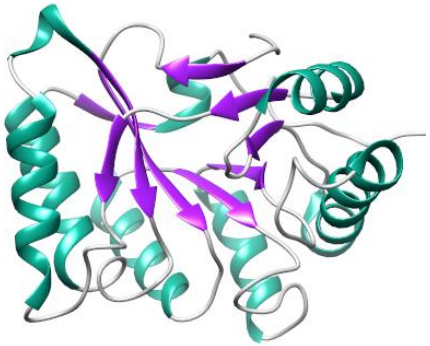
Physical contributions in protein

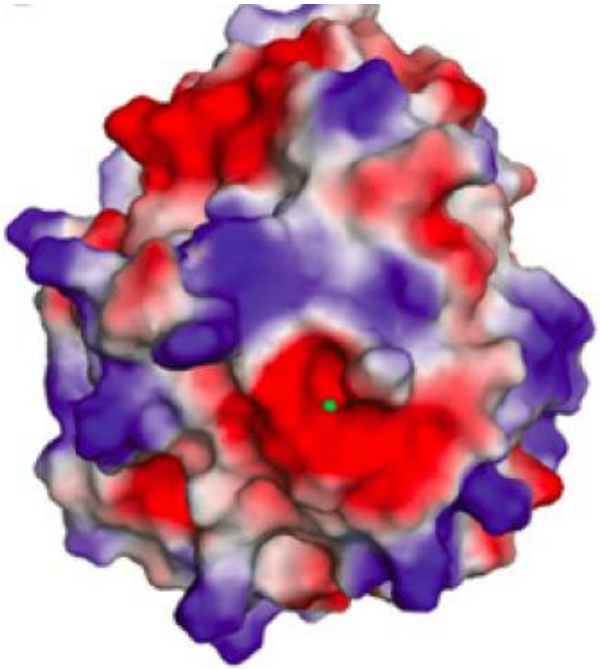
Different physical forces, which sometimes counteract and compensate one another, contribute to the stability of the native state of proteins.



-Hydrophobic effect
-Electrostatic interactions between charged and polar groups
-van der Waals interactions
-Conformational (chain) entropy

Electrostatic Potential Map

Illustrate the charge distributions on 3D structure

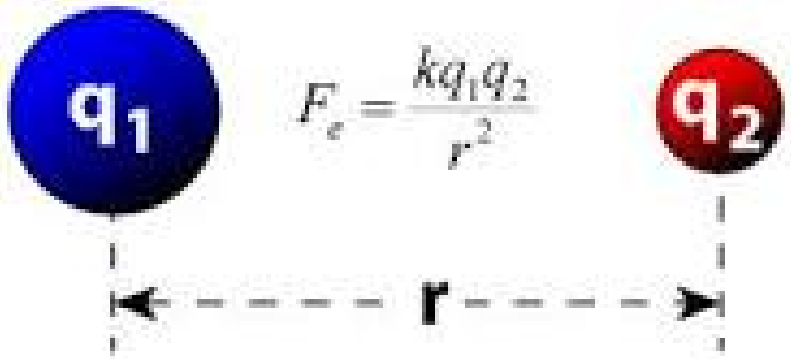


Electrostatic potential energy is fundamentally a measure of the strength of the nearby charges, nuclei and electrons, at a particular position.

Electrostatic Potential Energy

Electrostatic energy with Columb law

$$E_{ij} = \frac{q_i \, q_j}{\varepsilon \, r_{ij}}$$



The potential Φi at atom i is given by

$$\varphi_i = \frac{q_j}{\varepsilon \, r_{ij}}$$

Electrostatic Potential Map

→ Electrostatic potential maps for proteins are typically calculated by numerically solving Poisson-Boltzmann equation based on molecular mechanics-based point charges.

