SYNTHESIS, ANTIINFLAMMATORY AND ANTIMICROBIAL ACTIVITIES OF NEWER SERIES OF 1,3,4-OXADIAZOLES

Dissertation Submitted to

The Tamil Nadu Dr. M.G.R. Medical University,

Chennai – 600 032.

In partial fulfillment for the award of Degree of

MASTER OF PHARMACY

(Pharmaceutical Chemistry)

Submitted by

VENKATRAMAN. S

Reg. No.26106038

Under the Guidance of

Mr. A. THIRUGNANA SAMBANTHAN, M.Pharm., (Ph.D.)
Assistant Professor,
Department of Pharmaceutical Chemistry.

ADHIPARASAKTHI COLLEGE OF PHARMACY,

(Accredited by “NAAC” With CGPA of 2.74 on a Four point Scale at “B” Grade)

MELMARUVATHUR – 603319.

MAY – 2012.

SYNTHESIS, ANTIINFLAMMATORY AND ANTIMICROBIAL ACTIVITIES OF NEWER SERIES OF 1,3,4-OXADIAZOLES

Dissertation Submitted to

The Tamil Nadu Dr. M.G.R. Medical University,

Chennai – 600 032.

In partial fulfillment for the award of Degree of

MASTER OF PHARMACY

(Pharmaceutical Chemistry)

Submitted by

VENKATRAMAN. S

Reg. No.26106038

Under the Guidance of

Mr. A. THIRUGNANA SAMBANTHAN, M.Pharm., (Ph.D.)
Assistant Professor,
Department of Pharmaceutical Chemistry.

ADHIPARASAKTHI COLLEGE OF PHARMACY,

(Accredited by “NAAC” With CGPA of 2.74 on a Four point Scale at “B” Grade)

MELMARUVATHUR – 603319.

MAY – 2012.

CERTIFICATE

This is to certify that the dissertation entitled **"SYNTHESIS, ANTIINFLAMMATORY AND ANTIMICROBIAL ACTIVITIES OF NEWER SERIES OF 1,3,4-OXADIAZOLES"** submitted to The Tamil Nadu Dr. M.G.R. Medical University in partial fulfillment for the award of the Degree of Master of Pharmacy (Pharmaceutical Chemistry) was carried out by **VENKATRAMAN.S** (Reg. No.26106038) in the Department of Pharmaceutical Chemistry under my direct guidance and supervision during the academic year 2011-2012.

Place: Melmaruvathur

Date:

Mr. A. THIRUGNANA SAMBANTHAN, M.Pharm., (Ph.D.)
Assistant Professor,
Department of Pharmaceutical Chemistry,
Adhiparasakthi College of Pharmacy,
Melmaruvathur- 603319.

CERTIFICATE

This is certify that the dissertation entitled **"SYNTHESIS, ANTIINFLAMMATORY AND ANTIMICROBIAL ACTIVITIES OF NEWER SERIES OF 1,3,4-OXADIAZOLES"** is the bonafide research work carried out by **VENKATRAMAN. S** in the Department of Pharmaceutical Chemistry, Adhiparasakthi College of Pharmacy, Melmaruvathur, which is affiliated to The Tamil Nadu Dr.M.G.R Medical University under the guidance of **Mr. A. THIRUGNANASAMBANTHAN, M.Pharm., (Ph.D.)** Assistant Professor, Department of Pharmaceutical Chemistry, Adhiparasakthi College of Pharmacy during the academic year 2011-2012.

Place: Melmaruvathur

Date:

Prof. (Dr). T. VETRICHELVAN, M.Pharm., Ph.D.,
Principal,
Adhiparasakthi College of Pharmacy,
Melmaruvathur.

ACKNOWLEDGEMENT

I honestly acknowledge the blessing of **HIS HOLINESS ARULTHIRU ADIGALAR**, President and with respect to **THIRUMATHI AMMA,** Vice President, ACMEC Trust, Melmaruvathur for growing blessing in each step of the study.

I avail this opportunity with great pleasure and deep sense of gratitude to express my heartful thanks to my guide and **Mr. A. THIRUGNANA SAMBANTHAN, M.Pharm., (Ph.D.)** Assistant Professor, Department of Pharmaceutical Chemistry, Adhiparasakthi College of Pharmacy for his memorable guidance, criticism, and valuable advice, constant encouragement, patience and meaningful support in bringing this project to a great success.

I greatly indebted to **Prof. (Dr.) T. VETRICHELVAN, M.Pharm., Ph.D**., Principal, Adhiparasakthi College of Pharmacy for the abundant help rendered by him during the course of work. It has been my immense pleasure to have his invaluable guidance and encouragement in carrying out this work.

I would fail in my duty, if I do not mention the help and valuable advices and suggestions by **Mrs. D. NAGAVALLI, M. Pharm., Ph.D.,** Professor, **Mr. M. SUGUMARAN, M. Pharm., (Ph.D),** Assosiate Professor**,** Department of Pharmaceutical Chemistry and other faculty members of Adhiparasakthi college of Pharmacy, Melmaruvathur, for their valuable help and guidance during the course of my research work.

I have great pleasure in expressing sincere heartfelt thanks to **Mr. A.S.K. SANKAR, M.Pharm, (Ph.D),** Associate Professor, Department of Pharmaceutical Analysis, Vel's College of Pharmacy, Chennai, for his encouragement and support for the successful completion of my thesis work.

My Sincere thanks to our Lab Technicians **Mrs. S. Karpagavalli**, **D.Pharm, Mr. M. Gomathisankar D.Pharm, Mrs. N. Thatchayani D.Pharm.,** Electrician Assistant **Mr. H. NAGARAJ** and office assistant **Mr. I. Kumar** for their kind help through this work.

I extend my hearty thanks to **Mr. K. MARUTHAPANDIAN**, Ideal Analytical and Research Institution, Puducherry, for taking IR Spectroscopy studies and also thanks to **Mr. Avatar singh,** Executive officer, SAIF, Punjab University, Chandigarh, for taking NMR spectroscopy.

I extremely grateful to **Dr. R. Murugesan Ph.D.,** Sr. scientific officer Grade-I, SAIF, IIT- MADRAS, for carrying out MASS spectral analysis and I also thankful to **Mrs. E. Mohana Bharathi,** Microbiologist, Pharma analytical lab, Puducherry, for carrying out the antimicrobial studies.

My special thanks to **Mrs. S. Shoba, M. Pharm.,** Assistant Professor and **Ms. M. VijayaKumari, M. Pharm.,** Lecturer**,** Department of Pharmacology, for their kind help during successful completion of animal studies in my project work.

I extend my gratitude to Librarian **Mr. M. Suresh, M.L.I.S.**, for providing books, reference articles, and journals to make our project informative and thankful to all of our **Teaching staff members**, **Non teaching members**, **Lab assistants** and **workers** for their encouragement and co-operation throughout the project work.

I am indeed very much special thankful to the **Mr. R. Logesh, Mr. R. Sabarinathan**, **Mr. E. Ramajayam**, **Mr. R.E. Seenuvasan** and **Mr. R. Tamilselvan** for their valuable suggestion throughout the project work.

I must be thankful to all my colleagues, seniors, juniors and my lovable friends for their support and suggestion during my work.

Finally yet importantly, I am greatly obliged to my mother **Mrs. S. Vijaya,** my father **Mr. N. Sakthivel**, my well-wisher brothers **Mr. S. Manikandan**, **Mr. K. Rajesh Kumar,** and **Mr. K. NareshKumar,** my sister in law **Mrs. M. Sivaranjini** and **my relatives** for their inspiration, guidance, moral support, constant prayers for my successful endeavors.

S.VENKATRAMAN

DEDICATED

TO

MY PARENTS ,

BROTHERS

AND

RELATIVES

CONTENTS

LIST OF TABLES

LIST OF FIGURES

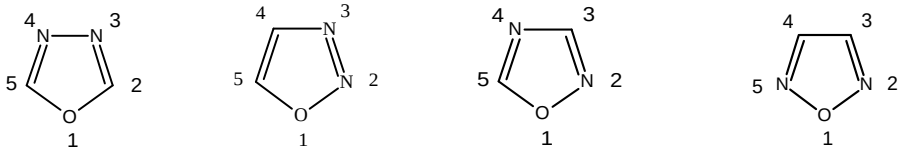
ABBREVIATIONS

$POCl_3$	:	Phosphorusoxychloride
CS_2	:	Carbondisulphide
^0C	:	Degree Celcius
µg	:	Microgram
%	:	Percentage
g	:	Grams
mg	:	Milligrams
ml	:	Milliliter
M.P	:	Melting point
pH	:	Hydrogen ion concentration
^1H-NMR	:	Proton Nuclear Magnetic Resonance
IR	:	Infra Red
h	:	Hours
mts	:	Minutes
mol	:	Mole
m/z	:	Mass / charge
R_f	:	Retardation factor
ppm	:	Parts per million
w/v	:	weight/volume
CMC	:	Carboxy methyl cellulose
CPCSEA	:	Committee for the Purpose of Control and Supervision of Experiments on Animals
ANOVA	:	Analysis of Variance

INTRODUCTION

I. INTRODUCTION

Oxadiazoles are five membered heterocyclic ring system containing one oxygen and two nitrogen atom in the ring. There are four isomeric types of oxadiazole depending on the positions of the nitrogen atoms in the oxadiazole ring and are numbered as

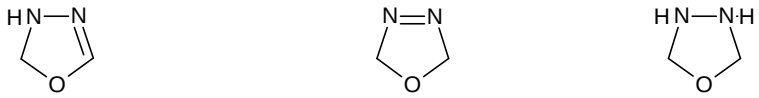


1,3,4-Oxadiazole 1,2,3-Oxadiazole 1,2,4-Oxadiazole 1,2,5-Oxadiazole

Out of these 1,3,4-oxadiazole are found to be biologically more potent. It is thermally stable aromatic heterocycle and exists in two partially reduced forms depending on the position of double bond.

(1) 2, 3-dihydro-1,3,4-Oxadiazole (1,3,4-Oxadiazoline)

(2) 2, 5-dihydro-1,3,4-Oxadiazole (1,3,4-Oxadiazolidine)



(1) (2) (3)

(3) The completely reduced form of 1,3,4-Oxadiazole is designated as 2,3,4,5-tetrahydro-1,3,4-Oxadiazole (1,3,4-oxadiazolidine).

Among different types of oxadiazole nucleus containing molecules, many 2,5-disubstituted 1,3,4-oxadiazoles had shown better biological activities. 2,5- di substituted -1,3,4-oxadiazoles are potent biological activities such as analgesic, anti inflammatory, anticonvulsant, sedatives, hypnotics, antiemetic, diuretic, CNS

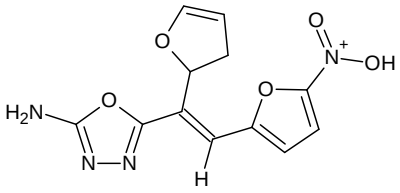
stimulant action, hypotensive, hypoglycaemic, muscle relaxant, herbicidal, bactericidal, and fungicidal activity. Some of them are strongly inhibit the enzymes such as monoamine oxidase, cyclooxygenase, lipoxygenase and succinate dehydrogenase etc.

Most commonly methods for the preparation of 1,3,4-oxadiazoles from acid hydrazides by treating with carbondisulphide and various aromatic acids separately. Oxadiazoles are very reactive in nature, undergoes for electrophilic and nucleophiilc substitution, thermal and photochemical reactions. (*Gupta R. R et al.*,)

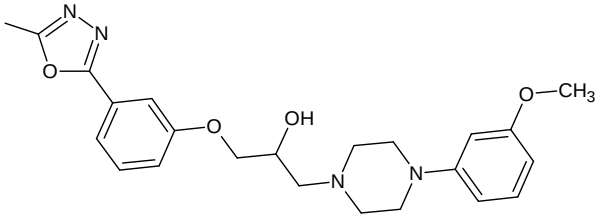
The thione derivatives of 1,3,4-oxadiazoles involved in mannich reaction, with formaldehyde and secondary amines to get the mannich bases of 1,3,4-oxadiazole derivatives.

Many drugs contain oxadiazoles; few familiar drugs are:

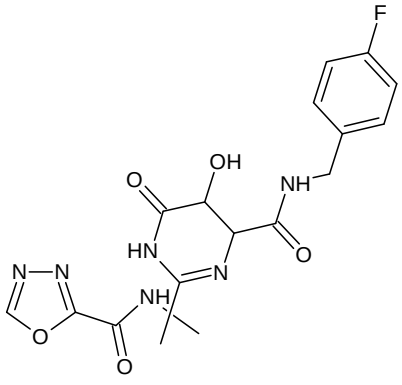
Furamizol (antibacterial)



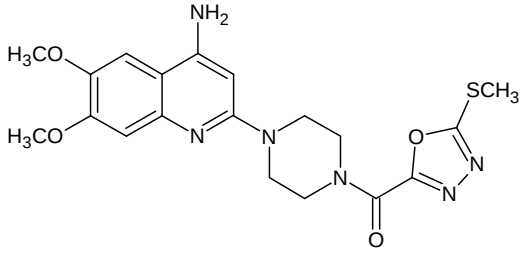
Nesapidil (vasodilator)



Raltegravir (antireteroviral)



Tiodazosin (antihypertensive)



INFLAMMATION

Inflammation is defined as the local response to living mammalian tissues to injury due to any agent. Specifically it is a series of molecular and cellular responses acquired during evolution designed to eliminate foreign agents and promote repair of damaged tissues.

Inflammation can be classified as two types,

- Acute inflammation
- Chronic inflammation

1. Acute inflammation

Acute inflammation is the initial response of the body to harmful stimuli and is achieved by the increased movement of plasma and leukocytes from the blood in to the injured tissues.

2. Chronic inflammation

Chronic inflammation is also known as prolonged inflammation, leads to a progressive shift in the type of cells which are present at the site of inflammation and is characterised by simultaneous destruction and healing of the tissue from the inflammatory process.

Antiinflammatory activity

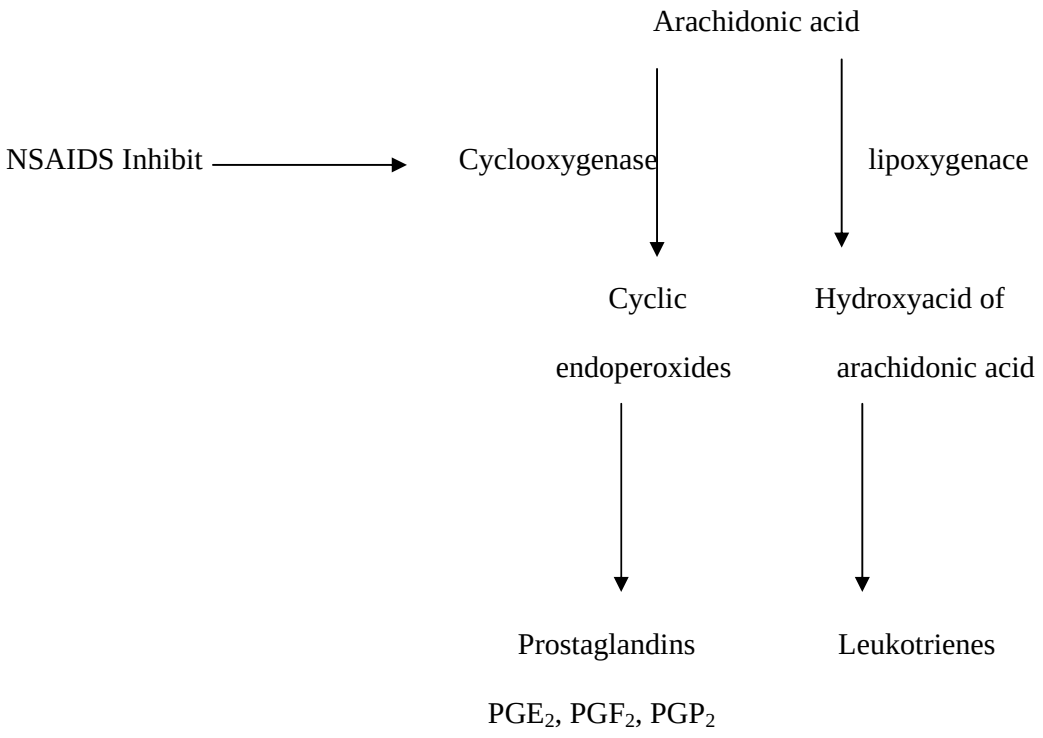
Nonsteroidal antiinflammatory drugs are used primarily to treat inflammation, mild to moderate pain and fever. It is also used to treatment of headaches, arthritis, sports injuries and menstrual cramps.

The commonly employed methods to evaluate the antiinflammatory activity of the compounds are Erythema assays, Edema assays, Granuloma assays and Experimental arthritis assays.

Mechanism of action

The nonsteroidal antinflammatory drugs act by inhibiting the biosynthesis of prostanglandins, which is the basis cause behind fever, pain and inflammatory condition. The biosynthesis of prostaglandin is catalysed by microsomal enzyme present in almost every mammalian cell type except erythrocytes.

Biosynthesis of prostaglandins



Antimicrobial activity

An antimicrobial agent is anything that can kill or inhibit the growth of bacteria such as high heat or radiation or a chemical substance. The antimicrobial agents are broadly classified into antibacterial, antifungal, antiviral, antiprotozoal, antiparasitic and anthelmintics. The antimicrobial agents act by interfering with cell wall synthesis, plasma membrane integrity, nucleic acid synthesis, ribosomal function and folate synthesis.

Antibacterial agents are used in relatively low concentrations in or upon the bodies of organisms to prevent or treat specific bacterial diseases without harming the host organism. The most commonly used methods for determination of antimicrobial activity are cylinder plate or cup plate method, turbidimetric or tube assay method and disc diffusion method.

Based on these reports, we have planned to synthesize the mannich bases of 1,3,4-oxadiazole derivatives and 2,5-disubstituted-1,3,4-oxadiazole derivatives and screen for antiinflammatory and antimicrobial activities.

OBJECTIVES OF THE WORK

II. OBJECTIVES OF THE WORK

- ✓ To synthesise the oxadiazole derivatives by cyclisation of acid hydrazides by two methods,

  (i) $POCl_3$ with aromatic acids and

  (ii) CS_2 with potassium hydroxide.

- ✓ To synthesise the mannich base of 1,3,4-oxadiazole by reacting thione oxadiazoles with appropriate amines and formaldehyde.

- ✓ To characterise the structure of synthesized compounds by Infra red spectroscopy, ^1H NMR spectroscopy and MASS spectroscopy.

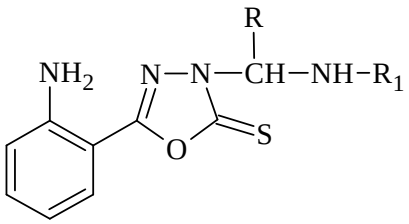
- ✓ To evaluate the antiinflammatory activity by carregenan induced rat paw edema method.

- ✓ To screen the antibacterial and antifungal activity by disc diffusion method.

LITERATURE REVIEW

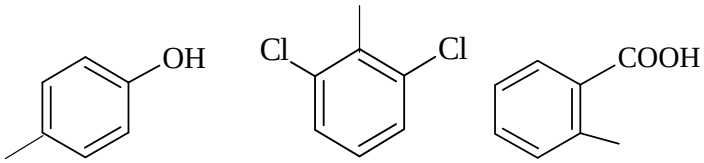
III. LITERATURE REVIEW

Selvakumar K. *et al.*, (2011) synthesized mannich bases of substituted 1,3,4-oxadiazole derivatives and evaluated for antimicrobial activity.

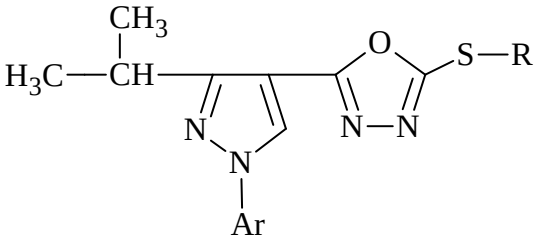


R = H, CH_3.

R_1 =

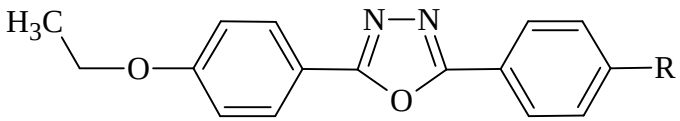


Abuzaid M. A. *et al.*, (2011) carried out the synthesis of novel thioglycosides incorporating 1,3,4 –oxadiazole, triazole and triazine and evaluated for antitumor activity.



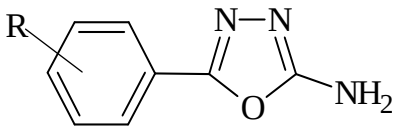
R = Thioglucosides.

Nadia Salih *et al.*, (2011) synthesized and estimated the potential antimicrobial activity of some 2,5-disubstituted-1,3,4-oxadiazole derivatives.



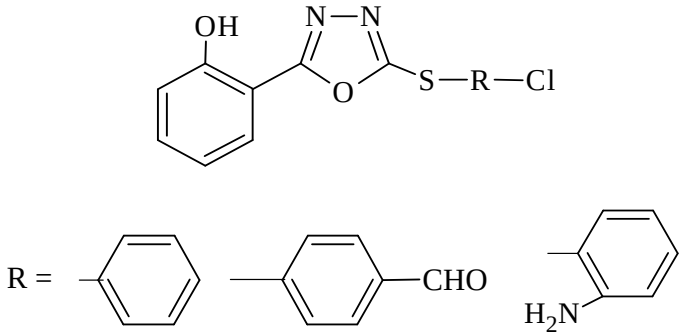
R = Br, NO_2, OCH_3.

Manish srivastava *et al.,* (2011) investigated the antibacterial and antifungal activities of newly synthesized 2-amino-5-substituted-1,3,4-oxadiazole derivatives.

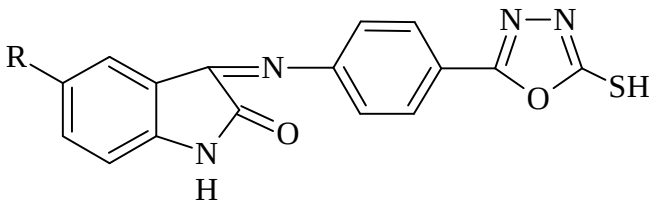


R = 3-Cl, 3-OCH_3, 3-NO_2, 4-Cl, 4-OCH_3, 4-NO_2.

Palak K Parikh *et al.,* (2011) synthesized some novel 1,3,4-oxadiazole derivatives and reported as potential antibacterial and antifungal activities.

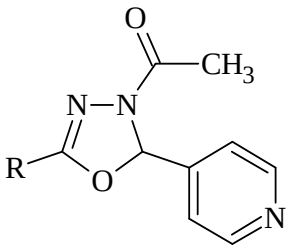


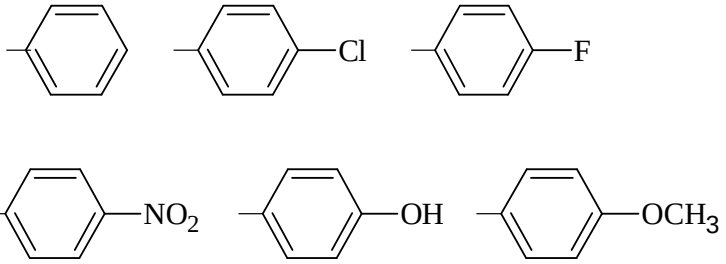
Sarangapani Manda *et al.,* (2011) synthesized 5-substituted-3-{4-(5-mercapto-1,3,4-oxadiazol-2-yl)phenylimino}-indolin-2-one derivatives and screened for anticancer activity against HeLa cancer cell lines using MTT assay.



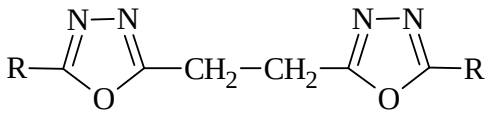
R = H, F, Cl, Br, NO_2, CH_3, COOH.

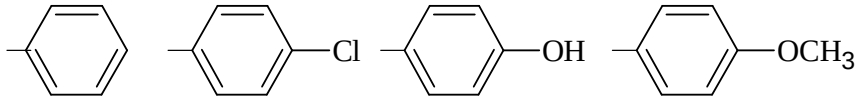
Sharma V. K. *et al.*, (2011) used microwave assisted method to synthesis some 1,3,4 oxadiazole derivatives and evaluated for their analgesic, antiinflammatory and antimicrobial activities.



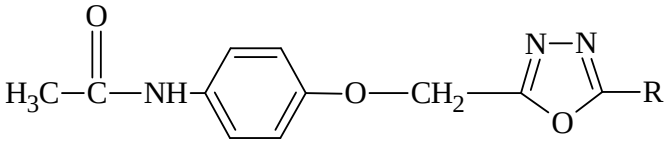
R = 

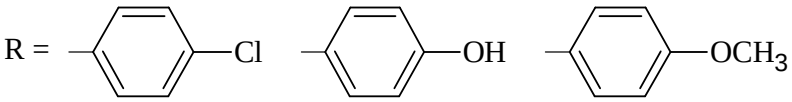
Vijay V Dabholkar *et al.*, (2011) investigated the antimicrobial activity of synthesized 2-substituted-1,3,4-oxadiazoles derivatives.

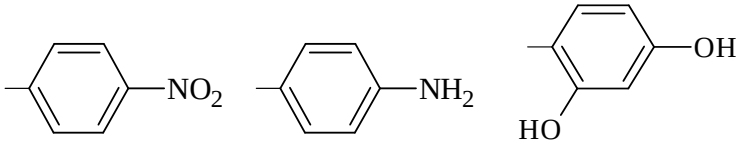


R = 

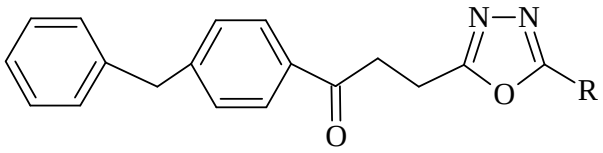
IIango K. *et al.*, (2010) carried out the antiinflammatory activity of synthesized 2,5-disubstituted-1,3,4-oxadiazoles by using carragenan induced rat paw edema method.

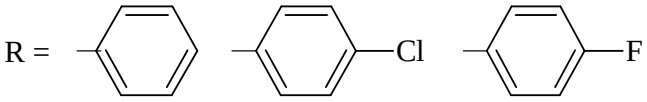


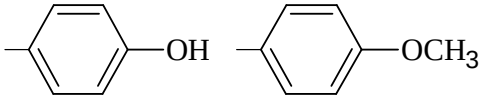




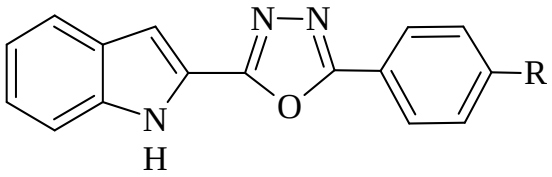
Husian A. *et al.*, (2010) synthesized β-aroyl propionic acid based 1,3,4-oxadiazoles and tested for antiinflammatory, analgesic, lipid peroxidation, ulcerogenic & antibacterial properties.





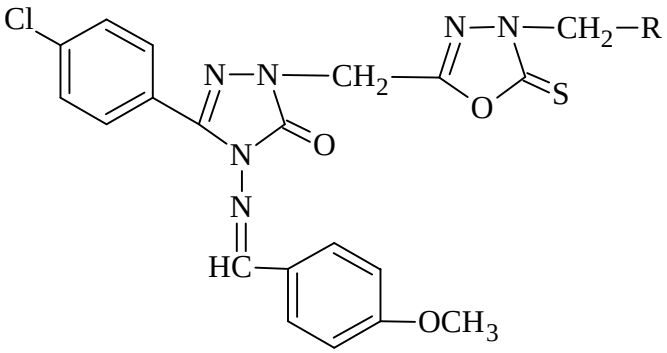


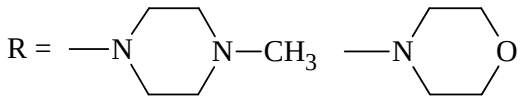
Somani R. R. *et al.*, (2010) studied the various central nervous system effects like depressant, sedative-hypnotic, anticonvulsant and anxiolytic activities of some mannich bases of 2,5-disubstituted-1,3,4-oxadiazole on albino mice.



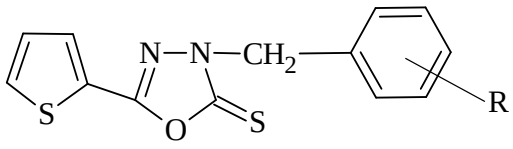
R = H, 2-Cl, 4-NO_2, 4-F.

Hakan Bektas *et al.*, (2010) synthesized and screened for antimicrobial activity of some new 1,2,4- triazole incorporated with 1,3,4-oxadiazole derivatives.



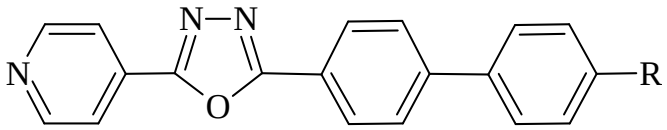


Mohamed A. *et al.*, (2010) synthesized some new 5-(2-thienyl)-1,2,4-triazoles and 5- (2-thienyl)-1,3,4-oxadiazoles and these derivatives were evaluated for antimicrobial activity.



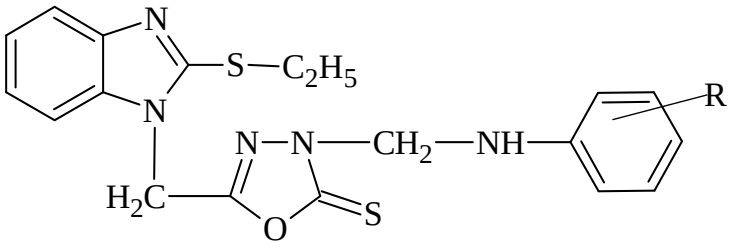
R= 2-F, 4-F, 4-Cl, 2-CF_3, 3-CF_3

Biju C. R. *et al.,* (2010) studied the analgesic, antiinflammatory and antimicrobial activities of synthesized biphenyl substituted-1,3,4-oxadiazole derivatives.



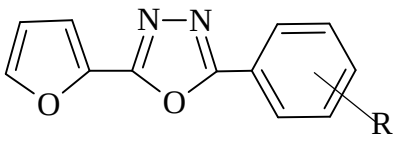
R = Cl, OH, COCl, OCH_3.

Somani R. R. *et al.,* (2009) synthesized some newer mannich bases by microwave assisted method and evaluated for their antimicrobial activity at three different concentrations.



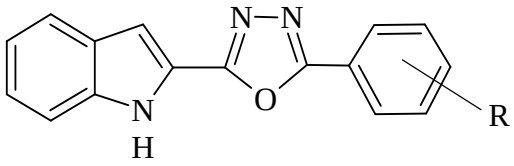
R = 3-Cl, 3-CH_3, 4-F, 4-OH, 4-NO_2.

Rina Das *et al.,* (2009) screened for antiinflammatory, antifungal and antibacterial activities of newly synthesized 1,3,4-oxadiazole derivatives substituted with furan and aromatic rings.



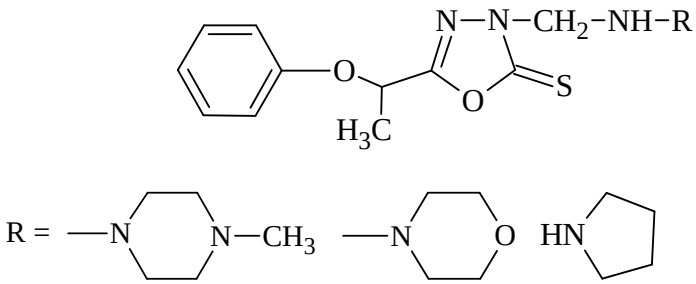
R = 2-NH_2, 2-OH, 4-Cl, 4-NO_2.

Niti Bharadwaj *et al.*, (2009) synthesized indole substituted oxadiazole derivatives and evaluated for their antimicrobial activity on different strains.

