Synthesis and Biological Activity of 5-Amino- and 5-Hydroxyquinolones, and the Overwhelming Influence of the Remote N_1-Substituent in Determining the Structure–Activity Relationship

John M. Domagala,* Alex J. Bridges, Townley P. Culbertson, Laura Gambino, Susan E. Hagen, Gregory Karrick, Kenneth Porter, Joseph P. Sanchez, Josephine A. Sesnie, F. Gregory Spense, DeeDee Szotek, and James Wemple

Parke-Davis Pharmaceutical Research Division, Warner-Lambert Company, Ann Arbor, Michigan 48105.
Received June 29, 1990

A series of 5-amino- and 5-hydroxyquinolone antibacterials substituted at C_7 with a select group of common piperazinyl and 3-aminopyrrolidinyl side chains was prepared. These 5-substituted derivatives were compared to the analogous 5-hydrogen compounds for antiinfective activity by using DNA gyrase inhibition, minimum inhibitory concentrations against a variety of bacteria, and in vivo efficacy in the mouse infection model. The influence on the structure–activity relationships of varied substituents at C_8 (H, F, Cl) and N_1 (ethyl, cyclopropyl, difluorophenyl) was also studied. The results showed that several of the structure–activity conclusions regarding side-chain bulk at C_7, the effect of halogen at C_8, and the effect of the C_5-amino group were greatly influenced by the choice of the N_1-substituent. Several outstanding broad spectrum quinolones were identified in this work. In particular, the spectrum and potency of the 7-piperazinyl quinolones could be greatly enhanced by the judicious choice of C_5-, C_8-, and N_1-substitutents.

With the discovery of norfloxacin[1] **1a–l** (Table II) and its 1,8-naphthyridine analogue enoxacin[2] in the late 1970's, the fluoroquinolone class of antibacterials **1** have generated a great deal of excitement around the world.[3] This is mainly due to the fact that no other class of antibacterials known today offers a greater potential for producing the first truly broad spectrum oral antiinfective with useful activity against Gram-negative, Gram-positive, and anaerobic organisms, and mycobacteria.[4] Almost all of the quinolone research conducted over the last 10 years has been focused on bringing this potential to reality. Much of this research has been the subject of several recent reviews covering: current quinolones approved or being tested in man,[3a,5] structure–activity relationships,[6] the mechanism of action,[7] the known adverse reactions,[8] and overviews of all the above topics.[9]

In general, the optimal substituents identified from previous quinolone SAR studies have been applied to the current day analogues. Typical examples are the widespread utilization of the fluorine at C_6,[1a,6a,d] and the ethyl, cyclopropyl, or fluorophenyl groups at N.[10,11]

Similarily, the great majority of the quinolones currently under development contain the 1-piperazinyl moiety as R_7 in **1**.[1a,3a,6] This group confers excellent Gram-negative potency and generally good therapeutic plasma levels in animals and man.[9,12] We have shown that replacing the

Table I. 5-Substituted-6,7-difluoroquinoline Starting Materials **9** Prepared and Utilized in This Study

compd 9	R_5	R_8	R_1	synthetic ref for known procedures[a]
a	H	H	Et	1a
b	H	F	Et	1a
c	H	Cl	Et	
d	H	H	2,4-diFPh	11
e	H		$OCH_2CH(CH_3)$	19
f	H	H		14
g	H	F		10
h	H	Cl		14
i	NH_2	H	Et	
j	NH_2	F	Et	
k	NH_2	Cl	Et	
l	NH_2	H	2,4-diFPh	
m	NH_2		$OCH_2CH(CH_3)$	
n	NH_2	H		
o	NH_2	F		17a
p	NH_2	Cl		
q	OH	F	Et	
r	OH	H		
s	OH	F		17a

[a] For starting materials **9** not referenced, procedures are included in the experimental section.

(1) (a) Koga, H.; Itoh, A.; Murayama, S.; Suzue, S.; Irikura, T. *J. Med. Chem.* **1980**, *23*, 1358. (b) Holmes, B.; Brodgen, R. N.; Richards, D. M. *Drugs* **1985**, *30*, 482.
(2) Speller, D.; Wise, R. *J. Antimicrob. Chemother.* **1988**, *21* (*Suppl. B*).
(3) (a) Bergan, T. *Antimicrobial Agents Annual 1*; Peterson, P. K., Verhoef, J., Eds.; Elsevier: New York, 1986; Vol. 1, p 164. (b) Neu, H. C. *Antimicrob. Newsl.* **1987**, *4*, 9. (c) Fernandes, P. B.; Chu, D. T. W. *Annu. Rep. Med. Chem.* **1988**, *23*, 133.
(4) Neuman, M.; Esanu, A. *Drugs Expt. Clin. Res.* **1988**, *14*, 385.
(5) (a) Wolfson, J. S.; Hooper, D. *Antimicrob. Agents Chemother.* **1985**, *28*, 581. (b) Fernandes, P. B. *J. Clin. Pharmacol.* **1988**, *28*, 156.
(6) (a) Domagala, J. M.; Hanna, L. D.; Heifetz, C. L.; Hutt, M. P.; Mich, T. F.; Sanchez, J. P.; Solomon, M. *J. Med. Chem.* **1986**, *29*, 394. (b) Stein, G. E. *Pharmacotherapy* **1988**, *8*, 301. (c) Fernandes, P. B.; Chu, D. T. W. *Antimicrob. Agents Chemother.* **1989**, *33*, 131. (d) Koga, H. *Kagaku no Ryoiki Zokan* **1982**, *136*, 177.
(7) (a) Hooper, D. C.; Wolfson, J. S. *Rev. Infect. Dis.* **1989**, *11* (*Suppl. 5*), S902. (b) Drlica, K.; Coughlin, S. *Pharmacol. Ther.* **1989**, *44*, 107. (c) Shen, L. L.; Mitscher, L. A.; Sharma, P. N.; O'Donnell, T. J.; Chu, D. T. W.; Cooper, C. S.; Rosen, T.; Pernet, A. G. *Biochemistry* **1989**, *28*, 3886.
(8) (a) Ball, P. *Rev. Infect. Dis.* **1989**, *11* (*Suppl. 5*), S1365. (b) Christ, W.; Lehnert, T.; Ulbrich, B. *Rev. Infect. Dis.* **1988**, *10* (*Suppl. 1*), S141.
(9) (a) Andriole, V. *The Quinolones*; Academic Press: New York, 1988. (b) Wolfson, J. S.; Hooper, D. C. *Quinolone Antimicrobial Agents*, American Society for Microbiology: Washington DC, 1989. (c) Fernandes, P. B. *International Telesymposium on Quinolones*, J. R. Prous Scientific Publishers: Barcelona, Spain, 1989. (d) Siporin, C.; Heifetz, C. L.; Domagala, J. M. *The New Generation of Quinolones*; Marcel Dekker: New York, 1990.
(10) Domagala, J. M.; Heifetz, C. L.; Hutt, M. P.; Mich, T. F.; Nichols, J. B.; Solomon, M.; Worth, D. F. *J. Med. Chem.* **1988**, *31*, 991.
(11) (a) Chu, D. T. W.; Fernandes, P. B.; Clairborne, A. K.; Maleczka, R. E.; Klock, P.; Shen, L.; Patel, J.; Pernet, A. *26 Interscience Conference on Antimicrobial Agents and Chemotherapy*, New Orleans, LA, 1986 Abstr. 428. (b) Chu, D. T. W.; Fernandes, P. B.; Clairborne, A. K.; Pihuleac, E.; Nordeen, C. W.; Maleczka, R. E.; Pernet, A. G. *J. Med. Chem.* **1985**, *28*, 1558. (c) Chu, D. T. W. *J. Heterocycl. Chem.* **1985**, *22*, 1033.

0022-2623/91/1834-1142$02.50/0

1-piperazinyl moiety with a 1-(3-amino or 3-aminomethyl) pyrrolidinyl ring greatly enhanced Gram-positive potency, providing the first group of broad spectrum quinolones with efficacy against *Staphylococcus* and *Streptococcus*.[13]

As newer work was published, certain exceptions to the accepted SARs began to appear. Sanchez and co-workers,[14] in a series of N_1-cyclopropyl analogues, demonstrated that an 8-halogen improved in vitro Gram-positive potency significantly. Meanwhile, when N_1 was difluorophenyl, Chu reported quite opposite effects.[15] Recently, a series of 5-amino-1-cyclopropylquinolones was reported with greatly enhanced Gram-positive potency, especially when the 1-piperazinyl side chain was at R_7.[16,17] These new derivatives represent another class of broad spectrum quinolones. Several literature accounts however, implied that substitution at R_5 for several types of substituents was detrimental.[6a,6d,17a,18]

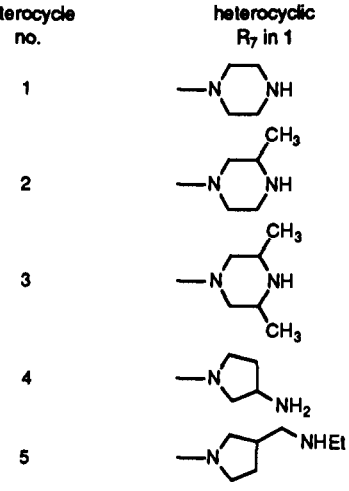
In this and the following paper, we wish to share a more detailed and extensive account of the nature of the 5-substituent (NH_2, OH, CH_3) as a function of the substituents at C_7, C_8, and N_1, and the surprising results which indicate that the conclusions of previous structure–activity relationships may have been overly influenced by the N_1-substituent chosen. We also wish to corroborate and rationalize the results published by earlier workers regarding the 5-substituent.

Chemistry

All of the 5-substituted quinolone precursors **9** were prepared according to Scheme I and are listed in Table I along with the 5-unsubstituted precursors for comparison purposes. The quinolone ring formation from the corresponding benzoic acid is well documented[10,11,14] and is unimpeded by the presence of the latent 5-substituent. All of the 5-amino derivatives, **9i–k,o,p**, were prepared beginning with the nitrobenzoic acid **4** ($R_5 = NO_2$). For the 5-amino quinolones, **9i,l,n**, which have a hydrogen at position 8, the amino group was introduced by the nucleophilic displacement of the *o*-fluorine in **3n** with benzylamine. The selectivity of this displacement was greater than 90:10 and the impurities were readily removed. The 5-hydroxy quinolones, **9q–s**, were prepared in a similar manner displacing the *o*-fluorine of **3r** with methanol. Compound **9s** was initially obtained with a 7-methoxy impurity, which was separated from the final product.

For the purposes of this study, a limited number of heterocyclic side chains were employed for the R_7 substituent. These side chains are shown in Chart I. The nucleophilic addition of the side chains to the quinolone nucleus to form the final products 1 has been extensively described.[6a,14] The physical properties or references for all the quinolones tested in this study are given in Table II. The numbering system employed in Table II and throughout this manuscript links the quinolone nucleus letter code (a, b, c, etc.) with the side chain number from Chart I (1–5).

