Asian Journal of Chemistry; Vol. 36, No. 12 (2024), 2855-2860

ASIAN JOURNAL OF CHEMISTRY

https://doi.org/10.14233/ajchem.2024.32708

Thiazolyl-Thiazolidinone Conjugated Pyrazoles as Potential Anticancer and Antibacterial Agents and Molecular Docking Studies

PREMANAND M. HONAVAR[1,2], BHUVANESH SUKHLAL KALAL[3,4], RANJITH RAJ[5], P.T. ANJANA[6], P.V. DEEPTHI[7], SHARATHA CHANDRA[8], A.M. VIJESH[9], RAMITH RAMU[10], VINITHA RAMANATHA PAI[3], NAVEEN KUMAR[11], VASANTHA KUMAR[11,*] and B. SHIVARAMA HOLLA[11,*]

[1]Department of Chemistry, Sri Dharmasthala Manjunatheshwara College of Arts, Science, Commerce, Honavar-581334, India
[2]Department of Chemistry, Tumkur University, Tumkur-572103, India
[3]Department of Biochemistry, Yenepoya Medical College, Yenepoya (Deemed to be University), Deralakatte, Mangaluru-575018, India
[4]Department of Pharmacology and Nutritional Sciences, College of Medicine, University of Kentucky, Lexington, Kentucky 40508, USA
[5]Department of Pharmacology, JSS Medical College, JSS Academy of Higher Education and Research, Mysuru-570015, India
[6]Department of Chemistry, Krishna Menon Memorial Government Women's College (Affiliated to Kannur University), Pallikkunnu, Kannur-670004, India
[7]Department of Chemistry, Nehru Arts and Science College (Affiliated to Kannur University), Kanhangad-671314, India
[8]Department of Biosciences, Mangalore University, Mangalagangothri-574199, India
[9]P.G. Department of Chemistry, Payyanur College (Affiliated to Kannur University), Payyanur-670327, India
[10]Department of Biotechnology and Bioinformatics, School of Life Sciences, JSS Academy of Higher Education and Research, Mysuru-570015, India
[11]Department of PG Studies and Research in Chemistry, Sri Dharmasthala Manjunatheshwara College (Autonomous), Ujire-574240, India

*Corresponding authors: E-mail: vasantha.kumar1886@gmail.com; hollabs@gmail.com

Received: 13 September 2024; *Accepted*: 26 October 2024; *Published online*: 30 November 2024; AJC-21827

In current study, five new compounds containing thiazolyl-thiazolidinone conjugated substituted pyrazoles were prepared and screened for anticancer and antimicrobial activities. Synthesized compounds were characterized by spectroscopic techniques. Anticancer activity was carried out against human melanoma cancer A375 and human breast cancer MDA-MB-231 cell lines. Also, these were screened for antibacterial activity against two Gram-positive *Staphylococcus aureus*, *Bacillus subtilis* strains and three Gram-negative *Pseudomonas aeruginosa*, *Klebsiella pneumonia* and *Escherichia coli* strains. Anticancer studies have identified compound **6e** as highly potent compound with IC_{50} = 13.08 µg/mL against A375 cell line and 14.21 µg/mL against MDA-MB-231 cell line. In addition, compound **6c** also showed very good activity against A375 cell line with IC_{50} = 13.22 µg/mL. Antibacterial activity results revealed compound **6a** and **6e** as highly promising agents against all the tested bacterial strains which is equipotent to the standard drug. Molecular docking studies on Aurora A kinase also revealed favourable interactions for target compounds through hydrogen bonds.

Keywords: Thiazolidinones, Pyrazoles, Anticancer activity, Antibacterial activity, Docking studies, Aurora A kinase.

INTRODUCTION

Thiazolidinone scaffolds are the privileged small molecules with unique properties in medicinal chemistry [1]. Owing to the extraordinary biological profile of these molecules, these are considered as magic moiety in medicinal chemistry research. It exhibits several different pharmacological activities such as antibacterial [2], antitubercular [3], anti-inflammatory [4], anticancer [5] and antiviral [6]. Thiazoles are also promising heterocyclic entity in pharmaceutical field [7]. Incorporation of this small motif in different molecules enhances the bioavailability of the drug candidates. It was noteworthy to mention the applications of clubbed pharmacophore of thiazole and thiazolidinones in medicinal chemistry especially in antimicrobial and anticancer field [8]. Thiazole endowed thiazolidinone derivatives were found to exhibit diverse biological activities such as antimicrobial [9], antitubercular [10], anti-inflammatory [11] and anticancer [12].

On the other hand, pyrazoles are considered as an auspicious heterocyclic system in synthetic and medicinal chemistry [13]. The importance of this heterocyclic system is evident from various drug candidates such as celecoxib, lonazolac, tepoxalin, *etc*. Medicinal chemists are continuously working towards the discovery of novel molecules either by derivatizing different functional groups in a heterocyclic system or attaching newer heterocyclic pharmacophores to other pharmacophores by a hybrid strategy. Hence, many pyrazole derivatives exhibited their potency in broad therapeutic areas such as antimicrobial [14], anti-inflammatory [15], anticancer [16], antiviral [17], *etc*. and research is still going on in this field to generate newer molecules that can combat various diseases.

