Phytochemical and Docking Evaluation of *Phyllanthus Niruri* L. Extract Reveals Cytotoxic Potential Against Cervical and Breast Cancer Cells

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ABSTRACT

Phyllanthus niruri L., a widely used traditional medicinal plant, has been reported to possess various therapeutic properties, including anticancer potential. However, its direct cytotoxic effects on specific cancer cell lines such as HeLa (cervical cancer) and MDA-MB-231 (triple-negative breast cancer) remain underexplored. This study aimed to evaluate the anticancer efficacy of the aqueous leaf extract of *Phyllanthus niruri* against these cell lines using in vitro and in silico approaches. The MTT assay revealed that the extract induced significant dose-dependent cytotoxicity in both cell lines, with an IC_{50} of 0.1924 µg/mL for HeLa and 0.017 µg/mL for MDA-MB-231, indicating a stronger effect on breast cancer cells even at lower concentrations. To further support the anticancer potential, antioxidant activity was assessed via DPPH free radical scavenging assay, which showed increased scavenging capacity with rising concentrations, suggesting the presence of bioactive phytochemicals. GC-MS analysis identified key phytoconstituents, including alkanes, alkenes, phenolic compounds, alcohols, thiadiazoles, and terpenes. Molecular docking studies were performed using the breast cancer target protein 6NM8, where most compounds exhibited weak to moderate binding affinities. Among these, 2-Hexyl-1-decanol showed the most promising interaction (-5.1 kcal/mol) with acceptable ADME properties. These findings highlight the cytotoxic efficacy of *Phyllanthus niruri* L., especially against MDA-MB-231 cells, and suggest its potential as a lead source for anticancer therapeutics. Further studies are warranted to isolate individual active compounds and elucidate their mechanisms of action.

Key Words: Anticancer, HeLa, MDAMB231, *Phyllanthus niruri*

INTRODUCTION

Phyllanthus niruri, a tropical shrub native to India and belonging to the family *Euphorbiaceae*, is widely distributed in coastal regions and has been extensively utilized in traditional medicine systems across the globe. This plant, known by various vernacular names such as "Bhumyamalaki" in Ayurveda and "Chanca Piedra" in South America, has been traditionally employed to treat a wide array of ailments, including jaundice, gonorrhea, diabetes, ulcers, and inflammation[1]. Its extensive use in traditional medicine is attributed to its diverse biological activities, which include anti-inflammatory, antioxidant, antimalarial, antimicrobial, antiviral, and anticancer properties [1].

The phytochemical exploration of *Phyllanthus niruri* dates back to 1861, when Ottow first isolated significant chemical constituents from the plant. Among these, lignan phyllanthin was identified as a key bioactive compound [2]. Subsequent research has revealed that *Phyllanthus niruri* is rich in a variety of phytochemicals,

including tannins, flavonoids, alkaloids, terpenes, coumarins, lignans, and phenylpropanoids, which are responsible for its pharmacological activities [3]. Notably, recent studies have identified nirtetralin and niranthin as potent anti-hepatitis B virus (HBV) compounds, further highlighting the plant's therapeutic potential[3][4]. Despite the extensive research on its biological activities, there is limited information on the potential side effects and toxicities of *Phyllanthus niruri*, with only a few studies addressing these concerns [5].

In recent years, there has been a surge in studies investigating the medicinal properties and chemical constituents of *Phyllanthus niruri*. However, despite its widespread use in ethnomedicine, much of the research has not progressed to clinical trials, leaving many of its potential therapeutic applications unexplored [6]. Among its many biological activities, *Phyllanthus niruri* has demonstrated significant antitumor properties, making it a promising candidate for cancer research [7].

Cancer remains one of the most pressing global health challenges, with cervical and breast cancers being particularly significant threats to women's health. The HeLa cell line, derived from cervical cancer cells taken from Henrietta Lacks in 1951, was the first human cell line to be successfully cultured and remains one of the most widely used cell lines in cancer research [8] [9]. Breast cancer, on the other hand, continues to be a leading cause of morbidity and mortality among women worldwide, with its incidence increasing annually[10]. Among the various subtypes of breast cancer, triple-negative breast cancer (TNBC) is particularly aggressive and associated with poor prognosis, making it one of the most dangerous forms of the disease [11].

