Startup protocol for the Horiba AquaLog Version 1.8 January 2022

Necessary files are located on the instruments computer. Important shortcuts are on the user accounts desktop toward the right side of the screen. Important shortcuts:

- AquaLog V4.2: The instrument software
- AL Data: The folder where all measurement data is to be stored
- FluorometerCheck: Shortcut to an app used for lamp and cuvette checks
- AL lamp hour tool: Shortcut to a tool that allows us to see the lamp hour count of the instrument.

Important locations on the hard drive:

- D:\AL Data: The folder where all data is to be stored.
- D:\AL startup routines: Contains the method files to be loaded for daily checks.

Prior to experiments

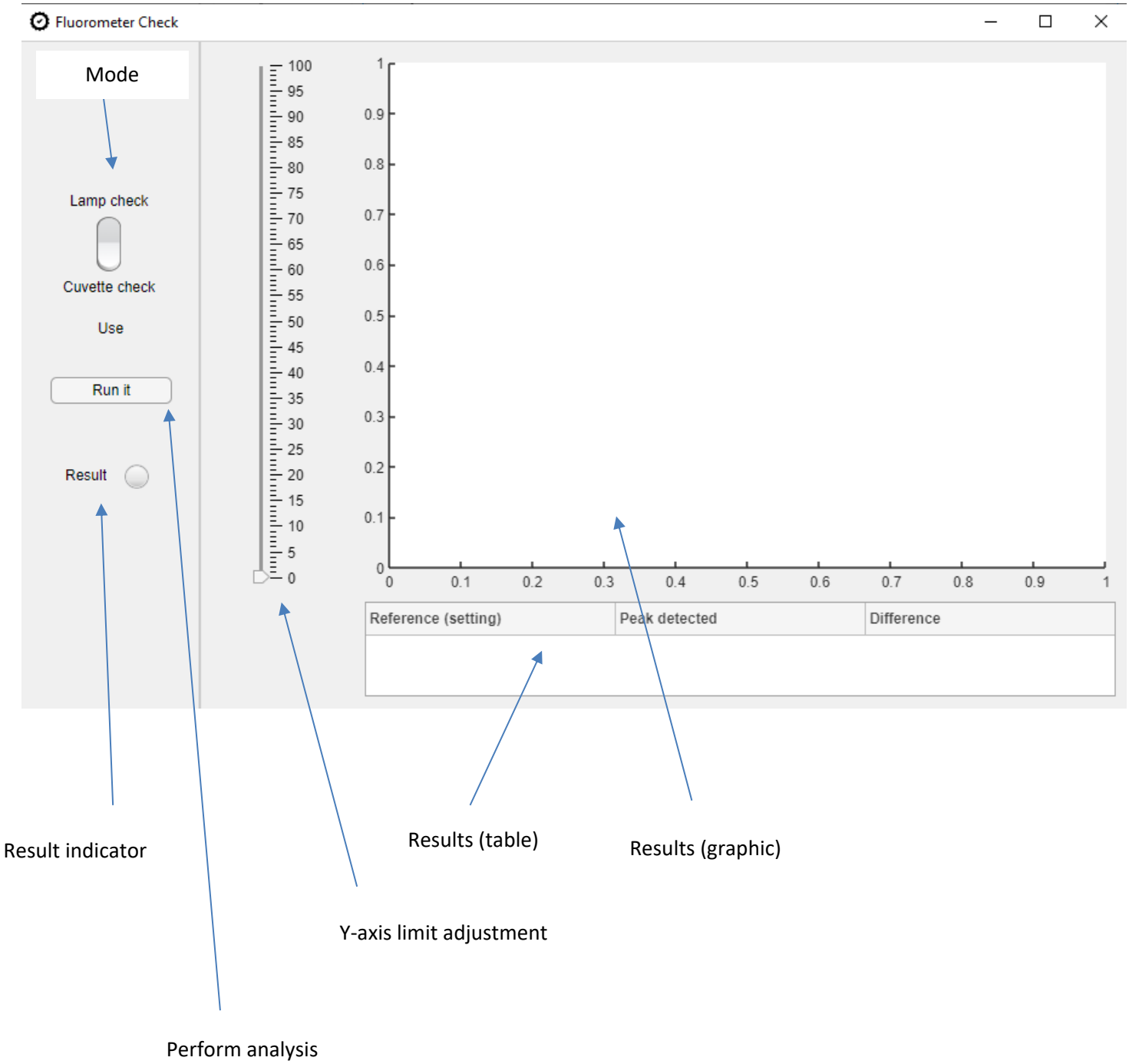
- Make sure your measurement cuvette is properly cleaned and has no scratches and cracks.
- Either make sure to use gloves while handling the cuvette, or be sure to touch your cuvette only in places where no contamination will occur as a result.

