

Important Notes

1. Ensure unique sequence names at all times.
 - a. **Never restart a run from an old sequence.** If the samples are the same, **this will overwrite your data.**
2. Sequence lists should have the sequence name printed with the header
3. If prompted **Choose load method options** when opening the software, choose "Download to instrument".

Aborting a run

1. If aborting a run, make sure that it has been removed from the sequence queue before trying to start a second run or troubleshooting. The software defaults to resuming where a sequence has been left off. **Do not pause your run.**
 - a. To abort a run, go to the run control drop down and select **Stop Run/Inject/Sequence.**
 - b. In the sequence queue, highlight the stopped sequence and hit the black X to delete it.
 - c. Ensure that the sequence queue is **empty** before adding a new sequence or troubleshooting.
 - d. If the autosampler stops working the run may also need to be aborted on the computer.

Creating and running a sample list on Agilent GCs

1. Check to ensure gas tanks have a sufficient amount for the run.
2. Ensure that the online module is open by clicking **(Instrument #) Online** icon on the desktop.
3. Ensure the method and run control tab is highlighted blue in the left pane
4. Click Sequence → New Sequence Template
 - a. In the top right sequence should change from the most recent sequence run to **Def_GC.S**
5. In the main screen, click the tab for **Easy Sequence**
 - a. Open the template: **RGC(#) Easy Sequence Template** by clicking the folder icon next to easy sequence setup.
 - b. Ensure box with method Information displays the correct dated method.
 - c. Under the data information box select the data location
 - i. The default location for data is the Tox Data folder. If this needs to be changed, select appropriate parent folder by clicking the (...). The subfolder is defaulted to have the same name as the sequence list.
 - d. Under the sequence information box, select the sequence location
 - i. The default location for sequences is the Tox Sequences folder. If this needs to be changed, select appropriate parent folder sequence location by clicking (...).
 - e. Next to Sequence name, name the sequence.

- i. When running multiple sequences in the same day, use a unique name for every sequence. (ex: if a second DSC needs to be run for the instrument, name it BACDSCYYYYMMDDRG#-1)
 - ii. It is preferred that A and B replicates are on different sequences.
 - f. Under sample Information for RGC5
 - i. Choose starting vial location.
 - 1. 101 starts the left Autosampler tray
 - 2. 201 starts the right Autosampler tray
 - 3. It is not possible to run on both trays with the same easy sequence
 - ii. Input number of samples.
 - g. Press **Fill Samples** button to load the sequence list
 - h. Name samples
 - i. **DO NOT USE DECIMALS (.) OR DUPLICATE NAMES IN THE SAMPLE LIST OR THE FILES WILL OVERWRITE**
 - ii. If you would like to label controls as 0.100g%, enter a name without decimals under the column **sample name** and fill out 0.100g% under **sample info**
 - i. When ready to run, click **Save and Add to Queue**. The green bar saying **Ready** should change to a blue bar for **Sequence Running**.
 - i. Navigate to the queue tab in the main window and ensure that the queue does not have warnings. If the queue is **not accepting sequences**. There is a problem.
 - j. If you are running on RGC3 or RGC4 remember to **start the Autosampler**. If you are injecting two trays, make sure both are correct and that when you start, you tell it to run all jobs in the sequence.
 - k. Repeat for the B set if necessary. (For RGC5, this can be done while the A set is running)
- 6. Once the run is complete, put the GC to sleep by clicking Method→Load Method→**SLEEP**RG#)
 - a. Instruments can also be put to sleep using the Chem Station Scheduler
 - i. In the top menu bar select view→ Chem Station Scheduler
 - ii. Input date and time (in military time) that you want the instrument to go to sleep. This means you need to know when your run should end.
 - iii. Under command write LoadMethod,SLEEPRG#).M
 - iv. Click save
 - v. The scheduler needs to remain open for command to be accepted.
 - b. Instruments without Chem Station Scheduler can also be scheduled to be put to sleep using Queue method.
 - i. In the top menu bar select Run Control→Queue Method
 - ii. In the Method box select browse and select the SleepRG#) method for that instrument.

