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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis and pharmacological investigations of novel 2-phenylquinazolin-4(3*H*)-one derivatives

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Abstract A series of novel 2-phenyl-3-(4-(5-substituted-phenylisoxazol-3-yl)phenyl)quinazolin-4(3*H*)-one **5a–5o** were designed and synthesized from anthranilic acid. All the synthesized compounds were 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 of biological studies revealed that all title 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 was discussed. Out of fifteen title compounds, 2-phenyl-3-(4-(5-(4-(trifluoromethyl)phenyl)isoxazol-3-yl)phenyl) quinazolin-4(3*H*)-one **5f** was found to be the most active compound.

Keywords Quinazoline · Isoxazole · Analgesic · Anti-inflammatory · Antimicrobial activity

Introduction

Pain is widely accepted to be one of the most important determinations of quality of life. A study reported by the world health organization demonstrated that individuals who live with persistent pain suffer fourfold more from depression (or) anxiety compared to healthy subjects (Gureje *et al*., 1998). The identification of compounds able to treat both acute and chronic pain with limited effects is one of the prominent goals in biomedical research.

Inflammation is a defensive, but exaggerated local tissue reaction in response to exogenous or endogenous insult. It is a fundamental physiological process that is essential for survival, but at the same time is one of the major causes of human morbidity and mortality (O'Neill, 2006; Cheeseright *et al*., 2009). A large number of non steroidal anti-inflammatory drugs (NSAIDs) are available clinically to treat inflammatory disorders (Van Ryn and Botting, 1995). However, long-term clinical usage of NSAIDs is associated with significant side effects such as gastrointestinal (GI) symptoms including mucosal damage, bleeding, nausea, heartburn, dyspepsia, abdominal pain, and renal toxicity (Van Ryn *et al*., 2000). Consequently, developing new therapeutic agents that can overcome gastrointestinal injury and at the same time could lead to an enhanced anti-inflammatory effect became an urgent need for inflammation patients (Van Ryn, 1971; Beuck, 1999).

Microbial infections are a growing problem in contemporary medicine, and the use of antibiotics is unavoidable (Chopra *et al*., 2008). The global sales of antibiotics are generally higher when compared to other drugs which are

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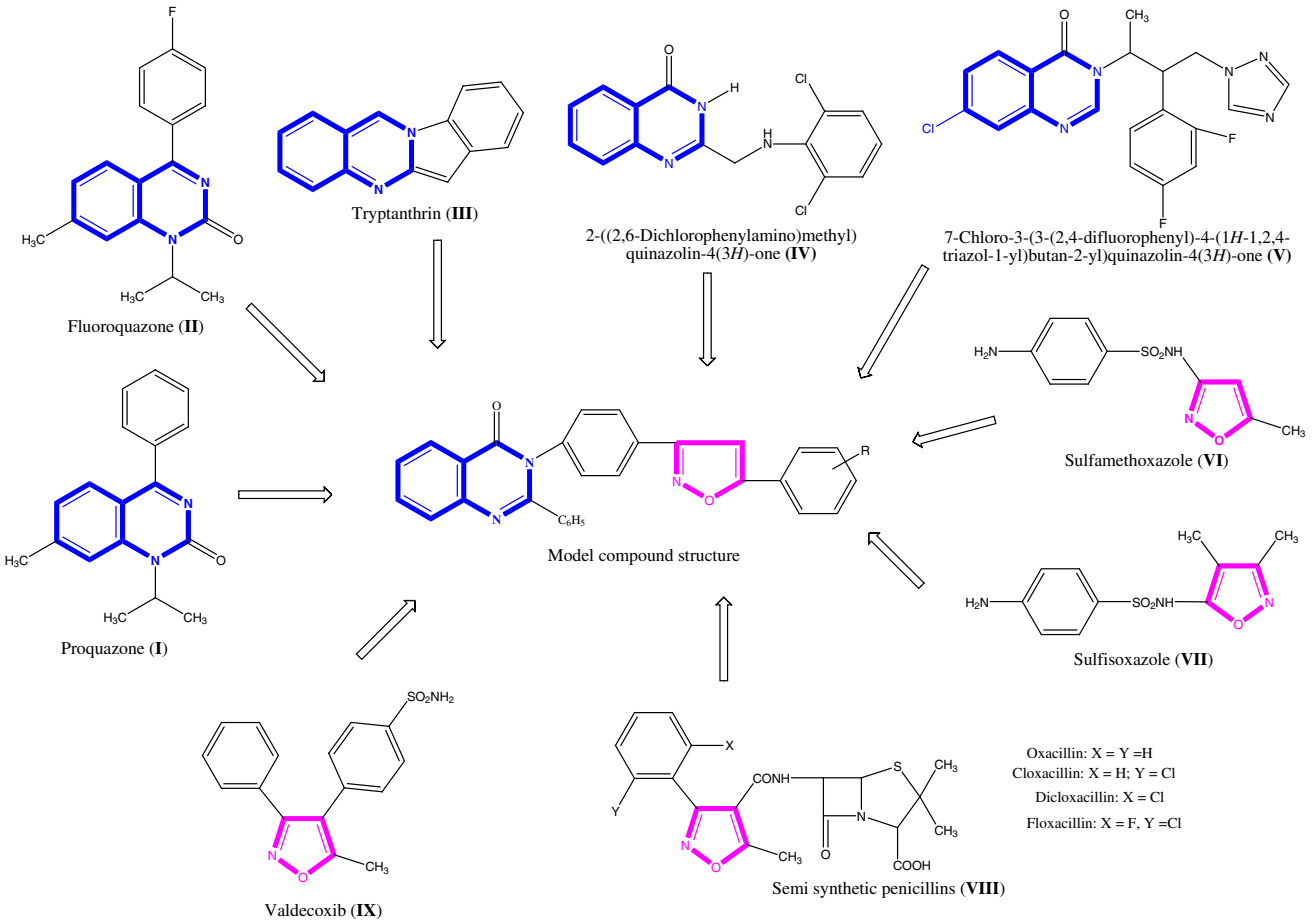


Fig. 1 Examples of structural similarities between known NSAIDs, antibiotics and 2-phenyl-3-(4-(5-substitutedphenylisoxazol-3-yl)phenyl)quinazolin-*4*(3*H*)-one (Model compound structure)

prescribed. Antibiotic resistance is a major problem in hospitals, as well as in community settings. Despite the many antibiotics and chemotherapeutics available, the emergence of old and new antibiotic-resistant microbial strains in the last decades constitutes a substantial need for new classes of antimicrobial agents (Devasia *et al*., 2006; Spellberg *et al*., 2008).

Bacterial infection often produces inflammation and pain. In normal practice, two group of agents (chemotherapeutic and NSAID) are prescribed simultaneously. The compounds possessing all these activities are not common. Quinazolines and condensed quinazolines have received the attention of medicinal chemist due to their wide spectrum of biological activities 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). Additionally, different known anti-inflammatory drugs such as proquazone **I** (Vanryzin and Trpold, 1980), fluproquazone **II** (Wheathly, 1982), and tryptanthrin **III** (Fankhauser *et al*., 1981; Cheristine and Matthias, 2004) are bearing quinazoline nucleus. Diverse examples of antimicrobial quinazolinone are presented in Fig. 1. 2-((2,6-Dichlorophenylamino)methyl)quinazolin-4(3*H*)-one **IV** exerted antibacterial effect against *Staphylococcus aureus* in a very low concentration (Jantova *et al*., 2004). 7-Chloro-3-(3-(2,4-difluorophenyl)-4-(1*H*-1,2,4-triazol-1-yl)butan-2-yl)quinazolin-4(3*H*)-one **V** exhibited high in vivo activity, low toxicity, and good pharmacokinetic profile (Bartroli *et al*., 1998).

On the other hand, isoxazole have gained importance because of the physiological and pharmacological activities associated with them. Moreover, antimicrobial drugs such as sulphamethoxazole **VI**, sulfisoxazole **VII** and semi synthetic penicillin (such as oxacillin, cloxacillin, dicloxacillin, and floxacillin) **VIII**, and NSAID valdecoxib **IX** possess isoxazole nucleus are represented in Fig. 1 (Panda *et al*., 2009; Gupta *et al*., 1999). In the present study, it was envisaged that a drug molecule possessing the above-mentioned pharmacophore could be advantage, since it might possess analgesic, anti-inflammatory, and antimicrobial activity. In view of the facts mentioned above and

as part of our efforts to discover potentially active new agents, various quinazolin-4(3*H*)-one derivatives **5a–5o** were synthesized and evaluated for their analgesic, anti-inflammatory, and antimicrobial activity.

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 were received as gift samples from Dr. Reddys 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. NMR spectra were recorded on Bruker Avance-500 NMR spectrometer in $CDCl_3$ using tetramethylsilane for ^1H NMR and $CDCl_3$ for ^{13}C NMR 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 Perkine Elmer model 2400 CHN analyzer and were within ±0.4 % of the theoretical values.

General procedure for the synthesis of title compounds

*Synthesis of 2-phenyl-4H-benzo-(1,3)-oxazin-4-one (**2**)* To a solution of anthranilic acid (13.7 g, 0.1 mol) dissolved in pyridine (60 ml), benzoyl chloride (28 g, 0.2 mol) was added to synthesize 2-phenyl-4*H*-benzo-(1,3)-oxazin-4-one. The mixture was stirred for 30 min followed by treatment with 5 % $NaHCO_3$ (15 ml). The solid thus obtained **2** was recrystallized from ethanol (Alagarsamy *et al*., 2002).