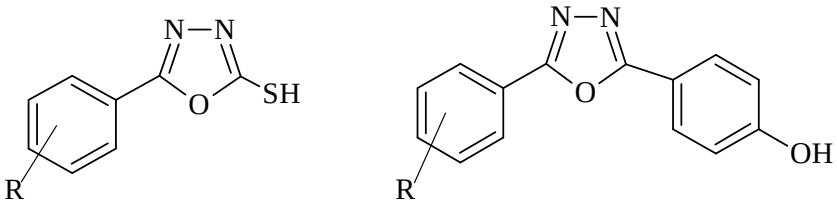


R= H, 2-Cl, 4-OH, C_6H_5.

Sujatha K. *et al.*, (2009) synthesized the new series of 5-(α-aryloxyethyl)-1,3,4-oxadiazole-2-thiones and tested for their antimicrobial activity.

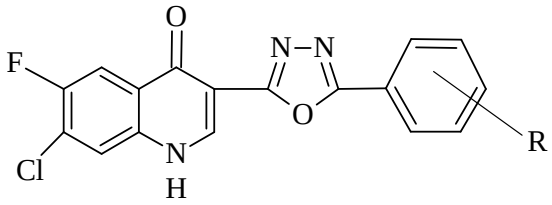


Keshari Kishore Jha *et al.*, (2009) carried out the antimicrobial activity of synthesized 2,5-disubstituted 1,3,4-oxadiazole derivatives.



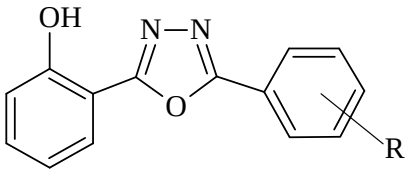
R = H, CH_3, OH, Cl

Shashikant R Pattan *et al.*, (2009) synthesized and evaluated of some new quinoline incorporated oxadiazole derivatives for their antiinflammatory activity.



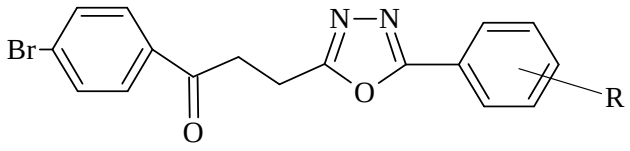
R = H, 2-OH, 4-$N0_2$, 4-NH_2, 4-NH_2-2-OH

Shashikant R Pattan *et al.*, (2009) synthesized and evaluated antitubercular activity of some novel hydroxyl phenyl substituted 1,3,4-oxadiazole derivatives.



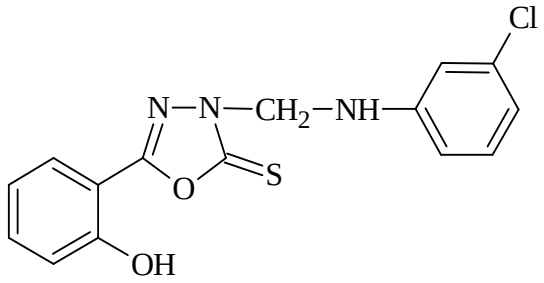
R = H, 2-OH, 2-Cl, 4-OH.

Asif Husain *et al.*, (2009) synthesized some novel 1,3,4-oxadiazole derivatives and evaluated their analgesic, antiinflammatory, ulcerogenic and antibacterial activities.



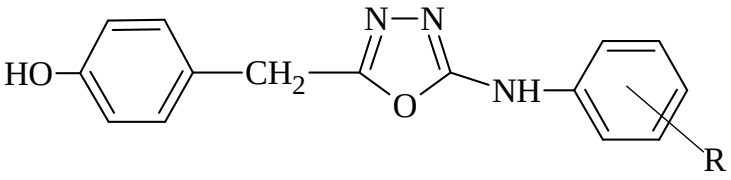
R = 2-OH, 4-F, 4-Cl, 4-CH_3, 4-NO_2, 4-OCH_3, 3,4-$(OCH_3)_2$.

Rakesh Saini *et al.*, (2009) synthesized newer 2,5-disubstituted-1,3,4-oxadiazole derivatives and evaluated their antimicrobial activity.



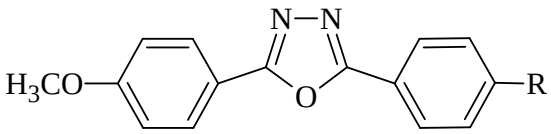
R = H, 2-Cl, 4-Cl, 4-NH_2.

Mohammad Amir *et al.*, (2008) synthesized some newer analogues of 4-Hydroxy phenyl acetic acid and evaluated antiinflammatory, ulcerogenic and lipidperoxidation activities.



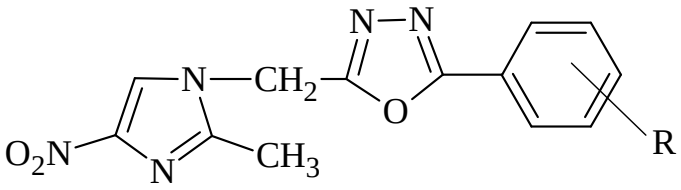
R = H, 2-Cl, 2-CH_3, 4-Cl, 4-Br, 4-F, 4-CH_3, 4-OCH_3.

Nagalakshmi G. *et al.*, (2008) studied the antimicrobial and antiinflammatory activities of some novel synthesized 2,5-disustituted-1,3,4-oxadiazole derivatives.



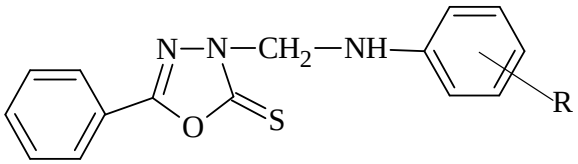
R = CH_3, NH_2, NO_2, OH.

Priya V Frank *et al.*, (2007) compared microwave assisted and conventional synthesis of nitro imidazole moiety containing oxadiazole derivatives and screened their antibacterial, antifungal, antiinflammatory activities.



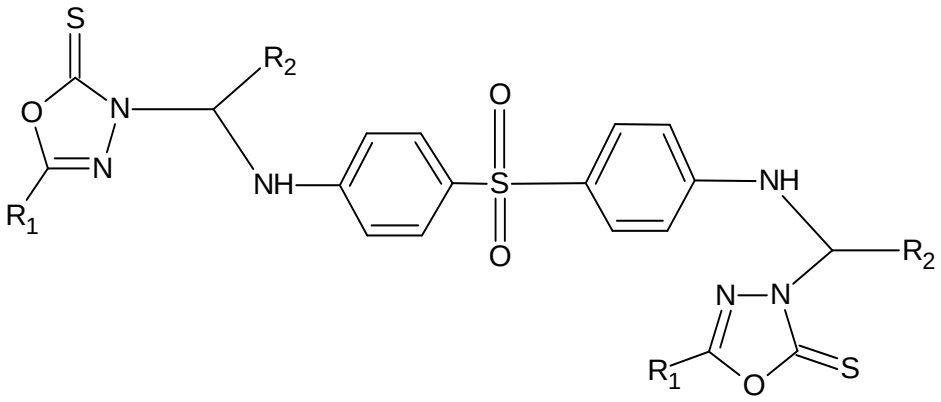
R = H, 2-CH_3, 4-CH_3, 4-Cl, 4-OCH_3.

Gheorghe R. *et al.*, (2007) synthesized some new mannich bases derived from 5-Phenyl-1,3,4-oxadiazole-2-thione and investigated their conformational isomers.

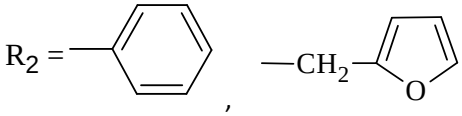


R = 4-OCH_3, 2,4,5$(Cl)_3$, 3,4$(Cl)_2$.

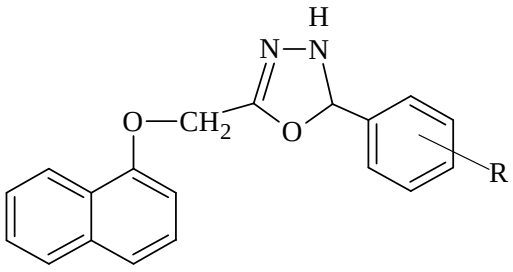
Mohamed Shaharyar *et al.*, (2007) synthesized oxadiazole mannich bases and evaluated their antimycobacterial activity against *M.tuberculosis* $H_{37}RV$ and INH resistant *M.tuberculosis*.

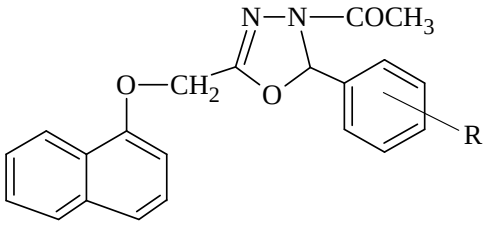


$R_1 = C_6H_5$, 4-NO_2- C_6H_5, C_6H_5-NH-C_6H_5, $C_6H_5OCH_2$, $C_6H_5CH_2$.



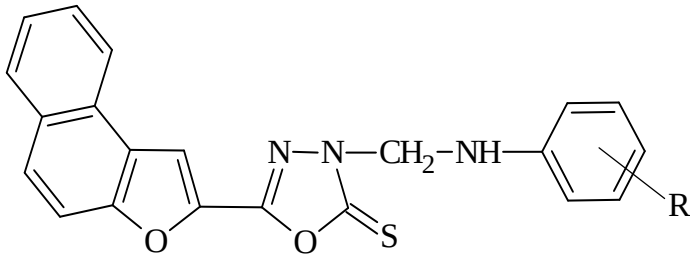
Harish R. *et al.*, (2007)synthesized some novel oxadiazole and oxadiazoline analoges and evaluated their antiinflammatory activity by carregenan induced rat paw edema method and cotton pellets-induced granuloma method.





R = 4-Cl, 4-OH, 4-NO_2, 4-CH_3, 4-OCH_3, 3,4-$(OCH_3)_2$.

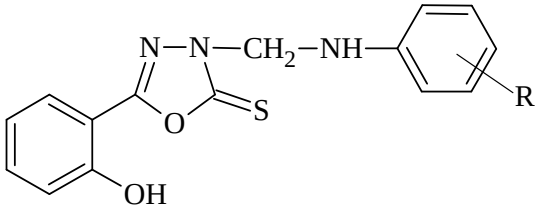
Ravindra K. C. ***et al.*****, (2006)** synthesized and evaluated their antimicrobial and antiinflammatory activities of 1,3,4-oxadiazoles linked to naptho [2,1-d]furan.



R = H, 2-Cl, 2-NO_2, 2-CH_3, 2-OCH_3,

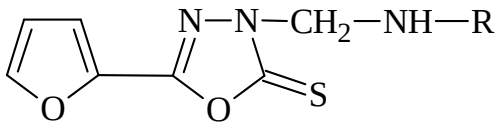
4-Cl, 4-NO_2,4-CH_3, 4-OCH_3.

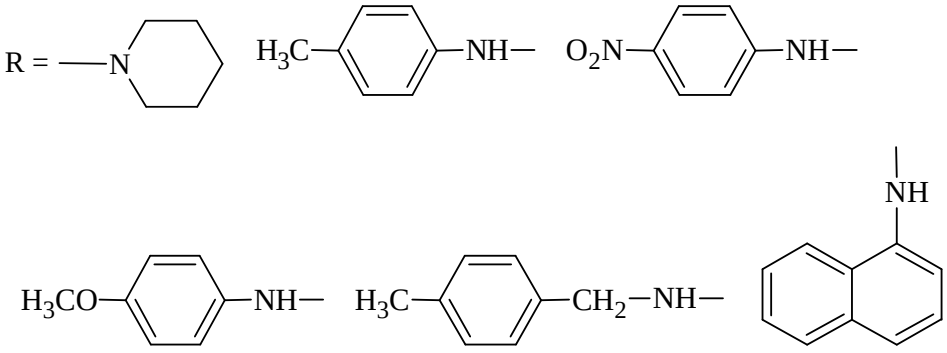
Aboria A. S. ***et al.*****, (2006)** synthesized series of 5-(2-hydroxyphenyl)-3-substituted –2,3-dihydro-1,3,4-oxadiazole-2-thione derivatives were evaluated for their *in-vitro* anticancer activity.



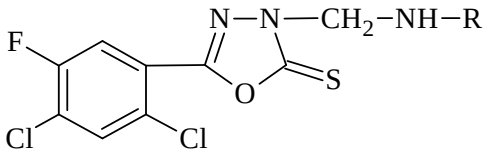
R = 2-Cl, 2-OH, 2-NO_2, 4-Cl, 4-OH.

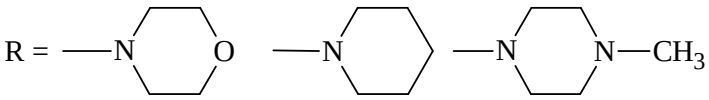
Koparir M. *et al.*, (2005) synthesized mannich bases of 5-Furan-2-yl-[1,3,4] oxadiazol-2-thiol and described their thiol-thione tautomerism. The structures were confirmed by elemental analysis.

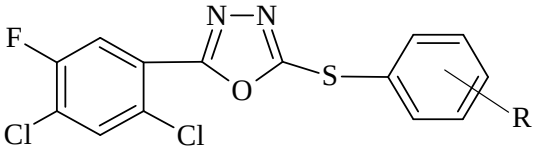




Subrahmanya Bhat K. *et al.*, (2004) synthesized some new fluorine containing 1,3,4-oxadiazole derivatives and screened for antibacterial and anticancer activities.

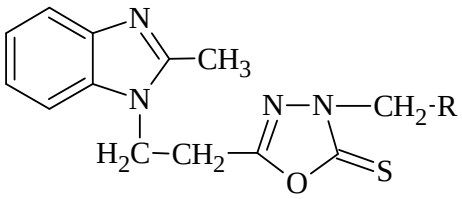


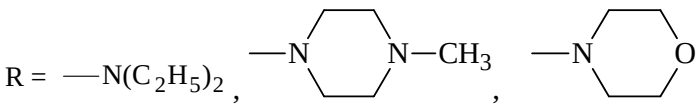




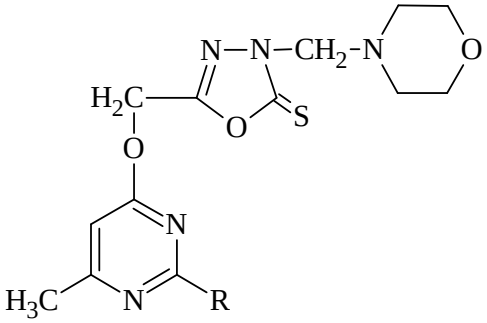
R = H, 4-Cl, 4-OCH_3, 2,4-$(Cl)_2$, 2-Cl-4-OCH_3

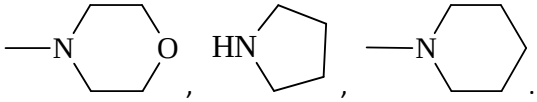
Afaf H. et al., (2005) synthesized and screened their antimicrobial activity of some new benzimidazole incorporated with oxadiazole derivatives.



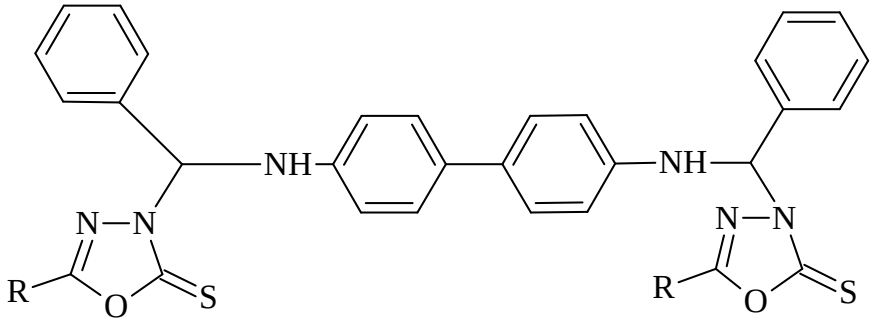


Virginija Jabukiene *et al.*, (2003) reported the antiinflammatory activity of synthesized 5-(6-methyl-2-substituted-4-pyrimidinyloxymethyl)-1,3,4-oxadiazole-2-thione and their 3-morpholino methyl derivatives.



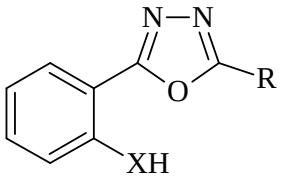
R= N-$(CH_3)_2$, $NHCH_2C_6H_5$, 

Feray Aydogan *et al.*, (2002) synthesized new aryl- and alkyl-substituted 1,3,4-oxadiazole-2- thione derivatives and tested for their antibacterial and antitubercular activities.



R = OCH_3, CH_2Cl, OC_2H_5, CH_2COCH_3, $CH_2COC_6H_5$, $COOC_2H_5$, CH_2COCH_3, C_6H_5.

Kee Jung Lee *et al.*, (2001) synthesized 1,3,4-oxadiazoles containing phenolic and thiophenolic group by appel's dehydration condition and the structures are confirmed by spectral studies.

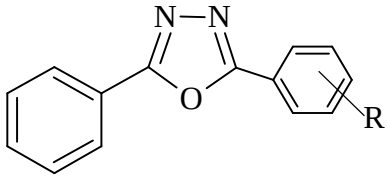


X = O or S

R = CH_3, CH_2Cl, $COOC_2H_5$, C_6H_5, C_6H_5NH,

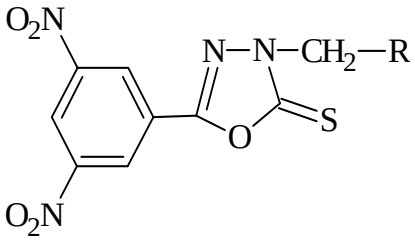
4-ClC_6H_4,4$MeOC_6H_4$.

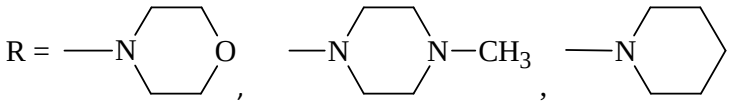
Girges M. M. *et al.*, (1994) carried out the synthesis of 2,5-disubstituted -1,3,4-oxadiazoles derivatives with different substituents and evaluated for their hypoglycemic activity.



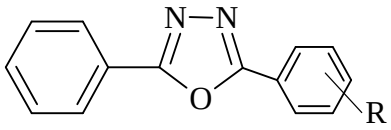
R = H, 2-CH_3, 2-OCH_3, 2-Cl, 4-CH_3, 4-Cl, 4-OCH_3.

Nigam R. *et al.*, (1992) synthesised some new 2-thio-3-(substitued-aminomethyl)-5-(3,5- dinitrophenyl)-1,3,4-oxadiazoles posses considerable antiinflammatory property.



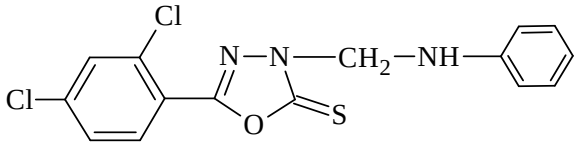


Kumar A. *et al.*, (1987) reported the potent antiinflammatory activity of 2-(substituted aryl)-5- (substituted phenyl)1,3,4-oxadiazoles.

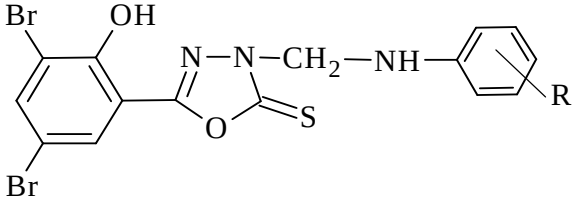


R = H, 2-OH, 2-Cl, 4-OH.

Nath S. C. *et al.*, (1984) synthesized new 3-arylaminomethyl-5-(2,4-dichlorphenyl)-1,3,4-oxadiazole-2-thiones and 3-alkyl-5-(2,4-dichlorophenyl)-1,3,4-oxadiazole-2-thiones for their fungicidial activity against *Curularia verruciformis and Alternaria tenuis.*

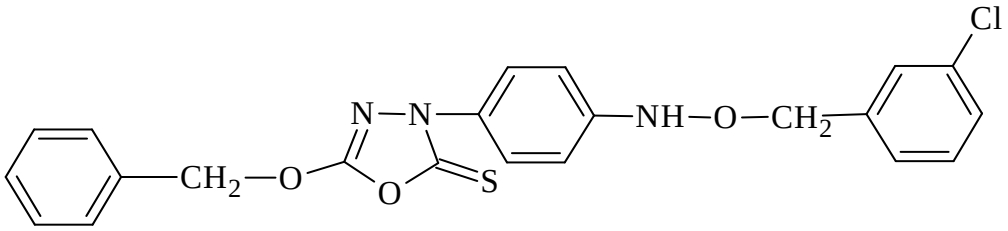


Soni N. *et al.*, (1982) reported the monoamine oxidase, pyruvaate oxidase and succinate dehydrogenase inhibitory properties for some 3-arylaminomethyl-5-(2-hydroxy-3,5-dibromophenyl)-1,3,4-oxadiazole-2-thiones.

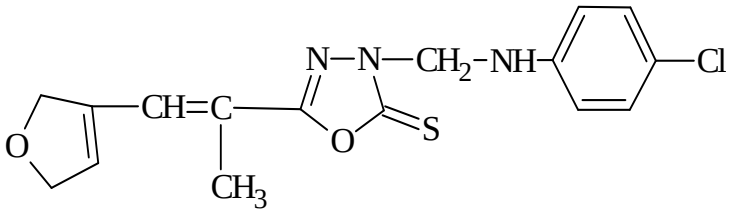


R = 2-Cl, 2-OH, 2-NO_2, 4-Cl, 4-OH, 4-NO_2.

Sengupta A. K. *et al.*, (1979) synthesized mannich bases derived from substituted oxadiazol-2-thiones found to be posses insecticidal activity and antimicrobial properties.



Choudhary S. K., *et al.*, (1978) synthesized some mannich bases of 5-(3,4-methylene dioxyphenyl)-3- arylaminomethyl-1,3,4-oxadiazol-2-thiones for their anticonvulsant activity.



EXPERIMENTAL SECTION

IV. EXPERIMENTAL SECTION

IV. A. MATERIALS AND METHODS

Table-1: List of chemicals and instruments used for the study

S.No	Chemicals / Instruments	Suppliers / Model
	Chemicals	
1	4-Amino benzoic acid	Loba Chem Pvt. Ltd.
2	4-Chloro benzoic acid	Loba Chem Pvt. Ltd.
3	4-Hydroxy benzoic acid	Loba Chem Pvt. Ltd.
4	4-Nitro aniline	Loba Chem Pvt. Ltd.
5	4-Nitro benzoic acid	Loba Chem Pvt. Ltd.
6	Carbondisulphide	Loba Chem Pvt. Ltd.
7	Cinnamic acid	SpectroChem Pvt. Ltd.
8	Con.Sulphuric acid	Qualigens fine Chemicals.
9	Dil.Hydrochloric acid	Qualigens fine Chemicals.
10	Ethyl alcohol	Loba Chem Pvt. Ltd.
11	Formaldehyde	SpectroChem Pvt. Ltd.
12	Hydrazine hydrate	Loba Chem Pvt. Ltd.
13	Nicotinic acid	Loba Chem Pvt. Ltd.
14	Potassium hydroxide	SpectroChem Pvt. Ltd.
15	Phosphorus oxy chloride	Loba Chem Pvt. Ltd.
16	Sodium bicarbonate	Loba Chem Pvt. Ltd.

	Instruments	
17	Heating mantle	Ajay Equipments
18	Hot air oven	Picses Instruments
19	Magnetic stirrer	Remi Equipments
20	Melting point apparatus	Sun bim Equipments
21	Plethysmograph	Remi Equipments
22	Weighing balance	Shzimadzhu 220V

METHODS

Melting points were determined using open capillary tubes and were uncorrected and the purity of the compounds were checked by thin layer chromatography using precoated silica gel plates and iodine vapours as a detecting agent.

The I.R spectra of the synthesized compounds were recorded in JASCO FT-IR spectrophotometer, Ideal analytical and research institution, Puducherry.

The NMR spectra of the synthesized compounds were recorded in BRUKER-500 MHZ NMR spectrophotometer, SAIF, Punjab University, Chandigarh.

The Mass spectra of the synthesized compounds were recorded in JEOL GC mate mass spectrophotometer by Electron impact method as ionization mode, SAIF, IIT, Chennai.

The antiinflammatory activity of the synthesized compounds was evaluated by carragenan induced rat paw edema method.

The antibacterial, antifungal activities were evaluated by disc diffusion method, Pharma analytical lab, Puducherry.

B. SCHEME

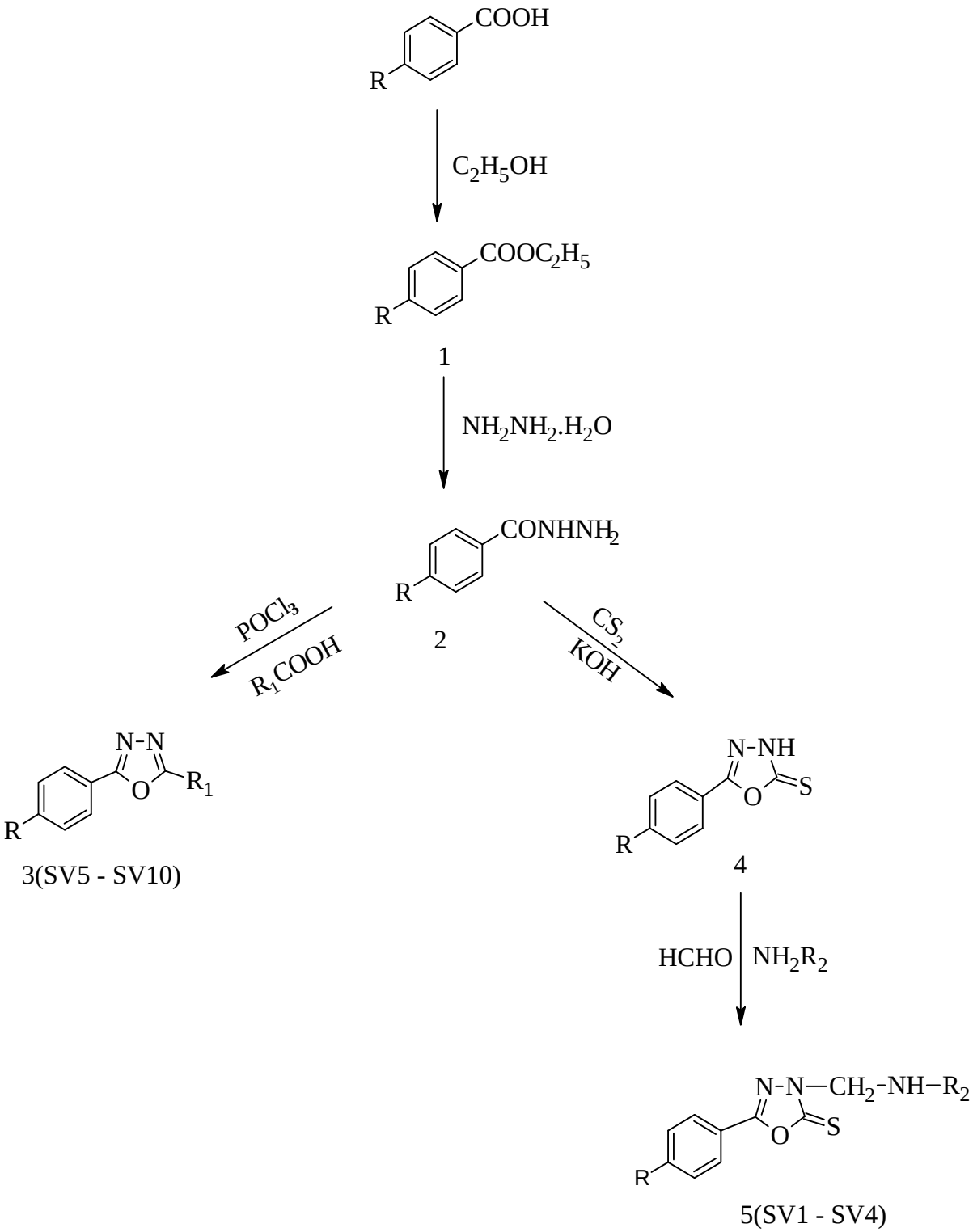
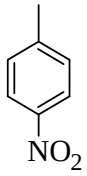
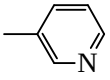
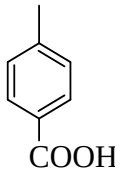
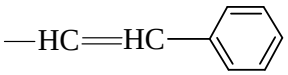
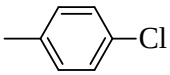
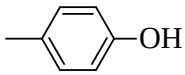


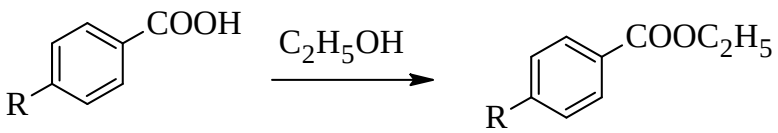
Figure 1: Scheme

CODE	R	R_2	CODE	R	R_1
SV1	Cl		**SV5**	Cl	
			SV6	OH	
SV2	Cl		**SV7**	NO_2	
SV3	NO_2		**SV8**		
			SV9	NO_2	
SV4	OH		**SV10**		

C. METHODOLOGY

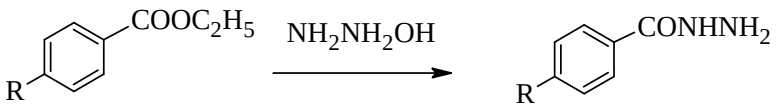
Esterification of aromatic acid (1):

4-Substituted aromatic acid (0.01 mol), ethyl alcohol (60ml), concentrated sulphuric acid (1.5 ml) were taken in a round bottom flask and refluxed for 4 h, the contents were cooled and poured into beaker containing crushed ice. The product was precipitated out. It was filtered, dried and recrystallized from ethanol. The completion of reaction was monitored by thin layer chromatography and chemical test.



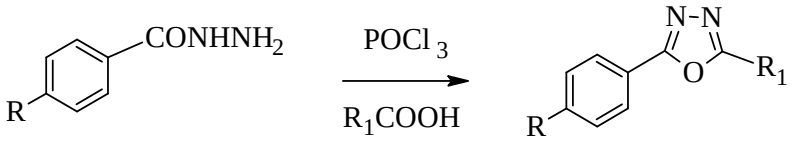
Preparation of acid hydrazides (2):

The 4-substituted ethyl benzoate (1) (0.01 mol), hydrazine hydrate (0.15 mol) and 30ml of ethanol was refluxed for 4 h and the excess of ethanol was distilled off and the contents were poured into a beaker and cooled in an ice bath, during which hydrazides separated out. The resulting solid obtained was filtered, dried and recrystallized from ethanol. (*Illango K. et al., 2010*)



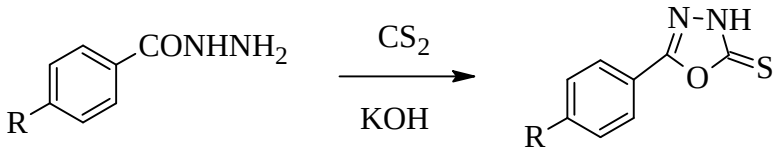
Preparation of 2,5-disubstituted-1,3,4-oxadiazoles (3):

The acid hydrazide (2) (0.01 mol) and an appropriate aromatic acid (0.01 mol) were refluxed in $POCl_3$ (5 ml) for 8 h cooled and poured on to crushed ice, neutralized with sodium bicarbonate solution. The precipitate was filtered off, dried and recrystallized from ethanol. The completion of reaction was monitored by thin layer chromatography. (*Keshari kishore jha et al., 2009*)



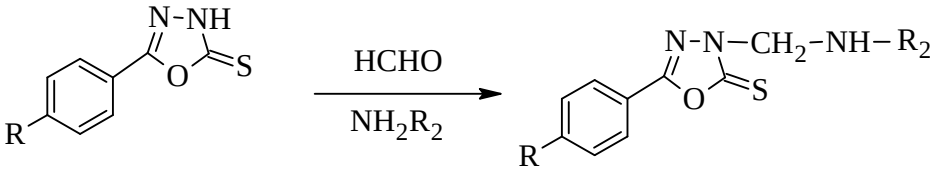
Preparation of 5-substituted-2-thio-1,3,4-oxadiazoles (4):

The acid hydrazides (3) (0.01 mol) in ethanol (15 ml) at 0□ C, carbon di sulphide (2ml) and potassium hydroxide (0.6 g) were added, and the reaction mixture was refluxed until the evolution of H_2S gas ceased (around 12 h). Excess solvents were 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 (*Ravindra K. et al., 2006*)



Preparation of Mannich bases of 1,3,4-oxadiazoles (5):

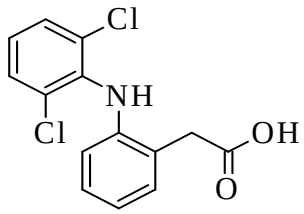
Equimolar quantity of 5-(4-substituted phenyl)-1,3,4-oxadiazol-2(3*H*)-thione (4) (0.0lmol) and appropriate amines were dissolved in ethanol(20ml) and to this add drop by drop of formaldehyde solution (0.0lmol). Stirred the contents for one hour at room temperature and then refluxed for 2-3 h. Then the content was kept at overnight in the refrigerator. The reaction mixture was concentrated and the product was separated out. It is filtered off, dried and recrystallized with ethanol. Practical yield and percentage yields were calculated. The completion of the reaction was monitored by thin layer chromatography. (*Koparir M. et al., 2005*)



D. EVALUATION OF ANTI-INFLAMMATORY ACTIVITY

Antiinflammatory activity of the compounds was evaluated by Carrageenan induced rat paw edema method in rats. A wistar rat of either sex (150-200g) was be divided into control, standard and test groups, each comprising of six rats. The study was approved by the Institutional Animal Ethics Committee (IAEC) (Reg No. 409/01/CPCSEA).

