RESEARCH ARTICLE

Study of Anti-inflammatory Activity of New Derivative of Indomethacin: A Computational Study

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Received: 17th June, 2022; Revised: 27th July, 2022; Accepted: 11th August, 2022; Available Online: 25th September, 2022

ABSTRACT

This article presents the synthesis of new derivative from thiodaizole, based on the reaction of Thiosemicarbazide with Indomethacin in the presence of Phosphorus (V) oxychloride as catalyst. COX enzyme is divided to two isoforms COX-1 and COX-2 with similar structure and high sequence identity. The novel Inhibitor used a thiodaizole ring in combination with Indomethacin to reduce the risk of analogs interfering with COX-1's small hydrophobic tunnel. Furthermore, the absence of a carboxylic group in the new Inhibitor reduces the Inhibitor's ability to form a salt bridge with Arg120 (Figure 2), preventing the inhibition of the COX-1 enzyme. The aim of this study is to investigate the Anti-inflammatory activity of Indomethacin against the human second isoform of prostaglandin synthase (cyclooxygenase, COX-2). COX-2 enzyme 3D structures (PDB ID: 1CVU) were extracted from the protein databank. The PDB format of Indomethacin and its derivative were prepared by use Discovery Studio 2016 Client. Molecular docking study appeared that the new derivative exhibited better binding energy (-10.9 kcal/mol) compared with Indomethacin that have binding energy (-10.09 kcal/mol) and it's have high selectivity towered COX-2 enzyme in the other hand In the present study, the predicted Pharmacokinetic (PK) values of Inhibitor (1) and Indomethacin (NSAID) deduce that Inhibitor (1) satisfies all PK parameters and has qualified as best lead candidate as an Anti-inflammatory agent compared to Indomethacin.

Keywords: Anti-inflammatory, Cyclooxygenase-2, Indomethacin, Molecular docking.

International Journal of Drug Delivery Technology (2022); DOI: 10.25258/ijddt.12.3.52

How to cite this article: Jaber, FA, Behget, SA, Shntaif, AH. Study of Anti-inflammatory Activity of New Derivative of Indomethacin: A Computational Study. International Journal of Drug Delivery Technology. 2022;12(3):1245-1248.

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

The non-steroidal Anti-inflammatory drugs (NSAIDs) in the treatment of rheumatoid arthritis, inflammation and pain by blocking the first step of the biosynthesis of prostaglandin from arachidonic acid through inhibition of cyclooxygenase (COX-2) activity.[1] NSAIDs are of great therapeutic benefit but gastrointestinal liabilities often limit it, including ulceration, gastrointestinal mucosal, bleeding and renal toxicity.[2] In recent years there is a great increase of inflammatory cases, leading to the design and development to production of safer and more active NSAIDs.[3] Computational chemistry, especially quantitative structure-activity relationship (QSAR) studies, have become an important part of the drug development process.[4] AutoDock tools, a widely distributed generic field molecular docking software, were used in this study. This software automatically processes the flexible docking of bonds into a known protein structure.[5] The default settings were used for all other parameters. Complex structure files were analyzed using DS visualization software. The AutoDock result gives a peptide's binding position and conformation and a rough estimate of its activity. In the present study, toxicity prediction data and Pharmacokinetic (PK) values of Indomethacin (NSAID) and synthesized derivative deduce that Inhibitor (1) is best as an Anti-inflammatory agent compared to Indomethacin.[6]

COMPUTATIONAL METHODS

Docking Studies

Ligand structures were generated using Chem Draw Professional 2016 as mol format and converted to 2D structures using DS visualization software and the ligands are saved in PDBQ format to identify torsions during docking.[7] From the Protein Data Bank, the crystal structures of COX-2 (PDB ID: 1CUV) were downloaded.[8] All non-standard ligands and residues were removed from the binding sites and the A chain of COX-2 was selected for docking studies and partial atomic charges were added to the protein structures and removed all water molecules from it.[9,10] Proteins are saved in (Pdb) format;

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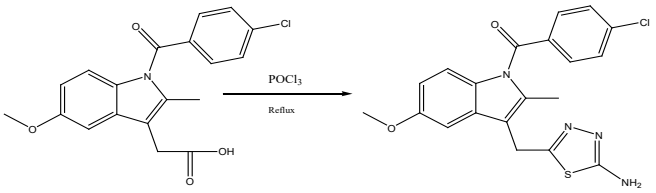


