Supplementary I – Experimental Procedures

Gold nanoparticle synthesis

0.4 mL of 0.5% (w/v) $HAuCl_4$ added to 15.6 mL of ultra pure H_2O was preheated in reflux reaction flask with continuous stirring for 30min at 60°C. 0.8 mL of 1% (w/v) citrate and 20 µL of 1% (w/v) tannic acid in 3.18 mL of water were preheated for 10 min at 60°C separately. Upon preheating, the two set of solutions were mixed quickly at the same temperature. Reaction temperature was maintained at 60°C for 10 min before increasing the temperature to 110°C. This temperature was then maintained for another 5 min before switching off the power. The product was finally cooled gradually to room temperature.

Au-dsDNA conjugation

Centrifuge a portion of nAu solution and discard the supernatant. Freshly prepared buffer containing 4, 4'-(phenylphosphinidene) bis-(benzenesulfonic acid) (PPBS) (20 mg PPBS/500 µL H_2O) was added to the residue for stabilization at 10 µL of PPBS per 500µL nAu solution ratio. The mixture was allowed to incubate at room temperature overnight. After washing with ultra pure H_2O to remove the excess PPBS, dsDNA with three times the amount of stabilized nAu, 5µL of 1 M Tris and 2.5 µL of 1 M NaCl was added to the stabilized nAu. The mixture was left to stand at room temperature condition for 3 h. dT_5 was then added to the mixture at 500 times the amount of nAu. NaCl concentration was increased to 75 mM and total volume of the mixture was topped up to 100µL with ultra pure H_2O. This mixture was left overnight for incubation. 200 mM increase in concentration of NaCl was then done every 3-4 hours till a final concentration of 600 mM was reached. A full day incubation was done before 400 µL of 50 mM Tris and NaCl buffer mixture was added. The mixture was then centrifuged and the supernatant was discarded to remove the excess reactants. The washing was done twice before the volume of the residue was topped up with 50 mM Tris and NaCl to a final volume of 50 µL.

Conjugate digestion

Conjugate digestion of each type of nAu-dsDNA consisted of (1) negative control, (2) digested product and undigested strands released by dithiothreitol (DTT) through thiol exchange reaction, and (3) sample with ethylenediaminetetraacetic acid (EDTA) added to inhibit EcoRV after digestion (See Scheme 1). EDTA was used to confirm that no unintended free DNA digestion occurred after the thiol exchange reaction. Except the negative controls, all conjugates were digested with 100 U of EcoRV under 37°C over 20 hours in a standard digestion buffer. Centrifugation was then performed to extract the supernatant containing the digested fragments and dehybridized fragments. The residue in Case (2) was incubated with 1M DTT overnight at room temperature condition to release the undigested strands. For Case (3), EDTA was added immediately after the digestion before centrifugation to inhibit EcoRV. All samples were run together in a 12% polyacrylamide gel electrophoresis (PAGE) at a constant current of 12A. The gel was stained with ethidium bromide before the digestion efficiency was quantified using a gel scanner (Gene Genius Bioimaging System, Syngene). The efficiencies of EcoRV digestions on the four types of nAu-dsDNA (conjugate digestions) were compared to their respective free DNA digestions to determine the minimum **d** needed for efficient digestion.

Free DNA digestion

The free DNA digestions conducted to benchmark the efficiencies were performed in each experiment such that 1 μl of 2 μM dsDNS was digested in a buffer system of 50 mM NaCl and Tris, 2 mM $MgCl_2$ and 1 X BSA forming a total volume of 40 μl. The mixture was subjected to 50 U of EcoRV at 37°C for 20 hours. Digested samples were processed and run in PAGE together with conjugate digestion samples.

Supplementary II – Gel Image of Free DNA Digestion

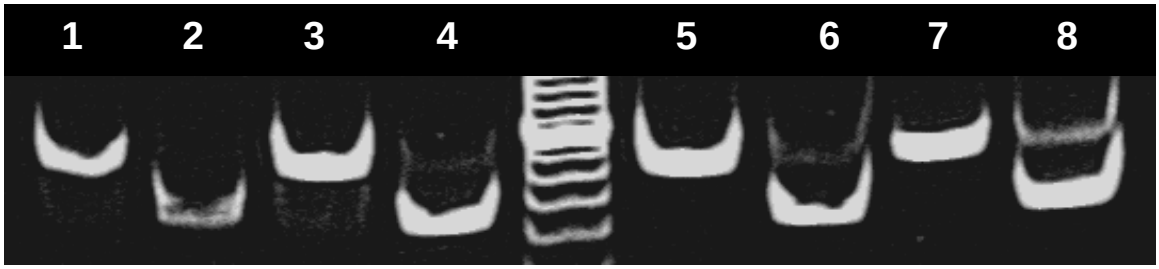

Figure S1: 20 hours free DNA digestion. Lanes 1-8, excluding the ladder, correspond to: (1) Undigested dsDNA (**d** = 5) as negative control, band detected at 80 bp; (2) Digested dsDNA (**d** = 5), band detected at 62 bp; (3) Undigested dsDNA (**d** = 4) as negative control, band detected at 80 bp; (4) Digested dsDNA (**d** = 4), bands detected at 62 bp and 80 bp; (5) Undigested dsDNA (**d** = 3) as negative control, band detected at 80 bp; (6) Digested dsDNA (**d** = 3), bands detected at 62 bp and 80 bp; (7) Undigested dsDNA (**d** = 2) as negative control, band detected at 80 bp; (8) Digested dsDNA (**d** = 2), bands detected at 62 bp and 80 bp.