Study of antimicrobial activity of new prepared seven membered rings (Oxazepine)

Ahmed Sa'adi Hassan[1] and Ahmed Solaiman Hame[2]*
1. Medical Lab Technique, College of Health and Medical Technical, Middle Technical University, Baghdad, IRAQ
2. Department of Applied Chemistry, College of Applied Science, University of Fallujah, Fallujah, IRAQ
*dr.ahmedalmohammedi@uofallujah.edu.iq

Abstract

In this work, ten new heterocyclic compounds bearing biologically active components were synthesized. α-Chloro-N, imidazole acetamide was synthesized and then used in the synthesis of target 7-membered heterocyclic ring systems (Oxazepine). Two nitrogen containing hetero cycles namely Imidazole were selected as biologically active components to be used in this work beside Oxazepine antibiotics.

The new heterocyclic compounds were synthesized by four steps: first one chloroacetylchloride was introduced in reaction with imidazole producing the corresponding α-Chloro-N,imidazole acetamide; second step via reaction with hydrazine hydrate and α-Chloro-N,imidazole acetamide under reflux conditions producing the target α-hydrazino-N, imidazole acetamide; in third step, mixture was refluxed consisting of ketones and hydrazino-N imidazole acetamide obtaining the imines compound and the last step is of reacted products of step three with different anhydrides (maleic, citraconic and dimathylmalic). Results of antimicrobial activity evaluation of the newly synthesized new heterocyclic compounds (Oxazepine) showed that the new Oxazepine exhibits very high antimicrobial activity against the tested bacteria and fungi.

Keywords: Seven membered rings, Oxazepine, antimicrobial activity, imidazole.

Introduction

Chemistry of 7-membered heterocyclic ring systems in recent years gained considerable attention due to the wide range of biological activities of these systems.[1] For the same reason, the natural products containing the seven rings brought the attention of the researchers and the focus was on the heterocyclic rings containing N, O, and S as hetero atoms and the fused rings system containing the benzo derivatives.[2] The chemistry of the bonds between carbon and nitrogen were essential in chemical sciences. This is due to the lon pair of isomethane group (>C=N) which is called Schiff bases relative to Schiff who prepared it for the first time.[3, 4]

Oxazepine derived was presented in this time for use in mental ease characterized by anxiety and stress.[5] Oxazepine is an unsaturated, seven membered containing heteroatoms and oxygen in the position 1 and nitrogen in position 3 in addition to five carbon atoms. It is synthesized by the pericyclic cycloaddition of Schiff bases with anhydrides.[6] Oxazepine and derivatives have medical and biological importance and they have medicinal and pharmaceutical application.[7] Imidazole 1, a histidine 2 component, is five membered heterocyclic compound containing nitrogen atoms in 1 and 3 positions. It has fascinating biological activities such as antibacterial, anticancer, antiviral and anti-depressant.[6, 7]

Bisimidazole is an important moiety in therapeutic. The divers interesting reviews of imidazoles and azolines[8- 10] either focus on synthetic methods for obtaining these compounds or their biological activity, or both of these topics but with greater emphasis given to one of them[11-13]. In the current review, we focus on a combination of azoline and imidazoles, which is directly involved in the preparation of precursor drugs and potential drug synthesis. According to all these facts we planned in this work to synthesize new compounds by combination of drug or heterocyclic molecules and oxazepine in a single molecule, thus the resulted new compounds having structural features of both Oxazepine and drug heterocycle imidazole would provide new derivatives possessing potent pharmacological activities.[14]

Material and Methods

Uncorrected melting points were recorded on Gallenkamp melting point apparatus. SHIMADZN FTIR-8400 Fourier Transform Infrared spectrophotometer was used for recording FTIR spectra of the prepared compounds. Bruker ultrasheild 300 MHz apparatus was used for recording 1H-NMR and 13C-NMR spectra using DMSO-d6 as solvent and TMS as internal standard. Hetashi model incubator was used for antimicrobial activity evaluation.

Synthesis of α-Chloro-N, imidazole acetamide (1): This compound was prepared using a modified procedure in leterature.[15] Chloroacetyl chloride (1.13 g, 0.01mol) in dry benzene (10ml) containing triethylamine (1ml) solution was added with stirring to a solution of 1, 3-imidazole (1.7g, 0.01 mol) in dry benzene (20ml). The resulting mixture was refluxed for 12 hours on a water bath. The solvent was distilled and the residue was washed with 5% sodium bicarbonate to remove the acid impurities, the residue was crystallized from suitable solvent m.p (90–92°C), yield 87%.

Synthesis of α-hydrazino-N, imidazole acetamide (2): To a solution of (1) (2.45g, 0.01mol) in absolute ethanol (25ml),

hydrazine hydrate (0.01 mol, 0.5 ml) was added with stirring and the final mixture was refluxed on a water bath. After cooling, crystal of yellow precipitate was formed. The crystals were filtered and recrystallized from suitable solvent m.p (131-133°C), yield 92 %.[16]

Synthesis of α-(2-Arylidine-hydrazino), N- imidazole acetamide (3,4): To a solution of compound (2) (1.23 g, 0.005mol) in absolute ethanol (25ml), appropriate aldehyde or ketone (0.005mol) was added. The resulting mixture was refluxed for 5-6 hours. After cooling, the precipitate was filtered, washed with ethanol and recrystallized from appropriate solvent.[6]

