Bioorganic Chemistry 88 (2019) 102965

Contents lists available at ScienceDirect

Bioorganic Chemistry

journal homepage: www.elsevier.com/locate/bioorg

Design, synthesis and molecular modeling studies on novel moxifloxacin derivatives as potential antibacterial and antituberculosis agents☆

Aslı Türe[a], Necla Kulabaş[a], Serap İpek Dingiş[a], Kaan Birgül[a,1], Arif Bozdeveci[b], Şengül Alpay Karaoğlu[b], Vagolu Siva Krishna[c], Dharmarajan Sriram[c], İlkay Küçükgüzel[a,*]

[a] Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Marmara University, Haydarpaşa 34668, İstanbul, Turkey
[b] Department of Biology, Recep Tayyip Erdoğan University, 53100 Rize, Turkey
[c] Medicinal Chemistry Research Laboratory, Pharmacy group, Birla Institute of Technology and Science, 333031 Pilani, India

ARTICLE INFO

Keywords:
Fluoroquinolones
Moxifloxacin
Antibacterial activity
Anti-tuberculosis activity
DNA supercoiling assay
Molecular docking
Resistant strains

ABSTRACT

Twenty-one novel alkyl/acyl/sulfonyl substituted fluoroquinolone derivatives were designed, synthesized and evaluated for their anti-tuberculosis and antibacterial activity. The targeted compounds were synthesized by the introduction of alkyl, acyl or sulfonyl moieties to the basic secondary amine moiety of moxifloxacin. Structures of the compounds were enlightened by FT-IR, ^{1}H NMR, ^{13}C NMR and HRMS data besides elemental analysis. Compounds were initially tested *in vitro* for their anti-mycobacterial activity against *Mycobacterium tuberculosis* H37Rv using microplate alamar blue assay. Minimal inhibitory concentration (MIC) values of all compounds were found between > 25.00–0.39 µg/mL while compounds **1**, **2** and **13** revealed an outstanding activity against *M. tuberculosis* H37Rv with MIC values of 0.39 µg/mL. Activities of compounds **1–21** against to a number of Gram-positive and Gram-negative bacteria and fast growing mycobacterium strain were also investigated by agar well diffusion and microdilution methods. According to antimicrobial activity results, compound **13** was found the most potent derivative with a IC_{50} value of < 1.23 µg/mL against *Staphylococcus aureus* and clinical strain of methicillin-resistant clinical strain of *S. aureus*.

1. Introduction

Bacterial infections and antimicrobial agent resistance are seen as a public health problem worldwide [1]. Fluoroquinolones (FQs) are broad-spectrum antibacterial agents that are used for the treatment of various bacterial infections such as urinary tract infections, sexually transmitted diseases, respiratory tract infections and etc. over the past three decades [2–5]. They are also recommended as second line anti-tuberculosis agents by World Health Organization (WHO) [6]. Structures of certain FQs in clinical use are presented in Fig. 1.

Tuberculosis (TB), caused by pathogen *Mycobacterium tuberculosis*, is an emerging global health threat and the ninth leading cause of death worldwide. According to the report by WHO published in 2017, TB is responsible for approximately 1.3 million deaths annually among HIV-negative people and additional 374,000 deaths among HIV-positive people [7]. The standard treatment regimen for TB includes a combination of rifampin, isoniazid, pyrazinamide and ethambutol. Bedaquiline and delemanid have been approved by the authorities for the treatment of multidrug resistant / rifampicin resistant tuberculosis (MDR/RR TB) [8]. MDR/RR TB is a growing problem and affects thousands of people's lives. Nearly 600,000 people were diagnosed with and 240,000 people died of MDR/RR TB in 2016 [7]. FQs show successful profile for drug-susceptible TB treatment and drug-resistant TB treatment. Especially moxifloxacin and gatifloxacin, which are fourth generation FQs, have shown promising anti-mycobacterial activity in clinical studies [9]. It is important to note that phase IV clinical trials on moxifloxacin for TB treatment are currently being conducted [10].

FQs are known as DNA topoisomerase inhibitors. DNA

Abbreviations: FQ, fluoroquinolone; TB, tuberculosis; WHO, World Health Organization; MDR/RR TB, multi-drug resistant/rifampicin resistant tuberculosis; MABA, microplate alamar blue assay

☆ This research was presented at the XXIV European Federation for Medicinal Chemistry International Symposium on Medicinal Chemistry (Manchester, UK) on August 28–September 1, 2016 and at the VII EFMC International Symposium on Advances in Synthetic and Medicinal Chemistry (Vienna, Austria) on August 27–31, 2017.

* Corresponding author.

E-mail address: ikucukguzel@marmara.edu.tr (İ. Küçükgüzel).

[1] Present address: Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Altınbaş University, Bakırköy 34145, İstanbul, Turkey.

https://doi.org/10.1016/j.bioorg.2019.102965
Received 10 November 2018; Received in revised form 27 April 2019; Accepted 29 April 2019

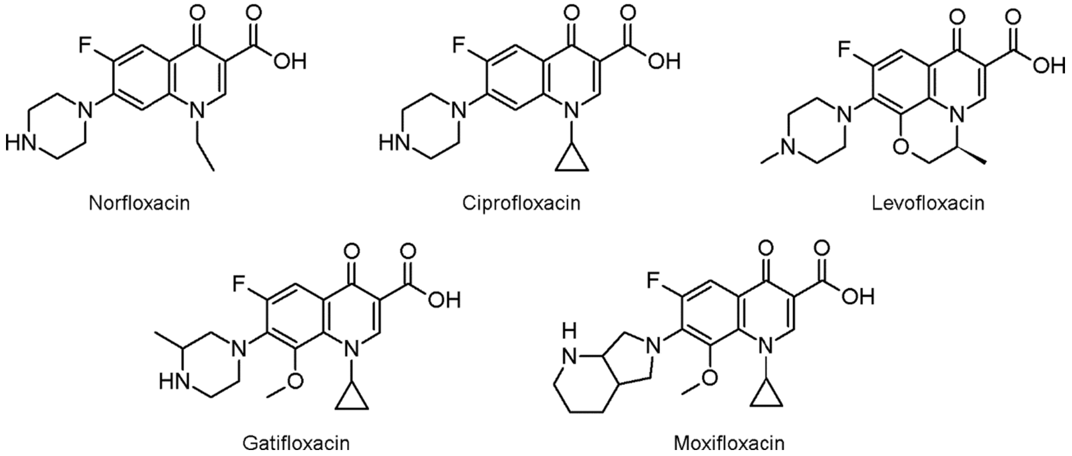


Fig. 1. Chemical structures of the FQ derivatives.

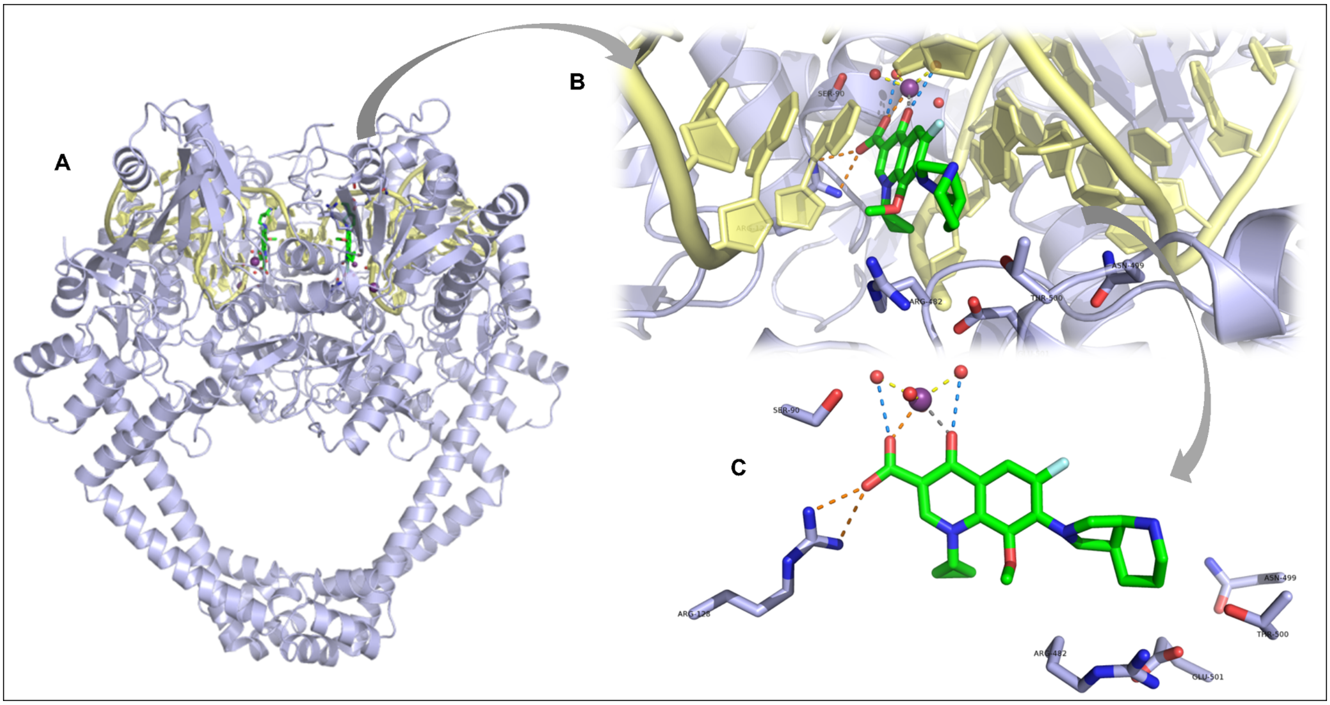


Fig. 2. (A) Crystal structure of *M. tuberculosis* DNA gyrase in complex with moxifloxacin (PDB ID: 5BTA). (B) Moxifloxacin in the binding site of *M. tuberculosis* DNA gyrase. (C) Interactions between moxifloxacin and *M. tuberculosis* DNA gyrase.

