Supporting Information

Bimetallic Au-Ag on Patterned Substrate derived from Discarded Blu-ray discs: Simple, Inexpensive, Stable and Reproducible SERS Substrates

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Optimization of r-AgBD-ROM substrate fabrication

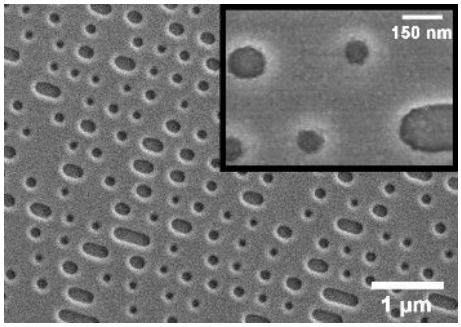

Figure S1. FE-SEM images of AgBD-ROM. Insert is an enlarged image.

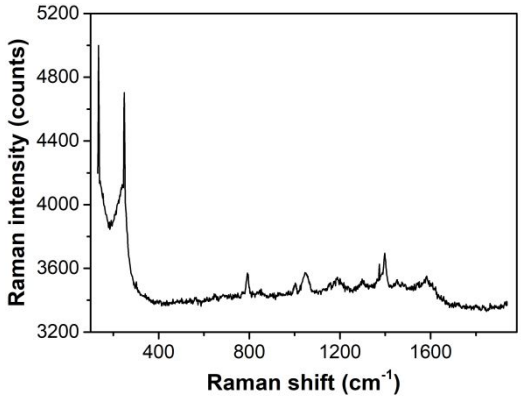

Figure S2. Raman spectra background of r-AgBD-ROM.

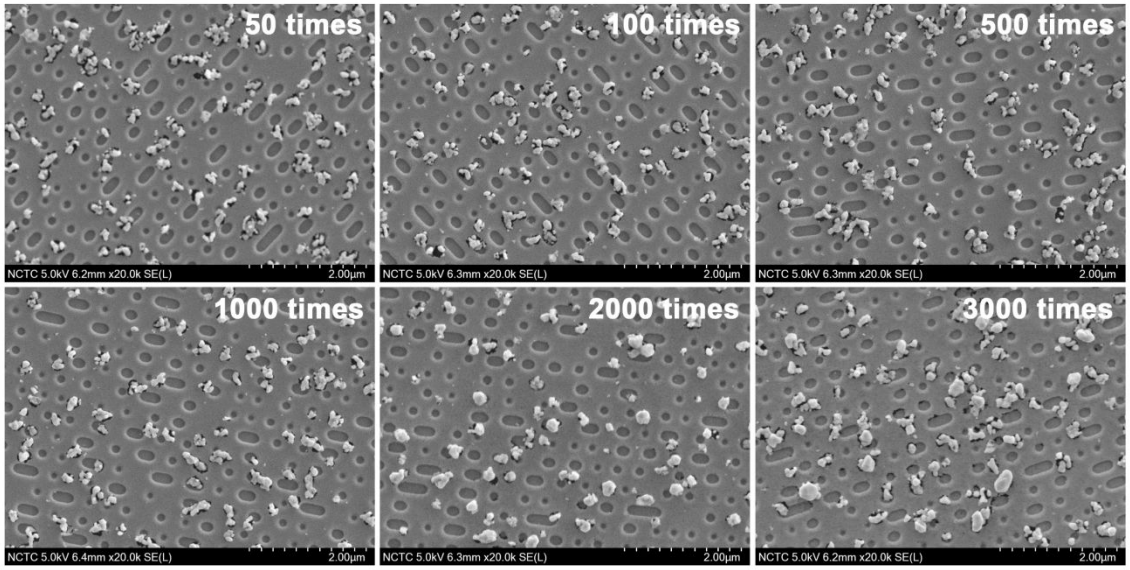

Figure S3. FE-SEM images of r-AgBD-ROM fabricated under different the number of step-scan.

