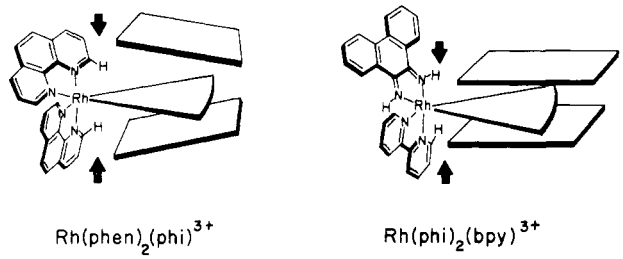


Figure 2. Matching the rhodium complex shapes to those of DNA sites. The schematic illustrates intercalation of $Rh(phen)_2(phi)^{3+}$ (left) and $Rh(phi)_2(bpy)^{3+}$ (right) into either an opened or canonical major groove site, respectively. For $Rh(phen)_2phi^{3+}$, the potential steric clashes between ancillary ligand hydrogen atoms and the base pair planes lead to the accommodation of the complex preferentially at such opened sites, whereas $Rh(phi)_2bpy^{3+}$, with recessed ancillary ligands, fits easily into the canonical intercalation site.

tercalated $Rh(phen)_2phi^{3+}$. The sequence selectivities found here do not appear to be dominated by such hydrogen bonding considerations, however, since it is the complex lacking hydrogen bonding groups in ancillary positions that shows the greater sequence selectivity.[11] Instead *the sequence selectivity observed must depend upon steric factors and a complementarity of the shape of the metal complex to the local conformation of the DNA site.* Figure 2 illustrates a model which rationalizes the different site selectivities observed. In B-DNA, at 5'-pyrimidine-purine-3' base steps, propeller twisting leads to steric clashes between the cross strand purine bases in the minor groove with a concomitant opening of the major groove.[12,13] The sequences cleaved preferentially by $Rh(phen)_2phi^{3+}$ are those which show the largest extent of such a major groove opening.[14,15] With the phi ligand inserted deeply between the base pairs, $Rh(phen)_2phi^{3+}$ appears to require sites with a more opened major groove; otherwise steric clashes may ensue between bases above and below the intercalation site and the overhanging phenanthroline H2 and H9 hydrogen atoms. For $Rh(phi)_2bpy^{3+}$, in contrast, the ancillary ligands do not overhang the metal center, and only the potentially hydrogen bonding imine protons abut the helical groove. Substantive intercalation by $Rh(phi)_2bpy^{3+}$ from the major groove, therefore, appears possible at all sites along the helix.

Rhodium(III) complexes of the phi ligand and its derivatives provide efficient photocleaving reagents, and they may find application both in vitro and in vivo. $Rh(phen)_2phi^{3+}$ becomes in particular a useful probe of the local variations in major groove size. Furthermore these results underscore the importance of considerations of shape in the design of sequence-specific molecules targeted to DNA.[16]

Acknowledgment. We are grateful to the NIH (GM33309 to J.K.B.; National Research Service Training Award for A.M.P., GM07216) and to the NSF for their financial support.

Supplementary Material Available: Synthesis and characterization data (NMR, FABMS, and UV-vis) for $[Rh(phen)_2phi]Cl_3$ and $[Rh(phi)_2bpy]Cl_3$ (2 pages). Ordering information is given on any current masthead page.

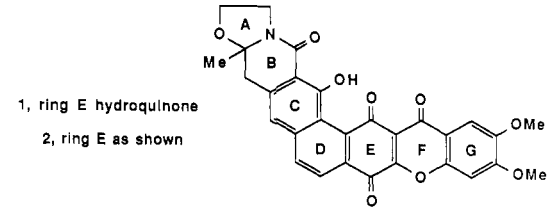
(11) The weak selectivity of $Rh(phi)_2bpy^{3+}$ for the 5'-TGC-3' site may be related to such hydrogen bonding interactions.
(12) Calladine, C. R. *J. Mol. Biol.* **1982**, *161*, 343.
(13) Dickerson, R. E. *J. Mol. Biol.* **1983**, *166*, 419.
(14) Nelson, H. C. M.; Finch, J. T.; Luisi, B. F.; Klug, A. *Nature* **1987**, *330*, 221. Satchwell, S. C.; Drew, H. R.; Travers, A. A. *J. Mol. Biol.* **1986**, *191*, 659.
(15) Intercalation may alter these conformational effects, although the stacking of phi between the base pairs could serve to reinforce the purine-purine stacking and roll.
(16) Recent crystal structures of protein-DNA complexes indicate that shape-selection may be an important feature also in the recognition of specific DNA sites by proteins. See, for example: Otwinowski, Z.; Schevitz, R. W.; Zhang, R.-G.; Lawson, C. L.; Joachimiak, A.; Marmorstein, R. Q.; Luisi, B. F.; Sigler, P. B. *Nature* **1988**, *335*, 321.

