WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 5.990
Volume 4, Issue 12, 1944-1953. Research Article ISSN 2277– 7105

SYNTHESIS AND ANTIMICROBIAL EVALUATION OF SOME NOVEL SUSTITUTED [1,2,4]TRIAZOLO[5,1-b][1,3,4]THIADIAZOLE DERIVATIVES

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Article Received on
18 Oct 2015,

Revised on 10 Nov 2015,
Accepted on 02 Dec 2015

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ABSTRACT

The search for potent bioactive agents in pharmaceutical industry has been directed towards the nitrogen containing heterocycles. In recent days the researchers think about synthesis of bridged derivatives of nitrogen containing heterocycles. The present study reports the synthesis of a library new conjugated heterocycles including [1,2,4]triazolo[5,1-b][1,3,4]thiadiazoles by cyclo condensation reaction of 1-amino-3-aryl-1H-1, 2, 4-triazole-5-thiol with various aromatic carboxylic acids and phosphorous oxychloride. The structures of newly synthesized compounds were established using IR, 1H-NMR, Mass and elemental analysis. The newly synthesized compounds were screened for antimicrobial activity using cup-plate agar diffusion method some of the compounds shows promising antibacterial and antifungal activity.

KEYWORDS: Antibacterial, anti fungal activity, [1,2,4]triazolo[5,1-b][1,3,4]thiadiazole.

INTRODUCTION

The searching of a new agent for the treatment of microbial infections is an important tool of medicinal chemistry. Microbes shows resistance to the most of marketed drugs hence to

overcome it there is continual need of newer antimicrobial agents. Microbial infections and diseases associated with it is a major health problem in the world from decades. A lot of chemical classes of heterocyclic and fused heterocyclic compounds have been identified through molecular biology, empirical screening and rational drug development for evaluation of antimicrobial agents during the past decades. In terms of searching, it could be considered that the conjugated triazolo thiadiazole derivates heterocyclic compounds are of major importance in their biological as well as synthetic approach of medicinal chemistry. From literature review it is suggested that triazolo thiadiazoles shows wide varieties of biological activities such as anti cancer[1], anti-inflammatory[2,3], antimicrobial[4,5], analgesic, anti-oxidant[6], Antitubercular activity.[7] In the view of above fact it is thought to synthesize and evaluate substituted[1,2,4]triazolo[5,1-b][1,3,4]thiadiazole derivatives for antimicrobial activity.

MATERIALS AND METHODS

Material

All the chemicals required for the synthesis were purchased from Modern Science, Nashik and are of AR grade.

Methods

ANTIBACTERIAL ACTIVITY

Anti bacterial study was carried out by using Cup-plate Agar diffusion method. The synthesized derivatives were tested in vitro for their anti bacterial activity against *E.coli* (NCTC 10418), *S. Aureus* (NCTC 6571) and *B. subtilis* which are pathogenic to human beings. Leavofloxacin had been used as a standard drug.

ANTI FUNGAL ACTIVITY

Anti-fungal activity was carried out by using Cup-Plate Agar diffusion method using nutrient agar as a culture media. The synthesized compounds were tested against *Candida albicans* (ATCC 10231) and *Aspergilus niger* (ATCC 16404). Amphotericin B had been used as a standard drug.

The synthesized compounds were dissolved in DMF and activities were carried out at a concentration of 200 µg/ml.

EXPERIMENTAL

Melting points were determined in open capillary method and are uncorrected. Purity of the compound was checked on Silica gel TLC plates. IR spectra were recorded on Thermo Nicolate IR 200 spectrophotometer using KBr disc method. ^{1}H NMR spectra were recorded on Bruker AMX-400, DMSO δ6 as solvent and TMS as internal standard. Combustion analyses were found to be within the limits of permissible errors.

Synthesis of 1-amino-3-aryl-1H-1, 2, 4-triazole-5-thiol[8]

0.01 mole of an acid hydrazide was stirred with a solution of 1.5 % w/v alcoholic solution of potassium hydroxide after which to it 0.01 mole of carbon disulfide was added slowly and the reaction mixture was left overnight at room temperature. The solid product of potassium dithiocarbazinate was filtered, washed with cold methanol and dried. It was directly used for next step without purification. Potassium dithiocarbazinate was then taken in water (8 mL) and hydrazine hydrate (0.02 moles) and refluxed for 4-5 hrs. During progress of the reaction, the reaction mixture turned to green with evolution of hydrogen sulphide and finally it became homogeneous. It was then diluted with little cold water and acidified with conc. hydrochloric acid. The white precipitated solid was filtered, washed with cold water and recrystallized from aqueous methanol.

