SYNTHESIS OF SUBSTITUTED AROMATIC CARBOXYLIC ACID DERIVATIVES AND THEIR BIOLOGICAL EVALUATION

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Abstract

The synthesis of new series of compounds was undertaken with the aim of exploiting immense potential of carboxylic acid derivatives and be able to come up with the most active biological activity profile. The titled compounds (1-8) were synthesized by stirring naphthoxy acetic acid hydrazide with corresponding aromatic aldehydes. The hydrazide was synthesized from the acid via its esterification followed by its nucleophilic substitution reaction i.e. hydrazinolysis with hydrazine hydrate. All the synthesized compounds were characterized on the basis of IR and ^{1}H-NMR spectroscopy techniques. All the final compounds were evaluated for their antimicrobial property using cup plate method. The antimicrobial result of newly synthesized compounds revealed that all the compounds showed moderate to good antimicrobial activity against the tested strains.

Keywords : Ester, Hydrazides, Hydrogenolysis, Antibacterial activity, Antifungal activity

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INTRODUCTION

The carboxylic group (-COOH or CO_2H) is one of the most widely occurring functional groups in chemistry as well as biochemistry.[1] The carboxylic group of a large family of related compounds called Acyl compounds or Carboxylic Acid Derivatives.[2] The stability of any type of carboxylic acid derivative is generally determined by the ability of its functional group to donate electrons to the rest of the molecules.[3] In essence, the more electronegative the atom or group attached to carbonyl group, the stable the molecule. The following derivative types are ordered in decreasing reactivity (the first is the most reactive):

Acyl halides (CO-X) > Acyl anhydrides (-CO-O-OCR) > Acyl Thioester (-CO-SR) >Acyl esters (-CO-OR) > Acyl Amides (-CO-NR_2)

Reactions of Carboxylic acid derivatives:

1) Acyl Group Substitution:

This is probably the single most important reaction of carboxylic acid derivatives. The overall transformation is defined by the following equation and may be classified either as nucleophilic substitution at an acyl group or as acylation of nucleophile.[4,5]

2) Reduction

Reductions of carboxylic acid derivatives might be expected to lead either to aldehydes or alcohols, functional groups having a lower oxidation state of the carboxyl carbon.[6]

Carboxylic Acid Derivatives and Acyl Groups:

The carboxylic acid derivatives can be distinguished from aldehydes and ketones by the presence of a group containing an electronegative heteroatom – usually oxygen, nitrogen, or sulphur-bonded directly to the carbonyl carbon.[7,8] You can think of a carboxylic acid derivative as having two sides. One side is the carbonyl group and the attached alkyl group; this is called an acyl group. The other side is the heteroatom-containing group.[9]

The relative reactivity of carboxylic acid derivatives:

Among the carboxylic acid derivatives, carboxylate group are the least reactive towards nucleophilic acyl substitution, followed by amides, then esters and (protonated) carboxylic acids, thioesters and finally acyl phosphates, which are the most reactive among the biologically relevant acyl groups.[10,11] The different reactivities of the functional groups can be understood by evaluating the basicity of the leaving group in each case. A thioester is more reactive than an ester.[12]

Esterification Reaction:

Esters are produced when carboxylic acids are heated with alcohols in the presence of an acid catalyst.[13] The catalyst is usually conc. H_2SO_4. Dry hydrogen chloride gas is used in some cases but these tend to involve aromatic esters. The esterification reaction is both slow and reversible.[14,15]

MATERIAL AND METHOD

Reagent, chemical and solvent used:

The reagents, chemical and solvents used during the course of these studies were commercially produced from various chemicals units.

Instruments/ Equipments/ Techniques used:

All the reactions were monitored by thin layer chromatography (TLC) by using silica gel as the stationary phase. The plates were visualizes in iodine chamber. The various solvent systems used for the same were cyclohexane: ethylacetate [7:3] and ethylacetate: cyclohexane [6:4].

Melting Point:

M.P. of synthesized compounds was determined by open glass capillary and was uncorrected. All the compounds show melting point in the range of 155-186^{0}C.

