

## Comparative modeling and rational design of $\omega$ -transaminase from *Caulobacter crescentus* for the increase in activity toward aromatic beta amino acid

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### Abstract

Accurate prediction of the 3-D structure of  $\omega$ -transaminase from *C. crescentus* exactly is essential for rational design of the enzyme. The 3-D structure of the  $\omega$ -transaminase from *C. crescentus* was constructed by comparative modeling. A homology model of the  $\omega$ -transaminase was constructed based upon the known structure of 2,2-dialkylglycine decarboxylase (PDB code 1DGE) as a template using the Genefold module in SYBYL 6.8.

The multiple alignments of the  $\omega$ -transaminases showed Lys281 was expected as the catalytic residue in the case of the  $\omega$ -transaminase from *C. crescentus*. In addition Trp53 and Arg260 were predicted to be positioned at the large binding pocket (on basis of *si* face at the PLP and Lys281) and predicted to interact with the carboxylate group of pyruvate and the side chain of amine substrate. Arg405 was also predicted as one of the conserved residues in the small binding pocket. Therefore, Trp53, Asp252, Arg260, and Arg405 should be positioned at the active binding pocket in the homology modeling.

The  $\omega$ -transaminase from *C. crescentus* showed high activities to aromatic amines and short aliphatic beta amino acids. To increase the activity for aromatic beta amino acid such as 3-amino-3-phenylpropionic acid, the large binding pocket of the active site should be enlarged to accept the large substrate. Several mutations were proposed to enlarge the binding pocket by means of the removal of steric hindrance between the substrate and the residues positioned at the entrance of the binding pocket and making the residues free where they were located behind the residues forming an active binding pocket by removal of the hydrogen bond. Mutant T284V and N285A showed the increase in the relative activity to 3-amino-3-phenylpropionic acid compared with 3-aminobutyric acid by two and eight-fold than the wild type.

### References

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