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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis, analgesic, anti-inflammatory, and in vitro antimicrobial activities of some novel quinazolin-4(3*H*)-one derivatives

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Abstract With the aim of developing potent analgesic, anti-inflammatory, and antimicrobial agents a series of novel quinazolin-4(3*H*)-one derivatives were synthesized and characterized by FT-IR, ^{1}H-NMR, mass spectroscopy and bases of elemental analysis. Tail-flick technique, carrageenan-induced foot paw edema test, and agar streak dilution test were performed for screening analgesic, anti-inflammatory, and in vitro antimicrobial activity, respectively. Moreover, all compounds were examined for its ulcerogenicity. Results revealed that entire series of compounds exhibited mild to good analgesic, anti-inflammatory, and antimicrobial activity with low to moderate ulcer index. The relationship between the functional group variation and the biological activity of the evaluated compounds were discussed. Compound 2-(2-(4-(trifluoromethyl)benzylidene)hydrazinyl)-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl) phenyl) acetamide **5e** was determined to be the most active compound.

Keywords Quinazolin-4(3*H*)-one · Analgesic · Anti-inflammatory · Antimicrobial activity

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Introduction

Non-steroidal anti-inflammatory drugs (NSAIDs) represent a heterogeneous family of pharmacologically active compounds used to alleviate acute and chronic inflammation, pain, and fever. In the past decade, numerous advances have taken place in the understanding of pathogenesis and as a result, significant progress has been made and is still being made in the development of novel NSAIDs (Bhandari *et al*., 2009). The most important mechanism of NSAIDs is closely related to their ability to inhibit both isoforms of the enzyme cyclooxygenase (COX) also known as prostaglandin H_2 (PGH_2) synthase i.e., COX-1 and COX-2 because it catalyzes the conversion of arachidonic acid to PGH_2 (Dannhardt and Kiefer, 2001). The constitutive COX-1 isoform is mainly responsible for the synthesis of prostaglandins which exert cytoprotective effect in the gastrointestinal tract and control renal function in the kidneys; while the inducible COX-2 is selectively activated by pro-inflammatory stimuli and facilitates the release of prostaglandins involved in the inflammatory process (Sondhi *et al*., 2002). However, long term clinical usages of NSAIDs are associated with significant side effects such as severe gastrointestinal ulceration, bleeding, in tolerance and nephrotoxicity (Bertolini *et al*., 2002; Sherif *et al*., 2009). Therefore, investigation of new NSAIDs is still a major challenge and production of safer and more active NSAIDs are needed.

Concomitant use of several drugs to treat inflammatory conditions that might be associated with some microbial infections may cause serious health problems, especially in patients with impaired liver or kidney functions. In addition, from pharmacoeconomic viewpoint, and seeking better patient compliance, the discovery of an analgesic, anti-inflammatory, and antimicrobial agent with potential

activity with fewer adverse effects is a priority of the current global research, and consequently has occupied prime interest in recent years. Unfortunately, none of the drugs possesses these three activities in a single component. Therefore, our aim is to find a compound having all these three (analgesic, anti-inflammatory, and antimicrobial) activities. While searching for such a compound, we have found that quinazoline ring is one of the moieties on which studies have been concentrated.

Quinazolines and condensed quinazolines have received the attention of medicinal chemist due to their wide spectrum of biological activity as many, such as analgesic, anti-inflammatory (Alagarsamy *et al*., 2005), antimicrobial (Mohamed *et al*., 2010), anticonvulsant (Kumar *et al*., 2011), anticancer (Abdel Gawad *et al*., 2010), antitubercular (Pattan *et al*., 2006), antiviral (Dinakaran *et al*., 2003), and antihelmintic activities (Shukla and Shukla, 1989).

Being involved in a research program aiming at finding out new structure leads that would act as potent analgesic, anti-inflammatory, and antimicrobial agents, it was of interest to synthesize a novel series of quinazolin-4(3*H*)-one derivatives as a trial to obtain safer and potent analgesic, anti-inflammatory, and antimicrobial agents. The ulcerogenic activity of the compounds was also determined.

Materials and methods

Chemistry

All solvents used were of laboratory grade and were obtained from SD fine chemicals (Mumbai, India), and Merck (Mumbai, India). Ciprofloxacin and Ketoconazole are received as gift samples from Dr. Reddy's laboratories, Hyderabad, India. Melting points were determined in open glass capillary tubes and are uncorrected. Compounds were routinely checked for their purity on Silica gel G (Merck) Thin layer chromatography (TLC) plates; iodine chamber and UV lamp were used for visualization of TLC spots. The IR spectra were recorded in KBr pellets on (BIO-RAD FTS) FT-IR spectrophotometer. ^{1}H-NMR spectra were recorded on Bruker DPX-300 NMR spectrometer in $CDCl_3$ using tetramethylsilane (TMS) as an internal standard. The chemical shifts are reported in ppm scale. Mass spectra were obtained on a JEOL-SX-102 instrument using electron impact ionization. Elemental analyses were performed on a Perkin Elmer model 240C analyzer and were within ±0.4 % of the theoretical values.

*Synthesis of 2-methyl-4H-benzo-(1,3)-oxazin-4-one (**1**)*

For the synthesis of 2-methyl-4*H*-benzo-(1,3)-oxazin-4-one derivative, a mixture of anthranilic acid (1.37 g, 0.01 mol) and acetic anhydride (10.2 mL, 0.1 mol) was refluxed on gentle flame for 1 h. The excess of acetic anhydride was distilled off under reduced pressure and the residue was dissolved in petroleum ether and kept aside for 1 h. The light brown solid **1** which obtained was filtered and dried (Alagarsamy *et al*., 2003). Yield: 71 % m.p. 181–183 °C. IR: (KBr cm^{-1}) 3096 (Ar–CH), 2882 (CH_3–CH), 1712 (C=O), 1055 (C–O–C). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 6.92–7.40 (4H, m, Ar–C<u>H</u>), 2.38 (3H, s, C<u>H</u>$_3$). ESI–MS: MS: *m/z* 161. Anal. Cald for $C_9H_7NO_2$: C, 67.07; H, 4.38; N, 8.69. Found: C, 67.16; H, 4.40; N, 8.66.

