Electronic Supplementary Information (ESI)

Unzipping and Shearing DNA with Electrophoresed Nanoparticle in Hydrogel

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S1. DNA sequences

Anchor: acrydite-5'-TGGAGACGTAGGGTATTGAATGAGGGCCGTAAGTTAGTTGGAGACGTAGG-3'

U15-GNP: CTTACGGCCCTCATTTT-3'-thiol-GNP

S15-GNP: GNP-thiol-5'-TTCTTACGGCCCTCATT-3'

U20-GNP: 5'-TAACTTACGGCCCTCATTCATT-3'-thiol-GNP

S20-GNP: GNP-thiol-5'-TTTAACTTACGGCCCTCATTCA-3'

U25-GNP: 5'-ACTAACTTACGGCCCTCATTCAATATT-3'-thiol-GNP

S25-GNP: GNP-thiol-5'-TTACTAACTTACGGCCCTCATTCAATA-3'

U30-GNP: 5'-CCAACTAACTTACGGCCCTCATTCAATACCTT-3'-thiol-GNP

S30-GNP: GNP-thiol-5'-TTCCAACTAACTTACGGCCCTCATTCAATACC-3'

U49-GNP: 5'-CCTACGTCTCCAACTAACTTACGGCCCTCATTCAATACCCTACGTCTCCTT-3'-thiol-GNP

S49-GNP: GNP-thiol-5'-TTCCTACGTCTCCAACTAACTTACGGCCCTCATTCAATACCCTACGTCTCC-3'

	U15	S15	U20	S20	U25	S25	U30	S30	U49	S49
Tm	68°C		75°C		78°C		82°C		94°C	

Figure S1. Melting temperature for the DNA duplex (calculated using Nupack)

S2. DNA-GNP's preparation

DNA strands and modified DNA strands are purchased and HPLC purified from Eurofins Genomics. After arrival, all DNA strands are hydrated with mQ water.

Three types of GNPs, originally coated with citrate acid, are used: 5 nm and 10 nm (Tanaka company, Japan). To passivate GNPs with Bis(p-sulfonatophenyl)phenylphosphine dihydrate dipotassium (BSPP) salt, six microtubes are filled with 250 µl of GNPs and 50 µl of BSPP solution (50 mg/ml in mQ). Microtubes are incubated at 50°C for 30 min in a thermocycler (Eppendorf Mastercycler personal). Microtubes are centrifuged with 20,000 G at 4°C for 2 h in a centrifuging machine (Kubota 3780). Supernatant is carefully

discarded. Remained gold become the stock gold solution.

Thiol-modified DNA (50 µM) is incubated with 100 µM TCEP for 1h at room temperature; final DNA concentration is 40 µM.

To prepare 10 µl of DNA-GNPs, the GNPs stock is diluted to a final concentration of 1 mg/mL BSPP, 1xTBE, 200 mM NaCl, and 4 µl mQ. After this, 1 µl of deprotected thiol-modified DNA (26 µM) is added. The mixture is pipetted thoroughly. The sample is heated at 50°C for 1 h and, finally, stored at room temperature. Results of characterization by 2% agarose gel electrophoresis (1.25 V cm^{-1} for 15min) is shown in Fig. S2.

