Ind. J. Pharm. Edu. Res., 2025; 59(1s):s312-s322.
https://www.ijper.org

Original Article

Unlocking Anti-Inflammatory Potential: Virtual Discovery and ADMET Evaluation of Novel Pyrazole-Based COX-II Inhibitors

Vipul M. Patil*, Harinath N. More

Department of Pharmaceutical Chemistry, Bharati Vidyapeeth College of Pharmacy, Near Chitranagari, Morewadi, Kolhapur, Maharashtra, INDIA.

ABSTRACT

Background: Inflammation, a complex biological process mediated by arachidonic acid metabolites, plays a crucial role in various diseases like arthritis, psoriasis and neurodegenerative disorders. The Cyclooxygenase (COX) pathway, particularly COX-2, is a well-established anti-inflammatory target. **Materials and Methods:** This study aimed to discover and evaluate novel pyrazole derivatives as potential COX-2 inhibitors via virtual screening. The 3D crystal structure of Cyclooxygenase-II (PDB: 1CX2) was prepared and optimized for in-silico investigations. Molecular docking analysis using AutoDock Vina assessed ligand-protein interactions, guided by CASTp3.0-predicted binding sites. Ligands were energy minimized and docked against the protein and drug-likeness/synthetic accessibility was predicted using SwissADME and pkCSM. Biological activity and medicinal chemistry were assessed using network diagrams, Bioavailability Radar and BOILED-Egg model for absorption and brain penetration prediction. This integrated approach facilitates the identification of potential COX-2 inhibitors with favorable pharmacokinetic profiles for further development. **Results:** Through molecular docking, three compounds (D202, D305 and F505) exhibited the highest binding affinity for COX-2, surpassing the native ligand's residual binding. Subsequent ADMET prediction revealed promising pharmacokinetic properties, including significant oral bioavailability scores (0.55) and synthetic feasibility scores ranging from 3.05 to 3.74. **Conclusion:** These findings suggest the potential of these pyrazole derivatives as promising lead candidates for further development as novel anti-inflammatory agents.

Keywords: Inflammation, COX 2, Molecular docking, ADMET, Pyrazole.

Correspondence:
Vipul M. Patil
Department of Pharmaceutical Chemistry, Bharati Vidyapeeth College of Pharmacy, Near Chitranagari, Morewadi, Kolhapur-416013, Maharashtra, INDIA.
Email: vipulpatil1230@gmail.com
ORCID: 0000-0002-1443-8797

Received: 18-02-2024;
Revised: 22-04-2024;
Accepted: 05-01-2025.

INTRODUCTION

Usually, infectious agents like bacteria, fungi, or viruses enter the body, lodge in the tissues, or move through the bloodstream. This results in inflammation.[1,2] Various biological events like tissue injury, ischemia, cancer and degradation may be responsible for the inflammation.[3] Mostly, in the formation of inflammation there is involvement of both adaptive and innate immune response. Inflammation is a vital part of the immune system's response in contradiction to a hostile world.[2] Many immune system diseases are associated with inflammation. Acute or chronic pain can be caused by many factors, including physical damage, chemicals, antibiotics and pathogenic microbial infections. Nonsteroidal Anti-Inflammatory Drugs (NSAIDs) have long been used to treat pain.[4] They are much more beneficial in treatment of different types of ailments caused due to inflammation such as rheumatoid arthritis, acute fever as well as relieving common daily pains.[5,6] The main enzyme involved in the process of prostaglandin synthesis is Cyclooxygenase (COX) or prostaglandin endoperoxide synthase. These two enzymes are mediators of pain, inflammation and increased body temperature (hyperthermia).[7] The mechanism of action involved in the activity of NSAIDs is to competitively inhibit the action of Cyclooxygenases (COX), which leads to the disruption of the biotransformation of downstream inflammatory mediators. Based on their distinct structures and functions, two functional COXs have been identified and defined as COX-1 and COX-2.[8] COX-1 is an intrinsic enzyme prevalent in a majority of cells. COX-1-catalyzed prostaglandins protect the gastrointestinal system. COX-2 is a triggered enzyme that generates a lot of prostaglandins, consequently it's regarded as a detrimental enzyme that promotes inflammation.[9]

Computational chemistry and biology provide useful tools to virtually confirm the binding pattern of ligands with target proteins.[10,11] Molecular docking is one of the widely used *In*

DOI: 10.5530/ijper.20256457

silico tools providing accurate calculation of binding energies between protein-ligand complexes and is used to demonstrate the interaction between ligand molecules.[12] It is a time-saving computational methodology intended to carry out research to identify and investigate the binding behavior of novel drug candidates.[13] Therefore, molecular docking studies have been used to elucidate the structural requirements for ligand interactions with multiple anti-inflammatory drug targets. In the present study, molecular docking of several newly pyrazole derivatives against cyclooxygenase II to check the anti-inflammatory potential of selected compounds.

MATERIALS AND METHODS

Protein and ligand preparation

The 3D crystal structure of PDB:1CX2 of cyclooxygenase-II at 3.00 Å resolution was obtained from the RCSB Protein Database (https://www.rcsb.org/).[14] Charged protein structures are created by removing water molecules and adding previously bound ligand molecules and polar hydrogen atoms to specify the right ionization and tautomeric states of amino acid residues. The produced protein structure was then tested using the PROCHECK program to measure protein quality.[15] The binding pocket was estimated using the CASTp3.0 server.[16] These found active binding locations were used to determine the optimal ligand binding site in the target protein. ChemSketch was used to construct the compound structures, which were then imported into BIOVIA Discovery Studio (DS) 2020 for optimization. Hydrogen atoms were added and the compounds were subsequently converted into PDB file format for additional in-silico research. Ligand designing scheme is elaborated in Figure 1 and Table 1.

