

# **DOTS Flow Cell user guide**

**A handbook for pH and DO monitoring in  
flow loop applications**



## Table of contents

<b>Introduction and general considerations .....</b>	<b>5</b>
<b>The DOTS Fiber Optic system with Flow Cells .....</b>	<b>7</b>
DOTS Flow Cells .....	7
DOTS Fiber Optic Sensor .....	8
Fiber Optic Cables .....	9
DOTS Software .....	9
<b>Technical Specifications .....</b>	<b>10</b>
DOTS Flow Cells – pH .....	10
DOTS Flow Cells – DO .....	11
<b>Recommended operating conditions .....</b>	<b>12</b>
DOTS Flow Cells .....	12
DOTS Fiber Optic Sensor and cables .....	12
<b>Absolute maximum ratings .....</b>	<b>13</b>
<b>Warnings .....</b>	<b>14</b>
<b>Declarations and certificates .....</b>	<b>15</b>
CE conformity .....	15
WEEE conformity .....	16
FCC compliance .....	17
<b>Setup and Handling .....</b>	<b>18</b>
<b>Connecting DOTS Flow Cells into a flow loop .....</b>	<b>18</b>
<b>Connecting DOTS Flow Cells to the DOTS Fiber Optic Sensor .....</b>	<b>24</b>
<b>Connect Pt100 temperature sensors .....</b>	<b>27</b>
<b>Connecting the DOTS Fiber Optic Sensor to the DOTS Software .....</b>	<b>27</b>
<b>Powering on the DOTS Fiber Optic Sensor .....</b>	<b>29</b>
<b>Starting your experiment .....</b>	<b>30</b>
<b>Calibration .....</b>	<b>31</b>
Oxygen flow cells .....	31
pH flow cells .....	32
Preparing calibration solutions using DOTS Cal Caps .....	33



<b>Sterilization and Storage .....</b>	<b>35</b>
<b>DOTS Flow Cell's measurement principles .....</b>	<b>36</b>
Optical pH and DO measurements .....	36
Principle of measurement – pH flow cells.....	37
Principle of measurement – DO flow cells.....	38
<b>Troubleshooting .....</b>	<b>39</b>
Table of common problems and solutions .....	39
Updating the DOTS Fiber Optic Sensor firmware.....	39
<b>Contact .....</b>	<b>40</b>

## Introduction and general considerations

Welcome to the DOTS Flow Cell user guide and congratulations on choosing the DOTS Fiber Optic system to monitor the pH and DO of your culture. This user guide will give you access to all information required to install, integrate, and use the fiber optic system within your laboratory everyday life. It is strongly recommended to read this user guide prior to any installation or operation of the system.

**Important aspect:**

This kind of grey box will be used throughout the complete document for the indication of important aspects, hints, or summaries.



**Caveats are indicated by yellow warning signs**



**Dangers and risks are indicated by red danger signs**

To ensure that this user guide provides you all information you need during your work with the fiber optic system, we at SBI are reliant on your feedback. Do not hesitate to contact us to share your ideas regarding errors, missing information, or incomprehensibilities so that we can improve this document and keep it up to date with your requirements.

This user guide, the DOTS Fiber Optic Sensor, and the DOTS Flow Cells may be subject to changes and improvements without advanced notice.

You may request the latest manual from our team of Application Scientists.  
[insights@scientificbio.com](mailto:insights@scientificbio.com)

In case of any questions that might arise during your work with the fiber optic system, do not hesitate to contact us.



## **DOTS Flow Cell user guide revisions:**

Revision 0	02.09.2022	Initial document release
Revision 1	21.11.2023	Updated SBI North America address

## The DOTS Fiber Optic system with Flow Cells

The DOTS Fiber Optic system consists of the DOTS Flow Cells, DOTS Fiber Optic Sensor, fiber optic cables, and the DOTS Software.

---



Figure 1: The DOTS Fiber Optic system with flow cells – pH and dissolved oxygen monitoring for flow loops.

### DOTS Flow Cells

DOTS Flow Cells enable continuous, in-line monitoring of pH and DO (dissolved oxygen) within a flow loop of, e.g., a bioreactor.

The DOTS Flow Cells contain the optical sensing elements that measure pH or DO of the liquid passing through the flow cells. The flow cells are color coded to easily distinguish between the type of included sensor: pH flow cells have a green band around the cable lock nut while DO flow cells have a blue band (see Figure 2). Luer connectors can be used to couple two flow cells together (Figure 1) and to connect the flow cells in-line within a flow loop. Male-male Luer connectors and Luer-barb connectors with quarter or eighth-inch barbs are available for purchase from SBI.



