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Electronic detection of dsDNA transition from helical to zipper conformation using graphene nanopores

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Abstract

Mechanical manipulation of DNA, by forced extension, can lead to a structural transformation of double-stranded DNA (dsDNA) from a helical form to a linear zipper-like form. By employing classical molecular dynamics and quantum mechanical non-equilibrium Greens function-based transport simulations, we show the ability of graphene nanopores to discern different dsDNA conformations, in a helical to zipper transition, using transverse electronic conductance. In particular, conductance oscillations due to helical dsDNA vanish as dsDNA extends from helical to zipper form as it is transported through the nanopore. The predicted ability to detect conformational changes in dsDNA, via transverse electronic conductance, can widen the potential of graphene-based nanosensors for DNA detection.

Introduction

Study of the mechanical properties of DNA, enabled by single-molecule experiments, is essential to understanding many key biological processes such as DNA transcription, packing, and replication. The ability to manipulate the structure of DNA at the nanoscale and change DNA electronic and mechanical properties has led to synthetic DNA-based nanodevices [1]. For example, the mechanical structure of a DNA molecule can be manipulated using an atomic force microscope (AFM), where one end of the DNA is tethered to a glass surface and the other end is attached to the AFM cantilever. Displacement of the cantilever exerts a force on the DNA molecule and thereby stretches it [2, 3, 4]. Optical tweezers [5], magnetic tweezers [6] and nanopores [7, 8, 9, 10] likewise can be used to trap DNA molecules and stretch them. Forced extension of double-stranded DNA (dsDNA) transforms its structure from a canonical helical conformation (B-DNA) to a stretched conformation (S-DNA) [11]. The S-DNA can further take a zipper-like form (zip-DNA), where complementary base pairs, on the two strands, are broken and interdigitate giving a zipper-like resemblance [12, 13].

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Graphene, a two-dimensional arrangement of carbon atoms in a honeycomb lattice, is emerging as an attractive candidate for nanopore-based sensors which have the potential to ultimately sequence the human genome [14, 15, 16]. A DNA molecule, driven electrophoretically through the nanopore, transiently blocks the flow of ions leading to a blockade of the ion current. The atomically sharp graphene layer (≈ 3 Å in width) can probe translocating DNA at subnanometer resolution using ion blockade currents. Detection of dsDNA [17, 18, 19] and single-stranded DNA (ssDNA) [20], via ion blockade currents, using graphene nanopores, has been experimentally demonstrated. All-atom molecular dynamics (MD) simulations have been successful in investigating DNA electrophoresis through a wide variety of nanopores [21, 22, 23, 24] including graphene [25, 26].

Graphene is electrically active and can conduct electronic currents. Hence, another opportunity for sequencing DNA is based on transverse electronic currents [27, 28]. First-principle density functional theory (DFT)-based calculations of isolated individual base pairs inside a graphene nanopore have suggested the possibility of base pair discrimination by means of transverse electronic currents [29, 30, 31, 32]. A multi-scale non-equilibrium Greens function (NEGF)-based transport model, accounting for solvent effects, has demonstrated detection of rotational and positional conformation of dsDNA inside a graphene nanopore transistor [33]. Microampere sheet current conduction through graphene nanoribbons (GNR) with small pore diameters (2 nm) has been demonstrated experimentally [34]. Recent experiments have demonstrated furthermore use of electronic sheet currents for the detection of DNA employing a graphene nanopore in the context of a graphene nanoribbon transistor [35].

In the present study, we demonstrate that the change in dsDNA conformation arising in the so-called B-DNA to zip-DNA transition discussed in Introduction can be detected through transverse electronic conductance, which arises around a graphene nanopore translocating the DNA and being employed as a sensor. To achieve such detection, we performed molecular dynamics simulations to induce forced B-DNA to zip-DNA extension in a 15-bp-long helical dsDNA, i.e., B-DNA, and observed a structural transition from helical to zipper form. The conductance in a graphene membrane was shown to be affected by the different dsDNA conformations arising during the B to zip transition. For this purpose the structures arising in the molecular dynamics simulations were used to determine the electrostatic potential around the dsDNA and the result fed into NEGF calculations of the graphene membrane conductance. The conductance changed from an oscillating type response, at the B-DNA stage, to a constant conductance corresponding to the zip-DNA stage. The capability of detecting conformational changes in dsDNA using transverse electronic conductance can complement ionic current measurements and potentially extend the molecular sensing capability of graphene-based nanopores.

Model and Methods

Steered molecular dynamics (SMD) simulations were employed to induce forced extension of dsDNA from B-form to zip-form. The simulations were performed on poly(A-T)₁₅ and poly(G-C)₁₅ strands with the DNA placed in a water box of size 70 Å × 70 Å × 110 Å and neutralized at 1 M KCl, amounting to 357 K⁺ and 327 Cl⁻ ions. The simulated systems

contained about 52,000 atoms. To accommodate the molecular extension along the z -axis the length of the water box was chosen twice as large as the initial length of the DNA, whose axis was aligned along the z -direction. The simulated system is shown in Figure 1a. To gain better sampling of the force-induced extension five independent SMD simulations were performed on both poly(A-T)₁₅ and poly(G-C)₁₅ strands.

