Available online at www.pelagiaresearchlibrary.com

Pelagia Research Library

Der Chemica Sinica, 2011, 2 (3):147-153

Pelagia Research Library
ISSN: 0976-8505
CODEN (USA) CSHIA5

Synthesis, characterization and biological studies of novel azetidinones

R. R. Mishra[a*], K. S. Nimavat[b] and K. B. Vyas[c]

[a]*M/S Shree Tirupati Industries, Palanpur, Gujarat (India)*
[b]*Government Science College, Gandhinagar, Gujarat (India)*
[c]*Sheth L. H. Science College, Mansa, Gujarat (India)*

ABSTRACT

2-(5-(phenoxy methyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide(1) undergoes facile condensation with aromatic aldehydes to afford the corresponding N'-aryl-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide (2a-h) in good yield. Cyclocondensation of compounds (2a-h) with chloro acetyl chloride yields N-(3-chloro-2-aryl-4-oxoazetidin-1-yl)-2-(5-(phenoxy methyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetamide (3a-h). The structures of these compounds were established on basis of analytical and spectral data. The newly synthesized compounds were evaluated for their antibacterial and antifungal activities.

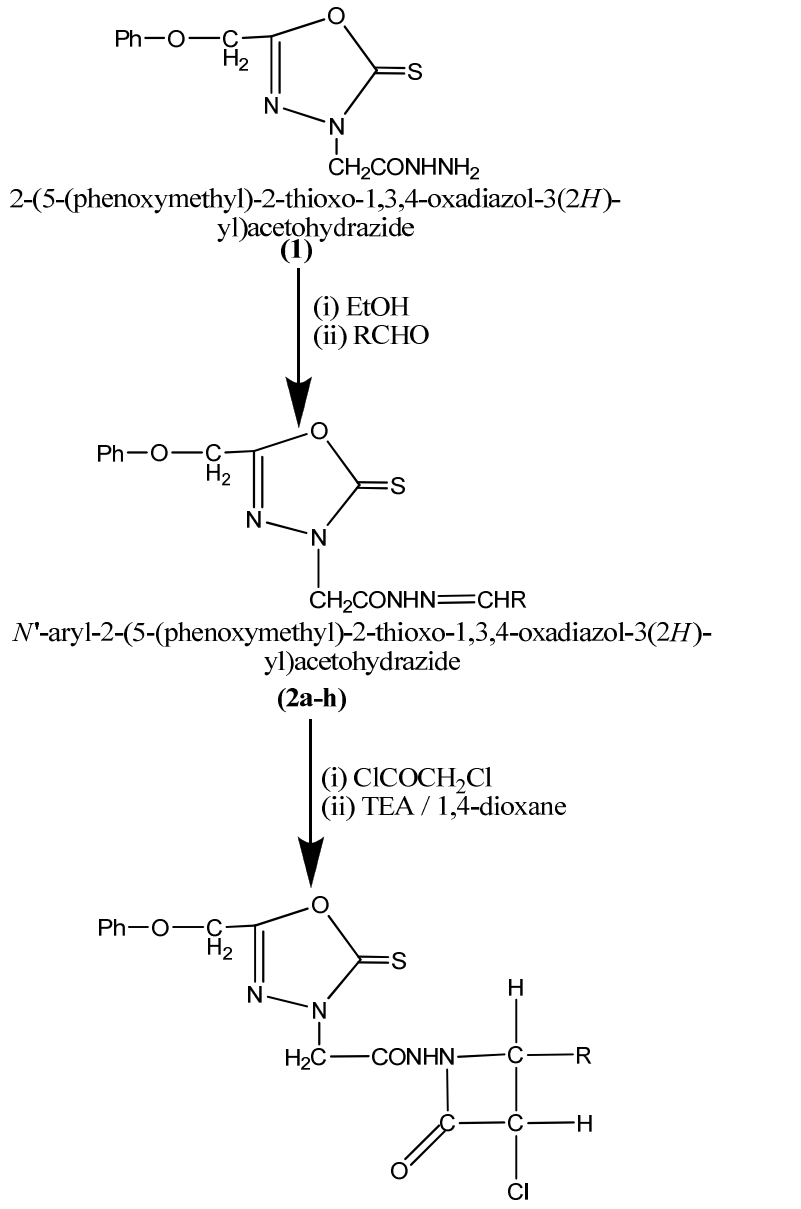
Keywords: 2-(5-(phenoxy methyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)aceto hydrazide, azetidinone, antibacterial activity, spectral studies.

