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Molecular Docking and ADME Profiling of 5-(Substituted Benzylidene)-2-(Arylamino)-1,3-Thiazol-4(5H)-ones: Insights into Pharmacokinetics and Binding Interactions

Prajwal Lourdes Lobo[1], Boja Poojary[2], Lydia J.[3], M. Suhas[4], P. A. Vivekanand[5,*], L. Ramya[5], B. Kalaivani[5], S. Harikumar[6], S. Thennarasu[7], P. Kamaraj[8], S. Jayanthi[9,*], Navaneetha K. Kumar[10], R. Arulnangai[11], Periasamy Anbu[12*]

[1]*Maharani Cluster University, Bangalore, India-560001*

[2]*Department of Chemistry, Mangalore University, Mangalagangothri, India*

[3]*Veterinary Assistant Surgeon, Rajakkamangalam, Nagercoil Division, Tamil Nadu, India*

[4]*Department of Anaesthesiology, Saveetha Medical College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai, 602 105, Tamil Nadu, India*

[5]*Centre for Catalysis Research, Department of Chemistry, Saveetha Engineering College, Chennai, India-602105*

[6]*Chemistry Division, Department of Humanities and Sciences, Rajalakshmi Institute of Technology , Chennai, India*

[7]*Department of Physics, Saveetha Engineering College, Chennai, India-602105*

[8]*Department of Chemistry, Bharath Institute of Higher Education and Research, Chennai 600 073, India*

[9]*Cauvery College for Women, Trichy, Tamil Nadu, India*

[10]*Saveetha Medical College & Hospital, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Thandalam, Chennai - 602 105, Tamil Nadu, India*

[11] *PG and Research Department of Chemistry, Jamal Mohammed College, Trichy India*

[12]*Department of Microbiology, Saveetha Medical College & Hospital, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Thandalam, Chennai - 602 105, Tamil Nadu, India*

Abstract

In the quest for effective cancer therapeutics, the optimization of pharmacokinetics, toxicity profiles, and efficacy is crucial. This study introduces a novel series of 5-(substituted benzylidene)-2-(arylamino)-1,3-thiazol-4(5H)-ones, synthesized to explore their potential as anti-cancer agents. These compounds were specifically designed based on the promising anti-tumour activity of 5-arylidene-4-thiazolidinone derivatives, known for their efficacy against MDA-MB-231 (human breast cancer cell line). To assess these new thiazol-4-ones, we used sophisticated in silico methods to perform pharmacokinetic ADME predictions and molecular docking simulations. Our molecular docking studies utilized FlexX to compare the binding affinities of these compounds with known drugs: Gestrinone (targeting EGFR alpha for breast cancer), Vandetanib (targeting VEGFR-2), and KU0058948 (targeting Poly ADP ribose polymerase for ovarian cancer). These comparative analyses revealed significant interactions with these key cancer targets. In addition, ADME predictions were performed using the iLOG predictor from Swiss ADMET software, demonstrating favourable properties for absorption, distribution, and bioavailability. Interestingly, compounds with fluorine

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****Corresponding Authors: pavivek@rediffmail.com; jaya_1110@yahoo.co.in; anbu25@yahoo.com***

substitutions at positions 2 or 4 of the acylamino ring showed encouraging activity and satisfied Lipinski and Veber's rules-based drug-likeness requirements, indicating that they could make good candidates for therapeutics. Furthermore, these compounds showed low toxicity levels, enhancing their suitability for further development.

Keywords: *ADME, Bezylidene, Binding, Docking, EGFR Alpha, Thiazole-4-Ones, VGEFR 2.*

Introduction

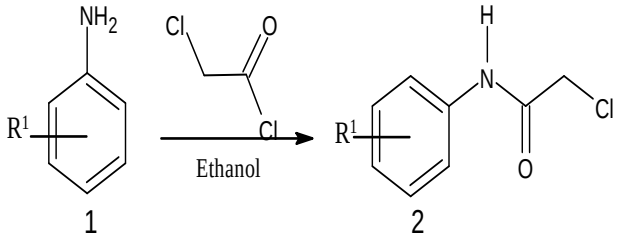
Cancer is characterized by the uncontrolled proliferation of cells that invade and destroy normal tissues, often leading to life-threatening consequences if not effectively managed [1]. It remains a significant global health challenge, with over ten million new cases diagnosed annually, contributing to more than six million deaths each year [2]. Despite the advancements in cancer treatment, chemotherapy remains the primary therapeutic approach. However, its effectiveness is often limited by adverse side effects, such as bone marrow suppression, alopecia, drug-induced secondary malignancies, and hepatotoxicity, as well as a restricted range of available anti-cancer drugs [3]. Among the diverse range of heterocyclic compounds explored for their therapeutic potential, 4-thiazolidinone derivatives have emerged as notable candidates due to their broad spectrum of biological activities [4] It has been demonstrated that these compounds exhibit antibacterial, antifungal, antiviral, anticancer, and anti-inflammatory qualities [5–11]. Particularly, 5-arylidene-4-thiazolidinone derivatives have shown notable anticancer activity against several cancer cell lines, such as human breast cancer (MDA-MB-231), paclitaxel-resistant (H460taxR), non-small cell lung cancer (H460), and human colon cancer (HT-29) [12].

