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Screening and Docking Molecular Studies of Natural Products Targeting overexpressed Receptors HER-2 in Breast Cancer

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ABSTRACT

Breast cancer is the first cancer to affect a community. Because of its extremely high mitotic activity, breast cancer that tests positive for HER 2 is considered to have a poor prognosis. Due to the side effects of chemical drugs, patients are increasingly turning to natural medicine, such as phytotherapy and nutritherapy. The study uses a bioinformatics approach (molecular docking) to searchfor new, non-toxic anti-cancer inhibitors. The studyscreens 102 ligands from natural and dietary compounds that are likely to interact with the HER-2. The virtual screening results of the allow us to select the 23 best compounds which can be proposed as the most effective HER-2 inhibitors. Lycopene would be a very promising ligand which presents a DeltaG of -9.82 kcal/mol. Other promising ligands include beta-carotene (DeltaG of -8.58), P-cumaric acid kcal/mol (DeltaG of -8.57) and Curcumin (DeltaG of -8.46). Other compounds, luteolin, anacardium (Anacardic acid) ,and alpha-Tocopherol, were found to have the strongest inhibitory effects with DeltaG values of -7.92 kcal/mol, -7.89 kcal/mol, and-7.85 kcal/mol, respectively. These compounds act directly on residues keys found in the hydrophobic pocket II (ATP binding site) and the hydrophobic region (the αC-β4 loop) of the EGFR domain. Pinoresinol, Kaempferol and Caffeic acid have DeltaGs of -7.48 Kcal/mol, -6.88 Kcal/mol and -6.34 kcal/mol, respectively. These three ligands are specific to the conserved regions of the HER-2 receptor and interact with the C-terminal, the C-lobe activation loop and the N-lobe P loop of the tyrosine kinase domain, respectively. Lapatinib (chemical compound) and quercetin (natural compound) have DeltaG of -7.58 kcal/mol and -7.28 kcal/mol, respectively, form a hydrogen bond with the same residue in the hydrophobic region. All the natural molecules seem very promising and, after *in vitro/in vivo* tests, could constitute good substitutes for the chemotherapies which are currently used to treat breast cancers as well as other cancers.

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1. Introduction

Breast cancer is the second most common malignancy and the second leading cause of cancer-related deaths in women (1). In early 2020, the World Health Organization (WHO) announced the incidence of breast cancer is rising in developing nations due to rising life expectancies, increased urbanization, and the adoption of Western lifestyles. It is estimated that 627,000 women died of breast cancer in 2020, accounting for 15% of all female cancer deaths. Estrogen and progesterone hormone receptor dysfunction is typically associated with breast tumors (2, 3). Furthermore, a great deal of research has been conducted on the overexpression of the human epidermal growth factor receptor 2 (HER2), overexpression of the epidermal growth factor receptor (EGFR1), and PI3Ka (dysregulation of the ER+ and ER−) signaling pathways in breast cancer (3, 4).

Therefore, it is essential to discover novel techniques and compounds that target these proteins. Approximately 20% to 25% of breast cancers are caused by the transmembrane protein receptor known as human epidermal growth factor receptor 2 (HER2), which is encoded by the HER2 gene located on the long arm of chromosome 17. The EGFR family consists of the four HER receptors: HER4, HER3, HER2, and HER (5). Upon HER2 receptor activation, specific tyrosine kinase residues are phosphorylated and signaling proteins are activated upon HER2 receptor activation, leading to the start of downstream signaling processes.The HER2 receptor regulates apoptosis, angiogenesis, cell proliferation, and survival through critical pathways that include mitogen-activated protein kinase (MAPK) and phosphatidylinositol triphosphate kinase (PI3K) signaling mechanisms (6).

In HER2+ breast cancers, HER2 receptor overexpression is known to be a HER2 activation mechanism. HER2-positive breast cancer remains a case study to this day. It is considered a cancer with a poor prognosis due to its high mitotic activity and ability to metastasize easily. However, improved molecular genetic techniques have made it possible to study resistance to trastuzumab (HERCEPTIN) treatment and develop new anti-HER2 targeted therapies. The monoclonal antibody pertuzumab and the tyrosine kinase inhibitor lapatinib specifically target HER2 receptors. The adverse effects of these two chemical and synthetic drugs include alopecia, nausea, vomiting, fatigue, fever, infection, diarrhea, muscle pain, paresthesia, cognitive disorders, cardiotoxicity, leukemia, and gastrointestinal and dermatological reactions. Several other drugs, such as tamoxifen, raloxifene, toremifene, and fulvestrant, are availablefor the treatment of breast cancer, but each has limitations that cause irreversible side effects (7). Researchers are searching for other, less toxic, and more natural molecules. Patients are therefore increasingly turning to natural medicine, such as phytotherapy and nutritherapy. There are various benefits to using natural products in food and medicine development, such as their superior chemical diversity, biological potency, structural complexity, and optimized regulation of natural product biosynthesis. Therefore, the goal of this study was to discovera more selective natural compound that targets breast cancer and can be used as a therapeutic agent using *in silico* methods.

2. Materials and Methods

2.1. Preparation of the protein

The crystal structure of the kinase domain of human HER2 was obtained from the Protein Data Bank (PDB) (https://www.rcsb.org) (8), with PDB ID: 3PP0 (9). The structure was downloaded in PDB format and further prepared for the docking process.

2.2. Preparation of ligands

Following an extensive literature search, 102 molecules with the potential to interact positively with the ErbB2 receptor tyrosine kinase domain in HER2+ breast cancer were selected. These molecules are derived from plants, microorganisms or food sources.

The ligand codes were obtained from PubChem (https://pubchem.ncbi.nlm.nih.gov/) (10), and Zinc Database (https://zinc.docking.org/) (11).

2.3. Pharmaco-Toxicity Study of Ligands

To test the toxicity of the plant-derived molecules, we used the PKCSM Database online server (http://structure.bioc.cam.ac.uk/pkcsm) (12).

