

Electroosmosis-Driven Polystyrene Particles Transport Across Polymer Membrane Containing a Conical-Shaped Nanopore

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This report presents a study of translocation of the charged polystyrene nanoparticles across two nanopores separately (a double conical-shaped and a single conical-shaped) contained within a polyimide (Kapton) film using the coulter Counter principle (or “resistive-pulse” method) in which the time-dependent current is recorded as the nanoparticle is driven across the membrane. The nanopores were fabricated by the track-etched method. Under the action of the electroosmotic, the particles transports across the nanopore and results in a direction-dependent and asymmetric triangular-shaped resistive pulse. When applied voltage was low, the transportation current decreased. With the increasing of the voltage, the transmembrane current decreased and increase alternately, the increase of current was less and the decrease of current was more. At higher voltage the current decreased. We contribute these to the change of the electroosmotic flow.

Keywords: Nanopore, Electroosmotic flow, Polystyrene particles, Ion transport.

INTRODUCTION

Nanopore single molecule detection is one of the research hotspots in recent years. It has been widely used in detection of various biological molecule and chemical analysis¹⁻³. The nanopore single molecule DNA sequencing technology was considered as the promising third generation sequencing technology by virtue of its advantage of fast detection and low cost and it is being drawn more and more attentions in the research fields about advanced DNA sequencing⁴.

According to the different attribute of nanopore, nanopore single molecule detection mainly focus on biological nanopore and synthetic nanopore. Compared to biological nanopore, synthetic nanopore is more stable and the pore size is adjustable. So, synthetic nanopore could be used in wider range of molecular detection. In recent years, because of the greatly improved detection sensitivity as the sizes of synthetic pores and channels are reduced to dimensions that are comparable to those of nanoparticles and molecules, synthetic pores and channels are widely used for sensing molecules, polymers and in the structural analysis of biopolymers⁵⁻⁸. DeBlois and Wesley⁹ reported that 60 nm diameter viruses may be rapidly sized in their natural hydrated state as they pass through a single pore. More recently, the crooks group reported Si₃N₄/PDMS membranes containing individual multiwalled carbon nanotubes (about

65 nm in radius). Saleh and Sohn¹⁰ utilized microfabricated nanopores/channels in quartz or PDMS for counting particles as small as 43 nm in radius (about 10⁸ particles/mL) and for the detection of antibody binding to latex colloids. Thornton *et al.*¹¹ has used electron beam lithography to fabricate cylindrical solid-state nanopores with diameters as small as 20 nm, capable of detecting polystyrene particles by the Coulter counting technique. Zhang *et al.*¹² also reported the analytical application of cylindrical-shaped silica nanochannels for sensing single 40 nm polystyrene particles and double-stranded DNA.

In order to have a more in-depth interpretation, much work also has been carried out on mimicing the complex chemo-electrical control that is observed in protein channels. Lan *et al.*¹³ investigated the pressure-driven translocation of nanoparticles across a conical-shaped glass nanopore and found that there was a linear dependence between the particle translocation frequency and the applied pressure. Holden *et al.*¹⁴ described the pressure-driven deformation and translocation of 570 nm radius soft microgel particles through glass nanopore membranes with radii as small as 375 nm. James *et al.*¹⁵ investigated ionic transport through nanopores in Si membranes and observed that rectification ratios could be enhanced by a factor of over 100 by voltage gating alone.

Most of the investigation focused on nanopores fabricated on inorganic matter. In this paper, we used a track-etched

double/single conical-shaped polymer nanopore to investigate the nanoparticle transmission property driven by electroosmotic flow. The track-etched method can fabricate nanopores in polymer materials, especially the geometry of the nanopores, including the pore-tip diameter and the cone angle, which can be controlled by monitoring the etching process. Because this method is relatively simple and the polymer materials surface is rich in many kinds of chemical active group, can be further functionally modified. In recent years, the nanopores fabricated by track-etched method have showed unique advantage in nucleic acid molecule sequencing and protein molecule detection.

