# CHM 317H1S
# Winter 2018

# Section B - GC & GC-MS

# B. Gas Chromatography and GC-Mass Spectrometry

## 1. List of Experiments

1. Qualitative & Quantitative Analysis by Gas Chromatography
2. TurboMass Tutorial* and Qualitative Analysis by GC-MS
3. Analysis of Foodstuffs for BHT and BHA

* This tutorial ***must*** be completed during the week ***before*** using the GC-MS!

## 2. Locker Inventory

Glassware for this experiment is set aside for you in a designated locker in room LM6. This should include the following items; please check that you have everything below at the beginning of your first laboratory period for this set of experiments.

| ✔ | Quantity | Common Items for Experiments B1–B3 |
|---|---|---|
| | 6 | Watch glasses to cover 50 mL beakers |
| | 6 | 50 mL beakers |
| | 3 | 100 mL beakers |
| | 8 | 10 mL volumetric flasks with stoppers |
| | 12 | pasteur pipettes with bulbs |
| | 2 ea. | 5 and 10 mL Mohr (graduated) pipettes |
| | 2 ea. | 1, 2, 5, and 10 mL transfer pipettes |
| | 2 | 3-Way bulb pipette filler |
| | 1 | Wash bottle for acetone |
| | 1 each | Large and small spatula |
| | 1 box | KimWipes™ |

| ✔ | Quantity | Extra Items for Experiment B3 |
|---|---|---|
| | 1 | 1.000, 5.00, and 10.00 mL volumetric flasks with stopper |
| | 1 | 100.0 mL volumetric flask with stopper |
| | 8 | 25.00 mL volumetric flasks with stoppers |

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## 3. Instrumentation

During this set of experiments, you will be using a variety of instrumentation featuring some of the more commonly-used detectors employed in gas chromatography:

- GC7, GC4, GC5, & GC6:
  Perkin-Elmer AutoSystem XL gas chromatograph with flame ionization detector (FID) and programmable split/splitless injector (PSSI). (Instruments are configurable to use other types of detector also.) Columns may include the following examples:
  (a) Supelco Simplicity 5 fused silica capillary (30 m long × 0.32 mm internal diameter, 0.25 μm thick liquid stationary phase, $T_{max}$ = 320°C)
  (b) Supelco Simplicity 1 fused silica capillary (30 m long × 0.32 mm internal diameter, 0.25 μm thick liquid stationary phase, $T_{max}$ = 320°C)
  Note that the actual make, type, and size of column will be indicated on a small label taped to the front of each instrument.

- GCMS:
  Perkin-Elmer AutoSystem XL gas chromatograph with autosampler and Perkin-Elmer TurboMass mass spectrometric detector (MSD) and programmable split/splitless injector (PSSI). Column 1 is a Phenomenex ZB 5 fused silica capillary column (30 m long × 0.25 mm internal diameter, 0.25 μm thick liquid stationary phase, $T_{max}$ = 320°C)

All the GCs in Analest use hydrogen gas as the mobile phase, generated electrolytically using a Whatman Hydrogen Generator. Instrument control and data collection is performed using Perkin-Elmer Client/Server TotalChrom Navigator software version 6.1.2 and/or Perkin-Elmer GC/MS TurboMass software version 4.1.1.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## 4. General Operating Instructions

The following pages provide general information on using the GC-FID and GC-MS instruments in the Analest laboratory. There are similarities and differences between the operating instructions for these instruments; please take time to read through these instructions carefully **before coming to the laboratory.**

### 4.1 Starting the Instrument:

(a) The GCs are normally up and running all the time. In the event that they are not, check that the $H_2$ carrier gas valve behind the instrument (in the narrow access space between the benches) is open and that the gauge indicates sufficient gas pressure. When using a GC with an FID, similarly make sure that the air valve is open and sufficient pressure is indicated on the gauge.

(b) If the computer adjacent to the GC is not on, start it up. Once it has booted, press the **Ctrl**, **Alt**, and **Del** keys simultaneously to call up the login window. Login to the Analest network using the user name and password for the particular instrument you wish to use:

GC7: username chm317gc7 and password gc7
GC4: username chm317gc4 and password gc4
GC5: username chm317gc5 and password gc5
GC6: username chm317gc6 and password gc5
GCMS: username chm317gcms2 and password gcms2

(c) Launch the software to control the instrument and collect the data:

**GC7/4/5/6:** double-click on the **TCNav** shortcut icon on the desktop to launch the TotalChrom software; **proceed to step 4.2**

**GCMS2:** double-click on **TurboMass** shortcut icon on the desktop to launch the TurboMass software; **proceed to step 4.5**

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## 4.2 Setting up the TotalChrom Software (GC–FID):

(a) On the main screen, locate the **Instruments** box listing all the instruments currently running under TotalChrom. If the button corresponding to your instrument is not already selected, click on it.

(b) Select **CAM Administration** from the **Admin** menu. In the **CAM Admin Tool** window, double-click on **TotalChrom Servers**, and then click on instrument to be used (e.g. **GC2**) to select it. Click on the first icon in the toolbar (the padlock) to unlock the instrument, and then close the **CAM Admin Tool** window.

(c) Linking the computer and instrument: In the main window, click on the **Run** box and select **Attach** from the popup menu. Wait until the **Status** box in the main window indicates that the instrument/interface has been successfully attached, and then select **Take Control** from the same popup menu.

(d) Setup and Method: click on the large **Setup** button in the main window. In the dialog box that opens, make sure that the **Method** radio control at the top is selected. There are a number of line items with text entry areas beside them. The **Method:** line should show the text:

C:\TC4\CHM317\FID1.mth

If not, you can either type this in or click on the button with the folder icon at the end of the line to browse to the appropriate file. Similarly, make sure that the **Data Path:** item contains the text:

C:\TC4\CHM317\

Enter a suitable name for the **Base File Name:** – the software will automatically add a sequential number to this name for each chromatographic run.

Under the **Processing** heading, make sure that **Suppress processing** is unchecked and that **Suppress reports/plots** is checked. Click on the **Bind** button – this makes sure that the operating conditions transferred to the instrument cannot be changed accidentally once you have started your experiment. Finally, click on the **Vial list** button.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

(e) Vial List: the vial list lets the software know how many samples you intend to run, and enables you to provide a title for each one. The sequence editor opens the vial list in a spreadsheet-style window. Enter a name for each one. Select **Insert** from the **Edit** menu or type a **ctrl–A** to add extra lines if necessary. Once you are done, save the vial list and exit the sequence editor.

If you do not have enough rows in your vial list, the software will disable the method after the last row has been used. You should *always* add more rows to the vial list than you think you will need!

(f) Now click the **OK** button in the **Setup Instrument** dialog box. Information will be transferred from the computer to the instrument; when setup is complete, all the lines in the **Status** box will be in green text and read "**Ready**". If you want to review the settings or check current values at any time after this, click on the large **Details** button in the main window. To print the setup parameters, click on the large **Method** icon in the main window. In the resulting window, select **Print** from the **File** menu; make sure that only those options you require are checked in the dialog box that comes up before printing (i.e. instrument parameters.)

(g) Summary of the FID1 method file (for Expt. B1 – see p.64 for B3):

| | |
|---|---|
| GC Method File: | CHM317FID1 |
| Column: | DB5 |
| Length: | 30 m |
| Carrier Gas: | Hydrogen |
| Split Ratio: | 25:1 |
| Column Pressure: | 4 psi |
| Oven Temperature: | 120 °C |
| Injector Temperature: | 250 °C |
| Injection Volume: | 1 μL |
| Detector: | Det1 |
| Detector Temperature: | 220 °C |

Note: Split Ratio = $(F_S + F_C)/F_C$ where $F_S$ = Split Flow (mL/min) and $F_C$ = Column Flow (mL/min).

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

(h) Lighting the Flame Ionization Detector: on the GC keypad, press the **Detector Control** button, then the **Enter** key, and finally the **Set** key, in that order. The ignition filament will glow and there should be a loud "pop" as the air–hydrogen flame ignites. You should also see the temperature and current reading on the instrument display panel. Once the flame is lit, press **Status Escape** on instrument keypad.

