

# LUNA FX7<sup>TM</sup>

Automated Cell Counter

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## User Manual

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## **DISCLAIMER**

The contents of this document are subject to change without notice.

The LUNA-FX7™ Automated Cell Counter is an electrical laboratory instrument for scientific research use only.

It is not a medical, therapeutic, or in vitro diagnostics device.

Do not disassemble the device on any occasion as this will invalidate your warranty.







## **TRADEMARKS**

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Logos Biosystems is the brand name of Life Science business of Aligned Genetics, Inc.

## CERTIFICATION MARKS

	<p>The WEEE (Waste Electrical and Electronic Equipment) symbol indicates that users of this instrument have responsibility of returning and disposing of WEEE in an environmentally friendly manner.</p> <p>Follow the waste ordinances of your region for proper disposal provisions.</p>
	<p>The CE mark indicates that this instrument conforms to all applicable European Community provisions for which this marking is required. Users must be aware of and follow the conditions described in this manual for operating the instrument. The protection provided by the instrument may be impaired if the instrument is used in a manner not specified by this manual.</p>
	<p>Protective earth (Ground)</p>
	<p>This device complies with Part 15 of the FCC Rules.</p>
	<p>This equipment complies with the requirement of UL 61010-1:2012, CAN/CSA C22.2 No.61010-1:2012.</p> <p>“Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use, Part 1: General Requirements.”</p>
	<p>The KC certification mark indicates that this instrument conforms with Korea's product safety requirements for electrical and electronic equipment and components for which this marking is required.</p>

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# Safety Precautions

## Instrument Safety

### General Safety

Operate the instrument in the conditions described in the Operating Conditions.

Install the instrument on a level and sturdy surface. Avoid vibrations from other devices.

Do not touch components with wet hands.

Use components provided or authorized by Logos Biosystems. If the proper combination of components is not used, product safety cannot be guaranteed.

Use only the power cord and AC adapter provided by Logos Biosystems. If the proper power cord and AC adapter are not used, electrical safety of the product cannot be guaranteed.

Ensure that the input voltage is compatible with the power supply voltage of the product.

Connect the grounding terminal of the instrument and electrical outlet properly. If the instrument is not grounded, electrical safety of the product cannot be guaranteed.

Turn on the instrument only after connecting the power cord and AC adapter to both the power source and the instrument. Turn off the instrument before disconnecting the power cord and/or moving the instrument.

Disconnect the power cord in the case of abnormalities.

Be careful with possible electric shock hazards as electric current may be still alive when the instrument stops.

Do not hold the slide slot while it is in motion.

Protect USB drives from being infected with viruses and malware.

### Operating Conditions

Operating Power	100 - 240 VAC, 1.5 A
Frequency	50/60 Hz
Electrical Input	12 VDC, 5.0 A
Installation Site	Indoor use only
Operating Temperature	10 - 35°C
Maximum Relative Humidity	10 - 80%
Altitude	≤ 2,000 m

### Instrument Disposal

Follow the rules and regulations of your local government.

### Instrument Disassembly

Do not disassemble the instrument in any event as this will invalidate your warranty.

If the instrument is damaged or malfunctioning, contact your local distributor or Logos Biosystems.

## Personal Safety

### Safety Guidelines

Read and understand all user manuals thoroughly before using the instrument.

Keep all user manuals in a safe and accessible place for future reference.

Read and understand all safety data sheets before storing, handling, or working with any reagents.

Wear appropriate personal protective equipment (PPE) when handling reagents and cell samples to avoid exposure.

When using toxic agents, radioactive materials, or pathogenic microorganisms belonging to WHO Risk Groups 2-4, follow national laws and regulations for biosafety level requirements.

This instrument is to be serviced by trained personnel only to avoid injury.

**Waste Disposal**

Do not reuse disposable slides. Used slides must be disposed as biohazardous waste according to the rules and regulations of your local government.

# Précautions de sécurité

## Sécurité des instruments

### Sécurité générale

Faites fonctionner l'instrument dans les conditions décrites dans les conditions de fonctionnement.

Installez l'instrument sur une surface plane et solide. Évitez les vibrations provenant des autres appareils.

Ne touchez pas les composants avec les mains mouillées.

Utilisez uniquement les composants fournis ou autorisés par Logos Biosystems. En cas d'utilisation d'une combinaison autre que celle qui a été recommandée, la sécurité du produit ne peut être garantie.

Utilisez uniquement le cordon d'alimentation et l'adaptateur fournis par Logos Biosystems. En cas d'utilisation du cordon et de l'adaptateur non appropriés, la sécurité électrique du produit ne peut être garantie.

Assurez que la tension d'entrée est compatible avec la tension d'alimentation du produit.

Connectez correctement la borne de mise à la terre de l'instrument et la prise électrique. Si l'instrument n'est pas mis à la terre, la sécurité électrique du produit ne peut pas être garantie.

Allumez l'instrument uniquement après avoir connecté respectivement le cordon d'alimentation et l'adaptateur à la source d'alimentation et à l'instrument. Éteignez l'instrument avant de débrancher le cordon d'alimentation et / ou de déplacer l'instrument.

Débranchez le cordon d'alimentation en cas d'anomalies.

Soyez prudent avec les risques d'électrocution, car le courant électrique peut être encore actif lorsque l'instrument s'arrête.

Ne tenez pas le tiroir de lame lorsqu'elle est en mouvement.

Protégez les clés USB contre les virus et les logiciels malveillants.

### Conditions de fonctionnement

Puissance de fonctionnement	100 - 240 VAC, 1.5 A
Fréquence	50 / 60 Hz
Entrée électrique	12 VDC, 5.0 A
Site d'installation	Utilisation en intérieur uniquement
Température de fonctionnement	10 - 35°C
Humidité relative maximale	10 - 80%
Altitude	≤ 2,000 m

### Destruction de l'instrument

Suivez les règles et réglementations de votre gouvernement local.

### Démontage de l'instrument

Ne démontez en aucun cas l'instrument car cela invaliderait votre garantie.

Si l'instrument est endommagé ou fonctionne mal, contactez votre distributeur local ou Logos Biosystems.

## Sécurité personnelle

### Consignes de sécurité

Lisez et comprenez attentivement tous les manuels d'utilisation avant d'utiliser l'instrument.

Conservez tous les manuels d'utilisation dans un endroit sûr et accessible pour référence future.

Lisez et comprenez toutes les fiches de données de sécurité avant de stocker, de manipuler ou de travailler avec des réactifs.

Porter un équipement de protection individuelle (EPI) approprié lors de la manipulation des réactifs et des échantillons cellulaires pour éviter toute exposition.

Lors de l'utilisation d'agents toxiques, de matières radioactives ou de micro-organismes pathogènes appartenant aux groupes de risque 2 à 4 de l'OMS, respectez les lois et réglementations nationales relatives aux exigences de niveau de biosécurité.

Cet instrument doit être entretenu par du personnel qualifié uniquement pour éviter les blessures.

### Traitement des déchets

Ne réutilisez pas les lames jetables. Les lames usagées doivent être éliminées comme des déchets biodangereux conformément aux règles et réglementations de votre gouvernement local.

# 1. Product Introduction

## Product Contents

**Product Contents** The LUNA-FX7™ Automated Cell Counter is shipped with the following components.

Component	Quantity
LUNA-FX7™ Automated Cell counter	1
Snap Holder (Inserted in the slide port)	1
Power Cord with AC Adapter	1
Cell Counting Slides Sample	2 ea / 4 slide types
Trypan Blue Stain, 0.4 %	2 x 1 mL
Acridine Orange/Propidium Iodide Stain	2 x 0.5 mL
LUNA-FX™ Calibration Beads Kit	1
WiFi Dongle	1
USB Drive	1
Installation Guide	1
Quick Start Guide	1

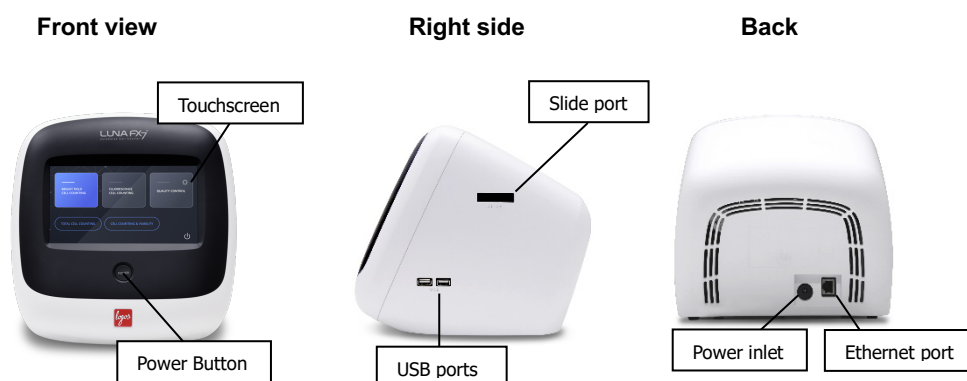
Inspect the product package upon delivery to ensure that all components have been included. Contact your local distributor or Logos Biosystems if anything is missing. Damage that may occur during shipping and handling is not covered by warranty and must be filed with the carrier.



## Product Description

### LUNA-FX7™ Automated Cell Counter

The LUNA-FX7™ is an automated, image-based cell counting device that features an accurate counting algorithm and increased counting volume and represents a fully automated solution for cell counting and viability analysis. The LUNA-FX7™ also provides flexible counting slide options from a single channel slide to a higher throughput, 8-channel slide.



#### Touchscreen

The LUNA-FX7™ has a 7-inch capacitive touchscreen for navigating the user interface.

#### Slide port

The automated slide port enables one-time slide insertion.

#### Power button

The power button is used for the main power control.

#### USB ports

USB ports allow the user to transfer or print cell count data. Data may be transferred via USB drive or the provided WiFi dongle. Counting data may be printed using the LUNA-FX7™ Printer (P17001).

#### Ethernet port

The Ethernet port allows the instrument to be connected to a computer network. The CountWire™ software package enables automated data synchronization and the ability to remotely operate the LUNA-FX7™.

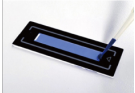




#### Power inlet

Connect the power inlet of the instrument to an electrical outlet with the supplied AC adapter and power cord.

### LUNA-FX7™ Cell Counting Slides

The LUNA-FX7™ gives you the flexibility to use various counting slide formats. The LUNA-FX7™ is compatible with the LUNA™ 1-, 3-, 8-Channel and Reusable Slide formats in addition to the standard LUNA™ Cell Counting Slides and PhotonSlides™. With single-time slide insertion, the LUNA-FX7™ is able to count all slide chambers at one time without needing

to remove and reinsert a slide. The increased counting volume yields more accurate and consistent results.

Channel No.	1 Channel	2 Channel	3 Channel	8 Channel	Reusable
Compatible Slides	LUNA™ 1-Channel Slides	LUNA™ Cell Counting Slides / PhotonSlide™	LUNA™ 3-Channel Slides	LUNA™ 8-Channel Slides	LUNA™ Reusable slides
					
Sample Throughput	1 sample	Up to 2 samples	Up to 3 samples	Up to 8 samples	1 sample
Sample Loading Volume	50 µL	10 µL/chamber	10 µL/chamber	10 µL/chamber	10 µL/chamber
Analysis Volume	5.1 µL	1.3 µL/chamber	1.3 µL/chamber	0.5 µL/chamber	1.3 µL/chamber

## 2. Getting Started

### Installation

#### Installation

Place the LUNA-FX7™ on a clean, level and sturdy surface.

- Avoid vibrations from other devices.
- Do not install the instrument in a location that will expose the device to intense ultraviolet light.
- Allow at least 5 cm (2 inches) free space at the back of the instrument to prevent overheating of the instrument.
- Allow at least 10 cm (4 inches) free space at the right of the instrument to insert/eject a cell counting slide easily.

Connect the instrument to electrical outlets using the supplied power cord and AC adapter.

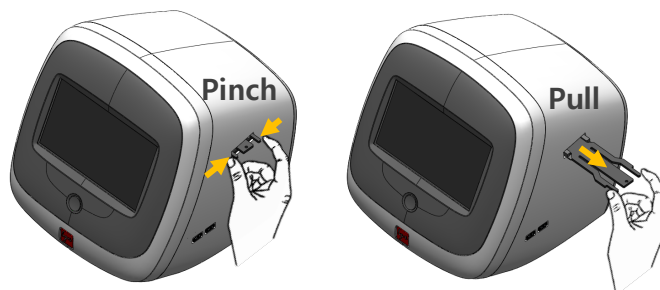
- Make sure the power cords are appropriate for your region.
- Always use power cord and AC adapter provided or approved by Logos Biosystems. If appropriate cord is not used, the electrical safety of the instrument cannot be guaranteed.

Connect the supplied WiFi dongle to a USB port.

(Optional) Connect a LUNA-FX™ Thermal Printer (P17001) to a USB port.

#### Snap Holder Removal

Remove the snap holder being inserted in the slide port. Pinch both handles to release the snap holder and pull it out of the slide port.

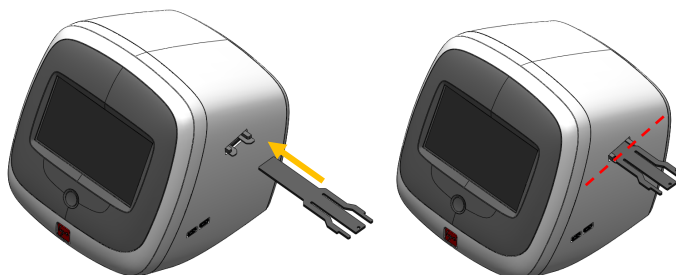


#### Snap Holder Insertion

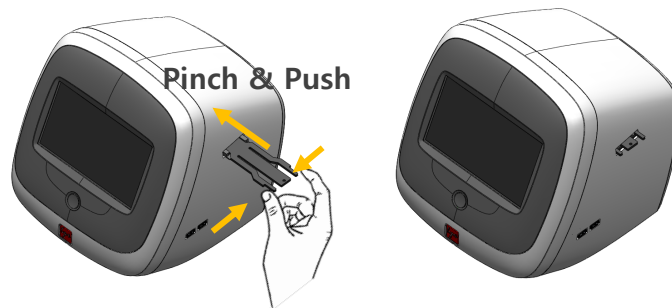
When moving the LUNA-FX7™ to another location, reinsert the snap holder and carry the instrument to avoid possible misalignment of the slide port due to excessive shaking.

Turn on the LUNA-FX7™ and enter any counting mode.

Press the EJECT button to have the slide port ejected.



Insert the snap holder, like attaching its bottom to the bottom of the slide port.  
Push the snap holder until the part where handles start.  
Press and hold the power button to turn off the LUNA-FX7™.  
Gently pinch both handles and push the snap holder inside until it reaches the end.  
The handle grooves should be caught and fixed inside.



## Setup

### LUNA-FX7™ Startup

Push the power button located below the touchscreen to turn on the instrument. After a short beep, the company logo will appear, followed by the home screen.