Chart I. Heterocyclic Side Chains Employed as the R_7 Substituent of 1 in This Study



Biological Assays

The quinolones 1 (Table II) were tested against 10 representative Gram-positive and Gram-negative organisms by using standard microtitration techniques,[13b] and their minimum inhibitory concentrations (MICs, μg/mL) were averaged from multiple experiments and recorded in Table III. To greatly simplify the data in the search for trends among compounds, the geometric mean of the MICs of the Gram-negative organisms and the Gram-positive organisms were calculated. These data, grouped by structural type are recorded in Tables IV–VI. The compounds were also tested for their inhibition of DNA gyrase which was isolated and purified from *Escherichia coli* H560.[6a] The initial cleavage assay was employed, which gives the lowest concentration of drug (μg/mL) that will cleave relaxed bacterial Col E1 plasmid DNA.[6a] The cleaved DNA was visualized by agarose gel electrophoresis and staining with ethidium bromide. The gyrase data is also the result of multiple experiments and are generally accurate to ±50% or one dilution.

The in vivo potency (Table VII), expressed as the median protective dose (PD_{50}, mg/kg), was determined in acute, lethal systemic infections in female Charles River CD-1 mice (16 mice per method of administration) as previously described.[16] Single doses of compound were administered with challenge.

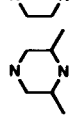
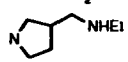
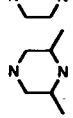
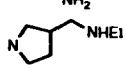
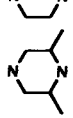
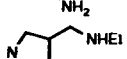
Results and Discussion

In light of the earlier literature, which indicated that 5-substitution was detrimental, we decided that our study of the nature of the C_5-substituent must include the commonly employed C_8-groups, H, F, and Cl, and the ethyl and cyclopropyl groups at N_1. In addition we included a few data points employing a difluorophenyl at N_1 and the ofloxacin (benzoxazine) nucleus. Vinyl, fluoroethyl, and cyclobutyl N_1-substituents were used to check the trends (data not included). For the side chain at C_7, we decided to employ the most common piperazines (1–3, Chart I) and pyrrolidines (4,5) described in the literature.

Effect of Alkylation on the Side Chain. Studying the effect of alkylation on the side chain was not our initial

(12) (a) Nix, D. E.; Schentag, J. J. *J. Clin. Pharmacol.* **1988**, *28*, 169. (b) Gerding, D. N.; Hitt, J. A. *Rev. Infect. Dis.* **1989**, *11* (*Suppl. 5*), S1046.
(13) (a) Domagala, J. M.; Heifetz, C. L.; Mich, T. F.; Nichols, J. B. *J. Med. Chem.* **1986**, *29*, 445. (b) Cohen, M. A.; Griffin, T. J.; Bien, P. A.; Heifetz, C. L.; Domagala, J. M. *Antimicrob. Agents Chemother.* **1985**, *28*, 766.
(14) Sanchez, J. P.; Domagala, J. M.; Hagen, S. E.; Heifetz, C. L.; Hutt, M. P.; Nichols, J. B.; Trehan, A. K. *J. Med. Chem.* **1988**, *31*, 983.
(15) Chu, D. T. W.; Fernandes, P. B.; Maleczka, R. E.; Nordeen, C. W.; Pernet, A. G. *J. Med. Chem.* **1987**, *30*, 504.
(16) Domagala, J. M.; Hagen, S. E.; Heifetz, C. L.; Hutt, M. P.; Mich, T. F.; Sanchez, J. P.; Trehan, A. K. *J. Med. Chem.* **1988**, *31*, 503.
(17) Miyamoto, T.; Matsumoto, J.; Chiba, K.; Egawa, H.; Shibamori, K.; Minamida, A.; Nishimura, Y.; Okada, H.; Kataoka, M.; Fujita, M.; Hirose, T.; Nikano, J. *J. Med. Chem.* **1990**, *33*, 1645. (b) Nakamura, S.; Akira, M.; Nakata, K.; Kurobe, N.; Kouno, K.; Sakaguchi, Y.; Kashimoto, S.; Yoshida, H.; Kojima, T. *Antimicrob. Agents Chemother.* **1989**, *33*, 1167.
(18) Verhist, L. *Pharm. Weekly, Sci. Ed.* **1986**, *8*, 22.

Table II. Physical Properties (or Literature Reference) for All Quinolones Tested in This Study

compd no.	R_5	R_8	R_1	R_7	method of preparation[a] base[b]/solvent/temp	method of purification[c] of final product (of intermediate)	MP or dec, °C	% yield of 1 from 9	analysis empirical form (analyzed elements)[d]
1a-1	H	H	Et		ref 1a				
1a-2	H	H	Et		-/pyridine/reflux	1. conc, EtOH wash 2. aqueous HCl, 2-PrOH	>300	66	$C_{17}H_{20}FN_3O_3 \cdot 1.3HCl$ (C,H,N,Cl)
1a-3	H	H	Et		-/pyridine/reflux	isoelect prec	207–208	68	$C_{18}H_{22}FN_3O_3 \cdot 1.6H_2O$ (C,H,N)
1a-4	H	H	Et		1. -/pyridine/reflux 2. HCl, EtOH	1. CH_3CN wash 2. isoelect prec	281–283	53	$C_{16}H_{18}FN_3O_3 \cdot 0.25H_2O$
1a-5	H	H	Et		ref 13a				
1b-1	H	F	Et		refs 1a, 6a				
1b-2	H	F	Et		-/CH_3CN/reflux	isoelect prec	237–240	85	$C_{17}H_{19}F_2N_3O_3 \cdot 0.8H_2O$ (C,H,N,F,H_2O)
1b-3	H	F	Et		Et_3N/CH_3CN/reflux	isoelect prec	231–233	89	$C_{18}H_{21}F_2N_3O_3 \cdot 0.8H_2O$ (C,H,N)
1b-4	H	F	Et		1. Et_3N/CH_3CN/reflux[e] 2. TFA	1. CH_3CN wash 2. isoelect prec	243–245	77	$C_{16}H_{17}F_2N_3O_3$ (C,H,N)
1b-5	H	F	Et		ref 13a				
1c-1	H	Cl	Et		-/DMF/60 °C	prec with HCl in THF, THF wash	285–289	88	$C_{16}H_{17}ClF_3O_3 \cdot HCl$ (C,H,N)
1c-2	H	Cl	Et		-/DMF/60 °C	isoelect prec	>250	76	$C_{17}H_{19}ClFN_3O_3 \cdot 4.0H_2O$ (C,H,N,H_2O)
1c-3	H	Cl	Et		-/DMF/60 °C	prec with HCl in THF, THF wash	287–292	85	$C_{18}H_{21}ClFN_3O_3 \cdot HCl$ (C,H,N)
1c-4	H	Cl	Et		1. Et_3N/DMF/60 °C 2. HCl EtOH	1. concentration 2. EtOH, THF wash	218–288	74	$C_{16}H_{17}ClFN_3O_3 \cdot HCl \cdot 2.25H_2O$ (C,H,N,Cl)
1c-5	H	Cl	Et		-/DMF/60 °C	isoelect prec	230–232	85	$C_{19}H_{23}ClFN_3O_3 \cdot 0.25H_2O$ (C,H,N)
1d-2	H	H	2,4-diFPh		ref 11a				

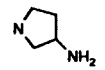
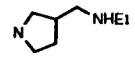
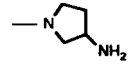
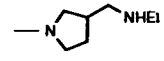
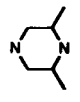
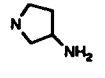
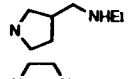
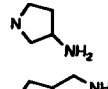
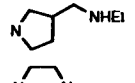
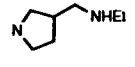
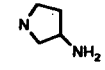
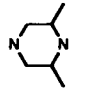
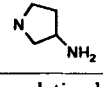
1e-4	H				ref 14			
1e-5	H				ref 14			
1f-1	H	H			ref 14			
1f-2	H	H			-/pyridine/reflux	isoelect prec	235–237	100	$C_{18}H_{20}FN_3O_3$ (C,H,N)
1f-3	H	H			-/pyridine/reflux	H_2O, ether wash	242–243	100	$C_{19}H_{22}FN_3O_3$
1f-4	H	H			ref 14			
1f-5	H	H			ref 14			
1g-1	H	F			ref 14			
1g-2	H	F			DBU/CH_3CN/reflux	EtOH wash	222–225	74	$C_{18}H_{19}F_2N_3O_3 \cdot 0.66H_2O$ (C,H,N)
1g-3	H	F			DBU/CH_3CN/reflux	concentration, prec from 2-PrOH	232–234	68	$C_{19}H_{21}F_2N_3O_3 \cdot 0.25H_2O$ (C,H,N,H_2O)
1g-4	H	F			ref 14			
1g-5	H	F			refs 10, 14			
1h-1	H	Cl			ref 14			
1h-2	H	Cl			Et_3N/CH_3CN/reflux	filter, H_2O, pH 11, HCl pH 2, freeze dry ether wash	>300	30	$C_{18}H_{19}ClFN_3O_3 \cdot 1.1HCl \cdot 1.1H_2O$ (C,H,N,Cl)
1h-4	H	Cl			ref 14			
1h-5	H	Cl			ref 14			
1i-1	NH_2	H			-/CH_3CN/reflux	CH_3CN, H_2O wash	235–237	86	$C_{16}H_{19}FN_4O_3 \cdot 2.3H_2O$ (C,H,N)
1i-5	NH_2	H			Et_3N/CH_3CN/reflux	isoelect prec	220–222	38	$C_{19}H_{25}FN_4O_3 \cdot 1H_2O$ (C,H,N)