INTRODUCTION

A large number of azetidinones containing β-lactam rings [1-5] are known to exhibit various biological activities like antibacterial, antifungal [6] and antibiotic [7] activities. More particularly and recently these types of compounds have been found in the treatment of T.B. and other chemotherapeutic diseases. Some azetidinons can also be prepared from hydazide derivaties, play medicinal activity like antibacterial, antifungicidal, analgesic, anti-inflammatory activity [8-22]. One of the hydrazide of 1,3,4-oxadiazole moiety can also give biological activity as 1,3,4-oxadiazole derivatives have also biological activity[23-25].Hence, it was thought of interest in merging of both azetidinone and 1,3,4-oxadiazole moieties may enhance the drug activity of compounds up to some extent or might posses some of the above mentioned biological activities. From this point of view, the objective of the present work is to prepare new derivatives of 1,3,4-oxadiazole containing an azetidinone moiety. Hence the present communication comprises the synthesis of N-(3-chloro-2-aryl-4-oxoazetidin-1-yl)-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetamide. The research work is scanned in **scheme-1**.

MATERIALS AND METHODS

Melting points were determined in open capillary tubes and were uncorrected. The IR spectra were recorded in KBr pellets on a Nicolet 760D spectrometer and ^{1}H NMR and ^{13}C NMR spectra were recorded in DMSO with TMS as internal standard on a Brucker spectrometer at 400 MHz and 100 MHz, respectively. LC-MS of selected samples taken on LC-MSD-Trap-SL_01046.



2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2*H*)-yl)acetohydrazide

(1)

N'-aryl-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2*H*)-yl)acetohydrazide

(2a-h)

N-(3-chloro-2-aryl-4-oxoazetidin-1-yl)-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2*H*)-yl)acetamide

(3a-h)

Scheme – 1

Where, R = (a) C_6H_5 (b) 4-OCH_3-C_6H_4 (c) 4-OH-C_6H_4

(d) 2-OH-C_6H_4 (e) 4-CH_3-C_6H_4 (f) 3,4-CH_2O_2-C_6H_4

(g) 4-OH-3-OCH_3-C_6H_3 (h) 3,4-C_2H_5-C_6H_4

The compound 2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl) aceto hydrazide (1) prepared in two steps. First steps involve the reaction between 4-(phenoxy methyl) benzohydrazide with CS_2 in presence of base yields 5-(phenoxymethyl)-1,3,4-oxadiazole-2(3H)-thione, by reported method.[26-28] Then 5-(phenoxymethyl)-1,3,4-oxadiazole-2(3H)-thione treated with bromoethyl acetate followed by hydrazine hydrate gives 2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide (1).

Table: 1 Analytical Data and elemental analysis of compounds (2a-h)

