Monatsh Chem (2018) 149:595–609
https://doi.org/10.1007/s00706-017-2079-5

ORIGINAL PAPER

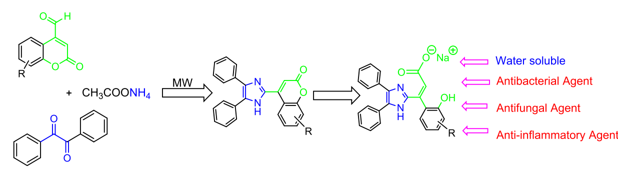
Design and synthesis of coumarin–imidazole hybrid and phenyl-imidazoloacrylates as potent antimicrobial and antiinflammatory agents

Megharaja Holiyachi[1] · S. Samundeeswari[1] · Bahubali M. Chougala[1] · Nirmala S. Naik[1] · Jyoti Madar[1] · Lokesh A. Shastri[1] · Shrinivas D. Joshi[2] · Sheshagiri R. Dixit[2] · Suneel Dodamani[3] · Sunil Jalalpure[3,4] · Vinay A. Sunagar[5]

Received: 9 August 2017 / Accepted: 16 October 2017 / Published online: 18 January 2018

Abstract An improved one pot, multi-component synthesis of tri- and tetrasubstituted coumarin–imidazole hybrid has been synthesized at C4 position in good to excellent yield. The reaction was performed under various catalysts and optimization condition results obtained are satisfactory. Wherein, trisubstituted coumarin–imidazole hybrid compounds were converted into phenyl-imidazole acrylates. Further, all the newly synthesized compounds were screened for their antimicrobial activity against Gram-positive *Bacillus flexus* and Gram-negative *Pseudomonas* spp. bacterial strains and two strains of fungi studies having *Scopulariopsis* spp. and *Aspergillus terreus* organisms. Similarly, antiinflammatory activity of all the compounds was screened against MMP-2 and MMP-9. Both antimicrobial and antiinflammatory results are excellent, among all compounds, sodium acrylate compounds are quite promising against microbial strains and matrix metalloproteins (MMPs). All the isolated compounds were characterized by IR, NMR, and mass spectral analysis.

Graphical abstract



Keywords Enzymes · Multi-component reaction · Gelatinase · Histidine · Matrix metalloproteins

Electronic supplementary material The online version of this article (doi:https://doi.org/10.1007/s00706-017-2079-5) contains supplementary material, which is available to authorized users.

✉ Lokesh A. Shastri
drlashastri@kud.ac.in

[1] Department of Chemistry, Karnatak University, Dharwad, Karnataka 580003, India

[2] Novel Drug Design and Discovery Laboratory, Department of Pharmaceutical Chemistry, S.E.T's College of Pharmacy, Dharwad, Karnataka 580002, India

[3] Dr. Prabhakar Kore Basic Science Research Center, KLE University, Belgaum, Karnataka 590010, India

[4] KLE University's College of Pharmacy, Nehru Nagar, Belgaum, Karnataka 590010, India

[5] Department of Chemistry, G.S.S. College, Belgaum, Karnataka 590006, India

Introduction

There are increasing antibiotic resistance properties among pathogens and on the other hand decreasing rate of new novel drug discovery is common drawback in medicine. Thus, multidrug resistance can cause infections as they are no longer response to most of the usual antibiotics [1, 2]. Moreover, imidazole is the integral part of the some naturally occurring molecules and is known for active scaffold in biomolecules, such as histidine, histamine, component of DNA base and Vitamin B_{12} imidazole derivatives exhibiting broad range of biological activities, such as antimicrobial [3–5], anticancer [6], antiinflammatory [7, 8], and analgesic activities [9].

Coumarin is also one of such natural product which has been isolated from a variety of plant sources to assess their potential therapeutic uses [10]. Coumarin derivatives comprise a vast array of biological activities and have been used in traditional medicine since long time. Thus, the biological investigation of coumarin derivatives shows the engrossment of several pathways by which coumarin act as potential anticancer [11, 12], anti-HIV [13, 14], anticoagulant [15], antimicrobial [16], antioxidant [17], and antiinflammatory agents [18]. The non-steroidal coumarin molecules (Fig. 1a) were synthesized and used for the treatment of breast cancer [19]. The synthetic analogous of bleomycin: histidine–pyridine (Fig. 1b) structure is potent anticancer agent [20].

In microorganism regulatory system, histidine protein kinase (HPKs) plays a key role in prokaryotic transduction [21]. Hence, HPKs is the target for most of the antibacterial and antifungal agents [22, 23]. The antimicrobial peptides (AMPs) are cationic amphiphils which are the first defense to protect organism from microbial infections [24], via amphiphilic structural conformations that is cationic and the hydrophobic groups segregate into distinct regions.

Therefore, the promising therapeutic perspectives of both the heterocycles inspired us towards the development of small molecules. Herein we employed distinct and optimized molecular hybridization strategy for the synthesis of a new imidazole nucleus directly linked at C4 position of coumarin. Further, hybrid molecules were transferred into amphiphilic molecules to mimic the structure, function and mode of action such as AMPs, which can display broad spectrum of antimicrobial agent. Due to the structural similarity with histidine, antimicrobial ability of these compounds manifested their competition with proteins inside the bacterial cell wall.

Results and discussion

Chemistry

In the literature, there are many synthetic methods reported for the synthesis of imidazoles, among these microwave-assisted method has driven much attention [25, 26]. The C4 position of coumarin ring bearing five membered imidazole derivatives are not available in the literature. The author has designed and synthesized C4-coumarin which is directly linked with C2 position of imidazole. The synthetic strategy followed for the synthesis of coumarin–imidazole hybrid structures are outlined in Scheme 1. The reaction of 4-formylcoumarin (**1**) [27] and 1, 2-diketone **2** with ammonium acetate in the presence of acid catalysts is to furnish the desired coumarin–imidazole hybrid molecules **3a–3g** in good yield (> 65%) under conventional method. For the purpose of scope and limitations of this reaction, we studied the reaction in different solvents and catalysts to optimize the condition. The optimized results are listed in Table 1, from which it is evident that acetic acid alone as a catalyst and solvent afforded excellent yield comparatively with other combinations of solvents and catalysts. Further, no changes in reaction yields were noted on increasing or decreasing the temperature. Similarly, on employing microwave (MW)-assisted method for the synthesis of coumarin–imidazole hybrid molecule **3**, significant improvement in product yield was observed with relatively short reaction time, the results on different solvents and catalysts combinations are summarized in Table 1. To understand more details of catalyst importance in both methods, that is conventional as well as MW conditions, we altered the molar ratio of all the catalyst (catalysts are recorded in Table 1) and reaction time. However, there is no change in product yield. It has been confirmed that the optimized yield, time and temperature were well satisfied. Further, knowing the importance of sulfonamide derivatives in the biological system, we focused our attention towards the synthesis of coumarin–imidazole sulfonamides; we carried out the reaction of compound **3** and *p*-toluenesulfonyl chloride in the presence of triethylamine (TEA) at 0–5 °C overnight to afford compound **4** in excellent yield. Compound **3** was also treated with sodium hydroxide (25%) in ethanol at 80 °C for about 4–5 h leads to compound **5** in good yields by ring opening, and compound **5** was isolated in the form of sodium salt. The structure of all newly synthesized compounds **3a–3g**, **4a–4f**, and **5a–5f** was confirmed by spectral analysis. IR spectrum of compound **3a** (R = C6–CH_3; R^1 = –H) shows 3430 and 1727 cm^{-1} due to –NH and carbonyl stretching bands, respectively; further formation

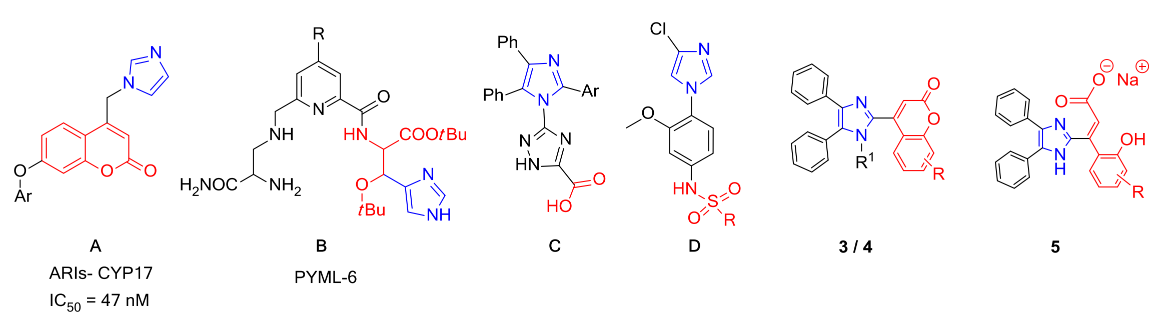


Fig. 1 **a–d** Potent anticancer and antimicrobial agents; **3**, **4**, and **5** are designed and synthesized bioactive molecules

Scheme 1

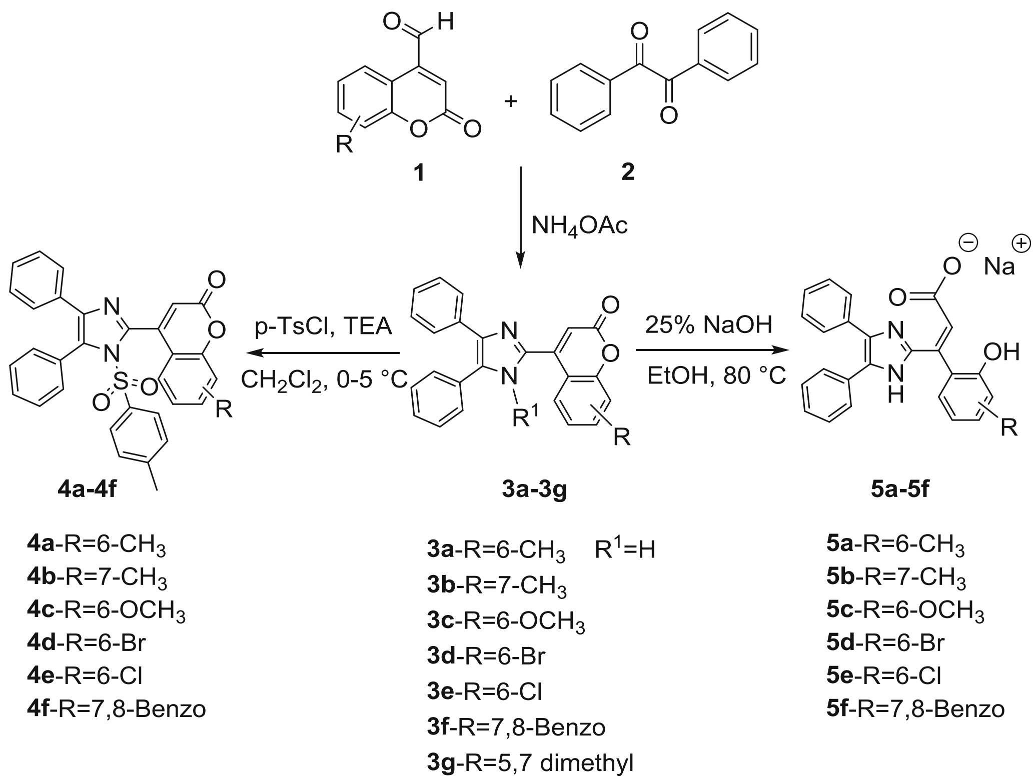


Table 1 Optimized condition for synthesis compound **3a** under conventional and microwave methods

