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Synthesis, biological activity evaluation and molecular docking studies of novel coumarin substituted thiazolyl-3-aryl-pyrazole-4-carbaldehydes

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ABSTRACT

A novel series of coumarin substituted thiazolyl-3-aryl-pyrazole-4-carbaldehydes (**4a–o**) were synthesized via an efficient, one-pot multicomponent approach involving 3-(2-bromoacetyl)coumarins (**1a–g**), thiosemicarbazide (**2**) and substituted acetophenones (**3a–c**) utilizing Vilsmeier–Haack reaction condition with good yields. The title compounds structure was elucidated by spectroscopic data (IR, NMR and Mass) and elemental analysis. All the synthesized compounds were screened for their in vitro cytotoxic activity against MCF-7, DU-145 and HeLa cell lines and studied detailed about molecular interaction of probable target protein human microsomal cytochrome CYP450 2A6 using docking simulation. These coumarin derivatives were exhibiting moderate to appreciable cytotoxic activities. The compounds **4m** and **4n** exhibited significant cytotoxic activity with IC_{50} values having 5.75 and 6.25 μM against HeLa cell line. Similarly compound **4n** also exhibiting good anti cancer property and antibacterial activity against DU-145 cell line and Gram negative bacterial strains.

According to World Health Organization (WHO), cancer is the second leading cause of death in humans after cardiovascular disease across the globe. Breast cancer and Prostate cancer are among the most notorious cancer types in women and men, respectively, and a threat for both the developed and developing countries. Numerous cancer therapeutic reports and literature reveal that there is no anticancer agent showing 100% efficacy without side effects. Therefore, across the globe, there is a huge thrust among the researchers to develop new chemotherapeutic drugs which would have maximum efficacy with specific mechanism of action to overcome the difficulties associated with the present clinically used drugs.

Multicomponent reactions (MCRs) have emerged as an efficient and powerful tool in modern synthetic chemistry for the generation of highly complex molecules in a single operation without isolation of intermediate.[1–3] MCRs construct an effective synthetic route in minimal time with maximum selectivity, high atom economy and high purity with excellent yields. Similarly, Vilsmeier–Haack reaction has been used for the construction of many heterocyclic compounds.[4]

Coumarin is an important scaffold present in various natural and synthetic compounds,[5,6] and has versatile pharmacological properties that include antifungal,[7] antioxidant,[8–10] anticoagulant,[11] antiviral,[12] antiproliferative,[13] antialzheimer,[14] anticancer[15–17] and anti-HIV[18,19] activities. In addition to the therapeutic applications, these are widely used as food additives, in cosmetics, perfumery, as optical brighteners, and in dispersed fluorescence and lasers dyes.[20]

Among the heterocyclic compounds, thiazole and pyrazole derivatives are one of the most important five member heterocyclic molecules. Thiazole is the most prevalent motif with attractive pharmacological activities. Several COX-2 inhibitors have pyrazoles as a common chemical unit. In recent years these five member molecules have attracted the attention of researchers in the field of medicine and agriculture. Thiazole and pyrazole derivatives were also found to take part in several biological activities.[21–27] The structures of Warfarin, Meloxicam, and Celecoxib and their pharmacological activities are given in Figure 1.

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Figure 1. Biologically active coumarin, thiazole and pyrazole derivatives.

In view of the importance of coumarins, thiazoles and pyrazoles in the field of medicinal chemistry and also to explore the scope of these motifs, we have focused on the design of a novel structural entity that comprises these three structural moieties along with a potential functional group into a single molecular framework. In the current scenario the use of computational approach is very important in the field of computer aided drug designing to elucidate the molecular level interaction and active site residues property in relation to activity. Therefore, we elicit synthesis, molecular docking and biological evaluation of coumarin substituted thiazolyl-3-aryl-pyrazole-4-carbaldehydes.

3-(2-Bromoacetyl)coumarins (**1a–g**) were prepared by bromination of 3-acetylcoumarins in dry chloroform.[28] The title compounds (**4a–o**) were obtained by reaction of various 3-(2-bromoacetyl)coumarins (**1a–g**) with thiosemicarbazide (**2**) and acetophenones (**3a–c**) under Vilsmeier–Haack reaction conditions with good yields. The general schematic representation is outlined in Scheme 1.

To know the feasibility of the reaction, was carried out the reaction in one-pot multicomponent approach of 3-(2-bromoacetyl)-2*H*-chromen-2-one (**1a**), thiosemicarbazide (**2**) and acetophenone (**3a**) in presence of dimethylformamide. In this step substituted 3-(2-bromoacetyl)coumarins react with thiosemicarbazide and form the Hantzsch thiazole product and further reaction with substituted acetophenones leads to the formation of an intermediate 3-(2-hydrazino-4-thiazolyl)coumarino)phenyl methyl methane. Without isolation of this intermediate, the compound is subjected to Vilsmeier–Haack formylation reaction condition to generate the target product. This method is simple and found to be easier to synthesize the thiazolyl pyrazole carbaldehyde derivatives in shorter reaction time with good yields (Table 1).

