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Original Research Paper

EVALUATION OF THE ANTI-ANGIOGENIC EFFECT OF COREOPSIS GRANDIFLORA IN HEPATOCELLULAR CARCINOMA VIA VEGFR2 REGULATION

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ABSTRACT

Background: Hepatocellular carcinoma (HCC) remains a leading cause of cancer-related mortality worldwide, with limited therapeutic efficacy and frequent resistance to current targeted agents. VEGFR2-mediated angiogenesis plays a central role in HCC progression, making it a clinically validated therapeutic target. Natural products are increasingly explored as safer, multitarget alternatives to synthetic inhibitors.

Aim: This study aimed to systematically evaluate phytochemicals from *Coreopsis grandiflora* as potential VEGFR2 inhibitors using an integrative computational pharmacology approach.

Methods: Twenty-five phytochemicals were retrieved from the IMPPAT database and subjected to molecular docking against VEGFR2 (PDB ID: 2OH4). Top-scoring compounds were further screened using Lipinski's Rule of Five, SwissADME pharmacokinetics, BOILED-Egg gastrointestinal and blood–brain barrier profiling, and PASS biological activity prediction. Target validation was performed through transcriptomic expression analysis and Kaplan–Meier survival assessment. In vitro cytotoxicity of the extract was evaluated in HepG2 cells using the MTT assay.

Results: Docking analysis identified several compounds with strong binding affinity, with the highest score reaching −7.8 kcal/mol. Interaction profiling revealed stable engagement within the ATP-binding pocket of VEGFR2, involving key catalytic residues. Eighteen compounds satisfied drug-likeness criteria, and selected candidates demonstrated favourable pharmacokinetic properties and predicted antineoplastic activity. VEGFR2 was overexpressed in HCC tissues, and higher expression levels were associated with poorer overall survival. Additionally, the extract exhibited dose-dependent cytotoxicity in HepG2 cells with an IC_{50} of 103.36 µg/mL.

Conclusion: This study identifies *Coreopsis grandiflora* as a promising botanical source of potential VEGFR2 inhibitors in HCC. By integrating docking, pharmacokinetic screening, biological prediction, and clinical target validation, the findings provide a rational foundation for further experimental investigation into plant-derived anti-angiogenic therapies.

Keywords: Cancer, disease, genetics, green product, drug

INTRODUCTION:

Hepatocellular carcinoma (HCC) is the most common primary malignancy of the liver and ranks as the third leading cause of cancer-related deaths globally (Chidambaranathan-Reghupaty S et al. 2021). Despite curative treatments like liver transplantation, surgical resection, and thermal ablation, recurrence rates remain high (up to 70% within five years), and the prognosis for HCC remains poor due to late-stage diagnosis, high recurrence rates, and limited treatment efficacy (Nevola R et al 2023). Recent developments in the treatment of hepatocellular carcinoma (HCC) have led to a multifaceted approach that extends beyond conventional therapies. Emerging strategies include immunotherapies, RNA-based therapeutics—particularly the use of small interfering RNA (siRNA) as a novel modality for HCC management—, and the development of prognostic nomograms for patients with double-negative HCC (AFP- and DCP-negative) to guide clinical decisions following local ablation (Mamdouh S et al. 2024, Manivannan HP et al. 2025, Xu JX et al. 2023). In addition, new molecular insights highlight the role of the transcription factor MAZ, which promotes angiogenesis via NEIL3-mediated aerobic glycolysis, thereby contributing to tumour progression (Zhang F et al. 2024).

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The therapeutic targets explored in hepatocellular carcinoma (HCC) to date include the RAF/MEK/ERK pathway, PD-1/PD-L1 immune checkpoints, c-MET receptor tyrosine kinase, and VEGF/VEGFR signaling, which are inhibited by sorafenib, nivolumab, cabozantinib, and bevacizumab, respectively (Wang Y et al. 2023, Fulgenzi CAM et al. 2022, Gordan JD et al. 2020). Inhibition of the RAF/MEK/ERK pathway by BRAF inhibitors has shown significant efficacy in cancers such as melanoma, colorectal cancer, and lung cancer, achieving tumor regression in 50–60% of patients, while combination therapy with BRAF and MEK inhibitors further improves progression-free survival and reduces resistance compared with monotherapy (Song Y et al. 2022). However, these treatments are frequently associated with major cardiovascular adverse effects—including heart failure, venous thromboembolism, and myocardial infarction—as well as more common toxicities such as hand–foot skin reaction, diarrhea, hypertension, and fatigue (Guha A et al. 2021). Immunotherapies targeting the PD-1/PD-L1 axis, such as nivolumab and atezolizumab, enhance antitumor immunity but provide durable responses only in a subset of patients and may lead to immune-related toxicities such as colitis, hepatitis, and endocrinopathies (Han Y et al. 2020). Similarly, inhibition of the c-MET receptor by cabozantinib has been used to block tumor invasion and metastasis; however, its clinical application is limited by gastrointestinal disturbances, hypertension, and hepatotoxicity (Murciano-Goroff YR et al. 2025, Chen SY et al. 2024).

