Antibacterial Activity of 6,8-Disubstituted-Quinolone-3-Carboxylic Acids

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Abstract □ Twelve 6,8-disubstituted-1-ethyl-1,4-dihydro-4-oxoquinolone-3-carboxylic acid derivatives were prepared and evaluated for their antibacterial activity. Among these, only the 6,8-difluoroquinolone-3-carboxylic acid showed moderate activity.

The third generation nalidixic acid derivatives (such as norfloxacin, ciprofloxacin, pefloxacin, and enoxacin)[1] exhibited potent antibacterial activity.[2] We initiated the synthesis of structural variants which bear the 6,8-substitution pattern.

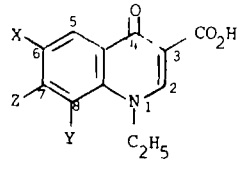
Substitution at the C-7 position in the quinolone system of 1 has been thoroughly investigated and the antibacterial activity of these compounds documented.[3] Contrary to this, substitution at the C-6 position of 1 with a fluorine at the C-8 position has not been previously reported. A series of compounds with substituents at the C-6 and C-8 positions in the aromatic ring system were prepared and the antibacterial spectrum determined.

The requisite 6,8-disubstituted-1-alkyl-1,4-dihydro-4-oxoquinoline-3-carboxylic acid was prepared by the usual route.

Aniline 2 was heated with diethyl ethoxymethylenemalonate to give the malonate 3 which, without purification, was cyclized to the 4-hydroxyquinoline-3-carboxylic acid ethyl ester (4; Scheme I). Alkylation of the ester by treatment with ethyl iodide and anhydrous potassium carbonate gave the 1-alkyl-1,4-dihydro-4-oxoquinoline ester 5. The 1-alkyl ester was hydrolyzed with dilute HCl to produce the carboxylic acid 6 whose structure was confirmed by spectral data.

The 6,8-difluoro-4-quinolinone 6 was treated with *N*-methylpiperazine to yield the desired 6-piperazinyl-8-fluoro-4-quinoline 7a. Evidence for the replacement of the 6-fluoro substituent was established by NMR. If the 8-fluoro substituent was replaced, the C-7 proton would appear as a doublet at δ 7.6 ppm ($J_{H\text{-}F}$ = 8.2 Hz) along with the doublet of the C-5 proton at δ 7.8 ppm ($J_{H\text{-}F}$ = 8.3 Hz). However, the appearance of the C-7 proton as a doublet at δ 7.8 ppm ($J_{H\text{-}F}$ = 8.2 Hz) with a singlet at δ 7.6 ppm corresponding to the C-5 proton confirmed the substitution of the C-6 fluorine by *N*-methylpiperazine.

The difluoroquinolinone was treated with a variety of piperazines to obtain the derivatives (7b–j) listed in Table I.



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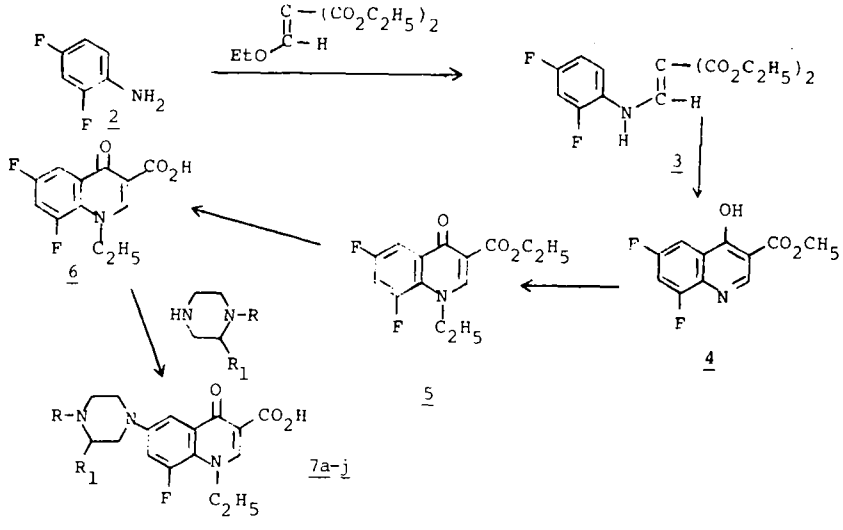
X = F, Y = H, Z = piperazinyl
X = piperazinyl, Y = F, Z = H

Experimental Section

All melting points were taken on a Mel-Temp apparatus and are uncorrected. Samples for elemental analysis were dried for 1–24 h under reduced pressure. The ^{1}H NMR spectra were obtained on a Varian model FT-80 spectrometer and chemical shift values are reported in δ downfield from the internal standard (Me_4Si). All spectra were taken in deuterated solvents. The analyses were within ±0.4% of the calculated value. The IR spectra were obtained using a Perkin Elmer 1310 spectrophotometer. Mass spectra were obtained on a Finnegan MAT model CH7 mass spectrometer.

