Synthesis and Characterization of 3-Amino-5-Arylimino-1,2,4 Thiadiazoles

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Abstract- **An efficient synthesis of the 3-amino-5 -arylimino-1,2,4 thiadiazoles by the oxidative cyclization of the intermediate 1-aryl-3-amidino thiocarbamide by iodine and ethanol has been worked out. The compounds were characterized on the basis of certain chemical transformations, their IR, ^{1}HNMR, ^{13}CNMR and mass spectral data. The synthesized compounds were screened for their antimicrobial activities. These compounds showed moderate to good antibacterial and antifungal activities.**

Key words: 3-Amino-5-arylimino-1, 2 , 4 thiadiazoles , oxidative cyclization, 1-aryl-3-amidino thiocarbamide

I. INTRODUCTION

Thiadiazole play a prominent role in nature. For example, the thiazolium ring present in vitamin B1 serves as an electron sink and its coenzyme form is important for the decarboxylation of α-keto acids[1]. Thiadiazole moiety acts as hydrogen binding domain and two-electron donor system [2] . Thiadiazole is a biologically identical to that of pyrimidine and oxadiazole and given the prevalence of pyrimidine in nature, it is not surprising that thiadiazole shown significant therapeutic potential properties, the sulfur atom of the thiadiazole imparts improved liposolubility and mesoionic nature reported as anti-parasitic, anti-convulsant and anti-coagulant[3], anti-microbial[4], anti-cancer[5], anti-inflammatory[6-7], anti-tubercular[8]. The literature survey reveals that the heterocyclic compounds having thiadiazole nucleus enhanced pharmaceutical, medicinal, agricultural and industrial values [9-12].

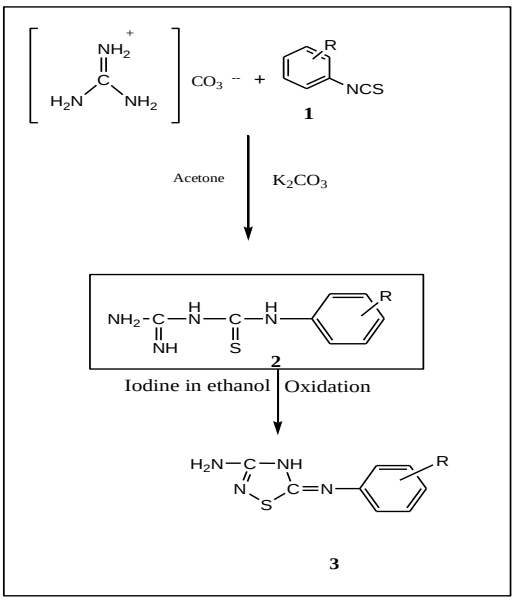
II. MATERIALS AND METHODS

The chemicals and reagents used in present work were of AR grade and LR grade purchased from SD fine chem. Ltd., and, Loba chem. Ltd., Amines used were aniline, o-toludine, p-toludine ,o-chloro aniline ,p-chloro aniline, o-anisidine, p-anisidine etc. The reaction progress was monitored by TLC technique by using suitable mobile phase of solvent. Purification of compounds were done by recrystalization method by using suitable solvent. Determination of melting point was done by using melting point apparatus and are uncorrected. IR spectra recorded on HAPP-GENZEL. ^{1}H NMR spectra on Bruker avance-II 400 NMR spectrometer at 400 MHz in $CDCl_3$ as solvent were recorded. The mass spectra were recorded on TOF MS ES+ 2.77e^{3} mass spectrometer. The compounds were screened for their antibacterial and antifungal activities by the agar diffusion method.

III. RESULTS AND DISCUSSION

We synthesized a series of 3-amino-5 arylimino-1,2,4 thiadiazoles by oxidative cyclization of 1-aryl -3-amidino thiocarbamides. For this 1-aryl -3-amidino thiocarbamides (1g, 0.005-0.0043 mole) were suspended in ethanol**.** To it iodine solution in ethanol was added in installments with continuous stirring. Initially, the colour of iodine disappeared, the addition was continued till the colour of iodine persisted. The reaction mixtures were left overnight at room temperature. The isolated residues were dissolved in ethanol and then were basified with dilute ammonia to get free bases. The ice was added to precipitate out the free bases. They were filtered, washed and crystallized from ethanol to yield 3-amino-5 arylimino-1,2,4 thiadiazoles .
The structures of the target compounds have been established by IR, ^{1}H-NMR, ^{13}C-NMR and mass structural data.

Reaction scheme



Where R in 1,2,3 a)-CH_3(o),b)-CH_3(m),c)-CH_3(p),d)-H, e)-Cl(p), f)-OCH_3(o), g)-OCH_3(p),h)-Cl(o)

The 3-amino-5 arylimino-1, 2, 4 thiadiazoline (3b-h) were prepared by the above reaction and the products were isolated in good yields. (Table 1.1).

