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Synthesis of 2-Quinuclidonium by Eliminating Water: Experimental Quantification of the High Basicity of Extremely Twisted Amides

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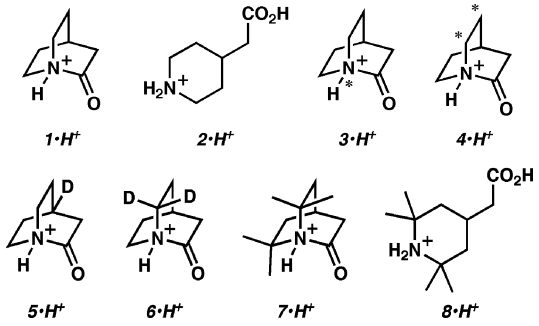
The amide or peptide bond plays an essential role in biology as the linking functionality between amino acids in proteins. Typically peptide bonds are stable, with half-lives in neutral aqueous solution exceeding hundreds of years.[1] In part, this stability is due to resonance stabilization between π-orbitals of the O−C−N linkage.[2] Resonance also leads to the planar geometry of the peptide bond. When this preferred geometry is disrupted and resonance no longer occurs, the stability and chemistry of the amide functionality changes dramatically.[3] This instability prevented the synthesis of 2-quinuclidone (**1**) until only recently,[4] despite the apparent simplicity of this compound.[5] Now that **1** is available, it is an excellent model for understanding twisted amide chemistry. Herein we present the first experimental results characterizing the basicity of **1**, which is found to be much higher than typical for amides. In addition, the unique gas-phase dissociation chemistry of **1**·$\mathbf{H}^+$ is described, and a second synthetic route to **1**·$\mathbf{H}^+$, which occurs only in the gas phase, is revealed.



The kinetic method, which relies on competitive fragmentation of proton bound dimers, was employed to determine the proton affinity (PA) of **1** relative to a series of reference bases (shown in Figure 1) according to previously established methods.[6] Briefly, dimers were introduced into an LTQ linear ion trap mass spectrometer by electrospraying dry acetonitrile containing the tetrafluoroborate salt of **1** and a reference base. The noncovalently bound dimers were then subjected to collision induced dissociation (CID) to determine the most basic site (which retains the proton more often). The results are shown in Figure 1. Analysis of the data yields a PA of 965.0 kJ/mol for **1** using the simple kinetic method. Application of the more rigorous extended kinetic method[7] yields a value of 964.2 kJ/mol, suggesting that entropic effects have a minimal impact on the measured PA. Calculations at the B3LYP 6-311++G** level of theory yield a PA of 944.3 kJ/mol. Previous calculations predicted a PA of 958.4 kJ/mol.[3b] Thus **1** is found to be very basic by theory and experiment.

By comparison, typical amides have PAs in the range of 880−900 kJ/mol.[8] In terms of basicity, **1** behaves more like a secondary

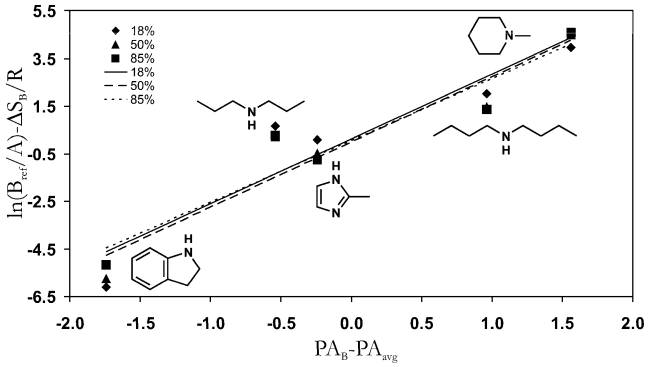


Figure 1. Data from kinetic method experiments showing the relative PA versus natural log of the ratio of ion intensities minus protonation entropies.[13] Three representative collision energies are shown for each reference base. The collinearity of all three lines indicates few entropic effects. The PA of **1** is determined to be 964.2 kJ/mol by the extended kinetic method.

or tertiary amine owing to the lack of resonance within the amide. In addition, the site of protonation differs for twisted amides with protonation at the nitrogen being favored by ~90 kJ/mol according to our calculations. In the process of collecting data to establish the PA of **1**, reference bases were found to separate into two groups. The less bulky bases give the data shown in Figure 1, which corresponds to dimers which are capable of hydrogen bonding to the nitrogen of **1**. The remaining reference bases are too bulky to access the nitrogen and presumably interact with the carbonyl oxygen of **1**.[9]

