

Nano-assembly of DNA Based Electronic Devices Using Atomic Force Microscopy

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Abstract—DNA electronics circuits require an efficient way to accurately position and individually manipulate DNA molecules. The recent development of Atomic Force Microscopy (AFM) seems to be a promising solution. We have recently developed an AFM based augmented reality system. This new system can provide both real-time force feedback and real-time visual feedback during nanomanipulation. We have shown that nano-imprinting and manipulation of nano-particles and nano-rods can be easily performed under assistance of the augmented reality system. In this research, the system's ability is extended to manipulation of DNA molecules. Using a polynomial fitting method, the deformation of DNA molecules is displayed in real time in the augmented reality system during manipulation. Indeed, DNA molecules adopt many different structures including kinks, bends, bulges and distortions. These different structures and inappropriate physical contacts may result in the controversy of DNA conductivity reported over the last decade. The AFM based nanomanipulation system can be used either as a nanolithography tool to make small gap electrodes or a nanomanipulation tool to elongate, deform and cut DNA molecules. The measurement of the conductivity of DNA molecules in their different shapes and structures is a promising method to find conclusive evidences, which will verify the electrical conductivity of DNA molecules.

I. INTRODUCTION

DNA plays a key role in biology as a carrier of genetic information in all living species. The Watson-Crick double-helix structure for DNA has been known for almost 50 years. During the last half century, the majority of research into DNA has been devoted to its biological properties, in particular its role in genetic inheritance, disease, aging, RNA synthesis and mutation. Recently, the DNA molecule has drawn much attention in engineering applications because of its appealing features for use in nanotechnology: it's minuscule size, with diameter of about 2.4 nm, and its short structural repeat of about 3.4-3.6 nm [1]. The desire to use DNA as the ultimate building blocks of electronic circuits has motivated the studies of DNA electrical properties and DNA manipulation.

In 1962, D. D. Eley and D. I. Spivey first suggested that DNA could serve as an electronic conductor [2].

The initial model for electron transfer through DNA is based on overlapped π -stack orbitals in adjacent base pairs. After much initial controversy over the past 15 years, the mechanism of charge-transfer is now moving towards a consensus view in the chemistry community [3]. The dominant charge-transfer mechanism in DNA appears to be distance-dependent coherent tunnelling through unit-step and weak-distance-dependent thermal hopping through multi-step [4]. Both charge-tunnelling and thermal-hopping have been verified by [5]. Contrary to the consensus in the chemistry society, the problem of whether DNA is conducive or insulating remains to be hotly debated among physics groups due to the disparate experimental results. A lot of experiments have been performed to measure the conductivity of DNA from indirect methods to direct methods (physical contact). Most indirect results suggest that DNA is a good conductor by microwave cells method [6], fluorescence quenching [7] and resonant cavity measurements [8], but only few results prove that it is insulating by measuring its dielectric property [9]. For direct measurement (with physical contact), the results are even more controversial. Some results suggest that DNA is a good conductive molecule wire [10] or even a super conductor at low temperature [11], some report that DNA is a semi-conductor [12], [13], [14], [15], while others find that DNA behaves as an insulator [16], [17], [18].

Due in part to the difficulty of individually connecting a single DNA molecule to an electrode, the results of almost all experiments are not repeatable. The only way to get a reliable measurement result is to make the experimental process repeatable and in a controlled environment. Therefore, having an efficient way to individually manipulate DNA molecules is key to DNA electronics circuits. DNA is a random coil in solution and it needs to be elongated, cut and fixed on a solid surface. Many methods have been used in the alignment of DNA, for example surface tension [19], dielectrophoretic force [20], [21], [22], viscous drag [23], laser trapping [24]. However, all these methods are still far from the requirement to accurately position and

build DNA into circuits.

Since the invention of the Atomic Force Microscopy (AFM), it has become a standard technique in imaging various sample surfaces down to the nanometer scale. Recent development of AFM has rapidly extended its ability from surface imaging to manipulation of nano-objects [25], [26], [27], [28]. The main problem of these manipulation schemes is the lack of real-time visual feedback. Each operation has to be verified by another new image scan before the next operation. Obviously, this scan-design-manipulation-scan cycle is time consuming and makes manipulation inefficient. Combining the AFM with virtual reality interface and haptic devices [29], [30] may facilitate the nanomanipulation by simplifying the off-line design, but the operator is still blind because he/she cannot see in real time the environment changes through the static virtual reality, therefore, a new image scan is still necessary after each operation. Thus any methods which can update the AFM image as close as possible to the real environment in real time will help the operator to perform several operations without the need of a new image scan.