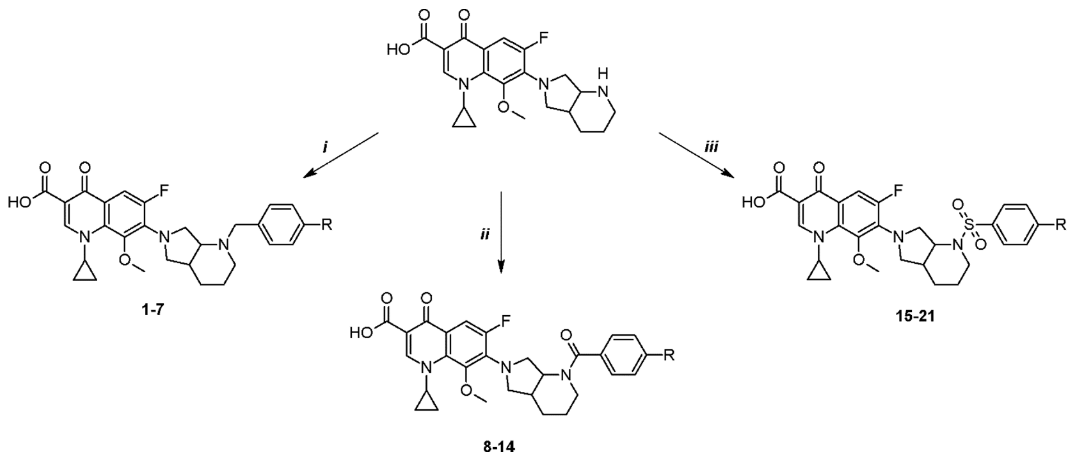
topoisomerases can be divided into two main types as topo I and topo II. Topoisomerase II enzymes are also classified as DNA gyrase and topo IV. Especially topo II enzymes are attractive targets for the development of antibacterial agents. FQs inhibit bacterial DNA gyrase enzymes which are closely related to topo IV [11,12]. Although several crystal structures of DNA topoisomerases obtained from different microorganisms were reported previously, the X-ray structure of *M. tuberculosis* DNA gyrase, co-crystallized with DNA and several ligands (moxifloxacin, gatifloxacin, levofloxacin, ciprofloxacin) has lately been published in 2016 by Blower et al (Fig. 2) [13]. This breakthrough development has opened the way to the discovery of new anti-tuberculosis agents which can specifically inhibit *M. tuberculosis* DNA gyrase [14].

Modification of already known FQ derivatives is a commonly used approach in drug discovery. Several reports highlight that increasing the lipophilicity of FQs can improve their activity [15–17]. In the literature, there are miscellaneous examples of the synthesis and activity evaluation of modified FQ derivatives [18–40]. Moreover, benzenesulfonyl-substituted FQs have recently been reported as a new class of FQs [41]. In the light of the literature and our recent report [42] that reveals the efficacy of modified FQ analogues, some novel FQ derivatives were designed and synthesized. Possible mode of action of the synthesized compounds was studied through enzymatic studies and docking calculations, as well.

2. Results and discussion

2.1. Chemistry

Novel moxifloxacin-derived compounds were designed and synthesized in this study. The synthetic strategy of the targeted compounds is described in the Scheme 1. To obtain compounds **1–7**, moxifloxacin was reacted with several substituted benzyl chlorides in tetrahydrofuran containing sodium hydroxide. For the synthesis of compounds **8–14**, moxifloxacin was reacted with the corresponding benzoyl chlorides in dimetylformamide in the presence of anhydrous K_2CO_3. Compounds



Scheme 1. Synthesis of the targeted derivatives. *i.* (Non) Substituted benzyl chloride, 5 N NaOH, THF. *ii.* (Non) Substituted benzoyl chloride, K$_2$CO$_3$, DMF. *iii.* (Non) Substituted benzenesulfonyl chloride, TEA, ACN.

15–21 were synthesized by the reaction of aryl sulfonyl chlorides with moxifloxacin in acetonitrile containing triethylamine.

Purity of the synthesized compounds was checked by using TLC and HPLC. The structures of the final compounds were confirmed by FTIR and ^{1}H NMR, ^{13}C NMR besides elemental analysis. Molecular weights and empirical formulae of compounds **1–21** were also determined by high resolution mass spectrometry (HRMS). According to ESI-HRMS findings, calculated and measured *m/z* values were found within satisfactory range.

Well documented spectral data is presented in Supplementary Material.

2.2. Biological studies

2.2.1. Anti-mycobacterial activity studies

MIC calculation of compounds **1–21** against *M. tuberculosis* were conducted with microplate alamar blue assay (MABA). Determination of cytotoxicity in RAW 264.7 cell culture and also *M. tuberculosis* DNA gyrase supercoiling assay for the determination of anti-tuberculosis activity were performed. Results are presented in Table 1.

According to the MABA results, introduction of alkyl moiety to moxifloxacin resulted in better anti-mycobacterial activity compared to the introduction of acyl or sulfonyl moieties. Compounds **1–7** possessed of MIC values at 0.39–3.13 μg/mL, compounds **8–14** possessed of MIC values at 0.39–25.00 μg/mL and compounds **15–21** possessed of MIC values at 25.00 – > 25.00 μg/mL. It is obvious that introduction of sulfonyl group to secondary amine's nitrogen does not enhance anti-tuberculosis activity. Compounds with acyl group substitution show moderate anti-tuberculosis activity.

Cytotoxicity of the compounds possessing MIC values less than 25.00 μg/mL against *M. tuberculosis* was evaluated at 25.00 μg/mL concentration. Percentage viability of RAW 264.7 cells which were exposed to 25.00 μg/mL concentrations of compounds were found between 85.20 and 60.40%. Cytotoxicity results are presented in the Table 1. Percentage viability of RAW 264.7 cells which were exposed to 25.00 μg/mL moxifloxacin was found 73.20%. Compared to moxifloxacin, compounds **1**, **2**, **4–6**, **9**, **12**, **14** were found as favorably less toxic to RAW 264.7 cell lines.

Related to the mycobacterial activity studies, compounds **1–3**, **5–6** and **13** possessing MIC values under 2.00 μg/mL were selected for *M. tuberculosis* supercoiling assay since they exhibited better anti-tuberculosis activity. DNA gyrase supercoiling assay was also applied to the reference drug moxifloxacin. Based on the supercoiling assay, IC$_{50}$ values of compounds **1–3**, **5–6** and **13** were found between 4.60 and 12.52 μM whereas IC$_{50}$ value of moxifloxacin was found 4.20 μM.

Compounds **1**, **2** and **13** were identified as the most active moxifloxacin derivatives among the synthesized compounds, by their MIC value of 0.39 μg/mL against *M. tuberculosis*. Of these selected derivatives, IC$_{50}$ value of compound **1** was 4.60 ± 0.12 μM on *M. tuberculosis* DNA supercoiling assay while viability percentage was found 74.24% on RAW 264.7 cells exposed to this compound. Further to the inhibition value which was found less than 30.00%, it can be asserted that compound **1** showed acceptable toxicity to RAW 264.7 cells. Besides, as seen in Table 1, it is observed that compound **1** is less toxic than moxifloxacin and supercoiling assay result of compound **1** reveals an IC$_{50}$ value comparable to moxifloxacin (4.20 ± 0.23 μM). In addition, compounds **2** and **13** which possess the same MIC value with compound **1**, reveal IC$_{50}$ values of 6.10 ± 0.32 and 8.69 ± 0.56 μM on supercoiling assay, respectively.

In this study, *Mycobacterium smegmatis* which is a rapidly growing mycobacterium was considered as a useful surrogate model for tuberculosis research. Compounds **1–21** were tested against *M. smegmatis* with agar dilution and microdilution assays. It was noteworthy that, the compounds which were found to be active against *M. smegmatis*, also possessed good inhibitory properties on *M. tuberculosis* (Tables 2 and 3).

Log P values of compounds were calculated theoretically and presented in Table 1. It should be stated that no correlation was determined between Log P values and anti-tuberculosis activity of the target compounds.

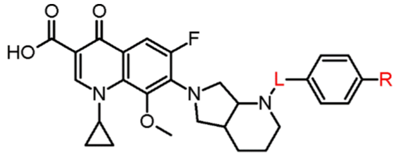
2.2.2. Antimicrobial activity studies

Antimicrobial activity of compounds **1–21** was first investigated by agar well diffusion method against to Gram-positive/Gram-negative bacteria, a mycobacterial strain and two yeast-like fungi (Table 2). MIC values of the compounds were investigated after the determination of the quantitative values of the compounds' antibacterial activity (Table 3).

Compounds **1–7**, in which alkyl moieties were substituted to moxifloxacin, showed significant antibacterial activity against both Gram-positive and Gram-negative species. Compounds bearing aroyl (**8–14**) or arylsulfonyl (**15–21**) moieties showed significant antibacterial activity against Gram-positive bacteria. It can be asserted that substitution of arylalkyl moiety to moxifloxacin results in broad-spectrum antibacterial activity, while substitution of either aroyl or arylsulfonyl moieties provides selectivity towards Gram-positive bacteria.

Among the synthesized FQ derivatives, compound **13** was found to be the most potent one in accordance with antibacterial activity results. It was shown to be active against both Gram-positive and Gram-negative strains without possessing selectivity. In addition, compound **13** showed a MIC value of < 1.23 μg/mL against *Staphylococcus aureus* and MRSA. Antibacterial activity of compound **13** against *S. aureus* was shown to be superior in comparison with the reference compounds.

Table 1
Anti-tuberculosis activity results of the compounds.



Compound	L	-R	MIC (µg/mL)	MIC (µM)	Cytotoxicity[a] (viability%)	IC$_{50}$ (µM) Supercoiling assay	GScore	Log P[b]
1	-CH$_2$-	H	0.39	0.79	74.24	4.60 ± 0.12	−12.939	3.42
2	-CH$_2$-	Me	0.39	0.77	79.24	6.10 ± 0.32	−12.847	3.88
3	-CH$_2$-	F	1.56	3.06	64.60	9.12 ± 0.41	−13.814	3.47
4	-CH$_2$-	OMe	3.13	6.00	81.05	ND	−13.177	3.33
5	-CH$_2$-	Cl	1.56	2.97	81.04	6.12 ± 0.18	−13.470	4.01
6	-CH$_2$-	NO$_2$	1.56	2.91	75.44	12.52 ± 0.68	−11.874	3.15
7	-CH$_2$-	Br	3.13	5.49	72.17	ND	−13.993	4.19
8	-CO-	H	25.00	49.45	ND	ND	−12.465	1.93
9	-CO-	Me	12.50	24.06	80.20	ND	−10.673	2.39
10	-CO-	F	12.50	23.88	60.40	ND	−11.703	2.15
11	-CO-	OMe	12.50	23.34	72.40	ND	−11.883	2.45
12	-CO-	Cl	6.25	11.57	77.17	ND	−11.666	2.69
13	-CO-	NO$_2$	0.39	0.71	66.39	8.69 ± 0.56	−12.509	1.89
14	-CO-	Br	12.50	21.39	85.20	ND	−12.000	2.87
15	-SO$_2$-	H	> 25.00	> 46.16	ND	ND	−11.259	2.92
16	-SO$_2$-	Me	> 25.00	> 44.99	ND	ND	−8.979	3.38
17	-SO$_2$-	F	25.00	44.68	ND	ND	−12.830	3.24
18	-SO$_2$-	OMe	> 25.00	> 43.73	ND	ND	−12.256	3.44
19	-SO$_2$-	Cl	> 25.00	> 43.40	ND	ND	−10.942	3.78
20	-SO$_2$-	NO$_2$	> 25.00	> 42.62	ND	ND	−5.055	3.12
21	-SO$_2$-	Br	25.00	40.29	ND	ND	−12.964	3.96
Moxifloxacin	-	–	0.18	0.44	73.30	4.20 ± 0.23	−13.569	1.60

[a] Cytotoxicity of the compounds was tested at RAW 264.7 cell line at 25.0 µg/mL. Results were presented as viability% of RAW 264.7 cells.

