



Insilico Screening of Bioactive Compounds Derived from Catharanthus Roseus against Anticancer Activity

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ABSTRACT

One of the most prevalent diseases in the world is cancer. In order to stop cancer from growing, developing, and spreading, cancer treatments still need to be carefully planned and targeted. Radiation, cellular stress, and cytotoxic medications are some of the triggers that activate the mitochondrial intrinsic apoptotic pathway. Native to the Mediterranean region, Vinca roseus is a blooming perennial plant that is primarily found in the northern hemisphere. They are native to tropical regions in South Asia. There are numerous uses for the Madagascar plant, Catharanthus roseus L. The 3D crystallographic structure of the Anti-cancer receptor (PDB ID-2KCE & 2HBS) was obtained from the Protein Data Bank and utilized as a protein target for in-silico experiments. Molecular docking was performed using Auto Dock 4 and Auto Dock Vina. A blind docking approach was employed to encompass all possible ligand binding sites. The binding free energy (kcal/mol) was utilized to calculate the binding affinity. This study reveals that Catharanthus roseus, a polyphenolic compound, may possess antioxidant, antibacterial, antifungal, & anticancer activity against the Anti-cancer receptor responsible for cancer disease, as predicted in silico. Molecular docking data suggest that Vincristine (-7.1 & -9.5 Kcal/mol) and Vindesine (-9.1 & -10.5 Kcal/mol) have greater activity than Vincristine. They have good Anticancer properties. & The best protein was found to be 2HBS for Anti-cancer activity.

keywords: Catharanthus roseus, Anti-cancer, Vincristine, Vindesine, 2HBS.

1. INTRODUCTION

Cancer is one of the world's most common illnesses. Cancer treatments must still be properly devised and targeted to prevent cancer growth, development, and spread. Radiation, cellular stress, and cytotoxic medications are some of the triggers that activate the mitochondrial intrinsic apoptotic pathway. These signals disrupt mitochondrial membrane proteins such as Bcl-2. The Bcl2 protein family contains both pro- and anti-apoptotic proteins. The proapoptotic Bcl-2 family has two subfamilies: BH123 and BH3-only proteins. The BH123 subfamily is mostly composed of BAK and BAX, whereas BH3-only proteins include BID, BIM, BAD, BIK, PUMA, and NOXA. In the absence of apoptotic triggers, the antiapoptotic Bcl-2 protein interacts with the proapoptotic BH123 proteins, BAK and BAX, on the outer membrane. Cancer is clearly a severe, perhaps deadly ailment. Cancer Treatment¹. It was responsible for around 10 million deaths worldwide in 2020, with a significant share of new cases related to stomach cancer and breast cancer, lung cancer, colon and rectum cancer, prostate cancer, skin cancer, and other cancers. These days, chemotherapy and radiation therapy are used in addition to chemically generated medications to treat cancer; chemotherapy has a negative effect on healthy cells.

1.1 Types of cancer:

Cancer is a multifaceted disease that can impact various bodily parts. The following are some of the most common types of cancer: Breast Cancer, Lung cancer, Prostate cancer, Colorectal cancer, Skin cancer (both melanoma and non-melanoma), Bladder Cancer, Leukemia, Lymphoma (both Hodgkin and non-Hodgkin lymphoma), Pancreatic cancer, Ovarian cancer, Cervical cancer, Brain cancer, Liver cancer, Kidney Cancer, Thyroid cancer, Esophageal Cancer, Stomach (gastric) cancer, Testicular carcinoma, Bone cancer, Sarcoma (which includes both soft tissue and bone sarcomas).

There are several more forms of cancer, some of which can attack several organs or systems in the body. Each form of cancer has unique features, treatment choices, and prognosis. Early identification and therapy are critical for improving cancer treatment results. There are many medicinal herbs in nature. Catharanthus roseus is one such species.

