Best Practices for Shipboard Underway Transmissometers

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1. Introduction

This best practice document was developed under the Rolling Deck to Repository project by a small working group comprised of the document authors. These recommendations are specifically for transmissometers installed within a research vessel's (RVs) scientific flow-through sea water (underway) system. They are not focused on transmissometers used for CTD/Rosette casts, though some information may be applicable. All underway transmissometers in the academic research fleet presently are WET Labs (henceforth Sea-Bird) C-Stars. Review of available sensors shows this will continue to be the most available and adaptive device.

The **Goal** of this document is to provide best practices for operators that will improve access to transmissometer data and metadata to support routine data quality evaluation and science application.

2. Data to Record and Provide to Users

What do science users want from an underway transmissometer? The primary output for users is the signal voltage, but providing both the calculated transmittance ratio and beam attenuation will be beneficial to some users. Although provision of sensor counts is not likely needed by most users, recording counts as opposed to signal voltage is dictated by the hardware used. The following values are of primary interest to users:

- 1. Signal voltage (V_{sig}) or raw counts; contingent upon device configuration
- 2. Calculated Transmittance Ratio (Tr)
- 3. Calculated Beam Attenuation (c)

3. Basic C-Star calculations

Factory calibration voltage values for V_d , V_{air} , and V_{ref} and instrument path length (z = 0.1 or 0.25 m) are necessary to transform the data into meaningful values that include Transmittance Ratio (**Tr**) and beam attenuation (**c**).

Additionally, V_{airs} and V_{darks} are required ship-board measurements recorded pre and post-cruise, to track sensor stability over time between factory calibrations. These ship-board measurements may change slightly over time, however significant deviations from the factory values provided are indicative of issues with sensor health and will require the sensor to be returned to the manufacturer for repair and/or calibration. Over time, small changes in V_{airs} and V_{darks} values can be expected and this will change the **Tr** and subsequently the **c** values. Required values for transformation are defined as follows:

- V_{sig} = recorded signal from instrument in volts
- V_d = dark (closed path) value from the Factory calibration

- Vair = air (open path, no water) value from the Factory calibration
- V_{darks} = dark (closed path) value from the *Ship* in-situ calibration
- V_{airs} = air (open path, no water) value from the Ship in-situ calibration
- V_{ref} = clean water signal measured during Factory calibration
- z = instrument path length in meters (0.1 or 0.25)

Calculate the Transmittance Ratio (Tr) including ship-board measures of V_{darks} and V_{airs} :

Equation 1:

Transmittance Ratio (Tr) = $[(V_{sig} - V_{darkS})/(V_{ref} - V_d)]^*[(V_{air} - V_d)/(V_{airS} - V_{darkS})]$

Transmittance is typically reported in % rather than decimal. Provide % transmission by converting the decimal **Tr** to %.

Calculate beam attenuation (c) using Tr from Equation 1:

Equation 2:

Beam Attenuation (c) = -1/z * ln (Tr)

Tr is entered as a decimal value and beam attenuation (c) units are in m⁻¹.

Note on C-star data output: Figure 1 depicts the data output for the transmissometer, it should be noted that lines 4 and 5 provide a corrected signal and calculated beam attenuation (beam c), respectively. With regard to values during operation, observations can be made on the calculated beam c to ensure the value is within the anticipated range. When operating in the underway configuration, very high or erratic values of beam c as well as corrected signal counts are indicative of bubble issues.

Column	Value	Example				
1	Instrument serial number	CST-1916DR 11531 15780 15783 -0.087 544				
2	Reference counts	CST-1916DR 11528 15777 15784 -0.087 543				
3	Signal counts	CST-1916DR 11526 15774 15784 -0.087 543				
4	Corrected Signal raw counts	CST-1916DR 11431 00000 00000 99.999 542				
5	Calculated Beam c, inverse meters	CST-1916DR 11430 00000 00000 99.999 542				
6	Internal thermistor, counts	CST-1916DR 11429 00000 00000 99.999 542				

Figure 1: Explanation of data output from the C-Star Transmissometer.

4. Instrumental metadata

The first four metadata items listed below have been noted as being essential to properly calculate transmittance ratio and beam attenuation from the signal voltage. The

remaining items are needed by scientists or data analysts to properly quality evaluate or apply these observations to scientific activities.

