

Thermo FlashSmart CHNS/O Elemental Analyzer

8/23/2020 SDC

Location: 1240 Hach Hall

Contact: Sarah Cady or Brett Boote, 1234 Hach Hall

Safety

All researchers working in 1232 Hach Hall must complete the EH&S Course “*Fire Safety and Fire Extinguisher Training*.” Aprons, safety glasses, and rubber gloves are available in 1238A Hach Hall if you wish to conduct your sample preparation in CIF facilities. Researchers should wear safety glasses in 1240 Hach. Researchers working directly with the FlashSmart CHNS/O Elemental Analyzer must also complete “*Laboratory Safety: Compressed Gas Cylinders*” training available on the [EH&S website](#).

Properly dispose of empty sample containers or glass vials from CHNS consumables in the broken glass container in 1240. Dispose of used CHNS combustion tubes in the box below the CHNS instrument in 1240.

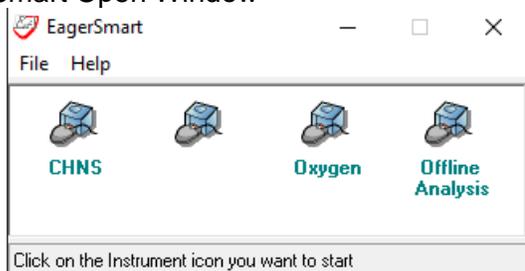
Some safety concerns specific to combustion EA include:

- Components of the system may be very hot. Use leather gloves when handling combustion tubes or engage the change reactor column feature of the software and ensure the system has cooled down sufficiently before handling.
- Users should be aware of compressed gas safety considerations when changing cylinders.
- Only trained users may independently use the CHNS for running their samples. You may also just be trained to weigh samples.

Preparing the System to Run Samples

Opening Application:

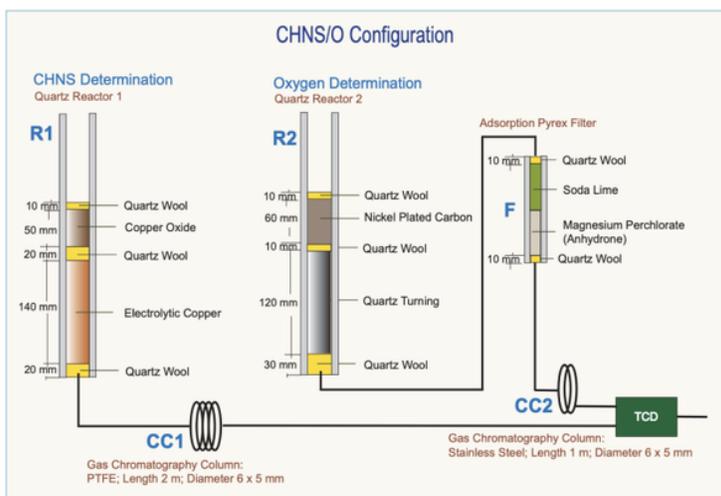
EagerSmart Open Window



There are three options for opening the EagerSmart application. Pick one based on what type of analysis will be carried out (offline analysis is useful for working on results during another run).

Reactor Packing and Configuration: CHNS/O Reactor Packing and Configuration Without Crucible

Size of the Filling Material (1)



Note In some cases, it is suggested to use Nickel Wool, in the top layer up the Nickel Plated Carbon, instead of Quartz Wool.

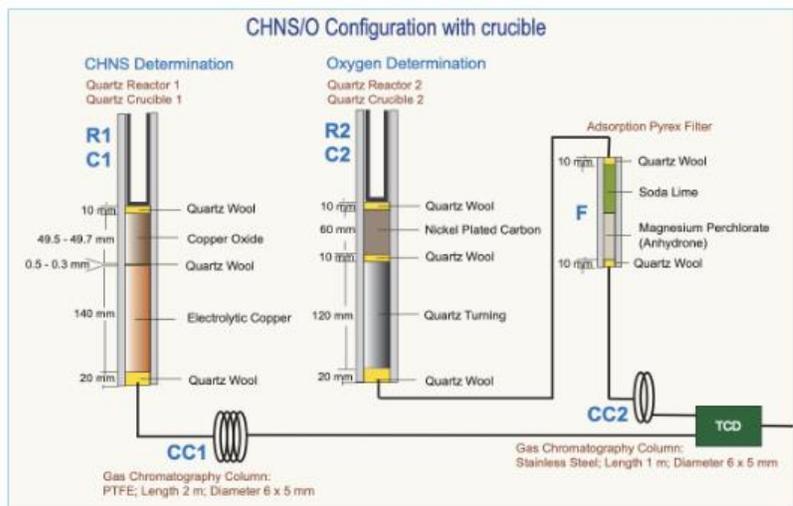
IMPORTANT CHNS Determination: If the sample has a high presence of inorganic material, we suggest to insert the quartz crucible into the oxidation/reduction reactor. In this case it is necessary to reduce the quartz wool between copper oxide and electrolytic copper up to obtain a thin layer and to reduce the copper oxide proportionally.

IMPORTANT Oxygen Determination: If the sample has a high presence of inorganic material, we suggest to insert the quartz crucible into the reactor of pyrolysis. In this case it is necessary to reduce from 30 mm to 20 mm the quartz wool in the lower section of the reactor.

Note Vanadium Pentoxide (V_2O_5) is an Oxygen donor: according to the sample nature, it is suggested to insert in the tin container with the sample, for a proper oxidation of material and consequently a quantitative Sulfur determination.

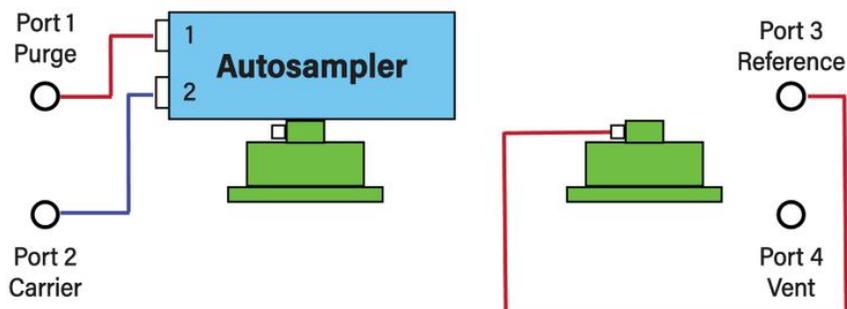
CHNS/O Reactor Packing and Configuration With Crucible

Size of the Filling Material (2)

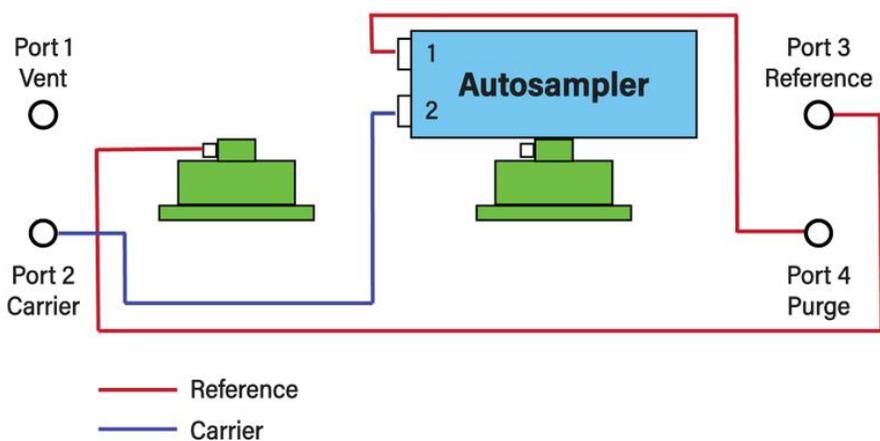


		EA Consumables	CE Elantech	
○	Quartz Wool	B1208	5 g	338 22200
○	Copper Oxide	B1248	50 g	338 21710
○	Electrolytic Copper	B1238	80 g	338 35314
○	Nickel Plated Carbon	B1086	5 g	338 23800
○	Quartz Turnings	B1271	50 g	338 22300
○	Soda Lime	B1211	100 g	338 35235
○	Magnesium Perchlorate (Anhydrous)	B1141	100 g	338 21900
--	Nickel Wool		2 g	338 25000
Reaction tubes (2)				468 20070
Quartz Crucibles (1)				252 04510

CHNS Configuration



Oxygen Configuration

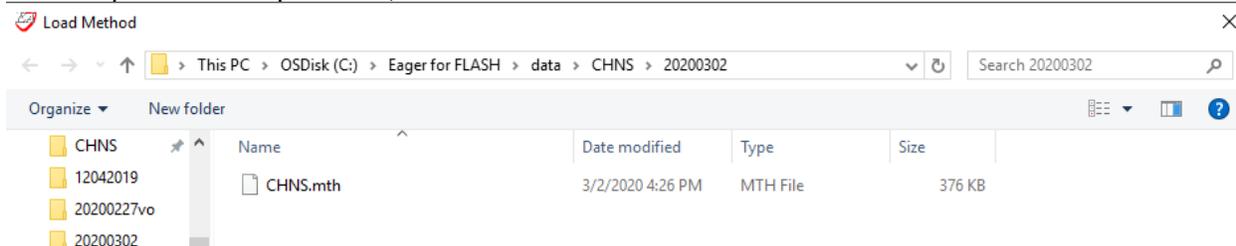


Setting Up Sample Table:

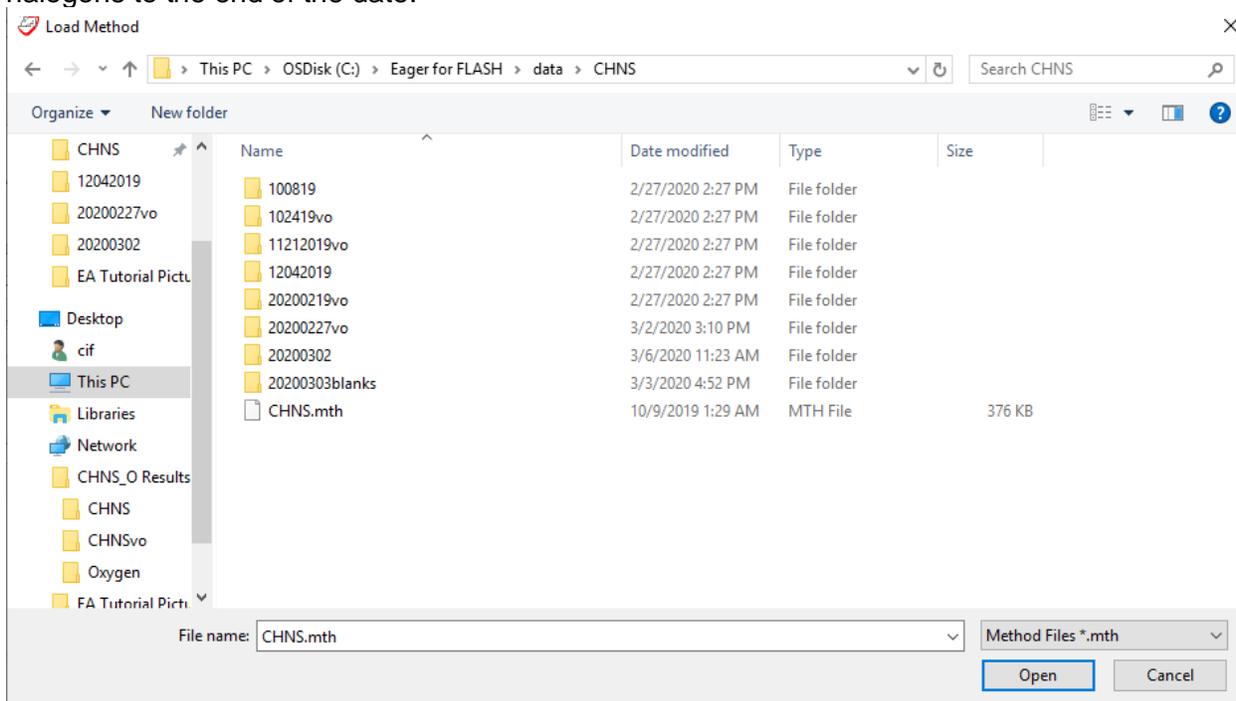
After opening the EagerSmart application and choosing a type of analysis, this window will pop up:

Actual	Level (uV)	Time	Channel status	Method	Filename of method in use
11 (No name)	2151	720.0 s	Waiting start	CHNS	c:\eager for flash\data\chns\20200302\chns.mth

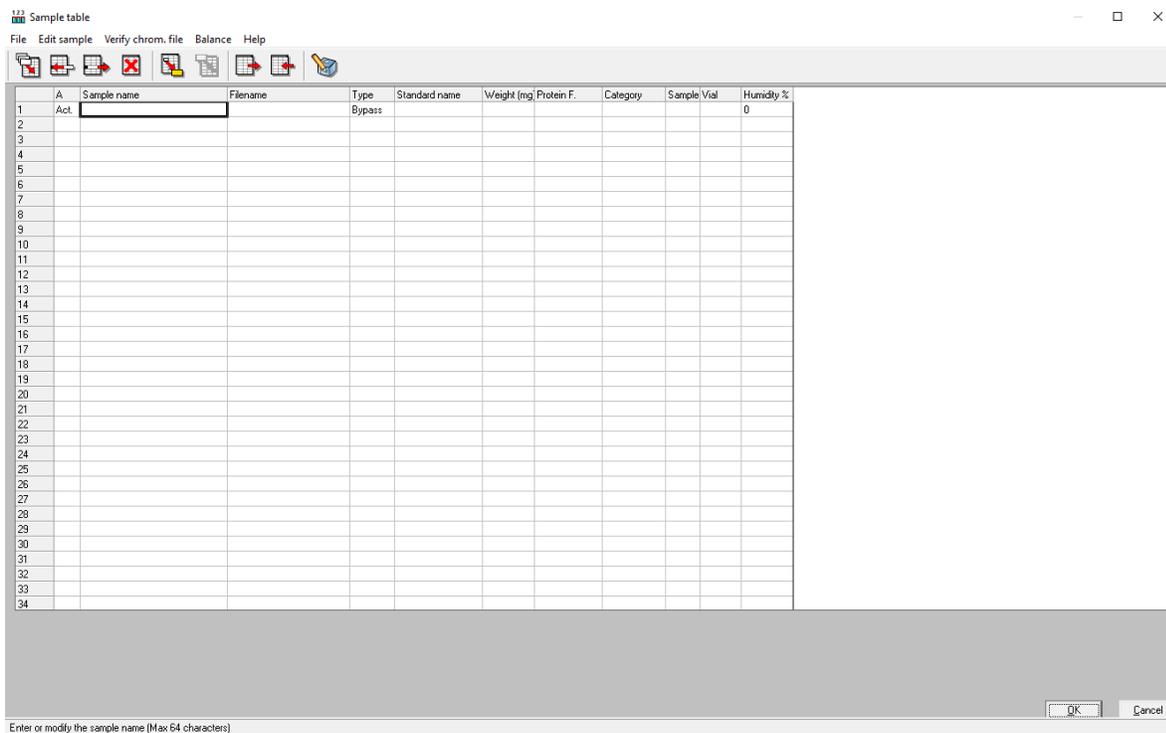
To set up a new sample table, select File and Load Method:



Under Data, select either CHNS or Oxygen (depending on type of analysis) and create a new folder titled “yyymmdd” using the date of set up. If creating a table for runs with halogens, add halogens to the end of the date.



Select the most recent data table used for the type of run (CHNS, oxygen, CHNS w/halogens). This is to ensure all properties of the method are correct for the type of run. Before making changes, save this method in the new folder. Then open the sample table and hit delete sample table, clearing the table for new data entry.

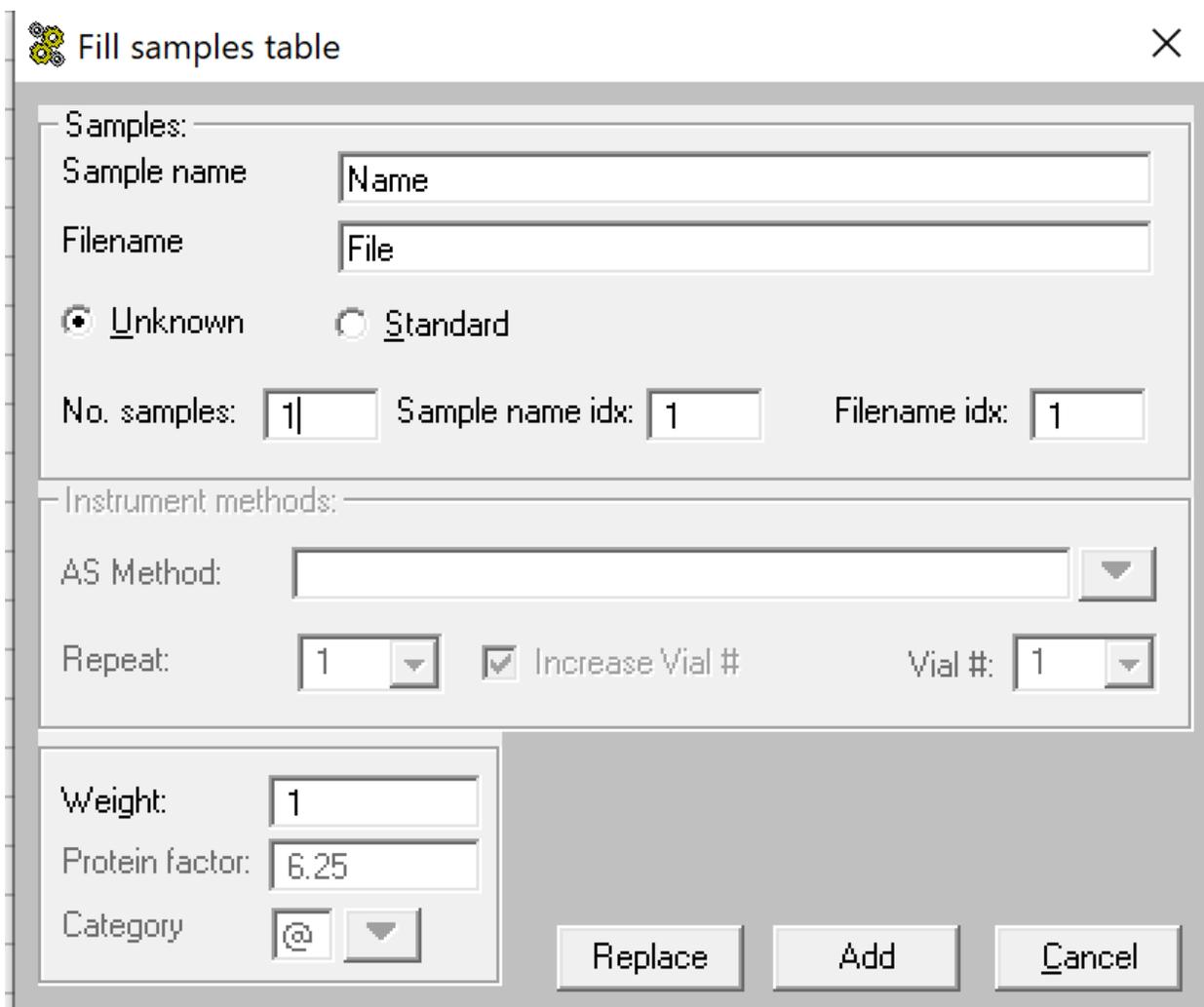


Click on the highlighted button, “Fill sample table”, and another window will open to set how the table will fill in.

