Available online at **www.joac.info**

ISSN: 2278-1862

Journal of Applicable Chemistry

2018, 7 (6): 1606-1616

(International Peer Reviewed Journal)

Synthesis, Antimicrobial and Insilico Study of Quinoline based1,2,4-Triazole Derivatives

S. Sreenivasa[1*], G. Shivaraja[1], Vivek Chandramohan[2] and Tadimety Madhu Chakrapani Rao[3]

[1]Department of Studies and Research in Organic Chemistry, Tumkur University, Tumkur-572103, Karnataka, **INDIA**

[2]Department of Biotechnology, Siddaganga Institute of Technology, Tumkur-572103, Karnataka, **INDIA**

[3]Tadimety Aromatics Pvt. Ltd, Hirehalli Industrial Area, Tumkur-572168, Karnataka, INDIA

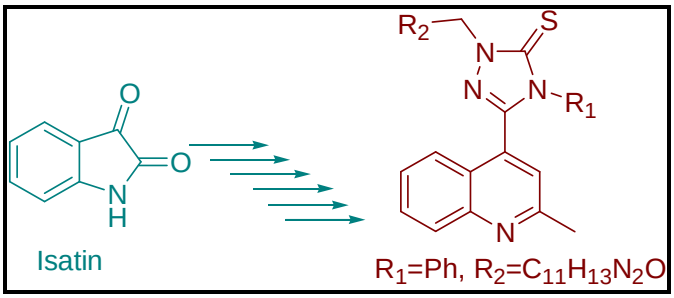
Email: drsreenivasa@yahoo.co.in

Accepted on 5th November, 2018

ABSTRACT

A series of Quinoline based 1,2,4-Triazole derivatives have been synthesized and characterized using FT-IR, ^{1}H and ^{13}C NMR spectroscopic techniques. Compounds were evaluated for their invitro antibacterial activity against selected Gram positive and Gram negative bacteria and antifungal activity against fungal pathogens by adopting broth dilution method of all the compounds ***8c****,* ***8d*** *and* ***8h*** *show good antibacterial activity and* ***8c*** *showed good antifungal activity. Molecular docking studies were performed for these compounds to understand the ligand-receptor possible intermolecular interactions.*

Graphical Abstract



Keywords: Quinoline, 1,2,4-Traizoles, Insilico study, Micro dilution assay, Antimicrobial activity.

INTRODUCTION

1,2,4-triazole and their derivatives represent one of the biologically active classes of heterocyclic compounds. 1,2,4-triazole derivatives are known to exhibit antimicrobial [**1-5**], antituberculosis [**6-10**] and anticancer properties [**11-13**]. They do possess anti-inflammatory [**14**], anticonvulsant [**15**], analgesic [**16**] and antiviral [**17**].

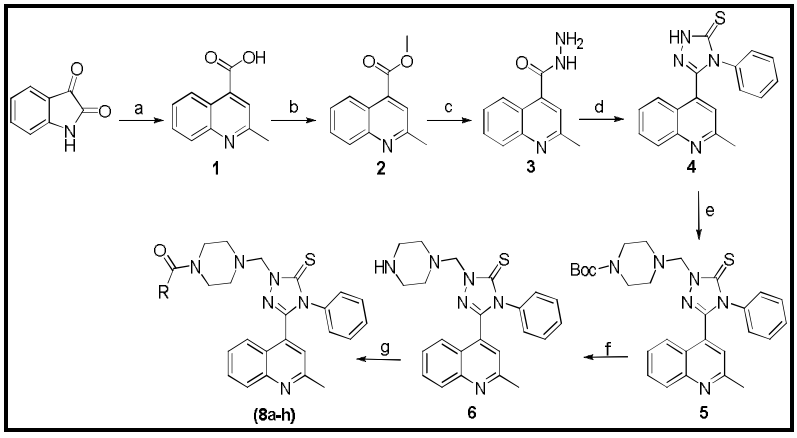
These heterocycles include ribavirin [**18**], rizatriptan [**19**], astazolam and alprazolam [**20-22**], letrozole and anastrozole [**23, 24**]. The number of medicines containing 1,2,4-triazole nucleus, such as itraconazole, fluconazole and posaconazole, used to treat mammalian infectious diseases [**25-28**]. rufinamide [**29**] has a similar structure containing 1,2,4-triazole.It has been noticed continuously over the years that interesting biological activities were possessed by 1,2,4-triazole derivatives.

There are several methods for synthesizing 1,2,4-triazole in the literature, which are used in a conventional one pot, multi-component, microwave-assisted, non-solvent-free were reported. Joshi et al. synthesized 1,2,4 triazoles by microwave technology quickly and efficiently [**30**]. Synthesis of 4-amino-3-mercapto-5-substituted 1,2,4-triazoles was carried out by many workers and few are summarized here, 5 mercapto-3 (3'-pyridyl) -4H-1,2,4-triazole were synthesized by Kshirsagar *et al* and evaluated their anticonvulsant and infection activity [**31**]. 1,2,4-triazole was found to have antifungal and anti-HIV activity [**32**]. According to researchers, new 1,2,4-triazole systems have new antibiotics, antiproliferative, antibiotics, antifungal and anti-aging biological actions. Furthermore, mercapto-1,2,4-triazoles found great benefit in the preparation of other heterocyclic compounds [**33**]. Mir and et al. Synthesized a new series of [5- (2-furyl) -1,2,4-triazol-3-ylthio] acetohydrazide and related compounds and evaluated antituberculous activity [**34**]. Aim of present study is to synthesize new bioactive quinoline based 1,2,4-triazoles. The clubbed 1,2,4-triazole ring with quinoline may lead to compounds that have more efficiency in biological activity [**35**].

MATERIALS AND METHODS

All the chemicals and solvents used were procured from Spectrochem and Sigma Aldrich make in the appropriate grade and used without further purification; purity of the chemicals and solvents received was confirmed by thin layer chromatography. Melting points were determined in open capillary and are uncorrected. IR spectra were recorded on Bruker alpha series in KBr (vmax cm^{-1}). ^{1}H and ^{13}C NMR Spectra were recorded on a BRUKER AV400, Agilent 400MR and Bruker AV III 500 MHz spectrometer using TMS as internal standard and chemical shift values are reported in δ ppm.

