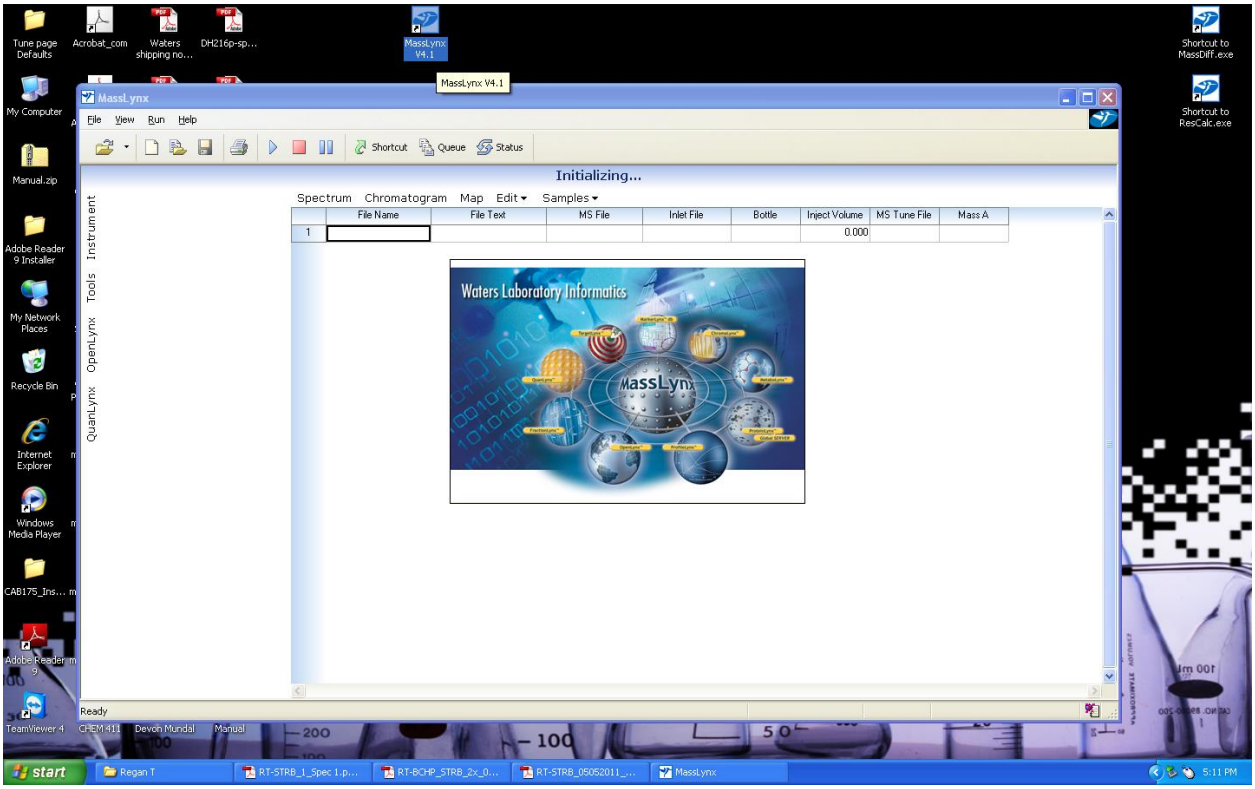
Gas Chromatography–Time of Flight Mass Spectrometry (GC-TOF MS)

Northwestern University IMSERC Facilities Manual
Version 1.0, A. Avestro

Gas Chromatography–Time of Flight Mass Spectrometry (GC-TOF MS) is an electron impact ionization technique that is well suited for small organic molecules with molecular weights up to ~1000 Da. Before running a GC-TOF sample, please ask yourself the following regarding your sample and sample preparation:

- Is my sample soluble in a common organic solvent (e.g. methylene chloride, methanol, chloroform, ethyl acetate, isopropanol, acetonitrile, etc.)?
- If any particulates or solid is present, has my sample by filtered?
- Is my sample capable of being volatized?
- Is my sample's molecular weight within the range of the instruments capabilities?

[1] Starting up the Software / Sample Setup



1. Double click on the GC-TOF software, MassLynx V4.1, located on the desktop. ***File >> Open Project…***

If the following message appears: "When changing to a new Project some services are automatically closed down. Continue?" – Click Yes.

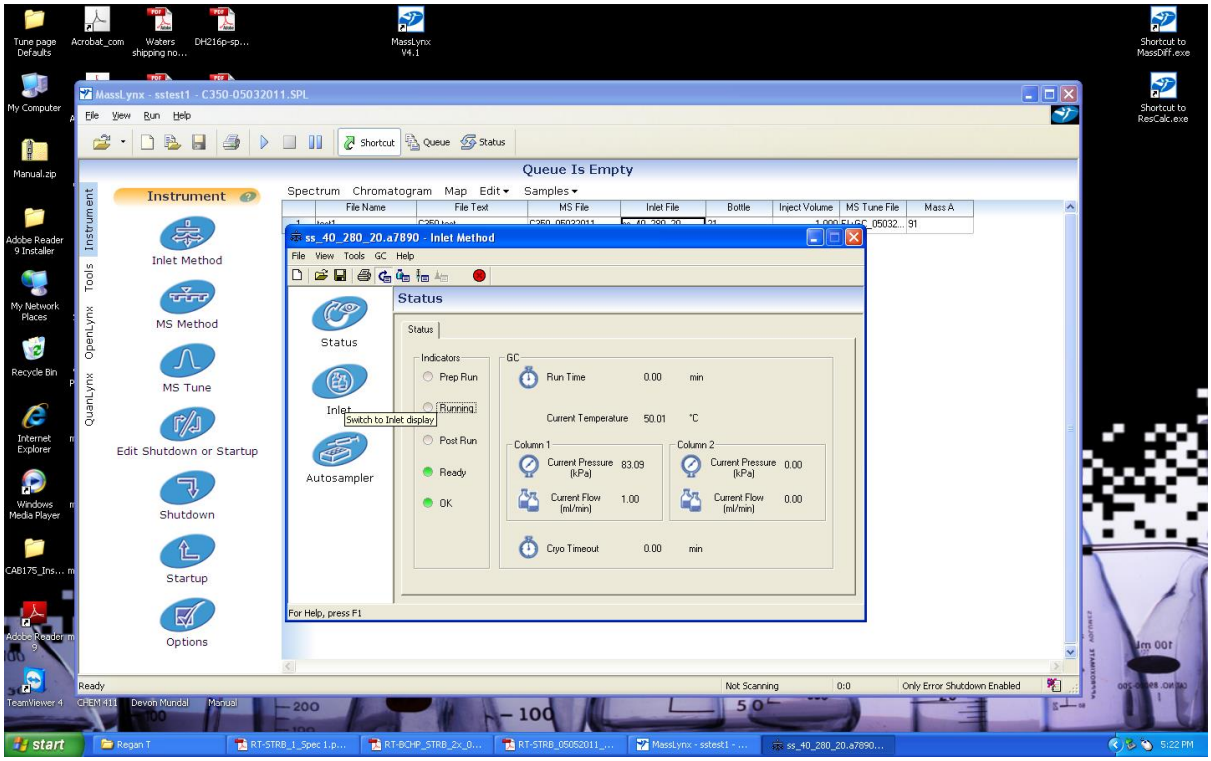
2. **PROJECT:** Select a project file (.PRO). The project file (.PRO) you select contains the project parameters that will be applied to your data acquisition.

