doric

Integrated Connectorized Fluorescence Mini Cubes

User Manual

Version 1.0.2

Contents

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Overview

1.1 Integrated Connectorized Fluorescence Mini Cube

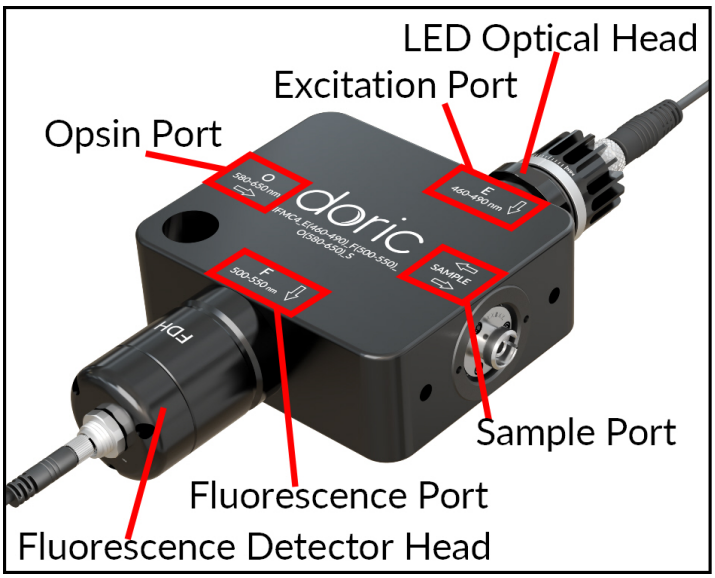


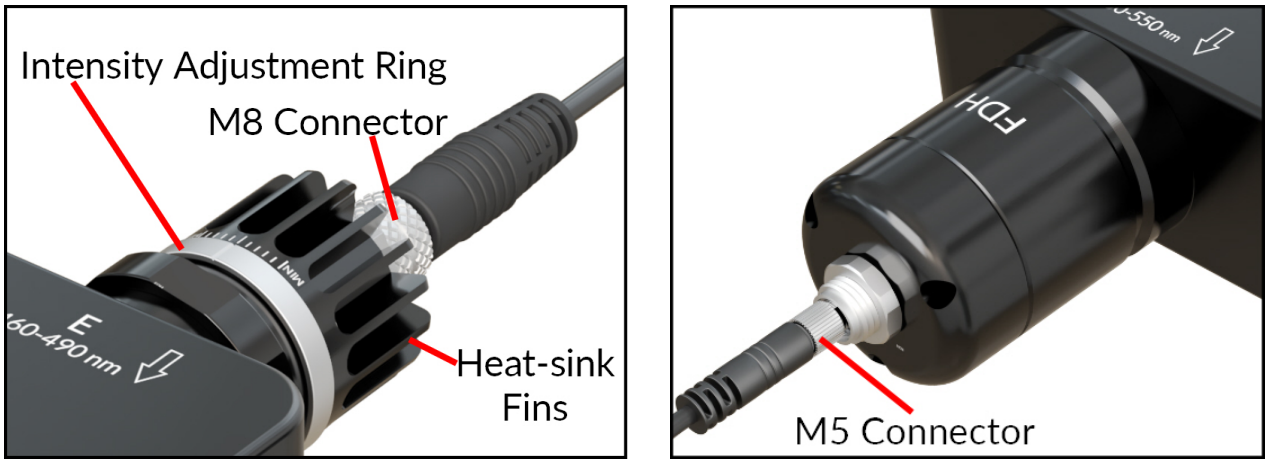
Figure 1.1: *4-port Fluorescence Mini Cube*

The *integrated Fluorescence Mini Cube* is an optical assembly that allows the combination of multiple excitation and detection signals. The cubes are classified by their number of ports, with 3, 4, 5, 6 and 7 port cubes available. The ports are each classified according to their usage, and are also qualified by a wavelength band corresponding to the bandwidth of optical filters within the cube.

- **E** and **IE** (Fig. 1.1) represent entry ports for fluorescence and isosbestic point excitation light. Each port of this type comes with a *Built-in LED Optical Head* that contains the following elements.
  - The **Intensity Adjustment Ring** (Fig. 1.2a) allows additional adjustment of the overall output intensity of light.
  - The **M8 connector port** (Fig. 1.2a) is used to connect the optical head to the *LED Driver* using a *Male-Female M8 Cable*.
  - The **Heat-sink Fins** (Fig. 1.2a) are used to evacuate heat from the light source, allowing stable output power. Ensure the fins are not blocked to allow proper cooling.
- **F** (Fig. 1.1) represents ports for fluorescence emission light. Each port of this type comes with a *Built-in Doric Fluorescence Detector Head*.