1 2 3 Sample table



Sample name does not need changed as this will be entered in as samples are entered. Change file name to `yyymmdd_`. This will fill in the file name for all samples. No. samples will be 200. Everything else can remain the same. Click add.

 Fill samples table

Samples:

Sample name:

Filename:

Unknown Standard

No. samples: Sample name idx: Filename idx:

Instrument methods:

AS Method:

Repeat: Increase Vial # Vial #:

Weight:

Protein factor:

Category:

Before entering in samples, standards must be run for calibration and checks. The first eight rows should be set up as follows:

	A	Sample name	Filename	Type	Standard name
1		BBOT Bypass	20200828_001	Bypass	
2		Tincup	20200828_002	Bypass	
3		Tincup	20200828_003	Blank	
4		BBOT Standard	20200828_004	Std	BBOT
5		BBOT Standard	20200828_005	Std	BBOT
6		BBOT Standard	20200828_006	Std	BBOT
7		BBOT Check	20200828_007	Unk	
8		BBOT Check	20200828_008	Unk	

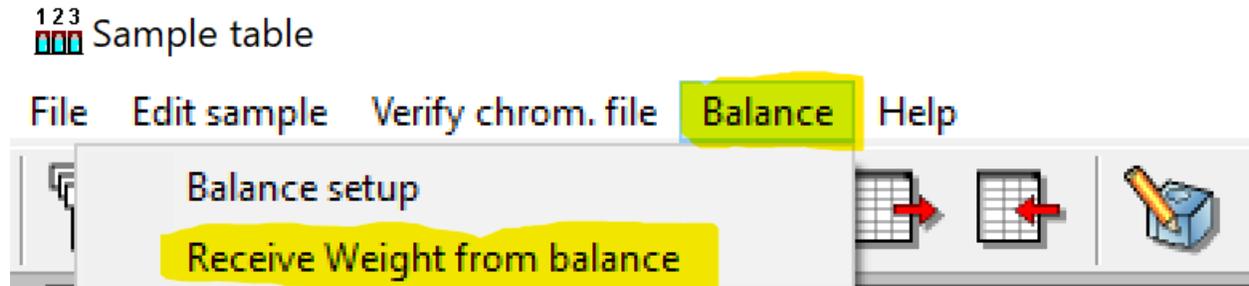
This setup is the same for halogenated samples, except silver cups should be used instead of tin.

From here on, samples can be entered to be run. Before each run, be sure to run two BBOT checks as unknowns.

Weighing and Wrapping Samples:

Samples should come prepared for analysis, although some might need further preparation. Samples should be a fine powder to allow for better combustion. If samples are too coarse, they should be ground into a finer substance. Liquid samples are trickier and depending on the thickness will decide how the sample will be packed for analysis.

To be able to enter the weights straight from the balance, on the sample table page, click balance=>retrieve weight from balance:



For solid samples:

1. Place a tin capsule on the balance.
2. Zero out the balance.
3. Retrieve tin capsule.
4. Add 8-10mg of Vanadium oxide now and zero the balance again with the capsule + vanadium.
5. Using a spatula tool, add a small amount of sample.
6. Place the tin capsule back on balance.
7. Try to keep the weight of the sample around 2.0mg, as adding too much sample can affect combustion.
8. Remove capsule and seal. Ensure that the sample is properly sealed and that none of the sample is falling out.
9. Place the sealed capsule back on the balance and press "Transfer"

For liquid samples:

1. Take one tin and flatten into a circle.
2. Place flat tin with another tin capsule on the balance.
3. Zero the balance.
4. Take the flattened capsule from balance.
5. Scoop a small amount of the sample onto the flattened capsule.
6. Place the flattened capsule back onto the balance.
7. Try to keep the weight of the sample around 2.0mg, as adding too much sample can affect combustion.
8. Transfer weight before adding vanadium
9. Add ~8-10mg vanadium oxide to the sample.
10. Remove flattened capsule and fold closed.

For halogenated samples:

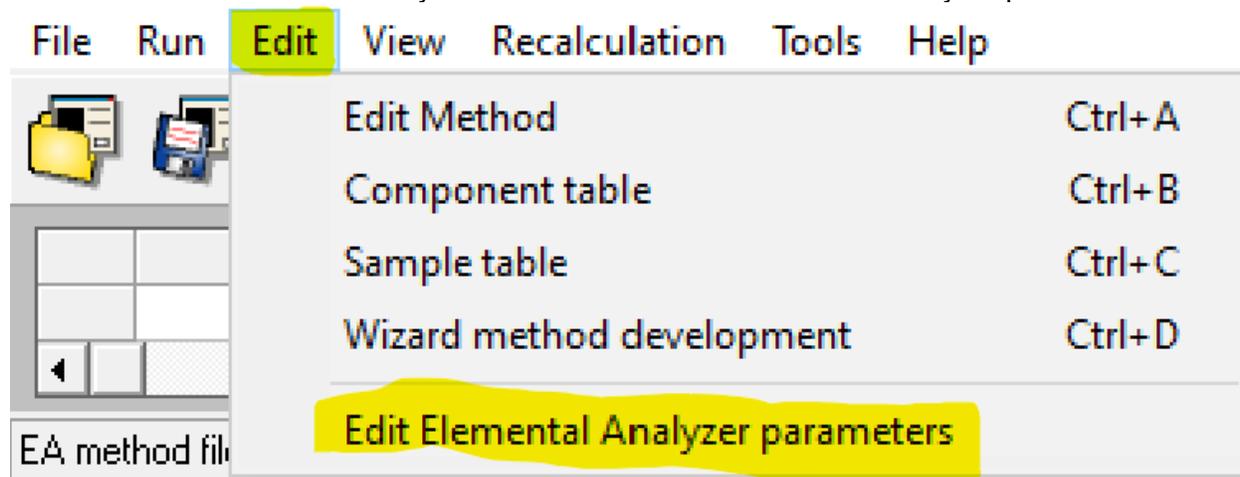
The process for weighing out halogenated samples is like regular samples, with a few changes. Instead of tin capsules, silver capsules are used to pack samples. Vanadium is still added to samples, but the weight of the samples should remain around 1mg instead of 2mg to aid in combustion.

For Oxygen analysis:

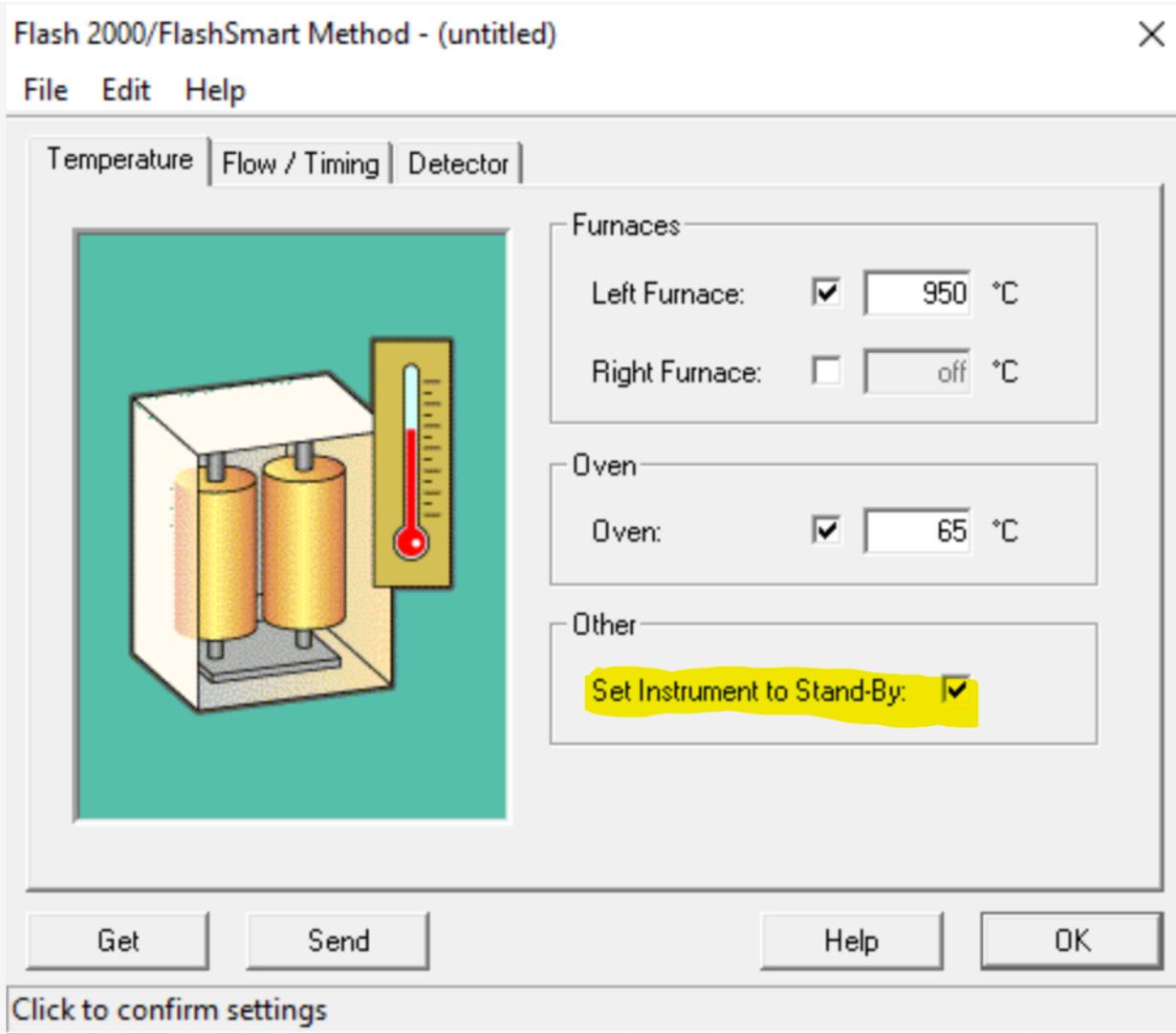
This process is also like normal CHNS samples, but silver capsules are used and no vanadium is added.