Given the rising incidence of cervical and breast cancers, there is an urgent need for novel therapeutic strategies. The present study focuses on evaluating the antiproliferative activity of *Phyllanthus niruri* against two cancer cell lines: HeLa (cervical cancer) and MDA-MB-231 (triple-negative breast cancer). These cell lines were chosen due to their clinical relevance and the increasing burden of these cancers globally [12] [13]. By investigating the potential of *Phyllanthus niruri* as a natural anticancer agent, this study aims to contribute to the development of new therapeutic options for these devastating diseases.

MATERIAL AND METHODS

Plant Collection and Preparation

The plant specimen of *Phyllanthus niruri* was collected from an open field near Nagari, Andhra Pradesh, India, during the month of December, which corresponds to the winter season in this region. The collection site was chosen due to the plant's natural abundance in this area and its accessibility for harvesting.

After collection, the whole plant was thoroughly washed under running tap water to remove surface impurities, soil particles, and any debris. To ensure complete cleanliness, the plant was then soaked in distilled water for approximately 5 minutes, followed by a second rinse with distilled water. This step was crucial to eliminate any residual contaminants that could interfere with subsequent processing or analysis.

Once cleaned, the plant was air-dried at room temperature (approximately 25–30°C) to remove excess moisture. After air-drying, the plant parts (leaves, stems, and roots) were carefully separated and further dried in a hot air oven at a controlled temperature of 35°C for one week. This low-temperature drying process was employed to preserve the thermolabile phytochemicals present in the plant, ensuring minimal degradation of its bioactive compounds.

Following the drying process, the leaves of *Phyllanthus niruri* were ground into a fine powder using an electric grinder. The powdered material was then sieved through a mesh to achieve a uniform particle size, which is essential for consistent extraction and analysis. The finely sieved powder was packed in an airtight container to protect it from moisture, light, and environmental contaminants, and stored at room temperature until further use. This storage method was adopted to maintain the stability and integrity of the plant material for subsequent phytochemical and pharmacological studies.

This standardized protocol for plant collection, cleaning, drying, and powder preparation ensures the reproducibility of results and the preservation of the plant's bioactive constituents, which are critical for downstream applications in research and traditional medicine.

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Soxhlet Extraction Procedure

For the extraction of bioactive compounds from *Phyllanthus niruri* leaves, the Soxhlet extraction method was employed. Initially, 5 grams of the finely powdered leaf material was accurately weighed and carefully packed into a thimble made of Whatman filter paper (Catalog No. 1004-125, GE Healthcare Life Sciences). The thimble containing the plant material was then placed in the Soxhlet apparatus, and 200 mL of distilled water was added as the extraction solvent.

The temperature of the heating mantle was set to 70°C to ensure optimal extraction efficiency while avoiding the degradation of heat-sensitive compounds. The extraction process was carried out continuously for 3 hours, allowing the solvent to cycle repeatedly through the plant material, thereby facilitating the efficient extraction of phytochemicals. After completion of the extraction process, the solvent containing the extracted compounds was collected and transferred to a clean container.

The collected solvent was then subjected to drying in a hot air oven set at 35°C to evaporate the liquid completely. This step was performed to isolate the solid extract from the solvent. Once the drying process was complete, the dried extract was carefully weighed, yielding a total of 1.165 grams. The dried extract was subsequently redissolved in 4 mL of distilled water to prepare a concentrated solution for further analysis.

To ensure sterility and remove any particulate matter, the extract was filter-sterilized using a syringe filter system. A 5 mL BD Emerald syringe (Reference No. 307757) was used in conjunction with a Whatman UNIFLO 25 mm syringe filter (Catalog No. 9913-2502). The filtered extract was collected in a sterile tube and stored at 4°C to maintain its stability and prevent microbial contamination until further use.

This standardized Soxhlet extraction protocol ensures the efficient recovery of bioactive compounds from *Phyllanthus niruri* leaves while maintaining the integrity of the extracted phytochemicals. The resulting extract is suitable for subsequent phytochemical, pharmacological, and biological studies.

