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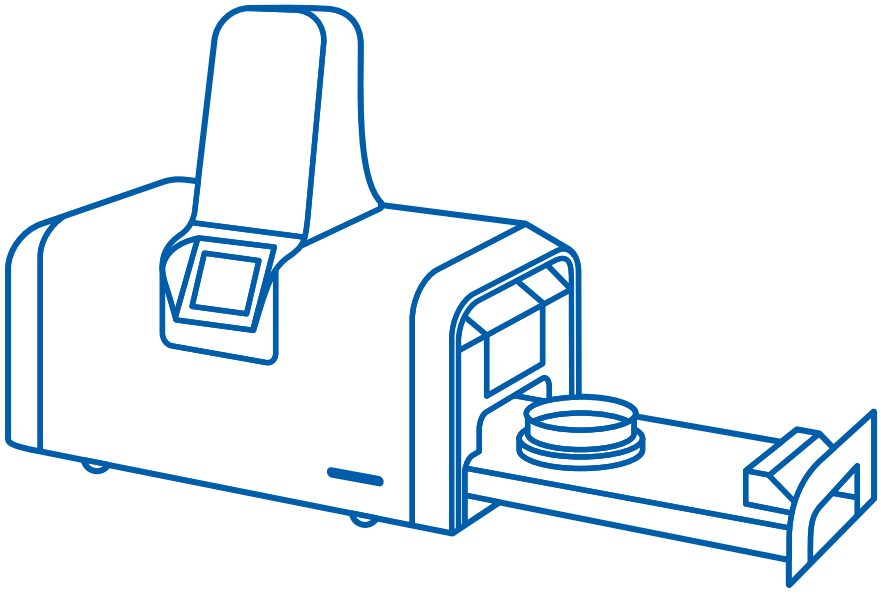
EZ-Fluo™ Rapid Detection System User Guide



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NOTE

This user guide is also available in the following languages: French, Italian, German, Spanish, Portuguese, Japanese and Chinese. Versions can be found on our website at www.millipore.com/techlibrary.

REMARQUE

Ce guide de l'utilisateur est également disponible dans les langues suivantes : français, italien, allemand, espagnol, portugais, japonais et chinois. Les versions peuvent être téléchargées depuis notre site Internet : www.millipore.com/techlibrary.

NOTA

Il presente manuale è disponibile anche nelle seguenti lingue: francese, italiano, tedesco, spagnolo, portoghese, giapponese e cinese. Tali versioni si trovano nel nostro sito web all'indirizzo www.millipore.com/techlibrary.

HINWEIS

Diese Bedienungsanleitung ist auch in den folgenden anderen Sprachen verfügbar: Französisch, Italienisch, Deutsch, Spanisch, Portugiesisch, Japanisch und Chinesisch. Diese Versionen finden Sie auf unserer Website: www.millipore.com/techlibrary.

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OBSER-VAÇÃO:

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Contents

Introduction

EZ-Fluo™ System is a rapid fluorescence-based technology for fast quantitative detection of contaminants over a broad range of filterable matrices.

This easy-to-use and simple system can detect viable microorganisms filtered on a membrane down to one CFU per sample. The non-destructive method allows the identification, by any method, of any microorganisms detected during the initial fluorescence reading. Sample preparation is performed using the EZ-Fit™ Manifold filtration system and membranes that ensure consistent and reliable results.

Chemistry of the Reaction

The EZ-Fluo™ system uses proven technology based on a universal enzymatic fluorescent staining of viable microorganisms.

After a filtration step, all microorganisms retained on the membrane are stained by a fluorescence viability marker. The reaction consists of an enzymatic cleavage by active microbial metabolism of a nonfluorescent substrate. Once cleaved by the cell's metabolism, the substrate liberates free fluorochrome into the microorganism cytoplasm that remains in the cell due to its hydrophilic nature.

The signal is thereby naturally amplified by accumulation of the fluorochrome inside the cells and can be easily detected by exposure to the excitation wavelength of the fluorescence dye in the EZ-Fluo™ Reader and visually counted or counted using the EZ-Fluo™ Camera and dedicated software.

Principles of the Procedure

The procedure is easy and requires limited training and skills: sample contaminants are collected by membrane filtration using the EZ-Fit™ Manifold filtration system.

After a short incubation time, media plates are removed from the incubator. Each membrane is then transferred onto a cellulose pad that has been pre-soaked with staining solution. These are then put in the incubator for 30 minutes.

Fluorescent micro-colonies can be counted two ways: directly through the viewing window of the EZ-Fluo™ reader, or using the EZ-Fluo™ Camera and dedicated software, which projects the image onto a computer screen. After reading, membranes can be re-incubated on media for later collection and identification of microorganisms if contamination of the sample was detected.

Operator and Equipment Safety

All employees who will operate and/or be near the EZ-Fluo™ must comply with the following:

• Read and understand the user guide of the EZ-Fluo™ before using this EZ-Fluo™. Failure to follow operating instructions could result in user injury or damage to the instrument.

• Read and understand all maintenance instructions in this user guide before performing maintenance on the pump. Failure to follow instructions could result in user injury or damage to the instrument.

• Any alteration of the pump from factory specification may cause unsafe conditions, and will void the product warranty.

• Any attempt to use the EZ-Fluo™ in a manner not specified in this user guide may result in damage to the instrument, operator injury, and will void the product warranty.

• Do not attempt to open and repair the EZ-Fluo™. Service should be performed by trained and authorized personnel only.

• Place the EZ-Fluo™ on a clean, flat, stable, horizontal surface, away from any source of excessive heat and close to an easily accessible, properly grounded power supply outlet. Do not locate the EZ-Fluo™ under direct lab light, as this could interfere with the reading of the membrane.

• The EZ-Fluo™ must be installed and used in a clean and dry area.

• The product is not intended to be used in explosive atmosphere

• Do not expose the Reader , Camera , power supply to liquid. If this happens, immediately switch off and disconnect the pump from the power outlet, wipe up the liquid

• Use only accessories and replacement parts designed for the pump. Using accessories not designed for the pump could result in user injury or damage to the instrument.

• When filtrating hazardous liquids, wear and use proper protective clothing and equipment in accordance with the MSDS for the handling and the disposal of the liquid to be filtrated.

• The system power cord is the main system disconnecting device, and must be easily accessible at all times.

• To avoid damaging the power cord or the outlet, it is recommended that you pull on the plug and not on the cord to unplug the system from a wall outlet or an electrical connector.

• Make sure the power cord is not a tripping hazard. Make sure that it is not gripped too tightly at the plug, the outlet or the point where it is connected to the system.

• Before use, check that the hoses are not unplugged, folded, or damaged.

• In case of skin contact with the filtrated liquid, refer to the safety datasheet of the filtrated liquid for first aid measures.

• Dispose the filtrated liquids according to local regulations. Before cleaning, shut down the EZ-Fluo™ and switch off and disconnect the power supply from power source.

