

**2013 Nanohub-U Course on
“Principles of Electronic Nanobiosensors”
Sensitivity of Potentiometric and Amperometric Sensors**

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In this 3rd HW set, we will discuss three topics: The origin of charges of a biomolecule such as DNA or protein, beating the screening limit by super-Nernst sensors.

Problem 3.1: Screening of charge on a spherical sensor surface

Solution:

Spherical coordinates are used in this solution to calculate the potential across a double layer. Consider a sphere of radius ‘a’, (typically a few nm). The Poisson-Boltzmann equation in the Debye approximation can be written as

$$\nabla^2 \psi = \frac{1}{r} \frac{d^2 (r\psi)}{dr^2} = \frac{\psi}{L_D^2}$$

The boundary conditions are

$$r \rightarrow \infty, \psi = 0$$

$$r = a, \text{ applying the Gauss law: } E_{r=a^-} = -\left(\frac{d\psi}{dr}\right)_{r=a^-} = \frac{Q}{4\pi\epsilon_r\epsilon_0 a^2}$$

The general solution is of the form:

$$r\psi = A_1 \exp\left(\frac{r}{L_D}\right) + A_2 \exp\left(-\frac{r}{L_D}\right)$$

Applying the boundary conditions

$$r \rightarrow \infty, \psi = 0 \Rightarrow A_1 = 0$$

$$r = a, E_{r=a^-} = \left(\frac{d\psi}{dr}\right)_{r=a^-} = \frac{Q}{4\pi\epsilon_r\epsilon_0 a^2} \Rightarrow \frac{A_2}{a} \left(\frac{1}{L_D} + \frac{1}{a}\right) \exp\left(-\frac{a}{L_D}\right) = \frac{Q}{4\pi\epsilon_r\epsilon_0 a^2}$$

The final solution is:

$$\psi = \frac{Q}{4\pi\epsilon_r\epsilon_0 r} \left(\frac{L_D}{L_D + a}\right) \exp\left(-\frac{(r-a)}{L_D}\right)$$

$\psi = \frac{Q}{4\pi\epsilon_r\epsilon_0 r} \exp(-r/L_D)$ with $a \rightarrow 0$. Apart from the exponential screening term, this is the potential profile associated with a point charge Q at a distance r from the source.

Problem 3.2: Biomolecules and Screening at low salt concentration.

Let us reconsider the protein discussed in Problem 3.3. We were told that the protein can be viewed as a sphere of radius $a=2\text{nm}$, with a total charge of $20q$.

- Calculate the screening length by using the formula $L_D^2 = \epsilon_r \epsilon_0 k_B T / 2z^2 I_0 N_{av} q^2$.
- Use the formula derived in Problem 3.2 to find the potential 3 nm from the center of the protein molecule (i.e. 1 μm away from the surface).

Solution:

For $I_0 = 0.05M$ for NaCl $z = 1$, $L_D^2 = \epsilon_r \epsilon_0 k_B T / 2z^2 I_0 N_{av} q^2 \Rightarrow L_D = 1.4\text{nm}$

Applying the potential formula $\psi = \frac{Q}{4\pi\epsilon_r \epsilon_0 r} \left(\frac{L_D}{L_D + a} \right) \exp\left(\frac{-(r-a)}{L_D} \right)$

we have $\psi(r = 3\text{nm}) = \frac{20q}{4\pi\epsilon_r \epsilon_0 r} \cdot \left(\frac{1.4}{1.4 + 2} \right) \cdot \exp\left(\frac{-(3-2)}{1.4} \right) \sim k_B T / q$

Problem 3.3: Debye length vs. Bjerrum lengths.

