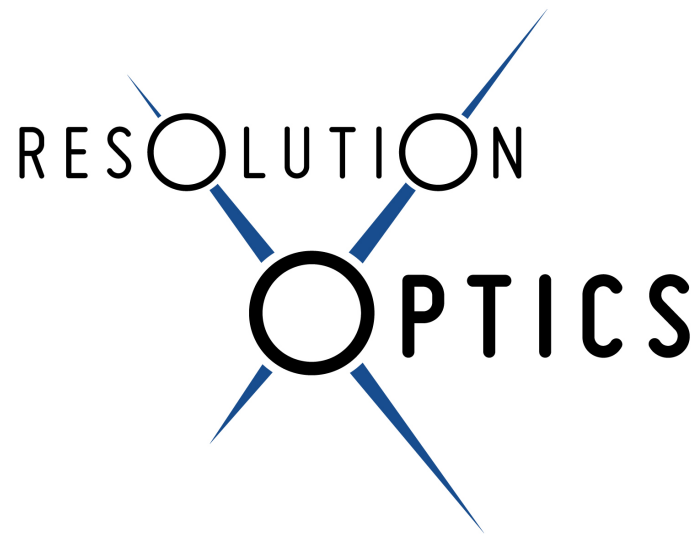




Submersible Microscope User Guide

Version 1.1.0

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Introduction

Congratulations on your purchase of the Submersible Microscope! Your microscope comes from a line of instruments that all operate under proprietary digital inline holography technology platform, developed by Resolution Optics Inc. With its new and unique method of data acquisition and analysis, this system is designed to optimize your research of the microscopic world.

General Safety Information

This instrument is of safety class 1 and has been built and tested according to EN 61010 1/IEC 1010-1/VDE 0411-1 safety standards for electrical measurement, control and laboratory equipment.



Attention!

This microscope contains a class IIIB Laser. Up to 100 mW at 405 nm is emitted from the laser housing itself. If properly assembled, the light emitted from the pinhole should not pose any threat of long term damage to an exposed eye. There are no user serviceable parts inside the light source. If problems are encountered regarding the light source, please return it immediately for service. Allow only a qualified technician to disassemble the point source. **Do not attempt to remedy any problems with the light source on your own!**



Attention!

Always make sure that the proper cable connector is attached to the proper bulkhead connector when setting up the microscope. Failure to do so could result in critical damage to the microscope.

What's in the Box

The following is the list of parts shipped as part of the Submersible Microscope.

1. Submersible microscope, pre-assembled. See the microscope description in the "Microscope and its Components" section.

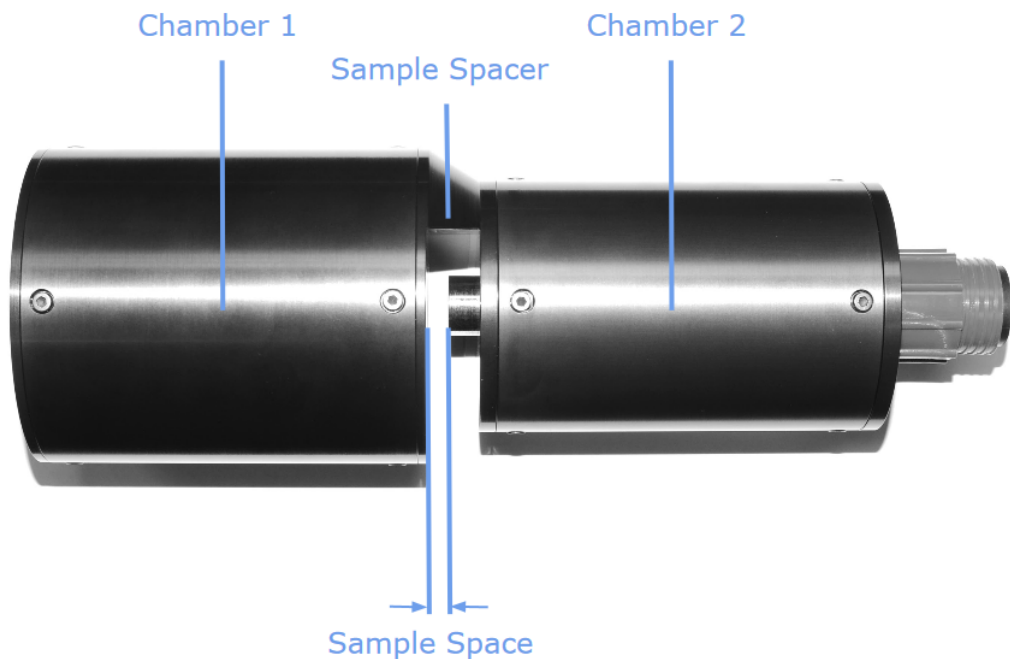
Follow the cable connection instructions in the "Microscope Setup" section.

