JOURNAL OF NATURAL REMEDIES
DOI: 10.18311/jnr/2025/48397
RESEARCH ARTICLE

An *In Silico* Computational Screening of Bioactive Molecules Retrieved from Selected Herbs against the Target Breast Cancer Gene 1

S. Karthik Nagarajan[1*], C. B. S. Bharath Christian[1], A. Siva[2] and S. Merish[3]

[1]National Institute of Siddha, Ministry of Ayush, Government of India, Chennai – 600047, Tamil Nadu, India; drkarthiksiddha2016@gmail.com

[2]Department of Clinical Research, Siddha Regional Research Institute (Central Council for Research in Siddha, Ministry of Ayush, Govt. of India), Puducherry – 605013, India

[3]Department of Pharmacology, ATSVS Siddha Medical College, Munchirai, Kanyakumari – 629171, Tamil Nadu, India

Abstract

Background: Breast cancer, one of the most prevalent cancers globally, affects millions of women each year, with increasing incidence rates. Conventional treatments often have limitations, including side effects and drug resistance. Molecular docking of bioactive compounds from medicinal herbs offers a novel approach to identifying natural inhibitors that target specific cancer genes, potentially leading to more effective, less toxic therapies. **Objective:** This study aims to analyse the molecular docking of bioactive molecules in the raw drugs of selective herbs against Breast Cancer Gene 1 by *in silico* computational screening. **Methodology:** From the literature survey, nine major bioactive compounds having a higher therapeutic value from the raw drugs of the selected herbs had been selected as ligands and Breast Cancer Gene 1 (PDB ID: 4Y2G) as a potential protein drug target for *in silico* molecular docking analysis using Autodock 4 software. **Results:** Among the 9 selected bio-active molecules in the selected 8 herbs, the results showed that the estimated binding energies in kcal/mol of the selected compounds towards the target Breast Cancer Gene 1 with its acidic and polar residues were as follows: Friedelin -8.21, Hinokiflavone -6.89, Curcumin -6.25, Picein -6.20, Bhilawanol A -5.93, Tinosporide -5.79, Kaempferol -5.47, β-pinene -4.02, Plumbagin -4.01. The ligand interactions with the target revealed the following scores in ascending order: β-pinene scored four, Plumbagin-five, Kaempferol-five, Hinokiflavone-six, Friedelin-seven, Tinosporide-eight, Curcumin-eight, Picein-eight, and Bhilawanol A-nine. Bhilawanol A exhibits the strongest interaction. **Conclusion:** Molecular docking studies of bioactive compounds derived from the raw drugs of the selected herbs suggest their potential as inhibitory potential of the Breast Cancer Gene 1, which is involved in the development of breast cancer.

Major Findings: The study identified Bhilawanol A as the most promising bioactive compound with the strongest inhibitory potential against Breast Cancer Gene 1, based on molecular docking analysis. Other compounds like Friedelin, Curcumin, and Picein also showed significant binding affinities, suggesting their potential as natural inhibitors for breast cancer therapy.

Keywords: Ayush, Breast Cancer, Breast Cancer Gene 1, Molecular Docking, Network Pharmacology, *Siddha* Medicine

Abbreviations: Akt/NF-kB = Protein Kinase B/Nuclear Factor kappa B; ASN = Asparagine; BRCA = Breast Cancer Gene 1; DNA = Deoxyribonucleic Acid; LEU = Leucine; LYS = Lysine; MMP-9 = Matrix Metalloproteinase-9; PDB = Protein Data Bank; PDBQT = Protein Data Bank, Partial Charge (Q), and Torsion (T); PHE = Phenylalanine; PRO = Proline; SER = Serine; THR = Threonine; VAL = Valine; VEGF = Vascular Endothelial Growth Factor

*Author for correspondence

Article Received on: 03.01.2025 Revised on: 21.05.2025 Accepted on: 27.05.2025

1. Introduction

Breast cancer is one of the most prevalent cancers worldwide, affecting millions each year. Globally, it is the most common cancer among women, with over 2 million new cases annually, accounting for nearly 25% of all female cancer cases[1]. In India, breast cancer is also the leading type of cancer among women, and its incidence has been rising due to lifestyle changes, genetic factors, and late detection. India faces unique challenges in breast cancer care, as a significant proportion of cases are detected at later stages, affecting treatment outcomes and survival rates[2].

Herbal compounds are increasingly researched for their potential as natural, less-toxic alternatives in cancer therapy[3]. Phytochemicals from plants have shown promising results in molecular docking studies against the BRCA1 gene and other cancer-related genes. These compounds may bind to critical sites on BRCA1, potentially preventing DNA repair in cancer cells and leading to their death[4].

Herbal-based molecular docking studies offer a promising route to develop effective, affordable, and accessible breast cancer treatments, particularly beneficial in countries like India, where herbal medicine is culturally integrated. In recent years, the quest for effective treatments against breast cancer has shifted focus towards the potential of various herbal plants[5].

Among these, *Vitex negunda* (Franch.) Rehder (*Nochi*), *Rhus succedanea* (L.) (*Karkadagasingi*), *Curcuma longa* (L.) (*Manjal*), *Picrorhiza kurroa* Royle ex Benth. (*Kadugurrhogni*), *Semicarpus anacardium* (L.) (*Serankottai*), *Tinospora cordifolia* (Thunb.) Miers (*Seenthil*), *Boerhavia diffusa* (L.) (*Mookirattai*), and *Plumbago indica* (L.) (*Kodiveli*) have emerged as a promising herb which was mentioned in the *Siddha* textbook, *Siddha* Materia Medica – Plant Division[6].

Research into targeted therapies has become essential, especially focusing on cancer-related genes like BRCA1, a gene responsible for DNA repair. Mutations or dysfunctions in BRCA1 significantly increase the risk of breast and ovarian cancers[7].

Molecular docking, a computational technique that models interactions between small compounds and specific gene targets, has become a valuable approach in cancer research[8]. This method helps identify potential inhibitors that can block cancer-related genes like BRCA1, effectively disrupting cancer progression at the molecular level[9].

