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Combining water analysis with protein electrostatics

Tim Cheeseright

The Hard Work

Susana Tomasio

Mark Mackey

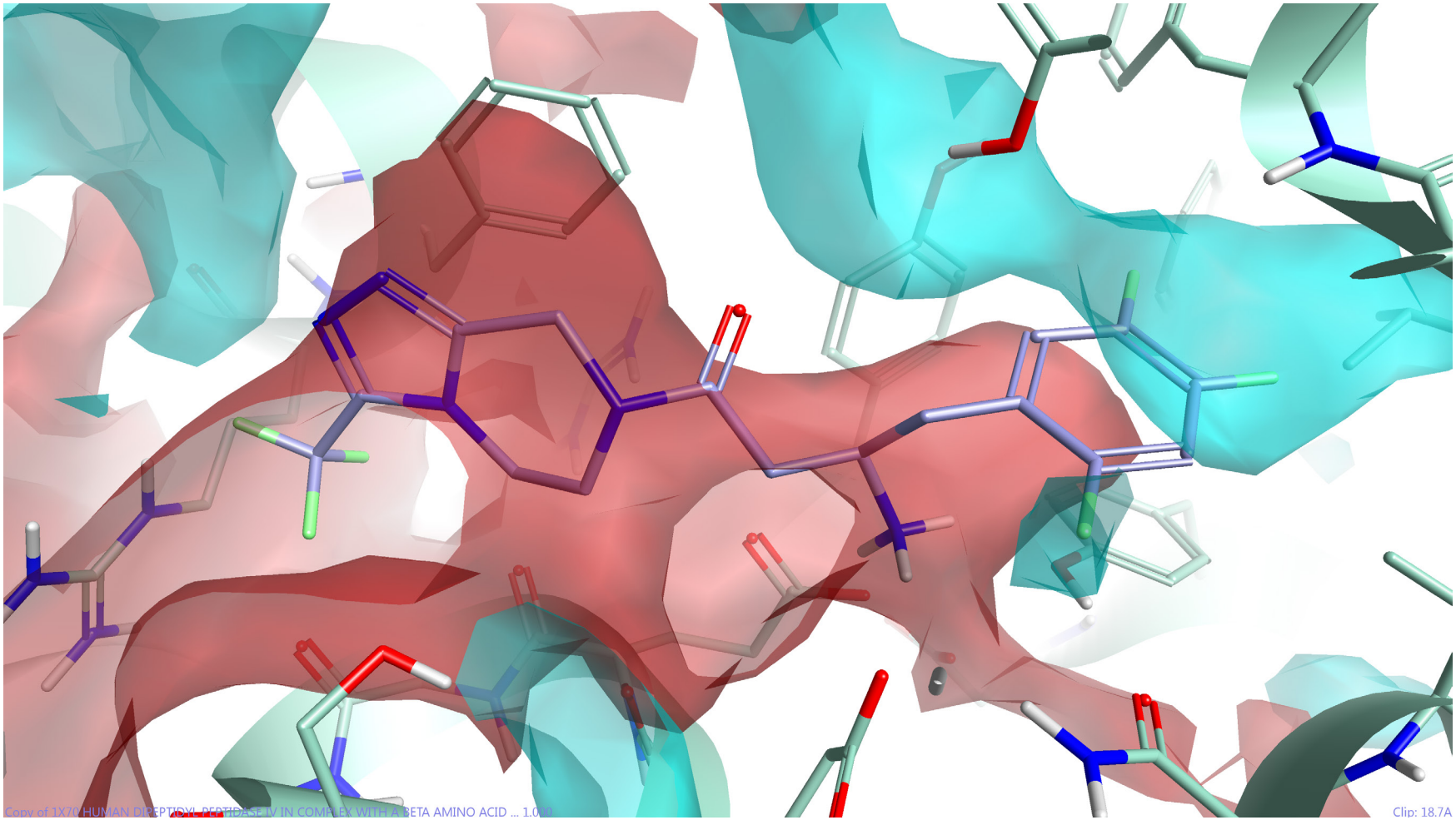
Paolo Tosco

Protein Interaction Potentials

Protein Electrostatics

- Cresset's electrostatic model is more detailed than other MM based approaches
- Understanding the subtleties of the protein active sites should help inform ligand design
- Similar to GRID and others but with Cresset's electrostatics
  - Flood active site with ligand atoms
  - Measure interaction potential at each point
  - Contour potentials as a surface