The home screen has three menus:

#### Brightfield Cell Counting

- Select Total cell counting mode or Cell counting & viability mode.
- Total cell counting mode is used to enumerate total cell numbers without staining cells.
- Cell counting & viability mode is used to count cells and calculate the viability of cells stained with the Trypan Blue Stain, 0.4% (T13001) or Erythrosin B Stain (Cat# L13002).

#### Fluorescence Cell Counting

- Select Cell lines & primary cells mode or Cell lines & primary cells, Advanced mode.

- Cell lines & Primary cells mode is used to count cells and calculate the viability of cells stained with fluorescence dyes, Acridine Orange/Propidium Iodide (AO/PI) (Cat # F23001).

Cell lines & Primary cell mode may also be used to count cells expressing GFP and/or RFP.

- Cell lines & Primary cells, Advanced mode is used to count cells and calculate the viability of cells with an improved cell detection and cell de-clustering capabilities.

### **Quality Control**

- Quality Control mode is only functional upon registration of Logos Biosystems brightfield or fluorescent validation slides.

- The Quality Control menu is used to monitor the accuracy and variability of the instrument.

- The validation slides contain pre-spotted patterns or pre-fixed beads with a known concentration and viability.

- Utilizing the Quality Control feature can provide daily, weekly, or monthly validation results that may be graphically displayed, and/or downloaded.

### **Screen Saver**

The screen backlight will automatically turn off after 10 minutes of inactivity. Touching the screen will reactivate the instrument.

# 3. Counting Cells

## Sample Preparation

### Sample Staining

#### Brightfield cell counting

For Total and viability cell counting, prepare a cell suspension according to standard procedures. Mix the sample, 1:1, with Trypan Blue Stain, 0.4% (T13001) or Erythrosin B (L13002). Mix gently, but thoroughly to ensure a homogenous suspension.

For total cell counting, load the sample directly onto the slide without staining the sample.

#### Fluorescent cell counting

Prepare a cell suspension according to standard procedures. Mix the sample, 9:1 (cells: stain), with Acridine Orange/Propidium Iodide Cell Viability Kit (F23001). Mix gently, but thoroughly to ensure a homogenous suspension.

### Sample Loading

Load the appropriate volume for each slide chamber according to the table below:

LUNA™ 1-Channel Slides	LUNA™ Cell Counting Slides & PhotonSlide™	LUNA™ 3-Channel Slides	LUNA™ 8-Channel Slides**	LUNA™ Reusable Slides
50 µL	10 µL	10 µL	10 µL	10 µL

\*\* The LUNA™ 8-Channel Slides are multi-channel pipette compatible.

For easy and accurate loading, hold the slides by their edges and pipette at a 45-60° angle to the slide. Take care not to overload or under-load the chamber.

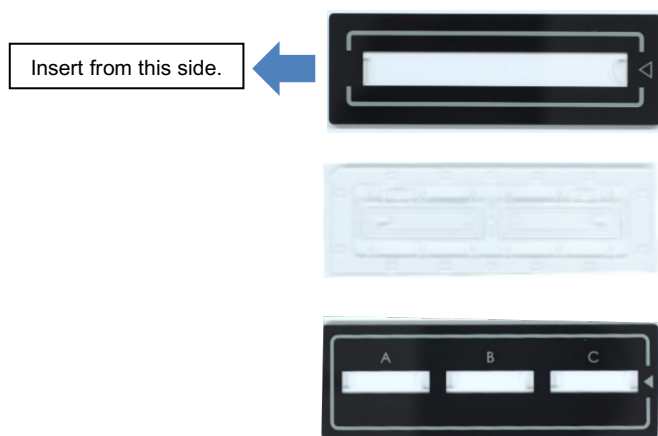
## Counting with the LUNA-FX7™

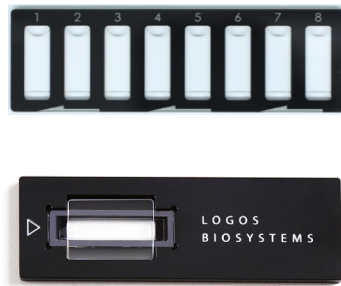
### Slide Insertion & Removal

Select appropriate counting mode and navigate to appropriate counting screen.

Press **EJECT**.

When inserting a slide into the instrument, ensure that the slide is facing up so that the arrow is showing on the right side and/or so that the lowest chamber designation is to the left.



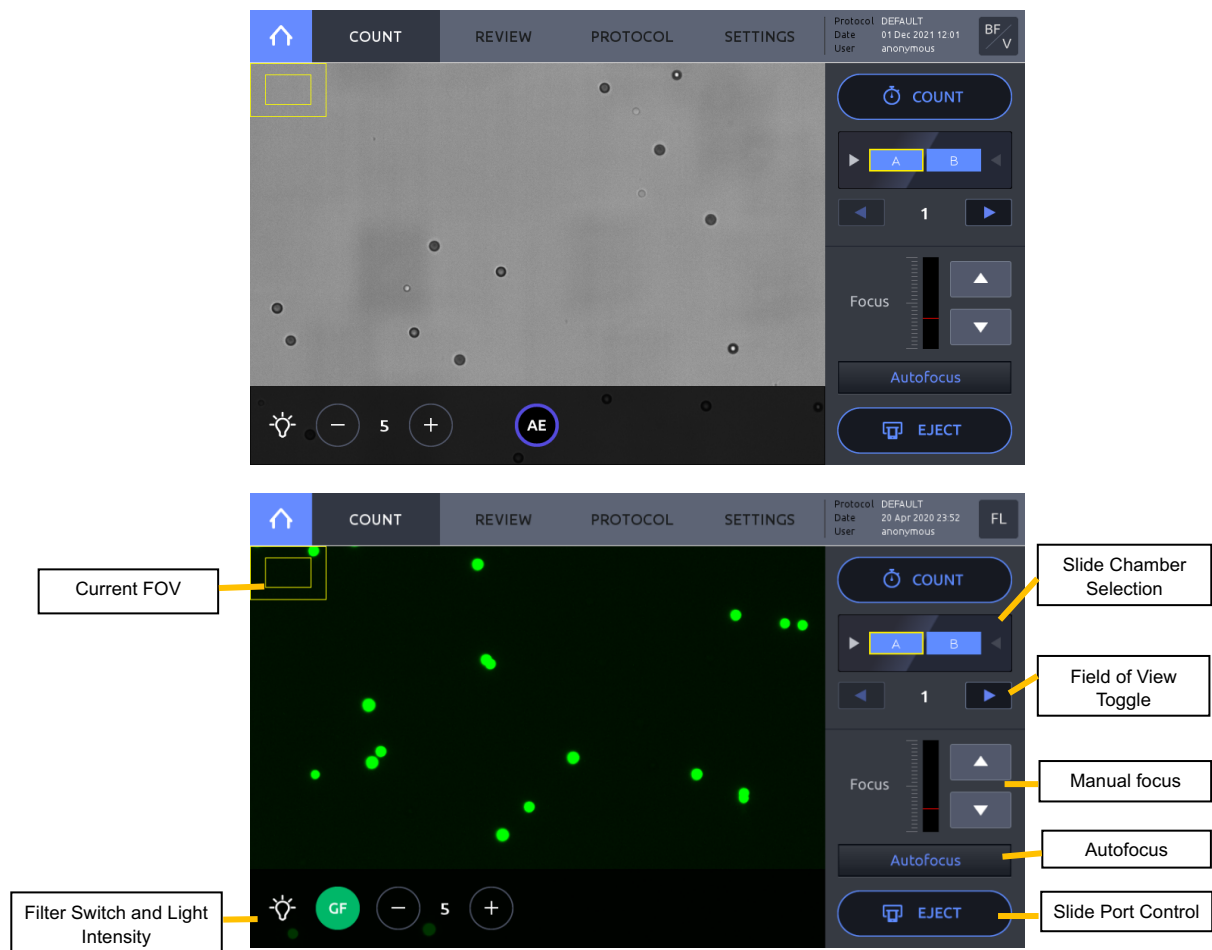


Press **INSERT**. The slide holder will automatically move into position.

To remove the slide, press **EJECT**. The slide holder will automatically extend out of the instrument and the slide may be removed.

## Viewing Images

By default, upon slide insertion, the viewing light will automatically turn on and the LUNA-FX7™ will perform an initial autofocus. Whether or not autofocus is performed upon slide insertion may be changed within **SETTINGS** (Section 8).



## Light

By pressing the lamp icon in the bottom left corner of the screen, a light control panel will appear. The intensity can be adjusted as needed. Press AE to use the auto-exposure function. When in the fluorescence counting screen, the filters also may be switched between the BF, Green, and Red channels. Photobleaching will occur with prolonged exposure, so work appropriately.

! **Important !** Adjusting light intensity levels in the COUNT screen will only be applied to the live view mode. Exposure levels for brightfield cell counting are automatically adjusted. Exposure levels for fluorescence cell counting may only be adjusted within a protocol (Section 5).

### Focusing

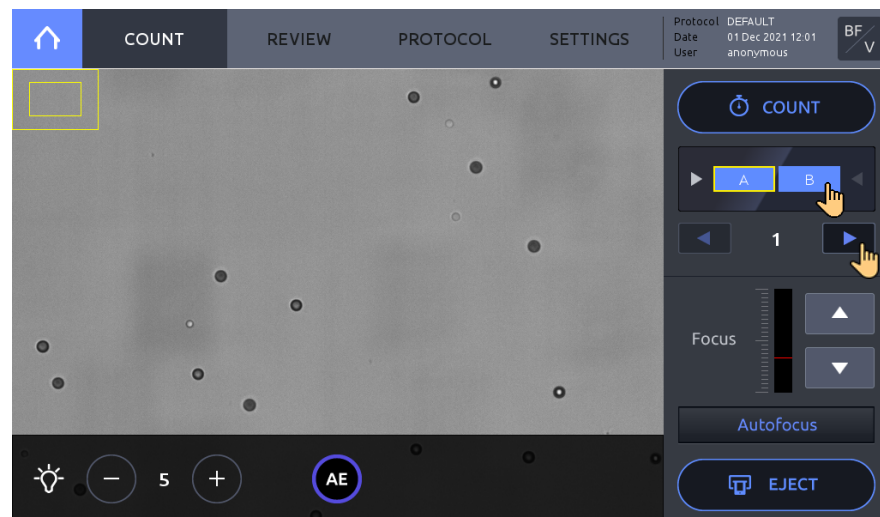
To bring cells into focus, press **Autofocus**. Focus may also be adjusted manually using the up & down arrows in the focus control bar.

### Zoom

Zoom in or out by spreading or pinching two fingers. The outer box in the upper, left hand corner of the viewing window represents the current field of view. The inner box represents the view on the screen. Zooming in or out will cause the inner box size to decrease or increase.

### Navigation

To view different slide chambers within a slide, select the chamber to be viewed by pressing a chamber on the slide image just under the **COUNT** button. To see different fields of view within a chamber, use the arrows located above the manual focus adjustment.



## Cell Counting

Prior to counting, confirm that the image is in focus for the first field of view. When the first field is in focus, press the **COUNT** button.

The LUNA-FX7™ will count all slide chambers as designated in **SETTINGS**.

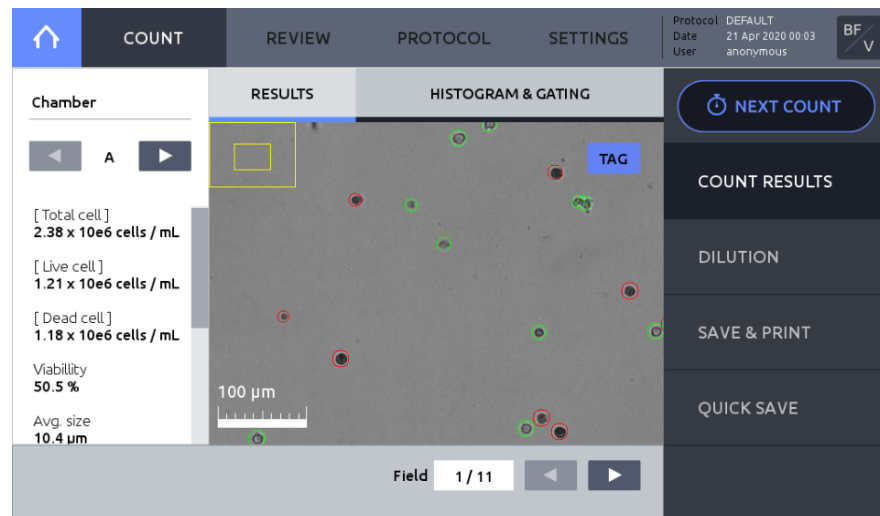
Counting time will vary depending on slide type, counting mode and the protocol used.



## Results

### Results

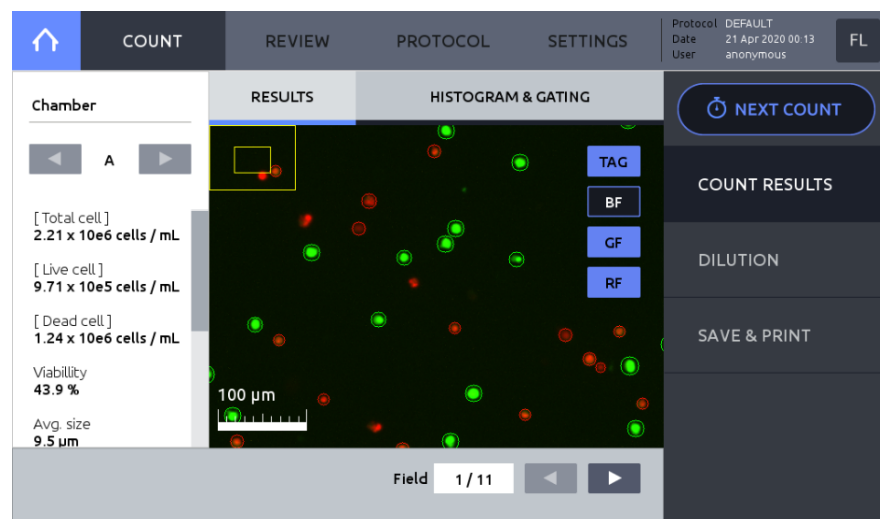
After counting is complete, the data and images will appear in the **RESULTS** window.



The counting results will be shown to the left of the screen.

Press the left or right arrows under **Chamber** to view the results and images for each counted chamber.

