Insilico Interaction Analysis of Single Wall Carbon Nanotube with Different Ion Channel

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ABSTRACT
Carbon nanotubes have now our day's major role. High usage of carbon nanotubes they are applied in various fields. But in another way it creates a toxicity which is harmful for living beings, animals, and other living organism. So, we identify how biological membranes like ion channel are blocked by carbon nanotubes. SWCNTs of certain diameters can efficiently block K+, Na and other channels. So the purpose of this study to identify the inhibition process by SWCNTs in different ion channels.

Keywords
Single wall Carbon nanotubes, Molecular interaction, Protein data bank.

1. INTRODUCTION
Ion channels are pore-forming proteins that help establish and manage the voltage gradient across the plasma membrane of cells by allocate the flow of ions down their electrochemical gradient in a regulated way. It is found in all types of membranes of cell (plasma membrane) and also intracellular organelles i.e. nucleus, mitochondria, endoplasmic reticulum, Golgi apparatus and so on. Such "multi-subunit" assemblies frequently engage a circular arrangement of identical or the same proteins closely packed around a water-filled pore through the plane of the membrane or lipid bilayer. [1,2] Ion channels make easy targets for these uniqueness external agents, such as natural toxins and synthetic drugs that react with them by set up electrochemical interactions. Thus, blocking agent's carbon nanotubes have been used not only as the basic mechanism for commercial pesticides and potential therapeutic drugs but also to conclude functional information. [3]

Nanotubes are used for biological purposes because of their unique mechanical, chemical, and electrical properties. [4] For example, nanotubes have been successfully used for the helical crystallization of proteins [5] and the growth of embryonic rat brain neurons [6] and as potential biosensors and bioreactors. [7] Carbon nanotubes are ahead increasing attention, due to possible health risks from occupational or environmental exposures. By the usage of nanotubes Lung injury, inflammation, and fibrosis were examined by histopathology, clinical chemistry, ELISA, or RT-PCR for cytokines/chemokines, growth factors, and collagen at 1 and 14 days after inhalation. [8] The identification of new molecules to block ion-channels is of significant interest in biological research, and therefore we required searching the prospect of using novel materials, such as chosen single-walled carbon nanotubes (SWNTs), as ion channel blockers.

2. TOXICITY
Carbon nanotubes can induce harmful effects such as inflammatory and fibrotic reactions. [9] CNTs can enter human cells and accumulate in the cytoplasm, causing cell fatality. [10] The process by which CNTs were synthesized and the types and amounts of metals they contained, CNTs were capable of producing redness, epitheloid granulomas (microscopic nodules), Fibrosis, and biochemical or toxicological changes in the lungs. [11] SWCNTs were more toxic than quartz; it is a serious occupational health hazard when chronically inhaled. As a control, ultrafine carbon black was shown to produce minimal lung responses. [12]

3. USING ION CHANNEL

3.1 1S1G
The family of calcium binding proteins called KChIPs associates with Kv4 family K (+) channels and modulates their biophysical properties. Here, using mutagenesis and X-ray crystallography, we explore the interaction between Kv4 subunits and KChIP1. [13, 14]

3.2 2ZXE
Sodium-potassium ATPase is an ATP-powered ion pump that establishes concentration gradients for Na (+) and K (+) ions across the plasma membrane in all animal cells by pumping Na (+) from the cytoplasm and K (+) from the extracellular medium. Such gradients are used in many essential processes, notably for generating action potentials. Na (+), K (+)-ATPase is a member of the P-type ATPases, which include sarcoplasmic reticulum Ca (2+)-ATPase and gastric H (+), K (+)-ATPase, among others, and is the target of cardiac glycosides. Here we describe a crystal structure of this important ion pump, from shark rectal glands, consisting of alpha- and beta-subunits and a regulatory FXYD protein, all of which are highly homologous to human ones. [15, 16]

3.3 2B8E - The P-type ATPases translocate cations across membranes using the energy provided by ATP hydrolysis. CopA from Archaeoglobus fulgidus is a hyperthermophilic ATPase responsible for the cellular export of Cu+ and is a member of the heavy metal P1B-type ATPase subfamily, which includes the related Wilson and Menkes diseases proteins. The Cu+-ATPases are distinct from their P-type counter-parts in ion binding sequences, membrane topology, and the presence of cytoplasmic metal binding domains,

suggesting that they employ alternate forms of regulation and novel mechanisms of ion transport. [17]

