Synthesis, Chemistry, and Antineoplastic Activity of α-Halopyridinium Salts: Potential Pyridone Prodrugs of Acylated Vinylogous Carbinolamine Tumor Inhibitors[1]

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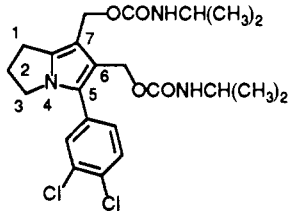
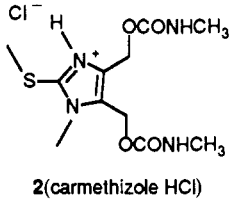
A series of 4- and 5-[2,3-dihydro-6,7-bis[[(*N*-alkylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]-2-halopyridinium iodides were synthesized. The rates of hydrolysis of the α-halopyridinium salts to the corresponding pyridones, and the reactivities of the carbamate moieties were studied as a function of pH, buffer composition, and ionic strength. The 4- and 5-pyrrolizinyl-2-halopyridinium iodides and the corresponding pyridones were evaluated against P388 lymphocytic leukemia in vivo. The α-fluoropyridinium compounds were active but the α-chloro compounds were not. This activity was correlated with the rates of hydrolysis of the α-halopyridinium compounds to the active pyridone. Compounds that were active in the P388 screen were evaluated in L1210 leukemia, M5076 carcinoma, and MX-1 mammary xenograft assays in mice.

The class of antineoplastic compounds known as "acylated vinylogous carbinolamines" contains a number of agents which have shown significant activity against a wide spectrum of experimental tumors.[2] Notable examples of drugs in this class include IPP (**1**)[3] and carmethizole hydrochloride (**2**).[4] The compounds were designed as



1(IPP)

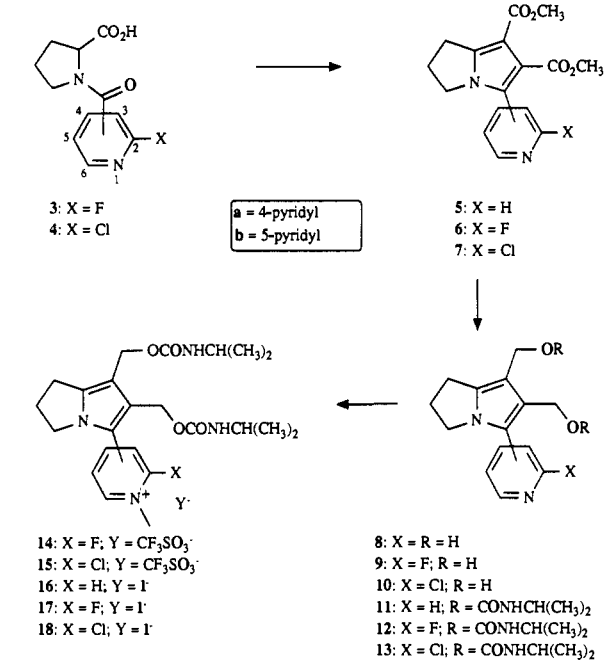


2(carmethizole HCl)

bifunctional electrophiles in which the carboxylates act as leaving groups in the alkyl–oxygen ester cleavage reaction.[5] The compounds are not carbamoylating agents; carbamate esters were chosen because they were reported to be more resistant to *esterase*-catalyzed hydrolysis than simple carboxylic acid esters.[6] The reactivities of the ester functions toward alkyl–oxygen cleavage has been shown to depend upon the electron density in the heteroaromatic ring. Thus, for example, in the pyrrolizine series of compounds related to **1**, the reactivity of the carbamates varies with the electronic influence of the phenyl substituents.[5,7]

The difficulty with IPP (**1**) is that it is insoluble in water and unstable in aqueous mixtures.[8] We previously reported the 2-hydroxy derivative of IPP (**1**) and polar, water soluble esters as potential prodrugs; the 2-hydroxy analogue of **1** was active against P388 lymphocytic leukemia in vivo but the water-soluble compounds were not.[9] We believe that the inactivity of the soluble compounds was due to facile hydrolysis of the carbamate ester(s) in aqueous solution.

Scheme I



This paper describes a new prodrug approach in which an α-halopyridinium moiety is used as the pyrrolizine C-5 substituent. The pyridinium moiety will impart water solubility to the compound, and as importantly, the strong electron-withdrawing influence of this group will reduce the reactivity of the carbamates (relative to **1**). Hydrolysis of the α-halopyridinium moiety to a pyridone will render the resulting compound more lipophilic, and because the pyridone is less electron withdrawing than the pyridinium moiety, the pyridone bis-carbamate will be more reactive. *Thus, this new prodrug approach offers both water solubility and latent reactivity.*

α-Halo-*N*-alkylpyridinium salts have been used as reagents for a number of different organic transformations. The synthetic utility is a consequence of the propensity

(1) Vinylogous Carbinolamine Tumor Inhibitors. 24. For part 23, see: Anderson, W. K.; Heider, A. R.; Raju, N.; Yucht, Y. A. *J. Med. Chem.* **1988**, *31*, 2097.
(2) (a) Anderson, W. K.; Chang, C.-P.; Corey, P. F.; Halat, M. J.; Jones, A. N.; McPherson, H. L., Jr.; New, J. S.; Rick, A. C. *Cancer Treat. Rep.* **1982**, *66*, 91. (b) Anderson, W. K. *Cancer Res.* **1982**, *42*, 2168. (c) Anderson, W. K. *Proc. Int. Congr. Chemother.* **1983**, *12*, (220), 39.
(3) Anderson, W. K.; New, J. S.; Corey, P. F. *Arzneim.-Forsch.* **1980**, *30(I)*, 765.
(4) Anderson, W. K.; Bhattacharjee, D.; Houston, D. M. *J. Med. Chem.* **1989**, *32*, 119.
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(8) El-Sayed, A.-A.; Repta, A. *Int. J. Pharm.* **1983**, *13*, 303.
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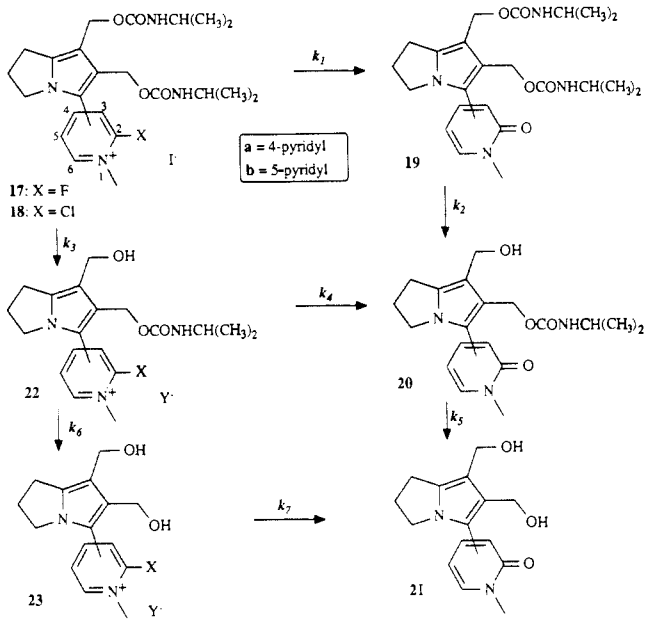
Scheme II



for these electron-deficient heterocycles to undergo nucleophilic substitution at the α-position.[10]

Chemistry

Synthesis. The 4- and 5-substituted 2-halopyridinium compounds **17** and **18** were synthesized according to the general method outlined in Scheme I (*note*: the 4- and 5-substituted pyridines are designated as **a** and **b**, respectively). Acylation of *l*-proline with the appropriate 2-haloisonicotinoyl chloride or 2-halonicotinoyl chloride gave the α-amido acids **3** or **4**. Treatment of **3** or **4** with acetic anhydride at 65–80 °C (higher temperatures gave significantly lower yields) generated, in situ, the mesoionic oxazolone intermediates that underwent smooth 1,3-dipolar cycloaddition with dimethyl acetylenedicarboxylate to give the 1,2-dihydro-1*H*-pyrrolizines **6** or **7**. Catalytic reduction of **7** (Pd/C) under acidic conditions effected dechlorination to give **5**. Reduction of diesters **5–7** with lithium aluminum hydride in THF at −15 °C gave diols **8–10** (reduction at higher temperatures led to attack on the α-halopyridine moiety). Acylation of the diols with isopropyl isocyanate in the presence of a catalytic amount of di-*n*-butyltin diacetate gave bis-carbamates **11–13**. Pyridine **11** was alkylated with iodomethane to give **16**, but the less reactive α-halopyridines **12** or **13** required the more reactive methyl triflate to prepare the pyridinium salts **14** or **15**. The trifluoromethylsulfonate counterion in **14** or **15** was exchanged for iodide by treatment with sodium iodide in anhydrous acetone to give **17** and **18**.

The *N*-methyl-α-fluoropyridinium compounds **14a** and **14b** were hydrolyzed to the corresponding pyridone bis-carbamates **19a** and **19b** by treatment with a two-phase mixture of acetone and saturated aqueous sodium bicarbonate solution (Scheme II). The hydrolysis of **14a** gave the pyridone bis-carbamate **19a** as the major product along with smaller amounts of the pyridone monocarbamate **20a** and the diol **21a**. Hydrolysis of **14b** under the same conditions gave the diol **21b** as the major product along with smaller amounts of the bis-carbamate **19b** and the monocarbamate **20b**. This latter result was due to the greater reactivity of **19b** compared to **19a** (see below).

Hydrolysis Studies. Three different hydrolytic rates must be considered for the compounds under study. These are (1) the activation rate, k_1 (Scheme II), where the α-halopyridinium salt is hydrolyzed to the pyridone; (2) the rate(s) of inactivation k_3 (and k_6) in which carbamate hydrolysis occurs before the α-halopyridinium moiety has been hydrolyzed, and; (3) the rates of carbamate ester hydrolysis on the pyridones **19** and **20**, k_2 and k_5. In the "acylated vinylogous carbinolamine" class of compounds, ester hydrolysis generally causes reduction or loss of antineoplastic activity. Thus, ester hydrolysis in either **17** or **18** can be considered an inactivation process. Obviously, in order for the prodrug to function, activation (k_1) must be faster than inactivation (k_3). Hydrolysis of the carbamate esters in the "acylated vinylogous carbinolamines" occurs by means of an alkyl–oxygen cleavage mechanism; therefore, the rate(s) of carbamate hydrolysis in **19** (k_2) and **20** (k_5) can be used as a comparative measure of the alkylating potential of these bifunctional electrophiles.

