Research Article

Synthesis and Pharmacological Investigations of Azetidinones Involving 3-Mercapto-4-amino-5-naphtho[2,1-*b*]furan-1,2,4-triazole

MN. Kumaraswamy[2], VP. Vaidya[1]*, C. Chandrasekhar[3], DA. Prathima Mathias[4], H. Shivakumar[5] and KM. Mahadevan[1]

[1]Department of PG Studies and Research in Chemistry, Kuvempu University, Shankaraghatta, Karnataka, India
[2]Department of PG Studies in Chemistry, Sir. M.V. Government Science College, Bhadravathi. Karnataka, India[3].
Department of Medicinal Chemistry, S.D.M. College, Ujjere, Karnataka, India.
[4]Department Chemistry, IDSG Govt. College, Chikkamagalur- Karnataka, India.
[5]Department of Pharmacology, SCS College of Pharmacy, Harapanahalli, Karnataka, India.

ABSTRACT

The reaction of naphtho[2,1-b]furan-2-carbohydrazide 1 with carbon disulphide and excess of hydrazine hydrate in ethanol produced 4-amino-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiol 2. The thiol 2 on treatment with aromatic aldehydes yielded 4-{[(4-aryl)methylene]amino}-5-(naphtho[2,1-b]furan-2-yl)-4H-1,2,4-triazole-3-thiols 3a-f. The title compounds, chloro-1-(3-mercapto-5-naphtho[2,1-b]furan-2-yl-1,2,4-triazole-4-yl)-2-(aryl)-azetidin-4-ones 4a-f were obtained by reacting compounds 3a-f with choloro acetyl chloride in presence of triethyl amine. Some of the newly synthesized compounds exhibited potent antibacterial, antifungal, anti-inflammatory, diuretic, anthelmintic and antipyretic activities.

Keywords: Naphtho[2,1-b]furan-2-carbohydrazide, azetidinones, antimicrobial activity.

INTRODUCTION

Azetidin-2-one, a four membered β-lactam skeleton, has been recognized as a useful building block for the synthesis of a large number of organic molecules by exploiting its ring strain[1]. Interesting reports appeared in the literature, where in azetidinone moiety has been connected to benzo[*b*]thiophene and benzothiazole nucleus; these, compounds exhibited significant antimicrobial and antitubercular activity[2-3]. The 2-azetidinone moiety is an essential part of the penicillin skeleton and a substructure found in □-lactamase inhibitors such as clavulanic acid or sulbactam[4]. Penams, cephems, monobactams, penems, carbapenems and triems are several structural variants of □-lactam antibiotics, which have been developed based on penicillin structure as novel approaches to antibacterial therapy[5]. Certain biheterocyclics connected to azetidine ring have been found to possess excellent antibacterial and anti-inflammatory activities[6]. Some of the derivatives of azetidinone were reported to be associated with antifungal[7] anti-herpes[8], and potent cholesterol absorption inhibitory activities[9-11]. The substituted triazoles exhibit broad spectrum of biological activities and medicinal properties. Substituted 1,2,4-triazole derivatives act as potential antimicrobial and antitubercular agents[12], antimicrobial activity[13], CNS depressant, mild hypocholesteremic and hypotensive activities[14], hypoglycemic[15], antiviral[16], insecticidal activities[17] and anti-inflammatory activity[18]. Several derivatives of naphtho[2,1-b]furan derivatives synthesized in our laboratory have been reported to possess many biological and pharmacological activities such as antimicrobial, analgesic, anti-inflammatory, diuretic, anthelmintic, antipyretic etc [19-22].

Encouraged from these facts, and the principle that the combination of two or

more biologically active heterocyclic systems enhances and/or changes the biological profile of molecules, intrigued us to extend our continued efforts to synthesize more potent derivatives of naphtho[2,1-*b*]furan derivatives. We now report the synthesis of novel compounds, 3-chloro-1-(3-mercapto-5-naphtho[2,1-*b*]furan-2-yl-1,2,4-triazloe-4-yl)-2-aryl-azetidin-4-ones 4a-f.

MATERIALS AND METHODS

All the reagents were A. R. grade and used without further purification. Melting points were determined with the open capillary and are uncorrected. IR spectra recorded in KBr pellets by using JASCO FT-IR 300E spectrophotometer. ^{1}H NMR and ^{13}C NMR spectra were recorded in DMSO-d_6 on Bruker Supercon FT-NMR 400 MHz instrument. Chemical shifts are reported in □□(ppm) relative to TMS as internal standard. Mass spectral data were obtained on a Jeol JMS-D 300 Mass spectrometer operating at 70 eV. Elemental analysis was performed using a Vario-EL elemental analyzer. All the reactions were monitored by TLC.