Pfitzinger reaction of isatins afforded 2-methylquinoline-4-carboxylic acid (1) [**36**] which on esterfication afforded quinoline ester (2). Carbohydrazide (3) was subsequently obtained by reacting qunoline ester with excess hydrazine hydrate. Obtained carbohydrazide was treated with phenylisothiocyanate to get condensed product which was further cyclised using strong base to get compound (4). Mannich reaction on compound (4) with N-boc piperazine afforded compound (5). compound (5) was deprotected using trifluro acetic acid to get the intermediate (6) which was coupled with different carboxylic acids using coupling agent carbonyl diimidazole (CDI) to get different acid amide derivatives (8a-h).



Scheme 1

Reagents and conditions: a) CH_3COCH_3, 33%KOH, reflux, 8h; b) MeOH, H_2SO_4, reflux, 6h; c) $NH_2NH_2.H_2O$, MeOH, relux, overnight; d) PhNCS,NaOH, EtOH, reflux, 3h; e) CH_2O, N-Boc piperazine, EtOH, stirred 12h; f)TFA,DCM, 0^OC, 2h; g) RCOOH 7a-h,CDI,THF,RT,18h.

Procedure for the synthesis of 2-methylquinoline3-carbohydrazide (3): Quinoline ester (2g, 0.009 mol) was dissolved in methanol (15 mL) and excess amount of hydrazine hydrate (0.95g, 0.029 mol) was added to the reaction mixture and refluxed overnight. Completion of the reaction was monitored by TLC. The reaction mixture was cooled to room temperature; white crystals separated out from reaction mixture. These crystals were filtered, washed with methanol to obtain the pure compound. These crystals were taken for the next step. Yield 89%, m.p. 221-223°C; IR (KBr): 3259-3141 (-NH-NH_2), 2928 (Ar-H), 1620 cm^{-1} (–C=O); ^{1}H NMR (400 MHz, DMSO-d_6): δ 9.6 (s,1H),8.18 (s, 1H, Ar-H), 7.93 (t, 1H,Ar-H), 7.7 (t, 1H, Ar-H),7.83 (d, 1H, Ar-H), 7.68 (s, 1H, NH),7.51 (s, 2H, NH_2), 1.8 (s, 3H, CH_3); LC-MS (ESI, m/z): 202(M+H).

Procedure for the synthesis of 3-(2-methylquinolin-4-yl)-4-phenyl-1H-1,2,4-triazole-5(4H)-thione (4): Compound **3** (0.5 g, 5mmol) was dissolved in ethanol, to this added phenyl isothiocyanate (0.675g, 0.05mol) and reaction mixture was stirred for 2 hours. The obtained solid was filtered, dissolved in 20 mL of 2M NaOH and refluxed for 3 hours. After cooling, reaction mixture was acidified with acetic acid, the precipitate obtained was filtered and recrystallized with ethanol. Light yellow color powder was obtained. Yield 35%,m.p. 248-250^{O}C; IR (KBr): 2853 cm^{-1} (C-H), 1609 cm^{-1}, 1553 cm^{-1} (C = N, C = C)and 1203 cm^{-1} (C = S). ^{1}H NMR (400 MHz, DMSO-d_6): δ 14.42 (s, 1H, NH), 7.93(d, 1H, *J* = 8.5Hz, Ar-H), 7.92 (d, 1H,*J*= 8.5Hz, Ar-H), 7.72(dd, 1H, *J*= 8.0Hz, Ar-H), 7.52 (dd, 1H, *J*= 8.0Hz,Ar-H), 7.31 (m, 5H, Ar-H), 7.46(s, 1H, Ar-H), 2.55(s, 3H, CH_3).

Procedure for the synthesis of tert-butyl 4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazine-1-carboxylate (5): Compound **4** (1g, 0.002 mol) was dissolved in absolute ethanol (100 mL). Then formaldehyde (37%, 1.0 mL) and N-Boc piperazine (1.17g, 0.002 mol) were added drop wise with vigorous stirring. After combining all reagents, the reaction mixture was stirred at room temperature for 12 h. The mixture was cooled, the solid product was filtered and washed with petroleum ether. The solid that separated was recrystallized from ethanol-dioxane (1:2) to yield the compound**5**. Yield 47%; m.p. 235-237 ^{O} C; IR (KBr): 1620 cm^{-1} (–C=O), 1553 cm^{-1} (C = N, C = C) and 1203 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.95-7.93(d, 1H, Ar-H), 7.77-7.70(m, 2H, Ar-H), 7.56-7.53(t, 1H, Ar-H), 7.20-7.17(t, 2H, Ar-H), 6.81(t, 1H, Ar-H), 6.58-6.56(d, 1H, Ar-H), 6.25-2.23(d, 2H, Ar-H), 4.52(s, 2H, CH_2), 3.20-3.18(t, 4H, $N(CH_2)_2$), 2.51-2.49(t, 4H, $N(CH_2)_2$), 2. 44(s, 3H, CH_3).

Procedure for the synthesis of 3-(2-methylquinolin-4-yl)-4-phenyl-1-(piperazin-1-ylmethyl)-1H-1,2,4-triazole-5(4H)-thione (6): A solution of starting material **5** (1.5 g, 0.019 mol) in DCM (50 mL) was treated with TFA (2 mL) stirred for 2 hours at 0 ^{O}C. The solvent was removed in vacuo and the crude was dissolved in 10% IPA/chloroform, washed with $NaHCO_3$, brine, dried with $MgSO_4$ and concentrated. The resulting material was purified by Column chromatography to get the product **6**as a solid. Yield 25%; m.p. 226-227 ^{O} C; IR (KBr): 1553 cm^{-1} (C = N, C = C) and 1203 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.95-7.92 (m, 3H, Ar-H), 7.78-7.70 (m, 4H, Ar-H), 7.56-7.50(t, 1H, Ar-H), 7.19-7.17 (t, 2H, Ar-H), 6.80 (t, 1H, Ar-H), 6.57-6.56 (d, 1H, Ar-H), 6.24-2.23 (d, 2H, Ar-H), 4.52 (s, 2H, CH_2), 3.20-3.18 (t, 4H, $N(CH_2)_2$), 2.51-2.49 (t, 4H, $N(CH_2)_2$), 2. 44 (s, 3H, CH_3).