We copied the ligand codes obtained from PubChem and Zinc Database to the pkCSM Database to eliminate toxic ligands based on the following criteria : AMES toxicity, hERG K^+ channel inhibitors toxicity and Hepatotoxicity.

2.4. Molecular Docking

Molecular docking was performed using the SwissDock server (http://www.swissdock.ch/) (13).

This server allows importing the target molecule,the "tyrosine kinase domain of the HER2 receptor",as well as

the ligands, for the purpose of testing their interactions. The goal is to study the interactions of these two molecules. Investigating the outcomes enables us to identify the binding energyand the hydrogen bonds formed, as well as the amino acids involved in these interactions.

2.5. Docking results visualisation

The visualization of the molecular docking results from the SwissDock server is done using the UCSF Chimera software (https://www.cgl.ucsf.edu/chimera/) (14).

3. Results

3.1. Ligand toxicity analysis

Initially, 102 natural compounds were obtained from the databases. These molecules can be found in food sources, such as plants or microorganisms. Some of these compounds were screened for their toxicity based on the predicted mutagenicity screening (AMES Toxicity), HERG K+ channel inhibitor toxicity ,and hepatotoxicity results. The results revealed eight potentially toxic ligands (Table 1). This toxicity could be mitigated by decreasing the administrated dose by 0.558 log mg/kg/day for Genipin, 0.36 log mg/kg/day for Sauchinone, 0.654 log mg/kg/day for Denbinobin, 0.144 log mg /kg/day for Xenognosin, and 0.82 log mg/kg/day for Kaempferol.

3.2. Interaction of ligands with HER2

This study investigated the interaction of the 3PP0 protein against 84 ligands. The results are shown in Table 2.

3.3. Ligands Interacting With Conserved Residues of the Tyrosine Kinase Domain of EGFR Family Receptors

It notes that there are 47 complexes formed between the ligands and the tyrosine kinase domain of 3PP0, which have the lowest energy scores compared to the other ligands. These complexesform hydrogen bonds with essential residues that are conserved in the EGFR family (Table 2). Therefore, according to interaction energy,the best ligand is Lycopene Table 2. Lycopene from tomato has an interaction energy of -9.82 kcal/mol and no predicted hydrogen bonds, suggesting the existence of other types of bonds by SwissDock (Figure 1). Taking into account what has been cited in the literature, including Met801, which is located in the Adenine region of the ATP binding site, and Cys805, which is located in of hydrophobic pocket II, it has noted that some ligands form interactions with the ATP binding site (Table 2) (15). Compounds that establish a hydrogen bond with the residue Met 801 in the adenine region are as follow:

- **Alpha-Tocopherol**

From sunflower oil has an interaction energy of –7.85 kcal/mol.

- **Isofraxidinde**

From the species *Eleutherococcus senticosus*,also known as Siberian ginseng, has an interaction energy of –7.77 kcal/mol**.**

- **Pyocyanin**

A blue green phenazine moleculeproduced specifically by the bacterium *Pseudomonas aeruginosa*, has an interaction energy of -7.65 kcal/mol.

- **Sulforaphane**

Which is mainly found in broccoli and cabbage, has an interaction energy of -7.10 kcal/mol.

- **Phloretic acid**

Which belongs to a class of organic compounds found in peanuts and avocados, has an interaction energy of –7.06 kcal/mol.

- **Xenognosin**

Which is present in common peas *Pisum sativumet* legumes, belongs to a class of organic compounds and hasan interaction energy of –6.86 kcal/mol.

- **Urocanic acid**

Which is essentially found in the fungus *Hippospongia communis*, has an interaction energy of –6.49 kcal/mol.

- **Sesamol**

Which is found in sesame seed, has an interaction energy of –6.09 kcal/mol.

Other compunds forming hydrogen bonds with cysteine residue 805 in the hydrophobic pocket II are: Syringic acid**,** found in from olive oil, and Anacardic acid, a component of cashew nuts, have interaction energies (DeltaG) of -6.52 and -7.89 kcal/mol, respectively.

These ligands interact with and form hydrogen bonds with residues Met801 and Cys805 in the ATP binding site of the aforementioned regions. They may have the potential to act as competitive inhibitors by blocking ATP access to its specific site on the tyrosine kinase domain (16, 17). In this study, Anacardic acid is the ligand with the best interaction energy value with the ATP binding site, with a DG of -7.89 kcal/mo (Figure 2).

Two of the 21 ligands, both of which are olive oil, act on the N-lobe of the tyrosine kinase domain.They were selected according to their interactions with residues in the (Cα) helix span residues (729-744).

Table 1. Toxicity parameters of some compounds.

Molecules	Mutagenicity	K+ hERG 1	Na + hERG 2	Hepatotoxicity
Genipin	Yes	No	No	No
Sauchinone	Yes	No	No	No
Denbinobin	Yes	No	No	No
Furanodiene	No	No	No	No
Chalcones	No	No	No	No
Isoliquiritoside	No	No	No	No
Xenognosin	Yes	No	No	No
Kaempferol	Yes	No	No	No
Luteolin	Yes	No	No	No
Silibinin	Yes	No	Yes	No
Daidzeine	Yes	No	Yes	No

Table 2. Docking results of total ligands with the tyrosine kinase domain obtained by Swiss dock.