*Synthesis of 3-(4-aminophenyl)-2-methyl-quinazolin-4(3H)-one (**2**)*

2-Methyl-4*H*-benzo-(1,3)-oxazin-4-one **1** (1.61 g, 0.01 mol) and *p*-phenylenediamine (1.08 g, 0.01 mol) was dissolved in 50 mL of anhydrous pyridine and heated on sand bath for 6 h. The resulting solution was cooled in ice bath and treated with 100 mL of dilute hydrochloric acid. The product thus separated **2** was filtered, washed with water, and crystallized from ethanol. Yield: 73 % m.p. 226–228 °C. IR: (KBr cm^{-1}) 3380 (NH), 3068 (Ar–CH), 2954 (CH_3–CH), 1730 (C=O). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 6.92–7.97 (8H, m, Ar–C<u>H</u>), 5.76 (2H, s, N<u>H</u>$_2$), 2.63 (3H, s, C<u>H</u>$_3$). ESI–MS: MS: *m/z* 251. Anal. Cald for $C_{15}H_{13}N_3O$: C, 71.70; H, 5.21; N, 16.72. Found: C, 71.92; H, 5.19; N, 16.67.

*Synthesis of 2-chloro-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**3**)*

3-(4-Aminophenyl)-2-methyl-quinazolin-4(3*H*)-one **2** (2.51 g, 0.01 mol) was dissolved in glacial acetic acid (50 mL) containing 50 mL of saturated solution of sodium acetate. The mixture was warmed to dissolve the substance completely. The solution was cooled in ice bath with stirring. A solution of chloroacetyl chloride (0.012 mol) was added drop wise to the above stirred solution so that the vigorous reaction did not take place. After half an hour the product separated is filtered. The product was washed with water and recrystallised using alcohol. Yield: 70 % m.p. 172–174 °C. IR: (KBr cm^{-1}) 3456 (NH), 3081 (Ar–CH), 2937 (CH_3–CH), 1746 (C=O quinazoline), 1662 (C=O amide), 772 (C–Cl). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.92 (1H, s, CON<u>H</u>), 6.95–8.01 (8H, m, Ar–C<u>H</u>), 4.38 (2H, s, C<u>H</u>$_2$), 2.56 (3H, s, C<u>H</u>$_3$). ESI–MS: MS: *m/z* 329 (M +2). Anal. Cald for $C_{17}H_{14}ClN_3O_2$: C, 62.30; H, 4.31; N, 12.82. Found: C, 62.11; H, 4.32; N, 12.86.

*Synthesis of 2-hydrazinyl-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**4**)*

Equimolar quantity of 2-chloro-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl)acetamide **3** (3.27 g, 0.01 mol)

and hydrazine hydrate (0.05 g, 0.01 mol) in 30 mL ethanol was refluxed for 10 h on a water bath. Cool the resulting solution, filtered, dried, and recrystallised from ethanol. Yield: 77 % m.p. 215–217 °C. IR: (KBr cm^{-1}) 3428 (NH), 3075 (Ar–CH), 2952 (CH_3–CH), 1723 (C=O quinazoline), 1650 (C=O amide). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.85 (1H, s, CONH), 7.07–8.19 (8H, m, Ar–CH), 5.62 (2H, s, NH_2), 4.51 (1H, s, CH_2NH), 4.06 (2H, d, CH_2), 2.39 (3H, s, CH_3). ESI–MS: MS: *m/z* 323. Anal. Cald for $C_{17}H_{17}N_5O_2$: C, 63.15; H, 5.30; N, 21.66. Found: C, 62.99; H, 5.32; N, 21.73.

General procedure for the synthesis of ***5a–5j***

Title compound **5a–5j** was synthesized by adding appropriate aromatic/heterocyclic aldehydes (0.01 mol) in fraction with the well stirred mixture of 2-hydrazinyl-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl)acetamide **4** (3.23 g, 0.01 mol) in 50 mL ethanol and 2–3 drops of glacial acetic acid. The pH was maintained to 5–6. The reaction mixture was then refluxed for a period of 4–6 h and kept aside for about 2 h. The products were separated by filtration, washed, and vacuum dried. Finally the products were recrystallized using ethanol to get pure form. The method used for the preparation and isolation of the compounds gave materials of good purity, as evidenced by their spectral analyses and by TLC. The title compounds are found to be soluble in chloroform, dimethyl sulfoxide, and dimethylformamide. The physicochemical properties of the synthesized compounds are given in Table 1.

*2-(2-Benzylidenehydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5a**)*

IR: (KBr cm^{-1}) 3454 (NH), 3037 (Ar–CH), 2973 (CH_3–CH), 1720 (C=O quinazoline), 1645 (C=O amide), 1531 (CH=N). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.94 (1H, s, CONH), 8.51 (1H, s, CH=N), 7.03–8.17 (13H, m, Ar–CH), 4.26 (2H, s, CH_2), 3.74 (1H, s, CH_2NH), 2.50 (3H, s, CH_3). ESI–MS: MS: *m/z* 411.

*2-(2-(4-Fluorobenzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5b**)*

IR: (KBr cm^{-1}) 3439 (NH), 3063 (Ar–CH), 2955 (CH_3–CH), 1747 (C=O quinazoline), 1674 (C=O amide), 1516 (CH=N), 1122 (C–F). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.87 (1H, s, CONH), 8.70 (1H, s, CH=N), 7.21–8.26 (12H, m, Ar–CH), 4.53 (2H, s, CH_2), 3.92 (1H, s, CH_2NH), 2.35 (3H, s, CH_3). ESI–MS: MS: *m/z* 429.

*2-(2-(4-Methylbenzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5c**)*

IR: (KBr cm^{-1}) 3462 (NH), 3055 (Ar–CH), 2988 (CH_3–CH), 1751 (C=O quinazoline), 1666 (C=O amide), 1547 (CH=N). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.90 (1H, s, CONH), 8.46 (1H, s, CH=N), 6.84–8.01 (12H, m, Ar–CH), 4.49 (2H, s, CH_2), 3.81 (1H, s, CH_2NH), 2.92 (3H, s, CH_3 benzylidene), 2.38 (3H, s, CH_3). ESI–MS: MS: *m/z* 425.

*2-(2-(4-Chlorobenzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5d**)*

IR: (KBr cm^{-1}) 3443 (NH), 3028 (Ar–CH), 2946 (CH_3–CH), 1734 (C=O quinazoline), 1642 (C=O amide), 1530 (CH=N), 775 (C–Cl). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.71 (1H, s, CONH), 8.65 (1H, s, CH=N), 7.12–8.08 (12H, m, Ar–CH), 4.62 (2H, s, CH_2), 3.94 (1H, s, CH_2NH), 2.47 (3H, s, CH_3). ESI–MS: MS: *m/z* 447 (M + 2).

*2-(2-(4-(Trifluoromethyl)benzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5e**)*

IR: (KBr cm^{-1}) 3435 (NH), 3046 (Ar–CH), 2961 (CH_3–CH), 1738 (C=O quinazoline), 1650 (C=O amide), 1515 (CH=N), 1137 (C–F). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.85 (1H, s, CONH), 8.33 (1H, s, CH=N), 7.04–8.19 (12H, m, Ar–CH), 4.38 (2H, s, CH_2), 3.72 (1H, s, CH_2NH), 2.56 (3H, s, CH_3). ESI–MS: MS: *m/z* 479.

