STRUCTURE CHARACTERIZATION METHODS AND STRUCTURAL APPROACH TO MEDICINE

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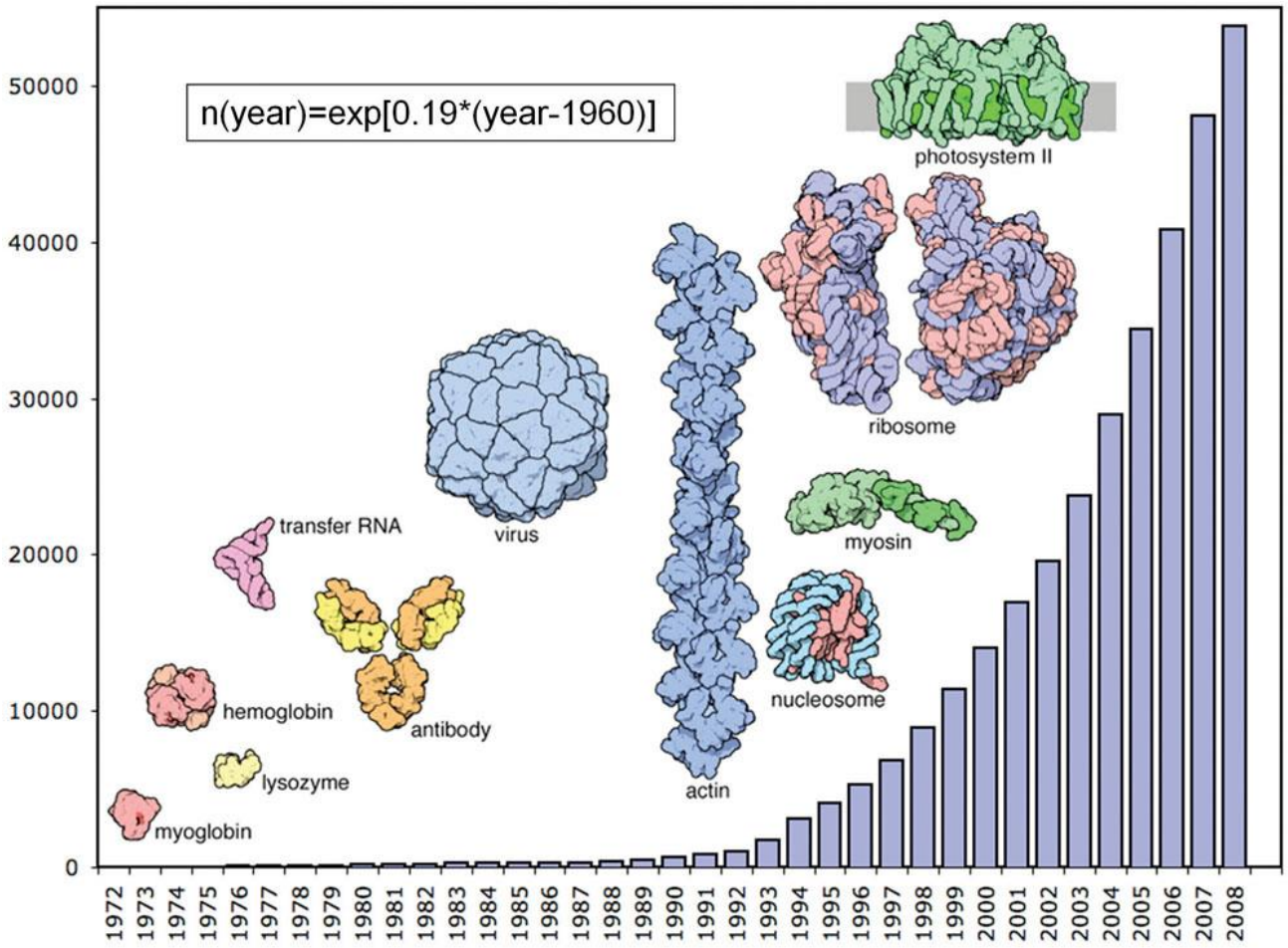
SUSTC

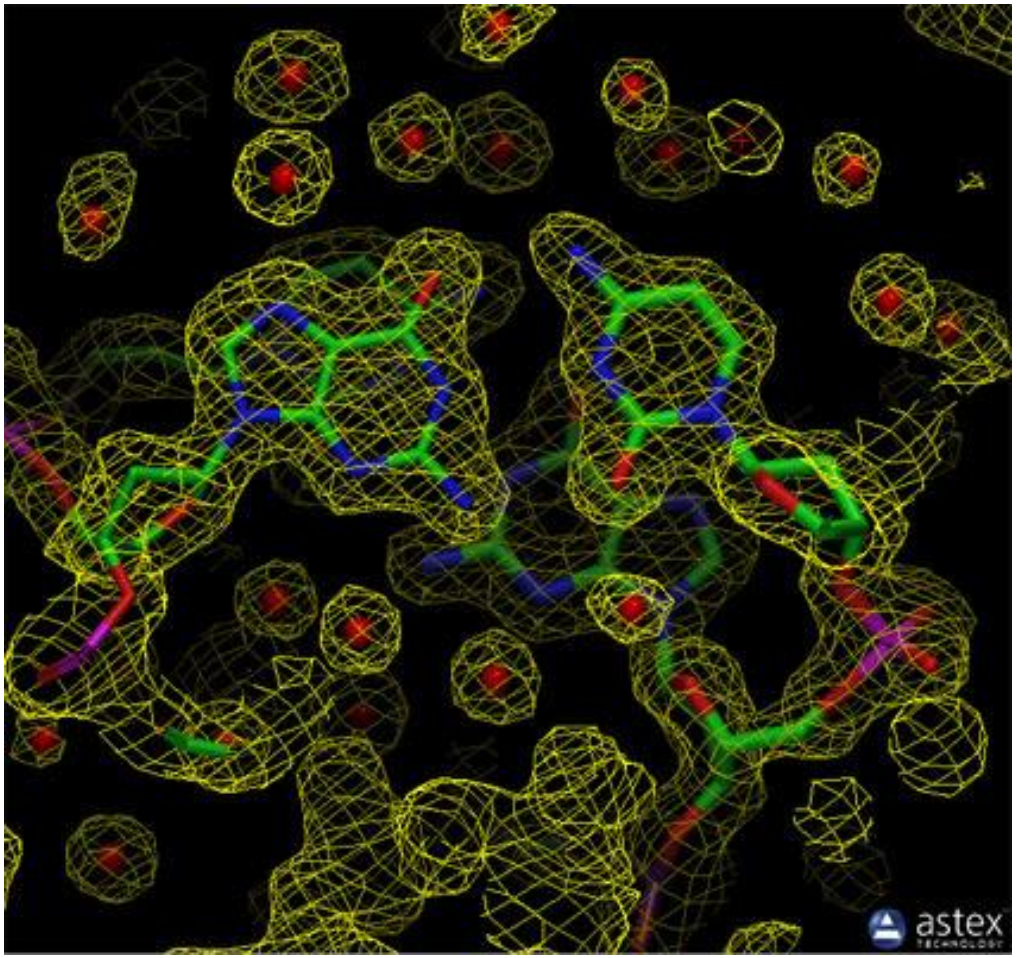
Explosive growth of solved protein structures



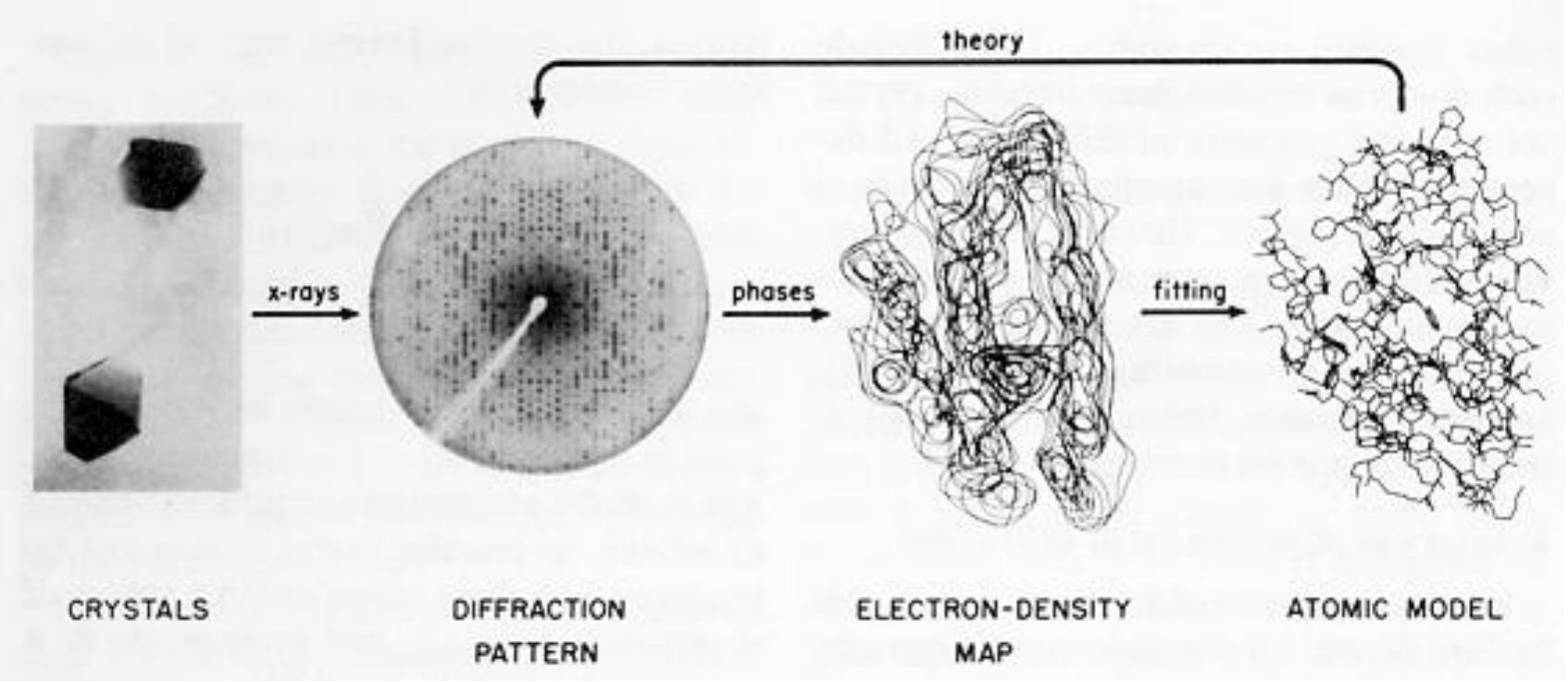
Structure determination methods

- Biophysics
  - Use of electromagnetic radiation
- X-ray crystallography
- Nuclear magnetic resonance (NMR) spectroscopy
- Electron microscopy (EM)
  - CryoEM