Wistar rats of either sex (150-200 g; National College of Pharmacy, Shimoga, Karnataka) and Swiss mice of either sex (20-30 g; National College of Pharmacy, Shimoga, Karnataka) were used for animal experiments. They were housed in polypropylene shoebox type cages with stainless steel grill top and bedded with rice husk. The animals were provided with pelleted diet (Goldmohur, Lipton India) and water *ad libitum*. They were allowed a one week acclimatization period before the experimental session. All the experimental protocols were met with the approval of Institutional Animal Ethics Committee (Reg. No. 14/1999/CPCSEA/5-7-99). The earthworms were procured from the Department of Environmental Sciences, Kuvempu University, Shankaraghatta, Shimoga, Karnataka).

EXPERIMENTAL

Synthesis of 4-amino-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiol 2.

To a solution of **1** (2.26 g, 0.01 mol) in ethanol (25 ml), carbon disulphide (1.14 g, 1.2 ml, 0.015 mol), and KOH (0.84 g, 0.015 mol) were added and stirred for 16 h at room temperature. The salt obtained was filtered and washed with diethyl ether. The mixture of this salt salt (3.40 g, 0.01 mol) in water (10 ml) and hydrazine hydrate (1.12 g, 1.5 ml) was refluxed for 4 h. The resulting solution was poured into ice cold water and neutralized with concentrated HCl. The product was recrystallized from aqueous DMF. (2.42 g, 86%). m.p. 245 ^{0}C.

Synthesis of 4-{[4-(methoxy phenyl) methylene]amino}-5-naphtho[2,1-b] furan-2-yl-4H-1,2,4-triazole-3-thiols 3b.

A mixture of 4-amino-5-(naphtho[2,1-*b*]furan-2-yl)-4*H*-1,2,4-triazole-3-thiol **2** (1.41 g, 0.005 mol) in dioxane (10 ml), 4-methoxybenzaldehyde (1.17 g, 0.0075 mol) and catalytic amount of conc. hydrochloric acid was refluxed for 2 h. The reaction mixture was poured into ice cold water, solid that separated out was filtered and dried. The product was recrystallised from dioxane. (1.38 g, 69%). m.p. 264 ^{0}C. Similarly the compounds 3a, 3c-f were synthesized by using benzaldehyde, 2-chlorobenzaldehyde, 4-chlorobenzaldehyde, 4-nitrobenzaldehyde and furfural, in place of 4-methoxybenzaldehyde.

Synthesis of 3-chloro-1-(3-mercapto-5-naphtho[2,1-b]furan-2-yl-1,2,4-triazloe-4-yl)-2-(aryl)-azetidin-4-one 4b.

A solution of chloroacetyl chloride (0.6 ml, 0.0055 mol) in dioxane (10 ml) was cooled to –10 ^{0}C using ice-salt bath and kept for stirring. To this, triethylamine (0.5 g, 0.005 mol) was added drop wise maintaining the temperature below 0 ^{0}C. A solution of 5-(naphtho[2,1-*b*]furyl-2-yl)-4-{[(4-methoxy phenyl)methylene]-amino}-3-mercapto-1,2,4-tirazole 3b (1.00 g, 0.0025 mol) in dioxane (10 ml) was then added drop wise to the above reaction mixture regulating the temperature <0 ^{0}C with stirring. After the addition was over the reaction mixture was refluxed for 16 h and then poured into ice cold water to obtain 4b as solid, which was filtered and dried. (0.85 g, 72 %). m.p. 280 ^{0}C.

The compounds 4a, 4c-f were synthesized by the same method. The sequence of

reactions is outlined in scheme-1. The physical and analytical data of the synthesized compounds is presented in Table-1.

Pharmacological investigations

Newly synthesized compounds 4a-f were studied for various pharmacological activities activities.

Antimicrobial activity

The *in vitro* antimicrobial activity was carried out against 24 h old cultures of two bacteria and two fungi by cup-plate method[23]. The compounds 4a-f have been investigated for their antibacterial activity against *Pseudomonas aeruginosa* and *Staphylococcus aureus* and antifungal activity against *Aspergillus niger* and *Curvularia lunata*. Chloramphenicol and fluconazole were used as standards for antibacterial and antifungal activity respectively. The compounds were tested at a concentration of 0.001 mol/ml in DMF against all the organisms. The zone of inhibition was compared with the standard drug after 24 h of incubation at 25 ^{0}C for antibacterial activity and 48 h at 30 ^{0}C for antifungal activity. The results are presented in Table 2.