- The **M5 Connector** (Fig. 1.2b) allows the *Fluorescence Detector Head* to be connected to the *Fluorescence Detector Amplifier* using an M5 male/male connection cable.
- For extremely low light level applications, the fluorescence ports (F, F1, etc.) can have a *Built-in Photomultiplier Tube* rather than a *Built-in Fluorescence Detector Head*.

- **O** (Fig. 1.1) represents optogenetic activation or silencing ports. These are always FC receptacles to allow connection to laser or fluorescence light sources.
- **S** (Fig. 1.1) represents the exit port to the sample. These are always FC receptacles to allow connection to an experimental subject.



(a) *Built-in LED Optical Head* (b) *Built-in Fluorescence Detector Head*

Figure 1.2: *ilFMC Built-in components*

1.2 Fluorescence Detector Head

The Fluorescence Detector Amplifier amplifies the signal coming from the detector head and transmits it to a recording system using a BNC output. It contains the following elements.

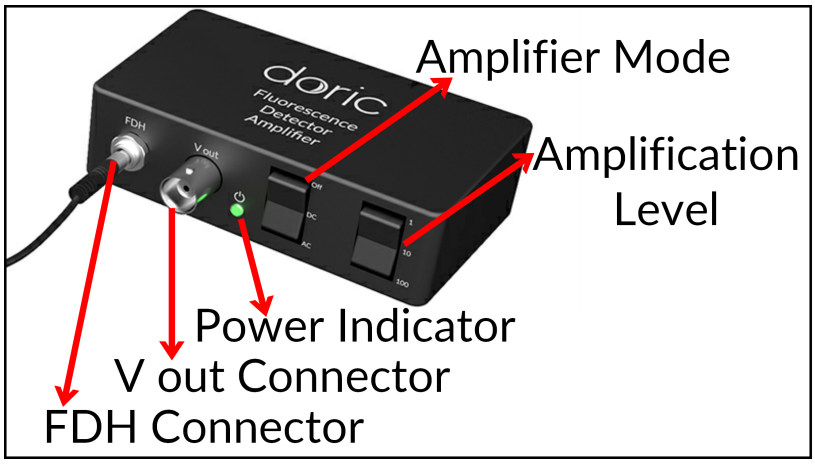


Figure 1.3: *Doric Fluorescence Detector Amplifier Elements*

- The **FDH Connector** (Fig. 1.3) is an M5 type connector used to link the amplifier and the head using a shielded M5 cable.

- The **V out Connector** (Fig. 1.3) is a BNC type connector used to connect the fluorescence detector with a DAQ system.
- The **Power Indicator** light (Fig. 1.3) shines green when the detector is on.
- The **Amplifier Mode** switch (Fig. 1.3) is used to switch the detection mode from **Off** to **AC** or **DC**.
- The **Amplification Level** switch (Fig. 1.3) allows the choice of amplification levels at 1, 10 or 100 times.
- The **Power Supply** connector, located on the back of the unit, is used to connect the 12 V power supply to the amplifier.

1.3 Battery Pack

(a) *Standard Battery Pack*

(b) *Rechargeable Battery Pack*

Figure 1.4: *Doric Fluorescence Detector Battery Packs*

There are two available battery packs.

- A *9V Battery Pack* (Fig. 1.4a) using standard 9V/E type battery, as well as a 9V/E batteries are shipped with each detector. These allow optimal usage of the detector when wall plugs are unavailable or when the current from a wall plug introduces significant noise in the signal. The pack can be connected directly to the **Power Supply** port and a single battery will last an average of 20 hours.
- A *Rechargeable Battery Pack* (Fig. 1.4b) can be provided when requested. These 12V 6000 mAh battery packs can keep a single detector powered for approximately 1 week without interruption. These power supplies are ideal for those who want to use the detectors solely using battery power.

1.4 Electrical Cables

The *Integrated Fluorescence Mini Cube* is provided with two electrical cable types. The *Fluorescence Detector Cable* is a shielded twisted pair M5 connectorized electrical cable used to connect the *Built-in Fluorescence Detector Head* and the *Fluorescence Detector Amplifier*. The *Optical Head Cable* is a male-female M8 connectorized cable using to connect the *Built-in Optical Head* to a *LED Driver*.

