International Journal of ChemTech Research
CODEN(USA): IJCRGG ISSN : 0974-4290
Vol. 3, No.2, pp 761-765, April-June 2011

Efficient Synthesis and Biological evaluation of 3-(phenyl)-6-(4-amino phenyl)[1,2,4]triazolo[3,4-b][1,3,4] thiadiazole and their Schiff base derivatives

Kokila Parmar*, Sarju Prajapati, Rinku Patel, Sejal Joshi, and Rekha Patel

Department of Chemistry,Hemchandracharya North Gujarat University, Patan, Gujarat-India

Corres.author: drkap_chem@yahoo.com

Abstract: Some new and biologically active1,2,4-triazolo[3,4-b][1,3,4] thiadiazole and their Schiff bases were synthesized by reaction of 3-(phenyl)-6-(4-N-acetylamino phenyl) [1,2,4]triazolo[3,4-b] [1,3,4]thiadiazole was hydrolysed by refluxing with methanole and Conc. HCL and alkaline with liquid ammonia. Biologically active Schiff bases are prepared by condensing these 1,2,4-triazolo[3,4-b][1,3,4] thiadiazole with different substituted aromatic aldehydes. All the newly synthesized compounds were confirmed by IR, ^{1}H NMR, and elemental analysis.
Keywords: Synthesis, Biological evaluation, 1,2,4-triazolo[3,4-b][1,3,4] thiadiazole, Schiff base.

Introduction

In recent literature is enriched with progressive findings about the synthesis and biological significance of fused heterocycls. Heterocycles bearing a thiadiazole moieties are reported to show biological properties such as antimicribial[1,2],anti tubercular[3,4], anti inflammatory[5] and anti convulsant[6]. A larg number of triazolo thiadiazoles have recently been reported to possess CNS depressant, anti bacterial, antifungal, anti tumor, anti-inflammatory[7], herbicidal, pesticidal and insecticidal properties[8,9]. In addition, the Schiff bases derived from 1,2,4-triazolo[3,4-b][1,3,4] thiadiazoles were also shown to possess an array of biological activities such as antifungal, antibacterial, and anti-inflammatory activity.[10,11].

In the present work, 1,2,4-triazolo[3,4-b][1,3,4] thiadiazole and their Schiff bases were synthesized by3-(phenyl)-6-(4-N-acetylamino phenyl) [1,2,4]triazolo[3,4-b] [1,3,4]thiadiazole was hydrolysed by refluxing with methanole and conc. HCL and alkaline with liquid ammonia. And their Schiff bases are prepared by condensing these 1,2,4-triazoleo[3,4-b][1,3,4] thiadiazole with different substituted aromatic aldehydes (a-j). Structure of these compounds were characterized by means of spectral data and elemental analysis.

Experimental Analysis

Synthesis of methyl benzoate (I).

Benzoic acid (0.01 mole) in 20 ml of methanol and 0.5 ml conc. Sulfuric acid was refluxed for 12 hrs. and poured into ice. The product was isolated and treated with standard sodium bicarbonate solution to give desired compounds. Yield: 96%.

Synthesis of benzoic acid hydrazide (II).

A mixture methyl benzoate (0.01 mole) and hydrazine hydrate (0.5 g, 0.01 mole) was heated for 9 hrs. and poured into ice. The product was isolated and crystallized from ethanol. Yield: 85%.

Synthesis of potassium-benzoic acid hydrazide dithiocarbamate (III).

A mixture of benzoic acid hydrazide (0.01 mole), KOH (0.84 g, 0.015 mole) and 1.5 ml CS_2 in absolute alcohol was stirred for 21 hrs. and product was isolated from diethyl ether. Yield: 87%.

Synthesis of 4-amino-5-phenyl-4H-1,2,4-triazole-3-thiol (IV).

Potassium salt (0.01 mole) was taken in hydrazine hydrate and heated up to the evolution of H_2S gas cused nearly 5 hrs. in oil bath. The reaction mixture was poured into crushed ice and treted with glacial Acetic acid .The product was filtered and purified by KOH treatement and crystallized from ethanol. Yield: 65%.

Synthesis of 3-(phenyl)-6-(4-N-acetylamino phenyl) [1,2,4]triazolo[3,4-b] [1,3,4]thiadiazole (V).

A mixture of n-acetyl-p-amino benzoic acid (0.01 mole) and 4-amino-5-phenyl-4H-1,2,4-triazole-3-thiol (0.01 mole) in $POCl_3$ (25 ml) was refluxed for 10 hrs. The reaction mixture was poured into crushed ice and thus solid separated out was filtered, washed with water and crystallized from ethanol.

