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In Silico Study of Ethanolic and Methanolic Extracts of *Tridax procumbens* (*Londotan*) as Anti-Inflammatory Agents Targeting the COX-2 Receptor

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Abstract

Inflammation is a major pathological process that occurs when the body's immune system responds to stimuli caused by various injury factors. The incidence of injuries in Indonesia has reached 9.7%, showing an increase compared to 2014 (7.3%) and 2007 (7.9%). One plant that has not been widely utilized as a medicinal agent but is known to possess anti-inflammatory activity is *T. procumbens* (commonly known as *londotan*). The aim of this study was to evaluate the interaction mechanisms of the bioactive compounds from *T. procumbens* with the COX-2 receptor through a molecular docking approach using the receptor structure obtained from the Protein Data Bank (PDB ID: 5IKR). Sodium diclofenac was employed as the positive control to identify potential new anti-inflammatory drug candidates with favorable pharmacokinetic profiles The molecular docking simulation results indicated that Quercetin-3-O-α-L-rhamnopyranoside the lowest binding free energy (ΔG = −9.73 kcal/mol) with an inhibition constant (Ki) of 73.71 nM, and formed significant hydrophobic interactions with the native ligand residues LEU B:352, VAL B:523, VAL B:349, and ALA B:527. In terms of pharmacokinetic properties, Quercetin-3-O-α-L-rhamnopyranoside demonstrated superior performance compared to sodium diclofenac and the native ligand, suggesting that *T. procumbens* has promising potential to be developed as an anti-inflammatory drug.

Keywords: COX-2; Inflammation; In Silico; Molecular Docking; *Tridax procumbens*.

INTRODUCTION

Inflammation is a fundamental pathological process that occurs when the body's immune system responds to stimuli caused by various injury factors, resulting in symptoms such as swelling, pain, redness, heat, and dysfunction (J. Wang et al., 2024). In 2022, the incidence of injuries in Indonesia was reported at 9.7%, showing an increase compared to 7.3% in 2014 and 7.9% in 2007. The prevalence of injuries among children aged 1–4 years was 8.2%, 12.1% among those aged 5–14 years, and the highest prevalence was observed in school-aged children at 13% (Nadira et al., 2025).

One of the therapeutic strategies for inflammation involves the inhibition of the COX-2 (cyclooxygenase-2) enzyme. cyclooxygenase-2 is one of the three isoforms of cyclooxygenase that catalyzes the conversion of arachidonic acid into prostaglandin H2, a key precursor of prostacyclin involved in the inflammatory process (Bruno et al., 2023). Drugs commonly used as cyclooxygenase-2 inhibitors belong to the class of nonsteroidal anti-inflammatory drugs (NSAIDs), including diclofenac, acetylsalicylic acid, naproxen, nimesulide, ibuprofen and paracetamol (Da Silva et al., 2022). However, NSAIDs inhibit both cyclooxygenase-1 and cyclooxygenase-2 either selectively or non-selectively, which may lead to adverse effects on the liver, kidneys, and stomach (Huyen Do et al., 2022). This limitation has encouraged the development of new candidate compounds that selectively inhibit cyclooxygenase-2 or the exploration of natural products with anti-inflammatory potential and minimal side effects, such as *T. procumbens* (commonly known as londotan).

According to Amagbegnon et al. (2021), *T. procumbens* is a wild herbaceous species commonly found in fields, grasslands, and along roadsides. It is traditionally used due to its abundance of active phytoconstituents with anti-inflammatory effects (Devi et al., 2022). This plant is easily accessible and possesses various medicinal benefits, yet remains underutilized due to limited awareness (Evbuomwan et al., 2023). The leaves of *T. procumbens*, traditionally used for therapeutic purposes, are rich in phytochemical

compounds with potential anti-inflammatory effects (Berlin Grace et al., 2020).

Kaushik et al. (2020) reported that the leaves of *T. procumbens* contain 23 flavonoid and 8 alkaloid compounds. Furthermore, Ingole et al. (2022) identified 91 compounds in the ethanol and methanol extracts of *T. procumbens*, including flavonoids, lignans, tannins, terpenoids, norisoprenoids, and benzoic acid derivatives. However, computational studies investigating the bioactive compounds of *T. procumbens* to analyze the interactions between its ligands and the catalytic residues of cyclooxygenase-2 have not yet been conducted.

In silico–based computational chemistry has become an effective strategy to accelerate the discovery of bioactive compounds with high biological activity. This approach holds great potential for optimizing drug candidate development in a more cost-efficient manner compared to conventional methods (Yuliana et al., 2022). Among the various computational methods used in in silico studies, molecular docking is one of the most widely applied techniques, aiming to predict the interactions between drug molecules and biological targets (Sanjaya et al., 2024). The present study aims to identify the active compounds of *T. procumbens* that have potential as cyclooxygenase-2 enzyme inhibitors for the treatment of inflammation using a molecular docking approach. The results of this study are expected to serve as preliminary data or a reference for future research.