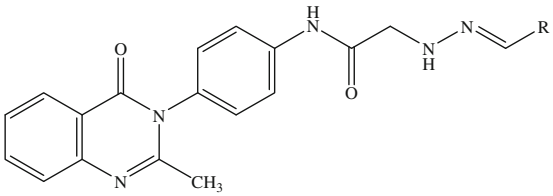
*2-(2-(4-Methoxybenzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5f**)*

IR: (KBr cm^{-1}) 3468 (NH), 3032 (Ar–CH), 2950 (CH_3–CH), 1755 (C=O quinazoline), 1679 (C=O amide), 1524 (CH=N), 1061 (C–O–C). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.93 (1H, s, CONH), 8.77 (1H, s, CH=N), 6.90–7.92 (12H, m, Ar–CH), 4.70 (2H, s, CH_2), 3.75 (1H, s, CH_2NH), 3.44 (3H, s, OCH_3), 2.31 (3H, s, CH_3). ESI–MS: MS: *m/z* 441.

*2-(2-(4-Nitrobenzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5g**)*

IR: (KBr cm^{-1}) 3450 (NH), 3074 (Ar–CH), 2989 (CH_3–CH), 1722 (C=O quinazoline), 1647 (C=O amide), 1542 (CH=N), 1505 and 1311 (C–NO_2). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.89 (1H, s, CONH), 8.62 (1H, s, CH=N), 7.09–8.20 (12H, m, Ar–CH), 4.55 (2H, s, CH_2), 3.98 (1H, s, CH_2NH), 2.57 (3H, s, CH_3). ESI–MS: MS: *m/z* 456.

Table 1 Structure and physicochemical properties of synthesized compounds **5a–5j**



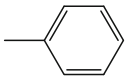
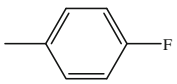
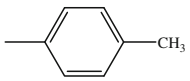
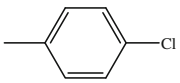
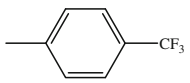
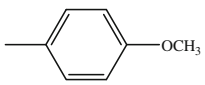
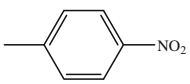
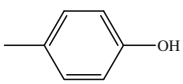
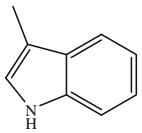
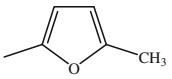
Compounds	R	m.p. (°C)	Yield (%)	Molecular formula	Log P^a	Analysis (%), found (calc.): C; H; N;
5a		232–234	70	$C_{24}H_{21}N_5O_2$	3.11	70.31 (70.06); 5.12 (5.14); 16.97 (17.02)
5b		251–253	78	$C_{24}H_{20}FN_5O_2$	3.27	66.95 (67.12); 4.70 (4.69); 16.26 (16.31)
5c		197–199	75	$C_{25}H_{23}N_5O_2$	3.60	70.81 (70.57); 5.43 (5.45); 16.41 (16.46)
5d		208–210	77	$C_{24}H_{20}ClN_5O_2$	3.67	64.87 (64.65); 4.51 (4.52); 15.67 (15.71)
5e		244–246	72	$C_{25}H_{20}F_3N_5O_2$	4.03	62.40 (62.63); 4.21 (4.20); 14.55 (14.61)
5f		188–190	79	$C_{25}H_{23}N_5O_3$	2.98	68.22 (68.01); 5.23 (5.25); 15.81 (15.86)
5g		262–264	71	$C_{24}H_{20}N_6O_4$	2.85	62.96 (63.15); 4.43 (4.42); 18.47 (18.41)
5h		203–205	75	$C_{24}H_{21}N_5O_3$	2.72	67.66 (67.44); 4.93 (4.95); 16.32 (16.38)

Table 1 continued

Compounds	R	m.p. (°C)	Yield (%)	Molecular formula	Log P^a	Analysis (%), found (calc.): C; H; N;
5i		276–278	72	$C_{26}H_{22}N_6O_2$	2.65	69.16 (69.32); 4.93 (4.92); 18.72 (18.66)
5j		239–241	76	$C_{23}H_{21}N_5O_3$	2.06	66.70 (66.49); 5.07 (5.09); 16.82 (16.86)

[a] Log *P* was calculated with Chem office 2009 software

*2-(2-(4-Hydroxybenzylidene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5h**)*

IR: (KBr cm^{-1}) 3530 (Ar–OH), 3426 (NH), 3051 (Ar–CH), 2977 (CH_3–CH), 1742 (C=O quinazoline), 1665 (C=O amide), 1538 (CH=N). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.82 (1H, s, CONH), 8.49 (1H, s, CH=N), 7.05–8.11 (12H, m, Ar–CH), 5.81 (1H, s, Ar–OH), 4.64 (2H, s, CH_2), 3.86 (1H, s, CH_2NH), 2.33 (3H, s, CH_3). ESI–MS: MS: *m/z* 427.

*2-(2-((1H-Indol-3-yl)methylene)hydrazinyl)-N-(4-(2-methyl-4-oxoquinazolin-3(4H)-yl)phenyl)acetamide (**5i**)*

IR: (KBr cm^{-1}) 3457 (NH), 3060 (Ar–CH), 2935 (CH_3–CH), 1754 (C=O quinazoline), 1652 (C=O amide), 1526 (CH=N). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.78 (1H, s, CONH), 9.03 (1H, s, NH indole), 8.31 (1H, s, CH=N), 6.92–7.98 (13H, m, Ar–CH), 6.06 (1H, s, CH_2NH), 4.25 (2H, s, CH_2), 2.42 (3H, s, CH_3). ESI–MS: MS: *m/z* 450.

*N-(4-(2-Methyl-4-oxoquinazolin-3(4H)-yl)phenyl)-2-(2-((5-methylfuran-2-yl)methylene)hydrazinyl)acetamide (**5j**)*

IR: (KBr cm^{-1}) 3431 (NH), 3059 (Ar–CH), 2943 (CH_3–CH), 1735 (C=O quinazoline), 1648 (C=O amide), 1534 (CH=N), 1072 (C–O–C). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ ppm: 9.95 (1H, s, CONH), 8.58 (1H, s, CH=N), 6.76–7.90 (10H, m, Ar–CH), 6.12 (1H, s, CH_2NH), 4.76 (2H, s, CH_2), 2.80 (3H, s, CH_3 furan), 2.37 (3H, s, CH_3). ESI–MS: MS: *m/z* 415.