In recent years, computational methods have become integral to rational drug design, offering an efficient, cost-effective approach to developing new and potent therapeutic molecules. Predicting the structure and binding affinity of ligand-protein interactions during drug discovery is made possible by these *in silico* methods, especially molecular docking studies. Molecular docking involves several key components: a target protein structure (with or without a bound ligand), a set of molecules or a database of existing or virtual compounds, and a computational framework to perform docking simulations and scoring. The accuracy of docking simulations is determined by the algorithm's ability to predict the correct conformation (http://poseview. zbh.uni-hamburg.de) and alignment of a ligand relative to its target protein, ideally mirroring experimental observations. By determining the lowest energy value and classifying the protein-ligand complexes according to their binding affinities, scoring algorithms determine the most advantageous binding position [13]. This study focuses on the binding interactions of newly synthesized 5-(substituted benzylidene)-2-(arylamino)-1,3-thiazol-4(5H)-ones with three key cancer-related drug targets: Poly[ADP-ribose]polymerase-1 (targeted for tumour treatment, PDB ID 3l3m), Vascular Endothelial Growth Factor Receptor-2 (targeted for angiogenesis, 3CPC), and the Estrogen Receptor (targeted for breast cancer). The results of these docking studies provide insights into the pharmacophoric features essential for further optimization and development of thiazol-4(5H)-one derivative as a potential anticancer agent.

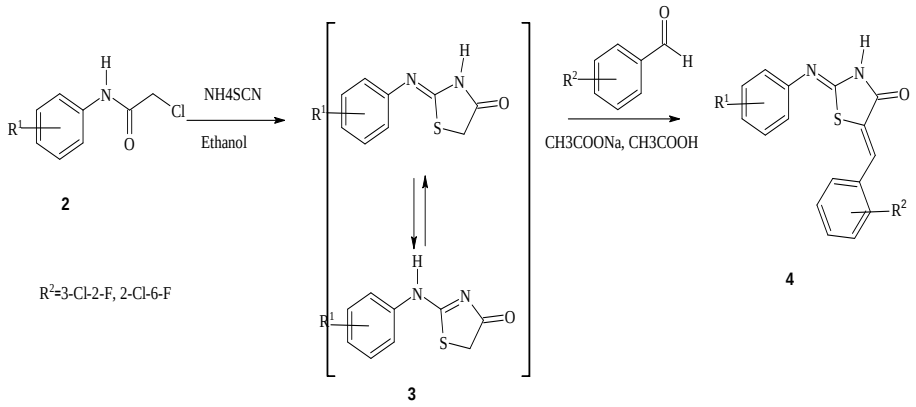
Materials and Methods

Synthesis of 5-(Substituted Benzyliledene)-2-(Arylamino)-1,3-Thiazol-4(5H)-Ones

Target ligands were synthesized by acylating the substituted anilines (1) with a two-acyl carbon linker in a dry ethanol medium using chloro-acetyl chloride, resulting in 2-chloro-N-arylacetamides (2). These 2-chloro-N-arylacetamides (2) yielded the cyclized 2-(phenylamino)-1,3-thiazol-4(5H)-one and 2-(phenylimino)-1,3-thiazolidin-4-one (3) by reacting with ammonium thiocyanates. These thiazolidine-4-ones were converted to (substituted benzylidene)-2-(arylamino)-1,3-thiazol-4(5H)-one (4) by condensing them with substituted benzaldehydes in acetic acid media while anhydrous sodium acetate was present [14-16]. The synthetic route is shown in Schemes I and II.



