HPLC Walkup Guide

1. Turn on power to all modules if they're not on. Each module should have a green lit power button.
2.  Open Chemstation Online.
3. Navigate to **Method and Run Control** if not already open.
4. Select the method for your analyte by double clicking a method file on the left to load it to the instrument.
5. If the pumps have been idle for several hours or if the solvents are changed, see **Purging** the pump below.
6. Check that there's enough solvent to complete the analysis. Update the bottle fillings by right clicking the pump module and selecting **Bottle Fillings**. Enter the approximate volume left in each channel's bottle. Failure to do this step may result in pumping air into the LC or premature abortion of the sequence.
7. Click the 

button to start the pump, column thermostat, and detectors.
8. Load your personal sequence template (**Sequence → Load Sequence Template**) or create a new one with your name (**Sequence → New Sequence Template**).
9. Open **Sequence → Sequence Parameters**. Edit the subdirectory name to the format "[Researcher]_[Analyte]_YYYYMMDD". You will be prompted to create the directory if it's new.
10. Check that **Post Sequence command/macro** is checked and **STANDBY** or **SHUTDOWN** are selected.
11. In the **Sequence Output** tab, check **Print sequence summary report**. Select the **Use Intelligent Reporting** radio button. Select your report template or use **XLS** or **XLS+chromatograms**. Check **Report to XLS**.
12. Click **OK**.

13. Right click the autosampler module and select **Assign Wellplates**. Change each sampler container to the appropriate sample plate type (54 vial plates or Micronic 0.75 mL). Click **OK**.

14. Open **Sequence → Sequence Table**. Enter your sample locations, sample names, and the method into the sequence table. You can copy sample names and locations from Excel and filldown the method name.

15. Click 

to start the sequence.

1 Data Analysis

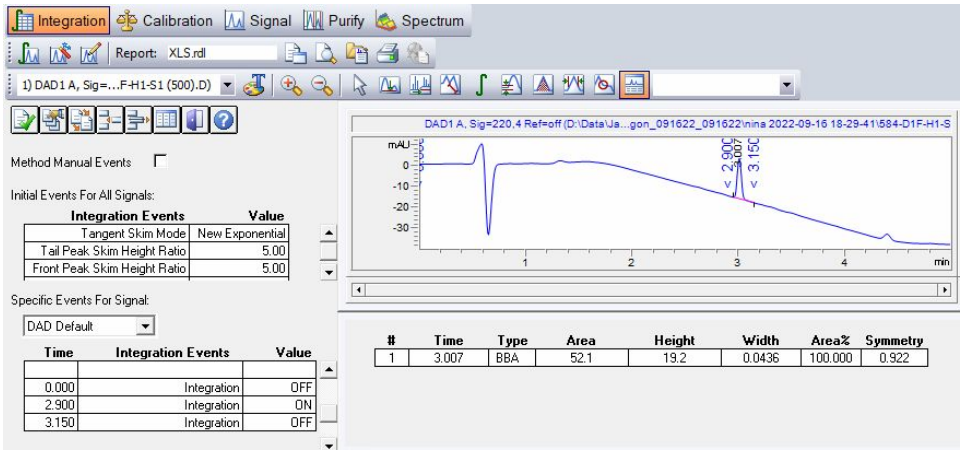
1. Open Chemstation Offline if not already open. 

2. Open **Data Analysis** from the bottom left menu.

3. Navigate to your sequence output folder and open your sequence.

4. Double click the data files and check that the integrations are correct. Otherwise, edit the integration parameters by selecting **Integration** and **Edit/Set Integration Events Table**.

5. Adjust the integration time windows and area/height thresholds to fit your data.

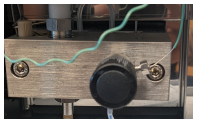


6. Click the green checkmark button to save the parameters. 

7. If the integration parameters had to be changed, click the green arrow to reprocess the sequence. 
8. Navigate to your sequence folder in Explorer and collect your xls report for your data.

2 Purging

These steps are necessary when changing solvent reservoirs or if the pump has been idle for a few hours or more.

1.  Open the purge valve on the pump by turning counter-clockwise 1/2 to 1 full rotation. This may look different or be in a different location depending on the specific pump model.
2. Right click the pump module window in ChemStation and select **Method**
3. Edit the solvent composition parameters so the pump is using 100% of the changed channel. Channel A composition is calculated from the composition of the other channels.
4. Set the flow rate to 5 mL/min and click OK.
5. Turn the pump module on in the software with the small green button if not already on. 

6. Allow the pump to run until all air bubbles have been purged from the solvent lines.
7. Reload the method without saving or restore the flow rate and compositions to their original settings.
8. Close the purge valve by rotating clockwise.
9. Continue with setting up your analysis.