Supporting Figures for

Digital Microfluidics for Microproteomic Analysis of Minute Mammalian Tissue Samples Enabled by Photocleavable Surfactant

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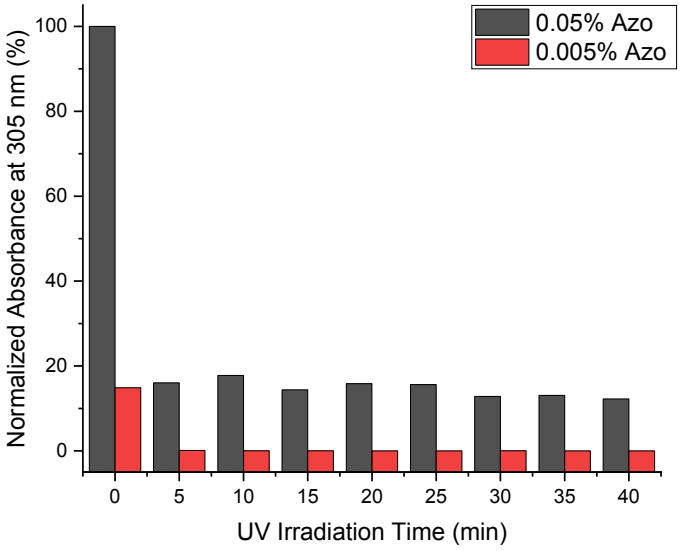

Figure S1. Plots of normalized absorbance at 305 nm for 0.05% (black) and 0.005% (red) wt/wt Azo in 50 mM TEAB as a function of UV irradiation time.

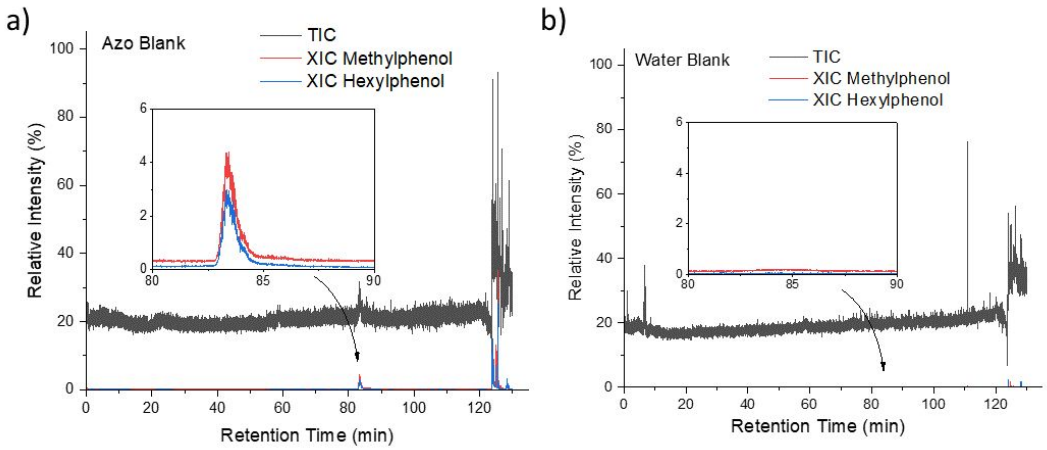

Figure S2. Base peak chromatograms from LC-MS/MS analysis of blanks containing (a) 0.005% Azo (after photodegradation) or (b) DI water. In each chromatogram, the total ion chromatogram (TIC) (black) is overlaid with extracted ion chromatograms

(XICs) of the Azo methylphenol fragment (red, m/z = 107.05±0.01) and hexylphenol fragment (blue, m/z = 177.13±0.01) normalized to $4x10^9$ intensity. The insets are magnified versions of the XIC data for the retention time window 80-90 min.