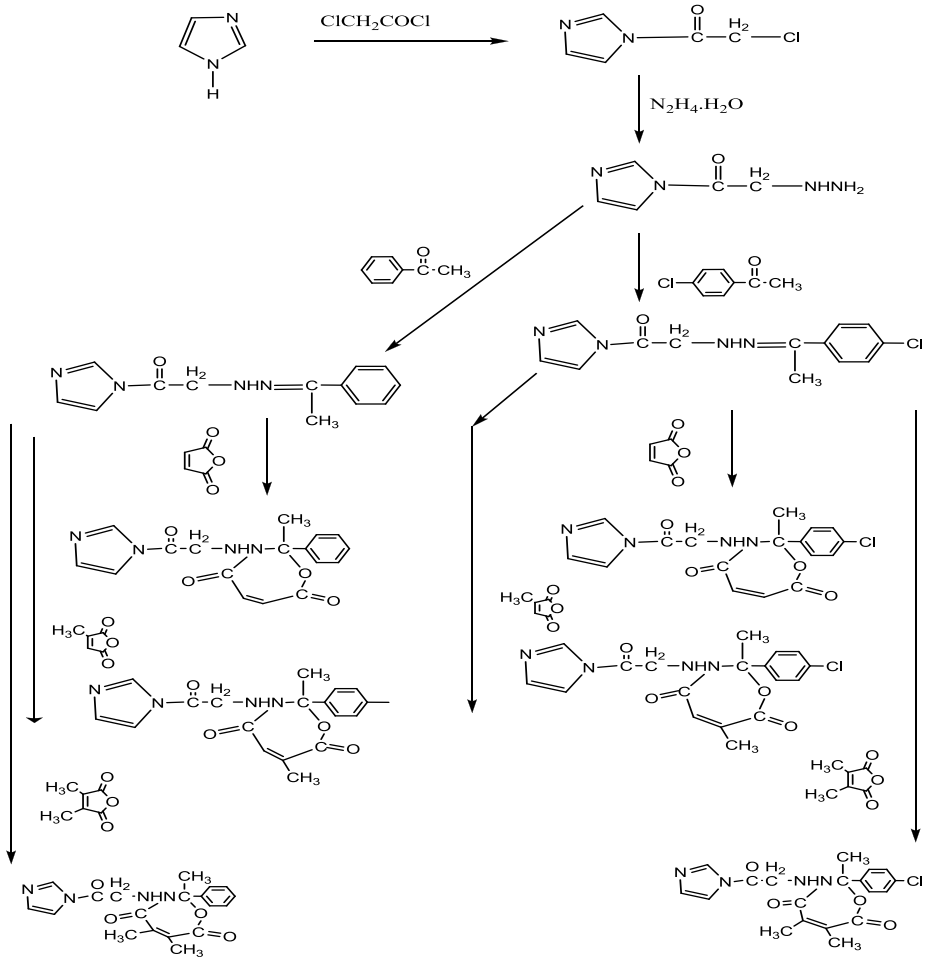
Synthesis of α [4,7-dioxa-2-aryl-1,3-oxazepine-3-yl amino] N- imidazole acetamide (5-10): It was prepared by using 0.01mol, 0.98g of maleic, citraconic and dimethyl malic anhydride and 0.01 mol of Schiff bases in abs. EtOH with stirring. Physical properties of synthesized oxazepine [5-10] are shown in table 2.[17]

Biological activity study: In a suitable conical flask, Mulerhonton was added to one liter of distilled water with stirring and heating until it was completely dissolved. The conical flask was sealed with cotton and sterilized at 121°C for 20 minutes under pressure of 15 bound/ inch, then cooled to 45-55°C, placed in 20 mL Petri dishes and left to cool and solidified. The studied organisms were placed on the agar surface, then by using a sterilized cork borer cups were scooped out of agar medium contained in a Petri dish and the solution of tested compound (0.1mL) was added in the cups and the Petri dishes were subsequently incubated at37°C for 48 hrs.[5-7] Ampicillin and fluconazole were used as reference and DMF as a negative control[7]. Inhibition zones caused by the various prepared compounds were determined and listed in table 1.

Results and Discussion

In this work we planned to synthesize new organic compounds containing two known pharmacologically active components namely oxazepine and drug molecules containing heterocycles nitrogen like imidazole. Thus, the newly synthesized compounds having structural features of both the mentioned were estimated. Combination of oxazepine with drug molecule like imidazole active heterocycles molecule would produce novel compounds having potent pharmacological activities.

Performing this target was made by many steps which are summarized in scheme 1.



