Pharmacy Reports

https://doi.org/10.51511/pr.73

RESEARCH ARTICLE

Open Access

Molecular docking analysis of guaiacin and chalcone from nutmeg (*Myristica fragrans*) as novel HSP90A inhibitors for skin cancer treatment

Muhammad Zhafran Arrafi, Ayu Sukma Cahyani, Nada Nikita Sitohang, Salsa Nabila Ahlika Ulya, Inggrid Anggraeni, Miranti Amalia, Gladys Ellnora Putri, Raihan M Dhiya Rajwa, Winni Nur Auli, Anjar Hermadi Saputro*

Department of Pharmacy, Faculty of Science, Institut Teknologi Sumatera, Indonesia

*Corresponding author: Jl. Terusan Ryacudu, Way Huwi, Jati Agung, South Lampung 35365, Lampung, Indonesia.
Email: anjar.saputro@fa.itera.ac.id

Abstract: Skin cancer represents one of the most prevalent malignancies globally, with Indonesia reporting the third highest incidence among cancer types. Despite advances in treatment, there remains a critical need for novel therapeutic agents. Heat Shock Protein 90 Alpha (HSP90A) has emerged as a promising target for cancer therapy due to its critical role in stabilizing oncogenic proteins. This study aimed to evaluate the potential of guaiacin and chalcone from nutmeg (*Myristica fragrans*) as HSP90A inhibitors for skin cancer treatment through computational analysis. Molecular docking was performed using AutoDock Tools with the HSP90A crystal structure (PDB ID: 2VCJ). The compounds were assessed for binding affinity, molecular interactions, and drug-likeness properties according to Lipinski's Rule of Five. Redocking validation yielded an RMSD of 1.24 Å, confirming protocol reliability. Guaiacin demonstrated promising binding affinity (-7.40 kcal/mol) with key hydrogen bonds to Asp93 and Lys58, while chalcone showed moderate affinity (-5.99 kcal/mol) with a single hydrogen bond to Thr184. Both compounds exhibited favorable drug-like properties with high predicted gastrointestinal absorption. Guaiacin emerges as a promising natural HSP90A inhibitor candidate with binding energy exceeding the stability threshold (-7.00 kcal/mol) and interactions with critical residues in the ATP-binding pocket, providing a foundation for further development of nutmeg-derived compounds as potential anticancer agents.

Keywords: HSP90A inhibitor, molecular docking, nutmeg, guaiacin, chalcone, skin cancer

Introduction

Skin is the outermost organ that serves as the primary protective barrier against environmental threats. Excessive ultraviolet (UV) radiation exposure represents a major risk factor for skin damage that can progress to skin cancer, one of the most common malignancies worldwide. Skin cancer incidence continues to rise globally, with particularly concerning rates in tropical regions where UV exposure is intense. In Indonesia, skin cancer ranks as the third most prevalent cancer type, highlighting the urgent need for improved therapeutic strategies [1].

Current treatments for skin cancer include surgery, radiation therapy, chemotherapy, and targeted therapy. Despite these options, challenges remain due to drug resistance, recurrence, and adverse effects associated with conventional therapies. This has driven the search for novel therapeutic agents with enhanced efficacy and reduced toxicity profiles [2]. Natural compounds derived from medicinal plants have gained significant attention as potential sources of anticancer agents due to their structural diversity and bioactivity.

Nutmeg (*Myristica fragrans* Houtt.), a spice used traditionally in various cultures for culinary and medicinal purposes, contains numerous bioactive compounds with therapeutic potential. Previous research has identified several secondary metabolites in nutmeg with anticancer properties, including guaiacin, a lignan with a complex structure containing two aromatic rings and four hydroxyl groups, and chalcone, a precursor of the flavonoid group with diverse biological activities [3]. These compounds have demonstrated antioxidant, cytotoxic, and anti-inflammatory properties, making them promising candidates for cancer therapy investigation.

