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Research

In Silico Evaluation of Curcumin Binding Affinity toward Key Inflammatory Targets: COX-2, TNF-α, and NF-κB

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Abstract:
Introduction — Inflammation is a biological response to tissue injury or infection involving key mediators such as cyclooxygenase-2 (COX-2), tumor necrosis factor-alpha (TNF-α), and nuclear factor kappa B (NF-κB). Curcumin, the major bioactive compound of Curcuma longa, has demonstrated potential anti-inflammatory properties. This study evaluated the anti-inflammatory potential of curcumin using an in silico approach.
Methods — Biological activity was predicted using PASS Online, drug-likeness was evaluated according to Lipinski's Rule of Five, molecular docking was performed with PyRx AutoDock Vina against COX-2, TNF-α, and NF-κB, and pharmacokinetic and toxicity profiles were predicted using ADMET analysis.
Results — Curcumin showed anti-inflammatory potential with a Probability of Activity (Pa) of 0.667 and a Probability of Inactivity (Pi) of 0.019. The compound satisfied all Lipinski's Rule of Five criteria without violations. Molecular docking produced binding affinities of −7.5, −5.8, and −6.1 kcal/mol against COX-2, TNF-α, and NF-κB, respectively, with ligand–protein interactions dominated by hydrogen bonds and hydrophobic interactions. ADMET prediction indicated high human intestinal absorption (82.19%), strong plasma protein binding (97.1%), and no predicted mutagenic or carcinogenic effects.
Conclusion — Curcumin exhibits favorable drug-like properties and promising multitarget anti-inflammatory activity, supporting its potential as a candidate for anti-inflammatory drug development. Further in vitro and in vivo studies are required to confirm these findings.

Keywords: Curcumin; anti-inflammatory; molecular docking; COX-2; ADMET.

1. Introduction

Inflammation is the body's response to tissue damage or infection in cells. This condition can be triggered by various factors, such as harmful substances, foreign bodies, toxins, infections, chemicals,

pathogens, immune reactions, or physical trauma. Typical symptoms of inflammation include redness, increased temperature, swelling, and pain [1]. Although inflammation plays a crucial role in fighting infections and the wound healing process, chronic inflammation can have detrimental effects, such as causing redness accompanied by pain that can progress into serious diseases, for example, joint inflammation due to rheumatoid arthritis or cancer [2]. The inflammatory process begins when body cells are damaged, which then triggers the release of various chemical substances known as mediators, such as prostaglandins, bradykinin, histamine, cytokines, and biological oxidants. These mediators play a role in regulating the inflammatory response, including increasing blood vessel permeability, recruiting immune cells, and causing inflammatory symptoms. Therefore, controlling inflammatory mediators is a primary target in anti-inflammatory therapy [3].

Clinical symptoms of an inflammatory reaction include redness caused by excessive blood flow to the injured area, heat caused by inflammation on the body's surface, swelling (tumor) caused by the influx of fluid and cells from the bloodstream into the interstitial space, pain (dolor) caused by pressure from edema on tissues, and dysfunction [4]. Besides, anti-inflammatory therapy generally employs steroid anti-inflammatory drugs (SIDs) and nonsteroidal anti-inflammatory drugs (NSAIDs). Both classes of drugs work by suppressing the inflammatory process through various mechanisms, such as inhibiting prostaglandin formation, reducing leukocyte migration, and suppressing the release of inflammatory mediators. However, long-term use can lead to various side effects [1]. Steroidal anti-inflammatory drugs can cause peptic ulcers, lower the body's resistance to infection, lead to osteoporosis, muscle and fat tissue atrophy, increase intraocular pressure, and have a diabetogenic effect. Meanwhile, nonsteroidal anti-inflammatory drugs can cause gastric ulcers and even bleeding, kidney dysfunction, and anemia [5].

These limitations have driven the development of alternative therapies based on natural ingredients, particularly herbal plants that have long been used in traditional medicine. One promising bioactive compound is curcumin, a polyphenol compound that exerts anti-inflammatory effects through various mechanisms [6]. Curcumin is known to possess a wide range of biological activities, including anti-inflammatory, anti-allergic, antioxidant, antiparasitic, antimutagenic, and antimicrobial effects. Additionally, this compound is utilized in the management of diseases such as cancer and Human Immunodeficiency Virus (HIV) infections [7]. This compound works by inhibiting the cyclooxygenase (COX-2) and lipoxygenase (LOX) enzymes, thereby reducing the production of pro-inflammatory mediators such as prostaglandins and leukotrienes. Furthermore, curcumin suppresses the activation of the NF-κB transcription factor, reduces the expression of pro-inflammatory cytokines such as TNF-α, IL-1β, and IL-6, and modulates the MAPK and JAK/STAT signaling pathways. Other activities include inhibiting the proliferation and activation of immune cells involved in the inflammatory process, making curcumin a promising candidate for an anti-inflammatory agent [6].

In the development of bioactive compounds, in silico approaches serve as effective and efficient methods. These methods can predict the activity of a compound with potential as a drug and eliminate other compounds with low activity. Additionally, the in silico approach offers advantages in saving energy, time, and costs. One commonly used technique is molecular docking, which is utilized to evaluate interactions between curcumin and inflammatory targets such as Cyclooxygenase-2 (COX-2), Tumor Necrosis Factor alpha (TNF-α), and Nuclear Factor kappa B (NF-κB) [8]. This method can also estimate the strength of binding affinity based on binding energy values and assess the stability of the ligand–protein complex through parameters such as Root Mean Square Deviation (RMSD) and analysis of amino acid residues at the active site. Furthermore, molecular docking allows for the identification of the types of interactions formed, such as hydrogen bonds, hydrophobic interactions, and van der Waals forces that contribute to complex stability. Thus, this approach provides a comprehensive overview of curcumin's potential as an anti-inflammatory agent at the molecular level. The use of this

method is considered effective and efficient as it can accelerate the initial screening of bioactive compounds before further in vitro or in vivo testing [6]. Although curcumin is known to possess anti-inflammatory activity, studies examining its interactions with several major inflammatory targets simultaneously remain limited. Therefore, this study was conducted to analyze curcumin's interactions with COX-2, TNF-α, and NF-κB using molecular docking to reveal its potential as a multi-target anti-inflammatory agent. The results of this study are expected to support the development of curcumin as a safer and more effective natural anti-inflammatory candidate.