MATERIALS AND METHODS

Materials

The three-dimensional (3D) structure of cyclooxygenase-2 (PDB ID: 5IKR) was downloaded from the Protein Data Bank in (.pdb) format. The two-dimensional (2D) and three-dimensional (3D) structures of 2-[(2,3-dimethylphenyl)amino]benzoic acid (compound a) were used as the native ligand, while sodium diclofenac (compound b) served as the reference ligand. The active compounds contained in the *T. procumbens* extract, obtained from the studies by Kaushik et al. (2020) and Ingole et al. (2022), were used as the test ligands.

Methods

Lipinski's Rule of Five Analysis

The analysis process began by identifying the active compounds present in *T. procumbens*, which were then evaluated based on Lipinski's Rule of Five. The parameters included a molecular weight of <500 Da, no more than 10 hydrogen bond acceptors, a log P value of <5, no more than 5 hydrogen bond donors, and a molar refractivity (MR) between 40-130 (Gade et al., 2023). According to this rule, only one violation is permitted (Saritha et al., 2024).

Ligand and Receptor Preparation

The ligands were obtained from the PubChem database and redrawn in 2D form using MarvinSketch software. Protonation was then performed at pH 7.4, and the resulting structures were saved for conformational analysis using the MMFF94 force field settings. The 3D structure of the cyclooxygenase-2 receptor (PDB ID: 5IKR) with the compound a was retrieved from the Protein Data Bank. The cyclooxygenase-2 receptor was subsequently separated from its ligand and water molecules using Molegro Molecular Viewer v2012.2.5 (Ruswanto et al., 2022).

Molecular Docking Validation

The validation process was carried out by redocking the compound a into the active site of the receptor with 100 repetitions. The validation process generated a grid box output and Root Mean Square Deviation (RMSD) values, where an optimal docking result was indicated by an RMSD value of less than 2 Å (Suciati et al., 2025). The validated grid box parameters were then applied for docking the test ligands that had successfully passed the Lipinski's Rule of Five screening.

Docking of Test Ligands

Docking of the test ligands was performed using PyRx software, which serves as an interface for AutoDock (Vyshnavi AM et al., 2023). The receptor and ligand structures were imported into the program and converted from .pdb to .pdbqt format. The previously validated grid box parameters were applied to ensure that ligand binding occurred exclusively within the defined active site region (Jeevitha et al., 2024). The Lamarckian Genetic Algorithm (LGA) was employed without repetition, and each docking simulation generated 100 conformations. The resulting outputs included the binding free energy (ΔG) expressed in kcal/mol and the inhibition constant (Ki) values (Ruswanto et al., 2022).

Visualization of Receptor–Ligand Interactions

The docking results of the receptor–ligand complexes were converted into .pdb format, followed by interaction analysis between the ligands and the receptor's active site. The 2D and 3D visualizations of the receptor–ligand interactions were then generated using Discovery Studio Visualizer v24.1.0 (Ruswanto et al., 2023).

Prediction of Pharmacokinetic and Toxicity Profiles

Screening of the test ligands was conducted to predict their physicochemical characteristics, pharmacokinetic properties, and toxicity profiles (Ruswanto et al., 2020). The predicted parameters included absorption, distribution, metabolism, excretion, and toxicity (Rahardhian et al., 2022). The pharmacokinetic profile and potential toxicity of the active compounds from *T. procumbens* were predicted using the pkCSM web server

to provide an initial assessment of their safety and bioavailability (Abdelazeem et al., 2024).

Protein Analysis

In protein structure validation for drug discovery, the receptor structure was verified using PROCHECK, and the Ramachandran plot was analyzed. The plot displays different regions represented by colored boxes: red for the core (most favored) regions, yellow for the allowed regions, and beige for the generously allowed regions (Parashar et al., 2025). The Ramachandran plot is used to evaluate the quality of the modeled or experimental receptor structure by providing information on the total number of amino acid residues located in the favored, allowed, and disallowed regions (Oladejo et al., 2023).

RESULTS AND DISCUSSION

Lipinski's Rule of Five Analysis

The evaluation of drug-likeness based on Lipinski's Rule of Five was carried out to assess five key physicochemical parameters that play a crucial role in drug discovery and in evaluating the bioavailability of candidate compounds (Saritha et al., 2024). Out of the 122 *T. procumbens* compounds that were preliminarily identified, only 45 compounds met the Lipinski's Rule of Five criteria, which are essential for drug-like molecules. These 45 compounds were therefore selected for further analysis, as presented in Table 1.

Table 1. Results of Lipinski's Rule of Five Analysis.