Biological activities

Pharmacology

The synthesized compounds were evaluated for analgesic, anti-inflammatory, and ulcerogenic activities. One-way analysis of variance (ANOVA) was performed to certain the significance of all the exhibited activities. The test compounds and the standard drugs were administered in the form of a suspension (1 % carboxy methyl cellulose as a vehicle) by oral route of administration for analgesic and anti-inflammatory but for ulcerogenicity studies by intraperitoneally as suspension in 10 % v/v Tween-80. Each group consisted of six animals. The animals were maintained in colony cages at 25 ± 2 °C, relative humidity of 45–55 %, under a 12 h light and dark cycle; were fed standard animal feed (Olfert *et al*., 1993). All the animals were acclimatized for a week before use.

Analgesic activity

The analgesic activity was performed by tail-flick technique using Wistar albino mice (25–35 g) of either sex selected by random sampling technique (Kulkarni, 1980; D'Amour and Smith, 1941). Diclofenac sodium at a dose level of 10 and 20 mg/kg was administered orally as reference drug for comparison. The test compounds at two dose levels i.e., 10 and 20 mg/kg were administered orally. The reaction times were recorded immediately before and 30 min, 1, 2, and 3 h after the treatment and cut-off time was 10 s. The percent analgesic activity (PAA) was calculated by the following formula. PAA = $[T_2-T_1/10-T_1] \times 100$; where T_1 is the reaction time (s) before treatment, and T_2 is the reaction time (s) after treatment. The observed results are presented in Table 2.

Anti-inflammatory activity

Anti-inflammatory activity was evaluated by carrageenan-induced paw edema test in rats (Winter *et al*., 1962). Diclofenac sodium 10 and 20 mg/kg was administered as standard drug for comparison. The test compounds were

Table 2 PAA of the synthesized compounds (tail-flick method)

Compounds	Dose (mg/kg)	PAA			
		30 min	1 h	2 h	3 h
5a	10	30 ± 1.09*	35 ± 1.52*	41 ± 1.33**	29 ± 0.81*
	20	42 ± 1.23*	50 ± 0.81**	57 ± 2.16*	32 ± 1.45*
5b	10	32 ± 0.84**	38 ± 1.52*	42 ± 1.07*	31 ± 0.69**
	20	44 ± 1.78*	54 ± 1.01**	58 ± 1.53**	35 ± 1.15*
5c	10	37 ± 0.70*	42 ± 1.47**	47 ± 0.64*	37 ± 2.03**
	20	48 ± 0.46*	59 ± 0.92***	63 ± 1.39**	41 ± 1.03*
5d	10	36 ± 1.23***	45 ± 0.53*	48 ± 1.47*	35 ± 1.61*
	20	50 ± 0.84**	58 ± 1.82***	64 ± 0.68**	40 ± 1.27*
5e	10	40 ± 0.55**	48 ± 1.42*	53 ± 0.83***	39 ± 1.16**
	20	53 ± 1.84*	63 ± 0.69**	70 ± 1.22***	45 ± 0.53**
5f	10	24 ± 1.61**	32 ± 1.86*	35 ± 2.15*	23 ± 1.72*
	20	35 ± 0.84*	40 ± 1.29*	48 ± 0.60**	28 ± 1.77*
5g	10	26 ± 1.27*	31 ± 2.05**	36 ± 1.13*	25 ± 0.62*
	20	38 ± 1.48*	42 ± 0.64*	49 ± 2.06*	27 ± 1.25**
5h	10	23 ± 0.79*	30 ± 1.38*	34 ± 1.02**	20 ± 1.84*
	20	36 ± 1.33*	43 ± 1.94**	47 ± 0.51*	26 ± 0.76*
5i	10	19 ± 0.88*	25 ± 2.24*	29 ± 1.46*	18 ± 2.51*
	20	30 ± 2.13*	35 ± 0.81*	41 ± 1.15*	20 ± 0.78*
5j	10	17 ± 0.95*	24 ± 1.83*	27 ± 1.06*	19 ± 1.41*
	20	29 ± 1.29*	33 ± 0.74*	38 ± 1.31*	21 ± 1.76*
Control		4 ± 0.72	5 ± 0.49	6 ± 0.84	3 ± 0.41
Diclofenac	10	34 ± 1.18*	44 ± 1.69**	49 ± 1.06***	32 ± 0.62*
	20	47 ± 1.42**	55 ± 1.35**	64 ± 1.08***	41 ± 0.87*

Each value represents the mean ± SEM ($n = 6$)

Significance levels * $p < 0.5$, ** $p < 0.01$, *** $p < 0.001$ as compared with the respective control

administered at two dose levels of 10 and 20 mg/kg. The paw volumes were measured using the mercury displacement technique with the help of plethysmograph immediately before and 30 min, 1, 2, and 3 h after carrageenan injection. The percent inhibition of paw edema was calculated according to the following formula, percent inhibition $I = 100[1 - (a - x)/(b - y)]$, where x is the mean paw volume of rats before the administration of carrageenan and test compounds or reference compound (test group), a is the mean paw volume of rats after the administration of carrageenan in the test group (drug treated), b is the mean paw volume of rats after the administration of carrageenan in the control group, y is the mean paw volume of rats before the administration of carrageenan in the control group. All the percent inhibition results are shown in Table 3.

Ulcerogenicity

Ulceration in rats was induced as reported method (Goyal *et al.*, 1985). Albino rats of Wistar strain weighing 150–200 g of either sex were divided into various groups each of six animals. Control group of animals were administered only with 10 % v/v Tween-80 suspension intraperitoneally. One group was administered with aspirin intraperitoneally in a dose of 200 mg/kg once daily for 3 days. Diclofenac was also administered as standard drug at 20 mg/kg once daily for 3 days to another group of animals in the same route. The remaining group of animals was administered with test compounds intraperitoneally in a dose of 20 mg/kg. On fourth day, pylorus was ligated as per previous reported method (Shay *et al.*, 1945). Animals were fasted for 36 h before the pylorus ligation procedure. Four hours after the ligation, animals were sacrificed. The stomach was removed and opened along with the greater curvature. Ulcer index was determined by earlier reported method (Ganguly and Bhatnagar, 1973) and presented in Table 3.

