Indonesian Journal of Pharmaceutical Education (e-Journal) 2025; 5 (3): 343– 356
ISSN: 2775- 3670 (electronic)
Journal Homepage: http://ejurnal.ung.ac.id/index.php/ijpe/index
DOI: 10.37311/ijpe.v5i3.33673

In Silico Evaluation of Tamarind Leaf Flavonoids Targeting ERα as Anti-Breast Cancer Agents Using Molecular Docking

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Article Info:
Received: 26 July 2025
in revised form: 20 September 2025
Accepted: 24 October 2025
Available Online: 31 October 2025

Keywords:
Tamarindus indica;
ERα;
Molecular docking;
Flavonoids;
Breast cancer

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ABSTRACT

Breast cancer particularly the estrogen receptor–positive (ER+) luminal subtype remains a major cause of mortality. Although tamoxifen is the gold-standard therapy, resistance and adverse effects motivate the discovery of new agents. This study identified tamarind (*Tamarindus indica* L.) leaf flavonoids with potential ERα inhibition using in-silico molecular docking. Twenty-two ethanol-extract compounds were screened by Lipinski's Rule of Five, docked to ERα (PDB: 1SJ0) with PyRx/AutoDock Vina, and analyzed in Discovery Studio; redocking of the native ligand yielded an RMSD of 0.68 Å, and tamoxifen served as the reference ligand. Four flavonoids exhibited strong ERα binding relative to tamoxifen (−9.1 kcal/mol): apigenin (−9.2), chrysin (−8.9), cynaroside (−8.4), and kaempferol (−8.0), forming stabilizing hydrogen bonds and hydrophobic contacts while largely complying with Lipinski criteria. These findings indicate that tamarind-derived flavonoids—particularly apigenin and chrysin—are promising ERα-targeting candidates and warrant in vitro/in vivo validation for anti-breast cancer development.

How to cite (APA 6th Style):
Rahmadani,A.,Putri,C.N.,Wachidah,A.K.,Kholifah,E. (2025). *In Silico Evaluation of Tamarind Leaf Flavonoids Targeting ERa as Anti-Breast Cancer Agents Using Molecular Docking. Indonesian Journal of Pharmaceutical Education (e-Journal), 5*(3), 343-356.

ABSTRAK

Kanker payudara, terutama subtipe luminal dengan reseptor estrogen positif (ER+) tetap menjadi penyebab utama mortalitas. Meskipun tamoksifen merupakan terapi standar, resistensi dan efek samping mendorong penemuan agen baru. Penelitian ini mengidentifikasi flavonoid daun asam jawa (*Tamarindus indica* L.) yang berpotensi menghambat ERα melalui pemodelan in silico. Sebanyak 22 senyawa dari ekstrak etanol disaring menggunakan Aturan Lima Lipinski, didocking ke ERα (PDB: 1SJ0) dengan PyRx/AutoDock Vina, dan dianalisis menggunakan Discovery Studio; validasi redocking terhadap ligan native menghasilkan RMSD 0,68 Å, dengan tamoksifen sebagai ligan referensi. Empat flavonoid menunjukkan ikatan kuat pada ERα dibandingkan tamoksifen (−9,1 kcal/mol), yakni apigenin (−9,2), krisin (−8,9), sinarosida (−8,4), dan kaempferol (−8,0), dengan pembentukan ikatan hidrogen dan interaksi hidrofobik yang stabil serta sebagian besar memenuhi kriteria Lipinski. Temuan ini menunjukkan bahwa flavonoid daun asam jawa, terutama apigenin dan krisin berpotensi sebagai kandidat penarget ERα dan memerlukan validasi in vitro/in vivo untuk pengembangan antikanker payudara.

Kata Kunci: *Tamarindus indica*; ERα; Molecular docking; Flavonoid; Kanker payudara

1. Introduction

Cancer is a genetic disease that arises from DNA damage, disrupting normal cell-cycle regulation and driving uncontrolled cell proliferation [1],[2]. Globally, it remains a leading cause of death; in Indonesia, breast cancer is among the most prevalent malignancies. According to GLOBOCAN 2020, breast cancer accounted for 68,858 new cases (16.6% of 396,914 total new cancers), with the luminal estrogen receptor–positive (ER+) subtype representing a substantial proportion of diagnoses [3]. Endocrine therapy provides clinical benefit for many ER-positive tumors, yet its effectiveness varies across populations and disease settings [4],[5],[6].