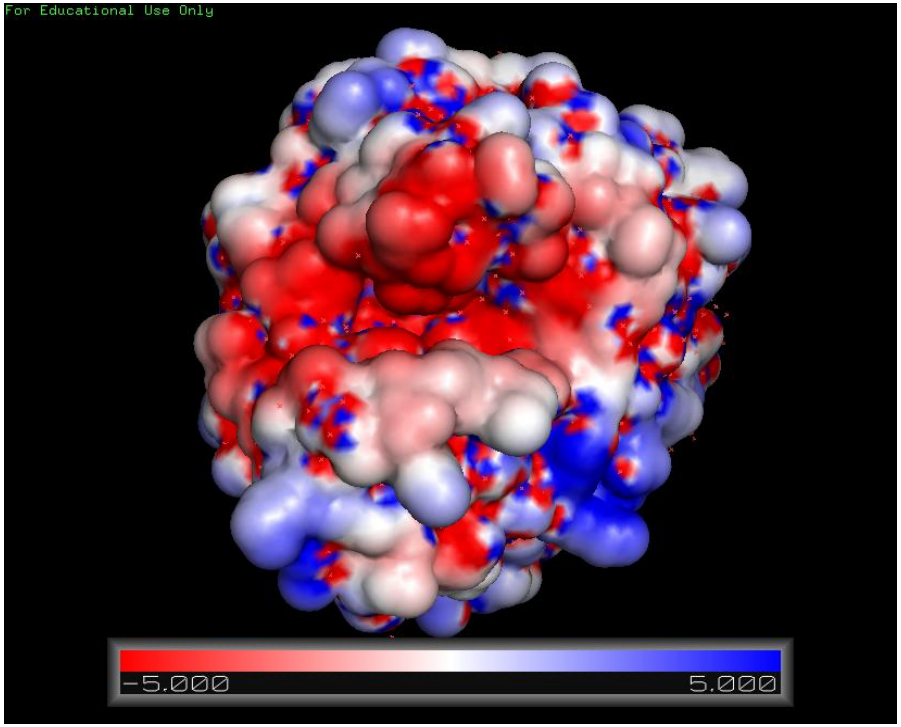
→ The charge assignment requires complete residues with hydrogen atoms and involves assessment of side-chain pKa's.

$$\nabla \varepsilon(r) \nabla f(r) = r_{macro}(r) + \sum_i q_i n_i^o \exp^{-q_i f / kT}$$

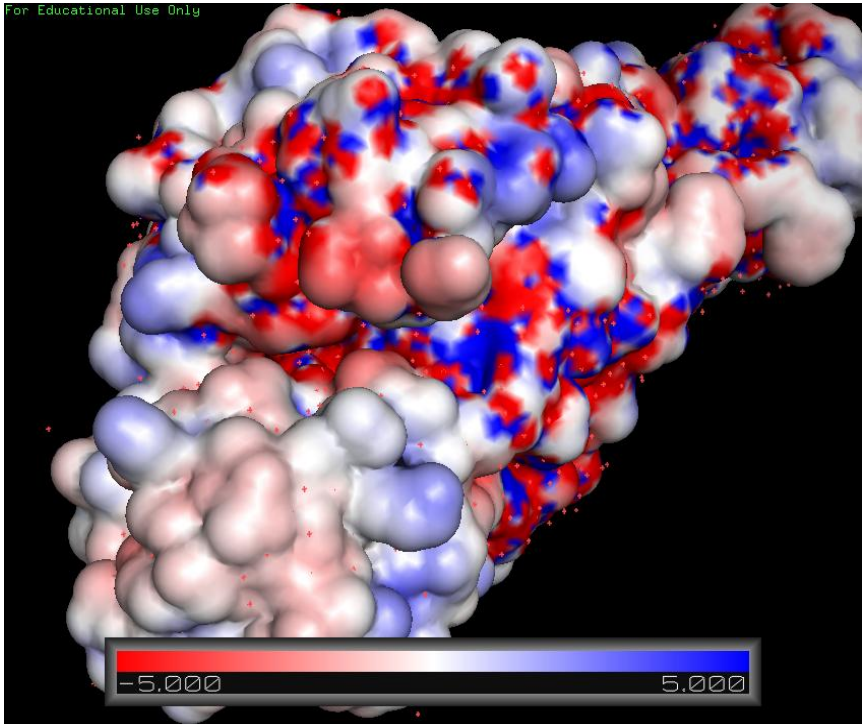
Charge distribution due to macromolecule