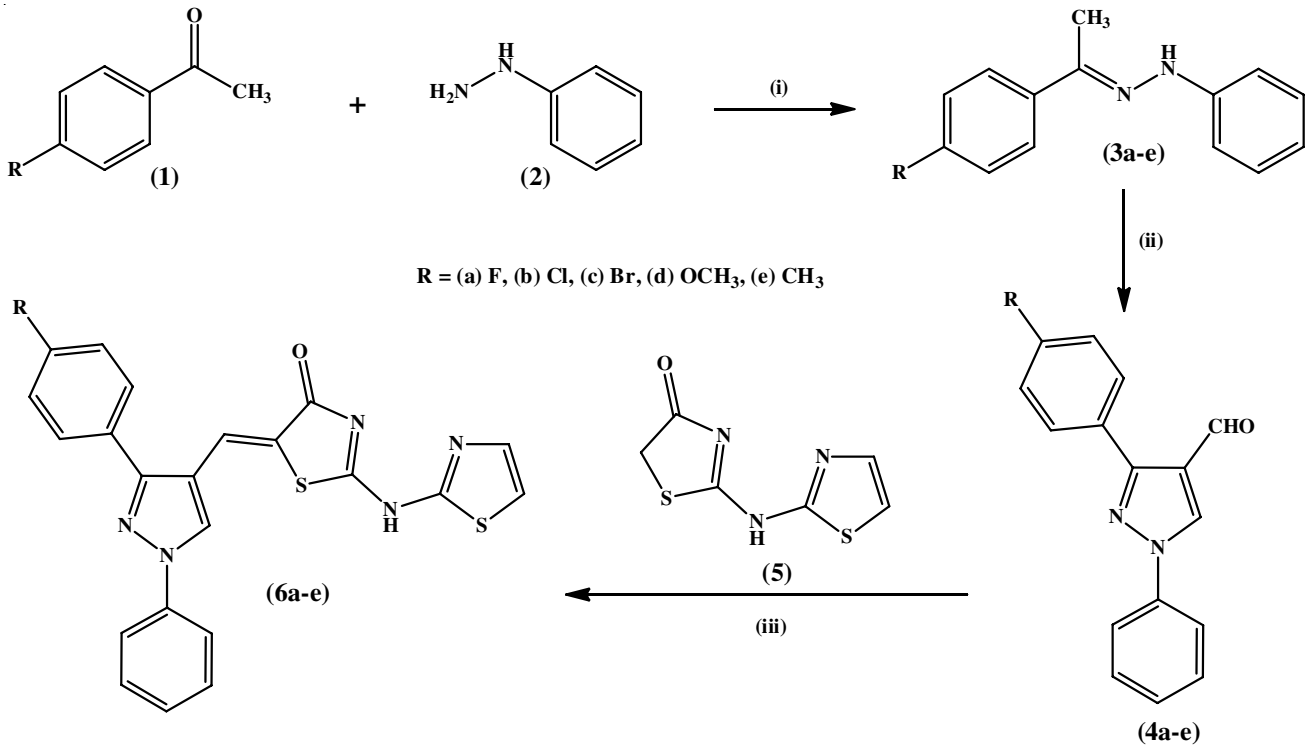
Based on the biological importance of thiazolidinones and pyrazoles and in continuation of our research on thiazolidinones [18-24] and pyrazoles [25,26], in the present study a new series of hybridized molecules were synthesized by combining these two pharmacophores in one framework. The synthesized compounds were screened for anticancer activity against two cancer cell lines A375 human melanoma cancer and MDA-MB-231 breast cancer cell lines using MTT assay. Also, these compounds were screened for antibacterial activity. Molecular docking studies was performed on Aurora A kinase (PDB: 4ZTR) to understand and to correlate the binding interactions with that of observed anticancer activity

EXPERIMENTAL

All the starting materials and reagents were procured from Sigma Aldrich, Spectrochem and were used as such. FTIR spectra were recorded on a Shimadzu ATR Spectrometer. Thin layer chromatography (TLC) was performed on a silica coated aluminium sheet (silica gel 60F254) using ethyl acetate and hexane (3:7, v/v). ^{1}H and ^{13}C NMR spectra were recorded on VNMRS 400 and 100 MHz instruments respectively using TMS as an internal standard. The chemical shifts are reported in δ units and the coupling constants (*J*) are reported in Hertz. Mass spectra were recorded in Agilent Technology LC-mass spectrometer.

General procedure for the synthesis of substituted phenyl hydrazones (3a-e): Mixture of substituted acetophenone (**1**) (0.019 mol) and phenyl hydrazine (**2**) (0.20 mol) in ethanol (25 mL) medium were taken in round bottomed flask. To this catalytic amount of glacial acetic acid (~3-4 drops) was added and the resulting reaction mixture was refluxed for 6 h. Progress of the reaction was monitored by TLC (thin layer chromatography) with combination of ethyl acetate and hexane (7:3 v/v) as solvent system. After completion of reaction, the reaction mixture was poured onto crushed ice and solid precipitate thus obtained was filtered, washed with cold water and dried to get substituted phenyl hydrazones (**3a-e**).

General procedure for the synthesis of 1,3-diphenyl-1*H*-pyrazole-4-carbaldehydes (4a-e): Substituted phenyl hydrazone (**3a-e**) (0.01 mol) and DMF (25 mL) were taken in the round bottomed flask, to this $POCl_3$ (0.05 mol) was added drop wise at 0-5 ºC. After complete addition, resulting mixture initially stirred at room temperature for 1 h and heated to 80 ºC for 10 h. Progress of the reaction was monitored by thin layer chromatography with solvent system ethyl acetate and *n*-hexane (8:2 v/v). After the completion of the reaction, the reaction mixture was poured into beaker containing crushed ice and solid precipitate thus obtained was filtered, washed with cold water and dried. Crude sample was recrystallized using DMF to get pure pyrazole-4-carbaldehydes (**4a-e**) (**Scheme-I**).