2. A cable with 13 pin connector to power laser, camera, and transmit data from the camera to the host computer.
3. Camera power supply.
4. Laser power supply.
5. Carrying case.

The Microscope and its Components

The Submersible Microscope consists of 3 primary components:

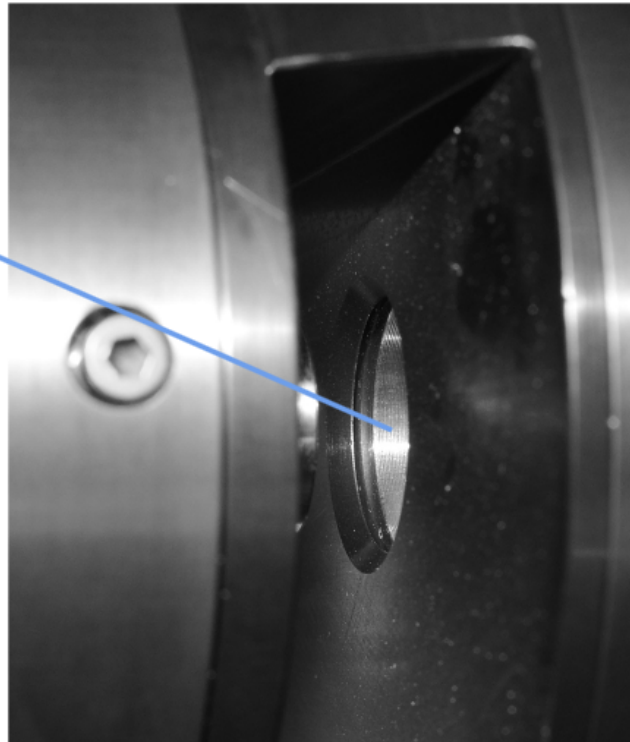
1. Chamber 1: houses the digital camera
2. Chamber 2: houses the laser point source
3. Sample Spacer: connects the two chambers, determines the sample volume that will be imaged.



Chamber 1

This Chamber contains a digital camera. The camera's image sensor is protected by a 2 mm thick sapphire glass window. This window is the most fragile external component of the microscope, and must be handled carefully.

Sapphire Window
(Camera side)



Sapphire Window: It is extremely important that this window is kept clean of dirt and scratches, and great care must be taken in cleaning this window. It is recommended that this window be cleaned under a microscope if possible. Use only lens wipes and organic cleaners for cleaning; do not use solvents as they can dissolve the adhesive material, releasing the window from its place. Avoid transporting the microscope along with loose hardware (Allen keys, screws, etc) as they will scratch the sapphire window surface if contact is made.

Chamber 2

This Chamber contains the laser point source, consisting of a 405 nm laser, a microscope objective and a pinhole. The point source provides the spherical wave of light necessary to produce images in inline holography. Similar to the camera in Chamber 1, the point source is protected by a 0.5 mm thick sapphire glass window.

Sample Spacer

The sample spacer provides a stable environment for imaging microorganisms and particles in addition to determining the source to screen distance of the instrument, which you will soon learn is essential for proper data analysis.

The **Sample Space**, determined by the Sample Spacer, allows for the non-destructive flow of organisms and particles. The Submersible Microscope's camera collects holograms of the samples as they, along with the fluid which contains them, pass through the Sample Space.

Microscope Setup

Step 1

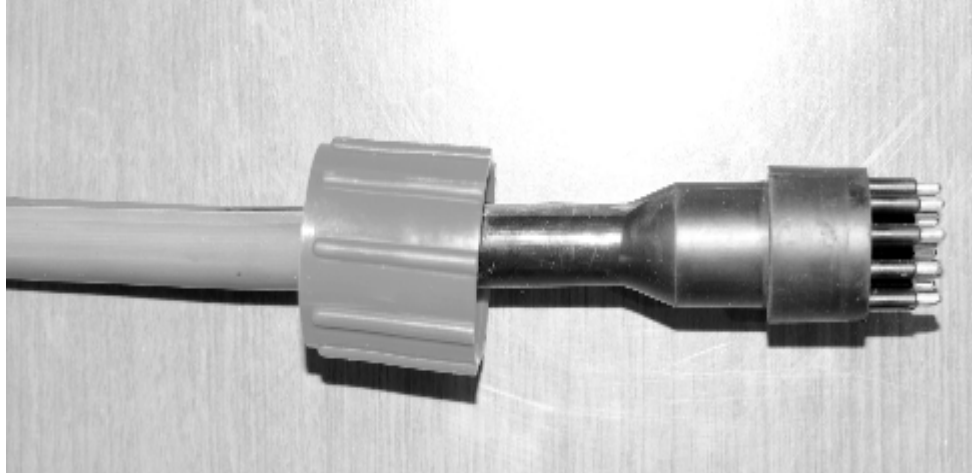
Firmly push the 13 pin connector at the end of the main underwater cable into the bulkhead connector. Try not to wiggle the cable too much as this may damage the pins. Push the cable connector all the way in.



