Supporting Information


Weakly Charged Cationic Nanoparticles Induce DNA Bending and Strand Separation

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In order to demonstrate that AuNPs bind to DNA by electrostatic interaction, DNA was bound to 2.8 micron streptavidin coated Dynabeads (Invitrogen) via a biotin-streptavidin link and stained with TOTO-1 (Invitrogen), AuNPs were added to quench the fluorescence by Forster resonance energy transfer (FRET). 364 nanograms of DNA was mixed with approximately $10^7$ Dynabeads and gently agitated for 10 minutes. TOTO-1 was added at 10 micromolar and the beads were imaged by fluorescence microscopy using a FITC filter (excitation wavelength of 480 nm, emission wavelength 512 nm) (Figure S1a,b). Fluorescence of the microbeads indicates that the DNA is attached and is double stranded. The beads were then re-imaged following addition of 14 pmol of AuNPs (Figure S1c, d), indicating loss of fluorescence due to FRET coupling between the TOTO-1 and AuNP. The FRET quenching phenomenon by AuNPs is highly distance dependent, so the dark substrate (Figure S1d) indicates the gold particles are close to the intercalated dye, implying the particles are bound to the DNA.
XPS was performed to determine the elemental composition of ligands (Figure S3). 10 microliters of 500 mM AuNP solution was drop cast onto a silicon wafer cleaned by nitric acid and HF. The fluid was evaporated and the substrate repeatedly washed with deionized water. XPS measurements were done at 10^{-10} Torr with a Cu Kα source on a molybdenum puck. Peak fitting was performed in commercially available Igor Pro software. 

To understand the changes in DNA structure induced by AuNPs with cationic ligands, atomistic molecular dynamics (MD) simulations were performed using AMBER 11[1] with the ff03 Cornell force field[2] for DNA (Details in Supplemental). In the simulations, AuNPs were considered to be a truncated octahedron with a 1.5 nm diameter and composed of 135 Au atoms. A total of 60 alkyl ligands were linked to randomly selected gold surface atoms, which produced a surface coverage of 11.78 Å^2/ligand (Figure S2). It has been shown that an Au_{140} nanoparticle is covered by about 60 ligands[3]. The assumed ligand structure in our simulations has been designed to simulate electrostatic and hydrophobic interaction of nanoparticles with DNA (Figure S2). The nanoparticle ligands are proprietary, but some forensic work was done for the benefit of the MD simulations. Our XPS measurements indicate, for example, a significant presence of amide groups (Figure S3). Follow up discussions with Nanoprobes indicated that all chains have this group which was used to attach terminal alkanes or amines. To summarize our best understanding from Nanoprobes and their patents, ligands with three carboxylic acid sites each are attached to the gold surface in the first step, and then molecules with primary amines are linked to these sites via amide linkage. The attached molecules may have either primary amines on both ends, leaving one exposed on the surface of the ligand shell, or they may end in an alkane. The NPs are then separated according to their charge via ion exchange chromatography and 6+ charge NPs are eluted and sold as ‘positively charged Nanogold’ by Nanoprobes. The good solubility of particles indicates enough amphiphilic character to be pulled into aqueous phase with contributions coming from amide and amine groups.
The AMBER force field has shown to reliably represent the conformational dynamics of both double-stranded and single-stranded DNA.[4-8] GAFF[9] force field, which has been parameterized specially for organic molecules made of C, N, O, H, S, P, F, Cl, Br, I, was used to represent alkyl ligands and includes the following terms:

\[
U = \sum_{\text{bonds}} k_1 (r - r_{eq})^2 + \sum_{\text{angles}} k_2 (\theta - \theta_{eq})^2 + \sum_{\text{dihedrals}} \frac{1}{2} (1 + \cos(n\phi - \gamma)) + \sum_{\text{nonbonded}} [4\varepsilon_{ij} \left( \frac{r_{ij}}{r_{ij}} \right)^{12} - 2\varepsilon_{ij} \left( \frac{r_{ij}}{r_{ij}} \right)^6] + \varepsilon_{eq}
\]

Here, \(r_{eq}\) and \(\theta_{eq}\) are equilibration structural parameters, \(k1, k2, V_n\) are force constraints, \(n\) is multiplicity and \(\gamma\) is phase angle for torsional angle parameters.

The partial charges for the ligands of the system were derived from the HF/6-31Gx electrostatic potential grid using Restrained Electrostatic Potential (RESP) method and the ANTECHAMBER component in the Amber suite[10]. The partial charge of gold atoms was set to zero. Each ligand of the same type is assumed to have an identical charge distribution.