Scheme-I: Synthesis of compounds **6a-e**. Reagents and conditions: (i) EtOH, glacial acetic acid, reflux, 6 h; (ii) DMF-$POCl_3$, 0-5 °C, 1 h at rt, then 80 °C, 10 h; (iii) anhydrous sodium acetate, glacial acetic acid, reflux, 10 h

General procedure for the synthesis of thiazolyl-thiazolidinone-pyrazole hybrids (6a-e): To a well stirred solution of 2-(thiazolamino) thiazole-4(5*H*)-one (**5**, 0.05 mol) in 30 mL of glacial acetic acid was buffered with anhydrous sodium acetate (0.12 mol). To this solution, appropriate pyrazole-4-carbaldehyde (**4a-f**, 0.06 mol) was added and the resulting solution was refluxed for 10 h. The progress of the reaction was monitored by TLC. After the completion of the reaction, the reaction mixture was cooled at room temperature and solid obtained was filtered and washed with water. The crude products were stirred in glacial acetic acid to remove the impurities and filtered off. Pure compounds of pyrazole-thiazolidinones derivatives (**6a-f**) were obtained by recrystallization process from DMF solvent.

(5*Z*)-5-((3-(4-Fluorophenyl)-1-phenyl-1*H*-pyrazol-4-yl)methylene)-2-(thiazol-2-ylamino)thiazol-4(5*H*)-one (6a): Yield: 78%; m.p.: > 260 ºC; ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm: 12.60 (s, 1H, N-H), 8.75 (s, 1H, pyrazole-H), 7.97 (d, 2H, Ar-H), 7.70-7.66 (m, 3H, Ar-H & thiazole-H), 7.56 (t, 2H, *J* = 8 Hz, Ar-H), 7.48 (s, 1H, =CH), 7.466 (d, 1H, thiazole-H), 7.41-7.36 (m, 3H, Ar-H); ^{13}C NMR (100 MHz, DMSO-d_6) δ ppm: 173.78, 171.66, 168.85, 159.74, 157.47, 145.49, 143.97, 135.87, 135.79, 134.74, 133.55, 133.03, 132.61, 129.05, 126.94, 124.67, 122.45, 121.13, 120.91, 120.63; LCMS (*m/z*): 448.24 (M+1); Anal. calcd. (found) % for $C_{22}H_{14}N_5OS_2F$: C, 59.05 (59.00); H, 3.15 (3.22); N, 15.65 (15.70).

(5*Z*)-5-((3-(4-Chlorophenyl)-1-phenyl-1*H*-pyrazol-4-yl)methylene)-2-(thiazol-2-ylamino)thiazol-4(5*H*)-one (6b): Yield: 78%; m.p.: > 260 ºC; ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm: 12.59 (s, 1H, N-H), 8.74 (s, 1H, pyrazole-H), 7.97 (d, 2H, *J* = 8.4 Hz, Ar-H), 7.80 (d, 2H, *J* = 8 Hz, Ar-H), 7.68 (d, 1H, *J* = 4 Hz, thiazole-H), 7.56-7.52 (m, 4H, Ar-H), 7.47 (s, 1H, =CH), 7.456 (d, 1H, *J* = 3.6 Hz, thiazole-H); ^{13}C NMR (100 MHz, DMSO-d_6) δ ppm: 173.70, 171.58, 159.66, 157.10, 145.44, 143.87, 137.45, 135.70, 134.70, 133.62, 133.65, 132.60, 130.55, 131.50, 128.45, 127.50, 126.72, 124.62, 122.40, 120.62; LCMS (*m/z*): 463.85.14 (M+), 465.75 (M+2); Anal. calcd. (found) % for $C_{22}H_{14}N_5OS_2Cl$: C, 56.95 (56.90); H, 3.04 (3.09); N, 15.10 (15.16).

(5*Z*)-5-((3-(4-Bromophenyl)-1-phenyl-1*H*-pyrazol-4-yl)methylene)-2-(thiazol-2-ylamino)thiazol-4(5*H*)-one (6c): Yield: 73%; m.p.: > 260 ºC; ^{1}H NMR (400 MHz, DMSO-d_6) , δ ppm: 12.61 (s, 1H, N-H), 8.74 (s, 1H, pyrazole-H), 7.97 (d, 2H, *J* = 8.4 Hz, Ar-H), 7.74 (d, 2H, *J* = 8 Hz, Ar-H), 7.675 (d, 1h, *J* = 4 Hz, thiazole-H), 7.59-7.53 (m, 4H, Ar-H), 7.48-7.41 (m, 3H, Ar-H & thiazole-H); ^{13}C NMR (100 MHz, DMSO-d_6) δ ppm: 173.76, 171.61, 159.71, 157.17, 145.49, 143.92, 137.00, 135.77, 135.59, 134.74, 133.69, 132.66, 129.33, 127.56, 126.79, 124.68, 122.45, 120.68; LCMS (*m/z*): 508.14 (M+), 510.10 (M+2); Anal. calcd. (found) % for $C_{22}H_{14}N_5OS_2Br$: C, 51.97 (51.91); H, 2.78 (2.85); N, 13.78 (13.86).