Compound	Molecular weight (<500 Da)	Hydrogen donor (<5)	Hydrogen acceptor (<10)	Log P (<5)	Molar refractivity (40-130)
Kaempferol	286,239	4	6	2,2824	117,313
Catechin	290,271	5	6	1,5461	119,662
(-)-Epicatechin	290,271	5	6	1,5461	119,662
(+)-Catechin	290,271	5	6	1,5461	119,62
Biochanin	282,251	0	5	1,6158	119,203
Apigenin	268,224	1	5	1,3128	112,519
Naringenin	272,256	3	5	2,5099	114,235
Daidzein	253,233	1	4	2,2392	107,725
Quercetin	300,222	3	7	0,724	122,108
Butein	272,256	4	5	2,4051	114,232
Robinetin	302,222	3	7	0,724	122,108
Baicalein	268,224	1	5	1,3128	112,519
Nobiletin	402,399	0	7	3,5116	167,007
Ellagic acid	300,178	2	8	0,0488	118,565
Luteolin	284,223	2	6	1,0184	117,313
Myricetin	316,221	4	8	0,4296	126,902
Isorhamnetin	314,24	2	7	1,027	128,792
Silymarin	482,441	5	10	2,3627	198,675
Caffeic acid	179,151	2	4	-0,1391	74,381
Ferulic acid	193,178	1	4	0,1639	81,065
Galgravin	372,461	0	5	4,8058	160,7
(3s)-16-17-didehydrofalcarinol or oxylipin	242,362	1	1	3,6229	111,302
2-propyl-1-heptanol	158,285	1	1	2,9753	70,818
3-octene-1-ol	128,215	1	1	2,1152	57,398
9,12 octadecadionic acid, ethylester	275,412	0	2	3,4442	123,897
2-methyl heptane	114,232	0	0	3,2227	53,294
2-propenyl butanoate	128,171	0	2	1,5157	55,514
2-4 dimethyl heptane	128,259	0	0	3,4687	59,659
Dibutylphthatate	278,348	0	4	3,6004	119,631
Dentoarboreol B	258,361	2	2	2,5937	116,096
S-(+)- dehydrovomifoliol	222,284	1	3	1,808	95,851
Vomifoliol	224,3	2	3	1,5998	96,484
Icaricide B1	386,441	5	8	-0,587	158,277
Loliolide	196,2446	1	3	1,4092	83,757
(-)-8- methoxyobliquin	274,272	0	5	2,5175	114,914
Esculetin	178,143	2	4	1,2042	72,668
Quercetin-3-O-α-L-rhamnopyranoside	446,364	5	11	-0,7753	179,107
6-methoxy-7,8-dihydroxyflavone	253,233	1	4	2,2392	107,725
7-methoxy-6,8-dihydroxyflavone	284,267	2	5	2,8798	119,203
Wogonin	283,259	1	5	2,2478	119,203

Compound	Molecular weight (<500 Da)	Hydrogen donor (<5)	Hydrogen acceptor (<10)	Log P (<5)	Molar refractivity (40-130)
Oroxylin	283,259	1	5	2,2478	119,203
4-hydroxybenzaldehyde	122,123	1	2	1,2047	52,752
2-hydroxybenzaldehyde	122,123	1	2	1,2047	52,752
Benzyl glucoside	270,281	4	6	-0,9969	110,384
Oleanolic acid	455,703	1	3	5,8989	201,354

Ligand and Receptor Preparation

At this stage, ligand protonation was performed to adjust to the physiological blood pH of approximately 7.4. Simultaneously, conformational analysis was conducted to obtain the lowest-energy or most stable molecular conformation, allowing optimal interaction with the active site of the cyclooxygenase-2 receptor (PDB ID: 5IKR). The 3D structure of the cyclooxygenase-2 receptor is presented in Figure 1. This receptor complex consists of the receptor bound to its compound a. Receptor preparation involved the removal of water molecules and coenzymes that could interfere with ligand–receptor interactions (Ruswanto et al., 2022; Sahayarayan et al., 2021).

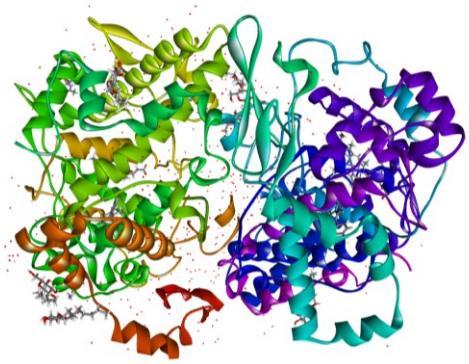


Figure 1. 3D structure of the cyclooxygenase-2 receptor (PDB ID: 5IKR).

Molecular Docking Validation

The validation of the docking method using the compound a aims to determine the ligand conformation that produces the most optimal biological effect. A drug molecule must stabilize a specific conformation of its target receptor to exert its pharmacological action. To evaluate the stability and accuracy of the interaction, the RMSD value was used, comparing the position of the compound a with that of the redocked conformation (Terefe et al., 2022).

Figure 2. 3D structure of the compound a before (magenta) and after (yellow) docking.

The RMSD value serves as a parameter that indicates the degree of deviation between the compound a before and its redocked conformation with the protein (Hanifan et al., 2024). The validation of the cyclooxygenase-2 receptor was performed with a grid center at x = 40.741, y = 38.189, and z = 85.878, and a grid box dimension of 40 × 40 × 40. The redocking process yielded an RMSD value of 0.55 Å, which was considered valid since it was below the acceptable threshold of 2 Å (Suciati et al., 2025).

This result indicates that the 3D structure of the compound a before and after redocking did not significantly differ (Figure 2) (F. Wang et al., 2022). Furthermore, a lower RMSD value corresponds to a more reliable docking model compared to the target crystal structure. Therefore, the same grid box dimensions and coordinates were applied for the docking of test ligands (Table 1).