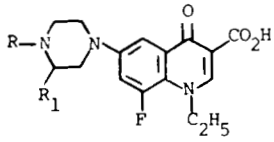
Ethyl 6,8-Difluoro-4-hydroxyquinoline-3-carboxylate (4)—A mixture of 2,4-difluoroaniline (2; 25 g; 0.19 mol) and diethyl ethoxymethylenemalonate (48 g; 0.22 mol) was heated at 120–130 °C. After



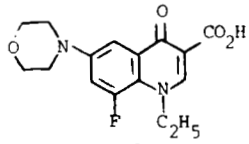
Scheme I

0022-3549/86/1200-1185$01.00/0

Table I—Difluoroquinolinone Derivatives



7a–j



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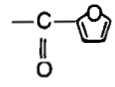
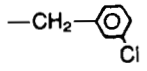
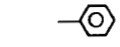
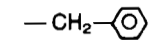
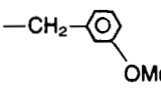
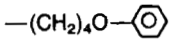
Compound	R	R_1	Yield, %	Recrystn. Solvent	mp, °C	Formula
7a	CH_3	H	69	DMF	218–220	$C_{17}H_{19}FN_3O_3$
7b	H	H	70	DMF	195–197	$C_{16}H_{18}FN_3O_3$
7c	H	CH_3	68	EtOH	175–177	$C_{17}H_{19}FN_3O_3$
7d		H	72	DMF	260–262	$C_{21}H_{20}FN_3O_5$
7e	$-(CH_2)_3-NMe_2$	H	69	DMF	185–187	$C_{21}H_{29}FN_4O_3$
7f		H	75	DMF	188–190	$C_{23}H_{23}FClN_3O_3$
7g		H	78	DMF	263–265	$C_{22}H_{22}FN_3O_3$
7h		H	63	DMF	118–120	$C_{23}H_{24}FN_3O_3$
7i		H	80	DMF	192–194	$C_{24}H_{26}FN_3O_4$
7j		H	80	EtOH	168–170	$C_{26}H_{30}FN_3O_4$
8	—	—	90	EtOH	253–255	$C_{16}H_{17}FN_2O_4$

4 h, the resulting EtOH had evaporated and the residual solid material was used in the successive reaction without further purification, mp 75–77 °C; ^1H NMR (Me_2SO-d_6): δ 1.2–1.4 (6, m, CH_3), 4.1–4.3 (4, m, CH_2), 7.2–7.65 (2, m, ArH), 8.3 (1, dd, J_{H-F} = 8 Hz), and 10.81 ppm (1, d, J_{H-F} = 13.0 Hz, NH); IR (KBr): 1685 cm^{-1} (ester); MS (70 ev): *m/z* 299 (M^+).

The aforementioned malonate (3; 29 g; 0.097 mol) was added to diphenyl ether (280 mL) and refluxed for 1 h. After the solution was cooled, the precipitate was removed by filtration, washed with hexane, dried and recrystallized from ethanol, mp 275–277 °C; ^1H NMR (Me_2SO-d_6): δ 1.34 (3, t, J_{H-H} = 7.0 Hz, CH_3), 4.3 (2, q, J_{H-H} = 8.1 Hz, CH_2), 7.56 (1, d, J_{H-F} = 8.0 Hz, ArH), 7.68 (1, dd, J_{H-F} = 8.1 Hz, ArH), and 8.38 ppm (1, s, ArH); IR (KBr): 1690 cm^{-1} (ester); MS (70ev): *m/z* 281 (M^+).

