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Controlled Translocation of DNA Segments through Nanoelectrode Gaps from Molecular Dynamics

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Molecular dynamics simulations show that electrophoresis of DNA segments through a nanoscale electrode gap can be controlled by applying appropriate biased voltages in the transmembrane direction. The translocation velocities are dependent on both the DNA molecular weight and nucleotide structure. Application of alternating driving fields results in oscillatory motion of DNA inside the gap. Interruption of the driving field can effectively pause the translocation of DNA segments. Results from this work are useful for designing novel sequencing devices.

Recently much research attention has been focused on the translocation of nucleotides through nanopores.[1−5] These studies are related to the development of novel DNA sequencing techniques based on threading DNA through nanopores. State-of-the-art micro-manufacturing technology has enabled us to fabricate nanopores at the scale of individual molecules. This made possible the screening of single molecules through nanopores by measuring molecule-specific properties. One of the methods proposed for sequencing DNA is to use a nanoelectrode gap as a screen; the identity of a nucleotide as it translocates through the gap is determined by measuring the tunneling current between the electrodes, which will be impacted by the presence of the molecule.[6−8] This technique could potentially serve as a novel tool to carry out DNA sequencing at a rate thousands of times faster than the current technology.[9]

However, fundamental questions remain concerning the feasibility of such a device. One of the primary concerns is the distinguishability of the conducting properties of the four types of nucleotides adenine (A), cytosine (C), guanine (G), and thymine (T). There are several first principal calculations on this topic, but the conclusions are still being debated.[10−12] Other questions are related to the design of the device, many of which are within the realm of investigation through molecular dynamics (MD) simulations. Among them is how to control the DNA when it is driven through the electrode gap. The essential idea behind the device is the base-by-base translocation of DNA at an appropriate rate to permit nucleotide identification. Rapid sequencing requires that the DNA segments thread the nanogap at a reasonably high velocity. On the other hand, the translocation process has to be controllable to allow sufficient time for the screening device to read the tunneling current signature unique to a base. In this report, we present a molecular dynamics simulation study of controlled translocation of DNA through nanogaps. We show that the electrophoresis of DNA segments in a nanoelectrode gap can be controlled by applying appropriate electric field in desired directions.

We have used the packages Nanoscale Molecular Dynamics (NAMD)[13] and Visual Molecular Dynamics (VMD)[14] to perform atomistic MD simulations of DNA translocation between two metallic nanoelectrodes in aqueous environment. The DNA and K^+ counterions were modeled by the AMBER (Assisted Model Building with Energy Refinement) potential.[15] The solvent water molecules were modeled by the transferable interaction potential (TIP3P).[16] The electrodes were modeled as two face-centered cubic lattices, one of platinum and one of gold, each with dimension of $3 \times 3 \times 3$ nm^3. The nano electrode gap widths investigated were 2.0 for single strand DNA (ssDNA) and 2.5 nm for double strand DNA (dsDNA). The effective gap widths between the solvent accessible surfaces are about 1.7 and 2.2 nm given the van der Waals radii of the metal atoms being about 0.3 nm. In our previous study, we have shown that those two gap widths are slightly wider than the minimum gap widths for single and double strand DNA threading.[17] Au and Pt atoms in the electrodes were fixed during simulations, and the interactions from them were calculated from the universal force fields.[18] MD simulations were performed up to 15 ns at constant pressure (1 bar) and constant temperature (300K).[19] A schematic setup of the simulation is shown in Figure 1. In the proposed device,[6] electrodes are attached between two insulating panels on the top and bottom. Two additional electrodes are placed along the *y*, or the transmembrane, direction to produce the field to drive the DNA through the gap between the Au and Pt electrodes. In our simulations, the panels and two additional nodes were excluded to reduce the simulation box to a computationally manageable size. We assume that an effective electric field can be produced along the transmembrane direction by the device. The initial momenta of the moving atoms (DNA, water, and ions) are set randomly, following a Gaussian distribution. The initial velocities conform to the temperature of interest, 300 K. Independent runs to test the repeatability of the results were

