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In Silico Study of Cladosporol and Its Acyl Derivatives as Anti-Breast Cancer Against Alpha-Estrogen Receptor

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Abstract. Breast cancer is a chronic health problem that causes 690,000 deaths worldwide. The development of secondary metabolite compounds from natural preparations through an *in silico* approach is needed as a predictive tool to prevent breast cancer, one of them is cladosporol from *Cladosporium* spp. This study aims to utilize an *in silico* approach to predict the potential of cladosporol against alpha-estrogen receptors. The alpha-estrogen receptor with code 6CBZ was selected based on group function as pharmacophore in ligand-receptor interaction. The methods used in this study are by using an *in silico* approach with Molegro Virtual Docker (MVD) Ver 5.5 for the docking process and CABS-flex 2.0 for identifying the stability of the complexes. ADMET properties analysis was also performed to know the pharmacokinetics attributes of cladosporol. Based on research conducted, stated that cladosporol octanoate has the lowest rerank score with a -84.3593 value and the RMSD value is 1.195 Å so it's valid for molecular docking. Exploration of cladosporol for anti-breast cancer from *Cladosporium* spp fungi can be a novelty for the development of future pharmaceutical research. Thus, the development of anti-cancer drugs for early prevention can be carried out to reduce the number of breast cancer cases worldwide.

Keywords: Acyl derivatives; ADMET properties; Alpha-estrogen receptor; Anti-breast cancer; Cladosporol

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INTRODUCTION

Indonesia is a developing country where life is faced with various kinds of problems, one of the major problems faced is health problems. The most prominent health problem in the world is chronic deadly cancer where cell growth and development are uncontrolled and very fast (Stefansson *et al*., 2015). According to Ekowati *et al*. (2018), breast cancer is the most common

malignancy in women worldwide, accounting for around 690,000 deaths and 2.3 million new cases each year. Breast cancer is one of the primary causes of cancer-related mortality, according to Harbeck *et al.* (2019).

Hormone-dependent, estrogen receptor-positive breast cancer affects over 80% of female patients with breast cancer globally. Since estrogen receptor alpha is essential to the onset and progression of breast cancer, it is the most important target for treatment (Anandan et al., 2022). Many scientists are working on creating new, powerful therapeutic drugs that specifically bind to estrogen receptor alpha to prevent the onset of hormone-dependent breast cancer (Harbeck et al., 2019). By virtually screening structural models, Shah *et al.*'s research from 2022 effectively found decetene curcumin derivative ligands as antagonists of the estrogen receptor alpha.

Fungi are widely recognized for their ubiquitous nature and are among the most plentiful sources of biodiversity (Greco *et al*., 2019). The genus Cladosporium is thought to be a rich natural resource of diverse natural and bioactive compounds that belong to various classes of secondary metabolites, including flavonoids, naphthalenones, alkaloids, lactones, benzofuoranthines, macrolides, coumarins, isocumarins, perilinaquinones, azafilones, sterols, and others. These compounds have been reported to have biological properties such as antimicrobial, quorum sensing inhibitory, cytotoxic, and phytotoxic activities (Salvatore *et al*., 2021; Ai *et al*., 2015). One of the species of the genus *Cladosporium*. Within 993 genera names, *Cladosporium* species are a diverse and global hyphomycetes genera that are recognized as common endophytes. It is reported that 123 bioactive chemicals are present in *Cladosporium* spp (Salvatore *et al*., 2021). Several bioactive compounds in *Cladosporium* spp. such as cladosporide, brefeldin A, malletinin E, hydroxyemodin, o-Hydroxyphenyl, plumbagin, cladosporin and cladosporol (Akpotu *et al*., 2017; Wang *et al*., 2020; Naseer *et al*., 2017; Salvatore *et al*., 2021; Venkateswarulu *et al*., 2018; Yehia *et al*., 2020; Wang *et al*., 2013).

Two bioactive compounds (Altertoxin X and Cladosporol H) had the highest potential drug-like binding mode, according to research by Anandan (2022) that used docking simulation to explore many bioactive compounds identified in *Cladosporium* spp. against estrogen receptor alpha. Cladosporol has several advantages over Altertoxin X, namely it does not have the possibility to induce mutations in DNA, has a lower LD50 than Altertoxin X, and is not toxic to the liver (Anandan *et al*., 2022). In this paper, molecular dynamics simulations of cladosporol and its derivatives that have been added with alkyl groups against alpha-estrogen receptors were performed with the purpose in order to predict the potential of cladosporol. Genomic analysis, proteomics, bioinformatics, and the effective in silico approach are all part of the modern drug development process (Lisdiana & Mustikaningtyas, 2023). The *in silico* approach can save significant money by preventing late-stage failures and expediting drug discovery (Chikhale, 2020). Critical ADMET factors, including absorption, distribution, metabolism, excretion, and toxicity, can be evaluated by in silico research (Vickers, 2017). Based on the 2D structure of small compounds and the molecules' oral bioavailability, *in silico* studies are very helpful in discovering possible therapeutic candidates (Chang et al., 2023). These models can also be used to align molecules or make molecular docking simulation easier (Hoque *et al*., 2017; Imam & Gilani, 2017; Agu *et al*., 2023). In this study, cladosporol compounds derived from the fungus *Cladosporium* spp. These compounds are known to have antibacterial, antioxidant, and anticancer activities. However, further research based on the basis of molecular interactions and affinity of compounds to each ligand for the body has not been done. Therefore, a study related to molecular docking of cladosporol compounds against alpha-estrogen receptors was conducted. Furthermore, this journal data can then be used as a pharmacological database for the synthesis of medicinal materials from the pharmaceutical field.