Molecular docking analysis

Molecular docking studies were conducted to examine the binding sites and interactions that take place between the chosen ligands and the target protein (PDB: 1CX2). Using the PyRx 0.8 AutoDock Vina package, molecular docking was carried out.[17] PyRx was used to import the SDF file format containing all ligands and the PDB file format including 1CX2 as a macromolecule. Afterwards, these ligands were utilized for molecular docking after being energy reduced and transformed to PDBQT format using the Open Babel plugin for PyRx. In the Vina workspace of PyRx, the grid box for PDB: 1CX2 was chosen to cover the binding site residues; its dimensions are X:110.746501541Å, Y:91.3735608101Å and Z:137.971333237Å. Its center is X:42.3332, Y:33.5901 and Z:36.0748. By default, the exhaustiveness was set to 8. For every ligand, nine distinct positions were anticipated in relation to the target protein. For every docked ligand, the optimal pose with the lowest binding affinity was also downloaded as a PDB. The BIOVIA Discovery Studio 2020 was used to analyze saved poses and visualize docking interactions.[18]

In silico prediction of drug-likeness and synthetic accessibility

Chemoinformatic techniques are now one of the most dynamic and frequently utilized approaches in pharmacokinetics (ADME) assessment, drug discovery and toxicity investigations. Quantitative computational methods can now be used to predict several Pharmacokinetic (PK) parameters.[19] Robust predictions are no worse than those obtained from in vitro experiments; an important advantage is that they require fewer techniques, resources and time. In addition to being critical, connections can be virtually filtered. The forecasts were made using the SwissADME web tool and pkCSM. Assessing the biological activity and medicinal chemistry of compounds using network diagrams.[20]

The Bioavailability Radar allows one to quickly assess a molecule's drug-like qualities. The pink section represents the ideal range for each property: solubility: log S, not more than 6, saturation: fraction of carbons in the SP3 hybridization not less than 0.25, flexibility: no more than 9 rotatable bonds, size: MW between 150 and 500 g/mol, polarity: TPSA between 20 and 130 Å and lipophilicity: XLOGP3 between -0.7 and +5.0. The Blood-Brain Barrier (BBB) and passive Human Gastrointestinal Absorption (HIA) are classified using the intrinsic graphical

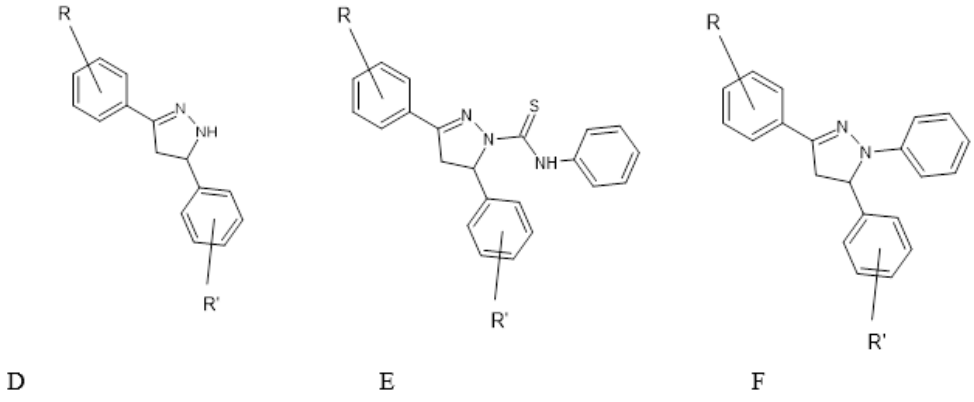


Figure 1: Designed pyrazole derivative D, E and F series.

Table 1: Designed pyrazole derivatives.

Code No.	R (Acetophenone sub.)	R′ (Benzaldehyde sub.)	Code No.	R (Acetophenone sub.)	R′ (Benzaldehyde sub.)
D/E/F101	-H	-H	D/E/F409	4-OH	4-NO_2
D/E/F102	-H	4-OCH_3	D/E/F410	4-OH	3-Cl
D/E/F103	-H	2-Cl	D/E/F411	4-OH	2-OH
D/E/F104	-H	4-Cl	D/E/F 501	3-NO_2	4-OCH_3
D/E/F105	-H	4-$N(CH_3)_2$	D/E/F 502	3-NO_2	2-Cl
D/E/F106	-H	3,4-dimethyl	D/E/F 503	3-NO_2	4-Cl
D/E/F107	-H	4-OH	D/E/F 504	3-NO_2	4-$N(CH_3)_2$
D/E/F108	-H	2-NO_2	D/E/F 505	3-NO_2	3,4-dimethyl
D/E/F109	-H	3-NO_2	D/E/F 506	3-NO_2	4-OH
D/E/F110	-H	4-NO_2	D/E/F 507	3-NO_2	2-NO_2
D/E/F111	-H	3-Cl	D/E/F 508	3-NO_2	3-NO_2
D/E/F112	-H	2-OH	D/E/F 509	3-NO_2	4-NO_2
D/E/F201	3-OH	-H	D/E/F 510	3-NO_2	3-Cl
D/E/F202	4-OH	-H	D/E/F 511	3-NO_2	2-OH
D/E/F203	3-NO_2	-H	D/E/F 601	4-NO_2	4-OCH_3
D/E/F204	4-NO_2	-H	D/E/F 602	4-NO_2	2-Cl
D/E205	2,4-dihydroxy	-H	D/E/F 603	4-NO_2	4-Cl
D/E206	2-NH_2	-H	D/E/F 604	4-NO_2	4-$N(CH_3)_2$
D/E207	3-NH_2	-H	D/E/F 605	4-NO_2	3,4-dimethyl
D/E208	4-NH_2	-H	D/E/F 606	4-NO_2	4-OH
D/E209	4-Br	-H	D/E/F 607	4-NO_2	2-NO_2
D/E/F301	3-OH	4-OCH_3	D/E/F 608	4-NO_2	3-NO_2
D/E/F302	3-OH	2-Cl	D/E/F 609	4-NO_2	4-NO_2
D/E/F303	3-OH	4-Cl	D/E/F 610	4-NO_2	3-Cl
D/E/F304	3-OH	4-$N(CH_3)_2$	D/E/F 611	4-NO_2	2-OH
D/E/F305	3-OH	3,4-dimethyl	D/E/F 701	4-Br	4-OCH_3
D/E/F306	3-OH	4-OH	D/E/F 702	4-Br	2-Cl
D/E/F307	3-OH	2-NO_2	D/E/F 703	4-Br	4-Cl
D/E/F308	3-OH	3-NO_2	D/E/F 704	4-Br	4-$N(CH_3)_2$
D/E/F309	3-OH	4-NO_2	D/E/F 705	4-Br	3,4-dimethyl
D/E/F310	3-OH	3-Cl	D/E/F 706	4-Br	4-OH
D/E/F311	3-OH	2-OH	D/E/F 707	4-Br	2-NO_2
D/E/F401	4-OH	4-OCH_3	D/E/F 708	4-Br	3-NO_2
D/E/F402	4-OH	2-Cl	D/E/F 709	4-Br	4-NO_2
D/E/F403	4-OH	4-Cl	D/E/F 710	4-Br	3-Cl
D/E/F404	4-OH	4-$N(CH_3)_2$	D/E/F 711	4-Br	2-OH
D/E/F405	4-OH	3,4-dimethyl	Total	D	76
D/E/F406	4-OH	4-OH		E	76
D/E/F407	4-OH	2-NO_2		F	71
D/E/F408	4-OH	3-NO_2		TOTAL	223

