α-(Acyloxy)dialkylnitrosamines: Effects of Structure on the Formation of *N*-Nitrosiminium Ions and a Predicted Change in Mechanism

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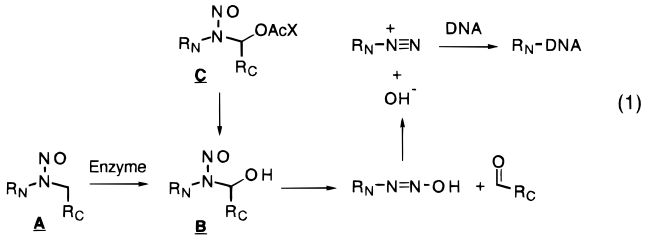
Abstract: The decay of α-(acyloxy)dialkylnitrosamines in aqueous solutions has been studied with a view toward elucidating mechanistic details and effects of structure on mechanism and reactivity. Rate constants (k_1) for the pH-independent decay of 43 α-(acyloxy)dialkylnitrosamines have been determined. Observations from these and other experiments rule out decomposition via an anchimeric assistance mechanism involving the *Z* isomer that had previously been suggested. All of the reported data for most of the compounds is consistent with a mechanism involving the formation of *N*-nitrosiminium ions in or before the rate-limiting step. Structure−reactivity correlations indicate that the stability of α-(acyloxy)dialkylnitrosamines is determined by electronic properties of substituents at R_N and R_C as well as by the ability of substituents R_C to engage in hyperconjugative interactions of C−H bonds with the developing cationic center in the transition state for nitrosiminium ion formation. Attachment of substituents of sufficient electron-withdrawing power at R_N and R_C results in a predicted change in mechanism to what appears to be an acyl group attack mechanism.

Introduction

Many dialkylnitrosamines (**A**) are enzymatically activated to their relatively unstable α-hydroxy derivatives (**B**), which give rise to DNA-alkylating diazonium ions and carbocations, as illustrated in eq 1.[1,2] α-(Acyloxy)dialkylnitrosamines (**C**) are



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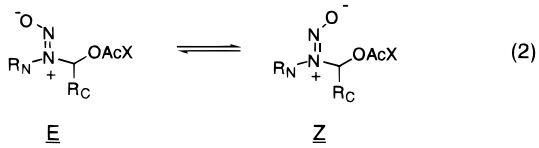
extensively used as precursors to α-hydroxydialkylnitrosamines in studies of nitrosamine carcinogenesis.[1,2] The esters vary widely in their reactivity, some being more unstable than the α-hydroxydialkylnitrosamine products.[3]

The effects of structure on the reactivity of the acyloxy compounds has been a source of confusion. The limited information available suggests that, for some substituents, the effects of attachment to nitrogen (R_N, eq 1) are opposite those attached to carbon (R_C, eq 1).[4,5] An early observation indicated that substituents that increase the relative proportion of the *Z* form of the ester, eq 2, increase reactivity.[5] This had been the



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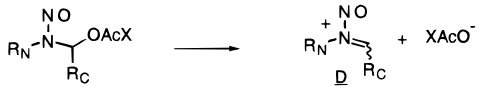
basis of the hypothesis that there was anchimeric assistance by the nitroso group of decomposition, possibly analogous to what had been observed in the case of β-(sulfatooxy)nitrosamines.[5] Recently, data, in the form of product analysis and structure activity effects, were summarized that indicated that nitroso oxygen attack at the carbonyl carbon was not a likely mechanism of reaction.[6] It was concluded that the dominant mechanism involved the formation of nitrosiminium ions (**D**) in, or prior to, the rate-limiting step, as in eq 3.[6−9] The onset of a carbonyl



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attack mechanism with electron-withdrawing substituents R_N and R_C (C, eq 1) was predicted.[6] An alternative mechanism or mechanisms of anchimeric assistance were not addressed, and the implications of the apparent correlation between isomeric composition and stability have not been identified. To clarify mechanistic aspects and the dominant structure−reactivity effects in this biologically important class of compounds, we have undertaken a more detailed study of the solvolysis of α-(acyloxy)dialkylnitrosamines.

In summary, correlations of electron-withdrawing power of substituents at R_N with reactivity and experiments to determine

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^{18}O-incorporation into products from solvent rule out the two conceivable mechanisms of anchimeric assistance. The stability of α-(acyloxy)dialkylnitrosamines is determined by electronic properties of substituents at R_N and R_C as well as by the ability of substituents R_C to engage in hyperconjugative interactions of C−H bonds with the developing cation in the transition state for formation of the nitrosiminium ion. The hyperconjugative effect dominates the reactivity effects of simple alkyl substituents and inverts the reactivity order expected on the basis of electronic considerations alone. Attachment of substituents of sufficient electron-withdrawing power at R_N and R_C results in the predicted change in mechanism to what appears to be an acyl group attack mechanism.

Experimental Section

Warning! Many α-(acyloxy)nitrosamines have been shown to be direct-acting carcinogens. Handling precautions entail the use of a well-ventilated fume hood and frequently changed double pairs of protective gloves. All materials suspected of contact are treated in 50% aqueous sulfuric acid baths containing the commercially available oxidant No-Chromix (Aldrich).

Reagents. Water that was used as solvent in the kinetic studies was redistilled from house-distilled water. Water for HPLC was redistilled from house-distilled water and filtered. Methanol and ethanol were dried and distilled from magnesium methoxide and ethoxide, respectively. Ethyl ether was dried and distilled from sodium. Acetonitrile, dichloromethane, chloroform, hexane, pentane, and ethyl acetate were dried and distilled from calcium hydride. Reagents for syntheses and kinetics were ACS grade or better, unless otherwise specified.

