Protein structure
(and biomolecular structure more generally)

CS/CME/BioE/Biophys/BMI 279

Sept. 28 and Oct. 3, 2017

Ron Dror

- Please interrupt if you have questions, and especially if you’re confused!

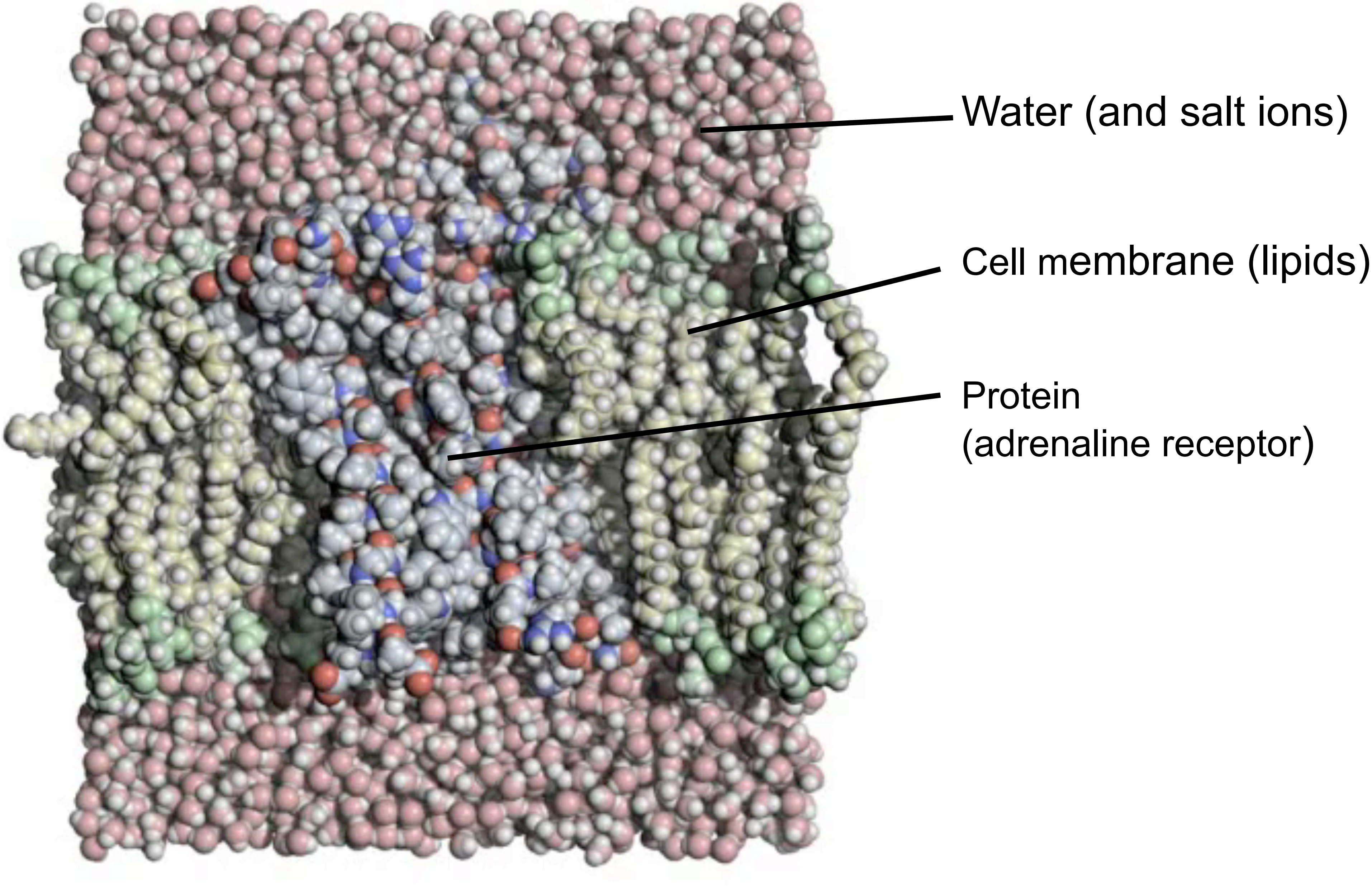
- Assignment 1 will be posted shortly
  - Look through it, especially the final problem, to decide whether to attend the tutorial next Wednesday (or, alternatively, ask the TAs for help during office hours)

Outline

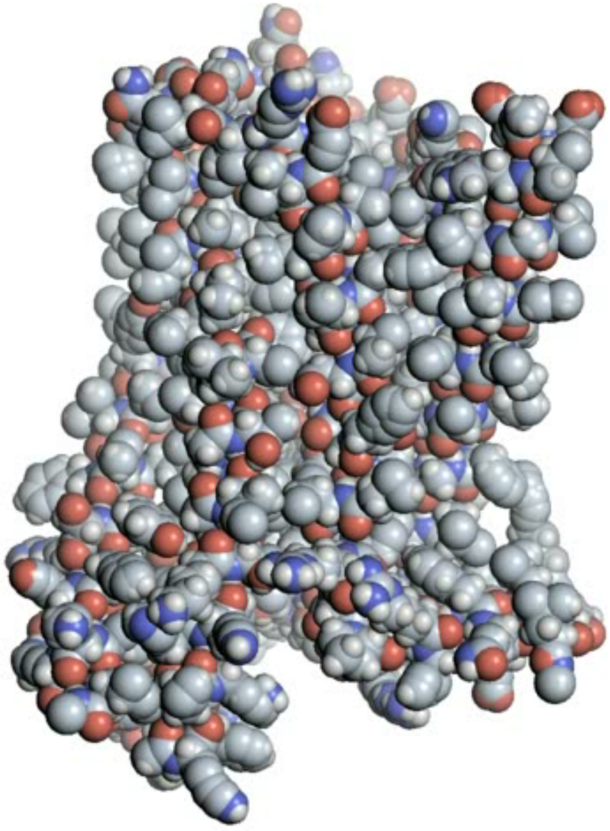
- Visualizing proteins
- The Protein Data Bank (PDB)
- Chemical (2D) structure of proteins
- What determines the 3D structure of a protein? Physics underlying biomolecular structure
  - Basic interactions
  - Complex interactions
- Protein structure: a more detailed view
  - Properties of amino acids
  - Secondary structure
  - Tertiary structure, quaternary structure, and domains
- Structures of other biomolecules

Visualizing proteins

Protein surrounded by other molecules (mostly water)

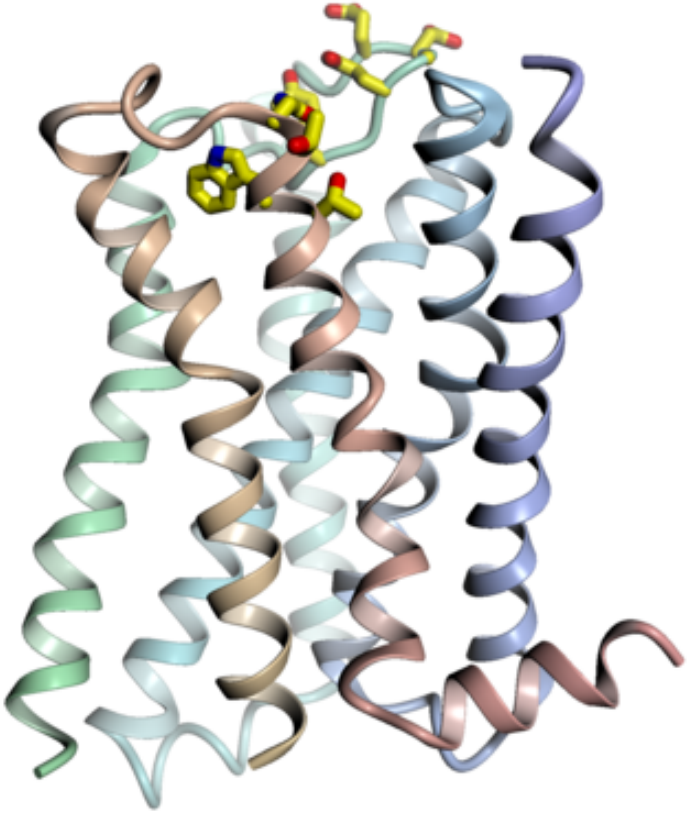


Protein only



Adrenaline receptor

Protein only — simplified representation



Adrenaline receptor

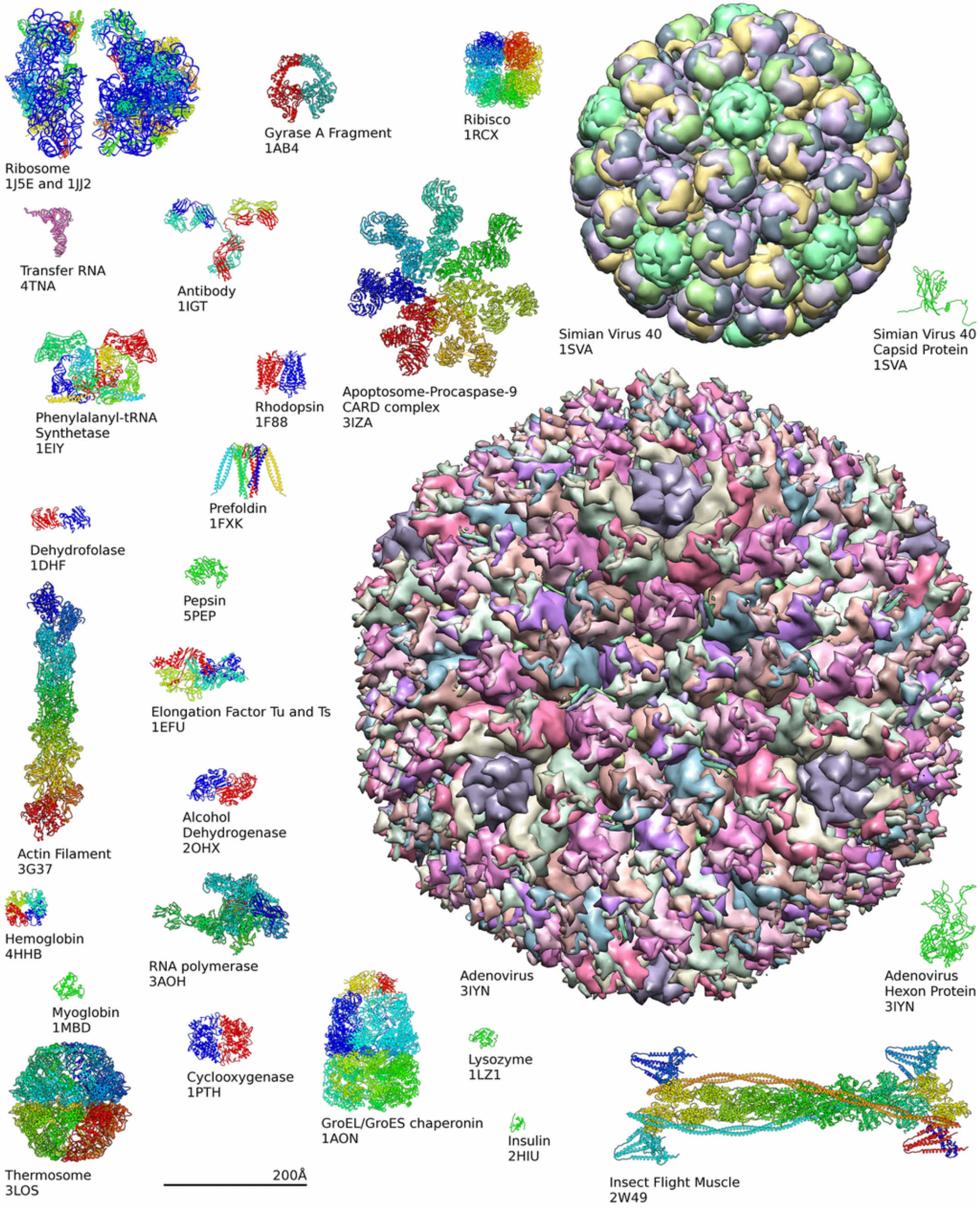
Key take-aways from these visualizations

- Protein is a long chain of amino acids.
- Protein and surrounding atoms fill space (close-packed).
- There are a lot of atoms. Simplified visual representations help you figure out what’s going on.

The Protein Data Bank (PDB)

The Protein Data Bank (PDB)

- Examples of structures from PDB.



https://upload.wikimedia.org/wikipedia/commons/thumb/2/24/Protein_structure_examples.png/1024px-Protein_structure_examples.png
(Axel Griewel)

You're not responsible for these; they're just examples.

The Protein Data Bank (PDB)

- http://www.rcsb.org/pdb/home/home.do
- A collection of (almost) all published experimental structures of biomacromolecules (e.g., proteins)
- Each identified by 4-character code (e.g., 1rcx)
- Currently ~134,000 structures. 90% of those are determined by x-ray crystallography.
- Browse it and look at some structures. Options:
  - 3D view in applet on PDB web pages
  - PyMol: fetch 1rcx
  - VMD: mol pdbload 1rcx

The Protein Data Bank (PDB)

An Information Portal to 111749 Biological Macromolecular Structures

A Structural View of Biology

This resource is powered by the Protein Data Bank archive-information about the 3D shapes of proteins, nucleic acids, and complex assemblies that helps students and researchers understand all aspects of biomedicine and agriculture, from protein synthesis to health and disease.

The RCSB PDB builds upon the data by creating tools and resources for research and education in molecular biology, structural biology, computational biology, and beyond.

Take an Interactive Tour of the PDB

September Molecule of the Month

Latest Entries *As of Tuesday Sep 01*

4Z7A

Structural and biochemical characterization of a non-functionally redundant M. tuberculosis (3,3) L,D-Transpep...

New Features

April 2015 Release

3D Structure Visualization
Support for modern web and mobile 3D viewers

Support For Large Structures
Access data files for large and complex entries

Protein Feature View Enhancements
Select views of proteins from related organisms with same gene name

Poster Prize Awarded at ACA

Congratulations to Marina E. Ivanova for *Molecular Interactions Within the Crumbs Cell Polarity Complex*, with an honorable mention to Kunhua Li for *High Resolution Crystal Structures of Antiviral, Glycosylated Y3 Protein from the Fungus*

Phased PDB Release Process » *08/24/15*

Announcing the 2015 EMDataBank Map Challenge » *08/11/15*

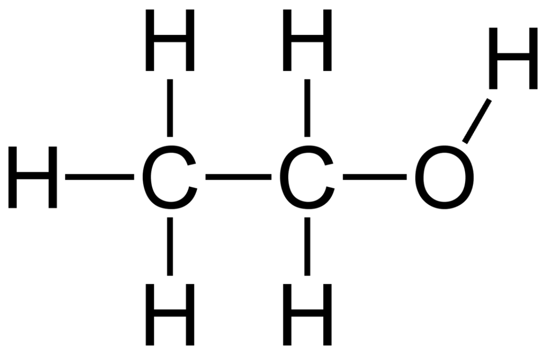
PDB at a Glance | 36224 Distinct Protein Sequences | 29324 Structures of Human Sequences | 7897 Nucleic Acid Containing Structures

Chemical (two-dimensional) structure of proteins

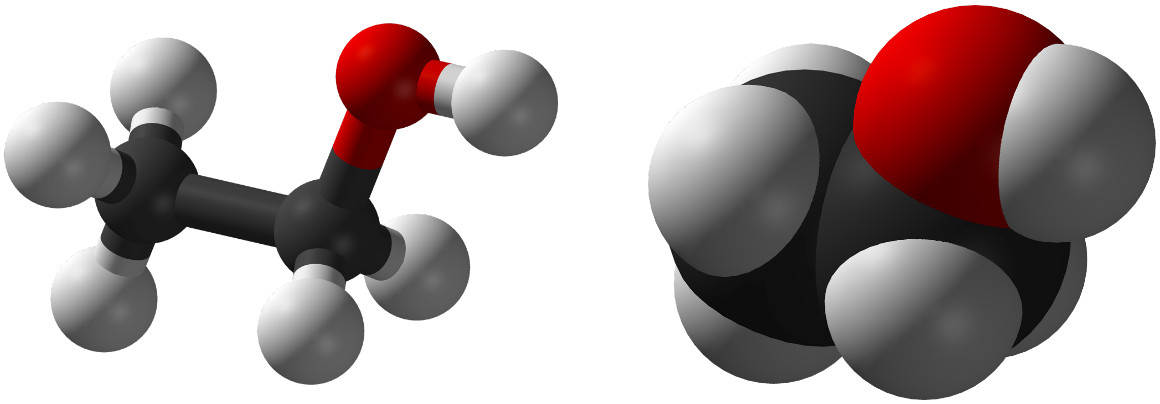
Two-dimensional (chemical) structure vs. three-dimensional structure

- Two-dimensional (chemical) structure shows *covalent bonds* between atoms. Essentially a graph.
- Three-dimensional structure shows relative positions of atoms.