It is found mostly in tropical regions across the world. *Vinca roseus* is a blooming perennial plant native to the Mediterranean region, mostly found in the northern hemisphere. They are endemic to South Asia and tropical countries. *Catharanthus roseus* L. from Madagascar has a variety of applications. Various names for *Vinca rosea* include Madagascar periwinkle, sparkling eyes, Cape Periwinkle, graveyard plant, old woman, and pink periwinkle. Rose periwinkle myrtle is one of its common names. It is used in a variety of hues for ornamentation.

Vinca alkaloids are chemical sub It is available in a range of hues, including pink, purple, and white.

It's also used for medicinal reasons. stances that are frequently present in plants and consist of carbon, hydrogen, nitrogen, and oxygen. Although the word "alkali" implies alkali, not all compounds have this quality. Alkaloids have physiological effects in addition to being poisonous, which makes them potentially useful as medicines. Vinca alkaloids are among the oldest plant alkaloids used to treat cancer. These nitrogenous bases, derived from the pink periwinkle plant *Catharanthus roseus* G., are either naturally occurring or semi-synthetic. Vincristine and vindesine have an anticancer impact because they bind to intracellular tubulin, which prevents DNA repair and RNA synthesis by blocking the enzyme DNA-dependent RNA polymerase. The biological functions that include protein synthesis, respiration, glycolysis, and nucleic acid^{7 8}.

Catharanthus roseus' leaves contain copious alkaloids vincristine and vinblastine, which suppress tumour development and contribute to its anticancer qualities. Vinblastine and vincristine were among of the earliest natural drugs⁹. The active component of *Catharanthus Roseus* varies based on its cultivation and harvest location. The Vietnamese cultivar has 0.1-0.2 percent total alkaloids, comparable to *Catharanthus Roseus* from Madagascar. Roots had more alkaloids (0.7-2.4%) than stems (0.46%) and leaves (0.37-1.15%). Several nations, including Korea, Japan, and China, are experimenting with this plant for medicinal purposes^{10 11}. *Vinca* plant contains vinblastine and vincristine, which are used in chemotherapy to treat various cancers. These are derived from the alkaloids catharanthine and vindoline, respectively¹². Vinorelbine agents are semi- synthetic chemotherapy drugs used to treat non-small cell lung cancers *Vinca* alkaloids vindoline and catharanthine, as well as luisine can be used to create these compounds¹³.

2. MATERIAL AND METHOD

2.1 Derivatives

There are many medicinal herbs in nature. *Catharanthus roseus* is one such species. It is found mostly in tropical regions across the world. *Vinca roseus* is a blooming perennial plant native to the Mediterranean region, mostly found in the northern hemisphere. They are endemic to South Asia and tropical countries. It contains important pharmaceutical, industrial, and extreme nutritional components. This study selected Vincristine, Vindesine, etc. the structure of these derivatives as 3D conformers in SDF format were received from the PubChem Database, also called the Library of Drug Molecule¹⁴.

Phytochemicals in *Vinca rosea*, including Vincristine, Vindesine, Vinblastine were isolated and analysed using PubChem. Pub Chem pro provides open access to knowledge about chemical compounds and their biological roles. The Substance database contains chemical information provided by individual data contributors to PubChem, whereas the compound database includes unique chemical structures retrieved from the Substance database¹⁵.

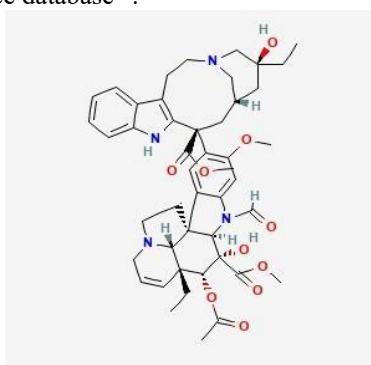


Figure 1: Vincristine

Vincristine is a vinca alkaloid with the chemical formula $C_{46}H_{56}N_4O_{10}$ discovered in the Madagascar periwinkle *Catharanthus roseus*. It is used as a chemotherapeutic medication to treat leukemia, lymphoma, myeloma, breast cancer, and head and neck cancer. It functions as a tubulin modulator, a microtubule-destabilizing agent, a plant metabolite, an anti- cancer agent, and a medication. It is a methyl ester, an acetate ester, a tertiary alcohol, a formamide, an organic heteropentacyclic, an organic heterotetracyclic, a tertiary amino, and a vinca alkaloid. It is the conjugate base of vincristine (2+). It is derived from the hydride of vincalukoblastine.