Despite these therapeutic challenges, targeting the VEGF/VEGFR2 pathway represents a promising approach, as VEGFR2 is the principal mediator of angiogenesis in HCC and is strongly correlated with tumor progression, metastasis, and poor prognosis (Mahaki H et al. 2025). While synthetic VEGFR2 inhibitors such as bevacizumab and ramucirumab are in clinical use, their benefits are offset by resistance, bleeding risks, hypertension, and high treatment costs (Wang L et al. 2024). Medicinal plants have long served as a valuable reservoir of bioactive compounds with diverse pharmacological properties, including anticancer activity. One promising plant in this regard is *Coreopsis grandiflora* (*C. grandiflora*), a perennial herb widely cultivated across various regions of Asia for both ornamental and medicinal purposes (Wu ZY et al. 2011). To address these challenges, our study introduces the novel exploration of phytochemicals from *Coreopsis grandiflora* as potential natural VEGFR2 inhibitors. This study aims to assess the anti-cancer potential of *C. grandiflora* against HCC through phytochemical screening, in vitro cytotoxicity assays, and molecular docking approaches. By evaluating the bioactive constituents and their interactions with molecular targets relevant to hepatocarcinogenesis, this work seeks to reveal new plant-based leads for future drug development.

METHODOLOGY:

Selection of Phytochemicals:

The initial phase of this study focused on the systematic identification and selection of phytochemicals present in *C. grandiflora*, a medicinal plant known for its therapeutic potential. To ensure the selection of compounds was comprehensive, evidence-based, and relevant to Indian medicinal plant systems, we utilized the IMPPAT (Indian Medicinal Plants, Phytochemistry and Therapeutics) database (https://cb.imsc.res.in/imppat/), a curated online resource developed for phytochemical and pharmacological data mining. The IMPPAT database integrates information on over 1700 Indian medicinal plants and more than 9500 phytochemicals. Using *C. grandiflora* as the search keyword, we retrieved a comprehensive list of phytochemicals reported in the plant. Each compound was manually screened to ensure relevance and non-redundancy. Structural details, including chemical names, molecular weights, SMILES notations, and PubChem Compound Identifiers (CIDs), were extracted. A total of 25 distinct phytochemicals were selected based on their reported bioactivity, structural diversity, and potential for interaction with cancer-related targets.

Ligand and Target Preparation:

Following the selection of 25 phytochemicals from *C. grandiflora*, the next step involved preparing the ligands for molecular docking analysis.

The 3D chemical structures of all selected phytochemicals were retrieved from the PubChem database (https://pubchem.ncbi.nlm.nih.gov/), an open-access repository that provides accurate and standardized structural information for small molecules. The files were downloaded in Structure Data File (.SDF) format, which preserves molecular geometry and atom connectivity.

The downloaded .SDF files were batch imported into Open Babel, a widely used open-source chemical toolbox integrated within PyRx (Python Prescription), a virtual screening software that incorporates AutoDock Vina. Each structure was then converted into Protein Data Bank, Partial Charge & Torsion Format (PDBQT), which is required for compatibility with the docking engine. Energy minimization of ligands using the Universal Force Field (UFF) to obtain an optimized, low-energy conformation, identification and assignment of rotatable bonds to allow conformational flexibility during docking were undertaken during the conversion process.

The molecular target selected for docking was Vascular Endothelial Growth Factor Receptor 2 (VEGFR2), a receptor tyrosine kinase encoded by the KDR gene. VEGFR2 plays a central role in angiogenesis, tumor growth, and metastatic progression, and is significantly upregulated in hepatocellular carcinoma (HCC), making it a promising therapeutic target. The crystal structure of the target protein, Vascular Endothelial Growth Factor Receptor 2 (VEGFR2), was retrieved from the Protein

Data Bank (PDB ID: 2OH4) in .PDB format. This standardized protocol ensured both ligands and receptor were properly formatted and chemically prepared for molecular docking simulations using AutoDock Vina integrated within PyRx.

Molecular Docking:
Molecular docking was performed using PyRx (version 0.8), a virtual screening software that integrates AutoDock Vina as its docking engine. The docking simulations aimed to predict the binding affinity and orientation of phytochemicals within the active site of the VEGFR2 protein. The grid box was strategically centered on the active site of VEGFR2 to ensure appropriate coverage of the binding pocket, with grid center coordinates set at X = 1.687599, Y = 34.131873, and Z = 17.159538. All 25 phytochemicals from *C. grandiflora*, along with a reference chemotherapeutic agent Carboplatin, were docked to the receptor. The docking algorithm evaluates the conformational space of each ligand and assigns a binding affinity score (in kcal/mol), which reflects the predicted strength of the ligand-receptor interaction.

