Asian Journal of Chemistry; Vol. 33, No. 3 (2021), 537-544

ASIAN JOURNAL OF CHEMISTRY

https://doi.org/10.14233/ajchem.2021.23062

Design, Molecular Docking and Biological Evaluation of Fused Thienopyrimidines and Quinazoline

S. Kaliraj[1], R. Jeyalakshmi[1,*], M.K. Kathiravan[2], T. Madhavan[3] and Arikketh Devi[3]

[1]Department of Chemistry, SRM Institute of Science & Technology, Kattankulathur-603203, India
[2]Department of Pharmaceutical Chemistry, SRM College of Pharmacy, SRM Institute of Science & Technology, Kattankulathur-603203, India.
[3]Department of Genetic Engineering, School of Bio-Engineering, SRM Institute of Science & Technology, Kattankulathur-603203, India

*Corresponding author: E-mail: jeyalakr@srmist.edu.in; rajyashree64@gmail.com

Received: 10 November 2020; *Accepted*: 28 December 2020; *Published online*: 16 February 2021; AJC-20246

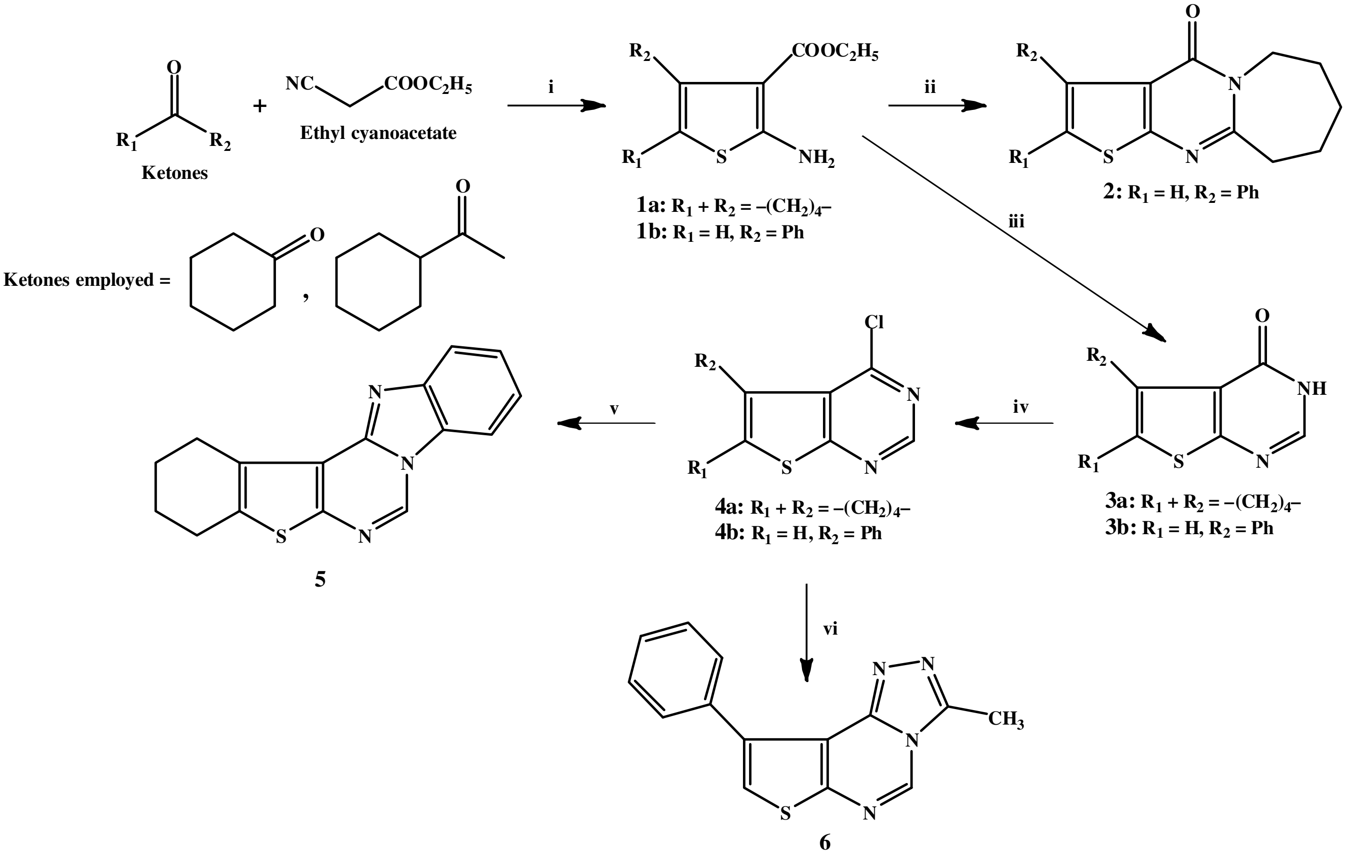
The anticancer activity of the condensed pyrimidine and quinazoline moieties are pronounced with a different pathway. Thienopyrimidine is considered as ring equivalent bioisosteres of quinazolines and present in other heterocyclic compounds including thienopyrimidine. The present investigation focused on the synthesis of thienopyrimidine and quinazoline derivatives for their anticancer activity against the human oral squamous carcinoma-3 (HSC-3) cell line. The synthesized compound confirmed for their structural characteristics from spectral analysis and tested for anti-proliferative activity from MTT assay. The electron-withdrawing group in 4-chloro thienopyrimidines and amino ester derivate/facilitate the inhibition of cancer cells. Further probing by docking studies revealed that the compounds exhibit possible interactions with VEGF, FGFR and c-Met proteins, which are known to have a role in the pathogenesis of oral squamous cell carcinoma. Among the derivatives a moderate activity demonstrated by substituted 4-chloro-5,6,7,8-tetrahydrobenzo[4,5]thieno[2,3-*d*]-pyrimidine (**4a**) and ethyl 2-amino-4,5,6,7-tetrahydrobenzo[*b*]thiophene-3-carboxylate (**1a**) derivatives. Lack of chirality and the presence of bulky substituents in few of the compounds were found to be the cause for lower potency.

Keywords: Thienopyrimidines, Quinazoline, lactams, Anti-proliferative activity, Molecular docking.

INTRODUCTION

Thienopyrimidines are made up of a thiophene ring fused with the pyrimidine moiety, like the imidazole moiety in purines [1]. Fused bioactive thienopyrimidine heterocycle is a versatile lead molecule finding many applications in the pharmaceutical field showing a wide spectrum of functions such as antibacterial [2,3], antimalarial [4], antiviral [5], analgesic [6], antioxidant [7], anti-inflammatory [8], antihypertensive [9], anti-histaminic [10], antiproliferative [11], anticovalent [12], antihyperlipidemic [13,14] and anticancer activity [15-18]. Pyrimidines, mainly thieno[2,3-*d*] pyrimidine derivative, were investigated for their anticancer activity through the inhibition of different protein kinase enzymes such as *c*-Met, VEGFR, dual EGFR/ErbB-2, EGFR, FGFR1, CDK-4 and Aurora [19-23]. The diverse biological activity of pyrimidines arises primarily because it is the fundamental building block of both DNA and RNA. Thienopyrimidine shows a broad spectrum of bioactivity similar to biogenic pyrimidines. Thieno[2,3-*d*]pyrimidine scaffold is part of 4-anilinoquinazoline scaffold, present in gefitinib, a widely marketed anticancer drug. Similarly, triazole fused thienopyrimidines derivatives have been gaining importance for their anticancer activities [24].