- 1. Reference voltage (V_{ref}) From factory sensor calibration
- 2. Dark voltage (V_d) and open-air voltage (V_{air}) From factory sensor calibration
- 3. Dark voltage (V_{darks}) and open-air voltage (V_{airs}) From routine shipboard in-situ cleaning and calibration (See Section 5)
- 4. Beam path length (**z** in meters)
- 5. Wavelength of light used: e.g., 650nm (red), 530nm (green), 470nm (blue), 715nm (infrared)
- 6. Instrument number (serial number from manufacturer) This traces the individual device to its associated metadata (e.g., items 1, 3, and 4, and ideally 2 based on operator practices).
- 7. Technician name Who did the ship in-situ calibration?
- 8. Ideally a technician/engineering log Noting problems, cleanings, repairs, etc.

Other items that have been noted to affect readings include water temperature, salinity, pressure, and whether or not the seawater source is pre-filtered. Pre-filtered seawater provides a filtered seawater signal that can then be used to monitor biofouling of both the sensor and upstream tubing and provide a running blank for corrections. This type of measurement is typically performed intermittently by using an actuator valve that switches between a filtered (typically 0.2µm) and a whole seawater signal. While uncommon in current ship-board practices, if this occurs, the time and duration of the signal should be recorded. It is important to document how your transmissometer is installed on your ship, noting whether or not water flow into the transmissometer happens downstream of a debubbler and if a flow meter exists to monitor the water rate and/or pressure in the pipe.

5. Underway Transmissometer Cleaning and In-situ Calibration

A clean transmissometer is crucial to obtaining quality data. Biofouling of both the sensor, tubing, and the system upstream can occur rapidly, even in oligotrophic waters. If the system does not include an intermittent filtered seawater signal to monitor biofouling, other means are necessary to ensure quality data. This includes performing consistent cleaning, tracking air and dark counts as well as visually inspecting the inside of underway tubing for fouling.

Recommendations for cleaning underway transmissometers and tracking signal counts:

- 1. Cleaning optics¹
 - a. **Recommended:** Before each cruise, prior to in-situ calibration for cruise.
 - b. Weekly as needed to support science or based on environmental conditions. It is best to perform an in-situ calibration prior to cleaning and post cleaning to track any drift that may occur between cleanings.
 - c. If the ship is to be dockside for more than a week, the optics should be cleaned and left dry until the next cruise. Just prior to the cruise (after > 1 week between cruises) the optics should be cleaned and a pre-cruise insitu calibration performed.
- 2. Cleaning tubing¹
 - a. **Recommended:** After each cruise, or for long duration cruises, every two weeks.
 - b. As soon as bio-fouling is suspected (see section 5.3 Cleaning Tubing)
 - c. Sometimes operators are requested to bleach their underway system. Sensors should **never** be bleached and should be bypassed when the bleach tablet is inserted and allowed to "pickle" the system. Following a freshwater rinse, instruments may be reconnected to the underway system.
- 3. Air and Dark ship in-situ calibrations should be done regularly by operator and new V_{darks} and V_{airs} values recorded and provided to users
 - a. Recommended: Before and after each cruise
 - b. Weekly as needed to support science
 - c. Science advisory panel also noted this calibration should be done whenever the instrument is cleaned or swapped.
- 4. *V_{airS}* and *V_{darkS}* ship calibration and optics cleaning should occur whenever a device is installed/swapped.
- 5. Factory calibrations **recommended** to be done yearly as a minimum, depending on the total time of use and trends of V_{airs} calibrations. This resets $V_{ref.}$
- 6. Operator should routinely provide calibration sheet from Sea-Bird with dataset.
- 7. Sensor and tubing redundancy would help with cleaning cycle turn around.

5.1 Recommended Cleaning and Calibration Kit:

- MeOH: 100% Methanol, HPLC grade, Examples include BDH85681, JT9830-2, MX0475P-6, this solution removes any organics that may be present
 - Alternative: Isopropyl alcohol, 70% or 91% (higher percentage is recommended); a high water content will increase likelihood of tainted optic lens
- Triton: Triton X-100, Laboratory grade, VWR #97062-208

¹ Cleaning schedule will depend on the length of time a vessel remains in port between cruises.

