IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

Human Journals
Research Article
September 2019 Vol.:16, Issue:2

Synthesis, Characterization and Pharmacological Evaluation of Some Novel Diphenyl 1-H-Pyrazole-4-Carbaldehyde Derivatives with Their Anti-Convulsant Activity

IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

***Ruby Tabassum, Vidyakant Kushwaha, Vivek Dwivedi, Shahnawaz Sameem, Sudhakar Singh, Ritesh Tiwari**

Chandra Shekhar Singh College of Pharmacy, Puramufti, Kaushambi India, 212213

Submission: 26 August 2019
Accepted: 31 August 2019
Published: 30 September 2019

HUMAN JOURNALS
www.ijppr.humanjournals.com

Keywords: Vilsmeier-Hack reagent, Pyrazole, Anticonvulsant activity

ABSTRACT

A series of eight new 1, 3-diphenyl 1-H-pyrazole-4-carbaldehyde of pyrazole derivatives have been synthesized and evaluated for anticonvulsant activity by using maximal electroshock method (MES method). Initially, substituted acetophenone was allowed to react with phenylhydrazine in the presence of ethanol to form 2-phenyl-1-(1-phenylethylamine) hydrazine. The compound was further treated with Vilsmeier-Hack reagent ($DMF+POCl_3$) to form 1, 3-diphenyl 1-H-pyrazole-4-carbaldehyde. The final compound was synthesized by the reaction of a compound with isonicotinahydrazide in the presence of 1, 4-dioxane as a solvent. The structure of the final analogs has been confirmed based on elemental analysis, FTIR, ^{1}H NMR&^{13}CNMR. All the values of elemental analysis, FTIR, ^{1}H NMR&^{13}CNMR were found to be prominent. The pharmacological screening by using Maximal Electroshock (MES Model) for anticonvulsant activity. The result of the current study showed that substituted pyrazole derivative bearing (1, 3-substituted phenyl-1H-pyrazole-4-yl-methylene) isoniazid evaluated as a potent anticonvulsant drug.

INTRODUCTION

Pyrazole refers to a class of simple aromatic ring organic compound of heterocyclic diazole series characterized by a 5-member ring structure composed of three carbon atom and two nitrogen atom in the adjacent position and to the unsaturated parent compound. Being so composed and has a pharmacological effect on human, they are classified as an alkaloid, although they are rare in nature.

Pyrazole unsubstituted in 1-position show NH-acidity, the pKa value of pyrazole is 14.21 and equal to that of imidazole. The partially reduced form of pyrazole is named as pyrazoline, basic in nature, these are aromatic molecules due to there planner conjugated ring structure with six delocalized pie electron and the aromatic nature arises from the four-electron and the unshared electron on the –NH.

An intermolecular conjugated charge transfer process has been reported to exist in an excited state. The conjugated part ($-N^1-N^2-C^3$) of the ring, the nitrogen atom at the 3 positions is respectively electron-donating and withdrawing moieties. The carbon atom at 4 and 5 positions do not conjugate with the remaining part of the ring (Jamwal et al., 2013).

Pyrazole derivative is of interest because of their potential biological activity such as antileishmanial (Alodeani et al., 2015), antifungal (Patil et al., 2014), anti-inflammatory(Martin et al., 2013),anticancer (Balbi et al., 2011), antibacterial (Gaikwad et al. , 2013),antitubercular (Kawale et al., 2013), antiproliferative (Maurizio et al., 2013).

Heterocycles are an organic compound containing at least one atom of carbon, and at least one element other than carbon, such as sulfur oxygen or nitrogen within a ring structure (Eicher et al., 2003). Heterocyclic chemistry is the branch of chemistry dealing with the synthesis, properties, and application of heterocycles. The history of heterocyclic chemistry began in the 1800s, in step with the development of organic chemistry (Compaigne et al., 1986).

Epilepsy is one of the more common and frequent neurological disorder in man, characterized by the excessive temporary neuronal discharges resulting in an uncontrolled convulsion that affect more than two million Americans and 60 million people worldwide. If not treated, it is associated with progressively impaired cognition and function, brain damage, and other neurological deficits. Although in many cases, epilepsy can be adequately controlled through

the administration of antiepileptic drugs (AEDs), it is estimated through that roughly 20-30% of patient have seizures that are resistant to available medical therapies. Conventional AEDs like phenobarbital, pyrimidine, phenytoin, carbamazepine, ethosuximide, and benzodiazepine are widely used (Wei et al., 2015).

Heterocyclic form by far the largest of the classical division of organic chemistry and are of immense importance biologically, industrially, and indeed to the functioning of any development human society. For more than a century, heterocyclic has constituted one of the largest areas of research in organic chemistry. Among the approximately 20 million chemical compounds identified the end of the second millennium, more than two-third are fully or partially aromatic (Balaban et al., 2004).

Even through compounds have frequently been detected in the environment, there is no publication about the toxic effects on embryos of danio rerio and the knowledge of their occurrence, the environmental fate, biological metabolism and toxic is limited (Blum et al., 2011; Feldmannova et al., 2006; Bleeker et al., 1998).

Thus, further investigations are needed to evaluate the toxicity of these compounds with a special focus on the development of an aquatic organism. In ecotoxicological testing, fish are an indispensable component of integrated toxicity testing strategies for the aquatic environment (Hote and Bhoyar, 2014).

MATERIALS AND METHODS

Reagents and solvents

The chemicals used for the experimental work were commercially procured from various chemical units from the Sigma Aldrich, HiMedia, Lobachem India Ltd. and CDH. These compounds were purified and dried before their use.

Instrument and Equipment

Melting point- Melting points of the synthesized compounds were determined by the open capillary method.

Solubility- The solubility of the compounds was checked in various solvents at room temperature.

^{1}H NMR –The proton magnetic resonance spectra (^{1}H NMR) were recorded on a Bruker 300 MHz instrument in DMSO/$CDCl_3$using tetramethylsilane as an internal standard. The ^{1}H NMR spectroscopy was done at CDRI, Lucknow.

IR - The infrared spectra of the compounds were recorded in KBr on PERKIN ELMER FTIR spectrometer. The FTIR spectroscopy was performed at CDRI, Lucknow.

TLC - Iodine chamber and U.V lamp were used for visualization of TLC spots.

