Molecular Recognition of Cell Adhesion Proteins Is Water the "Glue" That Holds Pathogens to Host Cells?

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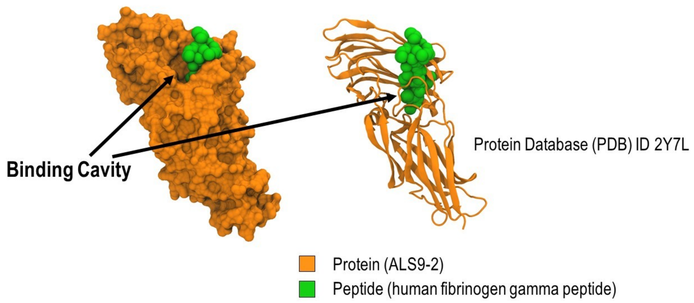
1. Introduction

1.1 Background

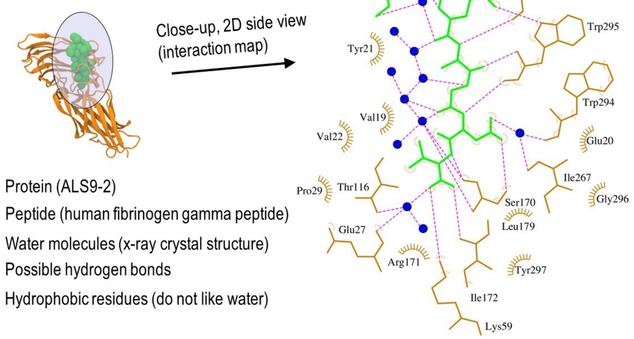
Candida is the most common fungal pathogens in humans. Though usually commensal, the species promotes nosocomial infections and can be life-threatening for those with weakened immune systems. Many pathogenic microorganisms express on their surfaces cell adhesion proteins which they use to stick to host tissues, such as our skin, to cause infection. *Candida albicans* expresses several kinds of agglutinin-like (Als) adhesion protein domains, which have the remarkable properties of being able to bind to a wide variety of polypeptides through their peptide binding cavity; however, the binding mechanism is not yet understood. In the X-ray structure of *Candida albicans* Als9-2 with human fibrinogen gamma peptide, a network of water molecules forms a bridge between the protein and the peptide in the binding pocket.

X-ray Crystal Structure of ALS9-2 in Complex with Human Fibrinogen Gamma Peptide



1.2 Question and Hypothesis

How does the peptide "stick" to the protein? Hypothesis: Water molecules form interactions between the peptide and protein, stabilizing the bound conformation.



2. Objective

The aim of this work is to determine whether the network of water molecules in the binding cavity plays a role in the molecular recognition of ALS9-2 to the human fibrinogen gamma peptide.

3. Methods

3.1 Molecular Dynamics

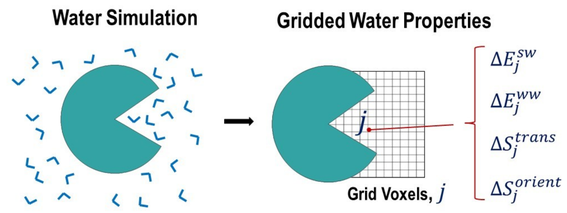
Molecular dynamics (MD) is a computational technique used to model the behavior of a system over an assigned period of time. We have performed a MD simulation of ALS9-2 without the peptide, using the TIP3P water model and ff14SB force field, and characterized the hydration properties of the binding site using solvation thermodynamics techniques.

