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"Weighing" Photon Energies with Mass Spectrometry: Effects of Water on Ion Fluorescence

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Fluorescence is widely used to detect molecules with high sensitivity and is an excellent probe of molecular structure and dynamics owing to its sensitivity to the microenvironment of the fluorophore. By comparison, fluorescence detection of gas-phase ions has been largely limited by challenges associated with efficiently collecting the isotropically emitted photons from a limited number of ions and from background scattering. Despite these challenges, laser-induced fluorescence of trapped ions has been reported[1] for various fluorinated benzene cations,[1a] laser dye cations,[1b−f] and dye tagged biomolecules.[1g,h] One of the advantages and ultimate goals of applying optical spectroscopy techniques to charged ions *in vacuo* is to better understand the effects of water solvation on the physical properties and structures of ions. For example, Parks and co-workers used Förster resonance energy transfer (FRET) to investigate how complementary strands of a dye labeled double stranded oligonucleotide helix complex dissociates in the gas phase.[1h] By investigating fluorescence or FRET of ions with a controlled extent of hydration, detailed information about how water affects molecular structure, excited state chemistry, and dynamics can potentially be obtained. However, such studies have not been performed largely due to low abundances and short lifetimes of extensively hydrated ions and relatively low fluorescence photon collection efficiencies.

When isolated hydrated ions are electronically excited, conversion of electronic to internal energy can occur rapidly. The resulting energy is released by evaporating water molecules from the cluster. For example, electron capture by size-selected, thermalized, and extensively hydrated metal ions results in a large number of water molecules lost from the reduced precursors.[2] The energy deposited into the ions can be obtained from the average number of water molecules lost from the clusters, the sum of the threshold binding energies of the lost water molecules, and the total energy that partitions into the translations, rotations, and vibrations of the products.[2a] However, if an excited ion fluoresces, the energy of the photon that is emitted is not available to convert into internal modes of the cluster, so fewer water molecules are lost. For laser-induced ion fluorescence, the energy of the fluorescent photon that is emitted can be obtained from the difference in energy of the photon absorbed by the ion and the energy required to evaporate the observed number of water molecules from the cluster that has fluoresced. A key advantage of this indirect detection method is that *all dissociation products resulting from emission are observed, irrespective of the direction of which the photon is emitted*; i.e., this is equivalent to 100% photon collection efficiency, which makes this indirect method for measuring fluorescence in the gas phase highly sensitive. Here, we report the first results using this method to measure laser-induced ion fluorescence of hydrated ions.

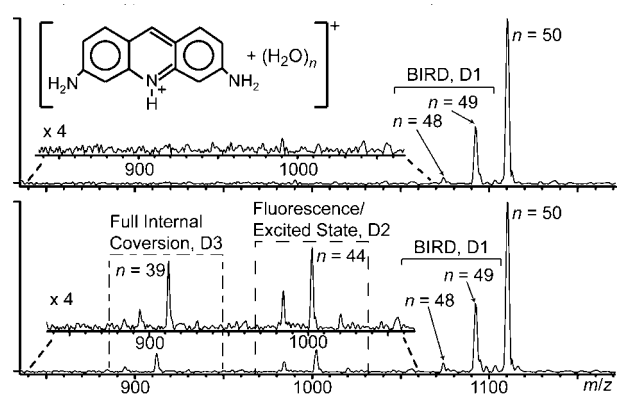

Figure 1. UV photodissociation spectrum of $PH^+(H_2O)_{50}$ irradiated for 250 ms (bottom) showing two resolved distributions in which ∼11 (D3) and ∼6 (D2) water molecules are lost compared to the reference spectrum (top) taken under identical conditions except without 248 nm laser irradiation.

Experiments were performed on a 2.75 T Fourier-transform ion cyclotron resonance mass spectrometer equipped with an external nanoelectrospray source and an ion cell cooled to 133 K.[3] Effects of temperature on nanocalorimetry experiments are discussed elsewhere.[3b] A KrF excimer laser (248 nm, ∼1 W average power) was used to irradiate size-selected and thermalized hydrated protonated proflavine ions, $PH^+(H_2O)_n$, $n = 13-50$, for 250 ms. After ion irradiation, a delay of 50 ms to 3 s prior to ion detection is used to investigate the dynamics of long-lived electronic excited states accessed upon absorption of a 248 nm photon.

