QAM-I-120

# Operation and Calibration of the SEAL AQ300 Autoanalyzer

Revision 0



Approval:

Laboratory Manager — Date: 6-30-21

Concurrence — Date: 06/30/2021

Effective date: 6-30-21

Renewal date: _____ Initials: _____

*Texas Institute for Applied Environmental Research*

## 1. Applicability and Purpose

This procedure applies to the operation and calibration of the SEAL AQ300 Autoanalyzer. This instrument is designed for use in the analysis of ammonia-nitrogen ($NH_3$), nitrate ($NO_3^-$)+nitrite ($NO_2^-$) nitrogen, orthophosphate (-$PO_4^{3-}$) phosphorus, and cadmium (Cd VI) in soil extracts. It also provides checklists for conducting daily, weekly, monthly, and semi-annual monitoring and maintenance of the SEAL AQ300 Autoanalyzer.

## 2. Purpose

The purpose of this procedure is to provide a method of operation and calibration of the SEAL AQ300 Autoanalyzer. By utilizing appropriate chemistry SOPs to perform the standard calibration for each set of samples, the analyst eradicates anomalies due to reagent mixture variations, sample preparation variations, and instrument sensitivity fluctuations. The operation of the instrument allows the analyst to determine the concentration of various chemical species which are in extracts analyzed by the TIAER chemistry laboratory.

## 3. Definitiions

a. Standard QA/QC definitions are found in QAM-Q-101, "Laboratory Quality Control".

## 4. Equipment, Reagents, and Standards

4.1 Equipment

4.1.1 Seal AQ300 Autoanalyzer
Seal Analytical: 6501 West Donges Bay Road, Mequon, Wisconsin 53092
Tel: 888-211-9829
Support: techsupport@seal-us.com

4.1.2 SEAL AQ2 software

4.1.3 SEAL sample cups

4.1.4 SEAL reagent wedges

4.1.5 SEAL reaction trays
4.1.6 Pump tubing
4.1.7 Assorted glassware, Class A, used in the preparation of reagents and standards

4.2 Reagents
4.2.1 Cuvette cleaning solution
4.2.2 Deionized (DI) water
4.2.3 Household bleach
4.2.4 For additional reagents, refer to the individual SOP for the analyte of interest.

4.3 Standards
4.3.1 For standards, refer to the individual SOP for the analyte of interest.



## 5. Procedure

a. Preparing SEAL AQ2 software for conducting analyses.
   i. Turn on the SEAL AQ300 Autoanalyzer at least 30 minutes prior to use, making sure that switches for both the motor and the fan are on. Both toggle switches are located on the back right of the machine.
   ii. Attach the USB cord to the SEAL AQ300 Autoanalyzer and a labtop computer on which the SEAL AQ2 software program has been downloaded.
   iii. Log onto the SEAL software
   iv. Navigate to the “Maintenance and setup” screen.
   v. Click on the “Test” button.
   vi. Select the test to be conducted from the list on the left.
   vii. For each selected test (e.g. $NH_3$ or -$PO_4^{3-}$), click on the various tabs associated with the test of interest to ensure that reagent, blank, standard, diluent, calibration data, wavelength, and reaction time are entered according to the information provided in the SEAL methodology for that test.
   viii. The software provides each test with a number. Note the number of the test listed on the main parameters tab and on

the calibration tab. This number will be critical to use in listing the reagents to be used in the analysis In the “Reagents” and “Reagent Set” tables.

ix. Assign reaction wedge locations for each of the reagents, standard, diluent, wave length, and reaction time in the test screen. Note: the blank and diluent are usually both the extraction solution. The CCB is DI water.

x. On the “Calibration” tab, ensure that the auto standard concentration is consistent with the value listed in the analyte SOP. Also, ensure that the wedge listed as the “auto standard” contains the concentration listed on the parameter list provided in SEAL methodologies. Place the wedge in location listed on the test parameter tab.

xi. In the “QC Pro” tab, list the CCV test first and the CCB test second. The CCV test concentration should be half the concentration of the standard concentration, which is the analyte concentration at the upper limit of detection for the analysis. The value for the low limit should be 0.5 mg/L below the value of the CCV while the high limit should be 0.5 mg/L above the value of the CCV. Place the CCV wedge in the reaction tray in the location listed on the test tab. The CCB is DI water. The low limit, value, and high value of the CCB should be < 3 x limit of detection. Place the wedge containing DI water in the location listed on the test tab.

xii. On the “Reagent” tab, click on the reagents button on the left side of the window and enter the names and locations of all the reagents, standards, diluents, CCV, and CCB solutions that will be used in the analysis. Enter the location number for each wedge in the column corresponding to the number of the test to be run.

