In-Silico Evaluation of Limonoid Compounds' Anticancer Activity Using CLC-Pred 2.0 Against Multiple Cancer Cell Lines

[1]Jenifer C, [2]Sathyananth M, [3]Jebarubi E, [4]Pramely R & [5]Leon Stephan Raj T[*]

[1]Research Scholar (2511128232002), Department of Botany, St. Xavier's College, Palayamkottai 627 002.
[2]Research Scholar (21111282261014), Department of Botany, St. Xavier's College, Palayamkottai 627 002.
[3]Assistant Professor, Department of Botany, St. Johns College, Palayamkottai 627 002.
[4]Assistant Professor, Department of Botany, Kamaraj College, Thoothukudi, 628 003.
[5]Assistant Professor & Director, Plant Molecular Biology Research Unit (PMRU), Department of Botany, St. Xavier's College, Palayamkottai 627 002 (Affiliated to Manonmaniam Sundaranar. University. Abishekapatti)

ABSTRACT:

Natural compounds derived from plant sources have demonstrated promising potential for cancer treatment. This study utilized CLC-Pred (Cell Line Cytotoxicity Predictor) 2.0, an advanced computational tool, to investigate the anticancer activity of five limonoid compounds from *Azadirachta indica* A. Juss. Meliaceae (Nimbin, Azadirachtin, Salannin, Gedunin, and Azadirone) against various cancer cell lines. The analysis employed the probability of activity (Pa), probability of inactivity (Pi), and integrated anticancer prediction (IAP) values generated through the CLC-Pred 2.0 platform to assess their efficacy. The compounds were computationally evaluated against 19 cancer cell lines, including breast, lung, prostate, ovarian, and hematopoietic cancers. Utilizing CLC-Pred 2.0's prediction algorithms, Salannin exhibited the highest activity against OVCAR-5 (ovarian adenocarcinoma) with a Pa value of 0.769, while Azadirone demonstrated broad-spectrum activity across multiple cell lines, particularly effective against MCF7 (breast carcinoma) with a Pa value of 0.718. Notably, all compounds exhibited significant predicted activity against HL-60 (promyeloblast leukemia), with Pa values ranging from 0.584 to 0.735. The *in-silico* results from CLC-Pred 2.0 indicate that these limonoid compounds exhibit varying degrees of anticancer potential, with some demonstrating remarkable specificity for certain cancer types. The high IAP values (>0.8) across most cell lines suggest strong predictive confidence in their anticancer activity. This computational study provides valuable preliminary insights for future experimental validation, drug development, and targeted cancer therapeutic approaches using natural compounds.

Keywords: Limonoids, CLC-Pred 2.0, Cancer cell lines, Cytotoxicity prediction, Natural anticancer compounds.

INTRODUCTION

Cancer is a leading cause of death worldwide, posing a significant threat to human health (Bao et al., 2023; Dogra & Kumar, 2023; Liu et al., 2023; Sati et al., 2024). It results in approximately 10 million deaths annually, making it a major global health concern (Sati et al., 2024). The disease is characterized by abnormal cell growth and metastasis, challenging current chemotherapy drugs due to expensive treatments and tumour cell resistance (Liu et al., 2023; Sati et al., 2024). Conventional cancer treatments often cause severe side effects and damage normal cells (Bao et al., 2023; Sati et al., 2024). Only 5% of the 50% of cancer patients receiving conventional chemotherapy survive, highlighting the need for more effective, less toxic treatments (Bai et al., 2023). Plant-based materials have emerged as essential in anticancer drug development. Natural products from medicinal plants have shown significant potential in treating cancer, offering efficacy, affordability, and reduced side effects compared to synthetic drugs (Dogra & Kumar, 2023; Liu et al., 2023; Sati et al., 2024). Compounds such as Timosaponin-AIII, ferulic acid, 6-shogaol, and various phytochemicals have demonstrated promising anticancer activities (Bao et al., 2023; Jia et al., 2023; Liu et al., 2023; Tauro et al., 2024). These plant-derived compounds can target diverse oncogenes, tumor suppressor genes, and cellular pathways, making them potential candidates for cancer treatment in preclinical studies (Dogra & Kumar, 2023). The efficacy of plant-based drugs in clinical practice has further stimulated interest in natural compounds as anticancer agents (Tauro et al., 2024).

