Chapter 16:

Gas Chromatography

Problem 16.1: In the development of a GC method, list three experimental variables under the control of the analyst.

Temperature and flow rate are easily controlled during the experiment. The selection of carrier gas can have an impact on chromatography, and this is a relatively simple modification to accomplish, although not without some planning and preparation time. Finally, the analyst can select from a wide variety of column types and stationary phases, but changing this parameter requires time and financial resources.

Problem 16.2:

(a) Use your knowledge of partition kinetics to speculate on how an analyte might behave if the temperature in a GC analysis was set too low.
(b) How might you exploit a "too low" temperature to your advantage in a GC analysis?

(a) a low temperature (assuming it is still high enough to vaporize the sample) would slow down the kinetics of partitioning between the two phases. This would have the effect of increasing the C term in the van Deemter equation, causing band broadening.

(b) we could use a "low" temperature to preferentially vaporize some components of the mixture. Those components with high boiling points would condense at the beginning of the column while those with low boiling points would move on. Then we could increase the temperature to mobilize the condensed components.

Problem 16.3: Using Examples 16.1, 16.2, and 16.3 as guides, propose a method for the separation of caffeine, catechin, and phenol. Justify your choice of columns and your temperature gradient.

*** Note – this should be catechol rather than catechin
There are many possible answers.

Boiling points: caffeine = 178°C catechol = 245°C phenol = 182°C

Caffeine and phenol are similar in boiling point, but phenol is more polar. Catechol has a high boiling point but is similar in polarity to phenol. We need to be able to achieve a temperature of about 270° to be certain to remove all of the catechol. We would want to use a column of moderate polarity in order to have all solutes interact with it. For instance, we could select the SPB-20 (See Example 16.2) column and use a start temperature of around 150°, increase the oven temperature slowly (perhaps 5 °C/min) until we reach a temperature of about 200°C, then increase more rapidly (20 °C/min) to more quickly pull off the catechol.

Problem 16.4: Go online and use one of the GC column supplier's "How to Select a Column" guides and select a column for the analysis of gasoline. (See Table 16.2 for a list of GC column suppliers.) Explain why you selected that particular column, and justify your selection based on chromatographic principles.

Browsed to www.restek.com
Selected Columns > GC
Used "Narrow your results:" and selected "Petroleum and Petrochemical"
Used "Narrow your results:" and selected "Gasoline"
One option remained: Rt-TCEP column
highly polar phase; 1,2,3-tris[2-cyanoethoxy]propane—not bonded

The n-chain hydrocarbons in various gasoline samples will be fairly similar among different brands. The modifications that are made by manufacturers to impart specific properties will be substituted aromatics and oxygenated species. This polar column will allow the separation and analysis of those polar species.

Problem 16.5: Go online and use one of the GC column supplier's "How to Select a Column" guides and select a column for the analysis of metabolic steroids. Explain why you selected that particular column, and justify your selection based on chromatographic principles

Answer will vary depending upon which supplier is chosen.

Problem 16.6: As noted above, nitrogen gas exhibits a fairly low range of useful flow rates as a carrier. Hydrogen exhibits the widest range of useful flow rates among common carrier gases. Postulate on the relative advantages and disadvantages of using H_2 versus N_2 to explain why N_2 is more commonly used than H_2.

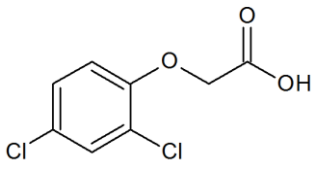
There are three main reasons. First, N_2 is significantly less expensive than H_2, so for continuous use in a laboratory, it represents a big cost savings. Secondly, N_2 is inert, while H_2 is reactive toward many species; using N_2 reduces the potential for reactions with analytes. Finally, the utilizing H_2 in a continuous use laboratory requires excellent ventilation ($$) in order to prevent the potential for build-up of the explosive gas.

