Indo. J. Chem. Sci. 13 (2) (2024)
Indonesian Journal of Chemical Science
http://journal.unnes.ac.id/sju/index.php/ijcs

Interaction of BCN OH (Bicyclononyn derivate) and Pertuzumab in the Development of Linker ADC (Antibody Drug Conjugation) Through Molecular Docking

Aden Dhana Rizkita[1]✉, Tris Ella Julita Gulo[1], Nisa Sri Wahyuni[1], Risma Eka Putri Selay[1], Nisa Alwia[1], Ayu Andini Rizki Lestari Rohman[1], Efriyana Oksal[2], Kumam Pinky Chanu[3], Asaduzzaman[4], Taufik Muhammad Faqih[5]

[1]Department of Pharmacy, Sekolah Tinggi Ilmu Kesehatan Bogor Husada, Road. Sholeh Iskandar No.4, Kedungbadak, District. Cereal Land, Bogor City, West Java 16164, Indonesia.
2Departement of Chemistry, Faculty of Mathematics and Natural Sciences, Universitas Palangkaraya, Road. Yos Sudarso Palangka Raya 73111, Central Kalimantan, Indonesia
[3]Ph.D. Program in Clinical Drug Development of Herbal Medicine, College of Pharmacy, Taipei Medical University, No.250, Wuxing St., Xinyi Dist., Taipei City 110, Taiwan
[4]Graduate Institute of Biomedical Optomechatronics Engineering, Taipei Medical University, No.301, Yuantong Rd., Zhonghe Dist., New Taipei City 235, Taiwan
[5]Department of Pharmacy, Faculty of Mathematics and Natural Sciences, Universitas Islam Bandung, Bandung, West Java, Indonesia

Article Info

Accepted : 14-03-2024

Approved : 20-06-2024

Published : 26-08-2024

Keywords:
ADC(antibody Drug Conjugate),
BCN-OH,
Linker,
Molecular Docking,
Pertuzumab

Abstract

Pertuzumab, a monoclonal antibody targeting Human Epidermal Growth Factor Receptor 2 (HER2), is pivotal in breast cancer therapy by inhibiting the critical heterodimerization process for tumor growth. Despite its efficacy, variable clinical responses prompt the search for enhanced treatments. Antibody Drug Conjugates (ADCs) merge antibody specificity with cytotoxic drugs, offering a promising therapeutic avenue. Bicyclononyn (BCN) emerges as a potent linker candidate due to its reactivity and stability. This study investigates BCN-OH, a BCN derivative, for conjugation with pertuzumab, identifying key residues via in silico docking simulations. The analysis reveals a robust interaction between pertuzumab and BCN-OH, exhibiting a binding affinity of -5.1 kcal/mol. Noteworthy residues involved include Cys4, Thr5, Gly6, and Thr7. Hydrogen bonds form between BCN-OH's hydroxyl group and Thr5 and Gly6 residues, while hydrophobic interactions occur with Cys4 and Thr7 residues. These interactions underscore the pertuzumab-BCN-OH complex's stability and specificity, endorsing its potential as an ADC linker for targeted cancer therapy. This study offers valuable insights into BCN-OH's conjugation with pertuzumab, facilitating the rational optimization of ADCs for enhanced breast cancer treatment outcomes

✉ Correspondence address:
Road. Sholeh Iskandar No.4, Kedungbadak, District. Cereal Land, Bogor City, West Java 16164, Indonesia.
E-mail: adhen@sbh.ac.id (choose Correspondence Style)

p-ISSN 2252-6951
e-ISSN 2502-6844

Introduction

Pertuzumab, a monoclonal antibody targeting the Human Epidermal Growth Factor Receptor 2 (HER2), has transformed the treatment landscape of breast cancer (Swain et al., 2023). Its mechanism of action involves inhibiting the heterodimerization process crucial for tumor cell growth by disrupting the interaction between HER2 and other HER family members, such as HER3 and HER4, as well as the Epidermal Growth Factor Receptor (EGFR) (Maadi et al., 2021). Pertuzumab binds to subdomain II of the HER2 extracellular domain, effectively blocking the heterodimerization induced by heregulin (HRG), thus impeding tumor progression (Ishii et al., 2019).

