GAS CHROMATOGRAPHY

Gas chromatography provides a means for the separation of volatile components of either a gaseous, liquid, or solid mixture based on a partitioning of the mixture's vapor components between two phases. One phase (the stationary phase) is a stationary bed of small particles through which the vapor component travels. The volatile components are generally interspersed in an inert gas such as helium (the mobile phase) as they travel through the stationary phase. In many cases, the stationary phase is a solid material. When it is such the technique is termed gas-solid chromatography (gsc). More often, the stationary phase is a liquid adsorbed on to the surface of a solid support. When liquids comprise the stationary phase the technique is called gas-liquid chromatography.

The objective of this lab is to explore gas chromatography, including the concepts of retention, height equivalent to a theoretical plate (HETP), retention parameters of polar columns, and flow rate using a simple mixture. In addition, the separation of volatile components in a more complex mixture will also be investigated. In this section, the effects of temperature programming of the GC column will be investigated.

Materials:

In this experiment you will use a Hewlett-Packard HP 5890A Gas Chromatograph. This instrument is programmable to allow a wide variety of operating conditions of use. The column used in this GC will consist of a solid stationary phase called Carbowax 20 M, a common polar support. The mobile phase will be helium. Detection of the eluted mixture components will be accomplished using a thermal conductivity detector (TCD) or flame ionization detector (FID). Chromatograms will be collected and stored digitally using *Logger Pro*, a commercial data acquisition package.

I. Set-up:

a. The He carrier gas should be on prior to heating the column. Allow the He gas to flow through the column for approximately 15 min before turning on the oven.
b. Adjust the flow rate of the carrier gas to approximately 40 mL/min.
c. After the 15 minute column conditioning period, set the oven temperature to 110°C. The injector temperature should be set at least 10° higher than the column temperature.
d. Verify that the voltage output from the detector is connected to the Serial Box Interface using the Instrumentation Amplifier. These components allow conversion of the analog signal at the detector to a digital signal, capable of being stored on the computer. For basic instruction on using *Logger Pro*, consult "**Data Collection for GC using Logger Pro**".

II. Injection Reproducibility

Inject three successive 0.5 μL n-propanol samples, including approximately 5 μL of air into the column at one minute intervals. You may need to adjust the attenuation and/or the range factors on the GC to get the peaks on scale. Measure the peak height and peak areas and calculate the average values, the standard deviation, and the relative standard deviation for your injections.

III. Effect of Data Acquisition Rate:

To better understand the implications of A/D data systems to accuracy and precision, perform the following before running your sample mixture. Select one analyte and run two 0.5 μL injections each at data collection rates of 0.25, 1, 2, 5, and 25 samples per second. What affect does changing acquisition rate have on the quality of your data? How will this affect the accuracy and precision of a result? What are the benefits and challenges of collecting at low rates compared to high rates? In your notebook, include a plot of one injection at each collection rate.

IV. HETP versus Sample Volume:

Obtain the chromatograms of 0.1, 0.2, 0.4, 0.6, and 0.8 μL of n-propanol. If the attenuation is changed during the injection of the sample, you must inject the same sample before and after the change in attenuation to allow for correlation of the samples. Calculate the HETP for each sample and plot HETP versus sample volume.

$$HETP = L/N$$

L = length of the column

N = number of theoretical plates

The number of theoretical plates is calculated using the following relationship.

$$N = 16(x/y)^2$$

The value x corresponds to the distance of the peak maxima from the point of injection; y represents the width of the transient chromatographic peak at the base.

V. Peak Area versus Sample Volume:

Measure the peak area for each of the peaks in the above samples. Use the height and width at half-height to approximate the area. Plot the area/volume ratio versus the sample volume. What relationship exists between the sample volume and the area/volume ratio? What relationship would you expect from the sample concentration and the peak areas?

VI. Retention Parameters - Qualitative Analysis:

Inject a 1 μL sample of a mixture of 1-heptane, tetrahydrofuran (THF), 2-butanone (MEK), and 1-propanol into the column (predict the order of elution of these components before injecting your sample.) Include air in your injection for an additional point of reference. Using pure samples and the retention time from each of the eluent peaks, identify the peak corresponding to each of the components of your unknown mixtures. Was your prediction of the order of elution correct?

VII. Effect of Carrier Gas Flow Rate:

Increase the rate of the carrier gas flow rate to approximately 45-50 mL per minute. Inject a sample of the four component mixture into the column and note the differences in elution of these components with an increased flow rate. What effects were noted?

VIII. Temperature Effects:

Adjust the flow rate of the system to 30 mL per minute, the column temperature to 90° C, the detector block temperature to 150° C, and the injector temperature to 150° C. Allow a few minutes for the system to equilibrate. Inject a 4 μL sample of mixture into the chromatograph. Record the chromatogram for approximately 15 minutes.

After the constant temperature chromatogram has been recorded adjust the parameters of the chromatograph to the following conditions.

initial column temperature:	45°C
time at initial column temperature	2 min.
rate of temperature increase	16°C per min.
final column temperature	120°C
time at final column temperature	2 min

Inject 4 μL of the mixture into the chromatograph and immediately start the program. Record the chromatogram and compare it to the constant temperature chromatogram. At the higher column temperature the components are eluted much more rapidly and the resolution decreases. If the resolution is not adequate for your needs, comment on alterations in the program which could be used to improve the resolution of the components of the mixture.

Attempt a modification to the temperature programming in order to improve the resolution of your mixture's components. Inject another sample and see if your modifications actually did improve the resolution of the components of interest. Comment on the method that you might use to identify the various components of the mixture (qualitatively).