The hydrolyses of the α-halopyridinium compounds to the pyridones were followed by UV spectroscopy [**17a** (UV_{max} 388 nm) or **18a** (384 nm) hydrolyzed to **19a** (319 nm); **17b** (334 nm) or **18b** (330 nm) hydrolyzed to **19b** (276 nm)]. The k_{obs} is a combination of rates because this method does not distinguish between k_1, k_4, and k_7. It is assumed that the degree of esterification in **17**, **18**, **22**, and **23** will not have a significant influence on the rate of α-halopyridinium salt hydrolysis. The 5-substituted 2-fluoropyridinium compound **17b** was more reactive (k_{obs} = 0.0072 s^{-1}; $t_{1/2}$ = 1.6 min) than the 4-substituted 2-fluoropyridinium compound **17a** (k_{obs} = 0.0337 s^{-1}; $t_{1/2}$ = 20.5 min) but both were much more reactive than the α-chloropyridinium compounds. The pseudo-first-order rate constants for the hydrolysis of the α-chloro compounds **18a** and **18b** were too slow to measure under the conditions studied (0.05 M phosphate buffer, pH 7.4, 25 °C), the compounds had half-lives greater than 7 days.

The hydrolysis of **17a** was studied in more detail and the effects of phosphate buffer concentration, pH, and ionic strength on the hydrolysis of the pyridinium moiety

Table I. Effect of Phosphate Buffer Concentration on the Hydrolysis of the α-Fluoropyridinium Moiety in **17a** at pH 6.5 and 8.0[a]

pH	buffer concn, M	K_{obs}, min^{-1}	half-life, min
6.5	0.02	0.0052	133.3
	0.05	0.0072	96.2
	0.10	0.0138	50.3
8.0	0.02	0.0132	52.5
	0.05	0.0200	34.65
	0.10	0.0360	19.25

[a] Ionic strength = 0.05 M, 25 °C.

Table II. Effect of pH on the Hydrolysis of the Pyridinium Moiety in **17a** in 0.05 M Phosphate Buffer[a]

pH	K_{obs}, min^{-1}	half-life, min
6.0	0.0044	157.5
6.5	0.0153	45.3
7.0	0.0208	33.8
7.4	0.0337	20.5
8.0	0.0520	13.3

[a] Ionic strength = 0.25 M, 25 °C.

Table III. Effect of Ionic Strength on the Hydrolysis of the Pyridinium Moiety in **17a** in 0.02 M Phosphate Buffer, pH 7.4, 25 °C

NaCl, M	K_{obs}, min^{-1}	half-life, min
0.10	0.024	29.1
0.25	0.020	33.9
0.50	0.011	63.2
1.0	0.007	99.1

(10) Mukaiyama, T. *Angew. Chem., Int. Ed. Engl.* **1979**, *103*, 2625.

Table IV. Effect of pH on Carbamate Hydrolysis of **19a** in Acetonitrile–Water (1:1), 0.05 M Buffer, at 25 °C[a]

pH	K_{obs}, min^{-1} [b]	half-life, min[b]	pH	K_{obs}, min^{-1} [b]	half-life, min[b]
2.0	0.330	2.11	6.0	0.0135	51.35
3.0	0.0304	22.80	7.4	0.0130	53.51
3.5	0.0214	32.46	8.0	0.0128	54.14
4.0	0.0149	46.69	9.0	0.0129	53.97
5.0	0.0139	50.05	10.0	0.0133	52.33

[a] Ionic strength = 0.136, adjusted by addition of KCl. [b] Data given are the averages of two determinations.

Table V. Effect of Buffer Concentration on Carbamate Hydrolysis for **19a** in Acetonitrile–Water (1:1) at 25 °C[a]

pH	buffer concn, M	K_{obs}, min^{-1} [b]	half-life, min[b]
3.0	0.05	0.0304	22.80
3.0	0.025	0.0342	20.27
3.0	0.01	0.0349	19.86
7.4	0.05	0.0130	53.51
7.4	0.025	0.0134	51.92
7.4	0.01	0.0133	52.31

[a] pH 3 buffer = 0.05 M citrate and pH 7.4 buffer = 0.05 M phosphate (ionic strength = 0.136, adjusted by the addition of KCl). [b] Data given are the averages of two determinations.

Table VI. Effect of Ionic Strength on Carbamate Hydrolysis for **19a** in Acetonitrile–Water (1:1) at 25 °C in 0.05 M Phosphate Buffer, pH 7.4

ionic strength	K_{obs}, min^{-1} [a]	half-life, min[a]
0.136 (0.025 M KCl)	0.0130	53.51
0.211 (0.10 M KCl)	0.0124	55.89
0.611 (0.50 M KCl)	0.0080	86.65

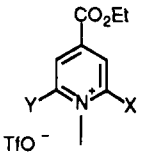
[a] Data given are the averages of two determinations.

to the pyridone are summarized in Tables I, II, and III, respectively. These data show that the reaction rate increases with either increased buffer concentration or increased pH and are characteristic of general-base catalysis.[11] The negative salt effect seen from the data in Table III was the expected influence of changes in the polarity of the reaction medium on the conversion of the ionic pyridinium salt to a neutral pyridone.

The hydrolysis of the pyridone bis-carbamate **19** was studied by HPLC. The reaction in pure water was too fast to follow, but the reaction was slower in acetonitrile–water (1:1) and could be followed by HPLC. The hydrolysis of **19** in acetonitrile–water (1:1), *with no added buffer*, gave monocarbamate **20**, which was subsequently hydrolyzed to diol **21**. The reaction was more complex in acetonitrile–aqueous phosphate buffer because phosphate also acted as a nucleophile. Therefore, in addition to **20** and **21**, mono- and diphosphates were also formed. These phosphates underwent slow hydrolysis to diol **21**. The hydrolysis [0.05 M phosphate buffer–acetonitrile (1:1), pH 7.4, 0.025 M KCl, 25 °C] of **19a** and **19b** were compared to that of **1**. The pseudo-first-order rate constants were determined by monitoring the disappearance of the bis-carbamate. 5-Substituted pyridone **19b** (k_{obs} = 0.214 min^{-1}; $t_{1/2}$ = 3.25 min) was more reactive than both **1** (k_{obs} = 0.0422 min^{-1}; $t_{1/2}$ = 16.44 min) and 4-substituted pyridone **19a** (k_{obs} = 0.0130 min^{-1}; $t_{1/2}$ = 53.51 min).

The data for carbamate hydrolysis in **19a** as a function of pH, buffer concentration, and ionic strength are given in Tables IV, V, and VI, respectively. The rate of hydrolysis was relatively constant over the pH range of 4–10,

Table VII. Hydrolysis Rates for Model α-Halopyridinium Salts[a]



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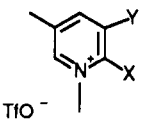

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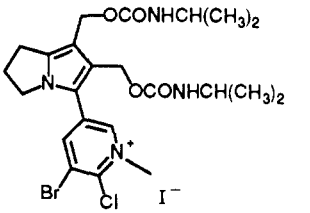
compd	X	Y	k_{obs}	half-life
24a	F	H	*b*	
24b	Cl	H	0.055 min^{-1}	6.0 h
24c	Br	H	0.012 h^{-1}	57.7 h
24d	Cl	Cl	0.114 min^{-1}	126 min
25a	F	H	0.0605 min^{-1}	11.4 min
25b	Cl	H	0.0425 h^{-1}	268.6 h
25c	Cl	Cl	0.055 h^{-1}	12.6 h
25d	Cl	Br	0.041 min^{-1}	17.0 h

[a] Hydrolyses (to the corresponding pyridones) were done at 25 °C in 0.05 M phosphate buffer (pH = 7.4). [b] The rate was too fast to measure under these conditions.

but a rapid rate increase was observed below pH 3. This corresponds to acid catalysis, below pH 3, where k_{obs} = $K_{rate}[H^+] + k^0$.[12] The data in Table V show that buffer concentration does not have a significant effect on the rate of hydrolysis at either pH 3.0 or 7.4. This indicates specific, and not general, acid catalysis. The negative salt effect seen from the data in Table VI is well established for hydrolysis reactions of this type.

Carbamate hydrolysis in the pyridinium salts **16a** and **16b** can be used to study the inactivation rates, k_3 (and k_6), of the prodrug. These salts lack the α-halogen substituent so the pyridinium moiety will be inert, but the carbamate esters in **16a** and **16b** can be hydrolyzed. The esters in the 4-substituted pyridinium salt **16a** (k_{obs} = 0.006 min^{-1}; $t_{1/2}$ = 115 min) were much more stable than in **16b** (k_{obs} = 0.071 min^{-1}; $t_{1/2}$ = 10 min) under the conditions studied (0.05 M deuterated phosphate buffer in deuterium oxide, pH 7.4, 25 °C). The reaction was run in deuterium oxide so it could be followed by NMR; no deuterium isotope effect should be observed since the rate-limiting step does not involve proton transfer.

A series of model α-halopyridinium compounds were prepared and the hydrolysis rates were studied. The structures of the model compounds, along with hydrolytic data, are given in Table VII. These data showed that substitution of the α-halopyridinium ring with electron-withdrawing substituents increased the rate of hydrolysis to the pyridone and that 2-chloro-3-bromopyridinium salt **25d** had a hydrolytic half-life that was intermediate between the α-fluoro- and the α-chloropyridinium salts **25a** and **25b**, respectively. The corresponding bis-carbamate **26** was prepared for additional study. Hydrolysis of the



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pyridinium salt in **26** to the corresponding pyridone [0.05

(11) If a buffer component was acting as a nucleophile in a slow step, this result would be observed for both general- and specific-base catalysis.

(12) If the Bronsted coefficient is very small, there would be no variation with buffer concentration in both general and specific catalysis.