- Use 0.1% solution: 0.1ml of Triton X-100 diluted to Milli-Q water to 100ml final volume, this solution solubilizes any proteins that may be present
- Milli-Q Water: Ultrapure or Nanopure water
- Box of Kimtech Wipes or kimwipes (delicate task wipes in green box) or Lens Paper (lens paper is recommended for all optical faces)
- Dry nitrogen (for drying the lenses without oil clouding the lens)
- 1/4" push rods, delrin or nylon, Grainger Model# 2XMG1 or # 2XNZ9
- Standard squirt bottles for Milli-Q water and Triton Solution
- Dispensing ESD bottle for methanol (Grainger Model# 35270)
- Bottle top filtration system 0.1µm, 500ml volume (VWR# 89220-696)
- Nitrile gloves
- 1x dark card (cardboard wrapped with electrical tape works great)
- 2x large plastic bags, 1x clean tubing, 1x dirty tubing
- Bleach (for cleaning tubing)

Note on Personal Protective Equipment (PPE): The use of gloves is required when handling Triton X and Methanol (MeOH), in addition to exercising sound safety practices with chemical use, gloves should also be worn whenever you are working close to any optical faces as they are sensitive to dirt, oil, and grease that could be introduced if using bare hands during cleaning and calibration procedures.

5.2 Cleaning the Transmissometer

As previously mentioned the recommended amount of cleaning the transmissometer should occur pre and post-cruise, if the ship is to be dockside for more than a week, as well as during long duration cruises or when biofouling is suspected. In addition to cleaning the optics, the tubing connected to the underway system that feeds the C-Star should also be cleaned regularly. A redundant set of tubing should be available to easily swap out tubing when necessary. Below are instructions for recommended cleaning practices:

- Remove the C-star from the underway system and remove the flow tube by loosening the set screws (9/64") on the flow sleeve, the flow sleeves only need to move ~1/2" to easily remove them.
- 2. Rinse the C-star body, both optical windows and the flow tube with Milli-Q. Check for evidence of salt crystals or debris, continue to rinse and flush until salt crystals and debris are not apparent.
- 3. Next, use a squirt bottle to distribute the diluted 0.1% Triton solution onto the optical windows and through the flow tube, follow with a rinse using Milli-Q water. Rinse until soapy residue is undetectable.

- 4. Inspect the o-rings on the optical windows and flow tube for debris or damage and replace if necessary (size 122).
- 5. Clean the flow tube. Use 2-3 kimwipes that are damp with MeOH and place over the opening of the flow tube. Using the delrin or nylon rod, carefully, slide the kimwipe through the flow tube (<u>Do NOT use a bottle brush</u>!). If you feel resistance, back out, reposition the kimwipes and go again. Repeat this process until the kimwipes come out clean.
- 6. Follow up by pushing through dry kimwipes, inspect the flow tube for any streaks, or particles left by kimwipes, and repeat with a dry kimwipe, if necessary.
- 7. Ensure that the flow sleeves are dry as well, loosen the set screws to easily remove them from the flow tube and dry the inside and outside of the flow sleeves. In some cases, it may be necessary to use the delrin or nylon rod to push kimwipes through the stainless nozzle fittings to remove any biofouling. MeOH damp kimwipes can be used in the event of heavy soiling. Dry flow sleeves ensure no moisture is introduced to the optical windows or flow tube when reattached.
- 8. Clean the optical windows, use 2 kimwipes that are damp with MeOH and carefully dab with firm pressure. If there is heavy soiling, you may very gently rub the kimwipe across the optical window, do not perform any scrubbing motion. Repeat, if needed.
- 9. If necessary, follow up with a dry kimwipe on the optical window, inspect for any streaks or particles left by the kimwipe. Repeat, if necessary, until the optical windows look clean, dry, and debris-free.

Other tips:

- If available, nitrogen gas can be used to dry the optical windows and flow tube. Ship supplied air or other air sources are not recommended as they may contain oil that will foul the optical surfaces of the sensor.
- When replacing the flow tube, secure flow sleeves onto optical window and tighten the set screws but do not overtighten, hand tight is good, otherwise leaks will occur.

