Multifunctional Enzymes

Substrate Channeling in Multifunctional Enzymes

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In Short

- Substrate Channeling
- Dynamical Properties
- Substrate Specifity

The metabolism of fatty acids is one of many important biological processes. Metabolism is done in many organisms by the multifunctional enzyme-type 1 (MFE-1)(Figure 1). This enzyme has two active sites: hydratase and dehydrogenase active site. It plays a crucial role in the beta-oxidation pathway and catalyzes three reactions. First, the isomerization of 3Z.enoyl-CoA to 3E-enoyl-CoA following the hydration at first active site with the help of water. Then the fatty acid substrate is released from the hydratase active site and diffuses to the dehydrogenase active site where the third (oxidation) reaction takes place with the help of NAD+ (Figure 2).

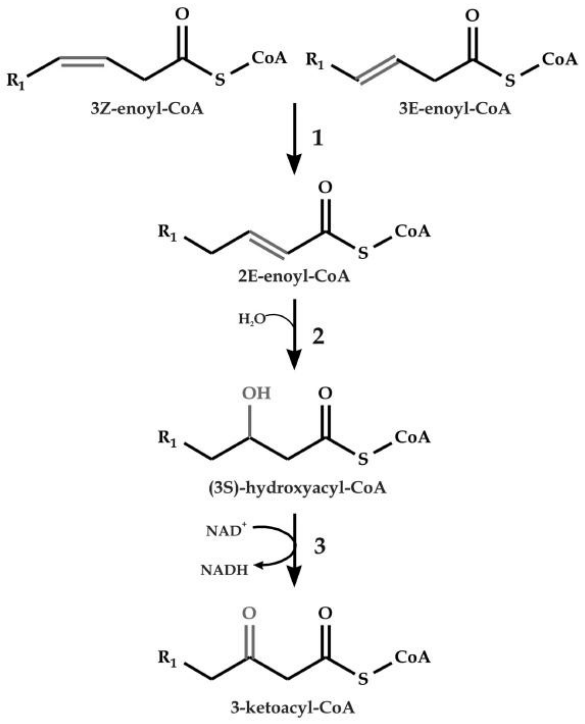

FIGURE 1. **Three steps of the overall reaction for the conversion of a 3-enoyl-CoA molecule into a 3-ketoacyl-CoA molecule as follows: *1*) the isomerization, *2*) the hydration, and *3*) the oxidation.**

But neither the mechanism of the substrate channeling nor the substrate specifity at the first active site has been investigated so far. Therefore, this project addresses these two issues at atomic level using classical molecular dynamics, accelerated molecular dynamics and hybrid QM/MM as well.

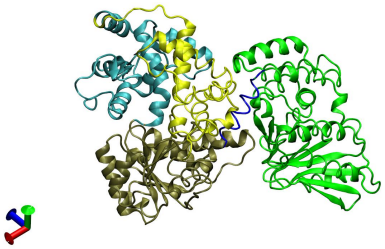

***Figure 1:** Overall Structure of MFE1. Domain A is green; the B-domain is blue; the C-domain is tan; the D-domain is cyan and the E-domain is yellow*

Classical molecular dynamics is a well-known computational approach and is usually used for studying the physical movements of atoms and molecules. The trajectories of these molecules are calculated by solving the Newton's equation of motion for interacting particles. In this case forces between the particles and also their potential energies are calculated by using molecular mechanics force fields (in this case CHARMM). Because of the huge molecular size of the MFE-1 it is necessary to capture those dynamics of the MFE-1 on a higher timescale. For this, we use accelerated molecular dynamics (aMD). This apporach modifies the potential by adding a boost potential. This potential raises the energy so that it improves the conformational space sampling that is not accessable via conventional molecular dynamics. Conventional molecular dynamics and accelerated molecular dynamics are fully covered and included in the well-known package of NAMD on the HLRN architecture. We will sample the structural and dynamical properties of MFE-1 using these computational methods.

Before addressing the substrate channeling between the two active sites, we will investigate in much greater detail the dynamical properties depending on substrate-, CoA- and NADH/NAD+-binding. For this our main interest is the change of the dynamical behavior of the hydratase active site depending on the protonation state.

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http://www.biomodeling.tu-berlin.de/

More Information

[1] Kasaragod, P., Venkatesan, R., Kiema, T.R., Hiltunen, J.K., Wierenga, R. *J. Biol. Chem.* **285**, 24089-24098 (2010).

[2] Kasaragod, P., Schmitz, W., Hiltunen, J.K., Wierenga, R. *FEBS J.* **280**, 3160-3175 (2013).

[3] Kiema, T.R., Taskinen, J.P., Piril, P.L., Koivuranta, K.T., Wierenga, R., Hiltunen, J.K. *Biochem. J.* **367**, 433-441 (2002).

[4] Adcock, S.A., McCammon, J.A. *Chem Rev* **106**, 1589-1615 (2006).

Project Partners

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