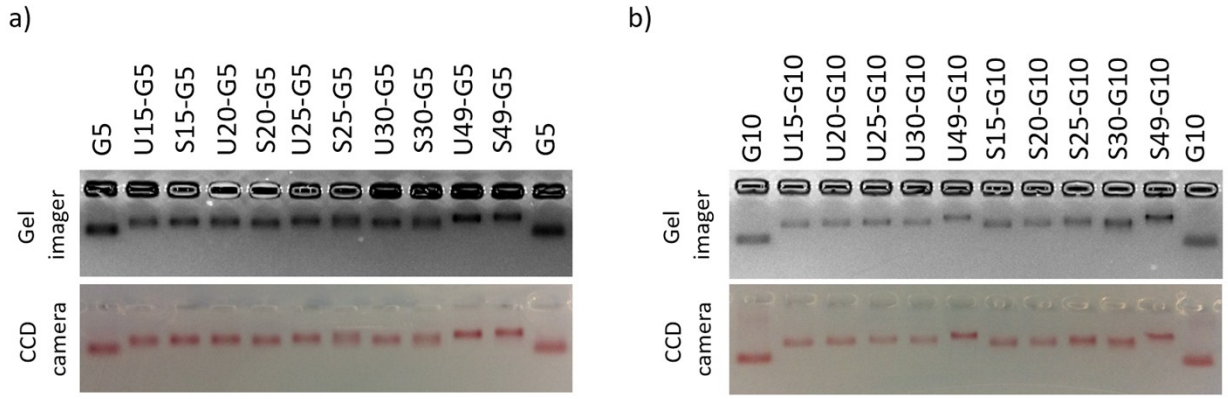

Figure S2. 2% gel electrophoresis assay for both 5nm and 10nm GNP. Gel imager uses white EPI light source and 530/28 filter. a) 5 nm GNP. b) 10 nm GNP.

S3. Preparation of capillary containing DNA-GNPs immobilized in hydrogel

Fig. S3 shows a general scheme of the preparation method.

For preparing the hydrogel 3 cm (+ 1 mm) capillaries are filled with 3% SOL hydrogel. Usually, 30 µl of hydrogel is prepared by mixing 4.5 µl of 20% acrylamide/bis solution (3.3% acrylamide:bis) (this is diluted from an original 30% acrylamide/bis solution) (SERVA), 6 µl of 5 µM Acrydite DNA, 3 µl of 10TBE, 9.9 µl of mQ, with 0.6 µl of 10% TEMED and 6 µl of 1% APS.

For immobilizing DNA-GNPs into hydrogel, capillaries with polymerized gel are carefully filled with 1xTBE buffer solution by using short-bevel needle syringes. Capillaries are pre-run in a vertical chamber, with the gel part close to the positive electrode, for 30 min under 5V (1.25V/cm) in order to remove undesired molecules. Then, 0.5 µl of DNA-GNPs in 8% sucrose are introduced through the top capillary end. The chamber is run at 5V for 2h, being this time enough for complete capturing of DNA-GNPs. Finally, the 1xTBE buffer in the capillary is extracted with the syringe and a new 1xTBE is injected.