During these reactions, thiazole and pyrazole rings are formed along with a potential functional group (—CHO) on pyrazole ring.

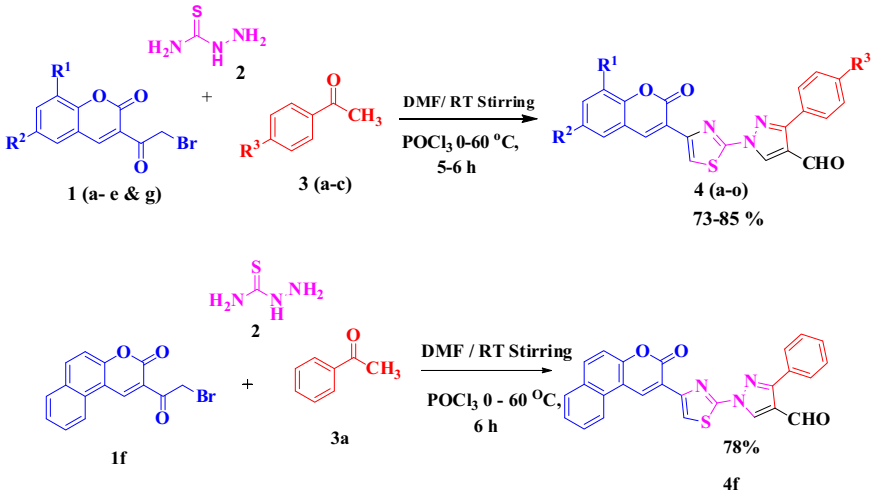
Table 1
A facile one-pot synthesis of coumarin substituted thiazolyl-3-aryl-pyrazole-4-carbaldehydes (**4a–o**)

Product	R^1	R^2	R^3	Time	Yield[a] (%)
4a	H	H	H	5.0	84
4b	H	Cl	H	5.5	80
4c	Cl	Cl	H	6.0	75
4d	H	Br	H	5.5	80
4e	Br	Br	H	6.0	85
4g	H	Cl	Cl	6.0	77
4h	Cl	Cl	Cl	5.5	73
4i	H	Br	Cl	5.0	83
4j	OCH_3	H	Cl	6.0	82
4k	H	H	CH_3	5.0	77
4l	H	Br	CH_3	6.0	82
4m	Cl	Cl	CH_3	5.5	81
4n	Br	Br	CH_3	5.0	85
4o	OCH_3	H	CH_3	6.0	82

Reaction condition: 3-(2-bromoacetyl)coumarin (**1a–g**, 1 mmol), thiosemicarbazide (**2**, 1 mmol), acetophenone (**3a–c**, 1 mmol), DMF (5 mL) and $POCl_3$ (5 mmol), 0–60 °C, 5–6 h.
[a] Isolated yields.

The formylation is highly regioselective, it took place only on pyrazole ring and not on thiazole ring. It is a novel observation, and as far as our knowledge is concerned, no such report so far is available in one-pot multicomponent manner to synthesize the coumarinyl thioazolo-pyrazole-4-carbaldehydes in the literature. This may trigger a new and interesting area of research in chemistry with reference to Vilsmeier–Haack formylation reaction.

All the synthesized compounds (**4a–o**) were fully confirmed on the basis of their analytical and spectral studies. IR spectra of the compounds, in general has shown strong bands in the range of 1547–1637 cm^{-1} (C=N), 1617–1685 cm^{-1} (C=O of aldehyde) and 1713–1737 cm^{-1} (C=O of lactone). The ^1H NMR of the compounds



Scheme 1. Synthesis of coumarin substituted thiazolyl-3-aryl-pyrazole-4-carbaldehydes **4**(**a–o**).

has shown characteristic peaks at 8.34–8.80, 8.85–9.64, 9.30–9.78 and 10.04–10.09 ppm corresponds to C4-H of coumarin, pyrazole-H, thiazole-H and aldehyde protons, respectively. The ^{13}C NMR of the synthesized compounds has shown peaks at 157.9–159.1, 158.8–163.5 and 184.7–185 ppm corresponding to imine, lactone and aldehyde carbons, respectively. Similarly, the molecular ion peak from the mass spectra and elemental analyses data are further evidence for the formation of the products. The spectral data of some of the compounds are given below.

