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In silico assessment of the anticancer effects of tecomaquinone I, brevilin A, erybraedin A, evodiamine, and sumadain C in oral squamous cell carcinoma

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Abstract

Oral squamous cell carcinoma (OSCC) is one of the most common malignancies globally. Current treatment modalities, including chemotherapy and radiotherapy, often result in adverse effects that compromise the quality of life for patients. An alternate approach involves using phytocomposites as substitutes for commercial chemotherapeutic agents, potentially offering improved therapeutic outcomes with minimal adverse effects. The study aims to investigate the anticancer effects of phytocomposites in OSCC. A literature search was performed to select relevant phytocomposites for the analysis. The chosen compounds included tecomaquinone I, brevilin A, erybraedin A, evodiamine, and sumadain C. Bioactive compound information was extracted using online software tools, such as ZINC15, SwissADME, PharmaGist, and ZINCPharmer. The target protein for this study was Harvey rat sarcoma virus oncogene and its structure was obtained from the Protein Data Bank. On evaluating the binding affinities of these phytocomposites to the target protein, it was found that ZINC94088045 and ZINC94241870 exhibited the highest binding affinities, with values of -18.71 and -15.75 kcal/mol, respectively. These compounds also demonstrated favorable pharmacokinetic properties, with total polar surface area values of 85.11 Å^2 and 122.39 Å^2, log P values of 2.59 and 1.77, and bioavailability scores of 0.55 and 0.56, respectively. Notably, these compounds also showed high gastrointestinal absorption and low cytotoxicity. The final phytocomposite combination displayed a strong binding affinity to the target protein while maintaining low cytotoxicity. This study highlights the potential anticancer effect of phytocomposites on OSCC with negligible cytotoxicity, offering a promising alternative to conventional treatments.

Keywords: Binding affinity; Oral squamous cell carcinoma; Phytocomposites; Harvey rat sarcoma virus oncogene

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1. Introduction

Oral cancer is a significant global health concern, ranking as the sixth most common cancer worldwide and the third most prevalent cancer in India, where it accounts for nearly 40% of all cancers. In India, oral cancer is the leading cause of death and is particularly prevalent among men, making it the most common cancer in this demographic. According to Petersen,[1] the age-standardized incidence rate of oral cancer in India is 12.6/100,000 people, with a rapidly increasing trend.[1] Approximately 90% of oral malignancies are oral squamous cell carcinoma (OSCC).[2] The causes of OSCC are multifaceted, including smoking, chewing tobacco, alcohol consumption, chronic inflammation, nutrient deficiencies, genetic mutations, infections, dental neglect, poor oral hygiene, and alterations in oral microbiota.[3] Current treatment options for OSCC include surgical excision of the cancerous tissue followed by radiotherapy and chemotherapy. However, these therapies are limited by the disease's heterogeneity and complex mechanisms, underscoring the need for advanced and alternative treatments.

Conventional chemotherapeutic drugs used to treat OSCC, such as antimetabolites (e.g., methotrexate), DNA interactive agents (e.g., cisplatin and doxorubicin), anti-tubulin agents (e.g., taxanes), hormones, and molecular targeting agents,[4] have significant anticancer effects but are often accompanied by severe adverse effects. These adverse effects, including recurrence, drug resistance, toxicity to non-target tissues, diarrhea, tingling or numbness in the hands or feet, itchy skin, damage to the oral mucosa, xerostomia, allergic reactions, memory issues, decreased kidney function, hearing loss or ringing in the ears, nausea, vomiting, fatigue, decreased appetite, changes in taste, alopecia, and constipation,[5] significantly impair the quality of life for post-operative patients. There is a pressing need for chemotherapeutic agents with fewer side effects and higher efficacy to improve patient outcomes. Recent scientific evidence highlights the potential of phytocompounds as anticancer agents. Given their diverse chemical composition and biological activities, phytocompounds have emerged as promising alternatives to widely used chemotherapeutic medications.

Phytocompounds play a variety of roles, including acting as cofactors of enzymatic reactions, inhibitors of enzymatic reactions, scavengers of reactive or toxic chemicals, enhancers of essential nutrient absorption, stabilizers of nutrients, selective inhibitors of deleterious intestinal bacteria, and fermentation substrates for beneficial oral, gastric or intestinal bacteria.[6]

Phytochemicals, including flavonoids, terpenoids, polyphenols, chalcones, quinolones, quinones, sesquiterpenes, coumarins, alkaloids, tannins, and amides, are bioactive compounds found in plants that exhibit multi-dimensional pharmacological effects. These compounds can serve as effective chemotherapeutic agents due to their ability to interact with cellular targets by selectively targeting cancer cells and sparing healthy cells. This selective cytotoxic is achieved by modulating specific pathways, such as those involving the epidermal growth factor receptor (EGFR), cytokine receptors, toll-like receptors, and tumor necrosis factor receptors, which ultimately inhibit tumor cell proliferation and induce programmed cell death.[7] One of the most advantageous properties of phytocompounds is their strong antioxidant and anti-inflammatory properties. These properties help reduce inflammation and reactive oxygen species, both of which play critical roles in cancer development and progression. Chronic inflammation creates a microenvironment that supports tumor growth, invasion, and metastasis, while free radicals can initiate apoptosis in cancer cells or contribute to cancer development if not involved in programmed cell death. Phytocompounds also regulate various cellular processes, including cell proliferation, cell cycle progression, DNA repair, metabolism, metastasis, angiogenesis, and the activation or inactivation of oncogenes and tumor suppressor genes.[8] By inhibiting dysregulated factors in cancer cells, phytocompounds can enhance the effects of conventional therapies or be developed into standalone treatments.[9] Their use as chemotherapeutic agents or adjuvants offers an improved therapeutic environment with reduced toxicity profiles and enhanced tolerance, potentially improving the quality of life for patients undergoing chemotherapy, reducing treatment interruptions, and encouraging long-term adherence.

The cell cycle is a complex series of events that a cell undergoes during its growth and division,[10] regulated by several checkpoints to ensure proper progression. Proteins controlling the cell cycle are encoded by oncogenes, with proto-oncogenes playing an important role in advancing the cell cycle from the G (Gap) phase to either the S phase (DNA replication) or mitosis (chromosomal segregation). Mutations in proto-oncogenes disrupt normal physiological processes, converting them into oncogenes that produce aberrant proteins. These proteins are hyperactive, promoting tumor growth and development. One such oncogene is rat sarcoma virus (RAS), a GTPase molecule involved in signaling pathways that regulate cell proliferation, differentiation, migration, and apoptosis.[11] Harvey RAS (HRAS) is a member of the RAS superfamily and participates in the Ras-Raf-mitogen-activated protein kinases (MAPK) and the phosphoinositide-3 kinase (PI3K) pathways, which are critical for signal transduction and determining cellular life processes. Mutations in *HRAS* lead to dysregulation of these pathways contributing to

tumorigenesis. *RAS* mutations are found in approximately 30% of all human tumors.[12] Specifically, Kirsten RAS (*KRAS*) mutations are prevalent in lung cancer (32%), colorectal cancer (40%), and pancreatic cancer (85 – 90%).[13-15] While *HRAS* mutations are less common than *KRAS* and neuroblastoma RAS (*NRAS*) mutations, they are the most frequently detected in head and neck cancers.[16,17]

