Journal of Enzyme Inhibition and Medicinal Chemistry, 2013; 28(1): 1–10

ISSN 1475-6366 print/ISSN 1475-6374 online
DOI: 10.3109/14756366.2011.611943

informa
healthcare

RESEARCH ARTICLE

Synthesis and biological evaluation of novel benzothiazole clubbed fluoroquinolone derivatives

Prabodh Chander Sharma[1], Ravinder Kumar[1], Monika Chaudhary[1], Archana Sharma[1], and Harish Rajak[2]

[1]*Institute of Pharmaceutical Sciences, Kurukshetra University, Kurukshetra, India and* [2]*Institute of Pharmaceutical Sciences, Guru Ghasidas University, Bilaspur, India*

Abstract

In the present investigation, synthesis and anti-bacterial, analgesic and anthelmintic evaluation of a novel series of fluoroquinolone derivatives clubbed with benzothiazole moeity has been described. The synthesized compounds were characterised by spectral analysis (IR and ^{1}H NMR). Preliminary results indicated that the most of the synthesized compounds demonstrated good activities against gram negative and gram positive bacterial strains. Compounds **5a, 5b, 5f** and **5k** demonstrated potent anti-bacterial activities. Compound **5a** exhibited most potent anti-bacterial activity with MIC values of 04, 03, 08 and 15 µg/ mL against *B. subtilis, S. aureus, E. coli* and *P. aeruginosa*. Analogs **5a, 5c, 5g** and **5h** showed promising anthelmintic activity against *Eisemia foetida* in a low concentration as compared to standard drug piperazine citrate with mean paralysis time ranging 22.60 ± 2.46 to 31.60 ± 3.07 min. All synthesized compounds depicted good *in vivo* analgesic activity with compound **5a** exhibiting the most potent activity of 55.19% inhibition of writhing in comparison to the standard drug.

Keywords: Fluoroquinolones, benzothiazoles, anti-bacterial, analgesic, anthelmintic

Introduction

Fluoroquinolone anti-bacterial agents are among the most attractive drugs in the field of anti-infective chemotherapy[1–3]. The relevant breakthrough within this class of agents occurred almost 20 years after the original discovery when the addition of the fluorine molecule at a position C-6 of the pharmacophore created the 'fluoroquinolones'[4]. These antibiotics exert their anti-microbial activity by binding to two type II bacterial topoisomerase enzymes, DNA gyrase (subunits encoded by gyrA and gyrB) and topoisomerase IV (subunits encoded by gyrA and gyr B for *Staphylococcus aureus*). This binding induces permanent double stranded DNA breaks, and results in cell death[5,6]. Ciprofloxacin is among one of the most successful drugs of fluoroquinolone category[7]. The market is heavily dominated by ciprofloxacin and levofloxacin, which together command 65% ($3.3 billion) of global sales[8]. Due to excellent pharmacokinetic and pharmacological profile, fluoroquinolones have been the centre of attraction of the scientists for long period. Several new derivatives have been synthesized and evaluated for anti-bacterial activity against many bacterial strains such as *P. aeruginosa, K. pneumoniae, S. pneumoniae, S. aureus, S. epidermis* etc.[9–12] Fluoroquinolones analogs with anti-tubercular[13–14], anti-fungal[15], anti-viral[16] activities are well known. Many research endeavours are being undertaken to elaborate the possible role of fluoroquinolones in carcinogenesis and mutagenesis[17–18].

Heterocycles bearing nitrogen, sulphur and thiazole moieties constitute the core for a number of biological interesting compounds[19–20]. The 2-aminobenzothiazoles scaffold is one of the privileged structures in medicinal chemistry. Indeed various examples featuring this particular scaffold have been prepared, many exhibiting remarkable biological activities. It has demonstrated a myriad spectrum of biological activities such as anti-microbial[21], anti-inflammatory, analgesic and anti-tumour activity[22].

Address for Correspondence: Dr. Prabodh Chander Sharma, Assistant Professor, Institute of Pharmaceutical Sciences, Kurukshetra University, Kurukshetra 136119, India. Tel: +91-9416025460. E-mail: sharma_prabodh@rediffmail.com

(Received 02 July 2011; revised 22 July 2011; accepted 04 August 2011)

The prevalence of newly emerging virulence traits and drug resistance of the pathogens towards antibiotics have become a serious problem. Thus, the design of newer agents active against the resistant micro-organisms is of critical importance. Looking at the importance of these heterocyclic nuclei, it is thought of interest to accommodate 2-aminobenzothiazoles and fluoroquinolones in single molecular framework and screen them for their various biological activities. Due to our continuous interest[23–26] and research program on design and synthesis of novel anti-microbial agents a number of potent fluoroquinolone derivatives[27–29] have been synthesized. Recently, we have reported synthesis of novel fluoroquinolone derivatives annulated with benzothiazoles with promising anti-bacterial activity[30]. It was clearly indicated that clubbing of fluoroquinolones with benzothiazoles is worthwhile and the synthesized compounds were of promising pharmacological significance. On the basis of these facts, we are here reporting novel synthesis and anti-bacterial, analgesic and anthelmintic activities of substituted benzothiazole derivatives clubbed with fluoroquinolones.

Materials and methods

Chemistry

Chemicals and all solvents used in this study were procured from Merck AG (Mumbai, India), SD Fines (Mumbai, India), Sigma Aldrich (Bangalore, India) and Qualigens (Navi Mumbai, India). Melting points were determined on a Labindia MR-VIS visual melting range apparatus (Mumbai, India) and are uncorrected. The infrared (IR) spectra were recorded on a Perkin Elmer (Waltham, MA), IR spectrophotometer (potassium bromide disk). ^{1}H NMR spectra were recorded using Bruker 400 (Fallanden, Switzerland) spectrometer and chemical shifts are expressed as δ (ppm) with tetramethylsilane as an internal standard.

General procedure for synthesis

Procedure of synthesis of 2-amino-substituted benzothiazoles (2a-g)

The requisite 2-aminobenzothiazoles **2a-g** were prepared by reaction of bromine (4mL, 0.05 mol) dissolved in glacial acetic acid (37.5 mL) which was added drop wise with stirring to a solution of (0.05 mol) of aniline **1a-g** and potassium thiocyanate (19.43 g) dissolved in 90 mL of 96% glacial acetic acid while the temperature was kept below 35°C. After all the bromine solution has been added, the mixture was stirred for ten hours and then filtered and the residue was washed with water. The combined filtrate and the washings were neutralized with ammonium hydroxide. The precipitate was collected on filter and dried.

