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COMPUTATIONAL ANALYSIS OF BIOMOLECULES OF *MURRAYA KOENIGII* AS A POTENT ANTICANCER AGENT

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Abstract:
We have used a computational approach in this study to predict and identify the potentially harmful protein in curry leaves. The ligand was chosen from the PubChem database, while the target was chosen from the PDB. We have added the polar hydrogen group and eliminated the water molecules in order to prepare the target. It was also possible to find the twisting root where docking can be done to get the ligand ready. There were PDBQT files for both the target and the ligand. For the purpose of molecular docking against the three anticancer medications that we acquired from PubMed, we have taken into consideration three breast malignant proteins: HER2 (PDB ID-1N8Z), oestradiol (PDB ID-3HB5), and NUDT-5 (PDB ID-5NQR). A well-known sex hormone that causes breast cancer in women is oestradiol. Breast cancer cells proliferate and differentiate when the HER2 protein is activated improperly. Breast cancer cell proliferation and gene regulation are significantly impacted by NUDT 5. After using Auto Dock to dock molecules, Vina Mahanine and Pyrayafoline D had the least amount of binding energy with the breast cancer protein.

Although we are primarily concerned with breast cancer, the carcinogenic protein under examination is also accountable for other forms of cancer. The study says that Murraya koenigii might be a good place to get bioactive chemicals that can help avoid cancer. The idea of making anticancer drugs from higher eukaryotic plants seems like it would be good for people because it could lead to new treatments that will help cancer patients right now.

Keywords: Anticancer, Bioactive, Docking, Drugs, Repositories,Signaling

Introduction:

With the growth of bioinformatics, computer methods used to guess how the surfaces of two molecules with biological origins will interact have become a lot more common (Wadanambi, et al., 2023). The time-consuming task of continually examining interactions between various molecules is made simpler by bioinformatics, which also yields the most ideal interface interaction (Surana, et al., 2021). The best strategy is provided by two parallel approaches: prediction and experimentation. The result shows great promise and a high degree of accuracy (Surana, et al., 2021). Because it eliminates undesirable compounds from compound libraries and offers a time- and money-efficient platform, the virtual screening method is frequently used (Saleem, et al., 2022).

Globally, breast cancer is a leading cause of death for women. In 2020, the proportion of newly diagnosed cases with breast cancer was 12.3% (Pandya, et al., 2020).In 2020, 2.26 million instances of breast cancer were reported worldwide. In India, women who receive a fresh diagnosis of breast cancer have a 50% death rate (Pathan, et al., 2023). Three commonly observed proteins that are linked to breast cancer are NUDT-5, HER2, and estrogen. Herbal medicine is a better option because conventional and mainstream medication can have a lot of side effects and be expensive (Surana, et al., 2021). There has also been a huge rise in the desire for drugs that come from natural sources. Asia is home to *Murraya koenigiiL.*, a member of the Rutaceae family that is commonly referred to as "curry leaf" in English. India used to grow curry leaf trees for their fragrant leaves (Karthic, et al., 2022). It gradually became more and more well-known in many Asian cuisines because of its remarkable and distinctive flavor. Numerous health advantages, such as anti-inflammatory, anti-cancer, anti-fungal, antibacterial, and antioxidant qualities, have been reported (Mondal, et al., 2022).

It is the study of how two or more molecular structures, like a drug and an enzyme or protein, can join to make a stable complex that uses the least amount of energy (Kaloni, et al., 2020). A molecular modeling method called docking is used to predict how tiny molecules (ligands) and proteins (enzymes) will interact (Iman, et al., 2016). Curry leaves have been shown to contain carbazole alkaloids, such as mahanine, pyrafoline-D, mahanimbine, and grinimbine, which are known to have antioxidant and anti-cancer qualities. Curry leaves are high in minerals, iron, copper, magnesium, fiber, and carbohydrates (Ahire, et al., 2020). It also contains a lot of vitamins, including as vitamin B, vitamin E, vitamin A, vitamin C, and nicotinic acid. Iron and folic acid are prevalent in curry leaves (Mathivadani, et al., 2020). The main way that folic acid helps the body absorb and move iron is through its transportation. Recently, Syam et al. found that this plant's carbazole alkaloid, girinimbine, efficiently inhibited the growth and initiated apoptosis in hepg2 cells, which are human hepatocellular carcinomas (Gaikwad, et al., 2023).

