

pH-DEPENDENCE OF THE OPTICAL BIO-SENSOR BASED ON DNA-CARBON NANOTUBE

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Abstract. *In 2006, Daniel A. Heller et al. [1] demonstrated that carbon nanotubes (CNNTs) wrapped with DNA can be placed inside living cells and detect trace amounts of harmful contaminants using near infrared light. This discovery could lead to new types of optical sensors and biomarkers at the sub cellular level. The working principle of this optical bio-sensor from DNA and CNNTs can be explained by a simple theoretical model which was introduced in [3]. In this paper, the pH-dependence of DNA and the pH-dependence of solution around CNNTs are shown by using data analysis method. By substituting them into the same model, the pH-dependence of DNA-wrapped CNNTs was elicited in this paper. The range of parameters for workable conditions of this bio-sensor was indicated that the solution should have pH from 6 to 9 and the concentration of ions should be more than a critical value. These results are according to the experimental data and the deduction about pH and salt concentration in solution. They are very useful as using such a new bio-sensor like this in living environment.*

I. INTRODUCTION

Since the first time they were described [2], Carbon nanotubes have been the focus of intensive theoretical and experimental studies, due to their potential applications which utilize their unique electronic and mechanical properties. Many works have been carried out to investigate the electronic and optical properties of carbon nanotubes. All these works provided a basic backgrounds and theories to understand the physical properties of CNNTs and open new prospects of CNNT applications on nanotechnology. For instance, CNNTs can be used for optical nanosensors, because they typically fluoresce in the near infrared (n-IR) spectral region, where human tissue and biological fluids are characteristically transparent.

At the same time, a new branch of nanotechnology which uses the bio-molecules as structural materials has also developed and opened many new prospects. As in the case of Deoxyribonucleic acid (DNA), it is very famous as a nucleic acid that contains the genetic instructions used in the development and functioning of all known living organisms and some viruses. The main role of DNA molecules is the long-term storage of information. Recently, DNA nanotechnology uses the unique molecular recognition properties of DNA and other nucleic acids to create nano-sensors, nano-machines, etc with useful properties. DNA is thus used as a structural material rather than as a carrier of biological information.

Furthermore, these two types of materials, inorganic and organic materials, can be combined in one micro device. As shown in [1], sigle-walled nanotubes (SWNTs) wrapped

with DNA can be placed inside living cells in order to detect trace amounts of harmful contaminants at the subcellular level. This combination is due to the Carbon-structure of SWNTs and net negative charge of DNA molecule. And DNA-wrapped carbon nanotubes serve as sensors in living cells. This is the first nanotube-based sensor that can detect analytes at the subcellular level. When the DNA is exposed to ions of certain atoms (e.g., calcium, mercury and sodium) the DNA changes shape, perturbing the electronic structure of sigle-walled nanotube(SWNT) and shifting the nanotube's fluorescence to lower energy. As shown in [3], D.P.Hung et al. have discussed the working principle of the this type of optical bio-sensor SWNT-DNA. By using the simple theoretical model and the theory of exciton in CNNTs, he has shown that the simple model can explain the working principle of this optical bio-sensor from DNA and CNNTs, vs the concentration of cations in solution.

On the other hand, pH is a measure of the acidity or alkalinity of a certain solution. In the living body, pH is different in different organs. Or even in the same organ, the pH is also various in different parts. Moreover, the pH of certain solution drastically impacts the bio-molecules such as DNA, RNA, proteins ect. Without doubt, this optical bio-sensor based from DNA and CNNTs is effected by pH of solution in some way, and investigating its properties vs pH is an indispensable job. In this paper, we have shown the working principle of this optical bio-sensor vs pH of solution. Our results indicated the range of parameters for workable conditions of bio-sensor.