2D structure



3D structure



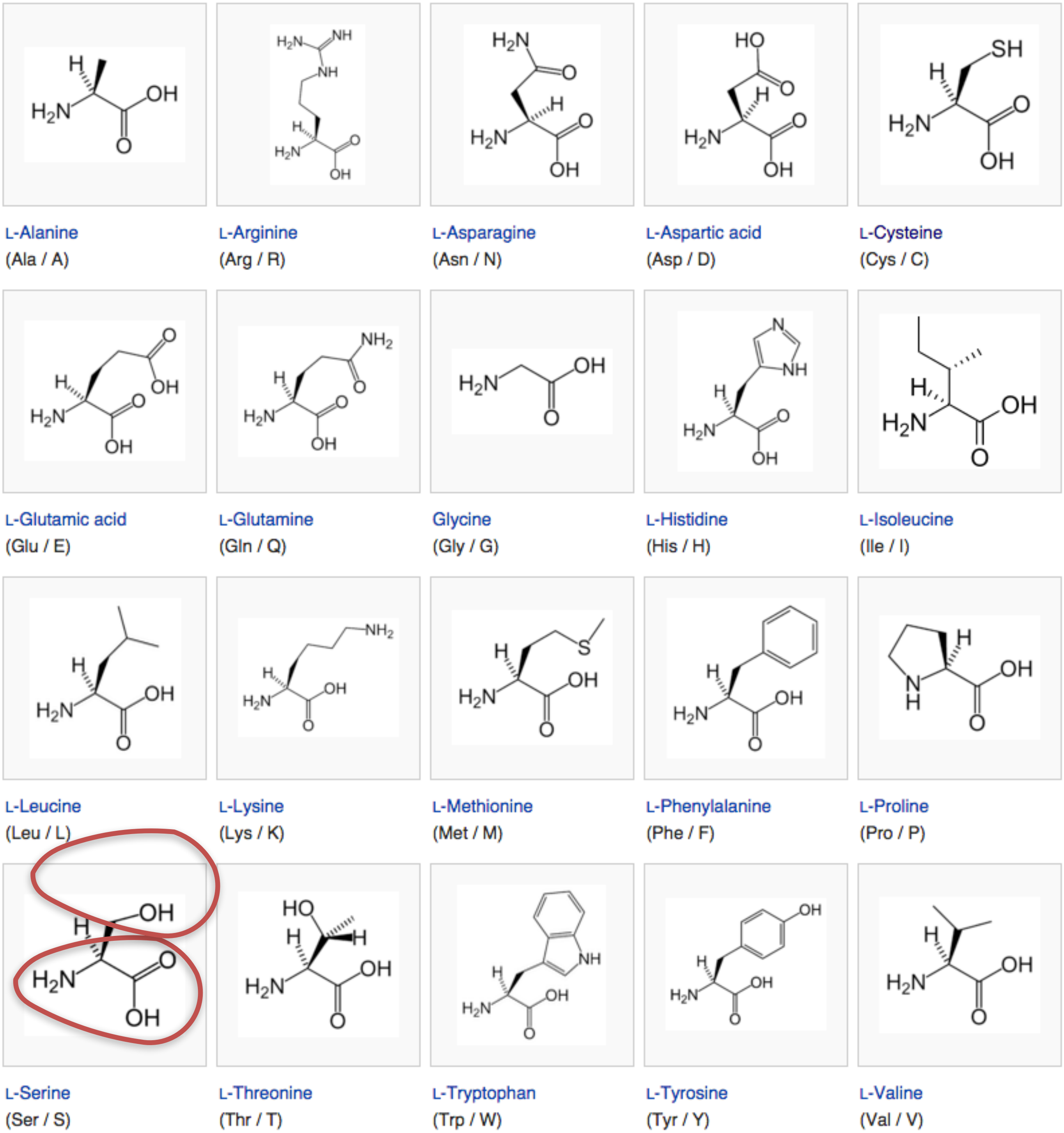
images from https://en.wikipedia.org/wiki/Ethanol

Proteins are built from amino acids

- 20 “standard” amino acids
- Each has three-letter and one-letter abbreviations (e.g., Threonine = Thr = T; Tryptophan = Trp = W)

The “side chain” is different in each amino acid.

All amino acids have this part in common.



L-Alanine
(Ala / A)

L-Arginine
(Arg / R)

L-Asparagine
(Asn / N)

L-Aspartic acid
(Asp / D)

L-Cysteine
(Cys / C)

L-Glutamic acid
(Glu / E)

L-Glutamine
(Gln / Q)

Glycine
(Gly / G)

L-Histidine
(His / H)

L-Isoleucine
(Ile / I)

L-Leucine
(Leu / L)

L-Lysine
(Lys / K)

L-Methionine
(Met / M)

L-Phenylalanine
(Phe / F)

L-Proline
(Pro / P)

L-Serine
(Ser / S)

L-Threonine
(Thr / T)

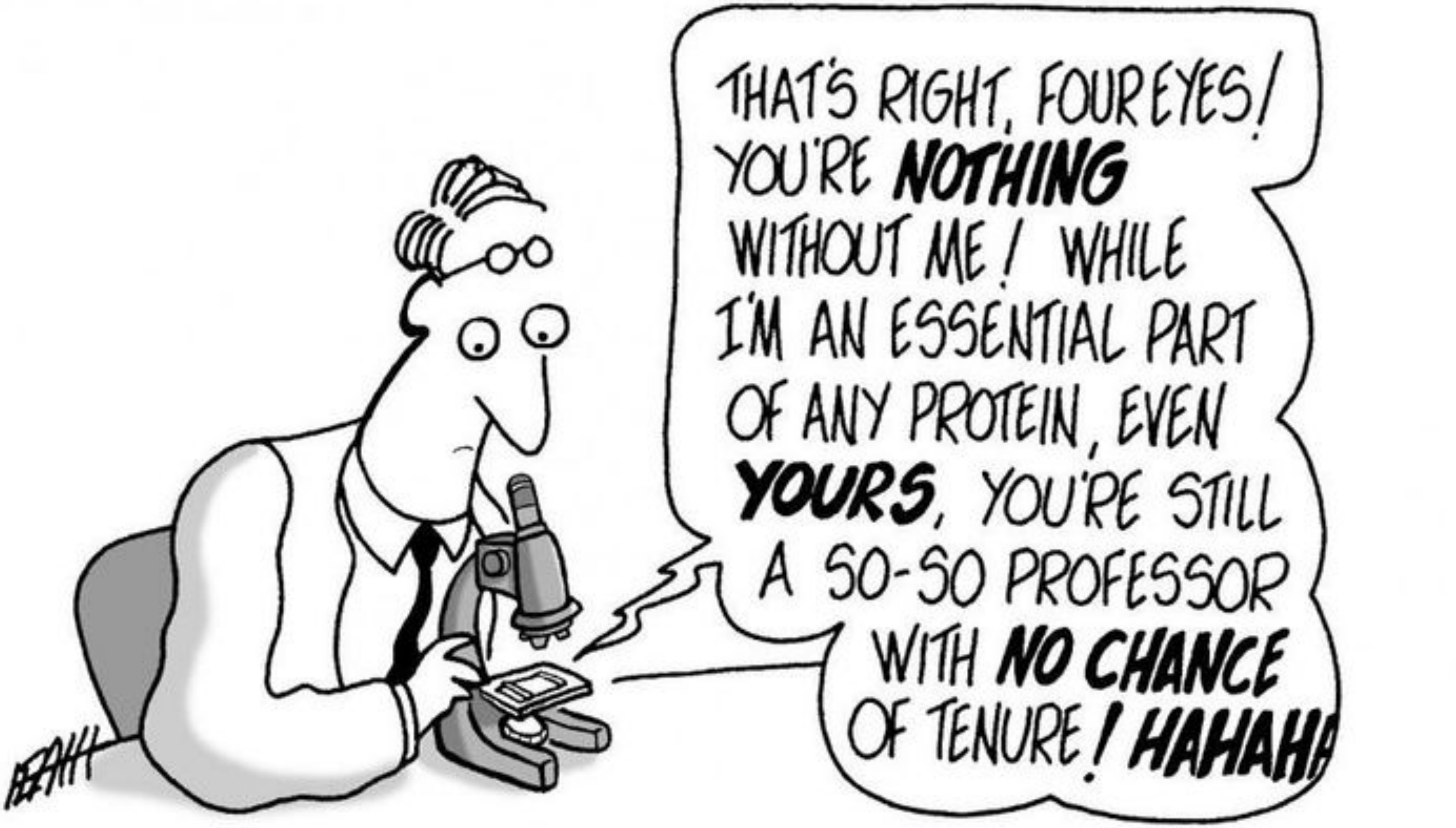
L-Tryptophan
(Trp / W)

L-Tyrosine
(Tyr / Y)

L-Valine
(Val / V)

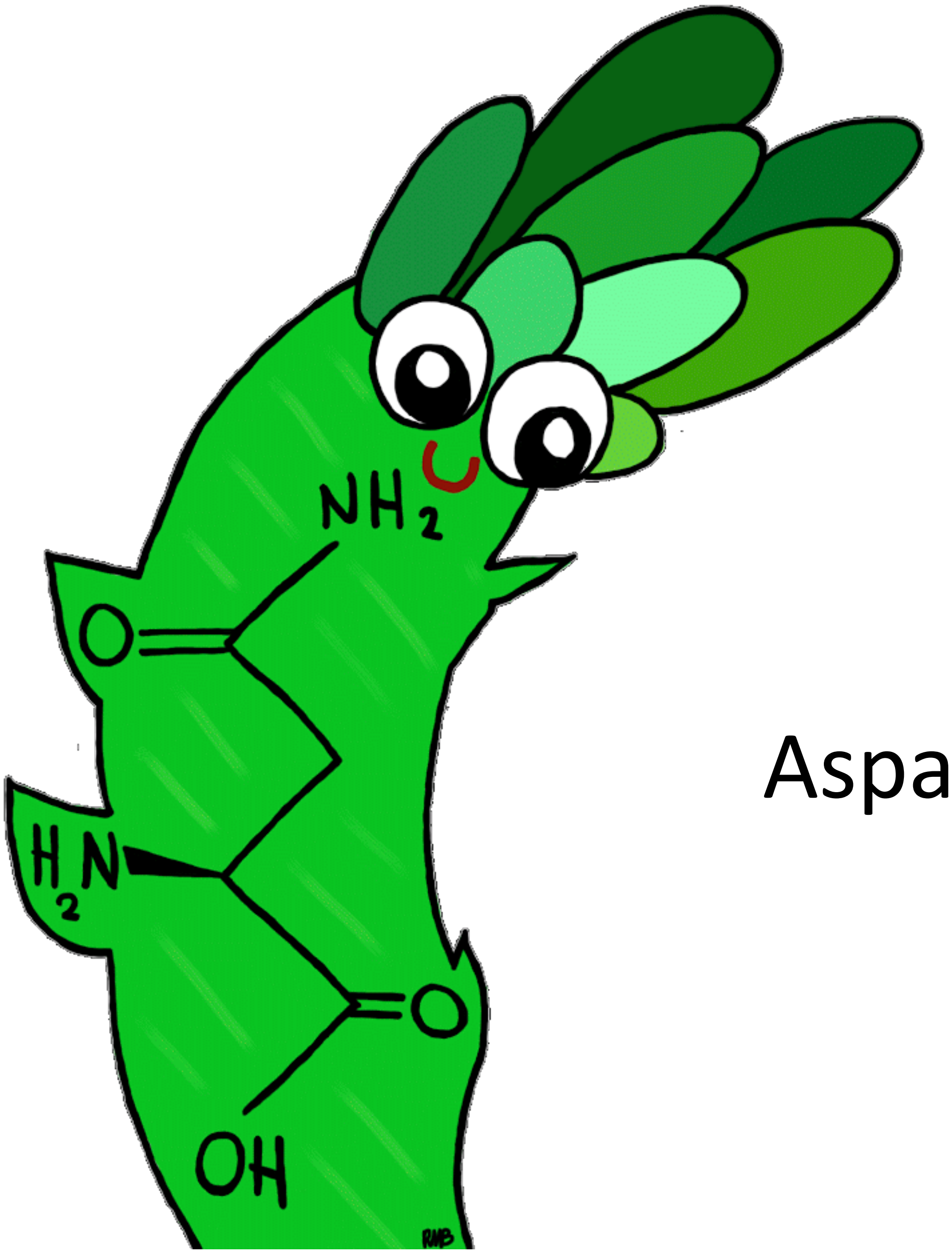
You don’t need to memorize all the structures

https://en.wikipedia.org/wiki/Proteinogenic_amino_acid



a mean o' acid

Source unknown. *American Scientist*?

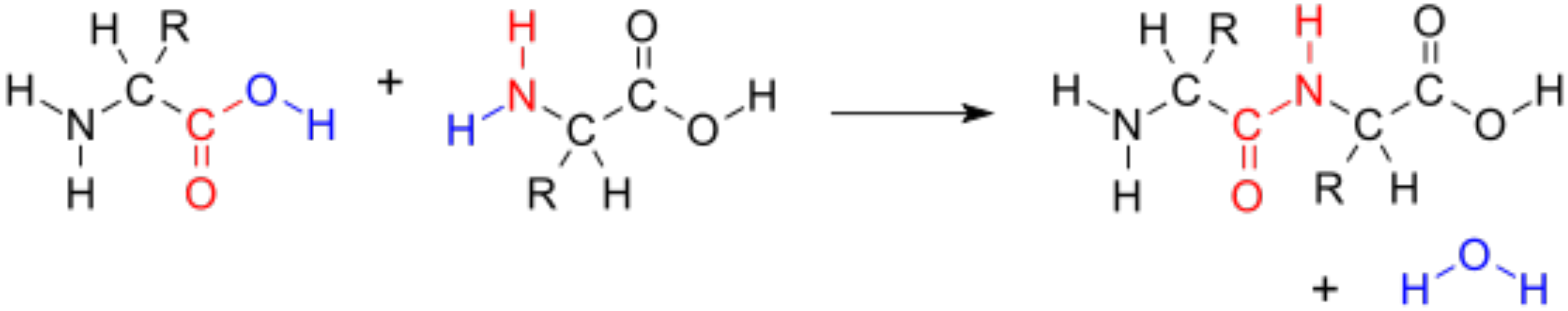


Asparagine

by Robin Betz

Proteins are chains of amino acids

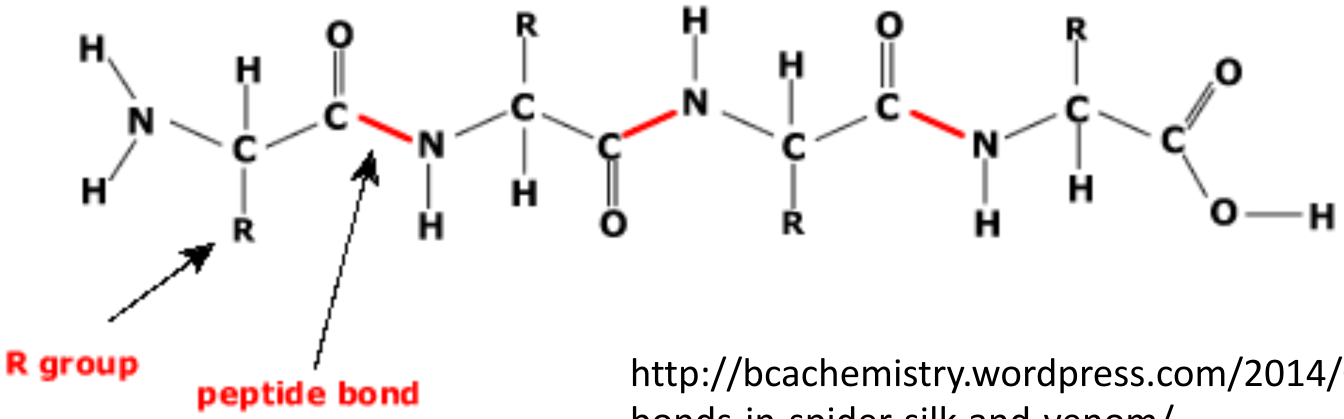
- Amino acids link together through a chemical reaction (“condensation”)



http://en.wikipedia.org/wiki/Condensation_reaction

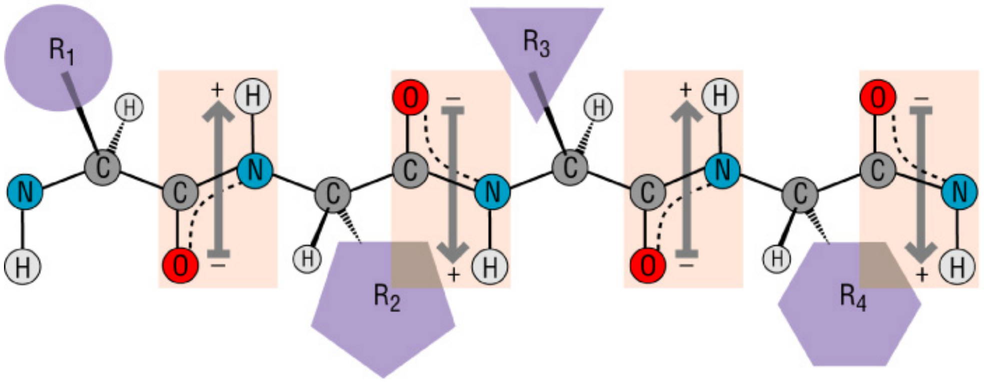
- Elements of the chain are called “amino acid residues” or just “**residues**” (important term!)
- The bonds linking these residues are “peptide bonds.” The chains are also called “polypeptides”

Proteins have uniform backbones with differing side chains



http://bcachemistry.wordpress.com/2014/05/28/chemical-bonds-in-spider-silk-and-venom/

From **Protein Structure and Function** by Gregory A Petsko and Dagmar Ringe



What determines the 3D structure of a protein?

