

## ***Supporting Information***

# Monitoring Thrombin Generation and Screening Anticoagulants through Pulse Laser–Induced Fragmentation of Biofunctional Nanogold on Cellulose Membranes

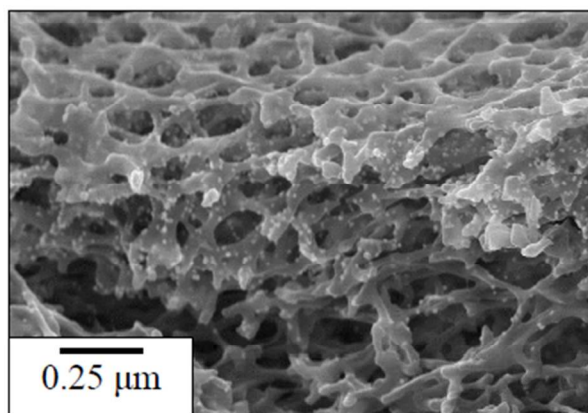
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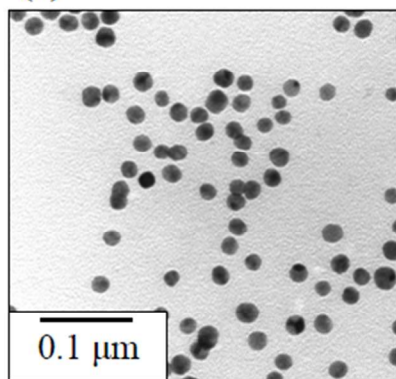
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**(A)**