Compd.	Molecular formula (Mol. wt.)	Yield	M.P. °C	Elemental Analysis							
				%C		% H		%N		%S	
				Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.
2a	$C_{18}H_{16}N_4O_3S$ (368)	87	235	58.6	58.68	4.3	4.38	15.2	15.21	8.6	8.70
2b	$C_{19}H_{18}N_4O_4S$ (398)	80	242	57.2	57.27	4.5	4.55	14.0	14.06	8.0	8.05
2c	$C_{18}H_{16}N_4O_4S$ (384)	78	238	56.2	56.24	4.1	4.20	14.5	14.57	8.3	8.34
2d	$C_{18}H_{16}N_4O_4S$ (384)	83	236	56.2	56.24	4.1	4.20	14.5	14.57	8.3	8.34
2e	$C_{19}H_{18}N_4O_3S$ (382)	79	243	59.6	59.67	4.7	4.74	14.6	14.65	8.3	8.38
2f	$C_{19}H_{16}N_4O_5S$ (412)	82	245	55.3	55.33	3.9	3.91	13.5	13.58	7.7	7.77
2g	$C_{19}H_{18}N_4O_5S$ (414)	78	245	55.0	55.06	4.3	4.38	13.5	13.52	7.7	7.74
2h	$C_{22}H_{24}N_4O_3S$ (424)	74	256	62.2	62.24	5.6	5.70	13.1	13.20	7.5	7.55

Table: 2 Spectral data of compounds (2a-h)

^{1}H NMR (δ, ppm)								
Compd.	**Ar-H**	**-CONH**	**-N=CH**	**-CH_3**	**-OCH_3**	**-OH**	**-OC_2H_5**	**-OCH_2O- cyclic**
2a	6.9–8.1 (m, 10H)	11.80(s)	8.4(s)	-	-	-	-	-
2b	6.9–8.1 (m, 9H)	11.80(s)	8.4(s)	-	3.9(s)	-	-	-
2c	6.9–8.1 (m, 9H)	11.80(s)	8.4(s)	-	-	11.20(s)	-	-
2d	6.9–8.1 (m, 9H)	11.80(s)	8.8(s)	-	-	11.20(s)	-	-
2e	6.9–8.1 (m, 9H)	11.80(s)	8.4(s)	2.4(s)	-	-	-	-
2f	6.9–8.1 (m, 8H)	11.80(s)	8.4(s)	-	-	-	-	6.09 2H (s)
2g	6.9–8.1 (m, 8H)	11.80(s)	8.4(s)	-	3.9(s)	11.20(s)	-	-
2h	6.9–8.1 (m, 8H)	11.80(s)	8.4(s)	-	-	-	4.0, 4H, (q,) (CH_2) 1.33, 6H, (t) (CH_3)	-

Table: 3 Analytical data and elemental analysis of Compounds (3a-h)

Compd.	Molecular formula (Mol. wt.)	Yield	M.P. °C	Elemental Analysis							
				%C		% H		%N		%S	
				Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.
3a	$C_{20}H_{17}ClN_4O_4S$ (444)	60	239	53.9	53.99	3.8	3.85	12.5	12.59	7.2	7.21
3b	$C_{21}H_{19}ClN_4O_5S$ (474)	62	250	53.1	53.11	4.0	4.03	11.7	11.80	6.7	6.75
3c	$C_{20}H_{17}ClN_4O_5S$ (460)	59	242	52.0	52.12	3.7	3.72	12.1	12.16	6.9	6.96
3d	$C_{20}H_{17}ClN_4O_5S$ (460)	59	242	52.1	52.12	3.7	3.72	12.1	12.16	6.9	6.96
3e	$C_{21}H_{19}ClN_4O_4S$ (458)	51	246	54.9	54.96	4.1	4.17	12.1	12.21	6.9	6.99
3f	$C_{21}H_{17}ClN_4O_6S$ (488)	53	249	51.5	51.59	3.4	3.50	11.4	11.46	6.5	6.56
3g	$C_{21}H_{19}ClN_4O_6S$ (490)	57	252	51.3	51.38	3.9	3.90	11.4	11.41	6.5	6.53
3h	$C_{24}H_{25}ClN_4O_4S$ (500)	51	261	57.5	57.54	5.0	5.03	11.1	11.18	6.3	6.40

Table: 4 Spectral data of compounds (3a-h)