Tamoxifen a selective estrogen receptor modulator (SERM)—has long served as a mainstay for ER-positive breast cancer by antagonizing ERα signaling [4],[7],[8]. However, prolonged exposure can lead to de novo or acquired resistance, which undermines therapeutic efficacy and contributes to treatment failure and mortality. Proposed mechanisms include mutations in ESR1, co-regulator alterations, and activation of alternative growth pathways, highlighting the need for new ERα-directed chemotypes that may circumvent or delay resistance [9],[10],[11],[12].

Natural products continue to attract interest as sources of chemically diverse and potentially safer anticancer agents with multi-target activity [13],[14]. *Tamarindus indica* L. (tamarind) has been reported to possess anticancer properties, and ethanol extract of tamarind leaves inhibited proliferation of ER-positive MCF-7 cells (IC_{50} 128.63 µg/mL), suggesting a possible ER-related mechanism that merits target-focused interrogation [15]. Building on this rationale, in silico approaches offer a cost and time efficient way to explore ligand–receptor recognition at atomic resolution and to prioritize plant-derived compounds acting on ERα before laboratory validation [16],[17],[18],[19].

Accordingly, this study investigates tamarind leaf constituents as prospective ERα modulators through a streamlined virtual-screening workflow. We profiled 22 ethanol-extract compounds for drug-likeness using Lipinski's Rule of Five, performed molecular docking against ERα (PDB: 1SJ0) with PyRx/AutoDock Vina, analyzed poses in Discovery Studio, internally validated the protocol by redocking the native ligand (RMSD = 0.68 Å), and benchmarked results against tamoxifen to prioritize leads for subsequent experimental confirmation.

2. Methods

Tools and Materials

We used Discovery Studio, ChemDraw, PyMOL, and PyRx (AutoDock Vina/Open Babel). Compound structures were retrieved from PubChem; the ERα structure (PDB: 1SJ0) was obtained from the Protein Data Bank; physicochemical profiling used SwissADME.

Lipinski's Rule of Five Analysis

Two-dimensional structures of 22 ethanol-extract constituents were prepared in ChemDraw and evaluated in SwissADME for molecular weight, logP, and hydrogen-bond donors/acceptors according to Lipinski's criteria [20].

Ligand Preparation

Ligand 3D structures were downloaded from PubChem (PDB), energy-minimized with Open Babel in PyRx, and exported as PDBQT for docking [21].

Protein Preparation

The ERα structure (PDB: 1SJ0) was cleaned by removing crystallographic waters and the native ligand, hydrogens were added, and the receptor was saved in PDB/PDBQT formats to preserve direct ligand–receptor contacts at the active site [21].

Docking Validation

Redocking of the native ligand reproduced the crystallographic pose with **RMSD = 0.68 Å**, indicating a valid protocol (RMSD < 2.0 Å) [22].

Molecular Docking and Scoring

Receptor/ligand files were converted to PDBQT in PyRx, the grid box was centered on the active site, and docking was performed using AutoDock Vina; binding affinities (kcal/mol) and best poses were recorded [21],[23],[24].

Interaction Analysis and Visualization

Top-ranked poses were analyzed in Discovery Studio to visualize 2D/3D contacts (hydrogen bonds, π–π/π-alkyl, hydrophobic) and to annotate key ERα residues.

Data Analysis

More negative binding affinity indicates stronger predicted interaction; test ligands were compared against tamoxifen as the reference to contextualize relative binding strength [24].

3. Results and Discussion

Lipinski Rule of Five Analysis

Leaves of *Tamarindus indica* L. are a rich source of bioactive phytochemicals with reported anticancer potential. In this study, 22 constituents from the ethanol extract were profiled for drug-likeness according to Lipinski's Rule of Five.