Solution:

- To solve for Bjerrum length (l_B)

$$\psi(r = l_B) = \frac{Q}{4\pi\epsilon_r \epsilon_0 (l_B)} \left(\frac{L_D}{L_D + a} \right) \exp\left(\frac{-(l_B - a)}{L_D} \right) = k_B T / q,$$

For large L_D

$$\psi(r = l_B) = \frac{Q}{4\pi\epsilon_r \epsilon_0 l_B} = k_B T / q \Rightarrow l_B = \frac{qQ}{4\pi\epsilon_r \epsilon_0 k_B T}$$

- For a unit charge $Q = q$, $l_B = \frac{q^2}{4\pi\epsilon_r \epsilon_0 k_B T}$, for air $\epsilon_r = 1 \therefore l_B = \frac{q^2}{4\pi\epsilon_0 k_B T} = 56\text{nm}$

For water $\epsilon_r = 78.9 \therefore l_B = \frac{q^2}{4\pi\epsilon_r \epsilon_0 k_B T} = 0.71\text{nm}$

- The center-to-center distance between base pair is $\sim 1\text{nm}$. Therefore, the repulsion between the charges of the DNA strands are considerably screened because the Bjerrum length is always lesser than the center-to-center distance between base pair, regardless the salt concentration in the water medium in which the DNA molecule is present.

Problem 3.4: Charge of a biomolecule as a function of pH.

A researcher is using a pH range from 2 to 8 to demonstrate a particular effect associated with DNA molecules. He claims that the charge of the DNA molecules is independent of pH. Based on the figure below, do you think that this claim is justified?

Solution:

Only half of the $[H_3PO_4]$ molecules are singly ionized at pH=2 (0.5 charge/atom), while the proportion of singly and doubly charged molecules are 1:1 at pH=7 (1.5/atoms). Therefore, the charge concentration changes by a factor of 3. Therefore, the assertion of the researcher is incorrect. Even if the sensor failed to catch any target biomolecules whatsoever, the change in the pH concentration will change the charge of the ionization of the receptor molecule, and thereby give a false impression of analyte capture.

Problem 3.5: Calculating charge of a protein molecule as a function of pH.

- (a) Calculate the electric charge of Prostate Specific Antigen (PSA) molecule (2ZCH:H) -- which has 229 amino acids connected by peptide bonds -- as a function of pH and show that point of zero charge occurs at $pH \sim 7.1$.

Use the total charge formula

$$Q = q \left(\sum_i^{Positive\ AA} i \frac{1}{1 + m_i} + \sum_j^{Negative\ AA} j \frac{-1}{1 + m_j} + \frac{1}{1 + m_{NH_2}} + \frac{-1}{1 + m_{COOH}} \right)$$

where i corresponds to K, R, and H, and j corresponds to D, E, C, and Y. The table below summarizes the number of the charged amino acids as well as their reaction constants (α , β , ... in the protein charge equation in lecture 14) in the PSA sequence.

Amino Acid	K	R	H	D	E	C	Y
Count (i or j)	11	5	2	11	5	8	11
pK	10	12	6.5	4.4	4.4	8.5	10
m	10^{pH-10}	10^{pH-12}	$10^{pH-6.5}$	$10^{4.4-pH}$	$10^{4.4-pH}$	$10^{8.5-pH}$	10^{10-pH}

Terminal group	NH ₂ (+ve)	COOH (-ve)
Count	1	1
pK	8	3.1
m	10^{pH-8}	$10^{3.1-pH}$

- (b) Follow the calculate the

more sophisticated calculator and compare the results obtained from the simple calculation above. How do the results compare.

instructions below to protein charge using a

- Step 1. Go to the Protein Data Bank website: <http://www.rcsb.org/pdb/home/home.do>
 Step 2. Search the protein that we are looking for by typing names like 'Prostate Specific Antigen' in the search box.

Step 3. When the results are displayed, scroll down the page, where you will see variants of protein specific antigen displayed. For now, choose one such as 2ZCH:H. It will take you to a page like <http://www.rcsb.org/pdb/explore/explore.do?structureId=2ZCH>. Note that if you had searched for a different protein, such as breast cancer marker HER2 and clicked 1N8Z variant from search results, you will be directed