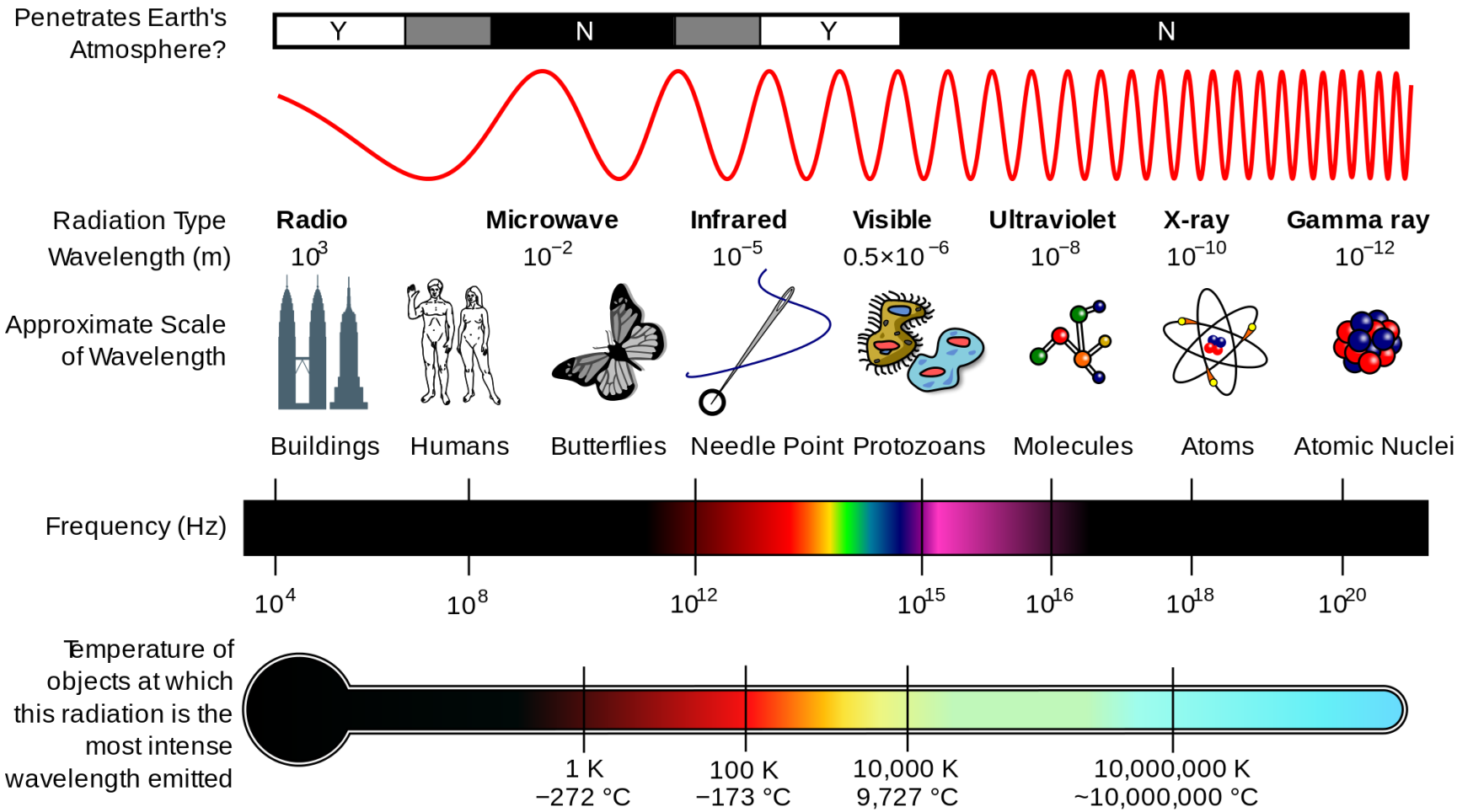
X-ray Crystallography



Structure determination by crystallography

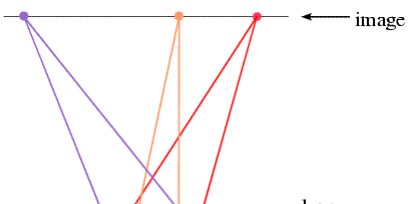


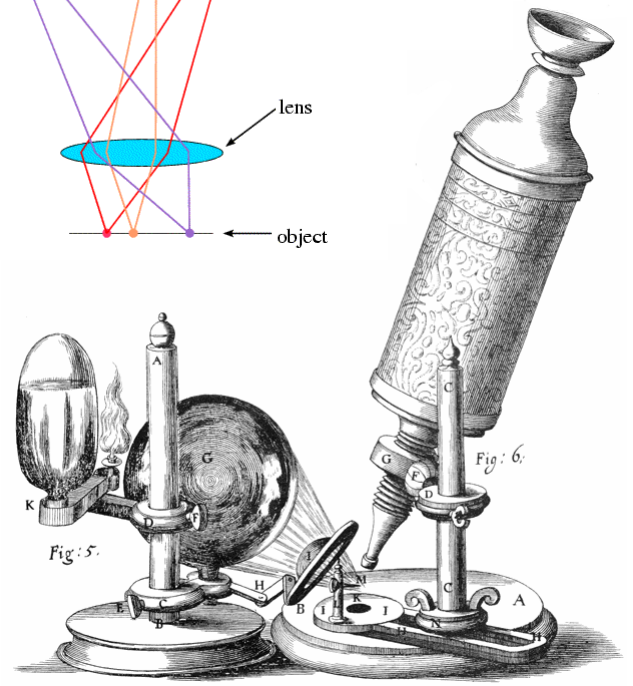
Why we need X-ray?



Why we need crystals?

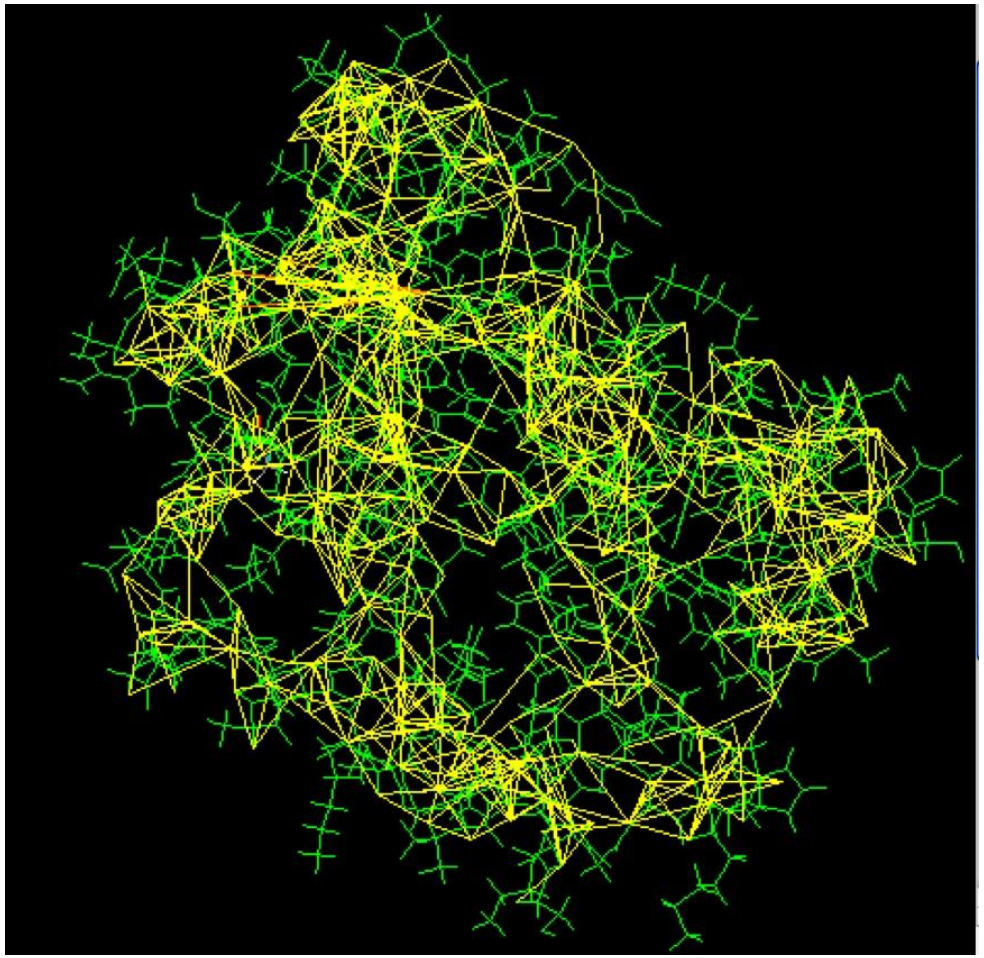
- X-ray lens are very hard to make, because of two difficulties
  - Focus X-ray
  - Atomic scale
- X-ray scattering from a single molecule would be incredibly weak and extremely difficult to detect above the noise level