Binding Affinity and Interaction Analysis:
Following docking, the binding affinity values for each compound were recorded. Compounds with lower (i.e., more negative) binding energies were considered to have stronger predicted binding to VEGFR2. To further evaluate the quality and specificity of these interactions, ligand-protein complexes were subjected to visual inspection using PyMOL and Discovery Studio Visualizer. These tools allowed for detailed analysis of the molecular interactions such as hydrogen bonds, hydrophobic contacts, pi-pi stacking, and van der Waals interactions. Visual assessment helped identify key amino acid residues within the VEGFR2 binding site involved in stabilizing ligand binding and provided insight into the structural compatibility of each phytochemical.

Drug-Likeness Prediction:
The drug-likeness of each docked compound was assessed using the SwissADME online tool (http://www.swissadme.ch/), which applies Lipinski's Rule of Five as a filter for oral bioavailability. The rule considers essential physicochemical properties such as molecular weight, lipophilicity (log P), number of hydrogen bond donors and acceptors, and topological polar surface area (TPSA). Compounds that violated more than one of Lipinski's parameters were deemed less likely to possess favorable pharmacokinetic properties and were excluded from subsequent analyses. This step helped narrow down the ligand set to those with better potential as drug-like molecules.

Pharmacokinetics and BBB Permeability:
Pharmacokinetic profiling of the filtered compounds was also conducted using SwissADME, specifically the BOILED-Egg model, which graphically predicts gastrointestinal (GI) absorption and blood-brain barrier (BBB) permeability. This model relies on key physicochemical properties to plot each compound on a 2D graph, enabling easy interpretation of their likely bioavailability. For this study, compounds with high GI absorption and no BBB permeability were prioritized, as ideal candidates for hepatocellular carcinoma therapy should exhibit systemic availability without crossing the blood-brain barrier, thereby minimizing potential central nervous system (CNS) side effects.

Biological Activity Prediction:
To predict the functional relevance of the selected compounds, biological activity prediction was performed using the PASS (Prediction of Activity Spectra for Substances) web server (http://www.way2drug.com/PASSOnline/). This tool estimates the probability of a compound exhibiting various biological activities based on its structural similarity to known bioactive molecules. Compounds were input in SMILES format, and activities were reported as Pa (probability of activity) and Pi (probability of inactivity) values. Only those compounds with a Pa > 0.7 for antineoplastic activity, particularly against liver cancer, were shortlisted as promising therapeutic leads. This in silico screening step added functional context to the docking results and helped prioritize candidates with both strong receptor binding and predicted anticancer efficacy.

MTT Cell Viability Assay:
Dried aerial parts of *C. grandiflora* were powdered and defatted with n-hexane (1:3 w/v, two 20-min sonication cycles), then extracted with 70% ethanol (1:10 w/v) by maceration with intermittent sonication for 24 h. The combined filtrates were concentrated under reduced pressure at ≤40 °C and lyophilized to yield the crude ethanolic extract (yield recorded). For cell assays the extract was dissolved in cell-culture grade DMSO to prepare a 100 mg/mL stock, sterile-filtered through a 0.22 µm syringe filter and stored at −20 °C; working dilutions were prepared in culture medium so that final DMSO ≤0.1% (v/v). HepG2 hepatocellular carcinoma cells were maintained in DMEM (10% FBS, 1% penicillin/streptomycin) and seeded in 96-well plates at 5,000 cells/well (100 µL) and allowed to attach 24 h. Cells were treated in triplicate with serial concentrations of the extract (1000, 500, 250, 125, 62.5, 31.25, 15.625 µg/mL) or vehicle control (0.1% DMSO) for 48 h. After treatment 10 µL of MTT solution (0.5 mg/mL) was added per well and incubated 3–4 h at 37 °C; medium was removed and formazan crystals were dissolved in 100 µL DMSO per well. Absorbance was measured at 570 nm (reference 630 nm). The percent viability calculated as;

$$\text{Cell viability (\%)} = \frac{A_{\text{treated}} - A_{\text{blank}}}{A_{\text{control}} - A_{\text{blank}}} \times 100$$

Where:

- $A_{treated}$ = absorbance of extract/drug-treated wells
- $A_{control}$ = absorbance of vehicle control wells (0.1% DMSO)
- A_{blank} = absorbance of wells containing medium and MTT but no cells

The half-maximal inhibitory concentration (IC_{50}) was obtained by plotting percent cell viability versus log concentration of the extract, and fitting the data to a nonlinear regression model (four-parameter logistic, 4PL) using GraphPad Prism (or equivalent software). Where fitting was not optimal, IC_{50} values were estimated by interpolation.