Synthetic procedure for substituted acetophenone (2a-h) (intermediate compounds

Synthesis of 2-phenyl-1-(1-phenylethylidene) hydrazine (2a)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water then dried.

Synthesis of 1-(1-(4-nitrophenyl)ethylidene)-2-phenylhydrazine (2b)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 4-nitro acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthesis of 1-(1-(3-hydroxyphenyl)ethylidene)-2-phenylhydrazine (2c)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 3-hydroxy acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthesis of 2-phenyl-1-(1-*p*-tolyethylidene) hydrazine (2d)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 4-methyl acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthesis of 1-(1-(3,4-dimethoxyphenyl) ethylidene)-2-phenylhydrazine (2e)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 3,4-dimethoxy acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthesis of 1-(1-(4-dimethoxyphenyl)ethylidene)-2-phenylhydrazine (2f)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 4-dimethoxy acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthesis of 1-(1-(2-bromophenyl)ethylidene)-2-phenylhydrazine (2g)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 2-bromo acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthesis of 1-(1-(4-aminophenyl) ethylidene)-2-phenylhydrazine (2h)

Phenylhydrazine (0.01mol) was added in glacial acetic acid (10ml) and water (10ml) to a solution of 4-amino acetophenone (0.01mol) in glacial acetic acid (20ml) contained in a beaker. After the completion of the reaction was checked by thin-layer chromatography. The

mixture was cooled in ice and shaken for 5min, colorless crystals obtained and filtered, washed with dilute acetic acid and water, dried.

Synthetic procedure for substituted 1, 3-diphenyl 1*H*-pyrazole 4-carbaldehyde (3a-h) (intermediate compounds)

Synthesis of 1, 3-diphenyl 1*H*-pyrazole 4-carbaldehyde (3a)

Phosphorous oxychloride ($POCl_3$ 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 2-phenyl-1-(1-phenylethylidine) hydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of 3-(4-nitrophenyl)-1-phenyl-1*H*-4-carbaldehyde (3b)

Phosphorous oxychloride ($POCl_3$, 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 1-(1-(-(4-nitrophenyl)ethylidene)-2phenylhydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of 3-(3-hydroxyphenyl)-1-phenyl-1*H*-4-carbaldehyde (3c)

Phosphorous oxychloride ($POCl_3$ 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 1-(1-(-(3-hydroxyphenyl)ethylidene)-2-phenylhydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of (1-phenyl-3-p-toly-1*H*-pyrazole 4-carbaldehyde (3d)

Phosphorous oxychloride ($POCl_3$ 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 2-phenyl-1-(1-p-tolyethylidene) hydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of 3-(3, 4-dimethoxy phenyl)-1-phenyl 1*H*-pyrazole 4-carbaldehyde (3e)

Phosphorous oxychloride ($POCl_3$ 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 1-(1-(-(3,4-dimethoxyphenyl) ethylidene)-2-phenylhydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of 3-(2-bromophenyl)-1-phenyl 1*H*-pyrazole 4-carbaldehyde (3f)

Phosphorous oxychloride ($POCl_3$0.01 mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 1-(1-(-(2-bromophenyl) ethylidene)-2-phenylhydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of 3-(4-methoxyphenyl)-1-phenyl 1*H*-pyrazole 4-carbaldehyde (3g)

Phosphorous oxychloride ($POCl_3$ 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0 5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 1-(1-(-(4-methoxyphenyl) ethylidene)-2-phenylhydrazine

(0.01 mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate.

Synthesis of 3-(4-aminophenyl)-1-phenyl 1*H*-pyrazole 4-carbaldehyde (3h)

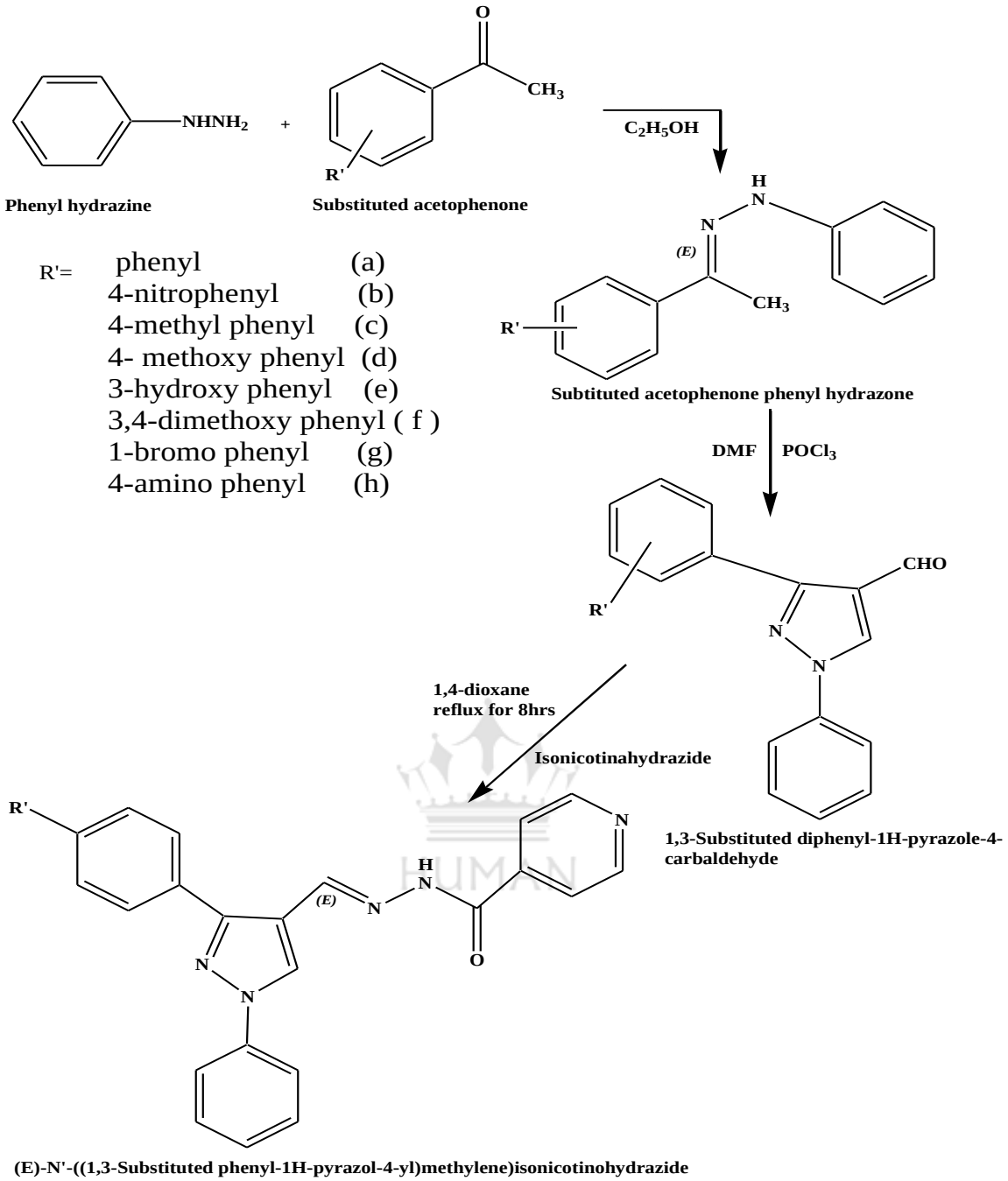
Phosphorous oxychloride ($POCl_3$ 0.01 Mol) was added dropwise to anhydrous N, N-dimethylformamide (DMF, 0.01mol) at 0-5°C with continuous stirring for 15 min at the same temperature. Intermediate mixture 1-(1-(-(4-aminophenyl) ethylidene)-2-phenylhydrazine (0.01mol) was added and reflux for 2hr at 100°C and after the completion of the reaction was confirmed by thin-layer chromatography. The reaction mixture was poured into crushed ice and neutralized with saturated sodium bicarbonate. The compound was obtained and filtered, washed with water and dried. The recrystallization of the compound from ethyl acetate (Dodiya and Pandit, 2012).

