 Amino Acids

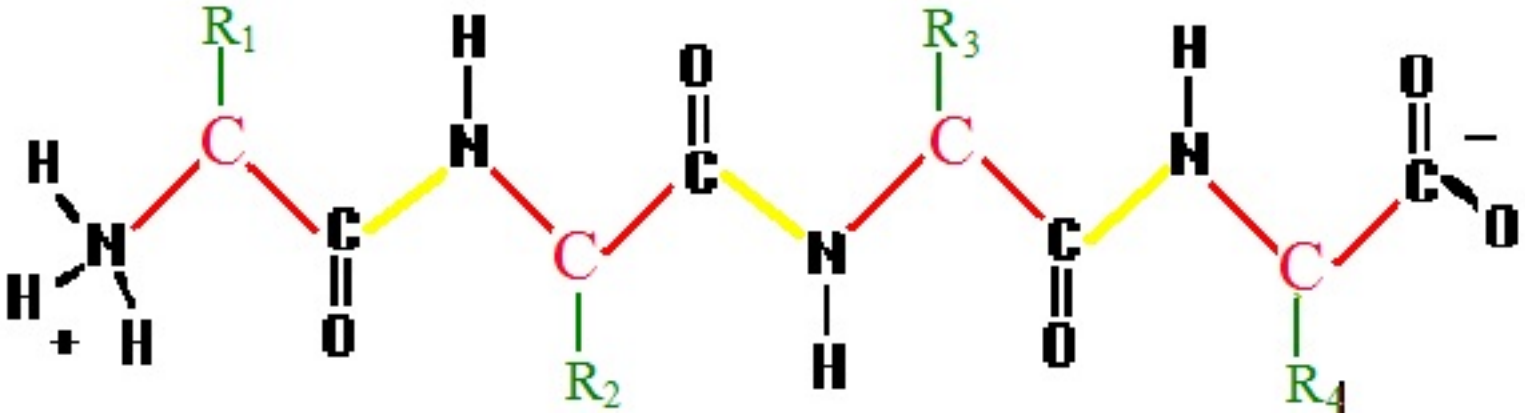
The Building Blocks of Proteins



The Peptide Chain, Primary Structure

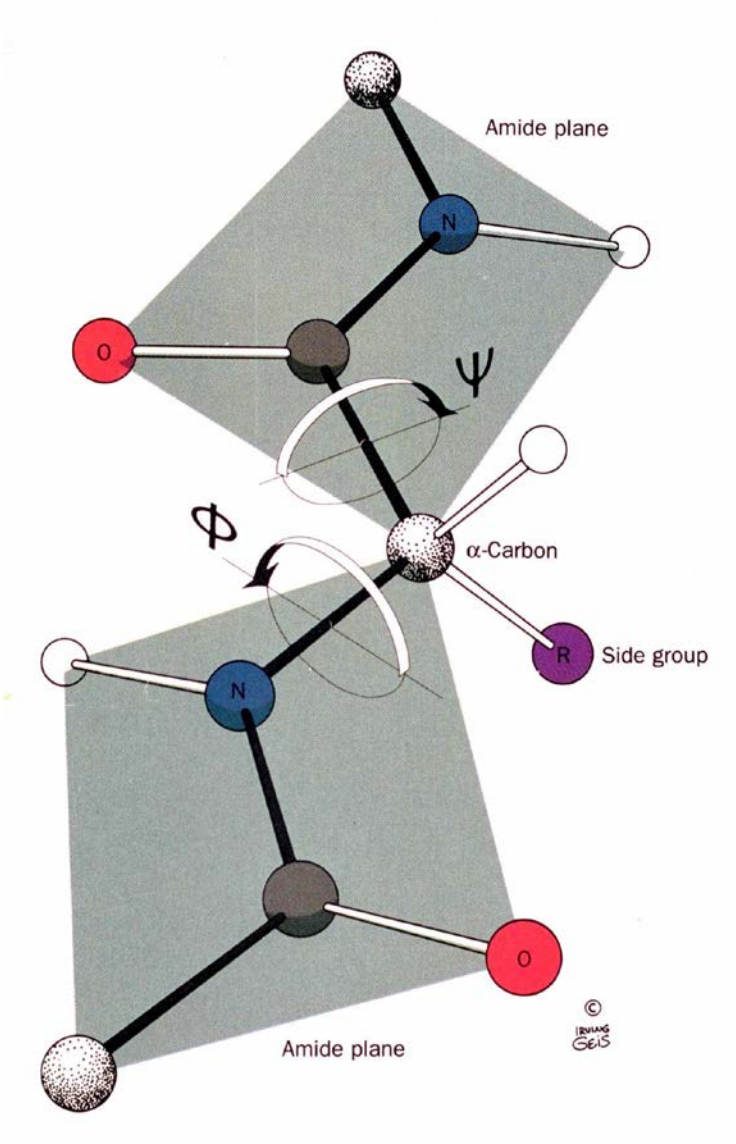
Assembled by the Ribosome During Translation





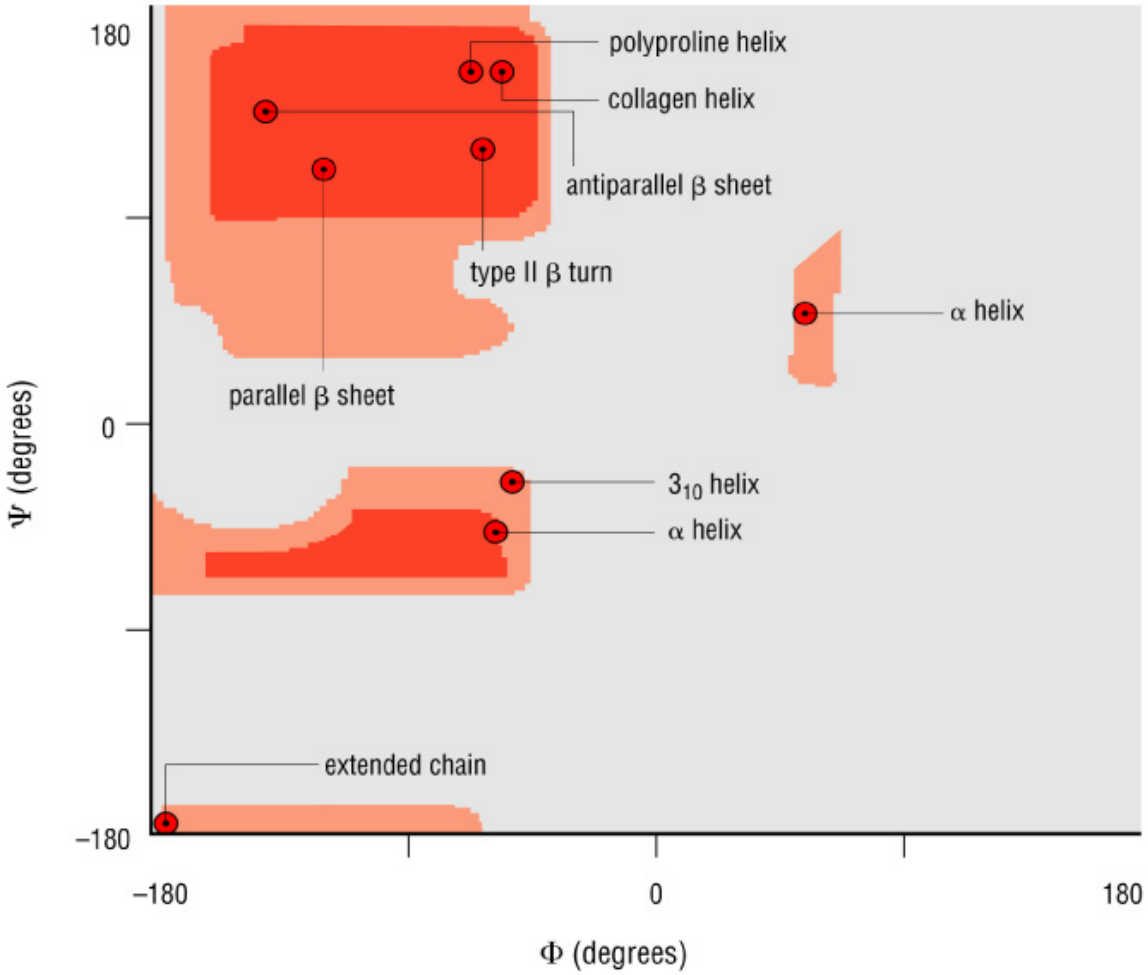
Peptide Backbone: Phi Psi Angles

Limits to their distribution



Ramachandran Plot

Energy limits to the Phi Psi Distribution

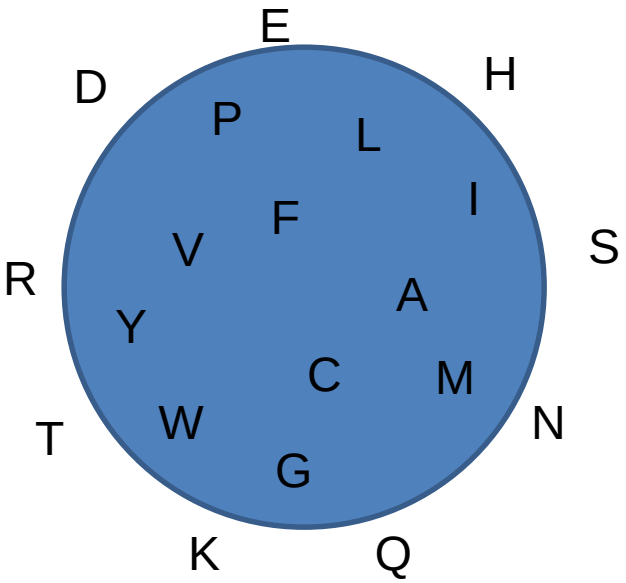


The Globular Protein Fold

Soluble Proteins:

Exterior: Hydrophillic

Interior: Hydrophobic



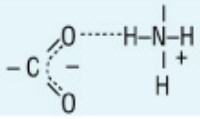
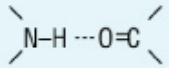
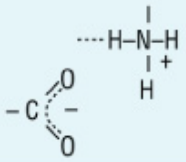
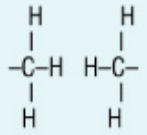
Packing

Cross Section of a Globular Protein

How would the organization of a Membrane Protein compare?

Interactions That Stabilize Protein Folds

Chemical Interactions that Stabilize Polypeptides

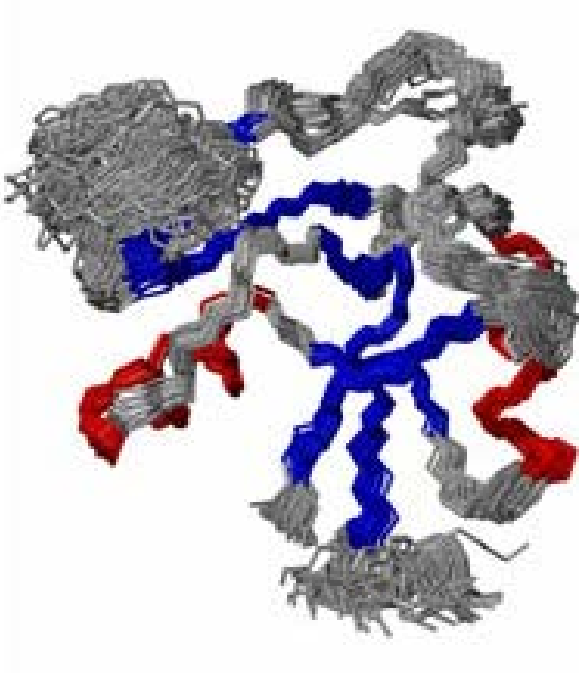
Interaction	Example	Distance dependence	Typical distance	Free energy (bond dissociation enthalpies for the covalent bonds)
Covalent bond	$-C_\alpha-C-$	-	1.5 Å	356 kJ/mole (610 kJ/mole for a C=C bond)
Disulfide bond	–Cys–S–S–Cys–	-	2.2 Å	167 kJ/mole
Salt bridge		Donor (here N), and acceptor (here O) atoms <3.5 Å	2.8 Å	12.5–17 kJ/mole; may be as high as 30 kJ/mole for fully or partially buried salt bridges (see text), less if the salt bridge is external
Hydrogen bond		Donor (here N), and acceptor (here O) atoms <3.5 Å	3.0 Å	2–6 kJ/mole in water; 12.5–21 kJ/mole if either donor or acceptor is charged
Long-range electrostatic interaction		Depends on dielectric constant of medium. Screened by water. 1/r dependence	Variable	Depends on distance and environment. Can be very strong in nonpolar region but very weak in water
Van der Waals interaction		Short range. Falls off rapidly beyond 4 Å separation. $1/r^6$ dependence	3.5 Å	4 kJ/mole (4–17 in protein interior) depending on the size of the group (for comparison, the average thermal energy of molecules at room temperature is 2.5 kJ/mole)

Secondary Structure
Helices
Strands
and Loops

First:
How do we determine structure?
Our Tools:

