Eur. J. Med. Chem. 33 (1998) 697–703
© Elsevier, Paris

Synthesis and antibacterial activity of 7-hydrazinoquinolones

Rajeshwar Singh[a]*, Rakhshandeh Fathi-Afshar[a], George Thomas[a], Maya Prakash Singh[a,1], Fusahiro Higashitani[b], Akio Hyodo[b], Norio Unemi[b], Ronald George Micetich[a]

[a]*SynPhar Laboratories Inc., Taiho Alberta Center, #2, 4290-91A Street, Edmonton, Alberta, T6E 5V2, Canada*
[b]*Taiho Pharmaceutical Co. Ltd., Tokushima, Japan*

(Received 17 November 1997; accepted 18 March 1998)

Abstract – A series of new C-7 substituted hydrazino quinolones and naphthyridines were prepared and tested for antibacterial activity. The hydrazine bridge at the C-7 position did not favor the antibacterial activity, whereas the nature of other substituents at N-1, C-5 and C-8 did noticeably influence the antibacterial activity. The 7-(1-aminomorpholino) derivatives exhibited superior antibacterial activity against Gram-positive and inferior activity against Gram-negative bacteria than the 7-(1-aminopiperazinyl) derivatives. Substitution of the quinolone at position-1 with cyclopropyl was the most beneficial for antibacterial activity among the series of compounds prepared. © Elsevier, Paris

fluoroquinolone antibacterials / 7-hydrazino substituted fluoroquinolones / structure–activity relationship of fluoroquinolone derivatives / antibacterial activity of fluoroquinolones

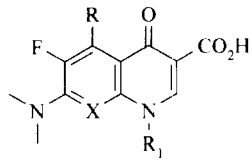
1. Introduction

The quinolone carboxylic acids constitute a class of extremely potent and orally active broad spectrum antibacterial agents [1–4]. These compounds have been shown to affect the bacterial growth by inhibiting the DNA gyrase, a key enzyme in bacterial DNA replication [5,6]. Various structural modifications of this class of compounds have provided the new agents such as ciprofloxacin [7], ofloxacin [8], lomefloxacin [9] and sparfloxacin [10] which are considerably more potent and have a broader spectrum of antibacterial activity (see *figure 1*). All the above compounds possess at least one fluorine atom at the C-6 position and a piperazine moiety at the C-7 position [11] of their structures. It was observed by Domagala et al. [12] that the piperazine group, although beneficial, is not essential either for low MICs or for low IC_{50} against the target enzyme, DNA gyrase. Domagala et al. has synthesized quinolone carboxylic acids with a 3-aminopyrrolidine substitution at C-7, in which the amino group has several degrees of freedom of rotation in comparison to the piperazinyl nitrogen. This class of compounds is more potent against Gram-positive bacteria, which may be due to the conformational differences between the five and the six membered rings. Based on the possibility of conformational effects of the C-7 substituents on the antibacterial activity, we have designed and synthesized 7-substituted hydrazino quinolonecarboxylic acids **6**, in which the N–N bond could have several degrees of freedom, and could provide optimal conformation for potent antibacterial activities. The present paper deals with the synthesis and in vitro antimicrobial activity of such compounds against selected bacterial isolates.

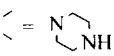
2. Chemistry

A generalized synthetic scheme for the synthesis of 7-hydrazino substituted fluoroquinolones **6** is given below in *figure 2* (see also *table I*). The pyridonecarboxylic acids **7** required as starting materials, were synthesized according to the literature methods [13–17], whereas the amines **8** (Y=O, N–CH_3) were commercially available.

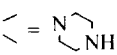
*Correspondence and reprints
[1]Present address: Wyeth-Ayerst Research, American Home Product Corporation, Pearl River, NY 10965, USA.



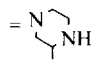
1. Norfloxacin R=H, R_1= C_2H_5, X=CH,



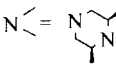
2. Ciprofloxacin R=H, R_1= c-C_3H_5, X=CH,



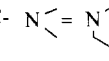
3. Lomefloxacin R=H, R_1= C_2H_5, X=CF,

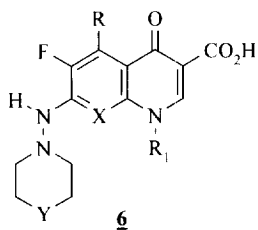


4. Sparfloxacin $R=NH_2$, R_1= c-C_3H_5, X=CF,



5. Ofloxacin R=H, R_1-X = -$CH(CH_3)CH_2OC$-





6

Figure 1.