Preparing Instrument for Analysis:

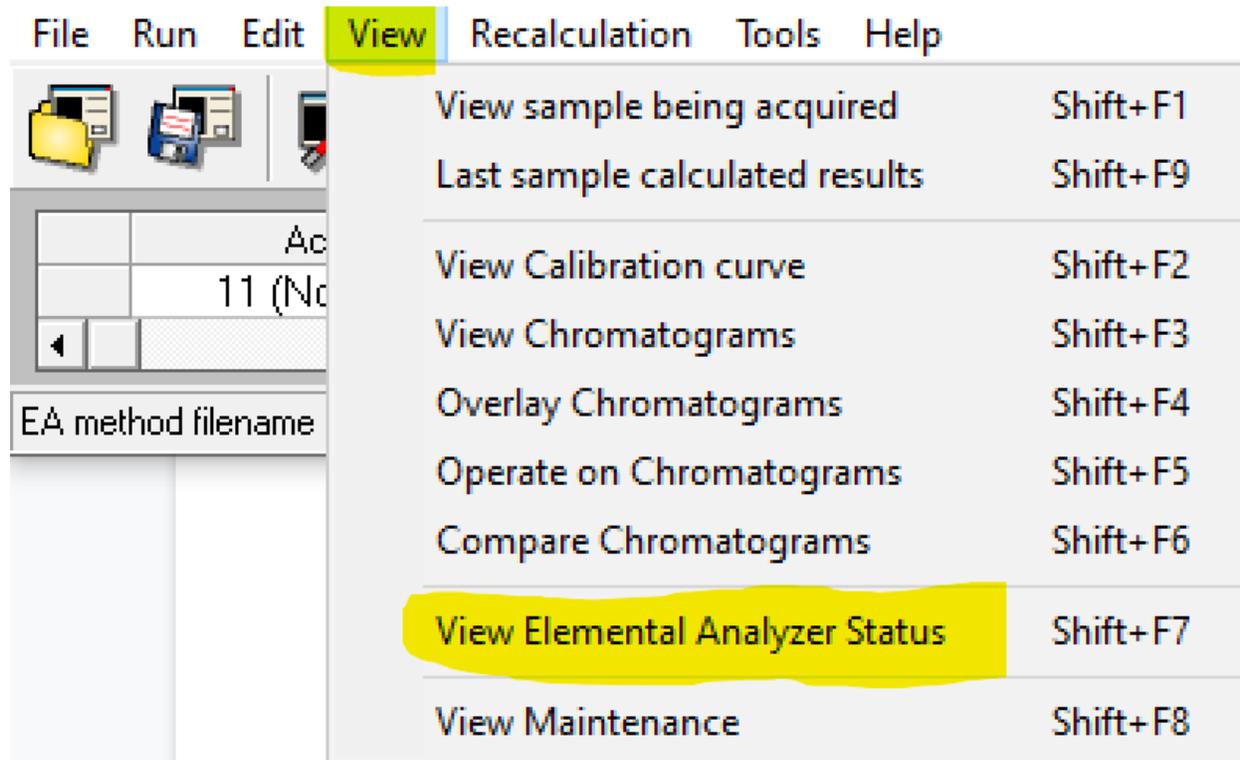
All the parameters should be set for the machine and saved in the method. The only parameter that will be of concern is standby mode. Under edit=>edit elemental analyzer parameters:



The following window will pop up. To begin heating up the furnace and to run samples, the machine must not be set to stand-by.



Now that the instrument is out of stand-by, the furnace must heat up. These settings are different for each furnace but should already be set. To check if the furnace is done heating up, click view=>view analyzer status:

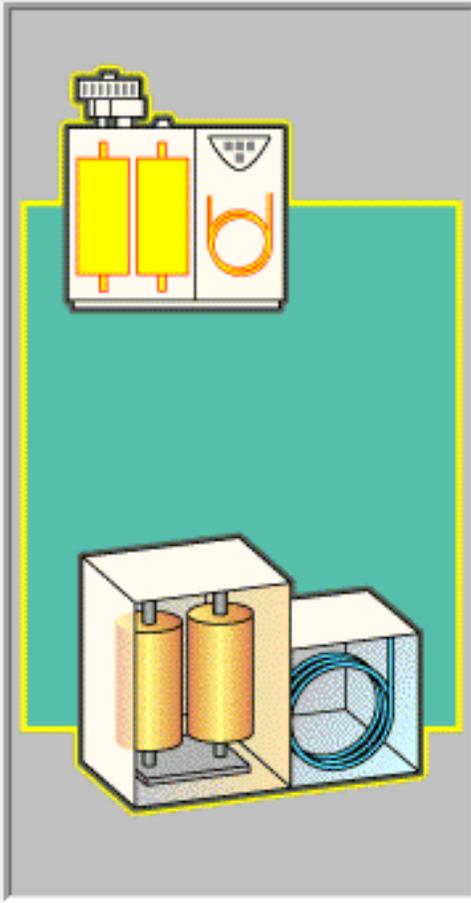


The following window will pop up:

Flash 2000/FlashSmart Status



- General
- Detector
- Auto-Ready
- Special Functions



	Set	Actual	
Left Furnace:	950	475	°C
Right Furnace:	0	24	°C
Oven:	65	65	°C
Temperatures Ready:		<input checked="" type="radio"/>	

	Set	Actual	
Carrier:	140	10	ml/min
Reference:	100	10	ml/min

Phase	
Run:	<input checked="" type="radio"/>
Sampling:	<input checked="" type="radio"/>
Oxygen Injection:	<input checked="" type="radio"/>