Despite its therapeutic efficacy, the clinical response to pertuzumab varies among individuals, necessitating the exploration of alternative strategies to enhance its effectiveness (Tsao et al., 2022). One such approach is the development of Antibody Drug Conjugates (ADCs), which combine the specificity of monoclonal antibodies with the cytotoxic potency of small molecule drugs (Peters & Brown, 2015). ADCs are composed of an antibody component designed to recognize and bind to specific cell surface antigens, a cytotoxic payload capable of inducing cell death, and a linker molecule that covalently attaches the antibody to the drug payload (Fu et al., 2022). This targeted approach enables the selective delivery of cytotoxic agents to cancer cells while sparing normal tissues, minimizing systemic toxicity, and improving therapeutic outcomes (Drago et al., 2021).

Central to the development of ADCs is the selection of an appropriate linker molecule that can facilitate stable conjugation between the antibody and the cytotoxic drug. Bicyclononyne (BCN) has emerged as a promising candidate for linker development due to its high reactivity and stability in forming covalent bonds with antibodies and drugs (Ponziani et al., 2020). BCN-OH, featuring a hydroxyl group and a bicycloheptane ring, offers stability and strong binding between the antibody and the drug payload. Moreover, BCN-OH possesses favorable drug-like properties, making it an attractive option for ADC development (Fu et al., 2022).

In this context, our study aims to investigate the ability of BCN-OH to conjugate with pertuzumab and elucidate the key residues involved in this conjugation. Through in silico docking simulations, we seek to predict the interactions between pertuzumab and BCN-OH, providing insights into the molecular mechanisms underlying their binding. By understanding these interactions, we aim to assess the potential of BCN-OH as an effective linker in ADCs and contribute to the advancement of targeted cancer therapy.

Understanding the molecular interactions between pertuzumab and BCN-OH is crucial for the development of novel ADCs with improved efficacy and specificity. Our study addresses this gap by leveraging computational approaches to investigate the binding affinity and key residues involved in the conjugation process (Meli et al., 2022). By elucidating the molecular mechanisms underlying the interaction between pertuzumab and BCN-OH, we aim to provide valuable insights that can inform the design and optimization of ADCs for targeted cancer therapy.

Method

Preparation pertuzumab as macromolecule or antibody in ADC

the Research Collaboratory for Structural Bioinformatics (RCSB) Protein Data Bank (PDB) was accessed at https://www.rcsb.org. The PDB ID "1S78" was entered into the search bar to retrieve the specific entry for pertuzumab. Upon locating the entry, detailed information and structural data were reviewed. The molecular structure of pertuzumab was then downloaded in the PDB format directly from the 1S78 entry page. To facilitate further computational analyses, the PDB structure was processed using molecular modeling software to convert it into the appropriate format required for subsequent studies, such as pyrx for docking simulations or other relevant formats. This comprehensive approach ensures the accurate preparation of the pertuzumab structure, suitable for various molecular modeling and docking applications (Mun et al., 2022).

Preparation of BCN-OH as Ligan or Linker in ADC

To prepare a ligand using the PubChem database with the keyword "Bicyclononyne", the following protocol was employed. First, the PubChem database (https://pubchem.ncbi.nlm.nih.gov) was accessed, and "Bicyclononyne" was entered into the search bar to initiate a compound search. From the search results, the appropriate compound was identified and selected for detailed information retrieval. Subsequently, the structure of the compound was downloaded in the desired format, such as SDF, MOL, or SMILES, from the compound's detailed information page. For automation, a Python script utilizing the PubChem PUG REST API was developed (Guidotti et al., 2023). The script executed a search query for "Bicyclononyne" retrieved the corresponding PubChem CID, and downloaded the structural data in the specified format. This methodological approach ensures the efficient preparation of the ligand for subsequent molecular modeling and docking studies (Astalakshmi et al., 2022).