model BOILED-Egg, which is based on the molecules' locations in the WLOGP-versus-TPSA referential. The yellow area (yolk) represents the high probability of penetration into the brain, whereas the white area represents the high possibility of passive absorption through the gastrointestinal tract.[21]

RESULTS

Molecular docking analysis

In silico molecular docking method increases drug discovery efficiency and reduces the cost and time of experiment. Designed compounds were subjeted for molecular heating with the BIOVA Discovery Studio 2020. The identified protein structure was subjected to quality review and binding site analysis utilizing the PROCHECK and CASTp servers. Almost 76.3% of residues are located in the most preferred regions, depicted in Ramachandran plot as shown in Figure 2. A single pocket detected in a targeted protein (PDB: 1CX2) having an area of 13821.595 along with the volume of 26280.022 depicted in Figure 3.

Molecular docking is a widely used computer-based smart tool to predict interactions between proteins and ligands. In this study, molecular docking was performed using the AutoDock Vina plug-in from PyRx 0.8 software.[17-18] Virtually designed pyrazole derivatives were docked with Cyclooxygenase-II (PDB: 1CX2). Table 2 shows that the binding of the compound to the target protein varies in the range of -6.7 and -10.7 kcal per mol. D202 and D305 demonstrated the highest binding affinities of -10.5 and -10.7 kcal per mol, respectively, out of all the ligands. BIOVIA Discovery Studio was used to visualize the thorough interaction the protein and ligand with the best possible conformation. The interaction between residues D202 and D305 and 1CX2 shows the highest interaction highlighted in the Figures 4A and B. PDB: 1CX2 receptor amino acid residues (PHE200, GLN203, LEU390, THR206, TRP387, TYR38, PHE210, LEU391, HIS207, LEU408, PHE404, HIS388, VAL444, ALA202, VAL295, PHE395 and PHE407) interact with D305, which forms conventional H bonds, Π-Alkyl, Π-Donor and Van der Waals interations. The interacting types and residues involved between 1CX2 and the docked compounds are represented in Table 2.

In silico prediction of drug-likeness and synthetic accessibility

The drug likeness and synthetic accessibility parameters of best fit docked molecules are depicted in Table 3. This is also present in PAINS (Pan-Assay Interferences) alerts along with synthetic accessibility of medicinal properties. There are no alerts (PAINS warnings=0) in any of the considered connections. Synthetic accessibility, ranges from 1-10, with 1-4 indicating easy synthesis,