Scheme 1. Synthesis of 2-chloro-*N*-arylacetamides



Scheme 2. Synthesis of (Substituted Benzylidene)-2-(Arylamino)-1,3-Thiazol-4(5*H*)-One

The open capillary method measured the synthesized compounds' melting points without any adjustments for atmospheric pressure. Infrared (IR) spectra were registered using a Shimadzu FT-IR-157 spectrophotometer, with samples prepared in potassium bromide (KBr) pellets. Proton nuclear magnetic resonance (1H NMR) spectra were recorded using a 400 MHz spectrometer (Bruker Avance), with tetramethylsilane (TMS) as the internal standard. Mass spectrometry was conducted using an Agilent Technology LC mass spectrometer, while direct analysis in real time (DART) mass spectra was recorded on a JEOL-AccuTOF JMS-T100LC mass spectrometer. For DART analysis, samples were exposed in front of the DART source, with ionization facilitated by dry helium at a flow rate of 4 LPM and a temperature of 350 °C. Elemental analyses for carbon, hydrogen, nitrogen, and sulfur (CHNS) were performed using a CHNS Elementar Vario EL-III analyzer. Using thin-layer chromatography (TLC) on silica gel plates, the reactions' the synthesis process.

3D Modeling and Ligand Minimization

The synthesized compounds were modelled using ArgusLab software [17]. Geometric optimization was performed for synthesized compounds and standard drugs using the AM1-NDDO Hamiltonian Quantum Mechanics Algorithm to minimize energy. The

optimization was performed using the Broyden Fletcher Goldfarb Shanno (BFGS) algorithm, an iterative technique for solving unconstrained nonlinear optimization problems. The Restricted Hartree -Fock (RHF) method was used with a closed-shell configuration for the self-consistent field (SCF) calculations, with a maximum number of SCF iterations set at 200 and convergence criteria set at 1.5936e-013 au (or 10^-10 kcal/mol). The ligand compounds' energy was minimized iteratively until the lowest feasible energy was reached, and the final energy-minimized molecular files were saved for further docking studies.

Docking Parameters

A FlexX module is utilized for target-ligand docking in the software package developed by BioSolveIT (LeadIT 2.1.2 version), which is an interactive structure-based platform for protein-ligand interaction prediction [18]. The FlexX tool visualizes the geometric arrangement of ligand-protein complexes while calculating the associated binding energies, operating under the assumption that the protein structure remains rigid during docking [19, 20]. The receptor structures for Poly[ADP-ribose] polymerase-1 (3L3M), Vascular Endothelial growth Factor Receptor-2 (3CPC), and the estrogen receptor (3LO3) were obtained from the RCSB Protein Data Bank (https://www.rcsb.org) and saved in PDB file format. To prepare the binding site for docking, the PDB files were modified and uploaded at the load or prepare receptor option. The docking procedure employed a hybrid algorithm that integrates enthalpy and entropy calculations to drive ligand-binding interactions [21]. Protein-ligand clash scoring was based on access scaling with default threshold parameters, and a maximum allowable overlap volume of 12 Å was used for clash handling.

Absorption, Distribution, Metabolism, and Excretion (ADME) Analysis

To evaluate the drug-likeness and pharmacokinetic properties of the synthesized ligands, a suite of algorithms was employed to predict their potential as drug candidates and their effects on biological systems. The reference drugs KU0058948, Vandetanib, and Gestrinone were included in this study for comparison. The Swiss ADME Tool and the iLOG predictor were utilized to estimate the drug ability of the ligands. The Swiss ADME Tool, accessible at http://www.swissadme.ch/, comprehensively analyzes physicochemical descriptors and predicts pharmacokinetic characteristics, including solubility, permeability, and bioavailability. The tool also assesses the drug-like nature of small molecules based on established criteria [22].

Results and Discussion

The synthesized compounds were characterized using elemental analyses, IR spectroscopy, 1H NMR, and mass spectral data, with the detailed characterization results provided in Table 1. For compound 4d (5-(2-chloro-6-fluorobenzylidene)-2-[(3-chloro-4-fluorophenyl) amino]-1,3-thiazol4(5H)-one), the IR spectrum exhibited strong absorption bands at 3201 cm^{-1} for the NH stretch, 3001 cm^{-1} for aromatic C–H, 1676 cm^{-1} for C=O, and 1625 cm^{-1} for C=N. The 1H NMR spectrum at 400 MHz revealed the aromatic protons of the 3-chloro-4-fluorophenyl and 2-chloro-6-fluorophenyl moieties resonating as a multiplet in the region of δ 6.98–7.63. The benzylidene proton appeared as a singlet at δ 8.112, while a singlet at δ 12.56 characterized the NH proton. The LC-MS analysis confirmed the molecular formula $C_{16}H_8N_2Cl_2F_2OS$, with an M+1 peak observed at m/z 385.

