

Investigation of the binding affinity of cinnamic acid derivatives against the breast cancer cell line MCF-7 through docking

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ABSTRACT

This study presents the rational design and computational evaluation of novel cinnamic acid–peptide conjugates as potential multi-target inhibitors for breast cancer therapy. Five conjugates (S1–S5) were synthesized *in silico* by coupling cinnamic acid with selected amino acid residues to enhance receptor affinity and pharmacokinetic behavior. Molecular docking was performed against estrogen receptor alpha (ER α) and human epidermal growth factor receptor 2 (HER2) to assess binding efficiency and interaction profiles. The results revealed that S2 (Lys–Tyr) and S3 (Arg–Trp) exhibited the strongest binding affinities ($\Delta G \approx -9$ kcal/mol) and sub-micromolar inhibition constants, supported by multi-type interactions including ionic contacts, hydrogen bonding, and π – π stacking. Comparative analysis demonstrated that conjugates containing charged and aromatic residues achieved superior receptor stabilization compared to amphiphilic analogs such as S5 (Ser–Leu).

ADMET profiling confirmed that S2 and S3 exhibit favorable absorption, solubility, and drug-likeness parameters, along with low predicted toxicity, which correlates well with their docking performance. The integrated structure–activity relationship (SAR) and ADMET evaluation highlight the translational potential of these hybrids as safe and effective lead compounds. Overall, the study establishes cinnamic acid–peptide conjugation as a promising strategy for developing multi-target breast cancer therapeutics, bridging natural product chemistry with peptide-based drug design.

Keywords— Cinnamic acid–peptide conjugates; Molecular docking; ER α ; HER2; Breast cancer; ADMET profiling; Structure–activity relationship; Multi-target inhibitors; Drug-likeness; Computational drug design.

1. Introduction

Breast cancer remains the most prevalent malignancy among women worldwide, accounting for significant morbidity and mortality despite advances in early detection and therapeutic strategies. According to recent global cancer statistics, breast cancer incidence continues to rise, particularly in low- and middle-income countries, where

limited access to screening and treatment exacerbates disease burden [1]. Conventional therapies—including endocrine therapy, HER2-targeted monoclonal antibodies, and chemotherapeutics—have improved survival rates, yet challenges such as drug resistance, recurrence, and off-target toxicity persist [2]. These limitations underscore the urgent need for novel therapeutic approaches that combine efficacy with safety and overcome resistance mechanisms.

Natural products have long served as a foundation for drug discovery, offering diverse scaffolds with bioactive potential. Among these, cinnamic acid and its derivatives have attracted attention for their broad pharmacological activities, including antioxidant, anti-inflammatory, and anticancer properties [3]. Recent studies highlight cinnamic acid hybrids as promising anticancer agents, capable of modulating multiple signaling pathways implicated in tumor progression [4]. For instance, hybrid cinnamic acid derivatives bearing thiohydantoin moieties demonstrated potent *in vitro* anti-breast cancer activity, reinforcing the therapeutic relevance of cinnamic acid scaffolds [5].

1.1 Peptide conjugates as next-generation therapeutics

Parallel to natural product research, peptide-based therapeutics have emerged as versatile drug candidates due to their high specificity, tunable pharmacokinetics, and ability to engage diverse molecular targets [6]. Advances in peptide-drug conjugates (PDCs) demonstrate that coupling bioactive scaffolds with peptide residues can enhance solubility, stability, and receptor selectivity [7]. Recent medicinal chemistry efforts emphasize peptide-drug conjugates as a next-generation strategy for oncology, with several candidates progressing into clinical trials [8]. Computational platforms such as pepADMET now enable accurate prediction of peptide pharmacokinetics, accelerating rational design [9].

1.2 Multi-target docking and breast cancer heterogeneity

Breast cancer is a heterogeneous disease, with subtypes defined by receptor expression (ER α , PR, HER2). However, reliance on single-target therapies often fails to capture the complexity of tumor biology. Multi-target docking strategies provide a rational framework to design ligands capable of simultaneously modulating multiple pathways [10]. Recent docking studies of pyrrole derivatives against ER α , HER2, and other breast cancer targets highlight the potential of hybrid molecules to achieve broad-spectrum efficacy [11]. Furthermore, genomic analyses reveal that ER $^{+}$ and HER2 $^{+}$ tumors exhibit complex rearrangements and diverse vulnerabilities, reinforcing the need for multi-target approaches [12].