Physics underlying biomolecular structure

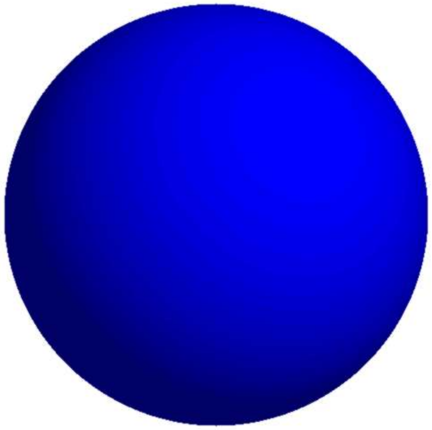
Why do proteins have well-defined structure?

- The sequence of amino acids in a protein (usually) suffices to determine its structure.
- A chain of amino acids (usually) “folds” spontaneously into the protein’s preferred structure, known as the “native structure”
- Why?
  - Intuitively: some amino acids prefer to be inside, some prefer to be outside, some pairs prefer to be near one another, etc.
  - To understand this better, examine forces acting between atoms

What determines the 3D structure of a protein?
Physics underlying biomolecular structure

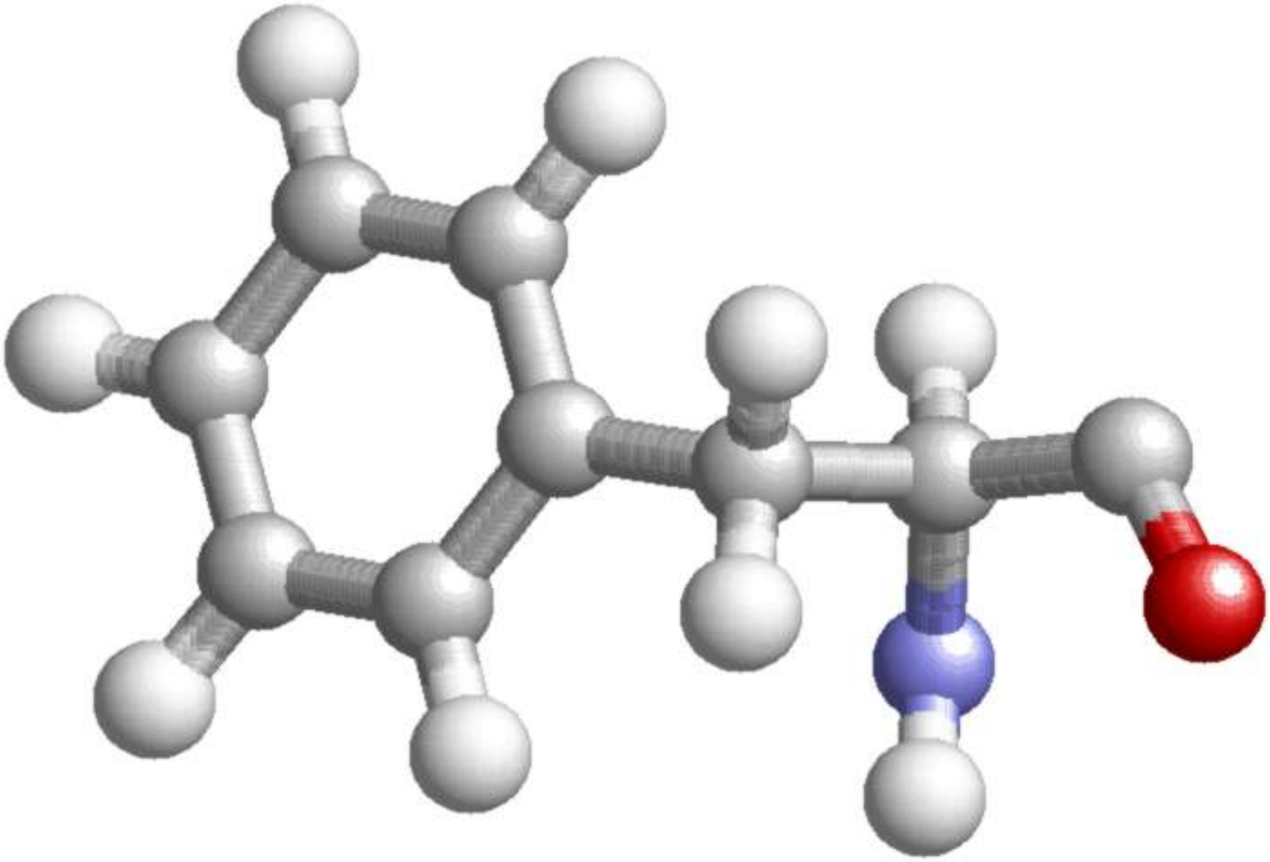
Basic interactions

Geometry of an atom



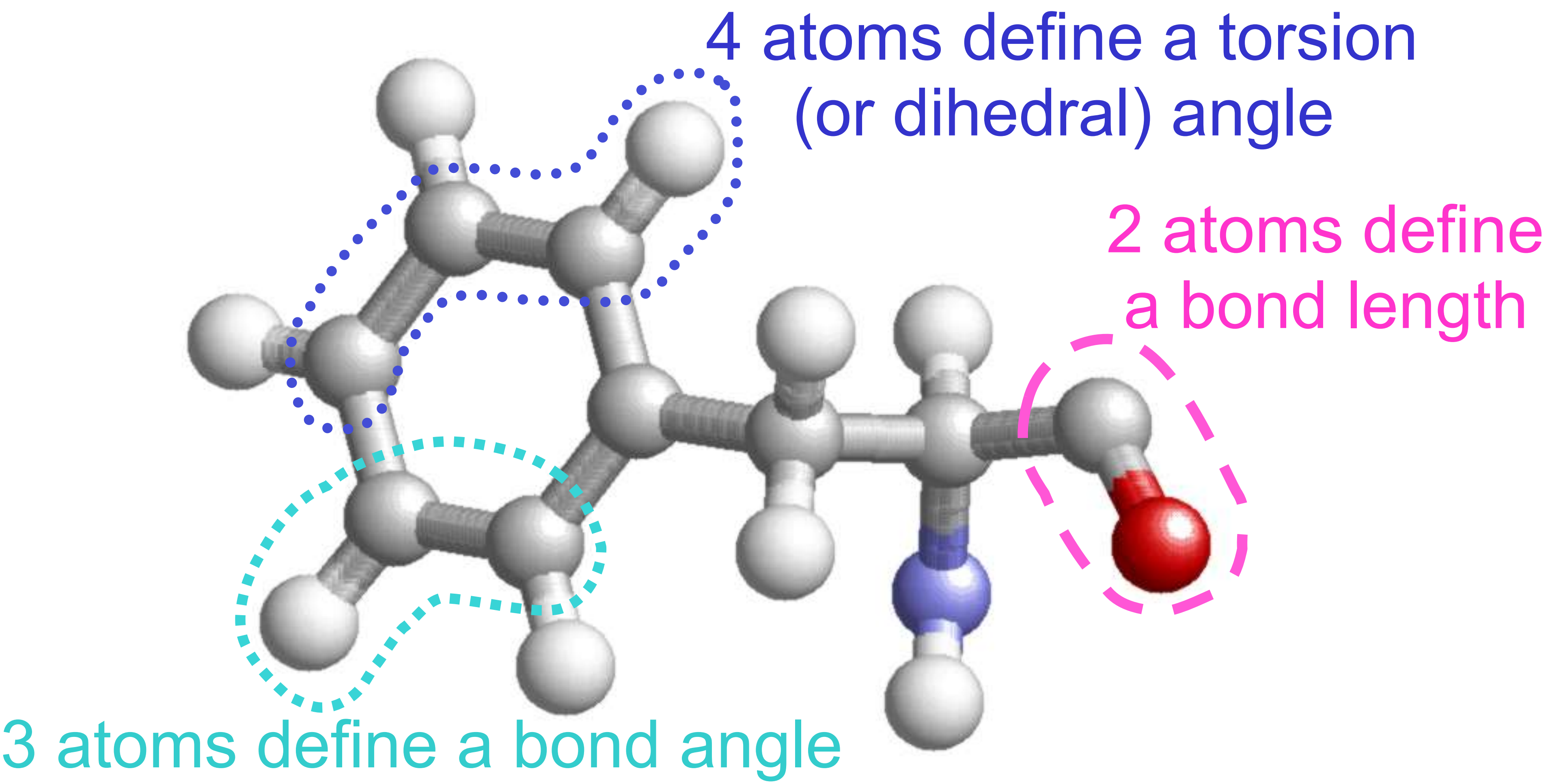
- To a first approximation (which suffices for the purposes of this course), we can think of an atom simply as a sphere.
- It occupies a position in space, specified by the (x, y, z) coordinates of its center, at a given point in time

Geometry of a molecule



- A molecule is a set of atoms connected in a graph
- (x, y, z) coordinates of each atom specify its geometry

Geometry of a molecule



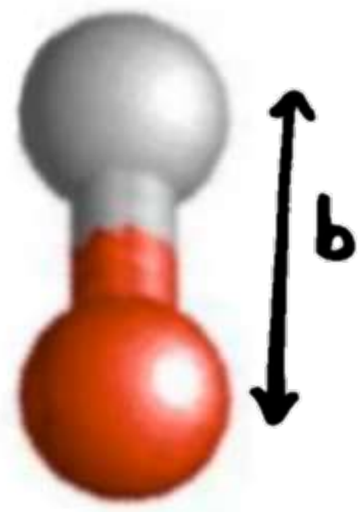
- Alternatively, we can specify the geometry of a molecule using bond lengths, bond angles, and torsion angles

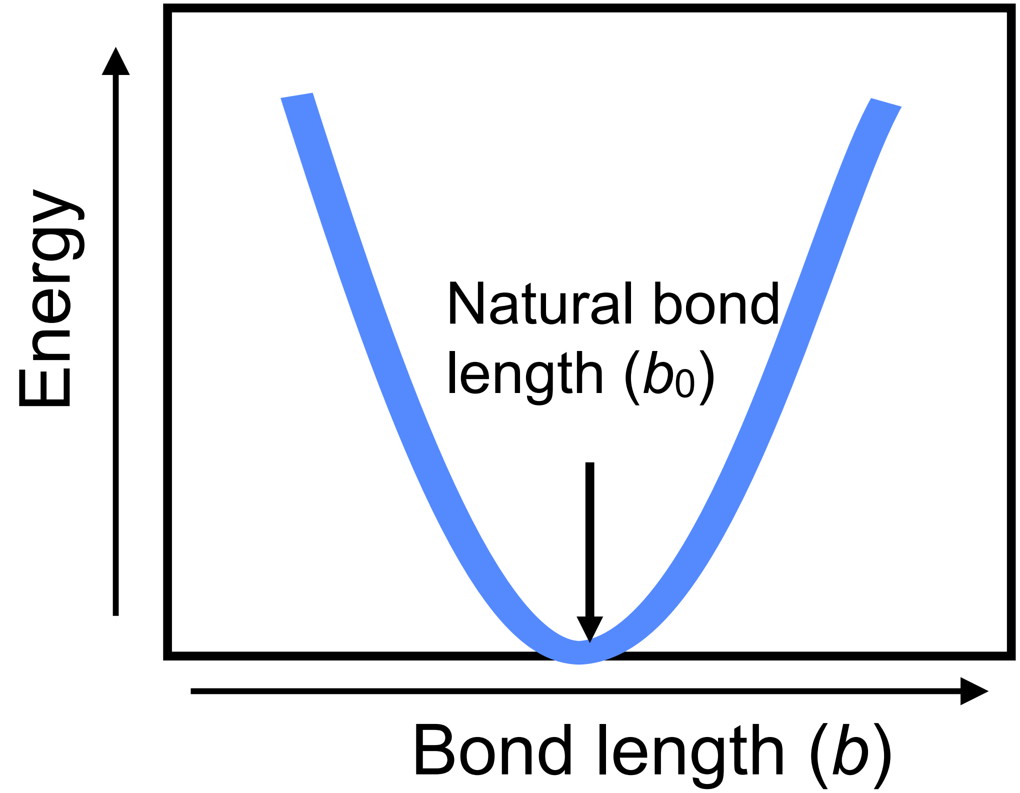
Forces between atoms

- We can approximate the total energy as a sum of individual contributions. Terms are additive.
  - Thus force on each atom is also a sum of individual contributions. Remember: force is the derivative of energy.
  - We will ignore quantum effects. Think of atoms as balls and forces as springs.
- Two types of forces:
  - Bonded forces: act between closely connected sets of atoms in bond graph
  - Non-bonded forces: act between all pairs of atoms

Bond length stretching

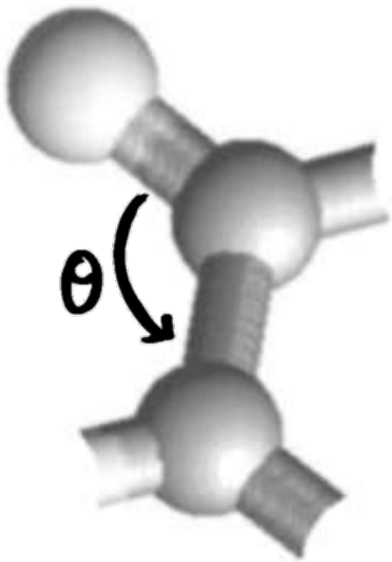
- A bonded pair of atoms is effectively connected by a spring with some preferred (natural) length. Stretching or compressing it requires energy.

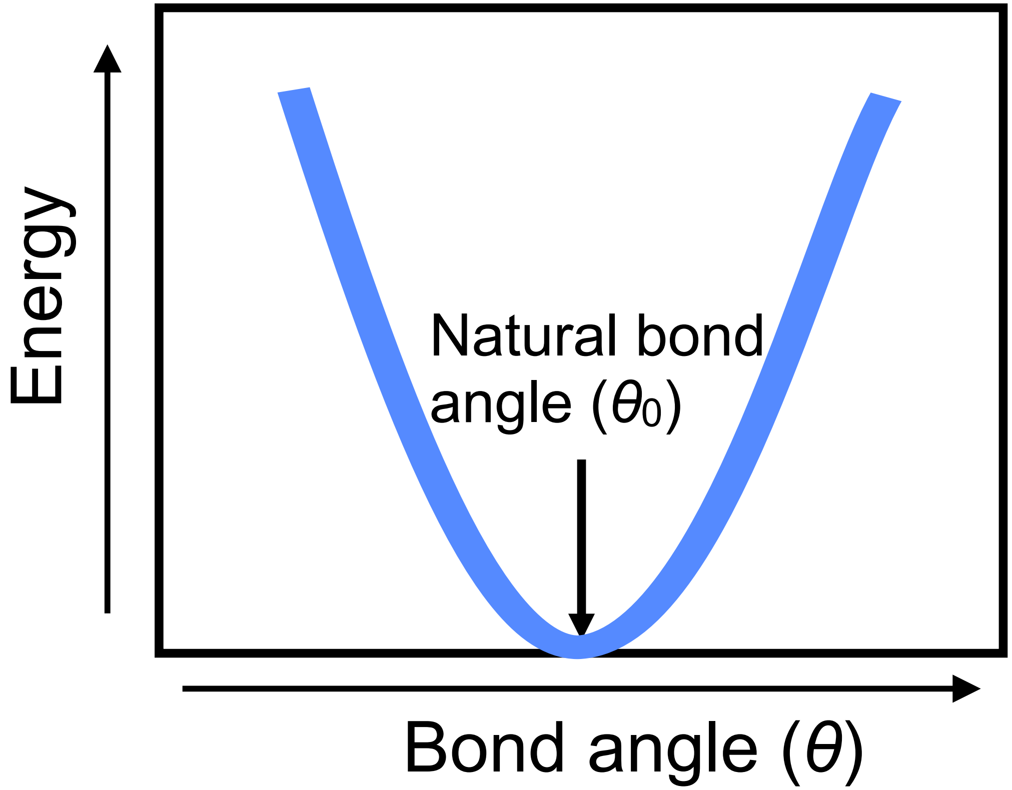




Bond angle bending

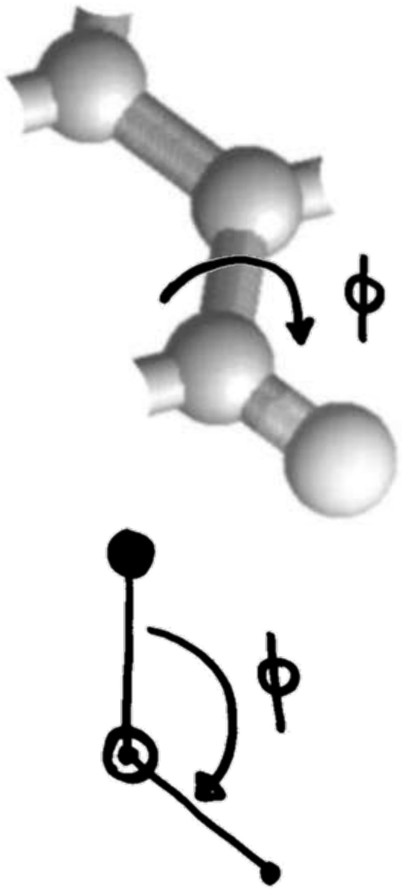
- Likewise, each bond angle has some natural value. Increasing or decreasing it requires energy.

