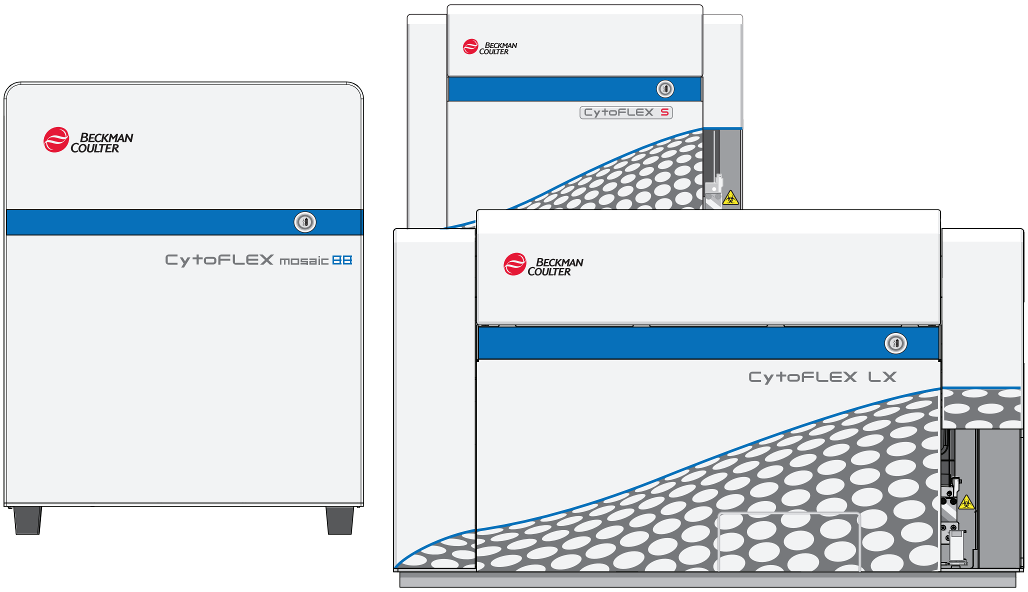
Instructions for Use

CytoFLEX mosaic

Spectral Detection Module for CytoFLEX S and CytoFLEX LX Flow Cytometers

For Research Use Only. Not for use in diagnostic procedures.



D17052AA
December 2024

Beckman Coulter, Inc.
250 S. Kraemer Blvd.
Brea, CA 92821 U.S.A.

CytoFLEX mosaic Spectral Detection Module for CytoFLEX S and CytoFLEX LX Flow Cytometers
PN D17052AA (December 2024)

Contact Us
If you have any questions, contact our Customer Support Center.

- Worldwide, find us via our website at www.beckman.com/support/technical.
- In the USA and Canada, call us at 1-800-369-0333.
- In Austria, call us at 0810 300484.
- In Germany, call us at 02151 333999.
- In Sweden, call us at +46(0)8 564 859 14.
- In the Netherlands, call us at +31 348 799 815.
- In France, call us at 0825838306 6.
- In the UK, call us at +44 845 600 1345.
- In Ireland, call us at +353 (01) 4073082.
- In Italy, call us at +39 0295392 456.
- In other locales, contact your local Beckman Coulter Representative.

EC REP
Beckman Coulter Eurocenter S.A.
22, rue Juste-Olivier
Case Postale 1044
CH - 1260 Nyon 1, Switzerland
Tel: +41 (0) 22 365 36 11

Glossary of Symbols is available at beckman.com/techdocs (PN C24689).

May be covered by one or more pat. - see www.beckman.com/patents

Original Instructions

Revision History

Initial Issue AA, 12/2024

Software Version 1.0

This document applies to the latest software listed and higher versions. When a subsequent software version affects the information in this document, a new issue will be released to the Beckman Coulter Web site. For labeling updates, go to www.beckman.com and download the latest version of the manual or system help for your instrument.

Safety Notices

Read all product manuals and consult with Beckman Coulter-trained personnel before attempting to operate the instrument. Do not attempt to perform any procedure before carefully reading all instructions. Always follow product labeling and manufacturer's recommendations. If in doubt as to how to proceed in any situation, contact us.

Beckman Coulter, Inc. urges its customers to comply with all national health and safety standards such as the use of barrier protection. This may include, but it is not limited to, protective eyewear, gloves, and suitable laboratory attire when operating or maintaining this or any other automated laboratory analyzer.

This manual assumes that users have basic knowledge of the Windows operating system, as well as experience working with laboratory testing technology. Users are invited to consult the appropriate documentation for such information.

Alerts for Danger, Warning, and Caution

⚠ DANGER

DANGER indicates an imminently hazardous situation which, if not avoided, will result in death or serious injury.

⚠ WARNING

WARNING indicates a potentially hazardous situation which, if not avoided, could result in death or serious injury.

⚠ CAUTION

CAUTION indicates a potentially hazardous situation, which, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices.

Safety Precautions

⚠ WARNING

Risk of operator injury if:

- **All doors, covers and panels are not closed and secured in place prior to and during instrument operation.**
- **The integrity of safety interlocks and sensors is compromised.**
- **Instrument alarms and error messages are not acknowledged and acted upon.**
- **You contact moving parts.**
- **You mishandle broken parts.**
- **Doors, covers and panels are not opened, closed, removed and/or replaced with care.**
- **Improper tools are used for troubleshooting.**

To avoid injury:

- **Keep doors, covers and panels closed and secured in place while the instrument is in use.**
- **Take full advantage of the safety features of the instrument.**
- **Acknowledge and act upon instrument alarms and error messages.**
- **Keep away from moving parts.**
- **Report any broken parts to your Beckman Coulter Representative.**
- **Open/remove and close/replace doors, covers and panels with care.**
- **Use the proper tools when troubleshooting.**

⚠ CAUTION

System integrity could be compromised and operational failures could occur if:

- **This equipment is used in a manner other than specified. Operate the instrument as instructed in the Product Manuals.**
- **You introduce software that is not authorized by Beckman Coulter into your computer. Only operate your system's computer with software authorized by Beckman Coulter.**
- **You install software that is not an original copyrighted version. Only use software that is an original copyrighted version to prevent virus contamination.**

⚠ CAUTION

If you purchased this product from anyone other than Beckman Coulter or an authorized Beckman Coulter distributor, and, it is not presently under a Beckman Coulter service maintenance agreement, Beckman Coulter cannot guarantee that the product is fitted with the most current mandatory engineering revisions or that you will receive the most current information bulletins concerning the product. If you purchased this product from a third party and would like further information concerning this topic, contact us.

⚠ CAUTION

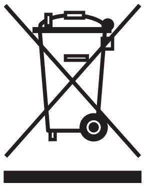
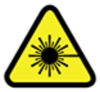
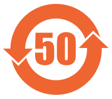
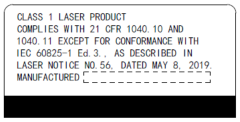
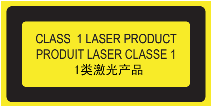
Risk of instrument damage. This device is intended for indoor use only. To avoid device damage, do not install the instrument outdoors.

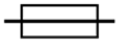
⚠ WARNING

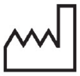
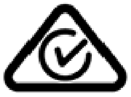
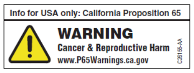
Risk of personal injury. Safety protection can be impaired if used in a manner not specified by the manufacturer. To avoid personal injury, use the instrument according to the manufacturer's instructions only.

Symbol Explanations

Symbol	Warning Condition	Action
	Biohazard Symbol	Consider all materials (specimens, reagents, controls, and monoclonal antibodies) and areas these materials come into contact with as being potentially infectious. Wear appropriate barrier protection and follow safe laboratory procedures when handling any material in the laboratory.
	Caution	To indicate that caution is necessary when operating the device or control close to where the symbol is placed, or to indicate that the current situation needs operator awareness or operator action in order to avoid undesirable consequences.
	Caution, Risk of Electric Shock	To indicate hazards arising from dangerous voltages.
	Warning, Sharp elements	To warn of sharp element.
	Warning, Crushing of hands	To warn of a closing motion of mechanical parts of equipment.

Symbol	Warning Condition	Action
	Recycling Symbol WEEE Wheeled Bin Symbol	The symbol of a crossed-out wheeled bin on the product is required in accordance with the Waste Electrical and Electronic Equipment (WEEE) Directive of the European Union. The presence of this marking on the product indicates: **1.** that the device was put on the European Market after August 13, 2005 and **2.** that the device is not to be disposed via the municipal waste collection system of any member state of the European Union. For products under the requirement of WEEE directive, please contact your dealer or local Beckman Coulter office for the proper decontamination information and take-back program which will facilitate the proper collection, treatment, recovery, recycling, and safe disposal of device. For the Japan market: This system is considered an industrial waste, subject to special controls for infectious waste. Before disposal of the system, refer to the Waste Disposal and Public Cleaning Law for compliance procedures.
	Laser Radiation	The laser radiation symbol indicates that there can be laser light radiation in the area. Take precautions to prevent exposure.
	RoHS Caution Symbol	This label indicates that the electronic information product contains certain toxic or hazardous substances. The center number is the Environmentally Friendly Use Period (EFUP) date, and indicates the number of calendar years the product can be in operation. Upon the expiration of the EFUP, the product must be immediately recycled. The circling arrows indicate the product is recyclable. The date code on the label or product indicates the date of manufacture.
CLASS 1 LASER PRODUCT COMPLIES WITH 21 CFR 1040.10 AND 1040.11 EXCEPT FOR CONFORMANCE WITH IEC 60825-1 Ed.3., AS DESCRIBED IN LASER NOTICE NO.56, DATED MAY 8, 2019. MANUFACTURED	Laser Compliance	This label indicates that the product is a Class 1 Laser Product and is in compliance with international standard and US requirements.
CLASS 1 LASER PRODUCT PRODUIT LASER CLASSE 1 1类激光产品	Laser Class I	This label indicates that this product is a Class I Laser product. Take precautions to prevent exposure.

