prising a central rod domain (approximately 2 000 A long) flanked by N- and C- terminal globular domains. Each of these domains contains several subdomains to which binding sites for various interaction partners have been mapped. Actin-binding domain (ABD) of plectin is located in proximity to its N-terminus. It consists of two so called calponin homology (CH) 1 and 2 subdomains.

Crystals of the plectin ABD were grown by the hanging drop diffusion method. Modification of crystallization conditions resulted in two crystal forms. Data from crystal form I (P21) were collected at room temperature to 2.0 A resolution and from crystal form II (P212121) at cryo temperature to 2.2 A resolution on the EMBL beamlines at the DORIS storage ring, DESY Hamburg. The structure was solved by molecular replacement method using utrophin ABD (PDB code 1QAG) as search model. Structures of both crystal forms were refined with the program REFMAC5. Recombinant molecule of the plectin ABD is an a protein consisting of 245 residues which form 11 helices. The structure is almost identical with the fimbrin ABD in spite of relatively low amino-acid sequence identity (23 %) and differs from those of utrophin and dystrophin mainly in orientation of CH1 and CH2 subdomains.

POSTERS

MOLECULAR DYNAMICS SIMULATION OF 1,2,3-TRICHLOROPROPANE IN THE ACTIVE SITE OF WILD TYPE AND MUTANT HALOALKANE DEHALOGENASE DHAA

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1,2,3-trichloropropane (TCP) is a toxic synthetic chlorinated hydrocarbon known to occur naturally. TCP is resistant to biological and chemical degradation and is often found as a water pollutant. Thermodynamics calculation shows that aerobic mineralization of TCP could provide sufficient energy to sustain microbial growth. The haloalkane dehalogenase (DhaA) from *Rhodococcus sp*. m15-3 hydrolyzes carbon-halogen bonds in a wide range of haloalkanes, including TCP, to the corresponding (halo)alcohol, releasing halide ions. Recently a way how to improve DhaA enzyme to utilize TCP as a substrate by double-point mutation (C176Y+Y273F) has been proposed by Bosma *et al*. [1].

The goal of the work was to explain by molecular modeling why is the mutated enzyme more effective than wild type DhaA. The molecular dynamics method was used to produce six 1 ns-long simulations; three of them with the DhaA wild-type in complex with TCP in tree different binding modes TCP(bm1), TCP(bm2), and TCP(bm3) and three of them with C176Y+Y273F double mutant of DhaA.. The binding mode TCP(bm1) corresponds to dehalogenation from C atom, while the binding modes TCP(bm2) and TCP(bm3) relates to the dehalogenation from either C atom.

Both simulations of wt/TCP(bm1) and C176Y+ Y273F/TCP(bm1) show that the dehalogenation from C is not possible, due to a sterical hindrance of TCP in the active site. Moreover, TCP changes its binding mode from TCP(bm1) to TCP(bm2) in both these simulations. The other simulations (wt /TCP(bm2), wt /TCP(bm3), C176Y+ Y273F/TCP(bm2) and C176Y+ Y273F/ TCP(bm3)) show that TCP frequently adopts a near attack conformation (NAC), i.e. conformation appropriate for SN2 attack during the whole simulation. Preliminary results indicate that NAC is more populated in simulation of double mutant (see Table 1).

Table 1: Population of NAC for studied systems.

System	TCP(bm2)	TCP(bm3)
wt-DhaA	20.7 %	6.5 %
C176Y+Y273F-DhaA	28.9 %	11.7 %

1. T. Bosma, J. Damborský, G. Stucki, D. B. Janssen, ***Appl. Environ. Microb.*. 68** (2002) 3582-3587.