In the case of compound 4g (5-(3-chloro-2-fluorobenzylidene)-2-[(4-fluorophenyl) amino]-1,3-thiazol-4(5H)-one), the IR

spectrum displayed absorption bands at 3192 cm^{-1} for NH, 3040 cm^{-1} for aromatic C–H, 1682 cm^{-1} for C=O, 1549 cm^{-1} for C=N, and 1160 cm^{-1} for C–F. The 1H NMR spectrum showed the aromatic protons of the 4-fluorophenyl moiety resonating as two multiplets in the regions of δ 7.20–7.24 and δ 7.79–7.82. The protons of the 3-chloro-4-fluorophenyl moiety appeared as multiplets at δ 7.43–7.47 and δ 7.91–7.93, while the benzylidene proton was noted as a singlet at δ 8.223. The NH-proton was also identified as a singlet at δ 12.35. The DART mass spectra confirmed the molecular formula $C_{16}H_9N_2ClF_2OS$, with a molecular ion peak recorded at m/z 350.

Table 1. Characterization Data of Compounds (4a-j)

Compounds	R^1	R^2	Mol. formula	Mol. Wt	M. P. (^{0}C).	Yield (%)	Smiles
4a	2,4,5-(Cl)3	2-Cl,6-F	C16H7N2Cl4FOS	436.11	152-154	75	O=C1N=C(S/C/1=C\c1c(F)cccc1Cl)Nc1cc(Cl)c(cc1Cl)Cl
4b	2, 4-(CH3)2	2-Cl,6-F	C18H14N2ClFOS	360.83	174-176	87	Cc1ccc(c(c1)C)NC1=NC(=O)/C(=C/c2c(F)cccc2Cl)/S1
4c	2, 4-(CH3)2	3-Cl,2-F	C18H14N2ClFOS	360.83	202-204	87	Cc1ccc(c(c1)C)NC1=NC(=O)/C(=C/c2cccc(c2F)Cl)/S1
4d	3-Cl,4-F	2-Cl,6-F	C16H8N2Cl2F2OS	385.21	204-206	82	O=C1N=C(S/C/1=C\c1c(F)cccc1Cl)Nc1ccc(c(c1)Cl)F
4e	4-Cl	2-Cl,6-F	C16H9N2Cl2FOS	367.22	214-216	88	O=C1N=C(S/C/1=C\c1c(F)cccc1Cl)Nc1ccccc1Cl
4f	2-Cl	2-Cl,6-F	C16H9N2Cl2FOS	367.22	198-200	78	Clc1ccc(cc1)NC1=NC(=O)/C(=C/c2c(F)cccc2Cl)/S1
4g	4-F	3-Cl,2-F	C16H9N2ClF2OS	350.77	210-212	84	Fc1ccc(cc1)NC1=NC(=O)/C(=C/c2cccc(c2F)Cl)/S1
4h	2-CF3	3-Cl,2-F	C17H9N2ClF4OS	400.77	208-210	77	O=C1N=C(S/C/1=C\c1c(F)cccc1Cl)Nc1ccccc1C(F)(F)F
4i	2, 4-Cl2	2-Cl,6-F	C16H8N2Cl3FOS	401.66	200-202	85	Clc1ccc(c(c1)Cl)NC1=NC(=O)/C(=C/c2c(F)cccc2Cl)/S1
4j	2-CF3	2-Cl,6-F	C17H9N2ClF4OS	400.77	162-164	73	O=C1N=C(S/C/1=C\c1cccc(c1F)Cl)Nc1ccccc1C(F)(F)F

The Swiss ADME analysis provided insights into the synthesized compounds' physicochemical properties, lipophilicity, and pharmacokinetics. These properties, which include solubility and bioavailability predictions, were assessed based on uncharged SMILES representations. The molar refractivity of the synthesized ligands fell within acceptable ranges, and the topological polar surface area (TPSA) values indicated favourable characteristics for drug-like behaviour (Table 2).

Lipophilicity

Lipophilicity was evaluated using the logarithm of the n-octanol/water partition coefficient (Log P), as determined by the Consensus Log P_o/w descriptor from SwissADME. Log P is critical for predicting membrane permeability and tissue distribution

[23]. A Log P value between 0 and 3 is generally indicative of good oral bioavailability [24]. The synthesized thiazol-4(5H)-ones exhibited Log P values ranging from 4.41 to 5.77, as detailed in Table 3. Notably, these values suggest higher lipophilicity, which could impact solubility and absorption. Predictions of solubility (Log Sw) and lipophilicity demonstrated a relationship, reinforcing the need for careful consideration of these properties in drug design.