Entry	Catalysts	Ratio	Solvent	Conventional			Microwave		
				Temp./°C	Time/h	Yield/%	Temp./°C	Time/min	Yield/%
1	Acetic acid	1:1	Ethanol	80	8	58	70	5	85
2	Acetic acid	1:2	PEG-400	100	8	64	120	5	78
3	Acetic acid	1:1	DMF	80	8	64	100	5	86
4	**Acetic acid**	**–**	**Acetic acid**	**90**	**8**	**88**	**100**	**5**	**92**
5	Silica H_2SO_4	1:1	Ethanol	80	5	58	70	4	68
6	Silica H_2SO_4	1:2	PEG-400	100	5	64	120	4	78
7	Silica H_2SO_4	1:1	DMF	80	5	62	100	3	74
8	Zinc oxide	2:1	Ethanol	110	8	48	100	6	53
9	Zinc oxide	2:2	PEG-400	120	8	54	120	6	57
10	Zinc oxide	2:1	DMF	100	8	50	100	6	52

Efficient protocol are in bold

of compound **3a** was supported by ^{1}H NMR, singlet was resonated at $\delta = 13.28$ ppm due to imidazole –NH and is D_2O exchangeable. A doublet was observed at 9.24 ppm for one proton ($J = 4.00$ Hz) due to C5–H of coumarin and this deshielding doublet was confirmed by C5 substitution (–CH_3 group substitution at C5, compound **3g**), four multiplets at 7.56, 7.48, 7.36, and 7.29 ppm corresponds to aromatic protons. Two singlets at 7.01 and 2.44 ppm corresponds to C3–H and C6–CH_3 of coumarin, respectively. ^{13}C NMR spectrum of compound **3a** shows distinct resonance in agreement with proposed structure. Formation of compounds **4a–4f** was confirmed by spectral analysis; compound **4a** shows the absence of –NH stretching band at 3430 cm^{-1} and strong SO_2 stretching band observed at 1380 cm^{-1}, further it was supported by ^{1}H NMR and ^{13}C NMR. Whereas, lactone ring opening from compound **3–5** was confirmed by IR spectra, compound **5a** shows the absence of lactone carbonyl and –NH and –OH broad band was observed at 3441 cm^{-1}. The ^{1}H NMR was recorded in deuterium water, spectrum shows the absence of –NH and –OH peaks and all aromatic protons are appeared in the respective region, ^{13}C NMR also supports the inferred structure.

Antimicrobial activity

Coumarin and imidazole molecules are very promising target compounds for antibacterial as well as antifungal activities. The synthesized compounds **3a–3g**, **4a–4f**, and **5a–5f** were screened for their antibacterial activity against Gram-positive *Bacillus flexus* and Gram-negative *Pseudomonas* spp. bacterial strains. Similarly, they were screened for their antifunal activity against *Scopulariopsis* spp. and *Aspergillus terreus* via agar well diffusion method. Ciprofloxacin and nystatin are the most effective antimicrobial agents and were used as the reference drugs. All the strains were incubated at 37 °C for about 48 h by inoculation into nutrient broth (Difco). The molten nutrient agar was inoculated with 100 mm^3 of the inoculums and poured into the Petri plate. The diameter of inhibition zones (in mm) was determined and MIC of the tested compounds was statistically determined by Turkey's fair wise test. A stock solution of the newly synthesized compounds **3a–3g** and **4a–4f** in DMSO and compounds **5a–5f** in sterilized water was prepared.

SAR study

Several important structural features of coumarin and imidazole derivatives are identified and constructed as coumarin–imidazole skeleton. The various substituents on coumarin exhibit better antimicrobial activity and deduced the following SAR study by scrutinizing their parameters in Table 2.

Antibacterial assay

Attachment of an imidazole nucleus at C4 position of the coumarin conjugates **3a–3g** was screened for their antibacterial activity and results obtained are summarized in Table 2. The MIC results indicate that all the synthesized compounds are highly active against both bacterial strains compared to standard. Substituents such as $–OCH_3$ (**3c**) and –Cl (**3e**) at C6 position of coumarin exhibited excellent activity (MIC values is 0.3 μg/cm^3). Moreover, C6 substitutions such as $–CH_3$ (**3a**) and –Br (**3d**) are less active compared to **3c** and **3e**. Substitutions at C7,8-benzo (**3f**) and C5,7-dimethyl (**3g**) are even less active compared to other substituents against both bacterial strains and compound **3b** is inactive.

The N-sulfonation of compounds **3a–3g** resulted in compounds **4a–4f**. The isolated compounds were screened for their antibacterial activity against Gram-positive and Gram-negative bacterial strains; the results obtained are listed in Table 2. The results of N-sulfonation of imidazole derivatives showed slight increase in the activity compared to compounds **3a–3g**, such as compounds **4d** (which contains bromo at C6 position of coumarin) and **4f** (which contains 7,8-benzo substitution on coumarin) with MIC value 0.2 μg/cm^3. Moreover, compound **4b** inactive against both the bacterial strains. The activity of compound **4c** remains same even after sulfonation of compound **3c** and the remaining substituent has not shown any changes in their activity.

The modified coumarin–imidazole hybrids to sodium salt of phenyl acrylate of imidazole **5a–5f** were tested against Gram-positive and Gram-negative bacterial strains. Minimum inhibitory concentration (MIC) was measured and the results were summarized in Table 2. The results show that all the compounds exhibited excellent antibacterial activities compared to standard and other series of compounds such as **3a–3g** and **4a–4f**. In this series, **3c** (which contains $–OCH_3$ and –OH on benzene ring), **5e** (chloro substitution and –OH on benzene ring) and **5f** (1-naphthol) are highly active with MIC values 0.1 μg/cm^3. Interestingly, the compounds **3b** and **4b** are inactive at a selected MIC of standard ciprofloxacin, wherein after lactone ring opening led to **5b** which showed promising activity against both Gram-positive and Gram-negative bacterial strains. The interesting findings in the activity difference between these three series of compounds **3a–3g**, **4a–4f**, and **5a–5f**, clearly evident that compounds **5a–5f** are more effective and highly potent against both bacterial strains, it might be the ionic nature which are completely soluble in water.

Antifungal assay

All the synthesized compounds **3a–3g**, **4a–4f**, and **5a–5f** were assessed for their antifungal activity against *Scopulariopsis* spp. and *A. terreus* organisms. The antifungal results obtained are listed in Table 2. In the series **3a–3g**, compounds **3a**, **3b**, **3e**, and **3g** are highly active against the both fungal strains with MIC values ranges from 1 to 4 μg/cm^3, while compounds **3c**, **3d**, and **3f** are inactive. In the case of N-sulfonation scaffolds **4a–4f**, except compound **4d** (bromo substitution on C6 position of coumarin) all the compounds are active against both fungal strains. Superior activity was noticed in the case of compounds **5a–5f**, these scaffolds are ionic in nature and completely soluble in water. All the compounds were more effective and potent against both fungal strains with MIC values ranges from 1 to 2 μg/cm^3. The structural activity views have been listed

Table 2 In vitro antimicrobial activity of compounds **3a–3g**, **4a–4f**, and **5a–5f**

Compounds	Minimum inhibitory concentration (MIC) in μg/cm^3			
	Gram-positive bacteria	Gram-negative bacteria	Antifungal	
	B. flexus	*P.* spp.	*S.* spp.	*A. tereus*
3a	0.4	0.4	0.1	0.4
3b	–	–	0.2	0.4
3c	0.3	0.3	–	–
3d	0.4	0.4	–	–
3e	0.3	0.4	0.2	0.2
3f	0.6	0.5	–	–
3g	0.7	0.6	0.1	0.2
4a	0.5	0.6	0.6	0.7
4b	–	–	0.2	0.2
4c	0.5	0.4	0.6	0.6
4d	0.2	0.2	–	–
4e	0.4	0.4	0.5	0.4
4f	0.2	0.2	0.6	0.4
5a	0.2	0.2	0.1	0.1
5b	0.3	0.3	0.2	0.1
5c	0.1	0.1	0.1	0.1
5d	0.2	0.2	0.1	0.2
5e	0.1	0.3	0.1	0.1
5f	0.1	0.1	0.2	0.2
Ciprofloxacin	0.7	0.7	–	–
Nystatin	–	–	0.7	0.7

below against both bacterial and fungal strains (Figs. 2, 3, 4).

From oral antimicrobial activity results, it was worth noting that all the synthesized compounds are highly active compared to standard ciprofloxacin and nystatin. Among synthesized series, compounds **5a–5f** are the most promising and highly potent scaffolds against both bacterial and fungal strains.

Antiinflammatory activity

The antiinflammatory activity of all newly synthesized compounds **3a–3g**, **4a–4f**, and **5a–5f** was screened using gelatin zymography method; this method can detect degrading gelatin which is generating by proteolytic enzymes. The two key matrix metalloproteinases such as 72 kDa called as gelatinase A (MMP-2) and 92 kDa gelatinase B (MMP-9), these two proteins have potent gelatin-degrading properties [28, 29]. In physiological system, various gelatinases are present and they play a key role in inflammation and autoimmunity stats [30]. Thus, the activated inflammatory cells can express several proteinases designated as matrix metalloproteinases (MMPs) these are able to degrade all connective tissue macromolecules. Among these gelatinases, MMP-2 and MMP-9 together forming collagenase play an important role in connective tissue remodeling [31]. MMP-2 and MMP-9 remains inactive, while they are with pro-domain, they get activates by denaturation, then it could be perceived on