Cell Culture

The two cell lines HeLa and MDAMB-231 were procured from National centre for Cell Science (NCCS Pune, India). The cells were grown in 35mm tissue culture dishes (LOT NO. 244D-20-240120) with complete media which is composed of DMEM/F12 (Ref No.12500-062, Gibco, Life Technologies Limited UK) and 10% FBS (Ref No.10271-106, Gibco, Thermo Fisher Scientific India Pvt, Ltd.). And the cells were incubated at 37^0C in CO_2 incubator (Panasonic, MCO-20AIC).

MTT Assay

The crude drug was screened for anticancer activity using MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide, a tetrazole] (Ref No.TC191-1G, HiMedia) assay. Briefly 2000 cells were seeded in each well of a 96 well plate and was incubated at 37^0C in a CO_2 Incubator for 24hrs (Panasonic, MCO-20AIC). After 24 hours incubation the medium was replaced with 100µl of drug dilutions at different concentrations [3mg, 1.5mg, 0.75mg, 0.37mg, 0.18mg, 0.09mg] and incubated overnight. Later the drug was replaced with 90µl of fresh medium to which 10µl of MTT solution was added and kept for 4-hour incubation at 37^0C. After the formation of formazan crystals 100ul DMSO was added to dissolve the crystals (CAS No.67-68-5, Thermo Fisher Scientific India Pvt Ltd) and incubated for another 10 minutes and read spectrophotometrically at 570nm to determine the IC_{50} of the drug.

DPPH Assay

To measure the free radical scavenging activity of the drug 0.5mM DPPH was prepared freshly by dissolving 10mg DPPH with 50ml of methanol (CAS No. 67-56-1, Thermo Fisher Scientific India Pvt. Ltd.). Different dilutions of the drug [600µg/ml, 300µg/ml, 150µg/ml, 75µg/ml, 37.5µg/ml, 18.75µg/ml] was prepared and 1ml of each concentration was taken in different tubes to which 0.5ml of DPPH was added and vortexed (Remi CM-

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101) respectively. This was incubated in dark at room temperature for 30 minutes and the intensity was read spectrophotometrically at 517nm and the free radical scavenging activity was calculated with the formula:

$$\% \text{ Of inhibition} = \frac{\text{control-test} \times 100}{\text{Control}}$$

GC-MS (Gas Chromatography-Mass Spectrometry) analysis

In GC-MS analysis 2µl of the sample was injected into the instrument in a slip mode using Rtx5MS-30m column with 0.25-mm and 0.25µm df. Then using the helium gas as the carrier gas the GC-MS was run with the flow rate of 1ml/mm and the analysis was recorded at 2 scan/sec with the scanning range of 40 to 850 m/z by mass spectra. And finally based on the peak areas and normalization of the internal standard obtained each component were quantified.

RESULTS

MTT Assay

The anticancer activity of the aqueous extract of *Phyllanthus niruri* leaves on HeLa and MDAMB 231 was assessed by MTT method. The mortality rate of the cells was found to raise with respect to increase in the concentration. In case of HeLa cells, the highest cell death was found to be 91.03% (Table1) at the highest concentration 3mg/µl with the average IC50 of 0.1924mg/µl. And in case of MDAMB231 cells, the highest cell death was found to be 94.79% at 3mg/µl.

Table 1 Anticancer activity of aqueous extract of Phyllanthus niruri L. against HeLa

Drug concentration (mg/µl)	OD at 570nm	Cell death in %
Media control	1.014	0
Vehicle control	1.003	1.09
Positive control	0.451	55.53
3	0.091	91.03
1.5	0.120	88.17
0.75	0.161	84.13
0.37	0.248	75.55
0.18	0.394	61.15
0.09	0.985	2.86