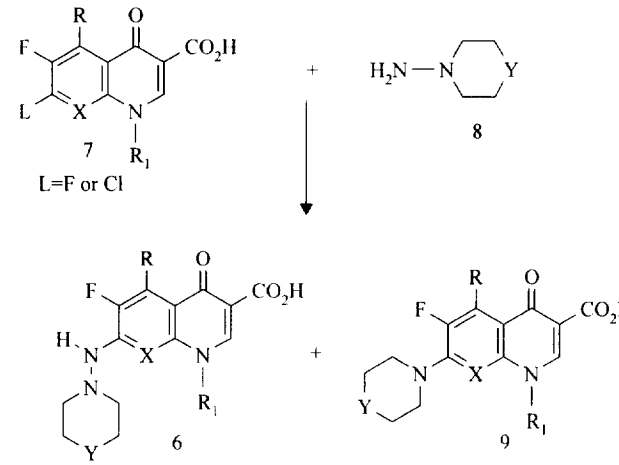
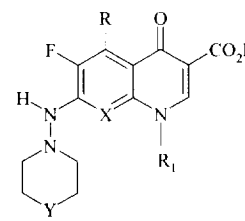


Figure 2.

Table I. List of synthesized 7-hydrazinofluoroquinolones.



6

Compound	R	R_1	X	Y
a	H	4-Fluorophenyl	CH	N–CH_3
b	H	4-Fluorophenyl	CH	O
c	H	4-Fluorophenyl	CF	O
d	H	4-Fluorophenyl	CF	N–CH_3
e	H	2,4-Difluorophenyl	CH	N–CH_3
f	H	2,4-Difluorophenyl	CF	N–CH_3
g	H	Cyclopropyl	CF	N–CH_3
h	H	Cyclopropyl	N	N–CH_3
i	NH_2	Cyclopropyl	CF	N–CH_3
j	H	Ethyl	CF	N–CH_3

The quinolonecarboxylic acids **7** (X=CH, CF) were reacted with amines **8** in solvent such as pyridine or N-methyl-2-pyrrolidinone at temperatures varying from 70 to 110 °C and naphthyridinecarboxylic acid **7** (X=N) with amines 8 at room temperature in acetonitrile. In most of these reactions, unlike the report of Ambros et al. [18], the desired hydrazino quinolones **6** were isolated along with another compound **9**, and the ratio of these two compounds was found to vary with changes in the reaction time, temperature and solvent. The structure of compounds **6** and **9** were established on the basis of elemental analysis, proton NMR and mass spectra, and by the synthesis of the corresponding authentic compound.

Formation of compound **9**, during the coupling reaction of pyridone-3-carboxylic acids **7** with amines **8**, can be explained by either of the following two possible mechanisms (*figure 3*): (i) the aminopiperazine is thermally unstable [19] and during heating it is converted to piperazine which reacts with the quinolonecarboxylic acid to give the deaminated product, or (ii) the more basic tertiary amine of the N-amino-4-substituted piperazine reacts at the 7-position of 7-halo pyridone-3-carboxylic acid followed by the deamination of the primary amine [20]. Condensation of N-aminopiperidine with o-halonitrobenzene in various solvents, similar to second reaction pathway, has been reported in literature [21]. During the reaction of 4-oxoquinoline-3-carboxylic acid with N-aminopiperazine, the 4-oxo group may be facilitating the attack of the more basic tertiary nitrogen at the

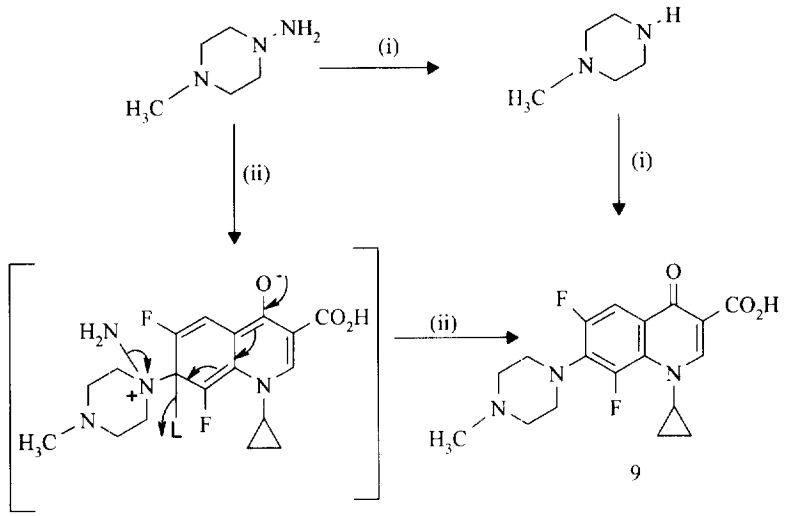


Figure 3. Possible mechanism of the formation of compound **9**.

