Abstract

Introduction: Non-small cell lung cancer (NSCLC) accounts for approximately 80% of all lung cancer cases. Clinical studies show a close relationship between NSCLC and epidermal growth factor receptor (EGFR). For this reason, first-generation epidermal growth factor receptor tyrosine kinase inhibitors play an important role in the targeted treatment of NSCLC. Despite the initial successful results, drug resistance due to the secondary T790M mutation has limited the use of these drugs. The present in silico study was conducted with the aim of proposing potential inhibitors with high binding potential to EGFRT790M by analogue-based rational design.

Method and Material: In this project, four groups of erlotinib analogs, including 58 molecules with quinazoline core, were evaluated for their anti-EGFR activity. These molecules were designed or selected based on the structure-activity relationship (SAR) of quinazoline compounds that inhibit the epidermal growth factor receptor and structural similarity to the first-line therapeutic erlotinib. First, the drug-like properties of the molecules were calculated using the SwissADME server. Then, the free energy of binding of the erlotinib analogs at the binding site of the wild-type receptor and mutant forms was calculated by molecular docking studies and using Autodock 4.2 software, and the superior structures in terms of binding potential to the co-crystallographic ligand site were determined. To verify the stability of the superior complexes, a molecular dynamics simulation (MD) was then performed for 50 nanoseconds in an aqueous environment using Gromacs 5.1.1 software. After we obtained the stable ligand-enzyme complexes, in the final phase of the study, the molecular interaction strength at the ligand-amino acid level for the stable complexes obtained by MD was calculated using the electron density dependence method (DFT) by the Gaussian 09 program.

Results: The mutant forms G719S, T790M, L858R, and the double mutant L858R/T790M were considered to be the most important mutations for the occurrence of resistance. Based on the molecular modeling results, the two designed molecules 6a ($EGFR^{T790M}$ ΔG_b -8.48 kcal/mol) and 22a ($EGFR^{T790M}$ ΔG_b -9.03 kcal/mol) showed the highest Gibbs binding free energy upon binding to the receptor site. All parameters of the molecular dynamics studies, including root mean square deviation (RMSD), root mean square fluctuations (RMSF), radius of gyration, Rg), intramolecular and intermolecular hydrogen interactions, and solvent accessible area, demonstrated the ability of both molecules a6 and a22 to stably interact with the T790M mutant form of the receptor. Moreover, all parameters confirmed each other and showed that the 6a molecule elicited a stronger and more stable ligand-protein complex.

Discussion and conclusion: Although experimental methods are also required to validate the molecules obtained, the in-silico design strategy used in this study allowed the recovery of new structures (6a and 22a) with high binding potential to important mutant forms. epidermal growth factor receptor (from a clinical point of view) compared with erlotinib. It should be noted that the basis for selecting 6a and 22a from the group of 58 analogues studied was the higher free energy of binding of these two molecules compared with the standard drug (erlotinib). In addition, both molecules had stable binding to the T790M mutation form. The results of this study not only confirm the importance of the position of

the 6 rings of quinazoline for the binding potential of these compounds, but also demonstrate the dominance of hydrophobic interactions over hydrogen interactions.

Key words: NSCLC, Erlotinib, CADD, SAR, Molecular dynamics