EXPERIMENTAL

Fabrication of the nanopores: The nanopores we used in this paper were prepared in a 12 μm thick foil of polyimide (Kapton). The foil was irradiated with a single heavy ion (Au) with an energy of 11.4 MeV nucleon⁻¹ at the UNILAC linear accelerator (GSI, Darmstadt, Germany). The foil was mounted into a home-made cell that had a chamber on each side of the foil. The etchant (13 % Cl NaClO) was filled in the chambers. Two platinum electrodes connected with a Keithley 6487 picoammeter were plugged into the solutions in each chamber. Supplying 1V voltage across the foil, we monitored the current from the picoammeter. The temperature according to the etching was maintained at 40 °C. At the moment that the pore was etched through, a big jump of current was observed. This method produced a double conical nanopore on the foil. We call the the larger opening of the conical nanopore the base and the smaller one the tip. The diameter of the base (D_{base}) was calculated by multiplying the bulk etching rate and the etching time. The diameter of the tip (D_{tip}) was estimated by an electrochemical method: $D_{\text{tip}} = 4LI/\pi KUD_{\text{base}}$, where L is the length of the nanopore, I is the ion current given an applied voltage U and K is the conductance of the solution we used for this measurement (typically 1 M KCl). The inner surface of the nanopore prepared by the above technique is negatively charged. The D_{base} and D_{tip} of the nanopore used in this measure were 1500 and 37 nm, respectively. The method of the single conical-shaped nanopore is fabricated by asymmetric track-etched method, two chambers filled with etchant (13 % Cl NaClO) and stopping medium (1 M KI) separately, the rest is the same as above method. The D_{base} and D_{tip} of the nanopore used in this measure were 3372 and 43 nm, respectively.

Comparing the single conical-shaped nanopore, the conical angle of the double conical-shaped nanopore is bigger, the electric field is more homogeneous, so it's easy to translocate through for the particles.

Ion current recordings: A foil with a conical nanopore on it was mounted into the center of our home-made cell. The chambers were filled with 0.1 M KCl solution, for three pH values (2.02, A3.7 and 5.9), polystyrene (PS) nanoparticles (diameter = 25 nm) were uniformly dispersed into the solution in one chamber. For the single conical nanopore, the chamber is close to the tip. A potential difference was applied across the foil using Ag/AgCl electrodes.

The direction of the voltage bias is as shown in the Fig. 1.

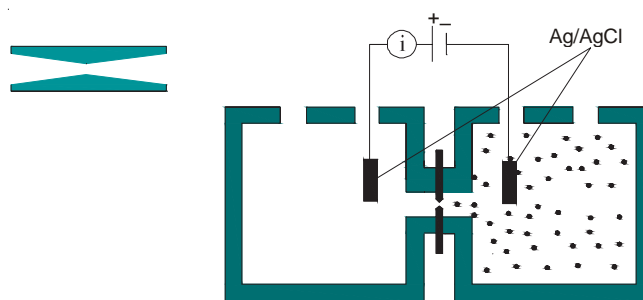


Fig. 1. Schematic illustration of the voltage-driven particle across a double conical-shaped nanopore analysis system. The insert shows the schematic of the double conical nanopore

The ion current was acquired by a patch clamp (Axon 200 B, Molecular Devices Inc.). The patch clamp was working in voltage-clamp mode and a low pass Bessel filter of 100 Hz was used. Then, the current signal was digitized by Digidata 1322A (Molecular Devices Inc.) at 2 kHz and followed by Clampex 9 with further analysis. Milli-Q water (18.2 M Ω) was used for preparing all the solutions used in our experiments. The voltage supplied across the foil was scanned from -0.3 to +0.3 V with a step of 0.1 V. A schematic illustration of the experimental setup is presented in Fig. 1. Particles are placed in solution on the base side and are driven through the double conical nanopore into the solutions on the base side by applying a voltage. The insert is the scheme of a double conical-shaped nanopore.