[b] Calculations were performed using Molinspiration online property calculation toolkit (http://www.molinspiration.com).

Synthesized compounds did not demonstrate substantive activity against *P. aeruginosa*. Compound **4** revealed the best anti-pseudomonal activity with the MIC value of 85.16 µg/mL. It was also determined that compounds **1**–**21** did not have inhibitory activity against yeast-like fungi (Table 2). It should be mentioned that the designed compounds did not show significant antibacterial activity against *Lactobacillus acidophilus*. *L. acidophilus* already presents in human microbiota and it has an important role in fermentation, especially in the dairy industry [43]. Less antibacterial activity of the compounds against *L. acidophilus* could be evaluated as a favorable effect, as this species is not a pathogenic strain.

Synthesized compounds were tested against *Bacillus cereus* strain, which is a spore-producing bacterium and one of the food-borne infectious agents. MIC values against *B. cereus* were found between < 1.23 and 18.00 µg/mL.

2.3. *In silico docking studies*

FQs are known to inhibit bacterial topoisomerase II enzymes [11,12]. The X-ray structure of *M. tuberculosis* DNA gyrase co-crystallized with DNA and moxifloxacin was published in 2016 [13].

To gain more molecular insight into the activity of the compounds, docking calculations were carried out. Docking studies with *M. tuberculosis* DNA gyrase in addition to *S. aureus* DNA gyrase were conducted to estimate the possible inhibitory activities of the designed compounds. Ligands were prepared as (1*S*,6*S*)-configured stereoisomers of 7-[2,8-diazabicyclo[4.3.0]nonan-8-yl moiety since 1*S*,6*S* configuration is present in the 3D structure of the starting compound moxifloxacin. DNA, Mg^{2+} and conserved water molecules were kept during the calculations.

GScores which were obtained from the docking studies of the compounds **1**–**21** and moxifloxacin with *M. tuberculosis* DNA gyrase and *S. aureus* DNA gyrase by using Schrödinger Glide-SP Flexible Docking are presented in Supplementary Material.

As a preliminary SAR finding, it can be said that compounds **1**–**7**, which showed better anti-tuberculosis activity compared to other series **8**–**21**, demonstrated higher GScores. There was no similar correlation with *S. aureus*, as most of the synthesized compounds were active against this strain regardless of their GScores.

According to MIC values and supercoiling assay results, compound **1** was demonstrated as the most active compound against *M. tuberculosis*. Fig. 3 shows the model conformation of compound **1** in the binding site of *M. tuberculosis* DNA gyrase.

Owing to the docking studies, carboxylate group of compound **1** and Arg128 make salt bridge interaction. Carboxylate group also makes attractive charge interaction with Mg^{2+}. As a metal acceptor, oxygen atom of quinolone ring interacts with Mg^{2+} as well. Hydrogen bonds between the two of the conserved water molecules and compound **1** were detected. Van der Waals contacts between Ser90 and compound **1** contribute to the binding. Conventional H bond, alkyl, pi-pi stacking and halogen interactions between compound **1** and DNA helix were observed additionally.

Asn499Lys, Thr500Asn and Glu501Val mutations are found in quinolone-resistance determining region [13]. In addition to docking studies with wild-type *M. tuberculosis* DNA Gyrase; whether compounds make significant interaction with any of these amino acid residues, docking was performed with mutated *M. tuberculosis* DNA Gyrase as well. Mutant form of *M. tuberculosis* DNA Gyrase was prepared by using MOE (Chemical Computing Group) rotamer explorer. However, no special interaction between mutated amino acid residues and compounds was observed through the docking studies of synthesized compounds into mutated proteins.

Compound **13** was shown as the most active compound against *S. aureus*. Fig. 4 represents possible binding conformation of compound **13**

Table 2
Screening for antimicrobial activity of the compounds using agar well diffusion method.

Compounds	Microorganisms and Inhibition Zone Values (mm)												
	Gram-Negative Bacteria				Gram-Positive Bacteria						Mycobacterial strain	Yeast-Like Fungi	
	Ec.	*Yp.*	*Kp.*	*Pa.*	*Sa.*	MRSA	*Ef.*	*Lm.*	*La.*	*Bc.*	*Ms.*	*Ca.*	*Sc.*
1	22	16	15	9	25	22	19	20	12	18	25	7	–
2	22	12	15	–	24	19	16	16	13	17	35	6	–
3	22	20	20	8	28	24	22	20	12	19	35	–	6
4	22	20	21	17	28	25	25	18	15	20	35	–	7
5	20	18	18	9	28	22	19	17	–	18	30	–	–
6	22	18	20	6	30	24	22	18	16	19	35	6	10
7	20	17	18	–	24	20	19	16	6	16	35	–	6
8	15	10	6	–	32	24	24	20	8	22	25	–	–
9	12	6	6	–	28	20	20	20	10	20	25	7	–
10	12	9	8	–	30	24	22	16	–	20	25	–	–
11	12	6	6	–	28	22	22	20	–	21	20	7	–
12	12	9	6	–	25	20	18	15	6	18	25	–	–
13	26	21	20	12	30	20	22	15	15	22	30	–	–
14	12	6	6	–	25	18	18	16	–	17	20	–	–
15	15	12	6	8	24	20	20	14	–	18	14	–	6
16	16	6	6	–	20	16	15	12	–	15	12	–	–
17	18	16	15	8	24	21	19	14	–	16	20	–	6
18	12	–	6	–	21	18	15	14	–	15	10	–	–
19	15	14	14	8	20	18	14	10	–	15	15	–	6
20	18	8	6	–	20	18	15	15	–	15	10	–	–
21	20	15	18	12	26	22	18	16	–	16	35	–	6
NFX	32	30	30	35	28	30	30	25	8	28	> 40	–	–
CFX	35	32	32	38	30	30	30	27	22	28	> 40	–	–
CFX.HCl	35	35	35	40	35	30	32	30	26	30	> 40	–	–
MFX.HCl	33	30	30	35	35	35	35	30	41	30	> 40	–	–

NFX, Norfloxacin; CFX, Ciprofloxacin; CFX.HCl, Ciprofloxacin hydrochloride; MFX.HCl, Moxifloxacin hydrochloride; *Ec., Escherichia coli* ATCC 25922*; Yp., Yersinia pseudotuberculosis* ATCC 911*; Kp., Klebsiella pneumoniae* ATCC13883*, Pa., Pseudomonas aeruginosa* ATCC 43288*; Sa. Staphylococcus aureus* ATCC 25923*; MRSA, Methicillin-resistant clinical strain of Staphylococcus aureus; Ef., Enterococcus faecalis* ATCC 29212*, Lm., Listeria monocytogenes* ATCC43251*; La., Lactobacillus acidophilus* RSKK 06029*; Bc., Bacillus cereus* Roma 709*; Ms., Mycobacterium smegmatis* ATCC607*; Ca., Candida albicans* ATCC 60193 *and Sc., Saccharomyces cerevisiae* RSKK 251, (–): no activity.

in *S. aureus* DNA gyrase binding site. Arg122 interacts with carboxylate group of compound **13**. Carboxylate group also interacts with Mg^{2+}. Oxygen atom of quinolone ring makes a metal acceptor interaction with Mg^{+2} ion as well. Oxygen atom of quinolone ring and carboxylate group make hydrogen bond interactions with conserved water molecules. Van der Waals contacts were observed between Lys417, Asn476 and Glu477. Carbon-hydrogen bond interaction was detected between methoxy group of compound **13** and Gly459. Contribution of alkyl, pi-alkyl and van der Waals contacts were observed additionally.

3. Conclusions

Novel FQ derivatives were designed as potential anti-tuberculosis and antibacterial agents. Targeted compounds were synthesized by the introduction of arylalkyl, aroyl or arylsulfonyl moieties to basic secondary amine group of moxifloxacin.

Anti-tuberculosis activity of the designed compounds **1–21** was evaluated *in vitro*. MABA method was used to determine the MIC values of the synthesized compounds against *M. tuberculosis* H37Rv. MIC values of all compounds were found between > 25 and 0.39 µg/mL. Compounds **1**, **2** and **13** expressed an outstanding activity against *M. tuberculosis* H37Rv with the MIC value of 0.39 µg/mL. Cytotoxicity of the compounds with the MIC values which were found less than 25.00 µg/mL against *M. tuberculosis* was tested. Viability% of RAW 264.7 cell lines were found between 85.20 and 60.40% whereas some of the designed compounds were less toxic than moxifloxacin to RAW 264.7 cells. DNA gyrase supercoiling assay was applied to compounds **1–3**, **5–6** and **13** as well as reference drug moxifloxacin and IC_{50} values of these selected compounds were found between 4.60 and 12.52 µM.

Antibacterial activity of the designed compounds against Gram-positive, Gram-negative and acidoresistant bacterial strains and yeast-like fungi was evaluated with zone value measurement and micro-dilution assays. Compounds **1–7** were found to be active against both Gram-positive and Gram-negative strains. Compounds **8–21** were found to be selectively active against Gram-positive bacteria. Remarkably, compound **13** showed a MIC value of < 1.23 µg/mL against *S. aureus* and MRSA. Thus, this compound can be considered as a useful antibacterial agent against antibiotic resistant *S. aureus*. Noteworthy, compounds which were active against *M. smegmatis* also showed good activities towards *M. tuberculosis*.

4. Experimental protocols

4.1. Chemistry

Apart from moxifloxacin.HCl (CAS number: 186826-86-8), all starting materials, reagents and solvents were of high-grade commercial products purchased from Aldrich or Merck. Purity determination of the compounds were performed by TLC on Merck silica gel 60 F_{254} plates. For the purification of benzoyl substituted FQs **8–14**, silica gel column chromatography was utilized using dichloromethane: methanol (90:10, v/v) as eluent and Merck silica gel 60 F_{254} (0.063–0.200 mm) as adsorbent. Melting points (°C, uncorrected) were determined using Schmelzpunktbestimmer SMP II basic model melting point apparatus. Elemental analysis was performed by using LECO CHNS-932 instrument and the results were found to be in consistent with the assigned structures. Infrared spectra were recorded on a Shimadzu FTIR 8400S and data are expressed in wavenumbers, ν (cm^{-1}). ^{1}H- and ^{13}C NMR (decoupled) spectra were recorded on Brüker AVANCE-DPX 400 at 300 MHz, the chemical shifts were expressed in δ (ppm) downfield from tetramethylsilane (TMS) using DMSO-d_6 or $CDCl_3$ as solvent. High resolution mass spectra were acquired by using Jeol JMS700 instrument.

