In silico estimation of COX-2 and 5-LOX inhibitory potential of some novel thiourea derivatives of naproxen

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Abstract: Design of dual COX-2/5-LOX inhibitors can be considered an adequate approach in the development of new anti-inflammatory drugs with less pronounced side effects. The aim of the present research was to examine the binding potential of the seven newly designed thiourea derivatives of naproxen towards COX-2 and 5-LOX. The binding analysis of ligand conformations was performed by OEDocking 3.2.0.2 software. The binding potential assessment revealed that thiourea derivatives of naproxen exhibited a comparable binding affinity as naproxen towards COX-2. The highest number of key binding interactions with 5-LOX was formed by compound **5**, whereas compound **6** established the most stable complex (-9.29 kcal/mol). According to the obtained results, derivatives **5** and **6** can be considered as dual COX-2/5-LOX inhibitors with potential anti-inflammatory activity. However, none of the investigated compounds were able to form three hydrogen bonds with the binding site of COX-2, as well as three key hydrogen bonds with the active site of 5-LOX.

Keywords: naproxen, thiourea, OEDocking, COX-2, 5-LOX

1. Introduction

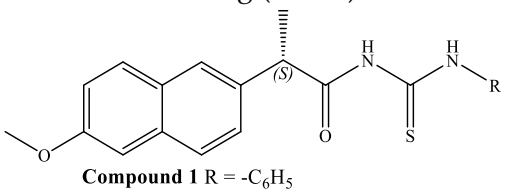
Naproxen and other aroyl propionic acid derivatives cause significant gastrointestinal toxicity due to non-selective inhibition of the cyclooxygenase enzymes (COX-1 and COX-2), so it is important to mask the free carboxyl group of the parent drug [1]. 5-lipoxygenase (5-LOX) is also involved in the metabolism of prostaglandins and catalysis of lipid peroxidation by producing lipid peroxides that lead to inflammatory response [2]. Therefore, the design of dual COX-2/5-LOX inhibitors can be considered an adequate

approach in the development of new anti-inflammatory drugs with less pronounced side effects.

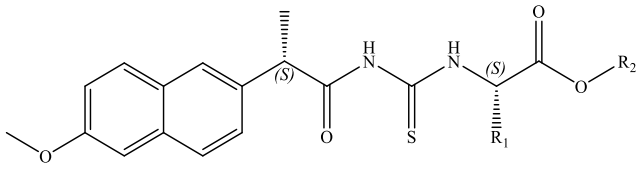
Encouraged by the results of previously published research [3], we decided to design seven new thiourea derivatives of naproxen with aromatic amines and amino acids. The aim of this research was to use a molecular docking analysis to examine the binding potential of these molecules towards COX-2 and 5-LOX.

2. Materials and Methods

Crystal structures obtained from the Protein Data Bank (http://www.rcsb.org/) were 3NT1 (naproxen bound to COX-2) and 6N2W (NDGA bound to 5-LOX). For the preparation of target enzymes MAKE Receptor 3.2.0.2 software was used [4]. The multiconformer data sets for each tested compound were prepared using OMEGA 2.5.1.4 [5,6]. Then, the binding analysis of ligand conformations into the active sites of target enzymes was performed by OEDocking 3.2.0.2 software [7–9] with the fast rigid exhaustive docking (FRED) tool.



Compound 1 R = -C_6H_5
Compound 2 R = -C_6H_4-CH_3-*p*
Compound 3 R = -C_6H_4-CH_3-*o*
Compound 4 R = -C_6H_4-OCH_3-*o*
Compound 5 R = -C_6H_4-F-*o*



Compound 6 R_1 = -CH_2-C_6H_5; R_2 = -CH_3
Compound 7 R_1 = -CH_2-(1*H*-indol-3-yl); R_2 = -CH_3

Figure 1. Structures of tested compounds.

3. Results and Discussion

Carbonyl and hydroxyl oxygen atoms of naproxen's carboxyl group interact as proton acceptors with a guanidine group of residue ARG120. Also, residue TYR355 with its phenolic group as hydrogen bond donor establishes hydrogen bond with carbonyl oxygen atom of the naproxen's carboxyl group. It can be observed that carbonyl and thiocarbonyl groups of aromatic amine derivatives **1**-**3**, and **5** (proton acceptors) establish a double hydrogen bond with residue ARG120 (Figure 2A). Carbonyl oxygen atom of derivative **4** (hydrogen bond acceptor) forms a double hydrogen bond with residue ARG120, whereas methoxy group attached to benzene ring in the side chain of the compound is involved in the formation of an additional hydrogen bond (proton acceptor) with ARG120. The phenylalanine methyl ester derivative (**6**) via thiocarbonyl group forms a single hydrogen bond (proton acceptor) with residue TYR355, while the thiourea nitrogen atom of tryptophan methyl ester derivative (**7**) as proton acceptor establishes a hydrogen bond with TYR355.

