RESEARCH PAPER

Screening of Phytochemicals to inhibit E7 protein of HPV to treat Cervical Cancer – A computational Approach

Kamali Manickavasagam Lekshmi[1*], Leelaprasaanth M G[1], Nanthini K[1], Yogasri T[1], Reshath R[1]

[1] Department of Biotechnology, KIT-Kalaignarkarunanidhi Institute of Technology, Coimbatore, Tamil Nadu, India.

[*] Corresponding author e-mail: kamalikitbt@gmail.com

ABSTRACT

Cervical cancer begins in the cervix, most often caused by various types of Human papillomavirus (HPV). A specific variant of HPV lead to changes in the cervix resulting in cervical cancer, whereas other variants cause genital or skin warts. Tumor formation is induced by the proteins E6 and E7 present in the HPV. The expression of genes encoding E6 and E7 is regulated by another protein E2. Current treatment method involves the removal of uterus along with some part of the vagina and the surrounding lymph nodes. This is an expensive method and have severe side effects demanding for a safer and more economical treatment options. In this research work, we selected seven phytochemical which has been reported to have anti-cancer activity. The phytochemicals were screened based on its pharmacokinetic property and their binding affinity with E7 protein. The predicted pharmacokinetics and bioavailability radar confirmed Asarinin as a promising phytochemical that possess anti-cancer properties.

Keywords: Cervical carcinoma, molecular docking, dynamics, phytochemicals, E7(2B9D), in silico study, Asarinin

How to cite this article: Kamali Manickavasagam Lekshmi, Leelaprasaanth M G, Nanthini K, Yogasri T, Reshath R. Screening of Phytochemicals to inhibit E7 protein of HPV to treat Cervical Cancer – A computational Approach. Int J Drug Deliv Technol. 2026;16(37s): 296-305. DOI: 10.25258/ijddt.16.37s.39

Source of support: Nil.

Conflict of interest: None

Abbreviations: HPV- human Papilloma Virus, OPRMI – Opioid receptor, mu 1, DCCM – Dynamics cross-correlation map, HPLC- High Performance Liquid Chromatography, PDB – Protein Data Base, GC/MS- Gas Chromatography, DPPH – 2,2-diphenyl-1-picry;hydrazyl, MTT – 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenylatetrazolium bromide assay, RMSD- Root Mean Square Deviation, RMSF- Root Mean Square Fluctuation, Rg – Radius of gyration, SASA- Solvent accessible surface area, PCA – Principal Component Analysis, MM-PBSA – Molecular Mechanics-Poisson-Boltzmann Surface Area,

1. Introduction

According to the cancer statistics of 2025, cancer is the second leading global cause of mortality and frequently strikes individuals younger than the age 85. The third edition of the 'International classification of Diseases for Oncology' is commonly used to categorize various cancers. Yet, it is not primarily utilized in categorizing cancers in children and adults. The predicted cancer death rate for the year 2025 was estimated using the previous year's data and the Joinpoint algorithm [1].

Cancer statistics 2024 and 2025 state that, out of all other cancers, cervical carcinoma remains the second leading cause of mortality among women since 2019. Even though it is preventable by HPV vaccination, women aged between 20 and 39 years are often prone to cervical cancer. It is classified into squamous cell carcinoma (easy to detect and more common) and Adenocarcinoma (low survival rate and hard to detect) [2] . Despite the medical advancements, there is no significant improvement in the survival rate of adenocarcinoma. Cytology screening (Pap smear) is the method used to detect cervical cancer, but it is not effective for detecting adenocarcinoma, which makes the increased count of deaths, therefore increasing the overall death rate.[3]

Cervical cancer is caused by multiple variants of HPV; among those variants, HPV16 is known to cause persistent infections by suppressing interferon responses. HPV16-infected cells disrupt the function of antigen-presenting cells and weaken the immune responses [4]. The E6 and E7 present in the cytoplasm and nucleus of the infected cells are early coat proteins transformed into the cell by the HPV16 DNA virus, which starts the growth of cervical carcinoma and disrupts key regulatory pathways. p53 is the major tumor suppressor gene that maintains the gene

correction mechanism of mutated genes [5]. It undergoes ubiquitination and proteasomal degradation when E7 binds to it, preventing apoptosis and allowing the damaged cells to multiply with the mutated genes, resulting in genetic instability and mutation. E7 works along with E6 to proliferate cervical cancer [6]. It binds with retinoblastoma, leading to uncontrolled proliferation of cells and disturbed cell signalling pathways. pRb is a key regulator of cell signalling pathways and maintains the activity of E2F transcription factors. It becomes inactive when E7 binds to it, which leads to uncontrolled cell cycle progression [7]

There are some clinical drugs found in the market for treating cervical cancer, like Cisplatin, Fluorouracil, Avastin, Paclitaxel, etc. Also, there are some FDA and WHO-approved preventive HPV vaccines like Gardasil 9, Gardasil (Quadrivalent), and Cervarix, which contain virus-like particles to prevent the infection but cannot treat the existing one. But there are possibilities that these vaccines can cause anaphylaxis, joint pain, muscle pain, etc [8] Also, cisplatin and paclitaxel are known to cause kidney damage, nerve disorders, hearing loss, etc. It is also a well-known fact that conventional therapies cause hair loss and other side reactions. Plants contain many bioactive compounds that are commonly called phytochemicals. They possess powerful properties that resist damage caused by toxic molecules, kill bacteria, inhibit viruses, combat fungal infections, prevent cancer, cause swelling to decrease and aid in healing [9] Certain medicinal plants were being explored for their anti-cancer activities and have been used as a conventional treatment method to treat cancers. *Piper longum* is a medicinal plant found commonly in India[10] (Karnataka, Kerala, Tamil Nadu) and is also well-known for its phytochemical composition. Its major composition includes alkaloids, lignans, oils, and terpenes [11].