Scheme 1

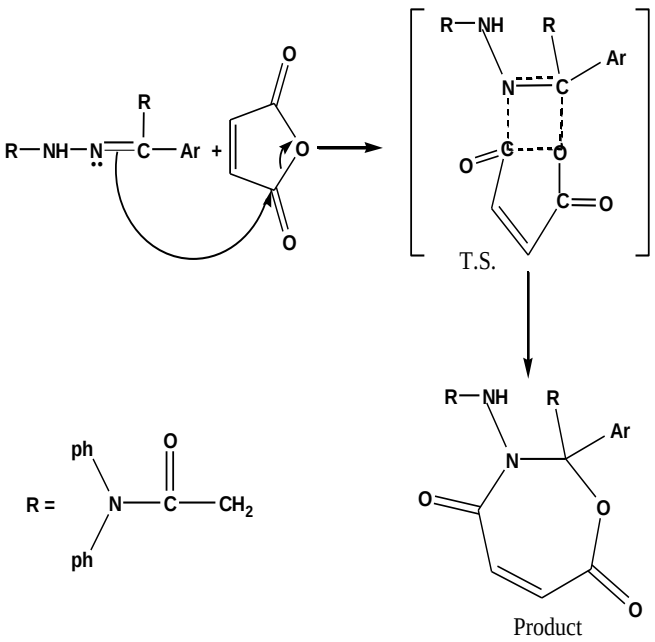
Synthesis of the target compounds was based on the reaction of chloroacetylchloride with imidazole moiety. In the first step compound [1] was prepared according to literature procedure[15] and then in the second step, compound [1] was introduced in reaction with hydrazine hydrate to produce compound [2]. The (hydrazino), N- imidazole acetamide reacted with ketones producing imines compounds [3,4] reacting with anhydride leading to ring closer producing oxazepine compounds Physical properties of new compounds are shown in table 1.

FTIR spectrum of α -Chloro –N-imidazole acetamide (1) showed the disappearance of (N-H) band of secondary amine at 3250 cm^{-1} and appearance of a strong (C=O) stretching band at 1697.24 cm^{-1}; bands of (C-H) aliphatic appeared at 2954.7 and 2858.3 cm^{-1}, bands at 3035.7 and 1519.2 cm^{-1} are due to C-H and C=C stretching of aromatic rings respectively. Also, bands at 671.18 cm^{-1} represent (C-Cl) stretching. Hydrazine (2) was found a suitable route for this synthetic approach.

FTIR spectrum confirmed the formation of compound (2) from the appearance of bands at 3285.4 and 3116.7 cm^{-1} which were assigned to the asymmetric and symmetric stretching bands of NH_2 and NH groups. In addition to the disappearance of C-Cl band at 671.18 cm^{-1} of compound (1), C=O B and appeared at 1670.2 cm^{-1} and 3053.1 and 1596.9 cm^{-1} were due to aromatic C-H and C=C stretching respectively and at 1427.23 cm^{-1} for δ(NH) bending.

The oxazepine derivatives [5-10] were obtained by refluxing equimolar amounts of the imines (3-4) and anhydride maleic, citraconic and dimathylmalic in dry benzene as in scheme 1. The synthesized compounds were characterized by their m.p and FTIR spectra listed in tables.

The suggested mechanism for the formation of the oxazepine derivatives is as follows:



FTIR spectra showed the appearance of (C=O) stretching band at 1704 cm^{-1} due to oxazepin dione ring (5) which is the most characteristic evidence for the success of the cyclization step. The FTIR Spectra are listed at table 2.

Finally, ^{1}H-NMR spectrum of compound [6] showed many signals including signal at (δ=1.3) ppm belonging to (N-H) amine proton which was caused by tautomerism with (N-H) amide as shown in this equation.

Other signals appeared at (δ=4.2) ppm due to (CH_3) protons and at δ=7.27-7.69 ppm belonging to (C-H) amide proton and aromatic protons.[13] C-NMR spectrum of the same compound [6] showed signals at 61.59 ppm belong to (CH_3) group, signals at (95.35-133.3) ppm due to aromatic ring carbons, signals at 157.8 and 164.33 ppm due to two carbon atoms in oxadiazole ring and signals at 168 and 169.1 ppm belonging to two carbonyl carbons. The ^{1}H-NMR spectrum of compound [10] showed signals at (δ=7.6-8) ppm belonging to aromatic protons.

^{13}C-NMR spectrum of the same compound [10] showed many signals including signals at 125.80-136.63 ppm belong to aromatic ring carbons, signal at 163.68 ppm due to two carbon atoms in oxadiazole ring and signal at 169.10 ppm due to two carbonyl carbons in amide ring.[18]

Biological Activity: Cup plate method using mulorhenton agar medium was employed in studying the antimicrobial activity of the prepared imides against four strains of bacteria and *Candida albicans* fungi. DMSO was used solvent and the concentration for all tested compounds was 100µg/mL. Inhibition zone was measured in mm and listed in table 3.

The results showed that the new compound [1] showed slight activity for gram positive bacteria. Compounds [3], [5], [6], [7] and [10] are highly active against *Staphylococcus aurous*, compounds [5] and [10] are highly active against *Streptococcuspyogenes*, compounds [8], [9] are highly active against *klebsiella pneumoniae* and compounds [8], [10] are highly active against *E.coli*.

The prepared compounds [3], [8] and [9] showed high activity against *Staphylococcus* aurous. *Streptococcuspyogenes*, compounds [5] and [9] showed high activity against *E. coli* and compounds [6], [7] and [8] showed high activity against *Streptococcuspyogenes* and *klebsiell pneumoniae.*

Compounds [3] and [10] showed high activity against *Candida albicans* fungi while the rest imides showed moderate activity against this fungus. On comparison, the obtained results of the prepared compounds with the results of the standard drugs it was noticed that incorporation of adduct moiety in drug molecule caused enhancement and increase in their antibacterial and antifungal activities.