Synthesis of (±)-Cervinomycins A_1 and A_2

T. Ross Kelly,* Christopher T. Jagoe, and Qun Li

Department of Chemistry, Boston College
Chestnut Hill, Massachusetts 02167
Received February 13, 1989

The cervinomycins[1] are recently reported members of a small but growing family[2] of naturally occurring antibiotics, all of which possess xanthone- and isoquinolone-based units embedded within a larger polycyclic framework. To date, no synthesis of any member of this group has been recorded.[3] We now report the synthesis of cervinomycins A_1 (**1**) and A_2 (**2**).



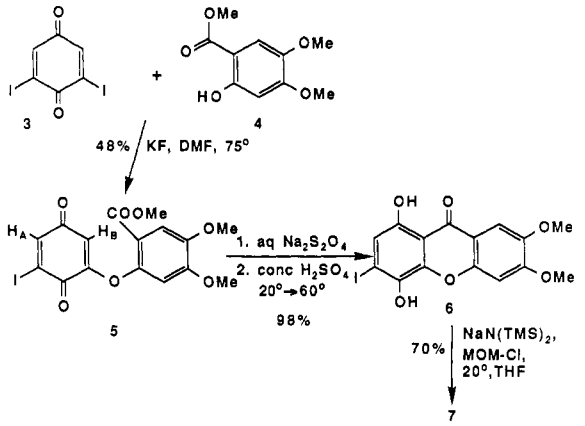
Considerations of synthetic economy dictated a convergent approach to the heptacyclic targets. A sequence based on the union of ABC and EFG fragments in the course of constructing the D ring appeared especially attractive. Preliminary studies indicated that the two extra carbons destined to become the phenanthrene bridge of the D ring could be effectively carried forward as an appendage to the ABC unit ($\rightarrow ABC_D$).

Construction of the EFG portion was straightforward[4] (Scheme I). Coupling of **3**[5] with **4**[6] proceeds via an addition/elimination mechanism to give **5**. That the reaction occurs with ipso and not cine[8] substitution of the iodine was established[4,9] by ^1H NMR (J_{AB} = 2.4 Hz). Reduction of **5** to the hydroquinone followed by cyclization[4] affords **6**. Findings later in the synthesis required

