LISST-HOLO

Operating Manual

LISST-HOLO version 1.0

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SEQUOIA

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Technical assistance

To obtain technical assistance related to your LISST-HOLO, please contact your local distributor (a list of our international distributors can be found on http://www.SequoiaSci.com/contact/international.aspx?SectionName=contact) or a Sequoia Technical Service Representative listed below.

Please be sure to include the instrument serial number with any correspondence.

Sequoia Technical Service Representatives

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Contents

1 Introduction

Dear LISST-HOLO Customer:

Thank you for purchasing the LISST-HOLO Digital Submersible Holographic Camera!

The intent with this introduction is to get you started using the LISST-HOLO as quickly as possible. Sequoia's goal with the LISST-HOLO is to make the user interface so friendly that no manual would be required for normal operation, except as a first-time introduction to the instrument.

The LISST-HOLO was shipped to you in a Pelican case with custom foam padding. Included in the shipping box are the LISST-HOLO, clamps, stands, External Memory Module with USB cable, Ethernet cable, Wireless Router, Serial Cable with Power Supply, and a spare 5-pin cable for connecting the LISST-Holo to alternate power source such as CTD.

Take the instrument out of the box and place it on a level surface using the plastic stands provided.

Sequoia Scientific, Inc. is pleased that you have chosen one of our products. Thank you! We strive to make our instruments rugged, reliable, and simple to operate. If you should have any difficulties or questions regarding this product, please feel free to contact us.

2 Contents and Technical Specifications

1) LISST-HOLO
2) Instrument stands
3) Clear box with power supplies and communication cables
4) Wireless Router
5) Chamber for testing
6) Manual and CD with software
7) 2 clamps (not shown)
8) 5-pin pigtail for external power (not shown)

2.1 General Features

- In-situ digital in-line holographic technology
- Self-contained with internal datalogger
- Ethernet connection to PC for programmable data collection—no software is needed for programming or offloading data
- Power via external battery pack (Optional, not included) or external power source (cable provided)
- Programmable data collection
- Optical path length: 50 mm standard
- Sample volume: 1.86 cm3
- Data processing yields in-focus particle images and volume distribution
- Automated firmware updates possible when instrument is connected to the Internet

2.2 Parameters Measured

- Particle images for observation and classification
- Particle volume distribution
- Temperature
- Depth

2.3 Particle Size and Concentration

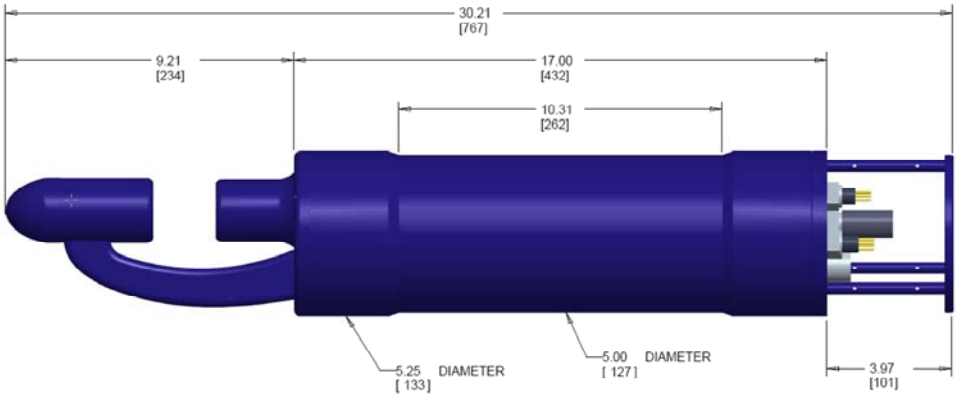
- Size range: 25-2500µm equivalent spherical diameter
- Concentration Range: Beam attenuation Coefficient of 0-4 m^{-1}.
- Resolution: Log spaced size classes, 18% bin widths.

2.4 Technology

- Solid state diode laser @ 658 nm
- 4.4 µm pixel size digital camera; 1600 × 1200 pixels

2.5 Mechanical and Electrical

- Dimensions 13.3 cm (5.25") ∅ × 75 cm (29.5") L
- Weight: 9.5/3.6 kg (21/8 lbs) in air/water
- 300 m depth rating
- 32 GB internal solid state drive
- 32 GB external USB flash drive (included)
- Battery life (Optional External Battery Pack): 10,000 holograms for standard 9V, 42Ah battery
- External power input: 9VDC nominal, 6-15VDC
- Sampling frequency up to 2 Hz



3 Getting Started

3.1 Introduction

This section is designed to enable the first-time user to unpack the LISST-HOLO and capture a hologram within a few minutes after opening the case for the first time. The experienced user may find this section useful for a quick review of the various procedures.

Section 4 describes how to install and use the software for processing the raw holograms.

Section 5 discusses how to configure and deploy the instrument in the field.

It is not necessary to install any software to go through this section and become familiar with the instrument and its operation.

3.2 Connect cables and power up

In order to program and offload data from the LISST-HOLO you need a PC and access to a power source.

Take out the instrument stands and place the LISST-HOLO on the stands.

Take out the clear plastic box and open it up.

The yellow cable is an Ethernet cable that is used for communication with the LISST-HOLO

The black cable is a 5-pin cable that is being used to power the LISST-HOLO when working in the laboratory. The 5-pin cable is being used to provide power to the LISST-HOLO; it is NOT being used for communication: The yellow Ethernet cable must be used for communication.

Look at the LISST-HOLO connector endcap.

Unscrew the dummies and connect the 5-pin cable to bulkhead connector 1 (just below the serial number) and the yellow Ethernet cable to bulkhead connector 2 (just above the serial number).

The External Memory Module and the Ethernet cable share the 8-pin bulkhead connector. Only one can be connected at a time.

The cables are now correctly connected to the LISST-HOLO.

It is necessary to have BOTH cables connected in order to power and communicate with the LISST-HOLO.

Connect the Ethernet cable to the wireless router.

DO NOT use the port to the far left.

Connect the router power cable to the router

Plug the router power supply into a 110-220VAC power source.

When the router completes its booting procedure it can be located as *LISST-Holo* on a wireless network. The password for connecting to the router is m*anyholograms* (this information is also located on the bottom of the router). Establish a Wi-Fi connection between the router and your PC before you proceed. If you do not have a Wi-Fi enabled PC, use the black Ethernet cable for connecting the router to your PC.

When you are connected to the router, plug in the power to the DB-9 connector.

NOTE: It is not necessary to have the DB-9 connector attached to the PC

IMPORTANT: MAKE SURE THAT THE POWER CABLE IS FIRMLY SEATED IN THE CONNECTOR.

EVEN MORE IMPORTANT: NEVER UNPLUG THE POWER TO THE HOLO WHILE IT IS RUNNING. ALWAYS PUT THE INSTRUMENT TO SLEEP.

Now plug in the power supply to the LISST-HOLO

The LISST-HOLO endcap LED will blink for ~ 30 seconds while the instrument is booting up. When it is ready, the green LED will blink every 5 seconds.

Please note: The LISST-HOLO has no internal power. When power is applied to the instrument, the internal processor is powered on and initiates its boot-up procedure.

3.3 Take a hologram

If you have followed the instructions in section 3.2 the LISST-HOLO is now powered up and connected to your PC. It is time to take a hologram!

Open up a web-browser on your computer, e.g. Firefox, Google Chrome or Internet Explorer.