SYNTHESIS OF FINAL COMPOUND (4a-h)

Synthetic procedure of Substituted (1, 3-diphenyl-1*H*-pyrazole-4-yl) isonicotinahydrazide (4a-h)

Compound (3a-h), 0.01mol and isonicotinahydrazide (0.01mol) were dissolved in 1, 4-dioxane (20ml) and the reaction mixture was refluxed for 8hrs after completion of the reaction was checked by thin-layer chromatography. When the reaction mixture was cooled the reaction mixture converts into crystal formed. The final compound (3a-h) was obtained and recrystallized from using absolute alcohol (Nisheeth et al., 2016).

Scheme



Characteristic data of synthesized compounds

Synthesis of (1, 3-diphenyl-1*H*-pyrazole-4-yl) isonicotinahydrazide (4a)

TLC *n*-hexane:Ethyl acetate: 7:3v/v, Rf 0.73

FTIR (KBr, cm^{-1}): 3015.21 (Ar, C-H str), 1546.21 (Ar, C=C str), 1659.95 (C=N str), 1660.92 (C=O str), 3014.22 (N-H str), 1665.95 (C=N str, Pyrazole), 1216.26 (C-N str, Pyrazole).

^{1}H NMR (DMSO-d_6(ppm)): 6.51-7.08 (m, 7H, Ar-H), 7.14-7.46 (m, 5H, Ar-H), 7.48-7.501 (d, 2H, Ar-H), 8.32 (s, 1H, CH=N), 8.58 (s, 1H, Pyrazole ring), 7.52 (m, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{22}H_{17}N_5O$: C, 71.92; H, 4.66; N, 19.06.

Synthesis of (3-(4-nitrophenyl)-1-phenyl-1*H* -pyrazole-4-yl) methylene) isonicotinahydrazide (4b)

TLC: *n*-hexane:Ethyl acetate: 7:3v/v Rf 0.69

FTIR (KBr *v*, cm^{-1}): 1640.62 (C=O str), 3118.10 (Ar, C-H str), 1645.62 (C=N str), 3118.10 (N-H str), 1542.40 (C=C str), 1210.92 (C-O str), 1542.40 (N-O str), 1645.62 (C=N str, Pyrazole ring), 1210.92 (C-N str, Pyrazole ring).

^{1}H NMR (DMSO-d_6(ppm)): 7.03-7.29 (m, 8H, Ar-H), 7.37-7.56 (m, 3H, Ar-H), 7.77-7.96 (d, 2H, Ar-H), 8.30 (s, 1H, CH=N), 8.52 (s, 1H, Pyrazole ring), 8.65 (s, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{22}H_{16}N_60_3$: C, 64.07; H, 3.91; N, 20.38.

Synthesis of (1-phenyl-3-p-toly-1*H*Pyrazole-4-yl) methylene) isonicotinahydrazide (4c)

TLC : *n*-hexane:Ethyl acetate: 7:3v/v Rf 0.72
FTIR (KBr, cm^1): 3019.22 (Ar, C- H str), 1409.9 (Ar, C=C str), 1661.93 (C=N str), 1658.89 (C=O str), 3017.20 (N-H str), 1661.93 (C=N str, Pyrazole ring), 1216.26 (C-N str, Pyrazole ring).

^{1}H NMR (DMSO-d_6(ppm)): 2.35 (m, 3H, CH_3), 6.01-7.18 (m, 4H, Ar-H), 7.32-7.47 (m, 5H, Ar-H), 7.57-7.62 (m, 4H, Ar-H), 8.31 (s, 1H, CH=N), 8.50 (s,1H, Pyrazole ring), 8.85 (s, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{23}H_{19}N_5O$: C, 72.42; H, 5.02; N, 18.36

Synthesis of (3-(4-methoxyphenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide (4d)

TLC: *n*-hexane:Ethyl acetate: 7:3v/v **Rf:** 0.72

FTIR (KBr, cm^{-1}): 3110.2 (Ar, C-H str), 1547.41 (Ar, C=C str), 1647.67 (C=N str), 3110.2 (N-H str) 1648.62 (C=O str), 1215.98 (Ar, C-O str), 1646.47 (C=N str, Pyrazole ring), 1215.98 (C-N str, Pyrazole ring).