II. MODEL

II.1. Theory framework

According to the experimental model of the CNNTs wrapped with DNA which serve as bio-sensor in living cell, a theoretical model was introduced in [3]. In this model, the exciton theory of CNNTs in [5] was used to explain the fluorescence of the bio-sensor. Here, to approximate the dielectric constant of the B- or Z-DNA wrapped CNNT, the effective medium and effective dielectric constant was introduced. The dielectric constant in the expression of exciton binding energy [5]

$$E_{\text{bind}} = AR^{\alpha-2}\mu^{\alpha-1}\varepsilon^{-\alpha}, \quad (1)$$

was substituted by the effective dielectric constant of effective medium surrounding SWNT (a.u.). In the expression, R is the radius of SWNT (nm); μ is reduced effective mass (m_0) here m_0 is mass of free electron; A and α are fitting constant they have value of $24.1(\text{eVnm}^{-3/5})$ and $1.4(\text{a.u.})$ respectively. DNA was considered as a ribbon with the width of w (nm). This ribbon regularly wraps around surface of cylinder radius of R (nm) with period along the axis of cylinder b (nm) (see Fig. 1). Therefore, the effective dielectric constant of medium surrounding SWNT can be written as [3]

$$\varepsilon = f\varepsilon_{\text{DNA}} + (1 - f)\varepsilon_s, \quad (2)$$

where ε_{DNA} and ε_s are dielectric constants of DNA and solution, respectively; f is the ratio of surface area covered by DNA per total cylindrical surface area.

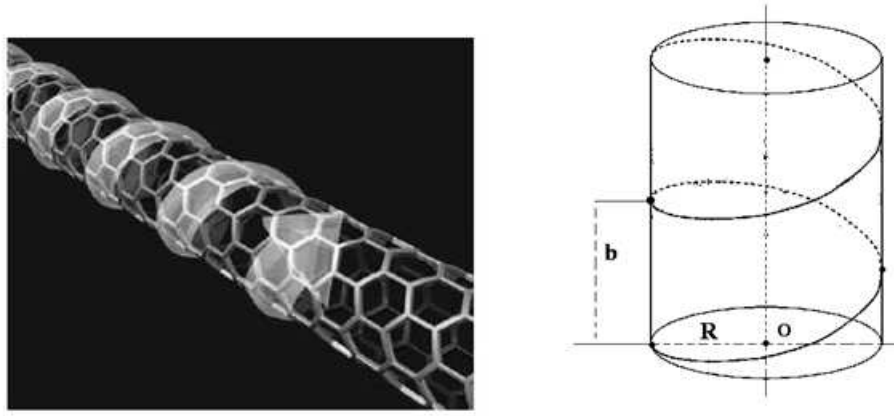


Fig. 1. Theoretical model of optical bio-sensor.

By using the model of helical spring for DNA [4], the expression of the ratio f was obtained as a function of structural parameters of DNA and radius of SWNT:

$$f = \frac{w}{2\pi R} \sqrt{\frac{4\pi^2 r_0^2 + b_0^2}{4\pi^2 r_0^2 + b_0^2 - 4\pi^2 R r_0}}, \quad (3)$$

Note that, the electronic structure of SWNT change with changing the structure of DNA, so that the energy of nanotube’s natural n-IR fluorescence is changed.

When the bio-sensor is in the certain medium where the ionic concentration exceed a critical value, the structure of DNA will be changed over from B to Z form. According to Eq. 1, the emission energy of SWNT was shifted. The deviation of exciton binding energy can be expressed as

$$\Delta E_{B \rightarrow Z} = E_{\text{bind-B}} - E_{\text{bind-Z}} = AR^{\alpha-2} \mu^{\alpha-1} \left(\frac{1}{\varepsilon_B^\alpha} - \frac{1}{\varepsilon_Z^\alpha} \right). \quad (4)$$

II.2. Experimental parameters

When the pH of solution varies, the dielectric constants of DNA and solution around the CNNTs change, it brings about the variation of effective dielectric constant. Therefore, the optical signals of our sensor change. Hence, we should start to investigate from the pH-dependence of dielectric constants of component parts, DNA and CNNTs.