In *Siddha* medicine, "*Sidalai*" refers to "*Puttru Noi*" or cancer, a term derived from the behaviour of white ants, which construct ant hills by eroding the material inside. The Tamil word "*Puttru*" translates to "white ant's anthill", and synonyms such as *Vanmegam* and *Purai Odiya Pun* are also used. *Siddhars* likely compared cancer's uncontrolled and destructive growth to the spread of an anthill, which expands while eroding its surroundings, leading to the term "*Sidalai*" for cancer. Various terms, including *Thunmongisam*, *Katti*, *Puttru*, *Soolai*, and *Vippuruthi*, are used in *Siddha* literature to describe growths or tumours, but there is no consistent standard for naming them. Some terms like *Katti*, *Soolai*, and *Vippuruthi* are applied to non-malignant conditions, such as abscesses or chronic inflammation with intense pain, while *Vippuruthi* specifically refers to neoplastic growths in internal organs[10].

Several classical *Siddha* texts discuss neoplastic growths, including *Theriyar Vaithiyam - 1001*, *Pullippani - 500*, *Theriyar Vaidhiya Kaviyam - 1500*, *Agasthiyar Munivar Kanma Kaandam - 400*, *Boghar Satha Kaandam - 7000*, *Agasthiyar Munivar Aruliya Chendooram - 300*, *Agasthiyar Vaidhya Kaaviyam - 1500*, *Agasthiyar Vaidhya Rathnatch Churukkam – 360*, *Agasthiyar Pallu - 200*, and *Agasthiyar Paripuranmnam - 400*.

Specific types of "*Puttru Noi*" (cancers) mentioned in these texts include *Yoni Puttru Noi* (vaginal cancer), *Linga Puttru Noi* (penile cancer), *Kanna Puttru Noi* (buccal mucosa cancer), *Eral Puttru Noi* (lung cancer), *Sevi Puttru Noi* (cancer of the ear), *Asana Puttru Noi* (rectal cancer), *Thondai Puttru Noi* (throat cancer), *Nakku Puttru Noi* (tongue cancer), *Vaai Puttru Noi* (buccal cancer), *Algul Puttru Noi* (vaginal cancer), *Maarbu Puttru Noi* (breast cancer), *Kadithada Puttru Noi* (cancer of the urinary tarct), *Kabala Puttru Noi* (brain tumour), *Paatha Puttru Noi* (leg cancer), and *Mooku Puttru Noi* (nasal cancer). These references demonstrate *Siddha* medicine's comprehensive approach to recognising and classifying various forms of cancer and neoplastic diseases.

Siddha medicine, with its ancient roots, comprehensively addresses various forms of cancer and their treatment, including the use of herbs, metals, and minerals. Among the cancers mentioned in *Siddha*

literature, *Maarbu Puttru Noi* refers to breast cancer, is also recognised. *Siddha* treatments often emphasise herbal remedies, along with the incorporation of metals and minerals, for addressing such disorders.

This study aims to evaluate the molecular docking of bioactive compounds from selected herbal raw drugs against Breast Cancer Gene 1 (BRCA1) using *in silico* computational screening methods.

2. Materials and Methods

Docking analysis was accomplished using AutoDock Version 4.2.6. A common molecular screening method for determining the binding energy between each ligand's three-dimensional structure and the target protein was used to undertake *in silico* molecular docking analysis for all selected bioactive molecules present in the raw drugs of herbs with the potential drug target Breast Cancer Gene 1 (BRCA1) [PDB ID: 4Y2G], for anti-breast cancer therapy.

2.1 Selection and Preparation of Ligands

Nine bioactive compounds from the selected raw drugs of herbs, such as Friedelin, Hinokiflavone, Curcumin, Picein, Bhilawanol A, Tinosporide, Kaempferol, β-pinene and Plumbagin were retrieved from published literature. Next, they were obtained in SDF format from the PubChem database. By converting these ligands to PDB format using the open babel program, and preparing them for docking analysis. The torsion requirements needed for proper binding were then defined using the AutoDock 4.2.6 application. Table 1 presents the vernacular and botanical names of the selected ligands for docking from the raw drugs of the herbs which were selected for the docking process. The ligands that were chosen for docking analysis are included in Table 2 along with their molar weight (g/mol), molecular formula, H-bond donor, H-bond acceptor, and rotatable bonds.

Table 1. Selected ligands for docking from the raw drugs of the selected herbs

Vernacular Name (Tamil)	Botanical name of the herbs	Selected Phytochemicals	PUBCHEM ID
Nochi	*Vitex negunda* (Franch.) Rehder	Friedelin[11]	5282213
		β-pinene[11]	439720
Karkadagasingi	*Rhus succedanea* (L.)	Hinokiflavone[12]	5310653
Manjal	*Curcuma longa* (L.)	Curcumin[13]	969516
Kadugurrhogni	*Picrorhiza kurroa* Royle ex Benth.	Picein[14]	5282290
Serankottai	*Semicarpus anacordium* (L.)	Bhilawanol A[15]	16153967
Seenthil	*Tinospora cordifolia* (Thunb.) Miers	Tinosporide[16]	6904392
Mookirattai	*Boerhavia diffusa* (L.)	Kaempferol[17]	5280863
Kodiveli	*Plumbago indica* (L.)	Plumbagin[18]	5281604

Table 2. Properties of Ligands selected for docking analysis

Compound	Molar weight (g/mol)	Molecular Formula	H Bond Donor	H Bond Acceptor	Rotatable bonds
Friedelin	426.7	$C_{30}H_{50}O$	0	1	0
β-pinene	136.23	$C_{10}H_{16}$	0	0	0
Hinokiflavone	538.5	$C_{30}H_{18}O_{10}$	5	10	4
Curcumin	368.4	$C_{21}H_{20}O_6$	2	6	8
Picein	298.29	$C_{14}H_{18}O_7$	4	7	4
Bhilawanol A	318.493	$C_{21}H_{34}O_2$	2	2	1
Tinosporide	374.4	$C_{20}H_{22}O_7$	1	7	1
Kaempferol	286.24	$C_{15}H_{10}O_6$	4	6	1
Plumbagin	188.182	$C_{11}H_8O_3$	1	3	0

2.2 Protein Preparation

The three-dimensional crystal structure of Breast Cancer Gene 1 (BRCA1) [PDB ID: 4Y2G] was extracted from the Protein Data Bank (PDB), shown in Figure 1, and its energy-minimised, and converted to the appropriate PDBQT formats. The ligand molecules were first positioned, oriented, and torsored randomly. When the device docked, all spinning torsions were relieved. Every docking experiment came from two distinct runs, each of which was intended to end after a maximum of 250000 energy assessments. The population size was set to 150. A translational step of 0.2 Å and quaternion and torsion steps of 5 were used in the search.

