Unraveling the molecular details of the pH-dependent trigger in diphtheria toxin T domain

Place of work: BioISI-FCUL (C8, 8.5.50D)

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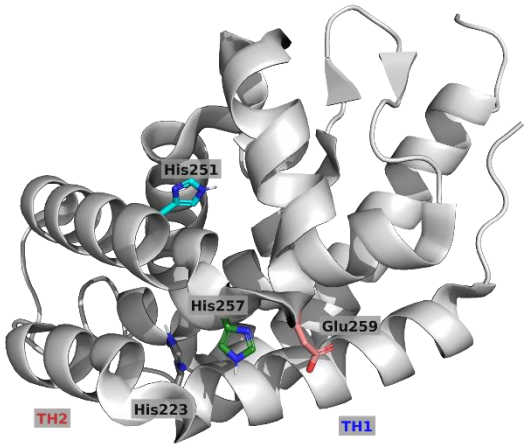
Diphtheria is a severe illness, which can be fatal if left untreated. The diphtheria toxin is produced by the *Corynebacterium diphtheriae* bacterium and is spread through respiratory droplets. The diphtheria toxin T (DTT) domain (see Figure) [1] translocates the toxin's catalytic domain across the endosomal membrane into the host cell cytoplasm, leading to cell death. The translocation process is triggered by endosome acidification and DTT undergoes a conformational change exposing a hydrophobic region (TH1-TH2) that enables it to insert into the endosomal membrane. The DTT domain is a potential target for therapeutics aimed at disrupting the translocation process and preventing the spread of the disease. The conformational destabilization of the DTT domain in the endosome is likely triggered by the protonation of key residues, although the molecular details of this process are still unknown. Several histidine residues have been proposed to be the pH sensors (see Figure), yet other typical protonatable residues, like aspartic and glutamic acids, cannot be excluded.



In this proposal, we aim to study the impact of endosomal acidity on the structural stability of DTT. We will use our state-of-the-art constant-pH molecular dynamics (CpHMD) method [2] to perform computational MD simulations under an acidic environment and characterize the conformational transitions triggered by the protonation of key residues, including the histidine residues that have been proposed by our collaborators (Prof. Alexey Ladokhin, Univ. Kansas, USA) [3]. The following tasks will be performed:

1. CpHMD simulations of the wild-type DTT protein at different pH values (3.0–7.0) to evaluate the impact of acidity on the protein.
2. Extend the previous simulations to several DTT mutants, including H223Q and H257Q, to help the experimentalists rationalize their experimental data and propose a detailed mechanism for the acid-induced conformational transition.
3. Perform CpHMD simulations in the presence of a membrane model at low pH to assess the role of the lipids in facilitating the exposure of the hydrophobic regions of DTT.
4. Compile all results in an MSc thesis and in a paper for an international scientific journal.

[1] Steere B, Eisenberg D. Worldwide Protein Data Bank; 2000. doi:10.2210/pdb1f0l/pdb
[2] Oliveira NFB, Pires IDS, Machuqueiro M. J Chem Theory Comput. 2020;16: 6368–6376.
[3] Rodnin, M. V., Li, J., Gross, M. L., Ladokhin, A. S. Biophys. J., 2016, 111, 1946–1953.