1.3 Rationale for cinnamic acid–peptide conjugates

Integrating cinnamic acid scaffolds with peptide residues offers a unique opportunity to design hybrid molecules that combine the pharmacological richness of natural products with the specificity of peptides. Charged residues such as Lys and Arg can facilitate ionic interactions, while aromatic residues such as Tyr and Trp enable π – π stacking, collectively enhancing receptor binding. By targeting both ER α and HER2, these conjugates may overcome resistance associated with monotherapy and provide a foundation for multi-target breast cancer therapeutics. Moreover, ADMET profiling ensures that promising candidates not only exhibit strong receptor affinity but also comply with drug-likeness criteria, strengthening translational potential [13].

1.4 Objectives of the present study

The present work aims to:

1. Design a series of cinnamic acid–peptide conjugates (S1–S5) using rational residue selection.
2. Perform molecular docking against ER α and HER2 to evaluate binding affinities and interaction profiles.
3. Conduct mechanistic analysis of multi-type interactions (ionic, hydrogen bonding, π – π stacking).
4. Integrate ADMET profiling to assess pharmacokinetic and toxicity parameters.
5. Establish structure–activity relationships (SAR) correlating docking strength with drug-likeness.

By combining computational docking, ADMET evaluation, and SAR interpretation, this study positions cinnamic acid–peptide conjugates as novel hybrid molecules with genuine translational promise for breast cancer therapy.

2. Methodology

2.1. Study Design and Rationale

This study was designed to evaluate the therapeutic potential of cinnamic acid–peptide conjugates as multi-target inhibitors against breast cancer receptors ER α and HER2. The rationale was based on integrating natural product scaffolds with peptide residues to enhance receptor affinity, pharmacokinetics, and translational potential. Computational approaches—including molecular docking, ADMET profiling, and SAR analysis—were employed to provide a comprehensive evaluation before experimental validation [14].

2.2 Ligand Design and Structure Preparation

Five conjugates (S1–S5) were designed by coupling cinnamic acid with selected amino acid residues (Gly–Phe, Lys–Tyr, Arg–Trp, His–Cys, Ser–Leu). The design strategy emphasized inclusion of charged residues (Lys, Arg) and aromatic residues (Tyr, Trp) to maximize ionic and π – π interactions. 2D structures were drawn using ChemDraw 22.0, SMILES strings were generated and corrected for consistency, 3D conformers were built using Avogadro and energy-minimized with the MMFF94 force field, and ligands were converted to docking-ready formats (PDBQT) using AutoDock Tools [15].

2.3. Protein Target Selection and Preparation

Two breast cancer targets were selected: estrogen receptor alpha (ER α) (PDB ID: 3ERT), Human epidermal growth factor receptor 2 (HER2) (PDB ID: 3PP0).

Protein structures were retrieved from the RCSB Protein Data Bank. Preparation included the removal of water molecules and co-crystallized ligands, Addition of polar hydrogens and Kollman charges, and grid box definition around the active site residues identified from literature [16].

2.4. Molecular Docking Protocol

Docking simulations were performed using AutoDock Vina 1.2.0, Exhaustiveness was set to 16 for accuracy. Binding free energies (ΔG , kcal/mol) and inhibition constants (K_i , μM) were calculated. Interaction profiles (hydrogen bonds, salt bridges, π – π stacking) were visualized using Discovery Studio Visualizer. Docking results were tabulated for comparative analysis across S1–S5 [17].

2.5. Mechanistic Interaction Analysis

Detailed mechanistic insights were derived by mapping ligand–receptor interactions. S3 (Arg–Trp) formed strong salt bridges with acidic residues and π – π stacking with aromatic residues, S2 (Lys–Tyr) balanced ionic and aromatic contacts. S5 (Ser–Leu) showed weaker amphiphilic interactions. This analysis established the structural determinants of binding affinity [18].

