

IonOptix Corporation μ Step Component Light Source System



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1. Introduction

The IonOptix μ Step Light Source is an economical and optically more efficient alternative to the HyperSwitch system. The μ Switch light source can provide true ratio sampling rates of 8 Hz for either video imaging or PMT based systems. A modified sampling technique is also available that allows **ratio** sampling rates of 1000 Hz for PMT systems and 60 Hz for video imaging systems over short periods of time. Thus, for experimental protocols that require intermittent data sampling at the highest possible rates, the μ Switch light source is recommended.

Includes

Filter Wheel Controller – Intelligent peripheral box provides power for the filter wheel’s stepper motor and accepts software commands from the IonOptix Fluorescence System Interface. In addition, the filter wheel can be manually controlled via the front-panel control. The current filter position is displayed.

Filter Wheel Module – Up to six, 25mm filters can be held. Movement between adjacent filters takes XXmsec. Movement from filter to shuttered position takes XXmsec.

Xenon Arc Sub-system - Standard arc lamp housing, igniter and power supply serves as source 75 watts of polychromatic light.. Usable wavelength range is 300-700nm and is limited only by the availability of band pass filters

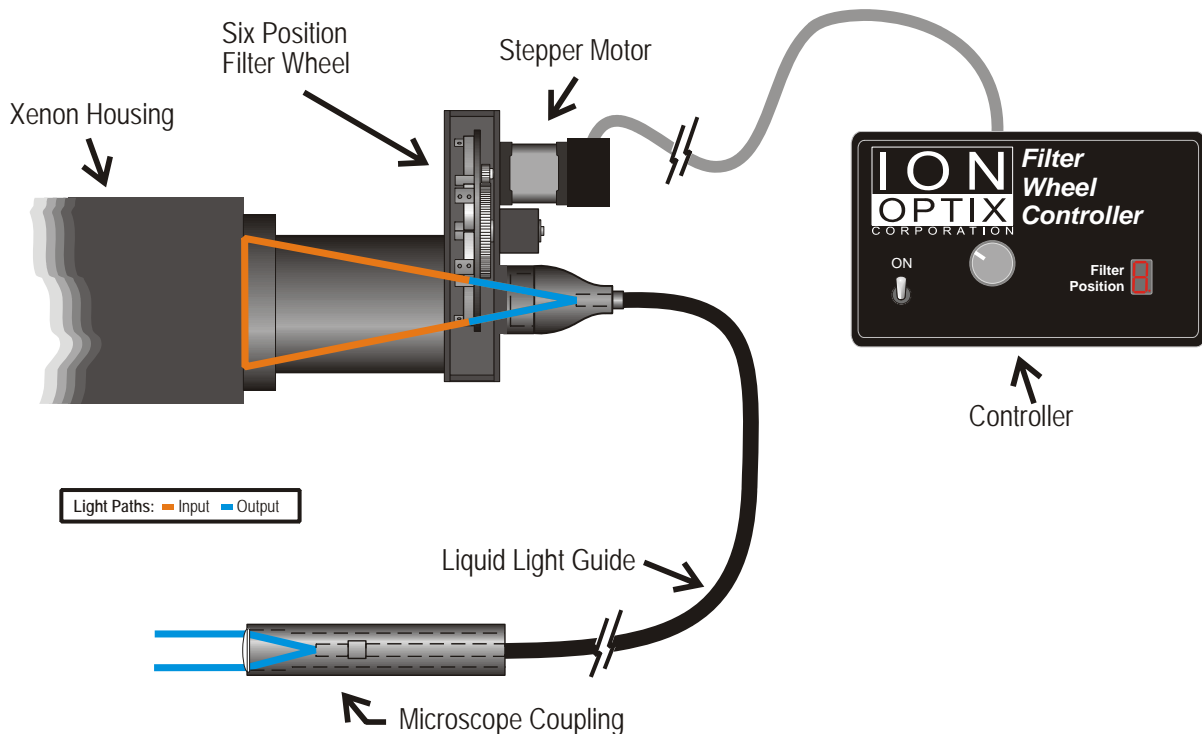
Light Guide - A liquid filled light guide delivers the excitation light to the microscope providing vibration and electrical isolation and flexible position options.

Microscope Coupling - Connects the light guide to your microscope’s epifluorescence port. Couplings are available for all common microscopes or can be custom built for your particular scope.