Compound **4b**: mp: 262–264 °C; FT-IR (KBr, ν_{max}, cm^{-1}): 1636 (—C=N), 1683 (—CHO), 1726 (—C=O); ^1H NMR (400 MHz, DMSO-d_6, ppm): δ 7.54 (t, 3H, J = 6.0 Hz, Ar-H), 7.70 (d, 1H, J = 2.0 Hz, Ar-H), 7.93 (t, 1H, J = 3.6 Hz, Ar-H), 8.01 (d, 1H, J = 2.4 Hz, Ar-H), 8.22 (s, 2H, Ar-H), 8.39 (s, 1H, C4-H of coumarin), 8.92 (s, 1H, C4-H of pyrazole), 9.38 (s, 1H, C5-H of thiazole), 10.07 (s, 1H, —CHO); ^{13}C NMR (100 MHz, DMSO-d_6, ppm): δ 117.95, 118.96, 120.35, 121.32, 123.13, 127.64, 128.91, 129.00, 129.95, 130.17, 130.29, 131.55, 131.90, 138.47, 145.30, 151.47, 158.94, 163.21, 184.78; HRMS (ESI) m/z calculated for $C_{22}H_{12}ClN_3O_3S$ $(M+Na)^+$ 456.0186, found 456.0199. Anal. Calcd for $C_{22}H_{12}ClN_3O_3S$: C, 60.90; H, 2.79; N, 9.69; S, 7.39. Found: C, 60.84; H, 2.84; N, 9.63; S, 7.42.

Compound **4c**: mp: 270–272 °C; FT-IR (KBr, ν_{max}, cm^{-1}): 1637 (—C=N), 1685 (—CHO), 1727 (—C=O); ^1H NMR (400 MHz, DMSO-d_6, ppm): δ 7.55 (d, 3H, J = 3.2 Hz, Ar-H), 7.71 (d, 1H, J = 8.4 Hz, Ar-H), 7.93 (d, 1H, J = 3.2 Hz, Ar-H), 8.00 (s, 1H, Ar-H), 8.18 (s, 1H, Ar-H), 8.39 (s, 1H, C4-H of coumarin), 8.92 (s, 1H, C4-H of pyrazole), 9.38 (s, 1H, C5-H of thiazole), 10.07 (s, 1H, —CHO); ^{13}C NMR (100 MHz, DMSO-d_6, ppm): 121.97, 123.02, 126.37, 128.54, 128.94, 129.02, 129.93, 130.33, 130.44, 131.37, 133.49, 135.60, 146.08, 152.92, 155.74, 159.18, 159.58, 185.09; MS (ESI) m/z: 468 $[M+2]^+$; Anal. Calcd for $C_{22}H_{11}Cl_2N_3O_3S$: C, 56.42; H, 2.37; N, 8.97; S, 6.85. Found: C, 56.46; H, 2.34; N, 8.99; S, 6.81.

Compound **4f**: mp: 257–259 °C; FT-IR (KBr, ν_{max}, cm^{-1}): 1635 (—C=N), 1683 (—CHO), 1725 (—C=O); ^1H NMR (400 MHz, DMSO-d_6, ppm): δ 7.55 (t, 2H, J = 3.6 Hz, Ar-H), 7.65–7.70 (m, 2H, Ar-H), 7.85 (t, 1H, J = 7.6 Hz, Ar-H), 7.94 (t, 2H, J = 3.6 Hz, Ar-H), 8.10 (d, 1H, J = 8.4 Hz, Ar-H), 8.25 (d, 1H, J = 9.2 Hz, Ar-H), 8.40 (s, 2H, Ar-H), 8.80 (s, 1H, C4-H of coumarin), 9.64 (s, 1H, C4-H of pyrazole), 9.78 (s, 1H, C5-H of thiazole), 10.09 (s, 1H, —CHO); ^{13}C NMR (100 MHz, DMSO-d_6, ppm): δ 113.20, 116.45, 118.47, 118.88, 122.41, 122.82, 126.31, 128.50, 128.63, 128.75, 128.97, 129.72, 130.05, 130.21, 133.65, 135.04, 135.71, 145.22, 152.45, 153.96, 158.63, 158.87, 184.87; HRMS (ESI) m/z calculated for $C_{26}H_{15}N_3O_3S$ $(M+Na)^+$ 472.0732, found 472.0784; Anal. Calcd for $C_{26}H_{15}N_3O_3S$: C, 69.48; H, 3.36; N, 9.35; S, 7.13. Found: C, 69.51; H, 3.32; N, 9.31; S, 7.17.

