Supporting information for

Complete Protein Characterization Using Top-Down Mass Spectrometry and Ultraviolet Photodissociation

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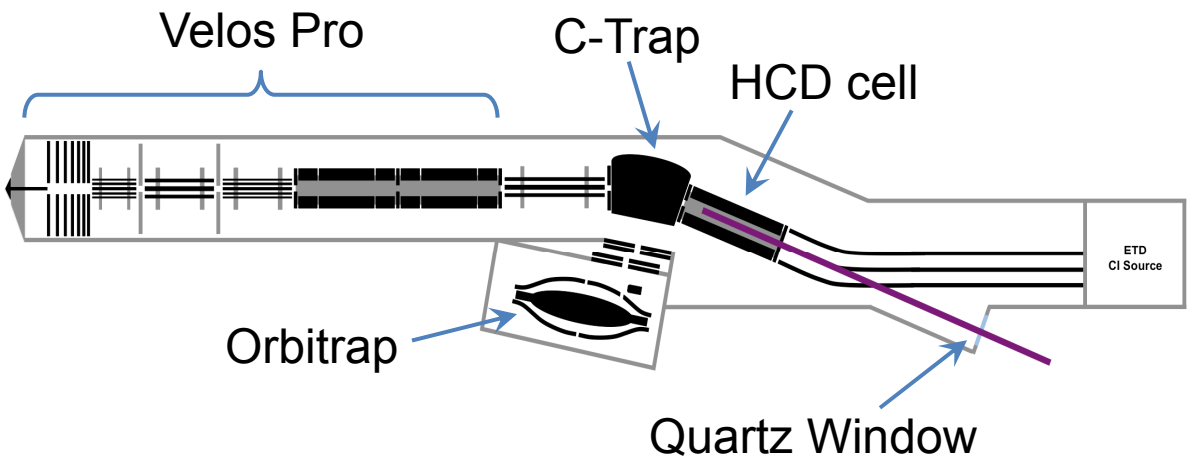

Figure S1. Schematic showing modifications made to the Thermo Scientific Orbitrap Elite mass spectrometer to allow photodissociation in the HCD cell.

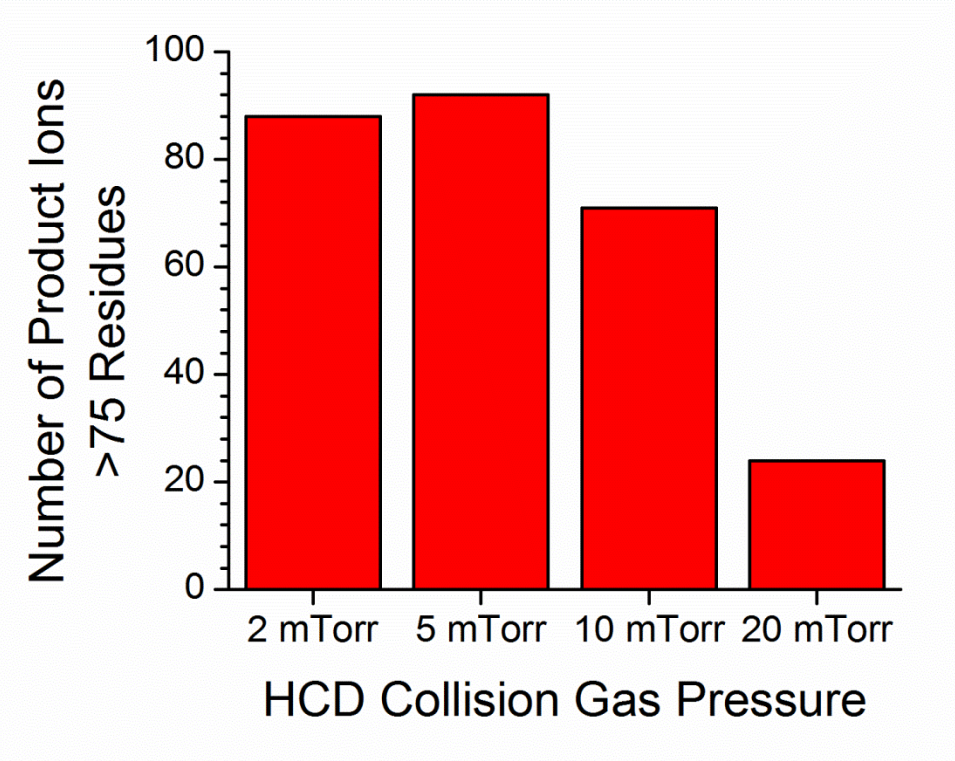

Figure S2. The number of UVPD product ions observed containing greater than 75 amino acids for the 20+ charge state of myoglobin as a function of HCD collision gas pressure. 10 mTorr is normal operating pressure and 5 mTorr was used for all other experiments.

