Available online at www.derpharmachemica.com

ISSN 0975-413X
CODEN (USA): PCHHAX

Der Pharma Chemica, 2023, 15(5): 59-66
(http://www.derpharmachemica.com/archive.html)

Synthesis, Characterisation and Anti-Breast Cancer and Antibacterial Evaluation of Novel 2-Phenylquinoline Amide Derivatives

Kailas W. Shinde*

Department of chemistry, Wilson College, Mumbai, India

***Corresponding author:** Dr. Kailas W. Shinde, Department of chemistry, Wilson College, Mumbai, India, **E-mail**: kailasshinde44@gmail.com*

Received: 13-June-2023, Manuscript no: dpc-23-102417, **Editor assigned**: 19-June-2023, PreQC No: dpc-23-102417, **Reviewed**: 17-July-2023, QC No: dpc-23-102417, **Revised**: 02-August-2023, Manuscript No: dpc-23-102417, **Published**: 30-September-2023, **DOI**: 10.4172/0975-413X.15.5.59-66

ABSTRACT

*A series of new class of 2-phenylquinoline amide derivatives **(5a-5j)** were synthesized from 5-nitroquinoline through multi-step reactions in good to moderate yields. All the compounds were confirmed by spectral characterization viz FT-IR, MS, ^{1}H-NMR and ^{13}C-NMR. All the molecules were evaluated for their anticancer activity against a breast cancer cell line, MDA-MB-231 by MTT assay and antibacterial activity against Gram-positive bacteria (Staphylococcus aureus 6538p and Bacillus subtilis) and Gram-negative bacteria (Escherichia coli and Pseudomonas aeruginosa) using agar well diffusion method. All the compounds exhibited significant anticancer activity as compared to standard cisplatin but not comparable to doxorubicin HCl. Compound **5g** exhibited better promising anti-breast cancer activity. Compounds **5d**, **5h**, **5i** and **5j** exhibited moderate antibacterial activity against all the tested organisms among all the synthesized analogues as compared to standard streptomycin. Based on the results, further modification and optimization of these compounds may be useful to develop potent anticancer and antibacterial drug.*

Keywords: Antibacterial; Anticancer; MDA-MB-231; 2-phenylquinoline amide

INTRODUCTION

Heterocyclic compounds compose the major and most diverse class of organic compounds and a significant number of synthetic and natural heterocyclic molecules are highly potent and are in therapeutic use. Heterocyclic compound is the heart of medicinal chemistry and lot of work have been done in this area but still there is scope to discover new entities. Among various pharmacologically important heterocyclic compounds, quinoline and its derivatives are considered as a privileged structure and occupy an exclusive place in medicinal chemistry and they are also very

important from industrial and biological perspective as they are very helpful in understanding life processes. Quinoline moiety has exhibited considerable application in pharmaceutical chemistry [1-3] and their wide-ranging occurrence in natural products such as camptothecin[4,5] and luotonin A [6] Synthetic members of quinoline are also an important and attracting attention of medicinal chemists in view of their profound range of biological activities such as antitubercular [7] anti-inflammatory [8] anti-platelet [9] antibacterial [10,11] anti-asthmatic [12] anticancer [13-14] antihypertensive [15] antimalarials [16] antifungals [17] and antiproliferative [18] Thus, we addressed our attention to synthesize novel quinoline derivatives by different modifications in their structure and explore the possibility of these derivatives becoming a potential candidate for the discovery of novel therapeutic drugs with enhanced biological activity.

During our literature survey, it is also observed that, the incorporation of amide group into quinoline nucleus enhances its biological activity. For instance, some quinoline-4-carboxamides were described as potent inhibitors of prolyl-t-RNA synthetase [19] Certain novel quinoline amides exhibited significant cytotoxic activity against breast cancer cell line MCF-7 [20] Some quinoline benzamides were reported as VEGFR-2 inhibitors [21] Certain quinoline-8-amide derivatives were found to be good inhibitors of human tankyrases [22] 2-Phenylquinoline-4-amides were also reported as STAT3 Inhibitors which provide a new therapeutic approach for cancer treatment [23]Various 2-phenyl-quinoline derivatives showed significant antibacterial activity [24-27] 6-methoxy-2-arylquinolines were reported as potential P-glycoprotein inhibitors [28] Certain 4-(imidazolylmethyl)-2-aryl-quinoline derivatives were described as aromatase inhibitors and anti-breast cancer agents [29].

In the light of these facts, we planned to synthesize a novel series of 2-phenylquinoline amide derivatives and to evaluate their anticancer activity against a breast cancer cell line (MDA-MB-231) by MTT assay method and antibacterial activity against Gram-positive bacteria (*Staphylococcus aureus 6538p* and *Bacillus subtilis*) and Gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*) by agar well diffusion method. The structure activity relationship (SAR) studies were also performed for all the bioactivities taken into account.