• The power supply must be connected to the earthing system

• The electrical installation must comply with local standards, power supply voltage: 100 - 240 Volt AC, 50-60 Hz.

• Use an electrical surge protector to prevent damage to the system.

• EZ-Fluo™ is designed for indoor use only.

Use and storage temperatures

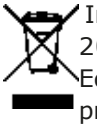
• The EZ-FluoTM Reader and/or its accessories must be kept away from any heat source. Do not expose the system to direct sunlight or to temperatures outside the ranges of values given below.

• Use and storage temperatures given below:

System operation	System storage	Altitude
10 to 40 °C	0 to 40 °C	< 2000 m

• Maximum relative humidity (storage and operation) 80% at temperatures up to 31 °C, then linear decrease to 50% RH at 40 °C.

• System dismantling – WEEE directive



In accordance with to the European Union Directive 2012/19/EC on Waste Electric and Electronic Equipment (WEEE), the presence of this logo on the product indicates that it should not be disposed of in the normal waste stream but collected separately.

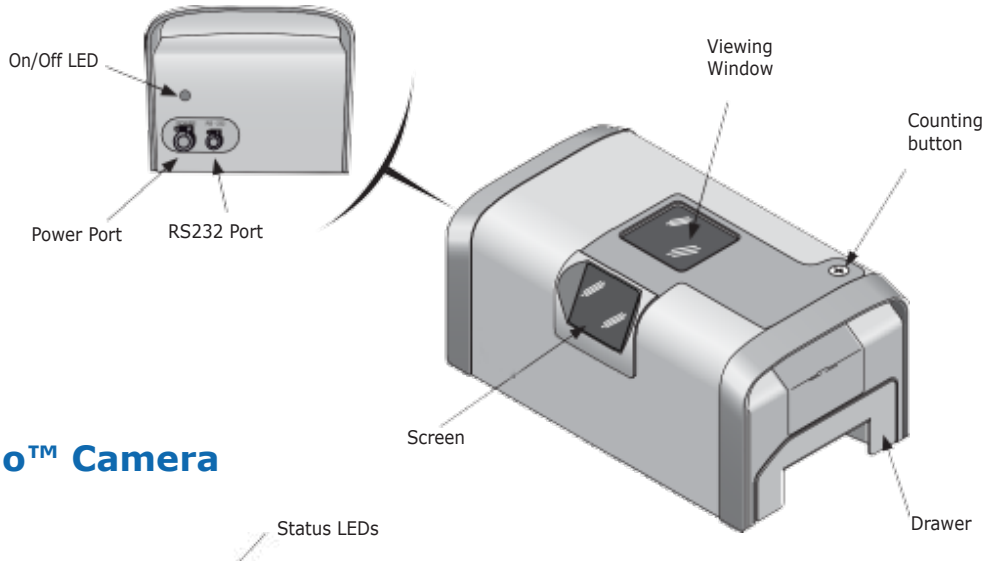
Go to www.millipore.com/weee for details on how to ensure proper treatment of the product in different countries.

Specifications and Operating Requirements

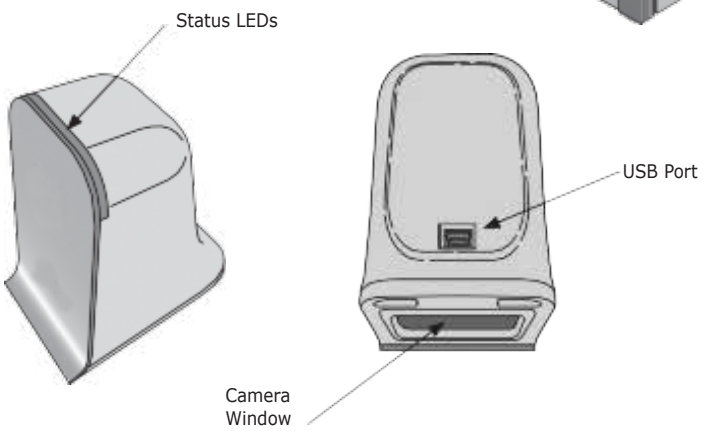
Dimensions (nominal)	**Component**	**Length**	**Width**	**Height**
	Reader	24.9 cm (9.8 in.)	14.2 cm (5.6 in.)	12.5 cm (4.9 in.)
	Camera	9.7 cm (3.8 in.)	6.6 cm (2.6 in.)	11.4 cm (4.5 in.)
Weight (nominal)	**Component**		**Weight**	
	Reader		4.4 kg (9.7 lbs.)	
	Camera		0.5 kg (1.1 lb.)	
Voltage	**Component**	**Supply Voltage Input**	**Supply Voltage Output**	
	Reader	100V – 240 V 50 Hz – 60 Hz	24V direct current	
	Camera	Powered by the computer		
Power	**Component**	**Consumption**		**Power**	
		Nominal	Maximal	Nominal	Maximal
	Reader	670 mA	2080 mA	16 watts	50 watts
	Camera	200 mA	430 mA	1 watt	2.15 watts
	Note: The reader is supplied with its power cord according to region.				
Environmental Operating Temperature	15 °C to 40 °C (59 °F to 104 °F)				
Humidity	< 90 %				
Altitude	< 3,000 m (9,842 ft.)				
Regulatory Information	We certify that the EZ-Fluo™ Reader and the EZ-Fluo™ Camera were designed and manufactured in application of the following European Council directives: • Electromagnetic compatibility 2004/108/EC • Restriction of the use of certain Hazardous Substances in electrical equipment (RoHS) 2002/95/EC			
Materials of Construction	**Component**	**Materials of Construction**		
	Reader			
	Housing	Painted aluminum sheet 1050		
	Optical chamber	304L stainless steel		
	Handle	304L stainless steel		
	Optical filter	Glass		
	LED protector	Transparent polycarbonate		
	Foot	Polyvinylchloride (PVC)		
	Labels	Polyester		
	Screen	Protector polyester		
	Camera			
	Front plate	304L stainless steel		
	Housing	Painted aluminium alloy		
	Window	Glass		
	Light Guard	Clear polymethyl methacrylate (PMMA)		

