Electronic Supplementary Material (ESI) for Analyst.

SUPPORTING INFORMATION

Re-configurable, Multi-mode Contained-electrospray Ionization for Protein Folding and Unfolding on Milliseconds Time Scale

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Supporting information is summarized in the table below

1. Control experiment involving disulfide bond containing lysozyme protein. Manipulation of charge state of lysozyme was less effective because the conformation of lysozyme is stabilized by four disulfide bonds which are unaffected by pH changes during contained-electrospray ionization

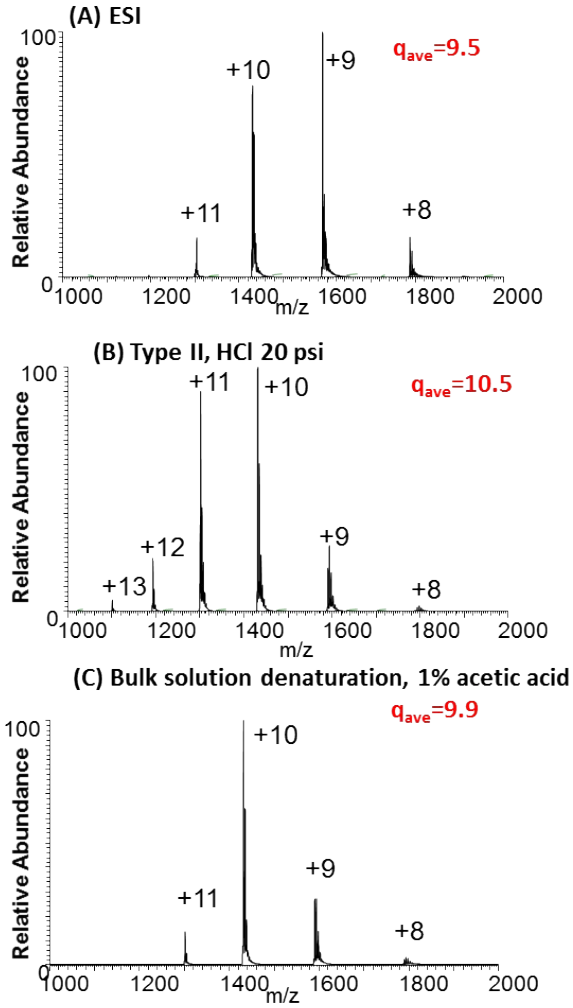

Figure S1. Positive ESI MS mass spectra for lysozyme solution (water) analyzed using (A) conventional ESI, in the absence of acid (B) contained-ESI, Type II mode, in the presence of HCl vapor and (C) conventional ESI, in the presence of 1% acetic acid.

2. Effect of protein pI on positive mode electrospray ionization. Ubiquitin, myoglobin, and cytochrome C were analyzed separately using traditional ESI, the absence of acidic. Cytochrome C with the highest pI (10.4) is the easiest to ionize and has the lowest noise. Ubiquitin with a pI of 5.2 is below the pH of the solution and has the highest level of noise