Table 1.1 Physical data of 3 amino-5 arylimino-1,2,4 thiadiazoline (3)

Reactants:- 1-Aryl-3-amidino thiocarbamides (2)
And Iodine in ethanol.

1-Aryl-3-amidino thiocarbamides (2)	3- amino-5 aryl/alkylimino-1,2,4 thiadiazoline (3)	Yield %	mp °C	Elemental analysis Found(cal) (%) N S
1-o-tolyl-3-amidino Thiocarbamide (2a)	3- amino-5 o-tolylimino-1,2,4 thiadiazoline (3a)	67.10 %	139	27.10 15.48 (27.16) (15.55)
1-m-tolyl-3-amidino Thiocarbamide (2b)	3- amino-5 m-tolylimino-1,2,4 thiadiazoline (3b)	77.33 %	141	27.11 15.50 (27.16) (15.55)
1-p-tolyl-3-amidino Thiocarbamide (2c)	3- amino-5 p-tolylimino-1,2,4 thiadiazoline (3c)	64.28 %	170	27.10 15.45 (27.16) (15.55)
1-phenyl-3-amidino Thiocarbamide (2d)	3- amino-5 phenylimino-1,2,4 thiadiazoline (3d)	83.33 %	Above 320	29.08 16.57 (29.14)(16.68)
p-chlorophenyl-3-amidino thiocarbamide (2e)	3- amino-5 chlorophenylimino-1,2,4 thiadiazoline (3e)	76.75 %	162	24.67 14.12 (24.72) (14.15)
1-o-anisyl-3-amidino Thiocarbamide (2f)	3- amino-5 o-anisylimino-1,2,4 thiadiazoline (3f)	61.23 %	145	25.10 14.39 (25.21) (14.43)
1-p-anisyl-3-amidino Thiocarbamide (2g)	3- amino-5 p-anisylimino-1,2,4 thiadiazoline (3g)	77.35 %	127	25.18 14.38 (25.21) (14.43)
1-o-chlorophenyl-3-amidino Thiocarbamide (2h)	3- amino-o-chlorophenylimino-1,2,4 thiadiazoline (3h)	50%	156	24.67 14.10 (24.72) (14.15)

3 Amino-5 o-tolylimino-1,2,4 thiadiazoline (3a)

IR (KBr) ν max cm^{-1}: 3352 (N-H),3050 (C-H aromatic),1585 (C=N),1539(C=C aromatic ring stretch),1315 (C-N),754(C-S) ; ^1H NMR ($CDCl_3$) δ ppm: 7.40 (s,1H,NH),7.40-6.95 (m,4H.,Ar-H),5.86 (d, 2H,NH_2),2.28 (s,3H, Ar-CH_3); C^{13}-NMR($CDCl_3$) δ ppm : 179.98 (1C, thiadiazoline ring carbon), 166.43(1C, another thiadiazoline ring carbon),138.28-121.89
(6C,Ar-carbons),17.97 (1C,Ar- CH_3 carbon) ;MS(m/z): 206 (M)+, 207 (M^++1) protonated.
The molecular formula of 3a was established as $C_9H_{10}N_4S$.Colour: cream.

3-Amino-5 m-tolylimino-1,2,4 thiadiazoline (3b)

IR (KBr) ν max cm^{-1}: 3305 (N-H),3077 (C-H aromatic),1647 (C=N),1564(C=C aromatic ring stretch),691(C-S)); ^1H NMR ($CDCl_3$) δ ppm: 7.42 (s,1H,NH),7.27-7.10 (m,4H.,Ar-H),5.98 (d, 2H,NH_2),2.31 (s,3H, Ar-CH_3); C^{13}-NMR($CDCl_3$) δ ppm : 177.56 (1C, thiadiazoline ring carbon), 166.47(1C, another thiadiazoline ring carbon),139.98-114.54(6C, Ar-carbons),20.96 (1C,Ar- CH_3) ;MS(m/z):207 (M^++1) protonated.
The molecular formula of 3b was established as $C_9H_{10}N_4S$.Colour: cream.

3-Amino-5 p-tolylimino-1,2,4 thiadiazoline (3c)

IR (KBr) ν max cm^{-1}: 3305 (N-H),3098 (C-H aromatic),1611 (C=N),1515(C=C aromatic ring stretch),1311 (C-N),732(C-S) ; ^1H NMR ($CDCl_3$) δ ppm: 7.44 (s,1H,NH),7.49-6.99 (m,4H.,Ar-H),5.77 (d, 2H,NH_2),2.28 (s,3H, Ar-CH_3); C^{13}-NMR($CDCl_3$) δ ppm : 177.86 (1C, thiadiazoline ring carbon), 166.31(1C, another thiadiazoline ring carbon),137.49-98-117.54(6C, Ar-carbons),20.47 (1C,Ar- CH_3) ;MS(m/z):207 (M^++1) protonated.
The molecular formula of 3c was established as $C_9H_{10}N_4S$.Colour: cream.