General procedure for the synthesis of (4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl) piperazin-1-yl) (substituted phenyl)methanone derivatives (8a-h): The carboxylic acids **7a-h** (152 mg, 0.0801 mol) was added to a solution of CDI (130 mg, 0.0801 mol) in dry THF(1 mL) at RT under N_2. The mixture was stirred for 90 min at RT, after which it was added over 25 min to a solution of the compound **6** (200 mg, 0.0400 mol) in THF (1.5 mL) at RT under N_2. The reaction mixture was stirred at RT for 16 h. after completion of reaction, the mixture was concentrated in vacuo and the crude obtained was extracted with ethyl acetate. The

organic phase was washed with brine, dried ($MgSO_4$), and concentrated to give oil. The oil was purified by column chromatography to get the products (8a-h).

Spectral interpretation

(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl) piperazin-1-yl)(phenyl)methanone(8a): IR (KBr): 1620 cm^{-1} (–C=O), 1554 cm^{-1} (C = N, C = C) and 1201 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ8.03-8.01(d, 2H, Ar-H), 7.95(d, 1H, Ar-H), 7.63-7.56(m, 6H, Ar-H), 7.20-7.17(t, 2H, Ar-H), 6.81(t, 1H, Ar-H), 6.58-6.56(d, 1H, Ar-H), 6.25-6.23(d, 2H, Ar-H), 4.52(s, 2H, CH2), 3.44-3.40(t, 4H, $N(CH_2)_2$), 2.55-2.53(t, 4H, $N(CH_2)_2$, 2.51(s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6):168.9, 167.5, 158.9, 149.7, 148.9, 139.3, 135.7, 135.2, 129.7, 129.0, 128.7, 128.5, 128.4, 128.3, 124.3, 120.7, 77.2, 52.6, 50.1, 24.3.

(4-chlorophenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8b): IR (KBr): 1622 cm^{-1} (–C=O), 1553 cm^{-1} (C = N, C = C) and 1205 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.97-7.95 (d, 2H, Ar-H), 7.77-7.56 (m, 5H, Ar-H), 7.21-7.18 (t, 2H, Ar-H), 6.80 (t, 1H, Ar-H), 6.56-6.54 (d, 1H, Ar-H), 6.24-6.23 (d, 2H, Ar-H), 4.51 (s, 2H, CH2), 3.43-3.40 (t, 4H, $N(CH_2)_2$), 2.54-2.53 (t, 4H, $N(CH_2)_2$, 2.51 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 168.9, 167.5, 158.9, 149.6, 148.8, 139.3, 135.6, 135.2, 129.5, 129.0, 128.7, 128.6, 128.4, 127.3, 124.3, 120.1, 77.2, 52.5, 50.5, 24.1.

(3-bromo-5-chlorophenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8c): IR (KBr): 1620 cm^{-1} (–C=O), 1554 cm^{-1} (C = N, C = C) and 1201 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.10 (s, 1H, Ar-H), 8.07(s, 1H, Ar-H), 7.95-7.56 (m, 4H, Ar-H), 7.22-7.19 (t, 2H, Ar-H), 6.82 (t, 1H, Ar-H), 6.55-6.53 (d, 1H, Ar-H), 6.24-6.23 (d, 2H, Ar-H), 4.50 (s, 2H, CH_2), 3.40-3.38 (t, 4H, $N(CH_2)_2$), 2.54-2.53 (t, 4H, $N(CH_2)_2$, 2.51 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 168.8, 167.5, 159.9, 149.7, 148.9, 139.3, 135.7, 134.2, 129.6, 129.0, 128.7, 128.5, 128.4, 127.3, 124.3, 121.7, 76.2, 51.6, 51.1, 23.3.

(4-methoxyphenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8d): IR (KBr): 1621 cm^{-1} (–C=O), 1550 cm^{-1} (C = N, C = C) and 1202 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.92-7.90 (d, 2H, Ar-H), 7.77-7.70 (m, 2H, Ar-H), 7.56-7.54 (t, 1H, Ar-H), 7.20-7.18 (m, 4H, Ar-H), 6.81 (t, 1H, Ar-H), 6.57-6.56 (d, 1H, Ar-H), 6.23-6.22 (d, 2H, Ar-H), 4.50(s, 2H, CH2), 3.34-3.30 (t, 4H, $N(CH_2)_2$), 2.54-2.53 (t, 4H, $N(CH_2)_2$, 2.51 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 167.9, 167.5, 158.9, 148.7, 147.9, 139.3, 134.7, 134.2, 129.6, 129.1, 128.4, 128.3, 128.2, 128.1, 124.3, 120.7, 77.2, 52.6, 50.1, 24.3.

(2-chlorophenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8e): IR (KBr): 1619 cm^{-1} (–C=O), 1553 cm^{-1} (C = N, C = C) and 1201 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.95-7.93 (d, 1H, Ar-H), 7.77-7.43 (m, 7H, Ar-H), 7.28-7.26 (t, 2H, Ar-H), 6.84(t, 1H, Ar-H), 6.68-6.66 (d, 1H, Ar-H), 6.35-6.33 (d, 2H, Ar-H), 4.55 (s, 2H, CH2), 3.45-3.43 (t, 4H, $N(CH_2)_2$), 2.58-2.57 (t, 4H, $N(CH_2)_2$, 2.52 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 168.7, 166.5, 159.9, 149.7, 148.8, 139.3, 135.3, 135.2, 129.1, 129.0, 128.7, 128.4, 128.3, 128.1, 124.3, 120.7, 77.2, 52.6, 50.1, 24.7.