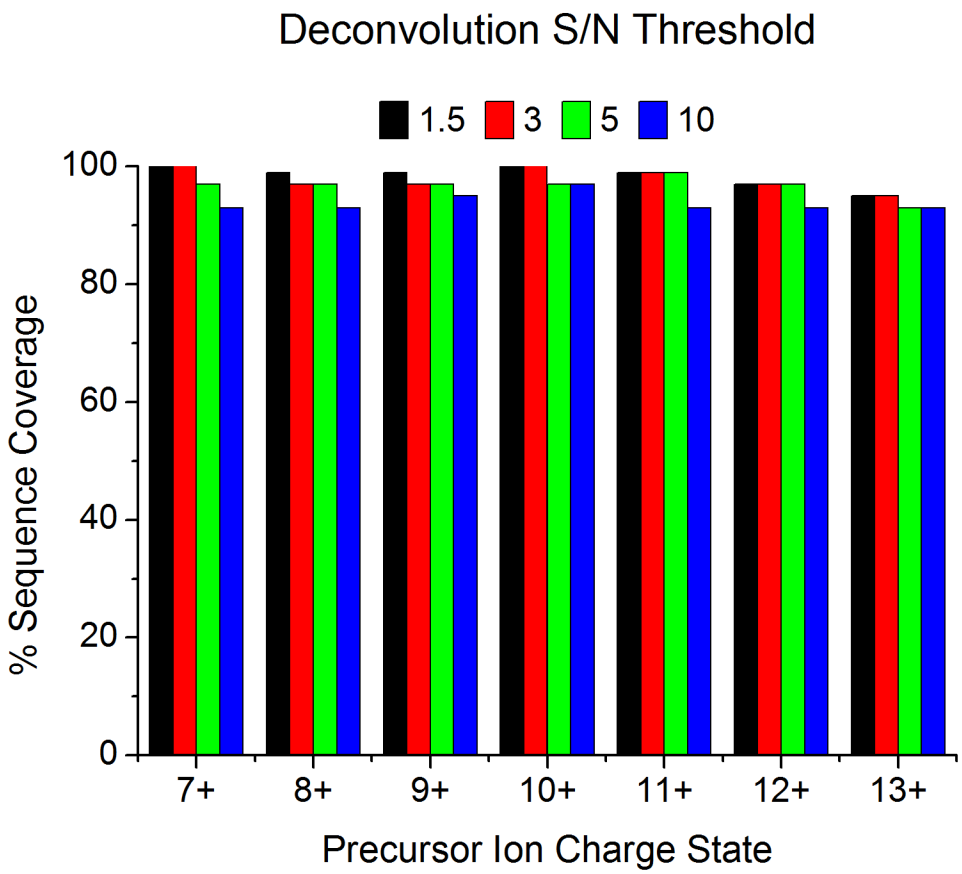

Figure S3. Graph of percent sequence coverage obtain for isolated precursor ion charge states of ubiquitin as a function of Xtract deconvolution S/N threshold.