Syntheses. Imine precursors to the α-(acyloxy)nitrosamines were synthesized by the method of Graymore[10] and were typically purified before use by distillation. Generally, the crude yield was 70%. α-(Acyloxy)nitrosamines were synthesized from the corresponding imines as described previously.[6−9] The yields after final purification were 20−50%. Assignment of *E* and *Z* configurations was based on the method of Karabatsos.[11] In all but two cases, assignment based on the shift in α-protons was in agreement with that made by use of the shift in *β*-protons. In the two remaining cases, the shift in *β*-protons gave assignment in accord with those for other compounds with sterically similar substituents. The *E*/*Z* ratios reported refer to the solvent $CDCl_3$. For two compounds, **13** and **18**, the *E*/*Z* ratios in 60% D_2O/40% CD_3CN were also determined and did not differ considerably from the ratios determined in $CDCl_3$.

For compounds **1**, **4**, **5**, and **6**, syntheses and analytical data have been published previously.[6−9]

A typical procedure is given for (*N*-nitrosomethylamino)ethyl acetate (**7**). *N*-Methylethyleneamine, 1.0 g, was placed in a 50 mL round-bottom flask. A volume, ∼25 mL, of dry CH_2Cl_2 was added to the flask, followed by cooling in an ethanol/water/$(CO_2)_s$ bath (−10 °C). A solution of 10 mL of dry CH_2Cl_2, 1.7 g of dry Et_3N, and 1.1 g of acetic acid was added to the flask dropwise. Then 2.0 g of $NOBF_4$ was added slowly to the reaction mixture, which bubbled and turned yellow. When the bubbling ceased, the ice bath was removed, and the reaction mixture was stirred at room temperature for $^1/_2$ h. CH_2Cl_2 was evaporated under a gentle stream of argon. The remaining material was extracted with ether three times, and the combined ether solutions were evaporated under a stream of argon. The product was purified by preparative silica gel TLC using 1:4 ether/hexane as the eluting solvent. See the Supporting Information for characterization of the compounds synthesized.

Deuterated compounds **18**-d_1 and **4**-d_3, with the isotopes incorporated at the carbon atom *β* to the ester oxygen, were synthesized starting from the deuterated aldehydes. The final products were >97% deuterated by ^1H NMR.

Kinetics. The kinetics of decomposition of nitrosamines were observed using either a Milton Roy 1001+ or 3000 diode array spectrophotometer, a Hewlett-Packard 8452A diode array spectrophotometer, or an Applied Photophysics DX.17MV stopped-flow spectrophotometer. Temperature was maintained at 25 °C by means of attached recirculating water baths.

The buffer solution in each reaction cell was prepared by diluting a concentrated stock buffer solution. Ionic strengths of reaction solutions were typically maintained at 1 M with $NaClO_4$. The buffers that were used include phosphate, ethanolamine, hydrazine, MOPS, MES, CHES, acetic acid, cyanoacetic acid, methoxyacetic acid, cacodylic acid, and morpholine.

The stock solution of the nitrosamine was prepared in freshly distilled dry CH_3CN. Reactions performed in the stopped-flow spectrophotometer were carried out by mixing 25 parts of the aqueous buffer with 1 part of the nitrosamine in CH_3CN solution using 2.5 and 0.1 mL syringes, respectively. Final concentration of the nitrosamine in the reactions was $\sim 10^{-4}$ M.

The pH values were obtained after the kinetic run using an Orion model SA 720 pH meter equipped with a Corning combination electrode. Prior to recording reaction pH values, we performed two-point calibrations using commercially available standards or those prescribed by ref 12.[12]

Product Analyses. Products were quantitated by ^1H NMR. The reaction solutions were buffered with either 0.05 M cacodylic acid (50% anion) or 0.05 M chloroacetate (50% anion) and prepared in screw-cap vials, which contained either 0.80 mL of D_2O and 0.20 mL of CD_3CN or 0.90 mL of D_2O and 0.10 mL of CD_3CN. $NaClO_4$ was used to maintain the ionic strength of the reaction solutions. About 0.01 g of a substrate was weighed out and transferred into the vial, and the reaction was allowed to proceed for more than 10 half-times. The reaction mixture was transferred into a 5 mm NMR tube, and the ^1H NMR spectrum of the reaction mixture was recorded. The integration values for signals of corresponding alcohols, ammonium ions in the cases of (*N*-nitrosomethylamino)-*p*-chlorobenzyl and -*p*-(trifluoromethyl)benzyl chloroacetates, and acetaldehyde and methanol in the case of (*N*-nitroso(methoxyethyl)amino)ethyl acetate were used to calculate their percentages on the basis of integration of C−H proton signals from acetate or chloroacetate leaving groups.

^{18}O Incorporation. The extent of incorporation of ^{18}O from solvent water into benzaldehyde in the decomposition of **42** and **43** was investigated. Decomposition of **42** was carried out at 25 °C in 0.1 mL of 0.05 M 50% anion cacodylic acid buffer solution ($\mu = 1$ M ($NaClO_4$)) which contained ∼50% $H_2{}^{18}O$ and ∼50% $H_2{}^{16}O$. The reaction was allowed to proceed for 3 min (∼8 half-lives). In the case of **43**, the reaction was carried out at 25 °C in 0.02 M 20% anion cacodylic acid buffer solution ($\mu = 1$ M ($NaClO_4$)) containing ∼50% $H_2{}^{18}O$ and ∼50% $H_2{}^{16}O$ and was allowed to proceed for 20 min (∼6 half-lives). At the end of the reaction times, in both cases, the product benzaldehyde was extracted into ether and analyzed by GC-MS with an HP-1 (cross-linked methylsiloxane), 12 m × 0.2 mm, capillary column. The relative amounts of ^{18}O and ^{16}O in benzaldehydes were determined by monitoring the signals at masses 107, 108 and 105, 106 for the benzaldehydes, respectively. The relative intensities for a given sample were the mean values of a minimum of four analyses in both cases. Determination of the $^{16}O/^{18}O$ ratio of the solvent was accomplished by recombination of the ether layers with the original reaction solvents and evaporation of the ether under a stream of dry argon followed by reincubation for 24 h. The $^{16}O/^{18}O$ content of the aldehydes was subsequently redetermined by repeating the analysis method above. Control experiments with ^{16}O -benzaldehyde indicated that oxygen exchange was complete after 24 h under these conditions.