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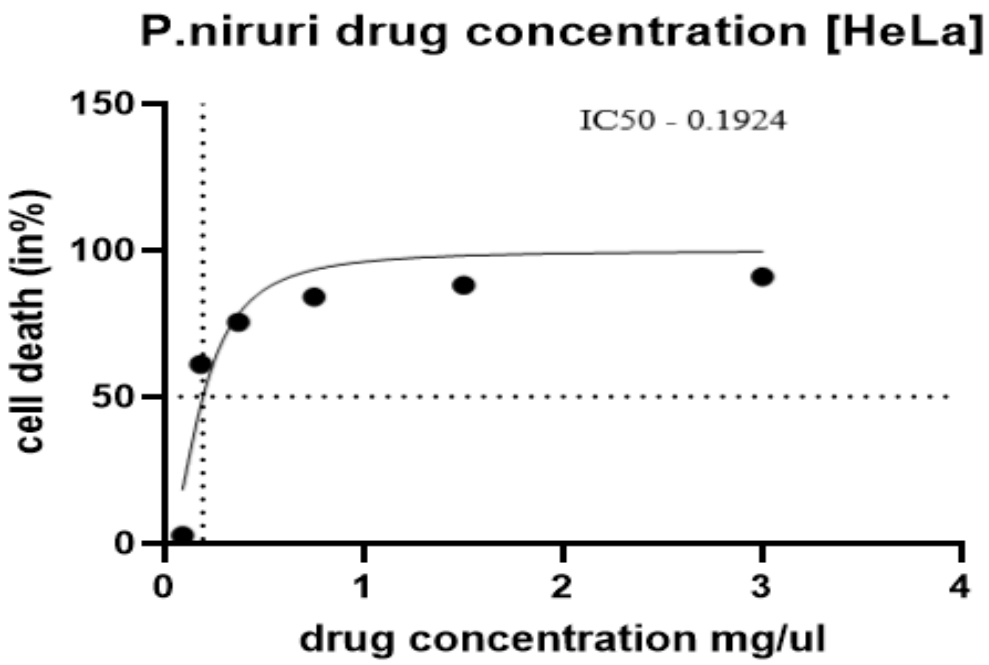

Figure 1 Graph showing anticancer activity of aqueous extract of Phyllanthus niruri L. against HeLa

Table 2 Anticancer activity of aqueous extract of Phyllanthus niruri L. against MDAMB231

Drug concentration µg/µl	OD at 570nm	Cell death in %
Media control	1.17	0
Vehicle control	1.105	5.56
Positive control	0.867	25.9
3	0.061	94.79
1.5	0.132	88.72
0.75	0.147	87.44
0.37	0.174	85.13
0.18	0.229	80.43
0.09	0.289	75.3
0.045	0.330	71.8
0.022	0.379	67.61
0	1.157	1.12

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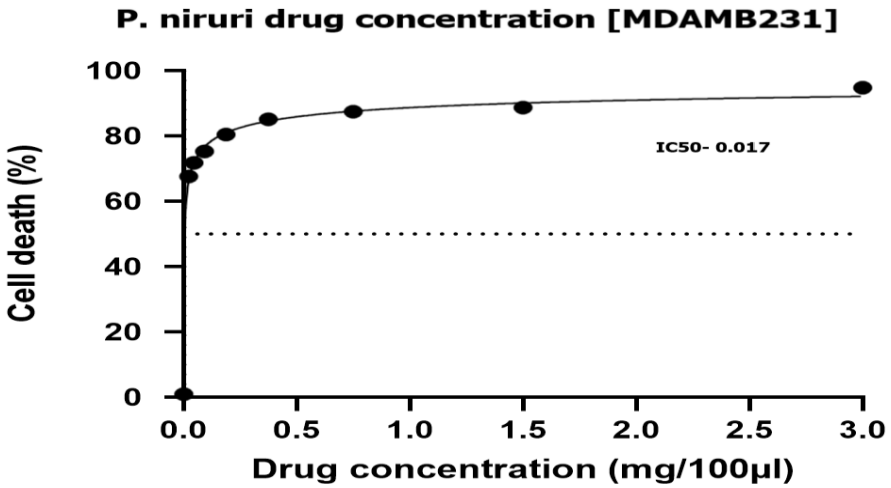

Figure 2 Graph showing anticancer activity of aqueous extract of Phyllanthus niruri L. against MDAMB231

DPPH Assay

The free radical scavenging activity of aqueous extract of *Phyllanthus niruri* was determined by the DPPH assay. The extract showed inhibition of DPPH in a dose dependent manner. The highest concentration 600µg/ml showed inhibition of 50.34% and the lowest activity was found to be 11.42% at 18.75µg/ml concentration as shown in the table 3, which shows a significant antioxidant activity of *Phyllanthus niruri*.