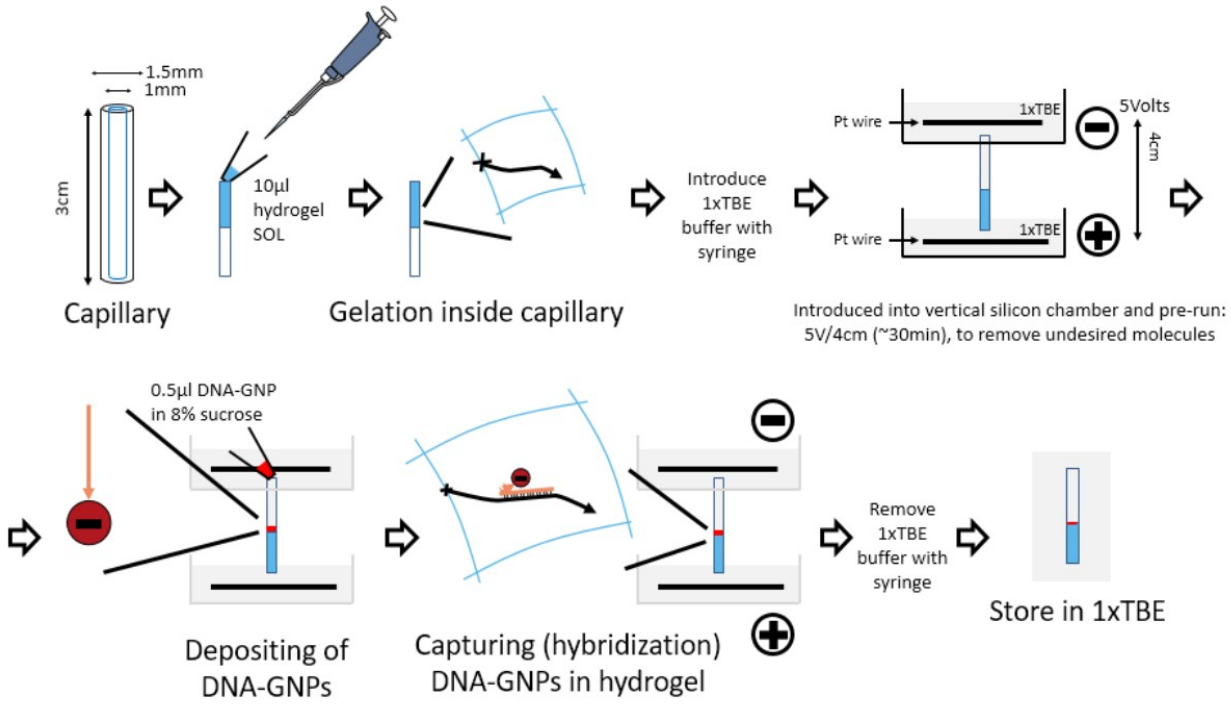

Figure S3. Scheme of the preparation of capillary

S4. Video recording

The CCD recording setup scheme is shown in Fig. S4. As the GNPs have a visible reddish color, we use a CCD camera (canon EOS Kiss X7) coupled to an optical microscope stage. A 1 mm thick polytetrafluoroethylene (PTFE) white sheet is illuminated by a white LED array, in order to provide a uniform background for the CCD recording. Note that the tubular capillary surface shows undesired reflection if directly illuminated from the camera side. Videos are recorded at maximum resolution.

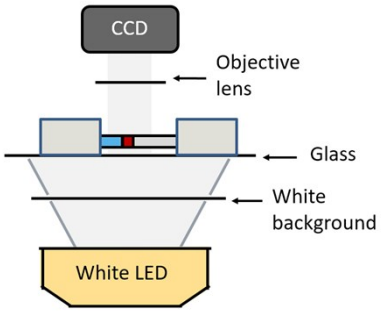

Figure S4. CCD recording setup for the dehybridization time evolution.

S5. Image processing and analyzing method

All CCD camera videos and microscope videos are processed in imageJ. The analysis scheme is given in Fig. S5.

