BioGecko

Vol 12 Issue 01 2023
ISSN NO: 2230-5807

SYNTHESIS AND EVALUATION OF SOME NOVEL HETEROCYCLES OF BIOLOGICAL INTEREST

Ms. Shital Dnyaneshwar Gaikwad*
Bhagwant University, Ajmer Rajasthan, 305001.

Dr. Ravindra B. Laware
Department of Pharmaceutics, Pravara Rural College of Pharmacy, Loni.413736.

ABSTRACT

The present research work is aimed to synthesize some novel substituted 1, 4- dihydropyrimidones. The twelve new derivatives of 1, 4- dihydropyrimidones were synthesized during the course of research work. The structures of these compounds have been established by means of IR, ^{1}H-NMR.The compounds were synthesized by using "Biginelli's reaction". The dihydropyrimidones, thus, prepared were treated with hydrazine hydrate to give respective hydrazides, which were then converted to thiadiazoles, oxadiazoles & pyrazoles. These derivatives were subjected to anticancer & antioxidant activity.A_2& A_3 have shown excellent anticancer activity as compared to the standard drug cyclophosphamide. Compounds **A_2, B_1, B_3, C_2 and C_3** have shown promising antioxidant activity at 250µg/mL, while the remaining compounds have also shown moderate antioxidant activity, when compared with standard drug ascorbic acid.The dihydropyrimidones were also treated with fenofibrate to give combination product. This was subjected to calcium antagonist & antihyperlipidemic activity.Compounds **D_2& F_2**have shown promising anticholesterol activity. Compounds **D_1 & F_2** have shown promising anti-triglyceride activity.Compounds **D_1 & F_2** have shown promising relaxing activity as compared to standard Nifedipine.

Key Words: 1, 4- dihydropyrimidones;anticancer; antihyperlipidemic; calcium channel antagonists & antioxidant.

INTRODUCTION

The chemistry and biological study of heterocyclic compounds has been an interesting field for a long time in medicinal chemistry. Heterocyclic compounds are very widely distributed in nature and are essential to life. A heterocyclic compound is one which possesses a cyclic structure with at least two different kinds of hetero atoms in ring. A number of heterocyclic derivatives containing nitrogen and sulphur atom serve as a unique and versatile scaffolds for experimental drug design. Nitrogen, oxygen, and sulphur are the most common heteroatoms.

Most of the sugars and their derivatives, including vitamin C, for instance, exist in the form of five-member (furan) or six-member (pyran) rings containing one oxygen atom. Most member of vitamin B group possess heterocyclic ring containing nitrogen. One example is vitamin B6 (pyridoxine), which is a derivative of pyridine, essential in amino acid metabolism.

All these natural and synthetic heterocyclic compounds can and do participate in chemical reactions in the human body. All the biological processes are chemical in nature. Such fundamental manifestations of life as the provision of energy, transmission of nerve impulses, sight, metabolism and the transfer of hereditary information are all based on chemical reactions involving the participation of many heterocyclic compounds, such as vitamins, enzymes, coenzymes, nucleic acids, ATP and serotonin. Heterocycles are able to get involved in an extraordinarily wide range of reaction types. Depending on the pH of the medium, they may behave as acids or bases, forming anions or cations. Some interact readily with electrophilic reagents, others with nucleophiles, some are easily oxidized, but resist reduction, while others can be readily hydrogenated but are stable toward the action of oxidizing agents. Certain amphoteric heterocyclic systems simultaneously demonstrate all of the above-mentioned properties. The ability of many heterocycles to produce stable complexes with metal ions

has great biochemical significance. The presence of different heteroatoms makes tautomerism ubiquitous in the heterocyclic series. Such versatile reactivity is linked to the electronic distributions in heterocyclic molecules. Evidently, all the natural products and the synthetic drugs mentioned above are good examples of nature's preference for heterocycles whose biological activity cannot be determined by one or a combination of two or three of the above mentioned properties.

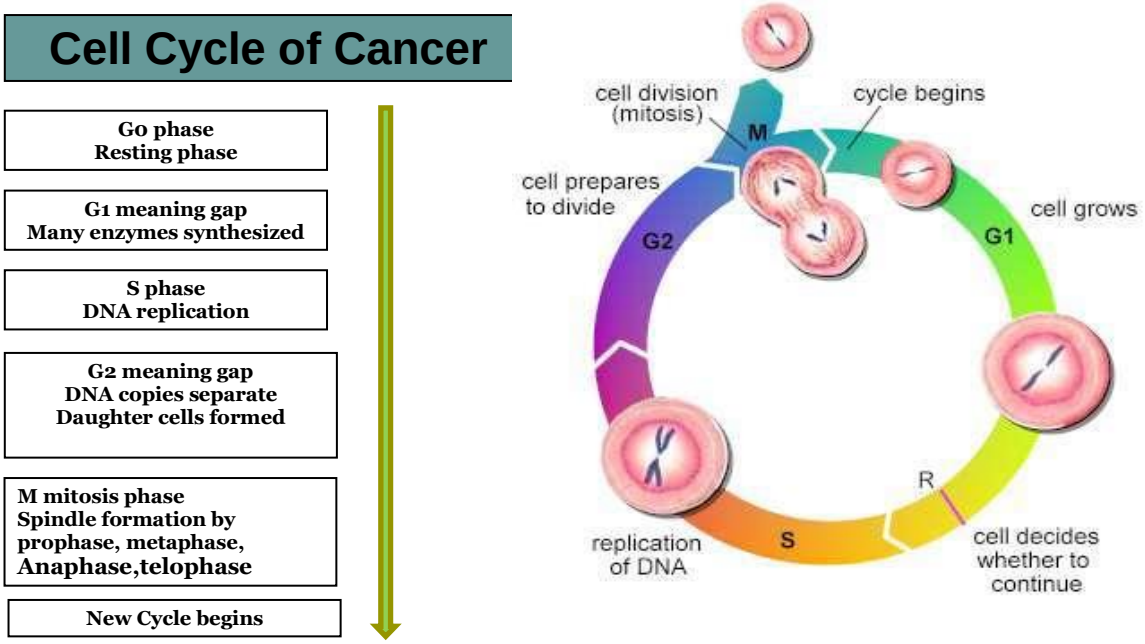
Pyrimidine (cytosine, thymine and uracil) and purine (adenine and guanine) derivatives are monocyclic and bicyclic heterocycles with two and four nitrogen atoms, respectively. They are key components of the deoxyribonucleic acid (DNA) molecules and participate directly in the encoding of genetic information. They also pass information to the related ribonucleic acid (RNA) molecules that control, in protein synthesis, the sequence of amino acids. The need for minute quantities of accessory dietary factors, the vitamins is well-known. Vitamins in the B group thiamine, folic acid, riboflavin, cyanocobalamine, are nitrogen-containing heterocycles and function either as coenzymes or their precursors. Other vitamins such as ascorbic acid (vitamin C) and tocopherol (vitamin E) are oxygen heterocycles.

Heterocycles are chemically more flexible and better able to respond to the many demands of biochemical systems. The constantly accelerating rate of research and development in heterocyclic chemistry suggested that enormous numbers of heterocyclic systems are well known and this number is increasing very rapidly. Heterocycles nucleus is present as a core structural component in an array of drug categories such as antimicrobial, anti-inflammatory, analgesic, antiepileptic, antiviral, antineoplastic, antihypertensive, antimalarial, local anaesthetic, antianxiety, antidepressant, antihistaminic, antioxidant, antitubercular, anti-Parkinson's, antidiabetic, antiobesity and immunomodulatory agents, and etc[1].

The impact of genomics and proteomics is additionally creating an explosion in the number of drug targets. Today's drug therapies are based solely on approximately 500 biological targets, while in 10 years' time, the number of targets could well reach 10,000. In order to identify more potential of drug candidates for all these targets, pharmaceutical companies have made major investments in high-throughput technologies for genomics and proteomics research, combinatorial chemistry andbiological screening. However, lead compound optimization and medicinal chemistry remain the bottlenecks in the drug discovery process. Developing chemical compounds with desired biological properties is time consuming and expensive. Consequently, increasing interest is being directed towards technologies that allow more rapid synthesis and screening of chemical substances to identify compounds with functional qualities.

CANCER

Today, the Greek term carcinoma is the medical term for a malignant tumor derived from epithelial cells. It is Celsus who translated *carcinos* into the Latin *cancer*, also meaning crab. Galen used "*oncos*" to describe *all* tumours, the root for the modern word oncology.[1] **Cancer** (medical term: malignant neoplasm) is a class of diseases in which a group of cells display *uncontrolled growth* (division beyond the normal limits), *invasion* (intrusion on and destruction of adjacent tissues), and sometimes *metastasis* (spread to other locations in the body via lymph or blood). These three malignant properties of cancers differentiate them from benign tumors, which are self-limited, and do not invade or metastasize. Most cancers form a tumor but some, like leukemia, do not. The branch of medicine concerned with the study, diagnosis, treatment, and prevention of cancer is oncology. Cancer affects people at all ages with the risk for most types increasing with age.[3] Cancer caused about 6 lakh deaths in India in 2011. Cancers are caused by abnormalities in the genetic material of the transformed cells.[6]these abnormalities may be due to the effects of carcinogens, such as tobacco smoke, radiation, chemicals, or infectious agents. Other cancer-promoting genetic abnormalities may randomly occur through errors in DNA replication, or are inherited, and thus present in all cells from birth. The heritability of cancers is usually affected by complex interactions between carcinogens and the host's genome.



ANTIOXIDANT

An **antioxidant** is a molecule capable of inhibiting the oxidation of other molecules. Oxidation is a chemical reaction that transfers electrons from a substance to an oxidizing agent. Oxidation reactions can produce free radicals. In turn, these radicals can start chain reactions. When the chain reaction occurs in a cell, it can cause damage or death. When the chain reaction occurs in a purified monomer, it produces a polymer resin, such as a plastic, a synthetic fibre, or an oil paint film. Antioxidants terminate these chain reactions by removing free radical intermediates, and inhibit other oxidation reactions. They do this by being oxidized themselves, so antioxidants are often reducing agents such as thiols, ascorbic acid or polyphenols. [15]

HYPERTENSION:

According to Medilexicon's medical dictionary, hypertension means "High blood pressure; transitory or sustained elevation of systemic arterial blood pressure to a level likely to induce cardiovascular damage or other adverse consequences."

Hypertension is classified as either primary (essential) hypertension or secondary hypertension; about 90–95% of cases are categorized as "primary hypertension" which means high blood pressure with no obvious underlying medical cause. The remaining 5–10% of cases (secondary hypertension) is caused by other conditions that affect the kidneys, arteries, heart or endocrine system.

Hypertension is a major risk factor for stroke, myocardial infarction (heart attacks), heart failure, aneurysms of the arteries (e.g. aortic aneurysm), and peripheral arterial disease and is a cause of chronic kidney disease. Even moderate elevation of arterial blood pressure is associated with a shortened life expectancy. Dietary and lifestyle changes can improve blood pressure control and decrease the risk of associated health complications, although drug treatment is often necessary in patients for whom lifestyle changes prove ineffective or insufficient.[42]

CALCIUM CHANNEL: [45]

A Calcium channel is an ion channel which displays selective permeability to calcium ions. It is sometimes synonymous as voltage-dependent calcium channel, although there are also ligand-gated calcium channels.

Types of Calcium Channels:

Three types of calcium channels have been identified voltage-sensitive, receptor operated (cardiac muscle &vascular smooth muscle) and stretch operated (in some blood vessels) channels. The regulation of calcium ions depends on both the entry and exit of calcium across the plasma membrane and on the sequestration and release of calcium within the cell. At the membrane level, calcium entry into the cell occurs partly through voltage gated calcium channels (VGCCs) which open when the cell membrane is depolarized. VGCCs belong to a family of homologous proteins that also includes channels for sodium and potassium. In addition, there are believed to be receptor-operated calcium channels (ROCCs), which are coupled to excitatory receptors either directly or via G-proteins and open in response to receptor ligands, such as noradrenaline acting on alpha1-adrenoceptor. In general, calcium channels are membrane-spanning, funnel-shaped glycoproteins that function like ion selective valves. They form a water-filled pore that open and close to permit calcium ions to move in the direction of its electrochemical concentration gradient. Each channel has outer and inner gates: the outer gates are specifically blocked by tetradotoxin in fast channels and by calcium channel blockers in slow channels. The inner gates, particularly in slow channels, appear to be dependent on the phosphorylation state of the membrane. The position of a channel gate, which is a portion at or near the inner side of the gate, indicates whether the channel is in the closed or opened state. Verapamil and diltiazem block slow channel conduction at the inner gate and possess some fast channel blocking activity as well. When conformational changes in the channel macromolecule occur, the activation and inactivation gates move into and out of an occluding position. This determines opening and closing of the channel pore. Calcium binding sites present in the pore ensure ion selectivity of the channels. Phosphorylation sites as well as drug and toxin binding sites of the channel macromolecule play important roles in the regulation of the channel. It should be emphasized that the exact macrostructure of the channel proteins, putative gates and other regulatory sites is unknown at this time. Though the direct evidence for ROCCs appears to be strong, they have so far eluded identification experimentally and some even doubt their existence. ROCCs do not appear to be targets for any of the known types of calcium antagonists which act only on VGCCs.

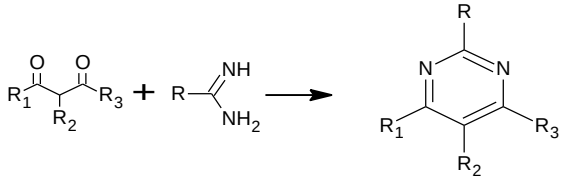
HYPERLIPIDEMIA: [58, 59]

Hyperlipidemia a broad term, also called hyperlipoproteinemia, is a common disorder in developed countries and is the major cause of coronary heart disease. It results from abnormalities in lipid metabolism or plasma lipid transport or a disorder in the synthesis and degradation of plasma lipoproteins. The term “dyslipidaemia” now a days is increasingly being used to describe abnormal changes in lipid profile, replacing the old term hyperlipidaemia. Hyperlipidemia means abnormally high levels of fats in the blood. These fats include cholesterol and triglycerides. These are important for our bodies to function but when they are high, they can cause heart disease and stroke. Hyperlipidemia is manifested as hypercholesterolemia and/or hypertriglycerolemia. However, hypercholesterolemia is the most common hyperlipidemia. The lipids that are involved in hypercholesterolemia are cholesterol, an essential component of cell membrane and a precursor of steroid hormone synthesis and triglycerides, an important energy source. They are transported in blood as lipoproteins.

METHOD OF SYNTHESIS OF PYRIMIDINE:

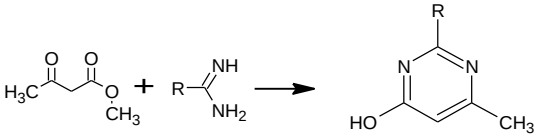
Pinner Pyrimidine Synthesis

The condensation of 1, 3-dicarbonyl compounds with amidinescatalyzed by acids or bases to give pyrimidine derivativesis regarded as the Pinner pyrimidine synthesis.[98, 99]



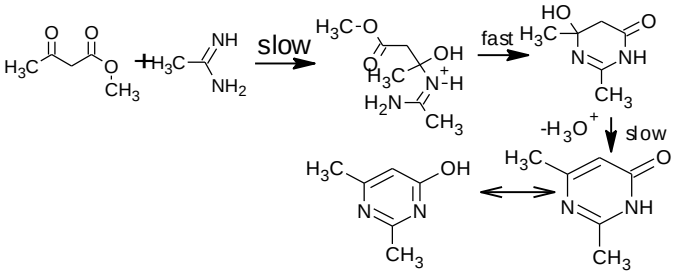
Historical Perspective

In the 1880s, Pinner found that the amidine derivativereacted with acetoacetic esterto furnish 2-substituted-6-hydroxy-4-methylpyrimidine. The condensation of amidinederivativewith other p-keto esters, malonic esters, and P-diketones proceeded similarly. [100-102]



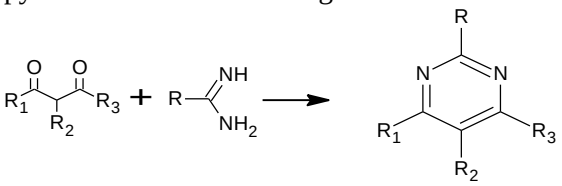
Mechanism

Although the Pinner pynmidine synthesis was discovered a century ago only a few reports on the reaction mechanism have appeared.[103, 104] T he condensation of acetyl acetone, methyl acetoacetate, or dimethyl malonate with acetamidinehas been studied by Katritzky etal. and the reaction mechanisms for these processes have been proposed by these authors.[104] Outlined below is the proposed mechanism of the condensation of methyl acetoacetate with acetamidine.[104]



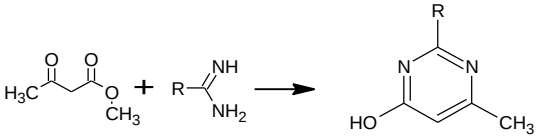
Pinner Pyrimidine Synthesis

The condensation of 1, 3-dicarbonyl compounds with amidinescatalyzed by acids or bases to give pyrimidine derivativesis regarded as the Pinner pyrimidine synthesis.[98, 99]



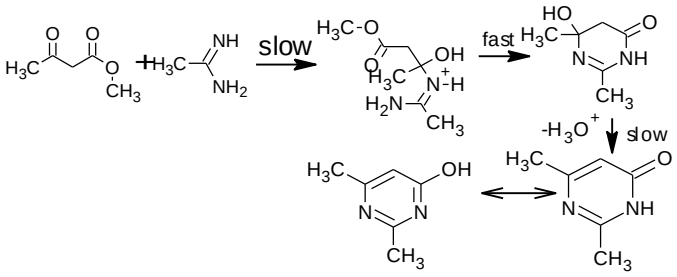
Historical Perspective

In the 1880s, Pinner found that the amidine derivativereacted with acetoacetic esterto furnish 2-substituted-6-hydroxy-4-methylpyrimidine. The condensation of amidinederivativewith other p-keto esters, malonic esters, and P-diketones proceeded similarly. [100-102]



Mechanism

Although the Pinner pynmidine synthesis was discovered a century ago only a few reports on the reaction mechanism have appeared.[103, 104] The condensation of acetyl acetone, methyl acetoacetate, or dimethyl malonate with acetamidinehas been studied by Katritzky etal. and the reaction mechanisms for these processes have been proposed by these authors.[104] Outlined below is the proposed mechanism of the condensation of methyl acetoacetate with acetamidine.[104]



MATERIALS & METHODS:

The identification and characterization of the prepared compounds were carried out by the following procedure to ascertain that all prepared compounds were of different chemical nature than the respective parent compound.

