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The structure–bioavailability approach in antifungal agents

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ABSTRACT

The thermodynamic and electrostatic properties of antifungal triazole and imidazole derivatives: itraconazole, fluconazole, miconazole and ketoconazole molecules were calculated. The main aim of our investigations was to identify molecular determinants that have an effect on bioavailability of studied compounds. This is solvation energy estimated by the ΔG values as well as electrostatic properties of the molecules. The influence of another substituents was also considered.

The presented values are discriminative and reflect very low solubility of ketoconazole in water in comparison with other molecules. The thermodynamic and electrostatic properties of molecules appeared to be good indicators of bioavailability of compounds studied here and they are likely to be used as screening parameters in Biopharmaceutical Classification System (BCS) development.

1. Introduction

Azole antifungal agents are the largest class of synthetic antimicotics. They are frequently used both systemically and topically in the treatment of systemic candida infections and mycosis [1,2]. This use is depended on the particular agent. The pharmacological activity stems from the presence of the heterocyclic aromatic five-membered ring, either imidazole or triazole.

Azole antifungal agents selectively inhibit CYP450 14α-demethylase in yeast and fungi [2–17]. This enzyme is involved in the convertion of lanosterol to ergosterol [18], which serves as a bioregulator of membrane fluidity and asymmetry and consequently of membrane integrity in fungal cells [10,11,14–17,19–22]. The basic nitrogen of the azole ring forms a tight hydrogen bonding with the heme iron of the fungal P450 shielding substrate and preventing oxygen binding [1,3–6,8,11,13,14,17]. The crucial for biological activity in humans is however, bioavailability of the active substance.

The inhibition of 14α-demethylase results in accumulation of sterol bearing a C14 methyl group changing the exact shape and physical properties of the membrane thus causing permeability changes and malfunction of membrane proteins and the consecutively, efficiently block of fungal growth [6,8,9,13,15,17].

First generation of azole drugs contain an imidazole ring (1-substituted), despite a wide spectrum of activity they have some shortcomings, e.g. miconazole, clotrimazole, econazole, for they are not absorbed orally.

Second generation azoles are more hydrophilic and can be administered orally.

The imidazole ring is still susceptible to metabolic degradation in vivo.

The best example is ketoconazole, which was the first member of this group with sufficiently good oral bioavailability to be used clinically for the treatment of deep-seated fungal infections [5,8].

Third generation azoles are based on triazoles – e.g. fluconazole, itraconazole, which provide effective oral therapy for many systemic fungal infections. These drugs are significantly more hydrophilic and so could be administered orally.

The triazole ring is much less susceptible to metabolic degradation in vivo.

The examples of direct use of the calculated ΔG_{solv} in the evaluation of a solubility of azole and similar antifungal agents in term of their bioavailability have been not reported in literature so far. Usually, the lipophilic properties of these compounds are being described on the base of the calculated log *P* [23–25]. However, available theoretical models in log *P* calculation do not consider the structure of studied molecules. In these approaches the values of log *P* are calculated as a sum of increments pertinent to particular chemical groups in the molecule of interest.

2. Results and discussion

The main aim of our investigations was to identify molecular determinants that have an effect on bioavailability of the following

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triazole and imidazole derivatives: itraconazole, fluconazole, miconazole and ketoconazole. This is solvation energy estimated by the free enthalpy (ΔG_{solv}) values as well as electrostatic properties of the molecules (Fig. 1).

The ΔG_{solv} of water and chlorbenzene solvation for four above mentioned medicinal products were calculated. Firstly, the conformational spaces were found for each compound in order to determine their most stable, lowest energy conformers. These calculations were carried out by Monte Carlo method with MMFF force field. Subsequently, the lowest energy conformers were initial structures in the further geometry optimization step of the calculations at HF (6-31G*) level. For the finally optimized molecules the ΔG_{solv} in water and chlorbenzene solutions were determined applying PCM model and HF method implemented in standard program packages [26,27]. Additionally, the electrostatic potential surfaces for each studied molecule were calculated. This characteristic seems to supplement well explanation of a solute–solvent interaction phenomenon.

The calculated values of ΔG_{solv} in water and chloroform and electrostatic potential for itraconazole, fluconazole, miconazole and ketoconazole are given in Table 1.

The presented values are discriminative and reflect very low solubility of ketoconazole in water in comparison with other molecules. In chlorobenzene the values of ΔG_{solv} indicate tremendous hydrophobicity of fluconazole possibly resulting in hyperbacteria-wall permeability and pharmacological activity. The very high value of ΔG_{solv} in chloroform has an indicative, rather than absolute value due to specifics of hydration model used.

