Available online at www.sciencedirect.com
ScienceDirect
European Journal of Medicinal Chemistry 41 (2006) 1048–1058
EUROPEAN JOURNAL OF MEDICINAL CHEMISTRY
http://france.elsevier.com/direct/ejmech

Original article

Heterocyclic system containing bridgehead nitrogen atom: synthesis and pharmacological activities of some substituted 1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles

V. Mathew[a], J. Keshavayya[b,*], V.P. Vaidya[b]

[a] *Acharya and B.M. Reddy College of Pharmacy, Bangalore, Karnataka, India*
[b] *Department of Studies in Chemistry, Kuvempu University, Jnana Sahayadri, Shankaraghatta, 577451 Shimoga, Karnataka, India*

Received in revised form 6 March 2006; accepted 9 March 2006
Available online 05 July 2006

Abstract

Several 3,6-disubstituted-1,2,4-triazolo[3,4-b]-1,3,4-thiadizoles were prepared by the condensation of 4-amino-3-aryl/aralkyl substituted-5-mercapto-1,2,4-triazoles **3**(**a**–**c**) with various substituted aromatic/hetero aromatic acids through a single step reaction. Elemental analysis, IR, ^{1}H NMR and mass spectral data confirmed the structure of the newly synthesized compounds. Synthesized triazolo thiadiazoles investigated for their antibacterial, antifungal, anti-inflammatory and analgesic activities. Some of the tested compounds showed moderate antimicrobial activity against various tested bacterial and fungal strains. None of the synthesized compounds have significant anti-inflammatory and analgesic activities.

Keywords: 4-Amino triazoles; Triazolo thiadiazoles; Phosphorous oxychloride; Spectral characterization; Pharmacological activities

1. Introduction

The recent literature is enriched with progressive findings about the synthesis and pharmacological action of fused heterocycles. Heterocycles bearing a symmetrical triazole or 1,3,4-thiadiazole moiety are reported to show a broad spectrum of pharmacological properties such as anti-inflammatory [1,2], antiviral [3] and antibacterial [4,5] activities. A survey of literature revealed that s-triazolo[3,4-b]-1,3,4-thiadiazole rings, have received much attention during recent years on account of their prominent utilization as antifungal [6], anti-inflammatory [7], antiviral, analgesic [8], anthelmintic [9] and antibacterial agents [10].

A triazolo thiadiazole system may be viewed as a cyclic analogue of two very important components—thiosemicarbazide [11,12] and biguanide [13], which often display diverse biological activities. Therefore it was planned to investigate a composite system, which combine these two biolabile components in a ring together to give a compact and planar structure and screened for their biological activities.

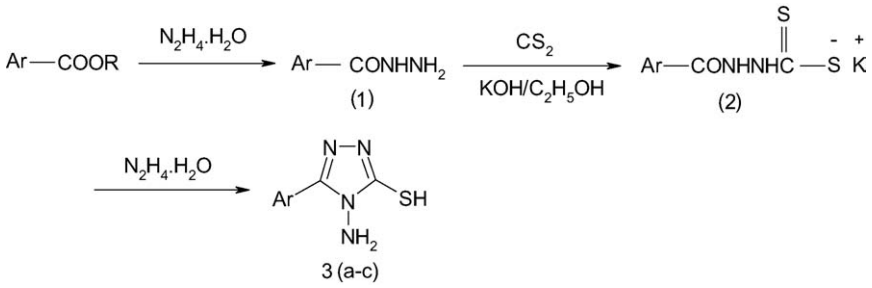
Prompted by these observations, we report herein the reaction of 4-amino-3-aryl/aralkyl substituted-5-mercapto-1,2,4-triazoles **3**(**a**–**c**) with aromatic acids in the presence of phosphorous oxychloride to give 3,6-disubstituted-1,2,4-triazolo [3,4-b]-1,3,4-thiadiazoles **4**–**11**(**a**–**c**) and their biological activities (Scheme 2).

Phosphorous oxychloride was necessary for this condensation, which activate the carbonyl group of aromatic acids and increases its electrophilicity to enhance the addition of triazole to it.

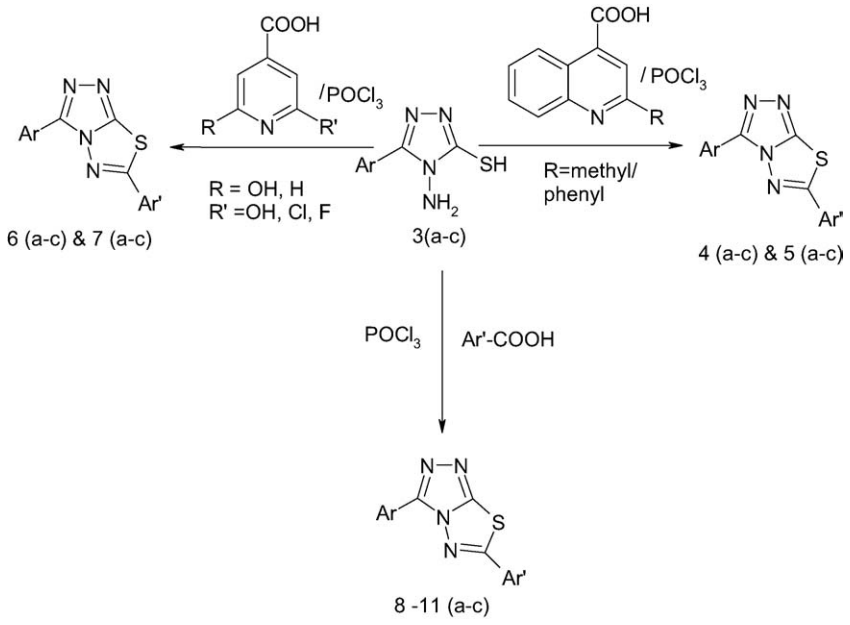
The required 4-amino-3-aryl/aralkyl substituted-5-mercapto-1,2,4-triazoles **3**(**a**–**c**) were prepared in good yield through multi step reaction by using the method of Reid and Heindel [14] with suitable modifications (Scheme 1). The structure of the intermediate triazole derivatives was based on their elemental analysis and other spectral data. The synthesis of 3,6-disubstituted-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles was accomplished in a single step by reacting 4-amino-3-aryl/aralkyl substituted-5-mercapto-1,2,4-triazoles **3**

* Corresponding author.
E-mail address: jkeshavayya@rediffmail.com (J. Keshavayya).

0223-5234/$ - see front matter © 2006 Elsevier SAS. All rights reserved.
doi:10.1016/j.ejmech.2006.03.018



Scheme 1. Heterocyclic system containing bridgehead nitrogen atom: synthesis and pharmacological activities of some substituted 1,2,4-triazolo [3,4-b]-1,3,4-thiadiazoles.



Scheme 2.

(**a**–**c**) with various aromatic acids in the presence of phosphorous oxychloride as per Scheme 2.

2. Chemistry

As depicted in scheme, in addition to triazolo thiadiazoles, we have synthesized three substituted 1,2,4-triazoles through a multiple step reaction (Scheme 1). Aromatic esters were hydrazinolysized to give aroyl hydrazides (**1**), which reacted with carbon disulfide and potassium hydroxide in ethanol to yield potassium dithiocarbazinate (**2**), which later cyclized to 4-amino-5-mercapto-1,2,4-triazole **3**(**a**–**c**) by reacting with hydrazine hydrate. The resulted triazoles **3**(**a**–**c**) further converted to triazolo thiadiazoles (**4**–**11**) by one pot reaction by the condensation with aromatic/hetero aromatic acids in the presence of phosphorous oxychloride (Scheme 2). This procedure afforded various triazolo thiadiazoles in 50–65% yields. The infrared spectra of the substituted-4-amino-5-mercapto-1,2,4-triazole (**3a**–**c**) showed two characteristic absorption bands, one of which appearing at 2585 cm^{-1}, was attributed to SH and the other at 3200–3300 cm^{-1}, was assigned to NH_2, which were disappeared by the formation of the triazolo thiadiazoles (**4**–**11**). Similarly the ^1H NMR spectra of the synthesized triazoles **3**(**a**–**c**) showed two characteristic broad signals at δ 5.5–5.7 and the other at δ 13.8–13.95, due to NH_2 and SH proton, respectively, which were absent in the ^1H NMR spectra of triazolothiadiazoles. The absence of these absorptions due to SH and NH_2 established that all the triazoles had converted to triazolo thiadiazoles by reacting with the –COOH group of the various acids. The ^1H NMR, mass spectra, IR and elemental analysis supported the structure of various synthesized triazolo thiadiazoles.

A slight modification has been done to the literature procedure, which called for the removal of excess phosphorus oxychloride under reduced pressure. This could result in damage to the rotatory evaporator and vacuum pump due to the strong acidity and high boiling point of phosphorus oxychloride. Hence alternative method is suggested where in the reaction mixture is poured into crushed ice with stirring. Some solid potassium carbonate and required quantity of solid potassium hydroxide were added till the pH of the reaction mixture was raised to 8.

