RECAPITULATING ALLOSTERY IN TRYPTOPHAN SYNTHASE ENZYME BY MEANS OF COMPUTATIONAL TECHNIQUES.

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Multimeric enzyme complexes, which perform many desirable chemical transformations, are often allosterically regulated by their protein partners, being the catalytic activity of isolated subunits seriously diminished.[1,2] Stand-alone enzyme subunits, however, are desirable for biosynthetic applications as lower metabolic loads on the host cell are needed, and the efforts to engineer activity, substrate specificity, etc. are drastically simplified. Tryptophan synthase (TrpS) is a heterodimeric complex that catalyzes the condensation of indole and L-Serine to form L-Tryptophan. Structural studies based on X-ray crystallography identified several key conformational states exhibiting open and closed conformations in both subunits.[1] For this enzyme, an allosteric network between the alpha-subunit (TrpA) and the beta-subunit (TrpB) keeps the proper conformational states along the catalytic itinerary, and allows indole diffusion from TrpA to TrpB through an internal tunnel. In an insightful work, Prof. Arnold and coworkers reactivated isolated TrpB subunits through directed evolution raising the catalytic efficiency to the level of the TrpS complex, by a stabilization of a closed/active conformation.[3] The presence of several activating mutations outside any allosteric site raised, however, the question of how distal mutations are able to recapitulate the allosteric regulation induced by TrpA remains to be answered. In this work, molecular dynamics simulations together with metadynamics calculations[4] were applied to map the effects of the introduced mutations, providing a detailed description of the enzyme most stable conformational states. This knowledge contributes to our current understanding on the effect of distal mutations on the protein conformational landscape, and how allosteric regulation can be recovered in isolated subunits.

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