Volume 8, Issue 6, 2018, 3744 - 3750 ISSN 2069-5837

Biointerface Research in Applied Chemistry

www.BiointerfaceResearch.com

Original Research Article Open Access Journal

Received: 03.09.2018 / Revised: 22.11.2018 / Accepted: 29.11.2018 / Published on-line: 15.12.2018

# A new approach for the synthesis of 8-((1,3-diphenyl-4,5-dihydro-1*H*-pyrazole-5 yl)methoxy)quinoline: a novel lead for breast cancer chemotherapy

**Kumbaradoddi B Umesha[1], Srikantamurthy Ningaiah[2], Nagarakere S Lingegowda[2], Vrushabendra Basavanna[2], Shridevi Doddamani[3,*]**

[1] Department of Chemistry, Yuvaraja's College, University of Mysore, Mysuru-570005, Karnataka, India
[2]Department of Chemistry, Vidyavardhaka College of Engineering, Mysore-570 002, Karnataka, India
[3]Chemical Sciences and Technology Division, CSIR-NIIST, Thiruvananthapuram 695 019, Kerala, India
*corresponding author e-mail address: ***shridevi20@gmail.com, shridevi20@niist.res.in***

## ABSTRACT

The present effort deals with the discovery of highly potent pyrazole bearing quinoline a novel lead for breast cancer chemotherapy. The target molecules were synthesized by 8-hydroxyquinoline and allyl chloride as precursors under basic medium followed by 1,3-dipolar cycloaddition reaction with benzaldehyde phenyl hydrazone under ethanol solvent medium. This reaction route cultivates many advantages such as easy work-up and environment friendly solvent system. At the same time they structurally characterized by $^{1}$H-NMR, $^{13}$C-NMR, IR spectral data and elemental analysis. In addition to that, the newly obtained compounds were screened for anti-breast cancer activity. Although the cytotoxicity of the obtained analogs was determined using human breast tumor cell lines MCF7, compounds 4b, 4c showed very good effect on MCF7 cancer cells and further, they may helpf to develop a perfect lead for breast cancer MCF7 cell line.

**Keywords:** *Pyrazole, 8-hydroxyquinoline, 1,3-dipolar cycloaddition, breast cancer.*

## 1. INTRODUCTION

Advanced Breast Cancer (ABC) [1] remains the furthermost precarious type of disease, which is diagnosed each year in most of the developed countries like the USA. On the basis of American Cancer Society and the National Cancer Institute survey, 40,610 women and 460 men die yearly in the ratio of 9:1 which is second death [2] causing disease among women. A lump in breast or armpit considered as breast cancer including nipple discharge, nipple redness, and breast skin texture and swelling symptoms. Most of the patients have hormone receptor positive ($HR^+$) which includes Estrogen receptor-positive [3-5] [$ER^+$] and/or progesterone receptor-positive [$PgR^+$] diseases [6]. MCF7 is an epithelial cancer cell line sensitive towards estrogen, progesterone, glucocorticoid and cytokeratin, which was first isolated from breast tissue and widely used for *in vitro* breast cancer studies [7]. When come to the treatment, some effective way to treat patients who are suffering from ABC-$HR^+$ type effectively by chemotherapy, advanced endocrine-based therapeutics [8-11], selective ER modulator (SERM) tamoxifen [12] and Androgen insensitivity syndrome (AIs) [13] followed by ESR mutation [14-15]. So here we have presented a work which aims to develop a novel lead for breast cancer disease chemotherapy.

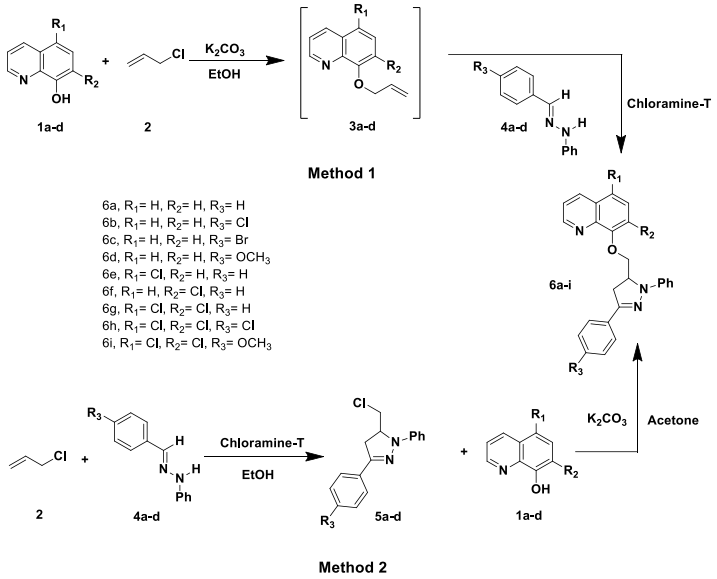
Quinoline moieties attract every chemist in recent trend because many quinoline bearing heterocycles were isolated from the plant [16-20] as an important ingredient, from which many alkaloids and their analogs can be prepared [21-23]. For example, chloroquine (CQ), quinine, camptothecin, luotonine-A are widely used alkaloid [24] for many diseases from ancient time. Among quinoline analogs, 8-Hydroxy Quinoline [25] (8HQ) is one of the most popular heterocycle quinoline with formula $C_9H_7NO$, which is widely used to develop many biologically important hetero molecules. That is why quinoline analogs have wide range of applications in pharmaceutical activities such as antibiotics [26], anticancer [27], anti-inflammatory [28-29], excellent selective cytotoxicity towards SMMC-772 cell line [30], potent MEK1 kinase inhibitors [31], irreversible inhibitors of human epidermal growth factor receptor-2 kinase activity [32], and inhibitors of tumor progression loci-2 (Tpl2) kinase and tumor necrosis factor alpha (TNF-α) production [33].

Pyrazolines [34] are two nitrogen-containing, electron rich five membered hetero molecule which found as a biologically important such as; anti-tumor [35], anti-bacterial [36], anti-fungal [37], anti-coagulant [38], anti-microbial [39], antiviral [40] and many more activities [41]. These flexible molecules have extensive application as synthons [42-49] in the production of many organic molecules. With a better understanding of chemical and biological efficiency with respect to quinoline and pyrazole analogs, we are attracted and prompted to obtain some pyrazole bearing quinoline derivatives through 1,3-dipolar cycloaddition reaction between nitrile imine [50] and olefin double bond.

