European Journal of Medicinal Chemistry 45 (2010) 5460–5464

Contents lists available at ScienceDirect

European Journal of Medicinal Chemistry

journal homepage: http://www.elsevier.com/locate/ejmech

Short communication

Synthesis, characterization and anti-microbial studies of some novel 2,4-disubstituted thiazoles

A.M. Vijesh [a,b], Arun M. Isloor [b,*], Vivek Prabhu [c], Shaoib Ahmad [c], Shridhar Malladi [b]

[a] SeQuent Scientific Ltd., No: 120 A & B, Industrial Area, Baikampady, New Mangalore 575 011, Karnataka, India
[b] Organic Chemistry Division, Department of Chemistry, National Institute of Technology-Karnataka, Surathkal, Mangalore 575 025, India
[c] Department of Microbiology, Veterinary College, KVAFSU, Hebbal, Bangalore 560 002, India

ARTICLE INFO

Article history:
Received 31 March 2010
Received in revised form
21 July 2010
Accepted 27 July 2010
Available online 6 August 2010

Keywords:
Thiazoles
Pyrazoles
Anti-microbial studies

ABSTRACT

In the present study a series of novel 2,4-disubstituted thiazole derivatives containing substituted pyrazole moiety was synthesized by the reaction of 3-Aryl-1*H*-pyrazole-4-carbaldehyde thiosemicarbazone with 6-Bromo/H-3-(bromoacetyl)-2*H*-chromen-2-one/phenacyl chloride. Structures of newly synthesized compounds were characterized by spectral studies. New compounds were screened for their antibacterial studies against *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa*. The results revealed that compounds **7c**, **8e**, and **8f** having 2,5-dichlorothiophene substituent and **8c**, **8d** having 2,4-dichlorophenyl substituent showed significant antibacterial activity against all tested microorganisms.

1. Introduction

In the recent years, thiazoles and their derivatives have attracted medicinal chemists because of their varied biological activities such as recently found application in drug development for the treatment of allergies [1], hypertension [2], inflammation [3], schizophrenia [4], antibacterial [5], HIV infections [6], hypnotics [7] and more recently for the treatment of pain [8], as fibrinogen receptor antagonists with antithrombotic activity [9] and as new inhibitors of bacterial DNA gyrase B [10].

Further, pyrazole derivatives have showed significant biological activities, such as anti-microbial [11], analgesic [12], anti-inflammatory [13]. Since several years the emergence of multi-drug resistant bacteria such as methicillin-resistant *Staphylococcus aureus* or vancomycin-resistant *enterococci* and also resistant fungi is reported worldwide, increasing the complexity of anti-infective therapies. Therefore novel, effective anti-microbial drugs are urgently required. Keeping in view of this and in continuation of our search on biologically potent molecules [14–16] we hereby report the synthesis and anti-microbial property of novel, thiazoles containing pyrazole nucleus. These compounds were evaluated for their antibacterial properties against *S. aureus*, *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa* bacteria.

2. Chemistry

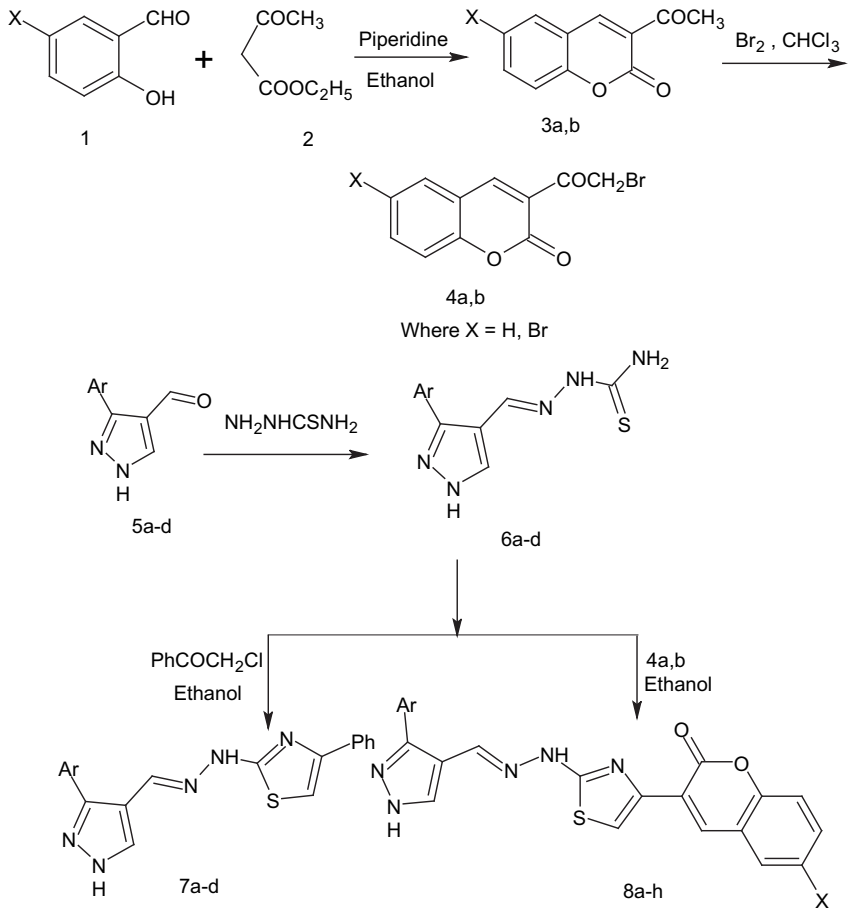
3-(Bromoacetyl)-2*H*-chromen-2-ones (**4a**,**b**) were synthesized as reported in the literature [17] and 3-substituted-1*H*-pyrazole-4-carbaldehydes (**5a**–**d**) were synthesized by the Vilsmayer Haack reaction of semicarbazones [18]. The targeted thiazoles (**7a**–**d** and **8a**–**h**) were obtained in good yield by refluxing substituted thiosemicarbazones (**6a**–**d**) with various phenacyl chlorides/bromoacetyl coumarins in ethanol for 8 h. The starting material **6a**–**d** in turn were synthesized by refluxing equimolar amount of 3-aryl-1-*H*-pyrazole-4-carbaldehyde with thiosemicarbazide in the presence of anhydrous sodium acetate in ethanol. The reaction pathway has been summarized in Scheme 1. Newly synthesized compounds (**7a**–**d** and **8a**–**h**) were characterized by IR, NMR, mass spectral and C, H, N elemental analyses.