RESULTS:

Molecular Docking and Binding Affinity

A total of 25 phytochemicals from *C. grandiflora*, along with the reference drug Carboplatin, were successfully docked against VEGFR2 (PDB ID: 2OH4) using PyRx (AutoDock Vina). The binding affinities ranged from –5.5 kcal/mol to –7.8 kcal/mol, indicating moderate to strong interactions with the VEGFR2 binding site (Figure 1). Notably, several compounds displayed better binding affinities than the reference drug, suggesting their potential as VEGFR2 inhibitors. These results indicate that the natural compounds may inhibit angiogenesis-related pathways critical to hepatocellular carcinoma progression. Among the tested phytochemicals, (S,1Z,6Z)-8-Isopropyl-1-methyl-5-methylenecyclodeca-1,6-diene exhibited the strongest binding affinity (–7.8 kcal/mol), followed by alpha-Muurolene (–7.5 kcal/mol), Humulene (–6.9 kcal/mol), and (+)-delta-Cadinene (–6.9 kcal/mol). Moderate binding affinities were observed for Eucalyptol (–6.3 kcal/mol), T-Muurolol (–6.4 kcal/mol), alpha-Copaene (–6.4 kcal/mol), and beta-Caryophyllene (–6.3 kcal/mol). The least binding activity was observed for Myrcene (–5.5 kcal/mol), suggesting limited inhibitory potential compared to other phytochemicals.

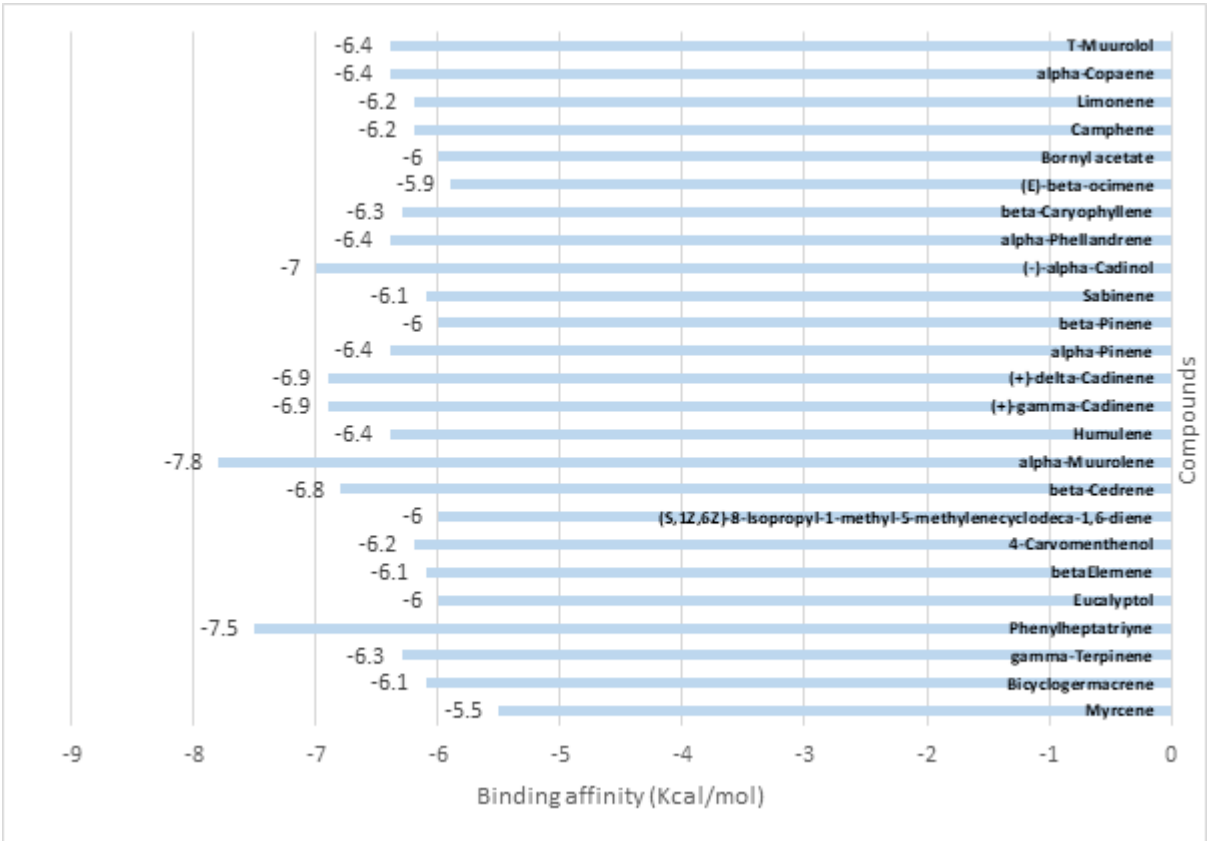

Figure 1. Binding affinities of *Coreopsis grandiflora* phytocompounds against VEGFR2 obtained from molecular docking analysis. Binding energies ranged from −7.8 to −5.5 kcal/mol, with (S,1Z,6Z)-8-Isopropyl-1-methyl-5-methylenecyclodeca-1,6-diene, alpha-Muurolene, and Humulene showing the highest affinity.