Heat Shock Protein 90 Alpha (HSP90A) has emerged as an attractive molecular target for cancer therapeutics. As a highly conserved molecular chaperone, HSP90A plays a crucial role in protein folding, stabilization, and activation of numerous client proteins involved in signal transduction, cell proliferation, and survival pathways [4,5]. Many of these client proteins are oncogenic drivers implicated in cancer progression, including kinases, transcription factors, and hormone receptors [6]. In cancer cells, HSP90A is often overexpressed, supporting the increased protein folding demands of rapidly dividing cells and stabilizing mutated or overexpressed oncoproteins.

Inhibition of HSP90A disrupts its chaperone function, leading to the degradation of client proteins through the ubiquitin-proteasome pathway and ultimately resulting in cancer cell death through multiple cellular pathways. This makes HSP90A inhibition a promising strategy that can simultaneously target multiple oncogenic pathways [7]. The ATP-binding site in the N-terminal domain of HSP90A has been the primary focus for inhibitor development, as ATP binding and hydrolysis are essential for its chaperone function.

Molecular docking represents a computational approach for predicting the binding modes and affinities of small molecules to target proteins. This technique allows for the rapid screening of compounds and identification of promising candidates for further development, significantly accelerating the drug discovery process while reducing experimental costs [8]. By modeling the interactions between ligands and the target protein, molecular docking provides valuable insights into the structural basis of inhibition and guides the optimization of lead compounds.

The present study aims to evaluate the potential of guaiacin and chalcone from nutmeg as HSP90A inhibitors through molecular docking analysis. By examining their binding affinities, interaction patterns with key residues in the ATP-binding pocket, and drug-like properties, we seek to determine their potential as novel natural product-based HSP90A inhibitors for skin cancer treatment. This computational approach provides a foundation for understanding the structural basis of HSP90A inhibition by these compounds and guides further experimental investigations to develop new therapeutic strategies against skin cancer.

Methods

Preparation of protein target and ligands

The three-dimensional structure of HSP90A (PDB ID: 2VCJ) was obtained from the RCSB Protein Data Bank (https://www.rcsb.org/). Water molecules were removed, and hydrogen atoms were added using AutoDock Tools v1.5.7. The structure was then assigned Kollman charges and saved in .pdbqt format for docking simulations. We selected HSP90A as our target due to its established role in cancer cell survival and proliferation [9].

The native ligand (2EQ, C23H24CIN3O5) was extracted from the HSP90A-ligand complex for redocking validation. The 3D structures of guaiacin and chalcone were obtained from the PubChem database (https://pubchem.ncbi.nlm.nih.gov/) and optimized using the MMFF94 force field in Avogadro to obtain energetically favorable conformations. All ligands were prepared for docking by adding Gasteiger charges, merging non-polar hydrogens, and assigning rotatable bonds using AutoDock Tools.

Molecular docking

The grid box was centered on the ATP-binding site of HSP90A with dimensions of 27 × 26 × 26 $Å^3$, spacing of 0.375 Å, and grid center coordinates of x = 33.105, y = 8.164, z = 26.071. These parameters were selected to encompass the entire binding pocket based on preliminary analysis of the co-crystallized ligand position [10].

Docking simulations were conducted using the Lamarckian Genetic Algorithm implemented in AutoDock Tools v1.5.7. The protocol included 100 independent docking runs for each ligand with a population size of 150, maximum number of energy evaluations set to 2,500,000, and maximum number of generations set to 27,000. The docking protocol was first validated by redocking the native ligand and calculating the RMSD between the docked and crystal structure poses, with an acceptance threshold of ≤ 2.0 Å as recommended by previous studies [7].

ADMET property prediction

Absorption, distribution, metabolism, excretion, and toxicity (ADMET) properties were predicted using the SwissADME web tool (http://www.swissadme.ch/) to assess the drug-likeness of the compounds. The following parameters were calculated: molecular

Table 1. Physicochemical properties and ADMET predictions for test compounds

Compound	Molecular weight (g/mol)	LogP	HBA[1]	HBD[2]	TPSA[3]	Lipinski violation	GI absorption	BBB permeant
Guaiacin	328.40	4.08	4	2	58.92 Å	1 (XLOGP3>3.5)	High	No
Chalcone	208.26	3.47	1	0	17.07 Å	0	High	Yes

