



## Synthesis of some Pyrazole Derivatives via Knoevenagel Condensation Proven Effective as Antibacterial and Antifungal Activity

Archana D. Jadhav<sup>1\*</sup>, Pankaj U. Barhate<sup>1</sup> and Ayesha N. Durrani<sup>2</sup>

1. Department of Chemistry, Loknete Ramdas Patil Dhumal Art's, Science and Commerce College Rahuri, Ahmednagar, Maharashtra, **INDIA**

2. Department of Chemistry, Dr. Rafiq Zakaria College of Womens, Naukhanda, Aurangabad, Maharashtra, **INDIA**

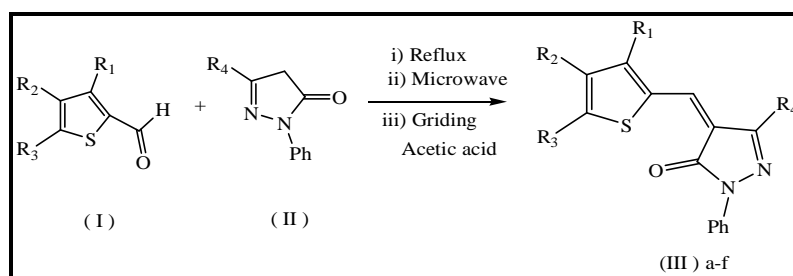
Email: [archu1990jadhav@gmail.com](mailto:archu1990jadhav@gmail.com)

Accepted on 20<sup>th</sup> May, 2020

### ABSTRACT

It deals with the preparation of pyrazole derivative synthesized by Knoevenagel condensation in acidic medium. The work represented by three different methods that is conventional, grinding and microwave assisted reaction. The entire synthesized compound was tested for their antibacterial activity against the Gram-positive and Gram-negative strains of bacteria. The investigation of antibacterial screening data revealed that most of the tested compounds showed less to moderate antibacterial and antifungal activity.

### Graphical Abstract



Synthesis of Pyrazole Derivatives.

**Keywords:** Citric acid, thiophene-2-carbaldehyde, Antifungal, Antibacterial, Spectra, etc.

### INTRODUCTION

Recently pyrazole containing compounds have attracted researchers because of their wide applications in the field of medicines. Pyrazolone is a derivative of pyrazole having an additional ketonic group. It is a heterocyclic nucleus incorporated in many drugs like Phenazone, Ampyrone, Propyphenazone which are potent antipyretics and analgesics [1]. It is an active pharmacophore due to various biological activities associated with it such as antimicrobial [2], antihyperlipidemic [3], anticonvulsant

[4], anti-inflammatory [5], antitumor [6], antiviral [7], antitubercular [8], antioxidant [9] and antidiabetic [10].

According to increasing microbial infections and the phenomenon of microbial resistance to current therapeutic agents due to mutations occurring at genetic level in micro-organisms are the too serious global problems. To overcome these problems there is a need of developing and designing new drug molecules with high potency. Currently much research is going on in this field and heterocyclic compounds are dominating scaffolds owing to their interesting chemistry and bioactivity. There are various synthetic approaches in synthesizing new pyrazole derivatives. The most common synthetic method is conventional method. In recent experiment we were use other two methods that is microwave and simple grinding method for synthesis of pyrazole derivatives with higher yields of product. One of the major concerns is to exhibit simple routes for the synthesis of pyrazole derivatives [11, 12], by Knoevenagel condensation in acidic medium and those synthesized compounds were subjected to antibacterial and antifungal activity to compare their potentials.