Table 1
Physical properties and structure of comp. (1-10)

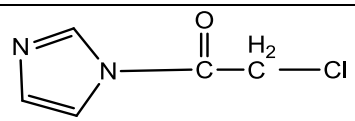
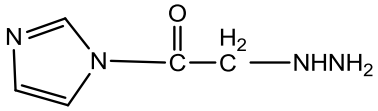
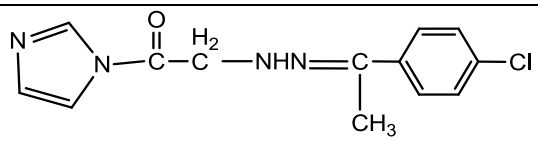
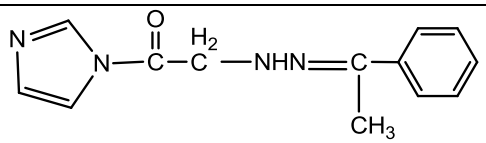
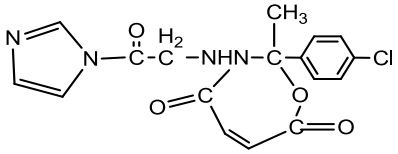
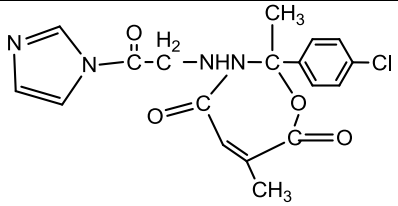
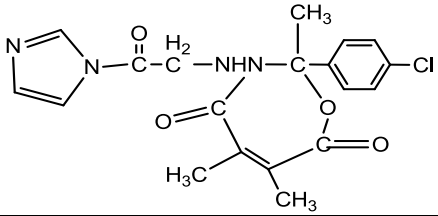
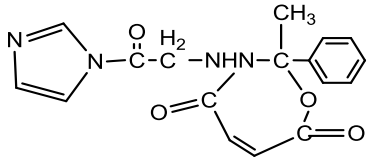
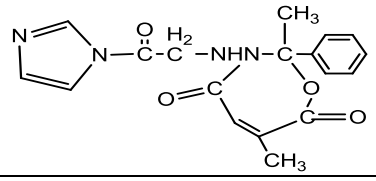
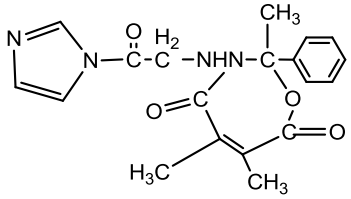
Comp. No.	Compound structure	Color	Melting Points °C	Yield %	Recrystallization Solvent
1		Dark gray	90-92	87	Ethanol
2		orange	131-133	92	Ethanol
3		Faint gray	202-204	93	Dioxane
4-		white	194-196	75	Dioxane
5		Dark orange	202-204	65	Ethanol
6		Faint Yellow	186-188	70	Acetone
7		Yellow	(92-94)	68	metanol
8		Yellow	120-122	76	Ethanol
9		orange	166-168	84	Acetone
10		Yellow	138-140	80	Ethanol

Table 2
FTIR Spectral Data $cm^{-1}$of the prepared compounds

Comp. No.	ν(N-H)	ν(C-H) aromatic aliphatic	ν(C=O) amide	ν(C=N)	ν(C=C) Aromatic	ν(C-Cl)	Others
1-	-	3035 2954	1697.2	1590	1519.2	671.18	dissapN-H 3250
2-	3285.4	3053 2875	1670	1607	1596.9	-	dissapC-Cl 671.18
3	3320 3280	3055 2835	1665	1610	1541	650	-
4	3253 3301	3070 2980	1645	1605	1530	-	-
5	3263.3 3109.04	3055 2993.3	1704	1658.6	1581.5	660	-
6	3260.3 3100. 4	2991.3 2931.5	1708.9	1657.9	1561.5	666.6	-
7	3244.7 3118.6	3021 2932.8	1679.8	1633.8	1525.4	668	-
8	3284.2 3131.6	3025.1 2962.2	1669.2	1643.4	1565.2		
9	3274.2 3148.2	3030.2 2942.5	1689.2	1653.3	1545.1		
10	3204.5 3109.6	3035.1 2972.1	1659.2	1653.2	1565.3		

Table 3
Antibacterial and antifungal activity of compounds [7-10]

Comp. No.	Gram-positive bacteria		Gram-negative bacteria		Fungi
	Staphylococcus aureus	***Streptococcus pyogenes***	***klebsiella pneumoniae***	***Escherichia coli***	***Candida albicans***
1-	+|	+	-	-	-
2-	+++	++	+	+	-
3-	++++	++	+	++	+++
4-	++	+	+	-	-
5-	++++	++++	++	+++	+
6-	++++	+++	++	++	++
7-	++++	+++	++	++	+
8-	+++	+++	++++	++++	++
9-	+++	++	++++	+++	++
10-	++++	++++	++++	++++	+++
Ampicillin	+++	++	++	++	-
Fluconazole	-	-	-	-	+++

Key of symbols: slightly active = + = inhibition zone 6-9 mm
Moderately active = ++ = inhibition zone 9-12 mm
High active = +++ = inhibition zone 13-17 mm
Highly active = ++++ = inhibition zone > -17 mm

