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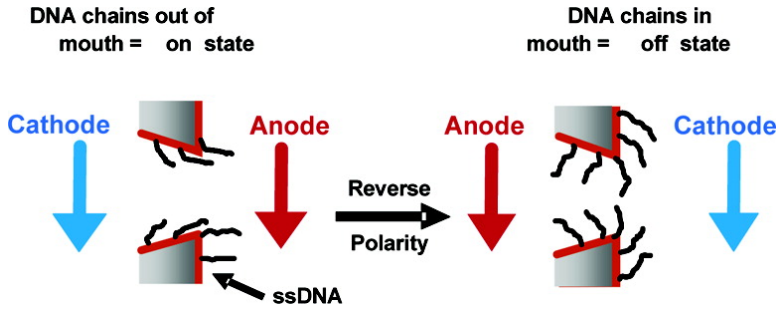


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# DNA−Nanotube Artificial Ion Channels

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There is considerable interest in developing chemical devices that mimic the function of biological ion channels.[1] We recently described such a device, which consisted of a single conically shaped gold nanotube embedded within a polymeric membrane.[1e] This device mimicked one of the key functions of voltage-gated ion channels: the ability to strongly rectify the ionic current flowing through it. The data obtained were interpreted using a simple electrostatic model.[1e]

While the details are still being debated,[2a−c] it is clear that ion-current-rectification in biological ion channels is more complicated and involves physical movement of an ionically charged portion of the channel in response to a change in the transmembrane potential.[2d] We report here artificial ion channels that were designed to rectify the ion current flowing through them via this "electro-mechanical"[3] mechanism. These artificial channels are also based on conical gold nanotubes, but with the critical electromechanical response provided by single-stranded DNA molecules attached to the nanotube walls.

Single conically shaped nanopores were etched into 0.07 $cm^2$ samples of a 12 $\mu$m thick polycarbonate membrane.[4] For most of the studies reported here, the large diameter opening of the pore was 5 $\mu$m (Figure 1A), and the small diameter opening was 60 nm (Figure 1B). An electroless plating method[5] was used to deposit a correspondingly conical gold nanotube within the pore. This entails coating the pore walls and membrane faces with a thin (∼10 nm) layer of gold. Because this layer is so thin, the large diameter opening of the conical gold nanotube remained 5 $\mu$m. An approximate measure of the diameter of the small opening, or "mouth", of the conical nanotube was obtained using a simple electrochemical method[4] (Table 1).

The final step is to covalently attach single-stranded DNA molecules to the gold surfaces. This was accomplished by using the following thiol-terminated DNAs: 5′HS-$(CH_2)_6$-CGAGTCCATTCA3′ (12-mer), 5′HS-$(CH_2)_6$-GACCGAGTCCATTCA3′ (15-mer), 5′HS-$(CH_2)_6$-CGCGAGAAGTTACATGACCTGTAGACGATC3′ (30-mer), 5′HS-$(CH_2)_6$-$A_{45}$3′ (45-mer), and 5′HS-$(CH_2)_6$-CGCGAGAAGTTACATGACCTGTAGCTCGCG3′ (30-mer hairpin).[6] In all of the experiments described here, the conically shaped gold nanotube was left embedded within the polycarbonate membrane. However, to prove that this nanotube is, indeed, conical, the membrane can be dissolved away and the liberated nanotube imaged via electron microscopy (Figure 1C).

Current−voltage ($I-V$) curves for these artificial ion channels were obtained by mounting the membrane sample between the two halves of a U-tube conductivity cell.[4] Each half-cell was filled with ∼5 mL of a 10 mM, pH 7, phosphate buffer that was also 100 mM in KCl. A Ag/AgCl reference electrode was inserted into each half-cell solution, and a Keithley instruments 6487 picoammeter/voltage source was used to apply the desired transmembrane potential and measure the resulting ionic current flowing through the gold nanotube.

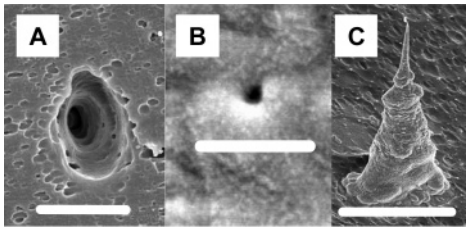


***Figure 1.*** Electron micrographs showing (A) large-diameter (scale bar = 5.0 $\mu$m) and (B) small-diameter (scale bar = 333 nm) opening of a conical nanopore, and (C) a liberated conical Au nanotube (scale bar = 5.0 $\mu$m).

***Table 1.*** Nanotube Mouth Diameter ($d$), DNA Attached, $r_{max}$, Radius of Gyration of DNA ($r_g$), and Extended Chain Length ($l$)

| $d$ (nm) | DNA attached | $r_{max}$ | $r_g$ (nm) | $l$ (nm)[a] |
|---|---|---|---|---|
| 41 | 12-mer | 1.5 | 1.4 | 5.7 |
| 46 | 15-mer | 2.2 | 1.6 | 6.9 |
| 42 | 30-mer | 3.9 | 2.9 | 12.9 |
| 38 | 45-mer | 7.1 | 4.0 | 18.9 |
| 98 | 30-mer | 1.1 | 2.9 | 12.9 |
| 59 | 30-mer | 2.1 | 2.9 | 12.9 |
| 39 | 30-mer | 3.9 | 2.9 | 12.9 |
| 27 | 30-mer | 11.5 | 2.9 | 12.9 |
| 13 | 30-mer | 4.7 | 2.9 | 12.9 |
| 39 | 30-mer hairpin | 1.4 | n/a | 6.9 |

[a] Includes the $(CH_2)_6$ spacer.