3. Biological results and discussion

The results of the antimicrobial effect of the newly synthesized compounds were reported as minimum inhibition concentration (MIC) against *Staphylococcus aureus*, *Bacillus subtilis* (gram-positive bacteria), *Pseudomonas aeruginosa*, *Escherichia coli* (gram-negative bacteria) and two fungi *Aspergillus niger* (*A. niger*) and *Candida albicans* (*C. albicans*). None of the synthesized compounds have important antimicrobial activities. While compounds **4a**, **4b**, **4c**, **11a**, **11b**, and **11c** exhibit moderate inhibitory activities against the tested strains of bacteria. Compounds **7a**, **7b**, and **7c** exhibit moderate inhibitory activity against *P. aeruginosa* and *E. coli*. The other compounds had no inhibitory activity. The results revealed that compounds **11a**, **11b** and **11c** exhibited the highest degree of inhibition against various microbial strains. Moreover, the biological activities of the other compounds against the tested organisms are very weak. However, the activities of the tested compounds are much less than those of standard antifungal and antibacterial agents used.

Some of the tested compounds **9a**, **9b**, **9c**, **11a**, **11b** and **11c** had shown weak anti-inflammatory and analgesic activity. Other compounds had no anti-inflammatory and analgesic activities. The anti-inflammatory and analgesic activity studies indicated that compounds **11a**, **11b** and **11c** exhibited the highest degree of anti-inflammatory and analgesic activity. However, the activities of the tested compounds are very much less than those of standard agents used.

4. Experimental protocols

Thin layer chromatography was used to reach the completion of the reaction and purity of the compounds synthesized. Melting points were taken in open glass capillary tubes by using Thiel's tube containing liquid paraffin and were uncorrected. IR spectra in KBr were recorded on a Shimadzu-8400 FTIR spectrophotometer, ^{1}H NMR spectra were recorded on Brucker spectrophotometer (400 MHz) in DMSO-d_6/$CDCl_3$ using TMS as an internal standard (chemical shifts are expressed in δ, ppm), mass spectra were recorded in Finnigan MAT 8230 mass spectrophotometer and micro analysis were recorded on Thermo Finnigan FLASH EA 1112 CHNS Analyzer. The purity of the compounds were checked on silica gel-G coated plates by using ethyl acetate and petroleum ether (1:1) as the eluent and observed in UV light. All the synthesized compounds gave satisfactory elemental analyses.

4.1. Pharmacological evaluation

4.1.1. Anti-inflammatory activity

All the synthesized compounds were tested for their anti-inflammatory activity using carrageenan induced rat hind paw edema method of Winter et al. [15]. Albino rats (Wistar strain) of either sex, weighing between 150 and 200 g, were used for the experiment. Tested compounds showed weak to moderate activity ranging from 6% to 32%, while phenylbutazone, standard showed 55% of inhibition. The animals were divided into various groups and each group consisting of six animals. One group served as control and received 0.1 ml of 1% gum acacia suspension orally. Group II served as standard and received phenylbutazone at the dose of 100 mg kg^{-1} as suspension in gum acacia orally. One hour after the administration of test compounds at the dose of 100 mg kg^{-1} as suspension in gum acacia, 0.1 ml of 1% carrageenan in normal saline was given subcutaneously to the sub plantar region of right hind paw. The paw volume was measured immediately ('0' h) and after 1–4 h, respectively, by using plethysmometer. The difference between the paw volume at 4th and 0 h measurement was calculated and taken as edema volume. Percentage inhibition in the paw edema was calculated by using the formula, percentage inhibition = 100 $(1 - V_t/V_c)$, where V_t = mean increase in paw volume of test, and V_c = mean increase in paw volume of control. Percentage inhibition shown by tested compounds is recorded in Table 1.

4.1.2. Analgesic activity

All the compounds were tested for their analgesic activity by using Eddy's hot plate technique [16] and exhibited weak to moderate activity ranging from 6% to 32%. Mice (Swiss strain) of either sex weighing between 25 and 35 g were used for the experiment. Diclofenac sodium at the dose of 50 mg kg^{-1} body weight as suspension in 1% gum acacia was used as standard, which showed percentage analgesic activity of 52%. In this method heat is used as a source of pain. Animals were individually placed on a hot plate maintained at constant temperature () and the reaction of animals, such as paw licking or jump response (whichever appears first) was taken as the end point. Tested compounds at the dose of 50 mg kg^{-1} body weight was given as suspension in 1% gum acacia orally to animals and observed the reaction time of animals on the hot plate at 15, 30, 60, 90 and 120 min after the compound administration. A cut off time of 15 s was taken as maximum analgesic response to avoid injury to the paws. Percentage analgesic activity shown by the tested compounds is recorded in Table 2.

4.1.3. Antibacterial and antifungal activities

The antibacterial activity of title compounds was determined in vitro by using paper disc method against variety of pathogenic micro organisms like *E. coli*, *P. aeruginosa* (gram-negative), *S. aureus*, *B. subtilis* (gram-positive) at 25, 50, 100 μg ml^{-1} concentrations, respectively, in the nutrient agar media by measuring the zone of inhibition in mm. The solutions of required concentrations (25, 50, 100 μg ml^{-1}) of test compounds were prepared by dissolving the compounds in DMF. Under identical conditions the standard antibiotics, amikacin at 100 μg ml^{-1} showed zone of inhibition 36 mm for *E. coli*, 43 mm for *P. aeruginosa* and vancomycin at 100 μg ml^{-1} showed zone of inhibition 41 mm for gram-positive organism. Antibacterial activity shown by the compounds towards various bacteria is recorded in Table 3.

Table 1
Anti-inflammatory activity of compounds (**4**–**11**)

Compounds	Change in paw volume (in ml) after (±S.E.)#				Percentage inhibition of edema volume after			
	1 h	2 h	3 h	4 h	1 h	2 h	3 h	4 h
4a	0.84 ± 0.02	1.17 ± 0.02	1.61 ± 0.05	1.90 ± 0.03	6.6**	8.6*	10.6	15.6
4b	0.82 ± 0.02	1.13 ± 0.03	1.54 ± 0.03	1.85 ± 0.02	8.9***	11.7**	14.4	17.8
4c	0.86 ± 0.03	1.21 ± 0.03	1.67 ± 0.06	1.98 ± 0.05	4.5	5.5	7.2	12.0
5a	0.76 ± 0.03	1.03 ± 0.02	1.50 ± 0.02	1.70 ± 0.02	5.0*	12.0**	14.3	17.5
5b	0.76 ± 0.02	1.05 ± 0.02	1.55 ± 0.07	1.74 ±	5.0*	10.3*	11.6	15.5
5c	0.77 ± 0.03	1.07 ± 0.03	1.56 ± 0.02	1.78 ± 0.03	3.8	8.6	10.9	13.6
6a	0.76 ± 0.03	1.08 ± 0.04	1.57 ± 0.02	1.80 ± 0.03	5.0*	7.7	10.3	13.1
6b	0.75 ± 0.03	1.06 ± 0.02	1.55 ± 0.02	1.82 ± 0.02	6.3*	9.4*	11.4	12.1
6c	0.76 ± 0.03	1.09 ± 0.03	1.58 ± 0.02	1.83 ± 0.05	5.0*	6.8	9.7	11.6
7a	0.74 ± 0.04	1.05 ± 0.03	1.55 ± 0.04	1.70 ± 0.04	7.5**	10.3*	11.4	17.9
7b	0.73 ± 0.03	1.04 ± 0.03	1.50 ± 0.03	1.72 ± 0.05	8.8***	11.2**	14.3	16.9
7c	0.75 ± 0.03	1.07 ± 0.04	1.57 ± 0.04	1.81 ± 0.02	6.3*	8.5*	10.3	12.6
8a	0.57 ± 0.02	0.83 ± 0.03	1.29 ± 0.02	1.75 ± 0.02	5.0*	5.8	6.5	8.9
8b	0.57 ± 0.02	1.83 ± 0.02	1.30 ± 0.04	1.74 ± 0.02	5.0*	5.4	5.8	9.4
8c	0.58 ± 0.04	0.84 ± 0.03	1.30 ± 0.02	1.80 ± 0.02	3.4	4.9	5.8	6.3
9a	0.82 ± 0.03	1.12 ± 0.02	1.42 ± 0.02	1.60 ± 0.03	8.9***	12.5**	21.1*	28.9*
9b	0.81 ± 0.02	1.11 ± 0.03	1.44 ± 0.02	1.65 ± 0.02	10.0***	13.3**	20.0*	26.5*
9c	0.84 ± 0.03	1.12 ± 0.04	1.45 ± 0.03	1.70 ± 0.03	6.7**	12.5**	19.4*	24.4*
10a	0.57 ± 0.02	0.81 ± 0.06	1.21 ± 0.04	1.60 ± 0.03	5.0*	8.2	12.5	16.5
10b	0.57 ± 0.02	0.82 ± 0.05	1.21 ± 0.02	1.62 ± 0.02	5.0*	7.1	12.5	15.4
10c	0.56 ± 0.02	0.82 ± 0.04	1.22 ± 0.02	1.66 ± 0.03	6.7**	7.2	11.7	13.4
11a	0.81 ± 0.04	1.06 ± 0.06	1.35 ± 0.02	1.54 ± 0.04	10.0***	17.2***	25.0**	31.5**
11b	0.80 ± 0.02	1.04 ± 0.03	1.37 ± 0.02	1.58 ± 0.04	11.1***	18.8***	23.9*	29.9*
11c	0.79 ± 0.03	1.08 ± 0.02	1.43 ± 0.05	1.64 ± 0.04	12.2***	15.6***	20.6*	26.2*
Phenyl butazone	0.72 ± 0.03	0.92 ± 0.03	0.97 ± 0.04	1.00 ± 0.03	10.0*	17.9**	37.1***	55.6***

= ±Standard error; * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$. For all other comparison $P > 0.05$.