Step Sampler Tray Position

Help

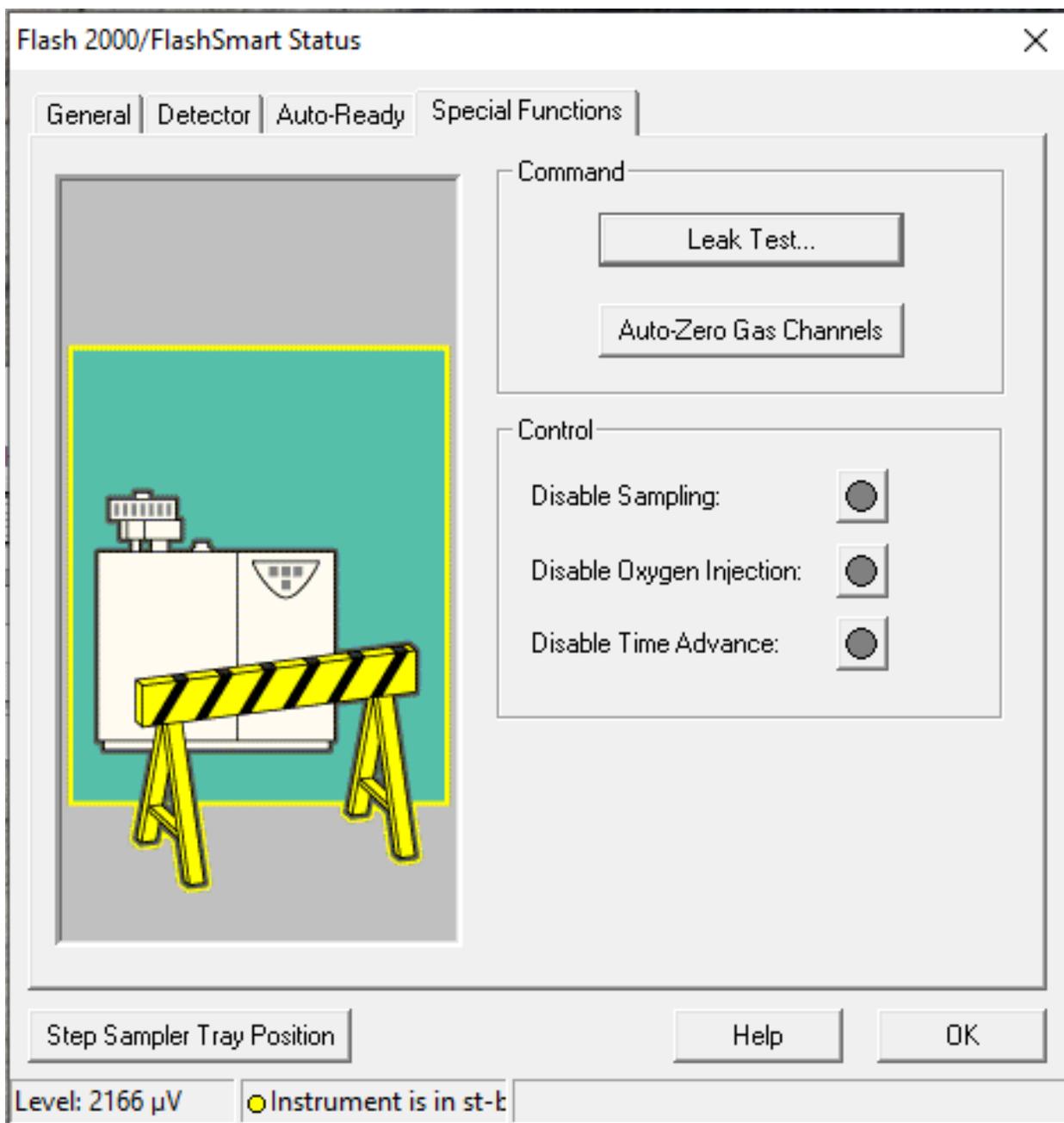
OK

Level: 2151 μ V Instrument is in st-b

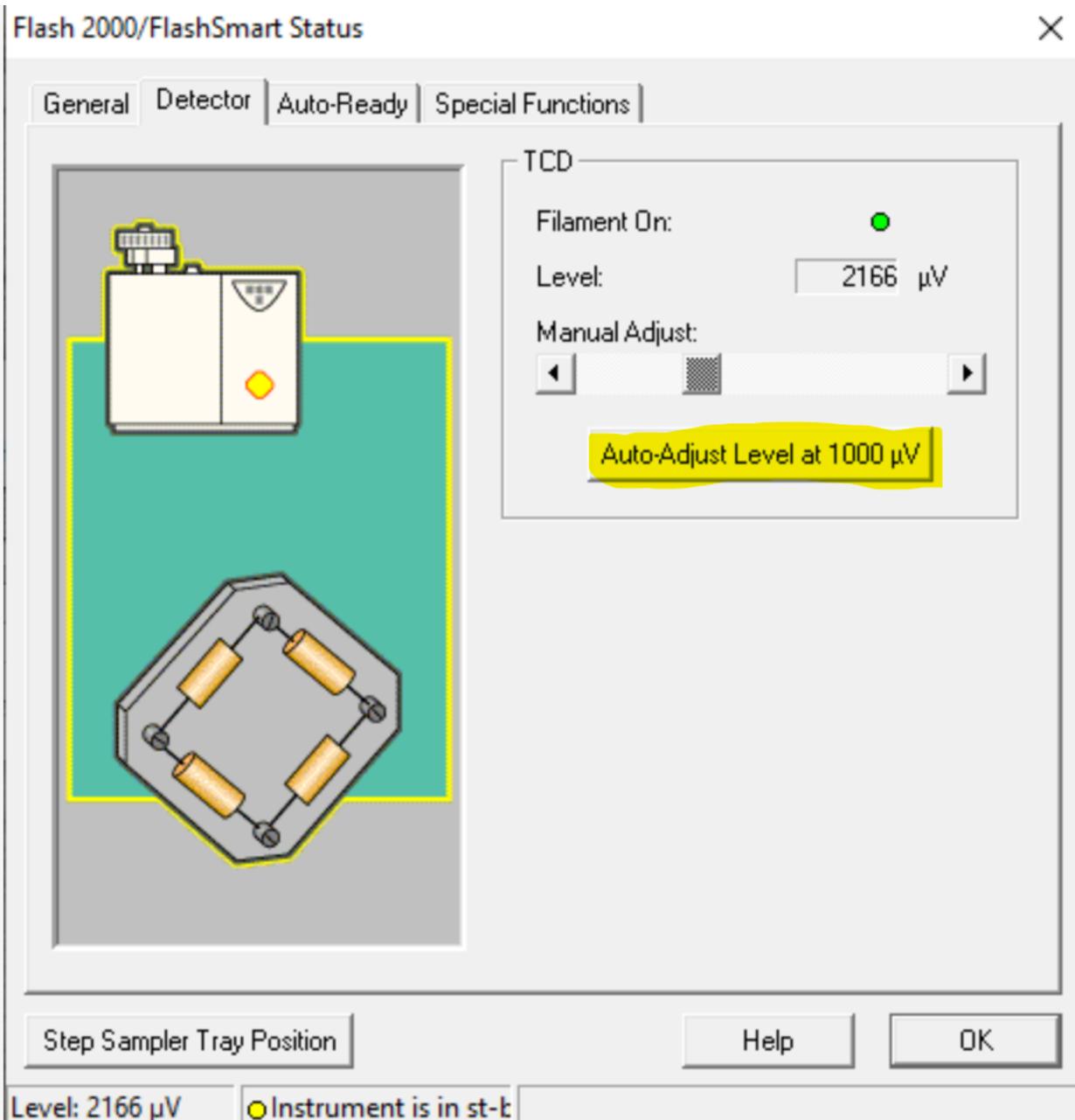
Under actual is what the furnace is currently at, and to be ready for analysis it should be what is under set. (Left: 950 for CHNS. Right: 1060 for Oxygen. Oven: 65 for normal CHNS/oxygen, 100 for halogen samples)

On this window there are other tabs that are needed to get the instrument ready for analysis. These tabs include Detector and Special Functions.

Special functions tab is where a leak test can be performed:



Both flow rates should drop below 5ml/min within a certain time. If a bit of time has passed (more than 90sec) then parts on the instrument must be tightened to prevent any leaks. Detector is used to adjust the machine to 1000 μV :



Click auto-adjust level at 1000 μV and the machine should begin adjusting to this level. It might take a few clicks for the machine to stay steady at this level. Now the autosampler can be loaded. Put the first sample to be analyzed in slot 1 and put subsequent samples from then on. Ensure that on the sample table in the first column, the first sample is listed as actual sample being acquired:

123 Sample table

File Edit sample Verify chrom. file Balanc

	A	Sample name
1	BBOT	BBOT
2		Tin Cup
3		BBOT Standard

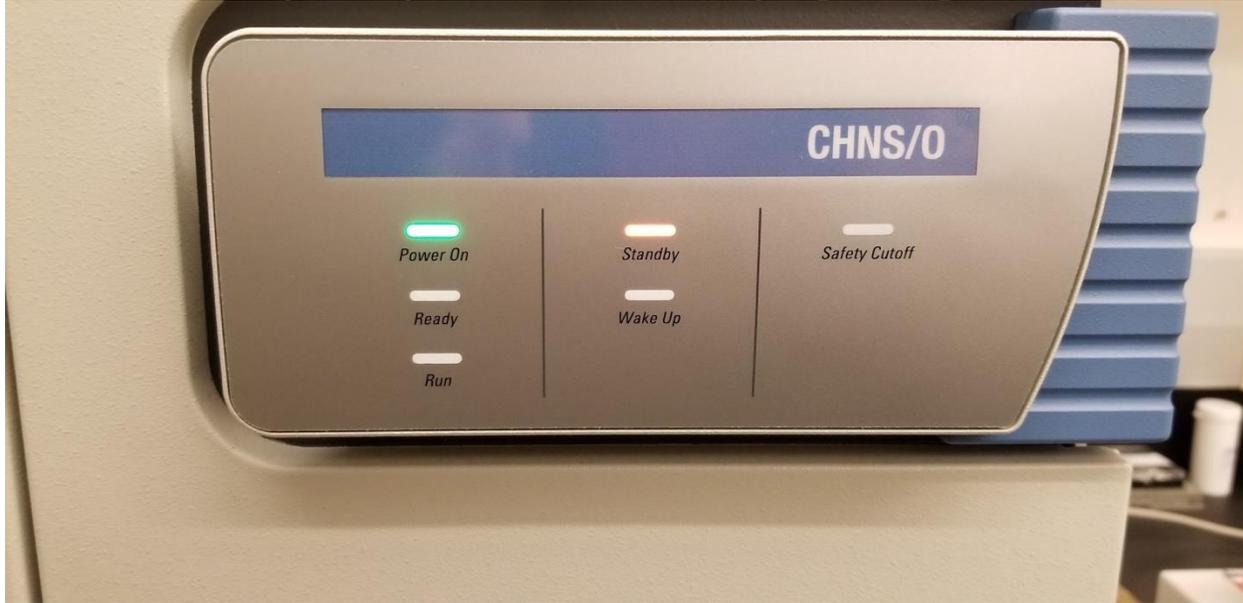
Actual sample

Sample # 1

Actual sample being acquired

OK Cancel

If the machine is ready for analysis, this will be indicated by a light turning on the machine:



To begin the run, select Run=>Start sequence of samples:

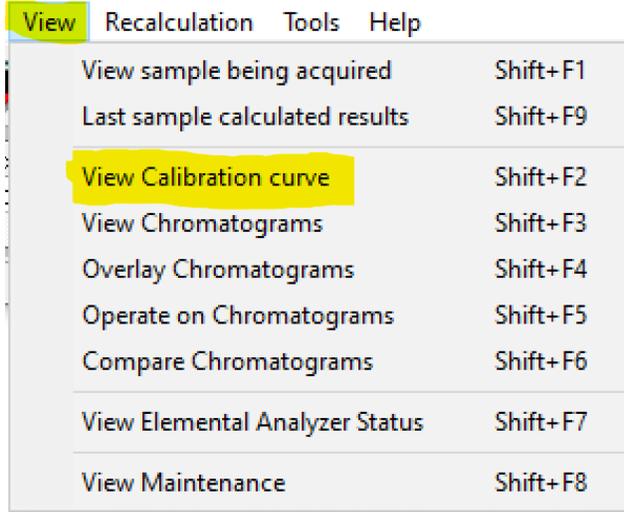
File Run Edit

- Start sequence of samples
- Stop sequence in progress
- Start single sample data acquisition
- Stop data acquisition
- Abort data acquisition
- Run Macro