2. Method

2.1 Research Design

This study employed an in silico computational approach to evaluate the anti-inflammatory potential of curcumin against three major inflammatory targets, namely cyclooxygenase-2 (COX-2), tumor necrosis factor-alpha (TNF-α), and nuclear factor kappa B (NF-κB). The computational workflow consisted of biological activity prediction, ligand preparation, drug-likeness evaluation, receptor preparation, molecular docking validation, molecular docking simulation, ligand–protein interaction analysis, and pharmacokinetic and toxicity prediction (ADMET). The overall research workflow is illustrated in Figure 1.

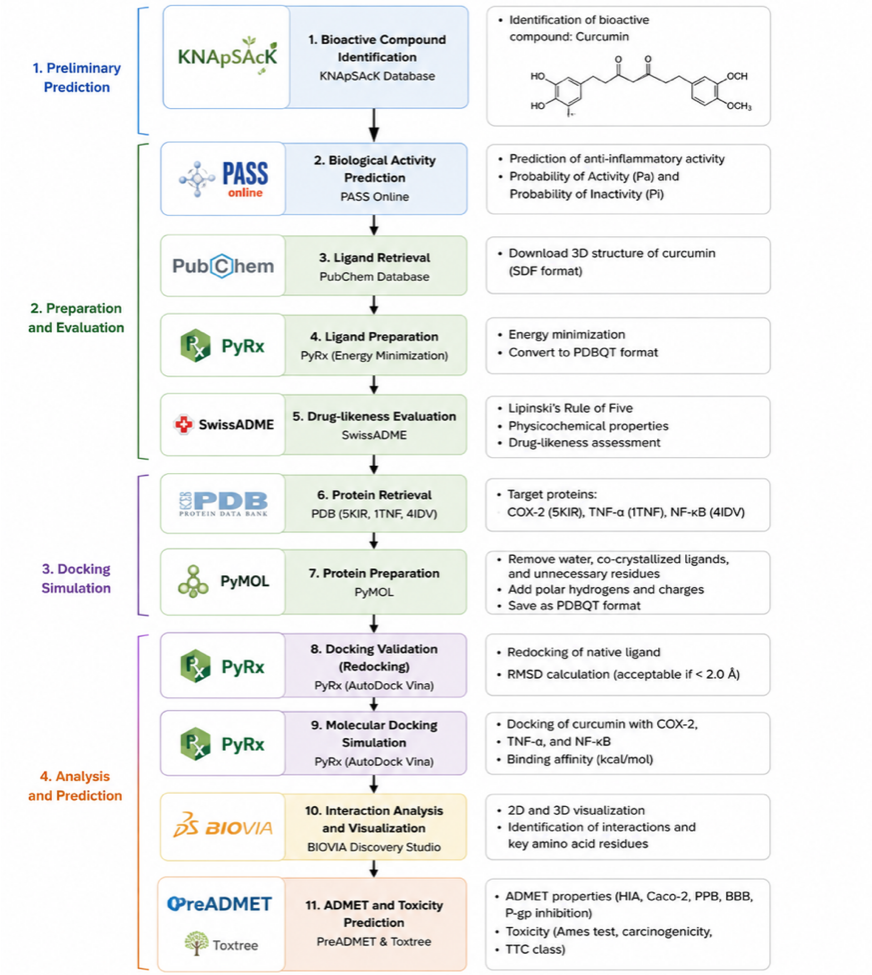


Fig. 1. Workflow of the in silico evaluation of curcumin against COX-2, TNF-α, and NF-κB.

2.2 Biological Activity Prediction

The preliminary biological activity of curcumin was evaluated using the Prediction of Activity Spectra for Substances (PASS) Online server (https://www.way2drug.com/passonline/). The three-dimensional

structure of curcumin was first identified from the KNApSAcK database and subsequently analyzed using PASS Online to predict its potential pharmacological activities. The prediction was based on the Probability of Activity (Pa) and Probability of Inactivity (Pi), where compounds with Pa values greater than Pi were considered to possess a higher likelihood of exhibiting the predicted biological activity.

2.3 Ligand Preparation

The three-dimensional structure of curcumin was retrieved from the PubChem database (https://pubchem.ncbi.nlm.nih.gov) in SDF format. Ligand preparation was performed using PyRx software, including geometry optimization through energy minimization to obtain the most stable molecular conformation before docking. The optimized ligand structure was then converted into PDBQT format for molecular docking simulations.

2.4 Drug-Likeness Evaluation

The physicochemical properties of curcumin were evaluated according to Lipinski's Rule of Five using the SwissADME web server (http://www.swissadme.ch/). The evaluated parameters included molecular weight, octanol–water partition coefficient (LogP), hydrogen bond donors, hydrogen bond acceptors, molar refractivity, and the number of Lipinski rule violations. This assessment was performed to determine the suitability of curcumin as a potential orally active drug candidate.

2.5 Protein Preparation

Three inflammation-related target proteins were selected for molecular docking, namely cyclooxygenase-2 (COX-2; PDB ID: 5KIR), tumor necrosis factor-alpha (TNF-α; PDB ID: 1TNF), and nuclear factor kappa B (NF-κB; PDB ID: 4IDV). The crystallographic structures were obtained from the Protein Data Bank (https://www.rcsb.org/). Protein preparation was performed using PyMOL by removing water molecules, co-crystallized ligands, and unnecessary heteroatoms while retaining the biologically relevant protein chain. Polar hydrogen atoms and Kollman charges were subsequently assigned, and the prepared receptors were saved in PDBQT format for docking analysis.

2.6 Molecular Docking Validation

Before docking simulations, the molecular docking protocol was validated by redocking the native ligand into the active site of each target protein using AutoDock Vina implemented in the PyRx platform. The validation aimed to assess the reliability of the docking protocol by comparing the predicted ligand pose with the crystallographic binding conformation. Root Mean Square Deviation (RMSD) values were used as the primary validation parameter, where RMSD values below 2.0 Å were considered acceptable for reliable docking simulations.