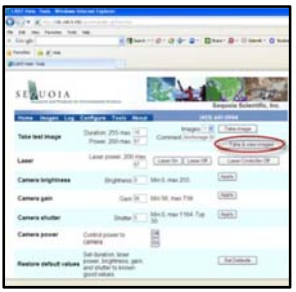
To connect to the LISST-HOLO, use URL http://192.168.0.160

Note: The IP address of the instrument can be changed. See the FAQ section for more information.

The LISST-HOLO main screen will then show.

Use the **Tools** screen to take a hologram.

In order to take a hologram, Click the **Tools** option.



On the tools page, click the **Take & View Images** button to take a hologram.

Note that the laser turns on, then off. It takes a few seconds to capture a hologram in this matter, because the laser has to warm up first. The actual hologram is taken a few milliseconds before the laser turns off.

You are automatically taken to the Images page, where all holograms on board the memory card are listed.

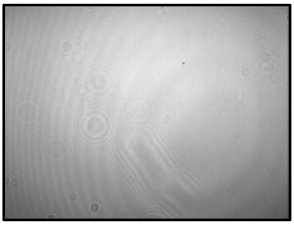
Click the hologram you wish to view. You will be asked if you want to save it to disk or to view it. Select the Save option, so that you can process the hologram. If you want to process a hologram, you always need to save it to a folder on your PC.

Holograms are stored in .pgm format, which is a lossless compression format. You may need to download and install OpenSeeIt (http://openseeit.sourceforge.net/), which

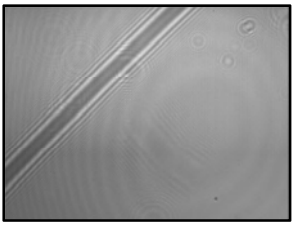
is a viewer for pgm files (see the FAQ on the About page in order to get information about OpenSeeIt).

You can of course also use OpenSeeIt to view holograms you have already downloaded and stored on your computer.

This page shows the FAQ where you can find more information about how download, install and use OpenSeeIt in order to view the unprocessed holograms.



If you took a hologram without anything in the laser beam, your hologram will typically look something like this.



Try taking another hologram, but hold a hair or thin piece of string into the laser beam.

You should then see something like the image to the left.

Take another 1-2 holograms with other types of material n the laser beam, e.g. a screw, or blow some dust through the laser beam when the hologram is captured.

The LISST-Holo comes with a test chamber that can be installed between the windows and be filled with water for testing. To install the chamber remove the U-shaped spacer and press the sliding part into the chamber leaving a small gap. Center the openings of the chamber on the windows and insert the U-shaped spacer so that the o-rings seal on the area around the window.

To put the LISST-HOLO to sleep, click the Home button, then click the Sleep button.

You can now safely disconnect the power to the LISST-HOLO.

To continue Getting Started with the LISST-Holo please read Section 4, Hologram Processing in Detail, or skip to Section 5, LISST-Holo Field Deployment.

4 Hologram Processing in Detail

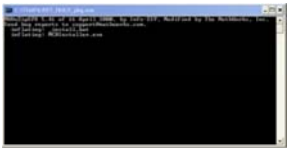
4.1 Intro

The holograms are processed using 2 MATLAB-based programs called Digiholomat and LISST_HOLO. The programs are compiled version of Matlab scripts. Matlab functions and scripts are available upon request. In addition to standard Matlab, the Image Processing Toolbox will also be required.

4.2 Install the hologram processing software

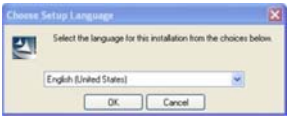
To install the software, insert the CD into your computer, and locate the LISST-HOLO software folder. Inside the folder you will see 2 files: LISST_HOLO_pkg.exe, and Digiholomat.exe

Copy both files to a temporary location on your computer, e.g. C:\Temp.

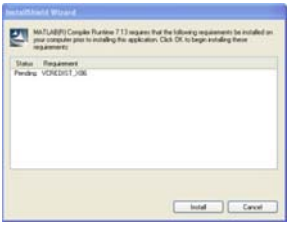


Double-click LISST_HOLO_pkg.exe to begin the installation.

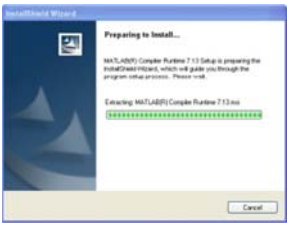
You will see a DOS window open up, detailing the progress of the installation.



Select the language for installation



Click Install to install any additional components that the installer requires.



The installer then prepares to install

Click Next

Enter your User Name and Organization

Confirm the destination folder for the installation of the Runtime Compiler

Click Install to begin

Wait while the installer installs the compiler. This typically takes several minutes

Click finish

Note that the number of files in C:\Temp has increased.

Now create a folder in your Program Files folder called Sequoia\LISST-HOLO (e.g the full path should be C:\Program Files\Sequoia\LISST-HOLO)

Move the LISST_HOLO.exe file and the Digiholomat.exe file to this folder.

NOTE: Be careful not to move the LISST_HOLO_pkg.exe file!

Delete all files in the C:\Temp folder

If you wish you can now create shortcuts for the LISST_HOLO.exe and Digiholmat.exe files and place them on your desktop for easy access.

4.3 Process holograms using LISST_HOLO and Digiholomat

Two different programs are supplied for processing holograms. LISST_HOLO will do batch processing of a selection of holograms, and automatically output the size distribution as well as composite images showing the in-focus particles.

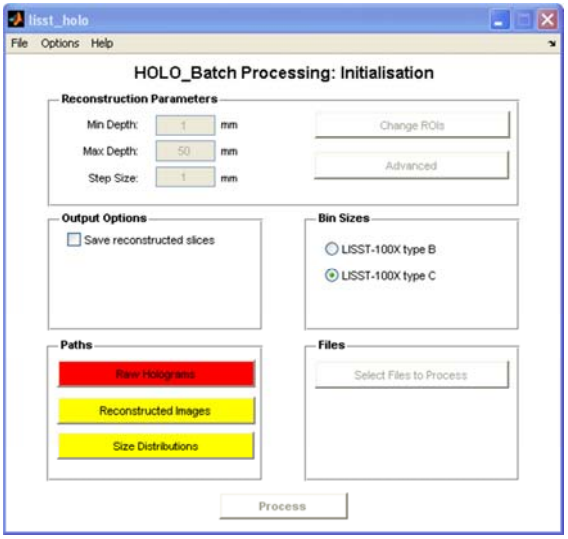
Digiholomat only processes one hologram at the time, but allows the user to zoom and analyze a hologram in great detail, in case more detailed image analysis is needed than what is provided with LISST_HOLO.

In the following sections the use of these 2 programs will be explained using example holograms included in the ExampleHolograms folder on your shipdisk.

Before you proceed, you should copy the 4 holograms in ExampleHolograms (Coffee.pgm, 70%transmission.pgm, 80%transmission.pgm, 90%transmission.pgm) to a folder on your PC.

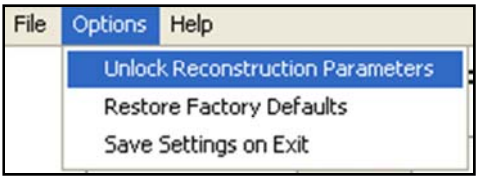
4.4 Batch process holograms (LISST_HOLO)

This section describes how to use the automated batch processing program, LISST_HOLO.



