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Quantitative relationships between lactam molecular properties and lactam/AcrAB-TolC complex geometry as determinants of MDR efflux

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INTRODUCTION

Multidrug resistance (MDR) of microbes and parasites as well as of cancer cells to currently used drugs is becoming one of the major problems in combating infectious and parasitic diseases and cancer, respectively. Among major mechanisms of multidrug resistance in cellular microbes and cancer cells are efflux pumps, macromolecular systems that extrude drugs and a large variety of structurally dissimilar substances from cell into the outside medium. Bacterial MDR efflux pump AcrAB-TolC exists in *E. coli*, *S. typhimurium* and several other Gram-negative bacteria as their major efflux system, being effective against -lactams and other antibiotics, organic dyes, detergents and many xenobiotics. It is a proton-motive device that connects the inner and outer cell membranes. The pump consists of the tube-like TolC trimer, the jellyfish-like AcrB trimer, and the AcrA oligomer. AcrB is responsible for attraction of substrates that are coming from periplasm/cytoplasm, their accumulation in its central cavity, and their expulsion through its channel and the TolC channel. Proton influx induces a series of allosteric changes in the pump and its components, enabling opening of the channels. Substrates in the central cavity must be placed and oriented in appropriate way to be extruded.

METHODS

In this work, the relationships between drug molecular properties and drug-receptor interaction geometry have been studied at quantitative level. Experimental 3D structures of four AcrB-drug complexes (dequalinium, ethidium, ciprofloxacin and ethidium [1]) were used. The drug-protein geometry and stereoelectronic molecular properties of these drugs and of 16 -lactams enabled prediction of positional and orientational parameters of these - lactams placed inside AcrB [2] *via* linear regression equations with the best variable selection. Prior to this analysis, the geometry of all drugs was modeled according to available experimental and modeled structural data, and then optimized at PM3 semi-empirical level after Montecarlo conformational search, taking into account the ionic state of the drugs at neutral pH and preserving the bioactive conformation of the four drugs as in the complexes with AcrB. Programs Titan and MOPAC 6.0 were used for all quantum-chemical computations. Crystallographc C_3 symmetry of AcrB (space group *R*32) facilitated definition of AcrB crucial axes and points, as well as drug-receptor geometry parameters (drug-central cavity distance and angle parameters). After predicting the drug-AcrB geometry parameters for - lactams, all drugs were superimposed in the common coordinate system of the uncomplexed AcrB. Molecular graphics was performed by using Titan, PLATON and WebLab Viewer programs. Experimental geometries of AcrB (complexed and uncomplexed) were from *E. coli*.

RESULTS AND DISCUSSION

Molecular descriptors of the four drugs (dequalinium, ethidium, ciprofloxacin and ethidium) that quantitatively correlated with these parameters (correlation coefficients above 0.82) were principal moments of inertia, molecular box parameters, dipole moment and its components, polarizability and hyperpolarizabilities. These molecular properties showed similar behavior as those for -lactams in terms of intercorrelations and correlations with the efflux activity of AcrAB-TolC pump in three strains of *S. typhimurium* (negative logarithm of Minimal Inhibitory Concentration) [3]. The new modeled AcrB/-l actam complexes show that drugs interact with the vestibule by electrostatic interactions, before binding in the central cavity and turning with their positive ends toward the opening of the AcrB channel. These results are consistent with known pump-mediated drug efflux mechanism and our previous quantitative structure-activity studies [4]. Elongated cylinder-like -lactam antibiotics with lipophylic side chains, significantly negative Y component of the dipole moment and low hydrogen bonding capacity seem to be good substrates of AcrAB-TolC MDR efflux pump

CONCLUSIONS

Stereoelectronic molecular properties of - lactams and structurally dissimilar compounds are quantitatively related to AcB-drug complex geometry. This explains the crucial point of the drug efflux, the orientation of an amphiphilic drug with respect to the inner membrane and the AcrAB-TolC pump in a Gram-negative bacteria.

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