Similarly, the antifungal screening of the compounds (**4**–**11**) was carried out in vitro by paper disc method against two fungi, *A. niger* and *C. albicans* by using griseofulvin (100 μg ml^{-1}) as the standard, which had shown (34 and 37 mm, respectively) as the zone of inhibition. Antifungal activity shown by the compounds towards fungi is recorded in Table 4. DMF was used, as the solvent control because the tested compounds is freely soluble in DMF, for both antibacterial and antifungal activities.

4.2. Preparation

4.2.1. General method for the synthesis of aryl acid hydrazide (*1*)

Dissolved the esters of substituted aromatic acids (0.1 M), in 30 ml of ethanol and hydrazine hydrate (0.1 M) was added drop wise to the mixture with stirring. The resulting mixture was allowed to reflux for 6 h. Excess ethanol was distilled out and the contents were allowed to cool. The crystals formed was filtered, washed thoroughly with water and dried. The completion of the reaction was monitored on TLC by using silica gel-G coated plates by using ethyl acetate and petroleum ether (1:1) as the eluent and observed in UV light.

4.2.2. General method for the synthesis of potassium dithiocarbazinate (*2*)

Potassium hydroxide (0.15 M) was dissolved in absolute ethanol (200 ml). To the above solution, aryl acid hydrazide (0.1 M) was added and cooled the solution in ice. To this, carbon disulfide (0.15 M) was added in small portions with constant stirring. The reaction mixture was agitated continuously for a period of 15 h. It was then diluted with anhydrous ether. The precipitated potassium dithiocarbazinate was collected by filtration. The precipitate was further washed with anhydrous ether (100 ml) and dried under vacuum. The potassium salt thus obtained was in quantitative yield and was used in the next step without further purification.

4.2.3. General method for the synthesis of 3-substituted-4-amino-5-mercapto-1,2,4-triazole *3(a–c)*

A suspension of potassium dithiocarbazinate of respective aromatic esters (**2**), (0.1 M) in water (5 ml) and hydrazine hydrate (15 ml, 0.3 M) was refluxed for 6–7 h with occasional shaking. The color of the reaction mixture changed to green with the evolution of hydrogen sulfide gas (lead acetate paper and odor). A homogenous reaction mixture was obtained during the reaction process. The reaction mixture was cooled to room temperature and diluted with water (100 ml). On acidification with concentrated hydrochloric acid, the required triazole was precipitated. It was filtered, washed thoroughly with cold water and recrystallized from ethanol. The completion of the reaction was monitored on TLC by using silica gel-G coated plates by using ethyl acetate and petroleum ether (1:1) as the eluent and observed in UV light.

4.2.4. 4-Amino-3-[4-(N,N-dimethyl amino) phenyl]-5-mercapto-1,2,4-triazole (*3a*)

M.p.: 248°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3313 (NH stretching), 1608 (C=N stretching); 3131 (aromatic CH stretching), 2586 (SH), 2978, 2813 (methyl CH stretch), 1232 (N–

Table 2
Analgesic activity of compounds (**4**–**11**)

Compounds	Reaction time (s) after drug administration			Percent increase in reaction time		
	30 min ± S.E.#	60 min ± S.E.#	90 min ± S.E.#	30 min	60 min	90 min
4a	5.20 ± 0.33	5.84 ± 0.55	6.71 ± 0.45	4.4	7.4	12.6
4b	5.87 ± 0.33	8.24 ± 0.60	9.17 ± 0.31	5.4	9.1	13.2
4c	7.22 ± 0.31	8.17 ± 0.34	9.63 ± 0.31	4.6	8.4	11.1
5a	6.44 ± 0.34	7.12 ± 0.37	8.08 ± 0.21	7.6	11.7	14.0
5b	5.50 ± 0.48	6.00 ± 0.17	7.45 ± 0.55	6.0	8.9	13.6
5c	6.41 ± 0.55	7.67 ± 0.45	8.17 ± 0.61	5.6	6.6	10.0
6a	8.17 ± 0.31	8.83 ± 0.48	9.83 ± 0.17	4.9	8.7	13.9
6b	6.00 ± 0.73	6.50 ± 0.43	7.50 ± 0.35	4.5	8.4	12.6
6c	4.46 ± 0.34	4.92 ± 0.55	5.36 ± 0.48	4.2	6.1	10.3
7a	4.56 ± 0.43	5.24 ± 0.48	6.04 ± 0.21	5.6	9.1	12.0
7b	7.84 ± 0.33	8.24 ± 0.60	9.17 ± 0.31	5.0	7.3	11.3
7c	4.02 ± 0.42	4.60 ± 0.26	5.83 ± 0.31	4.3	5.8	9.4
8a	8.17 ± 0.31	8.53 ± 0.48	9.00 ± 0.17	3.1	4.5	6.9
8b	7.45 ± 0.33	8.24 ± 0.60	9.17 ± 0.31	2.8	4.0	6.2
8c	–	8.67 ± 0.48	9.17 ± 0.55	–	3.2	5.8
9a	6.17 ± 0.34	7.15 ± 0.56	8.55 ± 0.37	10.3	19.2*	29.8*
9b	5.12 ± 0.33	5.78 ± 0.55	6.48 ± 0.45	11.8	21.7*	29.4*
9c	4.34 ± 0.37	5.24 ± 0.43	8.00 ± 0.31	9.3	19.5*	25.9*
10a	5.87 ± 0.58	6.36 ± 0.26	7.64 ± 0.31	4.6	7.8	13.2
10b	8.17 ± 0.43	8.83 ± 0.43	9.83 ± 0.21	5.2	6.6	12.3
10c	5.83 ± 0.37	6.67 ± 0.43	7.33 ± 0.31	4.7	7.3	10.5
11a	4.46 ± 0.34	4.92 ± 0.42	6.14 ± 0.34	11.8	20.3*	31.9*
11b	5.54 ± 0.56	7.67 ± 0.50	8.45 ± 0.61	10.5	20.8*	28.9*
11c	6.33 ± 0.43	7.33 ± 0.33	8.76 ± 0.22	9.2	19.4*	26.5*
Diclofenac sodium	5.67 ± 0.50	6.50 ± 0.43	7.33 ± 0.21	26.2***	37.2***	52.3***

= Standard error; * = $P < 0.05$; *** = $P < 0.001$. For all other comparison $P > 0.05$.

N=C), 1564, 1542, 1470, 1434 (C=C ring stretching), 1322 (C–N stretching); ^{1}H NMR: 13.65 (s, 1H, SH), 6.8, 7.9 (d, 4H, Ar-H), 5.73 (s, 2H, NH_2), 2.99 (s, 6H, N $(CH_3)_2$); MS *m/z* [% rel. int.]: 235 [24] (M+). Calcd. (%) for $C_{10}H_{13}N_5S$: C; 51.04, H; 5.57, N; 29.76, S; 13.63. Found: C; 50.96, H; 5.56, N; 29.75, S; 13.63.

*4.2.5. 4-Amino-3-[2-(N-methyl amino) phenyl]-5-mercapto-1,2,4-triazole (**3b**)*

M.p.: 221°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3355, 3300 (NH stretching), 1610 (C=N stretching), 3110 (aromatic CH stretching), 1579, 1510, 1467, 1428 (C=C ring stretching), 2587 (SH), 2940, 2837 (methyl CH stretching), 1286 (N–N=C), 1316 (C–N stretching); ^{1}H NMR: 13.89 (s, 1H, SH), 7.3 (d, 1H of Ar), 7.7 (d, 1H of Ar), 6.6–6.7 (m, 2H of Ar), 5.6 (s, 2H, NH_2), 6.18 (q, 1H, NH in NH–CH_3), 2.8 (d, 3H, CH_3 in NH–CH_3); MS *m/z* [% rel. int.]: 221 [38] (M+). Calcd. (%) for $C_9H_{11}N_5S$: C; 48.85, H; 5.01, N; 31.65, S; 14.49. Found: C; 48.90, H; 5.01, N; 31.62, S; 14.49.

*4.2.6. 4-Amino-3-(1-naphthyl methyl)-5-mercapto-1,2,4-triazole (**3c**)*

M.p.: 210°C yield (%): 70%; IR (KBr) ν (cm^{-1}): 3269 (NH stretching), 1625 (C=N stretching), 3162, 3044 (aromatic CH stretching), 1596, 1570, 1495, 1425 (C=C ring stretching), 2600 (SH), 2929 (methyl CH stretch), 1254 (N–N=C); ^{1}H NMR: 13.48 (s, 1H, SH), 7.3–8.1 (m, 7H, Ar-H), 5.67 (s, 2H, NH_2), 4.5 (s, 2H, CH_2); MS *m/z* [% rel. int.]: 256 [26] (M+). Calcd. (%) for $C_{13}H_{12}N_4S$: C; 60.91, H; 4.72, N; 21.86, S; 12.51. Found: C; 60.86, H; 4.73, N; 21.84, S; 12.52.

*4.2.7. Synthesis of 3-aryl/aralkyl substituted-6-(2-methyl/2-phenyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles **4** and **5(a–c)***

An equimolar mixture of respective triazole **3**(**a**–**c**) (0.02 M), 2-methyl/2-phenyl-quinoline-4-carboxylic acid (0.02 M) in dry phosphorous oxy chloride (10 ml) was refluxed for 7 h. The reaction mixture was cooled to room temperature and then gradually poured onto crushed ice with stirring. Finely powdered potassium carbonate and the required amount of solid potassium hydroxide were added till the pH of the mixture was raised to 8, to remove the excess of phosphorous oxychloride. The mixture was allowed to stand overnight and solid was separated. It was filtered, washed thoroughly with cold water, dried and recrystallized from hot ethanol.