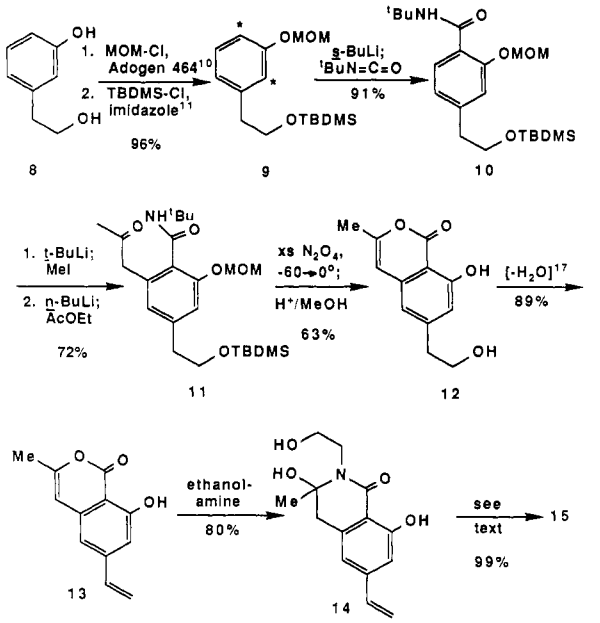
(1) Ōmura, S.; Nakagawa, A.; Kushida, K.; Lukacs, G. *J. Am. Chem. Soc.* **1986**, *108*, 6088-6089. Ōmura, S.; Nakagawa, A.; Kushida, K.; Shimizu, H.; Lukacs, G. *J. Antibiot.* **1987**, *40*, 301-308. For the original isolation, see: Ōmura, S.; Iwai, Y.; Hinotozawa, K.; Takahashi, Y.; Kato, J.; Nakagawa, A.; Hirano, A.; Shimizu, H.; Haneda, K. *J. Antibiot.* **1982**, *35*, 645-652. Qi, C.; Tian, J.; Marunaka, T.; Yamada, Y.; Yamawaki, I.; Ohtani, T.; Minami, Y.; Saito, H. Eur. Pat. Appl. EP 246,091, Nov 19, 1987; *Chem. Abstr.* **1988**, *108*, 130060d. For derivatives, see: Nakagawa, A.; Iwai, Y.; Shimizu, H.; Ōmura, S. *J. Antibiot.* **1986**, *39*, 1636-1638.
(2) Other members of the family include the following: (a) *Lysolipin I*, Dobler, M.; Keller-Schierlein, W. *Helv. Chim. Acta* **1977**, *60*, 178-185. Drautz, H.; Keller-Schierlein, W.; Zähner, H. *Arch. Microbiol.* **1975**, *106*, 175. (b) *Albofungin and chloroalbofungin*, for the initial structure assignments, see, inter alia: Gurevich, A. I.; Karapetyan, M. G.; Kolosov, M. N.; Omelchenko, V. N.; Onoprienko, V. V.; Petrenko, G. I.; Popravko, S. A. *Tetrahedron Lett.* **1972**, 1751-1754. Gurevich, A. I.; Deshko, T. N.; Kogan, G. A.; Kolosov, M. N.; Kudryashova, V. V.; Onoprienko, V. V. *Tetrahedron Lett.* **1974**, 2801-2804. A subsequent structure revision appears to have passed largely unnoted: Onoprienko, V. V.; Kozmin, Y. P.; Kolosov, M. N. *Biorg. Khim.* **1978**, *4*, 1418-1422; *Chem. Abstr.* **1979**, *90*, 54885u. (c) *LL-D42067α and LL-D42067β*, Labeda, D. P.; Kantor, S.; Kennett, R. L., Jr.; Carter, G. T.; Lee, T. M.; Borders, D. B.; Goodman, J. J.; Testa, R. T.; Maiese, W. M. U.S. Pat. Appl. 593,160, Mar 26, 1984; *Chem. Abstr.* **1986**, *104*, 62068c. (d) *Actinoplanones A-G*, Kobayashi, K.; Nishino, C.; Ohya, J.; Sato, S.; Mikawa, T.; Shiobara, Y.; Kodama, M. *J. Antibiot.* **1988**, *41*, 502-511, 741-750.
(3) (a) For a summary of model studies directed toward the synthesis of lysolipin I,[2a] see: Duthaler, R. O.; Heuberger, C.; Wegmann, U. H.-U.; Scherrer, V. *Chimia* **1985**, *39*, 174-182. For model studies relating to the synthesis of the cervinomycins, see: (b) Rama Rao, A. V.; Reddy, K. K.; Yadav, J. S.; Singh, A. K. *Tetrahedron Lett.* **1988**, *29*, 3991-3992. (c) Mehta, G.; Venkateswarlu, Y. *J. Chem. Soc., Chem. Commun.* **1988**, 1200-1202.
(4) Based on the method reported in Brassard, P.; Simoneau, B. *J. Chem. Soc., Perkin Trans. I* **1984**, 1507-1510.
(5) Commercially available from Lancaster Synthesis Ltd.
(6) Prepared in 73% overall yield from 2,4,5-trimethoxybenzoic acid (Aldrich) by selective[7] monodemethylation with BBr_3 in CH_2Cl_2 and esterification (CH_2N_2). For a lengthier synthesis, see: Hemmelmayr, F. V. *Monatsh. Chem.* **1914**, *35*, 1-8.
(7) Compare: Dean, F. M.; Goodchild, J.; Houghton, L. E.; Martin, J. A.; Morton, R. B.; Parton, B.; Price, A. W.; Somvichien, N. *Tetrahedron Lett.* **1966**, 4153-4159.
(8) For instances of cine substitution of haloquinones, see: Finley, K. T. In *The Chemistry of Quinonoid Compounds*; Patai, S., Ed.; Wiley Interscience: New York, 1974; p 895 and references thereto.