Table VIII. Activity of the α-Halopyridinium Compounds 17a, 17b, 18a, and 26 and the Pyridone 19a against P388 Lymphocytic Leukemia in Vivo[a,b]

compd	dose, mg/kg	TDS[c]	BWD:[d] T − C, g	%T/C[e]	KE[f]
17a[g]	100	1/6	−7.6		
	50	6/6	−0.8	157	1.32
	25	6/6	−1.0	141	1.25
	12.5	6/6	−1.3	149	0.75
18a	200[h]	0/6			
	100[g]	6/6	−0.8	121	−1.07
	50[g]	6/6	−0.5	112	−1.44
	25[g]	6/6	0.4	103	−1.56
	12.5[g]	6/6	−0.1	103	−1.56
17b[g]	400	0/6			
	200	2/6	−0.1		
	100	6/6	0.1	158	1.54
	50	6/6	0.6	141	0.32
	25	6/6	−0.1	137	0.06
	12.5	6/6	1.2	112	−1.42
18b[g,i]	50	0/4			
	25	4/4	−1.1	109	
	12.5	4/4	0.1	102	
	6.2	4/4	−0.4	98	
19a[h]	400	2/6	−1.0		
	200	6/6	−1.4		
	100	5/6	−1.8	148	0.69
	50	6/6	−0.6	132	−0.38
	25	6/6	−0.6	130	−0.50
	12.5	6/6	−0.1	116	−1.39
26[i]	200	4/4	−0.7	105	
	100	4/4	−1.0	122	
	50	4/4	−0.4	118	
	25	4/4	−0.6	124	

[a] Determined under the auspices of the National Cancer Institute. For general screening procedures and data interpretation, see: Geran, R. I.; Greenburg, N. H.; McDonald, M. M.; Schumacher, A. M.; Abbott, B. *J. Cancer Chemother. Rep., Part* 3 **1972**, *3*(2), 1. [b] Ascitic fluid containing ca. 10^6 cells was inoculated into CD_2F_1 mice. The drugs were given daily beginning 24 h after tumor inoculation. Fresh solutions (or suspensions) were prepared daily. [c] TDS: toxicity day evaluations were carried out on day 5; number of survivors/number of animals in the initial test group are given. [d] BWD refers to the body weight difference (grams) of the test animals on day 5 (for P388 and L1210) or day 14 (M5076) minus day 1. [e] %T/C refers to the percent increase of the median survival time of test animals compared to control animals. The MX-1 %T/C refers to tumor size. A positive value is the change in the treated tumor size relative to control. [f] KE refers to tumor cell kill and is the log of the tumor cell population at the onset of treatment minus the log of the tumor cell population at the end of treatment. [g] The drug was given in a saline solution. [h] The drug was given as a suspension in saline. [i] Determined by Mitsubishi Kasei Corp., Tokyo, Japan.

M phosphate buffer (pH 7.4, 25 °C)] occurred with a half-life of 110 h (k_{obs} = 0.0063 h^{-1}), a rate much faster than that of the corresponding α-chloro compound, **18b**.

The water solubilities of the α-fluoropyridinium compound **17a** and the pyridone **19a** were measured. The pyridinium salt **17a** had good water solubility, 13 mg/mL, and the pyridone **19a** was soluble to the extent of 5 mg/mL.

Biological Results and Discussion

The data from the P388 lymphocytic leukemia assays in mice are given in Table VIII. These data show significant activity for the α-fluoropyridinium compounds **17a** and **17b** but not for the α-chloropyridinium compounds **18a** and **18b**. The pyridone **19a** that corresponds to the pyridinium salts **17a** and **18a** was also active against P388 lymphocytic leukemia.[13] The two α-fluoro compounds **17a** and **17b** and pyridone **19a** were also tested against an expanded panel of experimental murine neoplasias. These data are given in Table IX. Some activity was apparent, but overall the three compounds failed to show sufficient activity to merit further development.

(13) Pyridone **19b** was not submitted for antitumor evaluation.

Table IX. Activity of 17a, 17b, and 19a against Selected Murine Tumors in Vivo[a]

compd	tumor system[b]	dose, mg/kg	TDS[c]	BWD:[d] T − C, g	%T/C[e]	KE[f]
17a[g]	L1210	100	0/6			
		50	5/6	−0.8	118	−1.94
		25	6/6	0.6	118	−2.08
	M5076	100	6/10	0.0		
		50	10/10	1.4	118	−1.59
		25	10/10	0.4	104	−1.98
	MX-1	150	0/6			
		75	4/6	0.2	34	
		37.5	4/6	0.3	69	
17b[g]	L1210	100	0/6			
		50	6/6	−0.7	123	−2.03
		25	6/6	−0.7	119	−2.09
	M5076	180	3/10	−1.4		
		90	10/10	−1.8	125	−1.40
		45	10/10	−0.5	115	−1.79
	MX-1	180	0/6			
		90	6/6	0.5	18	
19a[h]	L1210	166.7	5/6	−3.8	110	−2.29
		100	6/6	−4.2	146	−0.22
		60	6/6	−3.2	141	−0.50
		36	6/6	−0.3	135	−0.86
		21.6	6/6	−0.3	135	−0.86
	M5076	166.7	4/10	−5.8		
		100	10/10	−1.3	122	−1.49
		60	10/10	0.9	114	−1.79
		36	10/10	1.8	117	−1.69
		21.6	10/10	1.7	107	−1.93

[a] See footnote *a* in Table VIII. [b] For L1210 lymphocytic leukemia: ascitic fluid containing ca. 10^6 cells was injected ip in CD_2F_1 mice; drug treatment began 24 h after tumor inoculation and continued daily for a total of five doses. For *M5067 Sarcoma*: ascitic fluid containing ca. 10^6 cells was inoculated ip in CD_2F_1 mice; drug treatment began 24 h after tumor inoculation and doses were then given every 4 days for a total of four doses. For *MX-1 mammary xenograft*: the solid tumor (1 mm^3, 0.5 mg) was introduced in a subrenal capsule implant in NCr-nu mice, the first dose was given 24 h after tumor implantation and, thereafter, every three days for a total of three doses. [c–h] See footnotes *c–h* in Table VIII.

The inactivity of the α-chloropyridinium compounds can be attributed to the slow activation (hydrolysis of the pyridinium moieties to the *N*-methylpyridones) relative to the faster inactivation (carbamate hydrolysis). This argument is supported by the fact that the 2-chloro-3-bromopyridinium compound **26** has low but significant activity against P388 (Table VIII). Compound **26** is hydrolyzed to the pyridone at a faster rate than **18b**, but still slower than either **17a** or **17b** (data not shown). The low activities of the α-fluoropyridinium salts **17a** and **17b** are probably due to the relatively small difference between the rates of activation (k_1, Scheme II) and inactivation [k_3 (and k_6), Scheme II]. The ratios of k_1/k_3 were 6.0 and 5.6 for **17a** and **17b**, respectively. A second potential problem may be that the reactive pyridones are too water soluble (5 mg/mL). Earlier studies have shown that reactive, water-soluble compounds in this class have little or no activity. The reactivities of the carbamates in the pyridones **19a** and **19b** were comparable to that of IPP (**1**), a very active antineoplastic agent (the relative reactivities of the carbamates in **19b**, **1**, and **19a** were 0.2, 1.0, and 3.3, respectively).

This study demonstrates the potential utility of the α-halopyridinium moiety in drug design. More lipophilic pyridones can be prepared by homologation of the *N*-alkyl substituents on the pyridinium salt and on the carbamates. The reactivity data will be used in future drug-design

studies in this and other classes of agents.

Experimental Section

Melting points (uncorrected) were determined in unsealed capillary tubes on a Thomas-Hoover Unimelt apparatus. Infrared spectra were obtained for Nujol mulls (unless otherwise noted) on a Nicolet 7000 FT-IR spectrophotometer. NMR spectra were obtained on either a Varian EM390 spectrometer at 90 MHz or a Varian T60A spectrometer at 60 MHz in DMSO-d_6 (unless otherwise specified) with tetramethylsilane (TMS) as an internal standard. Elemental analyses were performed by Atlantic Microlabs, Inc., of Atlanta, GA, and were within ±0.4% of the theoretical value when indicated by the symbols of the element (unless otherwise noted). Anhydrous solvents were distilled from the following drying agents: tetrahydrofuran, diethyl ether, and dimethoxyethane from sodium/benzophenone; dichloromethane, 1,2-dichloroethane, benzene, and acetonitrile from calcium hydride; acetone from anhydrous potassium carbonate. Flash chromatography was performed with Merck 60 silica gel (mesh 200–245). Products were dried in vacuo for 12 h in a desiccator over anhydrous calcium sulfate (Drierite).

2-Fluoropyridine-4-carbonyl Chloride. 2-Amino-4-methylpyridine was diazotized in 48% fluoroboric acid to give 2-fluoro-4-methylpyridine as a colorless liquid: bp 63–64 °C (27 Torr) (lit.[14] bp 157 °C); NMR ($CDCl_3$) δ 2.30 (s, 3 H), 6.72 (s, 1 H), 6.99 (d, J = 6 Hz, 1 H), 8.05 (d, J = 6 Hz, 1 H).

A stirred mixture of 2-fluoro-4-methylpyridine (30 g, 0.27 mol) and potassium permanganate (100 g, 0.635 mol) in water (1.2 L) was heated at reflux. Additional potassium permanganate (50 g, 0.317 mol) was added after 1.5 h when the purple mixture became black, and the stirred mixture was maintained at reflux for 15 h. The reaction mixture was steam distilled to remove unreacted starting material, the hot residual aqueous solution was filtered, and the filtrate was concentrated to 450 mL in vacuo. The solution was cooled on an ice bath and acidified with concentrated HCl to pH 2.0 (pH test paper). The resulting precipitate was collected and crystallized from water to give 2-fluoropyridine-4-carboxylic acid (13.56 g, 36%): mp 220–224 °C; NMR δ 7.49 (d, J = 1.5 Hz, 1 H), 7.73 (dd, J = 5.5 and 1.5 Hz, 1 H), 8.39 (d, J = 5.5 Hz, 1 H), 11.87 (br s, 1 H, exchangeable with deuterium oxide); IR 3150, 2500, 1860, 1725, 1610, 1590 cm^{-1}.

A mixture of 2-fluoropyridine-4-carboxylic acid (10 g, 0.071 mol) and thionyl chloride (70 mL) was heated at reflux for 25 h. The excess thionyl chloride was removed by distillation at atmospheric pressure and the residue was distilled to give 2-fluoropyridine-4-carbonyl chloride (9.51 g, 93%): bp 88 °C (20 Torr).

2-Chloropyridine-4-carbonyl Chloride. A mixture of pyridine-4-carboxylic acid N-oxide (28 g, 0.2 mol) and phosphorus oxychloride (120 g, 0.8 mol) was heated at reflux for 7 h. The reaction mixture was cooled and poured into ice–water (500 mL). The precipitate was collected and crystallized from a large volume of ethyl acetate to give 2-chloropyridine-4-carboxylic acid (23.1 g, 73%) as a white granular solid: mp 225–227 °C (lit.[14] mp 250 °C); NMR δ 7.80 (m, 2 H), 8.62 (d, J = 5.5 Hz, 1 H).

2-Chloropyridine-4-carboxylic acid was converted to 2-chloropyridine-4-carbonyl chloride (83%): bp 144–148 °C (10 Torr).

2-Fluoropyridine-5-carbonyl Chloride. 2-Amino-5-methylpyridine (200 g, 1.84 mol) was diazotized to give 2-fluoro-5-methylpyridine as a pale yellowish oil (43%): bp 70–74 °C (30 Torr) (lit.[14] bp 155–156 °C); NMR ($CDCl_3$) δ 2.40 (s, 3 H), 6.90 (dd, J = 8 and 2 Hz, 1 H), 7.60 (dd, J = 8 and 2 Hz, 1 H); IR (neat) 1592, 1494 cm^{-1}.