Antimicrobial activity

In this study, all the synthesized compounds were screened for antimicrobial activity by agar streak dilution method. The antibacterial activity of the compounds were evaluated

Table 3 Percent anti-inflammatory activity (Carrageenan induced paw edema test in rats) and ulcer index of the synthesized compounds

Compounds	Dose (mg/kg)	Percent protection				Ulcer index
		30 min	1 h	2 h	3 h	
5a	10	24 ± 1.80**	29 ± 0.72*	34 ± 1.56**	28 ± 1.44*	0.70 ± 0.27
	20	35 ± 1.46*	46 ± 1.58**	51 ± 1.32*	32 ± 0.71*	
5b	10	26 ± 0.47*	33 ± 1.97*	36 ± 0.71**	29 ± 1.54**	0.64 ± 0.39
	20	37 ± 1.55*	49 ± 0.47**	52 ± 1.42*	36 ± 1.80**	
5c	10	30 ± 1.14*	35 ± 0.66**	41 ± 1.24*	31 ± 1.41*	0.61 ± 0.41
	20	41 ± 0.56*	54 ± 1.51*	60 ± 0.84***	39 ± 1.30**	
5d	10	31 ± 1.02***	37 ± 0.75*	43 ± 1.18*	34 ± 1.26*	0.59 ± 0.35
	20	43 ± 1.30*	52 ± 1.87*	59 ± 1.36**	42 ± 0.43***	
5e	10	35 ± 0.52*	40 ± 1.14**	46 ± 1.27***	39 ± 1.38*	0.55 ± 0.31
	20	46 ± 1.25*	57 ± 1.93***	65 ± 1.18***	47 ± 1.61**	
5f	10	22 ± 1.81*	25 ± 1.62*	29 ± 1.30**	19 ± 0.47**	0.86 ± 0.53
	20	31 ± 0.71**	37 ± 0.52*	42 ± 1.65*	30 ± 1.19*	
5g	10	20 ± 1.28*	26 ± 1.81**	31 ± 1.65*	20 ± 0.91*	0.80 ± 0.68
	20	29 ± 0.73*	36 ± 1.70*	40 ± 1.28*	31 ± 1.54**	
5h	10	21 ± 1.50*	25 ± 2.13*	28 ± 1.72**	22 ± 1.17*	0.82 ± 0.52
	20	30 ± 0.92**	39 ± 1.25*	43 ± 0.51*	29 ± 1.08*	
5i	10	20 ± 1.71*	23 ± 0.91**	29 ± 1.14*	21 ± 2.15*	0.72 ± 0.44
	20	28 ± 2.17*	36 ± 1.87*	41 ± 1.69*	32 ± 1.57*	
5j	10	19 ± 0.47*	25 ± 1.91*	30 ± 1.36*	23 ± 1.41*	0.77 ± 0.56
	20	31 ± 1.24*	35 ± 0.58*	39 ± 1.17**	30 ± 1.15*	
Control		4 ± 0.28	6 ± 0.63	5 ± 0.41	4 ± 0.75	0.13 ± 0.05
Diclofenac	10	31 ± 1.28*	36 ± 1.09**	42 ± 1.03***	29 ± 0.51*	1.61 ± 0.78
	20	40 ± 0.96*	51 ± 1.41*	60 ± 1.73***	43 ± 1.18**	
Aspirin		–	–	–	–	1.79 ± 0.62

Each value represents the mean ± SEM ($n = 6$)

Significance levels * $p < 0.5$, ** $p < 0.01$, *** $p < 0.001$ as compared with the respective control

against four Gram-positive bacteria *Staphylococcus aureus* ATCC 9144, *Staphylococcus epidermidis* ATCC 155, *Micrococcus luteus* ATCC 4698, and *Bacillus cereus* ATCC 11778 and three Gram-negative bacteria *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 2853, and *Klebsiella pneumoniae* ATCC 11298. The antifungal activities of the synthesized compounds were evaluated against two fungi *Aspergillus niger* ATCC 9029 and *Aspergillus fumigatus* ATCC 46645. Bacterial strains were cultured over night at 37 °C in Mueller–Hinton broth and the yeast was cultured overnight at 30 °C in YEPDE agar for antibacterial and antifungal activity tests. Test strains were suspended in nutrient agar to give a final density of 5×10^{-5} cfu/mL.

Minimum inhibitory concentration (MIC)

MIC of the compound was determined by agar streak dilution method (Hawkey and Lewis, 1994). A stock solution of the synthesized compound (100 μg/mL) in dimethyl formamide was prepared and graded quantities of the test compounds were incorporated in specified quantity of molten sterile agar (nutrient agar for antibacterial activity and Sabouraud's dextrose agar medium for antifungal activity). A specified quantity of the medium (40–50 °C) containing the compound was poured into a Petri dish to give a depth of 3–4 mm and allowed to solidify. Suspension of the micro-organism were prepared to contain approximately 5×10^{-5} cfu/mL and applied to plates with serially diluted compounds in dimethyl formamide to be tested and incubated at 37 °C for 24 and 48 h for bacteria and fungi, respectively. The MIC was considered to be the lowest concentration of the test substance exhibiting no visible growth of bacteria or fungi on the plate. The observed MIC is presented in Table 4.

Results and discussion

Chemistry

In this study, we synthesized a series of novel 2-methyl-quinazolin-4(3*H*)-one derivative by substituting different

Table 4 MIC (in μg/mL) of synthesized compounds

Compounds	*S. aureusus*	*S. epidermidis*	*M. luteus*	*B. cereus*	*E. coli*	*P. aeruginosa*	*K. pneumoniae*	*A. niger*	*A. fumigatus*
5a	31.25	31.25	62.5	31.25	62.5	31.25	15.62	31.25	31.25
5b	31.25	7.81	15.62	7.81	31.25	7.81	3.9	31.25	15.62
5c	125	62.5	125	31.25	62.5	62.5	31.25	62.5	125
5d	31.25	15.62	7.81	15.62	31.25	15.62	7.81	15.62	7.81
5e	15.62	15.62	7.81	3.9	31.25	7.81	7.81	7.81	15.62
5f	125	62.5	62.5	62.5	125	62.5	31.25	62.5	62.5
5g	31.25	7.81	15.62	7.81	31.25	15.62	7.81	15.62	15.62
5h	125	125	62.5	125	125	125	125	125	62.5
5i	62.5	31.25	31.25	31.25	62.5	15.62	15.62	31.25	62.5
5j	62.5	62.5	125	62.5	62.5	31.25	62.5	125	62.5
Ciprofloxacin	15.62	7.81	7.81	7.81	15.62	7.81	3.9	–	–
Ketoconazole	–	–	–	–	–	–	–	15.62	7.81

2-(2-methylenehydrazinyl)-*N*-phenylacetamide at 3rd position of quinazolin-4(3*H*)-one. Initially, anthranilic acid and acetic anhydride were used as starting materials to produce 2-methyl-4*H*-benzo-(1,3)-oxazin-4-one **1** by simple acetylation followed by ring closure reaction. Further this compound were treated with *p*-phenylenediamine gave 3-(4-aminophenyl)-2-methyl-quinazolin-4(3*H*)-one **2** with elimination of water. On stirring with chloroacetyl chloride in the presence of sodium acetate and glacial acetic acid, compound **2** gets converted to 2-chloro-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl)acetamide **3**. In the subsequent step, the compound **3** was treated with hydrazine hydrate results in 2-hydrazinyl-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl)acetamide **4**. Finally this compound **4** was treated with different aromatic/heterocyclic aldehydes in the presence of glacial acetic acid, and a variety of Schiff base derivatives have been isolated according to the synthetic Scheme 1.