Docking of Test Ligands

At this stage, the binding affinity was determined by analyzing the ΔG, Ki, and the types of interactions formed between the ligand and the receptor. The ΔG value represents the energy released during the binding process and is used to assess the strength of the protein–ligand interaction. A lower ΔG value indicates a stronger interaction (Chibuye et al., 2024).

The molecular docking results of the selected active compounds from *T. procumbens* against the cyclooxygenase-2 receptor are presented in Table 2. Out of the 45 phytoconstituents analyzed, 18 compounds exhibited lower ΔG values compared to the positive control and the compound a. Furthermore, these 18 compounds demonstrated amino acid interaction patterns

similar to those of the positive control, suggesting that the similarity in binding interactions may contribute to better binding affinity and potentially enhanced anti-inflammatory activity (Sohrab et al., 2022).

Table 2. Molecular docking results of compound b and test ligands against the cyclooxygenase-2 receptor. Bold text indicates interactions identical to the positive control.

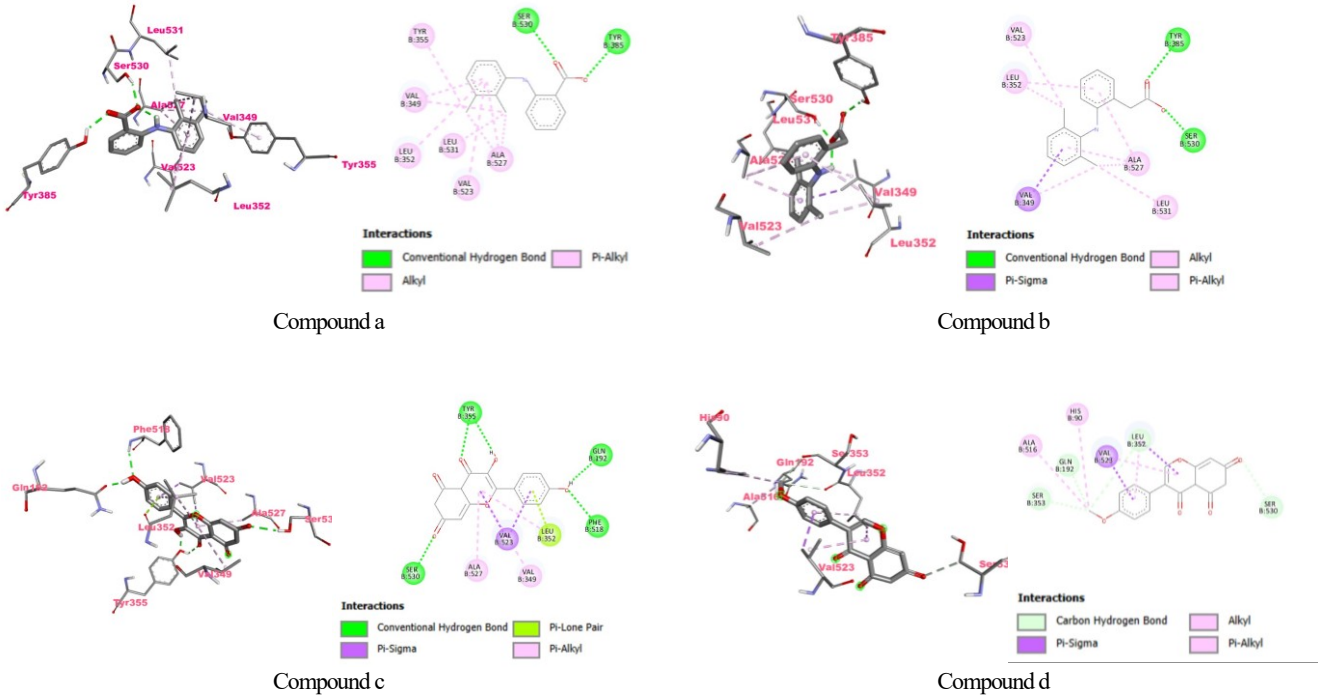
Compound	Run	ΔG (kkal/mol)	Ki	RMSD (Å)	Amino Acid Interactions	
					Hydrogen Bonds	Hydrophobic Bonds
Compound a	99	-7.03	7.01 uM	0.55	**SER B:530, TYR B:385**	**TYR B:355, VAL B:349, LEU B:352, VAL B:523, ALA B:527**
Compound b	23	105.43	2.64 uM	102.01	**TYR B:385, SER B:530**	**VAL B:349, VAL B:523, LEU B:352, ALA B:527,** LEU B:531
Kaempferol (compound c)	64	-8,52	570.15 nM	102.28	GLN B:192, TYR B:355, **SER B:530,** PHE B:518	**VAL B:523, LEU B:352, VAL B:349, ALA B:527**
Biochanin (compound d)	50	-8,33	789.02 nM	105.43	-	SER B:353, GLN B:192, **LEU B:352,** SER B:530, **VAL B:523,** ALA B:516, HIS B:90
Apigenin (compound e)	81	-8,17	1.03 uM	105.27	**SER B:530,** TYR B:355, GLN B:192, PHE B:518	ILE B:517, **VAL B:523, LEU B:352, ALA B:527, VAL B:349**
Daidzein (compound f)	62	-7,71	2.24 uM	105.90	GLN B:192	ILE B:517, **ALA B:527, VAL B:523, VAL B:349, LEU B:352,** ALA B:516
Quercetin (compound g)	7	-8,52	568.52 nM	102.18	**SER B:530,** TYR B:355, SER B:353, GLN B:192, PHE B:518	**VAL B:523, LEU B:352, ALA B:527, VAL B:349**
Butein (compound h)	54	-8,00	1.36 uM	105.71	PHE B:518, GLN B:192, SER B:353	HIS B:90, **VAL B:523, LEU B:352, ALA B:527,** ALA B:516
Robinetin (compound i)	56	-8,55	537.58 nM	106.36	PHE B:518, GLN B:192, SER B:353	**LEU B:352, VAL B:523,** MET B:522, **ALA B:527**
Baicalein (compound j)	42	-8,00	1.38 uM	104.07	-	**LEU B:352, VAL B:523**, MET B:522, ALA B:516
Luteolin (compound k)	93	-8,68	432.15 nM	103.19	HIS B:90, GLN B:192	SER B:530, **VAL B:523, LEU B:352, ALA B:527**
Myricetin (compound l)	24	-8,40	694.42 nM	105.30	SER B:353, PHE B:518, GLN B:192	SER B:530, MET B:522, **LEU B:352, VAL B:523**
Isorhamnetin (compound m)	77	-8,57	524.84 nM	102.59	**SER B:530,** TYR B:355, GLN B:192	ILE B:517, **VAL B:523, LEU B:352, VAL B:349,** ALA:527, HIS:90, ALA B:516
Galgravin (compound n)	73	-8,64	465.13 nM	98.50	**SER B:530**	**LEU B:352,** GLY B:526, **ALA B:527,** ILE:345, MET B:113, LEU B:359, LEU B:531, **VAL B:349, TYR B:355, VAL B:523,** TYR B:348, PHE B:381, TRP B:387, LEU B:384, TYR B:385
(-)-8-methoxyobliquin (compound o)	20	-7,72	2.19 uM	97.55	-	**VAL B:523,** TYR B:385, SER B:530, PHE B:381, LEU B:384, TRP B:387, **VAL B:349, ALA B:527, LEU B:352,** MET B:522, PHE B:518
Quercetin-3-O-α-L-rhamnopyranoside (compound p)	88	-9,73	73.71 nM	102.437	GLN B:192	SER B:530, **LEU B:352, VAL B:523,** LEU B:359, **VAL B:349, ALA B:527**
6-methoxy-7,8-dihydroxyflavone (compound q)	54	-7,80	1.93 uM	102.28	ARG B:120, TYR B:355, **SER B:530**	ALA B:527, **LEU B:352, VAL B:523,** MET B:522, GLY B:526, LEU B:531, **VAL B:349**