7-fluoro substituent of 6,7-difluoro-4-oxoquinoline-3-carboxylic acid, resulting in the formation of deaminated product **9**.

3. Result and discussion

A series of C-7 substituted hydrazinoquinolones and naphthyridines were prepared and tested for their antibacterial activity (*table II*). The hydrazino bridge at the C-7 position did not seem to favor the antibacterial activity (compare MICs of **6a–j** and **9**), however the nature of other substituents at N-1, C-5 and C-8 did noticeably influence the activity. The 7-(1-aminomorpholino) derivatives **6b** and **6c** were more active against Gram-positive and 7-(1-aminopiperazinyl) derivatives were more active against Gram-negative bacteria. The 4-fluorophenyl substituent at N-1 and a fluorine atom at the C-8 position increased the activity against *P. aeruginosa* and *S. aureus* and decreased the activity against *E. coli* and *K. pneumoniae*. The combination of the difluorophenyl at N-1 and fluorine at C-8 did not appear to favor antipseudomonal activity. The N-1 cyclopropyl derivatives were the most active among the present series against gram-negative bacteria tested. Compounds **6f** and **6g** were further tested against a panel of resistant clinical isolates of Gram-positive and Gram negative bacteria. Both the compounds have no better activity over reference compounds. The antibacterial activity results are summarized in *table III*.

The poor activity of the 7-hydrazinoquinolones may be explained on the basis of the tetrahedral molecular orbital state of the hydrazine amine. Two structural conformations of 7-hydrazinoquinolones, A and B are possible which seem to be unsuitable to fit the binding site of DNA gyrase in spite of having almost the same distance between C-7 and N-4 as that of the piperazine moiety (*figure 4*).

4. Experimental protocols

4.1. General methods

Melting points were determined on an Electrothermal digital melting point apparatus and the values are uncorrected. ^{1}H NMR

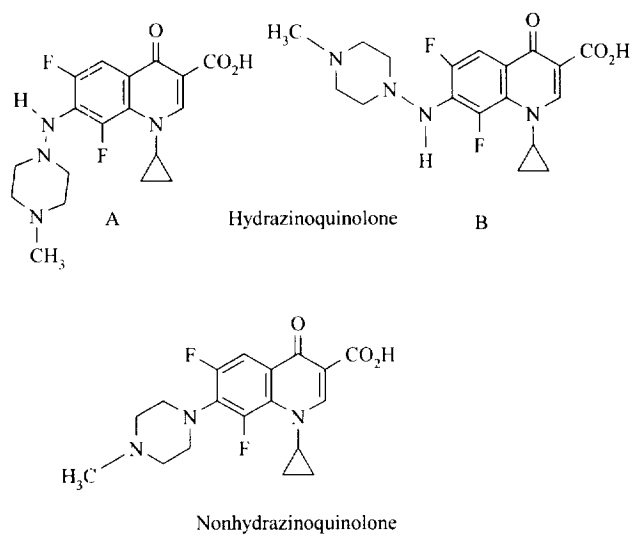


Figure 4.

Table II. Antibacterial activity (MIC, (g/mL) of 7-substituted hydrazinofluoroquinolones **6**.