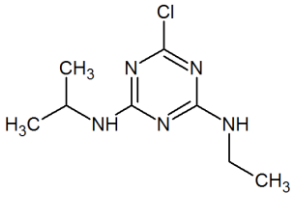
Problem 16.7: Do you think using CO_2 as a carrier gas in an instrument with an FID would be problematic? Explain.

No, it should not be a problem. The FID detector does not respond to CO_2 because CO_2 has carbon in its most oxidized form. That is, CO_2 is not combustible, therefore could not be seen with an FID detector. CO_2 is also chemically inert, so it is unlikely to react with analyte species, even at elevated temperatures.

Problem 16.8: In your job as a chemist for a regional analytical laboratory, you are expecting a large number of samples that you will need to test for potential herbicide contamination. Prepare

a plan (including explanation) for which type of detector you will need for the trace-level GC analysis of (a) 2,4-D, (b) atrazine, and (c) RoundUp. *Use an online search engine to find the structures of each of these substances.*

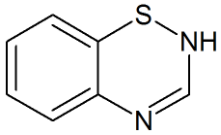
(a) 2,4-D 

(b) atrazine 

Both species (a) and (b) are combustible hydrocarbons, so an FID would respond to them. Due to the presence of chlorine in the molecule, we could use an ECD for both (a) and (b) as well. However, since we are measuring trace amounts, we would want to select the ECD to achieve the lower detection limits that detector is capable of.

(c) RoundUp contains three active ingredients, two of which thermally decompose (glyphosate and diquat), and one which is not volatile (imazapic). GC would not be an appropriate method for the analysis of RoundUp ingredients – we would want to develop an HPLC method instead.

Problem 16.9: You will be analyzing urine samples from patients taking low levels of thiazide diuretics. What type of GC detector would you choose? Explain.



Flame photometric detectors are sensitive to sulfur containing compounds. It would be suitable for this thiazide, but would also respond to other sulfur-containing substances in the urine. We would need to carefully select a column to ensure proper separation.

Problem 16.10: Calculate the compressibility correction factor, j, as a function of changing ambient pressures. Assume that the column head pressure remains constant at 70 mm Hg and the ambient pressure (in mm Hg) is:
(a) 760.0 (b) 767.3 (c) 752.6 (d) 749.7

We use Eq. 16.3 to calculate P, then use Eq. 16.2 to calculate j.

(a) $P = \frac{P_i}{P_o} = \frac{(760.0 + 70)}{760.0}$ = 1.0921 (4 significant figures, assuming the 70 mm Hg is exact)

$j = \frac{3}{2} \cdot \frac{(P^2 - 1)}{(P^3 - 1)} = \frac{3}{2} \cdot \frac{(1.0921^2 - 1)}{(1.0921^3 - 1)}$ = 0.95542 = 0.9554

(b) $P = \frac{P_i}{P_o} = \frac{(767.3 + 70)}{767.3}$ = 1.0912 (4 significant figures, assuming the 70 mm Hg is exact)

$j = \frac{3}{2} \cdot \frac{(P^2 - 1)}{(P^3 - 1)} = \frac{3}{2} \cdot \frac{(1.0912^2 - 1)}{(1.0912^3 - 1)}$ = 0.95578 = 0.9558

(c) j = 0.9549
(d) j = 0.9548

Problem 16.11: Given the following GC experimental parameters, your carrier gas flow at the column outlet measures 25.0 mL/min. The ambient temperature is 23°C. Ambient pressure is 758.8 mm Hg. The partial pressure of water at 23°C is 21.068 mm Hg. The column head pressure is 68.4 mm Hg. Calculate Fc for the following column temperatures:
(a) 150°C (b) 200°C (c) 300°C (d) 385°C

Eq. 16.8

(a) $$F_C = F_0\left(\frac{T_C}{T_a}\right)\left(1-\left(\frac{P_{H2O}}{P_0}\right)\right) = \left(25.0\,\frac{mL}{min}\right)\left(\frac{423.15K}{296.15K}\right)\left(1-\left(\frac{21.068\ mmHg}{758.8\ mmHg}\right)\right)$$

