A Synthesis of Substituted 1,3,4-oxadiazole, pyrazole deravatives and it's biological activities

ABSTRACT

Purpose: Nicotine Substituted 1,3,4 Oxadiazole and pyrazole moieties were synthesised and their antibacterial activity was evaluated.

Methods : TLC on silica gel G was used to check for homogeneity of the title compound. H NMR, Mass, and IR Spectra were used to characterise these compounds, and their antibacterial activity was tested.

Results : All of these compounds, including nicotine substituted ester, hydrazide, 1,3,4 Oxadiazole, and pyrazole, showed antibacterial activity. The maximum activity of enaminones was comparable to that of the standard drug ciprofloxacin (5mcg).

Conclusion : In the zone of inhibition studies, all six samples at MIC concentrations showed antimicrobial activity. Antibacterial activity was demonstrated for the compounds.

Keywords: Enaminones,Nicotinic Hydrazide, Nictonic Substituted Oxadiazole, Nicotin Substituted Pyrazole.

1. INTRODUCTION

Among the wide range of heterocycles Oxadiazole and pyrazole have been explored to develop new therapies[1-7]. Pyrazole and oxadiazole have attracted lot of attention to medical chemists. Wide spread use of Oxadiazole and pyrazole scaffolding a medical use . As a result, oxadiazole has a diverse set of biological properties, including antibacterial activity ,[8-9] antifungal[10-11] antitumor [12]antitubercular ,[13] anticonvulsant [14]and HIV-I inhibitory activities.[15]

The same range of medicinal significance is associated with compounds containing pyrazole. This article is an attempt to synthesize five membered heterocycles by using hydrazide bridge.

2. MATERIALS & METHODS

The open capillary method was used to determine the melting points, and the results are uncorrected. TLC on silica gel G was used to check for homogeneity of the compounds. Synthesised compounds were confirmed by 1HNMR ,Mass and IR spectra.

General procedure

1. Acid hydrazide Synthesis

By cyclo condensation process, ethyl 2-methyl-6- ethyl nicotinates was obtained via the reaction of enaminons again it is made to react with ethylene acetoacetate and ammonium acetate by refluxing with Using alkyl substituted Carboxylic acid and it is achieved via the hydrazinolysis of ester derivatives by refluxing in the presence hydrazine hydrate. [16]

2. Synthesis of compound 4:

Phosphorus oxide chloride, acid hydrazide, and aromatic substituted carboxylic acid were dissolved and refluxed for 16 to 22 hours.. The reaction mixture was slowly poured onto crushed ice and allowed to set overnight. Filtration was used to collect the separated solid, which was then dried and purified using ethanol.[17][18]

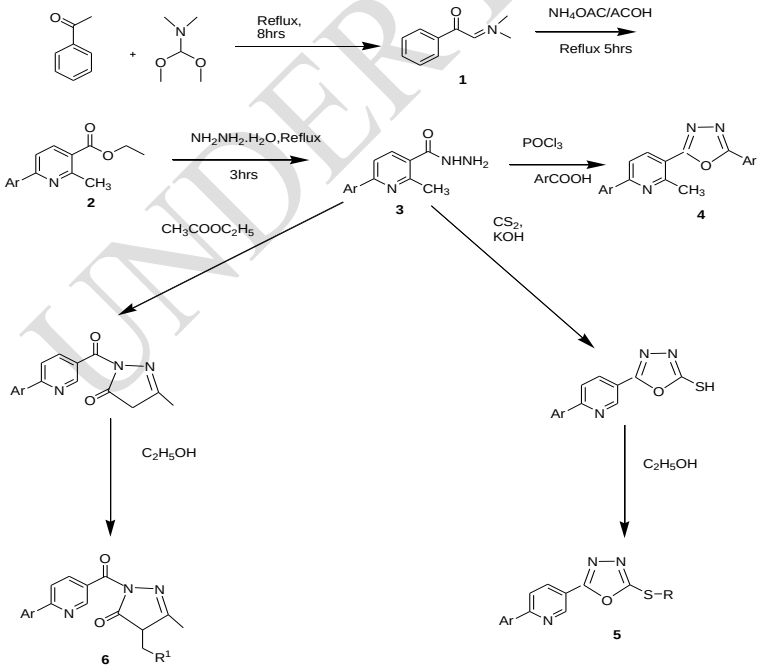
3. Synthesis of Compound 5:

Mixture was refluxed for 10-12 hours in 40 ml of 95 percent ethanol with 0.05 moles acid hydrazide, 0.05 moles (0.55 g) potassium hydroxide, and 12 ml carbon disulphide.. After cooling at room temperature, the resulting mixture was reduced. It was then acidified with dilute HCl. The solid mass was then filtered, dried, and purified using ethanol recrystallization. [17][18]