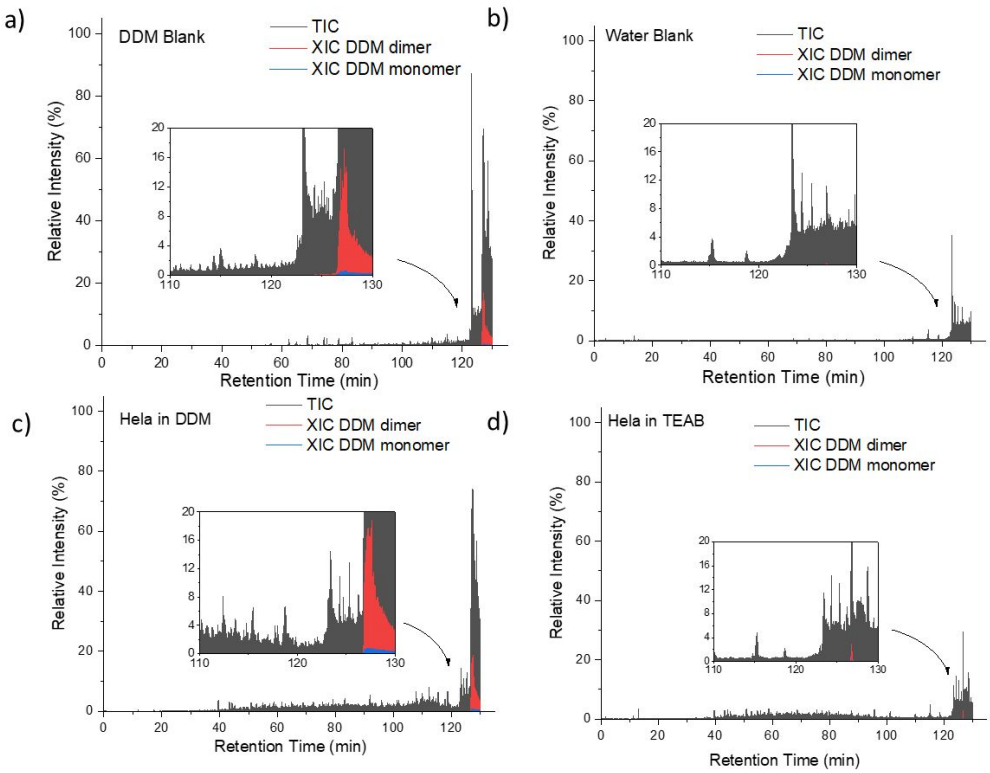

Figure S3. Base peak chromatograms from LC-MS/MS analysis of blanks containing (a) 0.005% DDM or (b) DI water, and 10 ng Hela standard digests in (c) 0.005% DDM or (d) 50 mM TEAB. In each chromatogram, the total ion chromatogram (TIC) (black) is overlaid with extracted ion chromatograms (XICs) of DDM dimer (red, m/z = 1021.61±0.01) and DDM monomer (blue, m/z = 511.31±0.01) normalized to 2.5×10^9 intensity. The insets are magnified versions of the XIC data for the retention time window 110-130 min.