Anticancer drug discovery is a complex, time-consuming, and expensive process that involves several stages, including target identification, chemical synthesis, preclinical testing using in vitro and animal models, and toxicity assessment. Successful compounds are then evaluated in clinical trials to assess their safety and efficacy in the target patient population, which can take several years to complete.[18] Recent advancements in technology have led to the development of computer-aided drug designing (CADD). CADD is an efficient way to identify potential compounds and aids in new drug development. CADD encompasses various methods, including structure-based drug designing (molecular docking and molecular dynamics) and ligand-based drug designing, such as ligand-based pharmacophore modeling, quantitative structure-activity relationship modeling, fragment-based drug designing, pharmacokinetics, and pharmacodynamics modeling.[19] These computational techniques simulate and predict the interaction of drug molecules with their target using computer algorithms on chemical and biological data. Several United States Food and Drug Administration (FDA)-approved anticancer drugs have been developed using CADD. For example, crizotinib, which targets hepatocyte growth factor receptors, anaplastic lymphoma kinase, and tyrosine kinase mesenchymal-epithelial transition, was approved in 2011 for treating lymphoma, esophageal cancer, and lung cancer.[20] Axitinib, a vascular endothelial growth factor receptor inhibitor for advanced renal cell carcinoma, was approved in 2012.[21] Recently, tubulin inhibitors have been developed using the ligand-based technique that targets tubulin polymerization, a vital process for cell cycle progression and division.[22] In addition, a human aromatase inhibitor has been developed through ligand-based approaches for the treatment of estrogen receptor-positive breast cancer.[23] Gefitinib, an EGFR inhibitor identified through structure-based design, was approved by the FDA in 2015 to treat advanced or metastatic non-small cell lung cancer (NSCLC).[24] Erlotinib, approved in 2004, targets EGFR kinases and is used for treating pancreatic cancer and NSCLC,[25] while lapatinib, approved in 2007, targets ERBB2/EGFR to treat breast cancer.[26] Other drugs, such as abiraterone, an androgen synthesis inhibitor[27] used for metastatic castration-resistant prostate cancer, and imatinib, a tyrosine kinase inhibitor[28] used for chronic myeloid leukemia, were approved by the FDA in 2011 and 2004, respectively. Moreover, several drugs like TAK-659, DZD9008, AZD5991, SHR- 3162, resveratrol, quercetin, and epigallocatechin gallate are currently undergoing clinical trials.[29-35] These examples highlight the importance of computational methods in designing anticancer drugs, paving the way for novel drug discoveries that can improve patient's quality of life. In this study, molecular docking was used to analyze the binding affinities, total polar surface area (TPSA) values, log P value, and bioavailability of the selected compounds (tecomaquinone I, brevilin A, erybraedin A, evodiamine, and sumadain C) with the target protein HRAS, which plays a role in cell proliferation and apoptosis. HRAS is a key oncogene involved in tumorigenesis, making it an important target for anticancer therapies. This study aimed to assess the anticancer effects of these phytocompounds in OSCC using *in silico* analysis like molecular docking. If the results are promising, further experimental and clinical studies could lead to the development of effective chemotherapeutic drugs.

2. Methods

Five phytocompounds with potential anticancer activity were selected through an extensive literature search. These phytocompounds are: (i) Tecomaquinone I, a quinone found in plants like *Tectona grandis* and *Lippia origanoides*, with a molecular formula of $C_{30}H_{24}O_4$, (ii) brevilin A, a natural sesquiterpene lactone found in *Centipeda minima*, with a molecular formula of $C_{20}H_6O_5$, (iii) erybraedin A, a flavonoid from the *Erythrina* species, with a molecular formula of $C_{25}H_{28}O_4$, (iv) evodiamine, an alkaloid from the dry unripened fruit of *Evodiae fructus*, with a molecular formula $C_{19}H_{17}N_3O$, and (v) sumadain C, a monoterpene-chalcone conjugate found in the seeds of *Alpinia katsumadai*, with a molecular formula of $C_{25}H_{26}O_5$.

The simplified molecular input line entry system (SMILES) notation for each compound is provided in Table 1. The SMILES form is a notation system used to represent the chemical structure of a drug in a linear format. The chemical structures of the compounds were retrieved from the PubChem database in.sdf format and converted into three-dimensional (3D) structures in.pdb format using the Open Babel software. The chemical structures of the selected phytocompounds are given in Figure 1. The 3D structure of the human HRAS protein was retrieved from the RCSB Protein Data Bank and is shown in Figure 2. The SMILES form and the structures of the selected phytocompounds were obtained from ZINC15, and a pharmacophore model was obtained from ZINCPharmer. A combination of approximately 20 phytocomposites was selected based on their log P values, TPSA values, and adherence to drug-likeliness rules

Table 1. Simplified Molecular Input Line Entry (SMILE) System form of phytocompounds tecomaquinone I, brevilin A, erybraedin A, evodiamine, and sumadain C

Phytocompound	SMILE
Tecomaquinone I	CC (C)=C[C@@H] 1Oc2c (c3c (c4ccccc24) OC (C)(C) C=C3) C2=C1C(=O) c1ccccc1C2=O
Brevilin A	C/C=C(/C) C(=O) O[C@H] 1[C@@H] 2[C@H](C) C(=O) O[C@@H] 2C[C@@H](C)[C@@H] 2C=CC(=O)[C@@]12C
Erybraedin A	CC (C)=CCc1c (O) ccc2c1O[C@H] 1c3ccc (O) c (CC=C (C) C) c3OC[C@@H] 21
Evodiamine	CN1c2ccccc2C(=O) N2CCc3c([nH] c4ccccc34)[C@@H] 21
Sumadain C	CC (C)[C@H] 1C=C[C@]2(C) Oc3c (C(=O)/C=C/c4ccccc4) c (O) cc (O) c3[C@H] 2C1

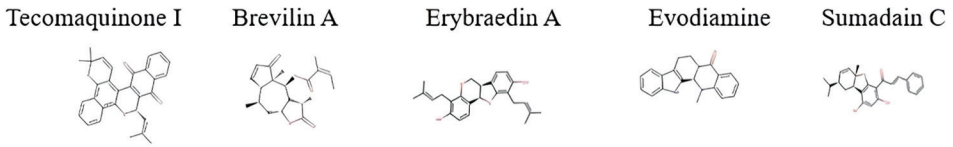


Figure 1. The structures of phytocompounds tecomaquinone I, brevilin A, erybraedin A, evodiamine, and sumadain C