(3-chlorophenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8f): IR (KBr): 1622 cm^{-1} (–C=O), 1552 cm^{-1} (C = N, C = C) and 1200 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.95-7.91 (m, 3H, Ar-H), 7.77-7.70 (m, 3H, Ar-H), 7.57-7.56 (m, 2H, Ar-H), 7.20-7.17 (t, 2H, Ar-H), 6.81 (t, 1H, Ar-H), 6.58-6.56 (d, 1H, Ar-H), 6.25-6.23 (d, 2H, Ar-H), 4.52 (s, 2H, CH2), 3.44-3.40 (t, 4H, $N(CH_2)_2$), 2.55-2.53 (t, 4H, $N(CH_2)_2$, 2.51 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 168.9, 167.5, 158.9, 149.7, 148.9, 139.3, 135.7, 135.2, 129.7, 129.0, 128.7, 128.5, 128.4, 128.3, 124.3, 120.7, 77.2, 52.6, 50.2, 24.2.

(2-methoxyphenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8g): IR (KBr): 1623 cm^{-1} (–C=O), 1554 cm^{-1} (C = N, C = C) and 1202 cm^{-1} (C = S); ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.95-7.92 (d, 1H, Ar-H), 7.82-7.70

(m, 3H, Ar-H), 7.59-7.56 (m, 2H, Ar-H), 7.24-7.20 (m, 3H, Ar-H), 6.90 (s, 1H, Ar-H), 6.81(s, 1H, Ar-H), 6.58-6.56 (d, 1H, Ar-H), 6.25-6.23 (d, 2H, Ar-H), 4.52 (s, 2H, CH2), 3.44-3.40 (t, 4H, $N(CH_2)_2$), 2.55-2.53 (t, 4H, $N(CH_2)_2$, 2.51 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 169.9, 168.5, 159.9, 150.7, 148.9, 139.3, 136.7, 135.2, 130.7, 129.0, 128.7, 128.5, 128.4, 128.3, 125.3, 120.7, 78.2, 52.6, 51.1, 25.7.

(4-bromophenyl)(4-((3-(2-methylquinolin-4-yl)-4-phenyl-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-yl)methyl)piperazin-1-yl)methanone(8h): IR (KBr): 1621 cm^{-1} (–C=O), 1552 cm^{-1} (C = N, C = C) and 1202 cm^{-1} (C = S); ^1H NMR (400 MHz, DMSO-d_6): δ 7.95-7.94 (d, 2H, Ar-H), 7.76-7.55 (m, 5H, Ar-H), 7.20-7.18 (t, 2H, Ar-H), 6.79 (t, 1H, Ar-H), 6.55-6.54 (d, 1H, Ar-H), 6.23-6.22 (d, 2H, Ar-H), 4.50 (s, 2H, CH2), 3.42-3.40 (t, 4H, $N(CH_2)_2$), 2.53-2.51 (t, 4H, $N(CH_2)_2$, 2.51 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): 168.9, 167.5, 158.9, 149.6, 148.8, 139.3, 135.6, 135.2, 129.5, 129.0, 128.7, 128.6, 128.4, 127.3, 124.3, 120.1, 76.2, 51.5, 49.5, 23.1.

Antimicrobial studies: The zone of inhibition of the synthesized compounds 8a-h was determined by agar diffusion technique [**37-39**]. The organism tested were *Staphylococcus aureus* (MTCC 96), *Escherichia coli* for antibacterial activity and *Aspergillus niger* (MTCC 1344), *Candida albicans* (MTCC 854) for antifungal activity. The agar media were inoculated with test organism and a solution of test compound (50 µL of 10 µg mL^{-1} in DMSO), 10 µg $mL^{-1}$was separately in cups (8 mm diameter) in the agar medium. Ciprofloxacin (10 mcg $disc^{-1}$) and Nystatin (25 mcg $disc^{-1}$) were used as a reference for antibacterial and antifungal activity respectively. The zones of inhibition were measured after 24 h incubation. MIC values were determined by disc diffusion method [**40**]. The stocks of each sample were prepared in the concentration 50 µg mL^{-1}. This was done by weighing 0.01mg of the sample and dissolving in 2 mL of DMSO. From stock solution, different concentrations of 10 µg mL^{-1}, 20 µg mL^{-1},30 µg mL^{-1}, 40 µg mL^{-1}, of each compound were prepared. Thus, proper amounts of the different concentrations of compounds were pipette on the blank disks, which were placed on the plates. The plates were incubated at 37°C for 24 h. The minimum inhibitory concentrations (MICs), the lowest concentration (µg mL^{-1}) of the test compound that gives no visible growth on the plates were recorded. DMSO was used as a solvent control to ensure that solvent had no effect on bacterial growth. Ciprofloxacin (10 µg 50 $µL^{-1}$) and Nystatin (10 µg 50 $µL^{-1}$) were used as a control drug. The results of the anti-microbial activities are summarized in table 2 and 3.

Molecular docking studies: The novel Quinoline based1,2,4-Triazole derivatives were subjected to dock in the active site of DNA gyrase enzyme using Lead IT software[**41**]. We investigated the theoretical binding mode of eight ligands at the binding site using molecular modeling. Molecular docking studies were performed for these ligands to understand the ligand-receptor possible intermolecular interactions in detail.

The crystal structure of the DNA gyrase (PDB code 1AB4) with resolution 2.3 Å was chosen as the protein model for the present study [**42**]. Water molecules and ligand were removed from the protein file. The resulting crystallography structure was imported in Discovery studio 3.1. The structures of the ligands were optimized using Discovery studio [**43**]. Using the MM+ molecular mechanical force field, 3D geometry optimization calculations for each ligand were performed. The ultimate conformations were calculated with the semi empirical parameterized model number 3 (PM3) methods. The molecular structures were optimized using the Polak-Ribiere algorithm until the root mean square gradient was 0.01 kcal $mol^{-1}Å^{-1}$. Geometry optimization was run many times with different starting points of each 8 ligand. Docking was performed using the routine procedure and default parameters of molecular docking Lead IT software and implemented empirical free energy function. In the docking protocol, ligands were assumed to be flexible molecules and the docking software was allowed to rotate all rotatable bonds of the ligands to obtain the best and optimized conformer of the ligands within the active site of the protein.

