

### **Abstract for the general public**

Our main objective is to develop an efficient methodology to study transport pathways connecting buried active sites of many enzymes to bulk solvents. Access to enzyme active sites is tightly regulated inside the cell to maintain the balance of chemical reactions responsible for the proper functioning of the cell. Small molecules such as substrates or water molecules require transport pathways, called tunnels to reach the active site. These tunnels can be seen across different classes of enzymes and in every other particular enzyme. The biological significance of these tunnels can be understood as they were linked to the development of various diseases, and inhibitors binding these transport pathways became viable drugs. The tunnels are quite dynamic being located inside the enzyme due to the dynamic nature of these macromolecules and cannot be studied efficiently through one crystal structure. Also, the transport pathways are coated with residues that form gates, and the opening and closing of these gates determine the open and close state of tunnels. Computer simulations prove to be an efficient method to study the dynamics of tunnels but due to the rare opening of gates, it requires extensive and time demanding simulations which are limitations of the standard method considering detailed movements of all atoms. To overcome these limitations, we want to study two different faster computer simulation methods and how efficient they are in capturing the details of tunnel behavior. These simulation methods are less computationally expensive since they do not consider details on the atomic level but rather consider the simulated system from a more coarse-grained view. To compare the effectiveness of these methods, we will test their sensitivity to capture the effects of small-scale mutations on tunnels as well as their ability to generalize the study of transport pathways on many diverse proteins irrespective of their type. We believe that a much less expensive methodology developed in this project will be very promising for tunnel exploration in larger protein systems, opening up new possibilities for the large-scale application needed for the rational engineering of enzymes with buried active sites and the development of drugs targeting transport pathways.