Compound	Run	ΔG (kkal/mol)	Ki	RMSD (Å)	Amino Acid Interactions	
					Hydrogen Bonds	Hydrophobic Bonds
7-methoxy-6,8-dihydroxyflavone (compound r)	46	-7,72	2.20 uM	105.20	PHE B:518, GLN B:192, SER B:353	**VAL B:523, ALA B:527, LEU B:352,** ALA B:516, HIS B:90, **VAL B:349**
Wogonin (compound s)	68	-7,63	2.56 uM	102.00	-	**VAL B:523, LEU B:352, ALA B:527,** ALA B:516
Oroxylin (compound t)	100	-8,51	579.66 nM	105.40	SER B:353, ILE B:517, PHE B:518	ALA B:527, GLN B:192, **VAL B:523, LEU B:352,** GLY B:526

Visualization of Receptor–Ligand Interactions

Molecular docking visualization was performed to evaluate the docking results of the compound a, compound b, and test ligands against the cyclooxygenase-2 receptor, as well as to analyze the type of bonds and ligand-receptors interactions. Figure 3 illustrates the two-dimensional (2D) and three-dimensional (3D) interactions between the ligands and amino acid residues. Observation of the interacting amino acid residues aims to identify the specific interactions formed between the ligands and the receptor (Teruel et al., 2023). The stability of the ligand–receptor complex is highly influenced by the number and strength of hydrogen bonds formed. The greater the number of hydrogen bonds, the more stable the ligand–receptor complex become (Mardianingrum et al., 2021).

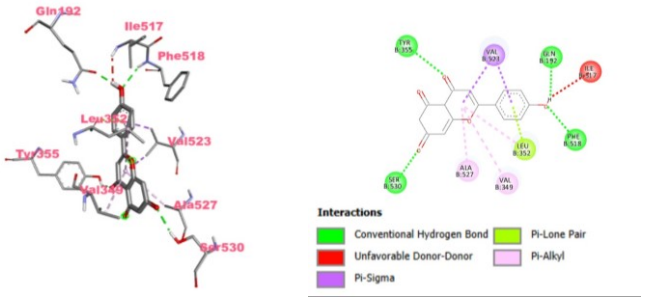
Identical amino acid residues observed in both the test ligands and the target receptor indicate stable interactions. Based on the amino acid residues obtained from each ligand (Figure 3), compound p demonstrated the most stable binding interactions and exhibited the lowest ΔG value compared to the compound a and the compound b, suggesting its strong potential as an anti-inflammatory candidate.



