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SYNTHESIS & EVALUATION OF SOME NOVEL 1,4 DIHYDROPYRIDINE DERIVATIVS FOR THEIR ANTICONVULSANT ACTIVITY

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ABSTRACT

The main of the study was to synthesize a series of 1, 4-dihydropyridine derivatives and tested for their anticonvulsants activity. All the synthesized novel compounds were screened for anticonvulsants activity. The anticonvulsant activities of these compounds were evaluated by using the maximal electroshock seizure (MES) and subcutaneous pentylenetetrazole (scPTZ) seizure models in mice. It was also found that P-03 compound was more potent as compared to other synthesized compounds with anticonvulsants activity. Whereas all the other compound have showed mild to moderate anticonvulsants activity as compared to standard drug. Syntheses have been carried out following simple methodology in excellent isolated yields. The structure and purity of the original compounds were confirmed or identified by melting point, TLC, IR spectra analysis and elemental analysis. These preliminary results indicate that some of compounds are exhibiting good anticonvulsants activity.

KEYWORDS: 1, 4-dihydropyridine, Ethylacetoacetate, Chloroacetylchloride, Synthesis, Silica Gel, anticonvulsants activity, maximal electroshock, Chemotherapeutic Science International.

INTRODUCTION

Calcium channel blockers (1,4-dihydro-pyridines) are reported to be effective against the whole range of convulsive procedures, including electro and pentylenetetrazole convulsions and sound and high pressure-induced seizures. Nifedipine and other dihydropyridine derivatives such as nimodipine, nitrendipine and nisoldipine are potent blockers of the calcium channels of smooth muscles and also bind with high affinity to the brain membranes. There are considerable evidences that calcium is an important factor for the induction of epilepsy. Specifically different seizure-inducing agents or procedures cause a rapid intraneuronal influx of calcium ions, which is mainly related to the subsequant epileptiform activity. Literature survey reveals that 1,3,4-Thiadiazoline derivatives are active against many Mycobacterias as anti-TBanti-bacterial, anti-fungal, anti- oxidizing agents, anti-cancer,anticonvulsant & antidepressantsactivity. Further, the anticonvulsant activity of several amide derivatives is attributed to the allosteric modulation of the GABA and affinity to the voltage-sensitive calcium channel receptors. In the present work we attemped to investigate some 1,4- dihydro-pyridines and their molecular hybrids with 1,3,4-thiadiazole linked by amide moiety in order to reveal higher anticonvulsant activity.

Prompted by the observed biological activities of the above mentioned derivatives and in continuation of our ongoing studies on novel biologically active molecules, we have designed and synthesized some 1, 4-dihydropyridine derivatives as potential anticonvulsant agents.

EPILEPSY[1]

Epilepsy is a neurological disorder of varied etiology. It is characterized by paroxysmal, excessive and hyper synchronous discharge of large number of neurons. Approximate 50 million people worldwide suffer from epilepsy, making this condition the second leading neurological disorder. It is estimated that 25% of the epileptic population have seizures that are not responsive to presently available medical therapies. Despite the optimal use of available antiepileptic drugs, many patients fail to experience seizure control and other do so only at the expense of significant toxic effects that range in severity from minimal brain impairment to death from aplastic anemia or hepatic failure. It is estimated that available medication controls the seizures in only 50% of patients or decrease incidence in only 75% of patients. These facts make the field of anticonvulsant drug discovery a high priority. The search continues for the ideal drug that should be potent, selective in raising seizure threshold and preventing seizures spread without causing serious side effects.

CLASSIFICATION OF EPILEPSY/ SEIZURES[1]

Seizures can be categorized into

Partial (focal) seizures: The symptoms of these seizures depend upon the site excessive neuronal discharge and the extent to which the abnormal electrical activity spreads to other brain neurons. It is of following types:

Simple partial seizures

Abnormal electrical activity is confined to a localized area of motor cortex so that the seizures are limited to one limb or a group of muscles controlled by that particular brain region. There is some sensory disturbance but no loss of consciousness. This seizure is also known as Jacksonian epilepsy and can occur at any stage.

Complex partial seizures (psychomotor or temporal epilepsy)

There is complex sensory hallucination, bizarre behavior and loss of consciousness. Motor movements may be repetitive in nature (stereotype). Incidences usually occur below the age of 20 years.

Generalized seizures

These seizures involve abnormal electrical discharge throughout both hemispheres. Though the seizures may be convulsive or non convulsive, there is usually an immediate loss of consciousness.

Tonic-clonic (grand mal) seizures

It is the commonest type of epilepsy. There is a loss of consciousness followed by tonic and then clonic phases. These seizures are generally followed by apostictal period of confusion, muscle weakness and exhaustion

Absence (petit mal) seizures: There is brief, abrupt and self-limiting loss of consciousness, but no convulsions. The patient is usually prepubertal and exhibits vacant stare with rapid eye blinking lasting for a few seconds.

Myoclonic seizures: These seizures consist of short episodic convulsions, which may reoccur after a few minutes. They may occur at any age and are usually due to underlying permanent neurological damage.

Febrile seizures: Young children may exhibit convulsions concomitant with hyperpyrexia. They are of tonic-clonic type and are of short duration. They do not cause any neurological damage and rarely require medication.

Status epilepticus: They are rapid recurrent grand mal seizures. This is emergency condition requiring immediate treatment.

CLASSIFICATION OF ANTIEPILEPTIC DRUGS[1,3]

- **First generation antiepileptic drugs**

Table 1.1: Shows the first-generation antiepileptic drugs.

Category of drug	Examples
Barbiturate	Phenobarbitone
Hydantoin	Phenytoin, Ethatoin
Iminostilbenes	Carbamazepine
Aliphatic acid	Valproic acid
Benzodiazepines	Diazepam
Succinimides	Phensuxmide, Ethosuxmide

- **Second generation antiepileptic drugs**

Table 1.2: Shows the second generation antiepileptic drugs.

Category of drug	Examples
Gaba analogs	Vigabatrin, Gabapentin
Carbamates	Felbamate
Triazines	Lamotrigine
Sulfonamides	Zonisamide
Nipecotic acid analogs	Tigabine
Sulfamates	Topiramate
Pyrrolidinone acetamide	Levetiracetam

- **Third generation antiepileptic drugs**

Retigabine, Rufinamide, Losigamone, Stiripentol, Remacemide.

1.5 MECHANISM OF ACTION OF ANTIEPILEPTIC DRUGS[1,3]

Table 1.3: Shows the mechanism of action of antiepileptic drugs.

S. No.	Drugs	Mechanism of action
1.	Phenobarbitone	It reduces neuronal discharge by inhibition of sodium and calcium conductance and also by depressing glutamate induced neuronal depolarization through AMPA receptors. It also enhances GABA (gamma amino butyric acid) effect on chloride channel.
2.	Phenytoin	It blocks high-frequency repetitive firing of action potential (post titanic potentiation) by blocking sodium channel. It also facilitates GABA activity and stabilizes neuronal membrane.
3.	Carbamezepine	Same as phenytoin.
4.	Valproic acid	It increases level of GABA by increasing its synthesis (stimulate glutamic acid decarboxylase) and by reducing its metabolism by inhibiting GABA-T.
5.	Phensuxmide	It reduces calcium conductance, inhibit sodium and potassium ATPase and GABA-T.
6.	Vigabatrine	It is irreversible inhibitor of GABA-T. It also potentates GABA

		action by inhibiting GABA transporter.
7.	Felbamate	It is believed to act by blocking NMDA receptor via glycine binding site.
8.	Zonisamide	It acts mainly on voltage gated sodium and calcium channel.

The basic mechanism of neuronal excitability is the action potential, a hyperexcitable state can result from increased excitatory synaptic neurotransmission, decreased inhibitory neurotransmission, an alteration in voltage-gated ion channels, or an alteration of intra- or extra-cellular ion concentrations in favor of membrane depolarization. A hyperexcitable state can also result when several synchronous subthreshold excitatory stimuli occur, allowing their temporal summation in the post synaptic neurons.

Action potentials occur due to depolarization of the neuronal membrane, with membrane depolarization propagating down the axon to induce neurotransmitter release at the axon terminal. The action potential occurs in an all-or-none fashion as a result of local changes in membrane potential brought about by net positive inward ion fluxes. Membrane potential thus varies with activation of ligand-gated channels, whose conductance is affected by binding to neurotransmitters; or with activation of voltage-gated channels, whose conductance is affected by changes in transmembrane potential; or with changes in intracellular ion compartmentalization.