1.5 Integrated Fluorescence Mini Cube With 3 Ports

Figure 1.5: *3-port Integrated Fluorescence Mini Cube*

The *3-port Integrated Fluorescence Mini Cube Configuration* (Fig. 1.5) has a single excitation port **E** and a single fluorescence port **F**. Currently available models include configurations for **GFP-like** (Table 1.1) and **RFP-like** (Table 1.2) fluorophores.

Table 1.1: *GFP-like Configurations*

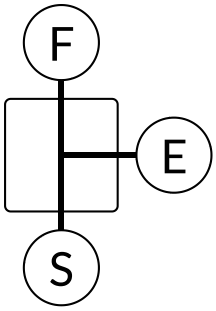
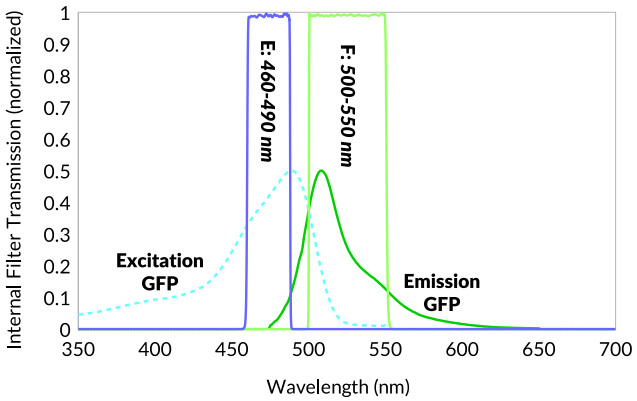
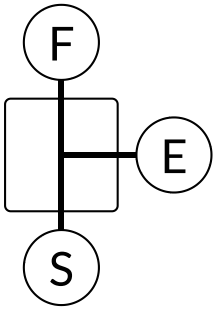
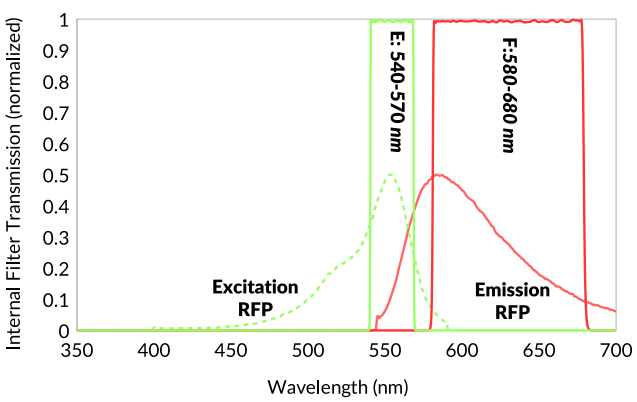
Configuration schematic	Filter Transmission
	

Table 1.2: *RFP-like Configurations*

Configuration schematic	Filter Transmission
	

1.6 Integrated Fluorescence Mini Cube With 4 Ports

Figure 1.6: *4-port Integrated Fluorescence Mini Cube*

The *4-port Integrated Fluorescence Mini Cube* (Fig. 1.6) has several different popular configurations.

- The first involves an excitation **E**, an optogenetic activation/silencing **O**, a fluorescence detection **F** and sample **S** ports. Such cubes are used for **GCaMP** fluorescence measurement combined with the activation of **red opsins** (Table 1.3).
- The second involves a fluorescence excitation **E**, isosbestic point excitation **IE**, a fluorescence detection **F** and sample **S** ports. This configuration can be used for **locked-in or sequential detection of autofluorescence** and **fluorophore excitation** (Table 1.4).
- The third involves a fluorescence excitation **E**, two fluorescence detection **F1/F2** and sample **S** ports. This configuration can be used for **CFP-YFP FRET experiments** (Table 1.5).