## 2. EXPERIMENTAL SECTION

**Method 1: General procedure for the synthesis of 6a.** To the refluxed solution of 8-hydroxyquinoline **(1a)** (1.45 g, 10 mmol) and potassium carbonate (2.07 g, 15 mmol, 1.5 eq) in dry ethanol (30 ml), added allyl chloride **(2)** (4.0 ml, 50 mmol, 5 eq) drop by drop over the period of 30 minutes [51]. Then the resultant solution was refluxed for 12 hours and progress was monitored by

TLC. After complete reaction (monitored by TLC), we have added prior generated nitrile imine obtained by treating benzaldehyde phenyl hydrazone **(4a)** (2.3 g, 12 mmol, 1.2 eq) with chloramine-T (3.37 g, 12 mmol, 1.2 eq) in dry ethanol solvent medium very slowly with constant stirring over the period of 30 min. After complete addition, the resultant solution stirred for 16 hours at room temperature. Further, the reaction mass was transferred into a beaker, evaporated, recrystallized with dry ethanol and obtained cream colored solid with 89% yield (3.38 g). The same procedure was followed for all the derivatives.




**Scheme 1**: Synthesis of quinolinyl-pyrazole scaffold *via* two different approaches

**Method 2: General procedure for the synthesis of 6a.** 8-hydroxy quinoline **(1a)** (1.45 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq) and dry acetone (25 ml) were taken in a 100 ml round bottom flask and added 5-(chloromethyl)-1,3-diphenyl-4,5-dihydro-1*H*-pyrazole **(5a)** (3.24 g, 12 mmol, 1.2 eq) in dry acetone (25 ml) drop by drop over the period of 30 minutes. The resultant solution was refluxed for 12 hours and reaction progress was monitored by TLC. The reaction mass was neutralized by adding dilute hydrochloric acid followed by washing successively with water. The reaction mass was extracted with ether (25 ml x 3), washed with brain (25 ml x 3) and finally with water (25 ml x 3). Finally, the ether layer was evaporated and the crude mass was recrystallized with hot ethanol which results in cream colored solid with 82% yield (3.11 g) and 48% overall yield.

**General procedure for the synthesis of Benzaldehyde Phenyl Hydrazone (4a-d)** [52, 53]**:** Benzaldehyde (1 ml, 10 mmol) and dry ethanol (10 ml) were taken in a dry round bottom flask and stirred thoroughly at 60$^{o}$C with the help of magnetic stirrer to obtain a homogeneous solution. To the above mixture, added phenyl hydrazine (1 ml, 10 mmol, 1 eq) in dry ethanol (5 ml) for a period of 30 minutes with the help of liquid addition funnel. After complete addition, the reaction mixture allowed to stir for one hour which yields pale yellow solid. Then the mixture cooled and the solid was filtered off. Then the solid was washed twice with ethanol and dried in the oven. Finally, the crude product was recrystallized with hot ethanol which results in a light yellow amorphous solid with a 93% yield (1.83 g). mp: 156°C. The same procedure was followed for all the derivatives.

**General procedure for the synthesis of 5a** [54]**:** Added allyl chloride **(2)** (8.1 ml, 50 mmol, 5 eq) very slowly to a mixture of benzaldehyde phenyl hydrazone **(4a)** (1.96 g, 10 mmol) and chloramine-T (3.37 g, 12 mmol, 1.2 eq) in dry ethanol (30 ml) solvent. After complete addition, the resultant solution was stirred for 12 hours and reaction progress was monitored for completeness by monitoring TLC. The resultant solid was filtered off, washed with ethanol and dried in the oven. Finally, the crude product recrystallized with hot ethanol and obtained a white solid with 60% yield (1.90 g), which was used for the further reaction. The same procedure was followed for all the derivatives.

## 3. RESULTS SECTION

The two novel approaches for the synthesis of pyrazole bearing quinoline derivatives were achieved; i) by one pot *O*-allylation of 8-hydroxyquinoline **(1)** with allyl chloride **(2)** under basic medium followed by *in situ* 1,3-dipolar cycloaddition reaction between nitrile imine and carbon-carbon double bond in the compound **3(a-d)**. ii) In the second method pyrazolines, **5(a-d)** were prepared first by treating benzaldehyde phenyl hydrazone **4(a-d)** with allyl chloride via 1,3-dipolar cycloaddition in the presence of Chloramine-T in ethanol as solvent. Then obtained pyrazolines were treated with 8-hydroxyquinoline in the presence of $K_2CO_3$ in dry acetone to get the target compound **6(a-i)**. When compared to method 2, it was envisioned that the yield obtained from method 1 was good (89%) than in method 2 (48%) due to the formation of undesired bi-cycloadduct [55] such as 1,3,4,6-tetraphenyl-1,4-dihydro-1,2,4,5-tetrazines during cyclization of nitrile imine and olefin double bond. In addition to that, electron rich dipolarophiles [56] such as oxygen bearing olefins drives regioselective 1,3-dipolar cycloaddition reaction to compare the electron deficient chlorine bearing olefins. Another important reason was that since the formation of pyrazole is purely electrophilic [57], the compound **3(a-d)** is most active towards electrophilic cycloaddition due to the presence of resonance stabilized electron rich oxygen of 8-HQ which in turn increases the double bond character for the cycloaddition reaction with nitrile imine. On the other hand, electron density on olefin double bond of allyl chloride will be less due to the negative inductive effect of chlorine atom which in turn reduces the reactivity towards 1,3-dipolar cycloaddition reaction with **4(a-d)**. And also, through **method 1** we have avoided the multi-step purification by following one pot tandem synthesis by adding prior generated nitrile imine to the reaction mass. Fortunately, we could able to get the compound **6a** in good yield (89%) from one step method which was never obtained from the multi-step method.

Aforementioned reasons prompted us to follow **method 1** to get target compounds **6(a-i)**.

All the newly synthesized compounds were analyzed by spectral ($^1$H NMR, $^{13}$C NMR, IR) analysis and they were reliable with the anticipated structure. A sharp characteristic doublet at $\delta$ = 2.8 for –$\underline{CH_2}$- group and at $\delta$ 3.7 for –$\underline{OCH_2}$- group and multiplet at $\delta$ 3.1 for –$\underline{CH}$< group and at $\delta$ 6.8-7.6 for aromatic ring reveals the formation of the title compound. Intense peak ($M^+$) at 379 in mass spectra supported the structure of the target molecule. Also, characteristic sharp peak at $\delta$ = 34.4 and $\delta$ = 66.1 for –$\underline{C}H_2$– group of pyrazole ring and for O–$\underline{C}H_2$– group in $^{13}$C spectra supports the formation of the target molecule. On the other hand, strong absorption band at 1265 cm$^{-1}$ in IR spectra confirmed the presence of alkyl aryl ether (Ar-$\underline{O}$-$CH_2$) group, which reveals the structure of the final compound.