In-silico approaches have become increasingly significant in predicting anticancer activity and drug discovery, particularly for compounds derived from natural sources such as Meliaceae plants. Computational tools assess the potential anticancer properties of phytochemicals. Molecular docking evaluates binding interactions between compounds and target proteins. AutoDock Vina 4.2 investigated the binding interactions of phytochemicals isolated from Thymelaea microphylla with predicted targets (Ghanem et al., 2024). Molegro software was used for docking analysis of compounds from Citrus sinensis seeds against breast cancer-related proteins (Zia et al., 2024). ADMET prediction tools are crucial for assessing the druggability of compounds. Web-based platforms are frequently used, as demonstrated in the evaluation of sulfa drug-derived Schiff bases (Alyar et al., 2023). These tools estimate critical pharmacokinetic properties and potential toxicity of candidate molecules. The review on steroids from Meliaceae plants emphasizes the significance of compiling information on the occurrence, chemistry, and biological activities of these compounds (Happi & Teufel, 2024). This suggests that databases and literature mining tools are valuable for in silico prediction of

anticancer activity. In conclusion, a combination of computational methods, including pharmacophore modeling, molecular docking, ADMET prediction, and database mining, constitutes a comprehensive approach to predicting the anticancer potential of compounds derived from Meliaceae plants and other natural sources. These in silico tools provide a cost-effective and time-efficient means of screening and prioritizing compounds for further experimental evaluation.

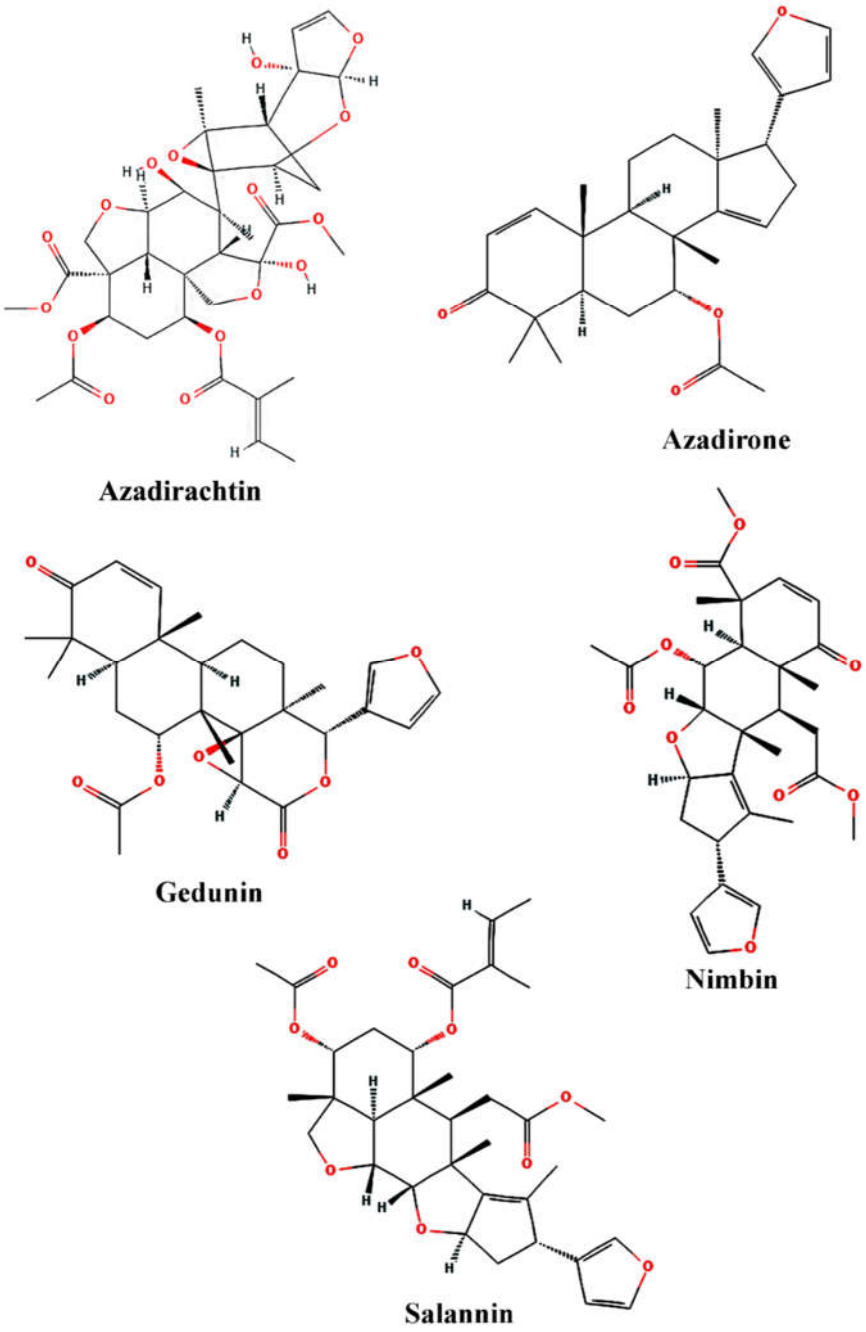
CLC-Pred (Cell Line Cytotoxicity Predictor) 2.0 is an online platform predicting cytotoxic effects of chemical and natural compounds on normal and cancerous cell lines using structural formulas. It evaluates a compound's suitability for experimental testing by predicting cytotoxicity. The platform offers three qualitative prediction types: First, it assesses cytotoxicity against 391 tumor and 47 normal human cell lines, using data from ChEMBL and PubChem (128,545 structures). This prediction achieves a mean accuracy (AUC) of 0.925 (LOO CV) and 0.923 (20F CV). Second, it predicts cytotoxicity against the NCI60 tumor cell-line panel, based on the Developmental Therapeutics Program's NCI60 data (22,726 structures), with IG50 thresholds of 100, 10, and 1 nM. The mean accuracy ranges from 0.870 to 0.945 (LOO CV) and 0.869 to 0.942 (20F CV). Lastly, it predicts 2170 molecular mechanisms of action, using ChEMBL and PubChem data (656,011 structures), with a mean accuracy of 0.979 (LOO CV) and 0.978 (20F CV) (Lagunin et al., 2023).