MATERIALS AND METHODS

Materials (2.1)

Chemistry (2.1.1)

All commercial chemicals and solvents are of LR-grade and AR-grade and were used without further purification. The thin layer chromatography (TLC) was performed on Merck pre-coated silica gel 60 F_{254} plates, with visualization under UV light. Melting points were determined with PEW-340MP melting point apparatus and are uncorrected. ^{1}H-NMR spectra were recorded on Bruker 400 MHz and ^{13}C-NMR spectra on Bruker 75 and 100 MHz AVANCE instruments, respectively and *J* values in Hertz and chemical shifts (δ) in ppm were reported relative to internal standard tetramethylsilane (TMS). FT-IR spectra (ν in cm^{-1}) using KBr discs were recorded on Perkin-Elmer FT-IR spectrophotometer. The mass spectra (MS) were measured with Thermo Finnigan-TSQ Quarter Ultra (triple Quad). The purity of all the compounds was determined by HPLC (Waters 2695 Alliance) using Kromasil C_{18} column (250 mm X 4.5 mm, 5 µ), with mobile phase containing ACN and buffer (0.01 M ammonium acetate + 0.5% triethylamine, pH 5.0, adjusted with acetic acid).

Anticancer Activity (2.1.2)

Cancer cell line MDA-MB-231 (breast adenocarcinoma) was purchased from National Centre for Cell Sciences, Pune, India. 3-(4,5-Dimethyl thiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT), Tris-HCl were obtained from SRL (Mumbai, India), Fetal bovine serum (FBS), Phosphate buffered saline (PBS), Dulbecco's modified eagle's medium (DMEM) and Trypsin-EDTA were obtained from CellClone (Delhi, India), antibiotics from Hi-Media Laboratories Ltd. (Mumbai, India).

Antibacterial Activity (2.1.3)

The Gram-positive organisms viz. *Bacillus subtilis* and *Staphylococcus aureus 6538p* and Gram-negative organisms viz. *Pseudomonas aeruginosa* and *Escherichia coli* cultures were obtained from neighbouring hospitals and pathological laboratories located in Mumbai.

METHOD

Chemistry (2.2.1)

In the present work, a series of novel 2-phenylquinoline amide derivatives (5a-5j) were synthesized from 5-nitroquinoline through multi-step reactions in good to moderate yields.

Procedure for the Synthesis of 5-nitroquinoline-N-oxide (2.2.2)

To a solution of 5-nitroquinoline (1 g, 5.7 mmol,1 eq.) in chloroform (10 mL), m-CPBA (2.47 g, 14.3 mmol, 2.5 eq.) was added and the mixture was stirred at room temperature for 12 h. Completion of the reaction was monitored by TLC in ethyl acetate- petroleum ether (2:8). The reaction mixture was then poured into water (10 mL) and neutralised with saturated sodium bicarbonate solution (25 mL). The aqueous mixture was extracted with dichloromethane (3X25 mL), the combined organic phases were dried (Na_2SO_4) and concentrated in vacuum. The crude product was purified by silica gel (100-200 mesh) flash column chromatography (10% Ethyl acetate/petroleum ether) to obtain the compound 1.

Yellow solid; Yield 85 %; mp 164-165 °C; ^{1}H NMR (DMSO-d_6, 400MHz, δ ppm): 8.25 (d, *J* = 8.8 Hz, 1H), 8.18 (d, *J* = 9.2 Hz, 1H), 7.95 (d, *J* = 7.2 Hz, 1H), 7.85 (dd, J_1 = 7.2 Hz, J_2 = 6.8 Hz, 1H), 7.77 (d, *J* = 8.8 Hz, 1H), 7.60 (dd, J_1 = 8.4 Hz, J_2 = 6.0 Hz, 1H); ^{13}C NMR (DMSO-d_6, 100MHz, δ ppm): 148.63, 142.86, 136.74, 132.78, 131.55, 127.27, 125.57, 123.38, 121.08; IR (KBr) ν_{max}/cm^{-1}: 3055, 1665, 1596, 1489, 1389, 1271; MS (ESI): *m/z* $[M+H]^+$ = 191.17; HPLC: 95.78%.

Procedure for the Synthesis of 2-Chloro-5-nitroquinoline (2.2.3)

Sulfuryl chloride (2.5 mL, 26.2 mmol, 10 eq.) was added to 5-nitroquinoline-N-oxide (1) (0.5 g, 2.6 mmol, 1 eq.) and the mixture was refluxed at 60 ^oC for 6 h. Completion of the reaction was monitored by TLC in ethyl acetate-petroleum ether (2:8). The reaction mixture was then cooled and dichloromethane (20 mL) and water (20 mL) were added and the mixture was stirred for 1 h to dissolve any insoluble material. The layers were separated and the organic phase was dried (Na_2SO_4) and concentrated in vacuum and the crude product was purified by silica gel (100-200 mesh) flash column chromatography (10% Ethyl acetate/petroleum ether) to obtain the compound 2.

Pale yellow solid; Yield 42 %; mp 134 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 8.99 (d, *J* = 9.2 Hz, 1H), 8.33-8.42 (m, 2H), 7.80-7.88 (m, 1H), 7.63 (d, *J* = 9.2 Hz, 1H); ^{13}C NMR (DMSO-d_6, 100MHz, δ ppm): 152.85, 151.18, 148.68, 141.27, 139.88, 135.23, 132.01, 126.11, 119.82; IR (KBr) ν_{max}/cm^{-1}: 3096, 1662, 1558, 1470, 1399, 788; MS (ESI): *m/z* $[M+H]^+$ = 208.60, *m/z* $[(M+2)-H]^+$ = 210.60; HPLC: 97.74%.