To examine the structure activity relationship (SAR) of the newly synthesized coumarin substituted thiazolyl pyrazole-carbaldehyde compounds having different substituents on coumarin as well as on phenyl ring, were evaluated for their cytotoxic activity against three human cancer cell lines (DU-145, MCF-7 and HeLa) by adopting the MTT assay method.[29,30] Doxorubicin was used as a standard reference drug. Initially all the fifteen coumarin derivatives were taken to evaluate the single dose (10 μM) screening. From this screening some of the compounds like **4g**, **4i**, **4m** and **4n** showed good inhibitory effect against HeLa cell line. Subsequently all the synthesized compounds were screened at three different concentrations (2.5, 5, 100 μM) to evaluate their cytotoxicity. All the synthesized compounds showed moderate to good anticancer activity with IC_{50} values ranging from 5.75 μM to 100 μM. The compound **4m** showed excellent cytotoxic activity against HeLa cell line, having IC_{50} value 5.75 μM. Similarly compounds **4g**, **4h**, **4i** and **4n** also exhibited good cytotoxic activity against HeLa cell line. Additionally the results obtained from MTT assay against DU-145 cell line, compounds **4f** and **4n** exhibited moderate cytotoxic activity. On the other hand, compounds **4**(**a**–**o**) exhibited poor activities against MCF-7 cell lines. From these anticancer biological studies, we were able to identify potent molecules which exhibited good anticancer activity in human cancer cell lines (Table 2).

Apoptosis is an important process of cell death of undesirable cells during development or homeostasis in multicellular organisms and during apoptosis, chromatin condensation takes place. To see whether coumarin derivatives (**4m** and **4n**) induced cytotoxicity occurs through apoptosis, HeLa cervical cancer cells were treated with 10 μM concentration of these compounds for a period of 24 h. Hoechst 33258 staining was used to visualize nuclear condensation. It was found that both these compounds caused a significant nuclear condensation as shown in Figure 2.

To evaluate the biological activities further, the title compounds were screened against various Gram positive (*Bacillus subtilis* [ATCC 6633], *Bacillus cereus* [ATCC 14579] and Methicillin-resistant *Staphylococcus aureus* [NCTC 13616]) and Gram negative bacterial (*Escherichia coli* [ATCC 8739], *Klebsiella pneumoniae* [ATCC 43816] and *Proteus vulgaris* [ATCC 13315]) strains by using agar well diffusion method.[31] Gentamycin sulfate was used as standard drug. The anti bacterial activity of the synthesized compounds exhibited significant zones of inhibition against tested Gram positive and Gram negative strains at 10, 20, 100, 200 and 300 μg/mL concentrations. The zone of inhibition (mm) of synthesized molecules at 100 μg/mL concentration was shown in Figure 3.

However, some of the strains exhibited resistance to tested compounds. Among, the tested compounds, compound **4n** exhibited highest activity against Gram negative *E. coli*, *K. pneumoniae* and *P. vulgaris* 22, 24 and 20 mm zone of inhibition at 100 μg/mL, respectively. On the other hand, compound **4m** also exhibited significant antibacterial activity with zone of inhibition 18, 20 and 18 mm against *E. coli*, *K. pneumoniae* and *P. vulgaris* at 100 μg/mL concentration. From the above results it is clear that the compounds **4n**, **4m** and **4h** are found to exhibit greater inhibition efficiency against various strains. Apart from these, compounds **4c**, **4d**, **4e**, **4g**, **4i**, **4j** and **4l** also showed significant to moderate antibacterial activity. On the other hand compounds **4a**, **4b**, **4k** and **4o** failed to exhibit activity against Methicillin-resistant *Staphylococcus aureus* [MRSA].

In order to evaluate the anti bacterial properties, apart from the zone of inhibition method we have calculated the minimum inhibitory concentration [MIC] and minimum bactericidal concentration [MBC] values for the fifteen synthesized molecules against various Gram-positive and Gram-negative bacterial strains by

Table 2
IC_{50} values for compounds **4**(**a**–**o**) against human cancer cell lines

Compound	IC_{50} values (μM)		
	DU-145	MCF-7	HeLa
4a	NA	NA	NA
4b	NA	NA	**12.82**
4c	41.05	76.33	**13.75**
4d	35.01	18.16	**11.02**
4e	27.97	22.23	**13.69**
4f	11.91	39.46	**13.11**
4g	**14.86**	18.67	**9.51**
4h	30.90	21.74	**10.29**
4i	22.32	85.03	**9.46**
4j	38.18	71.68	41.89
4k	50.23	69.45	37.36
4l	20.86	35.17	14.13
4m	**14.71**	**14.56**	**5.75**
4n	**10.81**	**24.52**	**6.25**
4o	31.42	42.57	28.19
Doxorubicin	2.49	3.18	3.92

NA = not active (IC_{50} values >100 μM).