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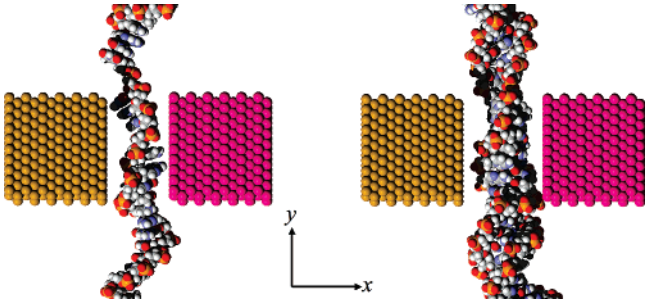

Figure 1. Snapshots from simulations of ssDNA translocation through a 2.0 nm electrode gap (left) and dsDNA through a 2.5 nm gap (right). The ssDNA includes 32 bases with sequence of eight repeated A−G−T−C's. The dsDNA includes 32 base pairs with the same sequence. Only parts of DNA segments are shown. The DNA and electrodes are wrapped in a simulation box filled with water and counterions (a typical simulation box consists of 31 000∼33 000 water molecules). Water molecules and counterions are not shown for clarity. The electric field is applied in the $\pm y$ direction.

performed by varying the random number seeds. More details of the simulation method can be found in the Supporting Information.

It is known that the dielectric properties of synthetic nanopores immersed in solution can be highly nonuniform due to the subsequent rearrangement of solvent and ions in response to the application of a uniform electric field.[20] Here we denote the driving field by the effective transmembrane voltage drop caused by the uniform electric field applied to the system. Given that the membrane thickness is 3 nm, the electric fields used in this study correspond to transmembrane voltage potentials of 150 and 300 mV. Previous experiments indicate that biased voltages of such magnitude are sufficient to drive a ssDNA molecule through a nanopore. For example, Meller et al.[21] demonstrated that DNA electrophoresis through a 1.8-nm protein pore [alpha-hemolysin (α-HL)] can be realized by an electrostatic potential of 70−300 mV through a 5 nm membrane.

The four types of nucleotides of which DNA is composed (A, C, G, and T) have different molecular weights and structures. To probe whether such mass and structural difference can influence the translocation of the DNA, we simulated the electrophoresis of four mononucleotide ssDNA under the same solvent environment and driving field. Each segment is composed of 20 mono-type bases. Namely, they are $polyA_{20}$, $polyC_{20}$, $polyG_{20}$, and $polyT_{20}$, respectively.

We found that, under the same driving force, the average translocation velocities of $polyC_{20}$, $polyT_{20}$, $polyA_{20}$, and $polyG_{20}$ are different and nucleotide dependent. The results are shown in Figure 2 for these four ssDNA migrating through a 2.0 nm wide gap under the influence of a transmembrane voltage of 300 mV in the y direction. The four solid lines represent the separations (Δy) between the center of each ssDNA segment (y_{DNA}) and the gap geometrical center (y_{gap}) along the driving force direction, $\Delta y = y_{DNA} - y_{gap}$. The two horizontal dashed lines denote the region where ssDNA enters and exits the gap. At the start of each simulation ($t = 0$), the first 2 bases of the ssDNA end (C terminus) are placed inside the gap entrance. The system was then relaxed for 0.2 ns. The electric field is turned on at $t = 0.2$ ns and ssDNA begins to translocate through the gap. There is a net charge of $-19|e|$ on each ssDNA. Therefore, the driving force exerted on each DNA molecule from the electric potential is consistently 0.30 nN. The simulations were monitored until the DNA is completely cleared from the nanogap. It can be seen from Figure 2 that the translocation velocities of these four types of nucleotides follow the sequence