Protein structures were cleaned by eliminating the lead components that were already there; water molecules were broken; polar hydrogens were included in the computation of Kollman's charges[19], and AutoDock 4.2.6 software was used to describe the merging of non-polar and rotatable bonds.

The selected ligands for the molecular docking studies against Friedelin, Hinokiflavone, Curcumin, Picein, Bhilawanol A, Tinosporide, Kaempferol, β-pinene, and Plumbagin, each characterised by distinct 2D and 3D structures, as Figure 2.

Friedelin features a rigid tetracyclic structure with a triterpenoid framework, characterised by multiple fused rings and hydroxyl groups, providing hydrophobic interactions. Hinokiflavone presents a flavonoid structure with two aromatic rings connected by a methylene bridge, while Curcumin features a linear, unsaturated diketone structure with two aromatic rings, allowing extensive conjugation. Picein, a flavonoid glycoside, has a flavonoid backbone with a sugar moiety attached, enhancing solubility. Bhilawanol A showcases a complex, oxygenated steroid-like structure, featuring fused rings and hydroxyl groups.

Tinosporide has a triterpenoid backbone with a complex ring system, contributing to its bioactivity. Kaempferol presents a flavonoid structure with a hydroxylated aromatic ring system, while β-pinene features a bicyclic monoterpene structure, offering rigidity and hydrophobicity. Plumbagin, a naphthoquinone, has a rigid aromatic core with two carbonyl groups, providing electrophilic properties for interactions. These structures exhibit diverse molecular conformations, from rigid frameworks to flexible and conjugated systems, contributing to their varied biological activities. These structural characteristics provide insights into the potential binding interactions with BRCA1.

2.3 Molecular Docking Methodology

Following the preparation of the protein target and ligand selection, Autodock 4.2.6 was used to establish

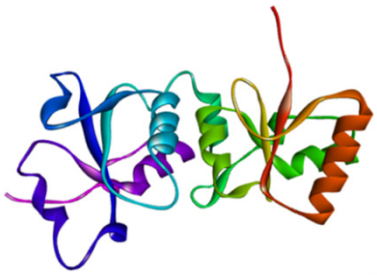


Figure 1. Receptor structure in 3D – Breast Cancer Gene 1 (BRCA1) [PDB ID 4Y2G].

Table 3. Summary of the molecular docking studies of the selected bioactive molecules against Breast Cancer Gene 1 (BRCA1)

Compounds	Estimated Free Energy of Binding (kcal/mol)	Estimated Inhibition Constant, Ki	Electrostatic Energy (kcal/mol)	Total Intermolecular Energy (kcal/mol)	Interaction Surface
Friedelin	-8.21	964.16 nM	-0.02	-8.21	639.440
Hinokiflavone	-6.89	8.90 uM	-0.21	-7.90	785.545
Curcumin	-6.25	26.40 uM	-0.17	-6.97	746.291
Picein	-6.20	28.63 uM	-0.08	-5.88	558.313
Bhilawanol A	-5.93	45.26 uM	-0.13	-7.54	532.218
Tinosporide	-5.79	56.65 uM	-0.30	-6.19	503.005
Kaempferol	-5.47	97.45 uM	-0.15	-5.83	540.761
β-pinene	-4.02	1.13 mM	-0.01	-4.02	357.102
Plumbagin	-4.01	1.15 mM	-0.03	-4.31	391.519

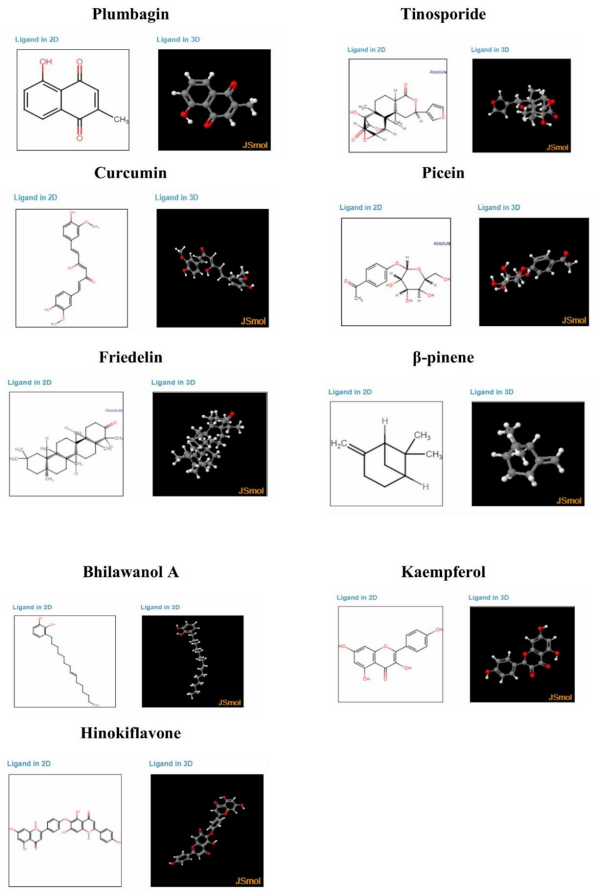


Figure 2. 2D and 3D Structure of selected ligands for molecular docking.