2.7 Molecular Docking Simulation

Molecular docking simulations were performed using AutoDock Vina integrated within the PyRx software package. Curcumin was docked against the active sites of COX-2, TNF-α, and NF-κB using identical docking parameters for all receptors. The binding affinity values (kcal/mol) were recorded to estimate ligand-binding strength, with more negative binding energies indicating stronger predicted interactions. The best docking pose for each receptor was selected based on the lowest binding affinity score for subsequent interaction analysis.

2.8 Ligand–Protein Interaction Analysis

The molecular interactions between curcumin and each target receptor were analyzed using BIOVIA Discovery Studio Visualizer 2021. Both two-dimensional (2D) and three-dimensional (3D) interaction profiles were generated to identify hydrogen bonds, hydrophobic interactions, π–π interactions, and the amino acid residues involved in ligand binding. These interaction profiles were subsequently interpreted to explain the observed binding affinity values.

2.9 ADMET Prediction

To evaluate the pharmacokinetic characteristics and toxicity profile of curcumin, computational ADMET prediction was performed using the PreADMET web server (https://preadmet.webservice.bmdrc.org/) and Toxtree software. The evaluated parameters included Human Intestinal Absorption (HIA), Caco-2 permeability, Plasma Protein Binding (PPB), Blood–Brain Barrier (BBB) penetration, P-glycoprotein inhibition, mutagenicity (Ames test), carcinogenicity, and Threshold of Toxicological Concern (TTC). These parameters provide preliminary insight into the drug-likeness, pharmacokinetic behavior, and safety profile of curcumin as a potential anti-inflammatory candidate.

3. Results

3.1 Biological Activity Prediction

The biological activity prediction was performed to provide an initial assessment of the anti-inflammatory potential of curcumin before conducting molecular docking simulations. PASS Online predicts the probability of biological activity based on the structural characteristics of the compound, expressed as the Probability of Activity (Pa) and Probability of Inactivity (Pi). The predicted biological activity of curcumin is presented in Table 1.

Table 1. Predicted biological activity of curcumin based on PASS Online analysis.

No.	Compound	Formula	Pa Value	Pi Value
1	Curcumin	$C_{21}H_{20}O_6$	0.667	0.019

3.2 Drug-Likeness Evaluation

The physicochemical properties of curcumin were subsequently evaluated to determine its suitability as an orally active drug candidate according to Lipinski's Rule of Five. Parameters including molecular weight, lipophilicity (LogP), hydrogen bond donors, hydrogen bond acceptors, and molar refractivity were assessed and compared with the accepted drug-likeness criteria. The evaluation results are summarized in Table 2.

Table 2. Drug-likeness properties of curcumin according to Lipinski's Rule of Five.

No	Compound	Massa (g/mol)	Log P	HBD	HBA	MR
1	Curcumin	368.38	3.20	2	6	102.80

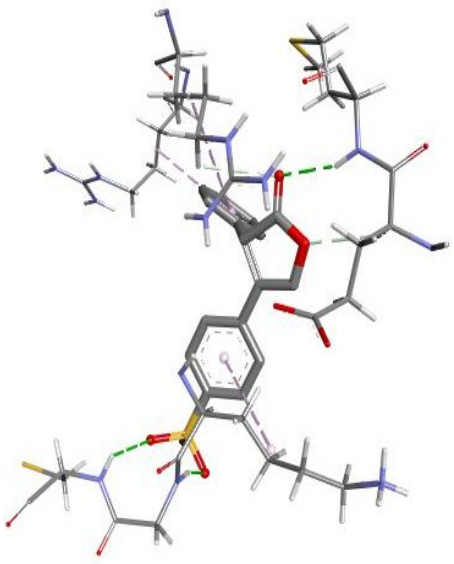
3.3 Molecular Docking against Cyclooxygenase-2 (COX-2)

To investigate the interaction of curcumin with cyclooxygenase-2 (COX-2), molecular docking simulations were performed using the prepared receptor structure. Docking performance was evaluated based on binding affinity values, docking validation (RMSD), hydrogen bond formation, and amino acid residues involved in ligand binding. The docking results for curcumin, the native ligand, and the reference compound are presented in Table 3.

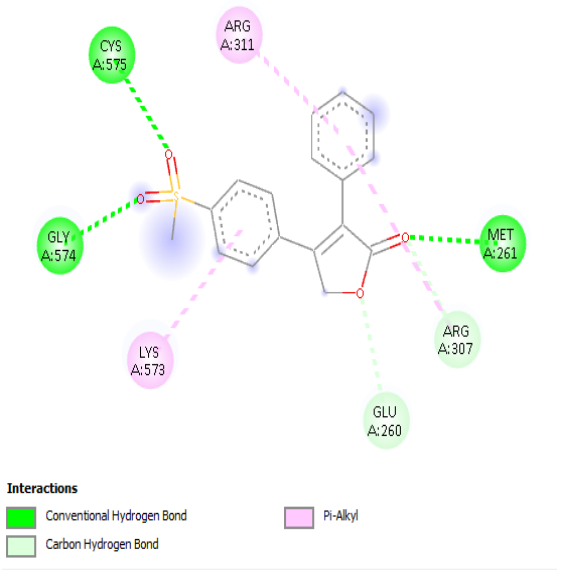
The molecular interactions observed during docking were further visualized to identify the binding orientation and intermolecular interactions between each ligand and the receptor active site. Both two-dimensional (2D) and three-dimensional (3D) interaction diagrams provide structural insight into the predicted binding mechanism. Representative docking poses are shown in Figure 1.

Table 3. Binding affinity and ligand–protein interactions of curcumin, the native ligand, and celecoxib against the COX-2 receptor.