0002-7863/89/1511-4522$01.50/0

Scheme I



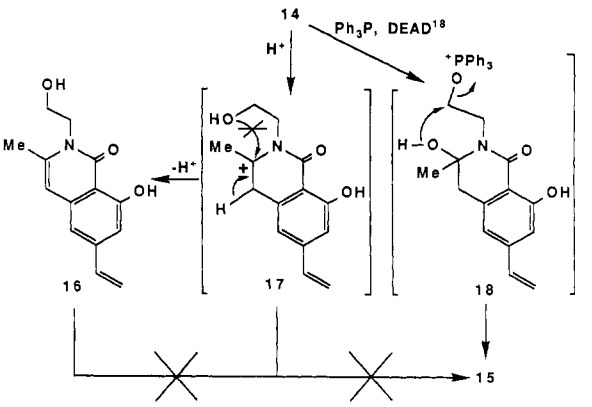
Scheme II



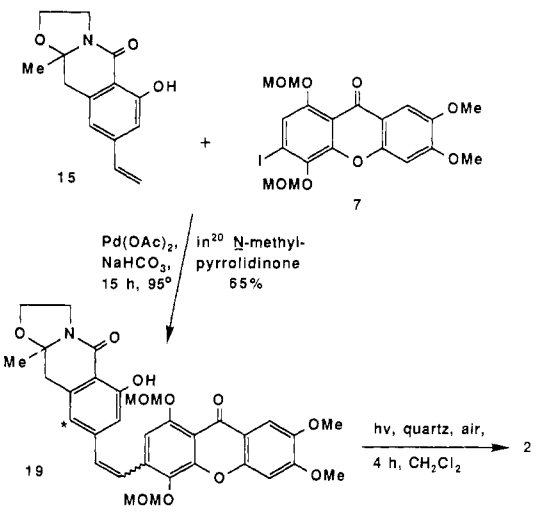
that the two OH's of **6** be protected; MOM groups (→ **7**) proved satisfactory.

The preparation of the ABC_D synthon is outlined in Scheme II. Proper choice[12] of experimental conditions allows one to direct ortho lithiation[13] to either asterisked position in molecules such as **9**. Under the conditions employed, reaction of the resulting anion with[14,12b] *tert*-butyl isocyanate gives amide **10** in 91% yield; none of the possible regioisomer was detected. The amide moiety in **10**, in addition to being the ultimate source of a C=O unit in **1** and **2**, also serves as the activating group for the two successive metalations[15] leading from **10** to **11**. Cleavage[16] of the three

Scheme III



Scheme IV



acid-labile groups in **11** occurs with simultaneous cyclization to give the isocoumarin **12**; dehydration[17] of **12** affords vinyl isocoumarin **13**.

Annelation of the oxazolidine ring onto **13** proved more difficult than expected. Reaction of **13** with ethanolamine proceeds smoothly to give **14**, but all efforts at acid-catalyzed cyclodehydration of **14** led to isoquinoline **16** (Scheme III). Attempts at cyclizing **16** to **15** failed. Mechanistic considerations eventually provided an avenue for redress. In particular, all the failed methods—which rely on conventional routes to oxazolidines—presumably share a common mechanism in which (see **17**) the tertiary hydroxyl functions as the leaving group and the primary hydroxyl acts as the nucleophile. Role reversal (**14** → **18** → **15**)[18] affords the solution.

With **15** and **7** in hand, elaboration to the cervinomycins proved refreshingly uncomplicated (Scheme IV). Pd^{II}-catalyzed arylation[19] of styrene **15** with **7** (but not with **6**) furnishes the corresponding stilbene. Irradiation[21] (medium pressure Hg lamp, quartz) of **19** in CH_2Cl_2 while open to the air precipitates a cascade of events which not only results in cyclization but also leads to cleavage of the MOM ethers and oxidation, providing (±)-cervinoymcin A_2 directly from **19** in a yield of 36%; none of the undesired regioisomer (resulting from cyclization at the asterisked carbon in **19**) was detected. The (±)-**2** so obtained is identical, except for properties dependent on optical activity, with an authentic sample of (−)-cervinomycin A_2 by direct comparison. Reduction of (±)-**2** with $NaBH_4$[22] gives (±)-cervinomycin A_1.[23]