Implicit solvent simulations where the solvent is treated by the continuum dielectric methods can significantly improve conformational searches and have already successfully predicted conformational preferences of experimentally known nucleic acid structures.[11-14]

And third self-guided MD simulations in explicit solvent and implicit solvent were performed for three AuNP-NH3 and DNA system. The SGLD method dramatically enhances conformational sampling by accelerating low-frequency, large-scale motions through the addition of an ad hoc momentum memory term. Thus, the conformational transition is promoted according to information extracted during the same simulation to achieve fast convergence in conformational sampling[15]. Use of the SGLD method has already led to
about 65 times speed up in peptide folding simulations[16] and hence show promise for our study. Even though the SGLD method introduce another level of approximations and requires accurate choice of the guiding factor, it can significantly speed up the conformational energy search in explicit solvent.

Initially, AuNPs were relaxed in implicit solvent and placed within a 5 Å proximity of DNA. All starting structures were subjected to minimization (10000 steps), followed by a different protocols for explicit and implicit simulations. For implicit simulations, the system was subjected to constrained 83.68 kJ mol\(^{-1}\) heating to 300 K (50 ps), and several consecutive MD equilibrations with a gradually declining constraint from 8.36 kJ mol\(^{-1}\) to 0.418 KJmol\(^{-1}\) (200ps). The main implicit solvent production MD runs were performed at 300 K and 0.5 M counterion concentration for at least 20 ns using 1 fs time step using an implicit solvent model (Generalized Born approximation)[17]. The temperature was maintained at 300 K using a Berendsen thermostat[18].

For explicit solvent MD minimized systems of DNA and AuNP were neutralized with Na\(^+\) ions and immersed in a water box with approximately 10 Å thick solvation shell using the TIP3P water model[19]. Additional Na\(^+\) and Cl\(^-\) ions were added to represent a 0.1 M effective salt concentration. The equilibration of a sample was carried out in 11 stages starting from a solvent minimization lasting 10000 steps while keeping the DNA-AuNP restrained for 836.8 kJ/mol. The system was then gradually heated up to 300 K in 40 ps time with the 836.8 kJ/mol restrain on the DNA-AuNP. A 200 ps NPT equilibration was performed to ensure the homogeneous solvent density with the DNA-AuNP restrain maintained at 836.8 kJ/mol. Another 10000 steps minimization followed by a 20 ps NPT run was executed with the DNA-AuNP restraint lowered to 104.6 kJ/mol. Subsequently four additional 1000 minimization steps were performed while relaxing the positional constraint from 83.68 kJ/mol to 20.92 kJ/mol. A final unconstrained 1000 step minimization was performed before heating the
system to 300 K at a constant volume within 40 ps. The long range electrostatic interactions were calculated by Particle Mesh Ewald summation[20] with a 0.00001 tolerance of Ewald convergence and the non-bonded interactions were truncated at 9 Å. The temperature was maintained at 300 K using a Berendsen thermostat[21]. SHAKE algorithm was used to constrain hydrogen atom vibrations[22]. The NVT production simulations were performed for 20 ns with a 2 fs time step.

A video is included here that shows the strand separation occurring over time in a self-guided mode. Three particles are used, and the video corresponds to the implicit MD methods described above.
Figure S1. Fluorescence microscopy images of TOTO-1 stained DNA attached to streptavidin coated magnetic beads (Dynabeads) 2.80 microns in diameter. Panels a and b are bright field and FITC fluorescence filtered images without AuNPs. Panels c and d are bright field and FITC fluorescence images with AuNPs added. Panel d is dark because the AuNPs bind the stained DNA and cause FRET quenching of emission from TOTO-1.
Figure S2. The DNA sequence modeled in the MD simulations, and the AuNP used in the MD simulations. The red spheres indicate the location of the primary amine groups.
Figure S3. XPS of the nanoparticles dispersed on silicon shows a carbon shoulder peak that indicates the presence of peptide bonds within the capping ligands which may contribute to the particles’ amphiphilic character. The peak shift is indicative of amines buried in this shoulder. The inset shows a survey scan.
**Table S-1.** Potential parameters for Au interactions

<table>
<thead>
<tr>
<th>Bonded</th>
<th>k1, kJ/mol/Å²</th>
<th>r&lt;sub&gt;eq&lt;/sub&gt;, Å</th>
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<tr>
<td>Au-Au[23]</td>
<td>1 271.27</td>
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<tr>
<td>Au-S[24]</td>
<td>158.44</td>
<td>2.65</td>
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<tr>
<td>Angle</td>
<td>k2, kJ/mol/rad²</td>
<td>Ø&lt;sub&gt;eq&lt;/sub&gt;, deg</td>
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<tr>
<td>Nonbonded -Lennard Jones</td>
<td>ε, kJ/mol</td>
<td>σ, Å</td>
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<tr>
<td>Au-organic[26]</td>
<td>0.163</td>
<td>1.467</td>
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REFERENCES