Ligands	Reesidue (s) target(s)	Length of hydrogen bong (A°)	Interraction energy (kcal/mol)
Crocetin	ALA 706 ALA 706	3.011 3.324	-9.46
Secoisolariciresinol diglucoside	GLU 757	1.841	-8.41
Lycopene	----------	No hydrogen bond	-9.82
P-coumaric acid	----------	No hydrogen bond	-8,57
Curcumin	----------	No hydrogen bond	-8.46
Pomiferin	----------	No hydrogen bond	-8.08
Formononetin	----------	No hydrogen bond	-8.07
Rosmarinc acid	----------	No hydrogen bond	-7.83
3,4-Dihydroxybenzoic	GLY 737	1.915	-6.22
4 P-hydroxybenzoique 1 acid	SER 779	2.213	-6.88
4 P-hydroxybenzoique 2 acid	----------	No hydrogen bond	-6.21
Gallic acid	VAL 777	2.517	-6.25
Gentisic acid	----------	No hydrogen bond	-6.93
Syringic acid	CYS 805	3.096	-6.51
Vanillic acid	VAL 777	2.191	-6.20
Catechines	VAL 777	3.187	-6.82
Epicatechines	GLN 943	1.819	-6.74
Biochanine A	LEU 726	3.347	-6.67
Ergostane	----------	No hydrogen bond	-6.18
Glycitein	----------	No hydrogen bond	-6.37
Daidzeine	VAL 777	2.114	-6.61
Genistein	CYS 947	1.995	-7.54
Malvidin	VAL 777	3.031	-7.32
Delphinidine	----------	No hydrogen bond	-6.70
Cyanidin	GLN 709 GLN 709	2.003 1.929	-7.00
Acetoxypinoresinol	----------	No hydrogen bond	-7.29
Pinoresinol	SER 728	2.523	-7.80
Hydroxytyrosol	ARG 849	2.215	-6.91
Tyrosol	ARG 849	2.129	-6.36
Secoisolariciresinol	----------	No hydrogen bond	-7.49
Enterodiol	PRO 945	2.169	-6.63
Enterolactone	----------	No hydrogen bond	-7.51
Capsaicin	----------	No hydrogen bond	-7.62
CAPE **Caffeic-acid-phenethyl ester**	----------	No hydrogen bond	-6.46
α-Linolenic acid	SER 728	3.160	-7,69
ChlorogeniqueHeriguard acid	GLN 943	2.140	-6.96
Ferulic acid	----------	No hydrogen bond	-7,37

Gingerol	----------	**No hydrogen bond**	**-7,27**
Petunidin	----------	No hydrogen bond	-7,03
Pelagronidine	GLY 778	2,052	-6,38
Homocastasterone	ASP838	2.716	-7.44
Cafeic	LEU1000	2.044	-6.71
Sinapic	CYS 805	2.052	-7.40
3-Hydroxybenzoic	---------	No hydrogen bond	-6.68
O-coumaric acid	----------	No hydrogen bond	-6.05
Diindolylmethane	----------	No hydrogen bond	-7,31
Naringenine	GLN943	1.841	-6.65
Indol 3-carbinol	----------	No hydrogen bond	-6.13
Kaempferol	ASP 863	3.572	-6.88
Dihydroresveratrol	---------	No hydrogen bond	-7.15
Resveratrol	----------	No hydrogen bond	-6.51
Sulforaphane	MET 801	3.708	-7.10
Myricetin	---------	No hydrogen bond	-7.03
Quercetin	ALA706 VAL 777	2.130 3.491	-7.28
Apigenin	CYS 947	2.064	-7.55
Luteolin	ALA 706 VAL 777	2.132 2.247	-7.92
Fisetin	VAL777	3.336	-6.55
Sauchinone	----------	No hydrogen bond	-6.83
Denbinobin	----------	No hydrogen bond	-7.01
Furanodiene	----------	No hydrogen bond	-6.82
Chalcone	----------	No hydrogen bond	-7.19
Lupane	----------	No hydrogen bond	-6.77
Genipin	VAL 777 VAL 777	2.696 3.142	-6.50
Opium	VAL777	2.054	-6.70
Pyocyanin	MET801	2.162	-7.65
Ginsenol	ALA706	2.131	-6.34
Menthol	VAL777	2.329	-6.10
Urocanic acid	MET801	2.182	-6.49
Anthranilic acid	ALA706 ALA706	2.410 2.177	-7.54
Anacardic acid	CYS805	2.049	-7.89
Diosmetin	VAL777 GLN709	2.073 2.348	-6.60
Khahalalide D	THR759	2.385	-7.39
Alpha-tocopherol	MET801	2.433	-7.85
Beta-carotene	----------	No hydrogen bond	-8.58
Choline	CYS802	2.442	-6.30
Sesamol	MET801	2.612	-6.09
Silibinin	VAL777 LEU711	2.197 2.546	-7.16
Xanthoxylin	VAL777	2.509	-6.26
Isofraxidin	MET801 VAL777	2.084 2.117	-7.77
Phloretic acid	MET801 VALL777	1.939 2.034	-7.06
Indole-3-carboxylic acid	GLN990 PHE731 GLN990	2.118 2.003 2.077	-7.81
Garlic	----------	No hydrogen bond	-6.69
Xenognosin	MET801	2.156	-6.86

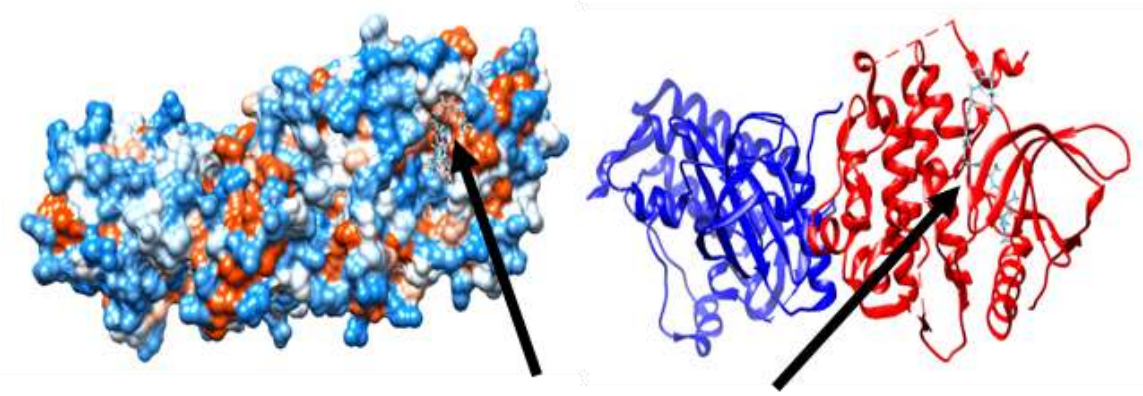

Figure 1. Three-dimensional illustration of the 3PP0-Lycopene complex using the molecular surface (A) and the ribbon model (B)