In recent days, Asarinin was found to exhibit significant anticancer activity against a wide range of diseases. Also, it has effective anti-inflammatory and anti-migraine activities. It is highly soluble in water, requires prolonged heating time, and has a complex separation process; hence, it is a tricky process to isolate Asarinin from its source [12].

Asarinin is a furofuran lignan that is abundantly present in *Asarum seiboldii, Asarum heterotropoides, Zanthoxylum ailanthoides,* etc., and present in native plants like Piper longum. Furofurans are a major class of lignans present in *P. longum*. It has the ability to block some liver enzymes like CYP2C19, CYP2D6, and CYP3A4, leading to a slow down the breakdown of some other drugs. Absorption is effective when taken orally, and it can cross the blood-brain barrier, but topical application does not produce significant effects [13]

Piper longum, which belongs to the family Piperaceae, has been reported for its tremendous medicinal values. It also consists of abundant phytochemicals of various classes. The common name of this plant is 'long pepper' and is also called 'Thippili.' It is found widely in the regions around Tamil Nadu, Kerala, Karnataka, and the Himalayas in India. It contains alkaloids, terpenes, essential oils, lignans, etc., in abundant percentages [14].

Molecular docking and dynamics studies are computational tools that help in the prediction of behaviour, stability, and binding affinity of the phytochemicals with the target proteins. AutoDock Vina is a tool used to dock the ligand with the target protein and to find its binding efficiency. Gromacs is used to simulate the protein and ligand interact, and shows how stable the protein is when it connects with phytochemicals [6].

This research targets the prediction of the cytotoxicity effect of crude extract of *Piper longum*. For measuring cytotoxicity, HeLa cell lines were exposed to different concentrations, and then cell viability was determined using MTT assay [13]

2. MATERIALS AND METHODS

This research focuses on a comprehensive range of materials was utilized to investigate the efficacy of potential therapeutics against the target protein. The materials include computational software tools such as AutoDock for molecular docking and Gromacs for dynamics analysis. Additionally, databases like the Protein Data Bank (http://www.rscb.org/pdb) and PubChem (https://pubchem.ncbi.nlm.nih.gov/) were accessed to retrieve protein and ligand structures. Open Babel GUI (http://openbabel.org/docs/) was used for the conversion of file formats for ligands.

2.1 Selection and Preparation of Ligands

Seven phytochemicals that possess anti-cancer activity were chosen based on the review of literature. The selected phytochemicals are Asarinin (CID 11869417), Carvacrol (CID 10364), Charantin (CID 169435928), Cucurbitacin (CID 119287), Militarine (CID 171638), Palmatine (CID 19009), and Picroside (CID 9849283). The 3D shapes of these plant compounds were downloaded from PubChem, and their SDF files were changed into PDB format using Open Babel. Other information like chemical structures, molecular

formula, molecular weight, CID, and Canonical SMILES was also obtained from PubChem.

2.2 Preparation of Protein

The coat protein of HPV16, called E7 protein (PDB ID: 2B9D), was selected as a target protein because of its high carcinogenic and asymptomatic properties. The PDB file of E7 protein was downloaded from the Protein Data Bank (http://www.rscb.org/pdb) with 1.60 Å from X-ray diffraction. PyMOL (pymol.org) was used for viewing the protein structure, and the water atoms and B chains were deleted from the protein structure.

2.3 In Silico Pharmacokinetics and Screening of Ligands

The therapeutic properties of the selected phytochemicals were analyzed using SwissADME (https://swissadme.ch/) and several parameters, such as bioavailability, the Lipinski rule of 5, water solubility, lipophilicity etc., were taken and used for screening.

2.4 Molecular docking of E7 protein with the phytochemicals

After the preparation of protein and ligands, those three phytochemicals were docked with the E7 protein (PDB ID: 2B9D) using Auto Dock Vina. Docking is used to find how strongly the ligand binds with the protein, and with this, the potential drug, which has high binding efficiency, can be identified along with its binding energy. The water molecules present in the protein structure were deleted, and gastric charge and polar hydrogens were also added. The non-polar hydrogen atoms were merged [15]

The Kollman charges were added to the ligand molecule, and PDB files were converted into PDBQT files. Then the grid boxes were set up, and the coordinates were noted for future references. The result files were saved, and the binding energy was analysed [5]

2.5 Molecular simulation and Dynamics

The crystal structure of the HPV E7 CR3 domain (PDB ID: 2B9D) was retrieved as described in [PMID:16249186]. Top-scoring ligands were determined by docking analysis, and their topologies were calculated by the ATB server [PMID: 30289710]. Hydrogen atoms were added to the heavy atoms by GROMACS tool pdb2gmx [PMID: 16211538] [15]. The first energy minimization was performed using the steepest descent algorithm for 1500 steps in a vacuum environment. The minimized structures were then solvated using a simple point charge (SPC) water model in a cubic periodic simulation box. Proper amounts of Na^+ and Cl^- counterions were added to neutralize the system and to have a physiological salt concentration of 0.15 M, as reported methodologies in past studies [PMID: 31514687, PMID: 32567989, PMID: 34844519].