^{1}H NMR (DMSO-d_6(ppm)): 3.73 (m, 3H, OCH_3), 6.48-6.89 (m, 6H, Ar-H), 7.08-7.26 (m, 5H, Ar-H), 7.31-7.46 (d, 2H, Ar-H), 8.20 (s, 1H CH=N), 8.30 (s, 1H, Pyrazole ring), 8.92 (s, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{23}H_{19}N_5O_2$: C, 69.51; H, 4.82; N, 17.62.

Synthesis of (3-(3--hydroxyphenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide (4e)

TLC: *n*-hexane:Ethyl acetate: 6:4v/v **R_f:** 0.79

FTIR (KBr, cm^1): 3020.90 (Ar, C-H str), 1552.82 (Ar, C=C str), 1665.80 (C=N str), 1750 .10 (C=O str), 3020.76 (N-H str), 1210.90 (C-O str), 1545.90 (C=N str, Pyrazolering) 1268.98 (C-N str, Pyrazole ring).

^{1}H NMR (DMSO-d_6(ppm)): 5.40 (s, 1H, O-H), 6.49-7.03 (d, 2H, Ar-H), 7.14-7.32 (m, 6H, Ar-H), 7.46-7.62 (m, 5H, Ar-H), 8.10 (s, 1H, CH=N), 8.32 (s, 1H, Pyrazole ring), 8.83 (s, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{22}H_{17}N_5O_2$: C, 68.92; H, 4.47; N, 18.27.

Synthesis of (3-(3, 4-methoxyphenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene)isonicotinahydrazide (4f)

TLC: *n*-hexane: Ethyl acetate: 7:3v/v **R_f:** 0.69

FTIR (KBr, cm^{-1}): 3019.95 (Ar, C-H str), 1550.89 (Ar, C=C str), 1663.69 (C=N str), 1752.07 (C=O str), 3020.89 (N-H str), 1215.98 (C-O str), 1602.28 (C=N str, Pyrazole ring), 1215.98 (C-N str, Pyrazole ring).

^{1}H NMR (DMSO-d_6(ppm)): 5.52 (d, 2H, Ar-H), 6.49-7.03 (m, 3H, Ar-H), 7.14-7.32 (m. 5H, Ar-H), 7.46-7.62 (m, 4H, Ar-H), 8.15 (s, 1H, CH=N), 8.35 (s, 1H, Pyrazole ring), 8.56 (s, 1H, N-H)

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{22}H_{17}N_5O_3$: C, 66.16; H, 4.29; N, 17.53.

Synthesis of (3-(2-bromophenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide (4g)

TLC: *n*-hexane: Ethyl acetate: 7:3v/v , **R_f:** 0.76

FTIR (KBr, cm^{-1}): 3015.81(Ar, C-H str), 1547.81 (Ar, C=C str), 1665.62 (C=N str), 1740.02 (C=O str), 3015.85 (N-H str), 550.92 (C-Br str), 1660.02 C=N str, Pyrazole ring), 1139.27 (C-N str, Pyrazole ring).

^{1}H NMR (DMSO-d_6(ppm)): 7.03-7.29 (m, 8H, Ar-H), 7.37-7.56 (m, 3H, Ar-H), 7.77-7.96 (d, 2H, Ar-H), 8.30 (s, 1H, CH=N), 8.52 (s, 1H, Pyrazole ring), 8.65 (s, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{22}H_{16}BrN_5O$: C, 59.21; H, 3.61; N, 15.69.

Synthesis of (3-(4-aminophenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide (4h)

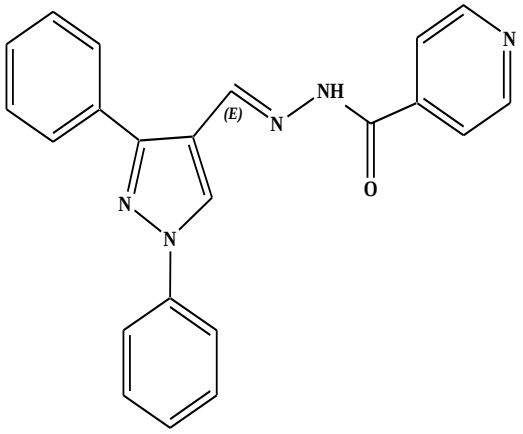
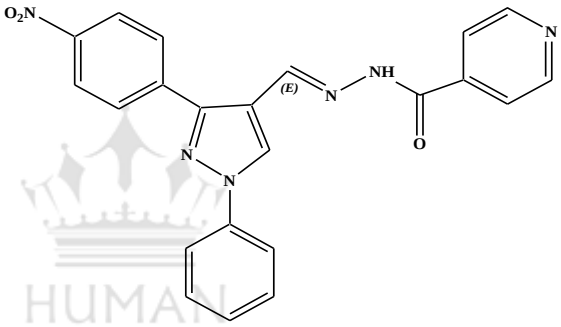
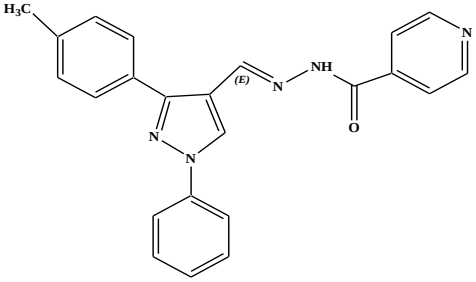
TLC *n*-hexane:Ethyl acetate: 7:3v/v **R_f :** 0.76

