Consider a hexapeptide with the following composition of amino acids: The N-terminal residue has the shortest side chain (R group) containing 2 methyl groups. The next residue's side chain contains an aromatic ring that can be phosphorylated. The next residue has a nonpolar aliphatic side chain containing only methylene groups. The next residue's side chain is readily oxidized to form covalent bonds. The next residue has the shortest side chain that is negatively charged at neutral pH. Lastly, the C-terminal residue has the longest side chain with an amido group.

Answer the following questions. Use the pKa values for R groups from Table 3-1 in Lehninger or from the lecture slides. You can assume the "common" values for the terminal carboxyl and amino groups: $pK_1=2.3$ and $pK_2=9.7$, respectively.

(A) Write the amino-acid sequence of this peptide in 3-letter and 1-letter abbreviations.

(B) What is the total charge on the peptide at pH = 12?

(C) Estimate the isoelectric point of this peptide. Show the equation you used and briefly explain your reasoning.

(D) What fraction of the peptide molecules is in the zwitterionic state at the isoelectric point? Show your calculation and briefly explain your reasoning.

(E) You want to use ion-exchange chromatography to purify this peptide by retaining it on the column. Do you need cation or anion column for this if the peptide is in TRIS buffer at pH=8.2? Explain your reasoning.