The Structural and Functional Studies of the Non-stereospecific α-Haloacid Dehalogenase (DehE) from *Rhizobium* sp. RC1

Azzmer Azzar Abdul Hamid[1], Ee Lin Wong[2], Kwee Hong Joyce-Tan[3], Mohd. Shahir Shamsir[2], Tengku Haziyamin Tengku Abdul Hamid[1] and Fahrul Huyop[2]

[1] Department of Biotechnology, Kulliyah of Science, IIUM, Bandar Indera Mahkota, 25200 Kuantan, Pahang, Malaysia
[2] Faculty of Biosciences and Medical Engineering, Universiti Teknologi Malaysia, 81310 Johor Bahru, Johor, Malaysia
[3] School of Bioscience and Technology, Faculty Science and Technology, Universiti Kebangsaan Malaysia, 43600 UKM Bangi, Selangor, Malaysia

Environmental pollution caused by the abundance of xenobiotic compounds in nature. For instance, synthetically halogenated compounds released from chemical industry were proven harmful to our health and environment. However, α-haloacid dehalogenases could catalysed the removal of halides from organic haloalkanoic acids and of interest for bioremediation. This study presents the first structural conformations and important residues of the non-stereospecific α-haloacid dehalogenase, DehE from *Rhizobium* sp. RC1. The enzyme was modeled using '*in silico*' technique and crystal structure of DehI from *Pseudomonas putida* PP3 was used as a template since both of them gets high similiarity to each other. DehE consists of only helices motif and depicted active site showed that the binding orientiations of both D- and L-2-chloropropionic acid by using substrate-docking analysis shared similar key binding residues among non-stereospecific α-haloacid dehalogenases. Twelve residues lining the active site has identified and some of them were verified using site-directed mutagenesis tests. Each residues was affected after mutation and Asp189 was proven to be as a catalytic residue for nucleophilic attack mechansim when its mutation resulted in total loss of activity. Three binding residues, Trp34, Phe37 and Ser188 were responsible for substrate recognition due to their mutation had diminish activity of the enzyme to below 20%. These details will promote more protein engineering studies to α-haloacid dehalogenases for future bioremediation and industrial applications.

Keywords: DehE, *Rhizobium* sp. RC1, non-stereospecific dehalogenase, α-haloacid dehalogenase, binding residues, catalytic residues, site-directed mutagenesis

References

Hamid AAA, Wong EL, Joyce-Tan, Shamsir MS, Hamid THTA, Huyop F (2013) Biotechnol Biotechnol Equip 27:3725-3736