5.3 Cleaning the Tubing

As previously mentioned, a spare set of tubing helps in both turnaround and ensuring tubing has ample cleaning time prior to installation. Biofouling of tubing can occur quickly, to check for fouling use a paper towel to wipe on the inside of the tube opening, if any fouling is observed tubing should be replaced with a clean set.

- 1. Remove dirty tubing from system and place in a large plastic bag labeled dirty tubing and replace with clean tubing
- 2. Run fresh water through the dirty tubing and rinse off salt crystals and debris on outside
- 3. Soak tubing in a bath of bleach solution (final concentration 1.25%) for several hours
 - a. Note: Household bleach is typically 8.25%, Laboratory bleach typically comes in 12.5%.
- 4. Remove from bath and rinse well with Milli-Q
- 5. Soak tubing in bath of Milli-Q for several hours
- 6. Allow to dry prior to placing in large plastic bag labeled for clean tubing

The debubbler should also be inspected frequently for biofouling and cleaned as recommended above. A black cover made of neoprene can be used to cover the window of the debubbler to protect it from biofouling, the addition of velcro allows for easy inspection, when required.

5.4 In-situ Air Calibration

To perform an effective air calibration the optical windows and flow tube must be clean and dry and there must be a means of completely blocking the sensors optical path shielding it from light. Below are step by step instructions to performing effective V_{airs} and V_{darks} in-situ calibration:

- 1. Turn the sensor on and allow to warm up for at least 5 minutes. For each test, collect data for 3 minutes.
- 2. Follow steps 1-9 of the cleaning procedures outlined in the section 5.2 and ensure both the optical windows and flow cell are clean and dry.
- 3. Perform V_{darks} by blocking the light source using a dark card, do not touch the optical window with your fingers and record data once the signal count has stabilized.
 - a. The signal count (Figure 1, column 3) and corrected signal raw count (Figure 1, column 4) should both read 00000.

- b. If signal count values are >50, ensure you are completely blocking the light source. Cycle the power and check the counts again, if values are still >50 proceed to send in the instrument for servicing.
- 4. It is recommended that the flow tube be reinstalled prior to running a clean air calibration. Ensure the flow tube is dry and carefully reinstall onto optical windows. Use small black caps, if available, over each of the nozzles on the flow tube, providing a dark environment, free of moisture and dust while collecting data.
- 5. Begin to record data and allow signal count to stabilize, with the flow tube in place and the optical path unobstructed.
 - a. The corrected signal counts should be close to that of the clean air value (V_{air}).
 - b. If values are within 100 counts of V_{air} record the sensor output
 - c. If values are >100 counts, this may indicate optical surfaces are not fully clean, repeat cleaning steps 5-9 until signal counts reach 100 or less, record the sensor output
 - d. If values are stable, but between 100 and 500 counts of the calibration sheet, record the sensor output
 - e. If, after repeated cleanings, the sensor fails to output a signal count within 500 counts of the manufacturer's calibration, the sensor may need to be serviced or re-calibrated.

5.5 In-situ Calibration: Clean Water Check

If supplies are available, perform an in-situ clean water calibration (V_{refs}) and record. This calibration is not required, but is considered helpful in determining sensor drift and if tubing should be replaced or cleaned. Note that it is important to conduct the clean water check in a particle free environment with pure water. Below are step-by-step instructions to performing an effective V_{refs} in-situ calibration.

- The sensors should have been cleaned according to steps 1-9 outlined in Section 5.2. It is recommended to perform this calibration after V_{darks} and V_{airs} since the sensor has been cleaned and the flow tube reattached.
- 2. With the flow tube attached, use pure water to fill the flow tube completely, until water is visible in both nozzle fittings, place the small black caps over the nozzles and invert the instrument to ensure bubbles rise into the nozzles. Remove the black caps and top off with pure water. Replace black caps.
 - a. "Pure water source" refers to fresh water (Milli-Q) that has undergone a second filtration (0.1 micron).
 - b. It is recommended that several hundred mls of pure water be filtered to allow for excess in the event multiple readings need to be taken to perform this calibration. The recommended filtration units have a capacity of 500ml

total volume. Excess volume can be stored and used for future clean water checks. Ensure the bottle is labeled properly (0.1µm filtered Milli-Q with date). It should be noted that these filtration units can be used multiple times. To filter water, ensure the top unit and bottom unit are secure (if an air-gap exists, it will not filter), remove the lid to the top of the unit and fill with Milli-Q water, place lid back on the top unit. Wrap your hands around the bottom of the bottle and apply firm pressure to gently squeeze. Filtration should begin, if filtration slows or halts simply re-apply pressure and squeeze to continue filtration.