Synthesis of 3-(phenyl)-6-(4-amino phenyl) [1,2,4]triazolo[3,4-b][1,3,4]thiadiazole (ATT) (VI).

3-(phenyl)-6-(4-N-acetylamino phenyl) [1,2,4] triazolo[3,4-b][1,3,4]thiadiazole was hydrolysed by refluxing with 75 ml of ethanol containing 15 ml of concentrated HCl for 4-5 hrs. it was then poured into ice-cold water and finally made just alkaline with liquid ammonia. The resultant product 3-(phenyl)-6-(4-amino phenyl) [1,2,4]triazolo[3,4-b][1,3,4]thiadiazole (ATT) is filtered off and washed with water and air dried. It was then recrystallised from ethanol to give product in near 60% yield. I.R. (KBr, cm^{-1}): 3362 (NH_2), 3030, 1500, 1600 (aromatic C-H), 1580 (C=N), 692, 1630 (NH-in and out plane), 1344 (C-S-C). PMR (δ ppm): 6.4-8.86 (m, aromatic CH). ^{13}CMR (δ ppm): 113, 130-150 (triazolo-thiadiazole), 120-129 (benzene).

Synthesis Arylidine-[3,6-(diphenyl)-[1,2,4]triazolo [3,4-b] [1,3,4]thiadiazole] (a-h) (VII)

A mixture of equimolar amount of 3-(phenyl)-6-(4-amino phenyl) [1,2,4]triazolo[3,4-b][1,3,4]thiadiazole (0.01 mole) and various aromatic aldehydes (0.01mole) in 50 ml acetic acid and refluxed for about 10-12 hrs. on oil bath. The reaction mixture was cooled and it was poured in to ice water and extracted with ethyl acetate and water and finally dried over anhydrous sodium sulfate. The solvent was evaporated to give the solid product. It was crystallized from ethyl acetate hexane using decolorizing charcoal to give various anils (a-h).

Spectral Analysis of Synthesized Schiff bases

(1) Benzylidine-[3,6-(diphenyl)- [1,2,4]triazolo 3,4-b] [1,3,4]thiadiazole (a)
I.R. (KBr, cm-1): 1040 (N-N), 3030-3080, 1600, 1500 (aromatic C-H stretching), 1630 (CH=N), 1095 (C-S). PMR (δ ppm): 6.4-8.86 (m, aromatic CH of CH=N protons). ^{13}CMR (δ ppm): 130-150 (triazolo-thiadiazole), 115-129 (benzene), 153 (CH=N).

(2) 4-Methoxy benzylidine-[3,6-(diphenyl)-[1,2,4] triazolo[3,4-b][1,3,4]thiadiazole (b)
I.R. (KBr, cm^{-1}): 1040 (N-N), 1200 (Ar-O-alkyl), 3030-3080, 1600, 1500 (aromatic C-H stretching), 1625 (CH=N), 1095 (C-S). PMR (δ ppm): 6.14-8.58 (m, aromatic CH of CH=N protons), 3.85 (3H, s, OCH_3). ^{13}CMR (δ ppm): 136-150 (triazolo-thiadiazole), 114-129 (benzene), 153 (CH=N), 113 (C-O), 56 (CH_3).

(3) 4-Hydroxy benzylidine-[3,6-(diphenyl)-[1,2,4] triazolo[3,4-b][1,3,4]thiadiazole (c)
I.R. (KBr, cm^{-1}): 1040 (N-N), 3370 (OH), 3030, 1600, 1500 (C-H stretching), 1627 (CH=N), 1095 (C-S). PMR (δ ppm): 6.3-8.1 (m, aromatic CH of CH=N protons), 3.85 (1H, s, OCH_3). ^{13}CMR (δ ppm): 130-150 (triazolo-thiadiazole), 114-129 (benzene), 153 (CH=N), 114 (C-O)

(4) 2-Hydroxy benzylidine-[3,6-(diphenyl)-[1,2,4] triazolo[3,4-b][1,3,4]thiadiazole (d)
I.R. (KBr, cm^{-1}): 1040 (N-N), 3370 (OH), 3030, 1600, 1500 (C-H stretching), 1625 (CH=N), 109.5 (C-S). PMR (δ ppm): 6.2-8.1 (m, aromatic CH of CH=N protons), 3.85 (1H, s, OH). ^{13}CMR (δ ppm): 136-150 (triazolo-thiadiazole), 114-129 (benzene), 153 (CH=N), 113 (C-O).