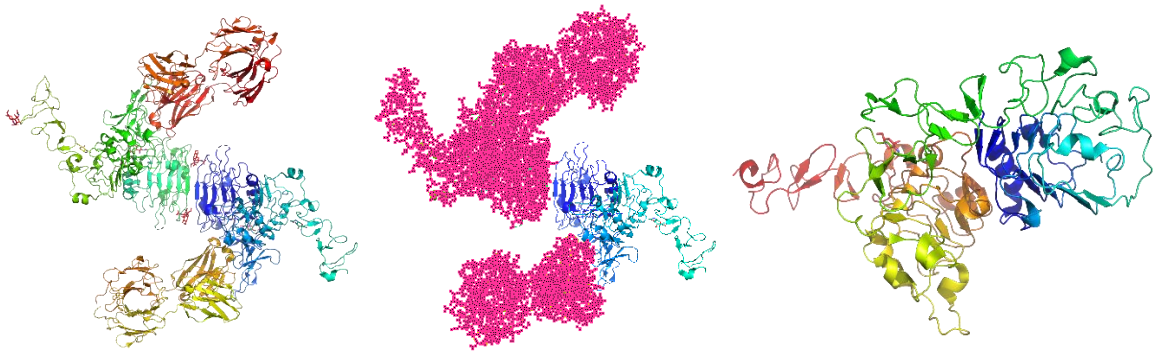
Molecular docking study

For molecular docking studies, the PyRx software, which includes Python, was employed (Vázquez-Jiménez et al., 2022). The preparation of pertuzumab, obtained from the PDB entry 1S78, involved initial cleaning steps where water molecules were removed using PyMOL. Pertuzumab was then prepared as a macromolecule for docking. Concurrently, the ligand Bicyclononyne was prepared using Open Babel to convert its structure to the appropriate format. The docking setup involved configuring the grid box to encompass the desired interaction sites, specifically targeting the lysine and cysteine residues. This setup facilitated the precise targeting of these residues for interaction analysis during the docking simulations.

Results and Discussion

Macromolecule preparation

The macromolecule pertuzumab with PDB ID 1S78 consists of 6 chains and 17 native ligands Fig 1A. In the process of macromolecule preparation for molecular docking, we focused on simplifying the protein structure. We removed all side chains, retaining only a single chain to facilitate molecular docking Fig 1B. Additionally, all native ligands were removed Fig 1C. Notably, no water molecules were present in this PDB file. After the removal of chains and native ligands, we obtained a purified pertuzumab structure in PDB format, which was subsequently used as the macromolecule model in the PyRx software for docking studies.



(A) (B) (C)

Figure 1. Macromolecule preparation, (A) Pertuzumab and native ligand, (B) Proses removing side chains and native ligand, (C) Pertuzumab after removing side chains and native ligand

Ligan preparation

The ligand preparation was conducted to obtain a chemical compound for interaction analysis, specifically visualized as a linker in Antibody-Drug Conjugates (ADC). It is crucial to analyze the drug-likeness properties of the ligand using Lipinski's Rule of Five to ensure its suitability (Qin & Gong, 2022). The ligand BCN-OH demonstrated drug-like characteristics that meet the Rule of Five criteria, Molecular Weight (MW): Less than 500 g/mol, LogP (octanol-water partition coefficient): Less than 5, Hydrogen Bond Donors: No more than 5, Hydrogen Bond Acceptors: No more than 10 (Leeson et al., 2021). The drug-likeness properties of BCN-OH are as follows: molecular weight (MW) = 150.22 g/mol, XLogP3-AA = 1.9, hydrogen bond donor count = 1, and hydrogen bond acceptor count = 1 its mean that BCN-OH has qualified for rule of 5 Table 1. The PubChem platform was used to obtain the ligand, and the structure was downloaded as an SDF file Fig 2. This file was then prepared for molecular docking studies.

