Electronic Supplementary Material (ESI) for Nanoscale

Supporting Information

Monitoring Enzymatic Degradation of Pericellular Matrices through SERS Stamping

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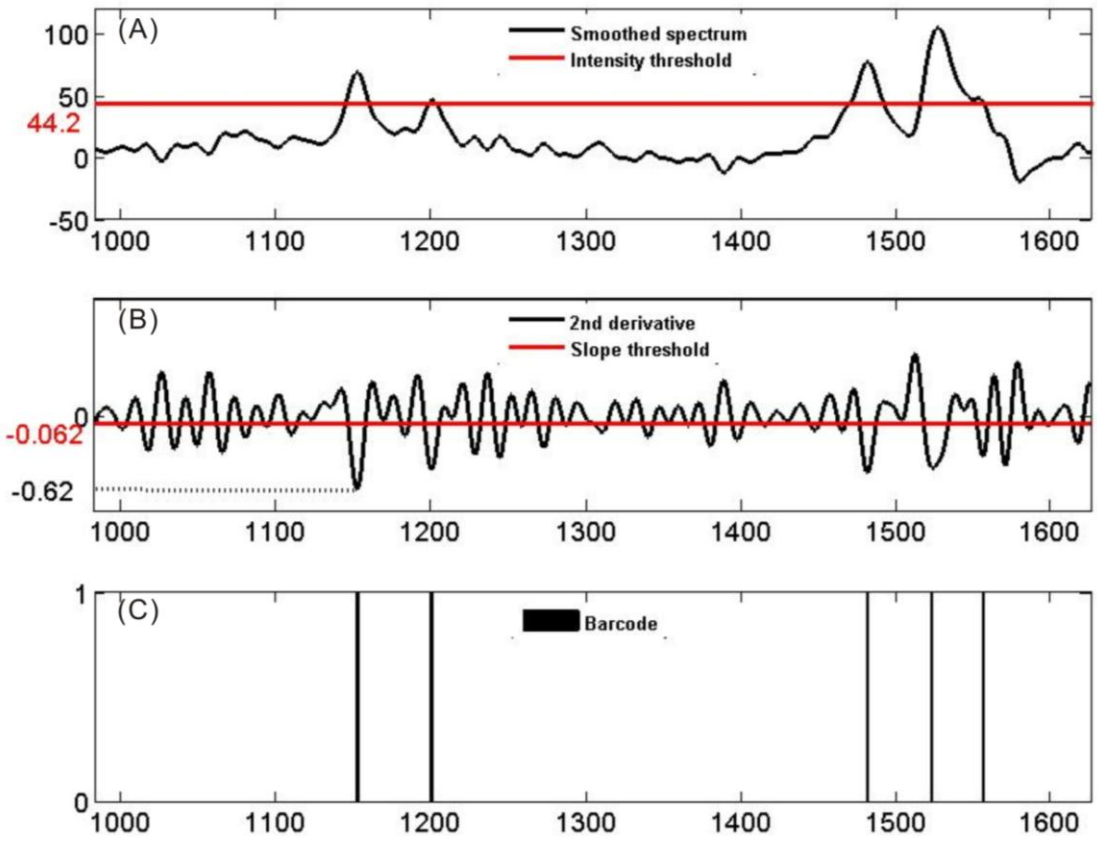

Figure S1. Conversion from the SERS spectrum to the barcode. (A). The first selection criterion is based on intensity. Only features in the SERS spectrum (black) above the intensity threshold (red), calculated as three times the average signal-to-noise of the backgrounds, are considered. (B). The second selection criterion is based on the second derivative (black), features with a negative second derivative of at least 10% of the global minimum of the second derivative (red) are considered. (C). Spectral peaks that fulfill both the intensity and second derivative selection criteria are included in the barcode.