System Components

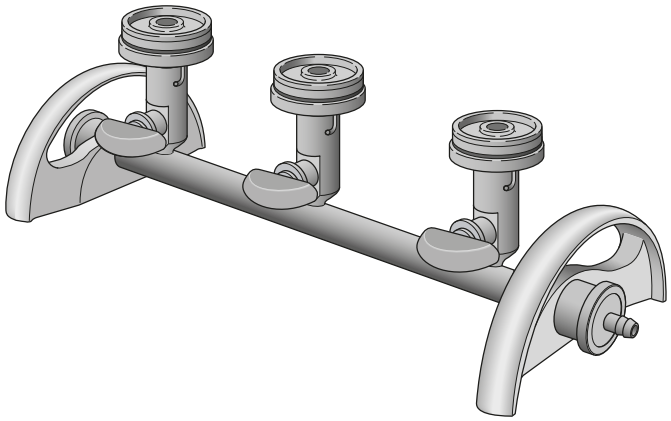
The EZ-Fluo™ Reader



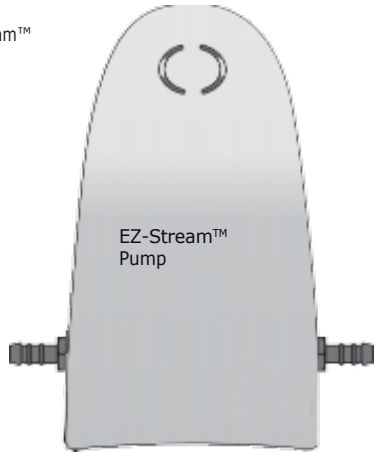
The EZ-Fluo™ Camera



The EZ-Fit™ Manifold



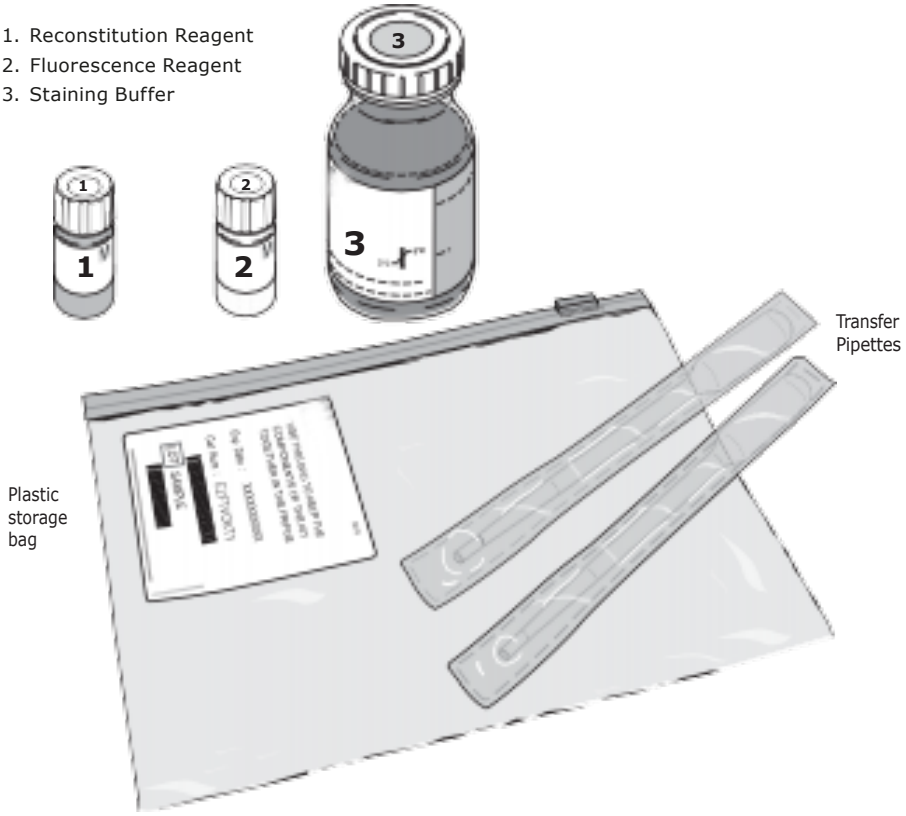
Recommended equipment



The EZ-Fluo™ Reagent Kit

The EZ-Fluo™ Reagent Kit consists of reconstitution reagent, fluorescence reagent, staining buffer, plastic storage bag, and transfer pipettes.

The kit components must be stored at 2 to 8 °C. One EZ-Fluo™ Reagent Kit contains sufficient material for 57 tests.



Installing the EZ-Fluo™ Reader

The EZ-Fluo™ Reader weighs 4.4 kg (9.7 lbs) and is designed for use on a laboratory workbench.

1. Remove all components and their accessories from the box.

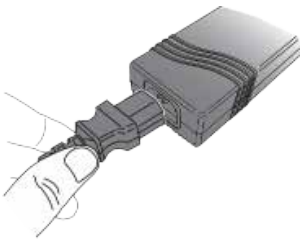
2. Remove the reader from the protective bag and place it on a flat, stable, horizontal surface.

Note: Please retain all packing materials. If the unit needs to be shipped, it should be packed in the original materials

3. Remove the protective film from the window.

4. Connect the end of the external power supply cord to the power outlet on the reader.

5. Connect the power cord to the external power supply. Connect the power cord to a power source.

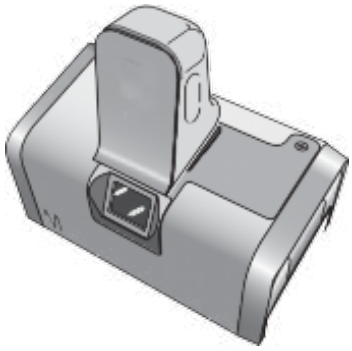


The system starts up. The auto test sequence runs and verifies that the EZ-Fluo™ Reader is ready to use:

Installing the Camera (Optional)

Installing the Camera onto the Reader

Place the camera on the top of the EZ-Fluo™ Reader as shown here. The camera magnetically locks onto the reader.



Installing the Software onto the Computer

System and Software Requirements

- 2 USB ports
- Minimum 512 MB of RAM memory; 1 GB recommended
- Minimum disk space: 500 MB
- Processor recommended: Intel® Core™2 Duo
- Minimum screen resolution 1024 x 768 – recommended: 1280 x 1024
- Operating system:
  - Microsoft Windows XP® 32 bits and 64 bits
  - Microsoft Windows Vista® 32 bits and 64 bits
  - Microsoft Windows 7® 32 bits and 64 bits
  - Microsoft Windows 10® 64 bits
  - Microsoft Windows 11®

Microsoft .NET Framework 2.0 SP2 is also recommended (Note: this is installed automatically during the installation of the EZ-Fluo™ Counter Software)

- Adobe Acrobat® Reader 9.0 or higher to display the user guide

Note: Administrator access to the computer is required to install the EZ-Fluo™ Spot Counter software. All screenshots shown are examples and may vary from one computer to another.

1. Switch on the computer.

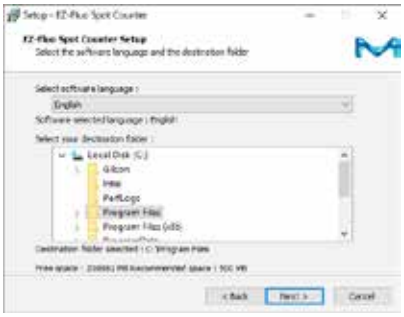
2. Log on with an administrator account.