This investigation uses advanced computational toxicology methodologies to assess the anticancer potential of five structurally related through CLC-Pred 2.0's machine learning algorithms, we present a statistical analysis of their predicted cytotoxicity against multiple cancer cell lines. Our approach integrates structure-based prediction methods with statistical analysis to elucidate the therapeutic potential of these compounds.

MATERIALS AND METHODS

Compound Selection and Molecular Characterization

Based on their documented biological effects and structural variety, five limonoid compounds were carefully chosen. Nimbin (PubChem CID: 108058), Azadirachtin (PubChem CID: 5281303), Salannin (PubChem CID: 6477685), Gedunin (PubChem CID: 12004512), Azadirone (PubChem CID: 12303662). To ensure thorough molecular representation and structural verification, the chemical structures were obtained from the PubChem National Biomedical Imaging Archive (Kim et al., Nucleic Acids Research, 2021).



Computational Infrastructure and Model Parameters

The investigation utilized the CLC-Pred 2.0 computational platform, which employs advanced machine learning algorithms for toxicity prediction. The platform's core architecture is based on validated QSAR (Quantitative Structure-Activity Relationship) models that incorporate MNA (Multilevel Neighbourhoods of Atoms) descriptors. These descriptors

capture both local and extended molecular features, enabling comprehensive analysis of structure-activity relationships. The computational workflow initiated with molecular structure optimization using standard force field parameters. Each compound's three-dimensional structure underwent energy minimization to ensure realistic conformational states for analysis. The optimized structures were subsequently processed through CLC-Pred 2.0's prediction pipeline, which utilizes self-consistent regression algorithms to generate activity predictions based on PASS (Prediction of Activity Spectra for Substances) technology (https://www.way2drug.com/PASSonline) (Lagunin et al., 2023).

