SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF NOVEL OXAZOLE-NICOTINAMIDE DERIVATIVES

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ABSTRACT

New Nicotinamide-Oxazole derivatives have been synthesized with EDC/HOBt as coupling reagent and characterized by IR, ^{1}H-NMR, ^{13}C-NMR and Mass spectra. *In vitro* antibacterial and antifungal studies were evaluated in zone of inhibition with new Nicotinamide-oxazole derivatives.

Keywords: Nicotinamide, Oxazole, antimicrobial activity, amide bond

1. Introduction

Since ancient times, huge amounts of increasing events of microbial infections has been observed and the frequent use of antibacterial, antifungal and cytotoxic drugs. Microbial species found resistance towards various existing antimicrobial agents. Hence, it was significant to synthesis new antimicrobial agents. In present work, amide linkage plays an important role as linker in between Pyridine and Oxazole side chain. The amide linkage has a biological significance especially towards antimicrobial area. The amides plays a key role for medicinal chemists[1,2] and its functionality is a common feature in small or complex synthetic or natural molecules[3,4]. An comprehensive analysis of Medicinal Chemistry database[5,6] revealed that the carboxamide group appears in more than 25% of known drugs and carboxamides[7] are neutral, stable and have both hydrogen-bond accepting and donating properties.

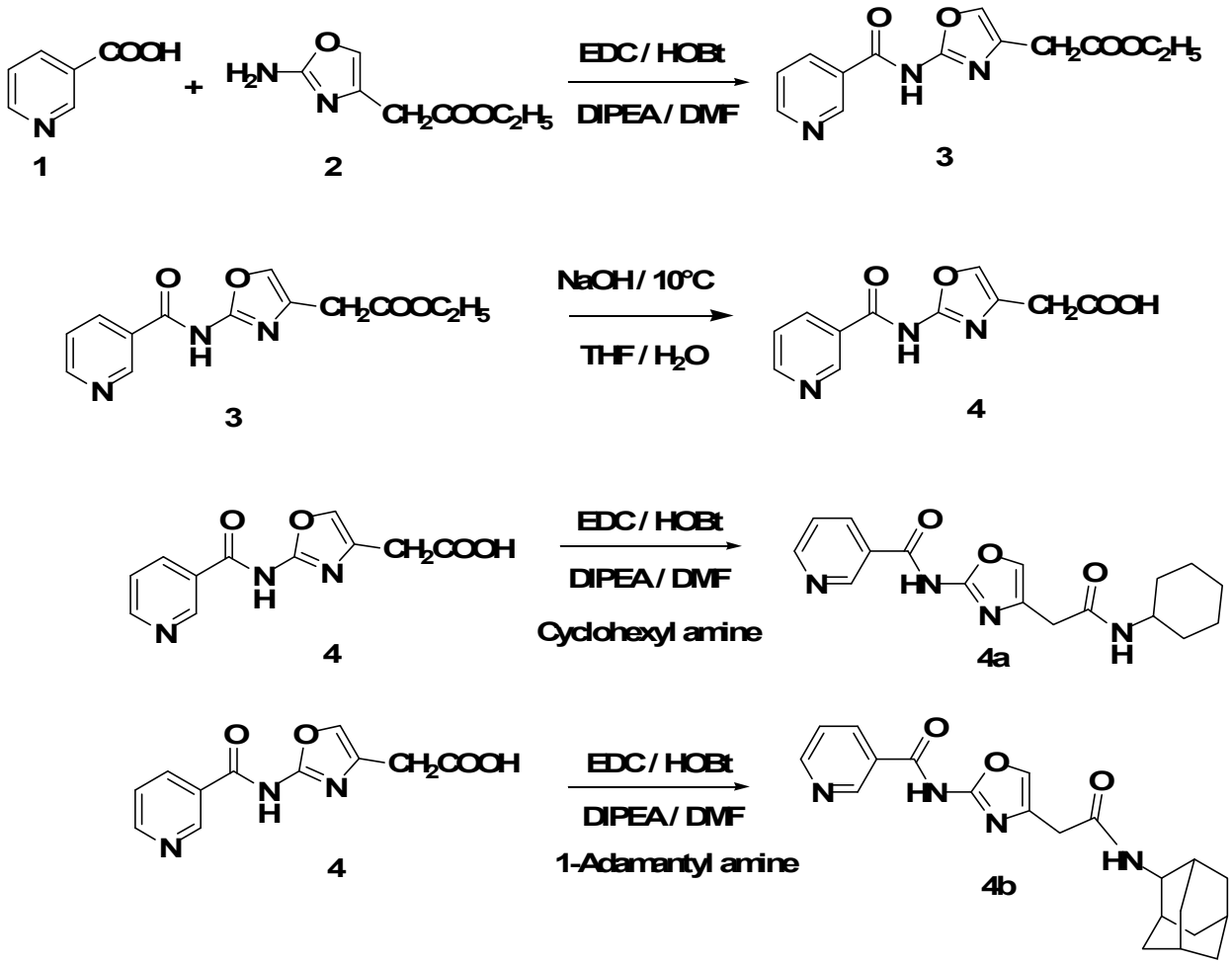
2. Materials and methods

The solvents and reagents used for the synthesis (AR Grade) were purified by the standard methods. DMF was dried over Type 3A molecular sieve for 72 hours, followed by distillation under reduced pressure. The chemicals and reagents were purchased from Sigma-Aldrich.

^{1}H NMR spectra were recorded on BRUCKER 400 MHZ Instrument (CDCl3 and DMSO-d^{6} used as solvent). TMS used as an internal standard. Chemical shifts are given in ppm. Infrared spectra were recorded on Brucker IFS 66v FT-IR SPECTROMETER using KBr pellets (ν in cm^{-1}).

3. Results and discussion

Reaction scheme:



Chemistry -Experimental section:

Ethyl {2-[(pyridine-3-ylcarbonyl)amino]-1,3-oxazol-4-yl}acetate (3)

To a stirred solution of pyridine-3-carboxylic acid (**1**) (5g, 0.0406 mol) in dry DMF (50 ml), EDC (1-**E**thyl-3-(3'-**D**imethylamino)**C**arbodiimide hydrochloride salt, 15.5g, 0.0812mol), HOBt (**H**ydr**O**xy**B**enzo**t**riazole) (5.48g, 0.0406 mol) were added and stirred under nitrogen atmosphere at 0°C for 30 minutes. Then ethyl-(2-amino-1,3-oxazol-4-yl)acetate (**2**) (9g, 0.0487mol), DIPEA (20ml, 0.122mol) were added. The mixture was stirred for about 4 hours at room temperature. The reaction was monitored by TLC. The reaction mixture was poured into ice water. The brown solid precipitated was filtered under vacuum. YIELD: 10 gm (85%). The mass spectrum of the ethyl{2-[(pyridine-3-ylcarbonyl)amino]-1,3-oxazol-4-yl}acetate (**3**) shows the peak corresponding to molecular ion (m/e=291). ^{1}H – NMR SPECTRA: (400MHZ, CDCl3) The NMR spectrum shows the chemical shifts corresponding to: δ 10.7(1H,b,Amide N-H), δ 9.92(1H,d, Ar-H), δ 8.82(1H,q, Ar-H), δ 8.28(1H,m, Ar-H), δ 7.48(1H,q, Ar-H), δ 6.87(1H,s, Ar-H), δ 4.16(2H,q, Ar-CH_2), δ 3.62(2H,s, ethyl ester-CH_2), δ 1.24(3H,m, ethyl ester-CH_3).

{2-[(pyridine-3-ylcarbonyl)amino]-1,3-oxazol-4-yl}acetic acid (4)

To a stirred solution of compound **3** (10 g, 0.034 mol) in THF (100ml), sodium hydroxide (2.72g, 25 ml of water) was added at 10°C. After the addition, the reaction mixture was stirred for 2 hours at room temperature. The reaction was monitored by TLC. THF was evaporated under high vacuum. The reaction mass was cooled to about 5-10°C and neutralized to pH 7 using 10 % HCl. The white precipitate formed was filtered and dried.

YIELD: 7 gm (78%). The mass spectrum of the {2-[(pyridine-3-ylcarbonyl)amino]-1,3-oxazol-4-yl}acetic acid (**4**) shows the peak corresponding to molecular ion (m/e=263). ^{1}H – NMR SPECTRA: (400MHZ, DMSO-D_6) The NMR spectrum shows the chemical shifts corresponding to: δ 12.8 (1H,b,Acid-H), δ 9.21(1H,d, Ar-H), δ 8.79(1H,q, Ar-H), δ 8.43(1H,m, Ar-H), δ 7.58(1H,q, Ar-H), δ 7.0(1H,s, Ar-H), δ 3.62(2H,s, Ar-CH_2).