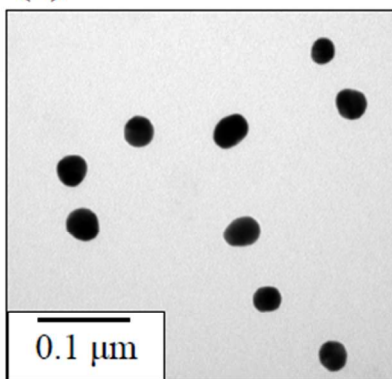


**(B)**

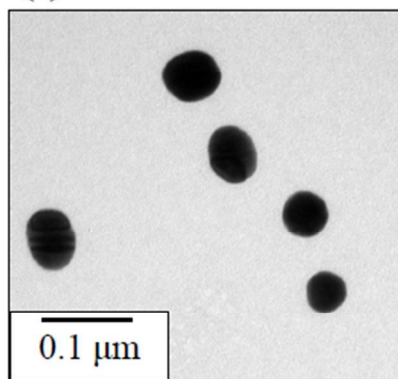
**(a)**



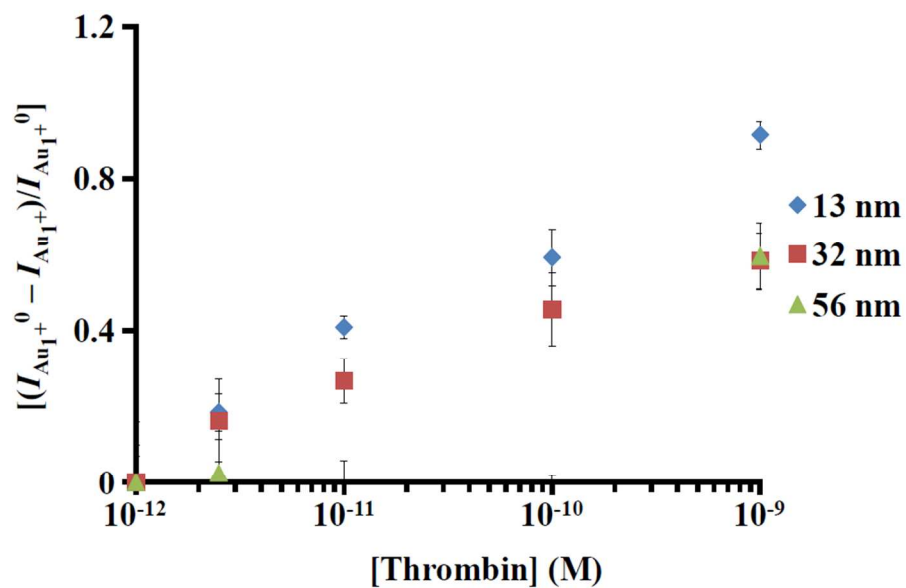
**(b)**



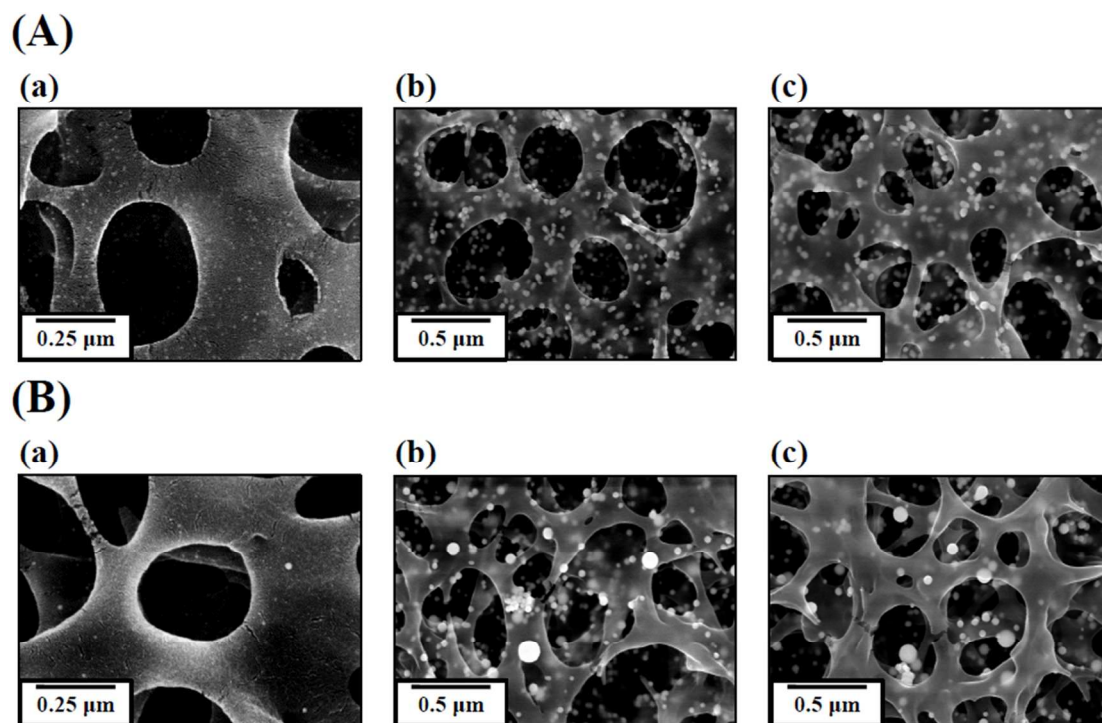
**(c)**



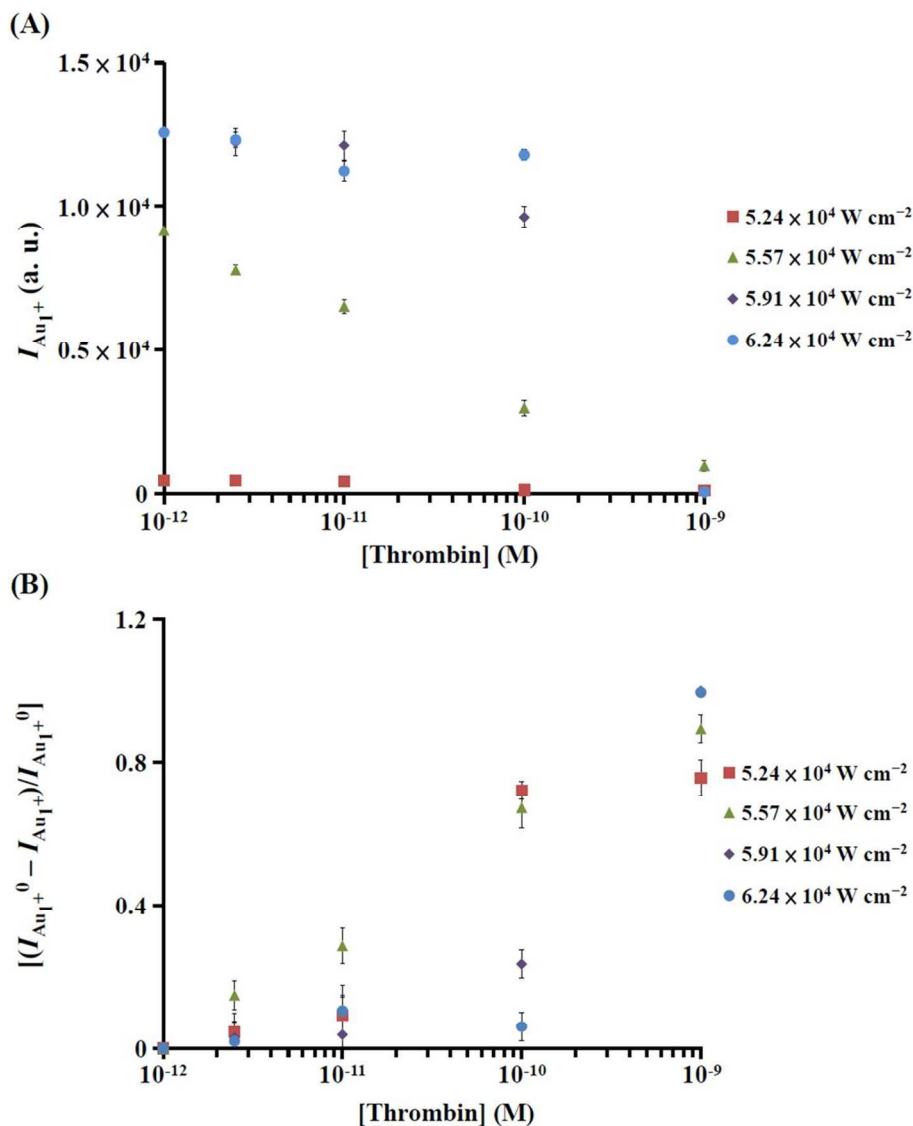
**Figure S1.** (A) Scanning electron microscopy (SEM) image (lateral view) of a Au NPs-MCEM and (B) TEM images of the synthesized (a) 13-, (b) 32-, and (c) 56-nm Au NPs.



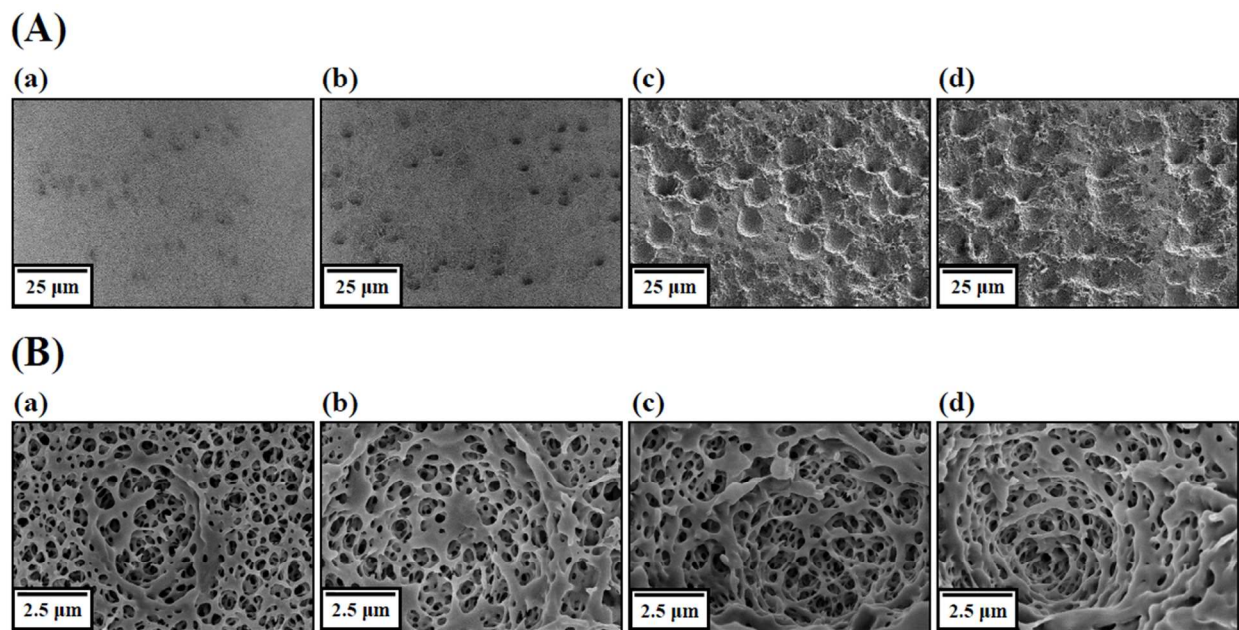
**Figure S2.** Effect of particle size of Au NPs on LDI-MS analysis of thrombin using Au NPs–MCEM substrates.  $I_{Au1+}^0$  and  $I_{Au1+}$  represent the signal intensities of  $[Au_1]^+$  ions in the absence and presence of thrombin, respectively. The Au NPs–MCEM was reacted with fibrinogen (1.0  $\mu$ M) and thrombin (0–1.0 nM) in PBS.



