Oxyhalogen−Sulfur Chemistry: Kinetics and Mechanism of the Oxidation of Thionicotinamide by Peracetic Acid

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The kinetics and mechanism of oxidation of an important xenobiotic, thionicotinamide (TNA), using peracetic acid in slightly acidic media were studied by spectrophotometric techniques. The reaction is characterized by a very rapid initial oxidation of the sulfur atom of the thioamide group to the *S*-oxide, followed by a much slower decomposition of the *S*-oxide to form additional oxidation products, mainly the thionicotinamide sulfinic acid. In excess thionicotinamide, the stoichiometry of the reaction was determined to be $CH_3CO_3H + (C_5H_5N)C(=S)NH_2 \rightarrow (C_5H_5N)C(=NH)SOH + CH_3COOH$, whereas in excess peracetic acid the stoichiometry was 2:1, $2CH_3CO_3H + (C_5H_5N)C(=S)NH_2 + H_2O \rightarrow (C_5H_5N)C(=NH)SO_2H + 2CH_3COOH + 2H^+$. The sulfoxide is quite stable, but there was no experimental evidence for the existence of any stable sulfone−sulfonic acid intermediates. Results show that the sulfur atom in thionicotinamide is the reactive center, undergoing a stepwise addition of oxygen to form successively the sulfenic acid and the sulfinic acid. There appeared to be no further oxidation past the sulfinic acid and no formation of sulfate was observed. A bimolecular rate constant of $(1.1 \pm 0.3) \times 10^3$ M^{-1} s^{-1} was evaluated for initial rapid formation of the *S*-oxide, and an upper limit rate constant of 0.10 ± 0.02 M^{-1} s^{-1} was evaluated for the slower decomposition of the *S*-oxide.

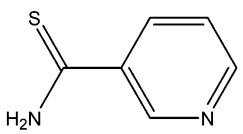
Introduction

The thioamide functional group is present in many drugs, chemicals, and agricultural chemicals, and exposure to man can result in inadvertent toxicity. Thus, a large number of potentially useful drugs containing the thioamide group are limited in their use due to the toxic side effects associated with these compounds. The thioamide, Prefex (2,6-dichlorothiobenzamide), has been used as herbicide;[1] quazepam (7-chloro-1-(2,2,2,-trifluoroethyl)-5-(*o*-fluorophenyl)-1,3-dihydro-2*H*-1,4-benzodiazepin-2-thione), has clinical efficacy as a hypnotic;[2] ethionamide is used as an antitubercular agent;[3] and thiobenzamide is a hepatotoxin that possesses powerful antibacterial properties.[4]

The associated toxicity of the thioamides has been linked to their oxidative metabolism, especially S-oxygenation.[5] Recent experimental evidence suggests that thioamides are easily oxidized to sulfoxides (sulfines), which under more vigorous oxidation conditions, are further oxidized to iminosulfinic acids (sulfenes). The toxicity, and possibly carcinogenicity of thioamides is caused by their metabolic oxidation to sulfines and sulfenes. The simple thioamide, thioacetamide (a carcinogen, tetratogen, and hepatotoxin), is oxidized to thioamide *S*-oxide and then to the *S*,*S*-dioxide, which is a highly reactive acylating intermediate with sufficient chemical reactivity to bind covalently to cellular macromolecules.[6]

Very few studies have addressed the mechanism and molecular basis for S-oxygenation, although it seems to be a prerequisite for bioactivation of most organosulfur compounds.[7,8] Although S-oxygenations of organosulfur xenobiotics by microsomes supplemented with NADPH and oxygen have been extensively studied,[9] the easy oxidation of sulfur compounds by hydrogen peroxide, oxyhalogens and other species of reduced oxygen such as the superoxide ion have not yet been systematically studied. It is generally accepted that metabolism of chemically stable organosulfur compounds depends on enzyme catalysis, with the microsomal cytochrome P-450 system and the flavin-containing monooxygenases, both, often implicated in S-oxygenations catalyzed by microsomes.[10]

Thionicotinamide is a known xenobiotic. It is a drug used for the treatment of *Chagas disease*, a common health problem in South America.[11,12] Some nitrofurans and nitroimidazoles have been used as drugs for this disease, but their carcinogenic and toxic effects, which are well-known, have precluded their use as effective therapeutic agents.[13] Some new pharmaceutical drugs, which include thionicotinamide, have been successfully tested[14] in an effort to renew and improve the toxic old ones which act to eliminate *Trypanosoma cruzi*, the etiologic agent of the Chagas disease.[15] This drug is related to homologues of nicotinamide or nicotinic acid because these compounds are used to synthesize NAD by *trypanosomes* and also by mammal, but the latter can produce it from other sources.[16] Thus, analogues of such compounds can mimic its action inhibiting the production of NAD by *trypanosomes*.



thionicotinamide

It has been surprising to us that no mechanistic studies have been performed on this very important compound. S-Oxygenation of thionicotinamide and its derivatives can form a good basis for further studies into the possible metabolic disposal of these types of compounds. We have thus embarked on a series of studies to provide both kinetic and mechanistic information regarding the oxidation of thionicotinamide. It is hoped that the

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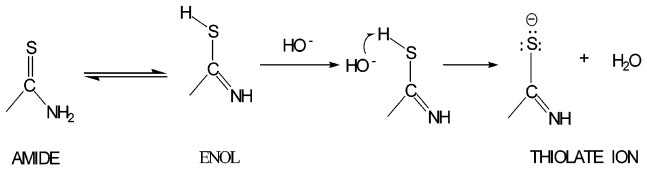
S-oxygenation mechanisms of thionicotinamide will aid in the understanding of both the physiological and toxicological implications of this vital xenobiotic as a pharmaceutical drug as well as its chemistry as a thioamide.