Implicit charge distribution due to counter ions

Electrostatic Potential Map of chitin and chitosanase

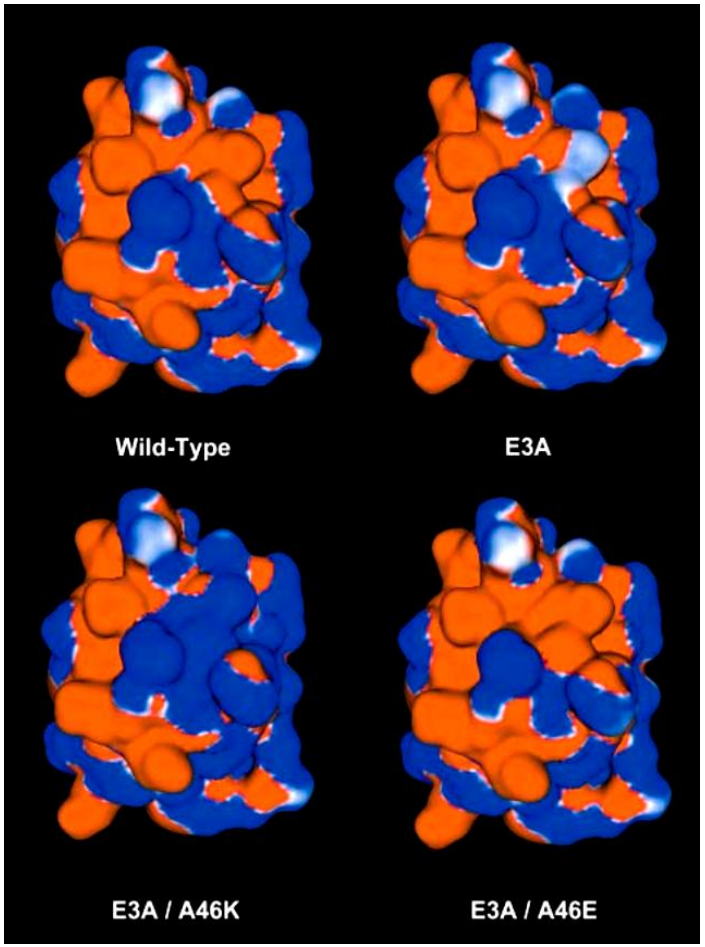


Chitosanase



Chitinase

Electrostatic potential maps can aid in understanding of relative stabilities



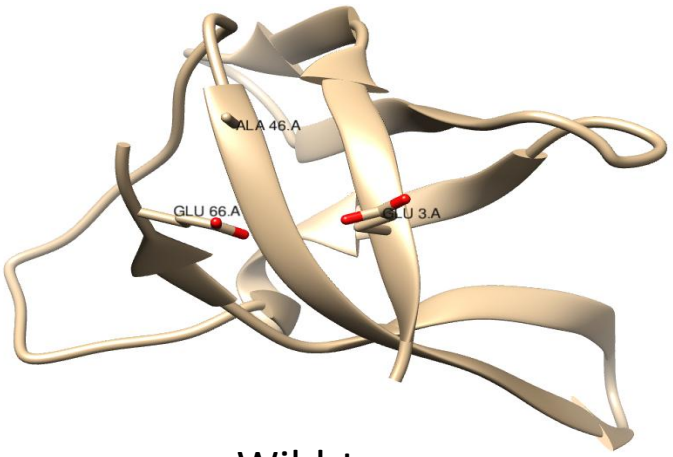
Electrostatic potentials is helpful in investigating the relative stability of mutant

The E3A/A46E mutant is destabilized

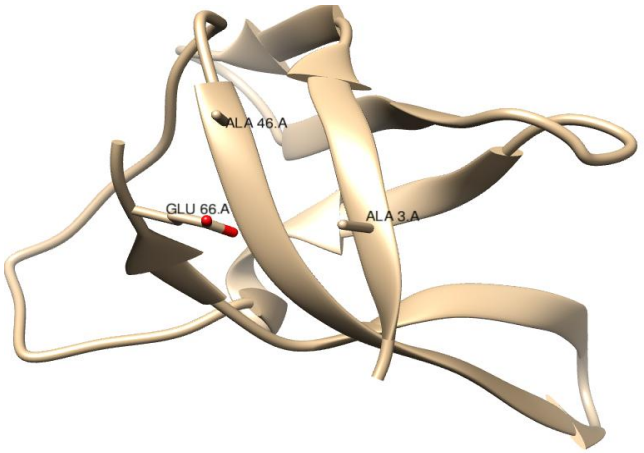
The E3A/A46K mutant is one of the most stable CSP mutants investigated.