Anti-inflammatory activity

The anti-inflammatory activity was evaluated by a rat paw edema method. This method is based on plethysmographic measurement of carrageenan-induced acute rat paw edema produced by sub plantar injection of carrageenan in hind paw of the rat[24-25]. Ibuprofen was used as standard and tween-80 (0.1%, 1 ml) solution as control for this study. The percentage inhibition of paw volume was calculated by using the formula

% Inhibition= (1 - Vt/Vc) x 100.

Where,
Vt = Mean increase in the paw volume in test animals group.
Vc = Mean increase in the paw volume in control group.

Analgesic activity

Analgesic activity was determined by the method based on acetic acid induced writhing in mice[26-27]. Acetyl salicylic acid (aspirin) was used as standard and Tween-80 (0.1%) solution as control. The percentage inhibition of writhing was calculated by using the formula

% Inhibition = (1-Nt/Nc) x 100,

Where,
Nt = Mean number of writhing in test animals
Nc = Mean number of writhing in control.
The results of anti-inflammatory activity and analgesic activities are given in Table 3.

Diuretic activity

The diuretic activity was evaluated on albino rats (Wistar strain) by literature method[28]. For this study aqueous solution of tween-80 (0.1%, 5 ml) served as control and frusemide as standard.

Anthelmintic activity

Anthelmintic activity was evaluated by using *Pheritima posthuma* (class-Annelida and order-Oligichaeta). The technique adopted was that described by Giand et al[29-30]. For this study 25 ml of 0.1% Tween-80 prepared in 6% dextrose solution was served as control. Albendazole suspended in 6% dextrose solution served as standard.

Antipyretic activity

The antipyretic activity was carried out on colony bred albino male rats as by a modified yeast induced hyperpyrexia method[31]. Tween–80 was used as control and paracetamol as standard drug. All the values are expressed as mean ± SEM. The results of diuretic, anthelmintic and antipyretic activities are presented in Table 4.

RESULTS AND DISCUSSION

The starting material, naphtho[2,1-*b*]furan-2-carbohydrazide 1 was synthesized by reacting ethyl naphtho[2,-b]furan-2-caroxyalate with hydrazine hydrate. The reaction of 1 with carbon disulphide and hydrazine hydrate in ethanol produced 4-

amino-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiol 2. The structure assigned to 2 has been established by spectral studies: IR (KBr): 3250 cm^{-1} (NH-asymmetric), 3168 cm^{-1} (NH-symmetric), 1600 cm^{-1} (C=N). ^1H NMR (DMSO-d_6): □□6.0 (s, 2H, NH_2); 7.5- 8.5 (m, 7H, ArH); 14.0 (s, 1H, SH); ^{13}C NMR (DMSO-d_6): δ 167.02, 159.98, 152.09, 141.05, 130.19, 128.82, 127.19, 125.28, 123.46, 122.60, 112.85, 112.59, 112.18, 108.91The synthesis of 4-{[4-(methoxyphenyl)methylene]amino}-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiols 3a-f was carried out by refluxing equimolar amount of 4-amino-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiol 2 with various aromatic aldehydes containing electron withdrawing and electron donating groups. The structure assigned to 3b has been established by spectral studies: ^1H NMR (DMSO-d_6): □□3.8 (s, 3H, OCH_3); 4.4 (s, 1H, SH); 7.6-8.6 (m, 11H, ArH); 8.7 (s, 1H, N=CH); 12.1 (s, 1H, NH, tautomeric); IR (KBr): 1597 cm^{-1} (C=N). The spectral data of compounds 3a and 3c-f is presented in Table 5. These hydrazones (3a-f) on treatment with chloroacetyl chloride in presence of triethyl amine at 0 ^0C furnished the desired products, 4-{[4-(aryl)methylene]amino}-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiols 4a-f. The IR spectrum of 4b exhibited the sharp absorption bands at 1657 cm^{-1} due to C=O carbonyl group of □-lactum ring and at 1600 cm^{-1} due to C=N. The ^1H NMR (DMSO-d_6) spectrum of 4b exhibited peaks at □□□□□□(s, 3H, OCH_3,); 4.1 (s, 1H, SH); 4,3 (d, 1H, CHPh); 4.9 (d, 1H, CHCl); 6.9-8.6 (m, 11H, ArH); Similarly ^{13}C NMR (DMSO-d_6) spectra was recorded which showed peaks at: δ 182.88, 131.52, 130.17, 128.85, 128.55, 127.74, 127.62, 127.52, 127.31, 127.08, 125.45, 125.32, 123.79, 123.66, 114.99, 114.52, 114.42, 114.16, 112.51, 112.32, 112.21, 55.68, 8.5, 7.54; MS: 478 (M+2). The spectral data of remaining compounds i.e. 3a, and 3c-f , 4a, and 4c-f is presented in Table 5. The compounds encompassing naphthofuran, triazole and azetidinone ring systems are known to exhibit wide spectrum of biological and pharmacological activities. Hence, it was contemplated to evaluate newly synthesized compounds for antimicrobial, anti-inflammatory, analgesic, diuretic, anthelmintic, and antipyretic activities by adopting literature procedure.