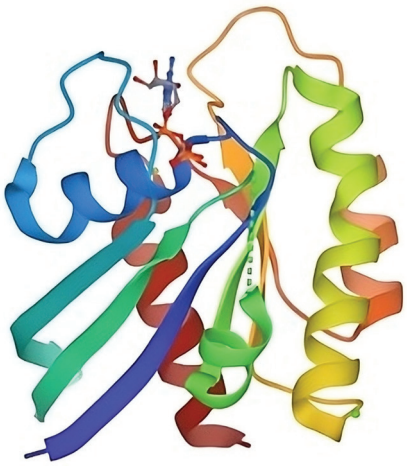


Figure 2. The structure of the Harvey rat sarcoma viral oncogene protein

(Lipinski, Ghose, Veber, Egan, Muegge). The log P value is a measure of lipophilicity, while the TPSA value is the measure of the drug's bioavailability and its ability to penetrate biological barriers. The rules of drug likeliness help determine the pharmacokinetics of a drug. The Lipinski rule assesses whether a chemical compound with certain pharmacological or biological activity possesses chemical properties and physical properties that would likely make it orally active in humans. The Ghose rule predicts drug likeliness based on factors such as molecular weight, molar refractivity, predicted log P value, and the total number of atoms. The Veber rule determines a molecule's oral bioavailability based on its structure, while the Egan rule predicts oral absorption using molecular weight, log P, hydrogen bond donors, hydrogen bond acceptors, and TPSA value. The Muegge rule predicts drug likeliness using molecular weight, Xlog P, TPSA, number of rings, carbons, heteroatoms, rotatable bonds, hydrogen bond acceptors, and hydrogen bond donors. The target protein and each phytocompound combination were docked using SwissDock software to evaluate their binding affinities.

3. Results

To assess the anticancer potential of selected phytocompounds, molecular docking was employed to evaluate their binding affinities with the target protein HRAS. The results showed that the binding affinities ranged from −7.25 to −18.71 kcal/mol. The pharmacokinetic properties of the selected phytocompounds, including log P values, TPSA values, bioavailability, and gastrointestinal absorption, were also analyzed. The details of these parameters for the selected 20 phytocompounds are given in Table 2. The structure of the docked protein and phytocompounds is shown in Figure 3.

4. Discussion

Oral squamous cell carcinoma has a significant mortality rate that is well-known. Several current treatment approaches for the disease include surgical intervention, radiation, and chemotherapy. However, existing chemotherapy medications have excruciating side effects that reduces the quality of life for patients. Therefore, pharmacologically active phytocompounds have gained increasing importance due to their lower adverse effects compared to current treatment modalities.

Genes contain the information necessary to produce proteins. Gene mutations can alter the way proteins function and some mutations may cause proteins to function abnormally, transforming normal cells into cancer cells. *RAS* is a proto-oncogene that is frequently mutated in cancer, with mutations observed in 10 – 30%

Table 2. Log *P* values, total polar surface area values, bioavailability, gastrointestinal absorption, and the binding affinities of the 20 selected phytocompounds

Molecule ID	Rule 1	Rule 2	Rule 3	Rule 4	Rule 5	Log P	TPSA	Bioavailability	GI	Binding affinity
ZINC87313354	Y	Y	Y	Y	Y	1.86	70.59 A^2	0.55	High	−7.38
ZINC58224231	Y	Y	Y	Y	Y	2.28	61.36 A^2	0.55	High	−7.25
ZINC94088045	Y	Y	Y	Y	Y	2.59	85.11 A^2	0.55	High	−18.71
ZINC85651314	Y	Y	Y	Y	Y	3.58	94.85 A^2	0.55	High	−7.26
ZINC94046548	Y	Y	Y	Y	Y	2.22	75.35 A^2	0.55	High	−7.62
ZINC73848468	Y	Y	Y	Y	Y	1.81	62.22 A^2	0.55	High	−7.65
ZINC86810909	Y	Y	Y	Y	Y	2.52	77.57 A^2	0.55	High	−7.46
ZINC75223456	Y	Y	Y	Y	Y	2.35	49.33 A^2	0.55	High	−7.76
ZINC91437757	Y	Y	Y	Y	Y	2.21	62.22 A^2	0.55	High	−7.36
ZINC94241870	Y	Y	Y	Y	Y	1.77	122.39 A^2	0.56	High	−15.75
ZINC32808915	Y	Y	Y	Y	Y	2.67	108.56 A^2	0.55	High	−7.39
ZINC94993303	Y	Y	Y	Y	Y	2.09	95.15 A^2	0.55	High	−7.16
ZINC69511953	Y	Y	Y	Y	Y	2.89	104.26 A^2	0.55	High	−7.49
ZINC65591349	Y	Y	Y	Y	Y	2.69	61.36 A^2	0.55	High	−8.07
ZINC66160667	Y	Y	Y	Y	Y	3.02	99.69 A^2	0.55	High	−8.03
ZINC58227918	Y	Y	Y	Y	Y	3.28	81.43 A^2	0.55	High	−7.36
ZINC12698794	Y	Y	Y	Y	Y	3.82	120.37 A^2	0.55	High	−8.21
ZINC58181828	Y	Y	Y	Y	Y	3.17	81.43 A^2	0.55	High	−7.7
ZINC58195891	Y	Y	Y	Y	Y	2.37	61.36 A^2	0.55	High	−7.45
ZINC66161033	Y	Y	Y	Y	Y	2.75	94.38 A^2	0.55	High	−8.09

Abbreviations: GI: Gastrointestinal absorption; TPSA: Total polar surface area.



Figure 3. The structure of the docked rat sarcoma virus protein and the phytocompounds tecomaquinone I, brevilin A, erybraedin A, evodiamine, and sumadain C

of the cancer cases. The RAS family of proteins, which includes small GTPases, is commonly involved in aberrant signaling pathways. Binary proteins known as RAS proteins are crucial components of signal transduction pathways.[36] The RAS protein family is encoded by three genes, *HRAS*, *KRAS*, and *NRAS*, and is involved in the regulation of cell division. These proteins play a key role in determining the destiny of cells during cell cycle[37] which is a well-planned process that coordinates DNA replication, cell development, and division. The cell cycle is crucial for growth and sustenance of all living organisms, and any disruptions in the cell cycle can eventually cause problems in cell growth and differentiation.

The HRAS protein functions as a molecular switch, cycling between active guanosine triphosphate (GTP)-bound and inactive guanosine diphosphate (GDP)-bound states.[38] The switch mechanism is crucial for signal transduction, promoting cell growth, differentiation, and proliferation by activating downstream signaling proteins. This switch mechanism has been found in a wide range of GDP/GTP-binding proteins, including heterotrimeric G-proteins, bacterial elongation factors, and several small GTPases with various biological activities. The transition from the inactive GDP-bound form to the active GTP-bound form is facilitated by guanine nucleotide exchange factors.[39]

The activation of HRAS is also influenced by post-translational modifications such as farnesylation, palmitoylation, and ubiquitination. Farnesylation, catalyzed by farnesyl

transferase, is essential for HRAS membrane localization and protein-protein interactions.[40] Palmitoylation, a lipid post-translational change, occurs when the palmitoyl chain is joined to the cysteine group.[41] It aids in maintaining the activity, stability, and trafficking of proteins while facilitating protein clustering and mediating the affinity of a protein for a lipid raft.[42] Palmitoylation is catalyzed by palmitoyl transferases.[43] Ubiquitination is the ubiquitin-conjugating system responsible for the post-translational alteration of proteins.[44] This process promotes protein breakdown by controlling proteasome activity, which is catalyzed by the conserved E1-E2-E3 three-enzyme cascade in an adenosine triphosphate (ATP)-dependent manner.[45,46]