(5) 4-Methyl benzylidine-[3,6-(diphenyl)-[1,2,4] triazolo[3,4-b][1,3,4]thiadiazole (e)
I.R. (KBr, cm^{-1}): 1040 (N-N), 2950, 1370 (CH_3), 3030, 1600, 1500 (C-H stretching), 1625 (CH=N), 1095 (C-S). PMR (δ ppm): 6.2-8.1 (m, aromatic CH of CH=N protons), 2.5(3H, s, CH_3). ^{13}CMR (δ ppm): 186-150 (triazolo-thiadiazole), 114-129 (benzene), 150 (CH=N), 20.9 (CH_3),

(6) 3,4-Methylrnedioxy benzylidine-[3,6-(diphenyl)-[1,2,4]triazolo[3,4-b][1,3,4] thiadiazole (f)
I.R. (KBr, cm^{-1}): 1040 (N-N), 2920, 2850, 1450 (CH_2), 3030, 1600, 1500 (C-H stretching), 1640 (CH=N), 1095 (C-S). PMR (δ ppm): 6.1-8.1 (m, aromatic CH of CH=N protons), 5.9 (2H, s, O-CH_2-O). ^{13}CMR (δ ppm): 136-150 (triazolo-thiadiazole), 114-129 (benzene), 153 (CH=N), 91.3 (O-CH_2-O)

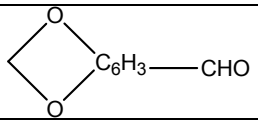
(7) 4-Hydroxy-3-methoxy benzylidine-[3,6-(diphenyl)-[1,2,4]triazolo[3,4-b] [1,3,4] thiadiazole (g)

I.R. (KBr, cm^{-1}): 1040 (N-N), 2950, 1370 (CH_3), 3030, 1600, 1500 (C-H stretching), 1640 (CH=N), 1095 (C-S). PMR (δ ppm): 6.2-8.1 (m, aromatic CH of CH=N protons), 3.36 (3H, s, OCH_3), 3.85 (1H, s, OH). ^{13}CMR (δ ppm): 136-150 (triazolo-thiadiazole), 114-129(benzene), 153 (CH=N), 56.3 (OCH_3).

(8) 3,4-Diethoxy benzylidine-[3,6-(diphenyl)-[1,2,4]triazolo[3,4-b][1,3,4] thiadiazole (h)

I.R. (KBr, cm^{-1}): 1040 (N-N), 2950, 2820, 1450 (CH_2), 3030, 1600, 1500 (C-H stretching), 1640 (CH=N), 1095 (C-S). PMR (δ ppm): 6.1-8.1 (m, aromatic CH of CH=N protons), 4.0 (4H, q, $2CH_2$), 1.33 (6H, t, $2CH_3$). ^{13}CMR (δ ppm): 136-150 (triazolo-thiadiazole), 114-129 (benzene), 153 (CH=N), 14.3 (CH_3), 65.4 (CH_2), 113 (C-O).

Table-1. Physical constant of arylidine-[3,6-(di phenyl)-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazole] (a-h)

Sr. No	R	Molecular Formula	Molecular Weight	M.P. °C	Yield %	% of C, H, N, S Cal / Found			
						C	H	N	S
a	C_6H_5-CHO	$C_{22}H_{15}N_5S$	381	176	77	69.2	3.9	18.3	8.3
						69.0	3.7	18.1	8.2
b	OCH_3-C_6H_4-CHO	$C_{23}H_{17}N_5OS$	411	247	73	67.1	4.1	17.0	7.7
						67.0	3.9	16.8	7.6
c	4-OH-C_6H_4-CHO	$C_{22}H_{15}N_5OS$	397	273	72	66.4	3.7	17.6	8.0
						66.0	3.7	17.4	7.8
d	2-OH-C_6H_4-CHO	$C_{22}H_{15}N_5OS$	397	246	77	66.4	3.7	17.6	8.0
						66.2	3.5	17.5	7.8
e	4-CH_3-C_6H_4-CHO	$C_{23}H_{17}N_5S$	395	232	78	69.8	4.3	17.7	8.1
						69.5	4.2	17.5	8.0
f		$C_{23}H_{15}N_5O_2S$	425	225	68	64.9	3.5	16.4	7.5
						64.7	3.4	16.3	7.4
g	4-OH-3-OCH_3-C_6H_3-CHO	$C_{23}H_{17}N_5O_2S$	427	234	72	64.6	3.9	16.3	7.4
						64.4	3.7	16.1	7.3
h	3-OC_2H_5-4-OC_2H_5-C_6H_3-CHO	$C_{26}H_{23}N_5O_2S$	469	238	63	66.5	4.9	14.9	6.8
						66.3	4.8	14.8	6.7