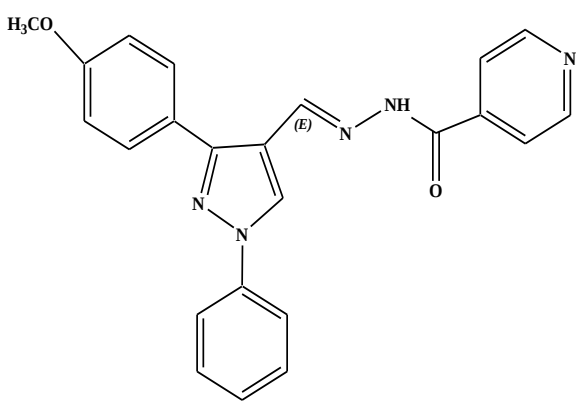
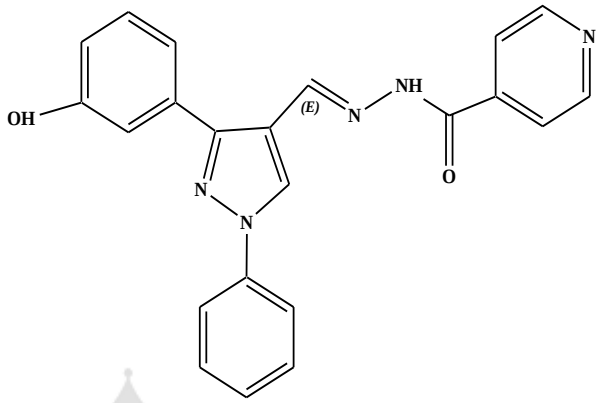
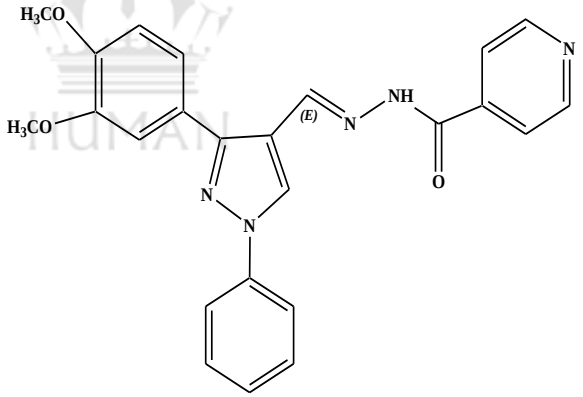
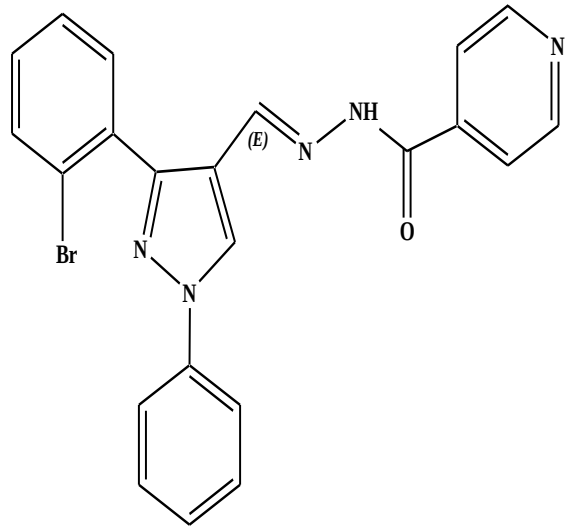
FTIR (KBr *v*, cm^{-1}) :3019.15 (Ar, C-H str), 1549.9 (Ar, C=C str), 1663.84 (C=N str), 3019.9 (N-H str), 1663.84 (C=O str), 1549.9 (C=N str, Pyrazole ring), 1215.33 (C-N str, Pyrazole ring).

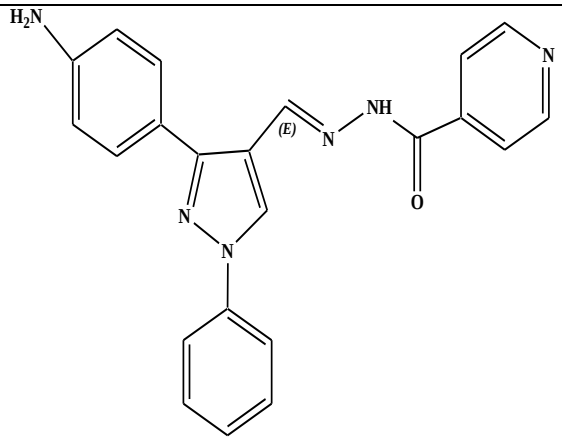
^{1}H NMR (DMSO-d_6(ppm)): 4.21 (d, 2H, N-H), 6.49-7.03 (d, 2H, Ar-H), 7.14-7.32 (m, 6H, Ar-H), 7.71-7.91 (m, 5H Ar-H), 8.21 (s, 1H, CH=N), 8.32 (s, 1H, Pyrazole ring), 8.74 (s, 1H, N-H).

ELEMENTAL ANALYSIS: Anal. Calcd. For $C_{22}H_{18}N_6O$: C, 69.10; H, 4.74; N, 21.98.

Table No. 1: IUPAC name and structure of synthesized compounds

S. No		Structure	M.P. (°C)
4a	1, 3-diphenyl-1*H*-pyrazole-4-yl) isonicotinahydrazide		160-162
4b	(3-(4-nitrophenyl)-1-phenyl-1*H* -pyrazole-4-yl) methylene) isonicotinahydrazide		150-152
4c	(1-phenyl-3-p-toly-1*H*Pyrazole-4-yl) methylene) isonicotin ahydrazide)		156-160

4d	3-(4-methoxyphenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide		140-142
4e	(3-(3--hydroxyphenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide		158-160
4f	(3-(3, 4-methoxyphenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene)isonicotinahydrazide		145-147
4g	3-(2-bromophenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide		148-150

4h	(3-(4-aminophenyl)-1-phenyl-1-*H*-pyrazole-4-yl) methylene) isonicotinahydrazide		152-154

***In vivo* Anticonvulsant activity (Maximal Electroshock Method)**

MATERIALS AND METHODS

Chemicals

The drug used in the study was of pharmaceutical grade. Phenytoin was supplied by SG Pharma, Mumbai, available in the laboratory (HIPER, LUCKNOW).

Animals

Male Wistar Albino rats weighing 100-150 gm were used for anticonvulsant activity. They were housed in standard environmental condition like- ambient temperature (25^0C ±1^0C), relative humidity (55^0C ±5%), and 12/12 hour light-dark cycle. Animals had free access to standard pellet diet and water. All animals' experiments were carried out by the guidelines of the Committee for Control and Supervision on Experiments on Animals (CPCSEA). The institute animal ethical committee has approved for conducting an animal experiment.