	^{1}H NMR (δ, ppm)								
Compd.	**C_2-H**	**C_3-H**	**Ar-H**	**-CH_3**	**-OCH_3**	**-OH**	**-OC_2H_5**	**-CONH**	**-OCH_2O- cyclic**
3a	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 10H)	-	-	-		7.8(s)	-
3b	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 9H)	-	3.9(s)	-		7.8(s)	-
3c	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 9H)	-	-	11.20(s)		7.8(s)	-
3d	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 9H)	-	-	11.20(s)		7.8(s)	-
3e	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 9H)	2.4(s)	-	-	-	7.8(s)	-
3f	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 8H)	-	-	-	-	7.8(s)	6.09 2H (s)
3g	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 8H)	-	3.9(s)	11.20(s)	-	7.8(s)	-
3h	5.0 (d, 1H)	5.5 (d, 1H)	6.9–7.6 (m, 8H)	-	-	-	4.0, 4H, (q) (CH_2) 1.33, 6H, (t) (CH_3)	7.8(s)	-

Preparation of N'-aryl-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide ***(2a-h)***

General procedure: – An equimolecular mixture of 2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide (1), (0.01mole) and the aromatic aldehydes (a-h) in ethanol (15ml) was refluxed on a water bath for 1-2 hrs. The solid separated was collected by filtration, dried and recrystallized from ethanol. The yields, melting points and other characterization data of these compounds are given in **Tables -1** & **2**.

Preparation of N-(3-chloro-2-aryl-4-oxoazetidin-1-yl)-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetamide ***(3a-h)***

General procedure: A mixture N'-aryl-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide (2a-h) (0.002 mole) and triethyl amine (TEA) (0.004 mole) was dissolved in 1,4-dioxane (50 ml), cooled, and stirred. To this well-stirred cooled solution chloro acetyl chloride (0.004 mole) was added drop wise within a period of 30 minutes. The reaction mixture

was then stirred for an additional 3 hours and left at room temperature for 48 hours. The resultant mixture was concentrated, cooled, poured into ice-cold water, and then air-dried. The product thus obtained was purified by column chromatography over silica gel using 30% ethyl acetate: 70% benzene as eluent. Recrystallization from ether/n-hexane gave white powered of N-(3-chloro-2-aryl-4-oxoazetidin-1-yl)-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetamide (3a-h), which was obtained in 53-72% yield. All the compounds were characterized by analytical and spectral data (**Table -3** & **4**) of the compounds is assigned in **scheme-1**.

BIOLOGICAL SCREENING

Antibacterial Activities

Antibacterial activities of all the compounds were studied against gram-positive bacteria (*Staphylococcus aureus and Bacillus subtilis*) and gram-negative bacteria (*E. coli, and klebsiella promioe*) at a concentration of 50μg/ml by agar cup plate method. Methanol system was used as control in this method. Under similar condition using tetracycline as a standard for comparison carried out control experiment. The area of inhibition of zone measured in mm. Compounds 3c, 3f and 3g were found more active against the above microbes. Other compounds found to be less or moderate active than tetracycline (**Table -5**).

Antifungal Activities

The fungicidal activity of all the compounds was studied at 1000 ppm concentration in vitro. Plant pathogenic organisms used were *Nigrospora Sp, Aspergillus niger, Botrydepladia thiobromine, and Rhizopus nigricum***,** *Fusarium oxyporium*. The antifungal activity of all the compounds (3a-h) was measured on each of these plant pathogenic strains on a potato dextrose agar (PDA) medium. Such a PDA medium contained potato 200gm, dextrose 20gm, agar 20gm and water one liter. Five days old cultures were employed. The compounds to be tested were suspended (1000ppm) in a PDA medium and autoclaved at 120^{o} C for 15 min. at 15atm.pressure. These medium were poured into sterile Petri plates and the organisms were inoculated after cooling the Petri plates. The percentage inhibition for fungi was calculated after five days using the formula given below:

$$\textbf{Percentage of inhibition} = 100(X-Y) / X$$

Where, X = Area of colony in control plate
Y = Area of colony in test plate

The fungicidal activity displayed by various compounds (3a-h) is shown in **Table - 6**.