Scheme 1: Synthesis of target selective COX-2 inhibitor

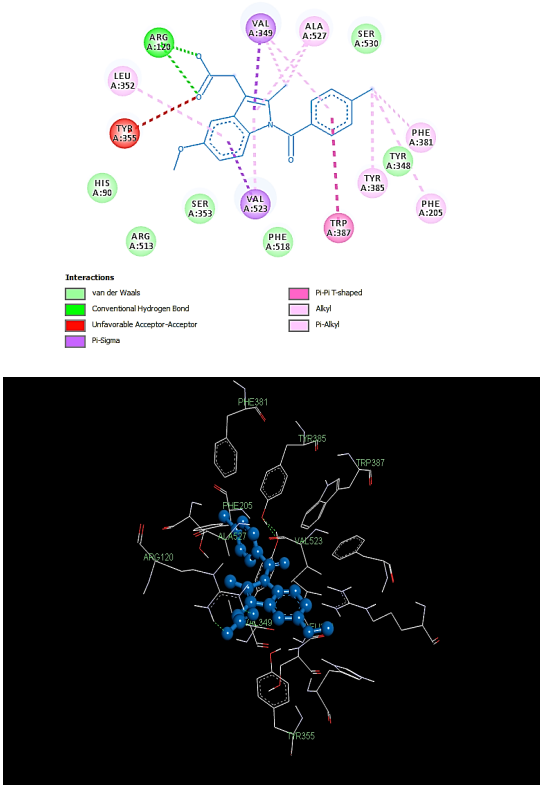


Figure 1: Predicted binding mode for Indomethacin with COX-2.

The atomic solubility coefficients were determined and finally converted into (Pdbqs) format.[11]

Docking Interactions

The Autodock Tools-1.5.6.Ink was used to analyze the formation of hydrogen bond and non-bonded between the ligand compound and the amino acid residues in the active site of COX-2. The docking area was defined using grid points of three-dimensional built-in x, y, and z affinity grid 48x54x62 A0. affinity grid with (0.375 Å) grid point spacing was placed around the active site of COX-2.2.[9]

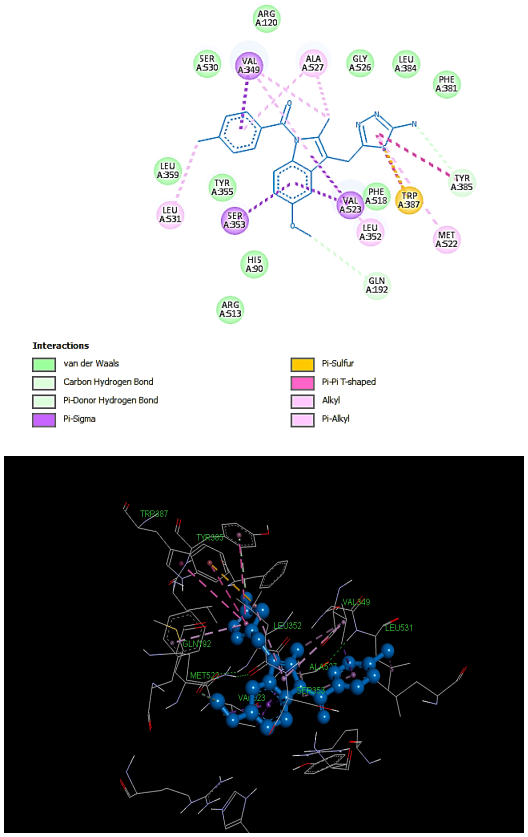
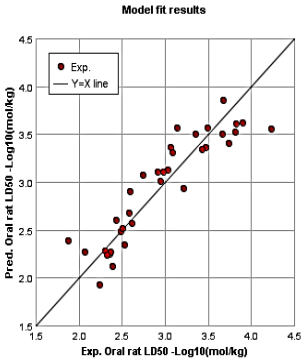


Figure 2: Predicted binding mode for inhibitor (1) with COX-2.

RESULTS AND DISCUSSION

The target COX-2 inhibitors (3-((5-amino-1,3,4-thiadiazol-2-yl) methyl)-5-methoxy-2-methyl-1H-indol-1-yl) (4-chlorophenyl) methanone was synthesized according to (Scheme 1) by reaction of Indomethacin (0.1mole) and thiosemicarbazide (0.1 mole), in (30 mL) $POCl_3$, was heated under reflux for (45 minutes), water (90 mL) was added and reaction mixture was

Parameter	Value
Endpoint	Oral rat LD_{50}
r^2	0.827
q^2	0.724
#chemicals	35
Model	FDA Model



Indomethacin Inhibitor (1)