Experimental Section

Materials. The following reagent grade chemicals were used without further purification: thionicotinamide (Sigma), nicotinamide (Acros), sodium chloride, perchloric acid, 60−72% (Fisher), barium chloride, D_2O, and peracetic acid, 32 wt % (Aldrich).

Methods. The rapid reactions of thionicotinamide with peracetic acid were followed on a Hi-Tech Scientific SF61-DX2 double-mixing stopped-flow spectrophotometer. Slower reactions involving the reactions of the *S*-oxide formed following oxidation of thionicotinamide were monitored on a conventional Perkin-Elmer Lambda 2S UV−vis spectrophotometer. ^{1}H NMR spectra of thionicotinamide and its oxidation products were obtained on a JEOL GX 270 spectrometer using D_2O as the solvent and internal standard. All kinetics experiments were performed at 25.0 ± 0.5 °C and an ionic strength of 0.5 M (NaCl). Thionicotinamide is sparingly soluble in water at neutral pH conditions, but its solubility increased in highly acidic and highly basic environments. Stock solutions of thionicotinamide were prepared by first dissolving a known sample of thionicotinamide in concentrated acid followed by serial dilutions with water to attain the desired strength. Thionicotinamide has an intense yellow color. Addition of base discolored the solution due to the formation of the thiolate anion. Addition of acid restored the coloration as the keto form of thionicotinamide was reestablished.



Stoichiometric determinations were carried out by mixing various ratios of thionicotinamide and peracetic acid in tightly sealed volumetric flasks and scanning them spectrophotometrically for depletion of thionicotinamde over periods of up to 24 h. Excess peracetic acid could be determined by adding excess acidified iodide and back-titrating with standard thiosulfate using freshly prepared starch as indicator.

$$RCO_3H + 2I^- + 2H^+ \rightarrow RCO_2H + I_2 + H_2O \quad (R1)$$

Results

Stoichiometry. The stoichiometry of this reaction was very complex and depended on the ratios of the oxidant to the reductant. In excess thionicotinamide a 1:1 stoichiometric ratio was quickly established in which there appeared to be a quick and facile oxygen atom transfer from the peracetic acid to thionicotinamide. The establishment of this stoichiometry was almost instantaneous:

$$CH_3CO_3H + (C_5H_5N)C(=S)NH_2 \rightarrow (C_5H_5N)C(=NH)SOH + CH_3COOH \quad (R2)$$

The product in reaction R2 was the thionicotinamide *S*-oxide which can be written, as above, in the form of a stable

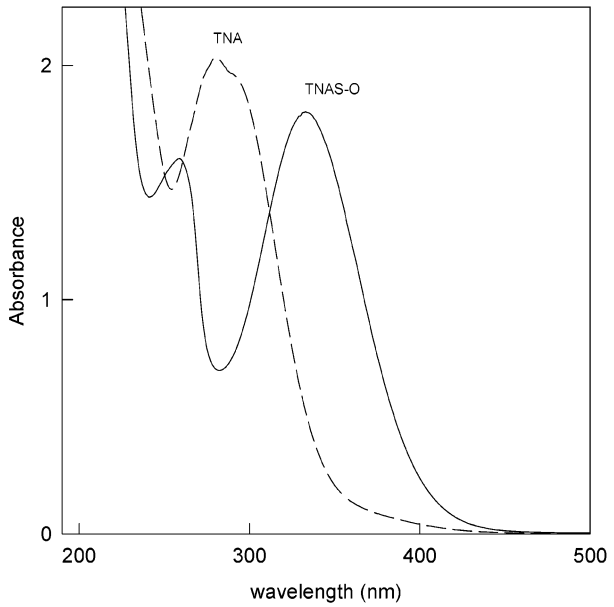
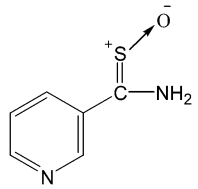


Figure 1. Spectra of thionicotinamide, TNA (dotted line) and its sulfoxide, TNAS−O (solid line) showing absorption peaks at 288 and 344 nm, respectively. The sulfoxide is formed from TNA in <1 s using peracetic acid as oxidant.

iminesulfenic acid but is better represented by the following structure:



In excess oxidant, the *S*-oxide is slowly oxidized to predominantly the sulfinic acid:

$$2CH_3CO_3H + (C_5H_5N)C(=S)NH_2 \rightarrow (C_5H_5N)C(=NH)SO_2H + 2CH_3COOH \quad (R3)$$

NMR data did not show the standard nicotinamide spectrum, which would suggest oxidation to sulfate, and instead showed a series of complex multiplets, and adding barium chloride did not detect sulfate in the form of insoluble barium sulfate. However, the exact ratio of oxidant to thionicotinamide was slightly higher than 2, suggesting possible formation of a sulfonic acid. Qualitative tests for ammonium ions were also negative.