Oral cancer is a bad biomarker and medication has been designed from natural products and synthetic substances. Naturally, the seed, leaf and other parts of plants and trees, as well as synthesized chemical compounds, were used in oral cancer medication. However, studies on the activity of thieno pyrimidines and quinazoline against the HSC3 are comparatively less. Thieno-pyrimidine fused with triazines, triazoles and pyrazoles have gained prominence for their potent biological activity. To cite a few, novel thiophene and thienopyrimidine derivatives were screened for five cell lines such as epidermoid carcinoma (larynx) Hep-2, hepatocellular carcinoma (liver) HepG-2, mammary gland (breast) MCF-7, epitheloid (cervix) carcinoma Hela and human prostate cancer PC-3. The results



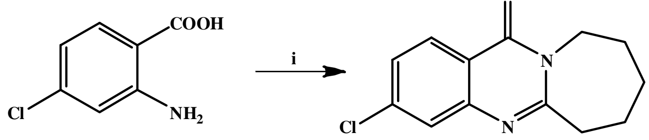
Scheme-I: Synthesis of benzimidazole, caprolactam, and triazole-fused thienopyrimidine. Reagents and conditions: (i) S, morpholine, ethanol room temperature stirring; (ii) caprolactam, $POCl_3$, toluene, reflux; (iii) formamide, 210 Temp, reflux; (iv) $POCl_3$, toluene, reflux; (v) *o*-phenylenediamine, microwave 350 W, 20 min, (iv) acetic hydrazide, ethanol, reflux

showed the highest anticancer activity against all the cell lines when compared to doxorubicin [25]. A series of compounds showed anti-lung cancer activity against A549 and colon HC-T-116 cells when a dithiocarbamate moiety was incorporated at the C2 position in thieno[2,3-*d*]pyrimidine scaffold. The anticancer activity of triazole fused heterocycles is well documented [24].

Based on these facts, recently the synthesis of fused thienopyrimidines, quinazolines compounds using butyrolactam, valerolactam, caprolactam, triazole and benzimidazole is reported [26,27]. Derivatives of these base compounds were prepared and screened for their potential anticancer activity. Lactam fused pyrimidines showed activity against human embryonic kidney cell line (HEK 293), derivatives of benzimidazole fused thienopyrimidines inhibited the breast cancer cell line (MCF-7) growth. More than 20 compounds were synthesized and subjected to test against human epidermoid carcinoma cell line (A431) and rat cardiomyocyte cells (H9c2). The results showed the promising anticancer activity against different cell lines. The docking studies on-base compounds and off molecules indicated the signaling pathways by inhibiting proteins with the crystal structure of vascular EGFR 2 kinase with PDB code 3VHK and EGFR kinase with PDB code 4RJ8. In continuation of our ongoing efforts in investing condensed pyrimidine potential activity, we herein report the synthesis, activity against the oral cancer cell line of the inhibition activity from molecular docking studies.

EXPERIMENTAL

Materials used in present study were of AR grade chemicals (ethyl cyanoacetate, diethylamine, toluene, acetic hydrazide, phosphorus oxychloride and *o*-phenylenediamine) and procured from SRL, India. Compounds **1-7** were synthesized as per the reported method [26,27]. The synthesis of benzimidazole, caprolactam and triazole-fused thienopyrimidine derivatives are shown in **Scheme-I**. **Scheme-II** corresponds to caprolactam fused quinazoline obtained from caprolactam under reflux with $POCl_3$ and toluene.



Scheme-II: Synthesis of caprolactam-fused quinazoline. Reagents and conditions: (i) caprolactam, $POCl_3$, toluene, reflux

The physico-chemical properties and spectral studies were carried out to assess the structure and functional groups. All ^1H & ^{13}C NMR spectra were obtained using Bruker spectrometer operating at 500 MHz. Chemical shifts were expressed relative to tetramethylsilane (TMS) and reported as δ (ppm).

The IR, ESI-MS and ^{1}H NMR spectra were analyzed and reported already in our previous work [26,27]. The ^{13}C NMR data are used to confirm the structure of compounds which are provided in this study.

Compound 1a: ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.97 (s, 1H), 4.27 (q, *J* = 7.0 Hz, 1H), 2.72 (s, 1H), 2.52 (s, 1H), 1.89-1.69 (m, 4H), 1.35 (t, *J* = 7.0 Hz, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 166.18, 161.72, 132.47, 117.65, 105.75, 59.40, 26.96, 24.55, 23.27, 22.84, 14.49.

Compound 2: ^{1}H NMR (500 MHz; $CDCl_3$) δ 5 7.54 (d, *J* = 7.3 Hz, 2H), 7.41 (t, *J* = 7.5 Hz, 2H), 7.37 (t, *J* = 7.4 Hz, 1H), 7.26 (s, 1H), 4.37 (t, *J* = 5.5 Hz, 2H), 3.07 (t, *J* = 6.5 Hz, 2H), 1.75-1.87(m, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 164.56, 161.70, 157.83, 139.02, 135.72, 129.73, 127.99, 127.83, 120.92, 118.15, 42.11, 36.81, 29.11, 27.35, 24.95.

Compound 4a: ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.72 (s, 1H), 3.06 (d, *J* = 7.6 Hz, 1H), 2.85 (d, *J* = 5.3 Hz, 1H), 1.90 (d, *J* = 6.0 Hz, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 168.88, 153.18, 151.55, 139.65, 128.85, 127.18, 26.18, 22.33.

Compound 5: ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.08 (s, 1H), 7.99 (d, *J* = 8.1 Hz, 2H), 7.55 (d, *J* = 7.0 Hz, 1H), 7.42 (d, *J* = 4.1, 1H), 2.69 (t, *J* = 8.7 Hz, 2H), 2.39 (t, *J* = 7.3 Hz, 2H), 2.25-2.18 (m, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 152.33, 144.78, 144.00, 136.57, 134.54, 130.16, 126.73, 126.09, 122.18, 119.84, 110.54, 25.68, 25.26, 22.81, 22.08.

Compound 6: ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.27 (s, 1H), 8.39 (s, 1H), 7.56 (d, *J* = 10.4 Hz, 2H), 7.44 (t, *J* = 7.4 Hz, 2H), 7.25 (t, *J* = 2.1 Hz, 1H), 2.48 (s, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 164.19, 156.25, 154.53, 149.42, 138.00, 136.42, 134.60, 129.53, 128.64, 125.82, 117.99, 14.64.