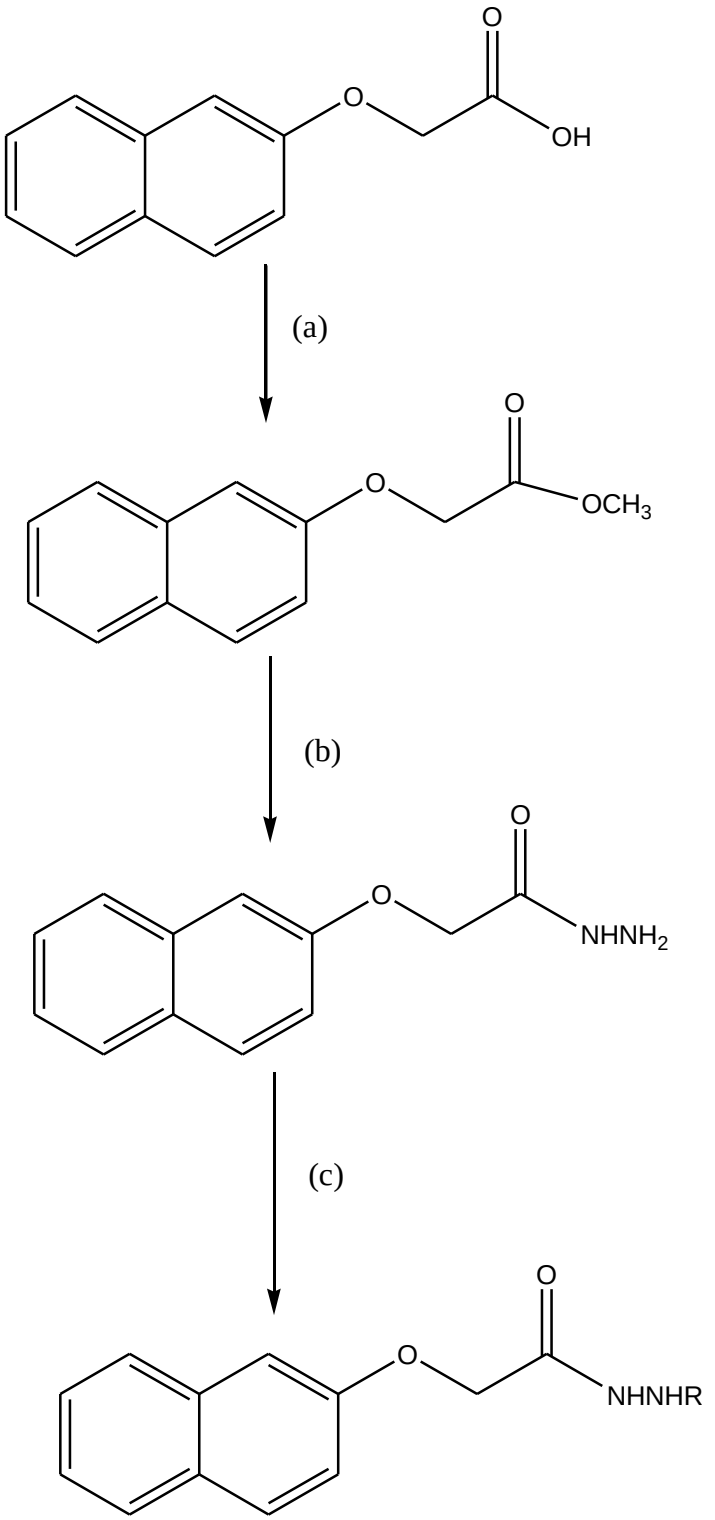
IR Spectrometer:

The IR Spectra were recorded by using SHIMADZU FT-IR spectrometer by maing KBr pellets.

NMR Spectrometer:

The proton nuclear magnetic resonance spectra (^{1}H-NMR) were recorded on Bruker 300 MHz and 400 MHz instrument in solvent (DMSO-d_6, $CDCl_3$: singlet; d: doublet; t: triplet; m: multiplet).