Synthesis of 1, 2, 4-triazolo [3, 4-b] [1, 3, 4] thiadiazoles (C_1-C_{16})[8,9]

0.01 mole of 1-amino-3-aryl-1H-1, 2, 4-triazole-5-thiol is mixed with a mixture of 0.01 mole of an aromatic acids and 5 ml of phosphorous oxychloride and was refluxed for 5-6 hrs. The reaction mixture was then cooled to room temperature and then poured on to the ice with continuous stirring. The reaction mixture thus obtained is then neutralized with sodium bicarbonate. Solid thus obtained is then washed with water and recrystallized from ethanol to offer final compounds.

SPECTRAL DATA

C_1: IR (cm-1) KBr disc: 3415.28 –NH str.; 3022.58 Ar-CH str.; 2865.36 –CH_3 str.; 1575.24 -C=N str.; 1010.36 –C-Nstr.; 690.25 –C-S-C str. **^{1}H-NMR (ppm):** 10.4 2H of –NH, 6.8-7.6 14 H of phenyl, 1.2-2.6 12H of –CH_3**; m/e (100%):** 518.

C_2: IR (cm-1) KBr disc: 3412.54 –NH str.; 3024.38 Ar-CH str.; 2865.32 –CH_3 str.; 1574.23 -C=N str.; 1008.28 –C-N str.; 691.38 –C-S-C str. 1091.23 –C-Cl str. **^{1}H-NMR (ppm):** 10.28 2H of –NH, 6.6-7.8 14 H of phenyl, 1.2-1.6 6H of –CH_3, 0.8-1.2 2H of –CH_2; **m/e (100%):** 532.

C_3: IR (cm-1) KBr disc: 3420.35 –NH str.;3240.28 –OH str., 3034.48 Ar-CH str.; 2820.28 –CH_3 str.; 1548.27 -C=N str.; 1007.24 –C-N str.; 694.18 –C-S-C str. **^{1}H-NMR (ppm):** 10.24 1H of –NH, 6.8-7.6 11 H of phenyl, 4.0 1H of –OH, 1.0-1.6 6H of –CH_3 ; **m/e (100%):** 415.

C_4: IR (cm-1) KBr disc: 3425.68 –NH str.; 3320.28 –CH=CH str., 3028.34 Ar-CH str.; 2835.27 –CH_3 str.; 1549.24 -C=N str.; 1012.32 –C-N str.; 695.37 –C-S-C str. **^{1}H-NMR (ppm):** 10.40 1H of –NH, 6.8-7.8 12 H of phenyl, 5.2-5.6 2H of –CH=CH, 1.0-1.4 6H of –CH_3 ; **m/e (100%):** 425.

C_5: IR (cm-1) KBr disc: 3426.58 –NH str.; 3025.69 Ar-CH str.; 2898.27–CH_3 str.; 1569.47 -C=N str.; 1007.32 –C-N str.; 1091.25 –C-Cl str., 690.18 –C-S-C str. **^{1}H-NMR (ppm):** 10.50 2H of –NH, 6.6-7.8 14 H of phenyl, 1.2-1.86H of –CH_3, 0.8-1.0 2H of CH_2; **m/e (100%):** 532.

C_6: IR (cm-1) KBr disc: 3412.24 –NH str.; 3015.49 Ar-CH str.; 2852.17–CH_3 str.; 1568.24 -C=N str.; 1008.27 –C-N str.; 1078.24–C-Cl str., 694.28 –C-S-C str. **^{1}H-NMR (ppm):** 10.25 2H of –NH, 6.8-7.6 14 H of phenyl, 0.8-1.2 4H of CH_2; **m/e (100%):** 546**.**

C_7: IR (cm-1) KBr disc: 3408.29 –NH str.; 3280.27 –OH str., 3025.08 Ar-CH str.; 2864.89 –CH_3 str.; 1598.47 -C=N str.; 1009.27 –C-N str.; 1085.27 –C-Cl str., 701.38 –C-S-C str. **^{1}H-NMR (ppm):** 10.25 1H of –NH, 6.8-7.6 11 H of phenyl, 4.0 1H of –OH, 0.8-1.2 2H of CH_2; **m/e (100%):** 429.

