

Fluorescence Photometry System

Complete, turnkey system for high-speed fluorescence recording

- 250 ratiometric or 1000 pseudo-ratiometric calcium measurements/second (configuration dependent)
- 1000 single and dual emission measurements/ second

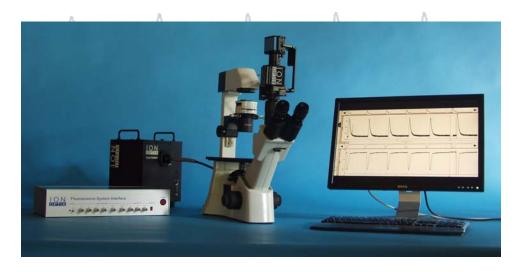




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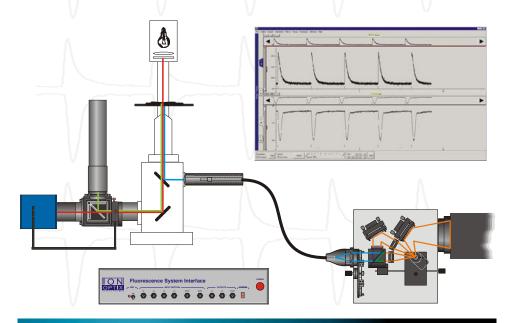


Fluorescence Photometry System Overview

Although the relationship between intracellular second messengers and downstream phenomena has been an area of intense investigation for decades, elucidation of the fundamental molecular and biophysical mechanisms remains at the forefront of cellular research. Impairment to the regulatory machinery governing cellular behavior correlates strongly with the onset and progression of many pathologies, including cardiomyopathies. Intracellular recordings of calcium and other important second messengers from isolated cells offer an important physiologic measurement while also providing key insights into the processes that affect tissue health.

IonOptix developed its Fluorescence Photometry Systems (<u>HyperSwitch configuration</u>: HPSYS and µStep configuration: MPSYS) over many years of collaboration with top researchers. We take pride in a line of precision products that are application driven and built to meet the needs of a demanding research environment. Since its inception in 1990 IonOptix has built and installed hundreds of high performance, turnkey systems in research laboratories worldwide.

Our complete photometry systems provide everything necessary for simultaneously acquiring quantitative fluorescence data with our new IonWizard 6 software. We also offer an inverted fluorescence microscope equipped with UV-transmitting photometry objective and brilliant optics, a digital framing camera and a suite of analog and digital connections for synchronous data collection.





IonWizard Software Suite

Acquisition of Quantitative Fluorescence Data

Our complete systems are built from components designed to work seamlessly with one another and our lonWizard core software, providing completely synchronous and accurate data acquisition. IonWizard's core functions are expanded through the PMTAcq acquisition module to record quantitative single-excitation, dual-excitation and dual-emission fluorescence data. Through this acquisition module, IonWizard communicates directly with hardware, including our fluorescence system interface and light source.

IW6 is capable of acquiring up to 4 channels of 1000 Hz analog data. It also supports analog data outputs and digital inputs. IW6 now features a signal generator function for programming voltages to drive and control external hardware.

PMTAcq supports PMT-based photometry recordings as well as coordinated wavelength control for dual excitation calcium acquisition.



- Coupled with either a Fluorescence System Interface, IonWizard and PMTAcq form a powerful integrated photometry and data recording system.
- Photometry data (such as cellular calcium levels) are simultaneously sampled at 1000 Hz for single excitation dyes, 250 Hz and 1000Hz for dual excitation dyes sampled ratiometrically and pseudo-ratiometrically, respectively.
- Multiple 'epochs' define separate acquisition regimes. Within each epoch, independent sampling rates for photometry and analog data are specified.

- Switches between different epochs occur automatically or via user intervention.
- Fluorescence data can be optionally output as an analog voltage in real time.



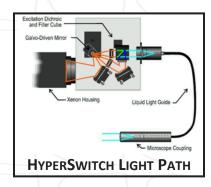
HyperSwitch

High Speed Dual Excitation Light Source

Cellular calcium levels rise and fall on millisecond time scales. For dual excitation indicator dyes such as Fura-2, the fluorescence excitation light source must switch between excitation wavelengths with speed and precision. The lonOptix HyperSwitch uses a galvanometer-driven mirror to steer light between two light paths. With sub-millisecond switching times, the HyperSwitch offers 250 true ratios per second when driven by our lonWizard core software and the PMTACQ acquisition module. The HyperSwitch comes equipped with a Xenon arc lamp for nearly uniform light intensity in the near ultraviolet and visible spectrum. The HyperSwitch delivers fluorescence illumination to your microscope through an efficient liquid light guide and a microscope-specific adapter.



- Fast switching. Galvanometer-based switching enables sub-millisecond switching. Complete dual excitation ratiometric collection achieved in 4 msec.
- 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.
- Excitation Optics. Excitation filters of your specification are included with each system. May be externally "flipped" out of light path for conventional fluorescence excitation. A manually controlled, 6-position filter wheel in the common excitation path provides a convenient means to select between neutral density filters (a 5-piece ND set is provided with HSW400).