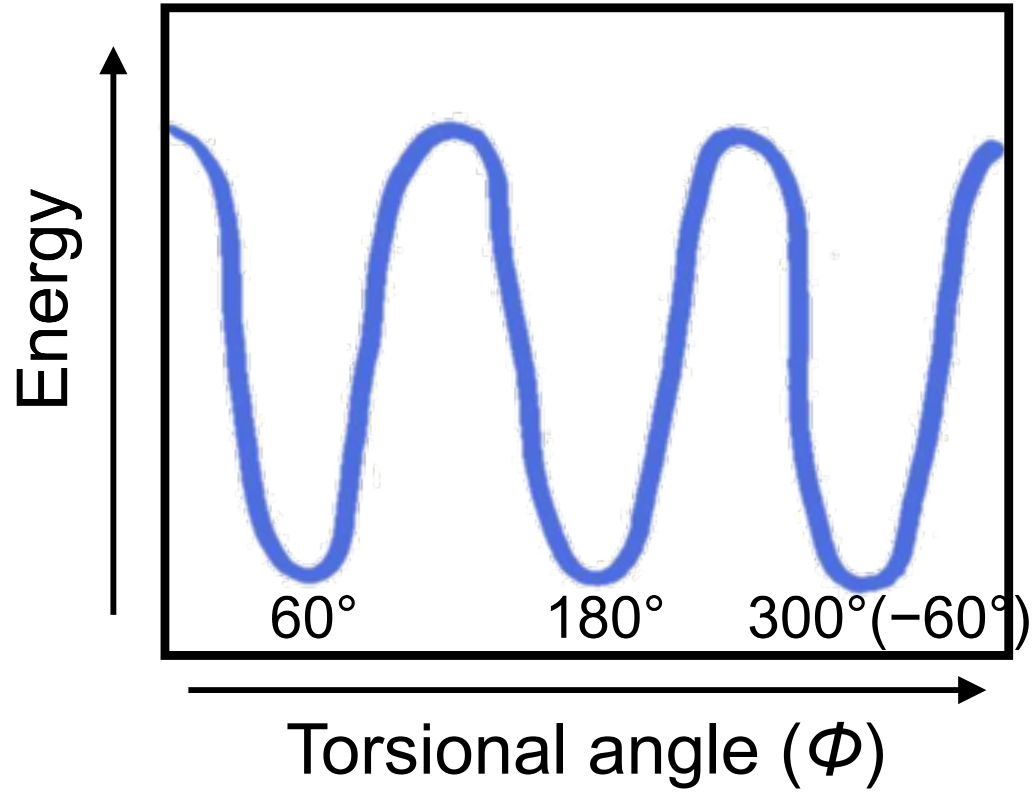




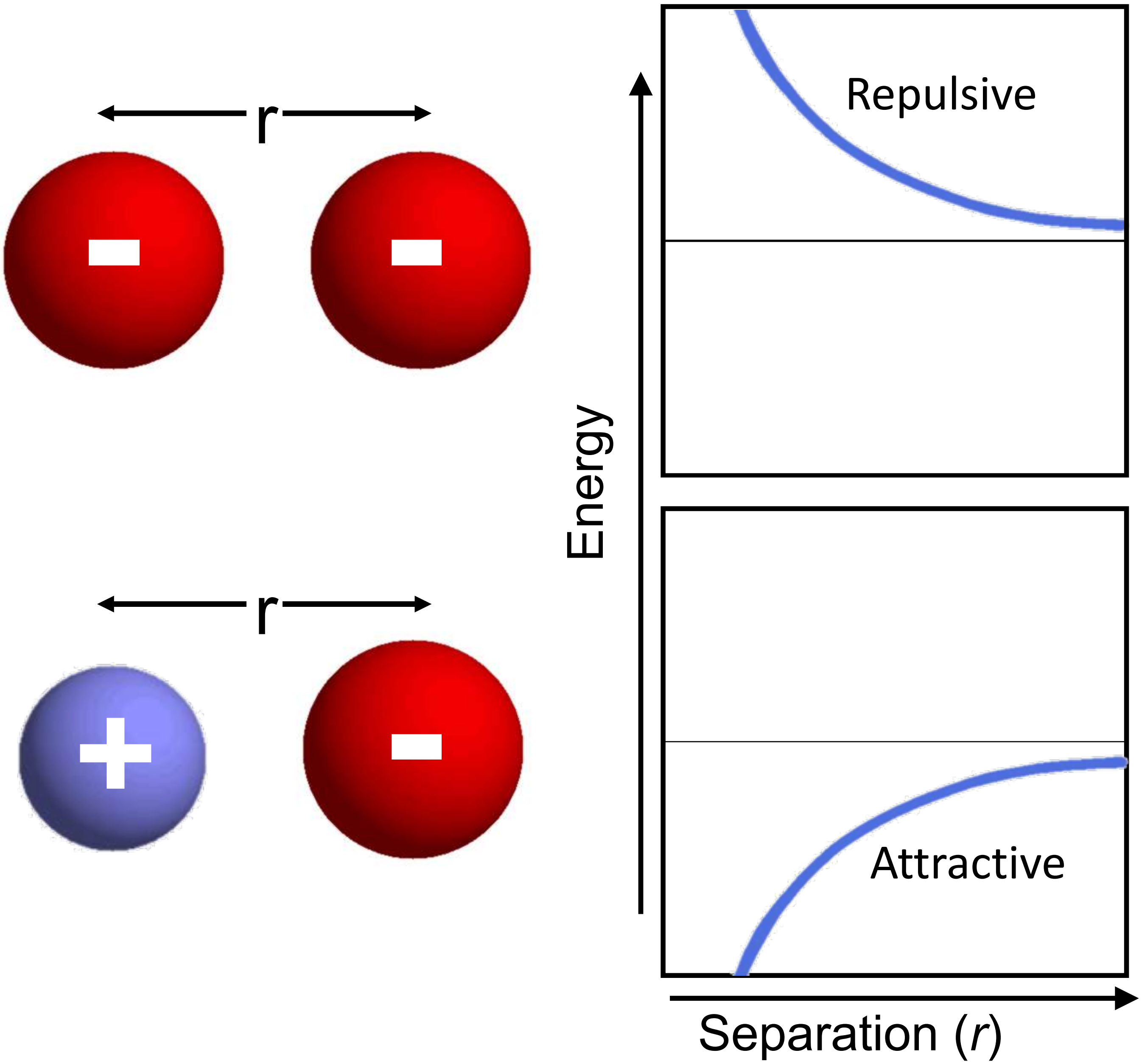
Torsional angle twisting

- Certain values of each torsional angle are preferred over others.



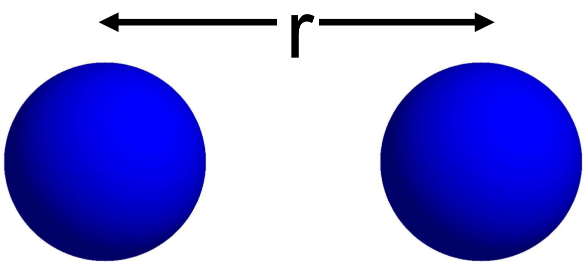


Electrostatic interaction

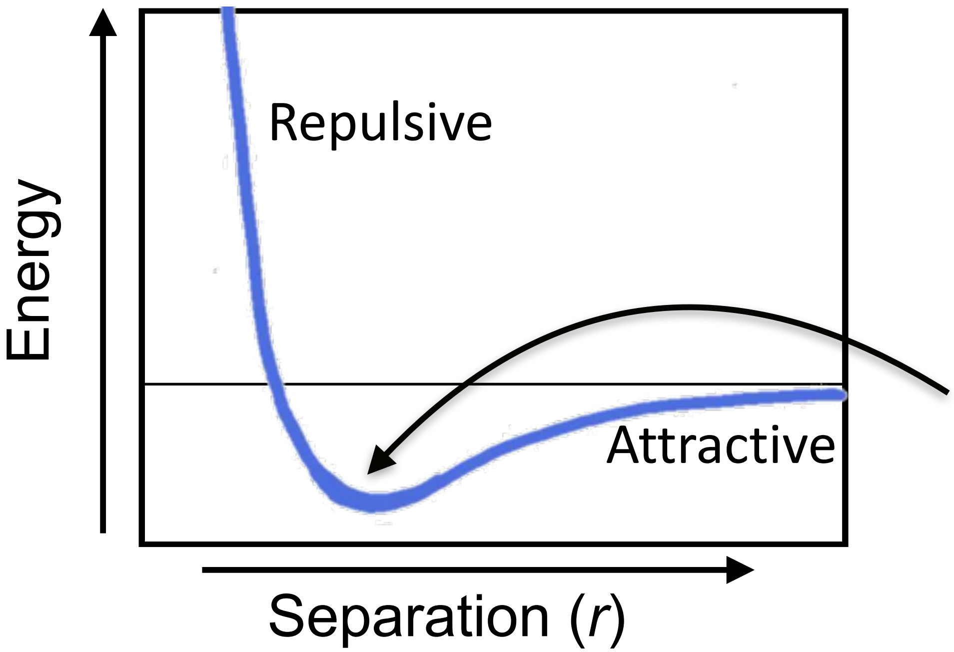


- Like charges repel. Opposite charges attract.
- Acts between all pairs of atoms, including those in different molecules.
- Each atom carries some “partial charge” (may be a fraction of an elementary charge), which depends on which atoms it’s connected to.

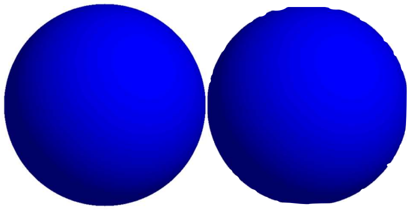
van der Waals interaction



- van der Waals forces act between all pairs of atoms and do not depend on charge.
- When two atoms are too close together, they repel strongly.
- When two atoms are a bit further apart, they attract one another weakly.



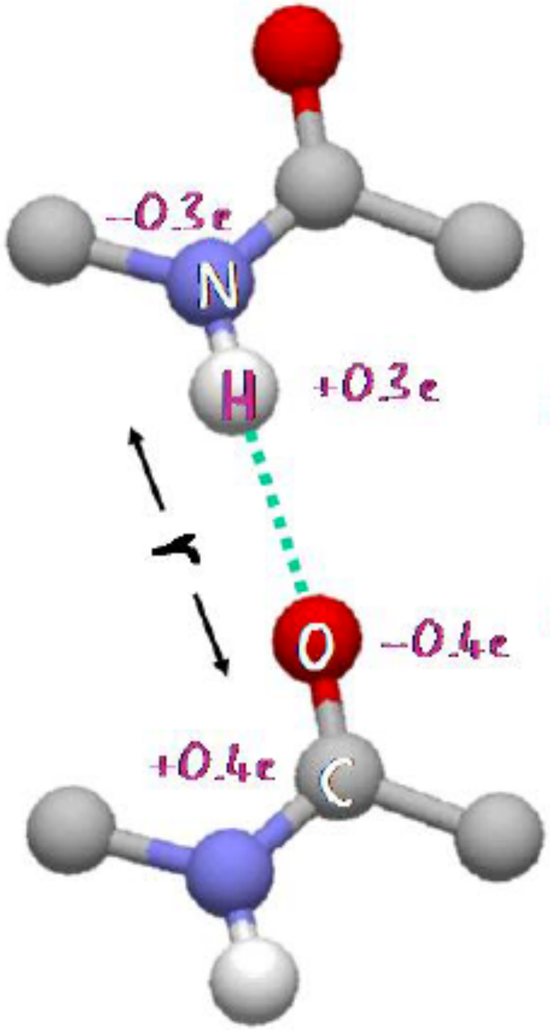
Energy is minimal when atoms are “just touching” one another

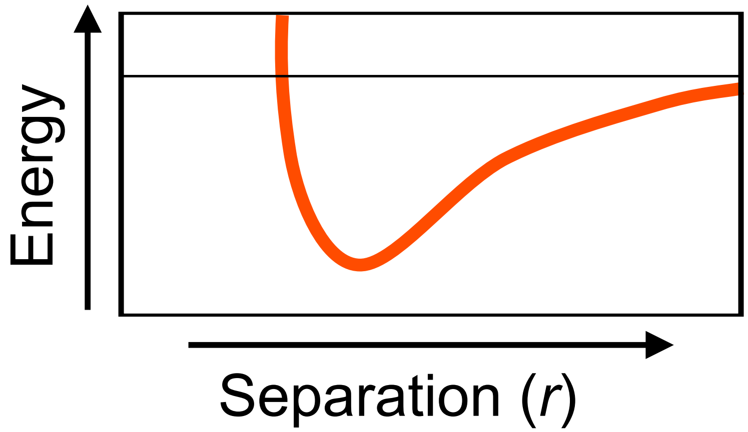


What determines the 3D structure of a protein?
Physics underlying biomolecular structure

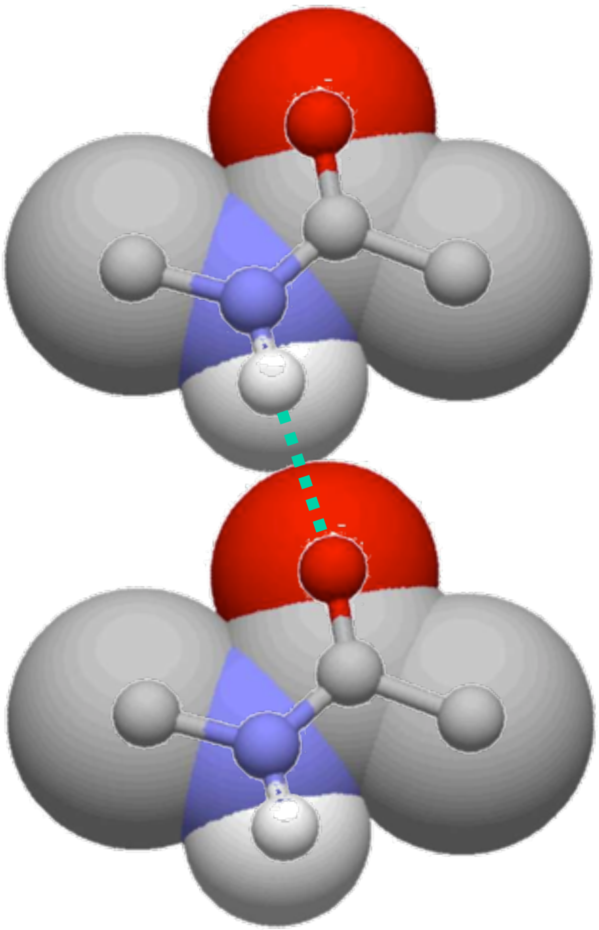
Complex interactions

Hydrogen bonds





- Favorable interaction between an electronegative atom (e.g., N or O) and a hydrogen bound to another electronegative atom
- Result of multiple electrostatic and van der Waals interactions
- Very sensitive to geometry of the atoms (distance and alignment)
- Strong relative to typical van der Waals or electrostatic forces
- Critical to protein structure