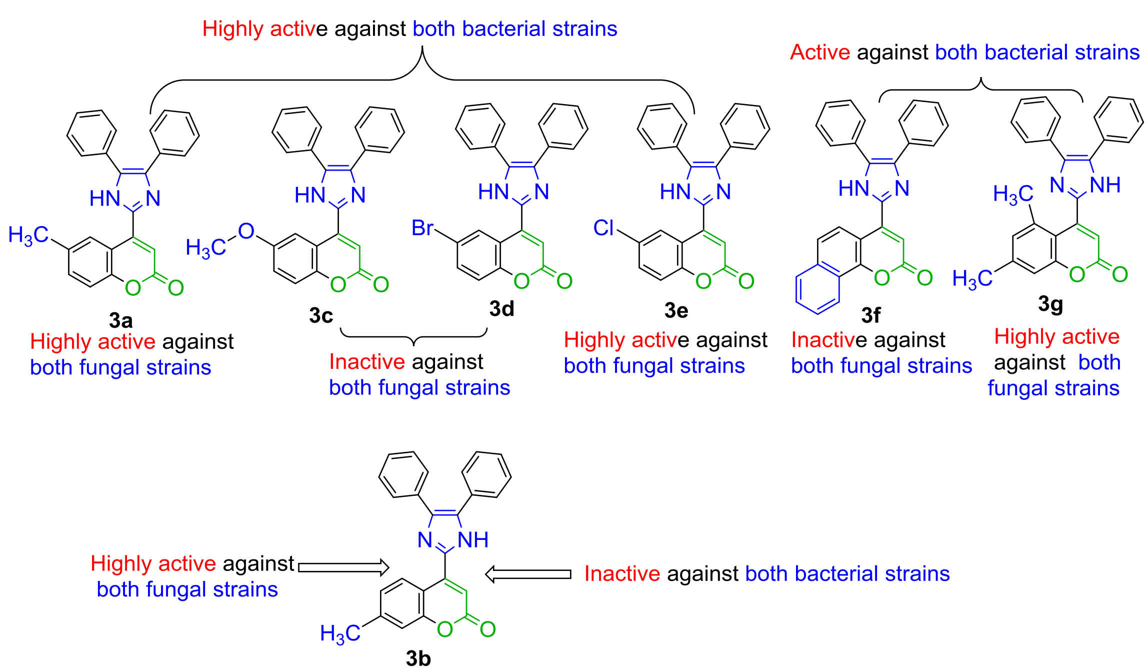


Fig. 2 Antimicrobial activity of compounds **3a–3g**

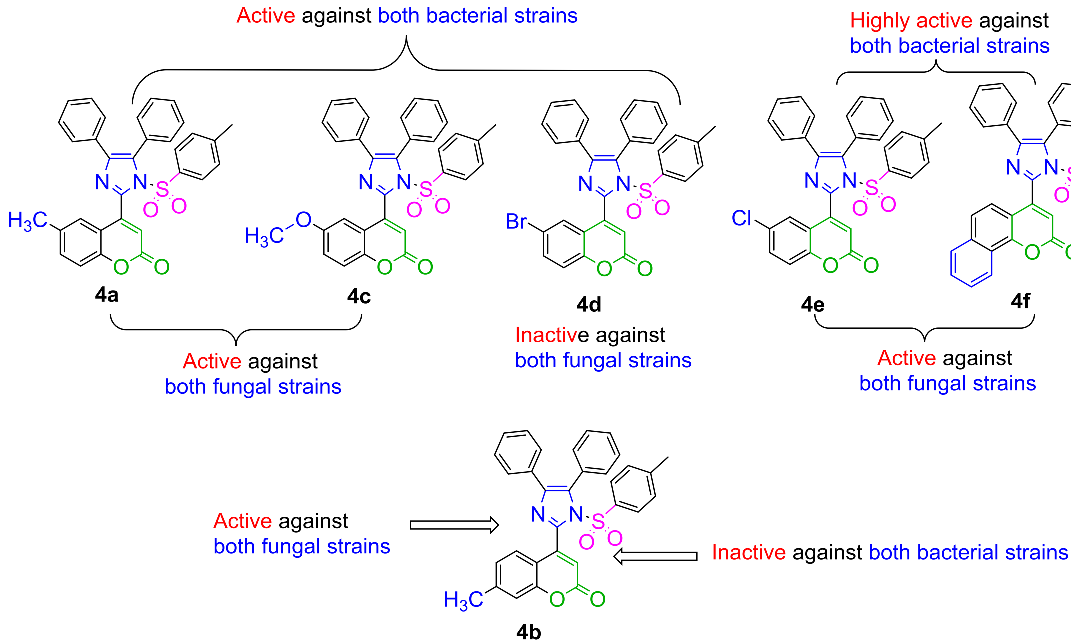


Fig. 3 Antimicrobial activity of compounds **4a–4f**

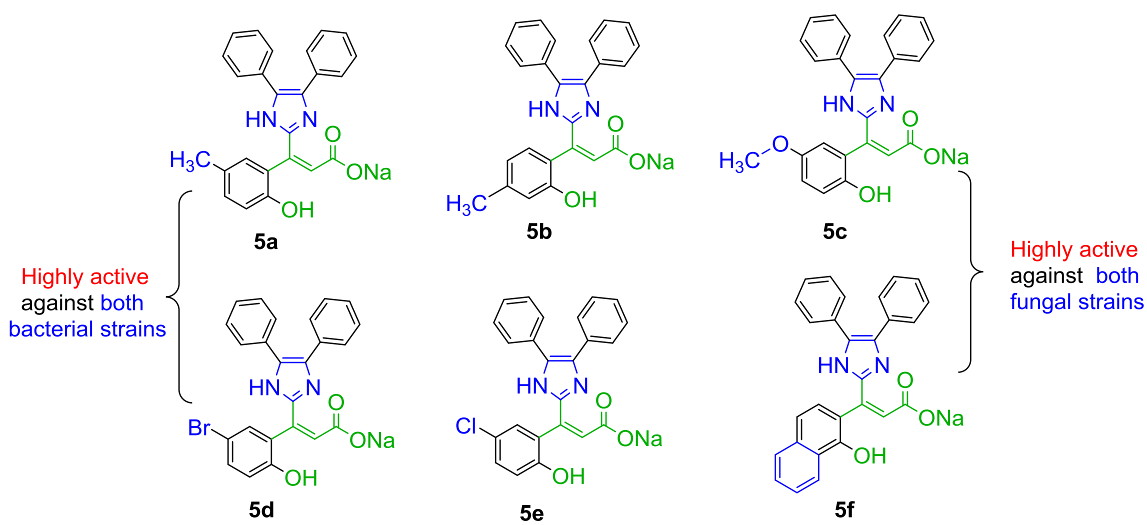


Fig. 4 Antimicrobial activity of compounds **5a–5f**

gelatin zymograms after staining with Coomassie blue as one or two bands which are designated as pro and activated forms. In vitro antiinflammatory activity results obtained are summarized in Table 3 and inhibition percentage of MMP-2 and MMP-9 for each compound was calculated by subtracting from 100 with % of bands obtained. The listed results notice that all the synthesized coumarin–imidazole hybrids (**3a–3g** and **4a–4f**) and phenyl-imidazole acrylates **5a–5f** are highly active against both MMPs (MMP-2 and MMP-9). Interestingly, among all these three series, compounds **5a–5f** are more potential and promising against both MMPs and the high potentiality might be the hydrophilic nature of the compounds. The obtained results are also presented in Fig. 5.

Molecular modeling studies

To define and elucidate the binding modes, molecular docking was performed for all the synthesized compounds on the active site of 4CAW and 1AI9 enzyme. The protein crystal structure of *Aspergillus fumigates* *N*-myristoyl transferase in complex with myristoyl CoA and pyrazolo sulfonamide ligand and *Candida albicans* dihydrofolate reductase were selected from the protein data bank (PDB ID: 4CAW; A-Chain and 1AI9). The proteins were prepared for docking by adding polar hydrogen atom with Gasteiger–Hückel charges and water molecules were removed. The 3D structure of the ligands were generated by the SKETCH module implemented in the SYBYL program [32] and its energy-minimized conformation was obtained with the help of the Tripos force field using Gasteiger–Hückel [33] charges. The molecular docking was performed with Surflex-Dock program that is interfaced with Sybyl-X 2.0 [34] and other miscellaneous parameters were assigned with the default values given by the software.

The docking study revealed that all the docked compounds are good antifungal agent against *A. fumigates* and *C. albicans*. Among all, compounds **5a**–**5f** series shown excellent interaction with enzyme, particularly compound **5a** and **5c** showed high C score values against PDB ID: 1AI9. As depicted (Fig. 6), compound **5a** involves six hydrogen bond interactions at the active site of enzyme (PDB ID: 1AI9). Nitrogen of imidazole ring involves an interaction with LYS57, two hydrogen bonding interactions are raised from oxygen of phenolic-OH with ARG79 and remaining three interactions are from the oxygen of –$COO^{-}Na^{+}$ with ARG56. The covalent structure of compound **5a** with amino acid residue is also shown (Fig. 6).

Similarly, compound **5c** involves six hydrogen bond interactions with the amino acid residues A/LYS57, A/ARG79 and A/GLU116 in the active site of the enzyme which are shown (Fig. 7). As depicted (Fig. 8), compound **5e** involves two hydrogen bond interactions at the active site of the enzyme (PDB ID: 4CAW; A-chain), possess interaction from hydrogen of hydroxyl group with GLY25 and another interaction from the oxygen of –$COO^{-}Na^{+}$ with GLY251. The docking score values are summarized in Table 4.

Therefore, the docking study corroborates the experimental findings, which suggest that the phenyl-imidazole acrylate derivatives act on both mechanisms such as inhibiting the dihydrofolate reductase and by inhibiting *N*-myristoyl transferase in complex with myristoyl CoA.

Conclusion

In summary, we have synthesized coumarin–imidazole derivatives **3a**–**3g**, **4a**–**4f**, and **5a**–**5f** both in conventional as well microwave irradiation method and evaluated for their antibacterial, antifungal, and antiinflammatory activities.

Table 3 In vitro antiinflammatory results of compounds **3a**–**3g**, **4a**–**4f**, and **5a**–**5f** with % bands and % inhibition of MMP-2 and MMP-9

Compounds	% Bands of MMP		% Inhibition of MMP	
	MMP-9	MMP-2	MMP-9	MMP-2
3a	20	10	**80**	**90**
3b	40	55	60	45
3c	100	15	Nil	**85**
3d	30	05	70	**95**
3e	10	04	**90**	**96**
3f	10	05	**90**	**95**
3g	85	65	15	35
4a	100	18	Nil	**82**
4b	90	13	10	**87**
4c	15	10	**85**	**90**
4d	70	20	30	**80**
4e	05	10	**95**	**90**
4f	90	85	10	15
Tetracycline	00	00	100	100
DMSO control	–	–	–	–
5a	10	08	**90**	**92**
5b	15	12	**85**	**88**
5c	05	05	**95**	**95**
5d	10	06	**90**	**94**
5e	05	04	**95**	**96**
5f	15	10	**85**	**90**
Tetracycline	00	00	100	100
Water control	–	–	–	–

Efficient protocol are in bold

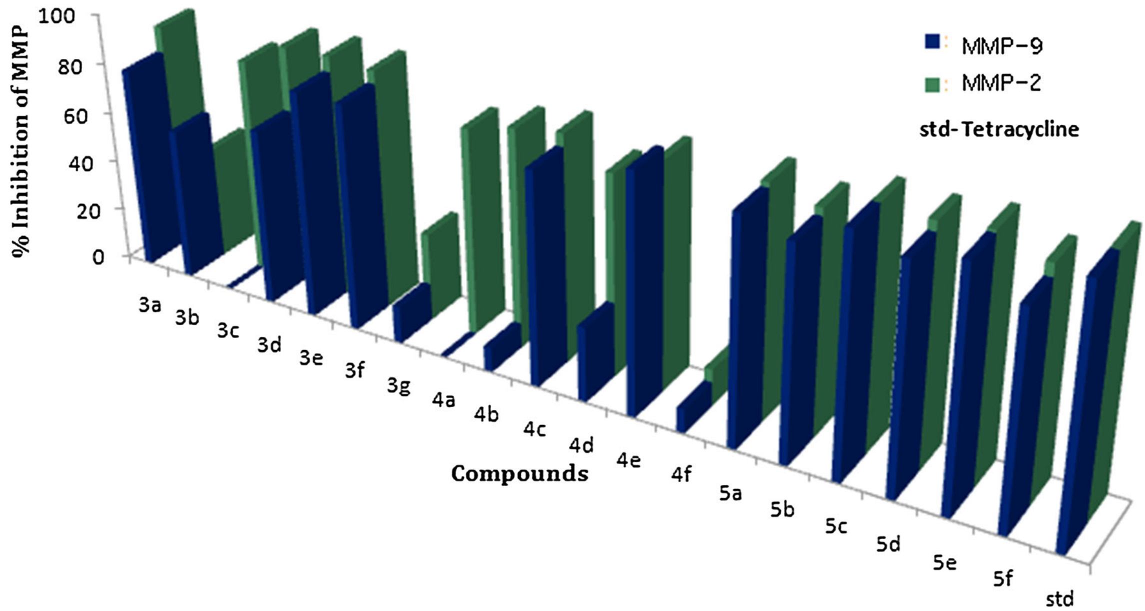


Fig. 5 Antiinflammatory activity of synthesized compounds **3a–3g**, **4a–4f**, and **5a–5f**