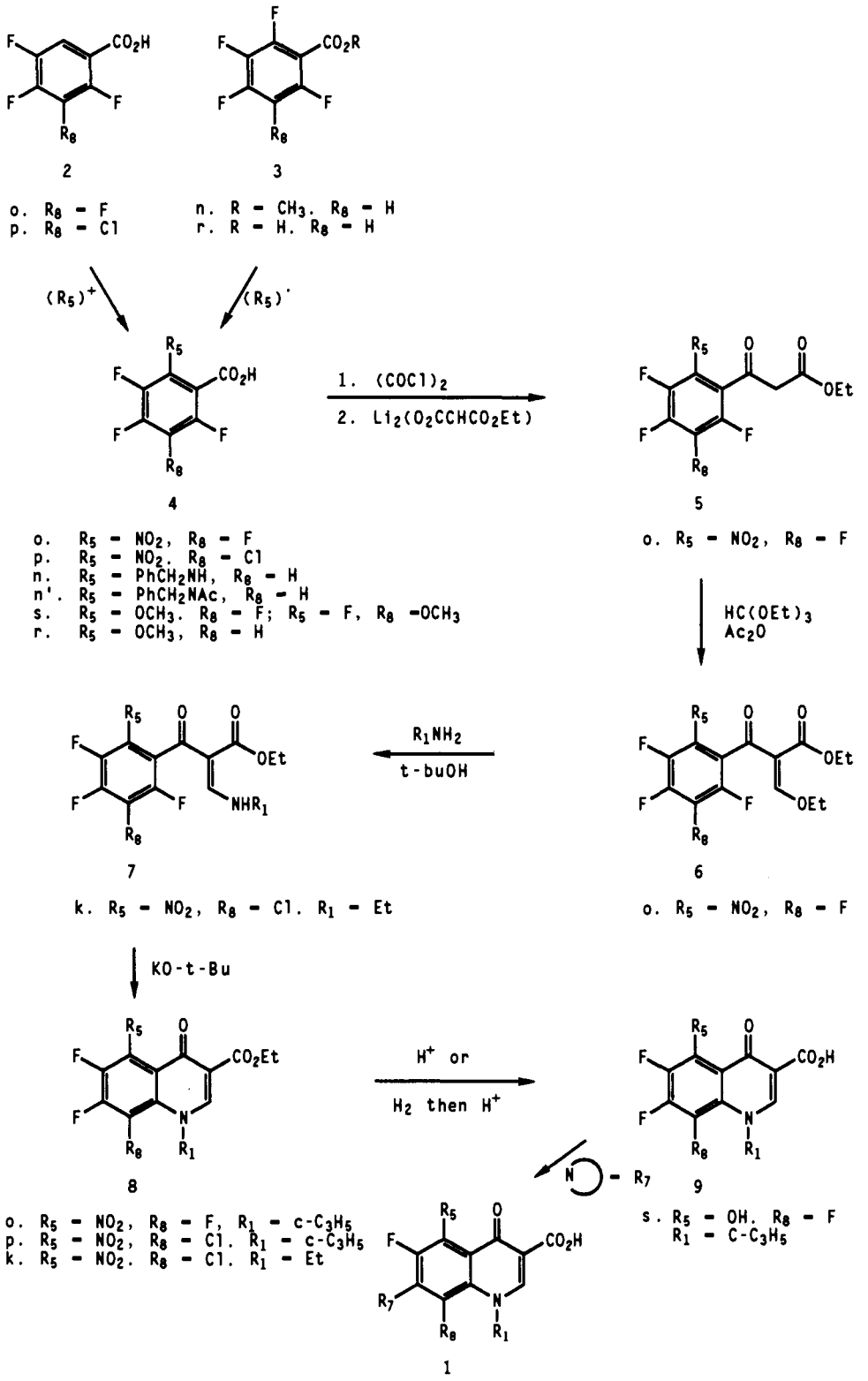
Table II (Continued)

compd no.	R_5	R_8	R_1	R_7	method of preparation[a] base[b]/solvent/temp	method of purification[c] of final product (of intermediate)	MP or dec, °C	% yield of 1 and 9	analysis empirical form (analyzed elements)[d]
1j-1	NH_2	F	Et		-/CH_3CN/reflux	H_2O, EtOH wash	255–258	84	$C_{16}H_{18}F_2N_4O_3 \cdot 0.3H_2O$ (C,H,N)
1j-2	NH_2	F	Et		-/CH_3CN/reflux	CH_3CN wash	240–250	91	$C_{17}H_{20}F_2N_4O_3 \cdot 0.5H_2O$ (C,H,N)
1j-3	NH_2	F	Et		-/CH_3CN/reflux	CH_3CN wash	250–257	76	$C_{18}H_{22}F_2N_4O_3 \cdot 1.25H_2O$ (C,H,N)
1j-4	NH_2	F	Et		Et_3N/CH_3CN/reflux	2-PrOH wash	265–280	89	$C_{16}H_{19}F_2N_4O_3 \cdot HCl \cdot 0.5H_2O$ (C,H,N)
1j-5	NH_2	F	Et		Et_3N/CH_3CN/reflux	EtOH ether wash	194–196	78	$C_{19}H_{24}F_2N_4O_3 \cdot 0.1H_2O$ (C,H,N)
1k-1	NH_2	Cl	Et		1. -/CH_3CN/80 °C/ 2. LiOH/THF	1. $H_2O/CHCl_3$ extraction 2. isoelect prec	210–212	50	$C_{16}H_{18}ClFN_4O_3 \cdot 1.2H_2O$ (C,H,N)
1k-3	NH_2	Cl	Et		1. -/DMSO/95 °C/ 2. LiOH/THF	1. Si gel chrom recryst EtOAc 2. EtOH wash	268–275	44	$C_{18}H_{22}ClFN_4O_3 \cdot Li \cdot 0.125$ H_2O (C,H,N)
1k-5	NH_2	Cl	Et		-/CH_3CN/reflux	H_2O wash	177–179	90	$C_{19}H_{24}ClFN_4O_3$ (C,H,N)
1l-2	NH_2	H	2,4-diFPh		-/pyridine/reflux	isoelect prec prec HCl EtOH	293–295	71	$C_{21}H_{19}F_3N_4O_3 \cdot HCl$ (C,H,N)
1m-4	NH_2				1. Et_3N/pyridine/reflux[e] 2. HCl AcOH	1. isoelect prec 2. concentration, prec from EtOH	275–276	60	$C_{17}H_{19}FN_4O_4 \cdot HCl \cdot 0.5H_2O$ (C,H,N)
1m-5	NH_2				-/CH_3CN/reflux	prec HCl MeOH	280–281	70	$C_{20}H_{25}FN_4O_4 \cdot HCl$ (C,H,N)
1n-1	NH_2	H			-/pyridine/reflux	conc prec from CH_3CN	212–216	64	$C_{17}H_{19}FN_4O_3 \cdot 0.4H_2O$ (C,H,N)
1n-2	NH_2	H			-/pyridine/reflux	1. concentration, prec from EtOH 2. isoelect prec	187–188	56	$C_{18}H_{21}FN_4O_3$ (C,H,N)
1n-4	NH_2	H			1. Et_3N/pyridine/reflux 2. HCl EtOH	EtOH wash	>300	77	$C_{17}H_{19}FN_4O_3 \cdot HCl$ (C,H,N,Cl)
1n-5	NH_2	H			Et_3N/pyridine/reflux	pH 6.8, EtOH to prec, EtOH wash	>300	33	$C_{20}H_{25}FN_4O_3 \cdot 1.5NaCl$ (C,H,N,Cl)
1o-1	NH_2	F			ref 16				
1o-2	NH_2	F			Et_3N/CH_3CN/reflux	isoelect prec	245–250	71	$C_{18}H_{20}F_2N_4O_3$ (C,H,N)

1o-3	NH_2	F		Et_3N/CH_3CN/reflux	isoelect prec	260–265	70	$C_{19}H_{22}F_2N_4O_3$ (C,H,N,F)
1o-4	NH_2	F		ref 16				
1o-5	NH_2	F		ref 16				
1p-1	NH_2	Cl		1. DBU/DMF/60 °C[g] 2. LiOH	1. Si gel chrom 2. isoelect prec	248–260	21	$C_{17}H_{18}ClFN_4O_3 \cdot 0.5HCl \cdot 1H_2O$ (C,H,N,Cl)
1p-2	NH_2	Cl		1. -/DMSO/95 °C[g] 2. LiOH THF	1. Si gel chrom recryst EtOAc 2. isoelect prec	199–202	35	$C_{18}H_{20}ClFN_4O_3 \cdot 1.25H_2O$ (C,H,N)
1p-3	NH_2	Cl		-/DMSO/95 °C	isoelect prec	183–283	37	$C_{19}H_{22}ClFN_4O_3 \cdot H_2O$ (C,H,N)
1p-4	NH_2	Cl		Et_3N/CH_3CN/reflux	isoelect prec	128–210	46	$C_{17}H_{18}ClFN_4O_3 \cdot 0.66H_2O$ (C,H,N)
1p-5	NH_2	Cl		-/CH_3CN/reflux	MeOH/H_2O wash	161–164	48	$C_{20}H_{24}ClFN_4O_3 \cdot 0.75H_2O$ (C,H,N)
1q-1	OH	F	Et 	-/CH_3CN/reflux	CH_3CN wash	251–253	58	$C_{16}H_{17}F_2N_3O_4 \cdot 1.3H_2O$ (C,H,N)
1q-2	OH	F	Et 	Et_3N/CH_3CN/reflux	CH_3CN wash	265–268	96	$C_{17}H_{19}F_2N_3O_4 \cdot 0.4H_2O$ (C,H,N)
1q-3	OH	F	Et 	Et_3N/CH_3N/reflux	CH_3CN wash	279–281	85	$C_{18}H_{21}F_2N_3O_4 \cdot 0.5H_2O$ (C,H,N)
1q-4	OH	F	Et 	1. Et_3N/CH_3CN/reflux 2. AcOH, HCl	isoelect prec	258–261	83	$C_{16}H_{17}F_2N_3O_4$ (C,H,N)
1q-5	OH	F	Et 	Et_3N/CH_3CN/reflux	CH_3CN wash	264–266	85	$C_{19}H_{23}F_2N_3O_4 \cdot 0.6H_2O$ (C,H,N)
1r-1	OH	H		-/CH_3CN/reflux	isoelect prec	273–277	44	$C_{17}H_{18}FN_3O_4$ (C,H,N)
1r-4	OH	H		1. DBU/CH_3CN/reflux 2. EtOH HCl	EtOH/ether wash	302–304	48	$C_{17}H_{18}FN_3O_4 \cdot HCl \cdot 1.4H_2O$ (C,H,N,Cl)
1s-1	OH	F		-/CH_3CN/reflux	CH_3CN wash	259–260	68	$C_{17}H_{17}F_2N_3O_4 \cdot 0.9H_2O$ (C,H,N)
1s-2	OH	F		-/CH_3CN/reflux	isoelect prec	272–274	50	$C_{18}H_{19}F_2N_3O_4 \cdot 1.5H_2O$ (C,H,N)
1s-4	OH	F		Et_3N/CH_3CN/reflux	trit with 2-PrOH	>300	74	$C_{17}H_{17}F_2N_3O_4 \cdot HCl \cdot 1.2H_2O$ (C,H,N,Cl)

[a] Reactions were monitored for completion by TLC or HPLC. [b] Where no base is given, excess side chain was employed as base. [c] Isoelectric precipitation (isoelect prec) includes: crude product (reaction concentrate or solids) is dissolved at pH 12, filtered, acidified with HCl to pH 6–8. Precipitate is washed with water and then dried to constant weight. [d] In addition to analytical data, all samples where >97% pure by HPLC. [e] The 3-[(*tert*-butoxycarbonyl)amino]pyrrolidine was employed as side chain requiring an acid deprotection step. [f] The ethyl ester of **9k** was employed as starting material. [g] The ethyl ester of **9o** was employed as starting material.

Scheme I



intention, but the data in Tables IV and V requires this issue to be addressed. Reading down any column in Tables IV and V reveals the effect of the side chain on a particular quinolone nucleus. Reading across any row reveals the effects of modification at C_5, C_8, and N_1 for a given side chain. It has been widely published that bulk in the side chain at R_7 is detrimental to a quinolone's in vitro potency.[6a,20,21] Our results in Tables IV and V corroborate this loss of quinolone potency with increasing piperazine alkylation when N_1 is ethyl (series a–c and i–k). The

(19) Hayakawa, I.; Tanaka, Y.; Hiramita, T. Eur. Pat. 0,047,005, 1982; *Chem. Abstr.* **1982**, *97*, 55821b.