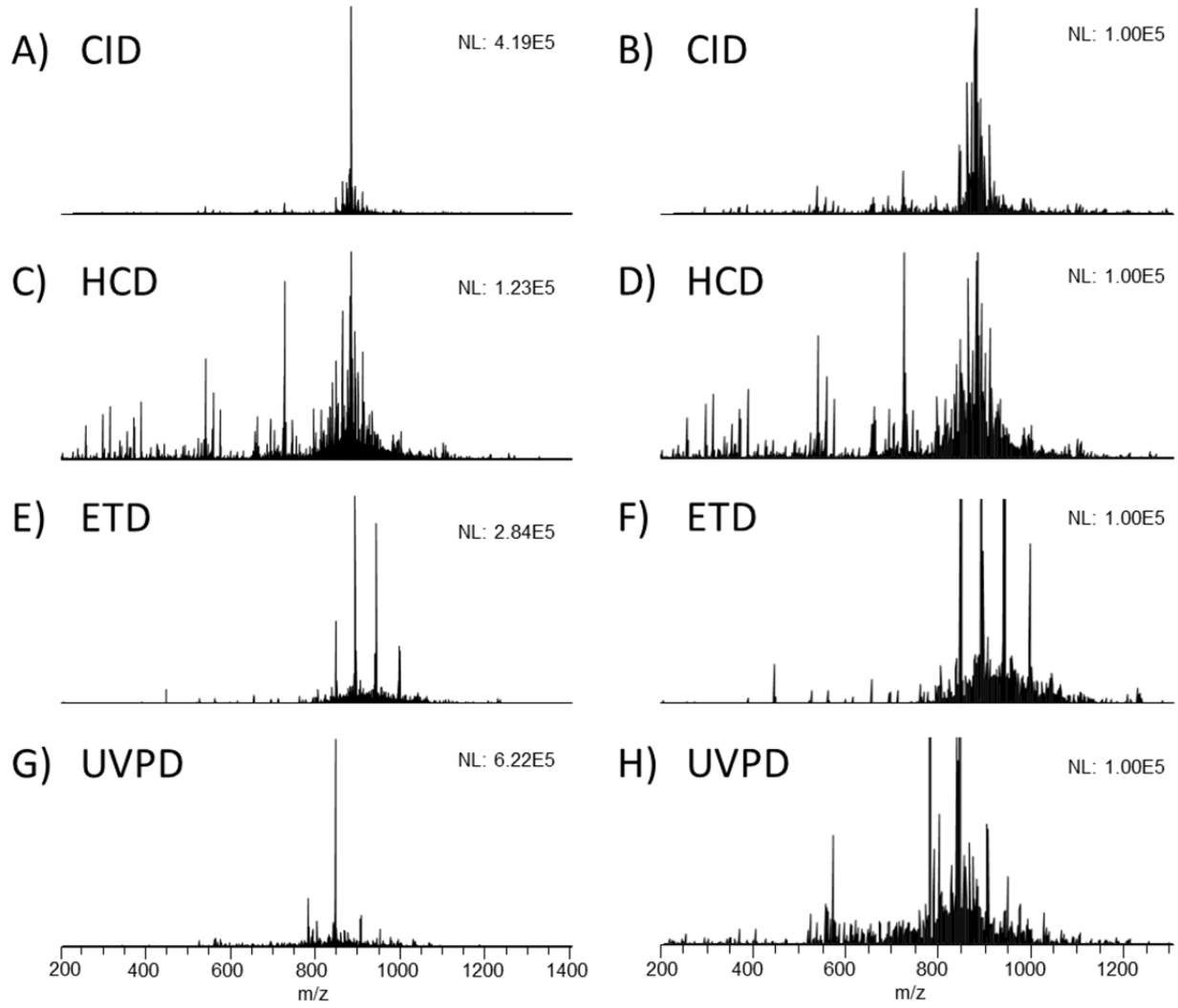

Figure S4. Full scale spectra produced by (A) CID, (C) HCD, (E) ETD and (G) UVPD of the 20+ charge state of myoglobin. The same (B) CID, (D) HCD, (F) ETD and (H) UVPD spectra of myoglobin scaled to the same absolute intensity (1.00E5).