## MATERIALS AND METHODS

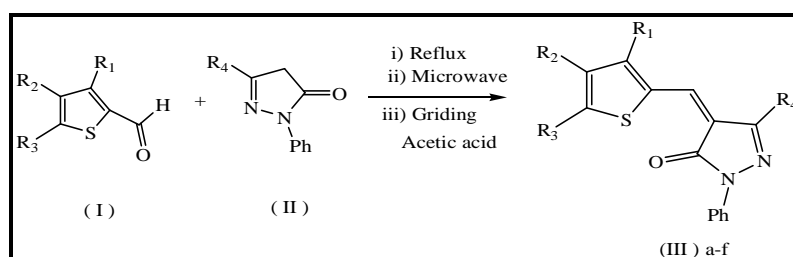
All the chemicals and solvents were obtained from E-Merck, and SD fine chemicals L.T.D. India (AR, LR grade). Melting points were determined in open capillaries in liquid paraffin bath and are uncorrected. Purity of the compound was routinely checked on silica gel TLC plates using  $\text{CHCl}_3$  as solvent.  $^1\text{H}$  NMR spectra were recorded on Bruker AV II, 400 MHz NMR spectrometers in appropriate solvents using TMS as internal standard or the solvent signals as secondary standards and the chemical shift was shown in  $\delta$  scale. Multiplicities of  $^1\text{H}$  NMR signals are designated as s (singlet), d (doublet), dd (doublet of doublet), dt (doublet of triplet), t (triplet), m (multiplet), etc. IR data were recorded an Alpha-T ATR-FTIR.

**Present work:** In present work pyrazole derivatives were synthesized by Knoevenagel condensation in acidic medium, according to conventional as well as non-conventional methods for preparation.

**Conventional method:** Condensation of compounds by the equimolar amounts (0.002 mol) of thiophene-2-carbaldehyde (I) and substituted pyrazolone (II) were taken in glacial acetic acid (4 mL). The reaction mixture was refluxed for 2 to 3 h. The completion of reaction was checked by TLC. After completion of reaction the content were allowed to cool and solid obtained was filtered by Buchner funnel. It was recrystallizing from acetic acid to afford pure compound.

**Microwave method:** Condensation of compound by the equimolar amounts (0.002 mol) of thiophene-2-carbaldehyde (I) and substituted pyrazolone (II) were taken in glacial acetic acid (2 mL). The mixture was subjected to microwave irradiation at 300 W for about 10 min. till completion of reaction checked by colour change and TLC. After completion of reaction, contents were allowed to cool and solid obtained was filtered and purified by recrystallization from acetic acid to afford pure compound.

### Experimental Scheme

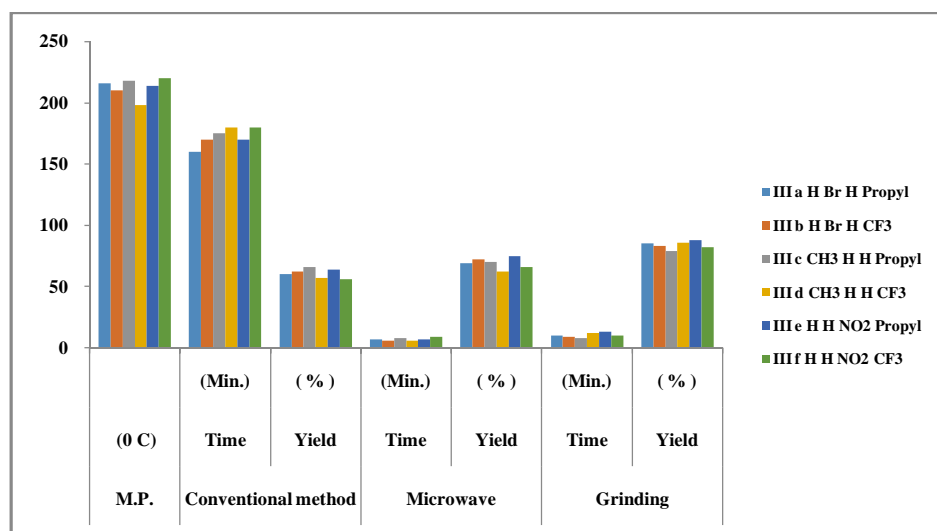


Scheme 1. Synthesis of some Pyrazole Derivatives.

**Grinding method:** Equimolar amounts (0.002 mol) of thiophene-2-carbaldehyde (I), substituted pyrazolone (II) and 2 gm of citric acid/tartaric acid were taken in mortar and the whole reaction mixture was then grind with the help of pestle for about 10 to 15 min. Progress of the reaction was monitored by TLC. After completion of reaction, the reaction mixture was transferred into ice cold water, the solid was obtained filtered, dried and recrystallize from acetic acid to get pure compound.