- Physical constants
- Thin Layer Chromatography
- Infrared Spectroscopy (IR)
- Nuclear Magnetic Resonance Spectroscopy (^{1}H-NMR)
- Elemental Analysis (C,H,N)

Procedures for Scheme-I

- Synthesis of 1, 3, 4- DHPM
- Synthesis of acid hydrazide derivative of 1, 3, 4- DHPM
- Synthesis of derivatives from hydrazide

a. Synthesis of mercapto thiadiazole derivative of 1, 3, 4- DHPM
b. Synthesis of pyrazole derivative of 1, 3, 4- DHPM
c. Synthesis of oxadiazole derivative of 1, 3, 4- DHPM

Step 1: Equimolar mixture of ethyl acetoacetate, substituted aromatic aldehyde and urea were taken in beaker. The reaction is carried out by simply heating a mixture of the three components dissolved in ethanol with a catalytic amount of hydrochloric acid at reflux temperature in microwave oven for 4-5 minutes. The product then precipitated on cooling. This reaction is known as "Biginelli reaction". Then the product was recrystallized from ethanol.[146]

Step 2: Dihydropyrimidone so obtained in step 1 was treated with hydrazine hydrate along with catalytic amount of sulfuric acid under microwave irradiation at high voltage for 5-6 minutes. The product was obtained on cooling the reaction mixture followed by neutralization of the mixture by pot. Hydroxide. Then the product was recrystallised from ethanol.[147]

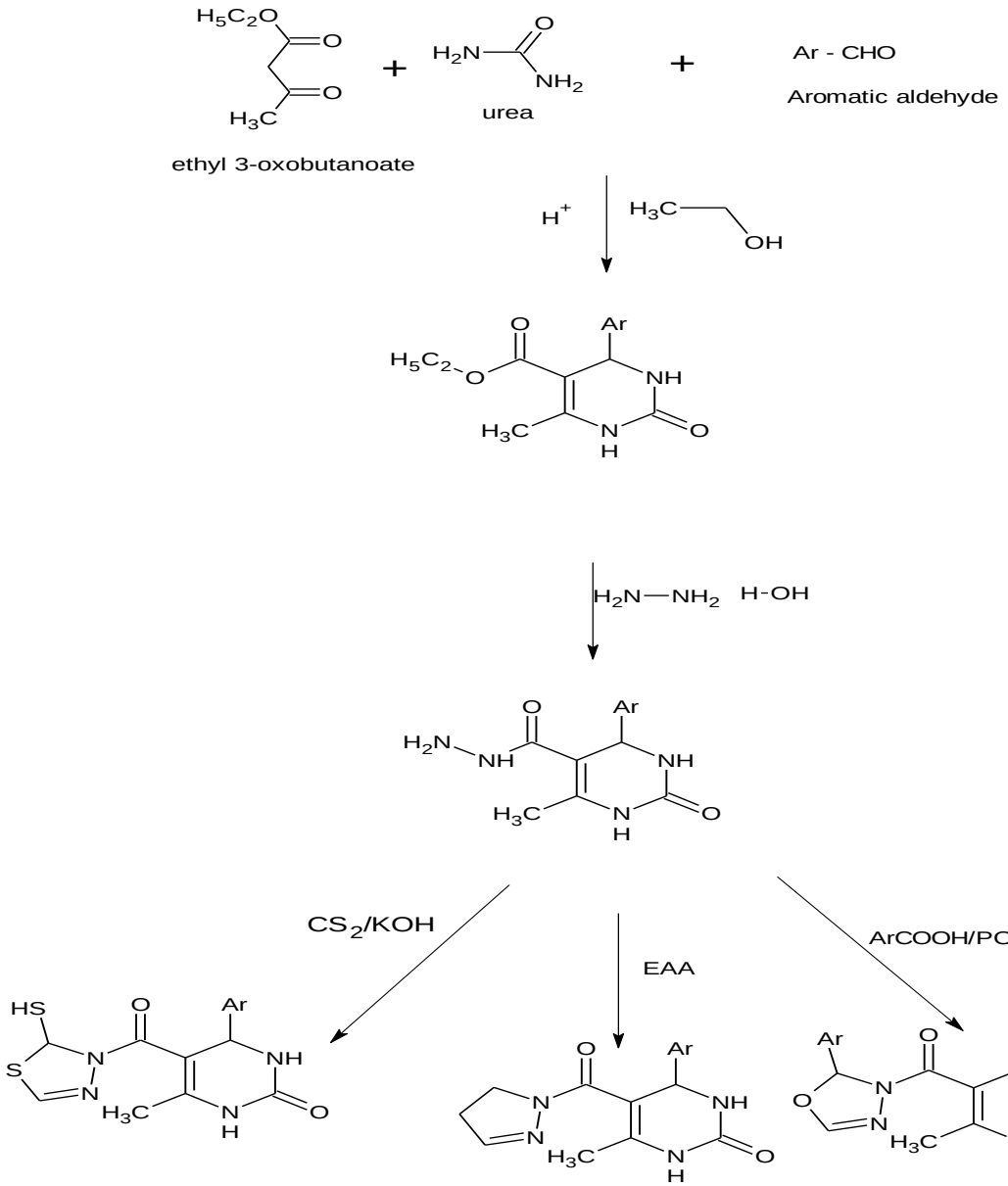
Step 3:

a. Thiadiazole synthesis: the acid hydrazide (0.01 mole) was treated under microwave with carbon disulfide (10 ml) and pot. hydroxide (0.01 mole) in 10 ml of ethanol to give solution which on addition of mineral acid gives precipitate of thiadiazole. (A_1, A_2, A_3.)[149]

b. Pyrazole synthesis: 0.01 mole acid hydrazide was treated under microwave with 0.1 mole of EAA to give pyrazole. (B_1, B_2, B_3.)[150]

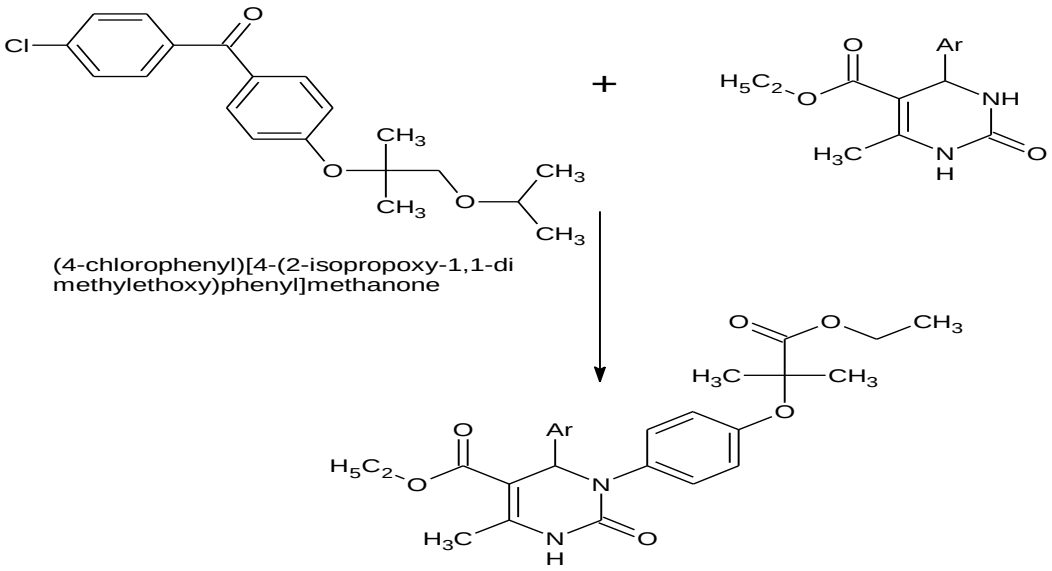
c. Oxadiazole synthesis: 0.01mole acid hydrazide was treated with 0.02 mole of aromatic acid & 10 ml of phosphoryl chloride that was refluxed for 7 hr that was again poured in water 20 ml with stirring to give solid ppt of oxadiazole. (C_1, C_2, C_3.)[148]

Scheme I



Scheme II

Combination of two anti-hypertensive drugs: Procedure: 1, 3, 4- DHPM was dissolved in ethanol and fenofibrate was added to it. The reaction mixture was heated under microwave until the solution. Then removed and cooled to get the precipitate. (D_1, D_2, D_3.)



Scheme III

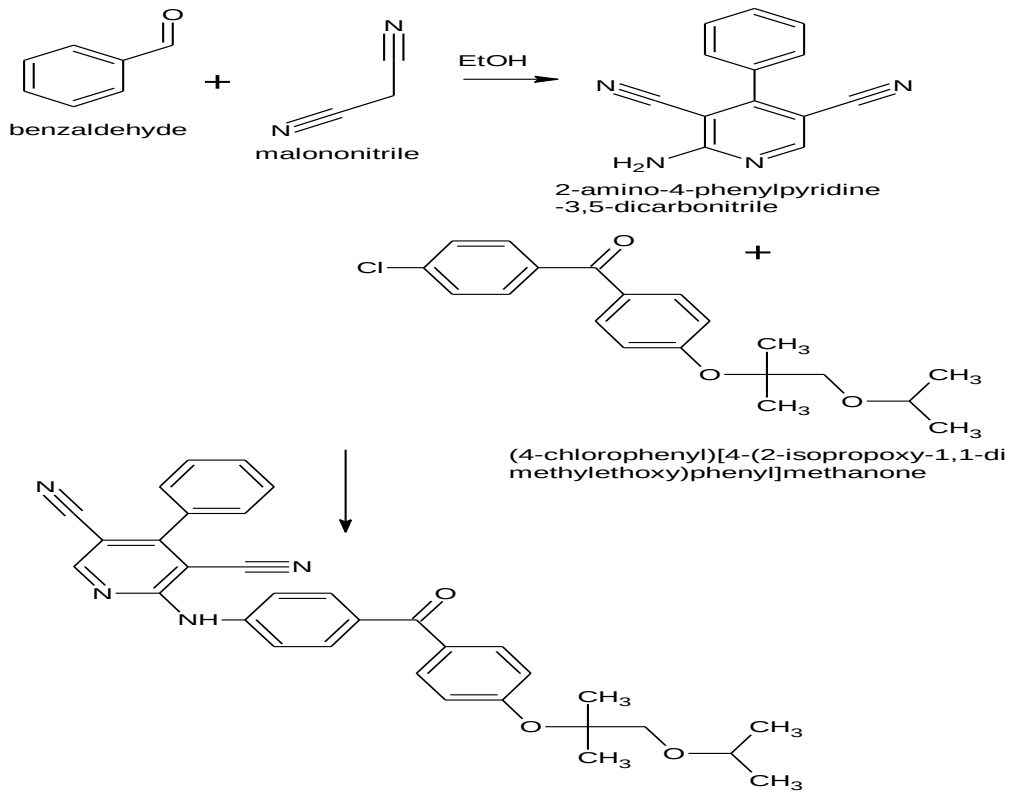
Procedure:

Step 1: Synthesis of 2- amino- 4- phenylpyridine- 3, 5- dicarbonitrile:

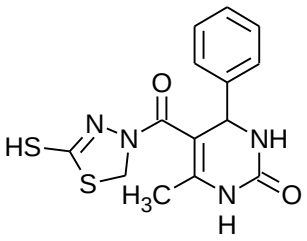
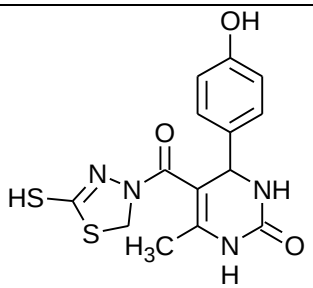
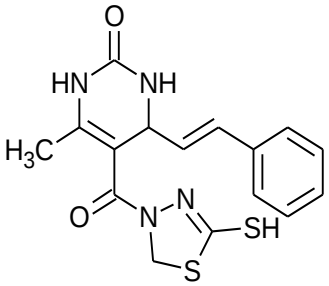
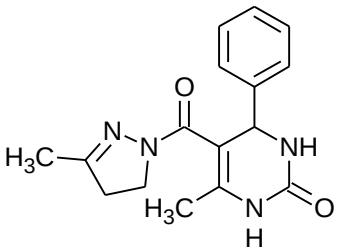
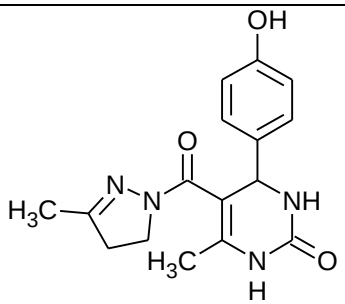
Equimolar mixture of benzaldehyde and malononitrile were treated with ethanol to dissolve and heated under microwave for 2 mins to solubilise completely. On cooling of the reaction mixture we got the precipitate of pyridine derivative.[151]

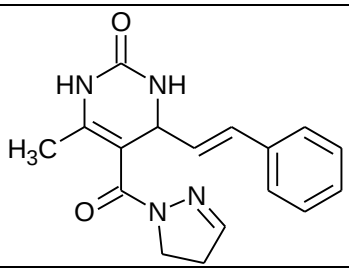
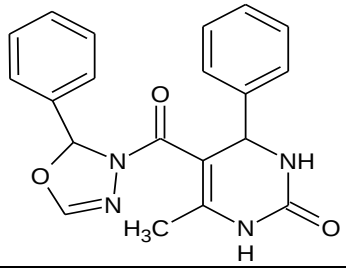
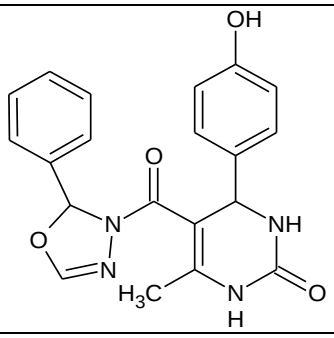
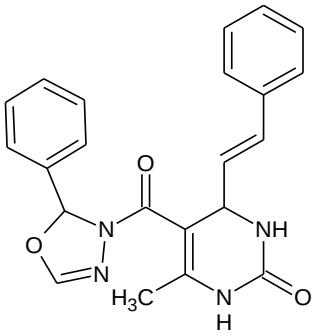
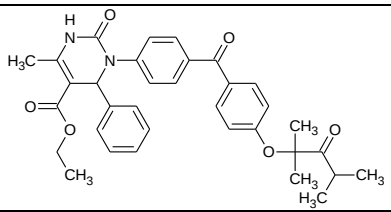
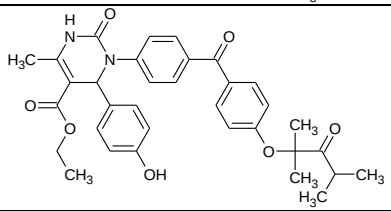
Step 2: combining the pyridine derivative and fenofibrate:

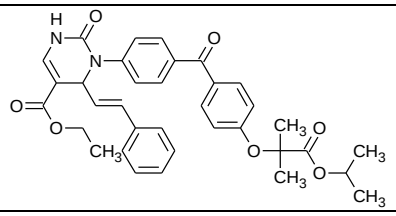
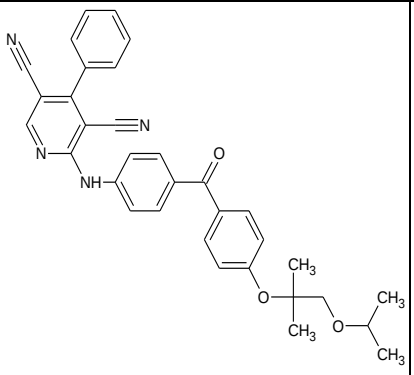
Equimolar mixture of step 1 product and fenofibrate were dissolved in ethanol with 2 drops of 10% NaOH solution under microwave for 30 sec. on cooling we get the product.