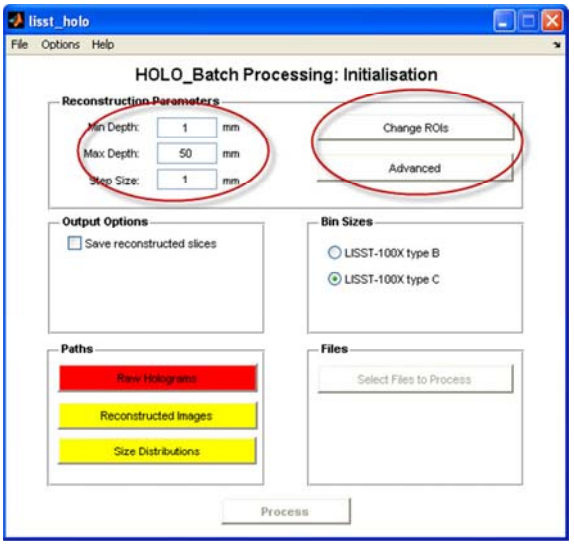
Open the program by clicking the LISST_HOLO executable.

There are 5 main procedures to be performed before processing can begin: Select Reconstruction Parameters, Select Output Options, Select Particle Bin Size, Select Paths (Folders) and Select Files.



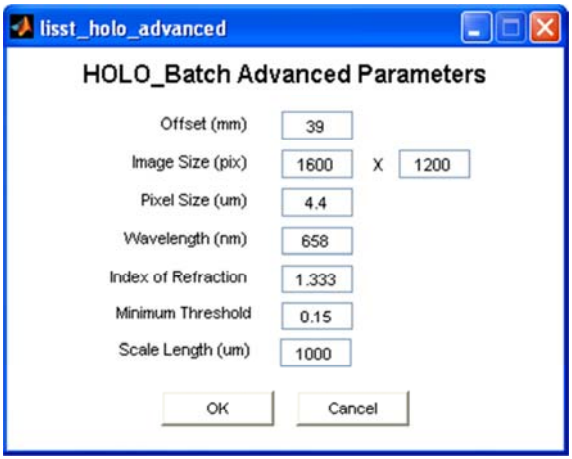
The LISST_HOLO is shipped with factory default settings for the Reconstruction Parameters, and these should normally not be changed.

However, if necessary they can be changed by selecting Unlock Reconstruction Parameters in the Options Menu



The Change ROIs and Advanced buttons then become active.

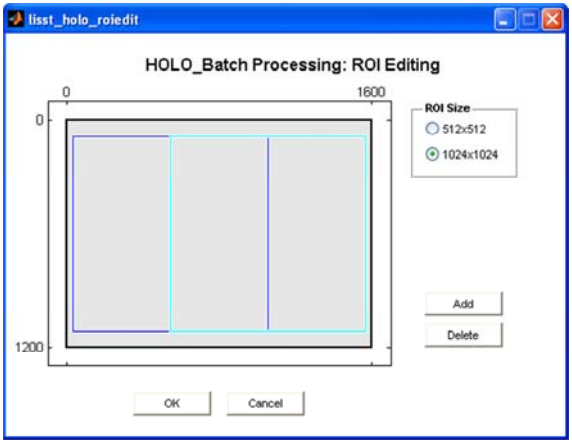
Also the three input boxes for selection of the minimum and maximum focal length, as well as the step size becomes active.



The advanced button opens up the settings menu for the advanced options.

The values listed in the window to the left are the factory default settings.

With the exception of Minimum Threshold and Scale Length, none of these values should ever be changed unless instructed by Sequoia support personnel to do so.

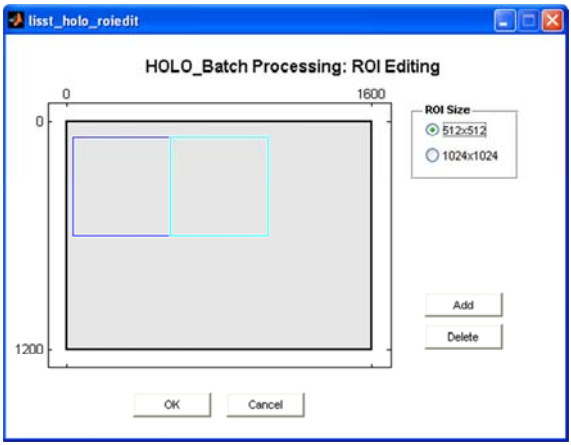


The Change ROIs button opens up the window for selecting the ROI (Region Of Interest) to be processed.

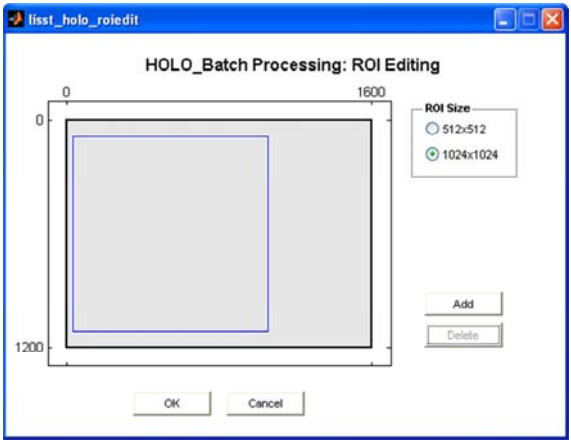
The ROI is the part(s) of the hologram that will be reconstructed. Particle counts will only be computed from the reconstructed part of a hologram.

Once selected, the ROI will be the same for all holograms.

In this example, 2 overlapping ROI's with a size of 1024×1024 pixels are chosen. Particle counts will then be obtained from both ROIs, and adjusted so that particles in the overlapping parts are only counted once.

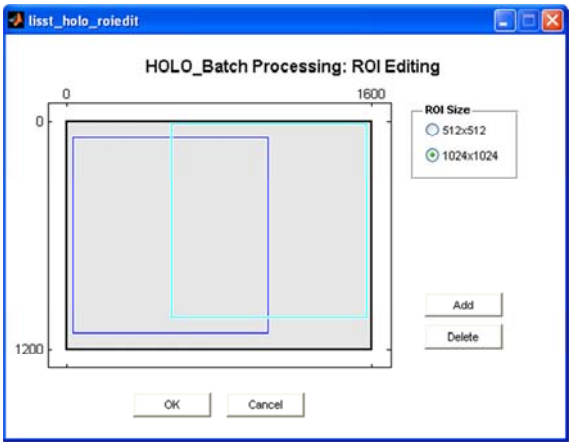


Reconstruction of holograms is memory demanding, so on PC's with smaller memory (<4GB), or slower processors (<2GHz), the 512×512 pixels ROI should be selected.



Note that you can delete and add ROI's using the Delete and Add buttons.

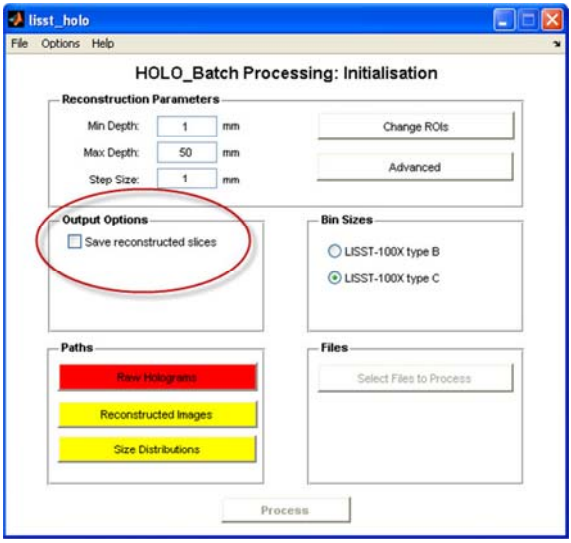
In this example to the left, one of the 1024×1024 pixels ROI's have been deleted.



