Kinetics and mechanism of acid hydrolysis of 1-methyl-1-nitroso-3-*p*-tolylsulfonylguanidine and 1-methyl-1-nitroso-3-benzoylguanidine

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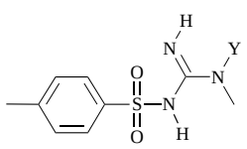
Kinetics of acid hydrolysis of 1-methyl-1-nitroso-3-*p*-tolylsulfonylguanidine 2 and of two 1-methyl-1-nitroso-3-benzoylguanidines (4-unsubstituted and 4-chloro) 5 and 6 have been studied. For the acid hydrolysis of 1-methyl-1-nitroso-3-*p*-tolylsulfonylguanidine 2, the absence of catalysis by thiocyanate ion, and the value of the kinetic solvent isotope effect indicate that either a rate determining proton transfer followed by fast denitrosation or a concerted pathway is involved in the mechanism. In the case of the acid hydrolysis of 1-methyl-1-nitroso-3-benzoylguanidines 5 and 6 it was observed that the protonated form decomposes *via* two parallel pathways. One involves a slow nucleophilic attack concerted with an intramolecular proton transfer, and the other a slow concerted denitrosation, where a second proton transfer and NO^+ expulsion are simultaneous.

Introduction

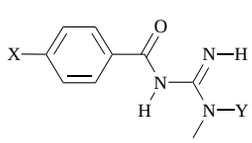
Over the last twenty years, reactivity of nitroso-compounds has been a topic of major interest, due to the potential cytotoxic nature of the resulting products.[1,2] The mechanism of acid hydrolysis of many nitroso-compounds has been reported;[3–13] nevertheless acid hydrolysis of polyfunctional nitroso-compounds and in particular guanidines has not been explored much.

Guanidines are usually considered as nitrogenated analogues of ureas; however, they are very basic compounds and, in this sense, they can be considered more similar to amines. In acidic media *N*-nitrosamines decompose to yield the parent amines and nitrous acid.[3–6] These reactions involve a fast protonation equilibrium on the nitrogen atom adjacent to the nitroso group followed by a rate limiting elimination of NO^+ catalysed by nucleophiles. *N*-Nitrosoamides and *N*-nitrosoureas decompose in acidic media by two parallel pathways: one involves denitrosation with a rate determining protonation followed by a rapid elimination of NO^+, and the other deamination that involves a fast protonation equilibrium followed by a rate determining deamination.[7–10] In previous work we have studied the kinetics of nitrosation of the antihypertensive drug clonidine,[14] a guanidine, that yields the mononitroso-derivative as a major product. We have also studied the kinetics of decomposition of *N*-nitrosoclonidine in acidic media. The reaction is acid catalysed and involves the rate limiting protonation of substrate as revealed by the solvent deuterium isotope effect of 1.2 and the independence of the denitrosation rate on the presence of nucleophiles.[13]

We then turned our attention to new combinations of the guanidine function with powerful electron withdrawing groups such as sulfonyl and carbonyl. We nitrosated compounds **1**, **3** and **4** obtaining the corresponding mononitroso-derivatives, 1-methyl-1-nitroso-3-*p*-tolylsulfonylguanidine (NTSG) **2**, 1-methyl-1-nitroso-3-benzoylguanidine (NBG) **5** and 1-methyl-1-nitroso-3-(4-chlorobenzoyl)guanidine (NClBG) **6** respectively. In this paper we report the kinetic study of the decomposition of **2**, **5** and **6**.



1 Y = H
2 Y = NO



3 Y = H, X = H
4 Y = H, X = Cl
5 Y = NO, X = H
6 Y = NO, X = Cl

Experimental

All chemicals and solvents used in the syntheses were of reagent grade. All reagents used for kinetic studies were of analytical grade or purified prior to use, and deionized water was used throughout this study.

All benzoylguanidines were prepared as described previously for the tolylsulfonylguanidine[15] and nitrosation was accomplished as already described.[13] Spectroscopic data for the new nitroso benzoylguanidines are as follows:

NTSG **2**: δ_H(300 MHz, $CDCl_3$) 2.44 (3H, s, CH_3 − Ar-), 3.18 [3H, s, $N(NO)CH_3$], 7.32 (2H, d, Ph) 7.87 (2H, d, Ph). *m*/*z* 256 (M^+), 226 (M^+ − NO). Mp 168–170 °C.