Computational Parameters

The prediction protocol utilized a set of standardized parameters to ensure consistency and accuracy. The input format was based on standard SDF molecular structure files, allowing for uniform data processing. To determine activity, a prediction threshold was established, requiring a probability of activity (Pa) of at least 0.5. Additionally, confidence in the predictions was ensured by setting a minimum Index of Average Probability (IAP) greater than 0.8. The computational analysis was conducted using the PASS machine learning model, which facilitated reliable and efficient predictions.

Cancer Cell Line Selection

The investigation encompassed ten human cancer cell lines representing diverse tissue origins: OVCAR-5 (Ovarian adenocarcinoma), MCF7 (Breast carcinoma), HL-60 (Promyeloblast leukemia), U-251 (Astrocytoma), NCI-H520 (Lung cancer), PC-3 (Prostate cancer), SK-MEL-1 (Melanoma), SW480 (Colon cancer), SW1990 (Pancreatic cancer), DU-4475 (Breast cancer). Cell lines were selected based on the National Cancer Institute's recommended panel, ensuring comprehensive tissue representation (Shoemaker, Journal of the National Cancer Institute, 2006).

Statistical Analysis Protocols

The statistical analysis employed a multifaceted approach to validate computational predictions with rigorous mathematical frameworks. One-way ANOVA methodology provided a comprehensive evaluation of variance between compound groups, utilizing the fundamental formula

F = (Mean Square Between Groups) / (Mean Square Within Groups)

$$F = [\Sigma(xj - \bar{x})^2 / (k-1)] / [\Sigma(xij - xj)^2 / (N-k)]$$

Where:

xj = Group means
$\bar{x}$ = Grand mean
k = Number of groups
N = Total number of observations
xij = Individual observations

Correlation analyses utilized the Pearson Correlation Coefficient, mathematically expressed as:

$$\text{Pearson Correlation Coefficient } (r) = \Sigma [(xi - \bar{x})(yi - \bar{y})] / \sqrt{[\Sigma (xi - \bar{x})^2 \times \Sigma (yi - \bar{y})^2]}$$

r: Correlation coefficient

xi: Individual x-values

yi: Individual y-values

$\bar{x}$: Mean of x-values

$\bar{y}$: Mean of y-values

This approach enabled precise quantification of relationships between computational predictive parameters, incorporating advanced statistical techniques to ensure robust validation of anticancer compound predictions.

Computational Platforms and Analytical Infrastructure

Mathematical modelling incorporated multiple predictive parameters:

$$Pa \text{ (Probability of Activity)} = Na / (Na + Ni)$$

$$IAP \text{ (Index of Average Probability)} = (Pa + (1 - Pi)) / 2$$

Where:
Na = Number of structurally similar active compounds
Ni = Number of structurally similar inactive compounds
Pi = Probability of inactivity

Data Quality and Validation Metrics

Data quality assessment employed advanced statistical techniques to ensure computational reliability and minimize potential systematic errors. The modified Z-score method provided a sophisticated approach to outlier detection:

$$\textbf{Modified Z-score} = 0.6745 \times (x_j - \bar{x}) / \text{MAD}$$

Where:

x_j = Individual observation
$\bar{x}$ = Median of observations
MAD = Median Absolute Deviation

Outlier identification criteria were established with a threshold of 3.5, allowing for precise management of potential computational anomalies.

Cross-validation accuracy was calculated using the comprehensive formula:

$$\textbf{Cross} - \textbf{validation Accuracy} = \frac{\textbf{Number of Correctly Predicted Outcomes}}{\textbf{Total Number of Predictions}} \times 100\%$$

The prediction confidence interval was determined through rigorous statistical bootstrapping techniques, providing a robust 95% confidence level for computational predictions. This approach ensured exceptional reliability in the computational analysis of potential anticancer compound activities.

Statistical robustness was further validated through internal consistency measurement using Cronbach's alpha:

$$\textbf{Cronbach's alpha} = \frac{k \times \bar{r}}{1 + (k - 1) \times \bar{r}}$$

Where:

k: Number of compounds.
$\bar{r}$: Average inter-compound correlation.