First, the pH dependence of the dielectric parameters of DNA was investigated in [7] by experiences.

In the Fig. 2 left, [7] the dielectric increment $\Delta\varepsilon$, which is a measure of the magnitude of the dielectric dispersion is given by: $\Delta\varepsilon = \varepsilon_s - \varepsilon_\infty$ where ε_s and ε_∞ are the low-frequency and high-frequency relative permittivities describing the relaxation process, respectively.

In our problem, we just paid attention in the dielectric constant at low-frequency. Because ε_∞ is quite invariable and its value is around water’s one. In our computation, we seted it equal to 80.

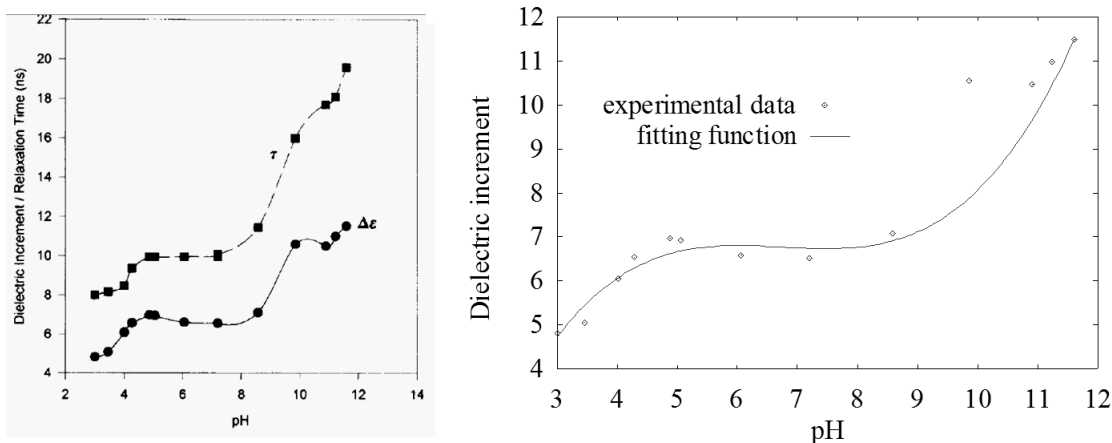


Fig. 2. Variation of dielectric increment and relaxation time with pH [7]. Right: Experimental data [7] and fitting function of DNA dielectric increment vs pH.

After fitting the experimental data with a polynomial of the third order (Fig. 2 right)

$$\Delta\epsilon_{DNA}(pH) = \epsilon_s - \epsilon_\infty = A \cdot pH^3 + B \cdot pH^2 + C \cdot pH + D, \quad (5)$$

the fitting constants A, B, C, D should have values of 0.044, -0.89, 5.93, -6.24 respectively.

For semiconducting nanotubes, reaction selectivity is easily followed in the absorption spectrum as a function of solution pH [8]. The marked pH dependence and complete reversibility suggest an equilibrium reaction of the nanotube (SWNT) with a number of free protons in solution ($n[H^+]$) resulting in a protonated nanotube complex [P] that has an overall diminished absorption cross section.



Here K_p is the reaction equilibrium constant. Scaling the absorption intensities to yield the fraction of reacted nanotubes and substituting the equilibrium relation above yield:

$$\frac{A(pH) - A_p}{A_d - A_p} = \frac{K_P}{[H^+]^n + K_P} = \frac{K_P}{10^{-(npH)} + K_P}. \quad (6)$$

In Figure 3 the absorbance for two different band gap semiconducting nanotubes is plotted as a function of pH, and values for n and K_p were regressed using this equilibrium-limited protonation model with A_p and A_d as the absorption intensities of the protonated and deprotonated states and K_p as the equilibrium constant. Values for $\ln(K_p)$ range from -36.39 for the (12,5) nanotube (0.83 eV band gap) to -33.97 for the (8,3) tube (1.3 eV band gap) The steepness of the curve in Figure 3 is a measure of n or the average number of protons reacting per protonated entity and was determined to be 3. Assignment of optical band gap energies with the (n,m) chirality index was possible using recently successes in nanotube spectroscopy.