*4.2.8. 3-[4-(N,N-dimethyl amino) phenyl]-6-(2-phenyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**4a**)*

M.p.: 224°C yield (%): 58%; IR (KBr) ν (cm^{-1}): 3050 (aromatic CH stretching), 1618 (C=N stretching), 1594, 1480, 1464 (C=C ring stretch), 2932, 2875 (methyl CH stretch), 1331 (C–N stretching), 1261 (N–N=C); ^{1}H NMR δ: 6.8–8.7 (m, Ar-H), 3.1 (s, 6H, N $(CH_3)_2$); MS *m/z* [% rel. int.]: 449 [24] (M+). Calcd. (%) for $C_{26}H_{20}N_6S$: C; 69.62, H; 4.49, N; 18.74, S; 7.15. Found: C; 69.59, H; 4.49, N; 18.75, S; 7.15.

Table 3
Antibacterial activity of compounds[#] (**3**–**11**)

Compounds	*E. coli* [1]			*P. aeruginosa* [2]			*B. subtilis* [3]			*S. aureus*[4]		
	25 μg ml^{-1}	50 μg ml^{-1}	100 μg ml^{-1}	25 μg ml^{-1}	50 μg ml^{-1}	100 μg ml^{-1}	25 μg ml^{-1}	50 μg ml^{-1}	100 μg ml^{-1}	25 μg ml^{-1}	50 μg ml^{-1}	100 μg ml^{-1}
	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*	±S.D.*
3a	3.00 ± 1.00	6.67 ± 0.58	10.33 ± 0.58	2.00 ± 0.00	5.33 ± 1.16	9.33 ± 0.58	1.67 ± 0.58	5.67 ± 0.58	11.33 ± 1.53	2.67 ± 0.58	5.67 ± 0.58	11.33 ± 0.58
3b	3.33 ± 1.16	7.67 ± 0.58	11.33 ± 0.58	4.67 ± 1.16	7.33 ± 0.58	13.67 ± 0.58	2.00 ± 0.00	6.00 ± 1.00	12.67 ± 1.16	3.00 ± 0.00	5.33 ± 0.58	11.00 ± 1.00
3c	3.00 ± 0.58	7.67 ± 1.16	11.00 ± 0.58	6.33 ± 0.58	11.67 ± 0.58	13.00 ± 1.00	1.33 ± 0.58	4.67 ± 1.16	10.33 ± 1.16	1.67 ± 0.57	4.67 ± 0.58	10.33 ± 1.53
4a	7.33 ± 1.16	12.33 ± 1.53	17.67 ± 0.58	6.33 ± 0.58	11.67 ± 0.58	17.00 ± 1.00	6.00 ± 1.00	12.30 ± 1.53	17.33 ± 0.58	5.67 ± 0.58	12.33 ± 0.58	18.33 ± 1.53
4b	7.00 ± 1.73	10.33 ± 2.52	17.00 ± 2.65	5.33 ± 1.16	10.33 ± 0.58	16.33 ± 0.58	6.00 ± 0.00	11.67 ± 1.16	17.33 ± 1.53	5.33 ± 0.58	10.67 ± 0.58	17.33 ± 0.58
4c	7.00 ± 1.73	10.33 ± 2.52	15.67 ± 2.89	4.67 ± 1.16	9.33 ± 0.58	14.67 ± 0.58	4.67 ± 0.58	8.33 ± 0.58	15.33 ± 0.58	5.33 ± 0.58	10.67 ± 0.58	16.33 ± 1.16
5a	6.67 ± 1.16	10.33 ± 1.53	16.33 ± 2.52	6.67 ± 0.58	10.67 ± 0.58	17.33 ± 0.58	6.33 ± 0.58	11.67 ± 1.16	17.00 ± 1.00	4.33 ± 0.58	9.33 ± 1.16	15.67 ± 0.58
5b	8.00 ± 1.00	11.67 ± 0.58	16.33 ± 0.58	7.67 ± 0.58	11.67 ± 0.58	17.33 ± 1.16	5.33 ± 1.16	10.67 ± 0.58	16.67 ± 0.58	5.33 ± 0.58	12.33 ± 0.58	17.00 ± 1.00
5c	5.33 ± 0.58	10.67 ± 0.58	14.67 ± 1.53	6.67 ± 0.58	10.33 ± 0.58	15.33 ± 1.16	7.33 ± 1.16	12.33 ± 0.58	18.00 ± 1.73	6.33 ± 0.58	11.33 ± 0.58	16.67 ± 0.58
6a	6.67 ± 1.16	10.00 ± 0.00	15.00 ± 0.00	8.33 ± 0.58	11.67 ± 1.16	16.33 ± 0.58	4.67 ± 1.16	9.33 ± 0.58	15.33 ± 0.58	4.33 ± 0.58	8.33 ± 0.58	14.67 ± 0.58
6b	5.00 ± 1.00	8.33 ± 1.16	14.33 ± 1.53	8.67 ± 1.16	11.67 ± 0.58	15.67 ± 1.53	5.33 ± 1.16	8.67 ± 0.58	15.00 ± 1.00	5.33 ± 0.58	10.33 ± 0.58	17.00 ± 1.00
6c	5.67 ± 0.58	8.33 ± 1.16	14.00 ± 1.73	9.00 ± 1.00	11.67 ± 1.53	15.33 ± 0.58	5.33 ± 0.58	9.67 ± 0.58	16.33 ± 1.53	6.00 ± 0.00	10.67 ± 0.58	16.67 ± 0.58
7a	4.33 ± 0.58	9.00 ± 1.00	18.67 ± 0.58	7.33 ± 0.58	12.33 ± 0.58	18.67 ± 0.58	4.33 ± 1.53	8.33 ± 0.58	18.00 ± 1.00	4.00 ± 0.00	8.67 ± 1.16	14.67 ± 0.58
7b	7.33 ± 0.58	12.33 ± 0.58	17.67 ± 1.16	6.67 ± 1.16	10.67 ± 0.58	18.00 ± 1.73	6.33 ± 1.16	11.33 ± 1.16	17.33 ± 1.53	3.33 ± 0.58	6.67 ± 0.58	14.33 ± 0.58
7c	6.00 ± 0.00	10.33 ± 0.58	15.33 ± 1.16	4.67 ± 1.16	8.67 ± 0.58	14.67 ± 0.58	7.67 ± 1.16	12.33 ± 0.58	18.00 ± 2.00	4.33 ± 0.58	7.33 ± 0.58	16.00 ± 1.00
8a	3.67 ± 1.16	8.67 ± 1.53	12.33 ± 0.58	3.67 ± 0.58	6.67 ± 1.16	11.67 ± 1.53	6.00 ± 1.00	9.67 ± 1.16	15.00 ± 1.00	2.67 ± 0.58	6.00 ± 1.00	12.33 ± 0.58
8b	4.67 ± 1.16	8.33 ± 1.16	13.00 ± 0.00	4.00 ± 0.00	6.00 ± 1.73	11.33 ± 2.31	3.33 ± 1.53	6.33 ± 0.58	10.67 ± 0.58	2.33 ± 0.58	6.33 ± 0.58	11.33 ± 1.16
8c	4.00 ± 0.00	6.67 ± 1.16	10.67 ± 2.68	1.33 ± 0.58	4.67 ± 1.16	10.67 ± 1.16	1.67 ± 0.58	4.00 ± 0.00	9.00 ± 0.00	2.33 ± 0.58	5.33 ± 0.58	11.00 ± 1.00
9a	6.00 ± 0.00	8.67 ± 1.16	12.67 ± 0.58	3.00 ± 1.00	7.33 ± 0.58	12.33 ± 0.58	2.33 ± 0.58	5.33 ± 0.58	10.67 ± 0.58	3.33 ± 0.58	6.33 ± 0.58	12.67 ± 0.58
9b	3.67 ± 1.16	6.67 ± 1.16	11.67 ± 0.58	3.33 ± 0.58	6.67 ± 0.58	12.67 ± 1.53	4.00 ± 0.00	6.33 ± 0.58	12.00 ± 0.00	2.00 ± 0.00	4.67 ± 0.58	9.67 ± 1.16
9c	2.00 ± 0.00	4.33 ± 0.58	7.67 ± 1.16	0.33 ± 0.58	2.67 ± 0.58	7.00 ± 1.73	1.33 ± 0.58	5.33 ± 0.58	8.33 ± 1.53	1.33 ± 0.58	3.33 ± 0.58	6.33 ± 0.58
10a	4.67 ± 1.16	7.67 ± 0.58	13.00 ± 1.00	1.67 ± 0.58	5.33 ± 0.58	11.33 ± 0.58	3.00 ± 1.00	5.67 ± 0.58	11.33 ± 0.58	2.67 ± 0.58	5.67 ± 0.58	12.33 ± 0.58
10b	3.33 ± 0.58	6.33 ± 0.58	7.33 ± 1.16	0.67 ± 0.58	3.67 ± 0.58	7.33 ± 0.58	1.67 ± 0.58	4.67 ± 1.16	10.67 ± 0.58	2.67 ± 0.58	5.33 ± 0.58	10.67 ± 1.53
10c	3.67 ± 0.58	6.67 ± 0.58	9.00 ± 1.00	1.33 ± 0.58	4.33 ± 0.58	8.00 ± 1.00	1.67 ± 0.58	4.00 ± 0.00	8.33 ± 1.53	1.33 ± 0.58	4.33 ± 0.58	8.33 ± 0.58
11a	8.00 ± 1.00	12.33 ± 1.16	17.67 ± 1.16	7.33 ± 1.53	12.67 ± 0.58	18.33 ± 0.58	8.33 ± 1.16	13.33 ± 1.16	19.00 ± 1.73	6.33 ± 0.58	11.67 ± 0.58	17.67 ± 0.58
11b	7.33 ± 0.58	11.67 ± 1.16	17.00 ± 1.00	8.00 ± 1.00	11.67 ± 0.58	17.33 ± 1.53	6.67 ± 0.58	11.67 ± 0.58	16.67 ± 0.58	6.33 ± 0.58	11.33 ± 0.58	17.00 ± 1.00
11c	8.33 ± 0.58	12.33 ± 0.58	17.33 ± 1.16	6.67 ± 1.16	12.33 ± 0.58	17.67 ± 1.16	5.33 ± 0.58	10.67 ± 0.58	16.33 ± 0.58	6.67 ± 0.58	12.33 ± 0.58	18.33 ± 0.58
Vancomycin	–	–	–	–	–	–	–	20.33 ± 1.53	41.00 ± 3.00	–	22.33 ± 0.58	40.67 ± 1.53
Amikacin	–	21.67 ± 0.58	36.00 ± 1.00	–	24.00 ± 2.65	43.00 ± 1.00	–	–	–	–	–	–