The newly synthesized compounds were evaluated for antimicrobial activity. Zone of inhibition was measured in mm and results are presented in Table 2. The compounds 4b, 4d and 4e displayed significant antibacterial activity. Rest of the compounds exhibited substantial activity against both the organisms. It is observed that electron withdrawing groups resulted in enhancement of activity. The compounds 4b and 4d showed promising antifungal activity, whereas remaining compounds are found to be considerable active. In this case also electron withdrawing groups have much more pronounced effect on antifungal activity.

Anti-inflammatory activity of the synthesized compounds was investigated by carrageenan induced rat paw edema method on albino rats (Wistar strain) using Ibuprofen as a standard drug. The percentage of inhibition of edema was calculated in each case and is presented in Table 3. The compounds 4a-f exhibited excellent activity, having percentage inhibition of 62.90, 65.32, 60.48, 61.29, 63.70 and, 62.09 comparable with that of standard drug having percentage inhibition of 79.59, while rest of the compounds were found to be moderately active. Combination of all the three heterocycles seems to be marked effect on activity. Electron donating methoxy group resulted in increase of activity to a greater extent.

Acetic acid induced writing method was adopted to evaluate analgesic activity of the synthesized compounds. The experiment was carried out on albino mice (Swiss strain) using aspirin as standard and percentage protection was calculated for each compound as well as standard, which is presented in Table-4. The results indicated that compounds 4a-f possess substantial analgesic activity and remaining compounds exhibited significant activity. The activity is independent of the substituent present in the molecule. The mechanism of action of all the tested compounds at present could not be

ascertained and needs further investigation.

Diuretic activity was evaluated on albino rats (Wistar strain) using Frusamide as a standard drug. Lipschitz values were calculated and presented in Table 5. The compounds 4b, 4d and 4e, were found to display promising activity and remaining compounds possessed moderate activity. Presence of electron donating groups in compound 4b may be responsible for the enhanced activity. Diuretics are drugs that increase the rate of urine flow. However, clinically useful diuretics also increase the rate of excretion of Na^+ and accompanying anion, usually Cl^-. The standard drug Frusemide used in this case, contains furan ring in its structure, hence the diuretic effect of the test compounds may due to the presence of naphthofuran moiety in their structures.

The synthesized compounds were screened for anthelmintic activity the time required paralysis and death of the worm were noted in each case. It was observed that none of the compounds exhibited considerable anthelmintic activity. Outer layer of the earthworm is a mucilaginous layer and composed of polysaccharides. This layer, being slimy, enables the earthworms to move freely. Any damage to the mucopolysaccharide membrane will expose the outer layers, and this restricts its movement and can cause paralysis. This action may lead to death of the worm and will be expelled out from the body. None of the compounds seems to have such an effect on earthworms.

Antipyretic activity of the synthesized compounds was determined by yeast induced hyperpyrexia method on albino rats (Wistar strain) using paracetamol as standard drug. Decrease in rectal temperature was recorded in each case. The results indicated that compound 4d exhibited excellent antipyretic activity showing the decrease in temperature to the extent of 0.5 ^{0}C. Rests of the compounds were either moderately active or less active. Chlorine atom which is present in position 4 increased the activity to considerable activity. The test compounds possess a significant antipyretic effect in yeast-induced elevation of body temperature in rats and this may be due to combined anti-inflammatory and analgesic effects.

CONCLUSION

A number of 4-{[4-(aryl)methylene]amino}-5-naphtho[2,1-b]furan-2-yl-4H-1,2,4-triazole-3-thiols 4a-f were synthesized and characterized by analytical and spectral studies. The newly synthesized compounds were evaluated for antibacterial, antifungal, anti-inflammatory, analgesic, diuretic, anthelmintic, and antipyretic activities. The results obtained hitherto indicated, that combination of naphtho[2,1-b]furan, triazole and azetidinone ring systems enhances the activity to a considerable extent. In many cases, presence of electron withdrawing groups results in increase of activity and in few cases electron donating methoxy group has marked influence in enhancing activity.

