

SERC: Seal Analytical AQ300 Protocols

Start Up Procedures

Refill DI container with fresh DI each day.

- *Even if it is full; still get fresh everyday (Flick sides to get bubbles off)*

Make sure the waste carboy is not full and labeled correctly for hazardous waste. This should ideally be transferred to a closed top container after every use.

Turn instrument on. The power button is on the back-right side of instrument just above the power cord. There is also a lamp only switch higher up on the back of the instrument so make sure the proper switch is used.

Log onto the computer and open the AQ300 software. Do not open the software without the instrument being on. The software does not like this. Log in name is "seal", password is "toledo". A checklist will pop up.

Perform the daily check list items:

- ☐ Empty and refill wash water reservoir (should have already been done via above).
- ☐ Empty the waste container (should have already been done via above).
- ☐ Replace reaction segments as required (see details below).

In software

Maintenance & Setup Tab -> Maintenance

- *The sash can be open, but it will move, and you want to look for smooth movements*

Instrument will initialize

- ☐ Observe movements during initialization
 - o Movements should be smooth and they should be the same each initialization.

All used reaction segments should have been replaced at the end of the last run, but you can check in the "Initialize" window.

Initialize -> Zero Segments

Pop up should say all reaction segments clean, if not replace the ones it indicates.

Diluter

- ☐ Prime syringe 10 times (change number on drop down list).
 - o Verify no air remains in syringe and is operating smoothly
 - *Start primes and look for bubbles in the syringe – you should have no bubbles by the end*
- ☐ Check operation of probe washer
 - o Turn on waste pump and wash valve.
 - *Look for leaks*
 - o Verify a vortex in chamber and no water dripping.
 - o **Turn off** wash valve and waste pump.

Cuvette

- Perform 5 auto washes (change number on drop down list).
 - Observe wash bath is filling and clean.
 - *Bath is in the back; look for water on the probe*
- Check/Adjust aspiration for both inner and outer wells (run check twice)
 - Verify that there is 1-2" of water (no bubbles) in the outlet tubing of cuvette.
 - *You don't want any bubbles close to the cuvette, but further up the tubing is okay.*
 - **REMEMBER TO PUT THE COVER BACK ON THE CUVETTE!!!!**
 - If you need to adjust, change the value in increments of ± 5 . This number is essentially pump rotations. You may also need to replace the tubing if large adjustments are needed OR if the pump is making a lot of noise.

Close out of maintenance window.

Make sure there is DI in reagent space 18 and cuvette cleaner in reagent space 1 then run the "Daily Startup" procedure and click continue when prompted. This will take just a minute or two. Once the small daily startup window disappears click on "print preview" and check that the gains from the last time the startup was ran (left columns) are not wildly different from current gains (right columns). If the numbers are quite different there might be a problem with the lamp and refer to the customer support manual. You can close the print preview window after comparing.

- *Replace the DI in space 18 everyday as well*
- *Cuvette cleaner is in the drawer if you need to refill it*

Running the Analyses

Click the Scheduling Tab

Choose a free tray and choose what reagent tray is appropriate for the methods you are running.

When a new scheduling tray appears click "Show stds/ctrls". Do not place any samples in the red highlighted rows.

You can now type in or import your samples into the sample ID column. You should insert a dup and spike every 10 samples. To do this highlight the 2 rows after the 10th sample. Click cup number pull down and click on same cup number as sample. This will assign these rows the same cup as your dup/spike sample. Type "dup" in 1st type column and "spka" in second type column. Repeat every 10 samples.

Once all your samples are entered, highlight all the rows (software won't allow you to scroll down and select so you may have to highlight a few times). Click the analysis you want run on these samples from the test box. Save.

Be sure that all reagents are in their proper locations on the reagent tray. You can check proper locations in the Maintenance tab -> Test -> Reagent Tab

- *If you are running phosphate, make sure you made fresh ascorbic acid (this needs to be made every day)*

Be sure standards and controls are also in the correct cup or reagent.

Be sure lid is closed

Double click "Run" and select the tray to begin the analysis.

- *You want to select Auto Calibration to do standard curve*
- *Turn on auto turn lamp off – especially if you have a long run into the evening.*

Data Results

Once run has finished:

Tray Manager -> Select correct tray - > Tray Log -> Ascending order -> Copy in Excel Format

Open Excel and paste data.