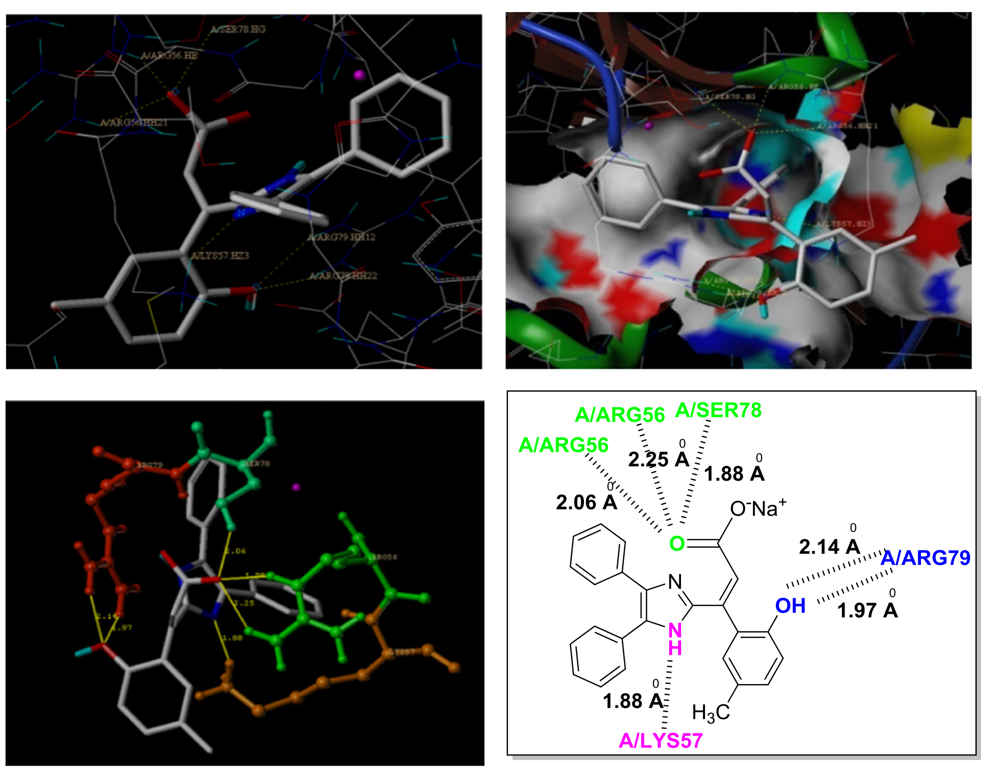


Fig. 6 Interactions of compound **5a** at the active site of enzyme PDB ID: 1AI9

Most of the compounds have exhibited excellent activity against bacterial and fungal strains. Among the compounds, all **5a–5f** were found to be more potent and have promising antibacterial and antifungal activities. Further, the molecular docking study of synthesized compounds showed promising interaction with enzyme and this also supports the compounds **5a–5f** being more potent than other series of compounds. Similarly, in vitro antiinflammatory activity of all compounds is more promising against MMP-2 and MMP-9, particularly all the compounds **3a–3g**, **4a–4f**, and **5a–5f** are found to be excellent against MMP-2. Moreover, we have seen that series **5a–5f** is quite promising and possess excellent activity against both MMP-2 and MMP-9 in comparison with the standard tetracycline. Findings from the antimicrobial and antiinflammatory studies indicats that the synthesized compounds are potential drugs.

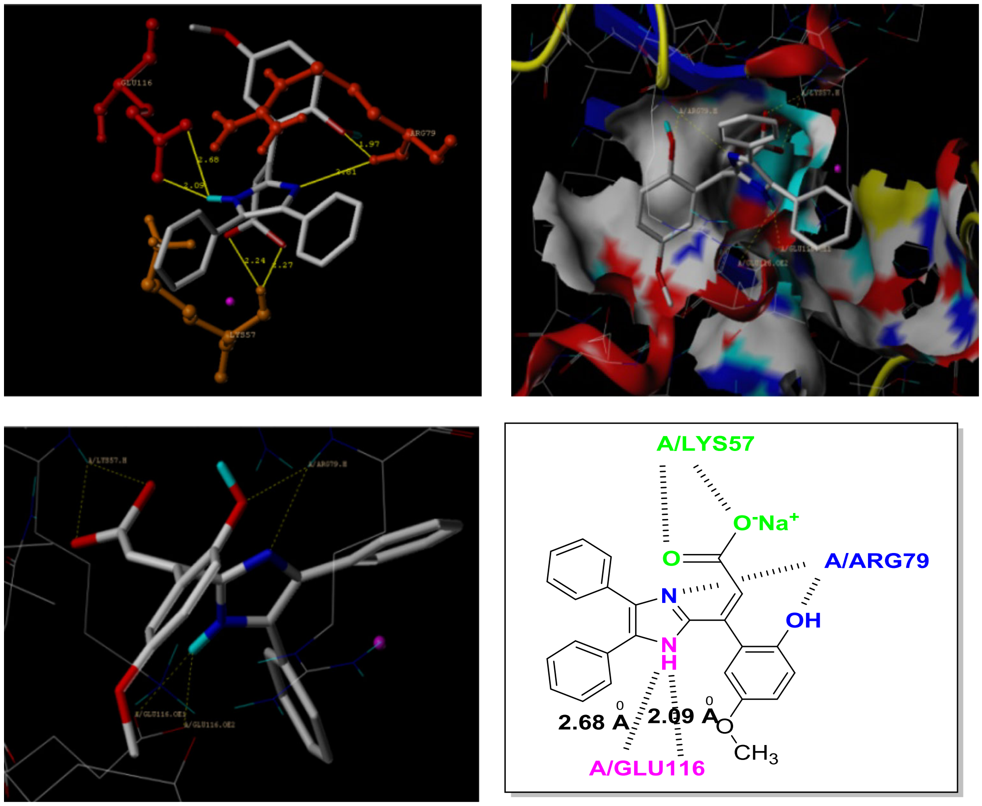


Fig. 7 Interactions of compound **5c** at the active site of enzyme (PDB ID: 1AI9)

Fig. 8 Interactions of compound **5e** at the active site of the enzyme PDB: 4CAW

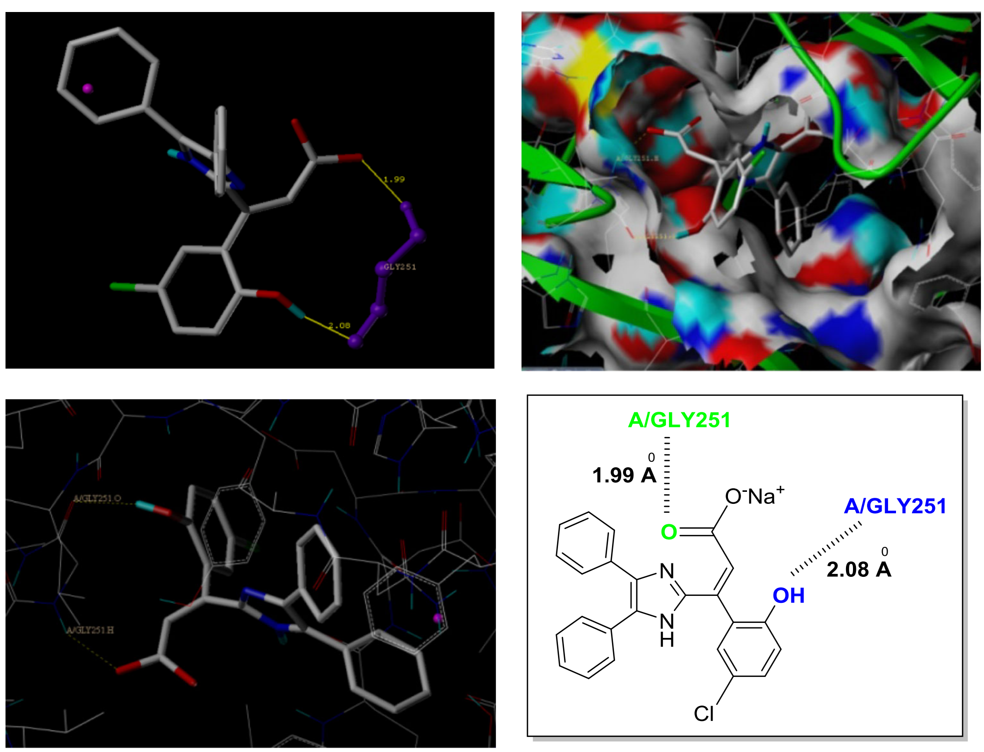


Table 4 Surflex docking scores (kJ/mol) of the coumarin–imidazole derivatives for *Candida albicans* (PDB ID: 1AI9)

Compounds	C score[a]	Crash score[b]	Polar score[c]	D score[d]	PMF score[e]	G score[f]	Chem score[g]
3a	18.66	− 8.61	4.30	− 7344.68	100.95	− 861.10	− 158.74
3b	19.53	− 3.09	0	− 6970.08	131.16	− 725.12	− 130.12
3c	10.08	− 5.10	0	− 7110.42	133.80	− 656.34	− 109.07
3d	16.94	− 1.92	0.29	− 4734.57	88.74	− 657.80	− 93.972
3e	18.20	− 2.05	0.92	− 4810.89	101.75	− 682.62	− 96.775
3f	19.91	− 3.97	6.56	− 6631.35	79.70	− 693.28	− 152.67
3g	20.45	− 3.05	4.18	− 6909.12	140.24	− 756.42	− 144.68
4a	18.82	− 11.08	6.48	− 5832.66	− 152.54	− 924.28	− 133.88
4b	9.70	− 4.058	4.26	− 4791.94	− 218.15	− 643.12	− 76.483
4c	8.40	− 23.13	7.65	− 7834.37	− 59.74	− 1161.23	− 147.73
4d	8.45	− 4.93	0.04	− 5555.56	− 174.97	− 718.72	− 76.818
4e	17.36	− 19.33	0.75	− 11780.5	285.22	− 1127.76	− 146.81
4f	15.06	− 9.41	0.16	− 10333	257.19	− 976.58	− 137.44
5a	42.76	− 7.57	26.52	− 5401.17	− 63.63	− 874.58	− 174.72
5b	19.41	− 8.45	17.53	− 2294.34	− 151.54	− 522.95	− 107.23
5c	30.91	− 8.11	15.69	− 2200.16	− 146.31	− 893.95	− 115.10
5d	27.78	− 5.69	19.58	− 2326.85	− 196.73	− 706.92	− 123.80
5e	28.07	− 8.28	16.69	− 1919.08	− 115.39	− 876.79	− 114.93
5f	28.70	− 8.49	15.60	− 4534.12	− 166.90	− 809.22	− 138.65