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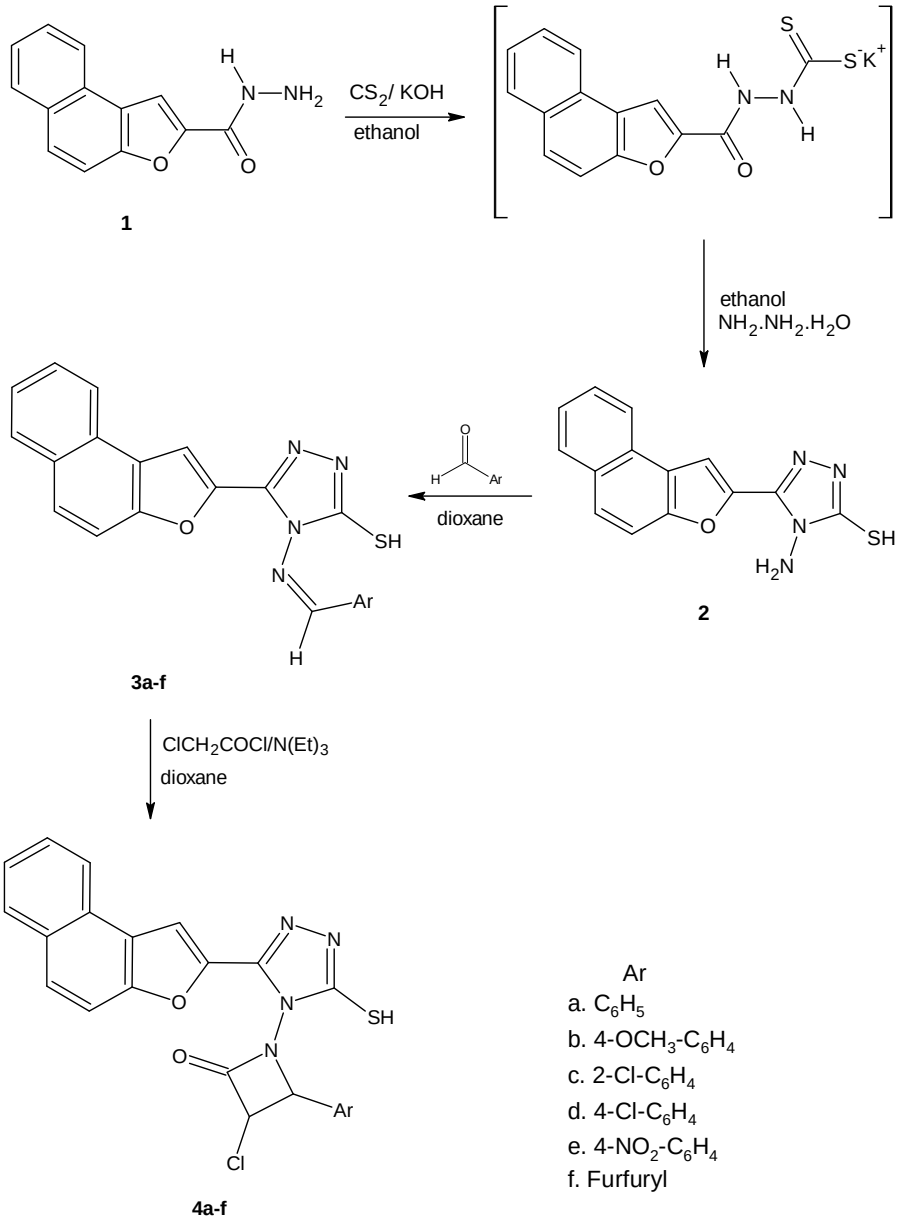

1

2

3a-f

4a-f

Ar

a. C_6H_5
b. 4-OCH_3-C_6H_4
c. 2-Cl-C_6H_4
d. 4-Cl-C_6H_4
e. 4-NO_2-C_6H_4
f. Furfuryl

Scheme 1:

Table 1: Physical and analytical data of the synthesized compounds

Comp.	R	M.p. ^{0}C	Yield (%)	Mol. Formula	Found (Calcd) %		
					C	H	N
3a	-C_6H_5	260	65	$C_{21}H_{14}N_4OS$	67.88 (68.09)	3.69 (3.81)	15.03 (15.12)
3b	4-OCH_3-C_6H_4	264	69	$C_{22}H_{16}N_4O_2S$	65.8 (65.9)	3.9 (4.0)	13.8 (13.9)
3c	2-Cl- C_6H_4	268	64	$C_{21}H_{13}N_4OClS$	62.21 (62.30)	3.15 (3.24)	13.77 (13.84)
3d	4-Cl- C_6H_4	271	61	$C_{21}H_{13}N_4OClS$	62.19 (62.30)	3.18 (3.24)	13.69 (13.84)
3e	4-NO_2- C_6H_4	275	58	$C_{21}H_{13}N_5O_3S$	60.59 (60.71)	3.06 (3.15)	16.79 (16.86)
3f	Furfural	257	56	$C_{19}H_{12}N_4O_2S$	63.19 (63.32)	3.28 (3.36)	15.49 (15.55)
4a	C_6H_5	274	68	$C_{23}H_{15}N_4O_2ClS$	61.69 (61.81)	3.29 (3.38)	12.39 (12.54)
4b	4-OCH_3- C_6H_4	280	72	$C_{24}H_{17}N_4O_3ClS$	60.2 (60.4)	3.4 (3.5)	11.5 (11.7)
4c	2-Cl- C_6H_4	284	60	$C_{23}H_{14}N_4O_2Cl_2S$	57.28 (57.39)	2.79 (2.93)	11.49 (11.64)
4d	4-Cl- C_6H_4	286	63	$C_{23}H_{14}N_4O_2Cl_2S$	57.29 (57.39)	2.78 (2.93)	11.48 (11.64)
4e	4-NO_2- C_6H_4	292	66	$C_{23}H_{14}N_5O_4ClS$	56.09 (56.16)	2.78 (2.87)	14.09 (14.24)
4f	Furfural	266	59	$C_{21}H_{13}N_4O_3ClS$	57.59 (57.73)	2.89 (3.00)	12.69 (12.82)

Table 2: Antimicrobial activity data of the compounds 4a-f

Compd.	Zone of Inhibition in mm			
	Antibacterial activity		Antifungal activity	
	P. aerugenosa	*S.aureus*	*A. niger*	*C. lunata*
Standard	24	26	24	22
4a	15	15	16	15
4b	18	19	18	17
4c	16	17	17	16
4d	17	18	17	17
4e	18	18	16	17
4f	16	16	15	16

Table 3: Anti-inflammatory and Analgesic activity of the compounds 4a-f

Compd.	Anti-inflammatory activity		Analgesic activity
	Group	Inhibition (%) of edema after 3 hrs	% Protection
Control	I	-----	-----
Standard	II	79.59	71.05
4a	III	62.90	58.29
4b	IV	65.32	60.00
4c	V	60.48	56.41
4d	VI	61.29	59.19
4e	VII	63.70	60.97
4f	VIII	62.09	55.77

Table 4: Diuretic, Anthelmintic and Antipyretic activities of the compounds 4a-f

Compd.	Group	Diuretic activity	Anthelmintic activity		Antipyretic activity		
		T/S (Lipschitz value)	Time in minutes		Mean rectal temperature		Decrease in temperature
			Mean time of paralysis	Mean death time	0 hr	3 hr	
Control	I	0.27	------	-----	38.7	38.5	0.2
Standard	II	1.00	33	46	38.4	37.7	0.7
4a	III	0.55	135	231	37.7	37.3	0.4
4b	IV	0.65	105	150	37.8	37.4	0.4
4c	V	0.58	125	235	---	---	---
4d	VI	0.62	125	225	37.6	37.1	0.5
4e	VII	0.62	111	214	37.5	37.1	0.4
4f	VIII	0.58	140	241	**---**	**---**	**---**