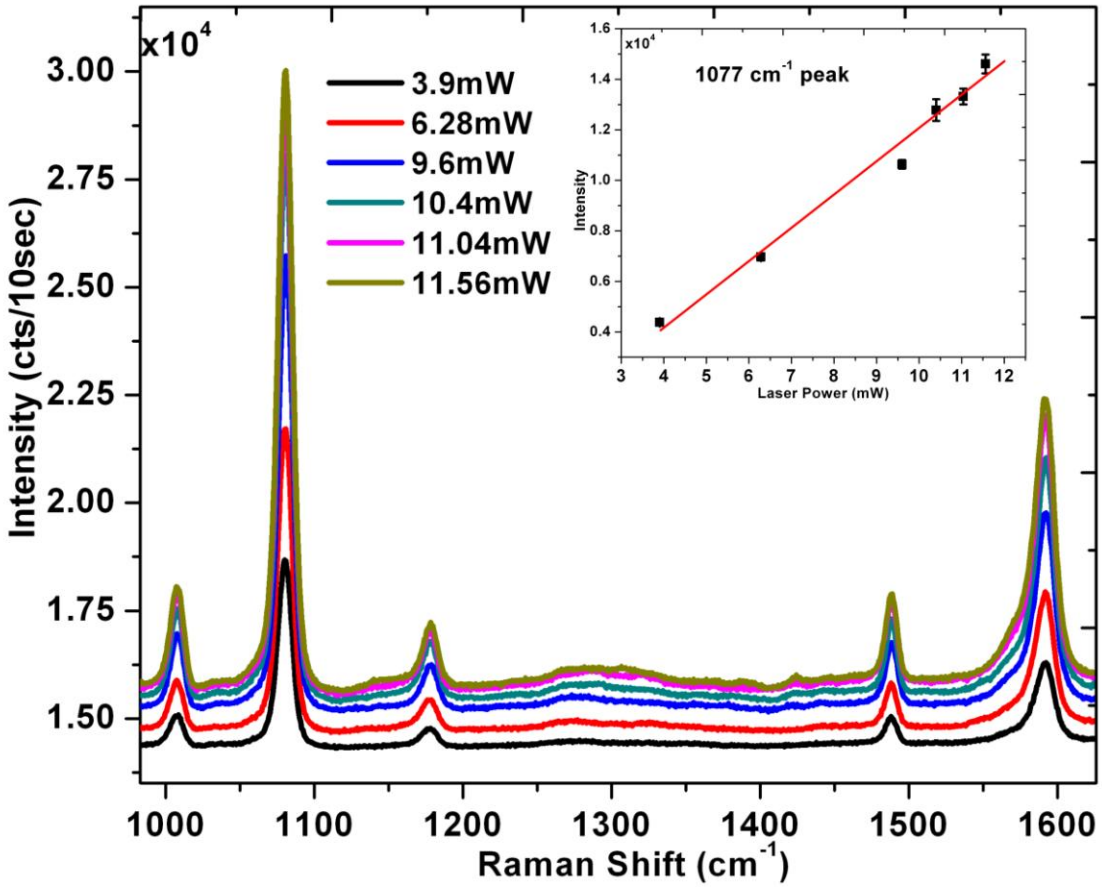

Figure S2. SERS signal of pMA as a function of laser excitation power.

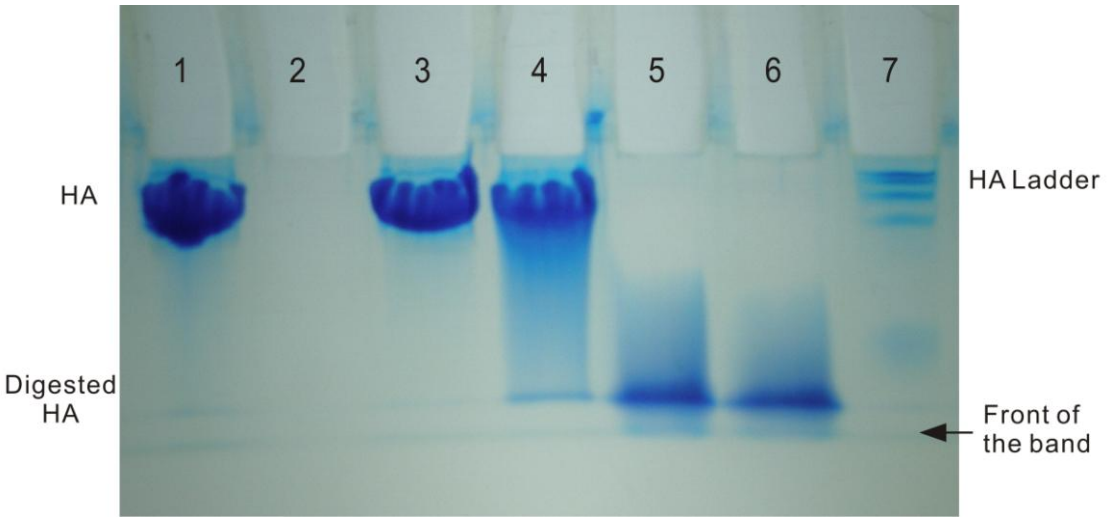

Figure S3. Gel electrophoresis (polyacrylamide gel) of synthesized HA under different enzymatic digestion conditions. Lane 1: 1 mg/mL HA (~150kDa); Lane 2: 1 mg/mL hyaluronidase (~55 kDa); Lane 3: HA pre-incubated with Hanks' buffer (pH 6) for 10 min; Lane 4: HA pre-incubated with 10 U/mL hyaluronidase for 10 min; Lane 5: HA pre-incubated with 600 U/mL hyaluronidase in Hanks' buffer (pH 6) for 30 min; Lane 6: HA pre-incubated with 600 U/mL hyaluronidase in PBS buffer (pH 6) for 30 min; Lane 7: HA ladder (ranging from ~25kDa to ~500kDa, got from *Sigma S0576*).