Farnesylation is a crucial step for HRAS to mature into an active state. When HRAS remains in an active state for an extended period, it becomes mutated.[47] Given that HRAS proteins play a crucial role in the cell cycle, which regulates cell proliferation and apoptosis, alterations in these proteins can suppress cell cycle arrest, thereby stimulating the growth of tumor cells.[48]

Farnesyl transferase inhibitors are a class of signal-transducing inhibitors designed to target the post-translational modifications of HRAS. These inhibitors prevent proteins from mislocalizing to cellular compartments due to their inability to localize on the cell membrane. Consequently, oncogenic activity is suppressed by interfering with downstream signaling pathways, which rely on the appropriate localization and interaction of the proteins on the membrane. Mislocalized proteins accumulate in intracellular compartments, activating cellular stress and inducing apoptosis in cancer cells. Thus, preventing farnesylation using a farnesyl transferase inhibitor is important for the proper function and localization of proteins.[49]

Farnesyl transferase inhibitors have demonstrated significant cytotoxic effects in various studies.[50] For instance, the farnesyl transferase inhibitor SCH66336 (lonafarnib) has shown antitumor activities in head and neck squamous cell carcinoma (HNSCC) both *in vitro* and *in vivo*. Its antitumor activity is mediated by insulin-like growth factor binding protein-3, which inhibits angiogenesis.[51] Tipifarnib, the first farnesyl transferase inhibitor to enter clinical development in 1997, has been shown to inhibit the growth of MCF-7 xenografts and induce cell death by increasing the expression of the cell cycle inhibitory protein, p21.[52] Moreover, combining tipifarnib and tamoxifen significantly increased tumor regression in endocrine therapies.[5]

Tecomaquinone I, a naturally occurring naphthoquinone, acts as a farnesyl transferase inhibitor. It prevents the farnesylation of target proteins, including RAS, thereby inhibiting RAS activation and stopping cell growth and cancer cell proliferation. In addition, tecomaquinone I exhibits strong anti-inflammatory, antioxidant, and anti-microbial effects. It has been identified as a potent natural inhibitor of human farnesyl transferase (IC_{50} 0.065 ± 0.004 μM).[53]

Cytokines are essential for controlling the response of the host immune system to infections but can also contribute to cancer development. They activate signaling pathways through the Janus kinase (JAK) and signal transducer and activator of transcription (STAT) pathways by binding to receptors and activating responses.[54] The JAK-STAT pathway is essential for cellular proliferation and differentiation. On cytokine binding to receptors, JAKs are activated, leading to the transfer of phosphate from ATP to the tyrosine residues of receptors. Subsequently, JAKs phosphorylate STATs, which become activated, migrate into the nucleus, and modify gene expression.[55] This shows the significance of JAKs and the need for JAK inhibitors in cancer treatment, as they can slow the spread of cancerous tissue. The JAK-STAT pathway undergoes various changes in solid tumors, including head and neck malignancies, lung cancers, breast cancers, and prostate cancers.[56]

A protein kinase inhibitor like abemaciclib can inhibit cancer cell development. JAK inhibitors, such as ruxolitinib, inhibit JAK proteins.[57] Ruxolitinib, when combined with cisplatin has been shown to reduce STAT3 activation and cell proliferation, promote apoptosis, and suppress tumor growth in preclinical *in vitro* and *in vivo* models of cisplatin-resistant NSCLC with elevated JAK2 and STAT3 activation levels.[58] The p38α mitogen-activated protein kinase inhibitor is a potential therapeutic agent for oral cancer. It inhibits p38 activation – a pathway that promotes cellular proliferation, migration, and invasion.[59] In addition, PKC412, an oral protein kinase inhibitor, inhibits the cell cycle and exhibits anti-angiogenic properties.[60]

Brevilin A, a natural JAK inhibitor, suppresses the STAT3 pathway by inhibiting STAT3 signaling and inhibiting STAT3-dependent cell proliferation.[61] It demonstrates high selectivity for STAT signaling pathway-dependent JAK activity without affecting other signaling pathways. Brevilin A also reduces STAT1 phosphorylation and its target gene, *IRF1*, and inhibits the JAK tyrosine kinase domain JH1.[62] Moreover, brevilin A has been shown to induce apoptosis and suppress STAT3 activation in human lung cancer cells.[63]

For cancer cells to survive, migrate, and metastasize, the extracellular matrix (ECM) must be properly controlled, as aberrant ECM adhesion can lead to cancer cell invasion and metastasis.[64] The focal adhesion kinase (FAK)-mediated phosphorylation of Src and the direct interaction

of integrins with Src through the cytoplasmic domain play key roles in the adherence of cancer cells to the ECM.[64,65] Src, a member of the membrane-bound non-receptor tyrosine kinase family, is frequently expressed in various malignancies and acts as a key mediator.[66] Specifically, Src activates the Akt-mediated pro-survival pathway, rendering cells resistant to anoikis, a form of detachment-induced cell death.[67] In addition, Src facilitates cell motility by activating Rac and cell division control protein 42 homolog, leading to cytoskeletal reorganization.[68] These alterations collectively enable cancer cells to acquire pro-metastatic characteristics. Src kinases are also involved in invadopodia formation through the phosphorylation of cortactin and other targets,[69] regulating invadopodium maturation by activating the Abl-related non-receptor tyrosine kinase Arg. Constitutively active Src kinase can enhance invadopodia formation, which is crucial for tumor invasion. Invadopodia are actin-rich structures that facilitate the formation of invasive cytoplasmic extensions, allowing cancer cells to breach the endothelium and project into the adjacent environment, leading to cancer cell extravasation. Src kinases have been seen as a desirable target for cancer treatment. Given their role in cancer progression, Src kinases are considered desirable targets for cancer treatment. Src inhibitors impede tumor cell migration and invadopodia development.[70] Dasatinib, the first Src inhibitor authorized by the FDA, is used to treat acute lymphoblastic leukemia with the Philadelphia chromosome and has shown anti-proliferative, anti-invasive, anti-angiogenic properties in solid tumors. Bosutinib has demonstrated a positive effect in patients with locally progressed or metastatic breast cancer by extending the time to progression.[71] Despite the efficacy of bosutinib, the clinical development of commercial Src inhibitors has been halted due to side effects.[72,73] Saracatinib is another Src inhibitor approved by the FDA, while PP2, an Src inhibitor, has been shown to inhibit the cell growth and progression of HNSCC by regulating the epithelial-mesenchymal transition pathway.[74]

Erybraedin A is a natural Src inhibitor that prevents the activation of the signals related to cell-ECM adhesion, thereby controlling cancer progression. It inhibits the activation of Src, FAK, and Akt, which downstream targets. Erybraedin A suppresses NSCLC cell viability, clonogenic survival, and tumorigenicity by inducing apoptosis.[75] In addition, it inhibits cell adhesion to fibronectin by inactivating Src in NSCLC.