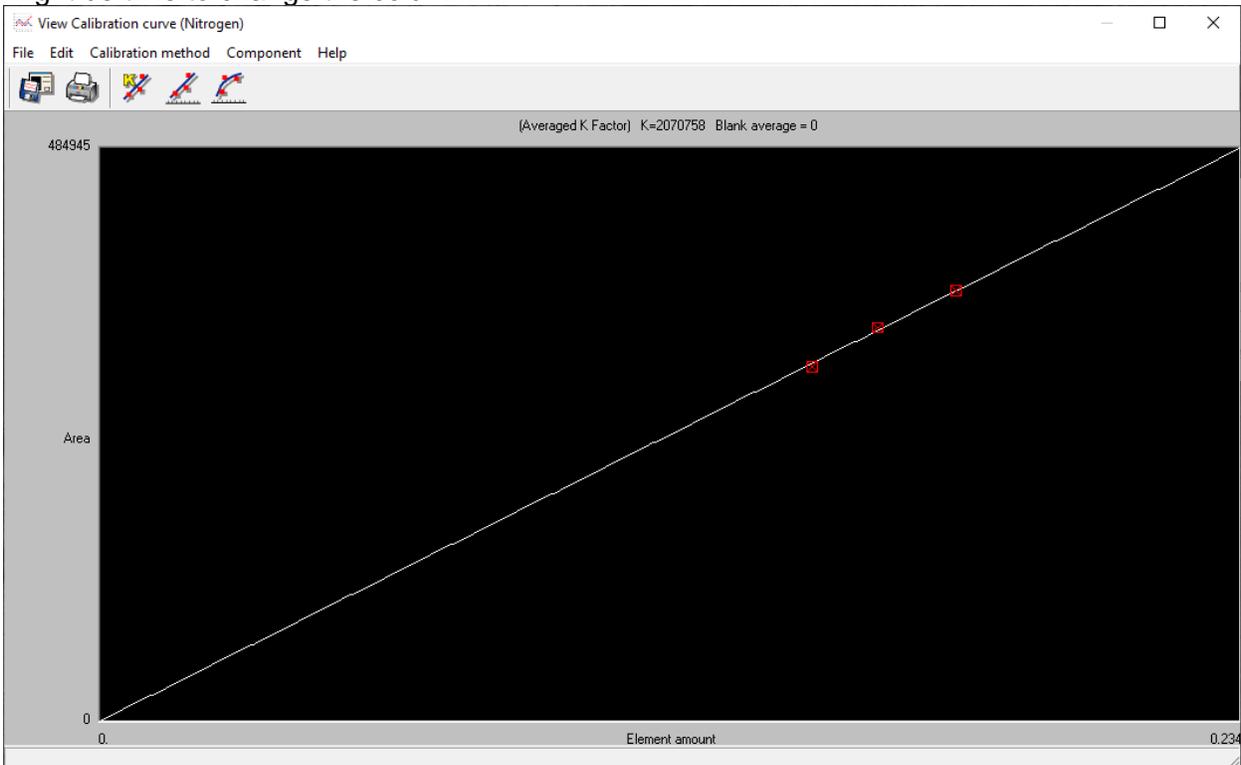
Another window will pop up and to make the machine go into stand-by mode to save energy after the run is completed, check the box Force to Stand-By.

Analyzing Results:

After the first run, the calibration curve formed from the results based on the standard runs should be checked. Select View=>View calibration curve:



As an example, the calibration curve shown below is what should be expected. If it seems off, it might be time to change the column.



If the calibration curve looks good, select Summarize Results to view initial results of the samples:



The window that pops up displays the results for each element. Under the column Group, number groups differing by each sample starting with 1. Then click Show summary sheet of all groups to get percent relative standard deviation, among other analysis. This is the important number to look at, as it should be below 3% for results to be trusted.

Summarize results (Element %)

File Select Result Edit View Print Help

Group	Sample name	Filename	Inj Date	Inj Time	Type
1	0 BBOT	20200302_001	03/04/2020	12:48	By-Pass
2	0 Tin Cup	20200302_002	03/04/2020	13:00	Blank
3	0 BBOT Standard	20200302_003	03/04/2020	13:12	STD
4	0 BBOT Standard	20200302_004	03/04/2020	13:24	STD
5	0 BBOT Standard	20200302_005	03/04/2020	13:36	STD
6	0 BBOT Check	20200302_006	03/04/2020	13:48	UNK
7	0 BBOT Check	20200302_007	03/04/2020	14:00	UNK

If the percent relative standard deviation is higher than 3 or the results seem odd, there are ways to change integration for each element for a sample.

The following are two methods to adjust integration after a run. From my experience, the second method is more powerful, but depending on the issue, you may need to try both. It is a good idea to understand both.

Changing the Integration parameters:

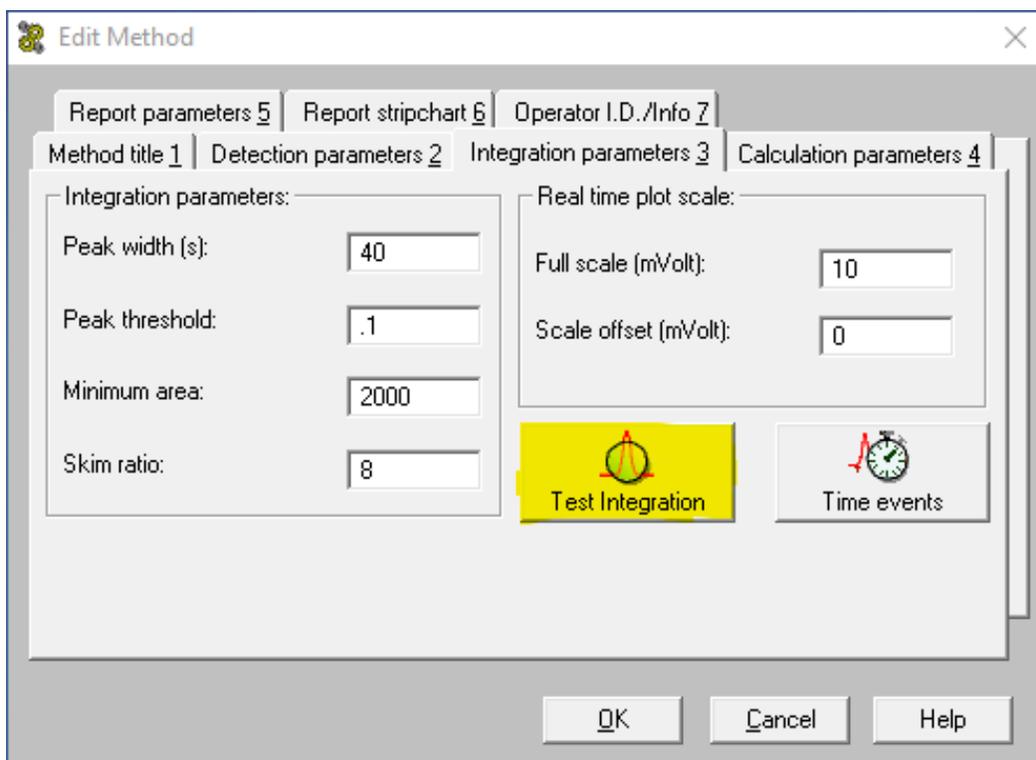
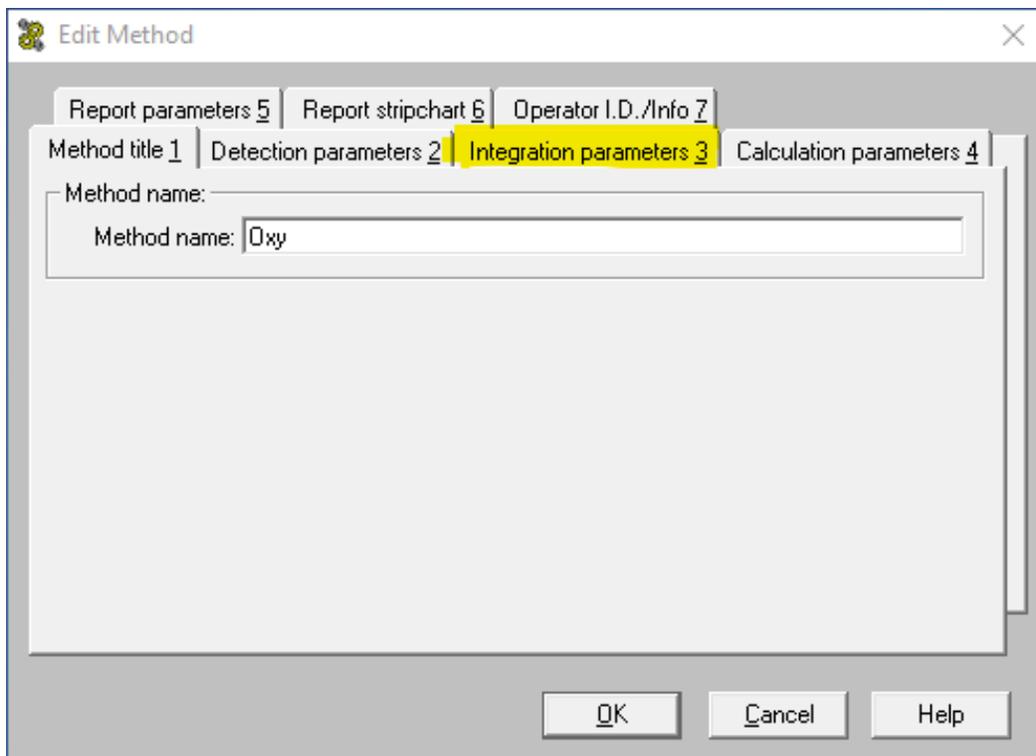
Actual	Level (uV)	Time	Channel status	Method	Filename of method ir
28 (BBOTcheck)	-2271	0.0 s	Waiting start	Oxy	c:\flash data\oxygen\20191009\oxygen.mth

EA method filename [Oxygen.eam]

Click “Edit” and then click “Edit Method.” This should be the first option in the edit drop-down.