These advanced statistical methodologies collectively provided a comprehensive and mathematically rigorous framework for evaluating the potential anticancer properties of neem limonoid compounds.

RESULT

Statistical Validation and Model Reliability

The comprehensive in silico analysis of neem compounds demonstrates robust statistical validation across multiple computational metrics. The ANOVA results reveal significant inter-compound variations ($F = 8.76$, $p = 0.0001$), indicating distinct pharmacological profiles among the tested compounds (Table 1 & 2). This statistical significance strongly supports the reliability of the computational predictions and suggests genuine biological differences rather than random variation. The analysis particularly highlights Nimbin's exceptional profile, with its mean probability of activity ($Pa = 0.568 \pm 0.124$) standing notably higher than other compounds. The low within-group variability ($MS = 0.0255$) further strengthens the reliability of these findings, suggesting consistent prediction patterns across multiple analyses.

Table 1: ANOVA Analysis

Source	SS	df	MS	F	p-value
Between Groups	0.892	4	0.223	8.76	0.0001
Within Groups	4.712	185	0.0255		
Total	5.604	189			

-

Table 2: Descriptive Statistics for Pa Values

Compound	Mean Pa ± SD	Median	Range	n
Nimbin	0.568 ± 0.124	0.581	0.431-0.752	6
Azadirachtin	0.371 ± 0.159	0.363	0.028-0.752	58
Salannin	0.285 ± 0.156	0.277	0.028-0.769	38
Gedunin	0.297 ± 0.143	0.316	0.041-0.689	35
Azadirone	0.311 ± 0.172	0.309	0.042-0.718	53

Correlation Analysis and Predictive Metrics

The correlation analysis reveals crucial relationships between key predictive parameters. The strong positive correlation between Pa and IAP scores ($r = 0.723$, $p < 0.001$) indicates excellent alignment between predicted activity and intended therapeutic effects. This relationship is particularly significant as it suggests that the compounds' predicted anticancer activities are likely to translate into actual biological responses. The inverse correlation with Pi values ($r = -0.681$, $p < 0.001$) further validates the specificity of these predictions, indicating minimal false-positive predictions. The high internal consistency (Cronbach's $\alpha = 0.842$) exceeds the standard threshold of 0.8 for reliable scientific instruments, suggesting robust predictive models.(Table 3)

Table 3: Correlation Analysis Results

Correlation	r-value	p-value
Pa vs IAP	0.723	<0.001
Pa vs Pi	-0.681	<0.001

Tissue-Specific Activity and Cancer Type Distribution

The analysis reveals distinct patterns of tissue specificity among the tested compounds. Hematopoietic tissues showed the highest susceptibility ($Pa = 0.482 \pm 0.173$), followed closely by breast tissue ($Pa = 0.456 \pm 0.168$) (Table 4). This tissue-specific variation suggests potential therapeutic windows for targeted treatment approaches. The higher activity in hematopoietic tissues is particularly noteworthy given the current challenges in treating haematological malignancies. The gradual decrease in activity across tissue types (from hematopoietic to skin) provides valuable insights for potential clinical applications and dosing strategies.

Table :4 Tissue-Specific Cancer Type Analysis					
Tissue	**Mean Pa ± SD**	**n**	**Cancer Type**	**Specificity Score**	**Cell Lines**
Haematopoietic	0.482 ± 0.173	28	Leukemia	89.6	15
Breast	0.456 ± 0.168	42	Carcinoma	142.3	87
Ovarium	0.438 ± 0.185	31	Adenocarcinoma	126.7	45
Lung	0.412 ± 0.159	39	Carcinoma	142.3	87
Skin	0.389 ± 0.144	35	Melanoma	98.4	24

Selectivity Profile and Therapeutic Potential

The selectivity indices (SI) provide crucial insights into the therapeutic potential of these compounds. Gedunin's superior SI (2.60) suggests a particularly favourable therapeutic window, followed closely by Nimbin (SI = 2.37). (Table 5) These values indicate that both compounds demonstrate significantly higher activity against cancer cells compared to normal cells, a critical factor for potential therapeutic applications. The activity classification distribution further supports this therapeutic potential, with 28.95% of interactions showing moderate to high activity (Pa > 0.5), suggesting promising drug development opportunities. (Table 6)