Three distinct product ion distributions are observed in these experiments (Figure 1, data for $n = 50$ shown). Up to two water molecules are lost from $PH^+(H_2O)_{50}$ as a result of absorption of blackbody photons (D1). Upon absorption of a single 248 nm photon (5.0 eV), two fully resolved distributions of product ions corresponding to the loss of ∼6 (D2) and ∼11 (D3) water molecules are observed. The widths of these distributions are remarkably narrow (≤3 water molecules) relative to the number of water molecules lost. Under these conditions, collisional and radiative cooling are negligible. A parallel distribution shifted by ∼11 water molecules from D2 and D3 is observed (17% of the sum of D2 and D3 abundances) as a result of absorption of an additional photon.

The distribution corresponding to the loss of ∼11 water molecules (D3) results from absorption from the ground electronic state (S_0) to an excited singlet state (S_i, $i \geq 4$, corresponding to a strong absorption band in solution; see Supporting Information) followed by full internal conversion of the electronic energy into internal modes of the precursor, i.e., nonradiative relaxation. The

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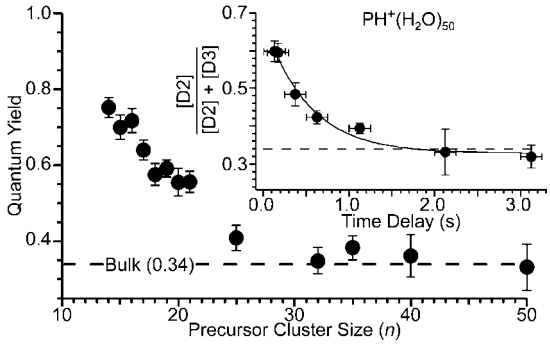

Figure 2. $PH^+(H_2O)_n$ laser-induced fluorescence quantum yield vs n and that for bulk $PH^+(aq)$ (dashed lines). The normalized product ion abundances of D2 as a function of the delay between laser irradiation and ion detection for $PH^+(H_2O)_{50}$ (inset).

energy removed per water molecule lost (0.47 eV) for this distribution is slightly higher than the binding energy of a water molecule to a large ionic water cluster (~0.4 eV)[4] because some energy partitions into translations and rotations of the products. The energy deposition necessary to form this product ion distribution can be determined from the average number of water molecules lost, the sum of the sequential water binding energies obtained from a discrete Thomson liquid drop model, and a statistical model for energy partitioning; this modeling is described in detail elsewhere.[2a] A value of 4.6 eV is obtained, indicating that the model underestimates the true energy deposition by ~8% for the $n = 50$ ion. The average deviation between the modeled and true value is 0.16 eV (~3%) over the cluster size range investigated. This excellent agreement indicates that protonated proflavine is *not* photodissociated in these clusters which would reduce the energy available for water evaporation.

The other laser-induced product ion distribution (D2) corresponds to a significantly fewer number of water molecules lost (an average of 5.9 for $n = 50$). The normalized abundance of D2 decreases exponentially with time (half-life of ~0.5 s for $n = 50$) to an asymptotic value of 0.33 ± 0.01 after 2 s, whereas the abundance of D3 increases over this same time frame (Figure 2, inset). Formation of D2 is attributed to two different photophysical processes. Upon absorption of a 248 nm photon, nonradiative relaxation to the first singlet excited state (S_1) rapidly occurs. A fraction of the S_1 population promptly fluoresces to S_0, and the remaining population undergoes intersystem crossing to a long-lived near degenerate triplet state which slowly undergoes nonradiative intersystem crossing to S_0. We conclude from these kinetic data for $PH^+(H_2O)_{50}$ that, upon absorption of a 248 nm photon, ~1/3 of the ions nonradiatively relax to S_0 directly forming D3, ~1/3 of the ions fluoresce to form D2, and ~1/3 of the ions form a long-lived triplet state (initially D2) that undergoes nonradiative intersystem crossing to S_0 ($\tau \sim 0.5$ s) forming D3. The asymptote of 0.33 to which D2 decays (Figure 2 inset) corresponds to the fluorescence quantum yield of $PH^+(H_2O)_{50}$, which is the same as the fluorescence quantum yield for $PH^+(aq)$ (0.34 ± 0.02, $\lambda_{excite} = 436$ nm).[5] The fluorescence quantum yield as a function of hydration extent (obtained with a 2 s delay prior to ion detection) is shown in Figure 2. These values decrease from 0.75 ± 0.03 for $n = 13$, to an average of 0.36 ± 0.02 for $n \geq 30$, indicating convergence to the bulk value for these larger clusters. The higher fluorescent yield at smaller cluster sizes is likely due to less efficient nonradiative relaxation from S_1 to S_0 in the cluster as a result of decreased fluorophore solvation; i.e., there are fewer water molecules to facilitate the conversion of electronic-to-vibrational energy.