[1]Hydrogen bond acceptor
[2]Hydrogen bond donor
[3]Topological Polar Surface Area (Å^2)

Table 2. Binding energies and molecular interactions of ligands with HSP90A

Ligand	Binding energy (kcal/mol)	Number of hydrogen bonds	Key interacting residues
Native ligand	-8.99	4	Gly97, Met98, Thr184, Leu107
Guaiacin	-7.40	2	Asp93, Lys58
Chalcone	-5.99	1	Thr184

weight, octanol-water partition coefficient (LogP), number of hydrogen bond acceptors (HBA), number of hydrogen bond donors (HBD), and topological polar surface area (TPSA). Compliance with Lipinski's Rule of Five was evaluated to predict oral bioavailability [8]. Additionally, gastrointestinal absorption was predicted to assess the potential for oral administration of these compounds.

Interaction analysis

The docking results were visualized and analyzed using BIOVIA Discovery Studio 2021. Hydrogen bonds, hydrophobic interactions, and other non-covalent interactions between the ligands and HSP90A were identified and characterized based on their types, distances, and participating amino acid residues. Key interactions with residues known to be important for HSP90 function were specifically examined to understand the potential inhibitory mechanism [11].

Results

Validation of molecular docking

Redocking of the native ligand 2EQ into the ATP-binding site of HSP90A yielded an RMSD value of 1.24 Å compared to the crystallographic pose. This value is well below the accepted threshold of 2.0 Å, confirming the reliability of our docking protocol and parameter settings for predicting binding modes of test compounds.

Physicochemical properties and ADMET prediction

The physicochemical properties and ADMET predictions for guaiacin and chalcone are summarized in Table 1. Both compounds demonstrated favorable drug-like properties according to Lipinski's Rule of Five, with molecular weights below 500 Da (guaiacin: 328.40 g/mol; chalcone: 208.26 g/mol), appropriate LogP values (guaiacin: 4.08; chalcone: 3.47), and acceptable numbers of hydrogen bond donors and acceptors.

Guaiacin showed one violation of Lipinski's Rule (XLOGP3 > 3.5), while chalcone fully complied with all criteria. Both compounds were predicted to have high gastrointestinal absorption, suggesting good potential for oral bioavailability. Chalcone was predicted to be blood-brain barrier (BBB) permeant, while guaiacin was not, indicating different tissue distribution profiles.

Binding affinity and molecular interactions

The binding energies and key molecular interactions obtained from docking simulations are presented in Table 2. Docking scores attempt to correlate with binding free energy, so more negative scores suggest stronger binding [12]. The native ligand 2EQ exhibited the strongest binding to HSP90A with a binding energy of -8.99 kcal/mol. Among the test compounds, guaiacin showed promising binding affinity (-7.40 kcal/mol), while chalcone displayed moderate affinity (-5.99 kcal/mol).