*Synthesis of 3-(4-acetylphenyl)-2-phenylquinazolin-4(3H)-one (**3**)* 2-Phenyl-4*H*-benzo-(1,3)-oxazin-4-one **2** (2.23 g, 0.01 mol) and *p*-aminoacetophenone (1.35 g, 0.01 mol) were dissolved in 50 ml of anhydrous pyridine and heated on sand bath for 8 h. The resulting solution was cooled in ice bath and treated with 100 ml of dilute hydrochloric acid. The product thus separated **3** was filtered, washed with water, and crystallized from ethanol. Yield 77 %, m.p. 205–207 °C. IR (KBr) cm^{-1}: 3049 (Ar–CH), 2,914 (CH_3-CH), 1,731 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 2.88 (s, 3H, $C\underline{H}_3$), 6.92–8.07 (m, 13H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 207.8 (C=O), 172.7 (C-2), 166.2 (C-4), 160.9 (C-9), 149.2 (C″-1), 144.6 (C-7), 143.6 (C″-4), 140.3 (C-5), 139.5 (C′-1), 135.6 (C′-4), 130.9 (C′-3), 130.2 (C″-3), 128.5 (C′-5), 127.8 (C″-5), 126.0 (C′-6), 125.1 (C″-6), 122.4 (C′-2), 121.3 (C″-2), 117.6 (C-6), 114.4 (C-8), 108.2 (C-10), 35.7 (CH_3). MS: *m/z* 340 [M^+]. Anal. Cald for $C_{22}H_{16}N_2O_2$: C, 77.63; H, 4.74; N, 8.23. Found: C, 77.35; H, 4.76; N, 8.26.

*Synthesis of 2-phenyl-3-(4-(3-substitutedphenylacryloyl) phenyl)quinazolin-4(3H)-one (**4a–4o**)* A mixture of 3-(4-acetylphenyl)-2-phenylquinazolin-4(3*H*)-one **3** (3.40 g, 0.01 mol) and different aromatic aldehydes (0.01 mol) was dissolved in a minimum quantity of ethanol. To this few drops of 10 % sodium hydroxide solution were added and stirred for the period of 4 h and further refluxed for 2 h. Then the reaction mixture was poured in crushed ice and stirred. The product separated out **4a–4o** was filtered, washed with water, dried, and recrystallized from ethanol.

*2-Phenyl-3-(4-(3-phenylacryloyl)phenyl)quinazolin-4(3H)-one (**4a**)* Yield 77 %, m.p. 231–233 °C. IR (KBr) cm^{-1}: 3,084 (Ar–CH), 1,725 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.53–7.79 (m, 18H, Ar–C<u>H</u>), 8.02–8.35 (m, 2H, C<u>H</u>=C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 199.5 (C=O), 170.9 (C-2), 164.4 (C-4), 162.3 (C-9), 159.7 (CH=CH), 145.1 (C″-1), 143.8 (C‴-1), 142.2 (C-7), 139.9 (C′-1), 138.4 (C″-4), 138.0 (C‴-4), 135.3 (C-5), 132.8 (C′-4), 131.6 (C″-3), 129.2 (C′-3), 125.7 (C‴-3), 126.1 (C″-5), 125.9 (C′-5), 124.0 (C‴-5), 120.4 (C‴-6), 119.5 (C′-6), 117.6 (C″-6), 116.8 (C‴-2), 115.3 (C′-2), 112.0 (C″-2), 110.6 (C-6), 109.9 (C-8), 105.5 (C-10), 103.9 (CH=CH). MS: *m/z* 428 [M^+]. Anal. Cald for $C_{29}H_{20}N_2O_2$: C, 81.29; H, 4.70; N, 6.54. Found: C, 81.52; H, 4.68; N, 6.52.

*3-(4-(3-(4-Methoxyphenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4b**)* Yield 71 %, m.p. 175–177 °C. IR (KBr) cm^{-1}: 3,106 (Ar–CH), 1,730 (C=O), 1,063 (C–O-C). ^1H-NMR ($CDCl_3$, 500 MHz) δ ppm: 3.41 (s, 3H, O<u>CH</u>$_3$), 7.06–8.12 (m, 17H, Ar–C<u>H</u>), 8.20–8.49 (m, 2H, C<u>H</u>=C<u>H</u>). ^{13}C-NMR ($CDCl_3$, 500 MHz) δ ppm: 202.3 (C=O), 179.5 (C-2), 170.8 (C-4), 162.2 (C‴-4), 159.4 (C-9), 158.1 (CH=CH), 149.1 (C″-1), 146.6 (C′-1), 143.8 (C‴-1), 140.1 (C-7), 139.0 (C″-4), 136.8 (C-5), 135.5 (C′-4), 133.7 (C″-3), 132.4 (C′-3), 129.9 (C″-5), 128.3 (C′-5), 125.4 (C‴-6), 122.0 (C′-6), 121.5 (C″-6), 119.8 (C‴-2), 118.9 (C′-2), 118.1 (C″-2), 114.2 (C-6), 112.9 (C-8), 109.6 (C-10), 107.3 (CH=CH), 105.4 (C‴-3), 104.6 (C‴-5), 67.8 (OCH_3). MS: *m/z* 458 [M^+]. Anal. Cald for $C_{30}H_{22}N_2O_3$: C, 78.59; H, 4.84; N, 6.11. Found: C, 78.30; H, 4.85; N, 6.13.

*3-(4-(3-(4-Aminophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4c**)* Yield 74 %, m.p. 198–200 °C. IR (KBr) cm^{-1}: 3,347 (NH), 3,060 (Ar–CH), 1,712 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.25 (s, 2H, N<u>H</u>$_2$), 6.

91–8.10 (m, 17H, Ar–CH), 8.18–8.42 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 198.6 (C=O), 170.1 (C-2), 168.4 (C-4), 163.2 (C-9), 160.7 (C‴-4), 155.9 (CH=CH), 149.3 (C″-1), 146.4 (C-7), 145.9 (C″-4), 141.6 (C-5), 140.2 (C′-4), 137.0 (C″-3), 135.8 (C′-3), 132.2 (C″-5), 131.5 (C′-5), 129.0 (C′-1), 126.6 (C‴-6), 125.9 (C′-6), 124.3 (C″-6), 122.7 (C‴-1), 119.2 (C‴-2), 117.5 (C′-2), 114.9 (C″-2), 113.0 (C-6), 110.5 (C-8), 109.2 (C-10), 105.7 (CH=CH), 102.6 (C‴-3), 101.4 (C‴-5). MS: *m/z* 443 [M^+]. Anal. Cald for $C_{29}H_{21}N_3O_2$: C, 78.54; H, 4.77; N, 9.47. Found: C, 78.33; H, 4.75; N, 9.50.

*3-(4-(3-(4-Hydroxyphenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4d**)* Yield 75 %, m.p. 189–192 °C. IR (KBr) cm^{-1}: 3,526 (OH), 3,098 (Ar–CH), 1,695 (C=O). ^{1}H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.64 (s, 1H, OH), 6.75–7.99 (m, 17H, Ar–CH), 8.05–8.31 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 190.5 (C=O), 165.3 (C-2), 162.9 (C-4), 158.1 (C‴-4), 157.4 (C-9), 151.6 (CH=CH), 146.2 (C″-1), 145.8 (C-7), 143.7 (C″-4), 142.5 (C-5), 141.9 (C′-4), 139.2 (C″-3), 138.7 (C′-3), 136.3 (C″-5), 133.4 (C′-5), 131.8 (C′-1), 128.2 (C‴-6), 126.5 (C′-6), 125.0 (C″-6), 124.4 (C"'-1), 123.2 (C‴-2), 119.6 (C′-2), 117.1 (C″-2), 114.8 (C-6), 113.9 (C-8), 109.0 (C-10), 107.6 (CH=CH), 103.7 (C‴-3), 101.3 (C‴-5). MS: *m/z* 444 [M^+]. Anal. Cald for $C_{29}H_{20}N_2O_3$: C, 78.36; H, 4.54; N, 6.30. Found: C, 78.65; H, 4.53; N, 6.28.

*3-(4-(3-(4-Nitrophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4e**)* Yield 79 %, m.p. 214–216 °C. IR (KBr) cm^{-1}: 3,053 (Ar–CH), 1,738 (C=O), 1,520 & 1,307 (NO_2). ^{1}H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.80–7.95 (m, 17H, Ar–CH), 8.07–8.33 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 194.1 (C=O), 174.8 (C-2), 165.2 (C-4), 163.4 (C-9), 152.0 (C‴-4), 150.9 (CH=CH), 147.3 (C‴-1), 146.5 (C″-1), 144.6 (C-7), 143.2 (C″-4), 142.0 (C′-4), 141.7 (C″-3), 137.5 (C′-3), 130.9 (C″-5), 129.9 (C′-5), 126.1 (C-5), 124.5 (C′-1), 120.4 (C‴-6), 119.7 (C′-6), 115.8 (C″-6), 115.0 (C‴-2), 113.3 (C′-2), 112.6 (C″-2), 112.1 (C-6), 109.5 (C-8), 109.0 (CH=CH), 106.2 (C‴-3), 105.7 (C‴-5), 104.8 (C-10). MS: *m/z* 473 [M^+]. Anal. Cald for $C_{29}H_{19}N_3O_4$: C, 73.56; H, 4.04; N, 8.87. Found: C, 73.81; H, 4.03; N, 8.89.

*2-Phenyl-3-(4-(3-(4-(trifluoromethyl)phenyl)acryloyl)phenyl)quinazolin-4(3H)-one (**4f**)* Yield 81 %, m.p. 220–222 °C. IR (KBr) cm^{-1}: 2,989 (Ar–CH), 1,703 (C=O), 1,136 (C–F). ^{1}H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.56–7.84 (m, 17H, Ar–CH), 7.95–8.26 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 189.5 (C=O), 180.2 (C-2), 177.6 (C-4), 170.3 (C-9), 164.8 (CH=CH), 155.5 (C‴-1), 150.9 (C″-1), 148.2 (C-7), 147.7 (C″-4), 145.6 (C′-4), 144.0 (C‴-4), 143.1 (C″-3), 142.8 (C′-3), 137.1 (C″-5), 132.4 (C′-5), 130.0 (C-5), 128.3 (C′-1), 125.5 (C‴-6), 124.7 (C′-6), 122.9 (C″-6), 120.8 (C‴-3), 119.2 (C‴-5), 118.7 (C‴-2), 118.0 (C′-2), 115.2 (C″-2), 114.6 (C-6), 112.5 (CF_3), 111.2 (C-8), 110.9 (CH=CH), 107.3 (C-10). MS: *m/z* 496 [M^+]. Anal. Cald for $C_{30}H_{19}F_3N_2O_2$: C, 72.58; H, 3.86; N, 5.64. Found: C, 72.34; H, 3.87; N, 5.66.