Test organisms	CPLX	**6a**	**6b**	**6c**	**6d**	**6e**	**6f**	**6g**	**6h**	**6i**	**6j**	**9**
Sa ATCC29213	0.5	0.12	0.25	1.0	2.0	2.0	2.0	2.0	> 8	1.0	4.0	0.125
Sa JHHD078	0.5	0.12	0.25	2.0	4.0	2.0	2.0	4.0	> 8	ND	4.0	0.06
Sa JHHM241	0.5	0.12	0.25	2.0	4.0	2.0	2.0	2.0	> 8	ND	4.0	0.125
Ec ATCC25922	0.015	0.25	1.0	4.0	1.0	1.0	0.5	0.25	0.05	0.25	0.5	0.015
Ec DCO	0.12	> 8	> 8	> 8	> 8	> 8	> 8	8.0	> 8	8.0	> 8	0.50
Ec DC2	0.06	1.0	1.0	> 8	4.0	1.0	1.0	1.0	4.0	ND	2.0	0.06
Ec SHV1	0.015	0.25	1.0	4.0	1.0	1.0	1.0	0.25	0.5	ND	0.5	0.015
Ec TEM1	0.015	0.25	1.0	4.0	2.0	1.0	0.5	0.25	0.5	ND	0.5	0.015
Ec TEM2	0.0075	0.50	0.12	4.0	0.06	1.0	0.5	0.25	0.5	ND	0.5	0.015
Ec OXA1	0.12	2.0	2.0	> 8	8.0	2.0	2.0	2.0	4.0	ND	4.0	0.06
Ec OXA3	0.015	0.25	0.5	4.0	2.0	1.0	0.5	0.25	0.5	ND	0.5	0.015
Kp ATCC13883	0.03	1.0	1.0	4.0	2.0	2.0	2.0	0.50	1.0	0.25	1.0	0.03
Pr ATCC29944	0.06	1.0	2.0	4.0	4.0	4.0	4.0	1.0	4.0	1.0	2.0	0.03
Pm ATCC2675	0.03	2.0	4.0	> 8	8.0	8.0	8.0	1.0	8.0	ND	2.0	0.03
Ecl ATCC23355	0.03	1.0	1.0	2.0	1.0	2.0	2.0	1.0	1.0	1.0	1.0	0.03
Cf (R-Ceph)	0.06	4.0	16	8.0	8.0	> 8	> 8	2.0	4.0	ND	4.0	0.25
Sm ATCC29882	0.06	2.0	4.0	4.0	4.0	2.0	8.0	1.0	2.0	1.0	8.0	0.125
Pa ATCC27853	0.03	4.0	4.0	4.0	4.0	4.0	> 8	> 8	> 8	> 8	> 8	0.50
Pa PSE1	0.12	2.0	4.0	1.0	4.0	4.0	> 8	8.0	8.0	ND	8.0	0.50
Pa PSE2	0.12	2.0	4.0	> 8	4.0	8.0	> 8	0.25	0.5	ND	8.0	0.015

Sa: Staphylococcus aureus; Ec: Escherichia coli; Kp: Klebsiella pneumoniae; Pr: Providencia rettgeri; Pm: Proteus mirabilis; Ecl: Enterobacter cloacae; Cf: Citrobacter freundii; Sm: Serratia marcescens; Pa: Pseudomonas aeruginosa; CPLX: Ciprofloxacin; ND: Not done.

spectra (ppm) were obtained on a Brucker AM-300 spectrometer in TFA with tetramethylsilane as an internal standard. The chemical purities of the synthesized compounds were checked on thin layer chromatography plates. Mass spectroscopic analyses (FAB) were performed on an AEI MS-9 (modified) mass spectrometer for molecular ion peaks.

4.2. Antibacterial activity

In vitro antibacterial activity against a variety of Gram-positive and Gram-negative organisms was determined by standard agar dilution method [22]. One replicator pin of an overnight culture of each test strain in saline was streaked to deliver 10^4 CFU/spot on Mueller Hinton agar containing different concentrations of the antibacterial agent. The MIC values (minimum inhibitory concentration in (g/mL at which the microbes failed to grow into a visible spot) were noted after incubation at 35 °C for 16–18 h. A single colony or a hazy growth was disregarded.

4.3. 6-Fluoro-1-(4-fluorophenyl)-7-N-(4-methylpiperazin-1-yl) amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6a***

A mixture of 6,7-difluoro-1-(4-fluorophenyl)-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.319 g, 1.0 mmole) and 1-amino-4-methyl piperazine (0.290 g, 2.5 mmole) in 5 mL of N-methylpyrrolidin-2-one was heated at 75 °C for 16 h under nitrogen atmosphere. The reaction mixture was then concentrated and the residue was diluted with ethanol. The separated solid was filtered and recrystallised from ethanol. Yield 0.232 g (55%); m.p. 280 °C (d); NMR (TFA) δ (ppm): 9.16 (s, 1H), 8.25 (d, J = 10.8 Hz, 1H), 7.75–7.43 (m, 4H), 7.07 (d, J = 6.6 Hz, 1H), 4.12–3.4 (m, 8H), 3.09 (s, 3H); Anal. Found: C, 59.69; H, 4.99; N, 12.94; requires for $C_{21}H_{20}F_2N_4O_3 \bullet 1/2H_2O$: C, 59.57; H, 4.99; N, 13.23%; Mass (FAB) *m/e*: 415 (M+H).

4.4. 6-Fluoro-1-(4-fluorophenyl)-7-N-(morpholin-4-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6b***

A mixture of 6,7-difluoro-1-(4-fluorophenyl)-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.250 g, 0.78 mmole) and 4-aminomorpholine (0.199 g, 1.95 mmole) in 4 mL of pyridine was heated at 85 °C for 24 h under nitrogen atmosphere in a pressure reaction vessel. The reaction mixture was concentrated and the residue was diluted with ethanol. The separated solid was filtered, washed successively with water and ethanol, to obtain 0.2 g (66%) of the desired product. m.p. 300–301 °C; NMR (TFA) δ (ppm): 9.17 (s, 1H), 8.25 (d, J = 10.8 Hz, 1H), 7.89–7.43 (m, 4H), 7.23 (d, J = 6.4 Hz, 1H), 4.10–3.85 (m, 4H), 3.35–3.00 (m, 4H); Anal. Found: C, 58.68; H, 4.26; N, 9.92; requires for $C_{20}H_{17}F_2N_3O_4 \bullet 1/2H_2O$: C, 58.54; H, 4.42; N, 10.23%.