3. Results and discussion

Formation of 3-aryl-1*H*-pyrazole-4-carbaldehyde(4-aryl-1,3-thiazol-2-yl)hydrazones was confirmed by recording their IR, ^{1}H NMR, ^{13}C NMR and mass spectra. All compounds are characterized after purification by column chromatography. IR spectrum of thiazole **7a** showed absorption at 3422 cm^{-1} which is due to the NH stretching. Band appeared at 1627 cm^{-1} is due to C=N. The C–S stretching frequency appeared at 1108 cm^{-1} further confirms the structure of the thiazole. The ^{1}H NMR spectrum of **7a** showed a singlet at δ 2.53 is due to SCH_3 protons. A doublet at δ 7.60

* Corresponding author. Tel.: +91 824 2474000; fax: +91 824 2474033.
E-mail address: isloor@yahoo.com (A.M. Isloor).

0223-5234/$ – see front matter © 2010 Elsevier Masson SAS. All rights reserved.
doi:10.1016/j.ejmech.2010.07.048



Scheme 1. Synthetic route for the 2,4-disubstituted thiazoles.

(J = 4.8 Hz) is due to aromatic protons of p-thioanisyl moiety. A singlet at δ 7.89 is due to thiazole 5H proton. Pyrazole-5H appeared as a singlet at δ 8.0. Similarly a doublet appeared at δ 7.41 (J = 4.8 Hz) is due to aromatic protons of p-thioanisyl group and doublet of doublet at δ 7.48 (J = 2.0 Hz) is due to aromatic protons of 2,4-dichlorophenyl moiety. A singlet appeared at δ 11.83 is due to pyrazole-NH protons. −N═CH Protons appeared as a singlet at δ 8.12 further confirmed the structure. The mass spectrum of **7a** showed molecular ion peak at m/z = 462 (m+), which is in agreement with the molecular formula $C_{20}H_{15}Cl_2N_5S_5$. Similarly the spectral values for all the compounds and C, H, N analyses are given in the experimental part and the characterization is provided in Table 1.

4. Anti-microbial studies

All the newly synthesized compounds were screened for their antibacterial activity. For this, *S. aureus*, *B. subtilis*, *E. coli* and *P. aeruginosa* microorganisms were employed. Anti-microbial study was assessed by Minimum Inhibitory Concentration (MIC) by serial dilution method [19]. Several colonies of *S. aureus*, *B. subtilis*, *E. coli* and *P. aeruginosa* were picked off a fresh isolation plate and inoculated in corresponding tubes containing 5 mL of trypticase soya broth. The broth was incubated for 6 h at 37 °C until there was visible growth. Mc Farland No.5 standard was prepared by adding 0.05 mL of 1% w/v $BaCl_2.2H_2O$ in Phosphate Buffered saline (PBS) to 9.95 mL of 1% v/v H_2SO_4 in PBS. The growth of all the four cultures was adjusted to Mc Farland No.5 turbidity standard using sterile PBS. This gives a 108 cfu/mL suspension. The working inoculums of aforementioned four different microorganisms containing 105 cfu/mL suspension was prepared by diluting the 108 cfu/mL suspension, 103 times in trypticase soya broth.

4.1. Preparation of anti-microbial suspension (1 mg/mL)

Dissolved 10 mg of each compound in 10 mL of Dimethyl formamide to get 1 mg/mL concentration.

4.2. Preparation of dilutions

In all, for each of the 12 anti-microbial compounds and standard anti-microbial i.e. Ceftriaxone, 24 tubes of 5 mL capacity were arranged in 4 rows with each row containing 6 tubes. Then 1.9 mL of trypticase soya broth was added in the first tube in each row and then 1 mL in the remaining tubes. Now, 100 μL of anti-microbial suspension dissolved in Dimethyl formamide was added to the first tube in each row and then after mixing the content, 1 mL was serially transferred from these tubes to the second tube in each of the rows. Then the contents in the second tube of each of the rows were mixed and transferred to the third tube in each of the rows. This serial dilution was repeated till the sixth tube in each of the rows. This provided anti-microbial concentrations of 50, 25, 12.5, 6.25, 3.125, 1.6125 μg/mL in the first to sixth tube respectively in each row. Finally, 1 mL of 10^5 cfu/mL of *S. aureus*, *B. subtilis*, *E. coli* and *P. aeruginosa* suspension were added to the first, second, third and fourth rows of tubes respectively. Along with the test samples

Table 1
Characterization data of the compounds **7a–d** and **8a–h**.