SCHEME:



Reagents and conditions :- (a) Menthol, Sulphuric acid, Reflux, 6 hrs (b) Hydrazine hydrate, Menthol,, Reflux, 3 hrs (c) Aromatic aldehyde, Stir, R = Aromatic aldehyde

SYNTHESIS:

Synthesis of esters:

A mixture of (0.01 mol) naphthoxy acetic acid and an excess of 25 ml of methanol with 0.7 ml of conc. Sulphuric acid as a catalyst was taken in a round bottom flask. This mixture was refluxed for 6 hrs. After refluxing, filter the product and washed with water and recrystallized from methanol. The product was collected and dried.

Synthesis of Hydrazide:

A mixture of methyl ester of (0.01 mol) naphthoxy acetic acid and (0.01 mol) hydrazine hydrate in 30 ml of methanol was refluxed for 3

hrs in round bottom flask. The product was filterd; washed and dried.

General procedure of synthesis of hydrazones:
A solution of hydrazide (0.01 mol) and aromatic aldehyde (0.01 mol) was treated in 25 ml of methanol/ DMF. The mixture was stirred at refluxing temperature 4-8 hrs. The solid product was collected and recrystallized from methanol to give pure compounds.

Physiochemical Data:
The physiochemical properties of synthesized compounds such as melting point, molecular weight, molecular formula, Rf value are examined carefully and are uncorrect. All the compounds were produced in appreciable yield. Their physiochemical properties are given in table.

Table 2 : Physiochemical data of synthesized compounds

Compound	Molecular Formula	Time (hrs)	Rf value	Yield(%)	M.P.(^{0}C)
1	$C_{19}H_{18}ClN_2O_3$	4	0.5	66	152-155
2	$C_{20}H_{18}N_2O_4$	5	0.6	70	150-153
3	$C_{21}H_{17}N_2O_3$	4	0.7	73	160-162
4	$C_{22}H_{22}N_2O_6$	5	0.8	65	180-183
5	$C_{21}H_{21}N_3O_3$	6	0.7	72	165-168
6	$C_{19}H_{16}N_2O_3$	5	0.5	67	170-172
7	$C_{19}H_{15}FN_2O_3$	8	0.6	63	184-186
8	$C_{20}H_{18}N_2O_4$	7	0.7	68	157-160

Table 3 : Solubility of synthesized compounds in different solvents

Compound	Water	Benzene	Chloroform	Ethanol
1	Insoluble	Slightly soluble	Soluble	Soluble
2	Insoluble	Slightly soluble	Soluble	Soluble
3	Slightly soluble	Slightly soluble	Soluble	Soluble
4	Insoluble	Slightly soluble	Soluble	Slightly soluble
5	Slightly soluble	Slightly soluble	Soluble	Soluble
6	Insoluble	Slightly soluble	Soluble	Slightly soluble
7	Insoluble	Slightly soluble	Soluble	Slightly soluble
8	Insoluble	Slightly soluble	Soluble	Soluble

BIOLOGICAL SCREENING
ANTIMICROBIAL ACTIVITY:
Antibacterial activity:
Past studies have shown that carboxylic acid derivative possess antimicrobial activity. In view of this an effort was made to study some synthesized compounds for their antimicrobial activity. The inhibition of the microorganism under standardized condition was utilized to demonstrate mcrobiological action of the ompounds.

Table 4 : Antibacterial activity of newly synthesized compounds

Compounds	Diameter of zone of inhibition (mm) Mean	
	E. coli	***S. aureus***
1	10.23	10.67
2	9.50	8.32
3	11.45	10.89
4	13.64	11.70
5	9.60	--
6	12.30	10.34
7	15.30	13.85
8	10.31	8.70
Ciprofloxacin	23.20	22.25
DMSO	00	00

Mean value of measured diameters of zones of inhibition at 100µg/ml.
MIC denotes minimum inhibitory concentration.
Nt denotes not tested.
"--" Means no activity.

Antifungal Activity:
The newly synthesized substituted phenacyl containing pyrazoles derivatives were screened for their antifungal activity against four different moulds by cup-plate method.

Table 5: Antifungal activity of synthesized compounds and standard drug

Compounds	Diameter of zone of inhibition (mm) Mean	
	C.albicans	***A.niger***
1	11.78	10.45
2	8.86	7.39
3	9.25	9.79
4	10.47	9.15
5	7.89	--
6	11.20	12.45
7	12.35	13.65
8	8.86	--
Fluconazole	23.25	22.20
DMSO	00	00

Mean value of measured diameters of zones of inhibition at 200 µg/ml for test compounds.
"—" Mean no activity.