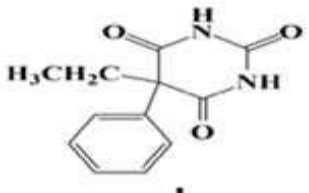
Neurotransmitters are substances that are released by the presynaptic nerve terminal at a synapse and subsequently bind to specific postsynaptic receptors for that ligand. Ligand binding results in channel activation and passage of ions into or out of the cells. The major neurotransmitters in the brain are glutamate, gamma-amino-butyric acid (GABA), acetylcholine (ACh), norepinephrine, dopamine, serotonin, and histamine. Other molecules, such as neuropeptides and hormones, play modulatory roles that modify neurotransmission over longer time periods.

The major excitatory neurotransmitter is the amino acid glutamate. There are several subtypes of glutamate receptors. Glutamate receptors can be found postsynaptically on excitatory principal cells as well as on inhibitory interneurons, and have been demonstrated on certain types of glial cells. The ionotropic subclasses are the alpha-amino-2,3-dihydro-5-methyl-3-oxo-4- isoxazolepropanoic acid (AMPA), kainate receptors, and N-methyl-D-aspartate (NMDA); these allow ion influx upon activation by glutamate). They are differentiated from one another by cation permeability as well as differential sensitivity to pharmacological agonists/antagonists. All ionotropic glutamate receptors are permeable to Na^+ and K^+, and it is the influx of Na^+ and outflow of K^+ through these channels that contribute to membrane depolarization and generation of the action potential. The NMDA receptor also has a Ca^{++} channel that is blocked by Mg^{++} ions in the resting state, but under conditions of local membrane depolarization, Mg^{++} is displaced and the channel becomes permeable to Ca^{++}; influx of Ca^{++} tends to further depolarize the cell, and is thought also to contribute to Ca^{++} mediated neuronal injury under conditions of excessive neuronal activation (such as status epilepticus and ischemia), potentially leading to cell death, a process termed excitotoxicity. The other major type of glutamate receptor is the metabotropic receptor, which functions by means of receptor-activated signal transduction involving membrane-associated G-proteins. There are at least 3 subtypes of metabotropic receptors, based on differential agonist potency, mechanism of signal transduction, and pre- versus post-synaptic localization.

The major inhibitory neurotransmitter, GABA, interacts with 2 major subtypes of receptor: $GABA_A$ and $GABA_B$ receptors. $GABA_A$ receptors are found postsynaptically, while $GABA_B$ receptors are found presynaptically, and can thereby modulate synaptic release. In the adult brain, $GABA_A$ receptors are permeable to Cl^- ions; upon activation Cl^- influx hyperpolarizes the membrane and inhibits action potentials. Therefore, substances which are $GABA_A$ receptor agonists, such as barbiturates and benzodiazepines, are well known to suppress seizure activity. $GABA_B$ receptors are associated with second messenger systems. Rather than Cl^- channels, and lead to attenuation of transmitter release due to their presynaptic location. The second messenger systems often result in opening of K^+ channels, leading to a hyperpolarizing current. Certain $GABA_B$ agonists, such as baclofen, have been reported to exacerbate hyper excitability and seizures.

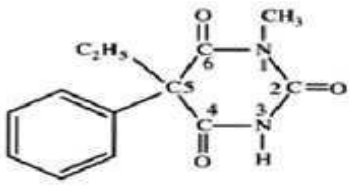
1.6. STRUCTURE OF ANTICONVULSANT DRUGS[1, 3, 4]

1. Phenobarbitone.



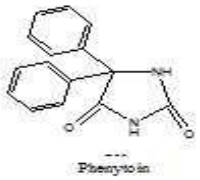
Phenobarbital

2. Mephobarbital

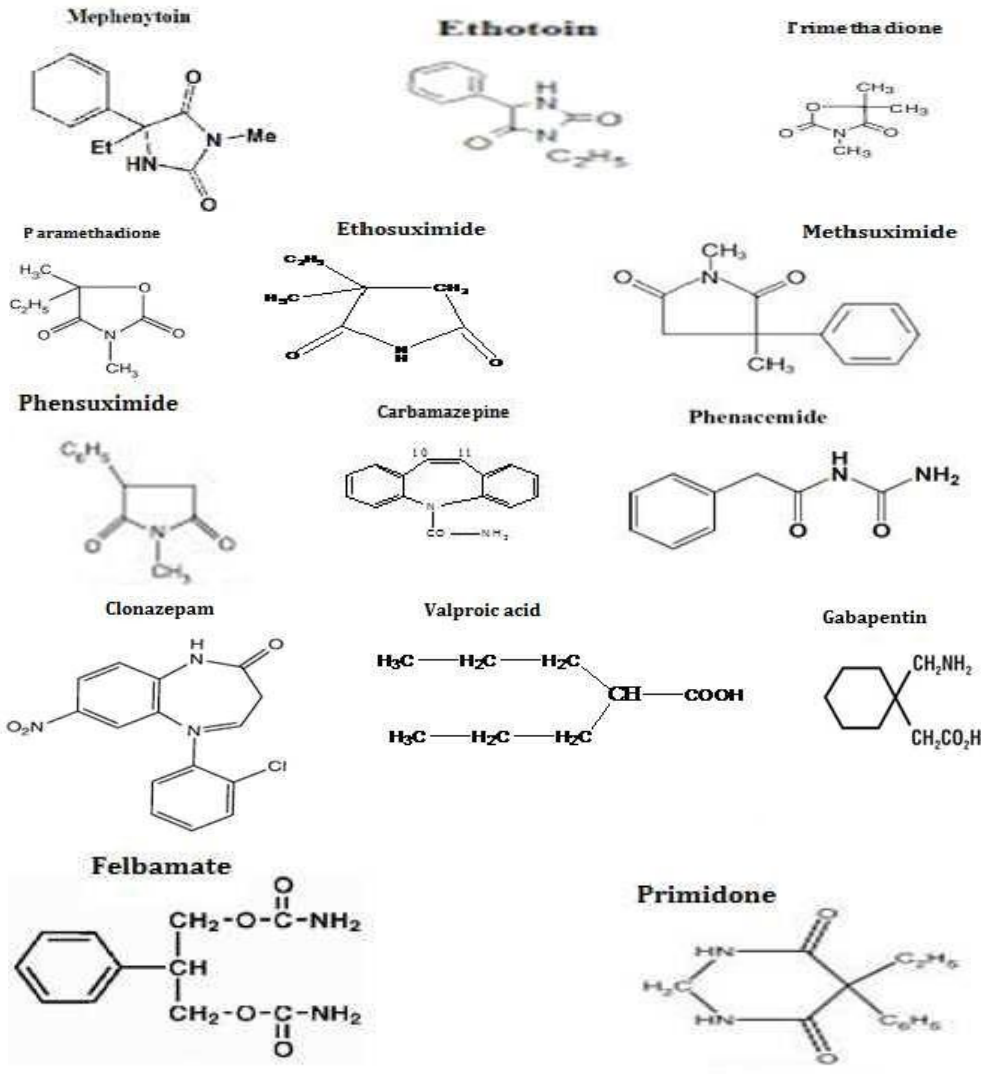


Mephobarbital

3. Phenytoin:



Phenytoin

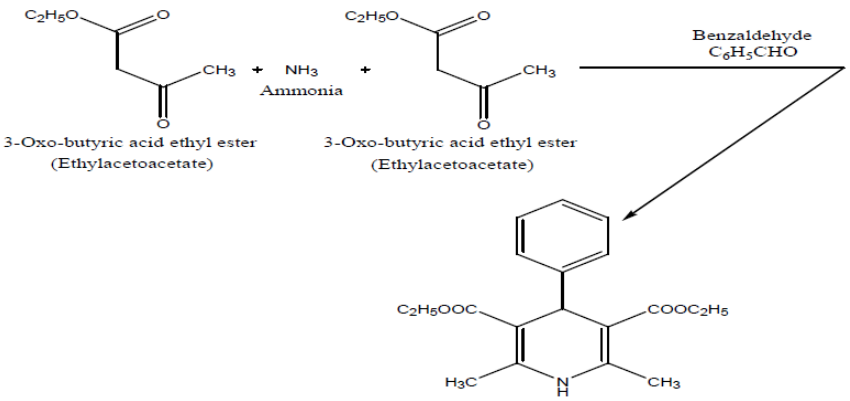


EXPERIMENTAL SECTION

1. Synthetic Scheme Product – I (P-01) formation

Step-1: Intermediate (IM-01) Product formation: Synthesis of 3,5-diethoxy carbonyl-1,4-dihydro-2,6-dimethyl- 4-phenyl-pyridine (IM-01)

Reaction



Procedure: In a 250-mL RBF, solution of benzaldehyde (0.2 mol), ethyl acetoacetate (0.2 mol), ammonium hydroxide (10 mL) in ethanol (60 mL) was taken and refluxed for 3 hours on water bath. To the resulting mixture, warm water (40 mL) was added and then allowed to cool. The separated product was filtered off, washed with 60% aqueous ethanol (10 mL) and recrystallized from ethanol.