IUPAC names of the synthesized compounds

Compound code.	Structure	Name	Empirical Formula
A_1		5-[(5-mercapto-1,3,4-thiadiazol-3(2*H*)-yl)carbonyl]-6-methyl-4-phenyl-3,4-dihydropyrimidin-2(1*H*)-one	$C_{12}H_{14}O_2N_4S_2$
A_2		4-(4-hydroxyphenyl)-5-[(5-mercapto-1,3,4-thiadiazol-3(2*H*)-yl)carbonyl]-6-methyl-3,4-dihydropyrimidin-2(1*H*)-one	$C_{13}H_{12}O_3N_4S_2$
A_3		5-[(5-mercapto-1,3,4-thiadiazol-3(2*H*)-yl)carbonyl]-6-methyl-4-[(*E*)-2-phenylvinyl]-3,4-dihydropyrimidin-2(1*H*)-one	$C_{16}H_{15}O_2N_4S_2$
B_1		6-methyl-5-[(3-methyl-4,5-dihydro-1*H*-pyrazol-1-yl)carbonyl]-4-phenyl-3,4-dihydropyrimidin-2(1*H*)-one	$C_{15}H_{15}O_2N_4$
B_2		4-(4-hydroxyphenyl)-6-methyl-5-[(3-methyl-4,5-dihydro-1*H*-pyrazol-1-yl)carbonyl]-3,4-dihydropyrimidin-2(1*H*)-one	$C_{15}H_{15}O_3N_4$

B_3		5-(4,5-dihydro-1*H*-pyrazol-1-ylcarbonyl)-6-methyl-4-[(*E*)-2-phenylvinyl]-3,4-dihydropyrimidin-2(1*H*)-one	$C_{16}H_{17}O_2N_4$
C_1		6-methyl-4-phenyl-5-[(2-phenyl-1,3,4-oxadiazol-3(2*H*)-yl)carbonyl]-3,4-dihydropyrimidin-2(1*H*)-one	$C_{20}H_{17}O_3N_4$
C_2		4-(4-hydroxyphenyl)-6-methyl-5-[(2-phenyl-1,3,4-oxadiazol-3(2*H*)-yl)carbonyl]-3,4-dihydropyrimidin-2(1*H*)-one	$C_{20}H_{17}O_4N_4$
C_3		6-methyl-5-[(2-phenyl-1,3,4-oxadiazol-3(2*H*)-yl)carbonyl]-4-[(*E*)-2-phenylvinyl]-3,4-dihydropyrimidin-2(1*H*)-one	$C_{22}H_{19}O_3N_4$
D_1		(E)-6-methyl-5-(2-phenyl-2, 3-dihydro-1,3,4-oxadiazole-3-carbonyl)-4-styryl-3,4-dihydropyridine-2(1H)-one	$C_{34}H_{36}O_7N_2$
D_2		Ethyl-3-(4-(4-((2,4-dimethyl-3-oxopentan-2-yl)oxy)benzoyl)phenyl)-4-(4-hydroxyphenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyridine-5-carboxylate	$C_{34}H_{36}O_8N_2$

D_3		(E)-ethyl-3-(4-(4((1-isopropoxy-2-methyl-1-oxopropan-2-yl)oxy)benzoyl)phenyl)-2-oxo-4-styryl-1,2,3,4,tetrahydropyridine-5-carboxylate	$C_{36}H_{38}O_7N_2$
F_2		Isopropyl-2-(4-(4-((3,5-dicyano-4-phenylpyridin-2-yl)amino)benzoyl)phenoxy)-2-methylpropanoate	$C_{33}H_{28}O_4N_4$

Physico- chemical constants of the synthesized compounds

Compound	Mol. Wt.	M. P.		% yield		Elemental analysis (calctd.)			Rf value	
		M. W.	S. F.	M. W.	S. F.	C	H	N	M. W.	S. F.
A_1	335.6705	262-264	260-262	96.9	99.7	50.09	4.50	16.76	0.6	0.59
A_2	336.605	264-266	264-266	96.96	98.9	49.95	3.59	16.71	0.58	0.59
A_3	359.6745	272-274	270-272	94.70	99.7	53.43	4.20	15.64	0.61	0.60
B_1	283.5435	278-280	276-278	80.13	95.33	63.53	5.33	19.84	0.58	0.56
B_2	299.5335	284-286	284-286	91.21	96.8	60.15	5.04	18.78	0.60	0.62
B_3	297.5703	296-298	294-296	90.90	96.66	64.58	5.75	18.91	0.63	0.62
C_1	361.6043	290-292	292-294	90.16	96.9	66.43	4.73	15.56	0.68	0.66
C_2	377.5943	294-296	292-294	90.9	97.65	63.61	4.53	14.90	0.66	0.67
C_3	387.6421	298-300	296-298	95.6	98.7	68.16	4.94	14.51	0.63	0.65

compound	Mol. Wt.	M. P.	% yield	Elemental analysis (calctd.)			Rf value
				C	H	N	
D_1	584.7284	302-304	85.10	69.83	6.20	4.81	0.66
D_2	600.7184	308-310	60.81	67.98	6.04	4.68	0.65
D_3	610.7662	314-316	92.10	70.79	6.27	4.60	0.66
F_2	545.8321	292-294	96.29	72.61	5.17	10.31	0.53

Infra-Red / 1H NMR spectral study of the synthesized compounds scheme

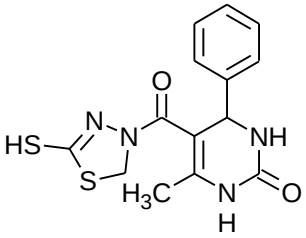
Compd. Code	IR Bands (cm^{-1})	Types of Vibrations	δ Values in ppm	No. Of Protons
A_1	3639.98 3050.83 3360.44 3338.27 1714.41 1605 1197.05 1144.02 1133.94	S-H str CH=CHstr NH-str CH_2 str C=O str C=C Ar C-O-C str C-S-C-str C-N-str.	8.892 7.283 7.4 & 7.5 3.9 & 4.0	1H of DHPM 5H of Ar CH 2H of thiadiazole 2H of 2NH
A_2	3650 3535 3355.62 3338.27 3325.73 1705.73 1678.73 1235.18 1200.47 1138.76	O-H S-H Ar-C CH_2 str NH-str C=O str C=C Ar C-O-C str C-S-C-str C-N-str		
A_3	3239.82 3222.47 3102.9 3023.84 2936.09 1705.73 1652.7 1108 1090 1089	Ar-CH str. CH_2 str. C-H Alkene NH-str. C=O str. C=C Ar C-O-C str C-S-C-str. C-N-str.		
B_1	3270.04 3029.9 2990.45 2392.54 2300.44 1725.26 1600.05 1108 1080	CH_2 str C-H Alkene C-H Ar NH-str C=O str C=C Ar C-O-C str C-N-str	8.994 7.14 – 7.93 1.068 – 1.26 2.238 & 2.494 3.925 – 3.99	1H of DHPM 5H of Ar-CH 6H of CH_3 2H of 2NH 4H of $2CH_2$

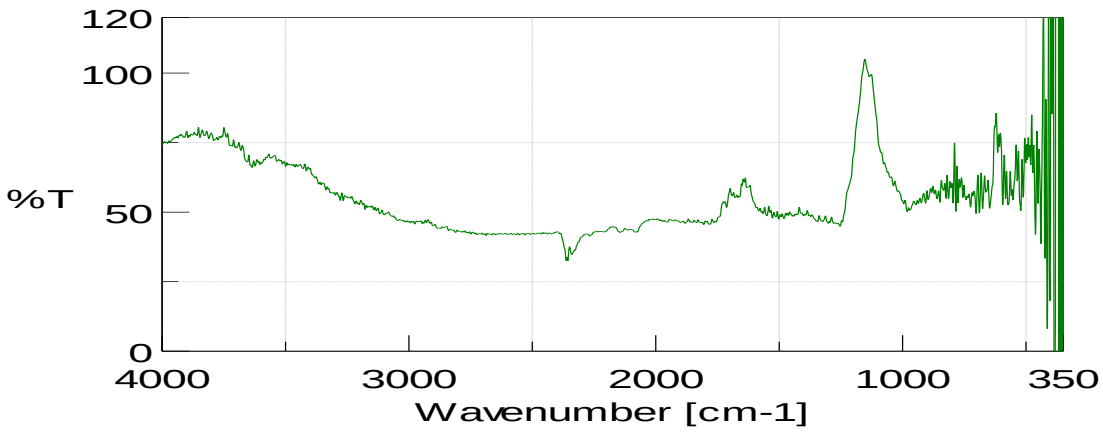
B_2	3511.74 3288.04 3129.9 2980.45 2492.54 2360.44 1718.26 1670.05 1180 1095	O-H str CH_2 str C-H Alkene C-H Ar NH-str C=O str C=C Ar C-O-C str C-N-str		
B_3	3248.5 3112.55 2971.77 2936.09 2355.62 2329.59 1731.76 1114.41 1157.52	Ar-CH str CH_2 str C-H Alkene C-H Ar NH-str C=O str C-O-C str C-N-str		
C_1	3194.23 2959.51 2915.16 2317.33 2255.62 2243.09 1722.62 1682.23 1009.23	C-H Ar C-H Alkene CH_2 N-H C=O C=C Ar C-O-C	8.997 7.3 – 7.9 1.096 – 1.11 7.336 & 7.357 5.24 & 5.25	1H of DHPM 10H of Ar 3H of CH_3 2H of 2CH of pyrazole 2H of 2NH
C_2	3062.23 3045.51 3001.16 2410.33 2305.62 2344.09 1755.62 1688.23 1090.23 1060.37	C-H Ar C-H Alkene CH_2 N-H C=O C=C Ar C-O-C		
C_3	3094.23 3059.51 3015.16 2417.33 2355.62 2343.09 1762.62 1692.23 1099.23	C-H Ar C-H Alkene CH_2 N-H C=O C=C Ar C-O-C		

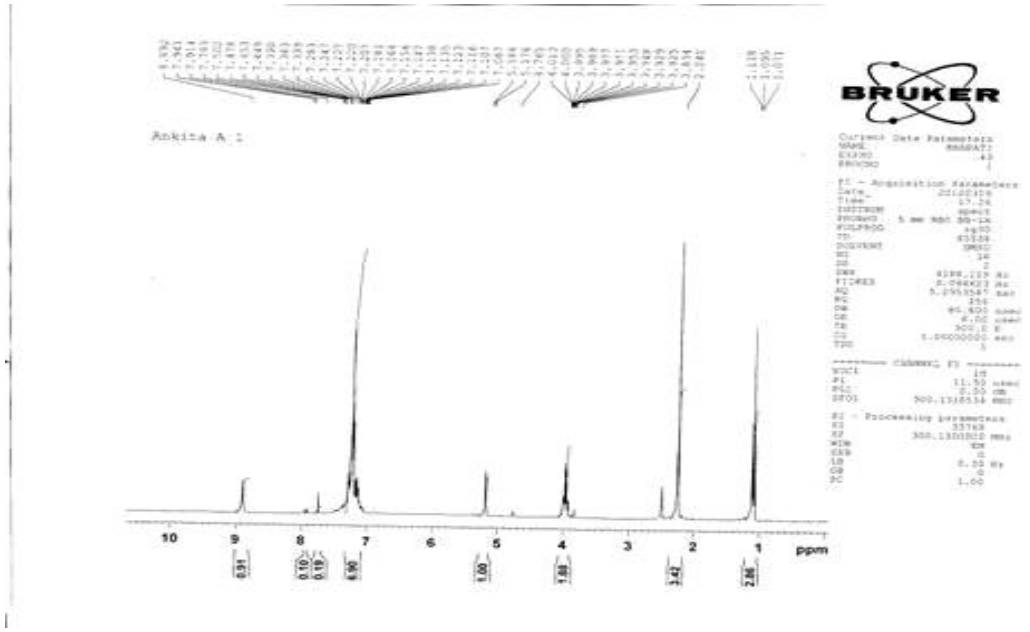
D_1	3248.5 3107.72 2980.45 2360.44 2343.09 1727.91 1714.41 1700.91 1648.84 1173.47 1094.4	C-H Alkene CH_2 N-H C=O C=C Ar C-O-C C-N	1.583 2.246 & 2.487 7.1 – 7.6 8.841 5.2	1H of CH 2H of CH_2 13H of Ar 1H of DHPM 1H of NH
D_2	3550 3248.5 3107.72 2980.45 2360.44 2343.09 1727.91 1714.41 1700.91 1648.84 1173.47 1094.4	O-H C-H Alkene CH_2 N-H C=O C=C Ar C-O-C		
D_3	3620 3228.5 3207.72 2880.45 2460.44 2443.09 1737.91 1724.41 1710.91 1658.84 1163.47 1084.4	O-H C-H Alkene CH_2 N-H C=O C=C Ar C-O-C		
F_2	3283.21 3076.87 2989.12 2932.23 2360.44 2286.2 1731.76 1678.73 1648.84 1010.52	C-H Ar CH_2 N-H Nitrile C=O C=C Ar C-O-C	6.76 – 7.64 7.654 1.14 2.487 2.499 2.941	13H of Ar 1H of DHP CH_3 CH_2 CH NH
A_{11}	3652.36 3235 3112.55 2980.45 2360.44 2343.09	S-H CH_2 C-H Ar N-H	8.950 5.217 1.068 – 1.115 7.156 – 7.249 3.935 & 3.954	1H of DHPM SH 3H of CH_3 5H of Ar 2H of NH

	2325.73 1736.58 1700.91 1648.84 1100 1090 1080	C=O C=C Ar C-S-C C-N C-O-C		
B_{11}	3248.5 3107.72 2980.45 2369.12 2355.62 2338.27 2320.91 1723.09 1700.91 1652.7 1080	C-H Ar CH_2 N-H C=O C=C Ar C-N	1.099 2.259 3.968 & 3.986 7.231 8.979	CH_3 CH_2 NH Ar 1H of DHPM
C_{11}	3248.5 3121.22 2984.3 2554.25 2355.62 2338.27 1723.09 1705.73 1182.15 1169.62 1099.23	C-H CH_2 N-H C=O C-O-C C-N	1.08 2.256 3.945 – 3.975 7.076 – 7.535 8.873	CH_3 CH_2 NH Ar 1H of DHPM

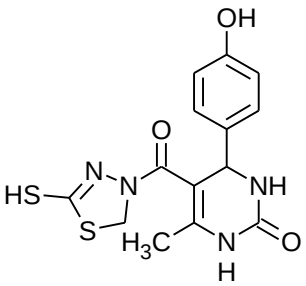
A1

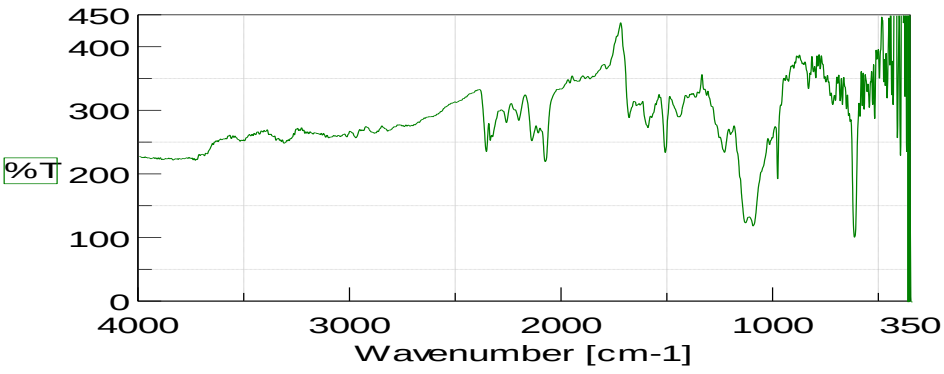




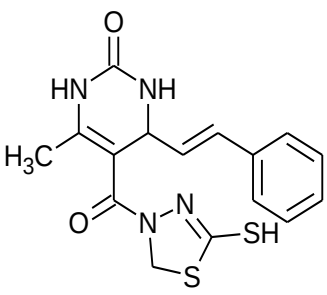

BRUKER
ppm

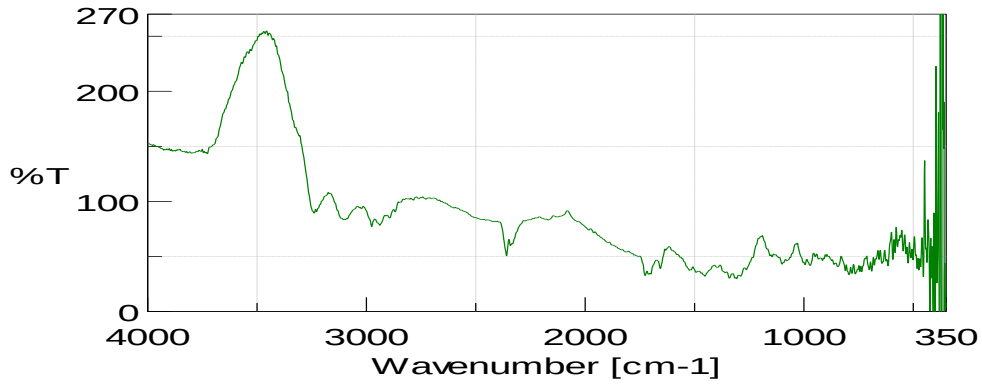
A2


OH
O
N
N
HS
S
NH
H3C
N
H
O

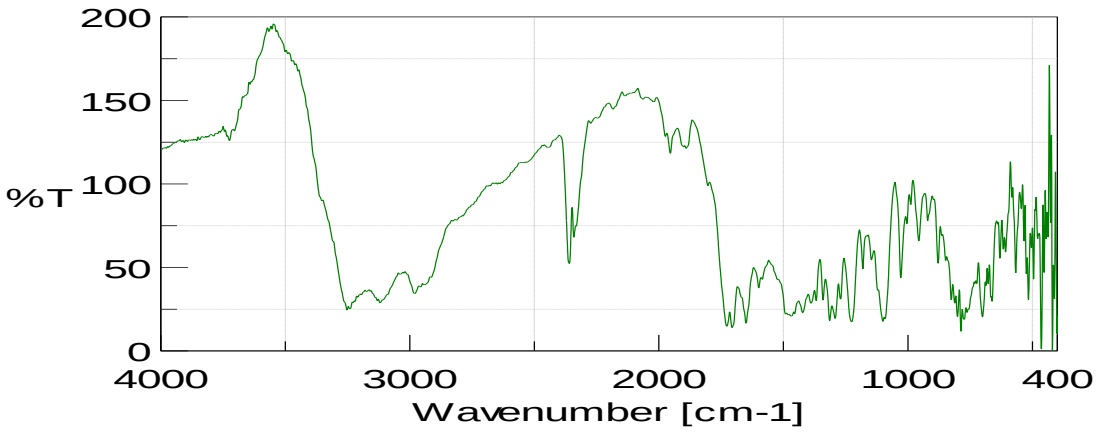

450
400
300
%T
200
100
0
4000
3000
2000
1000
350
Wavenumber [cm-1]

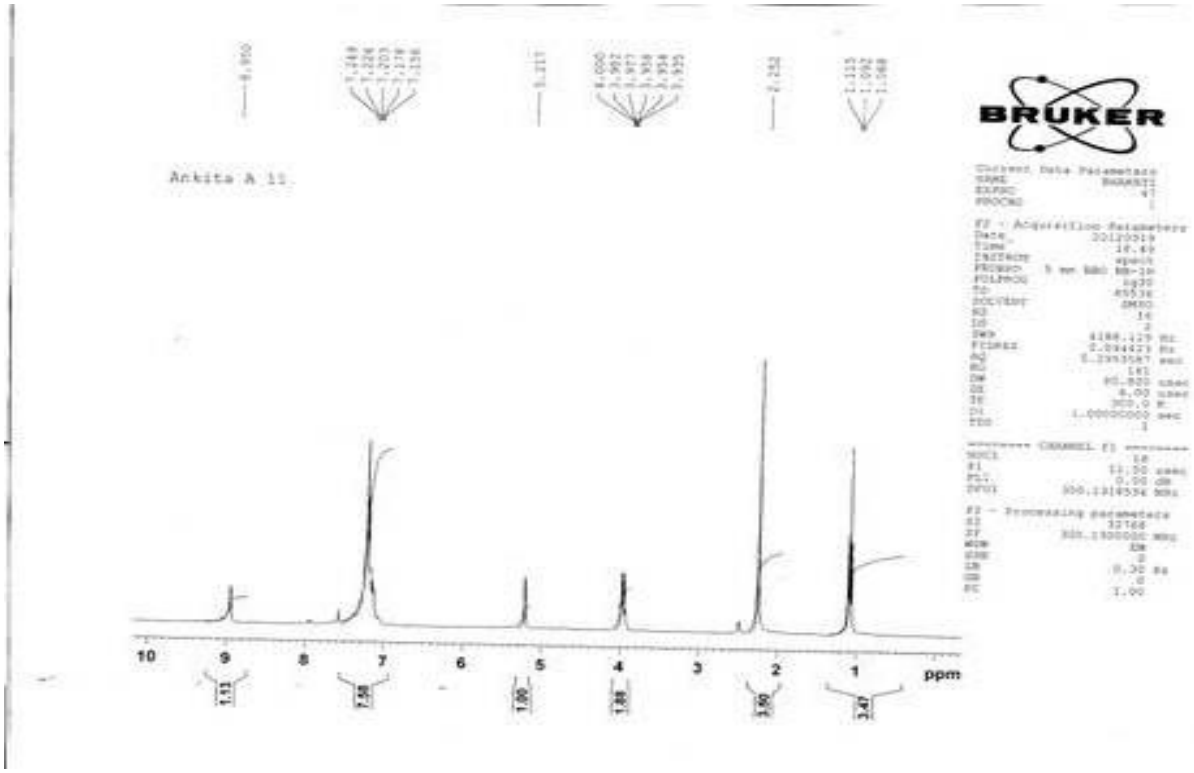
A3


O
HN
NH
H3C
O
N
N
SH
S

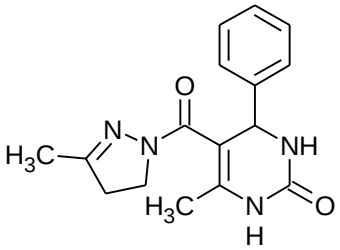

270
200
%T
100
0
4000
3000
2000
1000
350
Wavenumber [cm-1]