Table 3
Screening for antimicrobial activity of the compounds using microdilution method.

Compounds	Microorganisms and minimal inhibitory concentration (μg/mL)												
	Gram-Negative Bacteria				Gram-Positive Bacteria						Mycobacterial strain	Yeast-Like Fungi	
	Ec.	*Yp.*	*Kp.*	*Pa.*	*Sa.*	MRSA	*Ef.*	*Lm.*	*La.*	*Bc.*	*Ms.*	*Ca.*	*Sc.*
1	2.71	43.36	43.36	346.88	< 2.71	< 2.71	2.71	2.71	138.75	18.00	25.00	346.88	–
2	2.64	84.38	42.19	–	< 2.64	2.64	2.64	2.64	67.50	2.64	2.64	337.50	–
3	2.69	10.74	10.74	343.75	< 2.69	< 2.69	< 2.69	2.69	137.50	2.69	2.69	–	343.75
4	2.66	5.32	5.32	85.16	< 2.66	< 2.66	< 2.66	5.32	170.31	< 2.66	2.66	–	340.63
5	9.57	38.28	38.28	306.25	< 2.39	< 2.39	2.39	2.39	–	2.39	4.79	–	–
6	4.98	19.92	19.92	318.75	< 2.49	< 2.49	< 2.49	2.49	79.69	2.49	2.49	318.75	127.50
7	10.35	41.41	41.41	–	< 2.59	< 2.59	< 2.59	2.59	263.00	2.59	2.59	–	331.25
8	38.28	153.13	306.25	–	4.79	4.79	4.79	4.79	245.00	4.79	9.57	–	–
9	82.81	331.25	331.25	–	< 2.59	< 2.59	< 2.59	< 2.59	132.00	< 2.59	10.35	331.25	–
10	81.25	325.00	325.00	–	10.16	< 2.54	< 2.54	2.54	–	2.54	10.16	–	–
11	89.38	337.50	337.50	–	< 2.64	< 2.64	< 2.64	< 2,64	–	< 2.64	10.55	337.50	–
12	82.81	165.63	331.25	–	< 2.59	< 2.59	< 2.59	< 2.59	256.00	< 2.59	5.18	–	–
13	1.23	9.86	9.86	315.63	< 1.23	< 1.23	1.23	1.23	157.81	< 1.23	1.23	–	–
14	79.69	318.75	318.75	–	< 2.49	< 2.49	< 2.49	2.49	–	2.49	4.98	–	–
15	82.81	165.63	265	331.25	< 2.59	< 2.59	< 2.59	5.18	–	2.59	20.70	–	331.25
16	80.47	321.88	321.88	–	< 2.51	2.516	2.51	5.03	–	2.51	20.12	–	–
17	40.63	81.25	81.25	325.00	< 2.54	< 2.54	< 2.54	2.54	–	2.54	20.31	–	325.00
18	154.69	–	309.38	–	< 2.42	< 2.42	2.42	2.42	–	2.42	9.67	–	–
19	84.38	84.38	84.38	337.50	< 2.64	< 2.64	10.55	21.09	–	5.27	42.19	–	337.50
20	82.03	328.13	328.13	–	< 2.56	2.56	2.56	5.56	–	2.56	20.51	–	–
21	19.92	39.84	19.92	159.38	< 2.49	< 2.49	2.49	2.49	–	2.49	4.98	–	318.75
NFX	< 0.60	0.60	1.32	5.27	1.32	0.60	2.64	2.64	675.00	2.64	5.27	–	–
CFX	< 0.60	0.60	0.60	1.31	1.31	1.31	2.61	1.31	83.59	1.31	< 0.60	–	–
CFX.HCl	< 0.60	< 0.60	< 0.60	1.26	2.51	1.26	1.26	1.26	80.47	0.60	< 0.60	–	–
MFX.HCl	< 0.60	< 0.60	< 0.60	5.08	1.27	< 0.60	< 0.60	< 0.60	40.63	< 0.60	< 0.60	–	–

NFX, Norfloxacin; CFX, Ciprofloxacin; CFX.HCl, Ciprofloxacin hydrochloride; MFX.HCl, Moxifloxacin hydrochloride; *Ec., Escherichia coli* ATCC 25922; Yp., *Yersinia pseudotuberculosis* ATCC 911; Kp., *Klebsiella pneumoniae* ATCC13883, Pa., *Pseudomonas aeruginosa* ATCC 43288; *Sa. Staphylococcus aureus* ATCC 25923; MRSA, *Methicillin-resistant clinical strain of Staphylococcus aureus;* Ef., *Enterococcus faecalis* ATCC 29212, Lm., *Listeria monocytogenes* ATCC43251; La., *Lactobacillus acidophilus* RSKK 06029; Bc., *Bacillus cereus Roma 709; Ms., Mycobacterium smegmatis* ATCC607; (–): no activity.

*4.1.1. General procedure for the synthesis of compounds **1–7***

Moxifloxacin.HCl (1.50 mmol) was stirred in tetrahydrofuran (30 mL) in the presence of aq. 5 N NaOH (0.6 mL). Appropriate benzyl chloride (2.25 mmol) was added to the medium gradually afterwards. Reaction was maintained at 50 °C for 24 h. When moxifloxacin spot disappeared from TLC sheet, reaction medium was treated with ice-cold water. Solution was neutralized with 10% acetic acid. The precipitate was then filtered and washed with water. Crude product was recrystallized from EtOH or acetone.

4.1.1.1. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-benzyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid

1. (CAS Number: 2254156-04-0) M.p. 150 °C. Elemental analysis calculated for $C_{28}H_{30}FN_3O_4$; C 68.42, H 6.15, N 8.55; found C 68.12, H 5.67, N 8.51. IR (cm^{-1}): 1732, 1622 (C═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 15.11 (s, 1H, COO**H**), 8.80 (s, 1H, *C*$_2$-**H**), 7.93 (m, 1H, Ar-**H'**), 7.83 (d, 1H, C$_5$-**H**, *J* = 13.8 Hz), 7.62–7.29 (m, 3H, Ar-**H'**), 7.32–3-7.28 (m, 1H), 3.59 (s, 3H, OC**H$_3$**), 4.08–0.82 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{30}FN_3O_4$ [M + Na]$^+$, 514.2118, found 514.2124.

4.1.1.2. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-methylphenyl)methyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **2**. M.p. 138 °C. Elemental analysis calculated for

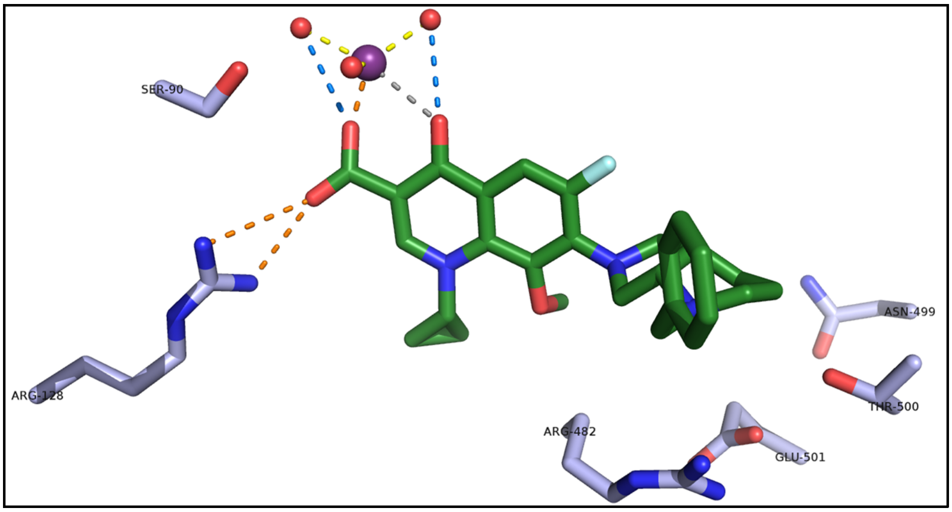


Fig. 3. Compound **1** in *M. tuberculosis* DNA gyrase binding site. Amino acids which can be mutated or which interact with the ligand are shown in the figure. Hydrogen bonds are depicted as blue dashes. Attractive charge and salt bridge interactions are shown as orange dashes. Metal acceptor interactions are shown as gray dashes. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

