

# Predicting proteinase specificities from free energy calculations

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Molecular dynamics (MD) simulations and the linear interaction energy (LIE) approach[1] have been used to study the role of the primary binding residue (P1) in complexes between three different subtilases (subtilisin Carlsberg[2], thermitase[3] and proteinase K[4]) and their canonical protein inhibitor eglin c. The binding free energy was calculated for all complexes differing only in the nature of the amino acid at the P1 position, and calculations for 19 different complexes for each protein were carried out including charged and uncharged states of the ionizable amino acids but excluding proline. The effects of substitutions at the P1 position on the binding free energies are found to be very large, and positively charged residues were found to be the weakest binders for all three enzymes. For subtilisin Carlsberg eglin c complex, eight of the twenty different association constants have been determined experimentally and the calculated absolute binding free energy for 5 of these are in good agreement with the experimental data. Such correlations can be usefully extended to other subtilase-inhibitor complexes to provide a general measure of pocket binding specificity.

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## References

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