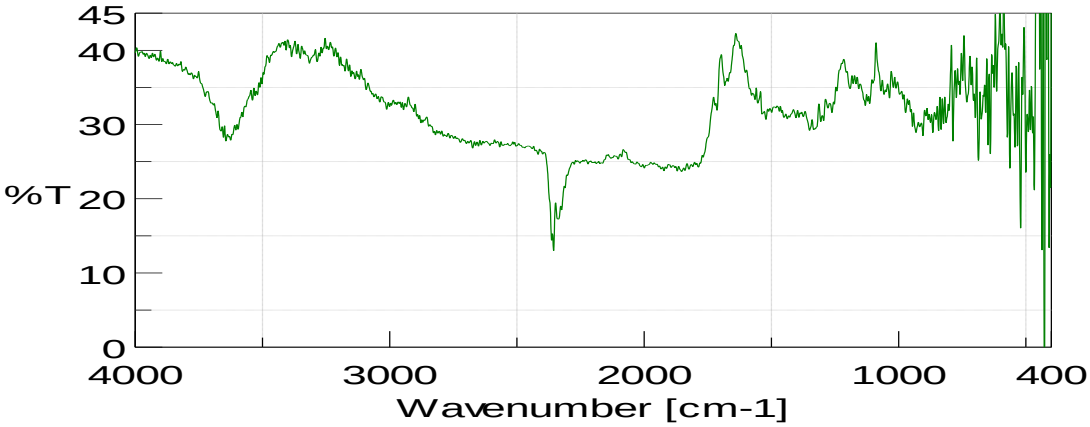
A11

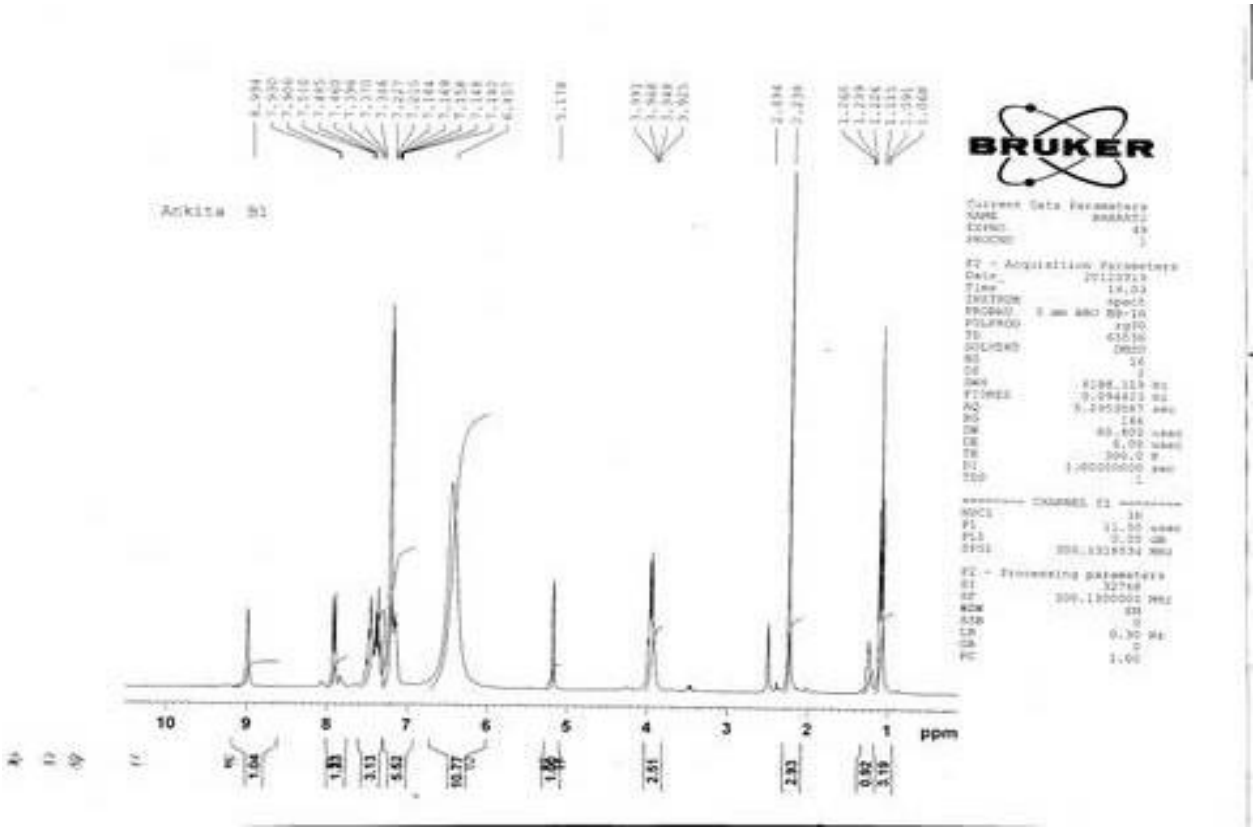

200
150
%T
100
50
0
4000
3000
2000
1000
400
Wavenumber [cm-1]


Ankita A 11
BRUKER
10
9
8
7
6
5
4
3
2
1
ppm

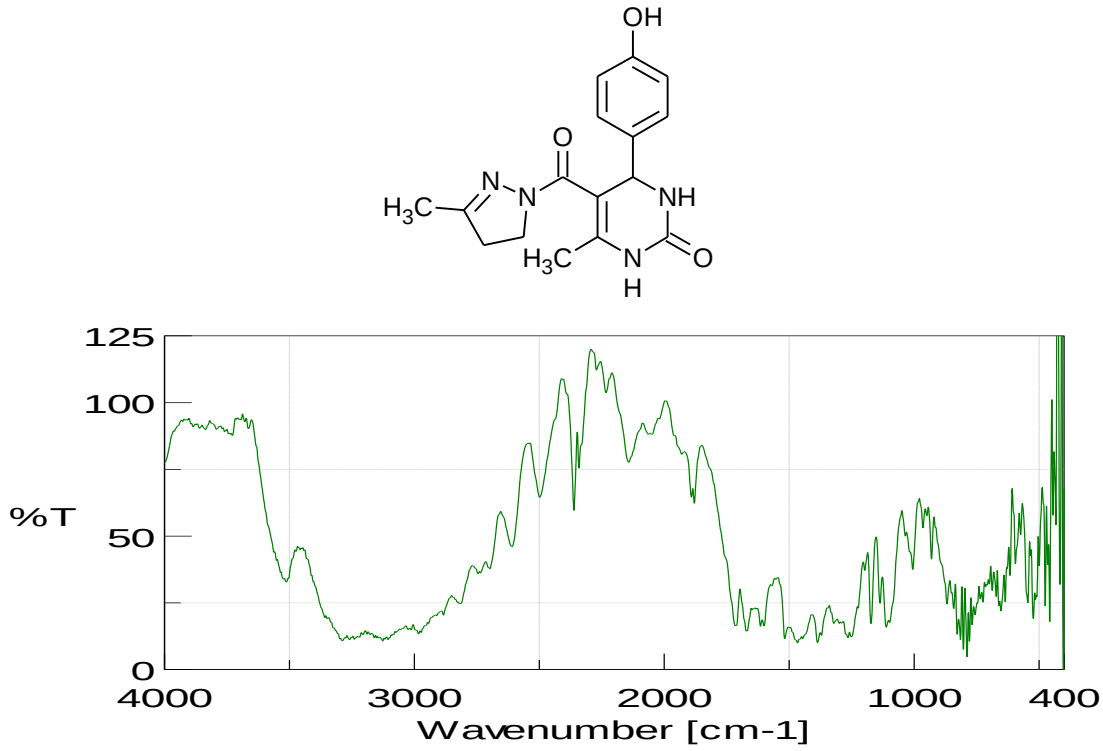
B1


O
N
N
NH
H3C
H3C
N
O
H

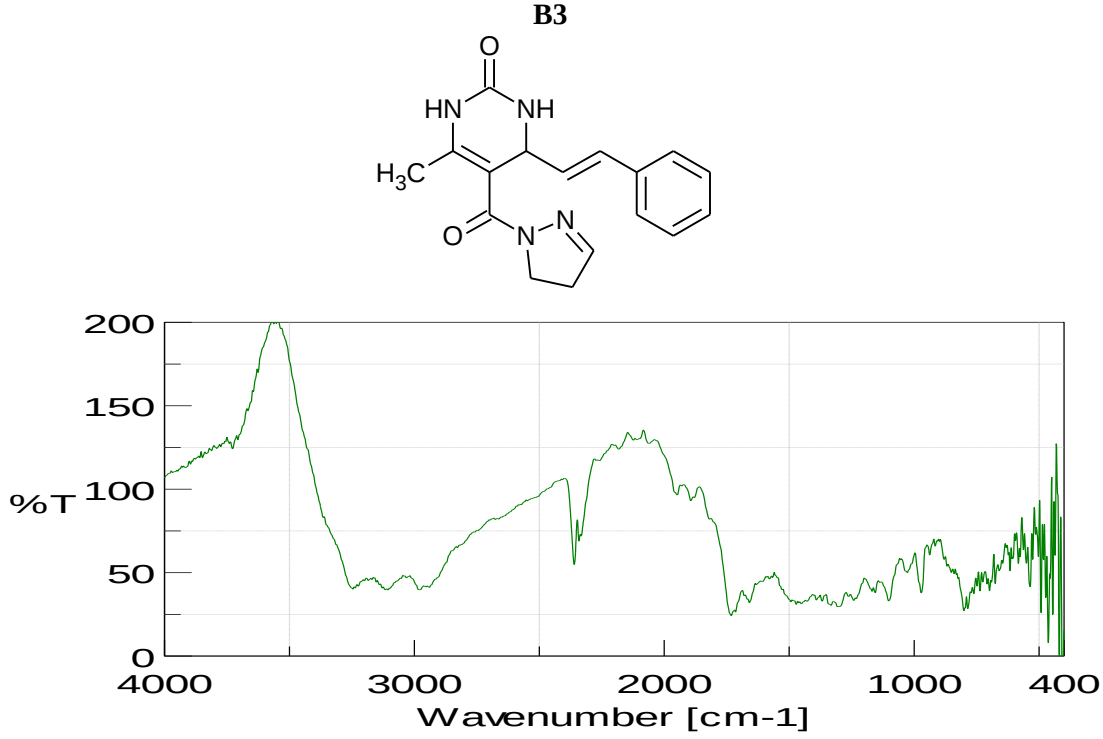

45
40
30
%T 20
10
0
4000
3000
2000
1000
400
Wavenumber [cm-1]


BRUKER
Ankita B1
10
9
8
7
6
5
4
3
2
1
ppm

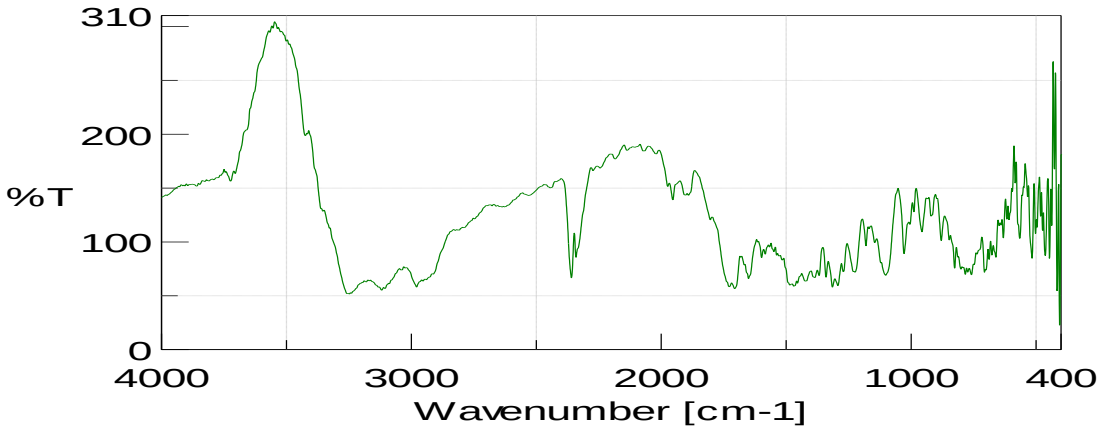
B2

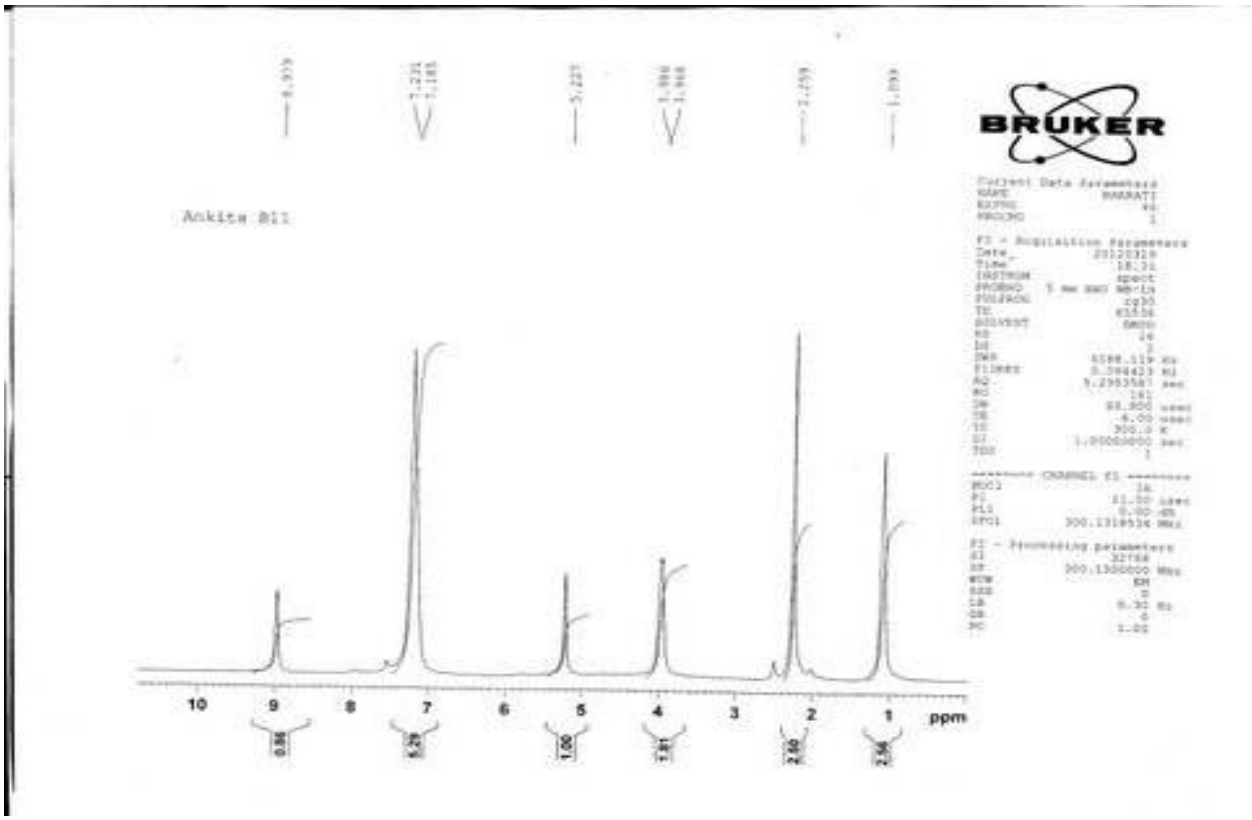

OH
O
N
N
H3C
NH
H3C
N
H
O
125
100
%T
50
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

B3

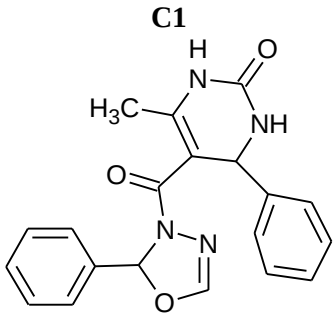

O
HN
NH
H3C
O
N
N
200
150
%T
100
50
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