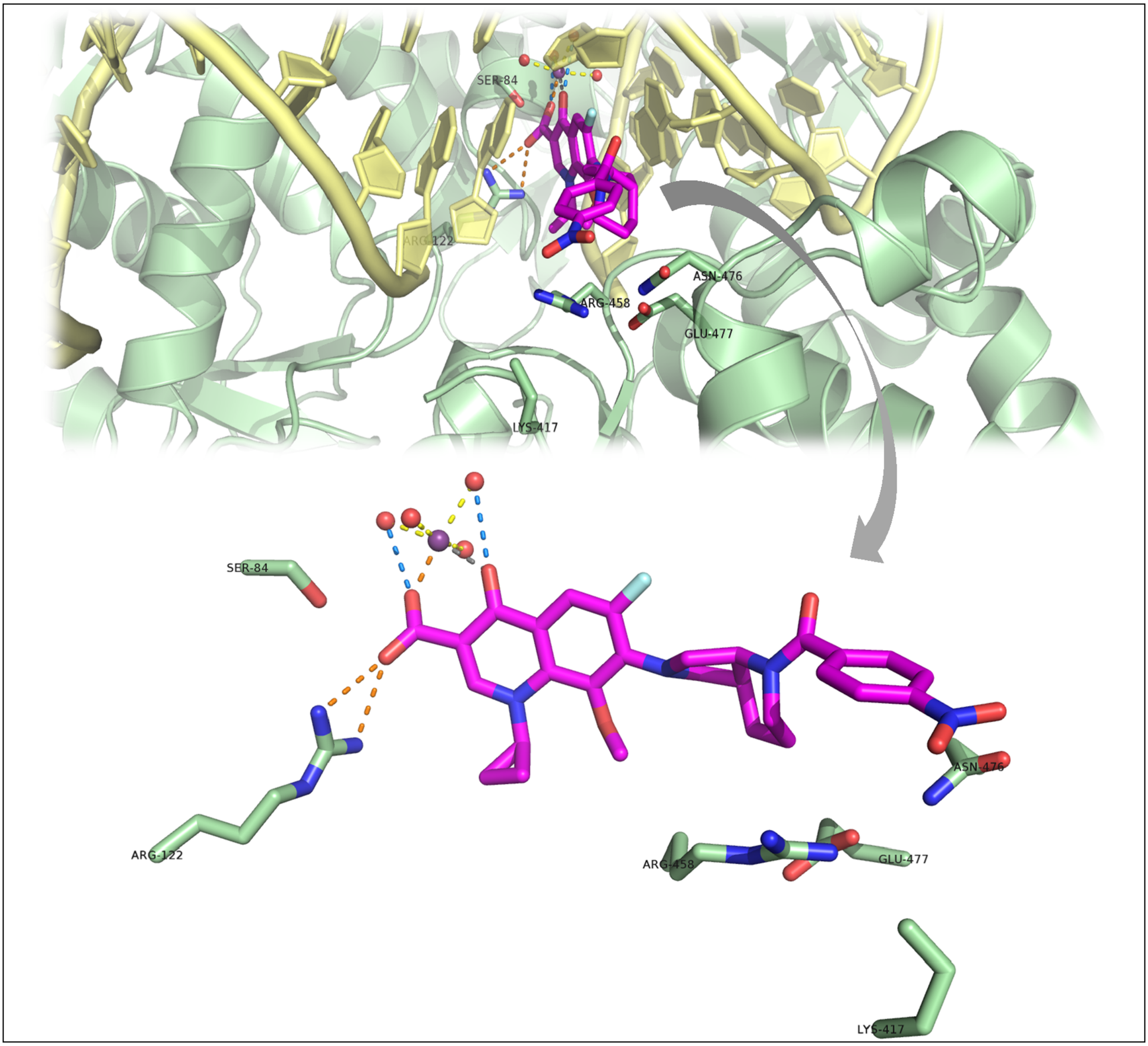


Fig. 4. Compound **13** in *S. aureus* DNA gyrase binding site. Amino acids which can be mutated or which interact with the ligand are presented. Hydrogen bonds are depicted as blue dashes. Attractive charge and salt bridge interactions are shown as orange dashes. Metal acceptor interactions are shown as gray dashes. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

$C_{29}H_{32}FN_3O_4$; C 68.89, H 6.38, N 8.31; found C 69.23, H 6.07, N 8.06. IR (cm^{-1}): 1722, 1620 (C=O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 15.14 (s, 1H, COO**H**), 8.77 (s, 1H, *C*$_2$-**H**), 7.80 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.14 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, *J* = 7.8 Hz), 7.05 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, *J* = 7.8 Hz), 3.59 (s, 3H, OC$\mathbf{H_3}$), 4.03–0.87 (aliphatic C–**H**). ^{13}C NMR δ ppm (75 MHz, $CDCl_3$): 176.69 (quinolone $\mathbf{C_4}$ = O), 167.19 (–**C**OOH), 153.68 ($\mathbf{C_6}$, *J* = 249.8 Hz), 149.44, 140.52, 140.42, 137.63 (quinolone $\mathbf{C_7}$, *J* = 11.3 Hz), 136.50, 135.93, 134.48, 128.88, 128.40, 117.88, 107.48, 107.55, 107.86, 62.20, 60.86, 59.67, 53.76, 53.27, 50.39, 40.48, 37.62, 23.78, 22.35, 21.03 ($\mathbf{C}H_3$), 9.86, 9.09. HRMS (ESI$^+$) *m/z* calculated for $C_{29}H_{31}FN_3O_4$ [M + Na]$^+$, 528.2280, found 528.2275.

*4.1.1.3. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-fluorophenyl) methyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **3***. M.p. 169 °C. Elemental analysis calculated for $C_{28}H_{29}F_2N_3O_4$; C 66.00, H 5.74, N 8.25; found C 65.81, H 5.64, N 8.12. IR (cm^{-1}): 1716, 1624 (C=O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 15.11 (s, 1H, COO**H**), 8.77 (s, 1H, *C*$_2$-**H**), 7.80 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.24–7.20 (m, 2H, Ar-$\mathbf{H_{2'6'}}$), 6.97–6.91 (m, 2H, Ar-$\mathbf{H_{3'5'}}$), 3.59 (s, 3H, OC$\mathbf{H_3}$), 4.00–0.88 (aliphatic C–**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{29}F_2N_3O_4$ [M + H]$^+$, 510.2199, found 510.2199.

*4.1.1.4. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-methoxyphenyl) methyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **4***. M.p. 164 °C. Elemental analysis calculated for $C_{29}H_{32}FN_3O_5$; C 66.78, H 6.18, N 8.06; found C 66.38, H 6.06, N 8.00. IR (cm^{-1}): 1620 (C=O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 15.13 (s, 1H, COO**H**), 8.77 (s, 1H, *C*$_2$-**H**), 7.80 (d, 1H, C$_5$-**H**, *J* = 13.8 Hz), 7.16 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, *J* = 8.7 Hz), 6.78 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, *J* = 8.7 Hz), 3.75 (s, 3H, Ar-OC$\mathbf{H_3}$), 3.59 (s, 3H, OC$\mathbf{H_3}$), 4.03–0.91 (aliphatic C–**H**). ^{13}C NMR δ ppm (75 MHz, $CDCl_3$): 176.71, 167.19 (–**C**OOH), 158.60 (phenyl $\mathbf{C_4}$), 153.69 (quinolone $\mathbf{C_6}$, *J* = 249.0 Hz), 149.45, 140.51, 140.42, 137.63 (quinolone $\mathbf{C_7}$, *J* = 10.5 Hz), 134.49, 130.92, 129.57, 117.90, 113.57, 107.87, 107.55, 107.48, 62.12, 60.86, 59.34, 55.20, 53.83, 53.15, 50.21, 40.49, 37.58, 23.82, 22.38, 9.11, 9.83. HRMS

(ESI$^+$) *m/z* calculated for $C_{29}H_{32}FN_3O_5$ $[M+H]^+$, 522.2399, found 522.2400.

4.1.1.5. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-chlorophenyl)methyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***5***. M.p. 119 °C. Elemental analysis calculated for $C_{28}H_{29}ClFN_3O_4$; C 63.94, H 5.56, N 7.99; found C 63.18, H 5.62, N 7.90. IR (cm^{-1}): 1726, 1622 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 15.11 (s, 1H, COO**H**), 8.77 (s, 1H, C_2-**H**), 7.82 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.26–7.20 (m, 4H, Ar-**H**'), 3.57 (s, 3H, OC$\mathbf{H_3}$), 4.00–0.87 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{29}ClFN_3O_4$ $[M+H]^+$, 526.1903, found 526.1903.

4.1.1.6. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-nitrophenyl)methyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***6***. M.p. 115 °C. Elemental analysis calculated for $C_{28}H_{29}FN_4O_4$; C 62.68, H 5.45, N 10.44; found C 62.41, H 5.35, N 10.16. IR (cm^{-1}): 1726, 1620 (C=O). ^{1}H NMR δ ppm (300 MHz, DMSO-d_6): 15.00 (s, 1H, COO**H**), 8.76 (s, 1H, C_2-**H**), 8.25–7.54 (m, 5H, Ar-**H**', C$_5$-**H**), 3.62 (s, 3H, OC$\mathbf{H_3}$), 4.19–1.00 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{29}FN_4O_6$ $[M+Na]^+$, 559.1968, found 559.1977.

4.1.1.7. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-bromophenyl)methyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***7***. M.p. 118 °C. Elemental analysis calculated for $C_{28}H_{29}BrFN_3O_4$; C 58.95, H 5.12, N 7.37; found C 59.04, H 5.35, N 6.87. IR (cm^{-1}): 1726, 1620 (C=O). ^{1}H NMR δ ppm (300 MHz, DMSO-d_6): 15.22 (s, 1H, COO**H**), 8.77 (s, 1H, C_2-**H**), 7.81 (d, 1H, C$_5$-**H**, *J* = 14.7 Hz), 7.33 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, *J* = 8.4 Hz), 7.13 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, *J* = 8.4 Hz), 3.58 (s, 3H, OC$\mathbf{H_3}$), 4.13–0.85 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{29}BrN_3O_4$ $[M + Na]^+$, 592.1231, found 592.1231.

4.1.2. General procedure for the synthesis of compounds 8–14

To a solution of moxifloxacin.HCl (1.50 mmol) in DMF (30 mL), K_2CO_3 (3.00 mmol) was added. The mixture was allowed to stir at 60 °C for 2 h. Related benzoyl chloride (2.25 mmol) was then added to the medium dropwise. When the reaction was complete, the mixture was poured into ice-water. The precipitate was filtered and the resultant raw material was purified by column chromatography using DCM: MeOH (90:10, v/v) as mobile phase.

4.1.2.1. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(phenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***8***. M.p. 162 °C. Elemental analysis calculated for $C_{28}H_{28}FN_3O_5$.3/2 H_2O; C 63.15, H 5.87, N 7.89; found C 63.63, H 5.55, N 7.85. IR (cm^{-1}): 3536 (O—H); 1732, 1618 (C=O). ^{1}H NMR δ ppm (300 MHz, DMSO-d_6): 15.14 (s, 1H, COO**H**), 8.66 (s, 1H, C$_2$-**H**), 7.66 (d, 1H, C$_5$-**H**, *J* = 13.8 Hz), 7.73–7.45 (m, 5H, Ar-**H**'), 3.58 (s, 3H, OC$\mathbf{H_3}$), 4.16–0.87 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{28}FN_3O_5$ $[M + Na]^+$, 528.1911, found 528.1920.

4.1.2.2. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-methylphenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***9***. M.p. 148 °C, Elemental analysis calculated for $C_{29}H_{30}FN_3O_5$; C 67.04, H 5.82, N 8.09; found C 66.63, H 6.04, N 8.11. IR (cm^{-1}): 1724, 1616 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 14.97 (s, 1H, COO**H**), 8.79 (s, 1H, C_2-**H**), 7.81 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.37–7.22 (m, 4H, Ar-**H**'), 3.59 (s, 3H, OC$\mathbf{H_3}$), 2.39 (s, 3H, Ar-C$\mathbf{H_3}$), 4.05–0.84 (aliphatic C—**H**). ^{13}C NMR δ ppm (75 MHz, CDCl$_3$): 176.75 (quinolone $\mathbf{C_4}$ = O), 171.95 (amide **C**=O), 166.94 (—**C**OOH), 153.69, (quinolone $\mathbf{C_6}$, *J* = 249.0 Hz), 149.73, 141.15, 140.06, 140.02, 137.10 (quinolone $\mathbf{C_7}$, *J* = 10.5 Hz), 134.39, 133.11, 129.22, 126.85, 118.90, 108.19, 107.87, 107.70, 61.22, 56.56, 56.45, 48.56, 40.41, 35.80, 25.59, 24.58, 21.40, 10.54, 8.53. HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{30}FN_3O_5$ $[M + Na]^+$, 542.2067, found 542.2072.