The samples were run on a precast polyacrylamide gel (*Thermo Scientific #25201*) at 120 V for 45 min in 0.5x TBE buffer. The pH of the running buffer was pre-adjusted to pH = 8.4 with HEPES. The gel was cleaned in water and then stained with Stains-all (*Sigma E9379*) overnight and washed again before imaging.

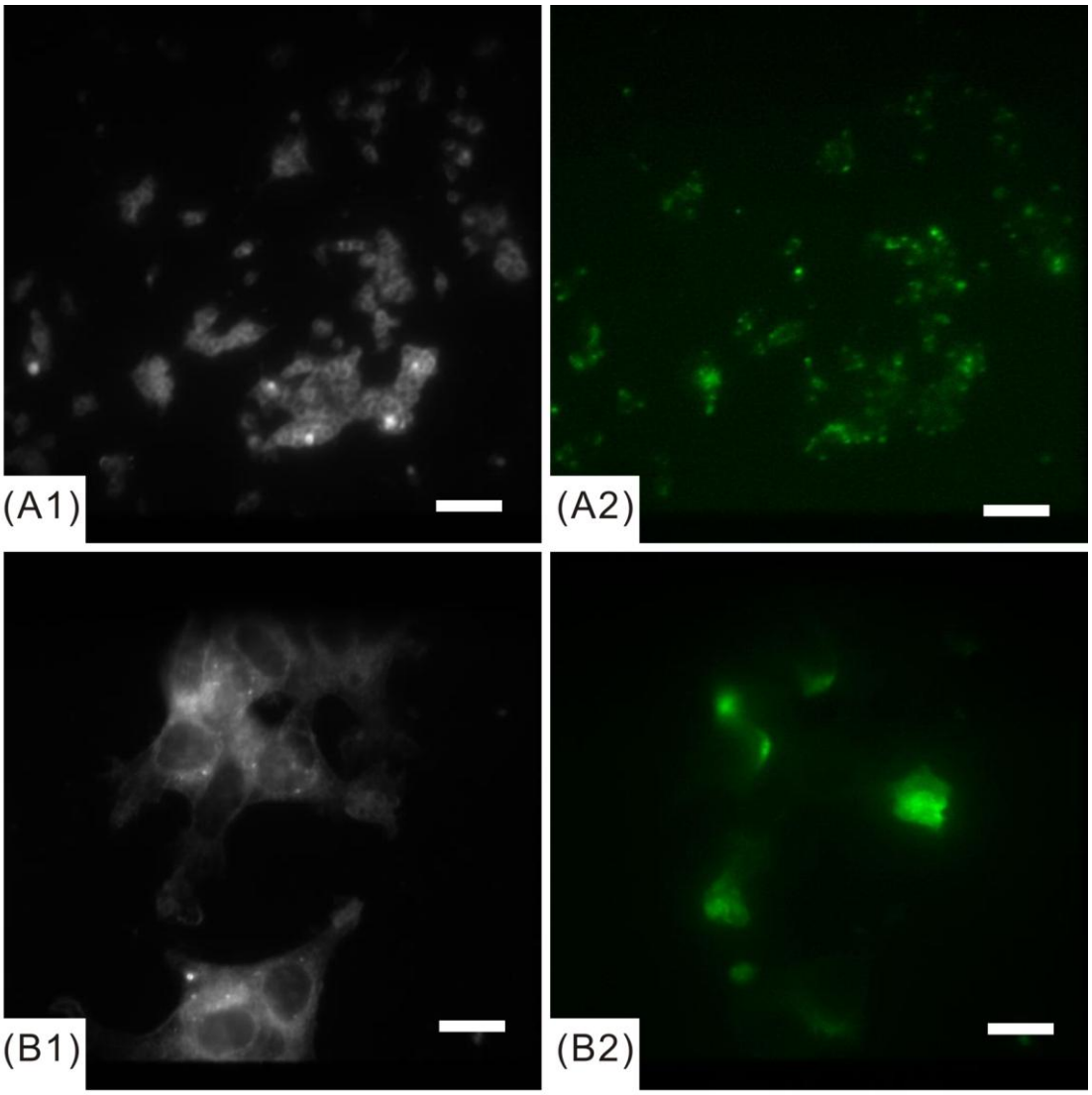

Figure S4 (A1)&(B1) Dark-field images of multiple MCF7 cells observed at 10X and 60X magnification. (A2)&(B2) Corresponding fluorescent images with antibody staining for HA. Scale bars denote 50 μm in (A) and 10 μm in (B).

The labeling was performed with an anti-HA primary antibody (*Abcam ab*53842) and a Dylight 488 conjugated IgG secondary antibody (*Abcam ab96943*).