The theoretical water solvation ΔG_{solv} data for miconazole and ketoconazole points at, in contrast to the experimental data, a stronger affinity to water of miconazole than ketoconazole. Undoubtedly, this inconsistency results from the simplicity of all available solvation models (e.g. PCM) which do not include solute–solvent interactions by hydrogen bonding, but first of all, from the possibility of formation the aggregates in solution by miconazole molecules [28]. The ketoconazole molecule contains more groups

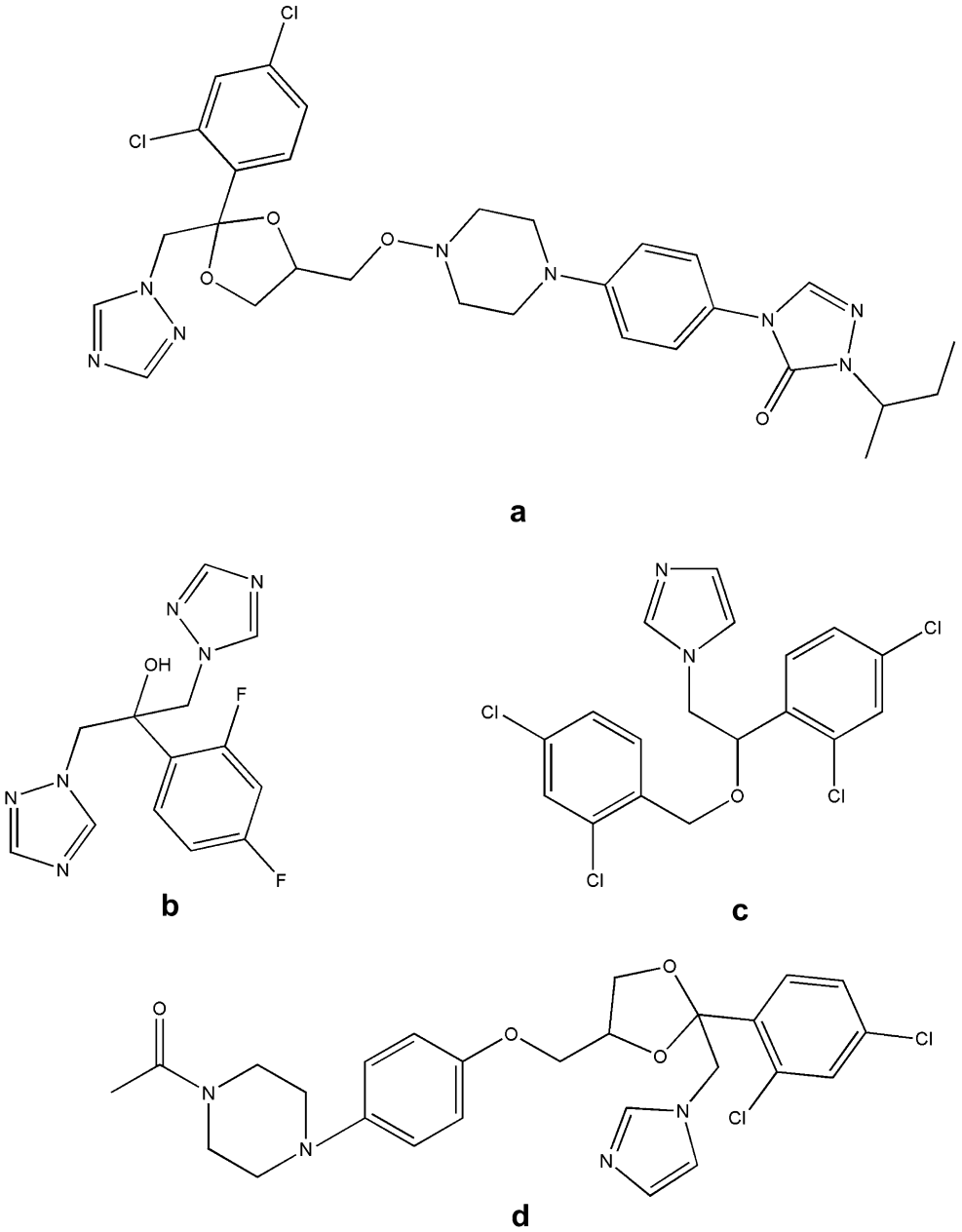

Fig. 1. The structural formulas of (a) itraconazole, (b) fluconazole, (c) miconazole, and (d) ketoconazole.

Table 1
The values of ΔG_{solv} and electrostatic potential.