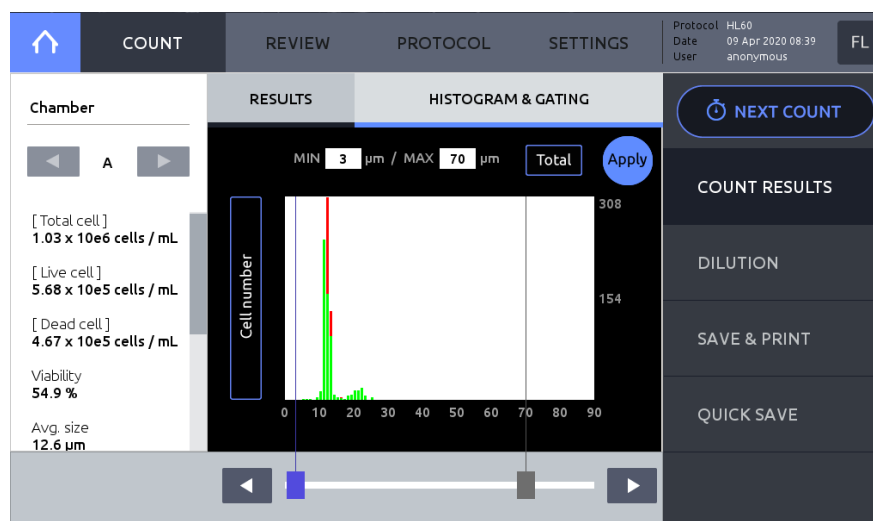
Press **TAG** to identify live (green circles) or dead (red circles) cells.



After fluorescence cell counting, BF, GF, and RF images can be viewed separately or in overlay.

## Histograms

Press **HISTOGRAM & GATING** to open the histogram window.



- ① Cell concentration or number can be graphed according to cell size.
- ② Each histogram for total, live, and dead cells can be displayed.
- ③ Cell size gating parameters may be changed by pressing the slider rectangles. An active slider will be highlighted in blue. Move the sliders by dragging or pressing the arrows.

Press **Apply** to set cell size gating parameters. Counting results will adjust accordingly.

To toggle between cell concentration, cell cluster, and cell number press ① the Y-Axis title.

To switch between total, live, and dead, press ② the title box on the left of **Apply**.

**Dilution Calculator** Press **DILUTION** to open the Dilution Calculator.

The screenshot shows the 'DILUTION' calculator interface. It includes input fields for 'Pre-dilution factor' (1: 10), 'Current Concentration' (1.36 x 10<sup>6</sup> /mL), 'Stock Concentration' (1.36 x 10<sup>7</sup> /mL), 'Desired Concentration' (1 x 10<sup>5</sup> /mL), and 'Final Volume' (10 mL). A 'CALCULATE' button is present. Below the inputs, a text box displays the result: 'Dilute 0.074 mL of cell sample in 9.926 mL of buffer.' On the right, there are buttons for 'NEXT COUNT', 'COUNT RESULTS', 'DILUTION', 'SAVE & PRINT', and 'QUICK SAVE'.

The dilution calculator starts out with the concentration of total cells (live and dead) as the current concentration. The current concentration options are **Total**, **Live**, **Dead**, and **Custom**, allowing users to set the current concentration to be the total cell concentration, live cell concentration, dead cell concentration, or a custom cell concentration by pressing the blue box below the Current Concentration value.

If the cell counting was performed with the diluted sample from the stock cell solution, dilution instruction can be calculated from the stock cell concentration by entering pre-dilution factor.

For example, enter “10” in the pre-dilution factor if the counted sample was 10-fold diluted from stock solution. Enter “1” (default value) if the counted sample was not diluted from stock solution.

Enter the values for the desired concentration and final volume. Press **CALCULATE** and dilution instruction will appear in the grey message box.

## Save

Press **SAVE & PRINT** to open the save window.



Select the desired saving options:

Save Options	File Type	Description
Analyzed image	TIF	Tagged images of cells
Raw image	TIF	Untagged images of cells
Report	PDF	Report with data, images, and histograms
Bioprocess	Onboard graph, .CSV	Growth curve displayed onboard Growth rate, doubling time

Press **Path** to select where files are to be stored.

Using the onscreen keyboard, provide the name and append the date and time by pressing the **+Date/Time** button.

Press **Save**. A folder name will be created with the name provided. The folder will contain subfolders matching each of the counted chambers, e.g. 'Chamber A', or 'Chamber 1'.

## Print

To print a text summary of the counting results, make sure a LUNA-FX7™ Thermal Printer (P17001) is connected to the LUNA-FX7™ and press **Print**.

The printer should be connected before powering on the instrument.

The printed report will contain the cell count results and protocol details.

Cell count report

---

Instrument : LUNA-FX7 Cell Counter  
Serial number : LU7-00-00000  
Software version : 0.0.0  
Firmware version : 0.0.0  
File name : HL60  
Date : 16 Apr 2017 14:35:37  
Security : Off  
User : anonymous  
File name : NoTitle  
Counting mode : Fluorescence cell counting  
Cell lines & Primary cells

---

Instrument setting

---

Slide type : 2 channel slide  
Counted chamber area : A, B  
Autofocused counting : On  
Last calibration : 10 Apr 2017 11:45  
Calibration value : 24 / 36 / 58 / 100 / 74

---

Protocol

---

Protocol name : DEFAULT  
GF exposure level : 5  
RF exposure level : 5  
Cell size calculation: BF  
Min. cell size : 3  
Max. cell size : 70  
GF threshold level : 5  
RF threshold level : 5  
Dilution factor : 1.11  
Size gating: 3 ~ 60 µm

---

Cell count results

---

[Total cell] : 1.04 x 10e6 cells / mL  
[Live cell] : 9.73 x 10e5 cells / mL  
[Dead cell] : 6.62 x 10e4 cells / mL  
Viability : 93.6 %  
Avg. size : 16.5 µm

## Quick Save

Press **QUICK SAVE** to save results with a default name and suffix designation. The appended suffix may be a sequential number or the date/time.

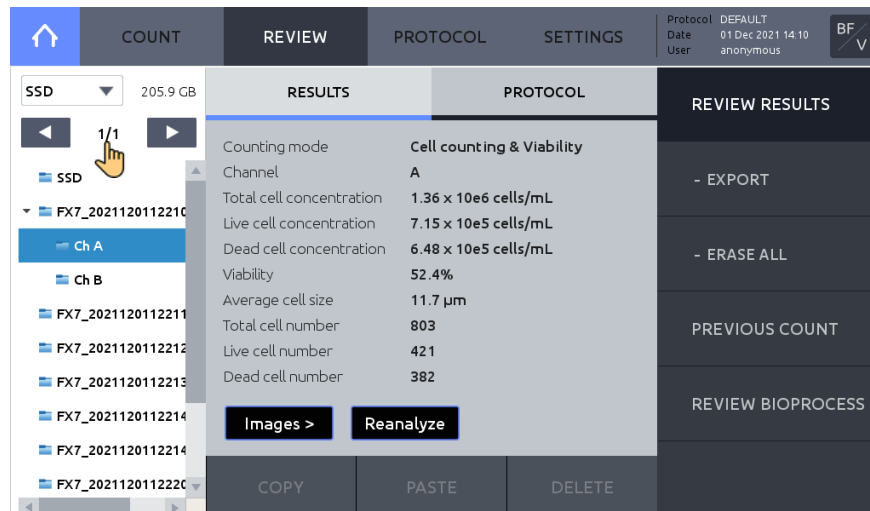
Default Quick save preferences may be pre-set in **SETTINGS: SAVE & REVIEW**.

## 4. Review

### Reviewing Data

#### Review Images

Press **REVIEW**.

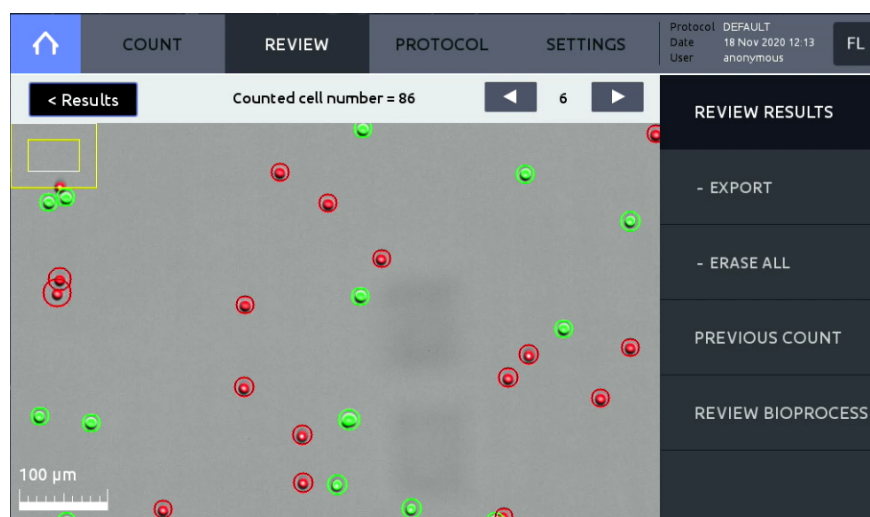


Select SSD or USB drive.

Navigate and open a folder from the internal or a USB drive. Select a subfolder, e.g., Ch A, Ch B. Cell counting results will appear. Max. 200 folders will be displayed per page. Press arrow button to navigate next page. Press page number to go to the specific page directly.

- **EXPORT TO USB:** You can copy the counting data of the user in the counting mode to a USB flash drive connected.
- **ERASE ALL:** You can delete all counting data of the user in the counting mode.

Press **Images >** to see images. Zoom in or out by pinching in or out with two fingers.



Scroll through the captured images using the arrows.

Press the **PROTOCOL** tab to check the protocol used.

To transfer files to a USB drive or delete files from the internal drive, press **< Results** to return to the main **Results** window. Use the command buttons at the bottom of the screen: COPY, PASTE, or DELETE.

## Reanalyze

Raw images may be reanalyzed using a different protocol.

Load or create the desired protocol.

! **Important !** During reanalysis, changing exposure levels in the protocol will have no effect on the results of the reanalyzed counting data.

Press **REVIEW** and select a folder from the internal or USB drive.

Select the subfolder/chamber to be reanalyzed.

Press **REANALYZE**.

## Previous Counts

Press **PREVIOUS COUNT** to see a list of previous counts.

						Protocol: DEFAULT	Date: 18 Nov 2020 12:15	User: anonymous	FL
⏏	COUNT	REVIEW	PROTOCOL	SETTINGS					
User/File	Date/Time	[Total cell]	[Live cell]	[Dead cell]	Viability				
anonymous B	12 Nov 2020 17:57:41	0.00 x 10e0 0	0.00 x 10e0 0	0.00 x 10e0 0	0.0	REVIEW RESULTS			
anonymous A	12 Nov 2020 17:57:41	1.05 x 10e6 1160	5.43 x 10e5 600	5.07 x 10e5 560	51.7	PREVIOUS COUNT			
lab1_lucy.br... B	12 Nov 2020 17:49:01	0.00 x 10e0 0	0.00 x 10e0 0	0.00 x 10e0 0	0.0	- EXPORT			
lab1_lucy.br... A	12 Nov 2020 17:49:01	1.05 x 10e6 1159	5.42 x 10e5 599	5.07 x 10e5 560	51.7	- ERASE ALL			
lab1_james.o... LU7-00-00020...	29 Oct 2020 19:27:40	0.00 x 10e0 0	0.00 x 10e0 0	0.00 x 10e0 0	0.0	REVIEW BIOPROCESS			
lab1_james.o... LU7-00-00020...	29 Oct 2020 19:27:40	1.05 x 10e6 1163	5.42 x 10e5 599	5.10 x 10e5 564	51.5				
lab1_james.o... LU7-00-00020...	26 Oct 2020 21:03:54	0.00 x 10e0 0	0.00 x 10e0 0	0.00 x 10e0 0	0.0				
lab1_james.o... LU7-00-00020...	26 Oct 2020 21:03:54	1.05 x 10e6 1158	5.40 x 10e5 597	5.08 x 10e5 561	51.6				
lab1_lucy.br... LU7-00-00020...	26 Oct 2020 20:42:06	0.00 x 10e0 0	0.00 x 10e0 0	0.00 x 10e0 0	0.0				
lab1_lucy.br... LU7-00-00020...	26 Oct 2020 20:42:06	1.05 x 10e6 1161	5.38 x 10e5 595	5.12 x 10e5 566	51.2				
lab1_james.o... LU7-00-00020...	26 Oct 2020 20:32:32	0.00 x 10e0 0	0.00 x 10e0 0	0.00 x 10e0 0	0.0				

A summarized version of each count that includes User/File, Date/Time, Total cell concentration, Live cell concentration, Dead cell concentration, Viability, Average size, and Protocol is automatically saved to the internal drive.

Live cell concentration, Dead cell concentration and Viability are not available In the Brightfield – Total cell counting mode.

Insert a USB drive and press **EXPORT TO USB** to save as a .CSV file.

Press **ERASE ALL** to delete all stored counts. This will not delete reports or images of the corresponding count, if they were saved to the internal drive.

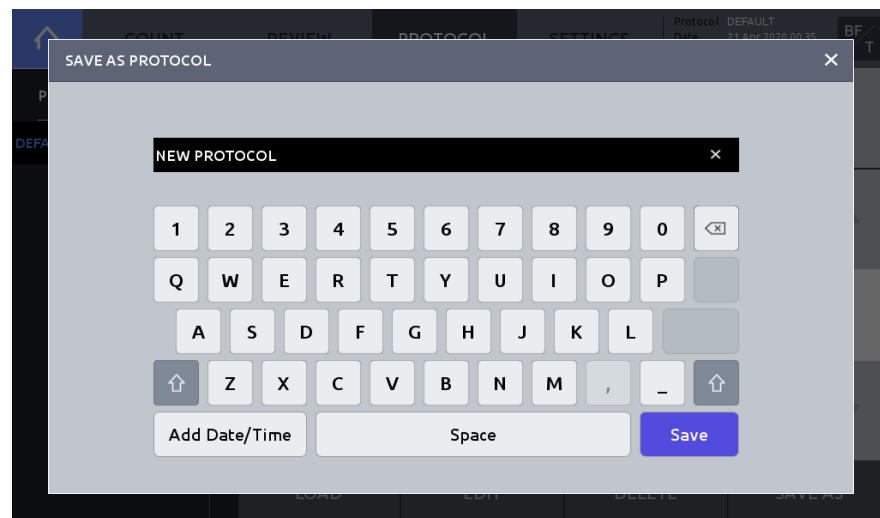
# 5. Protocols

## Protocol Selection

**Default Protocol** Customized protocols for specific cell types may be created.

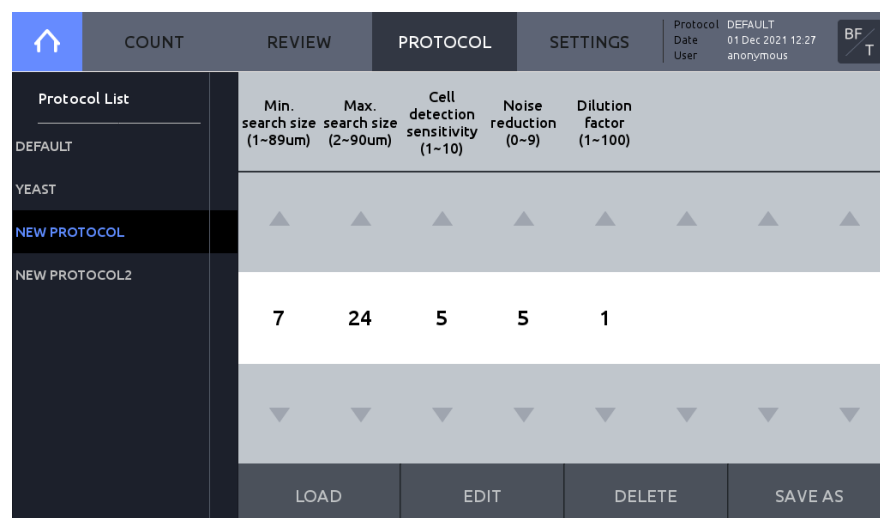
Each counting mode comes with a pre-set Default counting protocol. Bright Field Cell Counting & Viability mode and Fluorescence Cell Counting mode additionally have IQOQ protocols: IQOQ-BF or IQOQ-FL, applied only for quality control and validation purposes. These protocols cannot be edited. The default protocols, by design, will provide optimal results for most cell types, but protocols for specific cell types or applications may need to be optimized.