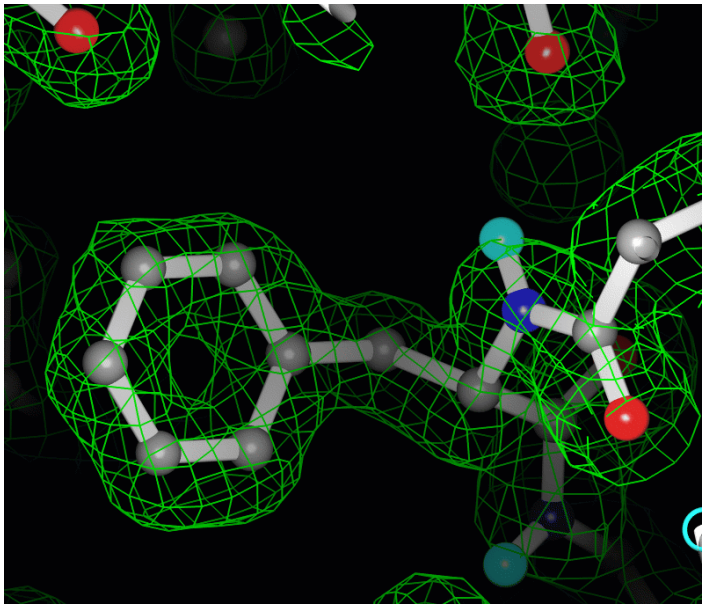
Solution NMR
Nuclear Magnetic Resonance

Distance Constraints
Dynamic Snapshot

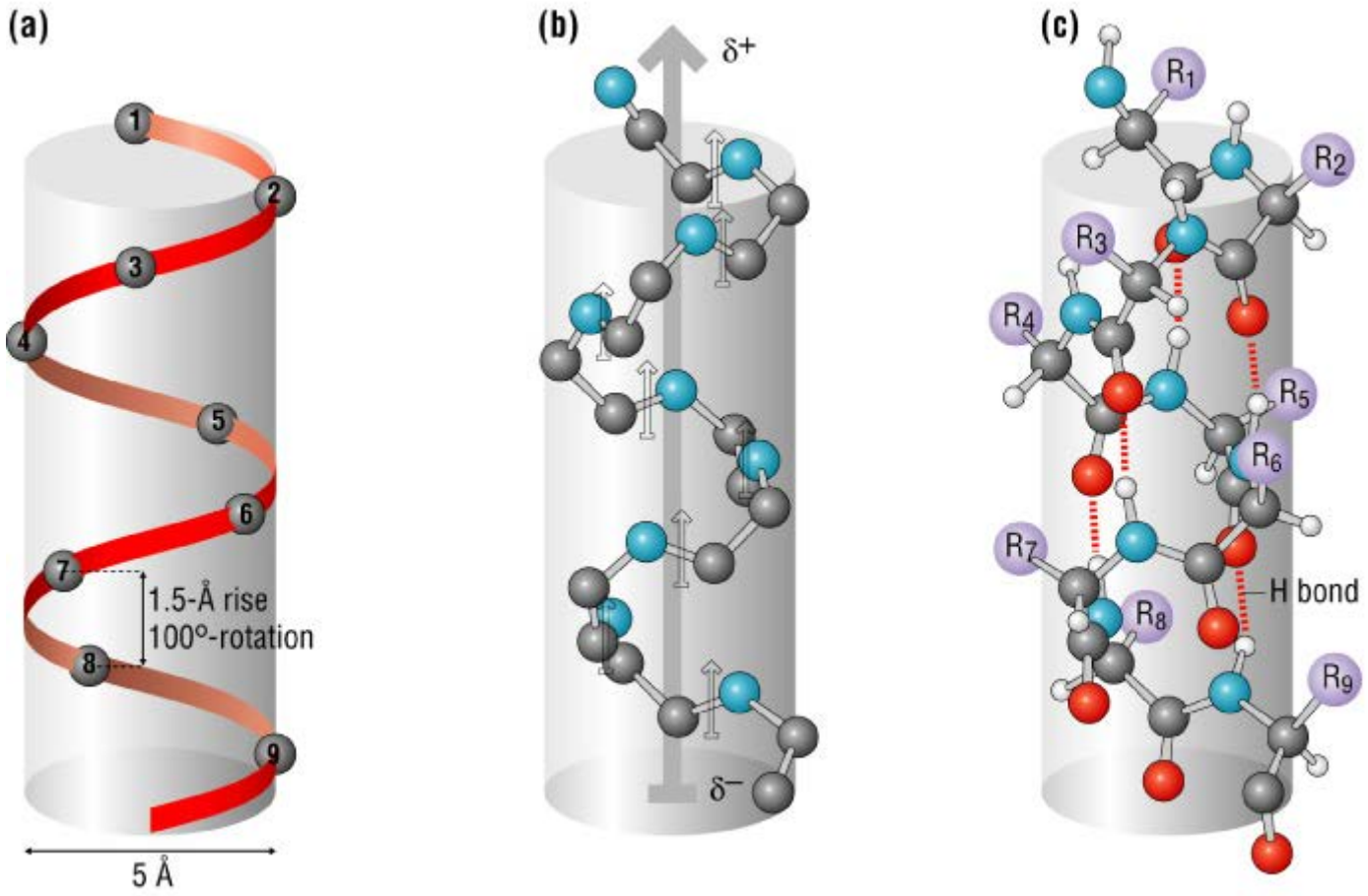


X-ray Crystallography

Electron Density
Static Snapshot

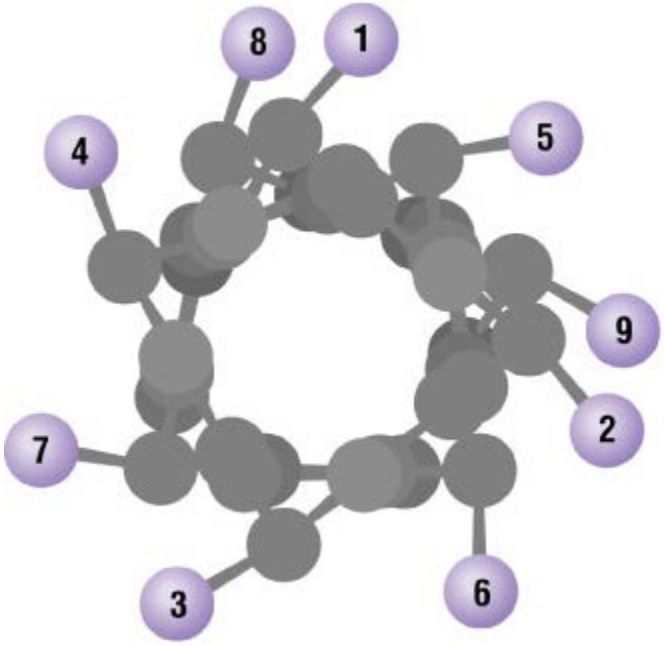


Secondary Structure: The α-Helix



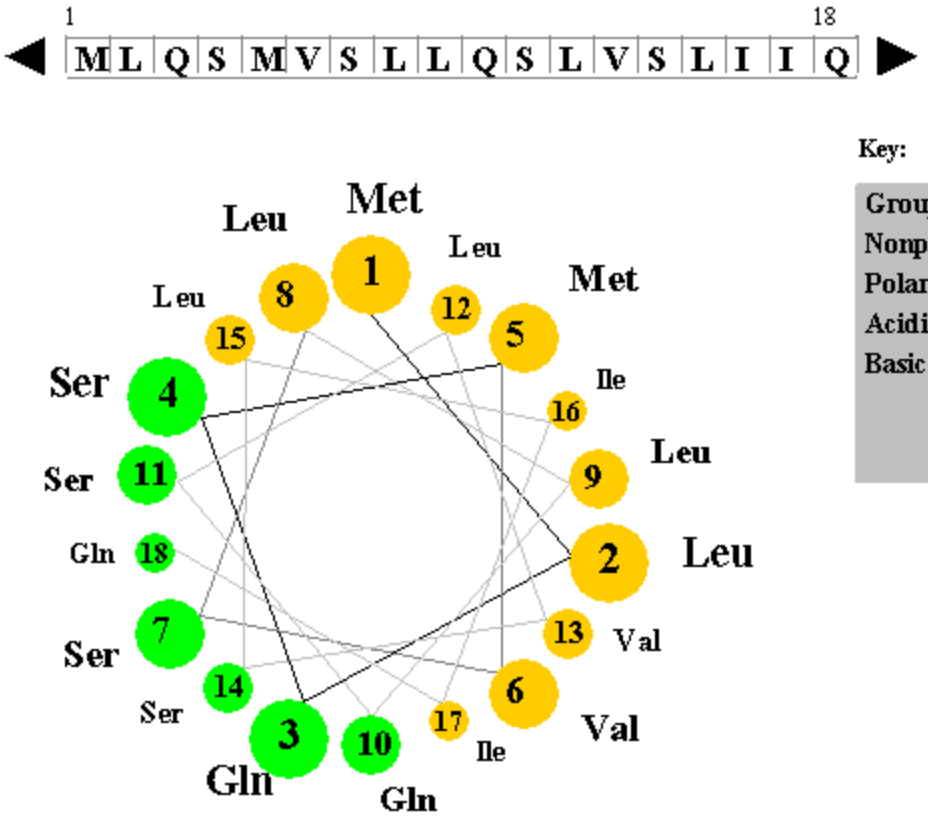
3.6 residues per turn

Secondary Structure: The α-Helix



Helical Axis

Secondary Structure: The α-Helix Helical Wheel – Hydropathy Plots

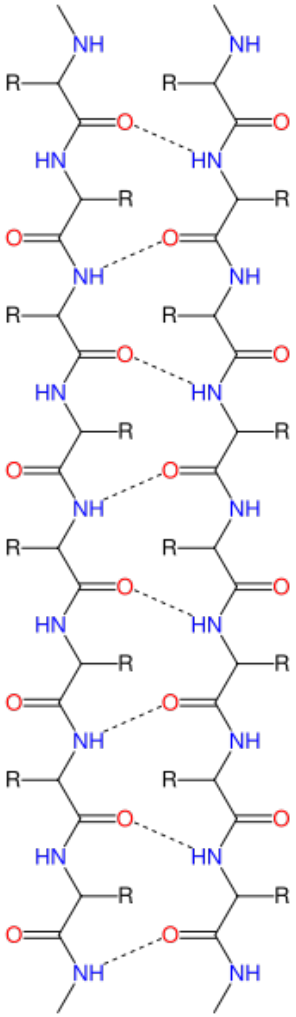


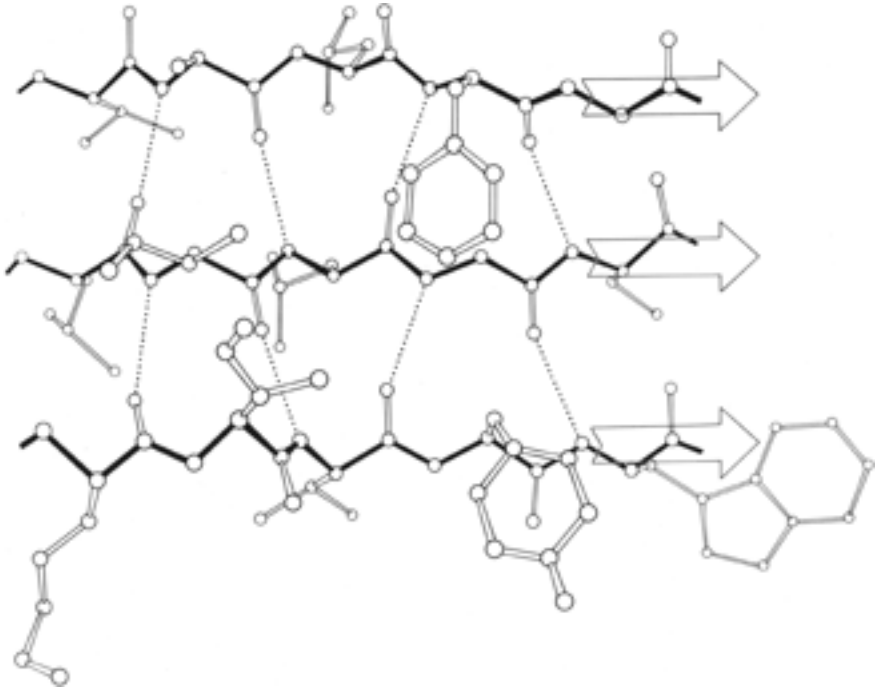
Helical Axis

Forms of Helices

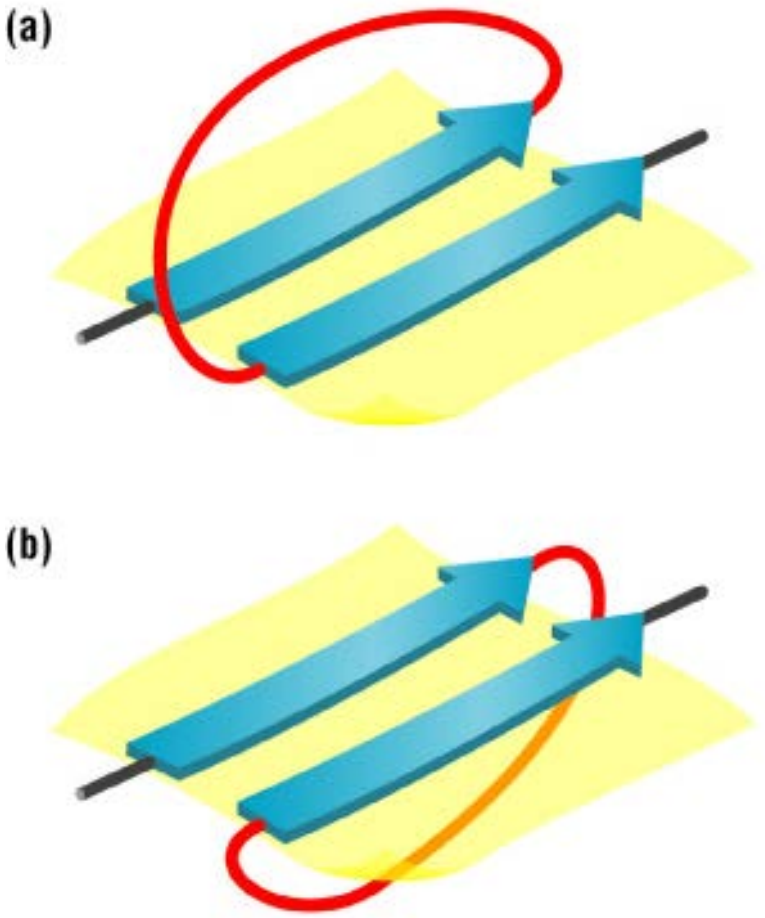
Average Conformational Parameters of Helical Elements					
Conformation	**Phi**	**Psi**	**Omega**	**Residues per turn**	**Translation per residue**
Alpha helix	–57	–47	180	3.6	1.5
3-10 helix	–49	–26	180	3.0	2.0
Pi-helix	57	–70	180	4.4	1.15
Polyproline I	–83	+158	0	3.33	1.9
Polyproline II	–78	+149	180	3.0	3.12
Polyproline III	–80	+150	180	3.0	3.1

Secondary Structure:
The β-Sheet: Parallel β-Strands

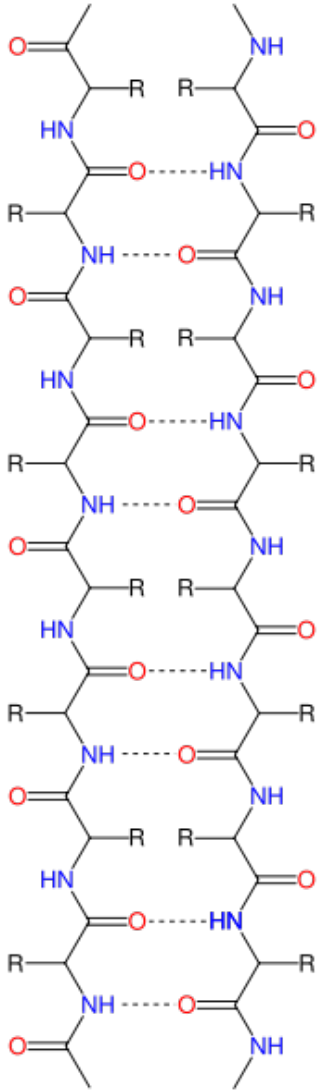


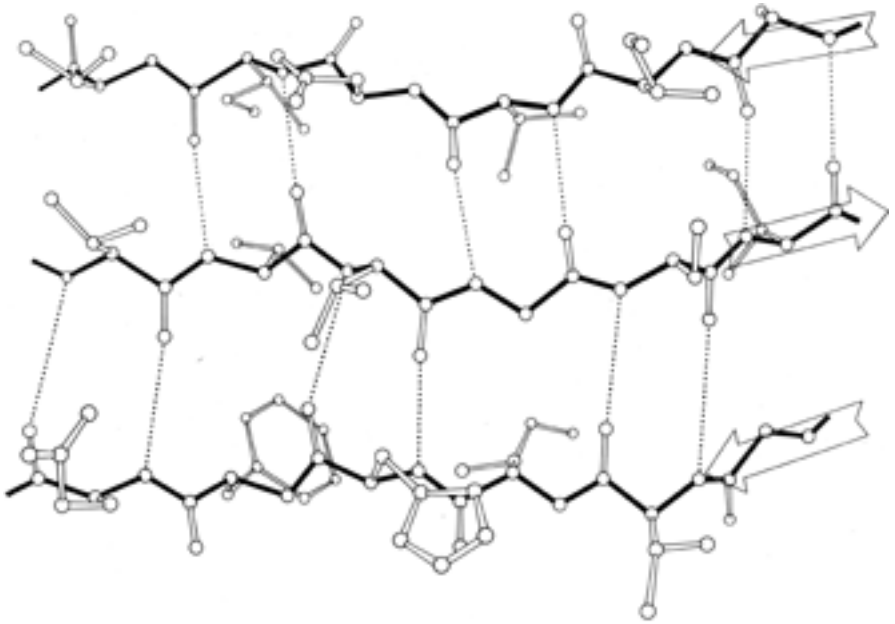


Secondary Structure:
The β-Sheet: Parallel β-Strands



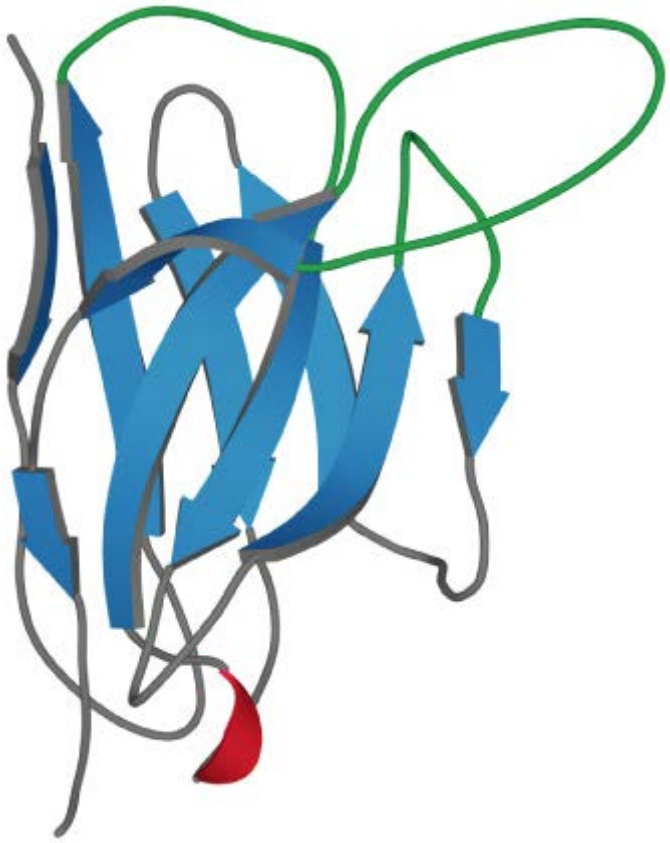
Secondary Structure:
The β-Sheet: Anti-Parallel β-Strands





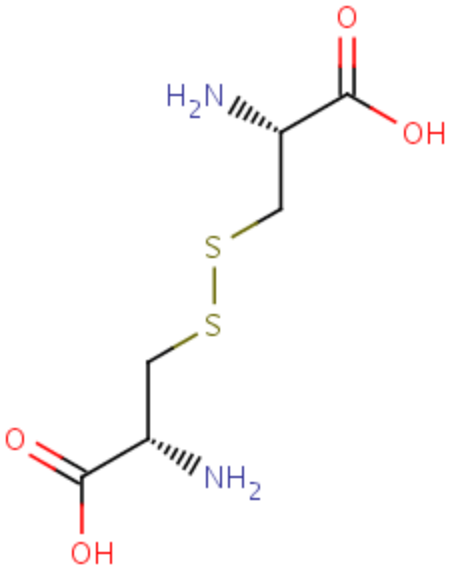
Secondary Structure: Loops

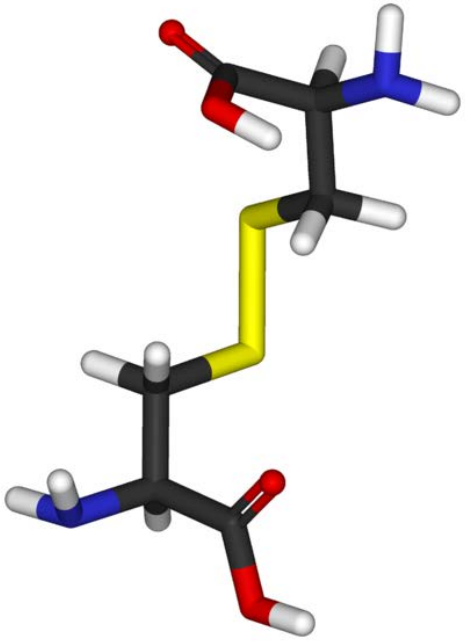
Linkers, Variable Structrure, Active Site

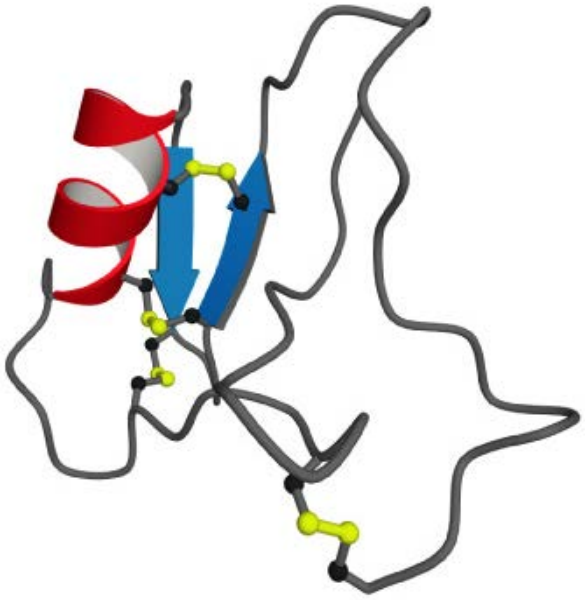


Cystine: The Disulfide Bond

Two Cysteines Covalently Bond (Oxidation)
Provides Added Stabilization to a Fold







Secondary Structure Prediction Tools

Online Sites:

Predict Protein
http://www.predictprotein.org/

Phyre
http://www.sbg.bio.ic.ac.uk/phyre/

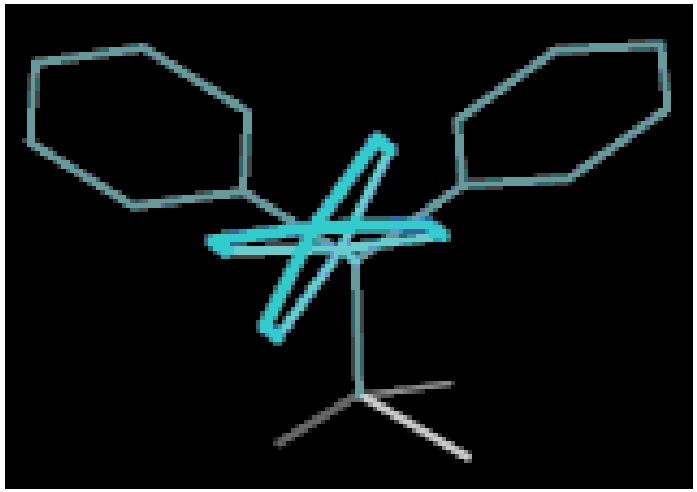
Jpred3
http://www.compbio.dundee.ac.uk/~www-jpred/

Secondary Structure:

Conformational Preferences of the Amino Acids

Amino acid	Preference		
	α-helix	β-strand	Reverse turn
Glu	**1.59**	0.52	1.01
Ala	**1.41**	0.72	0.82
Leu	**1.34**	1.22	0.57
Met	**1.30**	1.14	0.52
Gln	**1.27**	0.98	0.84
Lys	**1.23**	0.69	1.07
Arg	**1.21**	0.84	0.90
His	**1.05**	0.80	0.81
Val	0.90	**1.87**	0.41
Ile	1.09	**1.67**	0.47
Tyr	0.74	**1.45**	0.76
Cys	0.66	**1.40**	0.54
Trp	1.02	**1.35**	0.65
Phe	1.16	**1.33**	0.59
Thr	0.76	**1.17**	0.90
Gly	0.43	0.58	**1.77**
Asn	0.76	0.48	**1.34**
Pro	0.34	0.31	**1.32**
Ser	0.57	0.96	**1.22**
Asp	0.99	0.39	**1.24**

Structure is Conferred by Main Chain Conformation and Side Chain Conformation – Packing Rotamers: Favored Geometries/Energy States



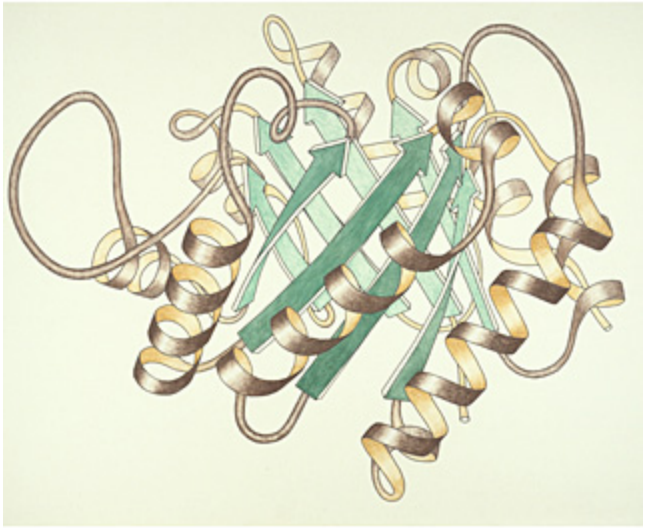
Phenylalanine

How Secondary Structure is Organized:

Motifs and Tertiary Structure

Topology Diagrams

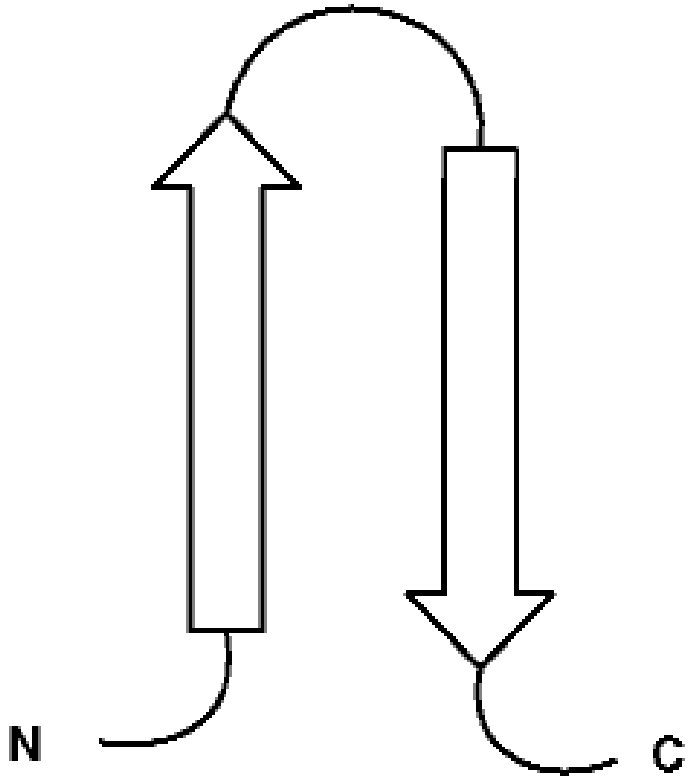
Jane Richardson (Duke Univ.)



Triose Phosphate Isomerase

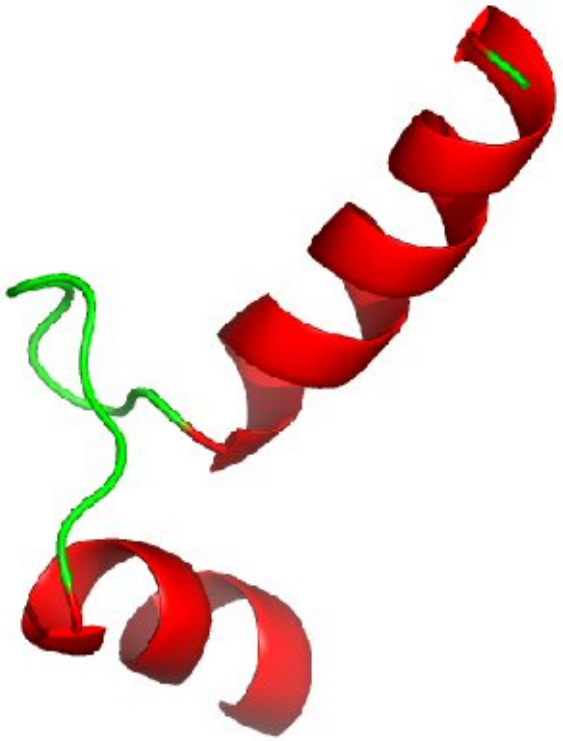
Structural Motifs

β Hairpin



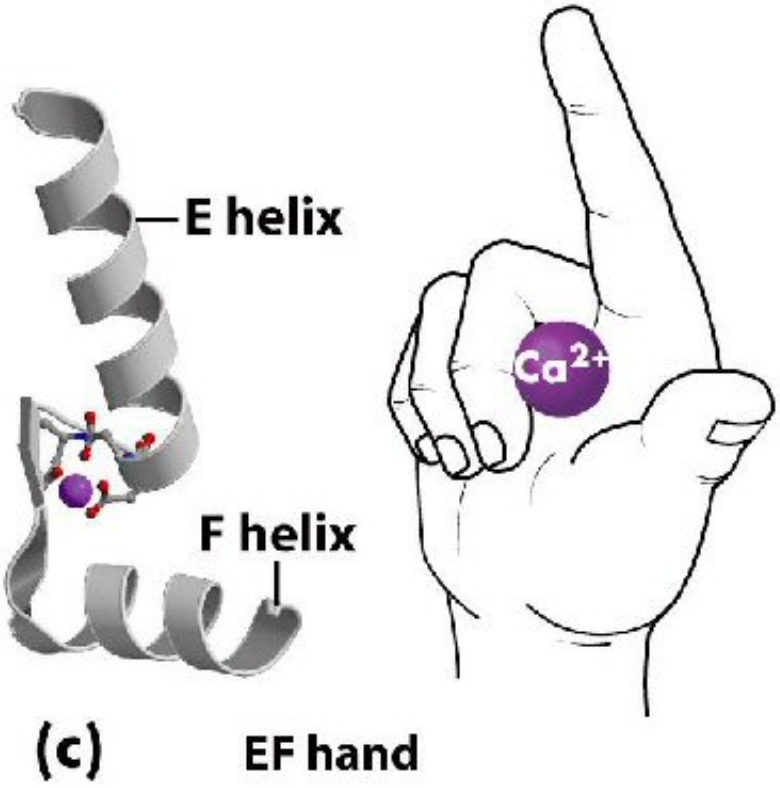
Structural Motifs

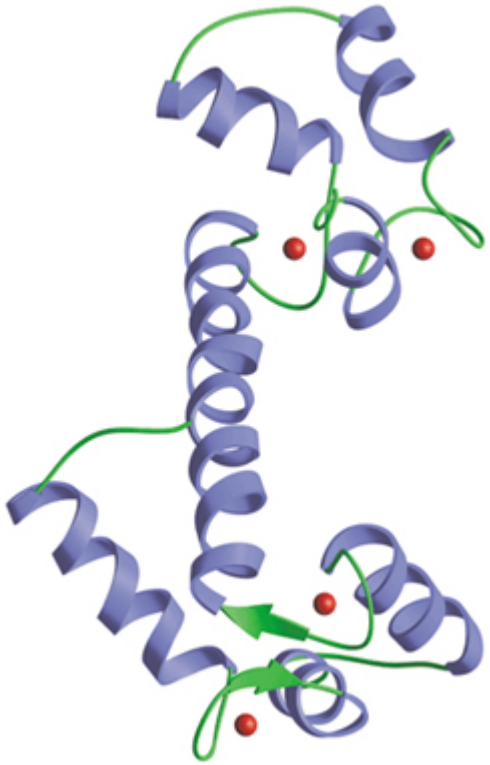
Helix-Loop-Helix



Structural Motifs

EF-Hand





Calmodulin: 4 EF Hands

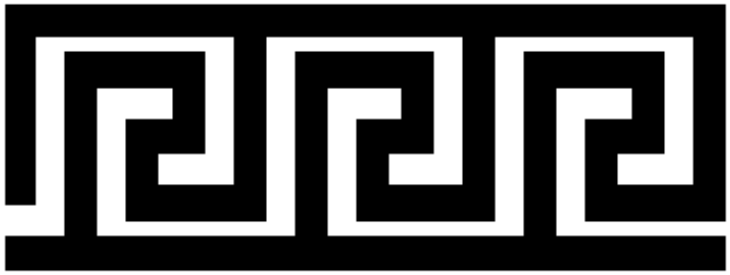
Structural Motifs

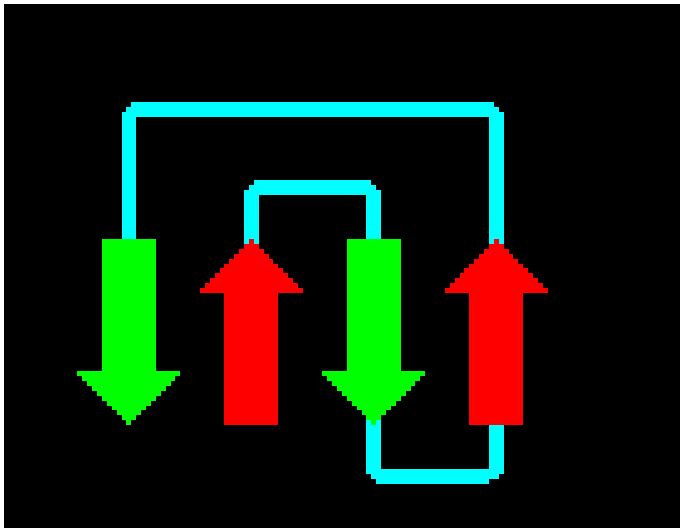
Zinc_Finger

Two Histidines on a Helix
Two Cysteines on a Loop

Structural Motifs

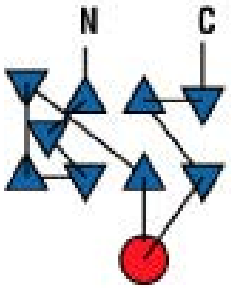
Greek Key





Structural Motifs

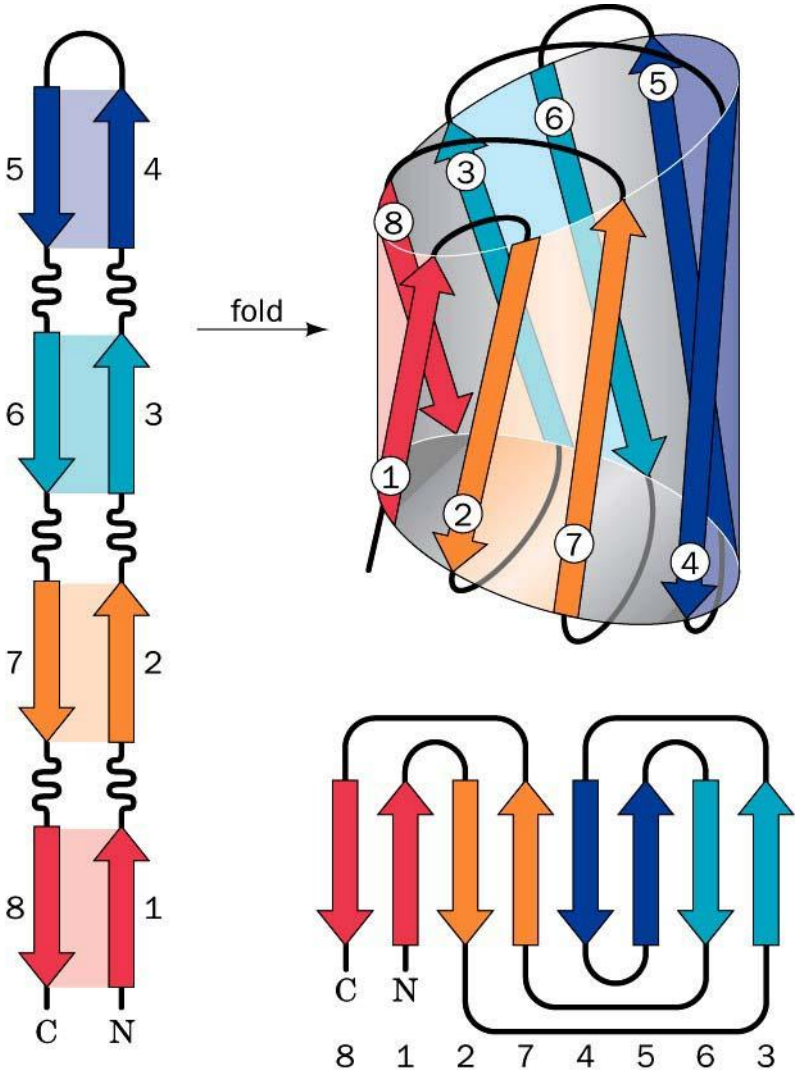
Greek Key



Pre-albumin

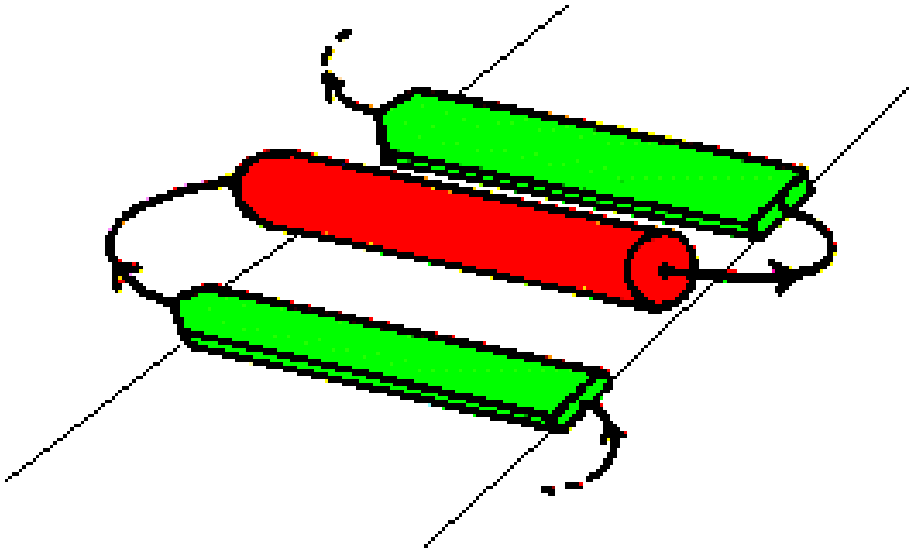
Structural Motifs

Jelly Roll



Structural Motifs

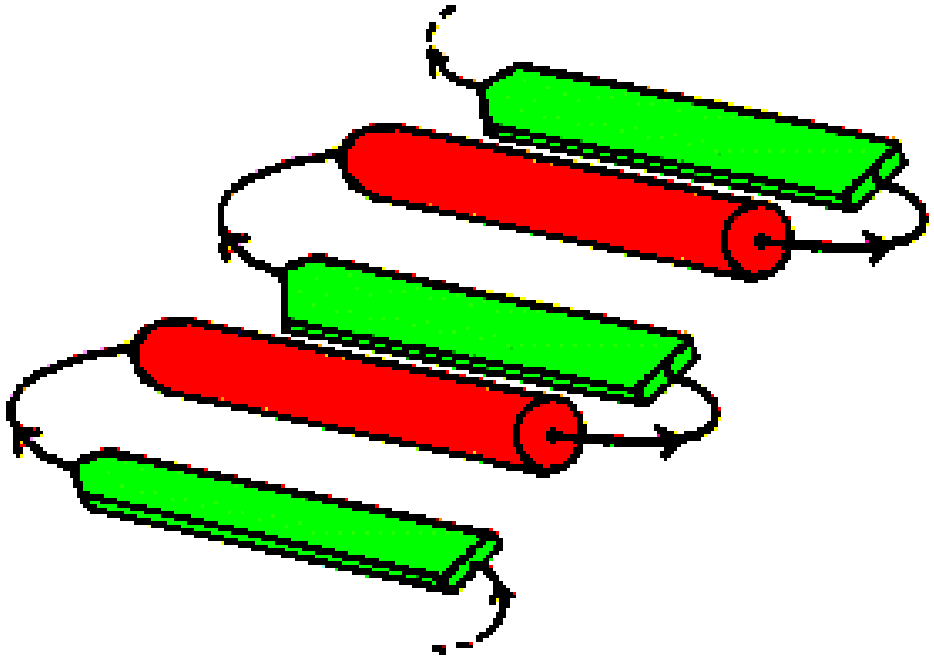
$\beta-\alpha-\beta$

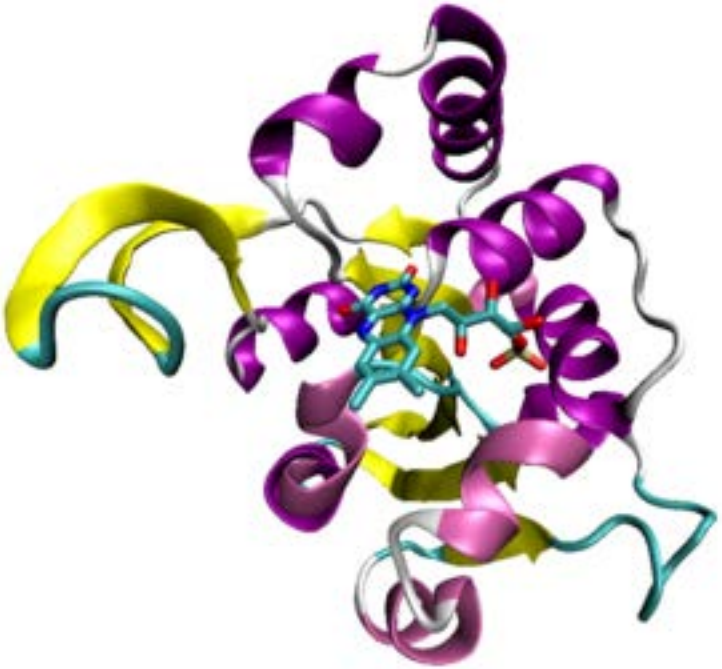


The right–handed beta–alpha–beta unit. The helix lies above the plane of the strands.