Table 1.3: *Excitation, Fluorescence and Opsin Activation*

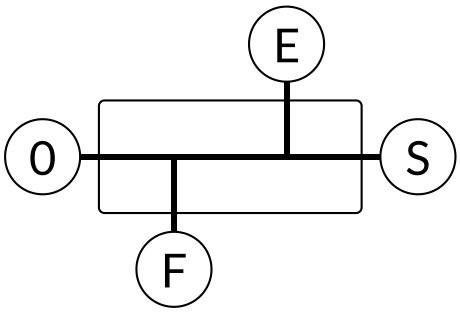
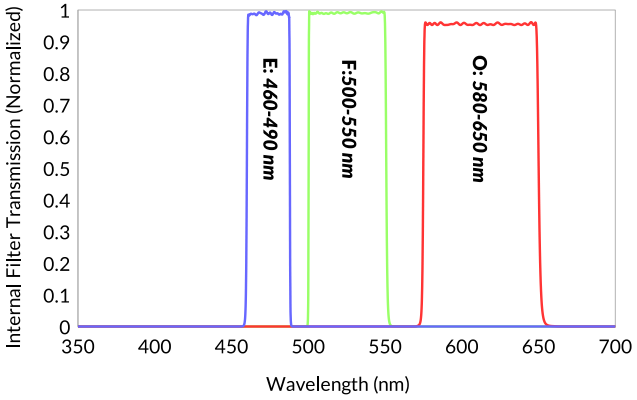
Configuration schematic	Filter Transmission
	

Table 1.4: *Locked-in or Sequential Detection of GCaMP Isosbestic and Functional Excitations*

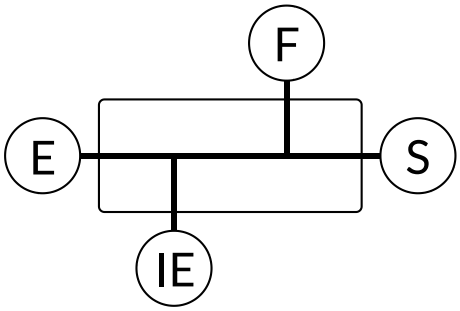
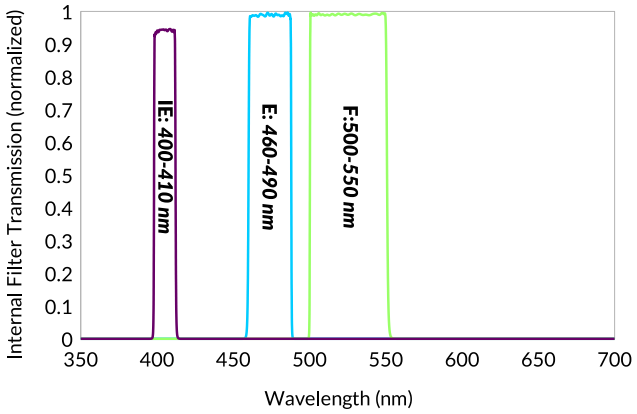
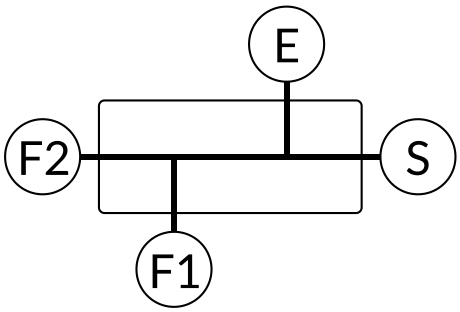
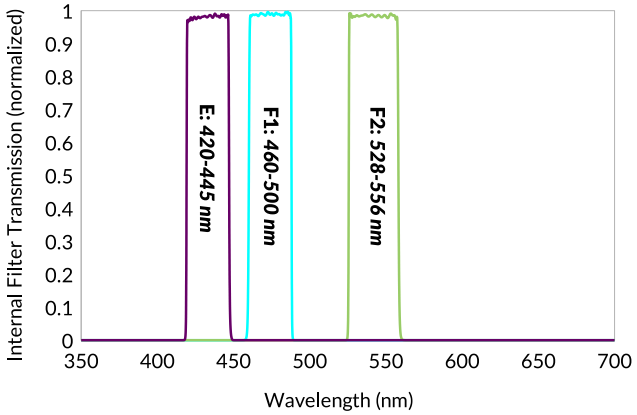
Configuration schematic	Filter Transmission
	

Table 1.5: *CFP-YFP FRET Cube (One Excitation and Two Fluorescence Detection Ports)*

Configuration schematic	Filter Transmission
	

1.7 Integrated Fluorescence Mini Cube With 5 Ports

Figure 1.7: *5-port Integrated Fluorescence Mini Cube*

The *5-port Integrated Fluorescence Mini Cube* (Fig. 1.7) has two different popular configurations.