**Creating Protocols** To create a new protocol, select the DEFAULT protocol and press SAVE AS. Rename the protocol and press Save. The newly created protocol will appear in the list of protocols.



**Editing Protocols** Select a protocol that is not the *Default* protocol.

Press **EDIT**. This will activate the arrows for each parameter, turning them black. Press the arrows to adjust the values of each parameter. Press **SAVE AS** to change the protocol name. Press **Load** to save the edited protocol under the selected name.



**Load Protocol** Select the desired protocol and press LOAD.

The current active protocol name can be seen in the upper right hand corner of the screen.

## Protocol Parameters

## Brightfield Cell Counting Parameters

Parameters for [Brightfield cell counting-Total cell counting]

		COUNT	REVIEW	PROTOCOL	SETTINGS	Protocol Date 01 Dec 2021 12:28 User anonymous		T	
Protocol List									
DEFAULT		Min. search size (1~89um)	Max. search size (2~90um)	Cell detection sensitivity (1~10)	Noise reduction (0~9)	Dilution factor (1~100)			
YEAST									
		▲	▲	▲	▲	▲	▲	▲	▲
		7	24	5	5	1			
		▼	▼	▼	▼	▼	▼	▼	▼
		LOAD		EDIT		DELETE		SAVE AS	

Parameter	Range	DEFAULT*
Min. search size (µm)	1-89	7
Max. search size (µm)	2- 90	24
Cell detection sensitivity	1-10	5
Noise reduction	0-9	5
Dilution factor	1-100	1

Parameters for [Brightfield cell counting-Cell counting &amp; Viability]

[illegible]

Parameter	Range	DEFAULT*
Min. search size (µm)	1-89	7
Max. search size (µm)	2- 90	24
Cell detection sensitivity	1-10	5
Live cell sensitivity	1-10	5
Noise reduction	0-9	5
Dilution factor	1-100	2



The IQOQ-BF on the Protocol List is the protocol that is used for IQOQ with the brightfield validation slide. Protocol YEAST is an optimized protocol that is used for yeast cell counting.

## Min./Max. search size

Search size refers to the approximate cell size that the algorithm recognizes as potential cell objects. By adjusting Min. and Max. search size, objects sized within the setting value will be listed as a potential cell candidate.

Clustered objects larger than the Max. search size will not be excluded from the search. Rather, the algorithm will utilize morphological information to identify individual objects within the cluster that are within the search size parameters. For the most accurate results, it is recommended to set the Min./Max. search size window as narrow as possible to encompass the expected cell size range.

## Cell detection sensitivity

Cell detection sensitivity refers to the sensitivity of object separation from the background. A higher Cell detection sensitivity value will increase detection of signals from weakly stained cells or smaller objects, but can also increase false positive calls.

## Live cell sensitivity

Live cells with intact cell membranes exclude trypan blue and erythrosin B. The dyes stain the cytoplasm of dead cells with compromised membranes. As a result, object intensity of unstained live cells is brighter than the stained dead cells. A higher Live cell sensitivity will decrease the intensity cutoff value and increase the number of live cells detected.

Live Cell Sensitivity is not available in the protocol of the Total cell counting mode.

## Noise reduction

This option allows for the adjustment of background noise during counting. With more noise reduction, the instrument will not detect weakly stained objects. With lower noise reduction, the instrument can detect objects with fainter signals.

## Dilution factor

The dilution factor is used to calculate cell concentrations accurately. The default dilution factor is preset as 1 for Total cell counting and as 2 for Total cell & viability counting (assuming a 1:1 ratio of stain to cell suspension).

This value can be modified according to the dilution of the original sample in increments of 1 between 1-10; and, increments of 10 between 10 -100. For users handling highly dense cell cultures. For highly dense cultures, serial dilutions and several counts with appropriately adjusted dilution factors may be necessary.

## Fluorescence Cell Counting Parameters – Cell lines & Primary cells mode

Parameters for [Fluorescence cell counting-Cell lines & primary cells]

		COUNT	REVIEW	PROTOCOL	SETTINGS	Protocol Date User			DEFAULT 12 Jul 2021 19:37 anonymous	FL C
Protocol List		GF exposure level (0.1~10)	RF exposure level (0.1~10)	Cell size calculation (BF / FL)	Min. cell size (1~89um)	Max. cell size (2~90um)	GF threshold level (1~10)	RF threshold level (1~10)	Dilution factor (1~10)	
DEFAULT										
IQOQ-FL										
		▲	▲	▲	▲	▲	▲	▲	▲	
		5	5	BF	3	70	5	5	1.11	
		▼	▼	▼	▼	▼	▼	▼	▼	
		LOAD	EDIT		DELETE		SAVE AS			

Parameter	Range	DEFAULT*
GF exposure level	0.1-10	5
RF exposure level	0.1-10	5
Cell size calculation	BF/FL	BF
Min. cell size	1-89	3
Max. cell size	2-90	70
GF threshold level	1-10	5
RF threshold level	1-10	5
Dilution factor	1-10	1.11

The IQOQ-FL on the Protocol List is the protocol that is used for IQOQ with the fluorescence validation slide.

! **Important !** The value of total concentration printed on the validation slide label, using the IQOQ-FL may differ from that of using the default protocol because the label value is determined with the IQOQ-FL protocol, which is for the purpose of IQOQ and Quality Control mode.

## GF/RF Exposure Level

The exposure for each channel can be adjusted. Increase exposure if the preview image is dim and only a few cells are visible. Lower exposure if the preview image is too bright and background noise is high. Determine optimal exposure values empirically.

## Cell Size Calculation

A mode between brightfield and fluorescence can be selected. The selected mode is used to measure the cell size.

If FL is selected, the cell size is measured based on the fluorescence signals so the results may change depending on the exposure level.

## Min./Max. cell size

Use these parameters to adjust the minimum and maximum cell sizes to be included in results. The base unit is 1 micrometer.

## GF/RF threshold level

Green and Red fluorescence threshold will determine the level of threshold during the image processing. Increasing the threshold will lead to fewer cells being detected by increasing the background level that is subtracted. Conversely, decreasing the threshold will lead to more cells being detected.

## Dilution Factor

The default dilution factor is pre-set as 1.11 for the standard Acridine Orange and Propidium iodide staining protocol, (e.g. 18  $\mu$ L cells + 2  $\mu$ L AO/PI.)

Assuming a final volume of 20  $\mu$ L, the dilution factor may be adjusted according to the table below:

Dilution factor	1	1.11	1.25	1.44	1.66	2	2.5	3.33	5	10
Sample volume	20 $\mu$ L	18 $\mu$ L	16 $\mu$ L	14 $\mu$ L	12 $\mu$ L	10 $\mu$ L	8 $\mu$ L	6 $\mu$ L	4 $\mu$ L	2 $\mu$ L

Parameters for [Fluorescence cell counting-Cell lines & primary cells, Advanced]

## Fluorescence Cell Counting Parameters – Cell lines & Primary cells, Advanced mode

[illegible]

Parameter	Range	DEFAULT*
GF exposure level	0.1-10	5
RF exposure level	0.1-10	5
Min. search size (µm)	1-89	7
Max. search size (µm)	2- 90	30
Declumping sensitivity	1-10	5
Min. FL intensity	0-10	0
Min. roundness	0-9	3
Dilution factor	1-10	1.11

The IQOQ-FL-A on the Protocol List is the protocol that is used for IQOQ with the fluorescence validation slide.

**! Important !** The value of total concentration printed on the validation slide label, using the IQOQ-FL-A may differ from that of using the default protocol because the label value is determined with the IQOQ-FL-A protocol, which is for the purpose of IQOQ and Quality Control mode.

!

Protocol YEAST is an optimized protocol that is used for yeast cell counting.

## GF/RF Exposure Level

The exposure for each channel can be adjusted. Increase exposure if the preview image is dim and only a few cells are visible. Lower exposure if the preview image is too bright and background noise is high. Determine optimal exposure values empirically.

## Min./Max. search Size

Search size refers to the approximate cell size that the algorithm recognizes as potential cell objects. By adjusting Min. and Max. search size, objects sized within the setting value will be listed as a potential cell candidate.

Clustered objects larger than the Max. search size will not be excluded from the search. Rather, the algorithm will utilize morphological information to identify individual objects within the cluster that are within the search size parameters. For the most accurate results, it is recommended to set the Min./Max. search size window as narrow as possible to encompass the expected cell size range.

## Declumping sensitivity

Declumping sensitivity refers to the sensitivity of cell separation from the cluster of cells. Higher levels of declumping sensitivity will lead to the detection of more cells in a cluster, but may recognize internal cell structure as separate objects.

**Min. FL intensity**

Min FL intensity is used to set the minimum Green and Red fluorescence intensity values to be detected as cells. Objects with fluorescence intensity lower than Min. FL intensity value are excluded from counting. Increased Min. FL intensity value will remove objects with weak fluorescence intensity. Conversely, decreased Min FL. Intensity value will detect more objects with weak fluorescence intensity.

**Min. roundness**

Roundness is the measure of how closely the shape of a cell approaches mathematically perfect circle. Roundness is expressed as a value between 1 and 0, Roundness = 1 for a perfect circle and Roundness = 0 for a line segment. Higher value will lead to the counting of rounder cells and excludes objects with less roundness. Lower values are suitable for counting cells with irregular shapes.

**Dilution Factor**

The default dilution factor is pre-set as 1.11 for the standard Acridine Orange and Propidium iodide staining protocol, (e.g. 18  $\mu$ L cells + 2  $\mu$ L AO/PI.)

Assuming a final volume of 20  $\mu$ L, the dilution factor may be adjusted according to the table below:

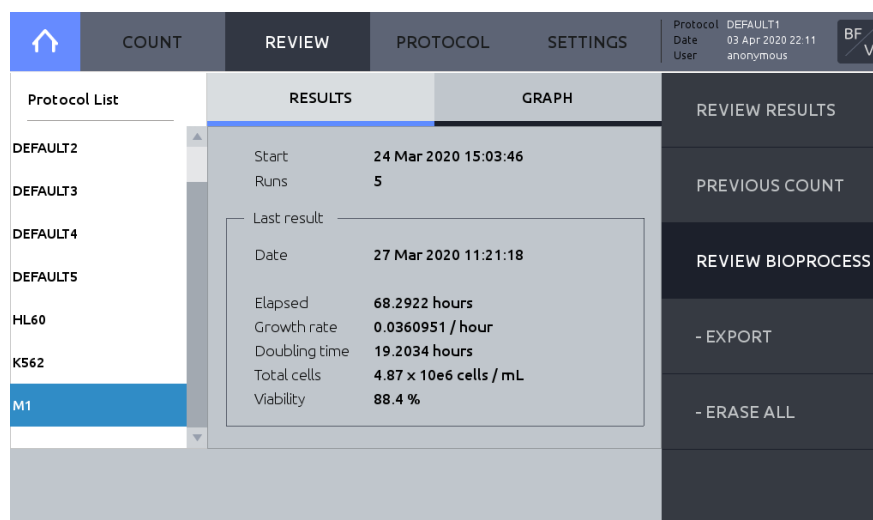
Dilution factor	1	1.11	1.25	1.44	1.66	2	2.5	3.33	5	10
Sample volume	20 $\mu$ L	18 $\mu$ L	16 $\mu$ L	14 $\mu$ L	12 $\mu$ L	10 $\mu$ L	8 $\mu$ L	6 $\mu$ L	4 $\mu$ L	2 $\mu$ L

## 6. Bioprocess Feature

### Bioprocess Feature

#### Bioprocess

The LUNA-FX7™ bioprocess feature enables automated tracking of multiple bioprocessing activities. The bioprocess feature tracks individual batches according to protocol and will calculate and chart growth rates, doubling times, and viabilities based on count data.



#### Creating a Bioprocess Protocol

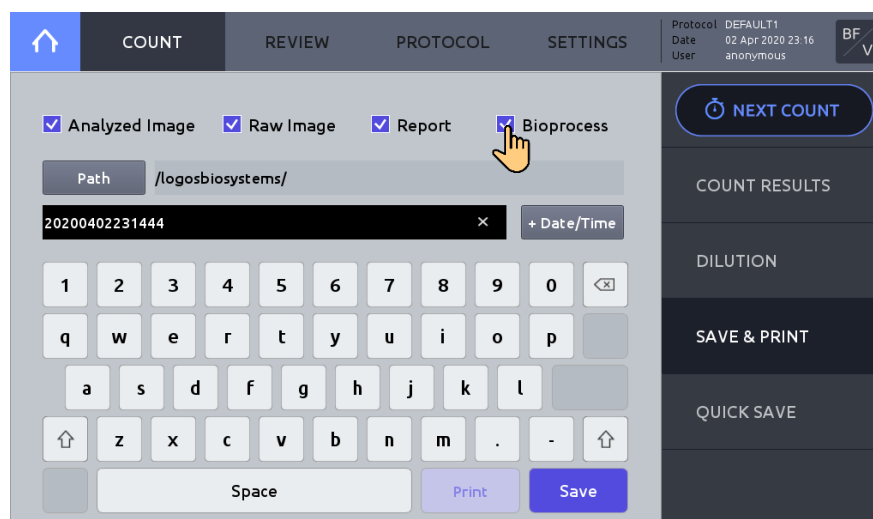
1. To create a new bioprocess, press **PROTOCOL** and create a new protocol.
2. Edit and save the new protocol as needed.

#### Record Bioprocess Data

1. To record bioprocess data, go to **SETTINGS** within a counting mode. Set Counting chamber area is set to 'Current'. If the Counting chamber area is not set to 'Current', bioprocessing data will not be saved.
2. Press **PROTOCOL** and load the appropriate protocol.

When you record bioprocess data of the same cell, make sure that the same protocol you have used is loaded before you count.