The Augmented Reality System developed in [31], [32] aims to provide the operator with real-time visual display and force feedback. The real-time visual display is a dynamic AFM image of the operating environment which is locally updated based on real-time force information and system models. Under the assistance of the Augmented Reality System, the operator can perform several operations without the need for a new image scan. Using the augmented reality system, we have successfully shown nano-imprints on a soft surface such as a polycarbonate surface [31], manipulation of latex particles on a glass surface [33], assembly of nano-rods on a polycarbonate surface [34]. In this paper, we extend the ability of the Augmented Reality System from manipulation of nano-particles and nano-rods to manipulation of DNA molecules. Using this system, the DNA molecule can be accurately positioned and easily modified by pushing, elongating, deforming and cutting. Solving the debate on DNA conductivity looks promising by measuring the conductivity of DNA molecules in their different shapes and structures. This technique starts an initial step towards fabricating DNA circuits.

II. DNA SAMPLES

In the study of DNA electrical conductivity, most researchers use commercial DNA samples with identical length and identical base-pair sequence such as λ -DNA. However, this may cause problems since DNA has almost unlimited base-pair sequences in nature and the nature of the sequence and length may affect its electrical conductivity. Instead of using commercial DNA samples, raw DNA samples with random length and random base-pair sequences are used in this research. The DNA sample was collected from mature seeds of peony species without restriction (cutting), therefore, some DNA molecules still keep their original length (Department of Plant Biology at Michigan State University). The AFM images of the DNA samples are shown in Fig. 1.

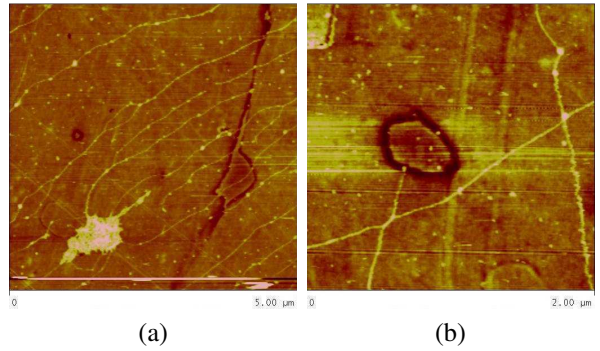


Fig. 1. AFM image of DNA samples: (a) Image of DNA sample in scanning range of 5 μ m. Most DNA molecules are longer than 5 μ m. (b) Image of DNA sample in scanning range of 2 μ m.

III. MANIPULATION OF SINGLE DNA MOLECULES USING THE AUGMENTED REALITY SYSTEM

In order to compare the electrical conductivity of DNA molecule in its different shape, kinks and deformation of DNA molecules can be created artificially using the AFM based nanomanipulation system. By controlling the pushing force between the tip and sample surface, the DNA molecules or DNA bundles can be either broken or deformed as shown in Fig. 2. A large pushing force in normal direction usually breaks the DNA molecule, and a small pushing force may only deform DNA molecule without breaking it. In Fig. 2(b), the big scratches on the surface indicate large pushing force applied on the AFM tip, and small scratches imply small pushing force used. We can see that the DNA bundle was broken when a big pushing force was applied but only deformed when a small pushing force was applied.

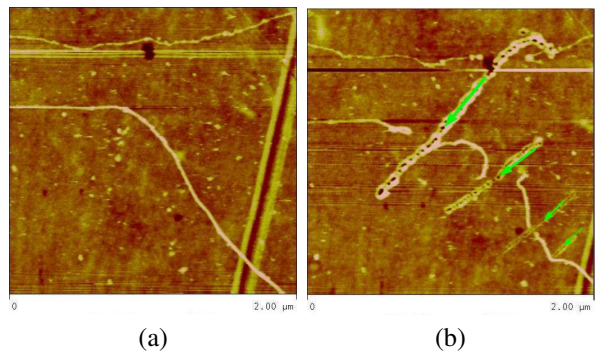


Fig. 2. (a). The AFM image of DNA ropes in its original shape. (b) The DNA ropes are cut by the AFM tip. The pushing force can be controlled in order to cut the DNA rope or only deform the DNA rope. The big scratches on the surface indicate strong pushing force applied, and small scratches imply small pushing force used. The arrows indicate the pushing directions.