[a]C score (consensus score) integrates a number of popular scoring functions for ranking the affinity of ligands bound to the active site of a receptor and reports the output of total score

[b]Crash score revealing the inappropriate penetration into the binding site. Crash scores close to 0 are favorable. Negative numbers indicate penetration

[c]Polar indicating the contribution of the polar interactions to the total score. The polar score may be useful for excluding docking results that make no hydrogen bonds

[d]D-score for charge and van der Waals interactions between the protein and the ligand

[e]PMF score indicating the Helmholtz free energies of interactions for protein–ligand atom pairs (potential of mean force, PMF)

[f]G-score showing hydrogen bonding, complex (ligand–protein), and internal (ligand–ligand) energies

[g]Chem-score points for H bonding, lipophilic contact, and rotational entropy, along with an intercept term

Experimental

Reagents were obtained from commercial sources and used without further purification until stated. Melting points were determined in open capillary tube on a Shital Scientific instrument, presented in degrees centigrade. Microwave irradiation experiments were carried out using CEM Discover SP Microwave Synthesizer equipped with IR sensor to monitor the reaction temperatures. Infrared spectra were recorded on a Nicolet 410 Fourier transform (FT) infrared spectrometer using KBr pellets. ^{1}H and ^{13}C NMR spectra were recorded on Bruker 400 MHz instruments, and further chemical shifts were measured in terms of parts per million with TMS as the internal standard. The mass spectra were recorded using Shimadzu GCMS-QP2010S instrument.

General procedure for synthesis of 4-(4,5-diphenyl-1H-imidazol-2-yl)-2H-chromen-2-ones ***3a–3g***

Conventional method A mixture of 93 mg benzil (0.44 mmol), 34 mg ammonium acetate (0.44 mmol), and 100 mg substituted 4-formylcoumarin (0.44 mmol) in acetic acid were stirred at 100 °C for the appropriate time mentioned in Table 1. After completion (monitored by TLC), reaction mixture was poured into ice cold water. The solid separated was filtered, washed with water, and dried. The crude product was crystallized from dichloromethane (DCM).

Microwave method 93 mg Benzil (0.44 mmol), 34 mg ammonium acetate (0.44 mmol) and 100 mg 4-formylcoumarin (0.44 mmol) were mixed in acetic acid and irradiated for the appropriate time mentioned in Table 1. The progress of the reactions was monitored by TLC. The mixture was added into ice cold water; solid separated was washed with water and crystallized from dichloromethane (DCM).

4-(4,5-Diphenyl-1H-imidazol-2-yl)-6-methyl-2H-chromen-2-one (**3a**, $C_{25}H_{18}N_2O_2$)
White solid; m.p.: 220–222 °C; IR (KBr): $\bar{\nu} = 3430$, 1725 cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 13.28$ (s, 1H), 9.24 (d, $J = 4$ Hz, 1H), 7.56 (d, $J = 4$ Hz, 4H), 7.48 (m, 3H), 7.36 (m, 2H), 7.29 (m, 3H), 7.01 (s, 1H), 2.44 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 160.21$ (C_2 of coumarin), 154.00 (C_4 of coumarin), 142.17 (C_9 of coumarin), 140.31 (C_3 of imidazole), 140.21 (C_6 of coumarin), 138.84 (Ar–C), 134.24 (Ar–C), 130.72 (C_4 of imidazole), 130.11 (C_5 of imidazole), 128.54 (Ar–C), 128.72 (Ar–C), 128.57 (Ar–C), 128.30 (Ar–C), 127.50 (Ar–C), 127.01 (Ar–C), 126.20 (Ar–C), 125.04 (Ar–C), 124.19 (Ar–C), 116.29 (Ar–C), 115.20 (C_5 of coumarin), 114.00 (C_8 of coumarin), 112.42 (C_{10} of coumarin), 110.21 (C_3 of coumarin), 21.10 (C of methyl) ppm; GC–MS: *m/z* calculated for $C_{25}H_{18}N_2O_2$ 378, found 378.

4-(4,5-Diphenyl-1H-imidazol-2-yl)-7-methyl-2H-chromen-2-one (**3b**, $C_{25}H_{18}N_2O_2$)
White solid; m.p.: 215–217 °C; IR (KBr): $\bar{\nu} = 3435$, 1720 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): $\delta = 13.25$ (s, 1H), 9.23 (d, $J = 8.8$ Hz, 1H), 7.55 (m, 4H), 7.45 (m, 3H), 7.35 (t, $J = 14.8$ Hz, 2H), 7.27 (t, $J = 14.4$ Hz, 4H), 6.99 (s, 1H), 2.41 (s, 3H) ppm; ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 160.15$ (C_2 of coumarin), 153.99 (C_4 of coumarin), 142.78 (C_9 of coumarin), 140.99 (C_3 of imidazole), 140.20 (C_6 of coumarin), 138.92 (Ar–C), 134.28 (Ar–C), 130.31 (C_4 of imidazole), 129.91 (C_5 of imidazole), 128.69 (Ar–C), 128.60 (Ar–C), 128.43 (Ar–C), 128.36 (Ar–C), 127.20 (Ar–C), 127.09 (Ar–C), 125.95 (Ar–C), 125.45 (Ar–C), 124.29 (Ar–C), 116.70 (Ar–C), 114.11 (Ar–C), 113.39 (C_5 of coumarin), 111.43 (C_8 of coumarin), 110.43 (C_{10} of coumarin), 110.81 (C_3 of coumarin), 20.97 (C of methyl) ppm; GC–MS: *m/z* calculated for $C_{25}H_{18}N_2O_2$ 378, found 378.

4-(4,5-Diphenyl-1H-imidazol-2-yl)-6-methoxy-2H-chromen-2-one (**3c**, $C_{25}H_{18}N_2O_3$)
White solid; m.p.: 218–220 °C; IR (KBr): $\bar{\nu} = 3313$, 1698 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): $\delta = 13.27$ (s, 1H), 9.25 (d, $J = 8.8$ Hz, 1H), 7.65 (m, 4H), 7.40 (m, 3H), 7.36 (m, 2H), 7.25 (m, 3H), 6.98 (s, 1H), 3.84 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 160.25$ (C_2 of coumarin), 153.98 (C_4 of coumarin), 142.88 (C_9 of coumarin), 140.96 (C_3 of imidazole), 140.00 (C_6 of coumarin), 138.94 (Ar–C), 134.22 (Ar–C), 130.41 (C_4 of imidazole), 129.99 (C_4 of imidazole), 128.79 (Ar–C), 128.62 (Ar–C), 128.41 (Ar–C), 128.34 (Ar–C), 127.22 (Ar–C), 127.00 (Ar–C), 125.85 (Ar–C), 125.45 (Ar–C), 124.20 (Ar–C), 116.16 (Ar–C), 114.14 (C_5 of coumarin), 113.40 (C_8 of coumarin), 111.42 (C_{10} of coumarin), 110.80 (C_3 of coumarin), 56.18 (C of methyl) ppm; GC–MS: *m/z* calculated for $C_{25}H_{18}N_2O_3$ 394.13, found 394.

6-Bromo-4-(4,5-diphenyl-1H-imidazol-2-yl)-2H-chromen-2-one (**3d**, $C_{24}H_{15}BrN_2O_2$)
White solid; m.p.: 216–218 °C, IR (KBr): $\bar{\nu} = 3424$, 1720 cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 13.24$ (s, 1H), 9.28 (d, $J = 8.8$ Hz, 1H), 7.66 (m, 4H), 7.40 (m, 3H), 7.35 (m, 2H), 7.26 (m, 3H), 6.96 (s, 1H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 160.26$ (C_2 of coumarin), 154.00 (C_4 of coumarin), 143.28 (C_9 of coumarin), 141.16 (C_3 of imidazole), 140.24 (Ar–C), 139.24 (Ar–C), 134.24 (C_4 of imidazole), 130.46 (C_5 of imidazole), 123.00 (Ar–C), 128.29 (Ar–C), 128.65 (Ar–C), 128.46 (Ar–C), 128.35 (Ar–C), 127.23 (Ar–C), 127.20 (Ar–C), 125.75 (Ar–C), 125.35 (Ar–C), 124.22 (Ar–C), 116.17 (Ar–C), 114.54 (C_5 of coumarin), 113.41 (C_8 of coumarin), 112.32 (C_{10} of coumarin), 110.61 (C_3 of coumarin) ppm; GC–MS: *m/z* calculated for $C_{24}H_{15}BrN_2O_2$ 442.03, found 442.

6-Chloro-4-(4,5-diphenyl-1H-imidazol-2-yl)-2H-chromen-2-one (**3e**, $C_{24}H_{15}ClN_2O_2$)
White solid; m.p.: 220–223 °C; IR (KBr): $\bar{\nu} = 3425$, 1724 cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 13.44$ (s, 1H), 9.26 (d, $J = 8.8$ Hz, 1H), 7.68 (m, 4H), 7.42 (m, 3H), 7.34 (m, 2H), 7.28 (m, 3H), 6.98 (s, 1H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 160.28$ (C_2 of coumarin), 154.02 (C_9 of coumarin), 143.29 (C_3 of imidazole), 141.14 (Ar–C), 140.25 (Ar–C), 139.26 (C_4 of imidazole), 134.25 (C_5 of imidazole), 130.48 (Ar–C), 123.01 (Ar–C), 128.28 (Ar–C), 128.66 (Ar–C), 128.48 (Ar–C), 128.36 (Ar–C), 127.24 (Ar–C), 127.22 (Ar–C), 125.76 (Ar–C), 125.34 (Ar–C), 124.20 (Ar–C), 116.18 (Ar–C), 114.55 (C_5 of coumarin), 113.43 (C_8 of coumarin), 112.33 (C_{10} of coumarin), 110.62 (C_3 of coumarin) ppm; GC–MS: *m/z* calculated for $C_{24}H_{15}ClN_2O_2$ 398.09, found 398.

4-(4,5-Diphenyl-1H-imidazol-2-yl)-2H-benzo[h]chromen-2-one (**3f**, $C_{28}H_{18}N_2O_2$)
White solid; m.p.: 218–220 °C; IR (KBr): $\bar{\nu} = 3427$, 1692 cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 13.33$ (s, 1H), 9.40 (d, $J = 8.8$ Hz, 1H), 8.43 (d, $J = 8$ Hz, 1H), 8.04 (d, $J = 7.6$ Hz, 1H), 7.91 (d, $J = 9.2$ Hz, 1H), 7.73 (m, 2H), 7.60 (m, 4H), 7.45 (m, 6H), 7.16 (s, 1H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 159.83$ (C_2 of coumarin), 150.98 (C_9 of coumarin), 141.45 (C_4 of coumarin), 141.13 (C_3 of imidazole), 141.05 (Ar–C), 139.06 (Ar–C), 134.28 (C_4 of imidazole), 134.20 (C_5 of imidazole), 130.44 (Ar–C), 129.90 (Ar–C), 128.92 (Ar–C), 128.71 (Ar–C), 128.62 (Ar–C), 128.51 (Ar–C),128.40 (Ar–C), 127.76 (Ar–C), 127.24 (Ar–C), 127.20 (Ar–C), 127.14 (C_{12} of coumarin), 124.08 (C_{11} of coumarin), 123.85 (C_7 of coumarin), 123.65 (C_6 of coumarin), 122.38, 121.75 (C_5

of coumarin), 121.43 (C_8 of coumarin), 112.46 (C_3 of coumarin), 111.44 (C_{10} of coumarin) ppm; GC–MS: *m/z* calculated for $C_{28}H_{18}N_2O_2$ 414.14, found 414.