**SPECTRAL ANALYSIS.**

**5-(chloromethyl)-1,3-diphenyl-4,5-dihydro-1*H*-pyrazole (5a).** Obtained from benzaldehyde phenyl hydrazone (1.96 g, 10 mmol), allyl chloride (8.1 ml, 50 mmol, 5 eq), chloramine-T (3.37 g, 12 mmol, 1.2 eq), in dry ethanol (30 ml). White solid. Yield: 60% (1.90 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.9 (m, 1H, CH), 3.1 (d, 2H, $CH_2$), 3.6 (d, 2H, $CH_2$), 6.7-7.3 (m, 5H, ArH), 7.5-7.7 (m, 5H, ArH). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 35.1, 52.1, 60.4, 116.6, 120.8, 128.3, 128.7, 129.4, 131.0, 136.5, 142.5, 151.9. IR spectrum, (KBr) ν: 3115, 3000, 1560, 1220, 1120. MS, m/z (% Rel. intensities): m/z: 270.1 ($M^+$), 272.1, 271.1, 273.0, 272.0. Found, %: C, 70.95; H, 5.59; N, 10.38; Cl, 13.08, Calculated, %: C, 70.98; H, 5.58; N, 10.35; Cl, 13.09.

**5-(chloromethyl)-3-(4-chlorophenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazole (5b).** Obtained from 4-chloro-benzaldehyde phenyl hydrazone (2.3 g, 10 mmol), allyl chloride (8.1 ml, 50 mmol, 5 eq), chloramine-T (3.37 g, 12 mmol, 1.2 eq) in dry ethanol (30 ml). Light yellow solid. Yield: 59% (1.78 g).

**3-(4-bromophenyl)-5-(chloromethyl)-1-phenyl-4,5-dihydro-1*H*-pyrazole (5c)).** Obtained from 4-bromo-benzaldehyde phenyl hydrazone (2.65 g, 10 mmol), allyl chloride (8.1 ml, 50 mmol, 5 eq), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Cream colored solid. Yield: 62% (2.09 g).

**5-(chloromethyl)-3-(4-methoxyphenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazole (5d).** Obtained from 4-methoxy-benzaldehyde phenyl hydrazone (2.27 g, 10 mmol), allyl chloride (8.1 ml, 50 mmol, 5 eq), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Pale yellow colored solid. Yield: 66% (1.98 g).

**8-((1,3-diphenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6a).** ***Method 1*:** Obtained from 8-hydroxy quinoline (1.45 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), phenyl hydrazone (2.3 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (50 ml). Cream coloured solid. % of Yield: 89% (3.38 g).

***Method 2*:** 8-hydroxy quinoline (1.45 g, 10 mmol), 5-(chloromethyl)-1,3-diphenyl-4,5-dihydro-1*H*-pyrazole (3.24 g, 12 mmol, 1.2 eq), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), dry acetone (50 ml). Cream coloured solid. % of Yield: 82% (3.11 g). Overall Yield: 48%.

$^1$H NMR (400 MHz, $CDCl_3$): δ 2.87 (d, 2H, $CH_2$), 3.15 (p, 1H, CH), 3.78 (d, 2H, $OCH_2$), 6.79 (m, 1H, ArH), 7.25 (m, 2H, ArH), 7.27 (m, 2H, ArH), 7.53 (m, 3H, ArH), 7.64 (m, 2H, ArH), 7.53 to 8.89 (m, 6H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.4, 57.6, 66.1, 107.5, 116.8, 117.5, 120.9, 121.8, 126.9, 128.5, 128.9, 129.7, 129.9, 131.1, 135.8, 136.3, 140.4, 142.6, 149.0, 151.8, 155.5. IR spectrum, (KBr) ν: 3124, 2941, 1597, 1265, 1194. MS, m/z (% Rel. intensities): 379.18 ($M^+$), 379.98, 381.18, 380.12. Found, %: C, 79.11; H, 5.57; N, 11.09; O, 4.23, Calculated, %: C, 79.13; H, 5.58; N, 11.07; O, 4.22.

**8-((3-(4-chlorophenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6b).** Obtained from 8-hydroxy quinoline (1.45 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), 4-chloro-phenyl hydrazone (2.76 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Light yellow amorphous solid. Yield: 84% (3.47 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.84 (d, 2H, $CH_2$), 3.14 (p, 1H, CH), 3.77 (d, 2H, $OCH_2$), 6.78 (m, 1H, ArH), 7.24 (m, 2H, ArH), 7.25 (m, 2H, ArH), 7.56 (m, 2H, ArH), 7.99 (m, 2H, ArH), 7.51 to 8.86 (m, 6H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.2, 57.6, 65.7, 107.3, 116.8, 117.4, 120.9, 121.4, 126.7, 128.4, 128.8, 128.9, 129.3, 129.6, 134.7, 135.7, 136.6, 140.4, 142.3, 149.3, 151.8, 155.5. IR spectrum, (KBr) ν: 3115, 2944, 1590, 1248, 1193, 784, MS, m/z (% Rel. intensities): 413.11 ($M^+$), 415.13, 414.15, 416.16, 415.11, 417.14, 414.12. Found, %: C, 72.59; H, 4.88; Cl, 8.56; N, 10.12; O, 3.85, Calculated, %: C, 72.55; H, 4.87; Cl, 8.57; N, 10.15; O, 3.87.

