Electrical detection of hybridization and threading intercalation of deoxyribonucleic acid using carbon nanotube network field-effect transistors

Ee-Ling Gui, Lain-Jong Li,[a] P. S. Lee, Anup Lohani, and S. G. Mhaisalkar
School of Materials Science and Engineering, Nanyang Technological University, 50 Nanyang Ave., Singapore 639798, Singapore

Qing Cao, Seong Jun Kang, and John A. Rogers
Department of Materials Science and Engineering, University of Illinois at Urbana-Champaign, Illinois 61801 and Department of Chemistry, University of Illinois at Urbana-Champaign, Illinois 61801

N. C. Tansil and Zhiqiang Gao
Institute of Bioengineering and Nanotechnology, 31 Biopolis Way, Singapore 138669, Singapore

(Received 7 September 2006; accepted 20 October 2006; published online 4 December 2006)

The authors study deoxyribonucleic acid (DNA) sensing characteristics of carbon nanotube network field-effect transistors (CNNFETs) by monitoring their electrical responses upon immobilization with a DNA probe, hybridization with DNA analytes, and intercalation with a N,N'-bis(3-propylimidazole)-1,4,5,8-naphthalene diimide modified with $Os(2,2'\text{-bipyridine})_2Cl^+$ pendants. The CNNFETs immobilized by single-stranded DNA molecules demonstrate the selective sensing of its complementary and single-base mismatched DNA (difference of ~16% in reduction of normalized drain current I_d). Subsequent intercalation demonstrates a further sensitivity enhancement (difference of ~13% in I_d reduction) due to specific binding between hybridized DNA and intercalators, corroborated by the x-ray photoelectron spectroscopy study. © *2006 American Institute of Physics*. [DOI: 10.1063/1.2399355]

The development of deoxyribonucleic acid (DNA) hybridized biosensors is motivated by the wide scale of genetic testing, clinical diagnostics, fast detection of biological warfare, and environmental testing, where cheap biosensors with miniaturized analytical systems are required. Carbon nanotubes (CNTs) have attracted much attention because their electronic structure is sensitive to molecular adsorption on the tube walls[1,2] or within the tubes.[3] Therefore there is a growing body of research on the development of carbon nanotubes in DNA sensing. The covalent functionalization of carbon nanotubes surfaces to attach DNA capture probes[4] and the fluorescent confocal detection of dye labeled complementary DNA have been reported.[5] Electrochemical detection of DNA based on vertically aligned multiwalled carbon nanotube nanoelectrode arrays has also been demonstrated.[6] Recently, noncovalent functionalization of carbon nanotube feld-effect transistor (FET) devices has been reported to show highly sensitive electronic detection of biomolecules such as antibodies.[7–9]. A very recent paper has demonstrated the label-free detection of DNA hybridization using CNT-FETs immobilized with single-stranded DNA.[10]

Here we report that the carbon nanotubes network field-effect transistors (CNNFETs) immobilized by a DNA oligomer encoded with a terminal NH_2 (NH_2-DNA) show reliable detection and differentiation of its complementary and single-base mismatched DNA analyte. We demonstrate that sensitivity improvement using threading intercalator can be achieved by transport measurements in dry FET devices rather than using conventional electrochemical detection. The intercalator we use is the N,N'-bis(3-propylimidazole)-1,4,5,8-naphthalene diimide (PIND) modifed with two $Os(2,2'\text{-bipyridine})_2Cl^+$ pendants (PIND-Os), which has been reported to bind strongly to double-stranded DNA because these two pendants interact with DNA via electrostatic forces.[11] The selective intercalation of PIND-Os with CNNFETs hybridized with complementary DNA further enhances the sensitivity of the devices and the $Os(bpy)_2Cl^+$ (bpy=bipyridine) pendant primarily contributes to the current reduction.