In this example, another 1024×1024 ROI has been added.

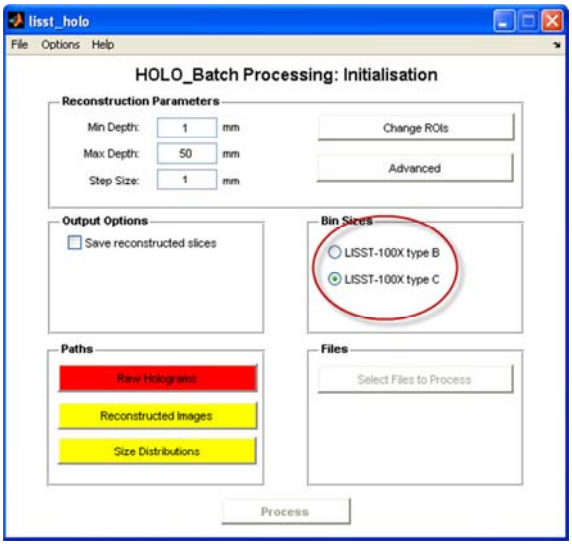
Note that the ROIs do not have to be aligned with each other.

Click the OK button when the ROI size and location is satisfying.



The next step is to select if output of individual, reconstructed slices of the hologram are to be saved.

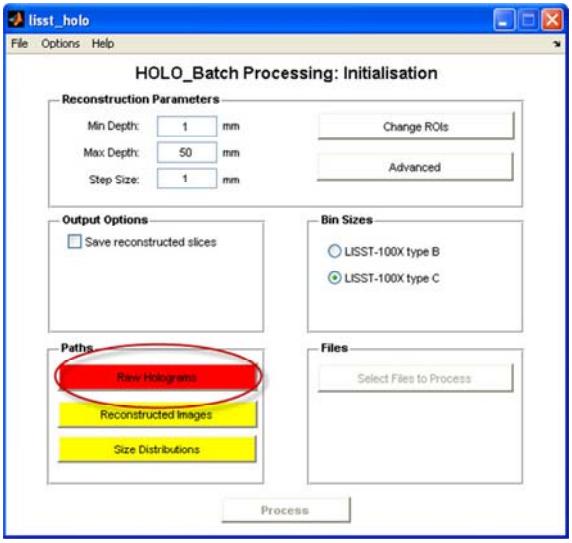
This is extremely memory-consuming so this option should only be selected on PC's with more than 4GB memory.



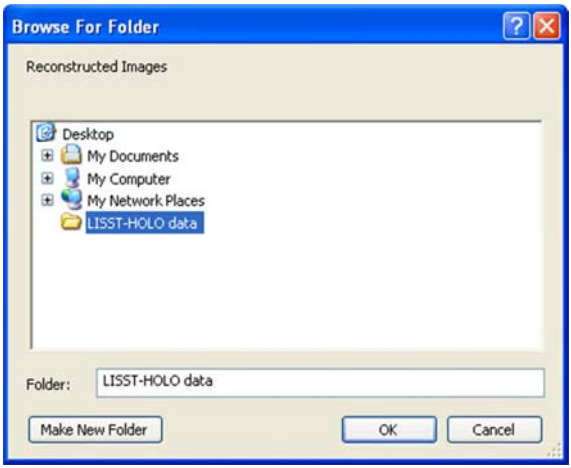
Select the LISST-100X equivalent size bins for your output size distributions.

The size bins for the LISST-HOLO are based on the LISST-100X so that data from the 2 instruments can be easily merged.

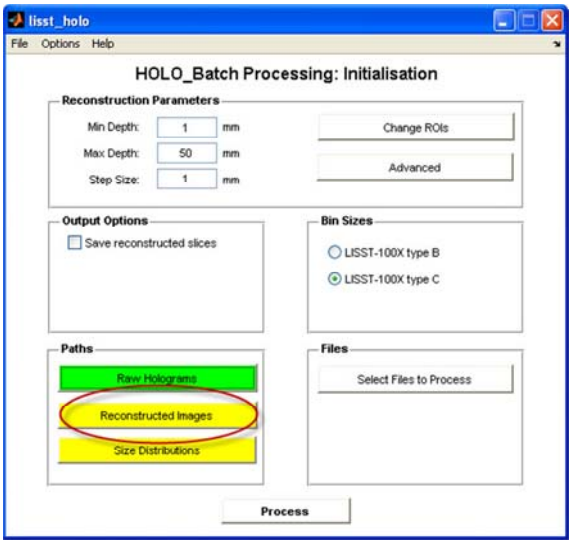
While the LISST-100X has an upper size bin of 250µm (type B) or 500µm (type C), the LISST-HOLO simply expands the upper limit of size bins up to 3000 µm (see appendix xx for details).



Now click the Raw Hologams button in order to select the path where LISST_HOLO will read your raw holograms.



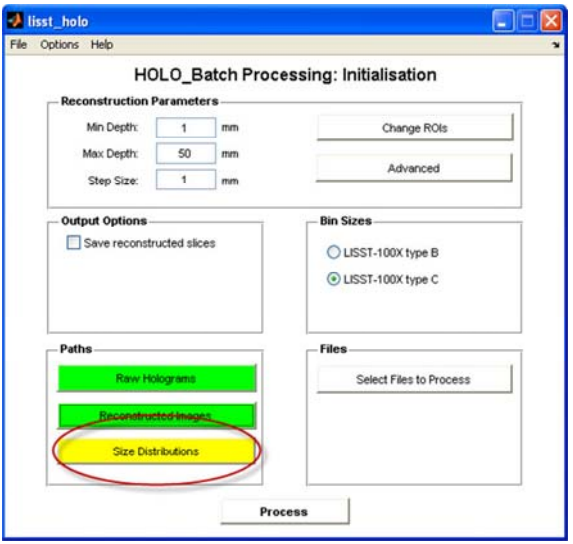
Select the folder where your raw holograms are stored and click OK.



Now click to select the folder where you want the reconstructed images to be stored.

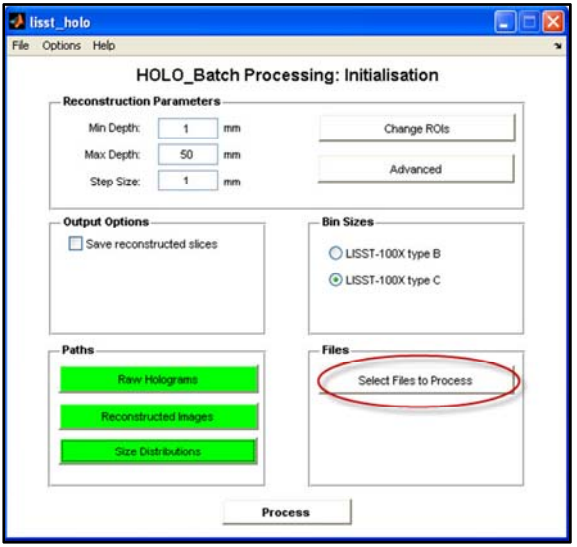
There are two reconstructed images (.jpg format) for each processed hologram.

As above, a dialogue box will open up prompting you to select a folder.