Hydrogen bonds

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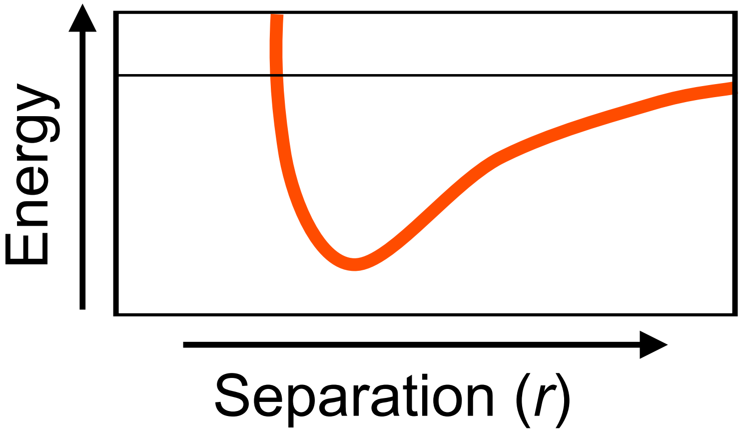
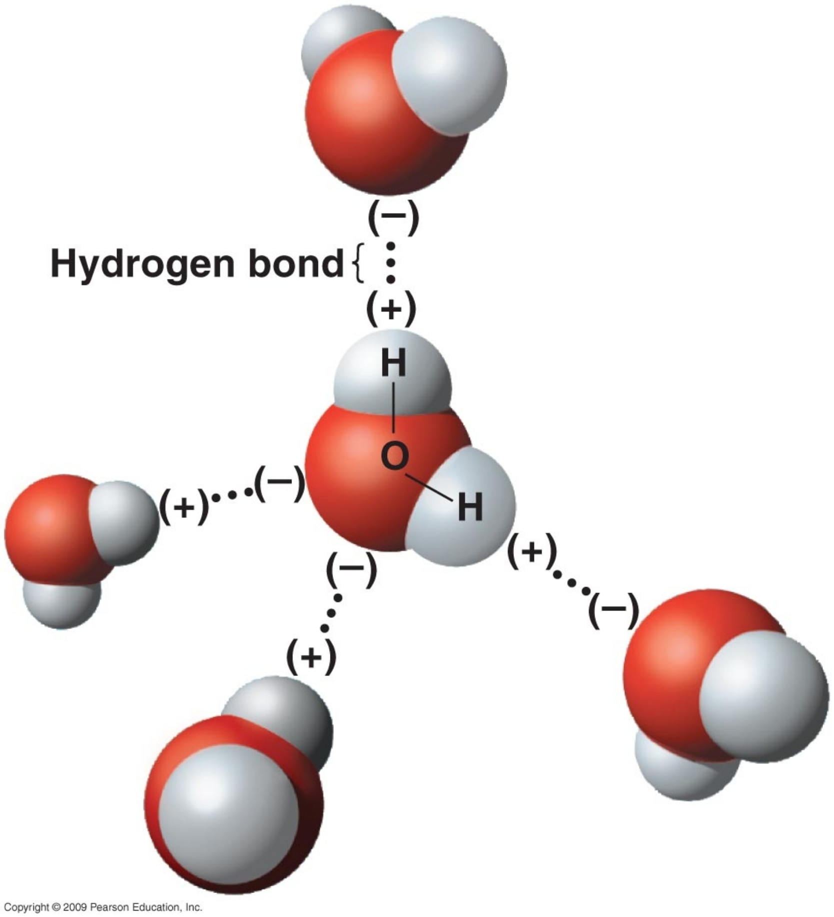


Water molecules form hydrogen bonds

- Water molecules form extensive hydrogen bonds with one another and with protein atoms
- The structure of a protein depends on the fact that it is surrounded by water

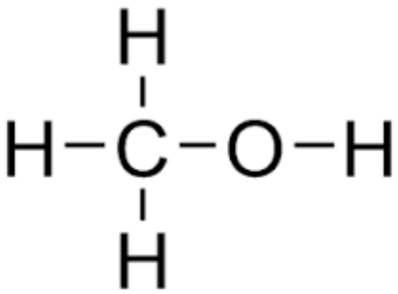


http://like-img.com/show/hydrogen-bond-water-molecule.html

Hydrophilic vs. hydrophobic

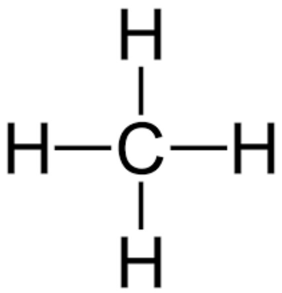
- Hydrophilic molecules are polar and thus form hydrogen bonds with water
  - Polar = contains charged atoms. Molecules containing oxygen or nitrogen are usually polar.
- Hydrophobic molecules are apolar and don't form hydrogen bonds with water

Hydrophilic (polar)



$$\mathrm{H-\overset{\displaystyle H}{\underset{\displaystyle H}{C}}-O-H}$$

Hydrophobic (apolar)



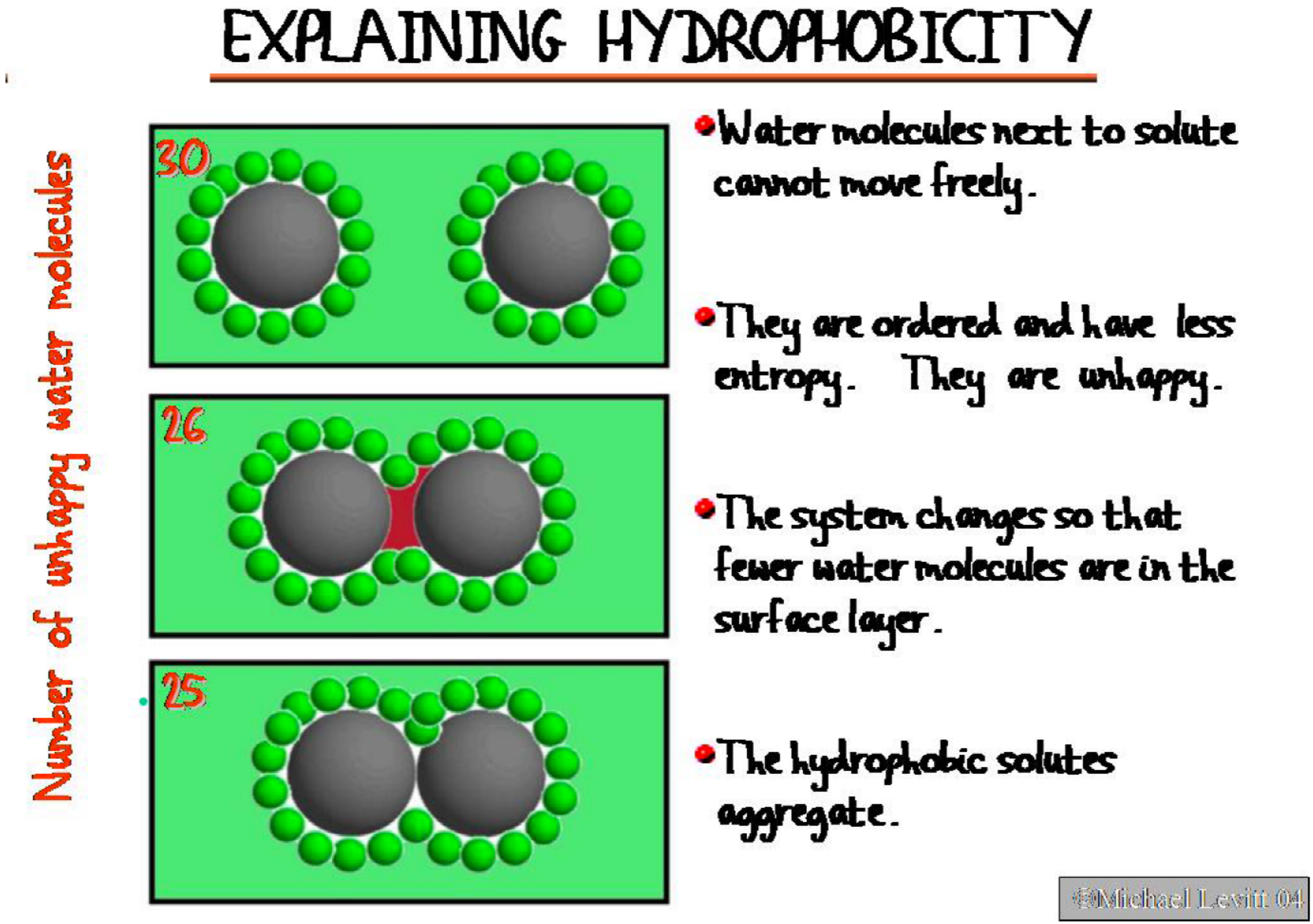
$$\mathrm{H-\overset{\displaystyle H}{\underset{\displaystyle H}{C}}-H}$$

Hydrophobic effect

- Hydrophobic molecules cluster in water
  - “Oil and water don’t mix”

http://science.taskermilward.org.uk/mod1/KS4Chemistry/AQA/Module2/Mod%202%20img/Oil-in-Water18.jpg

- This is critical to protein structure



EXPLAINING HYDROPHOBICITY

- Water molecules next to solute cannot move freely.
- They are ordered and have less entropy. They are unhappy.
- The system changes so that fewer water molecules are in the surface layer.
- The hydrophobic solutes aggregate.

- Slide from Michael Levitt
- We will discuss entropy next week. If this isn’t clear now, don’t worry.

Protein structure: a more detailed view

“Levels” of protein structure

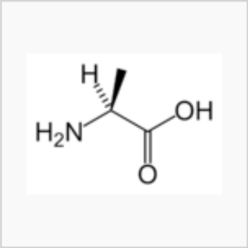
- Primary structure: sequence of amino acids
- Secondary structure: local structural elements
- Tertiary structure: overall structure of the polypeptide chain
- Quaternary structure: how multiple polypeptide chains come together

Protein structure: a more detailed view

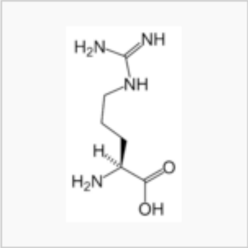
Properties of amino acids

Proteins are built from amino acids

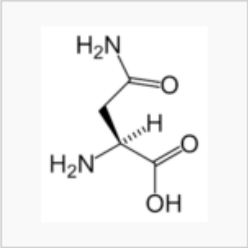
- 20 “standard” amino acids
- Each has three-letter and one-letters abbreviations (e.g., Threonine = Thr = T; Tryptophan = Trp = W)



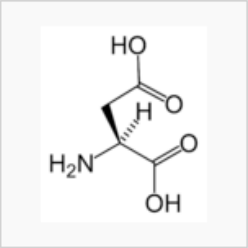
L-Alanine
(Ala / A)



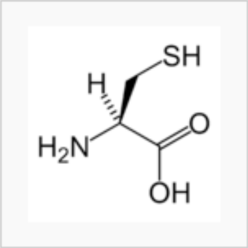
L-Arginine
(Arg / R)



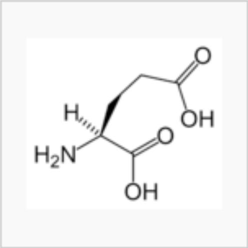
L-Asparagine
(Asn / N)



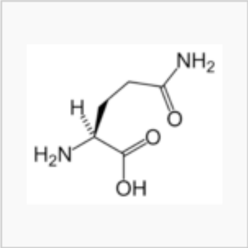
L-Aspartic acid
(Asp / D)



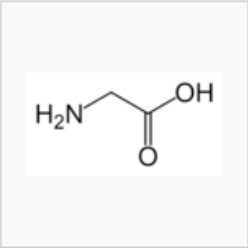
L-Cysteine
(Cys / C)



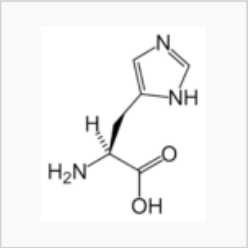
L-Glutamic acid
(Glu / E)



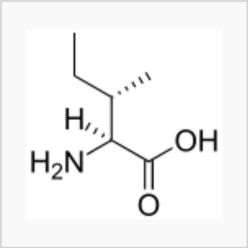
L-Glutamine
(Gln / Q)



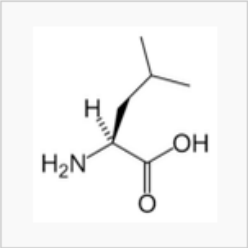
Glycine
(Gly / G)



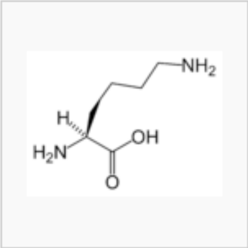
L-Histidine
(His / H)



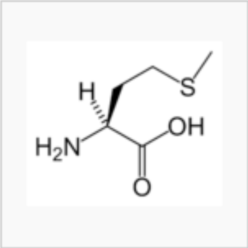
L-Isoleucine
(Ile / I)



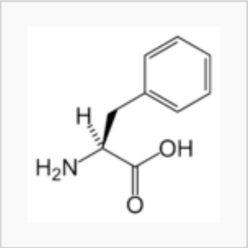
L-Leucine
(Leu / L)



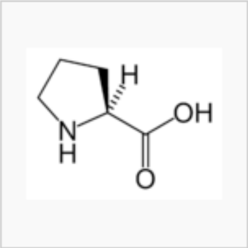
L-Lysine
(Lys / K)



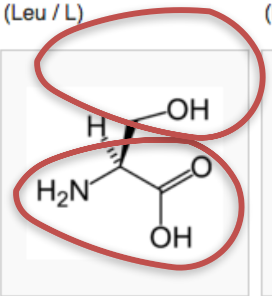
L-Methionine
(Met / M)



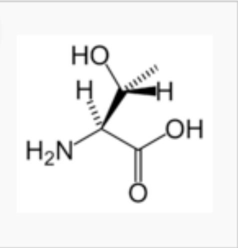
L-Phenylalanine
(Phe / F)



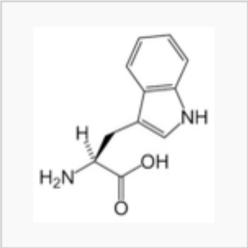
L-Proline
(Pro / P)



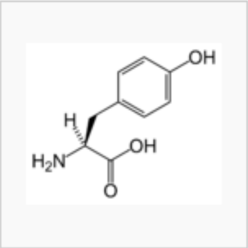
L-Serine
(Ser / S)



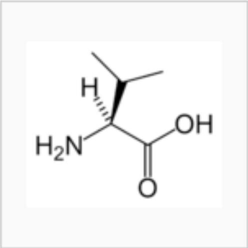
L-Threonine
(Thr / T)



L-Tryptophan
(Trp / W)



L-Tyrosine
(Tyr / Y)



L-Valine
(Val / V)

The “side chain” is different in each amino acid

All amino acids have this part in common.