Reaction Dynamics. The reaction displayed complex kinetics, which showed an initial rapid reaction that consumed peracetic acid but did not show a discernible change in the coloration of the thionicotinamide. Figure 1 shows data from the diode array stopped-flow spectrophotometer, which shows the UV−vis spectrum of TNA (dashed line). Upon mixing the reagents in the mixing chamber, the spectrum rapidly changes to a new one (solid line), which is not very different from the one of TNA. This is the spectrum of the thionicotinamide *S*-oxide (TNAS−O), which is almost instantaneously formed. The time lag between acquisition of the spectrum of TNAS−O after the acquisition of the TNA spectrum is less than 1 s. The experiment shown in Figure 1 was performed at excess oxidant. Further reaction will now proceed with the TNAS−O at a much slower rate. TNAS−O shows an absorption peak at 344 nm

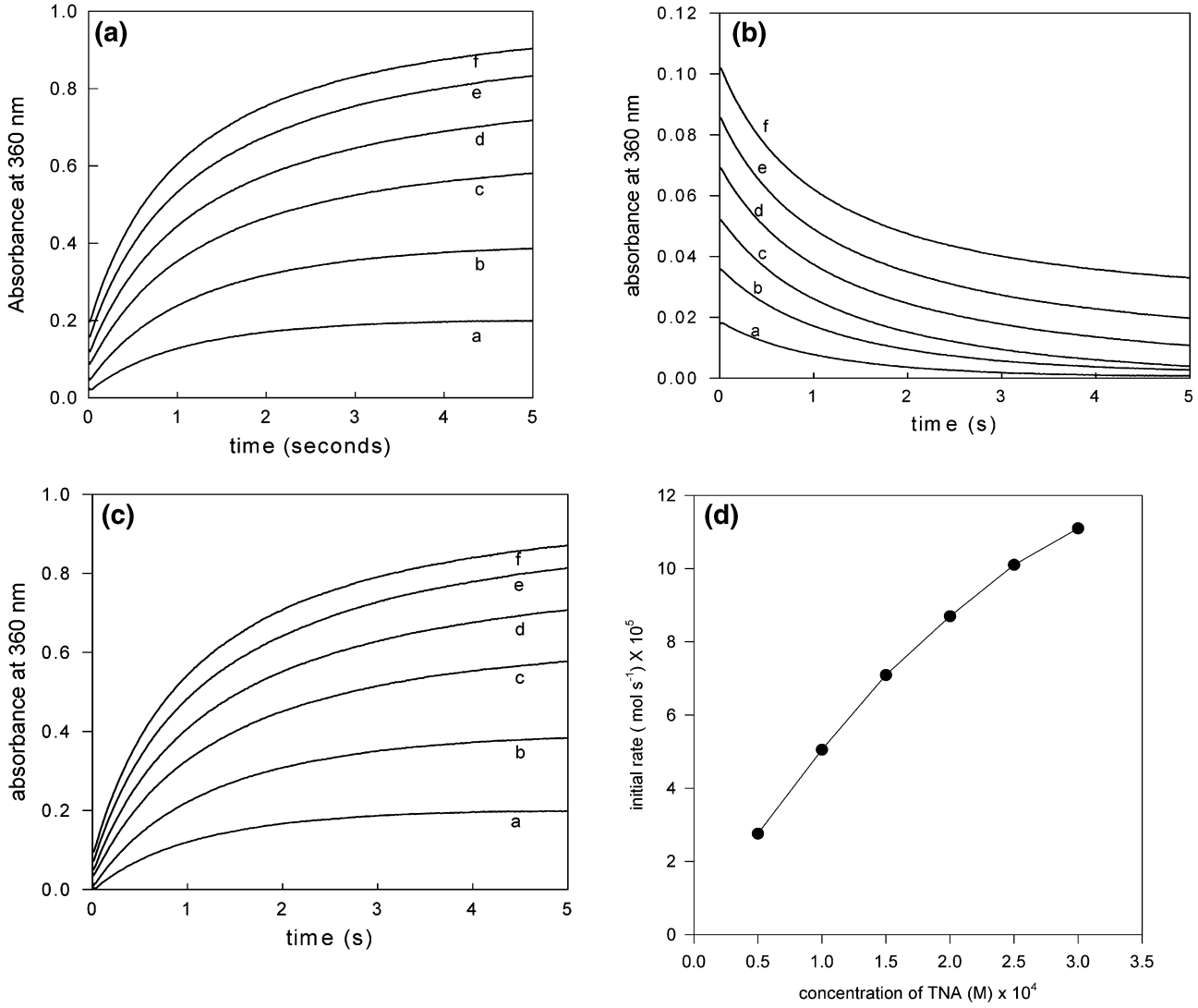


Figure 2. (a) Variation of [TNA] in its oxidation by peracetic acid showing total absorbance from TNA and TNAS−O at 360 nm. $[CH_3CO_3H] = 5.0 \times 10^{-4}$ M; $[H^+] = 0.05$ M; $I_{NaCl} = 1.0$ M; $[TNA]_o =$ (a) 5.0×10^{-5} M, (b) 1.0×10^{-4} M, (c) 1.5×10^{-4} M, (d) 2.0×10^{-4} M, (e) 2.5×10^{-4} M, (f) 3.0×10^{-4} M. (b) Concentration variations of TNA in its oxidation by peracetic acid showing unambiguous consumption of TNA at 360 nm. Conditions are the same as in Figure 2a. (c) Concentration variations of TNA in its oxidation by peracetic acid showing appearance of TNAS−O at 360 nm. These traces are obtained by subtracting at each data point the absorbances in Figure 2a from Figure 2b. Conditions are the same as in Figure 2a. (d) Plot of initial rates vs initial concentrations of TNA, $[TNA]_o$, from oxidation with peracetic acid. A very slight saturation is observed.

and TNA has a peak at 288 nm. TNAS−O has a large contribution to the absorbance observed at 288 nm and so does TNA at 344 nm. Absorbance traces at 344 nm would then show the resultant of the consumption of TNA and the formation of TNAS−O. An unambiguous rate of consumption of TNA is also not possible at 288 nm without including contributions from TNAS−O.