*3-(4-(3-(4-Chlorophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4g**)* Yield 70 %, m.p. 192–195 °C. IR (KBr) cm^{-1}: 3,081 (Ar–CH), 1,727 (C=O), 743 (C–Cl). ^{1}H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.93–7.90 (m, 17H, Ar–CH), 7.98–8.24 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 196.1 (C=O), 170.7 (C-2), 164.8 (C-4), 160.5 (C-9), 158.2 (CH=CH), 155.4 (C″-1), 151.9 (C‴-4), 150.0 (C″-4), 149.3 (C-7), 146.7 (C‴-1),145.5 (C′-4), 144.2 (C″-3), 140.7 (C′-3), 135.1 (C‴-3), 134.8 (C″-5), 133.0 (C′-5), 130.3 (C‴-5), 128.5 (C-5), 125.6 (C′-1), 122.9 (C‴-6), 120.7 (C-6), 119.5 (C′-6), 118.2 (C″-6), 115.1 (C‴-2), 112.7 (C′-2), 111.8 (C″-2), 108.9 (C-8), 105.1 (CH=CH), 101.5 (C-10). MS: *m/z* 464 [M^{+2}]. Anal. Cald for $C_{29}H_{19}ClN_2O_2$: C, 75.24; H, 4.14; N, 6.05. Found: C, 75.51; H, 4.13; N, 6.03.

*3-(4-(3-(4-Fluorophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4h**)* Yield 74 %, m.p. 156–158 °C. IR (KBr) cm^{-1}: 3,056 (Ar–CH), 1,733 (C=O), 1,119 (C–F). ^{1}H NMR ($CDCl_3$, 500 MHz) δ ppm: 7.18–8.13 (m, 17H, Ar–CH), 8.22–8.50 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 205.9 (C=O), 174.2 (C-2), 170.5 (C‴-4), 163.7 (C-4), 154.3 (C-9), 153.0 (CH=CH), 150.8 (C″-1), 148.1 (C″-4), 147.5 (C-7), 142.6 (C‴-1),141.4 (C′-4), 140.0 (C″-3), 137.2 (C′-3), 135.1 (C″-5), 134.5 (C′-5), 130.8 (C-5), 128.1 (C′-1), 125.7 (C‴-6), 124.9 (C‴-2), 121.3 (C-6), 120.8 (C′-6), 115.5 (C″-6), 114.7 (C-8), 113.2 (C′-2), 110.4 (C″-2), 109.8 (CH=CH), 106.6 (C-10), 102.5 (C‴-3), 100.9 (C‴-5). MS: *m/z* 446 [M^+]. Anal. Cald for $C_{29}H_{19}FN_2O_2$: C, 78.01; H, 4.29; N, 6.27. Found: C, 78.29; H, 4.30; N, 6.25.

*3-(4-(3-(3-Methoxyphenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4i**)* Yield 78 %, m.p. 238–240 °C. IR (KBr) cm^{-1}: 3,117 (Ar–CH), 1,716 (C=O), 1,054 (C–O-C). ^{1}H NMR ($CDCl_3$, 500 MHz) δ ppm: 3.69 (s, 3H, OCH_3), 6.84–7.88 (m, 17H, Ar–CH), 7.93–8.19 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 191.7 (C=O), 169.2 (C-2), 165.6 (C-4), 159.5 (C‴-3), 155.1 (C-9), 152.9 (CH=CH), 146.5 (C″-1), 144.2 (C‴-1), 143.0 (C′-1), 138.6 (C-7), 135.4 (C″-4), 134.9 (C-5), 133.4 (C′-4), 130.1 (C″-3), 128.3 (C‴-5), 127.8 (C′-3), 125.5 (C″-5), 120.9 (C′-5), 119.4 (C′-6), 118.6 (C″-6), 116.3 (C′-2), 111.7 (C″-2), 110.0 (C-6), 108.2 (C-8), 106.9 (C-10), 103.5 (CH=CH), 101.2 (C‴-6), 99.6 (C‴-4), 95.1 (C‴-2), 63.4 (OCH_3). MS: *m/z* 458 [M^+]. Anal. Cald for $C_{30}H_{22}N_2O_3$: C, 78.59; H, 4.84; N, 6.11. Found: C, 78.34; H, 4.83; N, 6.13.

*3-(4-(3-(3-Aminophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4j**)* Yield 73 %, m.p. 227–230 °C. IR (KBr) cm^{-1}: 3,320 (NH), 3,093 (Ar–CH), 1,739 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.12 (s, 2H, NH_2), 6. 65–7.86 (m, 17H, Ar–CH), 7.97–8.25 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 187.3 (C=O), 179.7 (C-2), 172.4 (C-4), 168.5 (C-9), 162.9 (C‴-3), 160.1 (CH=CH), 145.0 (C″-1), 144.2 (C‴-1), 143.8 (C-7), 142.6 (C″-4), 139.1 (C-5), 135.7 (C′-4), 134.3 (C″-3), 133.9 (C′-3), 131.5 (C″-5), 126.1 (C‴-5), 125.7 (C′-5), 123.4 (C′-1), 122.0 (C′-6), 121.5 (C″-6), 120.2 (C′-2), 118.7 (C″-2), 115. 1 (C-6), 114.6 (C-8), 110.5 (C-10), 108.3 (CH=CH), 105.1 (C‴-6), 103.9 (C'"-4), 100.4 (C‴-2). MS: *m/z* 443 [M^+]. Anal. Cald for $C_{29}H_{21}N_3O_2$: C, 78.54; H, 4.77; N, 9.47. Found: C, 78.80; H, 4.78; N, 9.43.

*3-(4-(3-(3-Hydroxyphenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4k**)* Yield 76 %, m.p. 163–165 °C. IR (KBr) cm^{-1}: 3,502 (OH), 3,079 (Ar–CH), 1,735 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.88 (s, 1H, OH), 7.10–8. 17 (m, 17H, Ar–CH), 8.29–8.58 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 197.3 (C=O), 172.8 (C-2), 166.4 (C-4), 159.6 (C‴-3), 155.0 (C-9), 148.9 (CH=CH), 140.1 (C″-1), 138.5 (C‴-1), 135.2 (C-7), 134.6 (C″-4), 130.9 (C-5), 129.3 (C′-4), 128.7 (C″-3), 125.5 (C‴-5), 123.9 (C′-3), 122.1 (C″-5), 121.5 (C′-5), 118.2 (C′-1), 117. 5 (C‴-6), 116.8 (C′-6), 115.4 (C′-2), 116.0 (C″-2), 114.2 (C-6), 112.6 (C-8), 110.7 (C-10), 106.3 (CH=CH), 103.1 (C″-6), 99.0 (C‴-4), 97.5 (C‴-2). MS: *m/z* 444 [M^+]. Anal. Cald for $C_{29}H_{20}N_2O_3$: C, 78.36; H, 4.54; N, 6.30. Found: C, 78.61; H, 4.55; N, 6.28.

*3-(4-(3-(3-Nitrophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4l**)* Yield 72 %, m.p. 245–247 °C. IR (KBr) cm^{-1}: 2,995 (Ar–CH), 1,708 (C=O), 1,536 & 1,322 (NO_2). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 7.02–8.01 (m, 17H, Ar–CH), 8.15–8.39 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 207.6 (C=O), 170.9 (C-2), 168.4 (C-4), 166.1 (C-9), 159.8 (C‴-3), 156.2 (CH=CH), 143.7 (C″-1), 142.0 (C‴-1), 141.5 (C-7), 139.4 (C″-4), 136.8 (C‴-6), 135.3 (C′-4), 134.9 (C″-3), 132.5 (C′-3), 131.0 (C″-5), 128.2 (C‴-5), 127.8 (C′-5), 127.0 (C-5), 125.9 (C′-1), 122.7 (C′-6), 121.4 (C″-6), 118.5 (C′-2), 117.9 (C″-2), 115.0 (C-6), 113.6 (C-8), 111.9 (CH=CH), 109.5 (C‴-2), 107.2 (C-10), 101.3 (C‴-4). MS: *m/z* 473 [M^+]. Anal. Cald for $C_{29}H_{19}N_3O_4$: C, 73.56; H, 4.04; N, 8.87. Found: C, 73.34; H, 4.05; N, 8.90.

*2-Phenyl-3-(4-(3-(3-(trifluoromethyl)phenyl)acryloyl)phenyl) quinazolin-4(3H)-one (**4m**)* Yield 75 %, m.p. 209–211 °C. IR (KBr) cm^{-1}: 3,059 (Ar–CH), 1,714 (C=O), 1,127 (C–F). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.76–7.82 (m, 17H, Ar–CH), 7.92–8.24 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 201.6 (C=O), 169.9 (C-2), 166.3 (C-4), 162.5 (C-9), 159.2 (CH=CH), 150.1 (C″-1), 148.7 (C‴-1), 147.0 (C-7), 145.3 (C‴-3), 144.8 (C″-4), 142.5 (C′-4), 140.7 (C″-3), 139.8 (C′-3), 138.2 (C″-5), 135.0 (C′-5), 133.5 (C-5), 130.9 (C‴-6), 127.6 (C‴-5), 125.1 (C′-1), 122.9 (C′-6), 121. 7 (C″-6), 120.0 (C‴-4), 117.4 (C‴-2), 116.5 (C′-2), 113.1 (C″-2), 110.2 (C-6), 108.7 (CF_3), 105.3 (C-8), 105.1 (CH=CH), 104.8 (C-10). MS: *m/z* 496 [M^+]. Anal. Cald for $C_{30}H_{19}F_3N_2O_2$: C, 72.58; H, 3.86; N, 5.64. Found: C, 72.30; H, 3.87; N, 5.62.