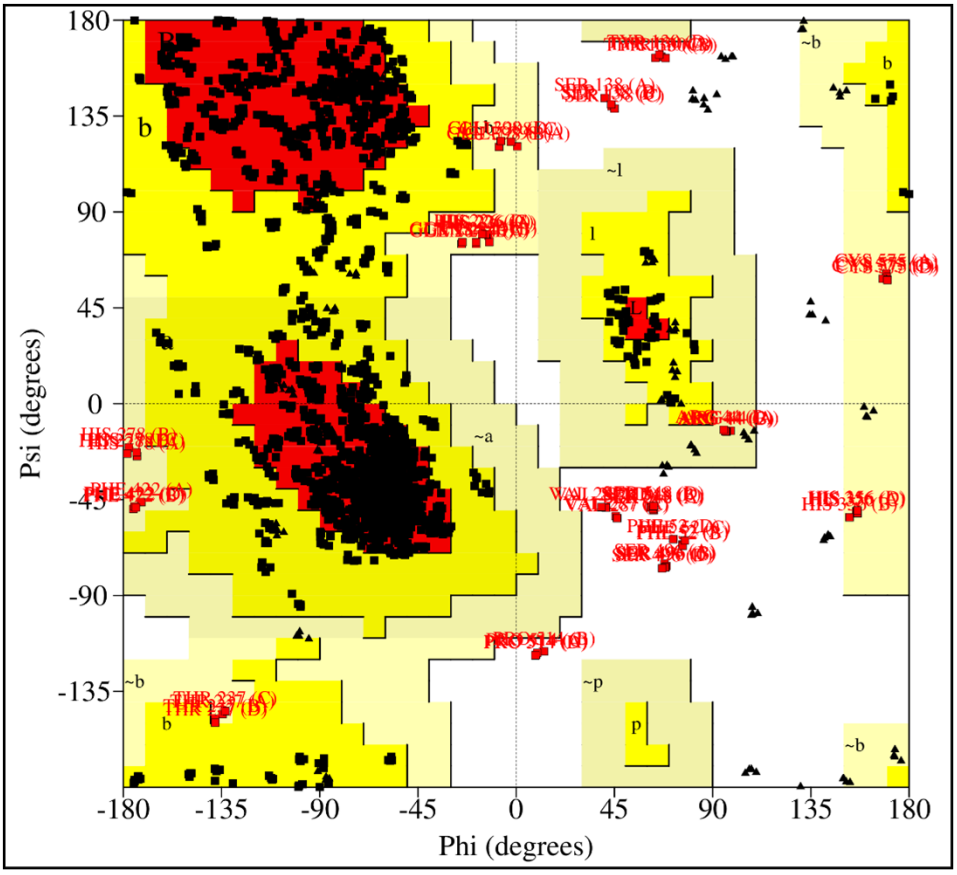


Figure 2: The Ramachandran plot for cyclooxygenase (PDB ID: 1CX2) shows a favored region with 76.3% residues.

4-7 indicating moderate and 8-10 indicating challenging to synthesize. All these molecules reside in very simple-to-synthesize range. In addition, Figure 5 shows Bioavailability Radar provides the information about drug-likeness of a molecule and Boiled-Egg describes the penetration of active compounds into the Blood-Brain Barrier (BBB) or Human Gastrointestinal Absorption (HIA). All compounds investigated had good physicochemical properties as their expected values were within the limits.

The synthesis access score ranges from 01 (very easy) to 10 (very difficult) based on 1024 Fragment contributions (FP2), adjusted for complexity and size penalty, as defined by the Swiss Institute of Bioinformatics, SwissADME.

DISCUSSION

Molecular docking analysis

The utilization of in-silico molecular docking methods in drug discovery offers notable advantages, including enhanced efficiency and reduced experimental costs and time. In this study, designed compounds were prepared for molecular docking using BIOVA Discovery Studio 2020. The selected protein structure, Cyclooxygenase-II (COX-II), underwent quality evaluation and binding site analysis through PROCHECK and CASTp servers, respectively. Analysis revealed a single pocket in the COX-II protein (PDB: 1CX2) with specific dimensions.

Molecular docking, a widely employed in-silico technique, was performed using the AutoDock Vina plugin within PyRx 0.8 software. The virtually designed pyrazole derivatives were docked against COX-II and their binding affinities ranged from -6.7 to -10.7 kcal/mol. Notably, compounds D202 and D305 exhibited the highest binding affinities, indicating strong interactions with COX-II.

Further analysis using BIOVA Discovery Studio allowed for detailed visualization of the interactions between the protein and ligands, with particular focus on compounds D202 and D305. It was discovered that compound D305 interacted with amino acid residues identified in the COX-II protein, including GLN203, LEU390, LEU391, LEU408, THR206, TYR38, PHE210, PHE200, PHE404, PHE395, PHE407, VAL444, VAL295, ALA202, HIS388, HIS207 and TRP387. These interactions involved conventional hydrogen bonds, van der waals forces, Π-alkyl and Π-donor interactions.

Overall, the results suggest that compounds D202 and D305 have strong binding affinities for COX-II, potentially making them promising candidates for further investigation as anti-inflammatory agents. The precise understanding of the interactions between these compounds and COX-II provides valuable insights for future drug design and development efforts targeting inflammation-related disorders.

In silico prediction of drug-likeness and synthetic accessibility

The analysis of the predicted drug-likeness and medicinal chemistry characteristics of the developed compounds, reveals promising attributes for further drug development endeavors. Notably, none of the considered compounds exhibited alerts for Pan-Assay Interference (PAINS), indicating a lack of potential

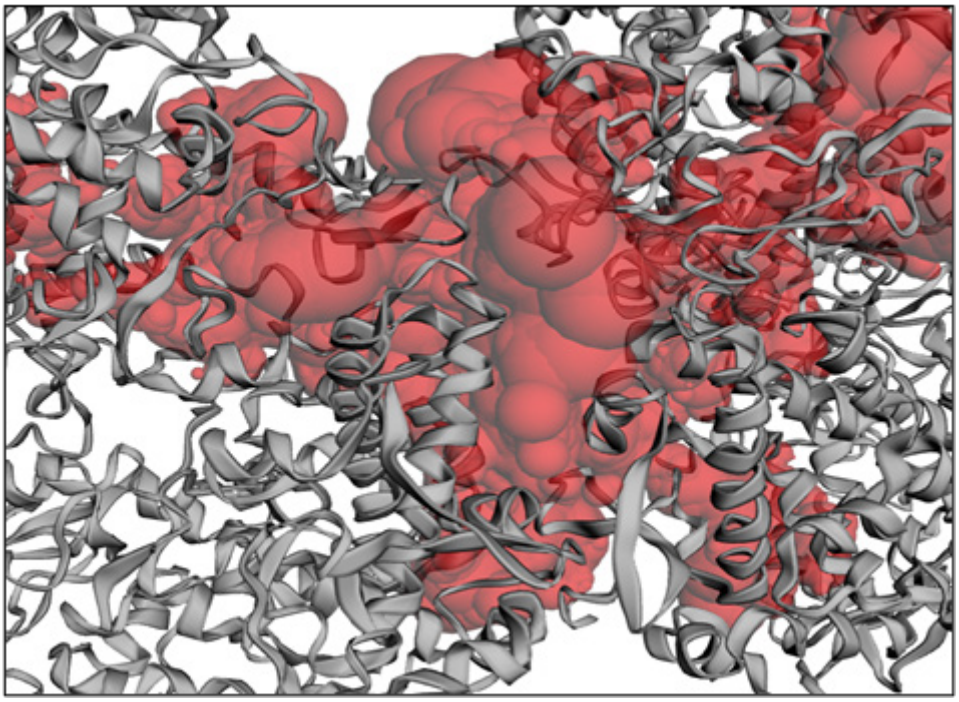


Figure 3: Binding pocket (red) is present in 1CX2.