Table 1. Experimental data

Comp.	R1	R2	R3	R4	M.P. (°C)	Conventional method		Microwave		Grinding	
						Time (Min.)	Yield (%)	Time (Min.)	Yield (%)	Time (Min.)	Yield (%)
III a	H	Br	H	Propyl	216	160	60	7	69	10	85
III b	H	Br	H	CF <sub>3</sub>	210	170	62	6	72	09	83
III c	CH <sub>3</sub>	H	H	Propyl	218	175	66	8	70	08	79
III d	CH <sub>3</sub>	H	H	CF <sub>3</sub>	198	180	57	6	62	12	86
III e	H	H	NO <sub>2</sub>	Propyl	214	170	64	7	75	13	88
III f	H	H	NO <sub>2</sub>	CF <sub>3</sub>	220	180	56	9	66	10	82



Graphical representation of observation.

**Antibacterial activity:** The plates were inoculated by specific microorganism by spread plate technique; bores were made in the solidified agar plate by using a sterile borer. The test solution of specified concentration was added in the bore by using sterile pipette and the plates were kept in freeze for one hour for diffusion and then incubated at 37°C for 24 h. After 24 h the plates were examined and zone of inhibition were recorded. All the synthesized compounds were screened for antibacterial activity against both gram positive *Staphylococcus aureus* and *Staphylococcus epidermidis* and gram negative *E. coli* and *Proteus vulgaris* bacteria at a concentration of 1000 µg mL<sup>-1</sup>. Azithromycin was used as internal standard for comparison of antibacterial activity.

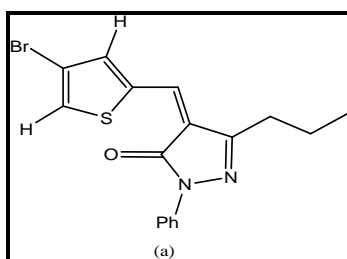
**Antifungal activity:** Antifungal activity study was carried out by cup-plate agar diffusion method using nutrient agar. Fungal culture were made in Sabouraud-Dextrose agar and incubated at 37°C for 18-24 h. The concentration was 1000 µg mL<sup>-1</sup>. The compounds were tested *in-vitro* for their antifungal activity against *Aspergillus niger* and *Candida albicans* at similar concentration as antibacterial. *Fluconazole* was used as internal standard for comparison of antifungal activity.

## RESULTS AND DISCUSSION

All the synthesized compounds are less active or inactive towards used antibacterial and antifungal strains.

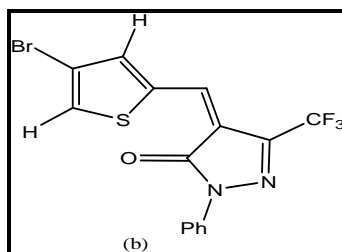
## Spectral Data

**a. (Z)-4-((4-bromothiophen-2-yl)methylene)-1-phenyl-3-propyl-1H-pyrazol-5(4H)-one:** IR: 1668 (C=O stretch.), 1593 (C=N stretch), 1112 (Ar-Br stretch.)  $\text{cm}^{-1}$ ;  $^1\text{H NMR}$  (DMSO- $d_6$ ):  $\delta$  ppm 7.18 (t, 1H, Ar-H,  $J = 7.3$  Hz), 7.42 (t, 2H, Ar-H,  $J = 7.5$  Hz), 7.92- 7.94 (m, 2H, Ar-H), 1.05 (t, 3H,  $\text{CH}_3$  of propyl,  $J = 7.32$  Hz), 1.77 (q, 2H,  $\text{CH}_2$  of propyl,  $J = 7.4$  Hz), 2.69 (t, 2H,  $\text{CH}_2$  of propyl,  $J = 7.4$  Hz), 8.08 (s, 1H), 8.18 (s, 1H, =CH), 8.25 (d, 1H,  $J = 1.4$  Hz); LC/MS  $m/z$  ( $M^{+1}$ ) = 375.