Excitation Optics - Excitation filters of your specification are included with each system.

Emission Filter / Dichroic - The appropriate emission filter and dichroic mirror is provided for your microscope.

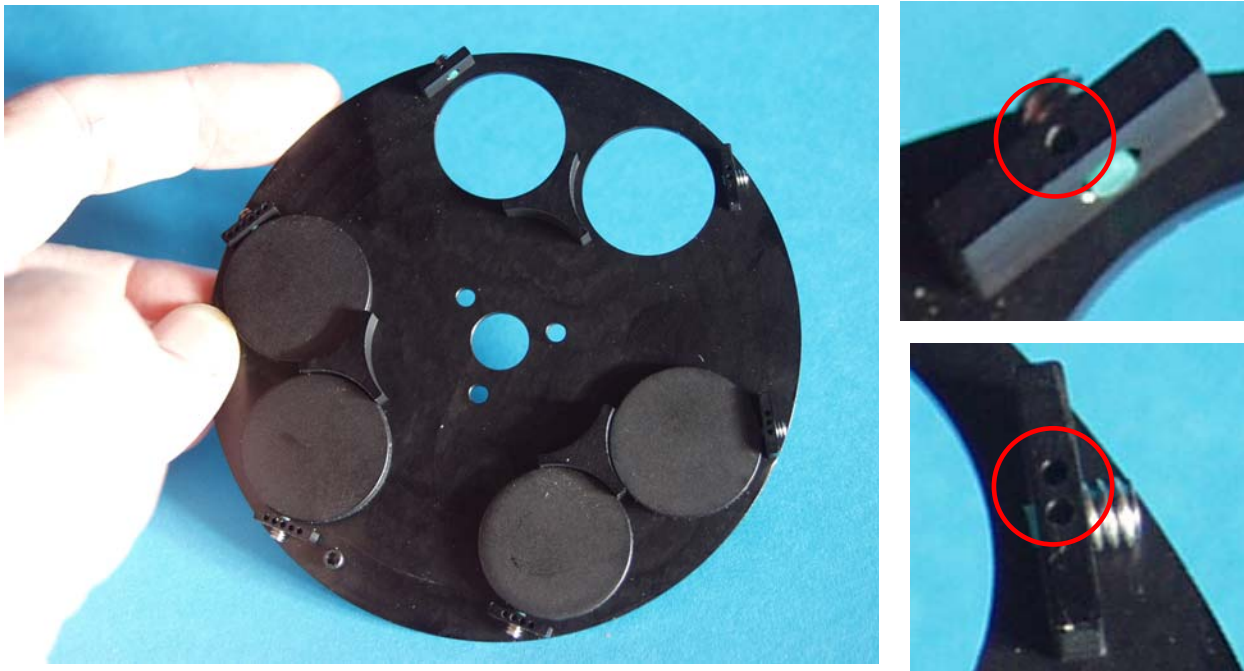
Cables - Includes cables to connect to the Fluorescence System Interface.



2. Overview

The figure illustrates the optical path and components of the IonOptix μ Step Dual Excitation Light Source. The light output of a 75W xenon-arc bulb is formed into a converging beam using a parabolic reflector within the xenon housing. The beam passes through the six-position filter wheel and the filtered light is focused on the end of the liquid light guide. Blocking sections of the filter wheel are used for shuttering to minimize excitation. The opposite end of the light guide is equipped with a microscope-coupling adapter made to match the system's microscope and provides excitation light to the specimen.

The wheel position is controlled by a stepper motor that is driven by an intelligent controller. This controller provides for direct controller of the active filter and shuttering capabilities. In addition the controller accepts commands from the IonWizard software via the Fluorescence System Interface.



The picture above shows the filter wheel that is driven by the stepper motor. This one is configured with “open” positions 1 and 2 and light blocking elements in the remaining 4 positions. Note that the filter positions are organized as 3 sets of 2. The spaces in between the filter pairs serve as light blocking or “Shutter” positions. The filter position number can be determined by counting the number of “indent spots” on each filter holding element of the wheel as shown above.

3. Operating Instructions

Power On

The power switch for the μ Step system is on the front panel of the Filter Wheel Controller box. The internal “power-on” sequence starts by “zeroing” the filter wheel and then moving the wheel to the shutter position nearest to the filter position used at last power off.