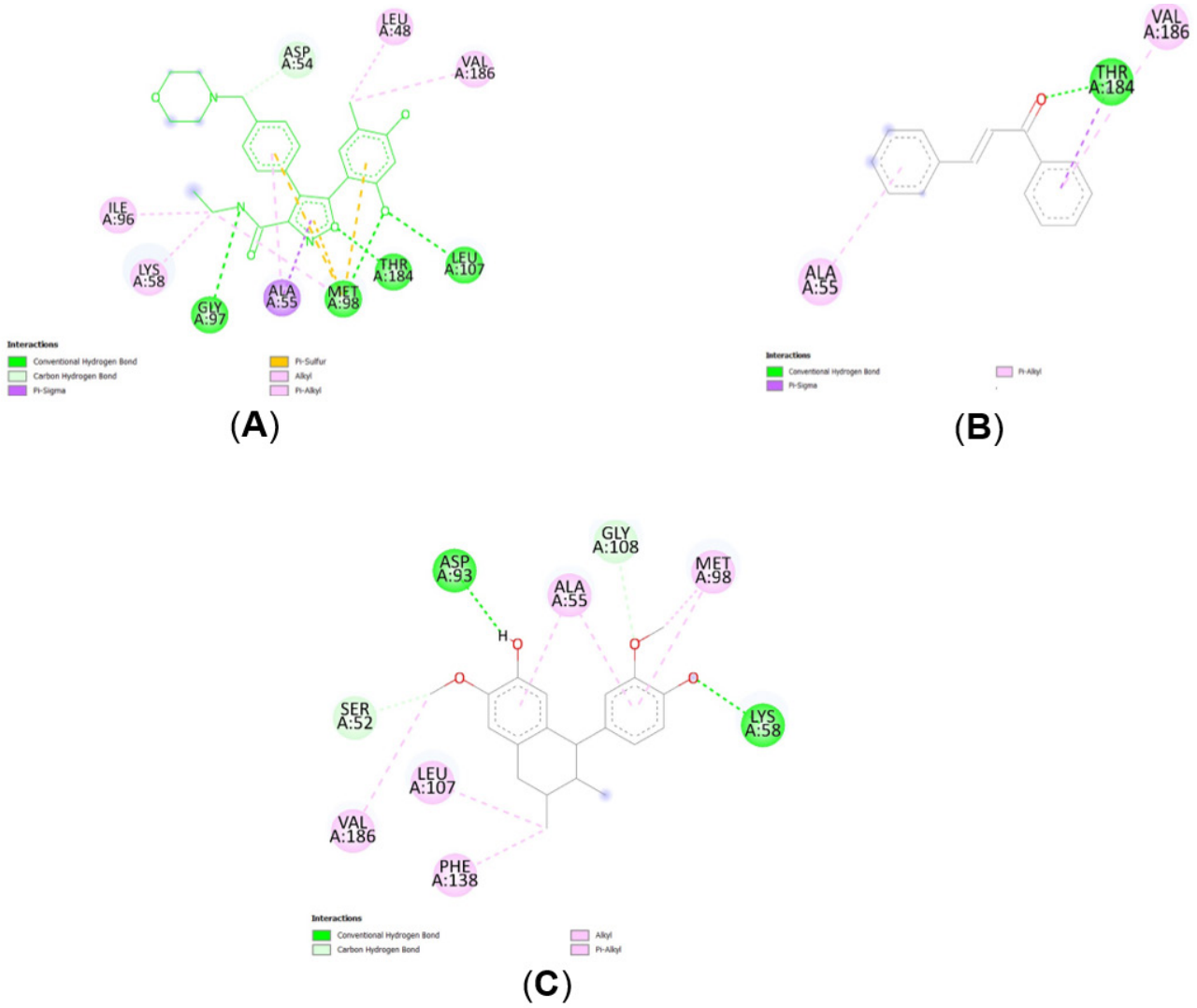

Figure 1. 2D visualization interaction of ligands docking with the HSP90A. (A) native ligand, (B) chalcone, (C) guaiacin.

Detailed analysis of molecular interactions revealed distinct binding patterns for each compound. The native ligand formed four hydrogen bonds with Gly97, Met98, Thr184, and Leu107, along with hydrophobic interactions with Ile96, Lys58, Ala55, Asp54, Leu48, and Val186. Guaiacin established two hydrogen bonds with Asp93 (bond distance: 2.11 Å) and Lys58 (bond distance: 1.95 Å), and formed hydrophobic interactions with Ala55, Gly108, Met98, Ser52, Val186, Leu107, and Phe138. Chalcone formed a single hydrogen bond with Thr184 (bond distance: 1.88 Å) and hydrophobic interactions with Ala55 and Val186.

Figure 1 illustrates the binding modes of the three compounds within the ATP-binding pocket of HSP90A. The binding pose of guaiacin overlapped significantly with that of the native ligand, occupying a similar region of the binding pocket, while chalcone displayed a somewhat different orientation.

Discussion

This study investigated the potential of guaiacin and chalcone from nutmeg as HSP90A inhibitors through molecular docking analysis. Our findings reveal differential binding affinities and interaction patterns that provide insights into their potential as anticancer agents.