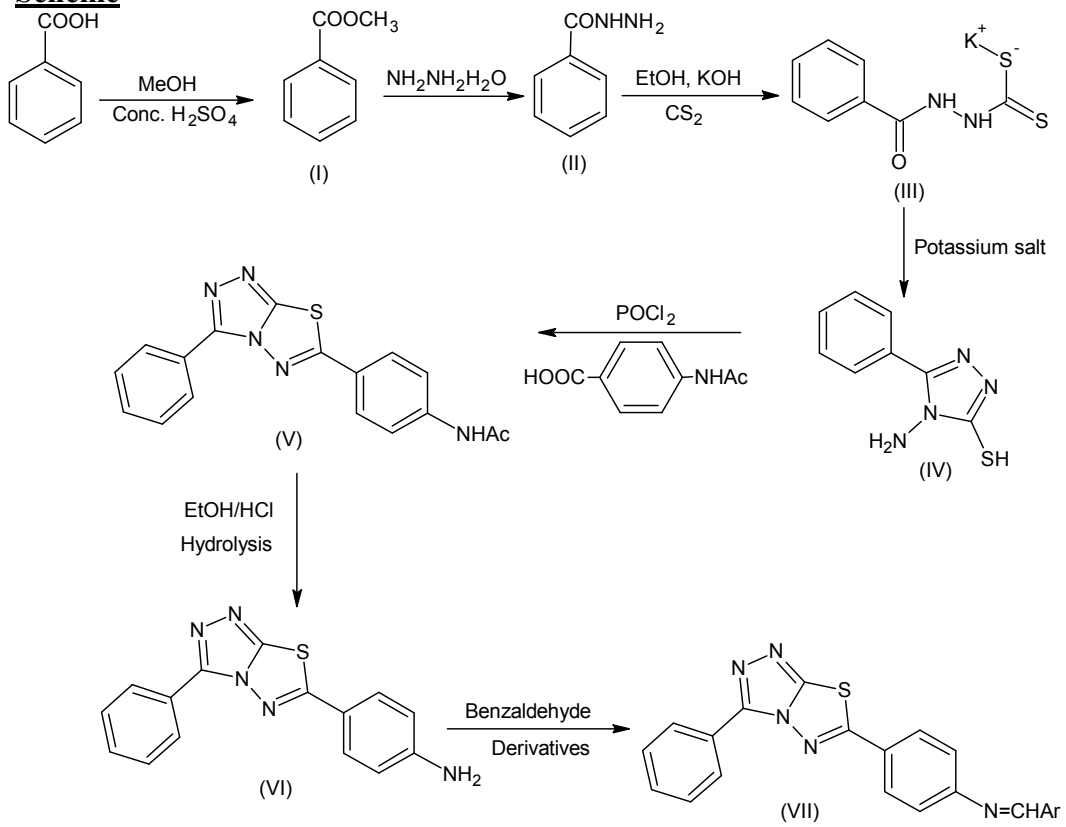
Results and Discussion

All the schiff bases of 1,2,4-triazolo[3,4-b][1,3,4] thiadiazoles were synthesized and the structure of schiff bases were established by means of IR, ^{1}H NMR spectral data as well as elemental analysis.

All the Schiff bases were evaluated for anti-microbial activity. The Schiff bases (a-h) were obtained by the treatment of 3-(phenyl)-6-(4-amino phenyl) [1,2,4]triazolo[3,4-b] [1,3,4]thiadiazole and various aromatic aldehydes in acetic acid and refluxed for about 10-12 hrs. on oil bath. The reaction mixture was cooled and it was poured in to ice water and extracted with ethyl acetate and water and finally dried over anhydrous sodium sulfate. The solvent was evaporated to give the solid product. It was crystallized from ethyl acetate hexane using decolorizing charcoal to give various anils (a-h).

All the synthesized structures showed satisfactory result. The IR data of the compounds clearly showed a strong C=N stretching band around 1640 cm^{-1} and a C-S-C linkage in thiadiazole of absorption band around 1344 cm^{-1}.This indicates that the formation of 1,3,4 thiadiazole derivatives along with a. azomethine linkage. The ^{1}H NMR also confirms the presence of shift value at 6.4-8.86 for CH=N groups respectively. Antibacterial screening of newly synthesized compounds was carried out against against *B. subtillis, S. aureus, P. aeruginosa and E. coli* in DMF solvent using cup-plate method. From the results obtained in the biological activity, it was observed that, the compound e, g and h were shown significant activities and compound a, b, c and d, have shown moderate activity.