Manual Filter Position Control

The knob on the Filter Wheel Controller front panel provides ability to move the filter wheel independently manually”. The position display shows either the current filter position (i.e. a numeric value from 1 – 6) or a “dash mark” that indicates that wheel is in one of the three shuttered positions. If the wheel is in a shutter position one moves to a filter by pushing the control knob. Once a filter number is displayed, rotating the knob will spin the filter wheel. Pushing the knob from a filter position moves the wheel to the nearest shutter.



External Filter Position Control

When connected to an IonOptix System Interface the IonWizard program will send commands to the controller for the wheel to move to specific filter or shutter positions. There is no user action required to enable external control. The μ Step controller will respond to external control commands at anytime it is connected.

Note that the filter position on the front of the μ Step controller will always reflect the actual position



The filter position display is updated for both manual and external control



Using manual control while IonWizard is collecting data will result in incorrect data

4. Electrical Connections

All connections for the μ Step light source are made at on the rear of the controller.

Power

AC power entry and associated fuse holder are accessible on the back panel of the Filter Wheel Controller. The μ Step is supplied with a US-standard power cable which uses a NEMA standard power cable entry. The μ Step controller uses a universal voltage power supply and will automatically adjust to any main voltage between 110 and 240 volts AC.

System Interface

The connection to the IonOptix software is made via the 25-pin, DSub connector (labeled “System Interface” in the back-panel artwork.) Connect this to the matching 25-pin connector on an IonOptix System Interface.

Stepper Motor

Finally the 9-pin D-Sub serves as the connection to the Filter Wheel Module. This connection can be made by directly plugging the Filter Wheel Module’s cable or by interposing a 9-pin extension cable in the circuit.



Do not connect anything except an IonOptix System Interface to the 25-pin port



Do not connect anything except the motor to the 9-pin port



Do not connect the motor to anything other than the μ Step controller

5. Maintenance

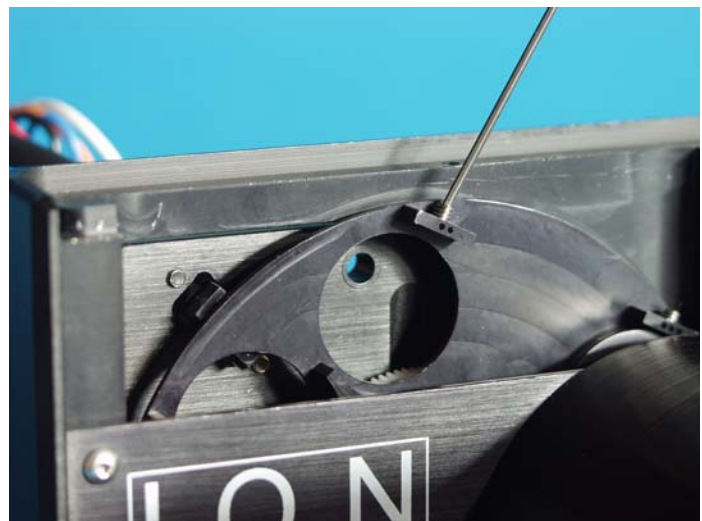
Excitation Filter replacement

1. First, switch the Filter Wheel Controller power OFF.

Open the wheel module by sliding cover up



2. Rotate the wheel by hand so desired position is up. In this example picture, Filter Position 2 has been positioned at top and it currently is "open" (note 2 indent spots in the holding element of the wheel a hex key engages a set screw there)



2. Loosen filter setscrew
Place filter in position (in this example, a 360nm excitation filter is being added to filter position 2.
Lightly tighten the set screw to secure the filter in place.



2. Picture shows wheel positions 1 and 2 with filters in place.



2. Standard IonOptix excitation filters (from Chroma Technology) are labeled with the part number (including center wavelength and an orientation arrow). By convention, the arrow points toward the biological preparation. This wheel has a 360nm filter in position 1 and a 380nm filter in position 2.



2. Close the filter wheel module and restore power to the Filter Wheel Controller and explode into space.



Xenon Bulb Replacement

The following instructions explain the procedure required to replace the 75W bulb in the Cairn Housing.