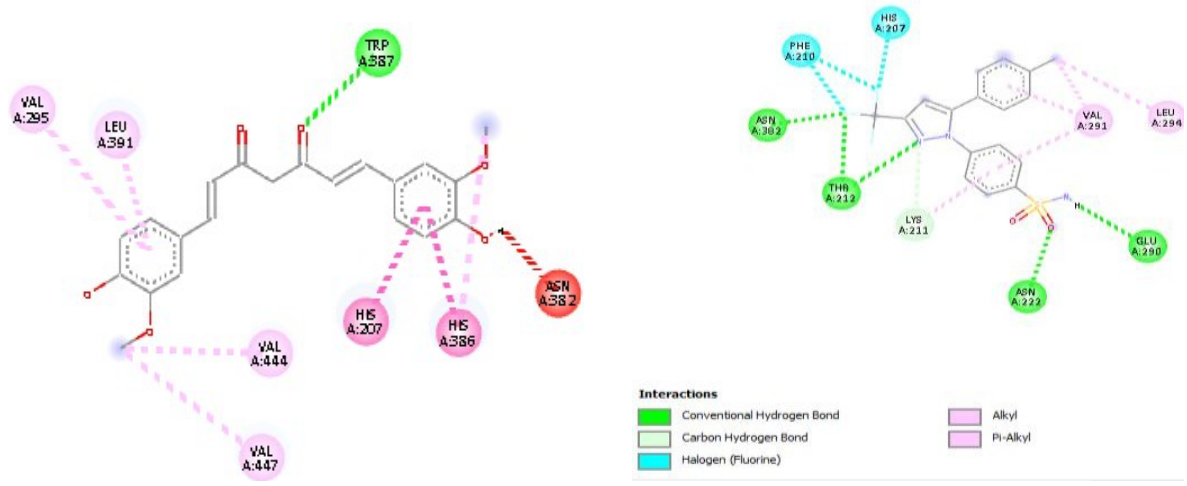
No	Compound	Binding Affinity (Kcal/mol)	RMSD	Acid Residue Amino	Hydrogen Bonds	Bond Distance
1	RCX Ligand	-6.9	0	CYS575 GLY574 MET261 ARG311 ARG307 LYS573 GLU260	CYS575 GLY574 MET261	2,88 2,21 2,33
2	Curcumin	-7.5	0	TRP387 LEU391 VAL295 VAL444 VAL447 HIS207 HIS386 ASN382	TRP387	2,62
3	Celecoxib	-8.3	0	ASN382 ASN222 THR212 GLU290 LYS211 PHE210 HIS207 VAL291 LEU294	ASN382 ASN222 THR212 GLU290	2,42 2,61 2,54 2,68



(a)



(b)



(c) **(d)**

Fig. 1. Molecular docking interactions of curcumin, the native ligand, and celecoxib with the COX-2 receptor. (a) Three-dimensional interaction of the native ligand; (b) two-dimensional interaction of the native ligand; (c) two-dimensional interaction of curcumin; and (d) two-dimensional interaction of celecoxib

3.4 Molecular Docking against Tumor Necrosis Factor-α (TNF-α)

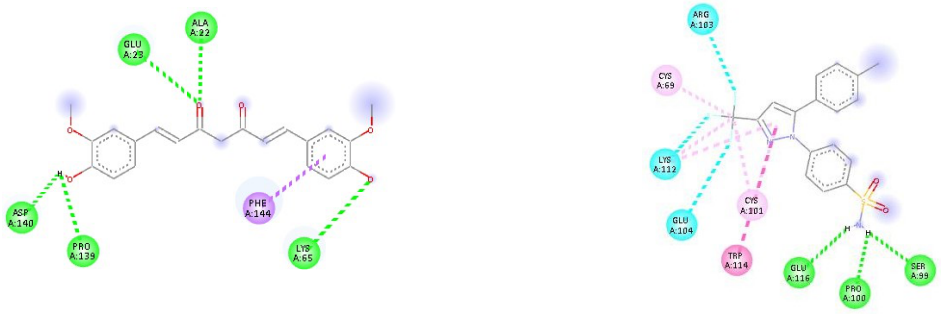
To further evaluate the multitarget anti-inflammatory potential of curcumin, molecular docking simulations were performed against tumor necrosis factor-alpha (TNF-α). Binding affinity values together with hydrogen bonding patterns and interacting amino acid residues were analyzed to characterize the predicted ligand–protein interactions. The docking results are presented in Table 4.

Table 4. Binding affinity and ligand–protein interactions of curcumin and celecoxib with the TNF-α receptor.

No	Compound	Binding Affinity (Kcal/mol)	RMSD	Acid Residue Amino	Hydrogen Bonds	Bond Distance
1	Curcumin	-5.8	0	ALA22 GLU23 ASP140 PRO139 LYS65 PHE144	ALA22 GLU23 ASP140 PRO139 LYS65	3,21 3,24 1,80 2,75 3,26
2	Celecoxib	-6.6	0	SER99 PRO100 GLU116 GLU104 CYS101 CYS69 LYS112 ARG103 TRP114	SER99 PRO100 GLU116	2,26 2,27 2,27

The interaction profiles obtained from molecular docking were subsequently visualized using two-

dimensional interaction diagrams to illustrate the binding pattern of each ligand within the active site of TNF-α. The docking interaction maps are presented in Figure 2.



(a) **(b)**

Fig. 2. Two-dimensional molecular interaction maps of (a) curcumin and (b) celecoxib with the TNF-α receptor.

3.5 Molecular Docking against Nuclear Factor Kappa B (NF-κB)

The binding interaction of curcumin with nuclear factor kappa B (NF-κB), an important transcription factor involved in inflammatory signaling, was also evaluated through molecular docking simulations. Binding affinity values and ligand–protein interaction profiles were analyzed to assess the binding characteristics of curcumin relative to the native ligand and celecoxib. The molecular docking results are summarized in Table 5.

Table 5. Binding affinity and ligand–protein interactions of curcumin, the native ligand, and celecoxib with the NF-κB receptor.