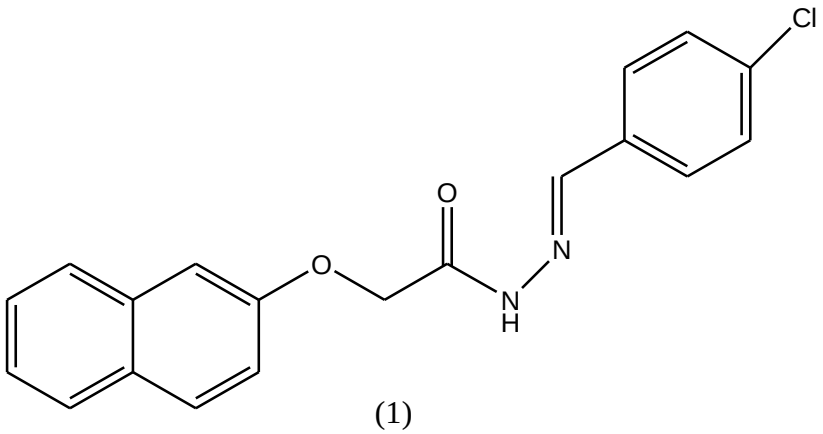
RESULT:
Chemistry:
The synthesis of compounds (1,2,3,4,5,6,7,8) was undertaken as per the scheme. The methyl ester of naphthoxy acetic acid was prepared. The hydrazide was prepared by the action of hydrazide hydrate on the methyl ester and the final products were prepared by the reaction of hydrazide and various aromatic aldehydes. Al the compounds were characterized by IR and ^{1}H-NMR.

The physicochemical data of hydrazones have been enlisted in Tables 2 and 3. The IR spectra of the final compounds showed different absorption bands.

The ^{1}H-NMR spectra of the compounds were recorded on Bruker 300 MHz instrument usimg DMSO-$d_6$or $CDCl_3$ as a NMR solvent. All the compounds were identified on the basis of chemical shift and splitting pattern.

***N'*-[(4-chlorophenyl) methylidene]-2-(napht ha len-2-yloxy) acetohydrazide (1)**
IR (KBr) spectrum of the compound showed absorption band at 3156 cm^{-1} was due to (Ar.st.). Absorption band at 1688 cm^{-1} was due to (C=O). Other band shows stretching at 618 cm^{-1} was due to (C-Cl), 3398 cm^{-1} was due to (N-H), 1599 cm^{-1} was due to (C=C). Absorption band at 1632 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of aryl group at $^\delta$ 7.2. The region showed multiplet at $^\delta$ 7.97-8.40 due to accounting of (11H, Ar-H). The region showed one singlet at $^\delta$ 8.25 of (1H, CH=N). One singlet at $^\delta$ 5.2 of (1H,-NH).

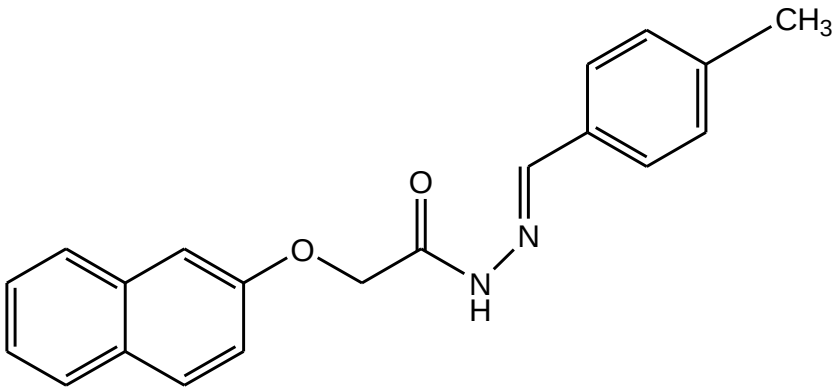


(1)

***N'*-[(4-methylphenyl) methylidene]-2-(naphthalen-2-yloxy) acetohydrazide (2)**

IR (KBr) spectrum of the compound showed absorption band at 3186 cm^{-1} was due to (Ar.st.). Absorption band at 1687 cm^{-1} was due to (C=O). Other band shows stretching at 3453 cm^{-1} was due to (N-H), 1505 cm^{-1} was due to (C=C). Absorption band at 1631 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of methyl group at δ 2.5. The region showed multiplet at δ 7.0-7.9 due to accounting of (11H, Ar-H) and one singlet of amine group at δ 9.4.The region showed one singlet at δ 8.5 of (1H, CH=N).