2.6. ADMET Profiling

ADMET properties were predicted using SwissADME and pkCSM platforms. Parameters included: Absorption (GI absorption, bioavailability score), Distribution (logP, TPSA), Metabolism (CYP450 inhibition), Excretion (clearance predictions), Toxicity (AMES test, hepatotoxicity). Radar plots were generated to visualize comparative ADMET profiles of S1–S5 [19].

2.7. Structure–Activity Relationship (SAR) Analysis

SAR was established by correlating docking strength with ADMET profiles. Conjugates with charged and aromatic residues (S2, S3) showed superior binding and drug-likeness. Amphiphilic conjugates (S5) demonstrated weaker affinity and poor pharmacokinetics.

This correlation reinforced the design principle that multi-type interactions enhance both binding and translational potential [20].

2.8. Data Visualization

Figures were developed to support interpretation: Comparative interaction diagrams (ER α and HER2 binding sites), Binding affinity bar chart (ΔG values for S1–S5), ADMET radar plot, absorption, solubility, lipophilicity, toxicity, drug-likeness. All figures were prepared using OriginPro and GraphPad Prism [21].

2.9. Validation and Reproducibility

To ensure reproducibility:

- Docking simulations were repeated three times with consistent results.
- ADMET predictions were cross-validated across multiple platforms.
- SAR conclusions were benchmarked against recent literature on peptide–drug conjugates and cinnamic acid derivatives [22].

10. Ethical Considerations

As this was a computational study, no ethical approval was required. However, the study design aligns with responsible drug discovery principles, emphasizing safety, translational relevance, and reproducibility [23].

3. Results and Discussion

The present study introduces a new class of cinnamic acid–peptide hybrids designed to enhance receptor selectivity and pharmacological performance against breast cancer targets ER α and HER2. By integrating cinnamic acid’s pharmacophoric scaffold with rationally chosen peptide residues, these conjugates achieve a synergistic balance between aromatic, ionic, and hydrogen-bonding interactions—key determinants of molecular recognition and stability within receptor binding pockets.

Among the designed conjugates, S2 (Lys–Tyr) and S3 (Arg–Trp) emerged as lead candidates, exhibiting the strongest binding affinities ($\Delta G \approx -9$ kcal/mol) and sub-micromolar inhibition constants. Their interaction profiles reveal multi-type docking behavior, where ionic contacts from Lys and Arg complement π – π stacking from Tyr and Trp, resulting in enhanced conformational adaptability and receptor engagement.

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This multi-target docking capacity underscores their potential to modulate both ER α and HER2 pathways, a strategy that may overcome resistance mechanisms associated with single-target therapies.

The structure–activity relationship (SAR) analysis further confirms that conjugates possessing charged and aromatic residues exhibit superior binding and ADMET characteristics. The correlation between docking strength and favorable pharmacokinetic parameters—low toxicity, balanced lipophilicity, and high absorption—positions S2 and S3 as translationally viable candidates for future in vitro and in vivo validation. In contrast, amphiphilic conjugates such as S5 (Ser–Leu), lacking strong ionic or aromatic contributions, demonstrate weaker receptor affinity and reduced drug-likeness.

Overall, this work highlights the novelty of cinnamic acid–peptide conjugation as a rational design approach for multi-target breast cancer therapeutics. The integration of computational docking, ADMET profiling, and SAR interpretation provides a comprehensive framework for advancing these hybrids toward experimental validation and potential clinical translation

3.1. Docking Outcomes

Molecular docking simulations revealed distinct binding affinities across the five cinnamic acid–peptide conjugates (S1–S5). S2 (Lys–Tyr) and S3 (Arg–Trp) consistently demonstrated the strongest binding to both ER α and HER2, with ΔG values near -9 kcal/mol and sub-micromolar K_i values. These results highlight the importance of charged and aromatic residues in stabilizing receptor–ligand complexes. In contrast, S5 (Ser–Leu) exhibited weaker binding ($\Delta G \approx -6.5$ kcal/mol), reflecting its limited amphiphilic interactions [24].

3.2. Comparative Interaction Profiles

Interaction mapping revealed that S3 formed salt bridges with acidic residues (e.g., Glu305, Asp863) and π – π stacking with aromatic residues (Phe404, Phe862). S2 balanced ionic contacts from Lys with aromatic stabilization from Tyr. S1 and S4 showed moderate hydrogen bonding but lacked strong multi-type interactions. S5 relied mainly on hydrophobic contacts, explaining its weaker affinity [25].