- The first involves a fluorescence excitation **E1**, an isosbestic point excitation **IE**, two fluorescence detection **F1,F2** and sample **S** ports. This configuration can be used to separate the **isosbestic point** and **fluorescence** with minimal cross-talk (Table 1.6).
- The second involves two excitation **E1/E2**, two fluorescence detection **F1/F2** and a sample **S** ports. This configuration can be used for **green fluorophore** and **red fluorophore** excitation and detection (Table 1.7).

Table 1.6: *GCaMP Isosbestic and Functional Excitations, and Opsin Activation*

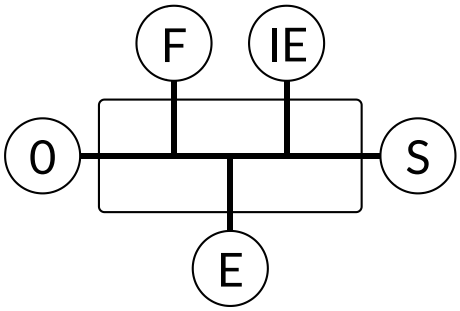
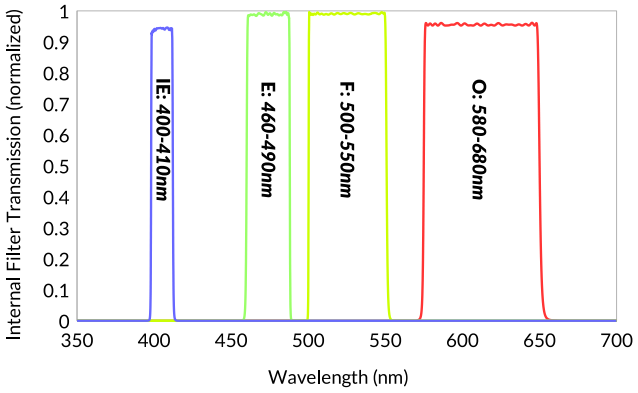
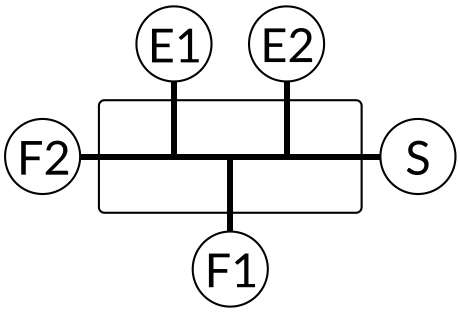
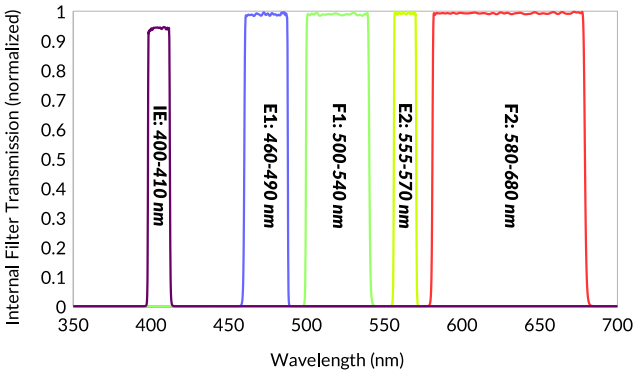
Configuration schematic	Filter Transmission
	

Table 1.7: *Separated Two Fluorophores Fluorescence Cube*

Configuration schematic	Filter Transmission
	

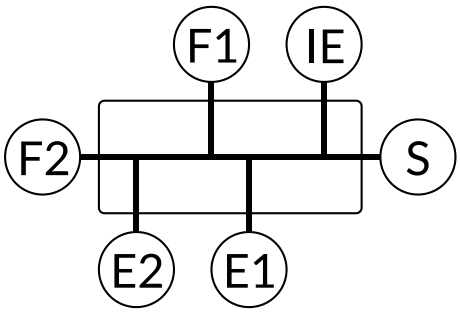
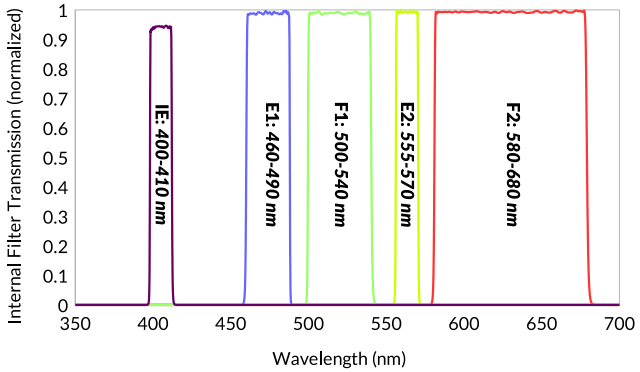
1.8 Integrated Fluorescence Mini Cube With 6 Ports

Figure 1.8: *6-port Integrated Fluorescence Mini Cube*

The *6-port Fluorescence Mini Cube* (Fig. 1.8) has one popular configuration.