If this the first time developing a Project Use File >>>Project Wizard

- Name the Project , and Store in **F:** drive.
- Choose option: Create using existing Project as template, choose Browse and use F:data/Template/Template.pro as your template.

Edit the *File Name* (must be unique as this will be the name that your data is saved under). *File Text* is optional space for you to enter additional identifying information about your sample

[2] Managing the Inlet File (GC conditions)



3. Open a pre-exisiting Inlet File — Right-click under ***Inlet File >> Browse***. Open the Inlet Method window to edit inlet parameters — Right-click under ***Inlet File >> Edit***.

The *Inlet File* contains acquisition conditions for the GC component of the instrument. GC-TOF *Inlet File* naming system: "(Start T)_(End T)_(Ramp Rate)"

Click on either "**Inlet**" or "**Autosampler**" on the left-hand side of the Inlet Method window (both lead to the same Inlet Setup table).

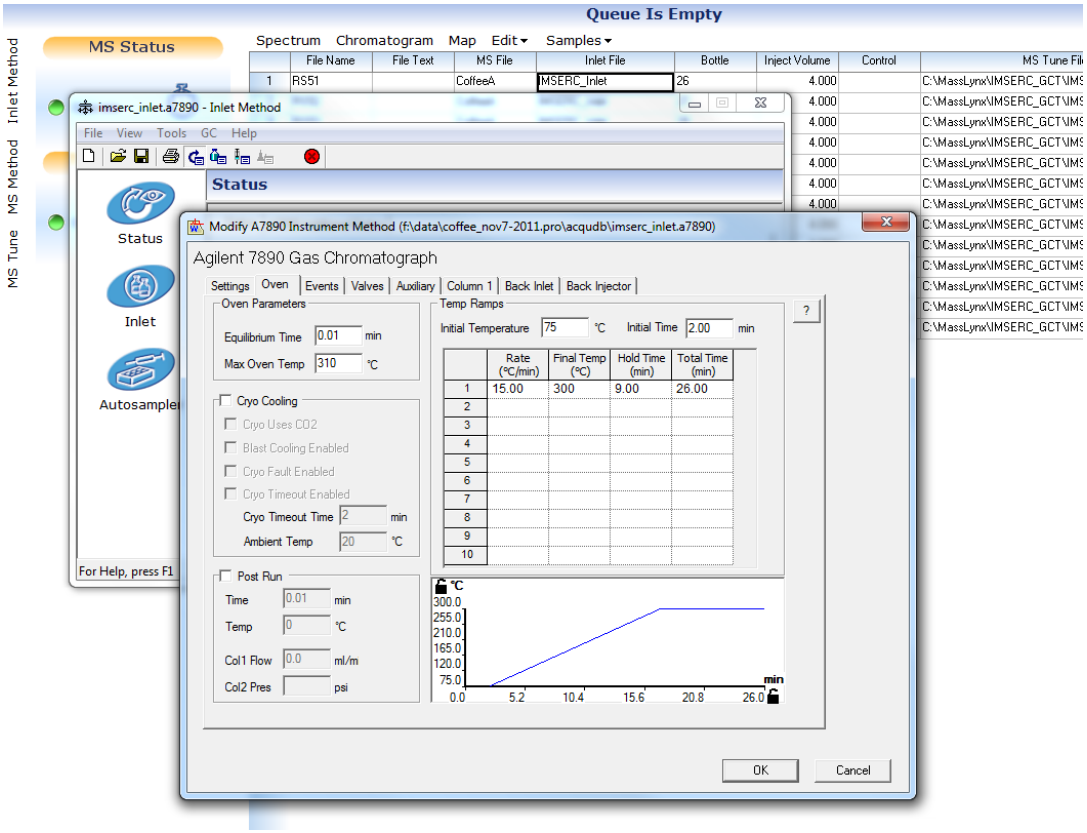
Settings

- *Column 1 Inlet* = Back Outlet = None
- *Injector = Back* *Type= 7683B*

Oven

- Set up a temperature program for your sample here. If using a "wax column," please be aware of the maximum operating temperature ~220 K. Please set up your temperature program accordingly to preserve the quality and lifetime of GC-TOF equipment.

After setting up your temperature program, take note of the total oven time (e.g. 22.5 minutes below), as this will come in handy during the *MS File* setup.