We can derive the following simple relationship for the absorbance at any wavelength, λ:

$$A_\lambda = A(\text{TNA}) + A(\text{TNAS}-\text{O}) \tag{1}$$

Using mass balance, the total thionicotinamide species are related as follows:

$$[\text{TNA}]_0 = [\text{TNA}] + [\text{TNAS}-\text{O}] \tag{2}$$

Assuming absorptivity coefficient of thionicotinamide and the *S*-oxide to be ϵ_1 and ϵ_2 respectively, the observed activity at any wavelength is given by

$$\frac{dA_\lambda}{dt} = \frac{\{[\text{TNA}](\epsilon_1 - \epsilon_2) + \epsilon_2[\text{TNA}]_0\}}{dt} \tag{3}$$

Equation 3 assumes a path length of 1 cm. $[TNA]_0$ is the initial concentration of thionicotinamide used and [TNA] is its instantaneous concentration. The instantaneous concentration of TNA at any time, *t*, can be derived from the integration of eq 3 and is analytically derived as

$$[\text{TNA}]_t = \frac{\{A_\lambda(t) - \epsilon_2[\text{TNA}]_0\}}{(\epsilon_1 - \epsilon_2)} \tag{4}$$

Equation 3 will be adequate in describing the total experimentally observed absorbance assuming no other species apart from TNA and TNAS−O. This is indeed the case in the initial stages of the reaction (reaction time less than 1 s) in which quantitative conversion of thionicotinamide to the *S*-oxide occurs and before any appreciable consumption of the *S*-oxide occurs. Peracetic acid has no contribution to the absorbance observed at either 288 or 360 nm and thus its variations will be ignored. Using eq 4, we can deconvolute the experimental data observed and obtain

unambiguous contributions from TNA and TNAS−O to the observed changes in absorbance.

The absorptivity coefficients of TNA and TNAS−O at 344 nm are both high and comparable. By shifting, however, to the 360 nm, the absorptivity coefficient of TNA went down to 368 M^{-1} cm^{-1} whereas that of TNAS−O was still high at 4141 M^{-1} cm^{-1}. For most of our kinetics experiments, we used 360 nm for kinetics measurements. Figure 2a shows the raw spectrophotometric data collected at 360 nm. It shows a fairly rapid increase in absorbance, which includes the formation of TNAS−O and the consumption of TNA. By using eq 4, we can trace the contribution to the absorbance due to TNA alone. This is shown in Figure 2b. The contribution from TNAS−O can be evaluated in the same manner and checked by subtracting the absorbance in Figure 2b from that in Figure 2a. These two treatments should give the same absorbance−time profiles. At $t = 0$, all the contribution to the observed absorbance is solely from TNA. At $t > 5$ s there is no contribution from TNA, as it would have been entirely consumed by then. Figure 2c shows the absorbance−time profile from TNAS−O contributions alone. Due to the much lower absorptivity coefficient of TNA at this wavelength, traces shown in Figure 2c are very similar to those in Figure 2a. However, the use of Figure 2a for kinetics data would have delivered erroneous initial rate values. There is a weak linear dependence of the consumption of TNA with $[TNA]_0$, with higher concentrations of TNA appearing to give a slight saturation (Figure 2d). As expected, the same type of dependence was observed for TNAS−O.

Figure 3a shows the effect of peracetic acid on the rate of reaction. Utilization, again, of eq 4 gives the absorbance changes derived solely from TNAS−O formation (Figure 3b). TNA is essentially completely consumed within 1 s. This is shown in Figure 3c. Concentrations of peracetic acid used in Figure 3a,b were much higher than those used in Figure 2a−d, and hence the observed rapid rates of consumption of TNA and formation of TNAS−O. A plot of initial rate of consumption of TNA vs initial concentrations of peracetic acid was linear with a slope slightly greater than unity with a slight saturation in higher peracid concentrations. A similar plot for the formation of TNAS−O gave the same dependence with respect to peracid. Equation 2, however, if correct in assuming that there are no other species formed in the initial stages of the reaction apart from the *S*-oxide, will insist on the same reaction order for both the formation of TNAS−O as well as consumption of TNA.

$$\text{initial rate} = -\mathrm{d}[\mathrm{TNA}]/\mathrm{d}t = \mathrm{d}[\mathrm{TNAS{-}O}]/\mathrm{d}t = k_0[\mathrm{TNA}]_0[\mathrm{CH_3CO_3H}]_0 \quad (5)$$

We evaluated a bimolecular rate constant $k_0 = (1.1 \pm 0.3) \times 10^3$ M^{-1} s^{-1}.