*3-(4-(3-(3-Chlorophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4n**)* Yield 79 %, m.p. 149–151 °C. IR (KBr) cm^{-1}: 3,072 (Ar–CH), 1,699 (C=O), 755 (C–Cl). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.98–8.11 (m, 17H, Ar–CH), 8.19–8.45 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 189.8 (C=O), 160.2 (C-2), 157.5 (C-4), 155.7 (C-9), 154.3 (CH=CH), 149.9 (C″-1), 147.5 (C‴-1), 144.0 (C‴-3), 143.1 (C″-4), 141.4 (C-7), 140.6 (C′-4), 137.2 (C‴-5), 136.5 (C″-3), 135.8 (C′-3), 133.4 (C″-5), 131.2 (C′-5), 129.0 (C-5), 125.1 (C‴-4), 124.6 (C′-1), 122.2 (C-6), 118.3 (C′-6), 115.6 (C″-6), 114.0 (C‴-2), 110.5 (C′-2), 106. 8 (C″-2), 102.3 (C‴-6), 99.7 (C-8), 98.4 (CH=CH), 95.6 (C-10). MS: *m/z* 464 [M^{+2}]. Anal. Cald for $C_{29}H_{19}ClN_2O_2$: C, 75.24; H, 4.14; N, 6.05. Found: C, 75.54; H, 4.15; N, 6.03.

*3-(4-(3-(3-Fluorophenyl)acryloyl)phenyl)-2-phenylquinazolin-4(3H)-one (**4o**)* Yield 80 %, m.p. 258–260 °C. IR (KBr) cm^{-1}: 3,122 (Ar–CH), 1,727 (C=O), 1,139 (C–F). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.50–7.78 (m, 17H, Ar–CH), 7.81–8.17 (m, 2H, CH=CH). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 194.5 (C=O), 178.9 (C-2), 174.2 (C‴-3), 169.4 (C-4), 165.8 (C-9), 160.1 (CH=CH), 148.3 (C″-1), 146.7 (C‴-1), 145.9 (C″-4), 143.0 (C-7), 141.2 (C‴-5), 140.6 (C′-4), 138.5 (C″-3), 137.7 (C′-3), 136.2 (C″-5), 133. 1 (C′-5), 131.4 (C-5), 126.8 (C′-1), 124.0 (C-6), 123.2 (C′-6), 122.9 (C″-6), 121.6 (C‴-6), 118.1 (C-8), 116.3 (C′-2), 112.9 (C″-2), 106.8 (CH=CH), 105.5 (C-10), 102.9 (C"'-4), 100.7 (C‴-2). MS: *m/z* 446 [M^+]. Anal. Cald for $C_{29}H_{19}FN_2O_2$: C, 78.01; H, 4.29; N, 6.27. Found: C, 78.25; H, 4.28; N, 6.29.

*General procedure for the synthesis of 2-phenyl-3-(4-(5-substitutedphenylisoxazol-3-yl)phenyl)quinazolin-4(3H)-one (**5a–5o**)* Title compound **5a–5o** was synthesized by adding hydroxylamine hydrochloride (0.69 g, 0.01 mol) in fraction with the well-stirred mixture of 2-phenyl-3-(4-(3-(substitutedphenyl)acryloyl)phenyl)quinazolin-4(3*H*)-one **4a–4o** (0.01 mol) in ethanol (25 ml). To this catalytic quantity of sodium acetate and glacial acetic acid was added. The reaction mixture was then refluxed overnight. Then the reaction mixture was cooled and poured in ice cold water. 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.

*2-Phenyl-3-(4-(5-phenylisoxazol-3-yl)phenyl)quinazolin-4(3H)-one (**5a**)* Yield 75 %, m.p. 165–167 °C. IR (KBr) cm^{-1}: 3,031 (Ar–CH), 1,712 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.25 (s, 1H, C<u>H</u> of isoxazole), 7.08–8.11 (m, 18H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 175.2 (C-5 of isoxazole), 172.6 (C-2), 170.9 (C-3 of isoxazole), 165.1 (C=O), 162.8 (C-9), 146.5 (C-7), 143.3 (C′-1), 141.0 (C‴-1), 140.7 (C″-4), 138.6 (C″-3), 137.2 (C‴-3), 135.4 (C‴-5), 134.9 (C′-3), 131.7 (C-5), 130.5 (C‴-4), 129.3 (C′-4), 127.6 (C″-1), 127.2 (C′-5), 126.8 (C″-5), 125.0 (C‴-2), 124.1 (C‴-6), 124.6 (C-6), 122.5 (C′-2), 120.9 (C″-2), 120.7 (C-8), 118.1 (C′-6), 117.4 (C″-6), 116.1 (C-10), 109.7 (C-4 of isoxazole). MS: *m/z* 441 [M^+]. Anal. Cald for $C_{29}H_{19}N_3O_2$: C, 78.90; H, 4.34; N, 9.52. Found: C, 78.62; H, 4.35; N, 9.55.

*3-(4-(5-(4-Methoxyphenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5b**)* Yield 72 %, m.p. 192–194 °C. IR (KBr) cm^{-1}: 3,018 (Ar–CH), 1,730 (C=O), 1,059 (C–O-C). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 3.16 (s, 3H, OC<u>H</u>$_3$), 6.02 (s, 1H, C<u>H</u> of isoxazole), 6.85–7.97 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 185.3 (C-5 of isoxazole), 182.9 (C-2), 179.4 (C-3 of isoxazole), 176.9 (C=O), 173.6 (C‴-4), 166.2 (C-9), 151.7 (C-7), 148.1 (C′-1), 147.4 (C″-4), 144.9 (C′-3), 144.2 (C″-3), 142.5 (C-5), 141.8 (C″-1), 140.1 (C′-4), 138.3 (C‴-2), 137.7 (C‴-6), 136.9 (C′-5), 133.1 (C″-5), 130.6 (C-6), 129.8 (C′-2), 129.0 (C″-2), 128.2 (C‴-1), 126.5 (C-8), 124.8 (C′-6), 123.4 (C″-6), 123.0 (C-10), 120.3 (C‴-3), 117.5 (C‴-5), 104.9 (C-4 of isoxazole), 68.4 (OCH_3). MS: *m/z* 471 [M^+]. Anal. Cald for $C_{30}H_{21}N_3O_3$: C, 76.42; H, 4.49; N, 8.91. Found: C, 76.69; H, 4.47; N, 8.88.

*3-(4-(5-(4-Aminophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5c**)* Yield 81 %, m.p. 236–238 °C. IR (KBr) cm^{-1}: 3,337 (NH), 3,134 (Ar–CH), 1,725 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.50 (s, 2H, N<u>H</u>$_2$), 6.19 (s, 1H, C<u>H</u> of isoxazole), 7.12–8.09 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 183.5 (C-5 of isoxazole), 180.3 (C-2), 176.8 (C-3 of isoxazole), 173.2 (C=O), 171.0 (C-9), 164.7 (C‴-4), 153.1 (C-7), 150.4 (C′-1), 148.6 (C″-4), 147.9 (C′-3), 144.2 (C″-3), 143.9 (C-5), 143.5 (C″-1), 139.4 (C′-4), 138.1 (C‴-2), 137.6 (C‴-6), 137.0 (C′-5), 134.7 (C″-5), 134.5 (C-6), 132.9 (C′-2), 132.0 (C″-2), 129.2 (C-8), 126.8 (C′-6), 126.4 (C″-6), 124.3 (C-10), 123.8 (C‴-1), 122.6 (C‴-3), 119.1 (C‴-5), 104.9 (C-4 of isoxazole). MS: *m/z* 456 [M^+]. Anal. Cald for $C_{29}H_{20}N_4O_2$: C, 76.30; H, 4.42; N, 12.27. Found: C, 76.59; H, 4.41; N, 12.22.

*3-(4-(5-(4-Hydroxyphenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5d**)* Yield 76 %, m.p. 150–153 °C. IR (KBr) cm^{-1}: 3,510 (OH), 3,015 (Ar–CH), 1,708 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.89 (s, 1H, O<u>H</u>), 6.33 (s, 1H, C<u>H</u> of isoxazole), 7.00–8.04 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 193.3 (C-5 of isoxazole), 189.6 (C-2), 187.1 (C-3 of isoxazole), 184.7 (C=O), 180.4 (C‴-4), 176.0 (C-9), 158.9 (C-7), 157.2 (C′-1), 155.9 (C″-4), 155.7 (C′-3), 152.8 (C″-3), 151.5 (C‴-2), 151.2 (C‴-6), 148.9 (C-5), 148.1 (C″-1), 147.5 (C′-4), 146.2 (C′-5), 143.6 (C″-5), 143.0 (C-6), 141.4 (C′-2), 140.6 (C″-2), 140.3 (C‴-1), 137.4 (C-8), 135.1 (C′-6), 134.8 (C″-6), 133.5 (C-10), 133.2 (C‴-3), 129.1 (C‴-5), 102.3 (C-4 of isoxazole). MS: *m/z* 457 [M^+]. Anal. Cald for $C_{29}H_{19}N_3O_3$: C, 76.14; H, 4.19; N, 9.19. Found: C, 75.85; H, 4.20; N, 9.21.

*3-(4-(5-(4-Nitrophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5e**)* Yield 70 %, m.p. 174–176 °C. IR (KBr) cm^{-1}: 3,107 (Ar–CH), 1,741 (C=O), 1,554 & 1,317 (NO_2). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.05 (s, 1H, C<u>H</u> of isoxazole), 6.71–7.82 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 180.1 (C-5 of isoxazole), 177.5 (C-2), 175.0 (C-3 of isoxazole), 169.7 (C=O), 166.1 (C-9), 161.3 (C‴-4), 149.1 (C‴-1), 147.6 (C-7), 146.5 (C′-1), 145.1 (C″-4), 143.7 (C′-3), 140.8 (C″-3), 140.4 (C-5), 138.2 (C″-1), 137.9 (C′-4), 134.3 (C‴-2), 133.7 (C‴-6), 132.2 (C′-5), 131.7 (C″-5), 131.4 (C-6), 130.6 (C′-2), 127.9 (C″-2), 126.4 (C-8), 125.8 (C′-6), 124.5 (C″-6), 124.0 (C‴-3), 120.3 (C‴-5), 117.8 (C-10), 109.2 (C-4 of isoxazole). MS: *m/z* 486 [M^+]. Anal. Cald for $C_{29}H_{18}N_4O_4$: C, 71.60; H, 3.73; N, 11.52. Found: C, 71.33; H, 3.72; N, 11.56.