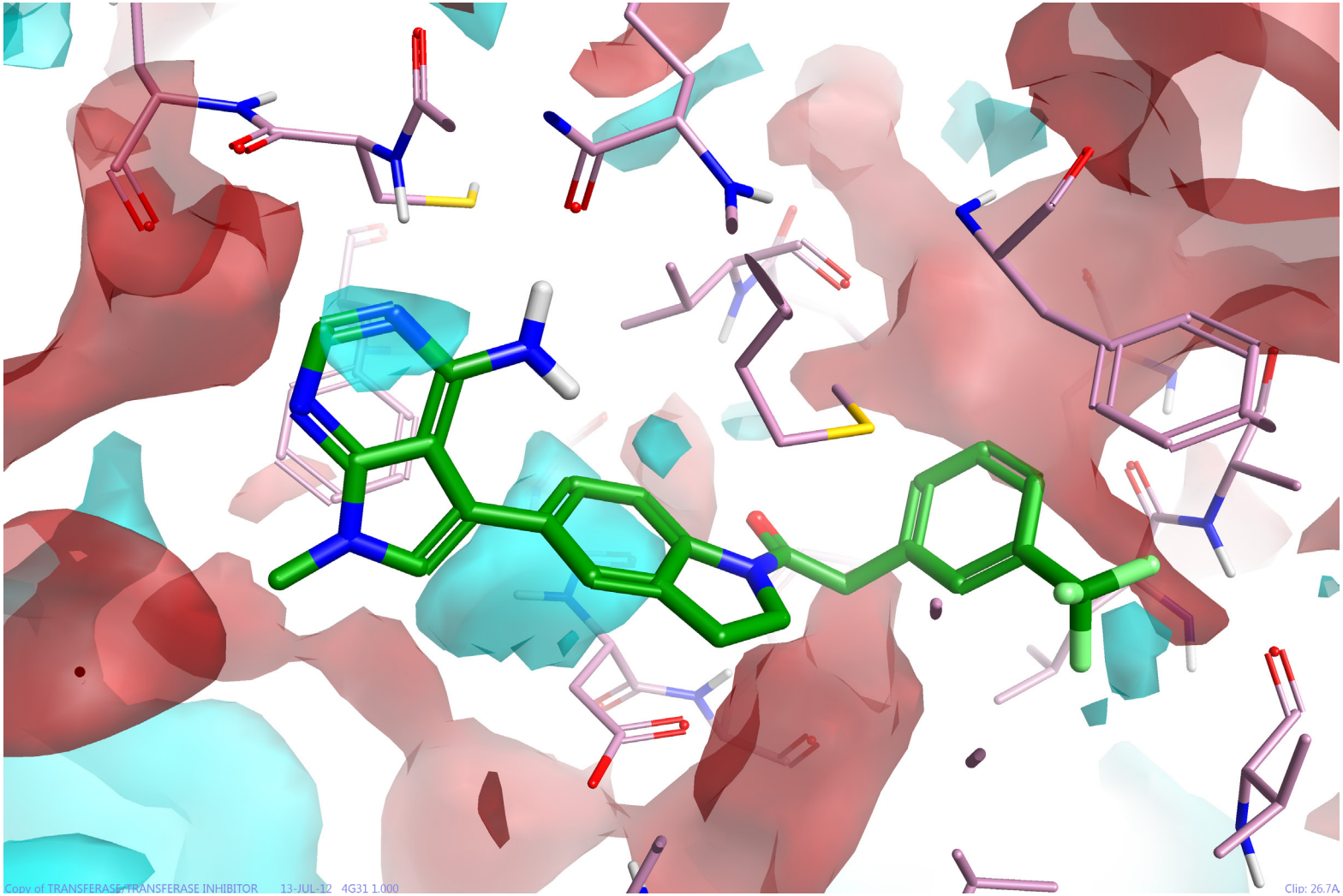
Example – DPP-IV from 1X70



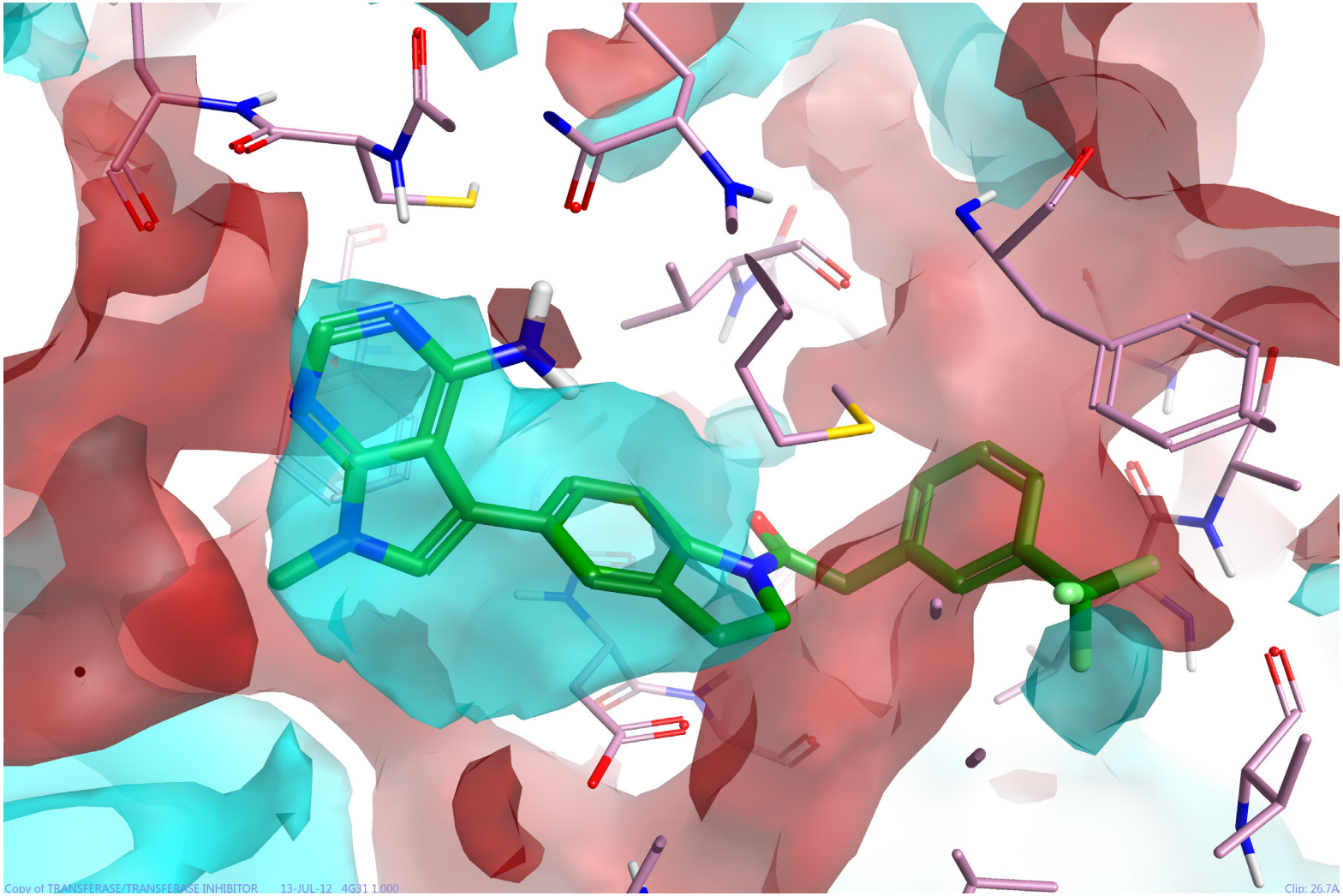
Ligand → Protein Notes

- To calculate the protein interaction potential requires a well prepared protein
- Requires a different approach to electrostatics as protein has large number of charged groups
  - → Uses a Distance Dependent Dielectric
- Calculations take 1-2 minutes
  - Frustratingly slow in practice
- Protein interaction potentials show what type of ligand atom is desired, not what type of ligand field
- Colored for ligand interpretation
  - Red → Positive ligand required