All molecular dynamics simulations were carried out using NAMD 2.9 [36], employing periodic boundary conditions, with the CHARMM27 force field parameters for dsDNA [37], ions and employing the TIP3P water model [38]. The dsDNA was built using the program X3DNA [39] and the topology of dsDNA along with the missing hydrogen atoms were generated using psfgen [36]. The system was energy minimized for 4000 steps, then heated to 295 K in 4 ps. A 0.5 ns equilibration under NPT ensemble conditions was performed to equilibrate the system to a desired pressure of 1 atm. This equilibration step was followed by a 1.5 ns equilibration under NVT ensemble condition. A temperature of 295 K was maintained by applying Langevin dynamics with a damping constant of 0.2 ps⁻¹; constant pressure of 1 atm was maintained using a Nosé-Hoover Langevin piston barostat [40] with a period and decay of 200 fs; long-range Coulomb interactions were computed using the particle-mesh Ewald (PME) method with a grid size of < 1 Å; van der Waals interactions were calculated using a 12 Å cutoff; an integration timestep of 1 fs was adopted, with a multiple timestepping algorithm [41, 42] employed to compute the bonded interactions at every timestep, the short-range non-bonded interactions every other timestep, and the long-range electrostatic forces every fourth timestep (employing so-called 1-2-4 timestepping). SMD simulations were then performed by harmonically restraining both, 3' and 5', terminal phosphate atoms on one end, while applying a time-dependent external force to the corresponding atoms at the other end along the z -direction as explained in detail below. A 2 fs integration timestep was adopted for SMD simulations with a 2-1-2 multiple timestepping scheme. The covalent hydrogen bonds in DNA and water were constrained using RATTLE [43] and SETTLE [44] algorithms, respectively. Each independent SMD simulation was 60 ns long, covering a total simulation time of 0.6 μs for five independent trajectories for each poly(A-T)₁₅ and poly(G-C)₁₅.

The electrostatics surrounding the DNA shifted through the graphene nanopore was described using the Poisson-Boltzmann equation

$$\nabla \cdot [\varepsilon(\vec{r}) \nabla \phi(\vec{r})] = -e[K^+(\vec{r}, \phi) - Cl^-(\vec{r}, \phi)] - \rho_{DNA}(\vec{r}) \quad (1)$$

where $\phi(\vec{r})$ is the electrostatic potential and $\varepsilon(\vec{r})$ is the local permittivity. The three-dimensional charge distribution due to the DNA, $\rho_{DNA}(\vec{r})$, was obtained from a prior MD trajectory. The ion solution obeyed Boltzmann statistics, namely [8]

$$K^+(\vec{r}, \phi) = c_0 \exp(-e\phi/k_B T), \quad Cl^-(\vec{r}, \phi) = c_0 \exp(e\phi/k_B T) \quad (2)$$

Here, $K^+(\vec{r})$ and $Cl^-(\vec{r})$ are the spatial ion concentrations, e is the electronic charge and c_0 is the molarity of the KCl solution (1 M). The non-linear equation (1) was solved self-consistently using a Newton multigrid scheme [45] on a 3-dimensional space ($L_x \times L_y \times L_z$).

The DNA charge density was translocated through an armchair edge graphene nanopore (diameter=2.4 nm) and a quantum point contact (QPC) edge graphene nanopore (diameter = 2.4 nm), which were placed at $z = L_z/2$ (see Figure 1b). Dirichlet boundary conditions, $\phi(z = 0) = \phi(z = L_z) = 0$, were assumed along the z -direction, and Neumann boundary conditions, $\nabla_x \phi(x = 0) = \nabla_x \phi(x = L_x) = \nabla_y \phi(y = 0) = \nabla_y \phi(y = L_y) = 0$, were assumed along the x - and y -directions. The Poisson equation (1) was discretized on a non-uniform, rectangular, three-dimensional grid consisting of $256 \times 256 \times 256$ grid points spanning a volume of $10 \times 8 \times 20$ nm³, with the mesh size ranging from $1/3$ Å near the pore mouth to 1 Å far away from the pore. The dielectric constant of water (ϵ_w) and graphene (ϵ_g) were set to 78 and 6, respectively [46]. In the present electrostatic model we have ignored the influence of surface charges on the electrostatic potential induced by the DNA molecule. The graphene membrane, in a practical graphene-based nanopore sensor, is typically sandwiched between oxide layers to insulate the graphene membrane from direct contact with solution [33]. The role of the additional insulating layers as well as the surface charge that spontaneously form on their surface has been investigated in detail elsewhere [47, 48]. The change in the electrostatic potential around the nanopore mouth is $< 5\%$, when the surface charge density changes from $10^{21}/\text{cm}^3$ to $10^{10}/\text{cm}^3$ [8], and ignoring the effect of surface charges is justified. In addition, a gate electrode can be used to adjust the carrier concentration and, thereby, offset the effects of the surface charge on the transverse electronic conductance [48].