No	Compound	Binding Affinity (kcal/mol)	RMSD	Amino Acid Residue	Bond Type
1	13V Ligand	-7.9	0	ASP519; GLU473 GLY409 SER476 ARG408	Hydrogen Bond Carbon Hydrogen Bond Pi–Carbon Hydrogen Bond n Pi–Alkyl Interaction
2	Curcumin	-6.1	0	GLU440; ARG432 LYS430 PHE411; HIS537	Hydrogen Bond Carbon Hydrogen Bond Pi-Pi Stacked
3	Celecoxib	-8.7	0	VAL431; TRP464 GLY412; LYS430; PHE411	Pi-Alkyl Interaction Fluorine

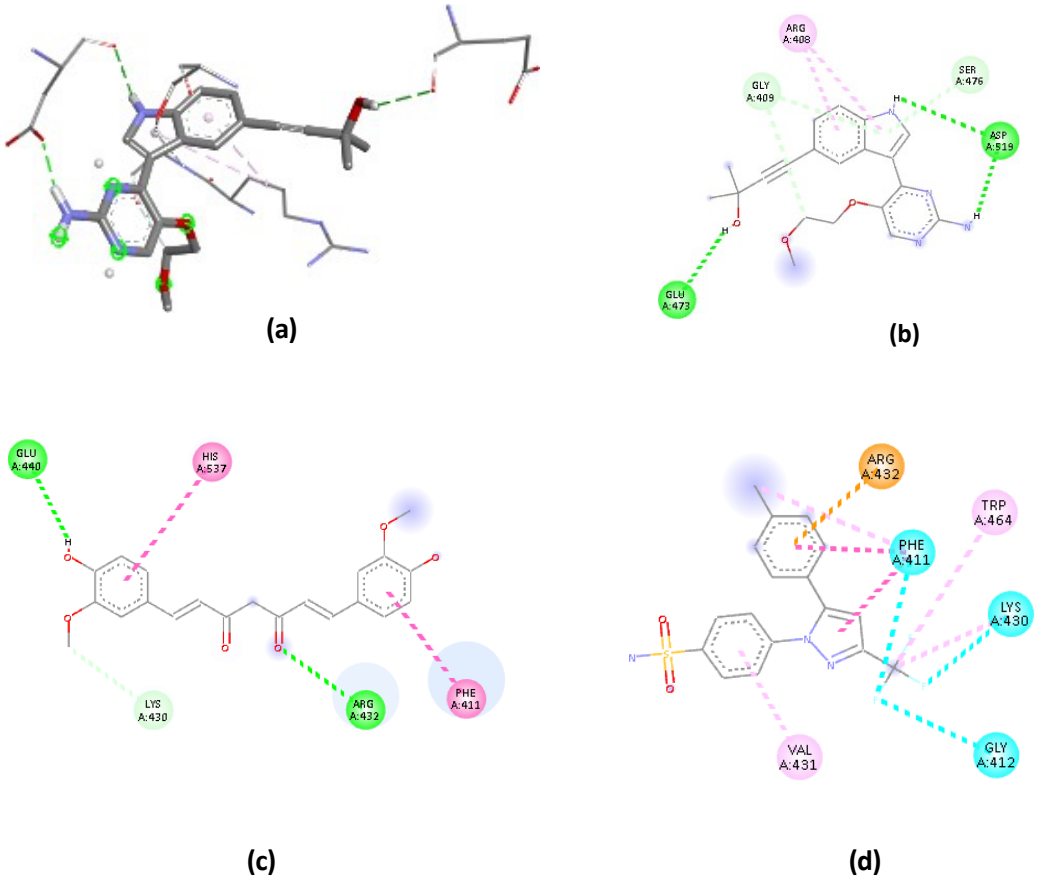


Fig. 3. Molecular docking interactions of curcumin, the native ligand, and celecoxib with the NF-κB receptor. (a) Three-dimensional interaction of the native ligand; (b) two-dimensional interaction of the native ligand; (c) two-dimensional interaction of curcumin; and (d) two-dimensional interaction of celecoxib.

3.6 ADMET Prediction

To complement the molecular docking analysis, the pharmacokinetic behavior and toxicity profile of curcumin were predicted using computational ADMET analysis. The evaluated parameters included intestinal absorption, plasma protein binding, blood–brain barrier permeability, P-glycoprotein inhibition, and toxicity endpoints, providing an initial assessment of the compound's drug development potential. The predicted ADMET properties are presented in Table 6.

Table 6. Predicted pharmacokinetic and toxicity (ADMET) properties of curcumin.

No.	Compound	Absorption		Distribution				Toxicity	
		Caco2	HIA	PPB	BBB	Pgb Inhibition	Kroes TTC	Carcinogen	Mutagen
1.	Curcumin	0.093	82.19	97.1	0.58	Inhibitor	Safe	-	-

4. Discussion

Compound potential prediction is performed to identify the characteristics and biological activities of a compound, particularly regarding its use as a drug candidate or natural product. This analysis uses the parameters Pa (*Potential Activity*) and Pi (*Potential Inactivity*). Predictions are made using Way2Drug PASS Online, accessible at https://www.way2drug.com/passonline/. Test results from PASS Online provide information on Pa (*Potential Activity*) and Pi (*Potential Inhibitory*) values. If the Pa value

is greater than 0.7, the compound is categorized as having very high biological activity, and the results tend to align with laboratory-scale testing. Meanwhile, if the Pa value falls within the range of 0.5 to 0.7, the compound is still considered to have fairly good biological activity thereby potentially demonstrating a high success rate when further tested *in vitro* or *in vivo* [9].

The results of the analysis of curcumin's anti-inflammatory activity potential are presented in Table 1. Based on these data, curcumin has a Pa value of 0.667 and a Pi value of 0.019. The Pa value indicates the probability that a compound is active, while the Pi value indicates the likelihood that the compound is inactive [10]. Thus, a higher Pa value compared to Pi indicates that curcumin is likely to possess biological activity. Overall, a Pa value of 0.667 falls into the moderate category, while the low Pi value suggests a low probability of inactivity. This confirms that curcumin has considerable potential as an anti-inflammatory agent [11].

Drug-likeness testing is an approach used to assess a compound's suitability as a drug candidate. One of the guidelines used is Lipinski's *Rule of Five*, which establishes several key parameters for determining a compound's potential as an oral drug. These parameters include a molecular weight ≤500 g/mol, the number of hydrogen bond acceptors (HBA) ≤10, the number of hydrogen bond donors (HBD) ≤5, a log P value ≤5, and *a molar refractive index* between 40–130 [12].

The analysis results indicate that curcumin meets the criteria of Lipinski's Rule of Five, thus demonstrating good potential as a candidate for an oral drug. Based on the data in Table 2, curcumin has a molecular weight of 368.38 g/mol, a log P value of 3.20, 2 hydrogen bond donors (HBD), and 6 hydrogen bond acceptors (HBA). These parameters fall within the limits required by Lipinski's rules, thus supporting good absorption and membrane permeability. Additionally, a molar refractivity value of 102.80 indicates that curcumin possesses structural characteristics suitable for interacting with biological targets. Thus, overall, curcumin exhibits a good drug-likeness profile and does not violate Lipinski's rules [13].