Procedure of synthesis of 2-(2-chloroacetylamino)-substituted benzothiazoles (3a-e)

Equimolar solution of benzothiazole (0.01 mmol) and chloroacetyl chloride (0.01 mmol) in chloroform (30 mL) in the presence of K_2CO_3 was refluxed at 80–85°C for 12 h. Excess of solvent was removed *in vacuo* and the residue was stirred with water (50 mL). The residue was washed with 5% $NaHCO_3$ and subsequently with water. The product was dried and recrystallised from methanol.

Procedure of synthesis of fluorquinolone derivatives bearing acetamide linkage (5a-o)

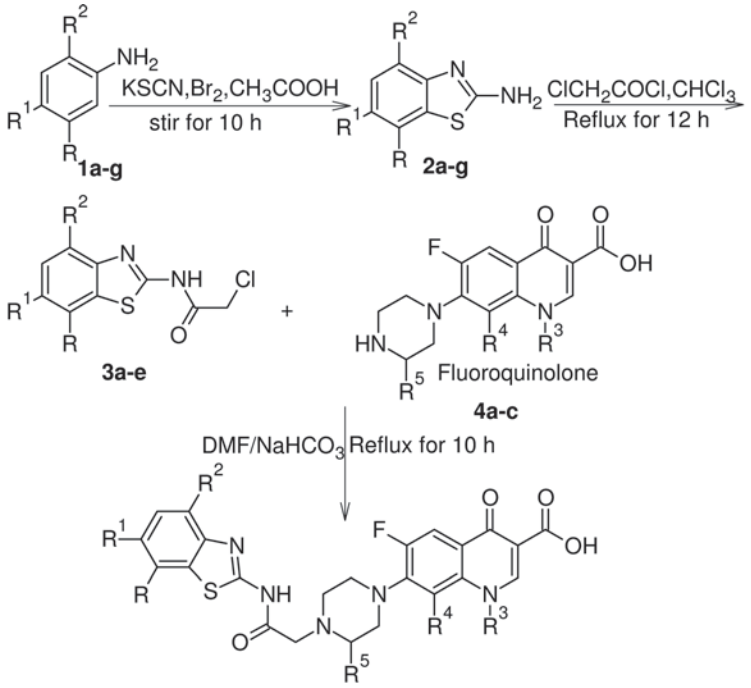
A mixture of benzothiazole **3a-e** (0.05 mmol), fluoroquinolone **4a-e** (0.05 mmol) and $NaHCO_3$ (0.05 mmol) in DMF (10 mL), was heated at 85–90°C. After consumption of fluoroquinolone (monitored by TLC), H_2O (20 mL), was added and the precipitate was filtered and washed with water to give product. Purification was achieved by passage through silica gel column (chloroform-ethanol; 95:5). The product was crystallized from DMF-H_2O to give **5a-o**. The novel derivatives were achieved through the versatile and efficient synthetic route outlined in Scheme 1 (Figure 1).

1-Cyclopropyl-6-fluoro-7-(4-(N-(6-chloro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl)piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5a)**. Yield 67%; m.p. 252–258°C; IR (cm^{-1}): 3340 (NH str), 3076–2890 (C-H str), 1715 (C=O str), 1666 (CONH str), 1628 (C=O str), 1528 (C=C str), 1257 (C-O str), 1142 (C-N str); **^{1}H NMR (DMSO-d$_6$) δ ppm:** 1.34 (m, 4H, -CH_2CH_2- cyclopropyl), 3.28–3.84 (m, 9H, piperazine-H and cyclopropyl-H), 4.16 (s, 2H, -CH_2 methylene bridge), 4.84 (s, 1H, -NH), 7.02–7.96 {m, 5H, aromatic (H_5, H_8-quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$-benzothiazole)}, 8.12 (s, 1H, H_2-quinolone), 15.08 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-7-(4-(N-(4,7-dichloro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1-,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5b)**. Yield 66%; m.p. 241–244°C; IR (cm^{-1}): 3320 (NH str), 3156–2877 (C-H str), 1705 (C=O str), 1674 (CONH str), 1628 (C=O str), 1535 (C=C str), 1257 (C-O str), 1188 (C-N str); **^{1}H NMR (DMSO-d$_6$) δ ppm:** 1.25 (m, 4H, -CH_2CH_2- cyclopropyl), 3.36–3.93 (m, 9H, piperazine-H and cyclopropyl-H), 4.32 (s, 2H, -CH_2 methylene bridge), 5.00 (s, 1H, -NH), 7.14–7.87 {m, 4H, aromatic (H_5, H_8- quinolone and $H_{5'}$, $H_{6'}$- benzothiazole)}, 8.79 (s, 1H, H_2-quinolone), 14.9 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-7-(4-(N-(6-nitro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5c).** Yield 68%; m.p. 262–265°C; IR (cm^{-1}): 3360 (NH str), 3155–2839 (C-H str), 1720 (C=O str), 1674 (CONH str), 1630 (C=O str), 1528 (C=C str), 1252 (C-N str), 1166(C-N str); **^{1}H NMR (DMSO-d$_6$) δ ppm:** 1.23 (m, 4H, -CH_2CH_2- cyclopropyl), 3.32–3.96 (m, 9H, piperazine-H and cyclopropyl-H), 4.12 (s, 2H, -CH_2 methylene bridge), 4.76 (s, 1H, -NH), 7.21–7.84 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.89 (s, 1H, H_2-quinolone), 15.05 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-7-(4-(N-(6-methyl-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5d).**



Compd	R	R^1	R^2	R^3	R^4	R^5	Mol. Formula	Compd	R	R^1	R^2	R^3	R^4	R^5	Mol. Formula
5a	-H	-Cl	-H		-H	-H	$C_{26}H_{23}ClFN_5O_4S$	**5i**	-H	-H	$-CH_3$	$-CH_2CH_3$	-H	-H	$C_{26}H_{26}FN_5O_4S$
5b	-Cl	-H	-Cl		-H	-H	$C_{26}H_{22}Cl_2FN_5O_4S$	**5j**	-H	-H	-F	$-CH_2CH_3$	-H	-H	$C_{25}H_{23}F_2N_5O_4S$
5c	-H	$-NO_2$	-H		-H	-H	$C_{26}H_{23}FN_6O_6S$	**5k**	-H	-H	-Cl		$-OCH_3$	$-CH_3$	$C_{27}H_{27}ClFN_5O_5S$
5d	-H	$-CH_3$	-H		-H	-H	$C_{27}H_{26}FN_5O_4S$	**5l**	-Cl	-Cl	-H		$-OCH_3$	$-CH_3$	$C_{27}H_{26}Cl_2N_5O_5S$
5e	-H	-F	-H		-H	-H	$C_{25}H_{23}F_2N_5O_4S$	**5m**	-H	-H	$-NO_2$		$-OCH_3$	$-CH_3$	$C_{27}H_{27}FN_6O_7$
5f	-H	-Cl	-H	$-CH_2CH_3$	-H	-H	$C_{25}H_{23}ClFN_5O_4S$	**5n**	-H	-H	$-CH_3$		$-OCH_3$	$-CH_3$	$C_{28}H_{30}FN_5O_5S$
5g	-Cl	-H	-Cl	$-CH_2CH_3$	-H	-H	$C_{25}H_{22}Cl_2FN_5O_4S$	**5o**	-H	-H	-F		$-OCH_3$	$-CH_3$	$C_{27}H_{27}F_2N_5O_5$
5h	-H	$-NO_2$	-H	$-CH_2CH_3$	-H	-H	$C_{25}H_{23}FN_6O_6S$								

Figure 1. Synthetic scheme for preparation of title compounds.