3. Insert the installation disc into the CD drive of the PC.

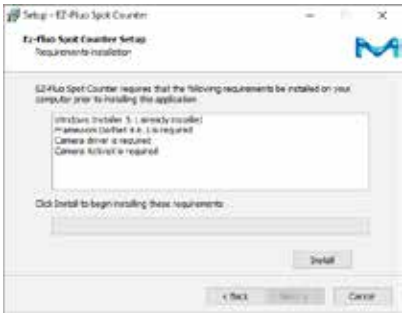
4. Using the file explorer, browse to the CD-Rom and double-click the Setup.exe file to start the installation.

5. The installation wizard for EZ-Fluo™ Spot Counter displays. Click **Next.**

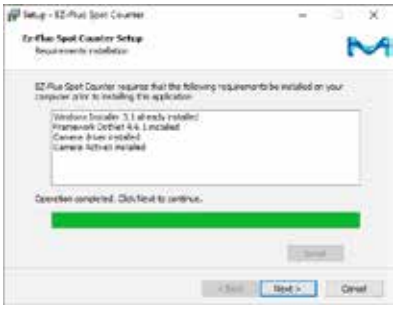
6. Select the language for the software. Select the destination folder for the software. Click **Next.**



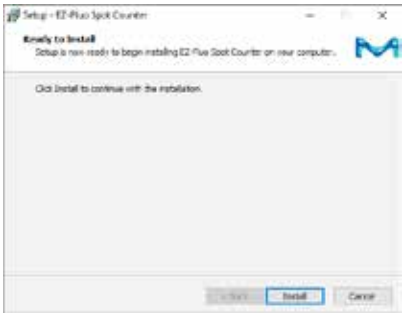
7. A new window displays, listing the components required to complete the EZ-Fluo™ software installation. Click **Install**.



8. When the installation of required components is completed, click **Next.**



9. A message indicating that the software is now ready to be installed displays. Click **Install.**



10. A message indicating that the computer must be restarted in order to complete the installation of the software displays. Save and close all the other applications. Then, select **Yes**, restart the computer now and click **Finish.**

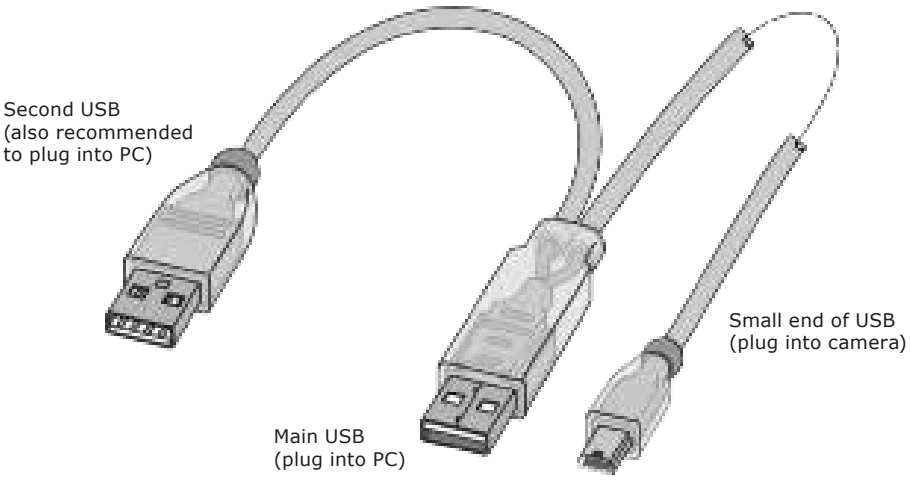
11. After the computer has restarted, the EZFluoSpotCounter icon is displayed on the desktop.

Note: Do not open the EZ-Fluo™ Spot Counter software before installing the EZ-Fluo™ Camera.

Connecting the Camera to the PC

1. Connect the small end of the USB cable to the camera.

2. The USB cable is equipped with two plugs. Connect the main plug to a USB port of the PC. Connect the second plug to another USB port of the PC, if available.



Note: The main plug of the camera USB cable must be connected to the same USB port during installation and use or the PC will not recognize the camera

3. The PC detects the EZ-Fluo™ Camera and automatically installs some components.
Wait until installation is finished.

4. The installation is complete.

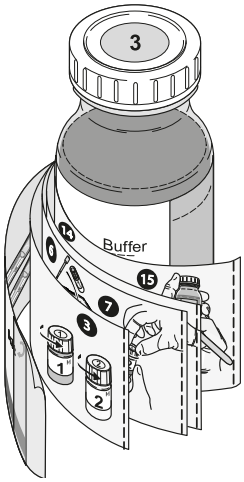
Preparing the Reagent Kit

Precautions and Methods

- Do not use any component in the kit that has expired.
- Remove the kit from the refrigerator one hour before starting the reconstitution.
- Reconstitute in a controlled-area environment.
- During the seven-day shelf life, do not let the Staining Solution remain at room temperature for more than four hours per day.
- After removing the Staining Solution from the refrigerator, wait one hour before performing a staining.
- Use appropriate personal protective equipment when using the Staining Solution.
- Mix the Staining Solution before performing the first test of the day.

Reconstituting the Staining Solution

Reconstitute the Staining Solution following the procedure on the Staining Buffer before the first use of the reagent kit. The reconstitution date may be noted on the Staining Buffer label.



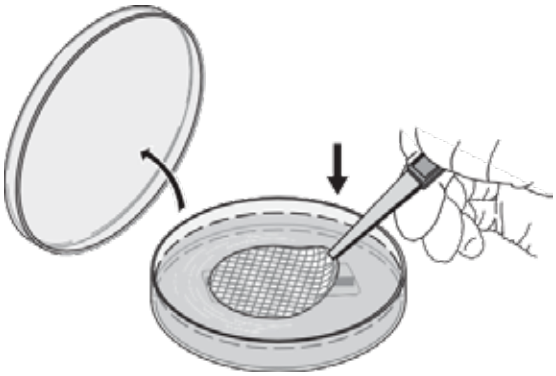
Store the reconstituted Staining Solution at 2 to 8 °C. One EZ-Fluo™ Reagent Kit contains sufficient material for 57 tests. The approximate number of remaining tests is indicated on the Staining Buffer (vial 3) label. The shelf life of the reconstituted Staining Solution is seven days.

Note: To extend the shelf life, the reconstituted Staining Solution can be distributed in up to four aliquots. These aliquots can be stored up to one month in the freezer. Once thawed, the aliquot must be stored at 2 to 8 °C, and protected from light. The shelf life of the thawed aliquot is seven days

Filtering the Sample

1. Perform the sample filtration using EZ-Fit™ Filtration System as described in the EZ-Fit™ Filtration Unit User Guide. The optimal vacuum is obtained using the EZ-Stream™ Pump (see EZ-Stream™ Pump User Guide).