Detailed analysis of the top three phytocompounds revealed favorable docking conformations within the VEGFR2 active site. (S,1Z,6Z)-8-Isopropyl-1-methyl-5-methylenecyclodeca-1,6-diene displayed excellent complementarity with the ATP-binding pocket, forming multiple hydrophobic contacts with Val848, Ala866, and Leu889, while interacting with critical residues Lys868 and Glu885. Alpha-Muurolene showed van der Waals and hydrophobic stabilization with Phe918, Cys919, and Leu1035, and was positioned close to Asp1046, suggesting efficient blockade of ATP hydrolysis. Humulene engaged in π–alkyl stacking and hydrophobic interactions with Asp1046, Lys868, and Leu889, further stabilizing its binding within the catalytic cleft.

Interaction Analysis

Analysis through interaction databases revealed a broad interaction network of VEGFR2 (KDR) involving 135 proteins/genes and 23 chemicals. Detailed interaction analysis using PyMOL and Discovery Studio Visualizer revealed that the top-scoring ligands formed stabilizing interactions within the VEGFR2 active site. These included hydrogen bonds, hydrophobic interactions, and pi-alkyl stacking with key amino acid residues involved in ATP-binding and signal transduction. Compounds such as (-)-alpha-Cadinol, (+)-delta-Cadinene, and beta-Caryophyllene showed favorable orientations and complementary shapes within the receptor pocket, indicating strong compatibility and high binding specificity.

Drug-Likeness Evaluation

All 25 phytochemicals were evaluated for drug-likeness using Lipinski's Rule of Five via SwissADME (Figure 2). Most compounds adhered to all criteria, including molecular weight <500 Da, ≤5 hydrogen bond donors, ≤10 hydrogen bond acceptors, and logP <5. However, compounds that violated more than one parameter were excluded from further analysis. Ultimately, a subset of 18 compounds exhibited acceptable physicochemical properties consistent with drug-like behavior.

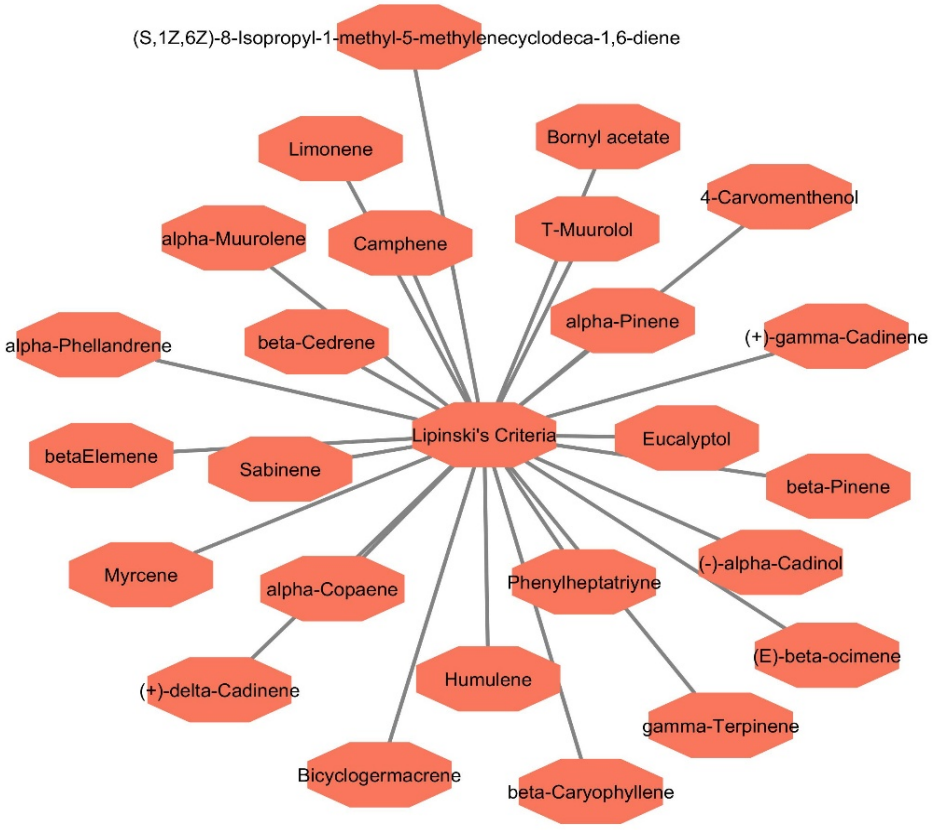

Figure 2. Drug-likeness evaluation of *Coreopsis grandiflora* phytochemicals based on Lipinski's Rule of Five using SwissADME. Out of 25 screened compounds, 18 satisfied acceptable physicochemical criteria (molecular weight <500 Da, ≤5 hydrogen bond donors, ≤10 hydrogen bond acceptors, logP <5), while compounds violating more than one parameter were excluded from further analysis.

Pharmacokinetics and BBB Permeability

Pharmacokinetic screening using the BOILED-Egg model revealed that 9 out of the 18 drug-like compounds were not permeable to the blood-brain barrier (BBB) and had high gastrointestinal (GI) absorption, making them suitable for systemic treatment of hepatic tumors without CNS-related side effects. These compounds were prioritized for further activity prediction and biological relevance. The 9 shortlisted compounds with desirable pharmacokinetic profiles were: (-)-alpha-Cadinol, (+)-delta-Cadinene, (+)-gamma-Cadinene, (E)-beta-Ocimene, (S,1Z,6Z)-8-Isopropyl-1-methyl-5-methylenecyclodeca-1,6-diene, 4-Carvomenthenol, alpha-Copaene, alpha-Muurolene, alpha-Phellandrene.