RESULTS AND DISCUSSION

Chemistry: The synthetic route of compounds is outlined in scheme 1. In present work 4-quinoline carboxylic acid was prepared by pfitzinger method which on esterification formed cinchoninic ester which was made to react with hydrazine hydrate to form carbohydrazide (3). Obtained carbohydrazide was treated with phenylisothiocyanate to get condensed product which was further cyclised using strong base to get compound (4). Mannich reaction on compound (5) with N-boc piperazine afforded compound (5). Using trifluro acetic acid compound (5) was deprotected to get the intermediate (6) which was coupled with different carboxylic acids afforeded derivatives (8a-h). The physical data of all the compounds are tabulated in table 1.

Table 1. Physical characterisation of all the synthesized compounds (8a-h)

Entry	R	Molecular fornula	Melting point (^{O}C)	Yield(%)
1	Ph	$C_{30}H_{28}N_6OS$	235	63
2	4-ClPh	$C_{30}H_{27}ClN_6OS$	254	56
3	3-Br-5-ClPh	$C_{30}H_{26}BrClN_6OS$	252	42
4	4-OMe	$C_{31}H_{30}N_6O_2S$	264	73
5	2-Cl	$C_{30}H_{27}ClN_6OS$	257	36
6	3-Cl	$C_{30}H_{27}ClN_6OS$	271	82
7	2-OMe	$C_{31}H_{30}N_6O_2S$	261	84
8	4-Br	$C_{30}H_{27}BrN_6OS$	252	39

Antibacterial activity: All compounds inhibit bacterial growth against Gram positive bacteria (*S. Aureus*) and Gram negative bacteria (*E.coli*). Compound 8c, 8d and 8h shows good inhibition (9-25 mm) in various concentrations whereas other compounds shows moderate inhibition (8-15mm) against gram negative and gram positive bacteria in various concentration. Control (Ciprofloxacin) shows good inhibition (23-14 mm) against all tested strains whereas negative control (DMSO) shows zero inhibition as shown in table 2 and Initially concentrations of 2.5, 5, 7.5 and 10 µg mL^{-1} of samples, control and standard were used to evaluate the dose dependent activity of test samples, at 10 µg mL^{-1} no growth of bacteria was observed this concentration was taken as MIC.

Table 2. Antibacterial activities of compounds 8a-h

Compounds	*S. aureus*(µg mL^{-1})				*E. coli* (µg mL^{-1})			
	10	20	30	40	10	20	30	40
8a	8	9	10	11	9	11	12	13
8b	8	10	13	14	8	9	11	14
8c	10	12	16	18	17	19	21	23
8d	9	12	14	17	17	18	23	24
8e	9	10	10	12	9	11	12	13
8f	8	9	11	12	7	9	11	15
8g	8	10	11	13	9	12	14	15
8h	11	12	15	17	18	19	24	25
CIPRO	22	22	22	22	28	28	28	28
DMSO	-	-	-	-	-	-	-	-

Zone of inhibition is expressed in mm at 10 µg mL^{-1}, 20 µg mL^{-1}, 30 µg mL^{-1}and 40 µg mL^{-1}.

Antifungal activity: All compounds inhibit fungal growth against *A. Niger*. *C. Albicans*at low concentration (10,20 µg mL^{-1}) as well as at higher concentration(30,40 µg mL^{-1}).Compound 8c shows good inhibition 10-19 mm against *C.albicans* and *A.niger* at low concentration (10 µg mL^{-1}).Control (Nystatin) shows good inhibition (12-25 mm) against all tested strains whereas negative control (DMSO) shows zero inhibition as shown in table 3. Minimum Inhibition Concentration of most of compounds is at 10 µg mL^{-1}.

A closer look at the results revealed that the title compounds possess better antibacterial activity as compared to antifungal activity. Among all the prepared compounds, **8c**, **8d** and **8h** were found to be the most promising antibacterial and **8c** as antifungal agents. Core structures consists of quinoline and 1,2,4-triazole,substituting electron withdrawing chlorine, bromine atom in various position of quinoline ring greatly increase antibacterial activity similarly.

Table 3. Antifungal activities of compounds 8a-h.

Compounds	*A.niger* (µg mL^{-1})				*C.albicans* (µg mL^{-1})			
	10	20	30	40	10	20	30	40
8a	5	6	10	12	10	13	11	12
8b	9	11	14	15	6	7	10	11
8c	10	13	15	17	13	15	18	19
8d	9	10	11	12	10	11	13	14
8e	8	9	10	11	8	10	11	13
8f	9	10	11	12	7	9	11	13
8g	8	10	11	13	9	12	14	15
8h	10	12	13	15	8	9	11	14
Nystatin	25	25	25	25	22	22	22	22

Zone of inhibition is expressed in mm at 10 µg mL^{-1}, 20 µg mL^{-1}, 30 µg mL^{-1}and 40 µg mL^{-1}.

Molecular docking: Table 4 summarizes the binding characterizations between synthesized compounds and DNA gyrase. According to the data presented in this table ligands interact with the DNA gyrase binding site through hydrogen bonding. Amongst various conformations of the ligands obtained from the docking procedure the conformation with the best scored pose, with the lowest binding energy (~ -22.891 – 21.419 kcal mol^{-1}) shown in table 4. However, the best scored pose for **8c**, **8d** and **8h** (-22.891, -21.876 and -21.419 kcal mol^{-1}, respectively) had a similar position and orientation with the reference structure.

