Exam 2, Chapters 13, 14, 15, 16, and 17

1. (10 points) One of the useful rules of thumb that you may have learned in Biochemistry, is that a 50 μg/ml solution of DNA has an absorbance of about 1.0 in a 1 cm cell.

A. I have a solution of DNA that has an transmittance of 0.15 when it is placed in a cell with a 200μm pathlength. What is the concentration of DNA in this solution (in units of μg/ml)?

B. Assume that this DNA is a synthetic oligo containing 25 base pairs, and that the MW of a single base pair has a molecular weight of 330. What is the molar concentration of duplex DNA molecules in the solution?

C. What is the Molar absorptivity of DNA in units of L cm^{-1} $(mol\ base\ pair)^{-1}$

2. (20 points) Compare and contrast a photodiode array spectrometer with a double beam dispersion type spectrophotometer. Start with a general description of how each instrument is built and how it works, then compare the instruments in terms of noise, resolution, reproducibility, sensitivity, accuracy, ease of use, speed for a scan, number of moving parts, ease of use, and cost, and any other factor you can think of.

3. (10 points) Compare $\sigma \rightarrow \sigma^*$, $n \rightarrow \sigma^*$, $n \rightarrow \pi^*$, and $\pi \rightarrow \pi^*$ transitions

Rank in order of highest E to lowest E, Where you would expect to find them in the UV/Vis region (short, medium or long wavelengths), Which transitions have strong absorptions, which have weak absorptions, which transitions undergo solvent shifts, and are these solvent shifts blue shifts or red shifts. Finally which of these transitions is usually involved in fluorescence or phosphorescence.

4. (20 points) Compare UV absorbance with UV fluorescence

How are machines measuring these two effects similar or different in construction?

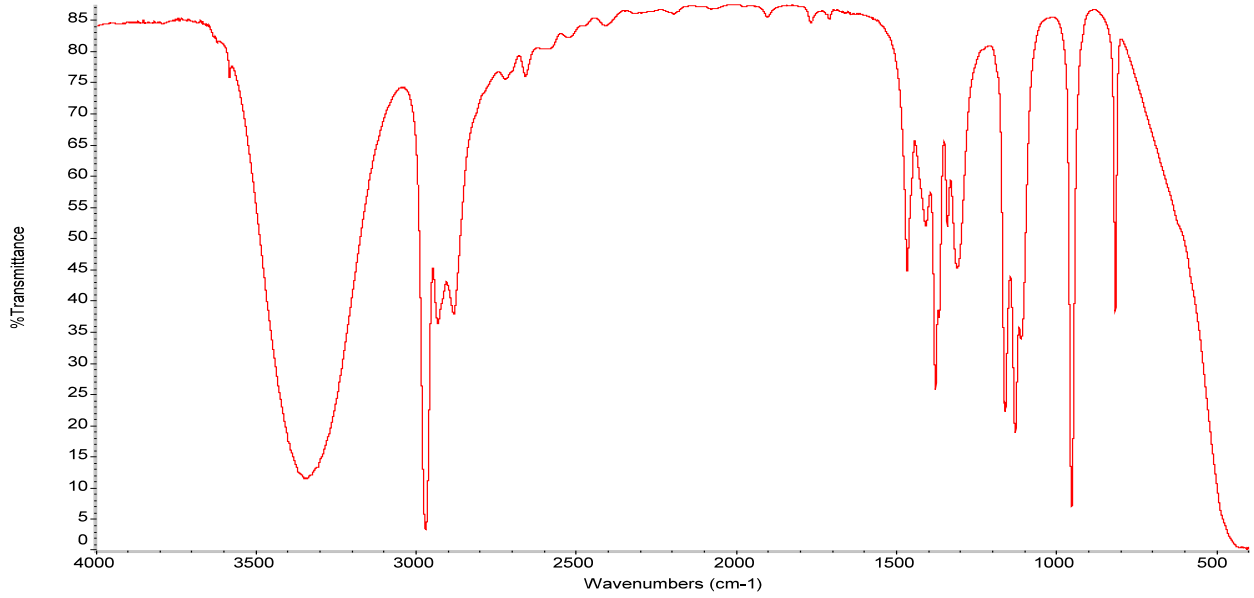
Which technique is more sensitive and why?

Which technique is more generally useful and why? (Can be applied to the most compounds)

5. (15 points) Make a block diagram of an FT-IR and describe how it works. Since this device does not have a monochrometer make sure you detail what your actual signal looks like, and how the movement or position of a mirror encodes frequency information.