The alkaloid evodiamine has been shown to modulate the activity of various molecular targets, including kinases, transcription factors, and multiple signaling pathways. It significantly downregulates the expression of several tumor markers, including vascular endothelial growth factor (VEGF) and cyclooxygenase-2, and blocks key signaling pathways Wnt/β-catenin, mTOR, NF-κB, PI3K/Akt, JAK-STAT, and others in different cancer cells. These findings contribute to elucidating the anticancer mechanism of evodiamine.[76] In human lung cancer cells, A549 and H1299, evodiamine was found to substantially enhance mitochondrial membrane depolarization, increase the cell death rate, and marginally decrease the Bcl-2/Bax ratio. In addition, it accelerated the mitochondrial release of cytochrome C into the cytoplasm, activating a series of pro-apoptotic pathways, including caspases 3 and 9.[77] Evodiamine also inhibited the PI3K/Akt and MAPK/ERK signaling pathways in human pancreatic cancer cells, Panc-1 and SW1990, as well as the phosphorylation of transcriptional activator 3 and signal transduction to inhibit autophagy.[78] Furthermore, evodiamine exhibits anti-metastatic properties by blocking cancer cell migration and invasion. This is achieved by regulating the expression and activity of matrix metalloproteinases, which are essential for ECM degradation and tumor cell invasion.[79] Metastasis, the spread of cancer to distant organs, remains a significant challenge in cancer treatment. Evodiamine's strong anti-angiogenic impact is crucial, as it involves inhibiting the growth of new blood vessels that supply oxygen and nutrition to tumors. By disrupting this supply, tumor growth and metastasis can be halted. The anti-angiogenic properties of evodiamine are primarily due to its ability to target angiogenic factors and signaling pathways, including VEGF and NF-κB.[80] In OSCC, high mobility group box-1 (HMGB1) or receptor for advanced glycation end products (RAGE) is responsible for the invasive behavior of cancer cells. Silencing RAGE has been shown to suppress the proliferation of HSC-4 cells. Notably, evodiamine demonstrates remarkable antitumor effects by directly binding to HMGB1 and reducing the activity of the RAGE pathway by affecting the interaction between HMGB1 and RAGE. By targeting RAGE, evodiamine significantly inhibited the proliferation, invasion, and angiogenesis of OSCC cells by affecting the AGE/RAGE signal transduction system.[81] In addition, evodiamine has been found to inhibit tumor growth and the expression of glutamyl-prolyl tRNA synthetase (EPRS), highly expressed in OSCC, in a xenograft model. It also impacts cell proliferation, glutamine metabolism, and EPRS expression in Cal27 and SAS cell lines.[82] A nanocomposite system comprising folic acid-modified graphene quantum dots loaded with evodiamine showed a high growth inhibitory rate of OSCC cells, exceeding 50% ($P < 0.01$). In a xenograft model using tumor-bearing nude mice, graphene quantum dots functionalized with folic acid loaded with 10% evodiamine significantly reduced tumor volume by 19%.[83]

Sumadain C, a naturally occurring monoterpene-chalcone conjugate derived from plant sources, has garnered significant attention due to its potential cytotoxic effects on various cancers. It induces programmed cell death in cancer cells through multiple apoptotic pathways, leading to cytotoxic processes. Sumadain C modulates pro-apoptotic proteins, inhibits anti-apoptotic proteins, and triggers mitochondrial malfunction, resulting in caspase activation and DNA breakage. In addition, it causes cell cycle arrest by disrupting numerous angiogenic signaling pathways and suppressing tumor vascularization.[84] It hinders cell survival, proliferation, and metastasis by modifying carcinogenic signaling pathways. Notably, sumadain C demonstrated potent cytotoxicity effects against three cell lines with IC_{50} values of 13.00, 15.93, and 12.78 μg/mL, respectively.[85] For comparison, cisplatin, used as a positive control, had IC_{50} values of 13.18, 31.91, and 16.39 μg/mL, respectively.[85] B-cell lymphoma 2 was found to be the target of sumadain C through network pharmacology techniques, like molecular docking, and was verified by a homogeneous time-resolved fluorescence assay.[86]

The selected phytocompounds have demonstrated promising anticancer effects in various cancer therapies. However, administering these compounds in combination with other chemotherapeutic treatments is expected to yield greater efficacy than using them alone. The synergistic interactions between phytocompounds and chemotherapeutic medications can enhance the overall effectiveness of the treatment and reduce the required dosage of conventional chemotherapeutic drugs. A combination of these drugs could be used to achieve better efficacy and effectiveness. Future studies should consider conducting *in vitro* experiments to analyze the effect of these phytocompounds on OSCC. The results may contribute significantly to the development and formulation of natural chemotherapeutic agents that can be administered alone or as an adjuvant.

5. Limitations

Although the *in silico* analysis was conducted comprehensively, the study has several limitations. The scoring functions used in molecular docking can produce inaccurate binding energies. In addition, the restricted sampling of both ligand and receptor conformations during pose prediction poses another challenge. Accurately modeling water molecules in the binding pocket also remains challenging. Furthermore, the variability in how docking tools treat the protein can further affect the results.

6. Conclusion

This study highlights the potential efficacy of phytocompounds with anticancer activity. The binding affinity of the selected phytocompounds is highly promising, suggesting that future experimental studies could develop them into chemotherapeutic agents. Integrating these phytocompounds into cancer treatment holds great promise for enhancing therapeutic outcomes and improving the overall well-being of affected patients. CADD emerges as a promising source of drug discovery, potentially revolutionizing cancer therapeutics.

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Conflict of interest

The authors declare no potential conflicts of interest to the research, authorship, and/or publication of this article.

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Ethics approval and consent to participate

This study was approved by the scientific review board with reference number SRB/PhD/OPATH-2001/22/047.

Consent for publication

Not applicable.

Availability of data

The data used in this work can be made available to the readers on request to the corresponding author.

References

1. Petersen PE. Strengthening the prevention of oral cancer: The WHO perspective. *Community Den Oral Epidemiol*. 2005;33(6):397-399.

   doi: 10.1111/j.1600-0528.2005.00251.x

2. Feller L, Lemmer J. Oral squamous cell carcinoma: Epidemiology, clinical presentation and treatment. *J Cancer Ther*. 2012;03(4):263-268.

   doi: 10.4236/jct.2012.34037

3. Sagar S, Ramani P. Tobacco as a trigger for oral squamous cell carcinoma arising in the background of oral submucous fibrosis: A clinicopathological study. *J Popul Ther Clin Pharmacol* 2023;30(6).

   doi: 10.47750/jptcp.2023.30.06.012

4. Nussbaumer S, Bonnabry P, Veuthey JL, Fleury-Souverain S. Analysis of anticancer drugs: A review. *Talanta*. 2011;85(5):2265-2289.

   doi: 10.1016/j.talanta.2011.08.034

5. Priyadarshini R, Ramasubramanian A, Ramani P, Doble M. Identification of novel potential herbal drug targets against beta-catenin in the treatment of oral squamous cell carcinoma. *Asian Pac J Cancer Prev* 2024;25(12):4181-4188.

   doi: 10.31557/apjcp.2024.25.12.4181

6. Dillard CJ, German JB. Phytochemicals: Nutraceuticals and human health. *J Sci Food Agric*. 2000;80(12):1744-1756.

   doi:10.1002/1097-0010(20000915)80:12<1744:AID-JSFA725>3.0.CO;2-W

7. Burcher JT, DeLiberto LK, Allen AM, Kilpatrick KL, Bishayee A. Bioactive phytocompounds for oral cancer prevention and treatment: A comprehensive and critical evaluation. *Med Res Rev*. 2023;43(6):2025-2085.

   doi: 10.1002/med.21969

8. Yadav N, Parveen S, Banerjee M. Potential of nano-phytochemicals in cervical cancer therapy. *Clin Chim Acta*. 2020;505:60-72.

   doi: 10.1016/j.cca.2020.01.035

9. Farrand L, Oh SW, Song YS, Tsang BK. Phytochemicals: A multitargeted approach to gynecologic cancer therapy. *BioMed Res Int*. 2014;2014:890141.

   doi: 10.1155/2014/890141

10. Liu J, Peng Y, Wei W. Cell cycle on the crossroad of tumorigenesis and cancer therapy. *Trends Cell Biol*. 2021;32(1):30-44.

    doi: 10.1016/j.tcb.2021.07.001

11. Li Z, Li D, Zhang G, *et al*. Methylation-associated silencing of MicroRNA-335 contributes tumor cell invasion and migration by interacting with RASA1 in gastric cancer. *Am J Cancer Res*. 2014;4(6):648-662.