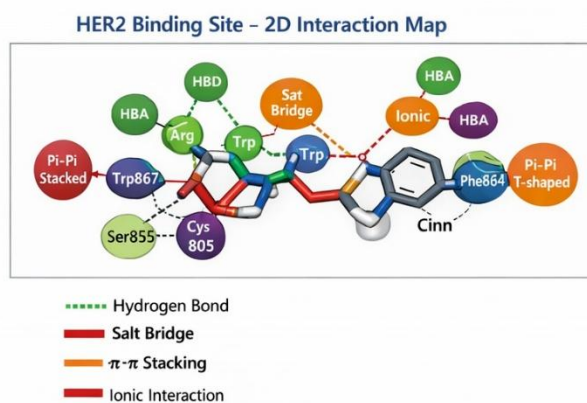
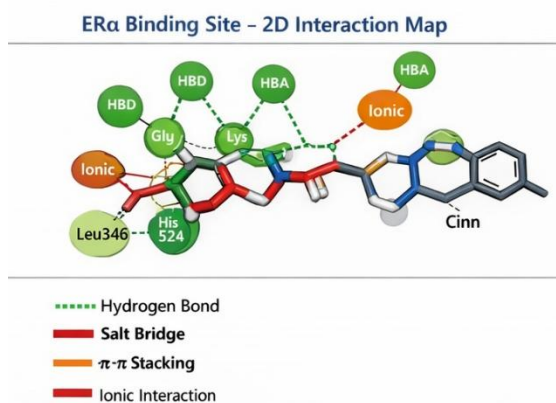
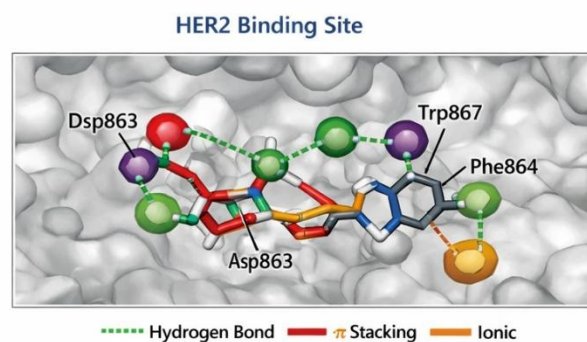
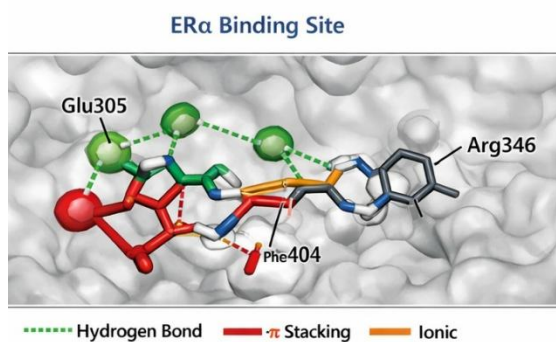
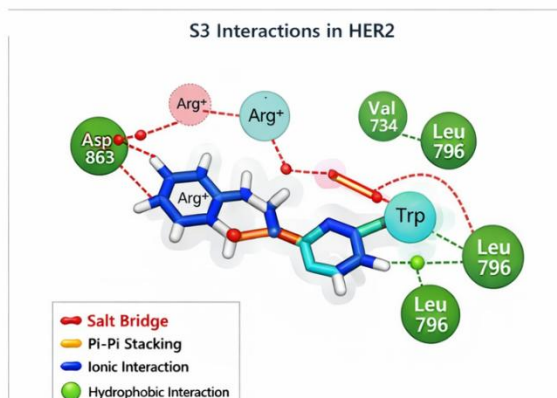
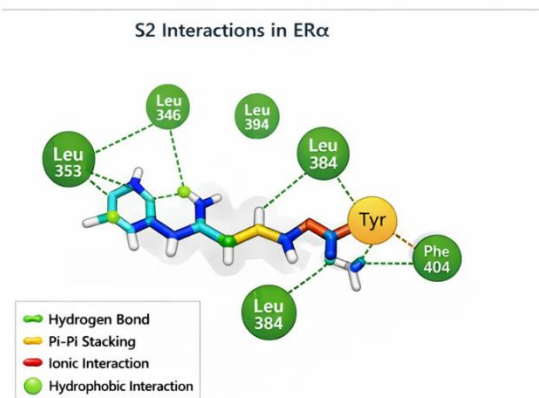
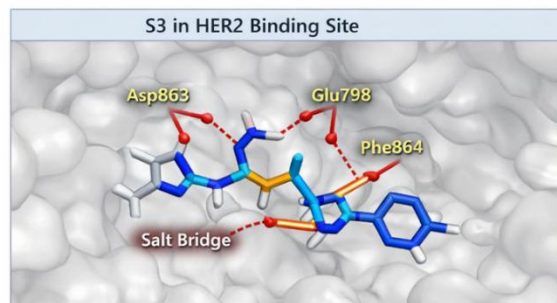
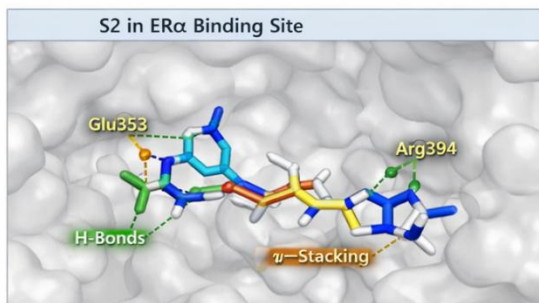
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Figure 3.1. Comparative interaction diagram