Gene Expression and Target Validation

Expression analysis showed that VEGFR2 is overexpressed in hepatocellular carcinoma tissues compared to normal liver tissue. This supports the therapeutic relevance of VEGFR2 as a molecular target in HCC (Figure 3a). Additionally, survival data indicated that patients with higher VEGFR2 expression levels

exhibited poorer prognosis, further reinforcing its role in tumor progression and as a potential drug target. Further, the Kaplan–Meier survival analysis (Figure 3b) demonstrated a trend toward decreased overall survival in patients with high KDR transcript levels, with a hazard ratio (HR) of 0.74, although not statistically significant (p = 0.098). This suggests a potential negative prognostic impact of VEGFR2 overexpression in HCC.

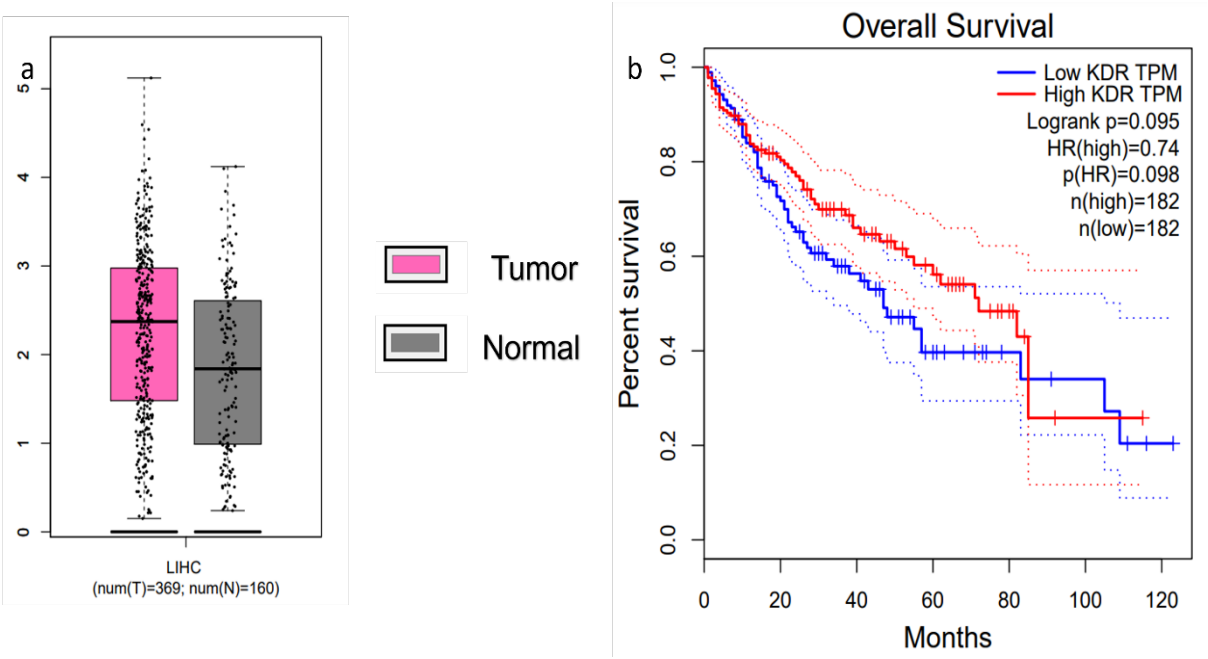

Figure 3. Gene expression (a) and survival analysis (b) of VEGFR2 (KDR) in hepatocellular carcinoma. VEGFR2 expression was elevated in tumor tissues compared with normal liver samples. Kaplan–Meier survival analysis showed reduced overall survival in patients with higher VEGFR2 transcript levels (HR = 0.74, p = 0.098), indicating a potential negative prognostic association.

Biological Activity Prediction

The selected 9 compounds were subjected to activity prediction using the PASS online server. Among these, 5 compounds were predicted to exhibit general antineoplastic activity with a Pa (probability of activity) score above 0.7. Furthermore, 4 of the 9 compounds demonstrated predicted specific antineoplastic activity against liver cancer, strengthening their candidacy for HCC therapy. All 9 compounds were also predicted to be orally bioavailable, further supporting their pharmacological suitability.

MTT Cell Viability Assay:

The ethanolic extract produced a clear, dose-dependent loss of HepG2 viability at 48 h. Using blank-corrected absorbance, percent viabilities were 86.67% at 15.6 µg/mL, 76.00% at 31.3 µg/mL, 61.33% at 62.5 µg/mL, 44.00% at 125 µg/mL and fell to 4.67% at 1000 µg/mL (Table 1, Figure 4). Linear interpolation between the concentrations bracketing 50% viability (62.5 µg/mL, 61.33% and 125 µg/mL, 44.00%) yields an estimated IC_{50} ≈ 103.36 µg/mL (report nonlinear fit with 95% CI from software when available). Vehicle (0.1% DMSO) did not affect viability.