Symbol	Warning Condition	Action
	Consider all materials (specimens, controls, monoclonal antibodies, and so forth) as being potentially infectious.	Wear standard laboratory attire and follow safe laboratory procedures when handling any material in the laboratory.
EC REP	EC Representative	Indicates the Authorized representative in the European Community.
CE	European Conformity (CE Mark) Regulatory Mark	A "CE" mark indicates that a product has been assessed before being placed on the market, and has been found to meet European Union safety, health, and/or environmental protection requirements.
UK CA	UKCA Mark	A "UKCA" mark indicates that a product has been assessed before being placed in UK market, and has been found to meet UK safety, health, and/or environmental protection requirements.
TÜVRheinland C US	TUV symbol	This test mark is proof of compliance with Canadian National standards and U.S. National standards. It demonstrates an electrical product has been successfully tested and certified by a Certification Body accredited by the Standards Council of Canada (SCC) and by a Nationally Recognized Testing Laboratory (NRTL) accredited by the Occupational Safety and Health Administration (OSHA).
○	Off (power)	To indicate disconnection from the mains, at least for mains switches or their positions, and all those cases where safety is involved.
|	On (power)	To indicate connection to the mains, at least for mains switches or their positions, and all those cases where safety is involved.
	Fuse	To identify fuse boxes or their location
	Manufacturer	Indicates the medical device manufacturer, as defined in EU Directives 90/385/EEC, 93/42/EEC and 98/79/EC.
	Consult the Instructions for Use Symbol	Indicates the need for the user to consult the instructions for use.

Symbol	**Warning Condition**	**Action**
	Date of Manufacture	Indicates the date when the medical device was manufactured.
SN	Serial Number	Indicates the manufacturer's serial number so that specific medical device can be identified.
#	Model Number	Indicates the model number or type number of a product.
	RCM Symbol	The "RCM" (Regulatory Compliance Mark) is depicted as a triangle with a partial circle and check. The mark is applied to products that comply with the EMC requirements of the Australian Communications Media Authority (ACMA) for use in Australia and New Zealand.
 Info for USA only: California Proposition 65 WARNING Cancer & Reproductive Harm www.P65Warnings.ca.gov C26155-AA	California Proposition 65	This product can expose you to chemicals known to the State of California to cause Cancer and Reproductive Harm. For more information go to https://www.P65Warnings.ca.gov.

Electromagnetic Compatibility (EMC)

This device complies with the emissions and immunity requirements as specified in the EN/IEC 61326 series of Product Family Standards for a “basic electromagnetic environment”. Such equipment is supplied directly at low voltage from public mains network. This equipment is not intended for residential use.

⚠ CAUTION

This device generates, uses, and can radiate unintentional radio-frequency (RF) energy. If this device is not installed and operated correctly, this RF energy can cause interference with other equipment. It is the responsibility of the end user to be sure that a compatible electromagnetic environment for the device can be maintained so that the device operates as intended.

In addition, other equipment can radiate RF energy to which this device is sensitive. If one suspects interference between this device and other equipment, Beckman Coulter recommends the following actions to correct the interference:

1. **Evaluate the electromagnetic environment before installation and operation of this device.**
2. **Do not operate this device close to sources of strong electromagnetic radiation (for example: unshielded intentional RF sources), as these can interfere with proper operation. Examples of unshielded intentional radiators are handheld radio transmitters, cordless phones, and cellular phones.**
3. **Do not place this device near medical electrical equipment that can be susceptible to malfunctions caused by close-proximity to electromagnetic fields.**
4. **This device has been designed and tested to CISPR 11, Class A emission limits. In a domestic environment, this device may cause radio interference, in which case, you may need to take measures to mitigate the interference.**

Contents

CHAPTER 4: Daily Startup, 4-1

CHAPTER 5: Instrument Quality Control and Standardization, 5-1

CHAPTER 6: Data Acquisition and Sample Analysis, 6-1

APPENDIX A: Instrument Installation, A-1

APPENDIX B: CytExpert for Spectral Electronic Record Management, B-1

Abbreviations

Beckman Coulter, Inc.
SOFTWARE END-USER LICENSE AGREEMENT

Related Documents

Illustrations

Tables

Introduction

Overview

This is an introductory chapter that gives a brief description of each document. It contains the following topics presented in this order:

- How to Use Your Manual
- About this Manual
- Conventions
- Graphics
- Advancing Your Skills

How to Use Your Manual

Your CytoFLEX mosaic Spectral Detection Module includes the manuals listed below:

- Use this **Instructions for Use** manual for information on the day-to-day operation of your CytoFLEX flow cytometer equipped with the CytoFLEX mosaic Spectral Detection Module. You can find detailed step-by-step procedures for Daily Startup and Quality Control, running samples, analyzing data, and performing Startup and Shutdown. This manual also contains physical and system specifications, safety and troubleshooting information, as well as information about what your CytoFLEX mosaic Spectral Detection Module does and the methods guiding operation. It also contains procedures for cleaning and maintenance.
- The **CytoFLEX Platform Instructions for Use** manual provides information on the day-to-day operation of your CytoFLEX flow cytometer not equipped with a CytoFLEX mosaic Spectral Detection Module.

NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

About this Manual

The information in the Instructions for Use manual is organized as follows:

CHAPTER 1, System Overview
Provides information regarding the individual components of the CytoFLEX flow cytometer and the CytoFLEX mosaic Spectral Detection Module, and the corresponding functions of these components.

CHAPTER 2, Using the CytExpert for Spectral Software
Provides an overview of each aspect of the software's functions.

CHAPTER 3, Operation Principles
Describes how the instrument measures scattered light and fluorescence as cells pass through the laser beam.

CHAPTER 4, Daily Startup
Provides instructions for starting your CytoFLEX flow cytometer equipped with a CytoFLEX mosaic Spectral Detection Module and navigating to the sample testing standby state.

CHAPTER 5, Instrument Quality Control and Standardization
Provides instructions for performing daily quality control (QC) on your CytoFLEX flow cytometer equipped with a CytoFLEX mosaic Spectral Detection Module to confirm the instrument is working correctly and to ensure accurate experimental data measurement. Quality control allows you to determine whether your instrument can provide adequate signal strength and precision.

CHAPTER 6, Data Acquisition and Sample Analysis
Provides instructions for operating the instrument, including data acquisition, analyzing and exporting results.

CHAPTER 7, Unmixing
Describes how to create an spectral unmixing experiment and automatically calculate unmixing values after acquiring the single color data. It also explains how to use these calculations for other experiments.

CHAPTER 8, Data Review
Describes how to use the Analysis screen to analyze data that has already been acquired.

CHAPTER 9, Daily Shutdown
Describes how to keep the instrument in optimal condition through daily cleaning during the shutdown procedure.

CHAPTER 10, Troubleshooting
Describes some common problems and their solutions in a basic troubleshooting matrix.

CHAPTER 11, Cleaning Procedures
Describes how to carry out certain routine and nonscheduled cleaning procedures.

CHAPTER 12, Replacement/Adjustment Procedures
Describes how to carry out certain routine and nonscheduled replacement and adjustment procedures.

APPENDIX A, Instrument Installation
Provides the instrument installation procedures for your CytoFLEX flow cytometer.

APPENDIX B, CytExpert for Spectral Electronic Record Management
Provides the instructions for using the CytExpert Electronic Record Management software option.

APPENDIX C, Sample Injection Mode Control Kit
Provides the instructions for using the CytoFLEX Sample Injection Mode Control Kit.

APPENDIX D, Deep Well Plate
Provides a list of deep well plates suggested for use on the Plate Loader DW.

APPENDIX E, Cyber Security
Provides instructions for cyber security.

APPENDIX F, Table of Hazardous Substances
Provides the table of hazardous substances with the hazardous substance name and concentration.

Conventions

This document uses the following conventions:

- **Bold face** font indicates buttons or selections that appear on the workstation screen.
- The term “select” is used to indicate the following action:
  - To click with a mouse.

  NOTE The verb “press” is reserved for mechanical buttons, such as keys on the keyboard.

- The software path to a specific function or screen appears with the greater than (>) symbol between screen options.
- Links to information in another part of the document for additional information are in blue and are underlined. To access the linked information, select the blue, underlined text.
- The information in your Instructions for Use manual applies to the CytoFLEX S instruments equipped with and without a plate loader, and the CytoFLEX LX, unless otherwise specified.

  NOTE When information in this document only applies to the plate loader configuration, it is marked [With Plate Loader]. When information in this document only applies to the configuration not equipped with a plate loader, it is marked [Without Plate Loader].

  When information in this document only applies to one instrument in the platform, it is marked either [CytoFLEX S] or [CytoFLEX LX].

  When information in this document only applies to the CytoFLEX mosaic Spectral Detection Module, it is marked [CytoFLEX mosaic Spectral Detection Module]. When information in this document only applies to one type of the CytoFLEX mosaic Spectral Detection Module, it is marked either [CytoFLEX mosaic 63] or [CytoFLEX mosaic 88].

  NOTE There are two kinds of plate loaders, the standard CytoFLEX Plate Loader and the CytoFLEX Plate Loader Deep Well (DW). The CytoFLEX Plate Loader DW (hereinafter called as Plate Loader DW) is compatible with CytoFLEX S and CytoFLEX LX instruments. When information in this document only applies to one plate loader, it is marked either [Plate Loader DW] or [Standard Plate Loader].