Comp-ounds	Ar	Ph or X	Molecular formula (Mol. wt.)	Yield (%)	M.p. (°C)
7a	4-SCH_3–C_6H_4	2,4-Dichlorophenyl	$C_{20}H_{15}Cl_2N_5S_2$ (461)	82	220–222
7b	2,4-Dichlorophenyl	2,4-Dichlorophenyl	$C_{19}H_{11}Cl_4N_5S$ (483)	85	210–212
7c	2,5-dichlorothiophene	2,4-Dichlorophenyl	$C_{18}H_9Cl_4N_5S_2$ (501)	78	236–238
7d	Biphenyl	2,4-Dichlorophenyl	$C_{25}H_{17}Cl_2N_5S$ (491)	81	226–228
8a	4-SCH_3–C_6H_4	H	$C_{23}H_{17}N_5O_2S_2$ (459)	83	202–204
8b	4-SCH_3–C_6H_4	Br	$C_{23}H_{16}BrN_5O_2S_2$ (539)	76	244–246
8c	2,4-Dichlorophenyl	H	$C_{22}H_{13}Cl_2N_5O_2S$ (483)	84	250–252
8d	2,4-Dichlorophenyl	Br	$C_{21}H_{12}BrCl_2N_5O_2S$ (549)	77	240–242
8e	2,5-dichlorothiophene	H	$C_{20}H_{11}Cl_2N_5O_2S_2$ (489)	79	218–220
8f	2,5-dichlorothiophene	Br	$C_{20}H_{10}BrCl_2N_5O_2S$ (567)	75	234–236
8g	Biphenyl	H	$C_{28}H_{19}N_5O_2S$ (489)	80	238–240
8h	Biphenyl	Br	$C_{28}H_{18}BrN_5O_2S$ (569)	76	254–256

and Ceftriaxone (standard), the inoculums control (without anti-microbial compound) and broth control (without anti-microbial compound and inoculum) were maintained. All the test sample and control tubes were then incubated for 16 h at 37 °C.

4.3. Interpretation

After incubation, the tubes showing no visible growth were considered to be representing the MIC. The details of results are furnished in Table 2. Inoculums control showed visible growth, where as the broth control showed no growth.

5. Conclusion

A series of novel 2,4-disubstituted thiazole derivatives were synthesized in reasonably good yields. They were characterized by ^{1}H NMR, ^{13}C NMR, mass spectrometry, IR studies and elemental analyses. All the newly synthesized compounds were screened for antibacterial activity by MIC method. Among the screened samples, **8e** and **8f** have showed excellent antibacterial activity at 1.6125 mg/mL concentration against *S. aureus* bacteria as compared to the standard drug Ceftriaxone which is active at 3.125 mg/mL concentration. They also showed similar activity as that of standard, against *B. subtilis*, *E. coli* and *P. aeruginosa,* at 1.6125 mg/mL concentration.

As regards the relationships between the structure of the heterocyclic scaffold and the detected antibacterial properties, it showed varied biological activity. Probably in this case the nature of the heterocyclic ring is not so important for anti-microbial activity. Moreover, the presence of different substituents causes a certain change of activity. Compounds **8e** and **8f** have 2,5-dichlorothiophene moiety, which is accounted for the enhanced antibacterial activity against all the four tested microorganisms. Similarly compounds **7c**, **8c** and **8d** have showed significant activity against the four bacteria, which are active at the same concentration as that of the standard drug. Compound **7c** has 2,5-dichlorothiophene and **8c** and **8d** have 2,4-Dichlorophenyl subsistent respectively, which is accounted for the activity of the compounds. On the other hand, for the remaining compounds have not showed any activity against any of the four tested bacterial strains. From the obtained results, it is clear that the major role for antibacterial activity is played by the substituent present on pyrazole and aromatic rings bonded to thiazole moiety. It is evident that thiazoles with 2,5-dichlorothiophene and 2,4-Dichlorophenyl are the most active compounds.

Table 2
Antibacterial activity data in MIC (μg/mL).

