

Ion Channel based biosensors: Ionic transport in carbon nanotubes

S. Joseph, R. J. Mashl^{**}, E. Jakobsson^{**} and N. R. Aluru

^{**}National Center for SuperComputing Applications.
Beckman Institute of Advanced Science and Technology.
University of Illinois, Urbana, IL-61801.

ABSTRACT

Molecular dynamics simulations were used to study ionic flow in carbon nanotubes with the goal of using carbon nanotubes as artificial protein channels found in cell membranes. Simulations show that ion occupancy in a carbon nanotube solvated in an electrolyte is very low. When partial charges were placed on the rim atoms of an uncapped (16, 16) tube and an external electric field was applied it was found that the ion occupancy increased significantly. To mimic an ion channel in a membrane, functional groups were attached at the ends and the tube was placed in a slab. The functionalized tube was found to conduct ions in the presence of an electric field and depending on the direction of the field it was selective to anions over cations.

Keywords: Ion channels, biosensors, carbon nanotubes, functionalization.

1 INTRODUCTION

Transport of molecules through macromolecular pores is of considerable importance in many biological and nanoelectromechanical systems. Ion channels in cell membranes are crucial for shaping electrical signals and controlling flow of ions and fluids across cells. Channels have interesting properties like gating and selective permeability that are still topics of interest in active research. In recent years engineered ion channels have been developed to function as single molecule detection systems [1], [2]. In an applied potential, an ionic current is carried by the ions that bath both the sides of the lipid bilayer. When the target molecule binds to the binding site in the pore, the current is modulated. The frequency of binding reveals the concentration of the analyte and the duration and amplitude of the events reveal its identity. Though engineered channels have significant advantages including high sensitivity, wide dynamic range and biocompatibility, their lack of durability makes them reliable only in a lab setting.

A review of recent literature shows that promising options for such a device that can be practically realized would be Gold nanotubule membranes [3],[4], ion beam etched silicon nitride membranes [5] or single carbon nanotubes. Carbon nanotubes being rigid and having exciting electrical and mechanical properties seem ideal for

an integrated circuit chip design but fundamental questions have to be addressed regarding transport of water and ions through it. Quantitative information on fundamental modes of mass transport and transport rates have been obtained through carbon nanotubes of diameters of about 150 nm [6] Hummer et al [7] showed by molecular dynamics simulations that water molecules enter nanotubes of as small as 8.4 Å diameter even though carbon is hydrophobic. In their experiments, Miller & Martin [8] show that it is possible to control the rate and direction of electrosmotic flow through nanotube membranes of large diameters (120 nm). Both side wall and end wall chemical functionalization attachments of nanotubes have been successfully realized in recent experiments[9], [10], [11].

In this paper we investigate the possibility of a nanoscale device that incorporates the functionality of ion channels into artificial nanotubes that could be bulk manufactured and is far less complex than a biological system. Molecular Dynamics simulations of functionalized nanotubes show that they can mimic ion channels and their features like selectivity and binding.

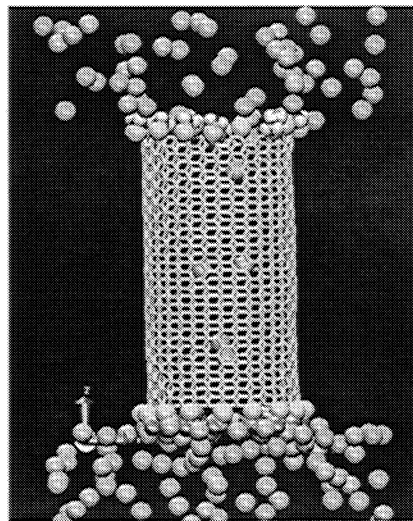


Figure 1: Bottom: Functionalized carbon nanotube in a bath of 1.5 M KCl. $\text{CH}_2\text{-NH}_3^+$ is attached one per carbon ring to the top and $\text{CH}_2\text{-COO}^-$ is attached at the bottom. The tube is inserted in a slab (not shown). Chloride ions are shown in darker shade. Water molecules are also not shown. Chloride occupancy is higher than potassium. A buildup of potassium ions near the COO^- is also observed.