12. Prior IA, Lewis PD, Mattos C. A comprehensive survey of RAS mutations in cancer. *Cancer Res*. 2012;72(10):2457-2467.

    doi: 10.1158/0008-5472.can-11-2612

13. Rodenhuis S, Slebos RJ, Boot AJ, *et al*. Incidence and possible clinical significance of K-ras oncogene activation in adenocarcinoma of the human lung. *Cancer Res*. 1988;48(20):5738-5741.

14. Vogelstein B, Fearon ER, Hamilton SR, *et al*. Genetic alterations during colorectal-tumor development. *New England J Med*. 1988;319(9):525-532.

    doi: 10.1056/nejm198809013190901

15. Shibata D, Almoguera C, Forrester K, *et al*. Detection of c-K-ras mutations in fine needle aspirates from human pancreatic adenocarcinomas. *Cancer Res*. 1990;50(4):1279-1283.

16. Lyu H, Li M, Jiang Z, Liu Z, Wang X. Correlate the TP53 mutation and the HRAS mutation with immune signatures in head and neck squamous cell cancer. *Comput Struct Biotechnol J*. 2019;17:1020-1030.

    doi: 10.1016/j.csbj.2019.07.009

17. Dou R, Zhang L, Lu T, *et al*. Identification of a novel HRAS variant and its association with papillary thyroid carcinoma. *Oncol Lett*. 2018;5(4):4511-4516.

    doi: 10.3892/ol.2018.7818

18. Cha Y, Erez T, Reynolds IJ, *et al*. Drug repurposing from the perspective of pharmaceutical companies. *Br J Pharmacol*. 2017;175(2):168-180.

    doi: 10.1111/bph.13798

19. Priyadarshini G, Sukumaran G, Dilipan E, Ramani P. Targeting oral cancer: *In silico* docking studies of phytochemicals on oncogenic molecular markers. *Asian Pac J Cancer Prev* 2024;25(6):2069-2075.

    doi: 10.31557/apjcp.2024.25.6.2069

20. Cui JJ, Tran-Dubé M, Shen H, *et al*. Structure based drug design of Crizotinib (PF-02341066), a potent and selective dual inhibitor of mesenchymal-epithelial transition factor (C-MET) kinase and Anaplastic Lymphoma Kinase (ALK). *J Med Chem*. 2011;54(18):6342-6363.

    doi: 10.1021/jm2007613

21. Cui W, Aouidate A, Wang S, Yu Q, Li Y, Yuan S. Discovering anti-cancer drugs via computational methods. *Front Pharmacol*. 2020;11:733.

    doi: 10.3389/fphar.2020.00733

22. Prada-Gracia D, Huerta-Yépez S, Moreno-Vargas LM. Application of computational methods for anticancer drug discovery, design, and optimization. *Bol Méd Hosp Infant Méx*. 2016;73(6):411-423.

    doi: 10.1016/j.bmhimx.2016.10.006

23. Spinello A, Ritacco I, Magistrato A. Recent advances in computational design of potent aromatase inhibitors: Open-eye on endocrine-resistant breast cancers. *Expert Opin Drug Discov*. 2019;14(10):1065-1076.

doi: 10.1080/17460441.2019.1646245

24. Ohbayashi N, Murayama K, Kato-Murayama M, *et al*. Structural basis for the inhibition of cyclin G-Associated kinase by gefitinib. *ChemistryOpen*. 2018;7(9):713-719.

doi: 10.1002/open.201800177

25. Masago K, Togashi Y, Fukudo M, *et al*. Good clinical response to erlotinib in a non-small cell lung cancer patient harboring multiple brain metastases and a double active somatic epidermal growth factor gene mutation. *Case Rep Oncol*. 2010;3(2):98-105.

doi: 10.1159/000310830

26. Xia W, Liu Z, Zong R, *et al*. Truncated ERBB2 expressed in tumor cell nuclei contributes to acquired therapeutic resistance to ERBB2 kinase inhibitors. *Mol Cancer Ther*. 2011;10(8):1367-1374.

doi: 10.1158/1535-7163.mct-10-0991

27. Asmane I, Céraline J, Duclos B, *et al*. New strategies for medical management of castration-resistant prostate cancer. *Oncology*. 2011;80(1-2):1-11.

doi: 10.1159/000323495

28. Roskoski R Jr. STI-571: An anticancer protein-tyrosine kinase inhibitor. *Biochem Biophys Res Commun*. 2003;309(4):709-717.

doi: 10.1016/j.bbrc.2003.08.055

29. Lam B, Arikawa Y, Cramlett J, *et al*. Discovery of TAK-659 an orally available investigational inhibitor of Spleen Tyrosine Kinase (SYK). *Bioorg Med Chem Lett*. 2016;26(24):5947-5950.

doi: 10.1016/j.bmcl.2016.10.087

30. Wang M, Yang JCH, Mitchell PL, *et al*. Sunvozertinib, a selective EGFR inhibitor for previously treated non-small cell lung cancer with EGFR exon 20 insertion mutations. *Cancer Discov*. 2022;12(7):1676-1689.

doi: 10.1158/2159-8290.cd-21-1615

31. Tron AE, Belmonte MA, Adam A, *et al*. Discovery of Mcl-1-specific inhibitor AZD5991 and preclinical activity in multiple myeloma and acute myeloid leukemia. *Nat Commun*. 2018;9(1):5341.

doi: 10.1038/s41467-018-07551-w

32. Wang L, Yang C, Xie C, *et al*. Pharmacologic characterization of fluzoparib, a novel poly(ADP-ribose) polymerase inhibitor undergoing clinical trials. *Cancer Sci*. 2019;110(3):1064-1075.

doi: 10.1111/cas.13947

33. Sahu RK, Verma VV, Kumar A, Tandon S, Das BC, Hedau ST. *In silico* prediction and interaction of resveratrol on methyl-CpG binding proteins by molecular docking and MD simulations study. *RSC Adv*. 2022;12(18):11493-11504.

doi: 10.1039/d2ra00432a

34. Shen LA, Peng X, Bao Y, *et al*. Design, synthesis and biological evaluation of quercetin derivatives as novel β-catenin/B-cell lymphoma 9 protein-protein interaction inhibitors. *Eur J Med Chem*. 2022;247:115075.