- iii. In subdirectory box add the date
- iv. Leave sample location empty (to indicate a blank run)
- v. Add sample name: SLEEP
- vi. Select **"Add to back of queue"**. MUST select this option only or your run WILL be disrupted and not acceptable for use.

Updating the DSC Retention Times (done post-maintenance or on an as-needed basis)

1. Ensure that only online mode is open.
2. Ensure that the correct method is loaded. If it is not, this can be selected under method→load method.
3. The software defaults to the most recent run being loaded. If this does not match the sequence you need:
 - a. Click the **Data Analysis** tab in the left pane under the file folders.
 - b. Select the Daily System Check's run from the folders on the left, double click to load.
4. The sample list be visible in the main window on the top. Double click to load a specific sample (it will be highlighted in blue). Right click and preview report brings up the report. Copy down the retention times.
5. Directly below the sequence list pane, click calibration (next to the scale icon) to load the calibration table. Both the front and back FID are in the same table. The retention times should be imputed under the column **RT** ensuring that it corresponds to the correct **Compound** and **Signal** (**A** for the Front FID, **B** for the back FID)
 - a. **Press OK at the top of the sequence table** after the RTs have been updated. The calibration table should disappear. This ensures no conflict can be generated by having the table open while reprinting the sequence.
6. From the drop down menus on top click Method→Save Method. You will be prompted to add a comment for the audit trail. (i.e. Updated retention times to reflect DSC-your initials)
 - a. Check the method audit trail to ensure that you did save your method.
7. **Reprint the DSC data with the updated method.**
 - a. This can be confirmed by the fact that the expected RTs will match the case RTs.

Data Printing

*Data automatically printed during a run (**preferred**):*

***Do not print your sample list until you have retrieved your data. Doing this will overwrite the last PDF created. It is preferred that you print your sample list after a run so that the easy sequence template will have the sequence name in the header.

1. To access individual data files, go to my computer
 - a. Data (D:)→RGC(#) data→Parent folder (ex: Tox Data)→Sequence→Sample name
 - i. The PDF is under the folder with the sample's name
 - ii. This can be copied to a new location (Using right click→copy) or dragged to a USB

- b. To print a second page, right click on the file and select **print**
 - i. Select the file you would like to print to
 - ii. Check box saying **append to existing pdf**.
 - iii. Save file
 2. To **print all files** from a run
 - a. Use the windows search function in the start menu with the following search parameters:
 - i. All files and folders
 - ii. Location: browse (Select folder from D: drive as referenced in section 1)
 - iii. Word or phrase: .pdf
 - b. The files can then be copied to a flash drive
 3. Printing a sequence
 - a. Before a run
 - i. Save your sequence, by using the disk icon in the easy sequence.
 - ii. The default file name in the upper right should now have your sequence name
 - iii. Hit the print preview button and print the sequence
 - b. After a run
 - i. PRINT/RETRIEVE ALL OF YOUR DATA FIRST
 - ii. Locate and load the easy sequence
 1. From the easy sequence window in the method run control tab
 2. Click load sequence in the header and choose the correct sequence
 - iii. Hit the print preview button and print the sequence

Data printed from offline mode

4. The offline module is used for data processing and accessed by clicking either **Instrument (GC#) Offline** on the desktop.
5. Load analysis method for current calibration BACRGC(#)(caldate)
6. Ensure the data analysis control tab is highlighted blue in the left pane
7. In the folders visible in the left pane, choose the appropriate run and double click to load
 - a. The sample sequence list should appear at the top of the main screen.
8. Double click on the desired sample to load its data.
9. To print, right click on the sample and select print report. Both front and back columns are printed on the same report.
 - a. Save as PDF with appropriate name
 - b. IMPORTANT: If saving an addition to a previous file select file and click the box for **append to existing PDF**. You will not be prompted as with Varian GCs. Failure to click box will overwrite your existing file.