Yield 61%; m.p. 252–255°C; IR (cm^{-1}): 3380 (NH str), 3183–2889(C-H str), 1713 (C=O str), 1674 (CONH str), 1628 (C=O str), 1551 (C=C str), 1227 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.24 (m, 4H, $-CH_2CH_2-$ cyclopropyl), 2.44 (m, 3H, $-CH_3$ benzothiazole), 3.60–3.94 (m, 9H, piperazine-H and cyclopropyl-H), 4.35 (s, 2H, $-CH_2$ methylene bridge), 4.86 (s, 1H, -NH), 7.21–7.87 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.26 (s, 1H, H_2-quinolone), 15.04 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-7-(4-(N-(6-fluoro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1-,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5e).** Yield 70%; m.p. 265–268°C; IR (cm^{-1}): 3360 (NH str), 3068–2876 (C-H str), 1710 (C=O str), 1666 (CONH str), 1635 (C=O str), 1535 (C=C str), 1150 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.14 (m, 4H, $-CH_2CH_2-$ cyclopropyl), 3.46–3.74 (m, 9H, piperazine-H and cyclopropyl-H), 4.24 (s, 2H, $-CH_2$ methylene bridge), 4.82 (s, 1H, -NH), 6.95–7.72 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.05 (s, 1H, H_2-quinolone), 15.10 (s br, 1H, -COOH).

1-Ethyl-6-fluoro-7-(4-(N-(6-chloro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4--oxo-quinoline-3-carboxylic acid **(5f).** Yield 58%; m.p. 232–235°C; IR (cm^{-1}): 3350 (NH str), 3056–2833 (C-H str), 1721 (C=O str), 1674 (CONH str), 1628 (C=O str), 1551 (C=C str), 1257 (C-N str), 1111 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.35 (t, 3H, $-CH_3$ ethyl), 2.24–2.36 (m, 2H, $-CH_2$ ethyl), 3.34 (m, 8H, piperazine-H), 4.02 (s, 2H, $-CH_2$ methylene bridge), 4.85 (s, 1H, -NH), 6.98–7.86 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.56 (s, 1H, H_2-quinolone), 14.94 (s br, 1H, -COOH).

1-Ethyl-6-fluoro-7-(4-(N-(4,7-dichloro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5g).** Yield 54%; m.p. 212–215°C; IR (cm^{-1}): 3350 (NH str), 3056–2833 (C-H str), 1713 (C=O str), 1688 (CONH str), 1621 (C=O

str), 1551 (C=C str), 1257 (C-N str), 1112 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.35 (t, 3H, -CH_3 ethyl), 2.24–2.36 (m, 2H, -CH_2 ethyl), 2.98–3.31 (m, 8H, piperazine-H), 4.31 (s, 2H, -CH_2 methylene bridge), 5.10 (s, 1H, -NH), 7.12–7.86 {m, 4H, aromatic (H_5, H_8- quinolone and $H_{5'}$, $H_{6'}$- benzothiazole)}, 8.92 (s, 1H, H_2-quinolone), 14.94 (s br, 1H, -COOH).

1-Ethyl-6-fluoro-7-(4-(N-(6-nitro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4--oxo-quinoline-3-carboxylic acid **(5h).** Yield 57%; m.p. 222–225°C; IR (cm^{-1}): 3350 (NH str), 3056–2833 (C-H str), 1721 (C=O str), 1674 (CONH str), 1628 (C=O str), 1551 (C=C str), 1257 (C-N str), 1162 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.38 (t, 3H, -CH_3 ethyl), 2.28 (m, 2H, -CH_2 ethyl), 3.16–3.72 (m, 8H, piperazine-H), 4.26 (s, 2H, -CH_2 methylene bridge), 4.87 (s, 1H, -NH), 6.98–7.94 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.24 (s, 1H, H_2-quinolone), 15.03 (s br, 1H, -COOH).

1-Ethyl-6-fluoro-7-(4-(N-(6-methyl-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5i).** Yield 52%; m.p. 196–199°C; IR (cm^{-1}): 3350 (NH str), 3086–2834 (C-H str), 1713 (C=O str), 1674 (CONH str), 1620 (C=O str), 1535 (C=C str), 1257 (C-N str), 1179 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.36 (t, 3H, -CH_3 ethyl), 2.24–2.34 (m, 2H, -CH_2 ethyl), 2.48 (m, 3H, -CH_3 benzothiazole) 3.12–2.37 (m, 8H, piperazine-H), 4.10 (s, 2H, -CH_2 methylene bridge), 4.87 (s, 1H, -NH), 6.87–7.86 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.32 (s, 1H, H_2-quinolone), 15.04 (s br, 1H, -COOH).

1-Ethyl-6-fluoro-7-(4-(N-(6-fluoro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4--oxo-quinoline-3-carboxylic acid **(5j).** Yield 53%; m.p. 206–209°C; IR (cm^{-1}): 3350 (NH str), 3156–2839 (C-H str), 1720 (C=O str), 1674 (CONH str), 1620 (C=O str), 1535 (C=C str), 1257 (C-N str), 1165 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.26 (t, 3H, -CH_3 ethyl), 1.96 (m, 2H, -CH_2 ethyl), 2.94–3.89 (m, 8H, piperazine-H), 4.14 (s, 2H, -CH_2 methylene bridge), 4.84 (s, 1H, -NH), 7.09–8.25 {m, 5H, aromatic (H_5, H_8- quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.24 (s, 1H, H_2-quinolone), 13.87 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-8-methoxy-7-(3-methyl-4-(N-(6-chloro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5k).** Yield 64%; m.p. 183–186°C; IR (cm^{-1}): 3340 (NH str), 3052–2854 (C-H str), 1710 (C=O str), 1674 (CONH str), 1628 (C=O str), 1551 (C=C str), 1257 (C-N str), 1174 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.24 (m, 4H, -CH_2CH_2- cyclopropyl), 2.19–2.94 (m, 3H, -CH_3 piperazine) 3.33–3.74 (m, 8H, piperazine-H and cyclopropyl-H), 3.75 (s, 3H, OCH_3), 4.16 (s, 2H, -CH_2 methylenee bridge), 4.86 (s, 1H, -NH), 6.96–7.86 {m, 4H, aromatic (H_5, quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.69 (s, 1H, H_2-quinolone), 15.13 (s, 1H, -COOH).