(5*Z*)-5-((3-(4-Methoxyphenyl)-1-phenyl-1*H*-pyrazol-4-yl)methylene)-2-(thiazol-2-ylamino)thiazol-4(5*H*)-one (6d): Yield: 76%; m.p.: > 260 ºC; ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm: 12.62 (s, 1H, N-H), 8.73 (s, 1H, pyrazole-H), 7.96 (d, 2H, *J* = 8.4 Hz, Ar-H), 7.68-7.65 (m, 3H, Ar-H & thiazole-H), 7.58 (t, 2H, *J* = 8 Hz, Ar-H), 7.49 (s, 1H, =CH), 7.454 (d, 1H, thiazole-H), 7.41 (t, 1H, *J* = 8 Hz, Ar-H), 6.95 (d, 2H, *J* = 8.4 Hz, Ar-H); ^{13}C NMR (100 MHz, DMSO-d_6) δ ppm: 173.74, 171.60, 162.56, 159.69, 157.15, 145.46, 143.90, 134.76, 133.66, 132.63, 129.55, 127.53, 126.72, 126.50, 124.64, 122.41, 120.65, 114.65, 56.50; LCMS (*m/z*): 459.68 (M+); Anal. calcd. (found) % for $C_{23}H_{17}N_5O_2S_2$: C, 60.11 (60.18); H, 3.73 (3.67); N, 15.24 (15.30).

(5*Z*)-5-((3-(4-Methylphenyl)-1-phenyl-1*H*-pyrazol-4-yl)methylene)-2-(thiazol-2-ylamino)thiazol-4(5*H*)-one (6e): Yield: 70%; m.p.: > 260 ºC; ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm: 12.60 (s, 1H, N-H), 8.73 (s, 1H, pyrazole-H), 7.97 (d, 2H, *J* = 8 Hz, Ar-H), 7.675 (d, 1H, *J* = 4 Hz, thiazole-H), 7.58 (t, 2H, *J* = 8.4 Hz, Ar-H), 7.49 (s, 1H, =CH), 7.41-7.46 (m, 4H, Ar-H), 7.23 (d, 2H, *J* = 8 Hz, Ar-H); ^{13}C NMR (100 MHz, DMSO-d_6) δ ppm: 173.75, 171.58, 159.68, 157.14, 145.49, 143.92, 136.55, 134.74, 134.00, 133.69, 132.66, 130.55, 128.00, 127.56, 126.79, 124.68, 122.45, 120.68, 23.5; LCMS (*m/z*): 443.80 (M+); Anal. calcd. (found) % for $C_{23}H_{17}N_5OS_2$: C, 62.28 (62.36); H, 3.86 (3.80); N, 15.79 (15.86).

Anticancer activity assay: Anticancer activity of the synthesized hybrid compounds **6a-e** was studied against two cancer cell lines, human melanoma A375 and breast cancer MDA-MB-231 cell lines by MTT assay as described earlier [27].

Antibacterial activity assay: Antibacterial activities for the synthesized hybrid compounds **6a-e** carried out against Gram-positive *Staphylococcus aureus, Bacillus subtilis* strains and Gram-negative *Pseudomonas aeruginosa, Klebsiella pneumonia* and *Escherichia coli* strain by tube dilution method. The MIC values were also determined by tube dilution method as described earlier [27].

Docking studies: The structures of the molecules **6a-e** were drawn in ChemDraw 11.0 (saved as mol files) and the energies were minimized using ADS. The minimized ligands and proteins were saved in structure data (.sd) and protein data bank (pdb) format respectively for further studies. The docking study was performed using Accelyrs Discovery Studio client version 3.5 software (Accelyrs Inc., http://www.accelrys.com). The X-ray crystallographic structures of all proteins (PDB ID: 4ZTR bound with other inhibitors were acquired from the protein data bank (PDB). The active site was defined around the bound inhibitor which covered all the active site amino acids of the target protein. A grid-based molecular docking method, C-DOCKER algorithm was used to dock the small molecules (ligands) into the protein active site. The designed structures were submitted to CHARMm (Chemistry at HARvard Macromolecular Mechanics) force field for structure refinement. All water molecules, bound inhibitor and other hetero atoms were removed from the macromolecule and polar hydrogen atoms were added. Energy minimization was carried out for all compounds using CHARMm force field to make stable conformation of protein with an energy gradient of 0.01 kcal/mol/Å. A final minimization of the ligand in the rigid receptor using non-softened potential was performed. For each final pose, the CHARMm energy (interaction energy plus ligand strain) and the interaction energy alone were calculated. The poses were sorted by CHARMm energy and the top scoring (most negative, thus favourable to binding) poses.

RESULTS AND DISCUSSION

The synthetic strategy adopted for the novel compounds thiazolyl-thiazolidinone conjugated pyrazoles (**6a-e**) is depicted in **Scheme-I**. Five substituted pyrazole aldehydes (**4a-e**) were synthesized by two step method. In the first step, substituted acetophenones (**1a-e**) and phenylhydrazine (**2**) were refluxed in ethanol medium in the presence of catalytic amount of glacial acetic acid for 3h to yield corresponding hydrazones (**3a-e**). The resulting hydrazones (**3a-e**) were upon Vilsmeir-Haack formylation reaction with DMF-$POCl_3$ reagent yielded substituted pyrazole aldehydes **4a-e**. The final compounds **6a-e** were prepared by refluxing **3a-e** with thiazolyl-thiazolidinone (**5**) in buffered glacial acetic acid.