Calibration

1. Create a new dated method for the calibration

- a. Load the most current dated method.
 - b. Click method→save method as
 - i. Save method with new date of calibration.
 - ii. Enter comment about new method creation for the audit trail
2. Load the most recent sequence calibration method: Sequence→Load Sequence
3. Correct the method in the sequence table
 - a. Open Sequence table (Sequence→Sequence Table)
 - b. The samples for calibration should already be named. To change the new method for calibration choose the newly create method in the method name dropdown, highlight the column for all samples and right click select **Fill down**
 - i. Check that the method change has occurred under the method column
 - ii. Ensure that the calibrators have the sample type:
 1. Calibrators: Calibration
 2. Verifiers: Sample
 - iii. The calibrators should have a level selected:
 1. 0.010g/dL = level 1
 2. 0.025g/dL = level 2
 3. 0.050g/dL = level 3
 4. 0.100g/dL = level 4
 5. 0.200g/dL = level 5
 6. 0.400g/dL = level 6
 7. 0.500 g/dL = level 7
 - iv. The first calibrator, should have **replace** selected from the drop down menu in columns **update RF** and **update RT**. Any subsequent levels/samples should have **average**.
 - c. Ensure that a negative is included after the 0.50g% calibrator and before running verifiers. This can double as the IS QC check.
 - d. After this is verified correct, click ok.
4. Update sequence parameters
 - a. Open Sequence Parameters (Sequence→Sequence Parameters)
 - b. Choose data path for the calibration files
 - i. For RGC5: D:\RGC5 data\Calibration
 - c. Name the subdirectory with the date and GC#
 - i. If prompted that this subdirectory does not exist, yes, create it.
 - d. Click ok
5. Save the new sequence template
 - a. Sequence→save sequence template as (use current date)
 - b. Ensure that this is now the name you see on the top right of the main window as the current sequence.
6. Start the sequence
 - a. Open the sequence table

- b. Click **Run**

Printing Calibration Results

Note: Process calibration in ONLINE MODE only so as to avoid the possibility of having two versions of the method in existence.

IF PRINTING ONTO THE SAME DOCUMENT REMEMBER TO CLICK "APPEND TO EXISTING PDF" BEFORE HITTING SAVE. YOU WILL NOT BE PROMPTED.

1. Ensure that the correct method is loaded
2. Print the sequence list
 - a. Under tab method and run control
 - b. Sequence→Print Sequence
 - c. Check:
 - i. Sequence parameters
 - ii. Sequence output
 - iii. Sequence summary
 - d. Uncheck:
 - i. Sample related custom fields
 - ii. Compound related custom fields
 - e. Click **Selected Columns**
 - i. Check: Vial, sample name, method name, cal level, update RF, update RT
 - ii. Click OK
3. Print method
 - a. Under method and run control tab
 - b. Method→print method
 - c. Hit **Check all**
 - i. Uncheck **Custom fields**
4. Print curves
 - a. Under Data analysis tabs
 - b. Select file→print→Calib Table+Curves
5. Print the file calibration data files from the data processing tab:
 - a. Ensure the method is the new calibration method: BACRGQ(#)(caldate)
 - b. In the folders visible in the left pane, choose the appropriate run and double click to load. The sample sequence list should appear at the top of the main screen.
 - c. Double click on the desired sample to load its data.
 - i. To print, right click on the sample and select print report. Both front and middle columns are printed on the same report.
 - ii. Save as PDF with appropriate name
 - iii. **IMPORTANT:** If printing to a single calibration file you must click the box for **append to existing PDF**. You will not be prompted. Failure to click box will overwrite your existing file.

6. Update the easy sequence setup template to reflect the new method
 - a. In online mode, selected method and run control tab selected from the left pane
 - b. In the main window, select **Easy Sequence Setup**
 - c. Load the easy sequence setup by clicking on the folder and selecting:
 - i. **RGC(#) Easy Sequence Template**
 - ii. In the method information box, click the (...) to the right of the method name.
 - iii. Select the new method.
 - iv. Save the change by clicking on the disk icon to the right of the file name.
7. After uploading to FA, scroll through the entire file again. On occasion the μ symbol in the method causes a print error. If this happens, print the file to PDF a second time and try uploading the new file.