Table: 5 Antibacterial Activity of Compounds (3a-h)

Compounds	Gram -Ve		Gram +Ve	
	E. coli	***Klebsiella promioe***	***Bacillus subtilis***	***Staphylococcus aureus***
3a	69	55	59	64
3b	58	66	65	65
3c	72	57	73	69
3d	63	64	63	58
3e	69	68	54	58
3f	67	62	76	74
3g	63	65	69	75
3h	64	47	67	72
Tetracycline	78	86	79	67

Table: 6 Antifungal Activity of Compounds (3a-h)

Zone of Inhibition at 1000 ppm (%)					
Compounds	***Aspergillus niger***	***Botrydepladia thiobromine***	***Fusarium oxyporium***	***Nigrospora Sp.***	***Rhizopus nigricum***
3a	64	65	72	64	54
3b	59	66	69	63	68
3c	62	68	72	59	64
3d	48	66	67	68	75
3e	67	67	63	63	72
3f	56	68	66	67	64
3g	62	74	62	60	57
3h	68	72	66	65	74

RESULTS AND DISCUSSION

It was observed that 2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl) acetohydrazide (1) on condensation with aromatic aldehydes to yield N'-aryl-2-(5-(phenoxymethyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl)acetohydrazide (2a-h).The structures of (2a-h) were confirmed by elemental analysis and IR spectra showing absorption band at 1628-1645(C=N), 3020-3080 cm^{-1} (C-H, of Ar.), 1720-1750 cm^{-1} (-CO), 2815-2850 cm^{-1} (-OCH_3),3450-3485cm^{-1}(-OH),2950, 1370 cm^{-1} (-CH_3),1185(C=S),1620(C=N ring),765(C-O-C ring). The C, H, N analysis and ^{1}H NMR data of all compounds are presented in **Table -1** & **2**.

The cyclocondensation of (2a-h) with chloroacetylchloride resulted in formation of N-(3-chloro-2-aryl-4-oxoazetidin-1-yl)-2-(5-(phenoxy methyl)-2-thioxo-1,3,4-oxadiazol-3(2H)-yl) acetamide (3a-h). The structures assigned to (3a-h) were supported by the elemental analysis and IR spectra showing absorption bands at 1750-1760 (C=O of monocyclic β-lactam), 3035-3090 cm^{-1} (C-H, of Ar.), 3450-3550 cm^{-1} (-OH), 2820-2850 cm^{-1} (-OCH_3), 2950, 1370 cm^{-1} (-CH_3), 1185(C=S),1620(C=N ring),765(C-O-C ring). The C, H, N analysis and ^{1}H-NMR data of all compounds are presented in **Table -3** & **4.**

The examination of data reveals that the elemental contents are consistence with the predicted structure shown in **scheme-1**.The IR data also direct for assignment of the predicted structure. The final structure of all compounds is confirmed by LC-MS data of selected samples. The LC-MS of samples 3b and 3e give the molecular ion peak (m/z) at 489 and 472 respectively. These values are corresponds to their molecular weight.

Acknowledgement
The authors are thankful to m/s Shree Tirupati Industries, Palanpur, Gujarat, India for providing chemicals and laboratory facilities for research work.

REFERENCES

[1] H. T. Clarke, J. R. Johnson, R. Robinson, *The chemistry of penicillin*, Princeton uni. Press, Princeton. **1949**.
[2] H. S. Patel, H. J. Mistry, H. D. Desai, *Oriental J. chem.* **2003**,19(1),187-192.
[3] H. S. Patel, H. J. Mistry, *Phosphorous, Sulfur and Silicon*,**2004,**179,1085-1093.
[4] H. S. Patel, V. K. Patel, *Ind. J. heterocyclic chem.*, **2003,**12,253-256.
[5] K. R. Desai, Bhanvesh Naik, *Ind.J.chem.*, **2006,**45(B),267-271.
[6] V. V. Mulwad, B. P. Choudhari, *Indian J. heterocyclic chem.*, **2003,**12,197-200.
[7] K. Akiba, M. Wada, *Chem. Abstr.*, **1989,**111,96964b.

