Gold nanoparticle amplification strategies for multiplex SPRi-based immunosensing of human pancreatic islet hormones

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Atomic Force Microscopy

Atomic force microscopy (AFM) measurements were performed using a Nanoscope III instrument (Digital Instruments, USA) and Nanoscope v 5.12r5 software. AFM images were acquired in tapping mode in air at room temperature with a silicon probe having a nominal spring constant of 42 N/m and a nominal resonance frequency 330 kHz (model PPP-NCHR, NANOSENSORSTM).
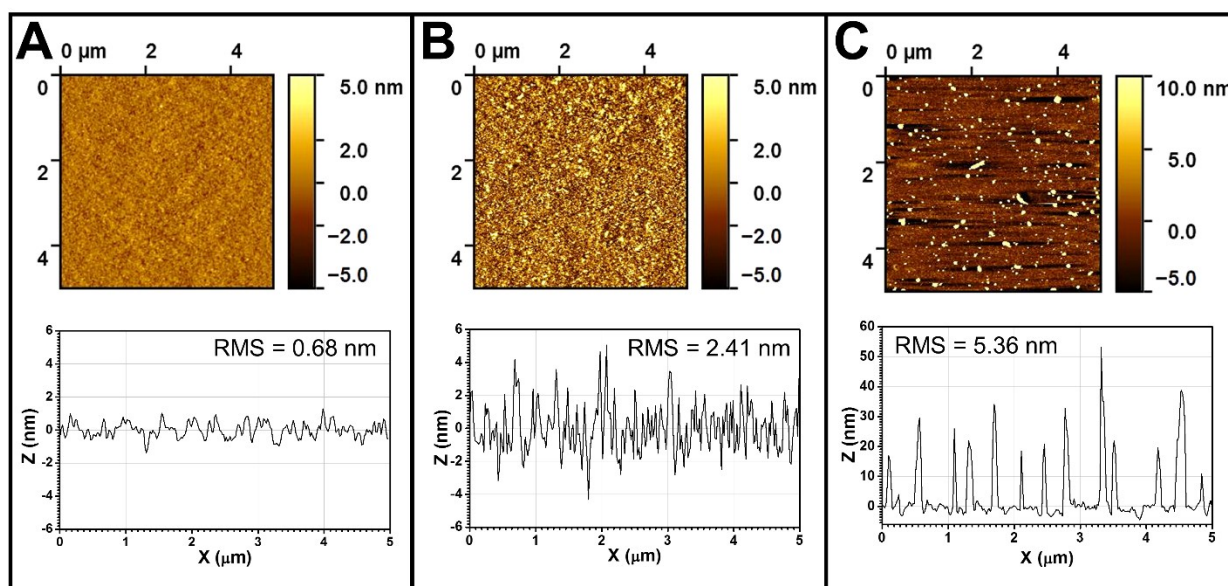

Figure S1. 2D AFM images and surface topography of: A) a clean gold surface, B) a gold surface functionalized with a self-assembled monolayer (SAM) of hexa(ethylene glycol) dithiol and C) a gold-SAM functionalized suface with gold nanopaticles.
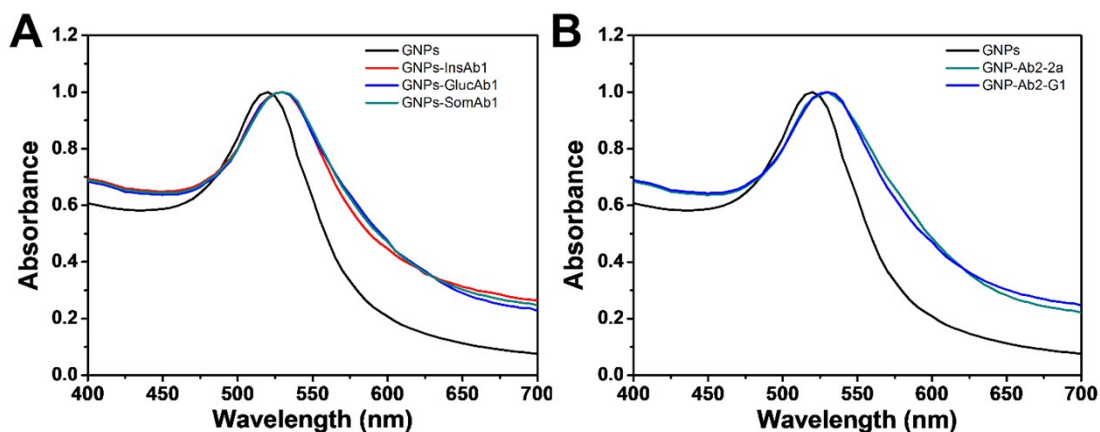
Figure S2. UV-vis absorption spectra of functionalized gold nanoparticles with: A) primary antibodies and B) secondary antibodies.
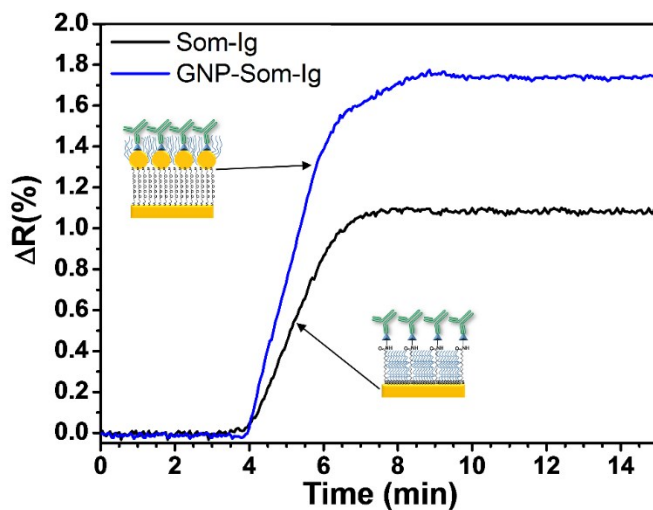

Figure S3. Real-time SPR angle shift sensorgrams comparison in response to the injection of 2μg/mL of anti-somatostatin antibody for a surface functionalized with gold nanoparticles and a surface without gold nanoparticles.

Table S1. Quantification of non-specific absorption of 1 mg/mL of BSA or LYZ on the sensor's surface during multiplexed analysis. Quantification was performed for two negative control: the bare SAM surface and spots functionalized with BSA (control). This calculation assume that $1RU = 1RIU = 1pg/mm^2$ of surface mass shift at a fixed wavelength of 800 nm. Only LYZ is reported in pg/mm^2 since it produced the largest sensor response.

Spot	ΔR (%) BSA [1mg/mL]	ΔR (%) LYZ [1mg/mL]	Amount of LYZ absorbed (pg/mm^2)
Insulin	0.04 ± 0.02	0.15 ± 0.02	90 ± 12
Glucagon	0.04 ± 0.01	0.11 ± 0.03	66 ± 18
Somatostatin	0.04 ± 0.02	0.16 ± 0.03	96 ± 18
Surface	0.06 ± 0.01	0.16 ± 0.04	96 ± 24
Control	0.03 ± 0.02	0.07 ± 0.04	42 ± 24