(20) Culbertson, T. P.; Domagala, J. M.; Hagen, S. E.; Hutt, M. P.; Nichols, J. B.; Mich, T. F.; Sanchez, J. P.; Schroeder, M. C.; Solomon, M.; Worth, D. F. *International Telesymposium on Quinolones*; Fernandes, P. B., Ed.; J. R. Prous: Spain, 1989; p 47.

(21) Matsumoto, J.; Miyamoto, T.; Minamida, Y.; Egawa, H.; Nishimura, H. *Current Therapy and Infectious Disease*; Nelson, J. D., Grassi, C., Eds.; American Society for Microbiology: Washington DC, 1980; Vol. 1, p 454.

Table III. Biological Testing Results from the Antibacterial Screen and the DNA Gyrase Supercoiling Inhibition Assay

	antibacterial activity (MICs),[a] μg/mL										
	Gram-negative organisms					Gram-positive organisms					gyrase DNA
						S. aureus					
compd no.	*E. cloac* MA 2646	*E. coli* Vogel	*K. pneum* MGH-2	*P. rettg* M1771	*P. aerug* UI-18	H 228	UC 76	*S. faecalis* MGH-2	*S. pneum* SV-1	*S. pyog* C 203	cleavage,[b] μg/mL
1a-1	0.1	0.025	0.05	0.025	0.2	0.8	0.05	1.6	1.6	0.8	1.0
1a-2	0.1	0.1	0.2	0.2	1.6	1.6	0.2	1.6	1.6	1.6	2.5
1a-3	0.2	0.2	0.4	0.8	6.3	1.6	0.4	1.6	1.6	1.6	2.5
1a-4	0.1	0.1	0.2	0.2	0.8	0.8	0.1	0.8	0.4	0.4	2.5
1a-5	0.4	0.1	0.4	1.6	3.1	0.2	0.1	0.2	0.2	0.1	2.5
1b-1	0.1	0.1	0.1	0.2	0.8	1.6	0.4	1.6	3.1	3.1	2.5
1b-2	0.1	0.1	0.2	0.4	1.6	1.6	0.4	3.1	1.6	3.1	2.5
1b-3	0.2	0.2	0.4	1.6	12.5	1.6	0.8	6.3	6.3	6.3	3.8
1b-4	0.1	0.1	0.2	0.2	0.4	0.4	0.1	0.4	0.4	0.2	2.5
1b-5	0.1	0.1	0.2	0.4	1.6	0.1	0.05	0.1	0.1	0.1	2.5
1c-1	0.1	0.1	0.2	0.4	3.1	0.4	0.2	3.1	3.1	3.1	2.5
1c-2	0.2	0.2	0.4	0.8	6.3	1.6	0.4	3.1	3.1	3.1	5.0
1c-3	3.1	1.6	3.1	6.3	12.5	3.1	1.6	12.5	12.5	12.5	5.0
1c-4	0.05	0.05	0.1	0.2	0.8	0.4	0.1	0.4	0.4	0.4	2.5
1c-5	0.2	0.2	0.4	0.8	12.5	0.1	0.025	0.1	0.05	0.05	2.5
1d-2	0.05	0.05	0.1	0.2	1.6	0.1	0.025	0.2	0.1	0.1	0.75
1e-4	0.4	0.4	0.4	0.8	0.4	1.6	0.1	1.6	0.8	0.2	0.75
1e-5	0.2	0.2	0.4	0.8	0.8	0.2	0.025	0.1	0.1	0.013	0.5
1f-1	0.013	0.013	0.05	0.05	0.4	1.6	0.2	0.8	0.8	0.8	0.5
1f-2	0.025	0.025	0.05	0.1	0.4	0.4	0.1	0.4	0.2	0.2	0.5
1f-3	0.025	0.025	0.05	0.2	0.4	0.2	0.05	0.2	0.2	0.2	1.0
1f-4	0.025	0.025	0.05	0.1	0.2	0.1	0.025	0.1	0.1	0.1	0.25
1f-5	0.4	0.2	0.2	0.8	1.6	0.4	0.013	0.2	0.2	0.05	1.0
1g-1	0.05	0.05	0.05	0.1	0.2	0.4	0.1	0.4	0.8	0.8	0.5
1g-2	0.05	0.025	0.025	0.1	0.4	0.2	0.1	0.4	0.4	0.4	1.0
1g-3	0.05	0.05	0.1	0.2	0.4	0.2	0.05	0.4	0.4	0.4	2.5
1g-4	0.013	0.013	0.025	0.05	0.1	0.05	0.013	0.1	0.05	0.05	0.1
1g-5	0.1	0.05	0.1	0.2	0.4	0.05	0.013	0.025	0.025	0.025	0.25
1h-1	0.025	0.025	0.05	0.1	0.4	0.1	0.05	0.4	0.1	0.2	0.5
1h-2	0.025	0.025	0.05	0.2	0.8	0.2	0.05	0.2	0.2	0.2	2.5
1h-4	0.025	0.025	0.025	0.025	0.05	0.025	0.013	0.05	0.025	0.05	0.5
1h-5	0.05	0.05	0.1	0.2	0.8	0.013	0.003	0.013	0.003	0.006	0.25
1i-1	0.2	0.2	0.4	1.6	3.1	0.4	0.05	0.2	0.1	0.05	2.5
1i-5	1.6	0.8	3.1	12.5	25	0.8	0.1	0.8	0.4	0.8	7.5
1j-1	0.2	0.1	0.2	0.4	0.8	1.6	0.2	1.6	1.6	3.1	2.5
1j-2	0.2	0.2	0.4	1.6	3.1	1.6	0.4	3.1	3.1	6.3	7.5
1j-3	0.8	0.4	0.8	3.1	12.5	1.6	0.8	6.3	12.5	25	7.5
1j-4	0.2	0.1	0.2	0.4	1.6	0.4	0.2	0.8	0.8	0.8	2.5
1j-5	0.4	0.2	1.6	1.6	6.3	0.2	0.1	0.2	0.2	0.2	3.8
1k-1	0.4	0.2	0.4	1.6	6.3	3.1	0.4	6.3	3.1	3.1	5.0
1k-3	1.6	1.6	6.3	6.3	12.5	3.1	1.6	12.5	25	25	7.5
1k-5	3.1	1.6	6.3	12.5	25	1.6	0.4	1.6	1.6	6.3	2.5
1l-2	0.2	0.1	0.2	0.4	1.6	0.2	0.05	0.4	0.2	0.4	2.5
1m-4	0.8	0.4	0.4	1.6	1.6	0.8	0.2	0.8	0.8	1.6	2.5
1m-5	1.6	0.8	0.8	3.1	12.5	0.8	0.1	0.4	0.4	0.1	7.5
1n-1	0.013	0.013	0.025	0.025	0.8	0.1	0.025	0.1	0.1	0.2	1.0
1n-2	0.025	0.013	0.025	0.1	0.4	0.2	0.05	0.2	0.1	0.2	0.5
1n-4	0.013	0.013	0.025	0.05	0.4	0.1	0.025	0.05	0.025	0.05	2.0
1n-5	0.1	0.1	0.2	0.8	0.8	0.2	0.025	0.025	0.05	0.1	1.0
1o-1	0.013	0.013	0.025	0.05	0.025	0.05	0.013	0.05	0.05	0.1	0.5
1o-2	0.013	0.013	0.025	0.05	0.4	0.05	0.025	0.1	0.05	0.2	1.0
1o-3	0.025	0.025	0.05	0.2	0.8	0.05	0.013	0.1	0.05	0.1	2.5
1o-4	0.006	0.006	0.013	0.025	0.1	0.05	0.013	0.05	0.025	0.025	0.5
1o-5	0.05	0.05	0.05	0.1	0.2	0.013	0.003	0.025	0.003	0.013	0.5
1p-1	0.025	0.025	0.05	0.1	0.8	0.1	0.025	0.1	0.1	0.2	1.0
1p-2	0.05	0.025	0.1	0.2	0.8	0.05	0.025	0.1	0.05	0.1	3.8
1p-3	0.1	0.05	0.2	0.4	1.6	0.1	0.025	0.4	0.1	0.2	
1p-4	0.025	0.025	0.05	0.1	0.8	0.1	0.025	0.1	0.1	0.1	
1p-5	0.2	0.1	0.4	0.8	1.6	0.1	0.025	0.1	0.025	0.025	1.0
1q-1	0.4	0.4	0.8	1.6	1.6	12.5	1.6	25	25	25	5.0
1q-2	0.4	0.4	0.8	1.6	12.5	6.3	1.6	12.5	12.5	12.5	5.0
1q-3	0.8	0.8	1.6	6.3	12.5	6.3	3.1	25	50	50	5.0
1q-4	0.2	0.2	0.4	0.4	0.8	1.6	0.2	3.1	1.6	1.6	2.5
1q-5	0.4	0.4	0.8	3.1	12.5	0.4	0.05	0.4	0.2	0.2	2.5
1r-1	0.05	0.025	0.1	0.1	0.4	3.1	0.2	0.8	0.8	0.4	1.0
1r-4	0.025	0.013	0.025	0.05	0.2	0.1	0.013	0.2	0.05	0.1	1.0
1s-1	0.1	0.05	0.2	0.2	0.8	1.6	0.2	1.6	1.6	1.6	0.5
1s-2	0.1	0.05	0.2	0.2	1.6	0.8	0.1	0.8	0.4	0.4	0.5
1s-4	0.013	0.013	0.025	0.05	0.2	0.1	0.025	0.1	0.05	0.05	0.5

[a] Minimum inhibitory concentration (ref 13b). [b] Concentration required to cause the first observable cleavage of circular DNA relative to oxolinic acid at 10 μg/mL (ref 6a).