After the system was prepared, all structures underwent an equilibration period within the NPT ensemble. Then a production molecular dynamics (MD) simulation of 100 nanoseconds was carried out. Simulation trajectory analysis was conducted with some of the tools available in the GROMACS package, such as Principal Component Analysis (PCA), hydrogen bond analysis (H-Bond), solvent accessible surface area (SASA), radius of gyration (Rg), root mean square deviation (RMSD), root mean square fluctuation (RMSF), and free energy landscape (FEL) analysis [PMID:33050786, PMID: 3466392], the free energy of binding between inhibitor and protein (2B9D-DRG, STD complex) was calculated through the Molecular Mechanics Poisson-Boltzmann Surface Area (MM-PBSA) method. To this aim, the g-mmpbsa tool provided by GROMACS was utilized [PMID: 24850022]. To get a precise estimate, the free binding energy of the 2B9D-STD complex was determined using 1000 frames taken from the last 50 ns of the simulation path [5]

3. RESULTS AND DISCUSSION

3.1 Screening of ligands:

Upon analysing the data from SwissADME (Table 1), it was identified that cucurbitacin, militarine, charantin, and picroside have high molecular weight and violate Lipinski's criteria. Hence, the other three phytochemicals, asarinin, carvacrol, and palmatine, have been further taken for the docking study [16]

3.2 Results of docking E7 oncoprotein with three phytochemicals

Three phytochemicals asarinin, carvacrol, and palmatine, were chosen for molecular docking against the E7 (Fig.1.a) oncoprotein of HPV16. Asarinin has been shown to induce caspase-dependent apoptotic cell death in cancer cells through the activation of caspase-3, caspase-8, and caspase-9 in human ovarian cancer cells. Carvacrol, a major bioactive component of essential oils from aromatic herbs, has multiple therapeutic activities ranging from anti-diabetic to anti-cancer, cardio protection, hepatoprotection, anti-obesity, as well as antimicrobial activities. palmatine is another key phytocompound possessing the ability to mitigate symptoms and complications of metabolic syndrome like cardiovascular diseases, osteoporosis, and osteoarthritis, and possess high antioxidant activity. After molecular docking, the binding energies of all phytochemicals with the E7 oncoprotein were estimated and ranked. Asarinin (Fig.1.b) has shown high binding

affinity towards E7 protein among the other phytochemicals with a binding energy of -9.69 kcal/mol (Fig.1.c) (Table 2). The binding pocket of asarinin was predicted using Castp server online (Fig 1.d). The compound with the highest binding affinity among them was chosen for further molecular dynamics simulation and structural analysis.

3.3 Molecular simulation and dynamics of apo-protein and bounded complex

All -atom molecular dynamics (MD) simulation is a very efficient method to study the structural dynamics of proteins and their interaction with ligands. This method has greatly boosted computer-aided drug discovery and design by enabling detailed examination of molecular systems at the atomic level. MD simulations were carried out in the current research to analyze the dynamics changes caused by ligands binding to the target protein. A number of important structural parameters were examined, such as root mean square deviation (RMSD), root mean square fluctuation (RMSF), radius of gyration (Rg), solvent- accessible surface area (SASA), and intermolecular hydrogen bonding. Free energy landscape (FEL) analysis and principal component analysis (PCA) were also performed on the 100 ns simulation trajectory to obtain further insights into the conformational behaviour of the system.

3.3.1 Root Mean Square Deviation (RMSD)

In order to examine the protein-tyrosine phosphatase complex stability and understand the systems behaviour better, the RMSD results were examined over time (as indicated in Fig.2). The results show that both systems reached equilibrium within less than 10 ns and were evenly distributed throughout the simulation. It was found that the average RMSD values for APO, DRG, and both were 0.99 ± 0.22 and 0.61 ± 0.07, respectively. The RMSD data analysis showed that the docked complex remained unchanged throughout the simulation and the APO and DRG were also stable up to 100 ns. This result implies that the system composed of APO and DRG is stable and did not show notable oscillations during the simulation.

3.3.2 Root Mean Square Fluctuation (RMSF)

For quantifying the change of every residue and flexible part of a protein, MD simulations use RMSF. By examining RMSF through simulations, the impact of ligand binding on the protein can be ascertained. As a general rule, tightly organized protein structures such as helices and sheets exhibit lower RMSF values and loosely organized loop regions exhibit greater RMSF values. The RMSF values of all complexes in this work were calculated and graphed for each residue in DRG and STD complex, which are presented in Fig.3. It was found that the average RMSF values for APO, DRG, and both were 0.39 ± 0.13 nm, 0.26 ± 0.12 nm, respectively. The results demonstrate that the combination of LIG and STD did not significantly alter the overall RMSF distribution.