4.5. 6,8-Difluoro-1-(4-fluorophenyl)-7-N-(morpholin-4-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6c***

A mixture of 6,7,8-trifluoro-1-(4-fluorophenyl)-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.337 g, 1.0 mmole) and 4-aminomorpholine (0.26 g, 2.5 mmole) in 5 mL of pyridine was heated at 75 °C for 24 h under nitrogen atmosphere in a pressure

Table III. Antibacterial activity (MIC (g/mL) of selected 7-substituted hydrazinofluoroquinolones against resistant clinical isolates.

Test organisms	**6g**	**6f**	CPFX	OFLX
Staphylococcus aureus (MRSA)CT-3	3.13	3.13	0.39	0.39
Staphylococcus aureus (MRSA)CT-5	> 100	> 100	> 100	50
Staphylococcus aureus (MRSA)CT-10	> 100	> 100	50	12.5
Staphylococcus aureus (MRSA) CT-23	> 100	> 100	25	12.5
Enterococcus faecium CT-29	> 100	> 100	> 100	> 100
Enterococcus faecium CT-30	100	100	3.13	12.5
Enterococcus faecium CT-38	50	50	3.13	12.5
Escherichia coli CT-59	0.39	1.56	< 0.012	0.05
Escherichia coli CT-69	25	> 100	1.56	3.13
Escherichia coli CT-70	> 100	> 100	6.25	12.5
Escherichia coli CT-73	> 100	> 100	25	25
Citrobacter freundii CT-76	> 100	> 100	25	25
Citrobacter freundii CT-83	3.13	50	0.10	0.39
Enterobacter cloacae CT-92	> 100	> 100	6.25	12.5
Enterobacter cloacae CT-93	0.78	3.13	0.025	0.10
Enterobacter cloacae CT-95	> 100	> 100	100	100
Serratia marcescens CT-98	> 100	> 100	100	100
Serratia marcescens CT-103	> 100	> 100	25	50
Proteus vulgaris CT-106	> 100	> 100	100	50
Proteus vulgaris CT-109	1.56	12.5	0.05	0.20
Morganella morganii CT-111	> 100	> 100	12.5	12.5
Morganella morganii CT-112	> 100	> 100	50	50
Morganella morganii CT-115	0.78	0.10	< 0.012	0.20
Pseudomonas aeruginosa CT-121	50	> 100	1.56	12.5
Pseudomonas aeruginosa CT-122	> 100	> 100	50	> 100
Pseudomonas aeruginosa CT-137	> 100	> 100	50	> 100
Pseudomonas aeruginosa CT-144	12.5	25	0.39	1.56

reaction vessel. The reaction mixture was concentrated and the residue was diluted with acetonitrile. The separated solid was filtered washed with water, dried and crystallized from acetonitrile, to obtain 0.25 g (59%) of desired product. m.p. 285–290 °C; NMR (TFA) δ (ppm): 9.26 (s, 1H), 8.44 (dd, J_1 = 11.34 Hz, J_2 = 1.5 Hz, 1H), 7.75–7.55 (m, 2H), 7.50–7.35 (m, 2H), 4.3–3.9 (m, 8H); Anal. Found: C, 54.72; H, 4.08; N, 9.50; requires for $C_{20}H_{16}F_3N_3O_4 \bullet H_2O$: C, 54.92; H, 4.12; N, 9.61%.

4.6. 6,8-Difluoro-1-(4-fluorophenyl)-7-N-(4-methylpiperazin-1-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6d***

A mixture of 6,7,8-trifluoro-1-(4-fluorophenyl)-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.337 g, 1.0 mmole) and 1-amino-4-methylpiperazine (0.29 g, 2.5 mmole) in 10 mL of pyridine was heated at 60–70 °C for 4 h under nitrogen atmosphere in a pressure reaction vessel. The red homogenous reaction solution was concentrated, the residue was dissolved in hot ethanol and cooled. The separated solid was filtered and the filtrate was concentrated under vacuum. The residue was crystallized from acetonitrile, to obtain 0.08 g (18%) of desired product. m.p. 255–257 °C; NMR (TFA) δ (ppm): 9.45 (s, 1H), 8.23 (dd, J_1 = 11 Hz, J_2 = 1.4 Hz, 1H), 7.80–7.53 (m, 2H), 7.48–7.43 (m, 2H), 3.88–3.25 (m, 8H), 3.11 (s, 3H); Anal. Found: C, 57.23; H, 4.40; N, 12.48; requires for $C_{21}H_{19}F_3N_4O_3 \bullet 1/2H_2O$: C, 57.14; H, 4.57; N, 12.69%.