3.4 1SU4 - Calcium ATPase is a member of the P-type ATPases that transport ions across the membrane against a concentration gradient. [18]

3.5 2OAU - The mechanosensitive channel of small conductance (MscS) responds both to stretching of the cell membrane and to membrane depolarization. [19]

3.6 1BL8

The structure of the potassium channel: molecular basis of K+ conduction and selectivity. The potassium channel from Streptomyces lividans is an integral membrane protein with sequence similarity to all known K+ channels, particularly in the pore region. Main chain carbonyl oxygen atoms from the K+ channel signature sequence line the selectivity filter, which is held open by structural constraints to coordinate K+ ions but not smaller Na+ ions. [20]

3.7 About PDB ID

- 2ZXE - Crystal structure of the sodium - potassium pump in the E2.2K+.Pi state.
- 2B8E – CopA ATP Binding Domain.
- 1S1G - Crystal Structure of Kv4.3 T1 Domain.
- 2OAU - Mechanosensitive Channel of Small Conductance (MscS).
- 1SU4 - Crystal structure of calcium ATPase with two bound calcium ions.
- 1BL8 - Potassium channel (kcsa) from streptomyces lividans

Table 1. For each complex, the PDB code, resolution, R-factor.

S. NO	PDB	Resolution (Ao)	R-value (obs.)	R-free	Organism
1.	1BL8	3.20	0.280	0.290	Streptomyces lividans
2.	1S1G	2.60	0.228	0.273	Homo sapiens
3.	1SU4	2.40	0.246	0.283	Oryctolagus cuniculus
4.	2B8E	2.30	0.238	0.297	Archaeoglobus fulgidus
5.	2OAU	3.70	0.293	0.321	Escherichia coli
6.	2ZXE	2.40	0.248	0.271	Squalus acanthias

3.8 APPLICATION

These findings postulate new uses for SWNTs in biological applications and provide unexpected insights into the current view of mechanisms governing the interaction of ion channels with blocking molecules. Ion channel play a physiological role and it has a unique structure. It includes the pore that provides passage for movement of ions across the plasma membrane by the charged, ligand and by electrical or mechanical stimuli. Therefore this makes target the ion channel by external agents such as natural toxin and synthetic drugs. Thus, blocking agents are not only harmful but also help to cure the disease like cancer treatment. .

4. OBJECTIVES

The objective of this study is to predict the carbon nanotubes create a toxicity which is harmful for living beings, animals, and other living organism. So, we identify how biological membranes like ion channel are blocked by carbon nanotubes. So the purpose of this study to identify the inhibition process by SWCNTs in different ion channels.

5. MATERIAL AND METHOD

We use SWCNTs which has a single layer of carbon nanotubes with diameter distributions peaked at (~0.9 and 1.3 nm), Single wall nanotubes (SWCNTs), are form by using nanoengineers and energy are minimizing by the nanoengineers and various types of PDB ID of ion channel are present in the rcsb site which is a protein data bank. By nanoengineer's different form of nanotubes like armchair, chiral and zigzag are formed. We have used different types of ion channel PDB ID: eg. 2OAU, 1BL8, 2B8E, 1SU4, 1S1G, 2ZXE by taking this, the docking process start which is a protein-protein interaction. For docking, patch dock and fire dock web server are used which is an online process. The academic algorithms have been designed to predict the three-dimensional structure of protein-protein and protein-ligand complexes by a procedure called docking. [21] By putting the value of receptor and ligand in the patch dock they give the result and go for the refinement in the Fire dock which gives the 10 best solutions and 1000 solution.

To prove our hypothesis that SWCNTs blocks the ion channel pores and thus interfere with the normal movement of ions across membrane. In a docking method we performed docking between SWCNTs model with different ion channel protein (PDB id: 2OAU, 1BL8, 2ZXE, 1S1G, 2B8E and 1SU4) using Patch Dock program. The result obtained by Patch Dock is further refined by Fire Dock program. [22, 23] Fire Dock utilizes Monte Carlo simulation and provides the ranking of docking results according to the binding energy. First of all we minimize the energy of nanotubes. Docking complexes with minimum energy have been simulated for a total of 900 frames with 1 frame per femto second at the temperature of 300 K. The interaction between each non-charged site in the nanotube was described by the Lennard Jones potential.