3.3.3 Radius of gyration (Rg)

The values of Rg were calculated and plotted against time (Fig. 4) in order to check how stable and compact the APO and DRG complexes were, the average Rg values were measured. APO had an average Rg of 1.92 ± 0.14 nm and DRG had 1.59 ± 0.04 nm.

3.3.4 Solvent accessible surface area (SASA)

When checking how much of the protein is exposed to the solvent, SASA is a useful statistic. To determine how APO and DRG complexes affect target's exposure to the solvent, this study calculated and graphed the SASA (Fig.5). The plot shows that the SASA values of APO and DRG follow a similar trend. APO and DRG were found to have average SASA values of 69.06 ± 4.92 nm and 69.90 ± 2.25 nm, respectively. The SASA results show no oscillations and fair equilibration throughout the simulation.

3.3.5 Intermolecular Hydrogen Bond

One significant factor determining the stability of protein and ligand interactions is the ability to form hydrogen bonds. In this study, it was discussed that the results of our research on the behaviour of hydrogen bonds between DRG over a period of simulation (Fig.6). What we observed was that at least four to five hydrogen bonds between docked and DRG complexes stabilized the complex during the simulation.

3.3.6 Principal Component Analysis (PCA)

In order to investigate the group motions within APO, DRG, by using PCA. The motility of a protein molecule varies based on its few eigenvectors (EVs). Thus utilized PCA to study how the shapes of APO and DRG changed during simulation (Fig.7). PCA time evolution displays stability by reducing the overall flexibility of the APO, DRG complex on both EVs, as evidenced by Figure F. The graph clearly shows that APO and DRG complexes overlapped and covered most of the shape changes. Generally speaking, the fewer movements seen in the APO, DRG had no appreciable impact on the target's dynamics and conformation, suggesting the complex's stability.

3.3.7 MM - PBSA

In order to find out how strongly DRG binds, then analysed its binding strength using the protein's energy data during the simulation. Table 3 illustrates the binding strengths of DRG and inhibitors by the MM_PBSA method. And compute residue-level

contributions to interaction energy over a stable simulation track.

4. CONCLUSION

Asarinin is a furofuran lignan that has potential therapeutic properties in various fields. Through molecular docking and dynamics study, it was found that it has good binding affinity with the E7 oncoprotein, and this ligand-protein complex is stable over a period of time. It has not shown notable fluctuations when the complex is simulated, and the protein stability is comparatively higher for the ligand-bound complex than the apoprotein. Asarinin can be used as an effective drug to inhibit the activity of E7 protein from disrupting the regular function of pRb genes.

Common sources of asarinin are *Asarum seiboldii*, *Asarum heterotropoides*, *Zanthoxylum piperitum*, *Chrysopogon zizanioides, Piper longum,* etc. All these plant species except for *P. longum* and *C. zizaniodes* are native plants of China, Korea, and Malaysia. *C. ziznioides* is commonly called Vettiver, and it is reported to have a lower concentration of asarinin than in *P. longum*. Also, *P. longum* has many other medicinal and therapeutic qualities, such as being an antimicrobial agent, an antitumor agent, and an anticancer agent.

Acknowledgements The authors are thankful to KIT-Kalaignarkarunanidhi Institute of Technology Coimbatore, for providing research facilities in Department of Biotechnology.

Author Contribution KML – conceptualization, methodology and supervision, LMG, NK, YT & RR - Data curation, original draft preparation, visualization, investigation, software validation, reviewing and editing.

Funding The authors received no financial support for the research, authorship, and/or publication of this article.

Data availability Data will be available on request.

Code availability Not applicable.

Declarations

Ethical Approval Not applicable.

Consent to participate Not applicable.

Consent for publication All authors read and approved for publication.

Competing interests the authors declare no competing interests.

References:

[1] V. D. Kharisma, A. N. M. Ansori, M. H. Widyananda, S. L. Utami, and A. P. Nugraha, "Molecular simulation: The potency of conserved region on E6 HPV-16 as a binding target of black tea compounds against cervical cancer," *Biochem Cell Arch*, vol. 20, pp. 2795–2802, 2020, doi: 10.35124/bca.2020.20.S1.2795.

[2] S. Prakash *et al.*, "Isolation of hesperetin - A flavonoid from Cordia sebestena flower extract through antioxidant assay guided method and its antibacterial, anticancer effect on cervical cancer via in vitro and in silico molecular docking studies," *J Mol Struct*, vol. 1207, May 2020, doi: 10.1016/j.molstruc.2020.127751.

[3] L. Zhang, J. Lv, M. Xiao, L. Yang, and L. Zhang, "Exploring the underlying mechanism of action of a traditional Chinese medicine formula, Youdujing ointment, for cervical cancer treatment," *Quantitative Biology*, vol. 9, no. 3, pp. 292–303, 2021, doi: 10.15302/J-QB-021-0236.