1 = *Escherichia coli*; 2 = *Pseudomonas aeruginosa*; 3 = *Staphylococcus aureus*; 4 = *Bacillus subtilis*; *S.D. = standard deviation; # = zone of inhibition in mm.

Table 4
Antifungal activities of compounds # (**3**–**11**)

Compounds	*C. albicans*[1]			*A. niger*[2]		
	25 (μg ml^{-1}) ±S.D.*	50 (μg ml^{-1}) ±S.D.*	100 (μg ml^{-1}) ±S.D.*	25 (μg ml^{-1}) ±S.D.*	50 (μg ml^{-1}) ±S.D.*	100 (μg ml^{-1}) ±S.D.*
3a	2.33 ± 0.58	5.33 ± 0.58	10.33 ± 0.58	2.33 ± 0.58	5.33 ± 0.58	12.33 ± 0.58
3b	2.33 ± 0.58	5.33 ± 1.16	11.00 ± 1.00	1.67 ± 0.58	4.33 ± 0.58	10.67 ± 1.53
3c	2.00 ± 1.00	5.00 ± 1.00	11.33 ± 0.58	2.33 ± 0.58	4.67 ± 0.58	10.33 ± 0.58
4a	6.00 ± 1.00	10.33 ± 1.16	16.33 ± 1.53	3.33 ± 0.58	7.33 ± 0.58	16.33 ± 1.16
4b	6.33 ± 0.58	10.67 ± 0.58	17.00 ± 1.00	5.67 ± 0.58	10.00 ± 1.00	15.67 ± 1.16
4c	4.67 ± 1.16	8.33 ± 0.58	13.33 ± 1.16	6.33 ± 0.58	10.67 ± 0.58	16.33 ± 0.58
5a	5.33 ± 1.16	9.33 ± 0.58	16.33 ± 0.58	4.67 ± 0.58	8.67 ± 0.58	14.67 ± 0.58
5b	4.67 ± 0.58	9.33 ± 0.58	16.33 ± 1.16	5.67 ± 0.58	10.33 ± 0.58	17.00 ± 1.00
5c	4.00 ± 1.00	6.67 ± 0.58	14.33 ± 0.58	5.33 ± 0.58	9.67 ± 0.58	16.33 ± 0.58
6a	4.67 ± 0.58	7.33 ± 0.58	15.00 ± 1.00	5.67 ± 0.58	10.33 ± 0.58	17.33 ± 0.58
6b	3.33 ± 0.58	6.33 ± 0.58	14.33 ± 0.58	±	10.00 ± 0.00	17.00 ± 0.00
6c	4.67 ± 0.58	7.67 ± 0.58	15.67 ± 0.58	4.67 ± 0.58	9.33 ± 0.58	16.00 ± 1.00
7a	4.67 ± 0.58	7.67 ± 0.58	16.00 ± 0.00	5.33 ± 0.58	9.67 ± 0.58	16.33 ± 2.08
7b	4.67 ± 0.58	7.33 ± 0.58	16.33 ± 1.53	5.33 ± 1.16	9.67 ± 1.16	16.33 ± 1.16
7c	5.00 ± 0.00	7.67 ± 0.58	16.33 ± 0.58	6.33 ± 0.58	10.67 ± 0.58	17.67 ± 1.16
8a	2.33 ± 0.58	5.67 ± 0.58	11.33 ± 0.58	1.67 ± 0.58	5.67 ± 0.58	10.67 ± 1.16
8b	2.67 ± 0.58	5.67 ± 0.58	13.33 ± 0.58	1.33 ± 0.58	5.33 ± 0.58	11.33 ± 0.58
8c	1.67 ± 0.58	4.33 ± 0.58	10.33 ± 0.58	1.33 ± 0.58	4.67 ± 0.58	9.33 ± 1.16
9a	1.33 ± 0.58	4.00 ± 0.00	10.33 ± 1.16	2.33 ± 0.58	5.67 ± 0.58	12.00 ± 1.73
9b	2.33 ± 0.58	5.00 ± 1.00	11.33 ± 1.53	3.33 ± 0.58	6.00 ± 0.00	12.67 ± 0.58
9c	–	2.33 ± 0.58	5.33 ± 0.58	–	2.67 ± 1.16	5.33 ± 1.53
10a	1.67 ± 0.58	4.67 ± 0.58	11.33 ± 1.53	2.33 ± 0.58	5.67 ± 0.58	12.33 ± 0.58
10b	1.33 ± 0.58	4.33 ± 0.58	11.00 ± 2.00	1.67 ± 0.58	4.33 ± 0.58	9.33 ± 1.53
10c	1.33 ± 0.58	4.33 ± 0.58	8.67 ± 0.58	1.67 ± 0.58	4.67 ± 0.58	9.00 ± 1.00
11a	6.33 ± 0.58	13.33 ± 1.16	18.33 ± 1.16	6.67 ± 0.58	13.33 ± 1.16	18.33 ± 2.08
11b	5.67 ± 0.58	11.67 ± 0.58	17.67 ± 1.16	6.67 ± 0.58	12.67 ± 1.16	18.33 ± 1.53
11c	5.33 ± 0.58	10.67 ± 0.58	17.33 ± 0.58	4.67 ± 0.58	9.67 ± 1.16	16.00 ± 1.73
Griseofulvin	–	20.33 ± 0.58	34.67 ± 1.16	–	21.67 ± 2.08	37.00 ± 2.00

*S.D. = standard deviation; 1 = *Candida albicans*; 2 = *Aspergillus niger*.

*4.2.9. 3-[2-(N-methyl amino) phenyl]-6-(2-phenyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**4b**)*

M.p.: 208°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3329 (NH stretching), 1610 (C=N stretching), 3090 (aromatic CH stretching), 1579, 1510, 1467, 1428 (C=C ring stretching), 2980, 2837 (methyl CH stretching), 1280 (N–N=C), 1320 (C–N stretching); ^{1}H NMR δ: 7.3–8.5 (m, Ar-H), 6.6–6.7 (m, 2H of Ar), 6.42 (q, 1H, NH in NH–CH_3), 2.9 (d, 3H, CH_3 in NH–CH_3); MS *m*/*z* [% rel. int.]: 434 [18] (M+). Calcd. (%) for $C_{25}H_{18}N_6S$: C; 69.10, H; 4.18, N; 19.34, S; 7.38. Found: C; 69.06, H; 4.18, N; 19.35, S; 7.38.

*4.2.10. 3-(1-Naphthyl methyl)-6-(2-phenyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**4c**)*

M.p.: 198°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3080 (aromatic CH stretching), 1612 (C=N stretching), 1518, 1483 (C=C ring stretching), 2930, 2850 (methyl CH stretch), 1280 (N–N=C); ^{1}H NMR δ: 7.2–8.4 (m, Ar-H), 5.02 (s, 2H, CH_2); MS *m*/*z* [% rel. int.]: 469 [24] (M+). Calcd. (%) for $C_{29}H_{19}N_5S$: C; 74.18, H; 4.08, N; 14.91, S; 6.83. Found: C; 74.09, H; 4.06, N; 14.92, S; 6.83.

*4.2.11. 3-[4-(N,N-dimethyl amino) phenyl]-6-(2-methyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**5a**)*

M.p.: 184°C yield (%): 58%; IR (KBr) ν (cm^{-1}): 3050 (aromatic CH stretching), 1618 (C=N stretching), 1594, 1480, 1464 (C=C ring stretch), 2932, 2875 (methyl CH stretch), 1331 (C–N stretching), 1261 (N–N=C); ^{1}H NMR δ: 6.8–8.2 (m, Ar-H), 3.1 (s, 6H, N $(CH_3)_2$), 2.46 (s, 2H, CH_3); MS *m*/*z* [% rel. int.]: 386 [32] (M+). Calcd. (%) for $C_{21}H_{18}N_6S$: C; 65.26, H; 4.69, N; 21.75, S; 8.30. Found: C; 65.31, H; 4.69, N; 21.73, S; 8.30.