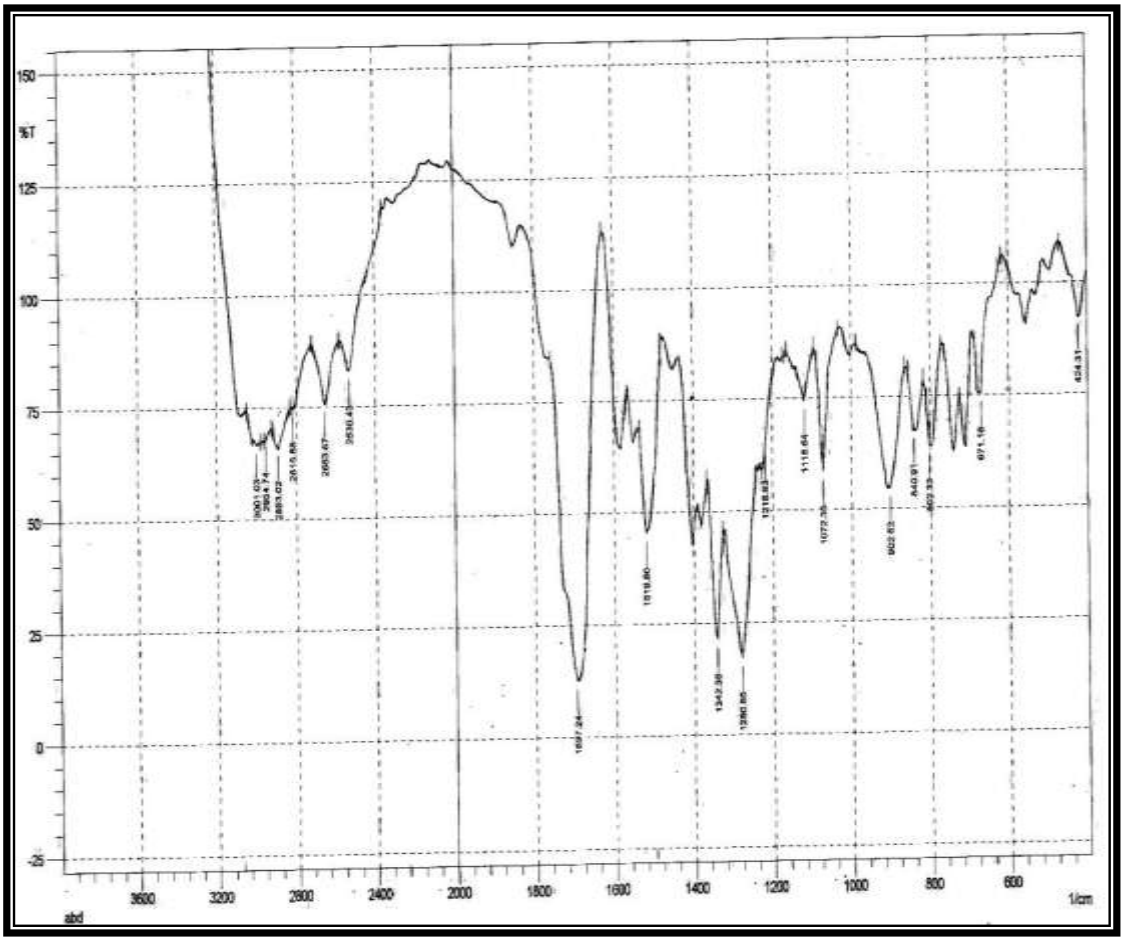


Fig. 1: FT-IR spectrum for compound [1]