Table 3 Free radical scavenging activity of aqueous extract of Phyllanthus niruri L

Concentration of P. niruri (µg/ml)	Absorbance at 517nm	% Inhibition
600	0.869	50.34
300	0.907	48.17
150	1.030	41.14
75	1.274	27.20
37.5	1.340	23.42
18.75	1.550	11.42

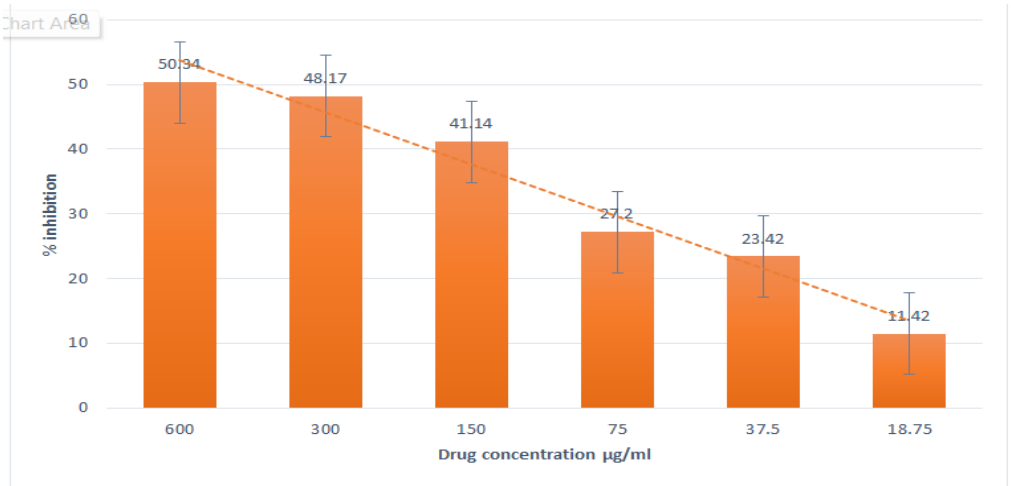

Figure 3 Graph showing free radical scavenging activity of Phyllanthus niruri L.

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GC-MS (GAS CHROMATOGRAPHY-MASS SPECTROMETRY) ANALYSIS

The GC-MS chromatogram of aqueous extract of *Phyllanthus niruri* showed a total of 18 peaks corresponding to bioactive compounds, which were identified by comparing peak, retention time, peak area and height of known compounds. This finding revealed that, different types of phytocompounds were present in the aqueous extract of *Phyllanthus niruri*. 1-Dodecene (3.84%), 1-Tetradecene (4.63%), 2,4-di-tert-butylphenol (11.42%), 1-Tridecene (9.05%), Dodecane 2-methyl (3.28%), 1-Nonadecene (6.32%), 1-Dococene (7.25%), 1-Tricosanol (10.27%), 1-Dodecanol 2-Octyl (6.62%), 1-Decanol 2-hexyl (4.14%), Tetradecyl Di-lactate (3.51%), 2-Methyltetracosane (3.37%), Squalene (6.21%), 1,2,4-Thiadiazol-2-amine N-ethyl (3.11%), 4-Methyldocosane (3.69%), 3-(3-Fluorophenyl)-7-(4-flurophenyl)-4H,6 (3.26%), Ethyl 2-acetanido-3,3,3-trifluoro-2-(o-toluid) (3.43%) and chloromethyl 3-chlorononanoate (6.60%) were predominantly present in aqueous extract of *Phyllanthus niruri.*