Results

Rate constants for the decay of α-(acyloxy)dialkylnitrosamines were determined from absorbance decay curves that exhibited good first-order behavior for 3−5 half-lives. Most of the determinations were in the pH range between 2 and 9 at 25

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Table 1. Kinetics Data for α-(Acyloxy)nitrosamines[a] at 25 °C in Aqueous Solution (Ionic Strength 1 M ($NaClO_4$))

compd	R_N	R_C	X	k_1[b], s^{-1}	% *Z* isomer[c]
1	*tert*-butyl	hydrogen	OAc	3.76(±0.21)e-5	4
2	methoxyethyl	hydrogen	OAc	4.08(±0.41)e-7	10
3	dimethoxyethyl	hydrogen	OAc	2.39(±0.53)e-7	11
4	*tert*-butyl	methyl	OAc	2.02e-3[c]	76
5	isopropyl	methyl	OAc	1.43e-3	23
6	ethyl	methyl	OAc	1.03e-3	10
7	methyl	methyl	OAc	6.86(±0.50)e-4	10
8	methoxymethyl	methyl	OAc	3.66(±0.10)e-4	0
9	benzyl	methyl	OAc	1.85(±0.12)e-4	5
10	dimethoxyethyl	methyl	OAc	1.72(±0.17)e-4	22
11	trifluoroethyl	methyl	OAc	2.31(±0.14)e-5	3
12	α-methylbenzyl	methyl	OAc	2.84(±0.02)e-4[d]	83
13	*tert*-butyl	ethyl	OAc	1.059(±0.04)e-3	72 (64)[e]
14	isopropyl	ethyl	OAc	6.41(±0.37)e-4	23
15	*n*-butyl	ethyl	OAc	4.75(±0.28)e-4	8
16	ethyl	ethyl	OAc	4.45e-4	15
17	methyl	ethyl	OAc	2.52(±0.28)e-4	0
18	*tert*-butyl	isopropyl	OAc	2.99(±0.12)e-4	70 (69)[e]
19	isopropyl	isopropyl	OAc	1.582(±0.07)e-4	29
20	ethyl	isopropyl	OAc	8.67(±0.39)e-5	0
21	methyl	isopropyl	OAc	6.39(±0.74)e-5	16
22	methoxyethyl	isopropyl	OAc	1.976(±0.12)e-5	4
23	ethyl	*tert*-butyl	OAc	1.29(±0.08)e-5	6
24	methyl	*tert*-butyl	OAc	6.65(±0.43)e-6	0
25	ethyl	$CH_2C(CH_3)_3$	OAc	1.24(±0.01)e-3	0
26	methyl	$CH_2C(CH_3)_3$	OAc	8.36e-4	0
27	ethyl	$PhCH_2CH_2$	OAc	2.03e-4	9
28	methyl	$PhCH_2CH_2$	OAc	1.0e-4	0
29	ethyl	$CH_3OCH_2CH_2$	OAc	1.34(±0.054)e-4	12
30	methyl	2-methoxyethyl	OAc	7.94(±0.88)e-5	0
31	methyl	2-methoxy-1-methylethyl	OAc	2.85(±0.33)e-5	0
32	ethyl	methoxymethyl	OAc	7.15(±0.93)e-6	19
33	ethyl	dichloromethyl	OAc	2.93(±0.31)e-6	17
34	$CH_3OCH_2CH_2$	H	OAcCl	8.55e-5	13
35	methyl	ethyl	OAcOMe	6.26(±0.39)e-3	0
36	methyl	isopropyl	OAcOMe	1.46(±0.09)e-3	<5
37	methyl	isopropyl	OAcCl	7.99(±0.11)e-3	7
38	methyl	*tert*-butyl	OAcCl	7.87e-4	0
39	*tert*-butyl	methyl	OcHex	7.92e-4	71
40	*tert*-butyl	*p*-chlorophenyl	OAc	1.31(±0.21)e-4	93
41	*tert*-butyl	*p*-chlorophenyl	O_2CPh	5.6e-4[d]	90
42	methyl	phenyl	OAcCl	4.6×10^{-2} [d]	4
43	*tert*-butyl	phenyl	OAcCl	3.30(±0.09)e-3	90

[a] See structure **C**, eq 1, for general formula. [b] The pH-independent rate constant for decay. The average value of the buffer-independent rate constant, k_0, for four or more determinations in the range of pH 2−9. [c] Determined in $CDCl_3$, except where noted. [d] Value from a single determination of k_0 at pH 6.1, cacodylic acid buffer. [e] Value in parentheses determined in 60% D_2O/40% DMSO-d_6.

°C and ionic strength 1 M. Rate constants were determined at a given buffer ratio typically at three buffer concentrations. In most cases, there was little effect on the magnitude of the rate constant k_{obsd}, over the range of concentration studied, 0.02−0.2 M. Increases in k_{obsd} of 20% or less were typical. In these cases, the buffers included cacodylic, chloroacetic, and methoxyacetic acids, CHES, and the ethylenediamine dication. Reactions in morpholine and bisphosphate buffers showed increases in k_{obsd} of up to 50% above the extrapolated buffer-independent value, k_0. Effects of increasing concentrations of acetate ions and acetate buffer solutions were variable and are discussed in more detail below. Values of k_0 were obtained from linear extrapolations to the *y*-intercept of plots of k_{obsd} against buffer concentration. Plots of log k_0 against pH were pH independent for most compounds over this pH range. The value of the rate constant k_1, for the pH-independent reaction, was taken as the average of all such experiments. For some compounds, a hydroxide ion dependent reaction was also observed in this pH range and the value of k_1 was taken from the best fit to the appropriate two-term rate law of eq 4. The

$$k_0 = k_1 + k_{OH}[OH^-] \quad (4)$$

values of k_{OH} will be reported elsewhere. In the case of the four compounds **4**, **12**, **41**, and **42**, the rate constants k_0 were measured at a single pH value of ∼6.1, which was in the pH-independent region in the case of all compounds for which the pH dependence was more completely characterized. The value of k_0 in these cases was taken as equal to k_1. All the values of k_1 are summarized in Table 1.