The fabrication of CNNFET devices has been reported elsewhere.[12–14] The electrode of the CNNFETs used was pure Au. The channel length separated by Au electrodes in the devices was varied from 5, 10, 25, 50, 75 to 100 μm. Figure 1(a) demonstrates the schematic illustration of the nanotube network devices and a typical atomic force microscope image showing strands of single-walled carbon nanotube (SWNTs) and catalyst particles on them. For immobilization of the capture probes the CNNFET devices were incubated in 1 μM of 12-mer synthetic oligonucleotides in a Tris-ethylenediaminetetraacetic acid (Tris-EDTA) buffer solution (10 mM tris-HCl/1.0 mM EDTA/0.10M NaCl) buffer solution for a period of 16–24 h. Following immobilization, the devices were washed thoroughly with buffer solution to remove the excess DNA and were subsequently dried. This ensured that the observed change in SWNT devices were not related to loosely bonded species on the device surface.

For hybridization experiments, 10 μl of DNA solution (500 nM complementary or single-base mismatched target analyte) was pipetted on top of the immobilized devices for 1 h, followed by washing. The sequences of the synthetic oligonucleotide and the target analytes are shown in Fig. 1(b). For the intercalation process 10 μl of PIND or PIND-Os [chemical structures shown in Fig. 1(c)], with the

[a]Electronic mail: ljli@ntu.edu.sg

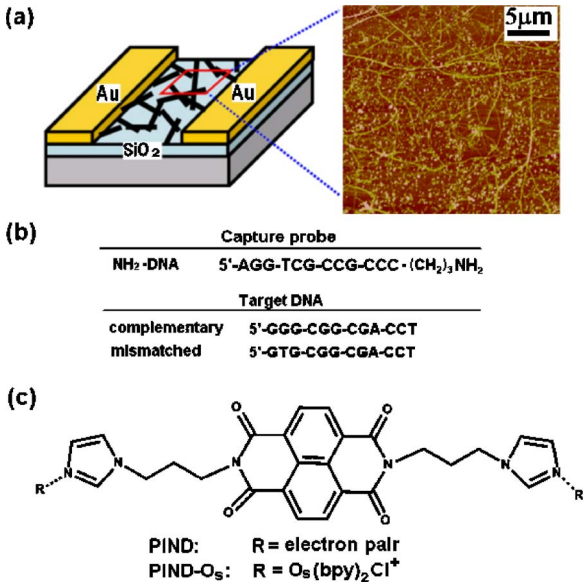

FIG. 1. (a) Schematic illustration of the network devices and a typical atomic force microscope image (5×5 μm^2) showing strands of SWNT and catalyst particles in the active channel area. (b) Sequences of the synthetic oligonucleotide, NH_2-DNA, and their complementary and single-base mismatched target analytes. (c) Structure of PIND and PIND-Os intercalators.