Compound 7: ^{1}H NMR (500 MHz; $CDCl_3$) δ 5 8.18 (d, *J* = 2.5 Hz, 1H), 7.60 (s, 1H), 7.18 (d, *J* = 7.5 Hz, 1H), 4.36 (t, *J* = 6.5 Hz, 2H), 3.06 (t, *J* = 6.5 Hz, 2H),1.81-1.87 (m, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 167.38, 165.87, 153.04, 144.38, 133.76, 132.03, 130.55, 123.47, 47.61, 41.65, 33.71, 32.32, 29.77.

Thus from the spectral data, the chemical structure of the synthesized substances are confirmed.

***in vitro* Cytotoxicity (MTT assay):** The cytotoxicity effects of the compounds were performed on the HSC-3 cell line derived from the human oral cancer tissue obtained from the Japanese Collection of Research Bioresources Cell Bank (JCRB). The cells were cultured in Dulbecco's modified eagle medium (DMEM) containing 10% fetal bovine serum (FBS) and 100 U/mL penicillin and 100 g/mL streptomycin. The cells were maintained at 37 ºC, 5% CO_2, 95% air and 100% relative humidity. The antiproliferative effect of the target compounds was measured by plating HSC-3 cells in a 96-well plate (7000 cells/well) and incubated at 37 ºC for 24h. The cells were treated with different concentrations of the compounds (250-1000 µM) and incubated for 48 h. After incubation, MTT (5 mg/mL in phosphate-buffered saline (PBS) was added to each well and incubated for 4 h. The culture media was removed and the formazan crystals were solubilized using 10% SDS. The absorbance at wavelength 595 nm was measured using a microplate reader to calculate the percentage of surviving cells and IC_{50} was determined using Graph Pad Prism software.

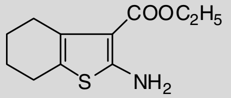
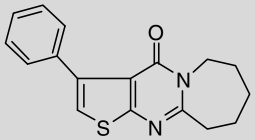
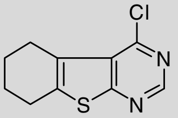
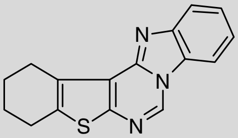
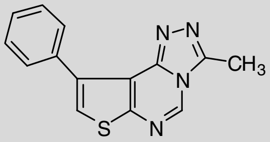
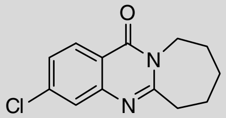
Molecular docking: The binding interaction of the compounds **1a**, **2**, **4a**, **5**, **6** and **7** with VEGF, FGFR and c-Met proteins using Autodock 4.2 software [28] were studied. Molecular docking was performed by considering the protein to be rigid and ligand as flexible where all rotatable bonds of the ligand were considered. The grid box was generated and centered on the binding site residues with 60 × 60 × 60 grid points in XYZ direction with 0.375 Å spacing. The Lamarckian genetic algorithm, which combines both genetic and local search algorithms, was employed for docking with default parameters. Also, the Glide module incorporated into Maestro version 10.2 [29] was used to perform docking because Glide reliably finds the correct binding modes and it outperforms other docking programs in achieving lower root mean square (RMS) deviations from native co-crystallized structures [30]. Glide docking calculations were run in the standard precision mode with default values. As being a general procedure, the docking setup was validated by ligand having protein binding with cells. A recent literature report of similar works on quinazoline derivatives with thienopyrimidines used EGFR and VEGFR receptors and the co-crystallized ligand Eriotinib and Tivozanib [31]. The base structure selected from the protein data bank of erlotinib co-crystallized with identified proteins of the present study utilized to determine ligand-receptor interactions followed by compound interaction.

RESULTS AND DISCUSSION

Aminothiophene carboxylate **1a** and **1b** (thiophene *o*-amino esters) precursors are synthesized by Gewald's reaction from the corresponding ketones, ethyl cyanoacetate and sulfur. In next step, thiophene *o*-amino esters were cyclized with caprolactam salts by treating with phosphorous oxychloride under refluxed to obtain the compound lactam-fused thienopyrimidine (**2**). Another synthetic pathway to benzimidazole fused thienopyrimidine (**5**) was accomplished by Niementowski condensations from thiophene *o*-amino esters with formamide followed by reacting with phosphorous oxychloride and finally microwave irradiation with *o*-phenylenediamine. Further, triazole fused thienopyrimidine (**6**) was synthesized from compound **4b** by the addition of acetic acid hydrazide in methanol under reflux. The next set of compounds synthesized, by following **Scheme-II** to lactam-fused quinazoline target compounds by ring annulation of pyrimidine. Lactam-fused quinazoline (**7**) prepared from 2-amino-4-chlorobenzoic acid by the addition of phosphorous oxychloride in toluene under reflux.

The physico-chemical properties of all the synthesized compounds are presented in Table-1. The ^{1}H NMR spectra provided data on the chemical shift of compounds **2** and **6**, showing the aromatic –CH– signal at δ 7.39-7.51 ppm as a triplet. Rather, eight positions of $–CH_3$ present in compound **6** showed a strong field at δ 2.48 ppm and all other three protons as a singlet. The sixth position –CH– in compounds **5** and **6** exhibited a weaker field at δ 9.08 and 9.27 ppm as a singlet and similarly, in the ^{13}C NMR spectrum, a weaker signal at δ 144 ppm can be observed. The observed six-proton multiplet $3\text{-}CH_2\text{–}(CH_2)_2$ in compounds **7** and **2** are seen in a stronger field of δ 1.81-1.87 ppm and δ 1.75-1.87 ppm, respectively.

TABLE-1
PHYSIO-CHEMICAL PROPERTIES AND IC_{50} VALUES OF THE COMPOUNDS

Compounds No.	Structure	m.f.	m.w.	m.p. (°C)	Yield (%)	HSC-3 cells IC_{50} (µM)
1a		$C_{11}H_{15}NO_2S$	227.31	110-112	90	183.2
2		$C_{17}H_{16}N_2OS$	296.39	163-165	60	491.5
4a		$C_{10}H_9N_2SCl$	224.02	114-116	75	101.6
5		$C_{16}H_{13}N_3S$	279.08	204-206	64	895.9
6		$C_{14}H_{10}N_4S$	266.90	190-195	50	424.1
7		$C_{13}H_{13}N_2OCl$	249.71	158-160	50	337.9
8	Doxorubicin	–	–	–	–	20

The structures in present study were further confirmed by ^{13}C NMR data. The chemical shift values of aromatic –C=C–at δ 127-130 ppm and C=O at a downfield chemical shift of δ 165 and 164 ppm can be seen. Thus both ^1H & ^{13}C NMR spectra confirmed all the relevant positions and number of protons and carbon of which completely correspond to the structure of the synthesized compounds.

Biological study: The anti-proliferative effect of the target compounds was examined by performing a 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay on the HSC-3 cell line. The oral cells were treated with increasing concentrations (250-1000 µM) of six derivatives after which the percentage of surviving cells calculated (Fig. 1). The results of the cytotoxic activity using HSC-3 cell lines showed that out of the 6 target compounds tested two compounds showed moderate activity with IC_{50} values of 183.2 and 101.6 for **1a** and **4a**, respectively, while the positive control doxorubicin showed IC_{50} 20 µM.