doi: 10.1016/j.ejmech.2022.115075

35. Olotu FA, Agoni C, Adeniji E, Abdullahi M, Soliman ME. Probing gallate-mediated selectivity and high-affinity binding of epigallocatechin gallate: A way-forward in the design of selective inhibitors for anti-apoptotic Bcl-2 proteins. *Appl Biochem Biotechnol*. 2018;187(3):1061-1080.

doi: 10.1007/s12010-018-2863-7

36. Simanshu DK, Nissley DV, McCormick F. RAS proteins and their regulators in human disease. *Cell*. 2017;170(1):17-33.

doi: 10.1016/j.cell.2017.06.009

37. Hancock JF. Ras proteins: Different signals from different locations. *Nat Rev Mol Cell Biol*. 2003;4(5):373-385.

doi: 10.1038/nrm1105

38. Ahmadian MR, Stege P, Scheffzek K, Wittinghofer A. Confirmation of the arginine-finger hypothesis for the GAP-stimulated GTP-hydrolysis reaction of Ras. *Nat Struct Biol*. 1997;4(9):686-689.

doi: 10.1038/nsb0997-686

39. Gimple RC, Wang X. RAS: Striking at the core of the oncogenic circuitry. *Front Oncol*. 2019;9:965.

doi: 10.3389/fonc.2019.00965

40. Sebti SM. Protein farnesylation: Implications for normal physiology, malignant transformation, and cancer therapy. *Cancer Cell*. 2005;7(4):297-300.

doi: 10.1016/j.ccr.2005.04.005

41. Sobocińska J, Roszczenko-Jasińska P, Ciesielska A, Kwiatkowska K. Protein palmitoylation and its role in bacterial and viral infections. *Front Immunol*. 2018;8:2003.

doi: 10.3389/fimmu.2017.02003

42. Das T, Yount JS, Hang HC. Protein S-palmitoylation in immunity. *Open Biol*. 2021;11(3):200411.

doi: 10.1098/rsob.200411

43. Guan X, Fierke CA. Understanding protein palmitoylation: Biological significance and enzymology. *Sci China Chem*. 2011;54(12):1888-1897.

doi: 10.1007/s11426-011-4428-2

44. Wang Y, Wang F. Post-Translational modifications of deubiquitinating enzymes: Expanding the ubiquitin code. *Front Pharmacol*. 2021;12:685011.

doi: 10.3389/fphar.2021.685011

45. Grillari J, Grillari-Voglauer R, Jansen-Dürr P. Madame curie bioscience database. In: *Post-Translational Modification of Cellular Proteins by Ubiquitin and Ubiquitin-Like Molecules: Role in Cellular Senescence and Ageing*. Austin, TX: Landes Bioscience; 2013. Available from: https://www.ncbi.nlm.nih.gov/books/NBK28487

46. Wang Y, Shi M, Feng H, *et al*. Structural insights into non-canonical ubiquitination catalyzed by side. *Cell*. 2018;173(5):1231-1243.e16.

doi: 10.1016/j.cell.2018.04.023

47. Cherfils J, Zeghouf M. Regulation of small GTPASes by GEFs, GAPs, and GDIs. *Physiol Rev*. 2013;93(1):269-309.

doi: 10.1152/physrev.00003.2012

48. Rowinsky EK, Windle JJ, Von Hoff DD. RAS protein farnesyltransferase: A strategic target for anticancer therapeutic development. *J Clin Oncol*. 1999;17(11):3631-3652.

doi: 10.1200/jco.1999.17.11.3631

49. Rane N, Prendergast GC. Farnesyltransferase inhibitors: Mechanism and applications. *Expert Opin Investig Drugs*. 2001;10(12):2105-2116.

doi: 10.1517/13543784.10.12.2105

50. Oh SH, Kim WY, Kim JH, *et al*. Identification of insulin-like growth factor binding protein-3 as a farnesyl transferase inhibitor SCH66336-induced negative regulator of angiogenesis in head and neck squamous cell carcinoma. *Clin Cancer Res*. 2006;12(2):653-661.

doi: 10.1158/1078-0432.ccr-05-1725

51. Kelland LR, Smith V, Valenti M, *et al*. Preclinical antitumor activity and pharmacodynamic studies with the farnesyl protein transferase inhibitor R115777 in human breast cancer. *Clin Cancer Res*. 2001;7(11):3544-3550.

52. Martin LA, Head JE, Pancholi S, *et al*. The farnesyltransferase inhibitor R115777 (tipifarnib) in combination with tamoxifen acts synergistically to inhibit MCF-7 breast cancer cell proliferation and cell cycle progression *in vitro* and *in vivo*. *Mol Cancer Ther*. 2007;6(9):2458-2467.

doi: 10.1158/1535-7163.mct-06-0452

53. Cadelis MM, Bourguet-Kondracki ML, Dubois J, Valentin A, Barker D, Copp BR. Discovery and preliminary structure-activity relationship studies on tecomaquinone I and tectol as novel farnesyltransferase and plasmodial inhibitors. *Bioorg Med Chem*. 2016;24(14):3102-3107.

doi: 10.1016/j.bmc.2016.05.024

54. Hedvat M, Huszar D, Herrmann A, *et al*. The JAK2 inhibitor AZD1480 potently blocks STAT3 signaling and oncogenesis in solid tumors. *Cancer Cell*. 2009;16(6):487-497.

doi: 10.1016/j.ccr.2009.10.015

55. Hosseini A, Gharibi T, Marofi F, Javadian M, Babaloo Z, Baradaran B. Janus kinase inhibitors: A therapeutic strategy for cancer and autoimmune diseases. *J Cell Physiol*. 2020;235(9):5903-5924.

doi: 10.1002/jcp.29593

56. Aittomäki S, Pesu M. Therapeutic targeting of the JAK/STAT pathway. *Basic Clin Pharmacol Toxicol*. 2013;114(1):18-23.

doi: 10.1111/bcpt.12164

57. Hu Y, Hong Y, Xu Y, Liu P, Guo DH, Chen Y. Inhibition of the JAK/STAT pathway with ruxolitinib overcomes cisplatin resistance in non-small-cell lung cancer NSCLC. *Apoptosis*. 2014;19(11):1627-1636.

doi: 10.1007/s10495-014-1030-z

58. Lin CM, Cooles FA, Isaacs JD. Basic mechanisms of JAK inhibition. *Mediterr J Rheumatol*. 2020;31(Suppl 1):100-104.

doi: 10.31138/mjr.31.1.100

59. Gill K, Kumar R, Mohanti BK, Dey S. Assessment of p38α in peripheral blood mononuclear cells (PBMC): A potential blood protein marker of head and neck squamous cell carcinoma. *Clin Transl Oncol*. 2013;15(11):969-973.

doi: 10.1007/s12094-013-1031-3

60. Monnerat C, Henriksson R, Chevalier TL, *et al*. Phase I study of PKC412 (N-benzoyl-staurosporine), a novel oral protein kinase C inhibitor, combined with gemcitabine and cisplatin in patients with non-small-cell lung cancer. *Ann Oncol*. 2004;15(2):316-323.