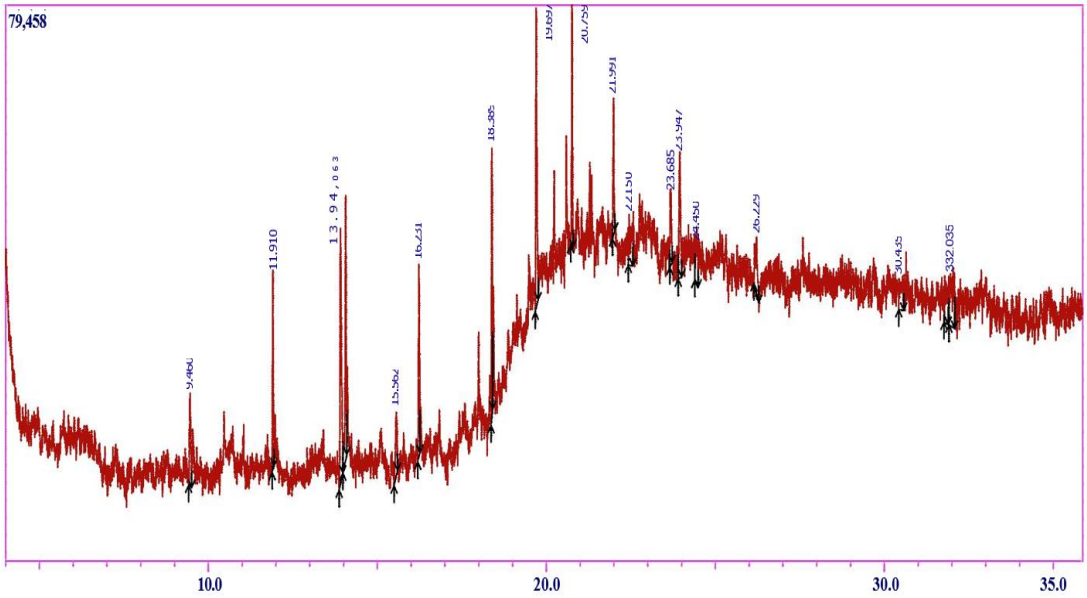

Figure 4 GC-MS chromatogram of the bioactive compounds present in aqueous extract of Phyllanthus niruri

Table 4 Phytocompounds identified in the aqueous extract of Phyllanthus niruri by GC-MS

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	9.460	33769	3.84	13256	3.72	2.55	1-Dodecene
2	11.910	40748	4.63	28444	7.97	1.43	1-Tetradecene
3	13.911	100475	11.42	36438	10.21	2.76	2,4-Di-tert-butylphenol
4	14.063	79601	9.05	37710	10.57	2.11	1-Tridecene
5	15.562	28855	3.28	9103	2.55	3.17	Dodecane, 2-methyl-
6	16.231	55554	6.32	27447	7.69	2.02	1-Nonadecene
7	18.389	63792	7.25	38423	10.77	1.66	1-Docosene
8	19.697	90310	10.27	42840	12.01	2.11	1-Tricosanol
9	20.759	58195	6.62	34412	9.64	1.69	1-Dodecanol, 2-octyl-

10	**21.991**	**36379**	**4.14**	**19449**	**5.45**	**1.87**	**1-Decanol, 2-hexyl-**
11	**22.450**	**30829**	**3.51**	**7204**	**2.02**	**4.28**	**Tetradecyl Di-Lactate**
12	**23.685**	**29682**	**3.37**	**10798**	**3.03**	**2.75**	**2-Methyltetracosane**
13	**23.947**	**54629**	**6.21**	**17945**	**5.03**	**3.04**	**Squalene**
14	**24.450**	**27342**	**3.11**	**5585**	**1.57**	**4.90**	**1,3,4-Thiadiazol-2-amine, N-ethyl-**
15	**26.229**	**32422**	**3.69**	**8246**	**2.31**	**3.93**	**4-Methyldocosane**
16	**30.435**	**28658**	**3.26**	**4389**	**1.23**	**6.53**	**3-(3-Fluorophenyl)-7-(4-fluorophenyl)-4H,6**
17	**31.790**	**30158**	**3.43**	**6577**	**1.84**	**4.59**	**Ethyl 2-acetamido-3,3,3-trifluoro-2-(o-toluid**
18	**32.035**	**58073**	**6.60**	**8539**	**2.39**	**6.80**	**Chloromethyl 3-chlorononanoate**
		879471	**100.00**	**356805**	**100.00**		

Interpretation Of Adme And Docking Results

Molecular docking was performed to evaluate the binding affinity of selected compounds with the target protein 6NM8 (Chain A), a structure associated with the SARS-CoV-2 main protease. The crystal structure of the protein was retrieved from the Protein Data Bank, and docking simulations were carried out using AutoDock or similar docking software. The binding interactions were visualized using a molecular visualization tool (e.g., PyMOL, Discovery Studio), as illustrated in **Figure 5**.