VL−J rij = 4ij ij/rij 12− ij/rij 6

And interaction of charged ions described by coulomb potential [24]

Vc (rij) =1/4Π€0 ZiZje/2ri

Where, rij is distance between the ith and jth atoms of different molecule, Zn is the charges on each nth site, e is electron charge, and are Lennard-Jones potential parameters used in the simulation and are calculated by the Lorentz Berthelot combining rule

ij = i +j 2 ij = √ij [25]

All complexes deposited at PDB with a resolution and R-factor less than 3.0 and 0.2 respectively, were taken for the study (http://www.rcsb.org/pdb/home/home.do). Six ion channel complexes were taken from Protein Data Bank in the form of PDB format.

5.1 RESULT AND DISCUSSION

In the present study Docking and simulation performed between SWCNTs and crystal structure of different ion channels (2OAU, 1SU4, 2ZXE, 1BL8, 2B8E, and 1S1G) provide insights in to the geo-metrical basis of molecular interactions. PatchDock and FireDock results suggest that the interaction between SWCNTs and ion channels with the extracellular domain shown and nanotube are red and blue in colour [Fig. 1 to 13]. However, the interaction is largely depends on the dimension and size of nanotubes [Table 2 & 3].This table shown SWCNTs with different armchair and chiral nanotube with different values of length, diameter and minimizing energy. Molecular dynamics simulation performed for these complexes using GROMACS shows that the interaction between SWCNTs and ion channel protein tend to acquire the minimum total energy and reach to the stability. In previous studies confirms that SWCNTs can be concerned about that as the ion channel is the new class of inhibitor as identified. [11–13] we suggest that toxicity by the SWCNTs is due to the blockage of the ion channel passage through which ions passes from extracellular medium to cytoplasm and vice versa. [25]

5.1.1.1 Subsubsections

The heading for subsubsections should be in Times New Roman 11-point italic with initial letters capitalized.

5.1.1.2 Subsubsections

The heading for subsubsections should be in Times New Roman 11-point italic with initial letters capitalized.

6. CONCLUSION

We use ion channels which have very important in biological field as target to identify the blockage by single walled carbon nanotubes (SWCNTs) as a novel material. Here we show that SWCNTs of certain diameters can efficiently block K^+, Na and other channels.

7. ACKNOWLEDGEMENT

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Table 2. SWCNTs have different types, diameter, length and minimizing atoms (M).

S.No	Types	N,M	Diameter (Å)	Length (Å)	M
1.	Armchair	5,5	6.8346	4.55	390 atoms
2.	Armchair	12,12	16.304	9.496	264 atoms
3.	Chiral	15,5	14.1458	6.497	170 atoms
4.	Chiral	12,1	9.844	6.688	114 atoms
5.	Chiral	12,5	11.8826	3.00	500 atoms
6.	Chiral	7,2	6.4608	5.99	72 atoms

Table 3. SWCNTs have different types, Finding Force rms value (F.F), evals, Modeling energy (M.E) and Bond Length (B.L).

S. No	Types	F.F rms	High	M.E	evals	B.L
1.	Armchair	658.37 pN	969.65pN	35.643 aJ	2047,1021	1.42
2.	Armchair	599.79 pN	945.076pN	4.833aJ	2900,1477	1.42
3.	Chiral	0.80275pN	1.341 pN	3.326aJ	3786,754	1.42
4.	Chiral	0.949pN	1.738 pN	4.136aJ	16873,1123	1.42
5.	Chiral	0.9611 pN	1.688 pN	16.893 aJ	14634,693	1.42
6.	Chiral	0.9946 pN	1.477 pN	5.071aJ	5368,426	1.42