  - Blue → Negative ligand required

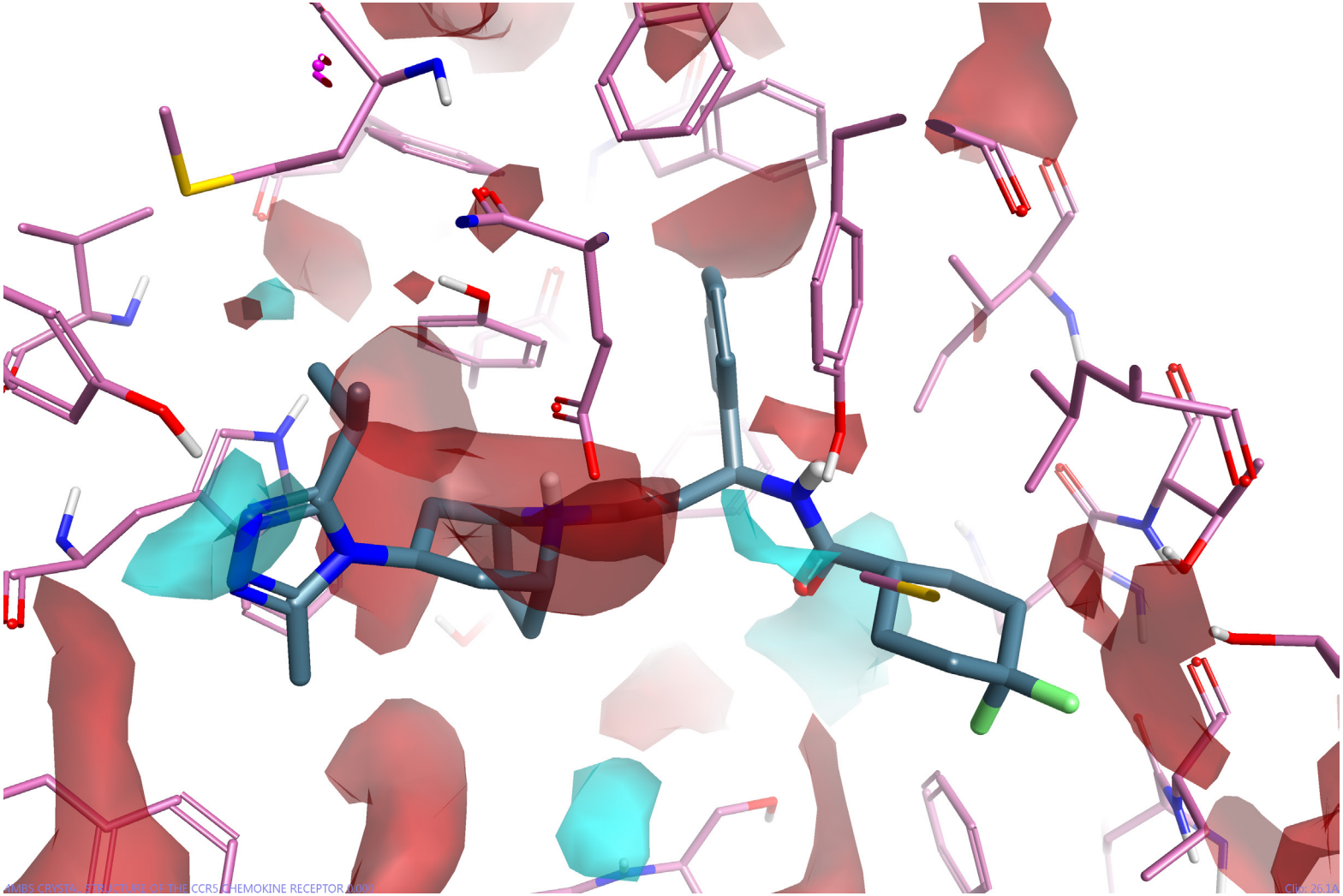
Example – PERK from 4G31



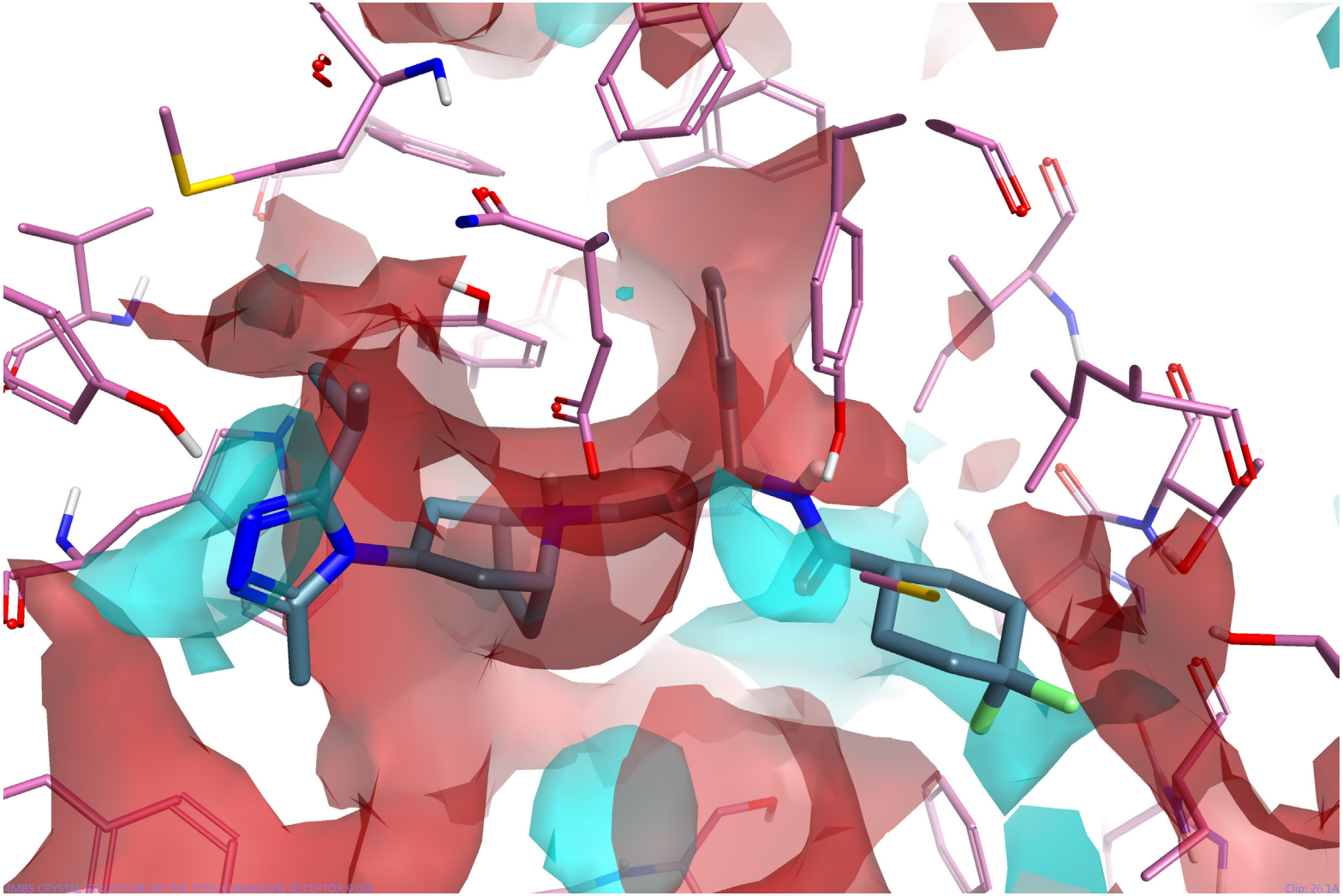
Example – PERK from 4G31



Example – CCR5 from 4MBS



Example – CCR5 from 4MBS



Protein Interaction Potentials Conclusions

- Looking at protein interaction potentials gives valuable insights into the electrostatics of the protein
- Useful in molecule design
- Still some research to identify useful contour levels

- Requires well prepared protein

- Scheduled for release in 2016 as part of a new application

3D-RISM

Better water positions through improved electrostatics?