A solution of 2-fluoro-5-methylpyridine (30 g, 0.269 mol) in water (1.2 L) was stirred with a mechanical stirrer, potassium permanganate (100 g, 0.635 mol) was added in portions, and the mixture was heated to 100 °C (just below reflux). When the characteristic purple color of the mixture had faded to black, another portion of potassium permanganate (50 g, 0.3175 mol) was added and the mixture was then stirred at 100 °C for 4.5 h. The mixture was filtered hot; the filtrate was cooled and extracted with ether to remove unreacted starting material. The aqueous layer was neutralized to pH 7 (pH test paper) with concentrated HCl and concentrated to a volume of 350 mL. The pH was lowered by addition of concentrated HCl to pH 4–2 at which point a large amount of precipitate appeared. The precipitate was filtered and the aqueous layer was extracted with ethyl acetate (500 mL). The organic layer was dried and evaporated, and the solids were combined to give 2-fluoropyridine-5-carboxylic acid (17.1 g, 45%) as a colorless solid: mp 275–278 °C (lit.[14] mp 280–282 °C); NMR δ 7.25 (dd, J = 6 and 1.5 Hz, 1 H), 8.50 (dt, J = 6.0 and 1.5 Hz, 1 H), 8.95 (d, J = 1 Hz, 1 H), 12.95 (s, 1 H, exchangeable with deuterium oxide); IR (KBr) 3070, 1682, 1590 cm^{-1}.

2-Fluoropyridine-5-carboxylic acid was treated with thionyl chloride as described above to give 2-fluoropyridine-5-carbonyl chloride as a clear oil (86%): bp 90–94 °C (15 Torr); NMR ($CDCl_3$) δ 7.35 (dd, J = 8 and 2 Hz, 1 H), 8.60 (dt, J = 8 and 2 Hz), 9.00 (d, J = 1.5 Hz, 1 H); IR (neat) 1787, 1583, 1478 cm^{-1}.

***N*-[(2-Fluoropyridin-4-yl)carbonyl]pyrrolidine-2-carboxylic Acid (3a).** A solution of 2-fluoropyridine-4-carbonyl chloride (9.51 g, 0.0663 mol) in anhydrous acetone (35 mL) was added dropwise (with simultaneous addition of 2 N sodium hydroxide to maintain the reaction mixture at pH 8–9) to a stirred solution (pH 9–10) of *l*-proline (8.85 g, 0.0768 mol) in 2 N sodium hydroxide (38.9 mL), 1 N sodium bicarbonate (56 mL), and acetone (70 mL) at 0 °C. The mixture was stirred for 2 h at 0 °C and then at room temperature for 1 h. The reaction mixture was concentrated to $^1/_3$ of the original volume in vacuo, cooled, and acidified with concentrated HCl (pH < 3). The mixture was extracted with dichloromethane (3 × 250 mL). The combined organic phase was washed with saline (2 × 100 mL), dried (sodium sulfate), and concentrated in vacuo. The oily residue was cooled and treated with *n*-hexane. The resulting solid was washed with *n*-hexane and crystallized from dichloromethane–*n*-hexane to give **3a** as a white, granular solid (13.8 g, 87%): mp 120–123 °C; NMR ($CDCl_3$) δ 2.15 (m, 4 H), 3.53 (m, 2 H), 4.73 (m, 2 H), 7.10 (d, J = 2 Hz, 1 H), 7.35 (dd, J = 5 and 2 Hz, 1 H), 8.33 (d, J = 5 Hz, 1 H); IR 3065, 2570, 1725, 1590 cm^{-1}.

***N*-[(2-Fluoropyridin-5-yl)carbonyl]pyrrolidine-2-carboxylic Acid (3b).** *l*-Proline (9.28 g, 0.0806 mol) was dissolved in distilled water (120 mL), acetone (60 mL) was added, and the solution was cooled to ice-bath temperature. The pH was adjusted to 8–9 (pH test paper) with 1 N sodium hydroxide and a solution of the 2-fluoropyridine-5-carbonyl chloride (10 g, 0.0696 mol) in acetone (40 mL) was added dropwise. Sodium hydroxide solution (2 N) was also added to keep the pH at 8–9 and the mixture was allowed to stir at room temperature for 15 h. The mixture was extracted with ether (100 mL), acidified to pH 2 with concentrated HCl, and extracted with ethyl acetate (2 × 400 mL). The ethyl acetate layer was washed with water (2 × 150 mL) and brine (150 mL) and dried (sodium sulfate), and the solvent was removed in vacuo to yield **3b** as a hygroscopic pinkish oil (11.0 g, 67%): NMR ($CDCl_3$) δ 2.00 (m, 2 H), 2.41 (m, 2 H), 3.62 (t, J = 6 Hz, 2 H), 4.45 (m, 1 H), 7.35 (dd, J = 8 and 2 Hz, 1 H), 8.20 (dt, J = 8.0 and 2.0 Hz, 1 H), 8.55 (s, 1 H).

***N*-[(2-Chloropyridin-4-yl)carbonyl]pyrrolidine-2-carboxylic Acid (4a).** 2-Chloropyridine-4-carbonyl chloride was treated with *l*-proline as described for **3a** to give 4a as a white, granular solid (dichloromethane–benzene, 94%): mp 129–131 °C; NMR ($CDCl_3$) δ 2.15 (br s, 4 H), 3.52 (m, 2 H), 4.70 (br t, J = 7 Hz, 2 H), 7.35 (s, 1 H), 7.64 (s, 1 H), 8.50 (d, J = 5 Hz, 1 H), 11.16 (s, 1 H, exchangeable with deuterium oxide); IR 3075, 2772, 2715, 1727, 1607, 1537, 1205, 1184 cm^{-1}.

***N*-[(2-Chloropyridin-5-yl)carbonyl]pyrrolidine-2-carboxylic Acid (4b).** *l*-Proline was acylated with 2-chloropyridine-5-carbonyl chloride [prepared from the acid by treatment with thionyl chloride (bp 128–130 °C, 23 Torr)] as described for the preparation of 3b to give **4b** as a hygroscopic colorless oil (90%): NMR ($CDCl_3$) δ 2.14 (m, 4 H), 3.59 (m, 2 H), 4.68 (m, 1 H), 7.37 (d, J = 8 Hz, 1 H), 7.90 (dd, J = 8.0 and 2.0 Hz, 1 H), 8.58 (d, J = 2 Hz, 1 H), 9.91 (s, 1 H, exchangeable with deuterium oxide).

Dimethyl 2,3-Dihydro-5-(4-pyridinyl)-1*H*-pyrrolizine-6,7-dicarboxylate (5a). A. Reduction with Zinc–Acetic Acid. A mixture of diester **6a** (1.16 g, 3.467 mmol) and zinc powder (15 g) in glacial acetic acid (40 mL) was stirred at room temperature for 48 h. Dichloromethane (200 mL) was added and the mixture

(14) (a) Minor, J. T.; Hawkins, G. F.; van der Were, A.; Roe, A. *J. Am. Chem. Soc.* **1949**, *71*, 1125. (b) Kappen, T. *Montash. Chem.* **1968**, *98*, 1858.

was filtered. Saturated sodium carbonate solution was added to the filtrate until the aqueous phase was alkaline (pH test paper). The residue was crystallized from ethyl acetate–*n*-hexane to give **5a** (0.63 g, 61%) as colorless fine needles: mp 180–181 °C; NMR ($CDCl_3$) δ 2.65 (q, J = 7 Hz, 2 H), 3.07 (t, J = 7 Hz, 2 H), 3.78 (s, 6 H), 4.03 (t, J = 7 Hz, 2 H), 7.28 (dd, J = 6 and 1.5 Hz, 2 H), 8.57 (dd, J = 6 and 1.5 Hz, 2 H); IR 1727, 1699, 1593 cm^{-1}. Anal. ($C_{16}H_{16}N_2O_4$) C, H, N.

B. Catalytic Hydrogenolysis. A solution of the diester **6a** (2 g, 5.97 mmol) in absolute ethanol (75 mL) was added to a suspension of 10% palladium on carbon (0.4 g) in 2 N HCl (15 mL). The mixture was mechanically shaken for 10 h at 35 psi of hydrogen atmosphere. The mixture was filtered through a pad of Celite and the ethanol was removed in vacuo. Sodium carbonate was added to the aqueous solution to make it basic (pH 9, pH test paper), and the solution was extracted with dichloromethane (3 × 150 mL). The organic solution was dried (sodium sulfate) and concentrated in vacuo. The slightly yellow-white solid residue was subjected to flash chromatography (ethyl acetate) to give **5a** (1.345 g, 76%) as a white solid. This compound was identical with that obtained by using method A.

Dimethyl 2,3-Dihydro-5-(3-pyridinyl)-1*H*-pyrrolizine-6,7-dicarboxylate (5b). Treatment of **6b** as in method B used for the synthesis of **5a** gave **5b** (80%) as a white solid: mp 180–182 °C; NMR ($CDCl_3$) δ 2.62 (q, J = 7 Hz, 2 H), 3.10 (t, J = 7 Hz, 2 H), 3.75 (s, 3 H), 3.82 (s, 3 H), 4.00 (t, J = 7 Hz, 2 H), 7.33 (dd, J = 8 and 4.5 Hz, 1 H), 7.78 (dd, J = 8 and 4.5 Hz, 1 H), 8.57 (dd, J = 4.5 and 1.5 Hz, 1 H), 8.65 (d, J = 1.5 Hz, 1 H); IR 1727, 1706, 1586, 1558 cm^{-1}. Anal. ($C_{16}H_{16}N_2O_4$) C, H, N.

Dimethyl 2,3-Dihydro-5-(2-fluoropyridin-4-yl)-1*H*-pyrrolizine-6,7-dicarboxylate (6a). A stirred mixture of **3a** (14.57 g, 0.0612 mol), dimethyl acetylenedicarboxylate (26.08 g, 0.1836 mol), and acetic anhydride (260 mL) was heated at 75 °C for 22 h until carbon dioxide evolution had ceased. The volatiles were removed from the reaction mixture in vacuo (75 °C) and the oily residue was purified by flash chromatography (hexanes–ethyl acetate, 1:1). The product was crystallized from methanol to give **6a** as fine needles (11.6 g, 60%): mp 142–143 °C; NMR ($CDCl_3$) δ 2.55 (q, J = 6.5 and 2 Hz, 2 H), 3.06 (t, J = 6.5 Hz, 2 H), 3.80 (s, 6 H), 4.06 (t, J = 6.5 Hz, 2 H), 6.95 (d, J = 2 Hz, 1 H), 7.20 (dd, J = 5 and 2 Hz, 1 H), 8.15 (d, J = 5 Hz, 1 H); IR 1720, 1615 cm^{-1}. Anal. ($C_{16}H_{15}N_2O_4F$) C, H, N.