2 SIMULATIONS

Simulations were performed by GROMACS [12] at a constant pressure of 1 bar and a constant temperature of 300 K. The system consisted of the carbon nanotube, water and ions and in some cases a slab and functional groups. The box size varied from 33 Å to 75 Å depending on the length and diameter of the tube and was at least twice as long as the tube length. The tube was fixed at the center of the box when there was no slab, but had limited freedom of motion when inserted into a slab which was fixed. The parameters for Lennard Jones potentials and bonded interactions were taken from GROMOS 96 force field with a carbon-carbon bond length of 1.42 Å. SPC/E model was used for water. Depending on the box size 1033 to 2970 water molecules were used to have a constant density of 1 g/cc. Concentrations of KCl varied from 1 M to 1.85 M. The slab consisted of pseudo atoms with the same LJ parameters as that of a lipid bilayer. The partial charges for NH₃⁺ and COO⁻ were taken from the side groups of lysine and glutamic acid respectively in the GROMACS amino acid database. Electrostatics was performed using Particle Mesh Ewald Summation. The simulation time varied between 2 and 7 ns and a time step of 2 fs was used in a leapfrog scheme.

2.1 Driving ions into CNTs

Knowing that water freely enters the tube from Hummer et. al [2001], we tested the ion occupancy in uncapped tubes of varying diameter in a solution of 1.85 M KCl. The tubes were fixed in the box center and were of length 13.4 Å. It was observed that in the absence of electric field and partial charges ion occupancy was very low (Figure 2) even for tubes with diameters as big as 21.696 Å. In the presence of an electric field of strength 0.015 V/nm, the ion occupancy increased significantly.

To attract even more ions into the tube, partial charges of +/-0.38 e were placed at atoms on the rim of the tube to create a dipole – the positive charges being on top rim and the negative charges on the bottom rim. The tube was then fixed in the center of a box of length 33 Å and an external electric field of 0.015 V/nm was applied along the axial direction to mimic the membrane potential. It was observed that the occupancy is even higher (See Figure 3).

Even though an electric field alone would drive ions into the tube, the partial charges on the rim increases the sensitivity and could be used to control the type and rate of ionic flow into the tube.

An interesting observation is the fact that the average occupancy of chloride ions is higher than that of the potassium ions (Figure 4). The reason for this could be explained by the water structure around the ions as well as

the van der Waals interaction between chloride ions and the nanotube.

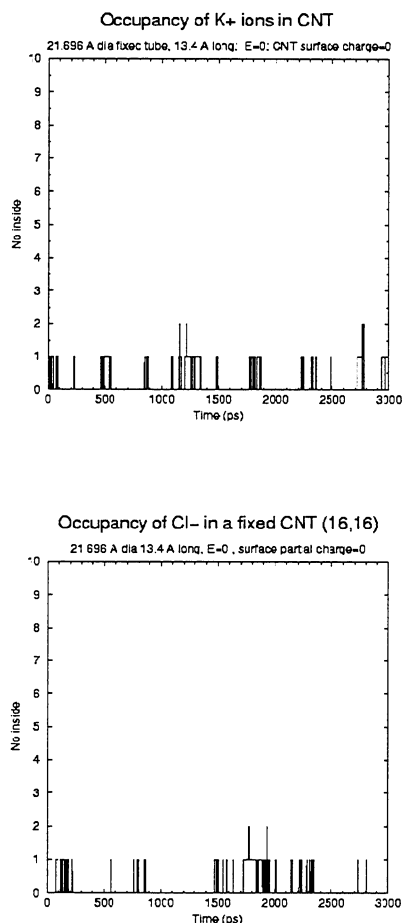


Figure 2. Very low ion occupancy in a (16,16) carbon nanotube (13.4 Å long, 21.696 Å diameter) fixed in a solution of 1.85 M KCl.