Figure 2: DOTS Flow Cells for pH and DO monitoring (left) can be connected in-line with standard Luer connectors (right).

### DOTS Fiber Optic Sensor

The DOTS Fiber Optic Sensor contains the light source that excites the sensing elements within the flow cells, collects the sensor signal, and communicates with the DOTS Software. DOTS Fiber Optic Sensors are available as either a single channel sensor for monitoring either pH or DO, or a dual channel sensor for monitoring both pH and DO in parallel. A dual channel sensor is shown in Figure 3 with key ports and LEDs labeled. Pt100 temperature sensors can be connected at each channel for temperature-corrected sensor measurements.

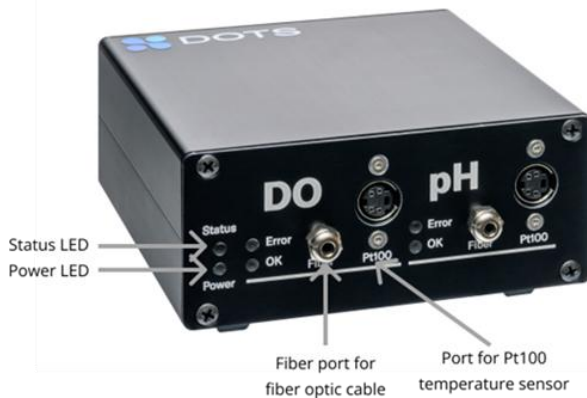


Figure 3: A DOTS Dual Channel Fiber Optic Sensor with ports labeled for fiber optic cables, Pt100 temperature sensors, and Status and Power LEDs



### **Fiber Optic Cables**

Fiber optic cables connect the DOTS Flow Cells to the DOTS Fiber Optic Sensor. The cables come color coded to match the two types of flow cells (green for pH, blue for DO).

---



---

Figure 4: Fiber optic cables with color coding

### **DOTS Software**

The DOTS Software allows users to setup experiments, visualize data, and control any connected sensors. Instructions for the DOTS Software are not included in this manual, but are provided in the DOTS Software User Guide.



## Technical Specifications

### DOTS Flow Cells – pH

#### Measurement range

DOTSFCPH57	5 – 7 pH (pKa 6)
DOTSFCPH68	6 – 8 pH (pKa 7)
DOTSFCPH79	7 – 9 pH (pKa 8)

#### Housing material

Nylon

#### Sterilization

Beta irradiated at 25 kGy

#### Response time (t90)<sup>1</sup>

Flow > 10 mL/min	< 90 seconds
Flow < 10 mL/min	< 120 seconds

#### Accuracy

± 0.05 (after a 2-pt calibration)

#### Resolution (at pKa value)

0.003 pH units

#### Drift

< 0.005 pH units per day at 25 °C

#### Shelf life

1 year in original packaging

#### Storage conditions

Dry, dark, and at room temperature

#### Cross sensitivities

Organic solvents at high concentrations, charged surfactants

<sup>1</sup> Time for 90% of the total sensor signal change in circulated media

## DOTS Flow Cells – DO

<b>Measurement range</b>	0 – 250% air saturation (approximately 50% DO)
<b>Housing material</b>	Nylon
<b>Sterilization</b>	Beta irradiated at 25 kGy
<b>Response time (t90)<sup>1</sup></b>	
Flow > 10 mL/min	< 20 seconds
Flow < 10 mL/min	< 30 seconds
<b>Accuracy</b>	
5% air saturation	± 0.1%
95% air saturation	± 1%
<b>Resolution</b>	
5% air saturation	± 0.05%
95% air saturation	± 0.25%
<b>Lifetime</b>	10 million data points
<b>Shelf life</b>	3 years in original packaging
<b>Storage conditions</b>	Dry, dark, and at room temperature
<b>Cross sensitivities</b>	Organic solvents at high concentrations, bleach

<sup>1</sup> Time for 90% of the total sensor signal change in circulated media

## Recommended operating conditions

### DOTS Flow Cells

<b>Temperature</b>		0 – 50 °C
<b>Pressure</b>		0 – 2 bars
<b>Flow rate</b>		1 – 500 mL/min
<b>Calibration</b>		
	pH	Factory calibration
	DO <sup>1</sup>	2-point calibration under environmental conditions
<b>Resolution</b>		
	5% air saturation	± 0.05%
	95% air saturation	± 0.25%
<b>Lifetime</b>		10 million data points
<b>Shelf life</b>		3 years in original packaging
<b>Storage conditions</b>		Dry, dark, and at room temperature
<b>Cross sensitivities</b>		Organic solvents at high concentrations, bleach

<sup>1</sup> Each 2-point calibration may be used for all sensors within the same production batch (as indicated by the sensor code on the DOTS Flow Cell packaging).

