Supporting Information

SERS-ELISA using silica-encapsulated Au core-satellite nanotags for sensitive detection of SARS-CoV-2

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Fig. S1. Schematic illustration of the synthesis of antibody-conjugated (a) HAuNP@SiO$_2$ and (b) AuNP@SiO$_2$ SERS nanotags.
Fig. S2. Characterization of CS@SiO$_2$ (top), HAuNP@SiO$_2$ (middle), and AuNP@SiO$_2$ (bottom) SERS nanotags. (a) Transmission electron microscopy (TEM) images of CS@SiO$_2$ (diameters of the core and satellite AuNPs = 75 and 32 nm, respectively; silica shell thickness = 21 nm), HAuNP@SiO$_2$ (diameter of the HauNPs = 58 nm; silica shell thickness = 18 nm), and AuNPs@SiO$_2$ (diameter of the AuNPs = 75 nm; silica shell thickness = 18 nm). (b) Corresponding UV/Vis spectra, (c) DLS distributions, and (d) Raman spectra. Here, BDMT was used as a Raman reporter for CS@SiO$_2$, and BT was used as a Raman reporter for HAuNP@SiO$_2$ and AuNP@SiO$_2$. The SERS peak intensity variations at 1597 cm$^{-1}$ (BDMT) and 1574 cm$^{-1}$ (BT), indicated with asterisks, were used for the SERS-ELISA quantitative evaluations of SARS-CoV-2.
Fig. S3. Changes in (a) UV/Vis absorption spectra, (b) size distributions, and (c) zeta potentials before and after antibody conjugation for CS@SiO$_2$ (top), HAuNP@SiO$_2$ (middle), and AuNP@SiO$_2$ (bottom).
Fig. S4. (a) Molecular structures of Raman reporter molecules and (b) corresponding Raman spectra of four different Raman reporters from CS@SiO$_2$. 
Fig. S5. SARS-CoV-2 assay results based on the absorbance measurements using (a) 96-well plastic plate and (b) 384-well glass plate. Assays were performed in the 0–1,000 PFU mL⁻¹ SARS-CoV-2 concentration range, and calibration curves were determined from their absorbance measurement data. Error bars indicate the standard deviations from five measurements.
Fig. S6. (a) Raman mapping images of the immunocomplexes with CS@SiO₂ nanotags for 1,000 PFU mL⁻¹ SARS-CoV-2, measured in pixel sizes of a 20 μm × 20 μm, in five randomly selected 400 μm × 400 μm regions, denoted as I–V. (b) Average Raman spectra measured for 400 pixels of area III to evaluate the pixel-to-pixel fluctuations. (c) Raman mapping images, measured for the five regions (I–V), to evaluate area-to-area fluctuations in Raman signal intensity. Raman mapping was performed using the characteristic Raman peak intensity variations of BDMT at 1597 cm⁻¹. Uniform yellow color distributions in the I–V areas indicate superior reproducibility in any area among the 384 wells. (d) Histograms of the Raman peak intensities for 400 pixels measured for the five regions, i.e., I–V.
Fig. S7. Determination of the optimal concentrations of (a) SERS nanotag and (b) BSA. When changing the concentration of the SERS nanotag from 10 to 60 pM under a SARS-CoV-2 concentration of 0 PFU mL⁻¹ (off) and 1000 PFU mL⁻¹ (on), the optimum concentration, for which the corresponding Raman intensity difference for the off and on conditions is the most significant, was determined to be 40 pM (red color), as shown in (a). The optimum BSA concentration condition was determined to be 2% (red color), as shown in (b).
Fig. S8. (a) Raman mapping images of immunocomplexes for 1,000 PFU mL⁻¹ SARS-CoV-2, measured in pixel sizes of 20 μm × 20 μm in randomly selected 400 μm × 400 μm regions in three different wells denoted as I–III. (b) Raman mapping images measured for the three regions in three wells to evaluate well-to-well fluctuations in Raman signal intensity. Raman mapping was performed using the characteristic Raman peak intensity variations of BDMT at 1597 cm⁻¹. (c) Average Raman spectra measured for 400 pixels in wells I–III. (d) Histograms of average Raman peak intensities for 400 pixels measured for the regions in wells (I–III).
Fig. S9. (a) Measurement of the Raman mapping images using CS@SiO$_2$ SERS nanotags for the 0–1000 PFU mL$^{-1}$ SARS-CoV-2 concentration range on a 384-well plate. (b) Raman mapping images measured for three well lines in the same SARS-CoV-2 concentration conditions. The color-decoding bar on the right shows that the Raman intensity gradually increases from dark red to bright yellow. As the concentration of SARS-CoV-2 increases, the Raman peak intensity increases consistently in the Raman mapping images.
Fig. S10. (a) Average Raman spectra and (b) corresponding histograms of average Raman peak (1597 cm$^{-1}$) intensities of 400 pixels of four respiratory viruses.