Submersible Microscope bulkhead with 13 pin connector

Step 2

Slide the locking sleeve over the cable connector and screw it onto the bulkhead connector.



Main power and Ethernet cable 13 pin connector

Step 3

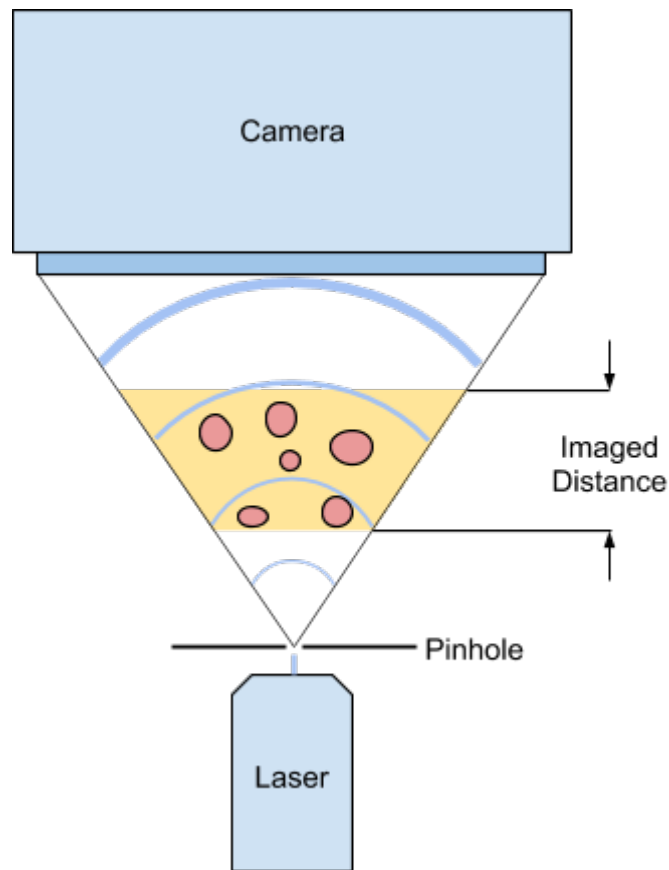
Connect the Ethernet connector of the main cable to the Ethernet port on the computer. Plug in the power adapter for the camera and plug in the laser power adapter to the connector on the same cable.

Step 4

Submerge the microscope into the body of water/fluid that you want to look at. Both sapphire windows should be fully submersed in water for the accurate imaging to be performed.

Principle of Operation

The Submersible Microscope operates on the principles of holography to image a volume in magnification. A 405 nm laser is focused on a 500 nm pinhole of the same order of magnitude as the wavelength of the light, which produces a spatially coherent light source as a reference wave. Light which scatters from the objects within the media (water) will interfere with the reference wave to produce an interference pattern which contains spatial and phase information of the objects within the volume. This interference pattern, the hologram, is recorded by a CCD camera, and reconstructed mathematically to build images of the objects within the volume.



The basic principle of digital in-line holographic imaging

The reconstructed images can be further analyzed, saved, or assembled into the volume reconstructions based on the application.

Software

The holograms acquired by the Submersible Microscope camera need to be properly processed in order to reconstruct the image.

Resolution Optics provides 2 standard software packages, **Swordfish** and **Octopus**, that incorporate our patented fast hologram reconstruction algorithm as well as additional functionality.

Swordfish is a particle counting software capable of detecting, counting and measuring particles in real time with the speed of up to 16 frames per second.

Octopus is a general purpose research-grade software that can reconstruct holograms acquired by the Submersible Microscope camera

at the rate of up to 16 frames per second. Octopus provides advanced 3D visualization and particle detection capabilities.

Please refer to the Swordfish and Octopus brochures and manuals available at www.resolutionoptics.com for the additional details on software installation and use.

Custom software solutions can be developed by Resolution Optics based on customer specifications. Contact us for details.