C_8: IR (cm-1) KBr disc: 3410.47 –NH str.; 3315.26 –CH=CH str., 3045.16 Ar-CH str.; 2864.23 –CH_3 str.; 1598.24 -C=N str.; 1009.27 –C-N str.; 1085.27 –C-Cl str., 710.35 –C-S-C str. **^{1}H-NMR (ppm):** 10.30 1H of –NH, 6.8-7.8 12 H of phenyl,5.6-5.8 2H of –CH=CH, 0.6.10 2H of CH_2; **m/e (100%):** 439.

C_9: IR (cm-1) KBr disc: 3456.27 –NH str.; 3228.21 –OH str., 3058.19 Ar-CH str.; 2856.37 –CH_3 str.; 1598.24 -C=N str.; 1009.27 –C-N str.; 695.28 –C-S-C str. **^{1}H-NMR (ppm):** 10.10 1H of –NH, 6.8-7.8 11 H of phenyl,4.0 1H of –OH, 1.2-1.8 6H of CH_3; **m/e (100%):** 415.

C_{10}: IR (cm-1) KBr disc: 3440.29 –NH str.; 3230.50 –OH str., 3028.14 Ar-CH str.; 2854.78 –CH_3 str.; 1610.27 -C=N str.; 1007.27 –C-N str.; 1094.27 –C-Cl str., 696.28 –C-S-C str.

^{1}H-NMR (ppm): 10.20 1H of –NH, 6.6-7.4 11 H of phenyl,4.0 1H of –OH, 0.8-1.2 2H of CH_2; **m/e (100%):** 429.

C_{11}: IR (cm-1) KBr disc: 3426.29 –NH str.; 3286.25 –OH str., 3078.14 Ar-CH str.; 1610.27 -C=N str.; 1007.27 –C-N str.; 694.48 –C-S-C str. **^{1}H-NMR (ppm):** 6.6-7.4 8H of phenyl,4.0 2H of –OH; **m/e (100%):** 312**.**

C_{12}: IR (cm-1) KBr disc: 3432.21 –NH str.; 3320.28 –CH=CH str., 3240.18 –OH str., 3028.19 Ar-CH str.; 1576.28 -C=N str.; 1054.29 –C-N str.; 697.58 –C-S-C str. **^{1}H-NMR (ppm):** 6.8-7.8 9H of phenyl, 5.8-6.0 2H of –CH=CH, 4.0 1H of –OH; **m/e (100%):** 322**.**

C_{13}: IR (cm-1) KBr disc: 3465.21 –NH str.; 3315.15 –CH=CH str., 3028.19 Ar-CH str.; 1578.24 -C=N str.; 1056.27 –C-N str.; 698.27 –C-S-C str. **^{1}H-NMR (ppm):** 10.40 1H of –NH, 6.4-7.6 12H of phenyl, 5.8-6.0 2H of –CH=CH,1.2-1.8 6H of $–CH_3$; **m/e (100%):** 425.

C_{14}: IR (cm-1) KBr disc: 3428.17 –NH str.; 3325.18 –CH=CH str., 3029.17 Ar-CH str.; 1585.14 -C=N str.; 1068.48 –C-N str.; 1094.28 –C-Cl str., 692.18 –C-S-C str. **^{1}H-NMR (ppm):** 10.35 1H of –NH, 6.4-7.6 12H of phenyl, 5.8-6.0 2H of –CH=CH, 0.8-1.4 2H of $–CH_2$; **m/e (100%):** 439.

C_{15}: IR (cm-1) KBr disc: 3418.29 –NH str.; 3305.19 –CH=CH str., 3240.18 –OH str., 3019.85 Ar-CH str.; 1578.14 -C=N str.; 1047.15 –C-N str.; 692.18 –C-S-C str. **^{1}H-NMR (ppm):** 6.6-7.6 9H of phenyl, 5.6-6.0 2H of –CH=CH,4.0 1H of –OH; **m/e (100%):** 322.

C_{16}: IR (cm-1) KBr disc: 3408.12 –NH str.; 3309.26 –CH=CH str., 3010.48 Ar-CH str.; 1587.14 -C=N str.; 1058.15 –C-N str.; 696.21 –C-S-C str. **^{1}H-NMR (ppm):** 6.6-7.8 10H of phenyl, 5.8-6.2 4H of –CH=CH; **m/e (100%):** 332.

