## **Homework Questions:**

1. (20%) You are performing a light scattering experiment with a 488 nm laser. What is the smallest particle size (in nanometer units) that will allow you to detect an angular dependence (and therefore some shape information)?

2. (30%) A macromolecule is moved from a non-polar solvent to a polar solvent, which causes a change in energy states as shown below. When moving to the polar solvent, do you expect a red or blue shift in the absorption profile, or none at all for (a) the n to  $\pi^*$  transition, and (b) the  $\pi$  to  $\pi^*$  transition? Explain why. Arrows on the right are drawn to scale for comparison.



3. (50%) A researcher carefully weighs out 10 mg of dry, lyophilized pure protein powder and dissolves it in a buffer, and prepares and measures the following dilution series on a spectrophotometer, which is blanked with distilled water:

mg/ml Protein Concentration	Absorbance Units (OD 280 nm)
0.1	0.412
0.2	0.490
0.3	0.638
0.4	0.690
0.5	0.814
0.6	0.932
0.7	1.046
0.8	1.089
0.9	1.164
1.0	1.236
1.2	1.298
1.3	1.337
1.4	1.369

- a. (5 %) At what OD level at 280 nm does this spectrophotometer become nonlinear? Explain with a graph.
- b. (5 %) What advantage or disadvantage does the researcher have by blanking the spectrophotometer with distilled water rather than with the buffer?
- c. (20 %) Is there any baseline absorbance present from the buffer? If so, how much?
- d. (20 %) What is the molar extinction coefficient at 280 nm for this protein, given this was measured in a 1 cm cuvette, and the molar mass of the protein is 45,000 Dalton? Make sure not to include

any non-linear data for this determination. Express your answer in  $\frac{ODL}{mol \ cm}$  units.