*2-Phenyl-3-(4-(5-(4-(trifluoromethyl)phenyl)isoxazol-3-yl)phenyl)quinazolin-4(3H)-one (**5f**)* Yield 78 %, m.p. 263–265 °C. IR (KBr) cm^{-1}: 3,052 (Ar–CH), 1,689 (C=O), 1,126 (C–F). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.20 (s, 1H, C<u>H</u> of isoxazole), 7.15–8.26 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 187.5 (C-5 of isoxazole), 184.9 (C-2), 179.4 (C-3 of isoxazole), 177.2 (C=O), 172.7 (C-9), 158.0 (C‴-1), 157.6 (C-7), 154.1 (C′-1), 150.8 (C‴-4), 148.4 (C″-4), 148.0 (C′-3), 147.6 (C″-3), 144.0 (C-5), 143.8 (C″-1), 142.3 (C′-4), 140.6 (C‴-2), 140.2 (C‴-6), 138.9 (C′-5), 137.3 (C″-5), 135.6 (C-6), 134.8 (C′-2), 134.5 (C″-2), 131.7 (C‴-3), 131.2 (C‴-5), 130.9 (CF_3), 128.5 (C-8), 127.3 (C′-6), 126.1 (C″-6), 122.4 (C-10), 107.6 (C-4 of isoxazole). MS: *m/z* 509 [M^+]. Anal. Cald for $C_{30}H_{18}F_3N_3O_2$: C, 70.72; H, 3.56; N, 8.25. Found: C, 70.99; H, 3.55; N, 8.22.

*3-(4-(5-(4-Chlorophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5g**)* Yield 85 %, m.p. 215–217 °C. IR (KBr) cm^{-1}: 2,996 (Ar–CH), 1,723 (C=O), 781 (C–Cl). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.38 (s, 1H, C<u>H</u> of isoxazole), 6.93–7.98 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 186.4 (C-5 of isoxazole), 183.8 (C-2), 178.3 (C-3 of isoxazole), 174.0 (C=O), 170.2 (C-9), 157.6 (C‴-4), 156.4 (C-7), 154.1 (C′-1), 151.9 (C″-4), 150.3 (C‴-3), 149.5 (C‴-5), 148.9 (C′-3), 148.1 (C″-3), 147.6 (C‴-2), 147.0 (C‴-6), 145.3 (C-5), 143.8 (C″-1), 143.5 (C′-4), 142.9 (C‴-1), 140.7 (C′-5), 140.2 (C″-5), 139.1 (C-6), 136.9 (C′-2), 136.7 (C″-2), 135.2 (C-8), 132.6 (C′-6), 132.2 (C″-6), 128.5 (C-10), 109.1 (C-4 of isoxazole). MS: *m/z* 477 [M^{+2}]. Anal. Cald for $C_{29}H_{18}ClN_3O_2$: C, 73.19; H, 3.81; N, 8.83. Found: C, 73.45; H, 3.80; N, 8.80.

*3-(4-(5-(4-Fluorophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5h**)* Yield 70 %, m.p. 181–183 °C. IR (KBr) cm^{-1}: 3,027 (Ar–CH), 1,738 (C=O), 1,115 (C–F). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.17 (s, 1H, C<u>H</u> of isoxazole), 6.69–7.86 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 191.2 (C-5 of isoxazole), 187.6 (C-2), 183.0 (C‴-4), 180.5 (C-3 of isoxazole), 174.8 (C=O), 168.1 (C-9), 152.4 (C-7), 150.7 (C′-1), 148.2 (C″-4), 146.9 (C"'-2), 146.3 (C‴-6), 144.7 (C′-3), 144.0 (C″-3), 143.1 (C-5), 142.5 (C″-1), 141.2 (C′-4), 139.8 (C′-5), 139.1 (C″-5), 138.6 (C-6), 137.9 (C′-2), 137.4 (C″-2), 136.9 (C‴-1), 135.3 (C-8), 133.7 (C′-6), 133.2 (C″-6), 131.8 (C-10), 130.5 (C‴-3), 128.6 (C‴-5), 112.3 (C-4 of isoxazole). MS: *m/z* 459 [M^+]. Anal. Cald for $C_{29}H_{18}FN_3O_2$: C, 75.81; H, 3.95; N, 9.15. Found: C, 76.07; H, 3.94; N, 9.11.

*3-(4-(5-(3-Methoxyphenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5i**)* Yield 83 %, m.p. 220–222 °C. IR (KBr) cm^{-1}: 3,002 (Ar–CH), 1,745 (C=O), 1,078 (C–O-C). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 3.07 (s, 3H, OC<u>H</u>$_3$), 6.03 (s, 1H, C<u>H</u> of isoxazole), 7.19–8.05 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 179.8 (C-5 of isoxazole), 177.1 (C-2), 174.2 (C-3 of isoxazole), 170.5 (C‴-3), 167.9 (C=O), 164.3 (C-9), 155.6 (C-7), 153.0 (C′-1), 152.1 (C‴-1), 151.5 (C‴-5), 149.4 (C″-4), 148.7 (C′-3), 145.3 (C″-3), 144.6 (C-5), 142.0 (C″-1), 141.2 (C′-4), 140.9 (C′-5), 137.5 (C″-5), 135.7 (C-6), 134.1 (C′-2), 132.4 (C″-2), 130.6 (C-8), 129.2 (C′-6), 127.5 (C″-6), 125.1 (C-10), 124.6 (C‴-6), 123.8 (C‴-4), 120.3 (C‴-2), 107.5 (C-4 of isoxazole), 65.9 (OCH_3). MS: *m/z* 471 [M^+]. Anal. Cald for $C_{30}H_{21}N_3O_3$: C, 76.42; H, 4.49; N, 8.91. Found: C, 76.71; H, 4.50; N, 8.89.

*3-(4-(5-(3-Aminophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5j**)* Yield 75 %, m.p. 189–191 °C. IR (KBr) cm^{-1}: 3,361 (NH), 3,129 (Ar–CH), 1,694 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.34 (s, 2H, N<u>H</u>$_2$), 6.26 (s, 1H, C<u>H</u> of isoxazole), 6.84–7.97 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 190.0 (C-5 of isoxazole), 186.7 (C-2), 181.9 (C-3 of isoxazole), 178.4 (C=O), 174.6 (C-9), 170.5 (C‴-3), 159.2 (C-7), 157.8 (C′-1), 156.3 (C‴-1), 152.1 (C″-4), 151.4 (C‴-5), 149.2 (C′-3), 148.5 (C″-3), 145.9 (C-5), 144.0 (C″-1), 142.6 (C′-4), 141.7 (C′-5), 140.3 (C″-5), 139.8 (C-6), 135.2 (C′-2), 134.5 (C″-2), 133.1 (C-8), 132.7 (C′-6), 131.2 (C″-6), 129.8 (C-10), 126.3 (C‴-6), 125.0 (C‴-4), 124.9 (C‴-2), 107.6 (C-4 of isoxazole). MS: *m/z* 456 [M^+]. Anal. Cald for $C_{29}H_{20}N_4O_2$: C, 76.30; H, 4.42; N, 12.27. Found: C, 76.55; H, 4.41; N, 12.24.

*3-(4-(5-(3-Hydroxyphenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5k**)* Yield 78 %, m.p. 232–235 °C. IR (KBr) cm^{-1}: 3,536 (OH), 3,033 (Ar–CH), 1,726 (C=O). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 5.95 (s, 1H, O<u>H</u>), 6.31 (s, 1H, C<u>H</u> of isoxazole), 7.22–8.31 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 183.5 (C-5 of isoxazole), 182.9 (C-2), 180.2 (C-3 of isoxazole), 177.4 (C=O), 175.1 (C‴-3), 171.8 (C-9), 155.3 (C-7), 154.7 (C′-1), 152.0 (C‴-1), 150.5 (C‴-5), 149.2 (C″-4), 146.9 (C′-3), 145.6 (C″-3), 143.1 (C-5), 141.7 (C″-1), 140.3 (C′-4), 138.6 (C′-5), 136.2 (C″-5), 134.5 (C-6), 133.0 (C′-2), 132.8 (C″-2), 132.1 (C-8), 130.9 (C′-6), 126.4 (C″-6), 125.9 (C-10), 122.0 (C‴-6), 121.7 (C‴-4), 117.5 (C‴-2), 104.8 (C-4 of isoxazole). MS: *m/z* 457 [M^+]. Anal. Cald for $C_{29}H_{19}N_3O_3$: C, 76.14; H, 4.19; N, 9.19. Found: C, 76.43; H, 4.18; N, 9.16.

*3-(4-(5-(3-Nitrophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5l**)* Yield 77 %, m.p. 248–250 °C. IR (KBr) cm^{-1}: 2,985 (Ar–CH), 1,701 (CvO), 1,515 & 1,329 (NO_2). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.08 (s, 1H, C<u>H</u> of isoxazole), 7.05–8.19 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 174.6 (C-5 of isoxazole), 173.9 (C-2), 171.1 (C-3 of isoxazole), 167.5 (C = O), 164.8 (C-9), 160.2 (C‴-3), 150.4 (C‴-6), 146.7 (C-7), 145.3 (C′-1), 143.9 (C‴-1), 141.0 (C″-4), 139.4 (C‴-5), 138.2 (C′-3), 137.9 (C″-3), 136.5 (C-5), 136.1 (C″-1), 134.8 (C′-4), 133.4 (C′-5), 132.6 (C″-5), 131.7 (C-6), 129.0 (C′-2), 125.3 (C″-2), 124.8 (C-8), 123.2 (C′-6), 120.9 (C″-6), 119.5 (C‴-2), 115.8 (C‴-4), 114.0 (C-10), 100.4 (C-4 of isoxazole). MS: *m/z* 486 [M^+]. Anal. Cald for $C_{29}H_{18}N_4O_4$: C, 71.60; H, 3.73; N, 11.52. Found: C, 71.39; H, 3.74; N, 11.55.