RESULTS AND DISCUSSION

Polystyrene particles transport the single conical nanopore: The surface charge of the track-etched Kapton nanopore can be adjusted by varying the pH values, low pH decreases density of negative charge. For the pH 3.7, 2.02, the surface charge on the pore wall is neutral and positive, respectively. Fig. 2 showed that the current was stable from -1000 to 1000 mV, revealing there wasn't particle transport across the nanopore. As shows in Fig. 3, at the pH value 3.7, there were resistant pulses when applied voltage exceeded -100 mV. It claimed that polystyrene particles transported across the nanopore driven by the electrophoresis. At the pH value 2.02, direction of the electroosmotic flow is from the negative to

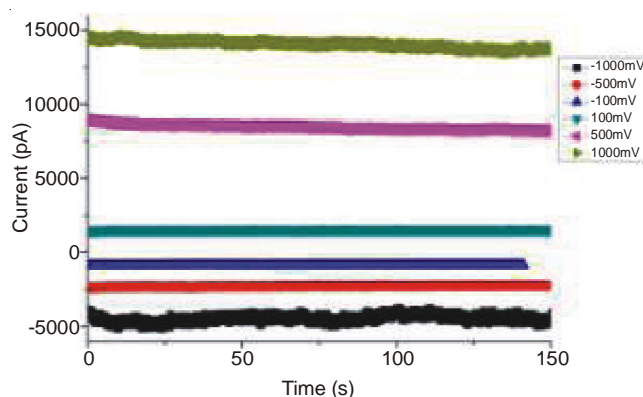


Fig. 2. i-t Recordings corresponding to the translocation of 25 nm radius polystyrene particles at different voltage using a 43 nm diameter single conical-shaped nanopore in a 0.1 M KCl solution at the pH value 2.02. The voltage from bottom to tip in order is: -1000, -500, -100, 100, 500 and 1000 mV

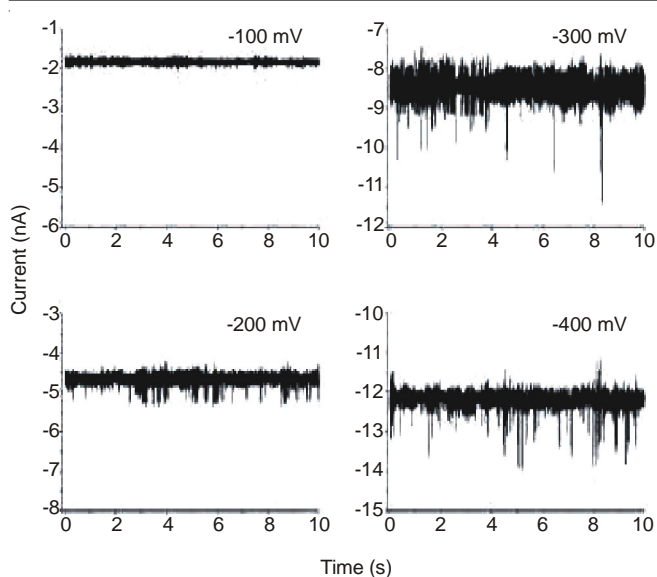


Fig. 3. *i-t* Recordings corresponding to the translocation of 25 nm radius polystyrene particles at different voltage using a 43 nm diameter single conical-shaped nanopore in a 0.1 M KCl solution at the pH value 3.7

positive electrode. The results showed that electroosmotic flow is reason for the transportation of polystyrene particle.

