Motion of dsDNA in a coupled nanochannel/nanopore system under an electric field

Yuliya Kuznetsova, Alexander Neumann, Olga Amosova, Xin Jin, S.R.J. Brueck
Armonica Technologies, Inc. Albuquerque, NM 87106
yuliya.kuznetsova@armonicatech.com

Investigation and analysis of genomic DNA samples is becoming critically important for clinical diagnostics and other biological applications where simple and inexpensive devices can create new capabilities and reduced analysis times. Additionally, interest in Lab-on-a-Chip micro/nanofluidic systems for analysis has tremendously increased in recent years. Here, we demonstrate a novel coupled-nanochannel/nanopore technique for DNA investigation and separation, as a route to accurate, inexpensive long-read genome sequencing.

The nanochannel/nanopore approach we have developed entails spin coating of multiple ~ monolayers of nominally 50-nm diameter colloidal silica nanoparticles on a nanopatterned photoresist surface. The nanoparticles "stack up" between the photoresist pattern features and ultimately form the walls and roof enclosing the resist lines. After the nanoparticles have been deposited, a 800°C calcination step (air ambient) is performed to sinter the nanoparticles and remove the resist (Fig. 1). Tortuous (convoluted) nanopores extending through the ceilings of the nanochannels are automatically formed in this process. Barriers, ~ 5- to 25-μm wide regions without nanochannels, were added with an additional optical lithography exposure in the same resist level to locally remove bands of the photoresist perpendicular to the grating lines. Wells for fluid access at the edges of the chip were introduced in a second lithography and etch process. Electrodes were mechanically placed in the wells to allow application of an electric field (Fig. 2).

We investigate DNA behavior in the nanochannel chip under the influence of an electric field (70 V across ~1 cm). Frames of a movie of ds-DNA transport (lambda-phage dsDNA 48,500 base pairs, visualized with YOYO-1 intercalated dye) through the tortuous nanopores in the roof and across the barriers are shown in Fig 3. DNA crosses the barriers by penetrating through the tortuous pores to the roof and then back into the channels as a result of the very thin layer of water atop the barriers. A time sequence along a single line of the movie is shown in Fig 3. The coupled nanochannel/nanopore system significantly slows the DNA translocation.

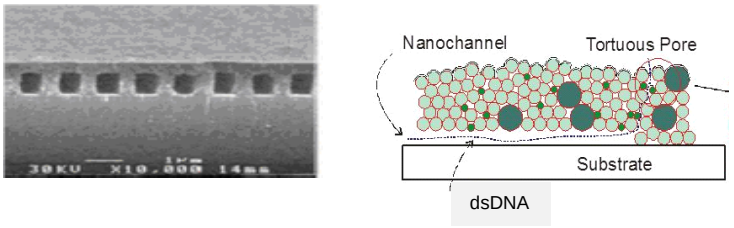

Fig. 1. (left) SEM of completed nanochannels; (right) Schematic of nanochannel structure. The different colors indicate the particle size dispersion.

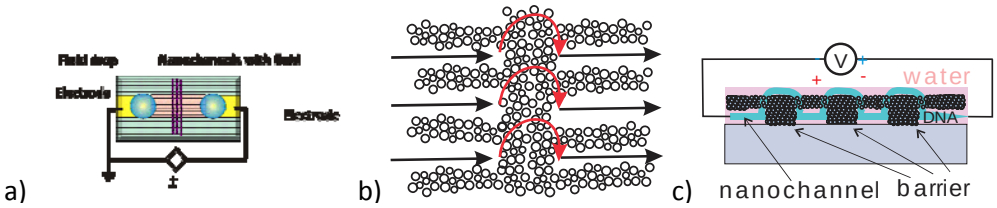

Fig. 2. a) Top view showing nanochannels, barriers, ports with an applied voltage across the chip. b) Schematic of particles along the centerline of the nanochannels showing the barriers. The arrows indicate the flow path of the DNA. c) Side view of nanochannels showing the porous roof. There is only a very thin film of water on the top of the roof.

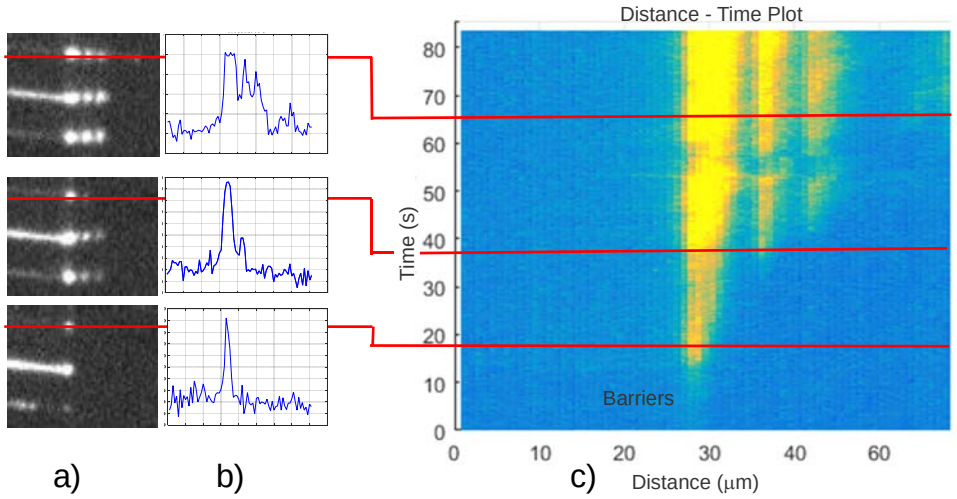

Fig. 3. a) Individual frames of a movie (at 18 s; 38 s; 65 s) of YOYO-intercalated lambda phage dsDA in nanochannels and transporting over three barriers under an applied electric field. b) Intensity vs. position for a single scan (red line) of each frame. c) False color intensity plot of the scans showing the progression of the DNA to the first barrier and then over each barrier. The DNA reenters the channels after moving across each barrier as a result of the very thin layer of water over the channels. At ~ 53s, the applied voltage was reversed for 5s, and the DNA flow was correspondingly reversed.