Comp. No.	*S. aureus*	*B.subtilis*	*E.coli*	*P.aeruginosa*
7a	***	50.00	50.00	50.00
7b	3.125	***	50.00	***
7c	3.125	1.6125	1.6125	1.6125
7d	***	***	***	***
8a	***	***	***	***
8b	***	***	***	***
8c	3.125	1.6125	1.6125	1.6125
8d	3.125	3.125	1.6125	1.6125
8e	1.6125	1.6125	3.125	1.6125
8f	1.6125	1.6125	1.6125	1.6125
8g	***	***	***	***
8h	***	***	***	***
Ceftriaxone (Standard)	3.125	1.6125	1.6125	1.6125
Inoculum control	***	***	***	***
Broth control	No growth	No growth	No growth	No growth

*** Indicates growth in all concentrations.

6. Experimental

6.1. Chemistry

Melting points were determined by open capillary method and were uncorrected. The IR spectra (in KBr pellets) were recorded on a JASCO FT/IR-4100 spectrophotometer. ^{1}H NMR and ^{13}C NMR spectra were recorded (DMSO-d_6) on a Bruker (400 MHz) and Varian (400 MHz) spectrometer using TMS as internal standard. Chemical shift values are given in δ scales. The mass spectra were recorded on a JEOL JMS-D 300 spectrometer operating at 70 eV. Elemental analyses were performed on a Flash EA 1112 series CHNS-O Analyzer. The completion of the reaction was checked by thin layer chromatography (TLC) on silica gel coated aluminium sheets (silica gel 60 F254). Commercial grade solvents and reagents were used without further purification.

6.2. General procedure for the synthesis of 3-acetyl-6-H/Br-2H-chromen-2-one (*3a–b*)

A mixture of salicylaldehyde/bromosalicylaldehyde (0.5 mol) and ethylacetoacetate (0.5 mol) were stirred in 20 ml of ethanol and cooled. To this mixture 10 g of piperidine was added with shaking. The mixture was maintained at freezing temperature for 2–3 h, which resulted in yellow colored solid mass, which was separated out. It was recrystalised from ethanol to get the target compounds.

6.3. General procedure for the synthesis of 6-Bromo/H-3-(bromoacetyl)-2H-chromen-2-one (*4a,b*)

To a solution of compound 3a-b (0.25 mol) in 200 ml of chloroform, bromine (0.25 mol) in 25 mL of chloroform was added

under stirring [20]. The mixture was stirred for 1 h at room temperature and then warmed to decompose the addition product. The mixture was heated to 55 °C for 15 min, cooled and filtered to get a solid mass, further which was washed with diethyl ether to get the desired product. It was recrystalised from acetic acid to give pale yellow colored needles.

6.3.1. Characterization of synthesized compounds

*6.3.1.1. 3-(2-Bromoacetyl)-2H-chromen-2-one (**4a**).* ^{1}H NMR (DMSO-d_6): δ 4.41 (s, 2H, $COCH_2$), 7.4–7.8 (m, 4H, aromatic H), 8.13 (s, 1H, Coumarin 4H).

*6.3.1.2. 6-Bromo-3-(2-bromoacetyl)-2H-chromen-2-one (**4b**).* ^{1}H NMR (DMSO-d_6): δ 4.43 (s, 2H, $COCH_2$), 7.31–7.62 (m, 2H, Coumarin 7H, 8H), 8.12 (s, 1H, Coumarin 4H), 8.19 (s, 1H, coumarin 5H).

*6.4. General procedure for the synthesis of 3-aryl-1H-pyrazole-4-carbaldehyde thiosemicarbazone (**6a–d**)*

An equimolar mixture of 3-aryl-1*H*-pyrazole-4-carbaldehyde (0.143 mol) and thiosemicarbazide (0.143 mol) in ethanol (25 mL) and sodium acetate (0.07 mol) was refluxed for 8 h. The solid separated was filtered, dried and recrystalised using ethanol–DMF mixture.

6.5. Characterization of synthesized compounds

*6.5.1. 3-(2,4-Dichlorophenyl)-1H-pyrazole-4-carbaldehyde thiosemicarbazone (**6a**)*

IR (KBr, υ_{max} cm^{-1}): 3451, 3324 (N–H-str), 1603 (C=N), 1537 (C=C), 1109 (C=S), 948, 835 (C–Cl). Yield 82%, M.p. 212–214 °C.

*6.5.2. 3-[4-(Methylthio)phenyl]-1H-pyrazole-4-carbaldehyde thiosemicarbazone (**6b**)*

IR (KBr, υ_{max} cm^{-1}): 3424, 3282 (N–H-str), 1591 (C=N), 1559 (C=C), 1128 (C=S). Yield 76%, M.p. 238–240 °C.

*6.5.3. 3-(2,5-Dichloro-3-thienyl)-1H-pyrazole-4-carbaldehyde thiosemicarbazone (**6c**)*

IR (KBr, υ_{max} cm^{-1}): 3432, 3272 (N–H-str), 3030 (C–H-str), 1606 (C=N), 1559 (C=C), 1036 (C=S), 945, 823, 706 (C–Cl). Yield 78%, M.p. 228–230 °C.

*6.5.4. 3-Biphenyl-4-yl-1H-pyrazole-4-carbaldehyde thiosemicarbazone (**6d**)*

IR (KBr, υ_{max} cm^{-1}): 3471, 3309 (N–H-str), 3046 (C–H-str), 1590 (C=N), 1576 (C=C), 1104 (C=S). Yield 81%, M.p. 234–236 °C.

*6.6. General procedure for the synthesis of 3-aryl-1H-pyrazole-4-carbaldehyde (4-aryl-1,3-thiazol-2-yl)hydrazone (**7a–d** and **8a–h**)*

An equimolar mixture of 3-Aryl-1*H*-pyrazole-4-carbaldehyde thiosemicarbazone 4a-d (0.01 mol) and substituted phenacyl bromide/chlorides (0.01 mol) in ethanol was refluxed for 4 h. After completion of the reaction, the reaction mixture was allowed to cool. The solid thus separated was collected by filtration and recrystalised using ethanol–DMF mixture.