Standard drug



Diclofenac sodium

Freshly prepared suspension of carrageenan (0.1ml, 1% w/v solution in 0.9% saline) will be injected under the planter aponeurosis of the left hind paw of each rat. One group kept as control, and the animals of the test and standard groups pretreated with the test and standard drugs suspended in 1% carboxymethylcellulose (CMC) given orally 30 min before carrageenan injection. The standard group receives 20mg/kg body weight of as standard diclofenac sodium drug, test groups receives 50mg/kg body weight of the synthesized compounds (SV3 and SV4) suspended in 1% carboxymethyl cellulose (CMC) and one group as control.

The paw volume was measured using the mercury displacement technique, with the help of plethysmograph in control, test, standard groups of animals, before and after 3 h of carageenan injection. The percentage inhibition of inflammation will be calculated using the formula,

Percentage inhibition = (1-Vt/Vc) X 100

Where, Vt and Vc are the mean relative changes in the volume of paw edema in the test and control respectively. (*Harish Rajak et al., 2007*)

E. EVALUATION OF ANTIBACTERIAL ACTIVITY:

The antibacterial activity can be evaluated by the following techniques.

A) Agar streak dilution method

B) Serial dilution method

C) Agar diffusion method

i) Cup plate method

ii) Cylinder method

iii) Paper disc method

D) Turbidimetry method.

The antibacterial activity of synthesized compounds were screened in the concentration of 50, 100, and 150 μg/ml in dimethyl formamide against gram positive *Bacillus cereus* and gram negative *Pseudomonas aeruginosa* in Muller Hinton agar medium by disc diffusion method.

Preparation of Muller Hinton Agar

Composition

Beef extract	10.0 g
Casein acid hydrolysis	17.5 g
Starch	1.5 g
Agar	20.0 g
Distilled water	1000 ml

All the ingredients are taken in 1000 ml of distilled water in a conical flask and heated in a steam bath to dissolve. The pH was adjusted to 7.0 ± 0.2 and sterilized in autoclave at 15 lb at 120^0C for 15 minutes. The sterile medium was poured into Petridishes and allowed to solidify.

Preparation of the Disk

Paper disk of 5 mm diameter and 2 mm thickness were sterilized by autoclaving at 121^0C for 15 minutes. Ciprofloxacin (10 µg/ml) was used as standard antibiotic for the comparison of antibacterial activity of the synthesized compounds.

Organism used:

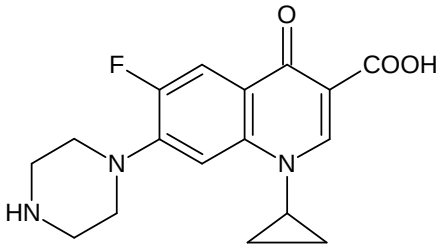
Gram positive Organisms

Bacillus cereus ATCC 11778

Gram negative Organisms

Pseudomonus aeruginosa ATCC 9027

Standard drug



Ciprofloxacin

Procedure for antibacterial activity

A suspension of the organism was added to sterile muller hinton agar medium at 45^oC. The mixture was transferred to sterile petridishes and allowed to solidify. Sterile disc 5mm in diameter was dipped in solution of different concentrations of test compounds, standard and control and they were placed on the surface of agar plates.

Left 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 the different solutions. Then the plates were incubated for 24h at 37 ^{o}C ± 1^oC and observed for antibacterial activity. The diameter of zone of inhibition was observed and recorded.

F. EVALUATION OF ANTIFUNGAL ACTIVITY:

The antifungal activity can be evaluated by the following techniques:

1. Cup and plate method/cylinder method
2. Turbidimetry / tube assay method

Organism used:

Aspergillus fumigates ATCC 46645

The antifungal activity of synthesized compounds was screened in the concentration of 50, 100, and 150 μg / ml in dimethyl formamide against *Aspergillus fumigates*. The antifungal activity was evaluated by measuring zone of inhibition in mm, details of the procedure are given below.

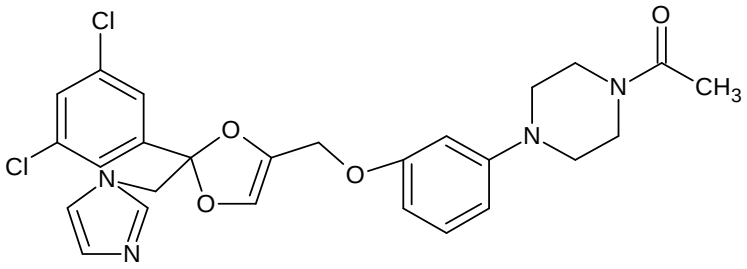
Preparation of Sabouraud's Agar Media

Composition

Dextrose	20 g
Peptone	10 g
Purified water	1000 ml
pH	5.4± 0.2
Agar	15 g

The media was prepared by dissolving the specified quantities of the dehydrated ingredients (Hi-media) in purified water and was distributed in petridishes to a thickness of 3-4 mm. The plates sterilized by autoclaving at 121^{o}C for 15 minutes. The sterile medium was poured into petridishes and allowed to solidify.

Standard drug



Ketaconazole

Procedure for antifungal activity

A suspension of the organism was added to sterile sabouraud's agar medium at 45^{o}C. The mixture was transferred to sterile petri dishes and allowed to solidify. Sterile discs 5 mm in diameter was dipped in solution of different concentration of test compounds, standard and control and they were placed on the surface of agar plates.

Left the plates to stand for 1 h at room temperature as a period of pre incubation diffusion to minimize the effects of variation in time between the applications of the different solutions. Then the plates were incubated for 48 h at 37^{o}C ±1^{o}C and observed for antifungal activity. The diameter of zone of inhibition were observed and recorded.

RESULTS AND DISCUSSION

V. RESULTS AND DISCUSSION

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 satisfactory yield was obtained for every reactions.

The completion of reaction was monitored by TLC using silica gel as stationary phase and ethylacetate - hexane as mobile phase. The spot in the TLC plate was detected by iodine vapours. The structures of the synthesized compounds were consistent with IR, ^{1}H NMR and MASS spectra.

A. PHYSICAL DATA OF THE COMPOUNDS

(i) Physical Data of Esters

The different 4-substituted aromatic acids were converted to their esters by esterification with ethanol. The physical data of these compounds are as follows.

Table-2

S. No	Esters	Molecular Formula	Molecular Weight	Physical State	M.P (^{0}C)	Yield (%)
1	4-Chloro Ethylbenzoate	$C_9H_9O_2Cl$	184.62	White powder	156	85.63
2	4-Nitro Ethylbenzoate	$C_9H_9NO_4$	195.17	Yellow powder	58	88.50
3	4-Hydroxy Ethylbenzoate	$C_9H_{10}O_3$	166.17	White powder	106	75.85

(ii) Physical Data of Aryl hydrazides

The esters were made to react with hydrazine hydrate in ethanol to get corresponding aryl hydrazides and the yields were calculated. The physical datas are

Table-3

S. No	Aryl hydrazides	Molecular Formula	Molecular Weight	Physical State	M.P (^{0}C)	Yield (%)
1	4-Chloro benzohydrazide	$C_7H_7N_2OCl$	170.59	Pale Yellow Powder	155	57.35
2	4-Nitro benzohydrazide	$C_7H_7N_3O_3$	181.14	Yellow powder	210	66.54
3	4-Hydroxy benzohydrazide	$C_7H_8N_2O_2$	152.15	White Powder	234	60.12

(iii) Physical Data of 5-aryl substituted-2-thio-1,3,4-oxadiazoles

The aryl hydrazides react with carbondisulphide and potassium hydroxide to get 5-aryl substituted-2-thio-1,3,4-oxadiazole by cyclisation mechanisms and the yields were calculated. The Physical data of 5- aryl substituted-2-thio-1,3,4-oxadiazoles are given below.

Table-4

S. No	5-aryl substituted -2-thio-1,3,4-oxadiazoles	Molecular Formula	Molecular Weight	Physical State	M.P (^{0}C)	Yield (%)
1	5-(4-Chlorophenyl) -2- thio-1,3,4-oxadiazole	$C_8H_5N_2OSCl$	212.65	Light Yellow powder	160	87.26
2	5-(4-Nitrophenyl) -2- thio-1,3,4-oxadiazole	$C_8H_5N_3O_3S$	223.20	Yellow powder	170	82.19
3	5-(4-Hydroxyphenyl) -2- thio-1,3,4-oxadiazole	$C_8H_6N_2O_2S$	194.21	White powder	182	53.57

(iv) Physical Data of Derivatives (SV1 - SV10)

The derivatives (SV-1 to SV-4) were synthesized by mannich reaction, the formaldehyde and appropriate amines was reacted with 5-aryl substituted-2-thio-1,3,4-oxadiazole to get the mannich bases of 1,3,4-oxadiazoles.

The derivatives (SV-5 to SV-10) were synthesized by condensing the aryl hydrazides and aromatic acids with $POCl_3$. The physical data of these compounds are

Table-5

Code	Molecular Formula	Molecular Weight	Yield (%)	Melting Point (□C)	*R_f value
SV-1	$C_{15}H_{11}N_4O_3SCl$	362.79	91.16	175	0.79
SV-2	$C_{16}H_{12}N_3O_3SCl$	361.80	78.45	180	0.58
SV-3	$C_{16}H_{12}N_4O_5S$	372.35	92.21	189	0.38
SV-4	$C_{16}H_{13}N_3O_4S$	343.35	86.87	196	0.56
SV-5	$C_{13}H_8N_3OCl$	257.67	70.81	162	0.5
SV-6	$C_{13}H_9N_3O_2$	239.22	63.02	186	0.55
SV-7	$C_{13}H_8N_4O_3$	268.22	73.30	210	0.53
SV-8	$C_{14}H_{11}N_3O_3$	293.27	76.79	293	0.59
SV-9	$C_{14}H_8N_3O_3Cl$	301.68	77.07	190	0.46
SV-10	$C_{14}H_9N_30_4$	283.23	65.37	205	0.35

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

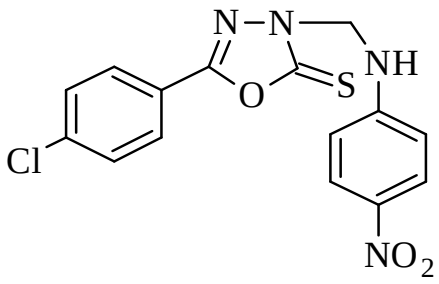
B. SPECTRAL ANALYSIS

The synthesized compounds were characterized by various methods such as IR, ^{1}H NMR and Mass spectroscopy.

i) Infra Red Spectral Analysis

The IR spectrum was recorded in JASCO FT-IR spectrophotometer. The significant IR values are measured in cm^{-1} and the results are given in the table.

Interpretation of IR spectrum of SV-1



The significant wave numbers of the compound and its functional groups are given below**.**

Table -6

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3366.10	N-H Stretching
2	1603.53	C-C Stretching in Aromatic ring
3	1488.21	C=C & C=N Stretching
4	1560.61,1531.18	N=O &C=N Stretching (Aromatic Nitro Group)
5	1329.29	CH_2 Stretching
6	1258.31	C=S Stretching
7	1111.69	C-O-C Stretching
8	752.59	C-Cl Stretching

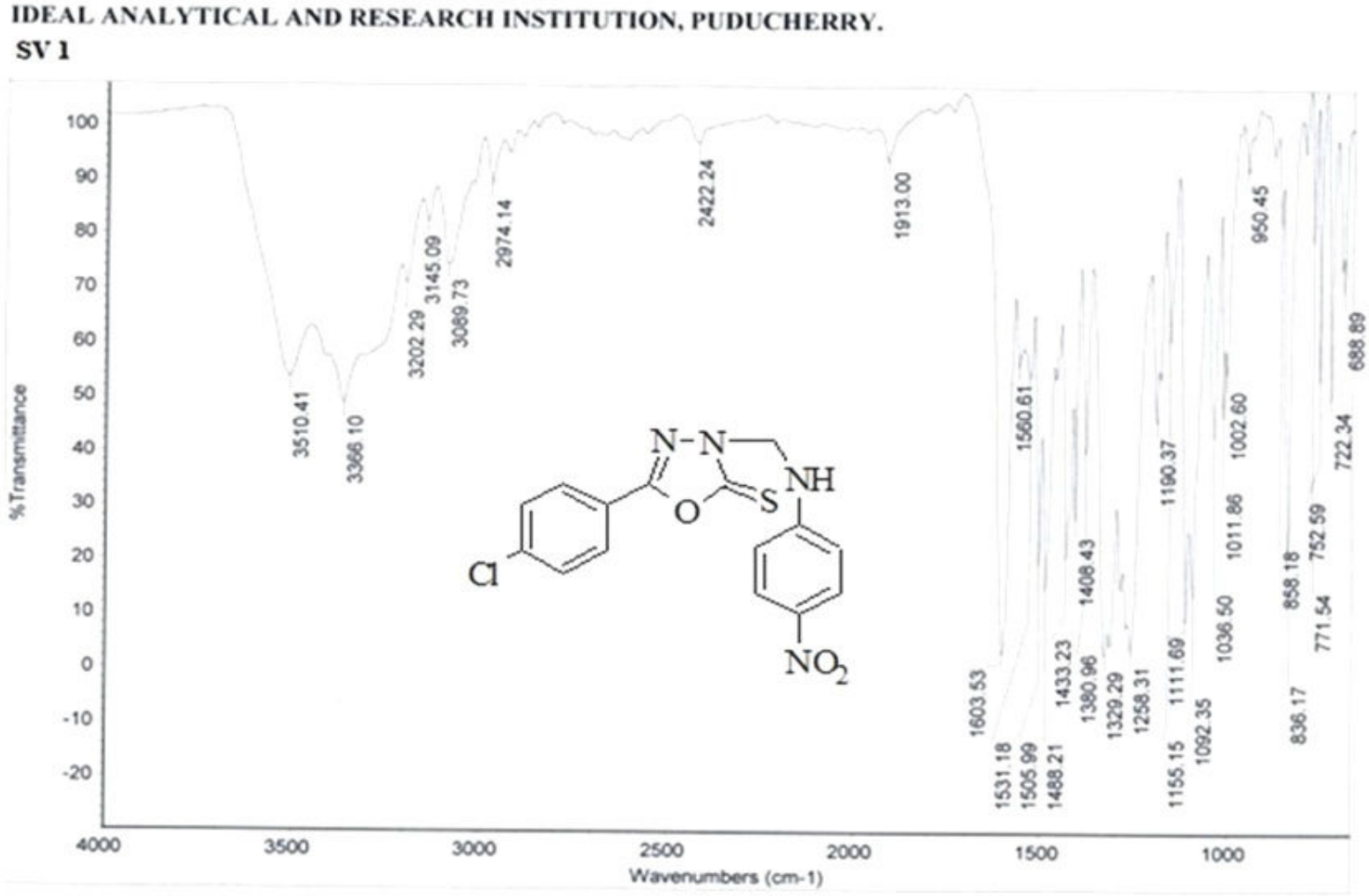
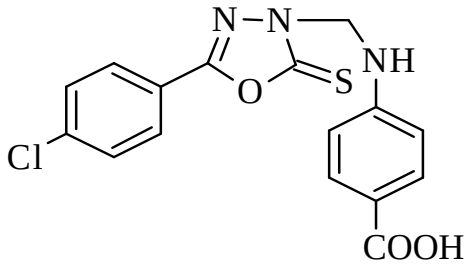


Figure 2: IR SPECTRA OF SV-1

Interpretation of IR spectrum of SV-2



The significant wave numbers of the compound and its functional groups are given below**.**

Table -7

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3316.90	N-H Stretching
2	3027.53	C-H Stretching in Aromatic Group
3	2666.37,2551.30	OH Plane bending & C-OStretching
4	1606.74	C-C Stretching in Aromatic ring
5	1490.16	C=C & C=N Stretching
6	1319.22	CH_2 Stretching
7	1253.83	C=S Stretching
8	1079.07	C-O-C Stretching
9	770.70	C-Cl Stretching

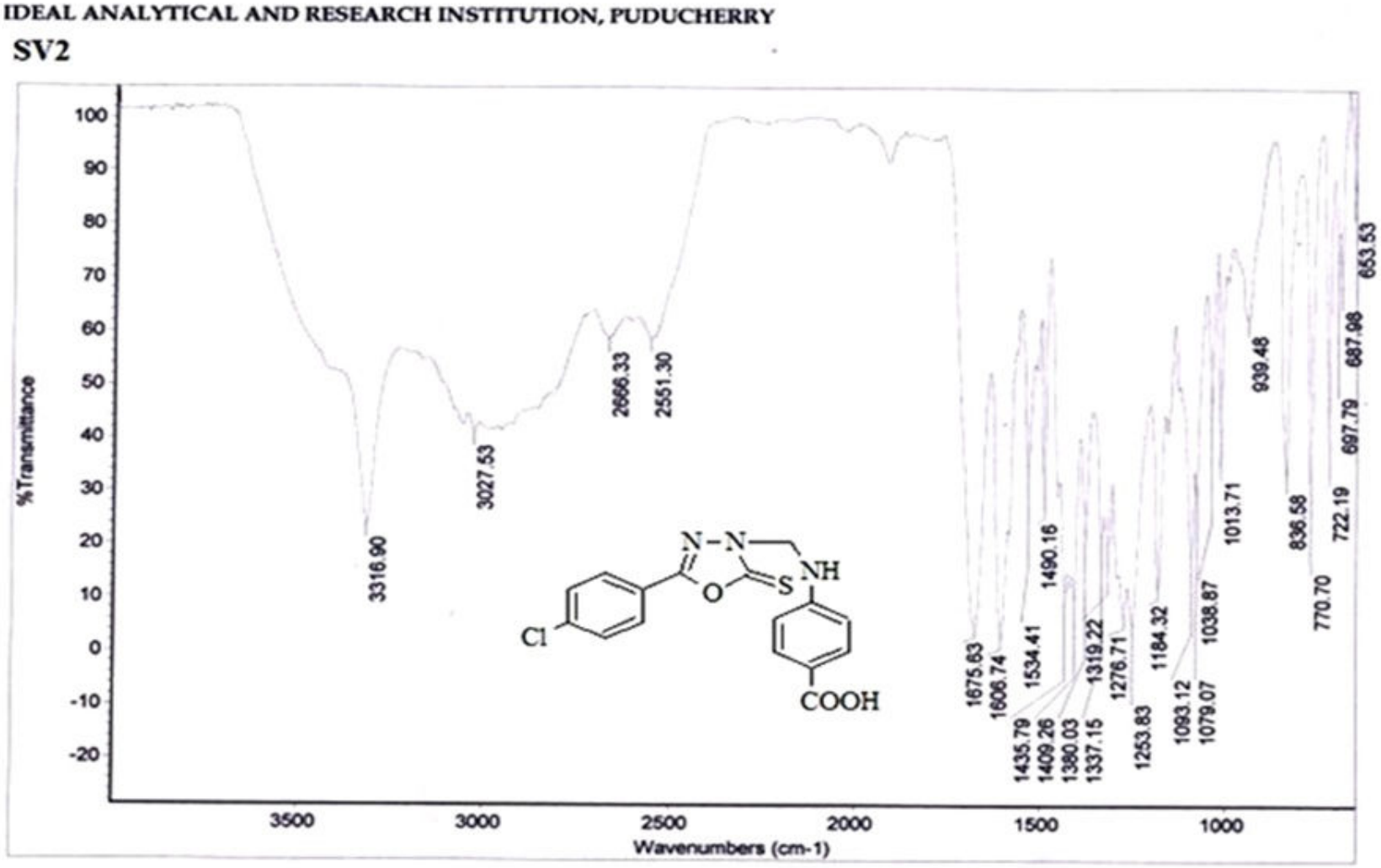
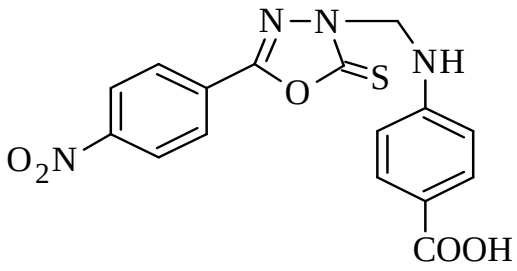


Figure 3: IR SPECTRA OF SV-2

Interpretation of IR spectrum of SV-3



The significant wave numbers of the compound and its functional groups are given below**.**

Table -8

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3423.41	N-H Stretching
2	3054.28	C-H Stretching in Aromatic Group
3	1604.98	C-C Stretching in Aromatic ring
4	1526.19	N=O &C=N Stretching (Aromatic Nitro Group)
5	1318.86	CH_2 Stretching
6	1255.17	C=S Stretching
7	1111.19	C-O-C Stretching

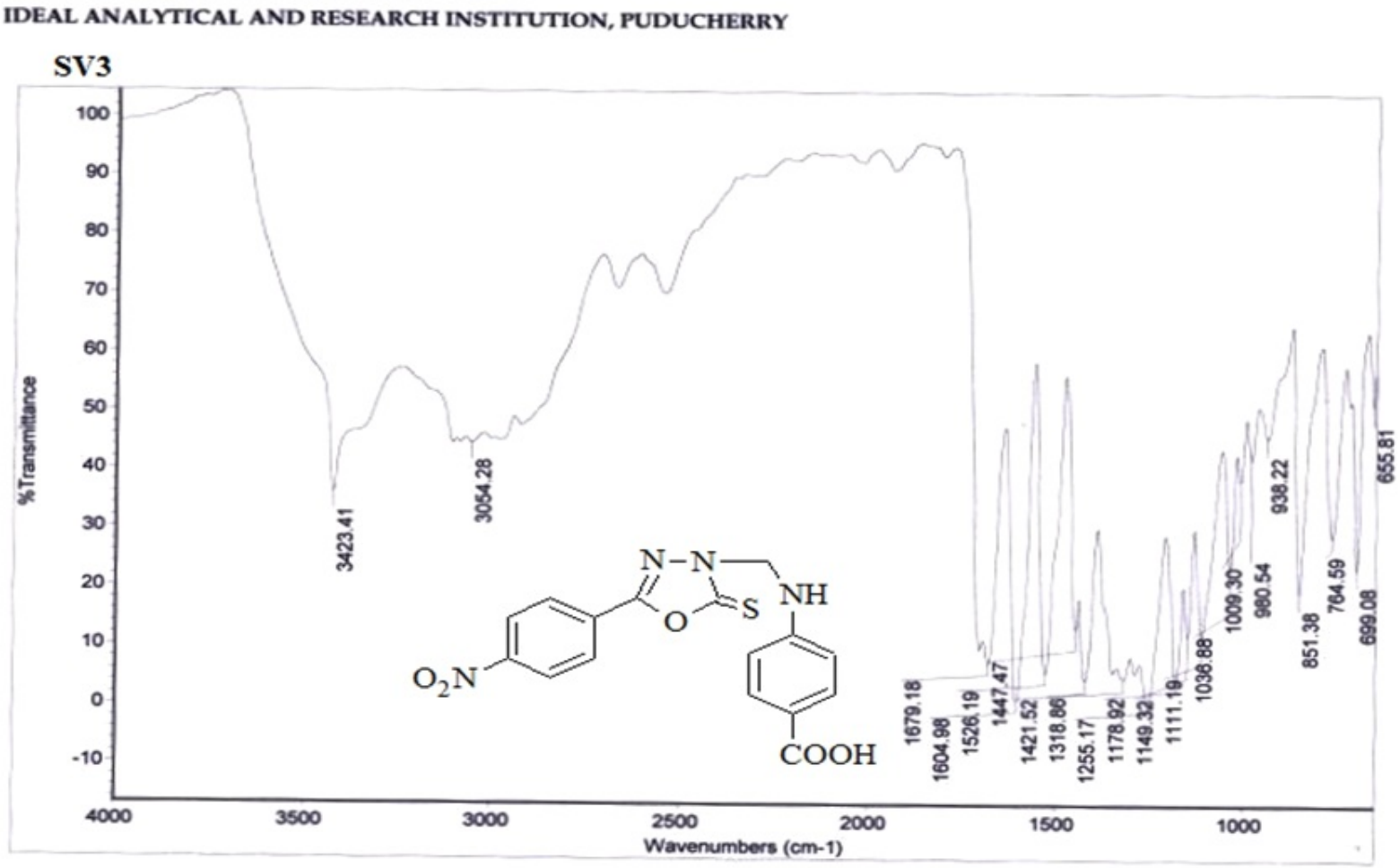
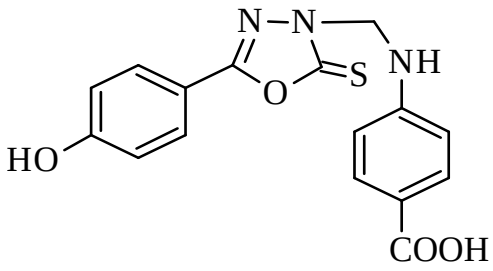


Figure 4: IR SPECTRA OF SV-3

Interpretation of IR spectrum of SV-4



The significant wave numbers of the compound and its functional groups are given below**.**

Table -9

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3298.94	O-H Stretching
2	1605.71	C-C Stretching in Aromatic ring
3	1531.94	C=C & C=N Stretching
4	1338.40	CH_2 Stretching
5	1280.14	C=S Stretching
6	1104.80	C-O Stretching (2° Alcohol)
7	1076.66	C-O-C Stretching

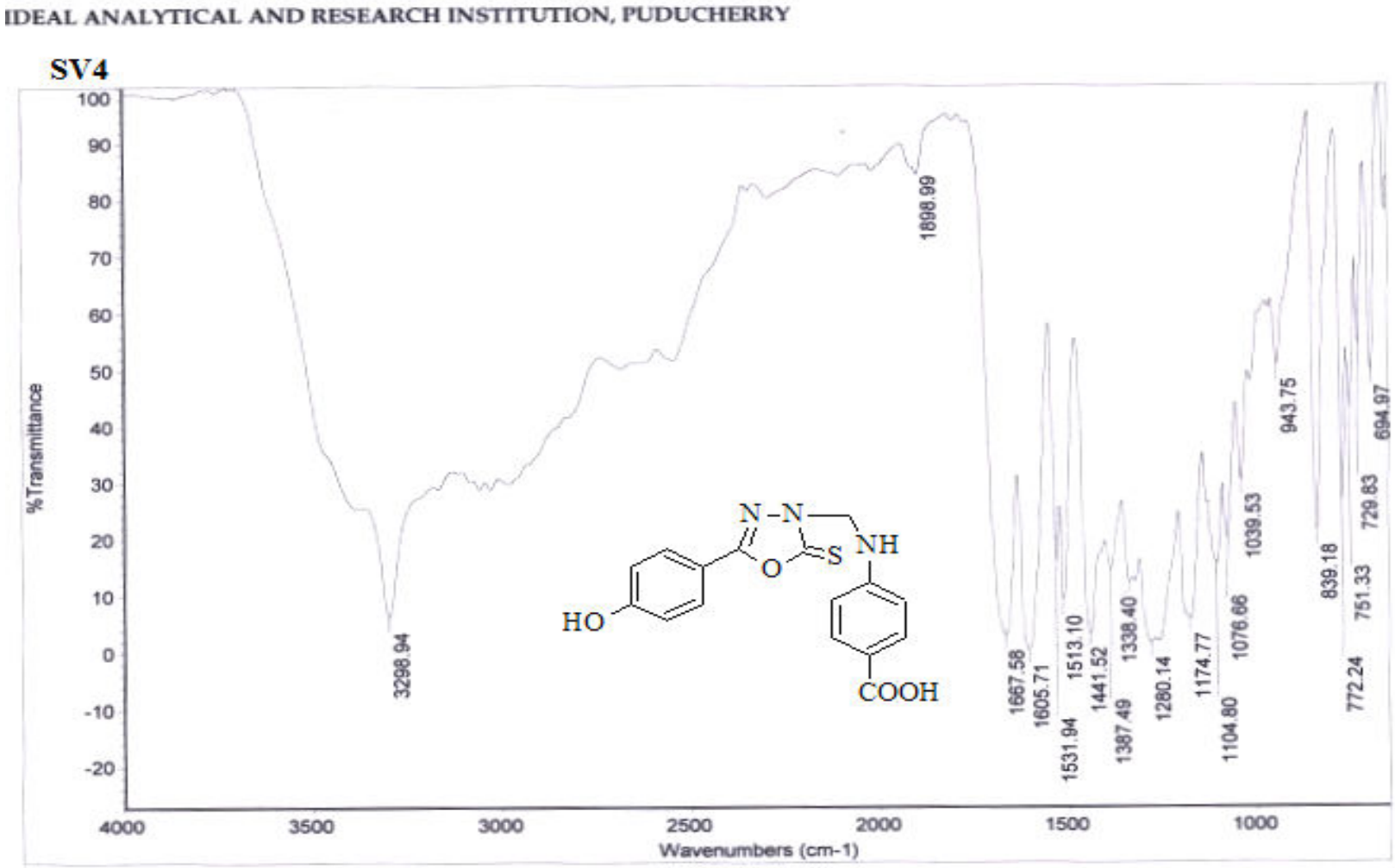
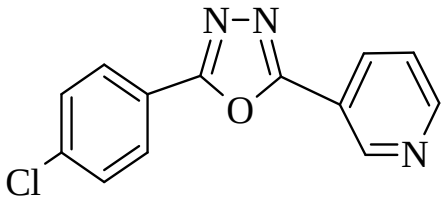


Figure 5: IR SPECTRA OF SV-4

Interpretation of IR spectrum of SV-5



The significant wave numbers of the compound and its functional groups are given below**.**

Table -10

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3417.14	N-H Stretching such as Furan,Thiophene,Pyridine
2	3060.55	C-H Stretching in Aromatic Group
3	1581.73	C=C & C=N Stretching
4	1326.18	C-N Stretching (Aromatic Teritary)
5	1106.94	C-O-C Stretching
6	1073.73	C-O Stretching
7	735.76	C-Cl Stretching