(9) Kelly, T. R.; Echavarren, A.; Behforouz, M. *J. Org. Chem.* **1985**, *50*, 3849–3851.
(10) van Heerden, F. R.; van Zyle, J. J.; Rall, G. J. H.; Brandt, E. V.; Roux, D. G. *Tetrahedron Lett.* **1978**, 661–662.
(11) Corey, E. J.; Venkateswarlu, A. *J. Am. Chem. Soc.* **1972**, *92*, 6190–6191.
(12) (a) For a leading reference, see: Ronald, R. C.; Winkle, M. R. *Tetrahedron* **1983**, *39*, 2031–2042. See, also: (b) Townsend, C. A.; Bloom, L. M. *Tetrahedron Lett.* **1981**, *22*, 3923–3924. (c) Townsend, C. A.; Davis, S. G.; Christensen, B. S.; Link, J. C.; Lewis, C. P. *J. Am. Chem. Soc.* **1981**, *103*, 6884–6888.
(13) For a review, see: Gschwend, H. W.; Rodriguez, H. R. *Org. React. (N.Y.)* **1979**, *26*, 1–360.
(14) Wakefield, B. J. *The Chemistry of Organolithium Compounds*; Pergamon: Oxford, 1974; p 128.
(15) Compare: Kelly, T. R.; Bell, S. H.; Ohashi, N.; Armstrong-Chong, R. J. *J. Am. Chem. Soc.* **1988**, *110*, 6471–6480.
(16) For general references to the use of NO_2/N_2O_4 in amide "hydrolysis", see: White, E. H. *J. Am. Chem. Soc.* **1955**, *77*, 6008–6010, 6013–6022. For an application to N-^{t}Bu amides, see: Kelly, T. R.; Echavarren, A.; Chandrakumar, N. S.; Köksol, Y. *Tetrahedron Lett.* **1984**, *25*, 2127–2130.
(17) Grieco, P. A.; Gilman, S.; Nishizawa, M. *J. Org. Chem.* **1976**, *41*, 1485–1486.
(18) Carlock, J. T.; Mack, M. P. *Tetrahedron Lett.* **1978**, 5153–5156.
(19) For a review, see: Heck, R. F. *Org. React. (N.Y.)* **1982**, *27*, 345–390.
(20) Chalk, A. J.; Magennis, S. A. *J. Org. Chem.* **1976**, *41*, 1206–1209.
(21) Mallory, F. B.; Mallory, C. W. *Org. React. (N.Y.)* **1984**, *30*, 1–456.

Acknowledgment. Partial support of this work by Grant CA37054 from the National Institutes of Health is gratefully appreciated. We thank Dr. S. Ōmura for samples of **1** and **2** and helpful information and the NIH-supported (Grant DRR00480) mass spectrometry facility at Michigan State University for high resolution mass spectra.

(22) Augustine, R. L. *Reduction*; Marcel Dekker: New York, 1968; p 40 and references thereto.

(23) Mp's (°C) of crystalline solids: (±)-**2**, 279–284 °C dec [lit.[1] for (−)-**2**, dec >290 °C]; **6**, 260 °C dec; **7**, 172–173 °C; **13**, 82–84 °C; **15**, 81–82 °C; **16**, 115–116 °C. ^{1}H NMR spectra ($CDCl_3$) of key intermediates: **7**, δ 3.58 (3 H, s), 3.79 (3 H, s), 3.96 (3 H, s), 4.00 (3 H, s), 5.26 (2 H, s), 5.32 (2 H, s), 6.84 (1 H, s), 7.45 (1 H, s), 7.60 (1 H, s); **15**, δ 1.36 (3 H, s), 3.09 (2 H, br s), 3.66 (1 H, dt, J = 6.6, 11.3 Hz), 3.98 (1 H, m), 4.22 (2 H, t, J = 6.6 Hz), 5.37 (1 H, d, J = 10.9 Hz), 5.83 (1 H, d, J = 17.5 Hz), 6.63 (1 H, dd, J = 17.5, 10.9 Hz), 6.76 (1 H, s), 6.87 (1 H, s), 11.85 (1 H, s); *trans*-**19**, δ 1.40 (3 H, s), 3.14 (2 H, s), 3.61 (3 H, s), 3.66 (2 H, m), 3.71 (3 H, s), 3.97 (3 H, s), 4.01 (3 H, s), 4.10 (2 H, m), 4.25 (2 H, t, J = 7.5 Hz), 5.26 (2 H, s), 5.40 (2 H, s), 6.88 (1 H, s), 6.91 (1 H, s), 7.07 (1 H, s), 7.14 (1 H, d, J = 15.0 Hz), 7.26 (1 H, s), 7.62 (1 H, d, J = 15.0 Hz), 7.64 (1 H, s), 11.95 (1 H, s). The structures assigned to **7**, **15**, and **19** as well as those of other compounds are supported by combustion analysis or exact mass determinations.