[4] Z. B. Li, J. Y. Wang, B. Jiang, X. L. Zhang, L. J. An, and Y. M. Bao, "Benzobijuglone, a novel cytotoxic compound from Juglans mandshurica, induced apoptosis in HeLa cervical cancer cells," *Phytomedicine*, vol. 14, no. 12, pp. 846–852, Dec. 2007, doi: 10.1016/j.phymed.2007.09.004.

[5] A. Hidayatullah *et al.*, "Concatenation of molecular docking and dynamics simulation of human papillomavirus type 16 E7 oncoprotein targeted ligands: In quest of cervical cancer's treatment," *An Acad Bras Cienc*, vol. 95, 2023, doi: 10.1590/0001-3765202320220633.

[6] D. Salaria *et al.*, "Phytoconstituents of traditional Himalayan Herbs as potential inhibitors of Human Papillomavirus (HPV-18) for cervical cancer treatment: An In silico Approach," *PLoS One*, vol. 17, no. 3 March, Mar. 2022, doi: 10.1371/journal.pone.0265420.

[7] S. K. Saha and A. R. Khuda-Bukhsh, "Berberine alters epigenetic modifications, disrupts microtubule network, and modulates HPV-18 E6-E7 oncoproteins by targeting p53 in cervical cancer cell HeLa: A mechanistic study including molecular docking," *Eur J Pharmacol*, vol. 744, pp. 132–146, Feb. 2015, doi: 10.1016/j.ejphar.2014.09.048.

[8] J. W. Lee, S. Park, S. Y. Kim, S. H. Um, and E. Y. Moon, "Curcumin hampers the antitumor effect of vinblastine via the inhibition of microtubule dynamics and mitochondrial membrane potential in HeLa cervical cancer cells," *Phytomedicine*, vol. 23, no. 7, pp. 705–

713, Jun. 2016, doi: 10.1016/j.phymed.2016.03.011.

[9] M. Kori, K. Y. Arga, A. Mardinoglu, and B. Turanli, "Repositioning of Anti-Inflammatory Drugs for the Treatment of Cervical Cancer Sub-Types," *Front Pharmacol*, vol. 13, Jun. 2022, doi: 10.3389/fphar.2022.884548.

[10] N. Ahmeda, M. Nazmul Hasan Zilani, ; Kazi, T. Md, T. ; Md, and K. Al-Din, "PHYTOCHEMICAL, ANTIBACTERIAL AND ANTI-OXIDANT ACTIVITY OF PIPER LONGUM LEAVES," vol. 1, pp. 27–35, 2019, [Online]. Available: http://pharmacologyonline.silae.it

[11] V. Yadav, A. Krishnan, and D. Vohora, "A systematic review on Piper longum L.: Bridging traditional knowledge and pharmacological evidence for future translational research," *J Ethnopharmacol*, vol. 247, p. 112255, 2020, doi: https://doi.org/10.1016/j.jep.2019.112255.

[12] Y. Xiao, Z. Liu, H. Gu, F. Yang, L. Zhang, and L. Yang, "Improved method to obtain essential oil, asarinin and sesamin from Asarum heterotropoides var. mandshuricum using microwave-assisted steam distillation followed by solvent extraction and antifungal activity of essential oil against Fusarium spp," *Ind Crops Prod*, vol. 162, Apr. 2021, doi: 10.1016/j.indcrop.2021.113295.

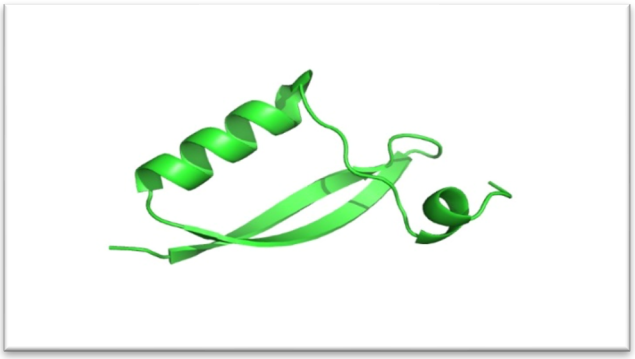
[13] M. Jeong, H. M. Kim, J. S. Lee, J. H. Choi, and D. S. Jang, "(−)-Asarinin from the roots of asarum sieboldii induces apoptotic cell death via caspase activation in human ovarian cancer cells," *Molecules*, vol. 23, no. 8, 2018, doi: 10.3390/molecules23081849.

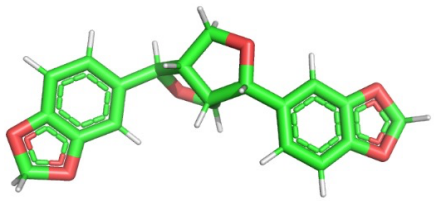
[14] K. Haribabu, "Simultaneous Determination of Asaranin and Sesamin in Piper chaba Fruit by using HPTLC-MS Method: Effect of Different Extraction Methods on the Yield of Marker Compounds," *J Anal Bioanal Tech*, vol. 5, no. 4, 2014, doi: 10.4172/2155-9872.1000199.

