Angewandte Chemie
Eine Zeitschrift der Gesellschaft Deutscher Chemiker

Supporting Information

Multiple Ionization of Free Ubiquitin Molecular Ions in Extreme Ultraviolet Free-Electron Laser Pulses

Thomas Schlathölter, Geert Reitsma, Dmitrii Egorov, Olmo Gonzalez-Magaña, Sadia Bari, Leon Boschman, Erwin Bodewits, Kirsten Schnorr, Georg Schmid, Claus Dieter Schröter, Robert Moshammer, and Ronnie Hoekstra*

anie_201605335_sm_miscellaneous_information.pdf

Supporting information

Experimental Section

The experiments were performed by interfacing a radiofrequency (RF) ion trap equipped with a time-of-flight (TOF) mass spectrometer with the BL3 beamline of the Free electron LASer in Hamburg (FLASH). Figure 1a shows a sketch of the apparatus. Tenfold protonated ubiquitin ions $[ubi+10H]^{10+}$ and sixfold deprotonated $[ubi-6H]^{6-}$ were produced by means of electrospray ionization (ESI). For $[ubi+10H]^{10+}$ a 30 μM solution of bovine ubiquitin was prepared using a 1:1 mixture of methanol and water to which 1% acetic acid was added. $[ubi-6H]^{6-}$ were produced from a 40 μM solution of bovine ubiquitin in a 6:4 mixture of methanol and water, to which 0.3% ammonium hydroxide was added. The ions were phase space compressed and mass selected by a radiofrequency (RF) ion funnel, an RF octopole ion guide and a RF quadrupole mass filter. The mass selected ions were then collected in a 3 dimensional RF ion trap and cooled by collisions with He buffer gas for about 50 ms. The diameter of the trapped ion cloud for typical trap amplitude settings was about 400 μm. The low mass cutoff of the trap was about 75 u, i.e. photofragments with smaller masses could not be studied.

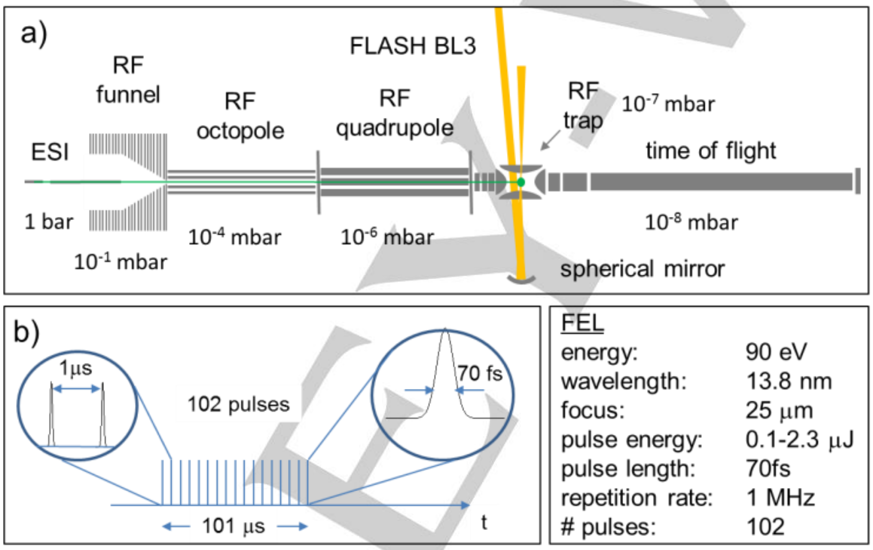

Figure 1 a) Sketch of the experimental setup. b) The pulse structure of the free electron laser. The FEL pulse parameters for the $[ubi+10H]^{10+}$ experiments are given as well.

For the FEL experiments on $[ubi+10H]^{10+}$, the photon beam from the FLASH BL3 beamline was back-reflected by means of a spherical multilayer mirror [1] and crossed the RF ion trap through two opposite bores in the ring electrode of the trap. The beam was focused into the trapped ion cloud in the center of the trap and the nominal focus diameter was 25 μm. FEL macropulses were produced at a repetition rate of 10 Hz and for photon energies of 90 eV (13.8 nm). A mechanical shutter was used to limit the beam exposure of the trap content to a single macropulse per experimental cycle. A macropulse consisted of 102 micropulses with fwhm length of 70±20 fs (see Figure 1b). A time separation of 1 μs between successive micropulses ensured a mixing of the trap between subsequent micropulses, implying only statistical chances for different micropulses acting on the very same protein. Pulse energies in the trap were varied between 0.1 μJ and 2.3 μJ. For the FEL experiments on $[ubi-6H]^{6-}$, the photon beam from the FLASH BL3 beamline was directly focused into the trap center. Here, 100 micropulses with with fwhm length of 90±20 fs at a pulse energy of 8.5 μJ were employed.