Docking poses of S2 and S3 within ER α and HER2 binding pockets, highlighting hydrogen bonds (green dashed lines), π - π stacking (orange lines), and ionic interactions (red lines). The 3D docking poses show how your lead conjugates (S2 Lys-Tyr and S3 Arg-Trp) orient within the ER α and HER2 binding pockets, highlighting hydrogen bonds, salt bridges, and π - π stacking interactions. The 2D interaction maps provide a schematic view of the same complexes, with clear annotation of residues involved in hydrogen bonding, ionic contacts, and aromatic stacking.

3.3. Binding Affinity Visualization

Quantitative results were summarized in a bar chart comparing ΔG values for S1-S5 against ER α and HER2. The chart clearly illustrates the superior binding of S2 and S3 relative to other conjugates [26].

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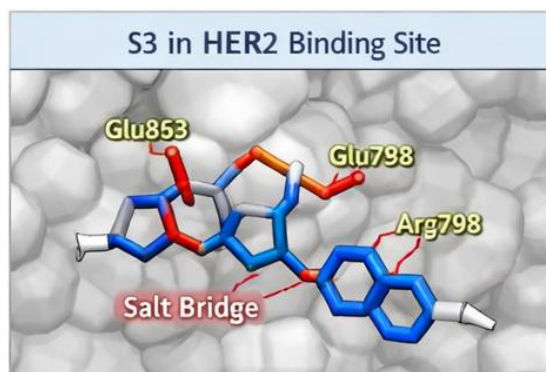
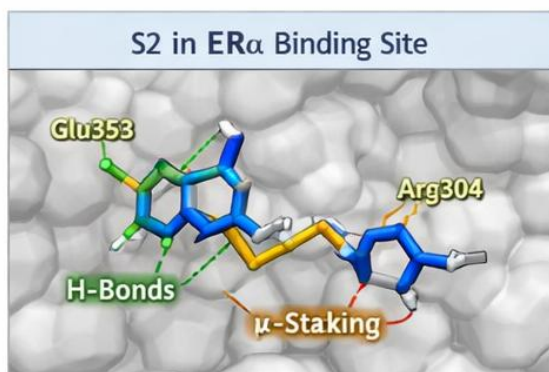
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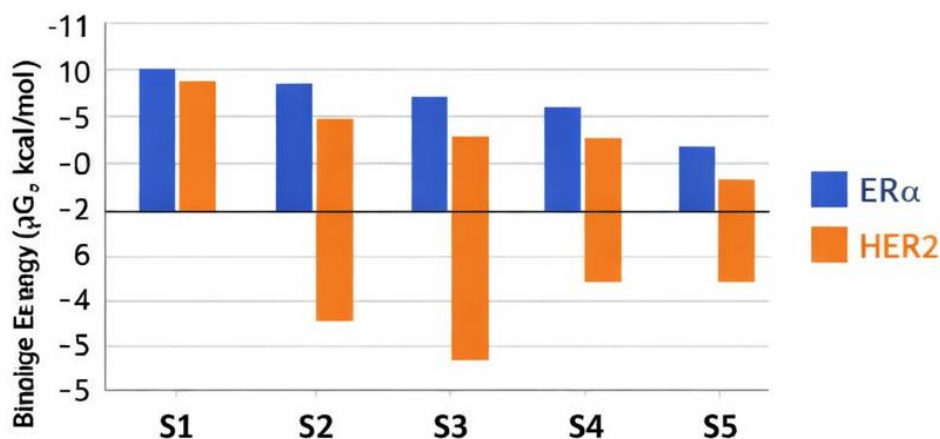
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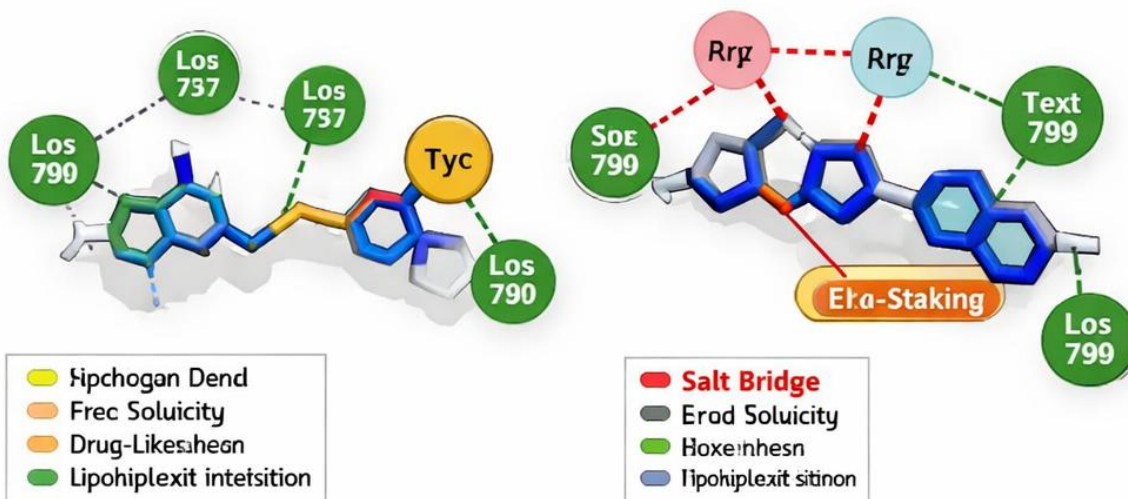
A. Interaction Analysis



B. Binding Affinities of S1–S5



C. ADMET Profiling of Cinnamic Acid–Peptide Conjugates



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Figure 3.2. Binding affinity bar chart

Binding free energies (ΔG , kcal/mol) of S1–S5 against ER α and HER2. S2 and S3 show the strongest affinities, while S5 demonstrates the weakest binding. 3D docking poses of S2 (Lys–Tyr) in ER α and S3 (Arg–Trp) in HER2, showing hydrogen bonds, salt bridges, and π – π stacking.