### DOTS Fiber Optic Sensor and cables

<b>Temperature</b>		15 – 45 °C
<b>Humidity</b>		0 – 80% (non-condensing)
<b>Input voltage</b>		5 VDC ± 5%
<b>Cable minimum bend radius</b>		80 mm

## Absolute maximum ratings

### Power supply (via USB)

voltage	4.5 – 5.5 VDC
current	0.5 A

### Operating temperature

0 – 60 °C
(10 – 50 °C recommended)

### Operating humidity

< 80 % (non-condensing)

## Warnings



Do not use the DOTS devices or any of its components in water bath! This might result in electric shocks, which could damage your health, the DOTS device and any other electric device around.



Do not look into the beam of any of the DOTS sensor fiber optic components! Their emitted light is of high intensity and might damage your eye or retina. Wear protective eye wear.



Do not touch, wet or electrically bridge the USB connectors! This might result in damage to your health and/or the DOTS device.



Do not spill liquids over the DOTS devices. Liquids may enter the housing and destroy the device by corrosion.



Do not introduce sharp bends into the fiber optic cables.



In general, do not use inorganic or organic acids and bases, organic solvents or detergents to clean the DOTS Fiber Optic Sensor or DOTS Flow Cells. Some organic solvents or detergents might be allowed for cleaning, but you should only use those being mentioned in the user guide.



Any kind of opening, manipulating or copying DOTS devices as well as decompiling, reverse-engineering, copying or distributing DOTS software or firmware or its components is strictly prohibited in accordance with German and international law and may lead to compensation claims.



SBI sensors, devices and other equipment are not intended for medical or military purposes or any other safety-critical applications. It is strictly prohibited to use SBI sensors, devices and other equipment for applications in humans or for applications where sensors are brought in direct contact with foods, drinks, tissues or other goods that are transferred into humans.

## Declarations and certificates

### CE conformity

The aquila biolabs GmbH, Arnold-Sommerfeld-Ring 2, 52499 Baesweiler, Germany, herewith declares under its sole responsibility that all devices and equipment being part of the DOTS Fiber Optic system and being manufactured by the aquila biolabs GmbH are in conformity with the Council Directives as described in EN IEC 61000-6-2:2019, EN IEC 61000-6-4:2019, EN 61326-1:2013, EN 55016-2-3:2017 + A1:2019, EN 61000-4-2:2009, EN IEC 61000-4-3:2020-09, EN 61000-4-4:2012, EN 61000-4-5:2014 + A1:2017, EN 61000-4-6:2014

This declaration applies to all products with the following identifiers:

DOTS Fiber Optic Sensors:

- DOTS-PHDO-BV Revisions: 1.00
- DOTS-PH-BV Revisions: 1.00
- DOTS-DO-BV Revisions: 1.00

Technical documentation is maintained at the aquila biolabs GmbH headquarter in Arnold-Sommerfeld-Ring 2, 52499 Baesweiler, Germany.

Date of declaration: 01.09.2022

Name, position of the undersigned: Konrad Herzog, Managing Director

Research & Development

### WEEE conformity

WEEE-Registration-No.: 61144888

The aquila biolabs GmbH, Arnold-Sommerfeld-Ring 2, 52499 Baesweiler, Germany, herewith declares compliance of all electronic components of the DOTS Fiber Optic system with the Council Directive 2012/19/EU.

Electronic components may contain various hazardous substances that could possibly exhibit negative impacts on your health and the environment. In order to avoid those effects aquila biolabs encourages you to make use of the appropriate local take-back and recycling systems for disposing electrical and electronic equipment. By doing this you are furthermore significantly reducing the pressure on natural resources and thus preserve our planet for subsequent generations.

Components that are affected by this declaration carry the following pictogram:



Date of declaration: 01.09.2022

Name, position of the undersigned: Konrad Herzog, Managing Director

Research & Development



### **FCC compliance**

This is a Class A product.

This device complies with Part 15 of the FCC Rules. Operation is subject to the following two conditions:

- (1) this device may not cause harmful interference, and
- (2) this device must accept any interference received, including interference that may cause undesired operation.