Table IV. Mean MICs (Gram-Negative/Gram-Positive, μg/mL) for All 5-Hydrogen Analogues

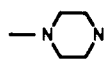
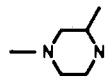
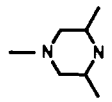
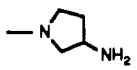
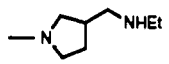
heterocycle R_7		quinolone nucleus with substituents: R_5, R_8, R_1							
		1a: H,H,Et	**1b**: H,F,Et	**1c**: H,Cl,Et	**1f**: H,H,c-C_3H_5	**1g**: H,F,c-C_3H_5	**1h**: H,Cl,c-C_3H_5	**1d**: H,H,$F_2C_6H_4$	**1e**: H,Oflox
	1	0.057/0.61	0.17/1.58	0.30/1.19	0.044/0.70	0.076/0.40	0.066/0.13		
	2	0.23/1.05	0.26/1.58	0.60/1.80	0.066/0.23	0.066/0.26	0.087/0.15	0.15/0.087	
	3	0.60/1.21	0.79/3.17	4.14/6.27	0.076/0.15	0.11/0.23			
	4	0.13/0.40	0.17/0.27	0.13/0.30	0.057/0.075	0.029/0.044	0.029/0.029		0.46/0.53
	5	0.60/0.15	0.26/0.087	0.69/0.057	0.46/0.10	0.13/0.025	0.13/0.006		0.40/0.057

Table V. Mean MICs (Gram-Negative/Gram-Positive, μg/mL) for All 5-Amino Analogues

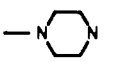
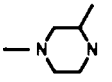
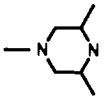
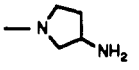
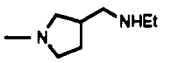
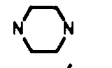
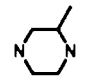
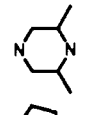
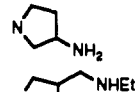
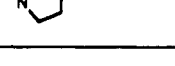
heterocycle R_7		quinolone nucleus with substituents: R_5, R_8, R_1							
		1i: NH_2,H,Et	**1j**: NH_2,F,Et	**1k**: NH_2,Cl,Et	**1n**: NH_2,H,c-C_3H_5	**1o**: NH_2,F,c-C_3H_5	**1p**: NH_2,Cl,c-C_3H_5	**1l**: NH_2,H,$F_2C_6H_4$	**1m**: NH_2, oflox
	1	0.60/0.11	0.26/1.15	0.79/2.37	0.038/0.087	0.022/0.044	0.076/0.087		
	2		0.60/2.07		0.058/0.13	0.038/0.066	0.11/0.057	0.30/0.20	
	3		1.58/4.79	4.18/8.27		0.087/0.050	0.23/0.11		
	4		0.30/0.53		0.038/0.044	0.016/0.029	0.076/0.076		0.80/0.70
	5	4.15/0.46	1.05/0.17	6.28/1.59	0.26/0.057	0.075/0.0082	0.40/0.044		2.09/0.26

Table VI. Mean MICs (Gram-Negative/Gram-Positive μg/mL) for All 5-Hydroxy Analogues

heterocycle R_7		quinolone nucleus with substituents: R_5, R_8, R_1		
		1q: OH,F,Et	**1r**: OH,H,c-C_3H_5	**1s**: OH,F,c-C_3H_5
	1	0.80/12.56	0.087/0.69	0.17/1.06
	2	1.21/7.23		0.20/0.40
	3	2.41/16.49		
	4	0.35/1.20	0.038/0.066	0.034/0.057
	5	1.38/0.20		

Gram-negative means show a 5- to 14-fold decrease in potency (for example **1c–1** vs **1c–3**), while the Gram-positive means fall by 2- to 5-fold. But when the cyclopropyl moiety is employed at N_1 (series f–h and n–p), the Gram-negative and Gram-positive potency barely change with additional piperazinyl alkylation and in a few examples the Gram-positive activity actually increases (**1f–3** vs **1f–1**)! In each case, a single alkylation shows an intermediate effect. The alkylation trends are mostly independent of the C_8-substituent. At the enzyme level (Table III), alkylation of the piperazine causes a very small but consistent diminution of gyrase inhibition. Indeed, it has been shown quantitatively that DNA gyrase has a relatively broad tolerance for steric bulk.[20] In conclusion, the effect of alkylation of the piperazine on in vitro antibacterial activity is primarily a function of the choice of the N_1-substituent. When N_1 is ethyl, the alkylated piperazines show a significant decrease in potency. When N_1 is cyclopropyl, in vitro potency was not significantly affected. In every case the cyclopropyl group was superior to the ethyl at N_1.

The Effect of the C_8-Substituent. When N_1 is ethyl, adding a halogen to the 8-position of the 7-piperazinyl analogues is detrimental (reading across series a–c in Table IV). The lost activity ranges from no change to a 7-fold decrease. This result is in direct conflict with those reported by Koga,[6d] but is readily explainable by the fact that Koga developed his SAR using a single organism. For the pyrrolidines at R_7, adding a halogen to C_8 is neutral for the 3-aminopyrrolidinyl analogues **4**, and beneficial for the [(ethylamino)methyl]pyrrolidinyl analogues **5** (**1a–5** vs **1b–5** or **1c–5**, and **1i** vs **1j**). In fact, the addition of an 8-fluoro group was required to make the pyrrolidinyl

Table VII. In Vivo Efficacy in Mouse Protection Tests (PD_{50})

compd series	substituents: R_5, R_8, R_1	PD_{50}, mg/kg[a,b]					
		E. coli		*Streptococci*			
				pyogenes		*pneumoniae*	
		PO	SC	PO	SC	PO	SC
1f-1	H, H, 	1	0.3	>100	19	>100	28
1f-2		1	0.4	39	12		
1f-3		0.7	0.4	15	12		
1f-4		3	0.5	43	4	97	11
1f-5		35	2				
1n-1	NH_2, H, 	4	0.3				
1n-2		2	0.4	66	21		
1n-4		8	0.7	110	6		
1n-5		90	3				
1g-1	H, F, 	0.5	0.3	59	29	33	9
1g-2		0.8	0.4				
1g-3		0.8	0.4				
1g-4		0.9	0.2	15	6	8	2
1g-5		4	1	5	2	2	0.5
1o-1	NH_2, F, 	0.8	0.2	31	23	68	27
1o-2		0.5	0.2	20	18	20	15
1o-3		1	0.4	14	13	17	17
1o-4		2	1	20	9	28	8
1o-5		16	2	16	5	5	3
1h-1	H, Cl, 	0.8	0.4				
1h-2		0.6	0.2	6	3		
1h-4		3	0.6	10	4	14	5
1h-5		4	1				
1p-1	NH_2, Cl,	2	0.5	26	10		
1p-2		0.7	0.3	6	4	12	9
1p-3		2	1	10	8		
1p-4		6	1				
1p-5		32	3	31	4		
1e-4	H,Oflox	29	2				
1e-5		>100	11				
1m-4	NH_2,Oflox	>100	23				
1m-5		>100	8				
1r-1	OH, H, 	3	0.1				
1r-4		3	0.4				
1s-2	OH, H, 	2	0.8	>50	>50		
1s-4		2	0.5	90	11	107	18

[a] Single dose given at challenge. [b] PO indicates oral administration by gavage, and SC indicates subcutaneous injection.

analogues competitive with the commonly used piperazines.[13a]

When the N_1-substituent is cyclopropyl (series f–h), addition of halogen at C_8 appears to improve Gram-positive activity significantly for the piperazinylquinolones **1g–1** and **1h–1**, but was neutral for the alkylated piperazinyl derivatives. For the 1-cyclopropylpyrrolidine series, addition of the halogens at C_8 is very beneficial (2–16-fold). Thus the structure–activity conclusions regarding the in vitro potency of the halogenated quinolones are dependent primarily on the choice of the N_1-substituent, and the side chain at R_7 as was observed by Chu[15] with the N_1-difluorophenyl analogues. The changes in Gram-negative antibacterial potency with halogenation of the 8-position do not correlate with the inhibition of DNA gyrase, which is generally unaffected.

Pyrrolidines vs Piperazines. The results seen in Tables IV and V regarding the differences between pyrrolidinyl and piperazinyl side chains are identical with those already published.[13a,14,20] In every case the pyrrolidines **4** and **5** confer superior Gram-positive activity relative to the piperazinyl analogues. Additionally, the 3-aminopyrrolidinyl side chain confers Gram-negative potency competitive with the best piperazines. These 3-aminopyrrolidinyl quinolones demonstrate the most balanced spectrum of activity throughout this study.

Given the intrinsically broad spectrum activity conferred by the 3-aminopyrrolidine group, the selective potency increases that come from the addition of the 8-halogen, and the 2–8-fold overall boost in activity from the cyclopropyl at N_1, it is not surprising that compounds **1g–4** (PD 117596) and **1h–4** (PD 127391, AM1091) are among the most potent quinolones ever reported.

5-Amino vs 5-Hydrogen. Matching the 5-hydrogen analogues, a–h (in Table IV) with the corresponding 5-amino derivatives i–p (in Table V), the effects of adding a 5-amino group can be determined. Once again the results show a striking dependence on the nature of the N_1-substituent. 5-Aminonorfloxacin (**1i–1**) is 10-fold less active than norfloxacin (**1a–1**) vs the Gram-negative organisms, but has five times improved potency against the Gram-positive organisms. The other N_1-ethyl-7-piperazinyl analogues (such as **1j–2,3** vs **1b–2,3** and **1k–1** vs **1c–1**) show sustained decreases in activity against all organisms when the 5-NH_2 is present. In the pyrrolidinyl series, analogue **1i–5** has host 3–7 times the potency of its 5-hydrogen counterpart **1a–5** against both the Gram-positive and Gram-negative organisms, respectively. In general, for every case where N_1 is not cyclopropyl, the antibacterial activity is reduced 2–5-fold, when the 5-amino group is introduced. These side by side results corroborate those suggested in the earlier literature, where 5-substitution was considered deleterious.[18]

However, when the N_1-substituent is cyclopropyl, the results are much different. Addition of the 5-amino group to ciprofloxacin (**1f–1** vs **1n–1**) increases Gram-positive potency by 8-fold and even improves the already good Gram-negative activity. The improved activity is seen throughout the N_1-cyclopropyl series peaking when R_8 is fluorine. Each of the 6,8-difluoro-5-aminoquinolones **1o–1** through **1o–5** shows a balanced spectrum and significant potency gains over their 5-hydrogen partners (**1g–1,5**). When R_8 is chloro, the gain over 5-hydrogen is smaller for the piperazines **1p–1** and **1p–2**. For the excellent 8-chloro-pyrrolidinylquinolones **1h–4** and **1h–5**, both Gram-positive and Gram-negative activity is reduced when the 5-amino group is added. Thus when N_1 is cyclopropyl, the 5-amino group is generally beneficial and the potency gain is somewhat dependent on the C_8-substituent improving significantly with addition of fluorine at C_8, and diminishing again with addition of chlorine. At the enzyme level, the 5-amino group generally has a neutral to negative effect on gyrase inhibition of 1–14-fold showing the greatest change for the pyrrolidinyl ofloxacin derivatives **1e–4,5** vs **1m–4,5** (Table III).

In the 5-amino-8-fluoro-1-cyclopropyl series **1o–1,3** three favorable structure–activity trends converge. First the 5-amino group boosts the Gram-positive potency of these piperazinyl analogues, then the 8-fluoro group further enhances overall activity. Finally alkylation of the piperazine is acceptable since N_1 is cyclopropyl. The net effect is that the piperazines **1o–1,2,3** are as potent overall as the 7-pyrrolidinyl derivatives. This level of Gram-positive activity has not been observed for any previous piperazinyl containing quinolones.