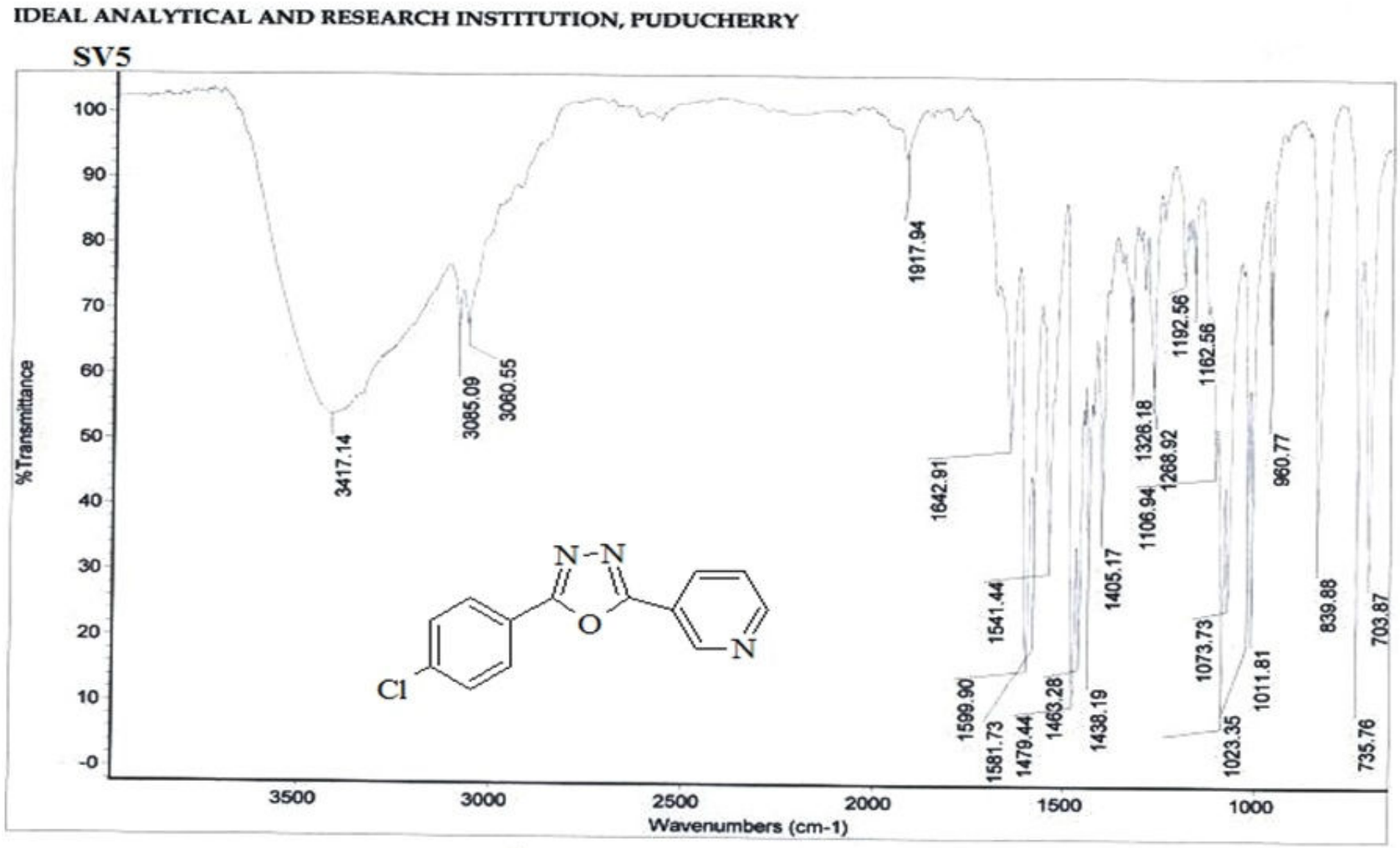
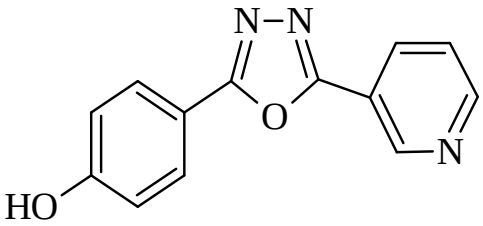


Figure 6: IR SPECTRA OF SV-5

Interpretation of IR spectrum of SV-6



The significant wave numbers of the compound and its functional groups are given below**.**

Table -11

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3417.82	N-H Stretching such as Furan,Thiophene,Pyridine
2	3052.32	C-H Stretching in Aromatic Group
3	2923.95,2844.75	O-H Stretching
4	1599.46	C=C Stretching in Aromatic Hydrocarbon
5	1575.61,1494.93	C=N stretching
6	1096.86	C-O-C Stretching
7	1074.56	C-O Stretching

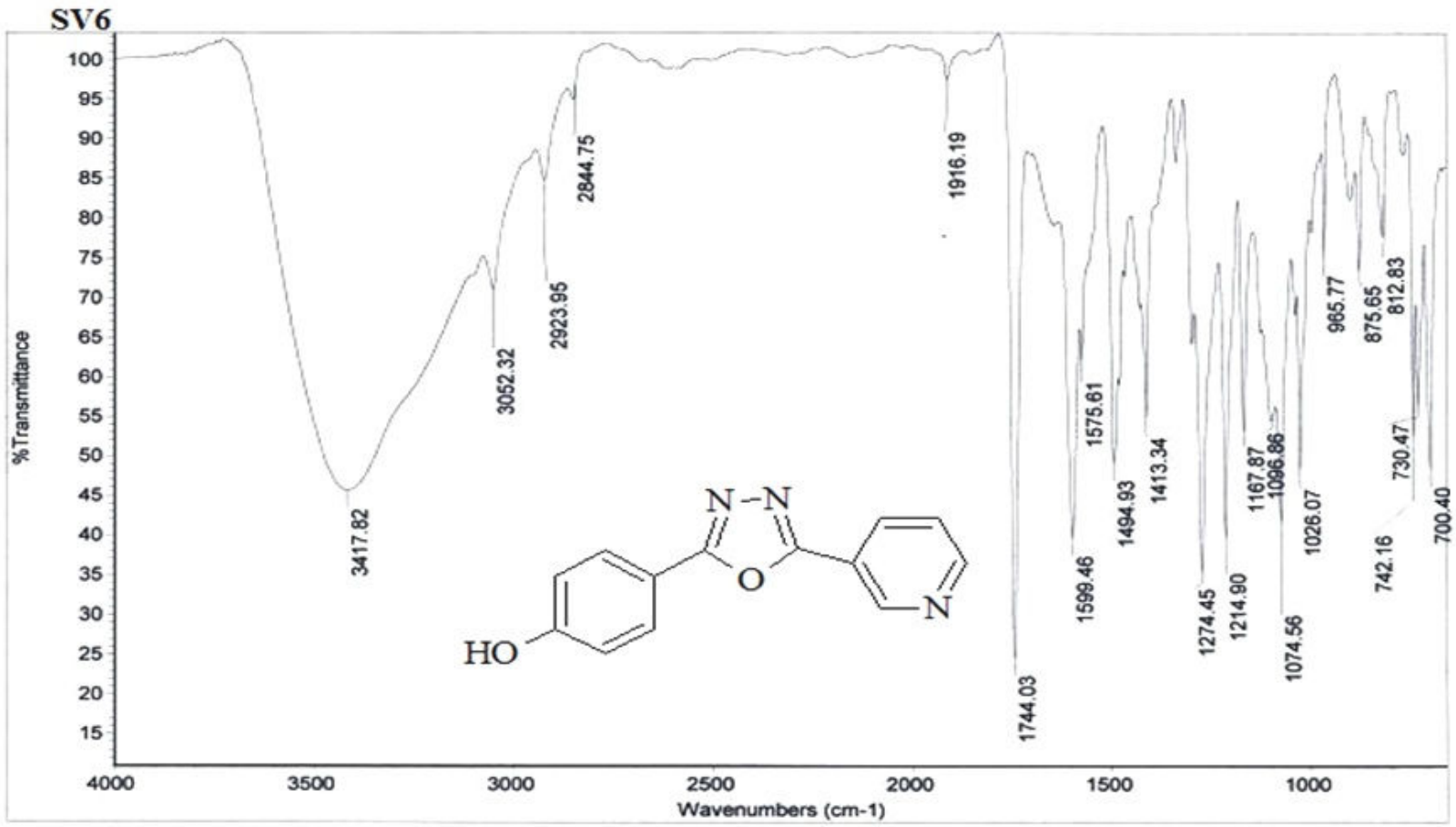
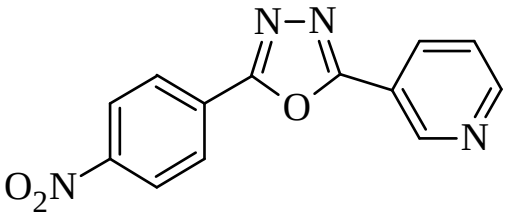


Figure 7: IR SPECTRA OF SV-6

Interpretation of IR spectrum of SV-7



The significant wave numbers of the compound and its functional groups are given below**.**

Table -12

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3109.67	N-H Stretching such as Furan,Thiophene,Pyridine
2	3063.24	C-H Stretching in Aromatic Group
3	1599.72	C=C Stretching in Aromatic Hydrocarbon
4	1552.81	N=O & C=N Stretching (Aromatic Nitro Group)
5	1481.84	C=C & C=N Stretching
6	1109.62	C-O-C Stretching
7	1085.23	C-O Stretching

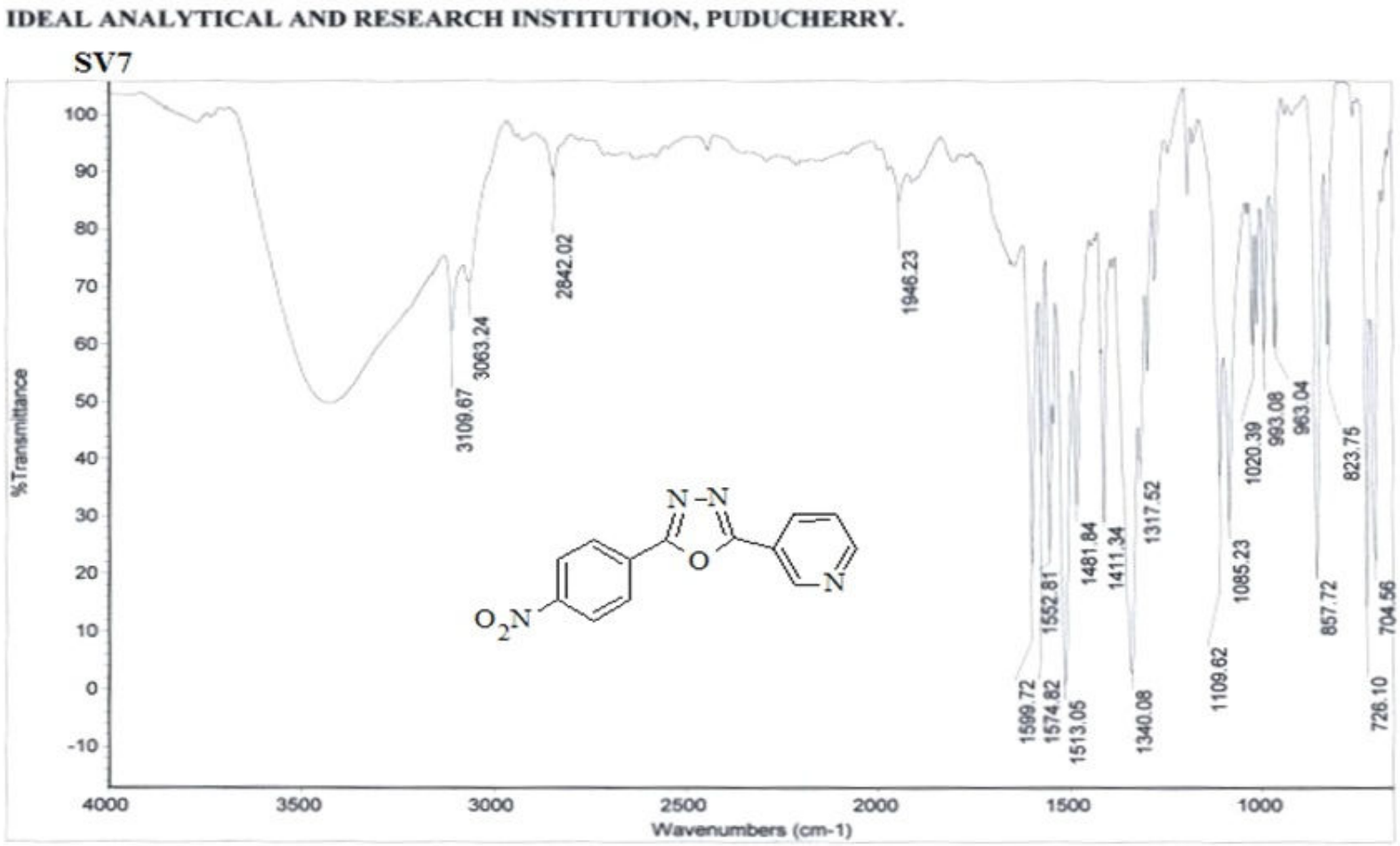
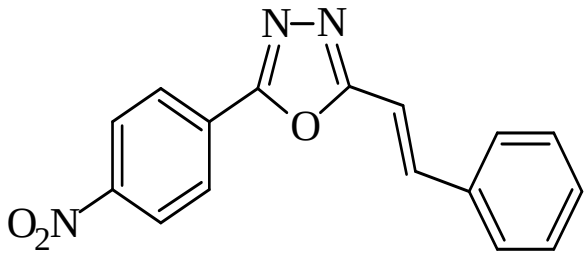


Figure 8: IR SPECTRA OF SV-7

Interpretation of IR spectrum of SV-8



The significant wave numbers of the compound and its functional groups are given below**.**

Table -13

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3447.20	N-H Stretching
2	1642.85,1606.49	C=C Stretching Non Conjugated diene
3	1551.76	N=O & C=N Stretching (Aromatic Nitro Group)
4	1578.82,1483.24	C=C & C=N Stretching
5	1086.52	C-O-C Stretching
6	1016.92	C-O Stretching

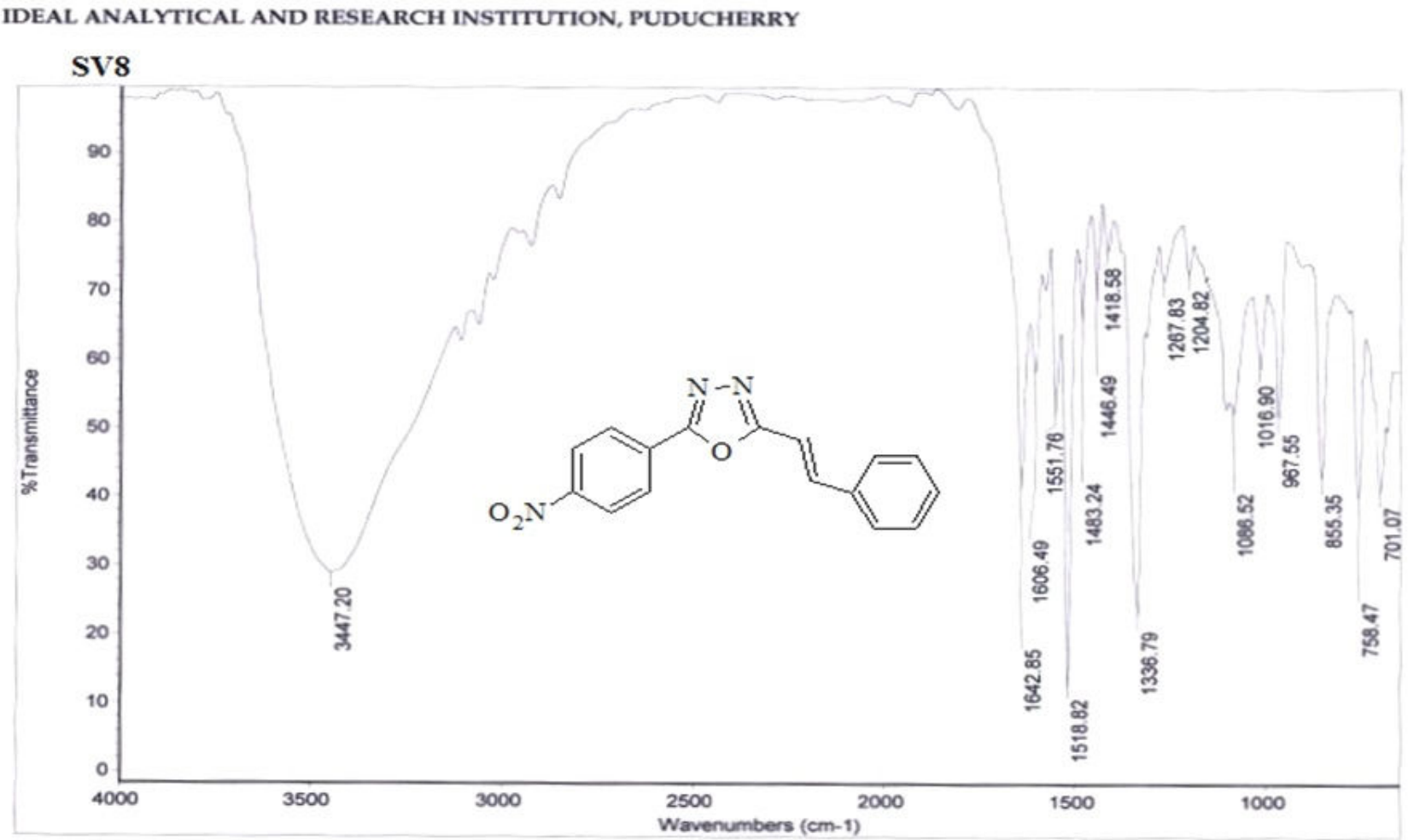
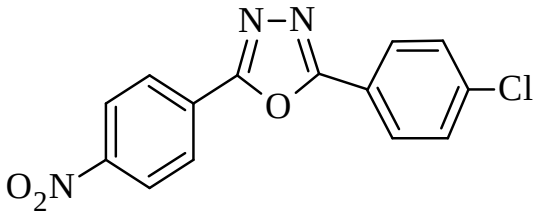


Figure 9: IR SPECTRA OF SV-8

Interpretation of IR spectrum of SV-9



The significant wave numbers of the compound and its functional groups are given below**.**

Table -14

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3451.50	N-H Stretching
2	2925.72	C-H Stretching in Aromatic Group
3	1604.42	C=C Stretching in Aromatic Hydrocarbon
4	1556.18	N=O & C=N Stretching (Aromatic Nitro Group)
5	1528.08,1480.13	C=C & C=N Stretching
6	1108.53	C-O-C Stretching
7	1071.64	C-O Stretching
8	708.91	C-Cl Stretching

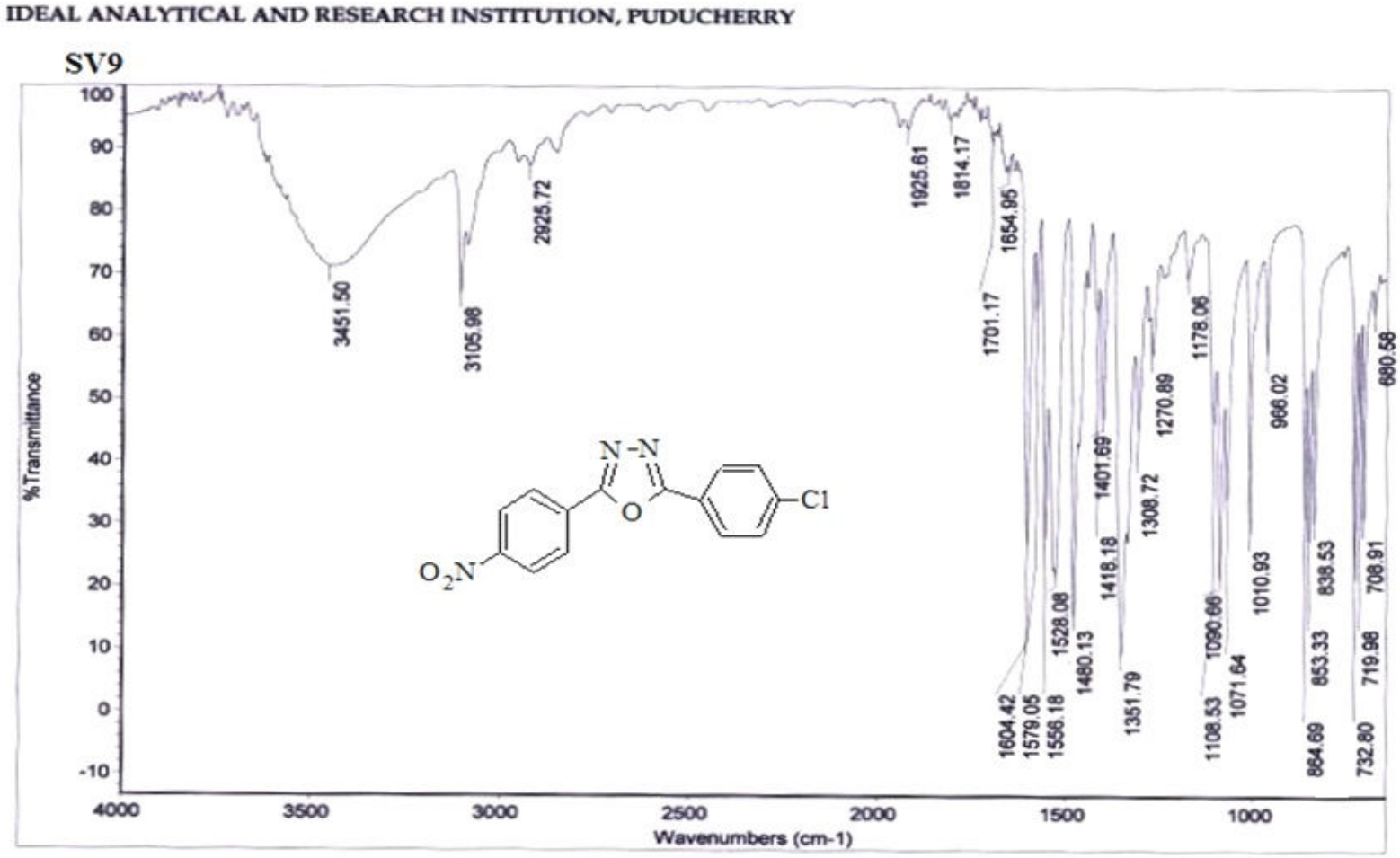
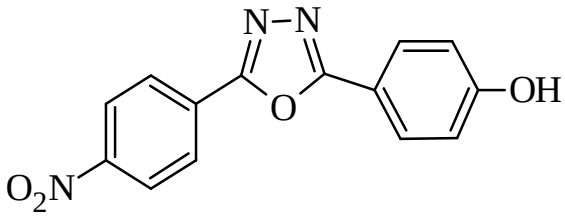


Figure 10: IR SPECTRA OF SV-9

Interpretation of IR spectrum of SV-10



The significant wave numbers of the compound and its functional groups are given below**.**

Table -15

S.No	WAVE NUMBER (cm^{-1})	FUNCTIONAL GROUP
1	3425.10	N-H Stretching
2	3108.04,2918.46	C-H Stretching in Aromatic Group
3	2849.42	O-H Stretching
4	1603.00	C=C Stretching in Aromatic Hydrocarbon
5	1555.48,1521.88	N=O & C=N Stretching (Aromatic Nitro Group)
6	1491.47,1441.30	C=C & C=N Stretching
7	1346.00	C-N Stretching (Aromatic Teritary)
8	1170.25	C-O-C Stretching
9	1074.39	C-O Stretching

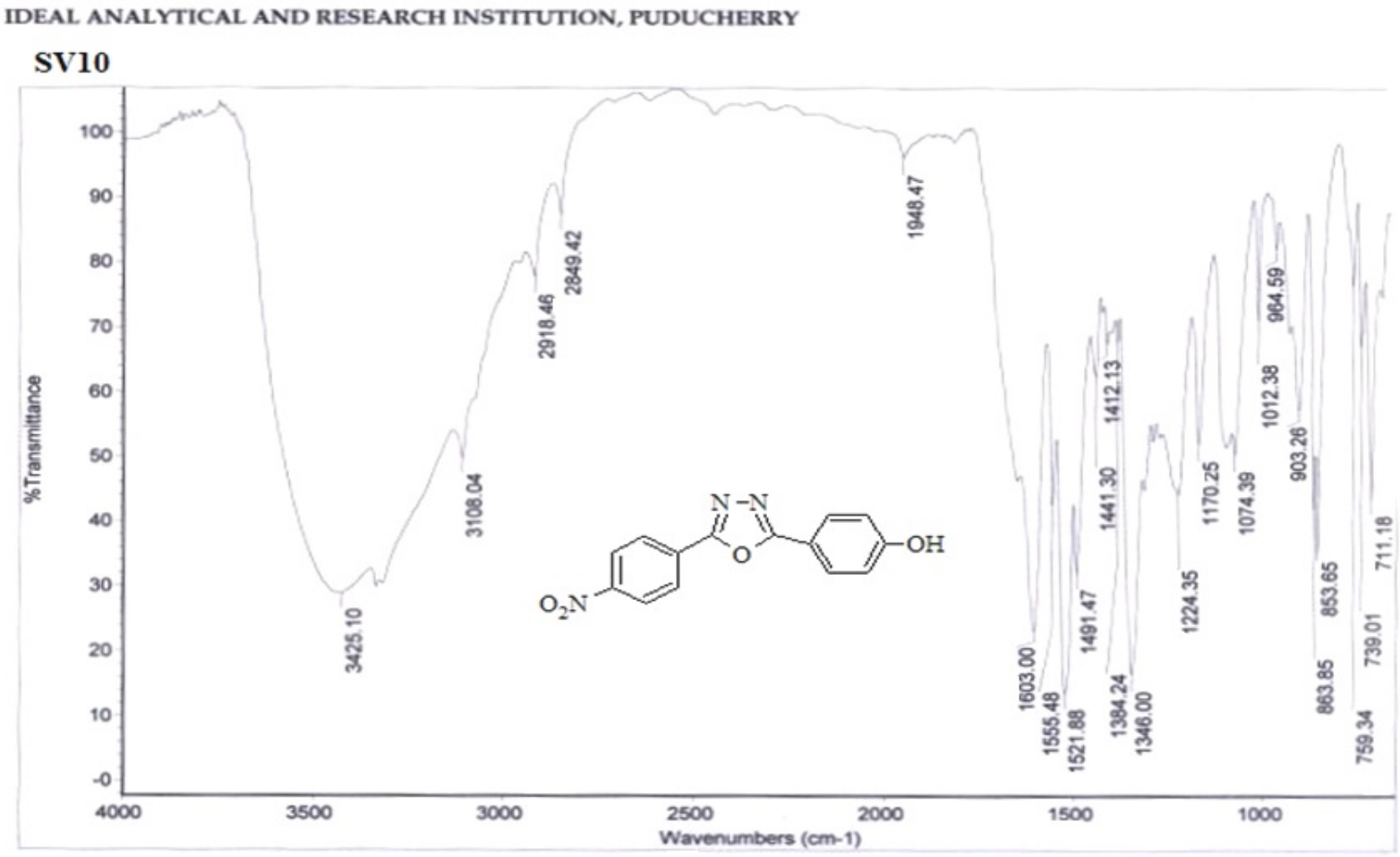
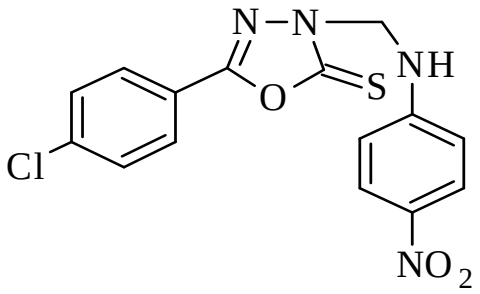


Figure 11: IR SPECTRA OF SV-10

ii) ^{1}H NMR Spectrum Analysis

The proton NMR values are recorded in the BRUKER- 400 MHz and the δ values with reference to the nature of protons are given in the table**.**

Interpretation of ^{1}H NMR spectra of SV-1



The δ values with reference to the nature of protons are given below.

Table-16

S. No	Values in ppm	Nature of protons
1	5.96	Protons in secondary amine group
2	6.79	Protons in methyl group
3	7.53-7.56	Phenyl Protons in Chloro benzene
4	7.79-8.14	Phenyl Protons in nitro benzene

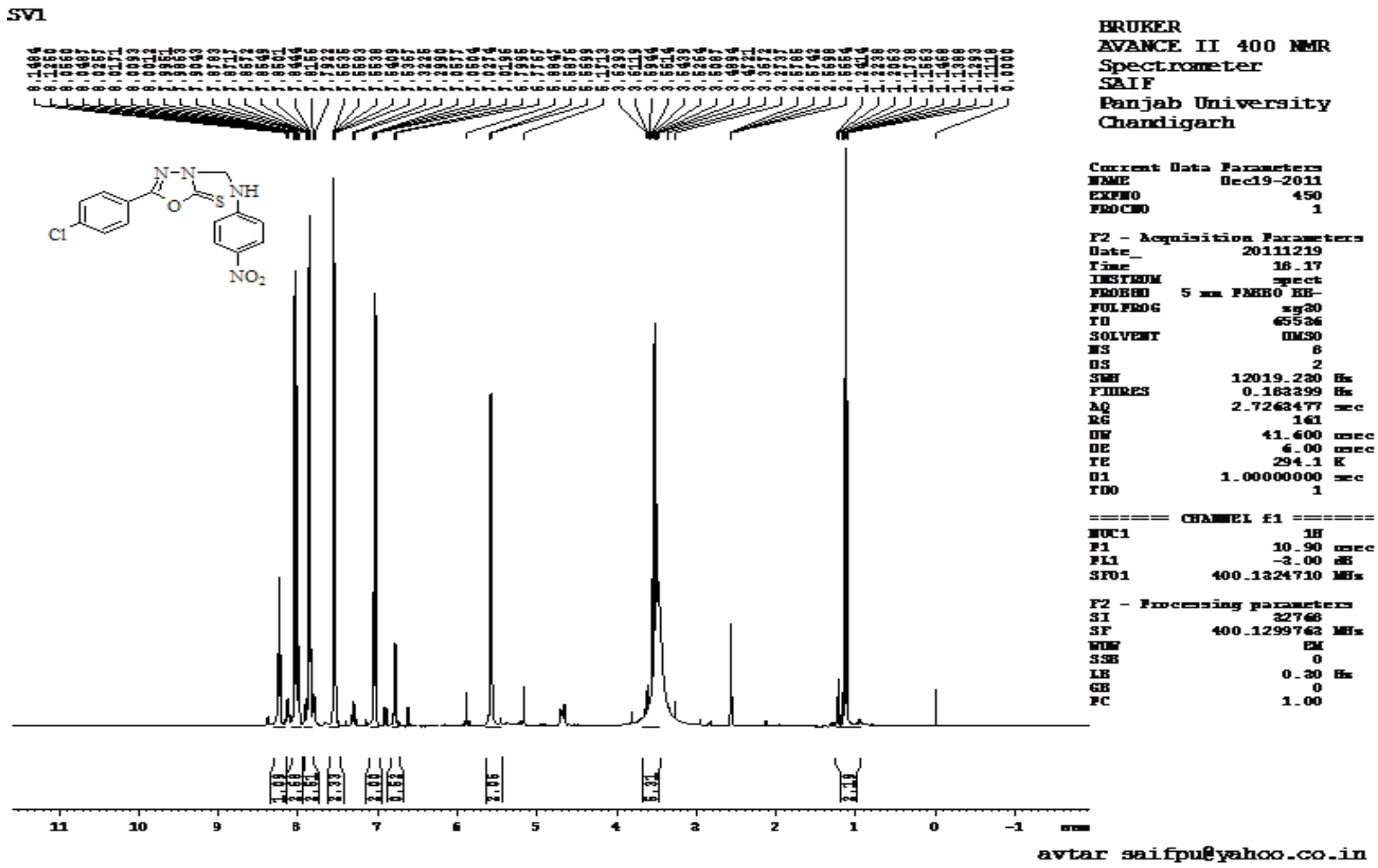
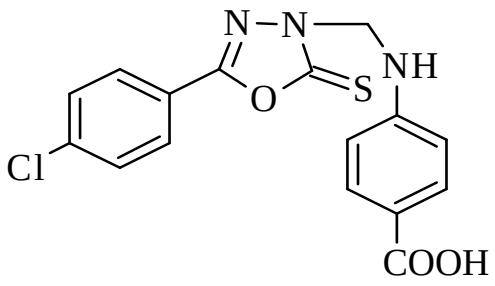


Figure 12: NMR SPECTRA OF SV-1

Interpretation of ^{1}H NMR Spectra of SV-2



The δ values with reference to the nature of protons are given below.