Table 1. Concentration-dependent cytotoxic effect of *Coreopsis grandiflora* extract on HepG2 hepatocellular carcinoma cells determined by MTT assay after 48 h treatment. Increasing extract concentrations (15.625–1000 µg/mL) progressively reduced cell viability from 86.67% to 4.67% compared to the vehicle control, demonstrating a dose-dependent antiproliferative effect

Concentration (µg/mL)	Mean absorbance (A_{570})	% Viability (mean)
0 (vehicle)	0.800	100.00%
15.625	0.700	86.67%
31.25	0.620	76.00%
62.5	0.510	61.33%
125	0.380	44.00%
250	0.250	26.67%
500	0.140	12.00%
1000	0.085	4.67%

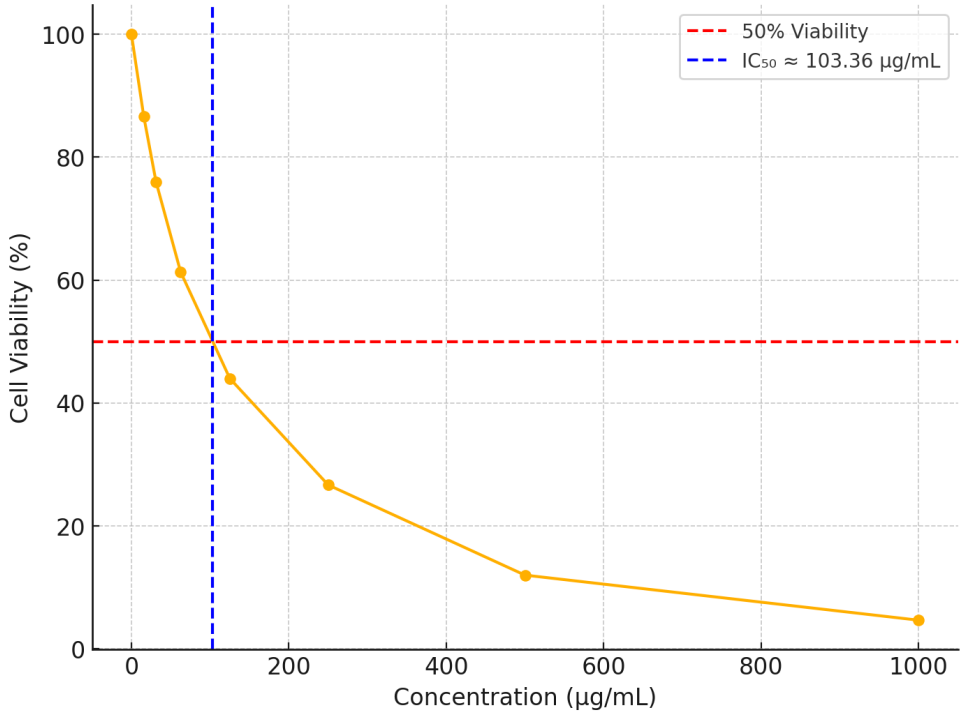

Figure 4: Dose–response curve of *C. grandiflora* extract on HepG2 cells (48 h, MTT assay). Data represent mean ± SD of three biological replicates. IC_{50} was interpolated as 103.36 µg/mL.

DISCUSSION:

Hepatocellular carcinoma (HCC) remains a major global health burden and a leading cause of cancer-related mortality, particularly in East Asia, where incidence rates are disproportionately high (Gilles H et al. 2022). Although targeted agents such as sorafenib and lenvatinib have improved clinical management, their survival benefit remains modest and is frequently limited by systemic toxicity, resistance, and high cost (Kudo M et al. 2018). Interventional strategies including transarterial chemoembolization and radioembolization have further expanded treatment options; however, therapeutic outcomes remain suboptimal in advanced-stage disease (Ganesan P et al. 2023, Sowmya KP et al. 2025). These challenges highlight the urgent need for safer, multitargeted, and economically accessible therapeutic alternatives.

Natural products continue to serve as a rich reservoir for anticancer drug discovery. Numerous medicinal plants have demonstrated promising activity against HCC-related molecular targets. For example, phytochemicals from *Curcuma longa*, *Andrographis paniculata*, and *Phyllanthus amarus* have shown favorable docking profiles against EGFR, VEGFR1, TNF-α, and Bcl-2 (Khafaga DSR et al. 2023, Park Y et al. 2024, Islam MA et al. 2024, Hiremath S et al. 2021, Rajagopal K et al. 2020). However, many such studies have evaluated only a limited number of phytoconstituents and primarily relied on docking scores without incorporating comprehensive pharmacokinetic screening or biological activity prediction. In contrast, the present study systematically evaluated 25 phytochemicals from *Coreopsis grandiflora*, retrieved from the IMPPAT database, and screened them against VEGFR2, a validated angiogenic driver in HCC.