1-Cyclopropyl-6-fluoro-8-methoxy-7-(3-methyl-4-(N-(4,7-dichloro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5l).** Yield 59%; m.p. 244–247°C; IR (cm^{-1}): 3350 (NH str), 3086–2854 (C-H str), 1713 (C=O str), 1666 (CONH str), 1620 (C=O str), 1528 (C=C str), 1273 (C-N str), 1161 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.21 (m, 4H, -CH_2CH_2- cyclopropyl), 2.19–2.94 (m, 3H, -CH_3 piperazine) 3.31–3.72 (m, 8H, piperazine-H and cyclopropyl-H), 3.78 (s, 3H, OCH_3), 4.08 (s, 2H, -CH_2 methylene bridge), 4.89 (s, 1H, -NH), 7.18–7.87 {m, 3H, aromatic (H_5, quinolone and $H_{5'}$, $H_{6'}$- benzothiazole)}, 9.04 (s, 1H, H_2-quinolone), 14.09 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-8-methoxy-7-(3-methyl-4-(N-(6-nitro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5m).** Yield 57%; m.p. 182–185°C; IR (cm^{-1}): 3310 (NH str), 3086–2839 (C-H str), 1720 (C=O str), 1670 (CONH str), 1628 (C=O str), 1551 (C=C str), 1257 (C-N str), 1154 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.12 (m, 4H, -CH_2CH_2- cyclopropyl), 2.46–2.76 (m, 3H, -CH_3 piperazine), 3.36–3.86 (m, 8H, piperazine-H and cyclopropyl-H), 3.72 (s, 3H, OCH_3), 4.24 (s, 2H, -CH_2 methylene bridge), 4.89 (s, 1H, -NH), 7.10–7.96 {m, 3H, aromatic (H_5, quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.76 (s, 1H, H_2-quinolone), 14.9 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-8-methoxy-7-(3-methyl-4-(N-(6-methyl-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5n).** Yield 53%; m.p. 203–206°C; IR (cm^{-1}): 3370 (NH str), 3056–2827 (C-H str), 1713 (C=O str), 1672 (CONH str), 1620 (C=O str), 1557 (C=C str), 1258 (C-N str), 1166 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.0 (m, 4H, -CH_2CH_2- cyclopropyl), 2.42–2.69 (m, 3H, -CH_3 piperazine), 2.48 (m, 3H, -CH_3 benzothiazole), 3.46–3.84 (m, 8H, piperazine-H and cyclopropyl-H), 3.72 (s, 3H, OCH_3), 4.16 (s, 2H, -CH_2 methylene bridge), 4.97 (s, 1H, -NH), 7.24–7.93 {m, 4H, aromatic (H_5, quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 8.27 (s, 1H, H_2-quinolone), 14.9 (s br, 1H, -COOH).

1-Cyclopropyl-6-fluoro-8-methoxy-7-(3-methyl-4-(N-(6-fluoro-1,3-benzothiazol-2-yl)amino)-2-oxoethyl) piperazin-1-yl)-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid **(5o).** Yield 56%; m.p. 226–229°C; IR (cm^{-1}): 3380 (NH str), 3155–2839 (C-H str), 1720 (C=O str), 1674 (CONH str), 1620 (C=O str), 1551 (C=C str), 1258 (C-N str), 1131 (C-N str); **^{1}H NMR (DMSO-d_6) δ ppm:** 1.12 (m, 4H, -CH_2CH_2- cyclopropyl), 2.92 (m, 3H, -CH_3 piperazine) 3.74 (m, 8H, piperazine-H and cyclopropyl-H), 3.78 (s, 3H, OCH_3), 4.04 (s, 2H, -CH_2 methylene bridge), 5.13 (s, 1H, -NH), 7.24–7.83 {m, 4H, aromatic (H_5, quinolone and $H_{4'}$, $H_{5'}$, $H_{7'}$- benzothiazole)}, 9.04 (s, 1H, H_2-quinolone), 15.04 (s br, 1H, -COOH).

Pharmacological screening

Anti-bacterial activity

The anti-bacterial activity was determined against two gram negative bacteria i.e., *E. coli* (NCDC 134), *P.*

aeroginosa (NCDC105) and three gram positive strains i.e., *B. subtilis* (NCDC 71), *B. polymoxa* (NCDC 64) and *S. aureus* (NCDC 110). The synthesized compounds were screened for anti-bacterial activity by minimum inhibitory concentration method[31]. Revival of these strains was done as per protocol provided by NDRI, Karnal. The bacterial cultures were stored at 4°C and culture was maintained on nutrient agar plate. MIC values of the synthesized compounds determined against various strains are reported in Table 1.

Calculation of log *P*

To investigate the activity of the synthesized compounds against gram negative and gram positive bacteria, the quantitative effects of lipophillicity on anti-bacterial activity were also studied. Partition coefficient, log *P* is an important parameter for anti-bacterial activity, usually related to its pharmacological activity. Log *P* is a measure of hydrophobicity, which is not only important for the penetration and distribution of a drug but also for the interaction of the drug with the receptor. Log *P* was

Table 1. Minimum inhibitory concentration (MIC) in μg/mL against *B. polymyxa, B. subtilis, E. coli, P. aeruginosa, S. aureus* bacterial strains.

	Gram (+)			Gram (–)	
Compounds	*B. subtilis* (NCDC 71)	*B. polymyxa* (NCDC 64)	*S. aureus* (NCDC 110)	*E. coli* (NCDC 134)	*P. aeruginosa* (NCDC 105)
5a	04	15	03	08	15
5b	10	30	05	06	65
5c	25	55	50	35	45
5d	35	110	40	20	45
5e	40	125	35	80	60
5f	02	25	15	50	10
5g	20	15	40	60	30
5h	15	75	105	125	55
5i	30	45	25	65	20
5j	65	55	45	35	40
5k	75	25	04	60	20
5l	25	25	10	70	30
5m	40	45	60	65	75
5n	120	60	70	75	85
5o	70	25	15	140	50
Ciprofloxacin	20	10	50	25	50
Norfloxacin	05	25	10	40	15
Gatifloxacin	80	10	05	100	40