Procedure for the synthesis of 5-nitro-2-phenylquinoline (2.2.4)

A solution of 2-chloro-5-nitroquinoline (2) (500 mg, 2.4 mmol, 1 eq.) and phenylboronic acid (440 mg, 3.6 mmol, 1.5 eq.) in 1:1 mixture of toluene/ethanol (10 mL) was degassed under reduced pressure and flushed with nitrogen. To this suspension, anhydrous caesium carbonate (1.5 g, 4.8 mmol, 2 eq.) and tetrakis(triphenylphosphine) palladium (0) (140 mg, 0.12 mmol, 0.05 eq.) was added and the system was degassed again. The reaction mixture was heated under reflux for 8 h. Completion of the reaction was monitored by TLC in ethyl acetate-petroleum ether (2:8). The reaction mixture was then allowed to cool to room temperature and filtered through celite. The filter cake was washed with ethyl acetate (3X25 mL) and the organic layer of the filtrate was separated, washed with brine, dried (Na_2SO_4) and concentrated in vacuum. The resulting residue was purified by silica gel (100-200 mesh) flash column chromatography (10 % Ethyl acetate/petroleum ether) to obtain the compound 3.

Yellow solid; Yield 76 %; mp 130-132 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 8.91 (d, *J* = 9.2 Hz, 1H), 8.41-8.47 (m, 3H), 8.31 (d, *J* = 7.2 Hz, 2H), 7.95 (t, *J* = 8.0 Hz, 1H), 7.55-7.60 (m, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 155.45, 146.64, 141.13, 134.86, 133.64, 130.02, 129.95, 129.07, 127.57, 126.78, 124.08, 121.72, 120.98; IR (KBr) ν_{max}/cm^{-1}: 3087, 1596, 1520, 1489, 1462; MS (ESI): *m/z* $[M+H]^+$ = 251.10; HPLC: 99.72%.

Procedure for the synthesis of 2-phenylquinolin-5-amine (2.2.5)

To a solution of 5-nitro-2-phenylquinoline (3) (500 mg, 2.0 mmol, 1 eq.) in ethyl acetate (5 mL), stannous chloride dihydrate (1.2 g, 5.0 mmol, 2.5 eq.) was added and the mixture was stirred at room temperature for 12 h. Completion of the reaction was monitored by TLC in ethyl acetate-petroleum ether (4:6). The reaction mixture was poured into water (10 mL) and neutralised with saturated sodium bicarbonate solution. The aqueous mixture was extracted with ethyl acetate (3X25 mL), the combined organic phases were dried (Na_2SO_4) and concentrated in vacuum. The crude product was purified by silica gel (100-200 mesh) flash column chromatography (20 % Ethyl acetate/petroleum ether) to obtain the compound 4.

Yellow solid; Yield 47 %; mp 142-144 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 8.90 (d, *J* = 8.8 Hz, 1H); 8.41-8.46 (m, 3H), 8.31 (d, *J* = 7.2 Hz, 2H), 7.54-7.59 (m, 3H), 6.97 (dd, J_1 = 6.8 Hz, J_2 = 1.6 Hz, 1H), 6.42 (s, 2H); ^{13}C-NMR (DMSO-d_6, 75 MHz, δ ppm): 153.40, 145.18, 141.67, 139.03, 134.02, 131.26, 131.18, 129.32, 129.18, 127.87, 127.29, 119.02, 118.03; IR (KBr) ν_{max}/cm^{-1}: 3461, 3341, 1633, 1592, 1465, 1358; MS (ESI): *m/z* $[M+H]^+$ = 221.30; HPLC: 95.55%.

General procedure for the synthesis of compounds 5a-5j (2.2.6)

To a solution of 2-phenylquinolin-5-amine **(4)** (500 mg, 2.3 mmol, 1eq.) in THF (5 mL), respective acid chloride (3.5 mmol, 1.5 eq.) and sodium

hydride (70 mg, 2.8 mmol, 1.2 eq.) was added at 0 ^{o}C and the reaction mixture was then stirred at room temperature for 1 h. Completion of the reaction was monitored by TLC in ethyl acetate-petroleum ether (4:6). The reaction mixture was then poured into ice cold water (10 mL) and extracted with ethyl acetate (3X25 mL), the combined organic phases were dried (Na_2SO_4) and concentrated in vacuum. The crude product was purified by silica gel (100-200 mesh) flash column chromatography (20 % Ethyl acetate/petroleum ether) to obtain the final compounds **5a-5j**.