Melting Point Range: 172-175^{0}C Percentage Yield: 47.56%

TLC Profile
Stationary Phase: Silica Gel
Mobile Phase: Toluene : Methanol :Ammonia :: 7.5 :2.0 : 0.5 Rf: 0.79

RESULTS AND DISCUSSION

The IR spectrum was scanned in the range of 400 to 4000 cm^{-1} and various peaks of different functional groups were observed. The IR spectral data of compound (IM-01) is given in following table while the IR spectrum is shown in Figure.

Table 1.4: Characteristic infrared absorptions of functional groups of compound (IM-01).

S.No.	Functional group	IR Absorbance (cm^{-1}) (Theoretical)	IR Absorbance (cm^{-1}) (Observed)	Inferences
1.	N-H str	3360-3330	3342.4	Indicate sec. amine
2.	Mono substituted benzene	770-730	749.3	Indicate free phenyl group
3.	CH_3 str	2982-2953	2981.7	Indicate methyl group
4.	C(=O)-O str	1230-1205	1211.2	Indicate ester group
5.	C=C str	1650-1450	1440.7	Indicate aromatic ring

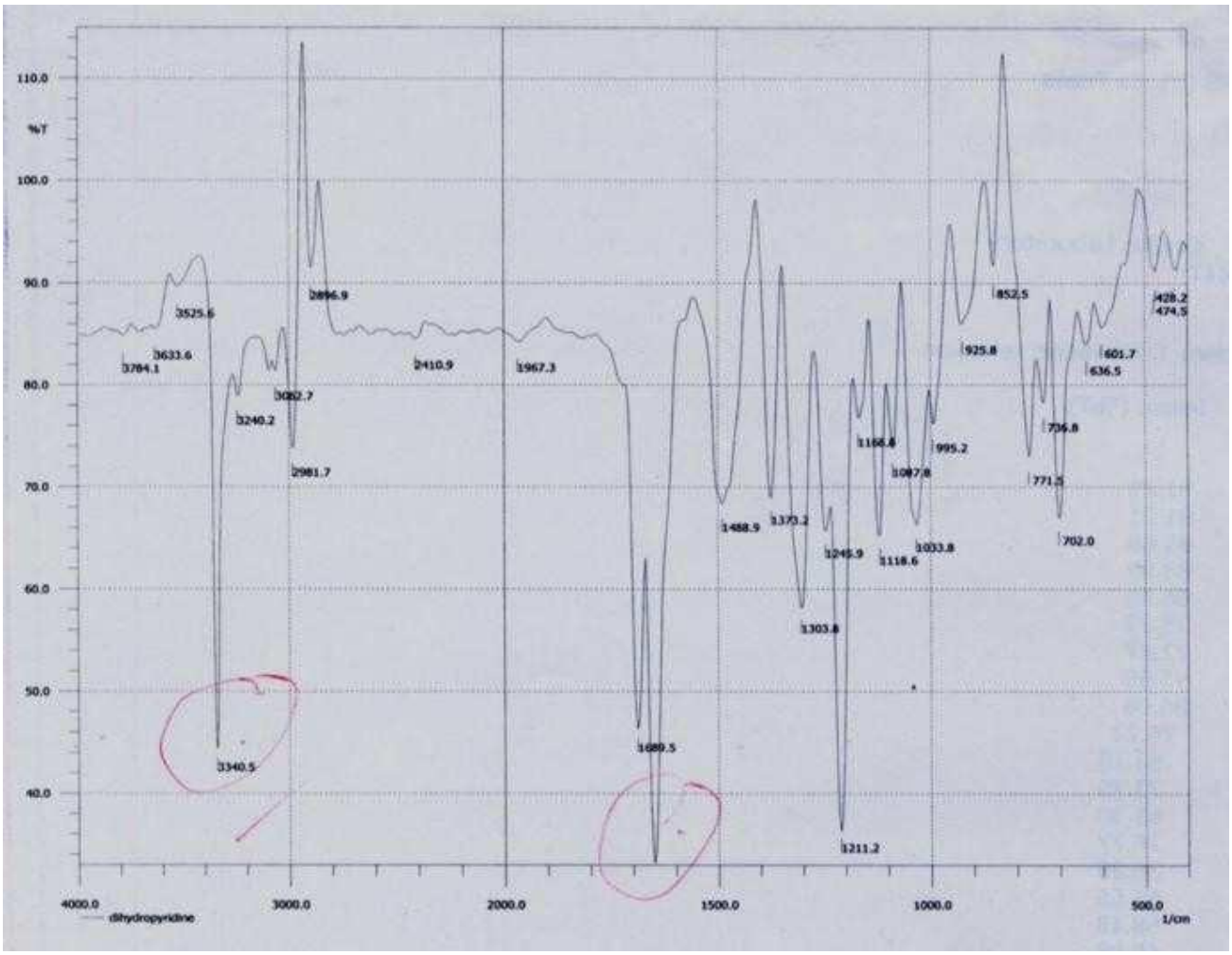
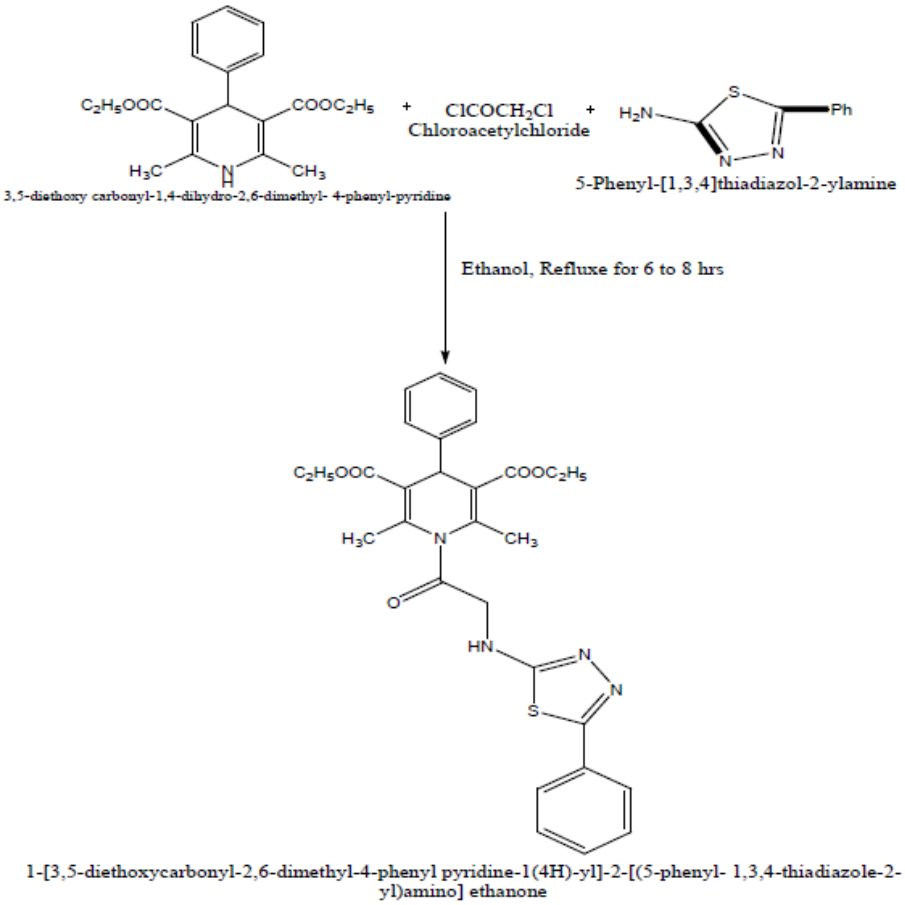


Fig. 1.1: Infrared absorptions of functional groups of compound (IM-01).

Step-2: Final Product (P - 01) formation: Synthesis of 1-[3,5-diethoxycarbonyl-2,6-dimethyl-4-phenyl pyridine-1(4H)-yl]-2-[(5-phenyl-1,3,4 thiadiazole-2-yl)amino] ethanone (P-01).