4-(4,5-Diphenyl-1H-imidazol-2-yl)-5,7-dimethyl-2H-chromen-2-one (**3g**, $C_{26}H_{20}N_2O_2$)
White solid; m.p.: 220–222 °C; IR (KBr): $\bar{\nu} = 3437$, 1727 cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): $\delta = 13.00$ (s, 1H), 7.51 (dd, $J = 8$, 8.4 Hz, 1H), 7.43 (t, $J = 12$ Hz, 2H), 7.38 (d, $J = 8$ Hz, 1H), 7.31 (t, $J = 12$ Hz, 2H), 7.25 (d, $J = 8$ Hz, 1H), 7.20 (s, 1H), 7.04 (s, 1H), 6.62 (s, 1H), 2.39 (s, 3H), 2.04 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 160.25$ (C_2 of coumarin), 154.20 (C_9 of coumarin), 142.18 (C_3 of imidazole), 140.32 (Ar–C), 140.27 (Ar–C), 138.82 (C_4 of imidazole), 134.25 (C_5 of imidazole), 130.76 (Ar–C), 130.31 (Ar–C), 128.79 (Ar–C), 128.31 (Ar–C), 127.53 (Ar–C), 127.08 (Ar–C), 126.22 (Ar–C), 125.64 (Ar–C), 124.21 (Ar–C), 116.31 (Ar–C), 115.23 (C_5 of coumarin), 114.10 (C_8 of coumarin), 112.44 (C_{10} of coumarin), 110.23 (C_3 of coumarin), 22.01 (C_7 of methyl), 21.18 (C_5 of methyl) ppm; GC–MS: *m/z* calculated for $C_{26}H_{20}N_2O_2$ 392.15, found 392.

General procedure for synthesis of 4-(4,5-diphenyl-1-tosyl-1H-imidazol-2-yl)-2H-chromen-2-ones ***4a–4f***
In a clean and dry flask containing 50 cm^3 methylene dichloride with 100 mg compound **3** (0.000241 mol), cool the solution to 0–5 °C and 69 mg solid *p*-toluenesulfonyl chloride (0.00036 mol) was added in two portion over the course of 30 min. After 10–15 min 0.1 cm^3 triethylamine (0.001 mol) was added drop wise over a period of 30 s. The reaction mixture was stirred overnight. Completion of the reaction was monitored by TLC. After completion, the reaction mixture was diluted in 150 cm^3 methylene dichloride and washed with 3 M HCl followed by the saturated $NaHCO_3$ solution and brine. The organic layer was dried with anhydrous Na_2SO_4 and evaporated under reduced pressure to obtain the desired product **4a–4f**.

4-(4,5-Diphenyl-1-tosyl-1H-imidazol-2-yl)-6-methyl-2H-chromen-2-one (**4a**, $C_{32}H_{24}N_2O_4S$)
Yield 80%; white solid; m.p.: 189–190 °C; IR (KBr): $\bar{\nu} = 1724$, 1380, 1184 cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): $\delta = 8.89$ (s, 1H), 8.13 (d, $J = 8$ Hz, 2H), 7.61 (d, $J = 8$ Hz, 2H), 7.55 (m, 2H), 7.50 (m, 3H), 7.48 (m, 3H), 7.41 (d, $J = 12$ Hz, 2H), 7.06 (d, $J = 12$ Hz, 2H), 6.85 (s, 1H), 2.49 (s, 3H), 2.48 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 159.66$ (C_2 of coumarin), 151.89 (C_4 of coumarin), 146.52 (C_9 of coumarin), 143.65 (C_4 of tosyl), 141.64 (C_3 of imidazole), 134.17 (C_1 of tosyl), 133.65 (C_6 of coumarin), 133.42 (Ar–C), 132.42(Ar–C), 132.82 (C_3 of tosyl), 132.00 (C_5 of tosyl), 131.21 (Ar–C), 129.27 (Ar–C), 129.44 (Ar–C), 129.88 (Ar–C), 129.00 (Ar–C), 128.52 (Ar–C), 128.42 (Ar–C), 128.52 (C_7 of coumarin), 127.97 (C_2 of tosyl), 127.99 (C_6 of tosyl), 126.32 (Ar–C), 124.42 (Ar–C), 122.47 (Ar–C), 120.21 (Ar–C), 116.75 (C_5 of imidazole), 116.34 (C_8 of coumarin), 115.44 (C_{10} of coumarin), 111.99 (C_3 of coumarin), 24.21 (C_6 of methyl), 23.00 (C_4 of methyl) ppm; GC–MS: *m/z* calculated for $C_{31}H_{22}N_2O_4S$ 518.13, found 518.

4-(4,5-Diphenyl-1-tosyl-1H-imidazol-2-yl)-7-methyl-2H-chromen-2-one (**4b**, $C_{32}H_{24}N_2O_2S$)
Yield 82%; white solid; m.p.: 190–192 °C; IR (KBr): $\bar{\nu} = 1728$ cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): $\delta = 8.88$ (s, 1H), 8.15 (d, $J = 8$ Hz, 2H), 7.60 (d, $J = 8$ Hz, 2H), 7.54 (m, 2H), 7.48 (m, 3H), 7.49 (m, 3H), 7.44 (d, $J = 12$ Hz, 2H), 7.08 (d, $J = 12$ Hz, 2H), 6.86 (s, 1H), 2.46 (s, 3H), 2.46 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 159.66$ (C_2 of coumarin), 151.89 (C_4 of coumarin), 146.52 (C_9 of coumarin), 143.65 (C_4 of tosyl), 141.64 (C_3 of imidazole), 134.17 (C_1 of tosyl), 133.65 (C_6 of coumarin), 133.42 (Ar–C), 132.42 (Ar–C), 132.82 (C_3 of tosyl), 136.00 (C_5 of tosyl), 134.21 (Ar–C), 129.27 (Ar–C), 129.44 (Ar–C), 129.88 (Ar–C), 129.00 (Ar–C), 128.52 (Ar–C), 128.42 (Ar–C), 128.52 (C_7 of coumarin), 127.97 (C_2 of tosyl), 127.99 (C_6 of tosyl), 126.32 (Ar–C), 124.42 (Ar–C), 122.47 (Ar–C), 120.21 (Ar–C), 116.75 (C_5 of imidazole), 116.34 (C_8 of coumarin), 115.44 (C_{10} of coumarin), 111.99 (C_3 of coumarin), 24.21 (C_6 of methyl), 23.00 (C_4 of methyl) ppm; GC–MS: *m/z* calculated for $C_{31}H_{22}N_2O_4S$ 518.13, found 518.

4-(4,5-Diphenyl-1-tosyl-1H-imidazol-2-yl)-6-methoxy-2H-chromen-2-one (**4c**, $C_{32}H_{24}N_2O_5S$)
Yield 78%; white solid; m.p.: 201–203 °C; IR (KBr): $\bar{\nu} = 1698$ cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): $\delta = 8.88$ (s, 1H), 8.12 (d, $J = 8$ Hz, 2H), 7.62 (d, $J = 8$ Hz, 2H), 7.56 (m, 2H), 7.55 (m, 3H), 7.50 (m, 3H), 7.43 (d, $J = 12$ Hz, 2H), 7.12 (d, $J = 12$ Hz, 2H), 6.89 (s, 1H), 3.84 (s, 3H), 2.49 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 158.65$ (C_2 of coumarin), 151.87 (C_4 of coumarin), 146.55 (C_9 of coumarin), 143.64 (C_4 of tosyl), 141.60 (C_3 of imidazole), 134.18 (C_1 of tosyl), 133.63 (C_6 of coumarin), 133.49 (Ar–C), 132.38 (Ar–C), 132.22 (C_3 of tosyl), 136.01 (C_5 of tosyl), 134.22 (Ar–C), 129.28 (Ar–C), 129.46 (Ar–C), 129.89 (Ar–C), 129.45 (Ar–C), 128.62 (Ar–C), 128.49 (Ar–C), 128.50 (Ar–C), 127.00 (C_7 of coumarin), 127.09 (C_6 of tosyl), 126.30 (C_2 of tosyl), 124.41 (Ar–C), 122.48 (Ar–C), 120.22 (Ar–C), 116.95 (Ar–C), 116.36 (C_5 of imidazole), 115.47 (C_8 of coumarin), 120.00 (C_{10} of coumarin), 110.26 (C_3 of coumarin), 56.23 (C_6 of methoxy), 23.04 (C_4 of methyl) ppm; GC–MS: *m/z* calculated for $C_{32}H_{24}N_2O_5S$ 548.14, found 548.

6-Chloro-4-(4,5-diphenyl-1-tosyl-1H-imidazol-2-yl)-2H-chromen-2-one (**4d**, $C_{31}H_{21}BrN_2O_4S$)
Yield 78%; pale yellow solid; m.p.: 194–196 °C; IR (KBr): $\bar{\nu} = 1730$ cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 8.89$ (s, 1H), 8.16 (d, $J = 8$ Hz, 2H), 7.62 (d, $J = 8$ Hz, 2H), 7.55 (m, 2H), 7.50 (m, 3H), 7.48 (m, 3H), 7.45 (d, $J = 12$ Hz, 2H), 7.04 (d, $J = 12$ Hz, 2H), 6.92 (s, 1H), 2.46 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 159.68$ (C_2 of coumarin), 151.90 (C_4 of coumarin), 146.50 (C_9 of coumarin), 143.62 (C_4 of tosyl), 141.58 (C_3 of imidazole), 134.18 (C_1 of tosyl), 133.72 (C_6 of coumarin), 133.44 (Ar–C), 132.46 (Ar–C), 132.84 (C_3 of tosyl), 136.02 (C_5 of tosyl), 134.24 (Ar–C), 129.28 (Ar–C), 129.46 (Ar–C), 129.90 (Ar–C), 129.02 (Ar–C), 128.55 (Ar–C), 128.47 (Ar–C), 128.48 (C_7 of coumarin), 127.00 (C_6 of tosyl), 127.96 (C_2 of tosyl), 126.32 (Ar–C), 124.34 (Ar–C), 122.46 (Ar–C), 120.22 (Ar–C), 116.77 (C_5 of imidazole), 116.32 (C_8 of coumarin), 115.42 (C_{10} of coumarin), 111.98 (C_3 of coumarin), 23.02 (C_4 of methyl) ppm; GC–MS: *m/z* calculated for $C_{31}H_{21}BrN_2O_4S$ 597.48, found 597.