Figure

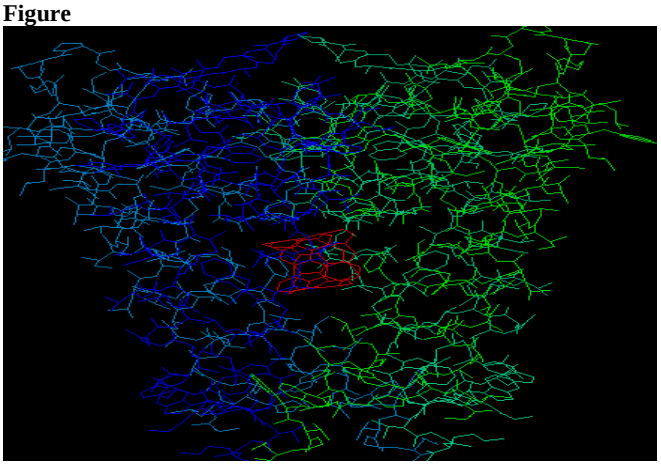

Figure1. 1bl8 ionchannel dock with nanotube 6

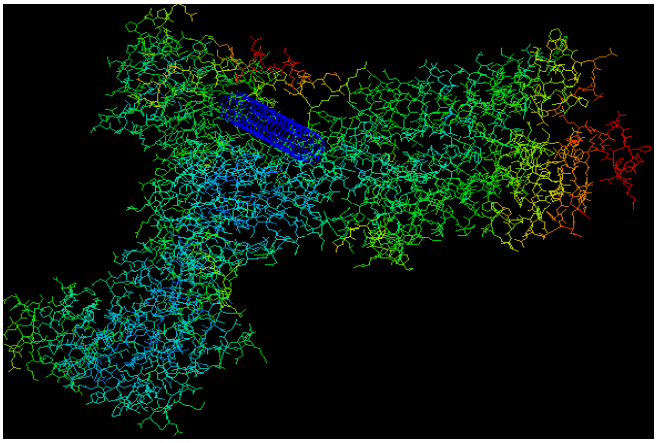

Figure2. 1su4 ionchannel dock with nanotube 3

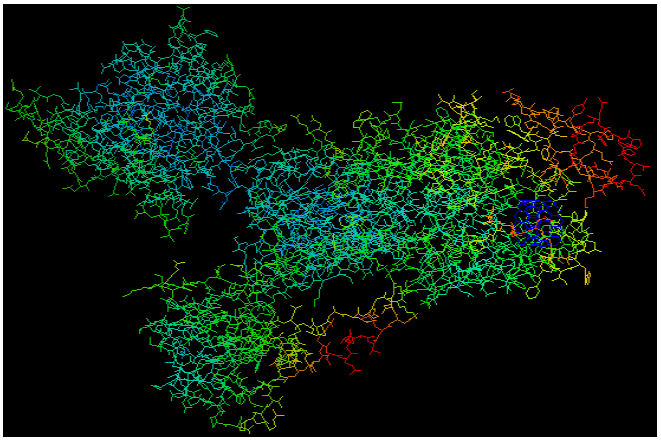

Figure3. 1su4 ionchannel dock with nanotube 4

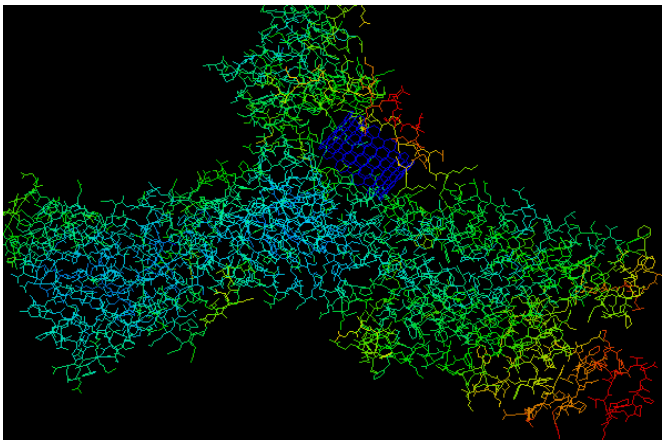

Figure 4. 1su4 ionchannel dock with nanotube 7

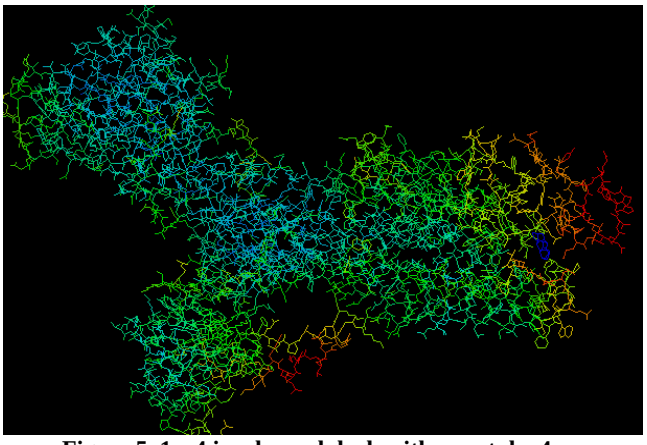