## 4.3 Running Samples on the GC–FID:

(a) Make sure that all the items in the **Status** panel are green and show "Ready". You can check the various current temperatures for the different parts of the instrument by clicking on the large **Details** icon. The instrument will take a little while for all temperatures to reach their set points.

(b) Once the gas chromatograph is fully equilibrated and the **Status** panel shows that everything is ready, you may inject your sample. Rinse the syringe with the appropriate solution a few times, fill it with the required volume of the solution, and wipe off the outside of the syringe needle with a KimWipe™ . Insert the syringe into the injection port, and depress the plunger smoothly and rapidly whilst **simultaneously** pressing the green **Run** key on the GC key panel. As soon as you have done this, remove the syringe from the injection port.

**Note:** The key to obtaining good GC results is to perform the sample injection as reproducibly as possible. A rapid injection is preferred; a slow injection will result in very broad peaks that may not be fully resolved. You can prevent the plunger from being accidentally depressed while inserting the syringe into the injection port by holding the shaft of the plunger in place with one finger. Ask for a demonstration of good injection technique, and practice with an <u>empty</u> syringe while waiting for the instrument to equilibrate.

(c) Monitoring the chromatogram: Click on the large **Real-Time Plot** icon in the main TotalChrom window. This will show the raw data while being collected by the computer. You can use the **Option** menu in this window to rescale the display if necessary.

(d) Processing the Chromatogram: This can be done at any time once a particular chromatographic run has been completed, i.e. you can start the

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

next run, then go back and analyse the data from the preceding one. Minimize the real-time plot window, and click on the large **Results** icon in the **Reprocess** area of the main window. Open the file corresponding to the experiment you have just run: this will have the base file name you set in the method, followed by a time-stamp, with a .rst suffix.

In the resulting chromatogram display, check that the software has correctly identified the various peaks in your sample, and that the baseline drawn across the bottom of each peak is reasonable. Also check that the software has not identified noise peaks as sample peaks: noise peaks are typically small and quite narrow, whereas your sample peaks will be broader and exhibit the expected Gaussian (or skewed-Gaussian) shape.

- to remove noise peaks: click on the **Process** button and choose **Noise/Area Threshold** from the drop-down menu. Click and drag along a region of the baseline containing the noise peaks you wish to remove. A window will appear suggesting appropriate thresholds based on the size of these peaks; either change these values or accept them, then click **OK**.

When you are satisfied, print the results by selecting **Print** from the **File** menu; choose the appropriate formats for **Report** ("landscape") and **Replot** (checked), then click **OK**.

## 4.4 Shutting Down the Software:

(a) Make sure that the instrument is not running (all the status text items should be green.)

(b) Click on the large **Setup** icon on the main page, click on the **Method Folder** icon, choose the **shutdwn.mth**, and click the **Select** button. Enter a suitable name in the **Base File Name** box (e.g. "shutdown") and click **OK**.

(c) Click on the large **Run** icon on the main page, and select **Release Control** from the drop-down menu. Then click on the **Run** icon again and select **Detach**. When the status box text turns red, close the TotalChrom program, and log off from the computer. Do **not** turn the GC off – the GC must be left on to prevent oxygen from the atmosphere diffusing into the column.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

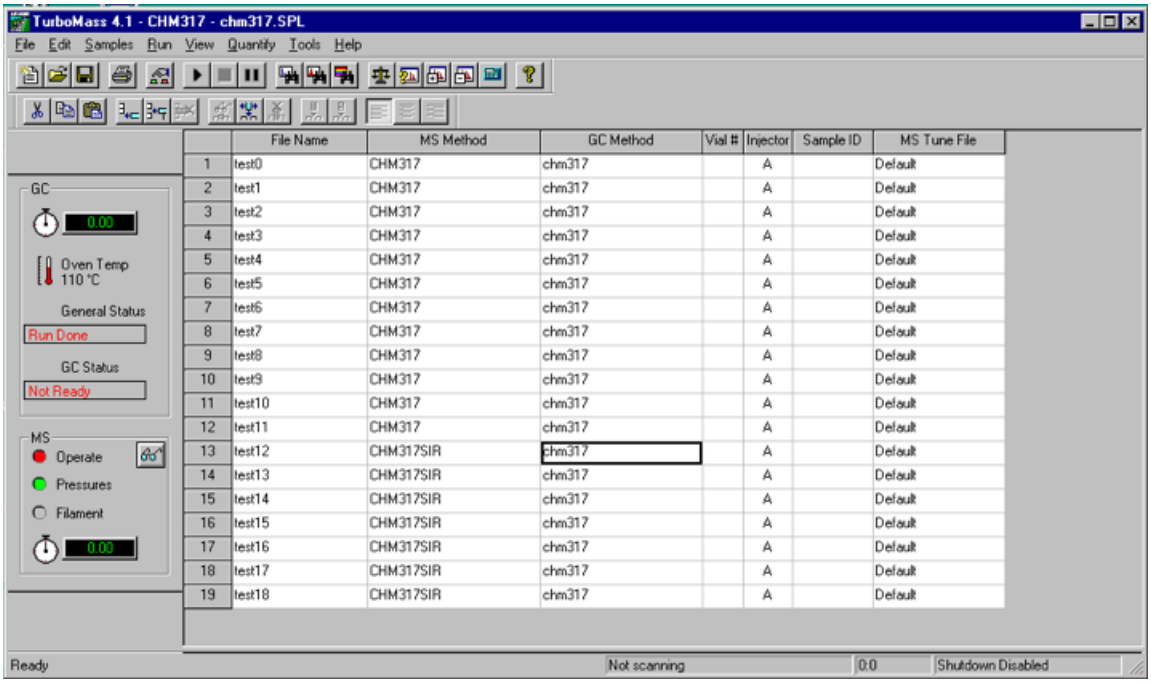
(d) Make sure you clean up any solutions, KimWipes™, etc. from the area around the GC, and that you have collected all your reports from the printer.

## 4.5 Setting up the GC–MS Software:

(a) When you launch the TurboMass software, you will be prompted for a user name and password again. Type:

User: `chm317gcms1`
Password: `gcms1`

The main page of the GC-MS software will open; note the two panels on the left-hand side indicating the current GC and MS status and conditions. The **GC Status** text is colour-coded: red means that no method has been transferred to the GC or MS, while blue means that the system is either setting up, running, or completing data transfer.

