Homology Modeling and Molecular Dynamics Simulation of Recombinant Laccase

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Abstract

Today, due to the abundant use of recombinant and engineered proteins in various fields, predicting the tertiary structure of proteins opens a new window in understanding their function. In this study, the sequence of recombinant laccase was used, and homology modeling was done by MODELLER software version 10.04 [1]. The maize laccase (PDB ID: 6KLG [2]) was considered as a template. A Python script did the prediction and construction of 100 models. DOPE and GA341 methods were employed to evaluate the quality of the models. To validate the 3D structures, the UCLA-DOE LAB SAVES tool was utilized. Following the identification of the best model, we assessed the structural stability of the enzyme through 10 ns molecular dynamics simulation using Gromacs 2022 [3]. The simulated structure exhibited notable stability, as indicated by consistent RMSD values observed during the simulation. In comparison to the reference protein, our novel enzyme displayed an RMSD value of 1.23 Å. The recombinant laccase formed a significant number of intra-structure hydrogen bonds, totaling 16,993, while the reference protein formed only 458. Additionally, the solvent-accessible surface area (SASA) of the structures differed, with values of 20,534.47 and 19,377.14 Å²/kcal for the reference protein and recombinant enzyme, respectively. The novel enzyme's 3D structure opens avenues for both experimental and computational investigations, such as predicting binding sites with ligands and engineering enhancements through residue design in future studies.

Keywords: tertiary structure, recombinant laccase, homology modeling, molecular dynamics simulation

References

[1] T. Xie, Z. Liu, and G. Wang, “Structural basis for monolignol oxidation by a maize laccase,” *Nat. Plants*, vol. 6, no. 3, pp. 231–237, doi: 10.1038/s41477-020-0595-5.

[2] A. Šali and T. L. Blundell, “Comparative protein modelling by satisfaction of spatial restraints,” *Journal of Molecular Biology*, vol. 234, no. 3. pp. 779–815, 1993. doi: 10.1006/jmbi.1993.1626.

[3] M. J. Abraham *et al.*, “Gromacs: High performance molecular simulations through multi-level parallelism from laptops to supercomputers,” *SoftwareX*, vol. 1–2, pp. 19–25, 2015, doi: 10.1016/j.softx.2015.06.001.