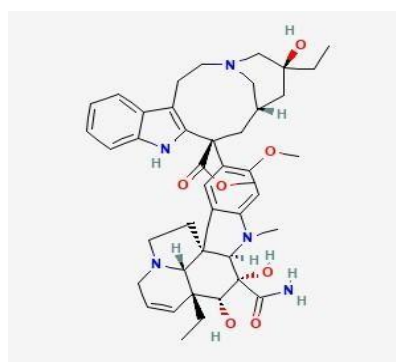


Figure 2: Vindesine

Vindesine is classified as a vinca alkaloid, methyl ester, organic heterotetracyclic compound, organic heteropentacyclic compound, tertiary alcohol, tertiary amino compound, and primary carboxamide. It serves as an antineoplastic agent.

It is functionally similar to vincalukoblastine.

2.2 Protein Preparation

3D crystallographic structure of Anti-cancer receptors (PDB ID- 2KCE, 2HBS) were received from Protein Data Bank and used as protein target for in-silico studies. subsequently, the protein's format was transformed to .pdb. pdbqt, 'q' denotes the addition of Kollman charges, and 't' signifies the removal of hetero atom. The Pymol software was employed to visualize the protein structure, serving as the receptor for the investigated molecule. Utilizing this tool, cartoon structures of the protein were generated¹⁶.

Table 1: Proteins & PDB ID

Sr.no	Protein	PDB ID
1	Cloacimonas Acidaminovorans	2KCE
2	Doxyhemoglobin S	2HBS

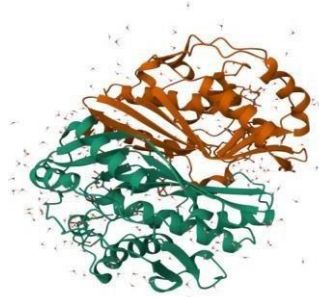


Figure 3: 2KCE

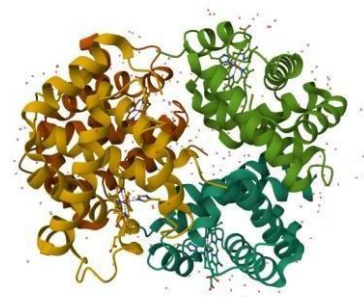


Figure 4: 2HBS

2.3 Docking Procedure

The PyRx tool, a comprehensive open-source platform that unifies Autodock 4, Autodock Vina, and Auto Wizard into a single environment, was used for docking investigations. The capacity of PyRx to effectively expedite the docking process for several ligands makes it stand out. PyRx does away with the necessity for prompt command knowledge, in contrast to Autodock Vina. PyRx also includes Open Babel, a useful tool for optimising ligand molecule SDF files and converting them to the appropriate.pdb format that is compatible with the receptor molecule. Moreover, PyRx provides the ease of doing energy minimization for every chosen ligand at the same time, which helps find stable molecular conformations¹⁷.