Aqualog startup procedure

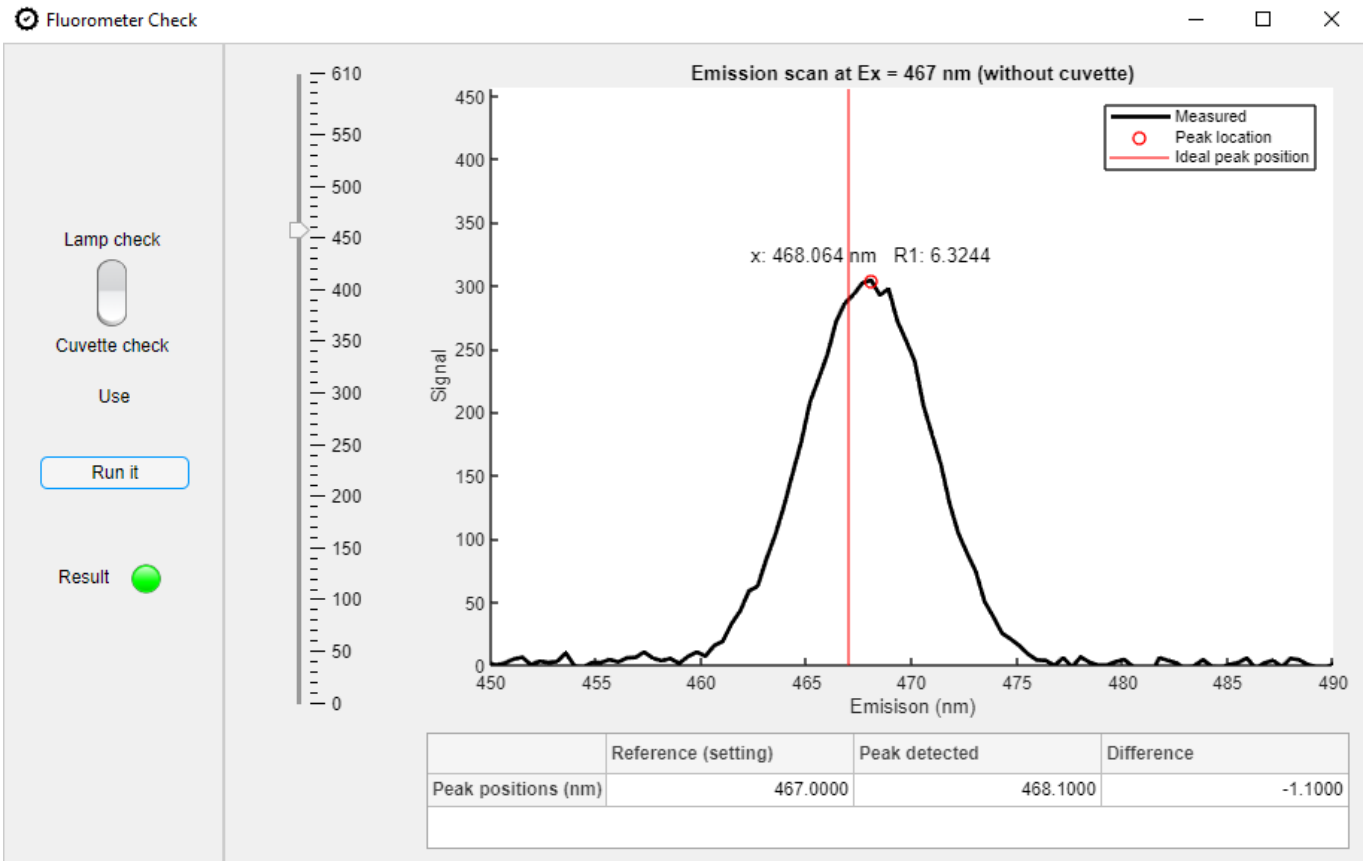
- Turn on the instrument (Power switch is on the left side of the instrument)
- Wait for 10s and turn on the Aqualog computer (if it's not already on)
- Start the software (Shortcut 'Aqualog' on the desktop)
- Open the *Aqualog Main Experiment menu* (blue symbol 'H2O' in the toolbox) to connect to the instrument
- Wait for the warmup of the Xe lamp (at least 45 minutes) before continuing with any measurements of samples. You may perform the daily checks immediately.