Table 5: The spectral data of remaining compounds. 3a, 3c-f, 4a and 4c-f

Comp.	Ar	IR (KBr) cm^{-1}		^1H NMR in ppm
		C=N	C=O	
3a	- C_6H_5	1585	-----	□□4.8 (s, 1H, SH), δ 7.4-8.5 (m, 12H, ArH), □ 8.7 (s, 1H, NCH), □ 12.0 (s, 1H, NH)
3c	2-Cl- C_6H_4	1620	-----	□□4.9 (s, 1H, SH), δ 7.6-8.5 (m, 11H, ArH), □ 8.9 (s, 1H, NCH), □ 12.2 (s, 1H, NH)
3d	4-Cl- C_6H_4	1610	-----	□□4.5 (s, 1H, SH), δ 7.3-8.4 (m, 11H, ArH), □ 8.6 (s, 1H, NCH), □ 11.9 (s, 1H, NH)
3e	4-NO_2- C_6H_4	1598	-----	□□4.6 (s, 1H, SH), δ 7.5-8.5 (m, 11H, ArH), □ 8.8 (s, 1H, NCH), □ 12.3 (s, 1H, NH)
3f	Furfural	1609	-----	□ 4.6 (s, 1H, SH), δ 7.5-8.3 (m, 9H, ArH), □ 8.5 (s, 1H, NCH), □ 11.9 (s, 1H, NH)
4a	- C_6H_5	1585	1673	δ 4.3 (s, 1H, SH), δ 4.5 (d, 1H, CHCPh), δ 4.8 (d, 1H, CHCl), δ 7.4-8.6 (m, 12H, ArH)
4c	2-Cl- C_6H_4	1606	1670	δ 4.5 (s, 1H, SH), δ 4.6 (d, 1H, CHCPh), δ 4.8 (d, 1H, CHCl), δ 7.4-8.4 (m, 11H, ArH)
4d	4-Cl- C_6H_4	1610	1660	δ 4.1 (s, 1H, SH), δ 4.4 (d, 1H, CHCPh), δ 4.8 (d, 1H, CHCl), δ 7.2-8.6 (m, 11H, ArH)
4e	4-NO_2- C_6H_4	1598	1666	δ 4.2 (s, 1H, SH), δ 4.3 (d, 1H, CHCPh), δ 4.7 (d, 1H, CHCl), δ 7.1-8.5 (m, 11H, ArH)
4f	Furfural	1592	1665	δ 4.3 (s, 1H, SH), δ 4.6 (d, 1H, CHCPh), δ 4.9 (d, 1H, CHCl), δ 7.1-8.0 (m, 10H, ArH).

REFERENCES

1. Duane AB. “β-Lactum cholesterol absorption inhibitors”. Current Medicinal Chemistry. 2004;11:1873-1887.
2. Vasoya SL, Patel MR and Dobaria SV, Joshi HS. Facile synthesis of new azetidinones and acetyl oxadiazoles bearing benzo[b]thiophene nuceus as potent biological active agent. Indian J Chem. 2005;44B:405-409.
3. Chavan AA and Pai NR. Synthesis and Biological Activity of N-Substituted-3-chloro-2-azetidinones. Molecules. 2007;12:2467-2477 .
4. George GI. Ed, The Organic Chemistry of β –Lactams. VCH Publishers, Newyork. 1993; 295-368.
5. Niccolai D, Tarsi L and Thomas RJ. The renewed challenge of antibacterial Chemotherapy. Chem. Commun. 1997;24:2333-2342.
6. Parmar JM, Khunt RC and Parikh AR. Azetidinone derivatives from 2-hydrazino pyrimidine. J Insti Chemists. 2001;73:133-135.
7. Jaish L and Srivastava SK. Synthesis and antimicrobial activity of some new N-methyl piperazinyl thiadiazoles and their azetidinone derivatives. J Sci and Indus Res. 2001;60:331-335.
8. Bonneau PR, Hasani F and Plouffe C et al. Inhibition of human cytomegalovirus protease by monocyclic β -lactum derivatives:

kinetic characterization using a fluorescent probe. J Am Chem Soc. 1999;121:2965-2973.
9. Annunziata R, Benaglia M, Cinquini N and Cozzi F. Stereoselective synthesis of 2-azetidinones as cholesterol-absorption inhibitors. Tetrahedron Asymmetry. 1999;10:4841-4849.
10. Rosenblum SB, Huynh T, Ayonso A and Davis HR. Synthesis of 3-arylpropenyl, 3-arylpropynyl and 3-arylpropyl 2-azetidinones as cholesterol absorption inhibitors: Application of the palladium catalysed arylation of alkenes and alkynes. Tetrahedron. 2000;56:5735-5742.
11. Burnett DA, Caplen MA, Domalski MS, Browne ME, Davis HR and Clader JW. Synthesis of fluorescent biochemical tools related to the 2-azetidinoone class of cholesterol absorption inhibitors. Bioorg Med Chem Lett 2002;12:315-318.
12. Bhat AR, Varadaraj Bhat G and Gautham Shenoy G. Synthesis and invitro antimicrobial activity of new 1,2,4-triazoles. J Pharm Pharmacol. 2001;53:267- 272.
13. Shobha RD, Laddi U, Rajani SB, Patil PA and Bennur S. Synthesis and pharmacological activities of some new 3-Substituted-4-Amino-5 Mercapto-1,2,4-triazoles. Indian J Pharm Sci. 2011;73(1):115-120.
14. Neeraj Upmanyu, Jeetendra Kumar Gupta, Kamal Shah and Pradeep Mishra. Anti-inflammatory and antinociceptive evaluation of newly synthesized 4-(substituted ethanoyl) amino-3-mercapto-5-(4-methoxy) phenyl-1,2,4-triazoles. J Pharm Bioallied Sci. 2011;3(2):259–265.
15. Mhasalkar MY, Shah MH, Pilankar PD, Nikam ST, Anantanarayanan KG and Deliwala CV. Synthesis and hypoglycemic activity of 3-aryl(or pyridyl)-5-alkyl(or aryl)amino-1,3,4-thiadiazoles and some sulfonylurea derivatives of 4H-1,2,4-triazoles. J Med Chem. 1971;14(10):1000-1003.
16. Al-Soud YA, Al-Dweri MN and Al-Masoudi NA. Synthesis, antitumor and antiviral properties of some 1,2,4-triazole derivatives. IL. Farmaco. 2004; 59(10):775-783.
17. Nidhi Goutam and Chourasia OP. Synthesis, antimicrobial and insecticidal activity of some 4H-1,2,4-triazole derivatives. Indian J Chem. 2010;49B:956-959.
18. Chandramouli1 MR, Shivanand, Nayanbhai TB, Bheemachari and Udupi RH. Synthesis and biological screening of certain new triazole Schiff bases and their derivatives bearing substituted benzothiazole moiety. J Chem Pharm Res. 2012;4(2):1151-1159.
19. Kumaraswamy MN, Prathima Mathias DA, Chandrashekhar C and Vaidya VP. Synthesis and pharmacological evaluation of 2-mercapto-4-substituted-naphtho [2,1-b]furo[3,2-d]pyrimidines. Indian J Pharma Sci. 2006;68(6):731-735.
20. Kumaraswamy MN, Prathima Mathias DA, Chandrashekhar C, Shivakumar H, Mahadevan KM and Vaidya VP. Novel approach towards the synthesis of 2-(1-naphtho[2,1-b]furan-2-ylcarbonyl)-3,5-disubstituted-2,3-dihydro-1H-pyrazoles: As a new class of antimicrobial and pharmacological agents. Indian J Pharma Sci. 2008;70(6):715-720.
21. Nagaraj GK, Prakash GK, Kumaraswamy MN, Vaidya VP and Mahadevan KM. Synthesis of novel nitrogen containing naphtho[2,1-b]furan derivatives and investigation of their antimicrobial activities. Arkivoc. 2006; XV; 160-168.
22. Vaidya VP, Shruthi E and Yamuna AJ. Synthesis of Substituted Urea Derivatives Encompassing Naphtho[2,1-b]Furan and Evaluation of Their Antimicrobial Activity. Res J. Pharma Bio Chem Sci 2011; 2(4); 35-43.

23. Indian Pharmacopoeia. Antimicrobial. Controller of Publications, Delhi. 1996; 2: 100-106.
24. Ghosh MN and Singh H. Inhibitory effect of a pyrrolizidine alkaloid, crotalaburnine, on rat paw edema and cotton pellet granuloma. Brit J Pharmacol. 1974;51:503-508.
25. Rashad S, Hemingway A and Rinford K. Effect of Nansteroidal anti-inflammatory Drugs on the course of Osteoarthritis. Lancet. 1989;2:519-522.
26. Shodhi SM, Shubhi Jain, Monica D, Rakesh Shukla and Ram Raghubir. One pot synthesis of pyrimidine and bispyrimidine derivatives and their evaluation for anti-inflammatory and analgesic activities. Bioorg Med Chem. 2007;15: 3334–3344
27. Saundane AR, Rudresh K, Satyanarayana ND and Hiremath SP. Pharmacological Screening of 6H,11H-indol[3,2-c]isoquinolin-5-ones and their Derivatives. Indian J Pharm Sci. 1998;60(6):379-383.
28. Lipschitz WL, Hadidian and Kerpcer A. Bioassay of Diuretics. J Pharmacol Expand Ther 1943;79:97-110.
29. Gaind KN, Mital HC and Khanna SR. Anthelmintic activity of Triphala. Indian J Pharm. 1964;26:106-107.
30. Gaind KN and Budhiraja RD, Antibacterial and anthelmintic activity of Withania coagulans Dunal. Indian J Pharm 1967; 29: 185–186.
31. Hukkeri VI, Patil BS, Savadi RV and Nagathan CV. Analgesic, Antipyretic and Diuretic Activities of Basella rubra Linn. Indian Drugs. 2004;41:536–539.