Thymidylate synthase (TS) is a protein enzyme catalyzing an crucial reaction essential for DNA synthesis in all living organisms. Importantly, TS is an established target in chemotherapy with several drugs targeting the enzyme by mimicking either substrate or cofactor (two molecules necessary for assembling DNA building block by the enzyme). Moreover, there is an ongoing research aiming at the creation of selective inhibitors of bacterial and nematode TS variants, which may be used as antibiotics and in treatment of parasitic diseases, respectively.

Thymidylate synthase is a homodimer, *i. e.* it consists of two identical subunits, each containing an active site, a region in protein structure where the reaction occurs. There exists a large body of experimental evidence that TS exhibits “half-the-sites” activity, which means that only one active site is operational at a time. This, in turn, supports the hypothesis of the “molecular communication” between the enzyme active sites. Experimental data seem to support this notion, however only qualitative description for the bacterial enzyme is currently available.

Given the fact that such “molecular communication” effects are difficult to investigate with the experimental methods and at the same time are crucial for the understanding of TS functioning, we propose to use computational molecular dynamics simulations. This method is a “microscope” that allows to simulate behavior of a protein structure in solution and to depict, at the molecular level, the dynamical changes. We plan to carry out many simulations of the enzymes from different species with and without bound ligands. This way, we will perform a comparative analysis of the enzyme dynamics and behavior of the active sites. By analyzing the simulated motions of thymidylate synthase structures we hope to identify communication “wires” that connect the two active sites and are responsible for the “half-the-sites” activity. To validate our findings we will perform additional simulations in which the predicted “wires” will be cut. We expect that such “broken” variants of the enzyme will show significantly altered behavior, thus supporting correctness of our predictions.

Successful completion of the project will not only allow to significantly increase the knowledge about the dynamics and ”molecular communication” in the thymidylate synthase but also, although this is not the direct goal of the project, will allow to describe the dynamics and subtle differences between the enzymes from different species, which may in future be used to improve existing drugs, or design new that can be used in the targeted therapy against e.g. bacterial infections. Lastly, the outcomes of this project will shed light on the general phenomenon of ”molecular communication” in homodimeric enzymes, which include many, also clinically important, enzymes.