Synthesis of N-(2-phenylquinolin-5-yl)benzamide (5a)

Yellow solid; Yield 78 %; mp 224-226 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.60 (s, 1H), 8.48 (d, J = 8.8 Hz, 1H), 8.30 (dd, J_1 = 7.2 Hz, J_2 = 1.2 Hz, 2H), 8.18 (d, J = 8.8 Hz, 1H), 8.12 (dd, J_1 = 6.8 Hz, J_2 = 1.2 Hz, 2H), 8.02 (d, J = 8.8 Hz, 1H), 7.83 (t, J = 7.6 Hz, 1H), 7.70 (d, J = 7.2 Hz, 1H), 7.65 (d, J = 7.2 Hz, 1H), 7.56-7.61 (m, 4H), 7.54 (d, J = 6.8 Hz, 1H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.32, 155.86, 146.85, 143.26, 141.87, 137.34, 133.03, 132.86, 131.34, 129.74, 129.53, 128.87, 128.61, 128.28, 126.11, 125.08, 120.85, 120.62; IR (KBr) ν_{max}/cm^{-1}: 3225, 3059, 1645, 1597, 1512, 1485; MS (ESI): m/z $[M+H]^+$ = 325.50; HPLC: 100%.

Synthesis of 2-methoxy-N-(2-phenylquinolin-5-yl) benzamide (5b)

Brown solid; Yield 80 %; mp 154-156 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.44 (s, 1H), 8.56 (d, J = 8.8 Hz, 1H), 8.24-8.31 (m, 3H), 7.96 (d, J = 8.4 Hz, 2H), 7.78-7.85 (m, 2H), 7.52-7.60 (m, 4H), 7.28 (d, J = 8.0 Hz, 1H), 7.13 (t, J = 7.6 Hz, 1H), 4.06 (s, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.52, 159.16, 156.85, 147.26, 141.71, 138.34, 133.83, 132.56, 130.34, 129.72, 129.55, 128.87, 128.60, 128.28, 127.19, 124.38, 123.48, 123.32, 120.67, 118.47, 55.87; IR (KBr) ν_{max}/cm^{-1}: 3372, 3026, 1666, 1619, 1551, 1481, 1232, 1015; MS (ESI): m/z $[M+H]^+$ = 355.60; HPLC: 98.96%.

Synthesis of 3-methoxy-N-(2-phenylquinolin-5-yl)benzamide (5c)

Green solid; Yield 74 %; mp 212-214 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.61 (s, 1H), 8.38 (d, J = 8.0 Hz, 1H), 8.21 (d, J = 8.0 Hz, 2H), 8.11 (d, J = 8.0 Hz, 1H), 7.55-7.58 (m, 2H), 7.46-7.53 (m, 7H), 7.17-7.20 (m, 1H), 3.84 (s, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.52, 159.68, 155.85, 147.26, 141.71, 136.68, 134.11, 132.56, 130.14, 129.72, 129.55, 128.87, 128.60, 127.32, 126.23, 123.81, 123.64, 121.58, 118.36, 116.20, 55.64; IR (KBr) ν_{max}/cm^{-1}: 3221, 3059, 1647, 1598, 1508, 1486, 1233, 1033; MS (ESI): m/z $[M+H]^+$ = 355.40; HPLC: 100%.

Synthesis of 4-methoxy-N-(2-phenylquinolin-5-yl)benzamide (5d)

Pale yellow solid; Yield 82 %; mp 194-196 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.68 (s, 1H), 8.49 (d, J = 8.8 Hz, 1H), 8.02-8.07 (m, 4H), 7.89 (m, 1H), 7.84 (d, J = 8.6 Hz, 1H), 7.77 (d, J = 6.8 Hz, 1H), 7.56-7.67 (m, 1H), 7.51-7.52 (m, 3H), 7.02-7.04 (m, 2H), 3.85 (s, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.87, 160.34, 154.85, 148.26, 141.71, 134.11, 131.90, 131.71, 130.70, 129.40, 129.28, 128.54, 128.36, 127.32, 126.23, 121.92, 121.23, 120.90, 55.52; IR (KBr) ν_{max}/cm^{-1}: 3214, 3087, 1640, 1621, 1509, 1486, 1250, 1032; MS (ESI): m/z $[M-H]^-$ = 353.10; HPLC: 99.86%.

Synthesis of 2-chloro-N-(2-phenylquinolin-5-yl)benzamide (5e)

Yellow solid; Yield 76 %; mp 220-222 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.78 (s, 1H), 8.59-8.61 (m, 3H), 8.25 (m, 3H), 8.18 (d, J = 8.0 Hz, 2H), 8.02-8.04 (m, 3H), 7.53-7.58 (m, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.45, 158.06, 146.92, 141.21, 140.95, 138.64, 134.95, 130.25, 130.13, 129.86, 128.19, 127.57, 127.31, 125.15, 124.58, 123.32, 121.76, 120.18, 118.02, 116.89; IR (KBr) ν_{max}/cm^{-1}: 3243, 3092, 1656, 1633, 1534, 1482, 777; MS (ESI): m/z $[M+H]^+$ = 358.25, m/z $[(M+2)-H]^+$ = 360.25; HPLC: 99.04%.