4.1.2.3. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-fluorophenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***10***. M.p. 115 °C. Elemental analysis calculated for $C_{28}H_{27}F_2N_3O_5$.1/3 H_2O; C 63.51, H 5.27, N 7.94; found C 63.05, H 5.38, N 7.90. IR (cm^{-1}): 1716, 1620 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 14.96 (s, 1H, COO**H**), 8.80 (s, 1H, C_2-**H**), 7.83 (d, 1H, C$_5$-**H**, *J* = 13.8 Hz), 7.48–7.10 (m, 4H, Ar-**H**'), 3.60 (s, 3H, OC$\mathbf{H_3}$), 4.10–0.86 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{27}F_2N_3O_5$ $[M+Na]^+$, 546.1816, found 546.1822.

4.1.2.4. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-methoxyphenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***11***. M.p. 187 °C. Elemental analysis calculated for $C_{29}H_{32}FN_3O_5$; C 65.04, H 5.65, N 7.85; found C 64.68, H 5.37, N 7.85. IR (cm^{-1}): 3473 (O—H); 1724, 1614 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 14.98 (s, 1H, COO**H**), 8.79 (s, 1H, C_2-**H**), 7.81 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.41 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, *J* = 8.4 Hz), 6.93 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, *J* = 8.7 Hz), 3.84 (s, 3H, Ar-OC$\mathbf{H_3}$), 3.59 (s, 3H, OC$\mathbf{H_3}$), 4.09–0.78 (aliphatic C—**H**). ^{13}C NMR δ ppm (75 MHz, CDCl$_3$): 176.77 (quinolone $\mathbf{C_4}$ = O), 171.73 (amide **C**=O), 166.94 (—**C**OOH), 160.88 (phenyl $\mathbf{C_4}$), 153.71, (quinolone $\mathbf{C_6}$, *J* = 249.8 Hz), 149.73, 141.17, 141.08, 137.10 (quinolone $\mathbf{C_7}$, *J* = 11.3 Hz), 137.02, 134.40, 128.86, 128.11, 119.96, 113.87, 108.24, 107.92, 107.75, 61.22, 56.57, 56.47, 55.38, 48.54, 40.39, 35.82, 25.62, 24.62, 10.53, 8.53. HRMS (ESI$^+$) *m/z* calculated for $C_{29}H_{31}FN_3O_6$ $[M+Na]^+$, 536.2191, found 536.2199.

4.1.2.5. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-chlorophenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***12***. M.p. 170 °C. Elemental analysis calculated for $C_{28}H_{27}ClFN_3O_5$.1/2H_2O; C 61.26, H 5.14, N 7.65; found C 61.07, H 5.05, N 7.50. IR (cm^{-1}): 3482 (O—H); 1732, 1614 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 14.95 (s, 1H, COO**H**), 8.79 (s, 1H, C_2-**H**), 7.81 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.44–7.35 (m, 4H, Ar**H**'), 3.59 (s, 3H, OC$\mathbf{H_3}$), 4.08–0.78 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{28}ClFN_3O_5$ $[M+Na]^+$, 562.1521, found 562.1517.

4.1.2.6. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-nitrophenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid ***13***. M.p. 138–142 °C. Elemental analysis calculated for $C_{28}H_{27}FN_4O_5$; C 61.09, H 4.94, N 10.18; found C 61.18, H 5.46, N 10.12. IR (cm^{-1}): 3587 (OH), 1723, 1618 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 15.04 (s, 1H, COO**H**), 8.77 (s, 1H, C_2-**H**), 8.12 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, *J* = 8.7 Hz), 7.80 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.46 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, *J* = 9.0 Hz), 3.60 (s, 3H, OC$\mathbf{H_3}$), 4.04–0.86 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{27}FN_4O_7$ $[M\text{-}CH_3 + H]^+$, 537.1780, found 537.2154.

4.1.2.7. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-bromophenyl)carbonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid 14. M.p. 160–162 °C. Elemental analysis calculated for $C_{28}H_{27}FBrN_3O_5{\cdot}H_2O$; C 55.82, H 4.85, N 6.97; found C 56.09, H 4.44, N 6.98. IR (cm^{-1}): 1726, 1620 (C=O). ^{1}H NMR δ ppm (300 MHz, CDCl$_3$): 15.13 (s, 1H, COO**H**), 8.76 (s, 1H, C_2-**H**), 7.79 (d, 1H, C$_5$-**H**, *J* = 14.1 Hz), 7.24–7.18 (m, 4H, Ar-**H**'), 3.59 (s, 3H, OC$\mathbf{H_3}$), 4.03–0.86 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{27}F_2N_3O_5$ $[M+Na]^+$, 584.1191, found 584.1205.

4.1.3. General procedure for the synthesis of compounds 15–21

To moxifloxacin. HCl (1.50 mmol) and triethlyamine (2.25 mmol) solution in acetonitrile (50 mL), related arylsulfonyl chloride (2.25 mmol) was added. The reaction medium was maintained at room temperature for 4 h. At the end of reaction, ACN was evaporated and the resultant precipitate was washed with water. Final crude product was recrystallized from EtOH.

*4.1.3.1. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-phenylsulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **15***. M.p. 165–168 °C. Elemental analysis calculated for $C_{27}H_{28}FN_3O_6S{\cdot}H_2O$; C 57.95, H 5.40, N 7.51, S 5.73; found C 57.70, H 5.55, N 7.45, S 5.76. IR (cm^{-1}): 1722, 1616 (C═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 8.80 (s, 1H, C_2-**H**), 7.87–7.33 (m, 6H, Ar-**H'**, C_5-**H**), 3.52 (s, 3H, OC$\mathbf{H_3}$), 4.67–0.78 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{27}H_{28}FN_3O_6S$ [M + Na]$^+$, 564.1575, found 564.1581.

*4.1.3.2. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-methylphenyl)sulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **16***. (CAS number: 1253700-48-9) M.p. 209 °C. Elemental analysis calculated for $C_{27}H_{28}FN_3O_6S$; C 60.53, H 5.44, N 7.56, S 5.77; found C 60.49, H 5.52, N 7.75, S 5.72. IR (cm^{-1}): 1737, 1622 (C═O); 1155 (S═O). ^{1}H NMR δ ppm (300 MHz, DMSO-d_6): 15.14 (s, 1H, COO**H**), 8.64 (s, 1H, C_2-**H**), 7.76 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, $J = 8.1$ Hz), 7.65 (d, 1H, C_5-**H**, $J = 13.8$ Hz), 7.43 (d, 2H, Ar- Ar-$\mathbf{H_{3'5'}}$, $J = 8.1$ Hz), 3.49 (s, 3H, Ar-OC$\mathbf{H_3}$), 2,39 (s, 3H, Ar-C$\mathbf{H_3}$). ^{13}C NMR δ ppm (75 MHz, $CDCl_3$): 176.74 (quinolone $\mathbf{C_4}$ = O), 166.89 (—**C**OOH), 153.70 (quinolone $\mathbf{C_6}$, $J = 249.0$ Hz), 149.75, 143.31, 141.23, 141.14, 136.98, 136.89 (quinolone $\mathbf{C_7}$, $J = 11.3$ Hz), 134.30, 129.83, 127.08, 119.02, 108.15, 107.83, 107.73, 61.20 (—O**C**H_3), 55.75, 53.97, 47.95, 40.93, 40.34, 35.55, 24.68, 23.66, 21.50 (—**C**H_3), 10.47, 8.57. HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{30}FN_3O_6S$ [M + H]$^+$, 556.1912, found 556.1936 [44].

*4.1.3.3. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-fluorophenyl)sulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **17***. M.p. 163 °C. Elemental analysis calculated for $C_{27}H_{27}F_2N_3O_6S$; C 57.95, H 4.86, N 7.51, S 5.73; found C 57.22, H 4.92, N 7.67, S 5.50. IR (cm^{-1}): 1726, 1620 (C═O); 1153 (S═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 14.92 (s, 1H, COO**H**), 8.78 (s, 1H, C_2-**H**), 7.90–7.85 (m, 2H, Ar-$\mathbf{H_{2'6'}}$), 7.78 (d, 1H, C_5-**H**, $J = 13.8$ Hz), 7.25–7.19 (m, 2H, Ar-$\mathbf{H_{3'5'}}$), 3.54 (s, 3H, OC$\mathbf{H_3}$), 4.65–0.78 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{27}H_{28}F_2N_3O_6S$ [M + H]$^+$, 560.1661, found 560.1661.

*4.1.3.4. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-methoxyphenyl)sulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **18***. M.p. 206 °C. Elemental analysis calculated for $C_{28}H_{30}FN_3O_7S{\cdot}H_2O$; C 57.04, H 5.47, N 7.13, S 5.44; found C 56.96, H 5.28, N 7.13, S 5.24. IR (cm^{-1}): 1726, 1620 (C═O); 1147 (S═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 14.94 (s, 1H, COO**H**), 8.78 (s, 1H, C_2-**H**), 7.80–7.76 (m, 3H, Ar-$\mathbf{H_{2'6'}}$, C_5-**H**), 6.99 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, $J = 9.0$ Hz), 3.90 (s, 3H, Ar-OC$\mathbf{H_3}$), 3.53 (s, 3H, OC$\mathbf{H_3}$), 4.63–0.79 (aliphatik C—**H**). ^{13}C NMR δ ppm (75 MHz, $CDCl_3$): 176.73 (quinolone $\mathbf{C_4}$ = O), 166.89 (—**C**OOH), 163.00 (phenyl $\mathbf{C_4}$), 153.67 (quinolone $\mathbf{C_6}$, $J = 249.8$ Hz) 149.75, 141.21, 141.12, 136.93 (quinolone $\mathbf{C_7}$, $J = 10.5$ Hz), 134.31, 131.41, 129.17, 118.98, 114.36, 108.13, 107.81, 107.60, 61.19, 55.78, 55.63, 53.93, 47.91, 40.89, 40.35, 35.63, 24.23, 23.69, 10.45, 8.55. HRMS (ESI$^+$) *m/z* calculated for $C_{28}H_{30}FN_3O_7S$ [M + Na]$^+$, 594.1686, found 594.1685.