Table 2. Physicochemical Properties of Ligands (4a-j)

Ligands	Molecular Wtg/mol	Heavy Atoms	RotatableBonds	H-Bond Acceptors	H-Bond Donors	Molar refracts	TPSA Å²
4a	436.11	25	3	3	1	107.66	66.76
4b	401.67	24	3	3	1	102.65	66.76
4c	360.83	24	3	3	1	102.56	66.76
4d	385.22	24	3	4	1	97.60	66.76
4e	367.22	23	3	3	1	97.64	66.76
4f	367.22	23	3	3	1	97.64	66.76
4g	350.77	23	3	4	1	92.59	66.76
4h	360.83	24	3	3	1	102.56	66.76
4i	400.78	26	4	6	1	97.63	66.76
4j	400.78	26	4	6	1	97.63	66.76
KU 058948	380.42	28	4	5	2	112.61	78.09
Gestrinone	308.41	23	1	2	1	92.48	37.30
Vandetanib	475.35	30	6	6	1	123.26	59.51

Table 3. Lipophilicity Properties of the Ligands (4a-j)

Ligands	Log Po/w logo	Log Po/w XLOGP3	Log Po/w SLOGP	Log Po/w MLOGP	Log Po/w SILICOS-IT	ConsensusLog Po/w
4a	3.53	6.44	6.26	5.70	6.71	5.77
4b	3.34	5.81	5.61	4.80	6.26	5.16
4c	3.36	5.29	4.92	4.26	6.00	4.77
4d	2.99	5.29	5.51	5.09	6.04	4.98
4e	3.08	5.19	4.95	4.30	5.62	4.63
4f	3.21	5.19	4.95	4.71	5.62	4.74
4g	2.94	4.66	4.86	4.18	5.40	4.41
4h	3.37	5.29	4.92	4.26	6.00	4.77
4i	3.26	5.44	6.47	5.05	6.04	5.25
4j	3.20	5.44	6.47	5.05	6.04	5.24
KU 0058948	2.51	1.88	1.75	2.92	4.07	2.62
Gestrinone	3.10	2.16	3.80	3.77	4.14	3.39
Vandetanib	4.31	4.93	5.04	3.32	4.31	4.38

Pharmacokinetics and Solubility

The pharmacokinetic profile of the synthesized compounds was evaluated using a Support Vector Machine model, which assessed their potential as P-glycoprotein substrates based on the characteristics of 332 molecules. Additionally, the model estimated the likelihood of high or low gastrointestinal absorption from a dataset of 597 compounds. The predicted log Kp values, derived from a

Quantitative Structure-Activity Relationship (QSAR) model for skin permeation, indicated permeability coefficients ranging from -4.39 to -5.13. A negative log Kp value suggests reduced skin permeability, which is crucial for evaluating the transdermal transport potential of these compounds [25].

SwissADME provided three distinct linear models to estimate water solubility, including quantitative assessments by log S and qualitative assessments. As shown in Table 4, the log S values, calculated using the ESOL algorithm, indicate that all synthesized compounds (4a–j) possess log S values between -5.14 and -6.76. According to the SwissADME classification, these values categorize the compounds as weakly soluble, with solubility expected to be limited for bioavailability from the gastrointestinal tract. The drug-likeness of the synthesized compounds was further assessed using the Lipinski and Veber rules. According to Lipinski's rule of five, good absorption or permeation is typically expected when molecular weight (MW) is <500 Da, hydrogen bond donors (HBDs) are < 5, log P is < 5, and hydrogen bond acceptors (HBAs) are < 10. The Veber et al. criteria also stipulate that the number of rotatable bonds (NBR) should be < 10 and polar surface area (PSA) < 140 Å^2. Most compounds adhered well to these rules, although compounds 4a, 4b, 4i, and 4j violated the log P criterion with values exceeding 5. Despite this, compounds 4b, 4c, 4d, 4f, 4g, and 4h exhibited favourable profiles for absorption and skin permeation.

Table 4. Pharmacokinetic Parameters for the Ligands (4a-j) and their Solubility in Water

Ligands	**P-Glycoprotein substrate**	**GI-tract absorption**	**Log Kp (skin permeation) cm/s**	**Log S (ESOL) Solubility in Water**
4a	No	High	-4.39	-6.76 poorly soluble
4b	No	high	-4.63	-6.16 poorly soluble
4c	No	high	-4.75	-5.58 Moderately soluble
4d	No	high	-4.89	-5.73 Moderately soluble
4e	No	High	-4.86	-5.57 Moderately soluble
4f	No	high	-4.86	-5.57 Moderately soluble
4g	No	high	-5.13	-5.14 Moderately soluble
4h	No	high	-4.75	-5.58 Moderately soluble
4i	No	high	-4.88	-5.83 Moderately soluble
4j	No	high	-4.88	-5.83 Moderately soluble
KU 058948	Yes	high	-7.29	-3.54 soluble
Gestrinone	No	high	-6.65	-3.05 soluble
Vandetanib	Yes	high	-5.70	-5.92 moderately soluble

The bioavailability score for all synthesized compounds (4a-j) and standard drugs such as KU0058948, Gestrinone, and Vanetanib was calculated to be 0.55, indicating moderate bioavailability potential. The medicinal chemistry of these prospective drug molecules was assessed using the PAINS (pan assay interference compounds) algorithm, which flagged one alert for all synthesized compounds. Additionally, the Brenk filter revealed one alert for each ligand, indicating potential issues with promiscuity or undesirable interactions. Synthetic accessibility of the ligands was also in the acceptable range and is presented in Tables 5 and 6. Energy minimization of the ligand structures was performed using Argus Lab software, optimizing their conformations to achieve stable configurations conducive to effective docking interactions [26]. The minimization process, which involved conserving deformation energies over successive cycles, was tailored to ensure the ligands were well-prepared for docking studies.