3-Amino-5 phenylimino-1,2,4 thiadiazoline (3d)

IR (KBr) ν max cm^{-1}: 3448 (N-H),3150 (C-H aromatic),1432 (C=N),1515(C=C aromatic ring stretch),712(C-S); MS(m/z):192 (M^+) .
The molecular formula of 3d was established as $C_8H_8N_4S$.Colour: white

3-Amino-5 p-chlorophenylimino-1,2,4 thiadiazoline (3e)

IR (KBr) ν max cm^{-1}: 3294 (N-H),3096 (C-H aromatic),1616 (C=N),1570(C=C aromatic ring stretch),1311 (C-N),732(C-S) ; ^1H NMR ($CDCl_3$) δ ppm: 7.45 (s,1H,NH),7.57-7.31 (m,4H.,Ar-H),6.13 (d, 2H,NH_2); C^{13}-NMR($CDCl_3$) δ ppm : 177.09 (C-3, thiadiazoline ring carbon), 166.52(C-5, another thiadiazoline ring carbon),138.89-118.74(6C, Ar-carbons) ;MS(m/z):227 and 229 (M^+ +1) 3:1 ratio.
The molecular formula of 3e was established as $C_8H_7N_4SCl$.Colour: cream.

3-Amino-5 o-anisylimino-1,2,4 thiadiazoline (3f)

IR (KBr) ν max cm^{-1}: 3370 (N-H), 3068 (C-H aromatic),1605 (C=N),1542(C=C aromatic ring stretch),1336 (C-N),748(C-S)); The molecular formula of 3f was established as $C_9H_{10}N_4SO$.Colour: faint yellow

3-Amino-5 p-anisylimino-1,2,4 thiadiazoline (3g)

IR (KBr) ν max cm^{-1}: 3321 (N-H),3100 (C-H aromatic),1606 (C=N),1547(C=C aromatic ring stretch),1332 (C-N),729(C-S) ; ^1H NMR ($CDCl_3$) δ ppm: 7.48 (s,1H,NH),7.39-6.91 (m,4H.,Ar-H),6.40 (d, 2H,NH_2),3.77 (s,3H, Ar-OCH_3); C^{13}-NMR($CDCl_3$) δ ppm : 180.18 (C-3, thiadiazoline ring carbon), 162.60(C-5, another thiadiazoline ring carbon),141.60-121.68 (6C, Ar-carbons) 156 (Ar-OCH_3);MS(m/z): 223 (M^+ +1).
The molecular formula of 3g was established as $C_9H_{10}N_4SO$.Colour: faint yellow

3-Amino-5 o-chlorophenylimino-1,2,4 thiadiazoline (3h)

The molecular formula of 3h was established as $C_8H_7N_4SCl$ Colour: cream.

The compounds 3-amino-5-arylimino-1,2,4 thiadizoline (3a to 3h) synthesized here have exhibited fairly good antibacterial (table 1.2) and antifungal activity (table 1.3). Inhibition zone record of the compounds showed that, the compound 3e was highly sensitive against E.coli and S.aureus. Other compounds were inactive against E.coli as well as S.aureus. Compounds 2e showed high sensitivity against A.niger and C.albicans. Other compounds were resistant to both the fungus strains.

Table- 1.2:- Antibacterial activity of 3-Amino-5 arylimino-1,2,4 thiadiazoline(3a to 3h)

Organism	3a	3b	3c	3d	3e	3f	3g	3h
E.coli	-	-	-	-	+++ (50mm)	-	-	-
S.aureus	-	-	-	-	+++ (60mm)	-	-	-

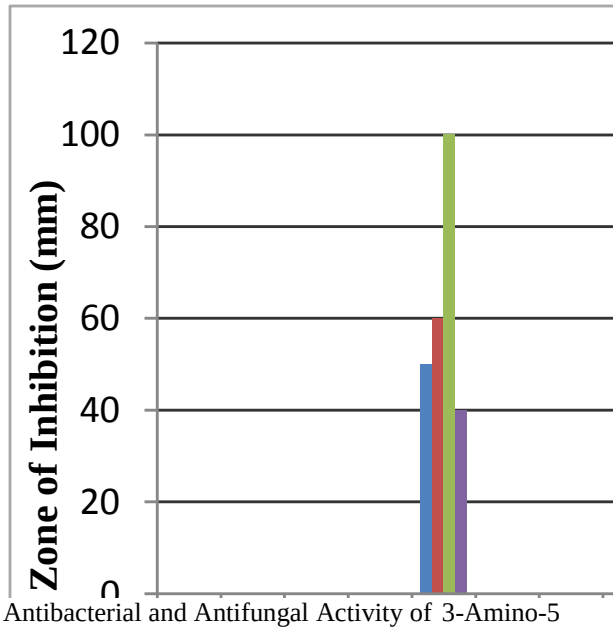
(Diameter of inhibition zone in mm)
(Concentration 500 µ g/ml)