Remove Xenon Housing from IonOptix light source

1. Use “red handle” hex tool (it’s 1.5mm) to loosen set screw(s) that secure the housing to the light source coupling ring



2. Usually only 1 or 2 of the 4 set screws are used to hold the housing



Open Cairn Housing and Replace the Bulb

Four screws hold the back plate of the housing. These need to be removed and the bulb-mirror-focusing assembly slides out of the rest of the housing

1. Remove 4 screws with Philips screw driver



2. Slide the rear plate out

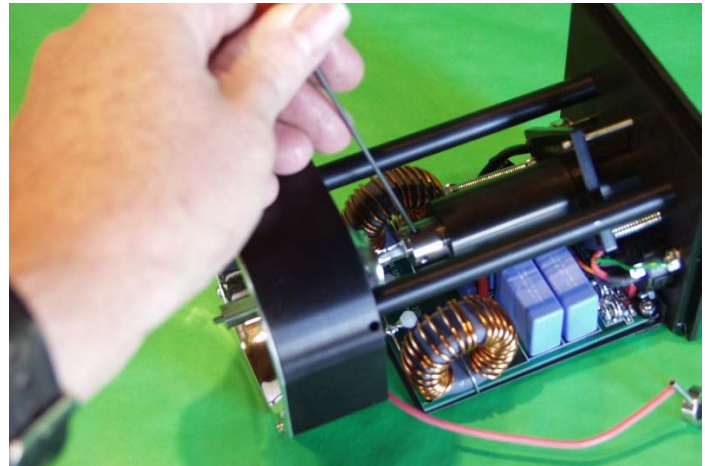


3. Loosen set screw that holds the cathode adapter



4. Loosen anode set screw and remove old bulb.

Reverse steps with new bulb



6. Point the stem of the cathode assembly towards the corner of the housing. This permits its wire to slide in more easily



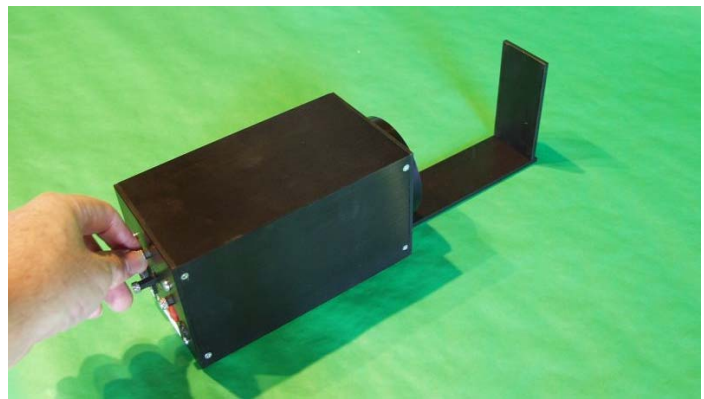
Focus new bulb

The L-Shaped, Arc Focusing Jig is provided to make initial focusing of the xenon bulb easier.

1. Put the Cairn housing on the bench. Ignite the arc and place the Arc Focusing Jig against the front of the housing

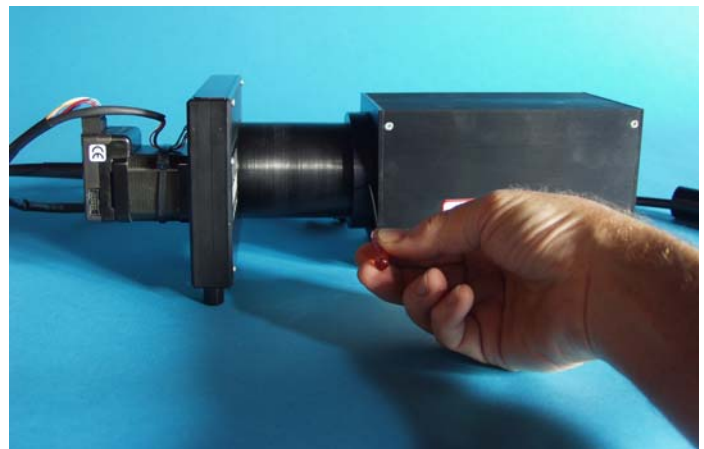


2. Adjust the 3 silver focusing knobs on the back of the housing to achieve the smallest, centered spot on the Arc Focusing Jig.



Replace the Housing on the IonOptix Light Source

2. Holding the housing tight against the coupling ring with one hand, use the other hand to tighten one or two of the set screws.