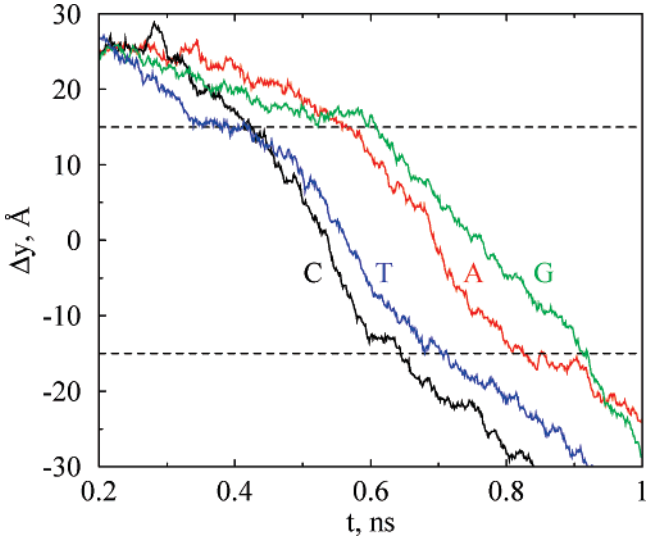

Figure 2. Simulated electrophoresis of four different mono-nucleotide ssDNA through a 2.0 nm gap. Lines from left to right are for $polyC_{20}$, $polyT_{20}$, $polyA_{20}$, $polyG_{20}$, as labeled in the figure. The two dashed lines confine the region inside the gap.

of $polyC_{20} > polyT_{20} > polyA_{20} > polyG_{20}$. The time needed for them to reach the gap entrance is 0.46, 0.49, 0.61, and 0.65 ns, respectively. It took them 0.12, 0.14, 0.16, and 0.21 ns, respectively, to pass the gap region. If each nucleotide is 0.33 nm long (corresponding to the base-to-base distance for standard DNA double helix in B form) along the ssDNA axis, the observed translocation rates correspond to threading about 51, 43, 38, 29 bases/ns for the four ssDNA segments respectively. Simulations have been repeated for three times for each case, and the results are quantitatively consistent within the simulation fluctuations.

The difference in the average translocation velocities for the four ssDNA segments is due to the differences in the molecular weights and the structure of bases C, T, A, and G. The masses of the four ssDNA segments simulated are about 5.8, 6.1, 6.2, and 6.6 kDa, respectively, following the order polyC < polyT < polyA < polyG, which is exactly the reverse order of the average translocation velocities observed. In particular, the molecular weight difference between $polyT_{20}$ and $polyA_{20}$ are smaller compared with that between $polyC_{20}$ and $polyT_{20}$. However, the difference in the time needed for $polyT_{20}$ and $polyA_{20}$ to enter the gap is substantially larger than that for $polyC_{20}$ and $polyT_{20}$ (Figure 2). This suggests that not only the mass differences but also the structural differences in the bases play a role in affecting the average translocation velocity. Indeed, A and G are double-ring structures, whereas C and T have only a single ring. Such structural differences would result in different interaction forces from the solvent when the ssDNA segments are migrating in the aqueous environment. Moving ssDNA in solution at a high speed inevitably involves the rearrangement of water molecules around the bases. For wider A and G bases, this would imply a higher energetic penalty compared to the narrower C and T bases. In turn, their moving speeds will be lower given that the driving force is the same. Further detailed comparative study of the interaction of water molecules with the different bases would be helpful to verify this possibility.

The controllability of DNA translocation is studied by simulations with alternating or intermittent electric fields. In these simulations we used slightly longer ssDNA and dsDNA, with 32 bases or base-pairs of mixed types of bases. The ssDNA consists of eight consecutive A−G−T−C repeats [poly-