- There are two excitation **E1/E2**, an isosbestic excitation **AE**, two fluorescence detection **F1/F2** and a sample **S** ports. This configuration can be used to detect the fluorescence of **two different fluorophores** in comparison with sample **isosbestic excitation** (Table 1.8).

Table 1.8: *Two Fluorophores Fluorescence and GCaMP Isosbestic Excitation*

Configuration schematic	Filter Transmission
	

1.9 Integrated Fluorescence Mini Cube With 7 Ports

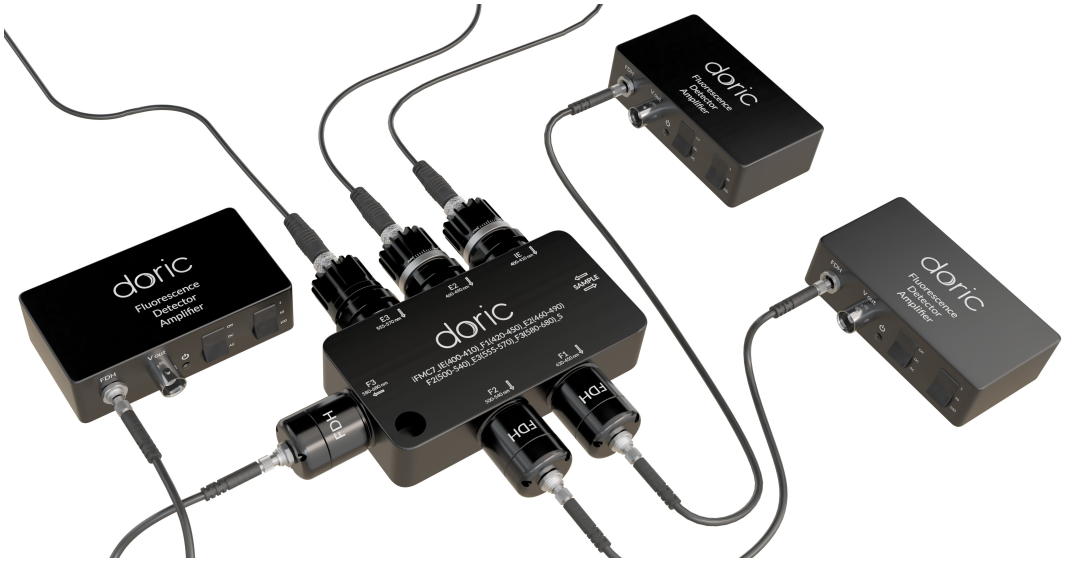
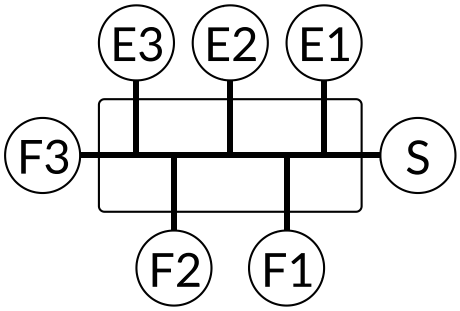
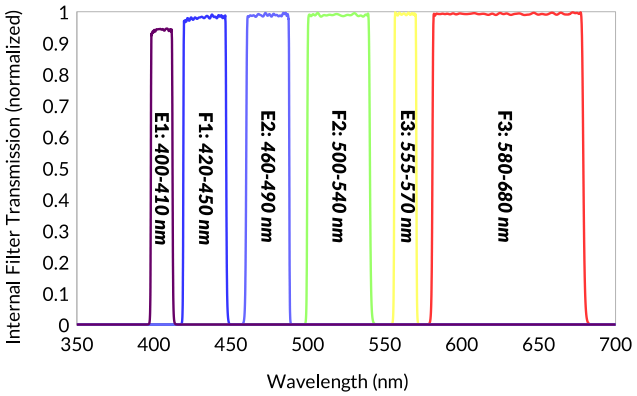


Figure 1.9: *7-port Integrated Fluorescence Mini Cube*

The *7-port Integrated Fluorescence Mini Cube* (Fig. 1.9) has a single popular configuration. The configuration involves three excitation **E1/E2/E3**, three fluorescence detection **F1/F2/F3** and one sample **S** port. This configuration can be used to detect the fluorescence of **three different fluorophores** simultaneously (Table 1.9).