Structural Motifs

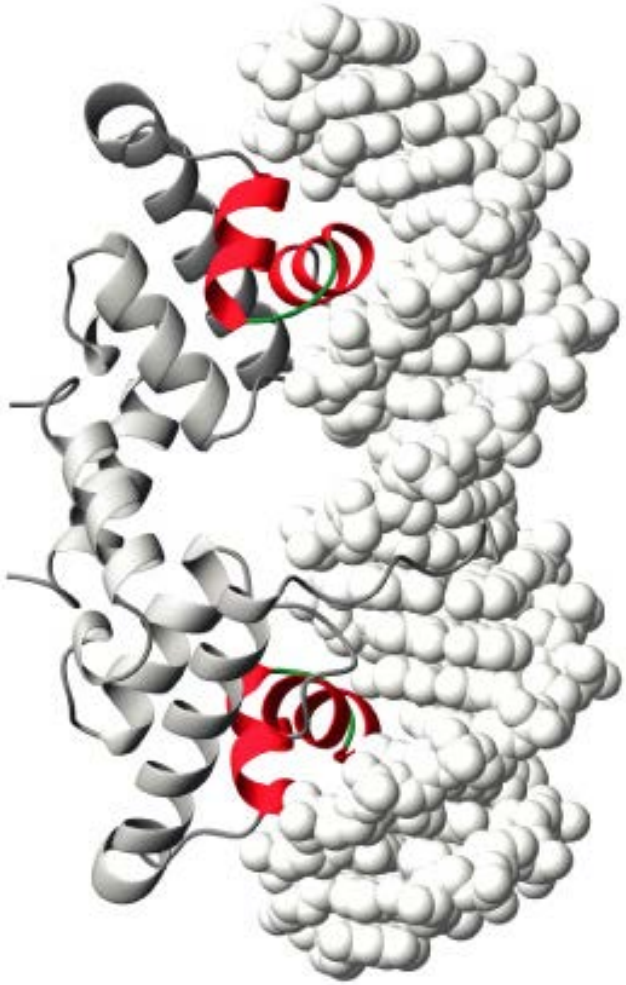
Rossmann Fold



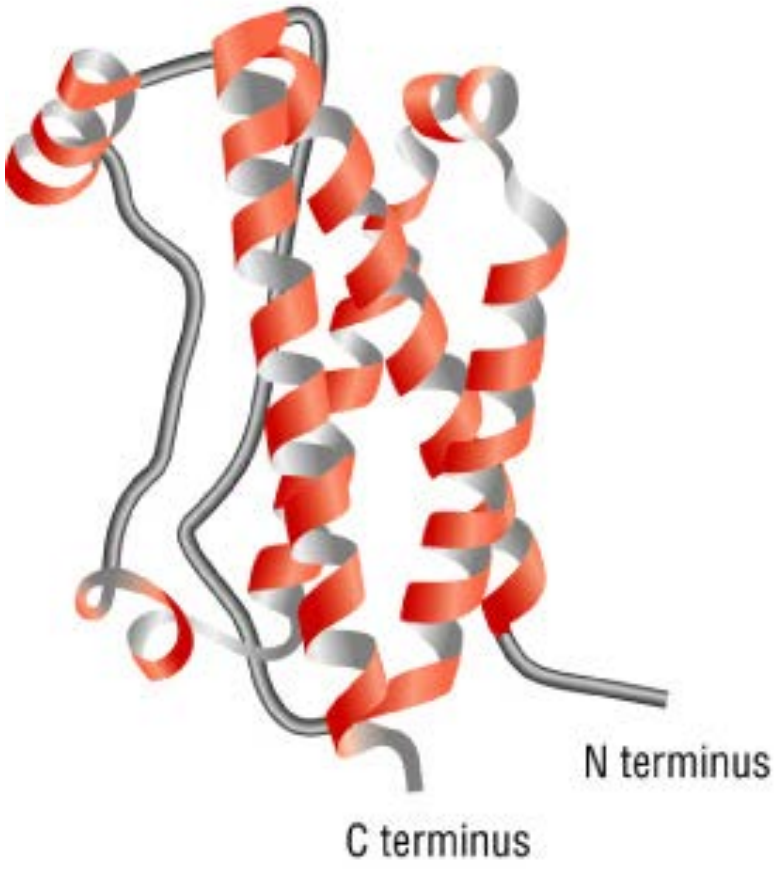


Nucleotide Binding

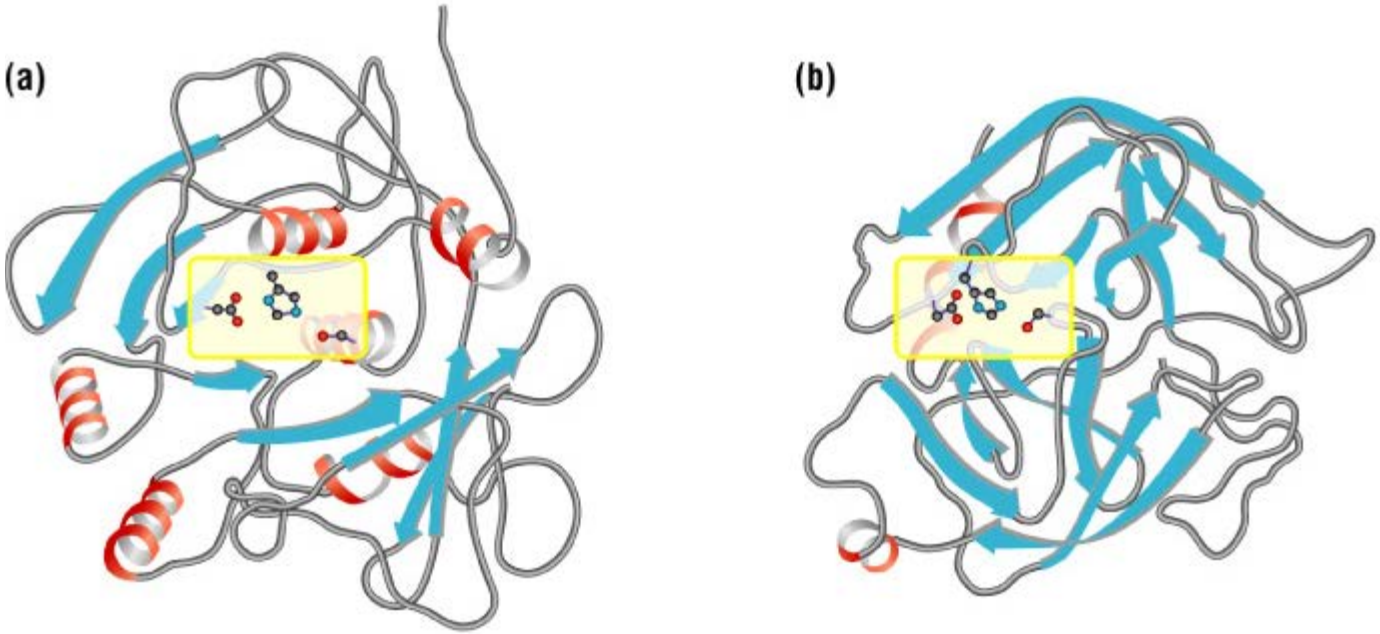
Helix-Turn-Helix
DNA Binding Proteins



Four-Helix Bundle

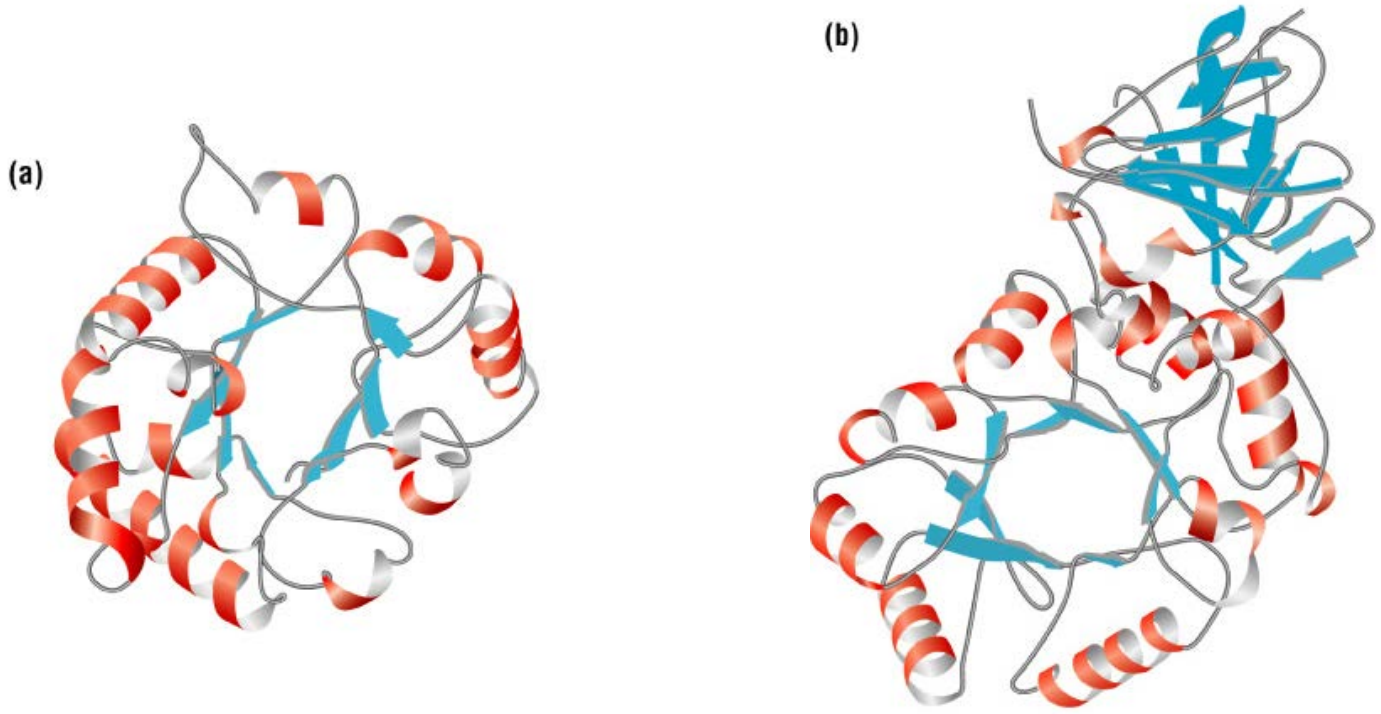


Catalytic Triad

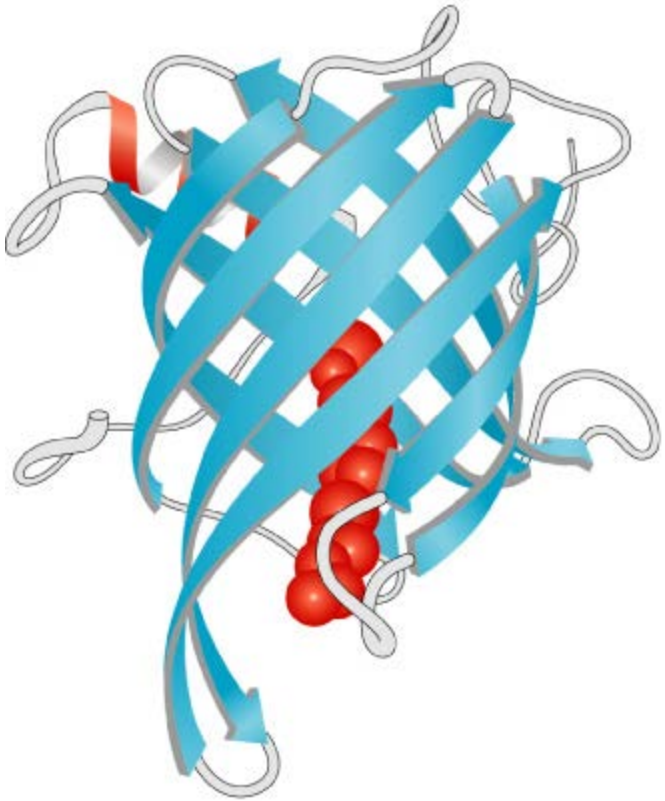


Aspartate, Histidine, Serine

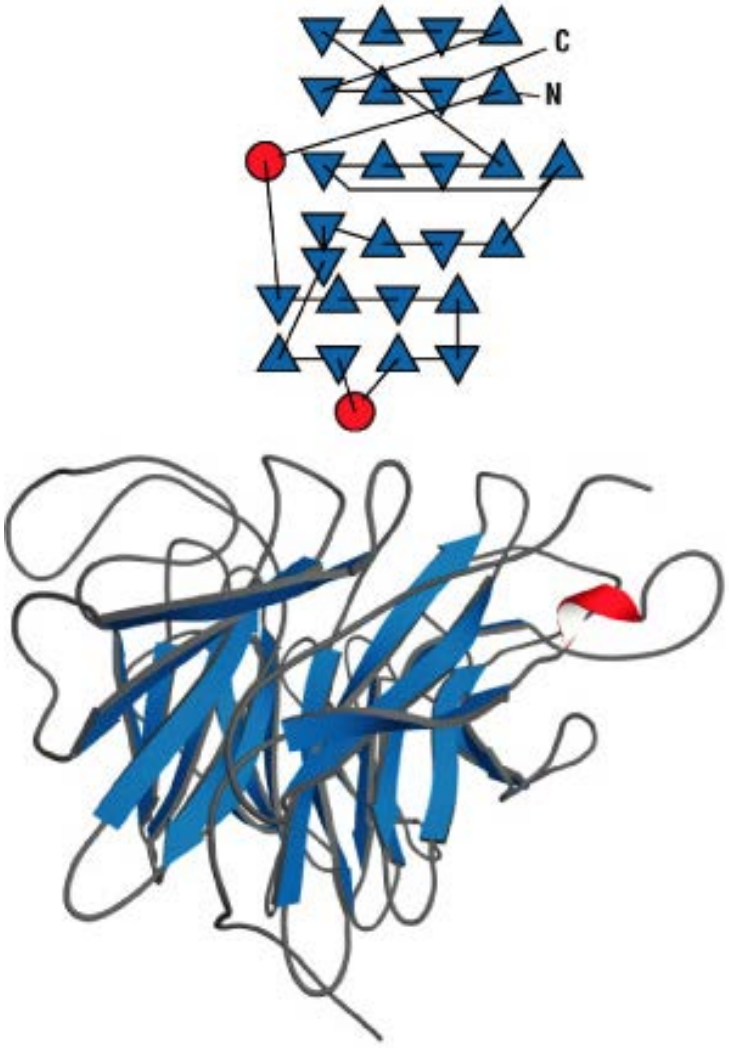
TIM Barrel Domain



β-Barrel

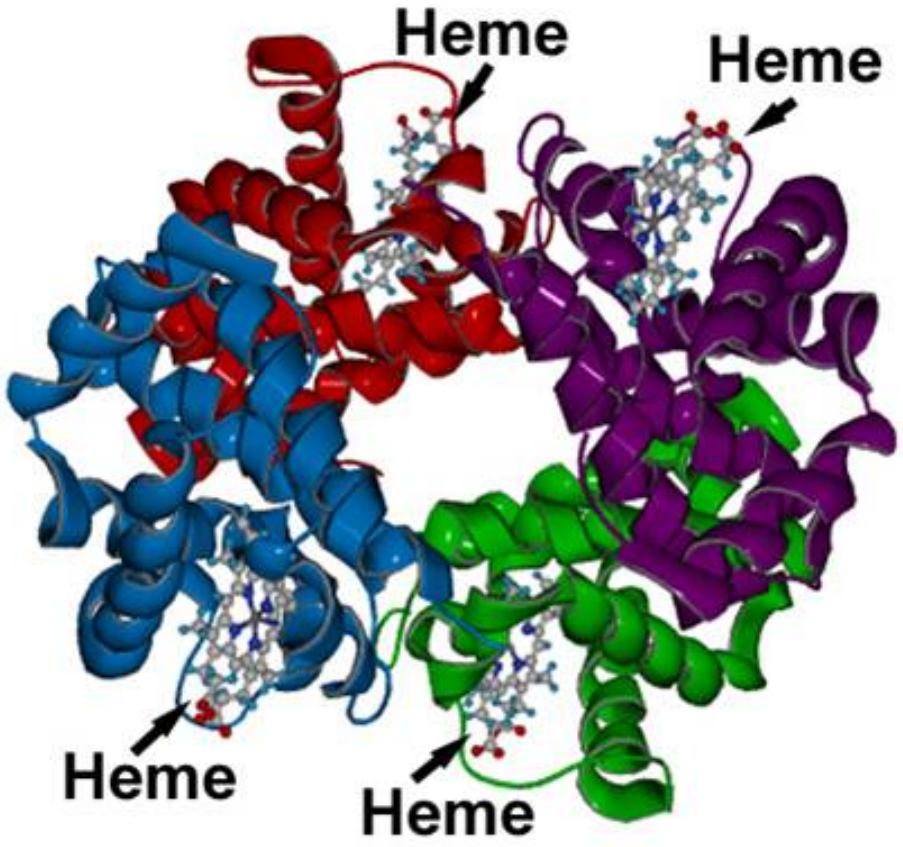


β-Propeller Domain



Neuraminidase

Non-Protein Molecules That Play a Role in Structure and Function

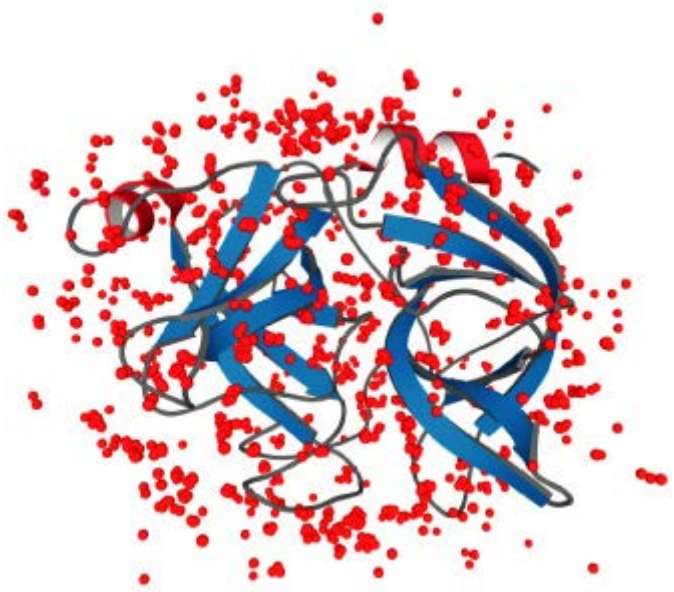


Cofactors

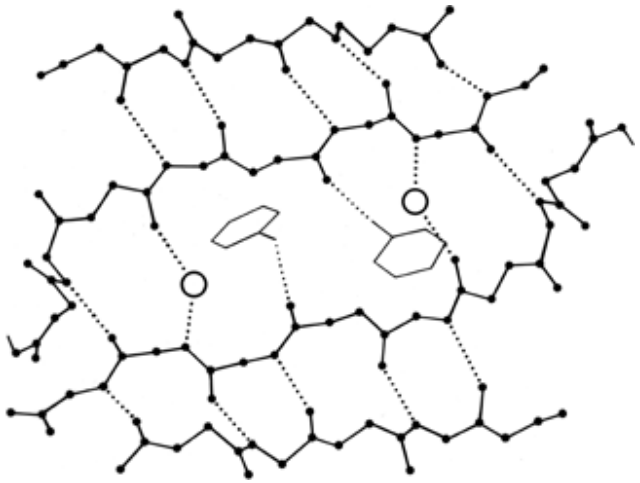
The Heme Group in Hemoglobin

Non-Protein Molecules That Play a Role in Structure and Function

Ordered Water

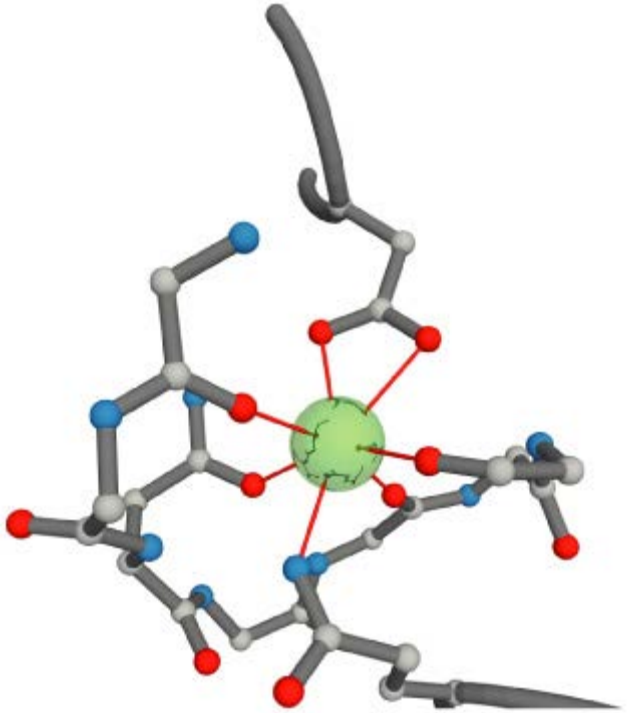


First Hydration Shell