METHODS

Docking Materials:

An *in silico* approach was performed in the hardware with specification model Dell Vostro 14 3000 and Intel® AMD Ryzen 5 3500U. The operating system used was Windows 10 Ultimate 64-bit with Radeon Vega MobilrGfx 2.10 GHz. The 2D compound structure of cladosporol was prepared by using Chemdraw Ver. 18 (PerkinElmer Informatics Inc.). Then, the 2D structure was converted into the 3D structure using Chem3D Ver.18 (PerkinElmer Informatics Inc.) to get a visualization of the three-dimensional shape of the molecule (Bajorath, 2011). The receptor also was prepared by using Protein Data Banks

(https://www.rscb.org/) with the code 6CBZ. In order to get the rerank score, the molecular docking of the ligand alpha-estrogen receptor (6CBZ.pdb) was conducted in the sampe protein cavity with Molegro Virtual Docker (MVD) Ver 5.5. Afterward, molecular dynamic simulations on CABS-flex 2.0 were used to validate the docking results (https://biocomp.chem.uw.edu.pl/CABSflex2/index). Last, the SMILE code of the cladosporol compound was inserted into the pkCSM online tool (http://biosig.unimelb.edu.au/pkcsm/) to get ADMET properties (Pires *et al*., 2015).

Receptor Preparation

The three-dimensional structure of the alpha-estrogen receptor was downloaded from Protein Data Banks (https://www.rscb.org/). The selected alpha-estrogen receptor was 6CBZ.pdb. Next, the crystal water molecule has been removed from the binding site. The alpha-estrogen receptor with PDB ID: 6CBZ was selected because it has a group function as a pharmacophore in the ligand-receptor interaction process. Pharmacophore is the precise arrangement of atoms, groups, or functionalities in a small molecule required for specific interactions with its biological target and its activity (Bajorath, 2011). The similarity of the pharmacophore group was high in the tested ligand. Pharmacophore fingerprints attempt to model binding-related structural or chemical properties of chemical compounds with the use of simple statistics of chemical features (Sahdev *et al*., 2023). The detail of the receptor is in Figure 1 (Maximov *et al*., 2018).

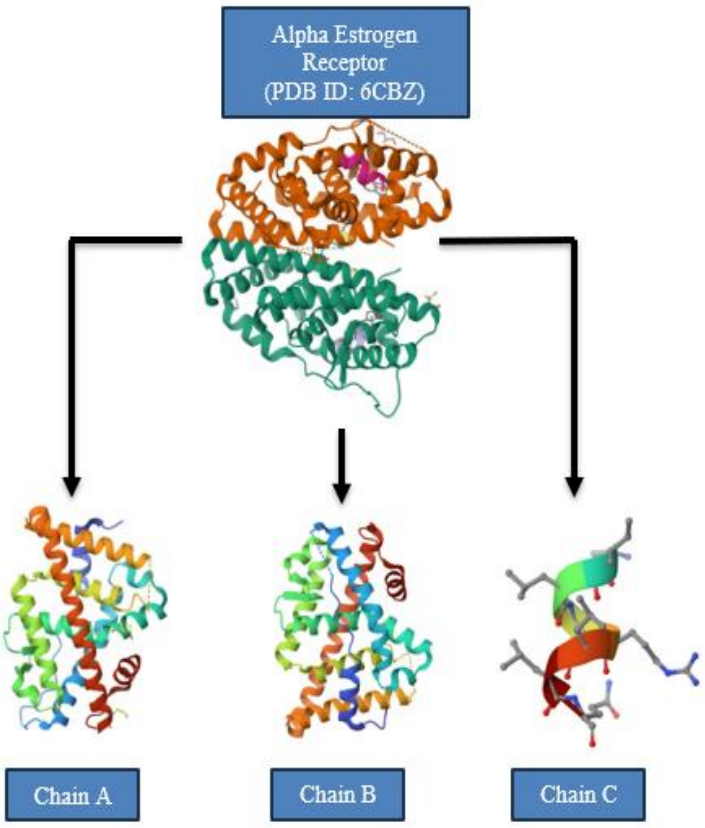


Figure 1. Alpha-Estrogen Receptor (PDB ID = 6CBZ; chain A length = 250; chain B length = 250; chain C length = 8)

Ligand Preparation

Cladosporol derived from *Cladosporium* spp. has a various type such as cladosporol A, cladosporol B, cladosporol C, cladosporol D, cladosporol E, cladosporol F, and cladosporol G (Jones *et al*., 1995). In this paper, we used cladosporol A as the compound model because it has a simple structure for adding the acyl. The 2D structure of the ligands (C and AC1-9) was drawn using ChemDraw Ver. 18. Then, the 2D structure was converted into the 3D structure using Chem3D Ver.18 to get a visualization of the three-dimensional shape of the molecule.