4.7. 1-(2,4-Difluorophenyl)-6-fluoro-7-N-(4-methylpiperazin-1-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6e***

A mixture of 6,7-difluoro-1-(2,4-difluorophenyl)-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.168 g, 0.5 mmole) and 1-amino-4-methylpiperazine (0.144 g, 1.25 mmole) in 3 mL of pyridine was heated at 100–110 °C for 5 h under nitrogen atmosphere in a pressure reaction vessel. The reaction mixture was concentrated and the residue was diluted with 10 mL of water. The separated solid was filtered, washed with water, dried and crystallized from ethanol. Yield 0.13 g (62%) m.p. 234–238 °C; NMR (TFA) δ (ppm): 9.14 (s, 1H), 8.26 (d, J = 10.8 Hz, 1H), 7.90–7.65 (m, 1H), 7.50–7.25 (m, 2H), 7.04 (dd, J_1 = 6.2 Hz, J_2 = 6.2 Hz, 1H), 3.90–3.65 (m, 2H), 3.50–3.00 (m, 6H), 3.10 (bs, 3H); Anal. Found: C, 57.36; H, 4.34; N, 12.55; requires for $C_{21}H_{19}F_3N_4O_3 \bullet 1/2H_2O$: C, 57.14; H, 4.57; N, 12.69%.

4.8. 6,8-Difluoro-1-(2,4-difluorophenyl)-7-N-(4-methylpiperazin-1-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6f***

A mixture of 6,7,8-trifluoro-1-(2,4-difluorophenyl)-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.25 g, 0.7 mmole) and 1-amino-4-methylpiperazine (0.21 g, 1.82 mmole) in 5 mL of pyridine was heated at 70 °C for 24 h under nitrogen atmosphere in a pressure reaction vessel. The reaction mixture was concentrated

and the residue was diluted with 10 mL of water. The separated solid was filtered dried and crystallized from ethanol. The crystallized solid was filtered and washed with a mixture of ether–hexane (1:1), to give 0.08 g (25%) of desired product. m.p. 224–226 °C; NMR (TFA) δ (ppm): 9.10 (s, 1H), 8.24 (dd, J_1 = 11.1 Hz, J_2 = 1.3 Hz, 1H), 7.85–7.55 (m, 1H), 7.35–7.08 (m, 2H), 3.97–3.23 (m, 8H), 3.12 (bs, 3H); Anal. Found: C, 53.64; H, 4.10; N, 11.71; requires for $C_{21}H_{18}F_4N_4O_3 \bullet H_2O$: C, 53.85; H, 4.30; N, 11.96%; Mass (FAB) *m/e*: 451 ((M+H)).

4.9. 6,8-Difluoro-1-cyclopropyl-7-N-(4-methylpiperazin-1-yl) amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6g***

A mixture of 1-cyclopropyl-6,7,8-trifluoro-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (1.42 g, 5.0 mmole) and 1-amino-4-methylpiperazine (1.44 g, 12.5 mmole) in 25 mL of pyridine was heated at 100–105 °C for 24 h under nitrogen atmosphere in a pressure reaction vessel. The reaction mixture was concentrated and the residue was diluted with water. The separated solid was removed by filtration and the filtrate was concentrated. The residue was crystallized with a mixture of ethanol–ether, to give 0.5 g (26%) of desired product. m.p. 208–210 °C; NMR (TFA) δ (ppm): 9.32 (s, 1H), 8.16 (dd, J_1 = 11 Hz, J_2 = 1.0 Hz, 1H), 4.61–4.39 (m, 1H), 3.84 (d, J = 10.9 Hz, 2H), 3.69–3.28 (m, 6H), 3.13 (s, 3H), 1.80–1.33 (m, 4H); Anal. Found: C, 56.02; H, 5.46; N, 14.46; requires for $C_{18}H_{20}F_2N_4O_3 \bullet 1/2H_2O$: C, 56.20; H, 5.46; N, 14.24%; Mass (FAB) *m/e*: 379 (M+H).