Back Injector

- *Viscosity delay* = 1.0 second
- *Plunger mode* = Slow

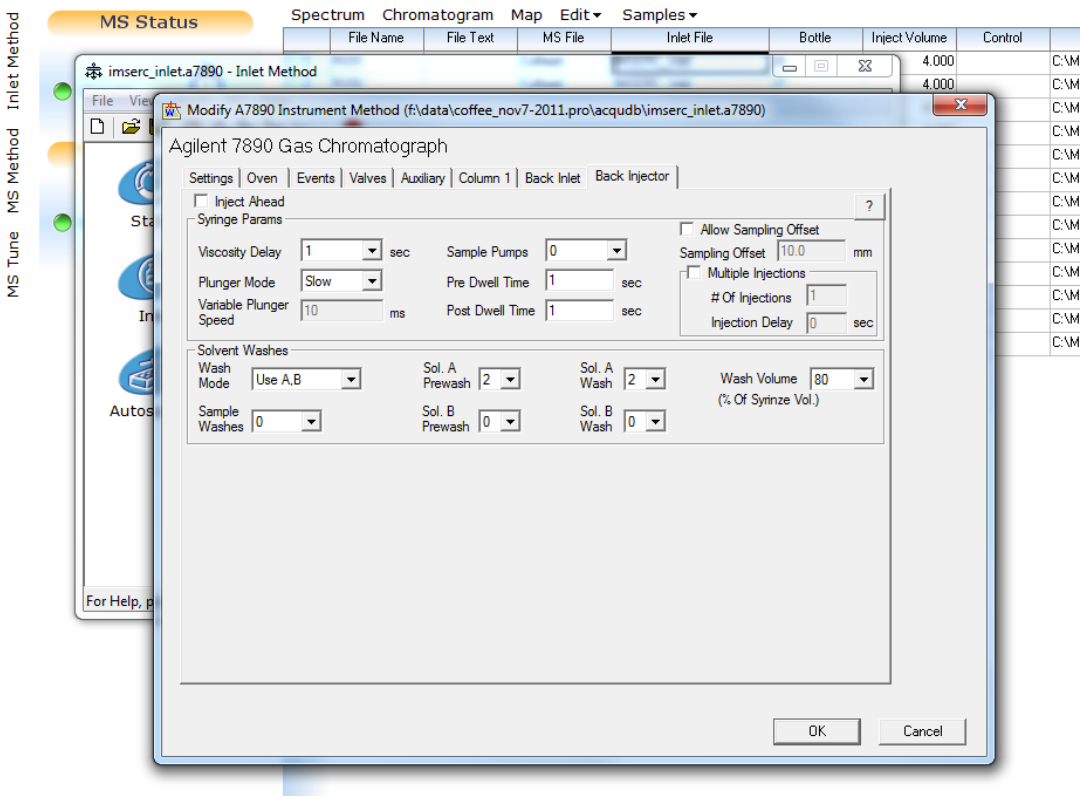
This will ensure that the plunger does not undergo any jerky, possibly harmful movements and injects at a steady speed.

- *Pre-dwell / Post-dwell Time* = 1.0 second

This setting will inform the robot to keep the needle inside the inlet for X amount of seconds before and after injection, allowing the needle temperature to equilibrate with the temperature of the injection chamber.

- *Sample wash* = optional, to prime syringe with sample pre-injection
- *Pre-wash* = 2 times for both Solvent A and Solvent B
- *Post-wash* = 1or 2 times for both Solvent A and Solvent B

Solvent A and Solvent B are located on the corral. The number selected from the drop-down menu indicates the number of washes for that solvent



Column 1

- *Length (m)* = 30.0 meters
- *Internal diameter (u)* = 250
- *Film thickness (u)* = 0.25
- *Carrier gas* = Helium (He)
- *Initial flow* = 1.0 mL/min
-

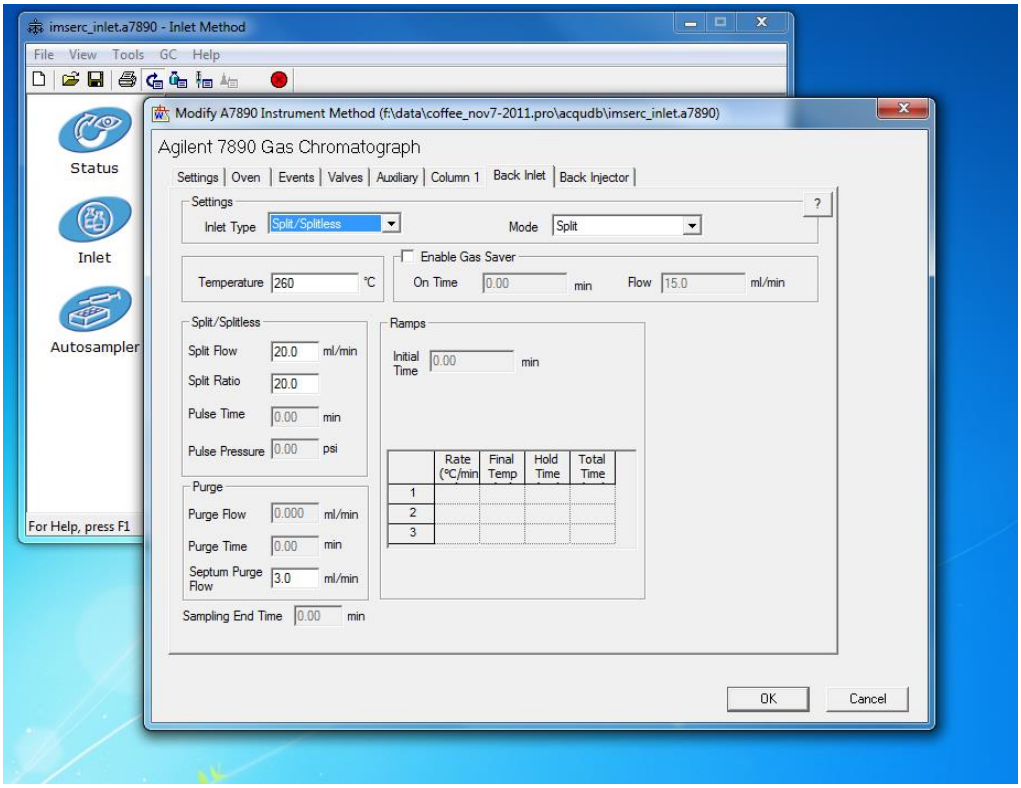
Back Inlet

- *Injection port type* = Split/Splitless
- *Mode* = Split
- *Septum purge flow* = 3.0 mL/min
- *Split ratio* = 100.0 (can go down to 5:1)

A split ratio of 100.0 (i.e. 100:1) means that of the sample amount injected, 100 parts will go to waste and 1 part will go to forward to the detector.

Click OK when finished adjusting Inlet File parameters. Back at the Inlet Method window, ***File >> Save As…*** to assign a new file name to your modified Inlet File. Close the Inlet Method window.

Suggested naming system mentioned above: "(Start T)_(End T)_(Ramp Rate)." Alternatively, you may choose to include additional letters or initials at the beginning to denote the file for future use, e.g., "C350_80_250_10."