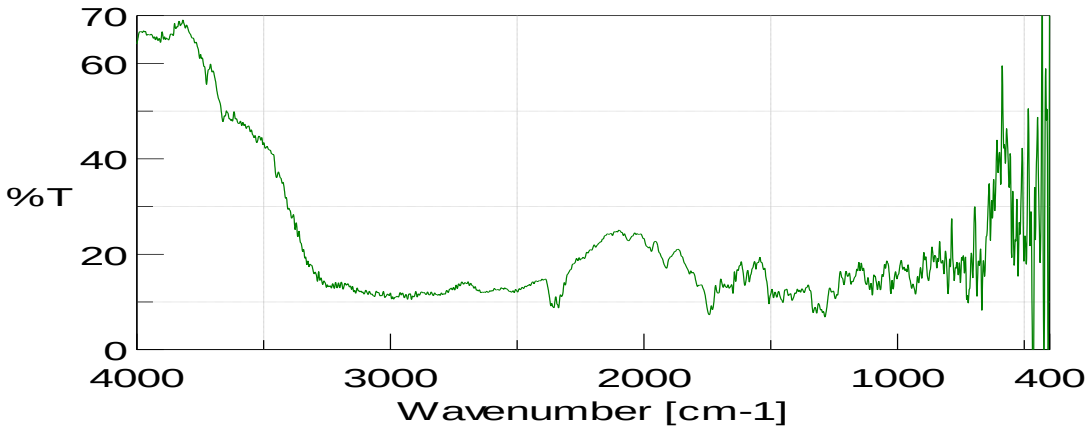
B11

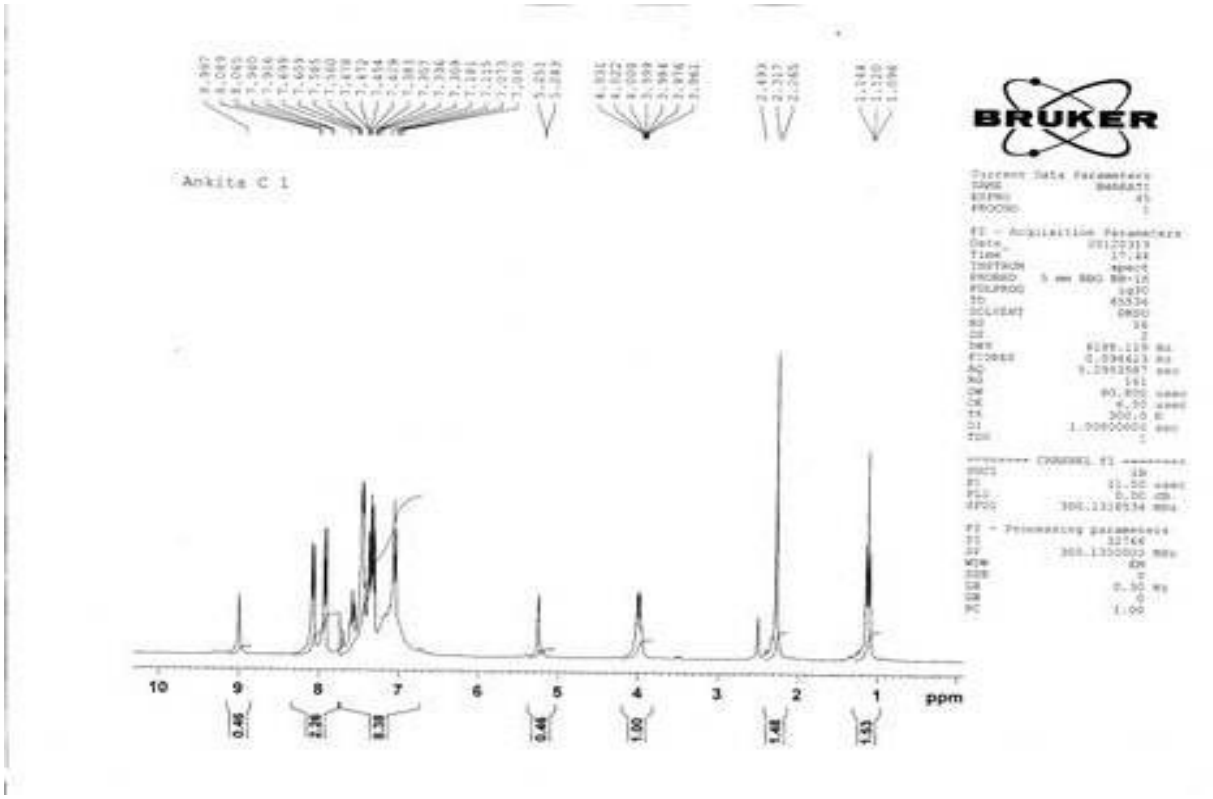

310
200
%T
100
0
4000
3000
2000
1000
400
Wavenumber [cm-1]


BRUKER
Ankita Bit
10
9
8
7
6
5
4
3
2
1
ppm

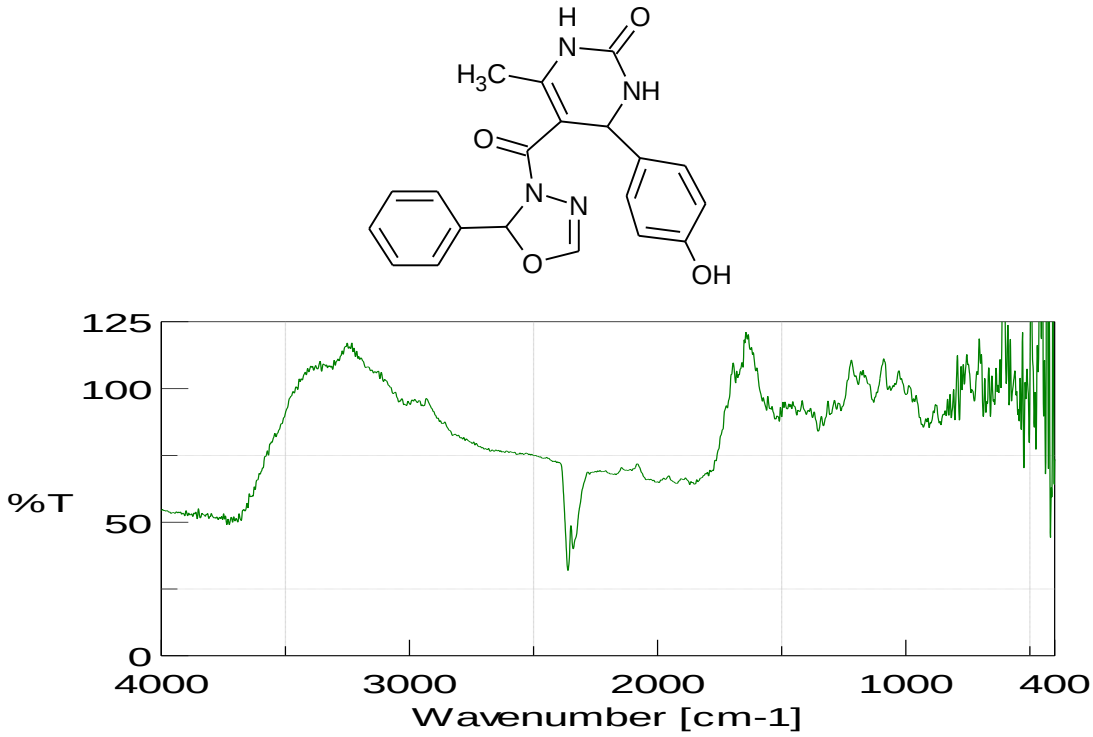
C1


H
O
N
H3C
NH
O
N
N
O

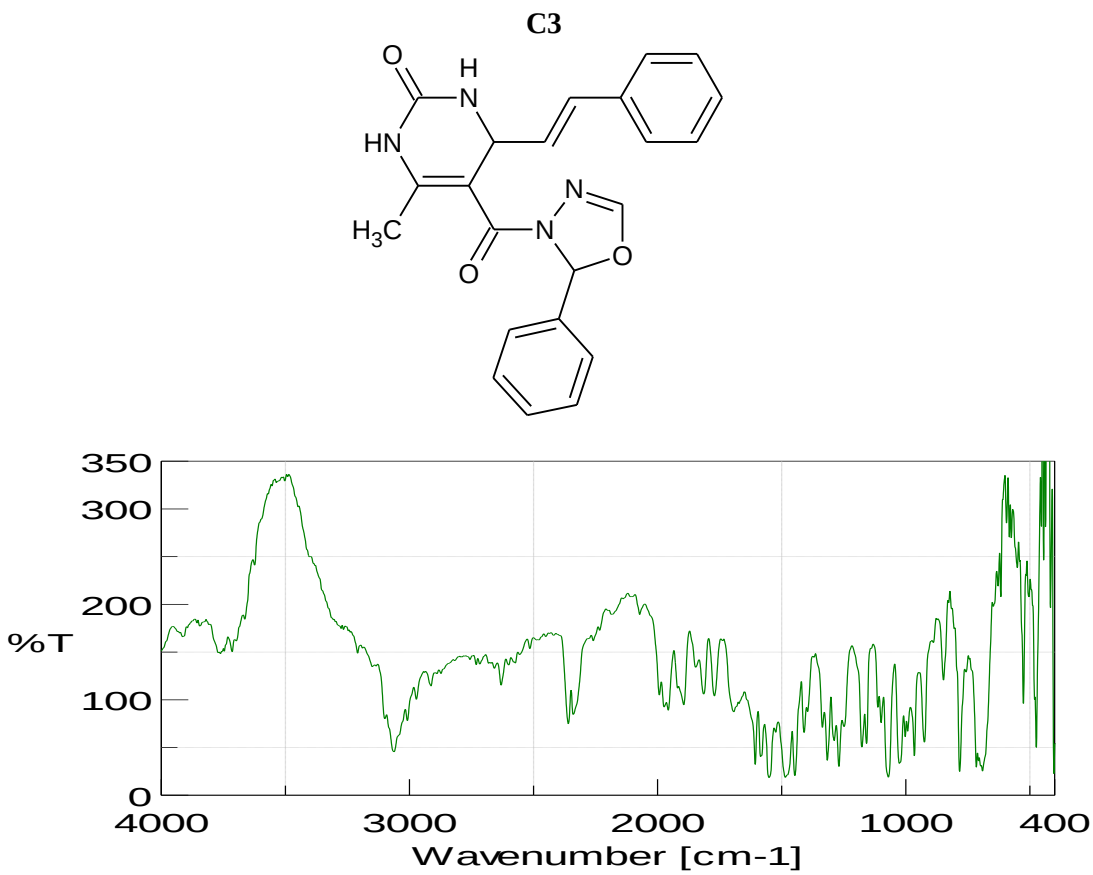

70
60
40
20
0
%T
4000
3000
2000
1000
400
Wavenumber [cm-1]


BRUKER
Ankita C 1
10
9
8
7
6
5
4
3
2
1
ppm

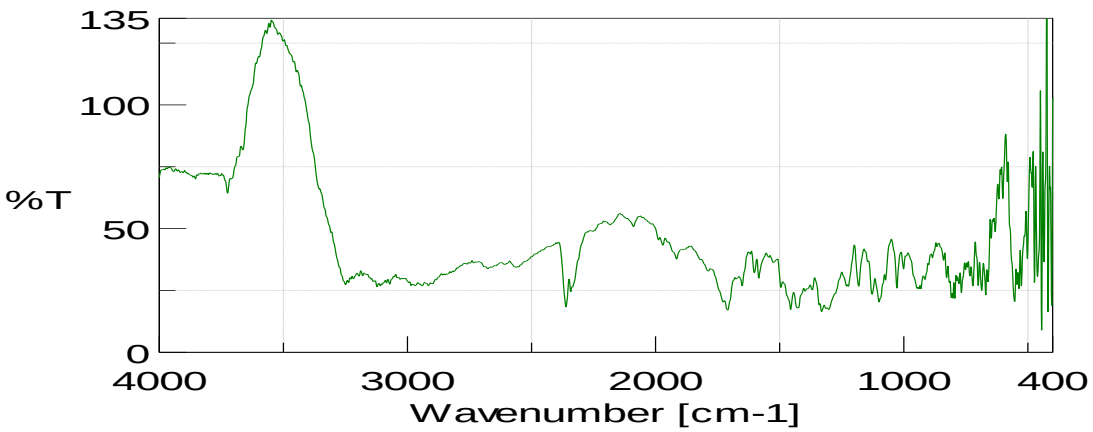
C2

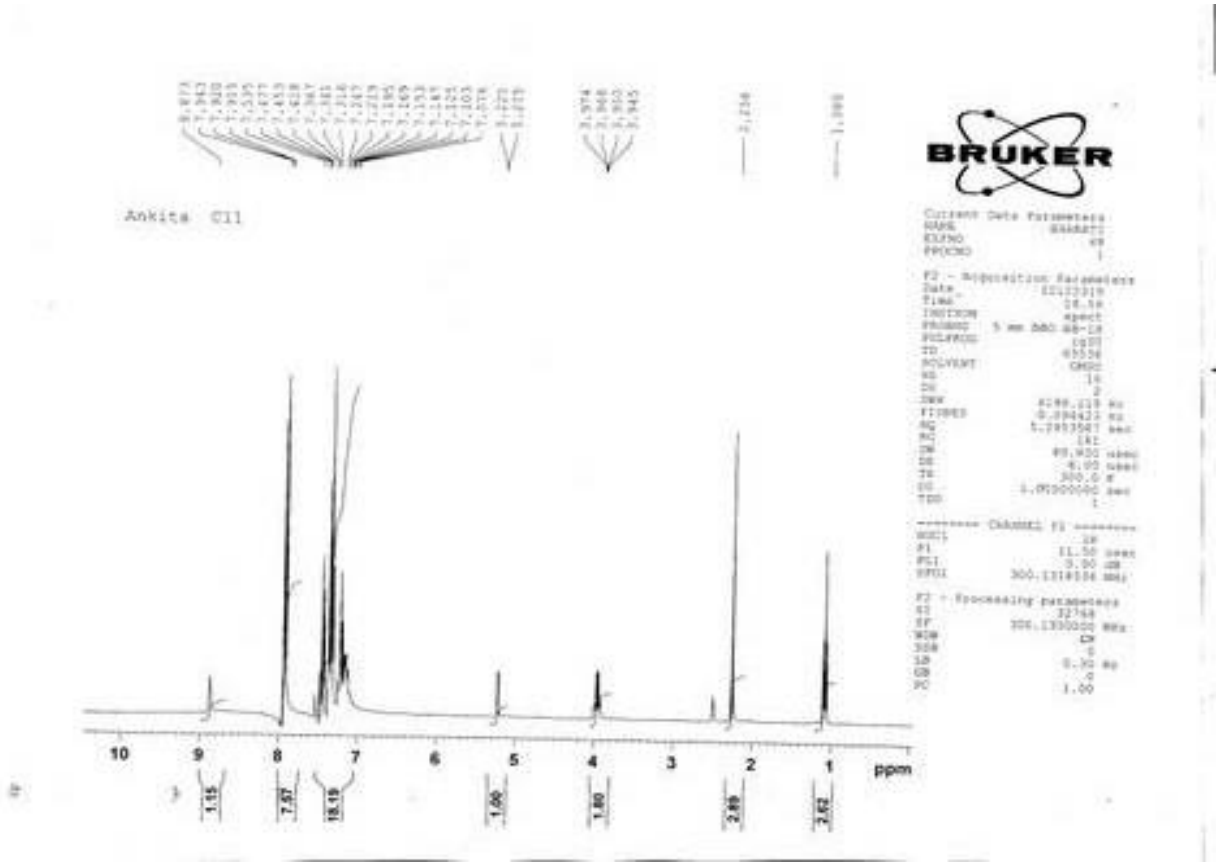

H
O
N
H3C
NH
O
N
N
O
OH
125
100
%T
50
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

C3

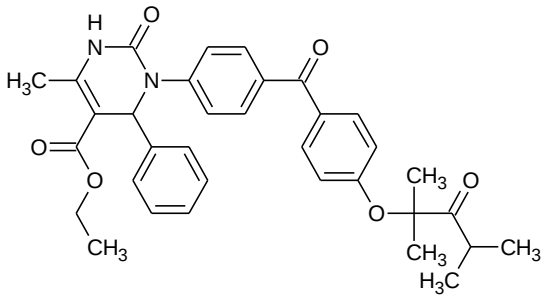

O
H
N
HN
H3C
N
N
O
O
350
300
200
%T
100
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

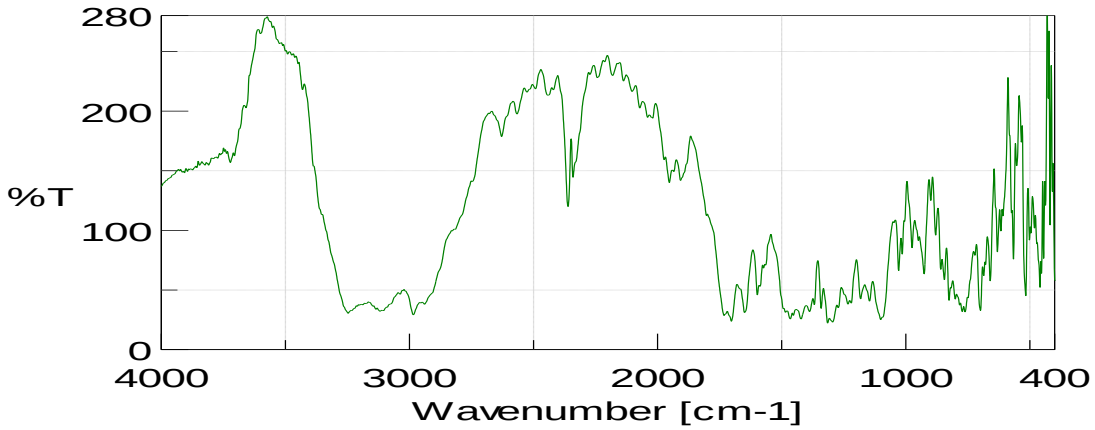
C11

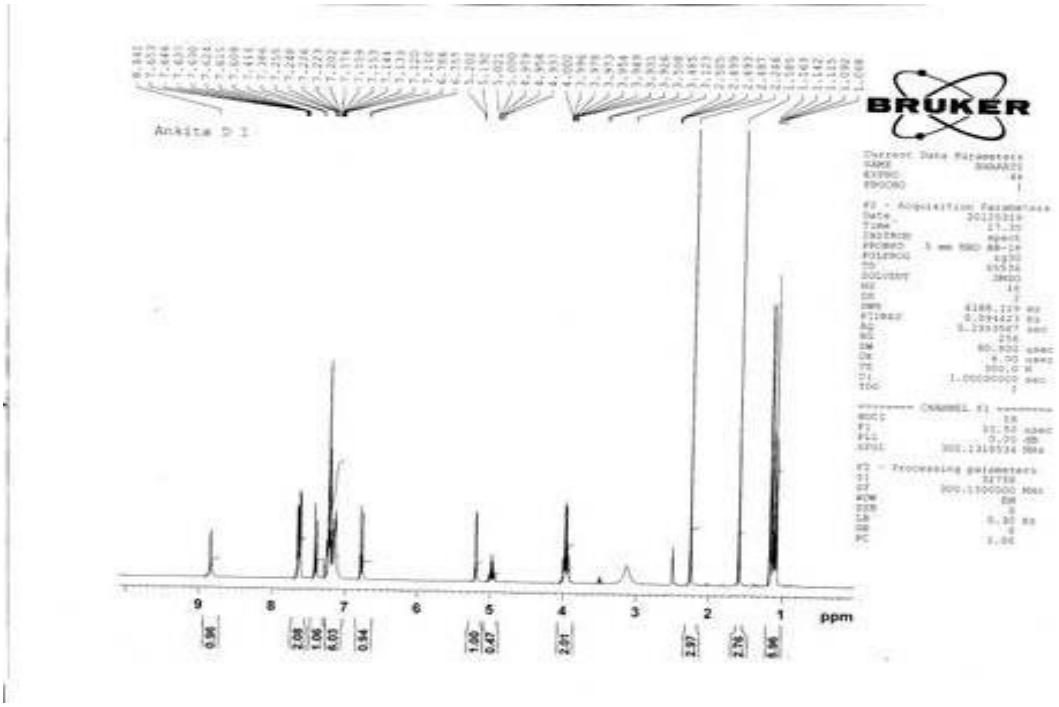

135
100
%T
50
0
4000
3000
2000
1000
400
Wavenumber [cm-1]