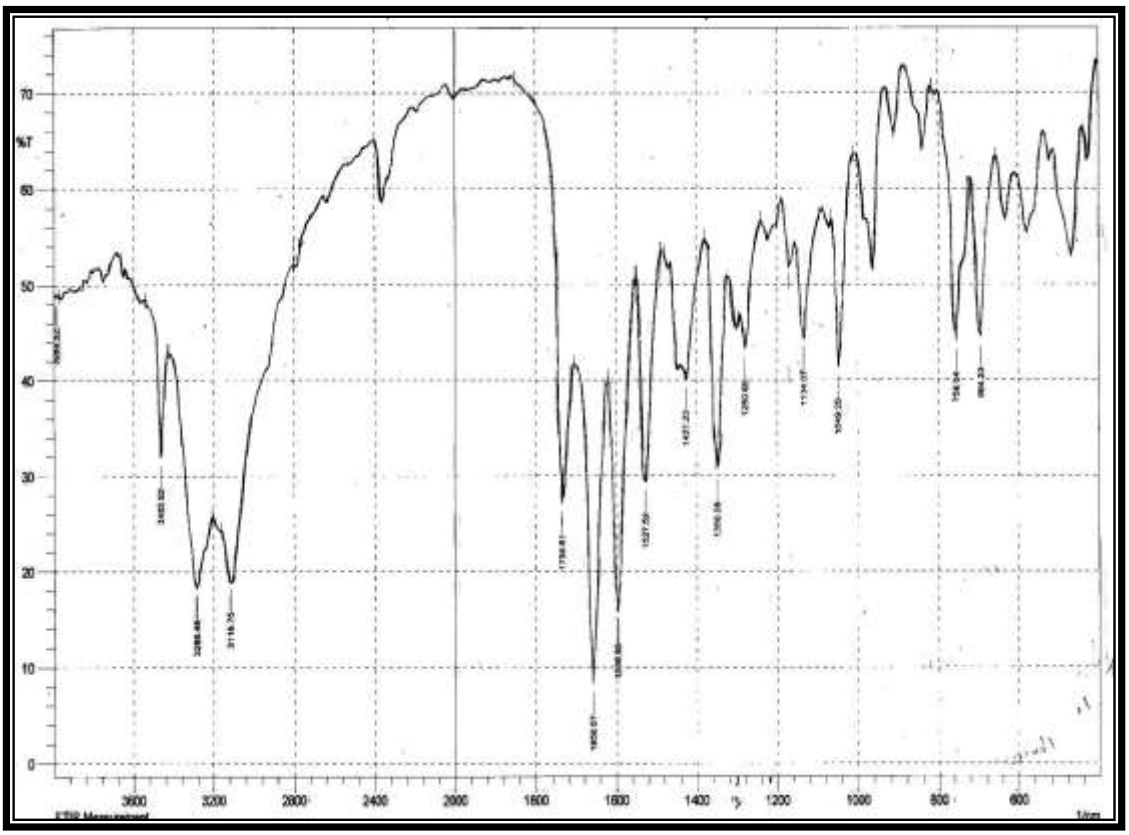


Fig. 2: FT-IR spectrum for compound [2]

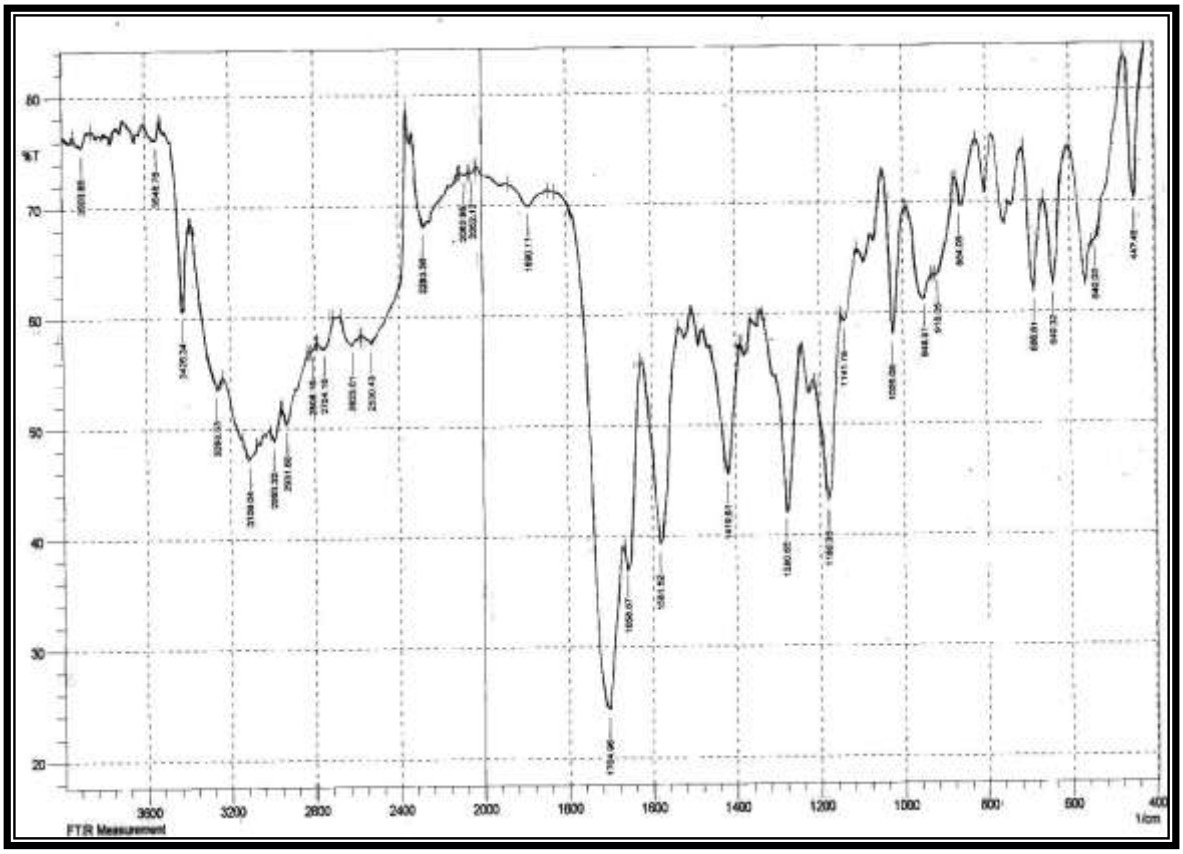


Fig. 3: FT-IR spectrum for compound [5]