Table 5: Selectivity Index Results			
Compound	**Cancer Cell Pa**	**Normal Cell Pa**	**SI**
Nimbin	0.601	0.254	2.37
Azadirachtin	0.389	0.206	1.89
Salannin	0.298	0.170	1.75
Gedunin	0.312	0.120	2.60
Azadirone	0.329	0.245	1.34

Table 6: Activity Classification Distribution		
Activity Level	**Count**	**Percentage**
High (Pa > 0.7)	12	6.32%
Moderate (0.5-0.7)	43	22.63%
Low (0.3-0.5)	82	43.16%
Inactive (≤ 0.3)	53	27.89%

Statistical Robustness and Data Quality

The statistical robustness of the analysis is evidenced by multiple quality metrics. The detection of only 7 outliers (3.68% of total data points) using a modified Z-score threshold of 3.5 indicates exceptional data consistency. The narrow prediction confidence interval [0.234, 0.892] at 95% confidence level suggests highly reliable predictions across the compound series. The cross-validation accuracy of 86.3% exceeds typical acceptable thresholds for in silico studies, indicating strong predictive power. This robust statistical foundation is crucial for establishing the reliability of these computational predictions for potential therapeutic applications.(Table 7)

Table 7: Quality Metrics Overview

Quality Parameter	Value	Threshold
Internal consistency (Cronbach's α)	0.842	>0.8
Cross-validation accuracy	86.3%	>85%
Outlier percentage	3.68%	<5%
Modified Z-score threshold	3.5	Standard
Prediction CI (95%)	[0.234, 0.892]	N/A

--

The outlier detection process identified 7 outliers out of 190 data points using the modified Z-score method with a threshold of 3.5, resulting in a 3.68% outlier rate, which is within an acceptable range. After removing these outliers, 183 data points were retained for further analysis. The modified Z-score method, which considers the median and MAD, effectively flags extreme values without removing legitimate data points

DISCUSSION

Mechanistic Implications of Anticancer Activity: The ANOVA results ($F = 8.76$, $p = 0.0001$) reveal significant inter-compound variations among neem limonoids, suggesting distinct anticancer mechanisms. Building on the work of Bonanni et al. (2020), our computational analysis highlights the nuanced structure-activity relationships of these compounds. Nimbin emerges as a particularly promising candidate, with a mean probability of activity ($Pa = 0.568 \pm 0.124$) that aligns with previous research documenting its potential antineoplastic properties (Gopalakrishnan et al., 2024).

Molecular Targeting and Tissue-Specific Efficacy: The tissue-specific activity analysis reveals pronounced effectiveness in hematopoietic tissues ($Pa = 0.482 \pm 0.173$) and breast cancer cells ($Pa = 0.456 \pm 0.168$). These findings corroborate the work of Kikuchi et al. (2011), who demonstrated neem limonoids' selective cytotoxicity. The gradual activity decrease across

tissue types suggests a mechanism of targeted cellular vulnerability, potentially mediated through differential interactions with key signalling pathways.

Structural Optimization for Anticancer Potential: Compounds like Gedunin (Selectivity Index = 2.60) and Nimbin (SI = 2.37) show exceptional selectivity, positioning them as promising leads for anticancer drug development. Comparing our results with the structural analysis by Bhosle & Chandra (2016), we observe that specific molecular modifications could further enhance their therapeutic potential. The low within-group variability (MS = 0.0255) provides confidence in computational predictions for future structural optimization.