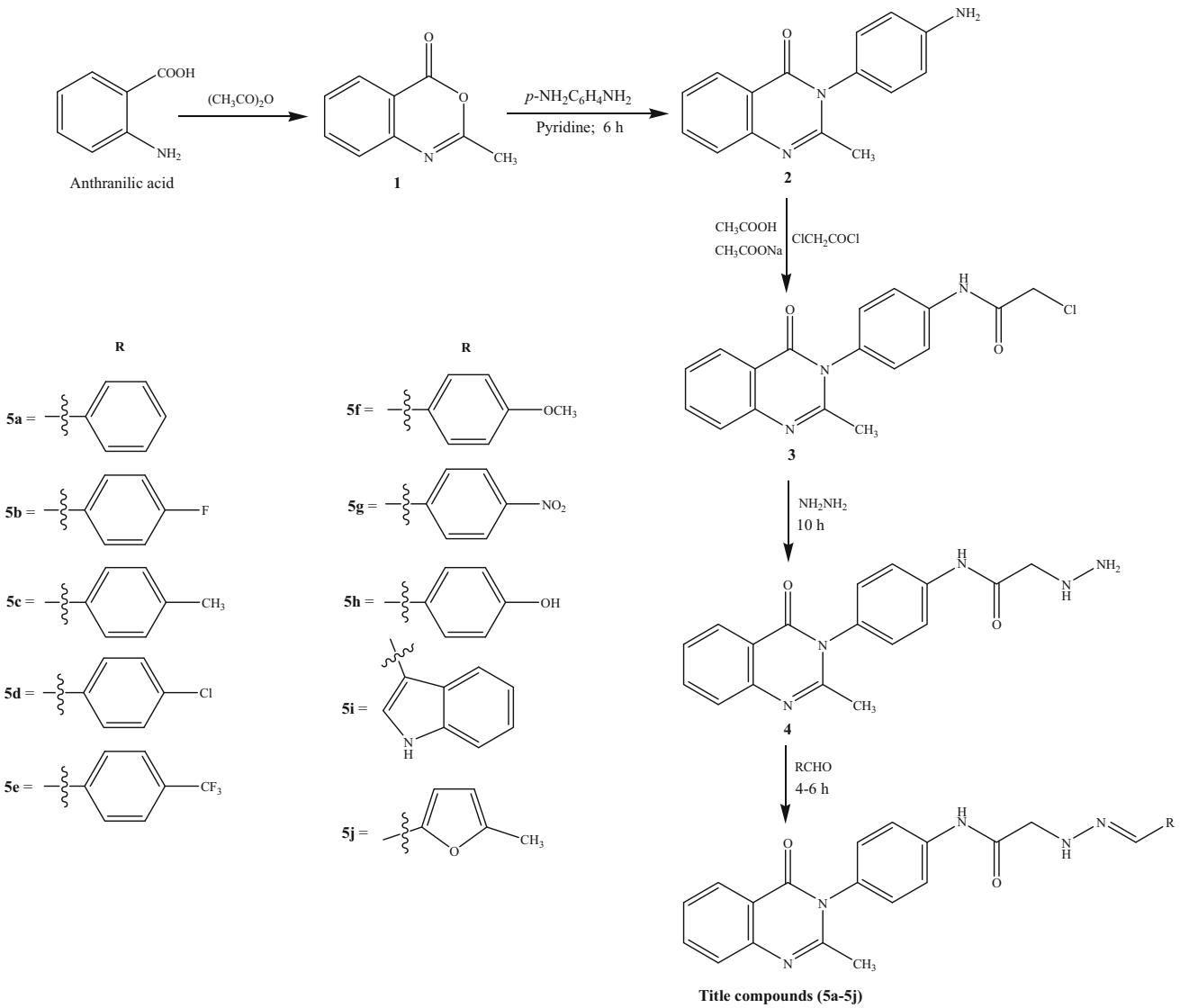
IR, ^{1}H-NMR, mass spectra and elemental analyses of the synthesized compounds are in accordance with the assigned structures. The IR spectra of all synthesized compounds showed some characteristic peaks indicating the presence of particular groups. Formation of 2-methyl-4*H*-benzo-(1,3)-oxazin-4-one **1** was confirmed by the presence of absorption peak at 1,712 and 1,055 cm^{-1} in IR due to presence of C=O and C–O–C stretching, respectively. The formations of compound **2** were confirmed by the absence of absorption bands around 1,050 cm^{-1} corresponds to C–O–C stretching and appearance of peak at 3,380 cm^{-1} in IR corresponds to N–H stretching of NH_2. Appearance of singlet at δ 5.76 ppm for two protons in its ^{1}H-NMR spectra which might be assigned to NH_2 group also confirms the formation of **2**. IR spectrum of compound **3** shows absorption peak at 1,662 and 772 cm^{-1} which might be due to C=O of amide and C–Cl, respectively, confirms the formation of compound **3**. This is further supported by the appearance of singlet for two protons at δ 4.38 ppm in its ^{1}H-NMR spectra which might be assigned to CH_2 group of acetamide. ^{1}H-NMR spectra of compound **4** showed a singlet peaks at δ 4.51 and δ 5.62 ppm corresponds to NH and NH_2 of CH_2NHNH_2, respectively, confirms its formation. The title compounds **5a–5j** showed absorption bands at 3426–3468, 1720–1755 cm^{-1}, and weak band at 1515–1547 cm^{-1}, which can be assignable to NH, C=O of quinazoline, and CH=N (azomethine linkage) vibrations, respectively. The proton magnetic resonance spectrums of synthesized compounds were recorded in $CDCl_3$. The following conclusions can be derived by comparing the NMR spectra of compounds **5a–5j**:

- A singlet at δ 2.31–2.57 ppm for C<u>H</u>$_3$,
- A singlet at δ 4.25–4.76 ppm for C<u>H</u>$_2$,
- A singlet at δ 3.72–6.12 ppm for CH$_2$N<u>H</u>,
- A multiplet at δ 6.76–8.26 ppm for Ar–C<u>H</u>,
- A singlet at δ 8.31–8.77 ppm for C<u>H</u>=N,
- A singlet at δ 9.71–9.95 ppm for CON<u>H</u>.