Compound	ΔG_{solv} in water (kcal/mol)	ΔG_{solv} in chloroform (kcal/mol)	Electrostatic potential range (kcal/mol)
Itraconazole	2.53	−1.23	−54.63 to 30.70
Fluconazole	−1.18	−0.68	−45.02 to 41.65
Miconazole	−1.63	3.24	−51.97 to 34.33
Ketoconazole	720.88	−23424.38	−56.19 to 31.65

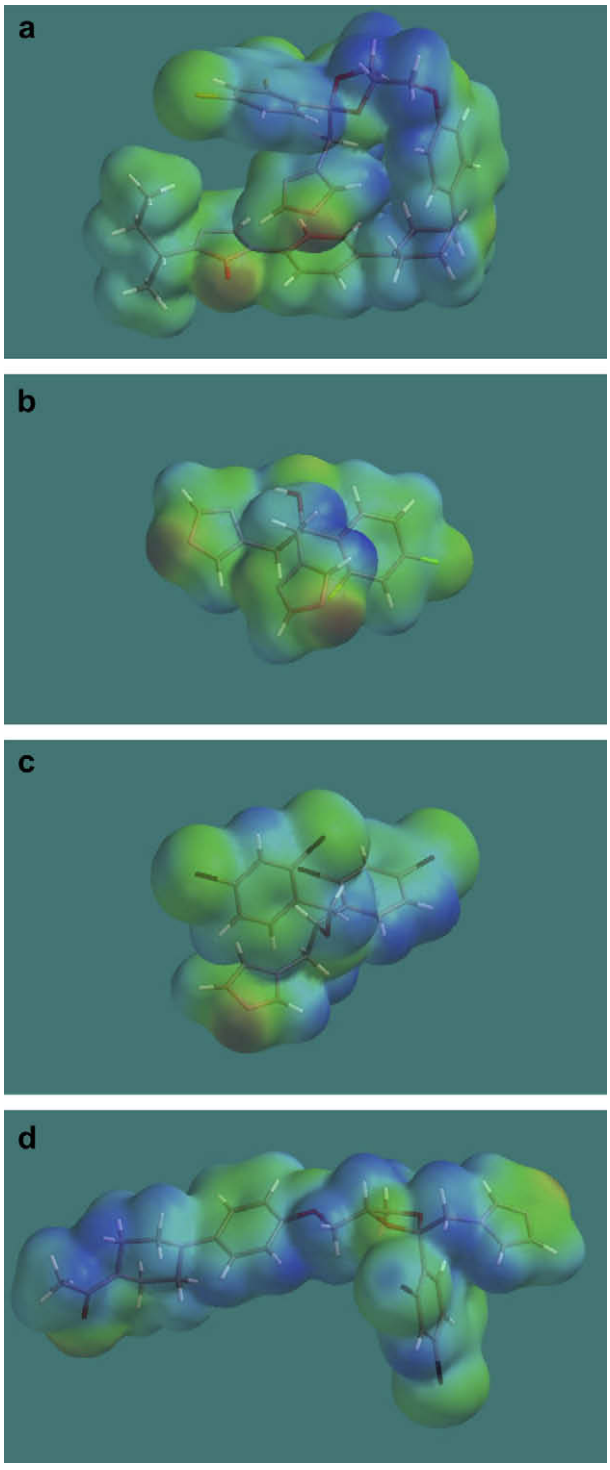

Fig. 2. The electrostatic potential distribution at isodensity surfaces of (a) itraconazole, (b) fluconazole, (c) miconazole, and (d) ketoconazole molecules (red-positive, blue-negative).

that are potentially able to form hydrogen bonding with water, than the miconazole one.

The other factor, the electrostatic potential distribution is consistent with ΔG_{solv} based findings. The map of electrostatic potential gives additional information on the distribution of solvent molecules in a solvation zone. From the energy of a solute–solvent interaction follows that the sites with the higher values of electrostatic potential in the environment of solute molecule concentrate more polar solvent molecules in the solvation zone (Fig. 2).

The electrostatic potential ranges define polarity of compounds and susceptibility to solute–solvent and molecular target interaction. The wider range potential distribution implies stronger interaction of the molecule with water medium. The ΔG_{solv} and electrostatic potential determinants should correlate with log *P* factor. The extremely low water solubility of ketoconazole did not allow to determine log *P* experimentally, due to detection problem at vanishable water concentration. However, this clearly indicates low hydrophilicity of ketoconazole, what comes from theoretical calculation.

Results of our investigation can also serve as description of properties relevant to Biopharmaceutical Classification System (BCS). Characteristics like solubility and permeability are considered when products are classified into group I–IV [29,30]. The calculated parameters describe both solubility and tendency to cross biological membranes. Here, the lipophilicity factor expressed as more positive ΔG_{solv} in water or and higher log *P* values can be a useful tool to order chemical substances.

The thermodynamic and electrostatic properties of molecules seem to be a good indicators bioavailability and can serve as classification factors in BCS system.

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