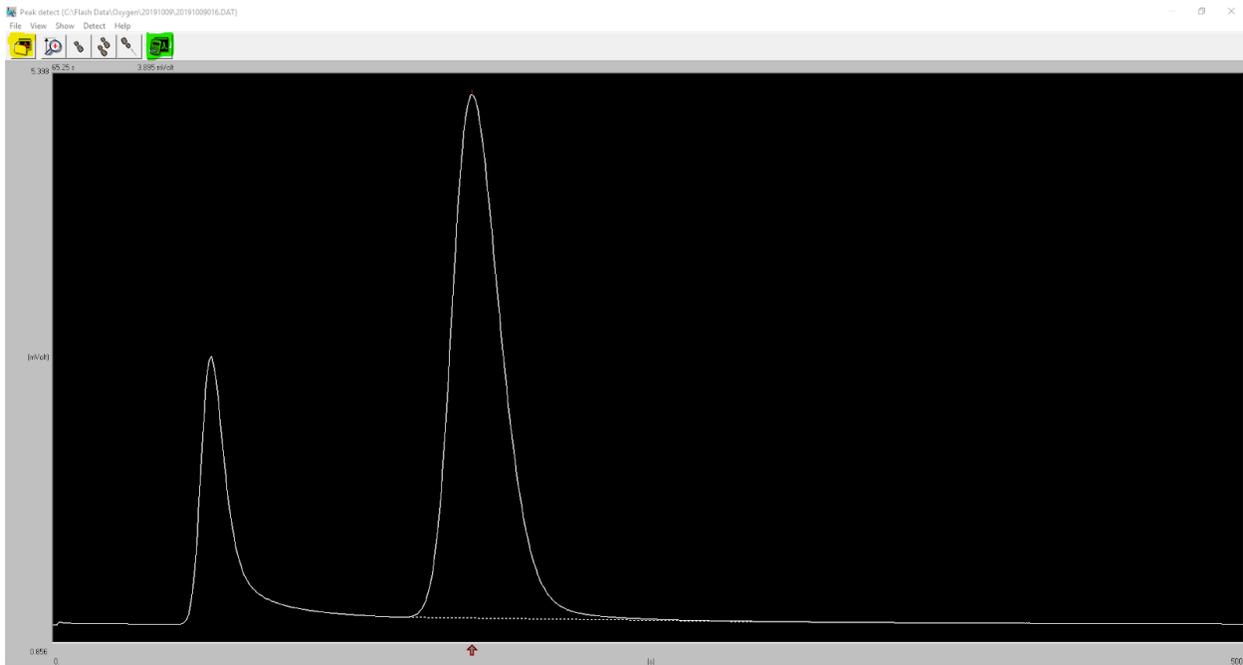
The Edit Method window will open and look as follows:

Click the “Integration parameters 3” tab. The Edit Method window will then look like:

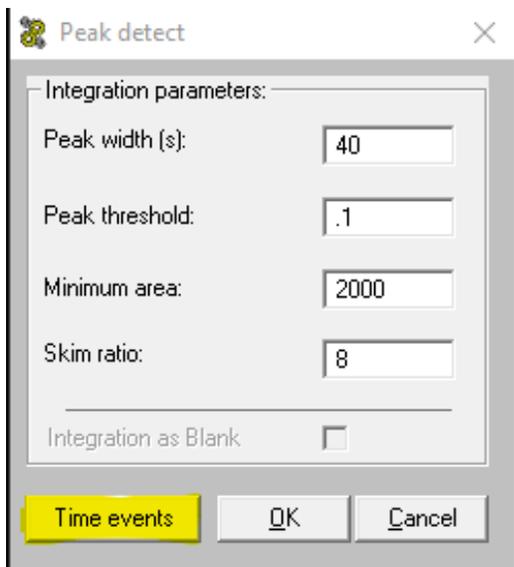


You can change any of the values in this window, but the current values are optimized for the instrument. You may have to decrease the minimum area if you know that a sample has a small amount of a certain element, but it is not being calculated. However, if you reduce it too much, you will pick up peaks that are due to residual sample from the run before (samples are not always perfectly cleaned out). Oxygen is usually set to 2000 and CHNS is usually set to 3000. I would not suggest going below 1000 (for minimum area).

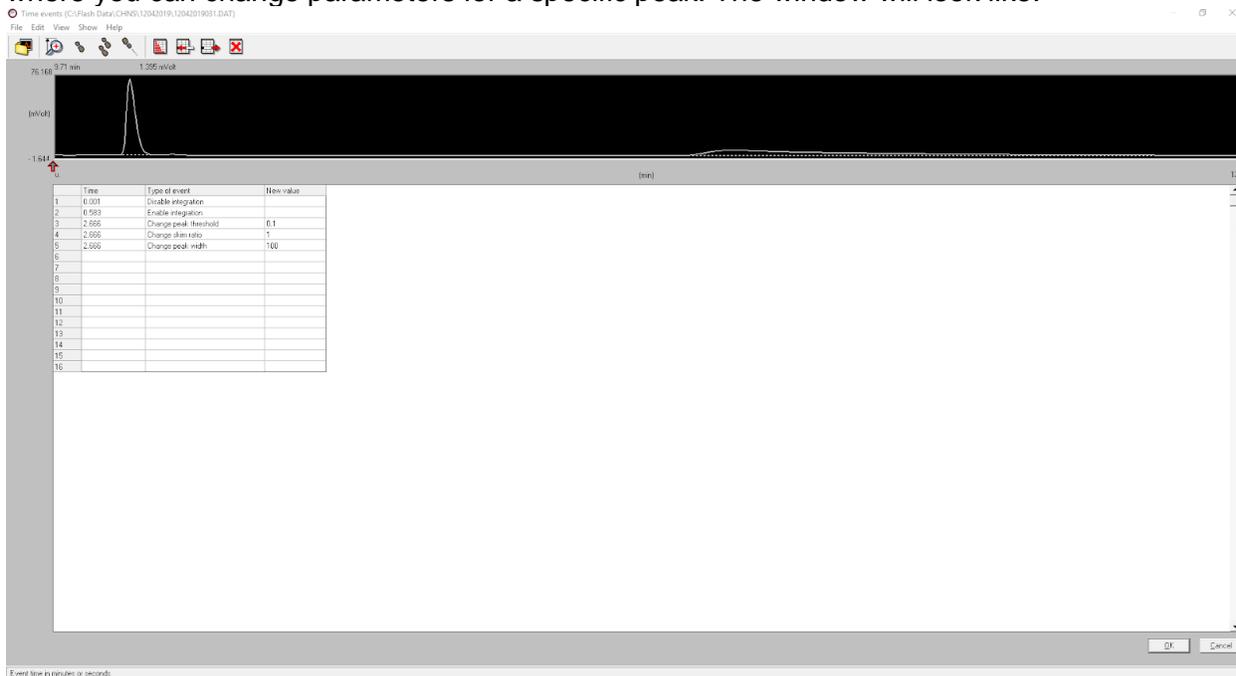
Most likely in this window, you will be using the “Test Integration” button. By clicking on this, you open:



From here, you can load your desired chromatogram (button in yellow). You can zoom in on any part of the chromatogram by clicking and dragging. You should be able to see how each peak is currently being integrated. If there is a problematic peak, you can click the “Test Integration” button that looks like a calculator (in green). You will open another window:



Changing minimum area and other parameters in the above window will change the parameters for all peaks in the chromatogram. By clicking on time events, you will open another window where you can change parameters for a specific peak. The window will look like:



The next image is of the table (zoomed in) from the figure above:

	Time	Type of event	New value
1	0.001	Disable integration	
2	0.583	Enable integration	
3	2.666	Change peak threshold	0.1
4	2.666	Change skim ratio	1
5	2.666	Change peak width	100
6			
7			
8			
9			
10			
11			
12			
13			
14			
15			
16			

You can click into the table to add lines. Drag the red arrow or type in a time to fill in the “time” column. You should pick a time before the peak you want to change parameters for, but make sure the time does not intersect with another peak before it. The type of event can be any of those listed above. If you would like to exclude a peak, you can disable integration around it. You can also change the peak width so that the integration looks for the full width of a peak. Changing the skim ratio and peak threshold will change how the software integrates but I usually just changed the peak width.

After you have changed time events settings, close out of the windows by clicking “ok.” When you are back to the main toolbar:

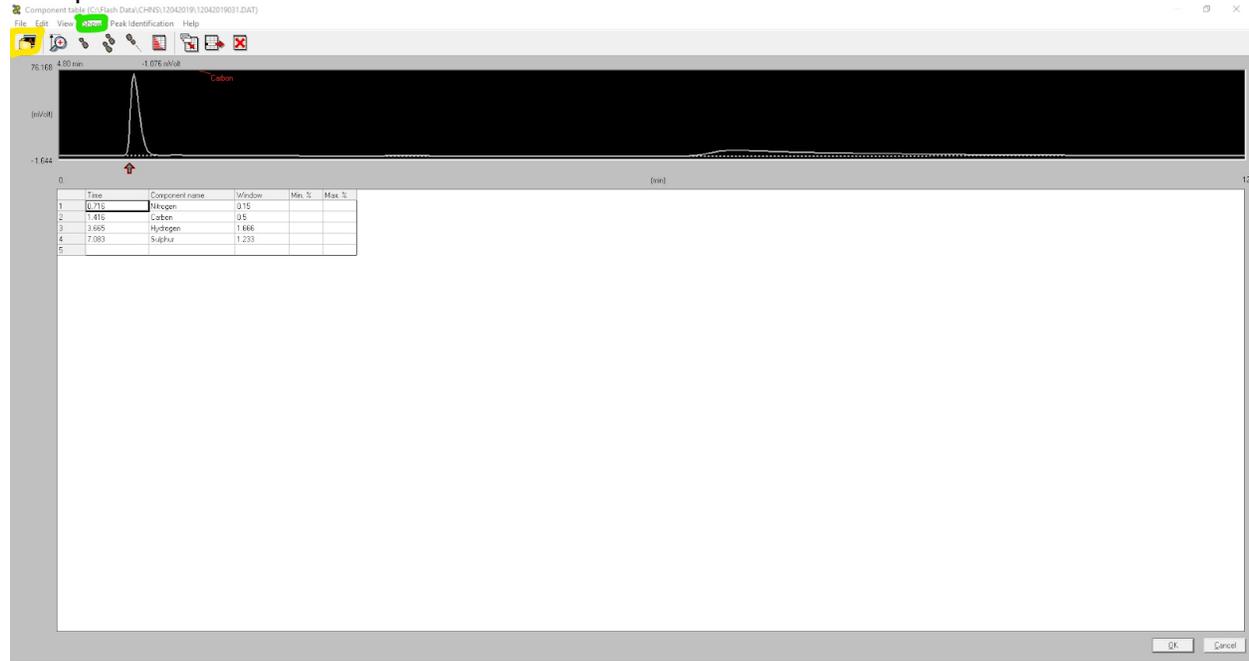


Click recalculation (not edit). Select to reintegrate and a window will pop up where you can choose to “save after integration” and then select a range of chromatograms to recalculate. If you do not do the recalculation step, then the changes you did in the steps above will not affect the calculations.