*4.1.3.5. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-chlorophenyl)sulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **19***. M.p. 206 °C. Elemental analysis calculated for $C_{27}H_{28}FN_3O_6S$; C 56.30, H 4.72, N 7.29, S 5.57; found C 56.08, H 4.72, N 7.35, S 5.35. IR (cm^{-1}): 1726, 1620 (C═O); 1147 (S═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 14.92 (s, 1H, COO**H**), 8.78 (s, 1H, C_2-**H**), 7.81–7.77 (m, 3H, Ar-$\mathbf{H_{2'6'}}$, C_5-**H**), 7.53 (d, 2H, Ar-$\mathbf{H_{3'}H_{5'}}$, $J = 9.0$ Hz), 3.53 (s, 3H, OC$\mathbf{H_3}$), 4.65–0.79 (aliphatik C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{27}H_{27}ClFN_3O_6$ [M + Na]$^+$, 576.1365, found 576.1378.

*4.1.3.6. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-nitrophenyl)sulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **20***. M.p. 199 °C. Elemental analysis calculated for $C_{27}H_{27}FN_4O_8S$; C 55.28, H 4.64, N 9.55, S 5.47; found C 54.53, H 4.56, N 9.58, S 5.44. IR (cm^{-1}): 1726, 1622 (C═O); 1151 (S═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 14.86 (s, 1H, COO**H**), 8.77 (s, 1H, C_2-**H**), 8.39 (d, 2H, Ar-$\mathbf{H_{3'5'}}$, $J = 9.0$ Hz), 8.06 (d, 2H, Ar-$\mathbf{H_{2'6'}}$, $J = 9.3$ Hz), 7.76 (d, 1H, C_5-**H**, $J = 13.8$ Hz), 3.56 (s, 3H, OC$\mathbf{H_3}$), 4.69–0.79 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{27}H_{27}FN_4O_8S$ [M + Na]$^+$, 609.1431, found 609.1430.

*4.1.3.7. 6-Fluoro-8-methoxy-4-oxo-1-cyclopropyl-7-(2-(4-bromophenyl)sulfonyl-2,8-diazabicyclo[4.3.0]nonan-8-yl)-1,4-dihydroquinoline-3-carboxylic acid **21***. M.p. 164 °C. Elemental analysis calculated for $C_{27}H_{27}BrFN_3O_6S{\cdot}H_2O$; C 52.26, H 4.39, N 6.77, S 5.17; found C 52.37, H 4.31, N 6.99, S 4.88. IR (cm^{-1}): 1726, 1620 (C═O). ^{1}H NMR δ ppm (300 MHz, $CDCl_3$): 14.92 (s, 1H, COO**H**), 8.78 (s, 1H, C_2-**H**), 7.80 (d, 1H, C_5-**H**, $J = 13.8$ Hz), 7.74–7.66 (m, 4H, Ar-**H'**), 3.53 (s, 3H, OC$\mathbf{H_3}$), 4.64–0.78 (aliphatic C—**H**). HRMS (ESI$^+$) *m/z* calculated for $C_{27}H_{27}BrFN_3O_6S$ [M + Na]$^+$, 642.0686, found 642.0690.

4.2. Biological studies

4.2.1. Anti-mycobacterial activity

Anti-tuberculosis activity of the designed compounds **1–21** was evaluated *in vitro*. MABA method was used to determine the MIC values of the synthesized compounds against *M. tuberculosis* H37Rv [45,46]. Results are represented in the Table 1. Method is explained in Supplementary Material.

4.2.2. Cytotoxicity

Compounds revealing MIC values less than 25.00 µg/mL were further examined for toxicity in a RAW 264.7 cell line at a concentration of 25.00 µg/mL [47]. Method is commented in Supplementary Material.

4.2.3. DNA supercoiling assay

DNA supercoiling [48] assay was performed using MTB DNA supercoiling assay kits (Inspiralis Ltd., Norwich, UK). Method is presented in Supplementary Material.

4.2.4. Antimicrobial activity

Detailed information can be found in Supplementary Material.

4.2.4.1. Zone inhibition assay. Simple susceptibility screening test using agar well diffusion was used to determine the antibacterial activity [49]. Antimicrobial activity was evaluated by the measurement of the zone inhibition of the tested organism.

4.2.4.2. Minimum inhibitory concentration assay. Dilution susceptibility testing method [50] was used to determine the minimal concentration of the compound needed to inhibit or kill the microorganism.

4.3. Molecular modeling studies

Molecular modeling studies with *M. tuberculosis* DNA gyrase and *S. aureus* DNA gyrase were carried out to simulate the potential inhibition profile of the synthesized compounds. Protein structures of *M. tuberculosis* and *S. aureus* DNA gyrase enzymes were obtained from protein data bank with 5BTA and 5CDQ PDB codes, respectively. During molecular modeling studies, prepared ligand structures were docked into protein binding site at pH: 7.0 ± 1.0 using Schrödinger Glide-SP Flexible Docking procedure. Carboxylic acid and tertiary amine moieties were generated as negatively and positively charged functions, respectively. 7-[2,8-Diazabicyclo[4.3.0]nonan-8-yl moieties of each compound were specially preserved as (1*S*,6*S*)-configured stereoisomers.

Docking results were evaluated according to ligand efficiency results alongside docking scores.

Furthermore; N499K, T500N, E501V mutated proteins *of M. tuberculosis* DNA gyrase were prepared using MOE (Chemical Computing Group) rotamer explorer. Docking calculations were run for mutant proteins. However, no special interaction between mutated amino acid residues and compounds was observed after docking of synthesized compounds into mutated proteins.

Figures were prepared by using PyMOL software.

Acknowledgments

The authors are grateful to Dr. Jürgen Gross from the Institute of Organic Chemistry, University of Heidelberg, for his generous help on obtaining high resolution mass spectra of the compounds. We would like to thank Prof. Dr. habil. Wolfgang Sippl from Institute of Pharmacy, Faculty of Natural Sciences, Martin Luther University, for giving us the opportunity of working in his molecular modeling research laboratory. Moxifloxacin hydrochloride was generously donated by Atabay Pharmaceuticals and Fine Chemicals Inc.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bioorg.2019.102965.

References

[1] F. Prestinaci, P. Pezzotti, A. Pantosti, Antimicrobial resistance: a global multifaceted phenomenon, Pathog. Glob. Health. 109 (7) (2015) 309–318, https://doi.org/10.1179/2047773215Y.0000000030.

[2] J.S. Wolfson, D.C. Hooper, Fluoroquinolone antimicrobial agents, Clin. Microbiol. Rev. 2 (4) (1989) 378–424.

[3] M.I. Andersson, A.P. MacGowan, Development of the quinolones, J. Antimicrob. Chemother. 51 (S1) (2003) 1–11, https://doi.org/10.1093/jac/dkg212.

[4] L.A. Mitscher, Bacterial topoisomerase inhibitors: quinolone and pyridone antibacterial agents, Chem. Rev. 105 (2005) 559–592.

[5] G.S. Bisacchi, Origins of the quinolone class of antibacterials: an expanded "Discovery Story", J. Med. Chem. 58 (12) (2015) 4874–4882, https://doi.org/10.1021/jm501881c.

[6] J. Crofton, P. Chaulet, D. Maher (contributions from J. Grosset, W. Harris, N. Horne, M. Iseman & B. Watt), Guidelines for the management of drug-resistant tuberculosis. Geneva: World Health Organization, 1997.

[7] Global Tuberculosis Report. World Health Organization, Geneva, 2017.

[8] Y. Fan, J. Wu, X. Cheng, F. Zhang, L. Feng, Fluoroquinolone derivatives and their anti-tubercular activities, Eur. J. Med Chem. 146 (2018) 554–563, https://doi.org/10.1016/j.ejmech.2018.01.080.

[9] Q. Ruan, Q. Liu, F. Sun, L. Shao, J. Jin, S. Yu, J. Ai, B. Zhang, W. Zhang, Moxifloxacin and gatifloxacin for initial therapy of tuberculosis: a meta-analysis of randomized clinical trials, Emerg. Microbes Infect. 5 (E12) (2016) 1–8, https://doi.org/10.1038/emi.2016.12.

[10] http://www.clinicaltrials.gov/ct2/results?cond=Tuberculosis&term=moxifloxacin&cntry=&state=&city=&dist= (data accessed on April 19th, 2019).

[11] F. Collin, S. Karkare, A. Maxwell, Exploiting bacterial DNA gyrase as a drug target: current state and perspectives, Appl. Microbiol. Biotechnol. 92 (2011) 479–497, https://doi.org/10.1007/s00253-011-3557-z.

[12] T. Khan, K. Sankhe, V. Suvarna, A. Sherje, K. Patel, B. Dravyakar, DNA gyrase inhibitors: progress and synthesis of potent compounds as antibacterial agents, Biomed. Pharmacother. 103 (2018) 923–938, https://doi.org/10.1016/j.biopha.2018.04.021.

[13] T.R. Blower, B.H. Williamson, R.J. Kerns, J.M. Berger, Crystal structure and stability of gyrase-fluoroquinolone cleaved complexes from Mycobacterium tuberculosis, Proc. Natl. Acad. Sci. USA 113 (7) (2016) 1706–1713, https://doi.org/10.1073/pnas.1525047113.

[14] V. Nagaraja, A.A. Godbole, S.R. Henderson, A. Maxwell, DNA topoisomerase I and DNA gyrase as targets for TB therapy, Drug Discov. Today. 22 (3) (2017) 510–518, https://doi.org/10.1016/j.drudis.2016.11.006.

[15] J. Azema, B. Guidetti, J. Dewelle, B. Le Calve, T. Mijatovic, A. Korolyov, J. Vaysse, M. Malet-Martino, R. Martino, R. Kiss, 7-((4-Substituted)piperazin-1-yl) derivatives of ciprofloxacin: synthesis and in vitro biological evaluation as potential antitumor agents, Bioorg. Med. Chem. 17 (2009) 5396–5407.

[16] C.G. de Almeida, C.G. Diniz, V.L. Silva, M.F. Saraiva, M. Le Hyaric, M.V. de Almeida, Antibacterial activity of lipophilic fluoroquinolone derivatives, Med. Chem. 5 (5) (2009) 419–421.

[17] M. Abdel-Aziz, S.E. Park, D. Abuo-Rahma Gel, M.A. Sayed, Y. Kwon, Novel N-4-piperazinyl-ciprofloxacin-chalcone hybrids: synthesis, physicochemical properties, anticancer and topoisomerase I and II inhibitory activity, Eur. J. Med. Chem. 69 (2013) 427–438.

[18] Y. Asahina, I. Araya, K. Iwase, F. Iinuma, M. Hosaka, T. Ishizaki, Synthesis and antibacterial activity of the 4-quinolone-3-carboxylic acidderivatives having a trifluoromethyl group as a novel N-1 substituent, J. Med. Chem. 48 (2005) 3443–3446.