to <http://www.rcsb.org/pdb/explore/explore.do?structureId=1N8Z>

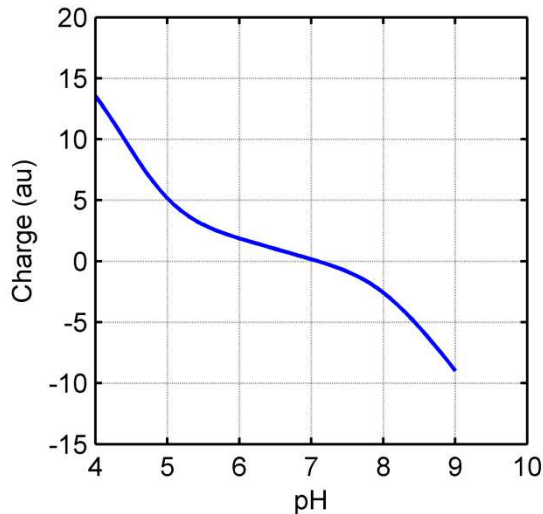
Step 4. Click the Display Files at the upper right corner of the webpage associated with the protein.

Step 5. Click FASTA sequence and then the sequence will be displayed on the webpage. FASTA is name of the format used by a program to align DNA and protein database searches. Here we use FASTA just to get the sequence of DNA or Amino acid -- we will not be using the FASTA program.

Step 6. For running protein calculator, copy the sequence it and paste to the designated place in the calculator. For example, you can use <http://www.scripps.edu/~cdputnam/protcalc.html>.

Solution:

The Charge vs. pH plot would then be:



Problem 3.6: Beating the Nernst response by NP-NW sensors.

Consider an accumulation-mode NP-NW transistor pair. What is the maximum pH sensitivity for a such device with AlGa_N serving as the channel of the planar FET (T1), and Si-NW as the NW-FET (T2) with the following properties?

- AlGa_N has mobility of 2000 cm²/V.s, Tox=45nm, W=1um (T1)
- Si-NW has mobility of 100 cm²/V.s, Tox=15nm, W=50nm (T2)
- $L_1=L_2$, $V_{DS1}=V_{DS2}$

Solution:

For a GN device we have:

$$\frac{\Delta V_G^{NP-NW}}{\Delta pH} = 59 \frac{mV}{pH} \left(\frac{\mu_1 (W/L)_1 V_{DS,1}}{\mu_2 (W/L)_2 V_{DS,2}} \right) \left(\frac{C_{OX,1}}{C_{OX,2}} \right)$$

Also, $\mu_1 / \mu_2 = 20$, $W_1 / W_2 = 1000 / 50 = 20$, and $C_{ox1} / C_{ox2} = T_{ox2} / T_{ox1} = 1 / 3$.

Therefore:

$$\text{Sensitivity} \leq (60)(20)(20) / 3 = 8 \text{ V/pH}$$

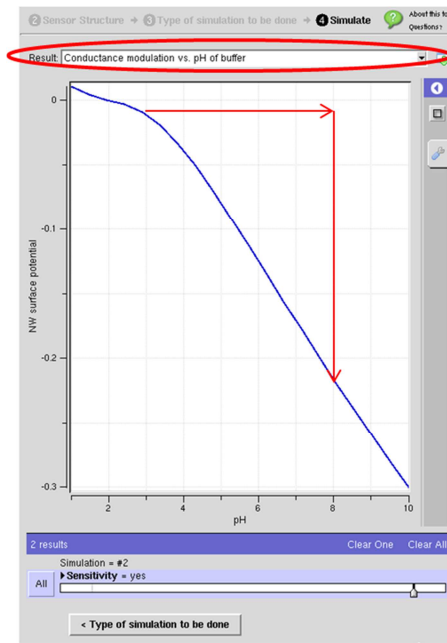
In the following problem set, we will explore the sensitivity of three different types of sensors, namely, pH-sensitivity of a NW sensor, Double-gate biosensor, and extended-gate biosensors.

Problem 3.7: pH response of a NW sensor

In this problem, we examine the sensitivity of the biosensor to change of pH in the fluid. This illustrative example is based on lectures on Sensitivity, specifically, lecture titled “ISFET as a pH-meter”.

Use Biosensor Lab (v2.0) to determine the following: If pH of the test fluid changes from 3 to 7, how does the surface potential change?

Solution:



- Select the graph “Conductance modulation vs. pH of buffer” for display.
- The plot below shows that the change in surface potential is 0.2V. Or equivalently, the sensitivity is ~50 mV/pH, close to the theoretical limit of 59 mV/pH.