In order to display in real time the deformation of DNA molecules or bundles in the Augmented Reality Environment, the deformation model of DNA molecules and bundles has to be found. Although some forces dominant in the nanoscale can be theoretically calculated [35], [36], it is not feasible to compute them because some parameters needed in the model are not available. For

example, object-surface distance, radii of curvature of the meniscus, and the thickness of the liquid layer, which are used to calculate the capillary force, are almost impossible to obtain. Therefore, instead of using mathematic model for the force-deformation of DNA molecules, an empirical method, as shown in Fig 3, is adopted in this research.

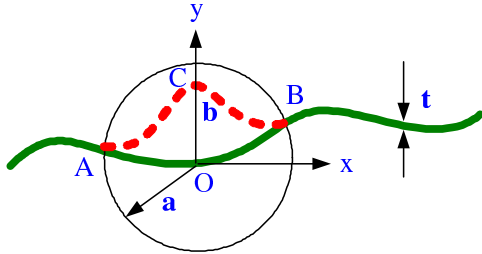


Fig. 3. Modeling deformation of DNA molecules and bundles under pushing by AFM tip. The solid curve is a DNA molecule (or DNA bundle) in its original status. The dash curve is the new status of the DNA molecules. The DNA molecule (bundle) is pushed by the AFM tip from the start point O to the end point C . The DNA molecule (bundle) is only deformed within an affecting region from point A to B , which is bounded by a circle with radius of a . t is the thickness of the DNA molecule (bundle).

As shown in Fig 3, the DNA molecule (bundle) is pushed by the AFM tip from the start point O to the end point C . The DNA molecule (bundle) is only deformed within an affecting region from point A to B , which is bounded by a circle with radius of a . a is determined by following empirical function f :

$$a = f(b, t)$$

where, b is the pushing distance before DNA broken, and t is the thickness of DNA itself. In order to display the DNA deformation in real time, a local coordinate system is defined such that y axis is along the pushing direction. The original shape of DNA inside the affecting circle can be removed first and then the new shape of DNA can be displayed through a 4th order polynomial function with respect to the local coordinate system. Define (x_A, y_A) , (x_B, y_B) and (x_C, y_C) as the position of points A , B and C ; and also define v_A and v_B as the slope of the tangent at points A and B . Then the 4th order polynomial function can be found as

$$y = a_4 t^4 + a_3 t^3 + a_2 t^2 + a_1 t + a_0$$

where

$$t = \frac{x - x_A}{x_B - x_A}$$

and the coefficients can be found through following equations

$$\begin{bmatrix} 0 & 0 & 0 & 0 & 1 \\ 0 & 0 & 0 & 1 & 0 \\ 1 & 1 & 1 & 1 & 1 \\ 4 & 3 & 2 & 2 & 1 \\ t_m^4 & t_m^3 & t_m^2 & t_m & 1 \end{bmatrix} \begin{bmatrix} a_4 \\ a_3 \\ a_2 \\ a_1 \\ a_0 \end{bmatrix} = \begin{bmatrix} y_A \\ v_A \\ y_B \\ v_B \\ y_C \end{bmatrix}$$

here

$$t_m = \frac{x_C - x_A}{x_B - x_A} = -\frac{x_A}{x_B - x_A}$$

It has been found that the AFM tip can go across the nano-objects if the pushing force is not strong enough to keep the tip in contact with the surface [33]. Therefore there exists a maximum value of b such that either the AFM tip goes across the DNA or the DNA is broken at that point if DNA is molecule is very thin. Namely,

$$b_{\max} = \text{Min}\{b_{\max}^1, b_{\max}^2\}$$

where b_{\max}^1 is determined by pushing force in normal direction F_n , DNA thickness t and surface properties. b_{\max}^2 is determined by DNA thickness t and surface properties. Unfortunately, these values have to be determined experimentally since it is almost impossible to obtain their mathematic models.

Using the model developed in this section, manipulation of DNA molecules can be displayed in real time in the augmented reality environment. An example of DNA manipulation is shown in Fig 4 in which Fig 4(a) shows the DNA molecules in their original shapes; Fig 4(b) shows the manipulation of DNA molecules displayed in the augmented reality environment; Fig 4(c) shows an AFM image after manipulation. It can be seen that several kinks have been created by slightly pushing the DNA molecules or bundles and the kinks created in the augmented reality environment are relatively identical to the real results. It has been also shown that the AFM tip goes across the kink created in the small circle as shown in Fig 4(b).