[19] D. Sriram, P. Yogeeswari, J.S. Basha, D.R. Radhab, V. Nagaraja, Synthesis and antimycobacterial evaluation of various 7-substituted ciprofloxacin derivatives, Bioorg. Med. Chem. 13 (2005) 5774–5778.

[20] A. Foroumadi, S. Ghodsi, S. Emami, S, Najjari, N, Samadi, M.A. Faramarzi, L. Beikmohammadi, F.H. Shirazi, A. Shafiee, Synthesis and antibacterial activity of new fluoroquinolonescontaining a substituted N-(phenethyl)piperazine moiety, Bioorg. Med. Chem. Lett. 16 (2006) 3499–3503.

[21] P. Senthilkumar, M. Dinakaran, P. Yogeeswari, A. China, V. Nagaraja, D. Sriram, Antimycobacterial activities of novel fluoroquinolones, Biomed. Pharmacother. 63 (2009) 27–35.

[22] S.R. Md-Saleh, E.C. Chilvers, K.G. Kerr, S.J. Milner, A.M. Snelling, J.P. Weber, G.H. Thomas, A.K. Duhme-Klair, A. Routledge, Synthesis of citrate-ciprofloxacin conjugates, Bioorg. Med. Chem. Lett. 19 (5) (2009) 1496–1498, https://doi.org/10.1016/j.bmcl.2009.01.007.

[23] A. Darehkordi, M. Javanmiri, S. Ghazi, S. Assar, Synthesis of N-aryl-2,2,2-trifluoroacetimidoyl piperazinylquinolone derivatives and their antibacterial evaluations, J. Fluorine Chem. 132 (2011) 263–268.

[24] S.T. Nguyen, X. Ding, M.M. Butler, T.F. Tashjian, N.P. Peet, T.L. Bowlin, Preparation and antibacterial evaluation of decarboxylated fluoroquinolones, Bioorg. Med. Chem. 21 (2011) 5961–5963.

[25] T. Plech, M. Wujec, U. Kosikowska, A. Malm, B. Rajtar, M. Polz-Dacewicz, Synthesis and in vitro activity of 1,2,4-triazole-ciprofloxacin hybrids against drug-susceptible and drug-resistant bacteria, Eur. J. Med. Chem. 60 (2013) 128–134, https://doi.org/10.1016/j.ejmech.2012.11.040.

[26] R. Cormier, W.N. Burda, L. Harrington, J. Edlinger, K.M. Kodigepalli, J. Thomas, R. Kapolka, G. Roma, B.E. Anderson, E. Turos, L.N. Shaw, Studies on the antimicrobial properties of N-acylated ciprofloxacins, Bioorg. Med. Chem. Lett. 22 (20) (2012) 6513–6520, https://doi.org/10.1016/j.bmcl.2012.05.026.

[27] P. Chen, W. Lin, A. Lee, M. Hu, New 7-[4-(4-(un)substituted)piperazine-1-carbonyl]-piperazin-1-yl] derivatives of fluoroquinolone: synthesis and antimicrobial evaluation, Molecules 18 (2013) 7557–7569, https://doi.org/10.3390/molecules18077557.

[28] B. Marquez, V. Pourcelle, C.M. Vallet, M. Mingeot-Leclercq, P.M. Tulkens, J. Marchand-Bruynaert, F. Van Bambeke, Pharmacological characterization of 7-(4-(piperazin-1-yl)) ciprofloxacin derivatives: antibacterial activity, cellular accumulation, susceptibility to efflux transporters, and intracellular activity, Pharm. Res. 31 (2014) 1290–1301.

[29] C. Ji, P.A. Miller, M.J. Miller, Syntheses and antibacterial activity of N-acylated ciprofloxacin derivatives based on the trimethyl lock, ACS Med. Chem. Lett. 6 (2015) 707–710, https://doi.org/10.1021/acsmedchemlett.5b00146.

[30] P.C. Sharma, A. Jain, M.S. Yar, R. Pahwa, J. Singh, S. Goel, Synthesis and antibacterial evaluation of novel analogs of fluoroquinolones annulated with 6-substituted-2-aminobenzothiazoles, Arab. J. Chem. 8 (2015) 671–677.

[31] R. Ge, Q. Zhao, Z. Xie, L. Lu, Q. Guo, Z. Li, L. Zhao, Synthesis and biological evaluation of 6-fluoro-3-phenyl-7- piperazinyl quinolone derivatives as potential topoisomerase I inhibitors, Eur. J. Med. Chem. 122 (2016) 465–474.

[32] E. Chugunova, N. Akylbekov, A. Bulatova, N. Gavrilov, A. Voloshina, N. Kulik, V. Zobov, A. Dobrynin, V. Syakaev, A. Burilov, Synthesis and biological evaluation of novel structural hybrids of benzofuroxan derivatives and fluoroquinolones, Eur. J. Med. Chem 116 (2016) 165–172.

[33] S.S. Panda, O.S. Detistov, A.S. Girgis, P.P. Mohapatra, A. Samir, A.R. Katritzky, Synthesis and molecular modeling of antimicrobial active fluoroquinolone–pyrazine conjugates with amino acid linkers, Bioorg. Med. Chem. Lett. 26 (2016) 2198–2205.

[34] R. Kant, V. Singh, G. Nath, S.K. Awasthi, A. Agarwal, Design, synthesis and biological evaluation of ciprofloxacin tethered bis-1,2,3-triazole conjugates as potent antibacterial agents, Eur. J. Med. Chem. 124 (2016) 218–228.

[35] H.K. Gençer, S. Levent, U.A. Çevik, Y. Özkay, S. Ilgın, New 1,4-dihydro[1,8]naphthyridine derivatives as DNA gyrase inhibitors, Bioorg. Med. Chem. Lett. 27 (2017) 1162–1168.

[36] X. Yan, Z. Lv, J. Wen, S. Zhao, Z. Xu, Synthesis and in vitro evaluation of novel substituted isatinpropylene-1H-1,2,3-triazole-4-methylene-moxifloxacin hybrids for their anti-mycobacterial activities, Eur. J. Med. Chem. 143 (2018) 899–904.

[37] H.M. Refaat, D.A.N. El-Din, Chemical and biological evaluation of moxifloxacin-benzimidazole mixed ligands complexes: anti-cancer and anti-oxidant activities, J. Mol. Str. 1163 (2018) 103–113.

[38] G. Zhang, X. Liu, S. Zhang, B. Pan, M. Liu, Ciprofloxacin derivatives and their antibacterial activities, Eur. J. Med. Chem. 146 (2018) 599–612.

[39] T.R. Towle, C.A. Kulkarni, L.M. Oppegard, B.P. Williams, T.A. Picha, H. Hiasa, R.J. Kerns, Design, synthesis, and evaluation of novel N-1 fluoroquinolone derivatives: probing for binding contact with the active site tyrosine of gyrase, Bioorg. Med. Chem. Lett. 10 (2018) 1903–1910, https://doi.org/10.1016/j.bmcl.2018.03.085.

[40] K. Dileep, S. Polepalli, N. Jain, S.K. Buddana, R.S. Prakasham, M.S.R. Mury, Synthesis of novel tetrazole containing hybrid ciprofloxacin and pipemidic acid analogues and preliminary biological evaluation of their antibacterial and antiproliferative activity, Mol. Divers. 22 (2018) 83–93, https://doi.org/10.1007/s11030-017-9795-y.

[41] A.A. Abdel-Aziz, Y.A. Asiri, M.H. Al-Agamy, Design, synthesis and antibacterial activity of fluoroquinolones containing bulky arenesulfonyl fragment: 2D-QSAR and docking study, Eur. J. Med. Chem. 46 (46) (2011) 5487–5497, https://doi.org/10.1016/j.ejmech.2011.09.011.

[42] A. Demirci, K.G. Karayel, E. Tatar, S.Ö. Okullu, N. Ünübol, P.N. Taşlı, Z.T. Kocagöz, F. Şahin, İ. Küçükgüzel, Synthesis and evaluation of novel 1,3,4-thiadiazole

{fluoroquinolone hybrids as antibacterial, antituberculosis, and anticancer agents, Turk. J. Chem. 42 (2018) 839–858.

[43] E.J.C. Goldstein, K.L. Tyrrell, D.M. Citron, Lactobacillus species: taxonomic complexity and controversial susceptibilities, Clin. Infect. Dis. 60 (Suppl 2) (2015) 98–107, https://doi.org/10.1093/cid/civ072.

[44] K.B. Deepika, E.S. Priya, K. Murali, N.R. Kumar, Synthesis of various sulphonamide-linked fluoroquinolones as antibacterial agents, Asian J. Chem. 22 (6) (2010) 4363–4370.

[45] B. Tanwar, A. Kumar, P. Yogeeswari, D. Sriram, A.K. Chakraborti, Design, development of new synthetic methodology, and biological evaluation of substituted quinolines as new anti-tubercular leads, Bioorg. Med. Chem. Lett. 26 (24) (2016) 5960–5966, https://doi.org/10.1016/j.bmcl.2016.10.082.

[46] P.B. Devi, S. Jogula, A.P. Reddy, S. Saxena, J.P. Sridevi, D. Sriram, P. Yogeeswari, Design of novel mycobacterium tuberculosis pantothenate synthetase inhibitors: virtual screening, synthesis and in vitro biological activities, Mol. Inf. 34 (2015) 147–159, https://doi.org/10.1002/minf.201400120.

[47] K.K. Alluri, R.S. Reshma, R. Suraparaju, S. Gottapu, D. Sriram, Synthesis and evaluation of 40,50-dihydrospiro[piperidine-4,70-thieno[2,3-c]pyran] analogues against both active and dormant Mycobacterium tuberculosis, Bioorg. Med. Chem. 26 (2018) 1462–1469.

[48] S. Saxena, J. Renuka, P. Yogeeswari, D. Sriram, Discovery of novel mycobacterial GNA gyrase B inhibitors: in silico and in vitro biological evaluation, Mol. Inf. 33 (2014) 597–609, https://doi.org/10.1002/minf.201400058.

[49] G.L. Woods, B.A. Brown-Elliott, E.P. Desmond, G.S. Hall, L. Heifets, G.E. Pfyffer, J. C. Ridderhof, R.J. Wallace Jr., N.C. Warren, F.G. Witebsky, Susceptibility testing of mycobacteria, nocardiae, and other aerobic actinomycetes; Approved Standard. NCCLS document M24-A 23 (18) (2003).

[50] P. A. Willanova, Methods for Determining Bactericidal Activity of Antimicrobial Agents; Approved Guideline. NCCLS document M26-A 19 (18) (1999).