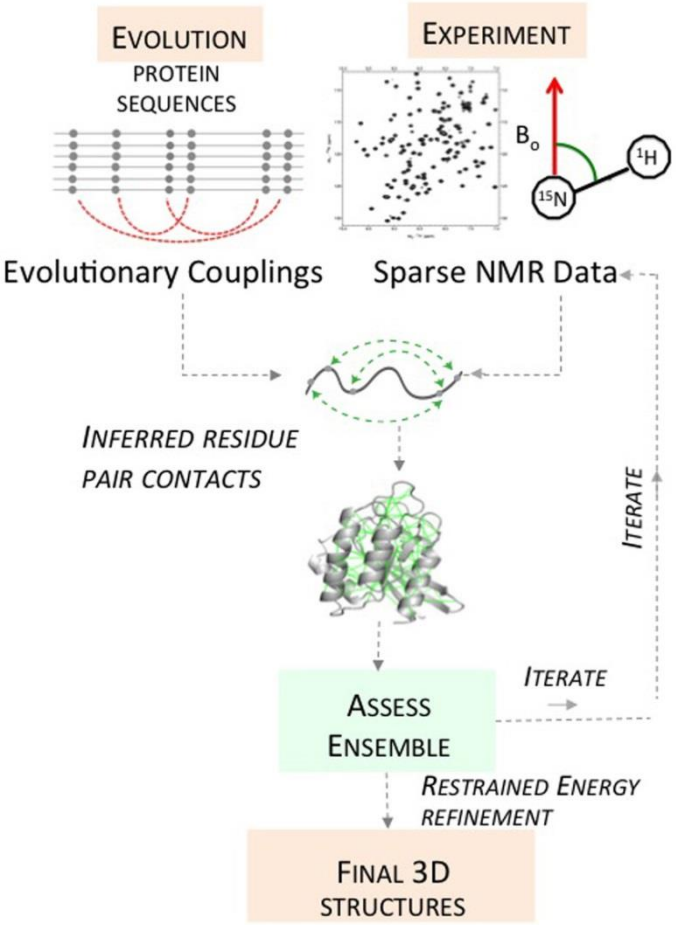


Hooke's microscope

NMR Spectroscopy



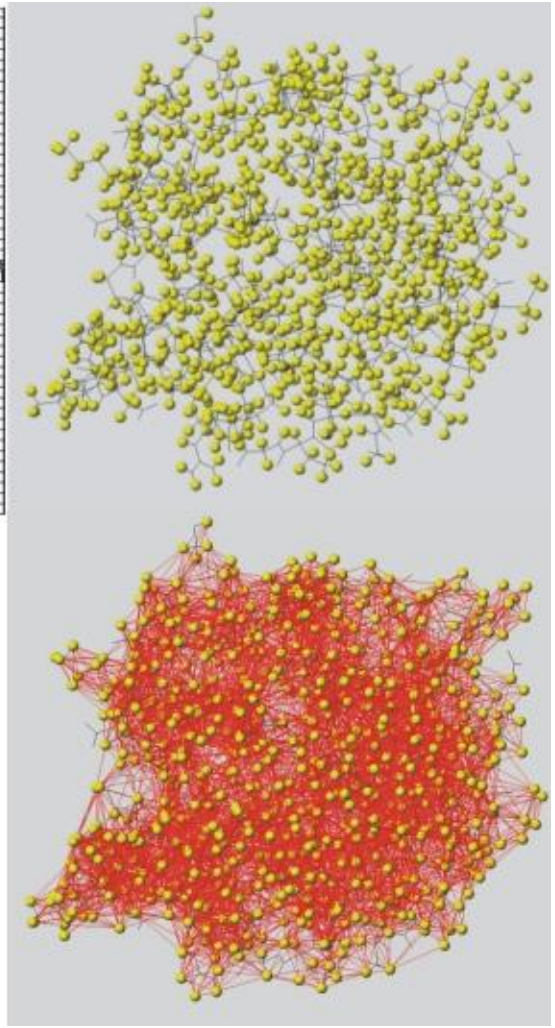
Structure determination by NMR



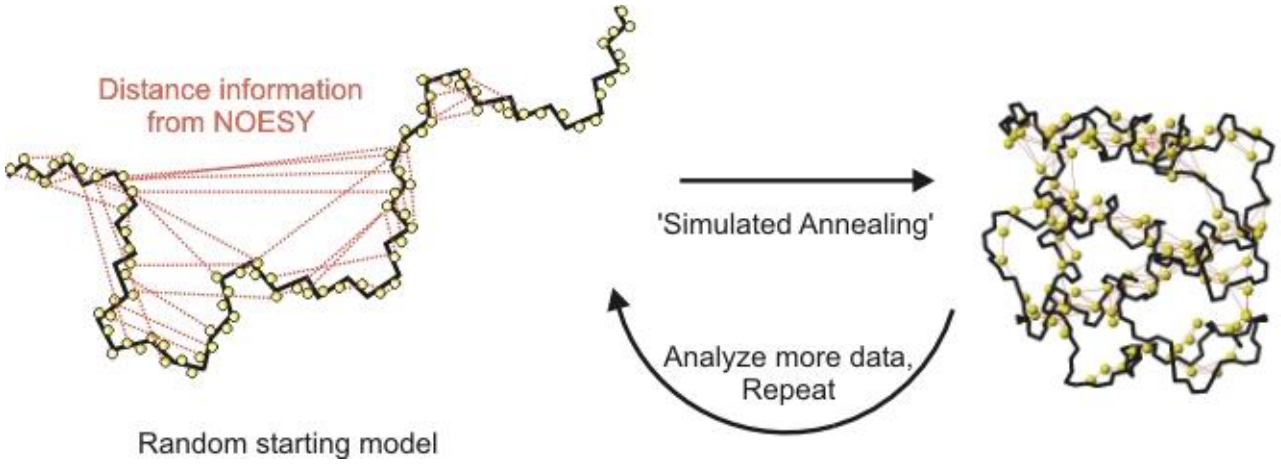
Generating powerful magnetic field

2D NOESY spectrum of a protein

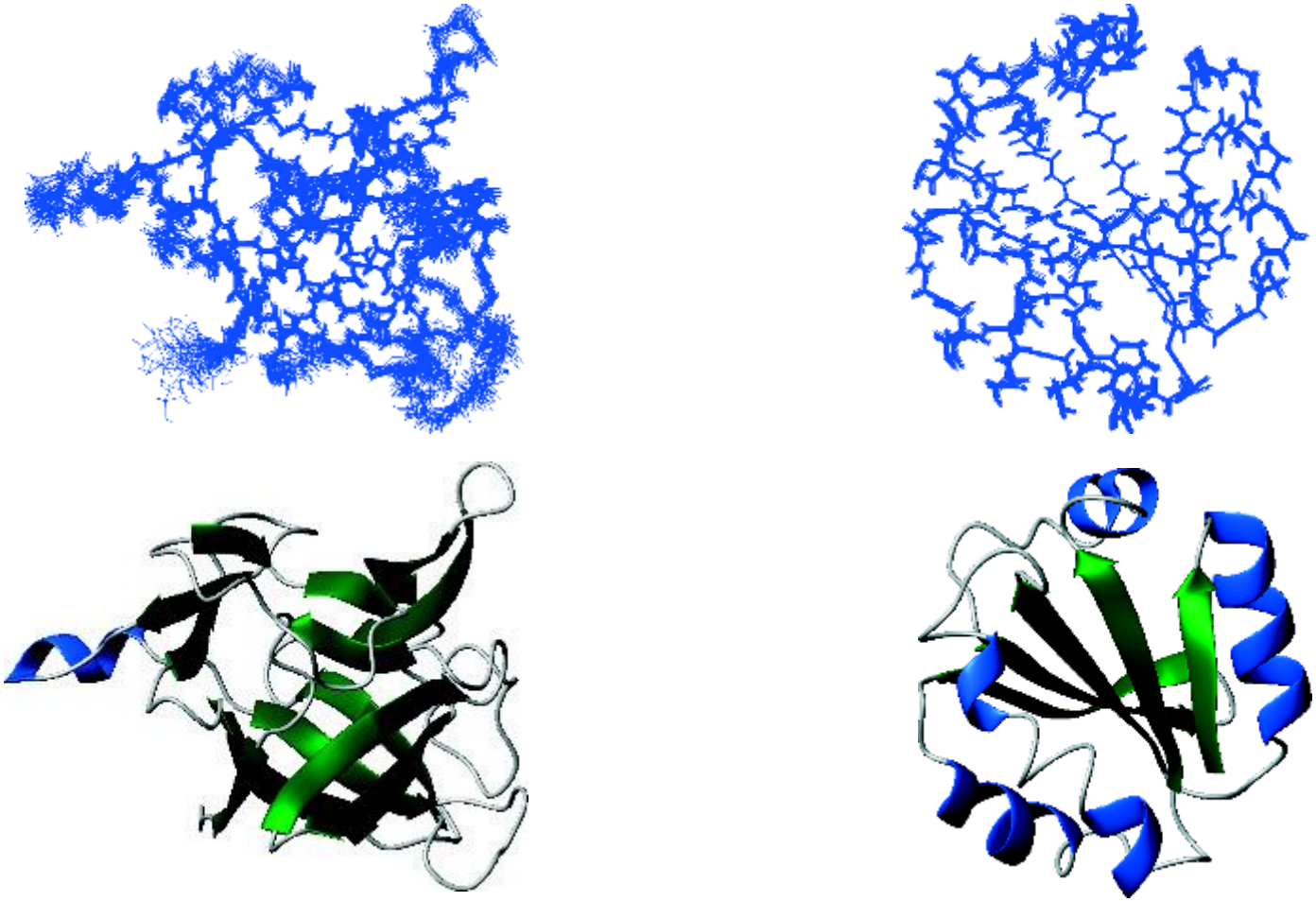
All pairs of protons closer than ~5 Å



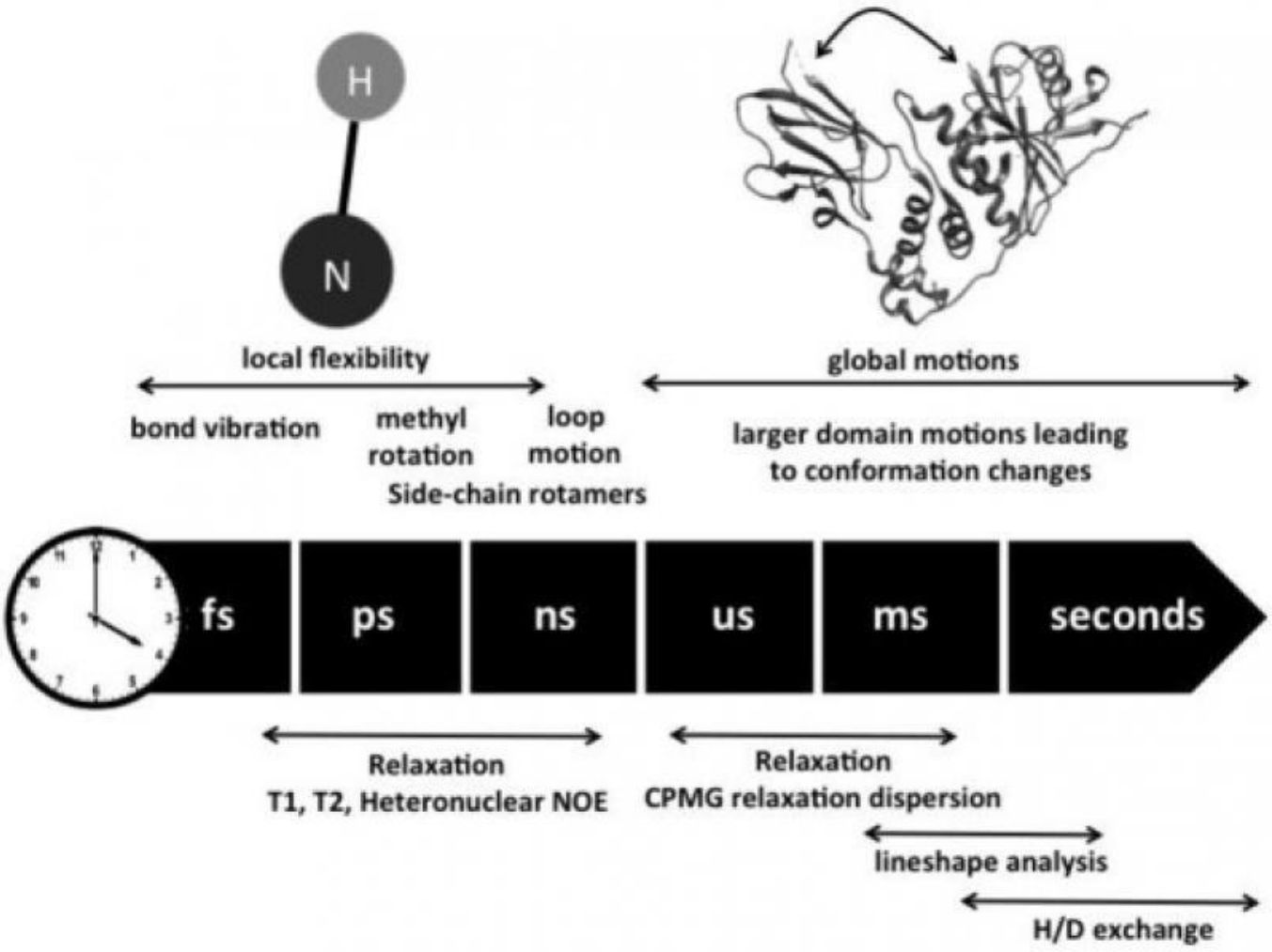
Standard approach to NMR protein structure