The results showed that compounds **1a** and **4a** exhibit better activity compared to the other modified compounds

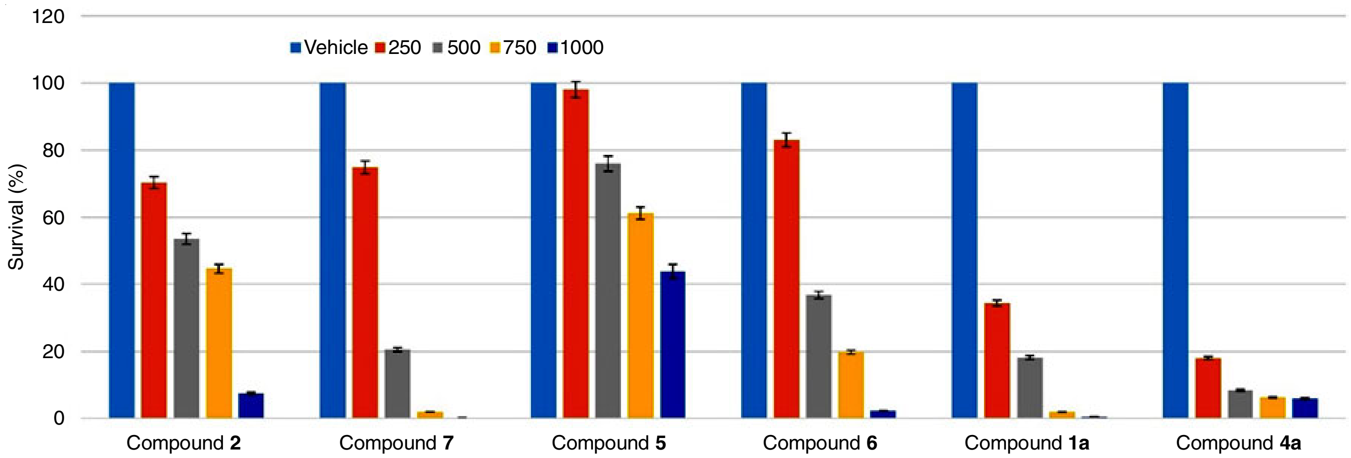


Fig. 1. Anti-proliferative effect of the compounds based on % cell survival in HSC-3 cells

while compound **5** possesses the least anti-proliferative effect. Compound **5** is benzimidazole-fused thienopyrimidine and the results of the anti-proliferative effect on the cell line HSC-3 show high IC_{50} when compared with compound **1a**. It implies that bulky group substitution in **5** hampers the anti-proliferative activity. It is worth to note that compound **1a** has a primary amine group, which is a hydrogen bond donor and facilitates binding to the amino acid residues, thereby inhibiting the growth of cells. However, the interactions are purely based on the position of substituents and orientation of the aryl groups.

It is suggested that the positioning to form intramolecular or intermolecular hydrogen bonding is required irrespective of weak or strong hydrogen acceptors. The most crucial part is pyrimidine nitrogen, which exerts ligand bonding mainly influenced by the electronic effects of substituents [32]. In the case of compound **4a**, chloro thienopyrimidine is found to have a lower IC_{50} value compared with other synthesized compounds. This is because the bulky group in the 7th position adjacent to the pyrimidine nitrogen changes the compound to becoming nonplanar without a chiral center [19,23]. This non-optimal positioning does not result in any intramolecular hydrogen bonding. Considering this, we infer the lower activity could be due to the improper positioning of the substituent, which could be further probed in our future study.

Docking study: The investigation was extended to understand the potency of the compound related to the structure through docking studies. Tertiary structures of VEGF2 (crystal structure of VEGFR2 kinase domain in complex with a pyrrazolone inhibitor PDB Id: 3U6J), FGFR1 (crystal structure of activated receptor tyrosine kinase in complex with substrates PDB Id: 3GQI) and c-Met (structure of the kinase domain of c-Met bound to XL880 (GSK1363089) PDB Id: 3LQ8) were downloaded from PDB database. The protein structures were prepared by removing all heteroatoms including co-crystallized ligands, ions and water molecules and subsequently adding hydrogen atoms and charges such as Kollman and Gasteiger charges. The structure of the ligand molecules was drawn and optimized using the SYBYLX-2.0 software.

Energy minimization was performed using the Tripos force field and Gasteiger Huckel charge methods. Using Autodock, the ADT atom type was assigned for both the protein and the ligand molecules and used for subsequent analysis. In Autodock, two approaches, namely blind docking and active site based docking were adopted. In blind docking, the ligands were made to bind randomly to the protein whereas, inactive site-based method, the ligands were made to bind inside the active site of VEGF2, FGFR1 and c-Met proteins. These approaches were performed to check the possibility of ligand binding on the whole protein and specifically inside the protein binding site. The binding site was selected based on the co-crystal ligand molecule in the target protein structure [33-35].

During docking, 10 different conformations were generated for each compound. The conformation having the lowest binding energy and better interaction with active site residues was considered as the active conformation for each compound. H-bond interaction, binding energy and orientation of the docked compound within the active site were also analyzed. It is observed that all these compounds showing better binding energy in the range of -5 to -8 kcal/mol for VEGF2, -4 to -6 kcal/mol for FGFR1 and -4 to -8 kcal/mol for c-Met. Erlotinib has shown the highest docking score greater than -12 kcal/mol. The detailed information about the docking score obtained from Autodock and Glide are given in Table-2. Fig. 2a-c show the binding of the compounds inside the binding cavity of VEGF2, FGFR1 and c-Met, respectively. It is observed that these compounds formed H-bond interaction with the active residues. The interaction of a ligand with active site residues along with the H-bond formation and H-bond distance are shown in Figs. 3-5 for VEGF2, FGFR1 and c-Met, respectively. While comparing with enzyme inhibition of erlotinib, present compounds exhibited lower potency though they have similar structures.