NBG **5**: δ_H(300 MHz, $CDCl_3$) 3.47 [3H, s, $N(NO)CH_3$], 7.24 (3H, m, Ph) 8.15 (2H, m, Ph). *m*/*z* 220 (M^+), 190 (M^+ − NO). Mp 99–105 °C.

NClBG **6**: δ_H(300 MHz, $CDCl_3$) 3.30 [3H, s, $N(NO)CH_3$], 7.50 (2H, d, Ph) 7.70 (2H, d, Ph). *m*/*z* 240 (M^+), 210 (M^+ − NO). Mp 105–107 °C.

The products of the acid hydrolysis of NTSG, NBG and NClBG were isolated by thin layer chromatography of the organic extract of the reaction medium, using dichloromethane as eluent.

Kinetic experiments were carried out in thermostatted cuvette cells, at 30 °C for NTSG, and 25 °C for NBG and NClBG. Kinetics runs were triggered by injecting a small ali-

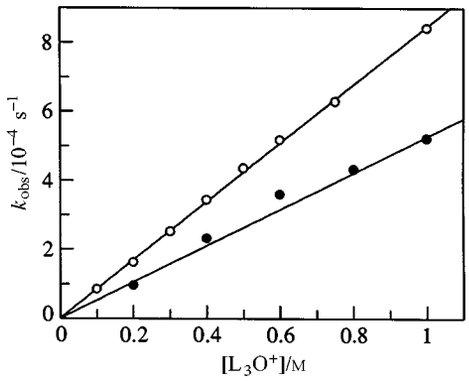


Fig. 1 Influence of [L_3O^+] (L = H or D) on k_{obs} for acid denitrosation of 1-methyl-1-nitroso-3-*p*-tolylsulfonylguanidine NTSG **2** (○, H; ●, D) (ionic strength = 1 M, T = 30 °C)

Table 1 Influence of [KSCN] on k_{obs} for acid denitrosation of 1-methyl-1-nitroso-3-*p*-tolylsulfonylguanidine **2** ([H^+] = 0.3 M; ionic strength = 1 M; T = 30 °C)

[KSCN]/M	$k_{obs}/10^{-4}$ s^{-1}
0.002	2.80
0.010	3.17
0.020	2.93
0.030	2.98

quot (30 μl) of a dioxane solution of the appropriate concentration of the nitroso substrate into the reaction medium. The ionic strength was kept constant at 1 M by addition of $NaClO_4$. Kinetic runs were monitored by UV spectroscopy following the decrease of the nitroso-compound absorbance (generally in the range of 200–300 nm) using a Milton Roy Spectronic 3000 Array or a Perkin-Elmer λ2 spectrophotometer. All experiments were carried out under pseudo-first-order conditions with large a deficit of nitroso-compound. Absorbance–time data always fitted the first-order integrated rate equation and the corresponding observed first-order rate constants, k_{obs}, were reproducible within 3% in all cases.

Results and discussion

1-Methyl-1-nitroso-3-*p*-tolylsulfonylguanidine

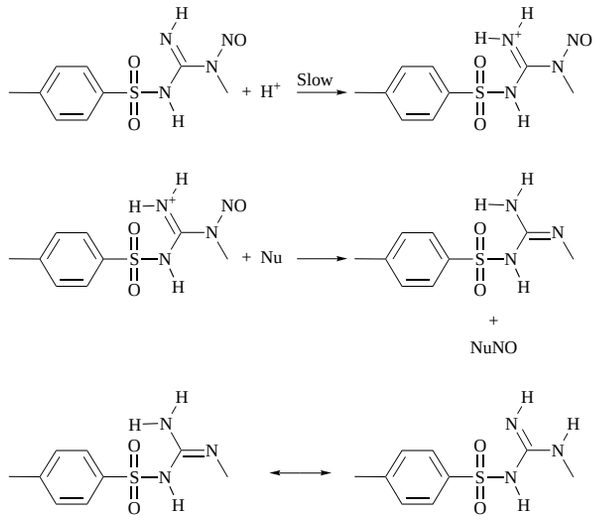
1-Methyl-1-nitroso-3-*p*-tolylsulfonylguanidine **4** is unstable in acid medium yielding the corresponding non-nitroso-derivative and nitrous acid. The reactions follow a first-order dependence on substrate concentration.