Dimethyl 2,3-Dihydro-5-(2-fluoropyridin-5-yl)-1*H*-pyrrolizine-6,7-dicarboxylate (6b). A mixture of **3b** (11 g, 0.04617 mol), dimethyl acetylenedicarboxylate (9.84 g, 0.06925 mol), and acetic anhydride (130 mL) was heated at 68 °c for 15 h until evolution of carbon dioxide had ceased. The product was isolated as described above to give **6b** as colorless crystals (10.5 g, 72%): mp 135–138 °C; NMR ($CDCl_3$) δ 2.60 (m, 2 H), 3.15 (t, J = 6 Hz, 2 H), 3.81 (s, 3 H), 3.90 (s, 3 H), 4.05 (t, J = 6 Hz, 2 H), 7.05 (dd, J = 8 and 2 Hz, 1 H), 8.00 (dt, J = 8 and 2 Hz, 1 H), 8.40 (s, 1 H); IR 1721, 1706, 1586, 1522 cm^{-1}. Anal. ($C_{16}H_{15}N_2O_4F$) C, H, N.

Dimethyl 2,3-Dihydro-5-(2-chloropyridin-4-yl)-1*H*-pyrrolizine-6,7-dicarboxylate (7a). α-Amido acid **4a** was treated as described in **6a** to give **7a** as yellow plates (methanol, 16%): mp 128–130 °C; NMR ($CDCl_3$) δ 2.56 (m, 2 H), 3.06 (q, J = 7.0 Hz, 2 H), 3.80 (s, 6 H), 4.06 (t, J = 7.0 Hz, 2 H), 7.25 (m, 2 H), 8.34 (d, J = 5.5 Hz, 1 H); IR 1713, 1593, 1516, 1170 cm^{-1}. Anal. ($C_{16}H_{15}N_2O_4Cl$) C, H, N.

Dimethyl 2,3-Dihydro-5-(2-chloropyridin-5-yl)-1*H*-pyrrolizine-6,7-dicarboxylate (7b). α-Amido acid **4b** was treated as described in **6b** to give **7b** (65%): mp 164–166 °C; NMR ($CDCl_3$) δ 2.60 (q, J = 6 Hz, 2 H), 3.08 (t, J = 6 Hz, 2 H), 3.76 (s, 3 H), 3.79 (s, 3 H), 4.01 (t, J = 6 Hz, 2 H), 7.32 (d, J = 8 Hz, 1 H), 7.75 (dd, J = 8 and 2.5 Hz, 1 H), 8.39 (d, J = 2.5 Hz, 1 H); IR 1723, 1692, 1558 cm^{-1}. Anal. ($C_{16}H_{15}N_2O_4Cl$) C, H, N.

2,3-Dihydro-5-(4-pyridinyl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine (8a). Diester **5a** (2.0 g, 6.66 mmol) was added portionwise over a period of 0.5 h to a stirred suspension of lithium aluminum hydride (0.632 g, 13.2 mmol) in anhydrous tetrahydrofuran (50 mL) that was cooled to −15 °C (ethylene glycol–dry ice) and maintained under an argon atmosphere. The mixture was stirred for 2 h at −15 °C. The excess hydride was destroyed by the cautious addition of 5% sodium hydroxide solution (10 mL). Dichloromethane (200 mL) was added, the mixture was filtered, and the inorganic precipitate was washed with hot dichloromethane (200 mL). The combined filtrate was washed with brine, dried (sodium sulfate), and concentrated in vacuo. The solid residue was dried (over phosphorus pentoxide in vacuo) to give **8a** (1.6 g, 98%) that was used immediately in the next step. Crude **8a**: mp 173–175 °C; NMR δ 2.45 (m, 2 H), 2.77 (m, 2 H), 4.04 (t, J = 6 Hz, 2 H), 4.53 (d, J = 4.5 Hz, 6 H), 7.44 (dd, J = 6.0 and 1.5 Hz, 2 H), 8.50 (dd, J = 6.0 and 1.5 Hz, 2 H); IR 3336, 1607, 1000 cm^{-1}.

2,3-Dihydro-5-(2-fluoropyridin-4-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine (9a). Diester **6a** was reduced as described for **8a** to give **9a** as white crystals (dichloromethane–benzene, 85%): mp 135–139 °C; NMR δ 2.47 (q, J = 6.5 Hz, 2 H), 2.79 (t, J = 6.5 Hz, 2 H), 4.05 (t, J = 6 Hz, 2 H), 4.35 (m, 5 H), 4.79 (t, J = 4.5 Hz, 1 H), 7.21 (s, 1 H), 7.41 (m, 1 H), 8.12 (d, J = 5.5 Hz, 1 H); IR 3379, 1614, 1548, 1516, 1487, 1000 cm^{-1}. Anal. ($C_{14}H_{15}N_2O_2F$) C, H, N.

2,3-Dihydro-5-(2-chloropyridin-4-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine (10a). Diester **7a** was reduced as described for **8a** to give **10a** as pale yellow crystals (dichloromethane–*n*-hexane, 91%): mp 145–149 °C; NMR δ 2.50 (m, 2 H), 2.82 (m, 2 H), 3.72 (s, J = 6 Hz, 4 H), 4.10 (t, J = 6 Hz, 2 H), 4.42 (d, J = 3.5 Hz, 2 H), 7.51 (dd, J = 5.5 and 1.5 Hz, 1 H), 7.69 (s, 1 H), 8.34 (d, J = 5.5 Hz, 1 H); IR 3330 cm^{-1}.

2,3-Dihydro-5-(4-pyridinyl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (11a). A mixture of diol **8a** (1.6 g, 0.655 mmol), 2-propyl isocyanate (1.23 g, 1.447 mmol), and dibutyltin diacetate (2 drops) in anhydrous dichloromethane (50 mL) was stirred at room temperature under an argon atmosphere for 15 h. The mixture was concentrated in vacuo (<30 °C) and the residue was subjected to flash chromatography (ethyl acetate) to give **11a** as an amorphous white powder (0.66 g, 41%): mp 160–162 °C; NMR ($CDCl_3$) δ 1.04 (d, J = 6.5 Hz, 2 H), 2.53 (m, 2 H), 2.89 (m, J = 6 Hz, 2 H), 3.79 (m, J = 6.5 Hz, 2 H), 3.99 (t, J = 6.5 Hz, 2 H), 4.71 (br d, J = 7 Hz, 2 H), 5.05 (s, 4 H), 7.24 (dd, J = 6 and 1.5 Hz, 2 H), 8.55 (dd, J = 6 and 1.5 Hz, 1 H); IR 3336, 1706, 1685, 1664, 1600, 1530, 1247, 1078, 1057 cm^{-1}. Anal. ($C_{22}H_{30}N_4O_4$) C, H, N.

2,3-Dihydro-5-(3-pyridinyl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (11b). Diester **5c** was reduced as described for **8a** and the crude product was immediately suspended in anhydrous dichloromethane (60 mL) and carbamoylated as described for **11a** (24-h reaction time) to give **11b** as colorless needles (76% over two steps): mp 156–158 °C; NMR ($CDCl_3$) δ 1.23 (d, J = 6 Hz, 12 H), 2.53 (q, J = 6.5 Hz, 2 H), 2.91 (t, J = 6.5 Hz, 2 H), 3.80 (m, J = 6.5 Hz, 4 H), 4.56 (m, 2 H), 5.01 (s, 2 H), 5.04 (s, 2 H), 7.30 (m, 1 H), 7.70 (m, 1 H), 8.75 (m, 1 H); IR 3336, 1706, 1684, 1663, 1529, 1247, 1078 cm^{-1}. Anal. ($C_{22}H_{30}N_4O_4$) C, H, N.

2,3-Dihydro-5-(2-fluoropyridin-4-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (12a). Diol **9a** was acylated as described for **11a** to give **12a** as fine needles (ethyl acetate, 77%): mp 160–162 °C; NMR ($CDCl_3$) δ 1.15 (d, J = 6.5 Hz, 12 H), 2.58 (q, J = 6.5 Hz, 2 H), 2.93 (t, J = 6.5 Hz, 2 H), 3.80 (m, 2 H), 4.03 (m, 2 H), 4.53 (br s, 1 H), 5.07 (s, 4 H), 6.90 (s, 1 H), 7.20 (m, 1 H), 8.20 (d, J = 5 Hz, 1 H); IR (Nujol) 3320, 1695, 1680, 1615 cm^{-1}. Anal. ($C_{22}H_{29}N_4O_4F$) C, H, N.

2,3-Dihydro-5-(2-fluoropyridin-5-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (12b). Diester **6b** was reduced and the resulting diol was carbamoylated, as described for **11a**, to give **12b** (85%, colorless needles from ethyl acetate): mp 178–180 °C; NMR ($CDCl_3$) δ 1.15 (d, J = 6 Hz, 12 H), 2.50 (m, 2 H), 3.03 (t, J = 6.0 Hz, 2 H), 3.6 (m, 2 H), 3.80 (m, J = 6 Hz, 2 H), 4.52 (s, 2 H), 5.00 (d, J = 4 Hz, 2 H), 7.02 (dd, J = 8 and 2 Hz, 1 H), 7.70 (dt, J = 8 and 2 Hz, 1 H), 8.20 (s, 1 H); IR (KBr) 3344, 1710, 1670, 1538 cm^{-1}. Anal. ($C_{22}H_{29}N_4O_4F$) C, H, N.

2,3-Dihydro-5-(2-chloropyridin-4-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (13a). Diol **10a** was acylated as described for **11a** to give **13a** as an amorphous white powder (isopropyl ether, 41%): mp 145–147 °C; NMR ($CDCl_3$) δ 1.15 (d, J = 6.5 Hz, 12 H), 2.60 (m, 2 H), 2.93 (m, 2 H), 3.88 (m, 2 H), 4.18 (br m, 2 H), 5.08 (s, 4 H), 6.90 (s, 1 H), 7.2–7.45 (m, 2 H), 8.34 (dd, J = 5 and 1.5 Hz, 1 H); IR 3386, 1593, 1473, 1000 cm^{-1}. Anal. ($C_{22}H_{29}N_4O_4Cl$) C, H, N.

2,3-Dihydro-5-(2-chloropyridin-5-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (13b). Diester **7b** was reduced and the resulting diol was carbamoylated as described for **11b** to give **13b** as colorless needles [flash chromatography (*n*-hexane–ethyl acetate, 1.5:1), 60% over two steps]: mp 174–176 °C; NMR ($CDCl_3$) δ 1.14 (d, J = 6.5 Hz, 12 H), 2.53 (m, 2 H), 2.90 (t, J = 6.0 Hz, 2 H), 3.58 (m, 4 H), 4.56 (br d, J = 8 Hz, 2 H), 4.99 (s, 2 H), 5.01 (s, 2 H), 7.30 (d, J = 8 Hz, 1 H), 7.64 (dd, J = 8 and 2 Hz, 1 H), 8.36 (d, J = 2 Hz, 1 H); IR (KBr) 3336, 1706, 1685, 1664 cm^{-1}.

