QM/MM (ONIOM) Calculation on FAK/Dasatinib Docking

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Introduction

The cells perform several activities at every level through biochemical signaling. In general, every signal pathway is mediated by a receptor protein[1,2]. The specific function of this class of enzyme is controlled by their molecular geometric covalent modification. These geometric molecular modifications are associated with the process know how phosphorylation. In this process phosphate groups originated from an ATP molecule react with the tyrosine amino-acids of the receptor tyrosine kinases modifying the original structure of the enzyme. This process is correlated with the normal division of the cell as well the control of cell proliferation through apoptosis. Cancer is a disease where an uncontrolled division of cells are observed. In this way, new cancer drugs are been developed from the knowledge of signal transduction[3,4,5,6,7,8,9]. Focal adhesion kinase (FAK) is a non specific receptor tyrosine kinase, localized inside cytoplasm, that is implicated in regulation of a number of cell signaling pathways, including spreading, motility and apoptosis[10,11,12,13,14,15,16]. The increase of phosphorylation in this kind of enzyme has been correlated with the interaction of integrins with fibronectins, which are adhesive proteins that help the cells adhere with cellular matrix.

Over expression of FAK has been correlated with several kinds of tumors[17,18,19,20,21]. Experimentally, it has been showed that the uncontrolled division of cell is verified by a phosphorylation of aminoacid tyronsine 397 in FAK[22,23,24,25,26,27]. The FAK phosphorylation is due of interaction between chemical signals and integrins with a non receptor tyrosine kinase. In the drug design context, molecular modeling has contributed to understand the interaction between FAK and pyrrolopirimidine inhibitors, helping in the discovery, development and optimization of new drugs[28,29,30,31].

Dasatinib is a known drug used in different cancers treatments and it is correlated with FAK inhibition35. However the molecular interaction between FAK and the dasatinib is unknown. In this work it has been performed molecular docking between FAK and the drug dasatinib. In order to understand the chemical interaction between dasatinib and the catalytic site of FAK, it was employed QM/MM calculation using PM6, HF, AM1, RMNDO approach for the higher layer and molecular mechanic for lower layer. UFF force field was employed to describe low layer in QM/MM calculations. Molecular dynamics was performed to determine the behavior of dasatinib inside the catalytic site. The amino acids of catalytic site were selected based in recent publications that show the interaction between FAK and the respective inhibitors. ONIOM approach based in RMNDO/UFF and HF/UFF are the more adequate quantum mechanic methods to explain the interaction between FAK and dasatinib without internal error coordinate during the optmimization.
The calculation results show that dasatinib interact via hydrogen bond with the aminoacid ARG426 (Arginine 426), that is the hydrogen bond found in others FAK inhibitors.

Methods

Based in recent studies[30], it was performed molecular docking using AutoDockVina following

the next scripts. The docking was performed keeping the protein FAK frozen. For the ligand the dihedral angles were retained free. It was used a grid with follow dimensions: x=-18, y=22 and z=16 angstroms. Center grid was centered in catalytic site of protein FAK. The size box used on the grid had the follow orientation: x=-0.444, y=10.627 and z=6.613 angstrons. The time employed to proceed the exhaustiveness search of ligand conformations in catalytic site was 500 seconds. In order to understand the interaction between the molecule dasatinib and FAK it was employed QM/MM calculations based in ONIOM approach present in Gaussian program. It was chosen the aminoacids CYS 502, LYS 454, MET 499, ARG 426, ALA 452, GLU 471, GLU 506, ARG 508, ARG 550, ASP 564 in order to compose the higher layer in according of current literature[30]. Quantum mechanics calculation based in PM6, B3LYP/6-31g, HF/6-31g, RMNDO were employed in higher layer that was optimized using the keyword quadmac, which does a quadratic step in the coordinates of all the atoms. 5000 SCF cycles were used in high layer optimization. UFF (Universal Force Field) was used in order to describe the Van-der Walls and electrostatic potential for the atoms in lower layer, that was maintained frozen during the optimization.

A short molecular dynamics with the classic force field CHARM and the program Hyperchem was employed in order to verify the dihedral angles displacements not observed during the optimization. For this purpose was used force field CHARM with one nanoseconds of simulation, a temperature of 300K and a dielectric constant equal 80 to simulate implicit solvent.

Results and Discussion

EQUILIBRIUM GEOMETRY AND MOLECULAR INTERACTIONS

Only the quantum mechanics calculation based in HF and MNDO approximation provided complete optimization of the system FAK-Dasatinb. The Table 1 show the energy associated with the interaction between FAK and the drug Dasatinib.

Table 1. Interaction obtained using HF/UFF and MNDO/UFF calculation.

METHOD	INTERACTION ENERY IN kcal
HF	-108
MNDO	-176

The optimized QM/MM calculation, based in MNDO/UFF and HF/UFF, show that the drug Dasatinib interact with the aminoacid GLU471 via hydrogen bonding, as can be seen in the Figure 1.