Table-17

S. No	Values in ppm	Nature of protons
1	2.57	Protons in Carboxylicacid group
2	5.78	Protons in amine group
3	6.94	Protons in methyl group
4	7.50-7.57	Phenyl Protons in Chloro benzene
5	7.74-7.93	Phenyl Protons in Benzoic acid

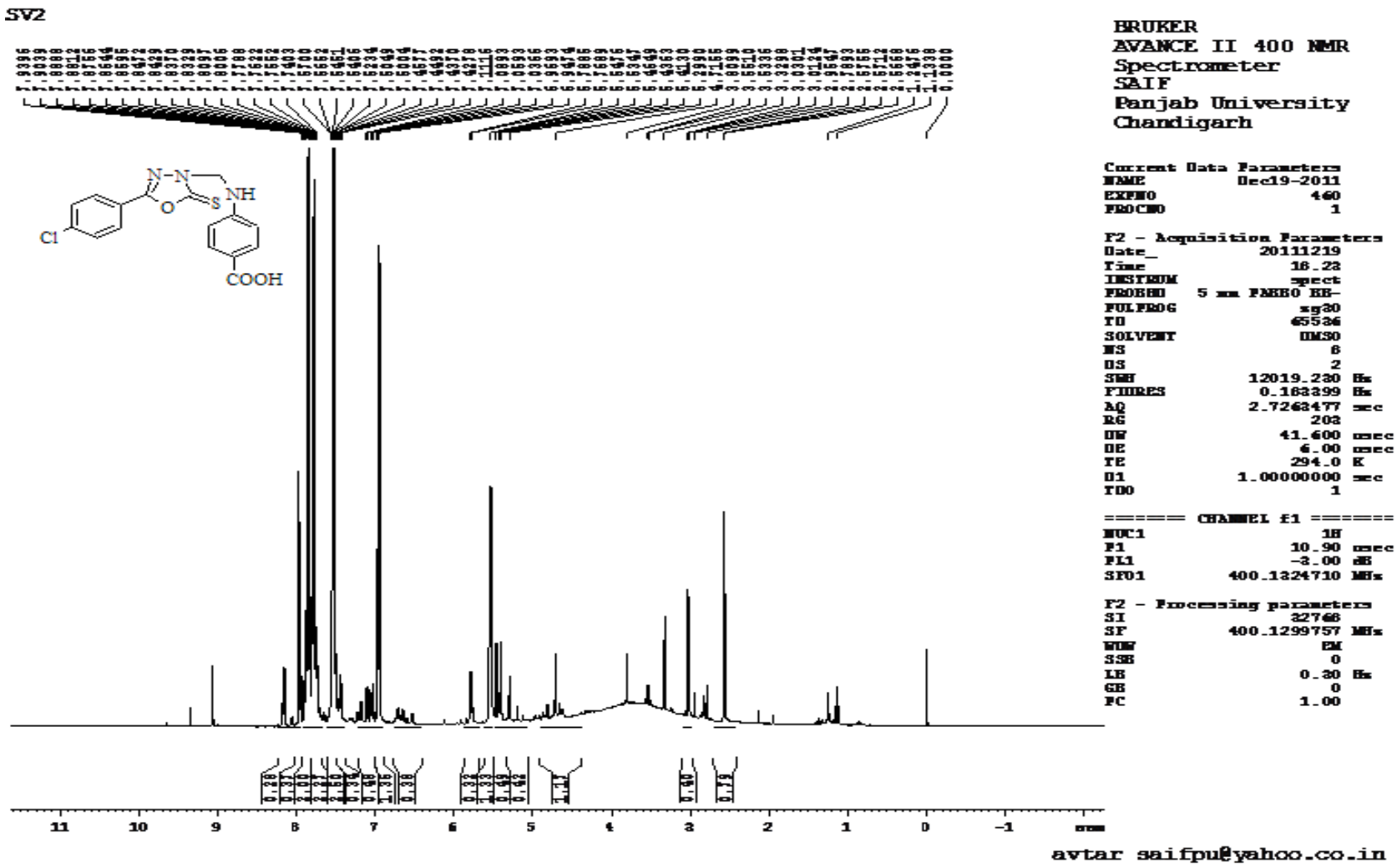
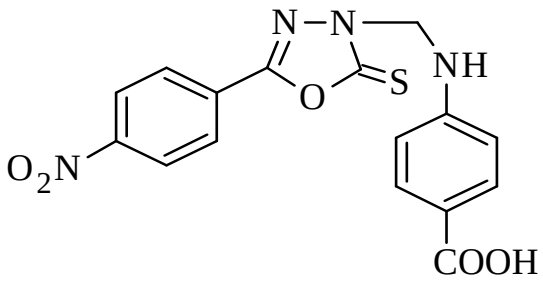


Figure 13: NMR SPECTRA OF SV-2

Interpretation of ^{1}H NMR Spectra of SV-3



The δ values with reference to the nature of protons are given below.

Table-18

S. No	Values in ppm	Nature of protons
1	2.57	Protons in Carboxylicacid group
2	5.09	Protons in amine group
3	6.95	Protons in methyl group
4	7.73-7.91	Phenyl Protons in Benzoic acid
5	8.09-8.39	Phenyl Protons in Nitrobenzene

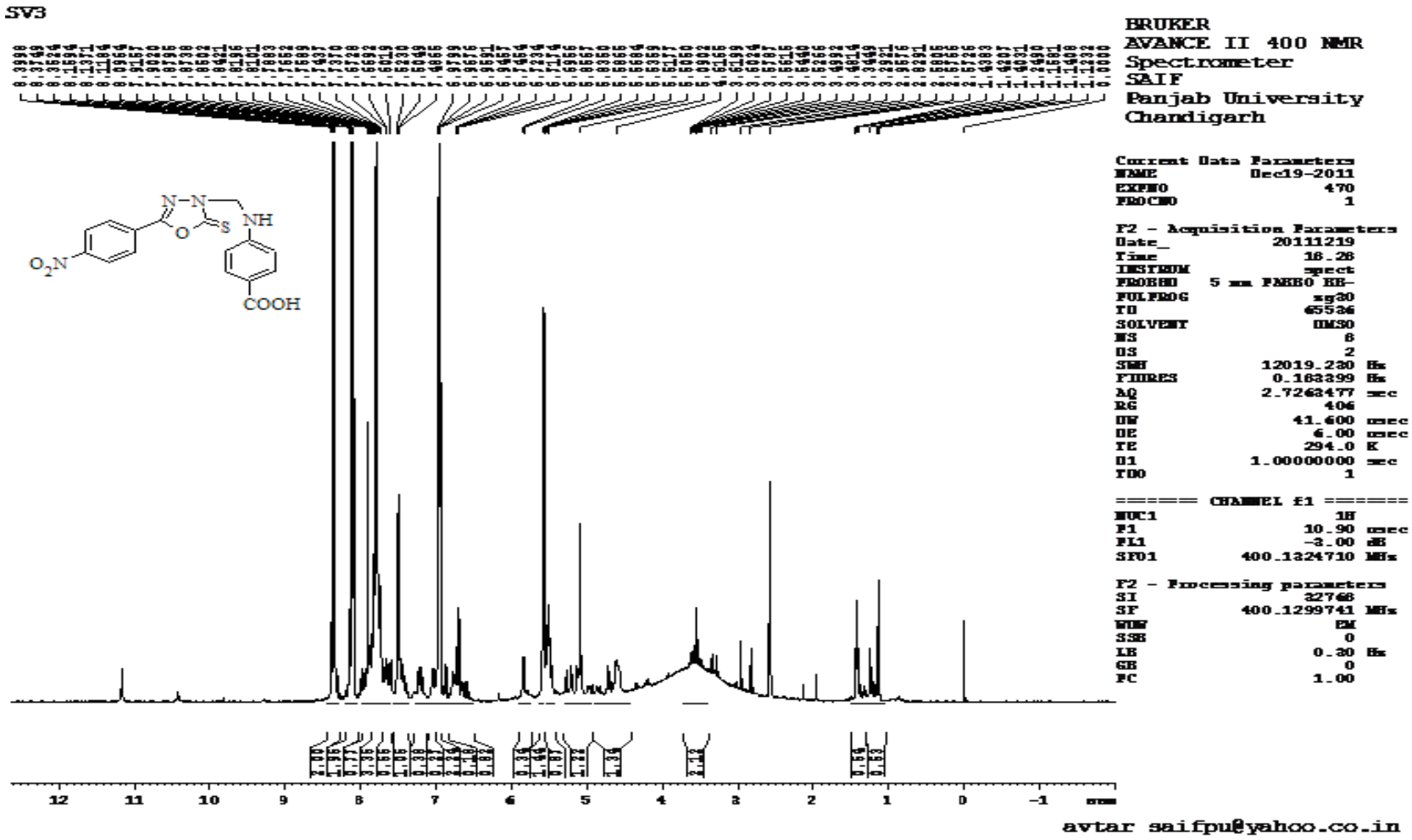
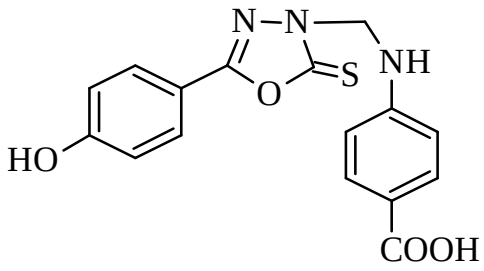


Figure 14: NMR SPECTRA OF SV-3

Interpretation of ^{1}H NMR Spectra of SV-4



The δ values with reference to the nature of protons are given below.

Table-19

S. No	Values in ppm	Nature of protons
1	2.57	Protons in Carboxylicacid group
2	5.06	Protons in amine group
3	5.76	Protons in methyl group
4	6.82-6.99	Phenyl Protons in Phenol
5	7.65-7.92	Phenyl Protons in Benzoic acid

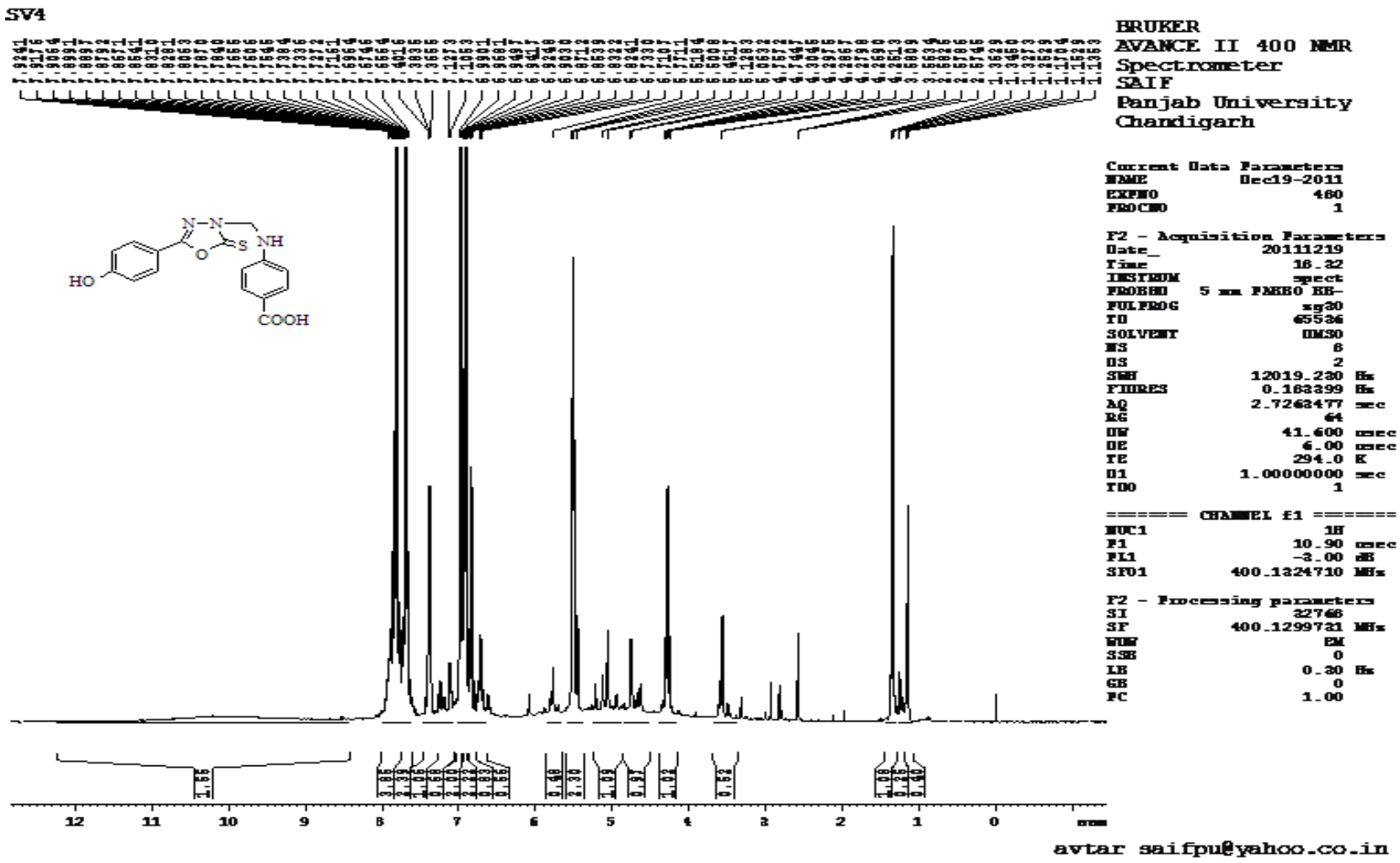
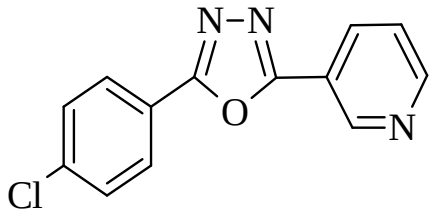


Figure 15: NMR SPECTRA OF SV-4

Interpretation of ^{1}H NMR Spectra of SV-5



The δ values with reference to the nature of protons are given below.

Table-20

S. No	Values in ppm	Nature of protons
1	7.42-7.55	Protons in Aromatic group
2	7.99-9.31	Protons in pyridine

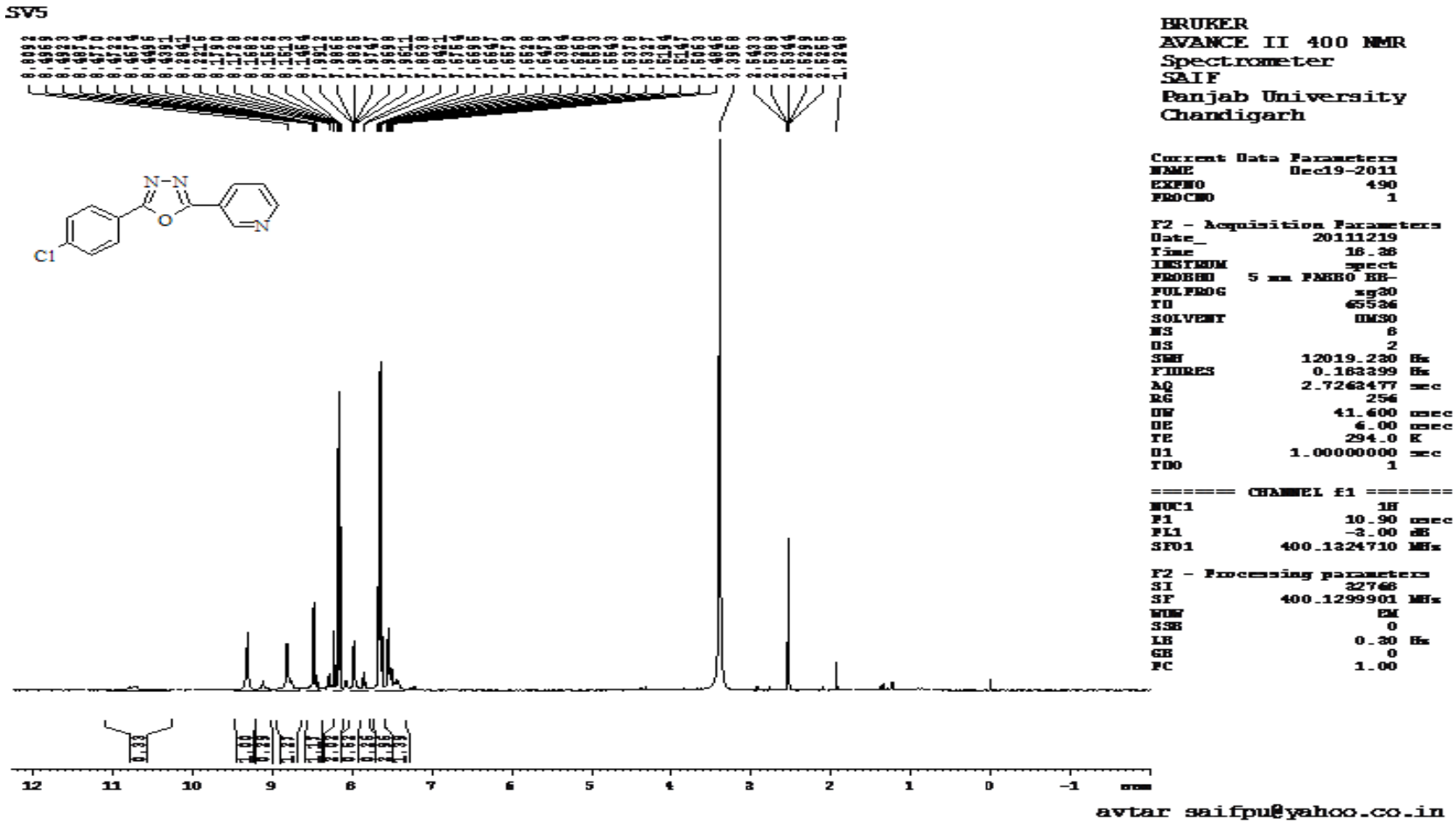
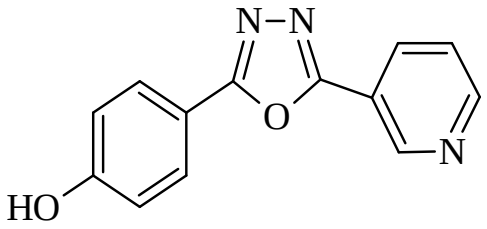


Figure 16: NMR SPECTRA OF SV-5

Interpretation of ^{1}H NMR Spectra of SV-6



The δ values with reference to the nature of protons are given below.

Table-21

S. No	Values in ppm	Nature of protons
1	5.50	Protons in hydroxygroup
2	7.63-7.86	Protons in Aromatic group
3	8.14-8.80	Protons in pyridine

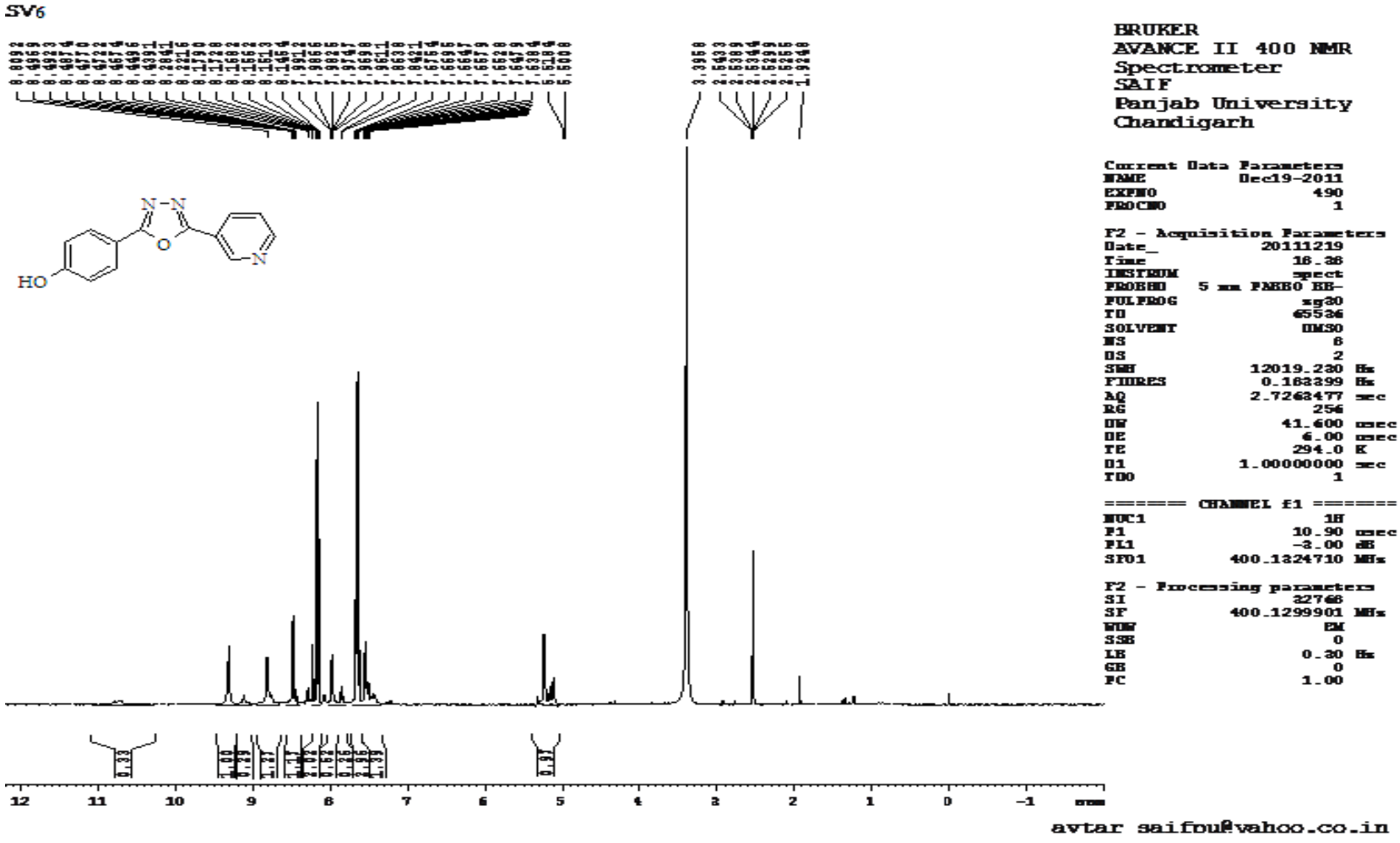
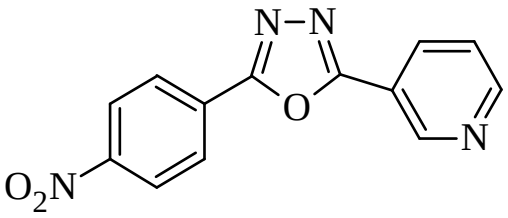


Figure 17: NMR SPECTRA OF SV-6

Interpretation of ^{1}H NMR Spectra of SV-7



The δ values with reference to the nature of protons are given below.

Table-22

S. No	Values in ppm	Nature of protons
1	8.44-8.46	Protons in Aromatic group
2	8.84-8.94	Protons in pyridine

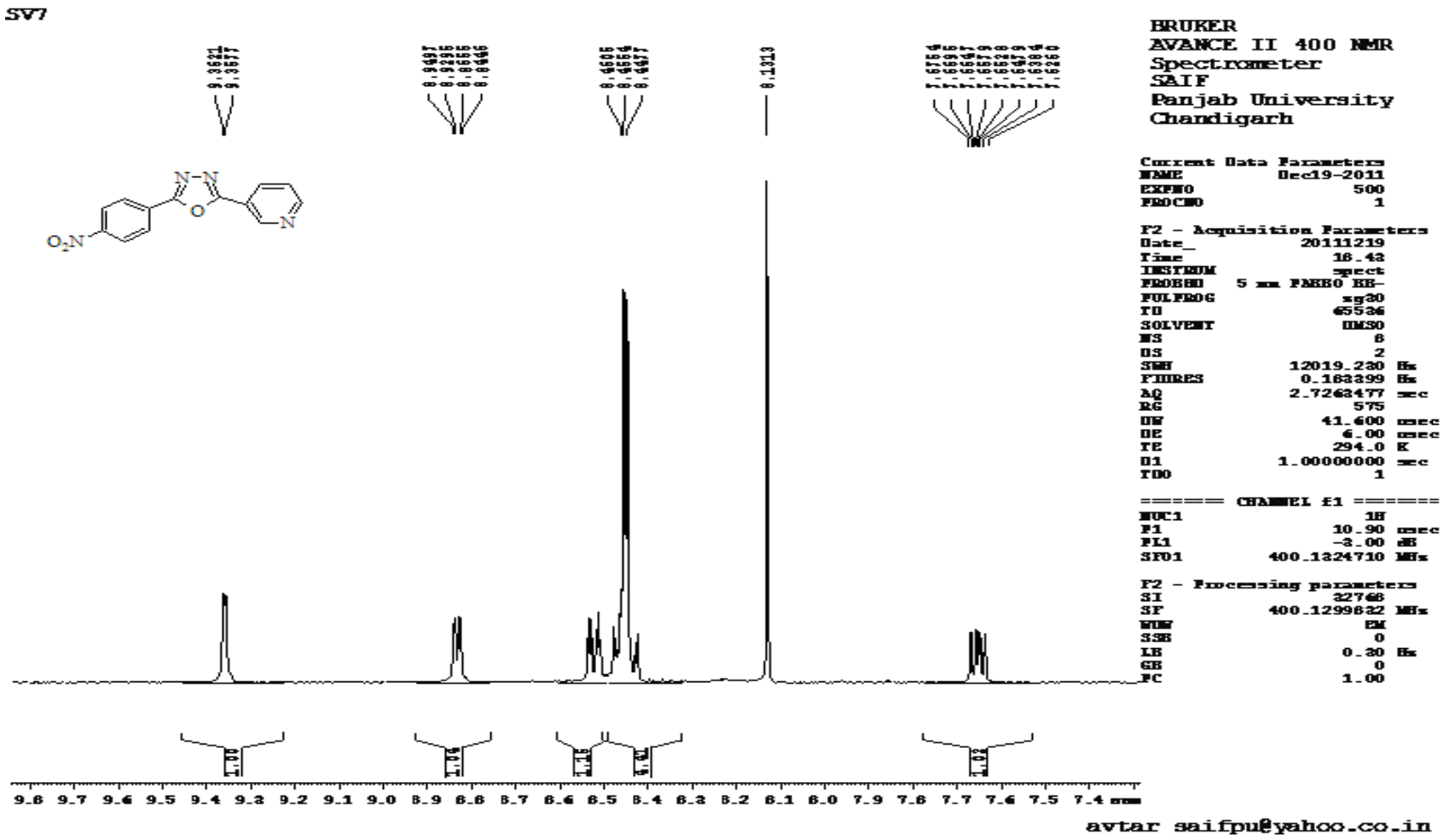
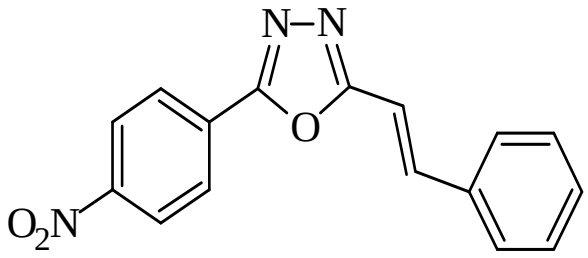


Figure 18: NMR SPECTRA OF SV-7

Interpretation of ^{1}H NMR Spectra of SV-8



The δ values with reference to the nature of protons are given below.

Table-23

S. No	Values in ppm	Nature of protons
1	6.76-6.79	Protons in ethene group
2	7.38-7.43	Protons in benzene
3	8.34-8.44	Protons in nitrobenzene

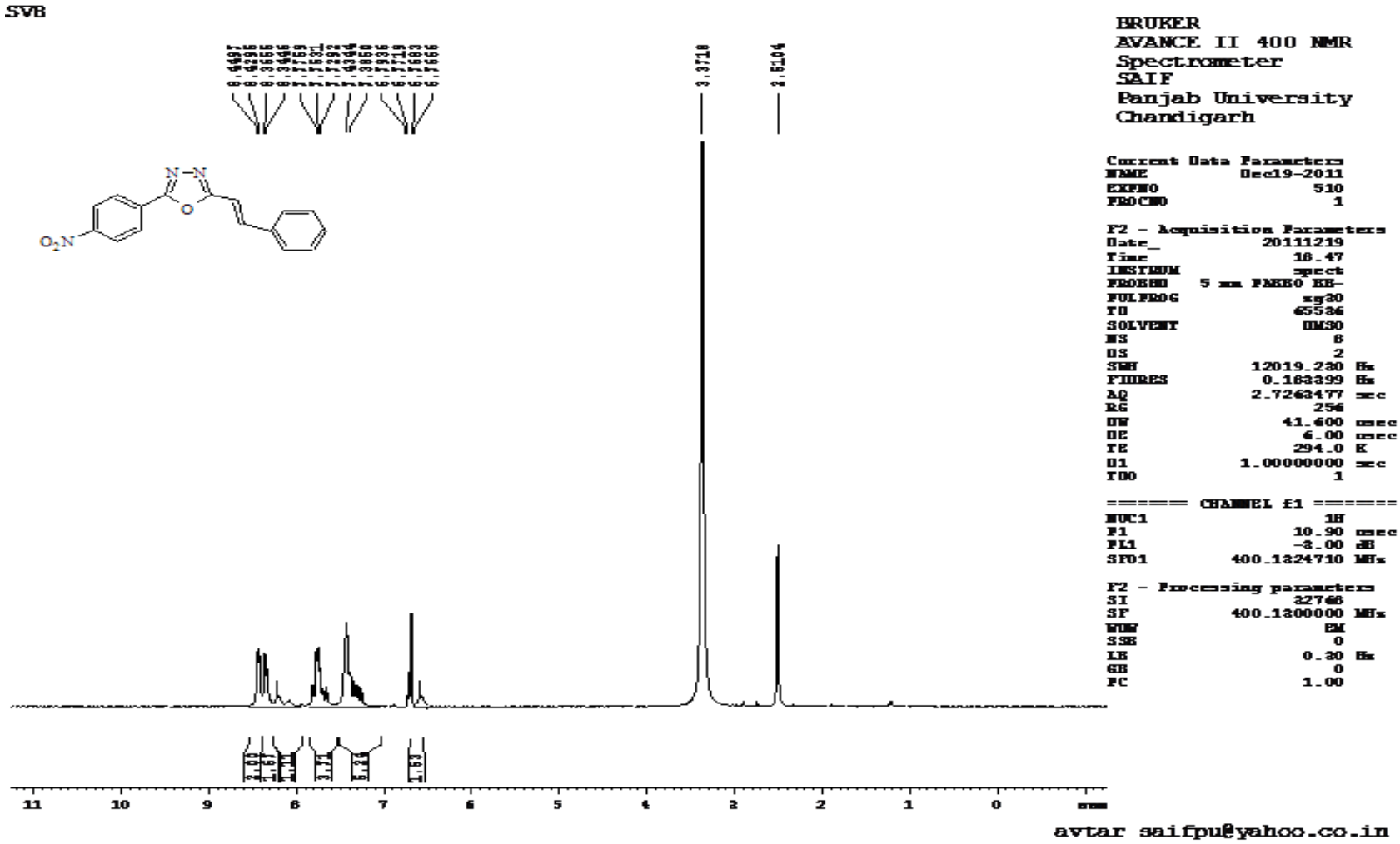
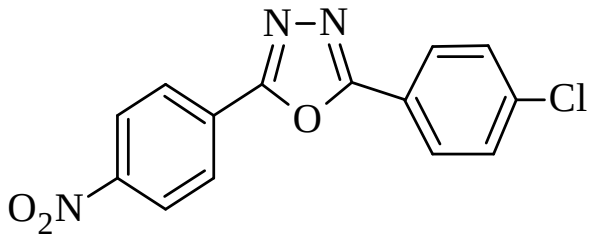


Figure 19: NMR SPECTRA OF SV-8

Interpretation of ^{1}H NMR Spectra of SV-9



The δ values with reference to the nature of protons are given below.