The secondary β-deuterium kinetic isotope effects for **18** and **4** were determined to be 1.02 ± 0.01 and 1.05 ± 0.02 per D, respectively.

Rate constants, k_H, for the acid-catalyzed decomposition of **7**, **17**, **21**, and **24** were measured below pH 2. The rate constants were determined as the slopes of plots of k_{obsd} against acid concentration. For **7**, **17**, **21**, and **24**, the values of k_H were 6.9 × 10^{-4}, 2.5 × 10^{-4}, 6.4 × 10^{-5}, and 6.7 × 10^{-6} M^{-1} s^{-1}, respectively.

Products of the pH-independent decay of the several (acyloxy)dialkylnitrosamines were characterized by ^1H NMR after decomposition of the compounds in D_2O solutions containing 10% or 20% CD_3CN (ionic strength 1 M, ($NaClO_4$)). The slightly higher concentration of acetonitrile in these studies compared to that in the kinetic determinations was necessitated

Table 2. Product Yields from the Decomposition of (Acyloxy)dialkylnitrosamines at 25 °C in Buffered D_2O with 10 or 20% CD_3CN[a]

compd[b]	% CD_3CN in reacn medium	% yield of R_NOD	% yield of $R_NND_3^+$	% yield of R_CCHO
7	10[c]	92	nd	99
8	20[d]	71[e]	nd	98
24	10[c]	92	nd	100
38	20[f]	99	nd	91
43	20[d]	89	10	99

[a] Yields determined by ^1H NMR and based on the intergration of the protons in the acetate anion leaving group. [b] See Table 1 for compound structure. [c] Buffered with 0.025 M $ClCH_2CO_2H$ buffer, 50% anion. [d] Buffered with 0.019 M $ClCH_2CO_2H$ buffer, 50% anion. [e] 28% CH_3OD, 27% CH_3CHO, and 15% $CH_3OCH_2CH_2OD$. [f] Buffered with 0.038 M cacodylic acid buffer, 50% anion.

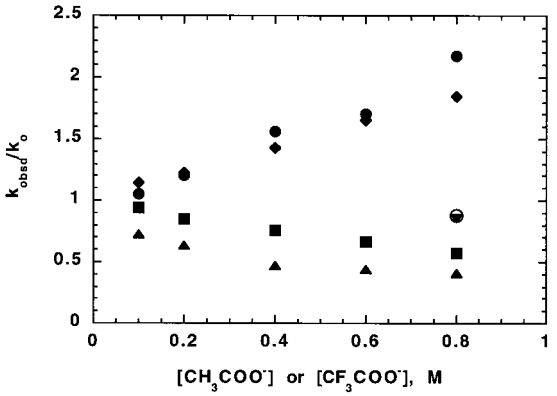


Figure 1. Plot of k_{obsd}/k_0 against acetate ion concentration for the decomposition of **7** (squares), **33** (solid circles), **1** (solid triangles), and **3** (diamonds) or against trifluoroacetate ion concentration for the decomposition of **1** (open circle) and **7** (inverted triangle) at 25 °C, ionic strength 1 M ($NaClO_4$).

by solubility considerations. Yields of the products were based on the C−H proton signal from the acetate or chloroacetate leaving groups. In cases where the yield of a product was low, identity was confirmed by spiking the NMR tube after quantitation with the authentic compound and observing the increase in signal in the appropriate frequency. The product yields are summarized in Table 2.

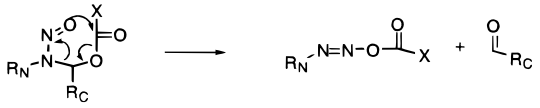
The effects of changing acetate ion or acetate buffer concentrations on the values of k_{obsd} were investigated in the case of four α-acetoxydialkylnitrosamines, and the results are depicted in Figure 1. Also included are data for control experiments in which the effect of changing salt from 0.1 M $Na^+CF_3CO_2^-$/0.9 M $NaClO_4$ to 0.8 M $Na^+CF_3CO_2^-$/0.2 M $NaClO_4$ upon k_{obsd} was determined for three α-acetoxydialkylnitrosamines.

Activation parameters were determined for the pH-independent solvolysis (k_1) for two compounds, **3** and **33**, by studying the effect of temperature on k_{obsd} over the temperature range from 20 to 50 °C. Linear Eyring plots of $\ln(k_{obsd}h/k_BT)$ against $1/T$(K) yielded values of $\Delta H^{\ddagger} = 14 \pm 1$ and 8 ± 0.2 kcal/mol and $\Delta S^{\ddagger} = -37 \pm 3$ and -56 ± 3 eu for **33** and **3**, respectively.

The results of experiments to measure the extent of ^{18}O incorporation into product benzaldehyde from the decomposition of **42** and **43** (natural abundance in ^{18}O) in ~50% ^{18}O-H_2O are summarized in Table 3. The compounds were decomposed for 6−8 half-lives, 3 and 20 min, for **42** and **43**, respectively. Control experiments with benzaldehyde (natural abundance in ^{18}O) incubated under the same conditions indicated the incorporation of ^{18}O from solvent into benzaldehyde to the extent of 0.8% and 6.6% after 3 and 20 min incubations, respectively. On the basis of the observation of 6.6% incorporation in 20 min, an upper limit of 2×10^{-4} s^{-1} can be calculated for the rate constant for exchange to equilibrium, 51.6% ^{18}O in this case, of the benzaldehyde oxygen with solvent water under these conditions.