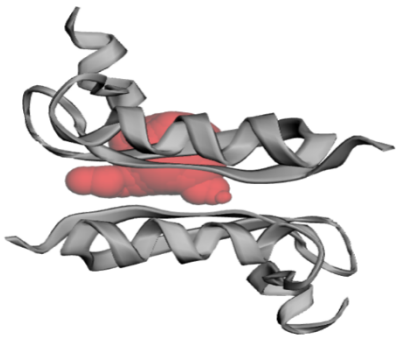
[15] A. Nag, P. Verma, S. Paul, and R. Kundu, "In Silico Analysis of the Apoptotic and HPV Inhibitory Roles of Some Selected Phytochemicals Detected from the Rhizomes of Greater Cardamom," *Appl Biochem Biotechnol*, vol. 194, no. 10, pp. 4867–4891, Oct. 2022, doi: 10.1007/s12010-022-04006-3.

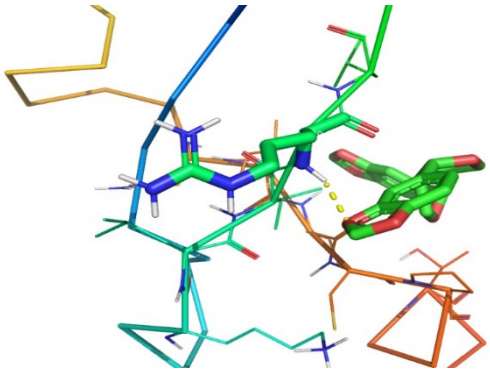
[16] W. Ahmad *et al.*, "In Vitro, Molecular Docking and In Silico ADME/Tox Studies of Emodin and Chrysophanol against Human Colorectal and Cervical Carcinoma," *Pharmaceuticals*, vol. 15, no. 11, 2022, doi: 10.3390/ph15111348.