Compounds with a molecular weight exceeding 500 Daltons tend to face difficulties penetrating cell membranes via diffusion. Furthermore, a drug candidate typically has fewer than five hydrogen bond donors and fewer than ten hydrogen bond acceptors. This relates to the energy requirements for absorption, where hydrogen bonding capacity is directly proportional to the energy needed. The fewer the hydrogen bonds, the lower the energy required for absorption, and vice versa. Furthermore, a compound must also possess appropriate lipophilicity, with a log P value ranging from -0.4 to 5. The log P value describes the solubility balance of the compound in the lipid and aqueous phases. The higher the log P value, the greater the compound's hydrophobicity. However, excessively high hydrophobicity can increase the risk of toxicity because the compound tends to accumulate in the lipid bilayer. Conversely, if the log P value is too low, the compound will have difficulty penetrating the lipid bilayer, thereby inhibiting the absorption process [14].

Ligands and receptors that have undergone the preparation stage are then analyzed using molecular docking methods with the assistance of the PyRx AutoDock software. At this stage, receptors that have been separated from their natural ligands still contain solvent molecules, such as water, as well as other residues that need to be eliminated. The presence of water molecules around the receptor can interfere with the docking process because they have the potential to interact with the ligand through hydrogen bonds, thereby inhibiting the formation of optimal interactions between the ligand and the receptor [15].

The analyzed parameters include *binding affinity* values, *root mean square deviation* (RMSD), interaction types, involved amino acid residues, and bond distances. The results of the molecular

docking simulations are presented in Table 3, Table 4, and Table 5. The *binding affinity* value is used to indicate the strength of the interaction between the ligand and the receptor. The smaller the *binding affinity* value, the stronger and more stable the interaction formed between the ligand and the receptor. A negative value indicates that the energy required for the interaction to occur is lower, allowing the binding process to occur spontaneously [9]. *Root mean square deviation* (RMSD) is a parameter used to evaluate the alignment of atomic positions between the predicted structure and the experimental structure, with a docking success criterion of an RMSD less than 2.0 Å [16].

Based on Table 3 for the COX-2 enzyme target, curcumin exhibits a *binding affinity* of -7.5 kcal/mol, which is lower than that of the native ligand RCX (-6.9 kcal/mol), yet still higher than that of the comparator ligand celecoxib (-8.3 kcal/mol). This indicates that curcumin has better affinity than the native ligand but remains inferior to celecoxib as a standard inhibitor. All compounds showed an RMSD value of 0, indicating that the conformations of the *docked* ligands closely match the reference structure and meet the *docking* validity criteria [16].

In Table 4, interactions with the TNF-α target show that curcumin has a *binding affinity* of -5.8 kcal/mol, while celecoxib has a lower value of -6.6 kcal/mol. This indicates that curcumin's affinity for TNF-α is weaker than that of the reference ligand. Nevertheless, an RMSD value of 0 indicates that the *docking* results remain valid and exhibit excellent structural fit [16].

Furthermore, in Table 5 for the NF-κB target, curcumin exhibits a *binding affinity* of -6.1 kcal/mol, which is lower than that of the native ligand 13V (-7.9 kcal/mol) and celecoxib (-8.7 kcal/mol). This indicates that curcumin's affinity for NF-κB is relatively weaker compared to both of these reference ligands. However, as with other targets, an RMSD value of 0 indicates that *the docking-derived* conformation exhibits a very high degree of fit [16].

Interactions between ligands and receptors result in various types of bonds, bond distances, and the involvement of specific amino acid residues. The bonds formed can include hydrogen bonds, van der Waals interactions, or other types of interactions. Compared to van der Waals bonds, hydrogen bonds are stronger because they can still form even when there is a distance between the ligand and the receptor. In the interaction between the receptor and the native ligand, four hydrogen bonds are formed, whereas the test compound produces three hydrogen bonds, and the comparison ligand forms two hydrogen bonds. A greater number of hydrogen bonds indicates a higher level of stability of the ligand-receptor complex. Additionally, the bond energy value reflects the ligand's affinity for the target protein. The lower the bond energy, the greater the tendency of the ligand to bind to the receptor. An increase in the number of hydrogen bonds also contributes to the high affinity of the ligand for the target protein [17].

Amino acid residue analysis is performed to identify ligand-receptor interactions, which become more stable and stronger when hydrogen bonds form [4]. Hydrogen bonding is an important non-covalent interaction that occurs when a hydrogen atom is bonded to an electronegative atom such as oxygen or nitrogen and interacts with another electronegative atom. In molecular docking, the ideal hydrogen-bonding distance is generally in the range of 1.5–2.5 Å. [17].

Based on Table 3, the interaction between the native RCX receptor and ligand indicates the involvement of three amino acid residues in hydrogen bonding: CYS575, GLY574, and MET261, with bond distances ranging from 2.21 to 2.88 Å. Meanwhile, in the interaction between the test compound (curcumin) and the receptor, only one amino acid residue forms a hydrogen bond, namely TRP387, with a bond distance of 2.62 Å. As for the interaction between the reference ligand celecoxib and the receptor, four amino acid residues were found to play a role in hydrogen bonding: ASN382, ASN222,

THR212, and GLU290, with bond distances ranging from 2.42 to 2.68 Å. Generally, the ideal hydrogen bond distance in molecular docking falls within the range of 1.5–2.5 Å. Hydrogen bonds are important non-covalent interactions that form when an H atom binds to an electronegative atom such as O or N, and then interacts with another electronegative atom [17].

Based on Table 4, the interaction between curcumin and the receptor involves five amino acid residues in hydrogen bond formation, namely ALA22, GLU23, ASP140, PRO139, and LYS65, with bond distances ranging from 1.80 to 3.26 Å. The number of residues involved with curcumin is greater than that of the comparator ligand celecoxib, which involves only three amino acid residues—namely SER99, PRO100, and GLU116—with bond distances between 2.26 and 2.27 Å. This indicates that curcumin has a fairly good ability to form hydrogen bonds with the receptor, although not all bond distances fall within the ideal range. It should be noted that the strength of the interaction is determined not only by the number of hydrogen bonds but also by the contribution of hydrophobic interactions and the ligand's fit within *the binding pocket* [18].