Simulation Details:

- Constant volume simulation
- Temperature = 300 K
- AMBER 14 software
- Time = 100 ns simulation

3.2 Solvation Thermodynamics

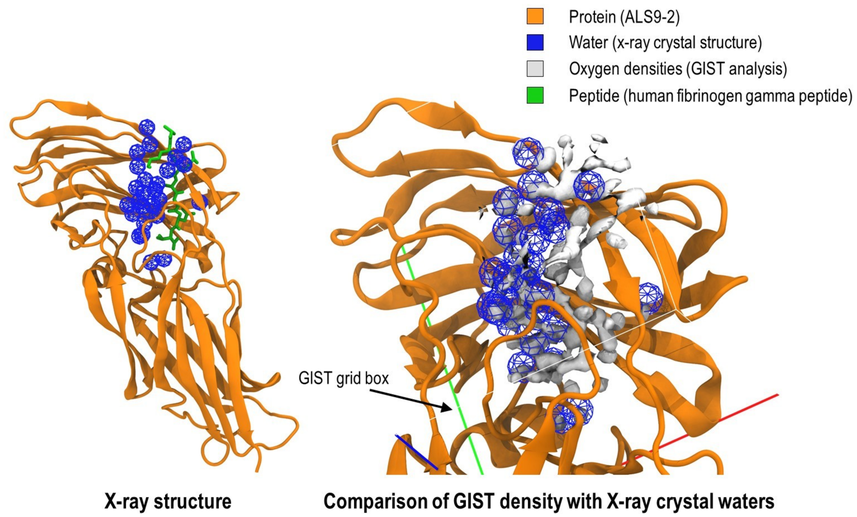
We characterized the hydration of the binding site using grid inhomogeneous solvation theory (GIST). GIST provides a solvation thermodynamic map of the region of interest regardless of density. GIST outputs include solvation energies and entropies as a function of the water molecules on the protein surface.



GIST uses explicit solvent simulations and solvation thermodynamic mapping to determine where specific hydration effects are important.

4. Results

Do We Observe the Same Hydration Patterns in the MD Simulation Without the Bound Peptide?



X-ray structure **Comparison of GIST density with X-ray crystal waters**

Many of the regions of high water density observed in the MD simulation correspond to the X-ray crystal waters found in the binding pocket of ALS9-2 bound to the human fibrinogen gamma peptide.

4. Results

4.1 Quantitative Analysis of GIST Density Relative to X-ray Crystal Waters

26 water molecules were found in the binding cavity of the X-ray structure. The GIST density was compared to locations of the X-ray waters to determine whether GIST density occupied the same regions.

GIST data was analyzed and compared to a TIP3P bulk water simulation (reference):

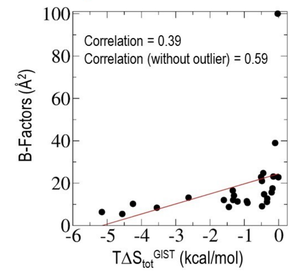
- **gO** = oxygen density of water in MD simulation relative to bulk water (TIP3P)
- **Total Energy (TIP3P)** = -9.53 kcal/mol per water molecule
- **Total Entropy (TIP3P)** = 0 kcal/mol per water molecule

Measure and Cutoff	GIST occupancy (out of 26 locations)
Oxygen Density (gO > 2)	24/26
Entropy (gO > 2, entropy < -2.00 kcal/mol)	15/26
Energy (gO > 2, energy < -9.53 kcal/mol)	24/26
Free Energy (gO > 2, ΔA < -9.53 kcal/mol)	20/26

4.2 Does a Correlation Exist Between Water Entropies and B-Factors?

B-factors of the 26 waters (from PDB) were compared with entropy values from GIST to determine the role of the peptide in ordering these molecules in the cavity.

A moderately positive correlation was observed between the B-factors and corresponding GIST entropies.



5. Conclusion and Future Study

Initial results suggest that the water network in the binding site is not due to the bound peptide because of the high occupancy of GIST density in the same locations as X-ray crystal waters. This GIST density is energetically favorable and entropically unfavorable, or ordered, relative to bulk water in the binding cavity without the peptide. A moderately positive correlation is observed between experimental B-factors and GIST entropies. It is predicted that this correlation will increase when the analysis is performed on the protein-peptide complex. This result suggests that the peptide does play a role in the structure of the water network. Further analysis is needed to quantify these differences with and without the peptide.

6. References

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7. Acknowledgements

This work was supported by CRSP and CSTEP.