- The CytExpert for Spectral Electronic Record Management software installation screens are shown in all instances unless otherwise specified.

IMPORTANT IMPORTANT is used for comments that add value to the step or procedure being performed. Following the advice in the IMPORTANT adds benefit to the performance of a piece of equipment or to a process.

NOTE NOTE is used to call attention to notable information that should be followed during use, or maintenance of this equipment.

Graphics

All graphics, including screens and printouts, are for illustration purposes only and must not be used for any other purpose. For example, software screens that show the CytoFLEX for Spectral software in the background may not depict the latest production version of the system.

Advancing Your Skills

To learn more about using your CytoFLEX mosaic Spectral Detection Module system, e-Learnings are available through the Beckman Coulter Learning Center.

- Instructor-Led Coursed
  - CytoFLEX mosaic In-Person Training
  - CytoFLEX mosaic Remote Training
- E-Learning Modules
  - CytoFLEX mosaic Basic Operator Training

For further information about the available courses, visit the Beckman Coulter Learning Center at http://www.beckman.com/service/training, and enter **CytoFLEX mosaic** in the Search field or contact us.

CHAPTER 1

System Overview

Overview

This chapter describes the individual components of the CytoFLEX flow cytometer, CytoFLEX mosaic Spectral Detection Module and the corresponding functions of these components.

This chapter contains information on:

Product Description

The CytoFLEX mosaic Spectral Detection Module is an optional accessory which can be attached to the CytoFLEX S and CytoFLEX LX flow cytometers to switch the cytometers from Conventional Mode to Spectral Mode. Spectral Mode enables the spectral flow functionality and allows users to run more complex assays and use spectral unmixing technology for spectral overlap correction.

The CytoFLEX S and CytoFLEX LX flow cytometers equipped with a CytoFLEX mosaic Spectral Detection Module are used for the qualitative and quantitative measurement of biological and physical properties of cells and other particles. These properties are measured when the cells pass through one or multiple laser beams in a single-file.

For Research Use Only. Not for use in diagnostics procedures.

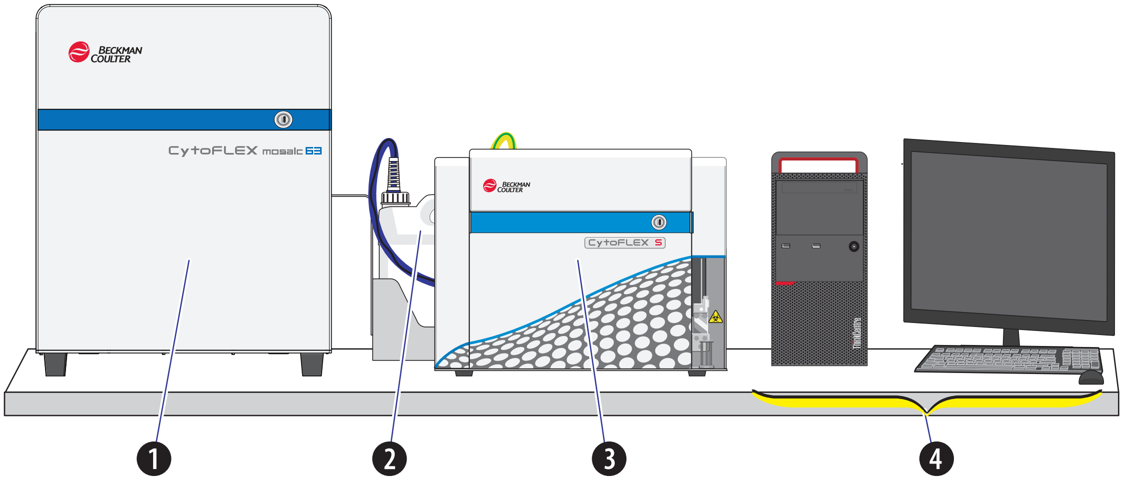
Main Components

⚠ CAUTION

Risk of instrument damage and/or instrument stability. Do not place any objects on top of the instrument, as this could cause warping of the top cover or affect the stability of the optical path.

The instrument consists of four main components: Fluid Containers/Cubitainers, Cytometer, the Workstation and CytoFLEX mosaic Spectral Detection Module.

Figure 1.1 Main Components [CytoFLEX S Without Plate Loader]



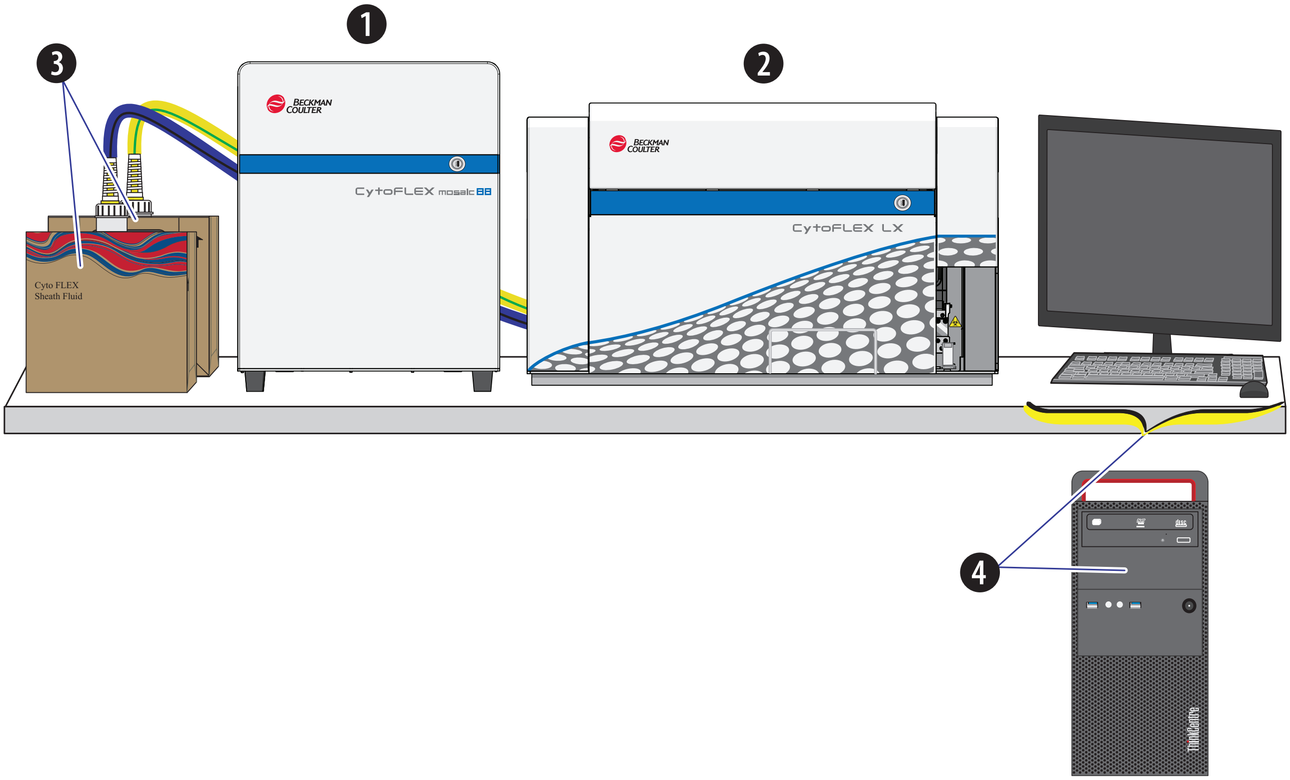
1. **CytoFLEX mosaic 63.** Provides signal collection with up to 63 channels using a CytoFLEX S configuration.

   NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

   NOTE Refer to the *CytoFLEX Platform Instructions for Use* manual if the instrument is being used without the CytoFLEX mosaic Spectral Detection Module connected.

2. **Fluid Containers.** Accommodates sheath fluid and waste liquid as required for operation of the instrument.
3. **Cytometer.** Provides signal generation.
4. **Workstation.** Displays the content of the workstation and data generated by the Cytometer.

Figure 1.2 Main Components [CytoFLEX LX]



1. **CytoFLEX mosaic 88.** Provides signal collection with up to 88 channels using a CytoFLEX LX configuration.

   NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

   NOTE Refer to the *CytoFLEX Platform Instructions for Use* manual if the instrument is being used without the CytoFLEX mosaic Spectral Detection Module connected.

2. **Cytometer.** Provides signal generation.
3. **Fluid Cubitainers.** Accommodates sheath fluid and waste liquid as required for operation of the instrument.

   NOTE The CytoFLEX LX does not have a fluid container holder.

4. **Workstation.** Displays and manipulates the contents of the Workstation and displays data generated by the Cytometer.

Optical Components

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Optical Components for detailed instructions.

NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

Wavelength Division Multiplexer (WDM)

Each WDM corresponds to a different laser, or in some cases two lasers. The WDMs are contained within the CytoFLEX mosaic Spectral Detection Module and cannot be altered to change filter configuration. Note that the CytoFLEX S equipped with a CytoFLEX mosaic 63 allows for up to 63 channel while the CytoFLEX LX equipped with a CytoFLEX mosaic 88 allows for 88 channels.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Wavelength Division Multiplexer (WDM) for detailed instructions.

NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

Optical Fiber

⚠ CAUTION

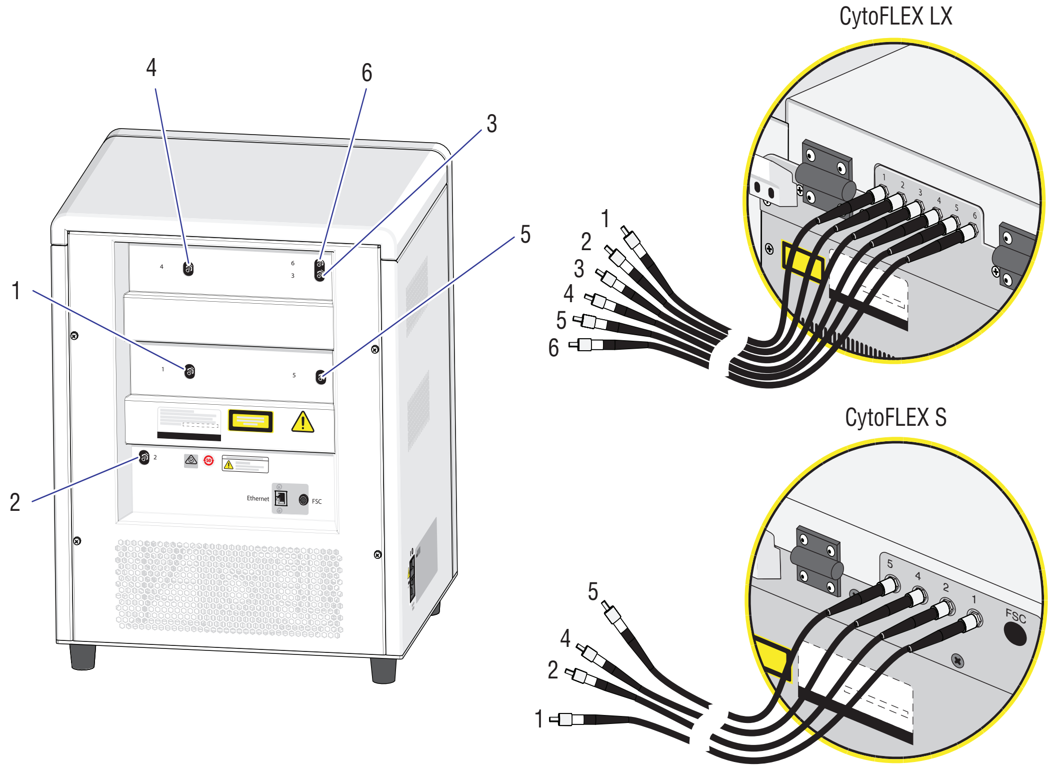
Risk of data integrity damage.

- **During use, verify that the optical fibers are securely connected between the fiber intake bracket at the back of the instrument and the back of the CytoFLEX mosaic Spectral Detection Module. A loose connection can alter the optical path and affect fluorescence detection.**
- **Take care when disconnecting the fibers as this could contaminate the tip and weaken the signal. Always ensure the fiber is reconnected right away.**
- **Do not kink the optical fibers.**

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Optical Fiber for detailed instructions.

NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

Fluorescence emitted by laser-excited fluorochromes is picked up and delivered by each optical fiber to the corresponding detector module. CytoFLEX S optical fiber has a colored ring on the end that connects to the WDM, indicating the color of the corresponding laser. CytoFLEX LX optical fiber has a numbered label on the end that connects to the WDM, indicating the number of the corresponding laser. Ensure that the correct fiber is properly connected to the corresponding WDM.



1. Violet laser fiber
2. NUV or UV laser fiber
3. Red laser fiber
4. Yellow laser fiber
5. Blue laser fiber
6. Infrared laser fiber

Fluidics System

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Fluidics System for detailed instructions.

Fluid Containers/Cubitainers

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Fluid Containers/Cubitainers for detailed instructions.

Fluidics Module

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Fluidics Module for detailed instructions.

Sample Station

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Sample Station for detailed instructions.

Sample Tube Holder Positions

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Sample Tube Holder Positions for detailed instructions.

Plate Loader Components

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Plate Loader Components for detailed instructions.

Plate Holder Components

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Plate Holder Components for detailed instructions.

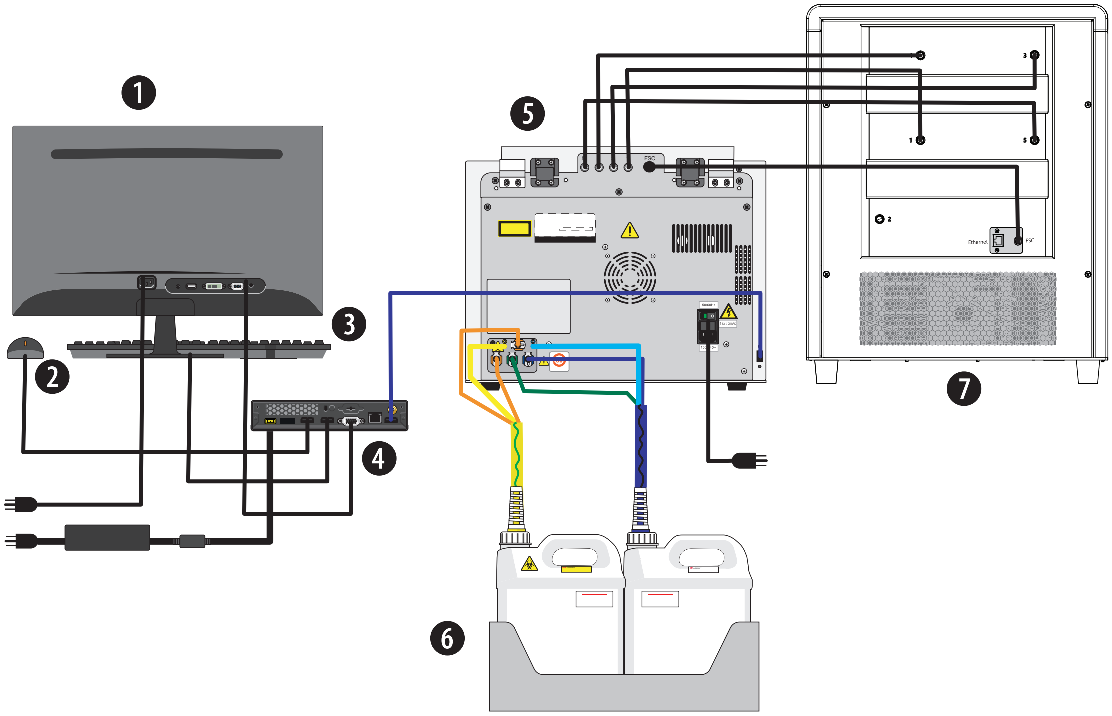
System Configuration

⚠ CAUTION

Risk of data loss and/or instrument damage. Never shut off the power or disconnect a data cable while the instrument is in the process of performing a task. This could cause data loss or damage to the system.

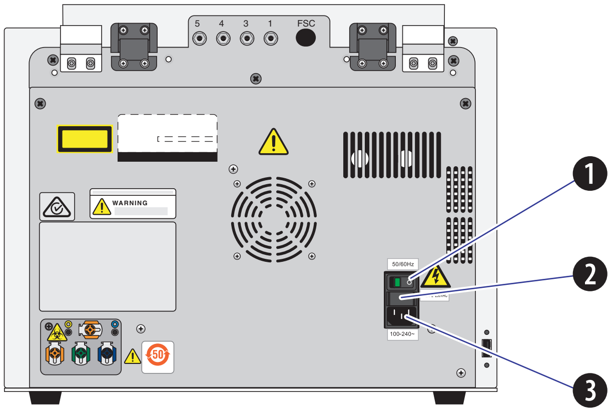
System Configuration [CytoFLEX S]

Figure 1.3 System Connections



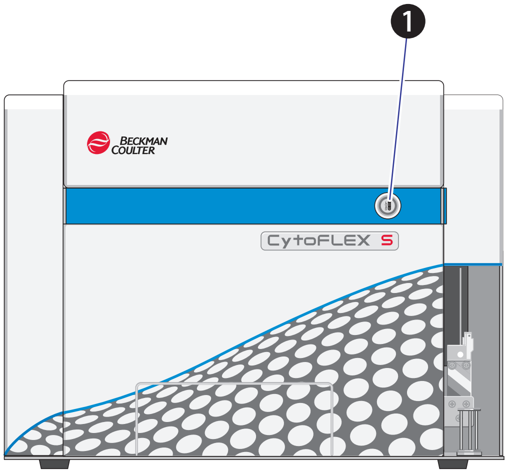
1. Monitor
2. Mouse
3. Keyboard
4. Computer
5. Cytometer
6. Fluid Container holder
7. CytoFLEX mosaic 63

Figure 1.4 Power Entry Module



1. **Power switch.** Turns Cytometer on and off. An indicator light glows when the power is on.
2. **Fuse.** Protects the internal system from damage by high electrical current.
3. **Power line socket.** Supplies the power to the Cytometer.