Fig. 1 shows that the values of the observed rate constants, k_{obs}, increase linearly with acid concentration [$HClO_4$]. From the slope a value for k_H^+ of (8.47 ± 0.05) × 10^{-4} dm^3 mol^{-1} s^{-1} [eqn. (1)] is obtained and the lack of intercept indicates that non-catalysed reaction is negligible.

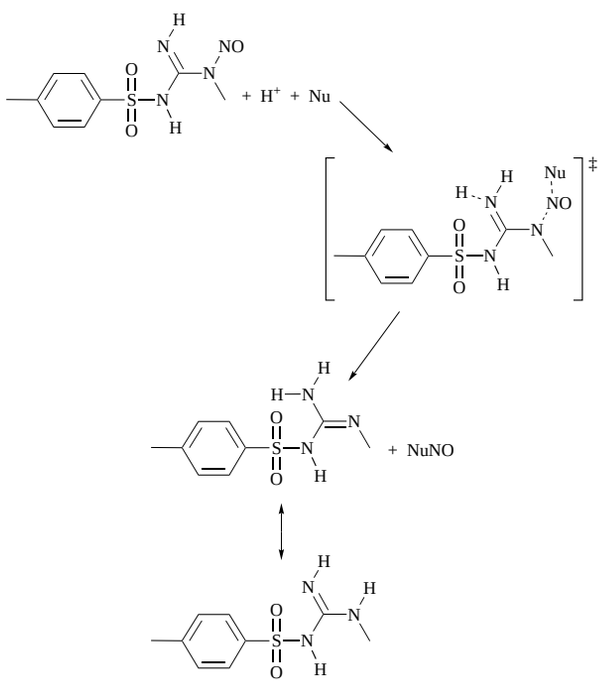
$$k_{obs} = k_H^+[HClO_4^+] \quad (1)$$

In Fig. 1 it is also shown that rate constants in D_2O are smaller than in H_2O and a value of 1.57 ± 0.04 for the solvent isotope effect for acid denitrosation, k_H^+/k_D^+, was obtained. No effect of nucleophile addition on the rate of hydrolysis of NTSG **2** (Table 1) was observed.

Both the solvent isotope effect and the absence of catalysis by nucleophiles point to a mechanism similar to that reported for the acid hydrolysis of the nitrosoamides[7,8] which involves a rate limiting protonation of substrate followed by fast expulsion of NO^+ with or without nucleophilic assistance from the protonated substrate (Scheme 1-A). Nevertheless a concerted protonation and NO^+ loss can also explain the experimental results (Scheme 1-B). Taking into account the microreversibility principle this mechanism is consistent with that proposed for the



Scheme 1-A



Scheme 1-B

nitrosation of 1-methyl-3-*p*-tolylsulfonylguanidine (TSG) **1**.[15] Since the nitrosation process showed a rate constant of 2.39 × 10^5 dm^3 mol^{-1} s^{-1} it is possible to compute an equilibrium constant for the nitrosation of TSG of K_{NO} = 2.82 × 10^8.

1-Methyl-1-nitroso-3-benzoylguanidines

The main products of acid hydrolysis of the 1-methyl-1-nitroso-3-benzoylguanidines **5** and **6** are the parent benzoylguanidines **3** and **4**, although 4-unsubstituted and 4-chloro benzoylureas are also detected as minor products due to hydrolysis of the benzoylguanidines. The amount of NO^+ liberated at the end of the reaction was measured as nitrite using a modification of Shinn's method.[16,13] In all cases a yield higher than 98% was detected. Acid hydrolysis of the 4-unsubstituted and 4-chloro benzoylnitrosoguanidine showed a first-order dependence on substrate concentration. At low H^+ concentrations the plot of k_{obs} *vs.* [$HClO_4$] showed a curved dependence while at lower pH values the observed first-order rate constants did not tend to a limit, but increased linearly with acid concentration (Figs. 2 and 4), implying complex dependence of k_{obs} on [H^+].