Ethyl 6,8-Difluoro-1-ethyl-1,4-dihydro-4-oxoquinoline-3-carboxylate (5)—A mixture of (4; 5 g; 0.0197 mol), K_2CO_3 (5.5 g; 0.039 mol), EtI (12.3 g; 0.078 mol), and dimethylformamide (60 mL) was heated at 80–90 °C with stirring. After 24 h, the mixture was evaporated to dryness and washed with ice-cold water. The crude ester was used in the successive reaction without further purification. The residue was recrystallized from ethanol to yield 5, mp 168–170 °C; ^1H NMR (Me_2SO-d_6): δ 1.3–1.5 (6, m, CH_3), 4.15–4.3 (4, m, CH_2), 7.7 (1, d, J_{H-F} = 8.0 Hz, ArH), 7.85 (1, dd, J_{H-F} = 8.1 Hz, ArH), and 8.62 ppm (1, s, ArH); IR (KBr): 1720 (ester), 1615 (C=O) cm^{-1}; MS (70 ev): *m/z* 281 (M^+).

6,8-Difluoro-1-ethyl-1,4-dihydro-4-oxoquinoline-3-carboxylic acid (6)—A mixture of 5 (10 g; 0.035 mol) and 1 M HCl (70 mL) was refluxed with stirring. After 3 h, the precipitate was removed by filtration, washed with water, and dried. The solid was recrystallized from ethanol to yield 6 (8.3 g, 92%), mp 210–212 °C; ^1H NMR (Me_2SO-d_6): δ 1.52 (3, t, J_{H-H} = 7.0 Hz, CH_3), 4.2 (2, q, J_{H-H} = 7.0 Hz, CH_2), 7.82 (1, d, J_{H-F} = 8.0 Hz, ArH), 7.93 (1, dd, J_{H-F} = 8.2 Hz, ArH), and 8.92 ppm (1, s, ArH); IR (KBr): 1715 (COOH), 1610 (C=O) cm^{-1}; MS (70ev): *m/z* 253 (M^+).

1-Ethyl-8-fluoro-1,4-dihydro-4-oxo-6-(*N*-methylpiperazinyl)quinoline-3-carboxylic acids (7a–j)—A typical procedure is presented. A mixture of 6 (1 g; 0.0039 mol) and *N*-methylpiperazine (2.5 g; 0.025 mol) was heated at 130–140 °C with stirring. After 5 h, the mixture was cooled, treated with methanol, and then the solid was removed by filtration and recrystallized from dimethylformamide to give 7a (0.96 g; 69%), mp 218–220 °C; ^1H NMR (Me_2SO-d_6): δ 1.79 (3, t, J_{H-H} = 7.0 Hz, CH_3), 2.1 (3, s, NCH_3), 3.7–4.1 (8, m, piperazine CH_2), 4.4 (2, q, J_{H-H} = 7.0 Hz, CH_2), 7.6 (1, s, ArH), 7.8 (1, d, J_{H-F} = 8.2 Hz, ArH), and 8.9 ppm (1, s, ArH); IR (KBr): 1730 (COOH), 1620 (C=O) cm^{-1}; MS (70ev): *m/z* 333 (M^+).

Results and Discussion

Table II summarizes the in vitro antibacterial activity against gram-positive bacteria (*Staphylococcus aureus* VGH 8445), gram-negative bacteria (*Escherichia coli* CLL 0311), and *Pseudomonas aeruginosa* (LLO 1244).

The results show that when a fluorine substituent is

Table II—In Vitro Antibacterial Activity[a]

Compound	Bacteria		
	S. aureus (VGH 8445)	*E. coli* (LL 0 311)	*P. aeruginosa* (LL 0 1244)
Ciprofloxacin	0.120	0.002	0.250
Ofloxacin	0.120	0.015	1.00
Pefloxacin	0.120	0.060	2.00
6	64	0.120	32
7a	128	128	128
7b	128	128	128
7c	128	128	128
7d	128	128	128
7e	>128	>128	>128
7f	32	>128	>128
7g	128	>128	>128
7h	64	>128	>128
7i	64	>128	>128
7j	128	>128	>128
8	128	128	128

[a] Expressed as μg/mL (ref 4).

present at the 6-position in **6**, moderate antibacterial activity exists, and that replacement of the 6-fluoro substituent by the piperazinyl moiety leads to compounds basically devoid of activity.

This indicates that the presence of a piperazinyl substituent at the 7-position with the fluorine in the 6-position of the aromatic ring is probably essential for the observed potent antibacterial activity of the third generation quinolones.

References and Notes

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2. Wentland, Mark P.; Cornett, James B. *Annu. Rep. Med. Chem.* 1985, *20*, 145–54.
3. Koga, Hiroshi; Itoh, Akira; Murayama, Satoshi; Suzue, Seigo; Irikura, Tsutomu *J. Med. Chem.* 1980, *23*, 1358–1363.
4. National Committee for Clinical Laboratory Study: code M7-A; 1985, *5*, 22.

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