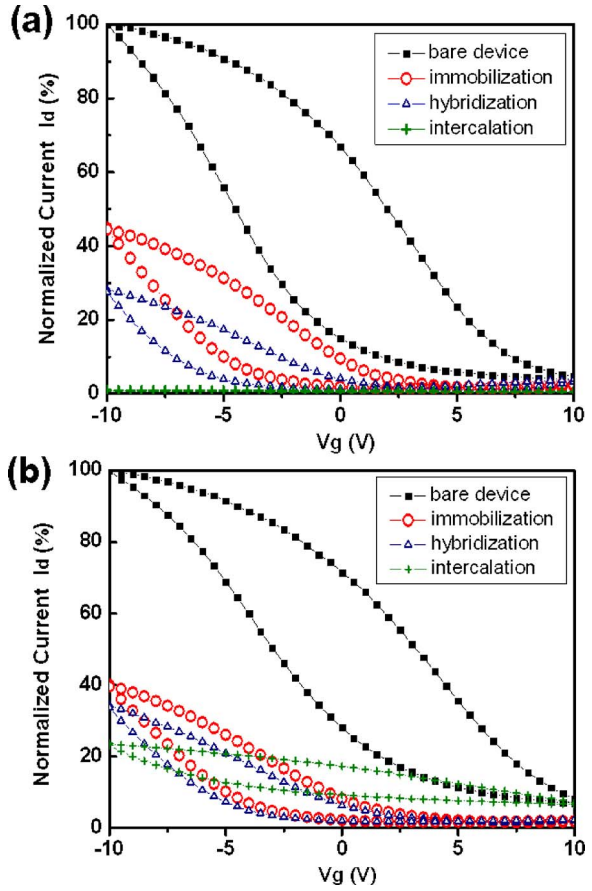

FIG. 2. Typical gate voltage dependence of the normalized drain current I_d (normalized by the initial drain current of their bare device at $V_g=-10$ V) for (a) a CNNFET bare device immobilized with NH_2-DNA, hybridized with complementary target analyte, (b) a CNNFET bare device, immobilized with NH_2-DNA, hybridized with single-base mismatched target analyte, and immersed with the PIND-Os intercalator (source-drain bias was kept at −0.5 V).

concentration of 100 μg/ml in Tris-EDTA buffer solution, was pipetted on the hybridized devices for 15 min, followed by thorough washing.

Figures 2(a) and 2(b) show the typical transfer curves for CNNFETs after immobilization with NH_2-DNA, where large reduction in normalized drain current (I_d) was observed with respect to corresponding bare devices. Reduction in drain currents has been attributed to attachment of DNA molecules on the sidewalls of CNTs resulting in electron doping to the CNT semiconductor channels.[10] On the other hand, in other reports, drain current reduction has also been attributed to the modulation of the metal-nanotube contact resistance due to gas environments or molecular absorption.[15,16] The cause for I_d reduction and the exact contribution of the DNA-CNT interaction and its effect on charge transport warrants further study.

The immobilized CNNFETs hybridized with their complementary target DNA show a relatively large percentage reduction of I_d [18.8% in Fig. 2(a)] as compared to those hybridized with the single-base mismatched target DNA [6.4% in Fig. 2(b)]. The significant reduction in I_d for complementary hybridization indicates the formation of a double-stranded DNA which may lead to the increase of scattering centers on semiconductor channels[10] or the shifts in Au work function further away from the valence band of carbon nanotubes.

Figure 3(a) summarizes the mean values of the sequential reduction in I_d after immobilization and hybridization. The DNA hybridization response was studied on devices with transistor lengths varying from 5 to 100 μm. The nanotube network is a random direction assembly of SWNTs with different diameters and tube lengths. Because the interaction between SWNT and DNA depends on tube diameter[17] this distribution may affect the relative change of I_d, as its influence on the absolute I_d value in pristine devices with identical device geometries.[13] Statistics carried out based on 14 devices with different channel lengths indicate that device-to-device variations, ~5% for complementary and for mismatched, are smaller than difference that result from different analytes. Therefore it allows us to conclude that the NH_2-DNA immobilized CNNFETs show significant differentiation in selective detection of complementary (21.5%) and single-base mismatched (5.1%) DNA analytes. The immobilized samples exposed to only the Tris-EDTA buffer solution on the other hand showed no change in I_d, thus confirming that the DNA-transistor interactions were directly responsible for the change in I_d.