6.7. Characterization of synthesized compounds

*6.7.1. 3-[4-(Methylthio)phenyl]-1H-pyrazole-4-carbaldehyde [4-(2,4-dichlorophenyl)-1,3-thiazol-2-yl]hydrazone (**7a**)*

IR (KBr, υ_{max} cm^{-1}): 3422 (N–H-str), 2852 (C–H-str), 1627(C=N), 1579 (C=C), 1108 (C–S), 956, 838 (C–Cl); ^{1}H NMR (DMSO-d_6): δ 2.53 (s, 3H, SCH_3), 7.36 (s, 1H, 2,4-dichlorophenyl), 7.41 (d, 2H, J = 4.8 Hz, p-thioanisyl), 7.48 (dd, 1H, J = 2.0 Hz, 2,4-dichlorophenyl), 7.6 (d, 2H, J = 4.8 Hz, p-thioanisyl), 7.68 (d, 1H, J = 2.4 Hz, 2,4-dichlorophenyl), 7.89 (s, 1H, thizole-5H), 8.0 (s, 1H, pyrazole-5H), 8.12 (s, 1H, N=CH), 11.83 (s, 1H, pyrazole-NH); MS (*m*/*z*, %): 462.0 (M^+, 64), 464.0 (M + 2), 466.0 (M + 4). Anal. calcd. for $C_{20}H_{15}Cl_2N_5S_2$: C, 52.06; H, 3.25; N, 15.18. Found: C, 52.04; H, 3.23; N, 15.20%

*6.7.2. 3-(2,4-Dichlorophenyl)-1H-pyrazole-4-carbaldehyde [4-(2,4-dichlorophenyl)-1,3-thiazol-2-yl]hydrazone (**7b**)*

IR (KBr, υ_{max} cm^{-1}): 3420 (N–H-str), 2923 (C–H-str), 1644(C=N), 1575 (C=C), 1105 (C–S), 1029 (C–Cl); ^{1}H NMR (DMSO-d_6): δ 7.26–7.77 (m, 6H, 2,4-dichlorophenyl), 7.84 (s, 1H, thizole-5H), 7.86 (s, 1H, pyrazole-5H), 8.08 (s, 1H, N=CH), 11.75 (s,1H, pyrazole-NH); MS (*m*/*z*, %): 484.0 (M^+, 100), 486.0 (M + 2), 488.0 (M + 4), 490.0 (M + 6), 492.0 (M + 8). Anal. calcd. for $C_{19}H_{11}Cl_4N_5S$: C, 47.20; H, 2.28; N, 14.49. Found: C, 47.16; H, 2.26; N, 14.45%.

*6.7.3. 3-(2,5-Dichloro-3-thienyl)-1H-pyrazole-4-carbaldehyde [4-(2,4-dichlorophenyl)-1,3-thiazol-2-yl]hydrazone (**7c**)*

IR (KBr, υ_{max} cm^{-1}): 3443 (N–H-str), 2931 (C–H-str), 1631(C=N), 1573 (C=C), 1111 (C–S), 948, 816, 735 (C–Cl); ^{1}H NMR (DMSO-d_6): δ 7.31 (s, 1H, 2,4-dichlorophenyl), 7.46 (d, 1H, J = 2.0 Hz 2,4-dichlorophenyl), 7.48 (d, 1H, J = 2.4 Hz, 2,4-dichlorothiophene), 7.65 (d, 1H, J = 2.0 Hz, 2,4-dichlorophenyl), 7.85 (s, 1H, thizole-5H), 7.97 (s, 1H, pyrazole-5H), 8.09 (s, 1H, N=CH), 11.88 (s, 1H, pyrazole-NH); ^{13}C NMR: 167.2, 145.5, 135.1, 132.4, 132.1, 132.0, 131.5, 129.6, 128.9, 127.3, 124.8, 123.7, 115.3, 108.6. MS (*m*/*z*, %): 502.0 (M^+, 100), 504.0 (M + 2), 506.0 (M + 4), 508.0 (M + 6), 510.0 (M + 8). Anal. calcd. for $C_{18}H_9Cl_4N_5S_2$: C, 43.11; H, 1.79; N, 13.97. Found: C, 43.10; H, 1.75; N, 13.94%.