**8-((3-(4-bromophenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6c).** Obtained from 8-hydroxy quinoline (1.45 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), 4-bromo-phenyl hydrazone (3.18 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Brown colored semi-solid. Yield: 80% (3.69 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.85 (d, 2H, $CH_2$), 3.15 (p, 1H, CH), 3.79 (d, 2H, $OCH_2$), 6.79 (m, 1H, ArH), 7.26 (m, 2H, ArH), 7.28 (m, 2H, ArH), 7.63 (m, 2H, ArH), 7.74 (m, 2H, ArH), 7.50 to 8.88 (m, 6H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.1, 57.5, 65.8, 107.1, 116.5, 117.5, 120.4, 121.8, 125.4, 126.8, 128.6, 129.4, 129.5, 131.4, 131.9, 135.2, 135.6, 140.3, 142.3, 149.1, 151.8, 155.6. IR spectrum, (KBr) ν: 3111, 2938, 1582, 1200, 1179, 692. MS, m/z (% Rel. intensities): 457.07 ($M^+$), 459.10, 458.10, 460.08, 459.08, 461.07, 458.07, 460.07. Found, %: C, 65.49; H, 4.41; Br, 17.40; N, 9.19; O, 3.51, Calculated, %: C, 65.51; H, 4.40; Br, 17.43; N, 9.17; O, 3.49

**8-((3-(4-methoxyphenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6d).** Obtained from 8-hydroxy quinoline (1.45 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), 4-methoxy-phenyl hydrazone (2.64 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). White colored semi-solid. Yield: 88% (3.58 g). $^1$H NMR (400 MHz, $CDCl_3$): $\delta$ 2.86 (d, 2H, $CH_2$), 3.13 (p, 1H, CH), 3.77 (d, 2H, $OCH_2$), 3.84 (s, 3H, $OCH_3$), 6.79 (m, 1H, ArH), 7.23 (m, 2H, ArH), 7.26 (m, 2H, ArH), 7.07 (m, 2H, ArH), 7.95 (m, 2H, ArH), 7.50 to 8.88 (m, 6H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.2, 55.7, 57.5, 65.5, 107.3, 114.5, 116.8, 117.4, 120.8, 121.8, 126.4, 128.5, 128.7, 129.4, 129.9, 131.8, 135.5, 140.7, 142.4, 149.2, 151.8, 155.7, 162.7. IR spectrum, (KBr) ν: 3121, 2899, 1585, 1203, 1190. MS, m/z (% Rel. intensities): 409.16 ($M^+$), 410.15, 411.10, 410.17. Found, %: C, 76.20; H, 5.68; N, 10.27; O, 7.85, Calculated, %: C, 76.26; H, 5.66; N, 10.26; O, 7.81.

**5-chloro-8-((1,3-diphenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6e).** Obtained from 5-chloro-8-hydroxy quinoline (1.79 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), phenyl hydrazone (2.3 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (50 ml). Cream colored semi solid. Yield: 79% (3.26 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.88 (d, 2H, $CH_2$), 3.13 (p, 1H, CH), 3.76 (d, 2H, $OCH_2$), 6.77 (m, 1H, ArH), 7.26 (m, 2H, ArH), 7.25 (m, 2H, ArH), 7.52 (m, 3H, ArH), 7.67 (m, 2H, ArH), 6.99 to 8.99 (m, 5H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.5, 57.7, 66.4, 106.9, 116.9, 118.4, 120.9, 122.6, 126.3, 127.8, 128.4, 128.9, 129.6, 131.4, 133.6, 136.5, 139.7, 142.4, 151.8, 152.3, 154.9. IR spectrum, (KBr) ν: 3120, 2971, 1589, 1211, 1189, 798. MS, m/z (% Rel. intensities): 413.14 ($M^+$), 415.15, 414.12, 416.18, 415.11, 417.11, 414.12. Found, %: C, 72.56; H, 4.86; Cl, 8.56; N, 10.14; O, 3.88, Calculated, %: C, 72.55; H, 4.87; Cl, 8.57; N, 10.15; O, 3.87.

**7-chloro-8-((1,3-diphenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6f).** Obtained from 7-chloro-8-hydroxy quinoline (1.79 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), phenyl hydrazone (2.3 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Yellow colored semi-solid. Yield: 82% (3.38 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.85 (d, 2H, $CH_2$), 3.14 (p, 1H, CH), 3.79 (d, 2H, $OCH_2$), 6.78 (m, 1H, ArH), 7.26 (m, 2H, ArH), 7.26 (m, 2H, ArH), 7.53 (m, 3H, ArH), 7.69 (m, 2H, ArH), 7.58 to 8.89 (m, 5H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.3, 57.8, 65.7, 111.3, 116.8, 117.9, 120.8, 122.3, 127.7, 128.4, 128.6, 128.8, 129.7, 131.4, 135.7, 136.3, 139.5, 142.6, 150.3, 151.8, 155.2. IR spectrum, (KBr) ν: 3119, 2977, 1588, 1209, 1199, 787. MS, m/z (% Rel. intensities): 413.13 ($M^+$), 415.15, 414.12, 416.14, 415.12, 417.11, 414.17. Found, %: C, 72.57; H, 4.86; Cl, 8.56; N, 10.17; O, 3.84, Calculated,%: C, 72.55; H, 4.87; Cl, 8.57; N, 10.15; O, 3.87.

**5,7-dichloro-8-((1,3-diphenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6g).** Obtained from 5,7-dichloro-8-hydroxy quinoline (2.13 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), phenyl hydrazone (2.3 g, 12 mmol, 1.2), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Brown colored semi solid. Yield: 70% (3.15 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.83 (d, 2H, $CH_2$), 3.12 (p, 1H, CH), 3.81 (d, 2H, $OCH_2$), 6.77 (m, 1H, ArH), 7.24 (m, 2H, ArH), 7.23 (m, 2H, ArH), 7.53 (m, 3H, ArH), 7.64 (m, 2H, ArH), 7.52 to 8.99 (m, 4H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.4, 57.8, 65.6, 110.9, 116.8, 118.5, 120.8, 122.9, 124.3, 127.6, 128.5, 128.6, 129.7, 131.4, 133.2, 136.5, 140.1, 142.4, 151.8, 153.3, 154.1. IR spectrum, (KBr) ν: 3119, 2939, 1588, 1221, 1187, 799. MS, m/z (% Rel. intensities): 447.01 ($M^+$), 449.07, 448.11, 450.14, 451.01, 449.09, 452.05, 451.04, 448.12. Found, %: C, 66.97; H, 4.25; Cl, 15.84; N, 9.39; O, 3.55, Calculated, %: C, 66.97; H, 4.27; Cl, 15.82; N, 9.37; O, 3.57