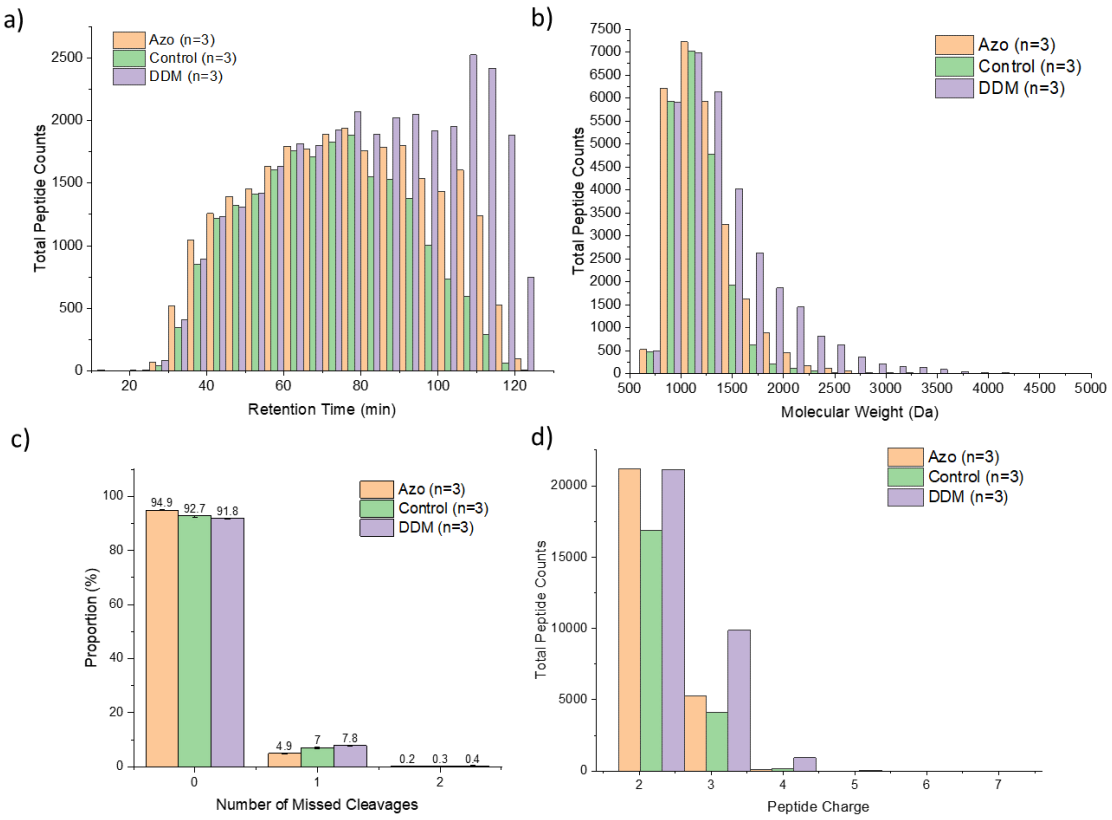

Figure S4. Analysis of peptides identified from 10 ng Hela digest standard in 0.005% Azo (orange), 50 mM TEAB as control (green), or 0.005% DDM (purple), including plots of total peptide numbers as a function of a) retention time, and b) molecular weight, c) proportions of peptides with missed cleavages (from zero cleavages on the left to two cleavages on the right), and d) total peptide numbers as a function of charge. The small error bars in c) are ± 1 st. dev. for n = 3 replicates per condition.