3. RESULT & DISCUSSION

3.1 Ramchandran plot

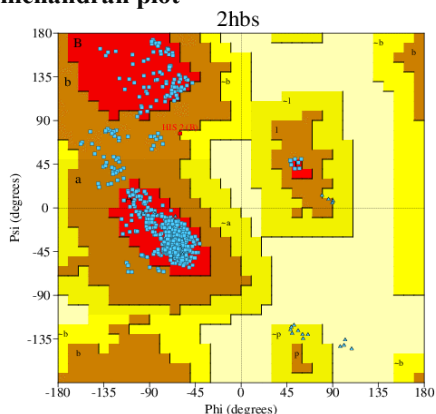


Figure 5: 2HBS

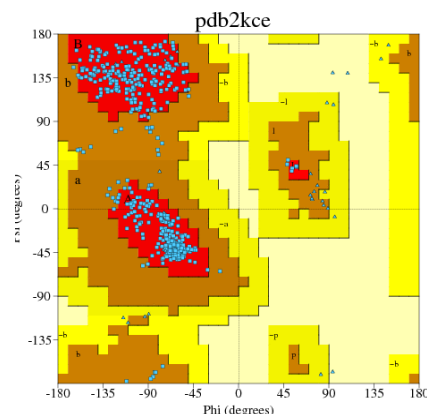


Figure 6: 2KCE

3.2 Ligand binding preparation



Figure 7:- 2KCE & Vincristine

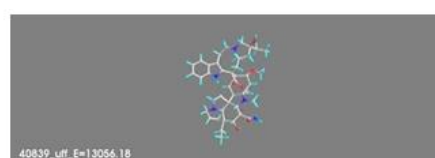


Figure 8:- 2KCE & Vindesine

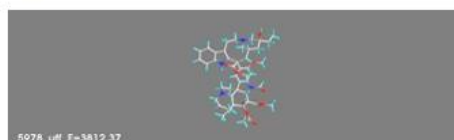


Figure 9:- 2HBS & Vincristine

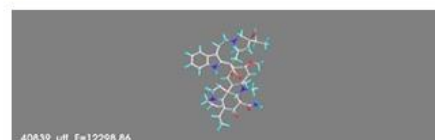


Figure 10:- 2HBS & Vindesine

3.3 Parameter gride and docking simulation

The molecular docking was performed to identify receptor ligand interaction. The Vincristine, Vindesine were used for docking with Catharanthus roseus. The grid map was created using AutoGrid and a grid box with a docking box. A grid box measuring $60 \times 60 \times 60 \text{ \AA}$ was used to establish the docking coordinates for TR α and $60 \times 60 \times 60 \text{ \AA}$ for TR β . Box spacing measured 0.375 \AA . Lastly, Auto Dock was executed with a maximum of 27,000 generations and 10,000 retries. Local search in combination with a genetic algorithm was used to determine the docking options. The stimulation for each Catharanthus roseus was made up of 1000 docking runs with the default AutoDock settings.

Using AutoDock 4 and AutoDock Vina as docking tools, molecular docking was carried out. A blind docking technique was used to include every potential binding site for the ligands. The binding free energy (kcal/mol) was used to express the binding affinity¹⁸.