IV. DNA ELECTRICAL CONDUCTIVITY MEASUREMENT

In [10], the DNA ropes were suspended across holes in a gold-coated carbon grid and a tungsten tip was then used to make contact with the ropes. Using this method, Fink and Schönenberger measured a single rope of λ -DNA 600nm long, which indicated efficient conduction through the rope. In [11], double-stranded DNA molecules were deposited by a combing process across a submicron slit between rhenium/carbon metallic contacts. The measured resistance per molecule is less than 100 kilohm at room temperature also indicates that DNA has good ohmic conduction behavior. Porath *et al* observed wide band gap semiconductor-like behavior for a single, double stranded DNA with only G-C base pairs sequence between two Pt electrodes with gap of 8nm in [13]. The semiconductor behavior of DNA is also observed by K.-H. Yoo and *et al* in [15] in which they measured the electrical transport through polyadenine-polythymine and polyguanine-polycytosine molecules containing identical base pairs between two electrodes with gap of 20nm. The gate-voltage dependence transport measurement by introducing a third electrode shows that polyadenine-polythymine behaves as an n-type semiconductor, whereas polyguanine-polycytosine behaves as a p-type semiconductor. However, the absence of dc-conductivity in λ DNA is observed by P.J. de Pablo and *et al* in [16]. In that experiment, the DNA molecule was partially covered by a gold electrode while the second electrode was a metallic atomic force microscopy tip. Since the thermal drift is quite significant,

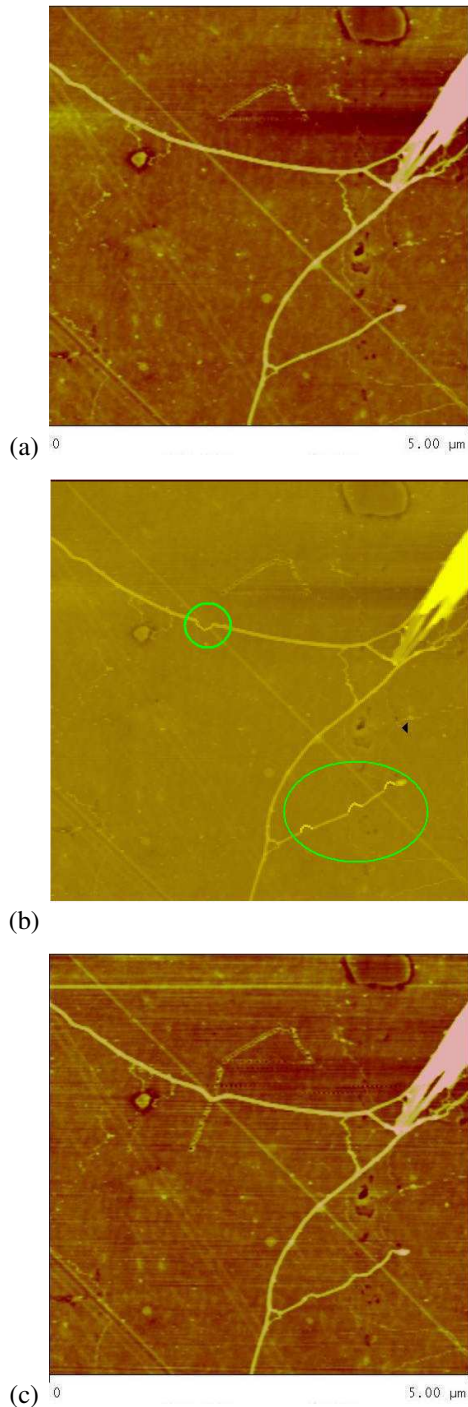


Fig. 4. Pushing DNA on a polycarbonate surface (scanning range of 5 μ m). (a) Image of DNA before pushing. (b) the real time display on the augmented reality during pushing; The kinks inside the circles are created by elaborately pushing. (c) A new scanning image after several pushing operations.

a reliable electrical contact between the tip and DNA is highly suspicious in this work. However, a series of experiments done by A.J. Storm in [17] prove that DNA is definitely an insulator at the length scale larger than 40nm. This result is clearly in disagreement with the work of the work of [10] and [11].

