In Silico Study of Binding of Camptothecin-Based Pro-Drugs to Human Carboxylesterase 2

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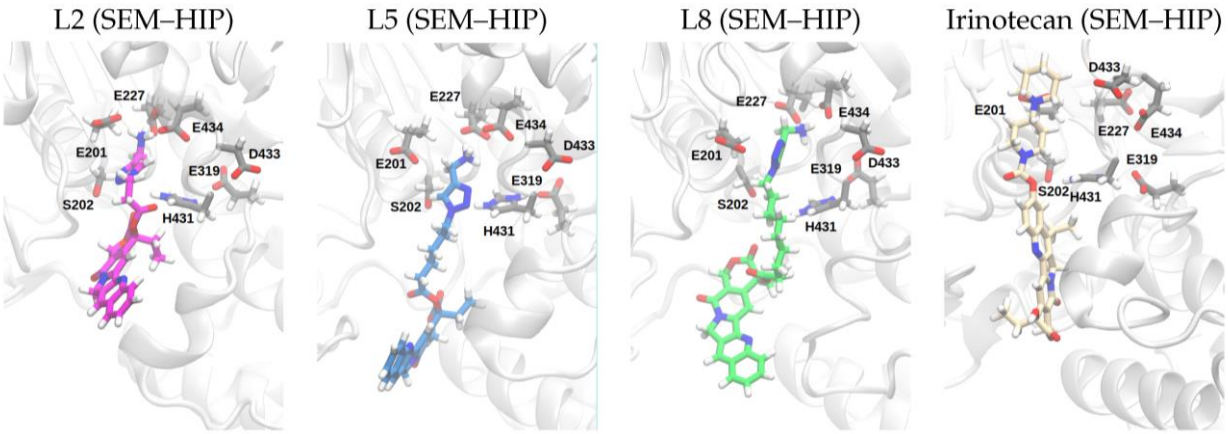

Pro-drugs, which ideally release their active compound only at the site of action, i.e., in a cancer cell, are a promising approach towards an increased specificity and hence reduced side effects in chemotherapy. A popular form of pro-drugs is esters, which are activated upon their hydrolysis. Since carboxylesterases that catalyse such a hydrolysis reaction are also abundant in normal tissue, it is of great interest whether a putative pro-drug is a probable substrate of such an enzyme and hence bears the danger of being activated not just in the target environment, i.e., in cancer cells.

In this work, we study the binding mode of carboxylesters of the drug molecule camptothecin, which is an inhibitor of topoisomerase I, of varying size to human carboxylesterase 2 (HCE2) by molecular docking and molecular dynamics simulations. A comparison to irinotecan, known to be a substrate of HCE2, shows that all three pro-drugs analysed in this work can bind to the HCE2 protein, but not in a pose that is well suited for subsequent hydrolysis. Our data suggest, moreover, that for the irinotecan substrate, a reactant-competent pose is stabilised once the initial proton transfer from the putative nucleophile Ser202 to the His431 of the catalytic triad has already occurred. Our simulation work also shows that it is important to go beyond the static models obtained from molecular docking and include the flexibility of enzyme–ligand complexes in solvents and at a finite temperature. Under such conditions, the pro-drugs studied in this work are unlikely to be hydrolysed by the HCE2 enzyme, indicating a low risk of undesired drug release in normal tissue.

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[2] I. Klemt, V. A. Reshetnikov, S. Dutta, G. Bila, R. Bilyy, I. C. Cuartero, A. Hidalgo, A. Wünsche, M. Böhm, M. Wondrak, L. A. Kunz-Schughart, R. Tietze, F. R. Beierlein, P. Imhof, S. Gensberger-Reigl, M. Pischetsrieder, M. Körber, T. Jost, A. Mokhir, *RSC Med. Chem.* **2024**, accepted. (DOI: 10.1039/D3MD00609C)