Supporting information

Materials. The 50-nm and 200-nm diameter mono-disperse silica beads (density 1.96 g cm^{-3}) functionalized with amino groups (100 and 80 nmol mg^{-1}, correspondingly) were purchased from G. Kisker GbR (Steinfurt, Germany). 100-nm diameter 3-aminopropyl functionalized silica particles, biotinamidohexanoic acid-3-sulfo-N-hydroxysuccinimide ester sodium salt (sulfo-NHS-biotin), deglycosylated avidin from hen egg white, sodium chloride, sodium hydroxide, phosphate buffered saline tablets (PBS) were obtained from Sigma-Aldrich (Dorset, UK). 10 kDa molecular-weight cut-off Slide-A-Lyzer Dialysis Cassettes and 2- (4'-hydroxyazobenzene) benzoic acid (HABA) which were purchased from Thermo Fisher Scientific Inc. (Rockford, USA). The Biacore sensor chips SA were purchased from GE Healthcare UK Ltd. (Little Chalfont, UK).

Biotin labelling. The labeling of the amino groups-containing silica particles with sulfo-NHS-Biotin was conducted according to the protocol recommended by manufacturer. 200 µL of silica particles suspension was diluted with PBS resulting in particle density equal 5 mg mL^{-1}. 10 mg mL^{-1} (17 mM) stock solution of sulfo-NHS-biotin in water was prepared fresh before the experiment and 15 µL were added to the particle suspension (final biotin concentration- 150 µg mL^{-1}). The suspension was incubated with biotin for 1 h at the room temperature. In order to remove the non-reacted biotin the silica particles were dialysed against 3 changes of 0.5 L of PBS solution in the Slide-a-Lyzer cassettes. The dialysis was conducted for 12 h at 4 °C upon constant steering. The biotinylated silica particles were removed from the dialysis cassettes and kept in the fridge at +4 °C. In order to obtain the nanoparticles with lower biotin density, 10 times lower concentration of sulfo-NHS-biotin (final biotin concentration- 15 µg mL^{-1}) was used for labeling. All other steps were conducted as described above.

Biotin quantification. The amount of biotin immobilized on the silica particles was measured using HABA assay. HABA assay is based on the replacement of the avidin by biotin from the HABA-avidin complex. HABA assay was performed according to the protocol recommended by manufacturer's instructions. 10 mg of avidin were mixed with 600 µL of HABA reagent and was adjusted to 20 mL with PBS. The HABA reagent was prepared by dissolving 24.2 mg of HABA in 9.9 mL of water, then adding 100 µL of 1 N NaOH. The measurements were conducted in the

microtiter plate in triplicates. 90 µL of avidin-HABA reagent were added to each well and mixed with 10 µL of the biotinylated sample which contained 50 µg silica particles. The mixture was incubated for 15 min and absorbance was measured using MRX-TC Revelation Microtiter plate reader equipped with 490 nm filter (Dynex Technologies, Chantilly**,** USA**).** The biotin quantification was performed accordingly to the calibration generated for a range of diluted samples with sulfo-NHS-biotin concentration ranging from 6-100 µg mL^{-1} (1-17 µM).

Biacore experiments. The interaction analysis was performed with a Biacore 3000 instrument (GE Healthcare Life Sciences, USA) at 25 °C using PBS (0.01 M phosphate buffer, 0.0027 M potassium chloride and 0.137 M sodium chloride, pH 7.4) as the running buffer at flow 20 µL min^{-1}. The suspensions of all biotin-labeled nanoparticles were filtered through the syringe filters with pore diameter 0.45 µm (Supelco, UK) and diluted in PBS for the analysis. Sensorgrams were collected sequentially for six analyte concentrations running in KINJECT mode. Dissociation constants (K_D) were calculated from plots of the equilibrium biosensor response (Figure 1S-3S) as a function of the biotin concentration using the BIA-evaluation 3.1 software.

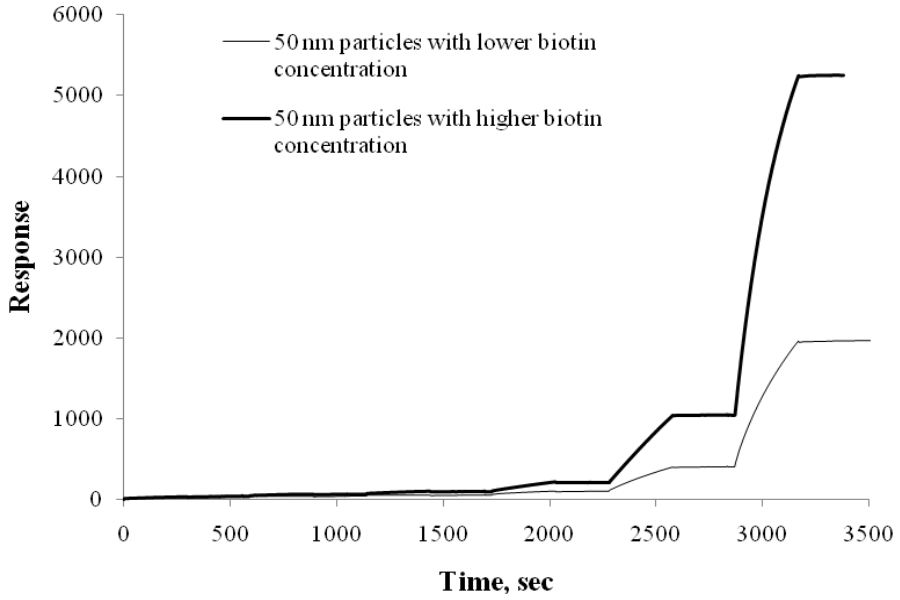

Figure 1S. Sensorgram of time-dependent binding of 50-nm silica nanoparticles functionalized with biotin to streptavidin-coated surface of Biacore sensor chip. The corresponding biotin concentrations for biotinylated particles with lower biotin concentration are 0.0028 nM, 0.028 nM, 0.28 nM, 2.8 nM, 28 nM, 280 nM and with higher biotin concentration- 0.005 nM, 0.05 nM, 0.49 nM, 4.85 nM, 48.5 nM, 485 nM.