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

## Setup and Handling

A typical workflow to set up an experiment with the DOTS Fiber Optic System includes the following steps:

1. Connect DOTS Flow Cells into a flow loop
2. Connect DOTS Fiber Optic Sensor to the lab computer and power on
3. Connect flow cells to the DOTS Fiber Optic Sensor using Fiber Optic Cables
4. *For non-temperature-controlled environments:* Connect at least one Pt100 temperature sensor to the DOTS Fiber Optic Sensor
5. Start your experiment using the DOTS Software

The individual setup steps for the hardware are explained in detail below. Information on all workflows of the DOTS Software can be found in the DOTS Software User Guide.

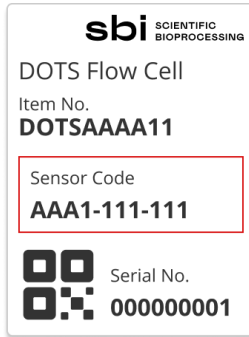
### Connecting DOTS Flow Cells into a flow loop

All steps in this section must be performed in a sterile environment, such as a laminar flow hood, biosafety cabinet, or clean bench, to maintain sterility of the pre-sterilized DOTS Flow Cells and Luer connectors.



**DOTS Flow Cells are sterile products. Integrate them into your flow loop in a sterile environment to maintain their sterility.**

Open the foil package(s) containing a single DOTS flow cell. Keep the package or save the sensor code shown on the package label (highlighted red in Figure 5). This code contains a factory calibration that can be used when setting up a new experiment in the DOTS Software.



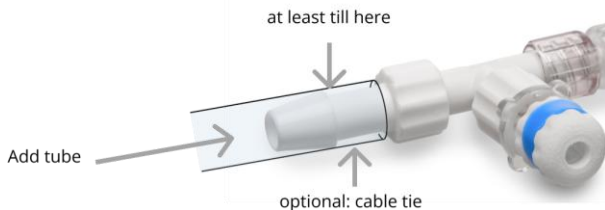
**Figure 5: Sensor code located on the exterior of each DOTS Flow Cell package.**

DOTS Flow Cells are T-shaped with Luer lock fittings on either side of the integrated sensing element. The flow cells are packaged with caps on the Luer locks to aid in maintaining sterility when handling and during setup. Unscrew the caps and replace them with the appropriate size of Luer connectors to integrate the flow cells into your flow loop.



**Figure 6: The DOTS Flow Cells are packaged with capped Luer locks (left) and can be purchased with separately packaged Luer-barb connectors, shown connected (right).**

The inner diameter of the connecting tubes on both sides of the flow cell should be of the same size to avoid pressure build-up inside the flow cell. If using tubing with extremely small inner diameter, allow sufficient time when setting up your system for the entire flow loop to fill with liquid. Ensure that the tubes fit tightly onto the connectors and that the barb-part of the connectors are pushed all the way into the tube. You can additionally secure the tubes using cable ties.



**Figure 7: Connecting and securing a tube onto the Luer-barb connector of a DO flow cell.**



**Always check that all tubes and connectors are tightly connected before starting an experiment.**

Use a male-male Luer connector to couple two flow cells together (i.e., one for pH and one for DO) and then connect the linked flow cells into your flow loop (Figure 8).



**Figure 8: A double-thread, male-male Luer connector (left) can be used to connect two flow cells together (right).**

The steps below detail the installation of a coupled pH and DO flow cell unit into a bioreactor flow loop.

Working in a sterile environment, remove the pH flow cell (indicated by the green ring on the upper lock nut) from its sterile packaging. Make sure to record the Sensor Code on the outside of the package (Figure 5).



Holding one side of the capped flow cell, remove the cap from the other Luer lock and replace it with a Luer-barb connector, or other tubing connector specific to your tubing type and diameter.

**Figure 9: Replacing a Luer cap with a Luer-barb connector on a sterile, pH flow cell.**



Connect one end of your sterile flow loop to the flow cell, making sure to push the tube all the way onto the barb portion of the Luer-barb connector.

**Figure 10: Connecting one side of a pH flow cell into a sterile flow loop.**

Repeat these steps with a dissolved oxygen flow cell. It is recommended to perform setup in this order so that sterility is maintained within the pH flow cell while handling the DO flow cell.



Holding the connected tubing of one of the flow cells, remove the remaining Luer cap and replace with a male-male Luer connector.



Figure 11: Replacing the remaining Luer cap from a pH flow cell with a dual-thread, male-male Luer connector.



Remove the remaining Luer cap from the second flow cell and connect to the other side of the male-male Luer connector.

Figure 12: Connecting a DO and pH flow cell together within a sterile flow loop via a male-male Luer connector.