Table 1. Results of the Lipinski Rule of Five Analysis

Ligand	Molecular Weight (≤500 Dalton)	Log P (≤5)	Hydrogen Bond	
			Donor (≤5)	Acceptor (≤10)
Apigenin	270.24	2.41	3	5
Chrysin	254.24	2.71	2	4
Cynaroside	448.38	-0.40	7	11
Kaempferol	286.24	2.30	4	6
Galangin	270.24	2.59	3	5
1,3,6-tri-O-galloyl-beta-D-glucose	636.47	-0.27	11	18
Pueraria Glycoside	432.38	-0.06	7	10
Adenosine	267.24	-1.98	5	8
Palmitic Acid	256.42	5.55	1	2
2-Amino-1,3,4-Octadecanetriol	317.51	3.11	5	4
L-Phenylalanine	165.19	0.64	3	3
L-Arginine	174.20	-1.54	7	6
2-(3- Methoxyphenyl) Acetamide	165.19	0.72	2	3
Hordenine	165.23	1.49	1	2
Methyl Isonicotinate	137.14	0.86	0	3
L-Pipecolinic Acid	129.16	0.21	2	3
L-Glutamic Acid	147.13	-0.73	4	5
L-Isoleucine	131.17	0.44	3	3
DL-Stachydrine	144.19	0.30	1	2
D-Proline	115.13	-0.17	2	3
Betaine	117.15	-1.55	0	2
Choline	104.17	-0.31	1	1

The results, presented in Table 1, revealed that 18 compounds adhered to the Lipinski Rule of Five criteria, while four exhibited violations. Pueraria Glycoside and L-Arginine violated one rule (hydrogen donor ≥5), Cynaroside violated two rules (hydrogen donor ≥5 and hydrogen acceptor ≥10), while 1,3,6-tri-O-galloyl-beta-D-glucose violated three rules (molecular weight ≥500 Da, hydrogen donor ≥5, and hydrogen acceptor ≥10). A drug compound is said to be able to penetrate the cell membrane if it meets the requirements of at least two Lipinski Rule of Five rules. Notably, 1,3,6-tri-O-galloyl-beta-D-glucose exceeded the limits for molecular weight (>500 Da), hydrogen bond donors (>5), and acceptors (>10), suggesting poor oral bioavailability and potential toxicity due to limited membrane permeability and inefficient excretion[25], [26].

Flavonoid compounds such as Apigenin and Chrysin demonstrated excellent compliance with RO5, exhibiting molecular weights below 500 Da, moderate Log P values (2.41 and 2.71, respectively), and optimal hydrogen bonding characteristics. These properties align with their known bioavailability and therapeutic potential, as reported in studies on flavonoid-based drug development[26]. In contrast, Cynaroside and Pueraria Glycoside, despite their promising binding affinities, showed deviations in hydrogen bonding capacity, which may necessitate structural modifications to enhance their drug-like properties[27].

Molecular Docking Validation

To ensure the reliability of our docking methodology, the native ligand (E4D) was redocked into the binding site of the estrogen receptor alpha (ERα; PDB ID: 1SJ0). The resulting root-mean-square deviation (RMSD) of 0.68 Å (**Figure 1**) confirmed the accuracy of our computational approach, as values below 2.0 Å are generally considered acceptable for validating docking protocols[28]. The close alignment between the docked and crystallographic ligand poses further supported the robustness of our methodology. These results are in agreement with prior studies where low RMSD values were correlated with high predictive accuracy in molecular docking experiments[27].