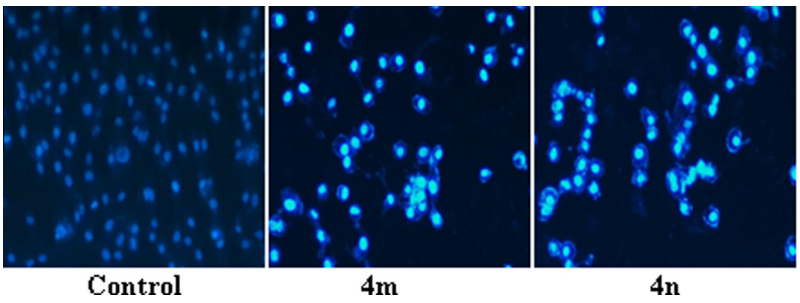


Figure 2. Coumarin derivatives cause apoptosis in HeLa cells. Cells were treated with **4m** and **4n** at 10 μM concentration for 24 h and washed with PBS, incubated with Hoechst-33258 stain (4 mg/mL) for 20 min to measure chromatin condensation. Images were taken using fluorescence microscopy equipped with DAPI filter.

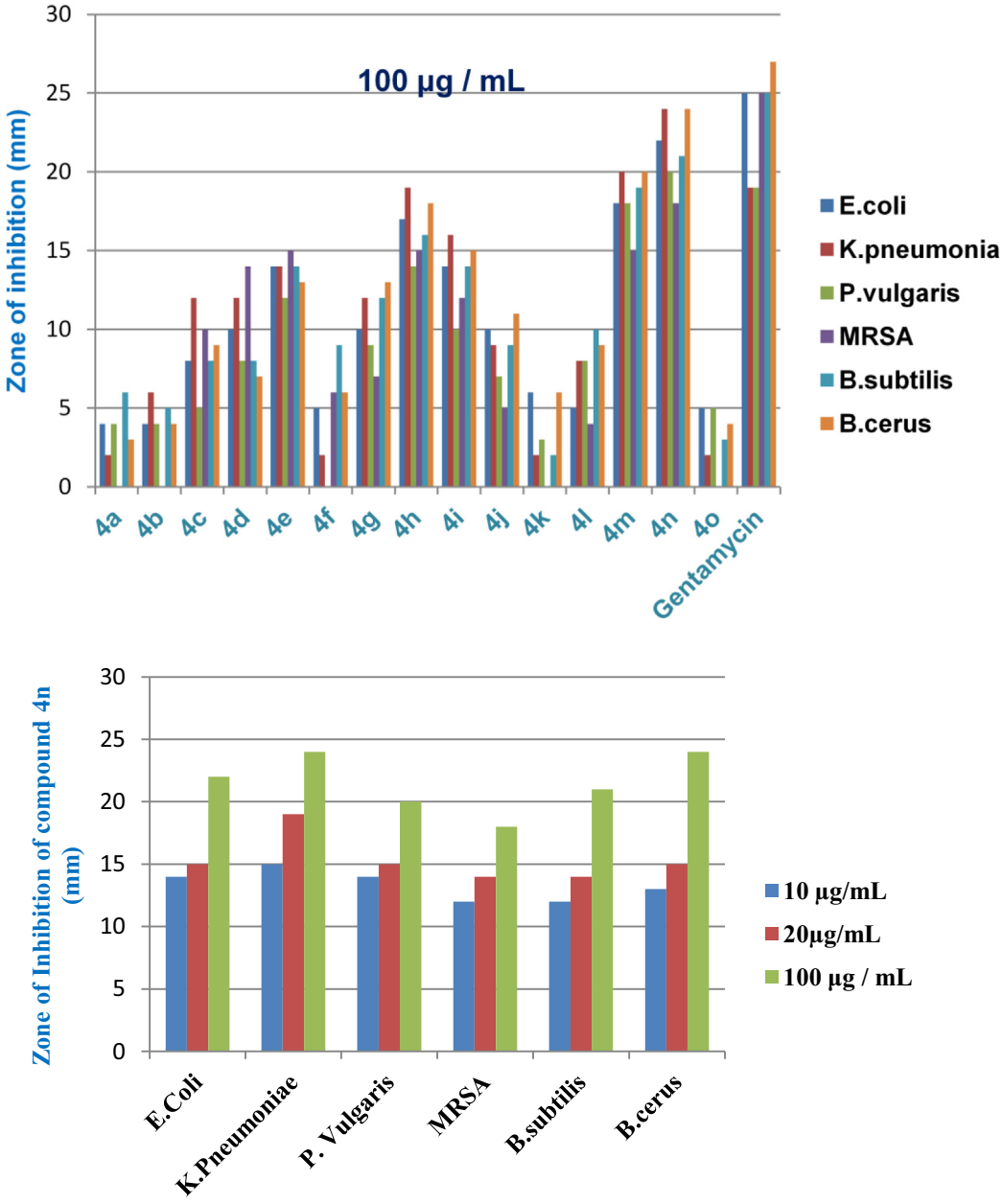


Figure 3. Antibacterial activity (zone of inhibition in mm) of compounds **4a–o**.