Indeed, the DNA molecule adopts many different struc-

tures which may affect the measurement results. These include kinks, bends, bulges and distortions along the molecule, as well as the flow of positively charged counterions along the negatively charged phosphate backbone. By measuring the height of a single DNA molecule, it has been found that there is very large compression deformation of the deposited DNA on the most common used substrates such as mica and silicon oxide surface [37]. In [38], the thickness of DNA molecules on the substrates treated by pentylamine is nearly 2.4 nm comparing to the thickness of 1.1 nm for DNA molecules on a clean substrate. Using atomic force microscopy (AFM) in spreading resistance mode, it has been found that the DNA is insulating on clean substrate but conductive on treated substrate. Another significant factor, which may affect the measurement result, is perhaps the electrical contacts. Ideally, these contacts should be ohmic so that any non-linearity in the conductivity of the molecular wire can be correctly attributed and studied. It has long been recognized that to make good electrical contact between a molecule and a conducting electrode, a chemical bond is required [39]. Therefore, a chemical bond approach for electrical contact and manipulation instrument for elongating, deforming or cutting DNA play key roles in verifying the hypothesis that the shape and structures of DNA molecules affect their conductivity.

In order to verify the distance dependence of DNA conductivity, a series of electrodes with gaps from 100nm to 3 μ m were fabricated on transparent substrates like polycarbonate or PMMA. PMMA is a transparent polymer which is commonly used for fabricating micro fluidic devices for biological application. The advantages of PMMA substrate are its transparency for easily locating the electrodes through an inverted optical microscope, and its softness to protect AFM tip. Most importantly, PMMA has hydrophobic surface property, which keeps DNA molecules in their original shape without strong compression due to adhesion force.

A. Fabrication of Electrodes Using a Standard Microlithographic Technique

We have demonstrated previously in [40] that gold (Au) microelectrodes with gap distances smaller than 2 μ m were fabricated using a 5 μ m gap distance mask design by over-developing technique during image reversal process. After patterning the AZ5214 positive photoresist, chromium (Cr) was first deposited as the adhesion layer between gold and silicon substrate. Electrodes were then patterned by lift-off process using acetone. However, it was found that PMMA is easily attacked by acetone, thus, lift-off process adopted for silicon substrates cannot be used. Because of this, wet etching was employed in the fabrication process. To enhance the adhesion of the microelectrodes on the polymer substrate, a thin parylene C layer of 0.1 to 0.5 μ m was first deposited on the PMMA substrate. The substrate was then coated with chromium and gold inside the electron beam evaporator and then spun with AZ5214 positive photoresist and was patterned to desired geometries. After lithography, the gold and chromium layers were etched by

gold etchant ($KI : I_2 : H_2O = 4 : 1 : 80$) and chromium etchant respectively. The photoresist was finally removed by oxygen plasma. The entire process is shown in Fig. 5. A pair of electrodes with gap of 2um fabricated by this method is shown in Fig. 6(a).

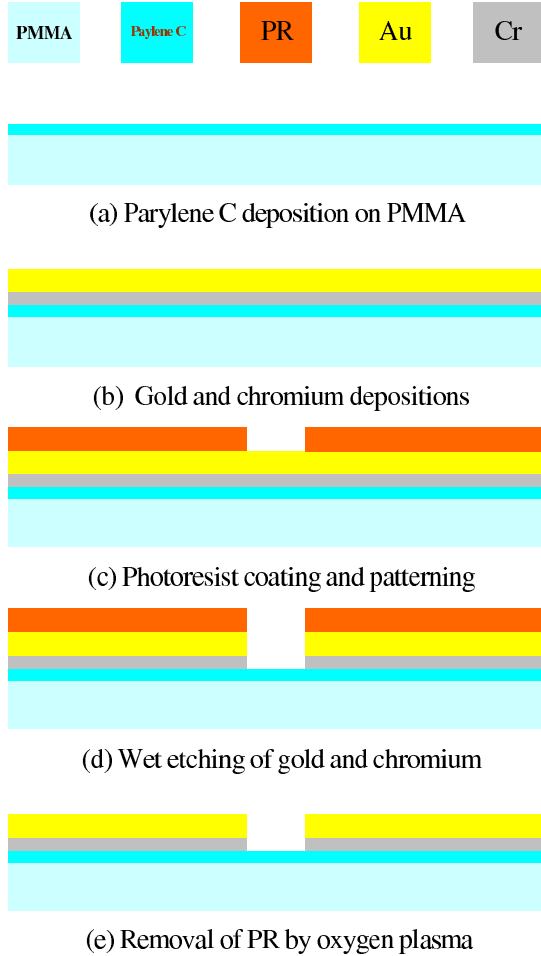


Fig. 5. Fabrication process of microelectrodes on a PMMA substrate.