xiii. Do not assign a standard or reagent to the wedge #1 position. The cuvette cleaning solution should be placed in this position but should not be listed as part of a methodology.

xiv. Close the “Tests” window and click on the “Reagent Sets” button. All reagents that were entered in the assessment column in the reagents table should show up in one of the tabs with various reagent set names within the reagent set table. The reagent set name should correspond to the name of the test to be conducted.

xv. If more than one test is to be conducted on the same samples, click on the test order tab and ensure that the correct order is listed for the assessments. If nitrogen tests are to be run at the same time, $NH_3$ must be run prior to running $NO_2^-$ and $NO_3^-$.

xvi. Make sure that all wedges are placed in the proper location in the reaction tray and that wedge volumes are recorded appropriately.

xvii. Double check to ensure that the location number for each solution being used is consistent between the reagent tab and the reagent table tab. Also, ensure that all solutions to be used in the analysis are listed and that no other solutions are listed in the test column.

b. SEAL instrument scheduling and start-up.

i. Once all the information required for defining the test to be run is entered correctly according to step 4.a, click on the “Scheduling” button. The “Scheduling” button is located both on the ribbon at the top of the screen and with the icons to the left of the screen.

ii. A “tray” pop up window will appear. In this window, select a free tray and then select a reagent set with the same name as the reagent set selected during step 4.a.vi.

iii. The default name for the tray is the date of the analyses. The name of the tray can be changed either on the tray selection tab or in the box under tray name on the scheduling screen.

iv. Once the tray and appropriate test have been selected in the pop-up window, the “Scheduling screen” will appear. On the

"Scheduling screen", ensure that the name of the test to be run is present in the "Available tests" box. If it is not present, go back to the "Tests" and "Reagents" tabs and tables to ensure that all solutions to be used are properly entered and that the wedges corresponding to each solution are in their proper place in the machine tray and are listed in the same wedge position on each of the screens. Also, make sure that the correct reagent set to be used in the analyses is listed for the tray selected.

v. The "Scheduling screen" will be used to list all standards, controls, and samples to be analyzed along with the analysis(es) to be conducted on each.

vi. Standards or controls can be conducted prior to sample analyses by pouring aliquots of standard or control solutions into sample cups that are placed in the first holes of the sampling wheel prior to placement of the samples. In the "Type" column on the scheduling sceen, type "S0", "S1", etc. for standards or "C1", "C2", etc. for various controls, such as CCV or CCB.

vii. To analyze samples, type "U" into the column labeled "Type" on the scheduling screen. Add the name of the unknown to be analyzed in the in the column labeled "Sample ID". If additional information about the sample needs to be provided, such as differences in concentration, enter this information into the column labeled "Sample Details".

viii. To select the analysis(es) to be conducted on each sample,place the cursor in the box under the column labeled "Requested Tests". Click on the box to highlight the box, then move the cursor to the test to be conducted as listed in the "Available Tests" box. Click on the test. This will enter the name of the test into the "Requested Tests" box. Requested tests can be added rapidly to multiple samples by highlighting several boxes using control + shift and then clicking on the name of the test to be used in the analysis.

ix. When all schedule information has been entered, click “Save” on the ribbon. The “Save” button is only visible on the scheduling screen and not on the maintenance and set-up screen.
x. Once the list of samples has been developed on the scheduling screen, obtain samples and standards to be analyzed from the refrigerator. Arrange the samples on the counter near the SEAL AQ300 in the order to be analyzed as listed on the scheduling screen. Also, set out the bag of sample cups and a beaker to collect waste.
xi. Fill a sample cup with the sample to be analyzed. Pour out the sample into the waste beaker and refill the sample cup. Place the sample cup in the hole on the sampling wheel labeled “1”. Fill a second sample cup in the same manner with the same sample to provide a duplicate analysis. Place this sample cup in the hole in the sampling wheel labeled “2”. Repeat this process until all samples listed on the scheduling screen are appropriately placed in the instrument.
xii. As samples are added, double check the cup position in the instrument to ensure that it is the same as the number of the sample listed on the scheduling screen.

c. SEAL instrument pre-run maintenance
   i. Once the machine has been turned on for more than thirty minutes and the machine is connected to the computer through the USB cord, click on the maintenance and set up tab.
   ii. Empty the DI water bottle, rinse with dilute bleach, rinse twice, and then add DI water to the DI water bottle and reattach the black top and the large siphoning mechanism.
   iii. Make sure that the waste bottle is emptied and that the cap with the many small hoses entering through the cap is attached.