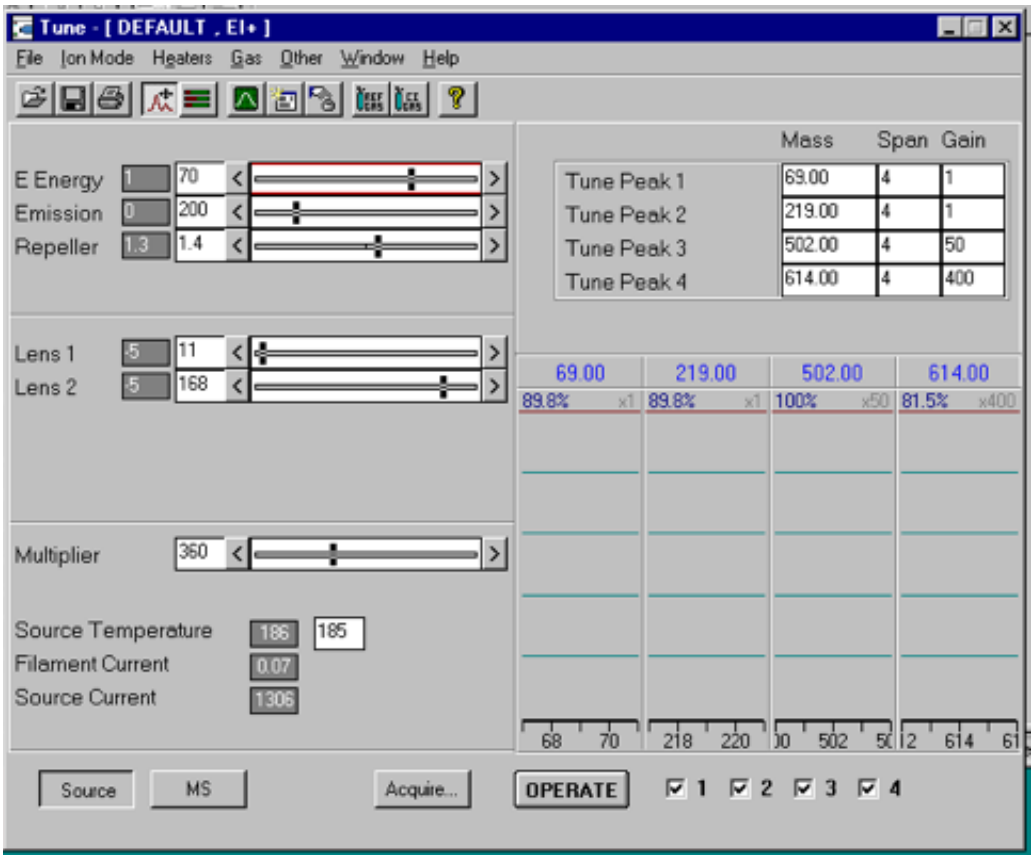



| | File Name | MS Method | GC Method | Vial # | Injector | Sample ID | MS Tune File |
|---|---|---|---|---|---|---|---|
| 1 | test0 | CHM317 | chm317 | | A | | Default |
| 2 | test1 | CHM317 | chm317 | | A | | Default |
| 3 | test2 | CHM317 | chm317 | | A | | Default |
| 4 | test3 | CHM317 | chm317 | | A | | Default |
| 5 | test4 | CHM317 | chm317 | | A | | Default |
| 6 | test5 | CHM317 | chm317 | | A | | Default |
| 7 | test6 | CHM317 | chm317 | | A | | Default |
| 8 | test7 | CHM317 | chm317 | | A | | Default |
| 9 | test8 | CHM317 | chm317 | | A | | Default |
| 10 | test9 | CHM317 | chm317 | | A | | Default |
| 11 | test10 | CHM317 | chm317 | | A | | Default |
| 12 | test11 | CHM317 | chm317 | | A | | Default |
| 13 | test12 | CHM317SIR | chm317 | | A | | Default |
| 14 | test13 | CHM317SIR | chm317 | | A | | Default |
| 15 | test14 | CHM317SIR | chm317 | | A | | Default |
| 16 | test15 | CHM317SIR | chm317 | | A | | Default |
| 17 | test16 | CHM317SIR | chm317 | | A | | Default |
| 18 | test17 | CHM317SIR | chm317 | | A | | Default |
| 19 | test18 | CHM317SIR | chm317 | | A | | Default |

The sample list window in the TurboMass software for running the GC-MS

(b) **Starting the MS:** click on the 'glasses' icon in the toolbar of the main window (see screen-shot above); this launches the tuning page for the MS. In the **Tune** page (below), click the **OPERATE** button to turn on the MS ionization filament. **Do *not* adjust any other parameters in this window.** Close the window and click the **OK** button when advised that the filament is on.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry




The TurboMass **Tune** window, showing the source controls and the **OPERATE** button.

(c) **Calibrating The MS:** this is performed periodically be a member of Analest staff, **and should *not* be attempted by students.**

(d) **Setup and Method:** In the main window, select **Open Project** from the **File** menu. Choose CHM317.PRO and click **OK**. This will open a spreadsheet in the main window with a number of different headings (see illustration on preceding page). To change or view the settings, double-click on the cell immediately below the corresponding heading. Each row will be used for a separate run; you should avoid over-writing existing files. Once you have the first row configured, you can use the software to copy your settings to the following rows.

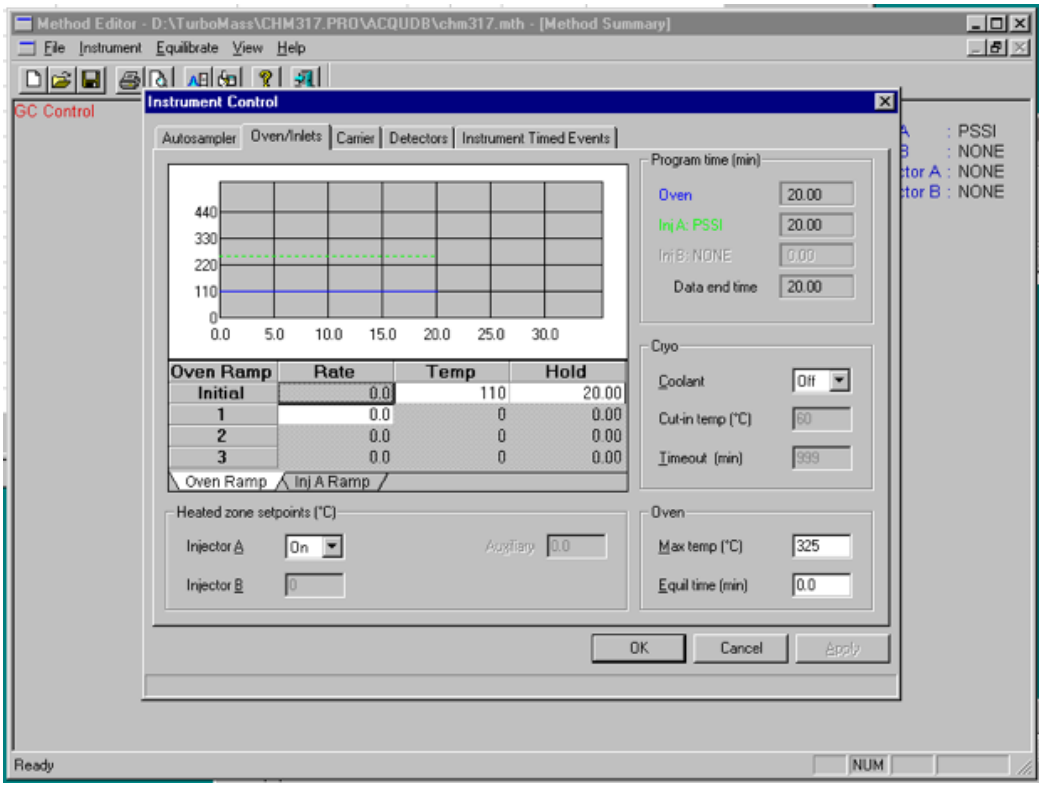
**File Name**: This is the file that will contain the raw chromatographic data. Choose a unique base file name containing *only* upper and lower case characters; **do *not* use spaces or punctuation marks or you will lose your data**! For example, you could use a combination of your initials. You can use the **Samples** menu (or right-click on a row) to add extra rows; you should always have more rows than you think you will need!

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

Click (or right-click) on the **File Name** column title, and select **Fill→Series...** from the **Samples** menu to set a series of incremented file names for your experiment.

**MS Method**: if this is not already set, double–click on the first cell in this column and choose **CHM317** from the drop-down menu list. Click (or right-click) on the **MS Method** column title, and select **Fill→Down...** to add the same method to all rows. To view the current method settings, right–click on the first cell in this column and select **Open** from the popup menu. A dialog box will open showing the range of mass values (40 – 250 amu) and solvent delay time (usually between 1.20 and 1.50 min – check with your demonstrator if any other value is shown.) Close the **MS Method** window, answering **Yes** when prompted.

**GC Method:** double-click on the cell below the **GC Method** heading and select **CHM317** from the drop-down menu list. Use the same procedure as in the previous step to make sure that all rows are using the same initial method. Right-click on the method name and select **Open** from the popup menu. In the new window that opens, select **Oven/Inlets** from the **Instrument** menu. This window (see below) allows you to examine the current settings for the column oven. Close the window, answering **Yes** when prompted.