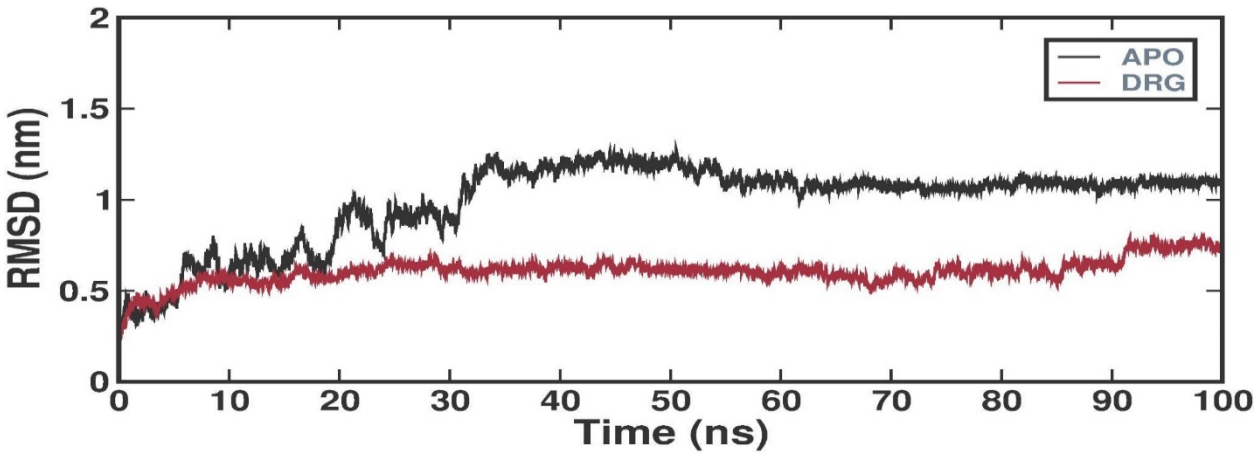
Figures

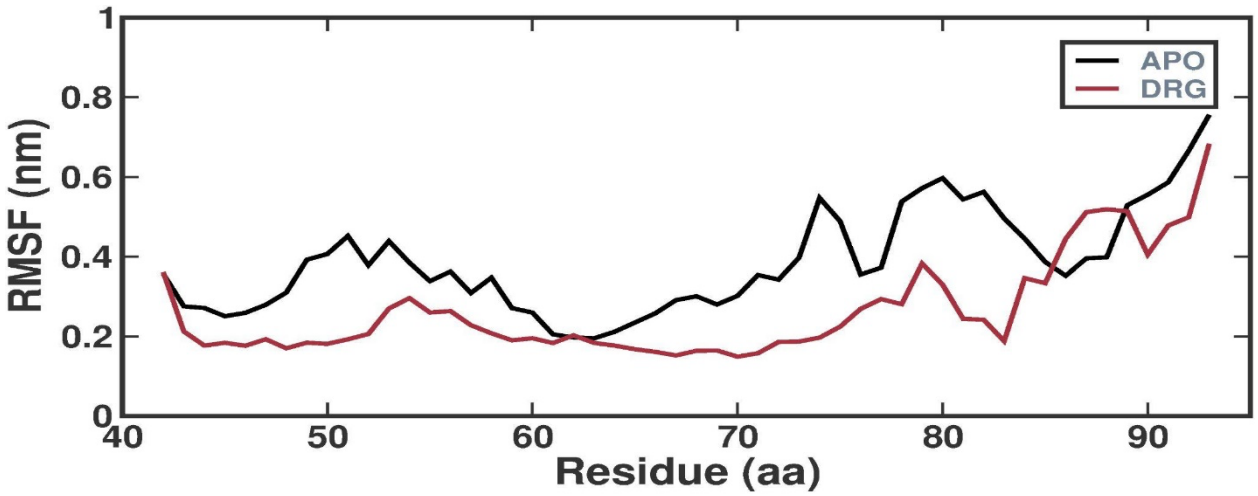


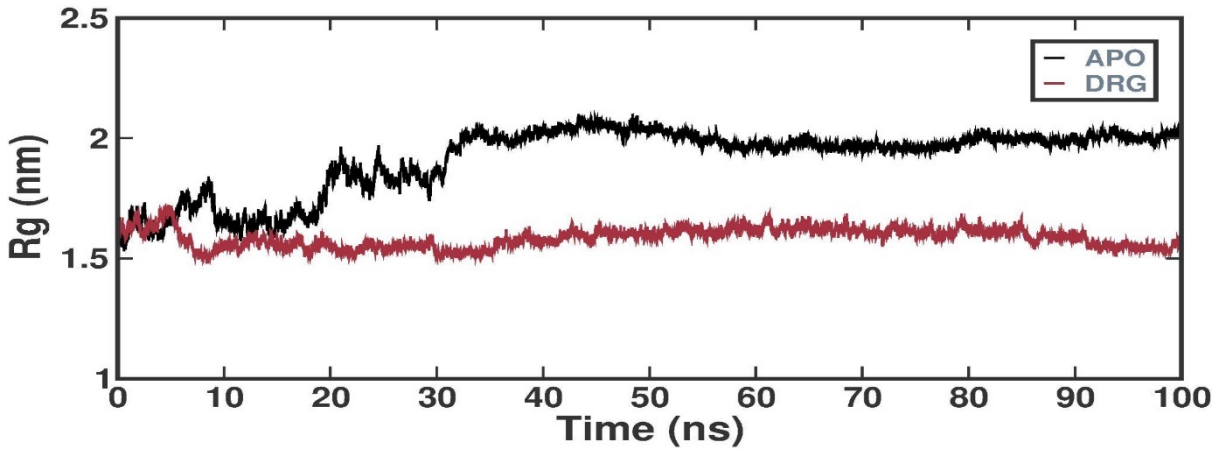


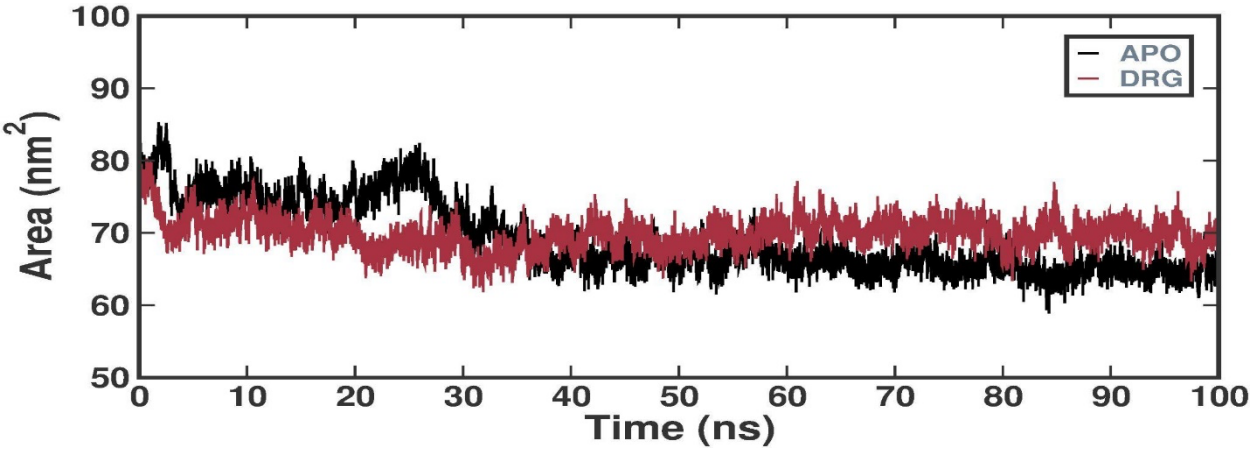


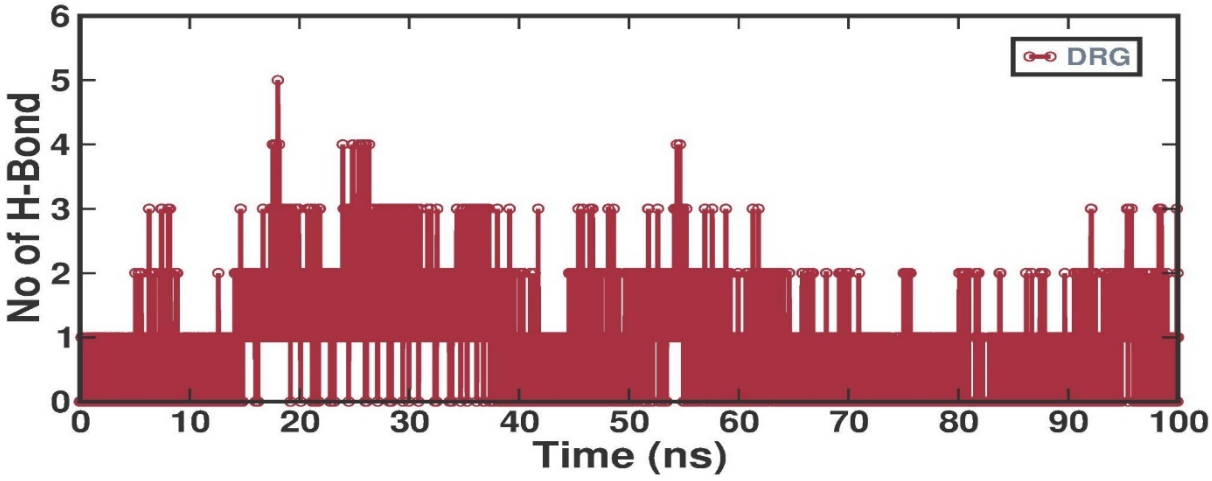





RMSF (nm)
Residue (aa)
APO
DRG


Rg (nm)
Time (ns)
APO
DRG


Area (nm^2)
Time (ns)
APO
DRG



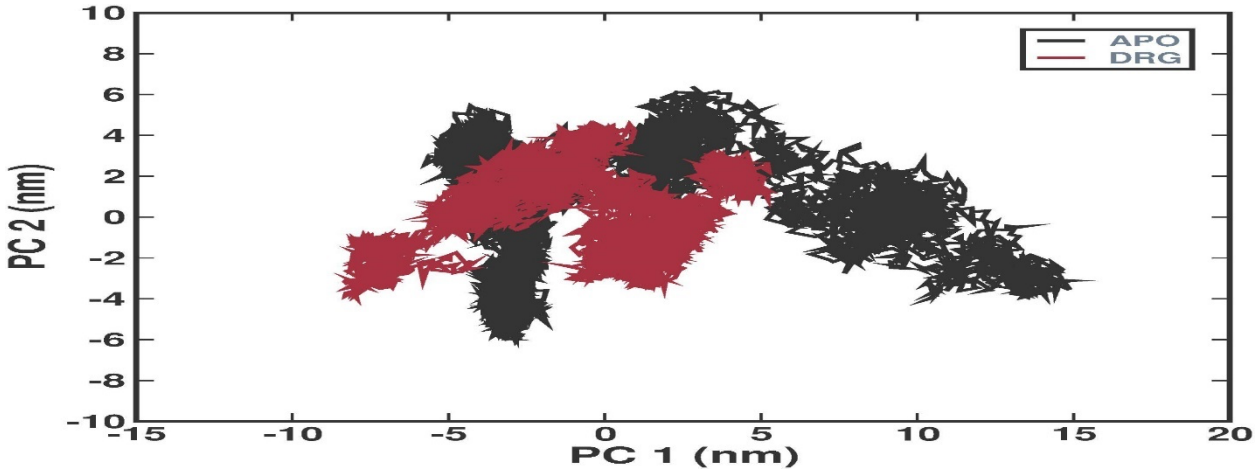


Figure Legends

Figure 1: a) E7 Protein b) Asarinin c) Binding pockets d) Asarinin docked with E7 protein

Figure 2: RMSD Conformational dynamics analysis of APO, DRG

Figure 3: Analysis of the APO and DRG complex using RMSF conformational dynamics

Figure 4: Rg Conformational dynamics analysis of LIG and STD complex

Figure 5: SASA Conformational dynamics analysis of APO, DRG complex

Figure 6: Intermolecular hydrogen bonds between protein and during the simulation time.

Figure 7: Principal component analysis 2D projection plot shows the conformation sampling of LIG and STD on PC1 and PC2.

Tables