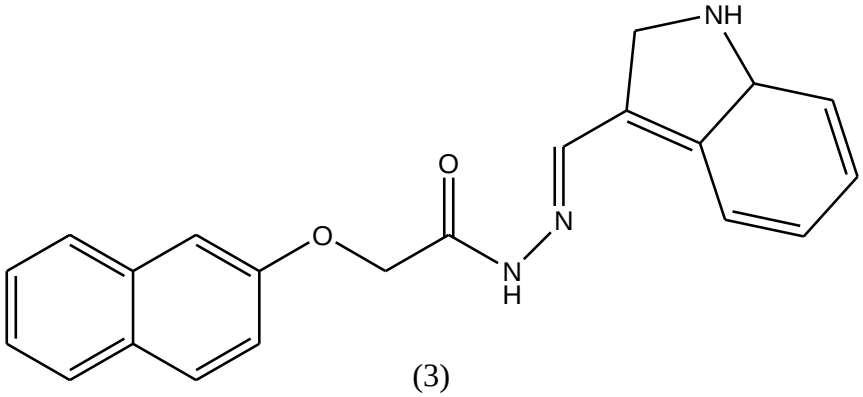


(2)

***N'*-[1*H*-indol-3-ylmethylidene]-2-(naphthalen-2-yloxy) acetohydrazide (3)**

IR (KBr) spectrum of the compound showed absorption band at 3037 cm^{-1} was due to (Ar.st.). Absorption band at 1629 cm^{-1} was due to (C=O). Other band shows stretching at 3313 cm^{-1} was due to (N-H), 1505 cm^{-1} was due to (C=C). Absorption band at 1514 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of amine group at δ 9.4. The region showed multiplet at δ 7.1-7.8 due to accounting of (11H, Ar-H).The region showed one singlet at δ 8.9 of (1H, CH=N).

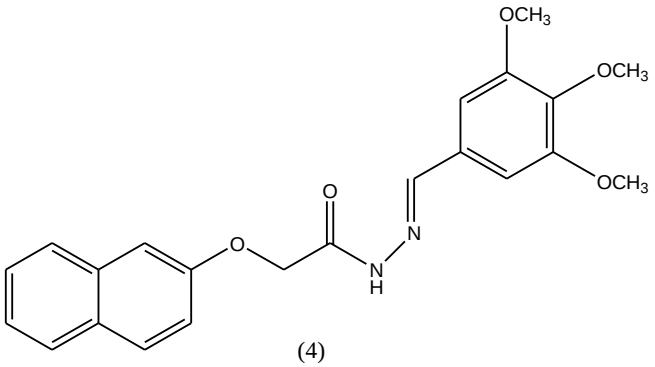


(3)

***N'*-[(3,4,5-trimethoxy)methylidene]-2-(naphthalen-2-yloxy) acetohydrazide (4)**

IR (KBr) spectrum of the compound showed absorption band at 3204 cm^{-1} was due to (Ar.st.). Absorption band at 1625 cm^{-1} was due to (C=O). Other band shows stretching at 3313 cm^{-1} was due to (N-H), 1542 cm^{-1} was due to (C=C). Absorption band at 1662 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed multiplet of methoxy group at δ 3.2-3.5. The region showed multiplet at δ 7.2-7.8 due to accounting of (11H, Ar-H).The region showed one singlet at δ 8.59 of (1H, CH=N).