Molecular Docking

Certain details about the protein-inhibitor

complex's binding modalities have been made known by molecular docking investigations (Aloui *et al*., 2024). All of the water molecules were eliminated for molecular docking purposes since they were taken into account during the scoring process (Singh *et al*., 2016). The molecular docking of the ligand alpha-estrogen receptor (6CBZ.pdb) was conducted in the sampe protein cavity with Molegro Virtual Docker (MVD) Ver 5.5. Next, we obtained a Rerank Score (RS) value that can predict the anticancer activity of the tested ligand through the inhibition of the alpha-estrogen receptor. The protein's amino acid residue and the functional group of the ligand have the optimal binding posture when RS has the lowest energy (Agustin *et al*., 2022; Rohmah *et al*., 2024).

Validation of Molecular Docking Method

To determine the stability of molecular complexes, the docking results were then validated using molecular dynamic simulations on CABS-flex 2.0 (https://biocomp.chem.uw.edu.pl/CABSflex2/index). The simulation parameters included protein rigidity, restraints, C-alpha restraints weight, Side-chain restraints weight, number of cycles, trajectory, temperature range, and RNG seed (Shivanika *et al*., 2020; Nandana *et al*., 2023).

ADMET Properties Analysis

Using the pkCSM site, physicochemical and pharmacokinetic predictions of active compounds were performed. The ADMET profile analysis, which encompasses absorption, distribution, metabolism, excretion, and toxicity, was then performed by uploading the SMILE file or copying the SMILE code and selecting the prediction mode. Redocking the native ligand on the cladosporol structure serves as internal confirmation. The purpose of this test is to identify prospective cancer-prevention candidate drugs by analyzing the interaction between ligands and receptor proteins.

RESULTS AND DISCUSSION

The development and identification of cladosporol compound characteristics through an *in silico* approach is important. This is because the molecular dynamics of compounds and their effects on the body can be known based on simulations of organic molecules. Technological advances in the field of computational chemistry have made rapid developments in all fields including pharmaceutical, biomolecular, and many more (Chander *et al*., 2017). The swift advancements in this domain have led to the appearance of numerous novel biological findings and accelerated the process of medication discovery. While CADD (Computer-Aided Drug Design) techniques leverage structural information from drug targets or ligands with known bioactivity to identify prospective drug candidates, biochemical methodologies are used to study the biological function of targets (Yu & MacKerell, 2018; Radityastuti *et al*., 2022). Nowadays, CADD plays a significant role in the drug development process because it may speed up the process by using its understanding of ligand-receptor interactions, structural optimization, and synthesis. Drug development success also largely depends on pharmacological characteristics including toxicity, excretion, metabolism, distribution, and adsorption. Drug discovery and development can be accelerated and improved by combining computational methods with biological data (Surabhi & Singh, 2018; Purnawati *et al*., 2022).

The structural sketch of cladosporol was visualized using Chemdraw and clarified using Chem3D (Figure 2).

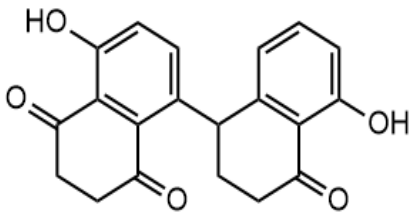


Figure 2. Cladosporol Structure

Validation of the docking method needs to be done before docking the test compound. Docking the native ligand to the chosen receptor served as a method of validation. The RMSD parameter, whose value indicates the amount of variation in the native ligand's position before and after the re-docking procedure, is examined in the docking method's validation. If the RMSD number is less than 2.0 Å, the approach is considered legitimate (Chander *et al*., 2017). The greater the deviation, the greater the error value of predicting ligand interactions with compounds (Brooijmans, 2009). From the results of method validation, the RMSD value is 1.195 Å so the docking method is said to be valid and can be continued molecular docking against the test compound.

Table 1. Test Compound of Molecular Docking

Code	Compound	RS
C	Cladosporol	-70.6869
AC1	Cladosporol acetate	-75.9126
AC2	Cladosporol propionate	-77.5446
AC3	Cladosporol butyrate	-81.8930
AC4	Cladosporol pentanoate	-82.0025
AC5	Cladosporol hexanoate	-82.2885
AC6	Cladosporol heptanoate	-61.3247
AC7	Cladosporol octanoate	-84.3593
AC8	Cladosporol nonanoate	-22.7957
AC9	Cladosporol decanoate	-63.4879

Based on the results of compound preparation, 10 cladosporol compounds and their derivatives were obtained. The prepared cladosporol compounds were analyzed using Molegro Virtual Docker and the Rerank Score (RS) value was obtained. The lower the RS value, the higher the affinity match for the compound and receptor. The compound with code AC7 (Cladosporol octanoate) has the lowest RS value compared to other compounds. This indicates that the cladosporol octanoate derivative compound has a high affinity and is compatible with the 6CBZ alpha-estrogen receptor. Molecular dynamic analysis results indicate that the interaction hotspot's total RMSF value is less than 3 Å (link for MD results https://biocomp.chem.uw.edu.pl/CABSflex2/job/50c57f1635a6d10/) (Figure 2), this indicates that the molecular complex consisting of 6CBZ alpha-estrogen receptor and AC7 interacts through stable bonds. Stable fluctuations formed at the interaction of ligand and protein constituent atoms must have an RMSF value of <3 Å (Wijaya *et al*., 2021).