Reaction



Procedure: In a 250 mL RBF, 3,5-diethoxy carbonyl-1,4-dihydro-2,6-dimethyl-4-phenyl- pyridine(0.005 mol, 1.64 gm) and chloroacetyl chloride (0.005 mol, 0.56 mL) was taken and kept overnight at room temperature. Then 2-amino-5-phenyl- thiadiazole (0.005 mol, 0.88 gm) in ethanol (12.5 mL) was added to the reaction mixture. The reaction mixture was refluxed for 6 hour. Then it was cooled and poured onto crushed ice with continuous stirring. The solid thus obtained was filtered, washed with cold water, dried and recrystallized from aqueous ethanol to yield 1-[3,5-diethoxycarbonyl-2,6-dimethyl-4-phenylpyridine- 1(4H)-yl]-2-[(5- phenyl-1,3,4-thiadi - azole-2-yl)amino]ethanone.

Melting Point Range: 163-166^{0}C Percentage Yield: 54.77%

TLC Profile

Stationary Phase: Silica Gel

Mobile Phase: Toluene : Methanol :Ammonia :: 7.5 : 2.0 : 0.5 Rf: 0.76

Results and Discussion:

The IR spectrum was scanned in the range of 400 to 4000 cm^{-1} and various peaks of different functional groups were observed. The IR spectral data of compound (P-01) is given in table while the IR spectrum is shown in figure.

Table 1.5: Characteristic infrared absorptions of functional groups of Compound (P-01).

S.No.	Functional group	IR Absorbance (cm^{-1}) (Theoretical)	IR Absorbance (cm^{-1}) (Observed)	Inferences
1.	C(=0)str.(tert amide)	1670-1630	1650.95	Indicate tert amide
2.	Mono substituted benzene	770-730	737.7	Indicate free phenyl group
3.	CH_3 str	2990-2953	2980.8	Indicate methyl group
4.	C(=O)-O str	1230-1205	1211.2	Indicate ester group
6.	N-H str	3360-3330	3342.4	Indicate sec amine
7.	C=N str	1600-1430	1488.94	Indicate nitro group
8.	C-S str	1120-1027	1090.67	Indicate C=S group

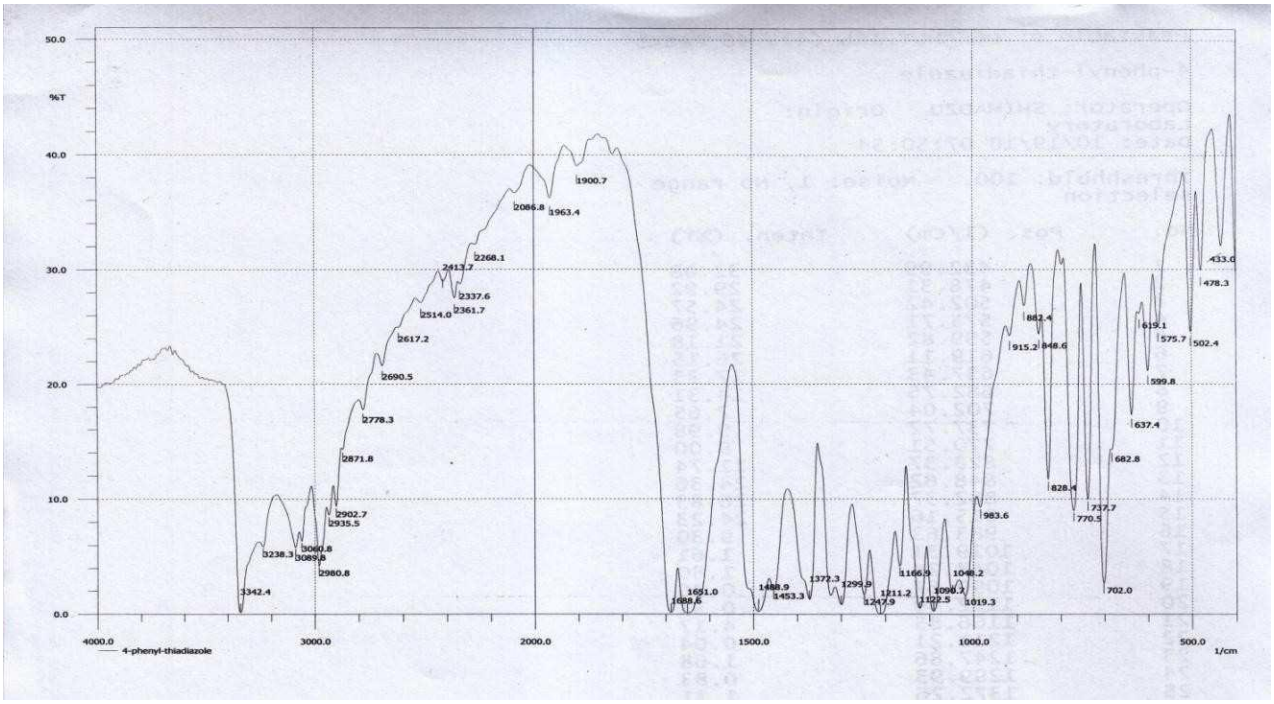
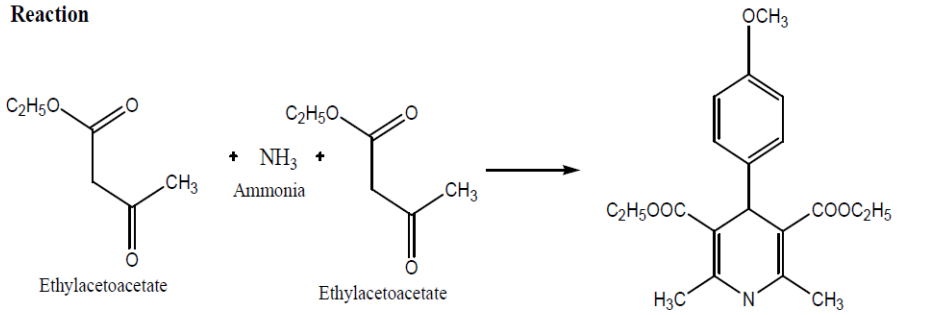


Fig. 1.2: Infrared absorptions of functional groups of compound (P-01) Product – II (P-02) formation.

Step-1: Intermediate (IM-02) Product formation: Synthesis of 5-diethoxy carbonyl-1, 4-dihydro-2,6-dimethyl- 4-(p-methoxy phenyl) pyridine (IM-02)

Reaction



Procedure: In a 250-mL RBF, solution of anisaldehyde (0.2 mol), ethyl acetoacetate (0.2 mol), ammonium hydroxide (10 mL) in ethanol (60 mL) was taken and refluxed for 3 hours on water bath. To the resulting mixture, warm water (40 mL) was added and then allowed to cool. The separated product was filtered off, washed with 60% aqueous ethanol (10 mL) and recrystallized from ethanol.

Melting Point Range: 174-177^{0}C
Percentage Yield: 55.13%

TLC Profile
Stationary Phase: Silica Gel
Mobile Phase: Toluene: Methanol: Ammonia: 7.5 : 2.0 : 0.5
Rf: 0.68

RESULTS AND DISCUSSION

The IR spectrum was scanned in the range of 400 to 4000 cm^{-1} and observed various peaks of different functional groups. The IR spectral data of compound (IM-02) is given in table while the IR spectrum is shown in figure.

Table 1.6: Characteristic infrared absorptions of functional groups of compound (IM-02).

S.No.	Functional group	IR Absorbance (cm-1) (Theoretical)	IR Absorbance (cm-1) (Observed)	Inferences
1.	N-H str	3360-3330	3342.4	Indicate sec. amine
2.	C-O-C str	1310-1240	1253.64	Indicate methoxy group
3.	CH3 str	2982-2953	2981.7	Indicate methyl group
4.	C(=O)-O str	1230-1205	1211.2	Indicate ester group
5.	C=C str	1650-1450	1440.7	Indicate aromatic ring