Table 4. Docking results of compounds 8a-h

Comp.	Docking energy (Kcal mol^{-1})	Number of interactions	Interacted amino acids via hydrogen bond
8a	-16.705	12	ASP87,ARG32,ILE174
8b	-18.088	7	ASP87,GLN267
8c	-22.891	17	ASP104,ARG126
8d	-21.876	11	ASP87,ARG32,ILE174,ARG91
8e	-18.188	15	ASP104,GLY105,ARG126,ALA93,GLY107
8f	-18.092	8	ASP115,GLN267,SER111,GLY114,ILE112
8g	-17.952	8	ASP87,ARG32,ILE174,GLN267
8h	-21.419	7	ASP87, ARG32,ILE174
CIPRO	-25.403	12	GLN267, TYR226, ARG91, ASP87

Figures 1 and 2 show 3D schematic presentations of compounds **8c**, **8d** and **8h** as well as Ciprofloxacin docked into the binding site and support the idea that the compounds are well incorporated into the binding pocket. Dock pose of each ligand was analyzed for interactions with the receptor. These hydrophobic sites of the ligands are conserved in all the structures (Fig. 1 and 2). The residue that interacts through a hydrogen bonding with the acceptor/donor site of the ligand differs depending on the ligand. The binding patterns of different ligands are also slightly different.

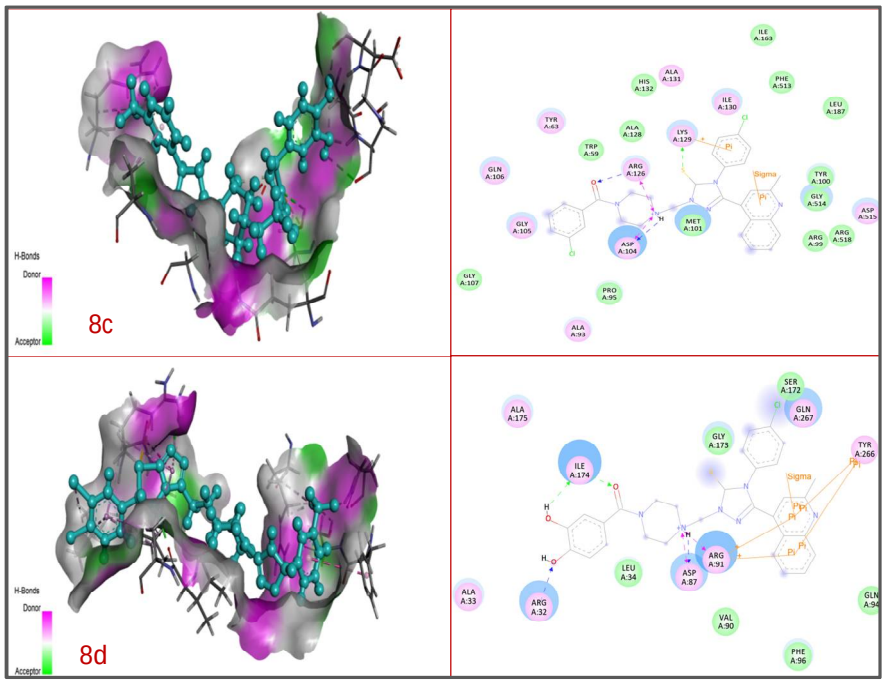


Figure 1. Binding poses of 8c and 8d with DNA gyrase protein.

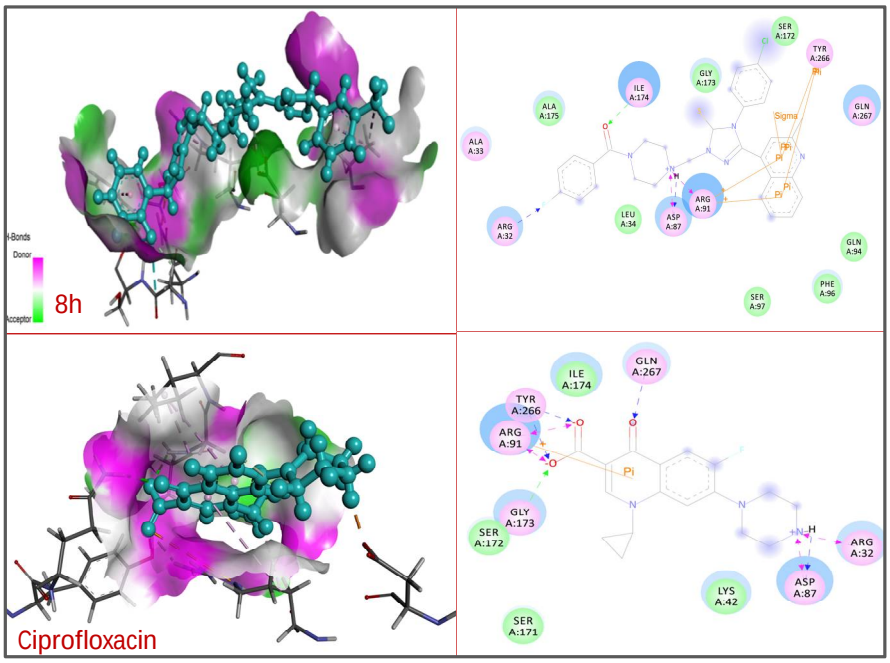


Figure 2. Binding poses of 8h and Ciprofloxacin with DNA gyrase protein.

In this study, the results have shown that synthesized novel quinoline 1,2,4-triazole derivatives followed Lipinski's RO5 (data not shown).

APPLICATION

The synthesized quinoline based 1,2,4-triazole derivatives are useful against the bacterial infections. Research on this type of heterocyclic molecules will be continued.

CONCLUSION

In conclusion, we have efficiently synthesized a series of novel qunoline derivatized 1,2,4-Triazole derivativesin good yield and their antimicrobial activity were evaluated. Synthesized compounds take part in an altered pattern of biological activity compared to the standard drug employed ciprofloxacin. Triazoles **8c**, **8d** and **8h** demonstrated potent inhibition against tested bacteria strains. Only compound **8c** showed good antifungal activity. It can be concluded that incorporation of a 1,2,4-triazole ring into quinoline moiety results in few promising antibacterial molecules. Docking score and binding of the most active compounds into the Gyrase crystal structure of *S. aureus* and *E.coli* is in compatible with the invitro study carried out.