The assigned structures of synthesized derivatives were characterized by ^1H & ^{13}C NMR and LC-MS spectrometry. ^1H NMR spectrum of the compound **6a** shows existence of singlet peaks at δ 12.60 and 8.75 ppm were integrated to the vinylic and pyrazole ring protons respectively. Two protons of thiazole ring were resonated at δ 7.99-7.97 ppm as doublet. Aromatic protons signals were observed in the range of δ 7.70-7.67 and 7.41-7.36 ppm along with a proton of amine group (NH) linking thiazole and thiazolidinones ring confirms the formation. Further, the formation of compound **6a** was supported by ^{13}C NMR spectrum with signal at δ 173.78, 171.66, 168.85, 159.74 ppm corresponds to -C=O; C=N & -C=S group carbon atoms, signals at δ 157.47 (C=C), 145.49, 143.97, 135.87, 135.79, 134.74, 133.55, 133.03, 132.61, 129.05, 126.94, 124.67, 122.45, 121.13, 120.91, 120.63 ppm are attributed to alkene as well as aromatic carbons atoms. The mass spectrum of compound **6a** shows molecular ion peak at *m/z* 448.24 (M+1) corresponding to its molecular mass+1 confirms the formation.

Anticancer activity studies: All the compounds **6a-e** were screened for their anticancer activity against two cancer cell lines namely A375 human melanoma cancer and MDA-MB-231 breast cancer cell lines using MTT assay. From the study, it is observed that all the compounds displayed good anticancer activity against MDA-MB 231 cancer cell lines than A375 cell lines (Table-1). In the series, compound **6e** having electron releasing methyl substitution at 4th position displayed excellent activity with IC_{50} = 13.08 μg/mL and it also exhibited good activity against A375 cell line with IC_{50} = 14.21 μg/mL. Next potent compound **6c** having electronegative bromine displayed very good activity with IC_{50} = 13.22 μg/mL against MDA-MB-231 and it did not show promising activity against A375 cell lines. Further, compound **6b** with 4-chloro substitution exhibited good activity with IC_{50} = 18.79 μg/mL and against A375 it showed moderate activity with IC_{50} = 27.02 μg/mL. other two compounds **6a** with 4-fluoro and **6d** with 4-methoxy substitution showed less activity in the series.

TABLE-1
ANTICANCER ACTIVITIES DATA OF SYNTHESIZED COMPOUNDS **6a-e**

Compd.	R	IC_{50} (μg/mL)	
		A375	MDA-MB-231
6a	F	33.41	39.13
6b	Cl	27.02	18.79
6c	Br	34.66	13.22
6d	OCH_3	38.72	34.83
6e	CH_3	14.21	13.08
Cisplatin	–	3.9	11.2

Antibacterial activity studies: Final compounds **6a-e** were screened for their antibacterial activity against various Gram-positive and Gram-negative bacterial strains by zone of inhibition and MIC values were determined by tube dilution method. Out of five derivatives, compounds **6a** and **6e** emerged as potent against all the strains (Table-2). Compound **6e** exhibited 2.5 times much better activity against *P. aeruginosa* (MIC 2.00 μg/mL), equipotent activity against *B. subtilis* (MIC 3.50 μg/mL) and *E. coli* (MIC 6.00 μg/mL) when compare to the standard drug ampicilin. Against other strains it showed reasonable activity (MIC 6.00-12.50 μg/mL). Compound **6a** showed pronounced activity against *P. aeruginosa* (MIC 5.00 μg/mL) and good inhibition against *B. subtilis* (5.00 μg/mL) but moderate towards *S. aureus, E. coli* but poor towards *K. pneumonia*. Compound **6b** exhibited reasonable activity against *P. aeruginosa* (MIC 6.00 μg/mL). Further compounds **6c** and **6d** exhibited moderate activity towards all the bacterial strains.

In the pyrazole conjugated compounds **6a-e**, SAR can be drawn by considering the substitutions in the aryl part at the 3rd position of pyrazole moiety. From the three halogenated compounds, **6a** emerged potent which contain a smaller fluorine atom at 4th position. The chloro and bromo substituted compounds resulted in decrease of activity. This decreasing in activity may be due to increase in size of halogen atom *i.e.* **6a > 6b > 6c**. From the two electron-releasing substituted compounds **6d** (4-OMe) and **6e** (4-Me); **6e** is more active than **6d**. The methyl group in **6e** may play a crucial role in the inhibition process.

Molecular docking studies on Aurora A kinase protein: Molecular docking studies was carried out for all the synthesized compounds on Aurora A kinase (PDB: 4ZTR) to under-

TABLE-2
ANTIBACTERIAL ACTIVITY DATA OF SYNTHESIZED COMPOUNDS **6a-e**

Compound	MIC (μg/mL)				
	Staphylococcus aureus	*Bacillus subtilis*	*Pseudomonas aeruginosa*	*Klebsiella pneumonia*	*Escherichia coli*
6a	8.00	5.00	5.00	17.0	12.5
6b	15.5	18.0	6.00	20.0	12.5
6c	18.0	12.5	12.0	20.0	15.0
6d	20.0	25.0	14.0	22.5	15.0
6e	12.5	3.50	2.00	15.0	6.00
Ampicillin	2.80	3.50	5.10	6.75	5.00

*Mean of triplicate value

stand ligand-protein interactions and to correlate the observed anticancer activity. The binding interactions of most potent compounds **6e** and **6c** with Aurora A kinase protein is depicted in Figs. 1 & 2 and corresponding binding score and binding interactions are depicted in Table-3. From the interaction score, it is clear that compound **6e** has highest score (-28.671) which has highest anticancer activity in the series (IC_{50} = 13.08 µg/mL). The N-H hydrogen of compound **6e** forms hydrogen bond with ASN261 and carbonyl oxygen of thiazolidinones forms two hydrogen bonds with LYS162 and PHE144. Also, thiazole ring formed a π-cation interaction with LYS258 residue. Moreover, N-phenyl ring of pyrazole involved in π-sigma interaction with LEU194 and there are 4 π-alkyl interaction between compound **6e** and protein. These strong interactions may be responsible for its higher anticancer activity.