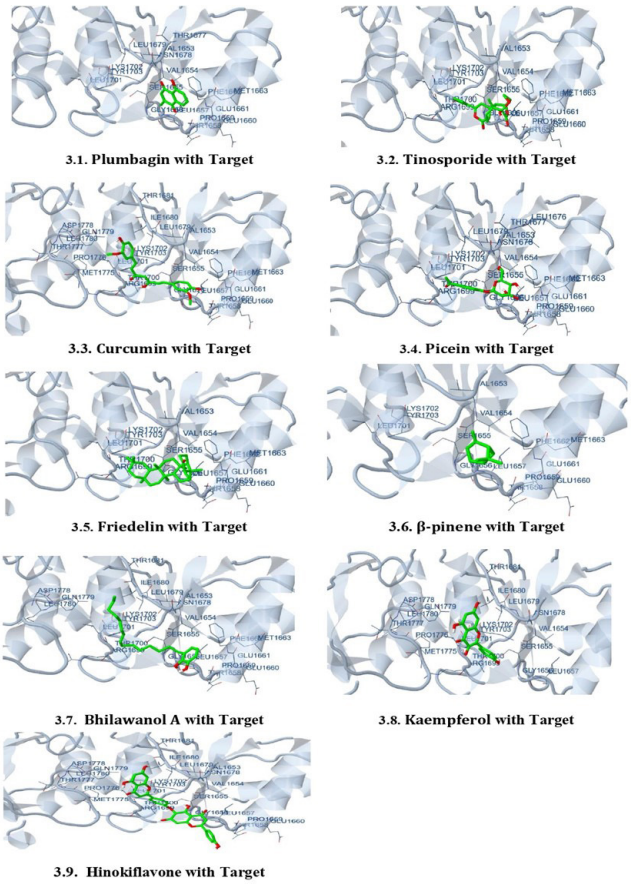


Figure 3. Docking Pose of the ligands with the target Breast Cancer Gene 1 (BRCA1).

Table 4. Amino acid Residue Interaction of Lead Breast Cancer Gene 1 (BRCA1) with selected bioactive molecules

Compounds	Inter actions	Amino Acid Residues										
Plumbagin	5	1654 VAL	1657 LEU	1662 PHE	1678 ASN	1702 LYS						
Tinosporide	8	1654 VAL	1655 SER	1657 LEU	1659 PRO	1662 PHE	1700 THR	1702 LYS				
Curcumin	8	1654 VAL	1655 SER	1657 LEU	1659 PRO	1662 PHE	1680 ILE	1700 THR	1701 LEU	1702 LYS	1776 PRO	1779 GLN
Picein	8	1654 VAL	1655 SER	1657 LEU	1662 PHE	1678 ASN	1700 THR	1701 LEU	1702 LYS			
Friedelin	7	1654 VAL	1655 SER	1657 LEU	1662 PHE	1700 THR	1701 LEU	1702 LYS				
β-pinene	4	1654 VAL	1657 LEU	1662 PHE	1702 LYS							
Bhilawanol A	9	1654 VAL	1655 SER	1657 LEU	1659 PRO	1662 PHE	1679 LEU	1680 ILE	1700 THR	1701 LEU	1702 LYS	1779 GLN
Kaempferol	5	1655 SER	1679 LEU	1680 ILE	1700 THR	1701 LEU	1702 LYS	1776 PRO	1779 GLN			
Hinokiflavone	6	1655 SER	1659 PRO	1679 LEU	1680 ILE	1700 THR	1701 LEU	1702 LYS	1776 PRO	1779 GLN		

molecular docking. The dimensions of x, y, and z were set at 60×60×60 with a resolution of 0.375 Å to generate binding. To obtain the best docking configurations, the grid box was centred. Once the grid file has been saved as a (.gpf) file, launch autogrid. Using a preset run count of 10, the Lamarckian genetic technique was used to compute the docking. Following the execution of AutoDock, the dock file was saved as a dpf. A (.dlg) file included the final docked findings, which included details on the inhibition constant, binding energy, and binding residues. The structures showing how ligands and proteins interact were seen using Discovery Studio[20,21].

The docking protocol involved defining the active site on the Breast Cancer Gene 1 (BRCA1) and allowing flexible ligand conformations, with the top-ranked binding poses selected based on the estimated free energy of binding and interaction profiles. This comprehensive approach facilitated the identification of potential BRCA1 inhibitors, contributing to the understanding of their therapeutic applicability[22].

3. Results

Table 3 summarises the molecular docking studies of bioactive molecules against Breast Cancer Gene 1 (BRCA1), detailing the estimated free energy of binding, estimated inhibition constant (Ki), electrostatic energy, total intermolecular energy, and interaction surface. Table 4 outlines the amino acid residue interactions of the bioactive molecules with Breast Cancer Gene 1 (BRCA1). Figure 3 illustrates the docking poses of the ligands with the target, Breast Cancer Gene 1 (BRCA1). Figure 4 presents the 2D interaction plot analysis of the ligands with the target, Breast Cancer Gene 1 (BRCA1). Figure 5