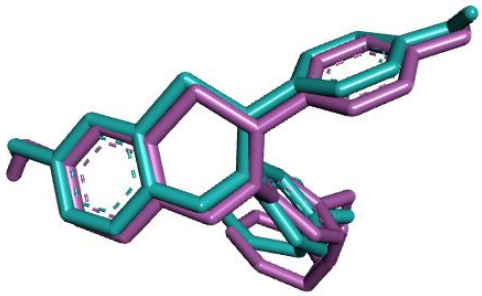

Figure 1. Superposition of the native ligand's crystallographic pose (purple) and the redocked pose (blue) within the estrogen receptor alpha (ERα) binding site (PDB ID: 1SJ0). The alignment yields an RMSD of **0.68 Å**, supporting the validity of the docking protocol

Binding Affinity and Molecular Interactions

The activity of the compound can be determined by docking the control ligand (tamoxifen) and test ligands (22 compounds) to the ERα receptor (ID 1SJ0). Tamoxifen, the reference drug, exhibited a binding affinity of –9.1 kcal/mol. Its complex structure contains an amino group, ethyl group, and benzene ring [29]. The interaction analysis revealed that tamoxifen established a hydrogen bond with CYS A:530, while additional stabilization was provided by hydrophobic interactions with residues ALA A:350, LEU A:525, ILE A:424, LEU A:346, MET A:388, LEU A:387, and PHE A:404. These interactions highlight tamoxifen's well-established antagonistic mechanism, in which hydrogen bonding secures its orientation and hydrophobic contacts enhance stability within the ERα binding pocket.

We interpret docking scores relative to tamoxifen, prioritizing ligands that combine more negative affinities with conserved ERα contacts. Relative to tamoxifen (−9.1 kcal/mol), apigenin achieved a slightly more favorable binding affinity (−9.2 kcal/mol) while preserving a comparable interaction pattern at the ERα pocket—combining multiple hydrogen bonds (e.g., ARG A:394, GLY A:521) with extensive hydrophobic contacts (ALA A:350, PHE A:404). Chrysin (−8.9 kcal/mol) lacked conventional hydrogen bonds yet compensated through dense hydrophobic/π-alkyl contacts around ALA A:350/LEU A:525/MET A:388, indicating a tamoxifen-like occupancy driven by aromatic stacking. Kaempferol (−8.0 kcal/mol) and Cynaroside (−8.4 kcal/mol) also engaged key residues; however, Cynaroside's RO5 deviations may impact drug-likeness despite favorable interactions. Together, these features rationalize why apigenin and Chrysin emerge as the most promising ERα-binding leads among the screened flavonoids [29],[30],[31].

Table 2. Binding affinity scores of ligands docking results against estrogen receptor alpha (ERα) protein (PDB ID: 1SJ0)

Ligand	Binding Affinity (kcal/mol)	Hydrogen Bonds (Residue:Chain:ID)	Hydrophobic/π-Interactions
Apigenin	−9.2	ARG A:394; GLY A:521; LEU A:387	ALA A:350; ILE A:424; PHE A:404; LEU A:525; LEU A:346; LEU A:391; MET A:388
Tamoxifen	−9.1	**CYS A:530**	ALA A:350; LEU A:525; ILE A:424; LEU A:346; MET A:388; LEU A:387; PHE A:404
Chrysin	−8.9	–	ILE A:424; ALA A:350; PHE A:404; LEU A:525; LEU A:346; LEU A:391; MET A:388
Cynaroside	−8.4	GLU A:353; LYS A:449; GLU A:323; LEU A:327; ARG A:394; ASP A:321; TRP A:393	ILE A:326
Kaempferol	−8.0	–	LEU A:525; LEU A:384; ALA A:350; LEU A:387; CYS A:530; LEU A:391; LEU A:346
Galangin	−7.9	LEU A:387	LEU A:525; LEU A:384; ALA A:350; CYS A:530; LEU A:346; LEU A:391
1,3,6-tri-O-galloyl-β-D-glucose	−7.8	GLU A:443; GLU A:353; GLU A:323; ASP A:321; TRP A:393; GLY A:442	LEU A:320
Pueraria Glycoside	−7.5	–	LEU A:525; LEU A:536; ALA A:350
Adenosine	−7.2	LYS A:449; GLU A:353; ARG A:394; TRP A:360	ILE A:326; PRO A:324
Palmitic Acid	−6.4	LEU A:387	ILE A:424; LEU A:346; LEU A:384; MET A:421; LEU A:525; ALA A:350; PHE A:404; MET A:388; HIS A:524
2-Amino-1,3,4-Octadecanetriol	−6.2	GLU A:353; LEU A:346; PHE A:404	ILE A:424; LEU A:525; ALA A:350; MET A:421; LEU A:384; HIS A:524
L-Phenylalanine	−6.1	PRO A:325; LEU A:327; ARG A:394	PRO A:324
L-Arginine	−6.0	ILE A:386; GLU A:353; PRO A:325	–
2-(3-Methoxyphenyl) Acetamide	−5.9	GLU A:353	ALA A:350; LEU A:346; PHE A:404; MET A:343