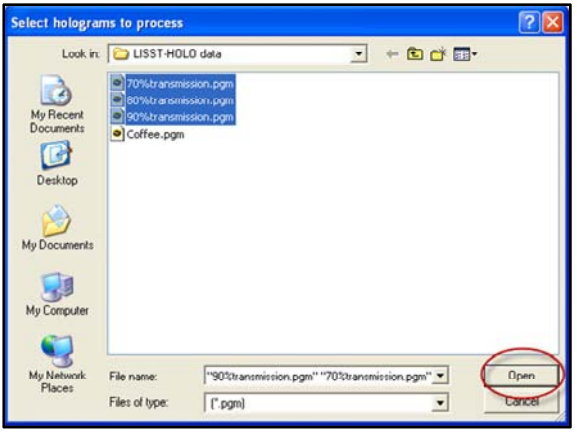


Finally, click the Size Distributions button to select the folder where you want the particle size distributions to be stored.

The output particle size distributions are in .csv format and can be read into most spreadsheets, e.g. Excel or Numbers, or MATLAB.

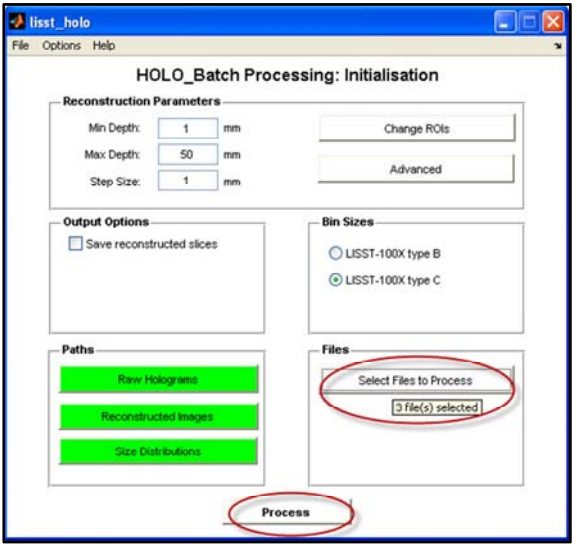


Now click Select Files to Process in order to select the holograms to Process.



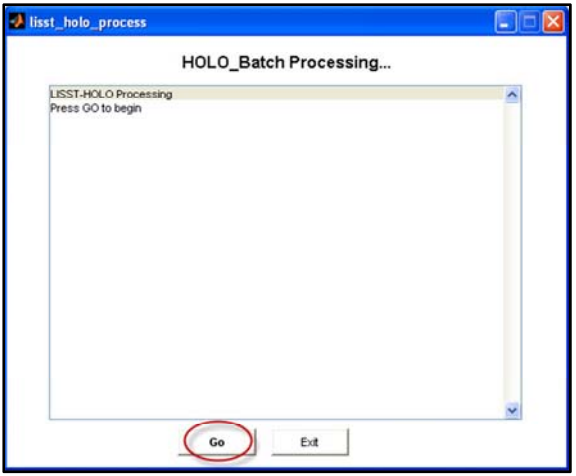
Select the 3 files named 70%transmission.pgm, 80%transmission.pgm and 90%transmission.pgm.

Then click the Open button.



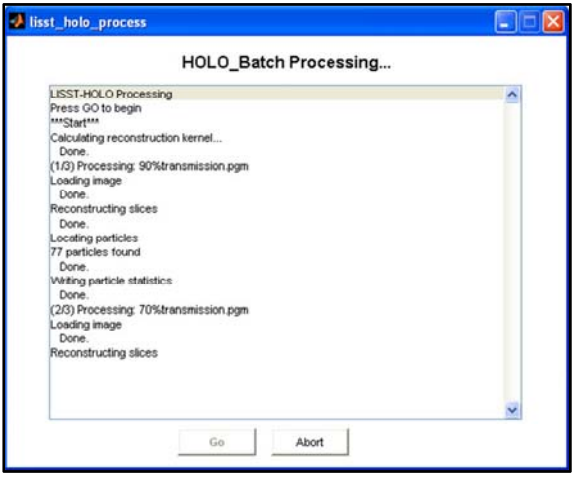
Note that if you now hover the mouse above the Select Files to Process button, you will see how many files (if any) are selected.

Click the Process button to begin processing.



A status window opens up. During processing, the progress can be followed in this window.

Click the Go button to start processing your holograms.



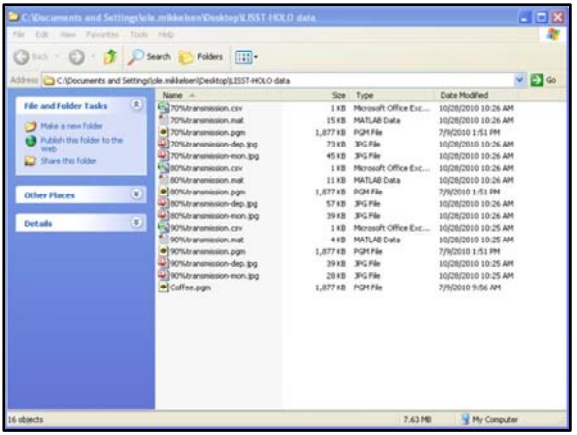
The typical processing speed is about 30 seconds per hologram for a 512×512 ROI.

However, the processing time varies strongly with the number of slices, total ROI area and the number of particles in a reconstructed hologram.

Processing times of up to 2 minutes per hologram is not uncommon for 1024×1024 ROIs with a large number of particles.

If you wish to cancel the processing, click the Abort button.

When processing is complete, the Abort button changes to Exit. Click to Exit the program.



Go to the folder(s) you specified as output paths for your processed data.

You will notice that 4 files have been created for each hologram; 2 .jpg files, a csv file and a .mat file.

In the following these files are briefly described.

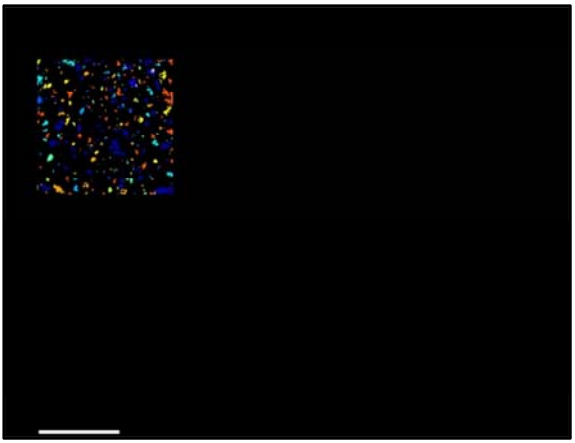
4.4.1 .JPG Files output by LISST_HOLO

The 2 .jpg files have a –dep and –mon suffix.

The –mon suffix image is a montage of all the particles that were found in all the planes within an ROI.

The scale bar length can be adjusted prior to processing in the Advanced section of LISST_HOLO.

Note that only the part of the hologram covered by the ROI is displayed on the –mon image. The rest of the hologram was not analyzed and therefore cannot bed displayed.



The –dep suffix image is a color coded image, where the particles within the ROI have been colored according to their depth of focus.

As with the –mon image, only the part of the hologram covered by the ROI is displayed on the –dep image.

Using a pair of 3-D glasses will display the particles in a 3-D effect.