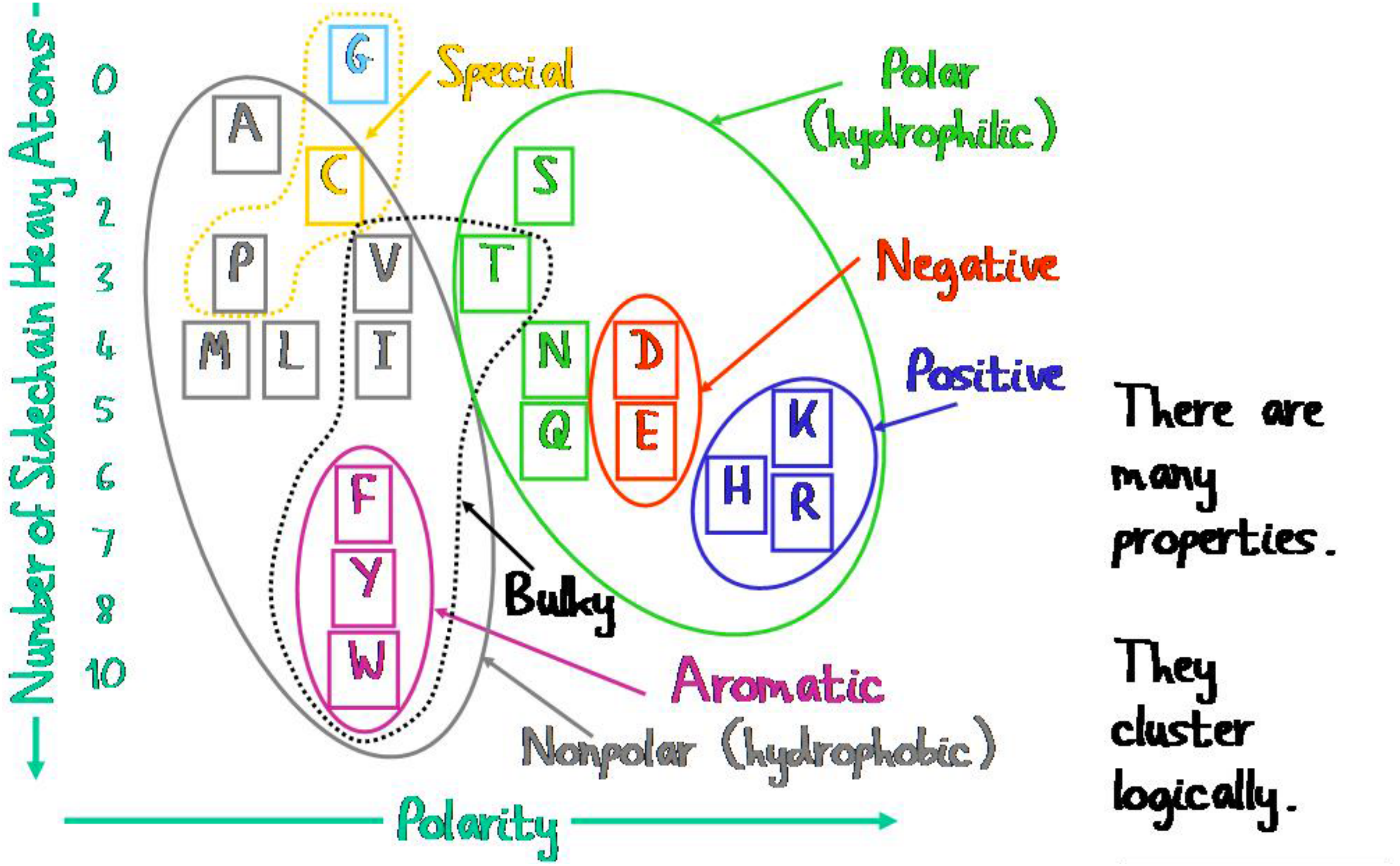
You don’t need to memorize all the structures

https://en.wikipedia.org/wiki/Proteinogenic_amino_acid

Amino acid properties

- Amino acid side chains have a wide range of properties. These differences bring about the 3D structures of proteins.
- Examples:
  - Large side chains take up more space than small ones
  - Hydrophobic side chains want to be near one another
  - Hydrophilic side chains form hydrogen bonds to one another and to water molecules
  - Negatively charged (acidic) side chains want to be near positively charged (basic) side chains

Amino acid properties



There are many properties.

They cluster logically.

Slide from Michael Levitt

You don't need to memorize which amino acids have which properties

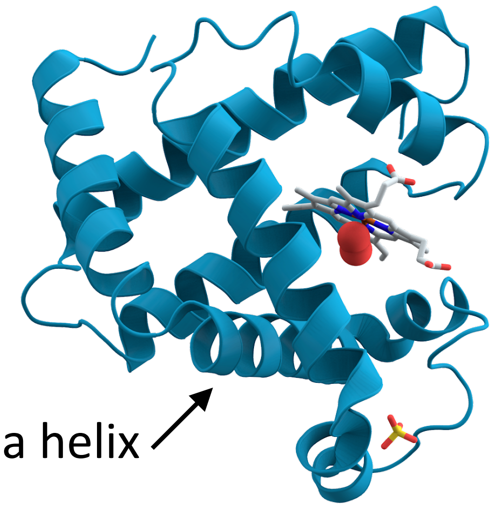
Protein structure: a more detailed view

Secondary structure

Secondary structure

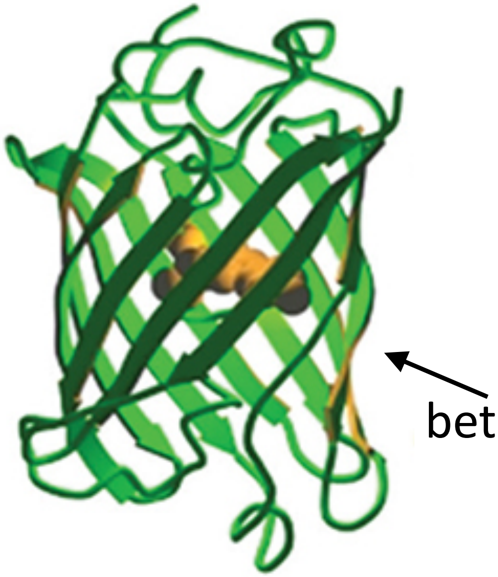
- “Secondary structure” refers to certain local structural elements found in many proteins
  - These are energetically favorable primarily because of hydrogen bonds between backbone atoms
- Most important secondary structure elements:
  - alpha helix
  - beta sheet

Myoglobin



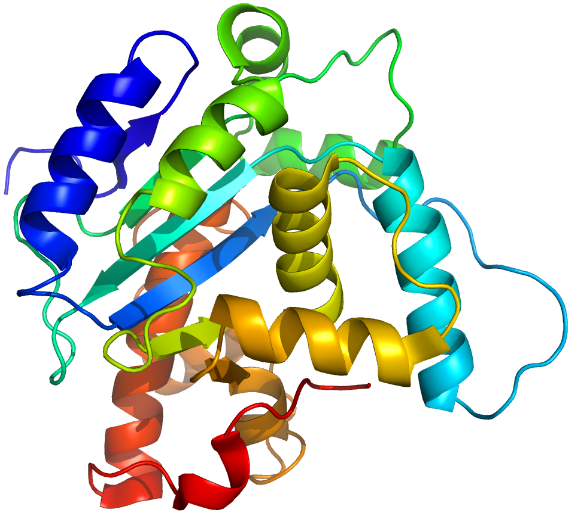
alpha helix

Green Fluorescent Protein



beta sheet

Pop2p



https://upload.wikimedia.org/wikipedia/commons/

http://www.biotek.com/assets/tech_resources/11596/figure2.jpg

http://upload.wikimedia.org/wikipedia/commons/e/e6/Spombe_Pop2p_protein_structure_rainbow.png

The alpha helix

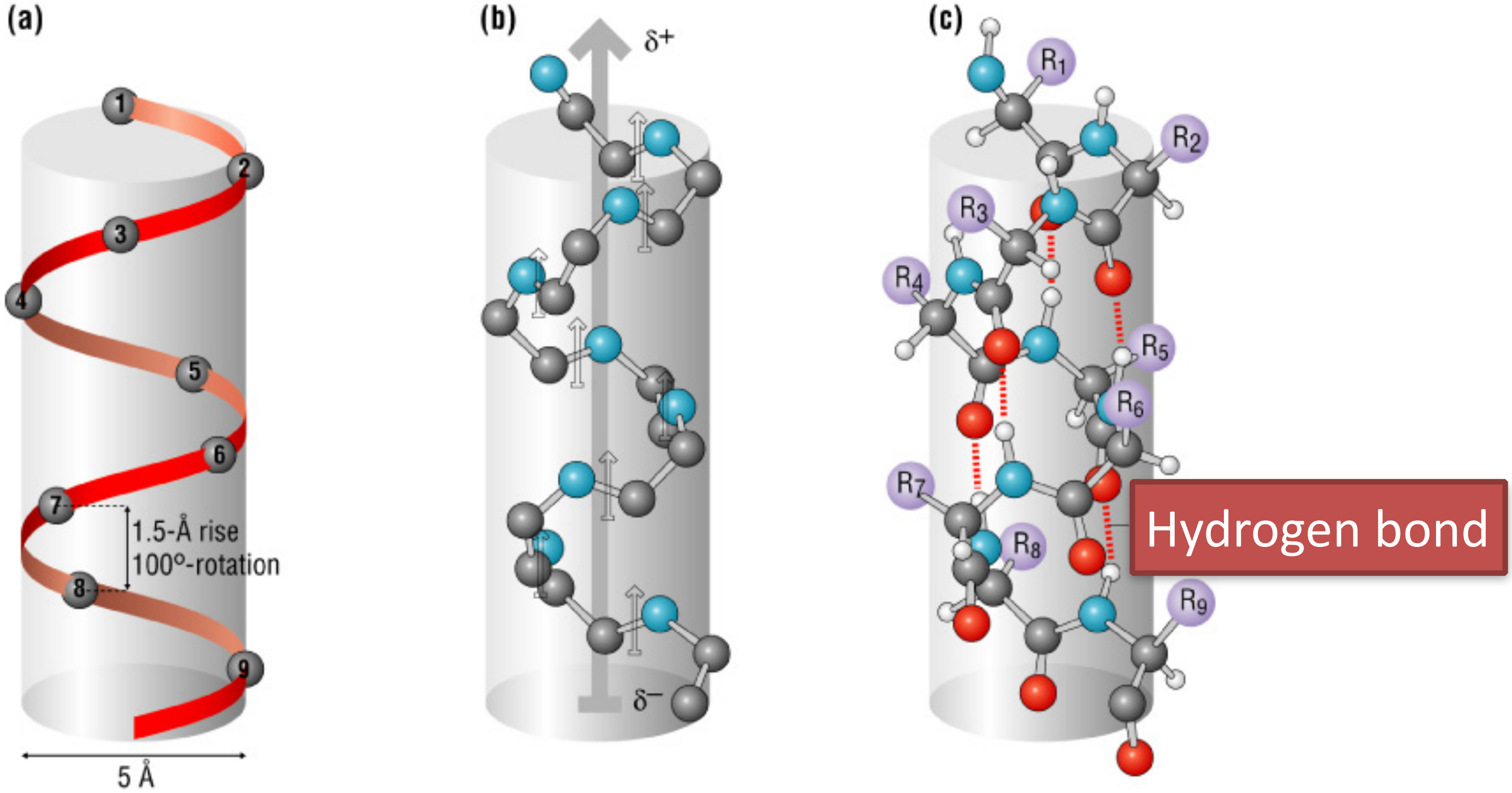
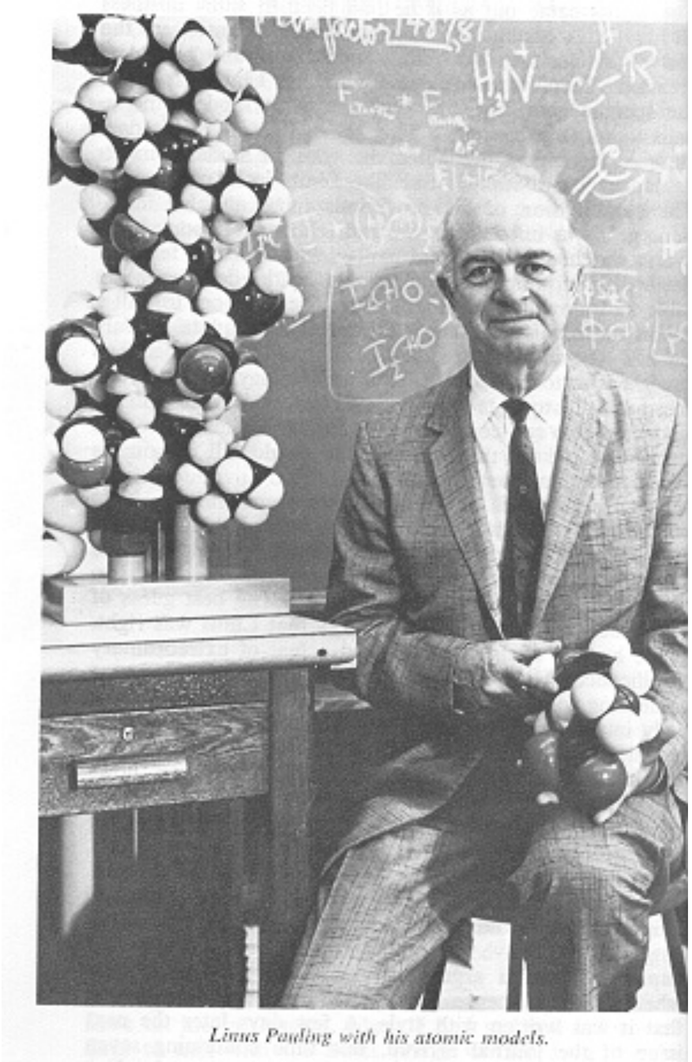


Image from "Protein Structure and Function"
by Gregory A Petsko and Dagmar Ringe

The alpha helix



Linus Pauling with his atomic models.

Linus Pauling

https://www.msu.edu/course/lbs/333/fall/images/PAULING.JPG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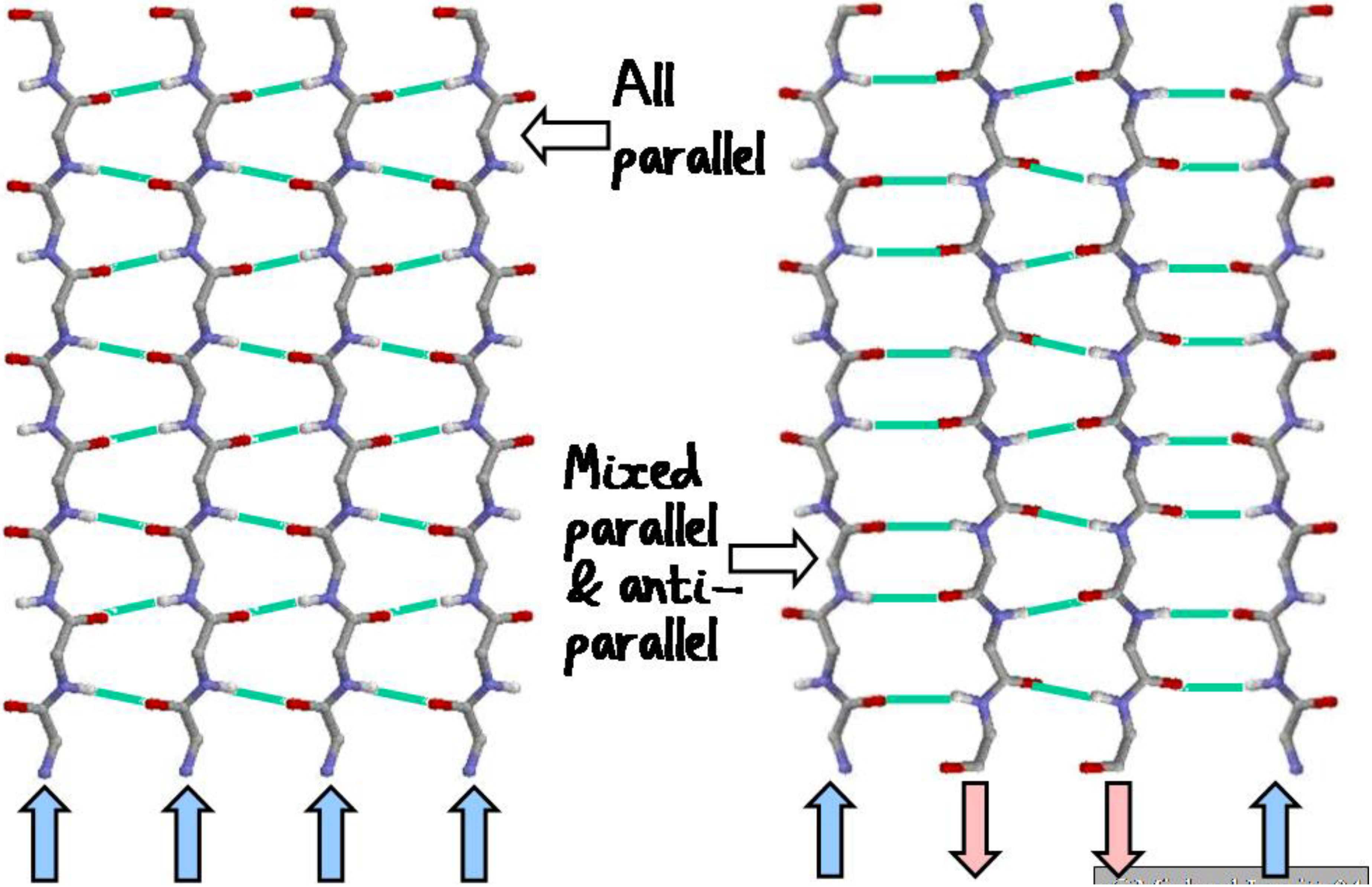
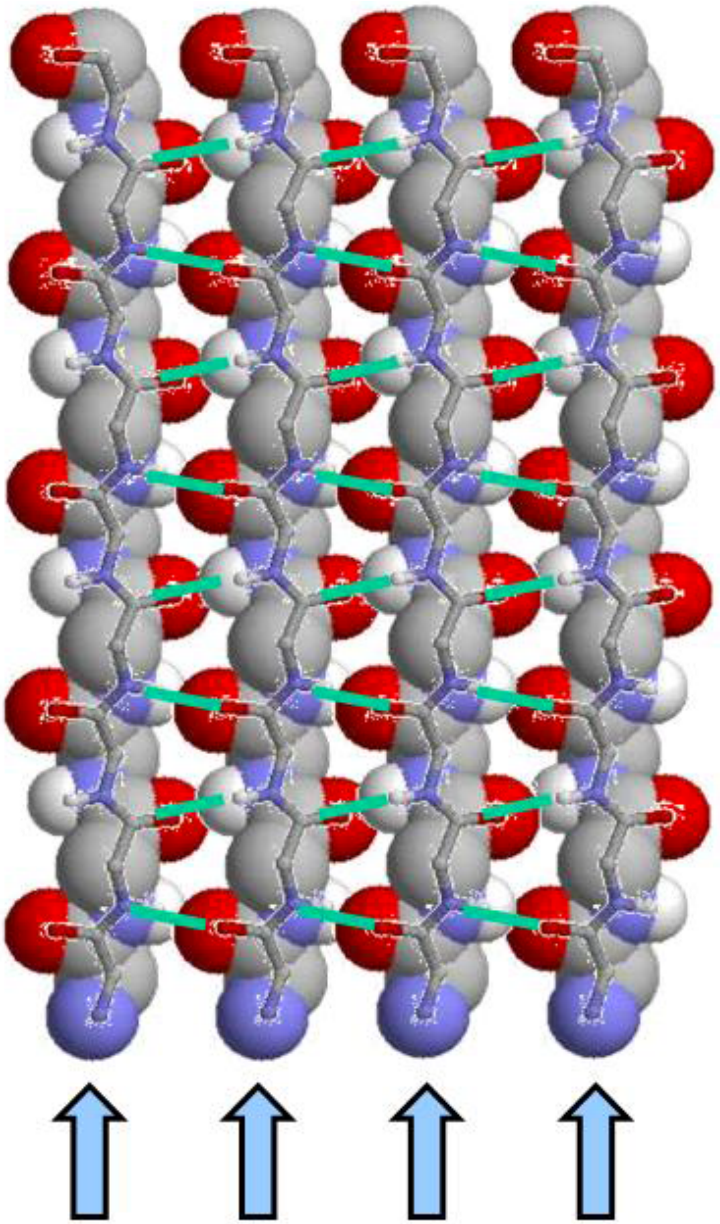
Stephen Isaacs
RESERVED PARKING
COLLEGE OF CHEMISTRY
9/30/2016
1:00 PM -6:00 PM
Prof. Ron Dror