Figure 1.5 Front of Cytometer **[CytoFLEX S Without Plate Loader Shown]**



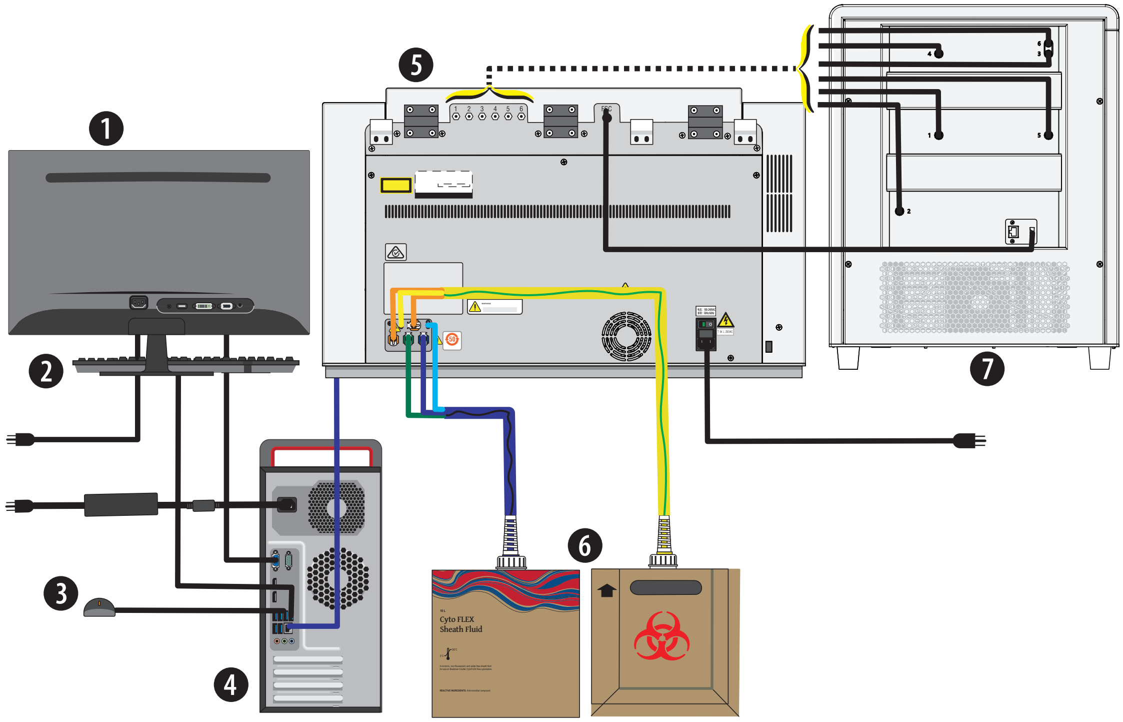
1. **Load button.** In addition to the software controls, this button can be used for automatic sample loading and data recording.

   NOTE This function is not available in the Plate Loader sample injection mode.

System Configuration [CytoFLEX LX]

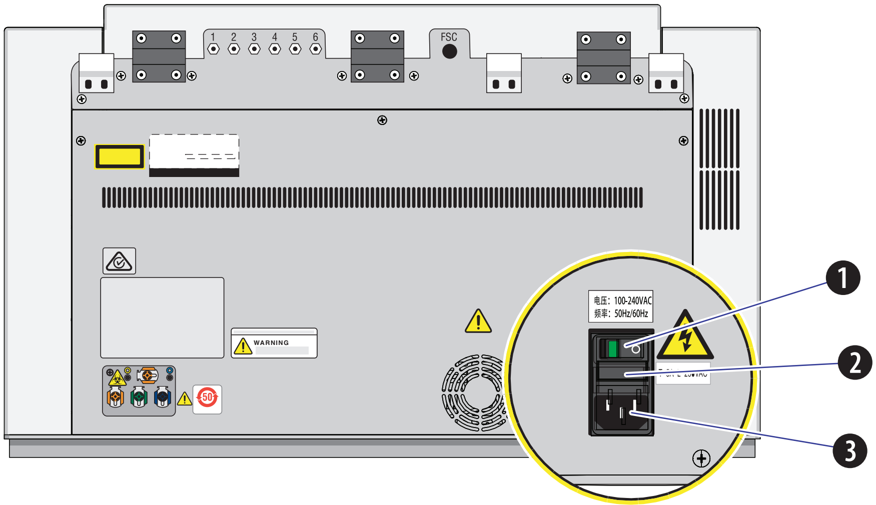
IMPORTANT The fluid cubitainers must be on the same level as the Cytometer. Do not place the fluid cubitainers on the floor.

Figure 1.6 System Connections



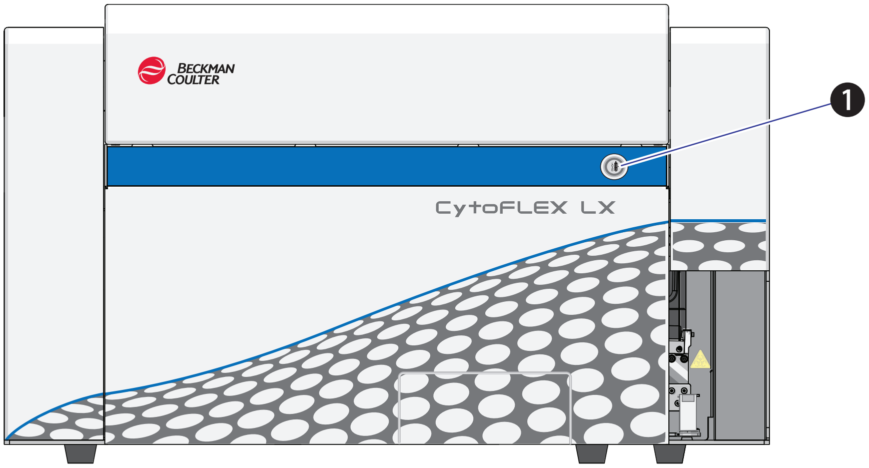
1. Monitor
2. Keyboard
3. Mouse
4. Computer
5. Cytometer
6. Fluid Cubitainers
7. CytoFLEX mosaic 88

Figure 1.7 Power Input Module



1. **Power switch.** Turns Cytometer on and off. An indicator light glows when the power is on.
2. **Fuse.** Protects the internal system from damage by high electrical current.
3. **Power line socket.** Supplies the power to the Cytometer.

Figure 1.8 Front of Cytometer

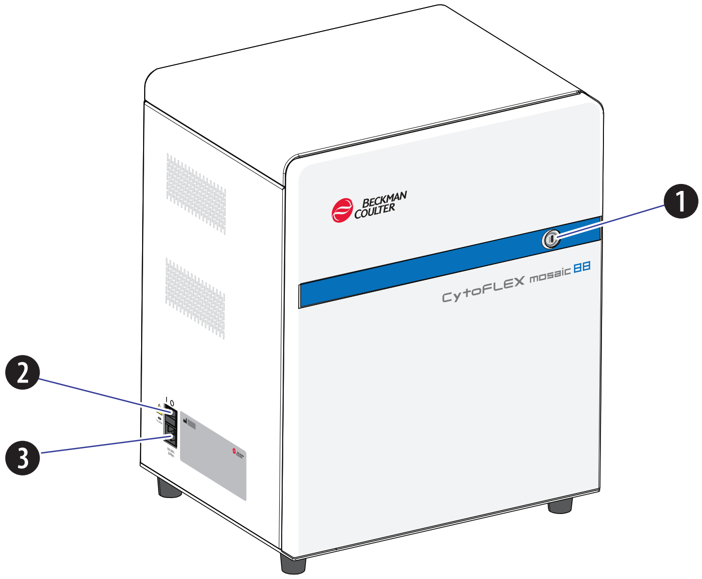


1. **Load button.** In addition to the software controls, this button can be used for automatic sample loading and data recording.

   NOTE This function is not available in the Plate Loader sample injection mode.

System Configuration [CytoFLEX mosaic Spectral Detection Module]

Figure 1.9 Front and Left Sides of the CytoFLEX mosaic Spectral Detection Module

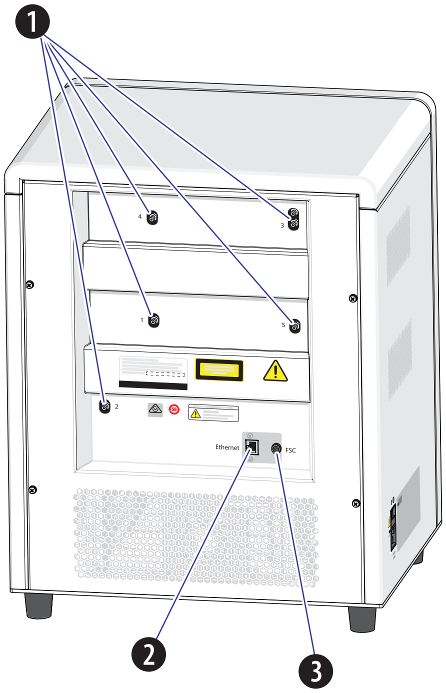


1. **Soft button.** In addition to the **Turn Off CytoFLEX mosaic** in the Cytometer menu on the software, this button can be used to turn the CytoFLEX mosaic Spectral Detection Module on/off. An indicator light glows when the power is on.

   NOTE To fully remove the instrument from power, the power switch on the left side of the instrument must be in the off position.

2. **Power switch.** Turns the CytoFLEX mosaic Spectral Detection Module on and off. An indicator light glows when the power is on.
3. **Power line socket.** Supplies the power to the Cytometer.

Figure 1.10 Back Cover Connections of the CytoFLEX mosaic Spectral Detection Module



1. **Optical fiber connectors.** Connects to the optical fibers.
2. **Ethernet connector.** Connects to the Ethernet cable of the workstation.
3. **FSC cable connector.** Connects to the FSC cable.

Consumables and Supplies

Reagents

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Reagents for detailed instructions.

NOTE The CytoFLEX Daily QC Fluorospheres are not intended to be used with the CytoFLEX mosaic Spectral Detection Module.

Ordering Information

Your instrument may be upgraded to a more highly configured model. For information on specific upgrades, replacement parts, or supplies, visit:

- www.beckman.com/supplies/cytoflex-platform-upgrades

Otherwise, contact us.

Instrument Specifications

Dimensions [CytoFLEX S]

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Dimensions [CytoFLEX S] for detailed instructions.

Dimensions [CytoFLEX LX]

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Dimensions [CytoFLEX LX] for detailed instructions.