Method

The anticonvulsant activity was performed by MES (maximal electroshock method). Adult male and female and female Albino rats (Wister strain) weighing 100-150gm were used. The animals were divided into three groups (control, standard, and test) and each group comprising of three rats. The test compounds were suspended in 1% aqueous CMC suspension and were injected i.p. according to their body weight. Phenytoin sodium was used as a standard drug which was given in the dose of 25mg/kg by i.p. which was observed to protect 100% against the induced convulsions. The control group received only 1% aqueous CMC suspension. The seizures were induced by electroconvulsiometer. The animals were

subjected to electroshock by delivering the current of 150 mA through the corneal electrodes for 0.2 seconds. The animals were observed for 30 min of convulsive responses. Different stages of convulsions i.e. the tonic flexion (towards the upper extremities), tonic extensor phase (extension of the lower extremities), clonic convulsions (intermediates jerking of limbs), stupor (unconsciousness) and recovery or death were observed for each animal (data as shown in table). The anticonvulsant effect of newly synthesized compounds was assessed by the absence or reduction of hind limb tonic extensor phase. Each value represents the mean SEM (standard error mean) of three rats significantly different from standard drug phenytoin.

BIOLOGICAL ACTIVITY

Anticonvulsant activity

The anticonvulsant activity was performed by MES (maximal electroshock method). This project has been approved by the Institutional Animal Ethical Committee at Hygia Institute of Pharmaceutical Education and Research, Lucknow (Ref. No. HIPER/IAEC/03/16/08). Adult albino rats (Wistar strain) of either sex weighing 150-200 gm were used. The animals were divided into three groups (control, standard, and test) and each group comprising of six rats. The test compounds were suspended in 1% aqueous CMC suspension and were administered *p.o*. according to their body weight. Phenytoin sodium was used as a standard drug which was given in the dose of 25 mg/kg by *i.p.*which was observed to protect 100% against the induced convulsions. The control group received only 1% aqueous CMC suspension. The seizures were induced by electroconvulsiometer. The animals were subjected to electroshock by delivering the current of 150 mA through the corneal electrodes for 0.2 seconds. The animals were observed for convulsive responses. Different stages of convulsions *i.e.* the tonic flexion (towards the upper extremities), tonic extensor phase (extension of the lower extremities), clonic convulsions (intermediates jerking of limbs), stupor (unconsciousness) and recovery or death were observed for each animal (as shown in **Table. 1**). The anticonvulsant effect of newly synthesized compounds was assessed by reduction (time) or absence of different phases induced by MES. Each value represents the mean SEM (standard error mean) of three rats significantly different from standard drug phenytoin.

RESULTS AND DISCUSSION

Compounds 4d, 4f, and 4e at 25 mg/kg exhibited significant anticonvulsant activity at all time intervals as compared to the control group. In the primary MES screening compounds 4d, 4fand 4eafforded protection against seizures confirming their potential utility as prototypic molecules. The anticonvulsant activity data revealed that all the compounds showed a remarkable reduction of hind limb tonic flexion, extensor, clonus and stupor phase when given in the dose of 25 mg/kg *p.o*. and compounds 4d and 4f were found to be the most potent compounds in the series. Moreover, anticonvulsant activity of the other test compounds was found to be much less effective than phenytoin used as a standard anticonvulsant drug. According to the results obtained it seems that the presence of the methoxy group and dimethoxy group attached on aryl ring increase the potency. Almost all the derivatives showed good to moderate anticonvulsant activity in comparison with standard phenytoin as shown in Table 2**.**

Table No 2. Anticonvulsant activity of titled compounds 25 mg/kg (Dose)

Compound Code	Flexion (seconds) (mean± SEM)	Extension (seconds) (mean± SEM)	Convulsion (seconds) (mean± SEM)	Stupor (seconds) (mean± SEM)	Recovery/ Death
Control	21.25±	16.05±	17.32±	121±	R
4a	18.35 ±	18.72±	19.33±	122.34±	R
4b	16.33±	17.66±	15.87±	117.23±	R
4c	15.33±	12.25±	14.43±	113.25±	R
4d	12.33±	11.46±	13.35±	107.23±	R
4e	12.32±	12.35±	13.21±	112.34±	R
4f	10.16±	11.33±	12.56±	106.34±	R
4g	16.16±	17.33±	18.33±	116.33±	R
4h	12.43±	11.32±	12.33±	119.68±	R
Standard (Phenytoin sodium)	Absent	9.66 ±	11.66 ±	105.66 ±	R

Data are expressed as Mean ± SEM for different stages. Statistical analysis was performed using two way ANOVA followed by Dunnett's test. ***P<0.001 *vs* control (MES); **P<0.05 *vs* control (MES); *P<0.01 *vs* control (MES)

CONCLUSION

In the present study, various novel pyrazole derivatives were synthesized and characterized by IR, NMR, and Elemental analysis spectroscopy. The entire synthesized compound was screened by anticonvulsant activity for using maximal electroshock (MES) model.

All compounds were synthesized by reaction between phenylhydrazine and substituted acetophenone in presence of ethanol afford the corresponding 2-phenyl-1-(1-phenylethylidene) hydrazine was allowed to react DMF and $POCl_3$ (Vilsmeier hack reaction) to give corresponding pyrazole derivative, such as 1,3-substituted diphenyl 1-H-pyrazole-4-carbaldehydeand then further reaction with 1,3-substituted diphenyl 1-H-pyrazole-4-carbaldehyde with isonicotinahydrazide in the presence of 1,4-dioxane to give corresponding pyrazole derivatives(4a-h).

Among all synthesized compounds, compound 4d and 4f were found to be most potent with compare to standard drug phenytoin in pharmacological evaluation by Maximal Electroshock method, the result of the current study showed pyrazole derivative containing methoxy and dimethoxy group found to be most potent.

The study stated that the pyrazole in combination with other heterocycles ring might be used as a lead for finding the potent anti-convulsant agents. The potency of the newly synthesized compounds was determined based on their reduction in convulsion at different time interval by using the Maximal Electroshock model of three rats significantly different from standard drug phenytoin.

Presence of an electron releasing group on the benzene ring also increases the potency. Substituted pyrazole derivative also increases the therapeutic value of pyrazole core is needed for the discovery of a potent anti-convulsant agent. Thus we observed that pyrazole in combination with other heterocycles might be used as a lead for further study in developing such compounds as a good lead molecule with the better pharmacological profile.

REFERENCES

1. Almansa, C., Gomez, L.A., Arriba, A.F. and Forn, J. 1996. Synthesis and structure-activity relationship of a new series of potent AT_1 selective angiotensin II receptor antagonists 5-(Biphenyl-4-yl methyl) pyrazoles. *Journal of Medicinal Chemistry*. Vol. 40, No. 4, pp. 547-558.
2. Alodeani1, E., Izhari1, A. and Arshad, M. 2015. Antileishmanial screening, physicochemical properties and drug-likeness of pyrazole carbaldehyde derivatives.*Asian Pacific Journal of Health Science*. Vol.2, Issue 2, pp.41-47.