3. Press the **COUNT** button.
4. Press **SAVE & PRINT**.
5. Select either 'Analyzed Image' or 'Raw Image' AND select 'Bioprocess'.



## Review/Export Bioprocess Data

1. Select **REVIEW**.
2. Press **REVIEW BIOPROCESSES**.
3. From the protocol list on the left, select the protocol used to create your bioprocess data.
4. To export bioprocess data as a .CSV file, press – **EXPORT TO USB**.
5. To delete selected bioprocessing data, press **ERASE ALL**. The data, but not the protocol will be deleted.

## Graph

Press **GRAPH** to view charted results.



Press the Y-axis title box to alternate between 'Total cell concentration', 'Live cell concentration', and 'Viability'.

Press 'Day', 'Month' or 'Year' to alter X-axis scale.

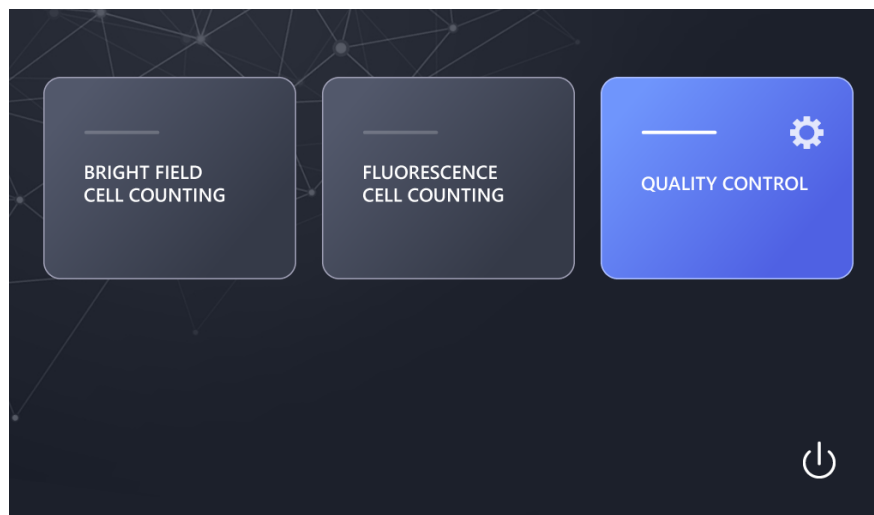
The bioprocess data is automatically saved to the internal drive and linked to the loaded protocol name.

## 7. Quality Control

### Quality Control Mode

#### Quality Control

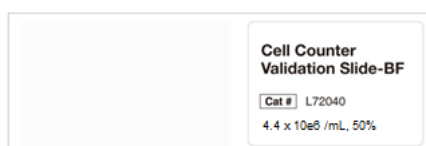
**Quality Control** mode is used to monitor the performance of the LUNA-FX7™. The Quality Control features may only be used in conjunction with the Logos Biosystems fluorescence or brightfield validation slides. Validation slides contain a pre-spotted pattern (brightfield) or pre-fixed beads (fluorescence) of known concentration and viability.



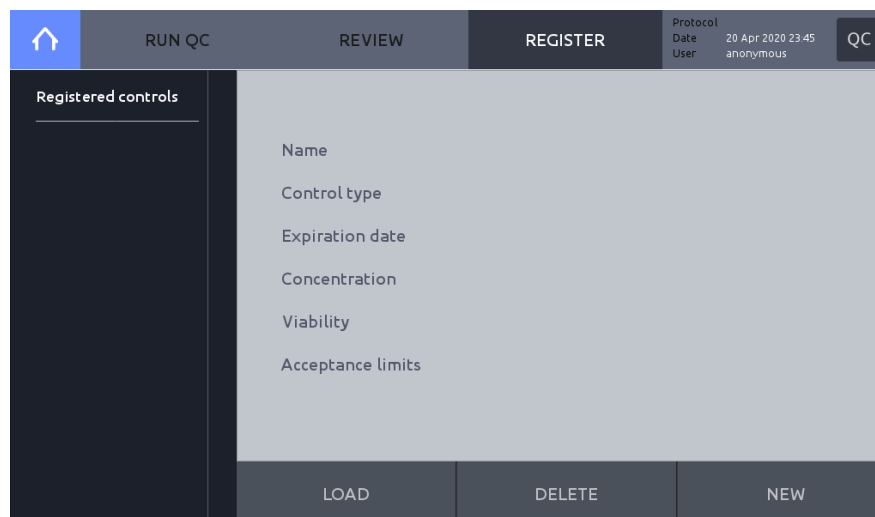
#### Validation Slide

Prior to use, validation slides *must* be registered.

#### Registration

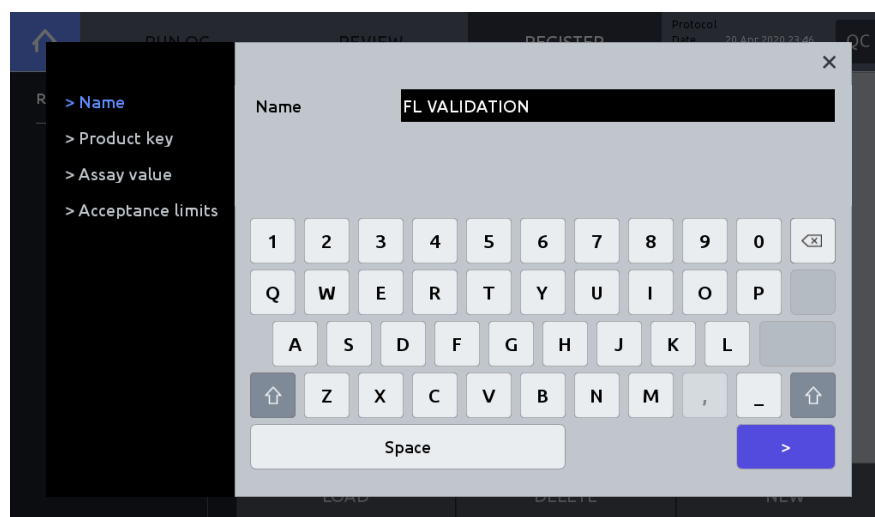


Press **Quality Control**, press **REGISTER**,



Press **NEW**.

Enter a **Name**, then press the **right arrow key [>]** to move to 'Product key'.



Enter the 20 digit **Product key**. Press the 'space' icon to advance. Press **OK**.

Confirm control type: Fluorescence or Brightfield and Expiration date.

- ! **Important !** The product key is included with the product information of the validation slide. Contact sales@logosbio.com if the product key is lost or missing.
- ! **Important !** The validation slides have a limited lifetime and the reliability of a validation slide can no longer be guaranteed. Therefore, to ensure the accuracy and reliability of the QC data, it is recommended that replacement validation slides are purchased prior their stated expiration.

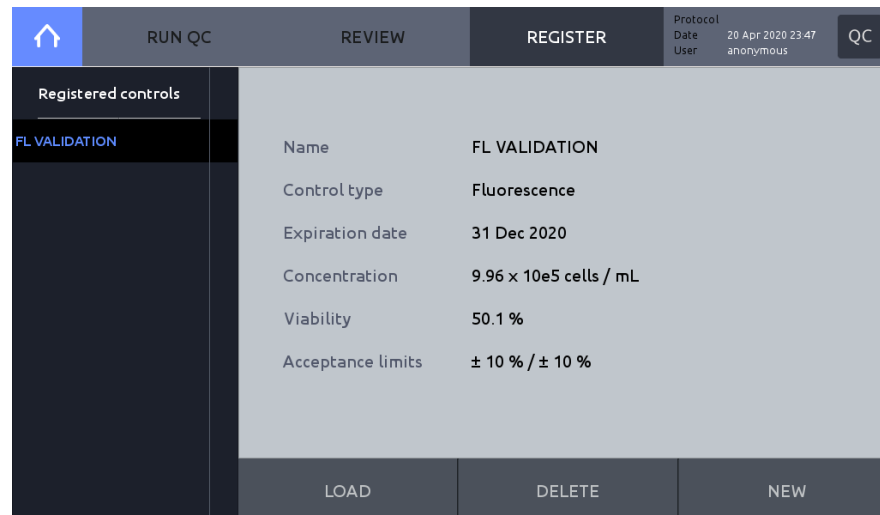


Press the **right arrow key [>]** to move to 'Assay value'.

Confirm that the Assay value is correct. If the Assay value differs from what was provided with the validation slide, check to ensure the product key was entered correctly. If entered, correctly, contact [sales@logosbio.com](mailto:sales@logosbio.com).

Set Acceptance limits (%). Acceptance limits produce upper and lower boundaries in QC graphing. Press **Save** to complete registration.

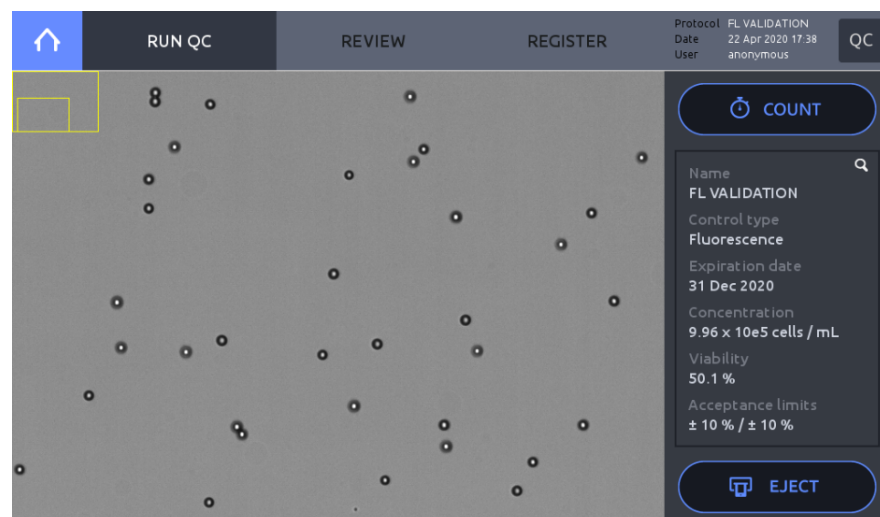
After completing registration, the validation slide information may be viewed by selecting the appropriate registered control in the **REGISTER** main page, and pressing **Load**.



## Performing Quality Control

Navigate to **Quality Control** mode and press **REGISTER**. Select a validation slide from the list of Registered controls. Press **LOAD**.

Press **RUN QC** in the main **Quality Control** screen, insert the validation slide, and press the **COUNT** button.

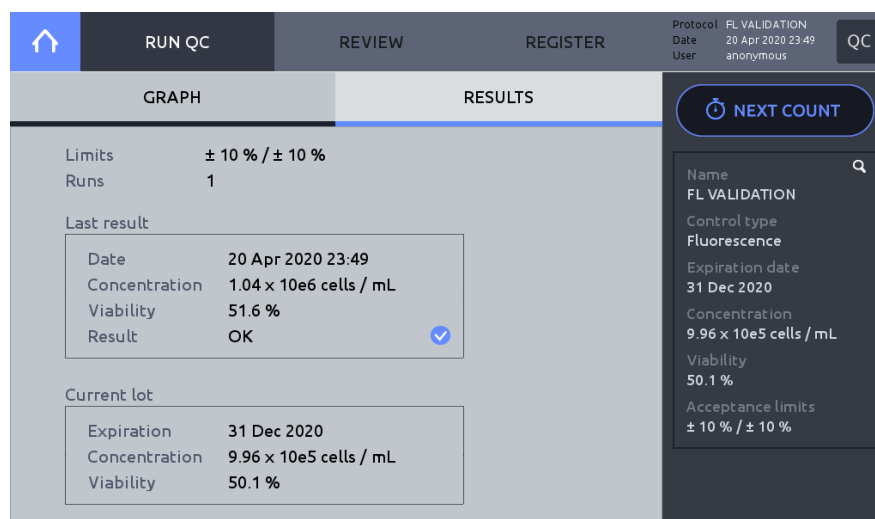


After counting, the QC graph will appear. Confirm that the measured value is within the acceptance limits established during slide registration.

Press **RESULTS** to see the counting data.

If the results are not within the acceptance range, redo RUN QC steps.

If not met again, contact your local distributor or Logos Biosystems.



To re-run QC, press **NEXT COUNT**, then press the **COUNT** button.

## Review

Press **REVIEW**.

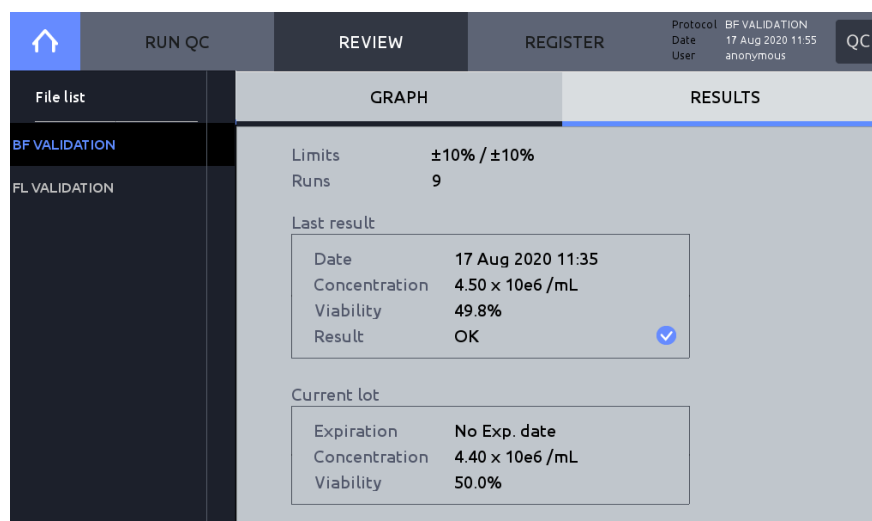
Select a validation slide from the File list on the left.



Press **GRAPH** to view a graphical representation. Press the Y-axis title box to switch between 'Concentration' and 'Viability'. Press 'Day', 'Month' or 'Year' to alter X-axis scale.

Press **EXPORT** to export a .CSV file with count data and graph images to a USB drive.

Press **RESULTS** to view the most recent QC count.



- ! **Important !** The value of total concentration printed on the slide label may differ from the counting result of using the default protocol because the label value is determined with a different protocol, which is for the purpose of IQOQ and Quality Control mode.
- ! **Important !** After the Quality Control counting is done, remove the validation slide from the device right away and store it in the case provided. The fluorescence pre-fixed beads may be photobleaching if they are exposed to light.

## 8. Settings

### LUNA-FX7™ Settings

#### Screen Settings

Navigate to either Brightfield or Fluorescence counting window and press **SETTINGS**. Within **SETTINGS**, software updates, date and time changes, and background calibrations may be performed. Additionally, options for 'Cell Counting', 'Save & Review', 'Network', 'CountWire' (only with CountWire™ package) and options may be adjusted.