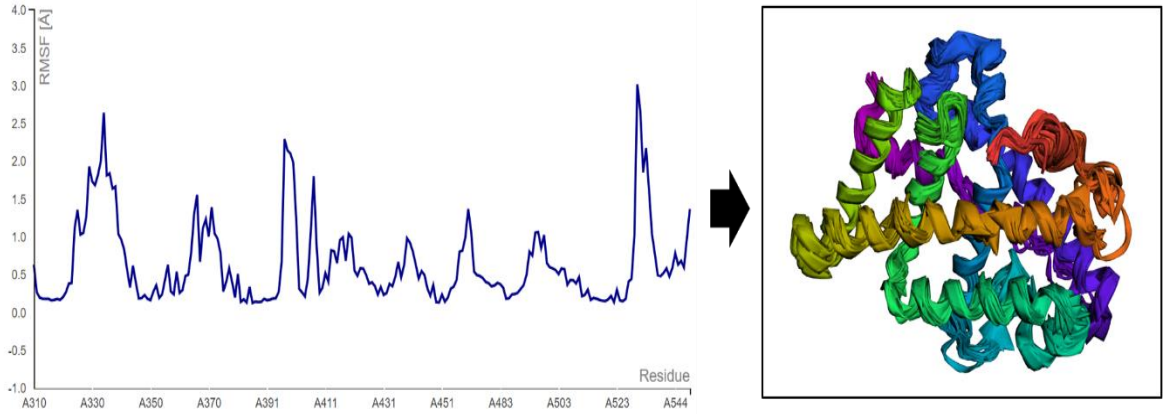


Figure 3. Molecular dynamic of alpha-estrogen receptors_AC7

In addition, a total of 14 ADMET property parameters—which are broken down into groups for toxicity, distribution, metabolism, excretion, and absorption—were detected (table 2). Drugs enter the human body through four different pathways: absorption, distribution, metabolism (biotransformation), and elimination. We refer to this process as ADME regulation. Similar to ADMET properties analysis, the pharmacokinetic approach is used to assess if a medication can have the required pharmacological effects (Zhong, 2017).

Table 2. ADMET Properties

ADMET Properties	Code Receptor 6CBZ									
	C	**AC1**	**AC2**	**AC3**	**AC4**	**AC5**	**AC6**	**AC7**	**AC8**	**AC9**
Water Solubility (Log mol/L)	-4.157	-4.66	-5.005	-5.342	-5.662	-5.957	-6.214	-6.42	-6.56	-6.645
CaCO2 Permeability (Log Papp in 10'-6 cm/s)	0.512	1.234	1.217	1.188	1.168	1.157	1.141	1.12	1.098	1.076
Intestinal Absorption (%Absorption)	92.879	95.014	95.104	94.729	93.896	93.301	92.944	92.594	92.25	91.906
VDss (Log L/Kg)	0.056	0.252	0.321	0.377	0.416	0.448	0.471	0.479	0.468	0.495
BBB Permeability (Log BBB)	-0.338	-0.244	-0.269	-0.293	-0.315	-0.332	-0.352	-0.375	-0.39	-0.419
CYP2D6 Substrate	No	No	No	No	No	No	No	No	No	No
CYP3A4 Substrate	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Inhibitor CYP1A2	Yes	No	No	No	No	No	No	No	No	No
Inhibitor CYP2C19	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Renal OCT2 Substrate	No	No	No	No	No	No	No	No	No	No
Total Clearance	0.079	0.182	0.876	0.884	0.914	0.941	0.471	0.483	0.495	0.507
AMES *Toxicity*	No	No	No	No	No	No	No	No	No	No
Max. Dosage Tolerance (*Human*) (Log mg/Kg/Day)	-0.224	-0.24	-0.15	-0.114	-0.082	-0.052	-0.023	0.005	0.034	0.065
hERG I Inhibitor	No	No	No	No	No	No	No	No	No	No
hERG II Inhibitor	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