The validation of our docking protocol, with an RMSD of 1.24 Å for the native ligand redocking, ensures the reliability of our results and is consistent with established standards in the field. Previous studies have indicated that RMSD values below 2.0 Å demonstrate good reproducibility of experimentally

determined binding modes [5,13]. This validation step is crucial for confidence in the predicted binding modes and affinities of our test compounds.

The binding energies obtained from our docking simulations indicate that guaiacin (-7.40 kcal/mol) forms a more stable complex with HSP90A compared to chalcone (-5.99 kcal/mol), though both exhibit lower affinity than the native ligand (-8.99 kcal/mol). This difference in binding affinity can be attributed to their structural differences and resulting interaction patterns.

Analysis of the molecular interactions provides further insight into the binding mechanisms of these compounds. Guaiacin forms hydrogen bonds with Asp93 and Lys58, both of which are crucial residues in the ATP-binding pocket of HSP90. Recent pharmacophore modeling studies have identified these residues, along with Ala55, Ile96, Met98, and Thr184, as key interaction points for effective HSP90 inhibition [11]. The interaction with Lys58 is particularly significant as this residue plays a central role in ATP binding and hydrolysis, which are essential for HSP90 chaperone function [9]. Disruption of this interaction by guaiacin suggests a potentially effective mechanism for inhibiting HSP90 activity.

Chalcone, despite its lower binding affinity, forms a hydrogen bond with Thr184, which is also involved in the ATP-binding mechanism of HSP90. This interaction is shared with the native ligand, indicating that chalcone may still interfere with ATP binding, albeit less effectively than guaiacin. The limited number of hydrogen bonds formed by chalcone (only one compared to two for guaiacin and four for the native ligand) likely contributes to its lower binding affinity. However, the interaction with Thr184 suggests that structural modifications to enhance binding at this position could improve its inhibitory potential.

The binding modes of our compounds can be compared to those of established HSP90 inhibitors such as geldanamycin and radicicol, which bind to the N-terminal ATP-binding pocket with binding energies typically ranging from -9 to -11 kcal/mol [14]. While our compounds show lower affinity than these optimized inhibitors, guaiacin's binding energy (-7.40 kcal/mol) is promising for a natural compound without pharmaceutical optimization. Natural products often serve as starting points for drug development, with chemical modifications enhancing their potency and pharmacokinetic properties [2].

From a structural perspective, guaiacin's superior binding can be attributed to its complex structure with two aromatic rings and four hydroxyl groups, providing multiple points for interaction with HSP90A. In contrast, chalcone's simpler structure limits its interaction potential. This structure-activity relationship provides valuable guidance for future optimization efforts. For instance, enhancing chalcone's structure with additional hydrogen bond donors or acceptors at positions that can interact with key residues like Asp93 and Lys58 might improve its binding affinity.

The ADMET predictions for our compounds suggest favorable drug-like properties, particularly for oral administration. Both compounds comply with Lipinski's Rule of Five, with guaiacin showing only one minor violation (XLOGP3 > 3.5). Recent studies have shown that approximately 20% of FDA-approved drugs violate at least one of Lipinski's criteria, suggesting that this minor violation does not necessarily preclude guaiacin's development potential [8]. The high predicted gastrointestinal absorption for both compounds indicates good potential for oral bioavailability, which is a significant advantage for patient compliance and treatment accessibility.

The differential BBB permeability prediction (chalcone: yes; guaiacin: no) suggests distinct tissue distribution profiles. While BBB permeability is not necessary for treating peripheral skin cancers, it could be relevant for potential applications in melanoma with brain metastases. Chalcone's predicted BBB permeability might offer an advantage in such cases, though this would require further investigation beyond the scope of our current study.

When compared to other natural product-based HSP90 inhibitors reported in the literature, guaiacin shows competitive binding properties. Recent studies have identified several plant-derived compounds, including flavonoids, withanolides, and diarylheptanoids, as potential HSP90 inhibitors with binding energies ranging from -6.5 to -8.5 kcal/mol [11,15]. Guaiacin's binding energy of -7.40 kcal/mol places it within this range, supporting its potential as a natural HSP90 inhibitor. Molecular dynamics studies on natural HSP90 inhibitors have also highlighted the importance of stable hydrogen bonding with key residues such as Asp93 and Lys58, which aligns with our findings for guaiacin [16].