- *Column F: Concentration*
- *Column G: Absorbance*
- *Column I: Dilution*
- *ALSO: RECORD the slope, intercept, and R2 for the standard curve on the QA QC log for the analysis you are doing.*

Shut Down Procedure

Remove sample cup tray from instrument and dispose of sample properly. Plastic cups can go in trash once emptied.

Make sure DI is placed in Reagent 18 slot and cuvette cleaner is placed in Reagent 1 slot.

Close lid.

Maintenance & Setup Tab -> Maintenance -> Cuvette -> Extra Wash

Extra wash will take a few minutes.

Cap and place reagents in fridge.

Remove used reaction wells and rinse and dispose of properly (hazardous waste). To see which reaction wells to replace:

Initialize -> Zero Segments

Pop up should indicate which wells to replace with new ones. Check that new wells are free from dust & dirt.

Click "Ok" once they are replaced.

Close out of maintenance window and close out of software.

Turn power off to instrument from back button just above the power cord.

COMPASS SEAL Notes:

General Notes:

- There is a USB on the table that has all the protocols and software on it if you ever need it.
- NH₄/NH₃ concentrations can be affected by freeze/thaw so you want to run these when you thaw the samples – prioritize this if you can only run one thing after thawing a set of samples.
- If you run samples for NO_x first, you can just top off the samples and run them for NH₄/PO₄ directly after to save time pouring
- If you have samples that seem to have particles in them, you need to refilter into the sample cup with a 0.45 µm filter – please make a note of this in the sample log.
- If the sample is brown/grey in color, you may need to dilute it
 - o First step is running it with an auto spike to check that it clears this
 - o You can add a dilution line to this sample in the software if needed
 - o *The machine should auto blank every sample so this should not usually be an issue.*

Sample Naming Scheme & File Naming Scheme:

- **PW Samples** should be named following: Site_YYYYMM_ZONE_LYSA/B/C_Depthcm
 - Ex. GWI_202208_LysA_10cm
 - Ex. GCrew_202208_LysC_45cm
- **Rhizon Samples** should be named following: Site_YYYYMM_RHZ_ZONE_SF_#
 - Ex. MSM_202208_RHZ_TR_SF_1
 - Ex. MSM_202208_RHZ_UP_SF_7
- **Files** should be named following: SEAL_COMPASS_Synoptic_Analysis_MonYYYY_#
 - Ex. SEAL_COMPASS_Synoptic_NO_x_Aug2022_2
 - Ex. SEAL_COMPASS_Synoptic_NH₃_PO₄_Aug2022_1
 - Ex. SEAL_COMPASS_Synoptic_NH₃_PO₄_Aug2022_2

NO_x Vanadium Run:

- Top Standard = 1ppm; Sample Cup #1
- Cup #2: Select REFF
 - o Cup 2 will be Nitrate reduction efficiency check
 - o Cup 3 will be Nitrite reduction efficiency check
- Cup #4: can start samples, but don't
- Cup#5: start samples here because then you can run the same set for NH₃/PO₄
- Run 3rd party check: peCheck 706ppm Nitrate
- Run Matrix Effect Checks
 - o Duplicate 10ppt artificial seawater with auto spikes
 - o Duplicate 20ppt artificial seawater with auto spikes
- CCV/CCBs will be run automatically every ten samples so you don't need to put those in
- Run spikes (SPKA) and duplicates (DUP) every couple samples (right now doing 3, but could do 6 or 9 later)

NH3 / PO4 Run:

- NH4 Top Standard = 2ppm; Sample Cup #1
- PO4 Top Standard = 0.3ppm; Sample Cup #2
- Cup #3: Leave blank
- Cup #4: Leave blank
- Cup #5: start samples at this cup
- Run 3rd party check: peCheck 908ppm NH4 and XXXppm PO4
- Run Matrix Effect Checks
 - o Duplicate 10ppt artificial seawater with auto spikes
 - o Duplicate 20ppt artificial seawater with auto spikes
- CCV/CCBs will be run automatically every ten samples so you don't need to put those in
- Run spikes (SPKA) and duplicates (DUP) every 10-12 samples

Matrix Effect Artificial Seawater:

- 10 ppt artificial seawater
 - o 10 g NaCl
 - o 0.2 g NaHCO3
 - o In 1000mL DI water
- 20 ppt artificial seawater
 - o 20 g NaCl
 - o 0.2 g NaHCO3
 - o In 1000mL DI water