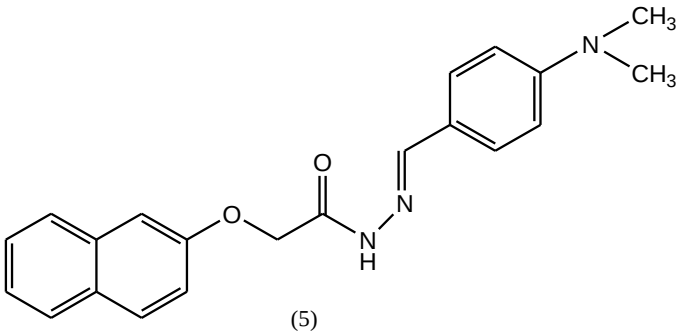


(4)

***N'*-[{4-(dimethylamino)phenyl}methylidene]-2-(naphthalen-2-yloxy) acetohydrazide (5)**

IR (KBr) spectrum of the compound showed absorption band at 3204 cm^{-1} was due to (Ar.st.). Absorption band at 1603 cm^{-1} was due to (C=O). Other band shows stretching at 3314 cm^{-1} was due to (N-H), 1514 cm^{-1} was due to (C=C). Absorption band at 1686 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of methyl group at $^{\delta}$ 2.9. The region showed multiplet at $^{\delta}$ 7.2-7.9 due to accounting of (11H, Ar-H) and one singlet of amine group at $^{\delta}$ 9.3.The region showed one singlet at $^{\delta}$ 8.5 of (1H, CH=N).

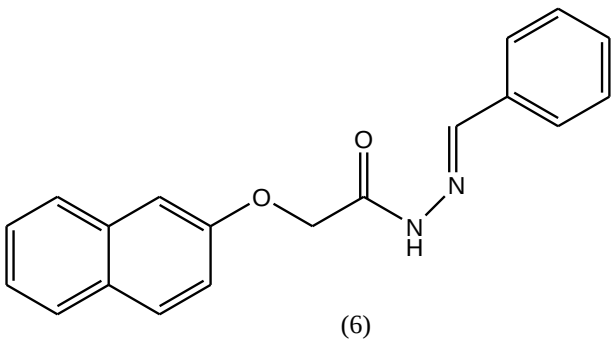


(5)

2-(naphthalen-2-yloxy-*N'*-[(E)-phenyl methylidene] acetohydrazide (6)

IR (KBr) spectrum of the compound showed absorption band at 3187 cm^{-1} was due to (Ar.st.). Absorption band at 1685 cm^{-1} was due to (C=O). Other band shows stretching at 3343 cm^{-1} was due to (N-H), 1599 cm^{-1} was due to (C=C). Absorption band at 1631 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of amine group at $^{\delta}$ 9.3. The region showed multiplet at $^{\delta}$ 7.2-7.8 due to accounting of (11H, Ar-H).The region showed one singlet at $^{\delta}$ 8.3 of (1H, CH=N).

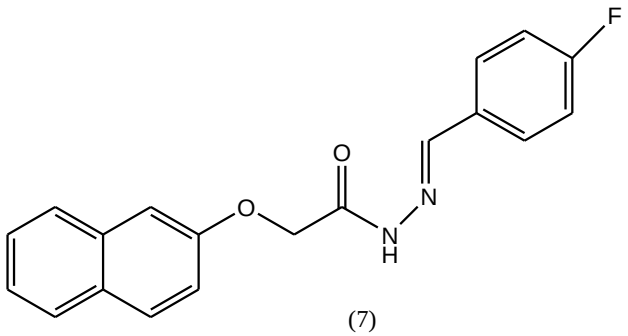


(6)

***N'*-[(4-fluorophenyl)methylidene]-2-(naphthalen-2-yloxy) acetohydrazide (7)**

IR (KBr) spectrum of the compound showed absorption band at 3168 cm^{-1} was due to (Ar.st.). Absorption band at 1632 cm^{-1} was due to (C=O). Other band shows stretching at 759 cm^{-1} was due to (C-F), 3314 cm^{-1} was due to (N-H), 1574 cm^{-1} was due to (C=C). Absorption band at 1519 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of amine group at $^\delta$ 9.8. The region showed multiplet at $^\delta$ 7.1-7.9 due to accounting of (11H, Ar-H).The region showed one singlet at $^\delta$ 8.4 of (1H, CH=N).