Hydrolysis Kinetics of the Ultimate Hepatacarcinogen *N*-(Sulfonatooxy)-2-(acetylamino)fluorene: Detection of Long-Lived Hydrolysis Intermediates

Markandeswar Panda, Michael Novak,* and Jozef Magonski

Department of Chemistry, Miami University
Oxford, Ohio 45056

Received February 21, 1989

N-(Sulfonatooxy)-2-(acetylamino)fluorene (**1**) is a putative ultimate hepatacarcinogen derived from metabolism of 2-(acetylamino)fluorene.[1] We report herein preliminary results of an investigation of the hydrolysis kinetics of **1**[2] and the discovery of several labile intermediates which may play a role in the in vivo chemistry of **1**.

Kinetics were monitored by UV spectroscopy in 5 vol % CH_3CN–H_2O (μ = 0.5 M (KCl)) at pH 1.0–9.5 and 20 °C.[3] Absorbance data were fit well by eq 1 (n varied from 1 to 4, depending on pH). Buffer independent rate constants, k_i, and experimental details are collected in Table I in the Supplementary Material. One rate constant, k_2, is dependent on $[\text{phosphate}]_T$ and $[\text{tris}]_T$. Much of this dependence in phosphate buffers is due

$$A_t = A_\infty + \sum_{i=1}^{n} A_i e^{-k_i (B_T) t} \qquad (1)$$

to nucleophilic catalysis (see below), but general acid catalysis also occurs in both buffers. Figure 1 shows that five pseudo-first-order processes occur. The rate constant k_1 is pH and buffer independent as are rate constants for hydrolysis of the more reactive *N*-(sulfonatooxy)acetanilides.[3] A plot of log k_1 for **1** (extrapolated to 40 °C from data at 5–25 °C) and six ring-substituted *N*-(sulfonatooxy)acetanilides[3a] vs σ^+ gives a ρ of −5.7 ± 0.6 (r = 0.97), which is in the range expected for heterolysis of the N–O bond.[3,4] Three of the other processes are pH dependent

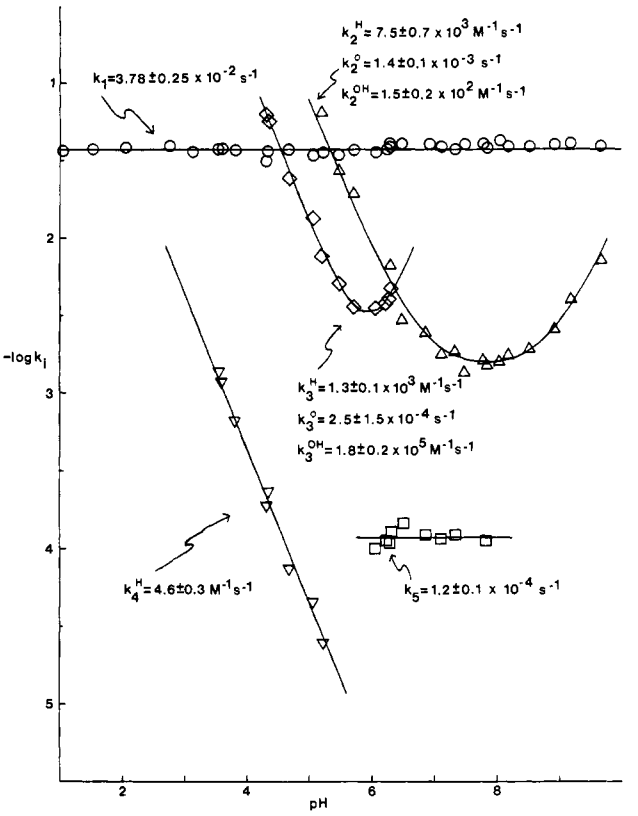
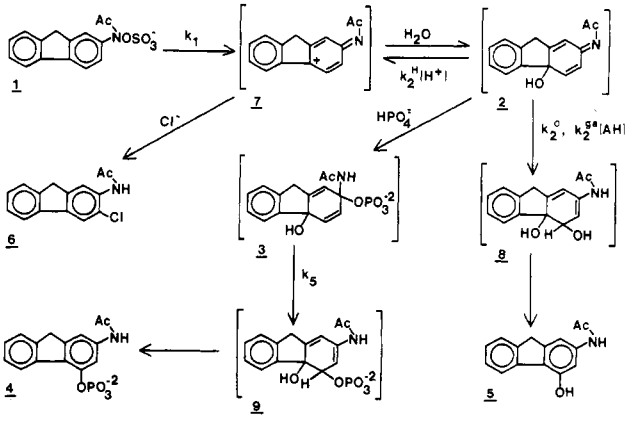