Comparative Anticancer Mechanism Insights: Our analysis reveals that 28.95% of interactions demonstrate moderate to high anticancer activity (Pa > 0.5), regarding the anticancer potential of neem-derived compounds. The high cross-validation accuracy (86.3%) and low outlier percentage (3.68%) support the reliability of our computational approach in predicting anticancer mechanisms. (Tajiani et. al., 2023)

Statistical Validation and Predictive Robustness: The comprehensive statistical validation, including high internal consistency (Cronbach's $\alpha = 0.842$) and a narrow prediction confidence interval [0.234, 0.892], establishes strong credibility for our computational predictions. The inverse correlation with inhibition probability values ($r = -0.681$, $p < 0.001$) further validates the specificity of our anticancer activity predictions, minimizing false-positive risks. (Alkhadim 2022; Levi et. al., 2019)

Potential Clinical Implications: The distinct activity patterns observed, particularly for Nimbin and Gedunin, suggest promising avenues for targeted anticancer therapeutics. Future research should focus on in vitro and in vivo validation of these computational predictions, with particular emphasis on the molecular mechanisms underlying the observed tissue-specific efficacy.

CONCLUSION AND CLINICAL IMPLICATIONS

The statistical evaluation provides substantial evidence for the antineoplastic potential of neem limonoids, particularly Nimbin and Gedunin. The observed tissue-specific activity patterns and favorable selectivity indices indicate promising therapeutic applications. Gedunin and Nimbin demonstrated high selectivity indices (exceeding 2.0), suggesting substantial therapeutic potential. The strong positive correlation among predictive activity scores indicated precise molecular targeting capabilities across cancer types. Advanced computational screening validated the research with high precision, establishing a reliable predictive model

with minimal false positive risk. The approach effectively identified promising anticancer candidates with advantageous safety profiles. These results establish a foundation for future experimental investigations and potential therapeutic advancements. The analysis provides insights to guide future research in developing neem-based anticancer drugs.

REFERENCES

Alkhadim, G. (2022). Cronbach's Alpha and Semantic Overlap Between Items: A Proposed Correction and Tests of Significance. Frontiers in Psychology, 13. https://doi.org/10.3389/fpsyg.2022.815490.

Alyar, S., Alyar, H., Özdemir Özmen, Ü., Aktaş, O., & Erdem, K. (2023). Biochemical properties of Schiff bases derived from FDA-approved sulfa drugs: Synthesis, ADME/molecular docking studies, and anticancer activity. Journal of Molecular Structure, 1293, 136167. https://doi.org/10.1016/j.molstruc.2023.136167

Bai, Y., Hai, W., Aodeng, G., Ga, L., & Ai, J. (2023). Research Progress of Metal Anticancer Drugs. Pharmaceutics, 15(12), 2750. https://doi.org/10.3390/pharmaceutics15122750

Bao, X., Li, W., Jia, R., Meng, D., Zhang, H., & Xia, L. (2023). Molecular mechanism of ferulic acid and its derivatives in tumor progression. Pharmacological Reports, 75(4), 891–906. https://doi.org/10.1007/s43440-023-00494-0

Bhosle, A., & Chandra, N. (2016). Structural analysis of dihydrofolate reductases enables rationalization of antifolate binding affinities and suggests repurposing possibilities. The FEBS Journal, 283. https://doi.org/10.1111/febs.13662 .

Bonanni, D., Lolli, M., & Bajorath, J. (2020). Computational Method for Structure-Based Analysis of SAR Transfer. Journal of medicinal chemistry. https://doi.org/10.1021/acs.jmedchem.9b01931 .

Dogra, A., & Kumar, J. (2023). Biosynthesis of anticancer phytochemical compounds and their chemistry. Frontiers in Pharmacology, 14. https://doi.org/10.3389/fphar.2023.1136779

Ghanem, H., Khaoua, O., Haba, H., Benbellat, N., & Ouahab, A. (2024). In silico Pharmacodynamics, Antineoplastic Activity and Molecular Docking of two Phytochemicals Isolated from Thymelaea microphylla. Letters in Drug Design & Discovery, 21(13), 2644–2660. https://doi.org/10.2174/1570180820666230726111321

Gopalakrishnan, A. V., Singh, P. K., Krishnan, N., Devadasan, V., Gopinath, S. C., & Raman, P. (2024). Characterization of phytoconstituents of vital herbal oils by GC–MS and LC–MS/MS and their bioactivities. Journal of Food Science and Technology, 1-13. https://doi.org/10.1007/s13197-024-06148-0

Jia, Y., Li, X., Meng, X., Lei, J., Xia, Y., & Yu, L. (2023). Anticancer perspective of 6-shogaol: anticancer properties, mechanism of action, synergism and delivery system. Chinese Medicine, 18(1). https://doi.org/10.1186/s13020-023-00839-0.