Dimensions [CytoFLEX mosaic Spectral Detection Module]

Dimensions		
Instrument dimensions (Length x Width x Height)	**CytoFLEX mosaic 63**	42.5 cm x 34.5 cm x 53.5 cm
	CytoFLEX mosaic 88	42.5 cm x 34.5 cm x 53.5 cm
Weight	**CytoFLEX mosaic 63**	30 kg
	CytoFLEX mosaic 88	33 kg

Installation Category

Installation Category 2

Maximum Altitude

Do not operate at an altitude greater than 2000 m (6561 ft). Beckman Coulter assumes no responsibility for any problems resulting from operating instrument at an altitude greater than 2000 m (6561 ft).

Pollution Degree

Pollution Degree 2

Acoustic Noise Level

Measure Level: <65 dBA

Electrical Ratings

Voltage: AC 100-240 V ± 10%

Frequency: 50/60 Hz

Power:

- CytoFLEX mosaic Spectral Detection Module: 450 VA
- CytoFLEX S: 250 VA
- CytoFLEX LX: 300 VA

Cytometer

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Cytometer for detailed instructions.

CytoFLEX mosaic Spectral Detection Module

Optics	
Excitation optics	The optical system is alignment free. The laser delays are automatically adjusted by the daily QC system, if required. No user intervention is required to ensure optimum system performance. **CytoFLEX S**: The system can be configured with up to four spatially-separated lasers. **CytoFLEX LX**: The system can be configured with up to six spatially-separated lasers.
Emission optics	Alignment-free integrated optics quartz flow cell design with >1.3 NA. Flow Cell dimensions: 430 µm x 180 µm internal diameter.

Optics		
Laser devices	**CytoFLEX S standard wavelengths**	**Violet laser** - Wavelength: 405 nm, 80 mW - Beam spot size: 5 µm x 80 µm **Blue laser** - Wavelength: 488 nm, 50 mW - Beam spot size: 5 µm x 80 µm **Yellow laser** - Wavelength: 561 nm, 30 mW - Beam spot size: 5 µm x 80 µm **Red laser** - Wavelength: 638 nm, 50 mW - Beam spot size: 5 µm x 80 µm
	CytoFLEX LX standard wavelengths	**Ultraviolet (UV) laser** - Wavelength: 355 nm, 20 mW - Beam spot size: 5 µm x 80 µm **Near Ultraviolet (NUV) laser** - Wavelength: 375 nm, 60 mW - Beam spot size: 5 µm x 80 µm **Violet laser** - Wavelength: 405 nm, 80 mW - Beam spot size: 5 µm x 80 µm **Blue laser** - Wavelength: 488 nm, 50 mW - Beam spot size: 5 µm x 80 µm **Yellow laser** - Wavelength: 561 nm, 30 mW - Beam spot size: 5 µm x 80 µm **Red laser** - Wavelength: 638 nm, 50 mW - Beam spot size: 5 µm x 80 µm **Infrared (IR) laser** - Wavelength: 808 nm, 60 mW - Beam spot size: 5 µm x 80 µm
Forward scatter detection	Silicon photodiode with built-in 488/8 band-pass filter.	
Fluorescence and side scatter detection	Fluorescence and side scatter light collected by the objective lens is delivered by fiber optics to a patent-pending design with high performance, solid-state, high efficiency, low-noise detector array. Reflective optics with a single transmission band-pass filter in front of each detector.	
Side scatter (SSC) configuration	The system offers the ability to collect side scatter lights from each laser to better resolve particles from noise.	

Electronics	
Signal processing	7 decade data display
Digital sampling rate	6.25 MHz
Signal	Pulse area and height for every channel, width for two selectable channels

Data Management		
Software	CytExpert for Spectral software	
Language	English	
FCS format	FCS 3.1 and FCS 3.0	
Operating system	Windows 10 Enterprise LTSC 2019 64 bit	
Minimum workstation/ computer requirements [CytoFLEX S and CytoFLEX LX]	**Processor**	Intel Core i7-11700K Processor, 3.6GHz
	Memory	64 GB RAM
	Storage	1 TB SSD
	Ethernet	2 Ethernet interface
	USB	4 USB 3.0 ports
	Monitor resolution	1920 x 1080 or 2560 x 1440

Performance Characteristics

Performance Characteristics [CytoFLEX S]

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Performance Characteristics [CytoFLEX S] for detailed instructions.

Performance Characteristics [CytoFLEX LX]

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Performance Characteristics [CytoFLEX LX] for detailed instructions.

Performance Characteristics [CytoFLEX mosaic Spectral Detection Module]

Performance		
Sensitivity	**MESF**	FITC: <30 molecules of equivalent soluble fluorochrome (MESF-FITC) PE: <10 molecules of equivalent soluble fluorochrome (MESF-PE) PB450: <50 molecules of equivalent soluble fluorochrome (MESF-PB450) APC: <25 molecules of equivalent soluble fluorochrome (MESF-APC)
Fluorescence detection configuration	**Detection range**	**Ultraviolet (UV) laser:** • Detectors: 20 • Wavelength: 365-950 nm **Near Ultraviolet (NUV) laser** • Detectors: 18 • Wavelength: 420-950 nm **Violet laser:** • Detectors: 20 • Wavelength: 420-950 nm **Blue laser:** • Detectors: 16 • Wavelength: 498-950 nm **Yellow laser:** • Detectors: 12 • Wavelength: 567-950 nm **Red laser:** • Detectors: 10 • Wavelength: 649-950 nm **Infrared (IR) laser:** • Detectors: 3 • Wavelength: 830-950 nm
Fluorescence detection	Up to 79 fluorescent detectors are designed to cover a broad wavelength of spectrum	
Fluorescence resolution	rCV <3% The CytoFLEX Flow cytometer is capable of achieving <3% rCV. Using CytoFLEX Daily QC Fluorospheres, CytoFLEX Ready to Use Daily QC Fluorospheres or Daily IR QC Fluorospheres (for 808 nm Laser) for daily QC, the pass criteria is ≤5% for the Violet, Blue, Yellow, and Red lasers while the pass criteria is ≤7% for NUV, UV and IR lasers.	

Performance		
Blue/Yellow/Red/ Infrared/Ultraviolet/ Near Ultraviolet side scatter resolution	<300 nm	
Violet side scatter resolution	80 nm relative to polystyrene particles	
Forward and blue side scatter resolution	Scatter performance is optimized for resolving lymphocytes, monocytes, and granulocytes as well as nanoparticles.	
Carryover	**Single Tube Loader format**	≤1.0%
Signal acquisition speed	30,000 events/second	

Reagent Limitations

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 1, Reagent Limitations for detailed instructions.

Safety Data Sheets (SDS/MSDS)

To obtain an SDS or MSDS for CytoFLEX Platform reagents used on the CytoFLEX Platform systems:

1. On the Internet, go to www.beckman.com:
    a. Select **Safety Data Sheets (SDS/MSDS)** from the Support menu.
    b. Follow the instructions on the screen.
    c. Contact us if you have difficulty locating the information.
2. If you do not have Internet access, contact us.

CHAPTER 2

Using the CytExpert for Spectral Software

Overview

The CytExpert for Spectral software is a full-feature software package that controls the instrument operation, collection of experiment data, and analysis of the results. This chapter will explain the software functions and features.

NOTE The CytExpert for Spectral software only applies to the CytoFLEX S and CytoFLEX LX instruments equipped with the CytoFLEX mosaic Spectral Detection Module. If your CytoFLEX instrument is not equipped with a CytoFLEX mosaic Spectral Detection Module or if you are using Conventional Mode, please use the CytExpert software. Refer to the *CytoFLEX Platform Instructions for Use* manual for detailed instructions on using the CytExpert software.

IMPORTANT The CytExpert software and the CytExpert for Spectral software can be installed on the same computer, but you cannot use the CytExpert for Spectral software and the CytExpert software together.

This chapter contains information on:

- Launching the Software
- Main Software Screen
- User Management
- Role Management
- Account Policies
- User Management Operation Log
- Graphic and Gating Styles
- Software Settings

Launching the Software

Select the desktop shortcut to launch the CytExpert for Spectral software.

If there is no desktop shortcut, run the "CytExpert.exe" software directly from the software installation directory. The default installation path is C:/Program Files/CytExpert for Spectral. Or, select ⊞ **> All Programs > CytExpert for Spectral**.

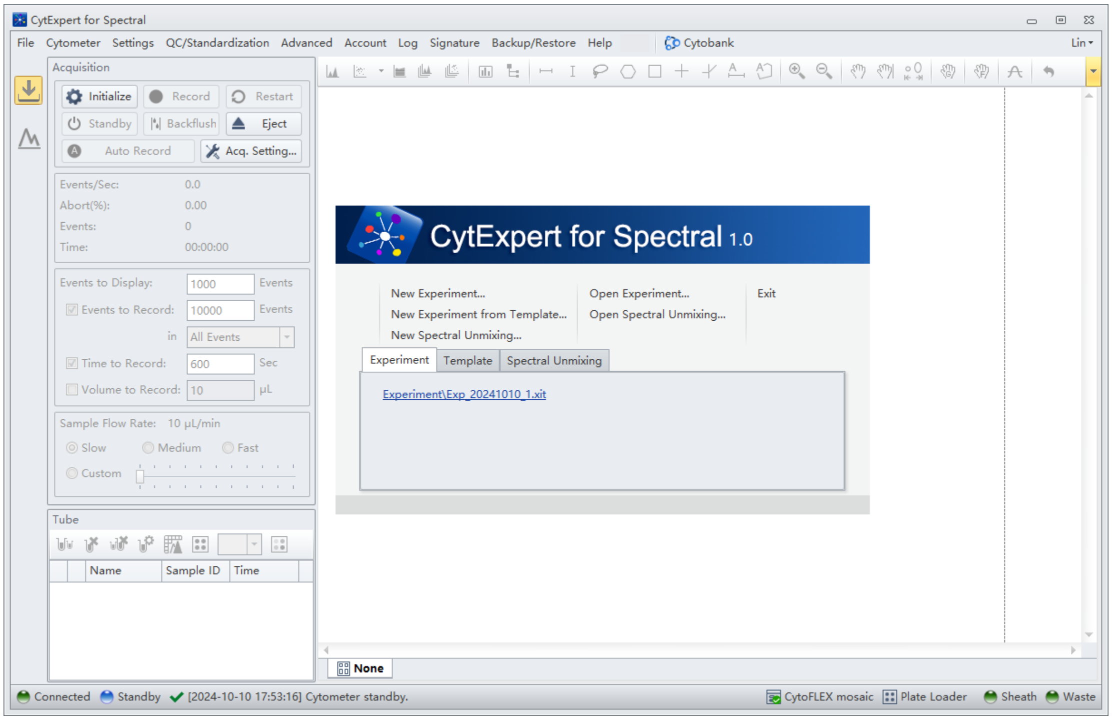
Refer to CHAPTER 4, Logging into the Software, for detailed instructions on opening the software and confirming the connection status.