The instrument control window within the TurboMass chromatographic method editor

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

**Sample ID:** leave this field blank for now. When you start collecting data, you can enter information into this field to help identify the sample or standard being run. This is better than trying to use the file name, as it avoids lost data!

**MS Tune File:** this should be set to **Default** for all sample rows.

## 4.6. Running Samples on the GC–MS:

(a) In the main TurboMass window, highlight the desired sample row of settings by clicking on the sample number (e.g. "run1"); the highlighted row will turn black. Click on the **Run** icon in the toolbar (the right–pointing arrow head); click the **OK** button in the resulting **Start Sample List Run** dialog box that appears, then click on **Yes**. Watch both the **GC status** and **MS status** entries; when these read "Waiting for injection" and "Ready", respectively, and all the status indicators are green, the instrument is ready.

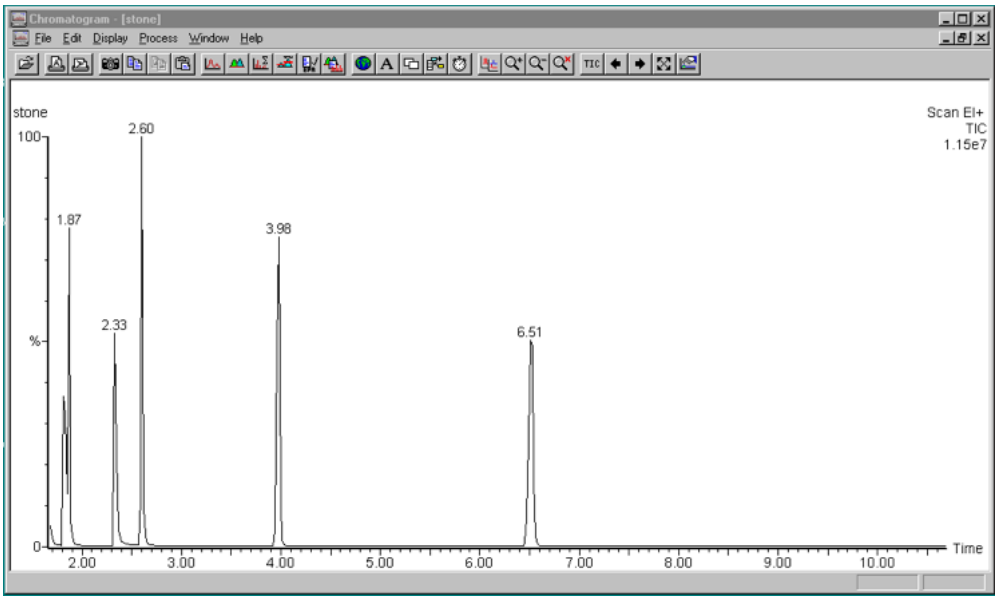


The status area (left) and button toolbar (above) of the main TurboMass sample window. After the first run, make sure that the general and GC status boxes look like this *before* clicking the "go" button to setup for another injection.

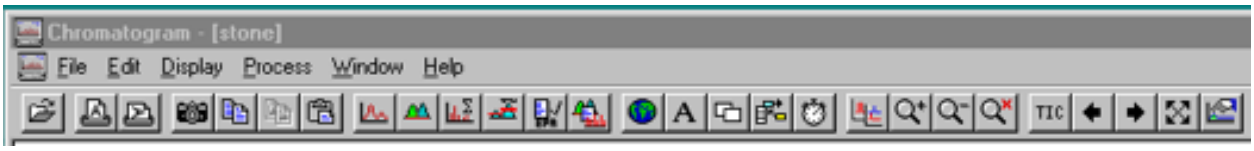
(b) Inject the sample in the same way as for the GC-FID (section 4.3(b)), while simultaneously pressing the green **RUN** key on the GC key panel; remove the syringe immediately following injection.

(c) Monitoring the chromatogram: Either select **Chromatogram** from the **View** menu or click on the middle of the three tool-bar buttons with **binoculars** in their icon (see above). In the resulting window, make sure that the **stopwatch** button in the toolbar is engaged (down); this enables the software to display the current chromatographic data in real time.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry




The TurboMass chromatogram window, showing a run in progress.




The menu and button bars from the TurboMass chromatogram window

Please note that there is another button that toggles between single and multiple chromatograms within the display window (the first one in the group with the magnifying glass icons); this should normally be disengaged (**up**). To remove a chromatogram from the window, click within the plot area and either press the **Delete** key or select **Remove...** from the **Display** menu. If you wish to examine the mass spectrum of individual peaks *while* data is still being collected, click on the stopwatch button to freeze the display, then follow the instructions below. To update the display, either click on the stopwatch a second time or on the rescale button (the one on the far right with four arrows).

(d) Stopping a run: You can stop a run in progress at any time, although you should *never* do this until *all* sample components have eluted form the column. Switch back to the main TurboMass window and click the red 'stop' button, then answer **Yes** to the following dialog boxes.

Because of the way the software is written, the status display will temporarily turn green *before* all the mass spectral data has been transmitted from the instrument to the computer. You ***must*** wait until the software has

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

finished collecting data and resetting the GC and MS status displays before attempting to start another run – the text in both boxes should then be **red** (see illustration at top of preceding page).

(e) Viewing the mass spectrum: you can view the mass spectra for any peak by double-clicking on it to open the mass spectrum display. Pressing the **F1** key on the keyboard will activate the library search software. Note that **you** have to decide which of the suggested structures is reasonable; problems can be encountered if you have overlapping (unresolved) peaks and/or very small peaks for which the parent ion is not detectable.

(f) Peak integration: the easiest way to obtain peak areas is to click on the "integrate peaks" button in the toolbar; this is the one showing two peaks filled with different colours (see above illustration). If your peaks exhibit *tailing*, or are not resolved at the baseline, you may need to adjust the start and end points used for calculating the area. To do this, click on the very last (right-hand) button in the toolbar. This will add boxes to each peak that can be dragged to change the integration. You can also zoom in on a particular region by clicking and dragging the mouse horizontally: a line with a vertical bar at each end will show the region you are selecting.

If you have unresolved peaks, or peaks with poor baseline separation, select **Edit→Integrated Peaks...** With this dialog open, right-click and drag across the peak to enter the start and end times for integration into the relevant fields in the dialog box (you can also enter them manually). Click on the **Add** button, and then click on the **Integrate** button to exit the dialog. The peak retention time and area will be displayed at the peak maximum, and the peak will be shaded so that you can see what has been included in the calculation.

## 4.7. Shutting Down the Software:

(a) Make sure that the instrument is not running (all the status text items should be green.) Click on the **Finger** icon in the main window toolbar to open the **Acquisition** screen; click on **GC** and choose **Release Control** from the drop-down menu, then exit the GC frame.

(b) Click on the **Glasses** icon in the MS frame; click on the **OPERATE** button in the resulting **Tune** page to turn the filament off.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

(c) Make sure that the **GC Status** display is empty, then exit the TurboMass software and logoff from the computer. Leave the GC-MS turned **on.**

(d) Note for TA: please notify lab technical staff that you are done with the instrument so they can set the correct shutdown conditions on the GC-MS (oven temperature of 40°C and carrier flow of 0.5).

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

# 5. Experiment B1: Qualitative & Quantitative Analysis by GC-FID

This experiment is adapted from expt. 12–2 and 12–3 from "Chemistry Experiments for Instrumental Methods" by Sawyer, Heineman, and Beebe. You will perform a qualitative analysis of a mixture of 1-octanol, 2-octanone, bromobenzene, decane, undecane, dodecane, and tridecane, dissolved in dichloromethane by retention time matching, as well as determine the concentration of one the components using area integration. You will also use nonane as an internal standard for quantitative analysis. All the measurements in this experiment are performed under *isothermal* elution conditions.

