1. For a reversed-phase chromatography experiment, it is noted that the retention time of an analyte decreases as the percent of acetonitrile (CH$_3$CN) increases in a CH$_3$CN/H$_2$O mobile phase. Explain why this happens.

(5)

As the percent CH$_3$CN increases, the mobile phase becomes less polar and more competitive with the nonpolar stationary phase for the analytes. This causes the analytes to elute more rapidly.

In chromatography, what is the significance of the theoretical plate height ($H$)? How is $H$ calculated from experimentally measured parameters? Be sure to define all terms.

(6)

Plate height is an indicator of column efficiency. The smaller the plate height, the more efficient the column.

$$H = \frac{L}{N}$$

where

$N = 16 \left( \frac{t_R}{w} \right)^2$

$L$ is the column length

$N$ is the number of plates

$t_R$ is the component retention time

$w$ is the baseline width of the peak

Indicate the major advantage that liquid chromatography has over gas chromatography in terms of analyzing a complex mixture.

(3)

Analytes do not have to be volatile or easily vaporized in order to use liquid chromatography.

Briefly explain why substances such as ethanol and hydrocarbons generate a signal in a flame ionization detector, but water, ammonia and carbon dioxide do not.

(4)

Column eluate flows into a small hydrogen flame. As the eluted material burns in the flame, any compounds like ethanol and hydrocarbons that contain oxidizable carbon atoms produce cations that are collected by an electrode positioned near the flame. This flow of cations constitutes the detector signal. Since water, ammonia and carbon dioxide do not contain oxidizable carbon atoms, these substances do not produce a signal.

What is a junction potential? Why do junction potentials develop?

(6)

A junction potential is a small voltage difference that develops at the interface when two dissimilar electrolyte solutions are in contact. It results from differing diffusion rates for the cations and anions of the electrolyte solutions.
1. Give an example of an ion-selective electrode. What is the fundamental difference between an ion-selective electrode and metal electrodes?

(6)

glass membrane electrode (specific for H\(^+\))

Ion selective electrodes differ from metal electrodes in that a potential develops across a thin membrane due to selective migration of ions across this membrane. No redox reaction occurs at the electrode surface.

What is a reference electrode?

(3)

A reference electrode is a half cell that has a constant, and usually known, half-cell potential.

2. Briefly explain why complexes in aqueous solution, such as Fe(phen)_3^{2+}(aq), have very broad absorption bands.

(5)

Vibrational excitation is superimposed on the electronic excitation occurring as a result of light absorption. In solution, these discrete transitions blend into a broad continuous absorption band because energy levels in such complexes become distorted due to interactions with other solutes and solvent.

What is Beer’s law? Define all terms. Indicate one limitation to Beer’s law.

(6)

Beer’s law: \[ A = \varepsilon l c \]

where

- \( A \) is absorbance;
- \( l \) is the pathlength the radiation takes through the sample (units of cm);
- \( c \) is the concentration of the light-absorbing substance (units of mol/L);
- \( \varepsilon \) is the molar absorptivity (units of L/mol-cm)

Limitations of Beer’s law:

- Beer’s law is successful in describing absorption behavior of dilute solutions only.

Beer’s law applies only to monochromatic radiation.

In terms of radiation output, what is the major difference between a hollow cathode lamp and a tungsten lamp? Why don’t we use a tungsten lamp for atomic absorption spectrophotometry?

(6)

The tungsten lamp emits all wavelengths in the visible (and near IR) whereas the hollow cathode lamp emits only the discrete wavelengths associated with electronic excitation/de-excitation in the cathode material.

The tungsten lamp, combined with a conventional monochromator, cannot provide bands of radiation that are narrow compared to the atomic absorption bands of the analytes. Thus, Beer’s law will not hold.
3. One can carry out an analysis for iodide potentiometrically by monitoring the potential of the following cell during the titration of an unknown iodide solution with Ag\(^{+}\)(aq).

\[
\text{Reference Electrode}
\]

\[
\text{Ag(s)} \mid \text{AgI(s)} \mid \text{analyte soln} \parallel \text{Cl}^{-}(\text{aq}) \mid \text{Hg(l)} \mid \text{Hg}_2\text{Cl}_2(\text{s}) \mid \text{Pt(s)}
\]

a) Write a balanced equation for the half reaction occurring in the reference electrode.

\[
\text{Hg}_2\text{Cl}_2(\text{s}) + 2e^- \leftrightarrow 2\text{Hg(l)} + 2\text{Cl}^-(\text{aq})
\]

b) Using activities, if the reference electrode has a potential of 0.253 V, what would be the potential of the above cell at the equivalence point in the titration of an iodide unknown with Ag\(^{+}\)(aq)? Assume that the activities of I\(^-\)(aq) and Ag\(^{+}\)(aq) are equal at the equivalence point.