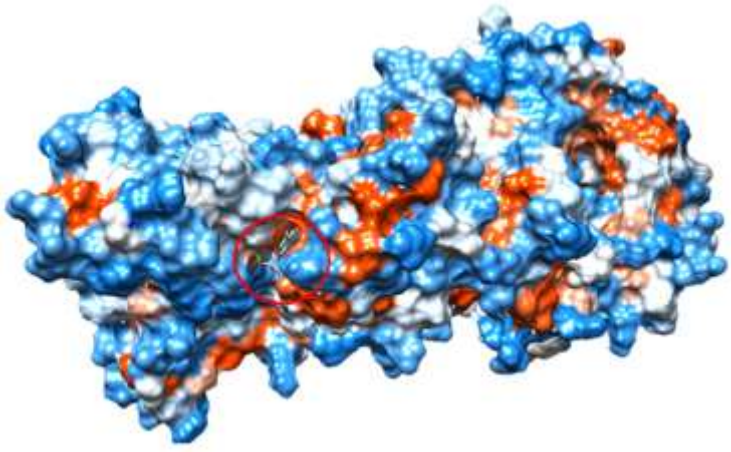

Figure 2. Three-dimensional illustration of the 3PP0 -Anacardic acide complex using the molecular surface.

3,4-Dihydroxybenzoic acid has an interaction energy of -6.22 kcal/mol and forms a hydrogen bond with the residue Gly737**.**

Sinapic acid has an interaction energy of -7.40 kcal/mol and forms a hydrogen bond with the residue Gly732.

Both ligands form hydrogen bonds:

3,4-dihydroxybenzoic acid forms a bond with residue Gly737 and Sinapic acid forms a bond with residue Gly732 within the (Cα) helix. This could destabilize the active open conformation of the activation loop (C-helix-in-DFG-in) and prevent ATP substrate binding to its specific site. It could also block of trans-autophosphorylation of the activation loop, which would maintain its inactive conformation. The ATP binding groove would no longer be accessible, and the tyrosines of the C-terminal tail would not be phosphorylated by the catalytic loop. This induces blockage of the downstream signaling cascade (18). In the present study, Sinapic acid is the target with the best interaction energy value acting on the N-lobe (Cα-helix), with a DG of -7.40 kcal/mol (Figure 3).

Three of these 47 ligands act on the A-loop of the C-lobe of the tyrosine kinase domain. According to the literature, the 20-30 residue sequence of the activation loop (Asp831-Val852 in the EGFR family) contains the conserved base DFG motif (Asp831-Phe-Gly833 in the EGFR family) and extends to an APE (Ala-Pro-Glu) motif also contains the Tyr845 residue, which is one of the target tyrosines for autophosphorylation by the catalytic loop (16-18).

- **Tyrosol** and **Hydroxytyrosol**

Found in olive oil, have interaction energies of -6.36 and -6.91kcal/mol, respectively. Each compound forms a hydrogen bond with an estimated length of 2.129Å for tyrosol and 2.215Å for hydroxytyrosol with residue Arg849 of chain A for tyrosol and chain B for hydroxytyrosol.

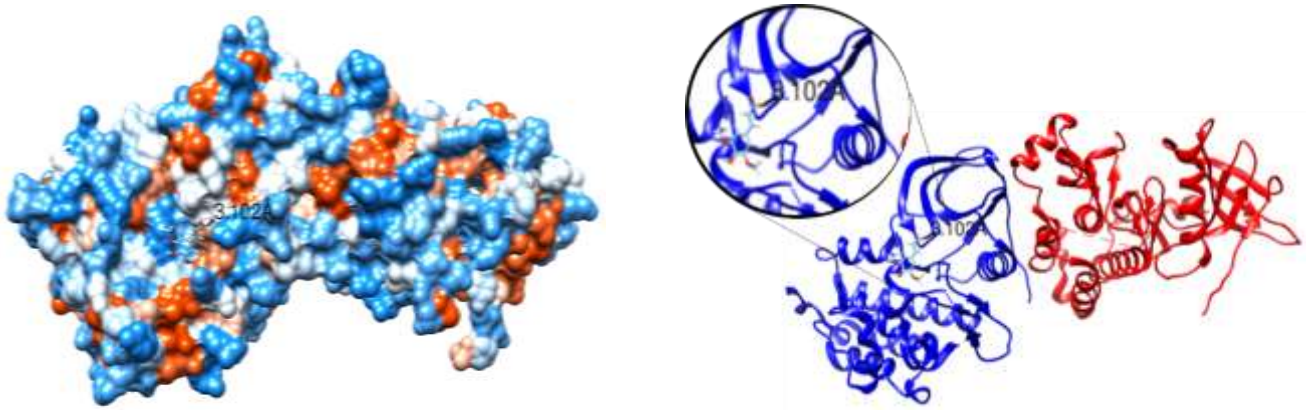

Figure 3. Three-dimensional illustration of the 3PP0 -Sinapic acide complex using the molecular surface (A) and the ribbon model (B).

• **Homocasterone**

Found in beans, has an interaction energy of -7.44 kcal/mol and forms a strong hydrogen bond with Asp838 of the A chain, with an estimated length of 2.716Å.

Thus, the ligand Homocasterone forms a hydrogen bond with the residue Asp838, the ligands Tyrosol and Hydroxytyrosoleach form a hydrogen bond with the residue Arg849 of the activation loop. These ligands can therefore block its activation and passage between conformations:

Inactive (C-helix-out-DF Gout), partially inactive (C-helix-in-DF Gout), and finally active (C-helix-in-DF Gin). This effect occurs by sequestering the trans-autophosphorylation of these tyrosine residues to phosphotyrosines by the catalytic loop after dimerization. Consequently, the ATP binding groove is inaccessible, preventing trans phosphorylation of the C-terminal tail and the recruitment of adaptor proteins (16-20).

This stdudy shows that the ligand with the best interaction energy value that acts on loop A and lobe-C is Homocasterone with a DG of -7.44 kcal/mol (Figure 4).