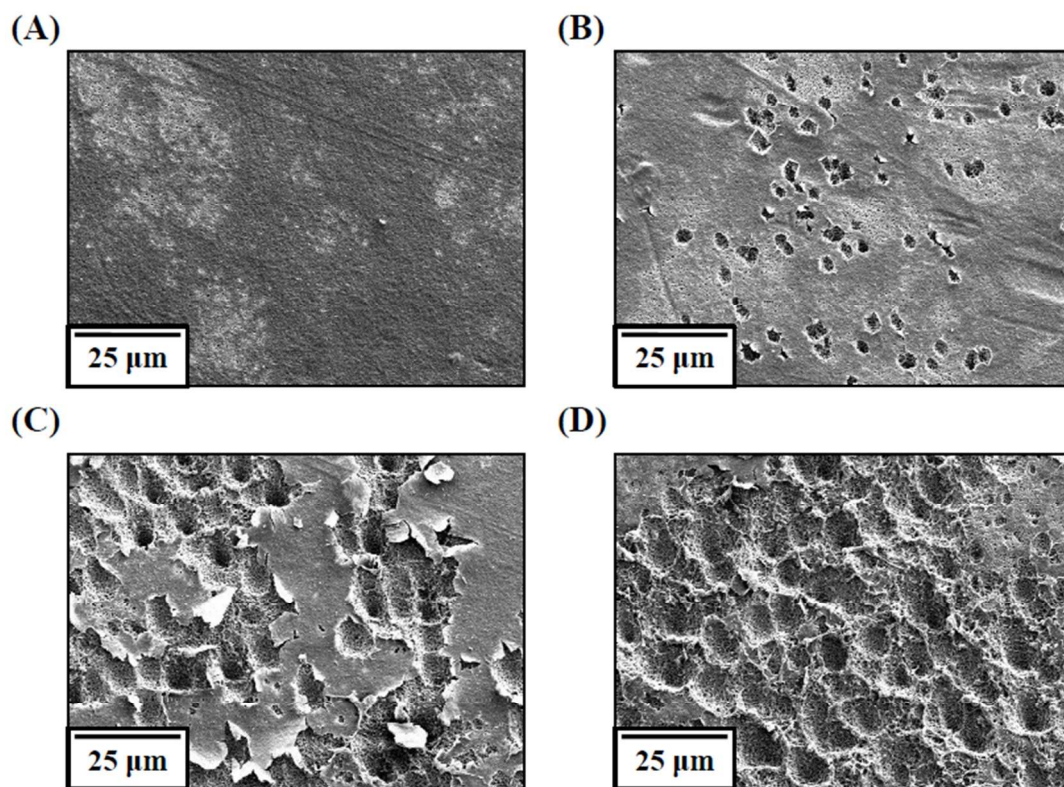
**Figure S3.** SEM images of Au NPs–MCEMs with Au NPs having dimensions of (a) 13-, (b) 32-, and (c) 56-nm (A) before and (B) after pulse laser irradiation ( $5.57 \times 10^4 \text{ W cm}^{-2}$ ). Other conditions were the same as those described in Figure 1.



**Figure S4.** (A)  $[\text{AuI}]^+$  intensities ( $I_{\text{AuI}^+}$ ) and (B) relative values of  $[(I_{\text{AuI}^+}^0 - I_{\text{AuI}^+})/I_{\text{AuI}^+}^0]$  of  $[\text{AuI}]^+$  ions obtained from Au NPs–MCEMs after irradiation from a pulsed Nd:YAG laser (power densities from  $5.24 \times 10^4$  to  $6.24 \times 10^4 \text{ W cm}^{-2}$ ). The Au NPs–MCEMs were reacted with fibrinogen ( $1.0 \mu\text{M}$ ) and thrombin ( $0$ – $1.0 \text{ nM}$ ) in PBS solution.  $I_{\text{AuI}^+}^0$  and  $I_{\text{AuI}^+}$  represent the signal intensities of  $[\text{AuI}]^+$  ions in the absence and presence of thrombin, respectively. Error bars represent standard deviations from four repeated experiments. Other conditions were the same as those described in Figure 1.