Table 5. Drug Likeliness of the Ligands 4a-j

Ligands	Lipinski	Ghose	Veber	Egan	Muegge	Lead-like	Bioavailability Score
4a	yes	1 violation	yes	1 violation	1 violation	2 violations	0.55
4b	yes	1 violation	yes	yes	1 violation	2 violations	0.55
4c	yes	Yes	yes	yes	I violation	2 violations	0.55
4d	yes	Yes	yes	yes	1 violation	2 violations	0.55
4e	yes	Yes	yes	yes	1 violation	2 violations	0.55
4f	yes	Yes	yes	yes	1 violation	2 violations	0.55
4g	yes	Yes	yes	yes	yes	2 violations	0.55
4h	yes	Yes	yes	yes	1 violation	2 violations	0.55
4i	yes	1 violation	Yes	1 violation	1 violation	2 violations	0.55
4j	yes	1 violation	yes	1 violation	1 violation	2 violations	0.55
KU 0058948	yes	Yes	yes	yes	yes	1 violation	0.55
Gestrinone	yes	Yes	yes	yes	yes	yes	0.55
Vandetanib	yes	Yes	yes	yes	yes	yes	0.55

Docking Results

The molecular docking of synthesized compounds (substituted benzylidene)-2-(arylamino)-1,3-thiazol-4(5H)-ones (4a-j) was conducted using the FlexX docking software, which incorporates a flexible optimization approach to accommodate torsional freedom in ligand placement. The scoring function utilized is based on the Bohm function, enabling effective evaluation of ligand-receptor interactions.

Table 6. Medicinal Chemistry of Ligands 4a-j

Ligands	PAINS	Brenk	Synthetic accessibility
4a	1 alert	2 alert(s)	2.08
4b	1 alert	1 alert	1.90
4c	1 alert	1 alert	1.96
4d	1 alert	1 alert	1.91
4e	1 alert	1 alert	1.83

4f	1 alert	1 alert	1.94
4g	1 alert	1 alert	2.02
4h	1 alert	1 alert	2.13
4i	1 alert	1 alert	2.08
4j	1 alert	1 alert	2.25
KU 058948	0 alert	0 alert	2.37
Gestrinone	0 alert	1 alert	5.46
Vanetanib	0 alert	0 alert	2.50

The docking scores for the synthesized compounds against the estrogen receptor (PDB ID: 3L03) were compared to the standard drug Gestrinone [27] (Table 7). Notably, compound 4g, 5-(3-chloro-2-fluorobenzylidene)-2-[(4-fluorophenyl)amino]-1,3-thiazol-4(5H)-one, achieved the highest docking score of -18.7649, to the target EGFR alpha which is far more potent than the reference standard drug Gestrinone, which scored -16.1457. The binding interactions of compound 4g with the receptor's binding pocket are illustrated in Figure 1. This compound did not violate the Ghose filter, indicating that its molecular weight exceeds the minimum threshold of 160 g/mol, while it also met the Muegge filter requirement of being over 200 g/mol. However, it did exceed the lead-like filter, which stipulates a maximum weight of 250 g/mol. Compound 4c, with a docking score of -17.2440, also exceeded the weight limit of the lead-like filter but demonstrated a strong binding affinity. Other compounds, including 4e, 4d, 4f, and 4j, displayed docking scores closely aligned with that of Gestrinone, suggesting that six of the ten synthesized compounds exhibit promising binding interactions with the estrogen receptor.