6-Chloro-4-(4,5-diphenyl-1-tosyl-1H-imidazol-2-yl)-2H-chromen-2-one (**4e**, $C_{31}H_{21}ClN_2O_4S$)
Yield 70%; pale yellow solid; m.p.: 190–192 °C; IR (KBr): $\bar{\nu} = 1700$ cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 8.90$ (s, 1H), 8.15 (d, $J = 8$ Hz, 2H), 7.60 (d, $J = 8$ Hz, 2H), 7.54 (m, 2H), 7.48 (m, 3H), 7.47 (m, 3H), 7.46 (d, $J = 12$ Hz, 2H), 7.10 (d, $J = 12$ Hz, 2H), 6.92 (s, 1H), 2.48 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 158.92$ (C_2 of coumarin), 152.01 (C_4 of coumarin), 146.52 (C_9 of coumarin), 143.64 (C_4 of tosyl), 141.50 (C_3 of imidazole), 134.16 (C_1 of tosyl), 133.74 (C_6 of coumarin), 133.46 (C_4 of tosyl), 132.48 (Ar–C), 132.80 (Ar–C), 136.04 (C_3 of tosyl), 134.28 (C_3 of tosyl), 129.30 (Ar–C), 129.44 (Ar–C), 129.92 (Ar–C), 129.04 (Ar–C), 128.54 (Ar–C), 128.48 (Ar–C), 128.40 (C_7 of coumarin), 127.02 (C_6 of tosyl), 127.98 (C_2 of tosyl), 126.34 (Ar–C), 124.36 (Ar–C), 122.40 (Ar–C), 120.32 (Ar–C), 116.78 (C_5 of imidazole), 116.33 (C_8 of coumarin), 115.40 (C_{10} of coumarin), 111.86 (C_3 of coumarin), 23.04 (C_4 of methyl) ppm; GC–MS: *m/z* calculated for $C_{31}H_{21}ClN_2O_4S$ 553.03, found 553.

4-(4,5-Diphenyl-1-tosyl-1H-imidazol-2-yl)-2H-benzo[h]-chromen-2-one (**4f**, $C_{35}H_{24}N_2O_4S$)
Yield 78%; white solid; m.p.: 192–194 °C; IR (KBr): $\bar{\nu} = 1696$ cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): $\delta = 8.88$ (s, 1H), 8.86 (d, $J = 8$ Hz, 2H), 7.68 (d, $J = 8$ Hz, 2H), 7.65 (m, 2H), 7.58 (m, 3H), 7.60 (m, 3H), 7.40 (d, $J = 12$ Hz, 2H), 7.10 (d, $J = 12$ Hz, 2H), 6.90 (s, 1H), 2.34 (s, 3H) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): $\delta = 158.70$ (C_2 of coumarin), 151.88 (C_4 of coumarin), 146.54 (C_{13} of coumarin), 143.68 (C_4 of tosyl), 141.62 (C_3 of imidazole), 134.20 (C_1 of tosyl), 133.64 (C_{12} of coumarin), 133.50 (Ar–C), 132.40 (Ar–C), 132.24 (C_3 of tosyl), 136.02 (C_5 of tosyl), 134.28 (Ar–C), 129.30 (Ar–C), 129.50 (Ar–C), 129.90 (Ar–C), 129.40 (C_3 of imidazole), 128.64 (Ar–C), 128.41 (Ar–C), 128.52 (C_2 of tosyl), 127.04 (C_6 of tosyl), 127.10 (Ar–C), 126.32 (Ar–C), 124.42 (Ar–C),122.50 (Ar–C), 120.24 (C_7 of coumarin), 116.96 (C_8 of coumarin), 116.40 (C_9 of coumarin), 115.47 (C_{11} of coumarin), 120.02 (Ar–C), 110.28 (Ar–C), 23.10 (C_4 of methyl) ppm; GC–MS: *m/z* calculated for $C_{35}H_{24}N_2O_4S$ 568.64, found 568.

General procedure for synthesis of sodium 3-(4,5-diphenyl-1H-imidazol-2-yl)-3-(2-hydroxyphenyl)acrylates ***5a–5f***
In a RB flask 4-(4,5-diphenyl-1*H*-imidazol-2-yl)-2*H*-chromen-2-one (**3**) with NaOH in 30–40 cm^3 ethanol (25%), was taken and heated an oil bath at 70–80 °C for 8 h. The reaction mixture was cooled, then excess ethanol was removed under pressure and reaction mixture was neutralized up to pH 7.5 with diluted HCl solution. The formed precipitate was filtered, washed with DCM, and dried to give the desired product **5**.

Sodium 3-(4,5-diphenyl-1-tosyl-1H-imidazol-2-yl)-3-(2-hydroxy-5-methylphenyl)acrylate (**5a**, $C_{25}H_{19}N_2NaO_3$)
Yield 72%; reddish solid; m.p.: 194–196 °C; IR (KBr): $\bar{\nu} = 3441$, 1643, 1417 cm^{-1}; ^{1}H NMR (400 MHz, D_2O): $\delta = 8.38$ (d, $J = 4$ Hz, 1H), 7.62 (d, $J = 4$ Hz, 4H), 7.50 (m, 3H), 7.38 (m, 2H), 7.29 (m, 3H), 6.92 (s, 1H), 2.18 (s, 3H) ppm; ^{13}C NMR (100 MHz, D_2O): $\delta = 167.21$ (C_1 of acrylate), 160.45 (C_3 of acrylate), 154.02 (Ar–C), 142.18 (C_3 of imidazole), 140.37 (Ar–C), 140.24 (Ar–C), 138.88 (Ar–C), 132.22 (C_4 of imidazole), 132.78 (C_5 of imidazole), 130.12 (Ar–C), 128.52 (Ar–C), 128.74 (Ar–C), 128.52 (Ar–C), 128.28 (Ar–C), 127.52 (Ar–C), 127.20 (Ar–C), 127.15 (Ar–C), 126.18 (Ar–C), 125.16 (Ar–C), 124.18 (Ar–C), 116.28 (Ar–C), 115.10 (Ar–C), 112.40 (Ar–C), 110.22 (C_2 of acrylate), 22.05 (C of methyl) ppm.

Sodium 3-(4,5-diphenyl-1H-imidazol-2-yl)-3-(2-hydroxy-4-methylphenyl)acrylate (**5b**, $C_{25}H_{19}N_2NaO_3$)
Yield 64%; reddish solid; m.p.: 196–198 °C; IR (KBr): $\bar{\nu} = 3442$, 1650, 1410 cm^{-1}; ^{1}H NMR (400 MHz, D_2O): $\delta = 8.40$ (d, $J = 4$ Hz, 1H), 7.60 (d, $J = 4$ Hz, 4H), 7.55 (m, 3H), 7.36 (m, 2H), 7.30 (m, 3H), 7.00 (s, 1H), 2.27 (s, 3H) ppm; ^{13}C NMR (100 MHz, D_2O): $\delta = 167.54$ (C_1 of acrylate), 160.55 (C_3 of acrylate), 154.32 (Ar–C), 142.19 (C_3 of imidazole), 140.47 (Ar–C), 140.44 (Ar–C), 138.80 (Ar–C), 134.21 (Ar–C), 130.75 (C_5 of imidazole), 130.10 (C_4 of imidazole), 128.58 (Ar–C), 128.72 (Ar–C), 128.54 (Ar–C), 128.30 (Ar–C), 127.50 (Ar–C), 127.24 (Ar–C), 127.18 (Ar–C), 126.20 (Ar–C), 125.18 (Ar–C), 124.19 (Ar–C), 116.31 (Ar–C), 115.12 (Ar–C), 112.43 (Ar–C), 110.27 (C_2 of acrylate), 22.21 (C of methyl) ppm.

Sodium 3-(4,5-diphenyl-1H-imidazol-2-yl)-3-(2-hydroxy-5-methoxyphenyl)acrylate (**5c**, $C_{25}H_{19}N_2NaO_4$)
Yield 72%; reddish solid; m.p.: 198–200 °C; IR (KBr): $\bar{\nu}$ = 3440, 1644, 1416 cm^{-1}; ^1H NMR (400 MHz, D_2O): δ = 8.36 (d, J = 4 Hz, 1H), 7.60 (d, J = 4 Hz, 4H), 7.48 (m, 3H), 7.32 (m, 2H), 7.30 (m, 3H), 6.90 (s, 1H), 3.84 (s, 3H), 2.24 (s, 3H) ppm; ^{13}C NMR (100 MHz, D_2O): δ = 167.20 (C_1 of acrylate), 160.44 (C_3 of acrylate), 154.10 (Ar–C), 142.32 (C_3 of imidazole), 140.36 (Ar–C), 140.25 (Ar–C), 138.80 (Ar–C), 134.24 (Ar–C), 130.82 (C_4 of imidazole), 130.14 (C_5 of imidazole), 128.50 (Ar–C), 128.70 (Ar–C), 128.54 (Ar–C), 128.30 (Ar–C), 127.54 (Ar–C), 127.28 (Ar–C), 127.12 (Ar–C), 126.20 (Ar–C), 125.14 (Ar–C), 124.20 (Ar–C), 116.30 (Ar–C), 115.12 (Ar–C), 112.00 (Ar–C), 110.24 (C_2 of acrylate), 54.02 (C of methoxy) ppm.

Sodium 3-(5-bromo-2-hydroxyphenyl)-3-(4,5-diphenyl-1H-imidazol-2-yl)acrylate (**5d**, $C_{24}H_{16}BrN_2NaO_3$)
Yield 70%; reddish solid; m.p.: 196–198 °C; IR (KBr): $\bar{\nu}$ = 3438, 1662, 1400 cm^{-1}; ^1H NMR (400 MHz, D_2O): δ = 8.42 (d, J = 4 Hz, 1H), 7.60 (d, J = 4 Hz, 4H), 7.55 (m, 3H), 7.35 (m, 2H), 7.32 (m, 3H), 6.90 (s, 1H) ppm; ^{13}C NMR (100 MHz, D_2O): δ = 168.20 (C_1 of acrylate), 160.44 (C_3 of acrylate), 154.04 (Ar–C), 142.20 (C_3 of imidazole), 140.38 (Ar–C), 140.26 (Ar–C), 138.86 (Ar–C), 134.24 (Ar–C), 130.80 (C_5 of imidazole), 130.14 (C_4 of imidazole), 128.54 (Ar–C), 128.75 (Ar–C), 128.55 (Ar–C), 128.32 (Ar–C), 127.54 (Ar–C), 127.22(Ar–C), 127.14 (Ar–C), 126.22 (Ar–C), 125.18 (Ar–C), 124.25 (Ar–C), 116.24 (Ar–C), 115.12 (Ar–C), 112.00 (Ar–C), 110.20 (C_2 of acrylate) ppm.

Sodium 3-(5-chloro-2-hydroxyphenyl)-3-(4,5-diphenyl-1H-imidazol-2-yl)acrylate (**5e**, $C_{24}H_{16}ClN_2NaO_3$)
Yield 64%; reddish solid; m.p.: 195–197 °C; IR (KBr): $\bar{\nu}$ = 3440, 1666, 1402 cm^{-1}; ^1H NMR (400 MHz, D_2O): δ = 8.44 (d, J = 4 Hz, 1H), 7.62 (d, J = 4 Hz, 4H), 7.54 (m, 3H), 7.36 (m, 2H), 7.28 (m, 3H), 6.92 (s, 1H) ppm; ^{13}C NMR (100 MHz, D_2O): δ = 168.22 (C_1 of acrylate), 160.45 (C_3 of acrylate), 154.22 (Ar–C), 142.32 (C_3 of imidazole), 140.36 (Ar–C), 140.24 (Ar–C), 138.78 (Ar–C), 134.26 (Ar–C), 130.82 (C_5 of imidazole), 130.18 (C_4 of imidazole), 128.55 (Ar–C), 128.21 (Ar–C), 128.04 (Ar–C), 128.00 (Ar–C), 127.56 (Ar–C), 127.24 (Ar–C), 127.00 (Ar–C), 126.24 (Ar–C), 125.16 (Ar–C), 124.24 (Ar–C), 116.04 (Ar–C), 115.16 (Ar–C), 112.04 (Ar–C), 110.04 (C_2 of acrylate) ppm.