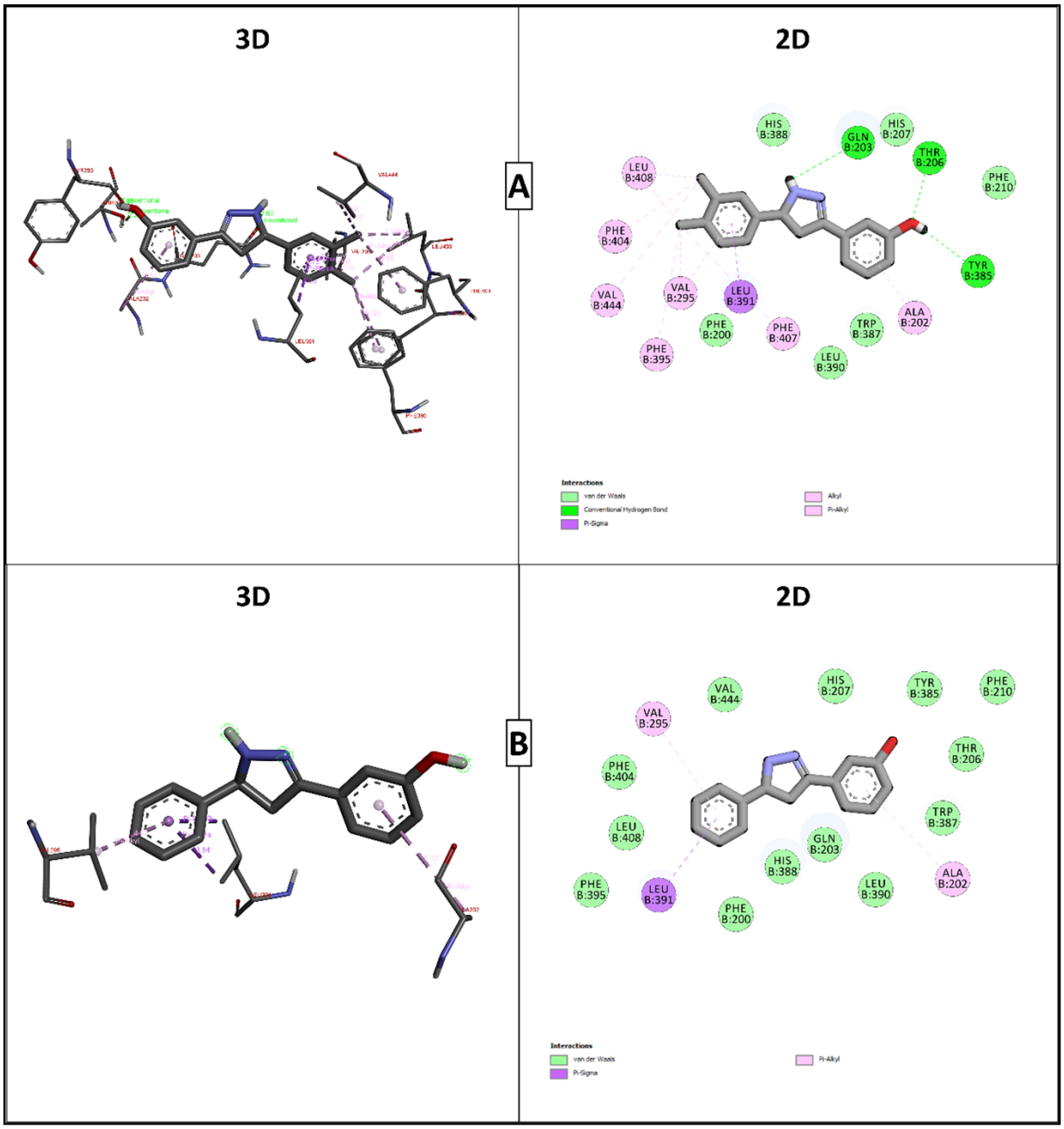


Figure 4: 3-Dimentional and 2-Dimentional interaction for A) D305 and B) D202 with Cyclooxygenase-II (PDB: 1CX2).

interference in biochemical assays commonly used in drug discovery research.

Moreover, the evaluation of synthetic accessibility scores indicates that all compounds fall within the range of very simple-to-synthesize molecules (scores of 1-4), suggesting feasibility in synthesis processes. This aspect is crucial for expediting the drug development pipeline by facilitating efficient and cost-effective synthesis routes. Additionally, assessment using the Bioavailability Radar and BOILED-egg models underscores favorable physicochemical properties of the investigated compounds. The absence of alerts in PAINS warnings, combined with easy synthetic accessibility and good physicochemical properties, bodes well for the drug-likeness and potential efficacy of these compounds.