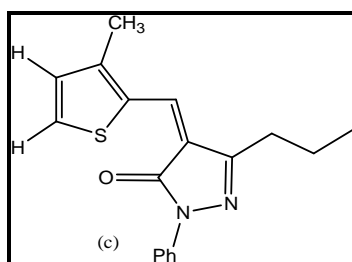
(Z)-4-((4-bromothiophen-2-yl)methylene)-1-phenyl-3-propyl-1H-pyrazol-5(4H)-one

**b. (Z)-4-((4-bromothiophen-2-yl)methylene)-1-phenyl-3-(trifluoromethyl)-1H-pyrazol-5(4H)-one:** IR: 1668 (C=O), 1591 (C=N), 1114 (Ar-Br)  $\text{cm}^{-1}$ ;  $^1\text{H NMR}$  (DMSO- $d_6$ )  $\delta$ : 7.21 (t, 1H, Ar-H,  $J = 7.0$  Hz), 7.49 (t, 2H, Ar-H,  $J = 7.4$  Hz), 7.86 (d, 2H, Ar-H,  $J = 8.0$  Hz), 8.29 (s, 1H, =CH), 8.39 (s, 1H, Ar-H), 8.46 (s, 1H, Ar-H); MS:  $m/z$  ( $M^{+1}$ ) = 401.



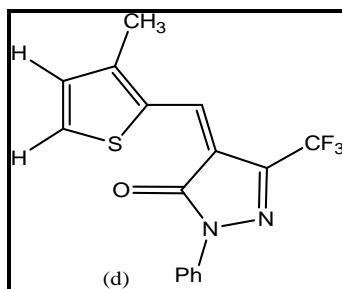
(Z)-4-((4-bromothiophen-2-yl)methylene)-1-phenyl-3-(trifluoromethyl)-1H-pyrazol-5(4H)-one

**c. IR:** 1665 (C=O), 1592 (C=N)  $\text{cm}^{-1}$ ;  $^1\text{H NMR}$  (DMSO- $d_6$ ):  $\delta$  1.08 (t, 3H,  $\text{CH}_3$ ), 1.79 (m, 2H,  $\text{CH}_2$ ), 2.35 (s, 3H,  $\text{CH}_3$ ), 2.65 (t, 2H,  $\text{CH}_2$ ), 7.20 (t, 1H, Ar-H,  $J = 7.1$ ), 7.40 (t, 2H, Ar-H,  $J = 7.5$  Hz), 7.96 (d, 2H, Ar-H,  $J = 8.2$  Hz), 8.06 (s, 1H, =CH), 8.19 (d, 1H, Ar-H,  $J = 4.2$  Hz), 8.27 (d, 1H, Ar-H,  $J = 4.2$  Hz); MS:  $m/z$  ( $M^{+1}$ ) = 311



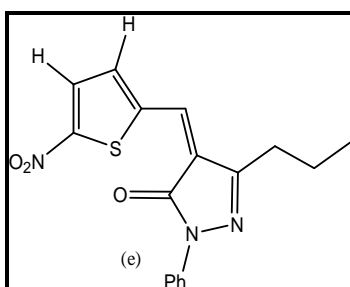
(Z)-4-((3-methylthiophen-2-yl)methylene)-1-phenyl-3-propyl-1H-pyrazol-5(4H)-one

**d). (Z)-4-((3-methylthiophen-2-yl)methylene)-1-phenyl-3-(trifluoromethyl)-1H-pyrazol-5(4H)-one:** IR: 1667 (C=O), 1596 (C=N)  $\text{cm}^{-1}$ ;  $^1\text{H NMR}$  (DMSO- $\text{d}_6$ ):  $\delta$  2.40 (s, 3H,  $\text{CH}_3$ ), 7.32 (t, 1H, Ar-H,  $J=7.2$  Hz), 7.51 (t, 2h, Ar-H,  $J=7.5$  Hz), 7.98 (d, 2H, Ar-H,  $J=8.1$  Hz), 8.30 (s, 1H, =CH), 8.26 (d, 1H, Ar-H,  $J=4.1$  Hz), 8.39 (d, 1H, Ar-H,  $J=4.1$  Hz); MS:  $m/z$  ( $\text{M}^{+1}$ ) = 337.