Effect of Acid. Acid, surprisingly, has a very profound effect on the dynamics of this reaction. Figure 4a shows that high acid concentrations retard the consumption of TNA and also increase the amount of transient TNA left before consumption of TNAS−O commences. High acid also reduces the final amount of TNAS−O formed before the observation of its slow consumption. A plot of initial rates vs inverse acid concentration is linear within a range of high acid concentrations (Figure 4b). This linearity was lost at low acid concentrations. The reaction rate rapidly decreased in highly acidic environments.

Consumption of TNAS−O. Further monitoring of the absorbance in Figure 3a,b will show a much slower decrease in the absorbance (consumption of TNAS−O). In conditions of excess oxidant, on the basis of the kinetic traces shown in Figures 2b, 3a, and 4a, we could assume that all the TNA would have been completely consumed at the point where consumption of TNAS−O commenced. Because the formation of TNAS−O is much more rapid than its consumption, the absorptivity

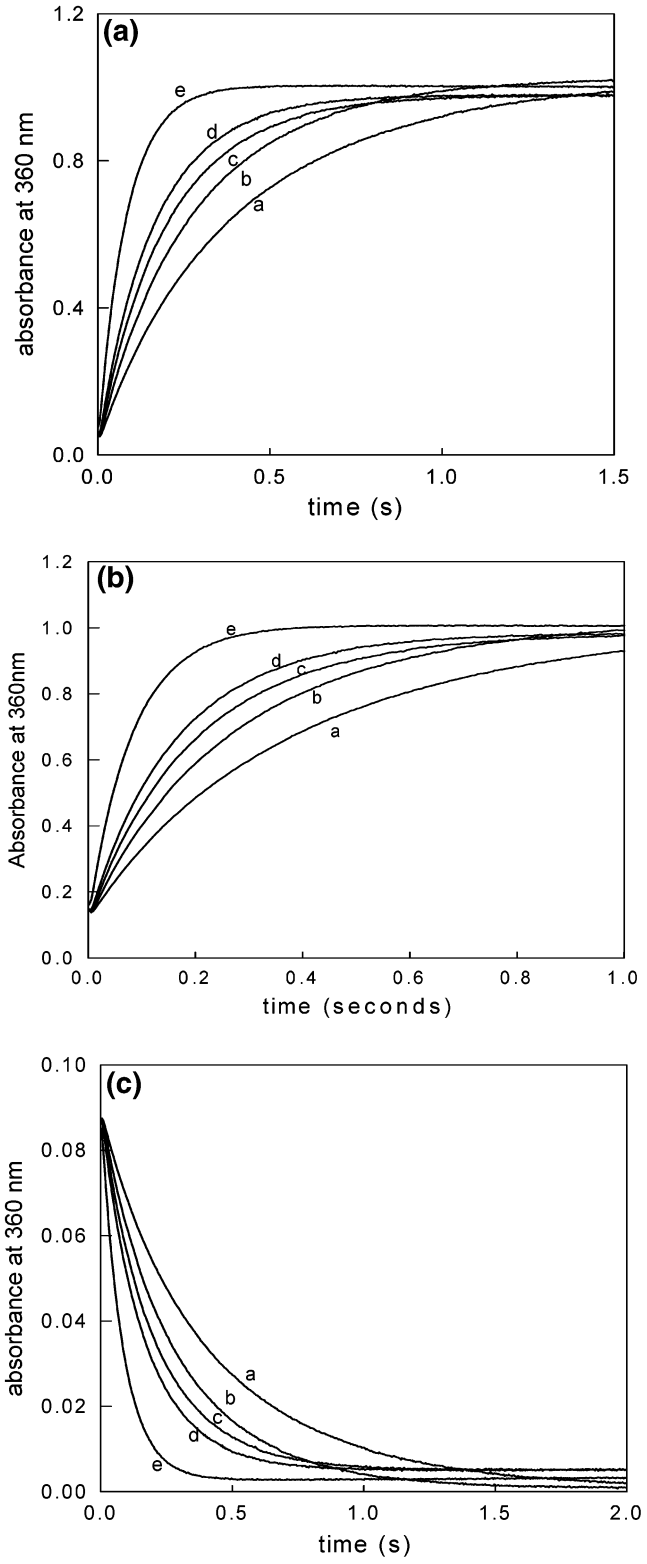


Figure 3. (a) Variation of $[CH_3CO_3H]$ in TNA oxidation by peracetic acid showing combined absorbance from TNA and TNAS−O. [TNA] $= 2.5 \times 10^{-4}$ M; $[H^+] = 0.05$ M, and $I_{NaCl} = 1.0$ M; $[CH_3CO_3H] =$ (a) 1.0×10^{-3} M, (b)1.5×10^{-3} M, (c) 2.0×10^{-3} M, (d) 2.5×10^{-3} M, (e) 5.0×10^{-3} M. (b) Variation of $[CH_3CO_3H]$ in TNA oxidation by peracetic acid showing formation of TNAS−O. The conditions are the same as in Figure 3a. (c) Variation of $[CH_3CO_3H]$ in TNA oxidation by peracetic acid showing consumption of TNA. The conditions are the same as in Figure 3a.