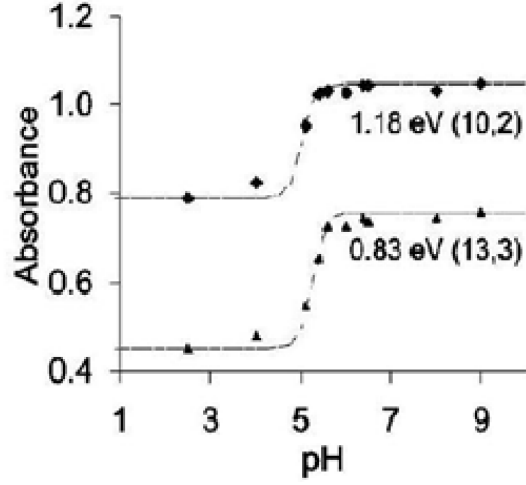


Fig. 3. Spectrophotometric titration of individually dispersed carbon nanotubes in SDS suspension. Absorbance plotted as a function of pH for two particular semiconducting nanotubes. The trend is characteristic of an equilibrium-limited surface reaction. Smooth curves represent the regressed model (eq 6) with $n = 3$ and best fit value of K_P for each nanotube.

According to equation 6, the pH - dependent absorbance of SWNT can be written as follow:

$$A(pH) = (A_d - A_p) \frac{K_p}{10^{-(npH)} + K_p} + A_p. \quad (7)$$

Otherwise, the absorbance of photons is given by the golden rule

$$A(pH) = Const \cdot \frac{e^2}{\varepsilon_s(pH)} |\langle \hat{\varepsilon} \cdot p_{cv} \rangle|^2 \delta(E_v + \hbar\omega - E_c). \quad (8)$$

Here, the matrix element $\langle \hat{\varepsilon} \cdot p_{cv} \rangle$ is between the one-electron initial and final states. It does not depend on the pH. Therefore, the pH-dependent parameter can only be dielectric constant.

If the SWNT is in the normal solution at a neutral pH, the dielectric constant is ε_0 (equal to the dielectric constant of water of 80), the neutral absorbance of photons A_0 would be:

$$A_0 = \frac{e^2}{\varepsilon_0} |\langle \hat{\varepsilon} \cdot p_{cv} \rangle|^2 \delta(E_v + \hbar\omega - E_c). \quad (9)$$

Dividing (8) to (9), and substituting (7) into the obtained equation then gives:

$$\frac{\varepsilon_s(pH)}{\varepsilon_0} = \frac{A_0}{(A_d - A_p) \frac{K_p}{10^{-(npH)} + K_p} + A_p}. \quad (10)$$

Thus, the dielectric constant versus pH curves is illustrated in Fig. 4.

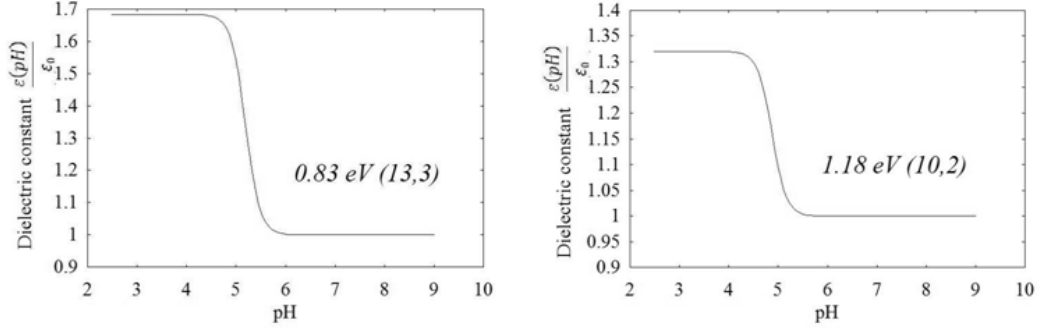


Fig. 4. The dielectric constant of solution around SWNT (13,3) and (10,2) vs pH.