The absorption group consists of water solubility (Log mol/L), $CaCO_2$ permeability (Log Papp in 10^{-6} cm/s), and intestinal absorption (%Absorption) parameters. The water solubility of cladosporol derivatives tended to be lower than the parent compound (Table 2). The longer the carbon chain of the derivative compound, the lower the solubility. This is because the carbon chain is classified as a non-polar bond that makes the solubility of the bond between water and the compound decrease (Solomon *et al*., 2011). Then for $CaCO_2$ permeability, a compound has good $CaCO_2$ permeability if the log Papp value is >0.90 x 10^{-6} cm/s (Pires *et al*., 2015). Table 2 shows that the parent compound cladosporol has a log Papp value below 0.90 which means the compound does not have good permeability. Meanwhile, other cladosporol-derived compounds have log Papp values above 0.90 which indicates that the compound has a high ability to penetrate the $CaCO_2$ cell membrane. Furthermore, in the intestinal absorption parameter, a compound is

said to be difficult to absorb by the receptor if the percentage value is <30% and easy to absorb if the value is >80% (Chander *et al*., 2017). Based on the data obtained, it can be interpreted that all compounds have a percentage proportion above 90%, which means that the compounds are able to be absorbed easily in the small intestine. The derivative compound coded AC1 with replication of the lowest alkyl group addition (-COOH) has the best intestinal absorption compared to other compounds and even the parent compound itself. Meanwhile, the derivative compound with the longest alkyl group addition replication with the code AC9 tends to have the lowest intestinal absorption when compared to other compounds.

In the distribution group, there are several parameters used including VDss (Log L/Kg) and BBB Permeability. The distribution group is one of the critical parameter groups for pharmacokinetic analysis, especially in the potential toxicity and systemic effects of drug ingredients. The likelihood that the pharmacological ingredient will be transported to tissue as opposed to the plasma membrane increases with increasing VDss value in the VDss parameter. The purpose of adding alkyl groups to the cladosporol chemical is to raise its VDss value, which will enable it to be dispersed throughout breast tissue to the greatest extent possible and enhance its anticancer properties. According to Pires et al. (2015), a compound is considered to have a low VDss value if the log VD value is less than -0.15 and a high VDss value if the log VD value is greater than 0.45 L/Kg. Based on Table 2, shows that the parent compound (code C) and derived compounds (code AC1-AC5) do not meet the criteria for high distribution (medium category) with values in the interval -0.15 to 0.45. Therefore, these compounds are predicted to be difficult to distribute into breast tissue. This contrasts with the derivative compound (code AC6-AC9) which has a VDss value above 0.45. Derived compounds with these codes have great potential to be maximally distributed in breast tissue. In addition, to improve the drug delivery system, several other delivery systems can be used such as liposomes, polymers, nanoparticles, and micelles (Liyanage *et al*., 2019). Furthermore, for BBB (Blood Brain Barrier) permeability parameters, compounds that have a logBB value > 0.3 have a high potential to be distributed in the blood-brain barrier. Meanwhile, compounds with a logBB value < -1 are difficult to distribute in the blood-brain barrier. Based on the data, all compounds, both the parent compound cladosporol and its derivatives, have a logBB value < -0.1, which means that all compounds are difficult to distribute in the blood brain barrier.

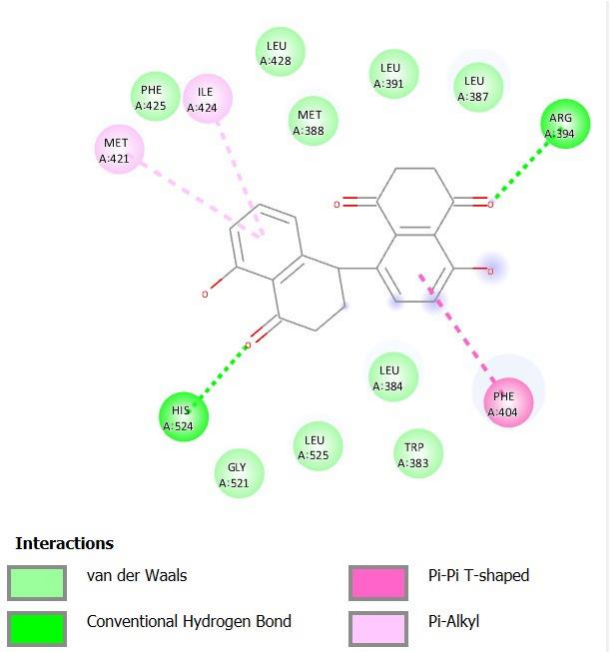
In the metabolism group, CYP2D6 substrate, CYP3A4 substrate, CYP1A2 inhibitor, and CYP2C19 inhibitor were among the parameters that were noted. CYP inhibitors are crucial for understanding how drugs interact with and function in the body (Lonsdale *et al*., 2013). Based on the data in Table 2, shows that all test compounds were unable to bind to the CYP2D6 substrate. However, all compounds can bind to the CYP3A4 substrate. Then based on the inhibitory ability in the form of CYP1A2 inhibitors, all derivative compounds have no inhibitory ability, except for the parent compound cladosporol which has inhibitory ability. The inhibitory ability in the form of CYP2C19 inhibitors of all compounds is also shown in CYP2C19 which is a cytochrome P-450 group for the metabolism of several drug compounds (Kirchmair *et al*., 2015).