In a first approach this non-linear dependence could be explained by the existence of two simultaneous pathways for the acid decomposition of *N*-nitrosobenzoylguanidines: denitro-

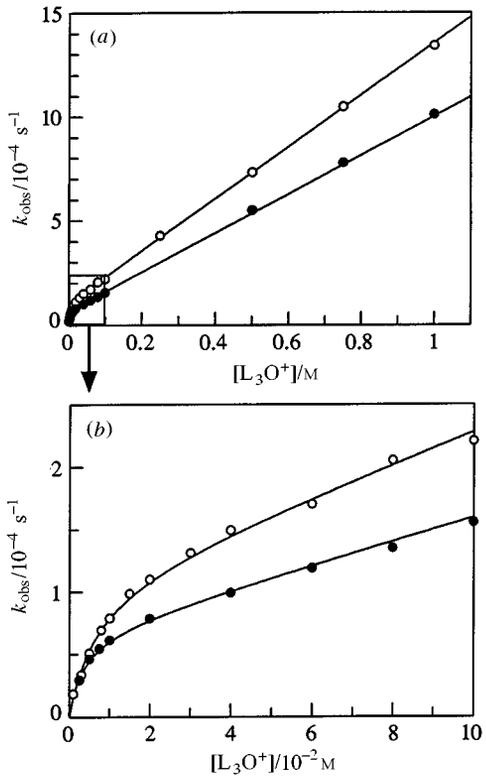


Fig. 2 (*a*) Influence of $[L_3O^+]$ on k_{obs} for acid denitrosation of 1-methyl-1-nitroso-3-benzoylguanidine NBG **5** (○, H; ●, D) (ionic strength = 1 M, T = 25 °C); (*b*) enlarged inset of (*a*)

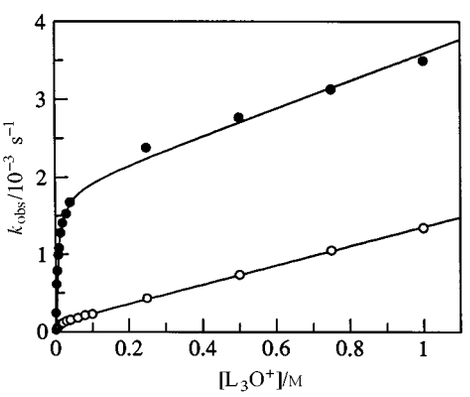


Fig. 3 Influence of $[H_3O^+]$ on k_{obs} for acid denitrosation of 1-methyl-1-nitroso-3-benzoylguanidine NBG **5** (○, without added nucleophile; ●, [KSCN] = 1×10^{-3} M) (ionic strength = 1 M, T = 25 °C)

sation and deamination in analogy to what was found for nitrosamides[7] and ureas.[9] However this possibility was ruled out because of the quantitative formation of nitrous acid (always >98%) observed.

A strong catalytic effect was observed when studying the dependence of k_{obs} on $[HClO_4]$ in the presence of a constant concentration of the powerful nucleophile SCN^- (Figs. 3 and 4). Nevertheless at high concentrations of acid no effect of SCN^- was observed and the plots of k_{obs} *vs.* $[HClO_4]$ are parallel. Again these results point to the existence of two parallel pathways in the mechanism of the acidic hydrolysis of *N*-nitrosobenzoylguanidines: one showing nucleophilic catalysis and the other showing no nucleophile effect.

All experimental results can be explained by a mechanism involving a protonation equilibrium of the carbonyl oxygen atom followed by two parallel pathways: one *via* an intramolecular proton transfer to the guanidine nitrogen with NO^+ elimination assisted by nucleophiles, and the other involving protonation at the guanidine moiety with subsequent or simultaneous denitrosation (Scheme 2).