For the single photon absorption experiments, the MAXII I411 beamline (MAXlab, Lund, Sweden) and the BESSY II U49/2 PGM1 beamline (Helmholtz Zentrum Berlin, Germany) for 45 eV and for 90 eV, 288.5 eV and 310 eV, respectively. At both facilities, the photon beam of typically 10^{13} photons/s was focused into the trap center (focal diameters were below 100 μm) and photoexposure was controlled with a mechanical shutter.

The photoabsorption products were then extracted from the trap into a time-of-flight spectrometer, to obtain the resulting mass-spectra.

Modeling photoabsorption in the FEL pulse

For the $[ubi+10H]^{10+}$ data, the influence of FEL pulse energy was investigated. The pulse energy is defined as the number of photons per pulse, multiplied with the photon energy, i.e. 90 eV. The geometry of the beam crossing the ion cloud is sketched in Figure 2.

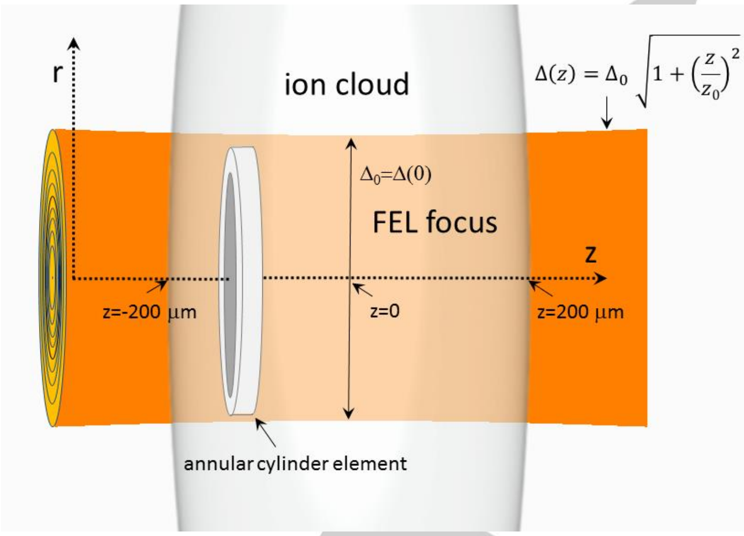

Figure 2 Sketch of the focus geometry crossing the ion cloud. z-axis and r-axis are not to scale.

The beam intensity is not constant over the cross section of the beam. We approximate the spatial beam intensity profile by a 2-dimensional Gaussian.

$$\boldsymbol{I(r,z) = I_0 \frac{4\,ln2}{\pi\Delta(z)^2} exp\left(-\frac{4ln2}{\Delta(z)^2} r^2\right)}$$

Here, $\boldsymbol{I_0}$is the total intensity and $\boldsymbol{r}$ is the distance from the beam axis, $\Delta(\boldsymbol{z})$ is the Gaussian width of the beam at a distance $\boldsymbol{z}$ from the focal point of the spherical mirror, i.e. from the trap center. $\Delta(\boldsymbol{z})$ is determined by the beam divergence, which is in turn given by the focal length of the mirror.

$$\Delta(z) = \Delta_0 \sqrt{1+\left(\frac{z}{z_0}\right)^2}$$

The Gaussian width of the beam in the focus (beam waist) is $\boldsymbol{\Delta_0}$= 25µm and the Rayleigh length (representing beam divergence) was $\boldsymbol{z_0}$=0.84 mm. As the beam crosses the ion cloud through the center, the length of the interaction volume can be approximated by the diameter of the ion cloud, which was determined as 400±50 µm. Computationally, we approximated the beam profile from $\boldsymbol{z = -200}$ µm to $\boldsymbol{z = 200}$ µm by 200 beam sections of constant diameter Δ and length 2 µm. Each of these sections was then divided into a series of annular cyclinders with radii starting from 0.1 µm and increasing in steps of 0.1 µm. The photoabsorption probability in each annular cylinder $\boldsymbol{P(z,r)}$ was then defined as the ratio of molecular photoabsorption cross section σ_{total}(90 eV) and annulus of the respective annular cylinder. Assuming a statistical distribution of independent photoabsorption processes, the probability for absorption of $\boldsymbol{n}$ photons $\boldsymbol{P_n(z,r)}$ by a single ubiquitin molecule can then be computed from the photon flux through the annular cylinder volume. Eventually, the charge state distribution was computed by summing up the contributions from all volume elements.

[1] Y. H. Jiang, A. Rudenko, O. Herrwerth, L. Foucar, M. Kurka, K. U. Kuehnel, M. Lezius, M. F. Kling, J. van Tilborg, A. Belkacem, K. Ueda, S. Duesterer, R. Treusch, C. D. Schroeter, R. Moshammer, J. Ullrich, *Phys. Rev. Lett.* 2010, *105*, 263002.