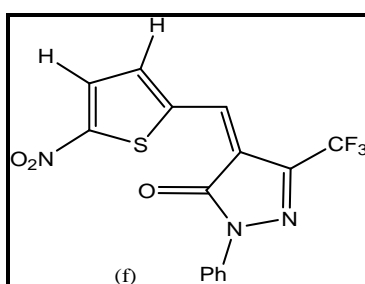
(Z)-4-((3-methylthiophen-2-yl)methylene)-1-phenyl-3-(trifluoromethyl)-1H-pyrazol-5(4H)-one

**e). (Z)-4-((3-nitrothiophen-2-yl)methylene)-1-phenyl-3-propyl-1H-pyrazol-5(4H)-one:** IR: 1669 (C=O), 1609 (C=C), 1598 (C=N), 1540 ( $\text{NO}_2$ ),  $\text{cm}^{-1}$ ;  $^1\text{H NMR}$  (DMSO- $\text{d}_6$ ):  $\delta$  1.09 (t, 3H,  $\text{CH}_3$ ), 1.75 (m, 2H,  $\text{CH}_2$ ), 2.62 (t, 2H,  $\text{CH}_2$ ), 7.22 (t, 1H, Ar-H,  $J=7.0$  Hz), 7.44 (t, 2H, Ar-H,  $J=7.4$  Hz), 7.96 (d, 2H, Ar-H,  $J=8.2$  Hz), 8.10 (s, 1H, =CH), 8.38 (d, 1H, Ar-H,  $J=4.0$  Hz), 8.49 (d, 1H, Ar-H,  $J=4.0$  Hz); MS:  $m/z$  ( $\text{M}^{+1}$ ) = 342.



(Z)-4-((3-nitrothiophen-2-yl)methylene)-1-phenyl-3-propyl-1H-pyrazol-5(4H)-one

**f). (Z)-4-((5-nitrothiophen-2-yl)methylene)-1-phenyl-3-(trifluoromethyl)-1H-pyrazol-5(4H)-one:** IR: 1665 (C=O), 1611 (C=C), 1594 (C=N), 1541 ( $\text{NO}_2$ ),  $\text{cm}^{-1}$ ;  $^1\text{H NMR}$  (DMSO- $\text{d}_6$ ):  $\delta$  7.35 (t, 1H, Ar-H,  $J=7.3$  Hz), 7.54 (t, 2H, Ar-H,  $J=7.7$  Hz), 7.92 (d, 2H, Ar-H,  $J=8.4$  Hz), 8.26 (s, 1H, =CH), 8.34 (d, 1H, Ar-H,  $J=4.6$  Hz), 8.45 (d, 1H, Ar-H,  $J=4.6$  Hz); MS:  $m/z$  ( $\text{M}^{+1}$ ) = 368.