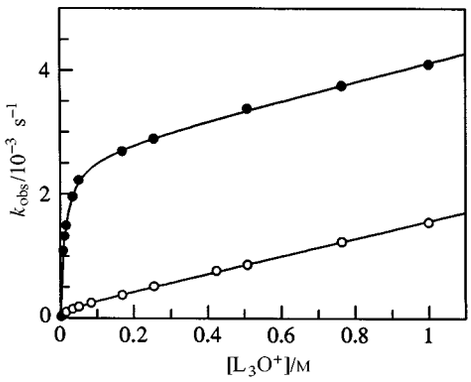
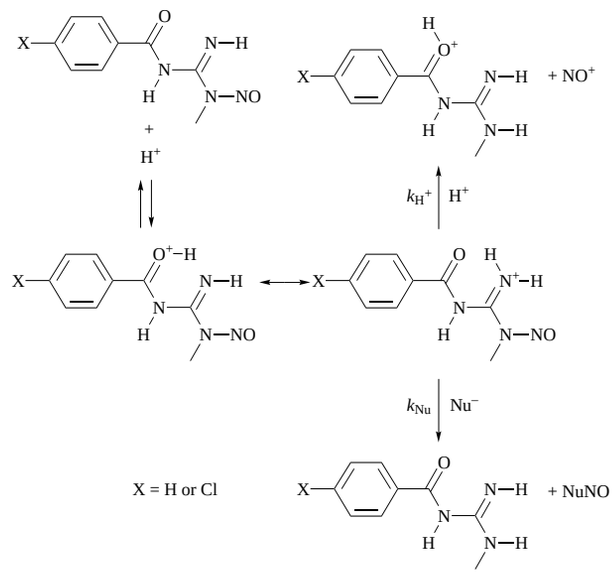


Fig. 4 Influence of $[H_3O^+]$ on k_{obs} for acid denitrosation of 1-methyl-1-nitroso-3-(4-chlorobenzoyl)guanidine NClBG **6** (○, without added nucleophile; ●, [KSCN] = 1×10^{-3} M) (ionic strength = 1 M, T = 25 °C)



Scheme 2

The site of this protonation is ambiguous. A tentative *O*-protonation followed by a rearrangement to the thermodynamically more stable *N*-protonated compound is proposed in analogy with that reported for the case of amides and ureas.[17] From Scheme 2 eqns. (2) and (3) can be easily deduced.

$$K_p = \frac{[NBGH^+]}{[NBG][H^+]} \quad (2)$$

$$k_{obs} = \frac{K_p k_{H^+}[H^+]^2 + K_p k_{Nu}[Nu][H^+]}{K_p[H^+] + 1} \quad (3)$$

Experimental data fitted satisfactorily eqn. (3) as can be observed in Figs. 2, 3 and 4 where solid lines represent calculated k_{obs} according to eqn. (3). In Table 2 the values obtained for K_p, the protonation constant, k_{H^+} and k_{Nu} are summarized.

As expected, protonation of 4-chloro-substituted nitrosobenzoylguanidine **6** on the oxygen atom is more difficult than that of the corresponding non-substituted compound **5**, K_p(Cl)/K_p(H) = 0.3. The values of k_{H^+} for both nitrosobenzoylguanidines are about the same, which agrees with the proposal of a rate-determining step involving a second protonation far from the substituted aromatic ring. The effect of thiocyanate ion as nucleophile was found to be stronger (by a factor of 10^6) in comparison to water. Thiocyanate ion is known to be one of the most effective catalysts of denitrosation reactions.[3]

Kinetic solvent isotopic effect

In order to further confirm the mechanism, we have carried out

Table 2 Kinetic parameters obtained from the fit of k_{obs} for acid denitrosation of 1-methyl-1-nitroso-3-benzoylguanidine (NBG) and 1-methyl-1-nitroso-3-(4-chlorobenzoyl)guanidine (NClBG) to eqn. (3) (ionic strength = 1 M; T = 25 °C)

	$K_p/10^2$ dm^3 mol^{-1}	$k_H^+/10^{-3}$ dm^3 mol^{-1} s^{-1}	k_{Nu}[Nu]/10^{-4} dm^3 mol^{-1} s^{-1}	k_{Nu}/dm^3 mol^{-1} s^{-1}
NBG(H_2O)	1.49 ± 0.16	1.3 ± 0.3	1.2 ± 0.2	(2.2 ± 0.3) × 10^{-6} [a]
NBG(D_2O)	2.79 ± 0.22	0.9 ± 0.2	0.7 ± 0.1	(1.2 ± 0.2) × 10^{-6} [a]
NBG, [KSCN] = 10^{-3} M	1.45 ± 0.07	1.75 ± 0.25	18.6 ± 1.6	1.86 ± 0.16
NClBG	0.443 ± 0.032	1.4 ± 0.3	1.9 ± 0.2	(3.4 ± 0.4) × 10^{-6} [a]
NClBG, [KSCN] = 10^{-3} M	0.784 ± 0.028	1.53 ± 0.17	26 ± 2	2.6 ± 0.2

[a] [H_2O] = 55.55 M.

kinetic runs of the acid denitrosation of the NBG **5** in deuterated water, the results are shown in Fig. 2 and summarized in Table 2.