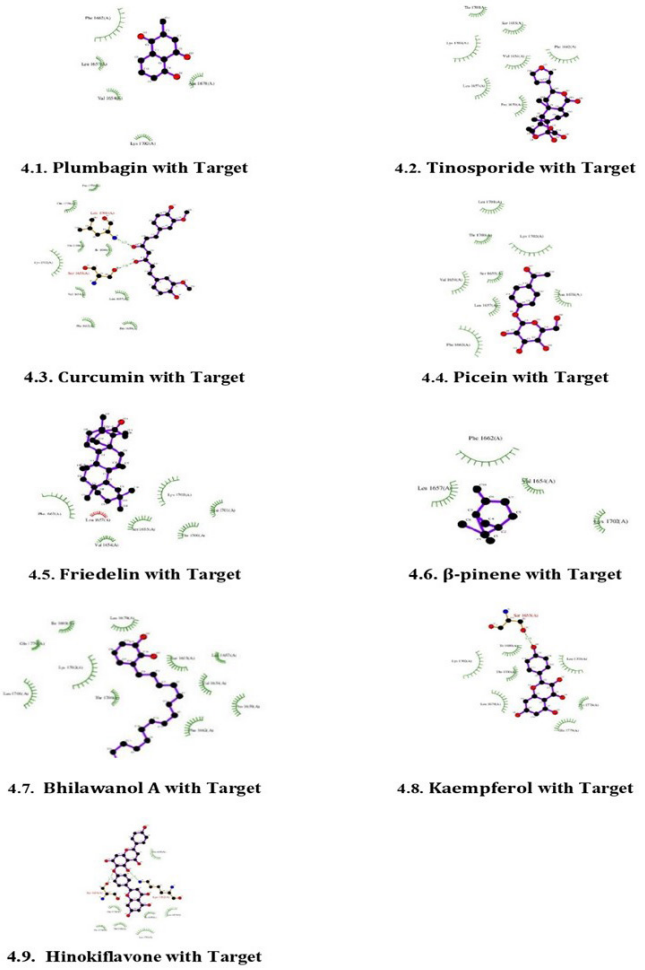


Figure 4. 2D interaction plot analysis of the ligands with the target Breast Cancer Gene 1 (BRCA1).

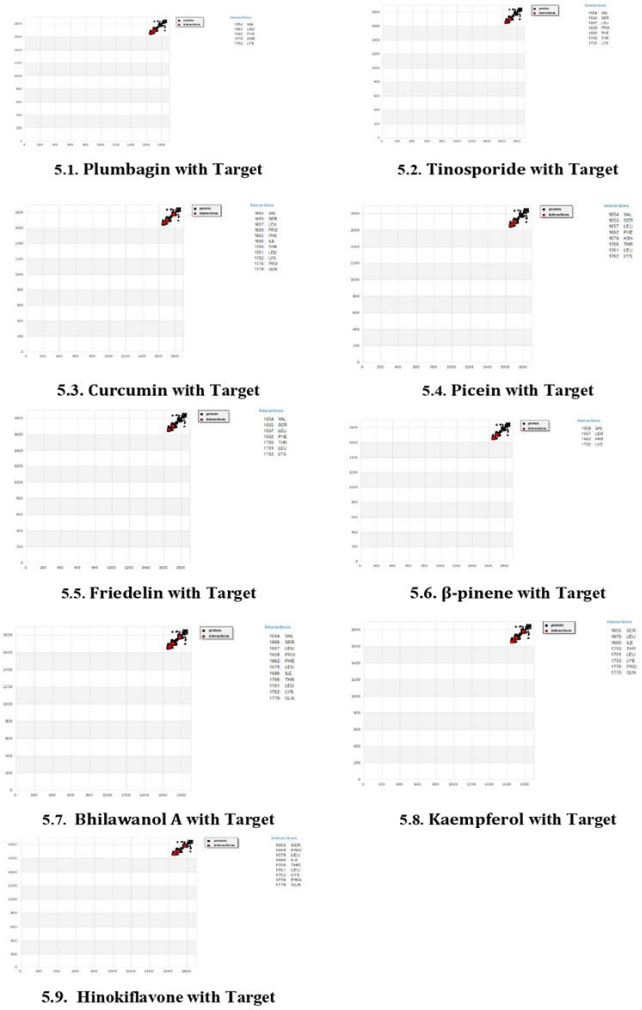


Figure 5. Hydrogen bond interactions along with core amino acid analysis of the ligands with the target Breast Cancer Gene 1 (BRCA1).

illustrates the hydrogen bond interactions along with core amino acid analysis of the ligands with the target, Breast Cancer Gene 1 (BRCA1).

4. Discussion

Molecular docking has become an essential tool in exploring potential therapies for breast cancer, one of the most common cancers globally. It enables the evaluation of how bioactive compounds from herbs interact with specific molecular targets involved in cancer progression, offering insights into the mechanisms through which these herbs exert their anticancer effects. By applying molecular docking, researchers can enhance the efficacy of *Siddha* treatments for breast cancer, bridging traditional herbal knowledge with modern pharmacology.

Additionally, docking studies can uncover synergistic effects between multiple herbal compounds, aligning with the holistic approach of *Siddha* medicine. This approach not only validates the therapeutic potential of herbal compounds in breast cancer treatment but also paves the way for developing targeted therapies rooted in ancient wisdom.

From Table 2, the selected ligands for molecular docking against BRCA1 vary significantly in their molecular properties, influencing their binding potential. *Friedelin*, with a molar weight of 426.7 g/mol, has no hydrogen bond donors or rotatable bonds, which may support stable binding. *β-Pinene*, a small molecule with a weight of 136.23 g/mol, similarly lacks hydrogen bond donors and acceptors, as well as rotatable bonds, potentially reducing flexibility in binding. *Hinokiflavone* and *Curcumin*, larger and more complex compounds, possess numerous hydrogen bond donors and acceptors (5 and 10, and 2 and 6, respectively) and multiple rotatable bonds, which can enhance interaction versatility with the BRCA1 binding site. Compounds like *Picein* and *Kaempferol* also show multiple hydrogen bond donors and acceptors, indicating strong binding potential through hydrogen bonding. In contrast, *Plumbagin* and *Bhilawanol A* exhibit simpler profiles with fewer donors and acceptors, suggesting varied but potentially less robust interactions. These molecular properties give insight into each compound's binding flexibility and potential effectiveness as BRCA1 inhibitors.