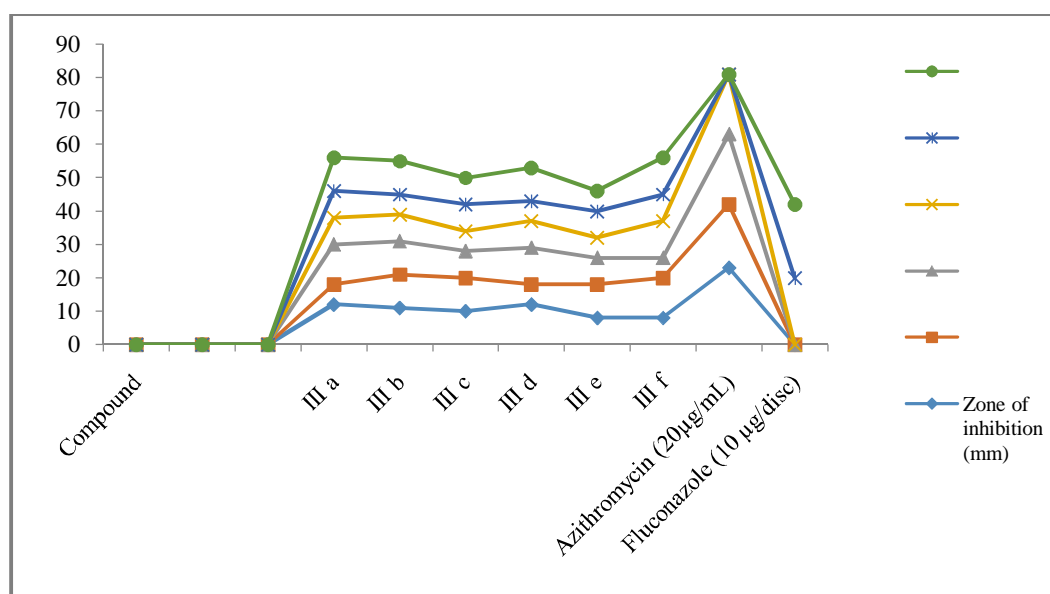


(Z)-4-((5-nitrothiophen-2-yl)methylene)-1-phenyl-3-(trifluoromethyl)-1H-pyrazol-5(4H)-one

**Antibacterial and Antifungal activity:** The results of zone of inhibition of synthesized compounds are reported in table 2. It was observed that less to moderate antibacterial and antifungal activity compared to the control drug against all the tested species.

Table 2. Zone of inhibition for minimal bactericidal and fungicidal concentration

Compound	Zone of inhibition (mm)					
	Antibacterial test model				Antifungal test model	
	Gram positive		Gram negative			
	<i>S. aureus</i>	<i>S. epidermidis</i>	<i>E. coli</i>	<i>P. vulgaris</i>	<i>A. niger</i>	<i>C. albicans</i>
III a	12	06	12	08	08	10
III b	11	10	10	08	06	10
III c	10	10	08	06	08	08
III d	12	06	11	08	06	10
III e	08	10	08	06	08	06
III f	08	12	06	11	08	11
Azithromycin (20 µg mL <sup>-1</sup> )	23	19	21	18	-	-
Fluconazole (10 µg disc <sup>-1</sup> )	-	-	-	-	20	22



Graphical representation for bactericidal and fungicidal concentration.

## APPLICATION

The application of our reaction is to increase the product yield with minimum time span and in future it will apply for other bioactivities like anticancer, antiviral, etc. as relevant biological activity.

## CONCLUSION

The main target of our reaction is to reduce the reaction time and efficiency of the product. Here, we have presented an operationally simple, suitable, fast, efficient method for the preparation of Pyrazole derivative. The main focus of this research work was to synthesize, purify, characterize and evaluate antibacterial and antifungal activities of the synthesized compounds & which shows less to moderate antibacterial & antifungal activity.

## ACKNOWLEDGEMENTS

The authors wish to express a deep sense of gratitude to the respected principal of Dr. Maqdoom Farooqui of Dr. Rafiq Zakaria College for Womens, Aurangabad for providing the necessary lab facilities and constant encouragement and perceptive criticism while completing the research work. We thank Prin. Dr. S. A. Pathare of Loknete Ramdas Patil Dhumal Arts, Science and Commerce

College, Rahuri and all teaching and non-teaching staff Department of Chemistry, for their kind cooperation. We also thankful to Prof. Honde Bharat of Shri Vivekanand Nursing Home Trust's College of B. Pharmacy, Shrishivaji nagar.