#### Cell Counting

Choose appropriate slide format and the chamber(s) to be counted.

The image displays two screenshots of the LUNA-FX7 Settings application, specifically the 'CELL COUNTING' section. Both screenshots show a top navigation bar with 'COUNT', 'REVIEW', 'PROTOCOL', and 'SETTINGS' tabs, and a top status bar with 'Protocol: DEFAULT', 'Date: 07 Dec 2021 14:31', 'User: anonymous', and a 'BF/T' toggle.

**Top Screenshot:** The 'Slide setting' section has '1 Ch.' selected. The 'Counting chamber area' contains a single 'current' button. The 'Autofocused counting' and 'Autofocus upon slide insertion' buttons are both set to 'ON'.

**Bottom Screenshot:** The 'Slide setting' section has '2 Ch.' selected. The 'Counting chamber area' contains four buttons: 'current', 'all', 'A', and 'B'. The 'Autofocused counting' and 'Autofocus upon slide insertion' buttons are both set to 'ON'.

HOME

COUNT

REVIEW

PROTOCOL

SETTINGS

Protocol

DEFAULT

Date

07 Dec 2021 14:31

User

anonymous

BF

T

Slide setting

☐ 1 Ch.

☐ 2 Ch.

☒ 3 Ch.

☐ 8 Ch.

☐ Reusable

Counting chamber area

current

all

A

B

C

Combine results of selected chambers

OFF

Autofocused counting

ON

Autofocus upon slide insertion

ON

CELL COUNTING

SAVE & REVIEW

NETWORK

COUNTWIRE

DATE & POWER

S/W & CALIBRATION

⌂

COUNT

REVIEW

PROTOCOL

SETTINGS

Protocol  
Date  
User

DEFAULT  
07 Dec 2021 14:31  
anonymous

BF  
T

Slide setting

☐ 1 Ch.

☐ 2 Ch.

☐ 3 Ch.

☒ 8 Ch.

☐ Reusable

Counting chamber area

current

all

1

2

3

4

5

6

7

8

Autofocused counting

ON

Autofocus upon slide insertion

ON

CELL COUNTING

SAVE & REVIEW

NETWORK

COUNTWIRE

DATE & POWER

S/W & CALIBRATION

⌂

COUNT

REVIEW

PROTOCOL

SETTINGS

Protocol

DEFAULT

Date

07 Dec 2021 14:31

User

anonymous

BF

T

Slide setting

☐ 1 Ch.

☐ 2 Ch.

☐ 3 Ch.

☐ 8 Ch.

☒ Reusable

Counting chamber area

current

Autofocused counting

ON

Autofocus upon slide insertion

ON

CELL COUNTING

SAVE & REVIEW

NETWORK

COUNTWIRE

DATE & POWER

S/W & CALIBRATION

## Slide setting

Select a slide format and Counting chamber area option.

Counting chamber area:

- **Current:** Counts the chamber that is being viewed in the live view of the Count screen.

- **All:** All chambers are counted.
- **Chamber designation:** One or more chambers may be selected.

When the 3 Ch. slide option is selected, results of all chambers may be combined. If [Combine results of selected chambers] is ON, the Counting chamber is automatically set to [all]. When combining results, the PDF report and .CSV file will contain data from all three chambers.

### Staining options

In settings for

Option	Description
With Trypan blue	This option is used when cell samples are mixed with Logos Biosystems' Trypan Blue Stain, 0.4% (T13001) or Erythrosin B Stain (L13002). Adjust the dilution factor in the protocol as appropriate.
With Custom staining	This option is used when cell samples are mixed with all other commercial or lab-made vital stains. <b><u>Background calibration must be performed when switching between different brands or compositions.</u></b>

### Autofocused counting

Select On or Off for the LUNA-FX7™ to readjust the focus for each field of view during image capture (recommended to keep 'On'.)

### Autofocus upon slide insertion

Select On or Off to autofocus when the slide is inserted.

## Save & Review

Press **Save & review** on the right menu.

The screenshot displays the 'SETTINGS' menu of the LUNA-FX7 software. The top navigation bar includes 'COUNT', 'REVIEW', 'PROTOCOL', and 'SETTINGS'. The 'SETTINGS' menu is open, showing various configuration options. On the right, a vertical sidebar contains a menu with the following items: 'CELL COUNTING', 'SAVE & REVIEW', 'NETWORK', 'COUNTWIRE', 'DATE & POWER', and 'S/W & CALIBRATION'. The 'SAVE & REVIEW' option is highlighted. The main settings area shows 'Auto save' as OFF, 'Quick & Auto save rule' with 'Name' as FX7, 'Suffix' as Sequence 1, and 'Next name' as FX7\_00001. 'Scale bar' is also OFF. Under 'Customized reports', there is a section for using a custom logo in reports (160 x 160 pixels, PNG format) with 'Load' and 'Default' buttons.

### Auto save

Select ON or OFF to activate Auto save function. When Auto save is activated, cell counting results are automatically saved according to the Quick & Auto save rule.

### Quick & Auto save rule

- **Name**

This name will serve as the prefix for all saved counts.

- **Suffix**

Select **Sequence** to automatically add sequential numbers to the prefix name; OR, select **Date/Time** to automatically append date and time to the prefix name.

- **Next name**

Displays file name to be used for the next count to be saved.

### Scale bar

Includes or excludes scale bar for Tag (Analyzed) images.

### Customized reports

Allows PDF reports to be customized with preferred logo or image. Required image format: 160 x 160 pixels and PNG format.

## Network

The LUNA-FX7™ may be connected to a local network via Ethernet cable or WiFi.

Within **SETTINGS**, press **NETWORK**.

### Ethernet connection

Connect an Ethernet cable to the instrument.

When connected, an IP address will appear on the screen in blue color.

### WiFi connection

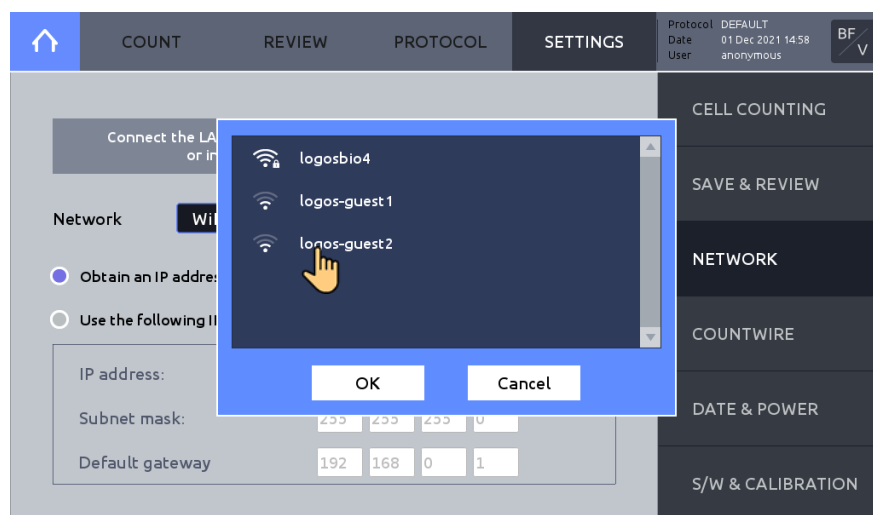
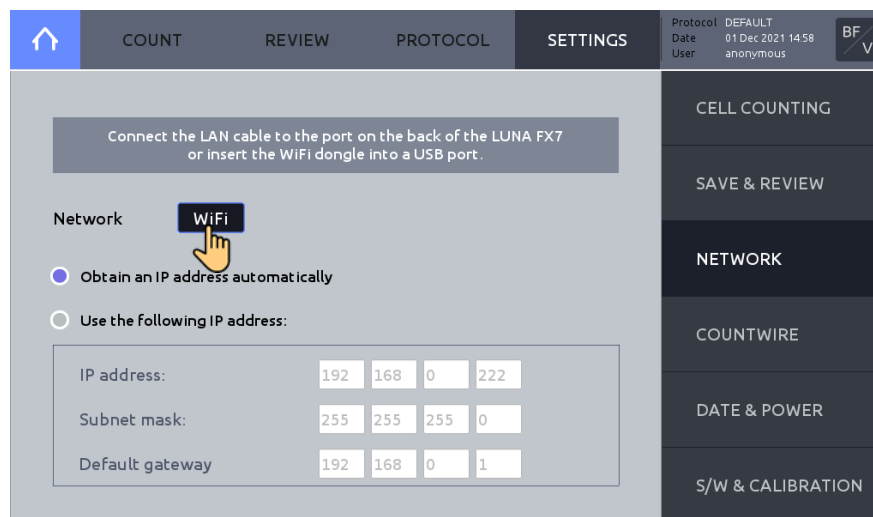
Insert the supplied WiFi dongle to a LUNA-FX7™ USB port.

Press **WiFi**.

Select appropriate WiFi, then press **OK**. Enter password, if necessary.

When the instrument is connected, an IP address will appear on the screen in blue color.

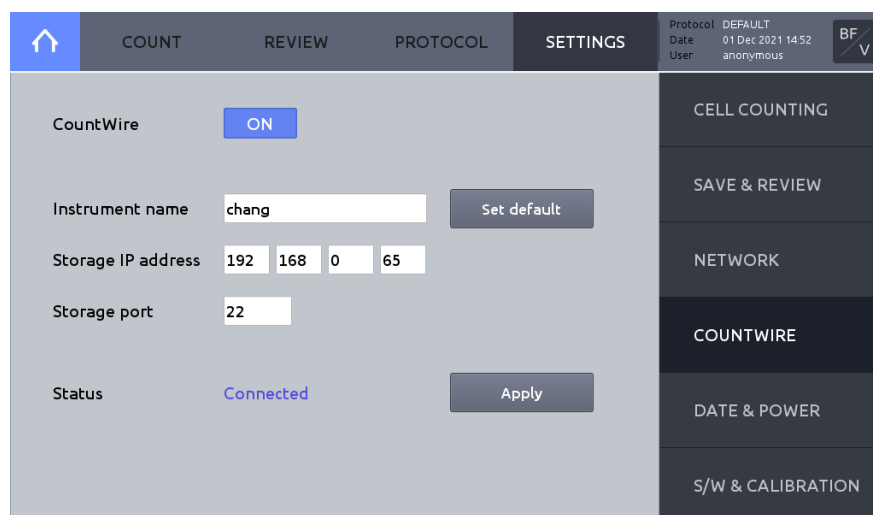




## CountWire

This setting is required to use the CountWire™ system.

Press **COUNTWIRE**.



## CountWire

It must be ON to utilize the LUNA-FX7™ as a part of the CountWire™ system.

! **Important !** If CountWire is on, data transfer via network by FTP freeware (FileZilla FTP Client) does not work.

### Instrument name

Required to distinguish the instruments in the CountWire™ system.

The default instrument name is the serial number, but the name may be changed.

Press **Set default** to initialize the instrument name to the serial number.

### Storage IP address

IP address of the CountWire™ Data Storage.

The same address that you input on the CountWire™ Client.

Ask network administrator for details.

### Storage port

Port number of the CountWire™ Data Storage. The Storage's port number is 22.

Ask network administrator for details.

Press the **Apply** button after entering the required information. If it is successfully connected, you can see the status "Connected".

For more details for the CountWire™, please refer to the CountWire™ User Manual.

## Date & Power

### Date & Time

The LUNA-FX7™ uses a 24-hour clock that is preset to Korean Standard Time. Adjust the settings to the local date and time.

Press **DATE & POWER**.

Press **Set Date & Time**. Input the desired values. Press **APPLY** to save changes.

The screenshot shows the 'SETTINGS' menu of the LUNA-FX7™. The top navigation bar includes 'COUNT', 'REVIEW', 'PROTOCOL', and 'SETTINGS'. The 'SETTINGS' menu is open, showing a sidebar with options: 'CELL COUNTING', 'SAVE & REVIEW', 'NETWORK', 'COUNTWIRE', 'DATE & POWER' (selected), and 'S/W & CALIBRATION'. The main content area displays the 'Date & Time' settings, showing 'Date' as '01 Dec 2021' and 'Time' as '15:20'. A 'Set Date & Time' button is present. Below this, the 'Power saver' section shows a toggle switch set to 'ON' and a text field indicating 'Turn off the instrument after 8 hours (1 ~ 24 hours)'.

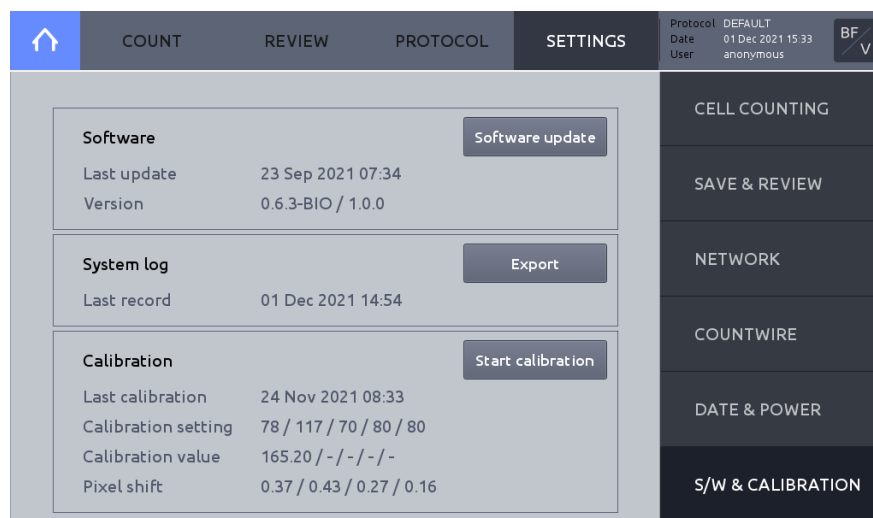
### Power saver

LUNA-FX7™ provides a **Power saver** to save energy and protect environment. Activate Power saver to automatically shut down the LUNA-FX7™.

## S/W & Calibration Software

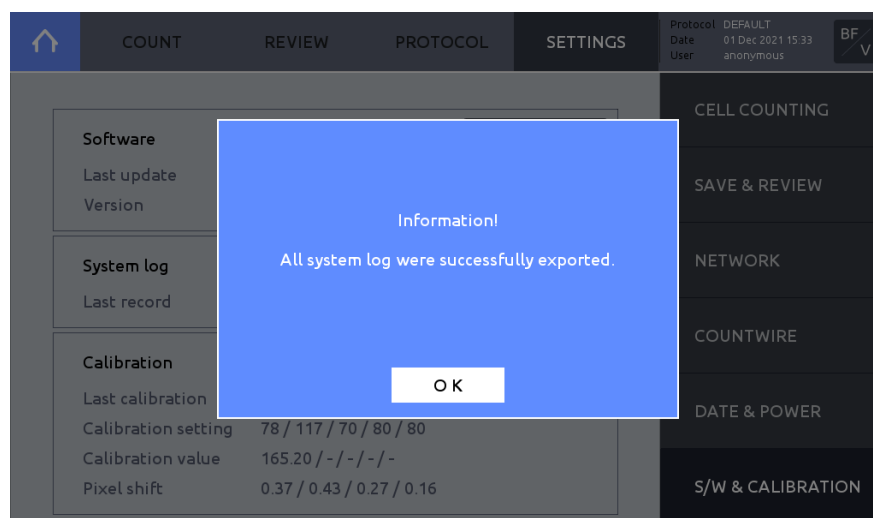
Logos Biosystems continually provides software updates to ensure optimal performance. The current software version is displayed in **SETTINGS: S/W & CALIBRATION**.