4.Synthesis of Compound 6

A water bath was used to heat a mixture of 0.01 moles acid hydrazide and 0.1 moles (12 ml) ethyl aceto acetate for 2 hours, stirring occasionally with a glass rod.A reddish syrup has been obtained, and it has been allowed to cool to room temperature. To remove coloured impurities, it was then thoroughly washed with ether. The separated solid had been filtered, dried, and purified using ethanol recrystallization. [19]

Scheme



Scheme. 1. Schematic diagram of synthesis

EVALUATION OF ANTIBACTERIAL ACTIVITY

Determination of minimum inhibitory concentration by tube dilution method [20]
Antibacterial activity as MIC is evaluated by tube dilution method. Double strength Nutrient broth was prepared and autoclaved. Prepared various concentration of stock solution using serial dilution method (100- 1.56 mcg/ml). Each 2.5ml of this solution double strength nutrient broth 2..5ml is added. Microorganism was inoculated (E. coli) in all test tubes were incubated at 37^{o}C for 48h

Determination of zone of inhibition by cup-plate method [21]
In this technique, melted agar inoculated with microorganisms is poured into Petri dishes. Wells were made in the agar plate and a specific volume of the antimicrobial substances were placed in them, plates were incubated at 37oC for the specified time. The antimicrobial substance diffuses through agar around its well and produces a clear zone of inhibition. The diameter of this zone gives an estimation of the degree of activity of the antimicrobial substance.

3. RESULTS

Spectral Values of compounds

Compound1:

1. Melting point :2450c

2.Yield :70%

3. IR Spectral values (Cm-1): (KBr): 1551, 1563(C=N)

4. Mass spectral values(m/z) (%) : 223 (25), 199 (40), 162 (54), 147 (48), 118 (100).

5.1H NMR :(DMSO-d6): δ 7.21-8.78(m, 9H, Ar-H)

Compound 2:

1.Melting Point :2400c

2.Yield:76%

3. IR Spectral Values (Cm-1):(KBr): 1123 (C-O-C), 1568, 1610 (C=N);

4.Mass Spectral values (m/z) (%) 237 (10), 227 (43), 189 (62), 147 (100).

5. 1H NMR (DMSO-d6): δ 3.79 (s, 2H, CH2), 7.05-8.76 (9H, Ar-H).

Compound 3:

1.Melting point :281oC.

2.Yield :75%

3.IR Spectral values (Cm-1): (KBr): 1146 (C-O-C), 1682, 1597 (C=N), 710 (C-Cl);

4. Mass Spectral values (m/z) (%) 259 (M+2, 15), 256 (19), 223 (31), 187 (42), 147(53), 118 (100).

5. 1H NMR (DMSO-d6): δ 6.32-8.76(m, 8H, C6H5-H);

Compound 4:

1. Melting point : 223oC.

2.Yield :73%

3 IRSpectral Values (Cm-1): (KBr): 1155 (C-O-C), 1620, 1642 (C=N), 1560 (C-NO2);

4.Mass Spectral Values (m/z)(%) 268 (18), 227 (28), 161 (63), 118 (100).

5. 1H NMR Spectral Values (DMSO-d6): δ 7.76-8.81 (m, 8H, Ar-H);

Compound 5:

1.Melting point :1700c

2.Yield :72%

3.IR spectral Values (Cm-1):(KBr): 2208 (CN), 1626 (C-C), 1238 (C–O–C), 754, 695 (SH) 2560-2600

4.Mass Spectral Values (m/z)(%): 589 [M+] (37), 509 (12.5), 470 (24.5), 259 (100), 236 (45), 145 (3.6), 117 (5.9), 77 (85).

5.1HNMR Spectral Values : (DMSO-d6): d: 5.41 (d, 1H), 5.81 (d, 1H), 7.26,7.89 (m, 13H, Ar-H), 8.00, 8.09 (2s, 2H,), 9.01 (s, 1H,).

Compound 6:

1.Melting Point :2400c

2.Yield :71%

3. IR spectral Values (Cm-1):(KBr): 2958, 2873, 2202 (CN), 1622 (C-C), 759, 648.

4.Mass Spectral Values (m/z)(%): 508 [M+] (12.5), 431 (35), 428 (12.9), 363 (17.8), 259 (45), 248 (18.9), 145 (12.6), 117 (64.6), 77 (100).

5.H NMR Spectral Values :(DMSO-d6): 7.11,7.91 (m, 14H, Ar-H and [CH=CH], 8.49, 8.79 (2s, 2H,), 9.59 (s, 1H,).

Biological Activity :

1.Determination of minimum inhibitory concentration by tube dilution method.[20]

Tube dilution method is used to assess antibacterial activity as MIC. Double the power The nutritional broth was made and autoclaved.. Prepared various concentration of stock solution using serial dilution method (100- 1.56 mcg/ml). Each 2.5ml of this solution double strength nutrient broth 2..5ml is added. Microorganism was inoculated (E. coli) in all test tubes were incubated at 37^{o}C for 48h.