BRUKER
Ankita C11
10
9
8
7
6
5
4
3
2
1
ppm

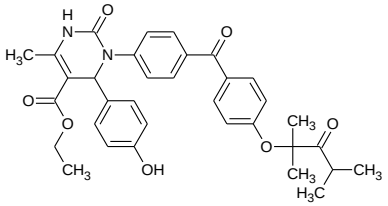
D1

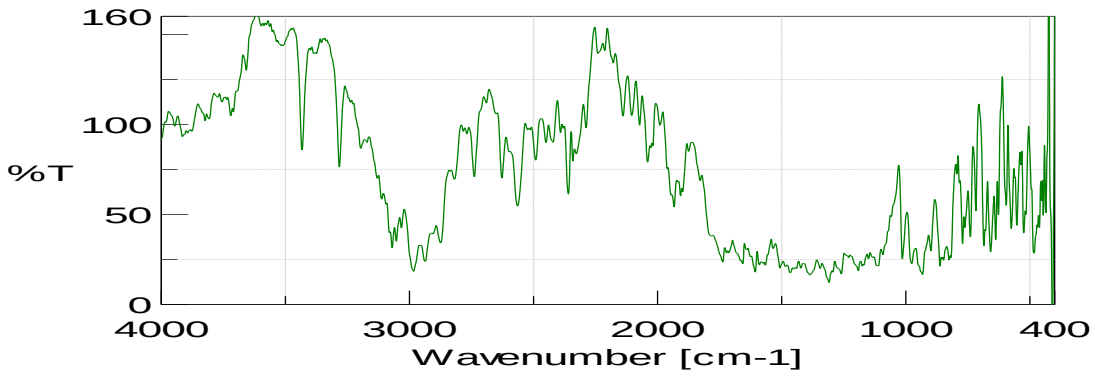

H
N
O
H3C
N
O
O
O
CH3
O
CH3
O
CH3
CH3
H3C
CH3


280
200
%T
100
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

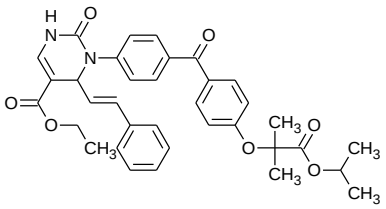

BRUKER
ppm

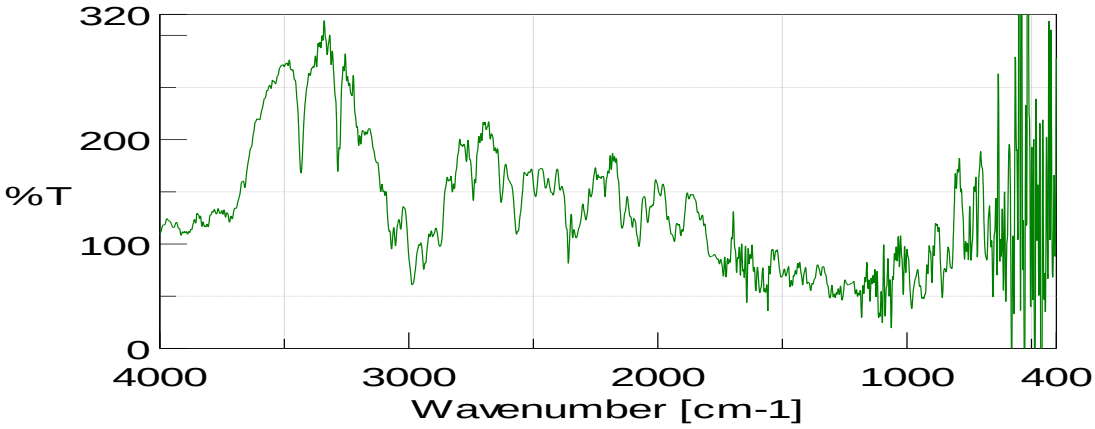
D2



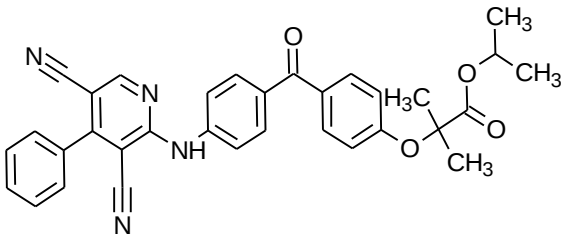

160
100
%T
50
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

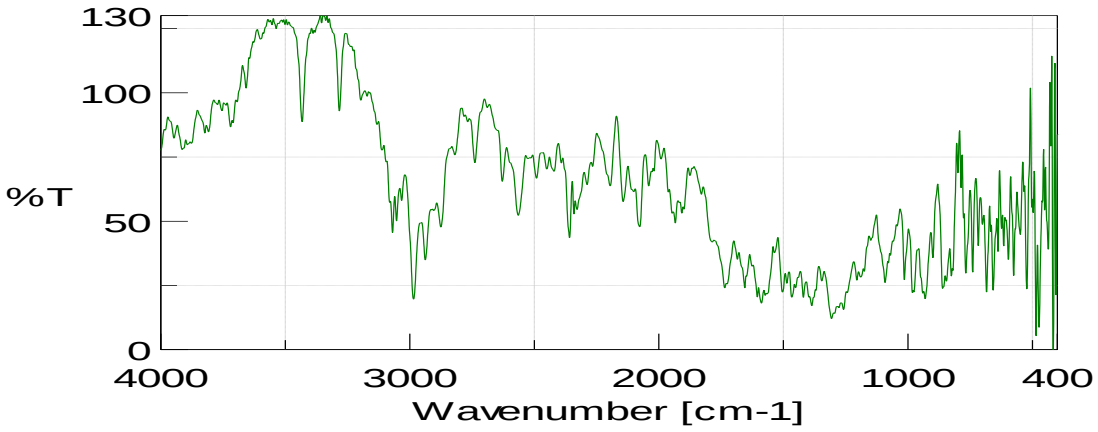
D3

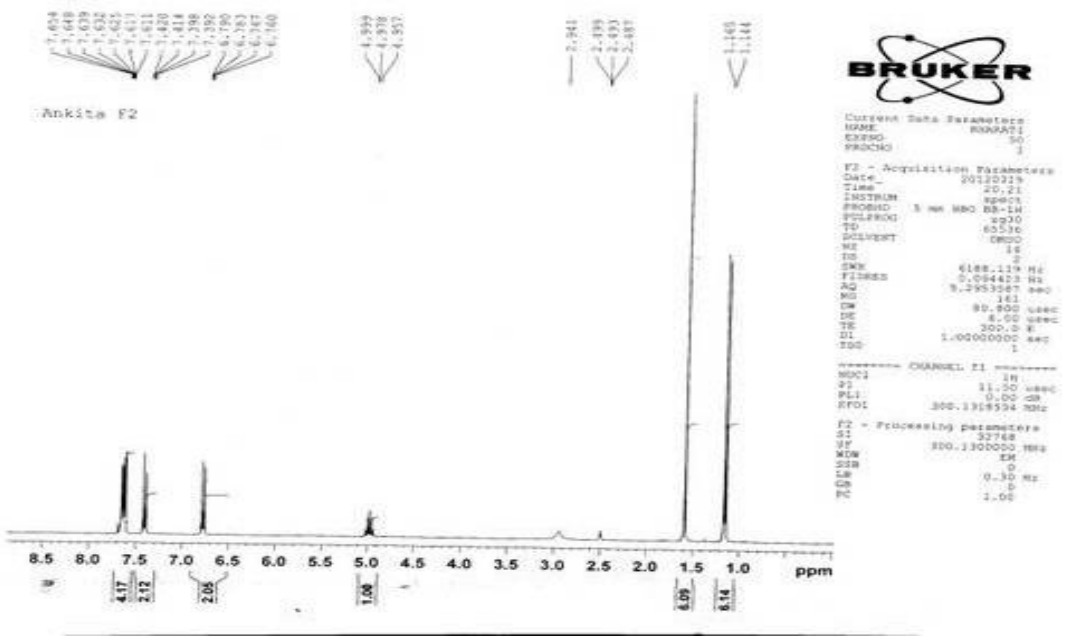

H
N
O
N
O
O
O
CH3
O
CH3
O
CH3
O
CH3
CH3


320
200
%T
100
0
4000
3000
2000
1000
400
Wavenumber [cm-1]

F2


N
N
NH
O
H3C
O
O
CH3
O
CH3
CH3
N





BIOLOGICAL EVALUATION

ANTI CANCER SCREENING:
Trypan Blue Assay [152]
Viable Cell Counts Using Trypan Blue
Trypan Blue is a vital dye. The reactivity of trypan blue is based on the fact that the chromophore is negatively charged and does not interact with the cell unless the membrane is damaged. Therefore, all the cells which exclude the dye are viable.
Procedure: Trypan Blue Staining of Cells
1. Place 0.5 ml of a suitable cell suspension (dilute cells in complete medium Without serum to an approximate concentration of 1 x 105 to 2 x 105 cells per ml) In a screw cap test tube
2. Add 0.1 ml of 0.4% Trypan Blue Stain. Mix thoroughly.
3. Allow to stand 5 min at 15 to 30°C (room temperature).
4. Fill a hemocytometer as for cell counting.
5. Under a microscope, observe if non-viable are stained and viable cells excludedthe stain

ANTICANCER ACTIVITY CARRIED ON THE THREE CELL LINES OF BREAST CANCER

Screening of anticancer activity

Sample	Conc. (ug/ml)	Observed Viable Cell	Total cell count	% Viability	Mean ±SEM
Control	-	89	113	78.76	--
A_1	1000	45	116	32.56	32.09± 0.4250
	100	47	112	34.45	34.12 ± 2.8850
	10	44	95	42.76	42.65 ± 0.5640
	1	45	82	59.45	58.12 ± 3.687
A_2	1000	18	112	16.07	16.44 ± 0.370*
	100	21	126	16.66	17.20 ± 0.540*
	10	27	123	21.95	21.38 ± 0.574*
	1	22	112	19.64	20.18 ± 0.540*
A_3	1000	16	110	14.54	14.13 ± 0.845*
	100	25	130	19.23	19.43 ± 0.675*
	10	29	135	21.48	22.24 ± 0.459*
	1	45	123	36.58	36.64 ± 0.794*
Cyclophosphamide	1000	16	102	15.68	16.42 ± 0.324*
	100	19	113	16.81	17. 32± 0.583*
	10	25	120	20.83	21.48 ± 0.276*
	1	21	108	19.44	20.08 ± 0.470*

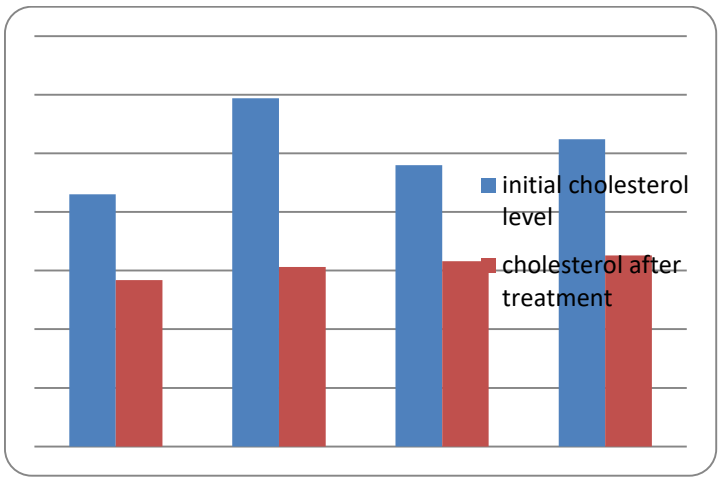
A_2 & A_3 are having excellent anticancer activity as compared to the standard drug Cyclophosphamide. A_1 is having moderate activity.

ANTIHYPERLIPIDEMIC ACTIVITY:

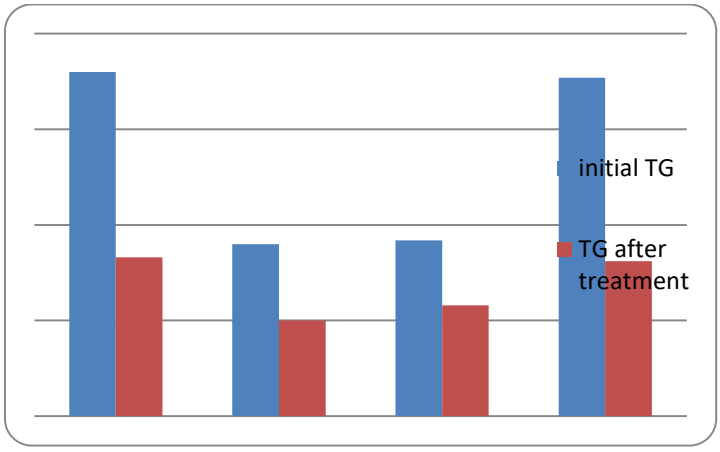
The serum samples containing high triglyceride & cholesterol level were collected from Pravara Medical Trust, Loni. These samples were treated with the test solutions of the synthesized compounds (10mcg/ml). These samples were then incubated for 24 hrs at 37°C. then the samples were tested for SGPT & SGOT to give the decreased triglyceride & cholesterol level.

Screening of antihyperlipidemic activity

Sample	**Test solution(mcg/ml)**	**Dosing (ml)**	**Initial**				**After treatment**			
			Chol.	**T.G.**	**SGPT**	**SGOT**	**Chol.**	**T.G.**	**SGPT**	**SGOT**
D1	10	0.1	215	180	26.2	33.59	142	83	15.247	12.38
D2	10	0.1	297	90	27.0	51.2	153	50	15.25	5.3
D3	10	0.1	240	92	36.7	157	158	58	18.74	33.59
F2	10	0.1	262	177	19.0	21.0	163	81	13.50	5.3



Decrease in cholesterol level



Decrease in TG level

CALCIUM CHANNEL BLOCKER ACTIVITY:

Calcium antagonism in the isolated cock ileum:

Purpose and rationale:

Contraction of ileum was induced by adding potassium chloride & calcium chloride to the organ bath containing slightly modified Tyrode solution (NaCl=8.0gm/l, KCl=0.2gm/l, $CaCl_2$=0.18gm/l, NaH_2PO_4=0.1gm/l, $MgCl_2$=0.1gm/l, Glucose=1.0gm/l, $NaHCO_3$=1.0gm/l). Test drugs with calcium channel blocking activity have a relaxing effect.[154]

Procedure

The assembly was being set up and arrangement was made for experiment. The cock ileum was obtained from the local slaughter house. It was placed in a petridish containing tyrode solution maintained at 37˚C. The mesentery of ileum was removed and the interior content1 was washed by blowing Tyrode solution (NaCl=8.0gm/l, KCl=0.2gm/l, $CaCl_2$=0.18gm/l, NaH_2PO_4=0.1gm/l, $MgCl_2$=0.1gm/l, Glucose=1.0gm/l, $NaHCO_3$=1.0gm/l) with help of pipette. The tissue was mounted in mammalian organ bath (35 ml capacity) and connected to isotonic frontal writing lever. The tissue was allowed to stabilize for 30min. The responses of acetylcholine were taken till the maximum effect was obtained. The responses of acetylcholine were recorded after addition of the test drug in the organ bath (200 mcg). The responses of acetylcholine were taken with same dose and continued till maximum effect obtained. The results were calculated as percent inhibition of contraction compared to the same dose as before addition of test drug solution.[153] Nifedipine was used as standard.

Percent inhibition= 100 - [(size of response after exposure to antagonist/size of response beforeexposure to antagonist) x 100]

Screening of CCB activity

Compound	Dose (ml)		Control (cm)	Test (cm)	% inhibition
	Ach*	Sample			
D_1	0.8	0.02	2.7	1.1	59.26
D_2	0.4	0.02	1.8	1.0	44.45
D_3	0.1	0.02	2.5	1.9	24.00
F_2	0.8	0.02	1.7	0.2	88.24
Nifedipine	12.8	0.02	2.5	0.0	100

*Acetylcholine
Compounds D_1 & F_2 have shown promising relaxing activity as compared to standard Nifedipine.