Once your single or coupled flow cell is connected to the flow loop, it will stay sterile. All further steps do not require a sterile environment.



**We recommend using a peristaltic pump with low pulsation for pumping liquid through your flow loop and the DOTS Flow Cells.**



**Double check all tubing and Luer connections prior to starting your experiment.**

## Connecting DOTS Flow Cells to the DOTS Fiber Optic Sensor

Place the DOTS Fiber Optic Sensor close to your flow loop. Choose a spot where it is safe from spills of chemicals or culture broth.

For each flow cell, take a fiber optic cable with matching color coding. Notice that there is a lock nut on one end of each fiber optic cable. Push this end of the fiber optic cable into the hole of the correct channel(s) (DO=blue, pH=green) of the DOTS Fiber Optic Sensor (Figure 13). The channels for the fiber optic cables are labeled "Fiber". The cables fit tightly so you may need to use some force to insert the cables. Make sure to push the cable all the way in until you feel a resistance.

### Connecting DOTS Flow Cells to the DOTS Fiber Optic Sensor



Figure 13: Connecting fiber optic cables to the Fiber ports of the DOTS Fiber Optic Sensor.



**Grab the fiber optic cable close to its edge when pushing it into the Fiber Optic Sensor channel to minimize the risk of damaging the cable.**



Push the screw nut down the cable and tighten onto the thread on the channel port to secure the fiber optic cable. You can check the connection by testing whether the cable still moves when you pull it gently (it should not).

Take the free end of the fiber optic cable and guide it to the correct flow cell. Loosen the color-coded lock nut on the flow cell and push the fiber optic cable inside until you feel a resistance (Figure 14).



Figure 14: Connecting a fiber optic cable to an in-line DOTS Flow Cell.



**Do not bend the fiber optic cables in sharp turns. Bending the cables beyond the minimum bend radius (80 mm) will damage the cables.**



**The end of the fiber optic cable must be flush with the bottom of the flow cell.**

Tighten the lock nut to secure the cable in the flow cell.

Proceed to connect the second Flow Cell (if you have prepared one) to the other channel of the Fiber Optic Sensor (Figure 15).



Figure 15: Completing the connection of two flow cells to the DOTS Fiber Optic Sensor via two fiber optic cables.



Double check the connection between fiber optic cable and flow cell if the LED labeled “Error” on the DOTS Fiber Optic Sensor illuminates red. This is an indication of poor signal quality being detected by the DOTS Software.



Double check all Luer connections to ensure that the tubing did not slip and that the connection between the coupled flow cells is still secure.

## Connect Pt100 temperature sensors

Changes in temperature affect the measurements of DOTS Flow Cells. DOTS Software can compensate for temperature influences if a suitable input of temperature data is provided. Pt100 temperature sensors are available for purchase with the DOTS Fiber Optic Sensor. We recommend the use of a real time temperature sensor for experiments in a non-controlled temperature environment.

To setup, simply plug in the cable end of the Pt100 to the port labeled “Pt100” to the right of the “Fiber” channel. Secure the metal end of the temperature sensor in a location near the flow cells, so that environmental temperature changes will be detected and controlled for within the DOTS Software.

If you are monitoring both pH and DO at the same location within a flow loop, only one Pt100 temperature sensor must be connected to the DOTS Fiber Optic Sensor for temperature compensation for both pH and DO measurements.



**Place the Pt100 temperature sensor as close as possible to the Flow Cell to ensure correct interpretation of Flow Cell data.**



**If monitoring pH and DO in separate locations use two Pt100 temperature sensors, one for the pH channel and one for the DO channel.**

For experiments where temperature is known and controlled, we recommend the use of a manual temperature input within the DOTS Software to simplify experiment setup.

## Connecting the DOTS Fiber Optic Sensor to the DOTS Software

The DOTS Fiber Optic Sensor uses a USB connection for both power input and data transfer. Connect the Type-B end of the USB cable to the USB port on the



back of the Fiber Optic Sensor (Figure 16). Use the attached screws to fix the cable. Connect the Type-A end of the USB cable to the computer that has DOTS Software installed. The LED labeled “Power” on the front of the Fiber Optic Sensor (Figure 3) should now light up green to show that the Fiber Optic Sensor is powered through the USB connection.



Figure 16: The USB port and Power button on the back of the DOTS Fiber Optic Sensor.