= 34.729 = 34.7 mL/min

(b) $$F_C = F_0\left(\frac{T_C}{T_a}\right)\left(1-\left(\frac{P_{H2O}}{P_0}\right)\right) = \left(25.0\,\frac{mL}{min}\right)\left(\frac{473.15K}{296.15K}\right)\left(1-\left(\frac{21.068\ mmHg}{758.8\ mmHg}\right)\right)$$

= 38.83 = 38.8 mL/min

(c) F_C = 47.0 mL/min
(d) F_C = 54.0 mL/min

Problem 16.12: Suppose we change the flow rate from 25.0 mL/min to 30.0 mL/ min. Recalculate Fc for each temperature in Problem 16.11.

See Problem 16.11 for calculation method.
(a) 41.7 mL/min (b) 46.6 mL/min (c) 56.4 mL/min (d) 64.8 mL/min

Problem 16.13: Imagine you are running GC analysis of gases trapped in ice core samples in a lab in the Arctic Circle. The lab is kept at a temperature of 12°C. Your other GC parameters include carrier gas flow at the column outlet of 25.0 mL/min. Ambient pressure is 762.8 mm Hg and the column head pressure is 78.4 mm Hg. Calculate Fc for the following column temperatures: (*Hint*: You will need to look up the partial pressure of water at 12°C.)
(a) 150°C (b) 200°C (c) 300°C (d) 385°C

P_{H2O} at 12° C = 10.5 mmHg

(a) $$F_C = F_0\left(\frac{T_C}{T_a}\right)\left(1-\left(\frac{P_{H2O}}{P_0}\right)\right) = \left(25.0\,\frac{mL}{min}\right)\left(\frac{423.15K}{285.15K}\right)\left(1-\left(\frac{10.5\ mmHg}{762.8\ mmHg}\right)\right)$$

= 36.59 = 36.6 mL/min

(b) 40.9 mL/min (c) 49.6 mL/min (d) 56.9 mL/min

Problem 16.14: Determine V_R^o for each peak in Figure 16.21.

Eq. 16.5

$P = \frac{P_i}{P_0} = \frac{(760.2 + 70.4)}{760.2}$ = 1.0926

$j = \frac{3}{2} \cdot \frac{(P^2 - 1)}{(P^3 - 1)} = \frac{3}{2} \cdot \frac{(1.0926^2 - 1)}{(1.0926^3 - 1)}$ = 0.95512

Vapor pressure of H_2O at 24.8°C = 23.475 mmHg

$$F_C = F_0 \left(\frac{T_C}{T_a}\right)\left(1 - \left(\frac{P_{H2O}}{P_0}\right)\right) = \left(28.1 \frac{mL}{min}\right)\left(\frac{558.15K}{297.95K}\right)\left(1 - \left(\frac{23.475\ mmHg}{760.2\ mmHg}\right)\right) = 51.014\ mL/min$$

First peak: $V_R^0 = jt_rF_C = (0.99512)(13.2\ min)(51.014\ ml/min)$ = 670.1 = 670. mL

2nd peak: 881 mL/min 3rd peak: 1110 mL/min 4th peak: 1210 mL/min
5th peak: 1340 mL/min 6th peak: 1430 mL/min

Problem 16.15: Determine V_N for each peak in Figure 16.21.

1st peak: $t_r' = t_r - t_m$ = 13.22 – 3.25 min = 9.97 min
See Problem 16.14 for calculation of j and F_c
Eq. 16.10: $V_n = jt_rF_C$ = (0.95512)(9.97 min)(51.014 mL/min) = 485.78 = 486 mL/min
2nd peak: t_r' = 14.10 min, V_n = 687.0 mL
3rd peak: t_r' = 18.60 min, V_n = 906.3 mL
4th peak: t_r' = 20.66 min, V_n = 1007 mL
5th peak: t_r' = 23.07 min, V_n = 1124 mL
6th peak: t_r' = 24.85 min, V_n = 1210 mL

Problem 16.16: Compare and contrast the ways a chromatographer performs a gradient separation in LC and GC.