The beta sheet



From Michael Levitt

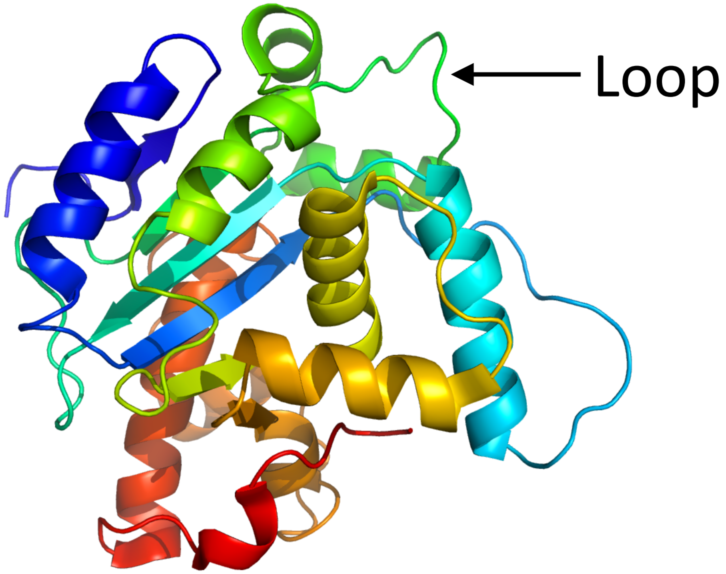
The beta sheet



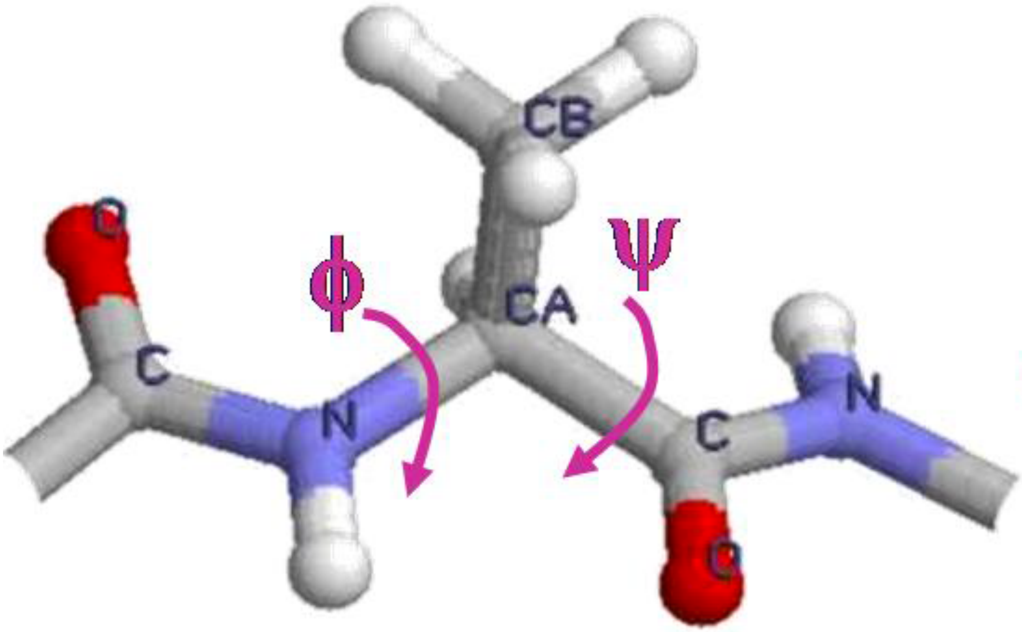
From Michael Levitt

Other secondary structure

- There are several less common secondary structures
- Regions connecting well-defined secondary structure elements are often referred to as "loops"



BACKBONE DEGREES OF FREEDOM



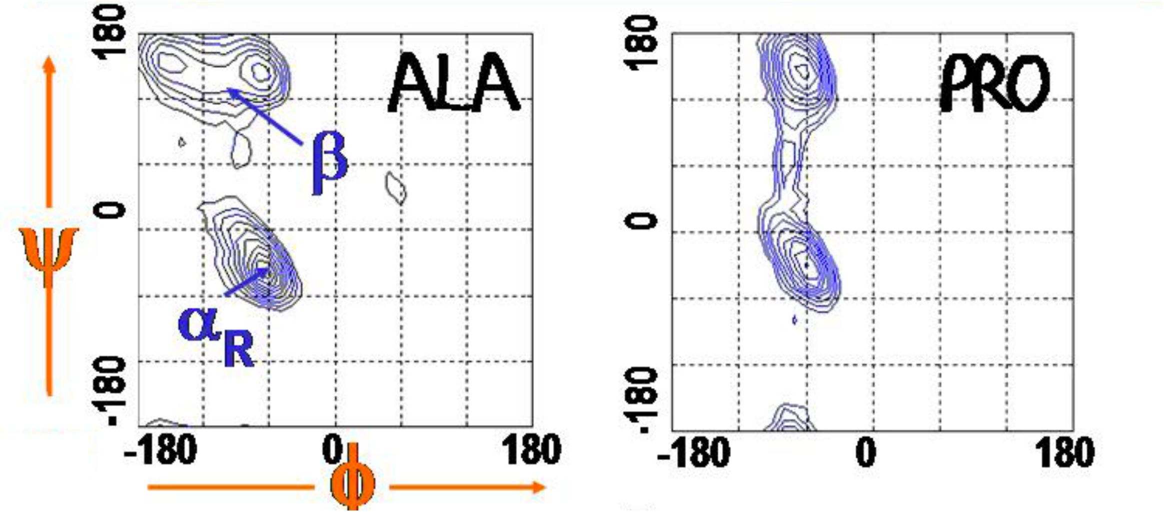
- The torsion angle rotating about the N–CA bond is called ϕ
- The torsion angle rotating about the CA–C bond is called ψ
- Together they are the (ϕ, ψ) angles

From Michael Levitt

- The remaining backbone bond (N–C, the "peptide bond") is rigid

Ramachandran diagrams

- A plot showing a distribution in the (Φ, Ψ) plane is called a Ramachandran diagram
  - Such a diagram can be a scatterplot, or a two-dimensional histogram visualized as a contour map or heat map
  - For example, one might make a Ramachandran diagram for many residues of the same amino acid type
- Some amino acid types have distinctive Ramachandran diagrams



Ala is typical
Pro is unusual

Image from
Michael Levitt

- Alpha helices and beta sheets have characteristic Ramachandran diagrams

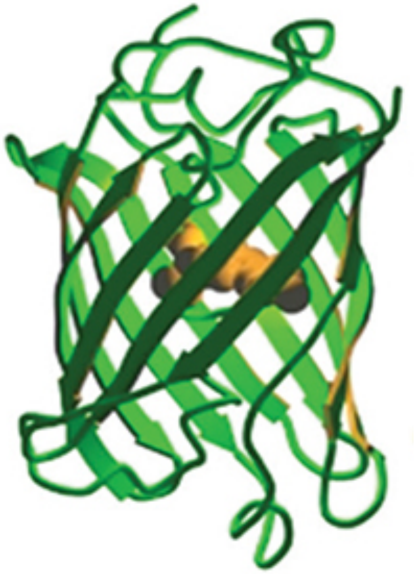
Protein structure: a more detailed view

Tertiary structure, quaternary structure, and domains

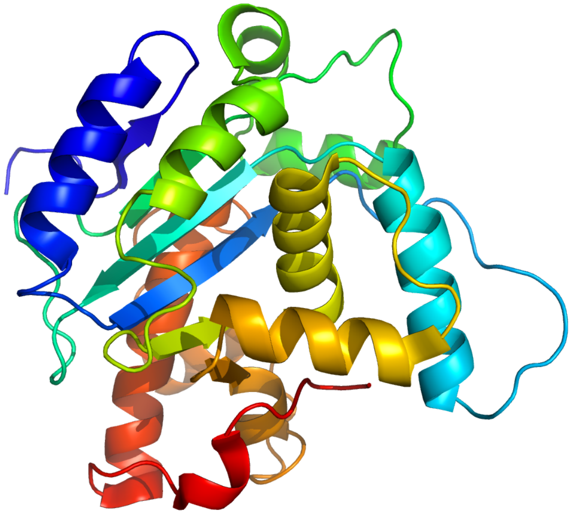
Tertiary structure

- Tertiary structure: the overall three-dimensional structure of a polypeptide chain

Myoglobin



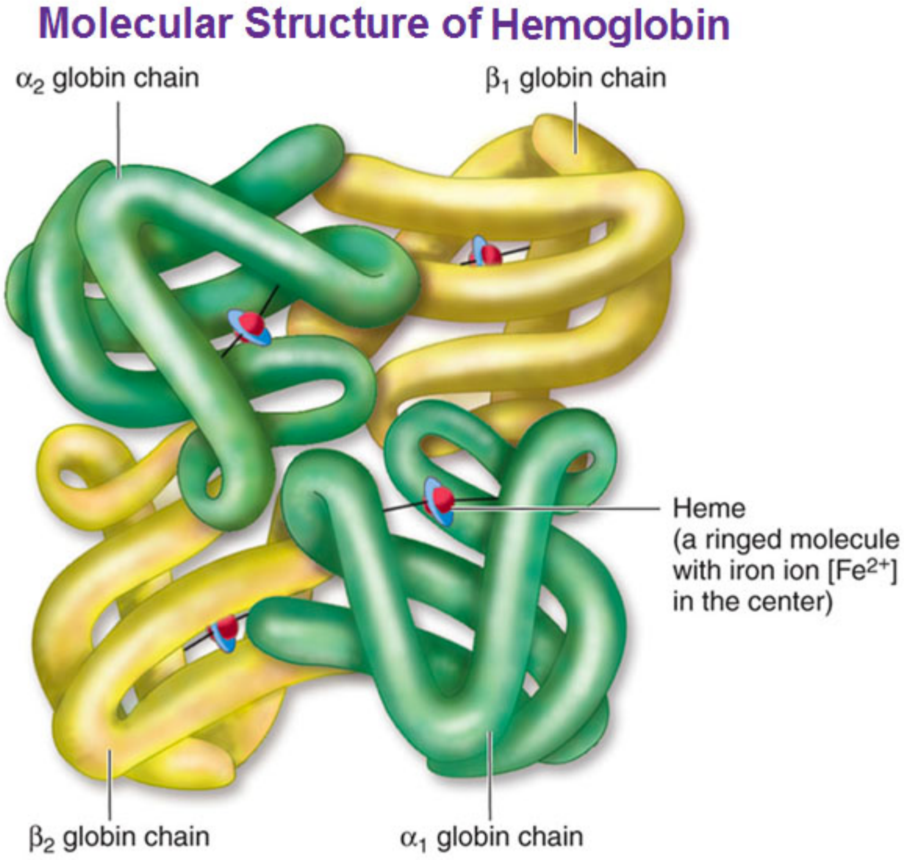
Green Fluorescent Protein

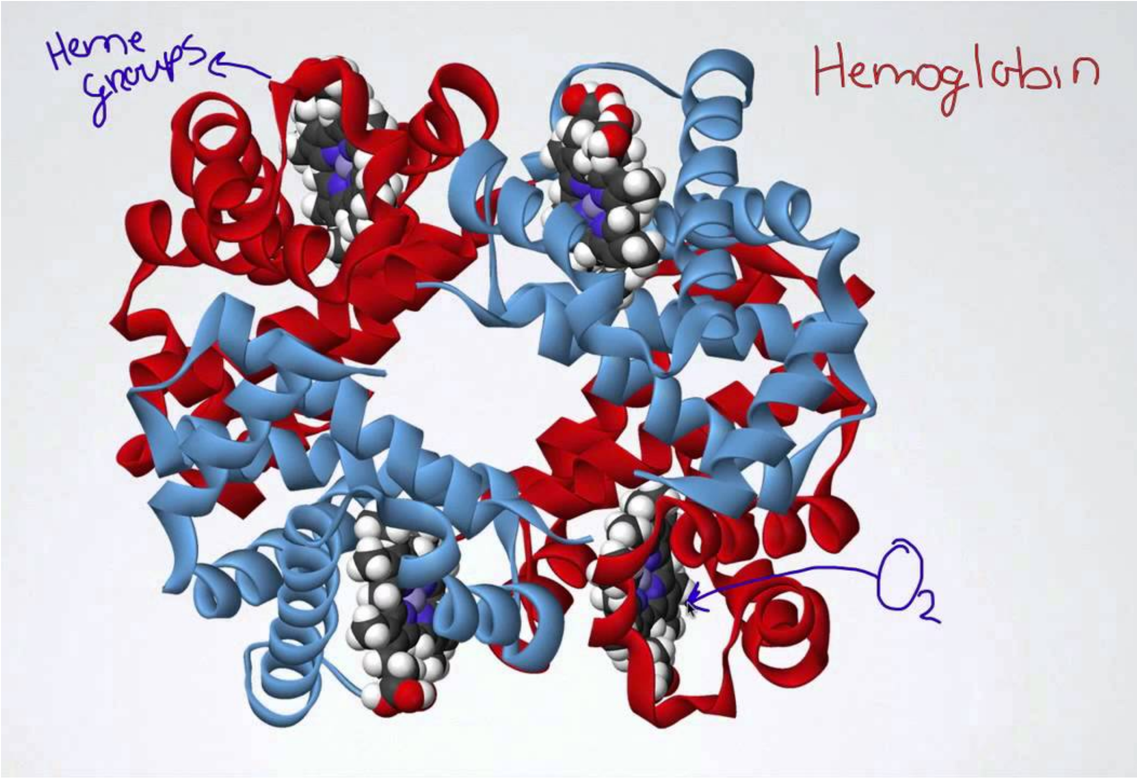


Pop2p

Quaternary structure

- Quaternary structure: the arrangement of multiple polypeptide chains in a larger protein