2.2 Functional Group attachment to carbon nanotubes to mimic ion channels

Once partial charges were shown to increase occupancy, the next step was to replace them with functional groups. In our simulations, an asymmetric functionalization of carboxylate and amino residues were used in place of the partial charges to mimic a real ion channel at either ends respectively. Even though functionalizing the inner wall would mimic the ion channel more closely, practically end wall functionalization is more feasible than inner wall functionalization for tubes of small diameter. The functionalized carbon nanotube was then placed in a slab with similar properties of that of a lipid bilayer with a surrounding bath of 1.5 M KCL solution. An electric field of 0.15 V/nm was used to drive the ions through the tubes

and the trajectories of K^+ and Cl^- ions are shown in Figure 5. The chloride current is much higher than potassium ion current even though the net partial charges at the mouths are same in magnitude. The attractive force between the COO^- and the K^+ ions is large that the K^+ ions are “bound” to the COO^- groups at the mouth causing an energy barrier thereby reducing the K^+ occupancy in the tube. Just like ionic channels, the functionalized carbon nanotube also seems to exhibit selective permeability of anions over cations. The phenomenon of binding of ions to functional groups can be used for modulation of the ionic current and this type of ion buildup causes inactivation of voltage gated K^+ channels.

Simulations of carbon nanotubes of a smaller diameter (10, 10) show that the time required for an ion passage is smaller but the principle of selectivity still applies. The smaller the tube diameter the water molecules arrange in smaller number of files and this lowers the mobility of ions. In fact we were not able to observe an ionic passage in a (6, 6) tube in a simulation time of 2 ns because the water molecules arrange themselves in a single file and mobility is considerably low.

The functionalized carbon nanotube system we have simulated is akin to multi-ion protein pores that contain several ions at a time. It was observed that for a (10,10) tube, even though the tube size is large enough for ions to pass each other, the electrostatic repulsion drives them apart so that they cannot do so. This phenomenon is observed in multi-ion pores. However in a larger diameter (16, 16) tube it was seen that chloride ions indeed do pass each other.

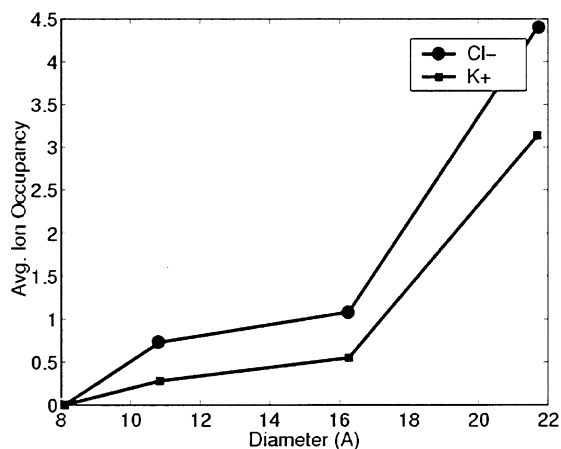


Figure 4: Variation of average ion occupancy with diameter in CNT (13.4 Å long) fixed in 1.85M KCl with an external applied electric field of 0.015 V/nm.

3 CONCLUSIONS

The possibility of incorporating the functionality of ion channels into carbon nanotubes has been demonstrated even though naturally carbon nanotubes have low ion occupancy. A much more detailed study is required for understanding physical mechanisms. Many theories including mean spherical approximation [13] have been used to describe selectivity in ionic channels. It is yet to be seen whether such a theory can explain selectivity here. Currently studies are being done to simulate these under biophysical conditions.

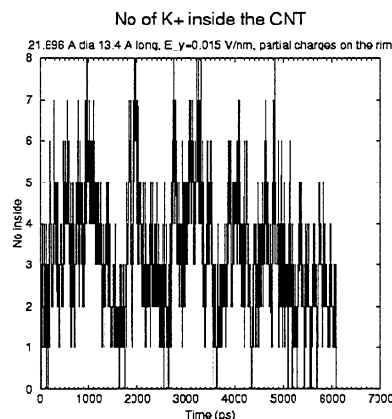
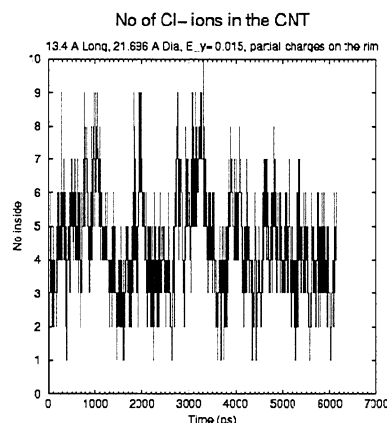


Figure 3: Ion occupancy in a (16, 16) carbon nanotube (13.4 Å long, 21.696 Å diameter) fixed in a solution of 1.85 M KCl with external electric field of $E=0.015$ V/nm and partial charges of $\pm 0.38 e$ on the rim atoms (positive on the top) and negative on the bottom.