4.10. 1-Cyclopropyl-6-fluoro-7-N-(4-methylpiperazin-1-yl) amino1,4-dihydro-4-oxo-1,8-naphthyridine-3-carboxylic acid ***6h***

A mixture of 1-cyclopropyl-7-chloro-6-fluoro-1,4-dihydro-4-oxo-1,8-naphthyridine-3-carboxylic acid (0.07 g, 0.25 mmole) and 1-amino-4-methylpiperazine (0.08 g, 0.70 mmole) in 3 mL of acetonitrile was stirred at room temperature for 24 h under nitrogen atmosphere. The reaction mixture was concentrated and the residue was diluted with water. The aqueous solution was extracted with chloroform. The chloroform extract was dried over sodium sulfate and concentrated to give 0.04 g (45%) of desired product. m.p. 251–252 °C; NMR (TFA) δ (ppm): 8.75 (s, 1H), 8.06 (d, J = 10.6 Hz, 1H), 3.87–3.67 (m, 1H), 3.33–2.95 (m, 4H), 2.87–2.55 (m, 4H), 2.36 (s, 3H), 1.50–1.00 (m, 4H); Anal. Found: C, 54.43; H, 5.59; N, 18.48; requires for $C_{17}H_{20}FN_5O_3 \bullet 3/4H_2O$: C, 54.47; H, 5.78; N, 18.68%; Mass (FAB) *m/e*: 362 (M+H).

4.11. 5-Amino-1-cyclopropyl-6,8-difluoro-7-N-(4-methylpiperazin-1-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6i***

A mixture of 5-amino-1-cyclopropyl-6,7,8-trifluoro-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.24 g, 0.8 mmole) and 1-amino-4-methylpiperazine (0.23 g, 2.15 mmole) in 5 mL of pyridine was heated at 110 °C for 20 h. The reaction mixture was concentrated and the yellow residue was triturated with water (10 mL). The separated solid was removed by filtration and the filtrate was concentrated. After concentration, the residue was triturated with ether, the separated solid was filtered, washed with water and dried, to give 0.027 g (9%) of desired product. m.p. 174–175 °C; NMR (TFA) δ (ppm): 9.15 (s, 1H), 4.32 (bs, 1H), 3.85 (m, 2H), 3.65–3.25 (m, 6H), 3.10 (s, 3H), 1.50–1.30 (m, 4H); Anal. Found: C, 51.78; H, 5.64; N, 16.47; requires for $C_{18}H_{21}F_2N_5O_3 \bullet 1.5H_2O$: C, 51.43; H, 5.75; N, 16.66%; Mass (FAB) *m/e*: 394 (M+H).

4.12. 6,8-Difluoro-1-ethyl-7-N-(4-methylpiperazin-1-yl)amino-1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***6j***

A mixture of 1-ethyl-6,7,8-trifluoro-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.271 g, 1.0 mmole) and 1-amino-4-methylpiperazine (0.29 g, 2.5 mmole) in 5 mL of pyridine was heated at 70 °C for 24 h under nitrogen atmosphere in a pressure reaction vessel. The reaction mixture was concentrated and the residue was diluted with ethanol. The separated solid was filtered and crystallized from ethanol, to give 0.23 g (63%) of desired product. m.p. 199–203 °C; NMR (TFA) δ (ppm): 9.24 (s, 1H), 8.2 (dd, J_1 = 12 Hz, J_2 = 1.4 Hz, 1H), 4.99 (q, 2H), 3.93–3.20 (m, 8H), 3.16 (s, 3H), 1.79 (t, 3H); Anal. Found: C, 50.87; H, 5.85; N, 13.72; requires for $C_{17}H_{20}F_2N_4O_3 \bullet 2H_2O$: C, 50.74; H, 6.00; N, 13.93% Mass (FAB) *m/e*: 367 (M+H).

4.13. 6,8-Difluoro-1-cyclopropyl-7-N-(4-methylpiperazin-1-yl) -1,4-dihydro-4-oxoquinoline-3-carboxylic acid ***9***

A mixture of 1-cyclopropyl-6,7,8-trifluoro-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (0.283 g, 1.0 mmole) and 1-amino-4-methylpiperazine (0.285 g, 2.5 mmole) in 5 mL of pyridine was heated at 100–105 °C for 24 h under nitrogen atmosphere in a pressure reaction vessel. The reaction mixture was concentrated and the residue was diluted with water. The separated solid was filtered, washed successively with water, ethanol and acetonitrile to give 0.07 g (19%) of desired product. m.p. 218–219.5 °C; NMR (TFA) δ (ppm): 9.41 (s, 1H), 8.23 (dd, J_1 = 11 Hz, J_2 = 1 Hz, 1H), 4.68–4.46 (m, 1H), 4.18–3.79 (m, 6H), 3.64–3.40 (m, 2H), 3.22 (s, 3H), 1.78–1.37 (m, 4H); Anal. Found: C, 58.81; H, 5.39; N, 11.32; requires for $C_{18}H_{19}F_2N_3O_3 \bullet 1/4H_2O$: C, 58.77; H, 5.30; N, 11.42%; Mass (FAB) *m/e*: 364 (M+H).

