11th International Conference
on Advanced Materials
Rio de Janeiro Brazil
September 20 - 25
ICAM2009

Design of a Nanobiosensor using Molecular Modeling Techniques

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Abstract – A nanobiosensor for detecting pesticides has been developed with the immobilization of the enzyme acetyl Co-enzyme A carboxylase (ACCase) on the tip of an Atomic Force Microscope (AFM). The electrostatic potential calculations provided a visual insight about the possible orientation of the enzyme on the AFM tip. The molecular docking calculation described the specific interaction between the pesticide diclofop and the enzyme ACCase (Fig.2), displaying an inhibition constant of 142.26 nM.

Among the many techniques and analytical processes used to analyze pesticides and other residues in the environment, the nanobiosensors are among the most efficient[1]. The aim of this project is to provide theoretical and experimental information to support the design of nanobiosensors for detecting pesticides (enzymatic inhibitors). The main goal is to simulate the chemical force microcopy (CFM) using molecular modeling techniques, with which the possible orientation of the enzyme on the AFM tip can be inferred. In this study, a 20ns explicit-solvent molecular dynamics simulation of ACCase enzyme was performed with 0.105 mol/L ionic strength. The OPLS-AA force field was used along with the GROMACS 3.3 package[2]. To characterize possible interactions between the enzyme and the AFM tip, the electrostatic potential for the enzymes was obtained by solving numerically the non-linear Poisson-Boltzmann equation applying a finite-difference procedure[3], performed using the APBS (Adaptative Poisson-Boltzmann Solver) program. To characterize the enzyme-pesticide interaction, a molecular Docking calculation of the pesticide diclofop-p on the ACCase active site was performed using the Autodock 4.0 program[4].

The molecular dynamics simulation showed that the ACCase enzyme has significative structural fluctuations in solution (1.0 nm) due to solvation of its polar chains and probably due to electrostatic repulsion among charged side chains. This hypothesis was confirmed by the electrostatic potential which displayed discrete regions with high charge concentration located in a different, specific region on the protein surface (Figure 1). This electrostatic potential landscape, which is weakly affected by the enzyme structural fluctuation, showed a high density of negatively charged groups far from the ACCase active sites, suggesting that AFM tips functionalized by any positively charged groups are suitable to immobilize the ACCase enzyme and preserve its biological activity. The results from Docking calculations in Figure 2 pointed to the best orientation of the pesticide diclofop in the ACCase active sites. This interaction between the 2 pesticides on the ACCase active site is favored by 9.34 kcal/mol, showing an inhibition constant (K_i) of 142.26 nM. These results suggest that the proposed biosensors are promising detectors for this class of enzymatic inhibitor pesticides.

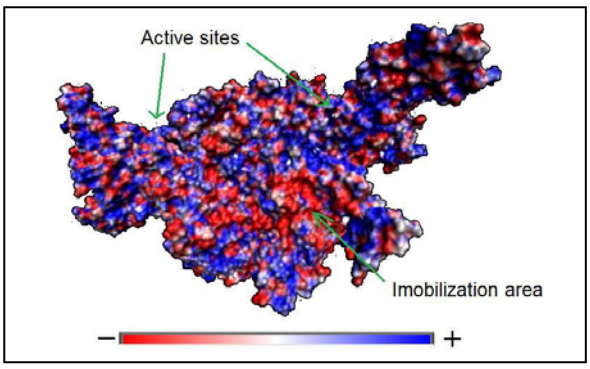

Figure 1: Electrostatic potential (-5 K_Bt/e to +5K_Bt/e) of the ACCase enzyme.

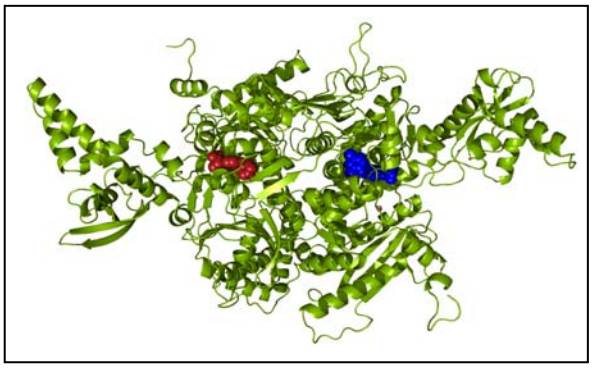

Figure 2: Diclofop pesticides docked on the ACCase active sites.

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