Synthesis of 3-chloro-N-(2-phenylquinolin-5-yl)benzamide (5f)

Brown solid; Yield 76 %; mp 230-232 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.70 (s, 1H), 8.48 (d, J = 8.6 Hz, 1H), 8.05-8.07 (m, 2H), 7.96-7.98 (m, 1H), 7.90-7.91 (m, 1H), 7.83 (d, J = 8.6 Hz, 1H), 7.78-7.81 (m, 2H), 7.66 (m, 1H), 7.51-7.59 (m, 4H) 7.45-7.46 (m, 1H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 164.97, 157.54, 148.89, 143.03, 141.38, 137.32, 133.08, 130.29, 129.94, 129.81, 128.07, 127.83, 127.54, 126.05, 124.13, 123.25, 122.08, 121.62, 119.90, 116.97; IR (KBr) ν_{max}/cm^{-1}: 3243, 3086, 1649, 1597, 1525, 1481, 777; MS (ESI): m/z $[M+H]^+$ = 358.20, m/z $[(M+2)-H]^+$ = 360.20; HPLC: 99.35%.

Synthesis of 4-chloro-N-(2-phenylquinolin-5-yl)benzamide (5g)

Yellow solid; Yield 80 %; mp 234-236 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.66 (s, 1H), 8.48 (d, J = 8.8 Hz, 1H), 8.29 (d, J = 6.8 Hz, 2H),

8.17 (d, J = 9.2 Hz, 1H), 8.12 (d, J = 8.4 Hz, 2H), 8.02 (d, J = 8.4 Hz, 1H), 7.82 (dd, J_1 = 8.4 Hz, J_2 = 7.6 Hz, 1H), 7.65-7.70 (m, 3H), 7.52-7.59 (m, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.33, 155.85, 147.26, 141.71, 136.68, 134.11, 132.10, 131.92, 130.44, 129.90, 129.68, 128.64, 128.06, 126.48, 124.32, 123.82, 123.63, 118.84; IR (KBr) ν_{max}/cm^{-1}: 3193, 3077, 1637, 1606, 1515, 1481, 779; MS (ESI): m/z $[M+H]^+$ = 358.33, m/z $[(M+2)-H]^+$ = 360.33; HPLC: 98.41%.

Synthesis of 2-nitro-N-(2-phenylquinolin-5-yl)benzamide (5h)

Brown solid; Yield 72 %; mp 278-280 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 11.00 (s, 1H), 8.77 (d, J = 8.8 Hz, 1H), 8.27-8.31 (m, 3H), 8.22 (d, J = 8.0 Hz, 1H), 8.12 (t, J = 7.2 Hz, 1H), 7.89-8.00 (m, 4H), 7.82 (t, J = 7.2 Hz, 1H), 7.57-7.63 (m, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 164.10, 157.45, 148.72, 141.06, 141.02, 138.64, 134.13, 132.86, 130.54, 130.32, 129.95, 129.77, 127.57, 126.78, 124.08, 121.72, 120.98, 118.87, 118.72, 116.98; IR (KBr) ν_{max}/cm^{-1}: 3223, 3090, 1674, 1644, 1593, 1525, 1492; MS (ESI): m/z $[M+H]^+$ = 370.12; HPLC: 99.54%.

Synthesis of 3-nitro-N-(2-phenylquinolin-5-yl)benzamide (5i)

Pale yellow solid; Yield 74 %; mp 254-256 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.99 (s, 1H), 8.87 (m, 1H), 8.48 (d, J = 8.0 Hz, 2H), 8.22 (m, 3H), 8.12 (d, J = 8.0 Hz, 2H), 8.06 (m, 2H), 7.88 (m, 1H), 7.52-7.58 (m, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 165.68, 159.34, 151.85, 141.26, 140.71, 136.23, 134.10, 133.90, 130.95, 129.90, 129.68, 128.94, 128.66, 127.42, 126.23, 123.32, 123.23, 120.32, 118.68, 116.36; IR (KBr) ν_{max}/cm^{-1}: 3238, 3063, 1649, 1619, 1529, 1520, 1494; MS (ESI): m/z $[M+H]^+$ = 370.40; HPLC: 99.66%.

Synthesis of 4-nitro-N-(2-phenylquinolin-5-yl)benzamide (5j)

Brown solid; Yield 78 %; mp 250-252 °C; ^{1}H-NMR (DMSO-d_6, 400 MHz, δ ppm): 10.96 (s, 1H), 8.51 (d, J = 8.0 Hz, 1H), 8.37-8.39 (m, 3H), 8.27 (m, 3H), 8.25 (m, 1H), 8.21 (d, J = 8.0 Hz, 2H), 8.15 (m, 1H), 7.54-7.57 (m, 3H); ^{13}C-NMR (DMSO-d_6, 100 MHz, δ ppm): 164.86, 158.32, 152.05, 147.01, 142.66, 142.35, 138.79, 132.52, 131.41, 129.63, 129.41, 129.19, 128.24, 128.14, 127.22, 126.15, 123.24, 118.19; IR (KBr) ν_{max}/cm^{-1}: 3249, 3086, 1656, 1650, 1600, 1525, 1482; MS (ESI): m/z $[M+H]^+$ = 370.30; HPLC: 98.50%.