The electronic structure of graphene around the nanopore was described using the tight binding Hamiltonian [49]

$$H = \sum_{i,\mu} [\epsilon_\mu - e\phi(\mathbf{r}_i)] a_i^{\mu\dagger} a_i^\mu + \sum_{\substack{\langle ij \rangle \\ \mu\nu}} V_{\mu\nu}(\vec{n}) a_i^{\mu\dagger} b_j^\nu + V_{\nu\mu}(\vec{n}) b_j^{\nu\dagger} a_i^\mu \quad (3)$$

where e_μ is the on-site occupation energy of an electron in state μ located at site i , e is the magnitude of the electronic charge, $\phi(\mathbf{r}_i)$ is the electrostatic potential at site i obtained from the Poisson-Boltzmann equation (1), and Fermion operators $a_i^{\mu\dagger}/b_i^{\mu\dagger}$ and a_i^μ/b_i^μ create and annihilate electrons in state μ at site i for the graphene A/B sublattice, respectively. The states μ, ν are described as superpositions of the p_z , d_{yz} , and d_{zx} orbitals of mono-atomic carbon. The values for all on-site energies and transfer integrals are taken from [49].

The Hamiltonian (Equation 3) can be used to obtain the transmission function $T(E)$, which when integrated over the distribution function $f(E)$, yields the electronic conductance across the conductor at a particular bias V_{DS} [48, 50]

$$G = \frac{2e}{V_{DS} h} \int_{-\infty}^{\infty} \bar{T}(E) [f_1(E) - f_2(E)] dE \quad (4)$$

where $f_\alpha(E) = f(E - \mu_\alpha)$ is the probability that an electron occupies a state at energy E in the lead α , $\mu_1 - \mu_2 = V_{DS}$ is the bias across the conductor. μ_1 is chosen to be equal to 0.0 eV for the QPC edge nanopore and equal to 0.16 eV for the armchair edge nanopore, which results

in maximum sensitivity of the electronic conductance to DNA charges. In our calculations we assumed $f(E)$ to be the Fermi-Dirac distribution, $V_{DS} = 5$ mV and a system temperature of 295 K was adopted.

Results and Discussion

The mechanical response of DNA to external forces can be studied *in silico* using SMD simulations [51, 52]. Earlier simulation studies investigated force-induced stretching of dsDNA by applying a pulling force to the 5' and 3' termini on one end of the dsDNA and by constraining the corresponding termini at the opposite end, causing the strands to stretch parallel to each other and to undergo the B-DNA to zip-DNA transformation [12, 13, 53]. Simulation studies performed under different pulling conditions, like pulling the 3' or 5' termini of both strands in opposite directions or torsionally constraining the dsDNA while pulling, can produce more complex structural changes including local DNA melting [54, 55, 56]. Pulling only one of the termini (3' or 5') and constraining the corresponding termini on the opposite end can lead to strand separation. In fact, various types of SMD simulations have been employed to study the effects of methylation and hydroxymethylation on DNA strand separation [57, 58].

In the present study, dsDNA was stretched by pulling both strands on one end of the dsDNA (atoms colored red in Figure 2) at a constant velocity of 1 Å/ns along the z -direction, while harmonically restraining the other end (atoms colored blue in Figure 2). The pulled atoms were attached to one end of a virtual spring; the other end of the spring, a dummy atom, was moved at a constant pulling speed v (1 Å/ns) along the z -direction. The pulled atoms experience a force $f = -k[z(t) - z(t_0) - v(t - t_0)]$, where $z(t_0)$ is the initial position of the dummy atom attached to the spring. The spring constant k was chosen to be equal to $3k_B T_0 / \text{\AA}^2$ (k_B , Boltzmann constant; $T_0 = 295$ K), which corresponds to a thermal RMSD deviation of $\sqrt{k_B T_0 / k} \approx 0.6$ Å, which is typical for SMD simulations [51, 52, 59, 60]. Figure 2 shows a sequence of snapshots, during forced stretching, of one independent SMD simulation for poly(A-T)₁₅ DNA. The DNA is seen to undergo a series of conformational changes starting from helical form, i.e., B-DNA (marked A in Figure 2) and gradually unwinding itself into planar zipper form, i.e., zip-DNA (marked E in Figure 2). During the A to E transition the hydrogen bonds between complementary base pairs break; the base pairs are seen to interdigitate and finally all stack on top of each other in a zipper like fashion. A 60-ns MD trajectory showing the untwisting and gradual elongation of the DNA from B-form to zip-form can be viewed in Supplementary Movie SM1. Also highlighted in Figure 2 are two sets of base pairs, P-P' and Q-Q', which are spaced half a pitch apart (5 bp apart) from each other. The length of the entire DNA changed from 52 Å to 103 Å over the course of the simulation. The extension of the DNA as a function of time is shown in Supplementary Figure S1.