Other ligands were selected according to their interactions with the following residues in the hydrophobic region of ErbB2: Val773, Met774, Gly776, Val777, Gly778, and Val782 in the αC-β4 loop (21). For exemple, Pelagronidin from grapes forms a hydrogen bond with Gly778. *Sixteen of these ligands form a hydrogen bond with the same Val777 residue (Table 2):

Vanillic acid from olive oil , *Catechin* from blackcurrant, *Malvidin* from grapes, *Fisetin* from strawberries and apples, *Genipin* from *Gardenia Jasminoides Ellis* species, *Opuim,.Gallic acid, Menthol* from peppermint and tea, *Diosmetin* from sage andthyme, Quercetin from red onions or buckwheat, *Luteolin* from green peppers, olive oil and carrots, *Xanthoxylin* from fats, oils, herbs and spices, Isofraxidin Phloretic acid, *Silibinin* extracted from the milk thistle flowers and *Daidzein* present in flax seeds. These two ligands have better interaction energy and strong bonds, but according to the Pharmaco-toxicity tests, these two molecules are mutagenic are toxic. These ligands form hydrogen bonds with residues (valine 777), which belong to the hydrophobic region of ErbB2 (αC-β4 loop). These residues interact with the activation loop andcan destabilize conformational changes from the inactive conformation (C-helix-out-DFGout) to the partially inactive conformation (C-helix-in-DFGout) and finally to the active conformation (C-helix-in DFGin). This occurs by sequestering trans autophosphorylation by the catalytic loop after dimerization. Thus, the ATP binding groove remains covered, there is no trans autophosphorylation of the C-terminal tail, and therefore, no recruitment of adapter proteins (21).

This results indicated that Luteolin, with a DG of -7.92 kcal/mol, is the ligand with the best interaction energy value interacts with the residues of the hydrophobic region in the loop (αC-β4) (Figure 5).

According to the literature, the residues of the hydrophobic region of loop A are Iso861, Thr862, Phe864, Leu866 and Leu869. The interaction between the two active and inactive conformations ,which takes place between Ser783 with in the hydrophobic region of ErbB2 (Cα-β4 loop) and residue Iso861 of loop A,allows the interaction between the latter and the αC-β4 loop (21).

The ligands Vanillic acid, Catechins, Malvidin, Fisetin, Genipin, Pelagronidin and 4 p-hydroxybenzoic acid form hydrogen bonds with residues belonging to the hydrophobic region of ErbB2 (Cα-β4 loop) and interact

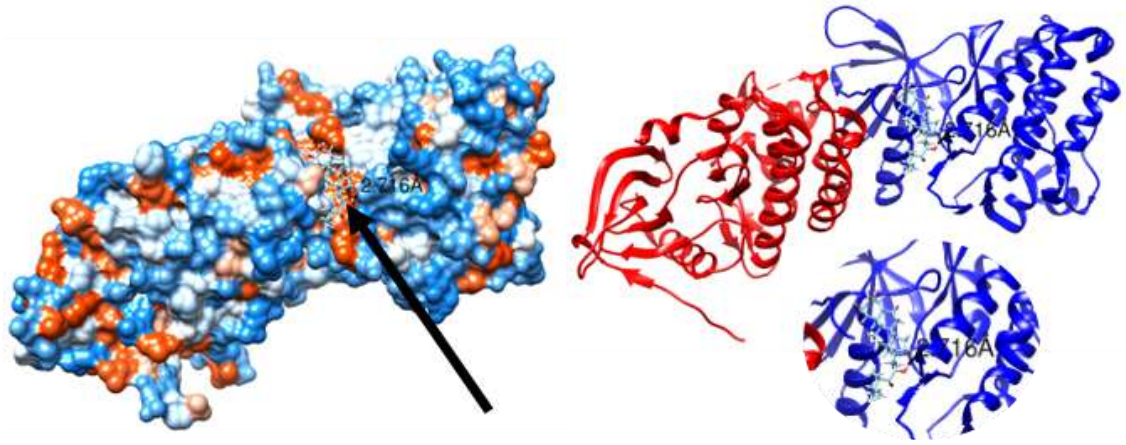

Figure.4: Three-dimensional illustration of the 3PP0 – Homocasterone complex using the molecular surface (A) and the ribbon model (B).

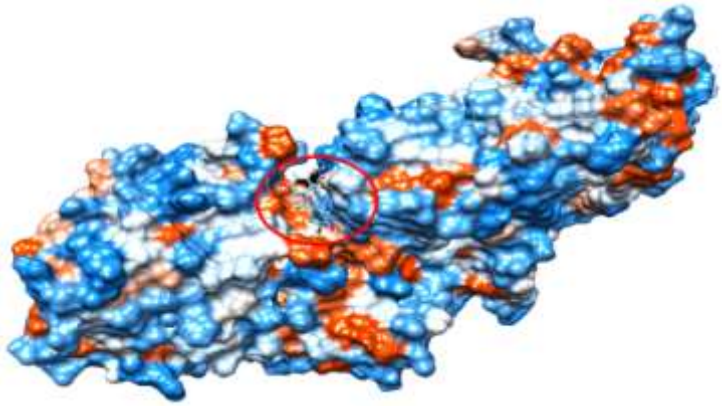

Figure 5. Three-dimensional illustration of the 3PP0 -Luteolin complex using the molecular surface.

with the activation loop. These ligands can destabilize conformational changes from the inactive conformation (C-helix-out-DFGout) to the partially inactive conformation (C-helix-in-DFGout) and finally to the active conformation (C-helix-in-DFGin). This occurs by sequestering trans autophosphorylation by the catalytic loop after dimerization. Consequently, the ATP-binding groove remains covered, preventing trans autophosphorylation of the C-terminal tail and the recruitment of adaptor proteins (21).

In this study, malvidin with a DG of -7.32 kcal/mol, is the ligand that with the best interaction energy value and interacts with residues in the hydrophobic region (in the Cα-β4 loop) (Figure 6).