The properties of **1** were explored further by collision induced dissociation (CID) experiments. The CID spectrum for **1**·$\mathbf{H}^+$ is shown in Figure 2a,*i*. Surprisingly, a single loss of 44 Da is the only major product that is observed, indicating that a single fragmentation pathway is energetically favored. Because of the bicyclic nature of **1**·$\mathbf{H}^+$, two covalent bonds must be broken to observe fragmentation. A loss of 44 Da further requires at least one hydrogen transfer. We propose the mechanism shown in Scheme 1 to account for the observed loss. Homolytic cleavage of the amide bond leads to abstraction of one out of two equivalent hydrogens facing the radical. Two possible McLafferty-type rearrangements (one is shown in Scheme 1) then lead to the second hydrogen transfer and the loss of ethenol. In order to verify this mechanism, a series of four compounds labeled with stable isotopes were prepared (**3**−**6**). CID of **3**·$\mathbf{H}^+$ yields a single product which retains the ^{15}N label as expected.[9] Similarly, **4**·$\mathbf{H}^+$ and **5**·$\mathbf{H}^+$ both fragment by yielding a single observable product with the ^{13}C and deuterium labels retained, respectively, in agreement with our mechanism.[9] Additionally, fragmentation of **6**·$\mathbf{H}^+$ confirms that hydrogen transfer occurs. In this case, two products are observed, with the difference between them being the loss or retention of a deuterium.[9] The loss of hydrogen is favored by a factor of 1.7, suggesting that isotope effects[10] may play a role in this reaction. Nevertheless, in each experiment the labeled atoms were lost or

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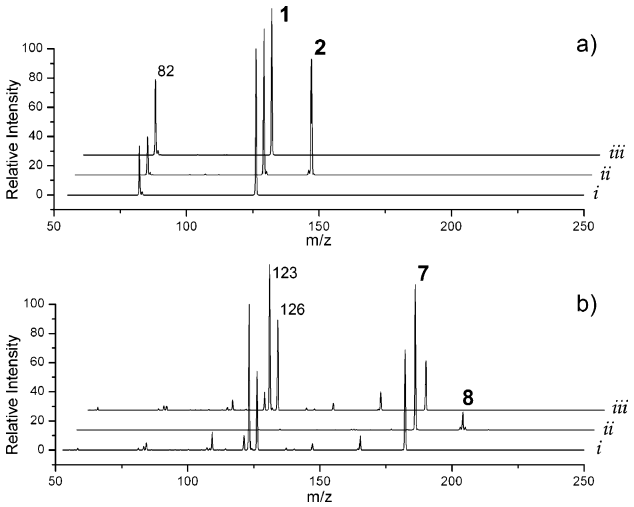
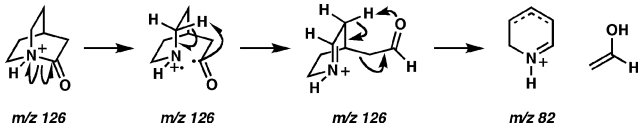
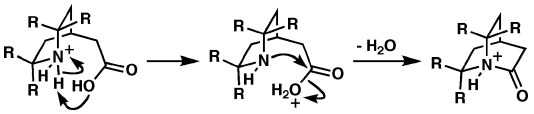


Figure 2. (a) (*i*) CID spectrum of $\mathbf{1}\cdot\mathbf{H}^+$, with a single fragment being detected. (*ii*) CID spectrum of $\mathbf{2}\cdot\mathbf{H}^+$. The loss of water generates $\mathbf{1}\cdot\mathbf{H}^+$ which spontaneously fragments. (*iii*) MS^3 CID spectrum of the reisolated peak at *m*/*z* 126 from spectrum *ii* confirming that $\mathbf{1}\cdot\mathbf{H}^+$ is generated by the loss of water. (b) (*i*) CID spectrum of $\mathbf{7}\cdot\mathbf{H}^+$. (*ii*) CID spectrum of $\mathbf{8}\cdot\mathbf{H}^+$. In this case, the synthesis proceeds cleanly without spontaneous fragmentation. (*iii*) MS^3 CID spectrum showing that all fragment peaks are reproduced when the gas-phase product is compared to the bona-fide sample in spectrum *i*.

Scheme 1



Scheme 2



retained in agreement with the mechanism shown in Scheme 1. As predicted, the amide bond is weakened by the lack of resonance stabilization and is the first bond to break upon collisional excitation.

Further insight into the chemistry of twisted amides can be obtained by synthesizing them in the absence of solvent. **1** is observed to rapidly hydrolyze in the presence of water.[4] Attempts to drive the reverse reaction in solution have been unsuccessful.[3a] Similarly, attempts to synthesize **1** with the acid chloride of **2** have met with frustration.[11] Nevertheless, collisional excitation of the hydrolyzed derivative $\mathbf{2}\cdot\mathbf{H}^+$ in the gas phase yields quantitatively a product with the same mass as $\mathbf{1}\cdot\mathbf{H}^+$ as shown in Figure 2a*ii*. Following reisolation and collisional cooling of this peak, the MS^3 CID spectrum is identical to that obtained by fragmenting $\mathbf{1}\cdot\mathbf{H}^+$ (compare Figure 2a*i* and 2a*iii*). Similarly, all isotopically labeled compounds react exclusively by eliminating water, followed by the same elimination that would be expected if $\mathbf{1}\cdot\mathbf{H}^+$ were generated as the product.[9] Thus it is possible to selectively synthesize $\mathbf{1}\cdot\mathbf{H}^+$ by eliminating water from $\mathbf{2}\cdot\mathbf{H}^+$ as shown in Scheme 2 (for R = H) if the water can be rigorously removed from the reaction system. This is not a difficulty in the gas phase; however, the data in Figure 2a*ii* also suggests that there is a high barrier to this process. Elimination of water to yield $\mathbf{1}\cdot\mathbf{H}^+$ also results spontaneously in further fragmentation. As mentioned above, this requires the cleavage of two covalent bonds. Therefore, this reaction appears to be difficult in solution for two reasons: a high barrier to activation and back reactions with water.