From Table 3, among the compounds tested, *Friedelin* shows the strongest binding affinity with a free energy of -8.21 kcal/mol, suggesting a stable interaction with BRCA1. Other compounds with notable affinities include *Hinokiflavone* (-6.89 kcal/mol), *Curcumin* (-6.25 kcal/mol), and *Picein* (-6.20 kcal/mol), indicating moderate binding stability. Compounds like *Bhilawanol A* and *Tinosporide* also demonstrate potential, though with slightly lower binding energies (-5.93 and -5.79 kcal/mol, respectively). *Kaempferol*, *β-pinene*, and *Plumbagin* show comparatively weaker affinities, with binding energies ranging from -5.47 to -4.01 kcal/mol. These observations suggest that compounds with higher binding affinities, particularly Friedelin, may have more potent inhibitory effects on BRCA1, supporting their further exploration as potential agents in breast cancer treatment.

Plumbagin, a bioactive compound primarily found in the Plumbaginaceae family, has been studied for its anticancer properties. It works by inactivating key pathways involved in cancer progression, including Akt/NF-kB, MMP-9, and VEGF[23]. Here, the plumbagin displays five interactions with BRCA1, mainly involving hydrophobic residues like VAL 1654, LEU 1657, and PHE 1662, along with polar residues ASN 1678 and LYS 1702. This suggests a moderate affinity for BRCA1 through both hydrophobic and electrostatic interactions, which may stabilise binding. While Plumbagin's lower interaction count compared to other compounds implies limited binding efficacy, its selective interactions with critical residues suggest that it may contribute to targeted BRCA1 inhibition.

Tinosporide engages in eight interactions with BRCA1, including residues VAL 1654, SER 1655, and LEU 1662. The presence of both hydrophobic (VAL, LEU) and polar (SER, PRO) residues in its binding profile suggests strong binding stability. Its extensive binding, particularly with residues like PHE 1662 and LYS 1702, indicates a high binding affinity that could enhance its potential to disrupt BRCA1-related pathways. This compound's diverse residue interactions imply a balanced and stable binding orientation, making it a promising candidate for further evaluation in breast cancer therapies focused on BRCA1 inhibition.

Curcumin exhibits eight interactions with BRCA1 residues, similar to Tinosporide, including VAL 1654, SER 1655, and LEU 1662. This compound is known

for its broad anti-cancer properties, and its ability to interact with both hydrophobic and polar residues may enhance binding affinity with BRCA1. Curcumin's interaction with residues like THR 1700 and LYS 1702, critical in BRCA1 stability, implies potential effectiveness in interfering with BRCA1 function. This dual-residue binding profile, balancing stability and versatility, supports further exploration of Curcumin as a BRCA1-targeted therapeutic with possible synergistic benefits when combined with other anti-cancer agents.

Picein shows eight interactions with BRCA1, binding to residues such as VAL 1654, SER 1655, and LEU 1662, similar to Curcumin and Tinosporide. This binding profile indicates strong binding potential and a stable orientation within the BRCA1 active site. Notably, Picein's interactions with key residues like THR 1700 and LYS 1702 suggest that it may disrupt BRCA1 activity. This combination of hydrophobic and polar residue engagement indicates that Picein could stabilise its binding and potentially inhibit BRCA1 function. Given its binding properties, Picein could be an effective option for further preclinical testing as a BRCA1-inhibiting compound.

Friedelin has seven interactions with BRCA1, involving residues VAL 1654, SER 1655, and PHE 1662, indicating moderate binding affinity. Friedelin's interactions cover both hydrophobic and polar sites, allowing for stability within the BRCA1 active site. However, its interaction profile is less comprehensive than other compounds like Tinosporide or Curcumin, which may reduce its potential binding efficacy. Nevertheless, Friedelin's binding with key residues, such as LYS 1702, implies it could still play a role in disrupting BRCA1 function.

With only four interactions, β-Pinene demonstrates a limited interaction profile, binding to residues VAL 1654, LEU 1657, PHE 1662, and LYS 1702. This suggests a comparatively lower binding affinity with BRCA1 due to fewer points of contact. However, its selective interaction with both hydrophobic and polar residues may still provide some stability in binding. Although it may not be as potent as other compounds, β-Pinene could still be of interest in combinatorial treatments where lower-affinity ligands contribute to a broader therapeutic effect in BRCA1 inhibition.

Bhilawanol A has the highest number of interactions with BRCA1, totalling nine, involving residues like SER 1655, LEU 1659, and PHE 1662. This extensive binding profile suggests a strong and stable affinity for BRCA1, potentially making Bhilawanol A a highly effective compound in targeting BRCA1 pathways. Its binding with critical hydrophobic and polar residues, including THR 1700 and LYS 1702, implies substantial potential to interfere with BRCA1 stability and function.

Kaempferol binds BRCA1 with five interactions, primarily involving SER 1655, LEU 1702, and LYS 1702, indicating moderate binding potential. While not as extensive as compounds like Tinosporide or Bhilawanol A, Kaempferol's selective interactions with key residues suggest that it could inhibit BRCA1 function with a focus on specific active sites. Kaempferol's moderate binding profile supports its consideration as a secondary agent that can be paired with higher-affinity compounds for optimal BRCA1 inhibition.

Hinokiflavone displays six interactions, binding residues such as SER 1655, LEU 1702, and LYS 1702, suggesting moderate binding stability. Although Hinokiflavone's binding profile is slightly less extensive than that of Bhilawanol A, its interaction with hydrophobic and polar residues may offer a stable orientation within the BRCA1 protein. Given its specific interaction pattern, Hinokiflavone could be a valuable candidate in multi-compound approaches to achieve enhanced BRCA1 pathway targeting.

The interactions of these natural compounds with the BRCA1 protein suggest potential roles in the prevention and treatment of breast cancer by targeting BRCA1 pathways[23-25].

The varied binding profiles, including both hydrophobic and polar interactions, indicate that these compounds may stabilise their binding to BRCA1, potentially disrupting its function. Since BRCA1 is a critical protein, it disrupts DNA damage response, transcriptional control, cell growth and apoptosis, particularly in breast cancer cells that rely on BRCA1 for survival[26].

By binding to specific residues in BRCA1, compounds like Bhilawanol A, Tinosporide, and Curcumin show promise in preventing cancer cells from repairing DNA damage, leading to apoptosis and reducing cancer cell proliferation.