[3] Managing the MS File

5. Open a pre-exisiting MS File —- Right-click under ***MS File >> Browse***.

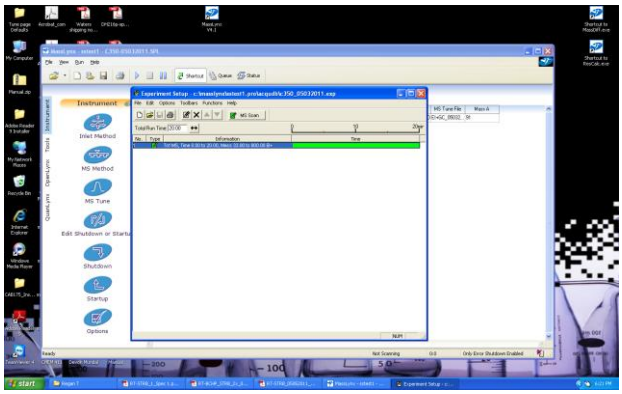
Again you can use the template file (F: Data/IMSERC.PRO/Acqudb/ IMSERC_MSfile)

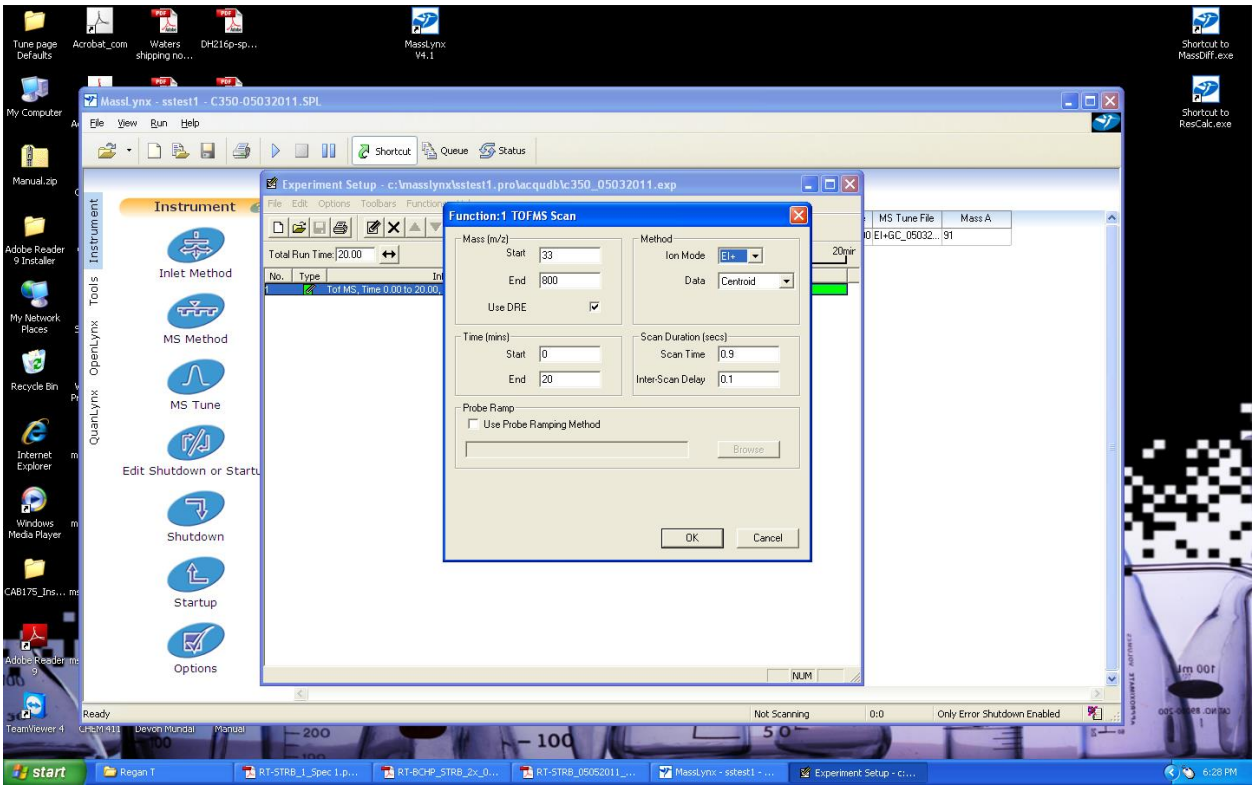
Open the MS File window to edit inlet parameters —- Right-click under ***MS File >> Edit***.

6. Double-click on the desired MS File to open the parameters setup window.

- *Mass (m/z)* = 33 – 800 m/z ("Use DRE" is checked.)
- *Ion Mode* = EI+ (electron impact ionization)
- *Data* = Centroid
- *Scan Duration —- Scan Time* = 0.9 sec, *Scan Interval* = 0.1 sec
- *Time (s)* = 0 – _(enter time from Inlet File setup here)_

Click OK when finished adjusting MS File parameters.