Main Software Screen

Hover your cursor over any button to display a text pop-up of the button's function.

Start Page

The login window automatically opens after the software has been launched. The start page automatically opens after logging into the software.



Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

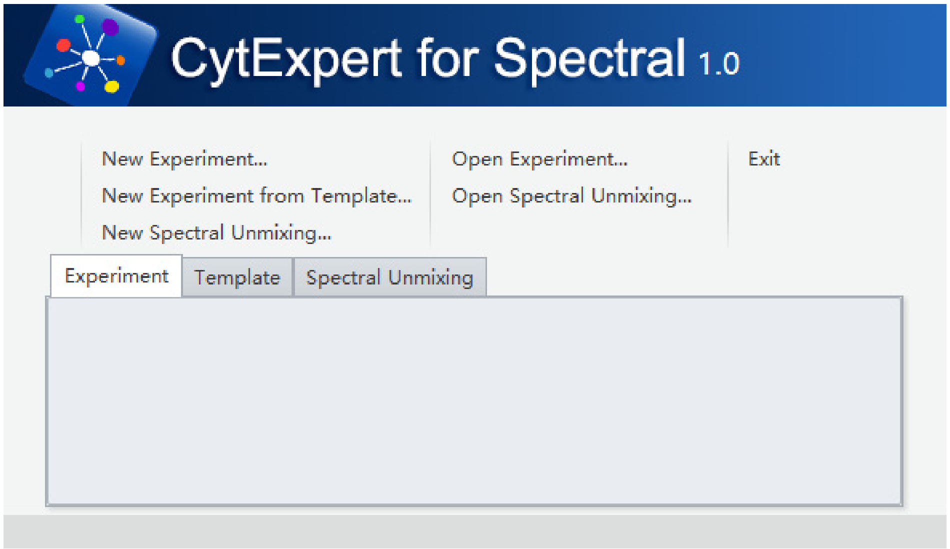
Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min

Experiment\Exp_20241010_1.xit

Connected Standby ✓ [2024-10-10 17:53:16] Cytometer standby.

The following operations can be selected from the start page:

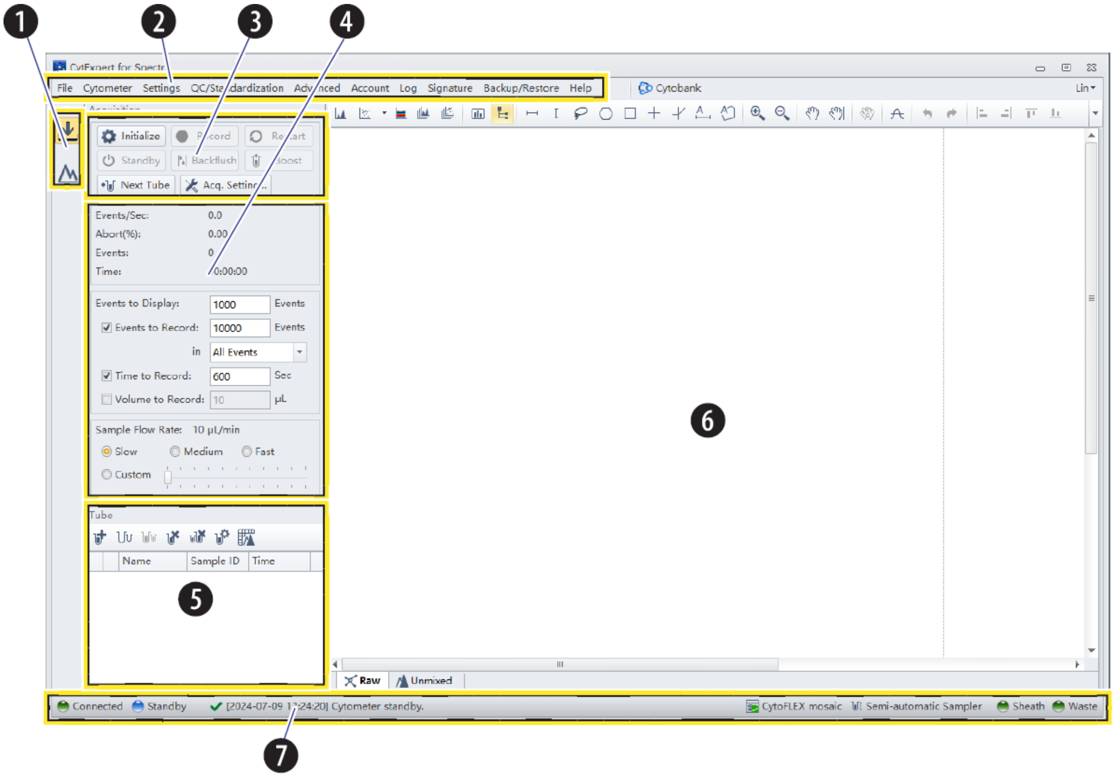


- **New Experiment**. For creating a new experiment. The process creates a file with the .xit extension and a folder with the same file name where the data (.fcs files) are kept.
- **New Experiment from Template**. For creating an experiment using a template saved from a previously saved experiment.
- **New Spectral Unmixing.** For creating a new spectral unmixing experiment. The process creates a file with the .xitsp extension and a folder with the same file name where the data (.fcs files) are kept.
- **Open Experiment**. For opening a previously created experiment.
- **Open Spectral Unmixing.** For opening a previously created spectral unmixing experiment.
- **Exit**. For exiting CytExpert for Spectral.

The Experiment, Template, and Spectral Unmixing tabs below give you the option of opening one of the 10 most recently opened experiments.

Acquisition Screen

Selecting **New Experiment**, **New Experiment from Template**, or **Open Experiment** automatically opens the Acquisition screen. The Acquisition screen can be accessed by selecting ⤓ on the left side of the page.



Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 0:00:00
Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL
Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

✓ [2024-07-09 1[illegible]:24:20] Cytometer standby.

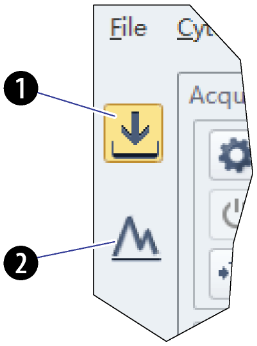
1. **Navigation.** Gives the option of accessing the Acquisition screen or Analysis screen.
2. **Menu.** Allows you to configure settings for sample acquisition, instrument operation, and software options.
3. **Instrument operation controls.** Controls sample loading/unloading and data acquisition and recording.
4. **Collection.** Establishes control over data recording options, displays the acquisition status, and controls the sample flow rate.
5. **Test tubes.** Allows you to configure and duplicate sample tubes, set display attributes, manage experimental data and unmixing.

   NOTE The Tube section of the screen can be expanded or retracted by dragging the top border of the Tube section of the screen. Expanding this section covers other elements of the screen, including: Events to Display, Events/Sec, and the Acquisition buttons.

6. **Plot area.** Includes plot and gating controls, as well as an area for creating plots and generating graphs. There are two worksheets display in this area, Raw worksheet and Unmixed worksheet.
7. **Status bar.** Displays instrument connection status and system information.

Acquisition Screen Navigation

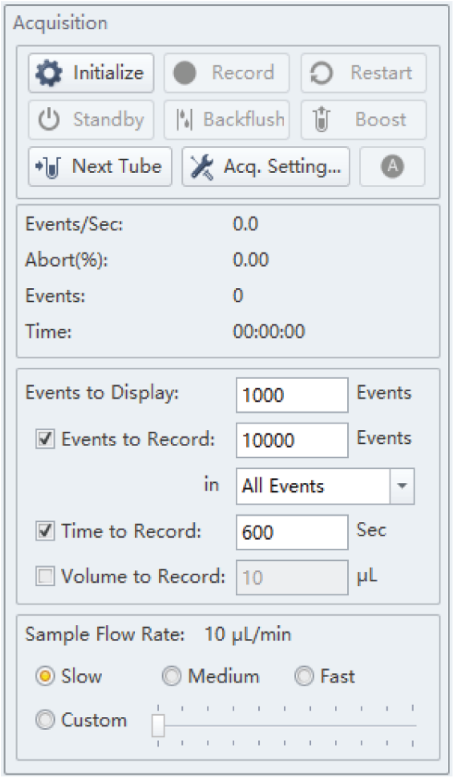
The Acquisition screens have two navigation icons, one for the Acquisition screen and the other for the Analysis screen.



1. **Acquisition screen icon.** Accesses the Acquisition screen.
2. **Analysis screen icon.** Accesses the Analysis screen.