*2-Phenyl-3-(4-(5-(3-(trifluoromethyl)phenyl)isoxazol-3-yl)phenyl)quinazolin-4(3H)-one (**5m**)* Yield 74 %, m.p. 157–159 °C. IR (KBr) cm^{-1}: 3,048 (Ar–CH), 1,740 (C=O), 1,122 (C–F). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.12 (s, 1H, C<u>H</u> of isoxazole), 6.94–8.08 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 190.7 (C-5 of isoxazole), 186.2 (C-2), 185.9 (C-3 of isoxazole), 181.6 (C=O), 177.5 (C-9), 152.0 (C-7), 151.3 (C′-1), 148.8 (C‴-3), 147.1 (C‴-6), 146.4 (C‴-1), 143.0 (C″-4), 141.5 (C‴-5), 140.9 (C′-3),

137.2 (C″-3), 135.8 (C-5), 135.1 (C″-1), 133.6 (C′-4), 132.0 (C′-5), 131.2 (C″-5), 130.5 (C-6), 128.7 (C‴-2), 127.4 (C′-2), 126.3 (C″-2), 125.9 (C‴-4), 125.5 (CF_3), 122.6 (C-8), 120.1 (C′-6), 119.3 (C″-6), 118.2 (C-10), 101.8 (C-4 of isoxazole). MS: *m/z* 509 [M^+]. Anal. Cald for $C_{30}H_{18}F_3N_3O_2$: C, 70.72; H, 3.56; N, 8.25. Found: C, 70.93; H, 3.55; N, 8.27.

*3-(4-(5-(3-Chlorophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5n**)* Yield 80 %, m.p. 250–253 °C. IR (KBr) cm^{-1}: 3,023 (Ar–CH), 1,739 (C=O), 764 (C–Cl). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.45 (s, 1H, C<u>H</u> of isoxazole), 7.10–8.23 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 182.7 (C-5 of isoxazole), 180.5 (C-2), 179.2 (C-3 of isoxazole), 175.0 (C=O), 173.8 (C-9), 152.4 (C‴-3), 151.9 (C-7), 151.1 (C′-1), 150.6 (C‴-1), 147.0 (C‴-5), 145.2 (C″-4), 144.7 (C‴-4), 143.4 (C′-3), 142.5 (C″-3), 140.1 (C-5), 138.9 (C″-1), 137.3 (C′-4), 136.5 (C′-5), 135.0 (C″-5), 133.2 (C-6), 132.7 (C‴-2), 131.9 (C′-2), 130.6 (C″-2), 126.1 (C‴-6), 125.4 (C-8), 123.0 (C′-6), 122.4 (C″-6), 119.5 (C-10), 102.9 (C-4 of isoxazole). MS: *m/z* 477 [M^{+2}]. Anal. Cald for $C_{29}H_{18}ClN_3O_2$: C, 73.19; H, 3.81; N, 8.83. Found: C, 72.95; H, 3.82; N, 8.86.

*3-(4-(5-(3-Fluorophenyl)isoxazol-3-yl)phenyl)-2-phenylquinazolin-4(3H)-one (**5o**)* Yield 73 %, m.p. 267–269 °C. IR (KBr) cm^{-1}: 2,990 (Ar–CH), 1,717 (C=O), 1,132 (C–F). ^1H NMR ($CDCl_3$, 500 MHz) δ ppm: 6.24 (s, 1H, C<u>H</u> of isoxazole), 7.06–8.15 (m, 17H, Ar–C<u>H</u>). ^{13}C NMR ($CDCl_3$, 500 MHz) δ ppm: 186.7 (C-5 of isoxazole), 182.9 (C-2), 179.5 (C‴-3), 174.2 (C-3 of isoxazole), 170.6 (C=O), 166.8 (C-9), 144.3 (C-7), 143.1 (C′-1), 141.0 (C‴-1), 140.4 (C‴-5), 138.9 (C″-4), 138.0 (C′-3), 135.7 (C″-3), 134.2 (C-5), 133.8 (C″-1), 132.7 (C′-4), 132.1 (C′-5), 131.0 (C″-5), 129.5 (C-6), 128.4 (C′-2), 127.9 (C″-2), 125.3 (C‴-6), 124.9 (C-8), 122.6 (C′-6), 121.8 (C″-6), 120.5 (C-10), 119.0 (C‴-2), 117.7 (C‴-4), 100.2 (C-4 of isoxazole). MS: *m/z* 459 [M^+]. Anal. Cald for $C_{29}H_{18}FN_3O_2$: C, 75.81; H, 3.95; N, 9.15. Found: C, 76.10; H, 3.94; N, 9.13.

Biological activities

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). Animals were maintained under standard conditions in an animal house approved by committee for the purpose of control and supervision on experiments on animals. Institutional Animal Ethics Committee approved the experimental protocol. 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 1.

Anti-inflammatory activity

Anti-inflammatory activity was evaluated by carrageenan-induced paw oedema test in rats (Winter *et al*., 1962). Diclofenac sodium 10 and 20 mg/kg was administered as a standard drug for comparison. The test compounds were 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 oedema 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, and 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 2.

Ulcerogenicity

Ulceration in rats was induced as reported method (Goyal *et al*., 1985). Albino rats of Wistar strain weighing

Table 1 Percent analgesic activity of the synthesized compounds **5a–5o** (Tail-flick method)

Compound	Dose (mg/kg)	Percent analgesic activity			
		30 min	1 h	2 h	3 h
5a	10	29 ± 1.82**	35 ± 0.59*	41 ± 1.50*	27 ± 1.08**
	20	40 ± 1.41*	47 ± 2.05**	56 ± 1.23**	37 ± 0.67*
5b	10	23 ± 0.48**	26 ± 1.06*	30 ± 0.93*	20 ± 2.31^{NS}
	20	35 ± 2.15*	37 ± 1.82**	47 ± 1.29**	32 ± 1.56**
5c	10	22 ± 1.22**	26 ± 1.57*	29 ± 2.11**	18 ± 0.53**
	20	33 ± 1.79^{NS}	37 ± 0.94**	45 ± 1.33*	31 ± 2.16*
5d	10	25 ± 1.09**	29 ± 2.28*	34 ± 0.72***	23 ± 1.46**
	20	36 ± 2.26*	40 ± 0.77**	51 ± 1.71*	35 ± 0.94*
5e	10	33 ± 0.67**	40 ± 1.05***	47 ± 1.59*	34 ± 2.40*
	20	45 ± 0.41**	55 ± 1.56*	62 ± 2.09**	40 ± 1.25*
5f	10	36 ± 1.16*	43 ± 1.82**	53 ± 1.54***	41 ± 0.87**
	20	48 ± 0.83**	60 ± 2.35*	67 ± 0.69**	45 ± 1.40**
5g	10	34 ± 0.59*	42 ± 0.68**	49 ± 0.90**	37 ± 1.95*
	20	46 ± 1.38*	55 ± 1.80***	64 ± 1.15*	42 ± 0.62*
5h	10	35 ± 1.72*	42 ± 2.15*	51 ± 2.34**	38 ± 0.96**
	20	47 ± 1.56*	58 ± 1.31**	65 ± 0.88***	45 ± 2.21*
5i	10	10 ± 0.81*	14 ± 1.65*	20 ± 0.60*	10 ± 1.08^{NS}
	20	18 ± 1.64*	24 ± 2.11^{NS}	30 ± 1.29*	16 ± 0.63**
5j	10	09 ± 1.56*	14 ± 1.38*	18 ± 1.43*	08 ± 0.82*
	20	16 ± 2.37*	23 ± 0.91*	27 ± 2.10^{NS}	13 ± 1.05*
5k	10	12 ± 1.21*	17 ± 1.57*	22 ± 0.84**	11 ± 1.38*
	20	21 ± 0.55*	27 ± 2.51*	32 ± 1.27*	17 ± 1.60*
5l	10	13 ± 2.07*	19 ± 1.38*	24 ± 2.26*	13 ± 0.90*
	20	22 ± 0.92*	28 ± 1.59*	32 ± 0.75*	20 ± 2.33**
5m	10	18 ± 1.23*	24 ± 2.67*	28 ± 1.58*	15 ± 0.53*
	20	27 ± 0.74*	32 ± 1.49*	38 ± 2.14**	26 ± 1.19*
5n	10	15 ± 1.82**	20 ± 1.57*	25 ± 0.69*	12 ± 2.28*
	20	25 ± 1.20*	31 ± 0.85*	33 ± 1.67**	23 ± 1.41*
5o	10	17 ± 1.87*	23 ± 1.21*	25 ± 1.50*	16 ± 0.79*
	20	25 ± 2.19*	32 ± 1.73*	35 ± 0.74*	26 ± 2.08*
Control	–	3 ± 0.39	6 ± 0.52	5 ± 0.63	4 ± 0.43
Diclofenac	10	34 ± 1.53*	42 ± 2.17**	48 ± 0.60**	35 ± 1.34*
	20	45 ± 1.21*	56 ± 0.78**	64 ± 1.66***	43 ± 0.57*

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

NS non significant

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

150–200 g of either sex were divided into various groups each of six animals. Control group of animals was 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 three days. Diclofenac was also administered as a standard drug at 20 mg/kg once daily for three 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 4th 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 2.

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 was evaluated against four Gram-positive bacteria *Staphylococcus aureus*

ATCC 9,144, *Staphylococcus epidermidis* ATCC 155, *Micrococcus luteus* ATCC 4,698 and *Bacillus cereus* ATCC 11,778 and three Gram-negative bacteria *Escherichia coli* ATCC 25,922, *Pseudomonas aeruginosa* ATCC 2,853, and *Klebsiella pneumoniae* ATCC 11,298. The antifungal activities of the synthesized compounds were evaluated against two fungi *Aspergillus niger* ATCC 9,029 and *Aspergillus fumigatus* ATCC 46,645. 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 compounds was determined by agar streak dilution method (Hawkey and Lewis 1994). A stock solution of the synthesized compound 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 petridish to give a depth of 3–4 mm and allowed to solidify. Suspension of the microorganism was 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 3.