Water in Prealbumin

Non-Protein Molecules That Play a Role in Structure and Function

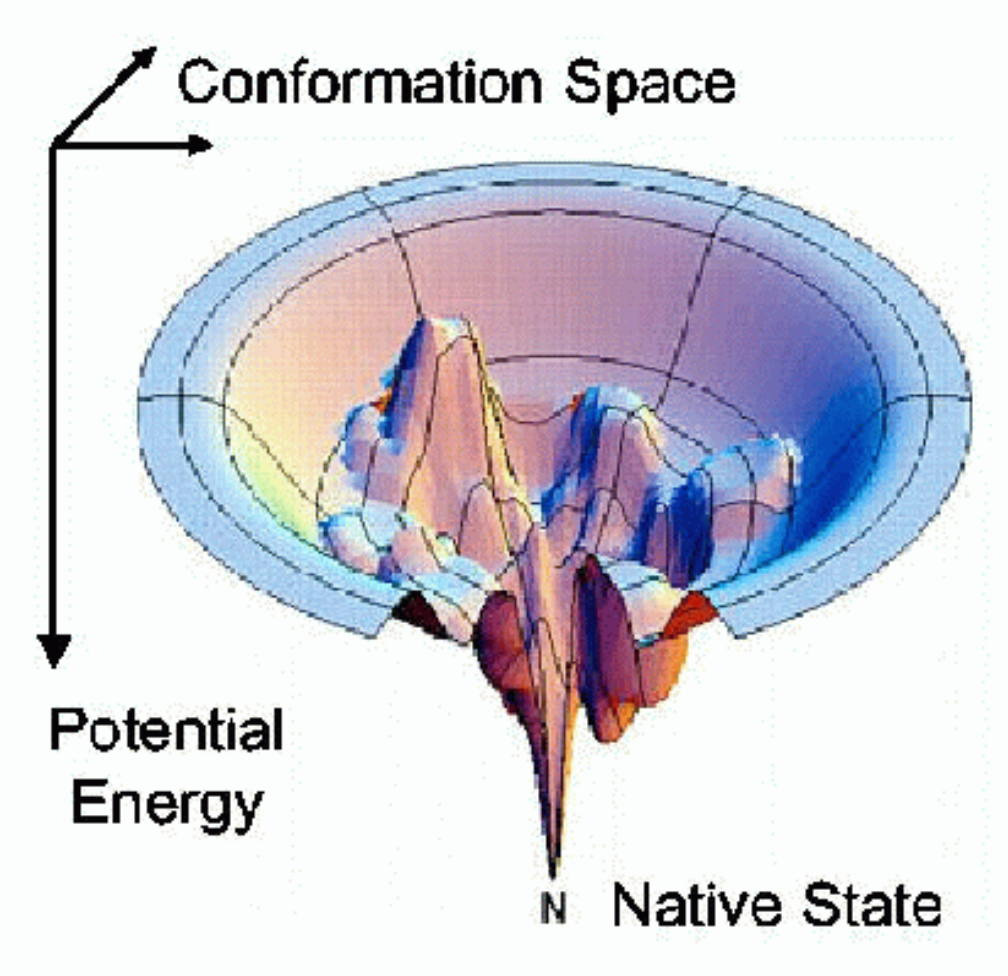


Water in Prealbumin

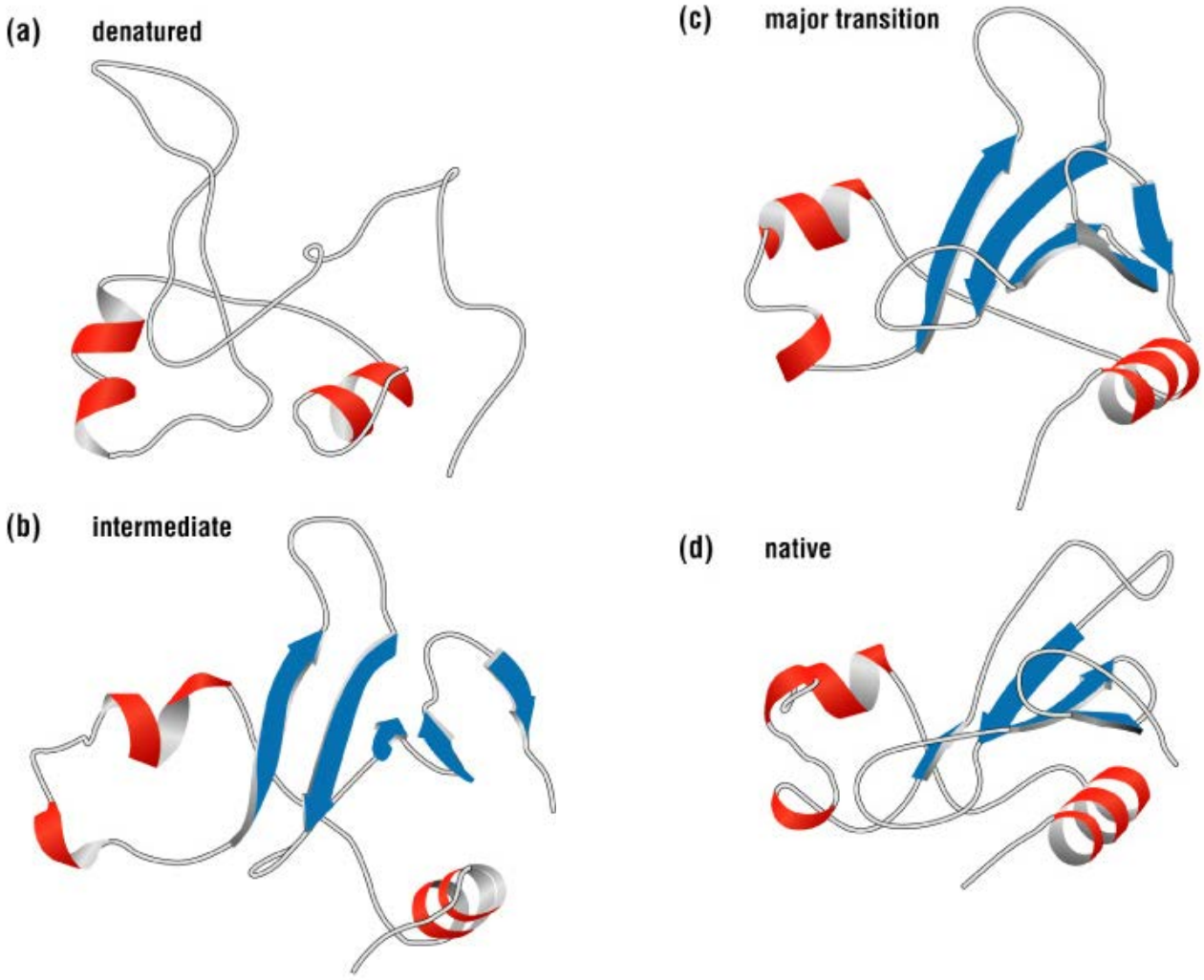
How Do Proteins Fold?

Moving from a linear peptide
To a Motif
To a Domain

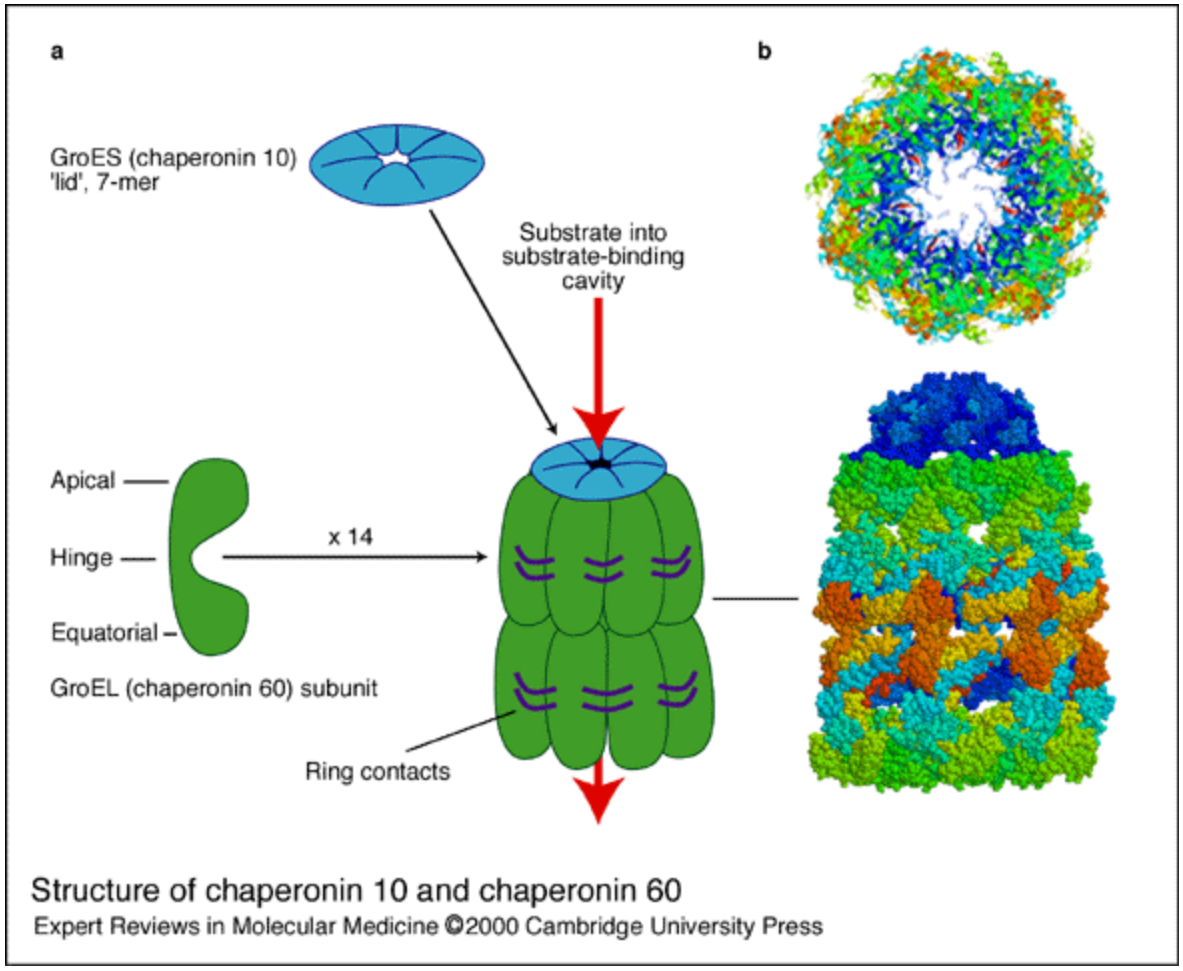
Protein Folding: Energy Landscape



Protein Folding: Pathways



Protein Folding: Help From Chaperones

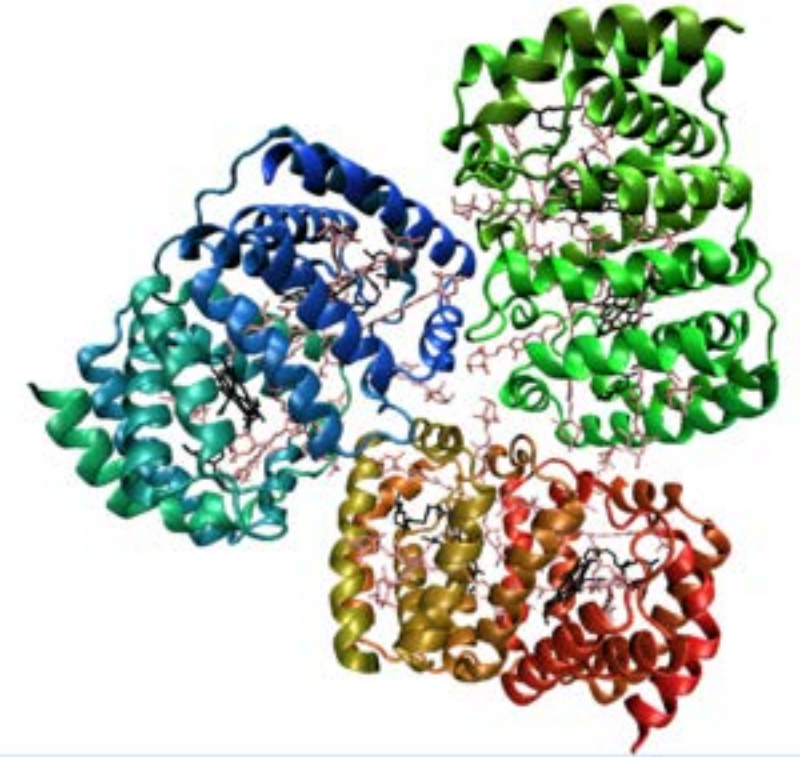


Structure of chaperonin 10 and chaperonin 60

Protein Domains Built from Motifs

α Domains

α solenoid – a curved structure

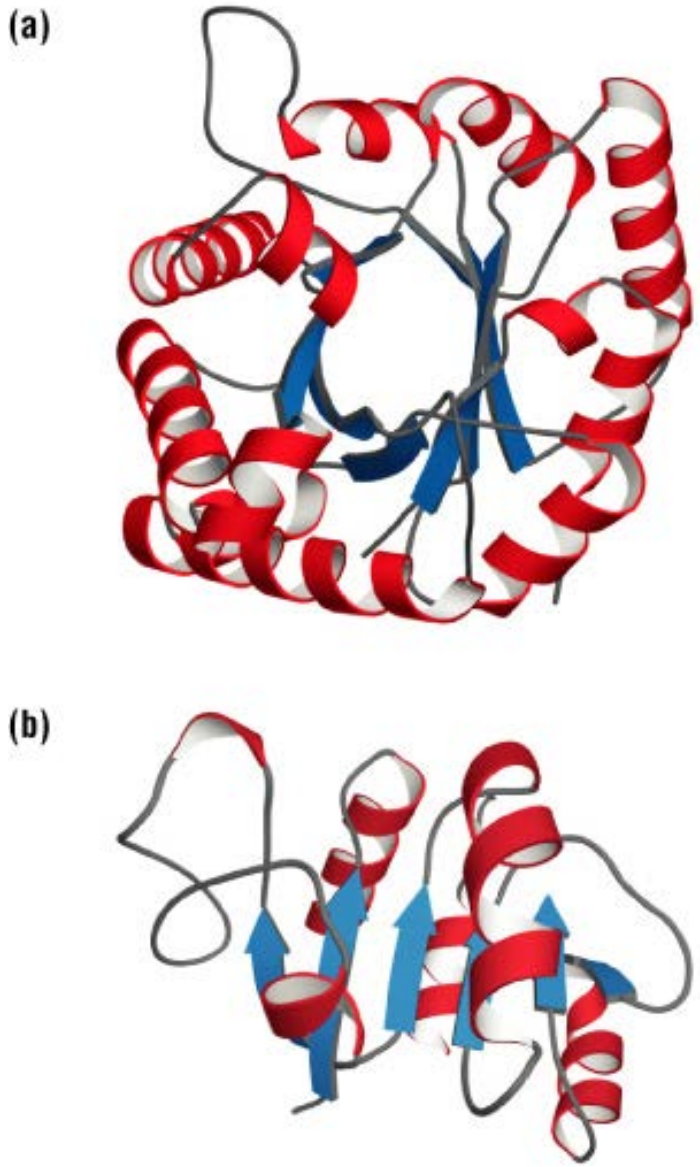


peridinin-chlorophyll-containing protein - trimer

β Domains

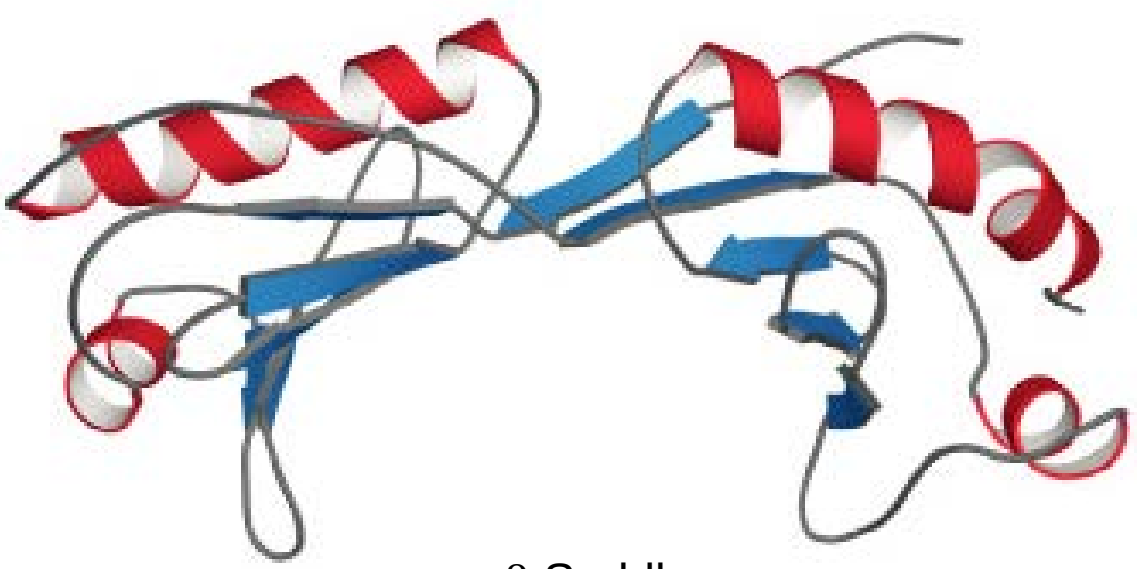
WD40 protein

α/β Domains



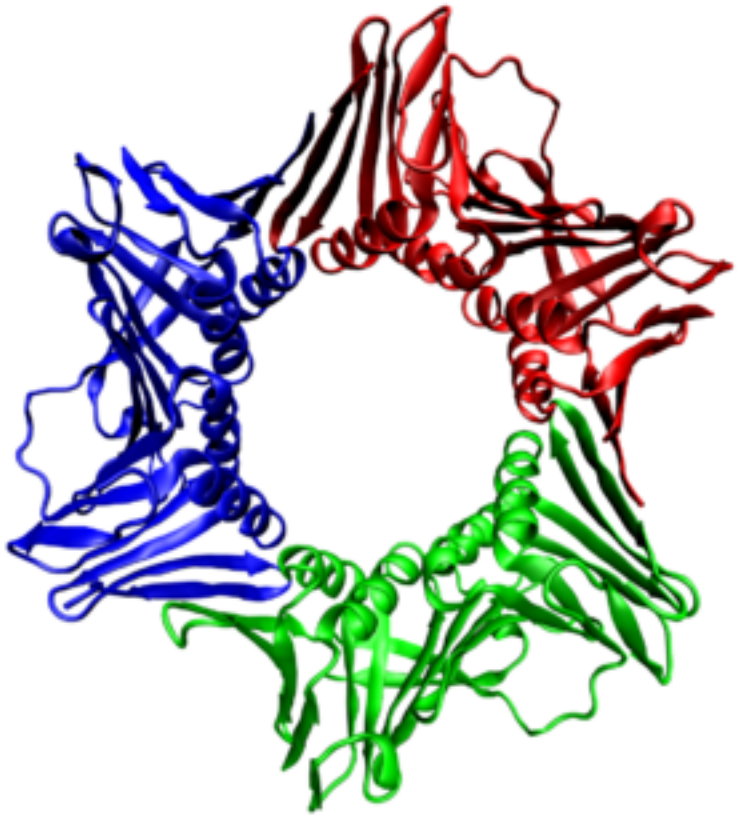
Helices and Strands
Alternate in the peptide