using standard MIC and MBC protocol.[32] Gentamycin sulfate and ampicillin were used as standard reference drugs. The MIC values were calculated after 24 h of incubation. Compound **4n** produced significant MIC values of 86.5, 79.1 and 72.8 μg/mL concentrations against *E. coli*, *K. pneumoniae* and *B. cereus*, respectively. The corresponding MBC values of 130, 115 and 110 μg/mL against *E. coli*, *K.*

pneumoniae and *B. cereus*. Similarly the compound **4m** also exhibited moderate MIC value of 98.2 μg/mL concentration against *B. subtilis*. Whereas the remaining compounds exhibited moderate to poor antimicrobial properties in the tested concentration range. The measured MIC and MBC values are shown in Table 3.

According to the data obtained in the present study, the compounds exhibited antibacterial activity in a concentration dependent manner. It has been clear that, the compounds are showing good activity against Gram negative species compared with that from Gram positive species. There are several reasons that can be speculated for immense activity of the compounds against Gram negative strains.

In order to explain the structure activity relationship of antimicrobial and anti cancerous properties of the newly synthesized coumarin substituted thiazolyl pyrazole-carbaldehydes, these compounds were examined by changing the functional groups such as Cl, Br, CH_3, —Ph and —OCH_3 at R^1 R^2 and R^3 positions and these were divided into two groups: group one was un-substituted coumarins and un-substituted phenyl ring on pyrazole nucleus and group two was substituted coumarins and substituted phenyl ring on pyrazole nucleus. The SAR studies clearly indicated the presence of substituents on both coumarin as well as on the benzene ring which are crucial for anticancer and antibacterial properties of the tested compounds. From the MIC values Br substitution at R^1, R^2 and —CH_3 substitution at R^3 position exhibit good antibacterial property. Similarly the presence of chlorine substituent at R^1, R^2 and —CH_3 substituent at R^3 position also showed good cytotoxic activity and antibacterial activity. On the other hand unsubstituted coumarin ring and unsubstituted phenyl ring on thiazolyl pyrazole carbaldehydes did not show any biological activities. From the above results, the presence of two halogen atoms (Br or Cl) on coumarin nucleus and methyl substituent on phenyl ring may be said to drastically enhance the properties.

For understanding the biological properties of the synthesized molecules, the lipophilicity values were determined through experimental procedure. Any molecule which exhibits pharmacological properties should have affinity for fat and high lipid solubility. Lipophilicity is a physicochemical property which can explain the drug like nature of the molecule and describe the partition equilibrium of solute molecule between aqueous medium and immiscible organic solvent. Experimentally, lipophilicity[33] of the biologically active compounds were measured by shake—flask method using *n*-octanol—phosphate buffer (50 mM, pH 7.4) biphasic system. Partition coefficient (*P*) values were calculated for all the active molecules and log*P* values (which is considered as lipophilicity) were measured. All the active molecule were found to show good lipophilicity values in the range of 3.0–3.6 (Table 4).

The docking simulation study was carried out using AutoDock 4[34] to validate the in vitro result and elucidate the importance of different types of interactions to inhibit the function of probable target human microsomal cytochrome P450 2A6 (1z11.pdb) enzyme.[35,36] The docking simulation results shows that the binding affinity of all the novel fifteen coumarin derivatives have stronger binding energies than that of the Methoxsalen (co-crystallized ligand of PDB molecule) and coumarin itself. The order of binding affinity of docked coumarin derivatives against the receptor is **4f** > **4l** > **4n** > **4o** > **4m** > **4g** > **4h** > **4j** > **4e** > **4d** > **4k** > **4a** > **4b**> and **4c** with the range of binding energy being −10 to −12.61 k cal/mol (Table 5).

The in silico study results show that the catalytic unit of Microsomal cytochrome P450 2A6 protein (1z11.pdb) active site residues Thr 305, Gln 360, Val 365, Pro 431, Phe 432, Cys 439 and Phe 440 are key interacting amino acids and common among 15, 15, 14, 15, 15, 16 and 14 ligand molecules. The number of hydrogen bonds vary from zero to three with the receptor molecule that show other interactions like electrophilic and hydrophobic which are responsible for forming strong bonds between ligands and receptor (1z11.pdb) to inhibit the function of enzyme and are responsible for the cytotoxic effect.