Phytochemicals	Molecular weight(g/mol)	No of H-bond acceptor	No of H-bond donor	Log $P_{o/w}$	TPSA $Å^2$	Bioavailability Score	Lipinski Rule
Carvacrol	150.22	1	1	2.24	20.23	0.55	Yes
Palmatine	352.4	4	0	0.10	40.80	0.55	Yes
Asarinin	354.4	6	0	3.37	55.38	0.55	Yes
Cucurbitacin	486.6	6	2	3.2	101	0.55	No
Picroside	512.5	13	6	-1.4	197	0.55	No
Militarine	726.7	17	9	-0.5	272	0.55	No
Charantin	1151.7	23	12	6.17	359.97	0.55	No

Table 1: Drug likeness of Phytochemicals

Phytochemicals	Binding energy(kcal/mol)
Asarinin	- 9.69
Palmatine	- 7.54
Carvacrol	- 5.45

Table: 2 Phytochemicals and their binding energies. Asarinin has high binding efficiency with the E7 protein and hence, it can be taken as potential phytochemical for binding with the target protein.

System	Van der Waal energy	Electrostatic energy	Polar solvation energy	Energy of binding
DRG	-146.623 +/- 10.344 kJ/mol	-13.989 +/- 12.262 kJ/mol	106.850 +/- 25.575 kJ/mol	-69.875 +/- 26.135 kJ/mol

Table: 3 MM-PBSA