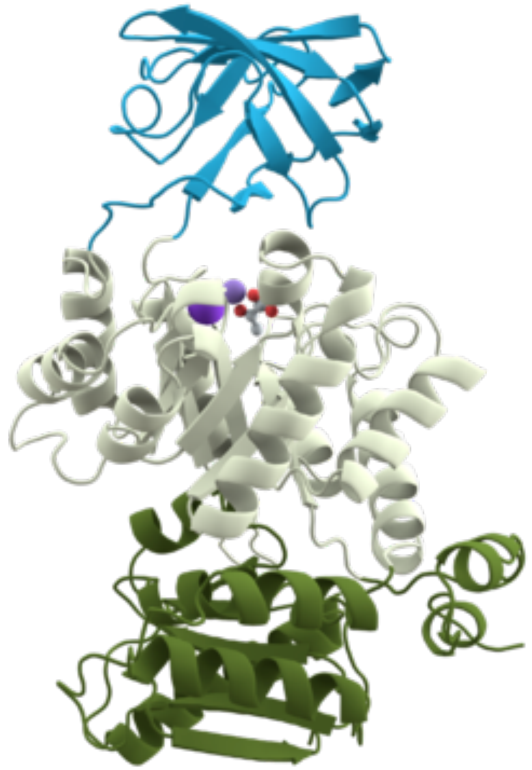




http://i.ytimg.com/vi/MKGhoC1Bf-I/maxresdefault.jpg

Domains

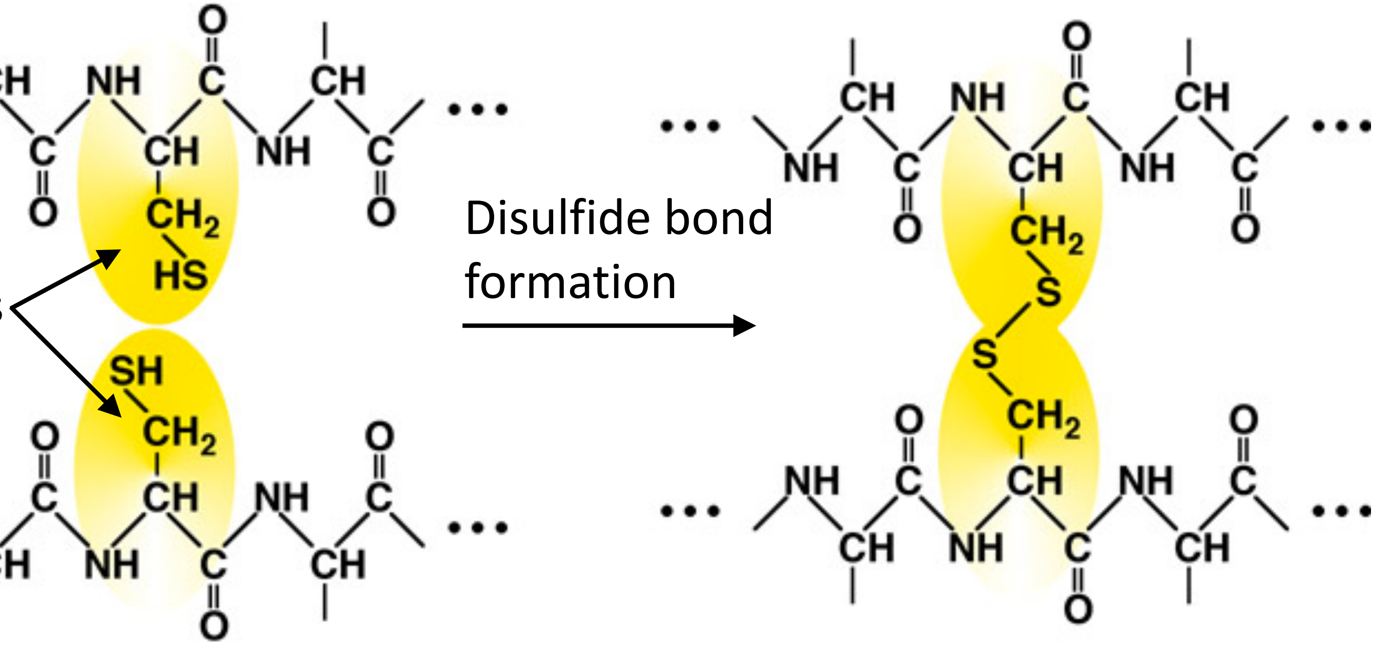
- Large proteins often consist of multiple compact 3D structures called *domains*
  - Many contacts with a domain. Few contacts between domains.
  - "Domain ≈ blob"
- One polypeptide chain can form multiple domains



http://en.wikipedia.org/wiki/Protein_domain

Disulfide bonds

- One particular amino acid type, cysteine, can form a covalent bond with another cysteine (called a disulfide bond or bridge)
- Apart from the bonds within an amino acid residue and the peptide bonds that connect residues, disulfide bonds are the only common covalent bonds within a protein
- In a typical cellular environment, disulfide bonds can be formed and broken quite easily



Cysteine side chains

Disulfide bond formation

http://www.crc.dk/yeast/yeasthome/yeasthome/images/ls_jpgs/fig2.jpg

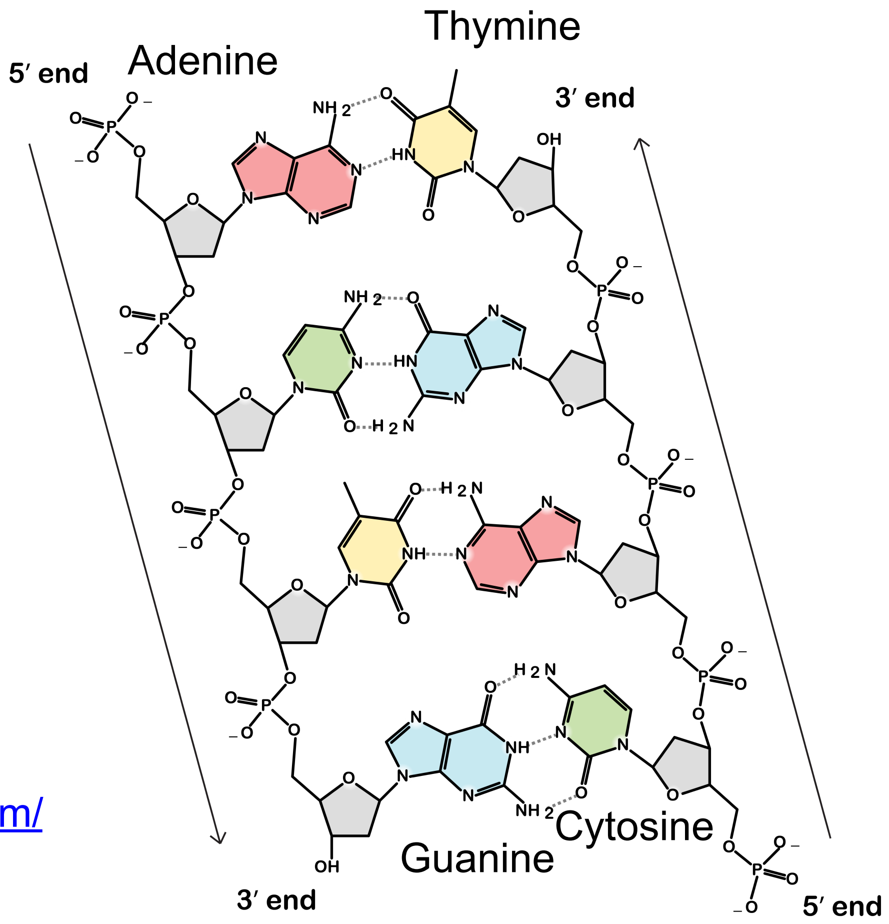
Structures of other biomolecules

What determines the structure of other biomolecules?

- The physical interactions that determine protein structure also determine the structures of other biomolecules
  - More generally, the great majority of the material covered in this course for proteins applies to other biomolecules as well

DNA

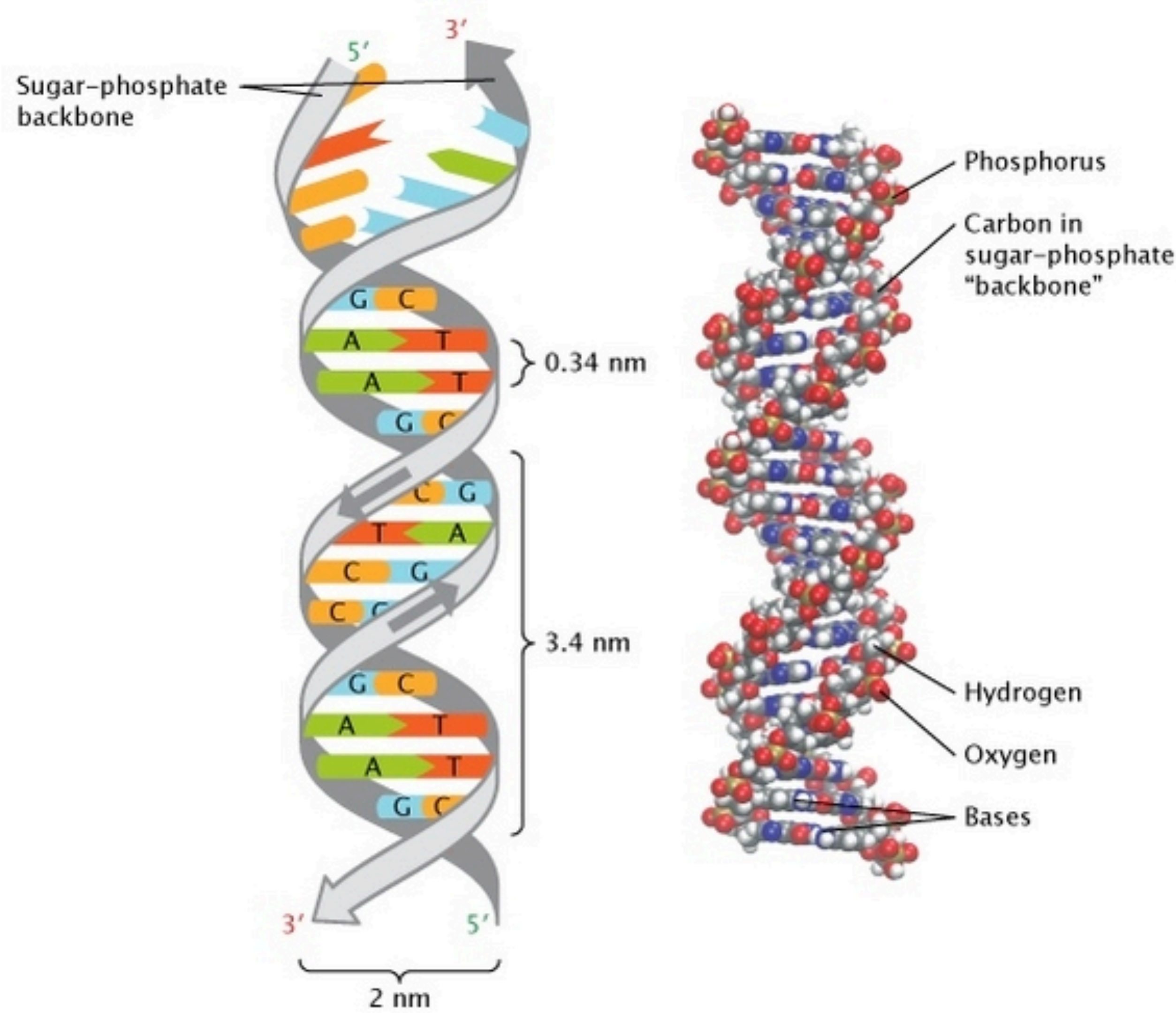
- DNA (deoxyribonucleic acid) stores the genetic code
- DNA, like protein, is a string of units with a uniform backbone
  - The units are nucleotides, instead of amino acid residues
  - Different nucleotides contain different nucleobases (bases) instead of side chains
- Only four common DNA bases
  - Adenine pairs with Thymine
  - Guanine pairs with Cytosine



Khan Academy (https://ka-perseus-images.s3.amazonaws.com/9d1d07df110f35ba532c792c73bceb164679a165.png)

DNA

- DNA forms one dominant 3D structure: a double helix
  - DNA acts more as information storage than as “machinery”
  - Long stretches of double helix can form coarser-scale structures, as we’ll see later on



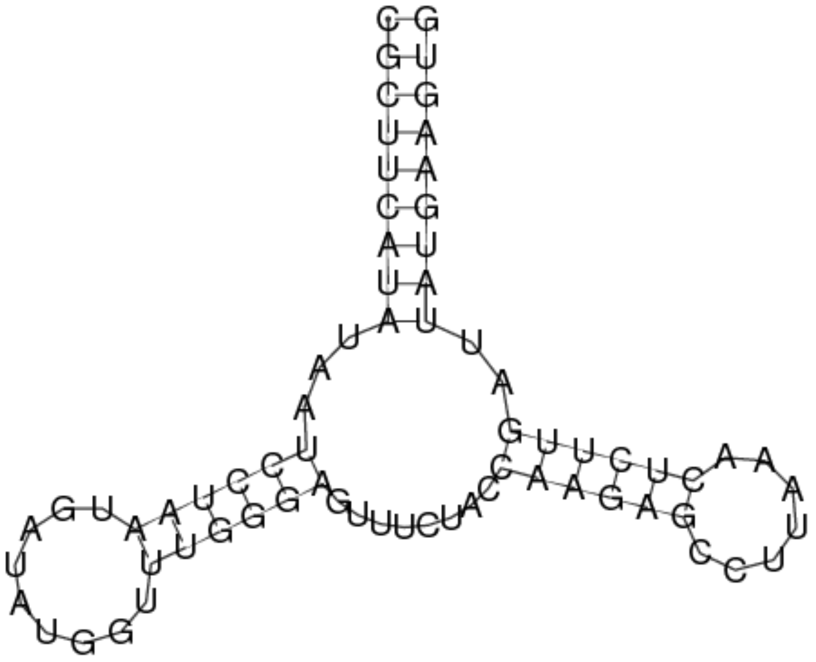
http://www.nature.com/scitable/content/ne0000/ne0000/ne0000/ne0000/104944953/73_1_2.jpg

Cambridge, 1953. Shortly before discovering the structure of DNA, Watson and Crick, depressed by their lack of progress, visit the local pub.

Source unknown

RNA

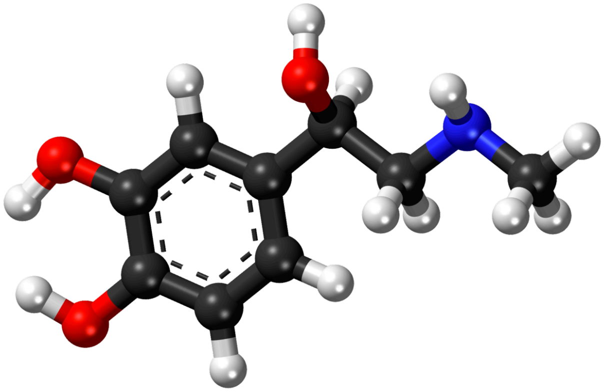
- RNA (ribonucleic acid) is a string of nucleotides, like DNA
- RNA, however, generally occurs as a single string (strand) rather than paired strands
- RNA bases often pair with other bases in the same RNA strand
  - Much work on RNA structure focuses on the “secondary structure”: which bases pair with one another
  - Note that “secondary structure” has different meanings for RNA and protein
- RNA can form machines with well-defined, varied 3D structure
  - Example: RNA in the ribosome



http://www.tbi.univie.ac.at/~pkerp/forgi/_images/1y26_ss.png

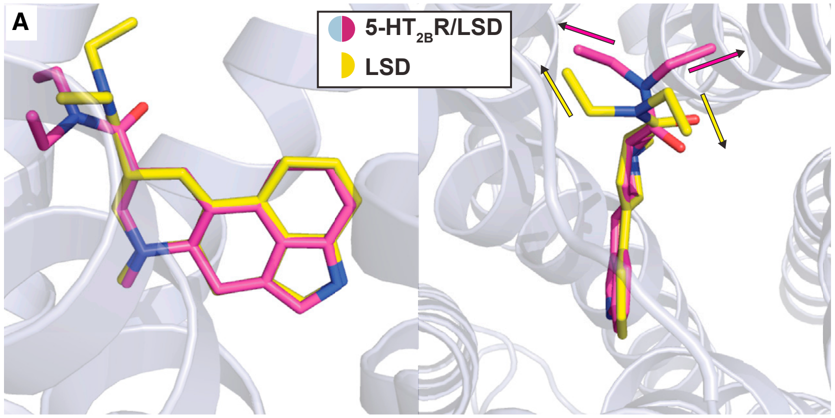
Small molecules

- Most drugs and many hormones, neurotransmitters, and other natural signaling molecules are “small molecules” (~100 atoms or fewer)
- Cambridge Structural Database is a repository of small molecule 3D structures, generally from x-ray crystallography
- However, these molecules are usually highly flexible and thus likely to take on a different 3D structure when bound to a protein



Adrenaline (epinephrine)

https://upload.wikimedia.org/wikipedia/commons/thumb/7/76/Epinephrine_ball-and-stick_model.png



LSD on its own (yellow) and receptor-bound (magenta)

Wacker et al., *Cell* (2017)