1. The most recent version may be downloaded from the Logos Biosystems website ([www.logosbio.com](http://www.logosbio.com)) into the root directory of a compatible USB drive.
2. Press Software in the SETTINGS screen.
3. Insert the USB drive with the downloaded file and authentication key into a USB port.
4. Press **Software update**.
5. Press **Start**. If a software update has been found, press **OK**.
6. Press Restart, then the instrument will automatically shut down and then restart.
7. Prior to the next count, perform calibration.



### System log

The LUNA-FX7™ records the system log for quick diagnosis and service of the instrument. Recorded system log file can be exported to a USB drive. Submit the exported system log file to the authorized distributors or sales representatives for a faster service of an abnormal instrument.

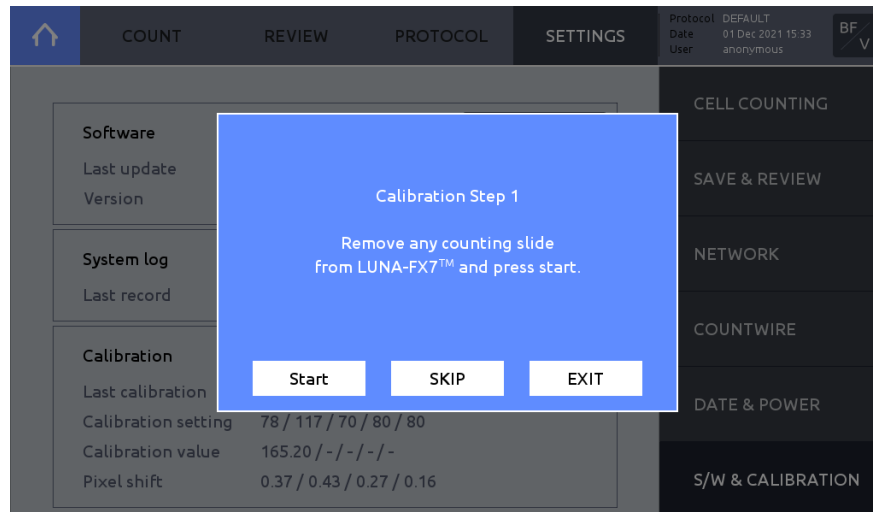


## Calibration

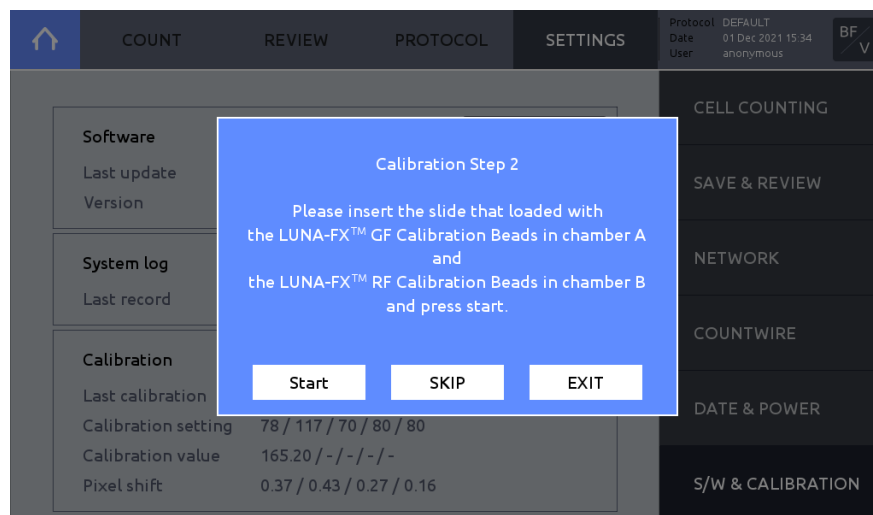
The LUNA-FX7™ is calibrated prior to shipping. Calibration only needs to be performed 1) after any software/firmware updates, and 2) after switching trypan blue or erythrosin B brands or formulations.

To perform calibration:

1. Press **S/W & CALIBRATION**.
2. Press **Start calibration**. The Calibration Step 1 window will appear.



3. Remove any counting slide from slide port.
4. Press **START**.
5. Press **START** or press **SKIP** if only fluorescence calibration is needed.
6. The Calibration Step 2 window will appear upon completion.



7. Load 10 µL of the LUNA-FX™ GF Calibration Beads into chamber A and 10 µL of the LUNA-FX™ RF Calibration Beads into chamber B of a new PhotonSlide™. Mix the calibration beads solution thoroughly before loading into the PhotonSlide™.
8. Wait for 30 seconds for the beads to settle down.
9. Insert the slide face up and chamber A first into the slide port.
10. Press **START**; or, press **SKIP**, if fluorescence calibration is not needed.
11. Upon completion, the calibration value(s) and calibration date will be updated.

## 9. Data Transfer via Network

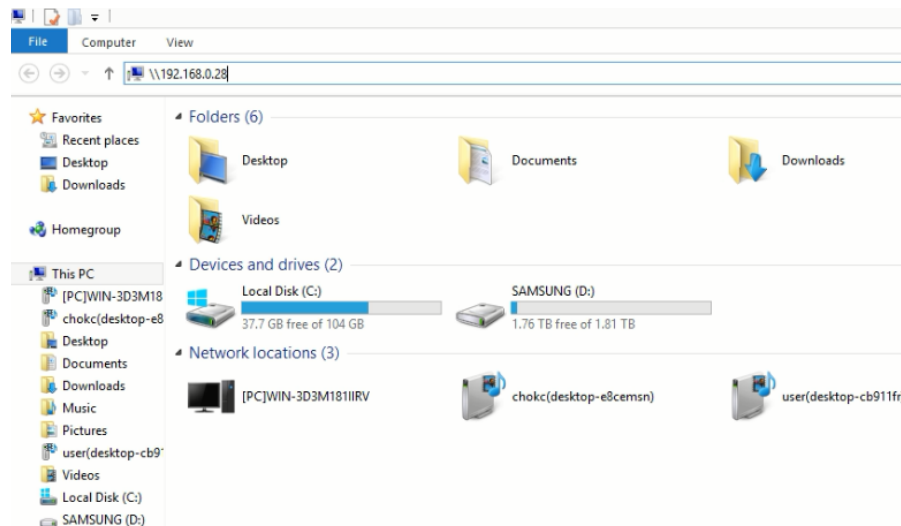
### Network sharing

**Connected to User PC** Connect the LUNA-FX7™ to a network. Make note of the LUNA-FX7™ IP address in the SETTINGS: NETWORK screen.

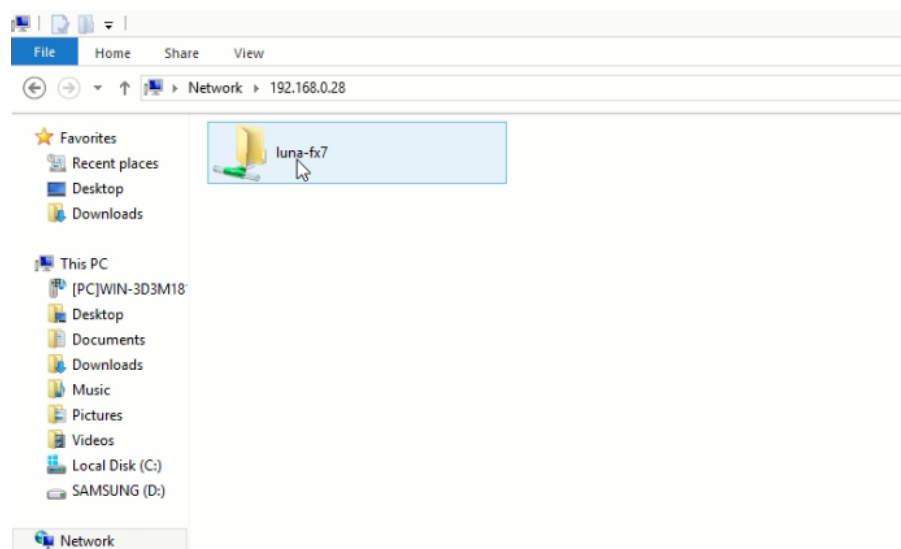
In your Windows PC, open File Explorer (Windows key + E)

Type the IP address connected to the LUNA-FX7™ in the location directory and press Enter.

e.g. \\192.168.0.28



You can double-click and open the **luna-fx7** folder.

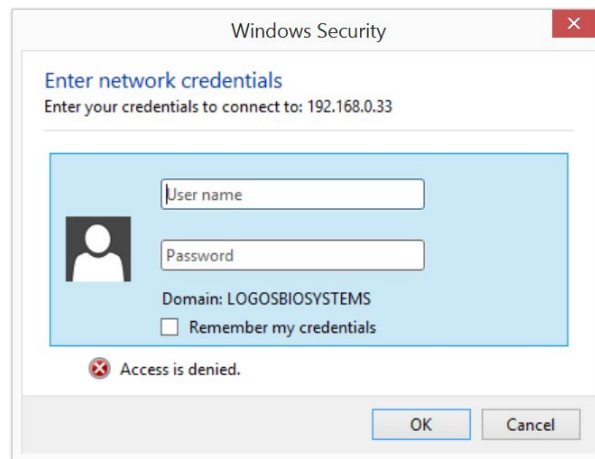


The data in the folder is not stored in the PC. You may move data from the luna-fx7 folder to another drive in the PC.

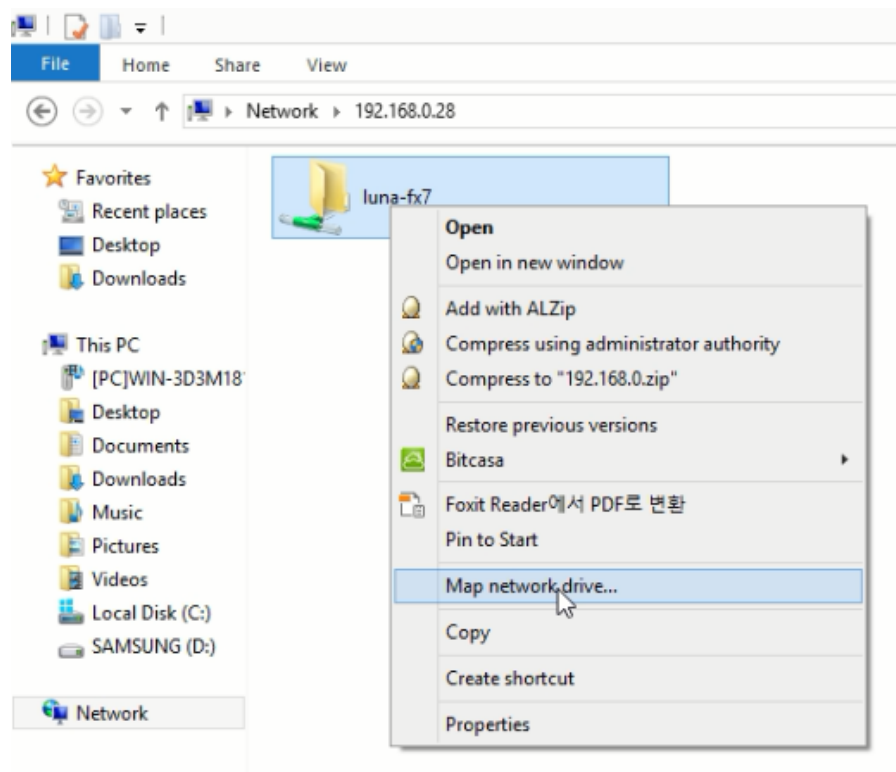
If it is the initial access, it may require a log-in with User name and Password.

- User name: logosbio

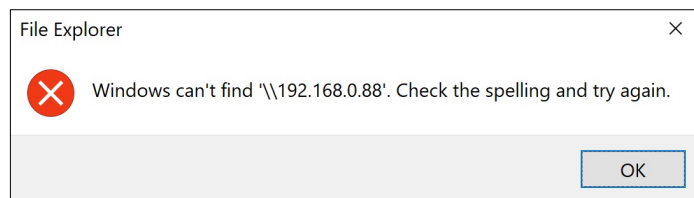
- Password: logosbio



You can right-click the folder to map the network drive or create shortcut for your convenience.



! **Important !** If you cannot access the folder with the message below,



- 1) Please check if the IP address is correct and the LUNA-FX7™ is well connected to the internet.

- 2) If there is no connection issue, contact Logos Biosystems or your local distributor to receive a script in a Zip file to clean Windows authentication caches.

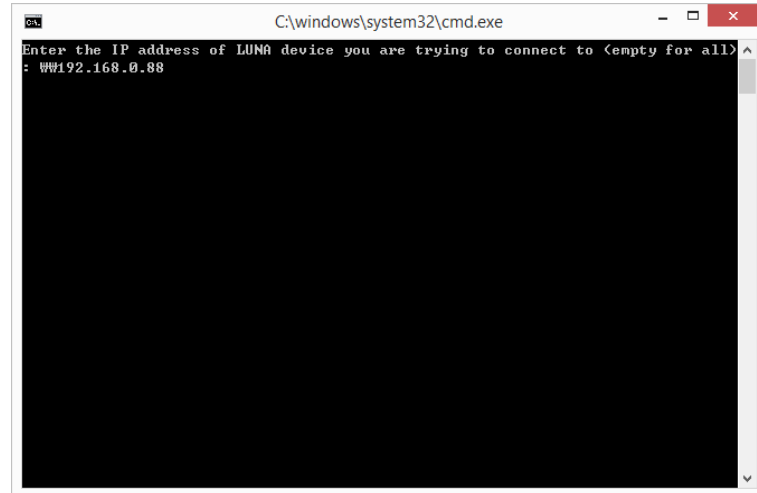
- Unzip the received file and run the command file.

- Type the IP address and press the Enter key.

e.g. 192.168.0.88

- Press any key.

- Try to go through from the beginning.



## FTP freeware

**FileZilla FTP Client** Download and install the **FileZilla FTP Client** from *filezilla-project.org* to a compatible computer.

Connect the LUNA-FX7™ to a network. Make note of the LUNA-FX7™ IP address in the SETTINGS: NETWORK screen.

Open the FileZilla FTP Client on the computer.