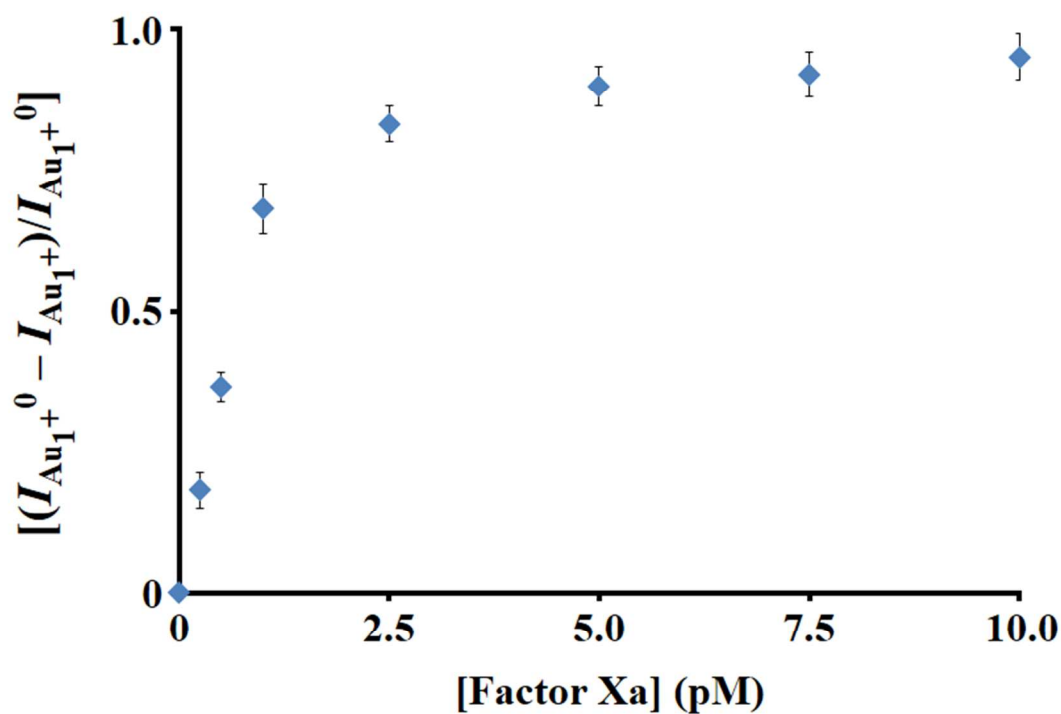


**Figure S5.** (A) Low- and (B) high-magnification SEM images of Au NPs–MCEMs after laser irradiation at power densities of (a)  $5.24 \times 10^4$ , (b)  $5.57 \times 10^4$ , (c)  $5.91 \times 10^4$ , and (d)  $6.24 \times 10^4$  W cm<sup>-2</sup>. Other conditions were the same as those described in Figure S4.



**Figure S6.** SEM images of a fibrin–Au NPs–MCEM after laser irradiation at power densities of (A)  $5.24 \times 10^4$ , (B)  $5.57 \times 10^4$ , (C)  $5.91 \times 10^4$ , and (D)  $6.24 \times 10^4$  W cm<sup>−2</sup>. The Au NPs–MCEM was reacted with fibrinogen (1.0 μM) and thrombin (1.0 nM) in PBS solution. Other conditions were the same as those described in Figure 1.





**Figure S7.** Validation of the use of the Fib–Au NPs–MCEM/LDI-MS system for the detection of Factor Xa (0–10 pM) spiked in 200-fold diluted plasma samples.  $I_{\text{Au}_1^+}^0$  and  $I_{\text{Au}_1^+}$  represent the signal intensities of  $[\text{Au}_1]^+$  ions in the absence and presence of spiked Factor Xa, respectively. Error bars represent standard deviations from three repeated experiments. Other conditions were the same as those described in Figure 4.