Scheme



Biological evaluation

Cup-plate agar diffusion method[12,13] was employed for *in vitro* study of antibacterial. Efficacy of the target compounds against *B. subtillis*, *S. aureus, Ps. aeruginosa and E. coli* in DMF solvent.

The study has been conducted according to the method adopted by Nutrient agar broth was melted in a water bath and cooked to 45^{O}C with gentle shaking to bring about uniform cooling. It was inoculated with 0.5-0.6 ml of 24 hour old culture especially and mixed well by gentle shaking before pouring on the sterilized Petri dish (25 ml each). The poured material was allowed to set (1.5 hour) and there after the "cups" were made by punching into the agar surface with a sterile cork borer and sooping out the punched part of agar. Into this "cups" 0.1 ml of test solution (prepared by dissolving 10gm of sample in 10ml DMF) was added by sterile micropipette. The plates were noted. Ampicillin, Tetracycline, Gentamycin, and Chloramphenicol were used as standard drugs. The biological activity test data are presented in Table 2. The compound e, g and h were shown significant activities and compound a, b, c and d have shown moderate activity.

Acknowledgement

The authors are thankful to Director, C.D.R.I, Lucknow (U.P.) and R.S.I.C,Chandigrah and National Chemical Laboratory, Pune for providing spectral and analytical data of the compounds. They are also thankful to the Microcare Laboratory, Surat (Gujarat) for the biological activity.

Table - 2. Antibactarial activity of standards and solvent (DMF)

Compounds	Zone of Inhibition in (mm)			
	Gram Positive		Gram Negative	
	B. Subtillis	S. Aureus	E. Coli	Ps. Aeruginosa
DMF	06	05	05	05
Ampicillin	19	15	20	21
Tetracycline	21	20	15	18
Gentamycine	20	18	19	22
Chloramphenicol	20	23	18	24

Tabel - 3. Antibacterial activity of arylidine-[3,6-(diphenyl)-[1,2,4]triazolo[3,4-b] [1,3,4]thiadiazole] (a-h)

Compounds	Zone of Inhibition in (mm)			
	Gram Positive		Gram Negative	
	B. Susbtillis	S. Aureus	E. Coli	Ps. Aeruginosa
a	10	14	14	11
b	11	12	11	09
c	13	12	13	11
d	10	12	09	08
e	16	17	13	18
f	12	11	09	14
g	19	20	13	16
h	14	15	16	17

References

1 Thomasco M.L. Godwood C.R. Weaver A.E. Ochoada M.J. and Ford W.C., Bioorg. Medi. Chem. Lett., 2003, 13, 4193.

2 Wadokar N.K. and Dhiman M.A., Indian J. of Chem., 2001, 40B, 636.

3 Joshi S.H. and Thakar K.A., Indian J. of Chem., 2005, 44B, 410.

4 Foroumadi A. Mirzaei A. and Shafiee A., II Farmaco, 2001, 56, 621.

5 Plaska E. Shahin G. Kelicen P. Duslu T.N. and Altino G., II Farmaco 2002, 57, 101.

6 Dogan N.H. Duran A. Rollas S. Sener G. Uysal K.M. and Gulen D., Bioorg. Med. Chem.,2002, 10, 2893.

7 Mohammad A. and Chhavi A.,Int.J.of ChemTech .Research., 2009, 1(4), 1200- 1205.

8 Ashok M. and Holla B. S., Phosphorous, Sulfur, and Silicon, 2007, 182, 981.

9 Prasad D.J. Karthikeyan M.S. Karegoudar P.B. and Holla B.S., Phosphorous, Sulfur, and Silicon, 2007, 182, 1083.

10 Mahadev B.T. Shoba R.D. Yallappa S.S. Subash C.M. and Shankar C.B., Ind. J. of hetro. chemi., 1996, 5, 215-218.

11 Silver stenin R.M. Bassler and Morrd T.C., spectrometric identification of organic compounds., 1979, 5, 328.

12 Seely H.W. and Van Denmark P. J., In. Microbes in action: A Lab. Manual of Microbiology, 2nd ed, 1975, 55.

13 A. L. Barry In. The Antimicrobial Susceptibility Test: Principle and practice,Lea and Febiger Philadelphia, 1976, 180.

*****