4.4.2 .CSV files output by LISST_HOLO

The .CSV (Comma-Separated-Values) file that is produced for each hologram contains detailed information about the mean and median size of the particle size distribution (computed from the volume distribution)

Column #	*Value*	*Unit*
1	Year (YYYY)	N/A
2	Month (MM)	N/A
3	Day (DD)	N/A
4	Hour (HH)	N/A
5	Minute (MM)	N/A
6	Second (SS)	N/A
7	1/100s of a second (hh)	N/A
8	Depth	m
9	Temperature	°C
10	Total Volume Concentration	µl / l
11	Total Cross-sectional area	cm^2 / l

12	Total Number concentration	l^{-1}
13	Mean size	µm
14	Mean size	Φ (phi)
15	Standard Deviation	Φ (phi)
16	D10	µm
17	D16	µm
18	D25	µm
19	D50	µm
20	D60	µm
21	D75	µm
22	D84	µm
23	D90	µm
24-30	Reserved for future use	N/A
31-80	Volume Concentration in each of the 50 LISST-HOLO size bins	µl / l
81-130	Area Concentration in each of the 50 LISST-HOLO size bins	cm^2 / l
131-180	Number Concentration in each of the 50 LISST-HOLO size bins	l^{-1}

4.4.3 .MAT file output by LISST_HOLO

The MATLAB .MAT file output contains a matrix called partstats. It has dimensions N×10, where N is the number of particles detected in the ROI. The 10 columns have the following

information about each particle:

Column #	*Particle Property*	*Unit*
1	Particle Area	μm^2
2	Particle Equivalent Diameter	µm
3	Major Axis Length	µm
4	Minor Axis Length	µm
5	Solidity	Dimensionless
6	Eccentricity	Dimensionless
7	Filled Area	μm^2
8	Convex Area	μm^2
9	Equivalent Convex Area Diameter	µm
10	Particle Volume	µm3

5 LISST-Holo Field deployment

5.1 Introduction

The LISST-Holo has many features that make it easy to deploy in the field. Similar to the LISST-100X, Start and Stop conditions can be programmed into the instrument in advance. Once the instrument is started it will wait for the Start Condition to occur before saving holograms. It will continue to save holograms at the programmed rate until the stop condition is reached. The holograms are stored on external submersible USB Flash memory module. There are some differences from the LISST-100X. On the LISST-Holo up to four different set of Start, Stop, and Sampling conditions can be pre-programmed. By using the external switch lever the desired set of conditions can be selected without connecting the instrument to a computer. In addition the switch lever is always used to start the selected sampling program. The sections below describe how to configure, start, and stop the data collection.

5.2 Pre-Programming

To configure the Start and Stop conditions you must connect to the instrument using the Ethernet connection as described in Sections 3.2 and 3.3. Clicking on the Home tab will show the current settings for the four sets of sampling conditions. Each of the four sets can be configured differently. The Start Condition, Sampling mode and rate, and the Stop Condition will need to be set.

To set the Start condition choose between a Date/Time start, delayed start, depth start, External Signal 1, External Signal 2, or the Magnetic Switch.

Date/Time start will wait until the selected GMT time is reached before sampling is started. If it is past the specified time the sampling will start immediately.

The Delay Start will start after the specified delay.

The Depth Start will wait until the depth is greater than the specified value before starting.

The External 1 and External 2 will wait for the voltage on the selected External input located in the Auxiliary 6-pin connector to be greater than about 2.4V.

The Magnetic switch option will start sampling when the switch is in the 1 position.

The sampling rates and modes also need to be selected. There are two modes: Fixed Rate Mode and Burst Mode. The Fixed rate mode will take one hologram at the specified interval. The Burst Mode can be used to take a short burst of holograms at fixed interval between the start of each burst.

The Stop Conditions also have multiple options: Date/Time, Fixed Duration, Greater than or less than Depth, External Signal 1 or 2, Magnetic switch, or fixed number of Samples.

The Date/Time Stop is used to stop collection at a particular time and date.
The Stop after setting will stop the sampling after the specified amount of time from the start of data collection.
The > Depth value will stop when the depth is greater than the specified depth.
The < Depth value will stop when the depth is less than the specified value.
The External 1 signal and External 2 signal setting will stop collection when the selected signal is less than 0.8 volts.
The Magnetic Switch option will cause sampling to stop when the switch is in the 0 position
The Samples setting will stop data collection when the specified number of samples have been saved.

After making changes to the settings press on the Apply button to save the settings and refresh the page.
All four sets of conditions can be programmed.

5.3 Preparing for Deployment

Now that the sampling parameters are configured we can prepare the instrument for deployment and start the sampling.
The first step is to put the instrument into low power sleep mode so that we can disconnect the Ethernet cable and plug in the External Memory Module. In order for the Memory Module to be recognized it must be in place before power is applied or while the instrument is in sleep mode.
To put the instrument to sleep press on the Sleep button on the Home page or on the Tools page. This will shut down the primary datalogger. The LED on the endcap should now be blinking every 30 seconds instead of every few seconds.
Disconnect the Ethernet cable and plug in the External Memory Module using the special threaded locking part to hold it in place. If the system is still powered up but not awake move the switch to the 1 position and then quickly back to the 0 position.

This will wake up the instrument. It will take about 30 seconds to boot. You will see the LED on the endcap flashing in different states during this process. When the green LED is flashing every few seconds the instrument is awake and ready to start sampling.
IMPORTANT: Regardless of the Start and Stop conditions selected the Switch on the endcap is used to start and stop the sampling mode. However, the actual Start and Stop conditions determine when holograms are stored. For example if a Start Time has been selected, moving the switch to the 1 position will cause the instrument to start looking for this start time. It will continue to sample until the selected Stop condition is reached or until the Switch is moved to the 0 position again.

5.4 Selecting Start/Stop Set

Now that the instrument is ready to start sampling we can select one of the four set of Start/Stop Conditions. This can be done by quickly moving the switch to the 1 position and back to the 0 position multiple times. The number of times the lever is in the 1 position will select the set of Start and Stop conditions to be used. For example, move the switch to the 1 position, back to 0, back to 1, then back to 0. The green LED on the Endcap should now be blinking twice every few seconds. This tells us that the Start/Stop Condition set 2 will be used. To go back to set 1 simply move the switch to the 1 position and back to 0. The LED will now be blinking once every few seconds.

5.5 Start Data Collection

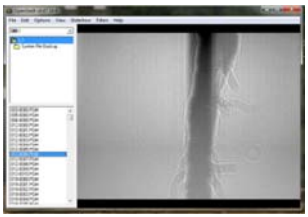
To start the data collection move the Switch to the 1 position. The LED on the endcap will change from green to red indicating that sampling has begun. It will blink slowly until the start condition is reached. It will be on continuously during the acquisition of a hologram. It will return to the green LED when the stop condition has been reached.
Remember: Moving the Switch to the 1 position will start the data sampling regardless of the actual Start Condition

setting. Similarly, moving the Switch to the 0 position will stop data collection if it is still sampling.

5.6 Stop Data Collection

Data collection will continue until the selected Stop Condition is reached. To Stop the data collection at any time move the switch to the 0 position. Within a few seconds the LED on the endcap should change from Red to Green. If the data collection has already stopped the LED will be blinking green regardless of the switch position. However, the switch should be placed in the 0 position when sampling is complete.

5.7 Downloading and Viewing Raw Holograms



The raw holograms have been saved to the External Memory module. These files can be downloaded using the provided USB cable. Remove the External Memory module from the instrument. Leave the locking ring attached to the connector on the instrument. Plug the mating underwater connector of the USB cable onto the Memory Module and then plug the USB end into the PC. The USB flash drive inside the module should now appear on your PC. You can now transfer files as you would from any USB flash drive.

To quickly view the raw holograms before offloading use the Browser mode of the OpenSeeIt software. If you have set OpenSeeIt to be used for all PGM files you can simply double click on one of the holograms on the Memory Module. You should see the raw hologram you selected. In the View menu of OpenSeeIt select the Browser mode option. Note: the short cut for doing this is just pressing enter when viewing a single hologram. Click on the list of files in the lower left and then use the arrow keys of the keyboard to display the different holograms.