α/β Domains



α–β Saddle

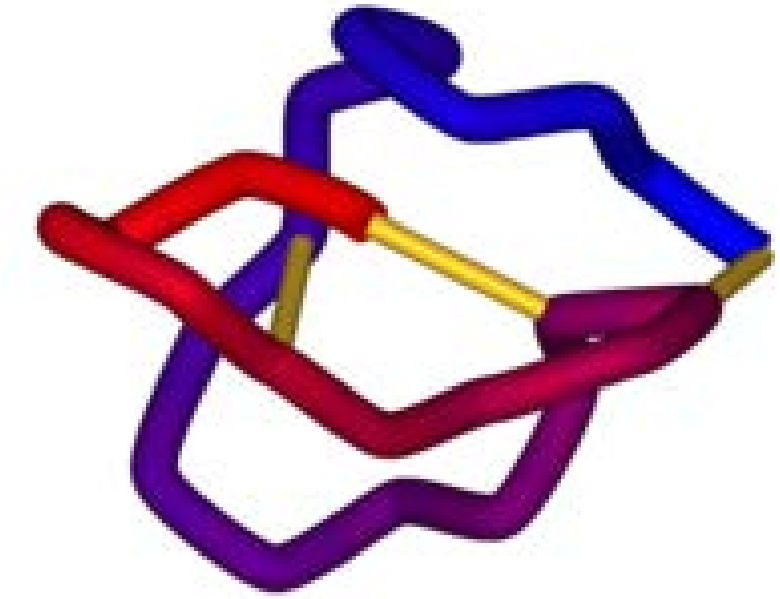
α and β Domains



DNA Clamp
PCNA

Helices and Strands
Are separated in the peptide

Irregular Folds

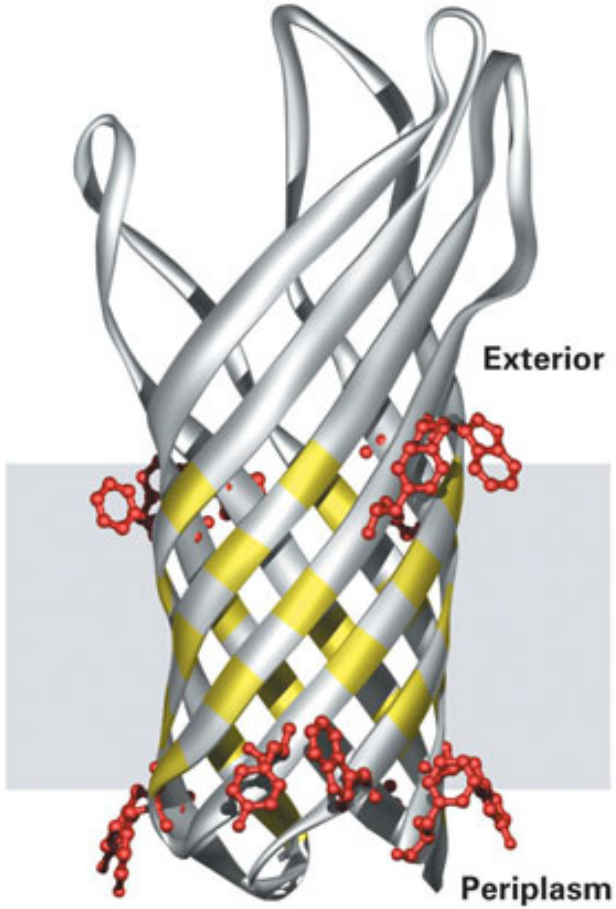


Conotoxin, disulfide in yellow

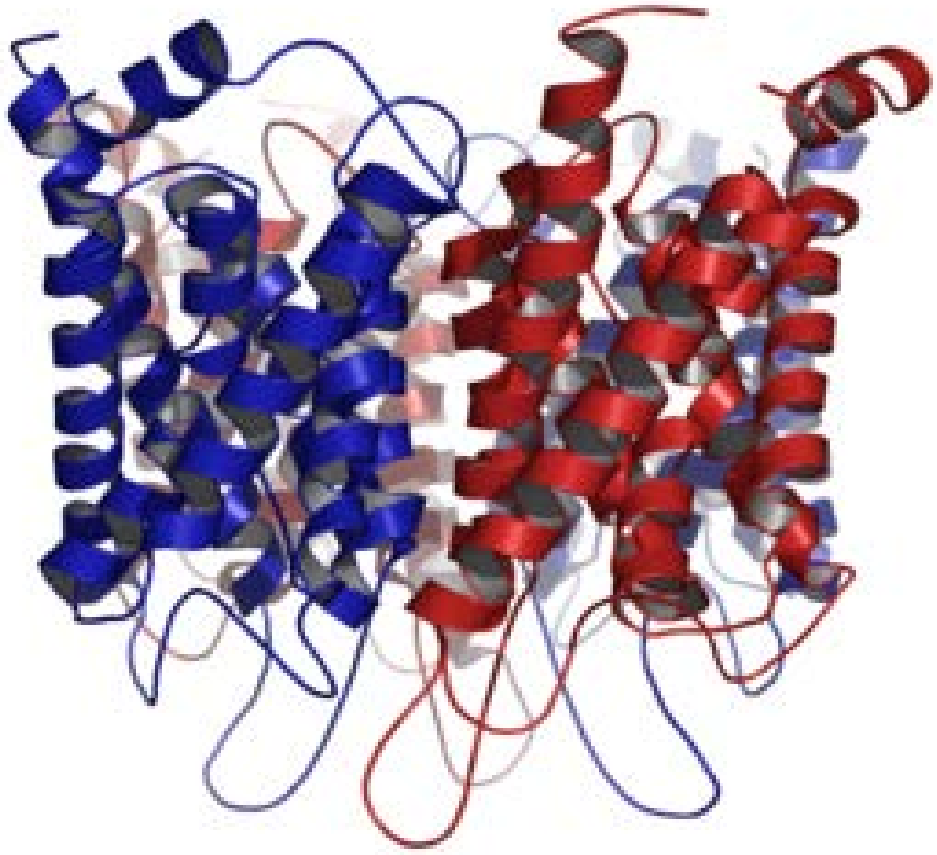
Examples of Protein Structure

Structure Confers Function

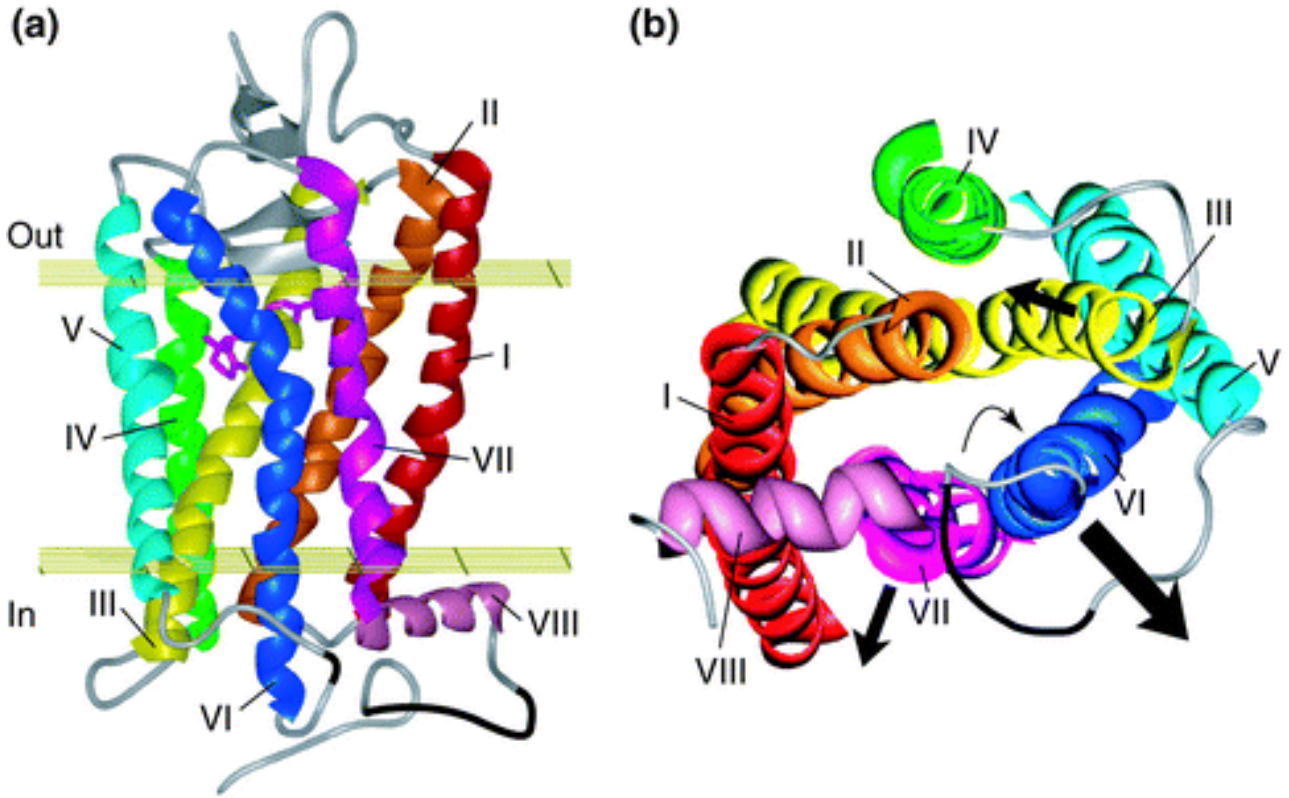
Membrane Proteins
Porins



Membrane Proteins
Aquaporins

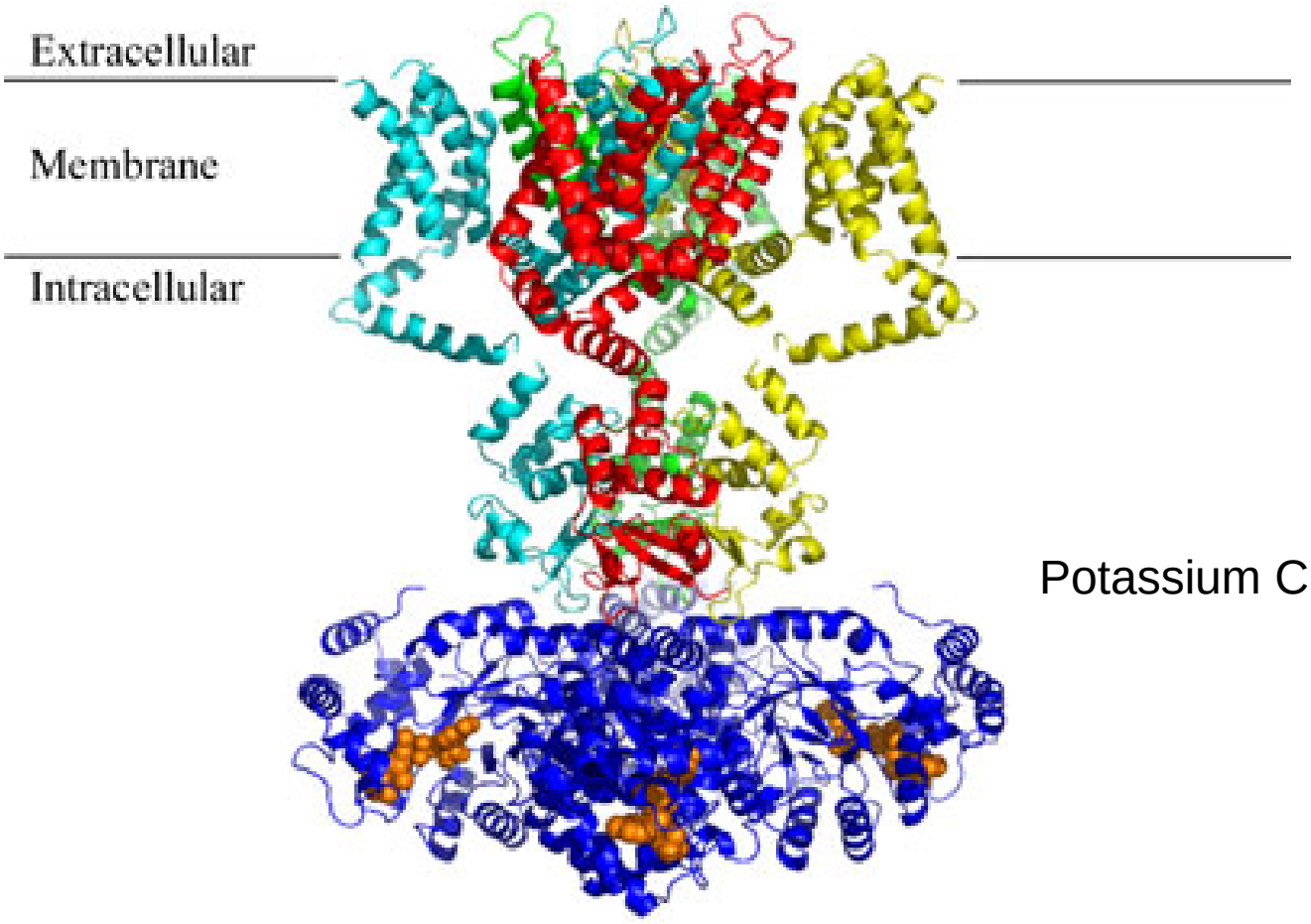


Membrane Proteins
G-Protein Coupled Receptors



GPCR: Rhodopsin

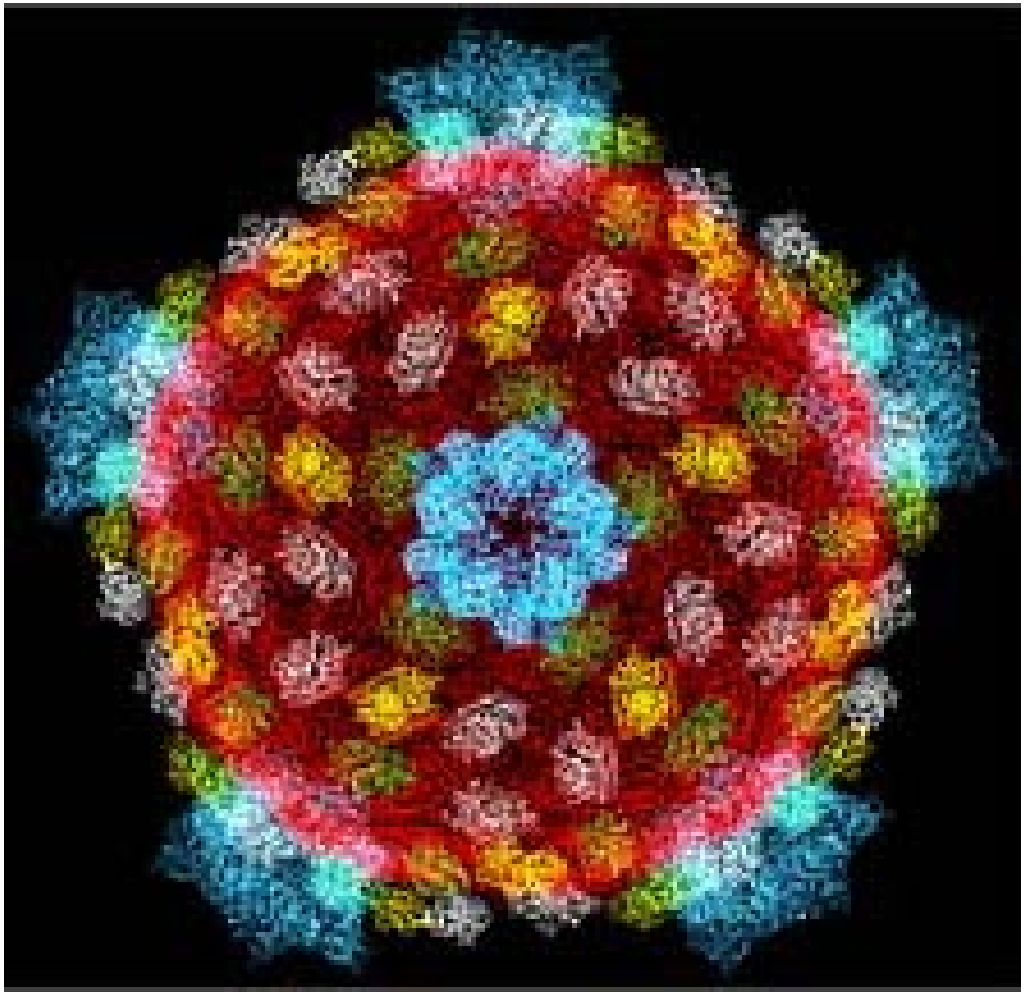
Membrane Proteins
Ion Channels



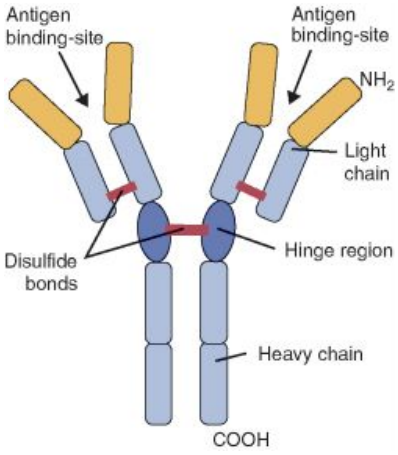
Potassium Channel