The remaining five ligands were selected according to their interactions with residues between the tyrosines of the C-terminal tail (Tyr874, Tyr992, Tyr1048, Tyr1068, Tyr1086, Tyr1101, and Tyr1173) (Table 2) (17).

The aforementioned tyrosines correspond to residues trans-autophosphorylated by the catalytic loop. Therefore,ligands that form hydrogen bonds with residues, that lie between these tyrosines, such as Epicatechin and Naringenin with residue Gln943, Apigenin with residue Cys947, Genistein with residue Cys947, may be susceptible to sequesteration of the interaction between the C-terminal tail and the catalytic loop. Therefore the tyrosines will not be trans-autophosphorylated, the adaptor proteins will not be recruited (17).

Apigenin has the best interaction energy value with residues between the tyrosines of the C-terminal tail, with a DG of -7.55 kcal/mol (Figure 7).

3.4. Ligands Interacting with Specific Residues of the HER-2 Receptor Tyrosine Kinase Domain

One ligand acts on the P-loop of the N-lobe of the tyrosine kinase domain. Non-conserved ligands specific to HER2 were selected based on their interaction with

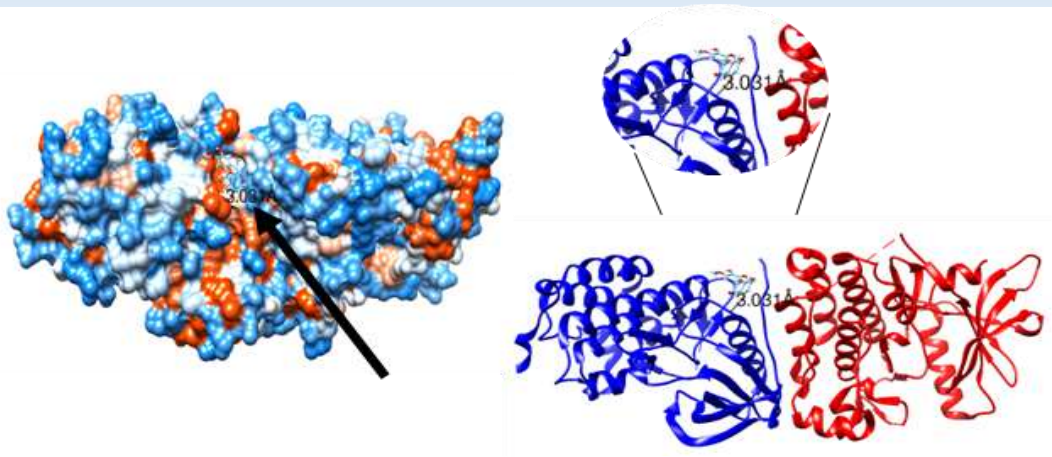

Figure 6. Three-dimensional illustration of the 3PP0 -Malvidin complex using the molecular surface (A) and the ribbon model (B).

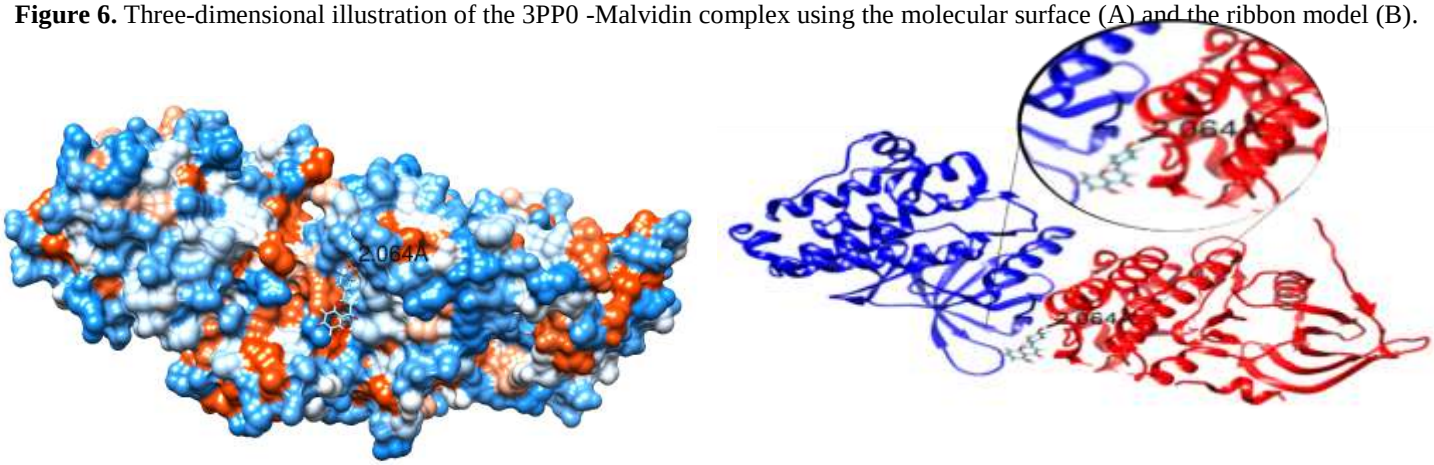

Figure 7. Three-dimensional illustration of the 3PP0- Apigenin complex using the molecular surface (A) and the ribbon model (B).

residuesfrom residue Leu726 to Val734 (9). Pinoresinol, found in olive oil, has an interaction energy of -7.48 kcal/mol and forms a strong hydrogen bond whose length, estimated at 2.613 Å with the residue Ser728 (Figure 8).

This ligand forms a hydrogen bond with the P-loop, which can destabilize the open active conformation of the activation loop (C-helix-in-DFGin) and prevent binding of the ATP substrate to its specific site. This blocks the downstream signaling cascade (16).

One ligand that acts on the C-terminal tail of the non-conserved tyrosine kinase domain in the EGFR family was selected based on its interaction with the residues extending from residue Pro999 to Leu1009 and was specific for HER2, The caffeic acid in olive oil has an interaction energy of -6.71 kcal/mol and forms a hydrogen bond with the residue Leu1000, estimated to be 2.044 Å long (9).

