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Research Article

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Synthesis and antimicrobial screening of some substituted 1, 3, 4- Oxadiazole derivatives

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ABSTRACT

A new series of ten compounds of 1,3,4 oxadiazoles were synthesized by ring closure reactions of different acid hydrazides with carbon disulphide and aromatic acids separately. Further, the 2-thio oxadiazoles were treated with aromatic amines to get mannich bases. The structures of the synthesized compounds were consistent with IR, ^1H NMR and MASS spectroscopy. The synthesized compounds were screened by disc diffusion method for their antibacterial and antifungal activity. Among the compounds synthesized 3d, 3e and 5b shown significant antimicrobial activity against both bacterial and fungal strains.

Keywords: 1,3,4-oxadiazole, Mannich bases, Antibacterial and Antifungal activity.

INTRODUCTION

1,3,4 oxadiazoles possess multi-range of biological actions such as antibacterial [1], anifungal [2], antiinflammatory [3 - 4], herbicidal, anticonvulsant, antitubercular [5] and anticancer activity, etc. In the present study newer analogs of 2,5-disubstituted oxadiazoles were synthesized from various acid hydrazides, which is obtained by reacting respective esters with hydrazine hydrate. The acid hydrazides were reacted with different aromatic acids to get 2,5-disubstituted 1,3,4-oxadiazoles (3a-e). Mannich bases of oxadiazoles were synthesized by converting acid hydrazide into 2-thio oxadiazoles followed by subjecting into mannich reaction (5a-e). All the synthesized compounds were screened for *in vitro* antimicrobial activity by disc diffusion method.