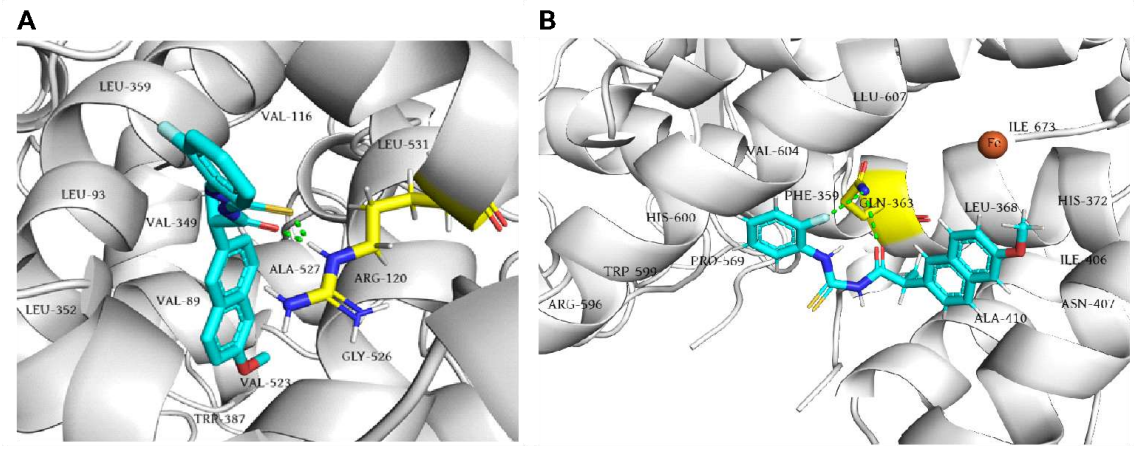


Figure 2. Molecular docking of compound **5** into the COX-2 (**A**) and 5-LOX (**B**) enzymes.

Taking into consideration the docking scores of aromatic amine derivatives (**1**: -14.53 kcal/mol; **2**: -14.57 kcal/mol; **3**: -14.27 kcal/mol; **4**: -12.77 kcal/mol; **5**: -14.62 kcal/mol), it can be observed that their values are lower but comparable to naproxen (-13.13 kcal/mol), with the exception of derivative **4**. Comparable docking score can be associated with the ability of aromatic rings of the tested compound's side chains to form additional binding interactions, whereby methoxynaphthalene core of these derivatives was docked into the active site of COX-2 in a similar fashion to naproxen. In contrast, ester derivatives have significantly higher docking scores (**6**: -11.28 kcal/mol; **7**: -8.99 kcal/mol) in comparison to naproxen. Regardless of the hydrogen bond formed by derivatives **6** and **7** with TYR355, the occurrence of sterically unfavorable interactions with VAL116, and in particular with ARG120 affects the higher value of docking scores.

The results of this *in silico* research showed that none of the tested molecules have the ability to chelate the iron ion in the binding site of the 5-LOX enzyme, so they can be considered as potential competitive non-chelating inhibitors. It was observed that none of the tested molecules was able to achieve the key hydrogen interactions that the co-crystal NDGA forms with the residues ARG596, ILE673, and HIS600. Namely, all derivatives form one hydrogen bond via the carbonyl oxygen of the amide group with GLN363, except for derivatives **2** and **3**, which failed to form any hydrogen bond.

The highest number of key binding interactions was formed by compound **5** (14 interactions) (Figure 2B). Namely, the naphthalene nucleus of compound **5** forms a double π-alkyl type interaction with the ALA410 residue. The same structural component of the tested ligand established simultaneously one π-alkyl contact with the tertiary carbon and one π-σ contact with the methyl group of the LEU368 side chain. The benzene ring of o-fluoroaniline formed a double π-π interaction with the π-electrons of the PHE359 and TRP599 aromatic cores. In terms of free binding energy values, all tested compounds (except derivatives **2** and **7**) achieved energies comparable to NDGA (-8.77 kcal/mol), but not to zileuton (-10.67 kcal/mol). This showed that molecules with the ability to chelate iron ion form more stable complexes with 5-LOX. Based on the free binding energy values, compound **6** established the most stable complex with the target

enzyme (-9.29 kcal/mol).

3. Conclusions

Molecular docking simulations suggest high binding potential of derivatives **5** and **6** towards COX-2 and 5-LOX. However, none of the investigated compounds were able to form three hydrogen bonds with ARG120 and TYR355 of COX-2, as well as three key hydrogen bonds with the active site of 5-LOX, like co-crystallized ligands.

Acknowledgment

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