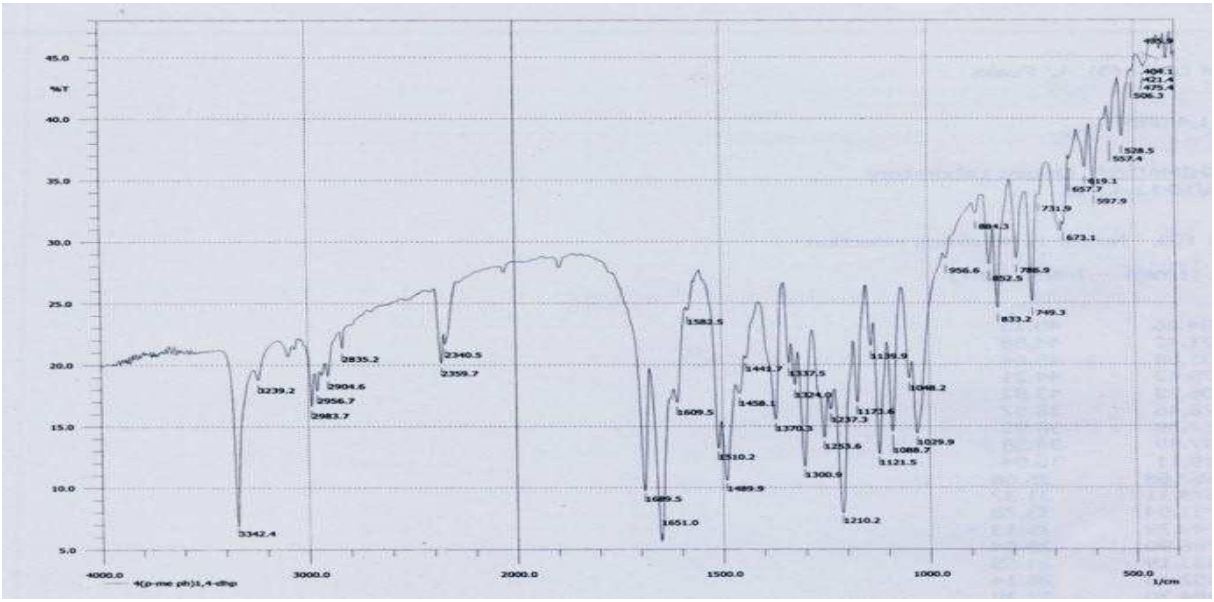
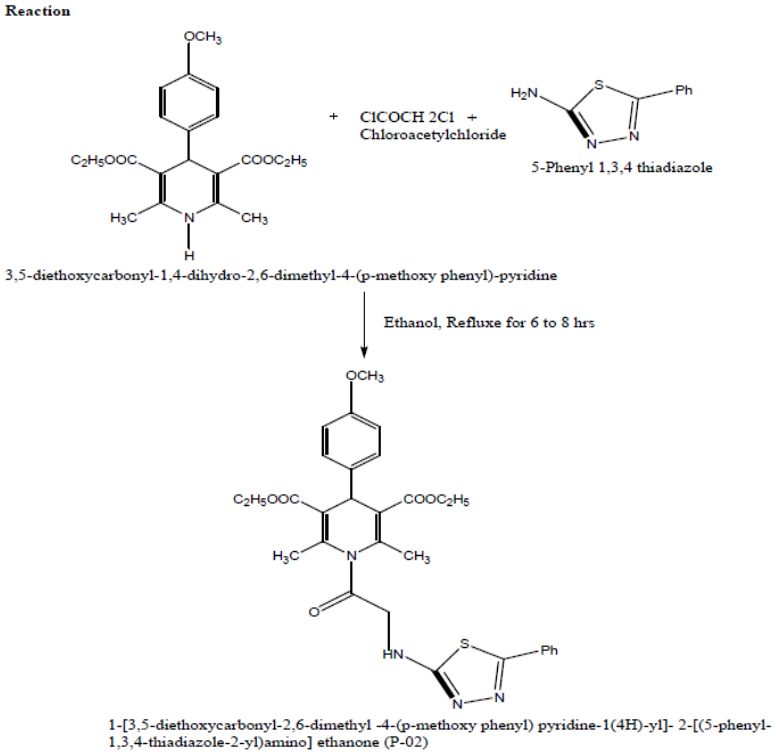


Fig. 1.3: Infrared absorptions of functional groups of compound (IM-02).

Step-2: Final Product (P - 02) formation: Synthesis of 1-[3,5-diethoxycarbonyl-2,6- dimethyl-4-(p-methoxy phenyl)pyridine- 1(4H)-yl]-2 – [(5-phenyl-1,3,4-thiadiazole-2-yl) amino] ethanone (P-02).



Procedure: In a 250 mL RBF, 3,5-diethoxy carbonyl-1,4-dihydro-2,6-dimethyl-4-(p-methoxy phenyl) – pyridine (0.005 mol, 1.79 gm) and chloroacetyl chloride (0.005 mol ,0.56 mL) was taken and kept overnight at room temperature. Then 2-amino-5- phenyl-thiadiazole (0.005 mol,0.88 gm) in ethanol (12.5 mL) was added to the reaction mixture. The reaction mixture was refluxed for 6 hour. Then it was cooled and poured onto crushed ice with continuous stirring. The solid thus obtained was filtered, washed with cold water, dried and recrystallized from aqueous ethanolto yield 1-[3,5-diethox carbonyl-2,6- dimethyl-4-(p- methoxyphenyl)-pyridine-1(4H)-yl]-2-[(5-phenyl-1,3,4-thiadiazole-2-yl)amino] ethanone.

Melting Point Range: 179-183^{0}C

Percentage Yield: 58.67%

TLC Profile:
Stationary Phase: Silica Gel
Mobile Phase: Toluene: Methanol: Ammonia: 7.5 :2.0 : 0.5
Rf: 0.74

Results and Discussion: The IR spectrum was scanned in the range of 400 to 4000 cm^{-1} and observed various peaks of different functional groups. The IR spectral data of compound (P-02) is given in table while the IR spectrum is shown in figure.

Table 1.7: Characteristic infrared absorptions of functional groups of compound (P-02).

S.No.	Functional group	IR Absorbance (cm^{-1}) (Theoretical)	IR Absorbance (cm^{-1}) (Observed)	Inferences
1.	C(=0)str.(tert amide)	1670-1630	1649.9	Indicate tert amide
2.	C-O-C str	1310-1240	1253.64	Indicate methoxy group
3.	CH_3 str	2982-2953	2957.64	Indicate methyl group
4.	C(=O)-O str	1230-1205	1210.5	Indicate ester group
5.	C=C str	1650-1450	1511.2	Indicate aromatic ring
6.	N-H str	3360-3330	3342.4	Indicate sec amine
7.	C-S str	1120-1027	1120.56	Indicate C=S group

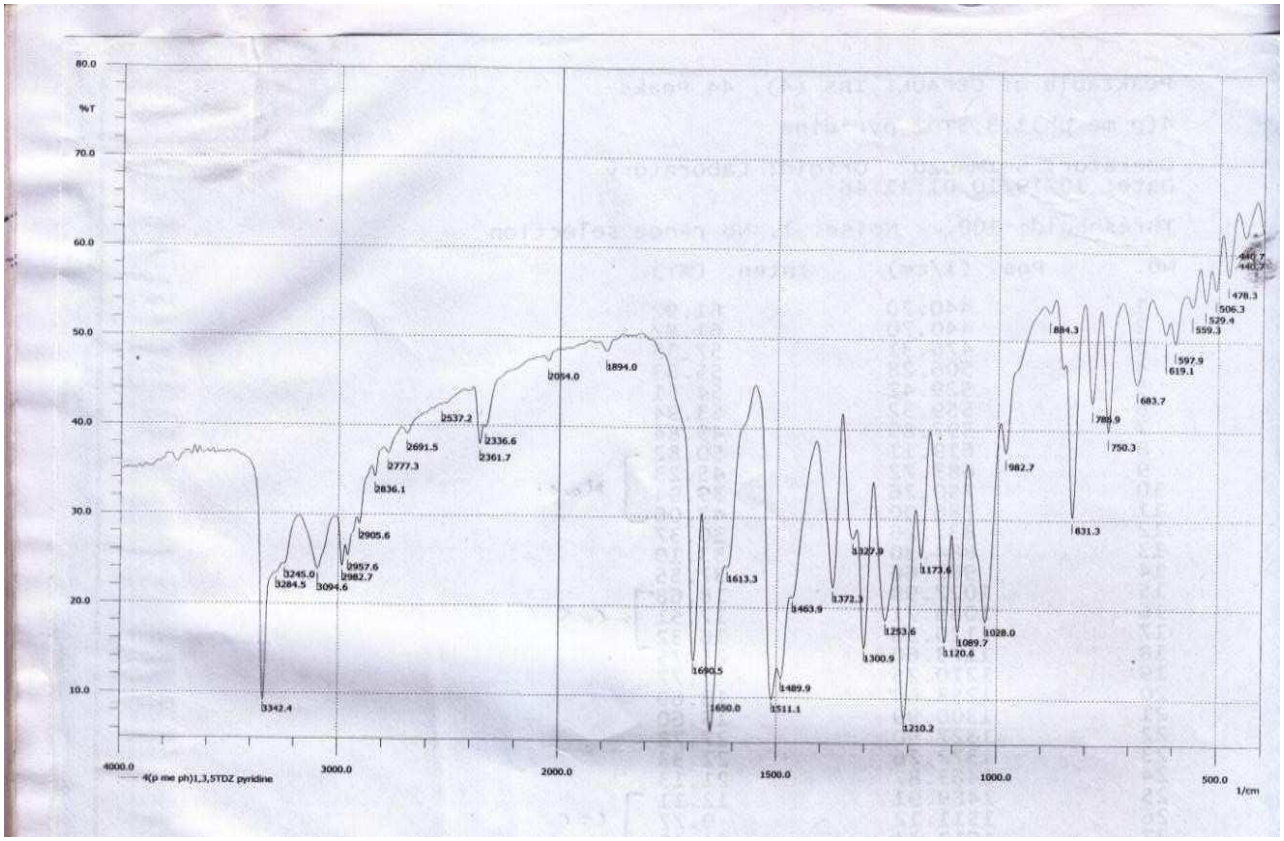
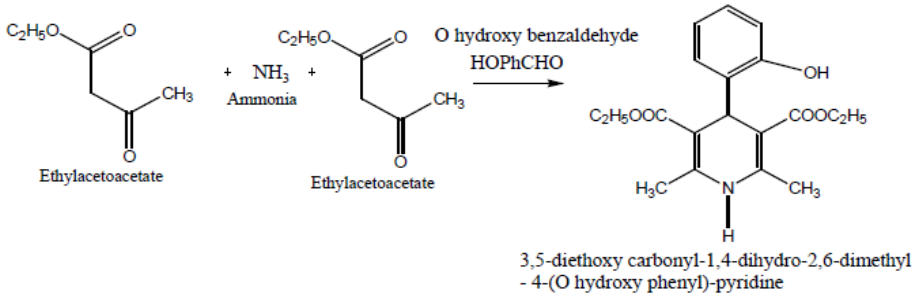