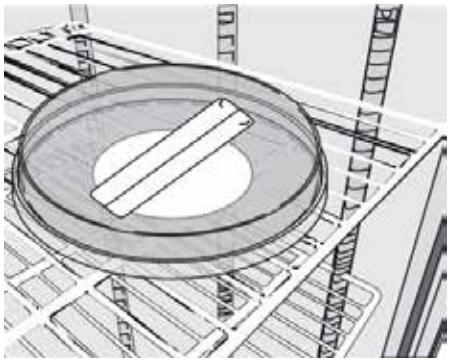
2. Transfer the membrane into a plate that contains culture medium. Ensure that the membrane is distributed directly and uniformly on the culture medium to avoid the formation of wrinkles and air bubbles.



3. Place the lid on the plate.

Incubating the Membrane

Place the plate that contains the membrane into the incubator, with the plate lid facing down.



Staining the Membrane

1. Remove the plate that contains the membrane from the incubator and transport it into a controlled environment area.

2. Remove the lid of a Petri-Pad™.

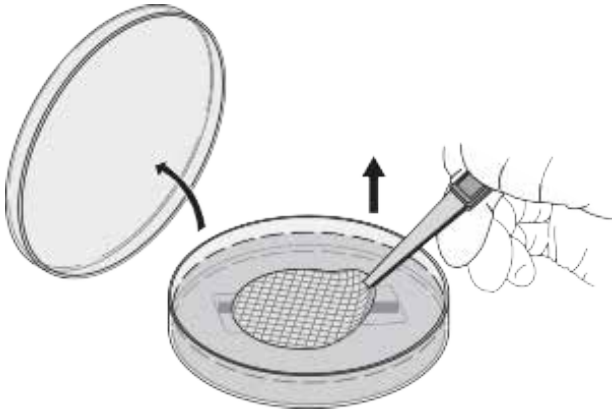
3. Using a pipette, add 1.7mL of Staining Solution on the center of the pad of the Petri-Pad™.

Note: If necessary, continue adding small amounts of Staining Solution until the pad is saturated.

4. Place the lid on the Petri-Pad™.

5. Remove the lid of the plate that contains the membrane.

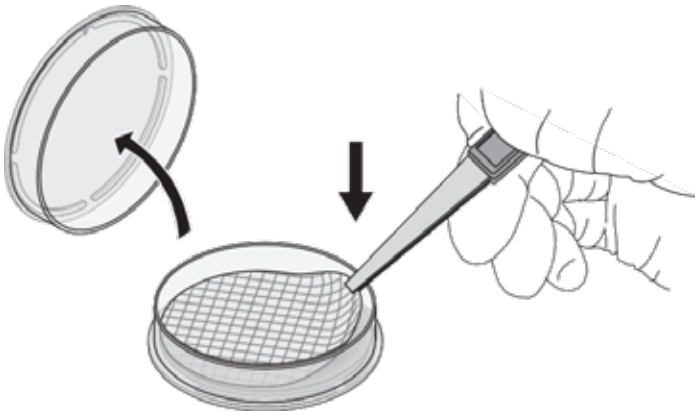
6. Using forceps, separate the membrane from medium.



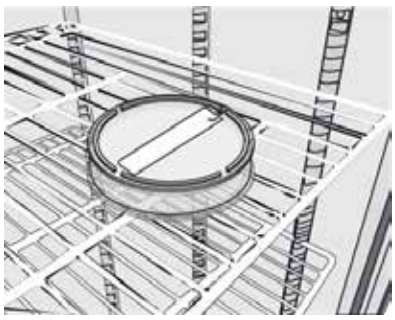
7. Open the lid of the Petri-Pad™ that is wetted with Staining Solution.

8. Transfer the membrane into the Petri-Pad™. Ensure that the membrane is distributed directly and uniformly on the medium to avoid the formation of wrinkles and air bubbles.

9. Close the lid.



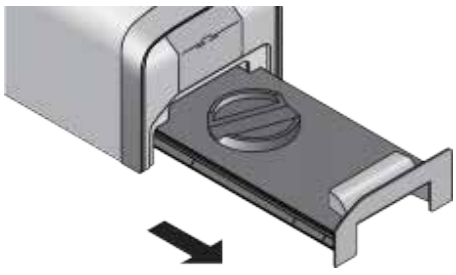
10. Incubate the Petri-Pad™ that contains the membrane for 30 minutes at 32.5 ± 2.5 °C with the Petri-Pad™ lid facing down.



Note: To ensure optimal results, do not stack multiple Petri-Pad™Dishes in the incubator.

Installing the Membrane in the Reader

1. After the staining step, remove the Petri-Pad™ that contains the membrane from the incubator.
2. Verify that the reader is on and ready to use.
3. Open the reader drawer and place the Petri-Pad™ in the drawer.



4. Close the drawer.

Reading the Membrane

1. When the reader drawer is closed, the reader lights the membrane.

2. Read the membrane two ways:

- Read the membrane manually, directly through the reader viewing window
- Read the membrane using the EZ-Fluo™ Camera and EZ-Fluo™ Spot Counter Software

This section contains instructions for both methods.

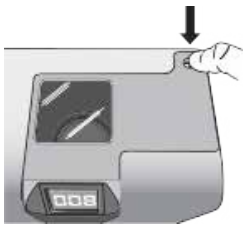
Note: If the EZ-Fluo™ Reader drawer remains closed for 10 minutes, and there is a Petri-Pad™ inside, the LED lights inside the reader switch off and the system enters standby mode.

To relight the membrane, open and close the drawer. This resets the counter on the reader screen. If the EZ-Fluo™ Reader is not used for more than 30 minutes, and there is no Petri-Pad™ inside, the reader goes into sleep mode. To reactivate the reader, push the counting button or open the drawer and insert a Petri-Pad™ containing a membrane. Close the drawer. When the reader is reactivated, the auto test sequence runs.

Reading the Membrane manually

To read the membrane manually, view the membrane directly through the reader viewing window.

1. Push the counting button to increase the count. The screen on the reader displays the counter.



Note: To make counting easier, align the membrane grid with the drawer

2. After the counting is complete, record the count and open the drawer to remove the Petri-Pad™ from the reader.

Note: Five seconds after the drawer opens, the counter disappears and the message Waiting for Petri-Pad™ is displayed on the screen.

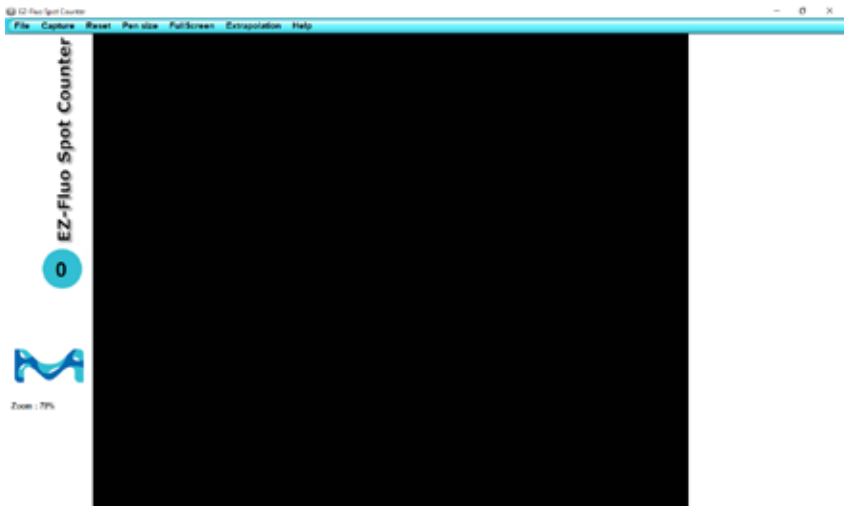
3. Close the drawer.