B. Fabrication of Electrodes Using AFM Based Augmented Reality System

In order to increase the possibility of more DNA molecules lying inside the electrode gap, a wide and short gap is preferred. However, it is very hard to make a gap smaller than 100nm. Fortunately, the AFM itself can also serve as a nanolithography tool under assistance of the augmented reality system. By controlling the AFM tip through a haptic joystick, a very small electrode gap can be made on the gold coated surface. Fig. 6(b) shows that a 150nm gap can be made by inscribing the AFM tip into the gold coated electrode (the thickness of gold coating is about 70nm, and it is prepared as described in Subsection IV-A).

C. Electrical Conductivity of Bundle DNA

Using the electrodes created by AFM nanolithography methods as shown in Fig. 6(b), conductivity of bundle DNA can be easily measured. The electrodes are connected by

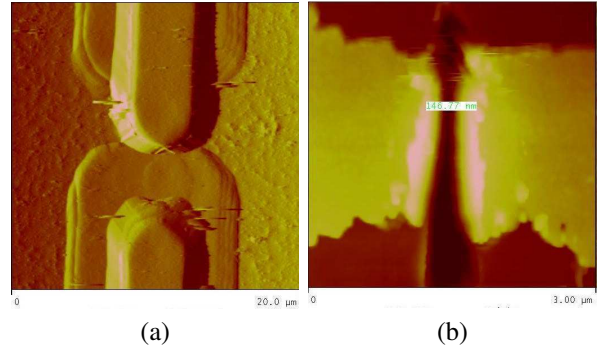


Fig. 6. (a). AFM image of the electrodes fabricated on a polycarbonate surface with a gap of 2um (20um scanning range). (b) AFM image of electrodes fabricated by AFM nanolithography on a gold coated polycarbonate surface with a gap of 150nm (3um scanning range).

a layer of gold 70nm thick. There is linear resistance of 15Ω only between the electrodes before nano-inscribing. After the nano-inscribing, a 150nm gap is created along the connection and the resistance between the electrodes becomes almost infinite. After the gap has been created, the DNA sample suspending in DI water was dropped on the electrodes and then blown with dry nitrogen. The resistance between electrodes becomes $180M\Omega$ after depositing DNA sample. The I-V curves before and after deposition of DNA sample are shown in Fig. 7 (The current is measured by a Picometer with resolution of 10 fA from Keithley Co.). This proves that DNA bundles are conductive in short distances. Using the electrodes with 2um gap and similar measurement method, we could not observe the electrical conductivity of DNA bundles in long distance. This experiment partially prove that the conductivity of DNA molecules is distance-dependent.

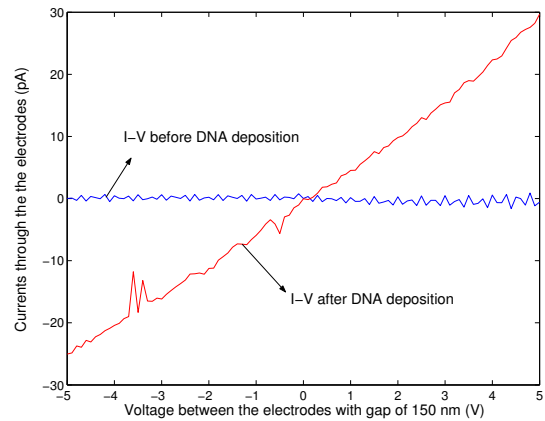


Fig. 7. I-V curves before and after deposition of DNA sample on the surface

V. DISCUSSIONS

The recent debate on DNA electrical conductivity requires more efficient experimental methods. We have developed an AFM based augmented reality system, in which the AFM system has been modified into a nanolithography and nanomanipulation tool. Using this system, small-gap

electrodes can be made by nanolithography and DNA molecules can be elongated deformed and cut by nanomanipulation. Our next step is to measure the conductivity of DNA molecules in different shapes between electrodes with different gaps. Through cross verification based on a series of experiments, conclusive evidences will be found to terminate the dispute on the electrical conductivity of DNA molecules.

ACKNOWLEDGMENT

This research work is partially supported under NSF Grants IIS-9796300, IIS-9796287 and EIA-9911077. The authors are pleased to acknowledge the support of Dr. Tao Sang in the Department of Plant Biology at Michigan State University who provides the DNA samples.

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