Daily instrument checks (2)

Start the app “FluorometerCheck” by double-clicking on the cuvette icon on the desktop. After the app has loaded, leave it at its default settings and carry on.



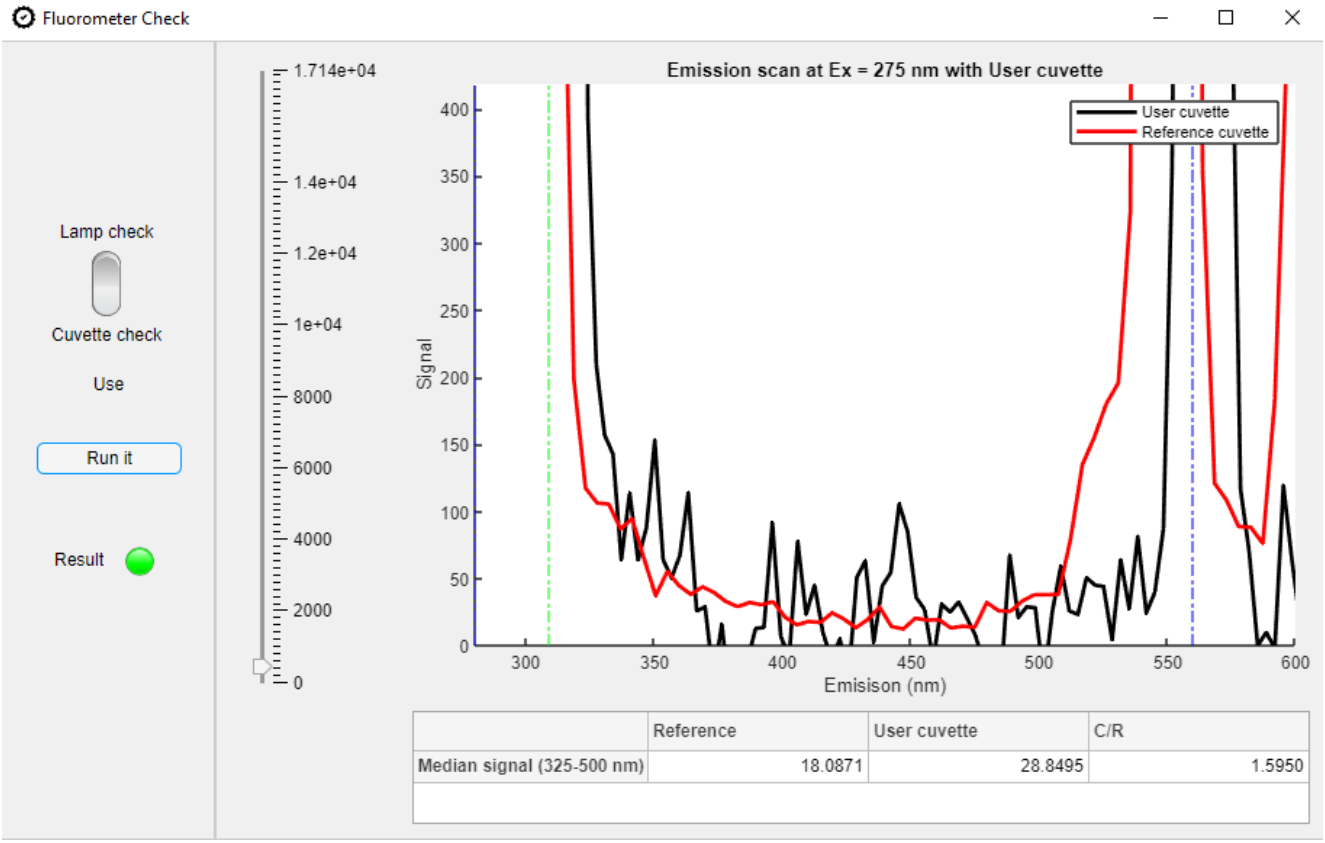
1. **Xenon lamp & monochromator check**
   - Leave the cuvette holder empty for this check
   - In the *Aqualog Main Experiment window*, click on *Spectra*, and select *Emission 2D*. In the top left field (*Experiment*), click Load, and navigate to 'D:\AL standard routines' and load the method *Lamp_Ex_Check_V2.xml*
   - Click Run and wait for the experiment to finish. You should see a graph with a flat baseline and a clear peak at around 460nm. Select the tab *Emission Sample Data* and the lower part of the experiment window. Copy the entire spreadsheet *A(X)* to *N(Y)* by clicking on the empty column space to the left of "A(X)" and above "Long Name". Press "Ctrl + C" to copy the data to the clipboard.
   - With the scan data now in the clipboard, switch to the app FluorometerCheck. The default settings need not be changed. Just click "Run it."