After performing this experiment, you should:

- Understand the basis of qualitative analysis in gas chromatography
- Understand the use of an internal standard for improved quantitative analysis
- Start to develop an understanding of retention mechanisms in GC

**IMPORTANT PRECAUTIONS:**

- ➔ **Do NOT wash your glassware with water or use compressed air to dry it**. When working with non-aqueous solvents such as dichloromethane or ethyl acetate, you should avoid introducing any water into the glassware. Rinse glassware *with the solvent you are going to use* instead. If this is insufficient to clean an item properly, use acetone, and then rinse with the solvent afterward.
- ➔ **Limit exposure to solvent fumes**. Make sure you stopper all flasks when transporting or weighing, and perform all rinsing and dilution operations in the fume hood. Do ***not*** leave open beakers of solvent on the bench.
- ➔ **Dispose of all organic waste properly**. Any solution containing dichloromethane or bromobenzene ***must*** be emptied into the halogenated waste container. Other hydrophobic organic waste (ethyl acetate, alkanes, benzene, *etc*.) can go in the large red waste can. Any water-soluble organics (*e.g*. alcohols) should go in the aqueous organic waste for the HPLC experiments in the other fume hood.

**Before the lab**:

1. In addition to your regular preparation, draw up a table showing how you will make up the standards outlined in step 4, and confirm this with your demonstrator before making your solutions.
2. In addition to your regular preparation, research the chemical resistance of the different types of gloves that are often used in chemical laboratories. Make sure you note which are the *most appropriate* type to wear when handling the chemicals you will be using in this experiment!

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## A. Instrument Setup and Retention Time Characterisation

1. Start up the assigned GC as outlined in section 4 above, and allow the instrument to equilibrate. It should be equipped with a capillary column having a stationary phase consisting of 5% phenyl and 95% methyl groups (often referred to simply as a 'phenyl' column). Make a note of the exact column type, dimensions, and manufacturer from the label on the front of the instrument. The following lists typical conditions used for this experiment – note that the column temperature may be varied slightly by lab staff in order to compensate for aging of the column:

| | |
|---|---|
| GC Method File: | CHM317FID1 |
| Column: | DB5 or equivalent (5% phenyl phase) |
| Column dimensions: | 30 m × 0.23 (or 0.32) mm × 0.25 μm |
| Carrier Gas: | Hydrogen |
| Split Ratio: | 25:1 |
| Column Pressure: | 4 psi |
| Oven Temperature: | 120 °C (isothermal) |
| Injector Temperature: | 250 °C |
| Injection Volume: | 1 μL |
| Detector: | Det1 |
| Detector Temperature: | 220 °C |

**Note:** Split Ratio = $(F_s + F_c)/F_c$ where $F_s$ = split flow rate (mL/min) and $F_c$ = column flow rate (mL/min); the split ratio reduces the sample volume entering the column from that injected by the syringe *i.e.* a split ratio of 25:1 results in 1/25$^{th}$ of the original injection volume actually entering the column.

2. While the instrument is equilibrating, accurately prepare 10.00 mL of a solution containing ~1.0 mg/mL of the first compound (1-octanol) in dichloromethane: dispense the liquids by *weight*, **not** volume, using an analytical balance.

➔ **Note:** the total mass required is approximately 1 drop from a disposable Pasteur pipette (~10–20 mg, depending on the liquid). Dispense one drop, and record its actual mass as accurately as possible; do **not** try to get exactly the right mass in the flask – you will be there all day!

3. When the GC-FID is ready, use a 2.0 or 5.0 syringe to inject 1.0 μL of your standard solution and record the chromatogram.

**Be very careful not to bend the needle, or pull the plunger out of the syringe!**

Use this to determine the retention time for 1-octanol under the instrument conditions being used. Note that you can stop the GC run as soon as both peaks (*i.e.* solvent and sample) have *fully* eluted: select **Run→Stop** from the menu in the real-time plot window. While this standard is running, start making solutions for each of the other compounds in dichloromethane in the same manner

4. Run each standard in turn as it becomes ready, taking turns to prepare the solutions and run the GC. As each run finishes, process the chromatogram by following the steps in section 4.3(d) (page 8). Compile a table in your lab notebook listing the retention time for

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

each compound *as you run each chromatogram*. You will use this information to identify each of the components in your sample mixture.

➔ Be sure to note any unexpected peak shapes in your lab notebook. Print only one copy of the chromatograms for each of your standards for reference; you will *not* need to submit these with your formal reports. Octanol is a particularly good compound to use in order to check the state of a GC column: if the stationary phase has degraded, the octanol peak will be much smaller than expected and exhibit an extremely long tail.

**B. Identification of an Unknown Mixture**

5. Inject 1.0 μL of the ***undiluted*** unknown mixture, and allow the run to go to completion. Use the results obtained in step 2 to identify the components in your unknown by matching the retention times. Note: these will vary slightly, depending on how accurately you performed you sample injection and pressed the **RUN** button on the GC. The software may report up to three decimal places for the retention time; generally, only the first two are actually significant. Make sure that you each have a copy of the chromatogram for the unknown mixture, that you write your name and demo group number on each copy, and that you label all the peaks.

**C. Quantitative Analysis by Internal Standard**

➔ You may choose any one of the standard compounds for this part of the experiment ***with the exception of*** octanol; check with your demonstrator before making up your solutions. You will be adding nonane as in internal standard to all your calibration solutions, as well as a known dilution of the unknown sample mixture.

6. When you have made the test solutions outlined above, choose **one** of the compounds for quantitative analysis. Accurately prepare 10.00 mL of a solution containing 1.0 mg/mL of your compound of choice and 1.0 mg/mL of nonane as internal standard, and dilute to the mark with dichloromethane.

➔ **Be sure to record the weight of both the compound *and* nonane in <u>each</u> flask, using an analytical balance.**

Using the *original* solution of this compound with **no** nonane, make an additional three standards covering the range 0.1 – 1.0 mg/mL, each containing 1.0 mg/mL of nonane. To do this, first accurately weigh one drop of nonane into a clean, dry 10.00 mL volumetric flask. Next, use a transfer pipette to add an appropriate volume of your standard solution, then dilute to the mark with dichloromethane. Repeat as necessary until you have the required number of standards.

7. Obtain chromatograms for each standard in turn, being sure that both the sample and internal standard (nonane) peaks are fully eluted. For best accuracy and precision, plot a calibration graph of peak area against concentration as you go, and redo any calibration solutions that do not lie close to the best-fit straight line through the data. (This may require simply re-running the solution, or making a fresh solution).

➔ Ideally, all injections should be performed in triplicate, but there is often insufficient time to allow this. For calibration solutions, a compromise is simply to ensure that there are

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

sufficient points in the calibration curve to compensate. All *sample* measurements should be at least duplicates, however, in order to determine precision.

8. While the standards are being run, prepare a solution of your unknown with nonane as internal standard: accurately weigh one drop of nonane into a clean, dry 10.00 mL volumetric flask, add 5.00 mL of the unknown mixture, and dilute to the mark with dichloromethane. Obtain *at least* two chromatograms of this mixture (preferably three.) Make sure that you each have a copy of the chromatogram for the unknown mixture, that you write your name and demo group number on each copy, and that you label all *relevant* peaks.

9. Check your calibration data using both regular and internal standard calibration procedures (see the Data Analysis section below). Repeat any measurements that appear suspect, re-making the solution if necessary.

## D. Finishing Up

10. Once you have finished your experiment, follow the procedure outlined above to shut down the GC system. Dispose of any solutions you no longer need (*check first!)*, along with any waste solvent, in the **appropriate waste container**. Any used Pasteur pipettes should be rinsed thoroughly first with acetone or methanol and then water before being disposed in the teal-coloured decontaminated glass bin. Clean all your glassware with acetone or methanol, using some alcohol on a KimWipe™ to remove any labels, and return it to the correct bench drawers.

➔ Please keep all the transfer and graduated pipettes in a different drawer to the rest of the glassware, in order to minimize accidental breakage

11. Check all areas where you have been working – balance, bench, and instrument – to make sure that they are clean and tidy, and that all chemicals have been returned to the correct shelves. When done, have your TA validate your lab notebooks before leaving.