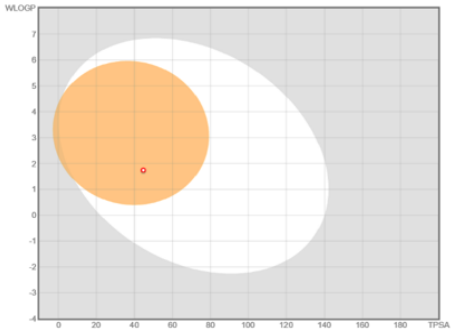
In conclusion, the comprehensive assessment of medicinal chemistry properties indicates that the developed compounds possess desirable characteristics for further exploration as potential drug candidates. These findings lay a solid foundation for future studies aimed at optimizing these compounds for therapeutic applications, particularly in the treatment of inflammation-related disorders.

Table 2: Ligand binding affinity for Cyclooxygenase-II (PDB: 1CX2) as well as interacting residues and interaction types.

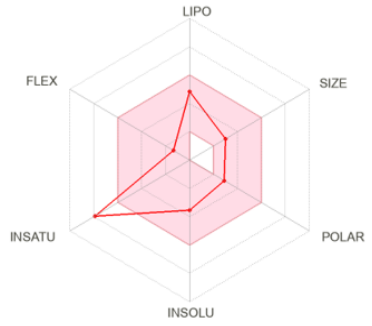
Molecule No.	Binding affinity (kcal/mol)	Conventional H-bond Interaction	Pi Sigma Interaction	Pi Alkyl Interaction	Van-der-Waals Interaction	Other Interactions
D104	-10.1	-	LEU391	VAL295, VAL444, HIS388, ALA202.	PHE200, GLN203, HIS207, THR206, TYR385, TRP387, ALA199, LEU390, VAL447, PHE404, LEU408, PHE395.	-
D106	-10.3	-	LEU391	PHE407, PHE200, PHE395, VAL295, LEU408, PHE404, VAL444, ALA202	LEU390, TRP387, TYR385, THR206, HIS207, GLN203, HIS388	-
D109	-10.2	HIS388	LEU391	VAL295, LEU390.	ALA199, THR206, GLN203, TYR385, HIS386, VAL444, PHE404, LEU408, PHE395.	C-H Bond: HIS207 π-π Stacked: TRP387 Amide π Stacked: ALA202.
D202	-10.5	-	LEU391	ALA202, VAL295.	VAL444, HIS207, TYR385, PHE210, THR206, TRP387, LEU390, GLN203, HIS388, PHE200, PHE395, LEU408, PHE404	-
D206	-10.1	TRP387, GLN203.	LEU391	VAL295, ALA202.	HIS207, THR206, TYR385, LEU390, HIS388, ALA199, VAL444, PHE404, PHE395, LEU408, PHE200.	-
D305	-10.7	GLN203, TYR385, THR206	LEU391	LEU408, ALA202, PHE404, VAL444, PHE395, VAL295, PHE407.	PHE200, HIS207, PHE210, TRP387, HIS388, LEU390.	-
D310	-10	THR206, TYR385, GLN203.	LEU391	ALA202, VAL295, PHE404, VAL444, LEU408.	ALA199, LEU390, HIS388, PHE200, PHE395, HIS207, HIS386, PHE210, TRP387.	-

Molecule No.	Binding affinity (kcal/mol)	Conventional H-bond Interaction	Pi Sigma Interaction	Pi Alkyl Interaction	Van-der-Waals Interaction	Other Interactions
E505	-10.1	GLY536, ASN375.	LEU145	LEU145	HIS226, GLY225, TRP139, GLY227, GLN374, GLN374, ARG376, ASN375, HIS226, LEU224, ARG376, GLY225, PHE142, SER143.	Carbon hydrogen Bond: ASN537, PRO538 Pi-Pi T Shaped: PHE142.
E604	-10.1	GLY536	LEU145	-	LEU145, TRP139, ARG376, GLN374, ASN375, ASN537, GLY536, PHE142, GLN374, ASN537, TRP139, GLY227, ASP229, HIS226, VAL228, ARG376, SER143, GLY225	Carbon hydrogen Bond: PRO538, ASN375 Pi-Pi T Shaped: PHE142.
E608	-10.1	SER126, GLN372, TYR122, LYS546.	-	PRO542, ARG61, LYS532.	GLN370, PHE367, SER121, ILE124, PHE371, ARG44, THR62, THR60, GLN543, PRO127, SER541, TYR373.	-
E609	-10	LYS468, LEU472, ASN39, ASN43.	-	LEU152, ARG469.	SER471, THR70, GLY63, LEU80, ARG44, GLN42, CYS41, PRO153.	Pi-Pi T Shaped: PHE64 Pi-Cation: GLU465.
F202	-10.1	GLN543	-	PRO542	ARG61, ILE124, SER126, SER541, PRO127, TYR373, GLN370, LYS532, PHE371, THR118, TYR122.	Amide Pi Stacked: SER121.
F308	-10.3	TYR130, ASN34, ALA156.	-	PRO153, CYS36, MET48.	ARG44, GLY45, ASN39, GLN461, CYS37, VAL155, PRO154, GLY135, TYR136, TRP323, GLN327, SER548, THR549, CYS47.	Carbon hydrogen Bond: LYS137 Pi-Cation: GLU46.
F505	-10.4	ASN39, TYR130.	-	LYS137, PRO153, TYR136, TYR136, MET48.	CYS41, LEU152, GLU465, GLY135, GLN327, ALA156, ASN34, THR549, SER548, GLU46, ARG44.	Carbon hydrogen Bond: CYS47.

