Секция «Биоинформатика»

Balance between reaction stages is crucial for the design of paraoxonase activity in butyrylcholinesterase

Научный руководитель – Злобин Александр Сергеевич

Макарикова Ольга Леонидовна
Студент (специалист)
Московский государственный университет имени М.В.Ломоносова, Факультет биоинженерии и биоинформатики, Москва, Россия
E-mail: omakarikova@bk.ru

Organophosphates (OPs) are highly toxic irreversible inhibitors of acetylcholinesterase. The development of OP hydrolyzing enzyme—organophosphatase (OPase)—is regarded as a promising strategy to treat OP poisoning with human butyrylcholinesterase (BChE) regarded as a promising scaffold for such. A series of BChE variants were constructed to date exhibiting low activity against model OP paraoxon. Among them the most prominent variant is G117H. The reaction proceeds in two major steps with the formation and hydrolysis of a covalent conjugate respectively. The exact mode of G117H action is yet to be reported, however, it was hypothesized that His residue acts as a general base to activate the catalytic water molecule. Based on this idea variants harbouring substitutions to Asp and Glu have been proposed throughout the decade. The underlying logic was to either introduce an alternate general base or to couple His to an activating residue to mimic the common architecture of catalytic triads. However, no such substitution reached expected values of overall catalytic activity. We hypothesised that this may be due to the disruption of substrate binding and set the goal of this work to examine this phenomenon.

We started by proving the beneficial effect of G117D substitution on the catalysis of the second reaction stage via QM/MM modeling. We then utilized enhanced-sampling molecular dynamics to study the whole binding process of paraoxon in both the wild-type BChE and its G117H and G117D variants. We found that our simulation points to the existence of the previously studied peripheral binding site and also yields correct estimates of binding kinetics. The productive pre-reaction conformation was found not to be the most energetically preferred one, thus pointing to the potential direction of further design efforts. The major difference between variants was found to be the huge energetic disadvantage in pre-reaction state formation in G117D due to the strong electrostatic repulsion. Thus G117D substitution while being potentially beneficial for the dephosphorylation is indeed deteriorating to the productive binding of the substrate.

Our results show that in order to engineer a better BChE-based OPase both catalytic stages as well as binding process should be considered simultaneously. While highlighting the complexity of this task, we also present a number of directions that may be considered in order to finally solve it.

The reported study was funded by RFBR, project number 19-34-51043. The research is carried out using the equipment of the shared research facilities of HPC computing resources at Lomonosov Moscow State University https://paperpile.com/c/3o6smR/RJ9z.

Источники и литература

1) Supercomputer Lomonosov-2: Large Scale, Deep Monitoring and Fine Analytics for the User Community // Supercomputing Frontiers and Innovations. 2019. Vol. 6, № 2.