## REFERENCES

- [1]. M. Himly, B. Jahn-Schmid, K. Pittertschatscher, B. Bohle, K. Grubmayr, F. Ferreira, H. Ebner C. Ebner, *J Allergy Clin Immunol*, **2003**, 111, 882.
- [2]. A. M. Akondi, M. L. Kantam, R. Trivedi, J. Bharatann, S. P. B. Vemulapalli, S. K. Bhargava, S. K. Buddana, R. S. Prakasham, *J Mole Catal A: Chem*, **2016**, 411, 325.
- [3]. M. Vijay Kumar, B. C. Revanasiddappa, *Indian J Heterocycl Chem*, **2015**, 25, 169.
- [4]. Z. Chilmonczyk, D. Siluk, R. Kaliszan, B. Lozowicka, J. Poplawski, S. Filipek, *Pure Appl Chem*, **2001**, 73, 1445.
- [5]. P. Khloya, P. Kumar, A. Mittal, N. K. Aggarwal, P. K. Sharma, *Org Med Chem Lett*, **2013**, <http://www.orgniedchemlett.coni/content/3/1/9>.
- [6]. K. C. Nicolaou, B. A. Pratt, S. Arseniyadis, M. Waetmann, A. Obrate, P. Giannakakou, *Chem Med Chem*, **2006**, 1, 41.
- [7]. S. A. P. Rostom, *Bioorg Med Chem*, **2010**, 18, 2767.
- [8]. S. R. Pattan, P. V. Patel, G. S. Athare, A. B. Jagnar, S. A. Nirmal, J. S. Pattan, *Bulgarian Chem Commun*, **2014**, 46(1), 125.
- [9]. M. Abdel-Aziz, G. El-Din, A. Abuo-Rahma, A. Hassan A, *Eur J Med Chem*, **2009**, 44, 3480.
- [10]. S. R. Shih, T. Y. Chu, C. R. Reddy, S. N. Tseng, H. L. Chen, W. F. Tang, M. S. Wu, J. Y. Yeh, Y. S. Chao, J. T. Hsu, H. P. Hsieh, J. T. Homg, *J Biomed Sci*, **2010**, <http://www.jbiomedsci.com/content/17/1/13>.
- [11]. A. Padmaja, T. Payani, D. G. Reddy, V. Padmavathi, *Eur j Med Chem*, **2009**, 44, 4557.
- [12]. J. Isaad, A. Perwuelz, *Tetrahedron Lett.*, **2010**, 51, 5328-5332.
- [13]. H. Balseven, M. M. Isgor, S. Mert, Z. Alim, S. Beydemir, S. Ok, Kasimogullari, R. Bioorg. *Med. Chem.*, **2013**, 21, 21-27.
- [14]. Z. Chilmonczyk, D. Siluk, R. Kaliszan, B. Lozowicka, J. Poplawski, S. Filipek, *Pure Appl Chem*, **2001**, 73, 1445.
- [15]. M. Abdel-Aziz, G. El-Din, A. Abuo-Rahma, A. A. Hassan, *Eur J Med Chem*, **2009**, 44, 3480.
- [16]. S. Keithellakpam, N. Moirangthem, W. S. Laitonjam, *Indian J Chem*, **2015**, 54B, 1157.
- [17]. R. V. Hangarge, D. V. Jarikote, M. S. Shingare, *Green Chem.*, **2002**, 4, 266.
- [18]. G. Mariappan, B. P. Saha, L. Sutharson, A. Singh, S. Garg, L. Pandey, D. Kumar, *Saudi Pharm J*, **2011**, 19, 115.
- [19]. Jag Mohan, *Indian J Chem*, **2005**, 44 B, 628.
- [20]. E. Palaska, G. Sah in, P. Kelicen, N. T. Durulu, G. Altmok, *Farmaco*, **2002**, 57, 101.
- [21]. Rathodls, M. T. Chhabria, J. D. Patel, *Indian J Heterocycl Chem*, **2005**, 14, 281.
- [22]. P. Erics, *Co-Ord Chem Rev*, **1996**, 153, 199.
- [23]. N. Gurn, *J Sci and Indust Res*, **2001**, 60, 601.
- [24]. V. Daniela, F. Marco, G. Enzo, *Bio-Org and Med. Chem. Lett.*, **2003**, 13, 1005.
- [25]. S. K. Jain, P. Mishra, *Indian J Heterocycl Chem*, **2005**, 14, 205.
- [26]. H. Girl, J. Singh, *Indian Chem Soc*, **1972**, 49, 175.
- [27]. V. K. Pandey, H. C. Lohani, A. K. Agrawal, *Indian J. Pharm. Sci.*, **1982**, 44, 155.
- [28]. Z. Muhi-eldeen, F. Al-Jawed, S. Eldin, Abdui-Kadirs, M. Carabet, *Eur J Med Chem*, **1982**, 17, 479.
- [29]. M. V. Sonawane, S. B. Chaudhari, J. P. Sonawane, S. S. Patil, S. K. Mali, *Int J Recent Sci Res*, **2015**, 6, 2737.
- [30]. A. Gadakh, C. Pandit, S. S. Rindhe, B. K. Karale, *Int J Chem*, **2010**, 2, 206.