Figure 3 shows force-extension curves for five independent SMD simulations performed on the poly(A-T)₁₅ DNA strand stretched with a constant pulling speed of 1 Å/ns. At the initial stage, the B-DNA undergoes an elastic transformation, where the force increases gradually from 0 to 100 pN accompanied by a molecular extension of 10 Å. Beyond this extension the

force-extension curve is characterized by a plateau region, where the DNA transforms cooperatively from the helical conformation, i.e., B-DNA to a zipper-like form, i.e., zip-DNA. The transition is characterized by coexistence of helical, stretched and zipper DNA domains. During the initial stages of the transition, a zip-DNA nucleation site appears near the pulling end of the DNA whereas the restrained end retains its helicity. Gradually, the DNA extends and unwinds, with parts of the DNA (middle portion) acquiring a stretched conformation (S-DNA), where the complementary base-pairs partially unwind but are still bound through hydrogen bonds. Eventually the S-DNA and B-DNA domains transform into zip-DNA domains and at an extension of 38 Å the entire DNA transforms into zip-DNA (see inset in Figure 3). The zip-DNA extends elastically beyond 38 Å and is marked by a steep increase in the force experienced by the DNA. The computed peak force (1 nN) experienced by the DNA is much higher than the experimental ones (≈ 150 pN) [2, 3, 4]. The discrepancy is attributed to the pulling speeds employed in simulations (1 Å/ns) which, due to limited computational resources, is much larger than the typical pulling speeds in experiments (1 Å/ μ s).

The evolution of the angle between base pairs P-P' and Q-Q' (see Figure 2), for the poly(A-T)₁₅ case is shown in Figure 4. The two base pairs are a half-pitch apart and the angle is initially, when the DNA assumes a helical form, -180° . After the transition to zip-DNA the base pairs are vertically stacked on top of each other, reducing the angle between the base pairs to 0° . The change in the angle (from -180° to 0°) occurs over a short range of extension, ranging from 10 Å to 25 Å. However, there is a significant variation in the observed dynamics across the five independent simulations. This variation can be attributed to fast pulling employed in the simulation, which does not allow dsDNA's slower degrees of freedom to relax completely during the simulation time covered (60 ns).

Charge distributions corresponding to DNA conformations at five intermediate steps of the B-DNA to zip-DNA transition, A through E in Figure 2, were extracted from the all-atom MD trajectory. The DNA charge distribution, for each of the intermediate stages, was then placed inside a graphene nanopore with a diameter = 2.4 nm, such that the base pair P-P' resides inside the graphene nanopore; in addition, the DNA axis was also aligned with the nanopore axis. These charge distributions were then "translocated" along the $-z$ direction in steps of 0.5 Å, until the base pairs Q-Q' reached the pore. At each step the electrostatic potential induced by the DNA on the graphene nanopore was calculated using the Poisson-Boltzmann approach. The electrostatic potential maps, in the plane of the graphene membrane, altered step-by-step due to DNA translocation, are provided for both B-DNA and zip-DNA cases in Supplementary Figures S4 and S5.

The electrostatic potentials determined according to Equation (1) were then included in the Hamiltonian of the graphene membrane (see Equation (3)) to calculate the resulting conductance across the graphene membrane by means of Equation (4) and as further outlined in Methods. Shown in Figure 5 is the transverse conductance as a function of DNA position inside the nanopore. The DNA inside the graphene nanopore was assumed to be stretched and for this purpose the different intermediate stages (A–E) during the B-DNA to zip-DNA transformation obtained from a SMD simulation performed on poly(A-T)₁₅ were adopted. The calculations described were carried out for two graphene nanopore geometries:

an armchair edge geometry with width of 5 nm, and a quantum point contact (QPC) edge with width of 8 nm. The QPC edge has an irregular edge shape, leading to more stringent boundary conditions for electron transport when compared to the flat armchair edge geometry [33]. (The exact lattice of the armchair and QPC edge are provided in Supplementary Figure S6). As one can see in Figure 5, the conductance varies sinusoidally for stage A DNA translocating through the nanopore, is constant for stage E DNA translocating, and adopts an intermediate variation for stage B, C, D DNA translocating.

Translocation of the DNA segment, comprising of nucleotides between P-P' and Q-Q', in the helical form results in an apparent rotation of the surrounding electric field inside the nanopore (see Supplementary Figure S4). As a result, the transverse electronic conductance in the graphene membrane being induced by the field oscillates. In case of zip-DNA being translocated through the pore the field is rotationally invariant and, as a result, the transverse conductance remains constant (see Supplementary Figure S5). In case of progressive extension of the DNA accompanied during a B-DNA to zip-DNA transition the sinusoidal variation of the transverse conductance diminishes as the translocating DNA passes through the A, B, C, D, and E stages shown in Figure 2. Translocation of the DNA segment, comprised of base pairs between P-P' and Q-Q', is equivalent to a rotation, in the plane of the graphene nanopore, by an amount equal to the angle between the base pairs P-P' and Q-Q' (see Figure 4). Thus, the conductance variation is a measure of the helicity of the DNA and can be used to detect the DNA conformation. The total variation in conductance for the B-DNA is about $1.5 \mu S$ for the arm chair edge¹ and about $10 \mu S$ for the QPC edge². The conductance variation is enhanced for the QPC edge case due to the constriction, which affects the electron transport near the nanopore. In addition, the boundaries also influence the shape of the response which no longer looks perfectly sinusoidal in case of a QPC edge pore.