Based on Table 5, the interaction between the receptor and the native 13V ligand involves residues ASP519 and GLU473 in the formation of hydrogen bonds, as well as an additional carbon-hydrogen bond at GLY409, supported by other interactions such as a π-carbon-hydrogen bond at SER476 and a π-alkyl interaction at ARG408. Meanwhile, in the interaction between the test compound (curcumin) and the receptor, there are two amino acid residues forming hydrogen bonds, namely GLU440 and ARG432, as well as additional interactions in the form of a carbon-hydrogen bond at LYS430 and a π-π *stacking* interaction involving PHE411 and HIS537. As for the interactions between the reference ligand celecoxib and the receptor, the interactions formed are dominated by hydrophobic interactions, such as π-alkyl interactions at VAL431 and TRP464, as well as other interactions involving fluorine and π-cation interactions involving GLY412, LYS430, PHE411, and ARG432, which collectively contribute to the stability of the ligand–receptor complex through a combination of both polar and nonpolar non-covalent interactions [19].

In the field of medicinal chemistry, ADMET analysis is used to evaluate a drug compound's ability to penetrate cell membranes and reach targets within the body [20]. ADMET analysis encompasses several key parameters: absorption, distribution, metabolism, and toxicity. The absorption parameter consists of Human Intestinal Absorption (HIA) and Caco-2 cell permeability, while distribution includes Plasma Protein Binding (PPB) and the Blood-Brain Barrier (BBB). The metabolism parameter relates to cytochrome P450 (CYP) enzymes, while toxicity is evaluated based on mutagenic and carcinogenic potential. HIA values describe the rate of compound absorption in the human intestine, classified into three categories: low (0–20%), moderate (20–70%), and high (70–100%). Predicting HIA is crucial for determining a compound's potential as a drug candidate. Additionally, the Caco-2 cell model is used as a representative *in vitro* approach for predicting oral absorption of compounds. Permeability levels in the Caco-2 model are classified into three categories: low (<4), moderate (4–70), and high (>70) [17].

Good drug distribution requires good PPB and BBB permeability. Table 6 shows that the curcumin compound has a PPB value of 97.1%. A PPB value exceeding 90% indicates that the compound is strongly bound to plasma proteins. However, a drug is generally more efficient when in its free form to cross cell membranes and reach its biological target compared to when it is strongly bound to plasma proteins. Meanwhile, the BBB value provides insight into the drug's permeability in crossing the blood-brain barrier. BBB values are categorized into three groups based on absorption strength: high (>2), moderate (2 to 0.1), and low (<0.1) [21]

Based on Table 6, curcumin has a BBB value of 0.58, so it can be concluded that this compound has moderate or sufficient ability to cross the blood-brain barrier. It should be noted that not all drugs

need to cross the blood-brain barrier optimally if the therapeutic target of the drug is not related to the central nervous system (*CNS*). Predicting distribution via P-glycoprotein (P-gp) inhibition and substrate parameters is important because P-gp is one of the drug transporters involved in the absorption and elimination of various drug compounds. P-gp also functions in regulating drug levels in plasma and peripheral tissues, thereby influencing its efficacy and potential toxicity. Additionally, toxicity aspects must be considered in drug candidate selection to ensure safe use. The toxicity parameters used include the Kroes TTC, mutagenic potential, and carcinogenic potential [17]. As shown in Table 6, the test compound is safe, non-mutagenic, and non-carcinogenic.

Thus, curcumin demonstrates potential anti-inflammatory activity through its ability to interact with several key targets in the inflammatory pathway, indicating a multi-target mechanism of action. These interactions support curcumin's role in inhibiting inflammatory mediators that contribute to the inflammatory process. However, when compared to celecoxib, curcumin's binding affinity is still relatively lower, reflecting that the strength of the interaction between the ligand and the receptor is not yet as optimal as that of the comparator drug.

Conclusion

Based on the results of the study, it can be concluded that curcumin has potential as a promising anti-inflammatory candidate based on an in silico approach. Predictions of biological activity indicate that curcumin exhibits a tendency to act as an anti-inflammatory agent with a higher Pa value compared to Pi. *Drug-likeness* test results show that curcumin meets all criteria of Lipinski's Rule of Five, making it a strong candidate for an oral drug. *Molecular docking* analysis indicates that curcumin is capable of interacting with key inflammatory targets—namely COX-2, TNF-α, and NF-κB—exhibiting sufficiently high *binding affinities* and valid conformations (RMSD = 0). Ligand-receptor interactions involve various types of non-covalent bonds, such as hydrogen bonds and hydrophobic interactions, which contribute to the stability of the complex. Additionally, ADMET analysis results indicate that curcumin has a favorable safety profile, although its bioavailability is limited due to high plasma protein binding. Thus, curcumin holds potential as *a lead compound* in the development of natural-based anti-inflammatory drugs, with the need for further optimization of its pharmacokinetic aspects.

Reference

[1] K. Dirga, M. Buana, and R. Mauludin, "Sumber Potensial Agen Antiinflamasi Tanaman Indonesia dari Usada Taru Premana dan Perkembangan Formulasinya: Telaah Pustaka," *J. Penelit. Farm. Indones.*, Vol. 13, No. 1, Pp. 1–11, 2024.

[2] R. Patala, V. Anggi, I. Paula, and N. Sakina, "Activity of Analgesic and Anti-Inflammatory Nanoemulsion of Ethanol Extract of *Curcuma longa* L. By In Vivo," *J. Pharm. Sci.*, Vol. 6, No. 4, Pp. 1795–1803, 2023.

[3] S. A. Rohaniah, D. Mulyanti, and T. M. Fakih, "Uji Aktivitas Antiinflamasi Senyawa Turunan Asetogenin Pada Daun Sirsak (*Annona muricata* L .) Terhadap Reseptor Siklooksigenase-2 (Cox-2) Secara in Silico," *Bandung Conf. Ser. Pharm.*, Vol. 3, No. 2, Pp. 217–224, 2023.

