Molecular docking study of ruthenium-*p*-cymene complexes with isothiazole derivatives as SARS-CoV-2 main protease inhibitors

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Abstract: Since proper treatment for COVID-19 still has not been developed, exploration of novel options is required. Activities of different metal complexes, promising results gained from examining different thiazole derivatives, and research in the field of natural products like *p*-cymene, produced an idea to test piano stool ruthenium *p*-cymene complexes with isothiazole derivatives as ligands. *In silico* methods are often used as the first step in a series of experiments during the development of new drugs, and docking simulations are a quick way to determine the feasibility of novel compounds as potential inhibitors of target enzymes. Existing compounds of ruthenium with published crystal structures were tested against the main protease of SARS-CoV-2. All of the tested compounds show a potential ability to bind to the target enzyme, while the compound with phenyl and morpholinyl substituents in isothiazole ligand shows the best activity among tested compounds. Authors feel confident that further research on this topic will yield compounds with even better potential activities against the main protease of the SARS-CoV-2.

Keywords: ruthenium, *p*-cymene, isothiazole, docking, SARS-CoV-2

1. Introduction

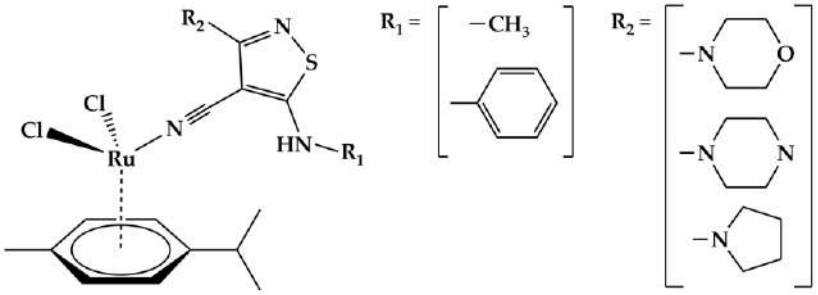
Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is a virus responsible for 2019 pandemic of disease COVID-19 [1]. In the early stages of pandemic, it seemed that vaccine development was in the spotlight, overshadowing therapeutic means of controlling this outbreak which can be seen from the extensive number of potential vaccines that were developed [2].

The use of metals as cures for ailments has a long history, while in modern medicine metal complexes were sometimes first-to-be-found or only effective treatments for certain diseases, like in the case of cisplatin with some types of cancer. Naturally, in the

fight against SARS-CoV-2 there was an effort to study established metallodrugs that already showed activities against viral diseases, like auranofin and bismuth citrate [3].

Large number of organic molecules were tested against SARS-CoV-2, selected from databases of known drugs and active compounds. The first step is usually *in silico* study of the select active compound like *p*-cymene [4] that is sent to later *in vitro* studies if the compound shows promise. Design of the novel drugs often starts from the promising small-molecule moiety present in many active compounds, like thiazole, from which potentially active derivatives are made by introducing diverse functional groups [5].

The search for effective therapy against COVID-19 is not over [6]. Considering promising results shown by metal complexes, *p*-cymene, and thiazole derivatives we decided to use molecular docking to examine the potential inhibitory power of existing [7, 8] piano stool *p*-cymene ruthenium complexes with isothiazole derivatives towards the active site of main protease of SARS-CoV-2 (M^{pro}). Scheme 1 describes selected compounds.



Scheme 1.

2. Methods

Structures of selected compounds were modeled following instructions in previously published work [8]. Structures are based on Scheme 1 and named according to Table 1.

Table 1. Naming convention of selected compounds.

Compound	R_1	R_2
1	methyl	morpholinyl
2	methyl	piperazinyl
3	methyl	pyrrolidinyl
4	phenyl	morpholinyl
5	phenyl	pyrrolidinyl

Structures of selected compounds were prepared for docking using AutoDockTools (ADT) [9] using previously calculated partial charges. Parameters for ruthenium interactions had to be added to the parameter file. The crystal structure of SARS-CoV-2 main protease with pdb code 6lu7 [10] was used as a docking target and was also prepared using ADT by removal of co-crystalized substrates and water, calculating Gasteiger charges, assigning atom types, and merging non-polar hydrogens. The structure of the compound designated as N3 from the 6lu7 file was used for method

validation. Docking simulations were performed using AutoDock 4 (AD4) [9]. Lamarckian AD4 hybrid genetic algorithm – local search method was used with 20 runs per tested structure, with maximum of $2.5 \cdot 10^7$ energy evaluations per run.

3. Results

After performing docking simulations, results were scored based on the estimated free energy of binding (ΔE_b), where lower energy indicates stronger binding and potentially higher potency of inhibitory effect. Derived results are presented in Table 2 with energy units as kilocalories per mole (kcal mol^{-1}) as is default for AD4.

Table 2. Docking results for each compound scored by estimated free energy of binding.

Compound	ΔE_b [kcal mol^{-1}]
1	-6.33
2	-6.07
3	-6.37
4	-7.21
5	-6.46

Compound N3 is potent inhibitor of M^{pro} with ΔE_b = -8.88 kcal mol^{-1} obtained from AD4 experiment used for validation. Compared to N3, tested compounds do not perform as well, but scores are good enough to indicate the possibility of binding at the active site of the enzyme. The best-scoring compound is compound 4. Every tested compound had access to the catalytic cavity of the enzyme and could be positioned adjacent to the HIS41 and CYS145 residues that are generally considered to be carriers of the enzyme's catalytic function [11]. Figure 1 shows the best scoring conformation of compound 4 in proximity of the catalytic dyad.

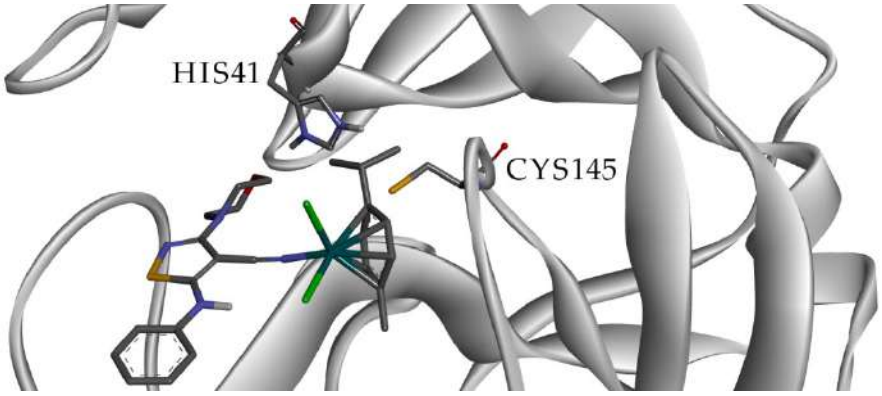


Figure 1. Best docking hit for compound 4 near catalytic dyad of M^{pro}.

4. Conclusions

Based on the results of docking experiments, isothiazole derivatives as ligands in piano stool ruthenium complexes show promising activity in *in silico* studies as

inhibitors of M^{pro}. Further modification of these ligands and their metal complexes may prove a fruitful endeavor during the search for treatment of the COVID-19.

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