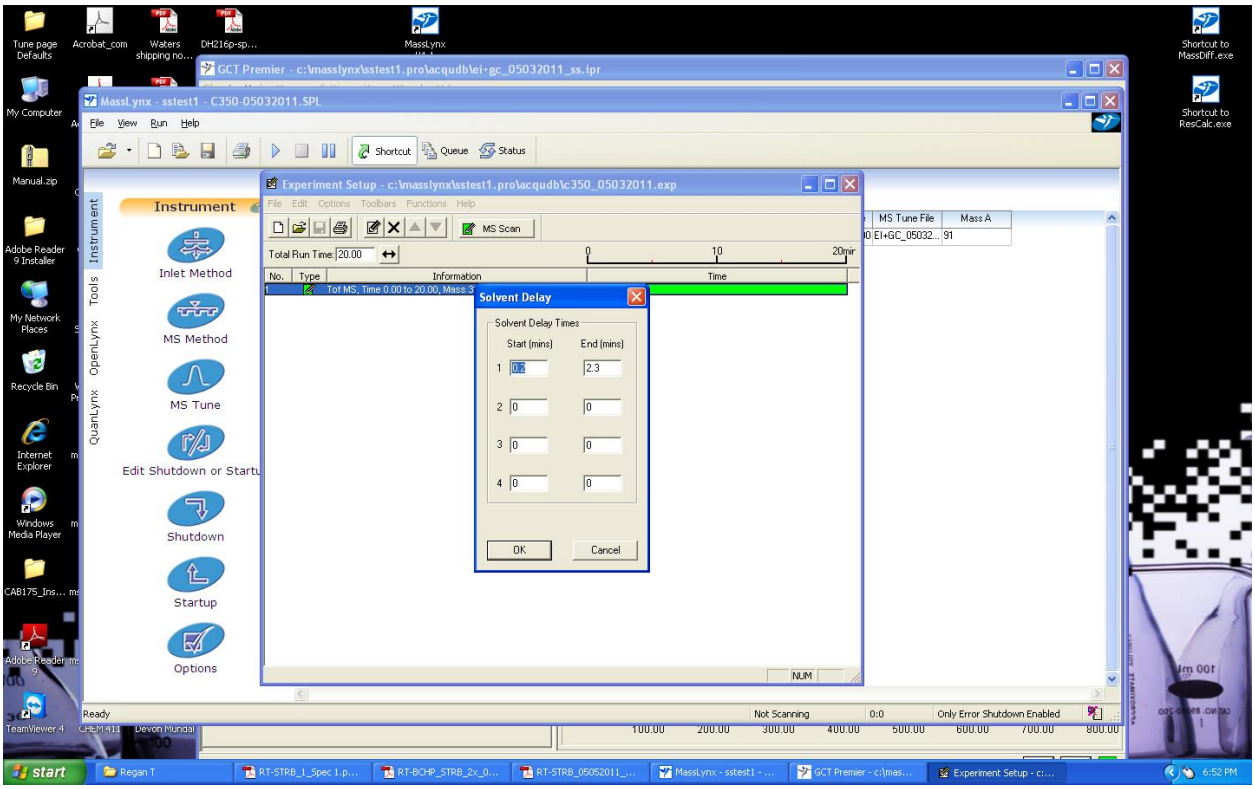


7. Edit the solvent delay time by going to ***Options >> Solvent Delay…***

- *Solvent delay* = 0.2 – 2.2 minutes (Do not start solvent delay at 0.0 min)

It is advisable to purposefully start the solvent delay a little later to ensure the filament is really operating before committing yourself to a complete data acquisition (you'll save a lot of time!)

Close the solvent delay window and go to ***File >> Save As…*** to assign a new file name to your modified MS File. Close the MS File edit page when done.



8. Finish filling in the missing entries at the MassLynx frontpage — Select the *most recent* MS Tune File. Keep the injection volume = 1.0 (microliters). You may optionally enter a target mass for your sample. Finally, enter the "Bottle" number for your sample vial located on the corral.

[5] Running a GC-TOF Sample

12. At this point, you may wish to add additional samples to your sequence. Doing so now (as opposed to in the beginning) will allow your newly modified MS and Inlet parameters to copy over to the next sample. Right-click >> ***Insert*** to add a new row.

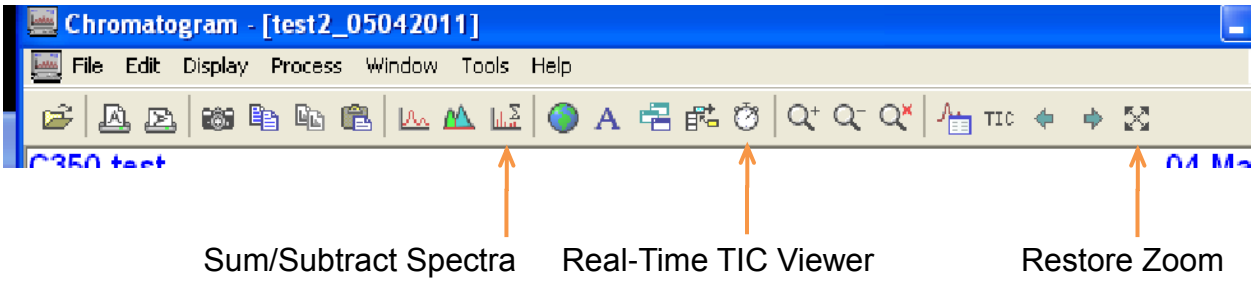
13. Double check at this point that the correct Inlet File is loaded by going to ***Settings >> Load…*** The "Remote" light should blink yellow if there is proper communication between the computer and instrument.

14. Go to ***Run >> Start*** to begin a sample sequence. The auto-injector will activate once the proper instrument temperature has been reached (visible on the GC console).

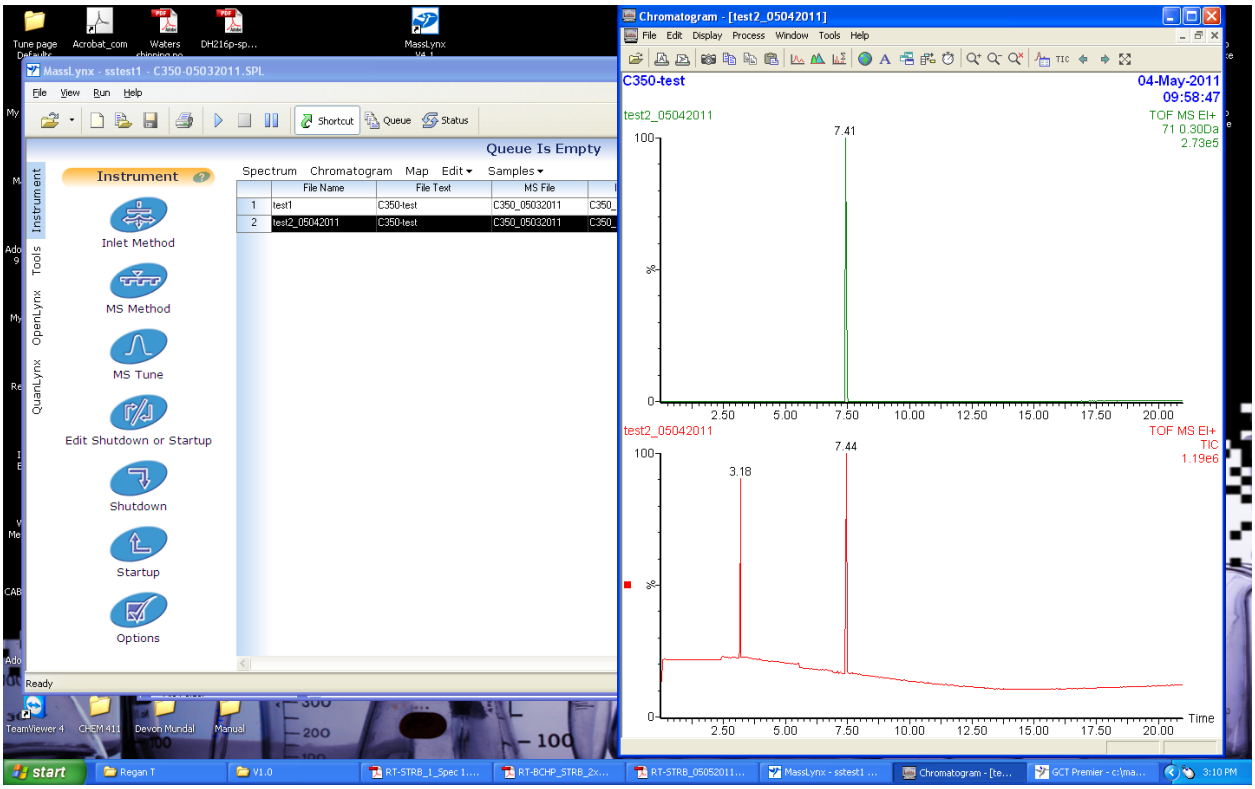
[6] Obtaining a Mass Spectrum

Manipulating the TIC and MS

- Left-click + drag = Zoom In
- Right-click = Single Scan Mass Spectrum
- Right-click + drag = Average Mass Spectrum