ACKNOWLEDGEMENTS

Authors acknowledge to UGC, New Delhi for financial support. Reference No: 43/226-2014(SR) Dated 12–08-2015.

REFERENCES

[1]. Z. A. Kaplancikli, G. Turan-Zitouni, Ozdemir A, G. Revail, New triazole and triazolo thiadiazine derivatives as possible antimicrobial agents, *Eur. J. Med. Chem.*, **2008**, 43, 155-9.
[2]. R. Mishra, R. Kumar, S. Kumar, J. Majeed, M. Rashid, S. Sharma, Synthesis and In vitro antimicrobialactivity of Some Triazole Derivatives, *J. Chil. Chem. Soc.*, **2010**, 55, 359-362.
[3]. Z. Muhi-Eldeen, M. Nadir, N. R. Aljobory, F. Husseen, S. Stohs, Synthesis of New biologically active Triazoles derivative, *Eur. J. Med. Chem.*,**1991**, 26, 237-241.
[4]. N. Upmanyu, S. Kumar, M. D. Kharya, K. Shah, P. Mishra, Synthesis and anti-microbial evaluation of some novel 1,2,4-triazole derivatives, *Acta. Pol. Pharm.*, **2011**, 68, 213-21.
[5]. M. Abdul Rahiman, D. R. Mamatha, G. L. Thejashree, M. G.Suresha, Synthesis, Characterization and Antimicrobial Studies of Some Novel Thiadiazoles derived from [1,2,4] Triazoles, *J. Applicable Chem.*,**2017**, 6, 01-07.
[6]. L. Zahajska, V. Klimesova, J. Koci, K. Waisser, J .Kaustova, Synthesis and antimycobacterial activity of pyridylmethylsulfanyl and naphthylmethylsulfanyl derivatives of benzazoles, 1, 2, 4-triazole, and pyridine-2-carbothioamide/-2-carbonitrile, *Arch. Pharm.(Weinheim)*, **2004**, 337, 549-55.
[7]. A. Foroumadi, Z. Kiani, F. Soltani, Antituberculosis agents VIII. Synthesis and in vitro antimycobacterial activity of alkyl alpha-[5-(5-nitro-2-thienyl)-1,3,4-thiadiazole-2-ylthio] acetates, *Il Farmaco*, **2003**, 58, 1073-6.
[8]. S. D. Joshi, H. M. Vagdevi, V. P. Vaidya, G. S. Gadaginamath, Synthesis of new 4-pyrrol-1-yl benzoic acid hydrazide analogs and some derived oxadiazole, triazole and pyrrole ring systems: a novel class of potential antibacterial and antitubercular agents, *Eur. J. Med. Chem.*,2008, 43, 1989-96.
[9]. B. P. Mallikarjuna, B. S. Sastry, G. V. Suresh Kumar, Y. Rajendraprasad, S. M. Chandrashekar, K. Sathisha, Synthesis of new 4-isopropylthiazole hydrazide analogs and some derived clubbed triazole, oxadiazole ring systems--a novel class of potential antibacterial, antifungal and antitubercular agents, *Eur. J. Med. Chem.*,**2009**,44, 4739-46.
[10]. G. V. Suresh Kumar, Y. Rajendraprasad, B. P. Mallikarjuna, S. M. Chandrashekar, C. Kistayya, Synthesis of some novel 2-substituted-5-[isopropylthiazole] clubbed 1,2,4-triazole and 1,3,4-oxadiazoles as potential antimicrobial and antitubercular agents, *Eur. J. Med. Chem.*, **2010**,45, 2063-74.
[11]. E. Pomarnacka, M. Gdaniec, Synthesis and anticancer activity of 2-amino-8-chloro-5,5-dioxo[1,2,4]triazolo[2,3-b][1,4,2]benzodithiazine derivatives, *Bioorg. Med. Chem.*, **2003**, 11, 1259-67.