MTT assay (2.2.7)

MTT assay was performed as reported previously [30] Briefly, cells were grown in DMEM media supplemented with fetal bovine serum (FBS) 10 % (50 µg/mL) and penicillin-streptomycin (50 µg/mL) at 37 ºC, CO_2 (5 %) and air (95 %). Cells were seeded (1x10^4 cells/well) in each of the 96-well plate for different concentration of synthesized compounds ranging from 0.01 to 100 µM. After incubation, 6 concentrations (triplicate) of test compounds (prepared in DMSO) were added to the cells and incubated at 37 °C and 5 % CO_2 for 48 h. 20 µL of MTT solution (5 mg/mL) was then added to each well. Plate was further incubated for a period of about 4 h, the supernatant was removed and 200 µL per well DMSO was added to solubilize formazan crystals. Plate was incubated for 10 min and absorbance was measured at 540 nm. (IC_{50} determination at concentrations: 0.01, 0.1, 1, 10, 50 and 100 µM). The statistical analyses were done using GraphPad Prism (version 6.0 forWindows; San Diego, CA, USA).

Agar well diffusion assay (2.2.8)

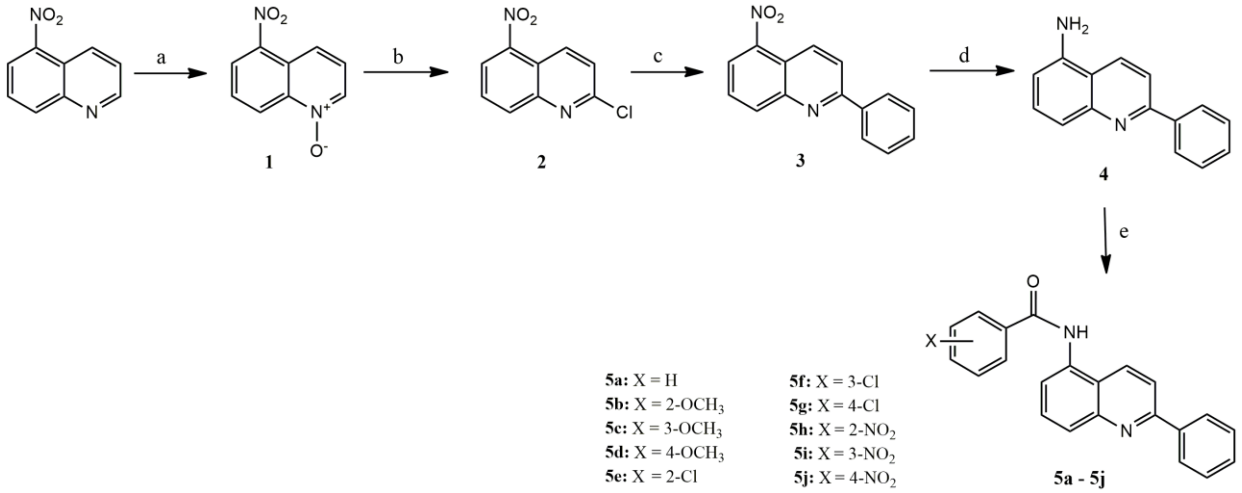
Agar well diffusion assay was performed as reported previously [31] All target compounds were diluted to obtain final concentration of 25 µg/mL using HPLC grade DMSO. The sterile molten Mueller and Hinton agar butt was seeded with 0.4 mL of 24 h old test pathogens (0.1 OD at 540 nm). The seeded NA butt was poured into sterile Petri plates. After solidification of medium, compounds were allowed to diffuse into the punched wells. After incubation at 37 °C for 24 h, the resulting zones of inhibition were measured in millimetres. The derivatives showing the maximum zone of inhibition against test pathogens were checked. The experiment was done in triplicates and the result was reported as mean standard deviation. A control was also prepared for the plates in the same way using solvent DMSO and streptomycin was used as a standard drug and zones of inhibition (mm) were noted.

RESULT AND DISCUSSION

Chemistry (3.1)

A series of novel 2-phenylquinoline amide derivatives (5a-5j) were synthesized from 5-nitroquinoline in five steps as shown in (**Scheme 1)**. The key intermediate 4 was synthesized by following methods described in literature [32-33] The first two step involved the treatment of 5-nitroquinoline with m-CPBA in chloroform to give 5-nitroquinoline-N-oxide (1) which was readily converted to 2-chloro-5-nitroquinoline (2) on treatment with SO_2Cl_2 in DMF at 60°C for 6 h. The third step is classical Suzuki coupling of 2-chloro-5-nitroquinoline with phenylboronic acid in the presence of tetrakis (triphenylphosphine) palladium (0) and caesium carbonate in 1:1 mixture of toluene/ethanol as solvent under thermal heating to give 5-

nitro-2-phenylquinoline (3). The key intermediate (4) was synthesized via reduction of intermediate (3) with $SnCl_2 \cdot 2H_2O$ in the presence of ethyl acetate as solvent at room temperature for 12 h. Finally, the target compounds (5a-5j) were obtained by coupling of 2-phenylquinolin-5-amine (4) with respective acid chlorides using sodium hydride in THF solvent stirred at room temperature for 1 h.