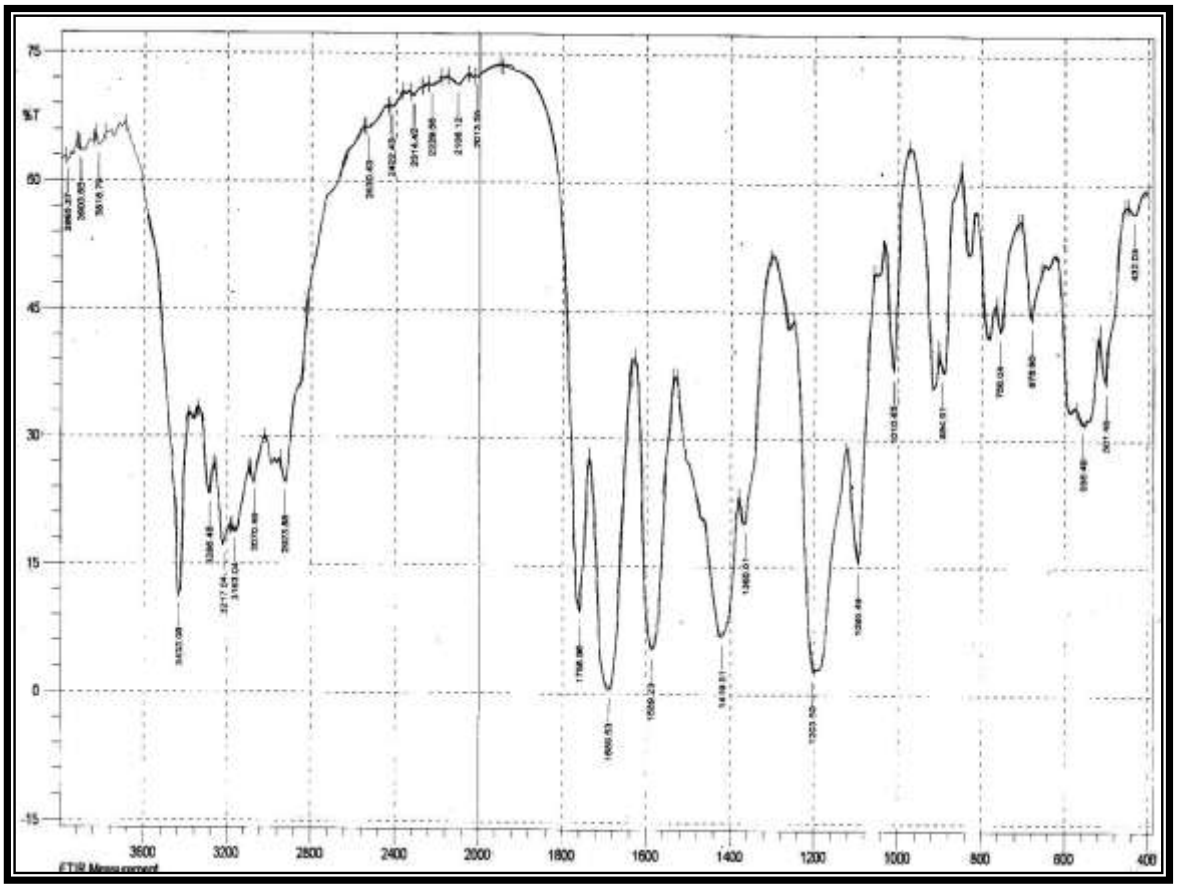


Fig. 4: FT-IR spectrum for compound [7]