Two ligands acting on the P-loop of the N-lobe of the tyrosine kinase domain wereselected according to their interaction with residues in the sequence extending from Leu726 to Val734 (9). Biochanin A from Soybean has an interaction energy of -6.67kcal/mol with Leu 726, and α-Linolenic acid from soybeanhas an interaction energie of -7.69 kcal/mol and forms a strong hydrogen bond with an estimated length of 3.160Å with the residue Ser728.

The Cα-helix and the P-loop are in close proximity and interact with ATP required for trans-autophosphorylation in the ATP -binding site (9, 17). The ligands such as Biochanin A, Pinoresinol and α-Linolenic acid form hydrogen bonds with the P-loopand can destabilize the active , open conformation of the activation loop (C-helix-in-DFGin), thus preventing no ATP substrate binding at its specific site and inducing blockage of the downstream signaling cascade (18).

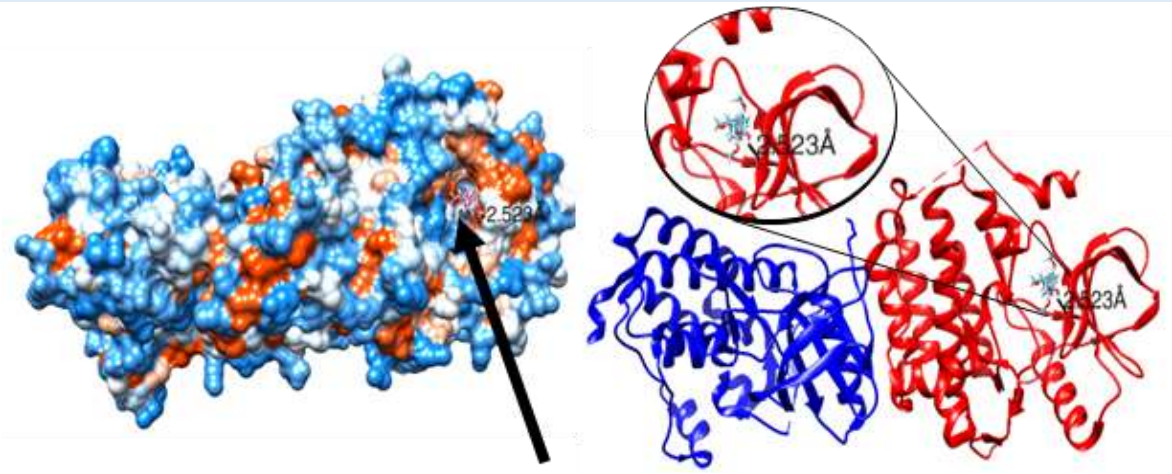

Figure 8. Three-dimensional illustration of the 3PP0-Pinoresinol complex using the molecular surface (A) and the ribbon model (B).

The ligand that has the best interaction energy value with the N-lobe P-loop residues specific for HER2 is α-Linolenic with a DG of -7.69 kcal/mol (Figure 9).

The remaining ligand that acts on the helix (Cα) of the N-lobe of the tyrosine kinase domain was selected according to its interaction with the residues in the stretch extending from Pro761 to Ala775.

One ligand acts on the C-lobe activation loop of the HER2 receptor-specific tyrosine kinase domain was selected based on their interaction with residues spanning from Asp863 to Val884, including the DFG motif (from residue Asp863 to residue Gly865). Kaempferol Present an interaction energy of -6.88 Kcal/mol and forms a hydrogen bond with the residue ASP863, estimated at 3.572 Å (Figure 10).

3.5. Visualization of Lapatinib Docking Results with 3PP0

Lapatinib is a ligand which is used as a chemical treatment in HER2+ breast cancer that is specific for inhibiting protein tyrosine kinase signaling pathways (22). Our findings indicate that it has an interaction energy of -7.58 kcal/mol and forms a hydrogen bond whose length is estimated at 2.303Å with the Val777 residue of the A chain. Lapatinib, therefore, interacts with the residues of the hydrophobic region of ErbB2 in the αC-β4 loop (Figure 11A).

3.6. Comparison of the Two Ligands Lapatinib and Quercetin

The quercetin found in red onions or buckwheatsssshas an interaction energy of -7.28 Kcal/mol and forms a hydrogen bond with the residue Val777 ,whose length is estimated at 2.130 Å (Figure 11A).

Based on the visualizing the results of the two ligands, Lapatinib and quercetin (the former a modified chemical ligand and the latter a natural ligand primarily found in red onions), it appears that each forms a hydrogen bond of different lengths: 2.303Å for Lapatinib and of 2.130Å for a quercetin, both with the residue valine777 in the hydrophobic region (in the αC-β4 loop).The interaction energies are -7.58kcal/mol for Lapatinib and-7.28kcal/mol for quercetin. Lapatinib certainly has a better interaction energy, but it is a chemical compound with side effects. Quercetin,on the other hand, has other beneficial effects on health in addition to its possible inhibition of 3PP0.

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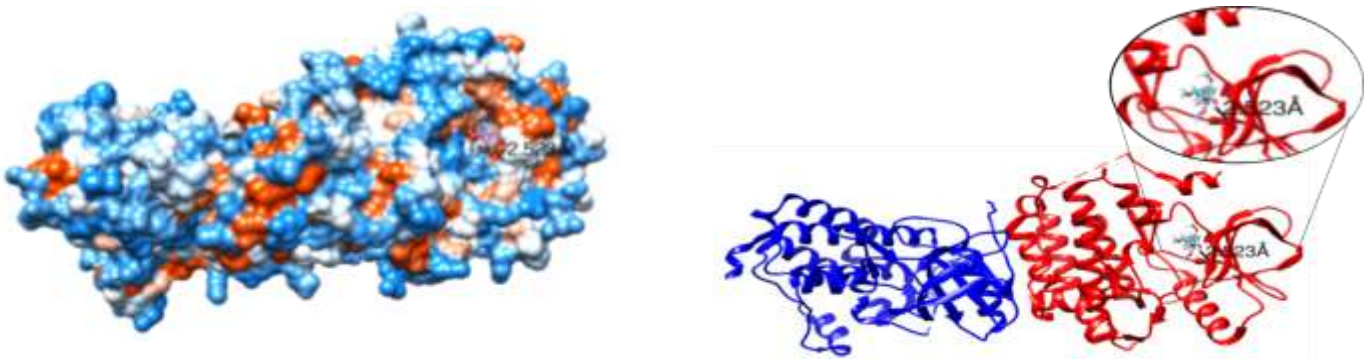

Figure 9. Three-dimensional illustration of the 3PP0 - α-Linolenic complex using the molecular surface (A) and the ribbon model (B).