6. (15 points) On the next 2 pages are the IR spectra and molecular formulas of four different compounds. Identify as many group frequencies as you can in these compounds, and, if possible, identify the compound. (You can use the book of reference spectra if you like)

C_3H_8O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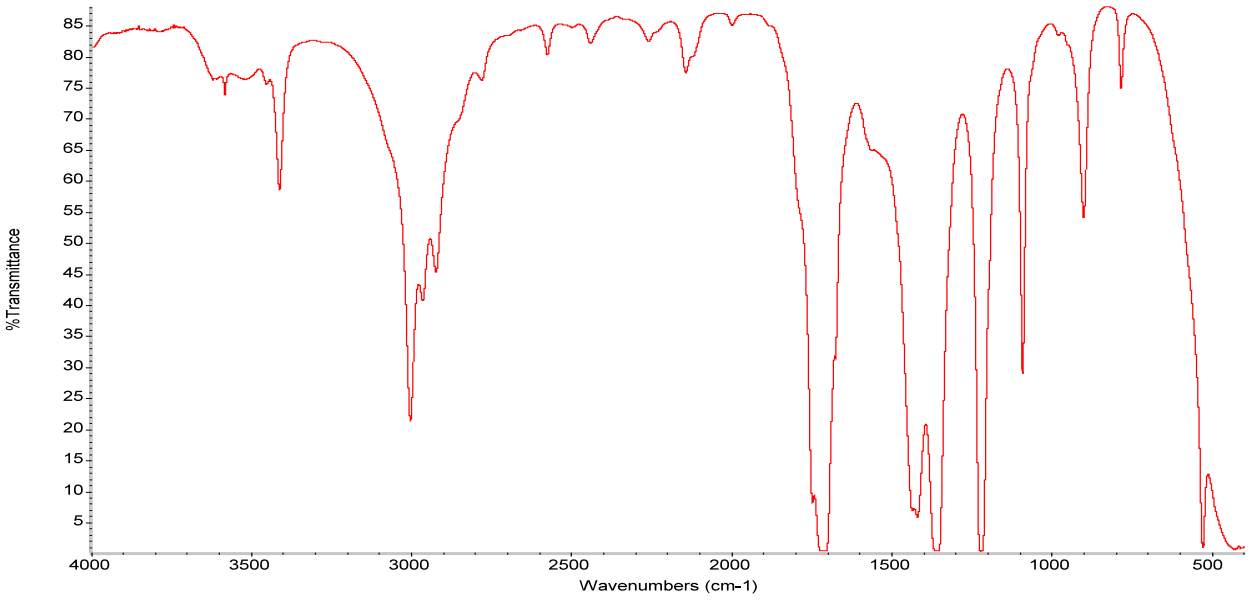
Wavenumbers (cm-1)

C_3H_6O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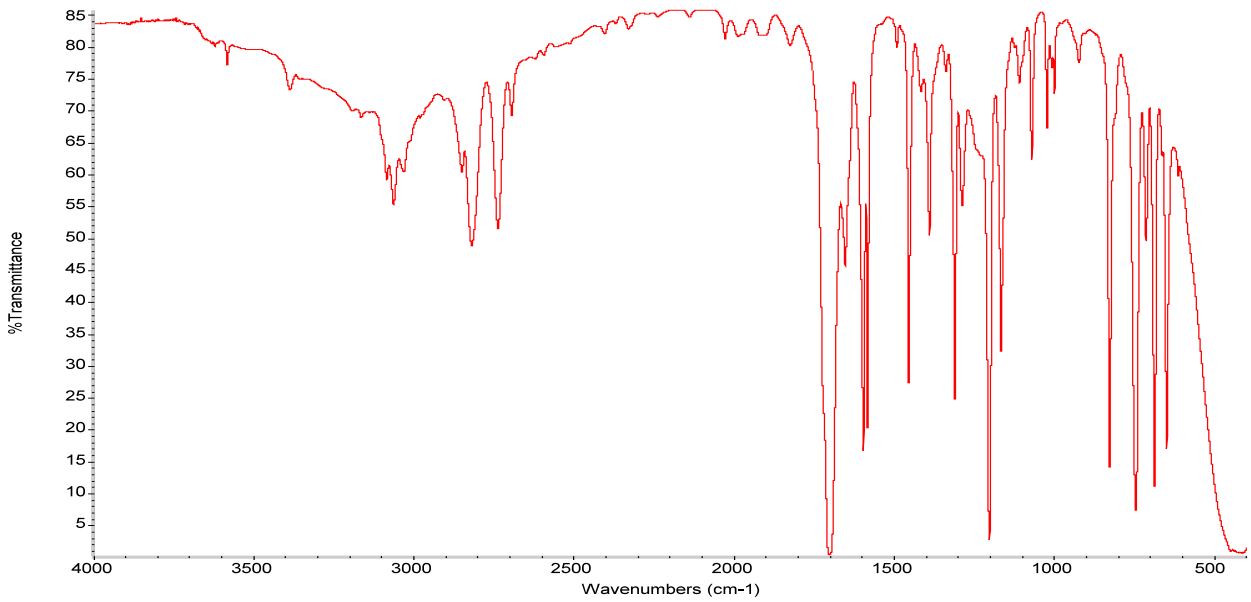
Wavenumbers (cm-1)

C_7H_6O



$C_4H_{10}O$