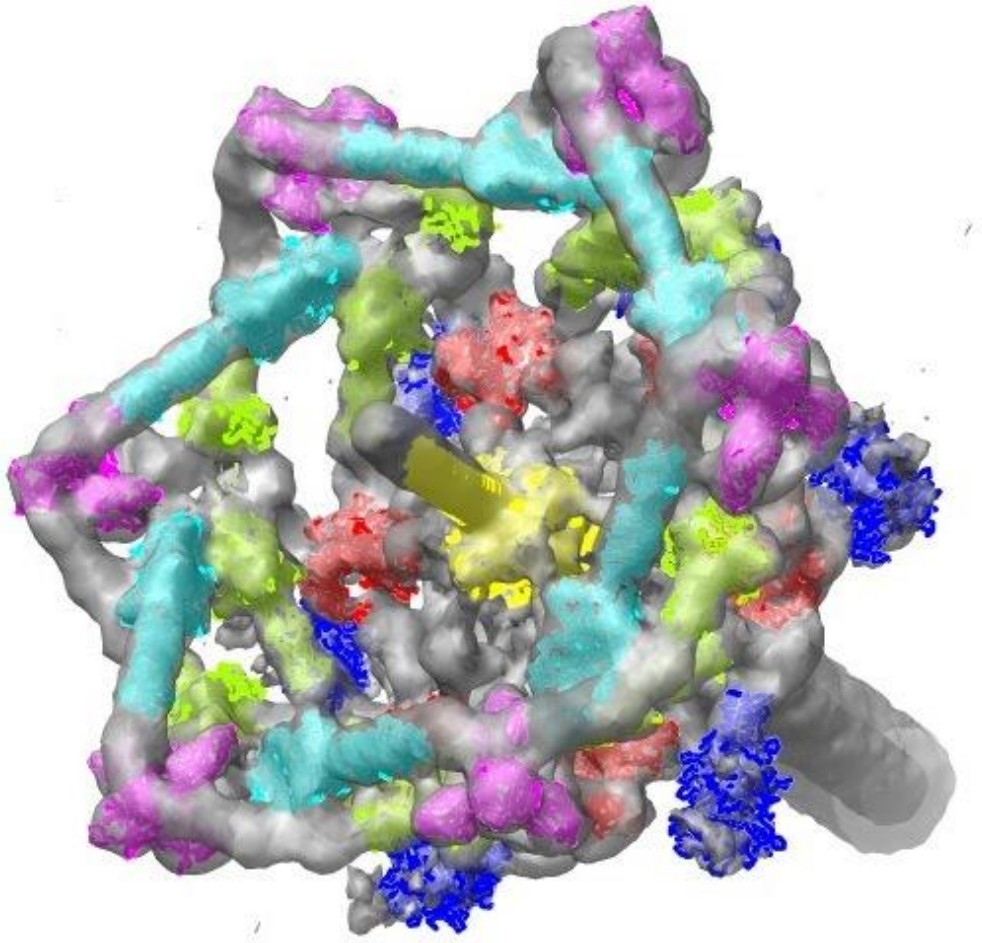
NMR structural ensemble



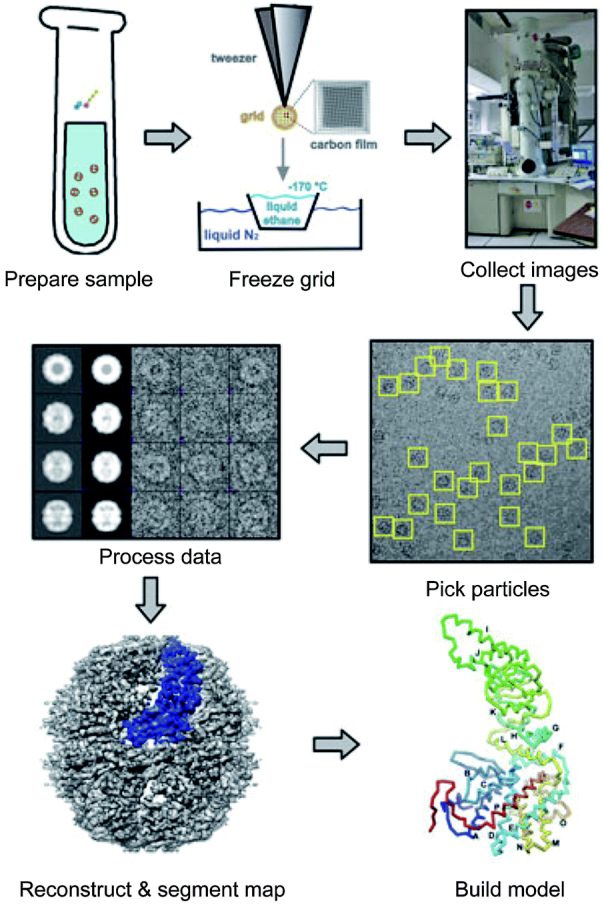
NMR and dynamics



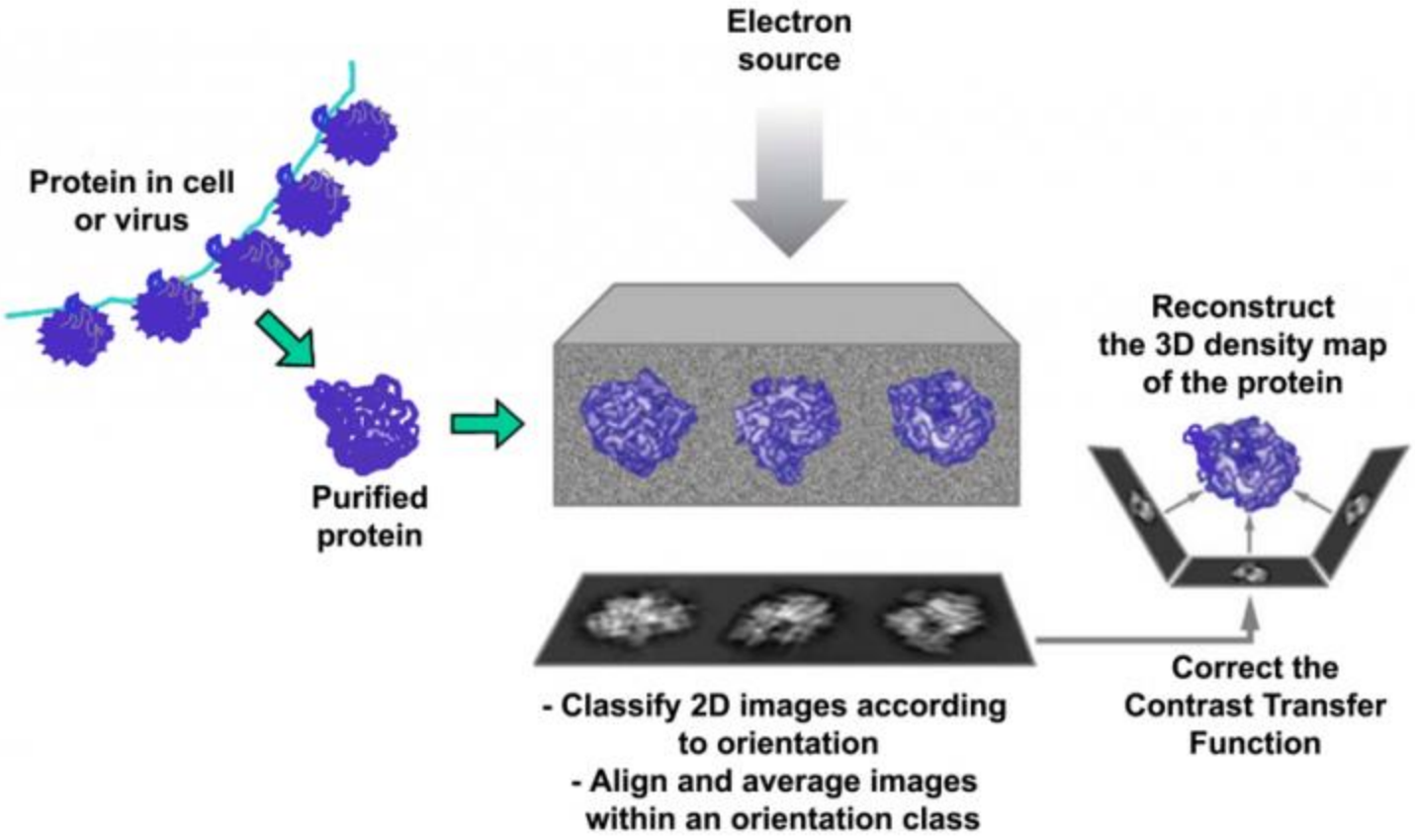
Electron Microscopy



Structure determination by cryoEM



Single particle technique

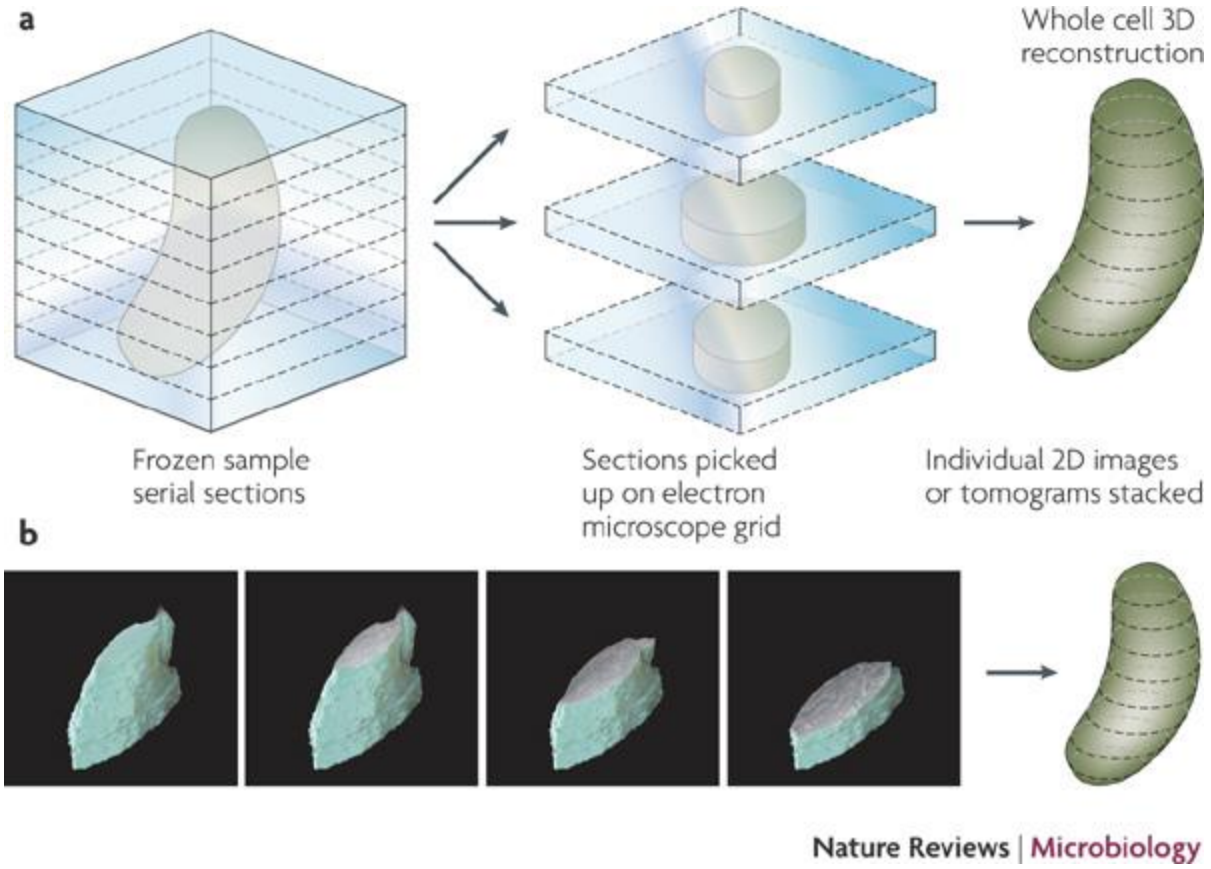


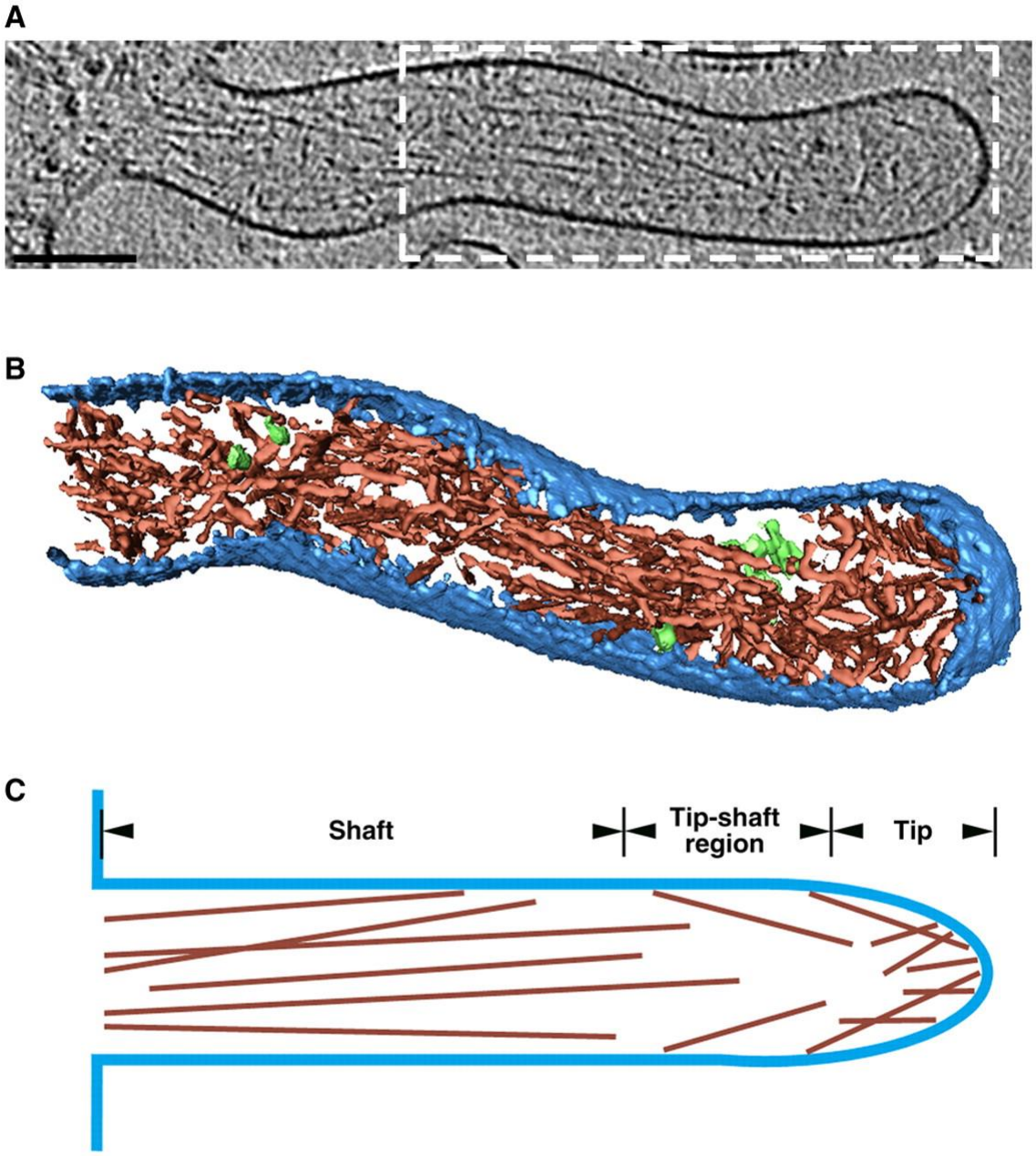