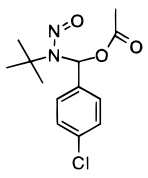
Discussion

Mechanism of the pH Independent Reaction. (A) No Anchimeric Assistance. An early study[5] on the reactivity of α-acetoxydialkylnitrosamines suggested a correlation between increasing reactivity and the percent of *Z*-isomer. It was concluded that the *Z* form decomposed with anchimeric assistance, though the nature of this assistance was not stipulated. It has been suggested[6] that there is nitroso oxygen attack at the acyl carbon to yield the ester products, formed via diazo ester intermediates, that are observed in the decomposition reactions of some α-(acyloxy)dialkylnitrosamines in nonpolar solvents. Such a process is indicated in eq 5. It was concluded[6] that such



(5)

a process was unlikely to be operative in aqueous solution due to the absence of a decrease in reactivity upon placement of carbonyl-stabilizing groups in the acyl function and the absence of anticipated products for the reaction in the presence of azide ion. The former point is buttressed in the comparison of reactivities of **40** and **41**. Both are >90% in the *Z* form, and



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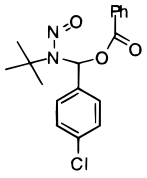
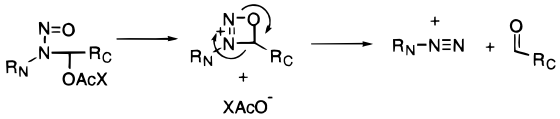


41

the latter is 4-fold more reactive—opposite what might be expected if carbonyl group attack is involved as in the mechanism of eq 5.[6]

An alternative possibility, involving nitroso oxygen attack on the α-carbon to yield initially a four-membered-ring intermediate that ultimately gives rise to products, as in eq 6, was



(6)

not addressed. Transition states and/or intermediates that are similar in structure to the four-membered ring in eq 6 are precedented by the nitro- and nitrosoamide decomposition[13,14] and by the nitrosative cleavage of imines to yield aldehydes.[15]

The observation in this study that the isotopic content of the oxygen in the product aldehyde is essentially identical to that of the solvent water (~50% ^{18}O)—and is very different from

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Table 3. Incorporation of ^{18}O from ^{18}O−Water in the Decay of ^{16}O-Containing α-(Acyloxy)dialkylnitrosamines at 25 °C, (Ionic Strength 1 M ($NaClO_4$))

compd[a]	%*Z* form [b]	%^{18}O-PhCHO after reaction	% ^{18}O in solvent[d]
42	4	50.6 (±0.3)	51.6 (±1.0)
43	90	48.4 (0.5)	46.4 (±1.2)

[a] Structure key in Table 1. Contained natural-abundance ^{18}O in all positions. [b] Determined in $CDCl_3$. [c] Reaction time was 3 min for compound **42** and 20 min for compound **43**. [d] Determined by equilibration of the product benzaldehyde with the reaction solvent for an additional 24 h.

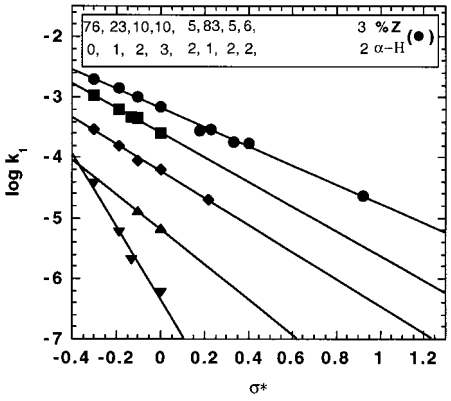


Figure 2. Plot of log k_1 against σ^* for substituents R_N (structure **C**, eq 1) in the solvolysis of α-acetoxydialkylnitrosamines at 25 °C, ionic strength 1 M ($NaClO_4$). R_C: $-CH_3$ (circles); $-CH_2CH_3$ (squares); $-CH(CH_3)_2$ (diamonds); $-C(CH_3)_3$ (triangles); $-H$ (inverted triangles—data from ref 6). Inset box applies only to circles: top row, percent of *Z* isomer measured by ^1H NMR; bottom row, number of α-hydrogens in R_N.

that in the starting α-(acyloxy)dialkylnitrosamine that contains only natural-abundance ^{18}O—rules out mechanisms of anchimeric assistance such as those in eqs 5 and 6. Table 3 indicates the isotopic contents of benzaldehyde formed in the decay of nitrosamines **42** and **43** (that contained only "natural-abundance" levels of ^{18}O) as well as the ^{18}O content of the water in which the reaction was carried out. The levels of ^{18}O in the product and solvent are the same within experimental error. If 10% of the reaction occurred by anchimeric assistance involving incorporation of oxygen from the starting material into the product aldehyde, the measured ^{18}O content of the benzaldehyde from **42** and **43** would be expected to be 46.4% and 41.6%, respectively. These values are different, outside 3 standard deviations from the observed values. Some ^{16}O-isotope retention in the product could be obscured due to oxygen exchange of the benzaldehyde with water, but allowance for this possibility still confirms that less than 10% of the product from **43**, and less still from **42,** can be due to anchimeric assistance pathways such as those in eqs 5 and 6.[16]

The observation, summarized in Figure 2, that α-acetoxydialkylnitrosamine reactivity toward solvolysis is a linear function of electron-donating ability of the substituent R_N, when R_C is held constant, and is independent of *E*/*Z* ratio is additional direct evidence against anchimeric assistance. In Figure 2, the solid circles are points for the log k_1 values of (*N*-nitrosoalkylamino)-ethyl acetates (R_C = methyl) plotted against σ^*. There is a good linear fit to the equation $\log k = -3.173 - 1.603\sigma^*$ ($R^2 = 0.995$).[17] The percent of *Z* isomer for each of these compounds is listed in the top row of the inset box of Figure 2, above the corresponding point for log k_1. It can be seen that the points for the values of log k_1 for compounds which are substantially or predominantly in the *Z* form do not noticeably deviate positively from the correlation, and they exhibit similar behavior for the correlation including only those compounds with 10% or less of the *Z* form. There is a possibility that the extent of *Z* conformer which we determined by ^1H NMR in $CDCl_3$ is different in the much more polar solvolysis medium (H_2O, ionic strength 1 M), thus confounding the analysis above. However, this seems unlikely because for two compounds, **13** and **18** (see Table 1), the *E*/*Z* ratios determined in 60% D_2O/40% CD_3CN were not substantially different from those determined in $CDCl_3$ (see Table 1). The absence of a rate enhancement due to increased *Z* conformation is a direct contradiction of the claim of anchimeric assistance, which, by definition, requires enhanced reactivity.