Table 1.9: *Three Fluorophores Fluorescence*

Configuration schematic	Filter Transmission
	

2

Operations Guide

2.1 Getting Started

The usage of the *Fluorescence Mini Cube* is straightforward.

1. Each type of *Fluorescence Mini Cube* has a screw hole to secure the cube. The hole accepts 1/4 (or M6) screws.
2. Clean the connector end tips of the patch cords before connecting them to *Fluorescence Mini Cube*. Use isopropyl alcohol or a similar cleansing solution.
3. With an FC connector, the connector key must be oriented to enter within receptacle slot to ensure good connection (Fig. 2.1).

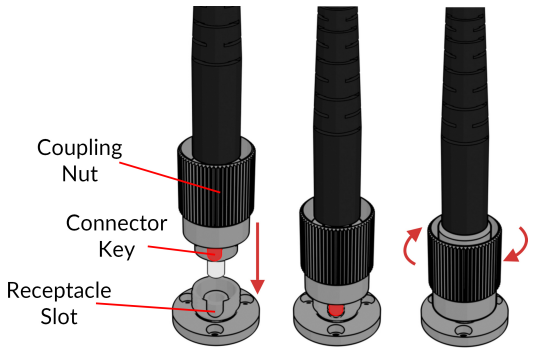


Figure 2.1: *FC connector, Fiber Installation*

- ⚠ To reduce the risk of eye injury, **it is sound practice to AVOID LOOKING DIRECTLY AT THE CONNECTOR OPENINGS** when the light source is turned on.

4. When not in use, place the plastic cap on the connectors for protection and cleanliness.

For its usage inside a system (Fig. 2.2), see Fiber Photometry Application Note. For use with a photomultiplier tube (Fig. 2.3), see the Photomultiplier Tube User Manual.

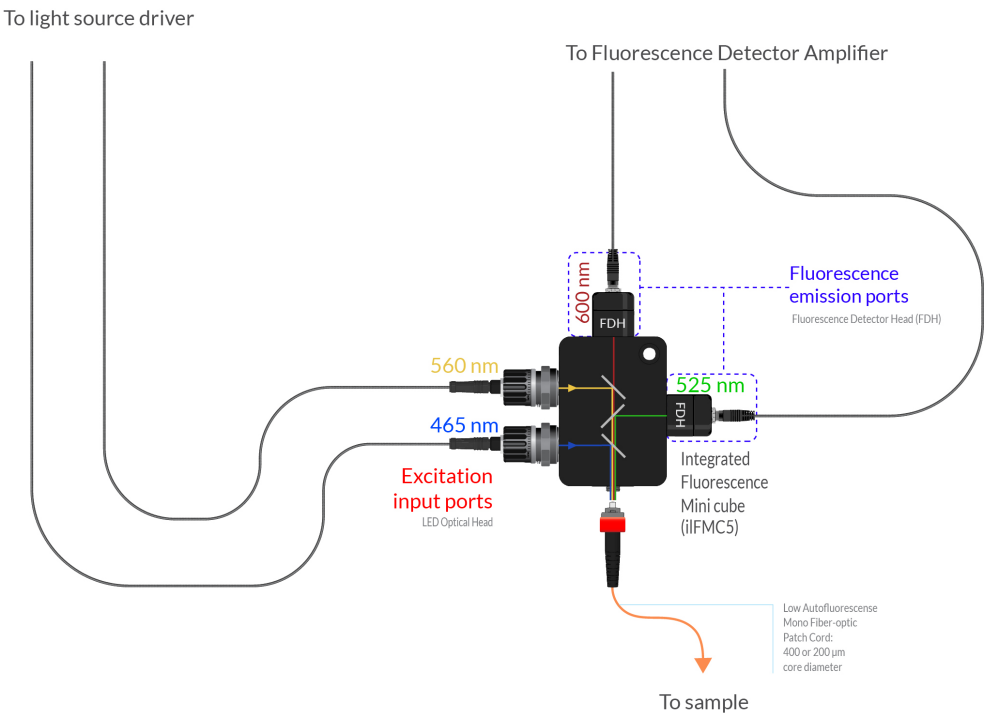


Figure 2.2: *Connections to the Integrated Fluorescence Mini Cube*

Figure 2.3: *Hamamatsu H10722-20 Photosensor modules directly attached to the Fluorescence Mini Cube*