[a]Reagents and conditions: (a) m-CPBA, $CHCl_3$ R.T., 12 h (b) SO_2Cl_2, 60°C, 6 h (c) phenylboronic acid, Cs_2CO_3, $Pd(PPh_3)_4$, toluene/ethanol (1:1), reflux, 12 h (d) $SnCl_2.2H_2O$, EtOAc, R.T., 12 h (e) respective acid chloride, NaH, THF, 0 °C → R.T., 1 h.

Scheme 1. Synthesis of novel 2-phenylquinoline amide derivatives.

Biological Evaluation (3.2)

Anticancer Activity (3.2.1)

All the derivatives were evaluated against MDA-MB-231 (breast adenocarcinoma) using MTT assay (colorimetric method). Cisplatin and Doxorubicin HCl were used as positive controls and the IC_{50} values are reported in µM. The results are shown in **(Table 1)**

It was observed that the IC_{50} values of compounds were found to be in the range of 33.05-16.28 µM. All the derivatives exhibited more or less similar potency and trends were observed when the substituent (X) was varied through its nature and position. Compound 5d (4-OCH_3) and 5g (4-Cl) demonstrated higher activity and the potency was decreased as the substituent (-OCH_3/Cl) was shifted to 2 (5b/5e) and 3 (5c/5f) position, while the compound 5g (4-Cl) was found to be the best molecule (IC_{50} = 16.28 µM) among all analogues (5a-5j). Among the compounds having -NO_2 substituent (5h-5j), compound 5i demonstrated least potency while 5h and 5j exhibited more or less same activity. It can be concluded from the above results that, the substituent (X) at 4 position possessed superior potency than at 2 and 3 position. All the molecules demonstrated potency less than 35 µM and were better than cisplatin but not comparable to doxorubicin.

Table 1. Anticancer activity of novel 2-phenylquinoline amide derivatives.

Compound No.	**X**	**MDA-MB-231[a]**
		IC_{50} ± SD (µM)[b]
5a	H	22.92±1.85
5b	2-OCH_3	24.35±2.05
5c	3-OCH_3	19.00±1.69
5d	4-OCH_3	18.37±0.85
5e	2-Cl	30.35±1.03
5f	3-Cl	19.24±0.59
5g	4-Cl	16.28±1.55
5h	2-NO_2	23.91±1.30
5i	3-NO_2	33.05±2.16
5j	4-NO_2	22.88±0.45
Doxorubicin.HCl		0.64±0.04
Cisplatin		47.95±1.26

[a] Breast adenocarcinoma cell line

[b] Results are mean of triplicate analysis

Antibacterial Activity (3.2.2)

All the compounds were screened against Gram-positive bacteria (*Staphylococcus aureus 6538p* and *Bacillus subtilis*) and Gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*). Streptomycin was used as a standard drug and zones of inhibition (mm) were noted. The results are shown in **(Table 2)**

From antibacterial activity data, it was confirmed that all the compounds showed less potency as compared to standard streptomycin. Among all the synthesized analogues, the compounds 5b, 5d, 5h, 5i and 5j exhibited moderate antibacterial activity against all the tested organisms except 5b with no activity against *Pseudomonas aeruginosa*. Compounds 5c, 5e, 5f and 5g were active against only Gram-positive bacteria (*Staphylococcus aureus 6538p* and *Bacillus subtilis*). Compound 5d (4-OCH_3) exhibited higher potency and the activity was decreased as the substituent (-OCH_3) shifted to 2 and 3 position. Compounds 5h, 5i and 5j having -NO_2 substituent showed almost similar activity. Compound 5a did not exhibit any antibacterial activity.

Table 2. Antibacterial activity of novel 2-phenylquinoline amide derivatives.

Compd. No.	**Zone of inhibition (mm)**			
	Gram-positive bacteria		**Gram-negative bacteria**	
	Staphylococcus aureus 6538p	***Bacillus subtilis***	***Escherichia coli***	***Pseudomonas aeruginosa***
5a	-	-	-	-
5b	12	12	10	-
5c	12	12	-	-
5d	13.5	13	11	11
5e	10	12	-	-
5f	13	11	-	-
5g	10	12	-	-
5h	11	13	10	10
5i	13	13	11	10
5j	12.5	11	11	10
Streptomycin	20	22	22	24

No inhibition

Results are mean of triplicate analysis

CONCLUSION

The present study attempts the synthesis of a novel series of 2-phenylquinoline amide derivatives and subsequent SAR investigations. Based on the observation made during the study, we can conclude that all the compounds showed significant anticancer activity and the trends were observed with variations in the substituent's (X). The substituent's (X) is favoured four position more than two and three position to exhibit superior potency. Compound **5g** exhibited better promising anticancer activity among various synthesized molecules. It was also revealed that, all the compounds showed less antibacterial activity as compared to standard streptomycin. Compounds **5d**, **5h**, **5i** and **5j** exhibited moderate antibacterial activity against all the tested organisms. Further modification and optimization of the reported molecules may lead to the discovery of novel potent drugs with significant anti-breast cancer and antibacterial activities in future.

CONFLICTS OF INTEREST

I declared that I have no known competing financial interests or personal relationships that could have appeared to influence the work reported in

this paper.