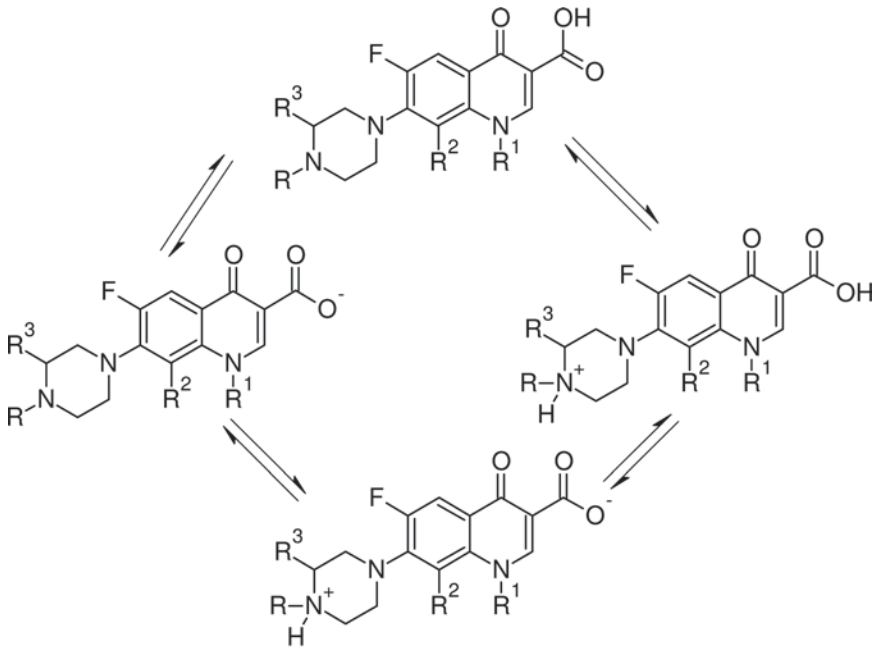


Figure 2. The possible protonation states of fluoroquinolones.[35]

calculated online by molinspiration software from the Website (http://www.molinspiration.com/cgi-bin/properties). The value of log *P* was used as such given by the software in the biological study.

Correlation between partition coefficient log *P* and log MIC

To determine the relationship between the calculated log *P* and log MIC value, a simple regression was performed. The partition of quinolones was generally consistent with the knowledge that the neutral species of like compounds is more lipophilic than the zwitterions species (Figure 2). The ratio of the zwitter ions to neutral species is very important feature to describe the lipophilicity of fluoroquinolones[32]. The following regression equation was, therefore, applied. The value of log MIC, log *P* and correlation coefficient (*r*) have been summarised in Tables 2 and 3.

$$Y = a\log X^2 + b\log X + c$$

where *Y*=log MIC; *X*=log *P*; and *a*, *b* and *c* are constants.

Correlation between molecular mass and log MIC

The molecular mass and bulkiness at the substituent of the substitution at C-7 position hinder penetration of quinolones into gram positive bacteria through the porine channel, although hydrophobic molecules appear to enter via lipopolysaccharide or across the lipid bilayer[33]. Conversely, gram positive bacteria do not possess an outer membrane, and so lack the outer membrane protein and lipopolysaccharide. Hence the accumulation of the drugs in gram positive bacteria takes place thorough simple diffusion across the cytoplasmic membrane and not affected by increasing the bulkiness of the N-7 substitution[34]. The value of log MIC, molecular mass and correlation coefficient (*r*) have been summarised in Tables 2 and 3. Correlation coefficient was calculated by the formula as given below:

$$r = \frac{N\sum XY - \sum X \sum Y}{\sqrt{N\sum X^2 - (\sum X)^2}\sqrt{N\sum Y^2 - (\sum Y)^2}}$$

where r is correlation coefficient, X series are Log *P* and molecular mass, and Y series are Log MIC.

Anthelmintic activity

In the present work, anthelmintic activity of newly synthesized compounds was evaluated using a variety of earthworms. Earthworms (*Pheritima posthuma*) were procured from Department of Agriculture, Gurukul, Kurukshetra. Suspension of the samples were prepared by triturating the samples with 0.5% tween 80 and distilled water. The resulting mixtures were then stirred using a mechanical stirrer for 30 min. The resulting suspensions were diluted to contain 200 mg in 5 mL of test samples. These suspensions were used for anthelmintic studies. The standard drug piperazine citrate was also used in the form of the suspension with the same concentration in the same way.

Table 2. Log *P*, molecular mass and log MIC of the compounds 5a-o.

	Correlation parameters		Log MIC				
Compounds	Log *P*	Mol. Mass	*B. subtilis*	*B. polymyxa*	*S. aureus*	*E. coli*	*P. aeruginosa*
5a	2.27	556.00	0.60	1.76	0.47	0.90	1.17
5b	2.88	590.41	1.00	1.47	0.69	0.77	1.81
5c	1.55	566.52	1.39	1.74	1.69	1.54	1.65
5d	2.04	535.51	1.54	2.04	1.60	1.30	1.65
5e	1.76	539.53	1.60	2.09	1.54	1.90	1.77
5f	2.28	543.94	0.30	1.39	1.17	1.69	1.00
5g	2.89	578.43	1.30	1.76	1.06	1.77	1.47
5h	1.56	554.55	1.17	1.87	2.02	2.09	1.74
5i	2.05	523.64	1.47	1.65	1.39	1.81	1.30
5j	1.77	527.51	1.81	1.74	1.65	1.54	1.60
5k	2.59	600.02	1.87	1.39	0.60	1.77	1.30
5l	3.19	634.52	1.39	1.39	1.00	1.84	1.47
5m	1.87	610.63	1.60	1.65	1.77	1.84	1.87
5n	2.36	579.65	2.07	1.77	1.84	1.87	1.92
5o	2.07	583.61	1.84	1.39	1.17	2.14	1.69

Table 3. Correlation coefficient (*r*) between log MIC of tested strains and log *P* and molecular mass of compounds 5a-o.

	Correlation coefficient (*r*)	
Bacterial strains	Log *P*	Molecular mass
B. subtilis	0.72	0.32
B. polymoxa	0.52	0.26
S. aureus	0.79	0.47
E. coli	0.58	0.36
P. aeruginosa	0.62	0.29

Five earthworms of a variety and similar size were placed in a petri dish of 4 inches diameter containing 50 mL suspension of the standard drug (piperazine citrate) at room temperature. Another set of earthworms was kept as control in 50 mL suspension of distilled water and 0.5% tween 80. Then, 50 mL each of the suspension of test samples were added into separate petri dish containing five earthworms in each. The time required for paralysis and death were recorded. The death time was ascertained by placing the paralysed worm warm water at 50°C, which stimulated the movement if the worm was alive. The mean paralysing time and mean death time was calculated[36]. The process was repeated with different varieties of earthworms. Results of anthelmintic activity have been summarized in Table 4.