Results and Discussion

Chemistry

In this study, we synthesized fifteen novel isoxazole containing quinazolin-4(3*H*)-one derivatives **5a**–**5o**. Initially, anthranilic acid and benzoyl chloride were used as starting materials to produce 2-phenyl-4*H*-benzo-(1,3)-oxazin-4-one **2** by simple benzoylation followed by ring closure reaction. In the next step, 2-phenyl-4*H*-benzo-(1,3)-oxazin-4-one **2** was treated with *p*-amino acetophenone gave 3-(4-acetylphenyl)-2-phenylquinazolin-4(3*H*)-one **3**. In the pre final step, on stirring with various aromatic aldehydes in the presence of sodium hydroxide, compound **3** was converted to corresponding 2-phenyl-3-(4-(3-substitutedphenylacryloyl)phenyl)quinazolin-4(3*H*)-one **4a**–**4o** with the loss of water molecule. Finally, the various chalcones **4a**–**4o** were treated with hydroxylamine hydrochloride in the presence of catalytic amount of sodium acetate and acetic acid results in corresponding isoxazole-substituted 2-phenylquinazolin-4(3*H*)-one **5a**–**5o** derivatives according to the synthetic Scheme 1.

IR, ^{1}H NMR, mass spectra, and elemental analyses of the synthesized compounds were 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-phenyl-4*H*-benzo-(1,3)-oxazin-4-one **2** was confirmed by the presence of absorption peak at 1,751 and 1,038 cm^{-1} in IR due to the presence of C=O and C–O–C stretching, respectively. The formations of compound **3** were confirmed by the absence of absorption bands in IR around 1,050 cm^{-1} corresponds to C–O–C stretching. Appearance of additional singlet at δ 2.88 ppm for three protons in its ^{1}H NMR spectra which might be assigned to $COCH_3$ group also confirms the formation of **3**. Formation of compounds **4a**–**4o** was confirmed by appearance of multiplet (double doublet) for two protons around δ 7.81–8.58 ppm in its ^{1}H NMR spectra which might be assigned to CH=CH group of chalcones. ^{1}H NMR spectra of compound **5a**–**5o** showed a singlet peaks at δ 6.02–6.45 ppm correspond to CH of isoxazole confirms its formation. The title compounds **5a**–**5o** showed absorption bands at 2,985–3,134 and 1,689–1,745 cm^{-1}, which can be assignable to Ar–CH and C=O vibrations, respectively. The proton magnetic resonance spectrums of synthesized compounds were recorded in $CDCl_3$. The NMR spectra of compound **5a**–**5o** showed a singlet at δ 6.02–6.45 ppm correspond to one proton of CH of isoxazole and a multiplet at δ 6.69–8.31 ppm for seventeen protons of Ar–CH.

Biological activities

Analgesic activity

Tail-flick method was used to assess the analgesic activity of title compounds **5a**–**5o** in Wistar albino mice, and the obtained results are documented in Table 1. From the study, it was observed that most of the title compounds exhibited significant activity and graded dose response. The title compounds displayed 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. Compounds possessing substituent at para position of phenyl ring attached at C-5 of isoxazole **5b**–**5h** exhibited better activity than the corresponding substituent at meta position **5i**–**5o**. At the same

Table 2 Percent anti-inflammatory activity (Carrageenan-induced paw oedema test in rats) and ulcer index of the synthesized compounds **5a–5o**

Compound	Dose (mg/kg)	Percent protection				Ulcer index
		30 min	1 h	2 h	3 h	
5a	10	21 ± 1.92**	30 ± 0.98*	34 ± 2.56**	27 ± 1.13*	0.78 ± 0.49
	20	35 ± 0.68*	44 ± 2.71**	57 ± 1.35*	38 ± 1.48*	
5b	10	16 ± 2.19*	23 ± 1.02*	28 ± 0.47*	20 ± 1.21*	0.83 ± 0.62
	20	29 ± 1.20*	41 ± 1.85*	51 ± 2.42*	31 ± 0.97**	
5c	10	14 ± 1.76*	21 ± 2.93*	27 ± 0.78*	19 ± 1.04*	0.86 ± 0.43
	20	28 ± 2.64**	39 ± 0.59*	49 ± 1.14*	28 ± 1.36*	
5d	10	17 ± 0.43*	25 ± 1.26*	31 ± 1.52**	22 ± 0.40*	0.81 ± 0.70
	20	30 ± 1.41*	41 ± 1.04*	53 ± 2.49**	32 ± 0.86*	
5e	10	28 ± 2.19**	37 ± 0.65**	40 ± 1.91***	32 ± 1.28*	0.74 ± 0.48
	20	42 ± 1.65*	50 ± 1.34*	60 ± 0.59**	43 ± 0.78**	
5f	10	33 ± 0.43*	39 ± 1.47***	46 ± 0.73**	35 ± 1.95*	0.68 ± 0.52
	20	45 ± 0.87**	54 ± 0.83*	63 ± 1.75***	48 ± 0.61*	
5g	10	29 ± 1.16*	36 ± 1.25**	42 ± 0.60*	34 ± 0.84*	0.71 ± 0.44
	20	41 ± 2.84*	51 ± 1.96*	59 ± 0.89***	44 ± 1.07**	
5h	10	31 ± 1.32**	38 ± 0.47*	43 ± 1.09***	35 ± 1.46*	0.70 ± 0.55
	20	43 ± 0.56*	53 ± 1.73*	62 ± 0.48**	45 ± 1.84*	
5i	10	09 ± 0.91*	12 ± 2.25*	15 ± 0.70*	10 ± 1.39*	1.05 ± 0.81
	20	15 ± 1.55*	19 ± 1.60^{NS}	24 ± 2.52*	15 ± 0.46*	
5j	10	06 ± 0.43*	10 ± 1.37*	14 ± 0.96*	09 ± 2.18*	1.16 ± 0.89
	20	12 ± 0.80*	16 ± 2.76*	21 ± 1.43*	10 ± 1.66^{NS}	
5k	10	09 ± 2.87*	13 ± 0.54*	15 ± 1.85*	11 ± 1.91*	0.99 ± 0.67
	20	17 ± 1.01^{NS}	20 ± 1.51*	24 ± 0.68*	17 ± 2.37*	
5l	10	10 ± 2.36*	13 ± 1.54*	17 ± 0.88*	11 ± 0.93*	0.97 ± 0.58
	20	17 ± 1.52*	21 ± 2.28*	26 ± 0.50*	20 ± 1.54**	
5m	10	13 ± 1.39*	16 ± 0.59*	21 ± 2.69*	17 ± 1.36**	0.90 ± 0.63
	20	21 ± 0.79*	26 ± 1.35*	32 ± 1.62*	25 ± 2.28**	
5n	10	11 ± 2.46*	15 ± 1.70*	18 ± 0.92*	13 ± 0.53*	0.93 ± 0.51
	20	19 ± 1.82*	22 ± 0.79*	29 ± 2.15**	21 ± 1.67*	
5o	10	13 ± 0.91*	16 ± 1.15*	20 ± 1.21*	14 ± 2.30*	0.92 ± 0.49
	20	20 ± 1.03**	24 ± 1.98*	32 ± 0.56*	21 ± 1.34*	
Control		4.1 ± 0.62	6.4 ± 0.95	4.9 ± 0.43	3.2 ± 0.58	0.13 ± 0.07
Diclofenac	10	29 ± 0.46*	37 ± 1.19***	42 ± 0.71**	33 ± 1.53*	1.61 ± 0.53
	20	42 ± 1.63**	51 ± 1.36**	60 ± 0.81**	44 ± 1.55*	
Aspirin	200	–	–	–	–	1.79 ± 0.65

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

NS non significant

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

position, compounds possessing electron-accepting group (**5e–5h** and **5l–5o**) displayed superior activity than the compounds containing electron-releasing groups (**5b–5d** and **5i–5k**). Compound having un substituted phenyl ring at 5th position of isoxazole **5a** showed moderate analgesic activity. Trifluoro methyl derivative exhibited good activity among the various electron-withdrawing groups tested, followed by fluoro, chloro, and nitro derivatives.

Anti-inflammatory activity

Carrageenan-induced paw edema test in rats was performed to evaluate the anti-inflammatory activity. The anti-inflammatory activity results are presented in Table 2. 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

Table 3 Minimum inhibitory concentration (MIC in μg/ml) of synthesized compounds **5a–5o**

Compounds	*S. aureus*	*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	62.5	31.25
5b	62.5	62.5	125	62.5	125	31.25	62.5	125	62.5
5c	125	62.5	125	125	125	62.5	62.5	125	125
5d	62.5	31.25	62.5	62.5	62.5	31.25	31.25	62.5	62.5
5e	31.25	15.62	7.81	31.25	31.25	15.62	7.81	31.25	7.81
5f	15.62	7.81	15.62	7.81	7.81	15.62	7.81	7.81	7.81
5g	15.62	15.62	31.25	15.62	31.25	15.62	3.9	31.25	15.62
5h	31.25	15.62	7.81	15.62	15.62	7.81	15.62	15.62	15.62
5i	125	62.5	125	62.5	125	62.5	62.5	125	62.5
5j	125	62.5	125	125	125	62.5	125	125	125
5k	62.5	62.5	62.5	62.5	62.5	31.25	31.25	62.5	125
5l	31.25	15.62	7.81	31.25	31.25	15.62	7.81	31.25	15.62
5m	15.62	7.81	31.25	7.81	15.62	15.62	7.81	15.62	7.81
5n	15.62	15.62	31.25	15.62	31.25	31.25	7.81	31.25	15.62
5o	31.25	15.62	7.81	15.62	31.25	7.81	15.62	15.62	15.62
Ciprofloxacin	15.62	7.81	7.81	7.81	15.62	7.81	3.9	–	–
Ketoconazole	–	–	–	–	–	–	–	15.62	7.81

maximum level at 2 h. Declining in activity was observed at 3 h. Compounds **5f** (p-CF_3 substituted derivative) and **5h** (p-F-substituted derivative) displayed a significant anti-inflammatory activity which is more than standard diclofenac. Compound **5g** (p-Cl-substituted derivative) and **5e** (p-NO_2-substituted derivative) protected rats from carrageenan-induced inflammation almost equally like standard diclofenac. All these compounds possessing electron-withdrawing group at para position of phenyl ring attached to isoxazole nucleus. The position of substituent's on phenyl ring attached at 5th position of isoxazole appeared to greatly influence the anti-inflammatory activity; compounds possessing substituent's at para position displayed better activity than corresponding substituent at meta position. Unsubstituted derivative **5a** exhibited moderate anti-inflammatory activity. Like analgesic activity, the compounds with electron-withdrawing substituents such as nitro, trifluoromethyl, chloro, and fluoro group exhibited more anti-inflammatory than corresponding compounds with electron-releasing substituents such as methoxy, amino, and hydroxyl group.