Table-24

S. No	Values in ppm	Nature of protons
1	7.59-7.63	Protons in chloro benzene
2	8.38-8.46	Protons in nitro benzene

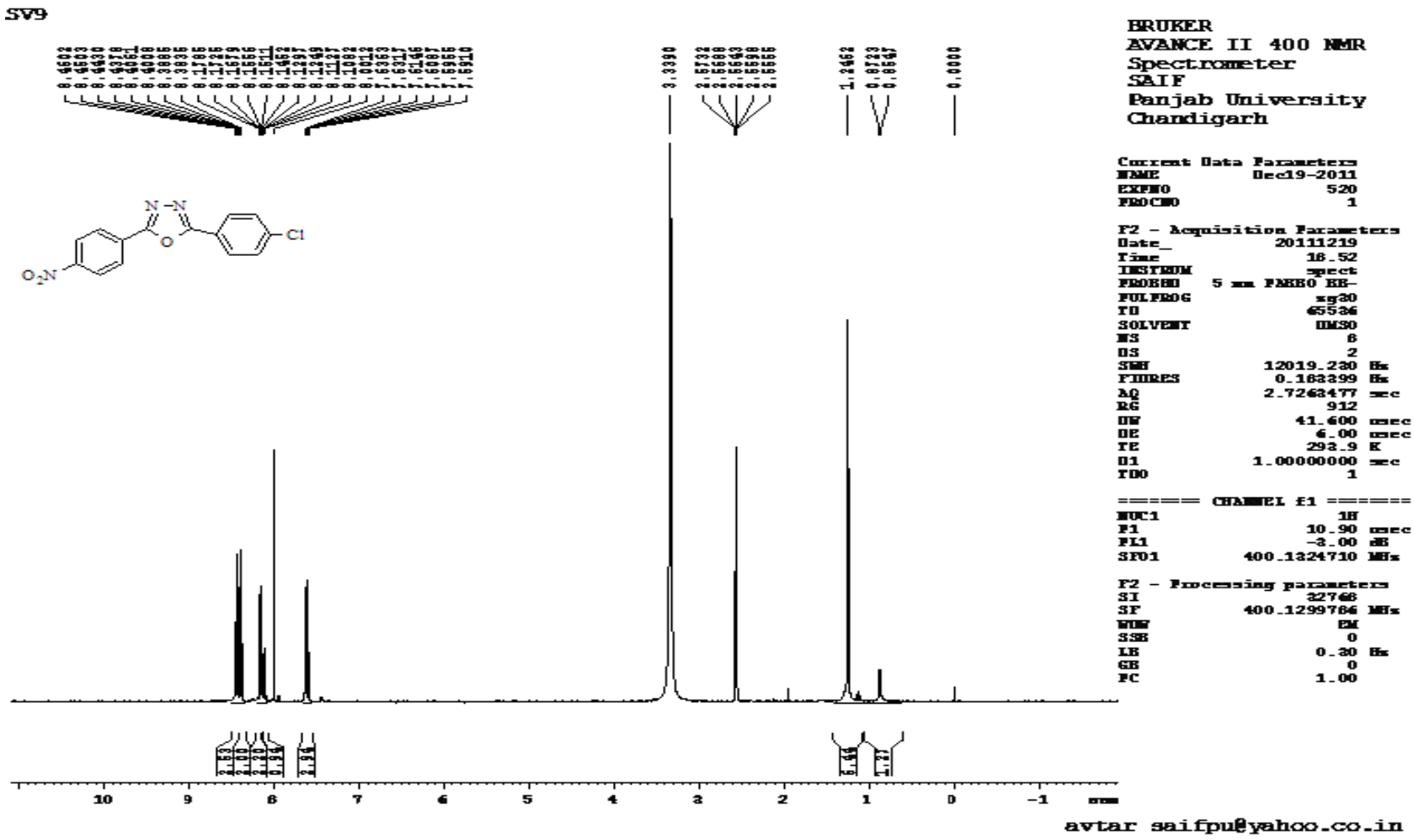
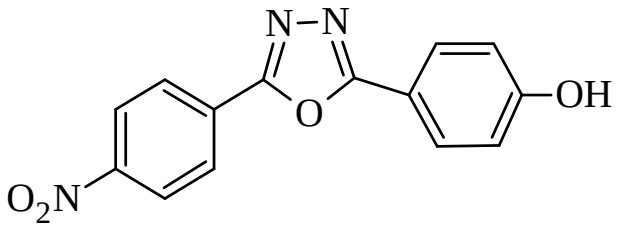


Figure 20: NMR SPECTRA OF SV-9

Interpretation of ^{1}H NMR Spectra of SV-10



The δ values with reference to the nature of protons are given below.

Table-25

S. No	Values in ppm	Nature of protons
1	5.14	Protons in hydroxyl group
2	6.54-6.99	Protons in hydroxy benzene
3	8.31-8.52	Protons in nitro benzene

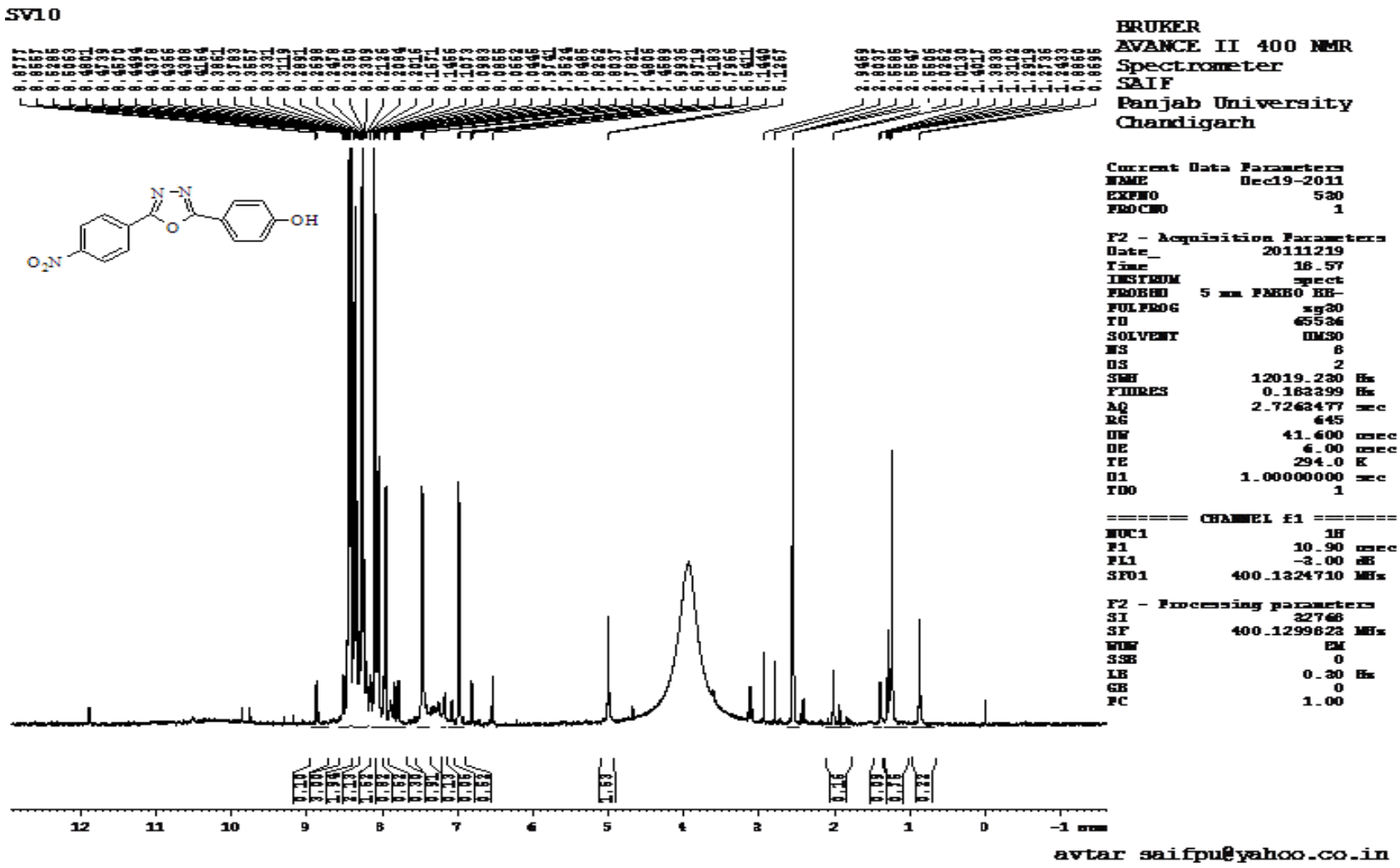
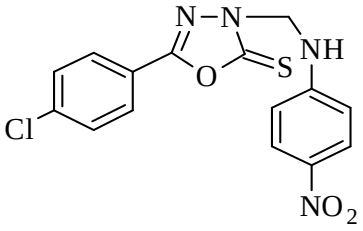


Figure 21: NMR SPECTRA OF SV-10

iii) Mass Spectral Analysis

Mass spectrum of the sample was recorded in JEOLGC mate by Electron impact method as ionization mode. The m/z values of the samples are given in the table.

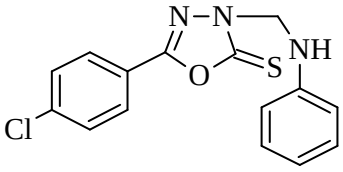
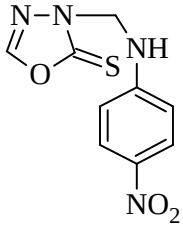
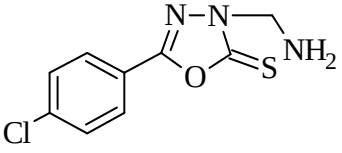
Interpretation of Mass spectra of SV-1

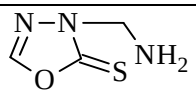
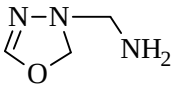
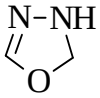


Molecular weight: 362.79 Molecular ion peak: 362.79

The possible fragments of the molecule with the relevant to its m/z values are:

Table-26

S.No	m/z	Fragments
1	315.71	
2	251.79	
3	240.02	

4	128.63	
5	98.01	
6	72.25	

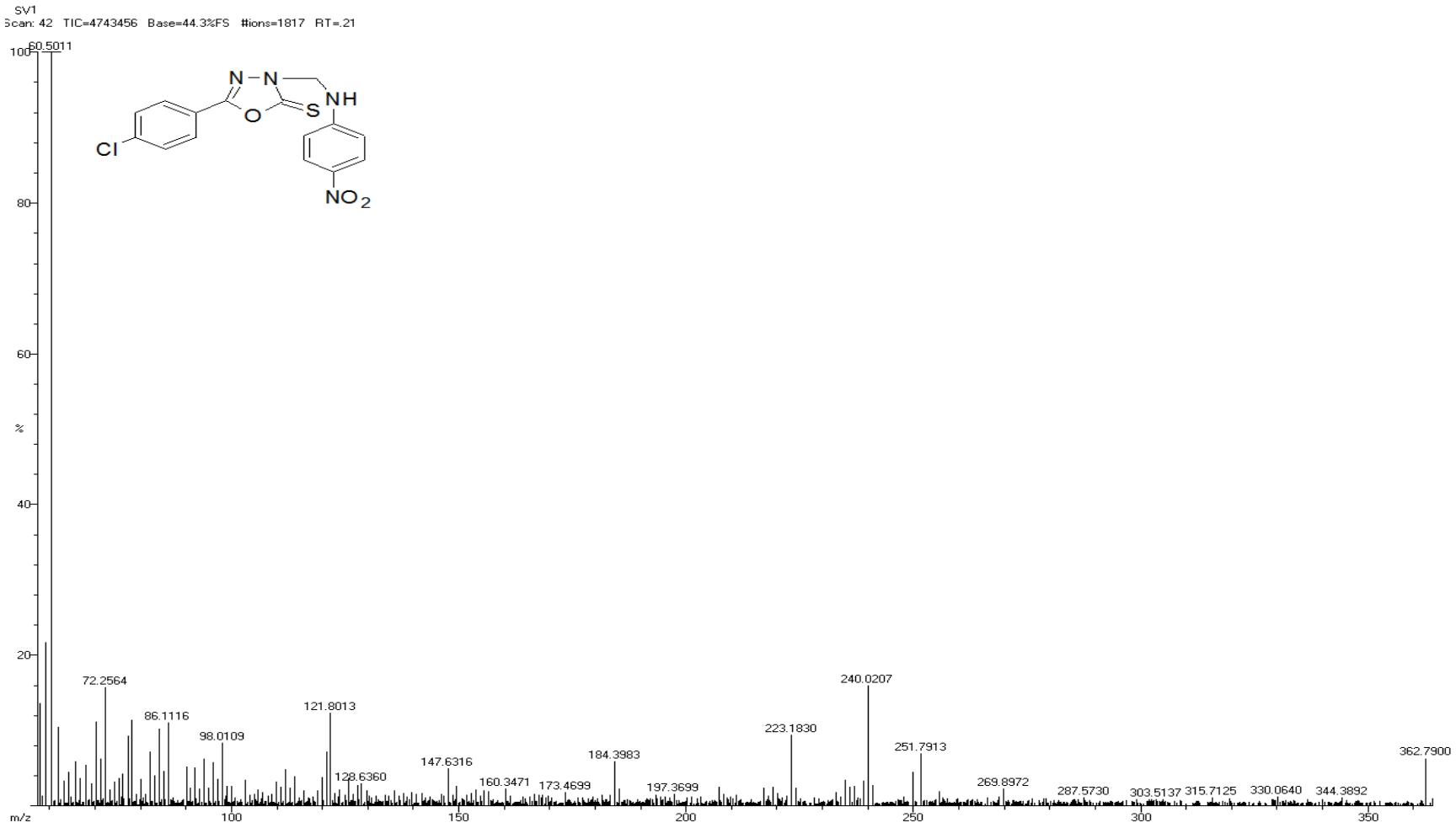
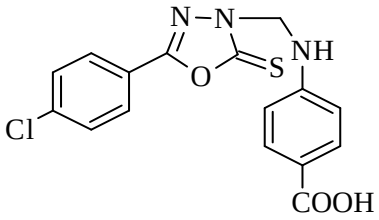


Figure 22: Mass Spectra of SV-1

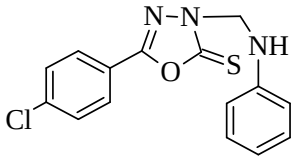
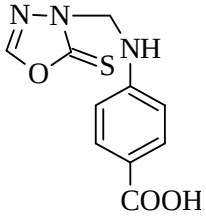
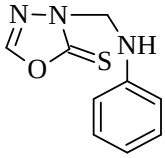
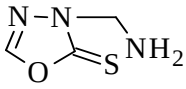
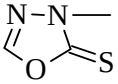
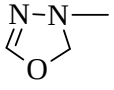
Interpretation of Mass spectra of SV-2



Molecular weight: 361.80 Molecular ion peak: 362.10

The possible fragments of the molecule with the relevant to its m/z values are:

Table-27

S.No	m/z	Fragments
1	317.11	
2	250.06	
3	206.48	
4	130.97	
5	115.07	
6	83.37	

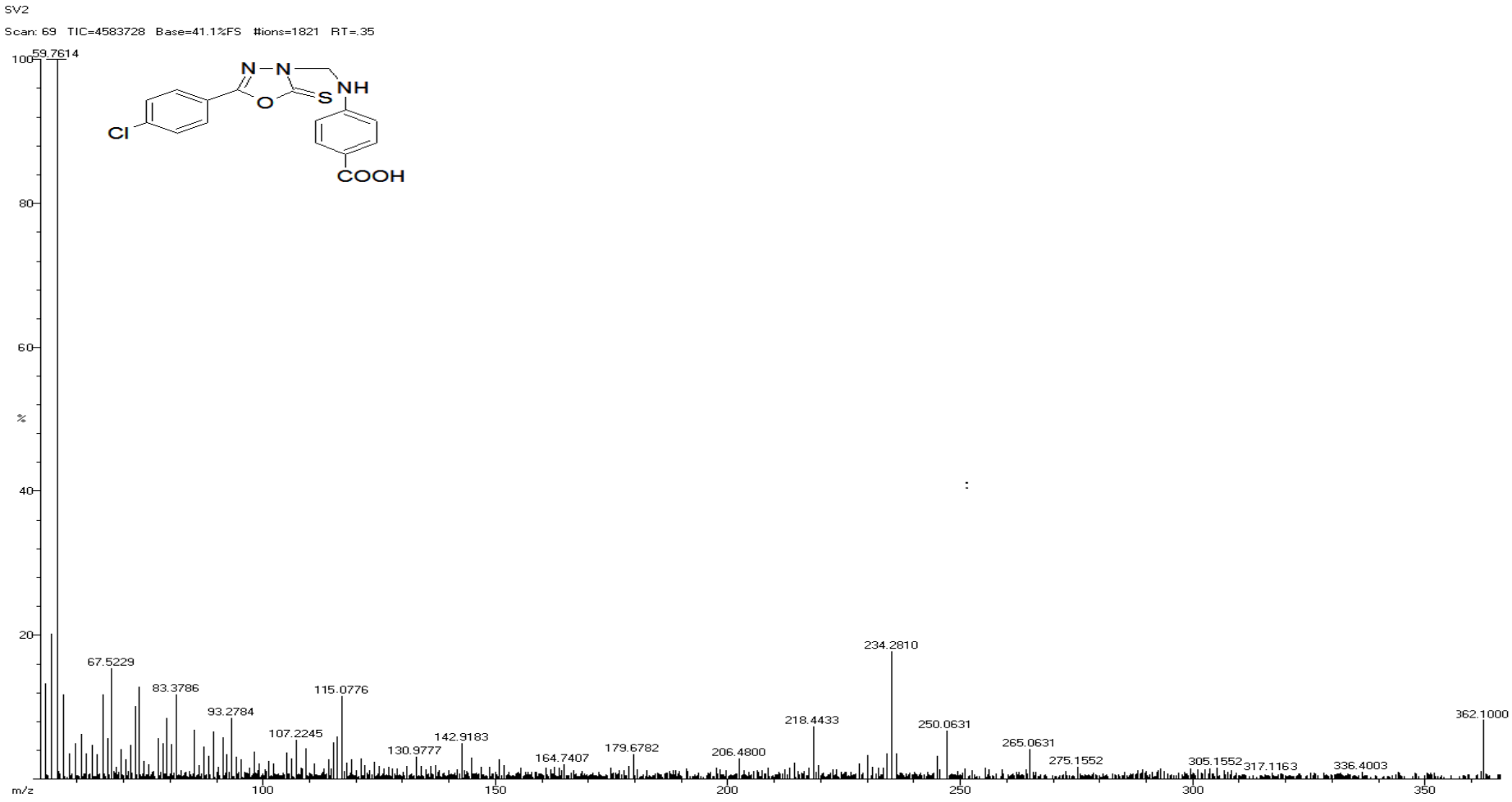
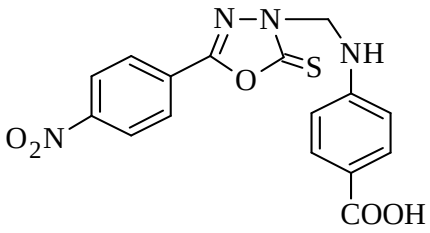


Figure 23: Mass Spectra of SV-2

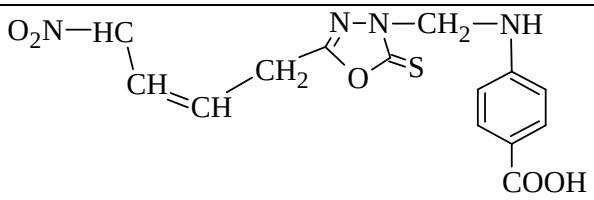
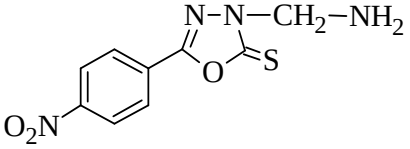
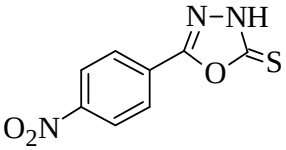
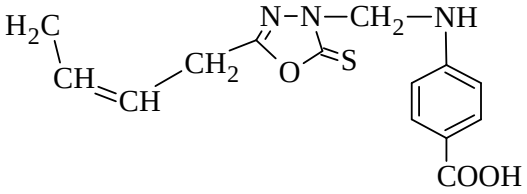
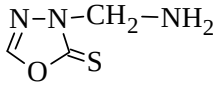
Interpretation of Mass spectra of SV-3



Molecular weight: 372.35 Molecular ion peak: 372.35

The possible fragments of the molecule with the relevant to its m/z values are:

Table-28

S.No	m/z	Fragments
1	346.39	
2	250.96	
3	222.36	
4	183.52	
5	128.89	
6	77.31	

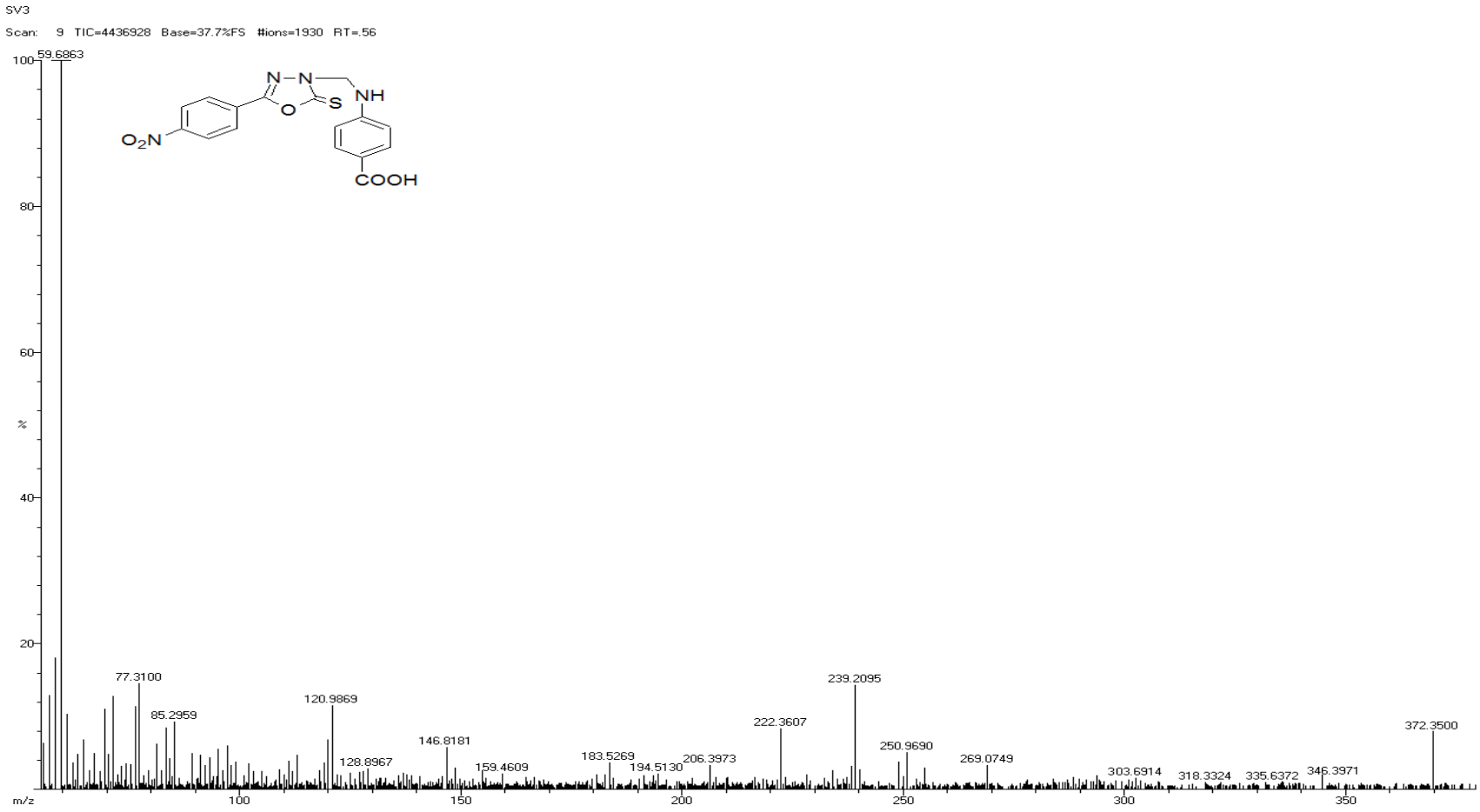
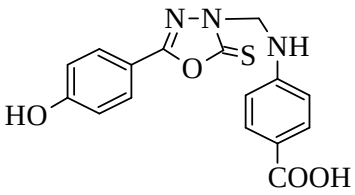


Figure 24 : Mass Spectra of SV-3

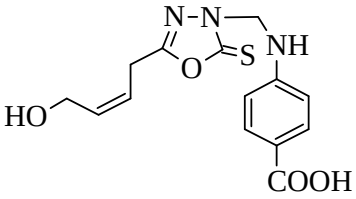
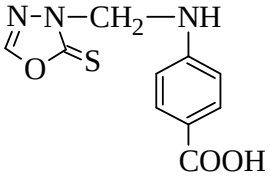
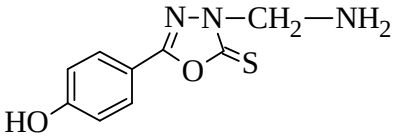
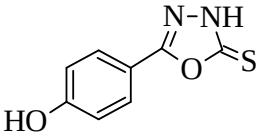
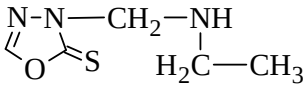
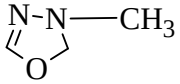
Interpretation of Mass spectra of SV-4



Molecular weight: 343.35 Molecular ion peak: 343.35

The possible fragments of the molecule with the relevant to its m/z values are:

Table-29

S.No	m/z	Fragments
1	317.00	
2	249.64	
3	221.03	
4	193.18	
5	158.13	
6	84.97	

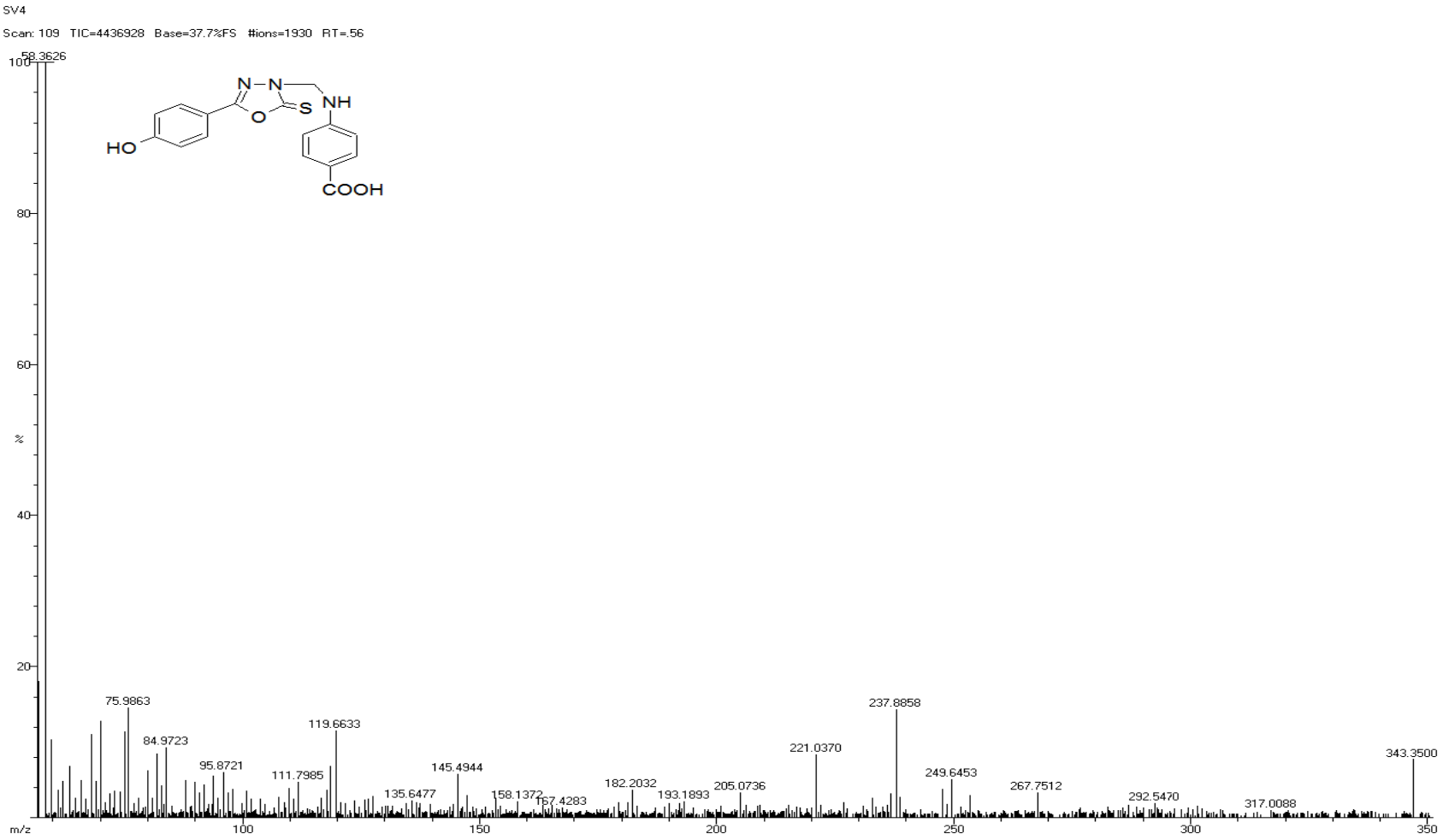
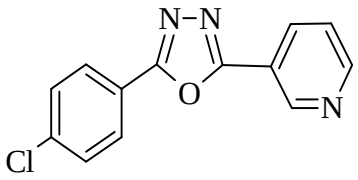


Figure 25: Mass Spectra of SV-4

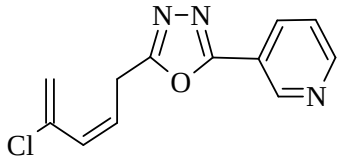
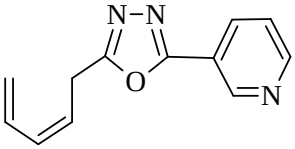
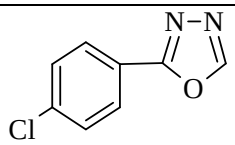
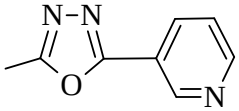
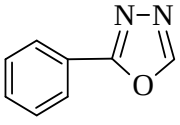
Interpretation of Mass spectra of SV-5



Molecular weight: 257.67 Molecular ion peak: 257.67

The possible fragments of the molecule with the relevant to its m/z values are:

Table-30

S.No	m/z	Fragments
1	244.80	
2	211.88	
3	180.80	
4	154.79	
5	142.82	
6	76.90	

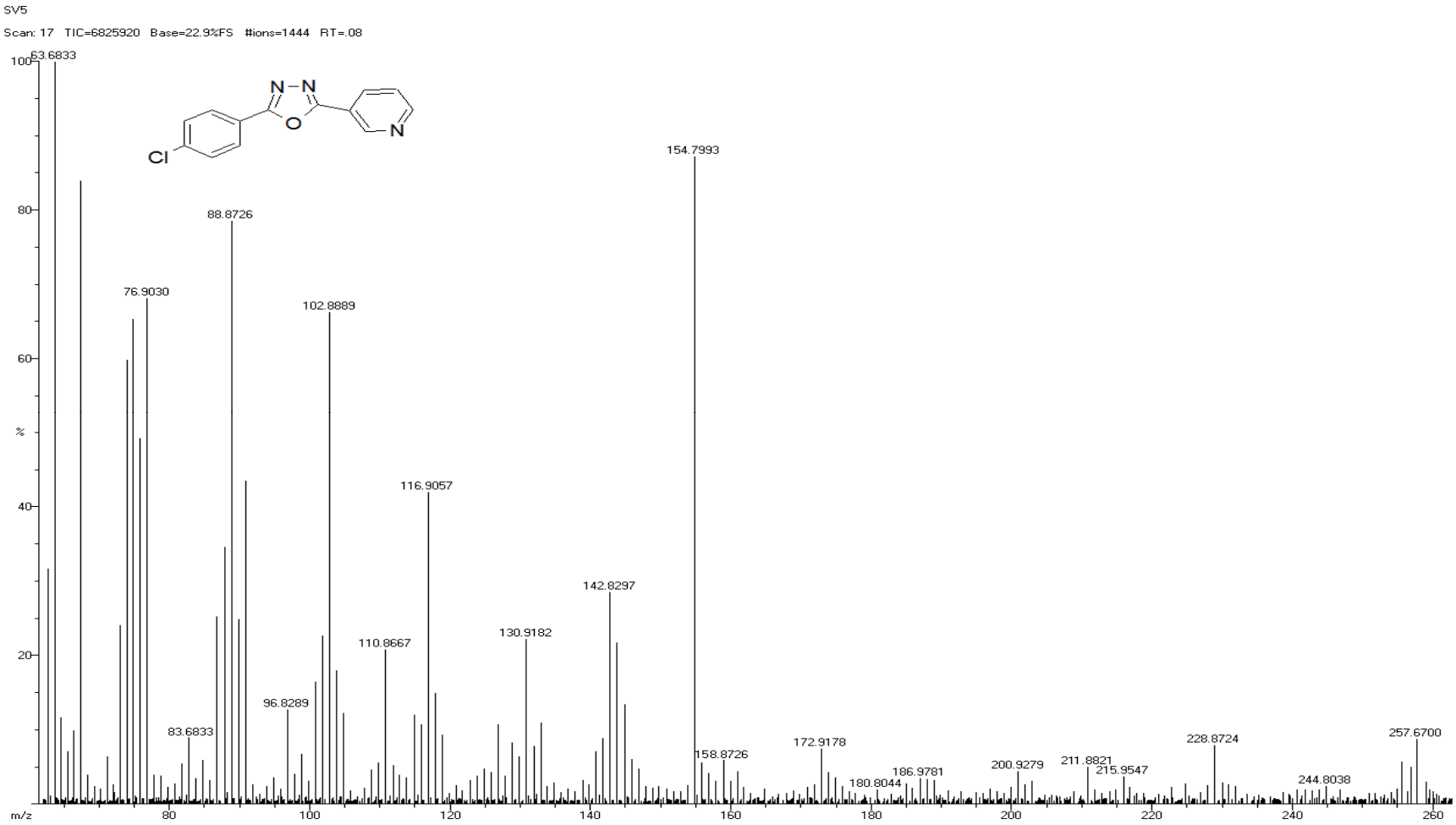
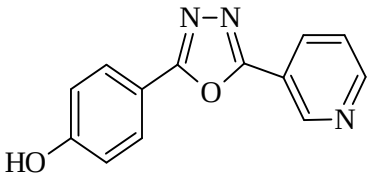


Figure 26: Mass Spectra of SV-5

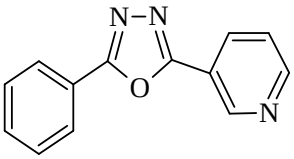
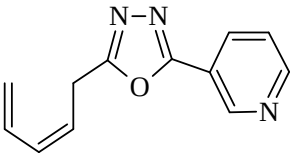
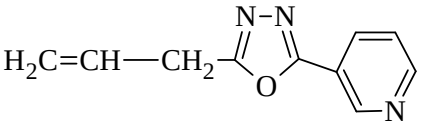
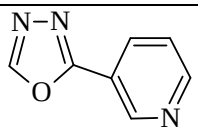
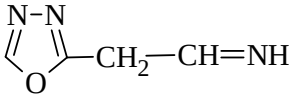
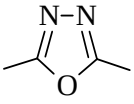
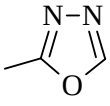
Interpretation of Mass spectra of SV-6



Molecular weight: 239.22 Molecular ion peak: 239.21

The possible fragments of the molecule with the relevant to its m/z values are:

Table-31

S.No	m/z	Fragments
1	223.10	
2	211.08	
3	185.18	
4	146.02	
5	111.13	
6	97.11	
7	83.09	

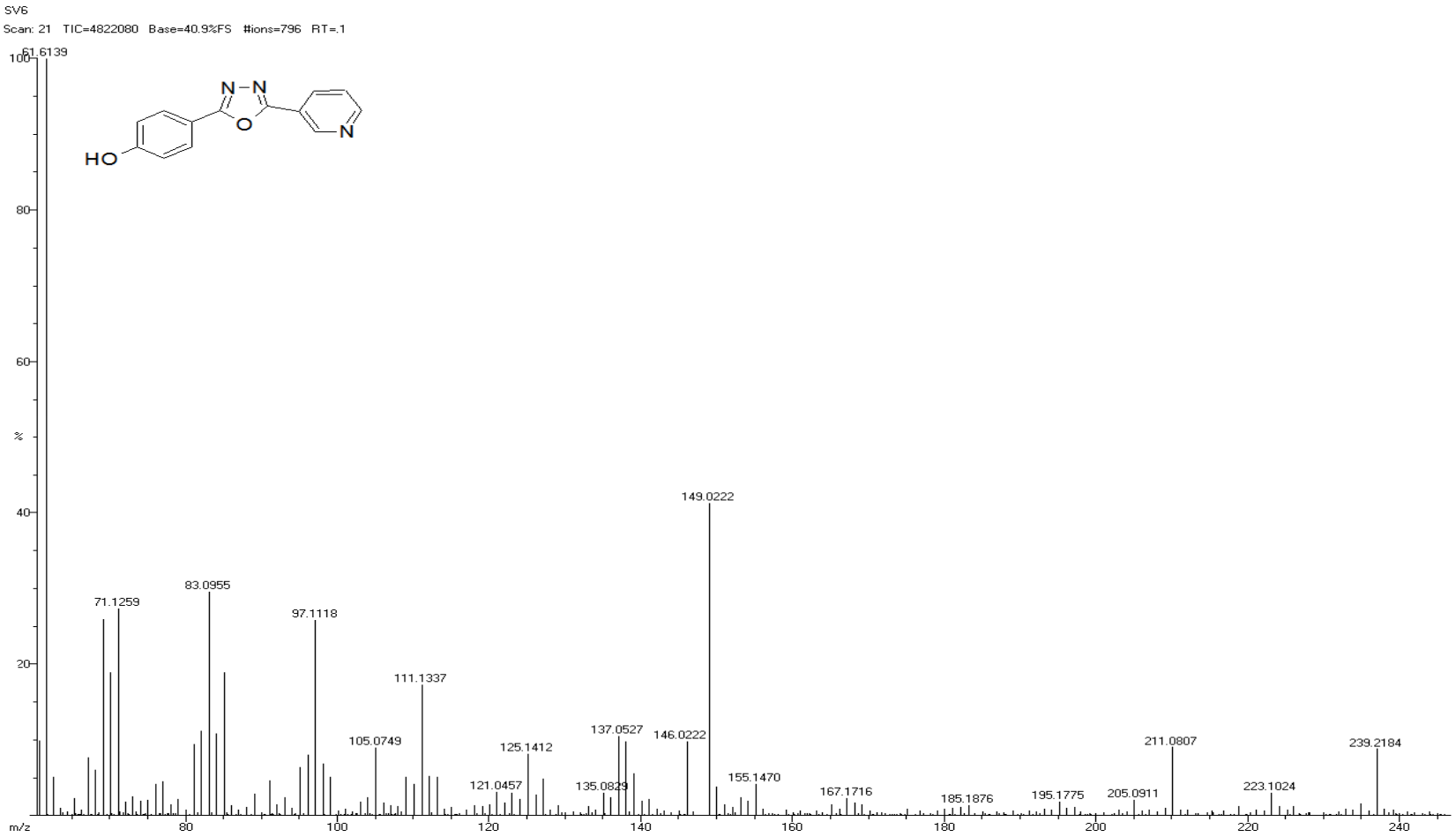
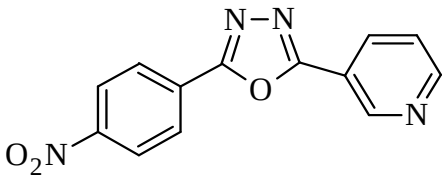


Figure 27: Mass Spectra of SV-6

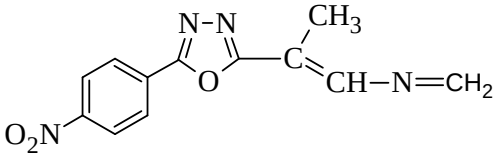
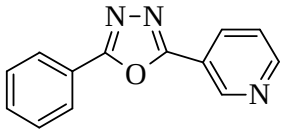
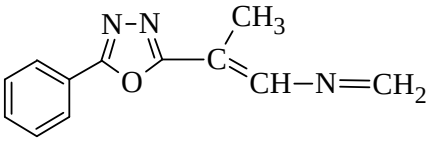
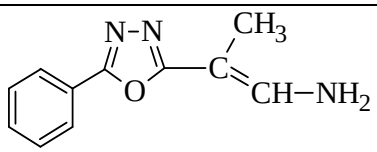
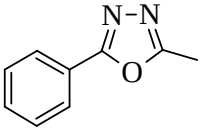
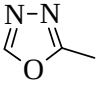
Interpretation of Mass spectra of SV-7



Molecular weight: 268.22 Molecular ion peak: 268.20

The possible fragments of the molecule with the relevant to its m/z values are:

Table-32

S. No	m/z	Fragments
1	258.88	
2	222.17	
3	212.15	
4	198.07	
5	157.02	
6	83.09	

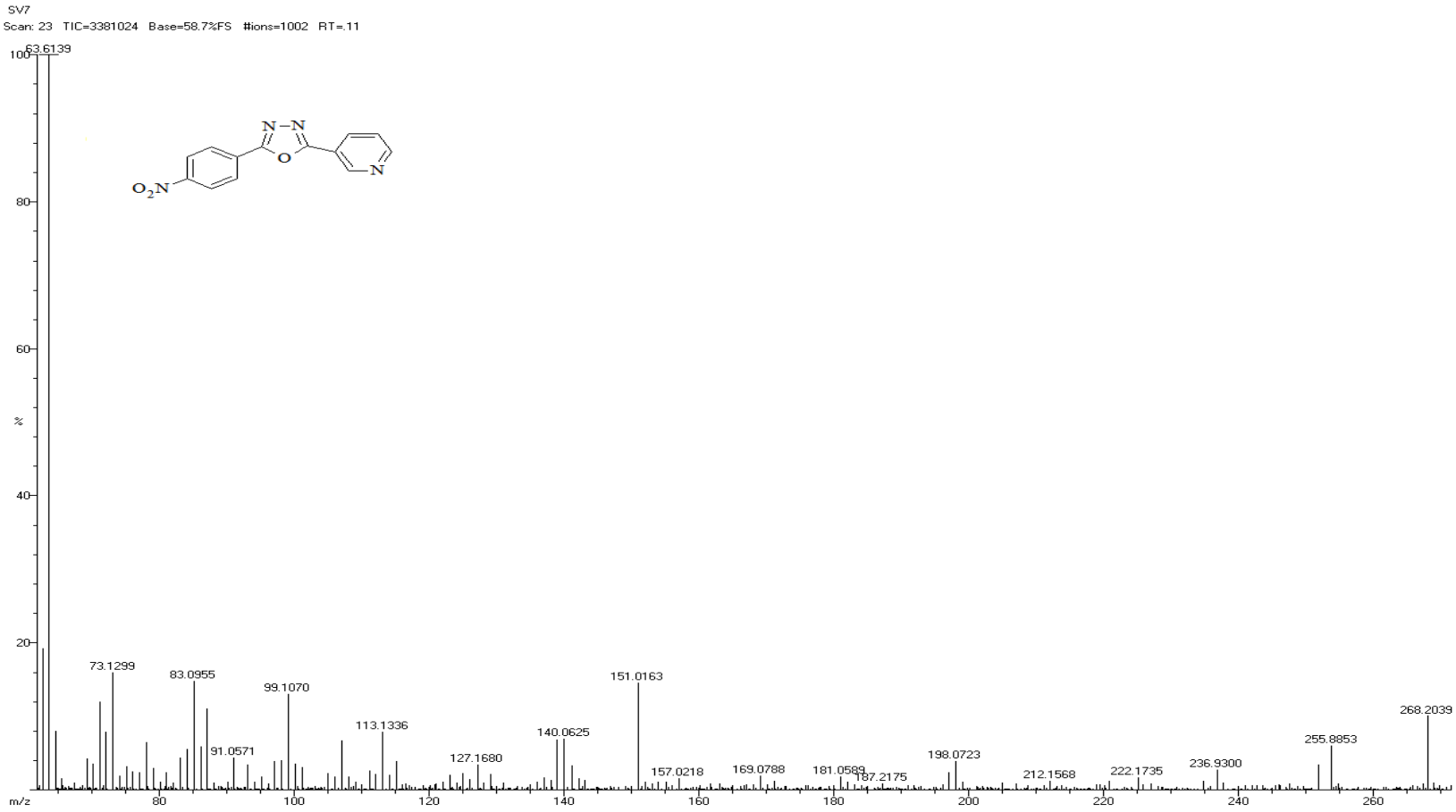
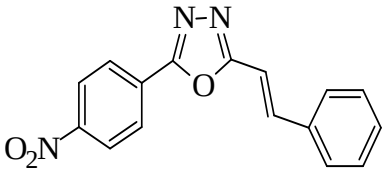


Figure 28: Mass Spectra of SV-7

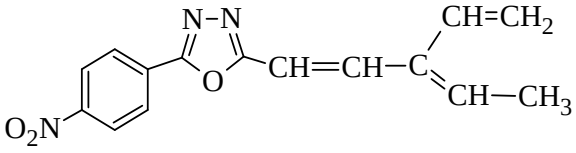
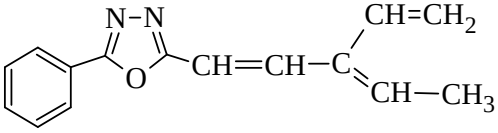
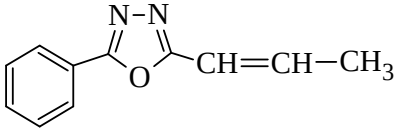
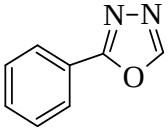
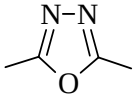
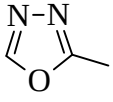
Interpretation of Mass spectra of SV-8



Molecular weight: 293.27 Molecular ion peak: 293.27

The possible fragments of the molecule with the relevant to its m/z values are:

Table-33

S.No	m/z	Fragments
1	280.89	
2	237.65	
3	182.90	
4	149.75	
5	97.84	
6	83.82	

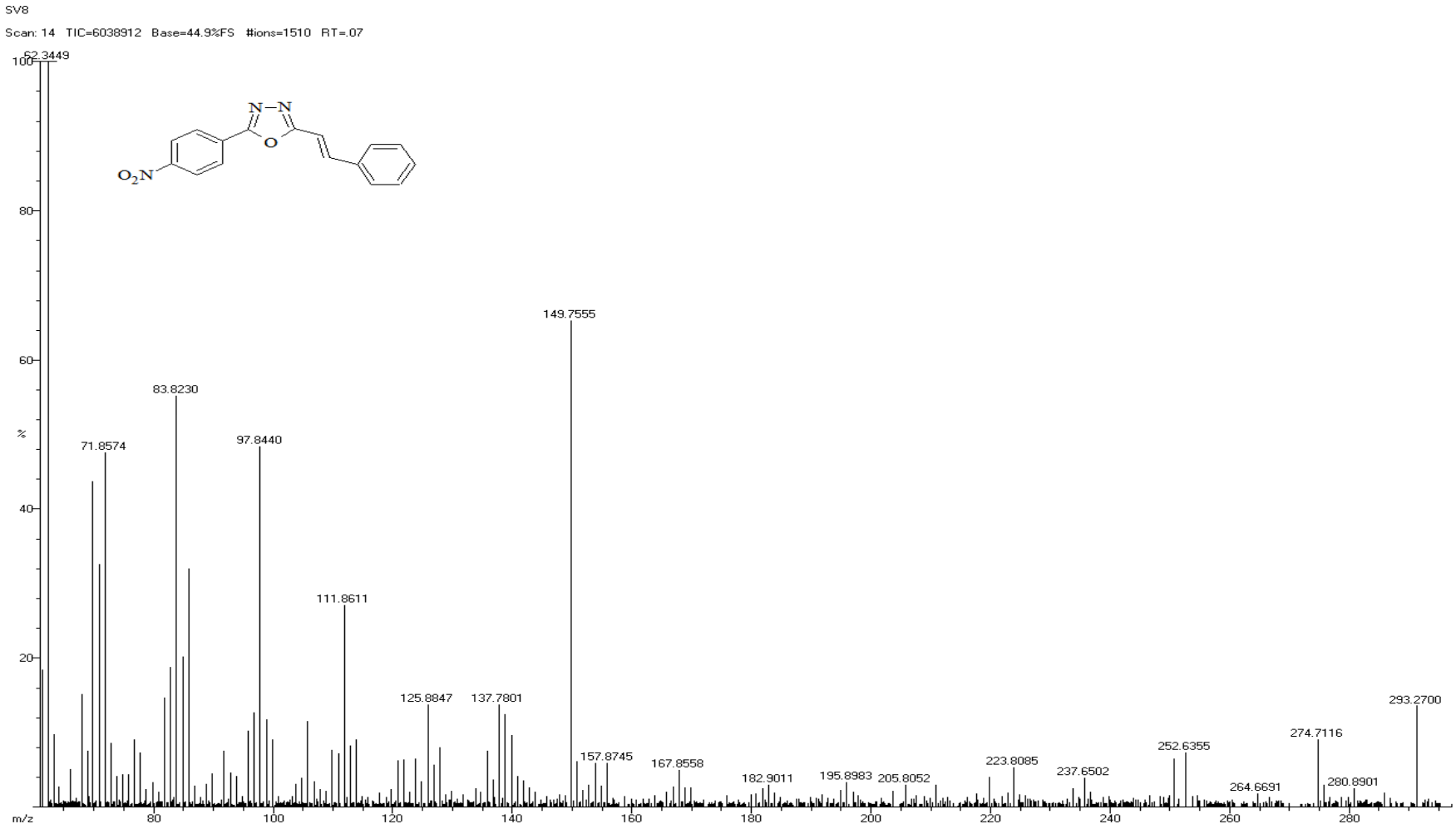
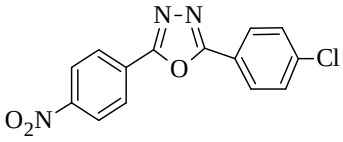


Figure 29: Mass Spectra of SV-8

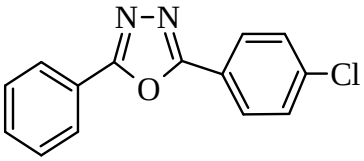
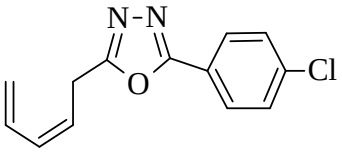
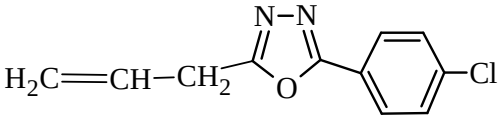
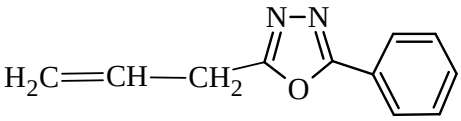
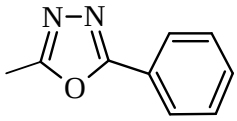
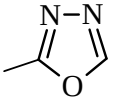
Interpretation of Mass spectra of SV-9



Molecular weight: 301.68 Molecular ion peak: 301.93

The possible fragments of the molecule with the relevant to its m/z values are:

Table-34

S.No	m/z	Fragments
1	256.20	
2	242.24	
3	218.93	
4	185.16	
5	159.15	
6	83.07	

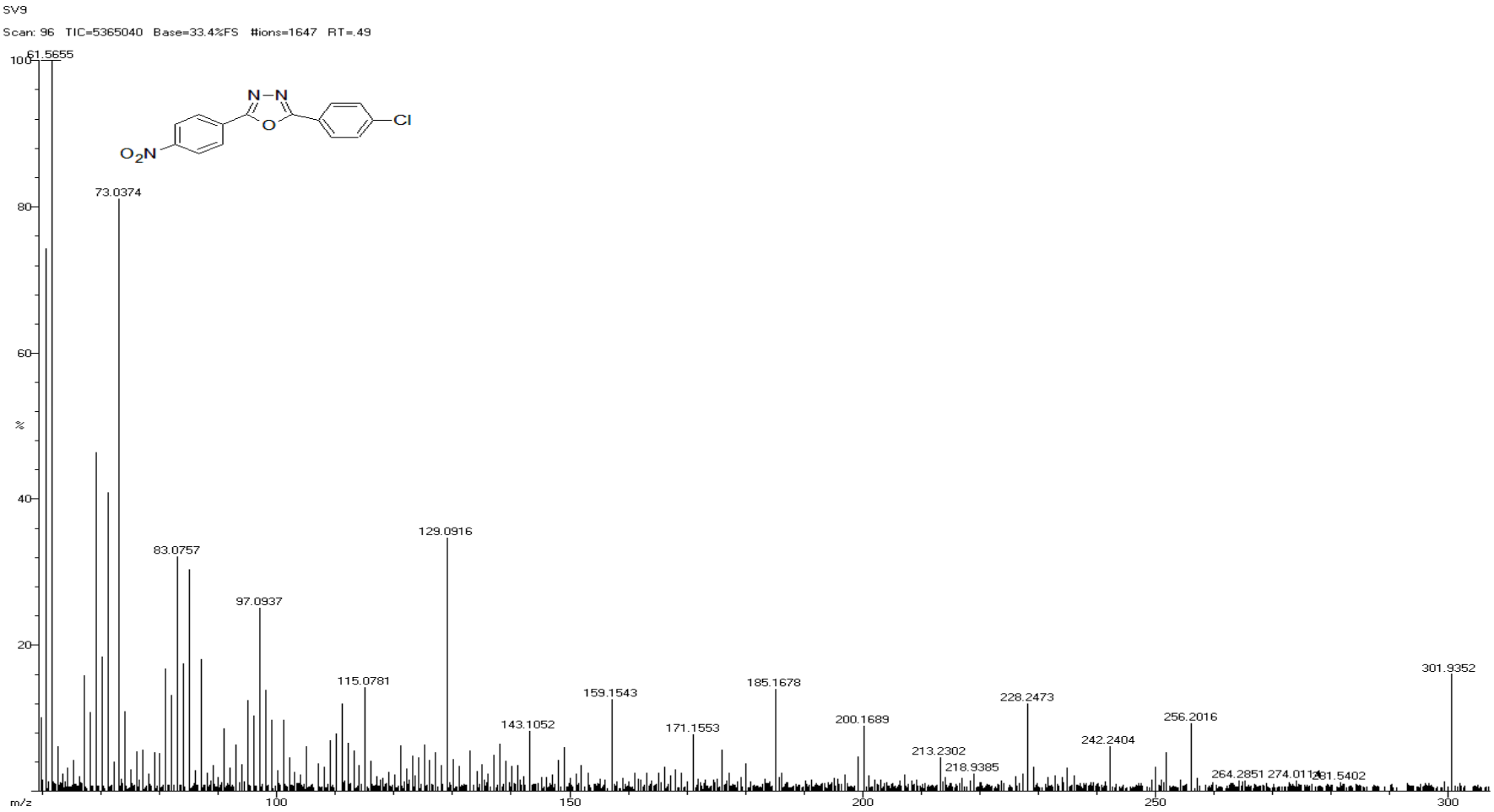
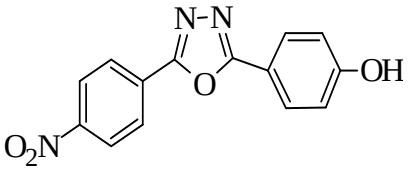


Figure 30: Mass Spectra of SV-9

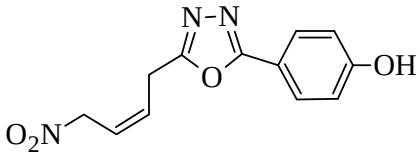
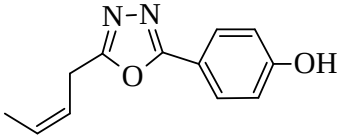
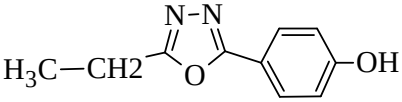
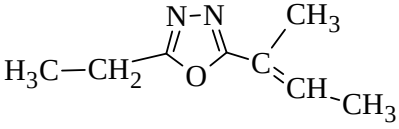
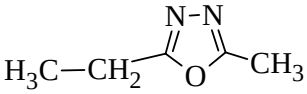
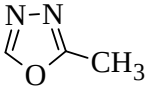
Interpretation of Mass spectra of SV-10



Molecular weight: 283.23 Molecular ion peak: 283.36

The possible fragments of the molecule with the relevant to its m/z values are:

Table-35

S.No	m/z	Fragments
1	257.65	
2	214.98	
3	187.12	
4	149.05	
5	110.12	
6	83.08	

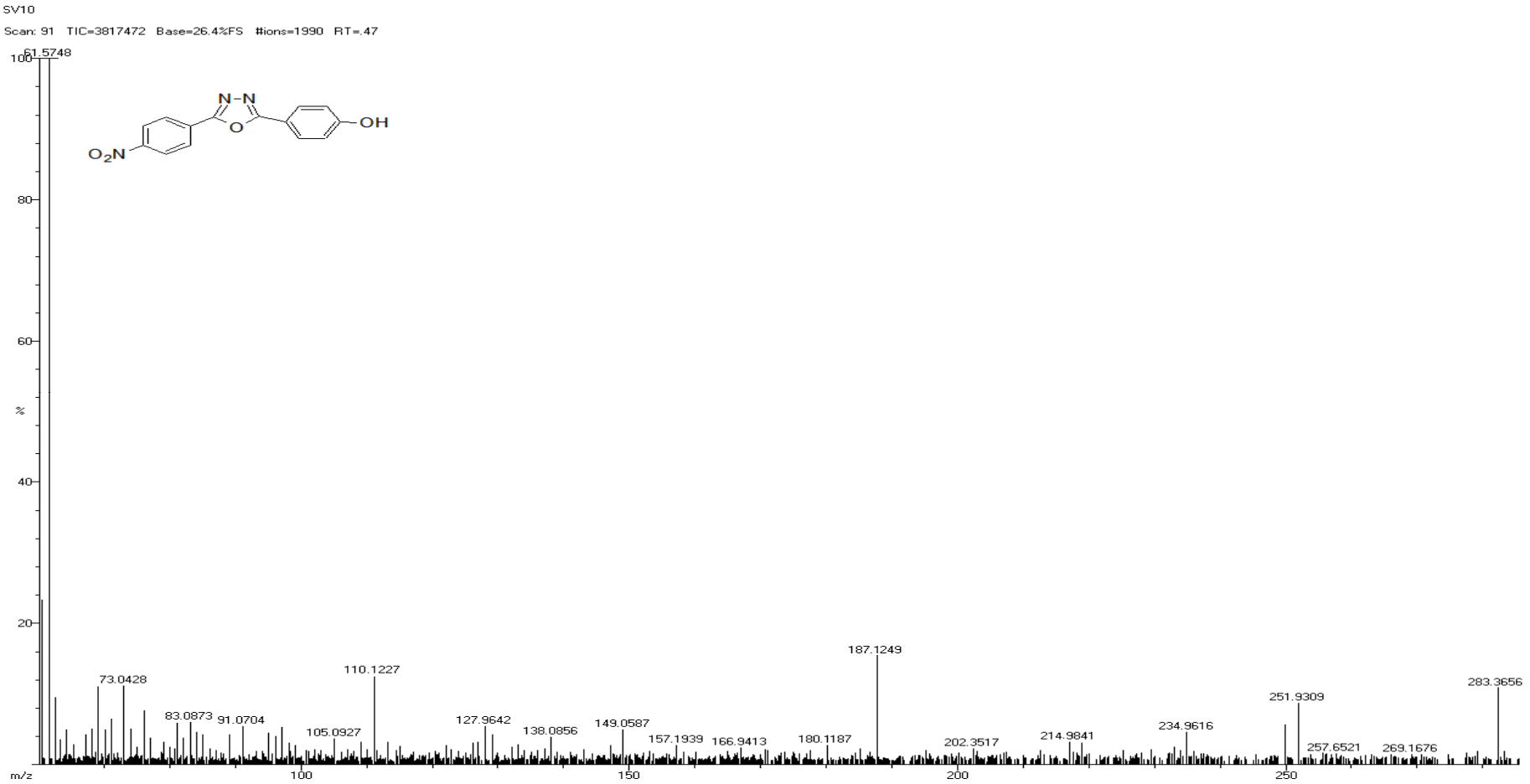


Figure 31: Mass Spectra of SV-10

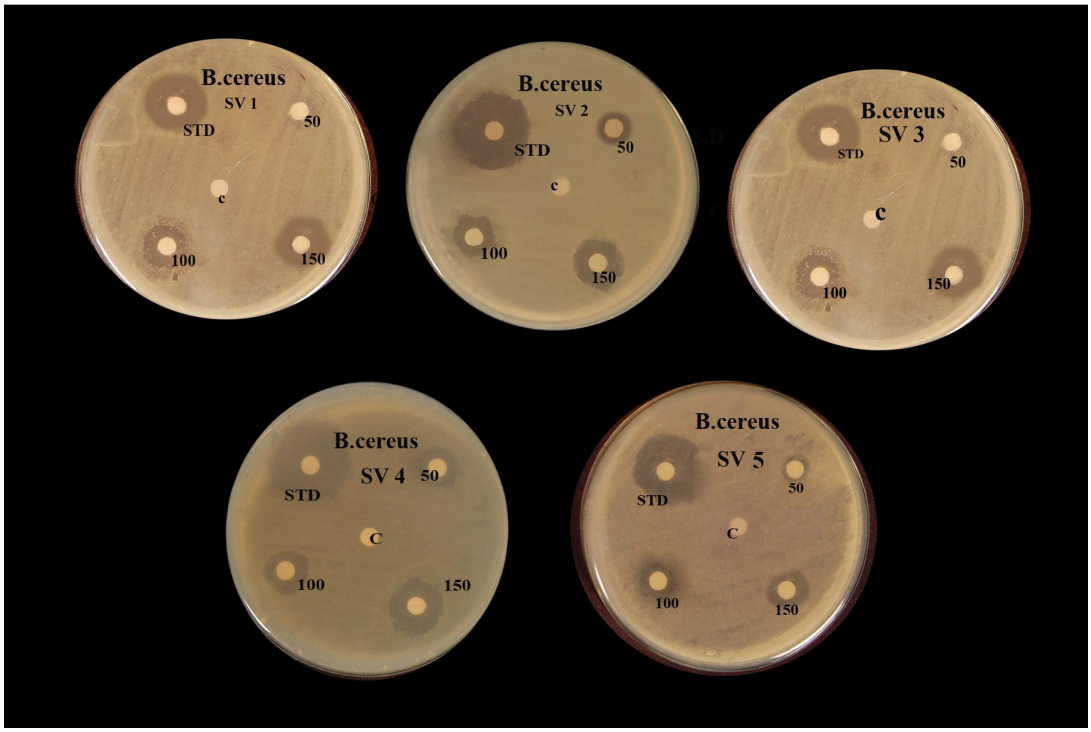
C. SCREENING OF ANTIMICROBIAL ACTIVITY

i) Antibacterial Activity

The antibacterial activity of the synthesized compounds was screened against both gram positive *Bacillus cereus* and gram negative *Pseudomonas aeruginosa* by disc diffusion method using ciprofloxacin as standard. The results are given below.