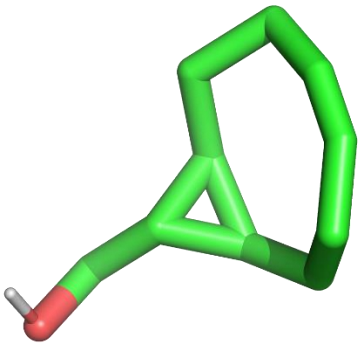


Figure 2. BCN-OH structure in 3D

Table 1. Druglikeness properties of linker BCN-OH

Property Name	Property Value	Reference
Molecular Weight	150.22 g/mol	Computed by PubChem 2.2 (PubChem release 2021.10.14)
XLogP3-AA	1.9	Computed by XLogP3 3.0 (PubChem release 2021.10.14)
Hydrogen Bond Donor Count	1	Computed by Cactvs 3.4.8.18 (PubChem release 2021.10.14)
Hydrogen Bond Acceptor Count	1	Computed by Cactvs 3.4.8.18 (PubChem release 2021.10.14)
Rotatable Bond Count	1	Computed by Cactvs 3.4.8.18 (PubChem release 2021.10.14)
Exact Mass	150.104465066 g/mol	Computed by PubChem 2.2 (PubChem release 2021.10.14)
Monoisotopic Mass	150.104465066 g/mol	Computed by PubChem 2.2 (PubChem release 2021.10.14)
Topological Polar Surface Area	20.2Å²	Computed by Cactvs 3.4.8.18 (PubChem release 2021.10.14)
Heavy Atom Count	11	Computed by PubChem
Formal Charge	0	Computed by PubChem
Complexity	189	Computed by Cactvs 3.4.8.18 (PubChem release 2021.10.14)
Isotope Atom Count	0	Computed by PubChem
Defined Atom Stereocenter Count	2	Computed by PubChem
Undefined Atom Stereocenter Count	0	Computed by PubChem
Defined Bond Stereocenter Count	0	Computed by PubChem
Undefined Bond Stereocenter Count	0	Computed by PubChem
Covalently-Bonded Unit Count	1	Computed by PubChem
Compound Is Canonicalized	yes	Computed by PubChem (release 2021.10.14)

Molecular Docking Analysis

The prepared pertuzumab and BCN-OH ligand were used in molecular docking studies conducted using the PyRx software. PyRx facilitated the molecular docking by integrating the AutoDock Vina tool, allowing for efficient docking simulations (Valdés-Tresanco et al., 2020). The docking setup involved configuring the grid box to encompass the desired interaction sites, specifically targeting the lysine and cysteine residues Fig 3. This setup facilitated the precise targeting of these residues for interaction analysis during the docking simulations. The molecular docking simulations conducted using AutoDock Vina within the PyRx software yielded 9 poses of the BCN-OH ligand bound to pertuzumab, each with varying binding affinities and RMSD (Root Mean Square Deviation) values. Among these poses, we selected the pose with the lowest RMSD value of 0 with highest binding affinity value for visualized binding site.

Choosing the pose with the lowest RMSD (Root Mean Square Deviation) value is a common practice in molecular docking due to several reasons. Firstly, the lowest RMSD typically indicates the most accurate representation of the ligand binding to the target macromolecule, with an RMSD value of 0 suggesting perfect alignment with the predicted binding conformation. This ensures high reliability and consistency, as lower RMSD values indicate less deviation from the expected binding interactions, thereby increasing confidence in the predicted binding site and interactions involved. Additionally, the lowest RMSD not only corresponds to optimal binding affinity but also ensures that the pose is structurally plausible, maintaining crucial interactions like hydrogen bonds and hydrophobic contacts.

Minimizing errors in docking predictions is another key benefit, as high RMSD values could indicate poor alignment and lead to incorrect conclusions about binding interactions and affinity. Lastly, for visualization and further analysis, the pose with the lowest RMSD provides a clear and accurate model of the ligand within the binding site, essential for understanding detailed interaction mechanisms and designing modifications to improve binding affinity or specificity. This pose was chosen for visualization of the binding site on pertuzumab, providing insights into the interaction mechanisms and potential binding affinities of BCN-OH with the target macromolecule. The visualized binding site enabled a detailed examination of the interactions between pertuzumab and BCN-OH, facilitating the understanding of key residues involved in ligand binding and supporting the optimization of ADC linker design.

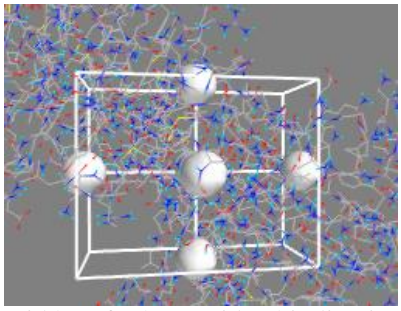


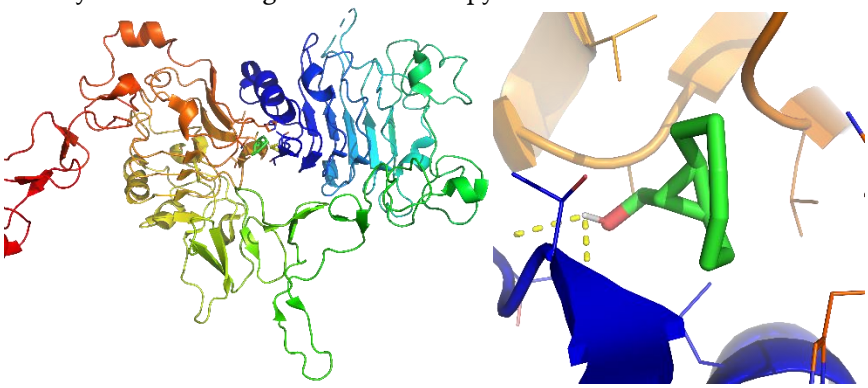
Figure 3. Grid box for key residue binding in Cys or Lys