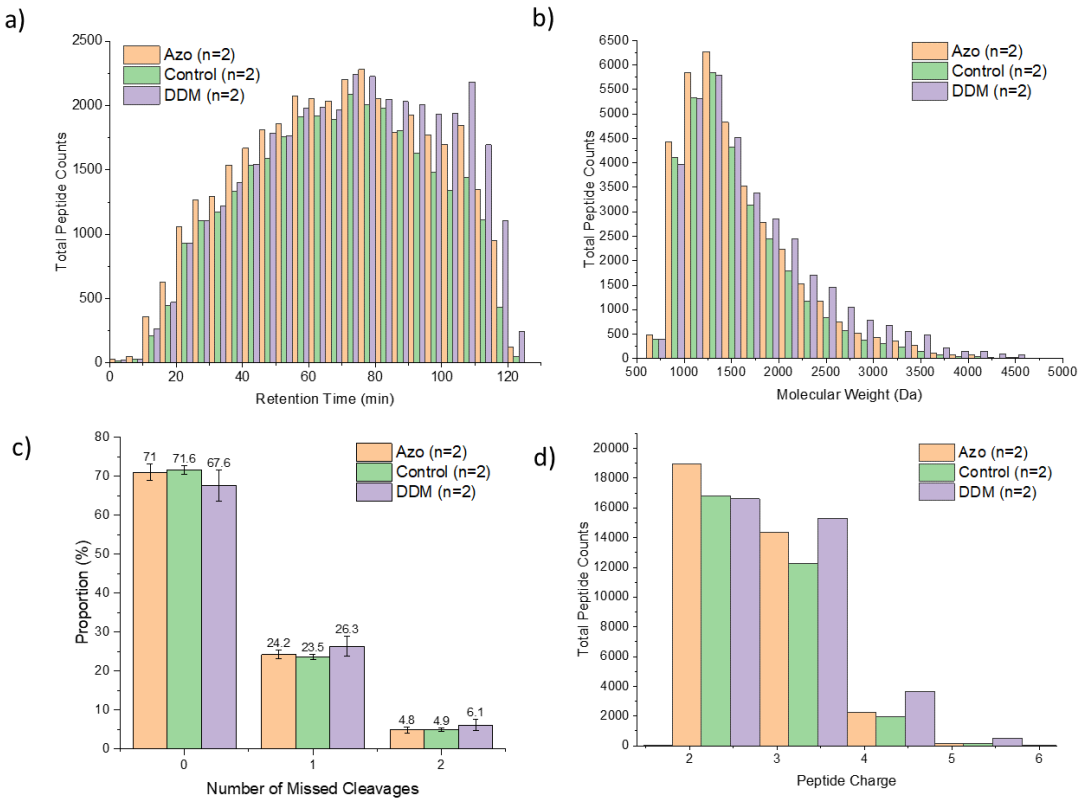

Figure S5. Analysis of peptides identified from digests of 100 MCF-7 cells in 0.005% Azo (orange), 50 mM TEAB as control (green), or 0.005% DDM (purple), including plots of total peptides as a function of a) retention time, and b) molecular weight, c) proportions of peptides with missed cleavages (from zero cleavages on the left to 2 cleavages on the right), and d) total peptides as a function of charge. Error bars for c) are ± 1 st. dev. for n = 2 replicates per condition