Figure 1. pH-rate profile for the hydrolysis of **1** at 20 °C in 5 vol% CH_3CN–H_2O (μ = 0.5 M (KCl)). The k_i were obtained by fits to eq 1. Rate constants shown in the figure were obtained by least-squares fits to appropriate rate equations.

Scheme I



(Figure 1). The rate constant k_5 is observed only in phosphate buffers, but its magnitude is independent of $[\text{phosphate}]_T$ and pH.

At pD 7.8 and 5 °C in 0.02–0.04 M KD_2PO_4/K_2DPO_4 (no KCl) **1** (ca. 2.8 mM) decomposes with a half-life of ca. 1 min into a longer lived species **2**, detected by 500 MHz ^{1}H NMR[5] (Scheme I), which also decomposes with a $[\text{phosphate}]_T$ dependent half-life of 3–7 min (consistent with k_2 at 5 °C) into **3**.[6] This species decomposes into **4**[7] with a half-life at 5 °C of 15–20 h

(1) DeBaun, J. R.; Miller, E. C.; Miller, J. A. *Cancer Res.* **1970**, *30*, 577–595. Weisburger, J. H.; Yamamoto, R. S.; Williams, G. M.; Grantham, P. H.; Matsushima, T.; Weisburger, E. K. *Cancer Res.* **1972**, *32*, 491–500.

(2) The synthesis of **1** (as its K^+ salt) has been described: Beland, F. A.; Miller, D. W.; Mitchum, R. K. *J. Chem. Soc., Chem. Commun.* **1983**, 30–31. Smith, B. A.; Springfield, J. R.; Gutman, H. R. *Carcinogenisis* **1986**, *7*, 405–411.

(3) (a) Novak, M.; Pelecanou, M.; Roy, A. K.; Andronico, A. F.; Plourde, F. M.; Olefirowicz, T. M.; Curtin, T. J. *J. Am. Chem. Soc.* **1984**, *106*, 5623–5631. (b) Novak, M.; Roy, A. K. *J. Org. Chem.* **1985**, *106*, 5623–5631.

(4) Gassman, P. G.; Campbell, G. A. *J. Am. Chem. Soc.* **1971**, *93*, 2567–2569. **1972**, *94*, 3891–3896. Gassman, P. G.; Granrud, J. E. *J. Am. Chem. Soc.* **1984**, *106*, 1498–1499.

(5) ^{1}H NMR for **2**: (500 MHz, D_2O) δ 2.32 (3 H, s), 3.64 (1 H, d, J = 18 Hz), 4.10 (1 H, d, J = 18 Hz), 6.1–6.35 obscured by **3**, 6.43 (1 H, d, J = 10 Hz), aromatic region obscured by **3**. The chemical shift of the acyl methyl group of **2** is consistent with that of other *N*-acylimines (ref 3b, 8, and 15).

(6) ^{1}H NMR for **3**: (500 MHz, D_2O) δ 2.07 (3 H, s), 3.28 (1 H, d, J = 17.5 Hz), 3.62 (1 H, d, J = 17.5 Hz), 6.10 (1 H, d, J = 10.1 Hz), 6.29 (1 H, d, J = 10.1 Hz), 6.35 (1 H, s), 7.41 (3 H, s, br), 7.58 (1 H, s); ^{31}P NMR (121.5 MHz, D_2O) δ 11.4 (relative to trimethyl phosphate). **3** rearranges into **4** and other unidentified materials upon attempted isolation. Only one diasteriomer appears to be present.

0002-7863/89/1511-4524$01.50/0