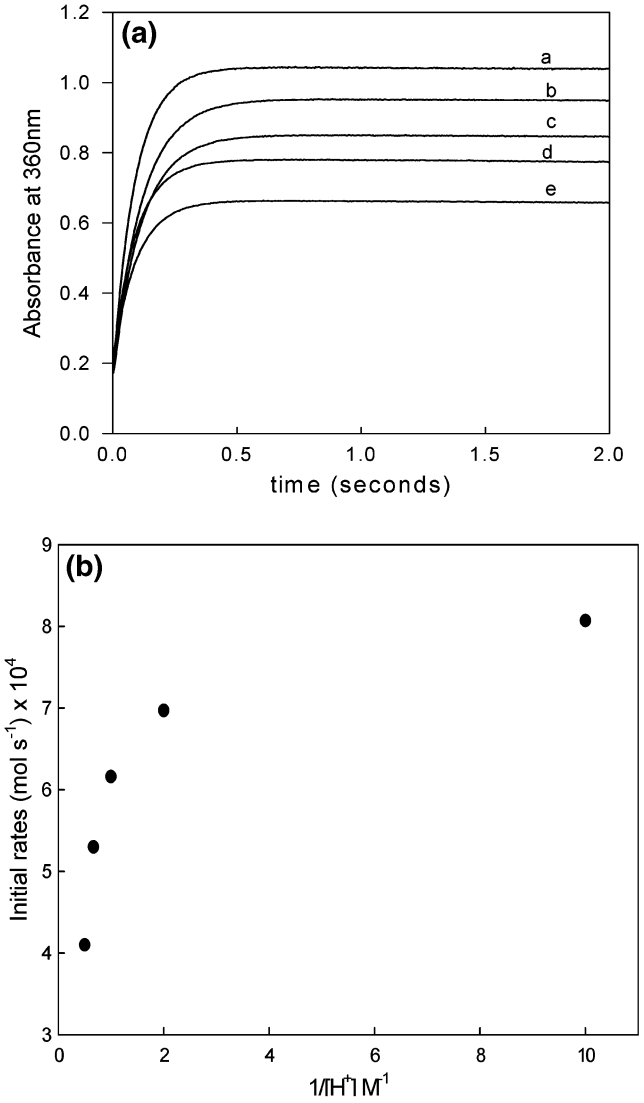
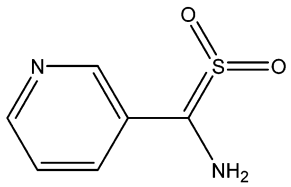


Figure 4. (a) Variation of $[H^+]$ in TNA oxidation by peracetic acid showing inhibiting effect of acid on reaction rates. [TNA] = 2.5×10^{-4} M; $[CH_3CO_3H] = 5.0 \times 10^{-3}$ M; $[H^+]$ = (a) 0.1 M, (b) 0.5 M, (c) 1.0 M, (d) 1.5 M, (e) 2.0 M. (b) Plot of initial rate as a function of reciprocal acid. Linearity is lost at low acid concentrations. [TNA] = 2.5×10^{-4} M; $[CH_3CO_3H] = 5.0 \times 10^{-3}$ M; $[H^+]$ = (a) 0.1 M, (b) 0.5 M, (c) 1.0 M, (d) 1.5 M, (e) 2.0 M.

coefficient of TNAS−O could be deduced in the region where contribution from TNA had decayed to zero and before the consumption of TNAS−O commenced. We evaluated an absorptivity coefficient for TNAS−O of 5105 M^{-1} cm^{-1} at 334 nm and a value of 4141 M^{-1} cm^{-1} at the observation wavelength of 360 nm. Figure 5a shows a series of traces that show this slow consumption of TNAS−O. The final product is the sulfinic acid of thionicotinamide after adding another oxygen atom to the *S*-oxide.



thionicotinamide S-dioxide

Data in Figure 5a gave a first order dependence on peracetic acid (see Figure 5a inset) Figure 5b shows the slow decay of