[4] L. Setyaningrum, L. S. Anggraeni, L. F. Ningrum, S. N. Hasana, M. N. Hafid, M. R. Usman, and E. F. Yanti, "Molekuler Docking Senyawa Daun Sirih Merah Terhadap Prostaglandin H2 Synthase - 1 Sebagai," *J. Ilm. Farm. Akad. Farm.*, Vol. 8, No. 1, Pp. 86–94, 2025.

[5] N. Ramadhani and S. A. Sumiwi, "Aktivitas Antiinflamasi Berbagai Tanaman Diduga Berasal dari Flavonoid," *Farmaka*, Vol. 14, No. 2, Pp. 111–123, 2016.

[6] L. N. Syamsi, Syamsudin, E. Mumpuni, W. Widowati, O. Nursanti, and R. Qodriah, "Analisis in Silico Senyawa Curcuma Xanthorrhiza dan *Moringa oleifera* Sebagai Kandidat Antiinflamasi," *J.*

Syifa Sci. Clin. Res., Vol. 7, No. 1, Pp. 57–66, 2025.

[7] A. Wilapangga and M. Arif, "Analisis Prediksi Potensi Farmakokinetik dan Toksisitas pada Senyawa Kurkumin dari Extrak *Curcuma aromatica* Secara in Silico," *J. Pharm. Umr.*, Vol. 2, No. 2, Pp. 156–165, 2026.

[8] M. R. Afnani, R. Ahmad, Z. Aziz, and M. F. Widiapratama, "Potential of Active Compounds in Water Hyacinth Leaves (*Eichhornia crassipes*) As Candidates for Leukemia Drugs: An in Silico Approach," *J. Bio-Digital Forensic Informatics*, Vol. 1, No. 1, Pp. 1–12, 2026.

[9] A. Nurfadillah, N. I. Amir, and S. Ramadhani, "In Silico Studies and ADMET Predictions Diterpenoid Compound *Andrographis paniculata* (Burm . F .) Nees as Inhibitor of Streptococcus Mutans," *Hayyan J.*, Vol. 2, No. 1, Pp. 12–24, 2025.

[10] B. K. Nadzifah, T. I. Sulistyowati, and S. G. Waluyo, "Analisis Potensi Antioksidan Senyawa Bioaktif Buah *Stelechocarpus burahol* Secara in Silico dan Rekomendasi Pemanfaatannya," *Semin. Nas. Sains, Kesehatan, dan Pembelajaran 5*, Vol. 5, No. 1, Pp. 767–774, 2025.

[11] A. M. Abdillah, A. Juniansa, and T. Kok, "In Silico Study of the Bioactivity Potential of Teak Leaves (*Tectona grandis*) for the Treatment of Chronic Inflammation-Related Disease," *Indones. J. Biotechnol. Biodivers.*, Vol. 9, No. 2, Pp. 53–61, 2025.

[12] F. A. Muslikh, N. Nahdhia, D. Susilawati, F. Sari, S. Nugroho, W. Werdiningsih, and D. Basuki, "Prediksi druglikeness dan potensi aktivitas biologis senyawa metabolit sekunder daun asam jawa (*Tamarindus indica*)," *J. Sintesis: Pen. Sains, Ter. dan Anal.*, vol. 6, no. 1, pp. 49–60, 2025.

[13] D. R. A. S. Syafrizayanti, Y. N. Alfath, R. Riga, and A. J. Kusnanda, "Studi Molecular Docking dan Farmakoinformatika Senyawa Kurkumin dan Arturmeron pada Tumbuhan Kunyit (*Curcuma longa* Linn.) Sebagai Antihipertensi," *J. Sains Dan Kesehat. (J. Sains Kes.)*, Vol. 6, No. 5, Pp. 657–665, 2024.

[14] N. Maftucha, R. Manalu, R. Amelia, P. Cordia, and R. Bupu, "Potensi Senyawa Turunan Xanton dari Kulit Buah Manggis (*Garcinia mangostana* L .) Sebagai Inhibitor Protein Mycobacterium Tuberculosis: Studi in Silico," *Pharm. J. Indones.*, Vol. 7, No. 2, Pp. 123–128, 2022.

[15] D. I. Dinata, M. I. Peni, and A. Asnawi, "Identification of Angiotensin Receptor Blocker Ii Ligands from Gotu Kola (*Centella asiatica* L .) Extract: An in Silico," *Indones. J. Pharm. Sci. Technol.*, Vol. 5, No. 2, Pp. 196–206, 2023.

[16] A. D. Pitaloka, C. Y. Nurhijriah, H. A. Musyaffa, and A. M. Azzahra, "Molecular Docking of Chemical Constituents of Dayak Onion (*Eleutherine palmifolia* (L.) Merr) Towards Vhr Receptors as Candidates for Cervical Anticancer Drugs," *Indones. J. Biol. Pharm.*, Vol. 3, No. 2, Pp. 83–95, 2023.

[17] A. Nurfadillah, J. Bunga, N. Indah, D. Toba, and A. I. Asdar, "Antituberculosis Potential of *Caesalpinia sappan* Bioactive Compounds using in Silico Approach," *Al Ulum J. Sains Dan Teknol.*, Vol. 12, No. 1, Pp. 1–10, 2026.

[18] T. Pantsar and A. Poso, "Binding Affinity via Docking: Fact and Fiction," *Molecules*, Vol. 23, No. 8, Pp. 1–11, 2018, Doi: 10.3390/Molecules23081899.

[19] G. G. Ferenczy and M. Kellermayer, "Contribution of Hydrophobic Interactions to Protein Mechanical Stability," *Comput. Struct. Biotechnol. J.*, Vol. 20, Pp. 1946–1956, 2022, Doi: 10.1016/J.Csbj.2022.04.025.

[20] R. Syahputra, D. Utami, And W. Widyaningsih, "Molecular Docking Study of Tyrosinase Enzyme Inhibition Activity of Sweet Potatoes (*Ipomoea batatas* L. Lam)," *Pharmacon Jurnalfarmasi Indones.*, Vol. 19, No. 1, Pp. 21–34, 2022.

[21] I. N. Lestari, E. Renada, D. Juliana, K. Iswara, And S. Sukmara, "Molecular Docking Study of Antidiabetic Compounds on Ppar -γ Receptors from Ciplukan (*Physalis angulata* Linn.) Plants," *Indones. J. Biol. Pharm.*, Vol. 3, No. 1, Pp. 1–9, 2023.