Table :1 MIC of the the compounds

Compounds	Minimum Inhibitory Concentration (mcg/ml)
C1	12.5
C2	25
C3	25
C4	50
C5	50
C6	25

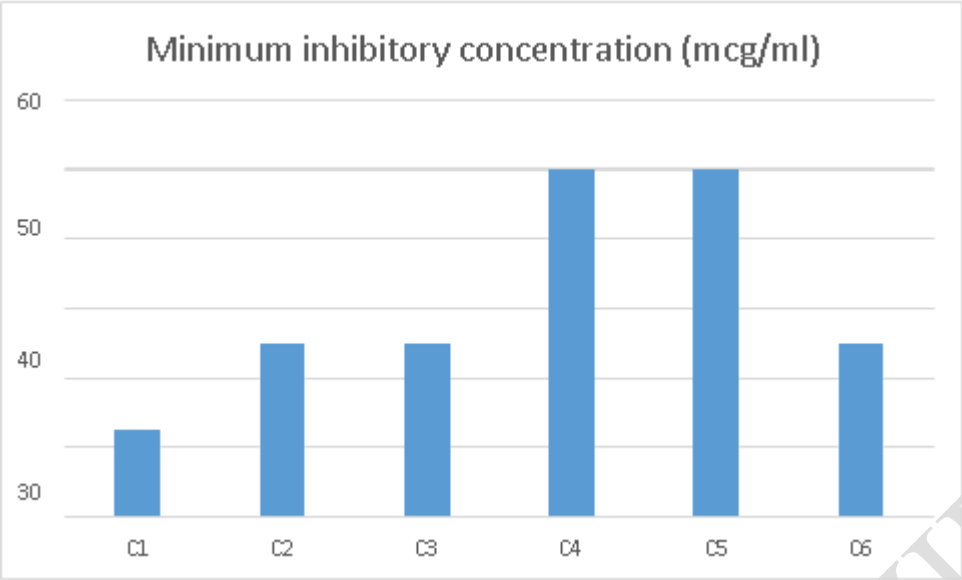
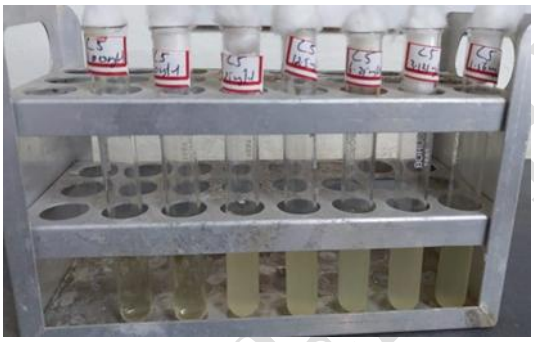


Fig 1: Minimum inhibitory concentration mcg/ml

The antimicrobial activity of the compounds was promising. The compounds' minimum inhibitory concentrations were in the range of 12.5 to 50 mcg/ml.Compound 1 exhibited maximum activity with minimum inhibitory concentration (mic) of 12.5 mcg/ml as given in table 1 and figure 1, 2, 3, ,4, 5, 6 and 7



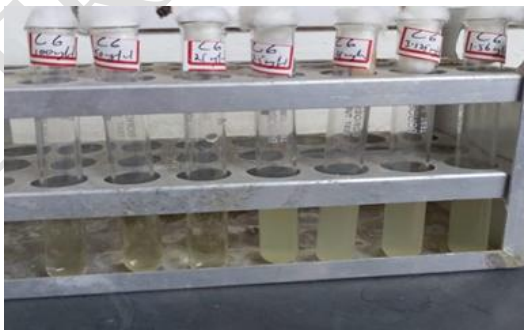
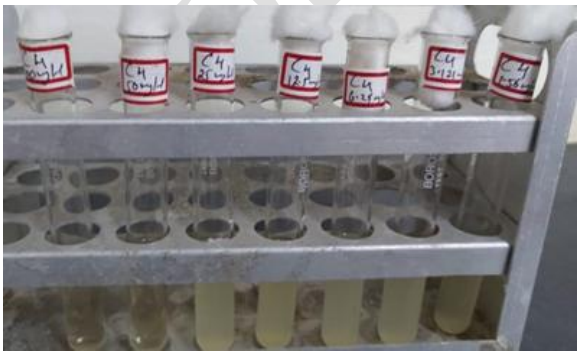