3D-RISM

- Analytical method for working out where water goes (Ornstein-Zernike equation)
- Conceptually equivalent to running an infinite-time MD simulation on the solvent and extracting the solvent particle densities

$$h(r_{12}, \omega_1, \omega_2) = c(r_{12}, \omega_1, \omega_2) + \rho \int dr_3 d\omega_3 c(r_{13}, \omega_1, \omega_3) h(r_{32}, \omega_3, \omega_2)$$

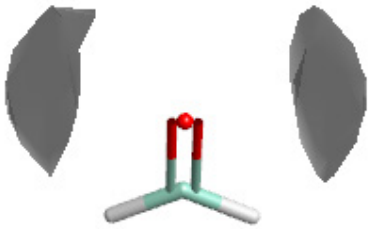
3D-RISM

- Analytical method for working out where water goes (Ornstein-Zernike equation)
- Conceptually equivalent to running an infinite-time MD simulation on the solvent and extracting the solvent particle densities
- Horribly complicated maths
- GPL implementation in Amber Tools
- Output is grid containing particle densities
- Thermodynamic analysis to assign ‘happiness’ to each water

Problems

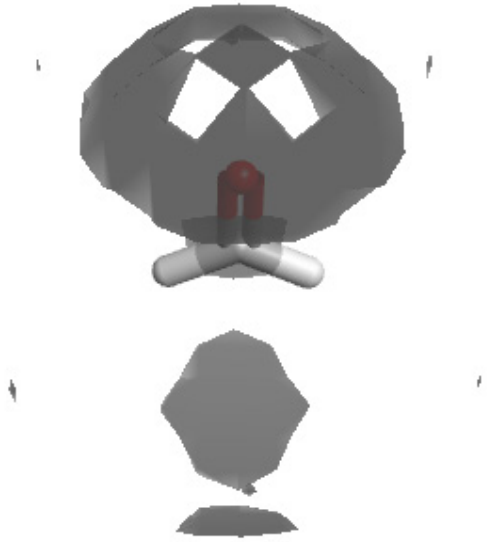
- Results depend on the potential function from solvent to solute $u(r_{12}, \Omega_1, \Omega_2)$
- In practise, this means vdW + electrostatics
- Results only as good as your potential functions
- Does the XED description of electrostatics improve the results?

