Analytical Chemistry Cume Based on the Announced Publications:

Please be sure to completely answer each question that is asked!

1. (10) What is a quantum dot? In particular, what specifically about it is "quantum"?
Confined size effects in semiconductor nanocrystals produce distinct quantum states rather than a continuous band structure within the dot. Adjusting size affects the emission wavelength in a manner that can be modeled as a particle-in-a-spherical-shell.

2. (10) Why does a quantum dot "blink"? Please start with a practical definition of blinking, then move to a physical/chemical description of the processes that drive the effect.
Blinking arises when the dot becomes trapped in a non-emissive long-lived surface excited state. It can only fluoresce again once it relaxes back to the ground state.

3. (15) The authors use total internal reflection fluorescence microscopy (TIRFM) to perform these measurements.
-(5) What is total internal reflection?
-(10) Draw an instrument schematic for a TIRFM instrument, indicating in particular how light is introduced to the microscope, how it is collected, and how it is finally detected. Please be as specific and detailed as possible in your drawing.

4. (15) The authors take an interesting approach toward quantification of the signals, using the experimental standard deviation about the mean of the intensity to quantify brightness. What is the mathematical definition of the experimental standard deviation?

$$\sigma = \sqrt{\frac{1}{N-1}\sum_{i=1}^{N}(x_i - \bar{x})^2}$$

5. (15) Let's explore the detection limits assessment reported in the paper. Biotin-avidin binding is among the strongest non-covalent interactions found in nature. Let's assume that 15 minutes is sufficient time to allow every SA-QD to bind to the biotinylated interface down to the lower reported detection limit of 5 fM. How many SA-QD "molecules" will be present in the 40 µL sample volume? Assuming all of them bind uniformly across the ~40 mm^2 surface area of the well, how many molecules will be surface-bound within a100 µm × 100 µm field of view? Are your answers in reasonable agreement with the experimental results of the study (e.g., Figure 4)?
~30

6. (15) The abstract indicates a reported detection limit of 5 fM in the paper. What is the analyte in this assessment? How would this strategy have to be modified for detection of a generic protein isolated directly from a mouse urine sample (for example)? Would you still expect to observe detection limits of 5 fM in this case? Why or why not?
The "analyte" was a quantum dot decorated with streptavidin. Typically, one would be more interested in detecting a functional unlabeled biomolecule, which would require at least one additional steps in order to attach a reporter quantum dot. Furthermore, one would next need to be able to discriminate between quantum dots that contained bound targets versus unbound QDs.

The final detection limits of the measurement will depend more on the chemistry associated with the target binding to the dot and to the surface. These contributions can only result in increases in the detection limits, both from the presence of undetected analyte, and from a nonzero background from QDs not bound to the target analyte.

7. (5) Consider Figure 4 of the manuscript, reproduced herein. What is the difference between x in the $f(x)$ term, t in the time-axis, and Z in the Z-projection?
They are all identical.

8. (15) From inspection of the time-traces shown in Figures 2 and 3, can you think of an alternative data analysis strategy that would likely improve the confidence in the recorded digital counts to a significantly greater extent than the approach proposed in the paper? Explain and defend!
Many possible answers.

Biochemistry Cumulative Exam

Title: Nucleic Acids

August 24th, 2013

1. (10 points). Assume DNA replication proceeds at a rate of 750 bps per second; calculate how long will it take to replicate the entire genome?

The *E. coli* genome is 4.6×10^6 base pairs. Assuming DNA replication proceeds at a rate of 750 bps per second, the time required to replicate this amount of DNA assuming two replication forks is given by:

$4.6 \times 10^6 / 2 \times 750 = 3{,}093$ sec = 51.6 min

2. (10 points) Why does IPTG cause induction of the lac operon? How does induction by IPTG differ from induction by lactose?

IPTG binds lac repressor and turns on the lactose gene expression. Induction by lactose is transient, because lactose is converted to galactose and glucose by β-galactosidase. IPTG is a stable molecule and induction is long lasting.

3. (10 points) Describe the structure of a rho-independent transcriptional terminator.

Rho-independent terminators are DNA elements that when transcribe produce a short sequence of mRNA capable of forming a G:C rich stem and loop followed by a stretch of Us.

4. (20 points) There is one nucleosome per 200 bp of eukaryotic DNA. How many nucleosomes are in a diploid cell? Nucleosomes can be approximately as disks 11 nm in diameter and 6 nm long. If all the DNA molecule in a diploid human cell are in B-conformation, what is the sum of their lengths? If this DNA is now arranged on nucleosomes in the "beads on the string motif", what is the approximate total length?