The conductance variation as a function of DNA extension is shown in Figure 6 for poly(A-T)₁₅ and poly(G-C)₁₅ strands computed at two different KCl molar concentrations, namely, 1 M (Figure 6a) and 0.1 M (Figure 6b). The error bars were obtained from sampling over five independent simulation trajectories for both poly(A-T)₁₅ and poly(G-C)₁₅. The QPC edge, because of its higher sensitivity compared to the armchair edge, was employed in the conductance calculations presented in Figure 6. For both high and low molarity cases, the conductance variation decreases as the DNA transitions from B-DNA to zip-DNA. The uncertainty in the conductance variation is significantly larger for the intermediate stages B, C, and D as compared to stages A (helical) and E (zipper), which can be attributed to the broader distribution in the unwinding pathways sampled by the independent MD simulations.

As discussed above, the conductance variation is a measure of the helicity and there is a large heterogeneity in the helical angle, between P-P' and Q-Q' (see Figure 4), observed in regions, where DNA transitions from B-DNA to zip-DNA. In the present study, limited though by small sampling, conductance variations are indistinguishable for poly(A-T)₁₅ and

¹ $G(\max) - G(\min) + G(\max) - G(0)$

² $G(\max) - G(\min) + G(0) - G(\min)$

poly(G-C)₁₅ cases. The general trend in conductance does not change when the molarity is changed from 1 M to 0.1 M. However, the magnitude of conductance variations is suppressed for the low molarity case, e.g., conductance variation for B-DNA (A in Figure 6) reduces from 10 μ S to 2 μ S for a change in molar concentration from 1 M to 0.1 M. In the low molarity case, due to reduced screening, the average potential induced on the graphene membrane is much larger in magnitude compared to the high molarity case (see Supplementary Figures S7 and S8), which is equivalent to a gating effect and changes the bias point of the QPC significantly [48]. Although reduced screening, at low molar concentrations, increases the magnitude of the potential induced at the pore mouth, the variation in conductance itself is not enhanced.

Conclusion

In summary, we propose a novel detection technique to sense dsDNA in different structural conformations (helical and zipper) by means of transverse electronic conductance in graphene nanopores. The oscillations in transverse electronic conductance arising due to the helical nature of the B-DNA vanish when B-DNA undergoes a structural transition to zip-DNA. Probing the structure of DNA using the transverse electronic conductance through graphene membranes can be a useful tool in the rapidly evolving field of DNA nanosensors. The computational approach used in the present study combines data from extensive classical mechanical all-atom molecular dynamics simulations (0.6 μ s in total) to describe intermediate conformations of dsDNA and from quantum mechanical NEGF-based calculations of transverse electronic conductance in graphene membranes: the two calculations are coupled through embedding the DNA charge distribution resulting from the MD simulation into a Poisson-Boltzmann description of the electrostatic potential in a physiological KCl solution and adding the resulting potential to the quantum mechanical calculation of the graphene sheet current. The results suggest the capability of graphene nanopores to detect conformational changes of dsDNA through measurement of the transverse electronic conductance (sheet current). Such measurement can supplement ionic blockade currents to assess the local conformation of translocating DNA in nanopore sensors. Observed trends in the membrane conductance for dsDNA are predominantly a signature of the backbone, which screens out the response due to the base pairs, making it difficult to observe any sequence dependent (AT/GC) changes. However, the prospects of sequencing ssDNA, where the nucleotide is better exposed in a much narrower pore ($d \approx 1.2$ nm) is being currently investigated.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