Analgesic activity

Swiss albino mice of either sex weighing 25–30 g were procured from diseases free animal house CCS, Agricultural University, Hisar, Haryana, India. The animals were housed in animal house of Institute of Pharmaceutical Sciences, Kurukshetra University, Kurukshetra, in polycarbonate cages in a room maintained under controlled room temperature 22 ± 2°C, relative humidity 60–70%, and provided with food and water *ad libitum*. All the experimental procedures and protocols used in the study were reviewed by the Institutional Animal Ethics Committee (Registration No 563/02/a/CPCSEA) and were in accordance with the guidelines of the CPCSEA, Ministry of Forests and Environment, Government of India. The animals were deprived of food for 24 h before experimentation but allowed free access to water throughout.

The analgesic activity was measured against chemical stimulus. For analgesic activity, the animals were divided into groups consisting of six mice each. The control group received normal saline: Tween 80 (95:5). The standard group received diclofenac sodium 50 mg/Kg body weight and the test groups received the synthetic compounds at a dose of 50 mg/Kg body weight. Thirty minutes later, nociception was induced by an intraperitoneal (IP) injection of acetic acid (1.0%), 0.1 mL/10 g[37]. The number of stretching or writhing was recorded from 5 to 15 min. Results have been summarized in Table 5.

The percentage protection was calculated by the following formula:

$$\%\ \text{Protection} = 100 - (\text{No. of writhes in test} / \text{No. of writhes in control}) \times 100$$

Results and discussion

Synthetic chemistry

The synthesis of various novel fluoroquinolone derivatives was achieved in a 3-step procedure employing various substituted anilines **1a-g** as starting material. Compound **2a-g** were synthesized from substituted anilines **1a-g,** in the presence of potassium thiocynate (ambidient nucleophile) from 1-phenyl urea in acidic medium. This substituted 1-phenyl thiourea in presence of oxidising agent i.e., bromine was cyclised into substituted 2-amino benzothiazoles. Compound **2a-g** on condensation with chloroacetyl chloride in the presence of K_2CO_3 as base and chloroform as solvent gave 2-(2-chloroacetyl amino)-substituted benzothiazoles **3a-e.** Compounds **3a-e** were reacted with fluoroquinolones **4a-c** in the presence of sodium bicarbonate as base and

Table 4. Anthelmintic activity of synthesized derivatives.

Compounds	Concentration of compounds (mg/100 mL)	Mean paralysing time (min.) + SE.	Mean death time (min) + SE.
5a	200	26.00 ± 2.00*	38.40 ± 0.92**
5b	200	32.60 ± 1.07	43.20 ± 1.24
5c	200	22.80 ± 1.86**	38.20 ± 0.58**
5d	200	23.20 ± 1.28**	44.40 ± 1.20
5e	200	27.80 ± 0.66	44.40 ± 1.28
5f	200	23.60 ± 0.67**	39.20 ± 1.15**
5g	200	26.80 ± 1.49*	43.20 ± 2.08
5h	200	27.00 ± 1.41	40.40 ± 1.03*
5i	200	28.40 ± 1.16	47.80 ± 1.49
5j	200	23.60 ± 1.80**	42.00 ± 1.00
5k	200	24.60 ± 1.63	43.60 ± 1.90
5l	200	29.00 ± 2.70	44.80 ± 1.39
5m	200	25.60 ± 1.20**	38.40 ± 0.67**
5n	200	32.40 ± 1.96	48.00 ± 1.30
5o	200	23.60 ± 1.20**	43.80 ± 1.53
Control	–	–	–
Standard	200	34.40 ± 1.03	44.80 ± 1.02

Statistical analysis: All the results were expressed as mean± standard mean error (SEM). Statistical analysis was done by using one way ANOVA followed by Dunnett's '*t*' test and critical range for significance difference between two group of observation was taken as *$p < 0.05$, **$p < 0.01$ and ***$p < 0.001$, compared with control.

Table 5. Analgesic activity of the title compounds by acetic acid-induced writhing method.

Compounds	Dose (mg/Kg)	No. of writhes mean ± SEM (% inhibition)	(%) Inhibition
5a	50	13.80 ± 0.80*	55.19
5b	50	15.60 ± 1.32	49.35
5c	50	14.20 ± 1.14*	53.89
5d	50	16.20 ± 0.86	47.40
5e	50	17.00 ± 0.23	44.80
5f	50	15.20 ± 1.20	50.64
5g	50	17.40 ± 1.66	43.50
5h	50	17.80 ± 2.87	42.20
5i	50	17.80 ± 2.47	42.20
5j	50	18.60 ± 1.63	39.61
5k	50	19.20 ± 1.65	37.66
5l	50	14.80 ± 0.73*	51.94
5m	50	17.60 ± 1.63	42.85
5n	50	20.80 ± 2.03	32.46
5o	50	15.00 ± 0.70*	44.80
Control	50	30.80 ± 0.37	–
Standard	50	10.40 ± 0.24	67.23

Statistical analysis: All the results were expressed as mean± standard mean error (SEM). Statistical analysis was done by using one way ANOVA followed by Dunnett's '*t*' test and critical range for significance difference between two group of observation was taken as $^{**}p < 0.01$ and $^{***}p < 0.001$, compared with control.

DMF was heated. The product was recrystallised from DMF-water to give **5a-o.**

The synthesized compounds are characterised by IR spectra of analog **5a-o** displaying characteristic absorption in the region 1721–1705 cm^{-1} showing the presence of C=O *str.* NMR spectral data of fluoroquinolone derivatives **5a-o** singlet at δ 4.32–4.12 (2H) ppm corresponding to proton of methylene bridge, clearly indicating the presence of acetamide linkage in them.

Pharmacological screening

Keeping in view the biological potential of fluoroquinolones, it was decided to perform biological evaluation of all the newly synthesized fluoroquinolone derivatives **5a-o.** Hence, all the synthesized compounds were subjected to anti-bacterial, analgesic and anthelmintic screening in order to evaluate their pharmacological potential.

Anti-bacterial activity

The newly synthesized compounds were screened for their *in vitro* anti-bacterial activity against bacterial strains. Compounds **5a-o** were evaluated for their anti-bacterial activity by MIC method, against two gram negative bacteria i.e., *E. coli* (NCDC 134), *P. aeruginosa* (NCDC105) and three gram positive strains i.e., *B. subtilis* (NCDC 71), *B. polymyxa* (NCDC 64) and *S. aureus* (NCDC 110). The major findings indicated that most of the compounds were more active against gram positive rather than gram negative bacteria. Compounds with chloro substituted benzothiazole substituted ring compounds were observed to be more potent than bromo, nitro, fluoro and alkyl substituted compounds. For example, **5a, 5b, 5f, 5k, 5l** showed potent activity against gram negative and gram positive bacteria.