1-Methyl-2-fluoro-4-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Trifluoromethanesulfonate (14a). Methyl trifluoromethanesulfonate (0.228 g, 1.387 mmol) was added to a stirred solution of **12a** (0.50 g, 1.156 mmol) in anhydrous dichloromethane (10 mL) at room temperature under an argon atmosphere. The mixture was stirred for 4 h at room temperature, anhydrous ether (50 mL) was added, and the mixture was stirred for an additional 1 h at room temperature. The resulting precipitate was filtered, washed with anhydrous ether (50 mL), and dried in vacuo. Reprecipitation from anhydrous dichloromethane–ether gave **14a** (0.660 g, 96%) as a yellow powder: mp ca. 70 °C (wet and bubbled) to 125 °C (complete dec); NMR (CD_3CN) δ 1.10 (d, J = 6.5 Hz, 12 H), 2.60 (q, J = 7 Hz, 2 H), 3.00 (t, J = 6 Hz, 2 H), 3.35 (m, 2 H), 4.06 (d, J = 3 Hz, 3 H), 4.25 (t, J = 7 Hz, 2 H), 5.10 (s, 2 H), 5.13 (s, 2 H), 5.3–5.9 (br s, 2 H), 7.6–7.9 (m, 2 H), 8.4 (m, 1 H); IR 3320, 1700, 1615, 1250, 1155 cm^{-1}. Anal. ($C_{24}H_{32}N_4O_7F_4S$) C, H, N.

1-Methyl-2-fluoro-5-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Trifluoromethanesulfonate (14b). Pyridine **12b** was methylated, as described for **14a**, to give **14b** (75%) as a hygroscopic yellow powder: mp ca. 80 °C (wet and bubbled) to 99 °C (complete dec); NMR (CD_3CN) δ 1.10 (d, J = 6 Hz, 12 H), 2.45 (m, 2 H), 2.90 (t, J = 6 Hz, 2 H), 3.60 (m, 2 H), 4.0 (m, 2 H), 4.30 (d, J = 4 Hz, 3 H), 5.00 (s, 2 H), 5.02 (s, 2 H), 5.50 (br s, 2 H), 7.95 (dd, J = 8 and 2 Hz, 1 H), 8.60–8.95 (m, 2 H); IR 3329, 1706, 1551, 1261, 1155 cm^{-1}.

1-Methyl-2-chloro-4-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Trifluoromethanesulfonate (15a). Pyridine **13a** was methylated as described for **14a** to give **15a** (95%) mp 83–87 °C; NMR (CD_3CN) δ 1.12 (d, J = 6.0 Hz, 12 H), 2.65 (m, J = 6.5 Hz, 2 H), 3.00 (m, 4 H), 3.62 (m, 2 H), 4.40 (m, 7 H), 5.07 (s, 2 H), 5.11 (s, 2 H), 8.0–8.3 (m, 2 H), 8.92 (d, J = 6.5 Hz, 1 H); IR 3329, 1699, 1621, 1529, 1247, 1155 cm^{-1}.

1-Methyl-2-chloro-5-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Trifluoromethanesulfonate (15b). Pyridine **13b** was methylated as described for **14a** to give **15b** (90%) as a hygroscopic yellow powder: mp ca. 70 °C (wet and bubbled) to 89 °C (complete dec); NMR δ 1.06 (d, J = 6.5 Hz, 12 H), 2.26 (m, 2 H), 2.85 (br d, J = 6 Hz, 2 H), 3.57 (m, 2 H), 4.05 (br t, J = 6.0 Hz, 2 H), 4.36 (s, 3 H), 4.95 (s, 4 H), 6.82 (br t, J = 7 Hz, 2 H), 8.32 (d, J = 8 Hz, 1 H), 8.57 (dd, J = 8 and 2 Hz, 1 H), 9.16 (d, J = 2 Hz, 1 H); IR 3322, 1692, 1621, 1529, 1459, 1254 cm^{-1}.

1-Methyl-4-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Iodide (16a). A solution of **11a** (0.20 g, 0.4825 mmol) and iodomethane (2 mL) in anhydrous acetone (30 mL) was stirred at room temperature for 27 h under an argon atmosphere. A yellow precipitate appeared. Anhydrous ether (100 mL) was added; the mixture was stirred for 0.5 h and filtered. The solid was washed with anhydrous ether (100 mL) and dried in vacuo over phosphorus pentoxide. Reprecipitation from anhydrous dichloromethane–ether gave **16a** as a yellow powder (0.25 g, 93%): mp 209–211 °C dec; NMR δ 1.03 (d, J = 6.5 Hz, 12 H), 2.48 (m, 2 H), 2.87 (m, 2 H), 3.57 (m, J = 6.5 Hz, 2 H), 4.23 (m, 5 H), 4.97 (s, 2 H), 5.00 (s, 2 H), 6.90 (br t, J = 7 Hz, 2 H), 8.01 (dd, J = 7 and 1.5 Hz, 2 H), 8.60 (dd, J = 7 and 1.5 Hz, 2 H); IR 3329, 1685, 1643, 1523, 1261 cm^{-1}. Anal. ($C_{23}H_{33}N_4O_4I$) C, H, N.

1-Methyl-3-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Iodide (16b). Bis-carbamate **11b** was methylated as described for **16a** to give **16b** as a yellow powder (0.26 g, 97%): mp 187–190 °C dec; NMR δ 1.10 (d, J = 6 Hz, 12 H), 2.49 (m, 2 H), 2.83 (m, 2 H), 3.56 (m, J = 6 Hz, 2 H), 4.08 (br t, J = 6 Hz, 2 H), 4.41 (s, 3 H), 4.93 (s, 4 H), 6.81 (br t, J = 6.5 Hz, 2 H), 8.12 (dd, J = 8 and 5.5 Hz, 1 H), 8.56 (d, J = 8 Hz, 1 H), 8.86 (br d, J = 5.5 Hz, 1 H), 9.03 (br s, 1 H); IR 3349, 1713, 1692, 1664, 1254 cm^{-1}.

1-Methyl-2-fluoro-4-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium Iodide (17a). A solution of anhydrous sodium iodide (0.160 g, 1.0 mmol) in anhydrous acetone (20 mL) was stirred at room temperature for 25 min. Trifluoromethanesulfonate salt **14a** (0.509 g, 0.833 mmol) was added portionwise and the reaction mixture was stirred at room temperature for 3 h under an argon atmosphere. Anhydrous ether (50 mL) was added and the resulting precipitate was collected and washed with anhydrous ether (100 mL). The product was dissolved in anhydrous 1,2-dichloroethane (20 mL), filtered through a pad of Celite, and reprecipitated with anhydrous ether (100 mL) to give **17a** as a slightly brownish-yellow powder (0.36 g, 73%): mp 100 °C (wet and bubbled) to 120 °C (dec); NMR δ 1.10 (d, J = 6.0 Hz, 12 H), 2.53 (m, 2 H), 2.90 (d, J = 6 Hz, 2 H), 3.53 (m, 2 H), 4.14 (d, J = 4 Hz, 3 H), 4.30 (m, 2 H), 5.00 (br s, 4 H), 6.95 (br t, J = 8 Hz, 2 H), 7.80 (s, 1 H), 7.90 (s, 1 H), 8.70 (br t, J = 5 Hz, 1 H); IR 3330, 1690, 1645 cm^{-1}. Anal. ($C_{23}H_{32}N_4O_4FI$) C, H, N.

1-Methyl-2-fluoro-5-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizinyl]pyridinium Iodide (17b). Salt **14b** was treated with sodium iodide for 3 h, as described for **17a**, to give **17b** as a light yellow powder (88%): mp 136 °C (wet and bubbled) to 150 °C (dec); NMR (CD_3CN) δ 1.10 (d, J = 6.0 Hz, 12 H), 2.50 (m, 2 H), 2.90 (t, J = 6 Hz, 2 H), 3.70 (m, 2 H), 4.10 (m, 2 H), 4.35 (d, J = 4 Hz, 3 H), 5.02 (s, 2 H), 5.05 (s, 2 H), 5.60 (br s, 2 H), 7.95 (dd, J = 8 and 2 Hz, 1 H), 8.69–9.0 (m, 2 H); IR 3357, 1685, 1537, 1254 cm^{-1}. Anal. ($C_{23}H_{32}N_4O_4FI$) C, H, N.

1-Methyl-2-chloro-4-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizinyl]pyridinium Iodide (18a). Salt **15a** was treated with sodium iodide for 1 h, as described for **17a**, to give **18a** (85%) as a light yellow powder: mp 149 °C (wet and bubbled) to 180 °C (dec); NMR δ 1.15 (d, J = 6.0 Hz, 12 H), 2.50 (m, 2 H), 2.88 (m, J = 6 Hz, 2 H), 3.53 (m, J = 6 Hz, 2 H), 4.23 (m, 5 H), 4.95 (s, 2 H), 5.02 (s, 2 H), 6.92 (br t, J = 8.5 Hz, 2 H), 7.98 (dd, J = 6 and 2 Hz, 1 H), 8.14 (d, J = 2 Hz, 1 H), 8.92 (d, J = 6 Hz, 1 H); IR 3343, 1692, 1621 cm^{-1}. Anal. ($C_{23}H_{32}N_4O_4ClI$) C, H, N.

1-Methyl-2-chloro-5-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizinyl]pyridinium Iodide (18b). Salt **15b** was treated with sodium iodide for 3 h, as described for **17a**, to give **18b** as a light yellow powder (87%): mp 160 °C (wet and bubbled) to 190 °C (dec); NMR δ 1.07 (d, J = 6.5 Hz, 12 H), 2.45 (m, 2 H), 2.84 (br t, J = 6 Hz, 2 H), 3.58 (m, 2 H), 4.11 (br t, J = 6 Hz, 2 H), 4.38 (s, 3 H), 4.95 (s, 4 H), 6.84 (br t, J = 7 Hz, 2 H), 8.34 (d, J = 9 Hz, 1 H), 8.62 (dd, J = 9 and 2 Hz, 1 H), 9.20 (d, J = 2 Hz, 1 H); IR 3350, 1685, 1621, 1530 cm^{-1}. Anal. ($C_{23}H_{32}N_4O_4ClI$) C, H, N.