Reading the Membrane with the EZ-Fluo™ Camera and EZ-Fluo™ Spot Counter Software (optional)

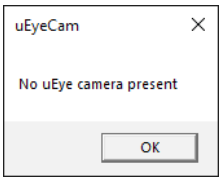
Note: In case of use of black membrane, do not use the camera to read the membrane

Starting the EZ-Fluo™ Spot Counter Software

1. Double-click the  icon on the desktop. The software opens.



Note: If the EZ-Fluo™ Camera is not connected to the PC, the message below displays before the opening of the software.



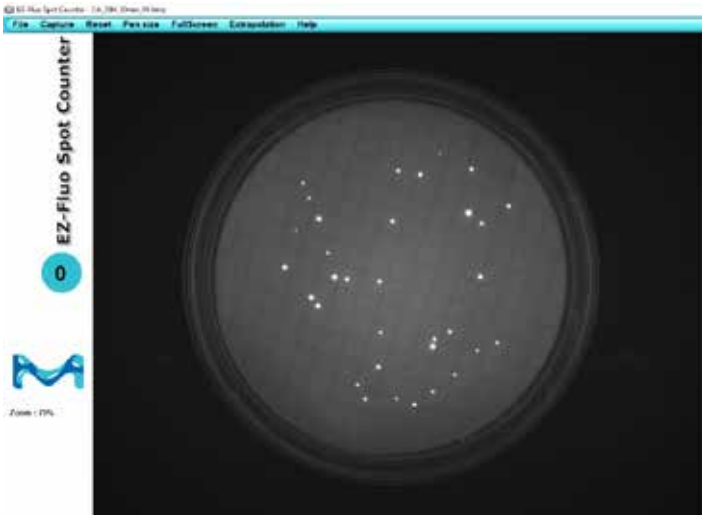
In order for the computer to recognize the camera, the camera must be connected to the same USB port during use that it was connected to when it was installed.

Capturing an Image

1. Place a Petri-Pad™ containing a membrane into the reader.
2. In the menu, click Capture (or press Ctrl + space).

Note: During the image capture, a progression bar displays and the camera LED light is orange.
A blinking orange LED light indicates an error.

3. The membrane image is displayed.



Counting the Spots

1. Mouse over the membrane image. The cursor changes to a cross.
2. Using the mouse, place the cross on a spot.
3. Left-click to mark the spot and increase the counter reading. The counter is located on the screen beneath the words "EZ-Fluo™ Spot Counter."

Note: To delete a marking, place the cursor on the spot you want to delete and right-click

Changing the Size of the Image

To increase the size of the image, use the scroll wheel of the mouse.

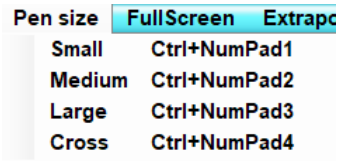
Changing to Full Screen Mode

1. In the menu, click **FullScreen**. The counter value and the zoom factor display in the menu.

Click **FullScreen** again to return to normal display mode.

Changing the Pen Size

1. In the menu, click Pen size.



2. Select the pen size you want to use. Several pen sizes can be used to mark the spots of one membrane.

Note: The standard pen size is Medium.

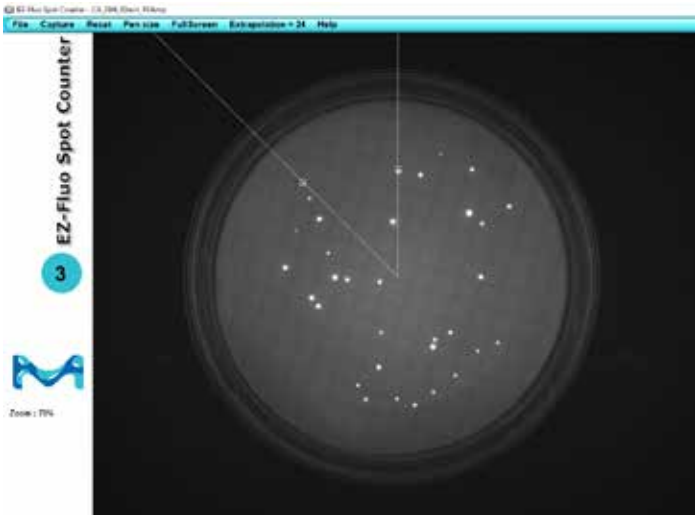
Counting by Extrapolation

When there are too many spots on the membrane to count one by one, use the Extrapolation function to estimate the number of spots contained on the entire membrane.

Note: When using the extrapolation function, mark spots only within the outlined segment.
Activating the extrapolation function resets the counter.

1. In the menu, click **Extrapolation.** A one-eighth segment of the membrane is outlined.

2. Using the method outlined above, count the spots in the outlined segment.



The extrapolated number of spots for the entire membrane is indicated at the top of the screen, in the menu.

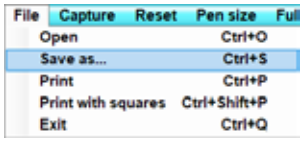
3. Click **Extrapolation** again to quit this function.

Resetting the counter

In the menu, click **Reset** to erase all markings and reset the counter.

Saving an Image

1. In the menu, click **File.** Select **Save as.**



2. Select the folder you want to save the image in.

3. Select the format for the saved image (.bmp or .jpg).

Note: For best image quality, choose .bmp format.
Images are saved without the counting marks

4. Enter the picture name in the File name field.

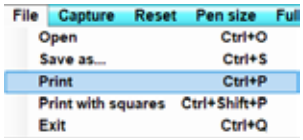
5. Click **Save.**

Printing an Image

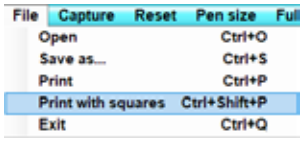
Images can be printed with or without counting marks.

1. In the menu, click **File.**

2. To print the image without counting marks, select **Print.**



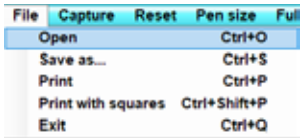
3. To print the image with counting marks, select **Print with squares.**



Opening a Saved Image

Note: Once the image opens, the image functions are available.
The EZ-Fluo™ Camera does not need to be connected to open and work on a saved image.