Comparing XED with GAFF – Hydrogen Density



formaldehyde_x:1

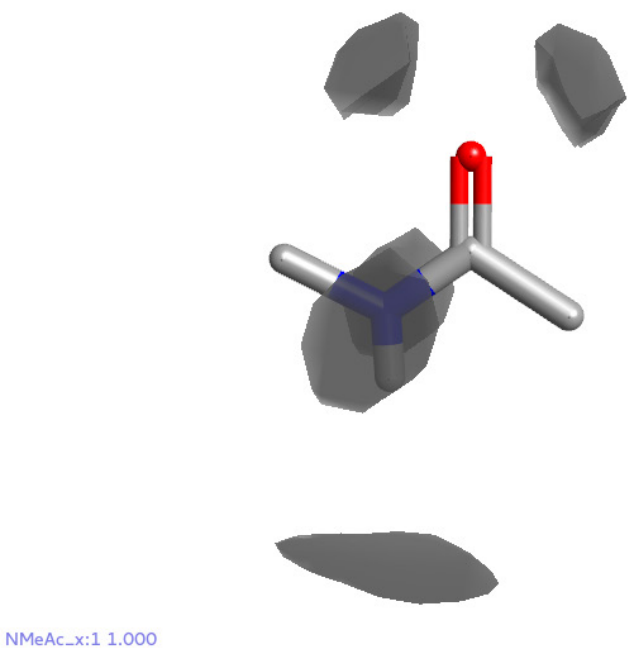
XED



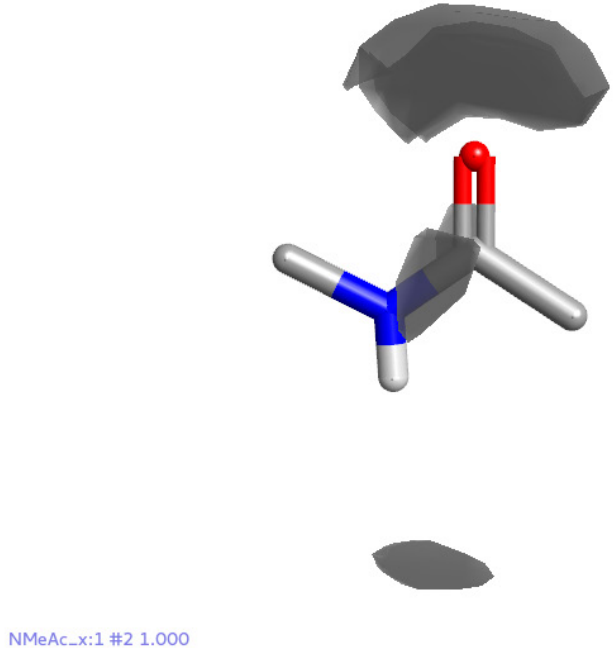
MOL 1.000

GAFF

Comparing XED with GAFF – Hydrogen Density

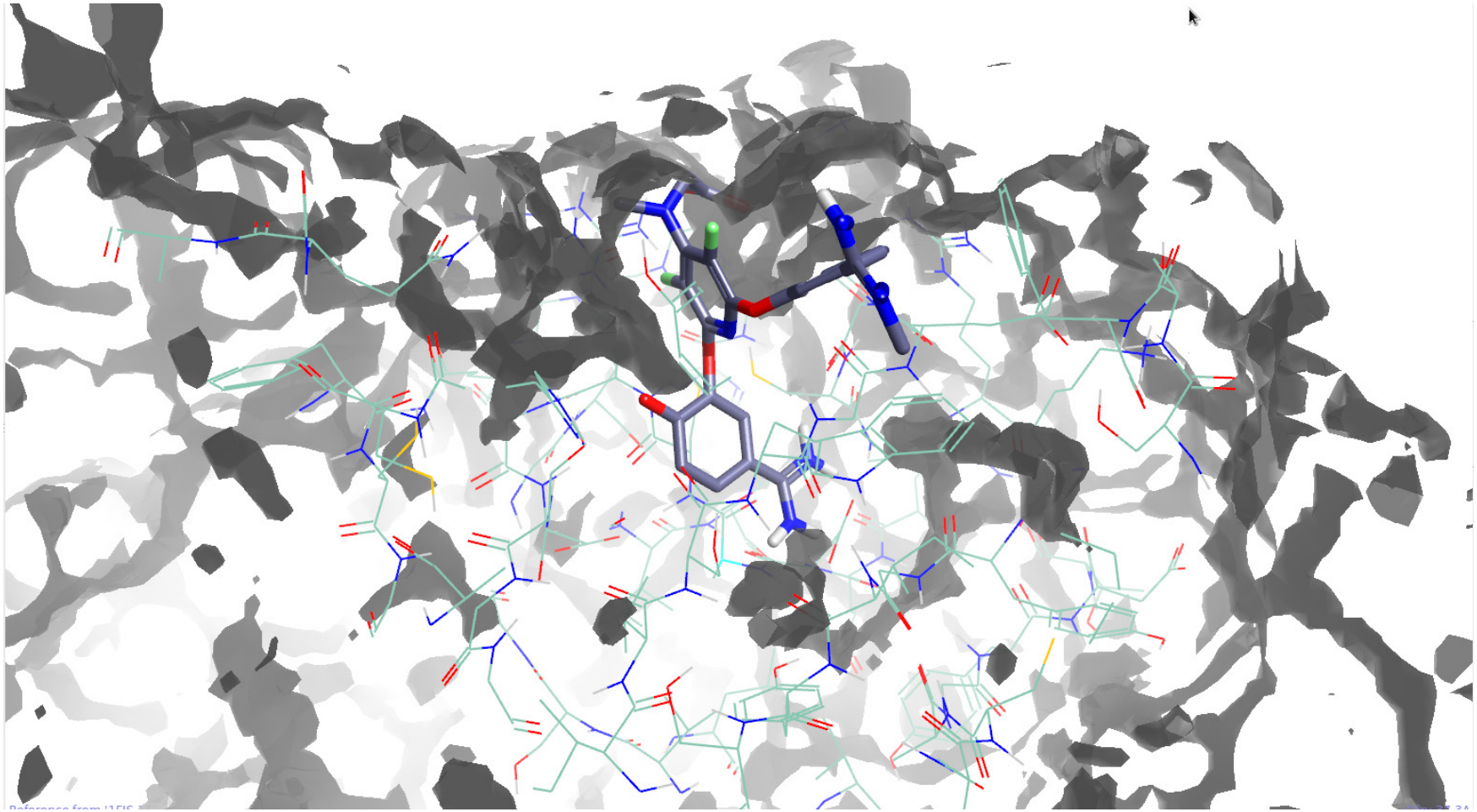


XED



GAFF

Extend to proteins – 1FJS



Assign water positions and delta-G

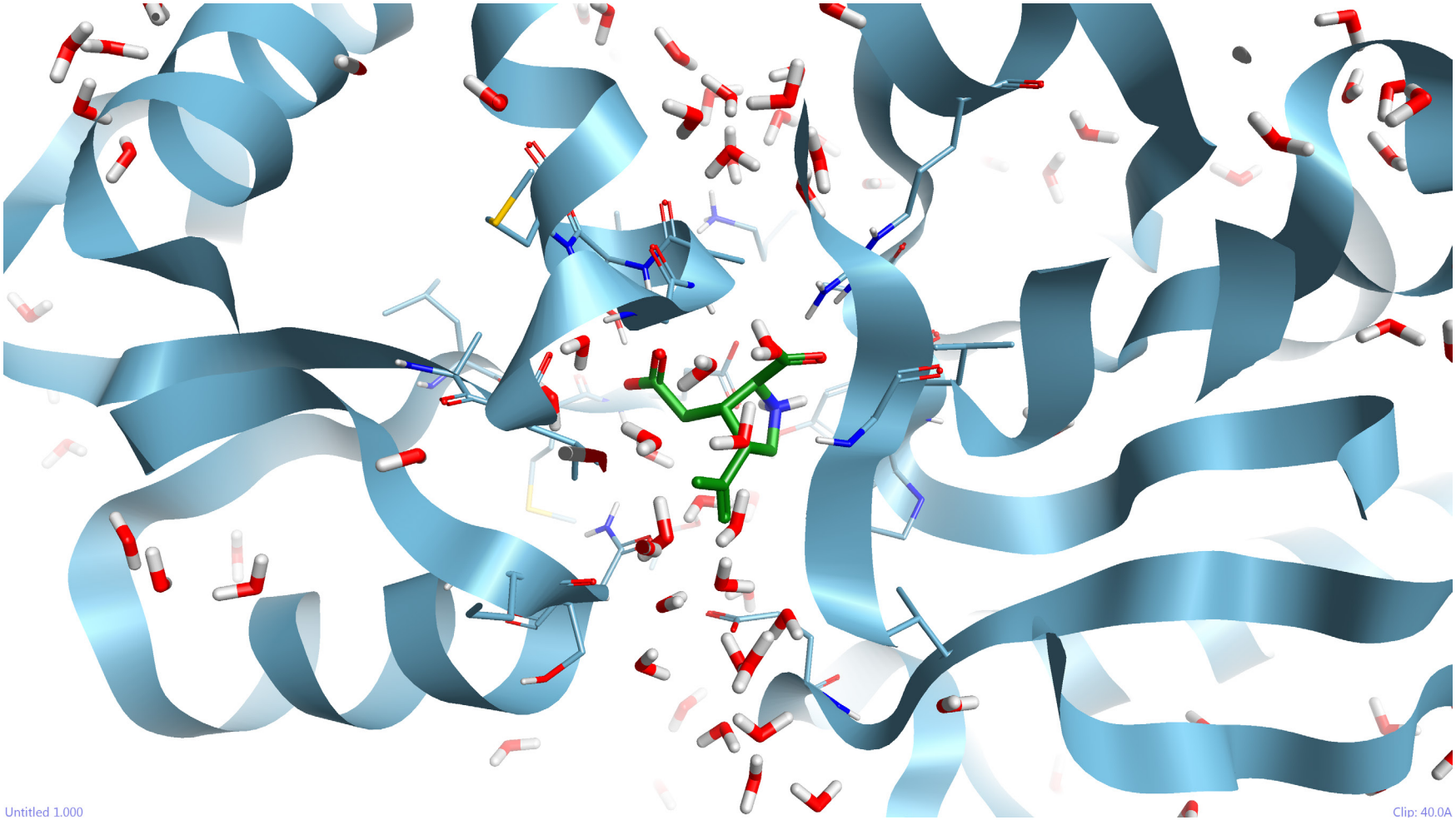
> From a 3D-RISM calculation we obtain thermodynamic data: solute internal energy, solute solvation free energy, average solute-solvent interaction energy, density, direct correlation and total correlation.

> This enables us to compute the position of the water molecules corresponding to the regions of high water density and the corresponding ΔG.

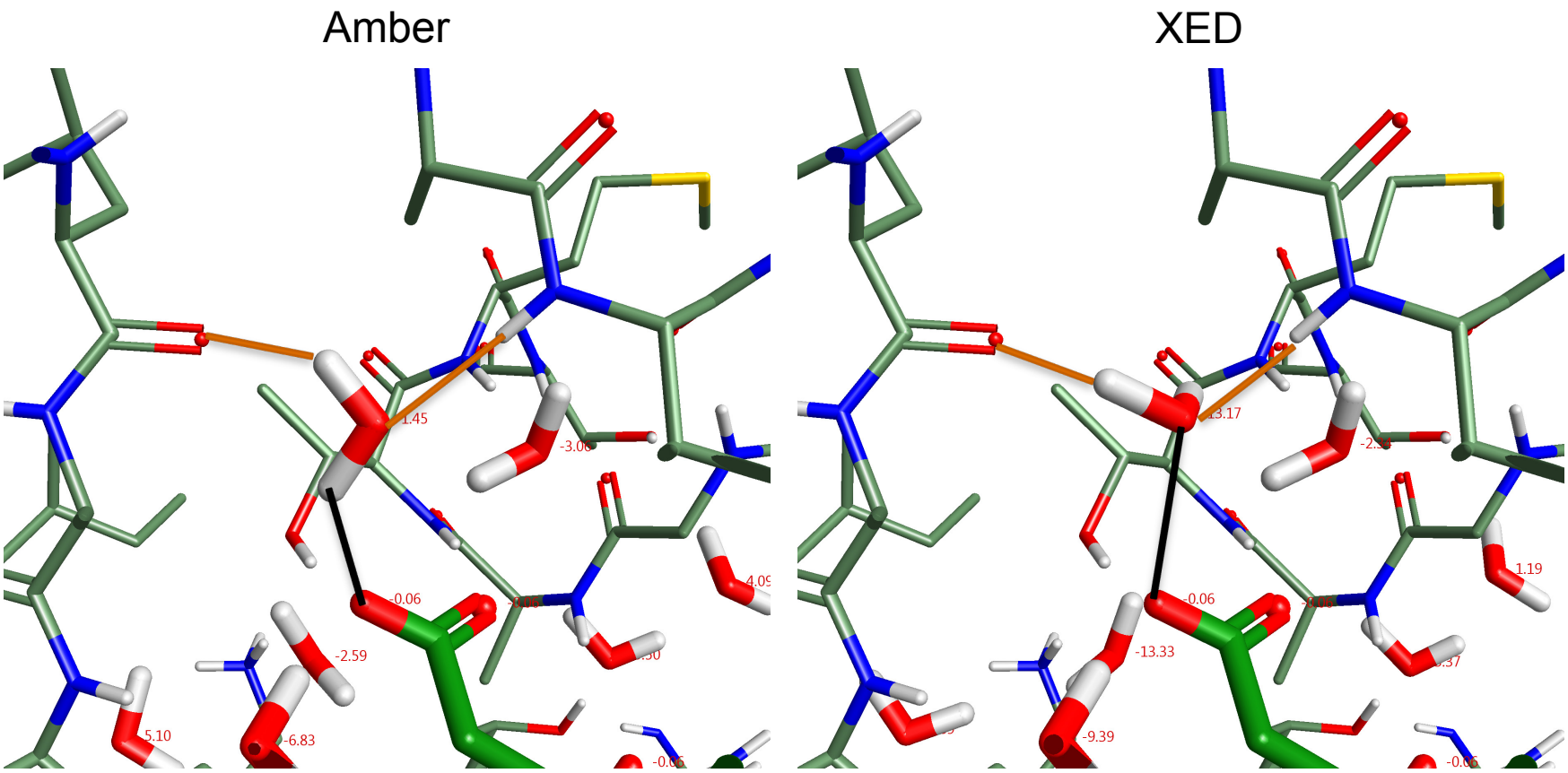
Examples & Comparisons

- Use Amber (GAFF) and XED force fields to calculate
  - Water positions & orientations
  - Water energies relative to solvent
- Compare for 3 proteins taken from the Iridium set
  - 1TT1 – Glutamate receptor with kainite ligand
  - 1N2V – tRNA-guanine transglycosylase, synthetic ligand
  - 1LPZ – FXa in complex with synthetic ligand