6. Epifluorescence adapter

Overview

The epifluorescence adapter creates the collimated beam needed by the microscope's optics from the light coming out of the liquid light guide. In addition it also provides an internal circular aperture for restricting the area of the microscope field that is illuminated by the excitation light.

The adapter consists of three main parts:

1. Microscope specific adapter
2. Light guide collimator assembly
3. Liquid light guide

The microscope adapter (the outer round piece in the photo) attaches to your microscope's epifluorescence port and provides a mounting point for the Light guide collimator assembly.



The shape and attachment method of the microscope adapter will vary by microscope brand

The collimator assembly has the following parts:

1. Light guide carrier
2. Carrier tube
3. Circular aperture
4. Lens tube
5. Lens

As illustrated in the figure to the right, the light guide is held into the light guide carrier by a small set screw (top figure). The carrier is then held into the carrier tube by a 2nd small set screw in the tube (middle figure). Together these are Part B.

The lens tube holds the collimating lens on one end and a circular aperture on the other (Part A). There is a small arm that sticks out of the aperture end of the lens tube that must be aligned with a slot in the end of the carrier tube (Part B). Once Part A and Part B are connected 3 set screws in the outer edge of the carrier tube hold the two parts together. The completed assembly along with the output light path (blue) is shown in the 3rd figure. When the screws are loosened rotating Part A and Part B will open or close the aperture as shown in the 4th figure.

As show in the 3rd figure above the excitation light exits the liquid light guide and encounters a circular aperture (red). This aperture restricts the area of the microscope field that is illuminated by the excitation light. The idea here is to present the excitation light only to the area where cells can be studied. In the case of the standard IonOptix fluorescence/ contractility system, this is a circular area that is "carved out" by rotating the rectangular aperture of the Cell Framing Adapter. Restricting the area of illumination decreases the background signal and the auto fluorescence of the fluorescence objectives themselves.

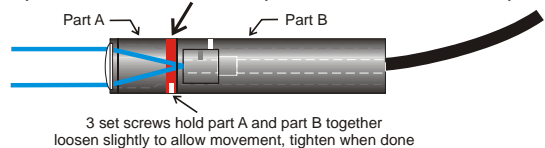
Light guide held into carrier by set screw



Carrier held into adapter by set screw



Circular aperture is connected to part A and Part B of adapter



Rotating Part A relative to Part B opens and closes aperture



Adjustment of excitation area

The excitation aperture is shipped at an “average” setting and is usually checked and, if necessary, adjusted at the time of the system installation. As discussed above the iris is adjust by loosening the screws and rotating part A relative to Part B. This mechanism was not designed to be easily adjusted rather it is meant to be set infrequently and fixed in the ‘correct’ position.

Judging the area of illumination is most easily done with a low magnification objective (like a 10x) and a piece of white paper. The process is as follows:

1. Remove the light guide collimator assembly from the microscope adapter
2. Loosen the set screws (one on each side) holding Part A and Part B together and close the aperture to its minimum position (this is done by holding the assembly with the lens pointed towards you and then rotating the collar clockwise)
3. Lightly tighten the screws holding part A and part B together
4. Install the light guide collimator back in the microscope.
5. Use the acquisition software to project one of the excitation wavelengths from the Excitation light source
6. Using the 10x objective it should be possible visualize a small, circular area of illumination roughly centered in the microscope’s field of view.
7. Now, repeat the process after opening the aperture a small amount.
8. Repeat this process until the edge of the illumination area is just within the microscope’s field of view.

This is the largest area that ever needs to be illuminated and is a reasonable starting point. Depending on the particular type of recording done by the system, it may be possible to further restrict the area of illumination

7. Specifications

1. IonOptix μ Step System
 - A. Controller box input power: 110-220VAC, 50-60Hz, 1.5A fuse
 - B. Active element: micro stepping motor/controller
 - C. Control signal: via 25 pin DSub connector
 - D. 6-position filter wheel holds 25mm diameter filters
 - E. Switching rate: __msec between adjacent filters
 - F. Excitation filters: for Fura-2, narrowband filters from Chroma Technology
 - G. Excitation output: directed w/o intervening optics to liquid light guide (1m in length w/ 5mm diameter active core)
2. Epifluorescence adapter: common element produces infinity projected beam via adjustable, circular field aperture. Microscope specific adapter provided
3. Cairn 75W Continuous xenon housing and power supply