Biological activities

Analgesic activity

Entire test compounds **5a–5j** was tested for their analgesic activity by tail-flick technique using Wistar albino mice. The results of analgesic study are summarized in Table 2. The reports indicate that all the test compounds exhibited significant activity and graded dose response. Moreover, this study revealed that test compounds showed moderate analgesic activity at 30 min of reaction time; the activity increased at 1 h, further it reached to peak level at 2 h and past its best in activity was observed at 3 h. Compound **5a** with unsubstituted phenyl derivative showed moderate analgesic activity compared to standard drug diclofenac



Scheme 1 Schematic representation of the synthesized compounds

sodium. With the increased lipophilicity *p*-fluoro derivative **5b** showed an increase in activity. Replacement of fluorine with methyl or chlorine further increases the lipophilicity results in enhanced activity which was found to be equipotent with reference standard diclofenac. Exchange of methyl or chlorine with trifluoromethyl further increases the lipophilicity led to moderate increase in activity which was found to be more potent than standard drug tested. Exchange of trifluoromethyl by methoxy or nitro or hydroxyl results in sharp fall in activity may be due to decreased lipophilicity. Introduction of heterocyclic moiety such as indole and furan instead of *p*-(methoxy/nitro/hydroxyl)phenyl group further decreases the activity. Compound 2-(2-(4-(trifluoromethyl)benzylidene)hydrazinyl)-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl)acetamide **5e** was found to be the most active analgesic agent and it is moderately more potent when compared to the reference standard diclofenac sodium.

Anti-inflammatory activity

Carrageenan-induced paw edema test was performed to assess the anti-inflammatory activity of test compounds using Wistar rats. The anti-inflammatory activity results (Table 3) showed that all the test compounds protected rats from carrageenan-induced inflammation reasonably at 30 min of reaction time; the activity increased at 1 h and it reached to maximum level at 2 h. Declining in activity was observed at 3 h. The compounds possessing unsubstituted phenyl ring **5a** exhibited moderate anti-inflammatory activity when compared to the reference standard diclofenac sodium. With increased lipophilicity the compound

with *p*-fluoro substituent **5b** showed moderately more activity than **5a**. Methyl and chlorine derivative **5c** and **5d** showed equipotent activity with reference standard diclofenac sodium. Among all tested compounds trifluoromethyl analog **5e** exhibited better activity which is more potent than diclofenac. A deep fall in activity was observed when trifluoromethyl group **5e** was replaced with methoxy or nitro or hydroxyl group **5f–5h**. Unlike analgesic activity replacement of these *p*-(methoxy/nitro/hydroxyl)phenyl group by heterocyclic moiety **5i–5j** retains the activity.

Ulcerogenicity

Further all the test compounds were examined for its ulcerogenicity and the results are summarized in Table 3. Entire test compounds exhibited less ulcer index compared to standard diclofenac and aspirin. Results of ulcer index revealed that the compounds bearing halogen and methyl substituents **5b–5e** showed negligible ulcer index, whereas replacement of *p*-(halogen/methyl)phenyl by phenyl/heterocyclic moiety leads to slight increases in ulcer index. Over other test compounds, **5f–5h** possessing methoxy or nitro or hydroxyl group exhibited higher ulcer index. The test compounds exhibited 34–53 and 31–48 % of the ulcer index when compared to the reference drug diclofenac (1.61 ± 0.78) and aspirin (1.79 ± 0.62), respectively. Among the tested compounds, 2-(2-(4-(trifluoromethyl) benzylidene)hydrazinyl)-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl) acetamide **5e** exhibited least ulcer index (0.55 ± 0.31) which is about one-third of the ulcer index of reference standards. Out of entire tested compounds, 2-(2-(4-methoxybenzylidene)hydrazinyl)-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl) phenyl)acetamide **5f** was found to possess highest ulcer index (0.86 ± 0.53) which is about 51 % of the ulcer index of diclofenac and aspirin.

Antimicrobial activity

All the synthesized compounds were evaluated for their in vitro antibacterial and antifungal activity by agar streak dilution method. A comparison of antimicrobial activity of the synthesized compounds with that of standard drugs was effectively presented in Table 4. The MICs of Ciprofloxacin and Ketoconazole were determined in parallel experiments to control the sensitivity of the test organisms. As seen in Table 4, except **5e** (MIC: 15.62 μg/mL) all other compounds showed lower activities (MIC: 31.25–125 μg/mL) than Ciprofloxacin against *S. aureus*. Compounds **5b** and **5g** displayed the equal activity (MIC: 7.81 μg/mL) against *S. epidermidis*, whereas rest of series exhibited lower activity (MIC: 15.62–125 μg/mL). Against *M. luteus* compounds **5d** and **5e** exhibited comparable activity (MIC: 7.81 μg/mL) as Ciprofloxacin, while others demonstrated lesser activity than standard. All compounds except **5b**, **5e**, and **5g** showed worse activities than standard against *B. cereus*, compounds **5b** and **5g** showed the same activity (MIC: 7.81 μg/mL); moreover **5e** demonstrated better activity (MIC: 3.9 μg/mL) than Ciprofloxacin. All the compounds exhibited lower activity against *E. coli* (MIC: 31.25–125 μg/mL). Compounds **5b** and **5e** displayed the equivalent activity (MIC: 7.81 μg/mL) as standard against *P. aeruginosa*. Only compound **5b** showed the same activity (MIC: 3.9 μg/mL) as Ciprofloxacin against *K. pneumoniae*, while rest of test compounds displayed poorer activity. Against *A. niger* compound **5e** demonstrated better activity (MIC: 7.81 μg/mL) than Ketaconazole, while compounds **5d** and **5g** exhibited similar activity (MIC: 15.62 μg/mL) as Ketaconazole; remaining compounds showed inferior activity (MIC: 31.25–125 μg/mL). Compound **5d** showed similar activity (MIC: 7.81 μg/mL) against *A. fumigatus* while the others have lower activity (MIC: 15.62–125 μg/mL) than standard.