TABLE-3
BINDING SCORE AND BINDING INTERACTIONS OF COMPOUNDS **6a-e** WITH AURORA A KINASE PROTEIN

Compd.	Docking score	H bonded residue
6a	-28.628	Lys141, Leu139
6b	-26.703	Lys162, Phe275, Asn261
6c	-26.902	Lys162, Phe275, Asn261
6d	-24.924	Ala273
6e	-28.671	Phe144, Lys162, Asn261

The next potent compound **6c** has also exhibited three hydrogen bonding interaction with target protein. As in the compound **6e**, here also N-H and carbonyl oxygen of thiazolidinone ring forms hydrogen bond with ASN261 and LYS162 residue. But thiazole nitrogen involved in hydrogen bonding interaction with PHE275. It also formed a π-donor hydrogen

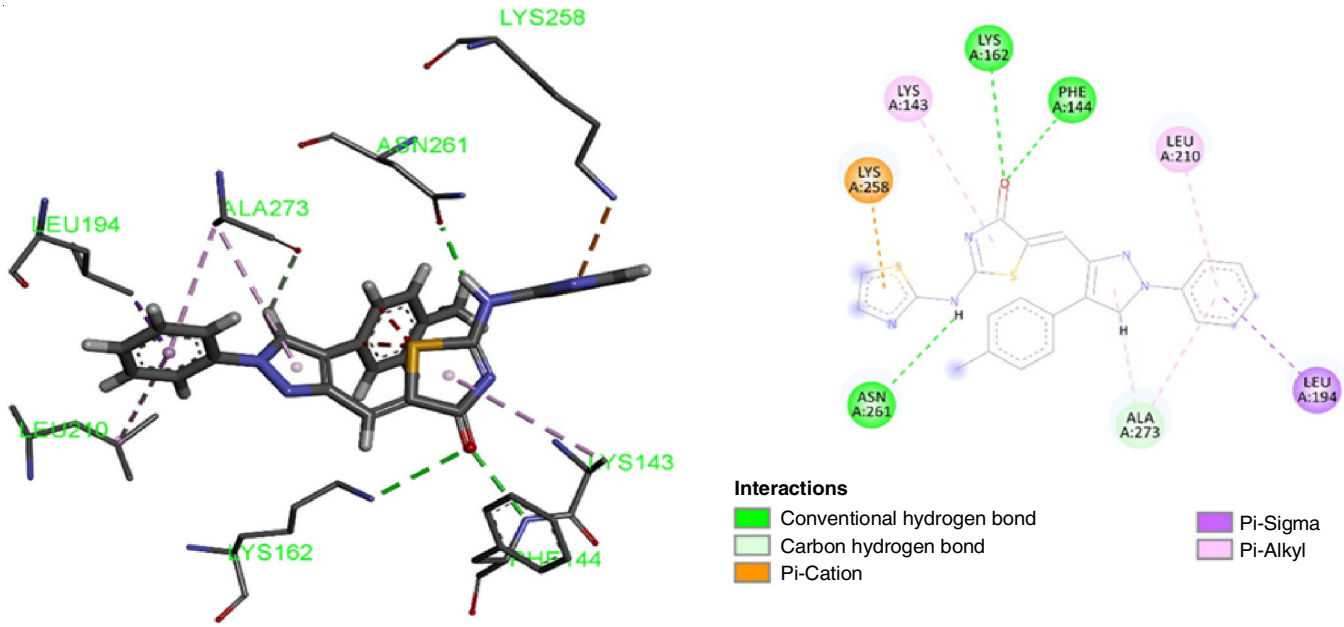


Fig. 1. (a) 3D and (b) 2D images of binding interactions of compound **6e** with Aurora A kinase protein (PDB: 4ZTR)