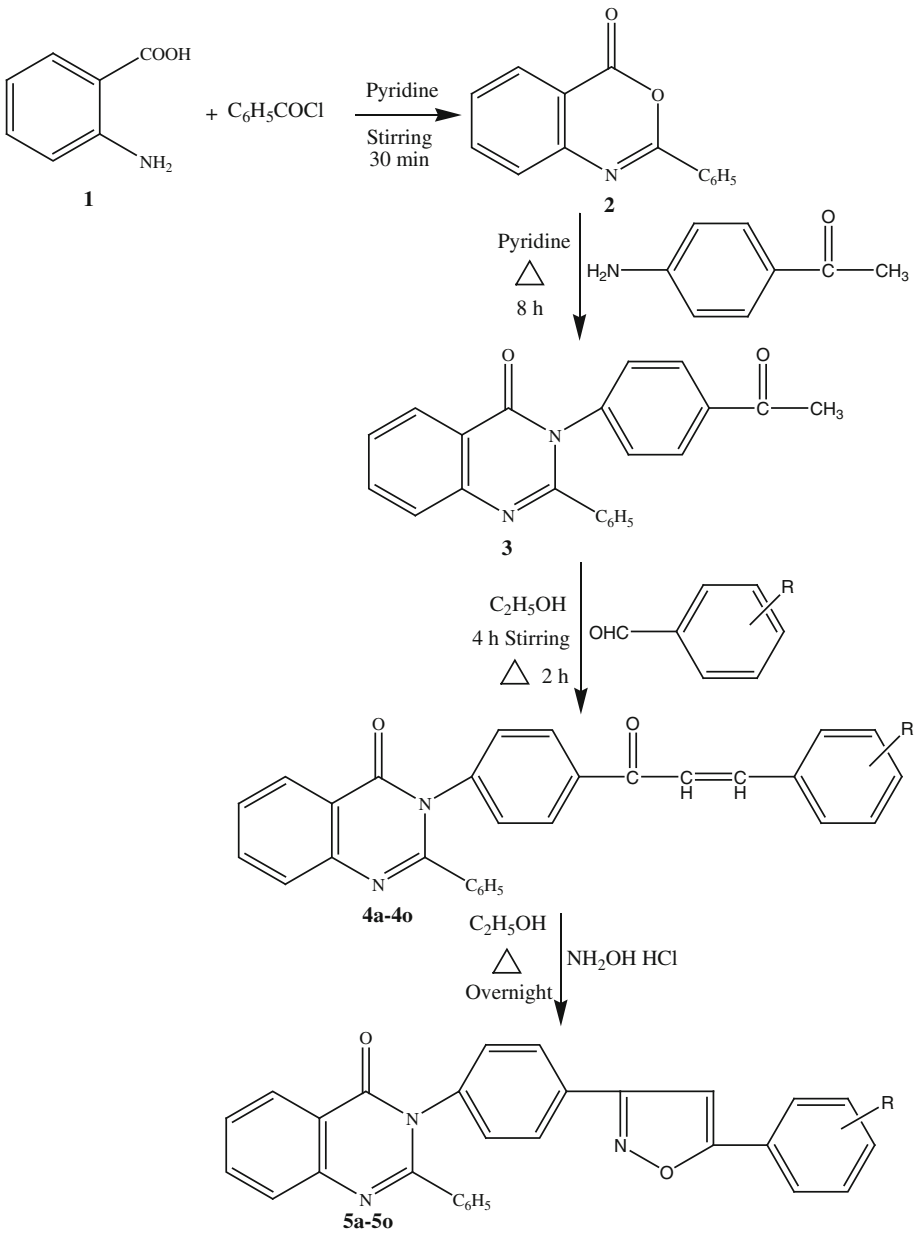
Ulcerogenicity

In addition, all the test compounds were examined for their ulcerogenicity, and the results are summarized in Table 2. The entire test compounds exhibited ulcer indexes less than those obtained by the standard diclofenac and aspirin. Results revealed that para-substituted analogs **5b–5h** displayed negligible or less ulcer index, whereas meta-substituted analogs **5i–5o** showed little increases in ulcer index. At the same position compounds bearing electron-donating groups exhibited higher ulcer index over compounds bearing electron-withdrawing groups; whereas un substituted one exhibited moderate ulcer index. The test compounds exhibited 42.24–72.05 % and 37.99–64.80 % of the ulcer index when compared to the reference drug diclofenac (1.61 ± 0.53) and aspirin (1.79 ± 0.65), respectively. Among the tested compounds, 2-phenyl-3-(4-(5-(4-trifluoromethylphenyl) isoxazol-3-yl)phenyl)quinazolin-4(3*H*)-one **5f** exhibited least ulcer index (0.68 ± 0.52) which is about 40 % of the ulcer index of reference standards. Out of entire tested compounds, 2-phenyl-3-(4-(5-(3-aminophenyl)isoxazol-3-yl)phenyl)quinazolin-4(3*H*)-one **5j** was found to possess highest ulcer index (1.16 ± 0.89) which is about 68 % of the ulcer index of diclofenac and aspirin.

Antimicrobial activity

Agar streak dilution method was used to analyze the in vitro antimicrobial activity of title compounds **5a–5o**. A comparison of antimicrobial activity of the synthesized compounds with that of standard drugs is effectively presented in Table 3. In order to control the sensitivity of the test organisms MICs of standard drugs (Ciprofloxacin and Ketoconazole) was determined in parallel experiments. The MIC values were determines as the lowest concentration that totally inhibited visible growth of the microorganisms. From the results, it was observed that compound **5f, 5g, 5m**

Scheme 1 Synthesis of title compounds (**5a–5o**)



R = H (**4a**, **5a**), 4-OCH_3 (**4b**, **5b**), 4-NH_2 (**4c**, **5c**), 4-OH (**4d**, **5d**), 4-NO_2 (**4e**, **5e**), 4-CF_3 (**4f**, **5f**), 4-Cl (**4g**, **5g**), 4-F (**4h**, **5h**), 3-OCH_3 (**4i**, **5i**), 3-NH_2 (**4j**, **5j**), 3-OH (**4k**, **5k**), 3-NO_2 (**4l**, **5l**), 3-CF_3 (**4m**, **5m**), 3-Cl (**4n**, **5n**), 3-F (**4o**, **5o**)

and **5n** (MIC: 15.62 μg/ml) displayed similar activity like Ciprofloxacin against *S. aureus;* whereas rest of series exhibited lesser activity (MIC: 31.25–125 μg/ml). Compared to Ciprofloxacin, compounds **5f** and **5m** exhibited equal activity (MIC: 7.81 μg/ml) against *S. epidermidis*, whereas rest of series exhibited lower activity (MIC: 15.62–62.5 μg/ml). Against *M. luteus* compounds **5e, 5h, 5l** and **5o** exhibited comparable activity (MIC: 7.81 μg/ml) as Ciprofloxacin, while others demonstrated lesser activity than standard. Compounds **5f** and **5m** showed the same activity (MIC: 7.81 μg/ml) as Ciprofloxacin, whereas rest of all compounds showed worse activities than standard against *B. cereus*. Compound **5f** exhibited better activity than Ciprofloxacin against *E. coli* (MIC: 7.81 μg/ml); whereas compounds **5h** and **5m** displayed equal activity (MIC: 15.62 μg/ml) and all other derivatives exhibited lower activity against *E. coli* (MIC: 31.25–125 μg/ml). Compounds **5h** and **5o** displayed the equivalent activity (MIC: 7.81 μg/ml) as standard against *P. aeruginosa*. Test compound **5 g** showed the same activity (MIC: 3.9 μg/ml) as Ciprofloxacin against *K. pneumoniae*. Against *A. niger* compound **5f** demonstrated superior activity and

compounds **5 h, 5 m** and **5o** showed similar activity (MIC: 15.62 μg/ml) as Ketoconazole; whereas remaining compounds showed inferior activity (MIC: 31.25–125 μg/ml). Compounds **5e, 5f** and **5m** 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 compound **5e–5h** and **5l–5o** might be due to the presence of electron-withdrawing substituent like nitro, trifluoromethyl, chloro, and fluoro groups. While other compounds, though they contain electron-donating substituents like methoxy, amino, and hydroxyl groups **5b–5d** and **5i–5k** exhibited less in vitro antimicrobial activity. Unsubstituted compound displayed moderate antimicrobial activity. Corresponding meta- and para-substituted derivatives exhibited almost equal activity, hence unlike analgesic and anti-inflammatory activity, the position of the substituent's influenced very less in the antimicrobial activity. 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-phenyl-3-(4-(5-(4-(trifluoromethyl)phenyl)isoxazol-3-yl)phenyl)quinazolin-4(3*H*)-one **5f** exhibited better activity against *E.coli* and *A. niger*; while it displayed equal activity as standard against *S. aureus*, *S. epidermidis*, *B. cereus*, and *A. fumigatus.*

Conclusion

In conclusion, a variety of novel quinazolin-4(3*H*)-one derivative were synthesized by introducing phenyl moiety at C-2 and 3,5-disubstituted isoxazole moiety at C-3 of quinazolin-4(3*H*)-one. All synthesized compounds were characterized by FT-IR, ^{1}H NMR, Mass spectroscopy, and elemental analysis. All title compounds were assessed for their analgesic, anti-inflammatory, ulcerogenicity, and in vitro antimicrobial activity. From the SAR studies, it was found that position of the substituent played major role in determining analgesic and anti-inflammatory activity than nature of substituent's; whereas nature of substituent's played important role in assessing antimicrobial activity than its position. Among several tested compounds, 2-phenyl-3-(4-(5-(4-(trifluoromethyl)phenyl)isoxazol-3-yl)phenyl)quinazolin-4(3*H*)-one **5f** showed better analgesic and anti-inflammatory activity which is more potent than reference standard Diclofenac. Interestingly this derivative possessed about only 40 % of the ulcer index of reference standards. In addition, compound **5f** also showed some excellent antimicrobial activity against some pathogenic strains of micro organism. 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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