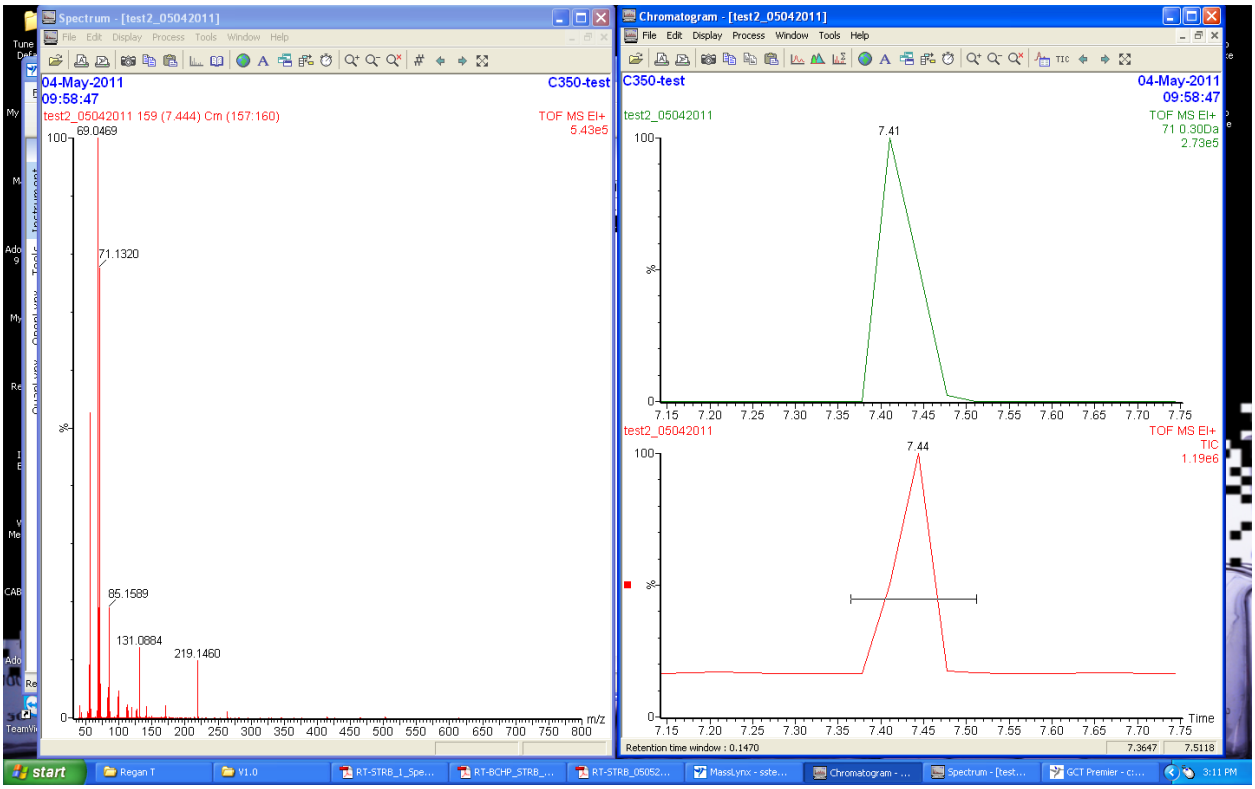
Sum/Subtract Spectra Real-Time TIC Viewer Restore Zoom



15. On the MassLynx frontpage, highlight the sample row of interest and click on "Chromatogram" above the sample table to open a snapshot (mid-run) or the complete total ion chromatogram (TIC).

During the middle of a run, the snapshot can be converted to a real-time chromatogram by clicking the Real-time TIC Viewer icon 

If a target mass for the was entered in the sample table ("Mass A"), then the window will show two spectra — the extracted ion chromatogram (top, **green**) and the TIC (bottom, **red**). This is denoted in the upper right-hand corner of each spectrum.