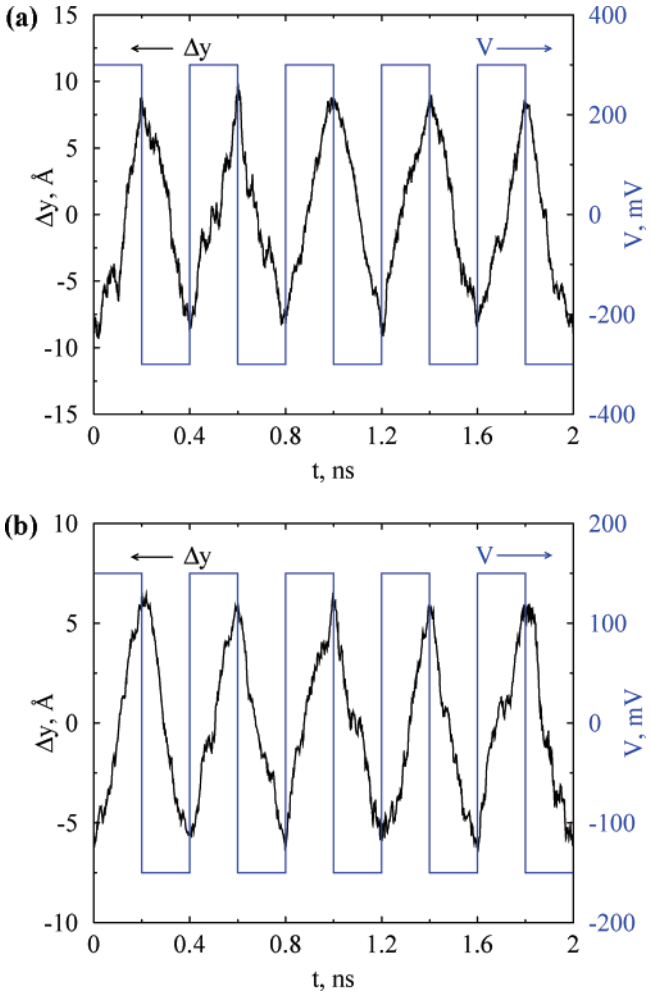

Figure 3. Oscillation of ssDNA (poly(AGTC)$_8$) in a 2.0 nm gap (a) and dsDNA (d-poly(AGTC)$_8$) in a 2.5 nm gap (b) driven by alternating electric fields along the $\pm y$ direction. Black lines correspond to the motion of the DNA middle section (Δy), and blue lines represent the transmembrane voltage potential (V) due to the electric field applied.

(AGTC)$_8$]. The dsDNA has the same sequence and length, but with full double helices. The nanogap width is 2.0 nm for ssDNA translocation, and 2.5 nm for dsDNA translocation. Two ending nucleotides in the C terminus of the DNA were placed inside the gap at the beginning of each simulation. An electric potential of 50−100 mV/nm was applied along the y direction at $t = 0$ to force the ssDNA into the gap. After the ssDNA threads about half way in the nanogap, voltage biases with alternating directions were applied and the trajectory of DNA was monitored. In each case, we have run three independent simulations to test the repeatability of the results.

The center-of-mass trajectories of the DNA middle section as a function of simulation time are presented in Figure 3. The middle section is defined as the 10 nucleotides confined in the gap region when the electric field begins to alternate. For ssDNA poly(AGTC)$_8$, it corresponds to the nucleotides from A_{13} to G_{22}. For dsDNA d-poly(AGTC)$_8$, it corresponds to the base pairs from $A_{13}-T_{52}$ to $G_{22}-C_{43}$. We noticed that the nucleotides outside the gap experience substantial deformation (especially for ssDNA) during the simulations. Therefore, the trajectory of the center of the whole ssDNA does not represent the actually trajectory of the DNA section inside the gap, which is the section of interest. Figure 3 shows the oscillatory behavior of the middle section of ssDNA and dsDNA along the $\pm y$ direction driven by the alternating electric field applied. It can be seen that translocation of ssDNA and dsDNA inside the gap responds instantaneously to the reversal of the electric field direction. Note that in our simulation we assume the switching of electric field direction is instantaneous, whereas in a real device switching will take some finite amount of time, e.g., ~ns or less for a GHz field.

For ssDNA translocation through the 2.0 nm nanogap, the electric field applied has an alternating frequency of 2.5 GHz. The transmembrane voltage potential is ±300 mV. The total driving force on the ssDNA from the electric field is 0.50 nN. The oscillation of ssDNA middle section follows the frequency of the alternating field. The oscillatory amplitude of the ssDNA is about 1.8 nm and remains roughly uniform during a simulation of 2 ns. The oscillatory curves of the ssDNA are very close to linear in each monotonically increasing or decreasing region. Therefore the moving velocity of the ssDNA under the electric field influence is a constant of about 27 bases/ns. Compared to translocation rates of polyC_{20}, polyT_{20}, polyA_{20}, and polyG_{20}, this rate is lower, but in the same order of magnitude.