TABLE-2
DOCKING SCORE AND H-BOND FORMING RESIDUE

Compd. No.	Autodock score			Glide SP score		
	VEGF2	FGFR1	*c*-Met	VEGF2	FGFR1	*c*-Met
1a	-5.08	-4.22	-4.68	-7.45	-4.98	-7.13
2	-7.67	-5.80	-5.29	-8.30	-5.52	-8.49
4a	-6.28	-4.34	-4.41	-6.32	-5.38	-8.08
5	-8.05	-5.33	-8.12	-9.17	-6.13	-9.65
6	-7.17	-5.55	-5.40	-7.76	-5.43	-8.88
7	-6.64	-4.67	-7.58	-7.82	-5.60	-9.17

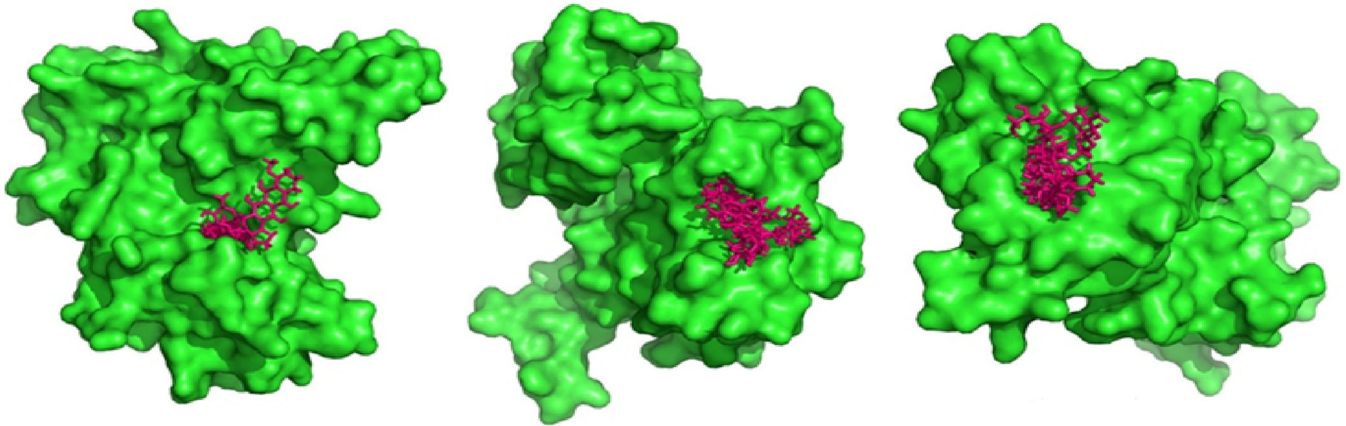

(a) VEGF2 (b) FGFR1 (c) *c*-Met

Fig. 2. The docked conformation of compounds **1a**, **2**, **4a**, **5**, **6** and **7** inside the binding site. A: VEGF2, B: FGFR1, C: *c*-Met

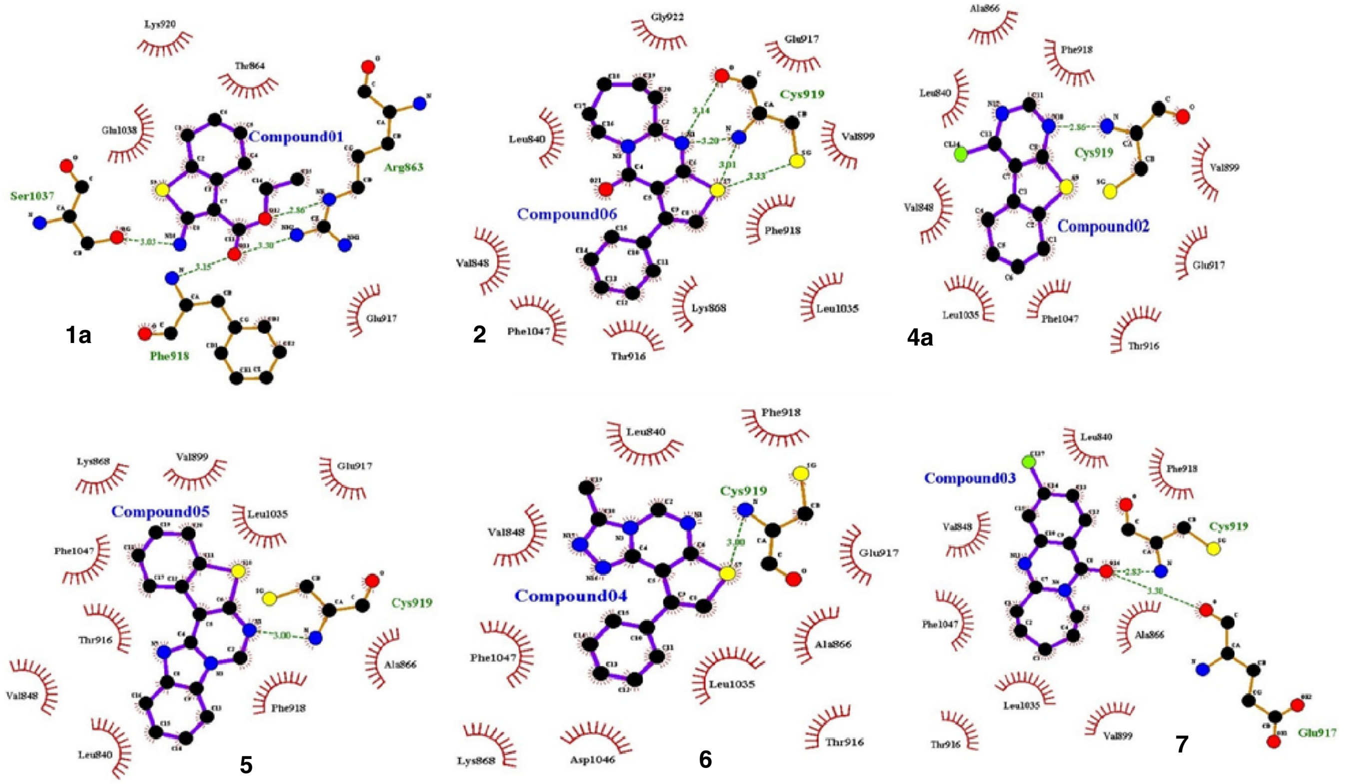


Fig. 3. Interaction of compounds **1a**, **2**, **4a**, **5**, **6** and **7** with VEGF2