Polystyrene particles transport the double conical nanopore: Fig. 4 shows the example *i-t* trace of a 37 nm radius double conical-shaped nanopore in 0.1 M KCl solution containing 0.1 % Triton X-100 in the presence of 25 nm radius polystyrene particles. The baseline current stabilized increased with the increasing of the voltage, the current at the positive voltage was stable, individual pulses are observed at the

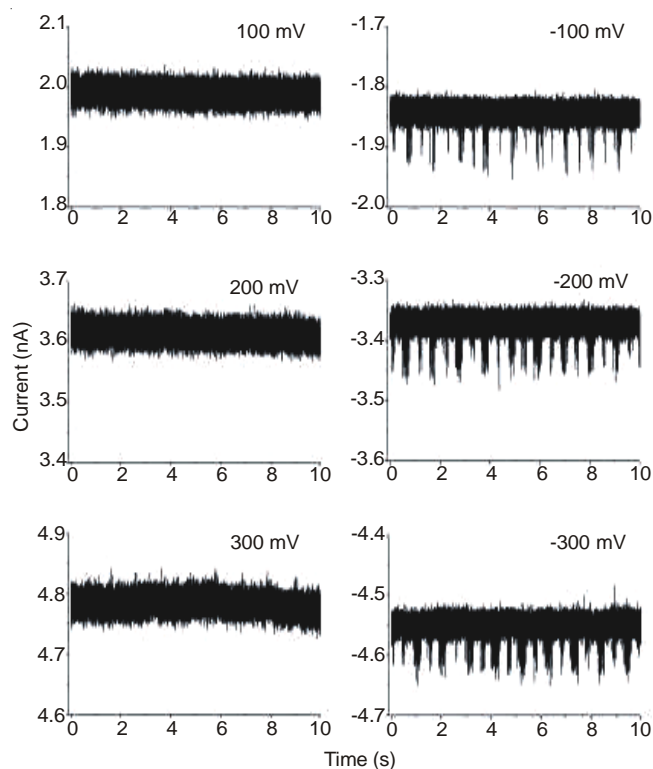


Fig. 4. *i-t* Recordings corresponding to the translocation of 25 nm radius polystyrene particles at different voltage using a 37 nm diameter double conical-shaped nanopore in a 0.1 M KCl solution (pH 5.9)

opposite voltage from -0.1 to -0.3 V, corresponding to the translocation of nanoparticles through the nanopore. At the applied voltage -0.1, -0.2 and -0.3 V the *trans*-membrane current became bigger when nanoparticle transported the nanopore, the particle translocation event rate increased with the increasing of the voltage.

At pH 5.9, the inner surface of the nanopore prepared by the above technique is negatively charged. The negative charges on the nanopore surface repel the negatively charged polystyrene nanoparticles, possibly hindering their entry into the pores. And the direction of the electroosmotic flow is from the positive to negative electrode. When applied a positive voltage to it, the electroosmotic flow and the repulsive interaction prevent the particles passing through the nanopore. At a negative voltage, the particles flow along with the electroosmotic flow, when the voltage increased, the particles could conquer the repulsive interaction and translocated through the nanopore, produced a resistive pulse. With the increasing of the negative voltage, the electroosmotic flow became bigger, more and more particles translocated through the nanopore, the particle translocation event rate increased. Fig. 5 shows that a linear dependence between the translocation frequency and the applied voltage. While the voltage is relatively uniform, a few larger pulses are also seen. This is due to the the translocation of aggregated particles, resulting in a relatively high pulse with longer duration time¹⁴⁻¹⁶. More coincident events are observed at higher voltage. When particle transported across the nanopore, the bulk currents would decrease due to the physical blocking by the particle as expected, but the polystyrene particle is negatively charged, resulting in an electric double layer around it, its translocation will also induce additional K^+ atoms within the channel and therefore the current increased.