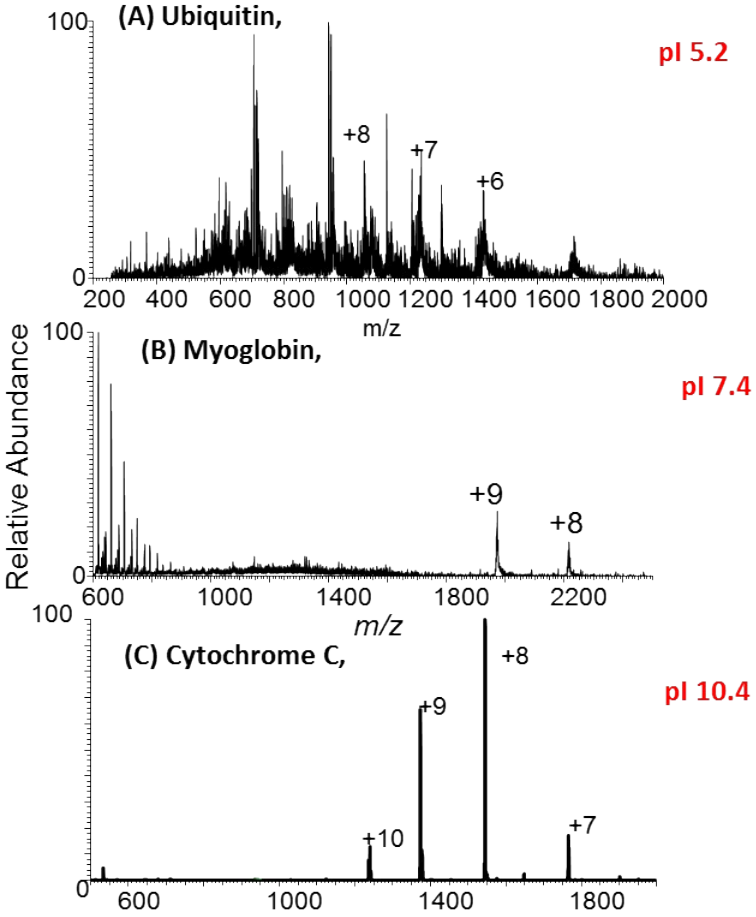

Figure S2. Positive ESI MS mass spectra for (A) ubiquitin, (B) myoglobin, and (C) cytochrome C prepared in 100% water. Spectra were recorded without addition of acid to protein solutions hence signal-to-noise is dictated by pIs of the proteins

3. Effect of chemical properties (pKa and vapor pressure) of modifying reagent on protein charge state. The chemical properties of acid reagents are shown below, which predicts HCl to be superior in protein denaturation because of its low pKa and high vapor pressure. The results in Figure S3 support this prediction

Reagent	pKa	Vapor pressure at 20°C (mbar)
HCl	-7	125
Acetic Acid	4.76	15.2
Formic Acid	3.75	44.8

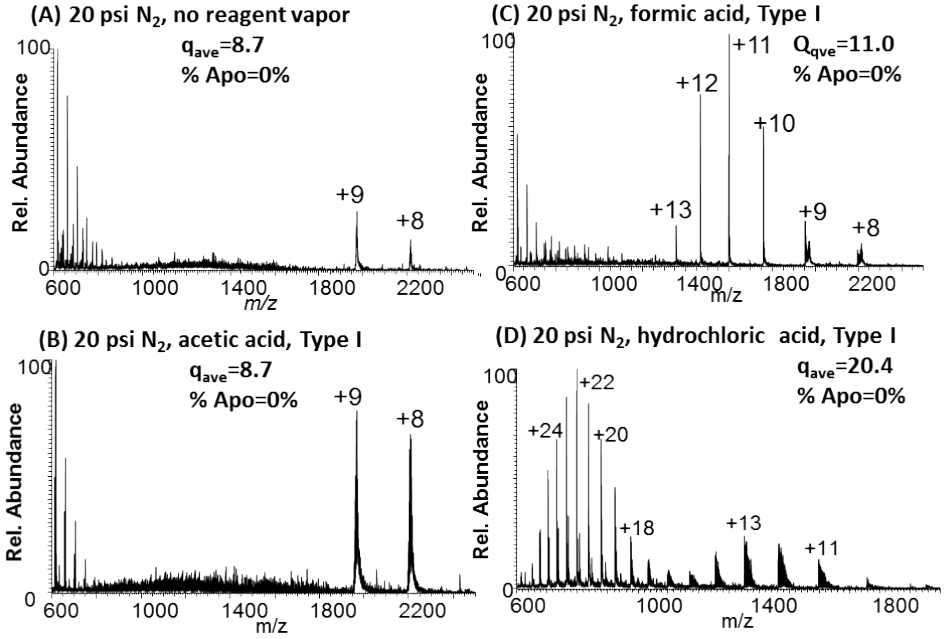

Figure S3. Myoglobin tested in type I mode using no reagent (A), acetic acid (B), hydrochloric acid (C), and formic acid (D)