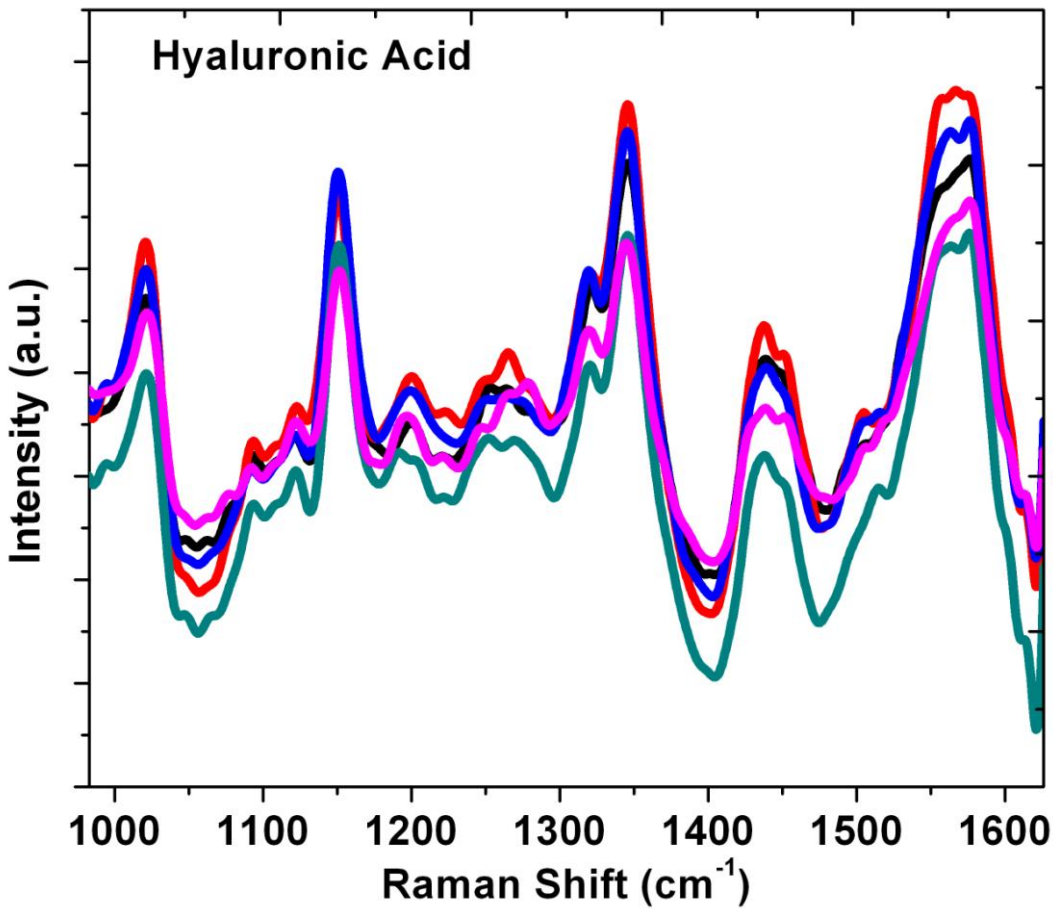

Figure S5. SERS spectra of synthetic HA measured with SERS stamps

Table S1. Tentative assignments of SERS features in Figure 7.

Raman Shift (cm^{-1})	Band Assignment[§]
1040	**C-C and C-O stretching (c)**
1105	**C-N (p)**
1145	**Oxygen bridge, C-C and C-O stretching (c)**
1205-1225	**Amide III rand coils (p)**
1330	**Amide III, C-H bending (c)**
1370	**COO^- stretching (c)**
1430	**CH_2, CH_3 bend (l) (p)**
1470	**CH_2, CH_3 bend (p) (l) (c)**
1500	**A, C, G (n)**
1525	**A, C, G (n)**
1620	**C=C (p) (l)**
999	**Amide III (p)**
1026	**C-H in plane stretching (p)**
1180	**Tyr, Phe (p)**
1215	**C-C_6H_5 stretching (p)**
1245-1265	**Amide III β sheet (p), Amide III (l), =CH (l)**
1560	**Amide II (p)**
1586	**Phe, Tyr (p)**

[§] Abbreviations: (p) protein, (l) lipid, (c) carbohydrates, (n) nucleotide.

Assignments based on the tables compiled in Refs.[1-8]:

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(4) Swain, R. J.; Kemp, S. J.; Goldstraw, P.; Tetley, T. D.; Steyens, M. M. *Biophys. J.* **2010**, *98*, 1703-1711.

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(8) Ochsenkuhn, M. A.; Jess, P. R. T.; Stoquert, H.; Dholakia, K.; Campbell, C. J. *ACS Nano* **2009**, *3*, 3613-3621.