*4.2.12. 3-[2-(N-methyl amino) phenyl]-6-(2-methyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**5b**)*

M.p.: 200°C yield (%): 56%; IR (KBr) ν (cm^{-1}): 3329 (NH stretching), 1610 (C=N stretching), 3090 (aromatic CH stretching), 1579, 1510, 1467, 1428 (C=C ring stretching), 2980, 2837 (methyl CH stretching), 1280 (N–N=C), 1320 (C–N stretching); ^{1}H NMR δ: 7.3–8.3 (m, Ar-H), 6.6–6.7 (m, 2H of Ar), 6.40 (q, 1H, NH in NH–CH_3), 2.94 (d, 3H, CH_3 in NH–CH_3), 2.44 (s, 2H, CH_3); MS *m*/*z* [% rel. int.]: 372 [36] (M+). Calcd. (%) for $C_{20}H_{16}N_6S$: C; 64.50, H; 4.33, N; 22.56, S; 8.61. Found: C; 64.43, H; 4.33, N; 22.58, S; 8.61.

*4.2.13. 3-(1-Naphthyl methyl)-6-(2-methyl-4-quinolinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**5c**)*

M.p.: 196°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3080 (aromatic CH stretching), 1612 (C=N stretching), 1518, 1483 (C=C ring stretching), 2930, 2850 (methyl CH stretch), 1280 (N–N=C); ^{1}H NMR δ: 7.2–8.2 (m, Ar-H), 5.0 (s, 2H, CH_2), 2.44 (s, 2H, CH_3); MS *m/z* [% rel. int.]: 407 [32] (M+). Calcd. (%) for $C_{24}H_{17}N_5S$: C; 70.74, H; 4.21, N; 17.19, S; 7.87. Found: C; 70.68, H; 4.21, N; 17.20, S; 7.87.

*4.2.14. Synthesis of 3-aryl/aralkyl substituted-6-(2-chloro/2,6-dihydroxy-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles **6** and **7(a–c)***

Equimolar proportions of respective triazole **3(a–c)** (0.02 M) and 2-chloro/2,6-dihydroxy-pyridinyl-4-carboxylic acid (0.02 M), were dissolved in 10 ml of dry phosphorous oxy chloride and the resulting solution was refluxed for 10 h. Cooled the reaction mixture to room temperature and then gradually poured onto crushed ice with stirring. Finely powdered potassium carbonate and the required amount of solid potassium hydroxide were added to the solution with stirring till the pH of the mixture was raised to 8, to remove the excess of phosphorous oxychloride. On allowing the reaction mixture to stand overnight, solid was separated. It was filtered, washed thoroughly with cold water, dried and recrystallized from aqueous DMF.

*4.2.15. 3-[4-(N,N-dimethyl amino) phenyl]-6-(2,6-dihydroxy-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**6a**)*

M.p.: 288°C yield (%): 55%; IR (KBr) ν (cm^{-1}): 3423 (OH stretching), 3045, 3010 (aromatic CH stretching), 1601 (C=N stretching), 1541, 1494, 1455 (C=C ring stretch), 2919, 2847 (methyl CH stretch), 1275 (N–N=C), 1328 (C–N stretching), 1225 (C–O stretching); ^{1}H NMR δ: 6.78, 7.86 (2d, 4H of Ar), 8.05 (s, 2H of pyridine), 6.32 (s, 2H, OH); MS *m/z* [% rel. int.]: 354 [40] (M+). Calcd. (%) for $C_{16}H_{14}N_6O_2S$: C; 54.23, H; 3.98, N; 23.71, S; 9.05. Found: C; 54.19, H; 3.98, N; 23.74, S; 9.04.

*4.2.16. 3-[2-(N-methyl amino) phenyl]-6-(2,6-dihydroxy-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**6b**)*

M.p.: 292°C yield (%): 55%; IR (KBr) ν (cm^{-1}): 3443 (OH stretching), 3317 (NH stretching), 3076, 3034 (aromatic CH stretching), 1607 (C=N stretching), 1590, 1538, 1478, 1448 (C=C ring stretch), 2970, 2934 (methyl CH stretch), 1334 (C–N stretching), 1262 (asymmetric C–O stretching), 1075 (symmetric C–O stretching), 1275 (N–N=C); ^{1}H NMR δ: 7.3, 7.8 (2d, 2H of Ar), 6.7–6.8 (m, 2H of Ar), 8.33 (d, 2H of Ar′), 6.05 (s, 2H, OH), 6.42 (q, 1H, NH in NH–CH_3), 2.94 (d, 3H, CH_3 in NH–CH_3); MS *m/z* [% rel. int.]: 340 [26] (M+). Calcd. (%) for $C_{15}H_{12}N_6O_2S$: C; 52.93, H; 3.55, N; 24.69, S; 9.42. Found: C; 52.89, H; 3.55, N; 24.71, S; 9.42.

*4.2.17. 3-(1-Naphthyl methyl)-6-(2,6-dihydroxy-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**6c**)*

M.p.: 284°C yield (%): 50%; IR (KBr) ν (cm^{-1}): 3456 (OH stretching), 3045 (aromatic CH stretching), 1610 (C=N stretching), 1572, 1484, 1458 (C=C ring stretch), 2970, 2920 (methyl CH stretch), 1239 (C–O stretching), 1280 (N–N=C); ^{1}H NMR δ: 7.4–8.2 (m, Ar-H), 6.3 (s, 2H, OH), 4.8 (s, 2H, CH_2); MS *m/z* [% rel. int.]: 375 [36] (M+). Calcd. (%) for $C_{19}H_{13}N_5O_2S$: C; 60.79, H; 3.49, N; 18.66, S; 8.54. Found: C; 60.71, H; 3.49, N; 18.70, S; 8.54.

*4.2.18. 3-[4-(N,N-dimethyl amino) phenyl]-6-(2-chloro-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**7a**)*

M.p.: 276°C yield (%): 62%; IR (KBr) ν (cm^{-1}): 3045, 3010 (aromatic CH stretching), 1601 (C=N stretching), 1541, 1494, 1455 (C=C ring stretch), 2919, 2847 (methyl CH stretch), 1329 (C–N stretching), 1275 (N–N=C); ^{1}H NMR δ: 6.82, 7.86 (2d, 4H of Ar), 7.34 (s, 1H of pyridine), 7.62 (d, 1H of pyridine), 8.12 (d, 1H of pyridine); MS *m/z* [% rel. int.]: 356 [34] (M+), 358 [12] (M + 2). Calcd. (%) for $C_{16}H_{13}N_6SCl$: C; 53.85, H; 3.67, N; 23.55, S; 8.99. Found: C; 53.90, H; 3.67, N; 23.56, S; 9.00.

*4.2.19. 3-[2-(N-methyl amino) phenyl]-6-(2-chloro-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**7b**)*

M.p.: 292°C yield (%): 58%; IR (KBr) ν (cm^{-1}): 3317 (NH stretching), 3076, 3034 (aromatic CH stretching), 1607 (C=N stretching), 1590, 1538, 1478, 1448 (C=C ring stretch), 2970, 2934 (methyl CH stretch), 1331 (C–N stretching), 1275 (N–N=C); ^{1}H NMR δ: 7.24, 7.68 (2d, 2H of Ar), 6.7–6.8 (m, 2H of Ar), 7.40 (s, 1H of pyridine), 7.54 (d, 1H of pyridine), 8.18 (d, 1H of pyridine), 6.38 (q, 1H, NH in NH–CH_3), 2.64 (d, 3H, CH_3 in NH–CH_3); MS *m/z* [% rel. int.]: 342 [45] (M+), 344 [14] (M + 2). Calcd. (%) for $C_{15}H_{11}N_6SCl$: C; 52.55, H; 3.23, N; 24.52, S; 9.35. Found: C; 52.50, H; 3.23, N; 24.50, S; 9.35.

*4.2.20. 3-(1-Naphthyl methyl)-6-(2-chloro-4-pyridinyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**7c**)*

M.p.: 284°C yield (%): 59%; IR (KBr) ν (cm^{-1}): 3045 (aromatic CH stretching), 1610 (C=N stretching), 1572, 1484, 1458 (C=C ring stretch), 2970, 2920 (methyl CH stretch), 1239 (C–O stretching), 1280 (N–N=C); ^{1}H NMR δ: 7.4–8.12 (m, Ar-H), 4.64 (s, 2H, CH_2); MS *m/z* [% rel. int.]: 377 [36] (M+), 379 [12] (M + 2). Calcd. (%) for $C_{19}H_{12}N_5SCl$: C; 60.40, H; 3.20, N; 18.53, S; 8.49. Found: C; 60.36, H; 3.20, N; 18.53, S; 8.49.

*4.2.21. Synthesis of 3-aryl/aralkyl substituted-6-(4-hydroxy-3-methoxy cinnamyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles **8(a–c)***

Respective triazole **3(a–c)**, (0.02 M) and 4-hydroxy-3-methoxy cinnamic acid (0.02 M) was added to

10 ml of dry phosphorous oxy chloride and the solution was refluxed for 5 h. The reaction mixture was cooled to room temperature and then gradually poured onto crushed ice with stirring. Finely powdered potassium carbonate and the required amount of solid potassium hydroxide were added till the pH of the mixture was raised to 8, to remove the excess of phosphorous oxychloride. The mixture was allowed to stand overnight and solid was separated. It was filtered, washed thoroughly with cold water, dried and recrystallized from hot ethanol.