*6.7.4. 3-Biphenyl-4-yl-1H-pyrazole-4-carbaldehyde [4-(2,4-dichlorophenyl)-1,3-thiazol-2-yl]hydrazone (**7d**)*

IR (KBr, υ_{max} cm^{-1}): 3419 (N–H-str), 2924 (C–H-str), 1626(C=N), 1573 (C=C), 1104 (C–S), 953, 838, 729 (C–Cl); ^{1}H NMR (DMSO-d_6): δ 7.34 (s, 1H, 2,4-dichlorophenyl), 7.39–7.51 (m, 4H, 2,4-dichlorophenyl, biphenyl), 7.66 (d, 1H, J = 2.4 Hz, 2,4-dichlorophenyl), 7.73–7.82 (m, 7H,biphenyl), 7.88 (s, 1H, thizole-5H), 8.02 (s, 1H, pyrazole-5H), 8.18 (s, 1H, N=CH), 11.87 (s, 1H, pyrazole-NH); ^{13}C NMR: 167.2, 139.9, 139.4, 136.0, 132.3, 132.1, 132.0, 131.5, 129.6, 128.9, 128.6, 127.6, 127.4, 126.8, 126.6, 113.9, 108.7. MS÷ (*m*/*z*, %): 492.0 (M^+, 60), 494.0 (M + 2), 496.0 (M + 4). Anal. calcd. for $C_{25}H_{17}Cl_2N_5S$: C, 61.10; H, 3.46; N, 14.26. Found: C, 61.07; H, 3.44; N, 14.23%.

*6.7.5. **3**-[4-(Methylthio)phenyl]-1H-pyrazole-4-carbaldehyde [4-(2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8a**)*

IR (KBr, υ_{max} cm^{-1}): 3311 (N–H-str), 2920 (C–H-str), 1728(C=O α-Pyrone), 1605(C=N), 1562 (C=C); 1095 (C–S), ^{1}H NMR (DMSO-d_6): δ 2.53 (s, 3H, SCH_3), 7.29–7.75 (m, 9H, coumarin, p-anisyl), 7.92 (s, 1H, thizole-5H), 8.09 (s, 1H, pyrazole-5H), 8.46 (s, 1H, N=CH), 11.79 (s, 1H, pyrazole-NH); MS (*m*/*z*, %): 460.0 (M^+, 100). Anal. calcd. for $C_{23}H_{17}N_5O_2S_2$: C, 60.13; H, 3.70; N, 15.25. Found: C, 60.12; H, 3.68; N, 15.23%.

*6.7.6. 3-[4-(Methylthio)phenyl]-1H-pyrazole-4-carbaldehyde [4-(6-bromo-2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8b**)*

IR (KBr, υ_{max} cm^{-1}): 3320 (N–H-str), 2919 (C–H-str), 1731(C=O α-Pyrone), 1631(C=N), 1573 (C=C); 1096 (C–S), ^{1}H NMR (DMSO-d_6): δ 2.53 (s, 3H, SCH_3), 7.36 (s, 1H, coumarin), 7.40 (d, 2H, J = 4.4 Hz p-thioanisyl), 7.59 (d, 2H, J = 4.8 Hz, p-thioanisyl), 7.72–7.75 (m, 2H, coumarin), 7.79 (s, 1H, thizole-5H), 8.12 (s, 1H, coumarin 4H), 8.13 (s, 1H, pyrazole-5H), 8.45 (s, 1H, N=CH), 11.84

(s,1H, pyrazole-NH); ^{13}C NMR: 167.5, 136.5, 133.7, 130.6, 128.5, 125.9, 121.1, 118.0, 111.0, 14.5. MS (*m/z*, %): 540.0 (M^+, 82), 542.0 (M + 2). Anal. calcd. for $C_{23}H_{16}BrN_5O_2S_2$: C, 51.21; H, 2.97; N, 12.99. Found: C, 51.18; H, 2.95; N, 12.96%.

*6.7.7. 3-(2,4-Dichlorophenyl)-1H-pyrazole-4-carbaldehyde [4-(2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8c**)*

IR (KBr, υ_{max} cm^{-1}): 3445 (N–H-str), 2925 (C–H-str), 1722 (C=O α-Pyrone), 1633(C=N), 1558 (C=C); 1099 (C-S), ^{1}H NMR (DMSO-d_6): δ 7.34–7.47 (m, 3H, coumarin, 2,4-dichlorophenyl), 7.55 (dd, 1H, *J* = 2.0 Hz 2,4-dichlorophenyl), 7.60–7.63 (m, 2H, coumarin), 7.76 (d, 1H, *J* = 2.0 Hz 2,4-dichlorophenyl), 7.81–7.83 (dd, 1H, *J* = 6.4 Hz coumarin), 7.88 (s, 1H, thizole-5H), 8.08 (s, 1H, pyrazole-5H), 8.47 (s, 1H, N=CH), 11.78 (s,1H, pyrazole-NH); ^{13}C NMR: 167.5, 159.0, 152.4, 138.0, 135.2, 134.0, 133.2, 131.5, 129.1, 128.7, 127.2, 124.6, 120.4, 115.8, 115.4, 110.0. MS (*m/z*, %): 484.0 (M^+, 68), 486.0 (M + 2), 488.0 (M + 4). Anal. calcd. for $C_{22}H_{13}Cl_2N_5O_2S$: C, 54.66; H, 2.69; N, 14.49. Found: C, 54.64; H, 2.66; N, 14.47%.