Table 2. Docking result in 9 different pose

Ligand	Binding Affinity	rmsd/ub	rmsd/lb
pertuzumab_clean_53380994_uff_E=1674.19	-5.1	0	0
pertuzumab_clean_53380994_uff_E=1674.19	-5	9.362	7.46
pertuzumab_clean_53380994_uff_E=1674.19	-4.7	7.82	6.744
pertuzumab_clean_53380994_uff_E=1674.19	-4.7	8.643	7.475
pertuzumab_clean_53380994_uff_E=1674.19	-4.6	4.805	3.328
pertuzumab_clean_53380994_uff_E=1674.19	-4.6	8.461	7.477
pertuzumab_clean_53380994_uff_E=1674.19	-4.6	4.745	2.68
pertuzumab_clean_53380994_uff_E=1674.19	-4.5	5.185	3.594
pertuzumab_clean_53380994_uff_E=1674.19	-4.5	8.809	7.324

Interaction of BCN-OH and Pertuzumab as linker and antibody in ADC

The interaction between BCN-OH and pertuzumab indicates that BCN-OH has the potential to serve as an effective linker in Antibody-Drug Conjugates (ADC). Our molecular docking studies demonstrated that BCN-OH can bind to pertuzumab Fig 4A,B, suggesting its ability to form a stable connection with the antibody. This is a crucial finding as the linker in an ADC plays a vital role in maintaining the stability of the conjugate, ensuring that the drug is delivered specifically to the target cells. The stable binding of BCN-OH to pertuzumab, observed in our docking simulations, supports its suitability as a linker that can facilitate the efficient delivery of therapeutic agents to target cells via pertuzumab. This interaction not only validates BCN-OH as a viable linker but also highlights its potential to enhance the efficacy and specificity of ADCs in targeted cancer therapy.



(A) **(B)**

Figure 4. Interaction mode between BCN-OH and Pertuzumab. (A) Full length Pertuzumab in complex with BCN-OH, (B) Interaction surroundings the BCN-OH

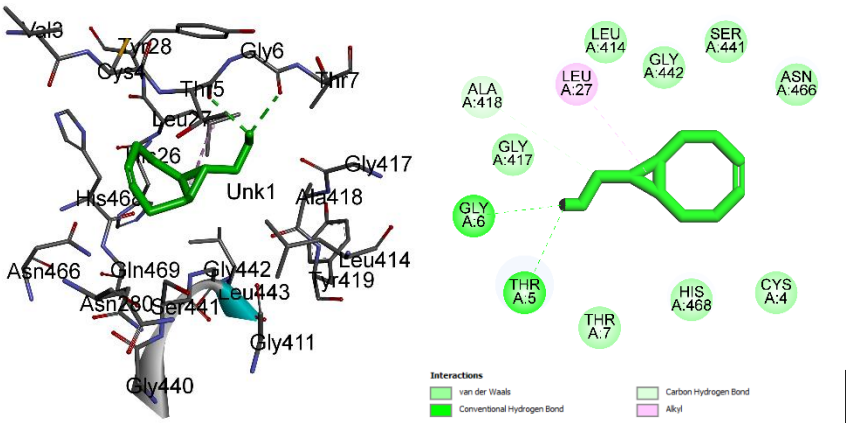
Key Residue involved in interaction

Our analysis of the interaction between BCN-OH and pertuzumab revealed key residues involved in the binding interface, providing insights into the molecular mechanisms underlying their interaction. Notably, the binding affinity of 5.1 kcal/mol signifies a favorable interaction between BCN-OH and pertuzumab, indicating the potential for BCN-OH to serve as an effective linker in Antibody-Drug Conjugates (ADCs).