In the simulations of dsDNA (d-poly(AGTC)$_8$) translocation in a 2.5 nm gap, similar oscillatory behavior was observed. A transmembrane potential drop of 150 mV along $\pm y$ direction was applied. The total electrical driving force on the dsDNA is the same as that on the ssDNA poly(AGTC)$_8$ because dsDNA has twice the net negative charges of that on the ssDNA. The dsDNA inside the gap responds instantaneously to the reversal of driving electric fields, and the oscillation frequency is virtually same as that of the voltage alternating frequency, like that observed in ssDNA simulations. However, the oscillation of dsDNA has a smaller amplitude (about 1.2 nm) than that of ssDNA, due to the heavier molecular weight and larger drag force experienced by the wider dsDNA. The oscillatory curve is roughly linear in each decreasing or increasing region, with a constant velocity of about 18 base pairs/ns.

The observed oscillatory behavior of ssDNA and dsDNA caused by reversing the electric field suggests that the translocation of DNA can be controlled as required if appropriate electric fields are applied to the system. The instantaneous response of DNA molecule to the applied field is particularly appealing since one can control the DNA without having to worry about the response time (also known as dead time) usually involved in control systems.[22]

One possible reason for the instantaneous response of DNA motion to the change in driving force is that the field applied is so strong that it can change the moving direction of ssDNA in a very short time scale; thus, the response observed would effectively look like it is instantaneous. The voltage bias used in this study (150−300 mV across a 3 nm membrane) is significantly large when compared with that used in previous experimental studies. For example, Meller et al.[21] found that a transmembrane potential as small as 70 mV in a 5 nm long pore can drive a DNA segment through a α-HL nanopore. We have performed simulations using a potential drop of 60 mV in the system but could not observe any translocation phenomenon within 15 ns of simulation. Simulations with much longer time scales are desired but limited by available computing power. Therefore, it is unclear if the DNA molecules will still respond to the change of electric field instantaneously if a much lower potential bias is used.

Another possible reason is that the nature of friction between the DNA and the water/nanogap surface is changed to plastic during the translocation. Previous work[23] has shown that confining water between two mica surfaces separated by less than 0.7 nm will result in the water molecules forming solid-

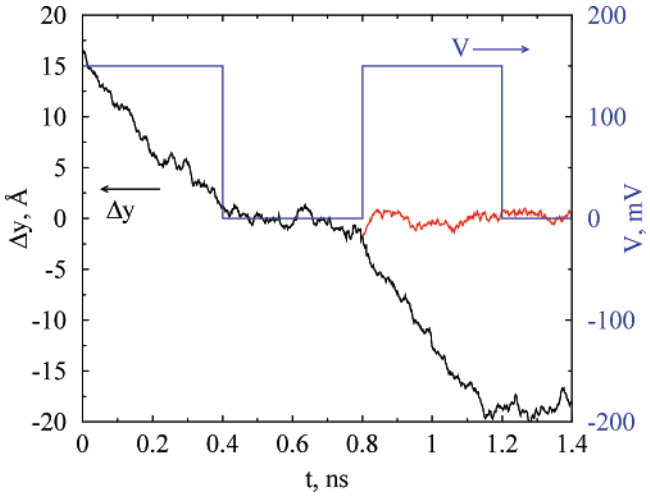

Figure 4. Translocation of ssDNA (poly(AGTC)$_8$) inside a 2.0 nm gap driven by a "braking'' electric field. Black and red lines present the motion of DNA (Δy), and the blue line shows the voltage bias in the y direction (V).

like (bilayer ice) structures. However, in this study, we performed analysis on the water structure inside the gap and found no convincing evidence of solidification. The effective viscosity of the nanoconfined water can be several orders of magnitude above the bulk water value, so that relative movement of the two surfaces confining the water may well be significantly impaired at the nanoscale compared to what might be expected on the basis of bulk water. Structural and tribological phenomena on the nanoscale are complicated and certainly the tribological properties of water nanoconfined between ssDNA or dsDNA and a metal surface have received little attention to date. Hence, it is not conclusive if the nature of the drag/friction force in the gap is altered before more extensive simulations studies are performed for comparison.