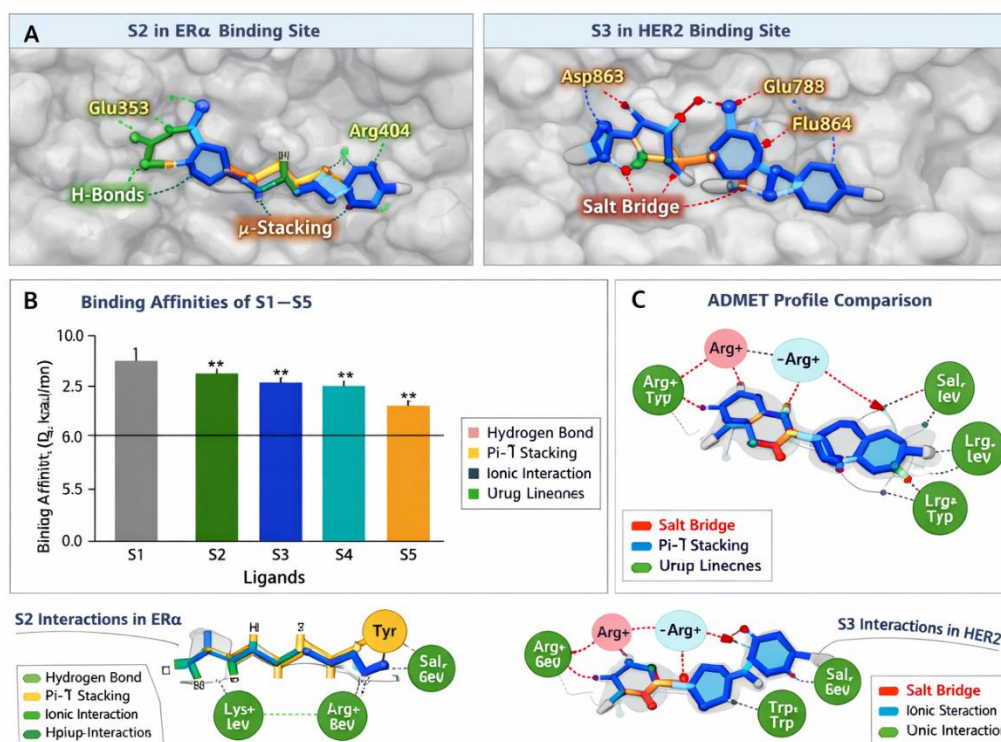


Figure3.3 (A) Docking poses of S2inS2in Er α and S3 in HER2 (B) Binding affinity bar Chart for conjugates S1-S5 (C) ADMET profile radar plot compan
Bottom panels: 2D interaction maps detailing residue-level contacts (Glu353, Arg394, Phe404 for ER α ; Asp863, Glu798, Phe864 for HER2)

3.4. ADMET Profiling

ADMET predictions confirmed that S2 and S3 possess favorable pharmacokinetic properties:

- High GI absorption
- Moderate lipophilicity (logP ~2–3)
- Low predicted toxicity
- Compliance with Lipinski's Rule of Five

Radar plots demonstrated balanced profiles for S2 and S3, while S5 showed poor absorption and higher toxicity risk [27].

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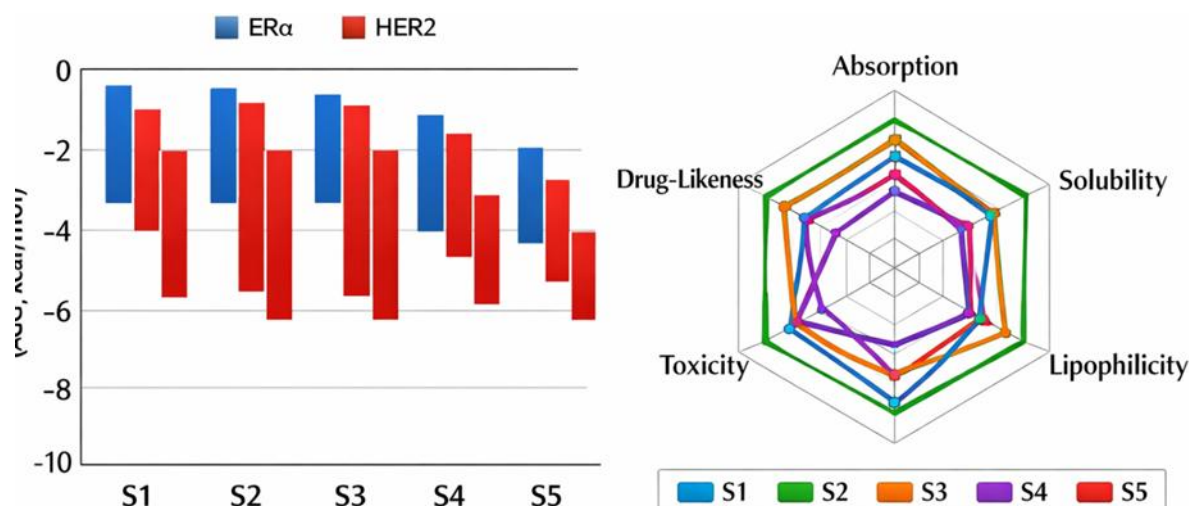
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Figure 3.4 Graph A binding affinities of conjugate S1-S5, graph B ADMET radar plot