Several computer, in vivo, and in vitro methods were used to test how well carbazole chemicals could fight cancer (Keservani, et al., 2020). Docking is one of these methods that has been used a lot in the development of medicines for breast cancer. Several researchers have studied the function of the carbazole alkaloids found in curry leaves in great detail over time (Mondal, et al., 2022). Their potential as anti-cancer drugs is justified by their inhibitory actions. Mahanine (PubChem CID:36689305), Girinimbine (PubChem CID:96943), and Pyrafoline-D (PubChem CID:375148) were the selected alkaloids (Suranaand Mahajan, 2022).

There is currently little research on curry leaves' potential as a breast cancer treatment. This makes the study important since it seeks to shed light on the efficacy of herbal medicine in the treatment of breast cancer (Ghasemzadeh, et al., 2014).

Curry-leaf trees, also known as *Murraya koenigii*, are mainly grown in the southern parts of India in the Indian subcontinent. Perennial trees are highly valued for their ability to provide taste to a variety of food dishes (Maheswari and Sankar, 2024). They also have a wide range of medicinal characteristics, including as antibacterial, antifungal, anticancer, and anti-inflammatory activities. This plant is native to Asia and a member of the Rutaceae family (Pakrashy, et al., 2022).

Material and Method:

Some biological materials that were used in this study were PubChem, ZINC, and RCSB-PDB (Protein Data Bank). Other tools that were used were Avogadro, PyMOL, Autodock vina, Discovery studio visualizer, and Open Babel GUI (Ahmadipour, et al., 2015). The Protein Data Bank (PDB), which was started in 1971 at Brookhaven National Laboratories

(BNL), is the only database in the world that has structural information about living macromolecules. It has information about the macromolecules' structures found by X-ray crystallography, NMR techniques, and other means (Kamalidehghan, et al., 2018). It lets you see the 3D structures of big living molecules like DNA, RNA, and proteins. Autodock Vina is not the same as Autodock 4.0. It is a better version of Autodock 4.0 that works well and accurately (El-Shiekh, et al., 2023). These days, the Scripps Research Institute in Florida, USA, takes care of both Autodock 4.0 and Autodock vina. Multiple drugs have been found easier with the help of AutoDock (Mohan, et al., 2013). A program called Discovery Studio Visualizer is used to simulate small molecular systems and examine large molecular systems. It has tools for docking receptors and ligands. PyMOL can be used to see biomolecules like proteins. The main job of Open Babel GUI is to change between different types of chemical files (Iman, et al., 2015).

Target selection:

Next, we inputted the name of the protein that was subjected to docking, such as Oestradiol or its PDB ID: 3HB5. We choose the option 'Download file from the drop-down menu'. Next, we selected the PDB File (text) and proceeded to download it. Subsequently, we accessed this text file and eliminated all the heteroatoms. The subsequent stage entailed the removal of X chains due to their identical nature, as the ligand has the capability to bind to any of these chains (Parkhe, et al., 2023).

Ligand Preparation:

Next, we inputted the name of the protein that was subjected to docking, such as Oestradiol or its pdb id: 3HB5. We choose the option 'Download file from the drop-down menu'. Next, we selected the PDB File (text) and proceeded to download it. Subsequently, we accessed this text file and eliminated all the heteroatoms. The subsequent stage entailed the removal of X chains due to their identical nature, as the ligand has the capability to bind to any of these chains (Patel, et al., 2019).

Preparation of Protein:

PDBQT files were produced by utilizing the Autodock tools application, which was acquired from MGL tools, to build the protein and ligand. Due to its unwanted interaction with other molecules (ligands) of interest, detach the water molecule from the target receptor. To improve the stability of the protein, add a polar hydrogen group. Add the Kollhman expenses. Determine the protein's Gasteiger charges. Transform the document into a pdbqt file. To aid in site-specific docking, acquire the active sites X, Y, and Z measurements. Now we made a text file with all the information about the PDBQT files of the protein, its ligand, and the SBD site sphere's properties (Sharma, et al., 2018).