N-{4-[2-(cyclohexylamino)-2-oxoethyl]-1,3-oxazol-2-yl}pyridine-3-carboxamide (5)

To a stirred solution of compound **4** (2g, 0.0076 mol) in dry DMF (20 ml), EDC (2.9g, 0.015 mol), HOBt (1g, 0.0076 mol) were added and stirred under nitrogen atmosphere at 0°C for 30 minutes. Then cyclohexylamine (0.9g, 0.0091 mol) was added and followed by DIPEA (3.7 ml, 0.023 mol) and stirring was continued for about 6 hours at room temperature. The reaction progress was monitored by TLC. The reaction mixture was poured into ice water. The brown precipitate obtained was filtered and dried over high vacuum. YIELD: 2 gm (77%). Mass spectrum: The mass spectrum of the N-{4-[2-(cyclohexylamino)-2-oxoethyl]-1,3-oxazol-2-yl}pyridine-3-carboxamide (**5**) shows the peak corresponding to molecular ion (m/e=344). ^{1}H–NMR SPECTRA : (400MHZ, DMSO-D_6) The NMR spectrum of compound **5** shows the chemical shifts corresponding to: δ 12.86(1H,s,Amide N-H), δ 9.2(1H,d, Ar-H), δ 8.77(1H,q, Ar-H), δ 8.41(1H,d, Ar-H), δ 7.57(1H,q, Ar-H), δ 7.49(1H,q, Ar-H), δ 6.94(1H,s, Ar-H), δ 3.51(3H,d),δ 1.73(2H,t),δ 1.66(3H,t),δ 1.64(1H,m),δ 1.25(2H,d). HPLC PURITY: - 99.32% at R.T 9.294. IR SPECTRA: In the IR spectra of compound **5**, amide N-H stretching frequency shows the strong band at 3292 cm^{-1}. Pyridine C-H strectching showed 3042 cm^{-1} ; amide C=O stretching shows at 1639.80 cm^{-1}. Amide N-H bending corresponds to 1554.44 cm^{-1}.

N-{4-[2-(adamantylamino)-2-oxoethyl]-1,3-oxazol-2-yl}pyridine-3-carboxamide (6)

To a stirred solution of compound **4** (2g, 0.0076 mol) in dry DMF (20ml), EDCI (2.9g,0.015 mol), HOBt (1g, 0.0076 mol) were added and stirred under nitrogen atmosphere at 0°C for 30 minutes. Then 1-adamantylamine (1.37g, 0.0091 mol) was added and followed by DIPEA (3.7 ml, 0.023 mol) and stirring was continued for about 6 hours at room temperature. The reaction progress was monitored by TLC. The reaction mixture was poured into ice water. The brown precipitate formed was filtered and dried over high vacuum. YIELD: 2.2 gm (67%). Mass spectrum: The mass spectrum of compound **6** shows the peak corresponding to molecular ion (m/e=396). ^{1}H – NMR SPECTRA: (400MHZ, DMSO-D_6) The NMR spectrum of compound **6** shows the chemical shifts corresponding to : δ 12.86(1H,s,Amide N-H), δ 9.2(1H,s, Ar-H), δ 8.78(1H,d, Ar-H), δ 8.41(1H,d, Ar-H), δ 7.58(1H,q, Ar-H), δ 7.49(1H,s, Ar-H), δ 6.94(1H,s, Ar-H), δ 3.47(2H,s),δ 2.0(3H,s),δ 1.93(6H,s),δ 1.61(6H,s). HPLC PURITY:- 97.61% at R.T 9.294. In the IR spectrum of compound **6**, amide N-H frequency stretching shows the strong band at 3323.84 cm^{-1}. Pyridine C-H strectching showed 3025 cm^{-1}; amide C=O stretching shows at 1635 cm^{-1}. Amide N-H bending corresponds to 1544 cm^{-1}.

Antimicrobial Activity

In the present study, the paper disc diffusion method was used to evaluate the *invitro* anti-microbial activity of New nicotinamide-Oxazole compounds. Nutrients agar medium and dextrose agar medium (Hi-Media Laboratories, India) were the media used for the study of anti-bacterial and anti-fungal respectively. The standard strains were procured from American Type Culture Collection (ATCC), Rockville, USA, and the pathological strains were procured from the Department of Microbiology, CEEAL ANALYTICAL LAB, Chennai. The anti-microbial activity of the synthesized compounds was screened against the following bacteria and fungi. Gram-positive organisms like 1. Staphylococcus aureus (ATCC 9144), 2. Staphylococcus epidermidis (ATCC 155), Gram-negative organisms like 1. Escherichia coli (ATCC 25922), 2. Klebsiella pneumonia (ATCC 11298), Fungi like Candida albicans (ATCC 9029) were Bio-assayed. The results were shown here.