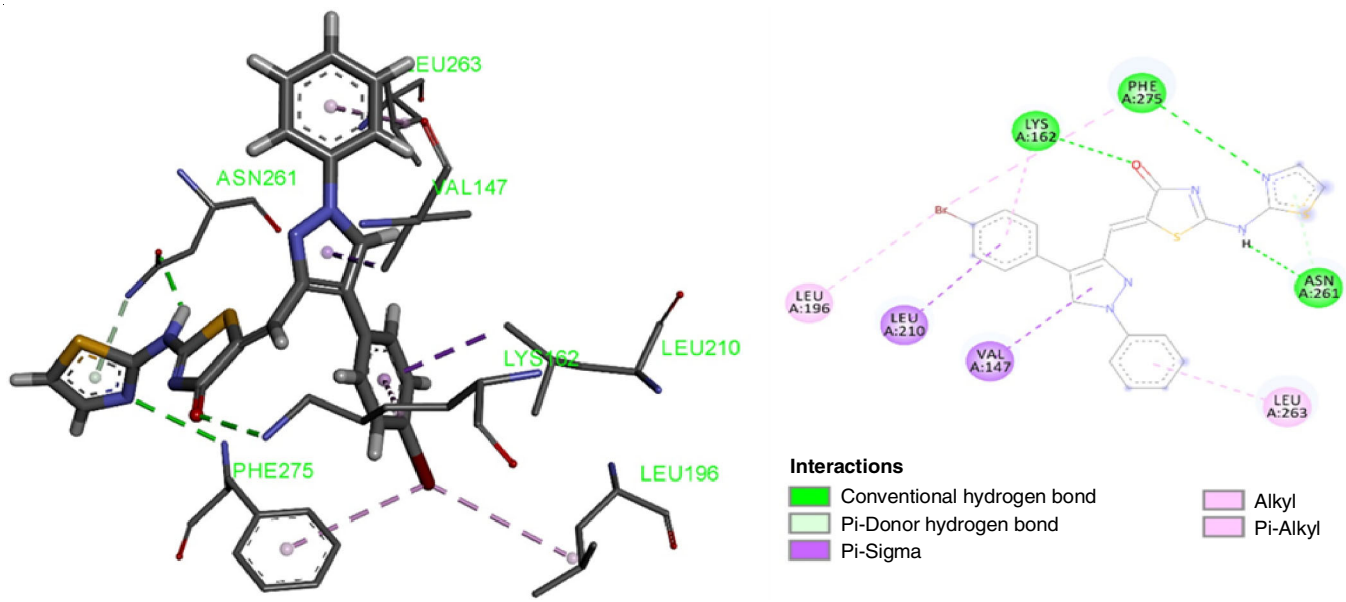


Fig. 2. (a) 3D and (b) 2D images of binding interactions of compound **6c** with Aurora A kinase protein (PDB: 4ZTR)

bond between thiazole ring and ASN261 and two π-sigma bonds between pyrazole ring and 4-bromopheny ring. Moreover, there are 4 π-alkyl interaction between **6c** and protein.

Further, compound **6b** has the similar pattern of interaction as that of compound **6c** and hence it showed good activity (IC_{50} = 18.79 µg/mL) against MDA-MB-231 cell lines. From the docking scores (Table-3), even though compound **6a** has highest interaction score there are only two hydrogen bond interactions which could be a reason for its lower activity and compound **6d** has only one hydrogen bonding interaction and hence, it showed least activity among the series.

Conclusion

The present study identifies a new class of hybrid molecules comprising of thiazolyl-thiazolidinone conjugated pyrazoles as potential anticancer and antimicrobial agents. Newly synthesized compounds exerted very good anticancer activity and in particular compound **6e** having methyl substitution displayed excellent activity against breast cancer MDA-MB-231 and melanoma cancer A375 cell lines with IC_{50} values 13.08 µg/mL and 14.21 µg/mL, respectively. Also, compound **6c** with bromo group exhibited very good activity against breast cancer MDA-MB-231 cell lines with IC_{50} values13.22 µg/mL. The antibacterial activity results revealed compounds **6a** and **6e** as potent agents with excellent MIC values against *P. aeruginosa* and *B. subtilis*, which is superior than the standard drug ampicillin. Molecular docking studies on Aurora A kinase revealed favourable interactions for compound **6e** involving strong hydrogen bonds with key amino acids in the binding pocket of protein. Hence, compound **6e** could be used as lead compound in further developing newer potent compounds.

ACKNOWLEDGEMENTS

The authors are thankful to S.D.M. College (Autonomous), Ujire, India for providing the research facilities to carry out research work. The authors are thankful to Yenepoya Research Centre, Yenepoya University, Deralakatte, India for carrying out the biological studies and IOE Vijnana Bhavana, University of Mysuru for the spectral analyses.