Table 7. Docking Results for the Compounds 4a-j with Breast Cancer Drug Target EGFR Alpha

Ligands	**TotalScore**	**Match**	**Lipo**	**Ambig**	**Clash**	**Rot**	**# Match**
4a	-12.4994	-10.6374	-15.1980	-3.8033	11.7893	0.0000	12
4b	-12.0926	-7.3920	-15.4966	-4.4024	9.7984	0.0000	13
4c	-17.2440	-13.6023	-14.2227	-2.8771	8.0591	0.0000	16
4d	-15.9385	-11.3085	-15.7091	-5.5240	11.2031	0.0000	16
4e	-16.6667	-14.5673	-11.9086	-4.0600	8.4702	0.0000	17
4f	-15.9828	-12.6705	-13.3551	-4.4943	9.1371	0.0000	16
4g	-18.7694	14.3465	-12.6556	-4.3879	7.2206	0.0000	18
4h	-12.1726	-12.4272	-12.3497	-3.4857	10.6900	0.0000	16
4i	-10.8781	-12.9084	-12.3087	-4.5555	12.0945	1.4000	17
4j	-15.4179	-14.0117	-12.6675	-8.3475	8.3475	1.4000	17
Gestrinone	-16.1457	-12.8548	-13.8934	-3.5408	5.9432	2.8000	3

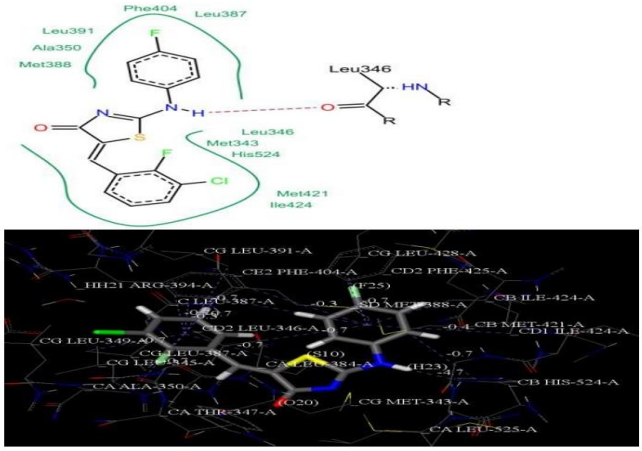


Figure 1. 5-(3-Chloro-2-Fluorobenzylidene)-2-[(4-Fluorophenyl)Amino]-1,3-Thiazol-4(5*H*)-Onedocked to EGFR Alpha

Vascular Endothelial Growth Factor Receptor-2 (VEGFR-2)

For the Vascular Endothelial Growth Factor Receptor-2 (PDB ID: 3CPC), compound 4g again emerged as the most effective ligand, achieving a docking score of -22.3284, which is comparable to the standard drug Vandetanib, with a score of -23.2287 (Figure 2) [28]. Compounds 4h and 4j also demonstrated substantial binding scores of -21.4453. The overall docking scores for compounds 4a-j ranged from -18 to -22, confirming their potential potency against VEGFR-2 (Table 8). These results underscore the strong binding interactions of the synthesized compounds with both EGFR alpha and VEGFR-2 targets, suggesting that further structural modifications could enhance their medicinal properties and inhibitory activities.

Table 8. Docking Results for the Compounds 4a-j with VGEFR-2

Ligands	TotalScore	Match	Lipo	Ambig	Clash	Rot	# Match
4a	-18. 4372	-13.6397.	-12.3701	-5.4317	7.6043	0.0000	17
4b	-19.5332	-12.9328	-14.1515	-5.2597	7.4108	0.0000	19
4c	-20.6161	-13.0286	-14.9780	-6.4732	6.5446	0.0000	16
4d	-20.0986	-15.4292	-13.9776	-3.4236	7.3318	0.0000	18
4e	-20.5107	-16.2620	-12.4183	-3.0674	5.8371	0.0000	10
4f	-19.9465	-12.4470	-13.3599	-5.8277	6.2881	0.0000	14
4g	-22.3284	-17.2233	-13.8574	-5.6692	9.0215	0.0000	18
4h	-21.4453	-13.6725	-16.3993	-5.3139	7.1405	1.4000	18
4i	-18.58521	-12.4945	-11.4226	-5.8096	5.7446	0.0000	16
4j	-21.4453	-13.6726	-16.3993	-5.3139	7.1406	1.4000	18
Vandetanib	-23.2287	-23.1163	-14.6101	-6.3949	9.8926	5.6000	18