iv. Place cuvette cleaning solution into a wedge and place this wedge in position 1 of the reagent tray.
v. Replace any used reaction segments on the reaction wheel.
vi. Click on the “Maintenance” tab in the “Maintenance and Set Up” section of the ribbon. Within this pop-up window, click on “Initialize” to properly align all components. Then, click on zero segments once all used reaction segments have been replaced.
vii. To check alignment, click on “set up”, then click on “start”, then go to “Maintenance – Setup” and then navigate to the “Sampler Test” tab. In the upper Horizontal Tests section, select the option Sampler over Reagent from the dropdown menu. Observe the alignment of the sample probe over the reagent wedge and make sure it is positioned over the cap. Adjust if necessary. Complete this test by clicking on Sampler return to horizontal.
viii. Click on cuvette function and conduct extra wash, test aspiration, and 5 autowashes.
ix. Click on diluter and conduct 5 - 10 primes
x. Prior to starting to run samples, click on “Daily Startup” to run primes and water baselines.
xi. After water baselines are run, return to the “Maintenance and Setup” screen and click on “Spectro”. This will produce a screen showing the accuracy for wavelength readings. If information on this screen shows that any of these readings are out of range, either the lamp filter needs to be cleaned or the lamp replaced.
xii. Record on the maintenance data sheet (see appendix) the completion of the above maintenance steps.

d. Conducting a sample run
   i. A sample run can be initiated after all tests, schedules, and start-up procedures have been conducted.

ii. Double click on the “Run” icon on the top left and select the name of the tray and the analysis method to be conducted.
iii. A pop-up window will appear asking whether standards and baselines should be run. Click to select both.
iv. Another pop-up window will appear showing the standards and diluent to use in conducting the standard curve. Click yes if all the information is correct.
v. Once the tray and analysis method has been selected, the “Scheduling screen” will appear. Check to ensure that all the information is listed as intended for the analyses. The analyses will commence, with the standard curve conducted initially. The standard curve will be developed automatically by the instrument based on pre-programmed levels of diluting the “standard solution.”
vi. Once the standard curve has been developed based on the running the standards, click on “Calibration” to ensure that the standard curve has a high $R^2$ value (>0.95) and that the values for each of the standard readings are within 5% of the expected values. If the values are not as expected, click on “Stop”, then “Orderly stop”. Review all the preparation steps and the machine setup and make appropriate changes prior to proceeding.
vii. If everything has been set up properly, the analyses will run automatically. The instrument will shut off and the lamp will turn off automatically at the end of the run.
viii. After the run has completed, wash the cuvette using the extra wash command from the “Maintanence and Setup” tab, empty the waste and DI water resevoir containers, discard used sample cups and reaction segments, and put the reagent tray in the refrigerator.
ix. To download or print results from the run, click on the “Reporting” tab. Select the format for the report and the information to be included in the report, and the trays to be included in the report. Report information can be in the form of

raw data or in the form of a formal report with logo, standard curve, CCV, and CCB data.

5. Weekly, monthly, and semi-annual maintenance
   a. Weekly maintenance
      i. Compare water baseline voltages to monitor lamp stability. If voltages drop suddenly (within 2 weeks) by more than 1V, the lamp may need to be replaced.
      ii. Clean sample wash bottle using bleach followed by rinses with DI water.
      iii. Clean the aspiration wash bath
      iv. Inspect the sampler and aspiration probes for looseness or any damage.
      v. Record on the maintenance data sheet (see appendix) the completion of the above maintenance steps.

   b. Monthly maintenance
      i. Replace the aspiration pump tube – ensure that water is being pulled through the cuvette. Optimize cuvette aspiration pump steps and check cuvette aspiration for both inner and outer reaction wells.
      ii. Clean/lubricate aspiration pump rollers (one pump tube)
      iii. Replace aspiration wash pump tubes – ensure that water is being pumped to the wash bath.
      iv. Clean/lubricate aspiration wash pump rollers (two pump tubes)
      v. Bleach wash water reservoir (2% solution)
      vi. Remove and clean fan air filter near cuvette
      vii. Run and record in the maintenance log results of measurement diagnostic test
      viii. Record on the maintenance data sheet (see appendix) the completion of the above maintenance steps.

   c. Semi-annual maintenance

i. Replace syringe assembly with O-ring.
ii. Clean/lubricate syringe screw drive.
iii. Replace lamp and optimize the water baselines using gain and offset.
iv. Replace reagent containers (wedges) and caps
v. Replace probe washer
vi. Inspect cuvette inlet and outlet tubing
vii. Inspect sampler and aspiration probes – straight, clean, not dripping. Replace if damaged.
viii. Record on the maintenance data sheet (see appendix) the completion of the above maintenance steps.

d. Maintenance documentation
   i. Record, date, and initial all inspection observations, part replacement, and instrument lubrication in the instrument maintenance log (I-120-1).



## 6. Quality and Control Safety Aspects

**a.** All aspects of this procedure comply with QAM-Q-101, “Laboratory Quality Control” and QAM-S-101, "Laboratory Safety".