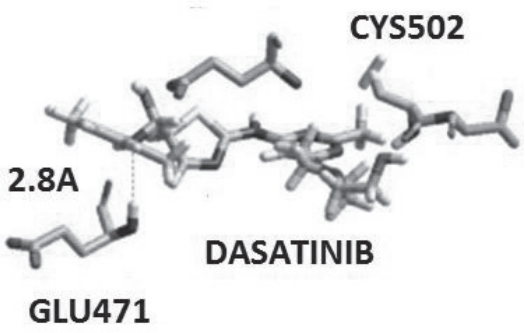

Figure 1. Interaction between Dasatinib and the aminoacid GLU471.

However when the molecular dynamics is performed, other hydrogen bonds into the catalytic site are found as shown in Figure 3. In these selected groups is observed dihedral angle rotations in dasatinib molecule during molecular dynamics as observed in Figure 2.

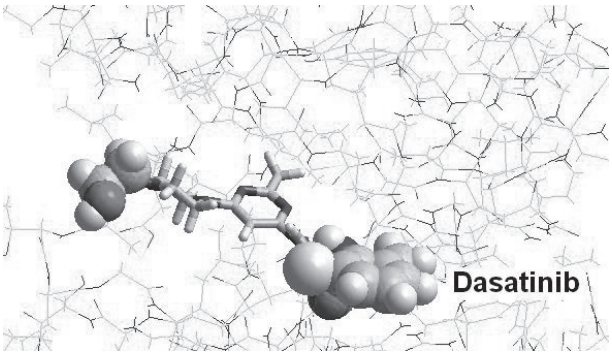

Figure 2. Chemical groups that showed dihedral rotation during the molecular dynamics.

The movement associated with the hydroxyl group allow the dasatinib perform hydrogen bond with the aminoacid ARG 426, as described in the Figure 3.

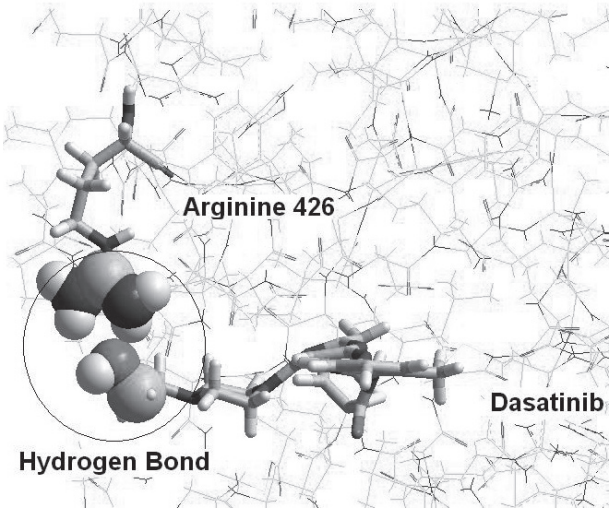

Figure 3: Hydrogen bond associated with the rotation of dihedral angle of group hydroxyl in the molecule of dasatinib.

This hydrogen bond is observed in others FAK inibitors as noted in the literature30.

THE PROBLEM ASSOCIATED WITH OPTIMIZATION

AM1, PM6 and B3LYP/3-21g were not useful to performed a complete optimization. These approximations revealed several errors in internal coordinates. In other hand HF/LANL2DZ and RMNDO performed a complete optimization of the complex FAK/dasatinib. In order to understand the optimization problem it was compared the molecular volume of Dasatinib with different FAK inhibitors The molecule dasatinib occupy a large parcel of volume of the catalytic site. This fact may be corroborated when the volume of molecule dasatinib is compared with the volume of two know inhibitors. These results are showed below in the Table 2.

Table 2. Molecular Volume occupied by different FAK inhibitos.

Inibidor	**Volume Molecular** A3
Pirrolo -pirimidina	1510.93
ATP	1044.64
Dasatinibe	2303.61

It is known that the semi-empirical method MNDO do not describe very well noncovalent interactions. The FAK/dasatnib bonding is essentially hydrogen bond and noncovalent interaction. In other hand the quantum-semi-empirical PM6 replace MNDO core-core approximation by Voityuk diatomic expression resulting in a corresponding increased hydrogen bond interaction energy[33]. There is also a problem associated with the HF method. The HF theory cannot describe the hydrogen bonding in some molecules[34]. Although DFT does not predict true dispersion interactions in the weak interaction region of zero overlap, it can still be useful for predicting correlation energy and even dispersion-like interactions in the region of overlap near the equilibrium geometry of even noncovalent complexes if one has an accurate enough functional[34]. Given these considerations we believe that during the energy optimization calculation via QM / MM , the inhibitor suffer deformation in their angles and dihedral , which cause errors in internal coordinates . These deformation are more pronounced for the functional B3LYP density and the quantum - semi-empirical PM6 method, that are more accurately methods to describe the interactions associated with dasatinib/ FAK docking.

Conclusions

Dasatinib establish hydrogen bonds with the aminoacid GLU471 in according with ONIOM optimization. In other hand, molecular dynamics reveals the possibility of the hydrogen bond with the aminoacid ARG426. The problematic associated with the convergence errors in some methods of simulation such as AM1, PM6 and B3LYP, are associated with the size of molecular dasatinib into the catalytic site and the quality of methods used. Methods with high quality in

the description of interactions and bonds provide more angles displacements in dasatinib into the catalytic site.

Acknowledgments

The authors are grateful for the support given from the FAPEG, CAPES, CNPQ and FINATEC.

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