*6.7.8. 3-(2,4-Dichlorophenyl)-1H-pyrazole-4-carbaldehyde [4-(6-bromo-2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8d**)*

IR (KBr, υ_{max} cm^{-1}): 3420 (N–H-str), 1715 (C=O α-Pyrone), 1627 (C=N), 1576 (C=C); 1105 (C–S), ^{1}H NMR (DMSO-d_6): δ 7.38–7.77 (m, 7H, coumarin, 2,4-dichlorophenyl), 7.88 (s, 1H, thizole-5H), 8.07 (s, 1H, pyrazole-5H), 8.11 (d, 1H, *J* = 2.0 Hz coumarin), 8.41 (s, 1H, N=CH), 11.77 (s, 1H, pyrazole-NH); ^{13}C NMR: 167.6, 158.2, 151.2, 143.5, 136.5, 135.3, 133.9, 133.2, 130.6, 129.1, 127.2, 121.4, 121.1, 118.0, 116.2, 115.3, 110.8. MS (*m/z*, %): 550.0 (M^+, 100), 552.0 (M + 2), 554.0 (M + 4), 556.0 (M + 6). Anal. calcd. for $C_{21}H_{12}BrCl_2N_5O_2S$: C, 45.90; H, 2.18; N, 12.79. Found: C, 45.87; H, 2.15; N, 12.76%.

*6.7.9. 3-(2,5-Dichloro-3-thienyl)-1H-pyrazole-4-carbaldehyde [4-(2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8e**)*

IR (KBr, υ_{max} cm^{-1}): 3420 (N–H-str), 1698 (C=O α-Pyrone), 1636 (C=N), 1576 (C=C); 1111 (C–S), ^{1}H NMR (DMSO-d_6): δ 7.2–7.8 (m, 6H, coumarin, 2,5-dichlorothiophene), 7.90 (s, 1H, thizole-5H), 8.02 (s, 1H, pyrazole-5H), 8.42 (s, 1H, N=CH), 11.78 (s, 1H, pyrazole-NH); ^{13}C NMR: 167.5, 158.7, 152.2, 138.0, 135.2, 131.6, 128.9, 128.7, 124.9, 124.6, 120.5, 119.1, 115.8, 115.3, 110.1, 85.6. MS (*m/z*, %): 490.0 (M^+, 100), 492.0 (M + 2), 494.0 (M + 4). Anal. calcd. for $C_{20}H_{11}Cl_2N_5O_2S_2$: C, 49.08; H, 2.25; N, 14.31. Found: C, 49.07; H, 2.23; N, 14.28%.

*6.7.10. 3-(2,5-Dichloro-3-thienyl)-1H-pyrazole-4-carbaldehyde [4-(6-bromo-2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8f**)*

IR (KBr, υ_{max} cm^{-1}): 3316 (N–H-str), 2924 (C–H-str), 1715 (C=O α-Pyrone), 1629(C=N), 1573 (C=C), 1109 (C–S), 949, 816 (C–Cl); ^{1}H NMR (DMSO-d_6): δ 7.20–7.65 (m, 3H, coumarin), 7.67 (d, 1H, *J* = 2.4 Hz 2,5-dichlorothiophene), 7.91 (s, 1H, thizole-5H), 8.02–8.04 (m, 2H, coumarin), 8.36 (s, 1H, pyrazole-5H), 8.86 (s, 1H, N=CH), 11.80 (s, 1H, pyrazole-NH); ^{13}C NMR: 167.5, 158.2, 151.2, 143.5, 136.5, 135.3, 133.7, 130.6, 128.9, 124.9, 121.4, 121.1, 118.0, 116.2, 15.3, 110.9. MS (*m/z*, %): 567.9 (M^+, 27), 570.0 (M + 2), 572.0 (M + 4), 574.0 (M + 6). Anal. calcd. for $C_{20}H_{10}BrCl_2N_5O_2S_2$: C, 42.33; H, 1.76; N, 12.35. Found: C, 42.31; H, 1.75; N, 12.33%.

*6.7.11. 3-(Biphenyl)-1H-pyrazole-4-carbaldehyde [4-(2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8g**)*

IR (KBr, υ_{max} cm^{-1}): 3420 (N–H-str), 2923 (C–H-str), 1729 (C=O α-Pyrone), 1633 (C=N), 1564 (C=C), 1100 (C–S); ^{1}H NMR (DMSO-d_6): δ 7.35–7.81 (m, 14H, coumarin, biphenyl), 8.01 (s, 1H, thizole-5H), 8.17 (s, 1H, pyrazole-5H), 8.43 (s, 1H, N=CH), 11.84 (s, 1H, pyrazole-NH); MS (*m/z*, %): 490.2 (M^+, 100). Anal. calcd. for $C_{28}H_{19}N_5O_2S$: C, 68.71; H, 3.88; N, 14.31. Found: C, 68.69; H, 3.86; N, 14.30%.

*6.7.12. 3-(Biphenyl)-1H-pyrazole-4-carbaldehyde [4-(6-bromo-2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]hydrazone (**8h**)*

IR (KBr, υ_{max} cm^{-1}): 3326 (N–H-str), 1733 (C=O α-Pyrone), 1633 (C=N), 1574 (C=C), 1105 (C–S); ^{1}H NMR (DMSO-d_6): δ 7.37–7.82 (m, 12H, coumarin, biphenyl), 8.02 (s, 1H, thizole-5H), 8.11 (d, 1H, *J* = 2.4 Hz coumarin), 8.20 (s, 1H, pyrazole-5H), 8.45 (s, 1H, N=CH), 11.88 (s, 1H, pyrazole-NH); ^{13}C NMR: 167.6, 158.2, 151.2, 143.5, 139.9, 139.4, 136.5, 136.2, 133.7, 130.6, 129.8, 128.9, 128.6, 127.6, 126.8, 126.6, 121.4, 121.1, 118.0, 116.2, 113.9, 111.0. MS (*m/z*, %): 570.0 (M^+, 82), 572.0 (M + 2). Anal. calcd. for $C_{28}H_{18}BrN_5O_2S$: C, 59.05; H, 3.16; N, 12.30. Found: C, 59.03; H, 3.14; N, 12.27%.