The DNA intercalators have been used since 1990 to avoid labeling of the target DNA and improved the detection limit by electrochemical methods.[11,18,19] In Fig. 3(a) we observe that further incorporation of PIND-Os intercalators results in an additional 21.8% of I_d reduction for the CNNFETs hybridized with complementary DNA, while relatively small percentage of change in I_d (9.1%) is detected for those hybridized with mismatched DNA. This indicates that a highly selective intercalation to double-stranded DNA is achieved and the process can be detected by electrical measurements for FET devices in dried states. Figure 3(b) provides the x-ray photoelectron spectroscopy (XPS) spectrum (Al as the x-ray source) where the Os $4f_{7/2}$ core level binding energy at 50.4 eV and Os (VIII) peak at 52.8 eV are identified.[20] After PIND-Os intercalation the Os content in the CNNFETs with complementary DNA hybridization is significantly enriched as opposed to the CNNFETs with mismatched DNA hybrid-

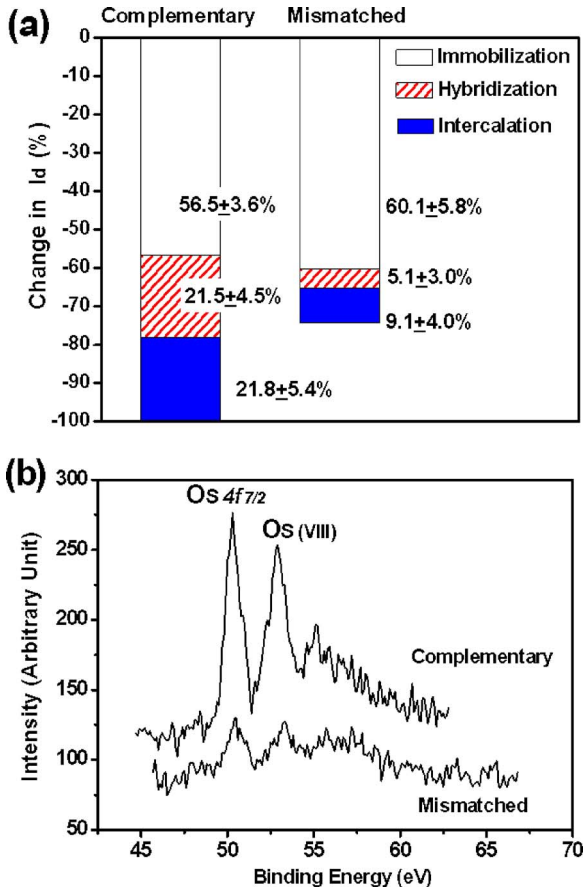

FIG. 3. (a) Statistical comparisons of sequential reduction in I_d for CNNFETs (left column: eight devices in total with channel lengths of 5, 10, 50, 75, and 100 μm; right column: six devices in total with channel lengths of 5, 25, and 75 μm) after immobilization, hybridization (complementary: hybridized with complementary DNA; mismatched: hybridized with single base mismatched DNA), and intercalation. (b) XPS comparison of the Os contents for two selected CNNFETs after PIND-Os intercalation.

ization. This corroborates the selective intercalation concluded from electrical measurements. To understand the causes of current reduction by intercalation we have also performed the parallel intercalation experiments using PIND intercalator, where the I_d is not reduced after intercalation to the CNNFETs hybridized with the complementary DNA. This suggests that the redox-active ligand $Os(bpy)_2Cl^+$ primarily contributes to the I_d reduction for PIND-Os intercalation.

In summary, we have performed the electrical measurements for CNNFETs immobilized with NH_2-terminated DNA. It is concluded that NH_2-DNA immobilized CNNFETs show reliable detection and differentiation for selective hybridization between complementary and single-base mismatched target DNA. A further sensitivity enhancement is achieved by including an intercalation process. The intercalation for double-stranded DNA can be detected by the transport measurement using a PIND-Os intercalator, where the redox-active ligand $Os(bpy)_2Cl^+$ is believed to cause an enhancement in the transduction mechanism based on the reduction of drain current. The contribution to the reduction in drain current could come from factors that include metal-CNT contact modulation as well as doping and/or charge transfer or interaction between the CNT and DNA. Further studies are required to understand the role of charge interaction between DNA and CNT, and how the intercalators participate in these charge interactions.

This research was supported by Nanyang Technological University, Singapore. One of the authors (E.L.G.) would like to thank the scholarship provided by EDB, Singapore.