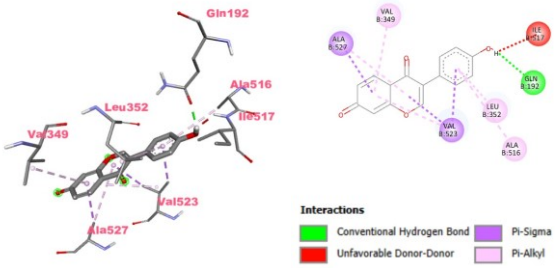
Compound a Compound b

Compound c Compound d

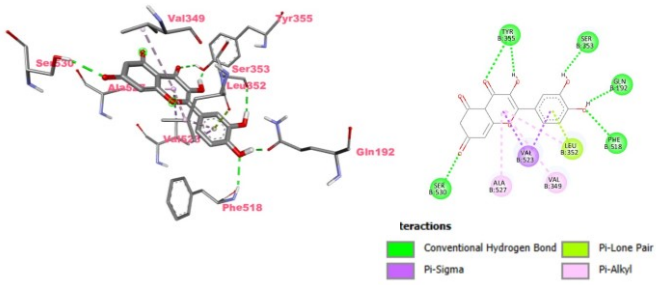
Figure 3. 2D and 3D interactions of the receptor with the compound a, compound b, and test ligands.



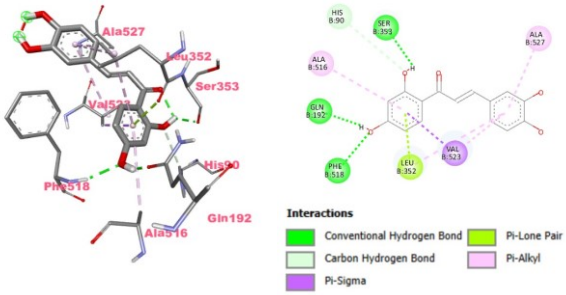
Compound e



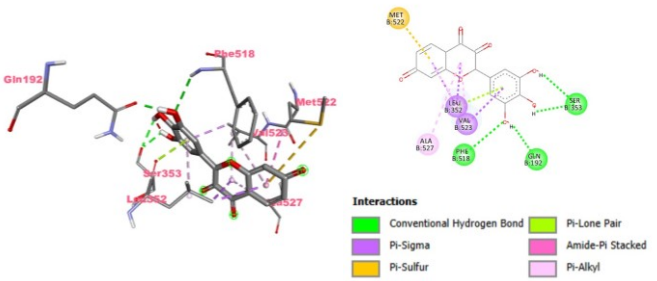
Compound f



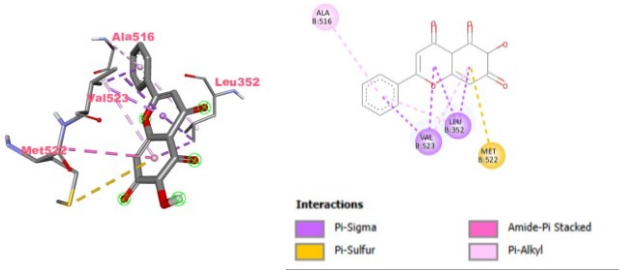
Compound g



Compound h

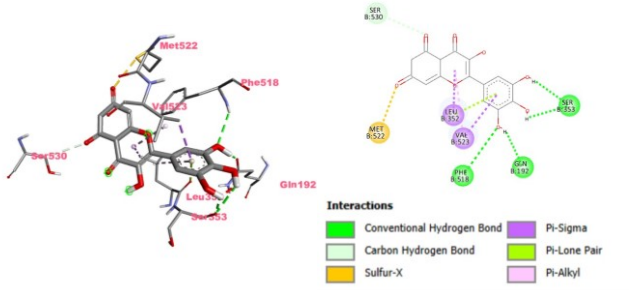


Compound i

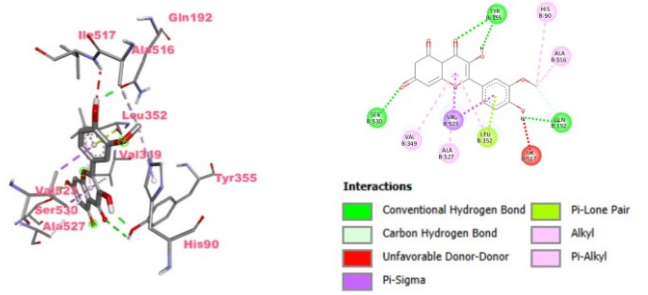


Compound j

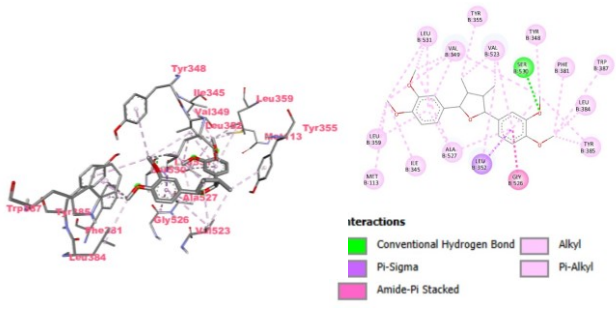
Compound k



Compound l



Compound m



Compound n

Figure 3 (Continued). 2D and 3D interactions of the receptor with the compound a, compound b, and test ligands.

