Paper-based Surface-enhanced Resonance Raman Spectroscopic (SERRS) Immunoassay Using Magnetic Separation and Enzyme-catalyzed reaction

Yuanyuan Chen^a, Hanwen Cheng^a, Kha Tram^b, Shengfeng Zhang^a, Yanhua Zhao^a, Liyang Han^a, Zengping Chen^a, Shuangyan Huan^{a,b}*

^a State Key Laboratory for Chemo/Biosensing and Chemometrics, College of Chemistry and Chemical Engineering, Hunan University, Changsha 410082, China (* Corresponding author: Dr. Shuangyan Huan, <u>shuangyanhuan@yahoo.com.cn</u>)

^b Department of Biochemistry and Biomedical Sciences and Department of Chemistry and Chemical Biology, McMaster University, 1200 Main Street West, Hamilton, Ontario L8N 3Z5, Canada



SI 1A The SERRS spectra of malachite green (10^{-6} M) addition: (a) after silver colloids were dried on the filter paper using stock solution of silver sol; (b) mixed with 10x concentrated silver sol prior to making the silver colloids substrate on filter paper; (c) before silver colloids were dried using 10x concentrated silver sol; (d) after silver colloids were dried on filter paper using 10x concentrated silver sol.



SIIB (a) MG molecules were mixed with silver colloids and then spotted onto the filter paper (b) Silver colloids were spotted onto the filter paper (but not dried) before addition of MG (c) Silver colloids were spotted onto the filter paper and dried before spotting the MG.



SI2 The SEM of silver colloid morphology on filter paper prepared with different chloride ion concentrations for pre-aggregation: (a) 0mM; (b) 10mM; (c) 15mM; (d) 20mM; (e) 25mM; (f) 50mM.



SI3 The SERRS intensity of MG (10^{-7} M) at 1615 cm^{-1} on different paper based silver colloids substrates prepared with different chloride ion concentrations corresponding to the SERS substrates shown in SI2. Each data plot is the average of at least 6 detections.



SI4 SERRS spectra of different MG concentrations on paper based silver colloid substrate prepared with 20mM chloride ions as shown in SI2 d and SI3: (a) 10^{-6} M, (b) 10^{-7} M, (c) 10^{-8} M, (d) 5×10^{-9} M, (e) 10^{-9} M, (f) 5×10^{-10} M, (g) 10^{-10} M.



SI5 Distribution of SERS enhancement effect on the paper based silver colloids substrate (a) with PVP modification and (b) without PVP modification. BCIP (5-bromo-4-chloro-3-indolyl phosphate) of 3mg/mL was reacted with ALP enzyme $(4.14 \times 10^{-9} \text{ M})$ to produce BCIP dimer for the SERS detections. The SERS intensity at 601 cm^{-1} was used for plotting. Plotted data is the average SERS intensity with standard error.



SI6 The SERS spectra of 3 mg/ml of BCIP on paper based silver colloids substrate before and after hydrolysis by ALP at 4.14×10^{-9} M.