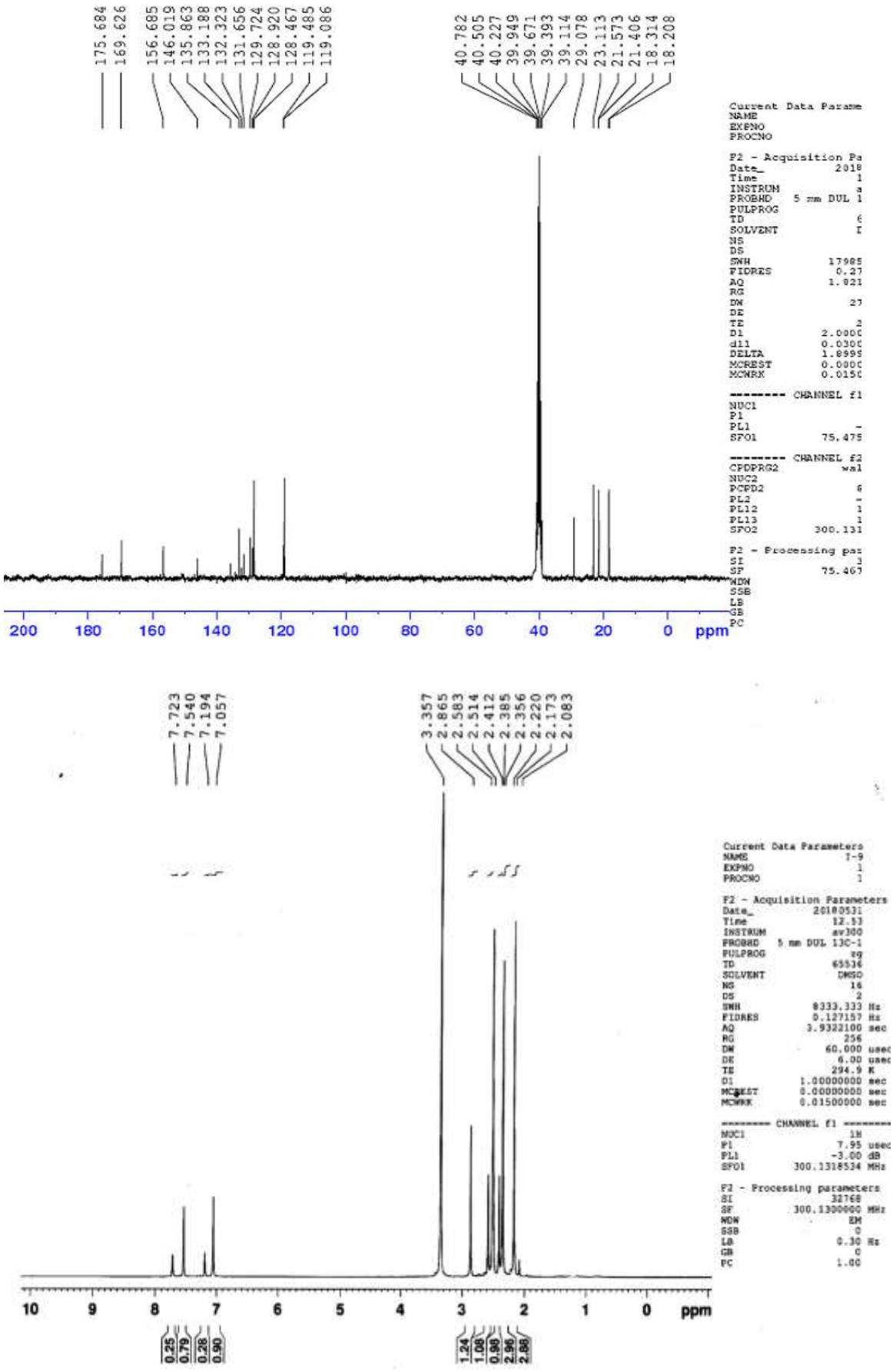


Fig. 5: ^{1}HNMR& ^{13}C-NMR spectrum for compound [6]

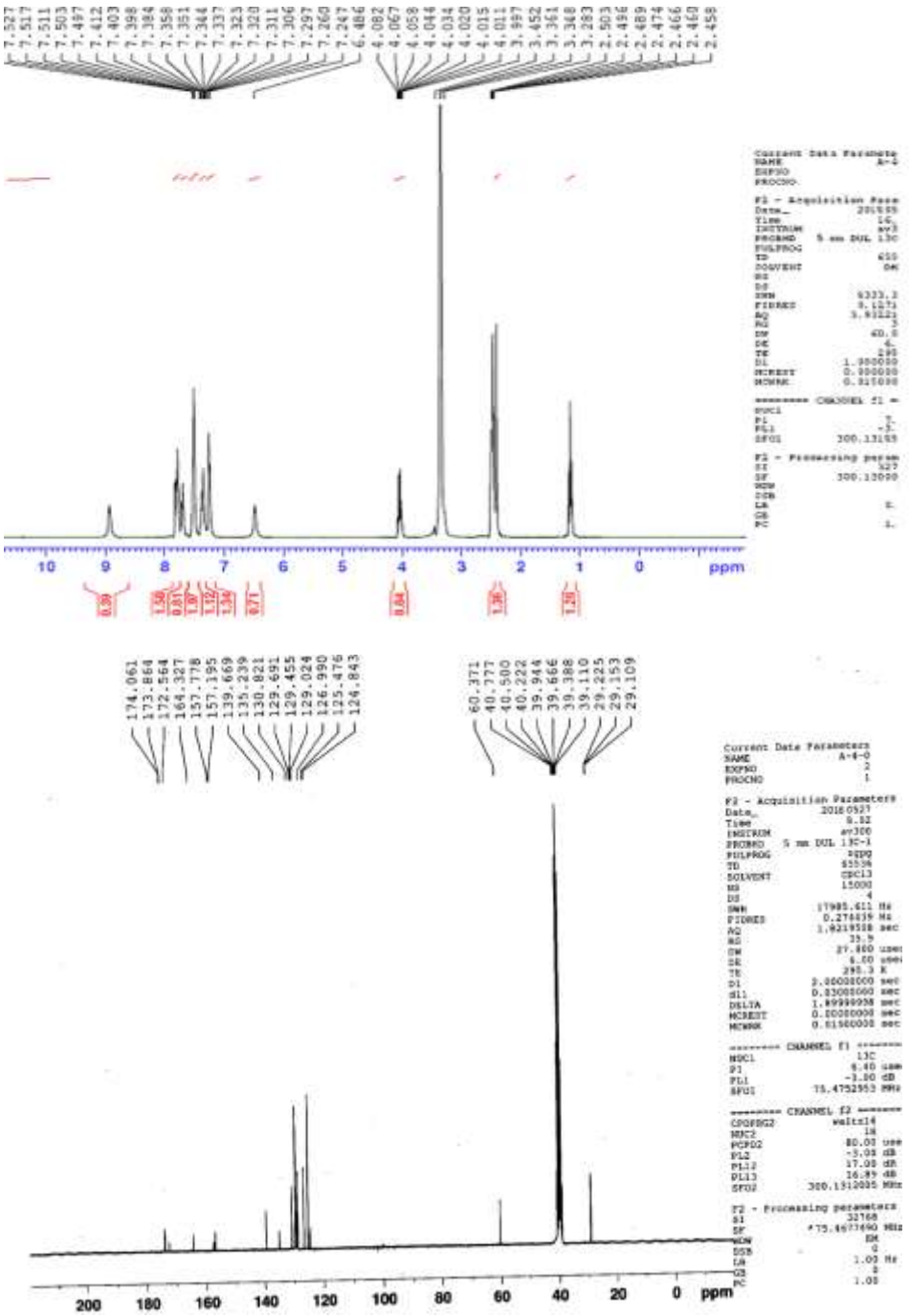


Fig. 6: [1]HNMR& [13]C-NMR spectrum for [10]

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