These results are further confirmed by examination of **7**, which has four additional methyl groups. This compound can be generated from the acid chloride in solution.[11,12] CID of the hydrolyzed product $\mathbf{8}\cdot\mathbf{H}^+$ yields exclusively $\mathbf{7}\cdot\mathbf{H}^+$ without the accompanying loss of additional fragments. The synthesis is again confirmed by comparing fragmentation with the genuine molecule; comparison of Figure 2b*i* with Figure 2b*iii* reveals that even very low abundance peaks are reproduced. In addition, the voltage amplitude required to carry out the reaction (Scheme 2, R = Me) is 20% lower in magnitude when compared to the voltage required where R = H. Thus, the energy required to generate $\mathbf{7}\cdot\mathbf{H}^+$ by eliminating water is much lower, in agreement with the observed synthetic routes in solution. The gas-phase syntheses suggest that **7** is more nucleophilic than **1** and should therefore be more basic as well. Attempts to determine the PA of **7** experimentally by the kinetic method met with frustration. The steric hindrance of the additional methyl groups prevents access to the bridgehead nitrogen. However, theory can be used to estimate the proton affinity. The calculated PA for **7** at the B3LYP/6-311++G** level is 982.0 kJ/mol. This value is significantly higher than that for **1**, which supports the idea of enhanced nucleophilicity for **7**. However, **7** is also much more stable toward hydrolysis, indicating that stability does not share a simple relationship with basicity for twisted amides.[12]

In summary, we have quantitatively assessed the basicity of an extremely twisted amide for the first time and established that the gas-phase chemistry of these molecules closely reflects the properties observed in solution.

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Supporting Information Available: Detailed syntheses and other experimental procedures. This material is available free of charge via the Internet at http://pubs.acs.org.

References

(1) Smith, R. M.; Hansen, D. E. *J. Am. Chem. Soc.* **1998**, *120*, 8910−8913.
(2) (a) Greenberg, A.; Venanzi, C. A. *J. Am. Chem. Soc.* **1993**, *115*, 6951−6957. (b) Wiberg, K. B. Origin of the Amide Rotation Barrier. In *The Amide Linkage*; Greenberg, A., Breneman, C. M., Liebman, J. F., Eds.; Wiley: New York, 33−45.
(3) (a) Johnson, J. R.; Woodward, R. B.; Robinson, R. The Constitution of the Penicillins. *The Chemistry of Penicillin*; Clark, H. T., Johnson, J. R., Robinson, R., Eds.; Princeton University Press: Princeton, NJ, 1949; pp 440−454. (b) Greenberg, A.; Moore, D. T.; DuBois, T. D. *J. Am Chem. Soc.* **1996**, *118*, 8658−8668.
(4) Prepared as the HBF_4 salt, please see: Tani, K.; Stoltz, B. M. *Nature* **2006**, *441*, 731−734.
(5) (a) Wasserman, H. H. *Nature* **2006**, *441*, 699−700. (b) Clayden, J.; Moran, W. J. *Angew. Chem., Int. Ed.* **2006**, *45*, 7118−7120.
(6) (a) Zheng, X.; Cooks, R. G. *J. Phys. Chem. A* **2002**, *106*, 9939−9946. (b) Bouchoux, G.; Sablier, M.; Berruyer-Penaud, F. *J. Mass Spectrom.* **2004**, *39*, 986−997.
(7) (a) Wu, Z.; Fenselau, C.; Cooks, G. R. *Rapid Commun. Mass. Spectrom.* **1994**, *8*, 777−780. (b) Cerda, B. A.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1995**, *117*, 9734−9739. (c) Armentrout, P. B. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 371−379.
(8) Hunter, E. P.; Lias, S. G. Proton Affinity Evaluation. In *NIST Chemistry WebBook, NIST Standard Reference Database Number 69*, Linstrom, P. J., Mallard, W. G., Eds.; National Institute of Standards and Technology, Gaithersburg, MD, 2005; 20899 (http://webbook.nist.gov).
(9) Please see Supporting Information for details.
(10) Westheimer, F. H. *Chem. Rev.* **1961**, *61*, 265−273.
(11) Pracejus, H.; Kehlen, M.; Kehlen, H.; Matschiner, H. *Tetrahedron* **1965**, *21*, 2257−2270.
(12) Greenberg, A.; Wu, G.; Tsai, J.-C.; Chiu, Y.-Y. *Struct. Chem.* **1993**, *4*, 127−129.
(13) Hunter, E. P.; Lias, S. G. *J. Phys. Chem. Ref. Data* **1998**, *27*, 413−656.

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