*4.2.22. 3-[4-(N,N-dimethyl amino) phenyl]-6-(4-hydroxy-3-methoxy cinnamyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**8a**)*

M.p.: 206°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3448 (OH stretching), 3075 (aromatic CH stretching), 1609 (C=N stretching), 1578, 1540, 1481, 1450 (C=C ring stretch), 2970, 2860 (methyl CH stretch), 1260 (asymmetric C–O–C stretching), 1021 (symmetric C–O–C stretching), 1286 (N–N=C), 1360 (C–N stretching); ^{1}H NMR δ: 6.85, 8.1 (2d, 2H of Ar), 7.1, 7.82 (2d, 2H of Ar′), 7.52 (s, 1H of Ar′), 3.0 (s, 6H, N $(CH_3)_2$), 3.89 (d, 3H, OCH_3), 6.8 (d, 1H, CH), 6.3 (d, 1H, CH), 6.1 (s, 1H, OH); MS *m/z* [% rel. int.]: 393 [44] (M+). Calcd. (%) for $C_{20}H_{19}N_5O_2S$: C; 61.05, H; 4.88, N; 17.80, S; 8.15. Found: C; 60.97, H; 4.88, N; 17.78, S; 8.14.

*4.2.23. 3-[2-(N-methyl amino) phenyl]-6-(4-hydroxy-3-methoxy cinnamyl)-1,2,4-triazolo[3,4-b] 1,3,4-thiadiazole (**8b**)*

M.p.: 170°C yield (%): 56%; IR (KBr) ν (cm^{-1}): 3444 (OH stretching), 3312 (NH stretching), 3090 (aromatic CH stretching), 1612 (C=N stretching), 1590, 1560, 1485, 1460 (C=C ring stretch), 2985, 2840 (methyl CH stretch), 1264 (asymmetric C–O–C stretching), 1021 (symmetric C–O–C stretching), 1286 (N–N=C), 1360 (C–N stretching); ^{1}H NMR δ: 7.3, 7.7 (2d, 2H of Ar), 6.8–6.9 (m, 2H of Ar), 7.02, 7.86 (2d, 2H of Ar′), 7.50 (s, 1H of Ar′), 3.9 (d, 3H, OCH_3 of Ar′), 6.44 (q, 1H, NH in NH–CH_3), 2.88 (d, 3H, CH_3 in NH–CH_3), 6.62 (d, 1H, CH), 5.82 (d, 1H, CH), 6.12 (s, 1H, OH); MS *m/z* [% rel. int.]: 379 [38] (M+). Calcd. (%) for $C_{19}H_{17}N_5O_2S$: C; 60.14, H; 4.52, N; 18.46, S; 8.45. Found: C; 60.06, H; 4.52, N; 18.45, S; 8.44.

*4.2.24. 3-(1-Naphthyl methyl)-6-(4-hydroxy-3-methoxy cinnamyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**8c**)*

M.p.: 188°C yield (%): 56%; IR (KBr) ν (cm^{-1}): 3438 (OH stretching), 3060 (aromatic CH stretching), 1606 (C=N stretching), 1544, 1478, 1453 (C=C ring stretch), 2950, 2920 (methyl CH stretch), 1235 (asymmetric C–O–C stretching), 1070 (symmetric C–O–C stretching), 1288 (N–N=C); ^{1}H NMR δ: 7.4–7.7 (m, 7H, Ar-H), 7.98, 7.88, 8.3 (m, 3H of Ar), 3.85 (d, 3H, OCH_3), 4.84 (s, 2H, CH_2), 6.8 (d, 1H, CH), 6.32 (d, 1H,CH), 6.1 (s, 1H, OH); MS *m/z* [% rel. int.]: 414 [26] (M+). Calcd. (%) for $C_{23}H_{18}N_4O_2S$: C; 66.65, H; 4.38, N; 13.52, S; 7.74. Found: C; 66.56, H; 4.38, N; 13.53, S; 7.74.

*4.2.25. Synthesis of 3-aryl/aralkyl substituted-6-(2-amino-3,5-dibromo phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles **9(a–c)***

Respective triazole **3(a–c)** (0.02 M) was dissolved in dry phosphorous oxy chloride (10 ml) and to the above solution, added 2-amino-3, 5-dibromo benzoic acid (0.02 M). The resulted mixture was refluxed for 5 h. The reaction mixture was cooled to room temperature and the mixture was gradually poured onto crushed ice with stirring. Finely powdered potassium carbonate and the required amount of solid potassium hydroxide were added until the pH of the mixture was raised to 8, to remove the excess of phosphorous oxychloride. Solid was separated on allowing the mixture to stand overnight. It was filtered, washed thoroughly with cold water, dried and recrystallized from aqueous DMF.

*4.2.26. 3-[4-(N,N-dimethyl amino) phenyl]-6-(2-amino-3,5-dibromo phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**9a**)*

M.p.: 224°C yield (%): 55%; IR (KBr) ν (cm^{-1}): 3280 (NH stretching), 3080, (aromatic CH stretching), 1611 (C=N stretching), 1587, 1510, 1467 (C=C ring stretch), 2970, 2838 (methyl CH stretch), 1316 (C–N stretching), 1285 (N–N=C); ^{1}H NMR δ: 6.9, 8.0 (2d, 2H of Ar), 7.7, 7.9 (2s, 2H of Ar′), 6.7 (s, 2H, NH_2), 3.0 (s, 6H, N $(CH_3)_2$); MS *m/z* [% rel. int.]: 494 [28] (M+), 496[36] (M + 2), 498 [26] (M + 4). Calcd. (%) for $C_{17}H_{14}N_6SBr_2$: C; 41.32, H; 2.86, N; 17.01, S; 6.49. Found: C; 41.37, H; 2.86, N; 16.99, S; 6.50.

*4.2.27. 3-[2-(N-Methyl amino) phenyl]-6-(2-amino-3,5-dibromo phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**9b**)*

M.p.: 218°C yield (%): 58%; IR (KBr) ν (cm^{-1}): 3298 (NH stretching), 3086 (aromatic CH stretching), 1614 (C=N stretching), 1582, 1475, 1456 (C=C ring stretch), 2976, 2845 (methyl CH stretch), 1280 (N–N=C), 1328 (C–N stretching); ^{1}H NMR δ: 7.3, 7.7 (2d, 2H of Ar), 6.8–6.9 (m, 2H of Ar), 7.63, 7.9 (2s, 2H of Ar′), 6.44 (q, 1H, NH in NH–CH_3), 2.88 (d, 3H, CH_3 in NH–CH_3), 6.62 (s, 2H, NH_2), MS *m/z* [% rel. int.]: 480 [28] (M+), 482 [38] (M + 2), 484 [24] (M + 4). Calcd. (%) for $C_{16}H_{12}N_6SBr_2$: C; 40.02, H; 2.52, N; 17.50, S; 6.68. Found: C; 39.99, H; 2.52, N; 17.53, S; 6.68.

*4.2.28. 3-(1-Naphthyl methyl)-6-(2-amino-3,5-dibromo phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**9c**)*

M.p.: 210°C yield (%): 56%; IR (KBr) ν (cm^{-1}): 3273 (NH stretching), 3090, 3047 (aromatic CH stretching), 1612 (C=N stretching), 1585, 1575, 1484, 1436 (C=C ring stretch), 2964 (methylene CH stretch), 1290 (N–N=C); ^{1}H NMR δ: 7.3–7.6 (m, 4H of Ar), 7.7–7.9 (m, 4H, Ar-H), 8.3 (1d, 1H of Ar), 5.78 (s, 2H, NH_2), 4.93 (s, 2H, CH_2); MS *m/z* [% rel. int.]: 515 [20] (M+), 517 [28] (M + 2), 519 [18] (M + 4). Calcd. (%) for $C_{20}H_{13}N_5SBr_2$: C; 46.62, H; 2.54, N; 13.59, S; 6.22. Found: C; 46.70, H; 2.54, N; 13.58, S; 6.23.

*4.2.29. Synthesis of 3-aryl/aralkyl substituted-6-(2-methyl-4-nitro phenyl)1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles **10(a–c)***

An equimolar mixture of respective triazole **3(a–c)** (0.02 M), 2-methyl-4-nitro benzoic acid (0.02 M) in dry phosphorous oxy chloride (10 ml) was refluxed for 4 h. The reaction mixture was cooled to room temperature and then gradually poured onto crushed ice with stirring. Finely powdered potassium carbonate and the required amount of solid potassium hydroxide were added till the pH of the mixture was raised to 8, to remove the excess of phosphorous oxychloride. The mixture was allowed to stand overnight and solid was separated. It was filtered, washed thoroughly with cold water, dried and recrystallized from hot ethanol.

*4.2.30. 3-[4-(N,N-dimethyl amino) phenyl]-6-(2-nitro-4-methyl phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**10a**)*

M.p.: 184°C yield (%): 62%; IR (KBr) ν (cm^{-1}): 3084 (aromatic CH stretching), 1606 (C=N stretching), 1587, 1556, 1475, 1451 (C=C ring stretch), 2956, 2845 (methyl CH stretch), 1284 (N–N=C), 1535 (asymmetric Ar–NO_2 stretch), 1339 (symmetric Ar–NO_2 stretch); ^{1}H NMR δ: 6.82, 7.94 (2d, 4H of Ar), 7.74, 8.1 (d, 2H of Ar′), 7.80 (s, 1H of Ar′), 2.62 (s, 3H, CH_3); MS *m/z* [% rel. int.]: 380 [34] (M+). Calcd. (%) for $C_{18}H_{16}N_6O_2S$: C; 56.83, H; 4.24, N; 22.09, S; 8.43. Found: C; 56.79, H; 4.24, N; 22.11, S; 8.44.