Other 5-Substituents. The successful implementation of the 5-amino group prompted us to explore the effects of a 5-hydroxy substituent (series qrs, Table VI). When N_1 is ethyl a significant loss of activity (2–10-fold) again occurs (series **1b** in Table IV vs **1q** in Table VI). When N_1 is cyclopropyl, the effect of the OH is neutral for the C_8-hydrogen examples (**1f** vs **1r**), and then generally decreases when C_8 is fluorine. Relative to the 5-amino series in Table V, the 5-hydroxy-1-cyclopropyl analogues are 2-

to 20-fold less active, especially vs Gram-positive organisms. The C_5-hydroxyl did not have any significant effect on solubility or other physical parameters. Thus, the 5-hydroxy derivatives offered little advantage over the corresponding 5-amino derivatives. In a few cases the 5-methoxyquinolones could be prepared if hydrolysis of the ester 8 (R_5 = OCH_3, R_1 = cyclopropyl, R_8 = fluoro or hydrogen) was performed under very mild (low yield) conditions. These analogues (data not shown), much like the previously reported 5-(methylamino)quinolones,[16] were inactive (mean MICs >3.1 μg/mL).

In Vivo Efficacy and Conclusions. The in vivo efficacy of select 1-cyclopropyl compounds is shown in Table VII. It has been previously noted how the halogen at R_8 improves in vivo efficacy.[12,14] The results in Table VII confirm this reported trend (series **1f** vs **1g** and **1h**; and **1n** vs **1o** and **1p**). Alkylation of the piperazine ring improves in vivo efficacy in several cases, most notably in the Streptococcus infections for **1f–3** vs **1f–1**, **1o–1** vs **1o–2** and **1o–3**, and for **1p–1** vs **1p–2** and **1p–3**. Finally in this broader list of comparisons, it is clear that the 5-amino group, as suggested earlier,[16] does reduce overall in vivo efficacy relative to a 5-hydrogen, especially when considering that it simultaneously improves Gram-positive in vitro potency.

In conclusion, using side by side comparisons we have clearly demonstrated that the effects of the 5-substituent, the 8-substituent, and even the C_7 side chain are determined primarily by the substituent at N_1. In particular, the 5-amino group increases in vitro potency when the N_1 group is cyclopropyl, but reduces potency when N_1 is ethyl or difluorophenyl. Similar results were witnessed for the effect of alkylation of the piperazine, and the addition of halogen of C_8. Clearly, the previous structure–activity results derived by workers in this field cannot be applied in general unless a variety of groups at the N_1 position are employed. Efforts to explain this phenomenon mechanistically are underway in our laboratories. Several compounds reported in this work have sufficiently good in vitro and in vivo potency such as **1o–2**, **1o–3**, **1o–4**, **1p–2**, and **1p–4** to warrant further development.

Experimental Section

All melting points were determined on a Hoover capillary melting point apparatus and are uncorrected. Infrared (IR) spectra were determined in KBr on a Nicolet FTIR SX-20 instrument. Mass spectra were recorded on a Finnigan 4500 GCMS or a VG Analytical 7070 E/HF spectrometer with an 11/250 Data System. Proton magnetic resonance (NMR) were recorded on either a Varian XL-200 or an IBM 100 WP100SY spectrometer. Shifts are reported in δ units relative to internal tetramethylsilane. Elemental analyses were performed on a Perkin-Elmer 240 elemental analyzer. All compounds prepared had analytical results ±0.4% of theoretical values. All organic solutions were dried over magnesium sulfate, and all concentrations were performed in vacuo at 10–30 mmHg. THF was dried over Na/benzophenone. All alkyl lithiums were from Aldrich and only new lots were analyzed. High-performance liquid chromatography (HPLC) was carried out by using an LKB 2150 pump, Rheodyne 7125 injector with 20-μL loop, Supelco LC18-DB column or ultramex C18 column (5-μm particle size, 250 × 4.6 mm i.d.), Perkin-Elmer LC-95 UV absorbance detector, and Hewlett-Packard 3390 integrator. Mobile phases were methanol/water (30:70–50:50), 0.05 M NaH_2PO_4, and methanol/water/acetonitrile (45:45:10). Column chromatography utilized Si gel (EM-60 230–400 mesh). All final products were analyzed by HPLC, but are reported here only where needed. No compounds tested were less than 97% pure by HPLC.

Preparation of 5-Amino-1-cyclopropyl-6,7,8-trifluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9o). General Sequence. 2,3,4,5-Tetrafluoro-6-nitrobenzoic Acid (4o). To a suspension of 125 g (640 mmol) of **2o** in 200 mL of H_2SO_4 at 60 °C was added 50 mL of fuming HNO_3 in 50 mL of H_2SO_4 (premixed with cooling). The dropwise addition was complete in 2 h. The mixture was stirred at 70 °C for 5.5 h and overnight at room temperature. The solids were filtered, suspended in cold H_2O saturated with NaCl, and filtered again. The solids were then dried to give 84 g (55%) of crude **4o** as a dark yellow solid. The solid was dissolved in ether and extracted with saturated NaCl solution three times. The ether layer was dried and concentrated to give 54 g (35%) of pure **4o** as a pale yellow powder: mp 135–136 °C; NMR ($CDCl_3$) δ 13.8 (br, s, 1 H); IR (KBr) 3300–2800, 1745, 1567 cm^{-1}; MS *m/z* 239 (M^+). Anal. Calcd for $C_7HF_4NO_4$: C, 35.16; H, 0.42; N, 5.86; F, 31.79. Found: C, 35.57; H, 0.54; N, 5.94; F, 31.50.

Ethyl 2,3,4,5-Tetrafluoro-6-nitro-β-oxobenzenepropanoate (5o). To a suspension of 64.2 g (269 mmol) of **4o** in 400 mL of H_2CCl_2 and 36.6 g (1.08 equiv) of oxalyl chloride was added 5 drops of DMF. The rapid gas evolution subsided after 16 h and all the solids had dissolved. The mixture was concentrated to give 68.6 g of crude 2,3,4,5-tetrafluoro-6-nitrobenzoyl chloride.

In a separate vessel, 62.8 g (475 mmol) of malonic acid monoethyl ester was dissolved in 1 L of THF with a catalytic amount of dipyridyl. At −30 °C, 310 mL of 1.6 M *n*-BuLi was added at a rate to keep the temperature between −20 and −30 °C. At −5 °C another 310–350 mL of *n*-BuLi was added until the pink color persisted for 5 min. The mixture was cooled to −78 °C, and the acid chloride in 100 mL of THF was added over 45 min. The mixture was then warmed to −35 °C for 1 h and was poured over ice water containing 40 mL (2 equiv) of concentrated HCl. The mixture was extracted with H_2CCl_2 three times, and the combined extracts were then washed with H_2O, 5% $NaHCO_3$, and 1 N HCl. The H_2CCl_2 was then dried and concentrated to give 67.6 g (91%) of **5o** as a thick oil: NMR ($CDCl_3$) δ 12.4 (bs, 1/2 H, enol), 5.5 (s, 1/2 H), 4.25 (m, 2 H), 3.9 (m, 1 H), 1.3 (m, 3 H); IR (LF) 1747, 1623 cm^{-1}; MS *m/z* 310 (M^+ + 1), 264 (M − OEt), 222 (M − CH_2CO_2Et).

Ethyl α-(Ethoxymethylene)-2,3,4,5-tetrafluoro-6-nitro-β-oxobenzenepropanoate (6o). A solution of 35 g (113 mmol) of **5o** in 29 mL of $HC(OEt)_3$ and 250 mL of Ac_2O was refluxed for 3 h. The mixture was concentrated under high vacuum to give 39.2 g (95%) of crude **6o**: NMR ($CDCl_3$) δ 8.08 (m, 1 H), 4.33 (m, 4 H), 1.25 (m, 6 H); IR (LF) 1717, 1555 cm^{-1}; MS *m/z* 360 (M^+).

Ethyl 1-Cyclopropyl-6,7,8-trifluoro-1,4-dihydro-5-nitro-4-oxo-3-quinolinecarboxylate (8o). To 39.0 g (107 mmol) of **6o** in 100 mL of *tert*-butyl alcohol was added 6.10 g (1.00 equiv) of cyclopropylamine. The mixture was heated for 3.5 h at 45 °C. To this mixture was added 12.0 g (1.00 equiv) of potassium *tert*-butoxide. After 6 h at 60 °C, the solids were collected, washed with water and ether, and dried to give 26 g (68%) of **8o**: NMR (Me_2SO-d_6) δ 8.57 (s, 1 H, C_2H), 4.23 (q, *J* = 7 Hz, 2 H), 4.04 (m, 1 H, c-C_3H_5), 1.15 (m, 7 H, c-C_3H_5 and CH_2CH_3); IR (KBr) 1734, 1627, 1560 cm^{-1}. Alternatively, the cyclization could be carried out in DMSO with K_2CO_3 (2.5 equiv) or with DMSO and triethylamine. The DMSO was removed to give a slurry which was alcohol precipitated.

5-Amino-1-cyclopropyl-6,7,8-trifluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9o). A suspension of 21.9 g (61.5 mmol) of the nitro ester **8o** in 300 mL of THF and 300 mL of EtOH was shaken with 3.0 g of RaNi at 20 psi H_2 for 12 h. The mixture was taken to 1.5 L with hot THF and was filtered to give 20 g (100%) of the ethyl ester of **9o** as an light yellow solid: mp 218–220 °C; NMR ($CDCl_3$) δ 8.44 (s, 2 H, C_2H), 6.5 (bs, 2 H, NH_2), 4.38 (q, *J* = 7 Hz, 2 H), 3.85 (m, 1 H, c-C_3H_5), 1.38 (t, *J* = 7 Hz, 3 H, CH_2CH_3), 1.05 (m, 4 H, c-C_3H_5); MS *m/z* 326 (M^+). Anal. Calcd for $C_{15}H_{13}F_3N_2O_3$: C, 55.22; H, 4.02; N, 8.59. Found: C, 55.20; H, 4.31; N, 8.37. This material was refluxed for 2 h in 6 M HCl. The solids were filtered, washed with ether, and air dried to give 18.0 g (100%) of **9o** as a yellow solid: mp 274–276 °C; NMR (Me_2SO-d_6) δ 14.23 (bs, 1 H, CO_2H), 8.43 (s, 1 H, C_2H), 7.55 (s, 2 H, NH_2), 4.05 (m, 1 H, c-C_3H_5), 1.12 (m, 4 H, c-C_3H_5); MS *m/z* 298 (M^+). Anal. Calcd for $C_{13}H_9F_3N_2O_3$: C, 52.36; H, 3.04; N, 9.39. Found: C, 52.55; H, 3.11; N, 9.27.

3-Chloro-2,4,5-trifluoro-6-nitrobenzoic Acid (4p). By using the procedure above for nitration of **2o**, 1.80 g (8.55 mmol) of 3-chloro-2,4,5-trifluorobenzoic acid (**2p**)[14] was converted to 1.8 g (82%) of **4p** as a bright yellow solid: mp 145–147 °C; NMR

(Me_2SO-d_6) δ 13.0 (very broad); MS *m/z* 255, 257 (M^+, M^+ + 2). Anal. Calcd for $C_7HF_3ClNO_4$: C, 32.90; H, 0.40; N, 5.48. Found: C, 32.95; H, 0.47; N, 5.33.