The obtained binding energies for the coumarin derivatives show similarity with the cytotoxic activity of among all the tested derivatives except **4f**, the good inhibitory potential as seen for the **4g**, **4i**, **4m** and **4n** demonstrated lowest binding energies (−11.61, −11.72, −11.72 and −11.95 kcal/mol) than the coumarin and methoxsalen (−6.37 and −7.77 kcal/mol). Particularly, the lowest cytotoxic activity of compound **4n** showed the lowest binding energy of −11.95 kcal/mol and other active compounds interestingly have nearly equal binding energies. The docking poses of these compounds revealed that they could also occupy the same active site as shown in Figure 4 and able to form strong hydrophobic and electrostatic interaction with the active site residues other than hydrogen bonding (Fig. 5). Among the most key interacting residues, Ile 366 and Cys 439 actively participate in the formation of hydrogen bonding with the compounds **4g** and **4g**, **4m** and **4n**.

Table 3
Minimum inhibitory concentration (μg/mL) and minimum bactericidal concentration (μg/mL) values of novel coumarinyl thiazolyl pyrazole carbaldehyde derivatives (**4a**–**o**)

Compound	Gram negative						Gram positive					
	E. coli		*K. pneumoniae*		*P. vulgaris*		MRSA		*B. subtilis*		*B. cereus*	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
4a	135	270	150	225	133	200	150	300	132	260	145	290
4b	134.6	265	129	190	130	260	150	300	129.5	190	138.6	275
4c	119.6	240	118	175	131.5	260	115	175	123	250	117	175
4d	118.1	240	116.8	175	124	250	116	175	125	250	129	260
4e	115.6	170	119	240	120	240	116.8	175	118	240	119	240
4f	132.8	260	150	225	150	300	135	270	122.8	240	138.9	275
4g	126.3	250	117.4	240	130.7	260	134	265	112.2	165	110.8	165
4h	110.9	220	105	155	113	225	112.5	225	110.7	165	112	220
4i	117.5	230	115	172	127.5	250	122.9	185	119.7	175	115	170
4j	125.3	185	129	260	132.3	260	139	275	132.6	260	126	250
4k	132.5	260	150	300	150	300	150	300	150	300	135	270
4l	134.6	270	124	250	121.8	240	138.1	275	125	250	126	250
4m	101.3	150	100.9	150	105	210	115.6	230	98.2	145	100.1	150
4n	86.5	130	79.1	115	100.7	150	105.9	175	92.4	150	72.8	110
4o	132.7	260	150	300	142	285	150	300	150	300	136	275
Gentamycin	2	8	4	16	4	8	22	50	45	80	10	22
Ampicillin	10	18	NT	NT	NT	NT	4	6	5	10	4	8

NT = not tested.

Table 4
Lipophilicity values of novel coumarinyl thiazolyl pyrazole carbaldehyde derivatives by shake flask method

Compound	Wave length (nm)	Concd of stock added (mM)	Concd obtained (mM)	Absorbance	*P*	Log*P*
4d	325	0.2857	0.01025	0.1251	2015.48	3.304
4e	330	0.2857	0.01563	0.1949	1295.92	3.112
4f	393	0.2857	0.00608	0.0913	3449.25	**3.537**
4g	335	0.2857	0.01754	0.2171	1146.63	3.059
4h	340	0.2857	0.01981	0.2483	1006.65	3.002
4i	329	0.2857	0.01723	0.2144	1168.61	3.067
4m	340	0.2857	0.00525	0.0773	4000.21	**3.602**
4n	335	0.2857	0.00955	0.1183	2168.71	**3.336**

Table 5
Docking results of novel coumarin derivatives with the Human Microsomal cytochrome P450 CYP2A6 [1z11.pdb] protein

Ligand name	Binding energy (kcal/mol)	Inhibition constant (nM)	No. of H bonds
Coumarin	−6.37	–	–
4a	−10.76	12.96	2
4b	−10.75	2.58	3
4c	−10.61	3.07	1
4d	−11.22	6.02	0
4e	−11.55	3.41	1
4f	−12.61	5.70×10^{-2}	1
4g	−11.61	16.62	1
4h	−11.6	2.7	2
4i	−11.72	13.2	1
4j	−11.58	3.27	2
4k	−10.86	10.97	1
4l	−12.02	1.54	1
4m	−11.72	2.57	1
4n	−11.95	1.73	1
4o	−11.89	1.93	2