*4.2.31. 3-[2-(N-methyl amino) phenyl]-6-(2-nitro-4-methyl phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**10b**)*

M.p.: 180°C yield (%): 60%; IR (KBr) ν (cm^{-1}): 3298 (NH stretching), 3086 (aromatic CH stretching), 1614 (C=N stretching), 1582, 1546, 1479, 1456 (C=C ring stretch), 2976, 2845 (methyl CH stretch), 1280 (N–N=C), 1530 (asymmetric Ar–NO_2 stretch), 1330 (symmetric Ar–NO_2 stretch); ^{1}H NMR δ: 7.3, 7.7 (2d, 2H of Ar), 6.7–6.8 (m, 2H of Ar), 7.8, 8.1 (d, 2H of Ar′), 7.88 (s, 1H of Ar′), 6.42 (q, 1H, NH in NH–CH_3), 1.86 (d, 3H, CH_3 in NH–CH_3), 2.64 (s, 3H, CH_3); MS *m/z* [% rel. int.]: 366 [40] (M+). Calcd. (%) for $C_{17}H_{14}N_6O_2S$: C; 55.73, H; 3.85, N; 22.94, S; 8.75. Found: C; 55.68, H; 3.85, N; 22.93, S; 8.75.

*4.2.32. 3-(1-Naphthyl methyl)-6-(2-nitro-4-methyl phenyl)-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**10c**)*

M.p.: 156°C yield (%): 65%; IR (KBr) ν (cm^{-1}): 3069 (aromatic CH stretching), 1608 (C=N stretching), 1587, 1545, 1463, 1431 (C=C ring stretch), 2916, 2848 (methyl CH stretch), 1525 (asymmetric Ar–NO_2 stretch), 1340 (symmetric Ar–NO_2 stretch); ^{1}H NMR δ: 7.3–7.6 (m, 4H of Ar), 7.8–7.9 (m, 4H, Ar-H), 8.3 (1d, 1H of Ar), 8.10 (d, 1H of Ar′), 2.68 (2s, 6H, CH_3), 4.9 (s, 2H, CH_3); MS (*m/z*, %): 401 (M+); MS *m/z* [% rel. int.]: 401 [38] (M+). Calcd. (%) for $C_{21}H_{15}N_5O_2S$: C; 62.83, H; 3.77, N; 17.45, S; 7.99. Found: C; 62.78, H; 3.77, N; 17.46, S; 7.99.

*4.2.33. Synthesis of 3-aryl/aralkyl substituted-6-[2-(2,4-dichloro phenoxy) ethyl] 1,2,4-triazolo[3,4-b]-1,34-thiadiazoles **11(a–c)***

To the equimolar proportions of respective triazole **3(a–c)**, (0.02 M) and 2-(2,4-dichloro phenoxy) propionic acid (0.02 M), 10 ml of dry phosphorous oxy chloride was added. The resulted mixture was further refluxed continously for 4 h on a water bath. The reaction mixture was cooled to room temperature and then gradually poured onto crushed ice with stirring. Finely powdered potassium carbonate and the required amount of solid potassium hydroxide were added till the pH of the mixture was raised to 8, to remove the excess of phosphorous oxychloride. The mixture was allowed to stand overnight and solid was separated. It was filtered, washed thoroughly with cold water, dried and recrystallized from petroleum ether.

*4.2.34. 3-[4-(N,N-dimethyl amino) phenyl]-6-[2-(2,4-dichloro phenoxy) ethyl]-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**11a**)*

M.p.: 114°C yield (%): 65%; IR (KBr) ν (cm^{-1}): 3085 (aromatic CH stretching), 1611 (C=N stretching), 1584, 1543, 1484, 1452 (C=C ring stretch), 2954, 2842 (methyl CH stretch), 1278 (N–N=C), 1357 (C–N stretching); ^{1}H NMR δ: 6.8, 8.1 (2d, 4H of Ar), 6.98, 7.42 (2d, 2H of Ar′), 7.16 (s, 1H of Ar′), 5.64 (q, 1H, CH in –CH–CH_3), 3.0 (s, 6H, N(CH_3)$_2$), 1.9 (d, 1H, CH_3 in CH–CH_3); MS *m/z* [% rel. int.]: 434 [40] (M+), 436[32] (M + 2), 438 [14] (M + 4). Calcd. (%) for $C_{19}H_{17}N_5OCl_2S$: C; 52.54, H; 3.95, N; 16.12, S; 7.38. Found: C; 52.48, H; 3.95, N; 16.10, S; 7.38.

*4.2.35. 3-[2-(N-methyl amino) phenyl]-6-[2-(2,4-dichloro phenoxy) ethyl]-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**11b**)*

M.p.: 92°C yield (%): 64%; IR (KBr) ν (cm^{-1}): 3086 (aromatic CH stretching), 3308 (NH stretching), 1614 (C=N stretching), 1575, 1549, 1477, 1446 (C=C ring stretch), 2948, 2832 (methyl CH stretch), 1283 (N–N=C), 1361 (C–N stretching); ^{1}H NMR δ: 7.3, 7.7 (2d, 2H of Ar), 8.3, 8.4, 8.8 (3d, 3H of Ar′), 6.7–6.8 (m, 2H of Ar), 5.7 (q, 1H, NH in NH–CH_3), 4.8 (q, 1H, CH in CH–CH_3), 1.9 (d, 3H, CH_3 in CH–CH_3), 1.7 (d, 3H, CH_3 in NH–CH_3); MS *m/z* [% rel. int.]: 420 [38] (M+), 422 [26] (M + 2), 424 [12] (M + 4). Calcd. (%) for $C_{18}H_{15}N_5OCl_2S$: C; 51.44, H; 3.60, N; 16.66, S; 7.63. Found: C; 51.52, H; 3.60, N; 16.67, S; 7.64.

*4.2.36. 3-(1-Naphthyl methyl)-6-[2-(2,4-dichloro phenoxy) ethyl]-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazole (**11c**)*

M.p.: 86°C yield (%): 55%; IR (KBr) ν (cm^{-1}): 3070 (aromatic CH stretching), 1607 (C=N stretching), 1586, 1538, 1477 (C=C ring stretch), 2982, 2929 (methylene CH stretch), 1284 (N–N=C); ^{1}H NMR δ: 6.8, 7.05 (2d, 2H of Ar′), 7.1 (s, 1H of Ar′), 7.3–7.6 (m, 4H of Ar), 7.8, 7.9, 8.3 (3d, 3H of Ar), 5.5 (q, 1H, CH in CH–CH_3), 1.78 (d, 3H, CH_3 in CH–CH_3), 4.9 (s, 2H, CH_2); MS *m/z* [% rel. int.]: 455 [44] (M+), 457 [36] (M + 2), 459 [14] (M + 4). Calcd. (%) for $C_{22}H_{16}N_4OCl_2S$: C;

58.03, H; 3.54, N; 12.30, S; 7.04. Found: C; 57.96, H; 3.54, N; 12.31, S; 7.04.

Acknowledgements

The authors are thankful to IISc, Bangalore, IIT, Chennai and STIC, Cochin, for the Spectral analysis. The authors are also thankful to the Chairman, Department of Chemistry, Kuvempu University, for providing laboratory facilities.

References

[1] D.H. Boschelli, D.T. Connor, D.A. Barnemeier, J. Med. Chem. 36 (1993) 1802.
[2] P.C. Unangst, G.P. Shrum, D.T. Connor, D.R. Dyer, D.J. Schrier, J. Med. Chem. 35 (1992) 3691.
[3] N. Srivastava, S. Bahadur, H.N. Verma, M.M. Khan, Curr. Sci. 53 (1984) 235.
[4] H. Saad, Indian J. Chem. 35B (1996) 980.
[5] X.P. Hui, C.H. Zang, Q. Wang, Q. Zhang, Indian J. Chem. 41B (2002) 2176.
[6] N. Tiwari, B. Chaturvedi, Nizamuddin, Agric. Biol. Chem. 52 (1988) 1229.
[7] A.R. Prasad, T. Ramlingam, A. Bhaskar Rao, P.V. Diwan, P.B. Sattur, Indian J. Chem. 25B (1986) 566.
[8] V. Srivastava, S. Sen, R. Shekar, Indian J. Chem. 33B (1994) 344.
[9] M. Imtiaz Hussain, V. Kumar, Indian J. Chem. 31B (1992) 673.
[10] Z.Y. Zhang, X.W. Sun, C.H. Chu, L. Zhao, J. Chin. Chem. Soc. 44 (1997) 535.
[11] K.C. Joshi, S. Giri, J. Indian Chem. Soc. 40 (1963) 42.
[12] M.C. Goldsworthy, Phytopath 32 (1942) 498.
[13] J. Haglind, Chem. Abstr 64 (1966) 16509.
[14] J.R. Reid, N.D. Heindel, J. Heterocyclic Chem 13 (1976) 925.
[15] C.A. Winter, E.T. Risley, G.W. Nuss, Proc. Soc. Exp. Biol. Med. 3 (1962) 544.
[16] R. Turner, Screening Methods in Pharmacology, Academic Press, New York and London, 1965 (100).