Transmission Cryo-Electron Microscopy

A tool used by structural biologists to study molecular nanomachines

Gabriel Lander, Thesis Defense 2009

Tomography technique



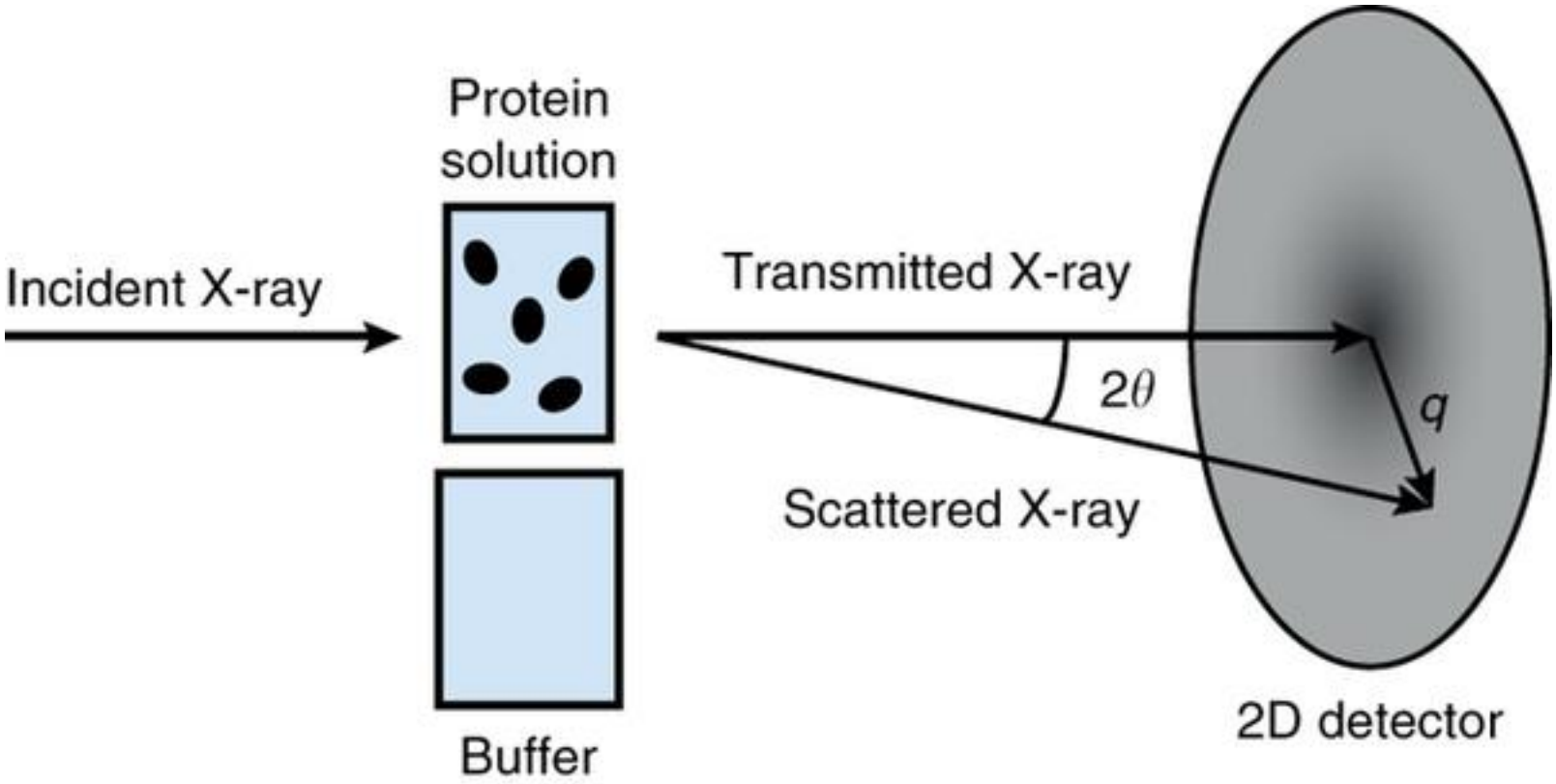


The architecture of *D. discoideum* filopodia

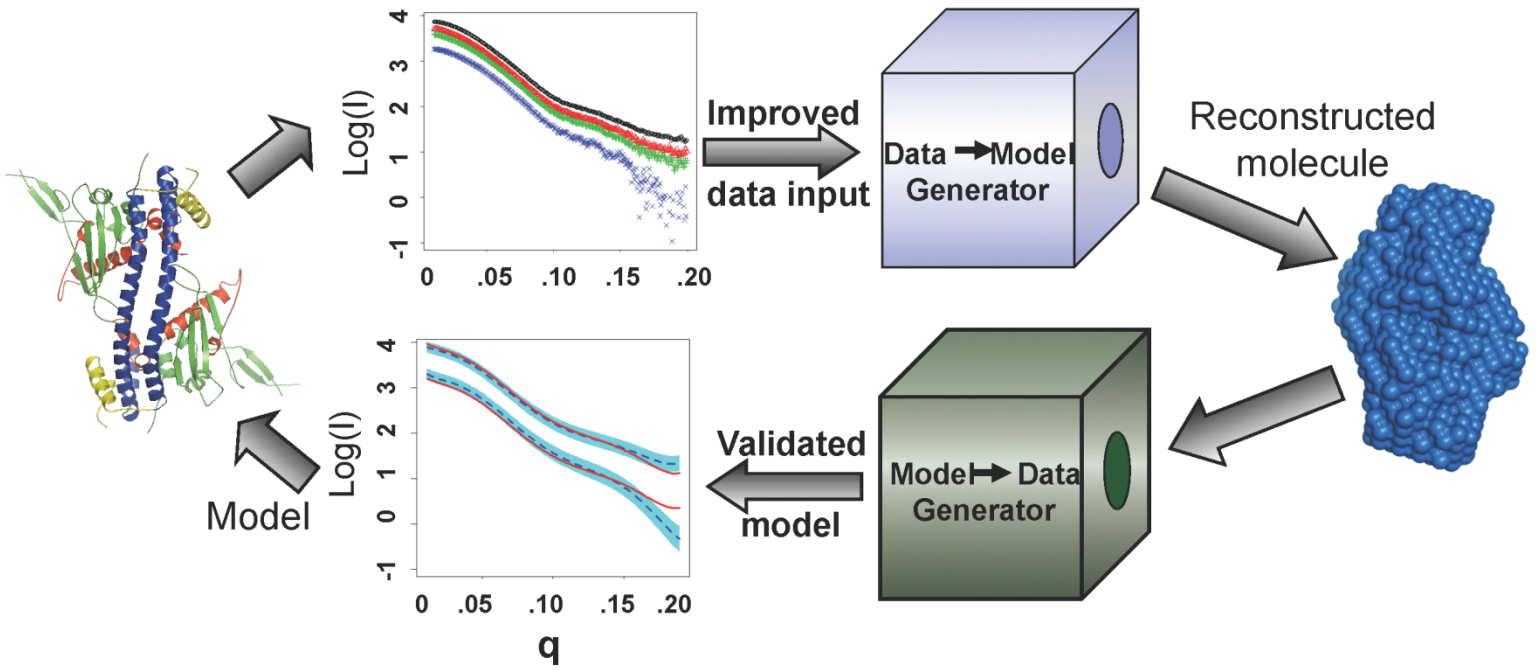
Other structure characterization methods