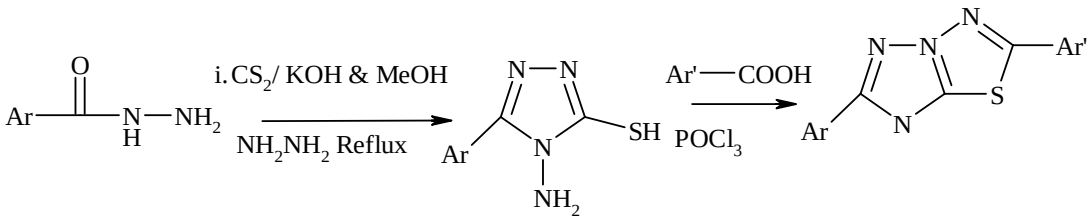
RESULT AND DISCUSSSION

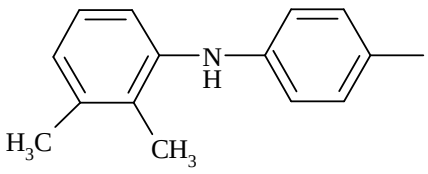
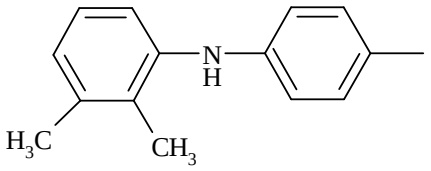
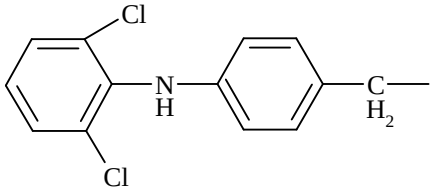
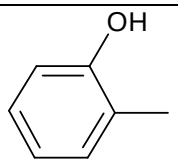
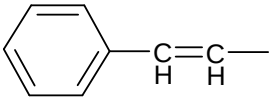
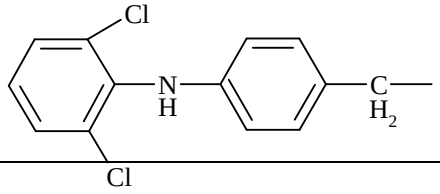
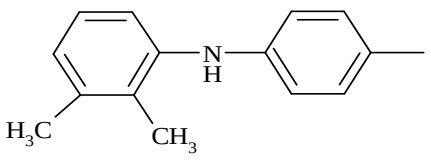
The desired compounds were synthesized as outlined in scheme mention below. The aromatic carboxylic acid were esterified in presence of an alcohol and conc. Sulphuric acid which on further treatment with hydrazine hydrate gives acid hydrazides. This acid hydrazide on further treatment with carbon disulphide and alcoholic potassium hydroxide give Potassium dithiocarbazinate which on further reaction with hydrazine hydrate offers 1-amino-3-aryl-1H-1, 2, 4-triazole-5-thiol. Which on refluxing with carboxylic acid and phosphorous oxychloride gives conjugated 1, 2, 4-triazolo [3, 4-b] [1, 3, 4] thiadiazoles. The cyclo

condensation was confirmed by appearance of new stretching bands and disappearance of characteristic peaks for thiol and amino indicates the formation of conjugated scaffolds. Also their proton NMR spectra lack in characteristics peaks for SH and NH protons respectively indicates the formation of conjugated system.

The synthesized compounds were subjected for anti bacterial activity. Out of sixteen compounds the compounds C_1,C_2,C_3,C_5,C_{11} and C_{14} had shown significant antibacterial activity. The structural features of the compounds like presence of sulphur along with nitrogen and some substituents like $–CH_3$, -OH and –Cl was thought to increase the biological activity. In case of anti fungal activity the compounds C_5, C_6, C_9 and C_{15} shows significant antifungal activity.