Acknowledgements

The authors wish to thank B. Lix for NMR spectral analysis, J. Ng for elemental analysis and the Mass Spectrometry Section, Department of Chemistry, University of Alberta, for providing FAB mass spectra of the compounds reported in this manuscript.

References

[1] Chu D.T.W., Fernandes P.B., In: Testa B. (Ed.), Recent Developments in the Field of Quinolone Antibacterial Agents, in Advances in Drug Research, Vol. 21, Academic Press, San Diego, 1991, pp. 39–144.

[2] Mitscher L.A., Devasthle P.V., Zovod R.M., In: Crumplin C.G. (Ed.), The 4-Quinolones: Antibacterial Agents In Vitro, Springer, Berlin, 1990, pp. 115–146.

[3] Wentland M.P., In: Siporin C., Heifetz C.L., Domagala J.M. (Eds.), The New Generation of Quinolones, Marcel Dekker, New York, 1990, pp. 1–44.

[4] Sanchez J.P., Gogliotti R.D., Domagala J.M., Gracheck S.J., Huband M.D., Sesmi J.A., Cohen M.A., Shapiro M.A., J. Med. Chem. 38 (1995) 4478–4487.

[5] Fan J.-Y., Sun D.K., Yu H., Kerwin S.M., Hurley L.H., J. Med. Chem. 38 (1995) 408–424.

[6] Palumbo M., Gatto B., Zagotto G., Palu G., Trends Microbiol. 1 (1993) 232–235.

[7] Wise R., Andrews J.M., Edwards L.J., Antimicrob. Agents Chemother. 23 (1983) 559–564.

[8] Hayakawa I., Hiramitsu T., Tanaka Y., Chem. Pharm. Bull. 32 (1984) 4907–4913.

[9] Chin N.X., Novelli A., Neu H.C., Antimicrob. Agents Chemother. 32 (1988) 656–662.

[10] Miyamoto T., Matsumoto J.-i., Chiba K., Egawa H., Shibamori K., Minamida A., Nishimura Y., Okada H., Kataoka M., Fujita M., Hirose T., Nakano J., J. Med. Chem. 33 (1990) 1645–1656.

[11] Egawa H., Miyamoto T., Minamida A., Nishimura Y., Okada H., Uno H., Matsumoto J., J. Med. Chem. 27 (1984) 1543–1548.

[12] Domagala J.M., Heifetz C.L., Mich T.F., Nichols J.B., J. Med. Chem. 29 (1986) 445–448.

[13] Chu D.T.W., Lico I.M., Claiborne A.K., Faubl H., Can. J. Chem. 70 (1992) 1323–1327.

[14] Domagala J.M., Heifetz C.L., Hutt M.P., Mich T.F., Nichols J.B., Solomon M., Worth D.F., J. Med. Chem. 31 (1988) 991–1001.

[15] Sanchez J.P., Domagala J.M., Hagen S.E., Heifetz C.L., Hutt M.P., Nichols J.B., Trehan A.K., J. Med. Chem. 31 (1988) 983–991.

[16] Chu D.T.W., Fernandes P.B., Maleczka R.E., Nordeen C.W., Pernet A.G., J. Med. Chem. 30 (1987) 504–509.

[17] Domagala J.M., Bridges A.J., Culbertson T.P., Gambino L., Hagen S.E., Karrick G., Porter K., Sanchez J.P., Sesnie J.A., Spense F.G., Szotek D., Wemple J., J. Med. Chem. 34 (1991) 1142–1154.

[18] Ambrose R., Ribera S.G., Munoz A.S., Barenys J.M.C., Hernandez J.A.O., ES patent 20003196; Chem. Abstr. 111 (1989) 174004e.

[19] ^{1}H NMR and HPLC indicates the presence of 4-methylpiperazine after heating the 4-methyl-1-aminopiperazine at 90°C for 5h in pyridine.

[20] The exact mechanism of the deamination is not clearly understood at this stage.

[21] Latham D.W.S., Meth-Cohn O., Suschitzky H., Tetrahedron Lett. (1972) 5365–5368.

[22] Thorns-berry C., Anhalt J., Barry A.L. et al., Approved Standard M7-A, National Committee for Clinical Laboratory Standards, Villanova (PA), USA, 1985.