3

Specifications

3.1 Specifications

Table 3.1: *Typical Connectorized LED Output Power vs Optical Fiber Core Diameter*

LED		TYPICAL OUTPUT POWER @1000 mA (mW)[1]			Overdrive @2000 mA (pulsed)	Intensity Adjustment
Central Wavelength (nm)	Bandwidth FWHM (nm)	Core 200 µm 0.53 NA	Core 400 µm 0.53 NA	Core 960 µm 0.63 NA		
365	~12	6.0	23	100	1	(TDB)
385	~12	6.0	23	100	1	(TDB)
405	~15	5.0	23	100	1	(TDB)
420	~15	5.5	23	100	1	(TDB)
450	~25	8.0	23	100	x1.7	(TDB)
465	~25	7.5	23	100	x1.7	(TDB)
505	~30	3.0	12	50	x1.6	(TDB)
515	~40	3.0	9.5	40	x1.5	(TDB)
560	~100	2.0	8.5	40	-	(TDB)
595	~20	2.0	8.5	40	x1.2	(TDB)
625	~20	3.5	14	70	x1.6	(TDB)
635	~20	6.5	25	100	x1.6	(TDB)
850	~35	6.0	22	40	-	(TDB)
940	~35	2.0	10	40	-	(TDB)
5500K	-	4.5	17	80	-	(TDB)

[1] All power values taken at a maximum current of 1000 mA, except for 365, 385, 405 and 420 nm LEDs (500 mA).

Table 3.2: *General specifications*

SPECIFICATION	VALUE	NOTES
Dimensions		
ilFMC 3	97.6 x 80.5 x 29.0 mm^3	Includes Connectors, LED and Detectors
ilFMC 4	124.1 x 110.5 x 29.0 mm^3	Includes Connectors, LED and Detectors
ilFMC 5	112.3 x 135.9 x 29.0 mm^3	Includes Connectors, LED and Detectors
ilFMC 6	138.1 x 130.9 x 29.0 mm^3	Includes Connectors, LED and Detectors
ilFMC 7	124.6 x 144.2 x 29.0 mm^3	Includes Connectors, LED and Detectors
*Typical fiber configuration for test		
Excitation path		
-From FMC to Sample	400 μm core, NA 0.57	-
Emission path		
-From Sample to FMC	400 μm core, NA 0.57	-

Table 3.3: *Typical filter transmission bandwidth*

Fluorescence Mini Cubes	Excitation (nm)	Emission (nm)
ilFMC3		
-GFP-like	460-490	500-550
-RFP-like	540-570	580-680
ilFMC4		
-Excitation, Fluorescence and Opsin Activation	460-490, 580-650	500-550
-Locked-in or Sequential Detection and Fluorophore Excitation	400-410, 460-490	500-550
-FRET (One Excitation and Two Fluorescence Detection Ports)	420-445	460-500, 528-556
ilFMC5		
-Separated Two-fluorophore Fluorescence	460-490, 555-570	500-540, 580-680
-GCAMP Isosbestic and Functional Excitations, and Opsin activation	400-410, 460-490, 580-650	500-550
ilFMC6		
-Two-fluorophore and GCaMP Isosbestic Excitation	400-410, 460-490, 555-570	500-540, 580-680
ilFMC7		
-Three-fluorophore Fluorescence	400-410, 460-490, 555-570	420-450, 500-540, 580-680

4

Support

4.1 Maintenance

The product does not require any maintenance. Do not open the enclosure. Contact Doric Lenses for return instructions if the unit does not work properly and needs to be repaired.

4.2 Warranty

This product is under warranty for a period of 12 months. Contact Doric Lenses for return instructions. This warranty will not be applicable if the unit is damaged or needs to be repaired as a result of improper use or operation outside the conditions stated in this manual. For more information, see our Website.

4.3 Contact us

For any questions or comments, do not hesitate to contact us by:

Phone 1-418-877-5600

Email sales@doriclenses.com

357 rue Franquet - Quebec, (Quebec)
G1P 4N7, Canada
Phone: 1-418-877-5600 - Fax: 1-418-877-1008
www.doriclenses.com