Parameter	Value
Endpoint	Oral rat LD_{50}
r^2	0.769
q^2	0.601
#chemicals	35
Model	FDA Model

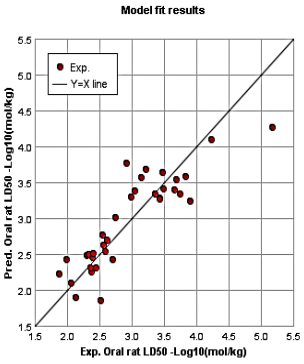


Figure 3: Statistical analysis and graph for indomethacin and inhibitor (1) prediction (data obtained from T.E.S.T. software)

Table 1: LD_{50} values Prediction in rats by T.E.S.T (Consensus method)

Generic name	Software estimation by T.E.S.T (Consensus method)		Statistical data validation by T.E.S.T
	Predicted LD_{50} Values (mg/kg)	Log LD_{50} value (mg/kg)	R^2 value#
Indomethacin	3.36	155.38	0.827
Inhibitor (1)	2.69	839.24	0.769

reflux for another 4 hour, on completion of reaction (TLC), cool to room temperature and poured in ice-cold water and basified (pH 8) with potassium hydroxide solution. The solid was filtered, washed with water, dried and crystallized from Ethanol.[1]

Yield: 90%, m.p.: 235°C. IR (KBr) υ max/cm^{-1} : 3487.06, 3348.19 (NH_2), 3093.6 ($CH_{aromatic}$), 2931.60, 2831.31 ($CH_{aliphatic}$) and 1681.81 (CO). ^{1}HNMR (δ ppm), (DMSO-d6 MHz),: d 2.509 (s, 3H, CH_3), 3.492 (s, 2H, CH_2), 3.751 (s, 3H, O-CH_3) 5.234 (s, 2H, NH_2), 6.663–7.653 (m,7H,$CH_{aromatic}$). ^{13}C-NMR(δ ppm), (DMSO-d6, MHz): 13.02, 32.06, 55.75, 102.4, 111.5, 114.09, 116.2, 129.4, 130.6, 131.5, 131.6, 134.6, 134.8, 137.04, 155.9, 168.2, 173.1.

Docking Interactions of COX-2

Docking interaction of Indomethacin, an amorphous ligand, with human (COX-2) yielded a best binding energy that is equal to 10.06 kcal/mol. This interaction is preferred by the formation of H-bond with the guanidinium group of Arg120 and hydrophobic interactions with Val349, Val523, Leu352, Ala527, Trp387, Tyr385, Phe205 and Phe381 (Figure 1). Inhibitor (1) docking reactions were observed with a binding energy of 10.9 kcal/mol. This interaction is supported by the formation of H bonds with (Gln192, Tyr385) and hydrophobic interactions with (Leu531, Val347, Ala527, tyr385, Met522, Trp387, Leu352, Val523, and Ser353) (Figure 2). Figure 1 shows complex docking and interaction graphs of crystallized, indomethacin and inhibitor (1) ligands with human COX-2 binding interactions. The amino acid Arg120 in human COX-2 was found to interact with the crystallized ligand indomethacin. While the amino acids Gln192 and Tyr385 are essential for interactions with the inhibitor (1), the interactions of the inhibitor (1) may explain the highest binding free energy and anti-inflammatory activity.

QSAR Modeling by Using T.E.S.T. Software

To calculate the LD50 values of Indomethacin and its derivative (Inhibitor (1)), the QSAR modeling software package (T.E.S.T. 4.1) developed by US EPA) was utilized. T.E.S.T. software was used to determine the intense toxicity prediction data in Table 1. According to the Canadian Center for Occupational Health (CCOH) and Safety and Ruiz *et al.,* 2012,[15, 16] toxicity ranges in mg/kg of (5), (5–50), (50–500), (500–5.000), (5.000–15.000), (5.000–15.000), (5.000–15.000), (5.000–15.000), and >(15.000) are super toxic, extremely toxic, very toxic, respectively have determined.

Table 2: Pharmacokinetic (PK) Properties for Indomethacin and Inhibitor (1)

Compound	*c log P*	*TPSA Ao*	*log BB*	*GI absorption*	*log S*
Indomethacin	2.76	68.63	+BBB	High	-4.86
inhibitor (1)	2.97	111.27	-BBB	High	-5.66

Pharmacokinetic Properties

In Table 2 the pharmacokinetic properties like (c log P), TPSA, blood brain barrier (log BB), GI absorption, and water solubility (log S) using Swiss ADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules.