**5,7-dichloro-8-((3-(4-chlorophenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6h).** Obtained from 5,7-dichloro-8-hydroxy quinoline (2.13 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), 4-chloro-phenyl hydrazone (2.77 g, 12 mmol, 1.2 eq), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Light grey semi solid. Yield: 65% (3.16 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.85 (d, 2H, $CH_2$), 3.13 (p, 1H, CH), 3.78 (d, 2H, $OCH_2$), 6.78 (m, 1H, ArH), 7.24 (m, 2H, ArH), 7.26 (m, 2H, ArH), 7.54 (m, 2H, ArH), 7.93 (m, 2H, ArH), 7.53 to 8.97 (m, 4H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.1, 57.4, 65.5, 110.9, 116.8, 118.8, 120.9, 122.9, 124.5, 127.7, 128.4, 128.6, 128.8, 129.6, 131.8, 133.8, 136.8, 140.3, 142.2, 151.6, 153.2, 154.1. IR spectrum, (KBr) ν: 3121, 2899, 1585, 1190, 788. MS, m/z (% Rel. intensities): 482.03 ($M^+$), 483.04, 485.12, 481.99, 484.09, 486.01, 483.05, 485.11, 487.06, 482.04, 487.05, 484.04. Found, %: C, 62.15; H, 3.77; Cl, 22.05; N, 8.72; O, 3.31, Calculated, %: C, 62.19; H, 3.76; Cl, 22.03; N, 8.70; O, 3.31

**5,7-dichloro-8-((3-(4-methoxyphenyl)-1-phenyl-4,5-dihydro-1*H*-pyrazol-5-yl)methoxy)quinoline (6i).** Obtained from 5,7-dichloro-8-hydroxy quinoline (2.13 g, 10 mmol), potassium carbonate (2.07 g, 15 mmol, 1.5 eq), allyl chloride (4.0 ml, 50 mmol, 5 eq), 4-methoxy-phenyl hydrazone (2.76 g, 12 mmol, 1.2 eq), chloramine-T (3.37 g, 12 mmol, 1.2 eq), dry ethanol (30 ml). Yellow colored amorphous solid. Yield: 71% (3.42 g). $^1$H NMR (400 MHz, $CDCl_3$): δ 2.84 (d, 2H, $CH_2$), 3.14 (p, 1H, CH), 3.79 (d, 2H, $OCH_2$), 3.87 (s, 3H, $OCH_3$), 6.79 (m, 1H, ArH), 7.23 (m, 2H, ArH), 7.26 (m, 2H, ArH), 7.05 (m, 2H, ArH), 7.99 (m, 2H, ArH), 7.53 to 8.97 (m, 4H, 8HQ). $^{13}$C NMR (400 MHz, $CDCl_3$): δ 34.3, 55.7, 57.7, 65.7, 110.7, 114.3, 116.7, 118.2, 120.9, 122.5, 124.4, 127.9, 128.7, 128.9, 129.9, 131.8, 133.1, 140.5, 142.3, 151.8, 153.3, 154.4, 162.9. IR spectrum, (KBr) ν: 3121, 2899, 1585, 1203, 1190, 796. MS, m/z (% Rel. intensities): 476.91 ($M^+$), 478.99, 478.15, 480.10, 481.09, 479.11, 482.00, 481.10, 478.09. Found, %: C, 65.25; H, 4.45; Cl, 14.83; N, 8.78; O, 6.69, Calculated, %: C, 65.28; H, 4.42; Cl, 14.82; N, 8.78; O, 6.69.

## BIOLOGICAL ACTIVITY.

**Evaluation of anti-breast cancer activity of compounds 6(a-i).** This assay identifies the decrease of MTT by mitochondrial dehydrogenase to blue formazan product, which mirrors the standard working of mitochondria and hence the cell viability. Here, Human breast adenocarcinoma cells (MCF7)($2x10^3$) were scattered in each well of a 96-well plate and were permitted to adhere and spread for 24 hrs. Afterward, cells were treated with different concentrations (0.1, 1.0, 10.0 and 100 µg/mL) of test compounds prepared in 10% dimethyl sulphoxide (DMSO) and incubated at 37$^o$C for 24 hr. And *Doxorubicin* was taken as a positive control and the first column containing no drug taken as negative control. After a period of 24 hrs, added MTT solution (10µL of 10 mg/mL) to each cultured well and further incubated for 4 hrs to allow the formation of formazan crystals. And again added 100 µL of MTT solution and incubation continued for 12 hrs. The absorbance value was measured in each well at 570 nm with the help of an ELISA plate reader (ELx800, BioTek, VT, USA) and matched with untreated control. Finally cell viability of was calculated using the following formula:

Cell viability % = (Absorbance of sample/Absorbance of control) x100

Further $IC_{50}$ values were calculated as the concentrations that show 50% inhibition of proliferation on any tested cell line.

**Discussion on the anti-breast cancer activity of compounds 6(a-i):** The MTT cell proliferation assay has been widely considered as a trustworthy way of measuring the cell

proliferation rate and conversely when metabolic events leading to apoptosis or necrosis. Even though the number of screened compounds in the present study is limited, some structural features that are essential for the enlightenment of their cytotoxic effect can be discussed. As per the present study quinoline bearing pyrazole nucleus containing chlorine are more potent compared to other moieties. So it was projected that, the integration of chlorine to pyrazole scaffold may intensifies the anti-breast cancer activity than the chlorine atoms on to quinoline moiety. It is also confirmed by the less activity shown by the compound **6i** which contain two chlorine atoms on quinoline. The results obtained are reported in percentage survival of the cells when compared to that of the untreated control cells ± standard deviation in the Table 1. A comparison of the para substituent on the phenyl rings of pyrazole part and 5,7-position of quinoline scaffolds demonstrated that an electron withdrawing group on the pyrazole phenyl ring have better activity. A contrast with -Cl, -Br and -$OCH_3$ compounds containing -$OCH_3$ had least and –Cl substituents had good effects. Among nine compounds which are screened for cytotoxicity, compound **6b** emerged as a potent anticancer agent with $IC_{50}$ value 14 µg/mL. The rate of cytotoxicity of compounds increases in the following order **6h>6c>6g** with $IC_{50}$ values 18, 26, 29 µg/mL respectively. All other compounds showed low to moderate cytotoxicity. The $IC_{50}$ for the standard drug *Doxorubicin (DOX)* was found to be 18 µg/mL.