III. RESULTS

Dielectric constants of DNA and solution around CNNT are varied when the solution pH changes. So, the pH dependence of effective dielectric constant will be expressed as

$$\varepsilon(pH) = f\varepsilon_{DNA}(pH) + (1 - f)\varepsilon_s(pH). \quad (11)$$

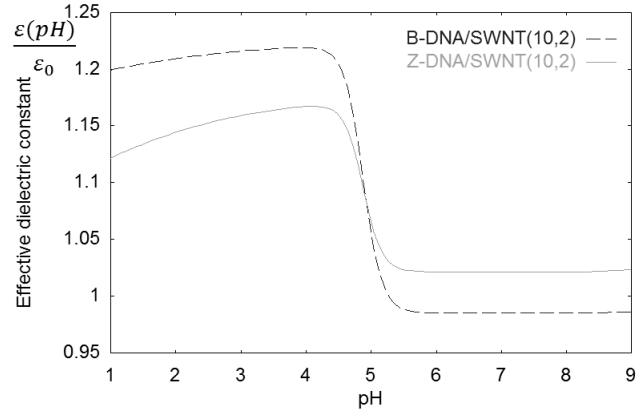


Fig. 5. The Effective dielectric constant in our model vs pH. The dash line insulates $\varepsilon(pH)$ of the sensor based on B-DNA around SWNT (10,2) and the solid one insulates $\varepsilon(pH)$ of sensor based on Z-DNA around SWNT (10,2).

And the effective dielectric constant of solution around sensor versus pH curves is illustrated in Fig. 5

According to the equation 1, the exciton binding energy of SWNT in the solution would be written as follow:

$$E_{\text{bind}}(pH) = AR^{\alpha-2}\mu^{\alpha-1}\varepsilon^{-\alpha}(pH), \quad (12)$$

and the neutral exciton binding energy is:

$$E_{\text{bind}}^{(0)} = AR^{\alpha-2}\mu^{\alpha-1}\epsilon_0^{-\alpha}, \quad (13)$$

thus

$$\frac{E_{\text{bind}}(pH)}{E_{\text{bind}}^{(0)}} = \left(\frac{\epsilon_0}{f\epsilon_{DNA}(pH) + (1-f)\epsilon_s(pH)} \right)^\alpha. \quad (14)$$

The pH-dependence of the exciton binding energy is insulated in Fig. 6.

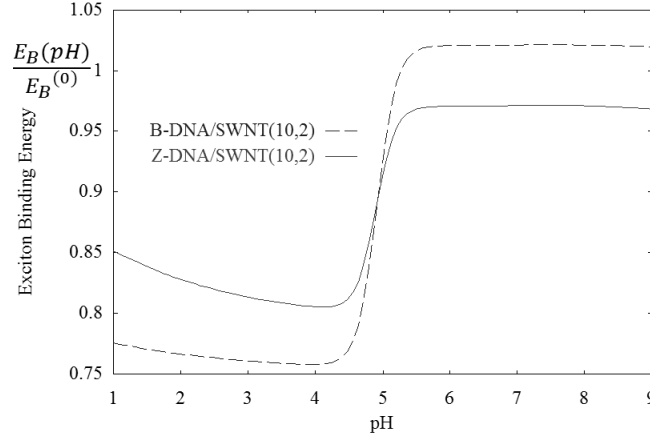


Fig. 6. The Exciton Binding Energy of model vs pH. The dash line insulates E_{Bind} of the sensor based on B-DNA around SWNT (10,2) and the solid one insulates E_{Bind} of sensor based on Z-DNA around SWNT (10,2).