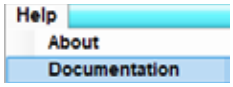
1. In the menu, click **File.** Select **Open.**



2. Navigate to the file to be opened.

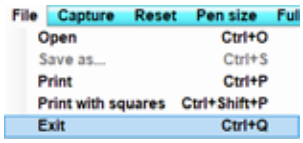
Using Help Menu

In the menu, click **Help** and select **Documentation** to access the online version of this user guide.



Closing the EZ-Fluo™ Spot Counter Software

In the menu, click **File** and select **Exit.**



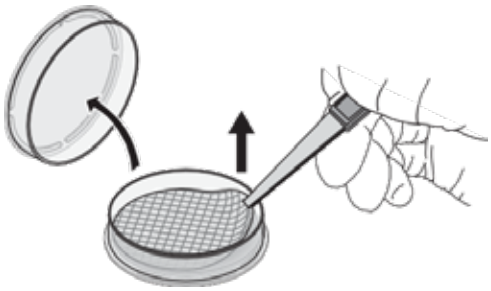
Performing a Reincubation (optional)

Follow this procedure if reincubation is necessary after the membrane is read.

1. Have ready a new plate that contains adequate medium.

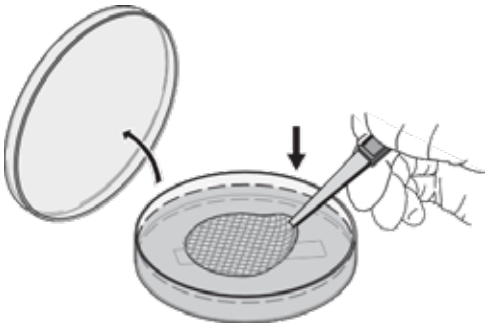
2. Remove the lid from the Petri-Pad™.

3. Using forceps, gently lift the membrane from the pad.



Note: Be careful to not damage the membrane.

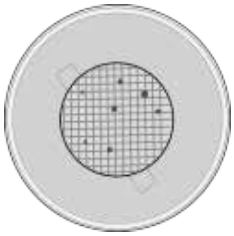
4. Transfer the membrane into a new plate.



5. Place the plate that contains the membrane in the incubator, with the lid facing down.

6. Remove the plate with the membrane from the incubator.

7. Count the colony forming unit (CFUs).



8. Take sample of the colonies and identify them following the appropriate standard protocol.

Storing the Reader

If the reader will not be used for an extended period of time, make sure the drawer is empty, close the drawer, and unplug the power cord from the power source.

Maintenance

Cleaning the EZ-Fluo™ Reader and Camera

1. Disconnect the reader from the power supply before cleaning.

2. Use only nonwoven wipes soaked in one of the products listed below. Do not spray any cleaning products, solvents, or abrasives on or into the EZ-Fluo™ Reader or Camera, as these could damage the instruments.

Cleaning Product	Dilution
Quaternary ammonium (NH4) Biguanide Propanol (10 to 40%)	Ready to use
Isopropyl alcohol 70% (aerosol)	Ready to use
Ethanol 70%	Ready to use
Sodium Hypochlorite	250 ppm
Peracetic acid at 5% Acetic acid at 7% Hydrogen peroxide at 27%	2.5%
Ethanol 41% Polyhexamethylene biguanide hydrochloride N-(3-aminopropyl)-N-dodecylpropane-1,3-diamine Didecyldimethylammonium chloride	Ready to use

Daily Cleaning: Reader and Camera

1. Gently clean the reader viewing window and the screen using a nonwoven wipe soaked in one of the products listed in the table in this section.

2. Gently clean the membrane site on the reader drawer using a nonwoven wipe soaked in one of the products listed in the table in this section.

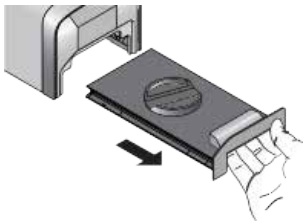
3. Clean the window of the EZ-Fluo™ Camera using a nonwoven wipe soaked in one of the products listed in the table in this section.

Weekly Cleaning: Reader and Camera

Disinfect all the exterior surfaces of the reader and the camera using a nonwoven wipe soaked in one of the products listed in the table in this section.

Monthly Cleaning: Reader

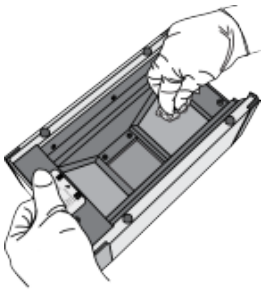
1. Remove the drawer from the reader by pulling firmly on the drawer.



2. Gently clean the drawer using a nonwoven wipe soaked in one of the products listed in the table in this section.

Note: The drawer is not autoclavable.

3. Invert the reader. Gently clean the interior of the reader using a nonwoven wipe soaked in one of the products listed in the table in this section.



4. Replace the drawer inside the EZ-Fluo™ Reader.

Annual Servicing

A complete check of the EZ-Fluo™ System should be performed by one of our authorized technicians annually.

Troubleshooting

Error Message or Symptom	Possible Cause	Remedy
The following message displays: Too bright! Check your reader.	The membrane is not correctly lit.	Verify that the camera is correctly placed on the reader.
		Open and close the drawer to reactivate the membrane lighting.
		Verify that the second plug of the camera USB cable is connected to a USB port on the PC. If the problem persists, disconnect other peripheral devices connected to USB ports of the PC. Then close and restart the software.
The following message displays: Nothing to capture! Check your reader.	The membrane is not correctly lit.	Verify that the camera is correctly placed on the reader.
		Verify that the reader is connected to power source.
		Open and close the drawer to reactivate the membrane lighting.
		Verify that the second plug of the camera USB cable is connected to a USB port on the PC. If the problem persists, disconnect other peripheral devices connected to USB ports of the PC. Then close and restart the software.
No image is displayed and the progress bar is blocked during an image capture.	Incorrect exposure time.	Verify that the camera is correctly placed on the reader.
		Verify that the reader is connected to power source.
	The membrane is not correctly lit.	Verify that the camera is correctly placed on the reader.
		Verify that the reader is connected to power source.

Protocol		
The Staining Solution is leaking from the Petri-Pad™.	The wrong side of the Petri-Pad™ has been used.	Make sure to put the staining solution into the Petri-Pad™ base (the face with the engraved logo).
Staining Solution droplets on the Petri-Pad™ lid inhibit the reading.	The Petri-Pad™ was inverted before the membrane transfer.	Do not invert the Petri-Pad™ before the membrane being transferred.
	The staining solution has not been completely absorbed by the pad and a droplet has fallen on the Petri-Pad™ lid during the staining step.	Wipe the Petri-Pad™ lid in a controlled environmental area.
During the reading step, presence of dark area(s) on the membrane cause some spots to be less well marked.	The volume of staining solution (1,7mL) applied to the pad was not sufficient to saturate the pad.	Add additional staining solution in small amounts until the pad is fully saturated.
	Air bubble(s) formed during the incubation step.	Ensure that the membrane is distributed directly and uniformly on the medium to avoid the formation of wrinkles and air bubbles.
	Air bubble(s) formed during the staining step.	Ensure that the membrane is distributed directly and uniformly on the medium to avoid the formation of wrinkles and air bubbles.