[12]. B. S. Holla, K. N. Poojary, B. Sooryanarayana Rao, M. K. Shivananda, New bis-amino mercaptotriazoles and bis-triazolothiadiazoles as possible anticancer agents, *Eur. J. Med.Chem.*, **2002**, 37, 511-7.
[13]. B. S. Holla, B. Veerendra, M. K.Shivananda, B. Poojary, Synthesis characterization and anticancer activity studies on some Mannich bases derived from 1,2,4-triazoles, *Eur. J. Med.Chem.*,**2003**, 38, 759-67.
[14]. M. F. El-Shehry, A. A.Abu-Hashem,E, M,El-Telbani, Synthesis of 3-((2,4-dichlorophenoxy) methyl)-1,2,4-triazolo(thiadiazoles and thiadiazines) as anti-inflammatory and molluscicidal agents, *Eur. J. Med. Chem.*, **2010,**45, 1906-11.
[15]. J. Chen, X. Y .Sun, K. Y. Chai, J. S. Lee, M. S. Song, Z. S.Quan, Synthesis and anticonvulsant evaluation of 4-(4-alkoxylphenyl)-3-ethyl-4H-1,2,4-triazoles as open-chain analogues of 7-alkoxyl-4,5-dihydro[1,2,4]triazolo[4,3-a]quinolines,*Bioorg. Med .Chem.*, **2007**, 15, 6775-81.
[16]. G. L. Almajan, S. F. Barbucenau, E. R.Almajan, C. Draghici, G. Saramet, Synthesis, characterization and antibacterial activity of some triazole Mannich bases carrying diphenylsulfone moieties, *Eur. J. Med. Chem.*, **2009**, 44, 3083-3089.
[17]. M. Kritsanida, A. Mouroutsou, P. Marakos, N. Pouli, S. Papakonstantinou-Garoufalias, C. Pannecouque, M. Witvrouw, E. De Clercq, *Il Farmaco*, **2002**,57, 253-257.
[18]. B. M.Rao, S.Sangaraju, M.K.Srinivasu, P.Madhavan, M. L.Devi, P. R.Kumar, K. B.Candrasekhar, C.Arpitha,T. S.Balaji, Development and validation of a specific stability indicating high performance liquid chromatographic method for rizatriptan benzoate,*J. Pharm.Biomed.Anal.*,**2006**, 41, 1146.
[19]. G. Hancu, A. Gaspar, A. Gyeresi, Separation of 1,4-benzodiazepines by micellarelektrokinetic capillary chromatography, *J.Biochem.Biophys.Methods*, **2007**, 69, 251-9.
[20]. Z. Z.Wang, Y. P.Wang, B.Yang, J. P.Wan,J.Wang, Clinical efficacies of treating refractory insomnia patients with estazolam plus alternating current magnetic field, *Chinese Med. J.*, **2012**,125, 3175-6.
[21]. E. Bajetti, N. Zilembo, E. Bichisao, P. Pozzi, L. Toffolatti, *Critical Reviews in Oncology Hematolog*, **2000**, 33, 137-168.
[22]. Ó. García-Algar, M. Á.López-Vílchez, I.Martín, A.Mur, M.Pellegrini, R.Pacifici, S. Rossi, S .Pichini, *Clinical Toxicol*, **2007**, 45, 295-8.
[23]. A. Demirbas, S. Ceylan , N. Demirbas,,Synthesis of some new five membered heterocycles, a facile synthesis of oxazolidinones, *J.Heterocycl. Chem*, **2007**, 44, 1271-1280.
[24]. C. Foulon, C. Danel, C. Vaccher, S. Yous, J. P.Bonte,J, F, Goossens, Determination of ionization constants of N-imidazole derivatives, aromatase inhibitors, using capillary electrophoresis and influence of substituents on pKa shifts, *J, Chromatograph A.*, **2004**,1035, 131-136.
[25]. L. T. Yu, M. T. Ho, C. Y. Chang, T. K. Yang, Asymmetric zinc Reformatsky reaction of evanschiral imide with acetophenones and its application to the stereoselective synthesis of triazole antifungal agents, *Tetrahedron: Asymmetry*, **2007**, 18, 949-962.
[26]. A. Gupta, J. D. Unadkat, Q. Mao, Interactions of azole antifungal agents with the human breast cancer resistance protein (BCRP),*J. Pharm.Sci*,**2007**, 96, 3226-35.
[27]. S. D. Schiller, H. B. Fung, Antimicrobial and antiurease activities of newly synthesized morpholine derivatives containing an azole nucleus, *Clinical Therapeutics*, **2008**, 29, 1862-1886.
[28]. H. A. Torres, R. Y. Hachem, R. F .Chemaly, D. P. Kontoyiannis, Posaconazole: a broad-spectrum triazole antifungal, *Lancet Infect Dis*, **2005**, 5, 775-85.
[29]. D. T. Hsieh,E. A. Thiele, Efficacy and safety of rufinamide in pediatric epilepsy, *Ther Adv Neuro Disord.*, **2013**,6, 189-98.
[30]. S. S. Joshi, A. V. Karnik, Rapid and efficient microwave-assisted synthesis of 4-amino-3-mercapto5-substituted-1,2,4-triazoles, *Indian J. Chem.*, **2006**, 45B, 1057-1059.
[31]. A. Kshirsagar, M. P. Toraskar, V. M. Kulkarni, S. Dhanashire,V. Kadam, *Int. J. Chem. Tech.Res.*, **2009**,1, 696-701.
[32]. A. K. Shirsagar, M. P. Toraskar, *Int. J.Chem.Tech. Res*, **2009**, 1, 696-701.

[33]. S. Ebrahimi, Synthesis of some pyridyl and cyclohexyl substituted 1,2,4 triazole, 1,3,4-thiadiazole and 1,3,4-oxadiazole derivatives, *Eur. J. Chem.*,**2010**, 1, 322-324.
[34]. I. Mir, M. T. Siddiqui, Antituberculosis agents. I. Alpha-(5-(2-Furyl)-1,2,4-triazol-3-ylthio) acethydrazide and related compounds, *Tetrahedron*, **1970**, 26, 5235-8.
[35]. K. Ilango, P. Valentina, Synthesis and biological activities of novel 1,2,4-triazolo-[3,4-b]-1,3,4-thiadiazoles, *Der Pharma. Chem.*, **2010**, 2, 16-22.
[36]. M. N. Zemtsova, P. L. Trakhtenberg, M. V. Galkina, A Procedure of 2-methylquinoline-4-carboxylic acids, *Russn. J. Org. Chem.*, **2003**, 49, 1803.
[37]. L. P. Carrod, F. D. Grady. Antibiotics and Chemotherapy, third ed, Churchill Livingstone Edinburg, **1972**, 477.
[38]. N. S. Habib, R. Soliman, A. E. Tombary, S. A. E. Hawash, G. Omaima, O. G. Shaaban, *Arch Pharm Res*., **2007**, 30, 1511-1520.
[39]. G. Shivaraja, S. Sreenivasa, A. R. Ramesha, T. M. Chakrapani Rao, H. Nagabhushana, Regioselective Synthesis, Antibacterial, Molecular Docking and Fingerprint Applications of 1-Benzhydrylpiperazine Derivatized 1,4-Disubstituted 1,2,3-Triazoles, *Chemistry Select.*, **2018**, 3, 8111 – 8117.
[40]. H. C. Anitha, S.Sreenivasa, N. R. Mohan, Vivek Chandramohan, G. Shivaraja,Preliminary Evaluation of Antimicrobial, Anthelmintic, Anti-Inflammatory Activities and In Silico Studies of Some Cinchoninic Acid Derivatives, *J. Applicable Chem.*, **2017**, 6, 543-558.
[41]. S. Sreenivasa, G. Shivaraja, V. Chandramohan, S. N. Shwetha, Aliyaarif, K. S. Guruprasad, *J. Applicable Chem.*, **2016**, 5, 1089-1096.
[42]. S. D. Sawant, A. A. Barvrkar, A. R. Chabukswar, S. D. Sarak, *J. Applicable Chem.*, **2013**, 2, 372- 384.
[43]. B. Sushma, Ch.V. Suresh, *J. Applicable Chem.*, **2012**, 1, 167-173.