## Powering on the DOTS Fiber Optic Sensor

To turn the Fiber Optic Sensor on, press the black “Power” button on the back of the DOTS Fiber Optic Sensor (Figure 16). After 1-2 seconds the LEDs on the front of the sensor will start blinking. The Fiber Optic Sensor is turned on and ready when the LEDs stop blinking and the LEDs labeled “Power” and “OK” are steadily illuminated green.



**Only press the Power button once or you will enter the boot mode of the Fiber Optic Sensor. For more information on this function refer to the Updating DOTS Fiber Optic Sensor firmware section of the user guide.**



**Make sure that the LEDs on the front of the Fiber Optic Sensor have blinked, and that the “OK” LEDs are illuminated green. If only the “Power” LED is green and nothing else is lit, the Fiber Optic Sensor is not turned on.**

It is possible to multiplex your system with additional sensors by using the DOTS Expansion Hub (Figure 17). The Hub includes 7 USB ports so that you can connect and monitor up to 7 Fiber Optic Sensors on the same DOTS computer using a single cable between the Hub and computer. All necessary cables are included with the purchase of the DOTS Hub and additional Fiber Optic Sensors.



Figure 17: The DOTS Expansion Hub for connecting up to 7 DOTS Fiber Optic Sensors to the same computer.



**The use of the DOTS Hub is strongly recommended for system expansion since the hardware performance has been extensively tested and validated.**

## Starting your experiment

Start your bioprocess, start the pump for the flow loop, and return to the DOTS Software to start your pH and/or DO measurements.

The process for how to create and start an experiment in the DOTS Software is explained in detail in the DOTS Software User Guide. The high-level steps are listed below.

1. Click the button to create an Experiment and select a suitable Application Template for your Object of interest.
2. Configure your pH and/or DO monitoring Tasks.
3. Assign a device to each Object.
4. Create your Experiment.



**Users will be prompted to select a Factory Calibration or Custom Calibration when creating an Experiment. Custom calibrations must be generated prior to starting an Experiment.**



Figure 18: The DOTS Fiber Optic Sensor and DOTS Flow Cells connected in a bioreactor flow loop.

## Calibration

DOTS Flow Cells come with a factory calibration that is recognized by the DOTS Software via the Sensor Code listed on the outside of each flow cell package. Sensor codes are batch-specific, meaning that each flow cell within a production batch will have the same factory calibration.

For most users, the factory calibration will be sufficient for monitoring overall trends of changing pH and DO since important parameters such as pressure, temperature, relative humidity, and salinity are compensated for within the DOTS Software.

### Oxygen flow cells

Multiple factors such as salinity, aeration, and media type can affect the saturated oxygen point of the solution. Using the factory calibration with these undefined conditions may result in the %DO measurement going out of range and being capped within the Software. Therefore, for users specifically interested in the %DO measurement it is recommended to generate a custom 2-point calibration under the intended experimental and environmental conditions.

The same custom calibration file can be applied to all experiments with the calibrated-for experimental conditions as long as flow cells from the same batch (with the same sensor code) are used.



**To ensure accuracy of DO measurements, it is recommended to generate a new calibration file at least once a month**

Important parameters that can affect the DO calibration are listed below. Along with each parameter is a brief explanation of how the parameter is compensated for within the DOTS Software.

Parameter	Compensation mechanism
<b>Temperature</b>	Manual input of constant temperature or Pt100 temperature sensor real time measurements
<b>Atmospheric Pressure</b>	Built-in pressure sensor within the DOTS Fiber Optic Sensor
<b>Humidity</b>	Built-in humidity sensor within the DOTS Fiber Optic Sensor



**If using a constant temperature input, ensure that environmental temperature is maintained at the specified degrees of Celsius for the duration of the calibration.**

### pH flow cells

Unlike the oxygen flow cells where an application-specific calibration is recommended to compensate for the variable saturated point of the measured solution, the pH flow cells return robust measurements regardless of application. A custom calibration can be generated periodically to verify the required accuracy of the pH measurements, but generally a one-point offset at the start of an experiment is sufficient when paired with the factory calibration.

Important parameters that can affect the pH calibration are listed below. Along with each parameter is a brief explanation of how the parameter is compensated for within the DOTS Software.

Parameter	Compensation mechanism
<b>Temperature</b>	Manual input of constant temperature or Pt100 temperature sensor real time measurements
<b>Salinity</b>	Manual input specifying salinity range of solution

**It is acceptable for calibration solution salinity to slightly differ from the salinity of the experimental solution.**





If using a constant temperature input, ensure that environmental temperature is maintained at the specified degrees of Celsius for the duration of the calibration.