Sodium 3-(4,5-diphenyl-1H-imidazol-2-yl)-3-(1-hydroxynaphthalen-2-yl)acrylate (**5f**, $C_{28}H_{19}N_2NaO_3$)
Yield 68%; reddish solid; m.p.: 196–198 °C; IR (KBr): $\bar{\nu}$ = 3444, 1644, 1418 cm^{-1}; ^1H NMR (400 MHz, D_2O): δ = 8.40 (d, J = 4 Hz, 2H), 7.62 (d, J = 4 Hz, 7H), 7.55 (m, 6H), 7.42–7.28 (m, 10H), 6.90 (s, 1H) ppm; ^{13}C NMR (100 MHz, D_2O): δ = 167.22 (C_1 of acrylate), 160.44 (C_3 of acrylate), 154.00 (Ar–C), 142.22 (C_3 of imidazole), 140.38 (Ar–C), 140.20 (Ar–C), 138.88 (Ar–C), 134.24 (Ar–C), 130.82 (C_4 of imidazole), 130.02 (C_5 of imidazole), 128.52 (Ar–C), 128.76 (Ar–C), 128.54 (Ar–C), 128.30 (Ar–C), 127.31 (Ar–C), 127.20 (Ar–C), 127.19 (Ar–C), 126.16 (Ar–C), 125.18 (Ar–C), 124.17 (Ar–C), 116.32 (Ar–C), 115.12 (Ar–C), 112.44 (Ar–C), 110.23 (C_2 of acrylate) ppm; GC–MS: *m/z* calculated for $C_{28}H_{19}N_2NaO_3$ 453.13, found 454, and $C_{28}H_{20}N_2O_3$ found 432.

Antibacterial activity

The antibacterial screening of all the synthesized coumarin–imidazole hybrid compound derivatives was carried out against a broad range of pathogenic microbial test strains using broth dilution technique. The stock solutions of synthesized compounds were reconstituted with a minimum amount of dimethylsulfoxide (DMSO). This solvent did not possess any antimicrobial activity of its own. To evaluate the antimicrobial activities against four pathogenic bacterial strains, namely Gram-positive (*Bacillus flexus*) and Gram-negative (*Pseudomonas* spp.) bacteria by broth dilution method.

Antifungal activity

Antifungal activity was done by broth dilution method. For assaying antifungal activity, *Scopulariopsis* spp. and *A. terreus* strain were recultured in DMSO. A close investigation of the MIC values indicated that all the compounds exhibited a varied range of MIC of antifungal activity against all the tested fungal strains.

Antiinflammatory screening

Inflamed tissue samples were collected from clinically diagnosed patients of chronic periodontitis and chopped it completely. The patients had no background of dental treatment or antibiotic or antiinflammatory therapy for at least 3 months prior to the study. Tris buffer (5 cm^3) was centrifuged along with the inflamed tissue samples at 3000 rpm for 15 min and stored at 20 °C for additional use. It was named as MMP extract. 10 mg of each target compounds was dissolved in 1 cm^3 of DMSO separately. Hence, each mm^3 contains 10 µg of the compound. For the initial evaluation, three trials were carried out with non-identical concentrations, namely 100 µg, 250 µg and 500 µg/cm^3 and found 500 µg/cm^3 with good inhibition. So 50 mm^3 of each sample which contained 500 µg/m^3 was prepared for all samples and used. 50 mm^3 of MMP extract and 50 mm^3 of the synthesized compounds were mixed separately and then incubated for 15–30 min. 29

non-reducing buffer in equal volume was added, mixed and 20 mm^3 was loaded into each well using gel loading tips and 10 mm^3 molecular weight marker was added to last well. 50 mm^3 of a tissue sample with 0.9% normal saline is used as the control.

Electrodes were coupled and the tank was closed with cover and cables were stippled into the power supply. Initially, the gel electrophoresis apparatus was run at about 50 V for 15 min and then at 100 V until the bromophenol blue reached the bottom of the plates. After electrophoresis, the equipment was disassembled and the gel was gently removed and the equipment was washed with zymogram renaturing buffer, i.e., 2.5% Triton X-100 for 1 h to remove SDS from the gel and let the proteins denature. The zymogram renaturing buffer was removed and the gel was incubated in zymogram incubation buffer at 37 °C overnight. The gel was smeared with Coomassie blue R-250 for 1 h. Gels should be smeared with an appropriate Coomassie R-250 smearing solution for about 2 h. The background stains blue with Coomassie stain where the gelatin is degraded. An appeared white band indicates the presence of gelatinases. The upper bands are gelatinase B (MMP-9) while the lower bands are gelatinase A (MMP-2) which are examined and using multi-image gel documentation systems the percentage of inhibition was assigned. The supporting document gives the gel photograph image for the in vitro antiinflammatory results for the detection of MMP-2 and MMP-9.

Docking study

The crystal structure used (PDB ID: 4CAW; A-Chain and 1AI9) for the docking studies was obtained from the Protein Data Bank. The proteins were prepared for docking by adding polar hydrogen atom with Gasteiger–Hückel charges and water molecules were removed. The 3D structure of the ligands was generated by the SKETCH module executed in the SYBYL program (Tripos Inc., St. Louis, USA) and its energy-minimized conformation was acquired with the help of the Tripos force field using Gasteiger–Hückel charges and molecular docking was executed with Surflex-Dock program that is interfaced with Sybyl-X 2.0 and other miscellaneous parameters were assigned with the default values given by the software.

Acknowledgements The authors express their appreciation to Karnatak University, Dharwad for necessary facilities and UGC-UPE (KU/Sch/UGC-UPE/2013-14/1107) fellowship and also, grateful to the DST, New Delhi, for the financial support. Authors also thank Dr. V. H. Kulkarni, Principal, S.E.T’s College of Pharmacy, Dharwad for necessary facilities. Authors extend their thanks to Anil H. A for recording some spectral analyses at CSIR-National Chemical Laboratory, Pune.

References

1. Scott RW, DeGrado WF, Tew GN (2008) Curr Opin Biotechnol 19:620
2. Tew GN, Scott RW, Klein M, Degrado WF (2010) Acc Chem Res 43:30
3. Sharma D, Narasimhan B, Kumar P, Judge V, Narang R, DeClercq EJ (2009) Eur J Med Chem 44:2347
4. Zampieri D, Mamolo MG, Vio L, Banfi E, Scialino G, Fermeglia M, Ferrone M, Pricl S (2007) Bioorg Med Chem 15:7444
5. Olender D, Zwawiak J, Lukianchuk V, Lesyk R, Kropacz A, Fojutowski A, Zaprutko L (2009) Eur J Med Chem 44:645
6. Ozkay Y, Iskdag I, Incesu Z, Akalın G (2010) Eur J Med Chem 45:3320
7. Suzuki F, Kuroda T, Tamura T, Sato S, Ohmori K, Ichikawa S (1992) J Med Chem 35:2863
8. Puratchikodya A, Doble M (2007) Bioorg Med Chem 15:1083
9. Pinza M, Farina Z, Cerri A, Pfeiffer U, Riccaboni MT, Banfi S, Biagetti R, Pozzi O, Magnani M, Dorigotti L (1993) J Med Chem 36:4214
10. Borges F, Roleira F, Milhazes N, Santana L, Uriarte E (2005) Curr Med Chem 12:887
11. Riveiro ME, Moglioni A, Vazquez R, Gomez N, Facorro G, Piehl L, deCelis ER, Shayo C, Davio C (2008) Bioorg Med Chem 16:2665
12. Feuer G, Kellen JA, Kovacs K (1976) Oncology 33:35
13. Kashman Y, Gustafson KR, Fuller RW, Cardellina JH, McMahon JB, Currens MJ, Buckheit RW, Hughes SH, Cragg GM, Boyd MR (1993) J Med Chem 36:1110
14. Shikishima Y, Takaishi Y, Honda G, Ito M, Takfda Y, Kodzhimatov OK, Ashurmetov O, Lee KH (2001) Chem Pharm Bull 49:877
15. Gage BF (2006) Am Soc Hematol Educ Program:467
16. Ostrov DA, Hernánde-Prada JA, Corsino PE, Finton KA, Le N, Rowe TC (2007) Antimicrob Agents Chemother 51:3688
17. Gormley NA, Orphanides G, Meyer A, Cullis PM, Maxwell A (1996) Biochemistry 35:5083
18. Fylaktakidou KC, Hadjipavlou-Litina DJ, Litinas KE, Nicolaides DN (2004) Curr Pharm Des 10:3813
19. Stefanachi A, Favia AD, Nicolotti O, Leonetti F, Pisani L, Catto M, Zimmer C, Hartmann RW, Carotti A (2011) J Med Chem 54:1613
20. Ali TF, Iwamaru K, Ciftci HI, Koga R, Matsumoto M, Oba Y, Kurosaki H, Fujita M, Okamoto Y, Umezawa K, Nakao M, Hide T, Makino K, Kuratsu J, Abdel-Aziz M, Abuo-Rahma GE, Beshr EA, Otsuka M (2015) Bioorg Med Chem 23:5476
21. Beier D, Frank R, Pylori H (2000) J Bacteriol 182:2068
22. Yamamoto K, Kitayama T, Minagawa S, Watanabe T, Sawada S, Okamoto T, Utsumi R (2001) Biosci Biotechnol Biochem 65:2306
23. Deschenes RJ, Lin H, Ault AD, Fassler JS (1999) Antimicrob Agents Chemother 43:1700
24. Patricia MS (2014) Infect Drug Resist 7:229
25. Frantz DE, Morency L, Soheili A, Murry JA, Grabowski EJ, Tillyer RD (2004) Org Lett 6:843
26. Wolkenberg SE, Wisnoski DD, Leister WH, Wang Y, Zhao Z, Lindsley CW (2004) Org Lett 6:1453
27. Holiyachi M, Samundeeswari S, Chougala BM, Shastri LA (2015) Synth Commun 45:1002
28. Loffek S, Schilling O, Franzke CW (2011) Eur Respir J 38:191
29. Tetlow LC, Adlam DJ, Woolley DE (2001) Arthritis Rheum 44:585
30. Galm U, Heller S, Shapiro S, Page M, Li SM, Heide L (2004) Antimicrob Agents Chemother 48:1307
31. Sakamoto S, Sakamoto M (1988) Mol Asp Med 10:299
32. SYBYL 7.0 (2004) Tripos Inc., St. Louis, USA
33. Gasteiger J, Marsili M (1980) Tetrahedron 36:3219
34. Tripos International (2012) Sybyl-X 2.0, Tripos International, St. Louis MO, USA