Several key residues were identified at the binding interface, each contributing to the stability of the BCN-OH-pertuzumab complex through different types of interactions. Thr5 and Gly6 were found to form hydrogen bonds with BCN-OH, enhancing the specificity and stability of the interaction. Additionally, residues Gly417, Leu414, Gly442, Ser441, Asn466, Cys4, His468, and Thr7 were involved in hydrophobic van der Waals interactions, further stabilizing the complex. Furthermore, specific interactions such as the carbon-hydrogen bond with Ala418 and alkyl interactions with Leu27 were observed, contributing to the overall binding affinity. These interactions play crucial roles in maintaining the structural integrity of the BCN-OH-pertuzumab complex and facilitating specific recognition and binding between the linker and the antibody.

This finding underscores the significance of Cys in mediating the interaction between the linker and the antibody. Cys residues are known for their ability to form disulfide bonds, which are critical for stabilizing the structure of proteins and protein complexes. In the context of ADCs, Cys residues play a pivotal role in conjugating the linker to the antibody, thereby facilitating the targeted delivery of therapeutic agents to cancer cells. The interaction between BCN-OH and the Cys residue in pertuzumab suggests the formation of a covalent bond or a strong non-covalent interaction, such as a thiol-hydrogen bond. This interaction is essential for anchoring the linker to the antibody and ensuring the stability of the ADC complex

Overall, the identification of Cys as a key residue in the binding interface between BCN-OH and pertuzumab highlights its crucial role in mediating the interaction and underscores its importance in the design and development of effective ADCs for targeted cancer therapy.



(A) (B)

Figure 5. (A) Key residue of pertuzumab in complex with BCN-OH in 3D, (B) in 2D

Table 3. Interaction profile of BCN-OH and Pertuzumab

Linker/Ligan	ΔG (kcal/mol)	Amino Acid Residue	
		Hydrogen Bond	Hydrophobic
BCN-OH	-5.1	Thr5, Gly6	**Van deer waals**: Gly417, Leu414, Gly442, Ser441, Asn466, Cys4, His468, Thr7 **Carbon hydrogen bond**: Ala418. **Alkyl**: Leu27

Conclusion

In conclusion, our study investigated the interaction between the linker BCN-OH and the antibody pertuzumab, aiming to elucidate the molecular mechanisms underlying their binding in the context of Antibody-Drug Conjugates (ADCs). Our findings demonstrate that BCN-OH has the potential to serve as an effective linker in ADCs, as evidenced by its favorable binding affinity of 5.1 kcal/mol with pertuzumab. Key residues, particularly Cys, were identified at the binding interface, highlighting their crucial role in mediating the interaction between BCN-OH and pertuzumab. The presence of Cys suggests the formation of a covalent bond or a strong non-covalent interaction, essential for anchoring the linker to the antibody and ensuring the stability of the ADC complex. Overall, our study provides valuable insights into the molecular interactions between BCN-OH and pertuzumab, contributing to the understanding of ADC design and optimization. These findings have implications for the development of targeted cancer therapies, where ADCs play a crucial role in delivering therapeutic agents specifically to cancer cells while minimizing off-target effects. Future research in this area may further explore the therapeutic potential of BCN-OH and other linker molecules in ADCs, advancing the field of precision medicine in cancer treatment.

References

Astalakshmi, Dr. N., T, G., K B, G. S., M, N., M R, H. H. S., S, G., Latha, Dr. S. T., & Kumar, Dr. M. S. (2022). Over View on Molecular Docking: A Powerful Approach for Structure Based Drug Discovery. *International Journal of Pharmaceutical Sciences Review and Research*, *77*(2), 146–157. https://doi.org/10.47583/ijpsrr.2022.v77i02.029

Drago, J. Z., Modi, S., & Chandarlapaty, S. (2021). Unlocking the potential of antibody–drug conjugates for cancer therapy. *Physiology & Behavior*, *18*(6), 327–344. https://doi.org/10.1038/s41571-021-00470-8.Unlocking

Fu, Z., Li, S., Han, S., Shi, C., & Zhang, Y. (2022). Antibody drug conjugate: the "biological missile" for targeted cancer therapy. *Signal Transduction and Targeted Therapy*, *7*(1). https://doi.org/10.1038/s41392-022-00947-7

Guidotti, I. L., Neis, A., Martinez, D. P., Seixas, F. K., Machado, K., & Kremer, F. S. (2023). Bambu and its applications in the discovery of active molecules against melanoma. *Journal of Molecular Graphics and Modelling*, *124*(July). https://doi.org/10.1016/j.jmgm.2023.108564