Tips for Optimal Hologram Acquisition

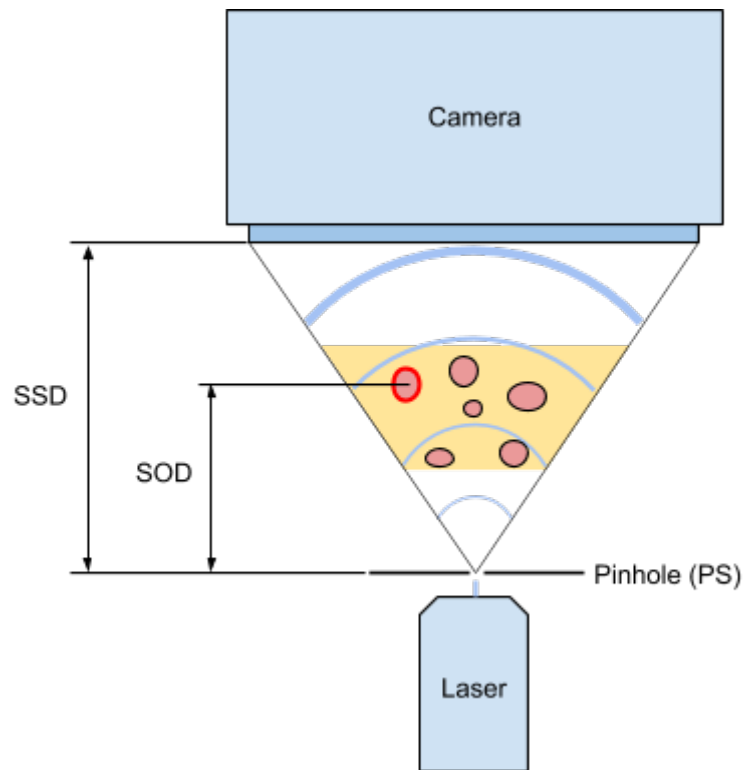


Image resolution is dependent on the SOD (Source to Object Distance). As this distance decreases, the resolution and the magnification increase. Maximum resolution is obtained by placing the sample/object as close as possible to the Point Source (PS). However, keep in mind that as the SOD distance decreases, the field of view becomes smaller.

The distance between the camera sensor and the point source - Source to Screen Distance (SSD) in the Submersible Microscope is fixed, and is

based on the ordered microscope configuration. The standard configuration provides 34 mm SSD, and offers 8 mm of Sample Space. Contact Resolution Optics for the development of custom configurations for special applications.

In order to achieve accurate images, inline holography requires that a reasonable amount of the reference wave (unscattered light) reaches the camera sensor. Too many objects between the PS and the camera can reduce the amount of reference wave reaching the camera and will therefore result in poor reconstructions.

For moving objects, keep the exposure time of the camera as short as possible while still maintaining a visible image. Long exposure times will result in blurry or smeared images. The exposure can be adjusted in the camera control of the Octopus software.

The PS does not produce a large amount of light; therefore background light from external sources must be reduced in order to obtain optimal images. Excessive background light can wash out the images and produce low contrast reconstructions.

If possible, always obtain a background image (a hologram containing only the light from the PS, without a sample/object causing scatter). A good background image will allow for background subtraction when reconstructing holograms, producing higher contrast reconstructions while reducing noise.

Care and Maintenance

Your Submersible Microscope requires very little maintenance. However, periodic cleaning of the windows must be undertaken to ensure the best possible images. It is recommended that after every experiment the microscope is washed in clean fresh water to minimize the potential for corrosion.

Troubleshooting

Problem: Camera is not detected by the software.

Solution: Make sure the cable connector is properly connected to the bulkhead connector. Make sure the camera cable is connected to the

Ethernet port on your computer. Make sure the camera power supply is connected to wall power and turned on.

Problem: No light from the point source.

Solution: Make sure the cable connector is properly connected to the bulkhead connector and is also plugged into a power source. Make sure the laser power supply is connected to wall power and turned on. If there is no light coming from the point source and it is properly connected there may be an alignment problem with the laser or the laser diode may have burned out. Send the microscope back to the manufacturer for re-alignment.

Specifications

Resolution	1 micron
Power	5 Watts
Size	30 cm x 10 cm
Weight (air / water)	1.8 Kg / 1.5 Kg
Data transfer	Gigabit Ethernet
Computer operating system	Windows XP SP3 or later
Communication & power	13 pin SeaConn
Hologram size	from 2048 x 2048 to 512 x 512
Frame rate	from 16 to 50 fps
Flow chamber space	8 mm