Molecular Docking using Autodockvina:

In the following method, set the directory where we placed the prepared file for auto-dock compilation.

The use of Discovery Studio Visualizer, analyze molecular docking to learn about the different ligand conformations on the protein:

Look at the different ways the ligand and protein connect and interact. Find out how far apart the ligand and the amino acid groups are. Find out about the different kinds of bonds that ligands and proteins use to connect. Find the best position for the ligand as it reacts with the protein. Look at how the protein and molecule interact with each other in two dimensions. Find the amino acid residues that help the ligand and protein form a link (Keservani, et al., 2019).

Results and Discussions:

These compounds have demonstrated potent anti-cancer properties. Three marker compounds were employed for conducting docking simulations on proteins associated with breast cancer. The protein-ligand relationship was determined for the docked ligands with the lowest binding energy. Table 1 presents This table displays the binding energy between proteins and their ligands.

Table 1: This table illustrates the affinity between proteins and their corresponding ligands, as measured by their binding energy

Sr. No	Compounds	Protein	Binding Energy (Kcal/mol)
1.	Girinimbine (Pubchem ID: 96943)	Oestradiol (PDB ID: 3H5B)	-9.1
		NUDT5 (PDB ID: 5NQR)	-9.5
		HER2 (PDB ID: 1N8Z)	-8.6
2.	Mahainine (Pubchem ID: 36689305)	Oestradiol (PDB ID: 3H5B)	-9.9
		NUDT5 (PDB ID: 5NQR)	-8.2
		HER2 (PDB ID: 1N8Z)	-9.6
3.	Pyrayafoline (Pubchem ID: 375148)	Oestradiol (PDB ID: 3H5B)	-9.9
		NUDT5 (PDB ID: 5NQR)	-7.3
		HER2 (PDB ID: 1N8Z)	-9.2

Ligand Protein interaction diagram of chemical compounds:

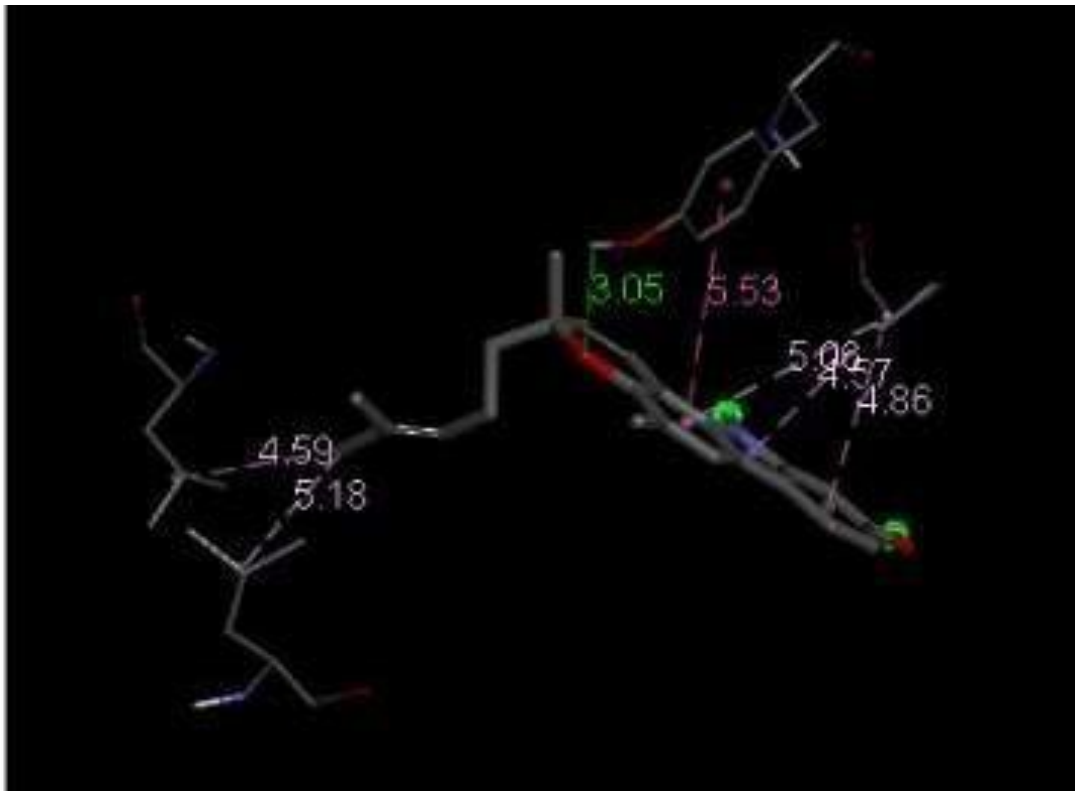

Figure 1: The lengths of the bonds between the mahanine molecule and the amino acid residues of the protein HER-2 are shown in this picture.

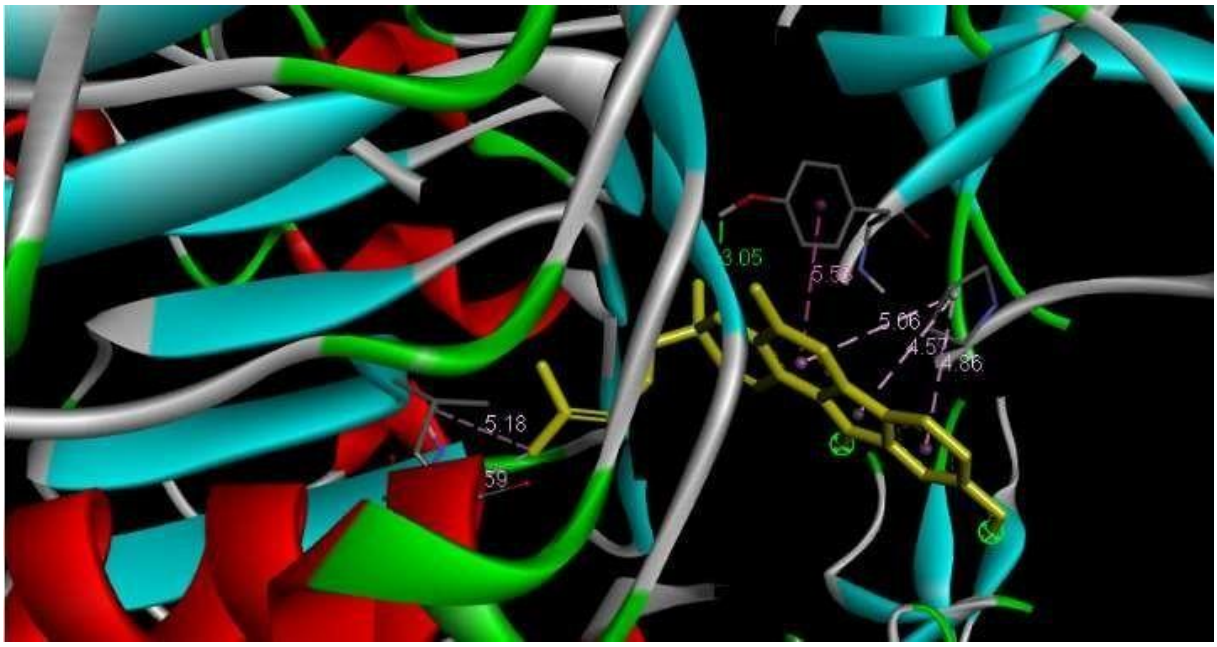

Figure 2: The illustration shows how Mahanine and HER-2 protein work together.