(B) Nitrosiminium Ion Formation. 1. Hyperconjugative Interactions of R_C and Electronic Effects at R_N. Figure 2 illustrates the dependence of log k_1, the rate constant for the pH-independent reaction, upon electron-withdrawing ability of substituents attached to the amino nitrogen (R_N). Inspection shows that there are separate correlations, for each distinct R_C group. The order of reactivity, for a given R_N group, is R_C = methyl > ethyl > isopropyl > *tert*-butyl > hydrogen. Among the first four groups, the rate constant is enhanced by increasing the number of hydrogen atoms β to the carbon atom undergoing bond cleavage. This is due the commensurate increase in the number of hyperconjugative interactions by C−H groups that can stabilize the developing nitrosiminium ion in the transition state.

There is evidence against the possibility that steric bulk, through inhibition of transition state solvation or some other means, is responsible for the observed order of reactivity:

First, steric bulk of the substituent attached to nitrogen (R_N) does not obscure the good linear correlation with σ^* in Figure 2 even though the correlation includes the bulky *tert*-butyl, isopropyl, and 1-phenylethyl groups. In a later section, it will be shown that electronic substituent effects at R_C are quantitatively similar to those observed for R_N substituents. This suggests a similar change in charge, at the nitrogen and carbon atoms of the developing iminium ion, in going from the ground state to the transition state. Given the similarity, it is difficult to see why steric hindrances of solvation should be so dramatically different at the two sites. In contrast, a difference in the involvement of hyperconjugative stabilization is indeed expected (vide infra).

Second, steric hindrance to solvation of the leaving carboxylic acid anion does not diminish reactivity beyond that expected simply from a consideration of leaving-group basicity effects. Both the (*N*-nitrosobutylamino)methyl benzoate and pivaloate are more reactive than the acetate, despite the lesser bulk of the acetate.[6] In addition, while the (*N*-nitroso-*tert*-butylamino)-ethyl acetate ester is more reactive, by less than a factor of 3, than the bulkier cylcohexanecarboxylic acid ester, most of this difference can be attributed to the greater basicity of the latter carboxylate leaving group. The value of log k_1 for this compound, adjusted for the pK_a difference, would only deviate negatively from the appropriate correlation for acetates (solid circles in Figure 2), by less than 0.2 log units.[18]

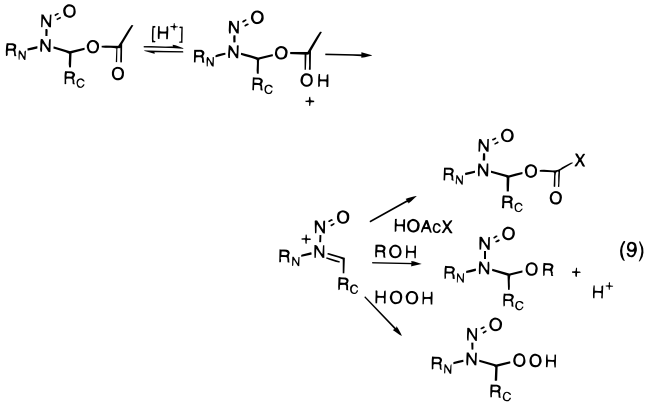
Third, steric hindrance to solvation of the anionic leaving group as a cause for the reactivity order is an untenable ex-

(16) A scheme and details on the method of calculation are included in the Supporting Information.

(17) A value of $\rho^* = -1.50$ was published on the basis of three substituents in ref 6.

(18) This is based on the reported difference in pK_a of 0.2 unit and the average value for β_{lg} of −1.09.

planation since the same order of reactivity ($k^{H}_{Me} > k^{H}_{Et} > k^{H}_{i-Pr} > k^{H}_{t-Bu}$) is observed in the acid-catalyzed decomposition reaction that has been shown to involve loss of neutral acetic acid with formation of the nitrosiminium ion. This mechanism, illustrated in eq 9, is indicated by the acid-catalyzed acyloxy

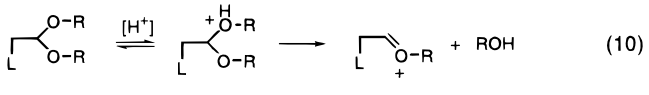


(9)

exchange and ether and hydroperoxide formation in alcoholic solvents.[19−21]

Fourth, two observations are inconsistent with the notion that steric bulk decreases reactivity by inhibition of solvation. Compound **25** is 3-fold more reactive than **16** despite the greater steric bulk at R_C in the former. The Taft steric parameter of the R_C substituent is −1.74 for **25** and only −0.07 for **16**. Similarly, despite the greater steric bulk of the $CH_2C(CH_3)_3$ ($E_s = 1.74$) compared to the *tert*-butyl substituent ($E_s = -1.54$),[22] the rate constant for nitrosiminium ion formation is 100-fold greater with the former substituent than with the latter in the case of the *N*-nitrosoethylamino compounds (**25** and **23**).