CONFLICT OF INTEREST

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

REFERENCES

1. A. Verma and S.K. Saraf, *Eur. J. Med. Chem.*, **43**, 897 (2008); https://doi.org/10.1016/j.ejmech.2007.07.017
2. C.G. Bonde and N.J. Gaikwad, *Bioorg. Med. Chem.*, **12**, 2151 (2004); https://doi.org/10.1016/j.bmc.2004.02.024
3. S.K. Pandey, U. Yadava, M.L. Sharma, A. Upadhyay, M.P. Gupt, A.R. Dwivedi and A. Khatoon, *Results Chem.*, **5**, 100726 (2023); https://doi.org/10.1016/j.rechem.2022.100726
4. Y.M. Omar, H.H.M. Abdu-Allah and S.G. Abdel-Moty, *Bioorg. Chem.*, **80**, 461 (2018); https://doi.org/10.1016/j.bioorg.2018.06.036
5. M.F. Ansari, D. Idrees, M.I. Hassan, K. Ahmad, F. Avecilla and A. Azam, *Eur. J. Med. Chem.*, **144**, 544 (2018); https://doi.org/10.1016/j.ejmech.2017.12.049
6. S.K. Yusufzai, H. Osman, M.S. Khan, B.M. Abd Razik, M.O. Ezzat, S. Mohamad, O. Sulaiman, J.A. Gansau and T. Parumasivam, *Chem. Cent. J.*, **12**, 69 (2018); https://doi.org/10.1186/s13065-018-0435-0
7. D. Das, P. Sikdar and M. Bairagi, *Eur. J. Med. Chem.*, **109**, 89 (2016); https://doi.org/10.1016/j.ejmech.2015.12.022
8. P. Vicini, A. Geronikaki, K. Anastasia, M. Incerti and F. Zani, *Bioorg. Med. Chem.*, **14**, 3859 (2006); https://doi.org/10.1016/j.bmc.2006.01.043
9. M. Fesatidou, P. Zagaliotis, C. Camoutsis, A. Petrou, P. Eleftheriou, C. Tratrat, M. Haroun, A. Geronikaki, A. Ciric and M. Sokovic, *Bioorg. Med. Chem.*, **26**, 4664 (2018); https://doi.org/10.1016/j.bmc.2018.08.004
10. M.F. Abo-Ashour, W.M. Eldehna, R.F. George, M.M. Abdel-Aziz, M.M. Elaasser, S.M. Abou-Seri and N.M. Abdel Gawad, *Future Med. Chem.*, **10**, 1405 (2018); https://doi.org/10.4155/fmc-2017-0327
11. A.A. Geronikaki, A.A. Lagunin, D.I. Hadjipavlou-Litina, I. Alam, P.T. Eleftheriou, D.A. Filimonov, V.V. Poroikov and A.K. Saxena, *J. Med. Chem.*, **51**, 1601 (2008); https://doi.org/10.1021/jm701496h
12. H. Zhou, S. Wu, S. Zhai, A. Liu, Y. Sun, R. Li, Y. Zhang, S. Ekins, P.W. Swaan, B. Fang, B. Zhang and B. Yan, *J. Med. Chem.*, **51**, 1242 (2008); https://doi.org/10.1021/jm7012024
13. M.F. Khan, M.M. Alam, G. Verma, W. Akhtar, M. Akhter and M. Shaquiquzzaman, *Eur. J. Med. Chem.*, **120**, 170 (2016); https://doi.org/10.1016/j.ejmech.2016.04.077
14. L.L. Xu, C.J. Zheng, L.P. Sun, J. Miao and H.R. Piao, *Eur. J. Med. Chem.*, **48**, 174 (2012); https://doi.org/10.1016/j.ejmech.2011.12.011
15. Y.R. Li, C. Li, J.C. Liu, M. Guo, T.Y. Zhang, L.P. Sun, C.J. Zheng and H.R. Piao, *Bioorg. Med. Chem. Lett.*, **25**, 5052 (2015); https://doi.org/10.1016/j.bmcl.2015.10.028
16. N. Yasser, F.M. Sroor, H.M. El-Shorbagy, S.M. Eissa, H.M. Hassaneen and I.A. Abdelhamid, *RSC Advances*, **14**, 21859 (2024); https://doi.org/10.1039/D4RA03375B
17. O.I. El-Sabbagh, M.M. Baraka, S.M. Ibrahim, C. Pannecouque, G. Andrei, R. Snoeck, J. Balzarini and A.A. Rashad, *Eur. J. Med. Chem.*, **44**, 3746 (2009); https://doi.org/10.1016/j.ejmech.2009.03.038
18. M. Bhat, B. Poojary, B.S. Kalal, P. Gurubasavaraja Swamy, S. Kabilan, V. Kumar, N. Shruthi, S.A. Alias Anand and V.R. Pai, *Future Med. Chem.*, **10**, 1017 (2018); https://doi.org/10.4155/fmc-2017-0191
19. A.S. Kumar, J. Kudva, B.R. Bharath, K. Ananda, R. Sadashiva, S.M. Kumar, B.C. Revanasiddappa, V. Kumar, P.D. Rekha and D. Naral, *New J. Chem.*, **43**, 1597 (2019); https://doi.org/10.1039/C8NJ03671C
20. P. Seboletswe, N. Cele and P. Singh, *ChemMedChem*, **18**, e202200618 (2023); https://doi.org/10.1002/cmdc.202200618
21. D. Ganavi, R. Ramu, V. Kumar, S.M. Patil, R.M. Martiz, P.S. Shirahatti, R. Sathyanarayana, B. Poojary, B.S. Holla, V. Poojary, K.N. Kumari and J.C. Shivachandra, *Arch. Pharm.*, **355**, 2100342 (2022); https://doi.org/10.1002/ardp.202100342
22. V. Kumar, P. Shetty, H.S. Arunodaya, K.S. Chandra, R. Ramu, S.M. Patil, A. Baliga, V.M. Rai, V. Udupi, V. Poojary and B. Poojary, *Chem. Biodivers.*, **19**, e202100532 (2022); https://doi.org/10.1002/cbdv.202100532
23. V. Kumar, V.M. Rai, V. Udupi, N. Shivalingegowda, V.R. Pai, L.N. Krishnappagowda and B. Poojary, *J. Iran. Chem. Soc.*, **19**, 793 (2022); https://doi.org/10.1007/s13738-021-02342-5
24. P. Nefisath, V. Kumar, S.M. Shashiprabha, S.M. Patil, R. Ramu, S. S and J.P. Dasappa, *ChemistrySelect*, **9**, e202303188 (2024); https://doi.org/10.1002/slct.202303188
25. N. Kumar, S. Sreenivasa, A. Prashant, V. Kumar, V. Chandramohan, B.S. Holla, P. Vishwantha and A.K. Yadav, *Chem. Data Coll.*, **32**, 100665 (2021); https://doi.org/10.1016/j.cdc.2021.100665
26. A. Ansari, A. Ali, M. Asifa and Shamsuzzaman, *New J. Chem.*, **41**, 16 (2017); https://doi.org/10.1039/C6NJ03181A
27. V. Kumar, V. K, B.S. Kalal, S.M. Kumar, T.E.S. Kumar, S. Thamotharan, M. Al-Ghorbani and V.R. Pai, *Chem. Data Coll.*, **28**, 100435 (2020); https://doi.org/10.1016/j.cdc.2020.100435