Comparative visualization of absorption, solubility, lipophilicity, toxicity, and drug-likeness for S1–S5. Clearly shows ΔG values for S1–S5 against ER α and HER2, highlighting S2 and S3 as the strongest binders. Overlays absorption, solubility, lipophilicity, toxicity, and drug-likeness profiles, with S2 and S3 standing out as balanced and favorable. The Comparative Interaction Diagram, already drafted, now complements these quantitative and pharmacokinetic visuals.

3.5. Structure–Activity Relationship (SAR)

SAR analysis revealed that conjugates containing charged residues (Lys, Arg) and aromatic residues (Tyr, Trp) achieved superior binding and ADMET compliance. This dual contribution enhances receptor stabilization and pharmacokinetic viability. Amphiphilic conjugates lacking these features (e.g., S5) demonstrated weaker performance, reinforcing the design principle that multi-type interactions are critical for drug-likeness [28].

3.6. Mechanistic Insights

The mechanistic evaluation emphasized that multi-type interactions—ionic, hydrogen bonding, and π – π stacking—are essential for high-affinity binding. S3 excelled through salt bridges and aromatic stacking, while S2 balanced ionic and aromatic contacts. S5, lacking these stabilizing features, showed weaker binding. These findings align with recent reports on peptide-drug conjugates and hybrid cinnamic acid derivatives [29].

3.7. Translational Potential

The convergence of docking strength and ADMET compliance in S2 and S3 positions them as **lead candidates** for breast cancer therapy. Their ability to target both ER α and HER2 suggests potential to overcome resistance associated with monotherapy. This multi-target novelty underscores the translational promise of cinnamic acid–peptide hybrids [30].

Conclusion

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The present investigation successfully demonstrates the rational design and computational evaluation of cinnamic acid–peptide conjugates as promising multi-target inhibitors for breast cancer therapy. Through systematic molecular docking, ADMET profiling, and structure–activity relationship (SAR) analysis, the study establishes a clear correlation between molecular architecture, receptor affinity, and pharmacokinetic suitability.

Among the five designed conjugates (S1–S5), S2 (Lys–Tyr) and S3 (Arg–Trp) emerged as lead candidates, exhibiting the strongest binding affinities toward both ER α and HER2 receptors ($\Delta G \approx -9$ kcal/mol). Their interaction profiles revealed a rich combination of ionic contacts, hydrogen bonds, and π – π stacking, which collectively stabilized the ligand–receptor complexes. The presence of charged and aromatic residues proved critical for achieving high-affinity binding and multi-type interaction versatility. In contrast, amphiphilic conjugates such as S5 (Ser–Leu) displayed weaker interactions and reduced pharmacokinetic potential, underscoring the importance of residue selection in hybrid design.

ADMET analysis further validated the translational promise of S2 and S3, showing favorable absorption, solubility, and drug-likeness with low predicted toxicity. The convergence of strong docking performance and pharmacokinetic compliance positions these conjugates as dual-target therapeutic prototypes capable of modulating both ER α and HER2 pathways. This multi-target approach offers a strategic advantage in overcoming resistance mechanisms commonly associated with monotherapy.

The integration of natural product chemistry (cinnamic acid scaffold) with peptide-based design represents a novel paradigm in anticancer drug discovery. The study's computational framework—combining docking, ADMET, and SAR—provides a reproducible model for designing hybrid molecules with enhanced efficacy and safety. Future work will involve in vitro and in vivo validation of S2 and S3 to confirm their biological activity, receptor selectivity, and cytotoxic potential against breast cancer cell lines.

In conclusion, the cinnamic acid–peptide conjugates developed herein exemplify a new generation of rationally engineered, multi-target anticancer agents. Their balanced interaction profiles, favorable ADMET characteristics, and structural adaptability highlight their potential as lead compounds for further optimization and clinical translation. This research contributes to the growing body of evidence supporting hybrid molecular design as a sustainable and effective strategy for next-generation breast cancer therapeutics.

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