2,3-Dihydro-5-(1-methyl-1,2-dihydro-2-oxopyridin-4-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (19a). Saturated sodium bicarbonate (10 mL) was added to a solution of **14a** (3 g, 5.03 mmol) in acetone (30 mL) at room temperature. The mixture was stirred at room temperature for 0.5 h and then concentrated in vacuo. The residue was subjected to flash chromatography (ethyl acetate) to give **19a** as slightly yellow fine crystals (1.58 g, 71%): mp 174–176 °C; NMR ($CDCl_3$) δ 1.13 (d, J = 6 Hz, 12 H), 2.5 (m, 2 H), 2.85 (m, J = 6.5 Hz, 2 H), 3.5 (m, 5 H), 3.90 (m, J = 6.5 Hz, 2 H), 4.56 (m, 2 H), 4.99 (s, 4 H), 6.21 (dd, J = 7.5 and 1.5 Hz, 1 H), 6.41 (d, J = 1.5 Hz, 1 H), 7.15 (dd, J = 7.5 and 1.5 Hz, 1 H); IR 3336, 1691, 1656, 1600 cm^{-1}. Anal. ($C_{23}H_{32}N_4O_5$) C, H, N.

2,3-Dihydro-5-(1-methyl-1,2-dihydro-2-oxopyridin-5-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine Bis(*N*-2-propylcarbamate) (19b). Hydrolysis of **14b** as described for **19a** (6 h reaction time) gave **19b** as colorless granular crystals (dichloromethane–benzene, 52.5 mg, 7%): mp 175–179 °C; NMR δ 1.10 (d, J = 6.5 Hz, 12 H), 2.45 (m, 2 H), 2.86 (t, J = 6.0 Hz, 2 H), 3.55 (m, 5 H), 3.95 (t, J = 6.0 Hz, 2 H), 4.92 (s, 4 H), 6.35 (m, 2 H), 6.80 (m, 2 H), 7.78 (d, J = 6 Hz, 1 H); IR 3306, 3047, 1696, 1654, 1591, 1059 cm^{-1}. Anal. ($C_{23}H_{32}N_4O_5$) C, H, N.

2,3-Dihydro-5-(1-methyl-1,2-dihydro-2-oxopyridin-4-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine (21a). Pyridinium

triflate **14a** (2.55 g, 4.27 mmol) was suspended in 100 mL of 1 N sodium bicarbonate and stirred at room temperature for 24 h. The solution was filtered and continuously extracted with dichloromethane (700 mL) for 24 h. The dichloromethane extract was concentrated in vacuo and the resulting tan residue was crystallized from hot dichloromethane to give **21a** as colorless crystals (0.50 g, 43%): mp 174–176 °C; NMR δ 2.45 (m, 2 H), 2.85 (t, J = 6 Hz, 2 H), 3.5 (s, 3 H), 4.0 (t, J = 6 Hz, 2 H), 4.46 (m, 6 H, two exchangeable with deuterium oxide), 6.42 (d, J = 3 Hz, 2 H), 7.53 (d, J = 6.5 Hz, 1 H); IR (KBr) 3407, 3153, 2955, 1642, 1564, 1006 cm^{-1}. Anal. ($C_{15}H_{18}N_2O_3 \cdot 0.5H_2O$) C, H, N.

Synthesis of the Ethyl Pyridine-4-carboxylate Precursors for 24a–d. 2-Bromo-4-methylpyridine was prepared from 2-amino-4-methylpyridine[15] and oxidized with permanganate to give 2-bromopyridine-4-carboxylic acid.[16] 2,6-Dichloropyridine-4-carboxylic acid was prepared from citrazinic acid.[17] The syntheses of 2-fluoro- and 2-chloropyridine-4-carboxylic acids were described in an earlier section. A solution of the appropriate carboxylic acid in anhydrous dichloromethane was treated with ethanol (1 equiv), 1,3-dicyclohexylcarbodiimide (1 equiv), and 4-(dimethylamino)-pyridine (0.15 equiv) and the mixture was stirred at room temperature for 6–24 h. The esters were purified by flash chromatography (*n*-hexane–ethyl acetate, 4:1 to 6:1).

Synthesis of the Pyridine Precursors to 25a–d. 2-Fluoro-[14] and 2-chloro-5-methylpyridine[18] were prepared from 2-amino-5-methylpyridine. 2,3-Dichloro-5-methylpyridine was prepared in a cuprous chloride–triphenylphosphine-induced cyclization of dichloroacetonitrile and methacrolein.[19,20] Bromination of 2-amino-5-methylpyridine and diazotization–chlorination gave 2-chloro-3-bromo-5-methylpyridine.[21]

Synthesis of the Pyridinium Salts 24 and 25. The appropriate pyridine was dissolved in anhydrous dichloromethane; the solution was cooled to 0 °C and treated with a slight excess of methyl trifluoromethanesulfonate and stirred for 0.5 h. The mixture was then stirred at room temperature for 3 h (5 h for **24c**, and 15 h for **24d** and **25d**). Anhydrous ether was added, the precipitate was collected and washed with ether.

Compound **24a** (80%): mp 100–103 °C; NMR (CD_3CN) δ 1.40 (t, J = 7 Hz, 3 H), 4.25 (d, J = 4 Hz, 3 H), 4.45 (q, J = 7 Hz, 2 H), 8.20 (m, 2 H), 8.75 (m, 1 H); IR (KBr) 3083, 1730, 1653, 1590, 1477, 1033 cm^{-1}. Anal. ($C_{10}H_{11}NO_5F_4S$) C, H, N.

Compound **24b** (78%): mp 79–81 °C; NMR (CD_3CN) δ 1.45 (t, J = 7 Hz, 3 H), 4.40 (d, J = 4 Hz, 3 H), 4.48 (q, J = 7 Hz, 2 H), 8.30 (dd, J = 2 Hz, 2 H), 8.50 (d, J = 2 Hz, 1 H), 9.0 (d, J = 4 Hz, 1 H); IR (KBr) 2985, 1727, 1583, 1456, 1111 cm^{-1}. Anal. ($C_{10}H_{11}NO_5F_3SCl$) C, H, N.

Compound **24c** (97%): mp 80–82 °C; NMR (CD_3CN) δ 1.40 (t, J = 7 Hz, 3 H), 4.45 (m, 5 H), 8.30 (dd, J = 6 and 2 Hz, 1 H), 8.75 (d, J = 2 Hz, 1 H), 9.10 (d, J = 6 Hz, 1 H); IR (KBr) 2990, 1725, 1592, 1464, 1102 cm^{-1}. Anal. ($C_{10}H_{11}NO_5F_3SBr$) C, H, N.

Compound **24d** (75%): mp 93–95 °C; NMR (CD_3CN) δ 1.45 (t, J = 7 Hz, 3 H), 4.45 (m, 5 H), 8.45 (s, 2 H); IR (KBr) 3091, 1731, 1555, 1407, 1153 cm^{-1}. Anal. ($C_{10}H_{11}NO_5Cl_2S$) C, H, N.

Compound **25a** (82%) was obtained as a clear viscous oil: NMR (CD_3CN) δ 2.45 (s, 3 H), 4.20 (d, J = 4 Hz, 3 H), 7.75 (dd, J = 8 and 4 Hz, 1 H), 8.45 (m, 2 H); IR (neat) 2983, 1649, 1543, 1268, 1148, 1028 cm^{-1}. Anal. ($C_8H_9O_3NF_4S$) C, H, N.

Compound **25b** (97%): mp 58–60 °C; NMR (CD_3CN) δ 2.51 (s, 3 H), 4.38 (s, 3 H), 8.00 (d, J = 7 Hz, 1 H), 8.35 (d, J = 7 Hz, 2 H), 8.88 (s, 1 H); IR 2918, 1623, 1580, 1297 cm^{-1}. Anal. ($C_8H_9O_3NClF_3S$) C, H, N.

Compound **25c** (95%): mp 90–92 °C; NMR (CD_3CN) δ 2.55 (s, 3 H), 4.42 (s, 3 H), 8.40 (s, 1 H), 8.8 (s, 1 H); IR 2914, 1611, 1456, 1379, 1266, 1146, 1034 cm^{-1}. Anal. ($C_8H_8O_3NCl_2F_3S$) C, H, N.

Compound **25d** (98%): mp 111–113 °C; NMR (CD_3CN) δ 2.50 (s, 3 H), 4.46 (s, 3 H), 8.50 (s, 1 H), 8.95 (s, 1 H); IR 2893, 1612, 1462, 1379, 1260, 1156, 1032 cm^{-1}. Anal. ($C_8H_8O_3NClBrF_3S$) C, H, N.

1-Methyl-2-chloro-3-bromo-5-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]-pyridinium Iodide (26). 2-Chloro-3-bromo-5-methylpyridine was oxidized to 2-chloro-3-bromo-5-pyridinecarboxylic acid[22] (mp 175–178 °C). The acid was converted to the acid chloride [bp 140–143 °C, 18 Torr; NMR ($CDCl_3$) δ 8.55 (d, J = 1 Hz, 1 H), 9.00 (d, J = 1 Hz, 1 H); IR (neat) 2504, 1737, 1575, 1461 cm^{-1}]. Acylation of *l*-proline, as described for **3a**, gave *N*-[(2-chloro-3-bromopyridin-5-yl)carbonyl]pyrrolidine-2-carboxylic acid as a hydroscopic, pinkish oil [NMR ($CDCl_3$) δ 2.21 (br m, 4 H), 3.70 (m, 2 H), 4.45 (m, 1 H), 8.20 (d, J = 2 Hz, 1 H), 8.60 (d, J = 2 Hz, 1 H), 9.30 (s, 1 H, exchangeable with deuterium oxide); IR ($CHCl_3$) 3025, 1741, 1600, 1452, 1254, 1085 cm^{-1}].

Treatment of the α-amido acid as described for **6a** gave dimethyl 2,3-dihydro-5-(2-chloro-3-bromopyridin-5-yl)-1*H*-pyrrolizine-6,7-dicarboxylate (66%): mp 121–124 °C; NMR ($CDCl_3$) δ 2.55 (q, J = 6 Hz, 2 H), 3.15 (t, J = 6 Hz, 2 H), 3.78 (s, 3 H), 3.81 (s, 3 H), 4.02 (t, J = 6 Hz, 2 H), 8.02 (d, J = 2 Hz, 1 H), 8.38 (d, J = 2 Hz, 1 H); IR (Nujol) 2900, 1710, 1456, 1379, 1153 cm^{-1}. Anal. ($C_{16}H_{14}O_4N_2BrCl$) C, H, N.