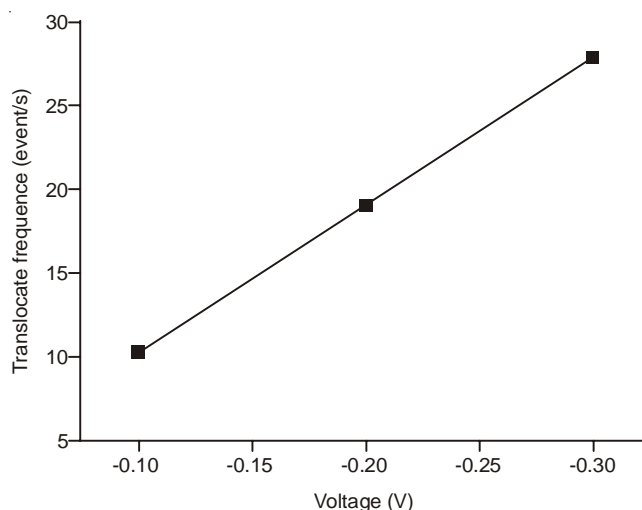


Fig. 5. Relationship between polystyrene particles translocation frequencies and applied voltage in a 0.1 M KCl solution (pH 5.9)

Fig. 6 shows the individual translocation pulse at different applied voltage. As can be seen, the pulse width almost the same when the voltage increased from -0.1 to -0.3 mV, the translocation time is about 300 μ s. It demonstrates the velocity of translocation didn't change linearly with the applied voltage.

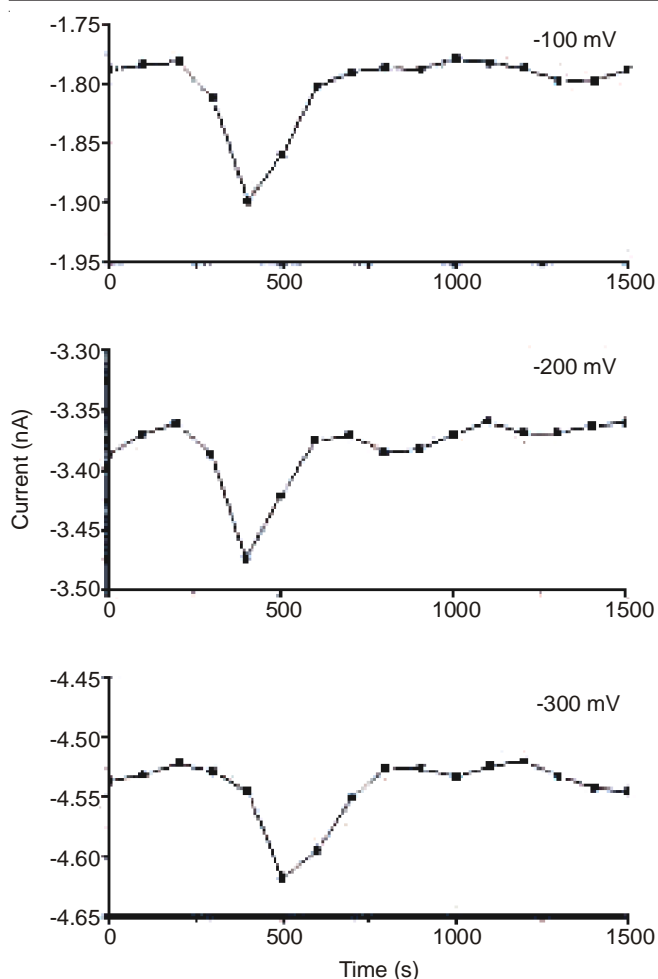


Fig. 6. Individual translocation pulse at different applied voltage in a 0.1 M KCl solution (pH 5.9)

Conclusion

The conical-shaped Kapton nanopores fabricated by the track-etched method have been used to analysis electro-osmotic-driven translocation of 25 nm radius polystyrene nanoparticles. The asymmetric triangular current pulse for

the translocation through has been found at the appropriate applied voltage. With the increasing of the applied voltage, the translocation frequency increases, the translocation time remains unchanged nearly. The investigation of transportation for pH values 3.7 and 2.02 of 0.1 M KCl demonstrated that electroosmotic flow is the driving force for polystyrene nanopore transportation.

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