Now you can check the results table. If the component values changed favorably, then you may be done. If there was no change (or very little change) then you can try another method of adjusting the integration.

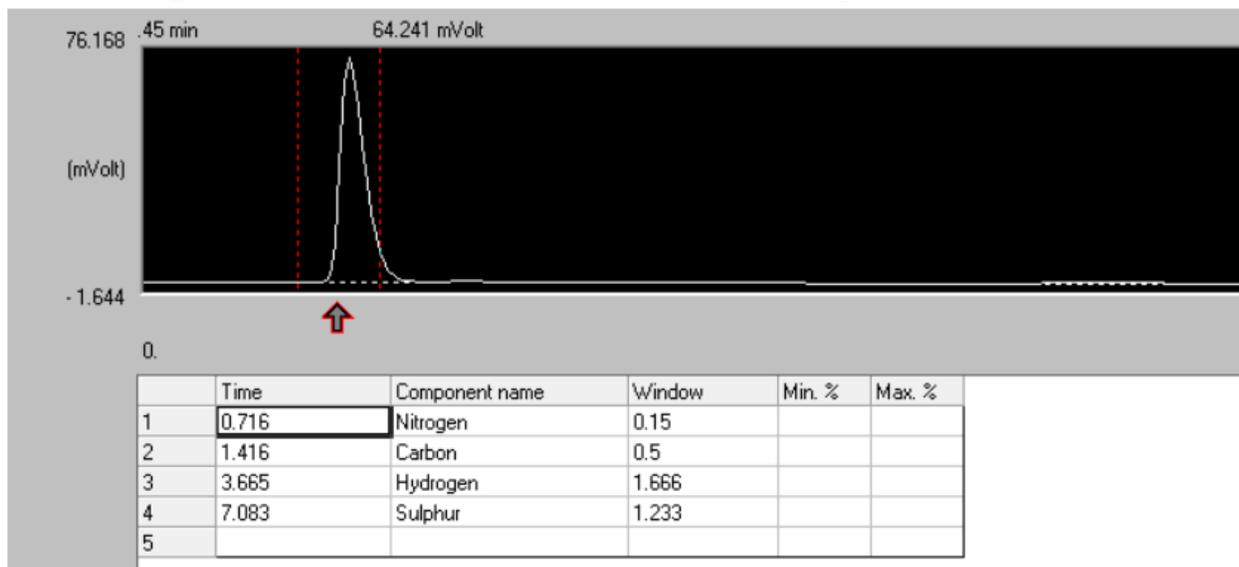
Editing the Component Table:

From the main toolbar, click on “Edit” and then “Edit component table.” The following window will open:



Component retention time in minutes or seconds

Load the problematic chromatogram (using the button in yellow) and then click “show” (button in green) to get a dropdown of show options. Click on “peak window.” This will cause red vertical lines to appear around the peaks. To move the peak window to the desired peak, click on the corresponding component in the table. It will look like the following:



The peak window is around the nitrogen peak because the nitrogen row is clicked on, which you can see by the darkened box around the nitrogen peak time. You can see in the figure that the peak window is cutting off part of the peak. To fix this, click into the nitrogen time and type in a time that is at the center of the peak. Then click into the nitrogen window and type in a number that is larger than the width of the peak. Make sure that the window does not intersect with any other nearby peaks. Additionally, you can click on any of the other components and make sure that the peak windows do not intersect.

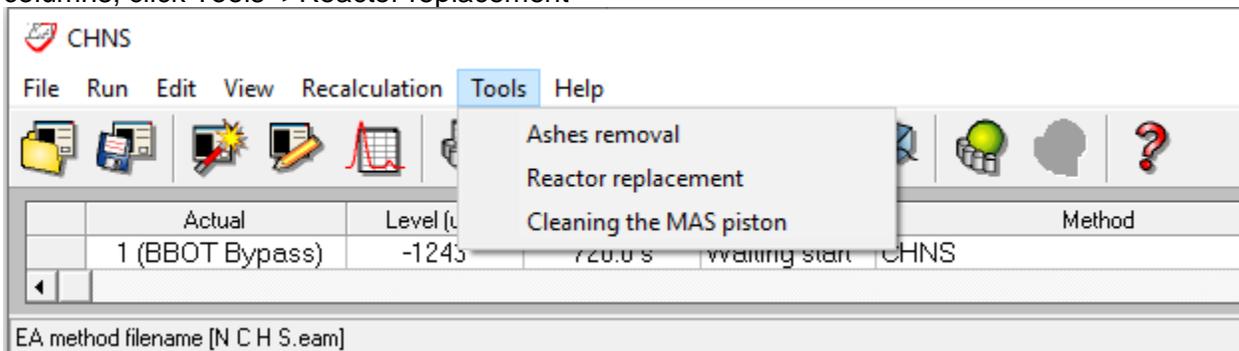
From this window, you can also reassign a peak to a component if the software incorrectly identifies the peak.

Once you have checked all the peak windows in the chromatograms, you can click “ok” to leave the window and save if it asks you to. Make sure to recalculate the results from the main toolbar and check the results to make sure the integration changed in a reasonable manner.

Maintenance

Combustion Reactor Column

The combustion reactor column must be changed every ~200 samples, because the copper in the reactor tube will become fully consumed after oxidizing the combusted samples. To change columns, click Tools=>Reactor replacement

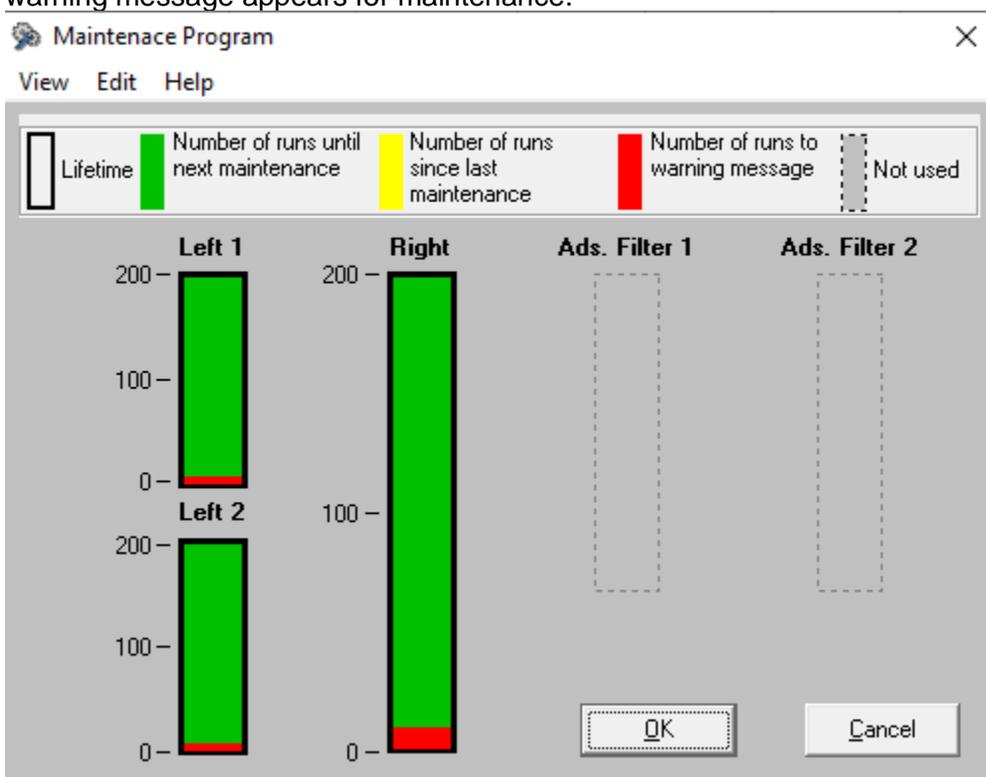


The reactor maintenance will stop the flow of gasses, turn off the filament, and cool down the reactor oven. If the instrument is in stand-by when reactor maintenance is initiated, the oven will be set to 450°C. It will take some time to cool down. It is best to wait until the reactor is ~150°C before handling. In this instance, use leather gloves to handle the reactor column. If you remove a hot reactor column, place it in the fume hood in 1238A to finish cooling down. If you do not want to handle a hot reactor column, wait until the system has completely cooled down. A used reactor column that has cooled to room temperature can be placed in the disposal box in the cupboard below the FlashSmart instrument in 1240.

To view maintenance for the reactor columns and to reset after changing the columns, press View=> View Maintenance

View	Recalculation	Tools	Help
View sample being acquired			Shift+F1
Last sample calculated results			Shift+F9
View Calibration curve			Shift+F2
View Chromatograms			Shift+F3
Overlay Chromatograms			Shift+F4
Operate on Chromatograms			Shift+F5
Compare Chromatograms			Shift+F6
View Elemental Analyzer Status			Shift+F7
View Maintenance			Shift+F8

The following window will pop up. Green represents the number of runs until next maintenance, yellow represents number of runs since last maintenance, and red is the number of runs until a warning message appears for maintenance.



To reset after changing the columns, press Edit=> Reset Maintenance Program. The following window will pop up

Reset Maintenance ✕

	Left 1	Left 2	Right	Ads. Filter 1	Ads. Filter 2
<input type="checkbox"/> Lifetime	<input type="text" value="200"/>	<input type="text" value="200"/>	<input type="text" value="200"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
 Number of runs to warning message	<input type="text" value="10"/>	<input type="text" value="10"/>	<input type="text" value="10"/>	<input type="text" value="10"/>	<input type="text" value="10"/>
 Number of runs until next maintenance	<input type="text" value="200"/>	<input type="text" value="200"/>	<input type="text" value="200"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
 Number of runs since last maintenance	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>

Hit reset for the appropriate furnace after changing the column.

Gas Cylinders

When running samples, it is important to keep an eye on the level in the helium and oxygen cylinders.



Helium



Oxygen

The left gauge reads the pressure, and the right reads to the amount left in the tank.