In the excretion group, predictions were made regarding 2 parameters, namely renal substrate OCT2 and total clearance. OCT2 (Organic Cation Transporter-2) renal substrate is a transporter that plays a vital role in renal disposition. The excretion process is the removal of chemicals from the body and clearance (CI) is a parameter to measure the elimination of drugs from the body (Varma *et* al., 2015). Based on the data in Table 2, it shows that all compounds, both the parent compound cladosporol and its derivatives, do not have a match with the OCT2 substrate and it is predicted that there is no side interaction with compounds that include OCT2 inhibitors. The total clearance was obtained from a combination of hepatic and renal clearance values (Abdel-Illah *et al*., 2017). All derivative compounds showed higher total clearance values than their parent compounds. This indicates that cladosporol derivatives are predicted to be easily excreted from the body compared to the parent compound cladosporol itself.

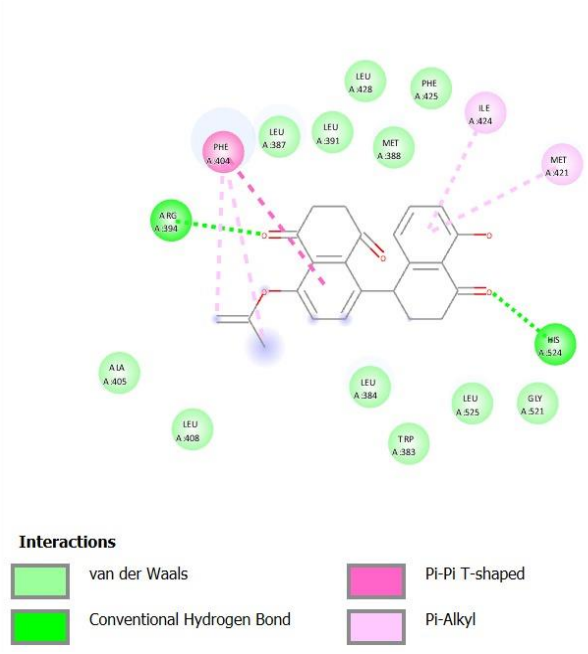
In the last group, the toxicity group, predictions were made regarding AMES toxicity, max dose tolerance, hERG I inhibitors, and hERG II inhibitors. hERG is a potassium channel whose blockage is considered to cause sudden death due to drug-induced QT syndrome. Drug inhibiting hERG thus would pose potential toxicity in high demand (Wang *et al*., 2012) All test compounds did not have AMES toxicity to the receptor. The presence of AMES toxicity is intended as a parameter for anticancer prediction for compounds. Furthermore, for the max dose

tolerance parameter of the compound for the body shows an increase in each replication of alkyl addition to the cladosporol compound. The longer the alkyl group is added to the compound, the greater the maximum dose tolerance of the compound for the body (Pires *et al*., 2015). Then, the hERG I inhibitor parameter showed that all compounds did not have the ability to inhibit HERG I. Meanwhile, for the hERG II inhibitor parameter, only the parent compound cladosporol does not have inhibitory activity against hERG II. All cladosporol-derived compounds have inhibitory activity against hERG II.

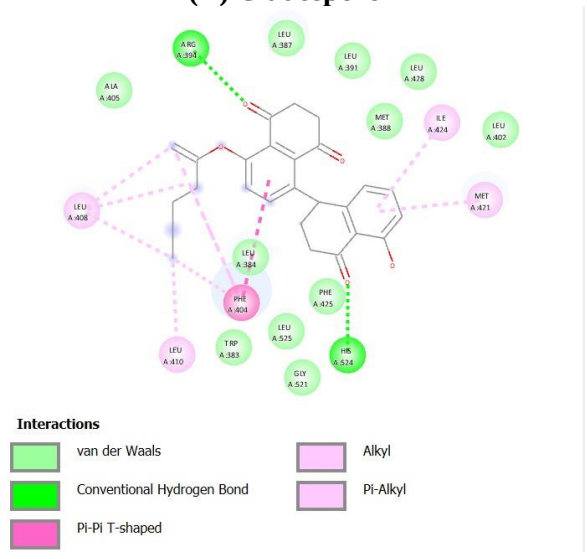
From the overall results and discussion described, all cladosporol test compounds and their derivatives have the potential to develop into anti-breast cancer. This is evidenced by ADMET and molecular docking data that show a match between the ligand and the alpha-estrogen receptor. The bioactivity of functional inhibition of breast cancer cells with alpha-estrogen receptors is also shown with all test samples having a rerank score value below 2.0 Å which means that all of them have good activity as anti-breast cancer (Chander *et al*., 2017). Further analysis using visualization of the binding between alpha estrogen receptor and cladosporol was performed to further investigate the receptor-compound match (figure 4).