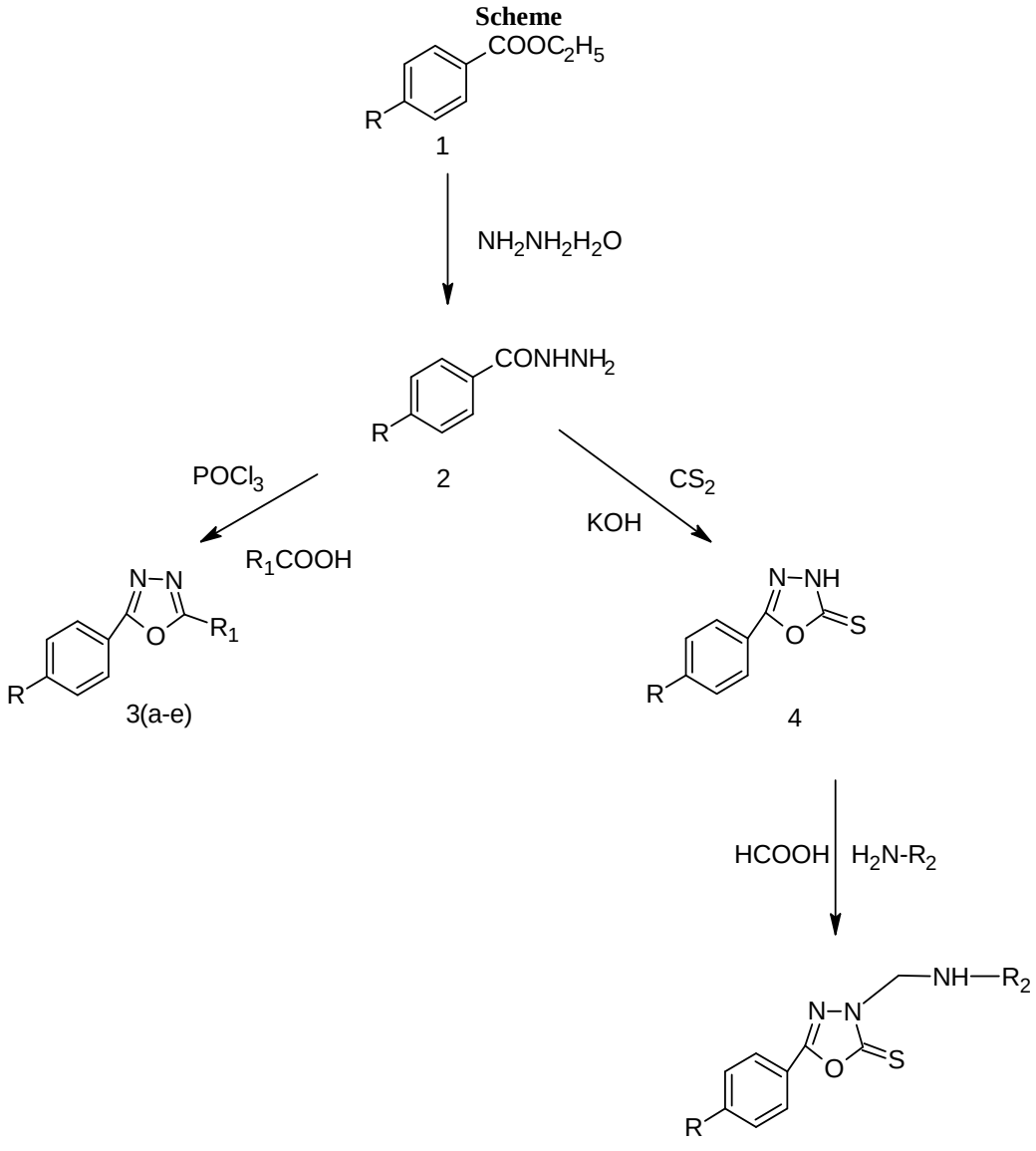
EXPERIMENTAL SECTION

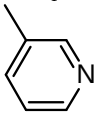
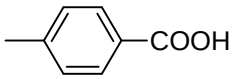
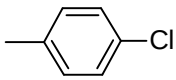
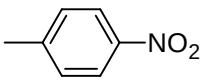
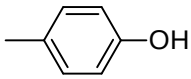
Melting points were taken in open capillary tubes and are uncorrected. The purity of the compounds was monitored by thin layer chromatography on pre-coated silica gel GF 254 plates. IR spectra were recorded through KBr pellet method in Perkin-Elmer FTIR spectrophotometer. ^1H NMR spectra was recorded on BRUKER ADVANCE II 400 NMR spectrometer with tetramethyl silane as an internal standard. The mass spectrum of compounds was recorded on JEOL GC Mate spectrophotometer.

Synthesis of 4-substituted acid hydrazides (2):
The starting material 4-substituted ethyl benzoate (1) was prepared by esterifying corresponding aromatic acids with ethanol in the presence of concentrated sulphuric acid [6]. A mixture of 4-substituted ethyl benzoate (0.01 mol), hydrazine hydrate (0.15 mol) and 30ml of ethanol was refluxed for 4 hr and the excess of ethanol was distilled off and the contents were poured into ice cold water and the precipitated hydrazides were filtered, dried and recrystallized from ethanol.

Synthesis of 2,5-disubstituted-1,3,4-oxadiazoles (3a-e):
The acid hydrazide (2) (0.01 mol) and aromatic acid (0.01 mol) were refluxed in $POCl_3$ (5 ml) for 8 h, cooled and poured into crushed ice and neutralised with sodium bicarbonate solution. The precipitate was filtered off, dried and recrystallized from ethanol [7].

Scheme



CODE	R	R_1	CODE	R	R_2
3a	Cl		**5a**	Cl	
3b	OH		**5b**	OH	
3c	NO_2		**5c**	NO_2	
3d	NO_2		**5d**	Cl	
3e	NO_2		**5e**	OH	

Synthesis of 5-(substituted)-2-thio-1,3,4-oxadiazoles (4):
The acid hydrazides (2) (0.01 mol) was dissolved in cold ethanol (15 ml) followed by added carbon disulphide (2 ml) and potassium hydroxide (0.6 g). The reaction mixture was refluxed until the evolution of hydrogen sulphide gas ceased (around 12 h). Excess solvent was evaporated and the residue was dissolved in water and then acidified with dilute hydrochloric acid (10%) to pH~5. The precipitate was filtered off, dried and recrystallized from ethanol.