The current results revealed that most of the synthesized derivatives exhibited significant antimicrobial activity. The potent antibacterial and antifungal activity exhibited by compounds **5b**, **5d**, **5e**, and **5g** might be due to the presence of electron withdrawing substituent like fluoro, chloro, trifluoromethyl, and nitro group, respectively. While other compounds, though they contain electron donating substituents like methyl, methoxy, and hydroxyl groups (**5c**, **5f**, and **5h**) exhibited less in vitro antimicrobial activity. The unsubstituted compounds showed moderate antimicrobial activity. The MIC values were determines as the lowest concentration that totally inhibited visible growth of the microorganisms. The chemical structure and antimicrobial activity relationship of the synthesized compounds revealed that the compounds having electron withdrawing moiety exhibited better activity when compared with compounds having electron releasing moieties. Among tested compounds, 2-(2-(4-(trifluoromethyl)benzylidene) hydrazinyl)-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl)phenyl) acetamide **5e** exhibited better activity against *B. cereus* and *A. niger*; while it displayed equal activity as standard against *S. aureus*, *M. leutus*, and *P. aeruginosa.*

Conclusion

In summary, a series of novel quinazolin-4(3*H*)-one derivative were synthesized and characterized by FT-IR, ^{1}H-NMR, Mass spectroscopy, and elemental analysis. These derivatives were evaluated for their analgesic, anti-inflammatory, ulcerogenicity, and in vitro antimicrobial activity. In general, halogen substituted compounds particularly trifluoromethyl analog exhibited potent analgesic and anti-inflammatory activity with negligible ulcer index.

Moreover, electron withdrawing group substituted derivatives showed remarkable antimicrobial properties than electron releasing group substituted compounds. Among several tested compounds, 2-(2-(4-(trifluoromethyl) benzylidene) hydrazinyl)-*N*-(4-(2-methyl-4-oxoquinazolin-3(4*H*)-yl) phenyl)acetamide **5e** showed better analgesic and anti-inflammatory activity which is more potent than reference standard diclofenac. Interestingly this derivative possessed about 32 % of the ulcer index of reference standards. In addition, compound **5e** also showed some excellent antimicrobial activity against *S. aureus*, *M. leutus*, *B. cereus*, *P. aeruginosa*, and *A. niger*. Hence, this analog could be developed as a new class of analgesic, anti-inflammatory, and antimicrobial agents. However, further structural modification is planned to enhance these activities with the low ulcerogenic index.

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References

Abdel Gawad NM, Georgey HH, Youssef RM, El-Sayed NA (2010) Synthesis and antitumor activity of some 2,3-disubstituted quinazolin-4(3*H*)-ones and 4,6-disubstituted-1,2,3,4-tetrahydroquinazolin-2*H*-ones. Eur J Med Chem 45:6058–6067

Alagarsamy V, Muruganantham G, Venkateshaperumal R (2003) Synthesis, analgesic, anti-inflammatory and antibacterial activities of some novel 2-methyl-3-substituted quinazolin-4(3*H*)-ones. Biol Pharm Bull 26:1711–1714

Alagarsamy V, Rajasolomon V, Meena R, Ramseshu KV (2005) Synthesis, analgesic, anti-inflammatory and antibacterial activities of some novel 2-butyl-3-substituted quinazolin-4(3*H*)-ones. Biol Pharm Bull 28:1091–1094

Bertolini A, Ottani A, Sandrini M (2002) Selective COX-2 inhibitors and dual acting anti-inflammatory drugs: critical remarks. Curr Med Chem 9:1033–1043

Bhandari SV, Dangre SC, Bothara KG, Patil AA, Sarkate AP, Lokwani DK (2009) Design, synthesis and pharmacological screening of novel nitric oxide donors containing 1,5-diarylpyrazolin-3-one as nontoxic NSAIDs. Eur J Med Chem 44:4622–4636

D'Amour FE, Smith DL (1941) A method for determining loss of pain sensation. J Pharmacol Exp Ther 72:74–79

Dannhardt G, Kiefer W (2001) Cyclooxygenase inhibitors-current status and future prospects. Eur J Med Chem 36:109–126

Dinakaran M, Selvam P, Declercq E, Sridhar SK (2003) Synthesis, antiviral and cytotoxic activity of 6-bromo-2,3-disubstituted-4(3*H*)-quinazolinones. Biol Pharm Bull 26:1278–1282

Ganguly AK, Bhatnagar OP (1973) Effect of bilateral adrenalectomy on production of restraint ulcers in the stomach of albino rats. Can J Physiol Pharmacol 51:748–750

Goyal RK, Chakrabarti A, Sanyal AK (1985) The effect of biological variables on the anti ulcerogenic effect of vegetable plantain banana. Planta Med 29:85–88

Hawkey PM, Lewis DA (1994) Medical bacteriology-a practical approach. Oxford University Press, Oxford, pp 181–194

Kulkarni SK (1980) Heat and other physiological stress-induced analgesia: catecholamine mediated and naloxone reversible response. Life Sci 27:185–188

Kumar P, Shrivastava B, Pandeya SN, Stables JP (2011) Design, synthesis and potential 6 Hz psychomotor seizure test activity of some novel 2-(substituted)-3-{[substituted]amino}-quinazolin-4(3*H*)-one. Eur J Med Chem 46:1006–1018

Mohamed MS, Kamel MM, Kassem EMM, Abotaleb N, Abd El-moez SI, Ahmed MF (2010) Novel 6,8-dibromo-4(3*H*)-quinazolinone derivatives of antibacterial and antifungal activities. Eur J Med Chem 45:3311–3319

Olfert ED, Cross BM, McWilliam AA (1993) Canadian council of animal care guide to the care and use of experimental animals, vol 1, 2nd edn. Canadian Council on Animal Care, Ottawa

Pattan SR, Krishna Reddy VV, Manvi FV, Desai BG, Bhat AR (2006) Synthesis of N-3[4(4-chlorophenylthiazol-2-yl)-2-aminomethyl]quinazolin-4(3*H*)-one and their derivatives for antitubercular activity. Indian J Chem 45B:1778–1781

Shay M, Komarov SA, Fels D, Meranze D, Grunstein H, Siplet H (1945) A simple method for the uniform production of gastric ulceration in the rats. Gastroenterology 5:43–61

Sherif AFR, Ibrahim MElA, Heba AAElR, Mona HB, Hayam MAA (2009) Design and synthesis of some thiazolyl and thiadiazolyl derivatives of antipyrine as potential non-acidic anti-inflammatory, analgesic and antimicrobial agents. Eur J Med Chem 17:882–895

Shukla JS, Shukla R (1989) Synthesis of 4-(5-substitutedarylidine-4-thiazolidinone-2-thione)-6,8-substituted quinazolines as potential antihelmintic agents. J Indian Chem Soc 66:209–210

Sondhi SM, Singhal N, Johar M, Reddy BSN, Lown JW (2002) Heterocyclic compounds as inflammation inhibitors. Curr Med Chem 9:1045–1074

Winter CA, Risley EA, Nuss GW (1962) Carrageenin induced edema in hind paw of the rat as an assay for anti-inflammatory drugs. Exp Biol Med 111:544–547