Table- 36 : Results of Antibacterial Activity

Compounds	Zone of Inhibition (in mm)					
	Bacillus cereus			***Pseudomonas aeruginosa***		
	50 (µg/ml)	**100 (µg/ml)**	**150 (µg/ml)**	**50 (µg/ml)**	**100 (µg/ml)**	**150 (µg/ml)**
SV-1	13	15	21	12	14	21
SV-2	14	16	19	13	15	18
SV-3	12	15	19	12	16	20
SV-4	15	18	21	15	18	24
SV-5	13	16	20	14	16	20
SV-6	12	15	18	15	17	20
SV-7	14	16	19	16	18	22
SV-8	13	15	18	15	19	23
SV-9	15	18	20	13	19	22
SV-10	16	19	21	15	18	25
Ciprofloxacin (10µg/ml)	39			38		



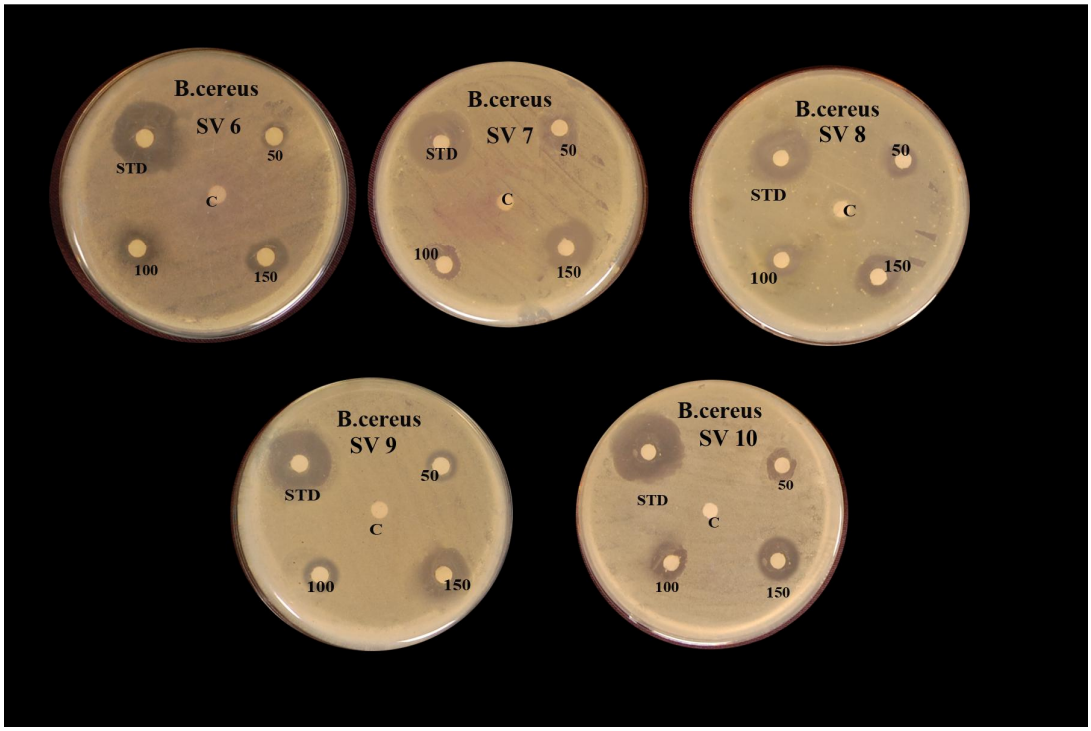


Figure 32: Antibacterial activity of synthesized compounds against *Bacillus cereus*

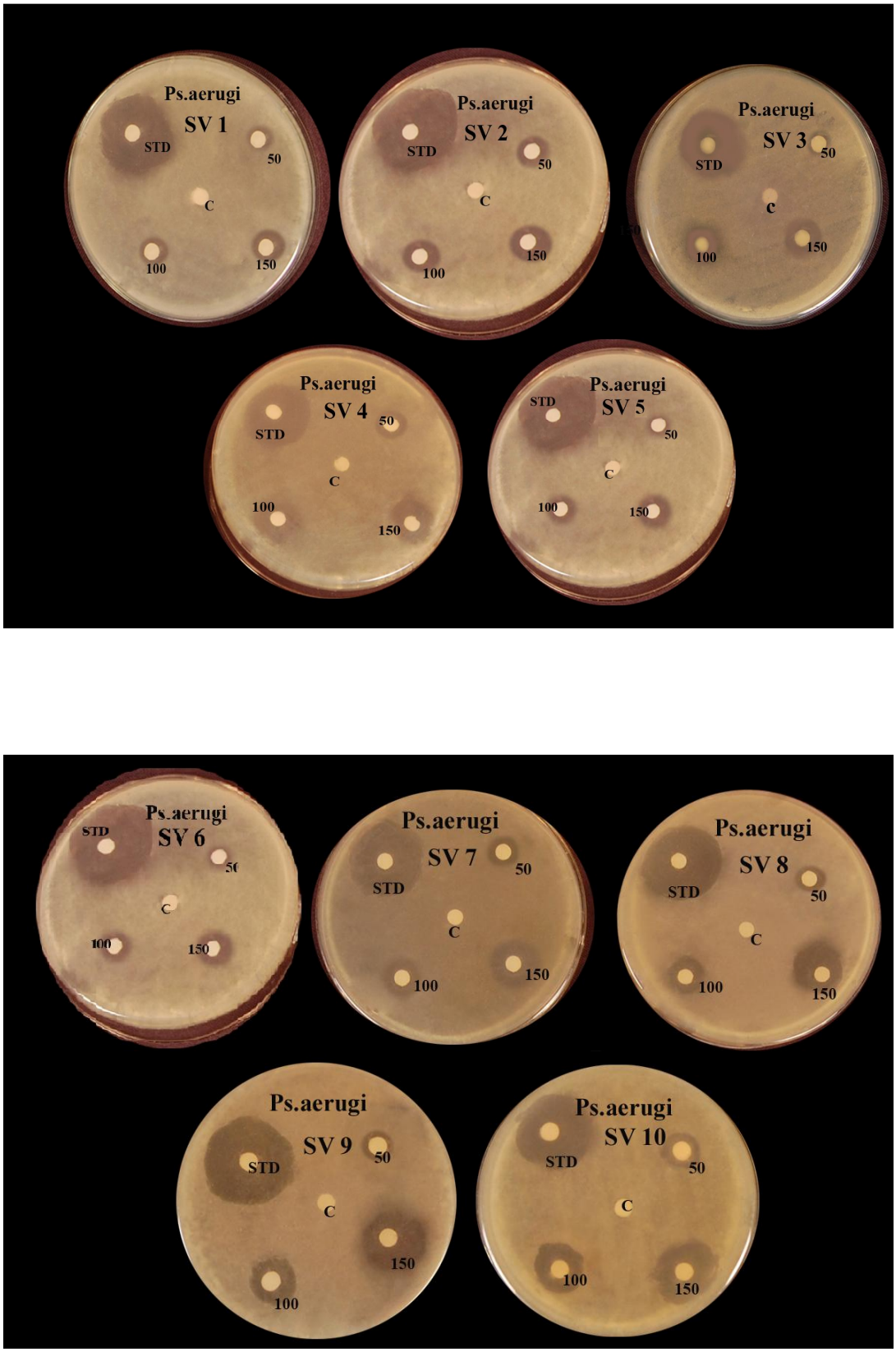


Figure 33: Antibacterial activity of synthesized compounds against *Pseudomonas aeruginosa*.

ii) Antifungal Activity

The antifungal activities of synthesized compounds were screened against *Aspergillus fumigates* by disc diffusion method using ketoconazole as standard drug.

Table-37 : Results of Antifungal Activity

Compounds	**Zone of Inhibition (in mm)**		
	Aspergillus fumigates		
	50 (µg/ml)	**100 (µg/ml)**	**150 (µg/ml)**
SV-1	16	18	20
SV-2	20	24	26
SV-3	15	18	20
SV-4	16	21	22
SV-5	14	19	21
SV-6	13	17	20
SV-7	15	20	22
SV-8	18	22	26
SV-9	16	22	24
SV-10	15	19	23
Ketoconazole (10µg/ml)	39		

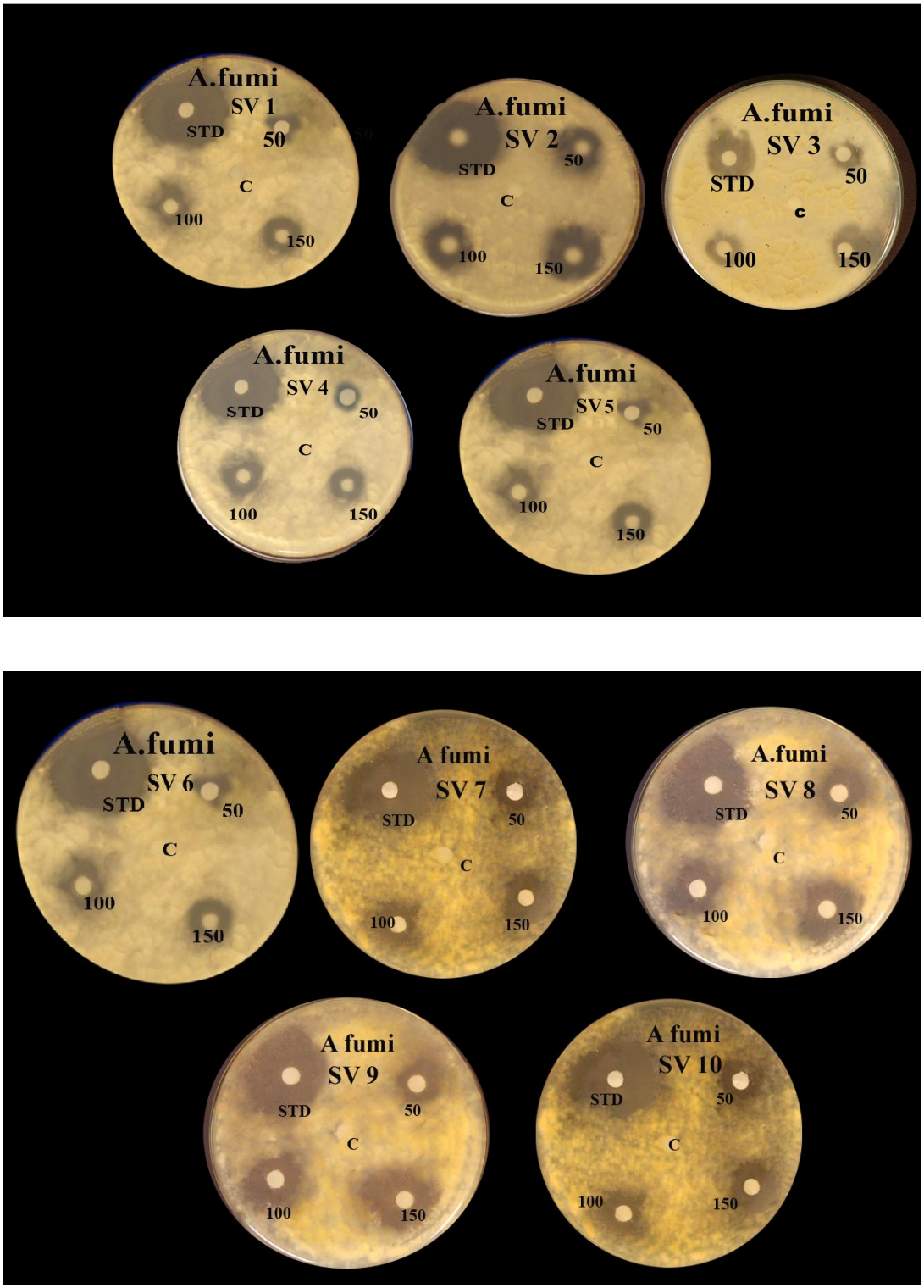


Figure 34: Antifungal activity of synthesized compounds against *Aspergillusfumigates.*

D. SCREENING OF ANTIINFLAMMATORY ACTIVITY

The antiinflammatory activity of the synthesized compounds (SV-3 and SV-4) was screened by carragenan induced rat paw odema method by using diclofenac sodium as standard. The results are given below:

Table -38 : Results of Antiinflammatory Activity

GROUP	SAMPLE	Paw edema Volume (in ml) at (mean ± SEM)			
		0 min	60 mins	120 mins	180 mins
I	Control	0.33±0.066	0.9±0.088	1.1±0.088	1.47±0.057
II	Standard (20mg/kg)	0.23±0.033	0.46±0.066*** (48.88)	0.53±0.088** (51.51)	0.55±0.082*** (62.14)
III	SV-3 (50mg/kg)	0.33±0.066	0.46±0.034* (48.81)	0.56±0.034** (52.09)	0.63±0.033*** (57.14)
IV	SV-4 (50mg/kg)	0.36±0.069	0.56±0.033* (33.63)	0.73±0.034* (37.77)	0.83±0.063*** (43.53)

n = 3 in each group.*p< 0.05, **p < 0.01 and ***p<0.001 when compared to control (One-way ANOVA followed by Bonferroni test)

Figures in the parenthesis indicate % inhibition of paw edema

DISCUSSION

The purity of the synthesized compounds was determined by their sharp endpoints and thin layer chromatography on pre-coated silica gel GF 254 plates using ethyl acetate - hexane as mobile phase. The purification was done by recrystallisation using methanol as solvent. The structures were confirmed by IR, proton NMR, and Mass spectroscopic methods.

Antiinflammtory activity

The synthesized compounds (SV-3 and SV-4) were tested for antiinflammatory activity by carragenan induced rat paw edema method using diclofenac sodium as standard. The activity was observed at 0, 60, 120 and 180 minutes.

The standard diclofenac sodium decreases the rat paw edema volume gradually at 180 minutes. Also, the compounds SV-3 and SV-4 shown moderate activity compared to that of standard but the compound SV-3 shows better activity than SV-4. The one way ANOVA followed by Bonferoni test was applied to find the results are statiscally or not used in population.

Antimicrobial activity

The antibacterial activity of the synthesized compounds was screened by disc diffusion method at the concentrations of 50, 100 and 150µg/ml against both gram positive *Bacillus cereus* and gram negative *Pseudomonas aeruginosa* using Muller Hinton agar media and the antifungal activity against *Aspergillus fumigates* by using Sabourand's agar media. The diameter of zone of inhibition were observed in mm and recorded. All the synthesized compound shows moderate activity at three different concentrations.

Antibacterial activity

Among the synthesized compounds screened for antibacterial activity SV-1, SV-4, SV-9 and SV-10 shown good antibacterial activity against gram positive organisms and the compound SV-4, SV-8, SV-9 and SV-10 could show better action against gram negative organisms. The rest of the compounds could show moderate to poor activity compared to 10µg/ml of ciprofloxacin as standard.

Antifungal activity

The antifungal activity of the synthesized compound was screened by disc diffusion method at three different concentrations. The compound SV-2, SV-8, SV-9 and SV-10 shows good antifungal activity and the remaining compounds show moderate to poor activity compared to 10µg/ml of ketoconazole as standard. All the compounds shown antifungal activity increases as the concentration increases.

SUMMARY

VI. SUMMARY

The ten derivatives of various substituted 1,3,4-oxadiazoles were synthesized with satisfactory yields. In our study, the 2,5-disubstituted-1,3,4-oxadiazoles were prepared from various aryl hydrazides react with different aromatic acids using $POCl_3$ as dehydrating agent. The aryl hydrazides were obtained by reacting respective esters with hydrazine hydrate.

A set of mannich bases of 1,3,4-oxadiazoles were prepared from 2-thione-1,3,4-oxadiazole derivatives. The thione derivatives were obtained from various aryl hydrazides reacting with carbondisulphide and potassium hydroxide.

The synthesized compounds melting points were taken in open capillary tubes and were uncorrected and purity was monitored by thin layer chromatography on pre-coated silica gel GF 254 plates using ethyl acetate-hexane as mobile phase. The purification was done by recrystallisation using methanol as solvent. The structures of the synthesized compounds were confirmed by IR, proton NMR, and Mass spectroscopic methods.

The synthesized compounds (SV-3 and SV-4) were tested for their antiinflammatory activity by carragenan induced rat paw edema method and the results of the compounds were compared with the standard drug diclofenac sodium. The compound SV-3 could show more activity than SV-4. The one way ANOVA followed by Bonferroni test was applied to find out the results are statistically significant or not in the used population.

The synthesized compounds were tested for antibacterial and antifungal activity using the standard drug Ciprofloxacin and Ketoconazole respectively. The antibacterial activity was screened against gram positive *Bacillus cereus* and gram

negative *Pseudomonas aeruginosa* and the antifungal activity was screened against *Aspergillus fumigates* by disc diffusion method at three different concentrations. All the compounds show moderate antibacterial and antifungal activities.

CONCLUSION

VII. CONCLUSION

The 2,5-disubstituted-1,3,4-oxadiazoles and mannich bases of 1,3,4-oxadiazoles shown moderate antibacterial and antifungal activities. Among the ten compounds synthesized only two were tested for antiinflammatory activity and it could show significant action compared to the standard. Hence less information is obtained in terms of antiinflammatory action for the synthesized compounds. On antimicrobial screening, though the compounds could show activities against bacterial and fungal strains but it is less compared to standard. Further research is needed to correlate the 1,3,4-oxadiazole with its antiinflammatory and antimicrobial activities.

BIBLIOGRAPHY

VIII. BIBLIOGRAPHY

- AbuZaid M. A., Telbani E. M., Elgemie G. H. and Nawwar G.A. Synthesis and invitro antitumor activity of new oxadiazole thioglycoside, *European Journal of Medicinal Chemistry,* **2011**, 46, 229-235.
- Aboria A.S., Abdel Rahman H. M., Mahfouz N. M. and Gendy M. A.Synthesis of some novel 5-(2-hydroxyphenyl)-3-substituted-2,3-dihydro-1,3,4-oxadiazol-2-thione derivatives Promising anticancer agents, *Bioorganic and Medicinal Chemistry Letters,* **2006**, 14, 1236–1246.
- Afaf H., Fahmy H. H. and Ali Abdelwahed S. H. Synthesis and antimicrobial activity of some new benzimidazole derivatives, *Molecules*, **2005**, 1429-1438.
- Ashutoskar. Medicinal Chemistry, 3rd ed, New Age International Private Limited, New Delhi. **2005**, 477-500.
- Asif Husian and Mohammed Ajmal. Synthesis of novel 1,3,4-oxadiazole derivatives and their biological properties, *Acta Pharma,* **2009**, 59, 223-233.
- Biju C.R., Manju P., Biju K. and Rekha K. Insilico design synthesis and screening of novel 1,3,4-oxadiazole derivatives for analgesic, antiinflammatory and antimicrobial activity, *International Journal of Biomedical Research,* **2010**, 1, (3), 109-123.
- Choudhary S. K., Choudhary M., Choudhary A. and Parmar S. S. Anticonvulsant and antiproteolytic properties of 3,5-disubstituted oxadiazol-2-thiones and their inhibition of respiration in rat brain homogenates. *Journal of Pharmaceutical Sciences,* **1978**, 67**, (**11**),** 1507–1509.

- Ferray Aydogan., Zuhal Turgut., Nuket Ocal and Sayesag Erde. Synthesis and electronic structure of new aryl- and alkyl-substituted 1,3,4-oxadiazol-2-thione derivatives, *Turkey Journal of Chemistry*, **2002**, 26, 159-169.
- Furnis B. S., Hannaford A. J., Smith P.W.J. and Tatchwel. Vogel's Textbook of Practical Organic Chemistry, 5th ed, Dorling Kindersley(India) Private Limited, New Delhi. **2008**, 1077-1078.
- Gheorghe R., Costel A., Ileana M., Eugenia C. and Lucia D. Synthesis of new mannich bases derived from 5-phenyl-1,3,4-oxadiazol-2-thione and investigation of the conformational isomers of dimethyl 5-(5-phenyl-2-thioxo-1,3,4-oxadiazole-3-methylamino)isophthalate, *Revue Roumanie de chimie*, **2007**, 52, (12), 1103-1110.
- Girges M. M. Synthesis and pharmacological evaluation of novel series of sulfonate ester containing 1,3,4- oxadiazole derivatives with anticipated hypoglycaemic activity. *European Journal of Medicinal Chemistry.* **1994,** 44, 490-494.
- Gupta R.R., Kumar M. and Gupta V. Heterocyclic Chemistry-II. 2nd ed, Springer (India) private limited, New Delhi. **2009**, 525-546.
- Hakan Bektas., Nesrin karaali., Deniz Sahin., Ahmet Demirbas., Sengul Alpay Karagolu and Neslihan Demirbas. Synthesis and antimicrobial activities of some new 1,2,4-triazole derivatives, *Molecules*, **2010**, 15, 427-2438.
- Harish R., Murali Dhar K. and Pradeep M. Synthesis of some new oxadiazole and oxadiazoline analogues for their antiinflammatory activity, *The Pharmaceutical Society of Japan*, **2007**, 10, 1757-1764.

- Husian A., Priyanka A. and Sarafoz. Synthesis and biological evaluation of β-aroyl propionic acid based 1,3,4-oxadiazoles, *Indian Journal of Pharmaceutical Sciences*, **2010**, 62-66.
- Illango K., Valentina P., Umarani N. and Kumar T. Synthesis and Characterisation of 2,5-Disubstituted -1,3,4-oxadiazoles as potent antiinflammatory agents, *Jounal of Young Pharma*, **2010**, 1, (1), 72-76.
- Ilango K. and Valentina P. Text Book of Medicinal Chemistry, 1st ed, Vol 1, Keerthi Publishers, Chennai. **2007**, 336-339.
- Jag Mohan. Organic Spectroscopy, Principles and Applications, 2nd ed, Narosa publishing home, New Delhi. **2005,** 76-95.
- John A Joule and Kirth Mills. Heterocyclic Chemistry at a glance, 1st ed, Blackwell publishing Limited, U.K. **2007,** 96-101.
- Kee Jung Lee., Chang Hoon Lee and Hyun In Cho. Synthesis of 1,3,4-oxadiazole having phenol or thiophenol group, *Bulletin of the Korean Chemical Society,* **2001**, 22, (10), 1153-1155.
- Keshari Kishore Jha., yatendra kumar and Shaharyar M. Design synthesis and Biological evaluation of 1,3,4-oxadiazole derivatives, *Asian Journal of Chemistry*, **2009**, 21, (9), 7403-7410.
- Koparir M., Cetin A. and Cansiz A. 5-Furan-2yl[1,3,4]oxadiazol-2-thiol,5-Furan-2-yl-4H[1,2,4]triazol-3-thiol and their thiol-thione tautomerism, *Molecules*, **2005**, 10, 475-480.
- Kulkarni S. K. Hand Book of Experimental Pharmacology, 3rd ed, Vallabh Prakashan, Delhi. **2007,** 128-130.

- Kumar A., Singh S., Verma M., Saxena A. K. and Shankar K. Potent anti-inflammatory activity of 2-(*o*-hydroxyphenyl)-5-(*p*dimethylaminophenyl)-1,3,4-oxadiazoles. *Indian Jounal of Pharmaceutical Sciences*, **1987**, 49, (6), 201–204.
- Manish srivastava., Deepak singh., Aashish singh Kushwah and Gokulan P.D. Synthesis and Biological evaluation of some new 1,3,4-oxadiazole derivatives, *Journal of Current Pharmaceutical Research*, **2011**, 4, 20-24.
- Mohammad Amir., Sadique A Javed and Harish Kumar. Synthesis of some newer analogues of 4-hydroxyphenyl acetic acid as Potent Antiinflammatory agents**,** *Journal of the Chinese Chemical Society*, **2008**, 55, 201-208.
- Mohamed A. and Omar A. Synthesis and antimicrobial activity of some new 5-(2 thienyl)-1,2,4-triazoles and 5-(2-thienyl)-1,3,4-oxadiazoles and related derivatives, *Molecules*, **2010**, 15, 502-514.
- Mohammad Shaharyar and Mohamed Ashraf Ali. Oxadiazole mannich bases Synthesis and antimycobacterial activity, *Bioorganic and Medicinal Chemistry Letters*, **2007**, 1-3.
- Nadia Salih., Jumat Salimon., Ayad Hameed and Emad Yousif. Synthesis Characterisation and antimicrobial evaluation of some 1,3,4-oxadiazole derivatives, *International Journal of Pharm Tech Research*, **2011**, 3, (2), 1097-1102.
- Nagalakshmi G. Synthesis antimicrobial and antiinflammatory activity of 2,5-disubstituted-1,3,4-oxadiazoles, *International Journal of Pharmaceutical Sciences*, **2008**, 70, (1), 49-55.

- Nath S. C. Synthesis of 3,5-disubstituted 1,3,4-oxadiazol-2-thiones as potential fungicidal agents, *Journal of Heterocyclic Chemistry*, **1984,** 21**,** 205–208.
- Nigam R., Swarup S., Saxena V. K., and Singh H. K. Synthesis and pharmacological screening of some new 2- thio-3-(substituted-aminomethyl)-5-(3,5-dinitrophenyl)-1,3,4-oxadiazoles, *Journal of Indian Chemical Society*, **1992,** 69, 692–693.
- Niti Bharadwaj., Saraf S.K., Pankaj Sharma and Pradeep Kumar. Synthesis evaluation and characterisation of some 1,3,4-oxadiazoles as antimicrobial agents, *E-Journal of Chemistry*, **2009**, 6, (4), 1133-1138.
- Palak K Parikh., Hiren M Marvaniya and Dhrubo Jyoti Se. Synthesis and biological evaluation of 1,3,4-oxadiazole derivatives as potent antibacterial and antifungal activities, *International Journal of Drug Devolopment and Research*, **2011**, 1, (3), 248-255.
- Parmar N. S. and Shiv prakash. Screening Methods in Pharmacology, Narosha Publishing House Private Limited, New Delhi. **2009**, 213-221.
- Priya V Frank., Girish K.S. and Balakrishna Kalluraya. Solvent-freee microwave-assisted synthesis of oxadiazoles containing imidazole moiety, *Journal of Chemical Sciences*, **2007**, 119, (1), 41-46.
- Raj K Bansal. Organic Name Reaction, 3rd ed, MC Graw-Hill publishing Company Limited, New Delhi. **2006,** 225-227.
- Rakesh Saini, Awani K Rai., Kesari A.N. and Shahar Yar. Synthesis and biological evaluation of 2,5-disubsstituted 1,3,4-oxadiazoles, *Asian Journal of Resaearch in chemistry*, **2009**, 2, (1), 34-36.

- Ravindra K.C., Vagdevi H.M., Vaidya V.P. and Basavaraj P. Synthesis antimicrobial and antiinflammatory activities of 1,3,4-oxadiazoles linked to naptho[2,1-b]furan, *Indian Journal of chemistry*, **2006**, 45B, 2506-2511.
- Rina Das and Dinesh Kumar Mehta. Synthesis, antiinflammatory and antimicrobial activity of some 2,5-disubstituted -1,3,4-oxadiazoles, *Asian Journal of Chemistry*, **2009**, 21, (6), 4419- 4424.
- Sarangapani Manda., Rajyalakshmi Gudipati and Rama Narshima Reddy Anreddy. Synthesis characterisation and anticancer activity of certain 3-{4-(5-mercapto-1,3,4-oxadiazol-2-yl)phenylimino}indolin-2-one derivative, *Saudi Pharmaceutical Journal*, **2011**, 19, 153-158.
- Selvakumar K., Anandarajagopal K., Rajamanickam V., Ajaykumar T. and Jesindha B. Synthesis characterisation and antimicrobial evaluation of 5-(2-amino phenyl) 1,3,4-oxadiazole-2-(3H)-thione derivatives, *International Journal of Pharmaceutical Sciences Review and Research*, **2011**, 6, (1), 64-67.
- Sengupta A.K., Garg M., and Chandran U. Synthesis of some new mannich bases derived from substituted1,3,4- oxadiazole-2-thiones and their biological activities, *Journal of Indian Chemical Society,* **1979**, 56**,** 1230–1232.
- Sharma V.K., Sudhir Bhardwaji., Bharat Parashar and Narendra Parashar. Microwave assisted synthesis and pharmacological evaluation of some 1,3,4-oxadiazole derivatives, *Archieves of Applied Science Research*, **2011**, 3, (2), 558-567.
- Sharma Y. R. Elementary Organic Spectroscopy, 4th ed, S. Chand and company Limited, New Delhi. **2007,** 210-233.

- Shashikant R Pattan., Hariprasad C.K., Nachiket S Dighe., Bhawar S.B., Hiremath S.V. and Ingalagi B. Synthesis and evaluation of some new quinoline derivatives for their antiinflammatory activities, *International Journal of Pharmaceutical Research and Development*, **2009**, 1, (9), 1-10.
- Shashikant R Pattan., Rabara P.A., Jayashri S Pattan., Bukitgar A., Wakale S. and Musmade D.S. Synthesis and evaluation of some novel substituted 1,3,4-oxadiazole and pyrazole derivatives for antitubercular activity, *Indian Journal of Chemistry*, **2009**, 48B, 1453-1456.
- Somani R. R., Kadam G., Vohra R., Vijayaraghavan S. and Shirodkar P.Y. Studies of CNS activities of some mannich bases of 1,3,4-oxadiazole, *International Journal of Pharmacology*, **2010**, 6, (5), 696-704.
- Somani R. R., Vijayaraghavan S., Shirodkar P.Y. and Kadam V. J. Microwave assisted synthesis and antimicrobial activity of some newer mannich bases, *International Journal of Pharmtech Research*, **2009**, 1, (3), 811-815.
- Soni N., Barthwal J. P., Saxena A. K., Bhargava K.P. and Parmar S. S. Monoamine oxidase and succinate dehydrogenase inhibitory properties of substituted 1,3,4-oxadiazol-2-thiones, *Jounal of Heterocyclic Chemistry*, **1982,** 19**,** 29–32.
- Subramanya Bhat K., Kartikeyan M.S., Shivarama Hola and Suchetha Shely N. Synthesis of some new fluorine containing 1,3,4-oxadiazole derivatives as potential antibacterial and anticancer agents, *Indian Journal of Chemistry*, **2004**, 43 B, 1765-1769.

- Sujatha K., Khadar A.M., Balakrishna Kalluraya and Babu M. Synthetic studies on 1,3,4-oxadiazoles, *Indian Journal of Heterocyclic Chemistry*, **2009**, 19, 33-36.
- Thomas L Gilchrist. *Heterocyclic Chemistry*, 3rd ed, Dorling Kindersley(India) private Limited, South Asia. **2007**, 335-339.
- Vijay V Dabholakar and Nitin V Bhusari. Synthesis of 2-substituted-1,3,4-oxadiazole derivatives, *International Journal of Chemical, Environmental and Pharmaceutical Research*, **2011**, 2, (1), 1-4.
- Virginija Jakubkiene., Milda Malvina Burbuliene., Giedrute Mekuskiene., Emilija Udrenaite., Povilas Gaidelis and Povila Vainilavicius. Synthesis and antiinflammatory activity of 5-(6-methyl-2-substituted-4-pyrimidinyl oxy methyl)-1,3,4-oxadiazol-2-thiones and their 3-morpholinomethyl derivatives, *Il farmaco*, **2003**, 58, 323-328.

ANNEXURE

ANNEXURE -1

CERTIFICATE

This is to certify that the project title ..... Anti - inflammatory ..... activity studies of Mannich Bases of 1,3,4 oxadiazole derivatives have been approved by the IAEC.

S.SHOBA
Name of Chairman/member Secretary IAEC:

Dr. P. Balakrishnamurthi
Name of CPCSEA nominee:

Signature with date
22/7/11
Chairman/Member Secretary of IAEC

1 Aug 11
CPCSEA nominee:

(Kindly make sure that minutes of the meeting duly signed by all the participants are maintained by office)

Dr.T.Vetrichelvan, M.Pharm., Ph.D.,
Professor and Principal
Adhiparasakthi College of Pharmacy
Melmaruvathur-603 319
Tamilnadu, India

ANNEXURE -2

Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2012, 4(2):1217-1221

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

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

A. Thirugnanasambanthan*, S. Venkatraman and M. Senthil Palaniappan

Department of Pharmaceutical Chemistry, Adhiparasakthi College of Pharmacy, Melmaruvathur, Tamilnadu, India

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.