[1]D. A. Heller, E. S. Jeng, T.-K. Yeung, B. M. Martinez, A. E. Moll, J. B. Gastala, and M. S. Strano, Science **311**, 508 (2006).
[2]J. Kong, N. R. Franklin, C. Zhou, M. G. Chapline, S. Peng, K. Cho, and H. Dai, Science **287**, 622 (2000).
[3]L. J. Li, A. N. Khlobystov, J. G. Wiltshire, G. A. D. Briggs, and R. J. Nicholas, Nat. Mater. **4**, 481 (2005).
[4]S. E. Baker, W. Cai, T. L. Lasseter, K. P. Weidkamp, and R. J. Hamers, Nano Lett. **2**, 1413 (2002).
[5]M. Hazani, R. Naaman, F. Hennrich, and M. M. Kappes, Nano Lett. **3**, 153 (2003).
[6]J. Li, H. T. Ng, A. Cassell, W. Fan, H. Chen, Q. Ye, J. Koehne, J. Han, and M. Meyyappan, Nano Lett. **3**, 597 (2003).
[7]R. J. Chen, S. Bangsaruntip, K. A. Drouvalakis, N. W. S. Kam, M. Shim, Y. Li, W. Kim, P. J. Utz, and H. Dai, Proc. Natl. Acad. Sci. U.S.A. **100**, 4984 (2003).
[8]A. Star, J.-C. P. Gabriel, K. Bradley, and G. Gruner, Nano Lett. **3**, 459 (2003).
[9]R. J. Chen, H. C. Choi, S. Bangsaruntip, E. Yenilmez, X. Tang, Q. Wang, Y.-L. Chang, and H. Dai, J. Am. Chem. Soc. **126**, 1563 (2004).
[10]A. Star, E. Tu, J. Niemann, J.-C. P. Gabriel, C. S. Joiner, and Christian Valcke, Proc. Natl. Acad. Sci. U.S.A. **103**, 921 (2006).
[11]N. C. Tansil, H. Xie, F. Xie, and Z. Gao, Anal. Chem. **77**, 126 (2005).
[12]S.-H. Hur, D.-Y. Khang, C. Kocabas, and J. A. Rogers, Appl. Phys. Lett. **85**, 5730 (2004).
[13]Y. Zhou, A. Gaur, S.-H. Hur, C. Kocabas, M. Meitl, M. Shim, and J. A. Rogers, Nano Lett. **4**, 2031 (2004).
[14]S.-H. Hur, C. Kocabas, A. Gaur, M. Shim, O. O. Park, and J. A. Rogers, J. Appl. Phys. **98**, 114302 (2005).
[15]X. Cui, M. Freitag, R. Martel, L. Brus, and P. Avouris, Nano Lett. **3**, 783 (2003).
[16]X. Tang, S. Bansaruntip, N. Nakayama, E. Yenilmez, Y.-L. Chang, and Q. Wang, Nano Lett. **6**, 1632 (2006).
[17]M. Zheng, A. Jagota, M. S. Strano, A. P. Santos, P. Barone, S. G. Chou, B. A. Diner, M. S. Dresselhaus, R. S. Mclean, C. B. Onea, C. C. Samsonidze, E. D. Semke, M. Usrey, and D. J. Walls, Science **302**, 1545 (2003).
[18]M. T. Carter, M. Rodriguez, and A. J. Brad, J. Am. Chem. Soc. **111**, 8901 (1989).
[19]S. Takenaka, K. Yamashita, M. Takagi, Y. Uto, and H. Kondo, Anal. Chem. **72**, 1334 (2000).
[20]C. K. Rhee, M. Wakisaka, Y. Tolmachev, C. Johnston, R. Haasch, K. Attenkofer, G. Q. Lu, H. You, and A. Wieckowski, J. Electroanal. Chem. **554**, 367 (2003).