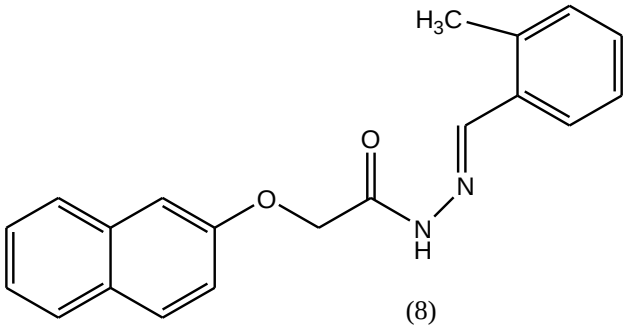


(7)

***N'*-[(2-methylphenyl) methylidene]-2-(naphthalen-2-yloxy) acetohydrazide (8)**

IR (KBr) spectrum of the compound showed absorption band at 3156 cm^{-1} was due to (Ar.st.). Absorption band at 1690 cm^{-1} was due to (C=O). Other band shows stretching at 3354 cm^{-1} was due to (N-H), 1560 cm^{-1} was due to (C=C). Absorption band at 1660 cm^{-1} was due to (O=C-NH) stretching.

The ^{1}H-NMR spectrum of the compound showed singlet of amine group at $^\delta$ 9.6. The region showed multiplet at $^\delta$ 7.2-7.9 due to accounting of (11H, Ar-H).The region showed one singlet at $^\delta$ 8.2 of (1H, CH=N). One singlet at $^\delta$ 2.9 of methyl group.



(8)

Antimicrobial activity:

The antimicrobial activity of newly synthesized compounds was determined in-vitro by simple susceptibility test using agar-well diffusion technique. All the compounds were tested for antibacterial activity against *Escherichia coli* (NCTC, 10418) and *Staphylococcus aureus* (NCTC, 65710). For antifungal activity two strains of fungi were employed *viz. C.albicans* and *A.niger*. A concentration of 100 µg/ml of tested compounds was prepared in DMSO. Ciprofloxacin and Fluconazole were used as standard drugs under the similar conditions for antibacterial and antifungal activity at 100 µg/ml. The results of antimicrobial activity have been presented in the **Table (4) and (5).**

Antibacterial activity:

The antibacterial results showed that of the compounds were found active against both the bacterial strains. The test compounds 1,2,3,4,5,6,7 and 8 showed moderate to good antibacterial activity against the test organisms. It was observed that the compound 7 showed highest zone of inhibition (mm) against test organisms. The higher antibacterial activity of compounds 7 and 3 may be attributed to the pharmacologically active 4-fluoro and indole substitutions attached to the benzoyl group of these compounds. It was observed that compound 7 showed the highest antibacterial activity against *E.coli* and *S.aureus* and the compounds 1,4 and 6 showed the moderate antibacterial activity against the test strains.

Antifungal activity:

Antifungal screening data of compounds revealed that the tested compounds showed considerable antifungal activity at concentration of 100µg/ml as shown by their measured zone of inhibition. However, the compound 7 which showed marked antibacterial activity was also found most active against both the fungal strains used in the studies.

CONCLUSION:

The synthesis of new series of compounds was undertaken with the aim of exploiting immense potential of carboxylic acid derivatives and be able to come up with the most active biological activity profile.

The titled compounds (1-8) were synthesized by stirring naphthoxy acetic acid hydrazide with corresponding aromatic aldehydes. The hydrazide was synthesized from the acid via its esterification followed by its nucleophilic substitution reaction i.e. hydrazinolysis with hydrazine hydrate.

All the synthesized compounds were characterized on the basis of IR and ^{1}H-NMR spectroscopy techniques. All the final compounds were evaluated for their antimicrobial property using cup plate method.

The physiochemical data of newly synthesized compounds have been enlisted in Table 2. The solubility chart of compounds has been enlisted in Table 3. The antibacterial activity and antifungal activity data were given in Table 4 and 5 respectively.

The antimicrobial result of newly synthesized compounds revealed that all the compounds showed moderate to good antimicrobial activity against the tested strains.

Moreover the compound 7 emerged as potent antimicrobial agent in the series and its framework constitute a fruitful model for further investigation in the development of a new class of antimicrobial agents.

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