1. Bath J, Turberfield AJ. DNA nanomachines. *Nature Nanotechnology*. 2007; 2:275–84.
2. Rief M, Clausen-Schaumann H, Gaub HE. Sequence-dependent mechanics of single DNA molecules. *Nature Structural Biology*. 1999; 6:346–349.
3. Ke C, Humeniuk M, Gracz HS, Marszalek PE. Direct measurements of base stacking interactions in DNA by single-molecule atomic-force spectroscopy. *Physical Review Letters*. 2007; 99:018302. [PubMed: 17678193]
4. Clausen-Schaumann H, Rief M, Tolksdorf C, Gaub HE. Mechanical stability of single DNA molecules. *Biophysical Journal*. 2000; 78:1997–2007. [PubMed: 10733978]
5. Smith SB, Finzi L, Bustamante C. Direct mechanical measurements of the elasticity of single DNA molecules by using magnetic beads. *Science*. 1992; 258:1122–1126. [PubMed: 1439819]
6. Smith SB, Cui Y, Bustamante C. Overstretching B-DNA: The elastic response of individual double-stranded and single-stranded DNA molecules. *Science*. 1996; 271:795–799. [PubMed: 8628994]
7. Keyser UF, Koeleman BN, Van Dorp S, Krapf D, Smeets RM, Lemay SG, Dekker NH, Dekker C. Direct force measurements on DNA in a solid-state nanopore. *Nature Physics*. 2006; 2:473–477.
8. Gracheva ME, Xiong A, Aksimentiev A, Schulten K, Timp G, Lebur-ton J-P. Simulation of the electric response of DNA translocation through a semiconductor nanopore-capacitor. *Nanotechnology*. 2006; 17:622–633.
9. Melnikov DV, Leburton J-P, Gracheva ME. Slowing down and stretching dna with an electrically tunable nanopore in a p-n semiconductor membrane. *Nanotechnology*. 2012; 23:255501. [PubMed: 22652932]
10. Heng JB, Aksimentiev A, Ho C, Marks P, Grinkova YV, Sligar S, Schulten K, Timp G. Stretching DNA using an electric field in a synthetic nanopore. *Nano Letters*. 2005; 5:1883–1888. [PubMed: 16218703]
11. Cluzel P, Lebrun A, Heller C, Lavery R, Viovy J-L, Chatenay D, Caron F. DNA: An extensible molecule. *Science*. 1996; 271:792–794. [PubMed: 8628993]
12. Lohikoski R, Timonen J, Laaksonen A. Molecular dynamics simulation of single DNA stretching reveals a novel structure. *Chemical Physics Letters*. 2005; 407:23–29.
13. Balaeff A, Craig SL, Beratan DN. B-DNA to zip-DNA: Simulating a DNA transition to a novel structure with enhanced charge-transport characteristics. *The Journal of Physical Chemistry A*. 2011; 115:9377–9391. [PubMed: 21598926]
14. Sint K, Wang B, K al P. Selective ion passage through functionalized graphene nanopores. *Journal of the American Chemical Society*. 2008; 130:16448–16449. [PubMed: 19554715]
15. Siwy ZS, Davenport M. Graphene opens up to DNA. *Nature Nanotechnology*. 2010; 5:697–698.
16. Bayley H. Holes with an edge. *Nature*. 2010; 467:164–165. [PubMed: 20829786]
17. Schneider GF, Kowalczyk SW, Calado VE, Pandraud G, Zandbergen HW, Vandersypen LMK, Dekker C. DNA translocation through graphene nanopores. *Nano Letters*. 2010; 10:3163–3167. [PubMed: 20608744]
18. Merchant CA, Healy K, Wanunu M, Ray V, Peterman N, Bartel J, Fischbein MD, Venta K, Luo Z, Johnson ATC, Drndi M. DNA translocation through graphene nanopores. *Nano Letters*. 2010; 10:2915–2921. [PubMed: 20698604]
19. Garaj S, Hubbard W, Reina A, Kong J, Branton D, Golovchenko JA. Graphene as a subnanometre trans-electrode membrane. *Nature*. 2010; 467:190–193. [PubMed: 20720538]
20. Garaj S, Liu S, Golovchenko JA, Branton D. Molecule-hugging graphene nanopores. *Proceedings of the National Academy of Sciences, USA*. 2013; 110:12192–12196.
21. Mathé J, Aksimentiev A, Nelson DR, Schulten K, Meller A. Orientation discrimination of single stranded DNA inside the α -hemolysin membrane channel. *Proceedings of the National Academy of Sciences, USA*. 2005; 102:12377–12382.
22. Aksimentiev A, Schulten K. Imaging alpha-hemolysin with molecular dynamics: Ionic conductance, osmotic permeability and the electrostatic potential map. *Biophysical Journal*. 2005; 88:3745–3761. [PubMed: 15764651]