3. Alu-Deeb A. O. and Alafeefy A. M. 2008. Synthesis of some new 3*H*-quinazoline-4-one derivatives as potential antitubercular agents. *World Applied Sciences Journal*. Vol. 5, Issue 1, pp.94-99.
4. Ananda Rajagopal, K., Sunilson, J. A., Illavarasu, A., Thangavelpandian, N. and Kalirajan, R. 2010. Antiepileptic and Antimicrobial Activities of Novel 1(unsubstituted/substituted)-3, 5-dimethyl-1*H* pyrazole Derivatives. *International Journal of ChemTech Research*. Vol. 2, pp.45-49.
5. Balbi, A., Anzaldi, M., Maccio, C., Aiello, C., Mazzei, M., Gangemi, R., Castagnola, P., Miele, M., Rosano, C. and Viale, M. 2011. Synthesis and biological evaluation of novel pyrazole derivatives with anticancer activity. *European Journal of Medicinal Chemistry*. Vol. 46, pp. 5293-5309.
6. Boyd, M.R. and Paull, K.D., 1995. Some practical considerations and applications of the national cancer institute *in vitro* anticancer drug discovery screen. *Drug Development Research*. Vol. 34, pp. 91-109.
7. Campaigne, E. and Albert, A. 1986. Rationalization of Heterocyclic Chemistry. *Journal of Chemical Education*. Vol. 6, pp. 806.
8. Deng, X., Mani, N.S. and Lett, O. 2008. Synthesis of tri- or tetrasubstituted pyrazoles by the reaction of hydrazones with nitro-olefins. *Journal of Organic Chemistry*. Issue 10, pp.1307-1310.
9. Gosselin, F., O'Shea, P. D., Webster, R. A., Reamer, R.A., Tillyer, R.D. and Grabowski J.J.2006. Arylhydrazi nes proceeds at room temperature in N, N-dimethylacetamide and furnishes pyrazoles. *Journal of Organic Chemistry*. Vol. 14, pp. 3267-3270.
10. Han, Q., Lam, P.Y.S., Chang, C.S. and Li, R. 1998. Cyclic HIV protease inhibitors: Design and synthesis of orally bioavailable, pyrazole P2/P2 cyclic ureas with improved potency. *Journal of Medicinal Chemistry*. Vol. 41, pp. 2019-2028.
11. Ikeda, M., Maruyama, K., Nobuhara, Y. and Okabe, S. 1996. Synthesis and cytoprotective antiulcer activity of 2- or 4-(1*H*-Pyrazol-1-yl)pyrimidine derivatives related to mepirizole and dulcerozine. *Chemical and Pharmaceutical Bulletin*. Vol. 44, pp. 1700-1706.
12. Jiang, J.A., Du, C. Y., Gu, Y.F. and Synlett, Ji. 2012. Synthesis of 4-Substituted 1, 5-Diaryl-1H-pyrazole-3-carboxylic Acids via a MeONa/LiCl-Mediated Sterically Hindered Claisen. *Journal of Organic Chemistry*. Issue 23, pp. 2965-2968.
13. Kawale, P. R., Doehate, P. P., Berad, B. N., Srivastava K. K. and Sharma, P. 2013. Synthesis of antituberculosis study of new amino acridine linked pyrazole. *Journal of Organic Chemistry*. Vol. 52 B, pp.1531-1535.
14. Keepers, Y.P., Piazao, P.E., Peters, G.J., Ark-Otte, J., Winogard, B. and Pinedo, H.M. 1991. Comparison of the Sulforhodamine B protein and Tetrazolium (MTT) assay for *in vitro* chemosensitivity testing. *European Journal of Cancer*. Vol. 27, No. 7, pp. 897-900.
15. Khunt, R.C., Khedkar, V.M., Chadwa, R.S., Chauhan, N.A., Parikh, A.R. and Cooutinho E.C. 2012. Synthesis, antitubercular evaluation and 3D-QSAR study of *N*-phenyl-3-(4-fluorophenyl)-4-substituted pyrazole derivatives. *Bioorganic and Medicinal Chemistry Letters.* Vol.22, pp. 666-678.
16. Kumar, D., Pal, S. S. and Singh, S. 2012. Importance of Pyrazole moiety in the field of cancer. *International Journal of Pharmacy and Pharmaceutical Sciences*. Vol. 4, Issue 2, pp.98-10.
17. Kudo, N., Furuta, S., Taniguchi, M., Endo, T. and Sato, K., 1999. Synthesis and herbicidal activity of 1, 5-Diarylpyrazole derivatives. *Chemical and Pharmaceutical Bulletin*, Vol. 47(6), pp. 857-868.
18. Kumar, S.V., Yadav, S.K., Raghava, B., Saraiah, B., Ila, H., Ragappa, K.S. and Hazra, A. 2013. 1, 3-bisaryl-monothio-1,3-diketone or 3-(methylthio)-1, 3-bisaryl-2-propenones with arylhydrazines furni shed 1-aryl-3,5-bisarylpyrazoles. *Journal of Organic Chemistry*. Issue 78, pp.4960 4973.
19. Mamolo, M.G., Zampieri, D., Falagiani, V., Vio, L. and Banfi, E. 2001. Synthesis and antimycobacterial activity of 5-aryl-1-isonicotinoyl-3-(pyridine-2-yl)-4,5-dihydro-1*H*-pyrazole derivatives. *Journal of IL Farmaco*. Vol. 56, pp. 593-599.
20. Mittapelli, V. and Padala, S. R. 2014. Synthesis and anti-diabetic activity of some 3-methyl quinazoline-4(3H)-one derivatives. *International Journal of ChemTech Research.* Vol. 6, Issue 14, pp. 5647-5652.
21. Nallasivan, P. K. and Gopal, N. 2015. Anti-inflammatory activity of pyrazoline containing quinazoline derivative. *Asian Journal of Pharmaceutical Analysis And Medicinal Chemistry*. Vol. 3, Issue 1, pp. 31-38.
22. Nitulescu, G.M., Draghici, C. and Missir, A.V. 2010. Synthesis of new pyrazole derivatives and their anticancer evaluation. *European Journal of Medicinal Chemistry*. Vol.45, pp. 4914-4919.