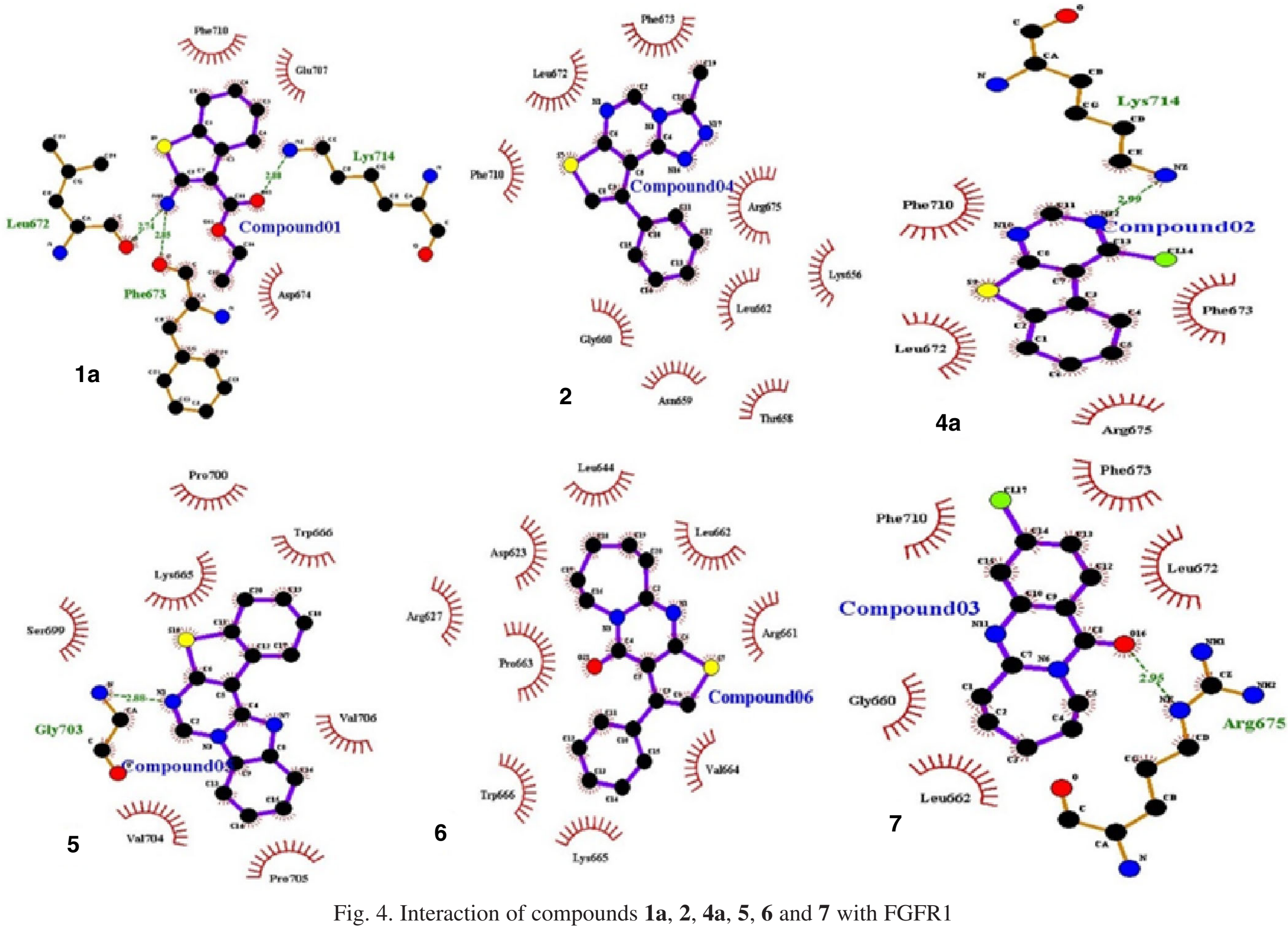


Fig. 4. Interaction of compounds **1a**, **2**, **4a**, **5**, **6** and **7** with FGFR1

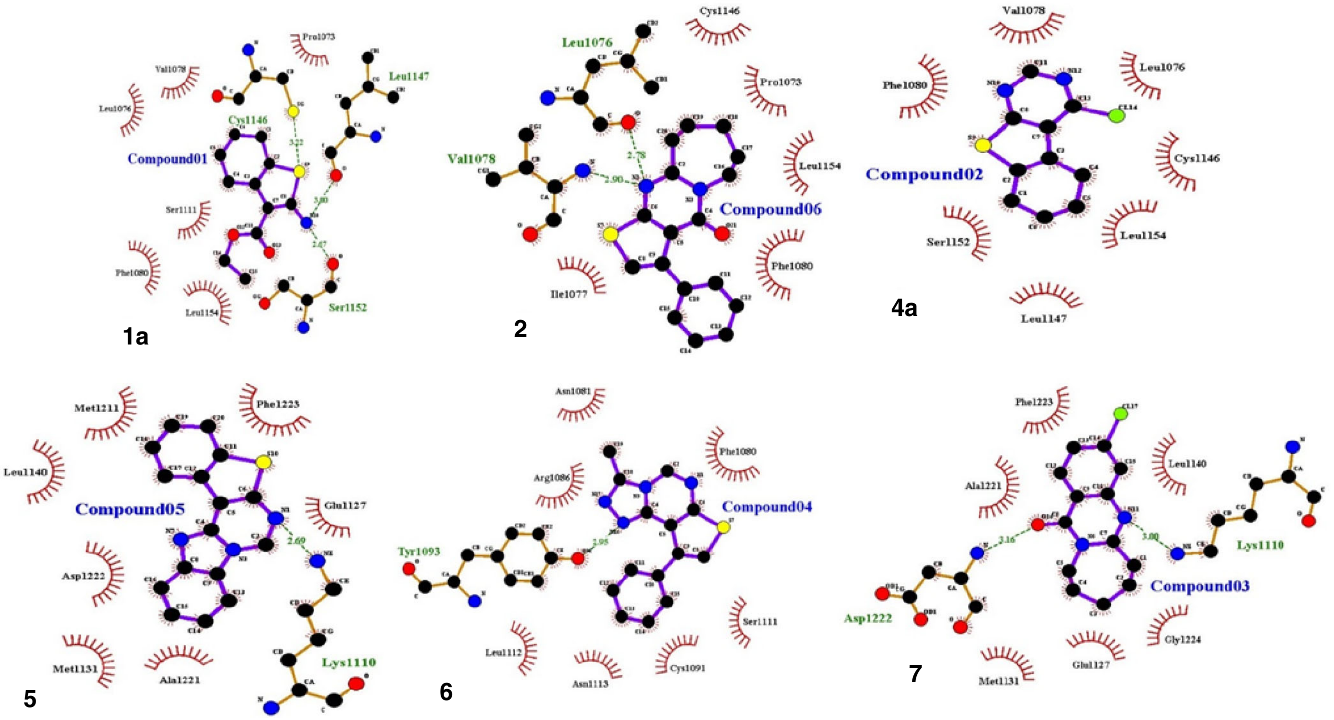


Fig. 5. Interaction of compounds **1a**, **2**, **4a**, **5**, **6**, and **7** with *c*-Met

