Bionanotechnology course, Exam 2016/17

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Part A: 45 points (1/3; recommendation spend no more than 5 minutes per questions; max one hour)
Part B: 46 + 23 + 9 + 12 + 9 = 99 points (2/3)
Part C / Bonus: 13 points

In all calculations, the end result should be given in significant numbers.

A) Literature and course questions (45 pts)

Answers should be ±50 words.

L1. You read the EU graphene roadmap and participated to the BNT course. Are the two compatible, complementary or have different visions? Argument and explain your thoughts in detail. What could be – in your view – a killer application for graphene (exclude propositions within the BNT course and proposals)? Explain why you think so. (5 pts)

L2. Drug delivery systems still face important drawbacks for efficient delivery of a drug to, for example tumors. What are those drawbacks and how are they solved? What are the important properties such a drug delivery system (i.e., for cancer therapy) need to fulfill? (5 pts)

L3. In the paper 'Tailoring the hydrophobicity of graphene nanopores' (Nat. Comm., 2013, 4, 2619), a self-assembled monolayer consisting of a pyrene and a short ethylene glycol species was proposed to prevent ssDNA to clog graphene nanopores. How was the thickness of the monolayer determined? Since 2017, wetting transparency of graphene has been investigated. Do you think that a coating is still necessary? And why? (5 pts)

L5. The review 'Single molecule detection with graphene' (Chem. Soc. Rev. 2016, 7, 45, 476-493), emphasizes on the technologies developed so far to detect single molecules with graphene nanopores and nanogaps. If one wish to sequence a biomolecule (take DNA as an example), what are the existing main limitations? Which design will you opt for (nanopore or nanogap)? Explain why? (5 pts)

L6. What is the working principle of a graphene field-effect sensor. How could it be used to detect, for instance, the binding of a drug to the active site of a protein? (5 pts)

L7. Light microscopy can be used to image nano-objects. Explain the methodology to do so. (5 pts)

L8. You visited the graphene and nanofabrication facilities. What do you think are the most essential instruments used to perform the research on nanopores and graphene field effect sensing. (5 pts)

L9. Is the random walk a phenomenon essential to consider at the nanoscale? Explain why. Cite a few (at least two) examples. (5 pts)

B) Problems

Problem #1: Working principle of a nanopore (46 points)

1a) Sketch the electrical and ionic circuit of a nanopore measurement setup. Include in the sketch the two redox electrodes, a battery (with + and – polarities), an ampere meter, the direction of the flow of current in the electrical circuit (with an arrow), the direction of the flow of electrons in the electrical circuit (with an arrow), determine the polarity of the electrodes and the direction of the flow of ions (consider KCl as the electrolyte). Identify the *cis* and *trans* chamber (the *cis* chamber is where the DNA will be injected, that is, at the negative electrode). (3 pts)

1b) If two Ag/AgCl redox electrodes are used, write the two half electrochemical reactions occurring at both electrodes. What is the degree of oxidation (i.e., the oxidation number) of silver in Ag and AgCl respectively? (2 pts)

1c) By applying an electrical potential across the two Ag/AgCl electrodes using the battery, an electrical current (I_{elec}) is measured using the ampere meter. What will be the measured ionic current (I_{ion})? (that is: express I_{ion} as a function of I_{elec}) (1 pt)

1d) Changing the two redox electrodes from Ag/AgCl to $KMnO_4/MnO_2$ will also change the mathematical relation between I_{ion} and I_{elec}. Determine this new relation. For simplicity, assume that KCl is replaced by $KMnO_4$ (only for this question). (3 pts)

In their 2011 Nanotechnology paper, Kowalczyk *et al.* approximated the resistance of a nanopore in a nanothick membrane by equation (1). They used Ag/AgCl as redox electrodes.

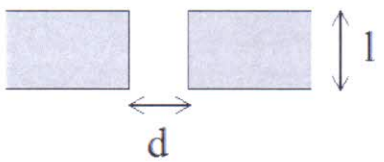
$$R = R_{channel} + 2R_{access} \tag{1}$$

Where $R_{channel}$ and R_{access} are expressed as

$$R_{channel} = \frac{1}{\sigma_e}\frac{4l}{\pi d^2} \tag{2}$$

$$R_{access} = \frac{1}{\sigma_e}\frac{1}{2d} \tag{3}$$

as derived from the model shown in Scheme 1. Here, σ_e is the bulk electrolyte conductivity, d the pore diameter and l the pore channel length.



Scheme 1.

1e) Explain the factor 2 in front of R_{access} in equation (1). (1 pt)

1f) Explain how $R_{channel}$ and R_{access} were determined (i.e., establish mathematically equation (2) and (3)). To answer this question, express the resistance of a cylinder as a function of the cyclinder dimensions (diameter d, and height l) and the conductivity of the electrolyte σ (i.e. for the access resistance). (3 pts for each, 6 pts total)

1g) Express the total conductance G of the SiN nanopore as a function of σ_e, l and d. (2 pts)

A nanopore of 20 nm diameter is created in a 30 nm thick SiN membrane.

1h) Describe at least one method to create nanopores in silicon nitride. (2 pts) Calculate separately the access resistance and the pore resistance of the SiN-pore in a 1M KCl electrolyte (σ_e = 10.5 Sm^{-1}) and express it in MOhms with three significant numbers. (5 pts)
According to equation 1, and using the Ohm's law (i.e. Voltage (U) = Resistance (R) * Current (I) what is the magnitude of the ionic current expected when a 200 mV bias voltage is applied over the pore? (express the current in nA with three significant numbers). (3 pts)

Double-stranded DNA is now added to the *cis* chamber and upon applying a 200mV bias voltage, individual DNA molecules translocate through the nanopore.

1i) With a background salt concentration of 1M KCl, the charges brought into the pore as DNA molecules translocate can be neglected, explain why. (2 pts)

1j) What will be the typical ion current traces observed upon DNA translocation? Assuming now that the pore diameter gets smaller and smaller (i.e. respectively d=6.5, 4.5, 2.5 and 0.5nm) what will the be the consequences on the shape of the observed translocation event (i.e. I vs time). To answer this question assume that a double-stranded DNA molecule has a diameter d_{DNA} of 2nm. (2 pts)

1k) Again, assuming that each DNA molecules have a diameter of 2nm, what will be the measured current blockade ΔI (in pA) when a DNA molecule translocate from head-to-tail in a non-folded conformation. (2 pts) What would be the current for a doubly and triply folded event? (2 pts) Why are folded events observed? (to proceed with the calculations assume that the ΔI is equivalent to the current $I_{substracted}$ that would be measured for a nanopore in SiN with the diameter of a DNA molecule. Explain why). (2 pts)

1l) Determine the numerical values of the conductance blockade (ΔG) in nS for non folded, doubly and triply folded translocation events. (Remember that for SiN nanopores, the ball park numbers for current blockades are in the order of 100pA per 100mV transmembrane potential). (3 pts)

1m) For an ultimately thin nanopore (l=0 nm), what will be the measured current blockade ΔI (respectively ΔG) for a non-folded event? (2 pts) Comment on why are nanopores in atomically thin membranes such as graphene or MoS2 regarded as potential DNA sequencer (2pts). What are the advantages and disadvantages of using graphene membranes for nanopore sequencing (2 pts).

Problem #2: Application of nanopores with carbon nanomembranes (23 points)

Carbon nanomembranes (CNMs) are carbon-based membranes that can be synthesized from self-assembled monolayers of small aromatic molecules, such as biphenyl thiols. The resulting membranes are graphene-like in the sense that they possess nanometer thickness.