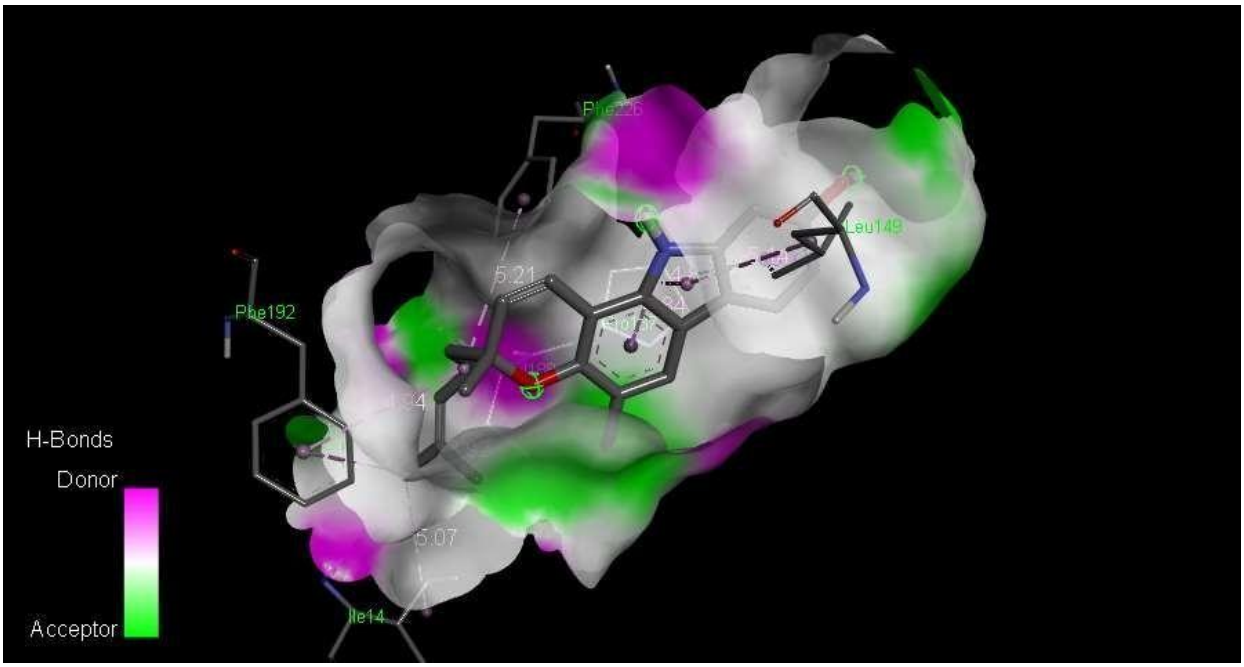

Figure3: shows how the H-Bonds of Oestradiol and Girinimbine look when they are docked.