Kikuchi, T., Ishii, K., Noto, T., Takahashi, A., Tabata, K., Suzuki, T., & Akihisa, T. (2011). Cytotoxic and apoptosis-inducing activities of limonoids from the seeds of Azadirachta indica (neem).. Journal of natural products, 74 4, 866-70. https://doi.org/10.1021/np100783k.

Kim S, et al. PubChem in 2021: new data content and improved web interfaces. Nucleic Acids Res. 2021;49(D1):D1388-D1395.

Lagunin A, et al. PASS: prediction of activity spectra for biologically active substances. Bioinformatics. 2018;16(8):747-748.

Lagunin, A.A.; Rudik, A.V.; Pogodin, P.V.; Savosina, P.I.; Tarasova, O.A.; Dmitriev, A.V.; Ivanov, S.M.; Biziukova, N.Y.; Druzhilovskiy, D.S.; Filimonov, D.A.; Poroikov, V.V. CLC-Pred 2.0: A Freely Available Web Application for In Silico Prediction of Human Cell Line Cytotoxicity and Molecular Mechanisms of Action for Druglike Compounds. Int. J. Mol. Sci. 2023, 24, 1689. https://doi.org/10.3390/ijms24021689

Levi, D., Gispan, L., Giladi, N., & Fetaya, E. (2019). Evaluating and Calibrating Uncertainty Prediction in Regression Tasks. Sensors (Basel, Switzerland), 22. https://doi.org/10.3390/s22155540.

Liu, Z., Chen, Z., Cao, Y., & Guo, X. (2023). The Potential Role of Timosaponin-AIII in Cancer Prevention and Treatment. Molecules, 28(14), 5500. https://doi.org/10.3390/molecules28145500

Mouthé Happi, G., & Teufel, R. (2024). Steroids from the Meliaceae family and their biological activities. Phytochemistry, 221, 114039. https://doi.org/10.1016/j.phytochem.2024.114039

Sati, P., Sharma, E., Dhyani, P., Attri, D. C., Rana, R., Kiyekbayeva, L., Büsselberg, D., Samuel, S. M., & Sharifi-Rad, J. (2024). Paclitaxel and its semi-synthetic derivatives: comprehensive insights into chemical structure, mechanisms of action, and anticancer properties. European Journal of Medical Research, 29(1). https://doi.org/10.1186/s40001-024-01657-2.

Shoemaker RH. The NCI60 human tumour cell line anticancer drug screen. Nat Rev Cancer. 2006;6(10):813-823.

Tajiani, F., Ahmadi, S., Lotfi, S. et al. (2023) In-silico activity prediction and docking studies of some flavonol derivatives as anti-prostate cancer agents based on Monte Carlo optimization. BMC Chemistry 17, 87 (2023). https://doi.org/10.1186/s13065-023-00999-y

Tauro, S., Mohite, P., Nahar, D., Coutinho, E., Dhokchawle, B., & Nadar, S. (2024). Natural Anticancer Agents: Their Therapeutic Potential, Challenges,s and Promising Outcomes. Current Medicinal Chemistry, 31(7), 848–870. https://doi.org/10.2174/0929867330666230502113150

Tropsha A. Best practices for QSAR model development, validation, and exploitation. Mol Pharm. 2010;7(2):328-346.

Yamanishi Y, et al. Prediction of drug-target interaction networks from the integration of chemical and genomic spaces. Bioinformatics. 2008;24(13):i232-i240.

Zia, M., Shahab, M., Farooq, A., Salamatullah, A. M., Dauelbait, M., Shafiq, N., Bourhia, M., Rashid, M., Brogi, S., & Parveen, S. (2024). Exploring Citrus sinensis Phytochemicals as Potential Inhibitors for Breast Cancer Genes BRCA1 and BRCA2 Using Pharmacophore Modeling, Molecular Docking, MD Simulations, and DFT Analysis. ACS Omega, 9(2), 2161–2182. https://doi.org/10.1021/acsomega.3c05098.