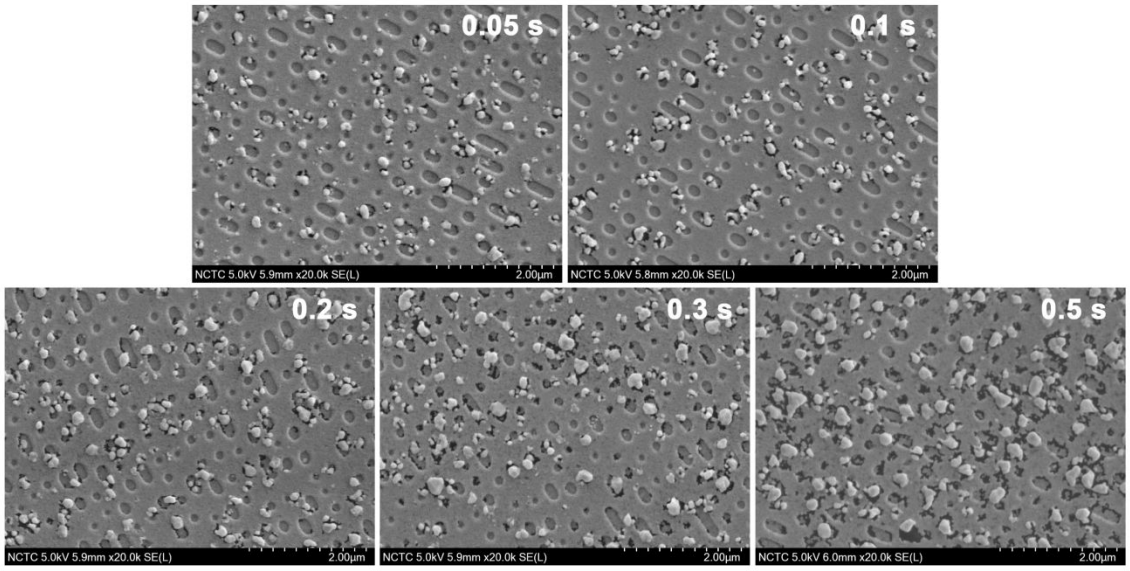

Figure S4. FE-SEM images of r-AgBD-ROM fabricated under different duration time.

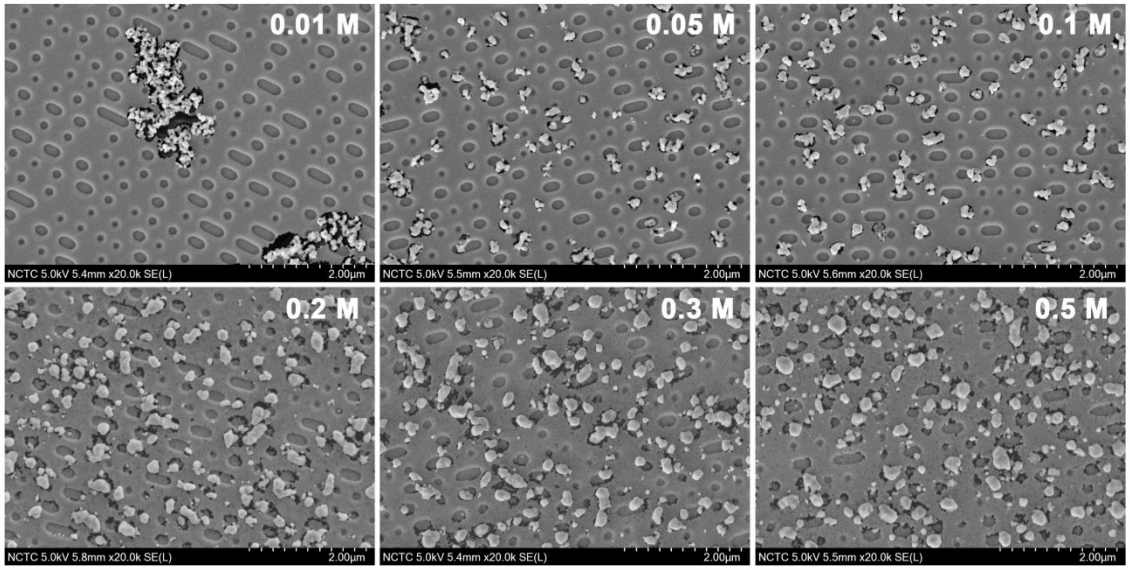

Figure S5. FE-SEM images of r-AgBD-ROM fabricated under different KCl concentrations.