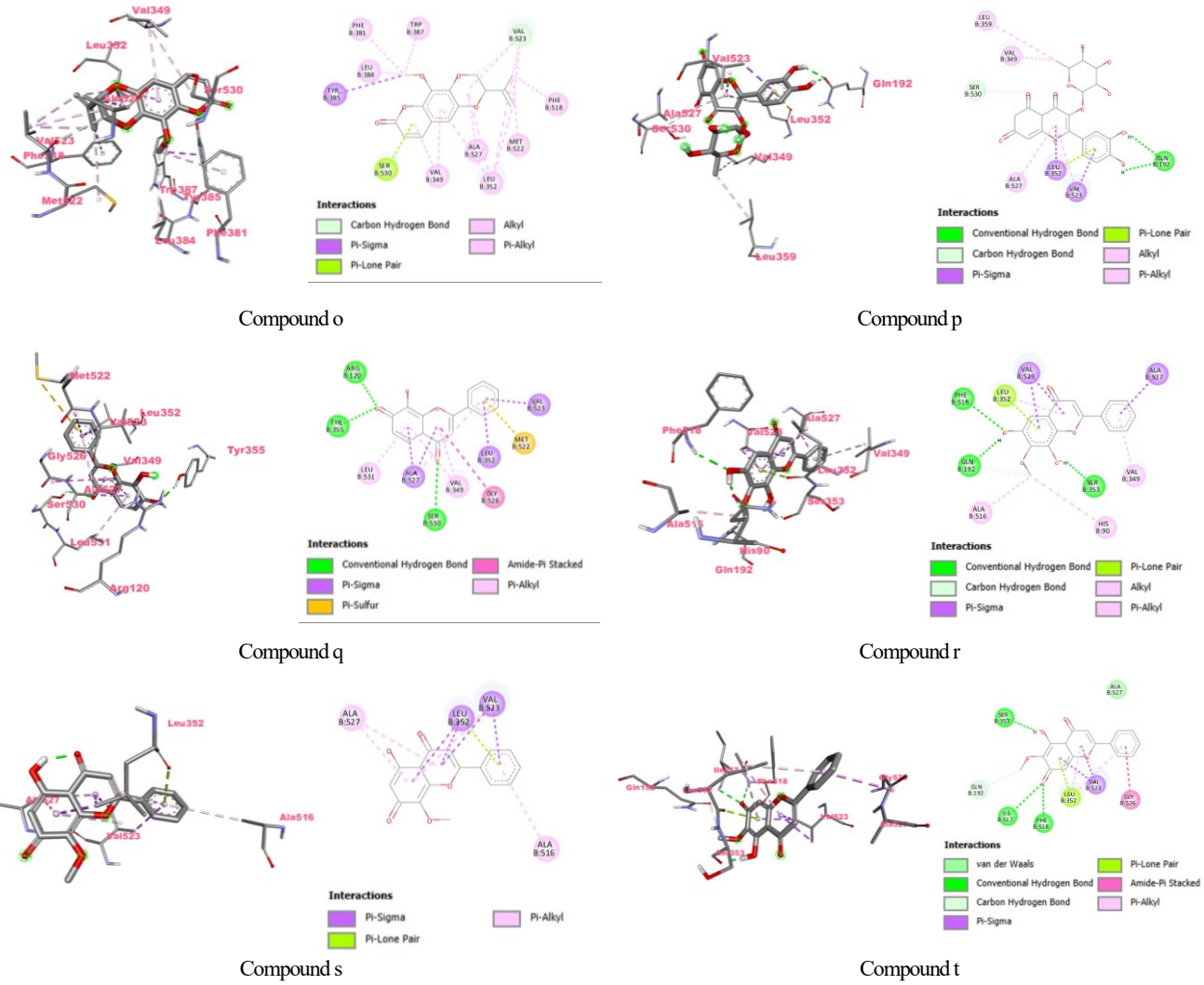


Figure 3 (Continued). 2D and 3D interactions of the receptor with the compound a, compound b, and test ligands.

Prediction of Pharmacokinetic and Toxicity Profiles

To achieve an optimal therapeutic effect, a drug must be able to reach its biological target within the body at an adequate therapeutic concentration while maintaining its bioactive form (Chibuye et al., 2024). To support this, the pharmacokinetic profiles were predicted using the pkCSM database, which applies graph-based classification and regression models to estimate ADMET parameters (Gadaleta et al., 2024).