1TT1 – X-ray



1TT1 – Amber vs XED

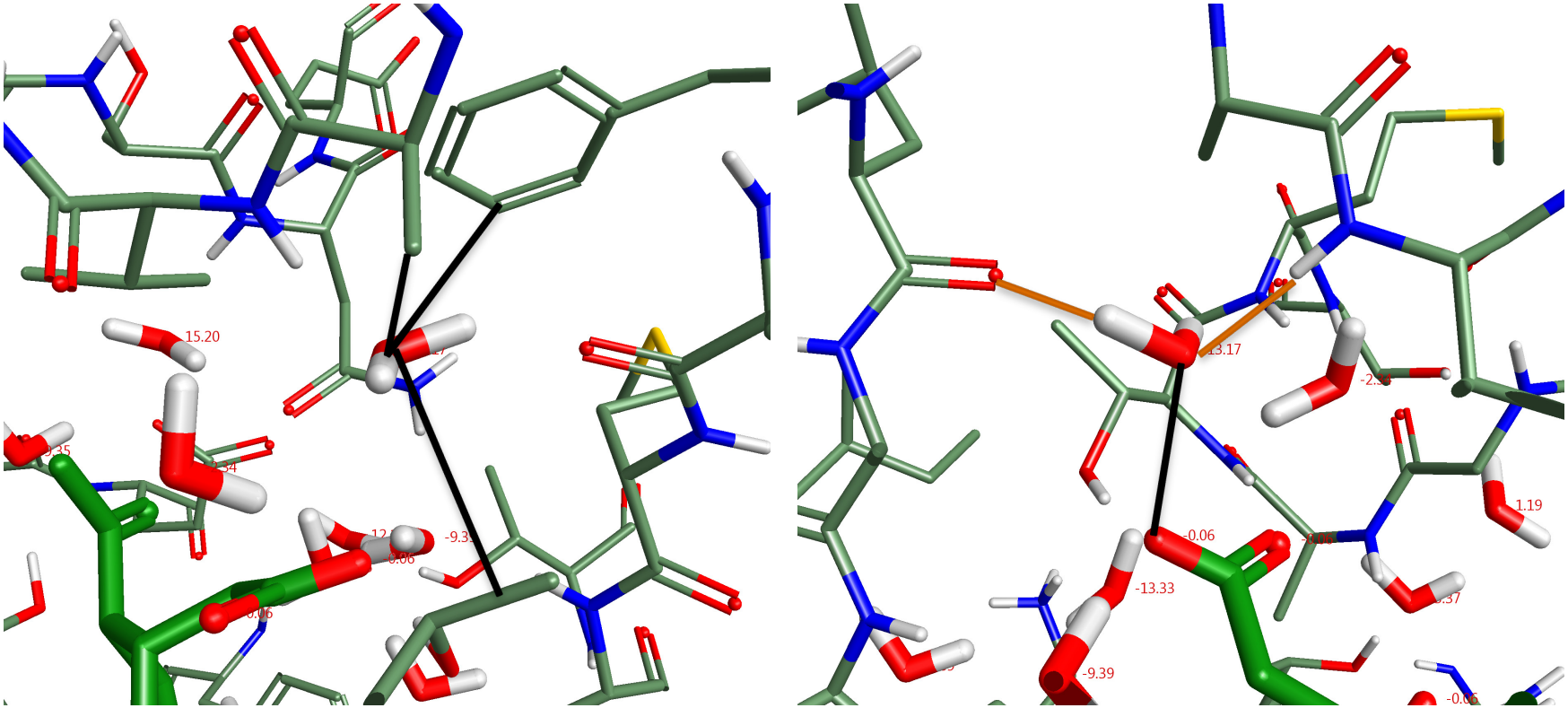


1.45Kcal → Unstable (just)

13.17Kcal → Unstable (very)

Both contain H-bonds but Amber has poor geometry to CO_2^-

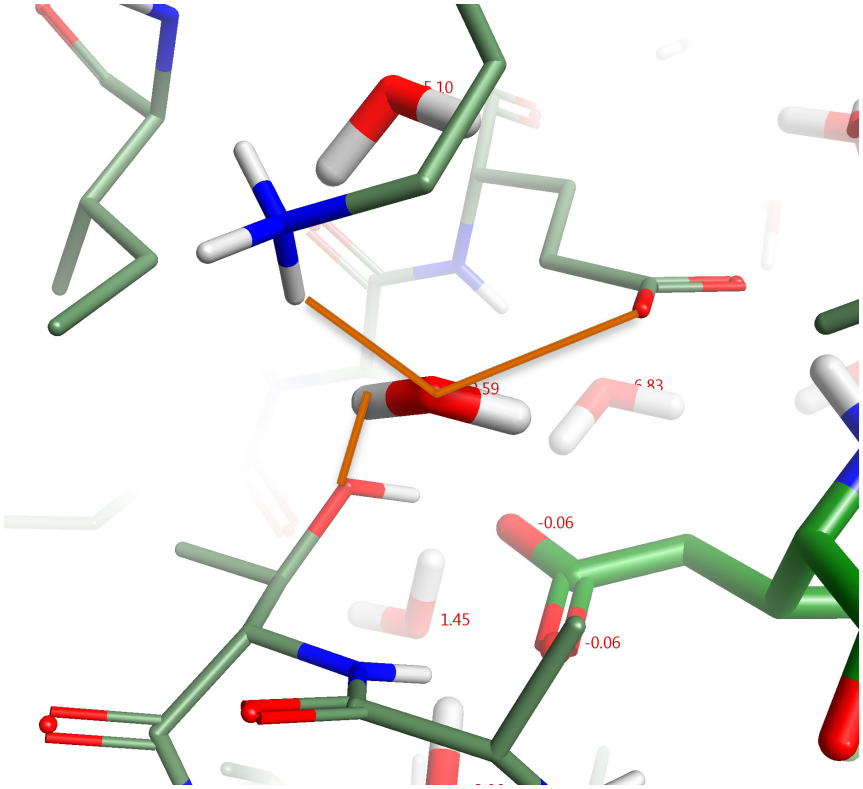
1TT1 – XED



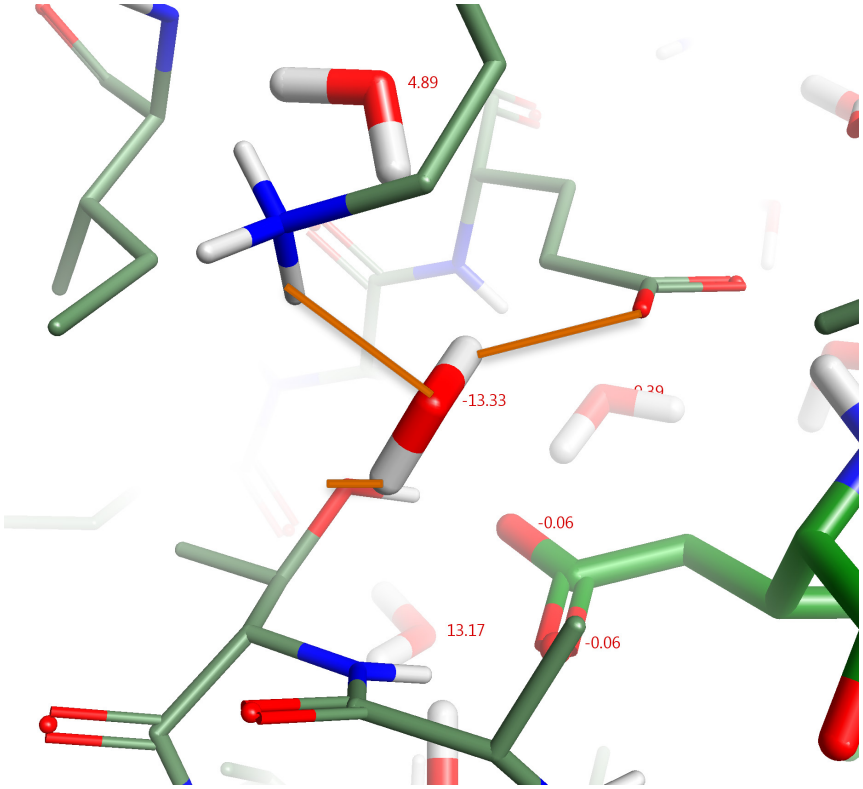
Environment at least partially hydrophobic

1TT1 – Amber vs XED

Amber



XED

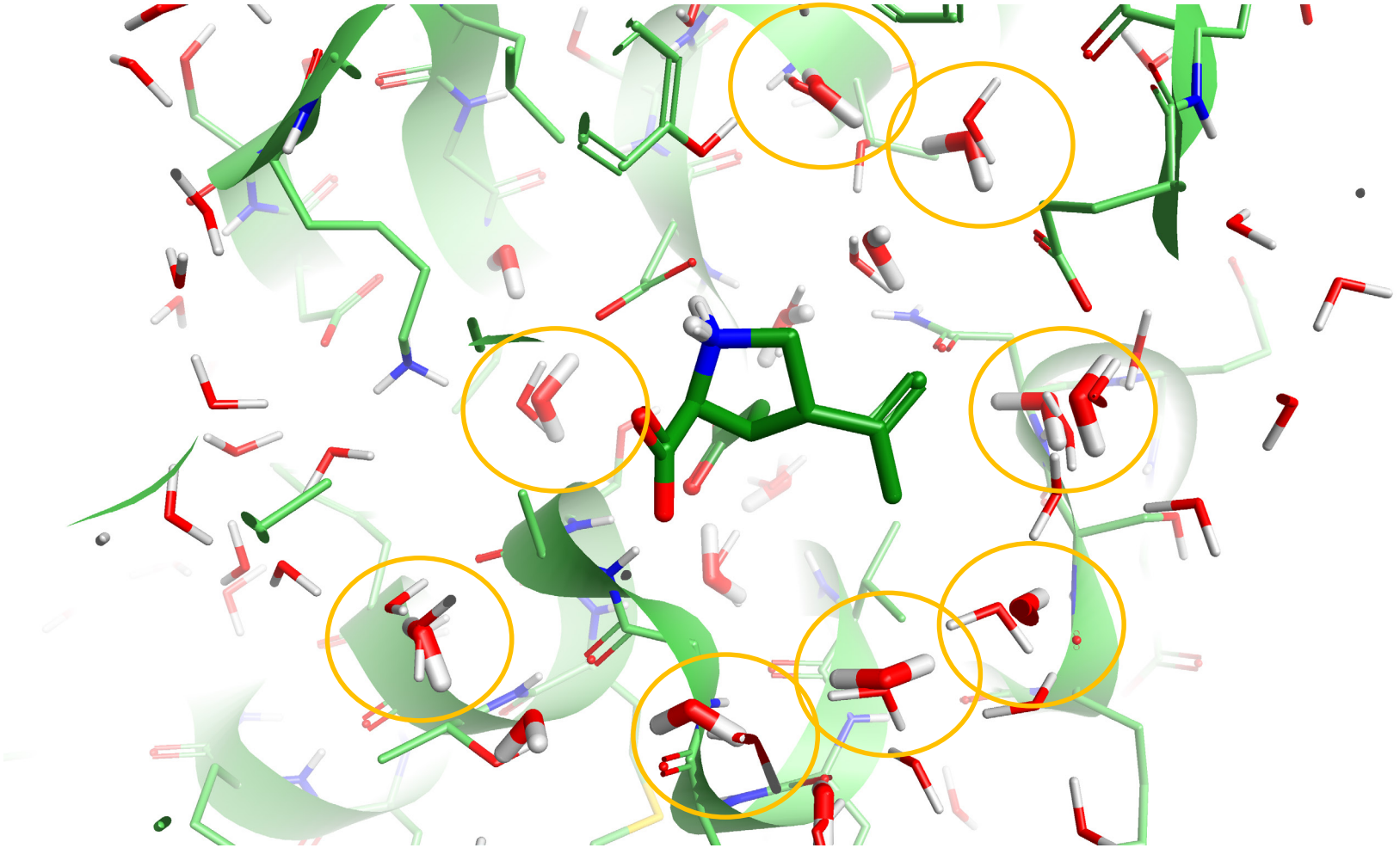


-2.59Kcal → Stable (just)