- X-ray scattering methods
  - Small angle X-ray scattering
- Neuron diffraction
- Optical spectroscopic methods
  - Absorbance: UV spectroscopy
  - Fluorescence: fluorescence microscopy
  - Circular dichroism
  - Vibrational spectroscopy
  - Raman spectroscopy
  - …

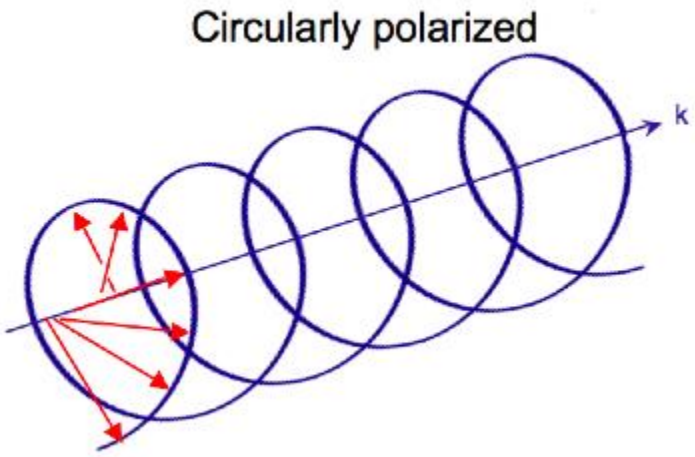
Small angle X-ray scattering (SAXS)

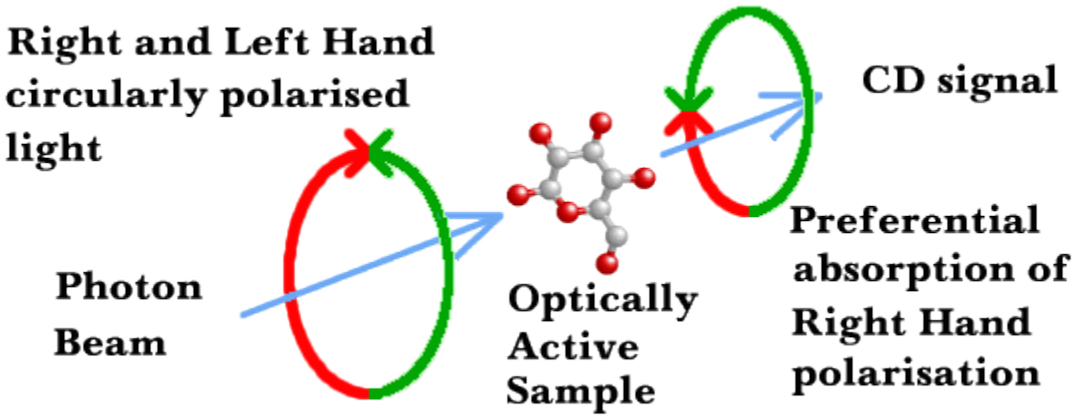


SAXS provides the shape information

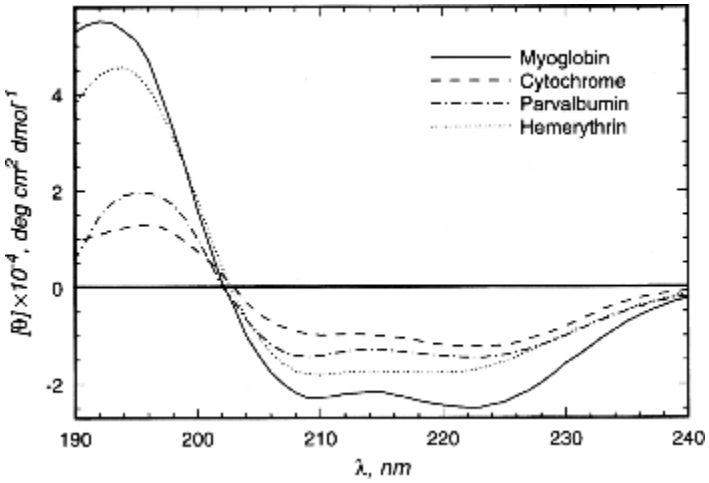


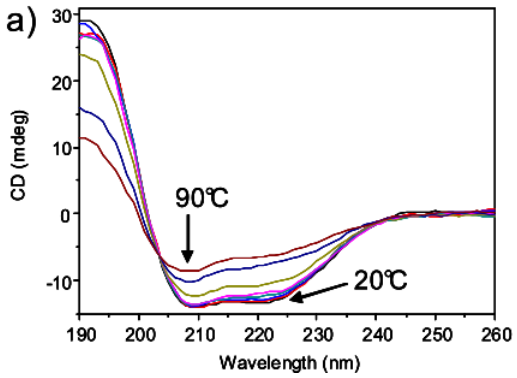
Circular dichroism (CD)

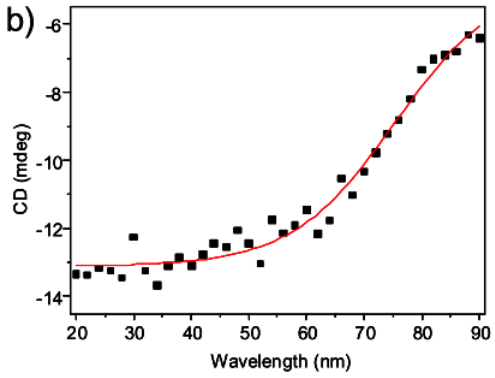




CD can be used for protein folding analysis





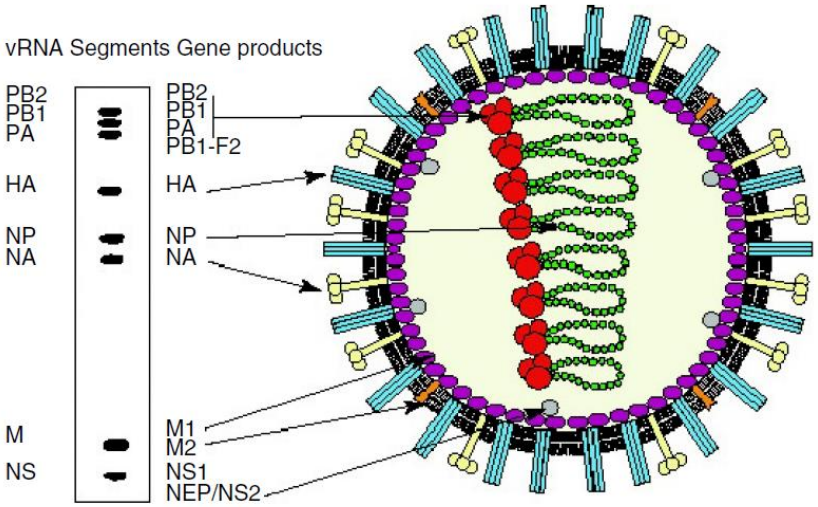


Protein structures provide molecular approach to medicine

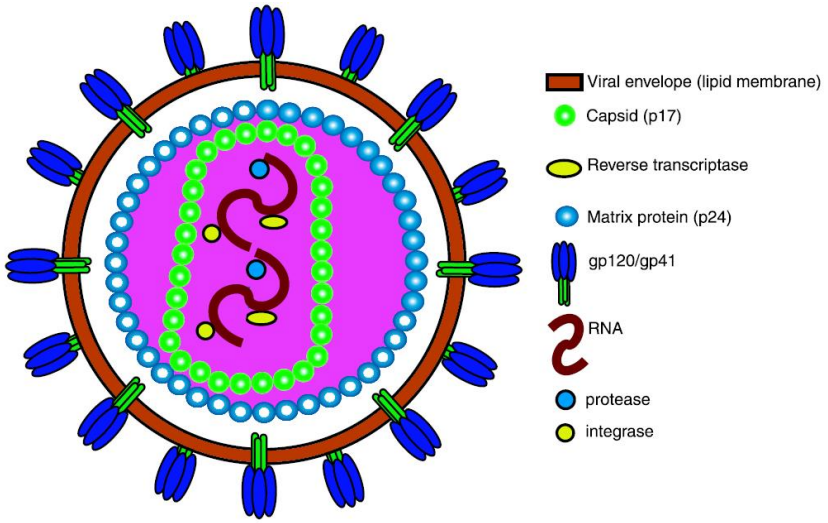
- Most diseases happen are due to defects in proteins
- Protein structure is a key to understand disease-causing mechanism
  - How disease-causing mutations affect structures and/or target recognition
- Protein structure based drug design