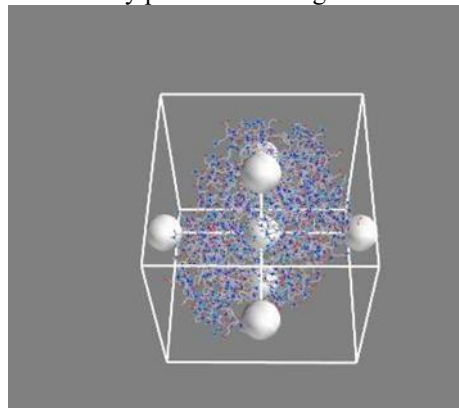


Figure 11: 2KCE & Vincristine

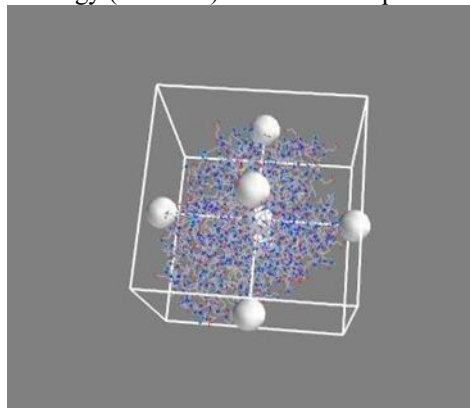


Figure 12: 2KCE & Vindesine

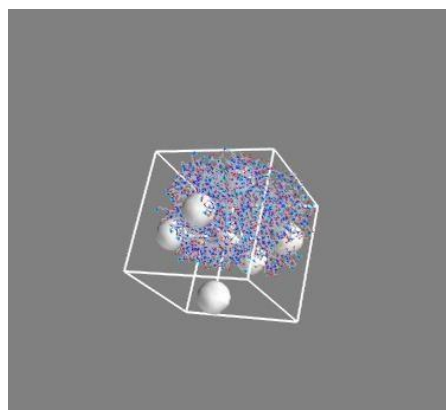


Figure 13: 2HBS & Vincristine

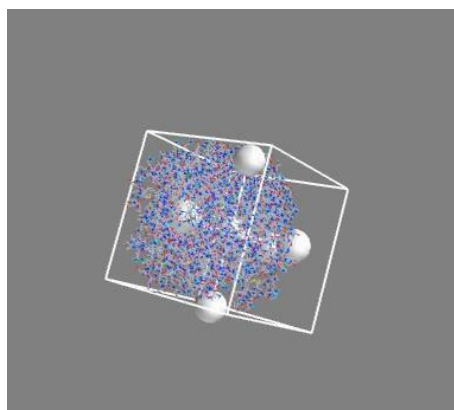


Figure 14: 2HBS & Vindesine

3.4 Binding result Structure

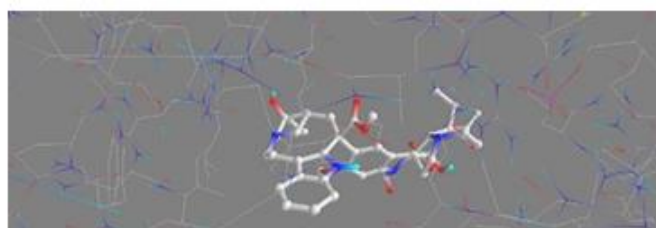


Figure 15:- 2KCE & Vincristine

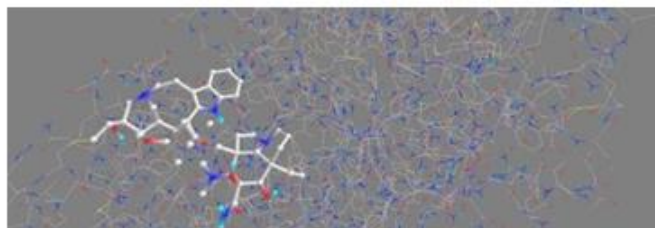


Figure 16:- 2KCE & Vindesine

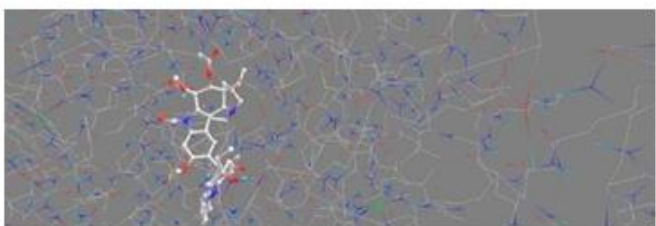


Figure 17:- 2HBS & Vincristine



Figure 18:- 2HBS & Vindesine

3.5. Biodiscovery Discovery Docking

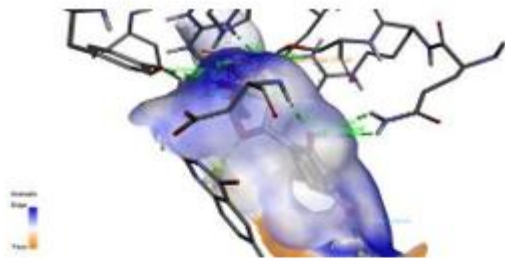


Figure 19:- Vincristine & 2KCE

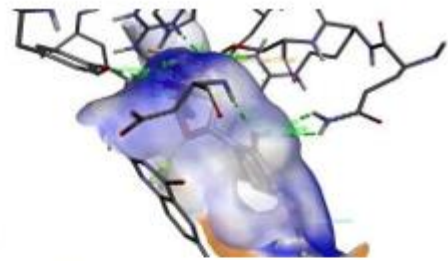


Figure 20:- Vindesine & 2KCE

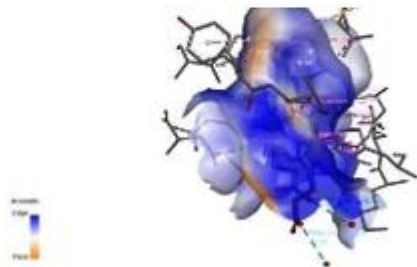


Figure 21:- Vincristine & 2HBS

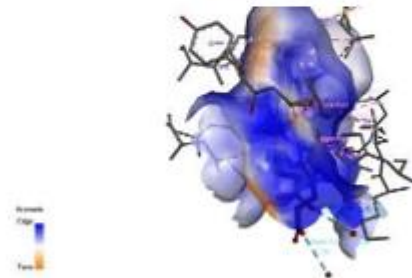


Figure 22:- Vindesine & 2HBS

3.6 Amino acid (2D Structure)

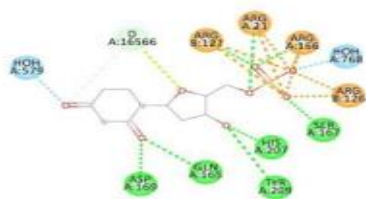


Figure 23:- Vincristine & 2KCE

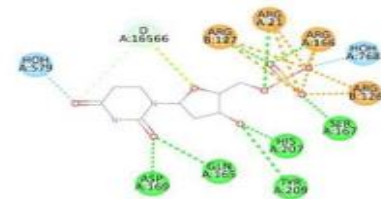


Figure 24:- Vindesine & 2KCE

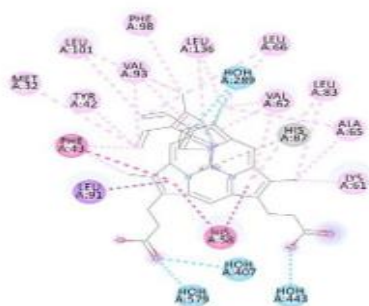


Figure 25:- Vincristine & 2HBS



Figure 26:- Vindesine & 2HBS

3.7 Binding energy

Table 2: Binding energy's of Protein & Receptors

Sr.no	Receptor /Protein	2KCE	2HBS
1	Vincristine	-7.2	-9.5
2	Vindesine	-9.1	-10.5

Our model is based on the ligand based pharmacophoric features of already known active compounds of the plant species. Structures of health beneficial phytochemicals i.e. Vincristine, Vindesine, present in *Catharanthus roseus* were extracted and analyzed using PubChem & the proteins are downloaded using protein data bank (PDB) & that is PDBID (2KCE, 2HBS). They have good binding energy.