-13.33Kcal → Stable (very)

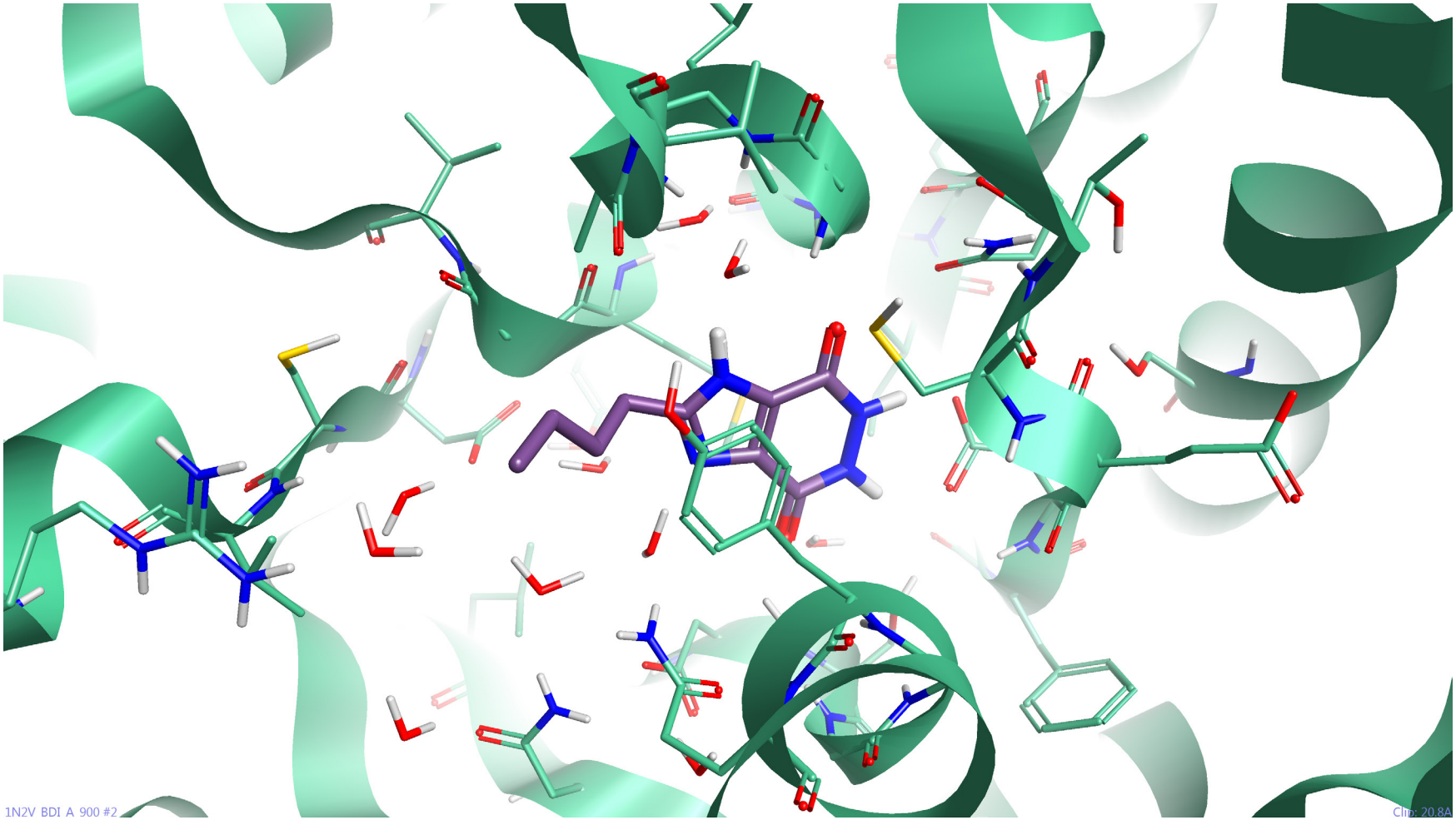
Extensive H-bonds suggest stability

1TT1 – XED shows good agreement with X-ray



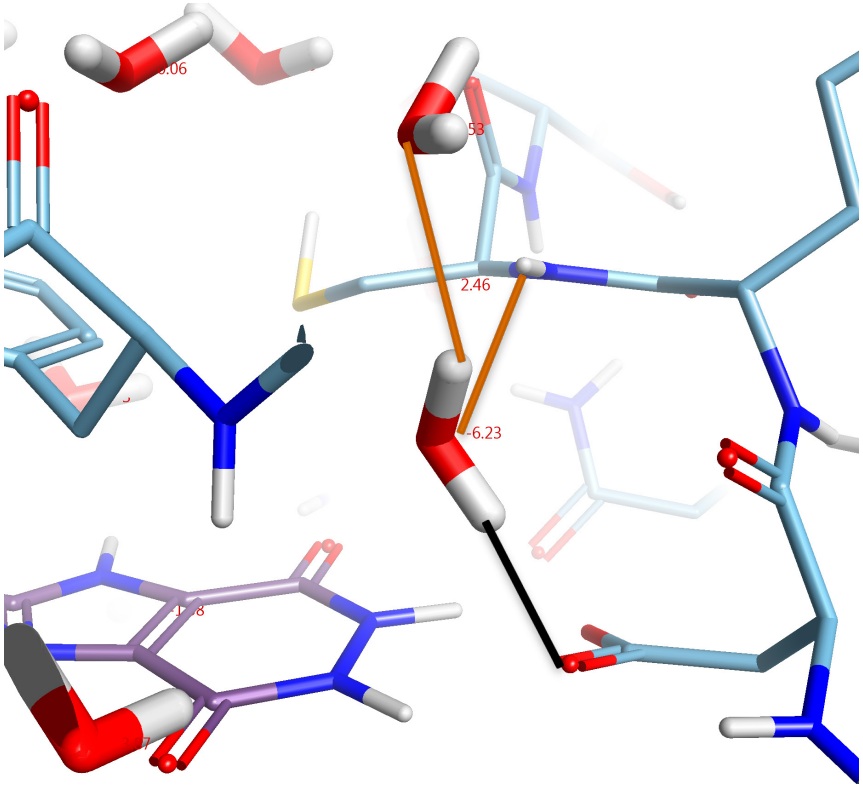
Thick sticks – XRAY, Thin Sticks - XED

1N2V – X-ray

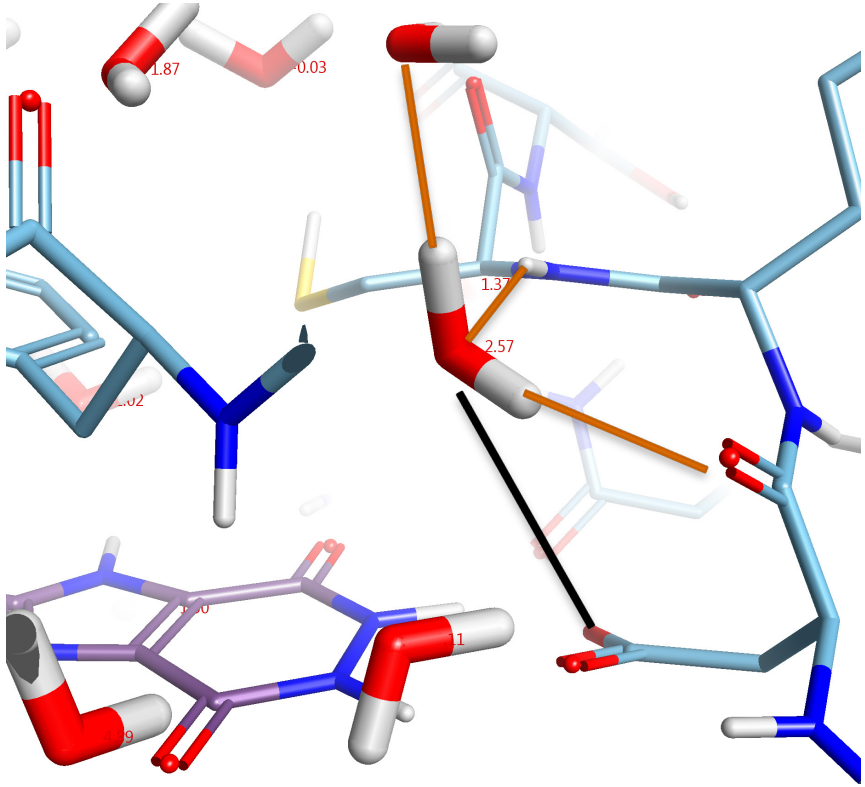


1N2V – Amber vs XED

Amber



XED

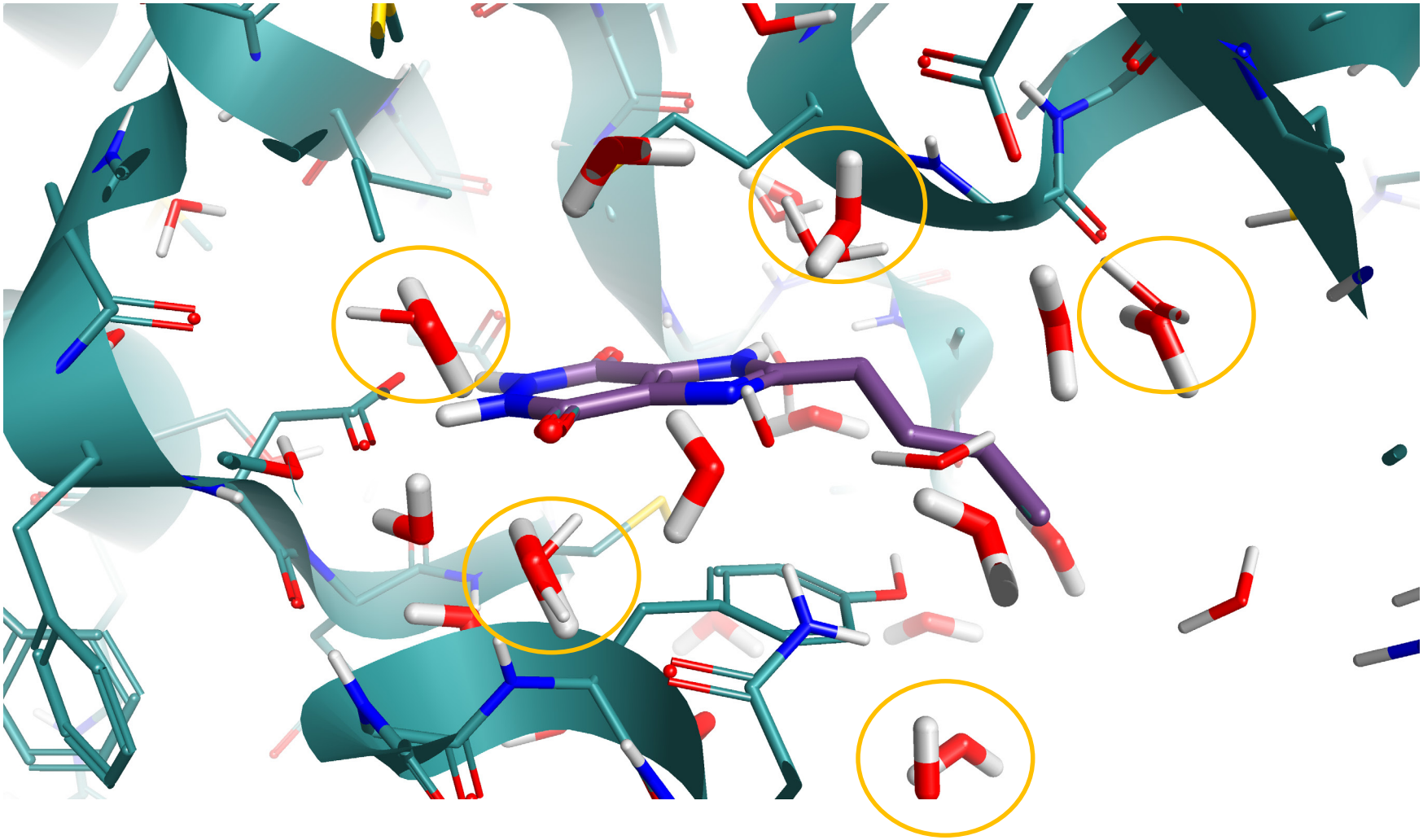


-6.21cal → Stable

2.57Kcal → Unstable (just)

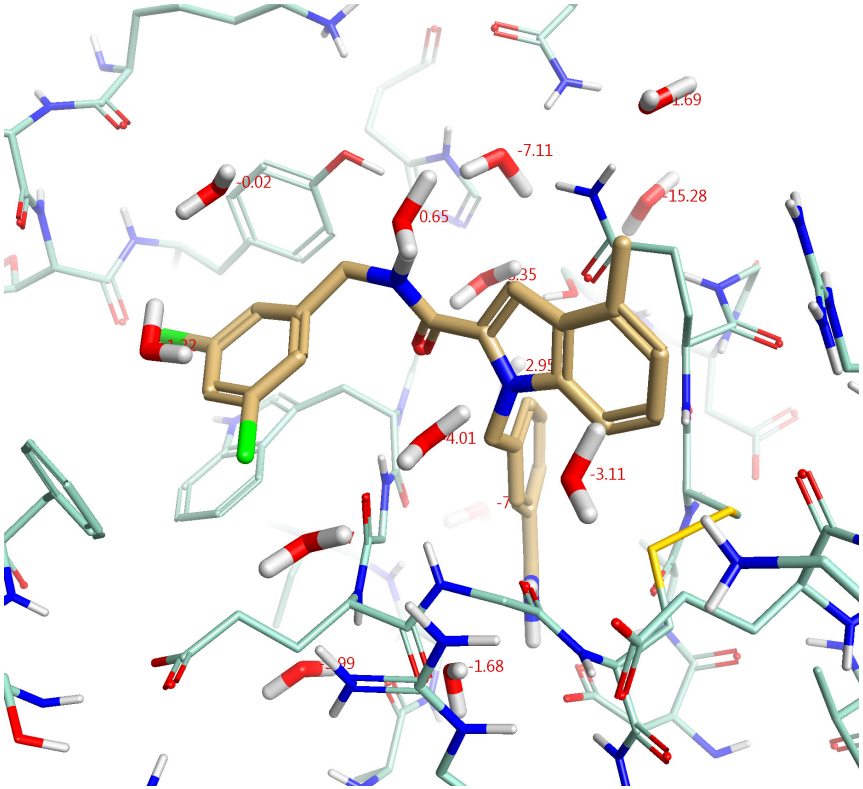
Both contain H-bonds but Amber has poor geometry to CO_2^-