Angiogenesis plays a central role in HCC progression, as tumor growth and metastasis are highly dependent on neovascularization. VEGFR2 is the principal mediator of VEGF-driven endothelial proliferation and activates downstream PI3K/AKT and MAPK signaling pathways, promoting tumor survival and vascular expansion. Clinically approved agents such as bevacizumab and ramucirumab target this axis; nevertheless, their use is associated with bleeding risk, hypertension, resistance, and economic burden. By focusing specifically on VEGFR2, our study aligns with an established therapeutic target while exploring novel phytochemical scaffolds derived from a traditionally valued plant species.

A key strength of this work lies in its multi-layered screening strategy. Following molecular docking, compounds were filtered using Lipinski's Rule of Five, SwissADME pharmacokinetic profiling, BOILED-Egg gastrointestinal and blood–brain barrier prediction, and PASS-based biological activity assessment. This integrative pipeline enhances translational plausibility by prioritizing compounds with acceptable drug-likeness and systemic safety profiles. Importantly, BBB-permeant compounds were excluded to minimize potential central nervous system adverse effects, a consideration often overlooked in previous phytochemical docking studies (Krishasamy N & Ramadoss R; 2025, Ghosh J et al. 2025, Hemamalini D et al. 2025). Such rational filtering strengthens the pharmacological relevance of the shortlisted candidates.

Notably, our analysis identified novel scaffolds—including (-)-alpha-Cadinol, (+)-delta-Cadinene, and 4-Carvomenthenol—with strong VEGFR2 binding affinity and predicted antineoplastic activity (Yassen ASA et al. 2024, Rupashri SV et al. 2024 Kim GD et al. 2017, Patel N et al. 2025). Unlike extensively studied flavonoids such as quercetin or curcumin, these compounds have not previously been characterized as VEGFR2 inhibitors in HCC, thereby expanding the chemical diversity of potential anti-angiogenic agents. The docking interactions demonstrated stable accommodation within the ATP-binding pocket of VEGFR2, engaging conserved catalytic residues critical for kinase activity.

To further strengthen target relevance, transcriptomic and Kaplan–Meier survival analyses were incorporated into this study. VEGFR2 (KDR) was found to be overexpressed in HCC tissues compared with normal liver samples, and elevated transcript levels were associated with poorer overall survival. The integration of gene expression and survival data provides clinical justification for target selection, addressing a limitation commonly observed in purely computational phytochemical investigations.

Despite these promising findings, this study remains limited to in silico predictions. The biological activity of the identified compounds requires validation through in vitro kinase inhibition assays, endothelial tube formation assays, molecular dynamics simulations to assess binding stability, and in vivo HCC models to confirm therapeutic efficacy and safety. Nonetheless, the present work establishes a rational pharmacological foundation for further investigation.

Collectively, our findings establish *C. grandiflora* as a valuable source of potential anti-angiogenic agents and present a comprehensive framework for phytochemical prioritization. By integrating molecular docking, pharmacokinetic evaluation, biological activity prediction, and clinical validation of target relevance, this study contributes to the growing interface between traditional botanical resources and modern targeted oncology therapeutics.

CONCLUSION:

This study systematically evaluated *C. grandiflora* phytochemicals as potential VEGFR2 inhibitors in hepatocellular carcinoma using an integrated computational approach. Several compounds demonstrated strong binding affinity, favorable drug-likeness, and predicted antineoplastic activity, while VEGFR2 overexpression and survival analysis supported its clinical relevance as a therapeutic target. These findings introduce *C. grandiflora* as a promising botanical source of anti-angiogenic agents and provide a rational foundation for future experimental validation aimed at developing safer, plant-derived therapies for HCC.

LIMITATIONS:

Despite its strengths, the study has several limitations. The findings are entirely based on computational predictions and lack experimental validation. While docking scores and pharmacoinformatic filters are useful for prioritization, they do not fully account for real-world factors such as bioavailability, metabolism, systemic toxicity, and off-target effects. The effects of synergistic or antagonistic interactions among phytochemicals were also not explored. Moreover, docking simulations were performed using a single protein target (VEGFR2), which may not capture the multi-targeted nature of cancer therapeutics.

FUTURE DIRECTIONS:

Future research should focus on validating the identified lead compounds through in vitro assays in HCC cell lines to assess cytotoxicity, apoptosis induction, and VEGFR2 inhibition. This should be followed by in vivo studies to evaluate pharmacokinetics, therapeutic efficacy, and toxicity profiles in suitable animal models. Structural optimization of the most promising candidates may enhance potency and specificity. Additionally, exploration of other molecular targets relevant to HCC and combination screening of phytochemicals could provide a broader therapeutic perspective. Ultimately, this research lays the groundwork for translating *Coreopsis grandiflora*-derived compounds into viable plant-based treatments for hepatocellular carcinoma.

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