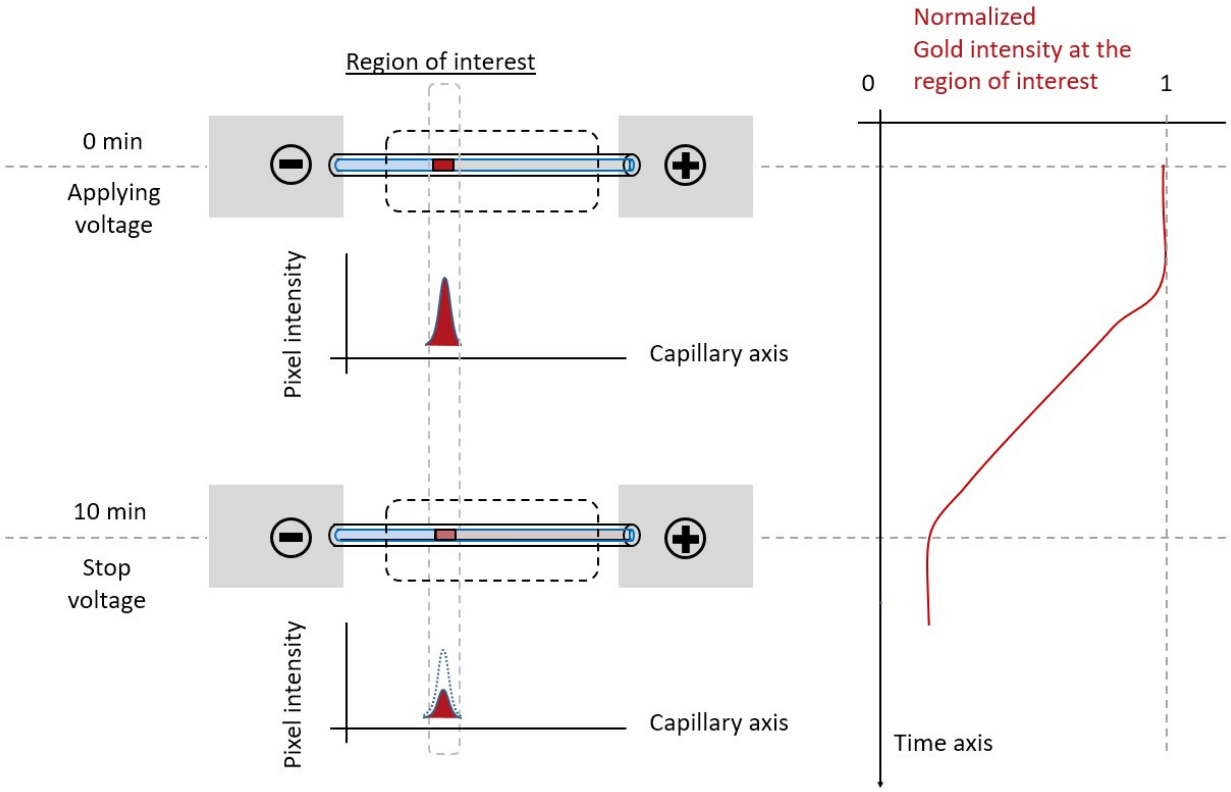

Figure S5. Method for obtaining hybridization ratio. The reddish color intensity distribution inside the capillary is shown at time zero and after 10 min (left), and its representation over time is shown (right)

S6. Voltage source and measurements of pH, temperature, electric current and zeta potential.

The regulated DC power source is a DC160-7.2 from NF corp (Japan) and its ripple noise is below 12 mV. pH measurements are taken near the electrodes by a glass electrode (Micro ToupH electrode 9618S-10D) connected to a pH meter device (Horiba D-54). Temperature measurements are taken on the capillary surface at its middle point by a Type-K thermocouple (Fluke 80PK-1 Bead Probe, fast response and 260°C max measurable temperature) connected to data logger (MCR-4TC). Electrical currents on the system are calculated by means of the voltage drop in a shunt resistor (1.2 Ω) in series with the silicon chamber. Zeta potential of -42 ± 1 mV for GNP5 is obtained in water with 10xBSPP by using Zetasizer Nano (Malvern) under default conditions.

S7. Microscopy of bear DNA duplex

Fluorescent microscopy of 15 nt DNA modified with FAM (DNA15-FAM) is done in a Nikon microscope at 1 frame per second and 0.5 sec LED light exposure. Fig. S6 shows the hybridization ratio over time of DNA15-FAM, U15-GNP5 and S15-GNP5. For the DNA15-FAM case, points are averaged over 10 sec windows.

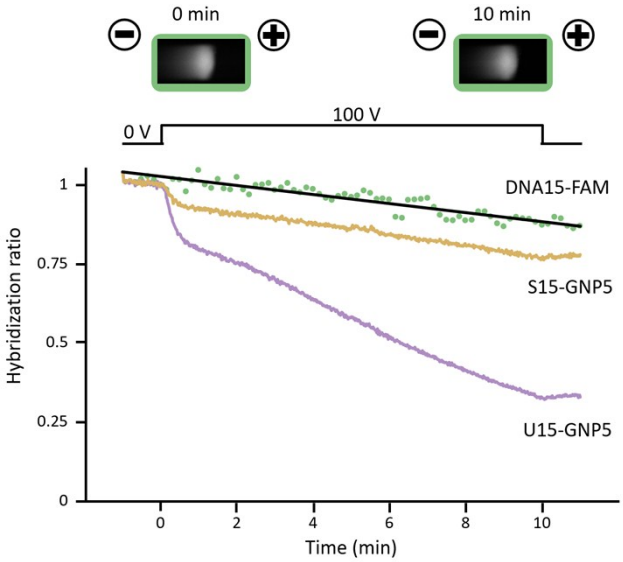

Figure S6. Control experiment for 15 nt DNA modified with FAM molecule (DNA15-FAM). Top: microscopy images for 0 min and 10 min. Below: hybridization ratio over time for DNA15-FAM and representative U15-GNP5 and S15-GNP5 for comparison.

S8. Release kinetics

The release kinetics of 15nt with GNP5 seems to be a two-step process: fast and then slow. The fast one has sigmoidal-like profile (Fig. S7a), which is faster than the profile of DNA-GNP in hydrogel without anchor (Fig. S7b). This may be due to DNA-GNPs partially hybridized to anchors or DNA-GNPs trapped in hydrogel pores. However, it is improbable since the pore size is large enough as verified with a single DNA-GNP band in hydrogel without anchor under 5V (Fig. S8). The slow process may obey a first-order kinetics, and if so it would agree with a reduced bimolecular dissociation where anchor concentration is constant.