ANTIOXIDANT ACTIVITY:
METHOD I
Hydrogen peroxide radical scavenging activity[154- 156]: 1mL of test drug/standard (Ascorbic acid) was added to 0.6mL of hydrogen peroxide solution (Ashwin fine chemicals and pharmaceuticals) in phosphate buffer (P^H - 7.4). After incubating for 10 minutes at 37°C the absorbance was measured at 230nm. Corresponding blanks were taken. The experiment was performed in triplicate. The absorbance of hydrogen peroxide in phosphate buffer as control was measured at 230nm. The scavenging effect (%) was measured using following equation. Hydrogen peroxide produces hydroxyl radicals in cells. Scavenging of these radicals by the test drug is used as a test for antioxidant activity. The reduction of these radicals is seen by the decreased absorbance at 230nm with increasing concentration of the test drug.

$$\text{Scavenging Effect (\%)} = \frac{\text{Control absorbance} - \text{Test absorbance}}{\text{Control absorbance}} \times 100$$

Anti-oxidant activity of the synthesized compounds (A_1- A_{16})

Comp. code	Concentration					
	50 µg/mL		150 µg/mL		250 µg/mL	
	Abs.	SE (%)	Abs.	SE (%)	Abs.	SE (%)
A_1	0.012(±0.001)	21.2	0.0095(±0.001)*	40.4	0.0063(±0.001)*	60.1
A_2	0.010(±0.001)	33.5	0.0087(±0.001)*	45.1	0.0041(±0.001)*	**74.3**
A_3	0.012(±0.015)	22.3	0.0069(±0.015)*	56.4	0.0063(±0.0015)*	60.1
B_1	0.010(±0.015)	32.5	0.0068(±0.015)*	57.1	0.0043(±0.015)*	**73.1**
B_2	0.012(±0.015)	22.3	0.0085(±0.015)	46.4	0.0065(±0.015)*	59.3
B_3	0.010(±0.001)	34.5	0.0057(±0.001)*	64.1	0.0037(±0.001)*	**76.7**
C_1	0.011(±0.001)	25.8	0.0095(±0.001)*	40.4	0.0054(±0.001)*	66.1
C_2	0.010(±0.001)	33.1	0.0059(±0.001)*	62.6	0.0039(±0.001)*	**75.4**
C_3	0.011(±0.020)	25.3	0.0074(±0.020)*	47.4	0.0065(±0.020)*	59.3
Ascorbic acid	0.007(±0.015)*	54.3	0.0052(±0.015)*	67.4	0.0023(±0.015)*	85.3

The results are expressed as mean ± SEM (n = 3). Significance was calculated by using one-way ANOVA with Dunnet's t- test. The difference in results was considered significant when $p<0.05$. *$p<0.05$ vs. control Compounds **A_2, B_1, B_3, C_2 &C_3** have shown promising antioxidant activity at 250 µg/mL, while the remaining compounds have also shown moderate antioxidant activity, when compared with standard drug ascorbic acid.

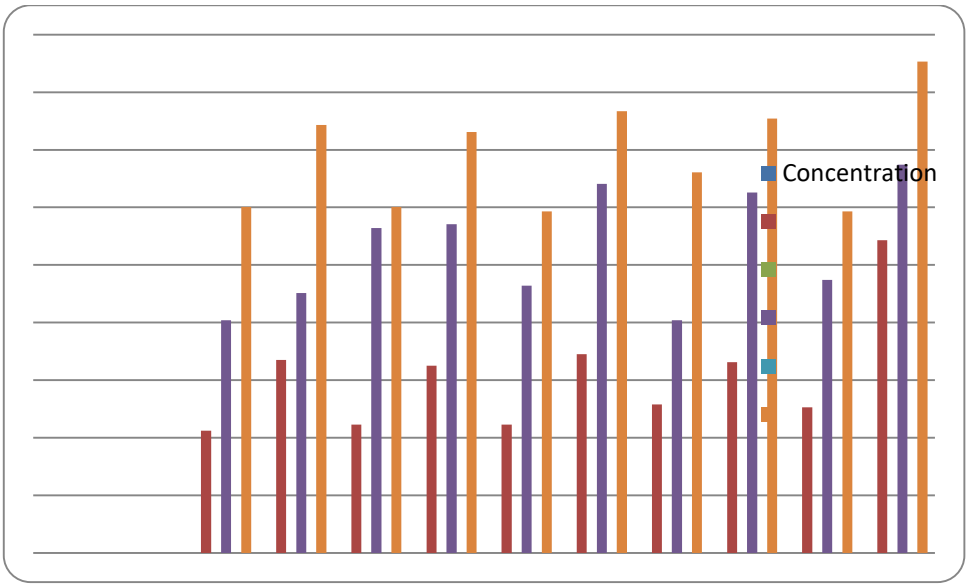
METHOD II:

Phosphomolybdenum method [157, 158]

An aliquot of 0.1 mL of compound solutions was combined with 1 mL of reagent solution (0.6 M sulfuric acid, 28 mm sodium phosphate and 4 mm ammonium molybdate). In case of blank 0.1 mL of methanol was used in place of compound. The tubes were capped and incubated in a boiling water bath at 95^0 C for 90 min. After the samples had cooled to RT, the absorbance of the aqueous solution of each was measured at 695 nm against blank in spectrophotometer. For compound of unknown composition, antioxidant capacity was expressed as equivalent of ascorbic acid (µM/mg of compound).

Evaluation of antioxidant capacity by Phosphomolybdenum method

The antioxidant activity for the synthesized compounds was evaluated by using phosphomolybdate method. It determines the total antioxidant capacity. This assay is based on the reduction of Mo (VI) to Mo (V) in presence of the antioxidant compounds and the subsequent formation of a green phosphate/Mo(V) complex at acidic pH, which is measured at 695 nm. The antioxidant capacity of the compounds was determined for 50 µg/mL, 150 µg/mL and 250 µg/mL concentrations. The antioxidant capacities of the compounds determined by phosphomolybdate method was expressed as µg of ascorbic acid equivalent/mg and showed in the Figure



Total antioxidant capacity of the synthesized compounds.

RESULTS AND DISCUSSION

The synthesized compounds were subjected to various antioxidants, anticancer, calcium channel antagonists and anti-screening by the standard methods.

ANTICANCER ACTIVITY

The compounds containing thiadiazole moiety (A_1, A_2& A_3) were screened for anticancer activity by Trypan blue assay. A_2& A_3 have shown excellent anticancer activity as compared to the standard drug cyclophosphamide.

ANTIHYPERLIPIDEMIC ACTIVITY

The compounds containing combination of fenofibrate (D_1, D_2, D_3 & F_2) were screened for antihyperlipidemic activity by SGPT & SGOT determination. Compounds **D_2& F_2**have shown promising anticholesterol activity. Compounds D_1 & F_2 have shown promising anti-triglyceride activity.

CALCIUM CHANNEL ANTAGONIST ACTIVITY

The compounds of scheme II & scheme III (D_1, D_2, D_3 & F_2) were subjected to calcium channel antagonist activity by using cock ileum relaxation method against acetylcholine. Compounds D_1 & F_2 have shown promising relaxing activity as compared to standard Nifedipine.

ANTIOXIDANT ACTIVITY

All the compounds were screened for antioxidant activity at different concentration. However Compounds **A_2, B_1, B_3, C_2 and C_3** have shown promising antioxidant activity at 250 µg/mL, while the remaining compounds have also shown moderate antioxidant activity, when compared with standard drug ascorbic acid.

The proposed work has given out many active anticancer, antihyperlipidemic, calcium channel antagonists & antioxidant agents. Some of the compounds have showen moderate activities. These compounds with suitable modification can be explored better for their therapeutic activities in the future.

CONCLUSION

1. The present work is a bonafide and novel for the synthesis of pyrimidine derivatives
2. In this view we have made extensive literature review on substituted pyrimidine derivatives for their medicinal values with the help of chemical abstracts, journals and internet surfing and text books.
3. For the synthesis of pyrimidine derivatives scheme was established based on the literature survey.
4. Around 13 new pyrimidine derivatives were synthesized, with the standard chemicals and well established procedures.
5. The synthesized compounds were tested for their Preliminary Tests, Physical Constants, and TLC etc.
6. The structures of the final compounds were confirmed by IR, ^{1}H-NMR Spectra and CHN analysis.
7. The proposed compounds were screened for their Anticancer, Antihyperlipidemic, calcium channel antagonist & Antioxidant activities with the standard drugs in the well-equipped pharmacology lab by using standard methods.
8. A_2& A_3 have shown excellent anticancer activity as compared to the standard drug cyclophosphamide.
9. Compounds **D_2& F_2**have shown promising anticholesterol activity. Compounds D_1 & F_2 have shown promising anti-triglyceride activity.
10. Compounds D_1 & F_2 have shown promising relaxing activity as compared to standard Nifedipine.
11. Compounds **A_2, B_1, B_3, C_2 and C_3** have shown promising antioxidant activity at 250µg/mL, while the remaining compounds have also shown moderate antioxidant activity, when compared with standard drug ascorbic acid.
12. The proposed work has given out many active anticancer, antihyperlipidemic, calcium channel antagonists & antioxidant agents. Some of the compounds have shown moderate activities. These

compounds with suitable modification can be explored better for their therapeutic activities in the future.

BIBLIOGRAPHY

1. Karpozilos A, Pavlidis N. "The treatment of cancer in Greek antiquity". *European Journal of Cancer***2004**. 40 (14): 2033–40.
2. Moss, Ralph W. "Galen on Cancer". CancerDecisions. **2004**.http://www.cancerdecisions.com/speeches/galen1989.html.
3. Cancer Research UK (January 2007). "UK cancer incidence statistics by age". http://info.cancerresearchuk.org/cancerstats/incidence/age/. Retrieved **2007-06**-25.
4. WHO. "Cancer". World Health Organization February **2006** http://www.who.int/mediacentre/factsheets/fs297/en/. Retrieved 2007-06-25.
5. American Cancer Society (December 2007). "Report sees 7.6 million global 2007 cancer deaths". Reuters. http://www.reuters.com/article/healthNews/idUSN1633064920071217. Retrieved **2008-08-07**.
6. Kinzler, Kenneth W.; Vogelstein, Bert. "Introduction". *The genetic basis of human cancer* (2nd, illustrated, revised ed.). New York: McGraw-Hill, Medical Pub. Division.**2002** p. 5. ISBN978-0-07-137050-9.
7. "WHO Disease and injury country estimates". *World Health Organization*. 2009. http://www.who.int/healthinfo/global_burden_disease/estimates_country/en/index.html. Retrieved Nov. 11, **2009.**
8. "Cancer" (PDF). *World Health Organization*. http://www.who.int/nmh/publications/fact_sheet_cancers_en.pdf
9. *World Health Organization*. http://www.who.int/mediacentre/factsheets/fs297/en/
10. Croce CM. "Oncogenes and cancer". *The New England journal of medicine***358,** January **2008** (5): 502–11. doi:10.1056/NEJMra072367. PMID18234754. http://content.nejm.org/cgi/content/full/358/5/502.
11. Knudson AG (November 2001). "Two genetic hits (more or less) to cancer". *Nature reviews. Cancer* November **2001**, **1** (2): 157–62. doi:10.1038/35101031. PMID11905807.
12. "Cancer prevention: 7 steps to reduce your risk". Mayo Clinic. 2008-09-27. http://www.mayoclinic.com/health/cancer-prevention/CA00024. Retrieved **2010**-01-30.
13. Cancer". *World Health Organization*. http://www.who.int/mediacentre/factsheets/fs297/en/
14. The Pharmacological Basis of Therapeutics by Alfred Goodman Gilman, McGraw- Hill Medical Publishing Division 10th edition, **2001**, 1284.
15. Adeline B, Juan CRD, Gilbert K, Denyse B. Synthesis and protective effects of coumarin derivatives against oxidative stress induced by doxorubicin. *Bioorg Med Chem Lett* 2008; 18:1102-5.
16. George K, Vassiliki T, Chryssa T. Synthesis and *in vitro* cytotoxicity of aminocoumarin platinum(II) complexes. Synthesis and *in vitro* cytotoxicity of aminocoumarin platinum(II) complexes. *Bioorg Med Chem Lett* 1997; 7: 2165- 68.
17. Irena K, Ilia M, Irina N, Spiro K, Margarita K. New lanthanide complexes of 4-methyl-7-hydroxycoumarin and their pharmacological activity. *Eur J Med Chem* 2001; 36:339-47.
18. Natala SR, Muralidhar R, Stephen C, Kiranmai G, Stanley CB, Premkumar R. Synthesis of new coumarin 3-(N-aryl) sulphonamides and their anticancer activity. *Bioorg Med Chem Lett* 2004; 14: 4093-7.
19. Irena K, Momekov G. New zirconium (IV) complexes of coumarins with cytotoxic activity. *Eur J Med Chem* 2006; 41: 717-26.
20. Wei-wei M, Ting-ting W, He-ping Z, Zhi-yu W, Jian-ping C, Jian-gang S. Synthesis and evaluation of novel substituted 5-hydroxycoumarin and pyranocoumarin derivatives exhibiting signi.cant antiproliferative activity against breast cancer cell lines. *Bioorg Med Chem Lett* 2009; 19: 4570-3.

21. Jolanta NM, Ewa N, Julita G. *In vivo* antitumor, *in vitro* antibacterial activity and alkylating properties of phosphorohydrazine derivatives of coumarin and chromone. *Eur J Med Chem* 2006; 41:1301-9.
22. Sathisha MP, Ullas N Shetti, Revankar VK, Pai KSR. Synthesis and antitumor studies on novel Co(II), Ni(II) and Cu(II) metal complexes of bis (3-acetylcoumarin) thiocarbohydrazone. *Eur J Med Chem* 2008; 43: 2338-46.
23. Stancho S, Georgi M, Frank J, Ilia M. Synthesis, computational study and cytotoxic activity of new 4-hydroxycoumarin derivatives. *Eur J Med Chem* 2008; 43:694-706.
24. Elzbieta B, Elzbieta B, Urszula K, Marek R. Cytotoxiceffects, alkylating properties and molecular modeling of coumarin derivatives and their phosphonic analogues. *Eur J Med Chem* 2003; 38:597-603.
25. Jae-Chul J, Ji-Ho L, Seikwan O, Jae-Gon L, Oee-Sook PS. Synthesis and antitumor activity of 4-hydroxycoumarin derivatives. *Bioorg Med Chem Lett* 2004; 14:5527-31.
26. Irena K, Ilia M, Spiro K, Margarita K. Synthesis, physicochemical characterization and cytotoxic screening of new complexes of cerium, lanthanum and neodymium with Warfarin and Coumachlor sodium salts. *Eur.J.Med.Chem* 1999; 34:63-68.
27. Irena K, Ilia M, Georgi M. Cytotoxic activity of new neodymium (III) complexes of bis-coumarins. *Eur J Med Chem* 2004; 39:765-75.
28. Irena K, Georgi M. New cerium (III) complexes of coumarins - Synthesis, characterization and cytotoxicity evaluation. *Eur J Med Chem* 2008; 43:178-88.
29. Irena K, Ilia M, Spiro K, Margarita K. Synthesis, physicochemical characterization and cytotoxic screeningof new complexes of cerium, lanthanum and neodymium with warfarin and coumachlor sodium salts. *Eur J Med Chem* 1998; 34:63-8.
30. Magda MFI, Heba SR, Mohammad MMH. Synthesis and docking studies of novel benzopyran-2-ones with anticancer activity. *Eur J Med Chem* 2010; 45: 3950-9
31. SergioV, Emilie B, Elodie V, Denyse B, Gilbert K. Synthesis and biological evaluation of novel coumarin based inhibitors of Cdc25 phosphatases. *Bioorg Med Chem Lett* 2010; 20:5827-30.
32. Yves J, Ioanna L, Anny C, Denis N, Laurent B, Bernard R *et al*. Synthesis, structure, and estrogenic activity of 4-amino-3-(2-methylbenzyl) coumarins on human breast carcinoma cells. *Bioorg Med Chem Lett* 2007; 15:2269-82.
33. Nermien M. Sabry , Hany M. Mohamed, Essam Shawky A.E.H. Khattab , Shymaa S. Motlaq , Ahmed M. El-Agrody, Synthesis of 4H-chromene, coumarin, 12H-chromeno[2,3-d]pyrimidine derivatives and some of their antimicrobial and cytotoxicity activities. *Eur J Med Chem* 46 (2011) 765-772
34. Marta P, Neves, Cidade H, Madalena P, Artur M.S, Prenylated derivatives of baicalein and 3,7-dihydroxy.avone: Synthesis and study of their effects on tumor cell lines growth, cell cycle and apoptosis. *Eur J Med Chem* 2011; 46: 2562-2574
35. Joanna T, Hejchman E, Kruszewska H, Wolska I. Synthesis and pharmacological activity of O-aminoalkyl derivatives of 7-hydroxycoumarin. Eur J Med Chem 2011; 46:2252-2263
36. Rangappa SK, Kallappa MH, Ramya VS, Mallinath HH. Analgesic, anti-pyretic and DNA cleavage studies of novel pyrimidine derivatives of coumarin moiety. *Eur J Med Chem* 2010; 45:2597-2605.
37. Ilia M, Danchev ND. Synthesis, toxicological and pharmacological assessment of some 4-hydroxycoumarin derivatives. *Eur J Med Chem* 1995; 30:531-5.
38. Ilia M, Caecilia M, Nicolay D. Synthesis, structure, toxicological and pharmacological investigations of 4-hydroxycoumarin derivatives. *Eur J Med Chem* 2006; 41:882-90.
39. Jeremiah JS, Ashley KW, Yousef RM, Michael DC, David EL. New adjuvants to enhance anticoagulant activity of Warfarin. *Bioorg Med Chem Lett* 2010; 20:1928-32.