CONCLUSION

This study includes the synthesis of novel (3-((5-amino-1,3,4-thiadiazol-2-yl)methyl)-5-methoxy-2-methyl-1H-indol-1-yl) (4-chlorophenyl) methanone to obtain safer and potent Anti-inflammatory agent. the newly synthesized derivative was docked into the COX-2 and the molecular docking study provided the interpretation of the biological activities of the active compound compared to the reference drug indomethacin. The present predicted intense toxicity results with special reference to LD_{50} values of rat orally were obtained that Inhibitor (1) is moderately toxic then Indomethacin (Figure 3).

REFERENCES

1. Betül K, Begüm N, Derya O, Serkan L, Ulviye A, Abdullah B, Yusuf Ö, Zafer A (2018) Synthesis and Biological Evaluation of New Thiosemicarbazone Derivative Schiff Bases as Monoamine Oxidase Inhibitory Agents. Molecules, 23(1): 60-78.
2. Shaymaa E, Mohammed A, Hamed I, Mohamed M (2017) Discovery of new indomethacin-based analogs with potentially selective cyclooxygenase-2 inhibition and observed diminishing to PGE2 activities. European Journal of Medicinal Chemistry, 141(2017): 306-321.
3. Menyhart-Botond S (2018) Binding of indomethacin methyl ester to cyclooxygenase-2. A computational study. Journal of Molecular Modeling, 24(7): 150 1-8.
4. John R, Cesar L, Yhors, C (2016) The health benefits of natural skin UVA photoprotective compounds found in botanical sources. Int J Pharm Pharm Sci., 8(3): 13-23.
5. Walaa S, Neama M, Emad K, Khaled M, Mounier M (2016) Synthesis, Biological Evaluation and Docking Analysis of Some Novel Quinazolin Derivatives as Antitumor Agents. Iran J Pharm Res, 15 (1): 179–196.
6. Anjugam C, Sridevi M, Gnanendra T (2018) Structure-Based Docking Studies Toward Exploring The Potential Anticancer Activity Of Morin Against Non-Melanoma Skin Cancer Therapeutic Drug Targets. Asian J Pharm Clin Res., 11(4): 61-66.
7. Muthusamy C, Thirunalasundari T (2014) Identification Of A Lead Molecule For Inflammation By Molecular Mechanics. International Journal Of Biological & Pharmaceutical Research, 5 (4): 378-382.
8. Asha B, Rabindra K, Lata P, Piyush A, Kishori G (2013) Synthesis, Biological Activity, Molecular Modelling Studies and 3D-QSAR Investigations of N-[2-(aryl/substituted aryl)-4-oxo-1, 3-thiazolidin-3-yl] pyridine-4-carboxamides. The Open Conference Proceedings Journal, 4 (13): 99-112.

9. Navya A, Nanda K, Hari P, Santhrani T, Uma P (2012) In vivo and in silico Analysis Divulges the Anti Inflammatory Activity of α-Mangostin. International Journal of Applied Biotechnology and Biochemistry, 2 (1): 69-80.
10. Rao P, Knaus E (2008) Evolution of non-steroidal Anti-inflammatory drugs (NSAIDs): cyclooxygenase (COX) inhibition and beyond. J. Pharm. Sci., 11(2): 81-110.
11. Tagreed N, Monther F, May M, Zainab B (2018) Synthesize of New Ibuprofen and Naproxen Sulphonamide Conjugate with Anti-Inflammatory Study and Molecular Docking Study. International Journal of Pharmaceutical Quality Assurance, 9 (2): 102-108.
12. Durrant J, Mccammon J (2010) Computer-aided drug-discovery techniques that account for receptor flexibility. Curr Opin Pharmacol, 10 (6): 770-4.
13. Ming Y, Yi J, Li-Png Y (2018) Screening for cyloxygenase 2 inhbitors from natural compounds of Radix Glycrhizae using computer simulation. Traditonal Medicne Rsearch, 3 (3): 115-130.
14. Ravi J, Kiran G, Sarangapani M, Sriram R (2015) Synthesis, In Vivo Anti-Inflammatory Activity, and Molecular Docking Studies of New Isatin Derivatives. International Journal of Medicinal Chemistry, 6(5): 243-248.
15. Canadian Center for Occupational Health & Safety, What is an LD50 and LC50, (2012), Available from: http://www.ccohs.ca/oshanswers/chemicals/LD50.html#_1_6.
16. Ruiz P, Begluitti G, Tincher T, Wheeler J, Mumtaz M (2012) Prediction of acute mammalian toxicity using QSAR methods: A case study of sulfur mustard and its breakdown products, Molecules, 17 (8): 8982-9001.