Optimization of Au-r-AgBD-ROM substrate fabrication

The Au-r-AgBD-ROM substrates fabricated under 0, 0.1 and 0.5 mM $HAuCl_4$ solution were chosen as representative substrates to investigate particle size and size distribution of AgNPs and Au-AgNPs. The histogram of particle size distribution evaluted from 78-91 particles in SEM images (3x3 μm^2) using imageJ software is shown in Figure S6. The average particle size and polydispersity index (PDI) were obtained by Gaussian fitting. The average particle size were 50 ± 14.3, 66.12 ± 30.12, and 146.62 ± 81.94 nm for Au-r-AgBD-ROM fabricated under 0, 0.1 and 0.5 mM $HAuCl_4$ solution, respectively. The PDI values were 0.29, 0.46, and 0.56 for Au-r-AgBD-ROM fabricated under 0, 0.1 and 0.5 mM $HAuCl_4$ solution, respectively.

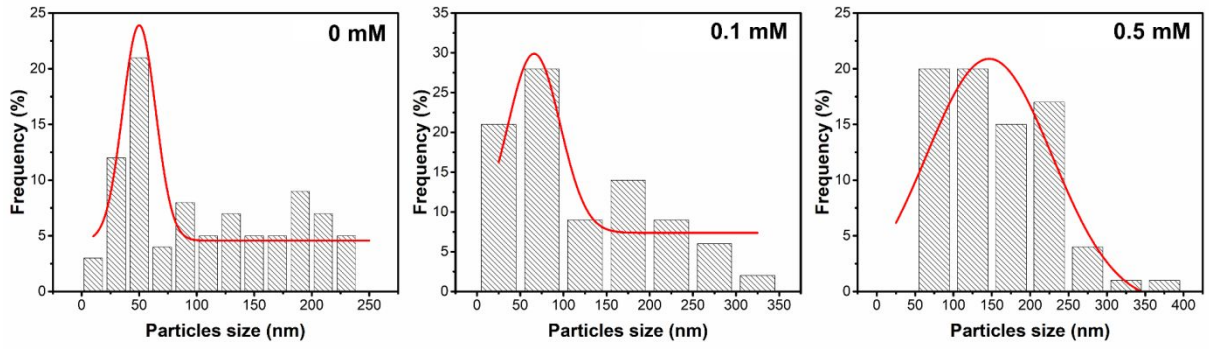

Figure S6. Histogram of NPs size distributions on the Au-r-AgBD-ROM fabricated under 0, 0.1 and 0.5 mM $HAuCl_4$ solution. The red curves represent a Gaussian fitting.

Elemental composition analysis of SERS substrates during EC treatment and Au coating methods

Table S1 The atomic percentage of elements on the as-prepared substrates obtained from XPS measurements

Peaks	**Atomic compositions (%)**

	AgBD-ROM	r-AgBD-ROM	Au-r-AgBD-ROM
$Ag3d_{3/2}$ (metal)	16.91	3.42	15.78
$Ag3d_{5/2}$ (metal)	24.98	5.04	23.32
$Ag3d_{3/2}$ (Ag-O-Ag)	-	6.86	4.56
$Ag3d_{5/2}$ (Ag-O-Ag)	-	10.13	6.74
O1s (C-O)	7.13	9.53	5.07
O1s (Ag-O-Ag/O-H/Au-O)	10.64	7.46	2.19
O1s (Absorb with H_2O)	-	1.54	2.19
C1s (C-C)	22.82	28.06	22.18
C1s (C-O)	8.34	17.37	8.25
C1s (C=O)	5.59	5.99	3.07
C1s (O=C-O)	3.59	2.43	2.25
C1s (shake-up)	-	2.17	-
$Au4f_{5/2}$ (metal)	-	-	1.51
$Au4f_{7/2}$ (metal)	-	-	2.01
$Au4f_{5/2}$ (Au^+)	-	-	0.38
$Au4f_{7/2}$ (Au^+)	-	-	0.50

Possible model for SERS enhancement phenomenon

Table S2 Surface roughness (RMS) and surface area of AgBD-ROM and r-AgBD-ROM obtained from AFM measurement.

substrate	**RMS**	**Surface area (nm^2)**
AgBD-ROM	2.232	25.008

r-AgBD-ROM	6.580	25.008

Surface area reported in Table S2 were calculated from S ratio, which represents the ratio between the real interfacial area and scan area (5 × 5 μm^2).