Fig. 1.4: Infrared absorptions of functional groups of compound (P-02).

Product – III (P-03) formation
Step-1: Intermediate (IM-03) Product formation: Synthesis of 3, 5-diethoxy carbonyl-1,4-dihydro-2,6-dimethyl- 4-(O-hydroxy phenyl)-pyridine (IM 03)

Reaction



Procedure: In a 250-mL RBF, solution of salicyldehyde (0.2 mol), ethyl acetoacetate (0.2 mol), ammonium hydroxide (10 mL) in ethanol (60 mL) was taken and refluxed for 3 hours on water bath. To the resulting mixture, warm water (40 mL) was added and then allowed to cool. The separated product was filtered off, washed with 60% aqueous ethanol (10 mL) and recrystallized from ethanol.
Melting Point Range: 125-127^{0}C Percentage Yield: 60.69%

TLC Profile:
Stationary Phase: Silica Gel
Mobile Phase: Toluene : Methanol :Ammonia :: 7.5 :2.0 : 0.5 Rf: 0.72

Results and Discussion: The IR spectrum was scanned in the range of 400 to 4000 cm^{-1} and observed various peaks of different functional groups. The IR spectral data of compound (IM-03) is given in table while the IR spectrum is shown in figure.

Table 1.8: Characteristic infrared absorptions of functional groups of Compound (IM- 03)

S.No.	Functional group	IR Absorbance (cm-1) (Theoretical)	IR Absorbance (cm-1) (Observed)	Inferences
1.	N-H str	3360-3330	3342.4	Indicate sec. amine
2.	CH3 str	2982-2953	2981.7	Indicate methyl group
3.	C(=O)-O str	1230-1205	1211.2	Indicate ester group
4.	C=C str	1650-1450	1440.7	Indicate aromatic ring

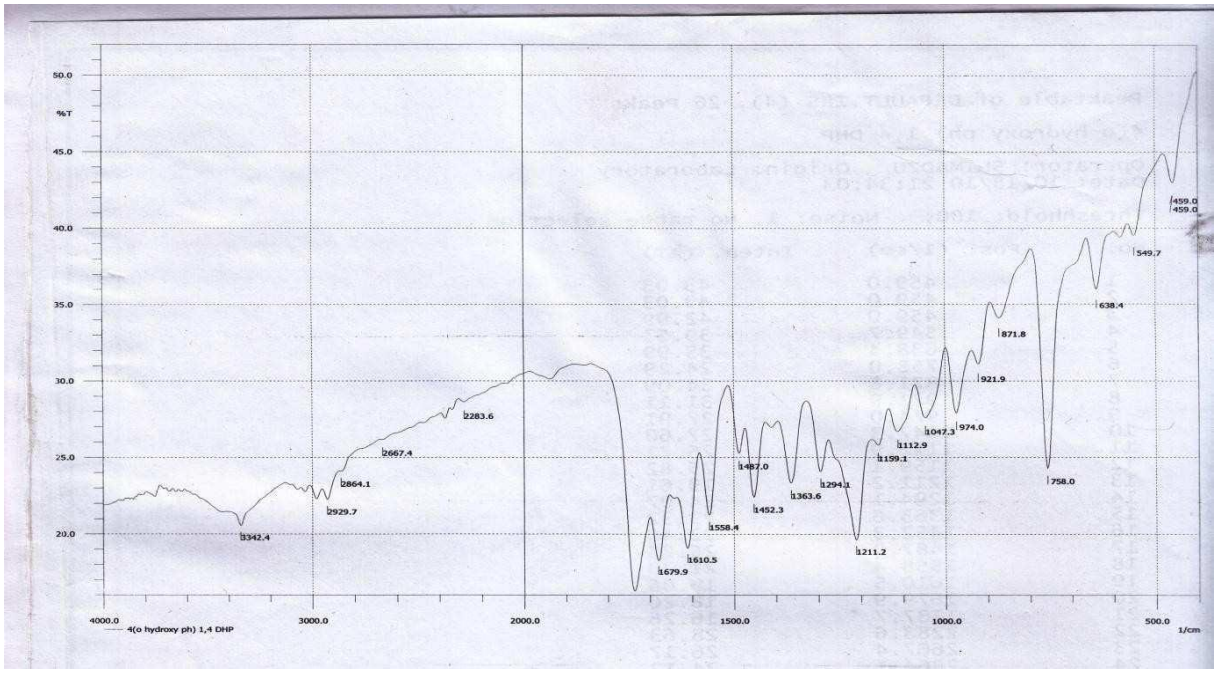
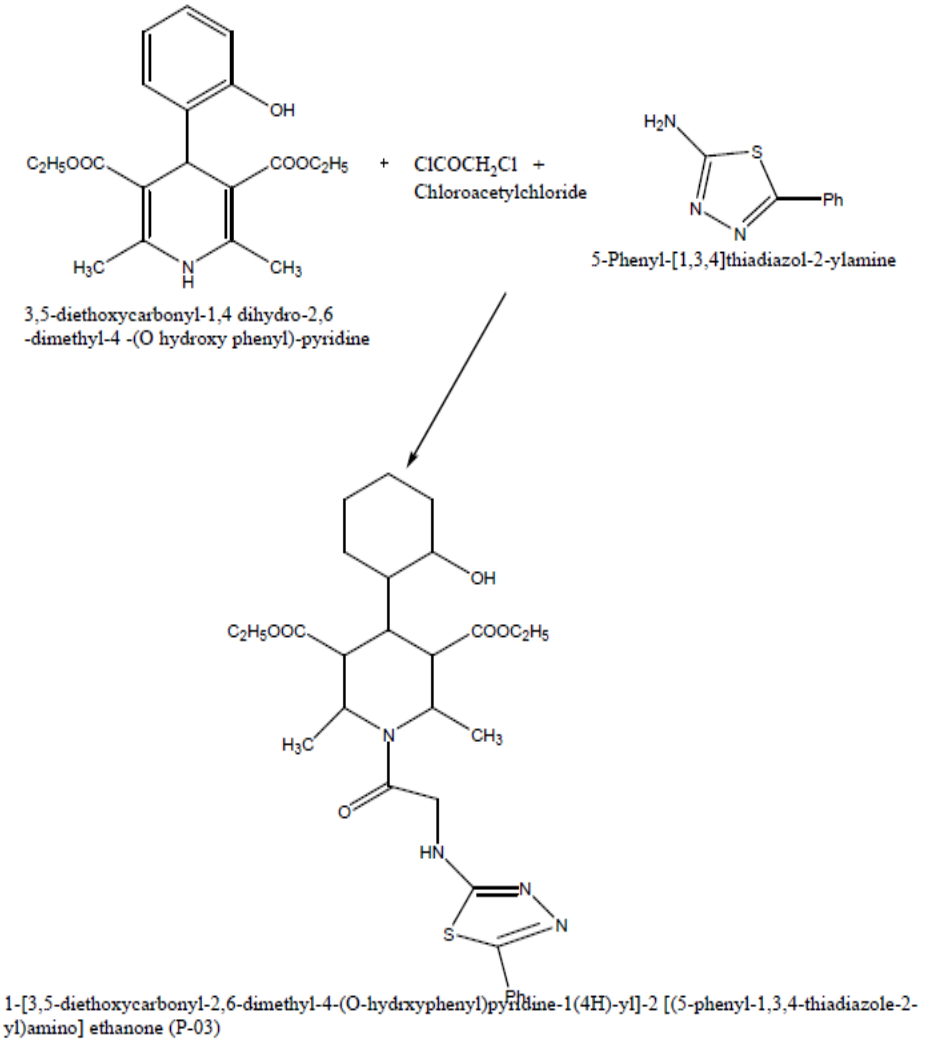


Fig.1.5. Infrared absorptions of functional groups of compound (IM-03).