The normal secondary β-deuterium kinetic isotope effects are consistent with hyperconjugative stabilization of the developing nitrosiminium ion character in the transition state. The values of $k_H/k_D = 1.02 \pm 0.01$ and 1.05 ± 0.02 per D for the solvolysis of **18** and **4**, respectively, are in the direction expected for loosening of the β-C−L bonds due to overlap in the transition state with the adjacent electron-deficient center.[23] The magnitude of these effects is small compared to the largest effects reported of $k_H/k_D = 1.14$ per D. Small effects are expected in the case of a developing cation that is strongly stabilized by electron donation from the adjacent amino group.[23] Secondary β-deuterium kinetic isotope effects of magnitude similar to those observed here have been observed in the acid-catalyzed cleavage of acetals in which electron donation by an adjacent oxygen atom stabilizes the transition state for cleavage of the carbon oxygen bond as in eq 10.[24] The importance of C−H hypercon-



(10)

jugative stabilization of the transition state for this reaction has been emphasized by the work of Kreevoy and Taft.[25,26]

The observed order of reactivity, Me > Et > *i*-Pr > *t*-Bu, is the so-called "Baker−Nathan order",[27] which is widely observed in organic reactivity.[27,28] It was originally ascribed to a hyperconjugative interaction,[27] but this explanation has been rejected.[29−31] The notion that steric inhibition of solvation[29] could account for the original observation was given credence by the subsequent observation that the order was reversed in the gas-phase equilibrium for protonation of alkylbenzenes.[30] This observation indicates that the dominant equilibrium-stabilizing factor *in the gas phase* is the well-known polarizability effect. The observation led to the conclusion that the Baker−Nathan order in solution was indeed due to hindrance of solvation.[30] This conclusion has since been generalized.[31]

This conclusion is not logically required by the observation but rather is dependent on unverified assumptions. There is no reason to expect that the relative strengths of various interactions should be constant in going from the gas phase to polar liquids. Group polarizability is strongly attenuated in polar liquid media.[32] Other types of stabilizing interactions, such as hyperconjugation, may have different dependencies on the influences of solvent. Beyond this, it is currently unclear whether there can be differential expressions of hyperconjugative and polarizability interactions in transition states compared to ground states.

The experimental facts summarized above and elsewhere[25,26] require that the transitions state stabilization by an adjacent C−H bond is greater than that by an adjacent C−C bond. That this is possible has been disputed.[33] On theoretical grounds, with optimal structure, the C−C bond interaction with an adjacent carbocation is stronger.[33,34] Again, however, the possibility of differential expressions of β-H versus β-C hyperconjugative interactions in *transition states*, due to intrinsic constraints on heavy-atom motion or steric constraints, has not been addressed.

In the case of nitrosiminium ion formation from α-acetoxydialkylnitrosamines, the extent of enhancement of the rate constant by hyperconjugative stabilization is not a simple function of the number of β-hydrogens, in contrast to what has been reported in the case of acid-catalyzed acetal and ketal hydrolysis.[31] Thus, in Figure 2, at $\sigma^* = 0$, the increase in log k_1 as β-methyl groups are replaced by β-hydrogen atoms, going from *tert*-butyl to methyl, is 0.96, 0.64, and 0.41 unit for each successive replacement. These differences cannot be accounted for by differences in polar effects upon each successive replacement since each hydrogen-for-methyl replacement in-

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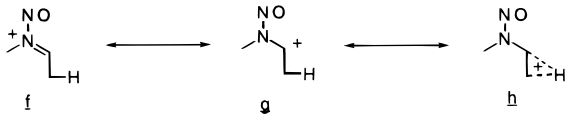
creases the value of σ^* of the R_C group by essentially the same amount, $\Delta\sigma^* \sim +0.1$. The differences may be the result of a saturation effect similar to that observed in classical resonance stabilization.[35]

The changes in slope in Figure 2 are consistent with either (1) a change in transition state structure or (2) a change in charge distribution in the transition state. The slopes of the lines, which indicate positive charge buildup in the transition state, increase in the order R_C = Me < Et < *i*-Pr < *t*-Bu with values of −1.60 (R^2 = 0.995), −2.05 (R^2 = 0.995), −2.23 (R^2 = 0.993), and −2.88, respectively. (1) The change in transition state structure would be consistent with a Hammond type effect[35] in which there is less carbon−oxygen bond cleavage as the reactivity of the starting material is enhanced by increasing hyperconjugative stabilization of the transition state in the order *t*-Bu < *i*-Pr < Et < Me. This is not ruled out, but the existing evidence does not suggest it. There is no indication of a significant change in the extent of charge buildup on the leaving acetate on the basis of the β_{lg} values, the slopes of plots of log k_1 against conjugate acid pK_a of the leaving acetate. The nearly constant slopes, = −1.05,[3] −1.13,[3] −1.10, and 1.09, of plots containing points for two or three different leaving groups for compounds in which the parent acetate is **6**, **17**, **21**, and **24**, respectively, do not suggest a changing transition state structure. The recently reported secondary α-deuterium kinetic isotope effects on the decay of methyl and propyl acetates average 1.15 ± 0.01 and 1.17 ± 0.02, respectively, for two compounds of each class,[8] again suggesting no difference. (2) The differences in slope can also be accounted for by a change in charge distribution in the transition state due to a change in the extent of hyperconjugative stabilization. Equation 11 illustrates three resonance forms that



(11)

can be considered to describe the structure of the nitrosiminium ion. Structure **h** represents that which describes hyperconjugative stabilization involving β-hydrogen atoms.

To the extent that structure **h** becomes more important in the order *t*-Bu < *i*-Pr < Et < Me, this results in diminution of the effective charge on nitrogen—structure **f** becomes a smaller contributor to the overall energy.

Finally, the *un*importance of hyperconjugative interactions for substituents attached to the amino nitrogen, R_N, is emphasized in Figure 2 for the ethyl acetates (filled circles). Given in the second row of the inset frame is the number of α-hydrogens (β to the nitrogen) for each R_N substituent, and it can be seen that the correlation is unperturbed as the number of such hydrogens varies from 0 to 3. This is expected because in no important resonance form of the nitrosiminium ion, eq 11, is the valence shell of the amino nitrogen less than completely occupied.