Synthesis of Mannich bases of 1,3,4-oxadiazoles (5a-e):
Equimolar quantity of 5-(4-substituted) phenyl-1,3,4-oxadiazol-2(3*H*)-thione (4) (0.01 mol) and different amines were dissolved in ethanol (20 ml) and followed by added drop by drop of formaldehyde solution (0.01 mol). Stirred the contents for 1 h at room temperature and then refluxed for 2-3 hours. Then the content was kept at overnight at the temperature of 5-8^{0}C. The reaction mixture was concentrated and the product was filtered, dried and recrystallized from ethanol [8].

Table 1- Physical data of the compounds

Code	Molecular formula	Molecular Weight	Yield (%)	Melting Point (°C)	*R_f value
3a	$C_{13}H_8N_3OCl$	257.67	70.81	162	0.50
3b	$C_{13}H_9N_3O_2$	239.22	63.02	186	0.55
3c	$C_{13}H_8N_4O_3$	268.22	73.30	210	0.52
3d	$C_{14}H_8N_3O_3Cl$	301.68	77.07	190	0.46
3e	$C_{14}H_9N_3O_4$	283.23	65.37	205	0.35
5a	$C_{16}H_{12}N_3O_3SCl$	361.80	78.45	180	0.58
5b	$C_{16}H_{13}N_3O_4S$	343.35	86.87	196	0.56
5c	$C_{16}H_{12}N_4O_5S$	372.35	92.21	189	0.38
5d	$C_{15}H_{11}N_4O_3SCl$	362.79	91.16	175	0.79
5e	$C_{15}H_{12}N_4O_4S$	344.34	84.58	182	0.75

** Solvent system: Ethylacetate : Hexane (1:4)*

Antimicrobial activity:
The synthesized compounds were tested for their *in vitro* antibacterial activity against *Bacillus cereus, Staphylococcus aureus* (gram positive) and *Pseudomonus aeruginosa, Klebsiella pneumonia* (gram negative) and the antifungal activity was screened against *Candida albicans* and *Aspergillus fumigatus* at the concentrations of 50, 100 and 150 µg/ml by disc diffusion method.

Ciprofloxacin and Ketoconazole were used as standard drugs for antibacterial and antifungal activity respectively. Dimethyl sulfoxide was used as a control and the zone of inhibition of the compounds are presented in the table.
Sterile disc of 5 mm in diameter made from Whatmann filter paper which is previously sterilized in U.V. lamp was dipped in solution of different concentrations of synthesized compounds, standard and blank and placed the disc on the surface of agar plates.

Allowed the plates to stand for 1 h at room temperature as a period of pre-incubation to minimize the effects of variation in time between the applications of different solutions. Then the plates were incubated for 24 h at 37^o C ± 1^o C for bacteria and 72 h at 25^{o}C± 1^{o}C for fungi. The diameter of zone of inhibition was measured.

RESULTS AND DISCUSSION

Five derivatives of each of 2,5-disubstituted and mannich bases of 1,3,4-oxadiazole were synthesized from acid hydrazides. The structures of the synthesized compounds were confirmed on the basis of IR, ^{1}H NMR and Mass spectroscopy. All the compounds were screened for their *in vitro* antibacterial and antifungal activities.

The compounds 3d, 3e, 5b and 5d shown good antibacterial activity against gram positive organisms and the compounds 3c, 3d, 3e and 5b could show better action against gram negative organisms. About antifungal screening, the compounds 3d, 3e, 5a and 5b shown significant activity.

To conclude, 3d, 3e and 5b are fruitful compounds among the series synthesized for both bacterial and fungal inhibition.