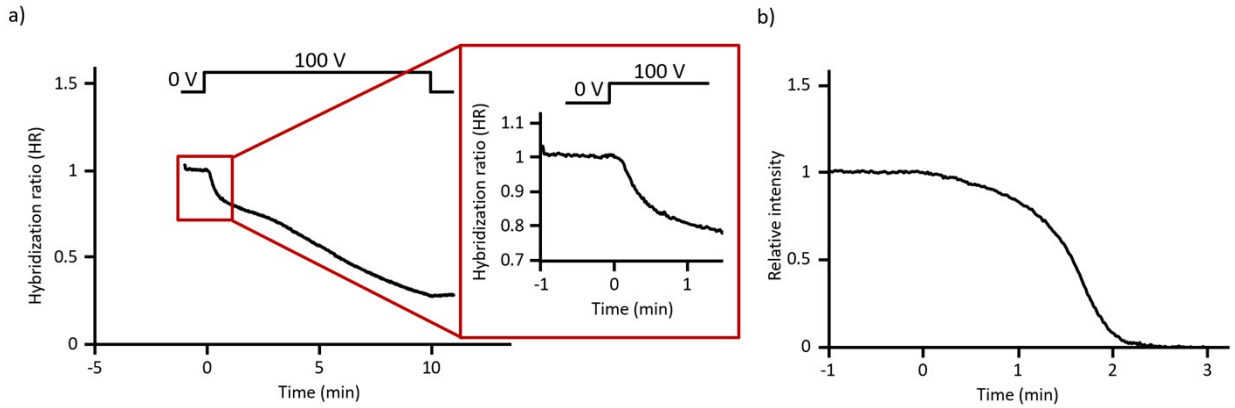

Figure S7. a) Hybridization ratio over time of U15-GNP5 under 100V (Fig. 1c from main text) and inset of the slow kinetics. b) Relative intensity of electrophoresing U15-GNP5 in a hydrogel without anchor under 100V in a vertical chamber.

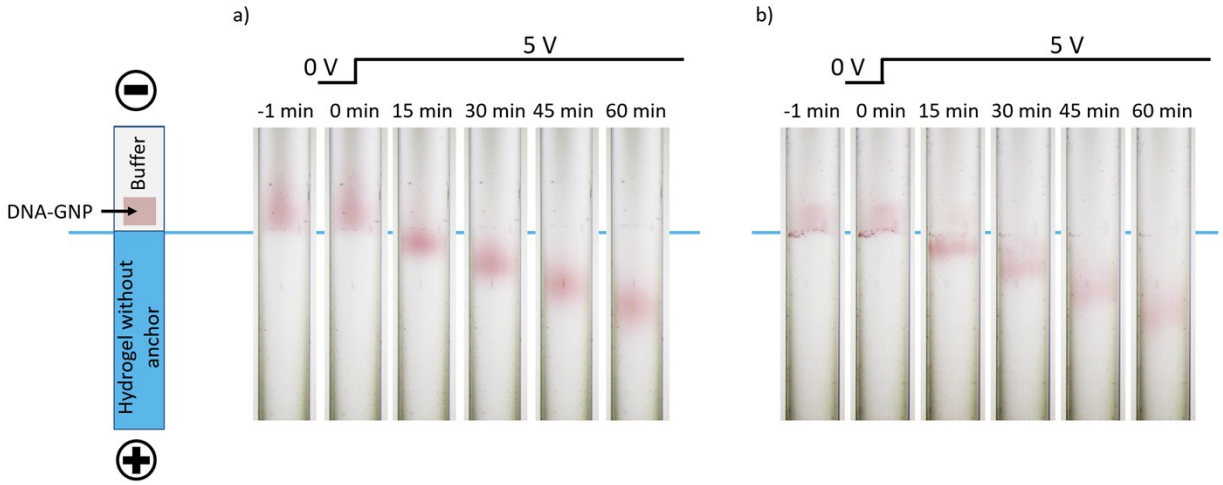

Figure S8. Band over time of DNA-GNP in a hydrogel without anchor under 5V in a vertical chamber. a) U15-GNP5. (b) U15-GNP10.