Step-2: Final Product (P - 02) formation: Synthesis of 1-[3,5-diethoxycarbonyl-2,6-dimethyl- 4-(O-hydroxy phenyl) pyridine- 1(4H)-yl]-2 – [(5-phenyl-1,3,4-thiadiazole-2-yl) amino] ethanone (P-03)



Procedure:- In a 250 mL RBF, 3,5-diethoxy carbonyl-1,4-dihydro-2,6-dimethyl-4-(O-hydroxy phenyl) – pyridine (0.005 mol, 1.74 gm) and chloroacetyl chloride (0.005 mol, 0.56 mL) was taken and kept overnight at room temperature. Then 2-amino-5- phenyl-thiadiazole (0.005 mol, 0.88 gm) in ethanol (12.5 mL) was added to the reaction mixture. The reaction mixture was refluxed for 6 hour. Then it was cooled and pouredonto crushed ice with continuous stirring. The solid thus obtained was filtered, washed with cold water, dried and recrystallized from aqueous ethanol to yield 1-[3,5-diethoxycarbony l-2,6-dimethyl- 4-(p- methoxyphenyl) pyridine-1(4H)-yl]-2-[(5-phenyl-1,3,4-thiadiazole-2-yl) amino]ethanone.

Melting Point Range: 155-158^{0}C Percentage Yield: 55.33%

TLC Profile:

Stationary Phase: Silica Gel

Mobile Phase: Toluene : Methanol :Ammonia :: 7.5 :2.0 : 0.5 Rf: 0.75

Results and Discussion: The IR spectrum was scanned in the range of 400 to 4000 cm^{-1} and observed various peaks of different functional groups. The IR spectral data of compound (P-03) is given in table while the IR spectrum is shown in figure.

Table 1.9: Characteristic infrared absorptions of functional groups of Compound (P-03).

S.No.	Functional group	IR Absorbance (cm^{-1}) (Theoretical)	IR Absorbance (cm^{-1}) (Observed)	Inferences
1.	C(=0)str. (tert amide)	1670-1630	1633.6	Indicate tert amide
2.	O-H str	3400-3000	3093	Indicate hydroxyl group
3.	CH_3 str	2982-2953	2964	Indicate methyl group
4.	C(=O)-O str	1230-1205	1209.28	Indicate ester group
5.	C=N str	1600-1430	1512.09	Indicate nitro group
6.	N-H str	3360-3330	3273	Indicate sec amine
7.	C-S str	1120-1027	1089.79	Indicate C=S group

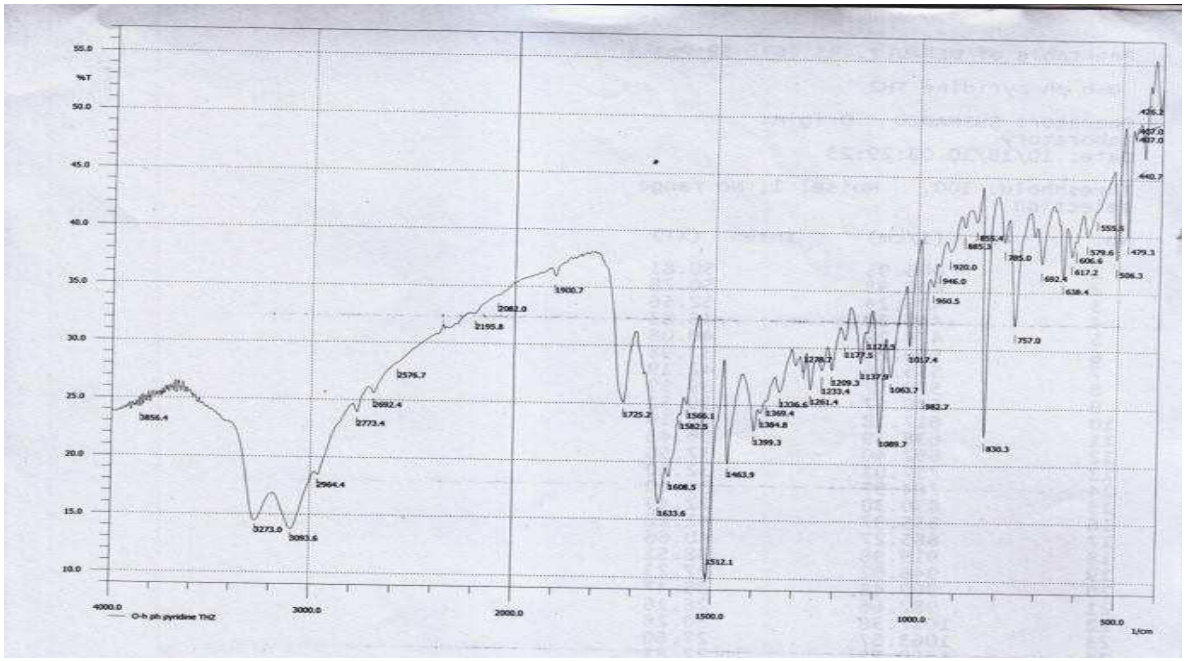


Fig. 1.6: Infrared absorptions of functional groups of compound (P-03).

5.4 Biological Studies

5.3.1 :- Introduction: A type of drug that is used to prevent or treat seizures or convulsions by controlling abnormal electrical activity in the brain. Anticonvulsants are used to treat epilepsy and other seizure disorders. They are also used to treat medical conditions, such as bipolar disorder, nerve pain, migraine headaches, fibromyalgia, and restless leg syndrome. There are many different types of anticonvulsants. Also called anti-seizure medication and antiepileptic.

The anticonvulsant activities of these compounds were evaluated following two methods as listed below:

1. *Maximal Electroshock Seizure (MES) test method*
2. *Subcutaneous pentylenetetrazole (scPTZ) seizure models in mice.*

5.3.2 :- Methods

1. Maximal Electroshock Seizure Test (MES): In the MES test, an electrical stimulus of sufficient intensity (25 mA, 500 V, 50 Hz, 0.2 s) was delivered via auricular electrodes by the electroshock generator (Rodent Shocker, Type 221, Hugo Sachs, March- Hugstetten, Germany) to induce maximal seizures. The endpoint was the tonic extension of the hind limbs.

Animals: Male CD-1 mice weighing 20–26 g were used in the in vivo experiment. The animals were housed in an environmentally controlled room (temperature of 22 ± 2 °C, humidity 55 ± 10%) on 12 h light/dark cycles (light on at 7:00 AM and off at 7:00 PM) and had free access to food (standard laboratory pellets) and water. The experimental groups consisted of 4–6 mice (anticonvulsant studies).

The experiments were performed between 8:00 a.m. and 3:00 p.m. For the experiments, the animals were selected in a random way and trained observers performed all measurements.

Procedure: *Maximal Electroshock Seizure (MES) test method:* The synthesized compounds in the present investigation have been tested for anticonvulsant activity by *"Maximal Electroshock Seizure (MES) test method*. The animals selected for anticonvulsant activity were mice. Anticonvulsant screening of all final compounds was performed initially in the MES and the 6 Hz (32mA) seizure tests at a fixed dose of 100 mg/kg, 0.5 h after intraperitoneal (i.p.) injection. Each screening group consisted of four animals. All compounds were evaluated as free bases. The preliminary pharmacological results showed that compounds P-1, P-2 and P-3 revealed in general weak anticonvulsant activity in the MES test, protecting at least 25% of mice (Table 5.10). The highest and significant anticonvulsant activity in this test was displayed by P-3 compound, which provides 100% protection (4/4) of animals from seizures. In general, more potent activity was observed in the 6 Hz test (32 mA), as three compounds showed anticonvulsant activity—namely, P-1, P-2 and P-3. Notably, half of them (P-1 and P-2) exhibited significant (at least of 50%) anticonvulsant protection. The highest activity, similar to MES seizures, revealed compound P-3, which protected 75% (3/4) of tested animals.

Table 1.10: Anticonvulsant activity (MES and 6 Hz tests) following i.p. administration of dose 100 mg/kg in mice.