Aluminum hydride–ether complex (1.2 g, 0.0217 mol) was suspended in anhydrous tetrahydrofuran (10 mL) at room temperature. A solution of the diester (2.0 g, 0.00483 mol) in ahydrous tetrahydrofuran (10 mL) was then added dropwise over a period of 30 min under an argon atmosphere. The mixture was stirred for 3 h at room temperature and then the reaction was carefully quenched with 5% sodium hydroxide (4 mL). Dichloromethane (30 mL) was added, and the inorganic salts were filtered and washed with hot dichloromethane (100 mL). The organic layers were combined, dried (sodium sulfate), and concentrated in vacuo to yield a tan solid, which was suspended in anhydrous dichloromethane (20 mL). Isopropyl isocyanate (1.23 g, 0.015 mol) and dibutyltin diacetate (2 drops) were added, and the reaction mixture was stirred under an argon atmosphere for 24 h. The mixture was then concentrated in vacuo to yield a colorless solid, which was subjected to flash chromatography (*n*-hexane–ethyl acetate, 1:1) to give 2,3-dihydro-5-(2-chloro-3-bromopyridin-5-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine bis(*N*-2-propylcarbamate) as colorless needles (2.1 g, 84% over two steps): 183–187 °C; NMR ($CDCl_3$) δ 1.17 (d, J = 6 Hz, 12 H), 2.51 (m, 2 H), 2.97 (t, J = 6.0 Hz, 2 H), 3.87 (m, 4 H), 4.55 (br d, J = 8 Hz, 2 H), 5.01 (d, s, 2 H), 5.03 (s, 2 H), 7.95 (d, J = 2 Hz, 1 H), 8.33 (d, J = 2 Hz, 1 H); IR (KBr) 3288, 2900, 1675, 1555, 1463, 1097 cm^{-1}.

Anal. ($C_{22}H_{28}O_4N_4ClBr$) C, H, N.

This bis-carbamate was methylated as described for **14a** to give 1-methyl-2-chloro-3-bromo-5-[2,3-dihydro-6,7-bis[[(*N*-2-propylcarbamoyl)oxy]methyl]-1*H*-pyrrolizin-5-yl]pyridinium trifluoromethanesulfonate (95%) as a hygroscopic yellow powder): mp ca. 97 °C (wet and bubbled) to 117 °C (complete dec); NMR ($CDCl_3$) δ 1.10 (d, J = 6 Hz, 12 H), 2.55 (m, 2 H), 2.92 (br t, J = 6 Hz, 2 H), 3.79 (m, 2 H), 4.12 (br t, J = 6 Hz, 2 H), 4.50 (s, 3 H), 5.25 (br s, 2 H), 8.60 (s, 1 H), 9.15 (s, 1 H); IR 3329, 1684, 1459, 1268 cm^{-1}. Anal. ($C_{24}H_{31}N_4O_7F_3ClBrS$) C, H, N.

The trifluoromethanesulfonate was converted to **26** as described for **16a**. Compound **26** (86%): mp 140 °C (wet and bubbled) to 152 °C (dec); NMR δ 1.05 (d, $J \approx 6$ Hz, 12 H), 2.50 (m, 2 H), 2.84 (br d, J = 6 Hz, 2 H), 3.60 (m, 2 H), 4.16 (br t, J = 6 Hz, 2 H), 4.45 (s, 3 H), 4.92 (s, 4 H), 6.93 (br t, J = 7 Hz, 2 H), 8.92 (d, J = 2 Hz, 1 H), 9.29 (s, 1 H); IR 2922, 1682, 1534, 1379, 1259 cm^{-1}. Anal. ($C_{23}H_{28}N_4O_4ClBr$) C, H, N.

Hydrolysis Rate Studies. A. Pyridinium Hydrolysis Rates. The hydrolysis rates of pyridinium prodrugs and model compounds were determined on a Cary 118 spectrophotometer by monitoring the decrease in pyridinium UV maximum ab-

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sorption as a function of time. Samples were kept at constant temperature in a thermostated circulating-water bath (accuracy ±0.1 °C).

A general procedure is as follows: a solution of the pyridinium compound in phosphate buffer of appropriate concentration and pH (adjusted via pH meter) was prepared so that the UV maximum absorption of the pyridinium chromophore remained on scale at the highest attenuation. The decrease in this absorption was then monitored as a function of time. Pseudo-first-order rate constants were obtained using the rate equation $\ln (Abs^0/Abs^t) = k_{obs}t$ (where Abs^0 = absorption of the pyridinium compound at time zero and Abs^t = absorption at time t). The slope of the linear plot of $\ln (Abs^0/Abs^t)$ vs time gave K_{obs}. Fast reactions were followed for a minimum of 3 half-lives and slow reactions for a minimum of 2 half-lives. All reactions displayed excellent first-order fit with correlation coefficients of 0.98 or better.

B. Bis-Carbamate Hydrolysis Rates. HPLC Studies. The hydrolysis of the bis-carbamate moieties was followed by HPLC using a Spectra-Physics 8000 system equipped with an Alltech C-8 reverse-phase column, a 30-μL autoinjector, a Varian Vari-Chrom UV detector, and a Hewlett-Packard 3392 integrator. Samples were kept at constant temperature in a jacketed column attached to a thermostated circulating-water bath (accuracy ±0.1 °C). The first-order rate constants were determined by following the relative decrease in the bis-carbamate peak area with time.

A general procedure is as follows: the appropriate buffer solution (2 mL) was added to a solution (2 mL, 0.5 mg/mL) of the bis-carbamate in acetonitrile. This mixture was loaded in the jacketed column reservoir and 30-μL samples were analyzed at time intervals such that a minimum of six injections were made for the faster reactions and about 12 were made for the slower reactions. All reactions were followed for a minimum of 3 half-lives. The C8 HPLC column was eluted with acetonitrile–water at a flow rate of 2 mL/min. Bis-carbamates **1**, **19a**, and **19b** had retention times of 5.0 min (65% acetonitrile in water), 4.1 min (42% acetonitrile in water), and 2.5 min (50% acetonitrile in water), respectively. Logarithmic values of peak area ratios ($area^0/area^t$) were plotted against time (min), and the slope of the line (determined by regression analysis) was the observed rate constant for the pseudo-first-order reaction. All reactions showed excellent first-order fit with correlation coefficient of 0.99 or better. The half-lives were not affected by the chromatographic conditions.

The buffers used in the pH–rate studies were sodium phosphate–HCl, pH 2.0; citric acid–NaOH, pH 3.0–3.5; succinic acid–NaOH, pH 4.0; sodium acetate–HCl, pH 5.0; sodium phosphate–HCl, pH 6.0–8.0; boric acid–NaOH, pH 9.0; and sodium carbonate–HCl, pH 10.0.

NMR Studies. Bis-carbamate hydrolysis rates were measured on a Varian EM390 NMR spectrometer at 90 MHz. Temperature was not controlled. A general procedure was as follows: a saturated solution of the bis-carbamate was prepared in a screw-cap vial by adding 50 mg to deuterium oxide (1.0 mL) containing 0.05 M deuterated phosphate buffer with the pD adjusted to 7.4. The vial was capped and the mixture placed in an ultrasonic bath and sonicated for 5 min. The solution was then filtered through glass wool into a NMR tube and capped. The decrease in the isopropyl carbamate methyl groups (δ 1.10) was followed as a function of time as determined by integration against an internal standard [3-(trimethylsilyl)propionic-2,2,3,3,-d_4 acid sodium salt]. Logarithmic values of peak area ratios ($area^0/area^t$) were plotted against time (min), and the slope of the line (determined by regression analysis) was the observed rate constant of the pseudo-first-order reaction. All reactions showed good first-order fit with a correlation coefficient of 0.96 or better.

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Registry No. **3a**, 123412-43-1; **3b**, 123412-44-2; **4a**, 123412-45-3; **4b**, 123412-46-4; **5a**, 123412-47-5; **5b**, 123412-48-6; **6a**, 123412-49-7; **6b**, 123412-50-0; **7a**, 123412-51-1; **7b**, 123412-52-2; **8a**, 123412-53-3; **9a**, 123412-54-4; **10a**, 123412-55-5; **11a**, 123412-56-6; **11b**, 123412-57-7; **12a**, 123412-58-8; **12b**, 123412-59-9; **13a**, 123412-60-2; **13b**, 123412-61-3; **14a**, 123412-63-5; **14b**, 123412-65-7; **15a**, 123412-67-9; **15b**, 123412-69-1; **16a**, 123412-70-4; **16b**, 123412-71-5; **17a**, 123412-72-6; **17b**, 123412-73-7; **18a**, 123412-74-8; **18b**, 123412-75-9; **19a**, 123412-76-0; **19b**, 123463-04-7; **21a**, 123412-77-1; **24a**, 123412-79-3; **24b**, 123412-81-7; **24c**, 123412-83-9; **24d**, 123412-85-1; **25a**, 123412-87-3; **25b**, 123412-89-5; **25c**, 123412-91-9; **25d**, 123412-93-1; **26**, 123412-94-2; *i*-PrNCO, 1795-48-8; 2-amino-4-methylpyridine, 695-34-1; 2-fluoro-4-methylpyridine, 461-87-0; 2-fluoropyridine-4-carboxylic acid, 402-65-3; 2-fluoropyridine-4-carbonyl chloride, 65352-95-6; pyridine-4-carboxylic acid *N*-oxide, 13602-12-5; 2-chloropyridine-4-carboxylic acid, 6313-54-8; 2-chloropyridine-4-carbonyl chloride, 65287-34-5; 2-amino-5-methylpyridine, 1603-41-4; 2-fluoro-5-methylpyridine, 2369-19-9; 2-fluoropyridine-5-carboxylic acid, 403-45-2; 2-fluoropyridine-5-carbonyl chloride, 65352-94-5; L-proline, 147-85-3; 2-chloropyridine-5-carbonyl chloride, 58757-38-3; dimethyl acetylenedicarboxylate, 762-42-5; dibutyltin diacetate, 1067-33-0; ethyl 2-fluoropyridine-4-carboxylate, 123412-95-3; ethyl 2-chloropyridine-4-carboxylate, 54453-93-9; ethyl 2-bromopyridine-4-carboxylate, 89978-52-9; ethyl 2,6-dichloropyridine-4-carboxylate, 1604-14-4; 2-fluoro-5-methylpyridine, 2369-19-9; 2-chloro-5-methylpyridine, 18368-64-4; 2,3-dichloro-5-methylpyridine, 59782-90-0; 3-bromo-2-chloro-5-methylpyridine, 17282-03-0; 3-bromo-2-chloro-5-pyridinecarboxylic acid, 29241-62-1; 3-bromo-2-chloro-5-pyridinecarbonyl chloride, 78686-84-7; *N*-[(2-chloro-3-bromopyridin-5-yl)carbonyl]-pyrrolidine-2-carboxylic acid, 123412-96-4; methyl 2,3-dihydro-5-(2-chloro-3-bromopyridin-5-yl)-1*H*-pyrrolizine-6,7-dicarboxylate, 123412-97-5; 2,3-dihydro-5-(2-chloro-3-bromopyridin-5-yl)-6,7-bis(hydroxymethyl)-1*H*-pyrrolizine bis(*N*-2-propylcarbamate), 123412-98-6; 1-methyl-2-chloro-3-bromo-5-[2,3-dihydro-6,7-bis(*N*-2-propylcarbamoyloxymethyl)-1*H*-pyrrolizin-5-yl]pyridinium trifluoromethanesulfonate, 123413-00-3.