## E. Data Analysis

You should compare two different calibration methods using the same data.

12. First, use the peak areas and concentrations for the analyte you chose to construct a conventional calibration graph. Calculate all the normal regression parameters ($r$, $s_{y/x}$, limits of detection and quantitation, *etc*.), and determine the concentration of the chosen compound in the unknown solution, together with the corresponding uncertainty expressed as both a standard deviation and a 95% confidence interval.

13. Second, use the internal standard to calculate normalised areas and concentrations, and repeat the regression analysis and concentration determination. Compare both the value and uncertainty you obtain by both methods using appropriate statistical tests. Internal standard calculations work in the following manner:

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

- For *each* concentration standard, calculate the ratio of the concentration of standard ($C_s$) to internal standard ($C_i$):

$$C'_s = \frac{C_s}{C_i}$$

- For *each* concentration standard, calculate the ratio of the area of the standard to internal standard peaks:

$$A'_s = \frac{A_s}{A_i}$$

- Plot a calibration graph of area ratio $A'_s$ *versus* concentration ratio $C'_s$
- For the sample, calculate the ratio of the peak area of the sample to internal standard:

$$A' = \frac{A}{A_i}$$

- Interpolate this value to determine the relative concentration of the sample using the concentration of internal standard *in that solution*, and derive the corresponding uncertainty.
- Use this interpolated value to calculate the concentration of the analyte in the original sample, taking into account the dilution factor preparing the sample with internal standard, and perform the appropriate error propagation for your final answer.

14. **Make sure you arrange to take the GC-MS tutorial during the week *before* the next lab period (experiment B2)**; you will be required to sign in to do so.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## 6. Experiment B2 Advanced Preparation: TurboMass Tutorial

The TurboMass Tutorial is an interactive, on-line tutorial that takes you through the basics of GC-MS. This tutorial is installed on a number of the computers in Analest, and is accessible through the appropriate desktop shortcut. You **must** complete this tutorial during the week prior to performing the GC-MS experiment. The key segments of this tutorial are:

1. Principles of Mass Spectrometry
2. Ion Sources: Gas Phase Sample Introduction and Electron Impact (EI)
3. Analyzers and Detectors: Quadrupole Mass Filter and Stability Diagram
4. Applications and Hyphenated Techniques: Qualitative, quantitative, GC-MS

## 7. Experiment B2: Qualitative Analysis by GC-MS

In this experiment, you will use a similar sample to that for experiment B1, but take advantage of the GC-MS combination for the identification of the components in the unknown. In addition, you will explore the effect of temperature on retention and resolution, and the use of selected ion detection modes.

**IMPORTANT PRECAUTIONS:**

- ➔ **Do NOT wash your glassware with water or use compressed air to dry it**. When working with non-aqueous solvents such as dichloromethane or ethyl acetate, you should avoid introducing any water into the glassware. Rinse glassware *with the solvent you are going to use* instead. If this is insufficient to clean an item properly, use acetone, and then rinse with the solvent afterward.
- ➔ **Limit exposure to solvent fumes**. Make sure you stopper all flasks when transporting or weighing, and perform all rinsing and dilution operations in the fume hood. Do ***not*** leave open beakers of solvent on the bench.
- ➔ **Dispose of all organic waste properly**. Any solution containing dichloromethane or bromobenzene ***must*** be emptied into the halogenated waste container. Other hydrophobic organic waste (ethyl acetate, alkanes, benzene, *etc*.) can go in the large red waste can. Any water-soluble organics (*e.g*. alcohols) should go in the aqueous organic waste for the HPLC experiments in the other fume hood.

### A. Instrument Setup and Qualitative Analysis of an Unknown

1. Start up the GC-MS as outlined in section 4.5, and allow the instrument to equilibrate. For the first part of this experiment, you should use the CHM317 methods for the MS and GC, along with the default tune file. The following lists typical conditions used for this experiment – note that the column temperature may be varied slightly by lab staff in order to compensate for aging of the column:

| | |
|---|---|
| GC Method File: | CHM317 |
| Column: | DB5 or equivalent (5% phenyl phase) |
| Column dimensions: | 30 m × 0.23 (or 0.32) mm × 0.25 μm |

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

| | |
|---|---|
| Carrier Gas: | Hydrogen |
| Split Ratio: | 25:1 |
| Column Pressure: | 4 psi |
| Oven Temperature: | 110 °C (isothermal) |
| Injector Temperature: | 250 °C |
| Injection Volume: | 1 μL |
| Detector: | Quadrupole MS |

The CHM317 method for the MS involves scanning the *m*/*z* range from 40 to 250 for the duration of the chromatographic run (set for a maximum of 20 minutes). When the GC-MS is ready, use a 2.0 or 5.0 syringe to inject 1.0 μL of the provided GC-MS unknown solution and record the chromatogram. Note that you do *not* need to dilute the sample.

**Be very careful not to bend the needle, or pull the plunger out of the syringe!**

When the chromatogram has been collected, use the features of the TurboMass software outlined in section 4.6 to identify each peak based on its mass spectrum. Make particular note of the number and identity of possible compounds suggested by the structure search software for each peak. (You do not need to list *all* the suggestions provided by the software, but you should note any potential matches with a quality factor of 800 or more.)

2. Compare the elution order (sequence in which the different compounds came off the column) and retention times with those obtained for the same sample mixture in experiment B, remembering that the solvent peak should *not* be observed in GC-MS due to the solvent delay on the MS. The absolute retention times will almost certainly be different, since you are using a different column and instrument, but the relative retention times should be similar. Make sure that you each have a labelled copy of the chromatogram for your reports. You may also wish to print out specimen copies of the mass spectra obtained for each sample component within your chromatogram.
3. Obtain peak areas for each peak in the sample chromatogram, and examine the peak shapes carefully. In particular, note how many data points in the total ion chromatogram (TIC) were obtained for each peak. Also note any peaks that are not fully resolved.
4. Repeat the injection of the unknown sample two times, so that you can obtain peak areas for each compound in triplicate. Calculated the %RSD for each peak.

## B. The Use of Selected Ion Measurement Modes

The default display mode when recording full-scan mass spectral data is to show the total ion count (TIC), which shows the sum of all ion fragments detected (as counts/second) over the selected range for each scan. One can also view the data for selected single ion fragments, which can be useful for determining if chromatographic peaks contain unresolved components, or for calculating peak areas for overlapping peaks.

Data can also be collected for specific ion fragments *instead of* obtaining the full scan; this can be advantageous for quantitative analysis. In this section, you will compare full scan and selected ion modes in GC-MS.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

5. Open one of your sample chromatograms in the viewer window; this should display the TIC view of the data. Review the mass spectra of two close or overlapping peaks within your chromatogram, and note the *m/z* values for two ion fragments; at least one (and ideally both) of these should be *unique* to one of the two compounds being separated.

Chromatograph viewer toolbar buttons related to viewing and integrating peaks.

Now click on the first button in the group having chromatographic peaks in their icons (see above left); the resulting dialog box will allow you to add additional views for specific ion fragments by entering the *m/z* value in the **Channel** field. Add views for the ion fragments you have selected, so that you can compare the TIC and selected ion chromatograms for the peaks of interest. (If the views are overlaid rather than stacked, switch the view display mode using either the menu or toolbar.) Use the **Integrate Peaks** button (second from left) to compare peak areas between the TIC and selected ion chromatograms.