**Table 1.** MTT assay of the compounds **6(a-i).**

| Product | $R_1$ | $R_2$ | $R_3$ | Vehicle Control | 0.1 µg/mL | 10 µg/mL | 100 µg/mL | $IC_{50}$ µg/mL |
|---|---|---|---|---|---|---|---|---|
| **6a** | H | H | H | 100 ± 3.75 | 98.9 ± 7.92 | 82.4 ± 3.08 | 56.7 ± 2.15 | 99 |
| **6b** | H | H | Cl | 100 ± 6.91 | 75.1 ± 4.66 | 63.9 ± 2.33 | 11.9 ± 7.19 | 14 |
| **6c** | H | H | Br | 100 ± 7.34 | 80.3 ±2.30 | 83.9 ± 2.85 | 21.9 ± 2.32 | 24 |
| **6d** | H | H | $OCH_3$ | 100 ± 4.14 | 100 ± 7.50 | 98.9 ± 2.47 | 84.1 ± 4.90 | >100 |
| **6e** | Cl | H | H | 100 ± 4.48 | 100 ± 5.01 | 95.7 ± 4.98 | 43.7 ± 2.49 | 89 |
| **6f** | H | Cl | H | 100 ± 4.30 | 100 ± 2.17 | 61.3 ± 4.75 | 53.3 ± 7.99 | 92 |
| **6g** | Cl | Cl | H | 100 ± 6.83 | 64.9 ± 4.04 | 72.4 ± 4.55 | 27.9 ± 7.55 | 65 |
| **6h** | Cl | Cl | Cl | 100 ± 3.09 | 83.0 ± 5.07 | 76.7 ± 2.13 | 27.8 ± 2.11 | 18 |
| **6i** | Cl | Cl | $OCH_3$ | 100 ± 1.96 | 93.2 ±4.80 | 88.6 ± 3.01 | 42.3 ±2.48 | 77 |
| ***DOX*** | - | - | - | - | - | - | - | 18 |

**The values are expressed as mean ± SD of six separate experiments, *DOX = Doxorubicin.***

## 4. CONCLUSIONS

The attempt reveals an efficient path to prepare new pyrazole bearing quinoline scaffolds which were structurally confirmed by spectroscopic techniques (NMR, Mass and IR). All the derivatives were screened for their breast cancer chemotherapeutic activity against MCF7 cell line. The study reveals that the chlorine substituted pyrazole (**6b**) exhibits excellent anti-breast cancer activity. The cytotoxicity of obtained compounds increases in the order of **6b**>**6h**>**6c**> with $IC_{50}$ values 14, 18, 24 µg/mL respectively.

## 5. REFERENCES


[1] Nicholas J Robert., Neelima Denduluri., Patient Case Lessons: Endocrine Management of Advanced Breast Cancer, *Clinical Breast Cancer*, 18, 3, 192-204, **2017**

[2] Smith R A., Cokkinides V., Brawley O W., Cancer screening in the United States, 2009: a review of current American Cancer Society guidelines and issues in cancer screening, *A Cancer Journal for Clinicians*, 59(1), 27-41, **2009**

[3] Pinhel I., Hills M., Drury S., Salter J., Sumo G., A'Hern R., Bliss J M., Sestak I., Cuzick J., Barrett-Lee P., Harris A., Dowsett M., ER and HER2 expression are positively correlated in HER2 non-overexpressing breast cancer, *Breast Cancer Research*,14:R46, 1-12, **2012**

[4] Harvey J M., Clark G M., Osborne C K., Allred D C., Estrogen receptor status by immunohistochemistry is superior to the ligand-binding assay for predicting response to adjuvant endocrine therapy in breast cancer, *Journal of Clinical Oncology*, 17, 1474-1481, **1999**

[5] Howlader N., Altekruse S F., Li Cl., Chen V W., Clarke C A., Ries L A., Cronin K A., US incidence of breast cancer subtypes defined by joint hormone receptor and HER2 status, *Journal of the National Cancer Institute*, 28, 106(5), 1-8, **2014**

[6] Cancer.net. Breast cancer: overview, Available at: http:// www.cancer.net/cancer-types/breast-cancer/introduction. Accessed: June 21, **2018**

[7] Fagan D H., Fettig L M., Avdulov S., Beckwith H., Peterson M S., Ho Y Y., Wang F., Polunovsky V A., Yee D., Acquired Tamoxifen Resistance in MCF-7 Breast Cancer Cells Requires Hyperactivation of eIF4F-Mediated Translation, *Hormones and Cancer*, 8, 4, 219-229, **2017**

[8] Luu T., Chung C., Somlo G., Combining emerging agents in advanced breast cancer, *Oncologist*, 16, 760-771, **2011**

[9] Cardoso F., Fallowfield L., Costa A., Castiglione M., Senkus E., ESMO Guidelines Working Group. Locally recurrent or metastatic breast cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up, *Annals of Oncology*, 22(6), 25-30, **2011**

[10] National Comprehensive Cancer Network. Clinical Practice Guidelines in Oncology. Breast Cancer. v.1.2017, Available at: http://www.nccn.org/professionals/physician_gls/pdf/breast.pdf. Accessed: March 13, **2017**

[11] Cardoso F., Costa A., Norton L., Senkus E., Aapro M., Andre F., Barrios C H., Bergh J., Biganzoli L., Blackwell K L., Cardoso M J., Cufer T., El Saghir N., Fallowfield L., Fenech D., Francis P., Gelmon K., Giordano S H., Gligorov J., Goldhirsch A., Harbeck N., Houssami N., Hudis C., Kaufman B., Krop I., Kyriakides S., Lin U N., Mayer M., Merjaver S D., Nordstrom E B., Pagani O., Partridge A., Penault-Llorca F., Piccart M J., Rugo H., Sledge G., Thomssen C., Van't Veer L., Vorobiof D., Vrieling C., West N., Xu B., Winer E., ESO-ESMO 2nd international consensus guidelines for advanced breast cancer (ABC2), *Annals of Oncology*, 25, 1871-1888, **2014**

[12] National Comprehensive Cancer Network. Clinical Practice Guidelines in Oncology. Breast Cancer. v.1.2017, Available at: http://www.nccn.org/professionals/physician_gls/pdf/breast.pdf. Accessed: June 21, **2018**

[13] Lake D E., Hudis C., Aromatase inhibitors in breast cancer: an update, *Cancer Control*, 9, 490-498, **2002**

[14] Alluri P G., Speers C., Chinnaiyan A M., Estrogen receptor mutations and their role in breast cancer progression, *Breast Cancer Research*, 16, 6, 494, 1-8, **2014**

[15] Jeselsohn R., Yelensky R., Buchwalter G., Frampton G., Meric-Bernstam F., Gonzalez-Angulo A M., Ferrer-Lozano J., Perez-Fidalgo J

A., Cristofanilli M., Gomez H., Arteaga C L., Giltnane J., Balko J M., Cronin M T., Jarosz M., Sun J., Hawryluk M., Lipson D., Otto G., Ross J S., Dvir A., Soussan-Gutman L., Wolf I., Rubinek T., Gilmore L., Schnitt S., Come S E., Pusztai L., Stephens P., Brown M., Miller V A., Emergence of constitutively active estrogen receptor-alpha mutations in pretreated advanced estrogen receptor positive breast cancer, *Clinical Cancer Research*, 20(7), 1757-1767, **2014**