Virus and their impact on health as seen through structure and function

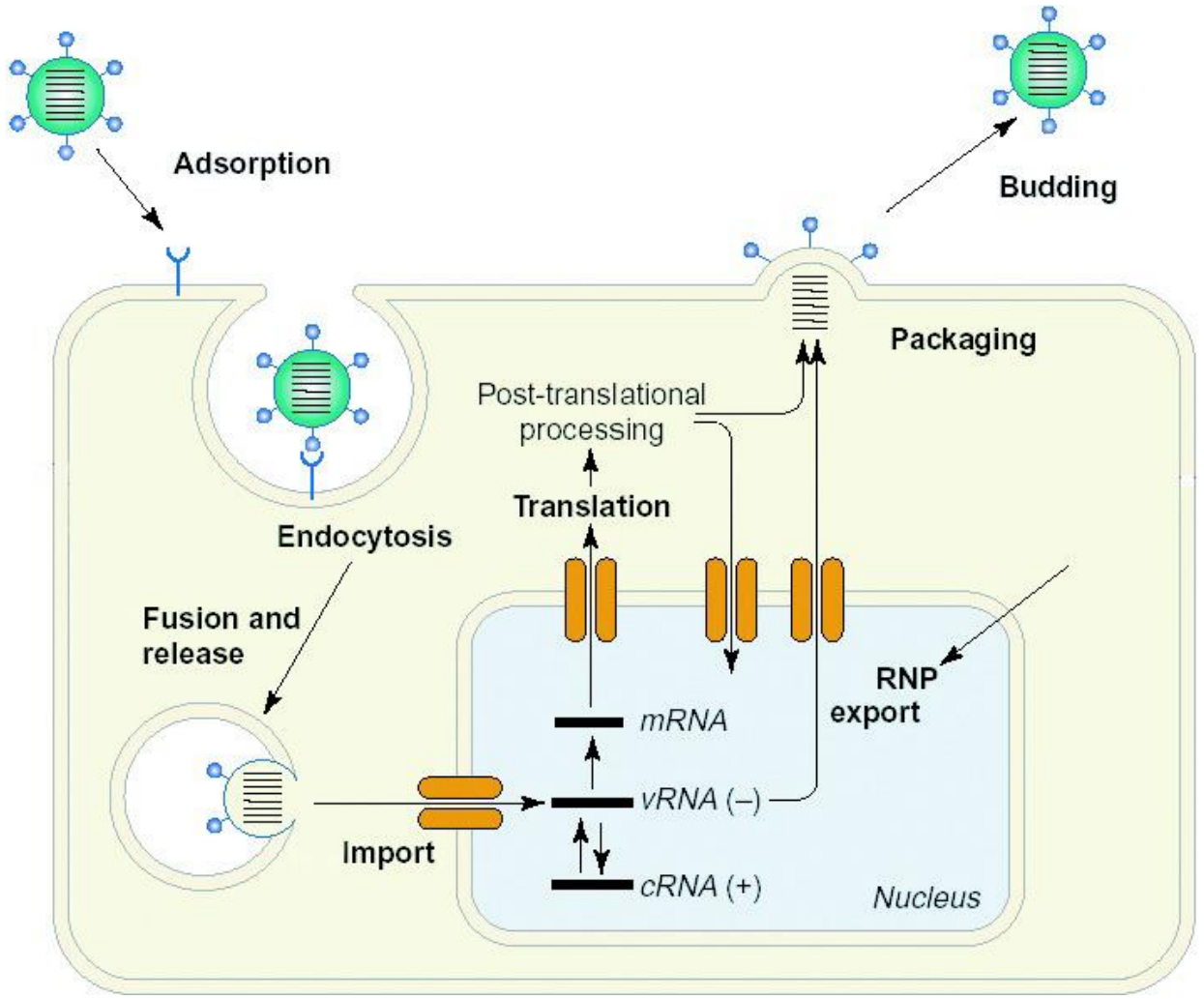
Influza

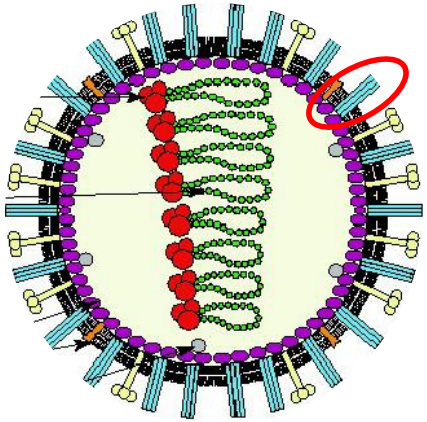


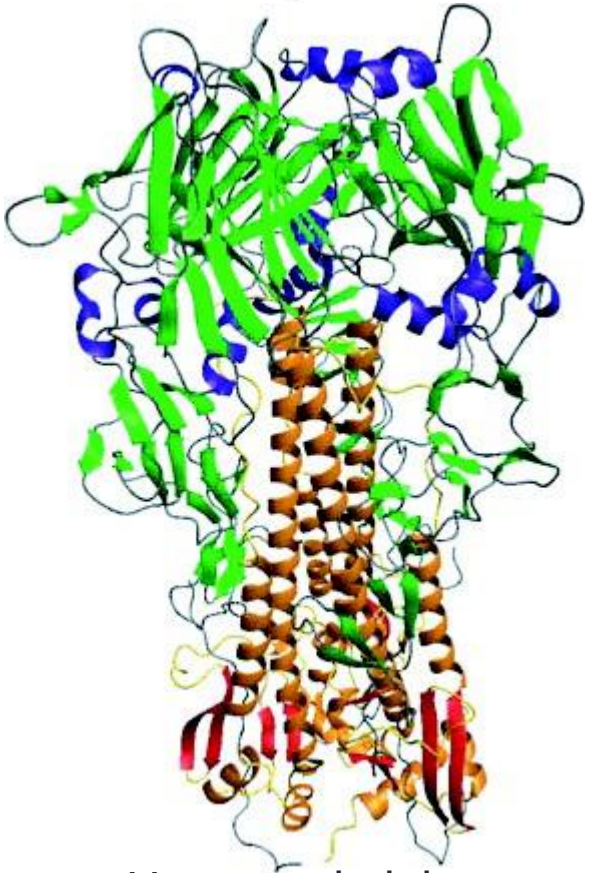
HIV



The pathway of viral entry and exit from host cells

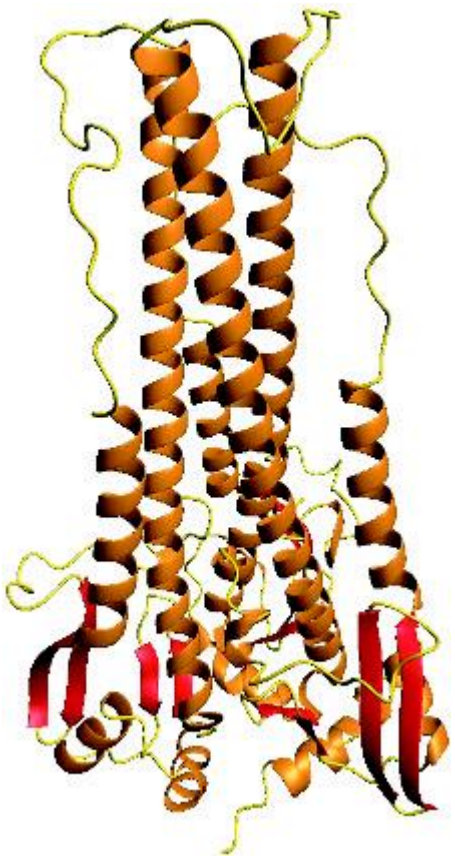


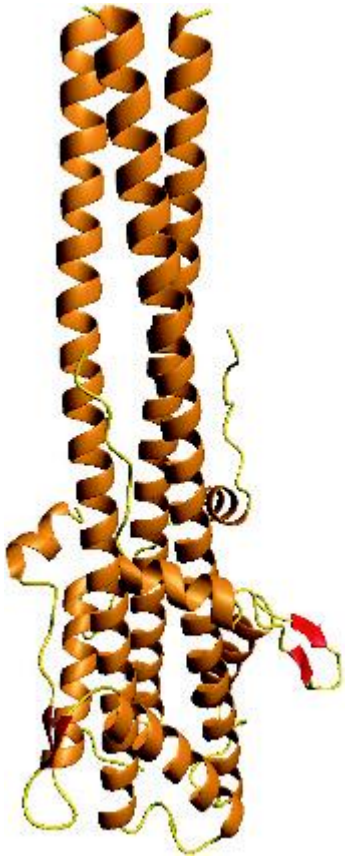




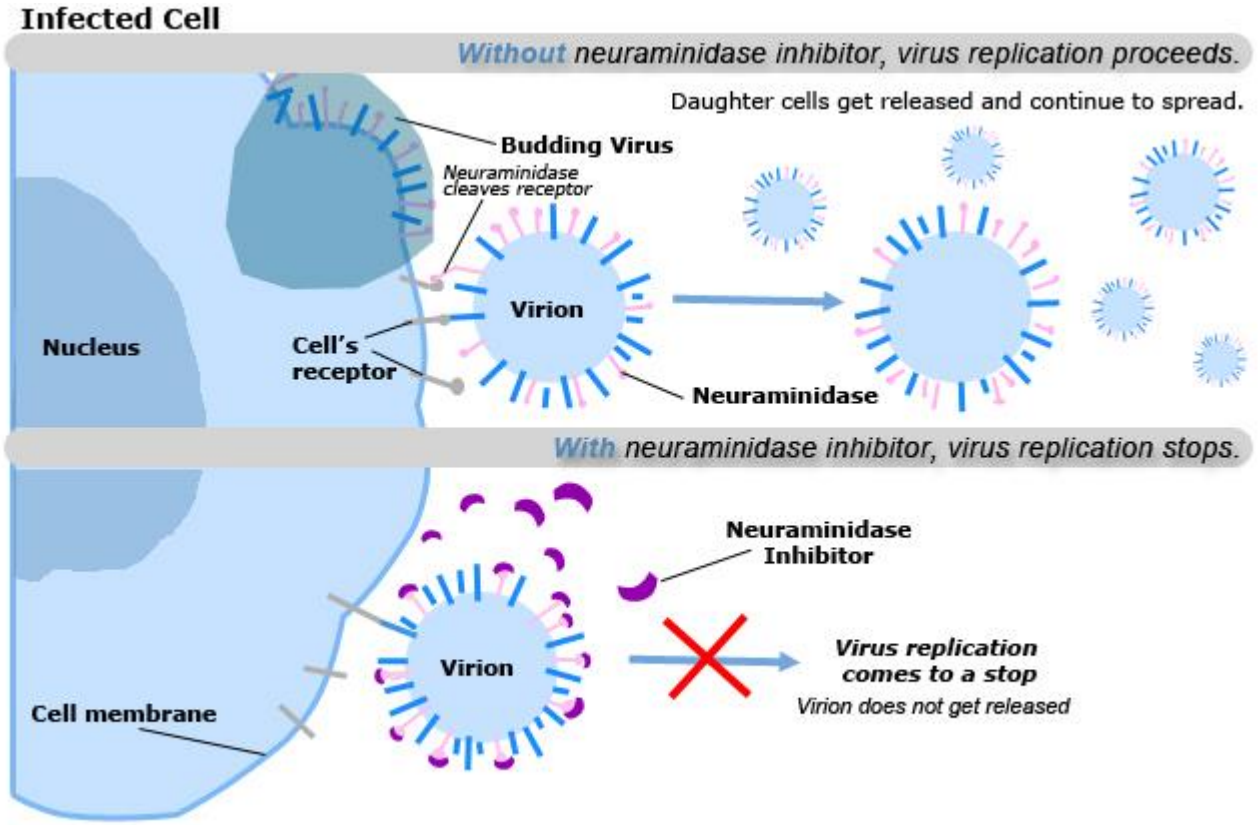
Haemagglutinin

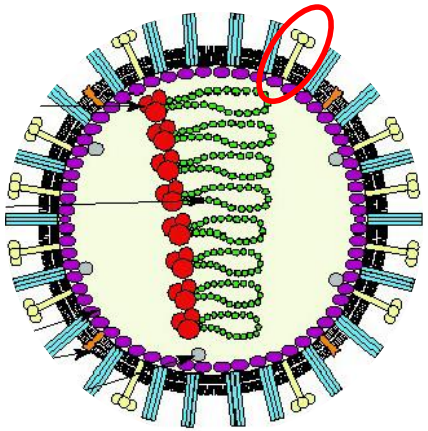
Haemagglutinin: the anchor



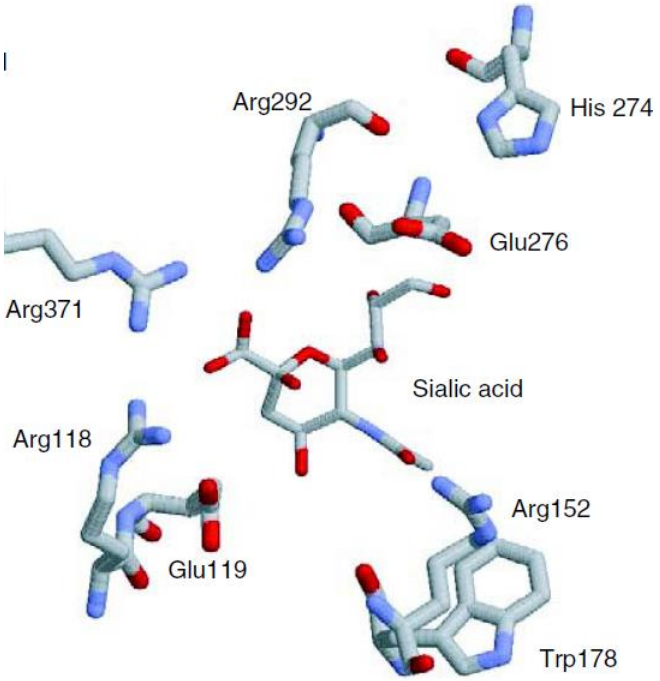


The conformational changes in the transition from neutral pH form to active haemagglutinin

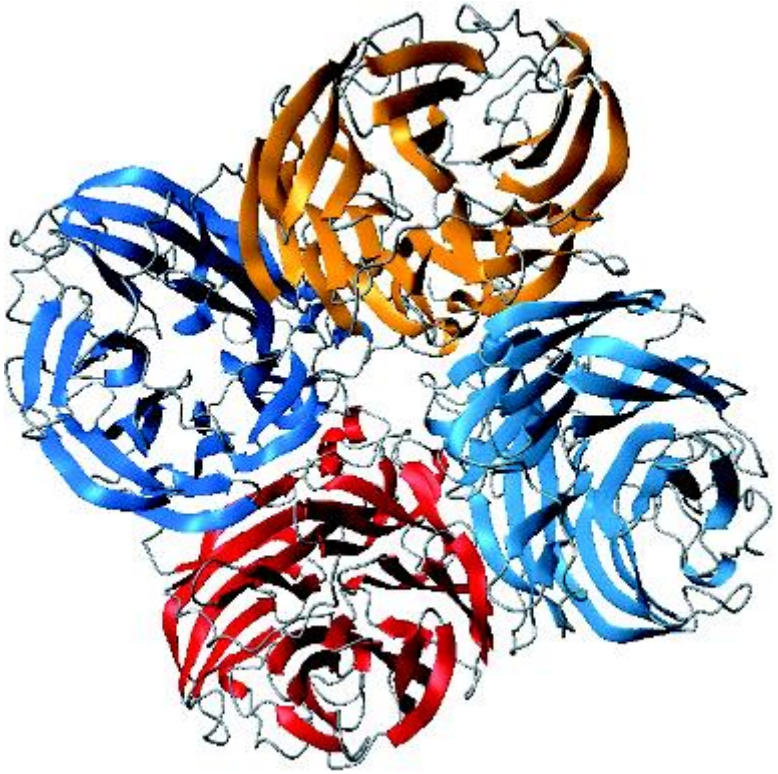

Infected Cell
Without neuraminidase inhibitor, virus replication proceeds.
Daughter cells get released and continue to spread.