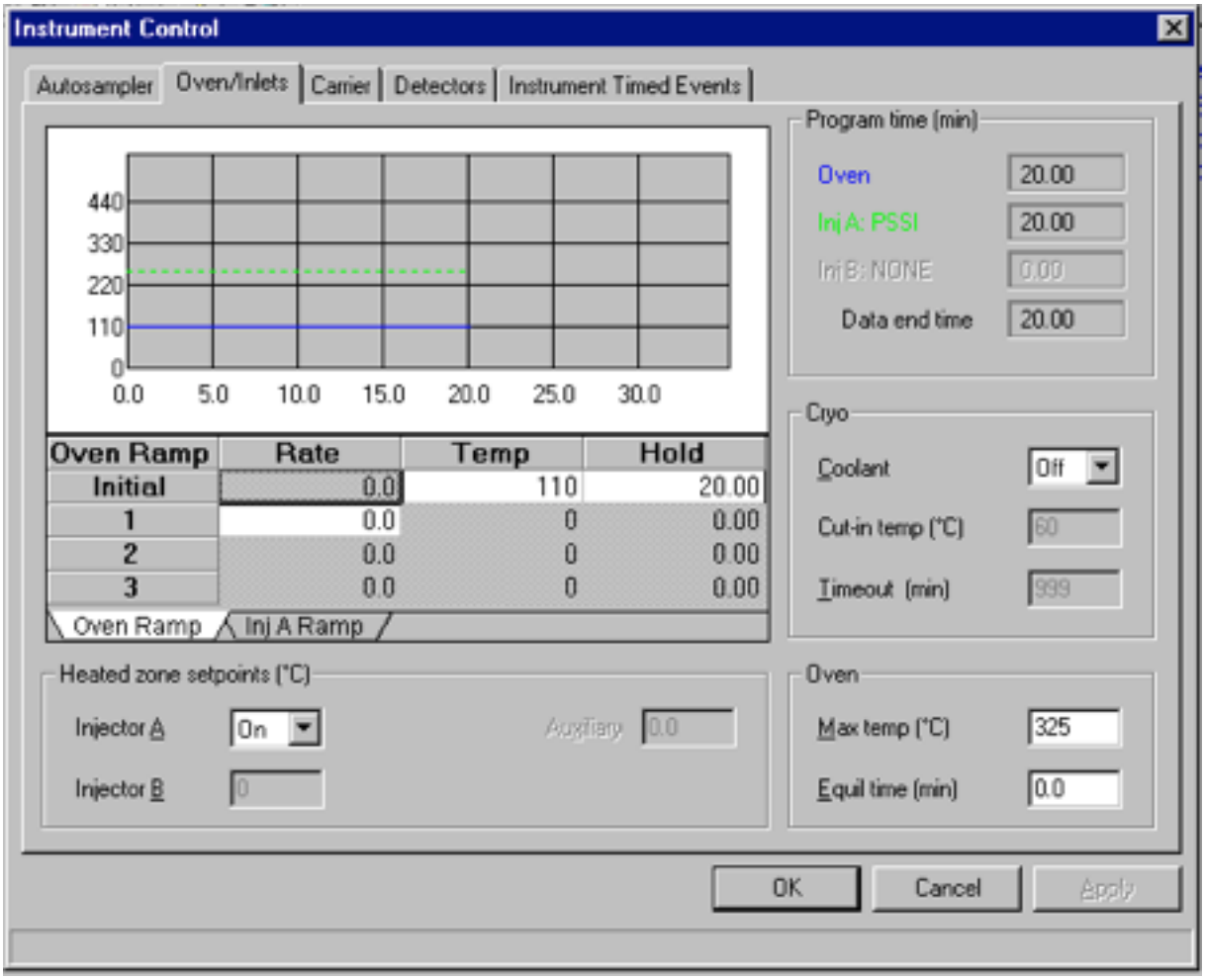
6. In the main TurboMass window, double-click on the MS method entry in the first unused row (add rows to the sample list if necessary), and select the **CHM317SIR** method from the drop down list. This method defines which *m/z* fragments to record during specific time periods corresponding to the expected elution times for each component in the unknown sample. You can view these conditions by right–clicking on the **CHM317SIR** entry and selecting **Open** from the pop-up menu. Once you have noted the conditions, select the row, click on the **Run** button and, once both the MS and GC are ready, inject your sample.

7. View the resulting chromatogram as usual, and use the same button as in step 5 to add the selected ion views: in the dialog box, click on the "Add all" button. Compare the number of points per peak and the area integration values with your previous full scan runs. Once you have done this, perform an additional two replicate injections, and calculate %RSD values for the peak areas obtained in this way.

## C. The Effect of Temperature on Retention Time

8. You will now examine the effect of varying the column oven temperature on retention time. Go back to the main TurboMass window and, for the next available unused row, set the MS method back to **CHM317** and the relevant GC method from the set **CHM31790** to **CHM317120** (corresponding to temperatures 90–120 °C); set these up in order from lowest to highest temperature. Open the GC method, and select **Ovens/Inlets...** from the **Instrument Control→Options** menu and confirm that the initial oven temperature is correct (*e.g.* 90°C – see screen–shot on next page), click **OK**, and save the updated method if asked to do so. Note that the run times for the lower temperatures should be longer than for your initial experiments, as retention times will also be longer.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry




The temperature program window within the GC Method editor

9. You can now select this row, and run a further injection of your unknown sample. Note that it will take longer for the GC to become ready, as the oven will need to cool down from the initial temperature; there will also be a short equilibration time once the new oven temperature has been reached. When both the MS and GC are ready, inject your sample as usual. Make sure that you allow *all* the sample components to completely elute before stopping the run, note the retention times for all the peaks, and print out a copy of the chromatogram.

10. Go to the next unused row in the main TurboMass window, making sure it is using the same MS and GC methods as step 8. Open the GC method as you did in step 8, but increase the initial oven temperature to 100°C. Following the same procedure as before, obtain a chromatogram at the new column temperature, note the retention times for all the peaks, and print out a copy of the chromatogram.

11. Repeat this procedure to obtain chromatograms at 110°C, 115°C, and 120°C. Be sure to note down if any peaks start to overlap, become unresolved, or move into the solvent delay window. Use the mass spectrum to confirm the identity of the peaks; you can use selected ion channels to clarify the retention times for overlapping peaks (see step 6).

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## D. Finishing Up

12. Once you have finished your experiment, follow the procedure outlined above to shut down the GC system. Dispose of any solutions you no longer need (*check first!)*, along with any waste solvent, in the **appropriate waste container**. Any used Pasteur pipettes should be rinsed thoroughly first with acetone or methanol and then water before being disposed in the teal-coloured decontaminated glass bin. Clean all your glassware with acetone or methanol, using some alcohol on a KimWipe™ to remove any labels, and return it to the correct bench drawers.

➔ Please keep all the transfer and graduated pipettes in a different drawer to the rest of the glassware, in order to minimize accidental breakage

13. Check all areas where you have been working – balance, bench, and instrument – to make sure that they are clean and tidy, and that all chemicals have been returned to the correct shelves. When done, have your TA validate your lab notebooks before leaving.

## E. Data Analysis

14. Compare the repeatability of peak area determinations obtained using full scan and selected ion recording; which has the better precision, and is the difference statistically significant? What factors might alter your findings? What implications does this have for quantitative analysis using GC-MS versus GC-FID?

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## 8. Experiment B3: Determination of BHA and BHT in Foods

This experiment is based in part on Yang *et al*, *Food Res. Int.*, **2002**, *35*, 627-633. Butylated hydroxy anisole (BHA) and butylated hydroxy toluene (BHT) are common antioxidants added to various foods in order to reduce the onset of rancidity through the breakdown of unsaturated fats. The Food and Drugs Act and accompanying Food and Drug Regulations (Health Canada, 2005) limit the maximum amount of these and similar compounds to 0.02% by weight of the fat content in fats and oils, and 0.01% by weight of the fat content in margarine. Not all margarines, fats, and oils contain either BHT or BHA, but some do.

Because of the high lipid content of fats and oils, methods for BHA and BHT require extensive extraction and clean-up procedures before samples can be injected into a chromatograph. This experiment uses a pre-column trap lined with glass wool and a megabore capillary column to enable simple solvent extracts to be injected directly into the GC for analysis. An internal standard (TBP) is used for accurate quantitative analysis. You may bring in your own oil sample for analysis if you prefer, although you should check the contents label carefully as not all oils contain these compounds. Otherwise, a sample will be provided for you.