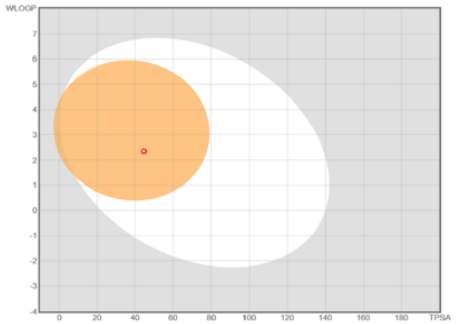
Molecule No.	Binding affinity (kcal/mol)	Conventional H-bond Interaction	Pi Sigma Interaction	Pi Alkyl Interaction	Van-der-Waals Interaction	Other Interactions
F508	-10	CYS159, ASN34, ALA156, GLN327.	-	PRO153, CYS36, LYS137.	GLY135, TYR130, GLU46, TRP3233, SER548, GLY551, SER138, LEU334, THR381, LYS47, TYR136, GLN461, ASN39, VAL155, CYS37.	Carbon hydrogen Bond: THR549 Halogen: MET48.



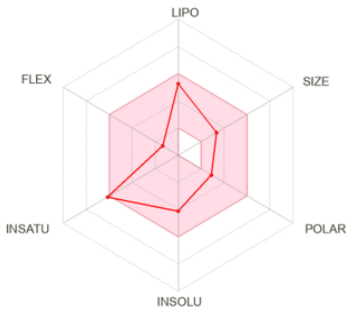
Boiled Egg representation of D202



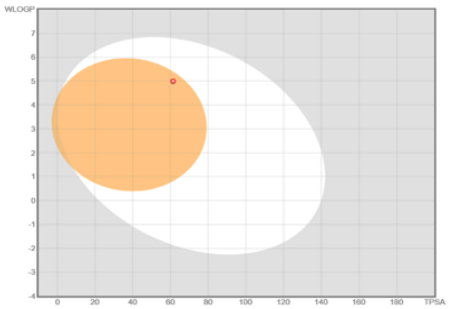
Bioavailability Radar representation of D202



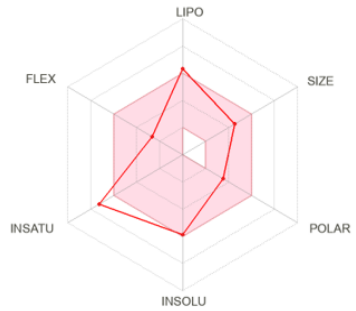
Boiled Egg representation of D305



Bioavailability Radar representation of D305



Boiled Egg representation of F505



Bioavailability Radar representation of F505

Figure 5: Boiled Egg representations with Bioavailability Radar images of the best docked molecule showing their physicochemical behavior.

Table 3: Drug likeness and synthetic accessibility properties of these best fit docked molecules.

Parameters		Molecule No.		
		D202	D305	F505
Drug Likeness	Lipinski	✓	✓	✓
	Egan	✓	✓	✓
	Veber	✓	✓	✓
	Ghose	✓	✓	✓
	Muegge	✓	✓	✓
	Bioavailability Score	0.55	0.55	0.55
Synthesis	PAINS Alert	0	0	0
	Synthetic Accessibility	✓	✓	✓
	Synthetic Score* (From 01 to 10)	3.05	3.27	3.74

Tickmark (✓) indicates the compounds showing drug likeliness and synthetic accessibility. *The synthesis access score ranges from 01 (very easy) to 10 (very difficult) based on 1024 fragment contributions (FP2), adjusted for complexity and size penalty, as defined by the Swiss Institute of Bioinformatics, SwissADME.

CONCLUSION

Inflammation is a vibrant part of the immune system's response in contradiction of a hostile world. PyRx is used to run molecular docking simulations, which are extremely beneficial in predicting and validating the type of ligand binding to cyclooxygenase-II proteins as an anti-inflammatory drug. The designed compounds are *in silico* screened against cyclooxygenase-II (PDB: 1CX2). The results of molecular docking show that all selected ligands have a considerable binding affinity for cyclooxygenase-II (PDB: 1CX2). Among all the virtually screened ligands D202, D305 and F505 were the three with the highest binding affinity for the targeted protein with anti-inflammatory activity, which binds to the residual binding as native ligands. In addition, the ADMET/PK predictors of these (three) active compounds were investigated and found to be orally bioavailable. Also it has therapeutic value as a treatment of inflammation after testing *in vivo* and *in vitro.*

ACKNOWLEDGEMENT

The author would like to thank Shivaji University, Kolhapur and Ashokrao Mane College of Pharmacy, Peth Vadgaon for supporting this work.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

NSAIDs: Non Steroidal Anti-inflammatory Drugs; **COX:** Cyclooxygenase; **PDB:** Protein Data Bank; **Å:** Angstrom Unit; **ADME:** Absorption, Distribution, Metabolism and Excretion; **PK:** Pharmaokinetics; **MW:** Molecular Weight; **TPSA:** Topological Polar Surface Area; **HIA:** Human Gastrointestinal Absorption; **BBB:** Blood-Brain Barrier; **PAINS:** Pan-Assay Interferences.

SUMMARY

The current study intended to unlock the anti-inflammatory potential studied using virtual discovery and ADMET evaluation of novel pyrazole-based COX-II inhibitors.

A total of 223 virtual pyrazole derivatives from series D (3,5-diphenyl-4,5-dihydro-1H-pyrazole), E (N-3,5-triphenyl-3,4-dihydropyrazole-2-carbothioamide) and F (2,3,5-triphenyl-3,4-dihydropyrazole) were synthesized.

The 3D crystal structure of Cyclooxygenase-II (COX-II) at 3.00 Å resolution (PDB:1CX2) was predicted for the active binding pocket using the CASTp3.0 server, revealing an area of 13821.595 and a volume of 26280.022 Å.

The selected protein's quality was assessed using the PROCHECK server, which determined a Ramachandran Plot showing that 76.3% of residues are present in the most favored regions.

Molecular docking performed with PyRx 0.8 software identified D202 and D305 as having higher binding affinities of -10.5 kcal/mol and -10.7 kcal/mol, respectively. Various interactions including conventional hydrogen bonds, van der Waals forces, Π-Alkyl and Π-Donor interactions were observed between the interacting residues and the pyrazole derivatives.