[16] Kam T S., Sim K M., Lim T M., Voastrictine, a novel pentacyclic quinolinic alkaloid from Tabernaemontana, *Tetrahedron Letters*, 42(28), 4721-4723, **2001**

[17] Muyard F., Bevalof F., Laube B., Vaquette J., Alkaloids from stem bark of *dutaillyea baudouinii*, *Phytochemistry*, 31, 3, 1087-1089, **1992**

[18] Fokialakis N., Magiatis P., Terzis A., Tillequin F., Skaltsounis A L., *Cyclomegistine*, the first alkaloid with the new cyclobuta[b]quinoline ring system from Sarcomelicope megistophylla, *Tetrahedron Letters*, 42, 5323-5325, **2001**

[19] Denis Zofou., Fidele Ntie-Kang., Wolfgang Sippl., Simon M N Efange., Bioactive natural products derived from the Central African flora against neglected tropical diseases and HIV, *Natural Product Reports*, 30, 8, 1098-1120, **2013**

[20] Mitaku S., Fokialakis N., Magiatis P., Tillequin F., Alkaloids from Sarcomelicope megistophylla, *Fitoterapia*., 78, 2, 169-170, **2007**

[21] Xiong T., Zhang Q., Zhang Z., Liu Q., A Divergent Synthesis of γ-Iminolactones, Dihydroquinolin-2-ones, and γ-Lactames from β-Hydroxymethylcyclopropanylamides, *The Journal of Organic Chemistry*, 71, 8005-8009, **2006**

[22] Jianguang Zhou., Nabi A Magomedov., Explorations on the Asymmetric Total Synthesis of Isoschizogamine, *The Journal of Organic Chemistry*, 72, 3808-3815, **2007**

[23] Magomedov N A., Efficient construction of cyclopenta[b]quinoline core of isoschizozygane alkaloids via intramolecular formal hetero-Diels-Alder reaction, *Organic Letters*, 5, 2509-2512, **2003**

[24] Verma G K., Raghuvanshi K., Kumar R., Singh M S., An efficient one-pot three-component synthesis of functionalized pyrimido [4, 5-b] quinolines and indeno fused pyrido [2, 3-d] pyrimidines in water, *Tetrahedron Letters, 53*, 399-402, **2012**

[25] Phillips J P., The Reactions Of 8-Quinolinol, *Chemical Reviews*, 56 (2), 271–297, **1956**

[26] Mahamoud A., Chevalier J., Davin-Regli A., Barbe J., Pages J-M., Quinoline derivatives as promising inhibitors of antibiotic efflux pump in multidrug resistant Enterobacter aerogenes isolates, *Current Drug Targets*, 7, 843-847, **2006**

[27] Xiao Z, Lei F, Chen X, Wang X, Cao L, Ye K, Zhu W, Xu S., Design, synthesis, and antitumor evaluation of quinoline-imidazole derivatives. *Arch Pharm*, 6, 351 (6), e1700407, **2018**

[28] Chaaban I, Rizk OH Ibrahim TM, Henen SS, El-Khawass EM, Bayad AE, El-Ashmawy, Nematalla HA, Synthesis, anti-inflammatory screening, molecular docking, and COX-1,2/-5-LOX inhibition profile of some novel quinoline derivatives, *Bioorganic Chemistry*,78, 220–235, **2018**

[29] AAM Abdel-Aziz, AS El-Azab, LA Abou-Zeid, KEH ElTahir, NI Abdel-Aziz, RR Ayyad, AM Al-Obaid, *Eur. J. Med. Chem*. 115, 121–131, **2016**.

[30] Zhang S L., Zhai X., Zhang S J., Yu H H., Gong P., Synthesis and cytotoxicity studies of quinoline-3-carbonitrile derivatives, *Chinese Chemical Letters*, 21, 939-942, **2010**

[31] Berger D M., Dutia M., Powell D., Floyd M B., Torres N., Mallon R., Wojciechowicz D., Kim S., Feldberg L., Collins K., Chaudhary I., 4-Anilino-7-alkenylquinoline-3-carbonitriles as potent MEK1 kinase inhibitors, *Bioorganic and Medicinal Chemistry*, 16, 9202-9211, **2008**

[32] Tsou H R., Overbeek-Klumpers E G., Hallett W A., Reich M F., Floyd M B., Johnson B D., Michalak R S., Nilakantan R., Discafani C., Golas J., Rabindran S K., Shen R., Shi X., Wang Y F., Upeslacis J., Wissner A., Optimization of 6,7-disubstituted-4-(arylamino)quinoline-3-carbonitriles as orally active, irreversible inhibitors of human epidermal growth factor receptor-2 kinase activity, *Journal of Medicinal Chemistry*, 48, 1107-1131, **2005**

[33] Green N., Hu Y., Janz K., Li H Q., Kaila N., Guler S., Thomason J., Joseph- McCarthy D., Tam S Y., Hotchandani R., Wu J., Huang A., Wang Q., Leung L., Pelker J., Marusic S., Hsu S., Telliez J B., Hall J P., Cuozzo J W., Lin L L., Inhibitors of tumor progression loci-2 (Tpl2) kinase and tumor necrosis factor alpha (TNF-alpha) production: selectivity and in vivo antiinflammatory activity of novel 8-substituted-4-anilino-6-aminoquinoline-3-carbonitriles, *Journal of Medicinal Chemistry*, 50, 19, 4728-4745, **2007**

[34] Anam Ansari., Abad Ali., Mohd Asif., Shamsuzzaman., Review: biologically active pyrazole derivatives, *New Journal of Chemistry*, 41, 16-41, **2017**

[35] Heba A Elhady, Refat El-Sayed and Hamedah S Al-nathali, Design, synthesis and evaluation of anticancer activity of novel 2-thioxoimidazolidin-4-one derivatives bearing pyrazole, triazole and benzoxazole moieties, *Chemistry Central Journal*, 12, 51, https://doi.org/10.1186/s13065-018-0418-1, **2018**

[36] Nauduri D., Reddy G B., Antibacterials and antimycotics: Part 1: Synthesis and activity of 2-pyrazoline derivatives, *Chemical and Pharmaceutical Bulletin (Tokyo)*, 46, 1254-1260, **1998**

[37] B'Bhatt H, Sharma S. Synthesis and antimicrobial activity of pyrazole nucleus containing 2-thioxothiazolidin-4-one derivatives. *Arab. J. Chem.* 10, S1590–S1596, **2017**.