ORGANISM	STD	3	4	4a	4b
STAPHYLOCOCCUS AUREUS (ATCC 9144)	*32 mm*	*17 mm*	*19 mm*	*22 mm*	*20 mm*
STAPHYLOCOCCUS EPIDERMIDIS (ATCC 155)	*29 mm*	*15 mm*	*17 mm*	*20 mm*	*22 mm*
ESCHERICHIA COLI (ATCC 25922)	*31 mm*	*14 mm*	*17 mm*	*22 mm*	*23 mm*
KLEBSIELLA PNEUMONIAE (ATCC 11298)	*28 mm*	*13 mm*	*15 mm*	*18 mm*	*19 mm*
CANDIDA ALBICANS (ATCC 9029)	*29 mm*	*17 mm*	*19 mm*	*25 mm*	*23 mm*

3. Conclusion

In recent study of four compounds of Nicotinamide-Oxazole derivatives was subjected to antibacterial and antifungal bio-assay. All four compounds were bio-active and it was concluded that all the prepared compounds were found to possess the significant activity with low toxicity. Further synthesis of new nicotinamide-oxazole derivatives and biological studies can be done as the present results encouraged to the limit where it can be future new antimicrobial agents.

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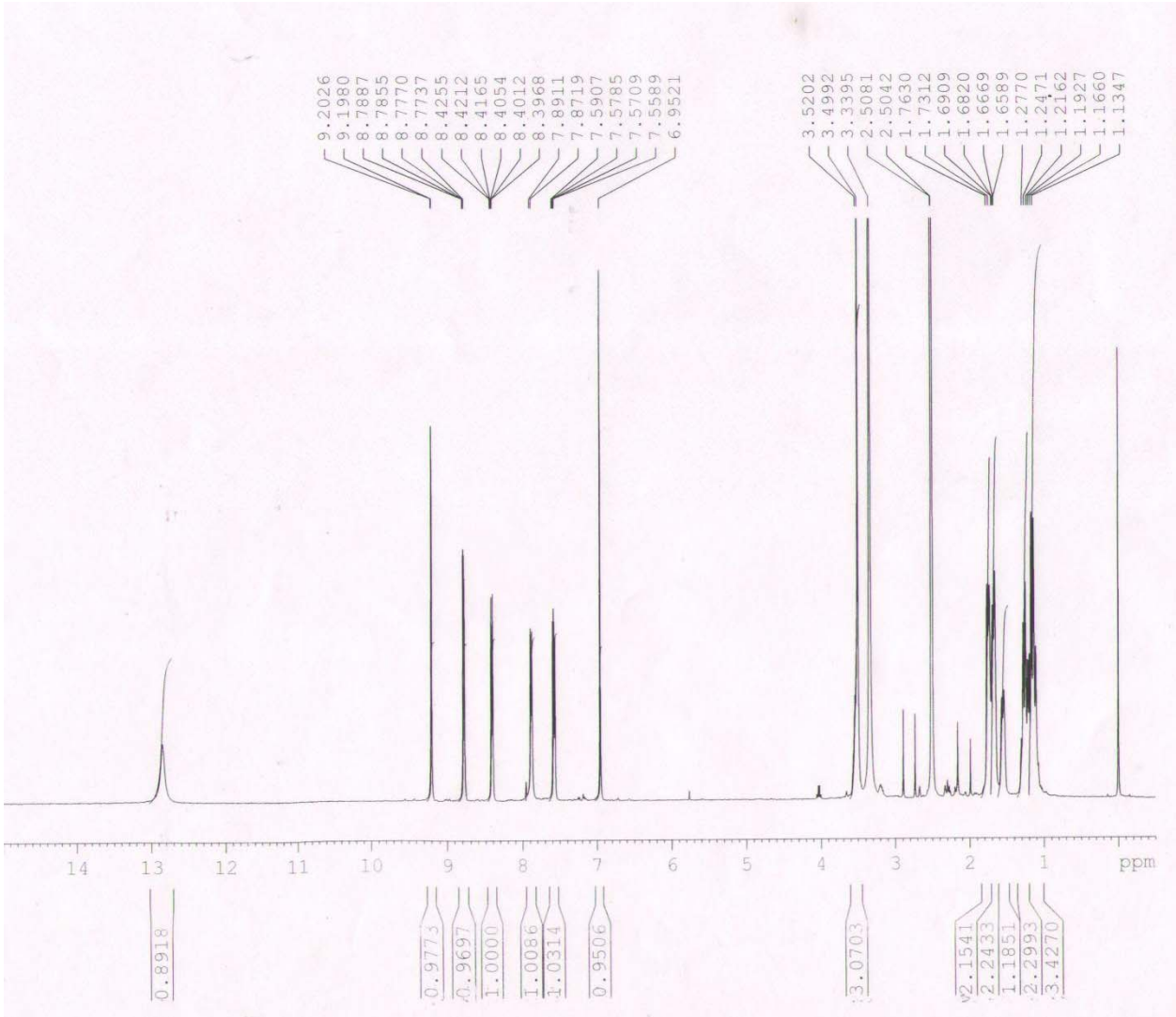