Compounds **5a, 5b, 5k, 5f** and **5l** were found to depict even better than standard antibiotics ciprofloxacin, norfloxacin and gatifloxacin against *S. aureus*. The compounds **5a, 5f, 5k** and **5l** (MIC = 15–25 μg/mL) showed even better MIC values as compared to the standard antibiotics ciprofloxacin, norfloxacin and gatifloxacin (MIC = 10, 20 and 10 μg/mL) when tested against *B. polymyxa*. Analogs **5a** and **5f** (MIC = 04 and 02 μg/mL) are even better than the standard antibiotics ciprofloxacin, norfloxacin and gatifloxacin (MIC = 20, 05 and 80 μg/mL) against *B. subtilis*. Compounds **5c, 5d** and **5g** (MIC = 20–35 μg/mL) showed comparable MIC against standard antibiotics ciprofloxacin, norfloxacin and gatifloxacin (MIC = 25, 40 and 100 μg/mL) when evaluated against *E. coli*. Compounds **5a** and **5f** (MIC = 15 and 10 μg/mL) showed even better MIC against the standard antibiotics ciprofloxacin, norfloxacin and gatifloxacin (MIC = 50, 15 and 40 μg/mL) when tested against *P. aeruginosa*. The results of MIC test against gram-positive and Gram negative bacteria revealed that ciprofloxacin derivatives were more potent than the norfloxacin and gatifloxacin derivatives. These data confirm that the effect of changes in side chain of 7-piperazinyl ring mainly depends on the substitution at N-4 position, linkage group and position of the substituted ring.

The correlation between calculated Log *P* and Log MIC **5a-o** in case of *B. subtilis* and *S. aureus* showed strong positive correlation ($r = 0.72$ and 0.79). However, such correlation could not be established in case of *B. polymyxa, E. coli, P. aeruginosa* ($r = 0.52$, 0.58, 0.62, respectively). These correlation results revealed that lipophilicity of the molecule and penetration into the cell membrane is not the only factor which can affect the activity. Indeed, other factors such as the affinity of the compounds for their target DNA gyrase and

topoisomerase IV can affect their MIC. Furthermore, the heterogeneous nature and difference in the steric effect of the tested *N-4*-piperazinyl substituted may play important roles in the accommodation of these molecules on the active sites of the enzyme. The correlation between the molecular mass and Log MIC for tested compounds **5a-o** results in a weaker correlation ($r = 0.32$, 0.26, 0.47, 0.36 and 0.29) for *B. subtilis, B. polymyxa, S. aureus, E. coli* and *P. aeruginosa*, respectively. These results indicate that the activity of compounds not affected by increasing the bulkiness of the N-7 substituent's in the present investigation.

Anthelmintic activity

The synthesized compounds were also evaluated for the anthelmintic activity done against *Eisemia foetida*. The results of this study revealed that all the compounds exhibited promising anthelmintic activity at low concentrations. Compound **5a, 5c, 5f, 5h** and **5m** demonstrated potent activity with mean paralysis time ranging from 22.60 ± 2.46 to 31.60 ± 3.07 min while most of the compounds showed death time in the range of 42.00 ± 1.00 min to 48.60 ± 1.07 min. The standard drug piperazine citrate at concentration of 200 mg/mL showed activity with mean paralysis time 34.40 ± 1.03 min and mean death time 44.80 ± 1.02 min. Results of anthelmintic activity showed that chloro and nitro substituted compounds were more potent than other substitution at benzothiazole ring. This showed that the increase electronegativity of the substituted group may increase the activity of the compounds.

Analgesic activity

All the derivatives were also screened for analgesic activity by adopting standard protocol viz. acetic acid-induced abdominal writhing test. Swiss albino mice weighing 25–30 g were used for carrying the evaluation. For abdominal writhing test, nociception was induced by an intraperitoneal (IP) injection of acetic acid and diclofenac sodium was used as a standard drug. The number of stretching or writhing were recorded for 5–15 min. All the compounds exhibited significant analgesic activities. Among all, some compounds were observed to be most active and depicted 47.40 and 55.19% inhibition of writhing in comparison to the standard drug diclofenac sodium (67.23%). From the results of analgesic activity, it was clearly revealed that compounds with chloro substitution at benzothiazole ring exhibit more potent activity than other groups.

Conclusion

In this paper, we present a novel array of fluoroquinolone clubbed with benzothiazole derivatives which are screened for anti-bacterial, anthelmintic and analgesic activities. The experimental data reveal that the synthesized compounds exhibit potent anti-bacterial activity as compared to the standard drugs ciprofloxacin, norfloxacin and gatifloxacin. They have also shown promising anthelmintic activity and good *in vivo* analgesic activity. These compounds may serve as useful lead molecules for new antibiotic discoveries.

Acknowledgement

Prof. Om Prakash, Director, Institute of Pharmaceutical Sciences and Lt. Gen. (Dr.) D.D.S. Sandhu, Vice Chancellor, Kurukshetra University, India, are duly acknowledged for providing necessary facilities.

Declaration of interest

The authors report no conflicts of interest. The authors are alone responsible for the content and writing of the paper.

References

1. Sharma PC, Jain A, Jain S. Fluoroquinolone antibacterials: A review on chemistry, microbiology and therapeutic prospects. Acta Pol Pharm 2009;66:587–604.
2. Sharma PC, Saneja A, Jain S. Norfloxacin: A therapeutic review. Int J Chem Sci 2008;6:1702–1713.
. Sharma PC, Jain A, Jain S. Therapeutic perspectives of fluoroquinolones: An update. In proceedings of the 1st Rashtriya Yuva Vagyanik Sammelan, Kurukshetra, November 28–30, 2008: pp. 237–239.
4. Tillotson GS. Quinolones: Structure-activity relationships and future predictions. J Med Microbiol 1996;44:320–324.
5. Sarkozy G. Quinolones: A class of anti-microbial agents. Vet Med Czech 2001;46:257–274.
6. Drlica K. Mechanism of fluoroquinolone action. Curr Opin Microbiol 1999;2:504–508.
7. Sharma PC, Jain A, Jain S, Pahwa R, Yar MS. Ciprofloxacin: Review on developments in synthetic, analytical, and medicinal aspects. J Enzyme Inhib Med Chem 2010;25:577–589.
8. Sharma PC, Alawadhi N, Sharma A, Pahwa R, Rajak H. Fluoroquinolones in urinary tract infections: A study on perception analysis of prescribers. Der Chem Sinica 2010;1:84–90.
9. Abuo-Rahma Gel-D, Sarhan HA, Gad GF. Design, synthesis, antibacterial activity and physicochemical parameters of novel N-4-piperazinyl derivatives of norfloxacin. Bioorg Med Chem 2009;17:3879–3886.
10. Foroumadi A, Ghodsi S, Emami S, Najjari S, Samadi N, Faramarzi MA et al. Synthesis and antibacterial activity of new fluoroquinolones containing a substituted N-(phenethyl)piperazine moiety. Bioorg Med Chem Lett 2006;16:3499–3503.
11. Asahina Y, Araya I, Iwase K, Iinuma F, Hosaka M, Ishizaki T. Synthesis and antibacterial activity of the 4-quinolone-3-carboxylic acid derivatives having a trifluoromethyl group as a novel N-1 substituent. J Med Chem 2005;48:3443–3446.
12. Asahina Y, Iwase K, Iinuma F, Hosaka M, Ishizaki T. Synthesis and antibacterial activity of 1-(2-fluorovinyl)-7-substituted-4-quinolone-3-carboxylic acid derivatives, conformationally restricted analogues of fleroxacin. J Med Chem 2005;48:3194–3202.
13. Sriram D, Aubry A, Yogeeswari P, Fisher LM. Gatifloxacin derivatives: Synthesis, antimycobacterial activities, and inhibition of Mycobacterium tuberculosis DNA gyrase. Bioorg Med Chem Lett 2006;16:2982–2985.
14. Senthilkumar P, Dinakaran M, Yogeeswari P, Sriram D, China A, Nagaraja V. Synthesis and antimycobacterial activities of novel 6-nitroquinolone-3-carboxylic acids. Eur J Med Chem 2009;44:345–358.