In LC we use a solvent gradient, starting with a solvent composition that is opposite in polarity to the column polarity; those species that are similar in polarity to the column interact more strongly with it than with the solvent and so move more slowly. As the experiment progresses, the polarity of the solvent is shifted more towards that of the column, allowing those retained species to interact more with the solvent and thus move along with the mobile phase.

In GC, we use a temperature gradient, starting with relatively low temperatures and gradually increasing to higher temperatures. The vapor pressure of analytes (proportional on their boiling points) is increased with temperature, as the kinetic relationship of their interaction with the stationary phase is changed as well.

A fundamental difference between LC and GC gradients is that in LC the solvent is playing a *chemical* role – that is, the chemistry of the interaction between the gradient solvent and the analytes is important. In GC, the temperature gradient changes the vapor pressure of different analytes and changes the kinetics of their interaction with the stationary phase, but the mobile phase does not play a chemical role – its only role is to move non-retained particles along.

EXERCISE 16.1: Define each of the following terms:
a. Eluate
b. Isothermal elution
c. Thermal gradient
d. Temperature programming
e. Selectivity factor
f. Retention factor
g. Carrier gas
h. Retention volume
i. Compressibility correction factor
j. van Deemter equation
k. Split injection
l. Hold-up volume
m. Adjusted retention volume
n. NET adjusted retention volume

These terms are defined in the text. To be included.

Exercise 16.2: Calculate the theoretical plate height, H, and the number of theoretical plates, N, for peak 4 in Figure 16.21. You might need to review Section 15.2 to answer this question. It is acceptable to estimate the values of the variables you need for the calculation from the axes of the chromatogram.

Peak 4 – t_r = 23.91
W_b = 1.6
$N = 16\left(\frac{t_r}{W_b}\right)^2 = 16\left(\frac{23.91}{1.6}\right)^2$ = 3573
$H = \frac{L}{N} = \frac{30\ m}{3570}$ = 0.00840 m = 8.4 mm

Exercise 16.3: In designing a separation method, we use the theoretical plate as a representation of the separation efficiency of the column. So it might seem reasonable to assume that more theoretical plates is always better than fewer theoretical plates. Having said that, why do we not automatically increase the column length anytime we need to improve separation?

The most practical reason is cost – to increase the column length, we would need to purchase a new column, install it, condition it, and tweak our method for the new column. Columns can be quite expensive, and there is a cost associated with the time it takes to install implement the column.
Another reason is that for any set of solutes, there is a practical limit to length – even with open tubular columns, the pressure required to move the mobile phase through increases with length, and there is always a sacrifice of peak width with length of time spent in a column.

Exercise 16.4: Describe the effect of each variable in Equation 16.11 on column efficiency.

A, Eddy diffusion. For packed columns, increased variability in eddy diffusion will increase H, which will decrease efficiency.

B, Longitudinal diffusion. Regardless of type of column, the B term always plays a part. Substances will diffuse away from a region of high concentration, so the longer any given solute band remains in the column, the greater the band broadening. A larger B term will increase H, decreasing N.

C, Mass transfer term. Slow kinetics of mass transfer within the stationary phase and between the stationary and mobile phases will increase H, decreasing efficiency.

v, mobile phase velocity. The effect v is relative to the B and C terms. A high flow rate will decrease the B term (increasing efficiency) but increase the C term (decreasing efficiency). The experimenter must seek an optimum flow rate to balance out these two terms.

Exercise 16.5: Rank the following compounds in terms of the expected elution order for a capillary GC separation run under isothermal conditions.

(a) Ethanol
(b) n-Propanol
(c) Methanol
(d) n-Pentanol
(e) n-Butanol

Without knowing the stationary phase, we must assume only boiling point plays a part; as such, the compounds will elute in order of increasing boiling point:
MeOH : EtOH : PrOH : BuOH : PeOH

Exercise 16.6: Sketch a schematic for a split flow injector and in your own words, describe each component.