Fig 2 and 3 compound C5 and C6



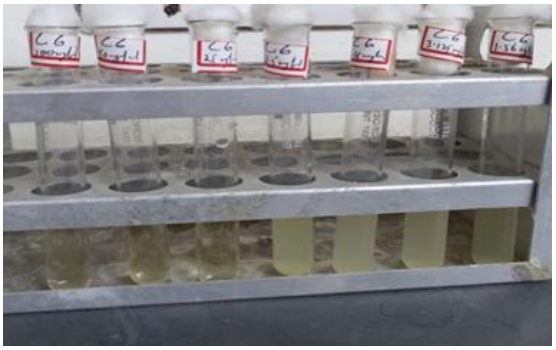
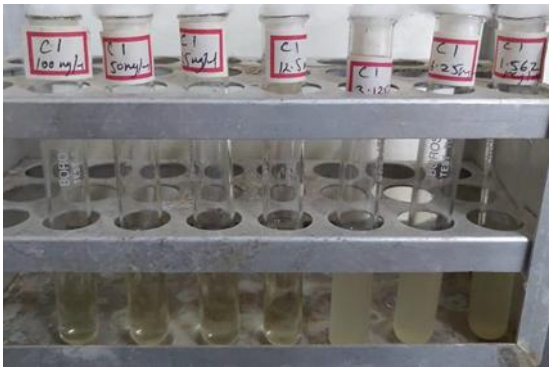


Fig 4 and 5 compounds C4 and C3



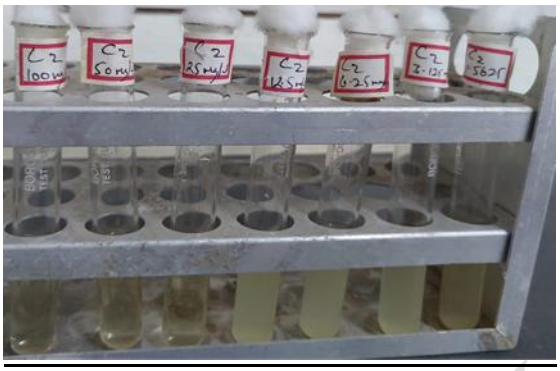


Fig6 and 7 compounds C1 and C2

3.Evaluation of antibacterial properties of the synthesized compounds were carried out against E. coli.

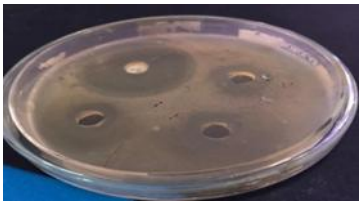


Fig8:C2

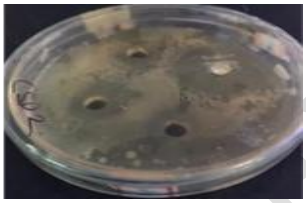


Fig9:C3

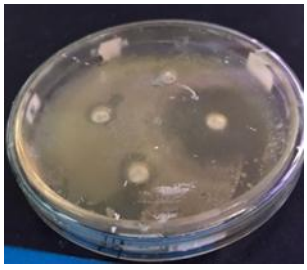


Fig 10: C4

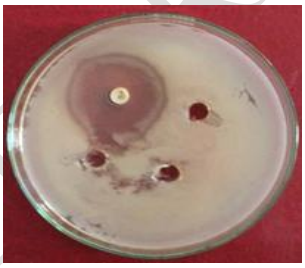


Fig11: C5

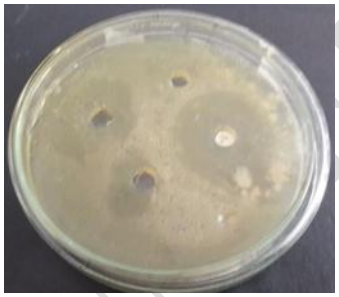


Fig12:C6

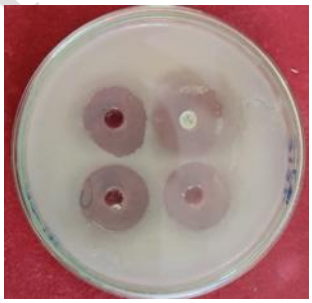


fig13: C1

Fig 8-13: Comp:1zone of inhibition.Std:Ciprofloxacin

Table: 2 **Ciprofloxacin 5mcg in different paltes**

Compounds	**Std (ciprofloxacin 5mcg)**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**

Zone of inhibition (mm)	32mm	32.01±0.034	20.56±0.106	19.28±0.12	12.01±0.135	10.75± 0.031	22.07 ±0.65

In the zone of inhibition studies, all six samples at MIC concentrations showed antimicrobial activity. The maximum activity of compound 1 was comparable to that of the standard drug ciprofloxacin (5mcg).

DISCUSSION:

Using acid hydrazide as an intermediate, a series of compounds were synthesised, including oxadiazole and pyrazole derivatives, and these compounds were screened for biological activity. Enaminones showed the highest activity, which was comparable to the standard drug ciprofloxacin (5mcg).

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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