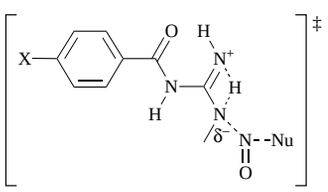
The protonation equilibrium shows an isotopic effect of $K_p(H)/K_p(D) = 0.53 \pm 0.11$ which corresponds to a solvent isotopic effect of 1.9 ± 0.3 for the dissociation of protonated NBG (K_{AH}^+).

The kinetic solvent isotope effect (KSIE) corresponding to the pathway catalysed by nucleophiles is $k_{Nu}(H)/k_{Nu}(D) = 1.7 \pm 0.5$ which indicates a proton transfer being involved in the rate determining step. A proton transfer preceding the nucleophilic attack on the nitroso group, as occurs in *N*-nitrosamines denitrosation,[7] must be ruled out because in that case a KSIE smaller than 1 should be expected. So proton transfer must be concerted although not necessarily synchronous with the nucleophilic displacement of the nitroso group.

The KSIE observed for the denitrosation pathway of the protonated substrate, $k_H^+/k_D^+ = 1.4 \pm 0.5$, is consistent with a slow protonation on the nitrogen atom adjacent to the nitroso group. Williams reported a similar KSIE for the acid denitrosation of *N*-methyl-*N*-nitrosotoluene-4-sulfonamide.[11]

Substituent effect

It is possible to discuss the values of the different kinetic parameters shown in Table 2 in terms of the substituent effects on the protonation equilibrium, K_p, acidic, k_H^+, and nucleophilic, k_{Nu}, denitrosation rate constants of the 1-methyl-1-nitroso-3-benzoylguanidines. As expected the protonation constant K_p decreases with electron withdrawing while the rate constant for the nucleophilic denitrosation, k_{Nu}, increases from $(1.2 \pm 0.2) \times 10^{-4}$ to $(1.9 \pm 0.2) \times 10^{-4}$ dm^3 mol^{-1} s^{-1}. These results suggest a significant development of negative charge in the transition state **7**, as consequence of N · · · N=O bond breaking.



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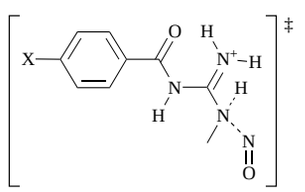
Quite surprising is the lack of any significant effect of electron withdrawing groups on the rate constant for acid denitrosation, k_H^+. This behaviour should be related to the balance between two simultaneous antagonistic effects. On the one hand the protonation of the nitrogen atom adjacent to the nitroso group, which would be favoured by the Cl group, causes the expected reactivity sequence to be NBG > NClBG. On the other hand, if N · · · N=O bond breaking is simultaneous with protonation, the development of a negative charge on the nitrogen atom adjacent to the nitroso group would be stabilized by electron withdrawing substituents, reversing the reactivity sequence to NClBG > NBG. These two factors can then account for the insensitivity of k_H^+ on substituents leading us to propose that acid denitrosation takes place *via* a nonsymmetrical concerted transition state **8** similar to that found for the acid hydrolysis of alkyl nitrites[18] and of *N*-methyl-*N*-nitrosobenzenesulfonamides.[19]



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The different behaviour towards acid hydrolysis shown by the sulfonyl and carbonyl guanidines could be attributed to the different Lewis basicity of the sulfonyl and carbonyl moiety. In carbonylguanidines the delocalization of the nitrogen lone pair of electrons increases electron density on the carbonyl oxygen atom, while in the case of the sulfonylguanidine this resonance is clearly lower.[20]

Acknowledgements

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