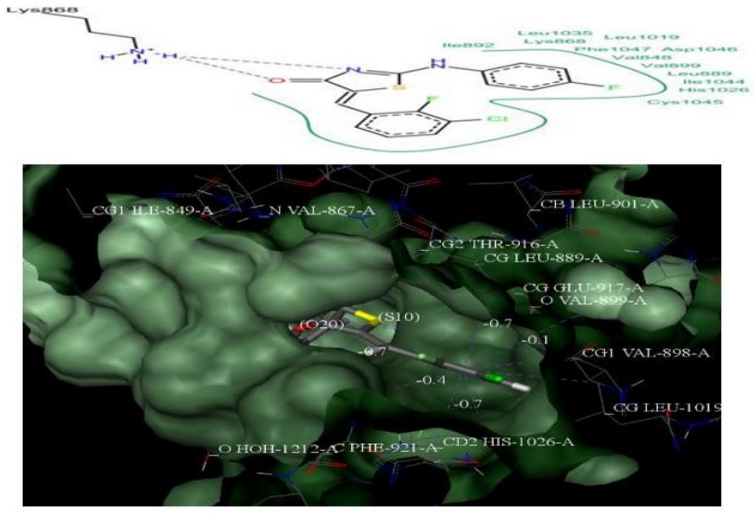


Figure 2. 5-(3-Chloro-2-Fluorobenzylidene)-2-[(4-Fluorophenyl)Amino]-1,3-Thiazol-4(5*H*)-One Docked to VEGFR-2

Poly [ADP-Ribose] Polymerase-1 (PARP-1)

The docking study against Poly[ADP-ribose]polymerase-1 (PDB ID: 3L3M) revealed that compounds 4d and 4g exhibited superior binding interactions, scoring -26.8680 and -25.6428, respectively [29,30]. In comparison, the standard drug KU0058948 had a docking score of -43.5813, indicating that while the synthesized compounds show promise, the reference compound remains significantly more potent (Table 9).

Structure-Activity Relationship Insights

Through the process of molecular docking, it was discovered that compound 4g 5-(3-chloro-2-fluorobenzylidene)-2-[(4-fluorophenyl)amino]-1,3-thiazol-4(5H)-one had favourable binding interactions, exhibiting the highest docking score in contrast to the other compounds. The favourable binding characteristics of compound 4g are notably attributed to the presence of a fluoro group at the para position of the arylamino ring, enhancing its interactions within the binding site. Similarly, compound 4d, which also includes a fluoro group and a chloro group, demonstrated effective binding, reinforcing the hypothesis that specific halogen substitutions can positively influence binding affinity. Overall, the synthesized ligands showed enhanced activity, particularly against EGFR alpha and VEGFR-2, suggesting a promising lead for further drug development.

Table 9. Docking Results for the Compounds 4a-j with Ovarian Cancer Drug Target Poly ADP Polymerase

Ligands	Total Score	Match	Lipo	Ambig	Clash	Rot	# Match
4a	-20.0124	-9.8318	-16.9478	-7.3661	8.7332	0.0000	6
4b	-19.9033	-10.6435	-11.5568	-7.0935	3.9905	0.0000	9
4c	-21.8077	-13.7135	-12.1314	-5.3559	3.9930	0.0000	13
4d	-26.8680	-14.4146	-14.7733	-8.9290	6.0688	0.0000	12
4e	-24.6097	-13.9490	-14.7227	-6.0457	4.7078	0.0000	10
4f	-23.9289	-10.8722	-14.1269	-8.0934	3.7637	0.0000	10
4g	-25.6428	-15.1746	-12.9414	-7.8534	4.9264	0.0000	11
4h	-23.3942	-12.4888	-11.6280	-6.5988	1.9214	0.0000	9
4i	-22.9418	-11.7370	-15.7876	-7.3787	5.1615	1.4000	8

4j	-22.4354	-12.9263	-15.0092	-8.7243	7.4243	1.4000	12
KU0058948	-43.5813	-33.2937	-13.1655	-8.5768	3.2547	2.8000	21

Conclusions

This study successfully synthesized a series of (substituted benzylidene)-2-(arylamino)-1,3-thiazol-4(5H)-ones with notable yields ranging from 73% to 88%. Characterization through IR, 1H NMR, and mass spectral data confirmed the formation of these compounds. Molecular docking studies indicated favourable interaction energies with key targets, including Poly[ADP-ribose]polymerase-1 (cancer tumour 3l3m), Vascular Endothelial Growth Factor Receptor-2 (3CPC), and the human estrogen receptor (breast cancer 3LO3), highlighting compound 4g as a promising drug-like candidate due to its favourable docking score and multi-target inhibitory profile. The presence of a fluoro group at the para position of the amino phenyl ring was found to play a crucial role in enhancing interactions with key residues in the docking sites. Additionally, most synthesized compounds exhibited strong docking scores against VEGFR-2 and EGFR alpha, indicating their potential as effective inhibitors. SwissADME analysis further supported the drug-like properties of these compounds, demonstrating good lipophilicity, low fraction unbound values, and appropriate distribution profiles, which are indicative of favourable bioavailability. Overall, this research not only identifies compound 4g as a potential lead molecule but also provides a valuable template for designing new compounds with improved binding affinities and enhanced ligand-receptor interactions. Future studies could build on these findings to explore further optimizations and validate the therapeutic potential of these novel thiazole derivatives.

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Conflicts of Interest

The authors declare that they have no conflict of interest in this study.

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