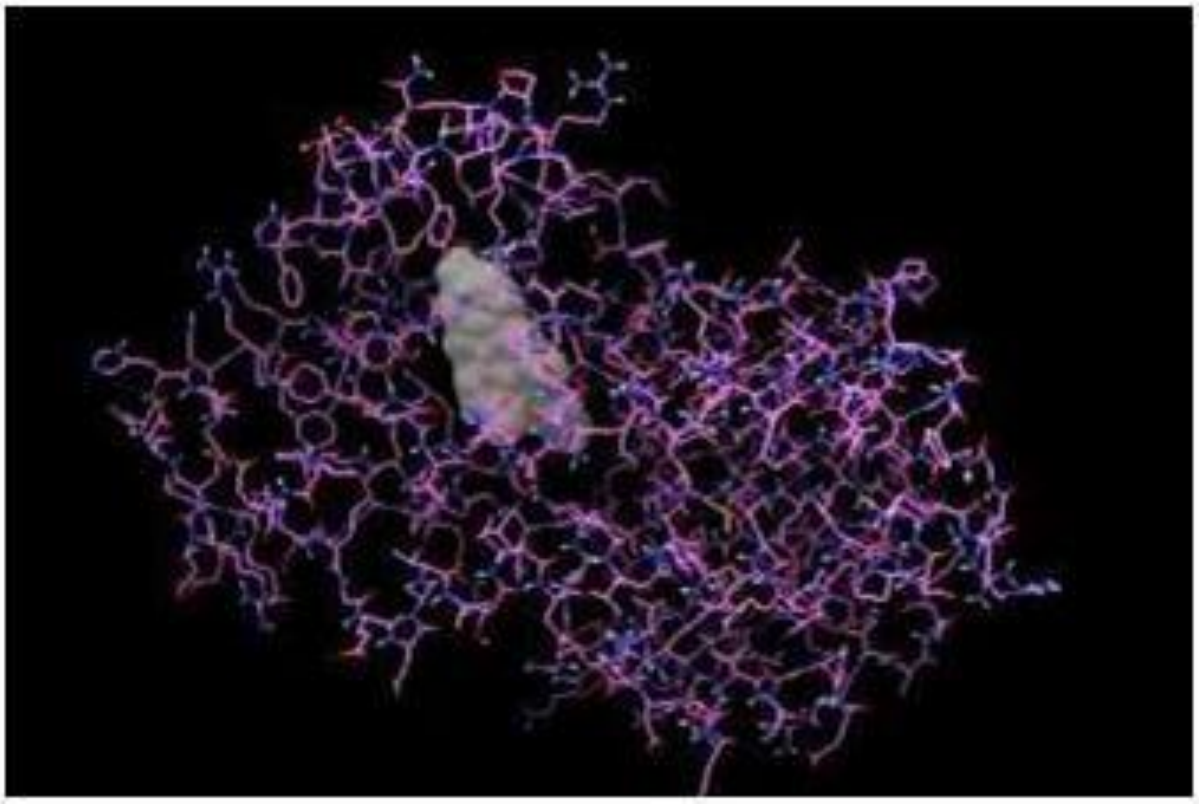

Figure4: shows how Oestradiol and Girinimbine fit together molecularly.

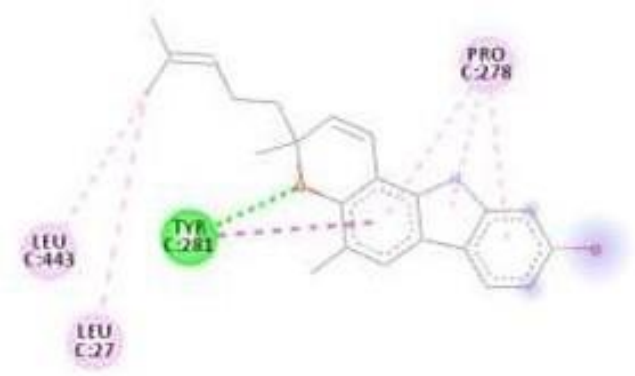

Figure 5: shows how Ligand Mahanine and HER2 protein combine in two dimensions.

In this study, we used computers to learn about and predict the protein in curry leaves that might be dangerous. PubChem was used to find the ligand, and the PDB was used to find the target. In order to get the target ready, we added the polar hydrogen group and got rid of the water molecules. It was also possible to find the twisting root where docking can be done to get the ligand ready. After using Auto Dock to dock molecules, Vina Mahanine and Pyrayafoline D had the least amount of binding energy with the breast cancer protein. The carcinogenic protein we are looking at is also responsible for other types of cancer, even though breast cancer is our main worry. The study says that *Murraya koenigii* might be a good place to get bioactive chemicals that can help avoid cancer. The idea of making anticancer drugs from higher eukaryotic plants seems like it would be good for people because it could lead to new treatments that will help cancer patients right now.

Conclusion:

Pyrayafoline D and mahanine had the most favorable binding energies with the proteins implicated in breast cancer. The current study's findings suggest that *Murraya koenigii* could serve as a valuable reservoir of bioactive compounds for the purpose of cancer prevention. The potential for developing anticancer drugs from higher plants appears promising since it enables the production of innovative pharmaceuticals, which are essential in today's society. Subsequent investigations can analyze the protein-ligand complex using molecular modeling and molecular dynamic simulations. In addition, these investigations can evaluate the ADME/T features of the substances in a laboratory setting and forward the inquiry towards clinical trials. In the future, research findings could be utilized in clinical trials to assess the

effectiveness of a treatment and benefit society. This would result in a reduction in both the time and cost associated with the drug discovery process.

Declarations:

Ethics approval and consent to participate:

Not applicable.

Consent for publication:

All the authors approved the manuscript for publication.

Availability of data and material:

All required data is available.

Competing interests:

All authors declare no competing interests.

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