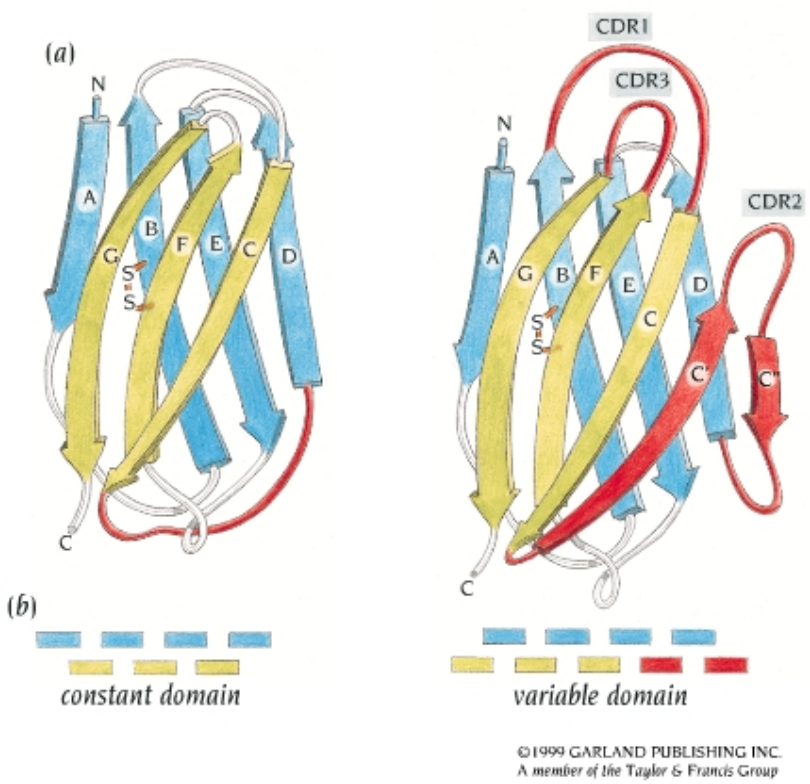
Viral Proteins

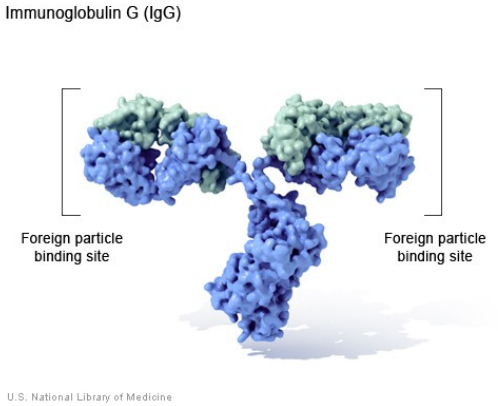
Reovirus Core



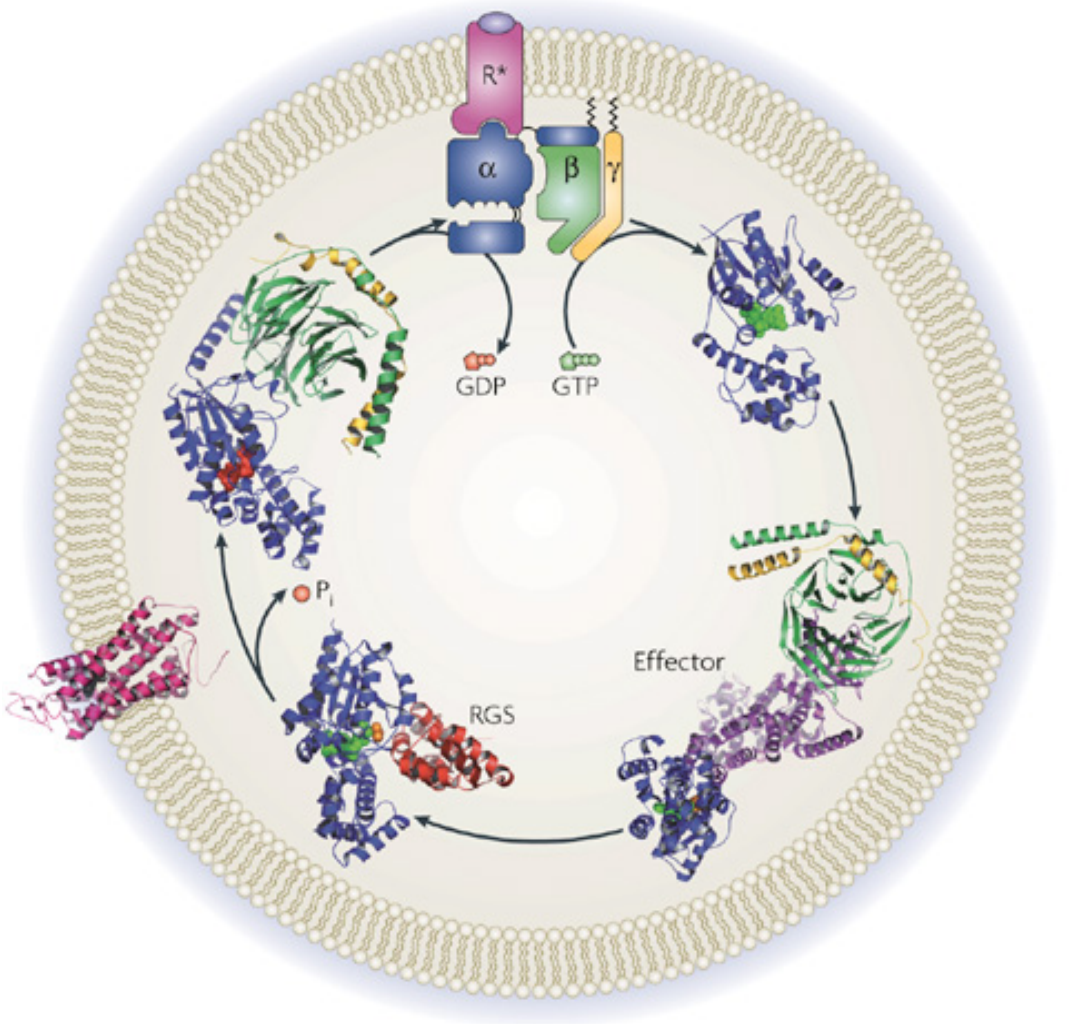
Immunoglobulins





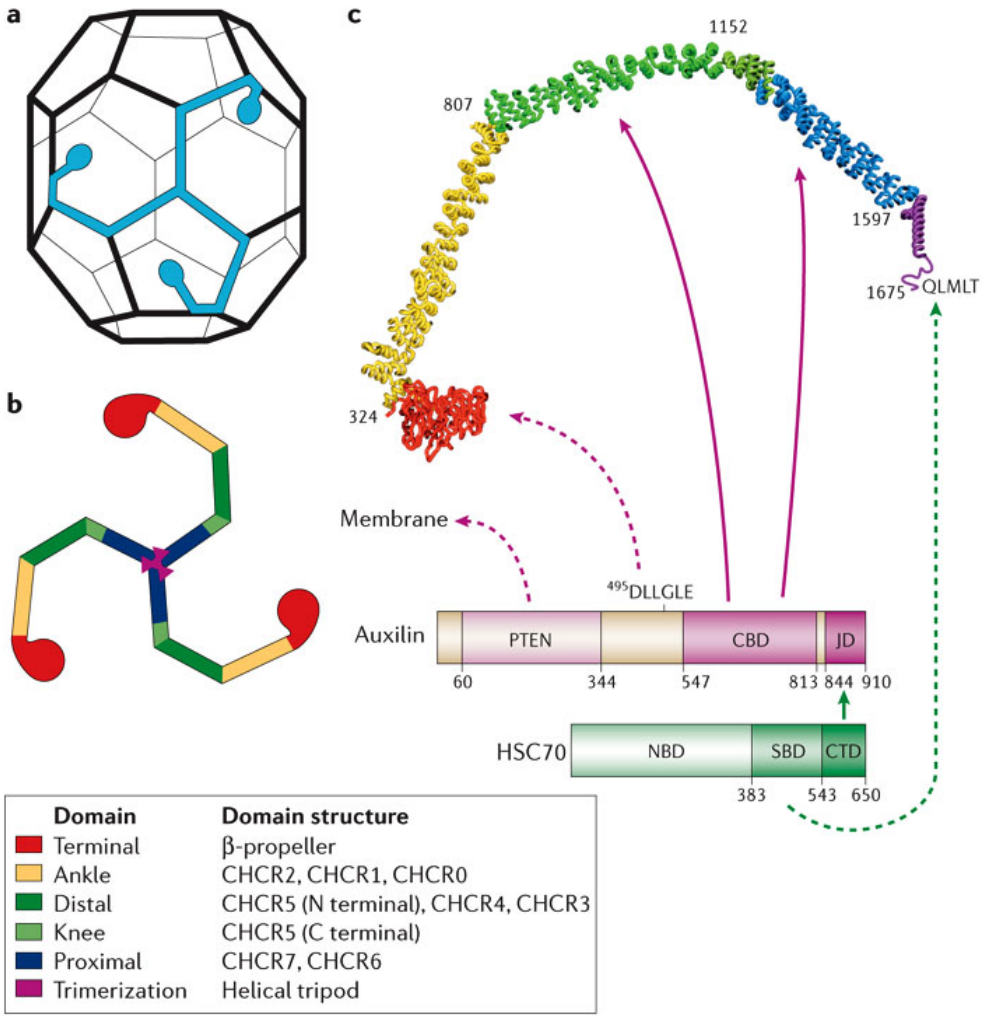


Signaling Molecules: G-protiens

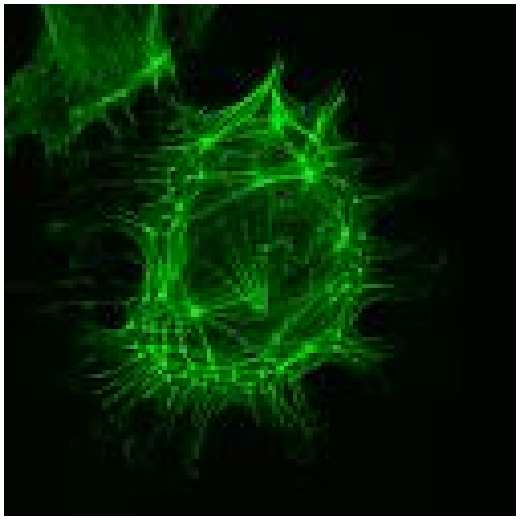


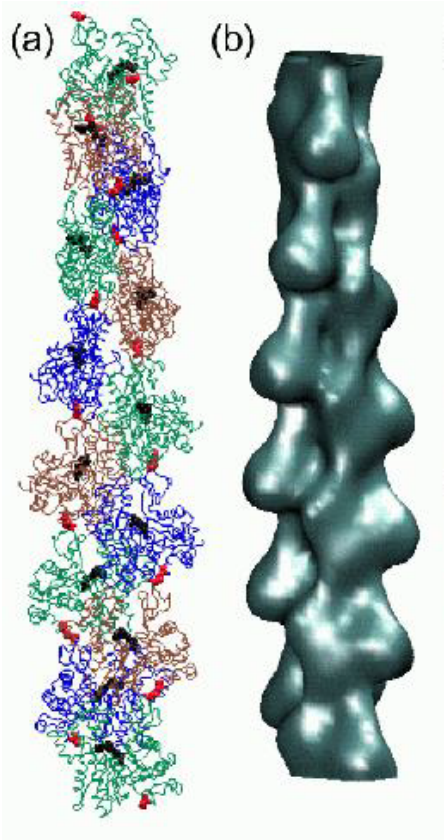
Nature Reviews | Molecular Cell Biology

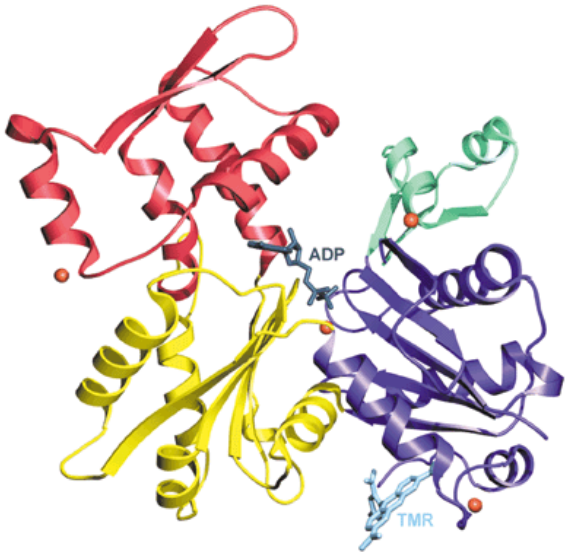
Structural Proteins
Clathrin



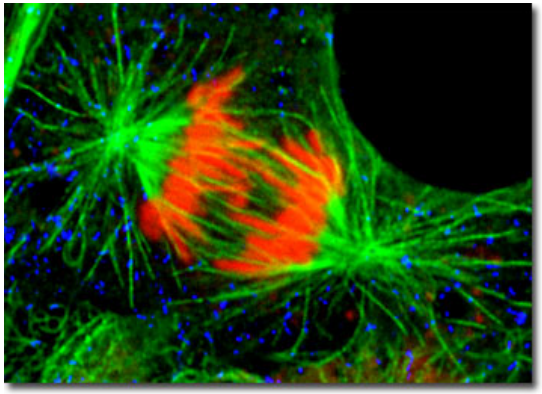
Structural Proteins
Actin Filaments: Actinh

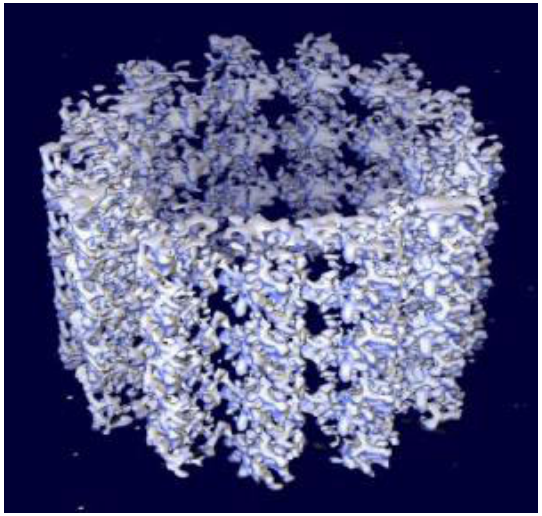


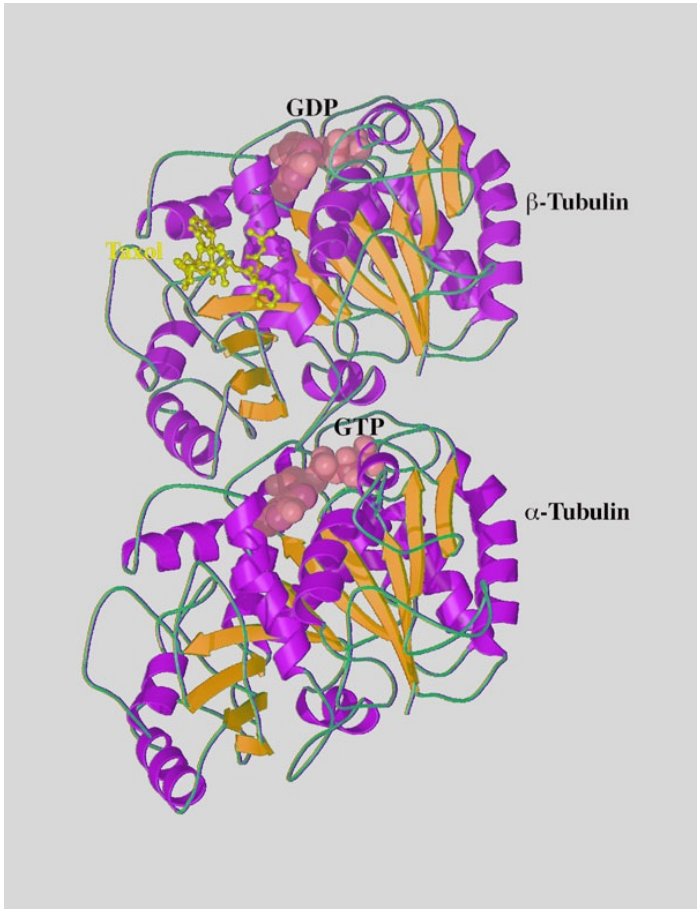




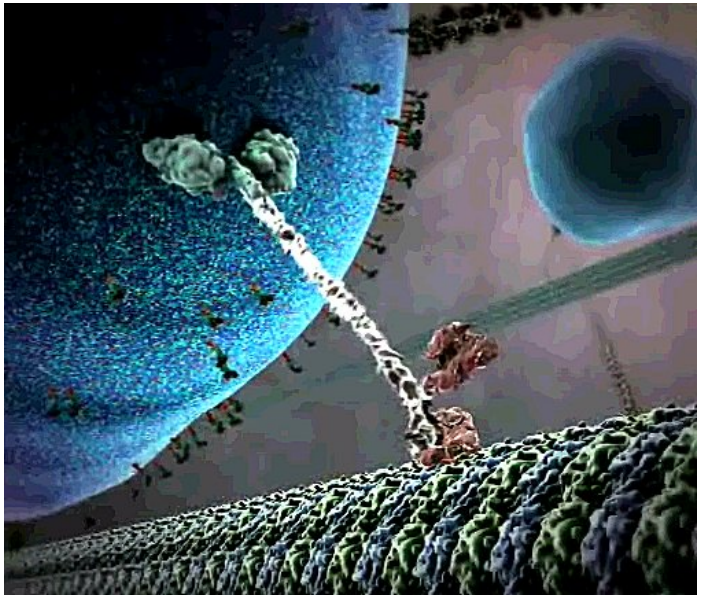
Structural Proteins
Microtubules: Tubulin