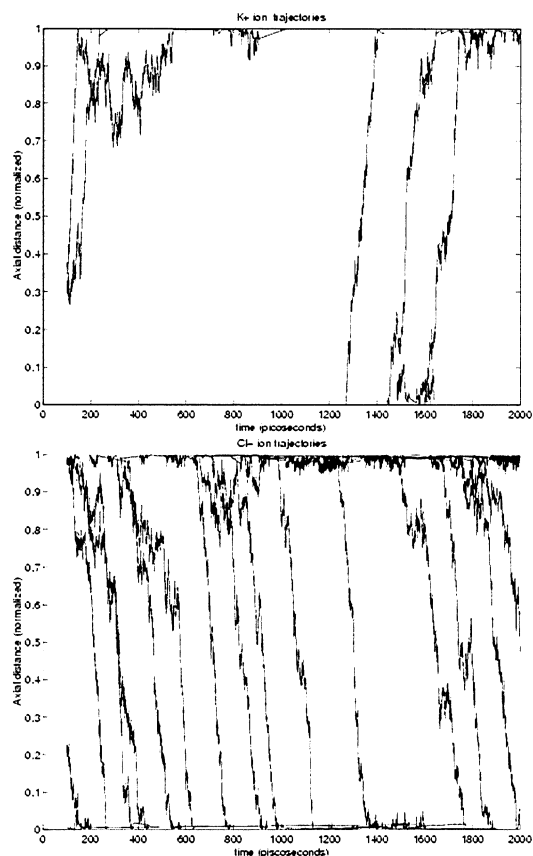


Figure 5: Positions of individual ions (K⁺ on the top and Cl⁻ on the bottom) inside the nanotube along the nanotube axis. The rate of chloride ion passage is higher than the K⁺ ion passage indicating a selectivity of anions over cations.

REFERENCES

- [1] Bayley, H. & Cremer, P. S., Stochastic sensors inspired by biology, *Nature* 413, pp 226-230, 2001
- [2] Woodhouse G. et. al., The ion channel switch biosensor, *J. Mol. Recognit.*, 12, pp 328-334, 1999.
- [3] Kang, Mun-Sik & Martin, C. R., Investigations of potential-dependent fluxes of ionic permeates in gold nanotubule membranes prepared via the template method, *Langmuir*, 17, pp 2753-2759, 2002.
- [4] Li, J. et al., Ion beam sculpting on nanoscale, *Nature*, 412, pp 166-169, 2001.
- [5] Martin, C. R., et al., Investigations of the transport properties of gold nanotubule membranes, *J. Phys. Chem.*, 105, pp 1925-1934, 2001.

[6] Sun, L. & Crooks, R.M., Single carbon nanotube membranes: A well defined model for studying mass transport through nanoporous materials *J. Am. Chem. Soc*, 122, pp 12340-12345, 2000.

[7] Hummer G., Rasaiah, J. C., Noworyta, J. P., Water conduction through the hydrophobic channel of a carbon nanotube, *Nature* ; 414, pp 188-190, 2001.

[8] Miller, S. A. & Martin, C. R., Controlling the rate and direction of electroosmotic flow in template prepared carbon nanotube membranes, *J. Electroanalytical Chemistry*, 522, No 1, pp 66-69, 2002.

[9] Chen, R. J. et. al., Noncovalent sidewall functionalization of single-walled carbon nanotubes for protein immobilization, *J Am Chem. Soc*, 123, pp 3838-3839, 2001.

[10] Chen, Y. et. al., Chemical attachment of organic functional groups to single-walled carbon nanotube material, *J Mater. Res.*, 13, No 9, pp 2423-2431, 1998.

[11] Halicoglu, T. & Jaffe, R. L., Solvent effect of functional groups attached to edges of carbon nanotubes, *Nano Letters*, 2, No. 6, pp 573-575, 2002.

[12] Lindahl, E., Hess, B. and van der Spoel, D., *J. Mol. Mod.*, 7, pp 306-317, 2001.

[13] Nonner W., Catacuzzeno, L., Eisenberg, B., Binding and selectivity in L-type calcium channels: a mean spherical approximation, *Biophysical J.*; 79, pp 1976-1992, 2000.