ACKNOWLEDGEMENTS

Authors are thankful to Dr. Sandip Gavade and Dr. Anand Burange for their help in recordings spectral data. Authors are also thankful to Dr. Ashish Keche for his valuable support in this work.

REFERENCES

1. Anjali Pathak D, Singh D. Quinoline: A Diverse Therapeutic Agent. IJPSR. **2016**; 7(1):p.1-13.
2. Myers AG, Tom NJ, Fraley ME, Cohen SB, Madar DJ. J Am Chem Soc. **1997**; 119(26):p. 6072-6094.
3. Shen W, Coburn CA, Bornmann WG, Danishefsky SJ.. J Org Chem. **1993**; 58(3):p. 611-617.
4. Rigby JH, Danca DM. Tetrahedron Lett. **1997**; 38(28):p. 4969-4972.
5. Comins DL, Nolan JM. Org Lett. **2001**; 3(26):p. 4255-4257.
6. Toyota M, Komori C, Ihara M. Heterocycles. **2002**; 56(1):p. 101-103.
7. Desai PK, Desai P, Machhi D, Desai CM, Patel D. Indian J Chem Sect B. **1996**; 35:p. 871-873.
8. Roma G, Di-Braccio M, Grossi G, Mattioli F, Ghia M. Euro J Med Chem. **2000**; 35(11):p. 1021-1035.
9. Ko TC, Hour MJ, Lein JC, Teng CM, Lee KH. Bio org Med Chem Lett. **2001**; 11(3):p. 279-282.
10. Kidwai M, Bhushan KR, Sapra P, Saxena RK, Gupta R. Bioorg Med Chem. **2000**; 8(1):p. 69-72.
11. Adana AK, Mirza Y, Aneja KR, Prakash O. Euro J Med Chem. **2003**; 38(5):p. 533-536.
12. Heitsch H. Curr Med Chem. **2002**; 9(9):p. 913-918.
13. Martirosyan AR, Rahim-Bata R, Freeman AB, Clarke CD, Howard RL. Biochem Pharmacol. **2004**; 68(9): p. 1729-1738.
14. Lamazzi C, Leonce S, Pfeiffer B, Renard P, Guillaumet G. Bioorg Med Chem Lett. **2000**; 10(19): p. 2183-2185.
15. Muruganantham N, Sivakumar R, Anbalagan N, Gunasekaran V, Leonard JT. Biol Pharm Bull. **2004**; 27(10): p. 1683-1687.
16. Korotchenko V, Sathunuru R, Gerena L, Caridha D, Li Q. J Med Chem. **2015**; 58(8): p. 3411-3431.
17. Vargas LY, Castelli MV, Kouznetsov VV, Urbina JM, Lopez SN. Bioorg Med Chem. **2003**; 11(7): p. 1531-1550.
18. Broch S, Aboab B, Anizon F, Moreau P. Eur J Med Chem. **2010**; 45(4): p. 1657-1662.
19. Yu XY, Hill JM, Yu G, Yang Y, Kluge AF. Bioorg Med Chem Lett. **2001**; 11(4): p. 541-544.
20. Ghorab MM, Alsaid MS. Acta Pharm. **2015**; 65(3): p. 271-283.
21. Yang Y, Shi L, Zhou Y, Li HQ, Zhu ZW. Bioorg Med Chem Lett. **2010**; 20(22): p. 6653-6656.
22. Narwal M, Venkannagari H, Lehtio L. J Med Chem. **2012**; 55(3): p. 1360-1367.
23. Matsuno K, Masuda Y, Uehara Y, Sato H, Muroya A. ACS Med Chem Lett. **2010**; 1(8): p. 371-375.
24. Wang X, Xie X, Cai Y, Yang X, Li J. Molecules. **2016**; 21(3): p. 340.
25. Benzerka S, Bouraiou A, Bouacida S, Roisnel T, Bentchouala C. Letters in Organic Chemistry. **2013**; 10(2): p. 94-99.
26. Labuschagne A, Lall N, Mphahlele MJ. The International Arabic Journal of Antimicrobial Agents. **2013**; 3(2): p. 1-12.
27. Benzerka S, Bouraiou A, Bouacida S, Roisnel T, Bentchouala C. Letters in Organic Chemistry. **2012**; 9(5): p. 309-313.
28. Aboutorabzadeh SM, Mosaffa F, Hadizadeh F, Ghodsi R. Iran J Basic Med Sci. **2018**; 21(1): p. 9-18.
29. Ghodsi R, Azizi E, Ferlin MG, Pezzi V, Zarghi A. Letters in Drug Design & Discovery. **2016**; 13(1): p. 89-97.
30. Shah CP, Kharkar PS. J Enzyme Inhib Med Chem. **2018**; 33(1): p. 972-977.
31. More G, Raut D, Aruna K, Bootwala S. Journal of Saudi Chemical Society. **2017**; 21: p. 954-964.
32. Kudo Y, Furumoto S, Okamura N. Tau imaging probe. U.S. Patent 9249101B2. **2016**.
33. Familoni OB, Klaas PJ, Lobb KA, Pakade VE, Kaye PT. Org Biomol Chem. **2006**; 4: p. 3960-3965.