The exciton energy would be written as: $E_{\text{exc}} = E_g - E_{\text{bind}}$, thus

$$\frac{E_{\text{exc}}(pH)}{E_{\text{bind}}^{(0)}} = \frac{E_g - E_{\text{bind}}(pH)}{E_{\text{bind}}^{(0)}} = \frac{E_g}{E_{\text{bind}}^{(0)}} - \left(\frac{\epsilon_0}{f\epsilon_{DNA}(pH) + (1-f)\epsilon_s(pH)} \right)^\alpha. \quad (15)$$

And the Fig. 7 shows the pH dependence of the exciton energy.

We should note you that, in our computations, we considered the concentration of salt is stable enough for DNA to stay in one form as the pH changes. After that, the increment energy for the SWNT (10,2) was calculated and compared with the experimental data [1] for this kind of SWNT (Fig. 8). The data was the energy shift of a DNA-SWNT solution with $0\mu M$ $HgCl_2$ and $52.37mM$ $HgCl_2$.

IV. CONCLUSIONS

By using a simple model, we have investigated the properties of sensor depends on environment around it, especially on solution pH. We have demonstrated that the sensor is influenced drastically by the protons (H^+) in solution, and the pH dependence is shown in (Fig. 7).

We can see that, the workable solution for sensor should has pH from 6 to 9. Beside the reason that beyond this range, the DNA will be denatured, the concentration of H^+ in that range is small enough so that their effects can be ignored comparing with the cation's.

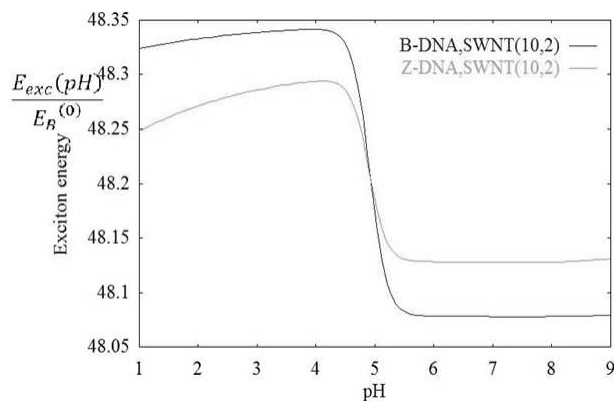


Fig. 7. The Exciton Energy of model vs pH. The dash line insulates E_{exc} of the sensor based on B-DNA around SWNT (10,2) and the solid one insulates E_{exc} of sensor based on Z-DNA around SWNT (10,2)

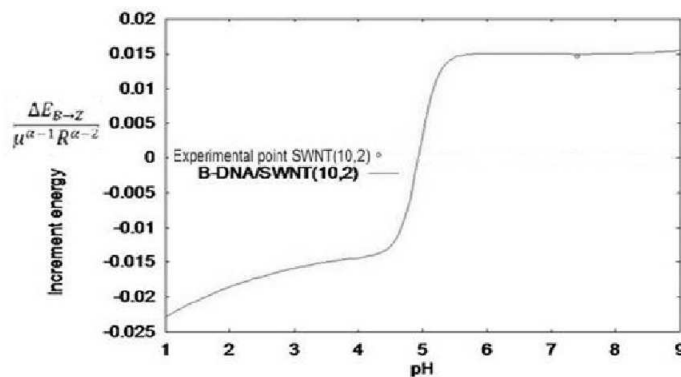


Fig. 8. The Increment Energy of SWNT (10,2) sensor vs pH and experiment point [1].

Another result, when using the sensor, we have to care about the workable conditions. The concentration should be more than a critical value so that, the proton effects can be ignored. Moreover, the sensor signal should be regulated because of pH disturbance.

This new combining structure of DNA and CNNTs is really interesting. [9, 10] are the newest papers about this combination. In the future, we will try to improve our model, by comparing with the experiment results. Therefore, we can choose the best parameters for the model. The sensors' properties depend on temperature, pKa, pressure, etc are still such interesting ways for studying more.

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