- 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.
- 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 microscope.



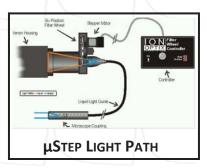
µStep

Single or Dual Excitation Light Source

The IonOptix μ Step also exploits our new filter wheel design that permits not only increased sampling speed (up to 8 ratio pairs per second), but also eliminates the need for a separate electronic shutter (movement to a shuttering position accomplished in 50 ms). It's a flexible device that can be used as part of a complete system or as a stand-alone device with six separate filter positions. The μ Step permits a modified sampling technique enabling pseudo-ratio sampling rates of 1000 Hz over short periods of time (visit us on the web for an application note on the interpolated numerator method). The μ Step delivers fluorescence illumination to your microscope through an efficient liquid light guide and a microscope-specific adapter.



- Controller with position display. A microcontroller provides the "intelligence" to drive the filter wheel and display current position. Filter position can be controlled either manually or by computer via a parallel or serial connection.
- Shuttering. The µStep is designed to use the filter wheel itself as a shutter between sampling periods to minimize light exposure, thereby avoiding dye bleaching and photodamage to the preparation. Separate electronic shuttering system is not required.
- 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.
- Excitation Optics. Excitation filters of your specification are included with each system. Sliders provide allow rapid placement of neutral density filters in the common excitation path, providing a convenient means to select between ND filters (a 5-piece ND set is provided with MUS200).



- Emission Filter / Dichroic. The appropriate emission filter and dichroic mirror is provided for your microscope.
- 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 microscope.
- Cables. Includes cables to connect to the Fluorescence System Interface.



Fluorescence System Interface

System Integration & Control

The IonOptix Fluorescence System Interface, model FSI700, provides all the standard non-video input, output and device control hardware needed for a typical dual-excitation fluorescence system. The FSI700 may be combined with a variety of IonOptix components to create the specific combination of system capabilities that are required in your experiments.

The FSI700 may be connected to external devices using the four analogi inputs, the two analog outputs or the start and mark trigger inputs. The FSI's inputs are used to collect output data from analog devices in real time. IonWizard's flexible device configuration allows the experimenter to specify the name and unit scaling of each auxiliary signal for easy-to-read data files.



Features

Inputs

- **Dual PMT inputs.** Provides connection to two PMT sensors.
- Start in. This TTL signal allows external initiation of data sampling.
- Mark in. This TTL signal is recorded during data acquisition to provide event synchronization information.
- Analog to Digital. Four channels of 16-bit A/D with input voltage range of ±5V.

Outputs

Digital to Analog. Two channels of ±5V.
 12-bit D/A outputs can be used as monitor or control signals.

Light Source Control

 IonOptix 25pin. Provides control signals to/from excitation light source.

Includes

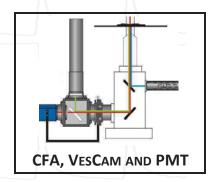
- Includes half-length, full-height PCI computer interface card and six foot cable.
- IonWizard driver software for Windows 2000, XP.



Fluorescence & Video Detection

Quantitative Fluorescence Detection

For detecting and quantifying fluorescence emission, we furnish systems with one or more photomultiplier tubes (PMTs). The PMT offers broader dynamic range, faster acquisition rates and greater photosensitivity than CCDbased sensors. To enable alignment and masking of the cell or region of interest we equip our photometry system with our cell framing adapter (CFA) and VesCam digital CCD camera. The CFA hosts several optical elements for filtering and directing light to the appropriate device. It holds an aperture for physically framing the image, preventing unwanted extracellular background from contributing to the fluorescent signal. The CFA also comes outfitted with our MyoHandle, a device for mechanically rotating the camera image for precise alignment of the interrogation window. Coupled with the appropriate optical filters, our CFA, camera and PMT offer precise photometry measurements ideally suited for intracellular indicator dyes.



PMT Sub-System

The new PMT400 sub-system offers true photon counting using an end-on bialkali PMT tube with integrated high-voltage power supply and amplifier discriminator. This combined package increases reliability and decreases cost. The PMT400 is directly powered and controlled by the IonOptix model FSI700 Fluorescence System Interface, where it is under software control. For stand-alone applications, a separate module (PTV100) is available that provides power to the PMT400 and a BNC analog "count" output for connection to other devices.

- Integrated Tube, Supply and Amplifier/Discriminator. Single package holds photon-quality blalkali PMT tube (180-600nm, 400nm peak sensitivity), integral, high-voltage power supply preset to the photon counting voltage, and amplifier/discriminator.
- C-Mount. Industry standard C-mount available to provide easy connection to standard microscope adapters.



- IonOptix Coupling and Emission Filter. An IonOptix to C-mount coupling holds the included emission filter and directly connects to the IonOptix Cell Framing Adapter.
- Cable. Integrated cable connects directly to Fluorescence System Interface. Standard extension cables allow the cable length to be easily increased.