REF: Torrez et al., Biophys J (2003) **85**, 2845-2853

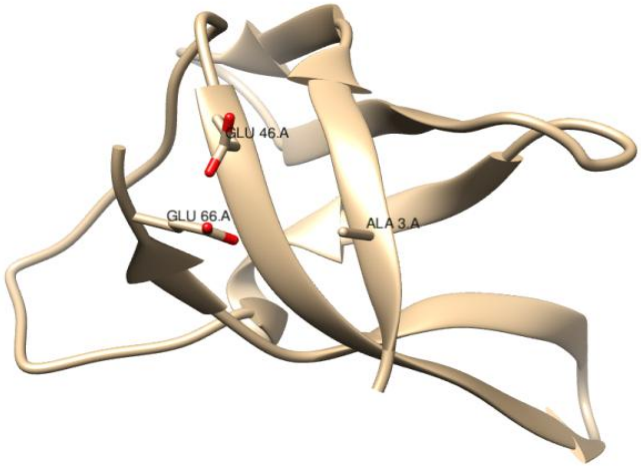
Relative stability of mutants



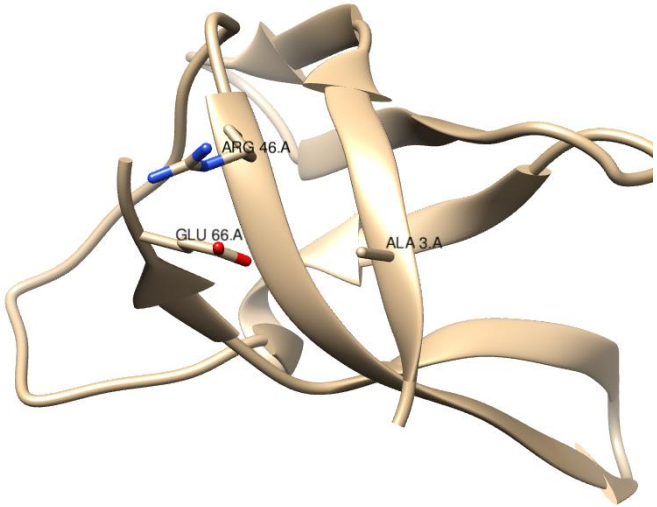
Wild-type



The E3A mutant is stabilized versus the wild-type



The E3A/A46E mutant is destabilized



E3A/A46K mutant is one of the most stable

Electrostatic Potential Map

→ modified version of your pdb is required with the .PQR extension.

→ Pdb2pqr software or server were designed to convert PDB-format structural information into PQR-format parameterized les.

→ PQR is a popular and compact way to include atomic parameters in a PDB-like format by replacing the occupancy column of a PDB

i)(P) with the atomic charge (Q) and the

ii) column with the radius (R)

Electrostatic Potential Map

Preparation PQR

Program that allows to convert pdb les to pqr format:

http://www.poissonboltzmann.org/pdb2pqr/

Before to run pdb2pqr with your pdb you need to make sure that:

1. Your pdb should not contain any residues in several conformers (mainly structure at high resolution)

2. Your pdb should contain complete residues information (missing lateral chain
have to be incorporated)

3. Bfactor of your pdb need to be <100

4. Remove non bonded atoms

Electrostatic Potential Map

→ APBS, the Adaptive Poisson-Boltzmann Solver, is a freely available macromolecular electrostatics calculation program.

→ PyMol currently supports the APBS plugin

→ CHARMM-GUI

→ Pymol can display the results of the calculations as an electrostatic potential molecular surface

Excercise

Generate electrostatic map on chitinase and chitosanase structure using PDB2PQR and PYMOL