After copying the files from the Memory Module they can be deleted using the standard windows techniques.

5.8 Next Deployment or Powering Down

The Memory Module can now be re-installed on the LISST-Holo and sampling can be started again by moving the switch to the 1 position again.

There is no need to connect the Ethernet cable and establish communication with the instrument unless the settings for the Start and Stop conditions need to be changed.

If the instrument is not going to be redeployed it can be powered down. If the instrument is not sampling or writing files the power can be disconnected. However, the safest time to remove power is when the instrument is in its low power sleep mode. The instrument will

automatically go into lower power sleep mode after 10 minutes of no activity. The green LED will blink every 30 seconds when in sleep mode. When the instrument is awake but idle the LED will blink every 5 seconds.

6 Reviewing Instrument Info and Other Settings

6.1 Intro to other Settings

In addition to the Start and Stop Conditions there are other parameters that can be changed using the Ethernet connection. This section will discuss some of the additional parameters that can be accessed from the home page.

6.2 Home

The Home page shows the Start and Stop conditions for the four different sets. In addition it also show System Status information such as available memory on the internal flash drive, input voltage, current depth and temperature, endcap switch position, and the state of the two digital inputs.

6.3 Images

In addition to the External Memory Module the LISST-Holo has an internal flash drive. When the External Memory Module is not attached the saved files will be stored to the internal memory. The Images page shows a listing of the files saved to the internal memory.

To see specific information about the file such as date and time, depth, and temperature click on the Details button. Use the Delete button to delete the hologram.

The file name is a hyperlink to the file on the internal drive. Click on the file name to download the file to a temporary folder and open the file in OpenSeeIt. To copy the file to a separate folder right click on the file name and choose “Save Target as” or “Save Link as” and then enter a folder to store the file to.

To download or delete multiple files you will need to use an FTP utility like Filezilla (http://filezilla-project.org).

6.4 Log

The LISST-Holo writes information to a file as it is operating. This file contains information that can be helpful in troubleshooting problems. This file can be sent to Sequoia for evaluation. The Log option of the main menu allows the user view the contents of the log file.

6.5 Configure

Selecting Configure on the home page main menu will bring up a window listing many parameters and allows their values to be changed. Values on this page should not be changed without detailed knowledge of the parameters use or without detailed instructions. By default this page has restricted access.

6.6 Tools

Selecting Tools on the main menu brings up a window that allows the user to take test image and to change additional parameters such as IP address, send Log file for analysis, check for firmware updates, synchronize time with standard time, and other settings. Values on this page should not be changed without detailed knowledge of the parameters use or without detailed instructions.

6.7 About

The About page contains information about the hardware and

firmware loaded in the instrument. It also contains some FAQs including links to the free OpenSeeIt and Filezilla software.

7 Handling, Cleaning, Maintenance and Storage

7.1 Handling

When handling the instrument, at all times please remember that the LISST-Holo is a high-precision optical instrument.

Avoid mechanical shocks and impact to the housing, as this may cause misalignment of the optical parts.

7.2 Cleaning

The glass windows of the instrument can be scratched. Do not use abrasive cleaners on glass or anodized surfaces.
When cleaning windows be careful not to drag and hard particles such as sand across the windows.
Use glass cleaner or liquid dish soap to clean windows.

7.3 Storage

The LISST-Holo should thoroughly cleaned before storage.
Store in a clean dry place or inside the provided ship case.
The instrument should be removed from power when not in use.

8 FAQ's

What is a hologram?	Fundamentally, holograms are 3-D images that have first been projected, and then stored on a 2-D surface. In the type of holograms captured by the LISST-HOLO (so-called in-line holograms), a laser beam is emitted and a small part of it is being scattered by the particles in the water. The part of the laser beam that is not being scattered by particles interferes with the scattered light. This creates an interference pattern - a hologram. The interference pattern is captured by a small digital camera. Upon processing of the captured interference pattern (i.e. the hologram), it is possible to recover the original image that created the hologram. Thus, the LISST-HOLO does NOT capture and store an image of the suspended particles. It is the hologram of the particles that is stored. During data processing, the image of the suspended particles is reconstructed from the hologram. Subsequently standard image processing methods can then be applied to get information about particle shape, size, area etc.
Why can't the LISST-HOLO be used when the flow speed is larger than 0.5 m/s?	It takes a few micro-seconds to capture the hologram using the digital camera. The image captured by any digital camera is made up by a large number of pixels. If the particles in the water move more than half a pixel size during the time it takes to capture and store the hologram, it is impossible to process the hologram and get reliable results. For the pixel size used in the LISST-HOLO, a flow speed of more than 0.5 m/s will begin to blur the hologram. Consequently, if the LISST-HOLO is used on a CTD, the CTD speed must be 0.5 m/s or less. Likewise, if the LISST-HOLO is deployed on a mooring or tripod, the current velocity around the instrument must be less than 0.5 m/s.
Why should I connect the LISST-Holo to the Internet?	Connecting the LISST-Holo to a local area network that is connected to the Internet has benefits. The first benefit is that the instrument is access from different locations across the network. Connecting the instrument to the Internet has other advantage such as checking for firmware updates, syncing the time with international standards, and transmitting log files to Sequoia. It is not necessary to have the instrument connected to a network or the Internet but it can be useful when needed.
How do I connect the LISST-Holo to the Internet?	The LISST-Holo comes configured with a pre-selected IP address, Netmask, Gateway, DNS 1, and DNS 2. These setting will work fine with the provided Wireless router but the will probably not work with your existing local area network. Because the LISST-Holo requires a static IP

	address an IP address must be selected so as not to interfere with other computers on the network. It is highly recommended that users work with their network administrators to determine the proper settings for their network. The new value can be entered on the Tools page. Be sure to double check the values before pressing the Apply button. Once the Apply button is changed the IP address will change and you will need to log into the new IP address before communication can be established. If the incorrect values are entered it is possible to change them manually through the serial port connections. Contact Sequoia for details of this procedure.
Why does it take 2 minutes to process one hologram?	Processing (or reconstruction) of a hologram is a mathematical intensive task. Therefore a LISST-HOLO hologram is split up into 4 sub-holograms, which are each processed separately. This is in order to avoid memory problems during processing. One the 4 sub-holograms have been processed they are merged into one image with particles and processed for size, area, shape etc. The 2 minute processing time per hologram is a maximum expected time on a standard 2.2GHz windows PC with 4GB of memory. However, since the processing is fully automated, it is possible to process holograms overnight or during weekends.
Is it possible to get data in real-time from the LISST-HOLO?	No. It typically takes 2 minutes to reconstruct a hologram into an image that can be analyzed for particle size, shape, area etc.

9 Ethernet Communications

9.1 Router setup

A static IP address is required for access to the LISST-Holo via a network. When the LISST-Holo may conflict with the user's or administrative concerns prevent connecting the LISST-Holo to the user's network, a router is required to communicate with the LISST-Holo.

The LISST-Holo comes with a Linksys E1000 router that has been programmed to communicate with the LISST-Holo. Plug your computer's Ethernet port and the LISST-Holo into any of the numbered ports on the E1000 router. You may also use a wireless connection to communicate with the LISST-Holo. You must still connect the LISST-Holo to the router; you computer may connect wirelessly. The SSID is LISST-Holo, the WAP PSK is manyholograms.