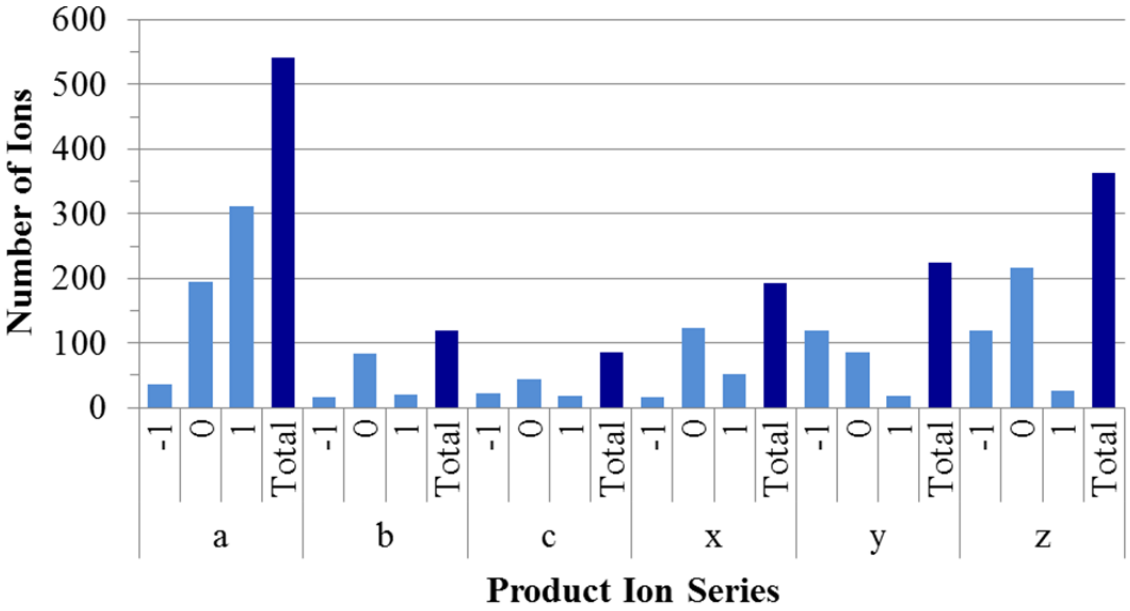

Figure S5. Distribution of UVPD product ions for myoglobin. Deconvoluted UVPD spectra were automatically compared to theoretical fragment ion masses using an altered version of ProSightPC created to annotate and aggregate single protein data with and without hydrogen atom shifts. Fragment ions were grouped based on the loss or gain of one hydrogen atom observed collectively for the 16+, 18+, 20+, 22+ and 24+ charge states of myoglobin. Zero represents the even electron species for all product ion series except for *z* ions where zero represents the radical species; -1 and +1 indicate the loss or gain of a hydrogen atom, respectively. Some amount of +/- 1 Da shifts are inevitable in automated interpretation of isotopic distributions in dense data sets like those created by UVPD; the amount of +/- 1 Da species for the *b*- and *c*-type ions reflect this approximate level. The +/- 1 Da shifts are enhanced for the *a, x, y* and *z* ions. Inspection of the analogous trends for other proteins in this study mirrored that found for myoglobin.

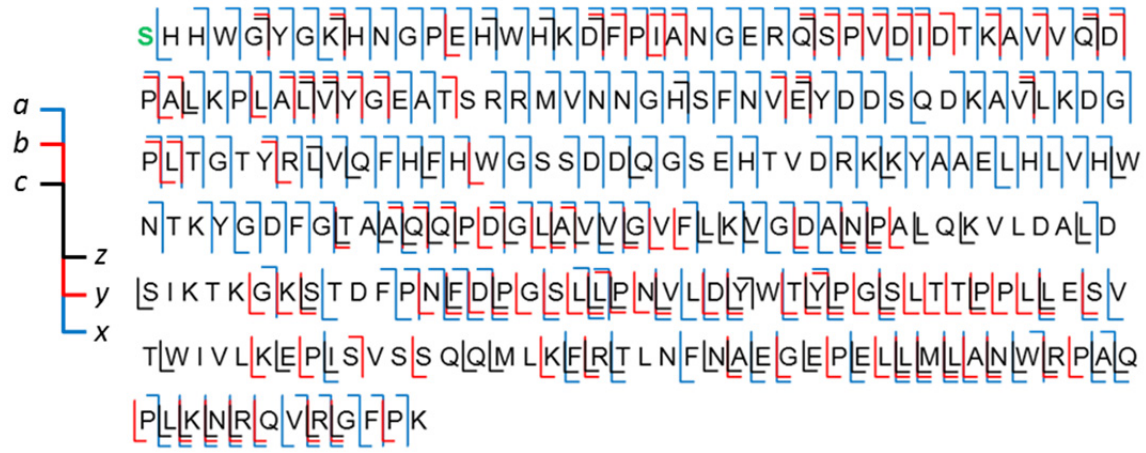

Figure S6. Fragment ion map of UVPD product ions from the 34+ charge state of bovine carbonic anhydrase II. Green "S" indicates N-terminal acetylation.