2. Substituent Effects at R_C. Because of the importance of hyperconjugation by hydrogen atoms on the R_C substituent, estimates of the polar effects of R_C substituents must be made by comparing substituents with identical numbers of α-hydrogen atoms. Figure 3 (solid points) contains such correlations. The slopes of plots of log k_1 against σ^* are −2.60 (R^2 = 0.974) and −2.82 (R^2 = 0.970) for *N*-ethylamino and *N*-methylamino compounds, respectively. Given the lower confidence in the correlations, it is uncertain whether these values of ρ^* represent

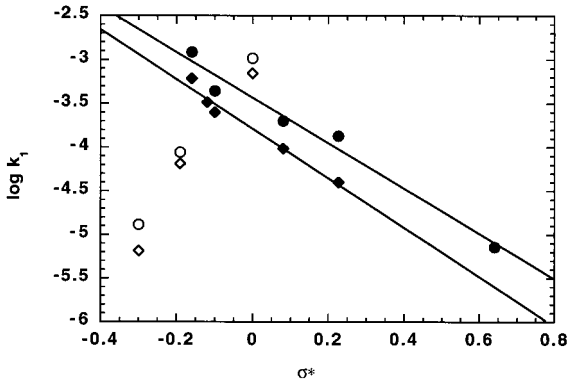


Figure 3. Plot of log k_1 against σ^* for substituents R_C (structure **C**, eq 1) in the solvolysis of α-acetoxydialkylnitrosamines at 25 °C, ionic strength 1 M ($NaClO_4$). For open and closed circles, $R_N = -CH_2CH_3$. For open and closed diamonds, $R_N = -CH_3$. For solid symbols, $R_C = -CH_2X$, X ≠ H. For open symbols, $R_C = -C(CH_3)_3$, $-CH(CH_3)_2$, and $-CH_3$.

real differences. The negative values of ρ^* are consistent with the formation of a transition state that is electron deficient relative to the ground state. The magnitudes indicate that the change in effective charge in going from the ground to transition state is similar to, in some cases slightly larger than, that experienced by substituents attached to the amino nitrogen for which the value of ρ^* varies from −1.6 to −2.9 (above). The relative *un*importance of steric bulk (and possible consequent hindrance of solvation) in determining the magnitude of rate constants is emphasized by the fact that, for the solid circles in Figure 3, proceeding from left to right, the value of the Taft steric constant changes as follows: −1.74, −0.07, −0.38, −0.77, and −0.19. This range of values is comparable with that traversed in passing through the series *t*-Bu, *i*-Pr, Et, Me but clearly does not obscure a reasonable correlation with σ^* alone. Rate constants for the Me, *i*-Pr, and *t*-Bu groups are plotted as open symbols and indicate substantially larger deviations than those observed for the solid points alone, indicative of the aforementioned hyperconjugative interaction.

(C) Change in Mechanism. The introduction of sufficiently strong electron-withdrawing groups at either R_N or R_C positions results in a change in mechanism, as previously predicted.[6] The expectation was based on the logic that such substituents would destabilize the transition state for nitrosiminium ion formation and simultaneously stabilize the transition state for nucleophilic attack at the carbonyl group of the ester. As noted (Results), the pH-independent reactions of **3** (R_N = dimethoxyethyl), **2** (R_N = methoxyethyl), and **33** (R_C = dichloromethyl) occur over relatively narrow regions compared to those of most of the α-acetoxydialkylnitrosamines. The rate constants for **3** and **2** lie 1.3 and 2.1 orders of magnitude above the line extrapolated in Figure 1 for other methyl acetates. The rate constant for **33** lies 3 log units above the line extrapolated for (*N*-nitrosoethylamino)alkyl acetates in Figure 3.[36] The positive deviations from the correlations for compounds that decompose through the formation of nitrosiminium ions are consistent with a change in mechanism.

The onset of a new mechanism is also indicated by the change in the effect on k_{obsd} of added acetate anion. In the case of reactions involving formation of *N*-nitrosiminium ions, the value

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(36) As the above discussion indicates, this does not take account of the fact that the $CHCl_2$ substituent has a single hydrogen atom that can stabilize the developing carbocation by hyperconjugation while those in Figure 3 have two; however, this means that the rate constant for **33** actually deviates by more than +3 log units from the appropriate correlation.

of k_{obsd} decreases with increasing acetate ion concentration due to diversion of the nitrosiminium ion intermediate to starting materials—the common ion inhibition.[9] Common ion inhibition is observed in Figure 1 for **1** and **7** (squares and triangles, respectively). The opposite effect of increasing acetate ion is observed in the case of **3** and **33** (diamonds and closed circles, respectively). Control experiments suggest that this effect is not likely due to a medium effect of replacing the $NaClO_4$ counterions with sodium acetate, which cancels a weak common ion effect. Replacement of 0.8 M $NaClO_4$ by 0.8 M sodium trifluoroacetate (open circle (**1**) and inverted triangle (**3**) in Figure 1) slightly diminishes the value of k_{obsd}—opposite the larger effect of sodium acetate upon decay of **1** and **7**. The enhancement of k_{obsd} with increasing sodium acetate concentration is consistent with rate-limiting attack of water on the carbonyl group that is assisted by general-base catalysis, as has been observed in related systems.[37]

The large negative values of entropies of activation observed for **3** and **33** are in contrast with what has been reported for reactions involving formation of nitrosiminium ions, but are consistent with what is known for ester hydrolysis involving water attack on the carbonyl group. Reported values of $\Delta S^{\ddagger}$ are near zero for the mechanism of eq 3. In contrast, $\Delta S^{\ddagger} = -37$ and -56 eu for **33** and **3**, respectively. These values are in the range reported for the mechanism of water attack on the carbonyl group.[38]

Summary. Experimental evidence has been adduced that rules out possible mechanisms of anchimeric assistance. The rate constants for formation of *N*-nitrosiminium ions are generally enhanced for electron-donating substituents in both R_N and R_C positions. In the case of simple alkyl groups in the R_C position, the number of hydrogens β to the cationic carbon of the developing nitrosiminium ion appears to dominate reactivity effects, increasing reactivity with increasing numbers of β-hydrogens due to hyperconjugative stabilization of the transition state. Attachment of substituents of sufficient electron-withdrawing power at R_N and R_C results in a predicted change in mechanism to what appears to be an acyl group attack mechanism.

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Supporting Information Available: Text giving spectroscopic and other analytical data for compounds not previously reported and a scheme and discussion of the analysis of calculated limits for ^{18}O incorporation experiments. This information is available free of charge via the Internet at http://pubs.acs.org.

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