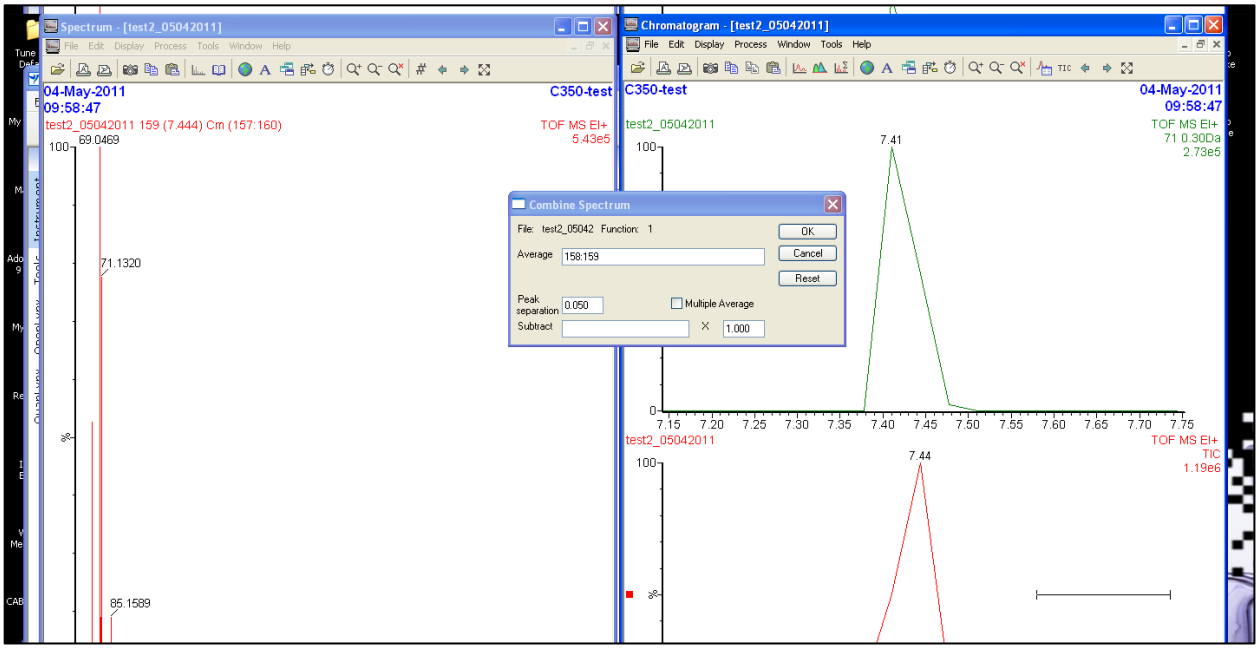


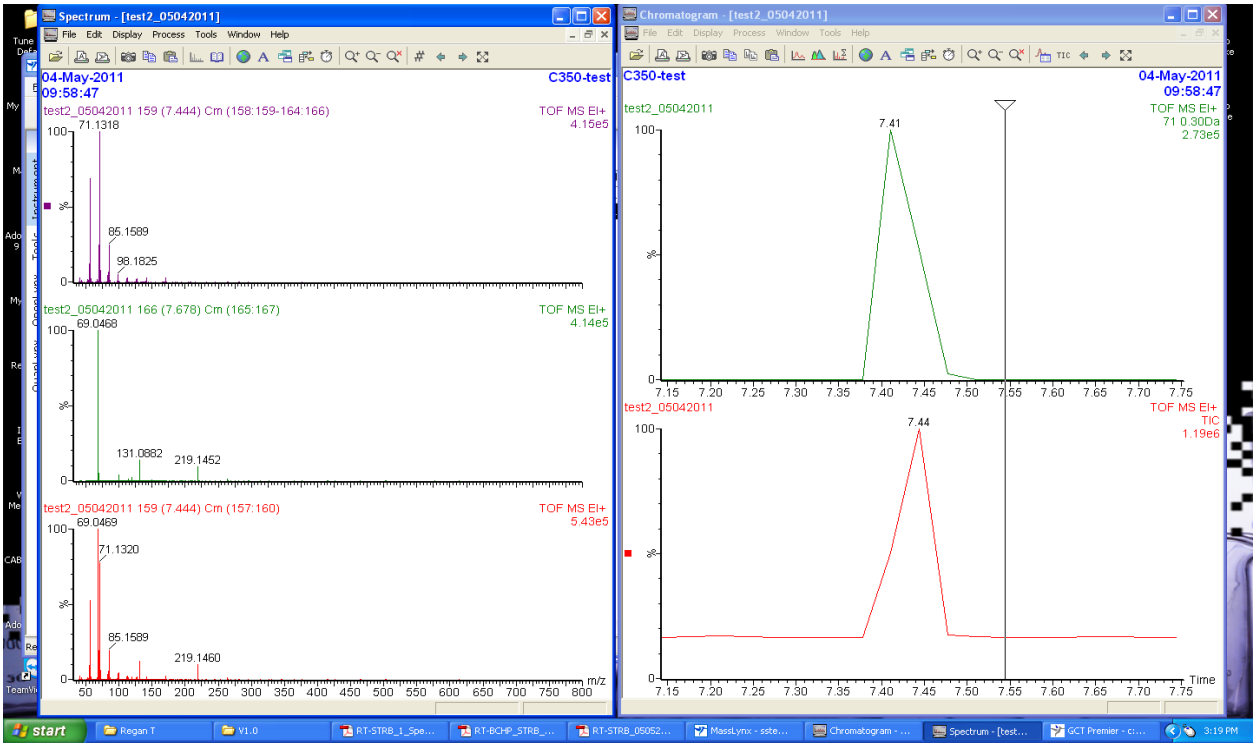
16. Zoom in on the peak of interest (left-click + drag) and pull up the average mass spectrum (right-click + drag across). You may also view the background signal by right-clicking + dragging across any other portion of the TIC.

Subtract the Background

- Click on the Sum/Subtract Spectra icon 
- Right-click + drag across the peak of interest on the TIC
- Right-click + drag across the background to eliminate the reference signals from the mass spectrum
- Click OK to view the background-corrected mass spectrum (top)

A mass spectrum of both the background (middle, **green**) and the background-corrected spectrum (top, **purple**) will appear stacked above the original spectrum (bottom, **red**) in the MS window.


Spectrum - [test2_05042011]
Chromatogram - [test2_05042011]
04-May-2011
09:58:47
C350-test
test2_05042011 159 (7.444) Cm (157:160)
TOF MS EI+
5.43e5
69.0469
71.1320
85.1589
Combine Spectrum
File: test2_05042
Function: 1
Average
158:159
OK
Cancel
Reset
Peak separation
0.050
Multiple Average
Subtract
1.000
7.41
7.44
TOF MS EI+
TIC
1.19e6


Spectrum - [test2_05042011]
Chromatogram - [test2_05042011]
04-May-2011
09:58:47
C350-test
test2_05042011 159 (7.444) Cm (158:159-164:166)
TOF MS EI+
4.15e5
71.1318
85.1589
98.1825
test2_05042011 166 (7.678) Cm (165:167)
4.14e5
69.0468
131.0882
219.1452
test2_05042011 159 (7.444) Cm (157:160)
5.43e5
69.0469
71.1320
85.1589
219.1460
m/z
7.41
7.44
TOF MS EI+
TIC
1.19e6
Time