Hordenine	−5.9	ARG A:394	PHE A:404; ALA A:350; LEU A:346; LEU A:391; LEU A:349
Methyl Isonicotinate	−5.5	–	GLU A:353; PRO A:324
L-Pipecolinic Acid	−5.3	GLU A:353; GLY A:390	PRO A:324; MET A:357
L-Glutamic Acid	−5.3	GLU A:353; LYS A:449; ARG A:394	–
L-Isoleucine	−5.1	ILE A:386; LYS A:449; GLU A:353	PRO A:324; MET A:357; TRP A:360
DL-Stachydrine	−4.9	–	–
D-Proline	−4.8	GLU A:353	–
Betaine	−4.0	GLU A:353; ILE A:386; ILE A:387	–
Choline	−3.6	GLU A:353; ILE A:386; ILE A:387	–

Note: *More negative values indicate stronger predicted affinity; compared to tamoxifen (−9.1 kcal/mol)*

The docking analysis revealed significant variations in the binding affinities of the tested compounds toward ERα (**Table 2**). Among them, apigenin emerged as the most promising candidate, with a binding affinity of –9.2 kcal/mol, slightly surpassing that of tamoxifen. Unlike tamoxifen, which relies on a single hydrogen bond, apigenin engaged in multiple hydrogen bonds with key residues (ARG A:394, GLY A:521, and LEU A:387) in addition to hydrophobic interactions with ALA A:350 and PHE A:404. This broader hydrogen-bonding network suggests that apigenin may achieve comparable or even greater binding stability than tamoxifen, despite lacking the bulky side chains characteristic of selective estrogen receptor modulators (SERMs).

These results are consistent with previous studies, which reported that hydroxyl group positioning is critical for ERα binding and that polyphenolic scaffolds can effectively mimic or compete with endogenous estrogens [30]. Prior work has also identified residues such as LEU A:387 and PHE A:404 as recurring interaction hotspots for flavonoids within ERα. The ability of apigenin to reproduce these interactions while achieving a binding affinity comparable to tamoxifen supports its potential as a natural ERα modulator. Moreover, these findings align with experimental evidence showing apigenin's ability to inhibit ERα-positive breast cancer cells through receptor antagonism and induction of cell cycle arrest and apoptosis [31].

Chrysin, another flavonoid compound, displayed a strong binding affinity (-8.9 kcal/mol) but lacked conventional hydrogen bonds, relying instead on hydrophobic interactions with residues like ILE A:424 and MET A:388 as shown in **Figure 2**. This observation aligns with previous studies suggesting that Chrysin's bioactivity may be more dependent on its hydrophobic character than on specific hydrogen bonding [32]. Similarly, Cynaroside exhibited a notable binding affinity (-8.4 kcal/mol) facilitated by six hydrogen bonds, highlighting the importance of glycoside moieties in stabilizing ligand-receptor interactions. However, its Lipinski Rule of Five violations may limit its therapeutic applicability unless structural optimizations are employed. Docking visualization shows the formation of six conventional hydrogen bonds at the residues GLU A:353, LYS A:449, GLU A:323, LEU A:327, ARG A:394, and ASP A:321, as well as one donor hydrogen bond at TRP A:393. This bond occurs between the hydrogen atoms of Cynaroside with the oxygen and nitrogen atoms of the ERα amino acids, contributing significantly to the stability of the complex. In addition, the hydrophobic interaction with

the ILE A:326 residue also maintains the stability of the bond between Cynaroside and ERα.