In the context of cancer prevention, these compounds offer significant potential as adjuncts to standard therapies, especially in individuals with a genetic predisposition to breast cancer due to BRCA1 mutations[27].

Compounds with strong binding affinities, such as Bhilawanol A and Tinosporide, may help inhibit BRCA1-mediated repair processes even at the early stages of carcinogenesis, potentially reducing the risk of tumour development. Regular administration of such compounds could theoretically lower the likelihood of cancerous cells surviving DNA damage, acting as a preventive measure.

The anti-inflammatory and antioxidant properties associated with compounds like Curcumin and Kaempferol further add to their potential, as chronic inflammation and oxidative stress are linked to cancer progression[28,29].

For treatment applications, these compounds might also enhance the efficacy of existing chemotherapy or radiotherapy by sensitising cancer cells to treatment[30]. By inhibiting BRCA1, these compounds could make cancer cells more susceptible to DNA-damaging treatments, as they would lack the BRCA1-mediated repair mechanisms. This synergy could allow for lower doses of chemotherapy, potentially reducing side effects for patients while maintaining or even enhancing therapeutic efficacy[31].

Furthermore, the ability of compounds like Hinokiflavone and Picein to target specific BRCA1 residues highlights their potential as candidates in personalised medicine, where treatment could be tailored based on a patient's BRCA1 status. Overall, these interactions illustrate the promising role of these natural compounds as complementary agents in both cancer prevention and treatment.

5. Conclusion

The molecular docking analysis identifies several natural compounds, including Bhilawanol A, Tinosporide, and Curcumin, as promising BRCA1 inhibitors. These compounds exhibit strong binding affinities, suggesting their potential roles in breast cancer prevention and treatment by disrupting DNA damage response, transcriptional regulation, cell growth, and apoptosis management. These compounds interact with critical BRCA1 residues, potentially disrupting DNA repair in cancer cells, making them more susceptible to apoptosis. With their additional antioxidant and anti-inflammatory benefits, compounds like *Curcumin* and *Kaempferol* could offer preventive effects, especially for individuals with BRCA1 mutations. Although these findings are promising, further *in vitro* and *in vivo* studies are needed to confirm their efficacy and bioavailability, paving the way for their use as complementary agents in targeted cancer therapies.

6. Acknowledgements

I express my sincere thanks to the library unit of the National Institute of Siddha, Ministry of Ayush, Government of India, Chennai.

7. Author Contribution

Karthik Nagarajan - Conceptualisation, Methodology, Study plan, Data Collection, Writing the original draft. Bharath Christian C.B.S and Siva A. - Data analysis, Validation, Formal analysis. Merish S. - Data interpretation, Final article drafting, grammar and spelling correction. All authors have read and agreed to the published version of the manuscript.

8. References

1. Arnold M, Morgan E, Rumgay H, Mafra A, Singh D, Laversanne M, *et al.* Current and future burden of breast cancer: Global statistics for 2020 and 2040. Breast. 2022; 66:15-23. https://doi.org/10.1016/j.breast.2022.08.010 PMid:36084384 PMCid:PMC9465273
2. Gupta A, Shridhar K, Dhillon PK. A review of breast cancer awareness among women in India: Cancer literate or awareness deficit? Eur J Cancer. 2015; 51(14):2058-2066. https://doi.org/10.1016/j.ejca.2015.07.008 PMid:26232859 PMCid:PMC4571924
3. Lin SR, Chang CH, Hsu CF, Tsai MJ, Cheng H, Leong MK, *et al.* Natural compounds as potential adjuvants to cancer therapy: Preclinical evidence. Br J Pharmacol. 2020; 177(6):1409-1423. https://doi.org/10.1111/bph.14816 PMid:31368509 PMCid:PMC7056458
4. Sohel M, Aktar S, Biswas P, Amin MA, Hossain MA, Ahmed N, *et al.* Exploring the anti-cancer potential of dietary phytochemicals for the patients with breast cancer: A comprehensive review. Cancer Med. 2023; 12(13):14556-14583. https://doi.org/10.1002/cam4.5984 PMid:37132286 PMCid:PMC10358259
5. McGrowder DA, Miller FG, Nwokocha CR, Anderson MS, Wilson-Clarke C, Vaz K, *et al.* Medicinal herbs used in traditional management of breast cancer: Mechanisms of action. Medicines (Basel). 2020; 7(8):47. https://