SERS substrate performance

LOD calculation

The equation used to estimate the limit of detection (LOD) in this study was displayed below.[1]

$$\boldsymbol{LOD} = \frac{I_{avg(blank)} + \mathbf{1.645}(SD_{blank} + SD_{low\ conc}) - \boldsymbol{b}}{\boldsymbol{a}} \quad \text{(S1)}$$

Where $I_{avg(blank)}$ and SD_{blank} are the average intensity and the standard deviation of a blank solution (n=10), respectively. $SD_{low\ conc}$ is the standard deviation of the low concentration sample. $\boldsymbol{a}$ and $\boldsymbol{b}$ are the slope and y-intercept of the sensitivity calibration curve, respectively.

EF calculation

The SERS enhancement factors (EFs) of our SERS substrates were evaluated using methylene blue (MB) as a Raman probe. SERS substrates (r-AgBD-ROM and Au-r-AgBD-ROM) and reference substrate (AgBD-ROM) in the dimension of 1 × 1 cm^2 were soaked in 1 mL of 10 μM MB aqueous solutions for overnight and dried in an N_2 stream. The standard equation used to calculate EFs in this study was displayed below.[2]

$$EF = \frac{I_{SERS}/N_{SERS}}{I_{REF}/N_{REF}} \quad \text{(S2)}$$

where I_{SERS} and I_{REF} are the average intensities of characteristic Raman peak of MB at 1628 cm^{-1} from SERS and reference substrates, respectively, in the same scattering geometry. N_{SERS} and

N_{REF} are the number of MB molecules absorbed on SERS substrates and reference substrate, respectively. N_{SERS} and N_{REF} were calculated by using the following equations:

$$N_{SERS} \text{ or } N_{REF} = C_{absorb} V_{soak} N_A \quad \text{(S3)}$$

$$C_{absorb} = C_{soak} - C_{remain} \quad \text{(S4)}$$

where C_{soak}, C_{absorb} and C_{remain} are molar concentrations of MB solution using to soak the substrates (10^{-6} M), absorbed on the substrates and remaining in solution, respectively. V_{soak} is the volume (L) of MB solution using to soak the substrates (10^{-3} L) and N_A is Avogadro's number (6.02×10^{23} mol^{-1}). C_{remain} was examined from the decrease of UV-VIS absorption intensity of remaining MB aqueous solutions. To calculate C_{remain}, the standard calibration curve of MB in a concentration range of 2 – 10 μM was constructed with UV-VIS absorbance measurements.

The calibration plot constructed from UV-VIS absorbance at 663 nm of MB standard solution (in water) was displayed in Figure S6. C_{remain} was estimated from the linear regression obtained from the calibration curve ($y = 0.0862x + 0.0854$, $R^2 = 0.999$). C_{absorb}, N_{SERS} ,and N_{REF} can be calculated using Equation S3 and Equation S4. The calculated parametric values of each substrate were reported in Table S3. N_{SERS} and N_{REF} calculated from Equation S3 are the number of MB molecules absorbed on the entire area of substrate. MB molecules under irradiated area (5 microns in diameter, area = 19.625×10^{-8} cm^2) on the substrate were also estimated as shown in Table S3. Based on the calculations in Equation S1, EFs from r-AgBD-ROM and Au-r-AgBD-ROM are determined to be 3.2×10^3 and 2.1×10^3, respectively.

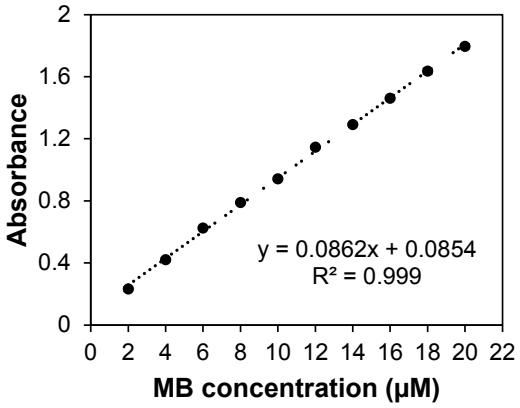

Figure S7. The calibration plot constructed from UV-VIS absorbance at 663 nm of MB standard solution (in water).

Table S3. The calculated parametric values of each substrate.