Figure 5. 1su4 ionchannel dock with nanotube 4

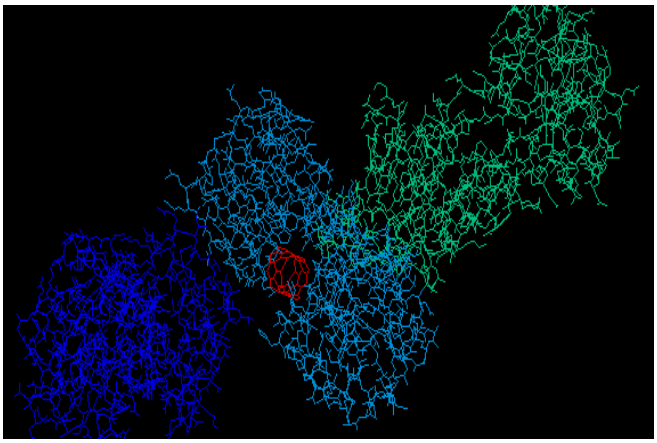

Figure 6. 2b8e ionchannel dock with nanotube 6

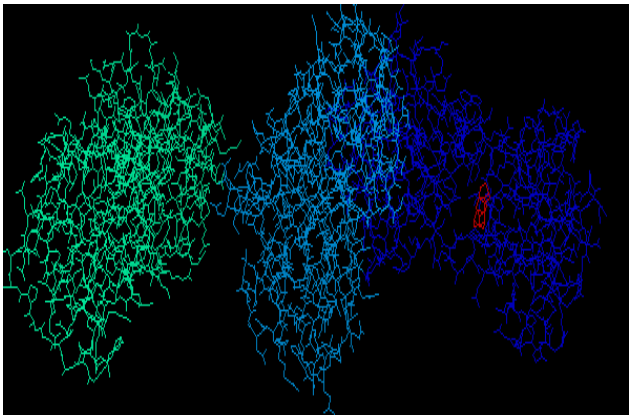

Figure7. 2b8e ionchannel dock with nanotube 1

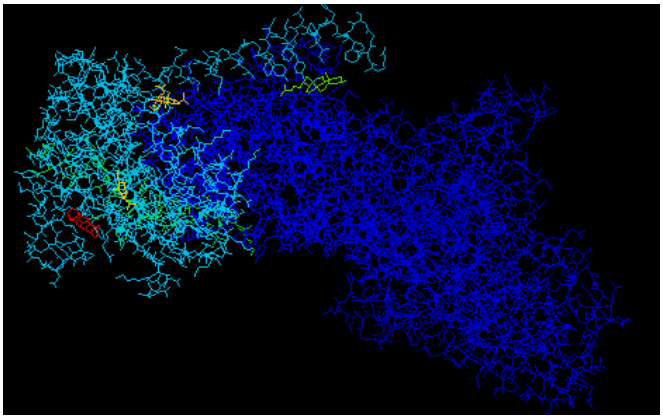

Figure8. 2zxe ionchannel dock with nanotube 1

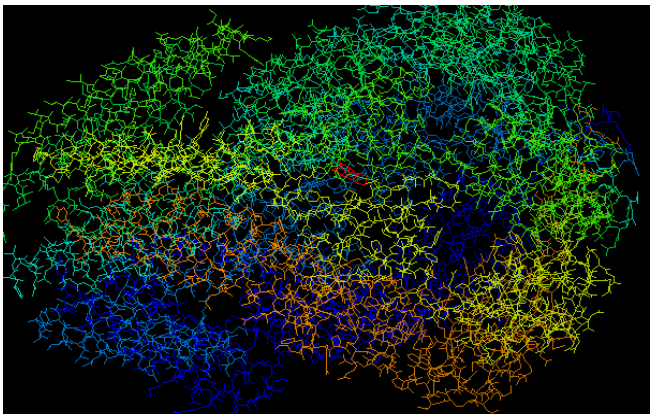

Figure10. 2oau ionchannel dock with nanotube 1

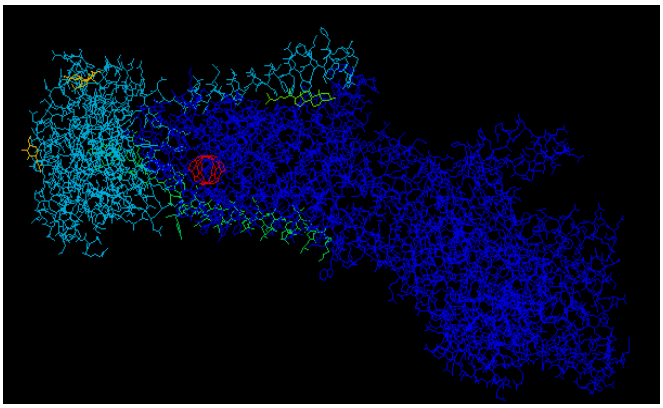