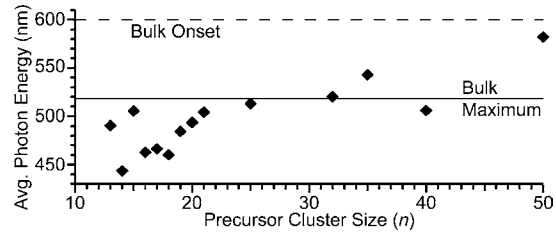

Figure 3. Average energies of fluorescent photons emitted from $PH^+(H_2O)_n$ upon laser-induced excitation at 248 nm. The bulk maximum and onset for $PH^+(aq)$ fluorescence[6] are indicated by solid and dashed lines, respectively.

The energy of the fluorescent photon is obtained from the difference in the average number of water molecules lost from the precursor for each of the distributions corresponding to ion fluorescence (D2, 2 s delay) and full internal conversion (D3) (see Supporting Information).[2a] The emitted photon energies as a function of hydration extent from $n = 13-50$ are given in Figure 3. The photon energies generally red shift from ~450 nm to ~580 nm with increasing hydration, although there are large oscillations at the smallest sizes investigated. The onset of the $PH^+(aq)$ fluorescence spectrum is 600 nm, and the maximum is 518 nm.[6] For $n = 25-40$, the emitted photon energies are within 30 nm of the bulk emission maximum, and the value for $n = 50$ is bracketed by the bulk fluorescence onset and maximum. The red shift is consistent with water stabilizing the excited state more effectively than the ground state.

This unorthodox and new method for indirectly probing ion fluorescence and triplet state formation has the advantage of high sensitivity (*the equivalent of 100% collection efficiency of the emitted photons)*, and no optical detection equipment is required. The high sensitivity of this method should make it possible to perform FRET experiments with gas-phase biomolecules in a microsolvated environment to investigate how a controlled number of water molecules facilitates dynamical motions in proteins or other molecules of interest.

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Supporting Information Available: Discussion of $PH^+(aq)$ electronic excited states and modeling the energy deposition. This information is available free of charge via the Internet at http://pubs.acs.org.

References

(1) (a) Cage, B.; McFarland, M. A.; Hendrickson, C. L.; Dalal, N. S.; Marshall, A. G. *J. Phys. Chem. A* **2002**, *106*, 10033–10036. (b) Khoury, J. T.; Rodriguez-Cruz, S. E.; Parks, J. H. *J. Am. Soc. Mass Spectrom.* **2002**, *13*, 696–708. (c) Friedrich, J.; Fu, J. M.; Hendrickson, C. L.; Marshall, A. G.; Wang, Y. S. *Rev. Sci. Instrum.* **2004**, *75*, 4511–4515. (d) Dashtiev, M.; Zenobi, R. *J. Am. Soc. Mass Spectrom.* **2006**, *17*, 855–858. (e) Sassin, N. A.; Everhart, S. C.; Dangi, B. B.; Ervin, K. M.; Cline, J. I. *J. Am. Soc. Mass Spectrom.* **2009**, *20*, 96–104. (f) Forbes, M. W.; Jockusch, R. A. *J. Am. Chem. Soc.* **2009**, *131*, 17038–17039. (g) Iavarone, A. T.; Patriksson, A.; van der Spoel, D.; Parks, J. H. *J. Am. Chem. Soc.* **2007**, *127*, 8606–8607. (h) Danell, A. S.; Parks, J. H. *Int. J. Mass Spectrom.* **2003**, *229*, 35–45.
(2) (a) Donald, W. A.; Williams, E. R. *J. Am. Soc. Mass Spectrom.* **2010**, *21*, 615–625. (b) Leib, R. D.; Donald, W. A.; Bush, M. F.; O'Brien, J. T.; Williams, E. R. *J. Am. Chem. Soc.* **2007**, *129*, 4894–4895.
(3) (a) Wong, R. L.; Paech, K.; Williams, E. R. *Int. J. Mass Spectrom.* **2004**, *232*, 59–66. (b) Donald, W. A.; Leib, R. D.; Demireva, M.; O'Brien, J. T.; Prell, J. S.; Williams, E. R. *J. Am. Chem. Soc.* **2009**, *131*, 13328–13337.
(4) Donald, W. A.; Williams, E R. *J. Phys. Chem. A* **2008**, *112*, 3515–3522.
(5) Melhuish, W. H. *J. Opt. Soc. Am.* **1964**, *54*, 183–186.
(6) Haugen, G. R.; Melhuish, W. H. *Trans. Faraday Soc.* **1964**, *60*, 386–394.

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