Scheme



Comp. Code	Ar	Ar’
C_1		
C_2		
C_3		
C_4		
C_5		

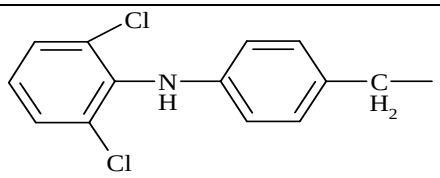
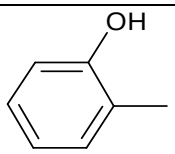
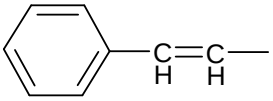
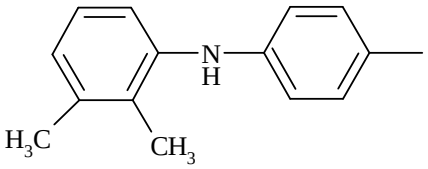
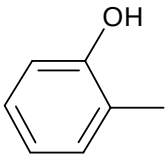
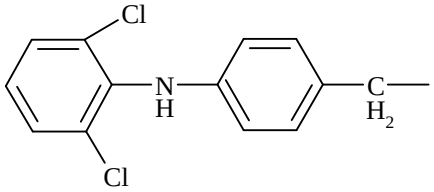
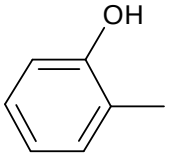
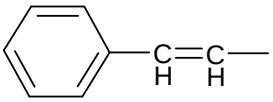
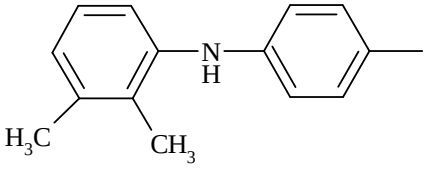
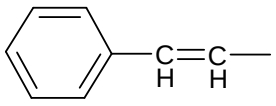
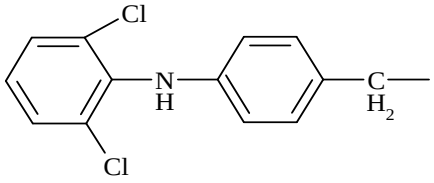
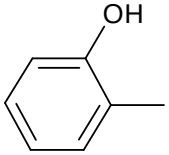
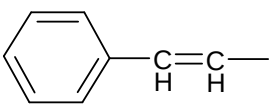
C_6		
C_7		
C_8		
C_9		
C_{10}		
C_{11}		
C_{12}		
C_{13}		
C_{14}		
C_{15}		
C_{16}		

Table no. 01: Analytical data of synthesized compounds (C_1-C_{16})

Comp. Code	Molecular Formula	Mole wt.	M.P. (0C)	Elemental analysis Found (cald.)			Rf Value	% Yield
				C	H	N		
C_1	$C_{31}H_{30}N_6S$	518.69	245-252	71.79 (71.42)	5.83 (5.38)	16.20 (15.92)	0.56	64
C_2	$C_{32}H_{32}N_6S$	532.72	236-245	72.15 (71.95)	6.05 (5.85)	15.78 (15.48)	0.61	59
C_3	$C_{23}H_{21}N_5OS$	415.52	278-281	66.48 (66.15)	5.09 (4.89)	16.85 (16.48)	0.63	62
C_4	$C_{25}H_{23}N_5S$	425.56	289-294	70.56 (7026)	5.45 (5.15)	16.46 (16.08)	0.64	69
C_5	$C_{32}H_{32}N_6S$	532.72	302-306	72.15 (71.96)	6.05 (5.85)	15.78 (15.43)	0.59	71
C_6	$C_{33}H_{34}N_6S$	546.74	288-293	72.50 (71.08)	6.27 (5.94)	15.37 (15.01)	0.64	48
C_7	$C_{24}H_{23}N_5OS$	429.55	278-284	67.11 (66.95)	5.40 (5.03)	16.30 (15.98)	0.68	53
C_8	$C_{26}H_{25}N_5S$	439.59	282-286	71.04 (69.54)	5.73 (5.38)	15.93 (15.68)	0.63	56
C_9	$C_{23}H_{21}N_5OS$	415.52	286-291	66.48 (66.05)	5.09 (4.89)	16.85 (16.49)	0.71	59
C_{10}	$C_{24}H_{23}N_5OS$	429.55	289-294	67.11 (66.98)	5.40 (5.06)	16.30 (15.98)	0.67	65
C_{11}	$C_{15}H_{12}N_4O_2S$	312.35	279-284	57.68 (57.38)	3.87 (3.54)	17.94 (17.58)	0.70	61
C_{12}	$C_{17}H_{14}N_4OS$	322.39	256-261	63.34 (62.98)	4.38 (4.07)	17.38 (17.05)	0.59	58
C_{13}	$C_{25}H_{23}N_5S$	425.56	263-268	70.56 (70.34)	5.45 (5.09)	16.46 (16.18)	0.62	63
C_{14}	$C_{26}H_{25}N_5S$	439.59	245-252	71.04 (70.95)	5.73 (5.36)	15.93 (15.68)	0.73	64
C_{15}	$C_{17}H_{14}N_4OS$	322.39	286-291	63.34 (62.98)	4.38 (4.12)	17.38 (17.05)	0.69	67
C_{16}	$C_{19}H_{16}N_4S$	332.43	278-285	68.65 (68.29)	4.85 (4.59)	16.85 (16.48)	0.68	70