### Preparing calibration solutions using DOTS Cal Caps

SBI offers DOTS Cal Caps that can be used for the preparation of calibration solutions for the DOTS Flow Cells pH and DO. The property of the final solution is indicated on each Cal Caps bottle.

We recommend the use of the pH 2 and pH 11 cal caps for preparation of acidic and basic pH calibration solutions.

We recommend the use of the 0% DO capsules for preparation of a deoxygenated solution if it is not possible to purge the flow loop system of oxygen with nitrogen (or similar) gas.



The capsules and powdered contents are not sterile. Any solutions prepared using the capsules must be passed through a sterile filter for to maintain sterility of the flow cells and flow system during calibration.



Use caution when preparing and using the acidic pH 2 and basic pH 11 calibration solutions. Use protective eyewear and gloves. Skin or eye contact with the calibration caps or calibration solutions may cause irritations or may otherwise considerably damage your health.

Follow the steps listed below to prepare the calibration solutions.

1. Add deionized water to a sealable container (i.e., a falcon tube or small bottle) at half of the final volume indicated on the Cal Caps bottle label (Figure 19, left).
2. Take one of the capsules from the bottle, pull it apart from either end with a twisting motion, and empty the powdered capsule contents into the container (represented by a beaker in Figure 19 right). Close the container and mix thoroughly until all the powder has dissolved.

3. Reopen the container and add in the remaining half volume of deionized water. Seal and mix well. Allow the 0% DO solution to sit in the sealed container for about 15 minutes before using to calibrate.



**Figure 19: DOTS Cal Caps for preparing pH 2, pH 11, and 0% DO calibration solutions.**

A prepared 0% DO solution may be reused for up to a week if stored in a tightly sealed container with no headspace.



**Always prepare a fresh pH solution when performing a new calibration since the Cal Caps are not delivered sterile, and bacterial growth could affect the pH of the buffer.**

The DOTS Flow Cells calibration is explained in detail in the DOTS Software User Guide.

## Sterilization and Storage

DOTS Flow Cells are delivered sterilized. They are designed as single-use products and cannot be re-sterilized once they have been unpacked and used. Secondary sterilization methods will cause sensor degradation and invalidate the factory calibration of the flow cells.



**Do not attempt to re-sterilize the flow cells following an experiment. We cannot provide troubleshooting support if flow cells have been re-sterilized.**

Store DOTS Flow Cells in their original packaging for maximum sensor shelf life and maintenance of the factory calibration.



**Do not store flow cells in static solution or in calibration buffers for an extended amount of time. Thoroughly rinse flow cells with deionized water following calibration with the Cal Caps buffers.**

The DOTS Fiber Optic Sensor and cables can be disinfected by wiping them softly with 70% ethanol wipes.



**Ensure that you have disconnected all DOTS devices and cables from any kind of power supply to prevent damages to the electronics, to connected devices, and to your health.**



**Ensure that all DOTS devices and cables are completely dry before you reconnect them to each other, to the power supply, and the USB cable.**

## DOTS Flow Cell's measurement principles

### Optical pH and DO measurements

DOTS Flow Cells are available for pH and DO monitoring in flow systems. Pumped solution passes through the flow cells and contacts the embedded chemosensor for either pH or DO. These chemosensors consist of a hydrogel that binds luminescent dyes. The dyes are the sensing element: Their luminescent properties are altered in response to concentration changes of the analytes  $\text{H}_3\text{O}^+$  (pH sensor) or dissolved oxygen (DO sensor).

Fiber optic cables transmit red light (610-630 nm) from the DOTS Fiber Optic Sensor to the chemosensors, causing them to luminesce in the near-infrared region (NIR, 760-790 nm). As analyte concentrations change, the DOTS Fiber Optic Sensor measures the signal intensity and phase shift which are then used to calculate pH and DO (% and mbar) in the DOTS Software.

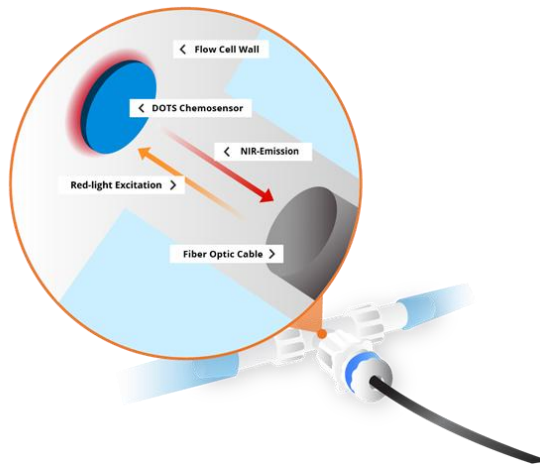


Figure 20: The interior of a DOTS Flow Cell with a connected fiber optic cable.