After performing this experiment, you should:

- Understand the difference between isothermal and temperature gradient elution methods in gas chromatography
- Understand the difference between megabore and regular capillary columns
- Know how to perform internal standard calculations in chromatography
- Be aware of sample matrix effects in sample injection for gas chromatography

**Chemicals:**

- BHT: 3,5–di–*tert*-butyl–4–hydroxytoluene
- BHA: a mixture of 2– and 3–*tert*-butyl–4–hydroxyanisole
- TBP: 2,6– di–*tert*-butyl–phenol
- Ethyl acetate
- Olive oil

**IMPORTANT PRECAUTIONS:**

➔ **Do NOT wash your glassware with water or use compressed air to dry it**. When working with non-aqueous solvents such as dichloromethane or ethyl acetate, you should avoid introducing any water into the glassware. Rinse glassware *with the solvent you are going to use* instead. If this is insufficient to clean an item properly, use acetone, and then rinse with the solvent afterward.

➔ **Limit exposure to solvent fumes**. Make sure you stopper all flasks when transporting or weighing, and perform all rinsing and dilution operations in the fume hood.

➔ **Dispose of all organic waste properly**. Any solution containing dichloromethane or bromobenzene ***must*** be emptied into the halogenated waste container. Other hydrophobic organic waste (ethyl acetate, alkanes, benzenes, *etc*.) can go in the large red waste can.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

Any water-soluble organics can go in the aqueous organic waste for the HPLC experiments.

1. Start up the assigned GC as outlined in section 4 above, and allow the instrument to equilibrate. The instrument conditions for the separation are as follows:

   Method: BHT2.mth
   Carrier pressure: 3.0 psi
   Injector oven: 250 °C
   Detector oven: 290 °C
   Column oven: temperature program (thermal gradient)
   Initial 150 °C; hold 1 min
   Ramp @ 7.0 °C/min to 190 °C; hold 2 min
   Ramp @ 45 °C/min to 300 °C; hold 1 min

   Column: DB-5 Megabore column 0.53 mm × 30.0 m, 1.5 μm film thickness

2. **Stock Solutions:** While the instrument is equilibrating, accurately prepare 25.00 mL of a solution containing about 2.0 mg/mL of BHA in ethyl acetate: weigh the analyte by difference using an analytical balance. Similarly prepare separate solutions containing 2.0 mg/mL of BHT and 1.0 mg/mL of TBP, respectively, in ethyl acetate. Calculate the actual concentrations in each of your solutions, to the correct number of significant figures.

➔ **Caution:** ethyl acetate is quite volatile, so you should dispense the solvent in a fume hood and keep the flasks tightly stoppered between operations to reduce evaporation of the solvent.

3. Determine the retention time for each compound by injecting 0.30 μL of each of standard in turn; you may stop each run once both the solvent and the standard have been eluted from the column. While these are running, start preparing your samples and calibration standards as follows.

**Caution:** The 0.5 and 1.0 μL syringes use a very fine wire connected to the plunger to control the liquid volume within the needle itself. When rinsing and filling the syringe, **DO NOT** pull the plunger all the way out of the syringe. If this *should* happen, notify the instructor or lab manager immediately; do *not* try to push the wire and plunger back inside the syringe!

4. **Calibration Standards:** Prepare a series of mixed standard solutions using the stock solutions prepared in step 2; make these calibration standards in 25.00 mL volumetric flasks. Each calibration standard should be made using 2.00 mL of the TBP stock solution as internal standard, together with equal volumes (2.00, 5.00, and 10.00 mL) of the BHT and BHA stock solutions *i.e.* each calibration solution should contain both BHT and BHA. Dilute the contents of each flask to the mark using ethyl acetate.

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

➔ **Caution:** ethyl acetate is quite volatile; dispense your solutions in a fume hood, and keep the flasks tightly stoppered between operations to reduce evaporation of the solvent.

5. **Oil sample preparation:** Accurately weigh out between 30–60 mg of the oil sample into a 1.00 mL volumetric flask (ask your demonstrator for this; be careful – they are very expensive!) Accurately add 0.50 mL of the stock TBP solution as internal standard using a glass transfer or Mohr pipette. Carefully fill the flask to the mark with ethyl acetate, stopper, and mix well be repeatedly inverting the flask.

6. As your calibration solutions are prepared, run each in turn by injecting 0.10 μL into the GC. For the calibration standards *only* you may stop the run once the solvent and all three peaks have eluted from the column. Remember to keep the flasks stoppered until you are ready to load the syringe; and that there will be a short delay for the column oven to return to the original temperature before you can start the next run.

7. Once you have successfully run all the standards, run the oil sample by injecting 0.10 μL into the GC. You *must* let the temperature program run to the end (~ 9 – 10 min) when chromatographing the sample in order to ensure that the high molecular weight components are properly removed before the next run. Perform replicate injections as time allows; ideally, you want at least three sample chromatograms.

8. While your sample is running, construct and check your calibration curve by plotting the area for each component relative to that of the internal standard (A') against concentration; be sure to calculate the actual concentrations of the compounds and internal standard from the dispensed weight, taking all dilution factors into account. A full description of the internal standard calibration method can be found in experiment B1, step 11.

9. After measuring all your samples and standards, ask your TA or lab instructor for assistance to modify the GC method to run isothermally at a temperature of 170 °C. Obtain an isothermal chromatogram for your most concentrated standard at this temperature, and note the new retention times for each of the compounds in your mixture.

10. Once you have finished your experiments and are satisfied with your data, follow the shutdown procedure outlined in section 4.4. Rinse the syringe with some ethyl acetate. Dispose of all your solutions and any waste solvent in the **appropriate waste container**. Any used Pasteur pipettes should be rinsed thoroughly first with acetone and then water before being disposed in the teal-coloured decontaminated glass bin. Clean all your glassware with acetone, using some alcohol on a KimWipe™ to remove any labels, and return it to the correct bench drawers.

➔ Please keep all the transfer and graduated pipettes, as well as the 1 mL volumetric flask, in a different drawer to the rest of the glassware in order to minimize accidental breakage

10. Check all areas where you have been working – balance, bench, and instrument – to make sure that they are clean and tidy, and that all chemicals have been returned to the correct shelves. When done, have your TA validate your lab notebooks before leaving.

11. **Data Analysis:** Construct calibration curves and determine the concentration of each analyte in the olive oil sample using both regular and internal standard calibration methods, as set out at the end of experiment B1. Don't forget to calculate the uncertainties in the

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

concentrations, and compare the results obtained using the two different calibration methods – which was the more precise method?

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry

## 9. Elements for Report Discussion

Your discussion for this report should address both the specific analysis results and the general lessons to be learned about GC from these experiments. In particular, you should discuss the factors governing elution order in GC and the factors governing the choice of column for any given sample separation. In doing so, make sure you compare and contrast the different mobile and stationary phases used in the various experiments, along with the properties of the sample components being separated. Also consider the following questions in respect to the specific experiments you have performed:

1. In GC-MS, it is not uncommon to find that peaks eluting with similar retention times have similar mass spectra, especially for samples containing trace quantities of a range of compounds and/or high molecular-weight species. Why is this, and what implications does it have for the identification of peaks in GC-MS chromatograms?

2. What steps can you take to improve the accuracy and precision of quantitative analysis by GC or GC-MS? What about sensitivity and limits of detection – how could you improve these for any given sample? How can the features of the MS detector be exploited to achieve this?

3. GC-MS (and its variants) is sometimes referred to as the 'gold standard' in forensic science – what does this mean, and why is this the case?

4. Compare and contrast the use of: (a) megabore with standard capillary columns; (b) isothermal and gradient elution techniques; and (c) split and splitless injection methods. If you did not have a trap and megabore capillary column for experiment B3, what additional steps would you have to take before your could inject a sample into the chromatograph?

© 2003-2018 Dr. D. C. Stone/University of Toronto Chemistry