Medicinal plants may treat a variety of ailments. *Catharanthus roseus* is a valuable medicinal plant that has traditionally been used to treat numerous health conditions, including Anti-cancer & antitumour activity. It is mostly used in cancer treatment because they have good Anti-cancer activity. It is also called as good Anti-cancer agent.

4. CONCLUSION

In silico molecular docking studies of *Catharanthus roseus* revealed potential phytochemical targets, including Vincristine, Vindesine phytochemical classes and structural manifolds. These phytochemicals are likely to target proteins of Anti-cancer disease or Anti-cancer agent¹⁹. This work suggests that *Catharanthus roseus*, a polyphenolic molecule, may have antioxidant, antibacterial, antifungal, & anticancer properties against the Anti-cancer receptors responsible for Anti-Cancer illness, as anticipated in silico. Molecular docking data indicates that Vincristine (-7.2, -9.5 Kcal/mol) and Vindesine (-9.1, -10.5 Kcal/mol). The polyphenolic molecule binds accurately to the receptor's active site, which is determined by the amino acids present. This study investigated pharmacokinetic characteristics and drug-likeness, identifying two molecules as potential lead compounds for future investigation²⁰. Vincristine has maximum binding (-9.5 Kcal/mol) against 2HBS receptor, Vindesine having maximum binding energy (-10.5 Kcal/mol) against 2HBS. Then In silico screening of bioactive compounds derived from *Catharanthus roseus* Against Anti-Cancer agent the 2HBS was found to be the best Protein for Anti-Cancer treatment from all the selected phytoconstituent.

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