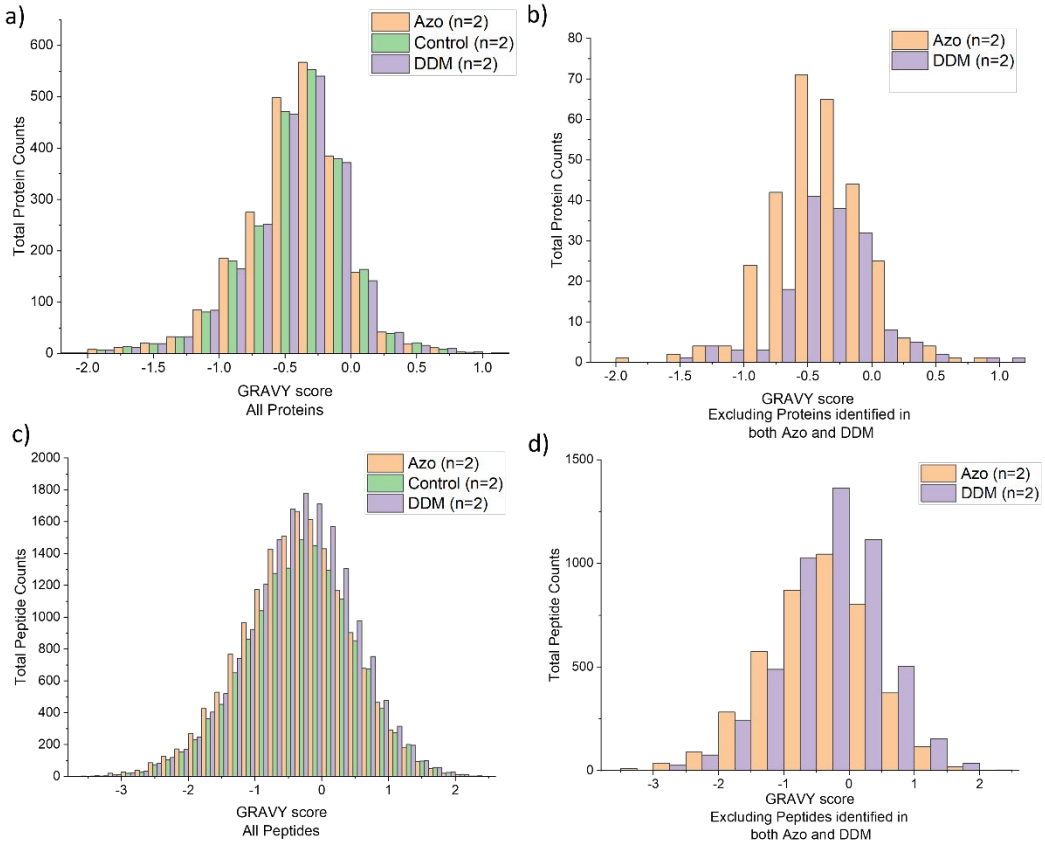

Figure S6. Hydrophobicity of proteins and peptides identified from digests of 100 MCF-7 cells in 0.005% Azo (orange), 50 mM TEAB as control (green), or 0.005% DDM (purple). Total protein counts as a function of GRAVY score for a) all proteins, b) all proteins excluding proteins identified in both Azo- and DDM-containing samples, and total peptide counts as a function of GRAVY score for c) all peptides, and d) all peptides excluding peptides identified in both Azo- and DDM-containing samples.

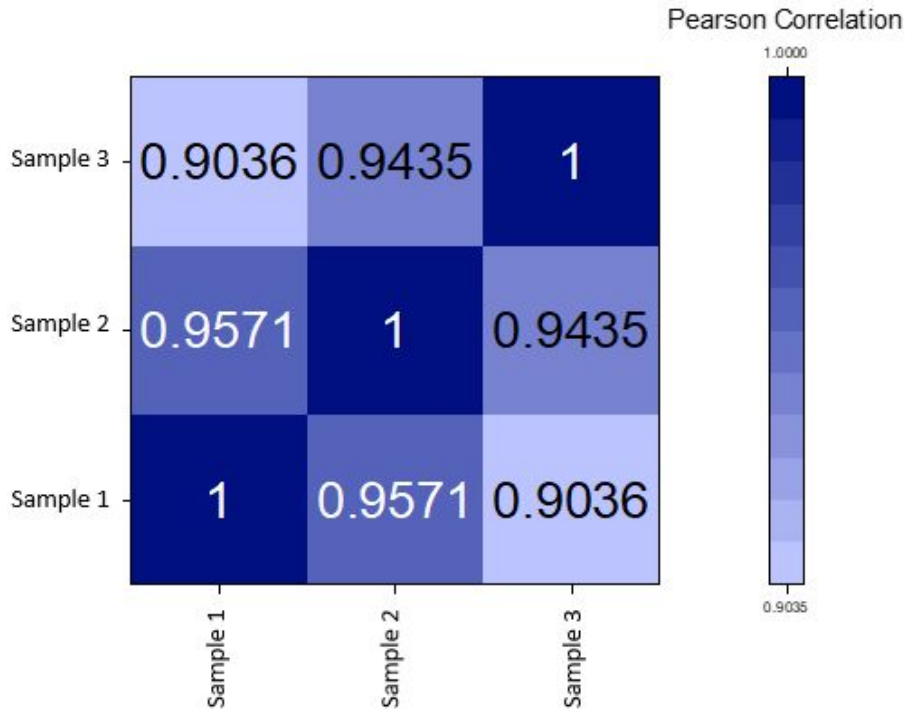

Figure S7. Pearson correlation values for quantified proteins identified from murine tissue biopsy samples (n=3) processed using Azo on DMF.