15. Rameshkumar N, Ashokkumar M, Subramanian EH, Ilavarasan R, Sridhar SK. Synthesis of 6-fluoro-1,4-dihydro-4-oxo-quinoline-3-carboxylic acid derivatives as potential antimicrobial agents. Eur J Med Chem 2003;38:1001–1004.
16. Hagihara M, Kashiwase H, Katsube T, Kimura T, Komai T, Momota K et al. Synthesis and anti-HIV activity of arylpiperazinyl fluoroquinolones: A new class of anti-HIV agents. Bioorg Med Chem Lett 1999;9:3063–3068.
17. Azéma J, Guidetti B, Dewelle J, Le Calve B, Mijatovic T, Korolyov A et al. 7-((4-Substituted)piperazin-1-yl) derivatives of ciprofloxacin: Synthesis and *in vitro* biological evaluation as potential antitumor agents. Bioorg Med Chem 2009;17:5396–5407.
18. Korolyov A, Dorbes S, Azéma J, Guidetti B, Danel M, Lamoral-Theys D et al. Novel lipophilic 7H-pyrido[1,2,3-de]-1,4-benzoxazine-6-carboxylic acid derivatives as potential antitumor agents: Improved synthesis and *in vitro* evaluation. Bioorg Med Chem 2010;18:8537–8548.
19. Sharma A, Kumar V, Jain S, Sharma PC. Thiazolidin-4-one and hydrazone derivatives of capric acid as possible anti-inflammatory, analgesic and hydrogen peroxide-scavenging agents. J Enzyme Inhib Med Chem 2011;26:546–552.
20. Kumar V, Sharma A, Sharma PC. Synthesis of some novel 2,5-disubstituted thiazolidinones from a long chain fatty acid as possible anti-inflammatory, analgesic and hydrogen peroxide scavenging agents. J Enzyme Inhib Med Chem 2011;26:198–203.
21. Huang S, Collony PJ. Synthesis of 2-N-alkyl (aryl) amino-7-nitrobenzothiazoles. Tetrahedron Lett 2004;45:9373–9375.
22. Baell JB, Duggan PJ, Forsyth SA, Lewis RJ, Lok YP, Schroeder CI. Synthesis and biological evaluation of nonpeptide mimetics of omega-conotoxin GVIA. Bioorg Med Chem 2004;12:4025–4037.
23. Kharb R, Yar MS, Sharma PC. New insights into chemistry and anti-infective potential of triazole scaffold. Curr Med Chem 2011;18:3265–3297.
24. Sharma PC, Kaur G, Pahwa R, Sharma A, Rajak H. Quinazolines analogs as potential therapeutic agents. Curr Med Chem 2011. Accepted manuscript.
25. Kharb R, Sharma PC, Yar MS. Pharmacological significance of triazole scaffold. J Enzyme Inhib Med Chem 2011;26:1–21.
26. Kharb R, Shahar Yar M, Sharma PC. Recent advances and future perspectives of triazole analogs as promising antiviral agents. Mini Rev Med Chem 2011;11:84–96.
27. Sharma PC, Jain S. Synthesis and in-vitro antibacterial activity of some novel N-nicotinoyl-1-ethyl-6-fluoro-1,4-dihydro-7-piperazin-1-yl-4-oxoquinoline-3-carboxylates. Acta Pol Pharm 2008;65:551–556.
28. Sharma PC, Jain S. Synthesis and *in vitro* anti-bacterial activity of certain novel N-nicotinoyl-1-ethyl-6-fluoro-1,4-dihydro-7-4-subsituted piperazin-1-yl-4-oxoquinoline 3-carboxylates. Acta Pharm Sciencia 2008;50:35–40.
29. Sharma OP, Sharma PC, Jain S. Synthesis and anti-bacterial evaluation of fluoroquinolone derivatives *In*: Proceedings of the International Conference on Bioscience, Biochemistry and Bioinformatics, held at Singapore, February 26–28, 2011: pp. 1–5.
30. Sharma PC, Jain A, ShaharYar M, Pahwa R, Singh J, Goel S. Synthesis and anti-bacterial evaluation of novel analogs of fluoroquinolones annulated with 6-subsituted 2-aminobenzothiazoles. Arabian J Chem 2011. Accepted manuscript doi: 10.1016/j.arabjc.2011.04.008.
31. Bauer AW, Kirby WM, Sherris JC, Turck M. Antibiotic susceptibility testing by a standardized single disk method. Am J Clin Pathol 1966;45:493–496.
32. Ross DL, Riley CM. Physicochemical properties of fluoroquinolone anti-microbials. Int J Pharm 1993;93:121–129.
33. Bazile S, Moreau N, Bouzard D, Essiz M. Relationships among antibacterial activity, inhibition of DNA gyrase, and intracellular accumulation of 11 fluoroquinolones. Antimicrob Agents Chemother 1992;36:2622–2627.
34. Piddock LJ, Jin YF, Griggs DJ. Effect of hydrophobicity and molecular mass on the accumulation of fluoroquinolones by Staphylococcus aureus. J Antimicrob Chemother 2001;47:261–270.
35. Pistos C, Tsantili-Kakoulidou A, Koupparis M. Investigation of the retention/pH profile of zwitterionic fluoroquinolones in reversed-phase and ion-interaction high performance liquid chromatography. J Pharm Biomed Anal 2005;39:438–443.
36. Dahiya R. Synthesis of a phenylalanine-rich peptide as potential anthelmintic and cytotoxic agent. Acta Pol Pharm 2007;64:509–516.
37. Collier HO, Dinneen LC, Johnson CA, Schneider C. The abdominal constriction response and its suppression by analgesic drugs in the mouse. Br J Pharmacol Chemother 1968;32:295–310.