Kaempferol exhibited notable binding characteristics with a binding affinity of -8.0 kcal/mol. As a typical flavonoid structure, its two benzene rings connected by a heterocyclic chain with hydroxyl groups facilitated multiple interactions with the receptor. The compound formed a Van der Waals interaction with THR A:347 and established significant hydrophobic contacts with LEU A:525, LEU A:384, ALA A:350, LEU A:387, CYS A:530, LEU A:391, and LEU A:346 as shown in **Figure 2.** These extensive hydrophobic interactions contribute to the stability of the kaempferol-ERα complex by minimizing aqueous interference, a characteristic feature of flavonoid-protein interactions [33].

Galangin, a structural analog of kaempferol, showed slightly reduced binding affinity (-7.9 kcal/mol) but similar interaction patterns. The hydroxyl group at position 3 appeared crucial for its hydrogen bond formation with LEU A:387 as shown in **Figure 2**, while its hydrophobic interactions mirrored those of kaempferol. This observation aligns with previous structure-activity relationship studies demonstrating the importance of hydroxylation patterns in flavonoid bioactivity.

The polyphenolic compound 1,3,6-tri-O-galloyl-beta-D-glucose displayed unique binding characteristics (-7.8 kcal/mol). Despite its larger molecular size, it formed multiple hydrogen bonds through its galloyl groups with GLU A:443, GLU A:353, GLU A:323, and ASP A:321 residues. However, its binding energy was somewhat compromised compared to smaller flavonoids, likely due to steric constraints within the binding pocket.

Pueraria Glycoside (-7.5 kcal/mol) and Adenosine (-7.2 kcal/mol) demonstrated moderate binding affinities through distinct mechanisms. The glycoside moiety of Pueraria Glycoside appeared to participate in hydrogen bonding, while Adenosine's ribose sugar and purine base engaged in multiple hydrogen bond interactions. These results are consistent with known structure-activity relationships for ERα ligands, where both aromaticity and hydrogen bonding capacity contribute to binding [34].

The study also identified several compounds with moderate (Palmitic Acid, -6.4 kcal/mol; 2-Amino-1,3,4-Octadecanetriol, -6.2 kcal/mol) to weak (Betaine, -4.0 kcal/mol; Choline, -3.6 kcal/mol) binding affinities. The saturated hydrocarbon chains of Palmitic Acid and 2-Amino-1,3,4-Octadecanetriol facilitated hydrophobic interactions but lacked the aromatic rings typically associated with strong ERα binding. This observation supports the established pharmacophore model for ERα ligands, which emphasizes the importance of aromatic systems [35],[36].Amino acid derivatives (L-Phenylalanine, L-Arginine, L-Glutamic Acid, L-Isoleucine) showed particularly weak binding, despite their potential for hydrogen bond formation. This suggests that while hydrogen bonding is important, the presence of aromatic systems is critical for high-affinity ERα binding, as demonstrated by the superior performance of flavonoid compounds [35],[36].

The comprehensive analysis of ligand-receptor interactions revealed several critical structural features that govern binding affinity to the estrogen receptor alpha (ERα). Foremost among these was the presence of aromatic systems, particularly the characteristic flavonoid scaffold composed of two benzene rings connected by a heterocyclic pyran ring. This structural motif demonstrated optimal geometry for establishing extensive hydrophobic interactions within the receptor's binding pocket, primarily through contacts with residues LEU A:525, LEU A:391, and ALA A:350 as shown in **Figure 2**. The planar nature of these flavonoid compounds allowed for

maximal surface contact with the hydrophobic regions of the binding site, a feature that has been consistently observed in high-affinity ERα ligands.