Table- 1.3:-Antifungal activity of 3-Amino-5 arylimino-1,2,4 thiadiazoline (3a to 3h)

Organism	3a	3b	3c	3d	3e	3f	3g	3h
A.niger	-	-	-	-	+++ (100mm)	-	-	-
C,albicans	-	-	-	-	+++ (40mm)	-	-	-

(Diameter of inhibition zone in mm)
(Concentration 500 µ g/ml)

Antibacterial and antifungal activity of 3-Amino-5-arylimino-1,2,4 thiadiazoline (3a to 3h)



Antibacterial and Antifungal Activity of 3-Amino-5 arylimino-1,2,4 thiadiazoline

IV.CONCLUSION

Literature on thiadiazoles revealed that less work is carried out on 1,2,4 thiadiazoles as compared to its other isomers The presented synthetic procedure is convenient and simple method for the synthesis of the various 1,2,4 thiadiazoles. The compound 3-amino-5 p-chlorophenylimino-1,2,4 thiadiazoline has shown significant biological activity. Presently antibacterial and antifungal activities are reported. Further screening for some other activities in related field may prove their more utility.

V.EXPERIMENTAL

Melting points are uncorrected and were measured using electro thermal apparatus. FT-IR spectra were recorded using KBr disk on Perkin Elmer FT-IR KBR spectrophotometer recorded with ν max in inverse centimeters. 1H NMR spectra were recorded on Bruker avance-II 400 NMR spectrometer at 400 MHz in DMSO/$CDCl_3$ as solvent. The spectra were recorded using tetramethylsilane as internal standard and chemical shifts being reported in parts per million (δ) relative to TMS. The mass spectra were obtained using Waters Q-TOF Micromass instrument. The progress of the reaction was monitored by TLC on Merck Silica Gel 60 F 254 plates with detection by UV light and I_2 vapours as visualizing agent. The compounds were screened for their antibacterial and antifungal activities by the agar diffusion method.

Preparation of aryl isothiocyanates: The aryl isothiocyanate were prepared by already known procedure.[13]

Preparation of 1-aryl-3-amidino thiocarbamide: 1-aryl-3-amidino thiocarbamides were prepared by the reaction of guanidine carbonate (1.8 g, 0.01 mole), and aryl isothiocyanate (1a-h) (0.01mole) in presence of potassium carbonate (1.42g, slightly more than 0.01 mole) by refluxing for 2 hours in acetone. The reaction mixtures were cooled, the solvent was distilled off when residues were obtained to yield 1-aryl-3-amidino thiocarbamides (2a-h)

Preparation of 3-amino-5 arylimino-1,2,4 thiadiazoline
1g 1-Aryl-3-amidino thiocarbamide (0.005-0.0043mole) (1a-1h) were suspended in 5 ml ethanol. To it iodine solution in ethanol was added in installments with continuous stirring. Initially, the colour of iodine disappeared, the addition was continued till the colour of iodine persisted. The reaction mixtures were left overnight at room temperature. The isolated residues were dissolved in 20 ml ethanol and they were basified with dilute ammonia to get free bases. The ice was added to precipitate out the free bases. They were filtered, washed and crystallized from ethanol to get 3a-3h.
Antibacterial: Initially, the stock cultures of bacteria were revived by inoculating in broth media (peptone-10g, NaCl-10 g and Yeast extract 5g, Agar 20g in 1000ml of distilled water) and grown at 37 °C for18 hours. The agar plates of the above media were prepared and wells were made in the plate. Each

plate was inoculated with 18 hour old cultures (100 µl,10^{-4} cfu) and spread evenly on the plate. After 20 minutes, the wells were filled with the solution of compounds at different concentrations. The control wells with Gentamycin were also prepared. All the plates were incubated at 37 °C for 24 hours and the diameter of inhibition zone were measured.[14] Antifungal: Potato dextrose agar 250g of peeled potato were boiled for 20 minutes and squeezed and filtered. To this filtrate 20 g of dextrose was added and the volume was made up to 1000 ml by distilled water. Initially, the stock cultures of fungi were prepared and wells were made in the plate. Each plate was inoculated with 48 hour old cultures (100 µl,10^{-4}cfu)and spread evenly on the plate .After 20 minutes, the wells were filled with solution of compounds at different concentrations .The control plates with antibiotic Amphotericin were also prepared. All the plates were incubated at 27 °C for 48 hours and the diameter of inhibition zone were measured.[15]

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