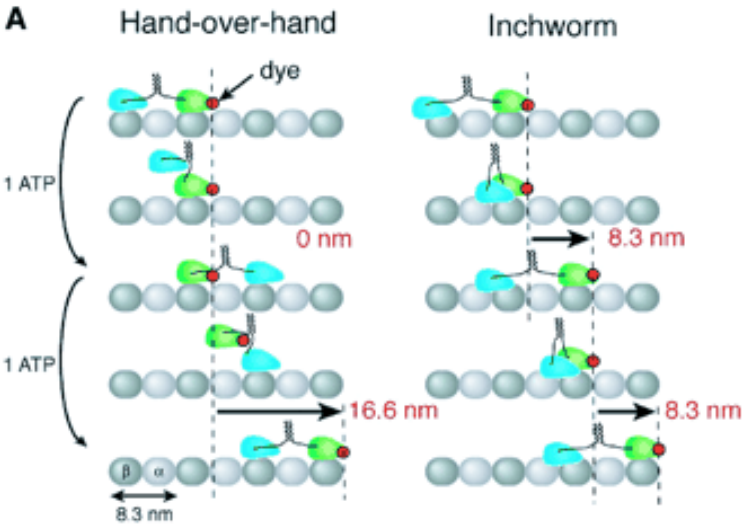


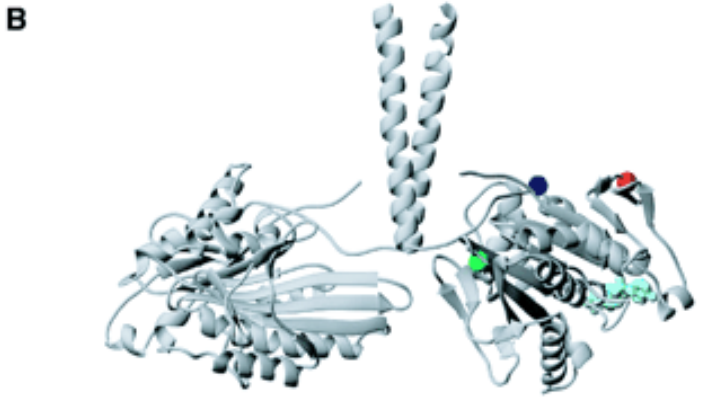




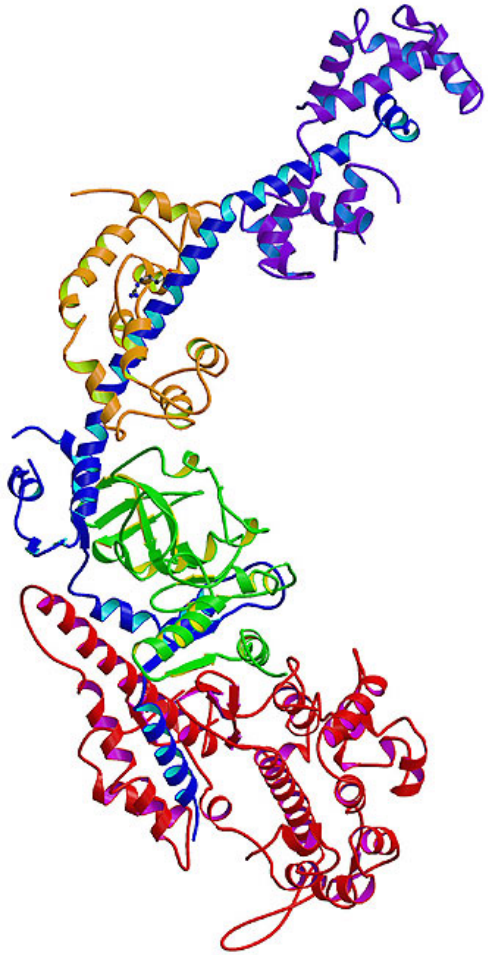
Motor Proteins
Kinesin







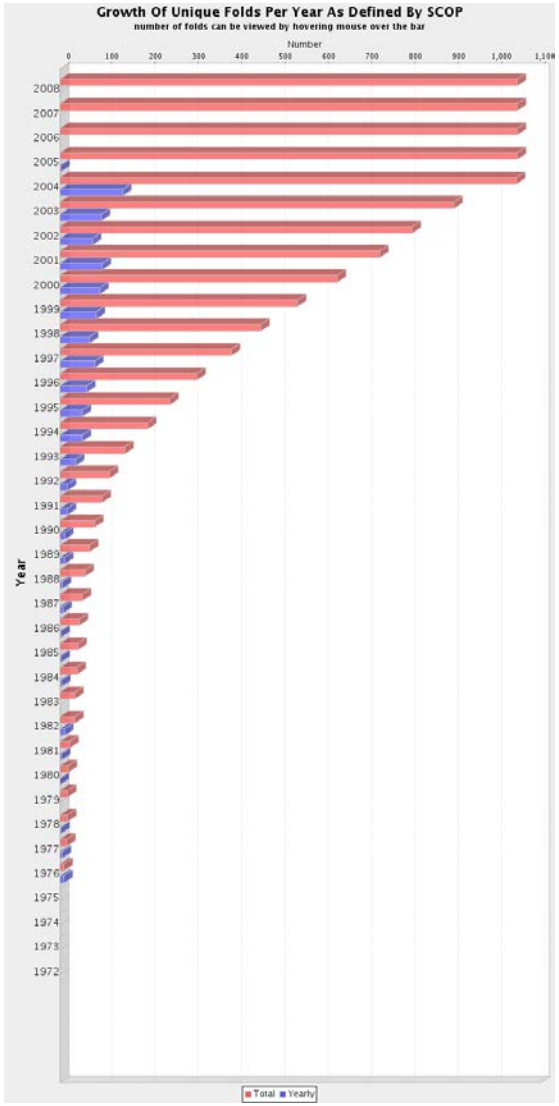
Motor Proteins
Myosin



Myosin Subfragment-1
(Gallus gallus)

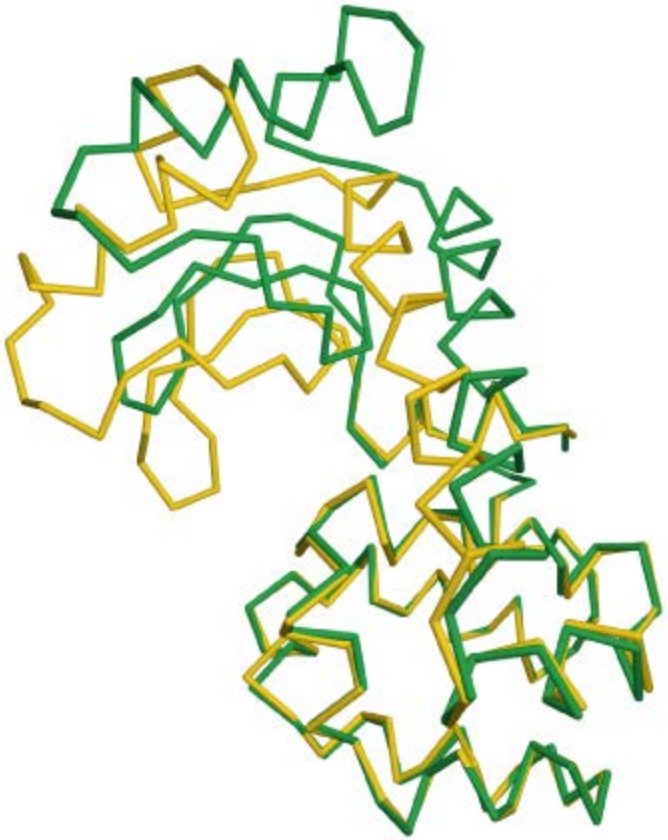
Protein Folds – as of 2008

2008



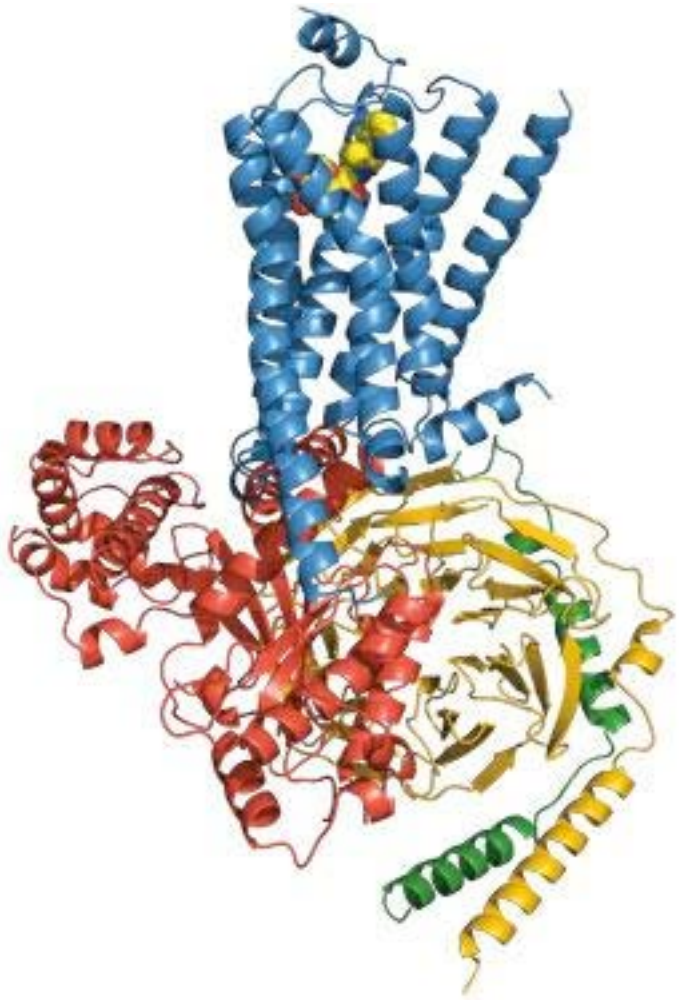
1056

Various Folds Confer Dynamic Properties



T4 Lysozyme

Various Folds Confer Dynamic Properties



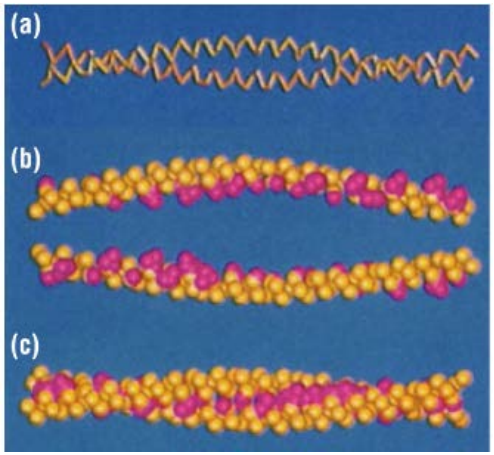
G-proteins
Nucleotide-dependent
Conformational change

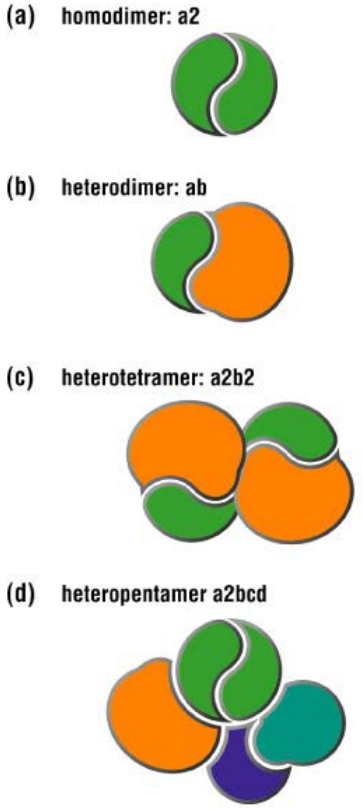
Quaternary Structure

Oligomerization of the same protein, or multiple different proteins

The Coiled Coil

A common dimerization motif





Homodimer

Homotrimer

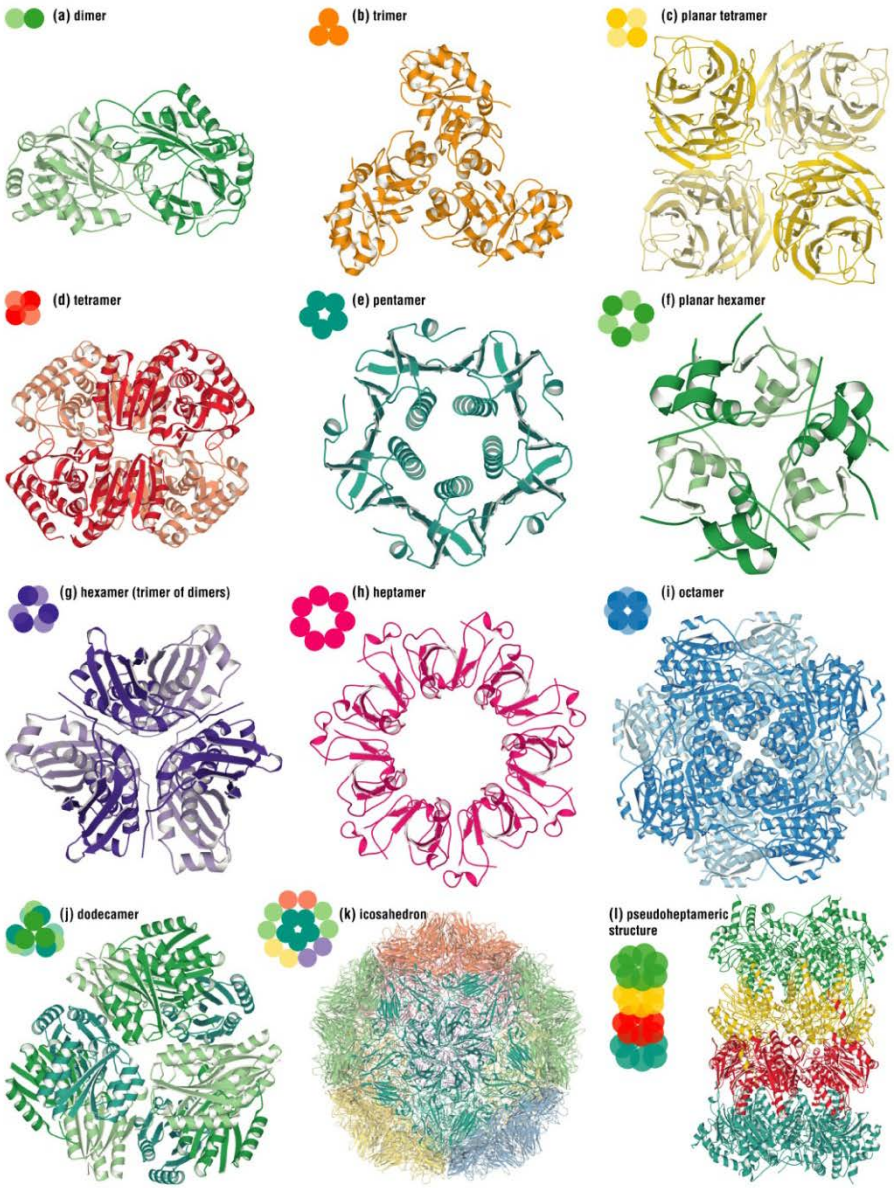
Homotetramer

Heterodimer

Heterotrimer

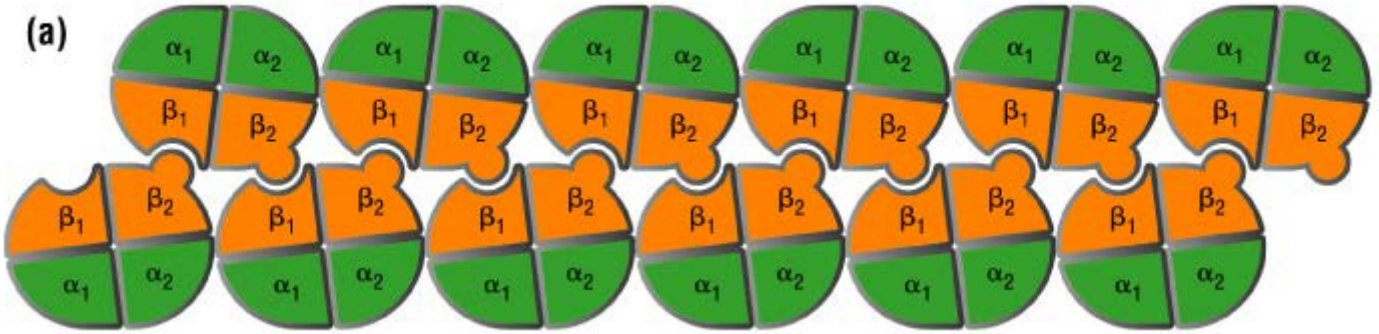
Heterotetramer

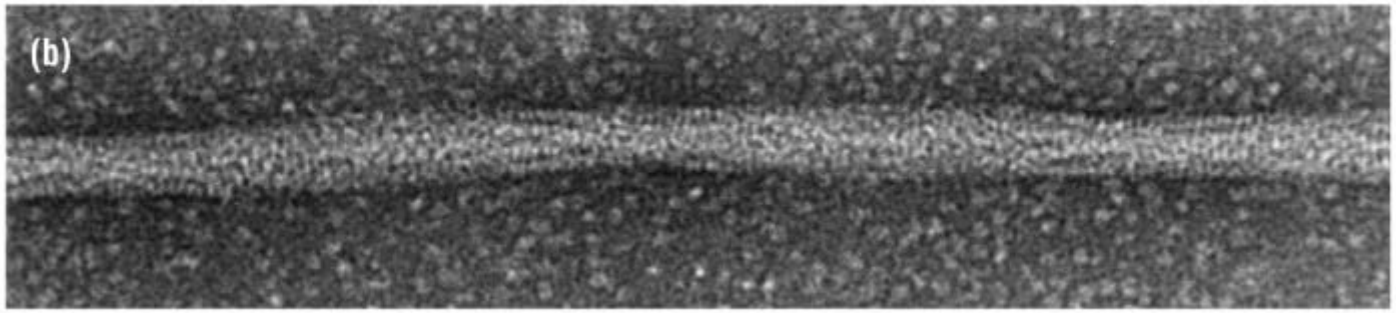
Quaternary Structure

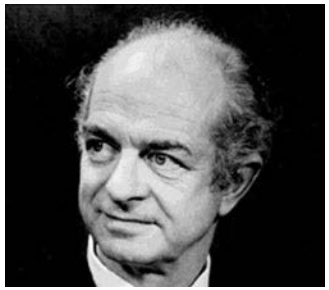


Quaternary Structure

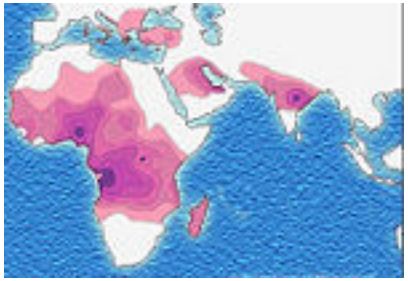
β-globin E6V Mutation: Sickle Cell Anemia







Pauling



Sickle Cell

Malaria

Hemoglobin is responsible Heterozygotes are protected from malaria

PyMol Demonstration

Discussion Questions

Open discussion on the PNAS paper by Pauling, Corey and Branson

What is the importance of tertiary structure for an active site?

Why is it important to study secondary and tertiary structure?

How important is it to know structure and what does structure tell us about function?

Can you predict the tertiary structure from primary or secondary structure.

Can you predict the quaternary structure from primary, secondary or tertiary Structure?

Why are some protein folds seen over and over again in nature – and used for different functions.