In our prior work, rectification was observed because the radius of the nanotube's mouth (5 nm) was comparable to the thickness of the electrical double layer associated with the fixed surface charge (due to adsorbed $Cl^-$) on the nanotube walls.[1e] Put another way, rectification was observed because such small mouth-diameter nanotubes are cation permselective.[1e,f] Because the nanotubes used here have much larger mouth diameters (Table 1), cation permselectivity is not observed. This is proven by the fact that without attached DNA, these nanotubes do not rectify, even though there is adsorbed $Cl^-$ on the nanotube walls (Figure 2A). Furthermore, we have used the Goldman−Hodgkin−Katz[7] method to show that even with the adsorbed DNA, such large mouth-diameter nanotubes are not permselective.[6] However, the DNA-containing nanotubes rectify the ion current; that is, they show an on-state at negative transmembrane potentials (anode facing the mouth of nanotube, Figure 2B) and an off-state at positive potentials (Figure 2C).

The extent of rectification may be quantified via the ratio, $r_{max}$, which is the absolute value of the current at −1 V (on-state) divided by the current at +1 V (off-state) (Table 1). The first four entries in Table 1 correspond to the nanotubes in Figure 2A. As is evident from the figure, $r_{max}$ increases with DNA chain length. The extent of rectification can also be controlled by holding the chain length constant and by varying the diameter of the nanotube mouth (Table 1). With one exception (vide infra), $r_{max}$ increases with decreasing mouth diameter.


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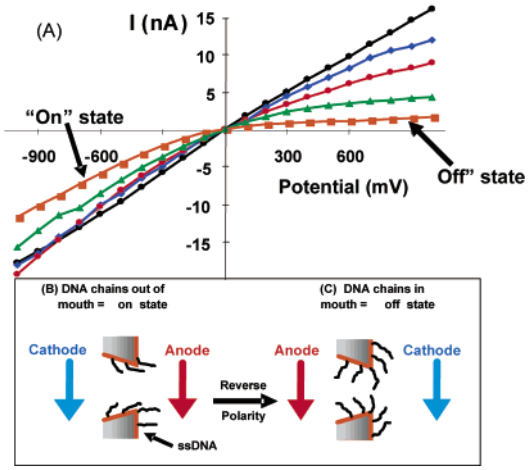


***Figure 2.*** (A) $I-V$ curves for nanotubes with a mouth diameter of 40 nm containing no DNA (black) and attached 12-mer (blue), 15-mer (red), 30-mer (green), and 45-mer (orange) DNAs. (B and C) Schematics showing electrode polarity and DNA chain positions for on (B) and off (C) states.

We propose that rectification in these nanotubes entails electrophoretic insertion of the DNA chains into (off-state, Figure 2C) and out of (on-state, Figure 2B) the nanotube mouth. The off-state is obtained because when inserted into the mouth, the chains partially occlude the pathway for ion transport, yielding a higher ionic resistance for the nanotube. There is ample evidence in the literature to support this hypothesis. First, because DNA chains are anionic, they can be driven electrophoretically through nanopores, and during translocation, occlusion of the nanopore by the DNA causes a transient increase in the pore's ionic resistance.[8] Furthermore, in complete analogy to our model, if one end of a DNA chain is immobilized in an electrophoresis experiment, the chain extends linearly in the direction of the anode.[9] In addition, simulations of the electric field strength in conical nanopores, identical to those used here, show that the field in the electrolyte solution in the mouth of the nanopore is $1 \times 10^6$ V m$^{-1}$ when the total voltage drop across the membrane is only 1 V.[10] This focusing of the electric field strength at the nanotube mouth means that there is ample field to extend the DNA chains toward the anode.

Further support for this model can be garnered from our experimental $I-V$ data. First, the magnitudes of the on-state currents in Figure 2A decrease with increasing DNA chain length. This is because even in the on-state, the DNA chains partially occlude the mouth of the nanotube and increase the nanotube resistance. Second, while the general trend is that $r_{max}$ increases with decreasing mouth diameter, one exception was noted; the nanotube with the ~13 nm mouth and the 30-base DNA rectifies less than does the nanotube with the ~27 nm mouth and this same 30-base DNA (Table 1). These data suggest that the DNA chain can be too long, relative to the mouth diameter, to allow for efficient rectification. Table 1 lists two relevant lengths of the DNA chains, the length if the chain were completely extended[8] and the radius of gyration[11] for the globular form. According to our model, in order to rectify, the chain must have the freedom to extend linearly toward the anode. The nanotube with the 13 nm mouth is the only case where the length of the extended DNA chain is equivalent to the mouth diameter. We suggest that this makes it sterically difficult for the chains to reorient and insert themselves into the mouth, and this is why relatively poor rectification is observed.

Third, according to our model, the DNA chain must have the flexibility to extend in the direction of the anode. This issue was explored by comparing the extent of rectification for a nanotube containing the conventional 30-base DNA and that of an identical tube containing a hairpin 30-base DNA. The hairpin DNA[12] folds back on itself because the bases at one end of the chain are complementary to the bases at the other end, making the chain much less flexible. The nanotube containing this 30-base hairpin DNA is a very poor rectifier, much worse than the same nanotube with the conventional 30-base DNA (Table 1).

We have described here the first example of artificial ion channels where the extent of rectification can be controlled at will by either a simple chemical method (varying the DNA chain length) or a simple physical method (varying the nanotube mouth diameter). With regard to the chemistry, our model suggests that nanotubes containing fixed, flexible polycations would rectify with opposite polarity as the DNA-based channels discussed here, and we have found this to be true.[6]

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**Supporting Information Available:** Attachment of DNA chains to the Au surface and artificial ion channels modified with polylysine that rectify with opposite polarity as the DNA-based channels (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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