Substrates	C_{remain} **(M)**	C_{absorb} **(M)**	N_{SERS} **or** N_{REF} **(molecules)**	N_{SERS} **or** N_{REF} **In scattering area (molecules)**	I_{SERS} **or** I_{REF} **(counts)**
AgBD-ROM (reference substrate)	7.99×10^{-6}	2.01×10^{-6}	1.21×10^{15}	23.74×10^{7}	10.39
r-AgBD-ROM (SERS substrate)	7.86×10^{-6}	2.14×10^{-6}	1.29×10^{15}	25.32×10^{7}	35940.21
Au-r-AgBD-ROM (SERS substrate)	7.66×10^{-6}	2.34×10^{-6}	1.41×10^{15}	27.67×10^{7}	25425.73

SERS practical application

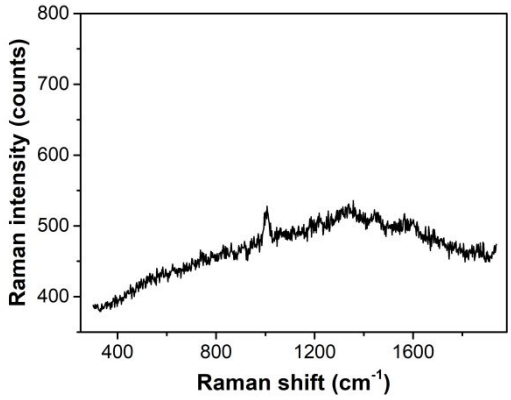

Figure S8. Raman spectra of bare Au-r-AgBD-ROM.

Table S4. Assignments of main peaks for Raman and SERS spectra of acetaminophen recorded on powder state and the Au-r-AgBD-ROM, respectively.

Acetaminophen		
Bulk powder	**SERS**	**Assignments**[3]
1654		amide I
1613	1618	*ν* CC (ring)
1562	1567	*ν* CC, σ^{ip} HNC
1450		σ^{as} CH_3
1374		σ^{s} CH_3
1328	1327	amide III band (*ν* CN/ *ν* CN (Ø)/CNH band)
1260	1275	*ν* CO, σ^{ip} HCC, *ν* CC
1241	1244	*ν* CC, σ^{ip} HOC
1172	1189	σ^{ip} HCC, *ν* CC
859	859	*ν* CC (ring)
797		*ν* CNC (ring)

Table S5. Assignments of main peaks for Raman and SERS spectra of ibuprofen recorded on powder state and the Au-r-AgBD-ROM, respectively.

Ibuprofen		
Bulk powder	**SERS**	**Assignments**[4]
1610	1613	ν_{ip} CC_{arom}
1571	1569	ν_{ip} CC_{arom} ν_{as} COOH
1344	1339	ω HCH
1210	1208	τ $HCCC_{alif}$
1186	1186	τ $HCCC_{alif}$
1010	1010	δ_{ip} HCC_{arom}
786		δ_{ip} CCOOH
748		δ_{oop} CCOOH
640	644	δ_{oop} CCC_{arom}

Table S6. Assignments of main peaks for Raman and SERS spectra of mefenamic acid recorded on powder state and the Au-r-AgBD-ROM, respectively.

Mefenamic acid		
Bulk powder	**SERS**	**Assignments**[5]
1626		β NH, ν CC (Ø2), β OH + ν C=O (f)
1605	1605	β NH, ν CC (Ø1 and Ø2), β OH + ν C=O
1585	1587	β NH (g), ν CC (Ø1 and Ø2)
1516	1518	ν CC (Ø2), β NH (g)
1448		β NH, β CH (Ø1 and Ø2), δ CH_3
1408		β NH, ν CC (Ø1 and Ø2), β OH + ν C=O (e)
1339	1334	β OH (j), ν CC (Ø2)
1281	1269	β CH in phase (Ø1, Ø2), ν CC (Ø1), ν C8N, β NH
1248	1246	β OH, β CH in phase (Ø1, Ø2), ν CCOOH
1165	1162	β CH (Ø2)
1097		β CH (Ø1, Ø2), ω CH_3

1085	1085	
1044	1025	ν CC (Ø2)
994		ω CH_3
812	817	γ CH in phase (Ø1), δ CCC in phase (Ø2), β COOH
777	786	γ CH in phase (Ø1), β COOH
705	709	CCC in phase puckering (Ø1, Ø2), γ CH_3 (C14, C15)
625	623	γ NH (k)
578		δ CCC in phase (Ø1, Ø2)

Abbreviations: Ø = phenyl group, Ø1, Ø2 = phenyl group labeled according to Figure 8 in main manuscript, ν = stretching, δ = bending, β = bending in plane, γ = bending out of plane, r = rocking, w = wagging, τ = twisting, s = symmetric, as = antisymmetric

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