Compound	Intraperitoneal Administration in Mice		
	MES [a]	6Hz [b]	
P-1	1/4	1/4	
P-2	0/4	2/4	
P-3	4/4		3/4

Ratios where at least two animals were protected or with motor impairment have been highlighted in bold for easier data interpretation. Data indicate number of mice protected or with motor impairment/number of mice tested. The animals were examined 0.5 h after compound administration.

RESULTS AND DISCUSSION

The final compounds P-1to P-3 were synthesized according to the procedure depicted in Scheme 5.3.1 to 5.3.6. From the starting materials an intermediate product IM-01 to IM-03 is prepared and from intermediate product in the next final step main product P-01 to P-03 is prepared. The purity and homogeneity of all compounds were confirmed by their sharp melting point and TLC. Melting Points were determined in open capillary method and are corrected. The chemical structures were confirmed by spectral analyses (infrared spectroscopy). IR spectra were recorded on Perkinelmer FTIR- spectrophotometer using KBr disc method. All the above result positively confirmed the formation of the synthesized compounds and hence correctness of the anticipated structures drawn for synthesized compounds. All the synthesized compounds have been tested for anticonvulsant activity. The compounds showed mild to good anticonvulsant activity.

SUMMARY AND CONCLUSION

This research work comprises of the synthesis of three 1,4-dihydropyridine derivatives. All derivatives are substituted at 1 and 4 position of basic nucleus other, possessing anticonvulsant activity. Therefore the present work accesses the potential of both the nucleus with different substitutions for better anticonvulsant activity. Hence, the final product synthesized is mentioned below.

I. 1-[3,5-diethoxy carbonyl-2,6-dimethyl-4-phenyl pyridine- 1(4H)-yl]-2-[(5- phenyl-1,3,4-thiadiazole-2-yl)amino]ethanone (P-01)
II. 1-[3,5-diethoxycarbonyl-2,6-dimethyl-4-(p-methoxy phenyl)pyridine- 1(4H)-yl]-2 -[(5-phenyl-1,3,4-thiadiazole-2-yl) amino] ethanone (P-02)
III. 1-[3,5-diethoxycarbonyl-2,6-dimethyl-4-(O-hydroxy phenyl) pyridine- 1(4H)-yl]-2 -[(5-phenyl-1,3,4-thiadiazole-2-yl) amino] ethanone (P-03):-

All the compounds synthesized were identified on the basis of melting point, TLC and IR spectra analysis. IR spectral data confirmed the identity of synthesized compounds. The summary of observed values is given in Table.

Table No.1.11: Physical Constant of Different Synthesized Compounds.

S.No.	Compound code & M. Formula	Mol.Wt.	% Yield	M.P. Range (°C)	R_f value	Solvent system (T.L.C)
1.	P-01 $C_{29}H_{30}O_5N_4S$	544	54.77	163-166	0.76	Toluene : methanol : ammonia (75:20:5, v/v/v)
2.	P-02 $C_{30}H_{32}O_6N_4S$	575	58.67	179-183	0.74	Toluene : methanol : ammonia (75:20:5, v/v/v)
3.	P-03 $C_{29}H_{30}O_6N_4S$	561	55.33	155-158	0.75	Toluene : methanol : ammonia (75:20:5, v/v/v)

All the synthesized compounds have been tested for anticonvulsant activity. These compounds showed mild to good anticonvulsant activity. The compound P-03 showed potent anticonvulsant activity.

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REFERENCES

1. Tripathi K.D., "Essential of medical pharmacology", 4th Edition, Jaypee Brothers Medical Publishers (P) Ltd, New Delhi, 2009; 264: 381-392.
2. Abrahm D.J., "Burger's Medicinal Chemistry and Drug Discovery", 6th Edition, 2003; 263- 321.
3. Pandey S.N, "Text Book of Medicinal Chemistry", 2nd Edition, 1999; 15.
4. Rajak H., Deshmukh R., Aggarwal N., Kashaw S., KharyaM.D., Mishra P., "Synthesis and Anticonvulsant Evaluation of 2,5-Disubstituted 1,3,4- Thiadiazoles: Pharmacophore Model Studies" Arch. Pharm. Chem. Life. Sci., 2011; 342: 453-461.
5. Swarnalata G, Prasanti G, Sirisha N. "1,4-dihydropyridines: A multifunctional molecule – A review". International journal of Chem Tech Research, Jan-Mar, 2011; 3(1): 75-89.
6. Pattan R. S. DigheN.S.Chavan P.A." Synthesis 1,4-dihydro pyridine derivatives and their anticonvulsant actiivity" J. Chem. Pharm. Res., 2010; 246-252.
7. Zonouz M. A, SahiranavardN. "Synthesis of 1,4-dihydro pyridine derivatives under aqueous media" E-Journel of chemistry, 2010; 372-376.
8. Pattanayak P. and Sharma R."2-Amino-5-Sulphanyl-1,3,4-thiadiazole derivatives as anticonvulsant agents: Synthesis and evaluation" Indian Journel of Chemistry, 2010; 49B: 1531-1534.
9. Subudhi B. B., Panda P.K, Swain P.S." Synthesis, characterization and anticonvulsant activity evaluation of some 1,4-dihydropyridines and 3,5 substituted) oxycarbonyl-1,4- dihydro-2,6-dimethyl-n- [2-(4-sulfamoyl pheny amino)-acetyl]-4-(substituted)pyridines" Acta Poloniae Pharmaceutica-Drug research, 2009; 147-153.
10. Kotharkar A. Sandeep, Shinde B.S. "Microwave assisted synthesis of 1,4- dihydropyridines"

Ukrainica bioorganica Acta, 2006; 1: 3-6.
11. Shafiee A., Rastakari N., "Anticonvulsant activities of new 1,4- dihydropyridine derivatives containing 4-Nitroimidazolyl substituents" DARU, 2004; 12(2).
12. Dogan H.N., Duran A., Rollas S., Sener G., Uysal M.K, Giilen D., "Synthesis of New 2,5-Disubstituted-1,3,4-Thiadiazoles and Preliminary Evaluation of Anticonvulsant and Antimicrobial ActivitiesBioorg. & Med. Chem., 2002; 10: 2893-2898.
13. Gulerman N.N., Rollas S., Ekinci A.C., "Anticonvulsant Activity of Some Substituted 1,3,4-Thiadiazoles" Acta Pharmaceutica Turcica, 2001; 161: 163.
14. Navidpour L., Shafaroodi H., Miri R., Reza D.A., Shafiee A.: Farmaco, 2004; 54: 261.
15. Ranke J., Molter K., F. Stock, U. Bottin-Weber, J. Poczobutt, J. Hoffmann, B.
16. Ondruschka, J.Filser, B. Jastorff, Ecotoxicol. Environ. Saf., 2004; 58: 396.
17. Jatav V., Mishra P., Kashaw S., Stables J.P., "Synthesis & CNS Depressant Activity of Some Novel 3-(5-Subatituted 1,3,4-Thiadiazole-2-yl)-2-Styryl Quinazoline-4(3H) –Ones" Eur. J. Med. Chem., 2008; 43: 135-141.
18. Stillings M.R., Welbourn A.P., WalyerD. S., "Substituted 1,3,4-thiadiazole with anticonvulsant activity. 2. Aminoalkyl Derivative" J. Med. Chem., 1986; 29: 2280-2284.
19. World Health Organization. Improving Access to Epilepsy Care. Available at: https://www.who.int/mental_health/neurology/epilepsy/en/.Accessed December 20, 2018.
20. Rehman R, Kelly PR, Husain AM, et al. Characteristics of Veterans diagnosed with seizures within Veterans Health Administration. *J Rehabil Res Dev*., 2015; 52(7): 751–62.
21. Kanner AM. Management of psychiatric and neurological comorbidities in epilepsy. *Nat Rev Neurol*, 2016; 12(2): 106–16.
22. Nevalainen O, Ansakorpi H, Simola M, et al. Epilepsy-related clinical characteristics and mortality: a systematic review and meta-analysis. *Neurology*, 2014; 83(21): 1968–77.
23. Thurman DJ, Logroscino G, Beghi E, et al. The burden of premature mortality of epilepsy in high-income countries: A systematic review from the Mortality Task Force of the International League Against Epilepsy. *Epilepsia*, 2017; 58(1).
24. Liporace J, D'Abreu A. Epilepsy and women's health: family planning, bone health, menopause, and menstrual-related seizures. *Mayo Clin Proc*., 2003; 78(4): 497–506.
25. Smaldone M, Sukkarieh T, Reda A, et al. Epilepsy and erectile dysfunction: a review. *Seizure*., 2004; 13(7): 453–9.
26. Morrell MJ. Stigma and epilepsy. *Epilepsy Behav*, 2002; 3(6S2): 21–25.