Knowing that the translocation of DNA responds instantaneously to the switch of electric driving force, we are further interested in studying the translocation of DNA inside a nanogap when a "braking'' electric field is applied. By "braking'' we mean the application of intermittent electric fields to force the DNA to move along the driving direction for a period and then the turning off of the electric field (pause period) to allow DNA to stop in the middle of the gap for base specific signature detection. This is the desired process for the sequencing device proposed.[6]

Simulation of ssDNA poly(AGTC)$_8$ translocation through a 2.0 nm gap under "braking'' voltage is performed. First, a transmembrane driving potential of 150 mV was applied in the y direction to force the ssDNA into the gap. After the ssDNA translocates about half way in the nanogap, "braking'' is initiated by turning off the field. In Figure 4, we show the trajectory of the ssDNA (black line) under the influence of a "braking'' voltage (blue line). Like that in Figure 3, the y direction motion presented in Figure 4 is for the $A_{13}-G_{22}$ section of the ssDNA poly(AGTC)$_8$. At $t = 0$, the center of the interested section is about 1.6 nm away from the gap center along the y direction. A voltage of 150 mV was applied from $t = 0$ to 0.4 ns. Correspondingly, the DNA was driven into the gap during this time period with a velocity of about 13 bases/ns. From $t = 0.4$ to 0.8 ns, the electric field is turned off. The movement of ssDNA along y direction during this period essentially comes to a halt, which is indicated by a roughly horizontal trajectory line with negligible decrease. At $t = 0.8$ ns, the electric field is turned on again to drive the ssDNA out of the gap. The trajectory represented by the red line after $t = 0.8$ ns shows the movement of $A_{13}-G_{22}$ if no electric field is applied for comparison. The slopes of the trajectories at $t = (0, 0.4)$ and $t = (0.8, 1.2)$ indicate that the velocities of DNA at those two time intervals are about the same. At $t = 1.2$ ns, the electric field is turned off again, and the monitored ssDNA section stops moving again. The movement of the ssDNA section was not monitored after $t = 1.4$ ns since the center of $A_{13}-G_{22}$ had exited the gap completely. The time between the entering ($t = 0.01$ ns) and exiting ($t = 1.06$ ns) of the ssDNA middle section was about 1.05 ns totally, including a 0.4 ns pause when the electric field is turned off.

Additional simulations using short DNA segments, such as segments with 10 or 15 bases (base-pairs), were performed. Qualitatively similar oscillation or pause behavior was observed. However, the fluctuations in the oscillatory amplitudes are much larger than those from simulations with 32 bases. This seems to suggest that longer DNA segments are easier to control than shorter ones in this particular application. Additional studies on this topic are still under way.

In order to sequence the DNA using the experimental device proposed, the ideal translocation velocity of a DNA segment should be in the range of several bases per microsecond, a rate under which the device can have sufficient time to measure the conducting properties across the electrodes.[6,7] We note that the translocation velocity of DNA calculated here is very high compared with that targeted in the experiment. As discussed earlier, in order to obtain nucleotide-specific information at the rates contemplated experimentally, simulations with much longer time scales (~ms) and smaller driving forces must be performed. Given the limitations of current computing power, one possible alternative is to use a coarse-grained method to model the systems to a time scale that is more meaningful for real device development.

In summary, we show through atomistic modeling that translocations of DNA through a nanogap can be tempered by applying the appropriate electric driving force to the system. The response of the DNA movement inside the nanogap to the switch of driving fields is instantaneous. The translocation velocity of a DNA segment is dependent on both the molecular weight and the structure of the nucleotides. "Braking'' the electric driving force can pause the translocation effectively. Although based on simulations of relatively short ssDNA segments, the results from this study can be potentially used in the developments of experimental devices for rapid sequencing of DNA.

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Supporting Information Available: Detailed description of the simulation methodology and the relevant references. This information is available free of charge via the Internet at http://pubs.acs.org.

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