- 3. With the flow path full of pure water and the nozzles capped, perform V_{refs} calibration by recording data once the signal count has stabilized.
 - a. The corrected signal count should be close to that of the clean water value (V_{ref}) from the factory calibration sheet.
 - b. If values are within 100 counts of V_{ref} , record the sensor output as V_{refs} .
 - c. If values are >100 counts, drain the flow tube and repeat step 2 above If the issue persists this may indicate optical surfaces are not fully clean. Repeat cleaning steps 5-9 until signal counts reach 100 or less, once stable record the sensor output as V_{refs}
 - d. If values are stable, and between 100 and 500 counts of the calibration page, record the sensor output
 - e. If, after repeated cleanings, the sensor fails to output a signal count within 500 counts of the manufacturer's calibration, the sensor may need to be serviced or re-calibrated.
- 4. Re-attach all in-line tubing so that the instrument is connected to science seawater flow-through system.

5.6 In-situ Calibration Log

When performing in-situ calibration for a transmissometer, it is essential to keep a log of the results of the calibration tests. A sample log sheet is provided in the Appendix. Alternatively, a log file can be based on the Sea-Bird Transmissometer Air Calibration Log template in App Note 106 (See Section 8).

6. Installation

- 1. A debubbler is recommended to be installed upstream of the instruments to reduce bubbles in the system.
- 2. In an underway system, the flow-tube offered by SeaBird is recommended in conjunction with black tubing to decrease the likelihood of biofouling.
 - a. Flow tube: <u>https://www.seabird.com/c-star-25-cm-leak-resistant-flow-tube/product-details?id=54627910474&callback=pfa</u>

b. Black tubing:

https://www.amazon.com/gp/product/B00XK18CK6/ref=ppx yo dt b sear ch asin title?ie=UTF8&psc=1

- 3. When installing the instrument, tubing should be installed so that water is flowing from the bottom to the top of the transmissometer to eliminate air pockets along the optical path. Figure 2 provides an example of an underway system with inline transmissometers on R/V Armstrong.
- 4. When mounting the transmissometer, it is recommended to include some rubber padding between the sensor and the mounting surface to reduce the influence of ship vibrations.
- 5. Usage of a power supply with a fixed voltage of 12V is recommended to ensure the 15V threshold for the Cstar is not exceeded.



Figure 2: Diagram showing input, flow path, and output of science sea water (SSW) system on the R/V Armstrong.

- 7. Recommendations for storage of the Transmissometer when ship is expected to be dockside for more than a week
 - 1. Conduct a fresh-water flush of the entire underway system to remove any seawater, this will burst any remaining cells adapted to seawater in the system but will not remove all biofouling.
 - 2. Drain the system of all water.
 - 3. Follow the Cleaning Procedures outlined in 5.2 and 5.3.
 - 4. Re-assemble the system, clean and dry.

8. Additional background reading

https://ioccg.org/wp-content/uploads/2019/05/beamc_protocol_april-2019.pdf

https://ioccg.org/wp-content/uploads/2019/11/inline-protocols-4.0-nov2019.pdf

App. Note 106 Sea-Bird Air Calibration Log Template (pp. 10-11), Note this is designed around CTD applications (see <u>https://www.seabird.com/transmissometers/c-star-transmissometer/family-downloads?productCategoryId=54627869913</u>)

Appendix

Example of a transmissometer in-situ calibration log sheet.

Device Metadata			Factory Calibration Values		Tranamiaaamatar In aitu			
Serial Number			Vd			Calibration Log		
Path Length (z)			Vair				Calibrat	IOT LOG
Wavelength			Vref					
								-
Date	Operator	<u>VairS</u> <u>Average</u> <u>Signal</u> Count	<u>VairS</u>	<u>VdarkS</u> <u>Average</u> <u>Signal</u> Count	<u>VdarkS</u>	<u>VrefS</u> <u>Average</u> <u>Signal</u>	<u>VrefS</u>	Notes
				Obditt		oount		