In VEGF2, Cys919 was found to interact with the compounds **2**, **4a**, **5** and **7** by forming hydrogen bonds whereas, in compound **1a**, Arg863, Phe918 and Ser1037 formed H-bonds. Residues such as Leu840, Val848, Ala866, Thr916, Glu917, Phe918, Leu1035 and Phe1047 formed hydrophobic interactions with all the compounds. In FGFR1, Lys714 and Arg675 formed an H-bond interaction and Leu672, Phe673, Asp674 and Lys714 and Phe710 formed hydrophobic interactions with compounds **1a, 4a, 6** and **7**. Hydrophobic interaction was further formed by Try666, Lys665, Ser699, Gly703, Val704, Pro705 and Val705 in compound **7** and Arg623, Arg627, Leu644, Arg661, Leu662, Pro663, Val664, Lys665 and Tyr666 in compound **2**. In c-Met, H-bond interaction was formed by Ser1152, Leu1147 and Cys1146 in compound **1a**, Lys1110, Asp1222 in compound **7,** Tyr1093 in compound **6**, Lys1110 in compound **5** and Leu1076, Val1078 in compound **2**. In compounds **1a** and **4a**, Val1070, Leu1076, Val1078, Phe1080, Leu1147 and Ser1152 were involved in hydrophobic interactions. Ala1221, Met1131, Leu1140, Asp1222, Phe1223, Gly1224 and Glu1127 were involved in hydrophobic interactions with compounds **5** and **7**. In compound **6**, residues such as Phe1080, Asn1081, Arg1086, Cys1091, Ser1111, Leu1112 and Asn1113 and in compound **2** Cys1146, Pro1073, leu1154, Phe1080, Ile1077 formed hydrophobic interactions. Leu840, Glu850, Lys920 and Lys838 residues from VEGF2, Phe673, Asp674, Arg675 and Glu707 residues from FGFR1 and Leu1147 and Ser1152 residues of c-Met have formed H-bond interaction with erlotinib. The score values much high compared to our prepared compounds but indicative of the inhibition of the proteins associated with OSCC. Typical compounds attached with ligands tested on cell lines such as A431 human epidermoid carcinoma and H9c2 rat cardiomyocyte cells, human embryonic kidney cell line (HEK 293) and breast cancer cell line (MCF-7) and their potency have been in the range of 80-85% growth inhibition. The change in the synthetic method by adopting microwave-assisted fusion of 4-chlorothieno[2,3-*d*]pyrimidines with *o*-phenylenediamine afforded target compounds also proved to be efficient against the MCF-7 cell line. However, there was no report on these derivatives with tested cell lines namely human oral squamous carcinoma-3 (HSC-3). Hence, it is of interest to screen their cytotoxicity and to identify the target proteins theoretically. It will help us to design the molecular structure to enhance the specific activity.

Conclusion

In present work, thieno[2,3,*d*]pyrimidines and quinazoline derivatives have been selected for anticancer activity. The compounds synthesized *via* Niementowski condensation. The yield was satisfactory and chemical structures were confirmed. The compounds were screened for anti-proliferative activity against the HSC-3 cell line. The preliminary investigations on the anti-proliferative effect of the HSC-3 cell line revealed that 4-chloro-5,6,7,8-tetrahydrobenzo[4,5]thieno[2,3-*d*]pyrimidine (**4a**) showed moderate inhibitory activity compared to that of other modified thienopyrimidine and quinazoline derivatives. When compared with thienopyrimidine derivatives, the parent compounds showed moderate inhibitory activity. Molecular docking results showed that these compounds could interact with the binding sites of VEGF, FGFR and c-Met proteins and inhibit the proteins responsible for OSCC. Lack of chirality and the presence of bulky substituents in a few of the compounds are found to be the cause for lower potency. Further designing of the compounds using inputs from molecular docking studies could yield better candidates for the treatment of cancer.

ACKNOWLEDGEMENTS

The authors are grateful to the authorities of SRM Institute of Science & Technology, SRM Nagar, Kattankulathur, India. Utilization of the facility sanctioned under DST-FIST, Government of India to the Department of Chemistry is greatly acknowledged.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