23. Panda, N. and Jena, A. K. 2012. Fe-Catalyzed One-Pot Synthesis of 1, 3-Di- and 1, 3, 5-Trisubstituted Pyrazoles from Hydrazones and Vicinal Diols. *Journal of Organic Chemistry*. Issue 77, pp. 9401-9406.
24. Patel, N.M. 2011. Design, synthesis, characterization and biological evaluation of certain new derivative of nitrogen heterocycles, *journal of dissertation Protocol*, Luqman college of pharmacy, Gulbarga, pp. 1-15.
25. Patil, A., Jadhav, R., Raundal, H., Sharma, L., Badgujar, R. and Bobade, V. 2014. Synthesis and antifungal activities of diaryl pyrazoles carboxamide derivatives. *Journal of Chemical and Pharmaceutical Research*. Vol. 6, pp.218-223.
26. Patra, P. K., Patra, N. and Pattnaik, S. 2014. Synthesis and antibacterial activity. *International journal of pharmaceutical sciences*. Vol. 6, Issue1, pp.801-805.
27. Pattan, S.R., Patel, G.S., Atharel, A.B., Nirmal, S.A. and Pattan, J.S. 2014. Synthesis and Evaluation of some substituted Pyrazole derivatives of biological interest. *Journal of Bulgarian Chemical Communications*. Vol.46, pp.125–134.
28. Priyadarsinia, P., Ujwalaa, B., Raob, C. V. and Rao, V. M. 2012. Synthesis and activity screening of some novel pyrazole derivative antimicrobial activity of some novel pyrazoles. Vol 4, pp.1123-1128.
29. Rajeev, J., Gupta, S., Jamwal, J., Javed, A. and Bhardwaj, V. 2013. A review on Pyrazole derivatives of pharmacological potential. *International journal of pharmaceutical and bioscience*.Vol. 3, pp. 114-123.
30. Sahu, S. K., Banerjee, M., Samantray, A., Behera, C., and Azam, M.A. 2008. Synthesis, Analgesic, Anti-inflammatory and Antimicrobial Activities of Some Novel Pyrazoline Derivatives. *Tropical Journal of Pharmaceutical Research*. Vol. 7, pp.961-968.
31. Sanchez, J.G.B. and Kouznetsov, V.K., 2010. Antimycobacterial susceptibility testing methods for natural products research. *Brazilian Journal of Microbiology*, Vol. 41, pp. 270-277.
32. Sha, Q. and Wei, A. 2013. 1, the 3-dipolar cycloaddition of diazo compounds and alkynyl bromides gives 3, 5-diaryl-4- sbromo-3H-pyrazoles. *Journal of Organic Chemistry*. Issue 45, pp. 413-420.
33. Shiota, T., Yamamori, T., Sakai, K., Kiyokawa, M., and Honma, T., 1999. Synthesis and structure-activity relationship of a new series of potent angiotensin II receptor antagonists: Pyrazolo[1,5-*a*]pyrimidine derivatives. *Chemical and Pharmaceutical Bulletin*, Vol. 47, No. 7, pp. 928-938.
34. Viale, M., Anzaldi, M., Aiello, C., Fenoglio, C., Albicini, F., Emionite, L., Gangemi, R. and Balbi, A. 2013. Evaluation of the anti-proliferative activity of three new pyrazole compounds in sensitive and resistant tumor cell lines. *Pharmacological Report*. Vol. 65, pp.717-723.
35. Yerragunta, V., Suman, D., Swamy, K., Anusha, V., Patil, P. and Naresh, M. 2014. Pyrazole and Its Biological Activity, *PharmaTutor*. Vol.2, Issue1, pp.40-48.
36. Zhang, G., Ni, H., Chen, W., Shao, J., Liu, H. and Chen, B. 2013. Vinyl azide, aldehyde, and tosylhydrazine afford 3, 4, 5-trisubstituted 1H-pyrazoles regioselectively. *Journal of Organic Chemistry*, Issue15, pp. 5967-5969.
37. Zhang, X., Kang, N. P., Wu, J. and Chang, J. 2006. Facile access to various di-, tri-, and tetrasubstituted (aryl, alkyl, and/or vinyl) pyrazoles from readily available α, β, unsaturated aldehydes/ketones. *Journal of Organic Chemistry*. Issue 79, pp. 10170-10178.
38. Reddy, S. K., Khan, K., and Nagaraja, C. 2106. A review on the determination of melting point measurement system. *International Journal of Advanced Research*. In electrical, electronic and instrumentation Engg. Vol, 5. Issue 2, pp. 975-979
39. Kumar, S., Jyotirmayee, K. and Sarangi, M. 2013. Thin-layer chromatography: A tool of biotechnology for isolation of bioactive compound from medicinal plants. *International Journal Pharm. Science.* Issue 18, pp.126-132.
40. Builla, J.A. and Barluenga, J. 2011. Heterocyclic compounds: An introduction. *Journal of Modern Heterocyclic Chemistry*. First edition. pp. 1-9.
41. Aljamali, N.M. 2015. Review paper on heterocyclic compounds. *Journal of plastic and polymer technology*, Vol, 1. Issue 1, pp. 49-64.
42. Desai, N.C., Kotadiya, G.M., Trivedi, A.R., Khedkar, V.M. and Jha, P.C. 2016 Design, synthesis and biological evaluation of novel fluorinated pyrazole encompassing pyridyl 1,3,4-oxadiazole motifs. *Journal of Medicinal Chemistry Research*. pp. 1-8.
43. Dodiya, A. and Pandit, U. 2012. Synthesis and antitubercular activity of novel pyrazole quinazolinone hybrid analog. *Journal of Medicinal Chemistry Research*.

Issue 22, pp. 3364-3371.
44. Cetin, A. 2015. International journal of current research in chemistry and pharmaceutical sciences. *International Journal of Current Research Chemistry Pharmaceutical Science.* Vol. 2, Issue 7, pp. 8-20.
45. Jadhav, P.B. and Pandey, P.S. 2013. Phase solubility analysis: A Technique of purity determination. *World research journal of pharmaceutical research*. Vol. 1, Issue 1, pp. 05-11.