Spectral data:
(3a): IR (cm^{-1}): 3085.09 (Ar CH str), 1479.44 (C=C, C=N str), 1106.94 (C-O-C str), 735.76 (C-Cl str). **^{1}H NMR (δ ppm):** 9.31 (m, 4H, pyridine), 7.53 (d, 4H, Ar). **m/z:** 257 (M^+), 212, 185.

(3b): IR (cm^{-1}): 3052.32(Ar CH str), 2844.75(Ar OH str), 1494.93(C=C,C=N str), 1096.86(C-O-C str). **^{1}H NMR (δ ppm):** 8.70 (m, 4H, pyridine), 6.94-7.82 (d, 4H, Ar), 5.07(s, 1H, OH). **m/z:** 239(M^+), 210, 185.

(3c): IR (cm^{-1}): 3063.24 (Ar CH Str), 1552.81(Ar NO_2 Str), 1481.84(C=C, C=N Str), 1085.23(C-O-C Str). **^{1}H NMR (δ ppm):** 9.35(m, 4H, pyridine), 8.44-8.46(d, 4H, Ar). **m/z:** 268(M^+), 212, 198.

(3d): IR (cm^{-1}): 2925.72 (Ar CH Str), 1556.18 (Ar NO_2 Str), 1480.13(C=C,C=N Str), 1071.64 (C-O-C Str), 732.80 (C-Cl bend). ^{1}**H NMR (δ ppm):** 8.46(m, 4H, Nitrobenzene), 7.59 (m, 4H, Chlorobenzene). **m/z:** 301 (M^+), 219, 185.

(3e): IR (cm^{-1}): 2918.46 (Ar CH Str), 2849.42 (Ar OH Str), 1521.88 (Ar NO_2 Str), 1491.47 (C=C,C=N Str), 1074.39 (C-O-C Str). ^{1}**H NMR (δppm):** 5.10 (s, 1H, OH), 6.79 – 8.20 (m, 8H, Ar). **m/z:** 283(M^+), 216, 188.

(5a): IR (cm^{-1}): 3316.90 (NH Str), 3027.53 (Ar CH Str), 1606.74 (Ar C=C str), 1252.83 (C=S str), 1074.39 (C-O-C Str), 722.19 (C-Cl str) ^{1}**H NMR (δ ppm):** 2.57(s, H, COOH), 5.78 (t, H, NH), 6.94 (d, 2H, CH_2), 7.57-7.86 (m, 8H, Ar). **m/z:** 362(M+), 240, 129.

(5b): IR(cm^{-1}): 3325.16 (NH Str), 3298.94 (OH Str), 3016.25 (Ar CH Str), 1605.71 (Ar C=C Str), 1280.14 (C=S Str), 1076.66(C-O-C Str). ^{1}**H NMR (δ ppm):** 2.59 (s, H, COOH), 5.06 (s, 1H, OH), 5.50 (m, H, NH), 6.73 (d, 2H, CH_2), 6.82 - 7.85 (m, 8H, Ar). **m/z:** 343(M^+), 250, 193.

(5c): IR (cm^{-1}): 3423.41 (NH Str), 3054.28 (Ar CH Str), 1604.98 (Ar C=C Str), 1526.19 (Ar NO_2 Str), 1255.17 (C=S Str), 1111.19 (C-O-C Str)**.** ^{1}**H NMR (δ ppm):** 2.56 (s, H, COOH), 5.42 (t, H, NH), 6.11 (d, 2H, CH_2), 7.87-8.35 (m, 8H, Ar). **m/z:** 372(M^+), 251, 129.

(5d): IR (cm^{-1}): 3366.10 (NH Str), 3089.73 (Ar CH Str), 1603.53 (Ar C=C Str), 1531.18 (Ar NO_2 Str), 1258.31(C=S Str), 1111.69 (C-O-C Str), 752.59 (C-Cl str). ^{1}**H NMR (δ ppm):** 5.96 (t, H, NH), 6.79 (d, 2H, CH_2), 7.53-8.05 (m, 8H, Ar). **m/z:** 363 (M^+), 251, 129.