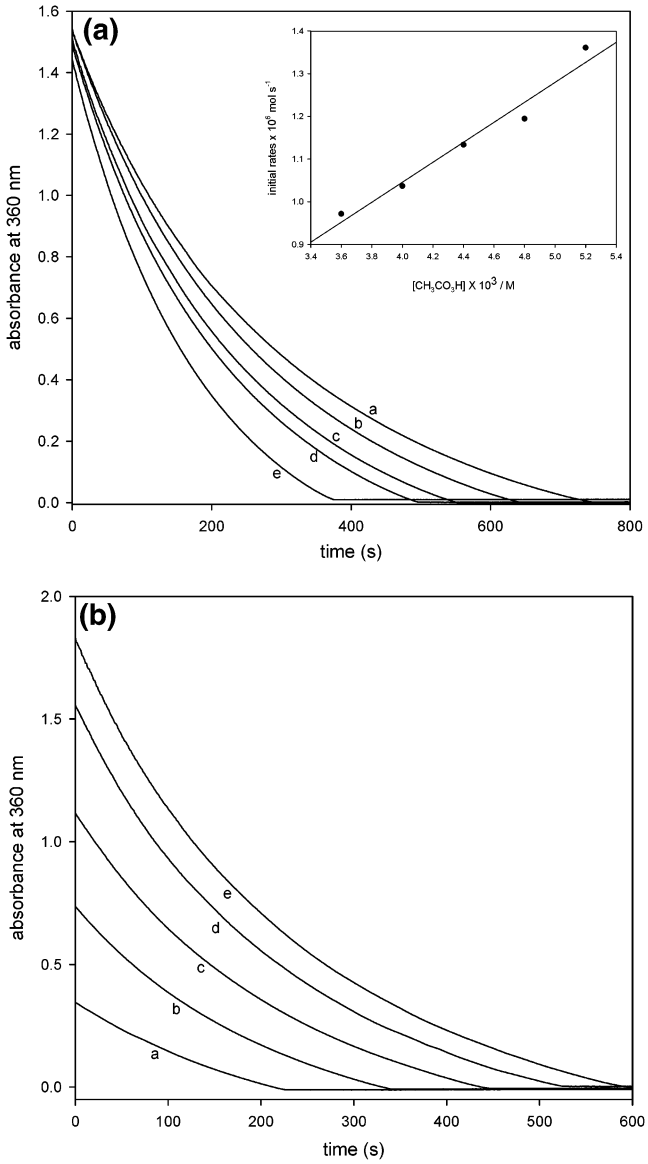


Figure 5. (a) Slow consumption of TNAS−O after instantaneous formation from TNA oxidation at different peracetic acid concentrations. [TNA] = 5.0×10^{-4} M, $[H^+] = 2.5 \times 10^{-4}$ M, I_{NaCl} = 1.0 M. $[CH_3CO_3H]$ = (a) 3.0×10^{-3} M, (b) 4.0×10^{-3} M, (c) 5.0×10^{-3} M, (d) 6.0×10^{-3} M, (e) 7.0×10^{-3} M. (b) Variation of $[TNA]_0$ in the slow oxidation of TNAS−O. This stage of the reaction is first order in $[TNA]_0$. $[RCO_3H]_0 = 4.0 \times 10^{-3}$ M; $[TNA]_0$ = (a) 1.0×10^{-3} M, (b) 2.0×10^{-3} M, (c) 3.0×10^{-3} M, (d) 4.0×10^{-3} M, (e) 5.0×10^{-3} M.

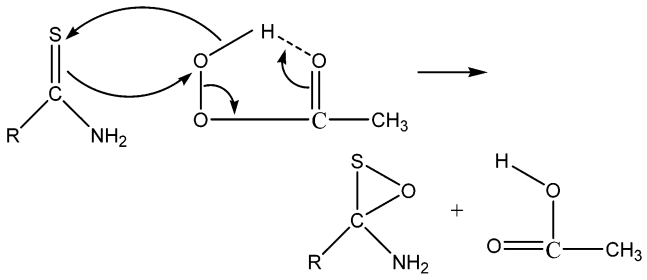
TNAS−O at varying TNA concentrations. Acid concentrations did not appear to exert much effect on the rate of consumption of TNAS−O. If we assume that all the TNA will have been transformed to TNAS−O at the point where the slow decomposition of TNAS−O is monitored; we can evaluate a lower limit bimolecular rate constant for the reaction of TNAS−O with peracetic acid.

$$-\mathrm{d[TNAS{-}O]}/\mathrm{d}t = k_1[\mathrm{TNAS{-}O}][RCO_3H]$$

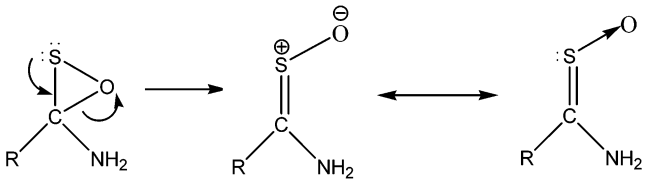
$$k_1 = 0.10 \pm 0.02\ M^{-1}\ s^{-1} \quad (6)$$

This assumption was checked by calculating the expected initial absorbance of TNAS−O based on amount of TNA used. In all cases, the observed absorbance was about 96% of that expected from a full conversion of TNA to TNAS−O. The small discrepancy could be explained by the longer sampling periods used for Figure 5a,b.

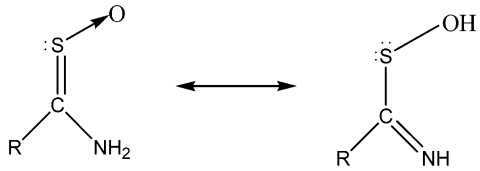
Mechanism. The vastly different rates of oxidation of TNA and of TNAS−O suggest that the mechanisms of these two oxidations are also quite different. Peracetic acid oxidations have been extensively studied and utilized in organic synthesis.[17] For compounds with double bonds, epoxidation appears to be the dominant pathway in which an oxygen atom can be transferred to the reductant.[18] We propose that the initial step involves an attack by the peracetic acid on the less hindered side of thioamide group to produce an epoxide-like intermediate, which rapidly rearranges to produce the *S*-oxide (in this mechanism, "R" represents the pyridine ring):



followed by the rearrangement



This *S*-oxide can also be represented, by resonance stabilization, as an iminesulfenic acid



Though we might expect the intermediate epoxide to be optically active, the final *S*-oxide should not be. This conversion is extremely rapid and, in excess peracetic acid, should be over within 1 s. In the absence of further oxidizing agent, the *S*-oxide is stable enough to be the final product.