Fig (1) shows Proton NMR spectrum of Compound **5**

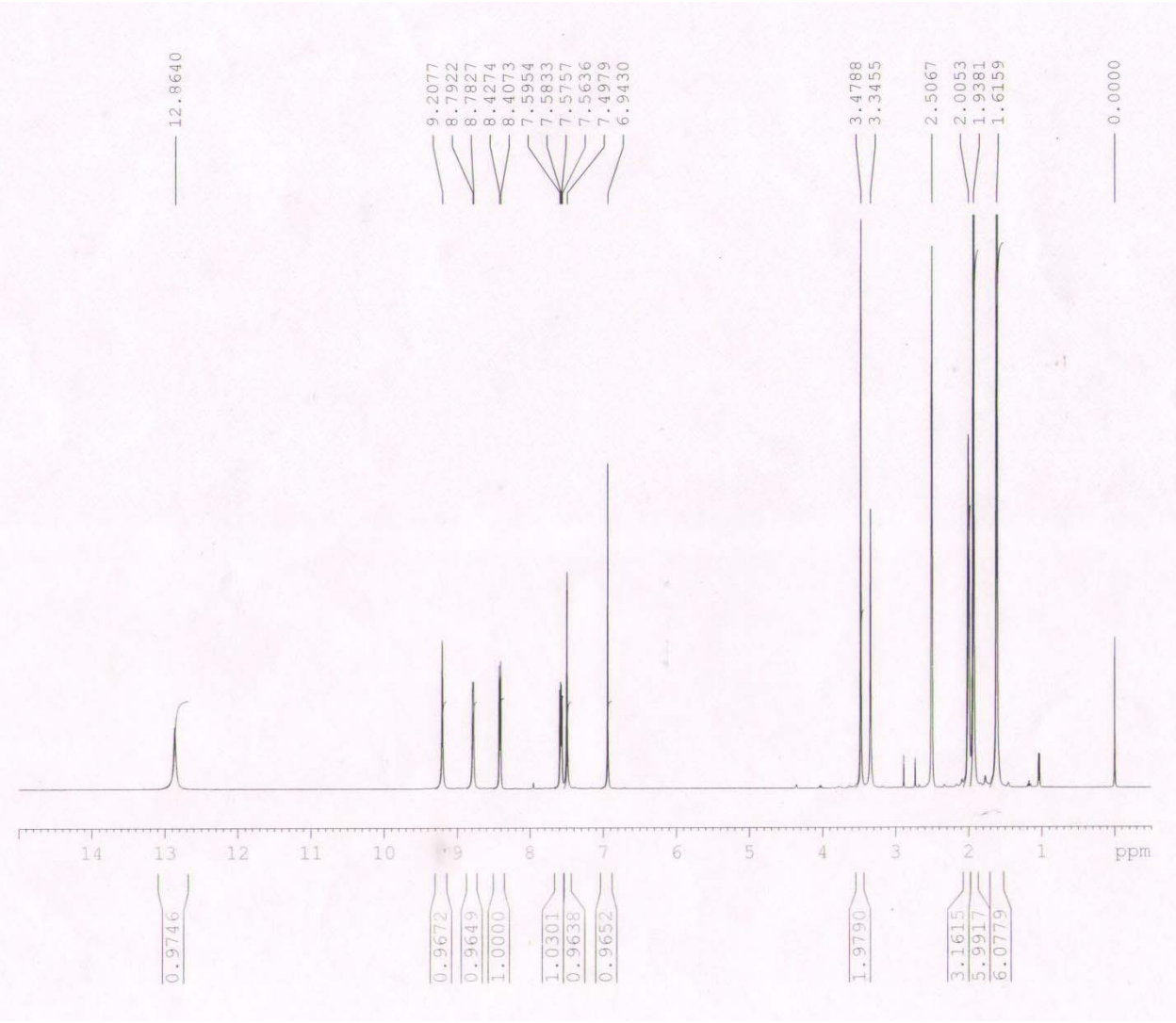

Fig (2) shows Proton NMR spectrum of Compound **6**