Among the evaluated parameters, hERG inhibition was used to assess the potential cardiotoxicity of the drug candidates (Creanza et al., 2021). A prediction of "no" for hERG inhibition indicates that the compound does not block cardiac hERG channels and is therefore unlikely to cause arrhythmia (Wilapangga et al., 2025). Additionally, skin sensitization was assessed to evaluate the potential of a compound to induce allergic reactions upon topical application, which is critical for ensuring the safety of candidates intended for topical use. A "no" prediction for skin sensitization indicates that the compound is unlikely to cause skin allergy and can be considered safe for topical administration (Huong Ta et al., 2021).

Based on Table 3, among the 18 *T. procumbens* compounds analyzed, the safest compounds and recommended candidates for new drug development are compound c, compound d, compound e, compound f, compound h, compound i, compound j, compound n, compound o, compound p, and compound q.

Table 3. Evaluation of pharmacokinetic properties and toxicity of compound a, compound b, and test ligands.

Compound	hERG I inh	hERG II inh	Skin Sensitisation
Compound a	No	No	No
Compound b	No	No	No
Compound c	No	No	No
Compound d	No	No	No
Compound e	No	No	No
Compound f	No	No	No
Compound g	No	Yes	No
Compound h	No	No	No
Compound i	No	No	No
Compound j	No	No	No
Compound k	No	Yes	No
Compound l	No	Yes	No
Compound m	No	Yes	No
Compound n	No	No	No
Compound o	No	No	No
Compound p	No	No	No
Compound q	No	No	No
Compound r	No	Yes	No
Compound s	No	Yes	No
Compound t	No	Yes	No

Protein Analysis

In drug discovery studies, protein structure validation is performed using Ramachandran plot analysis via the PROCHECK software. This plot illustrates the distribution of amino acid residues across three regions: the core region (red), the allowed region (yellow), and the generously allowed region (beige) (Parashar et al., 2025). A lower percentage of amino acid residues in disallowed regions indicates a more stable and higher-quality protein structure, as the presence of non-glycine residues in disallowed regions can cause steric hindrance that disrupts protein conformation during molecular docking (Ningrum et al., 2023).

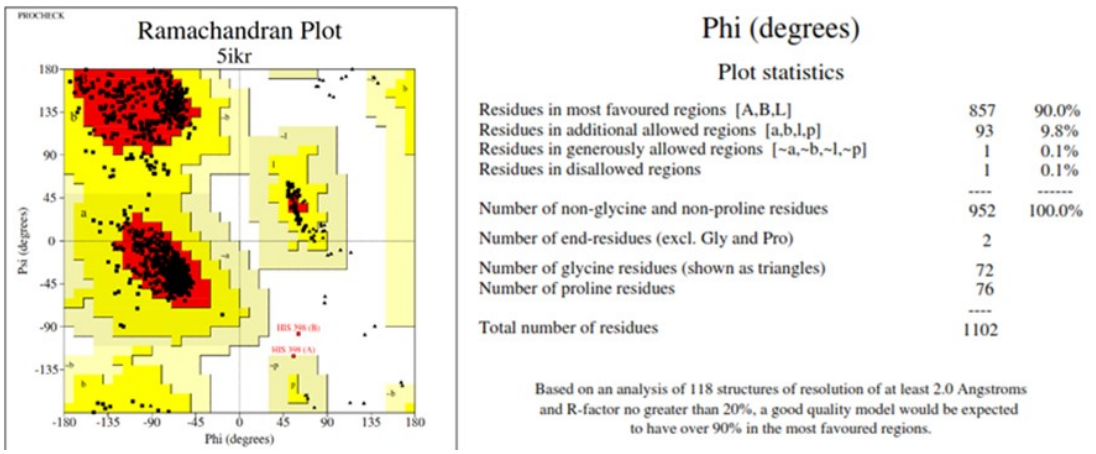


Figure 4. Ramachandran plot of the cyclooxygenase-2 receptor (PDB ID: 5IKR).

A protein structure is considered to have good quality if more than 90% of non-glycine residues are located in the most favored regions and fewer than 20% are in disallowed regions (Aziz et al., 2022). The analysis based on the Ramachandran plot is shown in Figure 4, indicating that the cyclooxygenase-2 receptor (PDB ID: 5IKR) contains 90% (857 residues) in the most favored regions, 0.1% (1 residue) in generously allowed regions, 9.8% (93 residues) in additional allowed regions, and 0.1% (1 residue) in disallowed regions. These results demonstrate that the cyclooxygenase-2 receptor structure is of sufficient quality to be used reliably as a docking target in molecular docking studies.

CONCLUSIONS

According to the results of the in silico analysis, 45 compounds identified from *T. procumbens* satisfied the physicochemical criteria of Lipinski's Rule of Five. Overall, this plant shows potential for development as an anti-inflammatory agent, as 18 compounds exhibitted stronger binding affinity than both compound a and the compound b. Among these, compound p demonstrated the most favorable binding properties, with the lowest ΔG value (-9.73 kcal/mol) and a KI of 73.71 nM. Moreover, this compound displayed similar amino acid interactions to those of compound a and the compound b, forming hydrophobic interactions with LEU B:352, VAL B:523, VAL B:349, and ALA B:527. These findings suggest that compound p is the most promising test compound from *T. procumbens* and a potential candidate for development as a novel cyclooxygenase-2 inhibitory anti-inflammatory drug.

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