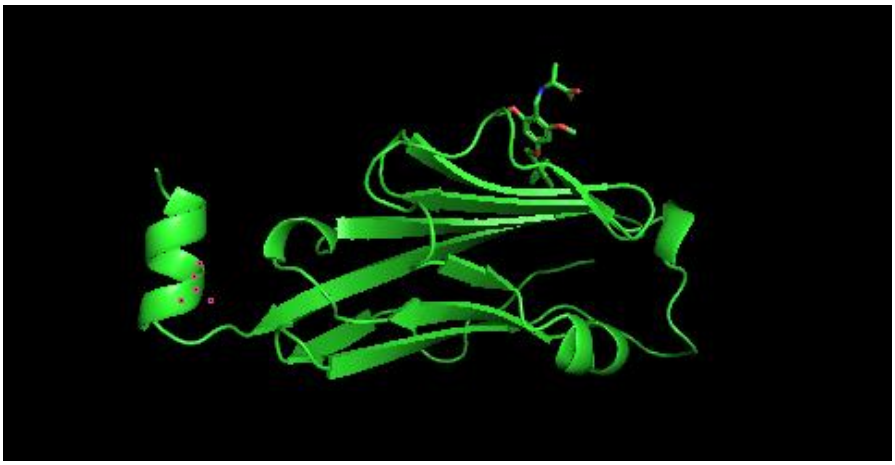

Figure 5. *Molecular docking interaction of selected ligand with 6NM8, Chain A.*

The figure shows the 3D structure of the target protein in green ribbon format, highlighting the active site region where the ligand is bound. The visualization demonstrates the spatial orientation of the ligand within the binding pocket, confirming successful docking interactions.

Figure 1 displays the 3D structure of the target protein (6NM8_A) with the docked ligand positioned within the binding pocket. The protein is represented in a cartoon model (green), showing distinct secondary structures such as α-helices and β-sheets. The ligand is shown in stick representation, indicating its precise orientation within the active site of the protein. This visual highlights the interaction between the ligand and key amino acid residues, suggesting potential hydrogen bonds or hydrophobic contacts that may stabilize the ligand-protein complex.

Docking results revealed that the selected compounds exhibited weak to moderate binding affinities, with binding energy values ranging from -3.1 to -5.1 kcal/mol. These values suggest moderate interactions, though not strong enough to indicate high inhibitory potential. The Root Mean Square Deviation (RMSD) values from docking poses were within acceptable ranges, indicating stable and valid docking conformations. A summary of the docking scores and RMSD values is presented in **Table 5**.

Compound	Solubility	Lipophilicity	Absorption	Best Docking Score (kcal/mol)	Conclusion
4-Methyldocosane	Very Poor	Very High	Low	-3.9	Weak binding and low drug-likeness
1-Docosene	Insoluble	High	Low	-4.1	Moderate docking but low potential
1-Dodecene	Poor	High	Low	-4.3	Moderate docking, weak ADME
1-Nonadecene	Very Poor	Very High	Low	-3.5	Unfavorable drug properties
1-Tetradecene	Poor	High	Low	-4.2	Low pharmacological viability
1-Tridecene	Poor	High	Low	-3.8	Limited potential
2-Hexyl-1-Decanol	Moderate	Acceptable	Moderate	**-5.1**	**Best candidate; promising profile**
2-Octyldodecan-1-ol	Poor	High	Low	-4.7	Moderate docking but poor solubility

DISCUSSION

Phyllanthus species, particularly Phyllanthus niruri, have shown strong anti-proliferative effects against various cancer types, including breast, lung, melanoma, liver, leukemia, and prostate cancers. Known for its anticancer potential and low toxicity, P. niruri has been widely studied, though its specific effects on MDAMB231 and HeLa cell lines remain underexplored [2].

In this study, the aqueous leaf extract of Phyllanthus niruri demonstrated significant cytotoxicity against both HeLa and MDAMB231 cells. At the highest concentration tested, the extract induced 91.03% cell death in HeLa cells (IC_{50} = 0.1924 µg/mL) and 94.79% in MDAMB231 cells (IC_{50} = 0.017 µg/mL), indicating a stronger effect on MDAMB231[7].

To further support its anticancer activity, the extract's antioxidant potential was assessed using a DPPH assay, which showed a dose-dependent increase in free radical scavenging activity. This antioxidant capacity is likely due to the diverse phytochemicals present in the crude extract [5].

GC-MS analysis of the extract revealed the presence of various bioactive compounds, including alkanes, alkenes, phenols, alcohols, thiadiazoles, and terpenes. Among these, phenolic compounds may play a major role in the observed anticancer and antioxidant activities. However, isolating and testing individual compounds is necessary for more targeted insights[1].