(5e): IR (cm^{-1}): 3398.14 (NH Str), 3251.27 (OH Str), 3021.39 (Ar CH Str), 1609.75 (Ar C=C Str), 1559.75 (Ar NO_2 Str), 1253.17 (C=S Str), 1089.96 (C-O-C Str). ^{1}**H NMR (δ ppm):** 5.59 (t, H, NH), 6.07 (d, 2H, CH_2), 6.75 (m, 4H, Hydroxybenzene), 8.15 (m, 4H, Nitrobenzene). **m/z:** 344(M^+), 222, 176.

Table 2-Antibacterial activity

Code	Zone of Inhibition (in mm)											
	Gram positive						Gram negative					
	Bacillus Cereus (µg/ml)			*Staphylococcus aureus* (µg/ml)			*Pseudomonus aeruginosa* (µg/ml)			*Klebsiella Pneumonia* (µg/ml)		
	50	**100**	**150**	**50**	**100**	**150**	**50**	**100**	**150**	**50**	**100**	**150**
3a	13	16	20	12	14	19	14	16	20	13	15	19
3b	13	15	18	13	15	18	12	15	19	12	14	16
3c	14	16	19	11	14	18	16	18	22	16	18	21
3d	15	18	20	15	18	20	13	19	24	12	18	23
3e	16	19	21	15	19	22	15	18	25	14	17	24
5a	14	16	19	14	17	20	13	15	18	12	15	17
5b	15	18	21	15	17	21	15	18	21	13	16	20
5c	12	15	19	12	15	18	12	16	20	12	15	18
5d	13	15	21	16	18	21	12	14	16	12	14	17
5e	14	17	19	12	15	18	12	15	18	13	16	18
Ciprofloxacin (10 µg/ml)	39			38			38			39		

Table 3- Antifungal activity

CODE	Zone of Inhibition (in mm)					
	Candida albicans (µg/ml)			*Aspergillus fumigates* (µg/ml)		
	50	**100**	**150**	**50**	**100**	**150**
3a	15	18	20	14	19	21
3b	12	16	19	13	18	20
3c	15	18	21	15	20	22
3d	17	21	23	16	22	24
3e	14	18	22	15	19	23
5a	17	20	23	20	24	26
5b	15	20	22	16	21	22
5c	16	19	22	15	18	20
5d	15	18	21	16	18	20
5e	13	17	20	15	19	22
Ketoconazole (10 µg/ml)	38			39		

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REFERENCES

[1] S Vijayaraghavan; RR Somani; PY Shirodkar; VJ Kadam. *Int. J. PharmTech Res*., **2009,** 1(3), 811-815.
[2] K Selvakumar; K Anandarajagopal; V Rajamanickam; T Ajaykumar; B Jesindha. *Int.J. Pharm Sci. rev. Res*., **2011**, 6(1), 64-67.
[3] V Priya Frank; KS Girish; Balakrishna Kalluraya. *J. Chem. Sci*., **2007,** 119(1), 41-46.
[4] SR Pattan; CK Hariprasad; S Nachiket; SB Bhawar; SV Hiremath; BM Ingalagi. *Int. J.Pharm. res. dev*., **2009**, 1(9), 1-10.
[5] SR Pattan; PA Rabara; S Jayashri; AA Bukitager; VS Wakale; DS Musmade. *Indian J. Chem*., **2009**, 48B, 1453-1456.
[6] BS Furniss; AJ Hannaford; WG smith and AR Tatchell. Vogel's Textbook of Practical organic Chemistry, 5th Edition, Dorling Kindersly Publishers Inc, New Delhi, **2007**; 1077-1078.
[7] Keshari Kishore Jha; Yatendra Kumar; Mohd.Shaharyar. *Asian J. Chem*., **2009**, 21(9), 7403-7410.
[8] KC Ravindra; HM Vagdevi; VP Vaidya; P Basavaraj. *Indian J. Chem*., **2006**, 45B, 2506-2511.