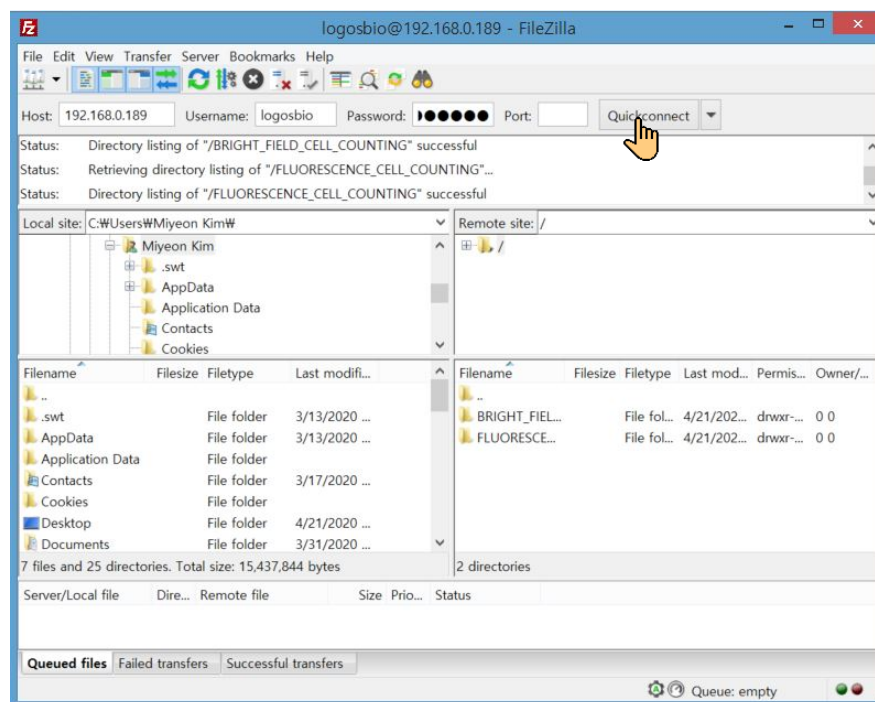
To connect, enter the following information:

- Host: IP address being used for the LUNA-FX7™. Type sftp:// and IP address.

Example: **sftp://192.168.0.189**

- Username: logosbio
- Password: logosbio

Click **Quickconnect**.



Folders/files saved to the LUNA-FX7™ will show up in the Remote site section.

Select the desired folders/files and right-click and select **Download** to import them to your computer.

For more details on using FileZilla FTP Client, visit the website ([filezilla-project.org](http://filezilla-project.org)).



# 10. Maintenance and Troubleshooting

## Maintenance

### Powering on/off

To turn on the LUNA-FX7™, push the power button below the touchscreen for at least a second.

To turn off the LUNA-FX7™, press the power icon in the menu bar or push the power button for at least three seconds. Turn off the LUNA-FX7™ at the end of each day.

### Cleaning

#### Safety

Turn the LUNA-FX7™ off and disconnect the power cable before cleaning. Make sure that liquids do not enter any part of the instrument during cleaning. Do not use abrasive cloths or bleach solutions as this can cause topical damage.

#### Surfaces

Clean the surfaces of the instrument with a soft cloth dampened with distilled water. Wipe dry immediately. Do not pour or spray liquids directly onto the instrument. Do not wet electrical wires or connections in order to avoid electrical shock or damage.

#### Touchscreen

Clean the touchscreen with a soft cloth lightly dampened with an authorized LCD cleansing detergent. Wipe dry immediately. Do not exert excessive force or pressure as this can damage the touchscreen.

## Troubleshooting

### Inaccurate Cell Count

#### Clumped cells

Gently but thoroughly pipette your cell suspension to break up aggregates prior to counting.

#### Too few or too many cells

Cell concentrations of  $5 \times 10^4$  to  $2 \times 10^7$  cells/mL are optimal for counting.

Dilute or concentrate cell suspensions accordingly.

#### Fluorescence signal too strong or weak

Adjust exposure level.

#### Visible cells uncounted

Adjust the protocol's detection sensitivity.

#### Poorly focused images

Make sure the first field is in focus before starting the count. This serves as a reference for the autofocus function.

#### Improper slide insertion

Make sure that the slide has been pushed completely to the end of the slide port.

#### Improper sample loading

Do not over- or under-fill the slide chambers.

**Optical components malfunctioning**

Optical components may be dirty or damaged.

Please contact your local distributor or Logos Biosystems.

**Damaged or contaminated slide**

Use a new slide if it is disposable.

Make sure that the counting area of the slide is transparent before loading the sample.

Wear gloves and handle by the edges to avoid smudging and contamination.

**Incorrect dilution factor**

Adjust the dilution factor in the selected protocol or create a new protocol.

Make sure the appropriate dilution factor has been selected.

**Slide Insertion****Not complete close of the slide port**

If the 8-channel slide is selected in SETTINGS: CELL COUNTING, but other types of slides are inserted, the slide port is not completely closed.

Select the right slide type in SETTINGS: CELL COUNTING, and insert the slide.

**Data Transfer and  
Saving****Incompatible USB drive**

Some USB devices are undetectable or incompatible. Use the USB supplied with the instrument or use a USB 2.0.

**Failed wireless connection**

Check that the WiFi dongle is connected to the LUNA-FX7™. Check that the LUNA-FX7™ is connected to a wireless network. Check that PC is connected to the same wireless network as the LUNA-FX7™. Check your wireless network connection.

**Failed Ethernet connection**

Ensure the Ethernet cable is connected to the LUNA-FX7™ and restart the LUNA-FX7™.

**Software Update  
Errors****Freezing during background calibration**

If calibration takes more than 10 minutes, reset the system by turning the power off and then on. If calibration fails repeatedly, contact your local distributor or Logos Biosystems.

**More than one software version on the USB drive**

Delete previous versions of software from the USB drive before downloading new software.

**Incompatible USB drive**

Some USB devices are undetectable or incompatible. Use the USB drive supplied with the instrument or use a USB 2.0.

**Incorrectly saved or damaged software**

Download the file again into the root directory of the USB drive. Insert the USB drive and press **Software update** in the SETTINGS: SOFTWARE. If the problem persists, contact your local distributor or Logos Biosystems.

# 11. Product Specifications






## LUNA-FX7™ Automated Cell Counter

### Physical and Technical Characteristics

	LUNA-FX7™ Basic Package	LUNA-FX7™ Advanced Package
Onboard storage	250 GB	1 TB
Additional software	-	Bioprocess software
Cell size range	1 - 90 µm	
Detection range	1 x 10 <sup>4</sup> - 5 x 10 <sup>7</sup> cells/mL (Optimal: 5 x 10 <sup>4</sup> - 1 x 10 <sup>7</sup> cells/mL)	
Cell detection method	Automated brightfield & fluorescence microscopy	
Slide options	1-Ch, 2-Ch, 3-Ch, 8-Ch, Reusable	
Measuring volume per chamber	0.5 - 5.1 µL/chamber (Each slide has different measuring volume.)	
Optics	Brightfield, Dual fluorescence	
Green fluorescence	Ex 470/40 nm, Em 530/50 nm	
Red fluorescence	Ex 530/40 nm, Em 620/60 nm	
Focusing	Autofocus with manual focus option	
Instrument type	Benchtop cell counter	
Display	7-inch TFT LCD multi-touch screen, 1024 x 600 pixels	
Data format	PDF, CSV, TIFF	
Data export	USB, WiFi, Ethernet	
Printer	External printer (optional)	
21 CFR Part 11	CountWire™ system (optional)	
User management	CountWire™ system (optional)	
IQ/OQ	Yes (optional)	
Dimensions	245 x 280 x 240 mm (9.6 x 11.0 x 9.4 inch)	
Weight	5.0 kg (11.02 lb)	
Rated line voltage	100 to 240 VAC	
Rated input current	1.5 A (at 100 VAC)	
Rated input frequency	50 to 60 Hz	
Output voltage / current	12 VDC / 5.0 A	

## LUNA™ Slides

### Physical Characteristics

Compatible Slides	LUNA™ 1-Channel Slides	LUNA™ Cell Counting Slides / PhotonSlide™	LUNA™ 3-Channel Slides	LUNA™ 8-Channel Slides	LUNA™ Reusable Slides
Image					
Material	Poly(methyl methacrylate) (PMMA)				Glass / Aluminium
Dimensions	25 x 75 x 2.1 mm	25 x 75 x 2.3 mm	25 x 75 x 2.1 mm	25 x 75 x 2.1 mm	25 x 75 x 2.5 mm

## 12. Ordering Information

### Instruments

Cat #	Product	Quantity
L70001	LUNA-FX7™ Automated Cell Counter, Basic Package	1
L70002	LUNA-FX7™ Automated Cell Counter, Advanced Package	1

### Slides and Reagents

Cat #	Product	Quantity
L72011	LUNA™ 1-Channel Slides, 50 Slides	1 box
L72012	LUNA™ 1-Channel Slides, 500 Slides	10 boxes
L72013	LUNA™ 1-Channel Slides, Sterile-gamma-irradiated, 500 Slides	10 boxes
L72001	LUNA™ 8-Channel Slides, 50 Slides	1 box
L72002	LUNA™ 8-Channel Slides, 500 Slides	10 boxes
L72003	LUNA™ 8-Channel Slides, Sterile-gamma-irradiated, 500 Slides	10 boxes
L72021	LUNA™ 3-Channel Slides, 50 Slides	1 box
L72022	LUNA™ 3-Channel Slides, 500 Slides	10 boxes
L72023	LUNA™ 3-Channel Slides, Sterile-gamma-irradiated, 500 Slides	10 boxes
L12001	LUNA™ Cell Counting Slides, 50 Slides	1 box
L12002	LUNA™ Cell Counting Slides, 500 Slides	10 boxes
L12003	LUNA™ Cell Counting Slides, 1000 Slides	20 boxes
L12005	PhotonSlide™, 50 Slides	1 box
L12006	PhotonSlide™, 500 Slides	10 boxes
L12007	PhotonSlide™, 1000 Slides	20 boxes
L12011	LUNA™ Reusable Slide	1 unit
L12014	LUNA™ Reusable Slide Coverslips	10 units
L72030	Cell Counter Validation Slide-FL	1 unit
L72040	Cell Counter Validation Slide-BF	1 unit
T13001	Trypan Blue Stain, 0.4% (200 tests)	2 x 1 mL
T13011	Trypan Blue Stain, 0.4%, Sterile-filtered	2 x 1 mL
L13002	Erythrosin B Stain (200 tests)	2 x 1 mL
F23001	Acridine Orange/Propidium Iodide Stain	2 x 0.5 mL
F23011	Acridine Orange/Propidium Iodide Stain, Sterile-filtered	2 x 0.5 mL
F73101	LUNA-FX™ Calibration Beads Kit	2 x 0.5 mL

### CountWire™

Cat #	Product	Quantity
L71001	CountWire™ Basic (1 CountWire™ Data Storage + 1 CountWire™ Verification Key)	1 set
L71002	CountWire™ Verification Key (additional)	1 unit

### IQ/OQ

Cat #	Product	Quantity
L74003	LUNA-FX7™ IQ/OQ Protocol	1 copy

## Accessories

Cat #	Product	Quantity
P17001	Thermal Printer	1 unit
P17002	Thermal Printer Refills, Paper and Ribbon	2 x 5 rolls

# 13. Purchaser Notification

## Limited Use Label License

**Research Use Only** The purchaser of this product should use this product only for research for the sole benefit of the purchaser. By use of this product, the purchaser agrees to be bounded by the terms of this limited use statement whether the purchaser is a for-profit or a not-for-profit entity.

If the purchaser is not willing to accept the conditions of this limited use statement and this product is unused, the Company will accept return of the product with a full refund.

The purchaser cannot resell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party for Commercial Purposes.

Commercial Purposes mean any and all uses of this product and its components by a party for monetary or other consideration, including but not limited to, (a) product manufacture, (b) providing a service, information, or data, (c) therapeutic, diagnostic, or prophylactic purposes, or (d) resale of this product or its components whether or not such product and its components are resold for use in research.

Aligned Genetics, Inc. ("Company") will not claim any consideration against the purchaser of infringement of patents owned or controlled by the Company which cover the product based on the manufacture, use or sale of a therapeutic, clinical diagnostic, vaccine, or prophylactic product developed in research by the purchaser in which this product or its components was employed, provided that neither this product nor any of its components was used in the manufacture of such product.

For any use other than this limited use label license of research use only, please contact the Company or email [info@logosbio.com](mailto:info@logosbio.com) for more information.

## Instrument Warranty

### Warranty

Aligned Genetics, Inc. ("Company") warrants to the original purchaser ("Purchaser") that the instrument ("Instrument"), if properly used and installed, will be free from defects in materials and workmanship and will conform to the product specifications for a period of one (1) year ("Warranty Period") from the date of purchase. If the Instrument under this limited warranty fails during the Warranty Period, the Company, at its sole responsibility, will: within and up to 30 calendar days of purchase, refund the purchase price of the Instrument to the Purchaser if the Instrument is in original conditions; or, after 30 calendar days of purchase, only replace or repair the Instrument for up to the Warranty Period without issuing a credit.

In no event shall the Company accept any returned instrument (including its components) that might have been used or contaminated in some labs, including but not limited to, HIV or other infectious disease or blood-handling labs. This limited warranty does not cover refund, replacement, and repair incurred by accident, abuse, misuse, neglect, unauthorized repair, or modification of the Instrument. This limited warranty will be invalid if the Instrument is disassembled or repaired by the Purchaser.

In case that the Company decides to repair the Instrument, not to replace, this limited warranty includes replacement parts and labor for the Instrument. This limited warranty does not include shipment of the Instrument to and from service location or travel cost of service engineer, the costs of which shall be borne by the Purchaser. Every effort has been made to ensure that all the information contained in this document is correct at its publication. However, the Company makes no warranty of any kind regarding the contents of any publications or documentation as unintended or unexpected errors including occasional typographies or other kinds are inevitable. In addition, the Company reserves the right to make any changes necessary without notice as part of ongoing product development. If you discover an error in any of our publications, please report it to your local supplier or the Company. The Company shall have no responsibility or liability for any special, incidental, indirect or consequential loss or damage resulting from the use or malfunction of the Instrument.

This limited warranty is sole and exclusive. The Company makes no other representations or warranties

of any kind, either express or implied, including for merchantability or fitness for a particular purpose with regards to this Instrument. To obtain service during the Warranty Period, contact your local supplier or the Company's Technical Support team.

### **Out of Warranty Service**

Please contact your local supplier or the Company's technical support team in order to obtain out-of-warranty service. If necessary, repair service will be charged for replacement parts and labor hours incurred to repair the Instrument. In addition, the Purchaser is responsible for the cost of shipping the Instrument to and from the service facility and, if necessary, the travel cost of a service engineer after 30 calendar days of purchase, only replace or repair the Instrument for up to the Warranty Period without issuing a credit.



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Aligned Genetics, Inc.

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