Fluorescence & Video Detection (cont.)

Cell Framing Adapter

The Cell Framing Adapter is used to simplify and optimize fluorescence recording using a PMT. An adjustable iris is used to frame a rectangular area of the microscope field of view maximizing selected cell or region fluorescence while masking background signal. The CCD camera displays the image area that the PMT will record. The CFA's D-Cube holds a dichroic mirror and emitter to reflect and filter fluorescence emission before collection by the PMT. The use of a red filter in the microscope condenser permits the transmitted image to be visualized concurrently with photometry recording. The Cell Framing Adapter comes with the matching mount to connect to the PMT Sub-System.



Features

- Microscope Coupling. A microscopespecific coupling attaches to the side port or trinocular head of all common microscopes.
- D-Cube. The D-Cube provides convenient access to emission optics. An appropriate dichroic mirror and emission filter for your selected emission band is included.
- Rectangular Aperture. Masks signal from cell and debris adjacent to the cell of interest.
- Magnification coupling. A range of demagnification couplers are available to optimize the image size presented to the system camera.
- MyoHandle. This mechanical element couples the rectangular aperture to the camera so that both elements can be rotated in tandem, facilitating cellular alignment.

Options

 Option D: Dual Emission. The CFA optics can be stacked to permit dual emission PMT recording.



Vessel Fluorescence Microscope

Motic AE31 & Olympus UIS2 Objective

The IonOptix-configured Motic AE31 inverted microscope provides an ideal platform for combined photometry and dimensioning measurements. It features upscale research functions, such as halogen Koehler illumination and epifluorescence capacity. The AE31 also incorporates Motic's Color Corrected Infinity Optical System [CCIS®] to produce crisp, flat and high contrast images. We equip our microscope packages according to the specific demands of the proposed IonOptix system. For calcium photometry systems, the AE31 comes configured with an Olympus fluorescence objective with high numerical aperture and UV transmission. Properly equipped with an IonOptix fluorescence illumination system, the Motic serves as an exceptional choice for researchers in search of reliable, high fidelity data acquisition at an affordable price.



MOTIC AE31 MICROSCOPE

Features

Motic AE31 Inverted Microscope

- CCIS Optics. Color corrected infinity optical system.
- Brightfield Illumination. Koehler illumination system w/ true DC 6V-30W output delivers bright, consistent illumination at all optical magnifications.
- Mechanical Stage. Precise control of sample position. Comfortable long wand allows user to rest forearm while manipulating stage.
- Trinocular Head. Easy access to auxiliary components. Efficient transmission of light for photometry and cellular dimensioning.
- Microscope Base. Wide base for strength and rigidity. Inverted "Y" support provides additional lateral support. Ergonomic design provides easy adjustment of focus and stage controls.
- Epifluorescence Package. 3 cassette capacity. Easy access to dichroic filter positioning. Direct and efficient fluorescence illumination port.

Olympus UApo/340 40X & 20X and UPLSAPO 10X objectives

- UV Transmission. High transmission of 340nm light; ideal for Fura-2 based Ca²⁺ photometry.
- High NA. Non-immersion lenses with excellent transmission and collection of light.



IonOptix Systems

Our goal is to support the scientific research community with an array of systems that meet experimental demands while adhering to our philosophy of developing innovative high-performance products at fair prices. The following are some of our application-driven systems currently available. Visit us at www.lonOptix.com for more information.

Calcium and Contractility/ Diameter

- Myocyte Calcium and Contractility Recording Systems
 - HyperSwitch Configuration (HMSYS)
 - µStep Configuration (MMSYS)
- Vessel Calcium and Diameter Recording System (HVSYS)
- Myocyte Contractility System (MCSYS)
- Vessel Diameter System (VDSYS)

Tissue Bath Fluorometry

FluoroPlex

General Photometry

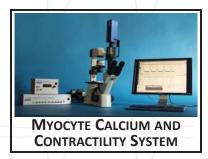
• Fluorescence Photometry System (HPSYS)

Cell Pacing

• Cell Culture Pacing

Myocyte Harvesting

Myocyte Harvesting System











"The Barn" — IonOptix Headquarters in Milton, Massachusetts

Company History

IonOptix makes quality ratiometric fluorescence and cell dimensioning data acquisition systems. We have been making reasonably priced, high performance systems since 1990.

IonOptix prides itself on post-sale customer support. Telephone and email support is available on an unlimited basis. More importantly, every system sale includes a one to two day installation visit to set up the system and train the customer. We consider this training to be critical as it gets the customer up and running as quickly as possible. We run experiments with your preparations during the visit to be assured that all technical issues particular to the experimenter's preparations have been covered.

Resellers

Our good friends at Cairn Research are currently our only resellers. They sell our components in conjunction with their fluorescence and electrophysiology systems.



Cairn Research

www.cairn-research.co.uk

Authorized Representatives

In efforts to afford this high level of service to all our customers, IonOptix has entered into agreements with local representatives in several countries. The representatives have been chosen based on their technical and biological expertise, familiarity with our products, and on their customer service skills.



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