CHEM 322 **Name**_____
Spring 2015 **Due date TBA**

Complete the following on separate paper. Show your work and **clearly identify your answers**.

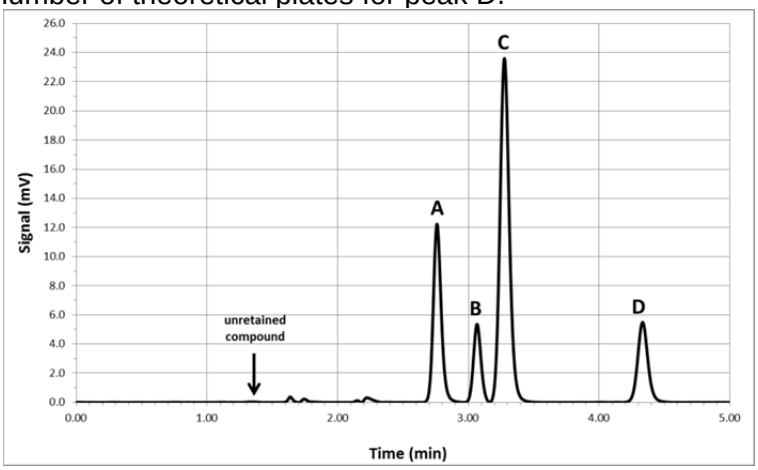
General Separations

1. Describe the relative contributions of the A, B, and C terms in the van Deemter relationship for the following four techniques: (a) packed column GC, (b) capillary GC, (c) HPLC, (d) capillary electrophoresis. Sketch a qualitative van Deemter curve (like Figure 26-10) for each technique.

2. How many theoretical plates must a column have to achieve a resolution of 1.5 between two peaks eluting at 8.0 and 8.2 min?

3. Why is dead volume more of a concern in gas chromatography than in liquid chromatography?

4. Consider the chromatogram below. For your convenience, the raw data for the chromatogram is available on our CHEM 332 website (under the "Homework" link) as an Excel file.
   a. Calculate the capacity factor (k') for each peak.
   b. Calculate the resolution for each pair of adjacent peaks (A-B, B-C, C-D).
   c. Calculate the number of theoretical plates for peak D.



Gas Chromatography

1. What are the key benefits for using open tubular (capillary) columns as opposed to packed columns for GC? Why might you choose packed columns anyway?

2. What is the effect of stationary phase film thickness on gas chromatograms?

3. Consider the GC separation of water, n-propanol, 1,1-dichloropropane and n-propane on a carbowax column. The separation is run three times, once using a TCD, once with and FID, and once with an ECD. Sketch the expected chromatograms for each separation and describe the key differences in each chromatogram.

4. What is the purpose a retention index (also known as a Kovats index)? For the data below calculate the retention index for 2-hexene.

Sample	Retention Time (minutes)
air	0.642
n-pentane	2.88
n-hexane	4.51
2 hexene	3.77

Liquid Chromatography

1. What is the purpose of a guard column in LC? How does it differ from a suppressor column?

2. Describe how an injection loop allows for improved precision for sample introduction. How does the autosampler on our HP1050 instrument function in a similar way?

3. Consider the table below. Both fluorescence and electrochemical detection offer lower detection limits and wider linear ranges than absorbance detection, yet absorbance is still the most common LC detection mode. Suggest reasons why this might be the case.

TABLE 28-1 Performance of HPLC Detectors

HPLC Detector	Commercially Available	Mass LOD* (typical)	Linear Range† (decades)
Absorbance	Yes	10 pg	3–4
Fluorescence	Yes	10 fg	5
Electrochemical	Yes	100 pg	4–5
Refractive index	Yes	1 ng	3
Conductivity	Yes	100 pg–1 ng	5
Mass spectrometry	Yes	<1 pg	5
FTIR	Yes	1 μg	3
Light scattering	Yes	1 μg	5
Optical activity	No	1 ng	4
Element selective	No	1 ng	4–5
Photoionization	No	<1 pg	4

4. Compare the benefits and limitations of using displacement pumps as compared to reciprocating pumps for HPLC. Why are reciprocating pumps more popular?

Capillary Electrophoresis

1. Why does pH affect the separation of amino acids by electrophoresis?

2. Explain how micellar electrokinetic chromatography combines electrophoretic and chromatographic separations principles. Why is the MEKC micelle called a “pseudostationary phase”?

3. Describe how electroosmotic flow might be intentionally “turned off” during an electrophoretic separation. Why might someone want to do this?

4. Capillary electrophoretic separations have the potential (no pun intended) to give very efficient separations, when compared to GC and LC. Why is this the case?