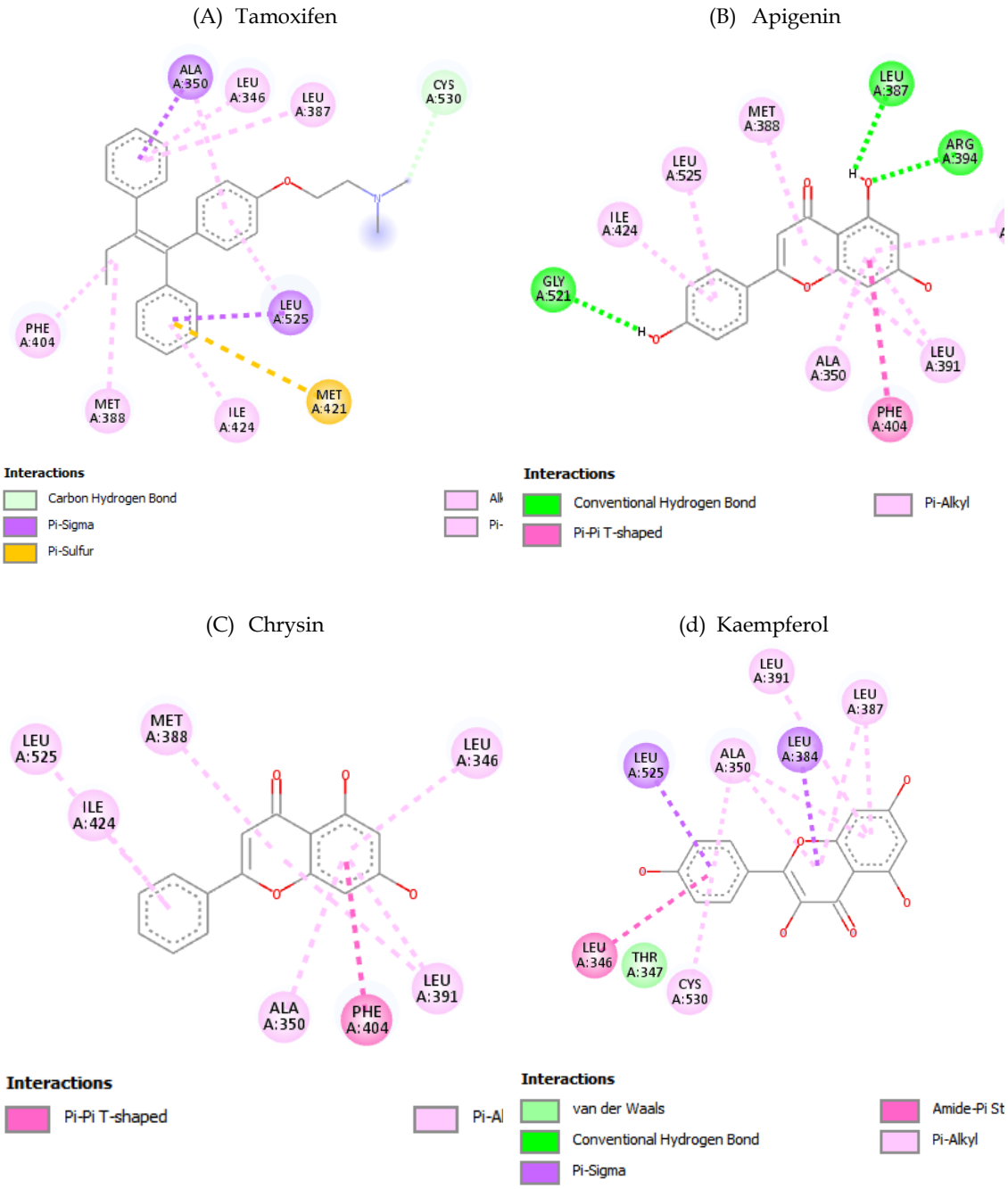

Figure 2. Two-dimensional interaction diagrams for (A) tamoxifen, (B) apigenin, (C) chrysin, and (D) kaempferol within the estrogen receptor alpha (ERα) binding pocket, highlighting conventional hydrogen bonds (green), π–π/π-alkyl contacts (purple/magenta), and hydrophobic interactions (orange) at key residues (e.g., ALA A:350, LEU A:525, PHE A:404, CYS A:530). This visualization supports the comparative analysis of ligand pose stability versus tamoxifen

Equally important was the positioning of hydroxyl groups on the flavonoid backbone. Our results demonstrated that specific hydroxylation patterns significantly

influenced binding affinity through the formation of crucial hydrogen bonds. Notably, the 3-hydroxyl group in galangin formed a stable hydrogen bond with LEU A:387, while the 4'-hydroxyl group common to many flavonoids interacted with GLU A:353. These observations align with previous structure-activity relationship studies that have identified these positions as pharmacophoric points for ERα binding [36]. The hydrogen bonding network not only contributed to binding energy but also helped properly orient the ligands within the binding pocket, maximizing favorable interactions.