[38] Xia Y., Dong Z W., Zhao B X., Ge X., Meng N., Shin D S., Miao J Y., Synthesis and structure-activity relationships of novel 1-arylmethyl-3-aryl-1H-pyrazole-5-carbohydrazide derivatives as potential agents against A549 lung cancer cells, *Bioorganic & Medicinal Chemistry*, 15, 6893-6899, **2007**

[39] Youssef M M., Mohammed S F., Kotb E R., Salma M A., A novel synthesis of some new pyrimidine, thiazolopyrimidine and pyrazole derivatives using diarylepoxypropanones as precursors, *World Journal of Chemistry*, 4, 149-156, **2009**

[40] Ansari A, Ali A. Asif M. Biologically active pyrazole derivatives. *New J. Chem.*, 41, 16–41, doi:10.1039/C6NJ03181A, **2017**

[41] Nagwa Mohamed Mahrous Hamada., Nadia Yousef Megally Abdo., Synthesis, Characterization, Antimicrobial Screening and Free-Radical Scavenging Activity of Some Novel Substituted Pyrazoles, *Molecules*, 20(6), 10468-10486, **2015**

[42] Yang J F., Cao H., Liu H., Li B Q., Ma Y M., Synthesis and Bioactivity of Novel Bis-heterocyclic Compounds Containing Pyrazole and Oxadiazoline, *Journal of the Chinese Chemical Society*, 58, 369–375, **2011**

[43] Mallikarjuna Rao R., Sreeramulu J., Ravindranath L K., Nagaraja Reddy G., Hanumanthurayudu K., Nageswara Reddy G., Jayaraju A., Madhusudhan P., Synthesis and biological screening of some Pyridine and Pyrrole Derivatives of Pyrazolo [3,4-*c*] pyrazoles, *Journal of Chemical and Pharmaceutical Research*, 4, 272–278, **2012**

[44] Mohareb R M., El-Sayed N N E., Abdelaziz M A., Uses of Cyanoacetylhydrazine in Heterocyclic Synthesis: Novel Synthesis of Pyrazole Derivatives with Anti-tumor Activities, *Molecules*, 17, 8449–8463, **2012**

[45] Kumar K A., Jayaroopa P., Pyrazoles: Synthetic Strategies and Their Pharmaceutical Applications-An Overview, *International Journal of PharmTech Research*, 5, 1473–1486, **2013**

[46] Piste P B., Facile Synthesis and Antimicrobial Screening of Pyrazole Derivatives, *World Journal of Pharmaceutical Research*, 3, 735–742, **2014**

[47] Kamal A., Shaik A B., Polepalli S., Reddy V S., Kumar G B., Gupta S., Krishna K V S R., Nagabhushana A., Mishra R K., Jain N., Pyrazole-Oxadiazole Conjugates: Synthesis, Antiproliferative Activity and Inhibition of Tubulin Polymerization, *Organic and Biomolecular Chemistry*, 12, 7993–8007, **2014**

[48] Bhalla A., Bari S S., Bhalla J., Synthesis of Novel Pyrazolylmethylene-Pyrimidine Heterocycles:Potential Synthons for Hybrid β-Lactams, *Canadian Chemical Transactions*, 3, 72–84, **2015**

[49] Abdelhamid A O., Zohdi H F., Sallam M M M., Ahmed N A., Reactions with Hydra-zonoyl Halides. 31. Synthesis of Some New Pyrrolidino [3,4-*c*] pyrazolines, Pyrazoles, and Pyrazolo [3,4-*d*] pyridazines, *Molecules*, 5, 967–973, **2000**

[50] Helene Pellissier., Asymmetric 1,3-dipolar cycloadditions, *Tetrahedron*, 63, 3235–3285, **2007**

[51] Fabrice Eckes., Emmanuel Deiters., Adrien Metivet., Veronique Bulach., Mir Wais Hosseini., Synthesis and Structural Analysis of Porphyrin-Based Polynucleating Ligands Bearing 8-Methoxy- and 8-

(Allyloxy)quinoline Units, *European Journal of Organic Chemistry*, 2011, 13, 2531–2541, **2011**

[52] Srikantamurthy N., Umesha K B., Shubakara K., Chethan J., Novel pyrazoline amidoxime and their 1, 2, 4-oxadiazole analogues: Synthesis and pharmacological screening, *Bioorganic & Medicinal Chemistry Letters*, 23(16), 4532-4539, **2013**

[53] Chandra., Srikantamurthy N., Umesha K B., Jeyaseelan S., Mahendra M., (*E*)-N′-Hydroxy-1, 3-diphenyl-4, 5-dihydro-1*H*-pyrazole-5-carboximidamide, *Acta Crystallographica Section E: Structure Reports Online*, E68(6), 1661-1662, **2012**

[54] Sureshbabu Dadiboyena., Ashton T Hamme I I., Environmentally Benign Lewis Acid Promoted [2+3] Dipolar Cycloaddition Reactions of Nitrile Imines with Alkenes in Water, *European Journal of Organic Chemistry*, 33, 7567–7574, **2013**

[55] Srikantamurthy Ningaiah., Shridevi D Doddaramappa., Chandra., Mahendra Madegowda., Shubhakara Keshavamurthy., Umesha K Bhadraiah., One-pot tandem synthesis of tetrasubstituted pyrazoles via 1,3-dipolar cycloaddition between aryl hydrazones and ethyl but-2-ynoate, *Synthetic communication*, 44, 2222-2231, **2014**

[56] Marco Chiericato., Piero Dalla Croce., Germano Carganico., Stefano Maiorana., 1,3-Dipolar cycloadditions. Reactions of nitrile imines with 1,2-disubstituted alkenes, *Journal of Heterocyclic Chemistry*, 16, 383-384, **1979**

[57] Aixue Sun., Jia Hai Ye., Haitao Yu., Wenchao Zhang., Xiaolong Wang., A new efficient synthesis of pyrazoles from hydrazonoyl halides and β-oxophosphonates, *Tetrahedron Letters*, 55, 4, 889-892, **2014**

## 6. ACKNOWLEDGEMENTS

The author KBU is grateful to the University Grant Commission, New Delhi, India for providing financial assistance (2162-MRP/15-16/KAMY002/UGCSWRO) to carry out the research.