doi: 10.1093/annonc/mdh052

61. Lee D, Kwak HJ, Kim BH, *et al*. Brevilin A isolated from *Centipeda minima* induces apoptosis in human gastric cancer cells via an extrinsic apoptotic signaling pathway. *Plants* (*Basel*). 2022;11(13):1658.

doi: 10.3390/plants11131658

62. Chen X, Du Y, Nan J, *et al*. Brevilin A, a novel natural product, inhibits janus kinase activity and blocks STAT3 signaling in cancer cells. *PLoS One*. 2013;8(5):e63697.

doi: 10.1371/journal.pone.0063697

63. Khan M, Maryam A, Saleem MZ, *et al*. Brevilin A induces ROS-dependent apoptosis and suppresses STAT3 activation by direct binding in human lung cancer cells. *J Cancer*. 2020;11(13):3725-3735.

doi: 10.7150/jca.40983

64. Guan X. Cancer metastases: Challenges and opportunities. *Acta Pharm Sin B*. 2015;5(5):402-418.

doi: 10.1016/j.apsb.2015.07.005

65. Mitra SK, Schlaepfer DD. Integrin-regulated FAK-Src signaling in normal and cancer cells. *Curr Opin Cell Biol*. 2006;18(5):516-523.

doi: 10.1016/j.ceb.2006.08.011

66. Augoff K, Hryniewicz-Jankowska A, Tabola R. Invadopodia:

Clearing the way for cancer cell invasion. *Ann Transl Med*. 2020;8(14):902.

doi: 10.21037/atm.2020.02.157

67. Windham TC, Parikh NU, Siwak DR, *et al*. Src activation regulates anoikis in human colon tumor cell lines. *Oncogene*. 2002;21(51):7797-7807.

doi: 10.1038/sj.onc.1205989

68. Playford MP, Schaller MD. The interplay between Src and integrins in normal and tumor biology. *Oncogene*. 2004;23(48):7928-7946.

doi: 10.1038/sj.onc.1208080

69. Leong HS, Robertson AE, Stoletov K, *et al*. Invadopodia are required for cancer cell extravasation and are a therapeutic target for metastasis. *Cell Rep*. 2014;8(5):1558-1570.

doi: 10.1016/j.celrep.2014.07.050

70. Eckert MA, Lwin TM, Chang AT, *et al*. Twist1-induced invadopodia formation promotes tumor metastasis. *Cancer Cell*. 2011;19(3):372-386.

doi: 10.1016/j.ccr.2011.01.036

71. Campone M, Bondarenko I, Brincat S, *et al*. Phase II study of single-agent bosutinib, a Src/Abl tyrosine kinase inhibitor, in patients with locally advanced or metastatic breast cancer pretreated with chemotherapy. *Ann Oncol*. 2011;23(3):610-617.

doi: 10.1093/annonc/mdr261

72. Gucalp A, Sparano JA, Caravelli J, *et al*. Phase II trial of saracatinib (AZD0530), an Oral SRC-inhibitor for the treatment of patients with hormone receptor-negative metastatic breast cancer. *Clin Breast Cancer*. 2011;11(5):306-311.

doi: 10.1016/j.clbc.2011.03.021

73. Mackay HJ, Au HJ, McWhirter E, *et al*. A phase II trial of the Src kinase inhibitor saracatinib (AZD0530) in patients with metastatic or locally advanced gastric or Gastro Esophageal Junction (GEJ) adenocarcinoma: A trial of the PMH phase II consortium. *Invest New Drugs*. 2011;30(3):1158-1163.

doi: 10.1007/s10637-011-9650-4

74. Lee S, Park S, Ryu JS, *et al*. c-Src inhibitor PP2 inhibits head and neck cancer progression through regulation of the epithelial-mesenchymal transition. *Exp Biol Med* (*Maywood*). 2022;248(6):492-500.

doi: 10.1177/15353702221139183

75. Min HY, Jung Y, Park KH, Oh WK, Lee HY. Erybraedin A is a potential Src inhibitor that blocks the adhesion and viability of non-small-cell lung cancer cells. *Biochem Biophys Res Commun*. 2018;502(1):145-151.

doi: 10.1016/j.bbrc.2018.05.137

76. Luo C, Ai J, Ren E, *et al*. Research progress on evodiamine, a bioactive alkaloid of Evodiae fructus: Focus on its anti-cancer activity and bioavailability (Review). *Exp Ther Med*. 2021;22(5):1327.

doi: 10.3892/etm.2021.10762

77. Mohan V, Agarwal R, Singh RP. A novel alkaloid, evodiamine causes nuclear localization of cytochrome-c and induces apoptosis independent of p53 in human lung cancer cells. *Biochem Biophys Res Commun*. 2016;477(4):1065-1071.

doi: 10.1016/j.bbrc.2016.07.037

78. Hong Z, Wang Z, Zhou B, *et al*. Effects of evodiamine on PI3K/Akt and MAPK/ERK signaling pathways in pancreatic cancer cells. *Int J Oncol*. 2020;56:783-793.

doi: 10.3892/ijo.2020.4956

79. Ogasawara M, Matsunaga T, Takahashi S, Saiki I, Suzuki H. Anti-invasive and metastatic activities of evodiamine. *Biol Pharm Bull*. 2002;25(11):1491-1493.

doi: 10.1248/bpb.25.1491

80. Yu H, Jin H, Gong W, Wang Z, Liang H. Pharmacological actions of multi-target-directed evodiamine. *Molecules*. 2013;18(2):1826-1843.

doi: 10.3390/molecules18021826

81. Ren L, Lou Y, Sun M. The anti-tumor effects of evodiamine on oral squamous cell carcinoma (OSCC) through regulating advanced glycation end products (AGE)/receptor for advanced glycation end products (RAGE) pathway. *Bioengineered*. 2021;12(1):5985-5995.

doi: 10.1080/21655979.2021.1972082

82. Lin L, Lv S, Ren H, *et al*. Evodiamine inhibits EPRS expression to regulate glutamate metabolism and proliferation of oral squamous cell carcinoma cells. *Kaohsiung J Med Sci*. 2024;40(4):348-359.

doi: 10.1002/kjm2.12803

83. Ma Y, Liu Y, Wang Y, *et al*. The nanocomposite system comprising folic acid-modified graphene quantum dots loaded with evodiamine in the treatment of oral squamous cell carcinoma. *Mater Des*. 2022;220:110838.

doi: 10.1016/j.matdes.2022.110838

84. Ma XN, Xie CL, Miao Z, Yang Q, Yang XW. An overview of chemical constituents from *Alpinia* species in the last six decades. *RSC Adv*. 2017;7(23):14114-14144.

doi: 10.1039/c6ra27830b

85. Hua SZ, Luo JG, Wang XB, Wang JS, Kong LY. Two novel monoterpene-chalcone conjugates isolated from the seeds of *Alpinia katsumadai*. *Bioorg Med Chem Lett*. 2009;19(10):2728-2730.

doi: 10.1016/j.bmcl.2009.03.117

86. You HL, Zhou B, Guo MJ, *et al*. Monoterpene-chalcone conjugates and diarylheptanoids isolated from the seeds of *Alpinia katsumadai* hayata with cytotoxic activity. *Phytochemistry*. 2024;225:114197.

doi: 10.1016/j.phytochem.2024.114197