1. E.Z. Elrazaz, R.A.T. Serya, N.S.M. Ismail, D.A. Abou El Ella and K.A.M. Abouzid *Future J. Pharm. Sci.*, **1**, 33 (2015); https://doi.org/10.1016/j.fjps.2015.09.001
2. N.G. Haswani and S.B. Bari, *Turk. J. Chem.*, **35**, 915 (2011).
3. M.F. El-Shehry, H.M. Hosni, A.E. Amr, A.A. Ibrahim, A.A. Fayed and D.H. Elnaggar, *Russ. J. Gen. Chem.*, **89**, 1528 (2019); https://doi.org/10.1134/S1070363219070247
4. A. Cohen, P. Suzanne, J.C. Lancelot, P. Verhaeghe, A. Lesnard, L. Basmaciyan, S. Hutter, M. Laget, A. Dumètre, L. Paloque, E. Deharo, M.D. Crozet, P. Rathelot, P. Dallemagne, A. Lorthiois, C.H. Sibley, P. Vanelle, A. Valentin, D. Mazier, S. Rault and N. Azas, *Eur. J. Med. Chem.*, **95**, 16 (2015); https://doi.org/10.1016/j.ejmech.2015.03.011
5. A. Angell, C. McGuigan, L. Garcia Sevillano, R. Snoeck, G. Andrei, E. De Clercq and J. Balzarini, *Bioorg. Med. Chem. Lett.*, **14**, 2397 (2004); https://doi.org/10.1016/j.bmcl.2004.03.029
6. H.M. Ashour, O.G. Shaaban, O.H. Rizk and I.M. El-Ashmawy, *Eur. J. Med. Chem.*, **62**, 341 (2013); https://doi.org/10.1016/j.ejmech.2012.12.003
8. Y. Kotaiah, N. Harikrishna, K. Nagaraju and C. Venkata Rao, *Eur. J. Med. Chem.*, **58**, 340 (2012); https://doi.org/10.1016/j.ejmech.2012.10.007
8. A.A. El-Tombary, S.A.M. El-Hawash, N.S. Habib, R. Soliman, I.M. El-Ashmawy and O.G. Shaaban, *Med. Chem.*, **9**, 1099 (2013); https://doi.org/10.2174/1573406411309080012
9. R.K. Russell, J.B. Press, R.A. Rampulla, J.J. McNally, R. Falotico, J.A. Keiser, D.A. Bright and A. Tobia, *J. Med. Chem.*, **31**, 1786 (1998); https://doi.org/10.1021/jm00117a019
10. B. Shireesha, K.U. Shankar, A.R. Rao, K.S. Rajan and M. Raghuprasad, *Int. J. Pharm. Sci. Nanotech.*, **3**, 136 (2008); https://doi.org/10.37285/ijpsn
11. S. Pédeboscq, D. Gravier, F. Casadebaig, G. Hou, A. Gissot, F. De Giorgi, F. Ichas, J. Cambar and J.-P. Pometan, *Eur. J. Med. Chem.*, **45**, 2473 (2010); https://doi.org/10.1016/j.ejmech.2010.02.032
12. A.S. Oganisyan, A.S. Noravyan, I.A. Dzhagatspanyan, I.M. Nazaryan and A.G. Akopyan, *Pharm. Chem. J.*, **41**, 588 (2007); https://doi.org/10.1007/s11094-008-0028-7
13. M.K. Kathiravan, K.D. More, V.K. Raskar, K.S. Jain, M. Maheshwar, S. Gadhwe, D.P. Jain, M.A. Nagras and M.S. Phoujdar, *Med. Chem. Res.*, **22**, 4286 (2013); https://doi.org/10.1007/s00044-012-0263-z
14. R. Tamboli, R. Amrutkar, K. Jain and M. Kathiravan, *Lett. Drug Des. Discov.*, **10**, 906 (2013); https://doi.org/10.2174/15701808113109990019
15. A.A. Gryshchenko, V.G. Bdzhola, A.O. Balanda, N.V. Briukhovetska, I.M. Kotey, A.G. Golub, T.P. Ruban, L.L. Lukash and S.M. Yarmoluk, *Bioorg. Med. Chem.*, **23**, 2287 (2015); https://doi.org/10.1016/j.bmc.2014.12.044
16. J. Li, W. Gu, X. Bi, H. Li, C. Liao, C. Liu, W. Huang and H. Qian, *Bioorg. Med. Chem.*, **25**, 6674 (2017); https://doi.org/10.1016/j.bmc.2017.11.010
17. A. TsMavrova, S. Dimov, D. Yancheva, A. Kolarevic, B.S. Ilic, Kocic G. and A. Šmelcerovic, *Bioorg. Chem.*, **80**, 693 (2018); https://doi.org/10.1016/j.bioorg.2018.07.009
18. S.N. Milik, A.K. Abdel-Aziz, D.S. Lasheen, R.A.T. Serya, S. Minucci and K.A.M. Abouzid, *Eur. J. Med. Chem.*, **155**, 316 (2018); https://doi.org/10.1016/j.ejmech.2018.06.011
19. C.H. Wu, M.H. Coumar, C.Y. Chu, W.H. Lin, Y.R. Chen, C.T. Chen, H.Y. Shiao, S. Rafi, S.Y. Wang, H. Hsu, C.H. Chen, C.Y. Chang, T.Y. Chang, T.W. Lien, M.Y. Fang, K.C. Yeh, C.P. Chen, T.P. Yeh, S.H. Hsieh, J.T. Hsu, C.C. Liao, Y.S. Chao and H.P. Hsieh, *J. Med. Chem.*, **53**, 7316 (2010); https://doi.org/10.1021/jm100607r
20. R. Rheault, T.R. Caferro, S.H. Dickerson, K.H. Donaldson, M.D. Gaul, A.S. Goetz, R.J. Mullin, O.B. McDonald, K.G. Petrov, D.W. Rusnak, L.M. Shewchuk, G.M. Spehar, A.T. Truesdale, D.E. Vanderwall, E.R. Wood and D.E. Uehling, *Bioorg. Med. Chem. Lett.*, **19**, 817 (2009); https://doi.org/10.1016/j.bmcl.2008.12.011
21. Y. Dai, Y. Guo, R.R. Frey, Z. Ji, M.L. Curtin, A.A. Ahmed, D.H. Albert, L. Arnold, S.S. Arries, T. Barlozzari, J.L. Bauch, J.J. Bouska, P.F. Bousquet, G.A. Cunha, K.B. Glaser, J. Guo, J. Li, P.A. Marcotte, K.C. Marsh, M.D. Moskey, L.J. Pease, K.D. Stewart, V.S. Stoll, P. Tapang, N. Wishart, S.K. Davidsen and M.R. Michaelides, *J. Med. Chem.*, **48**, 6066 (2005); https://doi.org/10.1021/jm050458h
22. W.J. McClellan, Y. Dai, C. Abad-Zapatero, D.H. Albert, J.J. Bouska, K.B. Glaser, T.J. Magoc, P.A. Marcotte, D.J. Osterling, K.D. Stewart, S.K. Davidsen and M.R. Michaelides, *Bioorg. Med. Chem. Lett.*, **21**, 5620 (2011); https://doi.org/10.1016/j.bmcl.2011.06.041
23. G.K. Packard, P. Papa, J.R. Riggs, P. Erdman, L. Tehrani, D. Robinson, R. Harris, G. Shevlin, S. Perrin-Ninkovic, R. Hilgraf, M.A. McCarrick, T. Tran, Y. Fleming, A. Bai, S. Richardson, J. Katz, Y. Tang, J. Leisten, M. Moghaddam, B. Cathers, D. Zhu and S. Sakata, *Bioorg. Med. Chem. Lett.*, **22**, 747 (2012); https://doi.org/10.1016/j.bmcl.2011.03.006
24. M.M. Kandeel, H.M. Refaat, A.E. Kassab, I.G. Shahin and T.M. Abdelghany, *Eur. J. Med. Chem.*, **90**, 620 (2015); https://doi.org/10.1016/j.ejmech.2014.12.009
25. M.M. Fouad, E.R. El-Bendary, G.M. suddek, I.A. Shehata and M.M. El-Kerdawy, *Bioorg. Chem.*, **81**, 587 (2018); https://doi.org/10.1016/j.bioorg.2018.09.022
26. S. Kaliraj and M.K. Kathiravan, *Curr. Bioact. Compd.*, **16**, 133 (2020); https://doi.org/10.2174/1573407214666180808124802
27. S. Kaliraj, R. Jeyalakshmi and M.K. Kathiravan, *Pharm. Chem. J.*, **54**, 258 (2020); https://doi.org/10.1007/s11094-020-02188-w
28. G.M. Morris, R. Huey, W. Lindstrom, M.F. Sanner, R.K. Belew, D.S. Goodsell and A.J. Olson, *J. Comput. Chem.*, **30**, 2785 (2009); https://doi.org/10.1002/jcc.21256
29. R.A. Friesner, J.L. Banks, R.B. Murphy, T.A. Halgren, J.J. Klicic, D.T. Mainz, M.P. Repasky, E.H. Knoll, M. Shelley, J.K. Perry, D.E. Shaw, P. Francis and P.S. Shenkin, *J. Med. Chem.*, **47**, 1739 (2004); https://doi.org/10.1021/jm0306430
30. T.A. Halgren, R.B. Murphy, R.A. Friesner, H.S. Beard, L.L. Frye, W.T. Pollard and J.L. Banks, *J. Med. Chem.*, **47**, 1750 (2004); https://doi.org/10.1021/jm030644s
31. A.E.-S. Mghwary, E.M. Gedawy, A.M. Kamal and S.M. Abuel-Maaty, *J. Enzyme Inhib. Med. Chem.*, **34**, 838 (2019); https://doi.org/10.1080/14756366.2019.1593160
32. S. Bugge, S.J. Kaspersen, S. Larsen, U. Nonstad, G. Bjørkøy, E. Sundby and B.H. Hoff, *Eur. J. Med. Chem.*, **75**, 354 (2014); https://doi.org/10.1016/j.ejmech.2014.01.042
33. M.H. Norman, L. Liu, M. Lee, N. Xi, I. Fellows, N.D. D’Angelo, C. Dominguez, K. Rex, S.F. Bellon, T.S. Kim and I. Dussault, *J. Med. Chem.*, **55**, 1858 (2012); https://doi.org/10.1021/jm201330u
34. J.H. Bae, E.D. Lew, S. Yuzawa, F. Tomé, I. Lax and J. Schlessinger, *Cell*, **138**, 514 (2009); https://doi.org/10.1016/j.cell.2009.05.028
35. F. Qian, S. Engst, K. Yamaguchi, P. Yu, K.A. Won, L. Mock, T. Lou, J. Tan, C. Li, D. Tam, J. Lougheed, F.M. Yakes, F. Bentzien, W. Xu, T. Zaks, R. Wooster, J. Greshock and A.H. Joly, *Cancer Res.*, **69**, 8009 (2009); https://doi.org/10.1158/0008-5472.CAN-08-4889