Ishii, K., Morii, N., & Yamashiro, H. (2019). Pertuzumab in the treatment of HER2-positive breast cancer: an evidence-based review of its safety, efficacy, and place in therapy. *Core Evidence*, *Volume 14*, 51–70. https://doi.org/10.2147/ce.s217848

Leeson, P. D., Bento, A. P., Gaulton, A., Hersey, A., Manners, E. J., Radoux, C. J., & Leach, A. R. (2021). Target-Based Evaluation of "drug-Like" Properties and Ligand Efficiencies. *Journal of Medicinal Chemistry*, *64*(11), 7210–7230. https://doi.org/10.1021/acs.jmedchem.1c00416

Maadi, H., Soheilifar, M. H., Choi, W. S., Moshtaghian, A., & Wang, Z. (2021). Trastuzumab mechanism of action; 20 years of research to unravel a dilemma. *Cancers*, *13*(14), 1–17. https://doi.org/10.3390/cancers13143540

Meli, R., Morris, G. M., & Biggin, P. C. (2022). Scoring Functions for Protein-Ligand Binding Affinity Prediction Using Structure-based Deep Learning: A Review. *Frontiers in Bioinformatics*, *2*(June), 1–30. https://doi.org/10.3389/fbinf.2022.885983

Mun, C. S., Hui, L. Y., Sing, L. C., Karunakaran, R., & Ravichandran, V. (2022). Multi-targeted molecular docking, pharmacokinetics, and drug-likeness evaluation of coumarin based compounds targeting proteins involved in development of COVID-19. *Saudi Journal of Biological Sciences*, *29*(12), 103458. https://doi.org/10.1016/j.sjbs.2022.103458

Peters, C., & Brown, S. (2015). Antibody-drug conjugates as novel anti-cancer chemotherapeutics. *Bioscience Reports*, *35*(4). https://doi.org/10.1042/BSR20150089

Ponziani, S., Di Vittorio, G., Pitari, G., Cimini, A. M., Ardini, M., Gentile, R., Iacobelli, S., Sala, G., Capone, E., Flavell, D. J., Ippoliti, R., & Giansanti, F. (2020). Antibody-drug conjugates: The new frontier of chemotherapy. *International Journal of Molecular Sciences*, *21*(15), 1–28. https://doi.org/10.3390/ijms21155510

Qin, Q., & Gong, L. (2022). Current Analytical Strategies for Antibody–Drug Conjugates in Biomatrices. *Molecules*, *27*(19), 1–17. https://doi.org/10.3390/molecules27196299

Swain, S. M., Shastry, M., & Hamilton, E. (2023). Targeting HER2-positive breast cancer: advances and future directions. *Nature Reviews Drug Discovery*, *22*(2), 101–126. https://doi.org/10.1038/s41573-022-00579-0

Tsao, L. C., Crosby, E. J., Trotter, T. N., Wei, J., Wang, T., Yang, X., Summers, A. N., Lei, G., Rabiola, C. A., Chodosh, L. A., Muller, W. J., Lyerly, H. K., & Hartman, Z. C. (2022). Trastuzumab/pertuzumab combination therapy stimulates antitumor responses through complement-dependent cytotoxicity and phagocytosis. *JCI Insight*, *7*(6), 1–18. https://doi.org/10.1172/jci.insight.155636

Valdés-Tresanco, M. S., Valdés-Tresanco, M. E., Valiente, P. A., & Moreno, E. (2020). AMDock: a versatile graphical tool for assisting molecular docking with Autodock Vina and Autodock4. *Biology Direct*, *15*(1), 1–12. https://doi.org/10.1186/s13062-020-00267-2

Vázquez-Jiménez, L. K., Juárez-Saldivar, A., Gómez-Escobedo, R., Delgado-Maldonado, T., Méndez-Álvarez, D., Palos, I., Bandyopadhyay, D., Gaona-Lopez, C., Ortiz-Pérez, E., Nogueda-Torres, B., Ramírez-Moreno, E., & Rivera, G. (2022). Ligand-Based Virtual Screening and Molecular Docking of Benzimidazoles as Potential Inhibitors of Triosephosphate Isomerase Identified New Trypanocidal Agents. *International Journal of Molecular Sciences*, *23*(17). https://doi.org/10.3390/ijms231710047