Acknowledgements

AMI thank Prof. A.V. Adhikari, HOD Chemistry and Prof. Sandeep Sancheti, Director, National Institute of Technology-Karnataka, India for providing the research facilities. Also thank Board for research in Nuclear Sciences, Government of India for 'Young Scientist' award. AMV thankful to the management, SEQUENT SCIENTIFIC LTD, New Mangalore, India for their invaluable support and allocation of resources for this work. The authors are also thankful to Head, NMR Research center, IISc, Bangalore for providing spectral data.

References

[1] K.D. Hargrave, F.K. Hess, J.T. Oliver, J. Med. Chem. 26 (1983) 1158–1163.
[2] (a) S. Bondock, W. Khalifa, A.A. Fadda, Eur. J. Med. Chem. 42 (2007) 948–954; (b) B.A. Steinbaugh, B.L. Batley, C.A. Painchaud, S.T. Rapundalo, B.M. Michniewicz, S.C.J. Olson, J. Med. Chem. 35 (1992) 2562–2572.
[3] P.K. Sharma, S.N. Sawnhney, A. Gupta, G.B. Singh, S. Bani, Indian J. Chem. 37B (1998) 376–381.
[4] J.C. Jaen, L.D. Wise, B.W. Caprathe, H. Tecle, S. Bergmeier, C.C. Humblet, T.G. Heffner, L.T. Meltzner, T.A. Pugsley, J. Med. Chem. 33 (1990) 311–317.
[5] K. Tsuji, H. Ishikawa, Bioorg. Med. Chem. Lett. 4 (1994) 1601–1606.
[6] F.W. Bell, A.S. Cantrell, M. Hogberg, S.R. Jaskunas, N.G. Johansson, C.L. Jordon, M.D. Kinnick, P. Lind, J.M. Morin Jr., R. Noreen, B. Oberg, J.A. Palkowitz, C.A. Parrish, P. Pranc, C. Sahlberg, R.J. Ternansky, R.T. Vasileff, L. Vrang, S.J. West, H. Zhang, X.X. Zhou, J. Med. Chem. 38 (1995) 4929–4936.
[7] N. Ergenc, G. Capan, N.S. Günay, S. Özkirimli, M. Güngör, S. Özbey, E. Kendi, Arch. Pharm. Pharm. Med. Chem. 332 (1999) 343–347.
[8] J.S. Carter, S. Kramer, J.J. Talley, T. Penning, P. Collins, M.J. Graneto, K. Seibert, C. Koboldt, J. Masferrer, B. Zweifel, Bioorg. Med. Chem. Lett. 9 (1999) 1171–1174.
[9] A. Badorc, M.F. Bordes, P. De Cointet, P. Savi, A. Bernat, A. Lale, M. Petitou, J.P. Maffrand, J.M. Herbert, J. Med. Chem. 40 (1997) 3393–3401.
[10] J. Rudolph, H. Theis, R. Hanke, R. Endermann, L. Johannsen, F.U. Geschke, J. Med. Chem. 44 (2001) 619–626.
[11] A.M. Isloor, B. Kalluraya, P. Shetty, Eur. J. Med. Chem. 44 (2009) 3784–3787.
[12] A.M. Isloor, B. Kalluraya, M. Rao, J. Saudi Chem. Soc. 4 (3) (2000) 265–270.
[13] B. Kalluraya, A.M. Isloor, P.V. Frank, R.L. Jagadesha, S. Shenoy, Indian J. Heterocycl. Chem. (Oct–Dec 2001) 159–162.
[14] A.M. Isloor, B. Kalluraya, K.S. Pai, Eur. J. Med. Chem. 45 (2010) 825–830.
[15] B. Chandrakantha, P. Shetty, V. Nambiyar, N. Isloor, A.M. Isloor, Eur. J. Med. Chem. 45 (2010) 1206–1210.
[16] U.S. Rai, A.M. Isloor, P. Shetty, A.M. Vijesh, N. Prabhu, S. Isloor, M. Thiageeswaran, H.K. Fun, Eur. J. Med. Chem. 45 (2010) 2695–2699.
[17] S.K. Sahu, A. Mishra, R.K. Behrea, Indian J. Heterocycl. Chem. 6 (1996) 91.
[18] P.G. Baraldi, C. Barbara, S. Giampiero, R. Romeo, B. Giovanni, N.Z. Abdel, J. Maria, D.L. Pineda, Synthesis (1998) 1140.
[19] Mackie, Mc. Cartney, Practical Medical Microbiology, 13th ed. Churchill Livingstone, Edinburgh, 1989.
[20] B. Kalluraya, P. Gunaga, M.V. Ramana, Indian J. Heterocycl. Chem. 8 (1999) 241–244.