\[
E_{\text{cell}} = E_{\text{ox}} + E_{\text{red}} = E_{\text{ox}} + 0.253 \text{ V}
\]

at the equivalence point

if \(a_{I^-} = a_{Ag^+}\), then \(K_{sp} = a_{I^-} \cdot a_{Ag^+} = a_{I^-}^2\) and \(a_{I^-} = (K_{sp})^{1/2} = (8.3 \times 10^{-17})^{1/2} = 9.11 \times 10^{-9}\)

anode reaction: \(\text{Ag(s)} + I^-(\text{aq}) \rightarrow \text{AgI(s)} + e^-\)

\[
E_{\text{ox}} = E_{\text{ox}}^0 - \frac{0.05916}{1} \log \frac{1}{a_{I^-}} = 0.152 \text{ V} - 0.05916 \log \frac{1}{9.11 \times 10^{-9}} = 0.152 \text{ V} - 0.478 \text{ V} = -0.324 \text{ V}
\]

\[
E_{\text{cell}} = -0.324 \text{ V} + 0.253 \text{ V} = -0.071 \text{ V}
\]

**DATA**

<table>
<thead>
<tr>
<th>Electrode</th>
<th>(E^0(\text{V}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ag(^{+})/Ag</td>
<td>0.799</td>
</tr>
<tr>
<td>AgI/Ag</td>
<td>-0.152</td>
</tr>
</tbody>
</table>

\(\text{AgI} \quad K_{sp} = 8.3 \times 10^{-17}\)
4. Ammonia can be determined spectrophotometrically by reaction with phenol in the presence of hypochlorite (OCl\(^-\)).

![Chemical Reaction Diagram]

A 4.37-mg sample of protein was chemically digested to convert its nitrogen to ammonia and then was diluted to 100.0 mL. Then 10.0 mL of this solution were placed in a 50-mL volumetric flask and treated with 5 mL of phenol solution plus 2 mL of sodium hypochlorite solution. This sample was diluted to 50.0 mL and the absorbance was measured in a 1.00-cm cuvet after 30 minutes. For reference, a standard solution was prepared from 0.0100 g of NH\(_4\)Cl (FW=53.49) dissolved in deionized water and diluted to 1.00 L. Then 10.0 mL of this standard solution were placed in a 50-mL volumetric flask and analyzed in the same manner as the unknown. A reagent blank was prepared using deionized water in place of the unknown.

The following data were collected.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Absorbance at 625 nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>blank</td>
<td>0.140</td>
</tr>
<tr>
<td>standard</td>
<td>0.308</td>
</tr>
<tr>
<td>unknown</td>
<td>0.592</td>
</tr>
</tbody>
</table>

a) Calculate the molar absorptivity of the blue product.

In the standard, [blue product] = [NH\(_4\)^+]

\[
[NH_4^+] = \frac{0.0100 \text{ g} \times \frac{1 \text{ mol}}{53.49 \text{ g}}}{L} \times \frac{0.0100 L}{0.050 L} = 3.73 \times 10^{-5} M
\]

\[
\varepsilon = 4.49 \times 10^3 \text{ M}^{-1} \text{ cm}^{-1}
\]

\[
A_{std} = 0.308 - 0.140 = 0.168 = 3.739 \times 10^{-5} \text{ M}(1.00 \text{ cm}) \varepsilon \quad \Rightarrow \quad \varepsilon = 4.49 \times 10^3 \text{ M}^{-1} \text{ cm}^{-1}
\]

b) Calculate the mass percent of nitrogen (AW=14.0) in the protein.

\[
\frac{A_{unk}}{A_{std}} = \frac{0.592 - 0.140}{0.308 - 0.140} = \frac{[NH_3]_{unk}}{[NH_3]_{std}} = \frac{[NH_3]_{unk}}{0.0100 \text{ g} \times \frac{1 \text{ mol}}{53.49 \text{ g}} L} = \frac{[NH_3]_{unk}}{1.87 \times 10^{-4} M}
\]

\[
\text{Mass percent of nitrogen} = \frac{[NH_3]_{unk}}{1.87 \times 10^{-4} M} \times \text{FW of NH}_3 \times 100\%
\]
\[ [\text{NH}_3]_{\text{unk}} = 5.03 \times 10^{-4} \text{ M} \]

\[
100.0 \text{ mL} \times \frac{5.03 \times 10^{-4} \text{ mol } N}{1000 \text{ mL}} \times \frac{14.0 \text{ g } N}{1 \text{ mol } N} = 7.04 \times 10^{-4} \text{ g } N
\]

\[
\frac{7.04 \times 10^{-4} \text{ g}}{4.37 \times 10^{-3} \text{ g}} \times 100 = 16.12\%
\]

16.1%