## Principle of measurement – pH flow cells

DOTS Flow Cells – pH contain embedded chemosensors with both a pH-sensitive luminescent dye and a pH-insensitive reference indicator. As the measured solution acidifies, increasing concentrations of protons bind to the pH-sensitive dye. As the pH indicator is protonated, it emits an increasingly bright luminescence. The DOTS Fiber Optic Sensor measures this luminescence as well as the reference indicator luminescence and correlates the high intensity signal to low pH.

As the measured solution becomes more basic, fewer protons bind to the pH-sensitive dye and little to no measurable luminescence is detected. The DOTS Fiber Optic Sensor measures only the reference indicator luminescence and correlates the lower signal intensity to high pH (Figure 21).

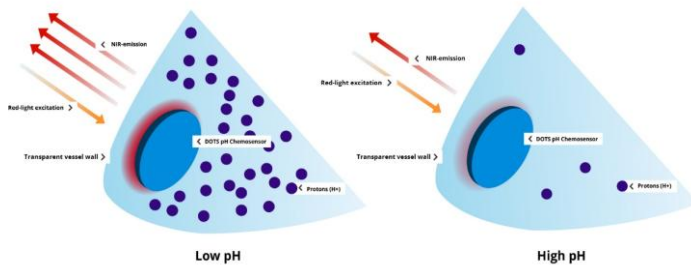


Figure 21: DOTS Flow Cells – pH principle of measurements.

## Principle of measurement – DO flow cells

DOTS Flow Cells – DO contain embedded chemosensors with an oxygen-sensitive indicator. As oxygen molecules collide with the indicator in increasing concentrations, the increased interactions result in a quenching effect on the chemosensor luminescence. This decrease in signal intensity is measured by the DOTS Fiber Optic Sensor and correlated to higher levels of DO.

As dissolved oxygen decreases in the measured solution, fewer interactions between the oxygen molecules and oxygen-sensitive indicator result in brighter luminescence from the red-light-excited chemosensor. This increase in signal intensity is measured by the DOTS Fiber Optic Sensor and correlated to lower levels of DO (Figure 22).

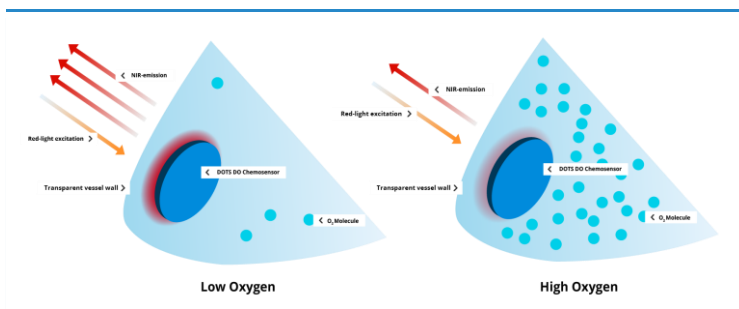


Figure 22: DOTS Flow Cells – DO principle of measurements.

## Troubleshooting

### Table of common problems and solutions

Problem	Solution
<b>Odd sensor signal</b> <b>Indicated by a red-lit “Error” LED on the DOTS Fiber Optic Sensor or a constant pH measurement of approximately 11.3</b>	Fiber optic cable not attached correctly. Ensure that one cable end is flush with the bottom of the flow cell and the other is inserted all the way into the Fiber Optic Sensor
<b>DOTS Fiber Optic Sensor does not connect to DOTS Software</b>	Ensure that the Fiber Optic Sensor is switched on (Power button on the back) and that the LEDs blinked before maintaining a steady state of illumination.

### Updating the DOTS Fiber Optic Sensor firmware

Contact our support team for instructions on how to update device Firmware via the boot mode of the Fiber Optic Sensor.



## Contact

**Website** [www.scientificbio.com](http://www.scientificbio.com)

**Newsletter** <https://www.scientificbio.com/subscription-center>

**subscription** Information on new Software versions, Device firmware updates, new products

**Email** [insights@scientificbio.com](mailto:insights@scientificbio.com)

**Headquarters** SBI North America

SBI Europe

Represented by:

aquila biolabs GmbH

201 Penn Center Blvd

Arnold-Sommerfeld-Ring 2

Pittsburgh, PA 15235

52499 Baesweiler, Germany

+1 413-230-8463

+49 2401 8049708