Human diploid cells contain 46 chromosomes and 6×10^9 bp. The number of nucleosome is given by:

$$\frac{6 \times 10^9 \text{ bp}}{} = 3 \times 10^7 \text{ bp nucleosomes}$$

200 bp/nucleosome

The length of B DNA, 6×10^9 bp is given by

B DNA, 6×10^9 bp in size is:

6×10^9 bp x 0.34 nm/bp $= 2.04 \times 10^9$ nm = 2.4 meters in length.

3×10^7 bp nucleosomes x 6 nm/nucleosome = 18 x 107 nm = 0.18 m = 180 mm

5. (20 points) Describe an experimental protocol for the preparation of two cDNA libraries, one from anaerobically grown yeast cells and the second from aerobically grown yeast cells.

mRNA is isolated from total RNA of both aerobically and anaerobically grown yeast during oligo dT column. The mRNA is used as a template with reverse transcriptase and oligo dT to produce ssDNA, which is then converted to dsDNA and cloned into a vector.

6. (30 points) (a) Why do *E coli* cells synthesize sigma subunits for transcription? What advantage do they confer in transcription process?

Because sigma subunit confers specificity to RNA polymerase to bind to certain promoter sequences, thereby controlling the transcription of the desired genes.

(b) Which RNA polymerase is responsible for the synthesis of rRNA in eukaryotes?

RNA Polymerase I and III.

(c) Why is uracil not utilized by DNA as a base?

Cytosine upon deamination can form uracil. DNA recognizes uracil as the non-natural base and replace it with cytosine. This prevents mutations in DNA sequences.

Inorganic Cumulative Exam
Bonding and Reactions in Inorganic and Organometallic Chemistry
August 24, 2013

1. (10 points) Draw the five d-orbitals and label them.

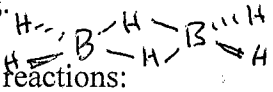
5-orbitals
two nodes

2. (15 points) CrF_2 adopts a distorted rutile structure in which two different Cr-F bond lengths are observed, 200 pm and 243 pm. What is the coordination number of Cr in CrF_2? Explain the presence of two different bond lengths. Jahn-Teller distortion

6-coord., d^4 Cr center, unequal occupancy of orbitals

3. (20 points) Construct a molecular orbital (MO) diagram for molecular oxygen. According to the Pauli exclusion-principle, show the electron occupancy in your MO diagram. What is the spin for ground state oxygen? See a general chemistry textbook. Two unpaired electrons (triplet).

4. (10 points) Discuss briefly the bonding in B_2H_6.



3-center-2-electron bonds

5. (30 points) Give the products for the following reactions:

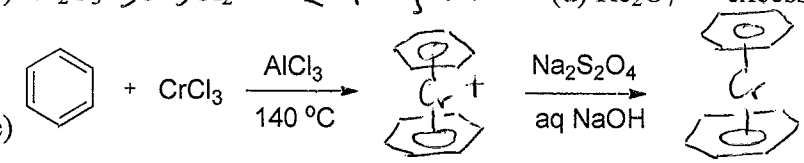
(a) $Xe + PtF_6 \rightarrow$ $Xe^+[PtF_6^-]$

(b) $Pt(py)Cl_3^- + py \rightarrow$ trans-$Pt(py)_2Cl_2$

(c) $Cr_2O_3 + 3C + 3Cl_2 \rightarrow$ $2CrCl_3 + 3CO$

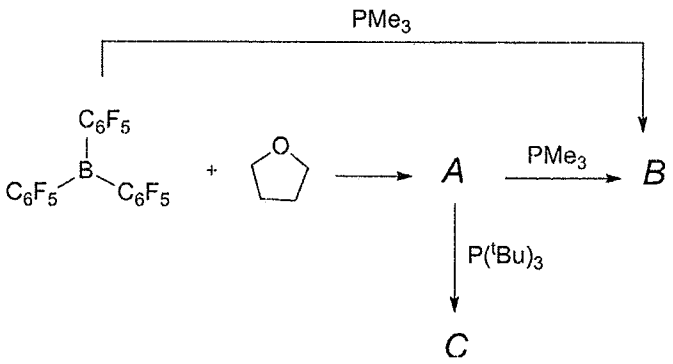
(d) Re_2O_7 + excess CO → $Re_2(CO)_{10}$

(e)



(f) $CO + H_2O \xrightarrow{\text{Cat./ high temp.}}$ $CO_2 + H_2$

6. (15 points) Identify A, B, and C in the scheme below. Draw structures please.



A: $(C_6F_5)_3B-O$

B: $(C_6F_5)_3B-PMe_3$

C: $(C_6F_5)_3\bar{B}-O-\overset{+}{P}(^tBu)_3$

frustrated Lewis Acid Base pair

Organic exam is crib

No Physical crib available
April 24, 2013
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