Figure9. 2zxe ionchannel dock with nanotube 6

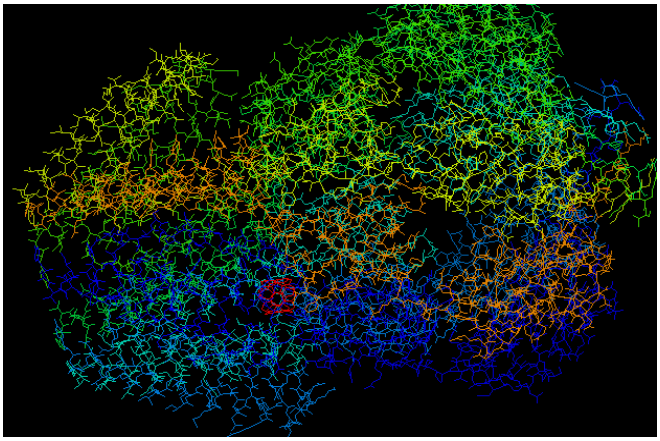

Figure11. 2oau ionchannel dock with nanotube 6

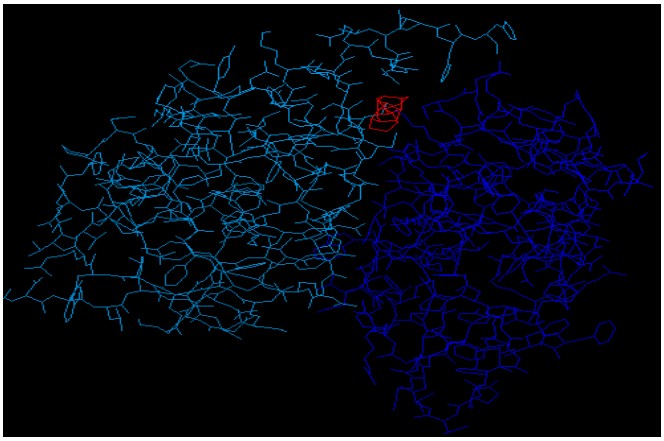

Figure12. 1s1g ionchannel dock with nanotube 1

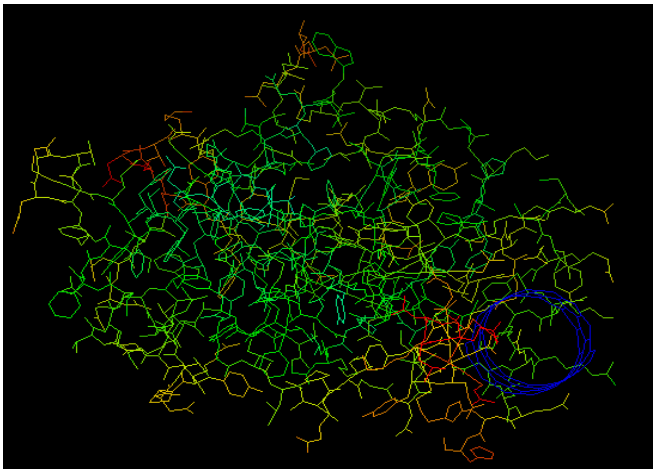

Figure13. 1s1g ionchannel dock with nanotube 4

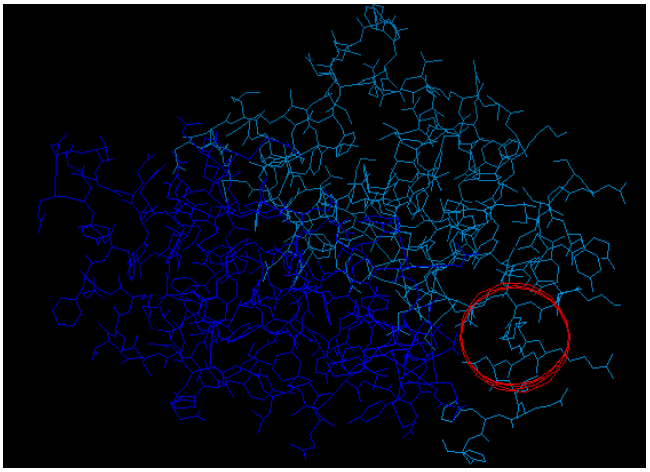

Figure13. 1s1g ionchannel dock with nanotube 4