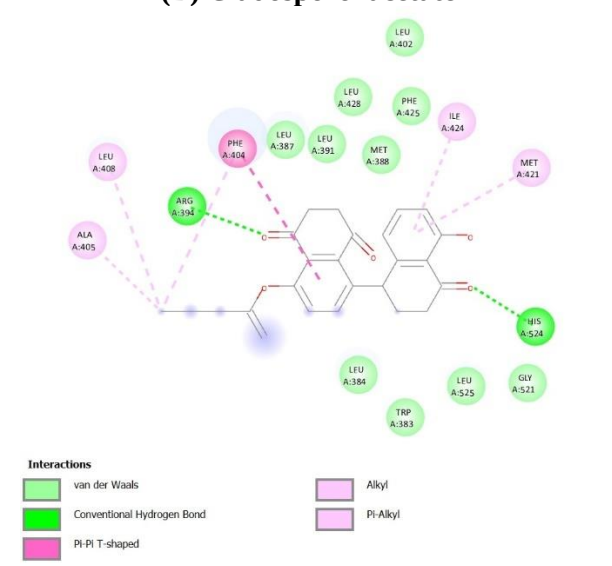
(A) Cladosporol



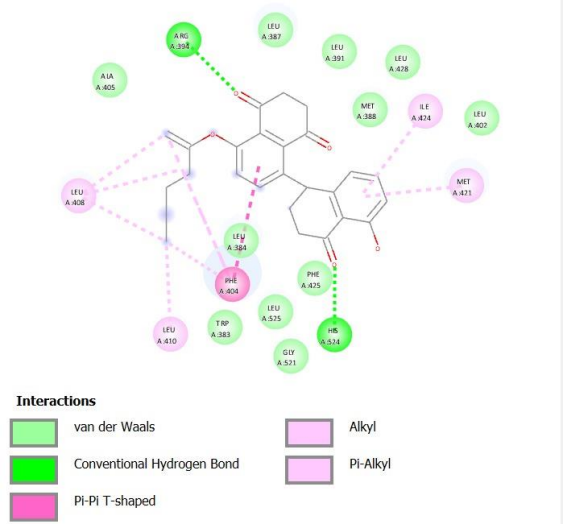
(B) Cladosporol acetate



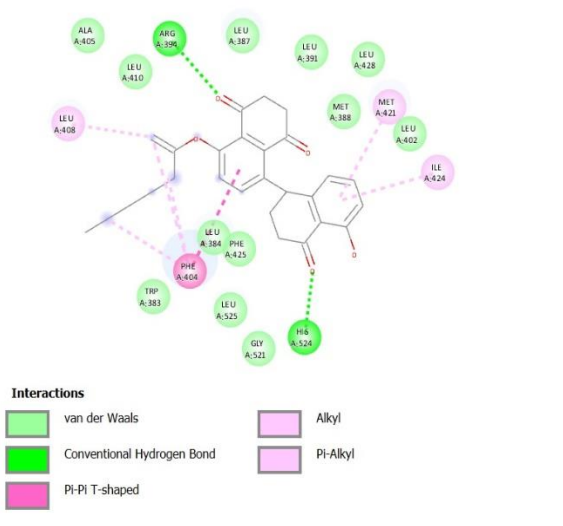
(C) Cladosporol propionate



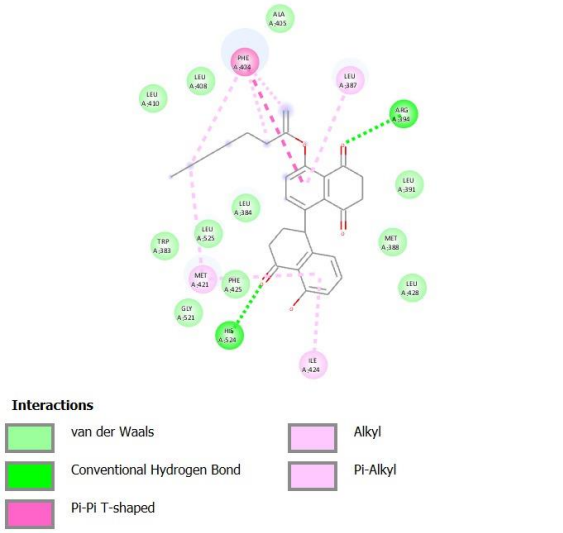
(D) Cladosporol butyrate



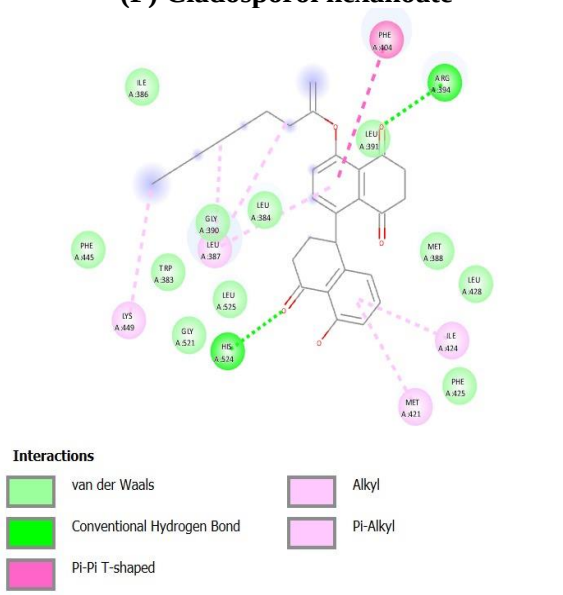
(E) Cladosporol pentanoate



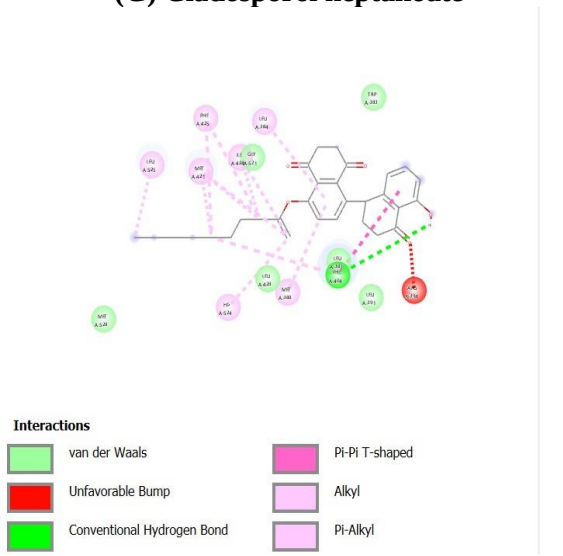
(F) Cladosporol hexanoate



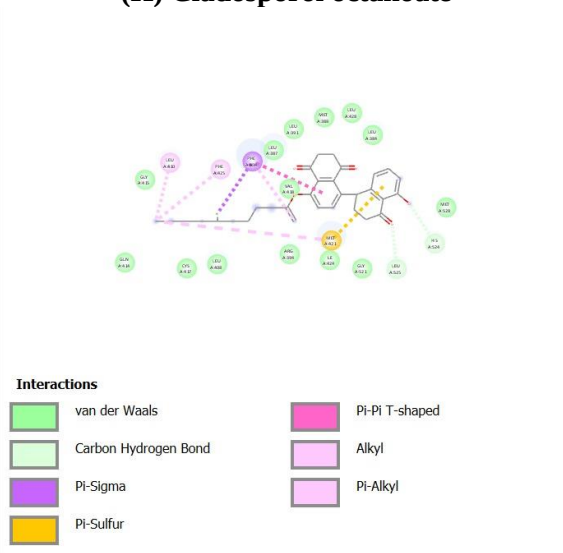
(G) Cladosporol heptanoate



(H) Cladosporol octanoate



(I) Cladosporol nonanoate



(J) Cladosporol decanoate

Figure 4. The binding visualization of cladosporol in targeting alpha-estrogen receptor

Figure 4 visualization aims to give a more thorough explanation of the docking outcome (Antonius *et al*., 2017). Cladosporol octanoate (AC7) showed the compounds with the greatest negative affinity. The complex as a whole has van der Wall interactions and conventional hydrogen bonds, which enable the molecule to attach to receptors strongly. According to Padmi *et al*. (2022), Cladosporol is a good indicator of van der Walls interactions, pi-alkyl, pi-pi T-shaped, and hydrogen bonding. Molecular docking research indicates that cladosporol in catalytic residues helps stabilize ligand complex contacts with targeted proteins, resulting in the right contact shape (Yang *et al*., 2016). Thus, additional in vitro and in vivo studies are needed to ascertain the importance of these connections (Abdullahi et al., 2021). A comparatively negative binding affinity is also formed by various interactions, such as those between pi-sulfur, pi-sigma, pi-alkyl, pi-pi T-shaped, and unfavorable bump bonds (Kan *et al*., 2018; Aini *et al*., 2022; Aini *et al*., 2022).