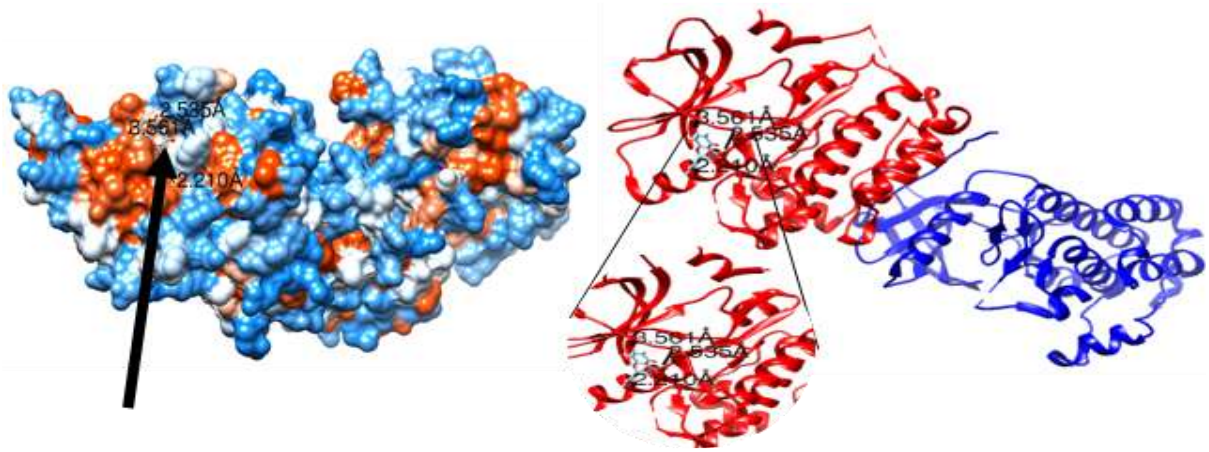

Figure 10. Three-dimensional illustration of the 3PP0 - Kaempferol complex using the molecular surface (A) and the ribbon model (B).

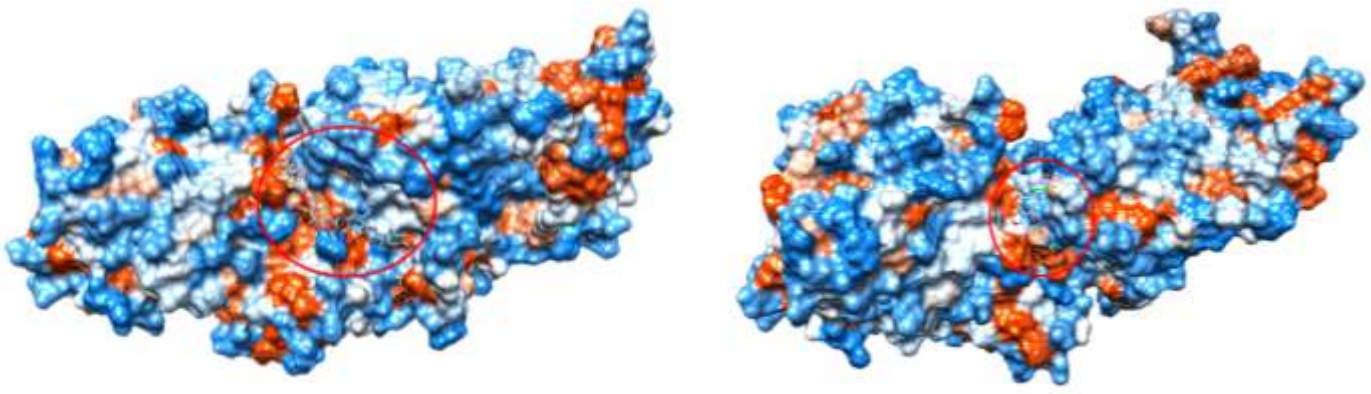

Figure 11. Three-dimensional illustration of the 3PP0 - Lapatinib (A) and 3PP0 - meletin (B) using the molecular surface

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4. Discussion

Indeed, quercetin, also known as vitamin P, is a food-derived compound and a bioflavonoid found in the pigments of colored fruits and vegetables. These include red onions, spinach, turmeric, apples, red grapes, carrots, berries, broccoli, green tea, lovage, chocolate or red wine. As a natural antioxidant, quercetin helps fight against oxidative stress by capturing and blocking the activity of free radicals and inhibiting the oxidation of lipids. Quercetin is also involved in regulation of signaling pathways, cell cycle proliferation and the immune response. In summary, investigating *in silico* before proceeding to the experimental stage can save a great deal of time and money. In silico technologies can predict a number of ADMET factors, toxicological effects, and likely active medication. In this study, the oral bioavailability of drugs was predicted using several prediction methodologies, which could lead to the development of safer, innovative pharmaceuticals. After analyzing the screening and molecular docking studies, we found that many natural products could be used as potential HER2 antagonists to treat of breast cancers. Additional wet-lab research is necessary to further evaluate these selected compounds.

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None.

Authors' Contribution

Conceived and designed experiments: N. L.
Conducted experiments: N. L.
Analyzed data: N. L, N. M, S KT.
Wrote the paper: N. L.

Ethics

In this article, the authors have observed all ethical points, including those related to plagiarism, double publication, data distortion and data manipulation.

Conflict of Interest

The author declares no known competing interests.

Grant Support

This study

Data Availability

The data supporting the findings of this study are available upon request from the corresponding author.

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