1N2V – XED vs X-ray shows good agreement

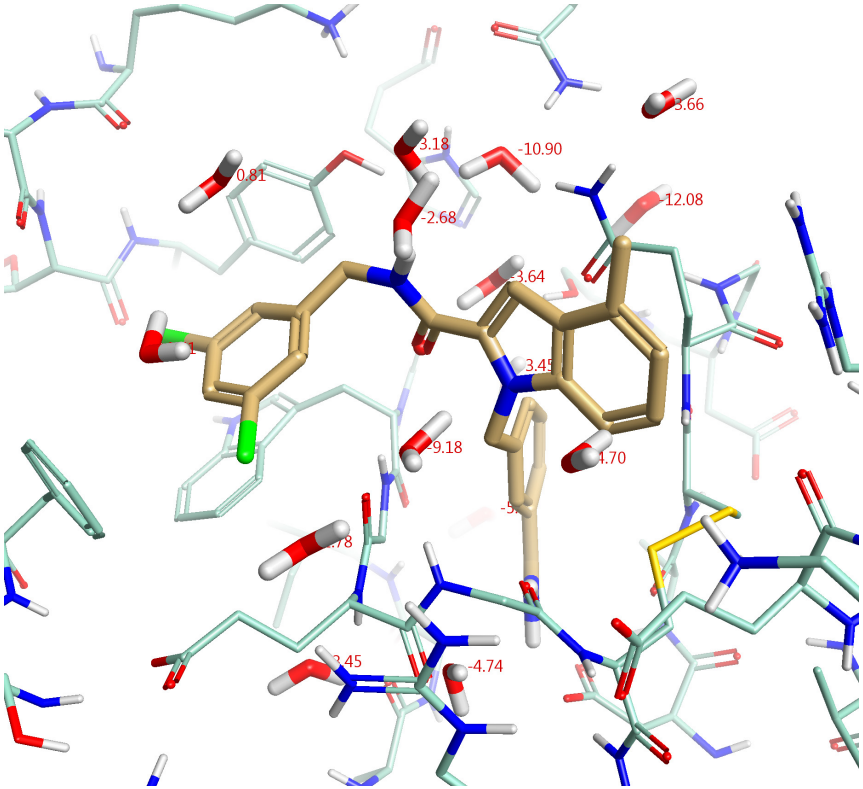


1LPZ – Amber vs XED

Amber

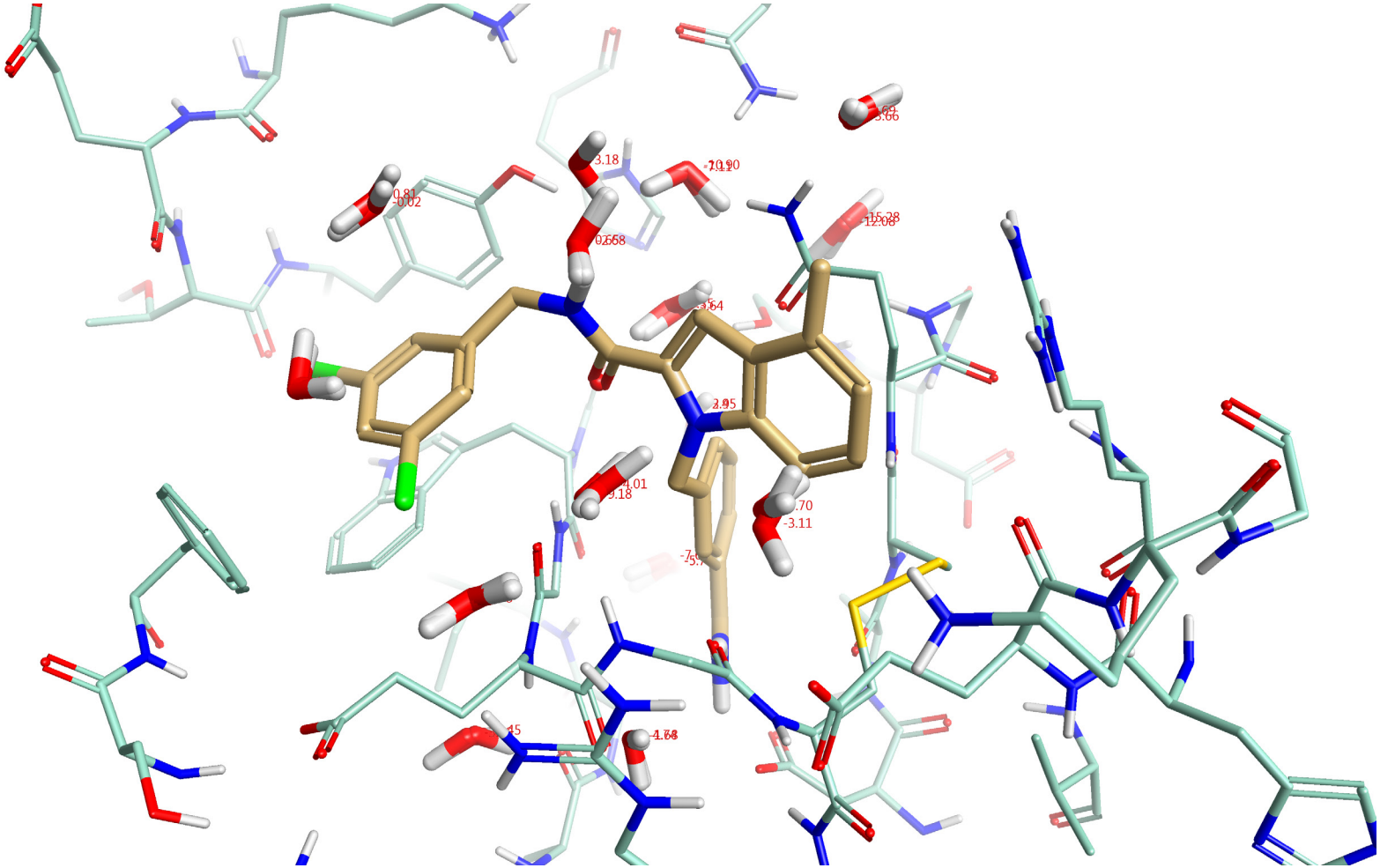


XED



Broad agreement in this case

1LPZ – Amber & XED give similar results



Broad agreement in this case

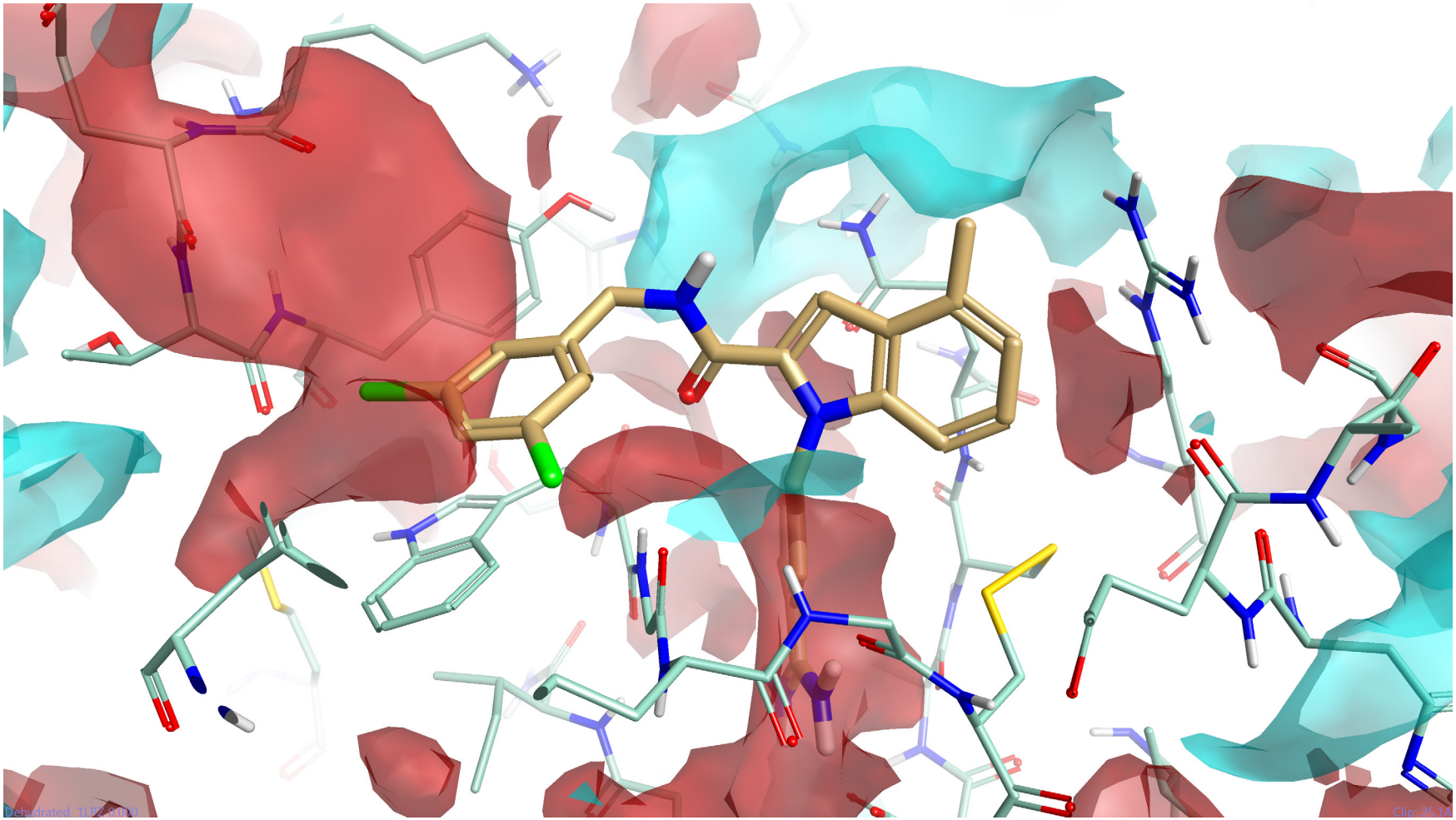
RISM with XED Conclusions

- Water patterns around small molecules look better with XED
- In proteins, XED provides better water patterns for most cases
  - A few limitations: it over-polarises amides
  - Validation is currently being performed
  - Difficult to find crystal data
- Release scheduled for 2016 as part of a new product

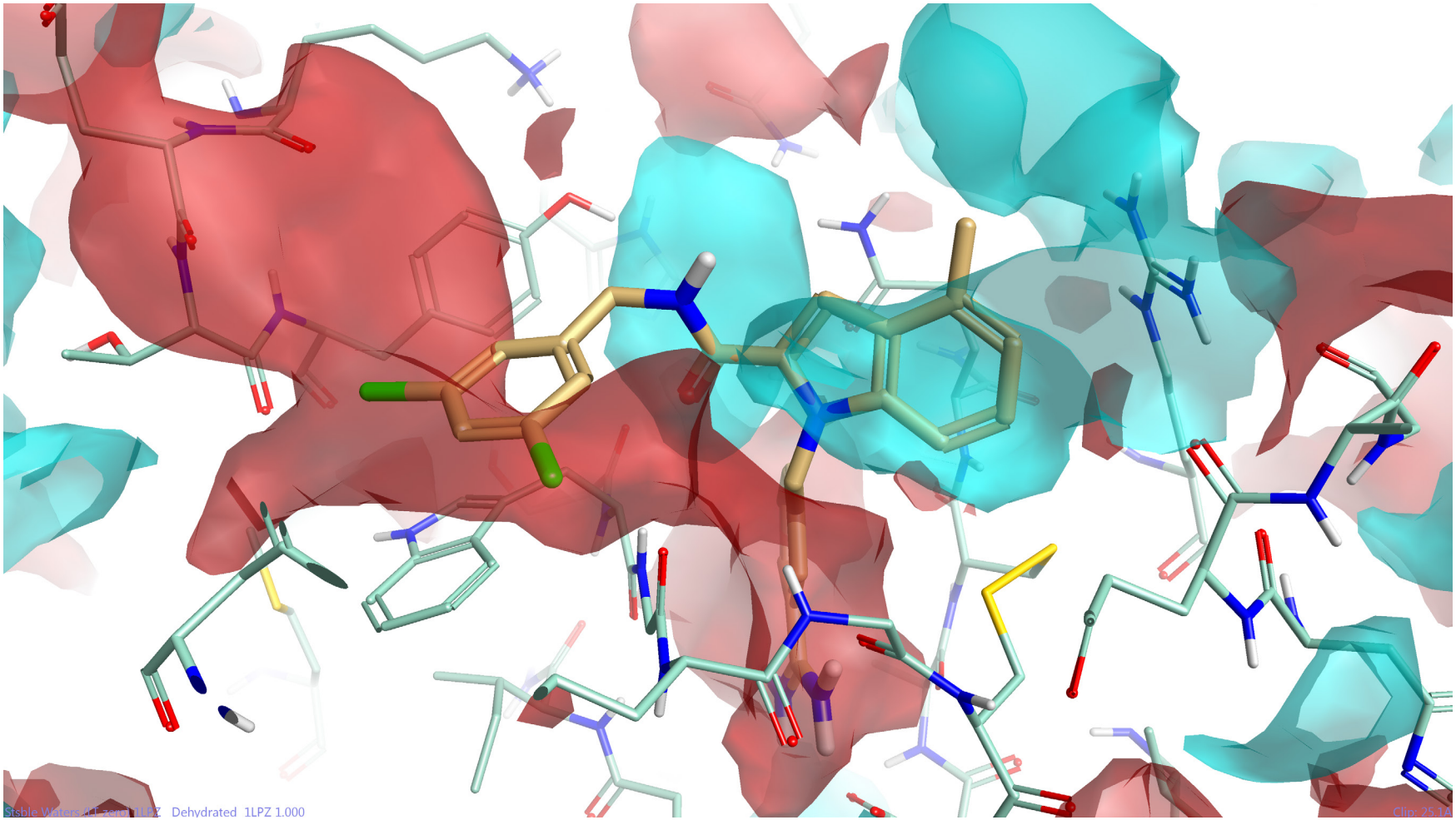
Bringing it all together

- Does using the stable waters change the protein interaction potential?
  - Using 1LPZ
  - Remove all waters with relative energy > -3 Kcal/mol
  - Merge remaining waters into protein
  - Add positive and negative interaction potentials

1LPZ Protein Int. Pot. – No Water



1LPZ Protein Int. Pot. – With Stable Water



Conclusions

- Protein electrostatics provide useful insights for molecule design
  - Well prepared protein essential
- 3D RISM using the XED force field promising method for assessing water in active sites
  - Validation in progress
- Combining water analysis enhances the view of protein electrostatics

Thank you

tim@cresset-group.com