5-Amino-8-chloro-1-cyclopropyl-6,7-fluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9p). By following the same sequence of steps as outlined for the 5-amino-8-fluoroquinolone **9o**, the 3-chloro-2,4,5-trifluoro-6-nitrobenzoic acid (**4p**) was converted to the ethyl ester of **9p** in 24% overall yield. Cyclization to the quinolone **8p** using KO*t*-Bu/*t*-BuOH proceeded in 36% yield. Alternatively Et_3N in DMSO for the cyclization improved the yield to 66%. Reduction of the nitro group in **8p** gave the ethyl ester of **9p**, which was isolated as a pale yellow powder: mp 197–199 °C; NMR ($CDCl_3$) δ 8.5 (s, 1 H, C_2H), 7.2 (bs, 2 H + $CHCl_3$), 4.4 (q, *J* = 7 Hz, 2 H, CH_2CH_3), 4.15 (m, 1 H, c-C_3H_5), 1.45 (t, *J* = 7 Hz, 3 H, CH_2CH_3), 1.2 (m, 2 H, c-C_3H_5), 0.85 (m, 2 H, c-C_3H_5).

Hydrolysis with HCl as described in the general procedure gave **9p** (92%) as a yellow powder: mp 285–291 °C; NMR (Me_2SO-d_6) δ 14.23 (bs, 1 H, CO_2H), 8.71 (s, 1 H, C_2H), 8.0 (bs, 2 H, NH_2), 4.32 (m, 1 H, c-C_3H_5), 1.17 (m, 2 H, c-C_3H_5), 1.02 (m, 2 H, c-C_3H_5); MS *m/z* 314, 316 (M^+, M^+ + 2). Anal. Calcd for $C_{13}H_9ClF_2N_2O_3$: C, 49.57; H, 2.86; N, 8.90; Cl, 11.28. Found: C, 49.61; H, 2.70; N, 9.11; Cl, 11.51.

5-Amino-8-chloro-1-ethyl-6,7-difluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9k). By following the general procedure for the synthesis of **9o**, but using triethylamine in DMSO at 25 °C for the cyclization of **7k** to **8k**, the acid **4p** was converted to **9k** in 18% overall yield: mp 286–291 °C; NMR (TFA) δ 9.3 (s, 1 H, C_2H), 5.3 (q, *J* = 7 Hz, 2 H, NCH_2CH_3), 2.1 (t, *J* = 7 Hz, 3 H, NCH_2CH_3); IR (KBr) 3430, 3315, 1710, 1600 cm^{-1}. Anal. Calcd for $C_{12}H_9ClF_2N_2O_3$: C, 47.60; H, 2.98; N, 9.26. Found: C, 47.53; H, 2.77; N, 9.17.

8-Amino-9,10-difluoro-2,3-dihydro-3-methyl-7-oxo-7*H*-pyrido[1,2,3-*de*]-1,4-benzoxazine-6-carboxylic Acid (9m). To a solution of 19.4 g (69.0 mmol) of 9,10-difluoro-2,3-dihydro-3-methyl-7-oxo-7*H*-pyrido[1,2,3-de][1,4]benzoxazine-6-carboxylic acid (**9e**)[19] in 200 mL of concentrated H_2SO_4 was added portionwise 10.1 g (100 mmol) of KNO_3. The mixture was stirred at room temperature for 18 h and was poured over ice. The solids were collected, washed with H_2O, EtOH, and Et_2O, and then dried to give 20.5 g (91%) of the 8-nitro **9e**: mp 275–259 °C; NMR (Me_2SO-d_6) δ 9.2 (s, 1 H, C_5H), 5.15 (q, *J* = 6 Hz, 1 H, $NCHCH_3$), 4.9 (d, *J* = 9 Hz, OCHH), 4.7 (d, *J* = 9 Hz, 1 H, OCHCH), 1.5 (d, *J* = 6 Hz, $CHCH_3$).

A suspension of 19.8 g (61.0 mmol) of 8-nitro **9e** in 200 mL of DMF and 1.5 g of 5% Pd/C was shaken at 50 psi H_2 for 18 h. The DMF was removed at reduced pressure and the mixture slurried with hot AcOH. Filtration and concentration gave a residue, which was dissolved in water at pH 11.0, filtered through Celite to clarify, and precipitated at pH 5.5 with HCl. The solids were collected to give 15.4 g (85%) of **9m** as a white solid: mp 327–329 °C; NMR (Me_2SO-d_6) δ 14.6 (s, 1 H, CO_2H), 8.86 (s, 1 H), 7.2 (s, 2 H, NH_2), 4.86 (m, 1 H, $NCHCH_3$), 4.46 (d, 11.5 Hz, 1 H), 4.19 (d, *J* = 11.5 Hz, 1 H), 1.38 (d, *J* = 6 Hz, 3 H). Anal. Calcd for $C_{13}H_{10}F_2N_2O_4 \cdot 0.25H_2O$: C, 51.91; H, 3.35; N, 9.32; H_2O, 1.5. Found: C, 51.74; H, 3.46; N, 9.20; H_2O, 1.3.

Methyl 2,3,4,6-Tetrafluorobenzoate (3n). To 179.7 g (0.926 mol) of 2,3,4,6-tetrafluorobenzoic acid[22] in 1.5 L of MeOH was added 45 mL of concentrated H_2SO_4. The mixture was refluxed for 68 h, poured over ice, and extracted twice with H_2CCl_2. The organic layer was extracted by 5% $NaHCO_3$, dried, and concentrated to 170.2 g (88%) of **3n** as a colorless oil: NMR ($CDCl_3$) δ 6.85 (m, 1 H), 3.97 (s, 3 H).

3,4,6-Trifluoro-2-[(phenylmethyl)amino]benzoic Acid (4n). To 43.16 g (208 mmol) of **3n** in 450 mL of CH_3CN was added 23.8 mL (218 mmol) of benzylamine and 31.6 mL (277 mmol) of Et_3N. The mixture was stirred overnight at room temperature and was refluxed for 2 h. The mixture was concentrated, poured into H_2O, and extracted three times with Et_2O. The Et_2O was in turn extracted with H_2O and brine. The Et_2O was dried and concentrated to give 58.57 g of syrup, which was flash chromatographed to give 43.1 g (70%) of the methyl ester of **4n** as a pale yellow solid, which was recrystallized from hexane, mp 56–57 °C. This material was dissolved in 400 mL of MeOH and 76.5 mL of 2 N NaOH. The mixture was refluxed for 72 h. It was concentrated, extracted with Et_2O twice, acidified to pH 3.5, and extracted with Et_2O again. This latter extract was dried and concentrated to give 38 g (93%) of **4n**, mp 140–141 °C.

2-[Acetyl(phenylmethyl)amino]-3,4,6-trifluorobenzoic Acid (4n'). To a suspension of 37.9 g (135 mmol) of **4n** in 675 mL of triethylamine and 0.34 g of 4-(dimethylamino)pyridine was added 14.4 g (141 mmol) of Ac_2O. The mixture was stirred vigorously overnight. It was added to 700 mL of 0.5 M NaOH and extracted with Et_2O three times. The aqueous layer was acidified with concentrated HCl to pH 1.8 and extracted with Et_2O. The Et_2O was dried and concentrated to give 41.4 g (95%) of crude **4n'**, which was recrystallized from toluene/isooctane to give 35.8 g (82%) of pure **4n'**: mp 150–153 °C; NMR ($CDCl_3$) δ 9.5–10.4 (bs, -2 H, CO_2H, H_2O), 7.23 (m, 5 H, Ph), 7.05 (m, 1 H, C_4H), 5.22 (d, *J* = 14 Hz, 1 H, PhCH*H*), 4.52 (d, *J* = 14 Hz, 1 H, PhC*H*H), 2.03 (s, 3 H, $COCH_3$). Anal. Calcd for $C_{16}H_{12}F_3NO_3 \cdot 0.5H_2O$: C, 57.83; H, 3.91; N, 4.21; F, 17.16. Found: C, 58.11; H, 3.68; N, 4.29; F, 16.98.

5-Amino-1-cyclopropyl-6,7-difluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9n). By following the general procedure as outlined for **9o**, the 2-[acetyl(phenylmethyl)-amino]benzoic acid (**4n'**) was converted to **9n** in 27% overall yield. The final ester hydrolysis step removes the acetyl and the benzyl groups along with the ester. Compound **9n** was isolated as a white solid: mp >300 °C; NMR (TFA) δ 9.29 (s, 1 H, C_2H), 7.68 (m, 1 H, C_8H), 3.92 (m, 1 H, c-C_3H_5), 1.67 (m, 2 H, c-C_3H_5), 1.41 (m, 2 H, c-C_3H_5); MS *m/z* 280 (M^+). Anal. Calcd for $C_{13}H_{10}F_2N_2O_3$: C, 55.71; H, 3.60; N, 10.00. Found: C, 55.63; H, 3.66; N, 9.88.

5-Amino-1-ethyl-6,7-difluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9i). By following the general procedure for **9o** the 5-amino-1-ethylquinoline **9i** was prepared in 66% overall yield from **4n'**. This compound, **9i**, was isolated as a tan solid: mp >300 °C; NMR (TFA, Me_2SO-d_6) δ 8.8 (s, 1 H, C_2H), 7.0 (m, 1 H, C_8H), 4.4 (q, *J* = 7 Hz, 2 H, CH_2CH_3), 1.45 (t, *J* = 7 Hz, 3 H, CH_2CH_3); MS *m/z* 268 (M^+).

5-Amino-1-(2,4-difluorophenyl)-6,7,8-trifluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9l). By following the general procedure for **9o**, the 5-amino-1-(2,4-difluorophenyl)quinoline **9l** was prepared in 58% overall yield from **4n'**. Compound **9l** was isolated as a white powder: mp >300 °C; NMR (TFA) δ 9.0 (s, 1 H, C_2H), 7.5 (m, 1 H), 7.1 (m, 2 H), 6.5 (m, 1 H). Anal. Calcd for $C_{16}H_8F_4N_2O_3$: C, 54.55; H, 2.29; N, 7.95. Found: C, 54.83, H, 2.26; N, 7.58.

2,3,4,5-Tetrafluoro-6-methoxybenzoic Acid (4s). To a solution of 50 mL of MeOH (1200 mmol) in 500 mL of Et_2O was added dropwise 140 mL of 2.05 M *n*-BuLi, maintaining the temperature at −20 °C. Twenty minutes after addition was complete, a solution of 70.7 g (267 mmol) of 2-(pentafluorophenyl)-4,4-dimethyl-2-oxazoline[23] in 100 mL of Et_2O was added quickly between −10 °C and −20 °C. After 2 h at −20 °C the mixture was brought to room temperature. The mixture was diluted with H_2O and extracted into EtOAc. The extract was dried and concentrated to give 76.1 g (95%) of 2-(2,3,4,5-tetrafluoro-6-methoxyphenyl)-4,4-dimethyl-2-oxazoline: NMR ($CDCl_3$) δ 4.12 (s, 2 H), 3.95 (m, 3 H, OCH_3), 1.39 (s, 6 H, $2CH_3$); MS *m/z* 277 (M^+), 289 (dimethoxy).