Researchers are still searching for anti-breast cancer compound candidates. Many *in vivo* and *in vitro* tests have been conducted using experimental animals. To determine the most appropriate and effective therapeutic ingredients to inhibit breast cancer cells, however, in silico predictive testing in the form of docking experiments must be conducted. In addition, the exploration of candidate anti-breast cancer compounds using cladosporol compounds from *Cladosporium* spp fungi can be a novelty for the development of future pharmaceutical research (Dibha *et al*., 2022).

In the future, the exploration of secondary metabolite compounds from *Cladosporium* spp. could be a strategy for the development of herbal ingredients from natural materials. In addition, cladosporol compounds that have known potential as anti-cancer drug candidates can be further tested using *in silico* and *in vivo* approaches in animal tests (Abdullahi *et al*., 2021). This is done to prove the effectiveness of the compound in inhibiting breast cancer cell growth through the alpha-estrogen receptor pathway. Thus, the development of anti-cancer drugs for early prevention can be carried out in order to reduce the number of breast cancer cases worldwide.

CONCLUSION

Recent studies of this *in silico* study show that cladosporol derivative compounds from *Cladosporium* spp. fungi with the code AC7 (Cladosporol octanoate) have the lowest rerank score which indicates the compound has high affinity and matches the 6CBZ alpha-estrogen receptor. Cladosporol octanoate has been found by MD research to have more stability, more non-bonded interaction capability, and lower binding energy. The outcome showed that the synthesis of cladosporol octanoate had great potential for use in the creation of organic synthesis anticancer medicines. Although cladosporol appears to be a promising treatment for alpha-estrogen receptor-related breast cancer, more in-vitro and in-vivo trials in a wet lab are required. The authors suggest that in future research, further exploration should be carried out regarding the potential of cladosporol for other types of cancer receptors in order to enrich the database of the bioinformatics approach.

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