If any of the above problems persists, contact Technical Support.

Troubleshooting

Error Message or Symptom	Possible Cause	Remedy
EZ-Fluo™ Reader		
The light (LED) and the screen on the reader are off.	The reader is not connected to power source.	Connect to power source.
	The reader is connected to power source.	Contact Technical Support.
The light on the reader is on but the screen is off.	The reader is in sleep mode.	To reactivate the reader, push the counting button or open the drawer and place a Petri-Pad™ containing a membrane inside.
The membrane is not lit when the reader drawer is closed.	The reader is not connected to power source.	Connect to power source.
	The drawer is not properly closed.	Close properly the drawer.
	The Petri-Pad™ moved away from the drawer membrane site when the drawer was closed.	Open the drawer and verify that the Petri-Pad™ is properly located in the drawer membrane site. Close the drawer.
The Petri-Pad™ is lit but the Millipore logo is visible instead of the membrane.	The Petri-Pad™ is inserted in the reader upside down.	Invert the Petri-Pad™.
The counting function does not work.	The counting button does not work properly.	Contact Technical Support.
The VERIFICATION NEEDED message displays at the top of each screen.	Some of the LEDs inside the reader are not functioning properly.	Contact Technical Support.
The CALIBRATION NEEDED message displays at the top of each screen.	30 days before the calibration due date, the message is displayed at the top of each screen.	Contact us to calibrate the reader. If the calibration due date is missed, the reader can be used; however, the performance is not guaranteed.
	The LED's on-time limit has been reached.	
The following message displays: System out-of-order – Please contact Technical Support.	The reader is out of order.	Contact Technical Support.

EZ-Fluo™ Camera and Spot Counter		
The following message displays: No uEye camera present.	The camera is not properly connected.	Verify that the main plug of the camera USB cable is connected to the USB port that was used during installation.
The following message displays: Could not open camera! Check your connection.		
The following message displays: The camera is not connected.		
The following message displays: Transfer error.	The camera is not properly connected.	Verify that the main plug of the camera USB cable is connected to the USB port used during the installation.
		Close and restart the software.
		Verify that the second plug of the camera USB cable is connected to a USB port of the PC. If the problem persists, disconnect other peripheral devices connected to USB ports of the PC. Then close and restart the software.

Ordering Information

Description	Qty/ Pk	Catalog No.
EZ-Fluo™ System Kits		
EZ-Fluo™ System standard kit including 1 reader, 1 camera and software card	1	EZFKIT001WW*
EZ-Fluo™ Hardware		
EZ-Fluo™ reader	1	EZFREAD01WW*
EZ-Fluo™ camera	1	EZFCAM001
EZ-Fluo™ Accessories		
EZ-Fluo™ validation protocol, US letter format	1	EZFLLTVP1
EZ-Fluo™ validation protocol, A4 format	1	EZFLA4VP1
Manifold 3-Place for EZ-Fit filtr.Units	1	EZFITEFUN3
EZ-Stream™ Pump	1	EZSTREAM1
Silicone Hose for EZ-Stream™ Pump Autoclavable Internal diameter: 9.5mm (3/8 in) Length: 5.0 m (196.8 in)	1	STREAMTUB
PVC Hose for EZ-Stream™ Pump Not autoclavable Internal diameter: 9.5mm (3/8 in) Length: 3.0 m (118.1 in)	1	XX6700034
Filter forceps	1	XX6200006P

	Description	Packaging / Format	Qty/ Pk	Catalog No.
PINK base — no pad	EZ-Fit® Filtration Unit, white plain PVDF membrane, 0.45 µm, 100 mL	Single	48	EFHVW10IS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 100mL	Single		EFHAW100IS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 100 mL	Multipack of 3 units		EFHAW10MS
	EZ-Fit Filtration Unit, white gridded MCE membrane, 0.45 µm, 100 mL	Bulk with protective bag		EFHAW10BS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 250 mL	Bulk with protective bag		EFHAW25BS
	EZ-Fit™ Filtration Unit, black gridded MCE membrane, 0.45 µm, 100 mL	Multipack of 3 units		EFHAB10MS
	EZ-Fit® Filtration Unit, black gridded MCE membrane, 0.45 µm, 100 mL	Bulk with protective bag		EFHAB10BS
	EZ-Fit® Filtration Unit, black gridded MCE membrane, 0.45 µm, 250 mL	Bulk with protective bag		EFHAB25BS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.22 µm, 100 mL	Multipack of 3 units		EFGSW10MS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.22 µm, 100 mL	Single		EFGSW10IS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.22 µm, 250 ML	Single		EFGSW25IS
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.8 µm, 100 mL	Bulk with protective bag		EFAAW10BS
	EZ-Fit® Filtration Unit, black gridded MCE membrane, 0.8 µm, 250 mL	Bulk with protective bag		EFAAB25BS

BLUE base — with pad	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 100 mL	Bulk	48	EFHAW100B
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 100 mL	Single		EFHAW100I
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 100 mL	Single with plug		EFHAW100IP
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 100 mL	Multipack of 3 units		EFHAW100M3
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 250 mL	Bulk		EFHAW250B
	EZ-Fit® Filtration Unit, white gridded MCE membrane, 0.45 µm, 250 mL	Single		EFHAW250I
	EZ-Fit® Filtration Unit, black gridded MCE membrane, 0.45 µm, 100 mL	Bulk		EFHAB100B
	EZ-Fit® Filtration Unit, black gridded MCE membrane, 0.45 µm, 100 mL	Single		EFHAB100I
	EZ-Fit® Filtration Unit, black gridded MCE membrane, 0.45 µm, 100 mL	Multipack of 3		EFHAB100M3
EZ-Fit® Manifold Base with High Throughput Application (HTA) Manifold Head, 1-place for EZ-Fit® filtration units			1	EZFITSAM1
EZ-Fit® Manifold Base with High Throughput Application (HTA) Manifold Head, 3-place for EZ-Fit® filtration units			1	EZFITSAM3
EZ-Fit® Manifold Base with High Throughput Application (HTA) Manifold Head, 6-place for EZ-Fit® filtration units			1	EZFITSAM6
EZ-FIT® Manifold Head High-Throughput Application (HTA), head for EZ-Fit filtration unit			1	EZFITMHTA1
EZ-FIT® Manifold Head High-Throughput Application (HTA), head for EZ-Fit filtration units			1	EZFITMHTA3

Standard Product Warranty

The applicable warranty for the products listed in this publication may be found at: **www.millipore.com/ec/cp3/terms**

(within the "Terms and Conditions of Sale" applicable to your purchase transaction).

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