A mixture of 68.6 g (240 mmol) of the above material and 400 mL of 6 N HCl was refluxed overnight. The solution was diluted with water and extracted with EtOAc. The organic layer was dried and concentrated. The residue was dissolved in water, taken to pH 10.5 with NaOH, was washed with Et_2O and was reacidified to pH 2 with HCl. The aqueous mixture was again extracted with EtOAc, which was dried and concentrated to give 37.4 g (69%) of **4s** as a tan solid, consisting of 4:1 mixture of 6-methoxy and 4-methoxy isomers: NMR ($CDCl_3$) δ 4.05 and 4.01 (2d, 4:1).

1-Cyclopropyl-6,7,8-trifluoro-1,4-dihydro-5-hydroxy-4-oxo-3-quinolinecarboxylic Acid (9s). By following the general procedure for **9o**, the 6-methoxybenzoic acid (4:1 mixture with 4-methoxy isomer) **4s** was converted to **9s** in 14% overall yield.

(22) Houlihan, W. J. US Pub. Pat. Appl. B. 390031, 1976; *Chem. Abstr.* **1976**, *84*, 121479f.

(23) Inukai, Y.; Takuma, K.; Toritani, K.; Sonoda, T.; Kobayashi, H. *Bull. Chem. Soc. Japan* **1984**, *57*, 225.

Compound **9s** was contaminated with 15% of 1-cyclopropyl-5,6,8-trifluoro-1,4-dihydro-7-methoxy-4-oxo-3-quinolinecarboxylic acid. This material washes away with the subsequent addition of the side chain and workup: NMR ($CDCl_3$) δ 14.4 (s, 1 H, 7-CO_2H), 12.9 (bs, 1 H, OH), 8.68 and 8.66 (2s, 1 H total, C_2H), 4.20 (d, 7-OCH_3 impurity), 3.92 (m, 1 H, c-C_3H_5), 1.20 (m, 4 H, c-C_3H_5); IR (KBr) 1743; MS m/z 299 (M^+), 313 (7-OCH_3 impurity).

1-Ethyl-6,7,8-trifluoro-1,4-dihydro-5-hydroxy-4-oxo-3-quinolinecarboxylic Acid (9q). By using the general procedure outlined for **9o**, the 1-ethyl-5-hydroxyquinoline **9q** was prepared from **4s** in 10% overall yield as a tan solid: mp 227–228 °C; NMR (Me_2SO-d_6) δ 14.6 (bs, 1 H, CO_2H), 13.0 (bs, 1 H, OH), 8.96 (s, 1 H, C_2H), 4.55 (m, 2 H, NCH_2CH_3), 1.42 (t, J = 6 Hz, 3 H, NCH_2CH_3); IR (KBr) 1743; MS m/z 287 (M^+). There was no trace of the 7-methoxy impurity observed in any of the spectral data or by HPLC analysis. Anal. Calcd for: C, 50.19; H, 2.81; N, 4.88. Found: C, 50.22; H, 2.78; N, 4.57.

8-Chloro-1-ethyl-6,7-difluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9c). The general procedure for **9o** was followed with slight modification. The base used for the cyclization step was K_2CO_3 (2.5 equiv) and THF was employed as solvent. By using this modification, the 3-chloro-2,4,5-trifluorobenzoic acid **2p** was converted to **9c** in 56% overall yield. **9c** was isolated as a white powder: mp 216–218 °C; NMR (TFA/Me_2SO-d_6) δ 9.0 (s, 1 H, C_2H), 8.45 (dd, J = 9 Hz, J = 6 Hz, 1 H, C_5H), 5.0 (q, J = 7 Hz, 2 H, CH_2CH_3), 1.75 (t, J = 7 Hz, 3 H, CH_2CH_3); MS m/z 287 (M^+), 289 (M^+ + 2); HPLC 98%.

3,4,6-Trifluoro-2-methoxybenzoic Acid (4r). To 19.4 g (100 mmol) of 2,3,4,6-tetrafluorobenzoic acid[22] (**3r**) in 50 mL of DMF was added over 30 min a slurry of 8.8 g (220 mmol, 60% oil dispersion) of NaH in 50 mL of DMF. When the vigorous gas evolution slowed, 3.26 g (102 mmol) of MeOH in 10 mL of DMF was added again accompanied with gas evolution and significant foaming. When the foaming ceased, the mixture was taken to 100 °C for 1 h. Once cooled, the mixture was poured into water and extracted three times with Et_2O. The water layer was acidified to pH 2.0 and extracted with Et_2O again. These latter extracts were dried and concentrated to give 17.23 g (84%) of **4r** as a light yellow solid: mp 91–94 °C; NMR ($CDCl_3$) δ 10.9 (bs, 1 H, CO_2H), 6.76 (m, 1 H, C_5H), 4.09 (d, J = 2.4 Hz, 3 H, OCH_3). Anal. Calcd for $C_8H_5F_3O_3$: C, 46.60; H, 2.43; F, 27.67. Found: C, 46.31; H, 2.28; F, 27.55.

1-Cyclopropyl-6,7-difluoro-1,4-dihydro-5-hydroxy-4-oxo-3-quinolinecarboxylic Acid (9r). By following the procedure for **9o**, the 2-methoxybenzoic acid **4r** was converted to **9r** in 25% overall yield as a white solid, which was recrystallized from DMSO/H_2O: mp 262–263 °C; NMR (Me_2SO-d_6) δ 8.71 (s, 1 H, C_2H), 7.67 (dd, J = 8 Hz, J = 5 Hz, 1 H, C_8H), 3.73 (m, 1 H, c-C_3H_5), 1.28 (m, 2 H, c-C_3H_5), 1.18 (m, 2 H, c-C_3H_5). Anal. Calcd for $C_{13}H_9F_2NO_4$: C, 55.52; H, 3.23; N, 4.98. Found: C, 55.31; H, 3.06; N, 4.76.

5-Amino-1-ethyl-6,7,8-trifluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic Acid (9j). By following the procedure for **9o**, the quinoline **9j** was prepared from **5o** in 19% overall yield: mp 288–295 °C dec; NMR (TFA) δ 9.06 (s, 1 H, C_2H), 4.8 (m, 2 H, CH_2CH_3), 1.67 (t, J = 7 Hz, 3 H, CH_2CH_3).

Registry No. **1a**-1, 70458-96-7; **1a-2**, 98079-48-2; **1a-2** free base, 98079-47-1; **1a**-3, 114506-56-8; **1a-4**, 131683-63-1; **1a-5**, 91187-95-0; **1b**-1, 99726-76-8; **1b-2**, 98079-51-7; **1b**-3, 98079-68-6; **1b-4**, 91188-12-4; **1b-5**, 91188-00-0; **1v**-1, 80076-54-6; **1c**-1 free base, 112282-53-8; **1c-2**, 111234-01-6; **1c**-3, 131683-64-2; **1c**-3 free base, 131683-65-3; **1c-4**, 131683-66-4; **1c-4** free base, 112282-61-8; **1c-5**, 112282-64-1; **1d-2**, 108319-06-8; **1e-4**, 131683-67-5; **1e-5**, 91196-82-6; **1f**-1, 85721-33-1; **1f-2**, 93107-32-5; **1f**-3, 93107-34-7; **1f-4**, 105112-37-6; **1f-5**, 104455-77-8; **1g**-1, 94242-53-2; **1g-2**, 103460-89-5; **1g**-3, 103460-90-8; **1g-4**, 99734-98-2; **1g-5**, 99734-97-1; **1h**-1, 99696-22-7; **1h-2**, 103460-91-9; **1h-2** free base, 101987-76-2; **1h-4**, 105956-97-6; **1h-5**, 104456-00-0; **1i**-1, 131683-68-6; **1i-5**, 131683-69-7; **1j**-1, 88488-43-1; **1j-2**, 131683-70-0; **1j**-3, 131683-71-1; **1j-4**, 131683-72-2; **1j**-4 free base, 119354-30-2; **1j-5**, 103772-17-4; **1k**-1, 131683-73-3; **1k**-3, 131683-74-4; **1k-5**, 131683-75-5; **1l-2**, 131683-76-6; **1l-2** free base, 131683-77-7; **1m-4**, 131683-78-8; **1m-4** free base, 131683-79-9; **1m-5**, 131683-80-2; **1m-5** free base, 103784-34-5; **1n**-1, 123016-42-2; **1n-2**, 123016-41-1; **1n-4**, 131683-81-3; **1n-4** free base, 131683-82-4; **1n-5**, 131683-83-5; **1n-5**, 131683-84-6; **1o**-1, 110236-78-7; **1o-2**, 110871-85-7; **1o**-3, 111542-93-9; **1o-4**, 112654-98-5; **1o-5**, 103784-28-7; **1p**-1, 131683-85-7; **1p**-1 free base, 111230-43-4; **1p-2**, 111230-45-6; **1p**-3, 130975-70-1; **1p-4**, 115904-58-0; **1p-5**, 115904-26-2; **1q**-1, 131683-86-8; **1q-2**, 131683-87-9; **1q**-3, 131683-88-0; **1q-4**, 131683-89-1; **1q-5**, 131683-90-4; **1r**-1, 124487-37-2; **1r**-4, 131683-91-5; **1r-4** free base, 131683-92-6; **1s**-1, 114038-14-1; **1s-2**, 111230-47-8; **1s-4**, 131683-93-7; **1s-4** free base, 114008-27-4; **2o**, 1201-31-6; **2p**, 101513-77-3; **3n**, 53001-68-6; **4n**, 123016-61-5; **4n** methyl ester derivative, 131683-94-8; **4n'**, 123016-62-6; **4o**, 16583-08-7; **4p**, 111230-48-9; **4r**, 124487-29-2; **4s**, 38512-77-5; **4s** 5-methoxy derivative, 131683-95-9; **5o**, 103772-11-8; **6o**, 104885-01-0; **8o**, 103772-12-9; **8p**, 111230-51-4;)9c, 80076-53-5; **9e**, 82419-35-0; **9e** 8-nitro derivative, 127625-16-5; **9i**, 131683-96-0; **9j**, 131683-97-1; **9k**, 131683-98-2; **9k** ethyl ester derivative, 131683-99-3; **9l**, 131684-00-9; **9m**, 84427-35-0; **9n**, 123016-57-9; **9o**, 103772-14-1; **9o** ethyl ester derivative, 103772-13-0; **9p**, 111230-53-6; **9p** ethyl ester derivative, 111230-52-5; **9q**, 131684-01-0; **9r**, 124487-36-1; **9s**, 114008-16-1; 2,3,4,5-tetrafluoro-6-nitrobenzoyl chloride, 103772-10-7; 2-(pentafluorophenyl)-4,4-dimethyl-2-oxazoline, 90619-70-8; 2-(2,3,4,5-tetrafluoro-6-methoxyphenyl)-4,4-dimethyl-2-oxazoline, 131684-02-1; 1-cyclopropyl-5,6,8-trifluoro-1,4-dihydro-7-methoxy-4-oxo-3-quinolinecarboxylic acid, 107564-13-6; 2,3,4,6-tetrafluorobenzoic acid, 32890-92-9.