40. Omaima MA, Kamelia MA, Rasha ZB, Timothy JM, Somaia AN, Shalini S. Synthesis, anticoagulant and PIVKA-II induced by new 4-hydroxy coumarin derivatives. *Bioorg Med Chem Lett* 2010; 18:3371-8.
41. Mashelkar UC, Audi AA. Synthesis of some novel 4-substituted coumarins having potential biological activity (Part III). *Indian J Chem* 2006; 45B:1463-9.
42. Destors JM, Boissel JP, Philippon AM, et al; Controlled clinical trial of bepridil, propranolol and placebo in the treatment of exercise induced angina pectoris. B.I.S. Research Group. Fundam Clin Pharmacol. 1989; 3(6):597-611. [abstract]
43. Singh S; Long-term double-blind evaluation of amlodipine and nadolol in patients with stable exertional angina pectoris. The Investigators of Study 152. Clin Cardiol. 1993 Jan; 16(1):54-8. [abstract]
44. Vliegen HW, van der Wall EE, Niemeyer MG, et al; Long-term efficacy of diltiazem controlled release versus metoprolol in patients with stable angina pectoris. J Cardiovasc Pharmacol. 1991; 18 Suppl 9:S55-60. [abstract]
45. Hall R, Chong C; A double-blind, parallel-group study of amlodipine versus long-acting nitrate in the management of elderly patients with stable angina. Cardiology. 2001; 96(2):72-7. [abstract]
46. Lawes CM, Bennett DA, Feigin VL, et al; Blood pressure and stroke: an overview of published reviews. Stroke. 2004 Apr; 35(4):1024. [abstract]
47. Sarkozi J, Bookman AA, Mahon W, et al; Nifedipine in the treatment of idiopathic Raynaud's syndrome. J Rheumatol. 1986 Apr; 13(2):331-6. [abstract]
48. Gjorup T, Kelbaek H, Hartling OJ, et al; Controlled double-blind trial of the clinical effect of nifedipine in the treatment of idiopathic Raynaud's phenomenon. Am Heart J. 1986 Apr; 111(4):742-5. [abstract]
49. Waller DG, Challenor VF, Francis DA, et al; Clinical and rheological effects of nifedipine in Raynaud's phenomenon. Br J Clin Pharmacol. 1986 Oct; 22(4):449-54. [abstract]
50. Phillips BG, Gandhi AJ, Sanoski CA, et al; Comparison of intravenous diltiazem and verapamil for the acute treatment of atrial fibrillation and atrial flutter. Pharmacotherapy. 1997 Nov-Dec; 17(6):1238-45. [abstract]
51. Siu CW, Lau CP, Lee WL, et al; Intravenous diltiazem is superior to intravenous amiodarone or digoxin for achieving ventricular rate control in patients with acute uncomplicated atrial fibrillation. Crit Care Med. 2009 Jul;37(7):2174-9; quiz 2180. [abstract]
52. Cohen AS, Matharu MS, Goadsby PJ; Electrocardiographic abnormalities in patients with cluster headache on verapamil therapy. Neurology. 2007 Aug 14; 69(7):668-75. [abstract]
53. Calcium channel blocker" at Dorland's Medical Dictionary; a b Nelson, Mark. "Drug treatment of elevated blood pressure". Australian Prescriber (33): 108–112. Retrieved August 11, 2010.
54. Iseri LT & French JH. “Magnesium: nature’s physiologic calcium channel blocker.” Am Heart J, 108, 188-193, 1984.
55. Lehne, Richard (2010). Pharmacology for Nursing Care, Seventh Edition. St. Louis, Missouri: Saunders Elsevier. pp. 505. ISBN 978-1-4160-6249-3.
56. Remuzzi G, Scheppati A, Ruggenenti P: Nephropathy in patients with type 2 diabetes. N Engl J Med 2002; 346:1145.
57. Choice of lipid-regulating drugs. Med Lett 2001; 43 (Issue 1105):43–48.
58. Collins R, Peto R, and Armitage J. The MCR/BHF: Heart Protection Study: preliminary results. Int J Clin Pract 2002; 56:53–56.
59. Contemporary management of lipid disorders: The evolving importance of statin therapy. Clin Courier 1998; 28 (No. 35):1–7.
60. Downs JR et al. Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels. Results of AFCAPS/TEXCAPS. JAMA 1998; 279:1615–1622.
61. Knopp RH. Drug treatment of lipid disorders. N Engl J Med 1999; 341:498–511.

62. Ornish D et al. Intensive lifestyle changes for reversal of coronary heart disease. JAMA 1998; 280:2001–2007.
63. Ross R. Atherosclerosis: An inflammatory disease. N Engl J Med 1999; 340:115–126.
64. Sacks FM et al.The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. N Engl J Med 1996; 335:1001–1009.
65. Shepherd J et al. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. N Engl J Med 1995; 333:1301–1307.
66. Tanne D et al. Blood lipids and first-ever ischemic stroke/transient ischemic attack in the Benzafibrate Infarction Prevention (BIP) Registry: High triglycerides constitute an independent risk factor. Circulation 2001; 104:2892–2897.
67. Nandeeshwariah S K. and Sarvottam Ambekar., *Ind. J. Chem.*, **1998**: 37B: 995.
68. Manish Shah., Khyati Parikh. and Hansa parekh., *Ind. J. Chem.*, **1998**: 37B: 73.
69. Upadhyay D N. and Vishnu J Ram., *Ind. J.Chem.*, **1999**: 38B: 173.
70. Filler R., *J. Fluorine Chem.*, **1999**: 33: 362.
71. Pramar J M., Modha J J. and Parikh A R., *Ind. J. Chem.*, **1999:** 38B: 440.
72. Sondhi S M., Verma R P., *Indian Drugs*, **1999:** 36(1): 50
73. Pratibha Sharma, Nilesh Rane and V. K. Gurram, *Bioorganic & Medicinal Chemistry Letters* 14 **(2004)** 4185–4190.
74. Dana Hockov,Anton_ın Hol, Milena Maso, raciela Andrei, Robert Snoeck, Erik De Clercq and Jan Balzarinib *Bioorganic & Medicinal Chemistry* 12 **(2004)** 3197–3202
75. M.Saxena, L.K Soni, *Indian J of Biochem. And Biophysics*, vol.43,Feb-**2006**,32-36.
76. W.S.Hamouly,*Indian J of Chem*.,45B,**2006,**2091-98.
77. Marcus H. Holschbach , Dirk Bier, Stefan Stüsgen, Walter Wutz, Wiebke Sihver, Heinz H. Coenen, Ray A. Olsson 6. *European Journal of Medicinal Chemistry* 41 (**2006**) 7–15.
78. R B Patel, P S Desai, K R Desai, Synthesis of pyrimidine based thiazolidinones and azetidinones: Antimicrobial and antitubercular agents, *Indian Journal of Chemistry*, Vol. 45B, Marc h **2006**, pp. 773-778.
79. Jack J. Kinsorac and Kenneth S. Kilgorec *Bioorganic & Medicinal Chemistry Letters* 17 **(2007)** 5396–5399.
80. Mehdi Adib, A new, one-pot, three-component synthesis of 4H-pyrido[1,2-a]- pyrimidines, 4H-pyrimido[1,2-a]pyrimidines, and 4H-pyrazino[1,2-a]pyrimidines, *Tetrahedron* 63 **(2007)** 11135–11140.
81. T. D. Venu , S. A. Khanum , Aiysha Firdouse , B. K. Manuprasad , Sheena Shashikanth, Riyaz Mohamed , Bannikuppe Sannanaik Vishwanth, *Bioorganic & Medicinal Chemistry Letters* 18 **(2008)** 4409–4412.
82. Jin Pei Zhou, Yi Wei Ding , Hui Bin Zhang , Lian Xu , Yue Dai ,*Chinese Chemical Letters* 19 **(2008)** 669–672.
83. Nilo Zanatta , Simone S. Amaral, Convergent synthesis and cruzain inhibitory activity of novel 2-(N0-benzylidenehydrazino)-4-trifluoromethyl- pyrimidines, *Bioorganic & Medicinal Chemistry* 16 **(2008)** 10236–10243.
84. Tarik El-Sayed Ali *European Journal of Medicinal Chemistry* 44 **(2009)** 4385–43.
85. Jing Bo Shi , Jing Gao , Ya Ping Wang , Qi Zheng Yao ,*Chinese Chemical Letters* 20 **(2009)** 404–406.
86. H. Cho, M. Ueda, K. Shima, A. Mizuno, M. Hayashimatsu, Y. Ohnaka, Y. Takeuchi, M. Hamaguchi, and K. Aisaka, *J. Med. Chem*., **32** (10), 2399(1989).
87. K. S. Atwal, B. N. Swanson, S. E. Unger, D. M. Floyd, S. Moreland, A. Hedberg and B. C. O'Reilly *J. Med. Chem.*, **34** (2), 806 (1991).
88. (a) R. Neumann and R. Morten, *Drugs of Todays*, **21**, 133 (1998). (b) R. K. Robin, *Chem EngNews*, 28(1986).

89. (a) F. Focher, D. Ubiali, M. Pregnolato, C. Zhi, J. Bambio, G. E. Wright and S. Spadri, *J. Med.Chem.*, 42, 2601 (2000). (b) A. Matsuda, H. Hattori, M. Tanaka and T. Sasaki, *Bioorg MedChemLett*, 6, 1887 (1996).
90. (a) H. Sherman & A. M. Kaplan, *Toxicol Appl Phermacol*, **34**, 189 (1975). (b) S. J. Stewart, *AUPatent* **2001**, 720032; *Chem. Abstr.*, 134, 67474 (2001). (c) G. D. Daves Jr, R. K. Robins and C. C. Cheng, *J. Org. Chem.*, **26**, 5256 (1961). Vol.2, No.3 (2009), 662-676 CHEMISTRY OF DIHYDROPYRIMIDINONES I. T. Phucho et al. 676
91. J. Stoltefuss, H. Boeshagen, M. Schramm and G. Thomas, *Chem. Abstr.*, 101, **1984**, 55110v (Bayer AG) *Ger Offen DE*, 3234 684(1984).
92. H. Cho, M. Ueda, K. Shima, A. Mizuno, M. Hayashimatsu, Y. Ohnaka, Y. Takeuchi, M. Hamaguchi, K. Aisaka, T. Hidaka, M. Kawai, M. Takeda, T . Ishihara, K. Funahashi, F. Satah, M. Morita and T. Noguchi, *J Med Chem*, 32, 2399 (1989).
93. F. Bossert and W. Vater, *Med. Res. Rev.*, **9**, 291 (1989).
94. (a) C. O. Kappe, *Acc Chem Res*, 879(2000). (b) C. O. Kappe, *Molecules* ,1 (1998). (c) B. Jauk, T. Pernant and C. O. Kappe, *Molecules*, 227 (2000).
95. A. D. Patil, N. V. Kumar, W. C. Kokke, M. F. Bean, A. J. Freger, C. Debrossi, S. Mai, A. Truneh, D. J. Faulkner, B. Carte, A. L. Breen, R. P. Hertzbery, R. K. Johnson, J. W. Westley and B. C. M. Potts, *J Org Chem*, 60, 1182 (1995).
96. M. C. Parlato, C. Mugnaini, M. L. Renzulli, F. Corelli and M. Botta, *Arkivoc*, 5, 349 (2004).
97. (a) K. S. Atwal, G. C. Rovnyak, B. C. O' Reilly and J. Schwartz, *J. Org. Chem.*, **54**,
98. 5898(1989). (b) C. O. Kappe, W. M. F. Fabian and M. A. Semones, *Tetrahedron*, **53**, 2803 (1997).
99. [Rl Kenner, G. W.; Todd, A. Pyrimidine and Its Derivatives. Hererocyclic Compounds; John Wiley & Sons: New York, 1957.
100. Brown, D. J. The Pyrimidines; John Wiley & Sons: New York, 1994.
101. Pinner, A. Ber. 1893,26,2122.
102. Pinner, A. Ber. 1884, 17.
103. Pinner, A. Ber. 1908,41,3517.
104. Ghosh, U.; Katzenellenbogen, J. A. J. Heterocycl. Chem. 2002, 39, 1101.
105. Katritzky, A. R.; Yousaf, T. 1. Can. J. Chem. 1986,64,2087.
106. Bowman, A. J. Chem. SOC. 1937,494.
107. Snyder, H. R.; Foster, H. M. J. Am. Chem. SOC. 1954, 76, 1 18.
108. Donleavy, J. J.; Kise, M. A. Org. Synth. 1943, Coll. Vol. 11, John Wiley & Sons, 422.
109. Todd, A. R.; Bergel, F. J. Chem. SOC. 1937,364.
110. Kenner, G. W.; Lythcoe, A. R.; Topham, A. J. Chem. SOC. 1943,388.
111. Maggiolo, A.; Phillips, A. P.; Hitchings, G. J. Am. Chem. SOC. 1951, 73, 106.
112. Biginelli, P. Gnu. Chim. Ital. 1893, 23,360.
113. [Rl (a) Kappe, C. 0. Tetrahedron 1993.49, 6937. [RI (b) Kappe, C. 0. Acc. Chem. Res. 2000,33,879. [R]
114. (c) Kappe, C. 0. Eur. J. Med. Chem. 2000,35,1043.
115. Yarim, M.; Sarq, S.; KiliG, F. S.; Erol, K. Farmaco 2003,58, 17. ,
116. (a) Overman. L. E.; Rabinowitz, M. H. J. Org. Chem. 1993.58.3235. (b) Coffey, D. S.; McDonald, A. I.;
117. Overman, L. E.; Rabinowitz; Renhowe, P. A. J. Am. Chem. SOC. 2000,122.4893. (c) Coffey, D. S.; Overman,
118. L. E.; Stappenbeck, F. J. Am Chem. SOC. 2000, 122,4904. (d) Cohen, F.; Overman, L. E. J. Am. Chem SOC. 2001, 123, 10782. (e) Overman, L. E.; Wolfe, J. P. J. Org. Chem. 2001, 66, 3167. (9 Cohen, F.; Collins, S.
119. K.; Overman. L. E. Org. Lett. 2003,5,4485. (g) Aron, 2. D.: Overman, L. E. Chem. Commun. 2004,253.
120. Folkers, K.; Johnson, T. B. J. Am. Chem. SOC. 1933,55,3784.

121. Sweet, F.; Fissekis, J. D. J. Am. Chem. SOC. 1973,95,8741.
122. Kappe, C. 0. J. Org. Chem. 1997,62,7201.
123. Bose, D. S.; Fatima, L.; Mereyala, H. B. J. Org. Chem. 2003,68,587.
124. Hu, E. H.; Sidler, D. R.; Dolling, U.-H. J. Org. Chem. 1998.63.3454,
125. Lu, J.; Bai, Y. Synthesis 2002,466.
126. Ranu, B. C.; Hajra, A.; Jana. U. J. Org. Chem. 2000,65,6270.
127. Ramalinga K.; Vijayalakshmi, P.; Kaimal, T. N. B. Synletr 2001,863.
128. Varala, R.; Alam, M. M.; Adapa, S. R. Synlett 2003,67.
129. Ma, Y.; Qian, C.; Wang, L.; Yang, M. J. Org. Chem. 2000.65, 3864.
130. Kappe, C. 0.; Falsone, S. F. Synlert 1998,718.
131. Fan, X.; Zhang, X.; Zhang, Y. J. Chem. Rex, Synop. 2002,436.
132. (a) Baruah, P. P.; Gadhwal. S.; Prajapati, D.; Sandhu, J. S. Chem. Len. 2002, 1038. (b) Maiti, G.; Kundu, P.;
133. Guin, C. Tetrahedron Lett. 2003,44,2757.
134. Fu, N.-Y.; Yuan, Y.-F. Cao, 2.; Wang, S.-W. Wang, J.-T.; Peppe, C. Tetrahedron 2002.58.4801.
135. (a) Lu, J.; Bai, Y.; Wang, Z.; Yang, B.; Ma, H. Tetrahedron Lett. 2000.41, 9075. (b) Lu, 1.; Bai, Y.-J.; Guo,
136. Y.-H. Wang,Z.-J.; Ma, H.-R. ChineseJ. Chem. 2002.20.681.
137. Chen, R.-F.; Qiang, C.-T. Chinese J. Chem. 2002,20,427.
138. Kumar, K. A,; Kasthuraiah, M.; Reddy, C. S.; Reddy, C. D. Tetrahedron Lett. 2001,42,7873.
139. Yadav, J. S.; Reddy, B. V. S. Srinivas, R.; Venugopal, C.; Ramalingam, T. Synthesis 2001,1341.
140. Peng, J.; Deng, Y. Tetrahedron Lett. 2001.42, 5917.
141. Folkers, K.; Hanvood, H. J.; Johnson, T. B. J. Am. Chem. SOC. 1932,.54,375 1.
142. Eynde, J. V.; Audiart, N.; Canonne, V.; Michel, S.; Haverbeke, Y. V.; Kappe, C. 0. Heterocycles 1997, 4S, 1967.
143. Jin, T.; Zhang, S.; Li, T. Synth. Comm. 2002,32, 1847.
144. Bussolari, J. C.; McDonnell, P. A. J. Org. Chem. 2000,6S, 6777.
145. Shaabani, A.; Bazgir, A.; Teimouri, F. Tetrahedron Lert. 2003,44, 857.
146. Phucho I. T., Nongpiur A., Tumtin S. Nongrum R., Nongkhlaw R. L., Recent progresses in chemistry of dihydropyrimidones, Rasayan J. Chem, 2009, 2 (3), 662-676.
147. Nagaraja G. K., Kumarswamy M. N., Vaidya V. P., Mahadevan K. M., ARKIVOC 2006 (x) 211-219.
148. Rajeeva B., Srinivasulu N., Shantakumar S. M., European J. Chemistry, 2009, 6(3), 775-779.
149. Pattan S. R., Kekare P., Dighe N. S., Nirmal S. A., Musmade D. S., Parjane S. K., Daithankar A. V., J. chemical and p'ceutical res., 2009, 1(1), 191-198.
150. Madhusudana Reddy M. B., Pasha M. A., Indian J. Chem., 2012, 51B, 537-541.
151. Lakshmikantam M., Mahendar K., Bhargava S., J. Chem. Sci., 2010, 122(1), 63-69.
152. Freshney R., Culture of animal Cells, A manual basic technique ,**1987**,20-25.
153. Ghosh MN, Editor, Fundamentals of Experimental Pharmacology, 2nd edition. Calcutta: Scientific Book Agency; 1984: 108.
154. Vilasrao J. Kadam, Yadunath M. Joshi, Harshad P. Sawant, Tej A. Jadhav Free Radical Scavenging Activity Of Aqueous Solution Of Black Salt .*Int J Pharmacy Pharm Sci* 2010: 2 : 95-96.
155. A. Shyam Sunder , A.Rama Narasimha Reddy, D. Krishna Prasad , K. Poorna Chander And Free Radical Scavenging Activity Of Methanolic Whole Plant Extract Of *Trianthema Portulacastrum* Linn (Aizoaceae)*Int.J.Ph.Sci.,May-Aug 2010;2(2):589-592.*
156. M. Elmastaş, I. Gülçin, Ö. Işildak, Ö.I. Küfrevioglu, K. Ibaoglu and H.Y. Aboul-Enein, Radical Scavenging Activity and Antioxidant Capacity of Bay Leaf Extracts *Journal of the Iranian Chemical Society*, Vol. 3, No. 3, September 2006, pp. 258-266.

157. Prieto Pineda M and Aguilar M, Spectrophotometric quantitation of antioxidant capacity through the formation of a phosphomolybdenum complex: , *Anal Biochem* 1999, 269, 337.
158. H. Vijay Kumar, C. R. Gnanendra, Nagaraja Naik And D. Channe Gowda, In Vitro Antioxidant Activity Of Benz[B,F]Azepine And Its Analogues, *E-Journal Of Chemistry* 2008;5(S2): 1123-1132.