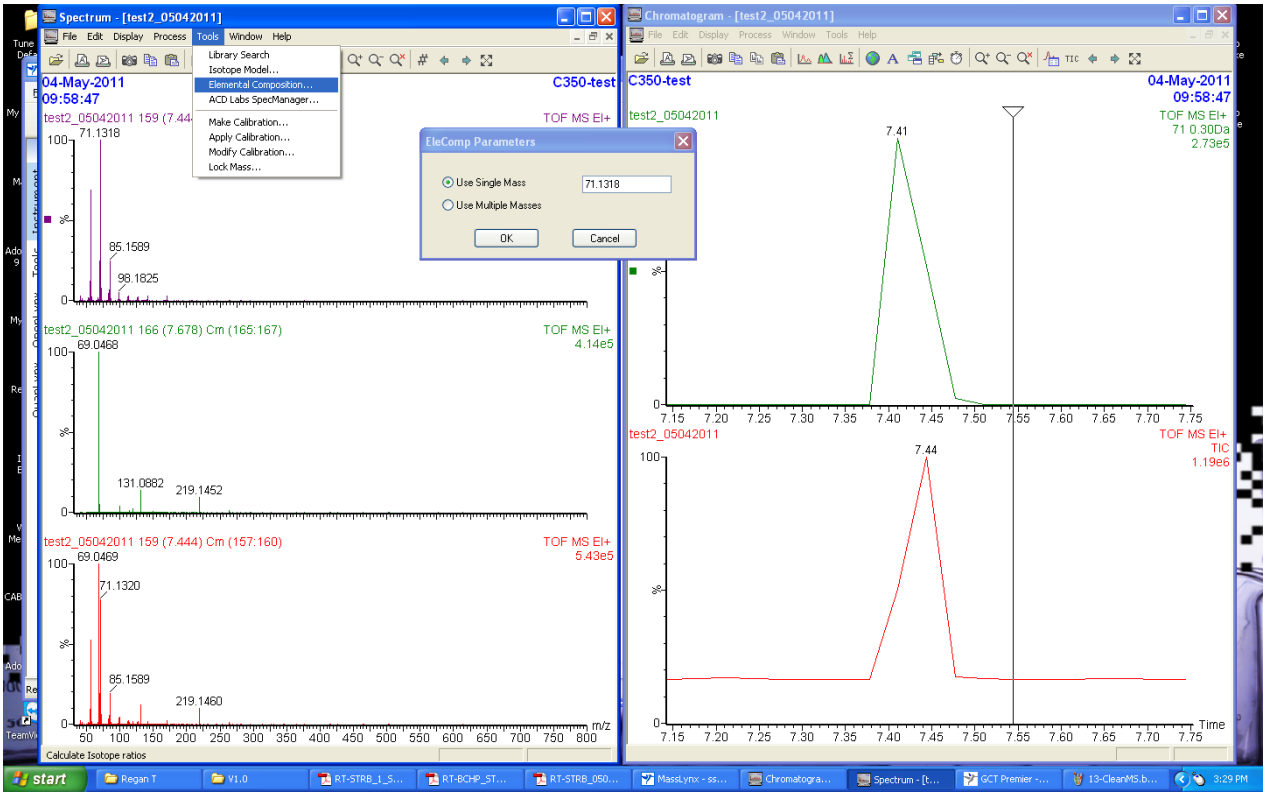
17. To print a single clean spectrum (as opposed to stacked spectra), select the undesired spectrum and hitting ‘Delete’ until only the desired spectrum is left. Complete the task by hitting Ctrl+P or ***File >> Print***.

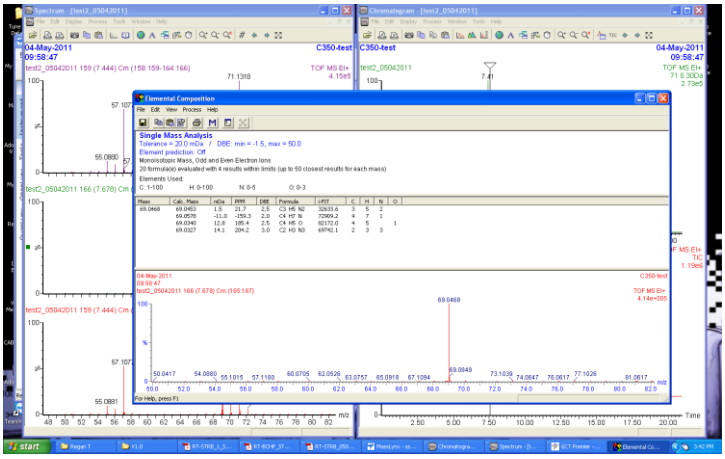
A selection is denoted by a similar-colored square to the left of the spectrum.

[7] Analyzing the Mass Spectrum

Elemental Composition

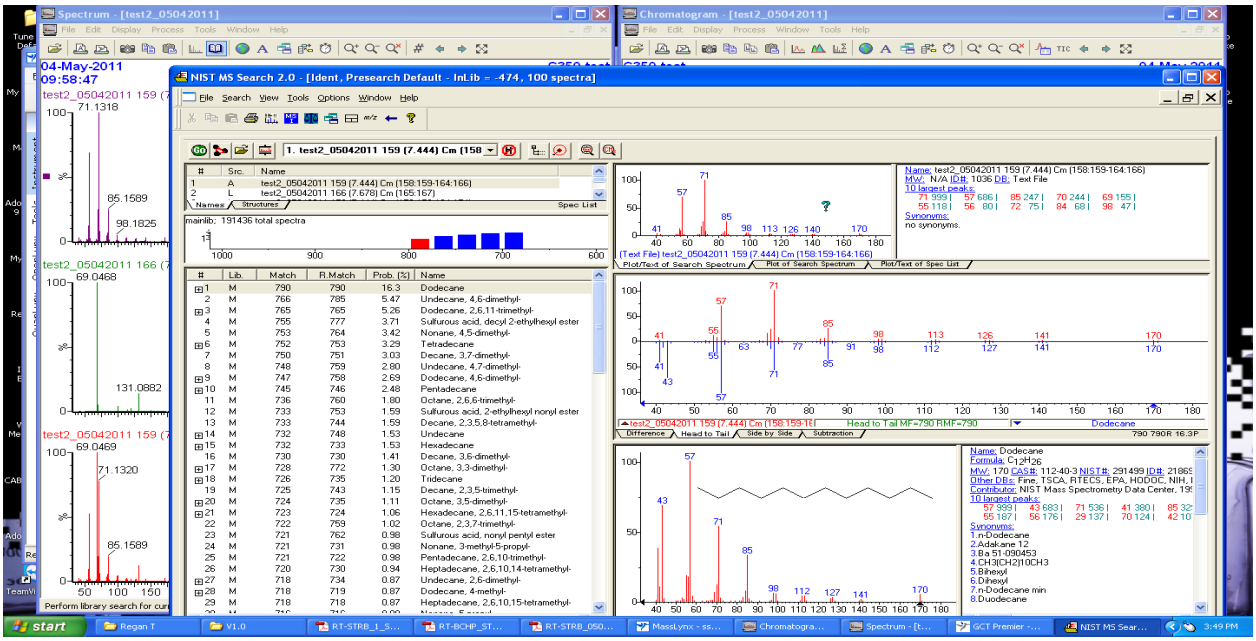
- ***Tools >> Elemental Composition…***
- Zoom-in and double left-click on the m/z signal of interest to select a single mass for elemental analysis. Click OK when finished.





NIST Database Comparison

- With the desired spectrum selected, click the NIST Database icon 
- The experimental spectrum will appear on top in **red**, the database spectrum of the best match below in **blue**.



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