23. Aksimentiev A, Heng JB, Timp G, Schulten K. Microscopic kinetics of DNA translocation through synthetic nanopores. *Biophysical Journal*. 2004; 87:2086–2097. [PubMed: 15345583]
24. Cruz-Chu ER, Ritz T, Siwy ZS, Schulten K. Molecular control of ionic conduction in polymer nanopores. *Faraday Discussion*. 2009; 143:47–62.
25. Sathe C, Zou X, Leburton J-P, Schulten K. Computational investigation of DNA detection using graphene nanopores. *ACS Nano*. 2011; 5:8842–8851. [PubMed: 21981556]
26. Wells DB, Belkin M, Comer J, Aksimentiev A. Assessing graphene nanopores for sequencing DNA. *Nano Letters*. 2012; 12:4117–4123. [PubMed: 22780094]
27. Zwolak M, Ventra MD. Electronic signature of DNA nucleotides via transverse transport. *Nano Letters*. 2005; 5:421–424. [PubMed: 15755087]
28. Postma HWC. Rapid sequencing of individual DNA molecules in graphene nanogaps. *Nano Letters*. 2010; 10:420–425. [PubMed: 20044842]
29. Nelson T, Zhang B, Prezhdov OV. Detection of nucleic acids with graphene nanopores: Ab initio characterization of a novel sequencing device. *Nano Letters*. 2010; 10:3237–3242. [PubMed: 20722409]
30. Prasongkit J, Grigoriev A, Pathak B, Ahuja R, Scheicher RH. Transverse conductance of DNA nucleotides in a graphene nanogap from first principles. *Nano Letters*. 2011; 5:1941–1945. [PubMed: 21495701]
31. Saha KK, Drndić M, Nikolić BK. DNA base-specific modulation of microampere transverse edge currents through a metallic graphene nanoribbon with a nanopore. *Nano Letters*. 2012; 12:50–55. [PubMed: 22141739]
32. Avdoshenko SM, Nozaki D, Gomes da Rocha C, González JW, Lee MH, Gutierrez R, Cuniberti G. Dynamic and electronic transport properties of DNA translocation through graphene nanopores. *Nano Letters*. 2013; 13:1969–1976. [PubMed: 23586585]
33. Girdhar A, Sathe C, Schulten K, Leburton J-P. Graphene quantum point contact transistor for DNA sensing. *Proceedings of the National Academy of Sciences, USA*. 2013; 110:16748–16753.
34. Puster M, Rodríguez-Manzo JA, Balan A, Drndić M. Toward sensitive graphene nanoribbon-nanopore devices by preventing electron beam-induced damage. *ACS Nano*. 2013; 7:11283–11289. [PubMed: 24224888]
35. Traversi F, Raillon C, Benameur S, Liu K, Khlybov S, Tosun M, Krasnozhan D, Kis A, Radenovic A. Detecting the translocation of DNA through a nanopore using graphene nanoribbons. *Nature Nanotechnology*. 2013; 8:939–945.
36. Phillips JC, Braun R, Wang W, Gumbart J, Tajkhorshid E, Villa E, Chipot C, Skeel RD, Kale L, Schulten K. Scalable molecular dynamics with NAMD. *Journal of Computational Chemistry*. 2005; 26:1781–1802. [PubMed: 16222654]
37. MacKerell AD Jr, Bashford D, Bellott M, Dunbrack RL Jr, Evanseck JD, Field MJ, Fischer S, Gao J, Guo H, Ha S, Joseph D, Kuchnir L, Kuczera K, Lau FTK, Mattos C, Michnick S, Ngo T, Nguyen DT, Prodhom B, Reiher IWE, Roux B, Schlenkerich M, Smith J, Stote R, Straub J, Watanabe M, Wiorkiewicz-Kuczera J, Yin D, Karplus M. All-atom empirical potential for molecular modeling and dynamics studies of proteins. *Journal of Physical Chemistry B*. 1998; 102:3586–3616.
38. Jorgensen WL, Chandrasekhar J, Madura JD, Impey RW, Klein ML. Comparison of simple potential functions for simulating liquid water. *Journal of Chemical Physics*. 1983; 79:926–935.
39. Lu X-J, Olson WK. 3DNA: a software package for the analysis, rebuilding and visualization of three-dimensional nucleic acid structures. *Nucleic Acids Research*. 2003; 31:5108–5121. [PubMed: 12930962]
40. Martyna GJ, Tobias DJ, Klein ML. Constant pressure molecular dynamics algorithms. *Journal of Chemical Physics*. 1994; 101:4177–4189.
41. Grubmüller H, Heller H, Windemuth A, Schulten K. Generalized Verlet algorithm for efficient molecular dynamics simulations with long-range interactions. *Molecular Simulation*. 1991; 6:121–142.
42. Schlick T, Skeel R, Brünger A, Kalé L, Board JA Jr, Hermans J, Schulten K. Algorithmic challenges in computational molecular biophysics. *Journal of Computational Physics*. 1999; 151:9–48.

43. Andersen HC. Rattle: A “velocity” version of the shake algorithm for molecular dynamics calculations. *Journal of Chemical Physics*. 1983; 52:24–34.
44. Miyamoto S, Kollman PA. SETTLE: An analytical version of the SHAKE and RATTLE algorithm for rigid water molecules. *Journal of Computational Chemistry*. 1992; 13:952–962.
45. Press, WH.; Teukolsky, SA.; Vetterling, WT.; Flannery, BP. *The Art of Scientific Computing*. 2. Cambridge University Press; New York, NY, USA: 1992. Numerical Recipes in C.
46. Hwang EH, Das Sarma S. Dielectric function, screening, and plasmons in two-dimensional graphene. *Phys Rev B*. 2007; 75:205418.
47. Gracheva ME, Leburton J-P. Electrolytic charge inversion at the liquid/solid interface in a nanopore in a doped semiconductor membrane. *Nanotechnology*. 2007; 18:145704.
48. Girdhar A, Sathe C, Schulten K, Leburton J-P. Gate-modulated graphene quantum point contact device for DNA sensing. *Journal of Computational Electronics*. 2014
49. Boykin TB, Luisier M, Klimeck G, Jiang X, Kharche N, Zhou Y, Nayak SK. Accurate six-band nearest-neighbor tight-binding model for the π -bands of bulk graphene and graphene nanoribbons. *Journal of Applied Physics*. 2011; 109
50. Datta, S. *Quantum Transport: Atom to Transistor*. Cambridge University Press; 2013.
51. Sotomayor M, Schulten K. Single-molecule experiments in vitro and in silico. *Science*. 2007; 316:1144–1148. [PubMed: 17525328]
52. Lee EH, Hsin J, Sotomayor M, Comellas G, Schulten K. Discovery through the computational microscope. *Structure*. 2009; 17:1295–1306. [PubMed: 19836330]
53. Santosh M, Maiti PK. Force induced DNA melting. *Journal of Physics: Condensed Matter*. 2009; 21:034113.
54. Lebrun A, Lavery R. Modelling extreme stretching of DNA. *Nucl Acids Res*. 1996; 24:2260–2267. [PubMed: 8710494]
55. Harris SA, Sands ZA, Laughton CA. Molecular dynamics simulations of duplex stretching reveal the importance of entropy in determining the biomechanical properties of DNA. *Biophysical Journal*. 2005; 88:1684–1691. [PubMed: 15626714]
56. Luan B, Aksimentiev A. Strain softening in stretched DNA. *Phys Rev Lett*. 2008; 101:118101. [PubMed: 18851334]
57. Severin PM, Zou X, Gaub HE, Schulten K. Cytosine methylation alters DNA mechanical properties. *Nucleic Acids Research*. 2011; 39:8740–8751. [PubMed: 21775342]
58. Severin PM, Zou X, Schulten K, Gaub HE. Effects of cytosine hydroxymethylation on DNA strand separation. *Biophysical Journal*. 2013; 104:208–215. [PubMed: 23332073]
59. Isralewitz B, Gao M, Schulten K. Steered molecular dynamics and mechanical functions of proteins. *Current Opinion in Structural Biology*. 2001; 11:224–230. [PubMed: 11297932]
60. Gao M, Sotomayor M, Villa E, Lee E, Schulten K. Molecular mechanisms of cellular mechanics. *Physical Chemistry - Chemical Physics*. 2006; 8:3692–3706. [PubMed: 16896432]