Table no. 02: Antibacterial and antifungal activity of synthesized compounds (C_1-C_{16})

Compd. code	Zone of inhibition at 200 g/ml (in mm)				
	E.coli	***B.Subtilis***	***S.aureus***	***A. niger***	***C.albicans***
C_1	25	24	26	20	21
C_2	25	26	26	21	20
C_3	24	25	26	15	22
C_4	20	23	25	16	21
C_5	26	25	21	21	22
C_6	23	24	23	22	24
C_7	25	23	22	18	23
C_8	22	24	20	19	22
C_9	24	25	26	21	23
C_{10}	23	25	26	20	22
C_{11}	26	23	26	20	21
C_{12}	24	23	25	19	21
C_{13}	20	24	25	19	22
C_{14}	25	26	23	20	21
C_{15}	24	23	26	21	22
C_{16}	20	22	24	18	23
Levofloxacin	26	25	26	-	-
Amphotericin-B	-	-	-	22	23

CONCLUSION

The concept laid down in this research describe the access of The present work describes the first access to the substituted [1,2,4]triazolo[5,1-b][1,3,4]thiadiazole ring system with potential antimicrobial activities. These synthesized compounds may serve as a starting material for further synthetic revolution in the field of synthetic chemistry. Some of the derivatives with minor molecular modifications can be explore as a prominent anti-microbial agents in future.

REFERENCES

1. Asif Husain, Mohd Rashid , Ravinesh Mishra , Shama Parveen , Dong-Soo Shin, Deepak Kumar, Benzimidazole bearing oxadiazole and triazolo-thiadiazoles nucleus: Design and synthesis as anticancer agents, Bioorganic & Medicinal Chemistry Letters., 2012; 22: 5438–5444.
2. Mohd. Wasim Akhter, Mohd. Zaheen Hassan, Mohd. Amir, Synthesis and pharmacological evaluation of 3-diphenylmethyl-6-substituted-1,2, 4-triazolo[3,4-b]-1,3,4-thiadiazoles: A condensed bridgehead nitrogen heterocyclic system, Arabian Journal of Chemistry., 2014; 7: 955–963.

3. V. Mathewa, J. Keshavayya, V.P. Vaidya, Heterocyclic system containing bridgehead nitrogen atom: synthesis and pharmacological activities of some substituted 1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles, European Journal of Medicinal Chemistry., 2006; 41: 1048–1058.
4. Yin Luo , Shuai Zhang , Zhi-Jun Liu , Wu Chen , Jie Fu , Qing-Fu Zeng , Hai-Liang Zhu, Synthesis and antimicrobical evaluation of a novel class of 1,3,4-thiadiazole: Derivatives bearing 1,2,4-triazolo[1,5-a] pyrimidine moiety, European Journal of Medicinal Chemistry., 2013; 64: 54-61.
5. Nasser S.A.M. Khalil, N- and S-a-L- Arabinopyranosyl[1,2,4]triazolo[3,4-b][1,3,4]thiadiazoles. First synthesis and biological evaluation, European Journal of Medicinal Chemistry., 2007; 42: 1193-1199.
6. N. Chidananda , Boja Poojary , V. Sumangala , N. Suchetha Kumari , Prashanth Shetty , T. Arulmoli, Facile synthesis, characterization and pharmacological activities of 3,6-disubstituted 1,2,4-triazolo[3,4-b][1,3,4]thiadiazoles and 5,6-dihydro-3,6-disubstituted-1,2,4-triazolo[3,4-b][1,3,4]thiadiazoles, European Journal of Medicinal Chemistry., 2012; 51: 124-136.
7. M.S. Karthikeyan, B.S. Holla, B. Kalluraya, N.S. Kumar, Monatsh. Chem., 2007; 138: 1309-1316.
8. Imtiaz Khan , Sumera Zaib , Aliya Ibrar , Nasim Hasan Rama , Jim Simpson, Jamshed Iqbal, Synthesis, crystal structure and biological evaluation of some novel 1,2,4-triazolo[3,4-b]-1,3,4-thiadiazoles and 1,2,4-triazolo[3,4-b]-1,3,4- Thiadiazines, European Journal of Medicinal Chemistry., 2014; 78: 167-177.
9. Imtiaz Khan , Aliya Ibrar , Naeem Abbas, Triazolothiadiazoles and triazolothiadiazines Biologically attractive Scaffolds, European Journal of Medicinal Chemistry., 2013; 63: 854-868.