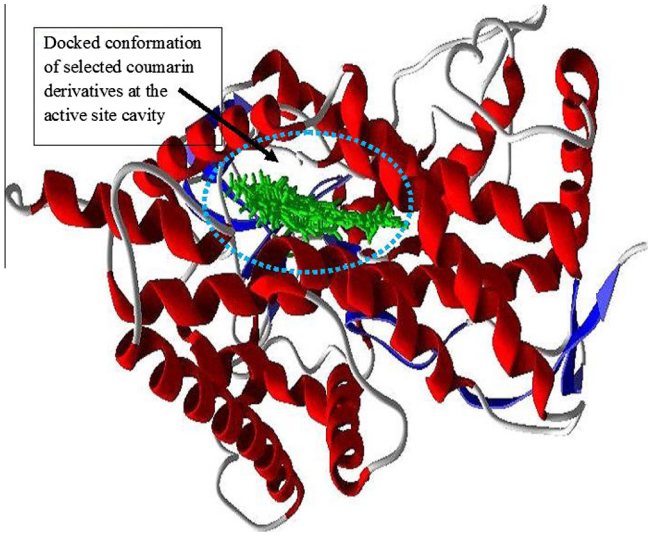


Figure 4. Secondary structure (cartoon) representation of target protein Human Microsomal cytochrome P450 2A6 together with the docked conformation of novel coumarin derivatives at the common active site.

In summary, we have developed a facile method for the synthesis of substituted thiazolyl pyrazole carbaldehyde derivatives under Vilsmeier–Haack reaction conditions via multicomponent approach and evaluated molecular interactions as well as cytotoxic activities against human cancer cell lines and antibacterial activity

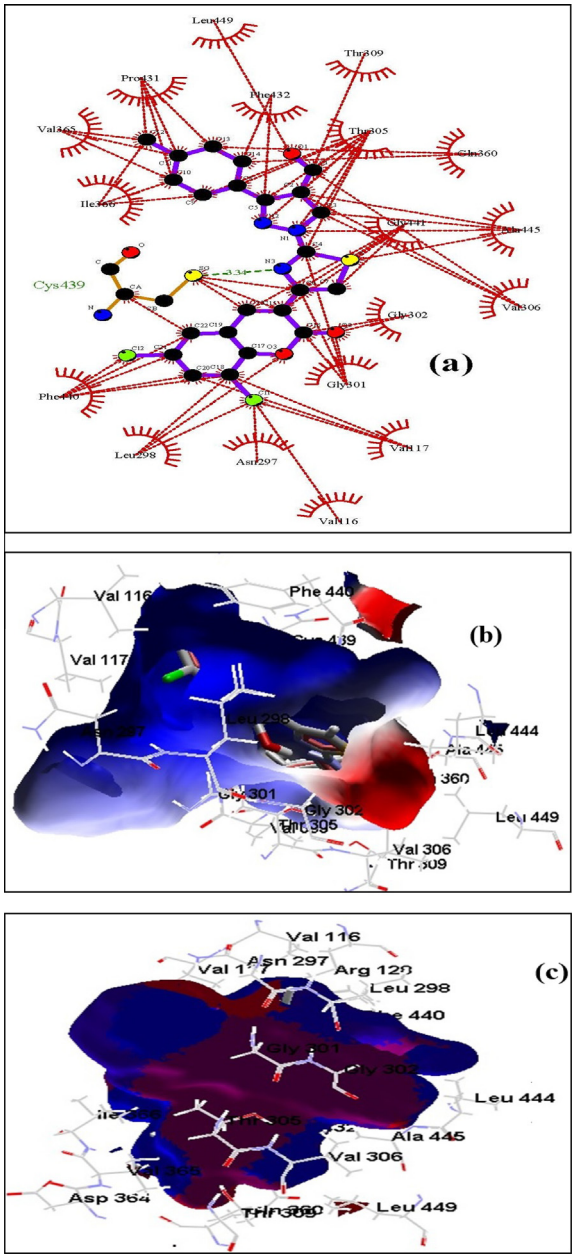


Figure 5. (a) Docked conformation of compound **4m** with hydrogen bonding view in form of 2-D using LigPlus[37] and (b) hydrophilic and (c) hydrophobic interactions using Molegro Molecular Viewer[38] at the active site cavity of receptor protein (1z11.pdb) (hydrogen bonds as green color dashed lines between the atoms involved), and hydrophobic contacts as an arc with spokes radiating towards the ligand atoms.

against Gram positive and Gram negative bacteria. Among the tested compounds, compounds **4m**, **4n** exhibited significant cytotoxic activity against HeLa cell line. Due to the presence of pharmacologically active moieties like coumarin, thiazole and pyrazole along with a potential functional group like aldehyde in their structures, they seem to be suitable candidates for further chemical modification and may be biologically active and useful as ligands in coordination chemistry.

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Supplementary data

Supplementary data (experimental procedures and final compounds characterization data) associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bmcl.2015.10.042.

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