Further oxidation of the sulfenic acid involves primarily an oxygen atom transfer without the benefit of a stabilizing epoxide intermediate. Sulfenic acids are well-known electrophiles and will combine with any nucleophile in solution (especially the parent thioamide to produce possible S−S bonds). The nucleophilic nature of the sulfur center in thionicotinamide, with two lone pairs of electrons will rapidly react with the electrophilic peracetic acid. Thus further reaction of the sulfenic acid will be comparatively slower due to electrostatic factors. The addition of the OH group on the sulfur atom also increases stearic hindrance around the sulfur atom, resulting in a much slower reaction.

Effect of Acid. Acid retards only the first stage of the reaction and reduces the amount of transient amount of TNAS−O formed before its consumption commences. Thioamides, in low pH conditions, will be protonated with the proton attached somewhere between the amine group and the sulfur center but with a stronger tilt toward the more nucleophilic sulfur atom.

$$R(NH_2)C{=}S + H^+ \rightleftharpoons [R(NH_2)C{=}S{-}H]^+ \qquad K_a \qquad \text{(R4)}$$

This protonated thioamide can be considered inert, or if it reacts, it would react very slowly, on the time scale of the oxidation of TNAS−O. If it is completely inert, then eq 5, for the rate of formation of TNAS−O becomes

$$\text{Rate} = -\text{d}[\text{TNA}]/\text{d}t = k_0[\text{TNA}]_0[\text{RCO}_3\text{H}]/(1 + K_a^{-1}[\text{H}^+]) \qquad (7)$$

The acid's retarding effect can be evaluated from eq 7. Low acid concentrations, in which $1 \gg K_a[H^+]$, will show limited to no acid effect. This is what was observed in Figure 4b. The value of k_0, which we evaluated of $(1.1 \pm 0.3) \times 10^3\ M^{-1}\ s^{-1}$, was an upper limit rate in the limit of low acid concentrations in which there is no discernible acid retardation.

Conclusion

In the absence of metal ion catalysts and standard P450-type enzymes[19] or the flavin-containing monooxygenases;[20] the oxidation of thionicotinamide by a mild oxidizing agent produces predominantly the sulfinic acid. The sulfinic acid formed is readily soluble in water when compared with the parent thioamide. This increase in solubility could be a mechanism by which such a xenobiotic can be eluted from the physiological environment.

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References and Notes

(1) Griffiths, M. H.; Moss, J. A.; Rose, J. A.; Hathway, D. E. *Biochem. J.* **1966**, *98*, 770−781.
(2) Tietz, E. I.; Roth, T.; Zorick, F. J.; Piccione, P.; Kramer, M. *Arzneimittelforschung* **1981**, *31*, 1963−1966.
(3) Shepard, C. C.; Jenner, P. J.; Ellard, G. A.; Lancaster, R. D. *Int. J. Lepr. Other Mycobact. Dis.* **1985**, *53*, 587−594.
(4) Waisser, K.; Celadnik, M.; Palat, K.; Odlerova, Z. *Cesk. Farm.* **1982**, *31*, 303−307.
(5) McCann, J.; Choi, E.; Yamasaki, E.; Ames, B. N. *Proc. Natl. Acad. Sci. U.S.A* **1975**, *72*, 5135−5139.
(6) Neal, R. A.; Halpert, J. *Annu. Rev. Pharmacol. Toxicol.* **1982**, *22*, 321−339.
(7) Park, S. B.; Howald, W. N.; Cashman, J. R. *Chem. Res. Toxicol.* **1994**, *7*, 191−8.
(8) Park, S. B.; Osterloh, J. D.; Vamvakas, S.; Hashmi, M.; Anders, M. W.; Cashman, J. R. *Chem. Res. Toxicol.* **1992**, *5*, 193−201.
(9) Benoit, E.; Buronfosse, T.; Moroni, P.; Delatour, P.; Riviere, J. L. *Biochem. Pharmacol.* **1993**, *46*, 2337−2341.
(10) Guo, W. X.; Poulsen, L. L.; Ziegler, D. M. *Biochem. Pharmacol.* **1992**, *44*, 2029−2037.
(11) Wanderley, D. M.; Correa, F. M. *Rev. Paul Med.* **1995**, *113*, 742−749.
(12) Wendel, S. *Curr. Opin. Hematol.* **1998**, *5*, 406−411.
(13) Murta, S. M.; Gazzinelli, R. T.; Brener, Z.; Romanha, A. J. *Mol. Biochem. Parasitol.* **1998**, *93*, 203-214.
(14) Kinnamon, K. E.; Poon, B. T.; Scovill, J. P.; Hanson, W. L.; Waits, V. B. *Mil. Med.* **1997**, *162*, 527−532.
(15) Kirchhoff, L. V. *Gastroenterol. Clin. North Am.* **1996**, *25*, 517−533.
(16) Zubrzycki, I. Z. *Biophys. J.* **2002**, *82*, 2906−2915.
(17) Morimoto, T.; Hirano, M.; Wachi, M.; Murakami, T. *J. Chem. Soc., Perkin Trans. 2* **1984**, 1949−1951.
(18) Murphy, A.; Dubois, G.; Stack, T. D. P. *J. Am. Chem. Soc.* **2003**, *125*, 5250−5251.
(19) Murataliev, M. B.; Feyereisen, R. *Biochemistry* **2000**, *39*, 5066−5074.
(20) Madan, A.; Faiman, M. D. *Drug Metab Dispos.* **1994**, *22*, 324−330.