See Figure 16.7 and associated text.

Exercise 16.7: Sketch a schematic of an FID and in your own words, describe how it functions.

See Figure 16.10 and associated text.

Exercise 16.8: Sketch a schematic of a TID and in your own words, describe how it functions.

See Figure 16.11 and associated text.

Exercise 16.9: Sketch a schematic of an ECD and in your own words, describe how it functions.

See Figure 16.12 and associated text.

Exercise 16.10: Sketch a schematic of a PID and in your own words, describe how it functions.

See Figure 16.13 and associated text.

Exercise 16.11: Sketch a schematic of an FPD and in your own words, describe how it functions.

See Figure 16.14 and associated text.

Exercise 16.12: Sketch a schematic of an AED and in your own words, describe how it functions.

See Figure 16.15 and associated text.

Exercise 16.13: Sketch a schematic of a "light pipe" interface for a tandem GC-FTIR and in your own words, describe how it functions.

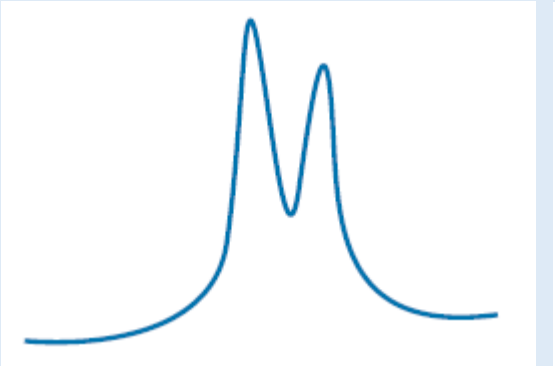
See Figure 16.18 and associated text.

Exercise 16.14: Sketch a schematic of a jet separator for a tandem GC-MS and in your own words, describe how it functions.

See Figure 16.19 and associated text.

Exercise 16.15: Imagine the following two peaks coeluted shortly after the hold-up volume. Which of the following actions would be the "*best*" way to improve the resolution? Defend your answer and explain why you rejected the other options.

(a) Use a longer column.
(b) Use a faster carrier gas velocity.
(c) Use a slower carrier gas velocity.
(d) Use a higher column temperature.
(e) Use a cooler column temperature.



Without knowing any other experimental details and assuming we want to accomplish the separation today (rather than in a couple of days), the best way would be (e), using a cooler column temperature. By decreasing the temperature, we can maximize the differential between the kinetics of solute interaction with the stationary phase, such that the later eluting peak takes just a bit longer to elute.

We can use the opposing argument to decide against option (d).

Option (a) is something that cannot be done in a timely fashion, unless we happen to have a longer column on the shelf. Generally, we would need to order a new, longer column. Using a higher carrier gas velocity (option b) would decrease the B term somewhat, but it is a reciprocal relationship and the relative gain in the B-term drops off dramatically as flow rate increases. We are not likely to gain much in terms of separation efficiency.

Similarly, using a lower gas velocity (option c) would decrease the C term, but at lower flow rates, the B term increases asymptotically. Without knowing where along the B and C curves we are, we cannot be sure that decreasing the velocity will help.

Exercise 16.16: Which of the various detector types discussed in this chapter is the detector of choice for the detection of halogenated organic compounds?

ECD – it is very sensitive to halogens with large electron clouds (chlorine and larger).

Exercise 16.17: In all chromatographic experiments, controlling the injection volume is an important consideration. Compare and contrast the methods used to control injection volume in LC and GC.

In LC, we use an injector fitted with a loop of fixed volume. That is, we can be fairly certain that each injection has, to a high degree of reliability, the same injection volume.

In GC, it is much more difficult with a typical GC syringe to generate reproducible injection volumes. Further, in many cases we use columns that require very small injection volumes – far smaller than we can physically create. In those cases, we can use a split flow injector to allow only a small fraction of the actual injected volume onto the column.