Pyrazole derivatives D202, D305 and F505 exhibited drug-like characteristics according to Lipinski, Ghose, Veber, Egan and Muegge parameters. Furthermore, they displayed a bioavailability score of 0.55, suggesting good bioavailability and pharmacokinetic properties, indicating a likelihood of greater than 10% bioavailability in rats.

REFERENCES

1. Azab, A., Nassar, A. and Azab, A.N., et al, Anti-inflammatory activity of natural products. Molecules, 2016;21(10):1321. Doi:10.3390/Molecules21101321.

2. Attiq, A., Jalil, J., Husain, K., et al, Raging the war against inflammation with natural products. Frontiers in pharmacology, 2018;9:976. Doi:10.3389/fphar.2018.00976.
3. Oniga, S.D., Pacureanu, L., Stoica, C.I., et al, COX inhibition profile and molecular docking studies of some 2-(trimethoxyphenyl)-thiazoles. Molecules, 2017;22(9):1507. Doi:10.3390/molecules22091507.
4. Zarghi, A. and Arfaei, S., Selective COX-2 inhibitors: a review of their structure-activity relationships. Iranian journal of pharmaceutical research: IJPR, 2011;10(4):655. PMID: 24250402; PMCID: PMC3813081.
5. Ju, Z., Li, M., Xu, J., et al, Recent development on COX-2 inhibitors as promising anti-inflammatory agents: The past 10 years. Acta Pharmaceutica Sinica B., 2022;12(6):2790-807. Doi:10.1016/j.apsb.2022.01.002.
6. Chakraborti, A.K., Garg, S.K., Kumar, R., et al, Progress in COX-2 inhibitors: a journey so far. Current medicinal chemistry, 2010;17(15):1563-93. Doi:10.2174/092986710790979980.
7. Wang, D. and Dubois, R., N., The role of COX-2 in intestinal inflammation and colorectal cancer. Oncogene, 2010;29(6):781-8. Doi:10.1038/onc.2009.421.
8. Dannhardt, G. and Kiefer, W., Cyclooxygenase inhibitors–current status and future prospects. European journal of medicinal chemistry, 2001;36(2):109-26. Doi:10.1016/s0223-5234(01)01197-7.
9. Chandrasekharan, N.V. and Simmons, D.L., 2004. The cyclooxygenases. Genome biology, 5, pp.1-7. Doi:10.1186/gb-2004-5-9-241.
10. Adelusi, T.I., Oyedele, A.Q.K., Boyenle, I.D., et al, 2022. Molecular modeling in drug discovery. Informatics in Medicine Unlocked, p.100880. Doi:10.1016/j.imu.2022.100880.
11. Pinzi, L. and Rastelli, G., Molecular docking: shifting paradigms in drug discovery. International journal of molecular sciences, 2009;20(18):4331. Doi: 10.3390/ijms20184331.
12. Salo-Ahen, O.M., Alanko, I., Bhadane, R., Bonvin, A.M., et al, Molecular dynamics simulations in drug discovery and pharmaceutical development. Processes, 2020;9(1):71. Doi:10.3390/pr9010071.
13. Pinzi, L. and Rastelli, G., Molecular docking: shifting paradigms in drug discovery. International journal of molecular sciences, 2019;20(18):4331. Doi:10.3390/ijms20184331.
14. Kurumbail, R.G., Stevens, A.M., Gierse, J.K., et al, Structural basis for selective inhibition of cyclooxygenase-2 by anti-inflammatory agents. Nature, 1996;384(6610):644-8. Doi:10.1038/384644a0.
15. Laskowski, R.A., MacArthur, M.W., Moss, D.S., et al, PROCHECK: a program to check the stereochemical quality of protein structures. Journal of applied crystallography, 1993;26(2):283-91. Doi:10.1107/S0021889892009944.
16. Tai, W., He, L., Zhang, X., et al, Characterization of the receptor-binding domain (RBD) of 2019 novel coronavirus: implication for development of RBD protein as a viral attachment inhibitor and vaccine. Cellular and molecular immunology, 2020;17(6):613-20. Doi:10.1038/s41423-020-0400-4.
17. Jain, A.S., Sushma, P., Dharmashekar, C., et al, *In silico* evaluation of flavonoids as effective antiviral agents on the spike glycoprotein of SARS-CoV-2. Saudi Journal of Biological Sciences, 2020;28(1):1040-51. Doi:10.1016/j.sjbs.2020.11.049.
18. Eberhardt, J., Santos-Martins, D., Tillack, A.F., et al, AutoDock Vina 1.2. 0: New docking methods, expanded force field and python bindings. Journal of chemical information and modeling, 2021;61(8):3891-8. Doi:10.1021/acs.jcim.1c00203.
19. Yadava, U., 2018. Search algorithms and scoring methods in protein-ligand docking. Endocrinol Int J, 6(6), pp.359-367. Doi:10.15406/emij.2018.06.00212.
20. Ajala, A., Uzairu, A., Shallangwa, G.A., et al., QSAR, simulation techniques and ADMET/pharmacokinetics assessment of a set of compounds that target MAO-B as anti-Alzheimer agent. Futur J Pharm Sci, 2023;9(4):1-20. Doi: 10.1186/S43094-022-00452-2.
21. Daina, A., Michielin, O. and Zoete, V., SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci Rep 2017;(7):42717. Doi:10.1038/srep42717.

Cite this article: Patil VM, More HN. Unlocking Anti-Inflammatory Potential: Virtual Discovery and ADMET Evaluation of Novel Pyrazole-Based COX-II Inhibitors. Indian J of Pharmaceutical Education and Research. 2025;59(1s):s312-s322.