Accelerated molecular dynamics simulation of glutamine recognition and binding by γ-glutamyltransferase.

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The enzyme-substrate molecular recognition process is a complex sequence of events which takes place on the microseconds to seconds time scale, thus its simulation requires the use of enhanced sampling methods to speed up phase space exploration. In particular, accelerated molecular dynamics (aMD) works by flattening the molecular potential energy surface, adding a non-negative boost potential when the system potential is lower than a reference energy [1]. We applied aMD to the study glutamine recognition and binding by γ-glutamyltransferase (GGT) [2]. Our approach allowed us to model the process of substrate recruiting by the enzyme and to identify a four step pathway describing the approach to GGT of a glutamine molecule, from the free state to its full insertion into the active site. In the first step (Fig. 1a), glutamine approaches the binding pocket without directly interacting with the inner residues and it is still able to depart from the protein surface. In the second step, glutamine orientation allows the formation of a stabilizing interaction between the negative charge on the carboxyl group and the N-terminus of Thr_{391}. Glutamine's amino group is also oriented towards a negatively charged pocket formed by Asn_{411} and Asp_{433} (Fig. 1b). In the third step of the proposed recognition pathway, the interaction established between glutamine amino group and Asn_{411} and Asp_{433} is associated to the weakening of the interaction between glutamine carboxyl group and Thr_{391}. This drives glutamine deeper into the binding pocket (Fig. 1c). Finally, in the fourth step glutamine attains its final binding pose into the catalytic site. The carboxyl group forms a salt bridge with Arg_{114}, the amino group is stabilized by the aforementioned negative pocket and the amidic group is now close to the hydroxyl oxygen on Thr_{391}, ready to react.

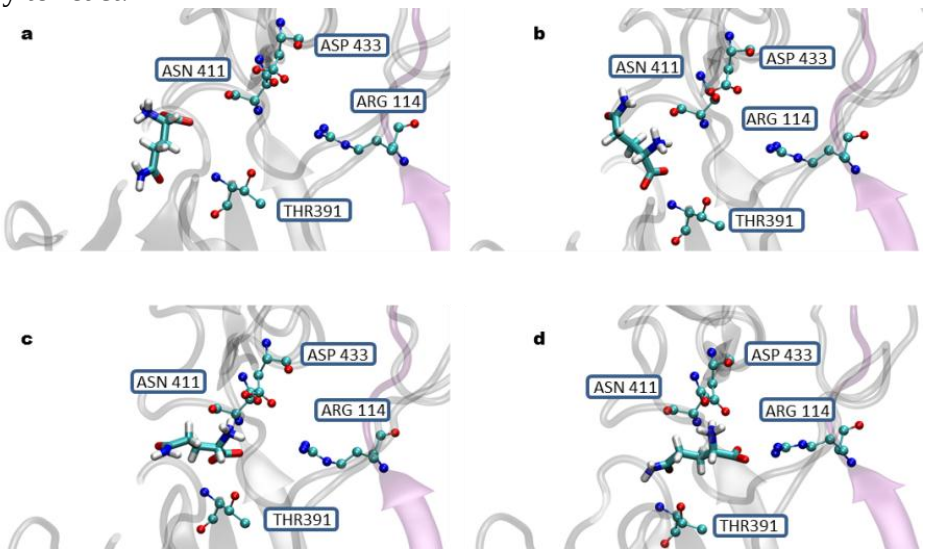

Figure 1: The four steps of our proposed binding pathway. Glutamine is represented in licorice, relevant GGT residues in CPK.

References

1. K. Kappel, Y. Miao and J.A. McCammon, *Q. Rev. Biophys.* **48** (2015) 479.
2. I. Castellano and A. Merlino, *Cell. Mol. Life Sci.* **69** (2012) 3381.