Budding Virus
Neuraminidase cleaves receptor
Virion
Nucleus
Cell's receptor
Neuraminidase
With neuraminidase inhibitor, virus replication stops.
Neuraminidase Inhibitor
Virion
Virus replication comes to a stop
Virion does not get released
Cell membrane



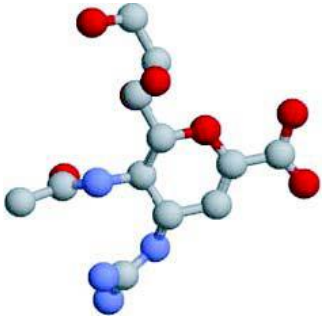
Neuraminidase: the releaser



Sialic acid (substrate) bound to active site

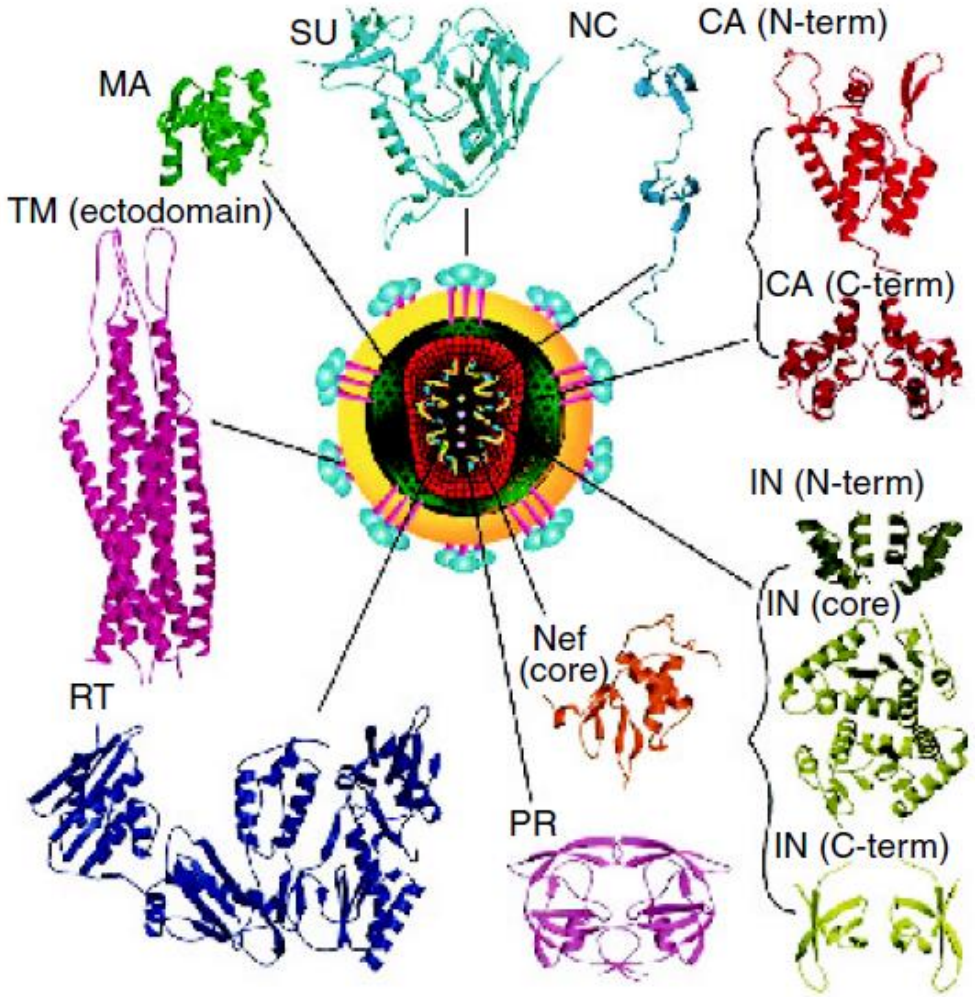


Neuraminidase

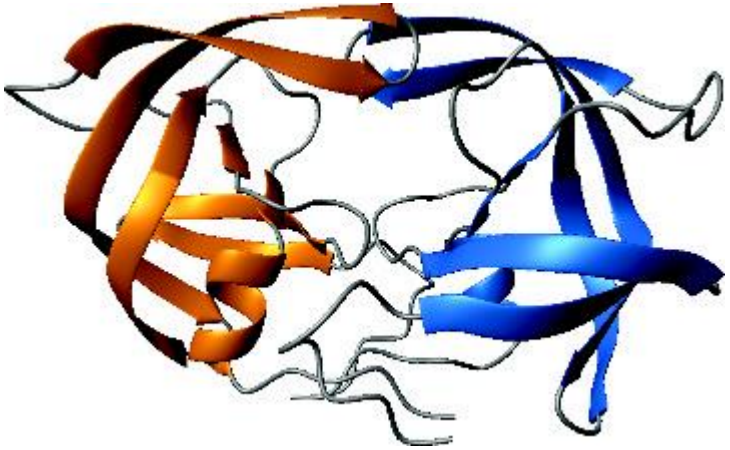


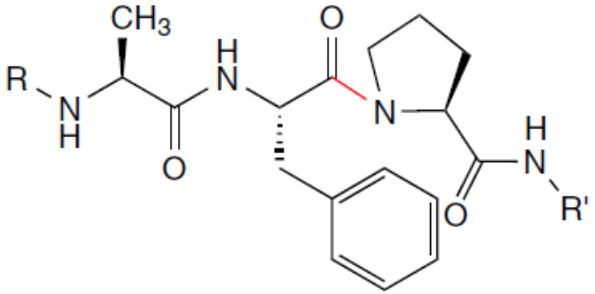
Zanamivir

The structure of HIV

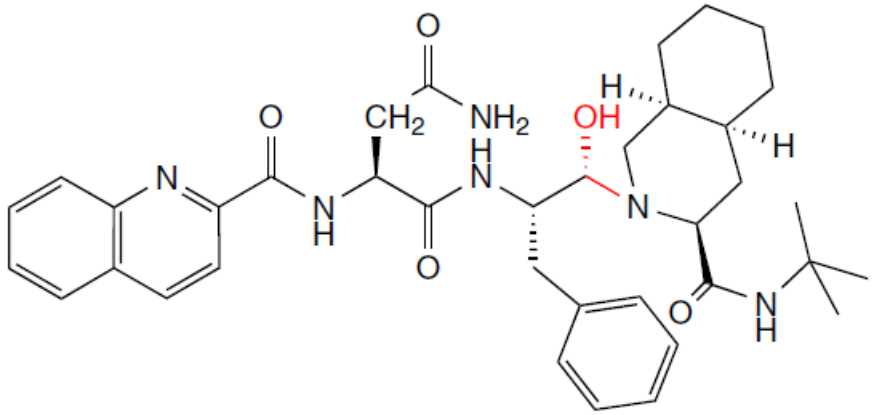


HIV protease





Ala-Phe-Pro sequence containing the target peptide bond (substrate) for HIV protease



The structure of saquinavir

Drug design strategies