doi.org/10.3390/medicines7080047 PMid:32823812 PMCid:PMC7460502
6. Muthaliyar KSM. Gunapadam Part -I (Siddha Materia Medica - Medicinalplant division), Department of Indian Medicine and Homeopathy, Chennai - 600 106, II Edition – Reprint; 2006. p. 198-200, 240-242, 381-386, 454-458, 488-490, 627-629, 720-722, 780-782.
7. Petrucelli N, Daly MB, Pal T. BRCA1- and BRCA2-associated hereditary breast and ovarian cancer. In: Adam MP, Feldman J, Mirzaa GM, *et al*., editors. GeneReviews®. Seattle (WA): University of Washington, Seattle; 1993-2024; Available from: https://www.ncbi.nlm.nih.gov/books/NBK1247/
8. Torres PHM, Sodero ACR, Jofily P, Silva-Jr FP. Key topics in molecular docking for drug design. Int J Mol Sci. 2019; 20(18):4574. https://doi.org/10.3390/ijms20184574 PMid:31540192 PMCid:PMC6769580
9. Chehelgerdi M, Chehelgerdi M, Allela OQB, Pecho RDC, Jayasakar N, Rao DP, *et al*. Progressing nanotechnology to improve targeted cancer treatment: overcoming hurdles in its clinical implementation. Mol Cancer. 2023; 22:169. https://doi.org/10.1186/s12943-023-01865-0 PMid:37814270 PMCid:PMC10561438
10. Pillai TVS. Tamil to English agarathi of medicine, chemistry, medicine and allied sciences, Vol V, Department of Indian Medicine and Homeopathy, Chennai; 1977. p. 3411, 3804.
11. Suganthi N. Phytochemical constituents and pharmacological activities of *Vitex negundo* Linn. J Chem Pharm Res. 2016; 8(2):800-807.
12. Lin YM, Anderson H, Flavin MT, Pai YH, Mata-Greenwood E, Pengsuparp T, *et al*. *In vitro* anti-HIV activity of biflavonoids isolated from Rhus succedanea and Garcinia multiflora. J Nat Prod. 1997; 60(9):884-888. https://doi.org/10.1021/np9700275 PMid:9322359
13. Hewlings SJ, Kalman DS. Curcumin: A review of its effects on human health. Foods. 2017; 6(10):92. https://doi.org/10.3390/foods6100092 PMid:29065496 PMCid:PMC5664031
14. Kant K, Walia M, Agnihotri VK, Pathania V, Singh B. Evaluation of antioxidant activity of *Picrorhiza kurroa* (Leaves) extracts. Indian J Pharm Sci. 2013; 75(3):324-329. https://doi.org/10.4103/0250-474X.117438 PMid:24082348 PMCid:PMC3783750
15. Gedam PH, Sampathkumaran PS, Sivasamban MA. Composition of bhilawanol from *Semecarpus anacardium*. Phytochemistry. 1974; 13(2):513-515. https://doi.org/10.1016/S0031-9422(00)91247-0
16. Sharma P, Dwivedee BP, Bisht D, Dash AK, Kumar D. The chemical constituents and diverse pharmacological importance of *Tinospora cordifolia*. Heliyon. 2019; 5(9):e02437. https://doi.org/10.1016/j.heliyon.2019.e02437 PMid:31701036 PMCid:PMC6827274
17. Mishra S, Aeri V, Gaur PK, Jachak SM. Phytochemical, therapeutic, and ethnopharmacological overview for a traditionally important herb: *Boerhavia diffusa* Linn. Biomed Res Int. 2014; 2014:808302. https://doi.org/10.1155/2014/808302 PMid:24949473 PMCid:PMC4053255
18. Jaisi A, Sakunphueak A, Panichayupakarn P. Increased production of plumbagin in *Plumbago indica* root cultures by gamma ray irradiation. Pharm Biol. 2013; 51(8):1047-1051. https://doi.org/10.3109/13880209.2013.775163 PMid:23746284
19. Kollman PA. Docking and scoring in a virtual screening protocol: A study of the human cyclin-dependent kinase 2 inhibitors. J Med Chem. 2000; 43(16):3070-3082.
20. Morris GM, Goodsell DS, Halliday SM, Huey R, Hart WE, Belew RK. Automated docking using a Lamarckian genetic algorithm and an empirical binding free energy function. J Comput Chem. 1998; 19(14):1639-1662. https://doi.org/10.1002/(SICI)1096-987X(19981115)19:14<1639::AID-JCC10>3.0.CO;2-B
21. Solis FJ, Wets RJ. Minimization of functions of several variables with an application to the traveling salesman problem. Math Program. 1981; 21(1):199-215.
22. Jayaraman S, Veeraraghavan V, Sreekandan RN, Mohan SK, Suga SSD, Kamaraj D, *et al*. Molecular docking analysis of the BRCA1 protein with compounds from *Justica adhatoda* L. Bioinformation. 2020; 16(11):888-892. https://doi.org/10.6026/97320630016888 PMid:34803264 PMCid:PMC8573473
23. Sadeghi F, Asgari M, Matloubi M, Ranjbar M, Yousefi NK, Azari T, *et al*. Molecular contribution of BRCA1 and BRCA2 to genome instability in breast cancer patients: Review of radiosensitivity assays. Biol Proced Online. 2020; 22:23. https://doi.org/10.1186/s12575-020-00133-5 PMid:33013205 PMCid:PMC7528506
24. Roy A. Plumbagin: A potential anti-cancer compound. Mini Rev Med Chem. 2021; 21(6):731-737. https://doi.org/10.2174/1389557520666201116144421 PMid:33200707
25. Haim W. BRCA1: An endocrine and metabolic regulator. Front Endocrinol. 2022; 13:844575. https://doi.org/10.3389/fendo.2022.844575 PMid:35432218 PMCid:PMC9009035
26. Roy R, Chun J, Powell SN. BRCA1 and BRCA2: Different roles in a common pathway of genome protection. Nat Rev Cancer. 2011; 12(1):68-78. https://doi.org/10.1038/nrc3181 PMid:22193408 PMCid:PMC4972490
27. Neiger HE, Siegler EL, Shi Y. Breast cancer predisposition genes and synthetic lethality. Int J Mol Sci. 2021; 22(11):5614. https://doi.org/10.3390/ijms22115614 PMid:34070674 PMCid:PMC8198377
28. Menon VP, Sudheer AR. Antioxidant and anti-inflammatory properties of curcumin. Adv Exp Med Biol. 2007; 595:105-125. https://doi.org/10.1007/978-0-387-46401-5_3 PMid:17569207
29. Qattan MY, Khan MI, Alharbi SH, Verma AK, Al-Saeed FA, Abduallah AM, *et al*. Therapeutic importance of kaempferol

in the treatment of cancer through the modulation of cell signalling pathways. Molecules. 2022; 27(24):8864. https://doi.org/10.3390/molecules27248864 PMid:36557997 PMCid:PMC9788613

30. Nisar S, Masoodi T, Prabhu KS, Kuttikrishnan S, Zarif L, Khatoon S, *et al.* Natural products as chemo-radiation therapy sensitizers in cancers. Biomed Pharmacol. 2022; 154:113610. https://doi.org/10.1016/j.biopha.2022.113610 PMid:36030591
31. Patel V, Casimiro S, Abreu C, Barroso T, de Sousa RT, Torres S, *et al.* DNA damage targeted therapy for advanced breast cancer. Explor Target Antitumor Ther. 2024; 5(3):678-698. https://doi.org/10.37349/etat.2024.00241 PMid:38966174 PMCid:PMC11220312