The molecular mechanism by which guaiacin and chalcone might inhibit HSP90A can be inferred

from our results. By binding to the ATP-binding pocket and interacting with residues critical for ATP binding and hydrolysis, these compounds would likely interfere with the ATPase activity essential for HSP90 chaperone function. This would prevent the maturation and stabilization of client proteins involved in cancer cell survival and proliferation, leading to their degradation through the ubiquitin-proteasome pathway [6]. The resulting simultaneous disruption of multiple oncogenic signaling pathways could effectively induce cancer cell death while potentially overcoming resistance mechanisms that target single pathways.

Our study has certain limitations that should be acknowledged. Molecular docking provides static binding predictions and does not account for the dynamic nature of protein-ligand interactions in a physiological environment. Further studies, including molecular dynamics simulations [17,18], would provide valuable insights into the stability of these interactions over time. Additionally, experimental validation through in vitro and in vivo studies is necessary to confirm the predicted inhibitory activity and therapeutic potential of these compounds.

Conclusion

This study demonstrates that guaiacin from nutmeg (*Myristica fragrans*) shows promising potential as an HSP90A inhibitor for skin cancer treatment, with a binding energy (-7.40 kcal/mol) and important interactions with key residues Asp93 and Lys58 in the ATP-binding pocket. While chalcone exhibited moderate binding affinity (-5.99 kcal/mol), its interaction with Thr184 suggests it may still interfere with HSP90A function. Both compounds demonstrated favorable drug-like properties according to Lipinski's Rule of Five and high predicted gastrointestinal absorption, supporting their potential for pharmaceutical development. These findings provide a strong foundation for further investigations, including molecular dynamics simulations to evaluate binding stability and experimental validation through in vitro and in vivo studies. The identification of natural compounds with HSP90A inhibitory potential contributes to the development of novel anticancer agents from traditional medicinal plants like nutmeg, potentially offering new therapeutic options with improved efficacy and reduced toxicity for skin cancer treatment.

Acknowledgement

The authors thank to the Institut Teknologi Sumatera for the support provided.

Funding

None.

Author contribution

Conceptualization: AHS, MZA, WNA; Methodology: ASC, NNS, SNAU, IA, MA, GEP, RMDR, WNA; Investigation: WNA, AHS; Writing – Original Draft: MZA, ASC, NNS, SNAU, IA, MA, GEP, RMDR, AHS; Writing – Review & Editing: AHS.

Declaration of interest

The authors declare that none of them has any conflict of interest with any private, public, or academic party related to the information contained in this manuscript.

Received: June 6, 2024
Revised: April 30, 2025
Accepted: May 3, 2025
Published: May 4, 2025

References

1. Hardianto A, Qodir N, Roflin E, Indra B. Incidence and Characteristics of Skin Cancer Patients at Dr. Mohammad Hoesin General Hospital, Palembang. Indonesian Journal of Cancer. 2025;19: 90-95. https://doi.org/10.33371/ijoc.v19i1.1280
2. Atanasov AG, Waltenberger B, Pferschy-Wenzig E-M, Linder T, Wawrosch C, Uhrin P, et al. Discovery and resupply of pharmacologically active plant-derived natural products: A review. Biotechnology Advances. 2015;33: 1582-1614. https://doi.org/10.1016/j.biotechadv.2015.08.001
3. Ginting B, Mustanir M M, Nurdin N N, Maulidna M M, Murniana M M, Safrina S S. Evaluation of Antioxidant and Anticancer Activity of Myristica fragrans Houtt. Bark. PJ. 2021;13: 780-786. https://doi.org/10.5530/pj.2021.13.99
4. Xie X, Zhang N, Li X, Huang H, Peng C, Huang W, et al. Small-molecule dual inhibitors targeting heat shock protein 90 for cancer targeted therapy. Bioorganic Chemistry. 2023;139: 106721. https://doi.org/10.1016/j.bioorg.2023.106721
5. Saxena AK, Saxena S, Chaudhaery SS. Molecular modelling and docking studies on heat shock protein 90 (Hsp90) inhibitors. SAR and QSAR in Environmental Research. 2010;21: 1-20. https://doi.org/10.1080/10629360903560504

