BC367, Fall 2018 Problem set #4 Due: Friday, September 28th

(Don't forget that each question should be answered on a separate sheet of paper. Also, please *type* your narrative answers.)

1. In addition to the more typically used modes of liquid chromatography (ion exchange, size exclusion, affinity, etc.) some scientists have found that a chromatographic stationary phase that includes the mineral hydroxyapatite (HA) can be useful as separating some proteins. Do some brief research on HA chromatography and describe what it is and how it could be used. (Your answer should be cited.)

2. In performing an MS/MS experiment on an unknown protein, your peptide fingerprint analysis software died before you can get the sequence of your last peptide fragment. Luckily, you have the CID spectrum. Using the following m/z values from this spectrum to identify the peptide sequence (assume perfect CID cleavage, *i.e*. each peak is a clean 'b' or 'y' fragment).

98.06009	334.14316
106.04991	455.19593
227.10268	463.18576
237.0904	542.22796
324.15544	560.23852

3. If you were using MS/MS to determine the identity of a short peptide and the mass of one of the residues corresponded to leucine, which of course could just as likely be isoleucine. How could you differentiate Leu and Ile experimentally?

4. You've purified a recombinant globular protein to 99% homogeneity. Because this protein is an enzyme, you can easily measure it's functional purity and determine that it has less than half the activity that it should have. You know that the enzyme has three cysteine residues, two of which are supposed to be in a disulfide bond. Discuss experiments that you could do to determine if, A) a disulfide bond is there, B) if the *correct* disulfide bond is there, and C) measures you could take to form or re-form the bond properly.