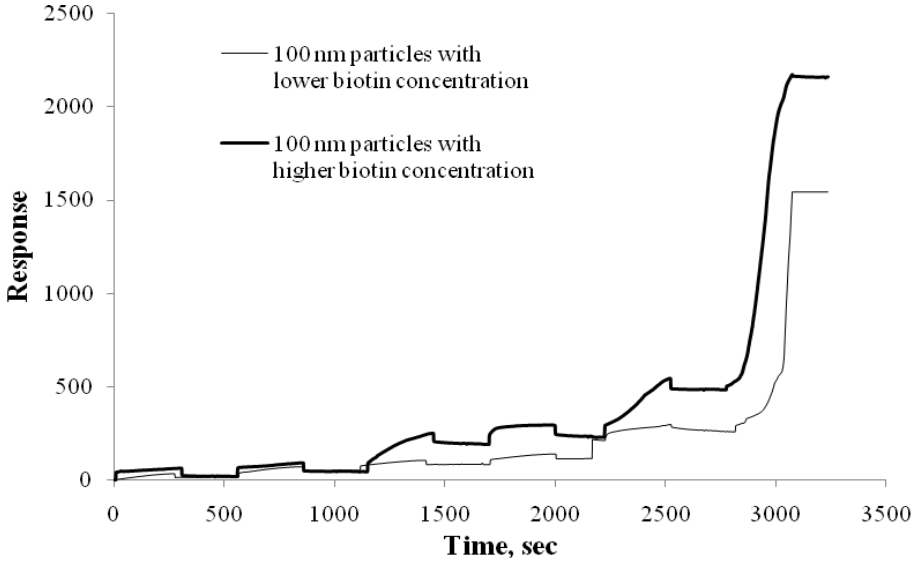

Figure 2S. Sensorgram of time-dependent binding of 100-nm silica nanoparticles functionalized with biotin to streptavidin-coated surface of Biacore sensor chip. The corresponding biotin concentrations for biotinylated particles with lower biotin concentration are 0.0038 nM, 0.038 nM, 0.38 nM, 3.8 nM, 38 nM, 380 nM and with higher biotin concentration- 0.0077 nM, 0.077 nM, 0.77 nM, 7.7 nM, 77 nM, 770 nM.

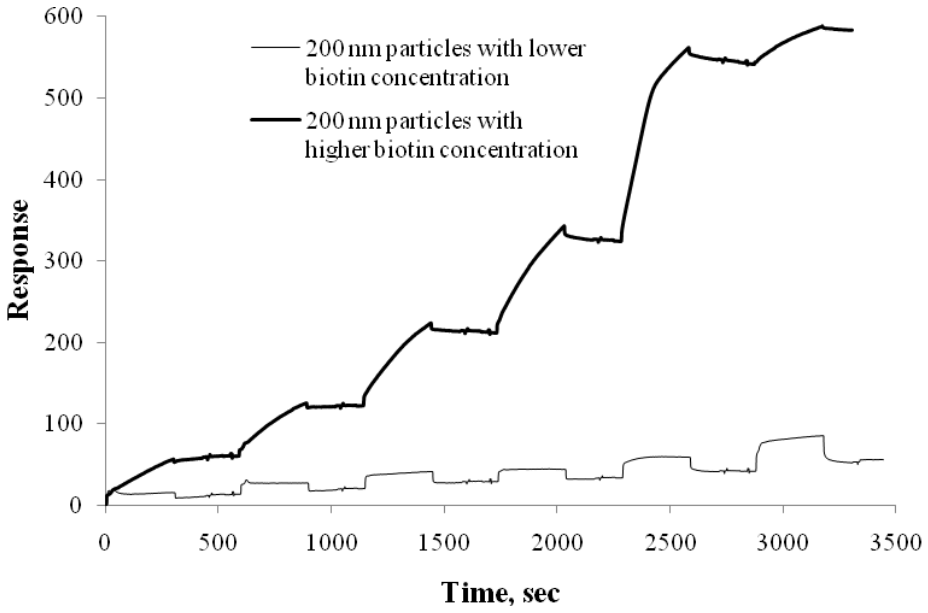

Figure 3S. Sensorgram of time-dependent binding of 200-nm silica nanoparticles functionalized with biotin to streptavidin-coated surface of Biacore sensor chip. The corresponding biotin concentrations for biotinylated particles with lower biotin concentration are 0.0024 nM, 0.024 nM, 0.24 nM, 2.4 nM, 24 nM, 240 nM and with higher biotin concentration- 0.0044 nM, 0.044 nM, 0.44 nM, 4.4 nM, 44 nM, 440 nM.