**b.** Safety
   **i.** The specific method being used may require use of hazardous substances. Waste is handled in accordance with QAM-W-101, "Disposal of Laboratory Waste".
   **ii.** Gloves, impact resistant safety glasses, and a lab coat are worn as needed during phases of this procedure.

c. Quality Control
   i. The precision and accuracy of the sample measurements are dependent upon the establishment of a calibration curve > 99%. To ensure accurate measurements of the analyte, all QC standards and samples being analyzed are treated

exactly as the blank and calibration standards during all phases of this procedure.

ii. To test the precision, accuracy, and sampling range of this instrument, analyses will be conducted at least semi-annually using samples that include standards from at least two different reagent sources and at concentrations representing the extremes of the sampling range.

iii. All data that do not meet quality requirements are handled in accordance with QAM-Q-105, “Corrective Actions.”

iv. All data are recorded in personal logbooks or E-logs in accordance with QAM-A-102, "Document and Data Control".

v. All instrument maintenance is documented in accordance with QAM-Q-103, "Equipment Maintenance".

## 7. References

a. Seal Analytical. 2017. AQ Software – Operations Manual. Publication No.: MB7-84EN-03. Rev. 1A. www.seal-analytical.co
b. Seal Analytical. 2017. Instrument Use Manual.
c. Seal Analytical. 2017. Instrument Maintenance Manual.
d. Seal Analytical – AQ2 Software

## 8. Attachments

8.1 Example SEAL AQ300 Maintenance Log

# Appendix1

## Example SEAL AQ300 Maintenance Logbook

| SEAL AQ300 Maintainance Logbook Daily Tasks | | | |
|---|---|---|---|
| Date | Initials | Task | Comments |
| | | Empty and refill wash water reservior | |
| | | Empty the waste container | |
| | | Replace reaction segments as required | |
| | | Observe movements during initialization: Movements should be smooth and the same with each initialization | |
| | | Prime syringe 5-10 times; verify no air remains in syringe and operating smoothly | |
| | | Check operation of probe washer: Turn on waste pump and wash valve; verify water movement through chamber | |
| | | Perform 1-5 auto washes of the cuvette; observe that the wash bath is filling and clean | |
| | | Check /adjust aspiration for both inner & outer wells: verify that there is 1-2" of water (no bubbles) in the outlet tubing of cuvette | |
| | | Run "Daily Startup" procedure: Water baseline voltages for filters 1-9 should range from 0.7 - 4.9 V. Filter 10/Dark should be near 0.03 V. | |
| | | Inspect Reagents for particulates or exessive color. Filter or replace reagents as needed. Check the method documents for information on reagent stability and symptoms of reagent degradation. | |
| | | Cadmium coil checks as required. See cadmium coil care section of "Technical tips" in the Customer Support Manual. | |

| SEAL AQ300 Maintainance Logbook Weekly Tasks | | | |
|---|---|---|---|
| Date | Initials | Task | Comments |
| | | Record and compare water baseline voltages to monitor lamp stability. If voltages drop suddenly (within 2 weeks) by more than 1V, the lamp may need to be replaced. | |
| | | Clean sampler wash bath | |
| | | Clean aspiration wash bath | |
| | | Inspect Sampler Probe -- straight, clean, not dripping | |
| | | Inspect Aspiration Probe -- straight, clean, not dripping | |

| SEAL AQ300 Maintainance Logbook Monthly Tasks | | | |
|---|---|---|---|
| Date | Initials | Task | Comments |
| | | Replace aspiration pump tube --ensure that water is being pulled through the cuvette | |
| | | Optimize cuvette aspiration pump steps and check cuvetter aspiration for both inner and outer reaction wells | |
| | | Clean/lubricate aspiration pump rollers (one pump tube) | |
| | | Replace aspiration wash pump tubes --ensure that water is being pumped to the water bath | |
| | | Clean/lubricate aspiration wash pump rollers (two pump tubes) | |
| | | Bleach wash water reservior (2% solution) | |
| | | Remove and clean fan air filter near cuvette | |
| | | Run and record results of measurement diagnostic test | |

| SEAL AQ300 Maintainance Logbook Semi-Annual Tasks | | | |
|---|---|---|---|
| Date | Initials | Task | Comments |
| | | Replace syringe assembly with O-ring | |
| | | Clean/lubricate syringe screw drive | |
| | | Replace lamp and optimize the water baselines using gain and offset | |
| | | Replace reagent containers (wedges) and caps | |
| | | Replace probe washer | |
| | | Inspect Cuvette Inlet and outlet tubing | |
| | | Inspect sampler probe -- straight, clean, not dripping | |
| | | Inspect aspiration probe -- straight, clean, not dripping | |