Molecular docking and ADMET analysis were carried out to evaluate the interaction of these compounds with the 6NM8 protein. Most compounds showed weak to moderate binding affinities and poor pharmacokinetic properties, particularly in solubility and GI absorption.

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However, 2-Hexyl-1-Decanol stood out with the best binding score (-5.1 kcal/mol), moderate solubility, acceptable lipophilicity, and moderate absorption—making it a strong lead candidate for further drug development. Other compounds, despite moderate docking scores, showed poor drug-likeness and would require structural modifications for further consideration.

Interestingly, no prior studies have reported docking plant-derived compounds with the 6NM8 protein. While various phytochemicals have been docked with other viral or cancer-related targets, 6NM8 remains relatively unexplored in the context of plant-based drug discovery—offering a novel and promising direction for future research.

Discussion

Phyllanthus niruri, renowned for its traditional medicinal use, exhibits potent anti-proliferative effects across various cancers, including breast, lung, melanoma, liver, leukemia, and prostate. Despite its widespread acknowledgment for low toxicity and therapeutic promise, specific effects on *MDAMB231* and *HeLa* cell lines have been underexplored until now.

Our findings reveal that the aqueous leaf extract induces substantial cytotoxicity: **over 91% cell death** in HeLa and MDAMB231 cells at the highest tested concentrations, with IC_{50} values of **0.1924 µg/mL** and **0.017 µg/mL**, respectively. Notably, the extract is more effective against triple-negative breast cancer (MDAMB231), underscoring its potential for targeting aggressive cancer subtypes.

Supporting its anticancer activity, the extract demonstrated a **dose-dependent increase** in antioxidant capacity via DPPH assay, likely attributable to its diverse phytochemicals. GC-MS profiling identified key bioactives—phenols, terpenes, alkanes—that may synergistically contribute to the observed effects. While phenolic compounds are well-known for their anticancer and antioxidant roles, isolating and testing individual constituents is crucial for precise mechanistic insights.

In silico docking and ADMET studies highlighted *2-Hexyl-1-Decanol* as a promising lead: it exhibited the best binding affinity (-5.1 kcal/mol), moderate solubility, and acceptable pharmacokinetic properties. Other phytochemicals showed weaker interactions and poor drug-likeness, indicating a need for structural optimization. Interestingly, this study pioneers docking plant-derived compounds against the *6NM8* protein, a relatively unexplored target opening new avenues in plant-based drug discovery.

Overall, this work positions *Phyllanthus niruri* as a potent source of anticancer agents, especially against resistant triple-negative breast cancer. Its phytochemical diversity and promising molecular interactions warrant further exploration—focusing on isolating active compounds, elucidating mechanisms, and optimizing drug-like properties for clinical translation.

CONCLUSION

The results of this investigation demonstrate that the aqueous leaf extract of *Phyllanthus niruri* possesses both potent anticancer activity particularly against the MDAMB231 breast cancer cell line and strong, dose-dependent antioxidant effects. GC-MS profiling revealed a diverse array of bioactive phytochemicals, among which 2-Hexyl-1-Decanol stood out for its high binding affinity to the 6NM8 target, favorable ADMET characteristics, and drug-like properties. Although several other constituents showed limited pharmacokinetic profiles, these findings lay the groundwork for future structural refinement and isolation efforts. Notably, this work represents one of the first reports of *P. niruri* metabolites engaging the 6NM8 protein, highlighting the plant's promise as a source of novel anticancer leads. To build on these insights, comprehensive in vitro and in vivo validation, along with advanced computational modeling, will be essential to advance the most promising candidates toward clinical development.

List of Abbreviations

- DMSO: Dimethyl sulfoxide

- DMEM/F12: Dulbecco's Modified Eagle Medium/Nutrient Mixture F-12
- DPPH: 2,2-Diphenyl-1-picrylhydrazyl
- FBS: Fetal bovine serum
- GC-MS: Gas chromatography-mass spectrometry
- IC50: Half maximal inhibitory concentration
- MTT: 3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide
- NCCS: National Centre for Cell Science
- PDB: Protein Data Bank
- RMSD: Root mean square deviation
- SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2
- TNBC: Triple-negative breast cancer

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Ethics approval

Not applicable.

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