Exercise 16.18: Both the TCD and FID are considered universal GC detectors, but the TCD is considered "more" universal than the FID. If the TCD is so much more universal, why use a FID at all?

The FID is far more sensitive and it has a larger linear dynamic range of concentrations over which we can measure a response. Further, many analyses that we care about involve combustible organic compounds, so the FID responds to the vast majority of the types of analytes we will want to study.

Exercise 16.19: Compare and contrast the operation of an atomic emission spectrometer (Chapter 9) with the operation of an AED.

At the most fundamental level, they are identical. The only real difference is in how we introduce the sample to the plasma. In AES, we nebulize a solution containing our sample and introduce the fine mist into the plasma, at which point it is vaporized and atomized. In an AED, our sample is already in the gas state (having been vaporized in the GC), and then it is introduced within the carrier gas into the AED plasma.

Exercise 16.20: Compare and contrast the features of the mobile phase in GC with that in LC. Describe the important properties of the mobile phase in each technique and its impact on the efficiency of the separation.

The instructor will need to determine how focused or how comprehensive the answer should be. A comprehensive answer will need to include reverse and normal phase LC techniques and relate each to the analogous stationary phases in GC. A discussion of analyte polarity with respect to the stationary phases should also be included.

Exercise 16.21: Answer the following questions for the chromatogram given in Figure 16.21.

(a) Calculate the selectivity factor and resolution for the first two peaks (see Section 15.2 for a review on calculating resolution).

(b) Calculate the number of theoretical plates for the first and last peak.

(a) $\alpha = \frac{k'_r}{k_r} = \frac{t_{r,2}-t_m}{t_{r,1}-t_m} = \frac{17.35-3.25}{13.22-3.25} = 1.41$

(b) First peak: $W_b = 1.24$ $\quad N = 16\left(\frac{t_r}{W_b}\right)^2 = 16\left(\frac{13.22}{1.24}\right)^2 = 1820$

last peak: $W_b = 1.43$ $\quad N = 16\left(\frac{t_r}{W_b}\right)^2 = 16\left(\frac{28.1}{1.43}\right)^2 = 6180$

Exercise 16.22: Which term of the van Deemter equation is most influential in a GC separation? Explain how this factors into the general elution problem.

The C term is most influential. In most cases with GC, there is no A term because we use an open tubular capillary. The B term drops off quickly with increasing flow rate and we will select a flow rate that optimizes the B term. However, we can modify the kinetics of mass transfer within the stationary phase and between the mobile and stationary phases by modifying temperature – GC's are specifically equipped to allow us to accomplish temperature programming to achieve a gradient elution.

Exercise 16.23: The peak capacity is the total number of observable peaks in a chromatogram under a given set of experimental conditions. The equation for determining the peak capacity is:

$$n_C = 1 + \int_{t_m}^{t_R} \left(\frac{\sqrt{N}}{4t}\right) dt$$

where n_c is the peak capacity, t is the separation time, tm is the hold-up volume, and tR is the retention time of the last eluting peak. If you ignore the fact that the number of theoretical plates varies with the retention time of an individual peak, determine the peak capacity of a system given, t_m = 6 min., t_r = 40 min, and N = 52,000.

Exercise 16.24: What type of detector would you select in the routine quantitative determination of Prozac in urine samples by GC?

Either an FID or a MS detector would be best. The fluorine atoms, while halogens, do not have the large electron cloud that would make them easily measured with an ECD, but the molecule is a large, complex hydrocarbon that will produce a strong FID signal and will produce many fragments in an MS.

Exercise 16.25: In the analysis of a series of n-alkanes, would you prefer to use a GC-FTIR or a GC-MS instrument? Explain.

The n-alkane will not show significantly different FTIR spectra – they will basically have the same CH and CC stretches and similar bending vibrations. While the fragmentation pattern in the MS will be similar, the mass of the precursor ion will be distinctive.