Figure 1.

Model of the simulated system. (a) All-atom MD model comprising of a 15-bp-dsDNA, K^+ and Cl^- ions, and water box. The dimensions of the simulated periodic cell were $70 \text{ \AA} \times 70 \text{ \AA} \times 110 \text{ \AA}$. (b) Schematic of graphene nanopore system used to calculate the transverse electronic conductance. Shown in the figure is the snapshot of a DNA conformation that arose during one of the MD simulations. The DNA being extended was placed after the actual MD simulations inside a QPC edge nanopore to mimic an actual translocation process through the pore. The electrostatic potential surrounding the DNA was calculated and fed into a NEGF calculation of the transverse electronic conductance induced by the potential in the graphene membrane. A pore diameter of 2.4 nm was assumed.

Figure 2.

Five representative snapshots (A–E) from a single SMD trajectory (see Supplementary Movie SM1) of poly(A-T)₁₅ DNA during a B-DNA (A) to zip-DNA (E) transition. The atoms colored in red were pulled in the *z*-direction at a rate of 1 Å/ns; the blue colored atoms were harmonically restrained to the initial positions. Also shown is the evolution of two sets of base pairs, P-P' and Q-Q', which are a half pitch (namely 5 bp) apart. The black arrows, joining P to P' and Q to Q', initially pointing in opposite directions corresponding to a pure helical conformation (B-DNA) align themselves in the same direction once the zipper conformation (zip-DNA) is reached. The numbers below each snapshot represent the corresponding molecular extension.

Figure 3.

Force-extension curves for poly(A-T)₁₅ DNA. Shown are the force-extension curves that resulted from five independent SMD simulations, Sim 1–5, performed at a pulling speed of 1 Å/ns. The force-extension curve begins with a region corresponding to the elastic extension of B-DNA followed by a B-DNA to zip-DNA transition plateau. In the region beyond the plateau the zip-DNA undergoes elastic extension, which is characterized by a sharp linear increase in force. The inset shows the zip-DNA conformation at the end of the transition plateau. Supplementary Figure S2 shows the force-extension curves for the poly(G-C)₁₅ strand.

Figure 4.

Evolution of the angle between base pairs P-P' and Q-Q' (see Figure 2) for five independent SMD simulations, Sim 1–5, performed on poly(A-T)₁₅ DNA; the angle changes from -180° to 0° as the DNA segment between P-P' and Q-Q' transitions from helical to zipper form. Supplementary Figure S3 shows the corresponding curve for the poly(G-C)₁₅ strand.

Figure 5.

Transverse electronic conductance as a function of poly(A-T)₁₅ DNA position (snapshot) for (a) graphene nanopore with armchair edge, and (b) graphene nanopore with QPC edge. Base pairs P-P' were initially aligned with the nanopore, and subsequently translocated at a rate of 0.5 Å, along $-z$ direction, per snapshot until base pairs Q-Q' reached the pore. The transverse electronic conductance changes from an oscillating type response, corresponding to B-DNA (A), to a constant conductance when the DNA adopts a zipper-like conformation, i.e., zip-DNA (E). Sinusoidal variation in the transverse electronic conductance diminishes as the DNA passes through the intermediate stages B, C, and D. A QPC edge geometry shows larger variations in transverse electronic conductance when compared to the armchair edge geometry.

Figure 6.

Variation in the transverse electronic conductance as a function of DNA extension for a QPC edge graphene nanopore. Shown in (a) and (b) are conductance variation, for the stages A, B, C, D, and E (see Figure 2) arising in the B-DNA to zip-DNA transition corresponding to KCl molar concentrations of 1 M and 0.1 M, respectively. The error bars are obtained from sampling over five independent force-extension simulations performed on poly(A-T)₁₅ as well as on poly(G-C)₁₅ strands.