The study also revealed important size constraints for optimal binding. Moderately sized flavonoids with molecular weights between 250-300 Da, such as apigenin (270.24 Da) and kaempferol (286.24 Da), showed superior binding compared to both smaller compounds and larger polyphenols. This size range appears to represent the ideal balance between sufficient interaction surface area and the ability to fit comfortably within the binding cavity. Larger molecules like 1,3,6-tri-O-galloyl-beta-D-glucose (636.47 Da) experienced steric hindrance despite their potential for multiple interactions, while smaller compounds lacked the necessary structural features for strong binding.

These structural determinants collectively explain the superior performance of certain flavonoid compounds in our binding studies. The combination of an optimal aromatic scaffold, strategic hydroxyl group placement, and appropriate molecular size creates an ideal pharmacophore for ERα interaction, consistent with the known characteristics of successful ER modulators. These findings provide valuable guidance for the future design or selection of natural product-derived compounds for targeting ERα, suggesting that flavonoid structures with these key features represent promising candidates for further development as potential therapeutic agents.

This study has several limitations that should be acknowledged. First, the computational nature of the investigation, while valuable for initial screening, requires subsequent experimental validation to confirm the biological relevance of the observed interactions. Second, the research focused exclusively on ERα (PDB ID: 1SJ0), which may not fully capture the complexity of estrogen receptor interactions in vivo. Additionally, the study did not consider important pharmacokinetic factors such as absorption, distribution, metabolism, and excretion (ADME) properties that would significantly influence the therapeutic potential of these compounds. The Lipinski Rule of Five analysis provided preliminary insights into drug-likeness, but more comprehensive ADME predictions would be valuable for future work. The Lipinski Rule of Five analysis provided preliminary insights into drug-likeness, but it is important to note that satisfying Lipinski's criteria does not guarantee adequate bioavailability. Natural products such as flavonoids often exhibit poor solubility, low membrane permeability, or extensive first-pass metabolism despite appearing "drug-like" on paper. Conversely, compounds with certain Lipinski violations may still demonstrate acceptable bioavailability through alternative mechanisms. Therefore, while docking provides useful predictions of binding affinity, biological efficacy can only be confirmed through subsequent in vitro and in vivo validation studies

Future studies are recommended to address these limitations by incorporating molecular dynamics simulations to better understand binding stability over time, investigating interactions with other estrogen receptor isoforms, and validating findings through in vitro and in vivo experimental models. Such improvements would provide more comprehensive insights into the therapeutic potential of these compounds and enhance the translational relevance of the findings. Furthermore, structure-activity relationship studies could explore chemical modifications to optimize both binding

affinity and pharmacokinetic properties of the most promising compounds identified in this work.

4. Conclusion

This in-silico study indicates that tamarind-derived flavonoids particularly apigenin and chrysin are the most promising ERα-binding candidates based on docking scores relative to tamoxifen and conserved contacts at key residues (e.g., ALA A:350, LEU A:525, PHE A:404, CYS A:530). The binding patterns highlight complementary hydrophobic interactions and, for apigenin, a broader hydrogen-bonding network that may underpin pose stability. Given the computational nature of these findings, experimental validation is required, including ER-positive cell-based assays, basic ADME/RO5 assessment, and where relevant molecular dynamics prior to in vivo studies.

Acknowledgements :

The authors acknowledge financial support from the Institute for Research and Community Service (LPPM), Sultan Agung Islamic University (Grant No. 69/B.1/SA-LPPM/VII/2023), which made this study possible.

Conflict of Interest:

The authors declare no conflict of interest related to this study.

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