[8] M. R. Shiradkar, K. K. Murahari, H. R. Gangadasu, T. Suresh, C. C. Kalyan, D. Panchal, R. Kaur, P. Burange, J.Ghogare, V.Mokale, M.Raut, *Bioorg.Med. Chem.*,**2007,** 15, 3997.
[9] Y. Janin, *Bioorg. Med. Chem.**2007,*** 15, 2479.
[10] E. Gursoy, N. Guzeldemirci-Ulusoy, *Eur. J. Med. Chem.* **2007,** 42, 320.
[11] M. R. Rao, K. Hart, N. Devanna and K. B. Chandrasekhar, *Asian J. Chem.* **2008,** 20, 1402.
[12] K. B. Kaymakcıoglu, E. E. Oruc, S. Unsalan, F. Kandemirli, N. Shvets, S. Rollas, D. Anatholy, *Eur. J. Med. Chem.***2006,**.41, 1253.
[13] R. Kalsi, M. Shrimali, T. N. Bhalla, J. P. Barthwal, *Indian J. Pharm. Sci.* **2006, 41,** 353.
[14] S. Gemma, G. Kukreja, C. Fattorusso, M. Persico, M. Romano, M. Altarelli,L. Savini, G. Campiani, E. Fattorusso, N. Basilico, *Bioorg. Med. Chem. Lett.* **2006,**16, 5384.
[15] D. Sriram, P. Yogeeswari, K. Madhu, *Bioorg. Med. Chem. Lett.* **2006,** 15, 4502.
[16] A. Nayyar, R. Jain, *Curr. Med. Chem.* **2006,** 12, 1873.
[17] R. M. Fikry, N. A. Ismael, A. A. El-Bahnasawy, A. A. Sayed El-Ahl., *Phosphorus Sulfur and Silicon.* **2006,** 179, 1227.
[18] A. Walcourt, M. Loyevsky, D. B. Lovejoy, V. R. Gordeuk, D. R. Richardson,*Int. J. Biochem. Cell Biol.* **2004,** 36, 401 .
[19] M. G. Mamolo, V. Falagiani, D. Zampieri, L. Vio, E. Banfi , G. Scialino,*Farmaco***2003,** *58*, 631.
[20] N. Terzioglu, A. Gursoy, *Eur. J. Med. Chem.*, **2003,** 38, 781.
[21] S. G. Kucukguzel, E. E. Oruc, S. Rollas, F. Sahin, A. Ozbek, *Eur. J. Med. Chem.* **2002,** 37, 197.
[22] S. Rollas, N. Gulerman, H. Erdeniz, *Farmaco*, **2002,**57, 171.
[23] Al- Mawsawi LQ, R. Dayam, L. Taheri, M. Witvrouw, Z. Debyser, N. Neamati, *Bioorg. Med. Chem. Lett.* **2007,** 17(23) 6472.
[24] C. Plasencia, R. Daym, Q. Wang, J. Pinski, T. R. Jr. Burke, D. I. Quinn, and N. Neamati, *Mol. Cancer Ther.* **2005,** 4(7) 1105.
[25] H. Zhao, N. Neamati, S. Sunder, H. Hong, S. wang; G. W. Milne, Y. Pommier,T. R. Jr. Burke, *J. Med. Chem.* **1997**,40(6) 937.
[26] R.W.Young and K.H.Wood,Journal of amarican Chem. Soc.,**1955**,77,400.
[27] B. Kalluraya, R. Chimbalkar and B. S. Holla, Indian J. Heterocyclic Chem.,**1995**,5,37.
[28] K. K. Jha, Y. Kumar and M. Shaharyar, *Asian Jour. of Chemistry*,**2009**,21(9),7403.