   - Once the graph is visible, a summary is placed in the clipboard. Paste the values into the instrument log (shortcut on the desktop) in columns C and D.
   - The first value should be 467 ±1-2nm. If out of range, write email to urbw@dtu.dk, but continue your experiment. Passing of this criterion will be indicated by a green light on the left. A red light indicates that something is wrong.

2. Cuvette check for contamination

- Rinse your measurement cuvette with plenty of water to rinse out any contamination.
- Place our measurement cuvette into the cuvette holder. Inspect the cell and make sure it's clear.
- Open the *Aqualog Main Experiment window* again and select *Spectra*, then *Emission 2D*. In the same directory as before, open the method file *Cuvette_check_V2.xml*. Run the method and wait for the results.
- Change from the *Emission Graph* tab to the *Emission Sample Data* tab. Copy the entire spreadsheet *A(X)* to *N(Y)* clicking on the empty column space to the left of "A(X)" and above "Long Name" and pressing the Ctrl key on the keyboard and then pressing Ctrl+C.
- Switch to the FluorometerCheck app and flip the switch to "Cuvette check". Then click "Run it". The result should look similar to this:



- Once the graph is visible, a summary is placed in the clipboard. In the instrument log, paste these in column E and F and the row corresponding to the day.
- To proceed, the following criteria should be met (the app indicates this with a green or red light):
  - Average signal reported for your cuvette is <2 (calculated in the excel worksheet).
  - The emission region between 1st and 2nd order Rayleigh and Raman scatter (350-500 nm) does not show signs of a broad contamination peak.
- If the one or both criteria are not met, rinse with ultrapure water repeatedly, polish cuvette with lens paper to remove potential seawater residue (inner and outer cuvette window).

Running samples

- Before running any blanks or samples, let the instrument warm up for one hour! The lamp needs to stabilize its output and the monochromator (next to the lamp) needs to equilibrate to the increasing temperature.
- Run samples with the desired EEM method (ask for a method file if unsure). Run the sealed cuvette as blank first (blank only) and a MilliQ afterwards (sample and blank, blank from file). Do not change sample names after running a sample (naming inconsistencies WILL occur).
- Always check that all four sides of your cuvette are clear and no bubbles appear inside the cuvette.
- After approx. 5-10 samples, the software slows down notably. Just start a new project (before you close the old one, consider performing the first step under "Finishing up").

Finishing up

- In the main window menu, click File, then HJY Export. Click on the … button and select the entire folder, or single graphs as desired. Click ok, and specify the export location.
- Clean your cuvette with MilliQ and store as desired.
- Note down the lamp hours by opening the desktop shortcut *AL lamp hour tool* in the instrument log. Also, note down any noteworthy occurrences in the column *Notes*.
- Close the Aqualog software and turn off the instrument. **DO NOT** shut down the computer.