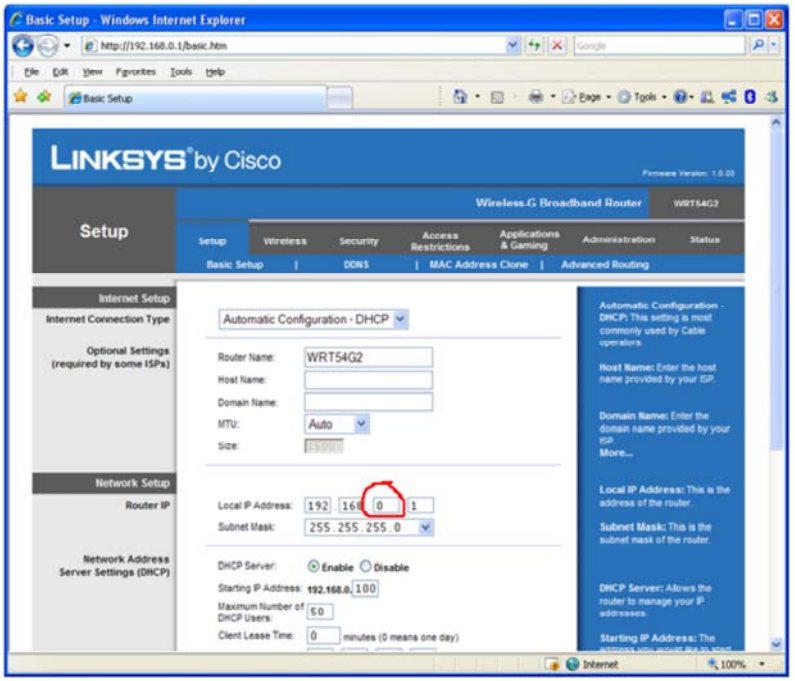
Optionally, the Internet port on the E1000 router can be connected to you LAN to give the LISST-Holo access to the Internet for software updates.

The E1000 router base address: 192.168.0.1. To log on, leave user name blank and enter admin for the password.

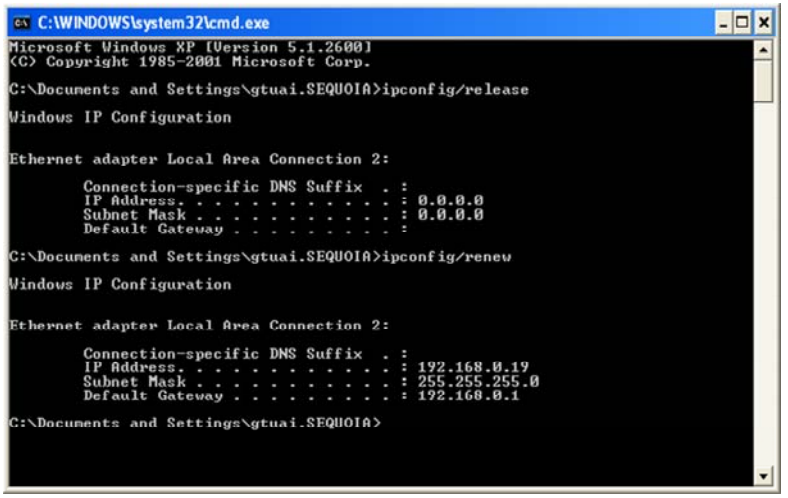
Programming the Linksys E1000 router

Apply power to the Linksys E1000 router.
Connect the Ethernet jack in a computer to Ethernet port 1 of the router. Light #1 should come on.
Open Internet Explorer and navigate to 192.168.1.1.
Leave the user name blank. The password is *admin*.

Change the Local IP Address from 192.168.1.1 to 192.168.0.1 unless the customer has requested a different address.

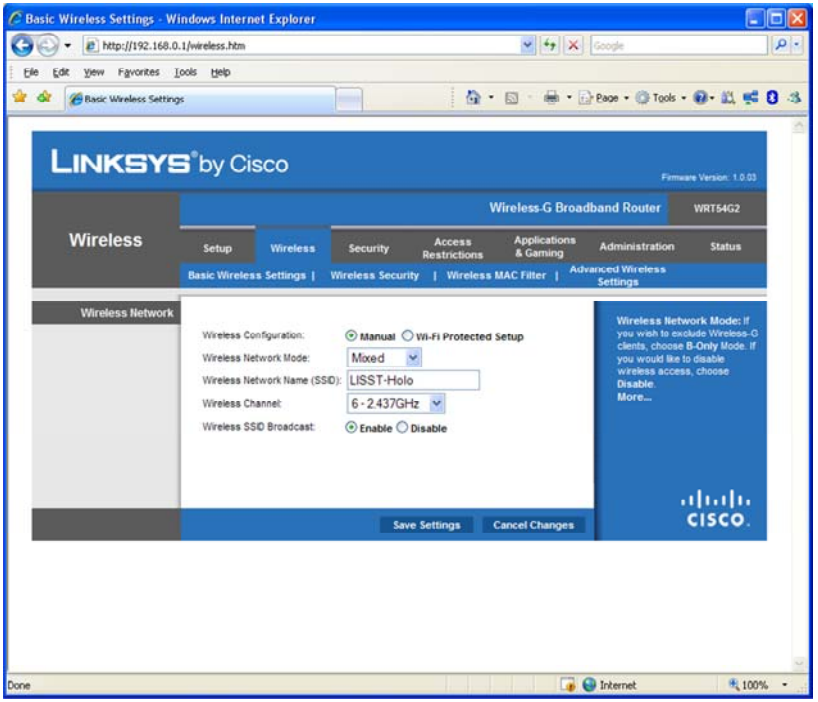


Click on Start/Run, type cmd, and click on OK. You should see a black window.
Type, `ipconfig/release` and press <Enter>.
Type, `ipconfig/renew` and press <Enter>.
Note that the IP address is now 192.168.0.xxx rather than 192.168.1.xxx.



Go back to Internet Explorer and change the URL to 192.168.0.1 or the address the customer has requested.

Click on the Wireless link. Under the Basic Wireless Settings, select Manual and enter the “LISST-Holo” for the SSID. Click on Save Settings.



Click on the Wireless Security link. Select WPA Personal. The WPA shared key is "manyholograms." Click on Save Settings.

9.2 Changing the IP settings via Serial Port

There are two ways to communicate with the Holo. The easiest way to change the IP information is via the web interface. Use a standard browser such as Internet Explorer or Firefox. Connect the Ethernet cable to the end cap. Plug the RJ45 connection into a hub. The default address of the Stokes is 192.168.0.151. Enter the IP address of the unit. If you do not know the IP address, you must use console communications below.

Linux console communication is available through the serial port on the end cap. Use a terminal emulator program such as HyperTerm set for 19.2K, 8 bits, 1 stop, no parity, no flow control. Use VT100 emulation. Under File/Properties in HyperTerm, click on the Settings tab. Uncheck, “Send line ends with line feeds.” The user name is root, the password is “password.”

The IP information can be changed from the serial console. After logging on, use the cip script to update the parameters. The format for the command is show below. All of the IP addresses must be present and separated by spaces. If DNS2 is unknown, duplicate DNS1.

cip <ip> <netmask> <gateway> <DNS1> <DNS2>

For example, if you wish the Holo to have the following IP information,
IP: 192.168.1.14
Netmask: 255.255.255.0
Gateway: 192.168.1.1
DNS IPs: 192.168.1.1 and 98.76.54.123

Enter the command:

```
cip 192.168.1.14 255.255.255.0 192.168.1.1
192.168.1.1 98.76.54.123
```

The LISST-Holo does not support DHCP.

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