6. Trepel J, Mollapour M, Giaccone G, Neckers L. Targeting the dynamic HSP90 complex in cancer. Nat Rev Cancer. 2010;10: 537-549. https://doi.org/10.1038/nrc2887
7. Ferreira L, Dos Santos R, Oliva G, Andricopulo A. Molecular Docking and Structure-Based Drug Design Strategies. Molecules. 2015;20: 13384-13421. https://doi.org/10.3390/molecules200713384
8. Chen X, Li H, Tian L, Li Q, Luo J, Zhang Y. Analysis of the Physicochemical Properties of Acaricides Based on Lipinski's Rule of Five. Journal of Computational Biology. 2020;27: 1397-1406. https://doi.org/10.1089/cmb.2019.0323
9. Neckers L, Workman P. Hsp90 Molecular Chaperone Inhibitors: Are We There Yet? Clinical Cancer Research. 2012;18: 64-76. https://doi.org/10.1158/1078-0432.CCR-11-1000
10. Huang N, Shoichet BK, Irwin JJ. Benchmarking Sets for Molecular Docking. J Med Chem. 2006;49: 6789-6801. https://doi.org/10.1021/jm0608356
11. Lauria A, Ippolito M, Almerico AM. Inside the Hsp90 inhibitors binding mode through induced fit docking. Journal of Molecular Graphics and Modelling. 2009;27: 712-722. https://doi.org/10.1016/j.jmgm.2008.11.004
12. García-Ortegón M, Simm GNC, Tripp AJ, Hernández-Lobato JM, Bender A, Bacallado S. DOCKSTRING: Easy Molecular Docking Yields Better Benchmarks for Ligand Design. J Chem Inf Model. 2022;62: 3486-3502. https://doi.org/10.1021/acs.jcim.1c01334
13. Hyun SY, Le HT, Nguyen C-T, Yong Y-S, Boo H-J, Lee HJ, et al. Development of a novel Hsp90 inhibitor NCT-50 as a potential anticancer agent for the treatment of non-small cell lung cancer. Sci Rep. 2018;8: 13924. https://doi.org/10.1038/s41598-018-32196-6
14. Kyle Hadden M, Hill SA, Davenport J, Matts RL, Blagg BSJ. Synthesis and evaluation of Hsp90 inhibitors that contain the 1,4-naphthoquinone scaffold. Bioorganic & Medicinal Chemistry. 2009;17: 634-640. https://doi.org/10.1016/j.bmc.2008.11.064
15. Sakkiah S, Thangapandian S, John S, Kwon YJ, Lee KW. 3D QSAR pharmacophore based virtual screening and molecular docking for identification of potential HSP90 inhibitors. European Journal of Medicinal Chemistry. 2010; 45: 2132-2140. https://doi.org/10.1016/j.ejmech.2010.01.016
16. Rampogu S, Parate S, Parameswaran S, Park C, Baek A, Son M, et al. Natural compounds as potential Hsp90 inhibitors for breast cancer-Pharmacophore guided molecular modelling studies. Computational Biology and Chemistry. 2019;83: 107113. https://doi.org/10.1016/j.compbiolchem.2019.107113
17. AbdElmoniem N, H. Abdallah M, M. Mukhtar R, Moutasim F, Rafie Ahmed A, Edris A, et al. Identification of Novel Natural Dual HDAC and Hsp90 Inhibitors for Metastatic TNBC Using e-Pharmacophore Modeling, Molecular Docking, and Molecular Dynamics Studies. Molecules. 2023;28: 1771. https://doi.org/10.3390/molecules28041771
18. Zarguan I, Ghoul S, Belayachi L, Benjouad A. Plant-Based HSP90 Inhibitors in Breast Cancer Models: A Systematic Review. IJMS. 2024;25: 5468. https://doi.org/10.3390/ijms25105468