Collection

Standby state

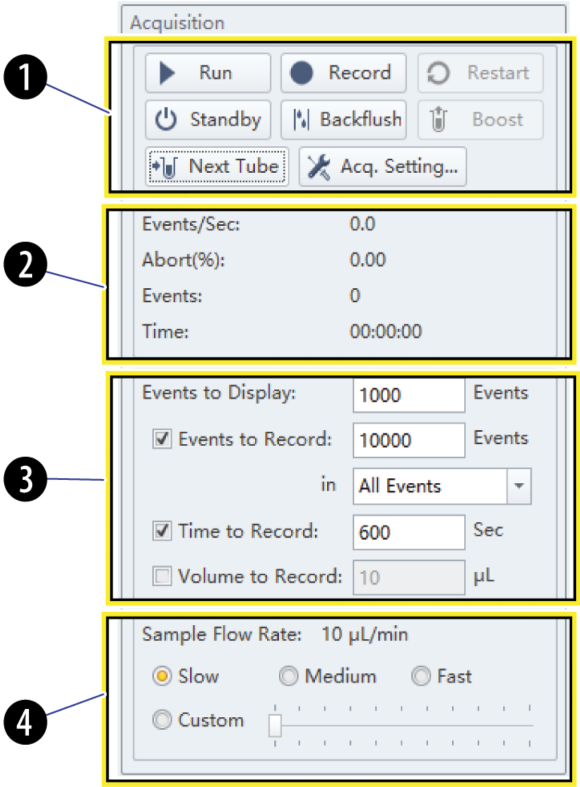


Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

Initialized state



Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

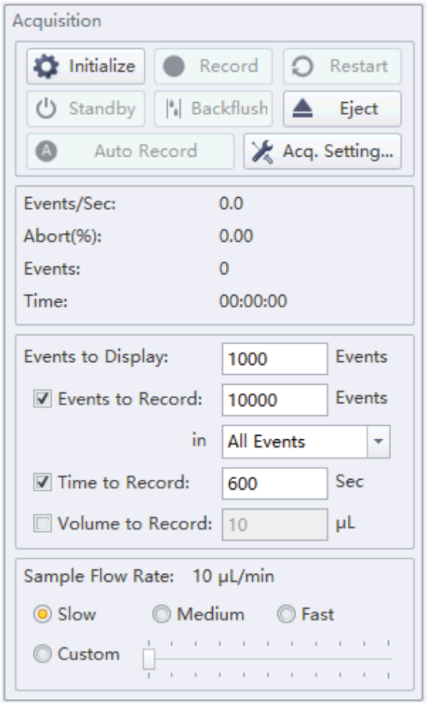
1. **Acquisition control.** Controls sample loading/unloading and data acquisition and recording.
2. **Acquisition status.** Displays such information as the acquisition rate (Events/Sec), event count, duration, and abort (%).
3. **Acquisition conditions.** Sets the necessary conditions for recording data.
4. **Sample flow rate.** Sets the acquisition rate for data collection.

NOTE High acquisition rate may increase the abort rate and measurement CVs.

Custom: The flow rate can be adjusted in 1 µL increments.

Collection [With Plate Loader]

Standby state

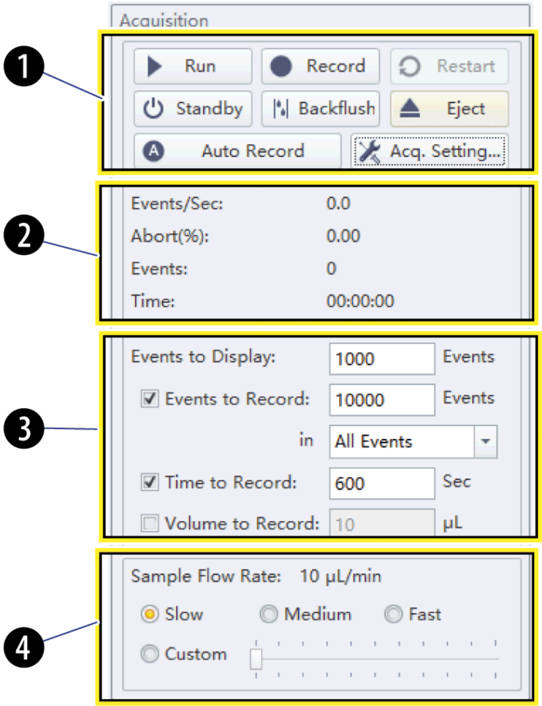


Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

Initialized state



Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

1. **Acquisition control.** Controls sample loading/unloading and data acquisition and recording.
2. **Acquisition status.** Displays such information as the acquisition rate (Events/Sec), event count, duration, and abort (%).
3. **Acquisition conditions.** Sets the necessary conditions for recording data.
4. **Sample flow rate.** Sets the acquisition rate for data collection.

NOTE High acquisition rate may increase the abort rate and measurement CVs.

Test Tubes

Manual/Semi-Automatic Sample Injection Mode

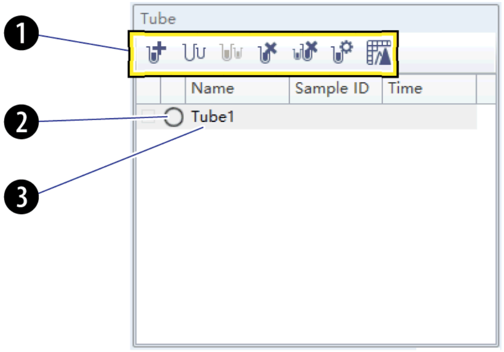
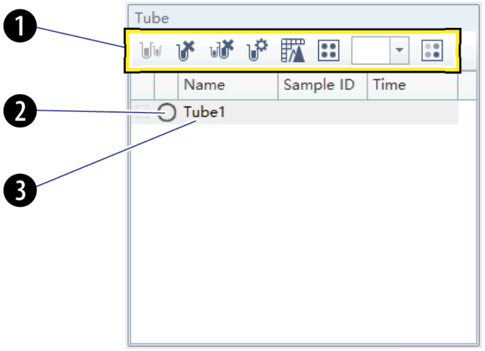
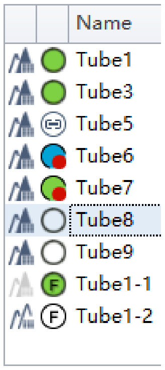


Plate Loader Sample Injection Mode



Test Tube Status



1. **Tube management controls.** Manages sample tubes. Used to add, copy, or delete attributes, and open the tube property.
2. **Test tube status indication.** Displays a colored symbol in front of each tube indicating the status of the tube processing.
   - ○ indicates that the tube data was not acquired.
   - ● indicates that the tube data was acquired by selecting **Run**, and the tube data can be overwritten.
   - ● indicates that the tube data was acquired by selecting **Record** or **Auto Record,** and the tube data cannot be overwritten.
   - Ⓕ indicates that the tube data was imported from FCS file.
   - Ⓕ indicates that the tube data was not acquired, and the unmixed data was imported from FCS file.
   - (!) indicates that the tube data imported from FCS file is damaged.
   - (!) indicates that the tube data was not acquired, and the unmixed data imported from FCS file is damaged.
   - ⊖ indicates that tube data was not acquired, and was linked to the Acquisition Catalog.
   - ⊖ indicates that the tube data was acquired by selecting **Run**, and was linked to the Acquisition Catalog.
   - ⊖ indicates that the tube data was acquired by selecting **Record** or **Auto Record**, and was linked to the Acquisition Catalog.
   - ● indicates that the tube data was acquired by selecting **Run**, and the tube data was saturated.

- ● indicates that the tube data was acquired by selecting **Record** or **Auto Record**, and the tube data was saturated.

NOTE In spectral unmixing experiments, ⚠ to the right of the test tube status indication symbol indicates that there is an unmixing gap.

NOTE Icons to the left of the test tube status indication symbol indicates the test tube's unmixing and fine adjustment status.

  - ☐ indicates that the tube data does not contain the unmixing matrix.
  - △ indicates that the unmixing matrix was not applied to the tube.
  - △ indicates that the unmixing matrix and fine adjustment matrix were not applied to the tube.
  - △ indicates that the unmixing matrix was applied to the tube, but the unmixing matrix is missing.
  - △ indicates that the unmixing matrix and fine adjustment matrix were applied to the tube, but the unmixing matrix is missing.
  - ▲ indicates that the unmixing matrix was applied to the tube.
  - ▲ indicates that the unmixing matrix and fine adjustment matrix were applied to the tube.

3. **Test tube list.** Displays the sample tubes used in the experiment. Right-click a tube in the list to perform additional operations.

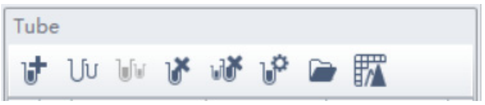
   NOTE In the Plate Loader Sample Injection mode the well number displays at the end of the tube name.

Test Tube Management Module

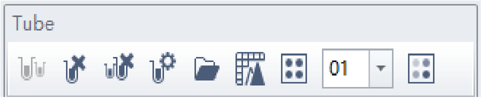
NOTE The 📂 is only available if the CytExpert for Spectral User Management software option is installed.

Select 📂 to open a previously created experiment.

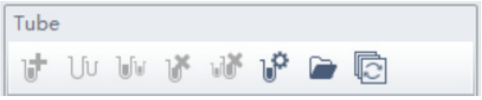
New Experiment Test Tube Management Module [Manual/Semi-Automatic Sample Injection Mode]



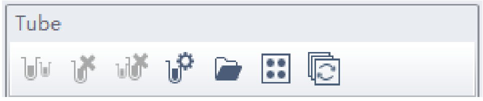
New Experiment Test Tube Management Module [Plate Loader Sample Injection Mode]



New Spectral Unmixing Experiment Test Tube Management Module [Manual/Semi-Automatic Sample Injection Mode]



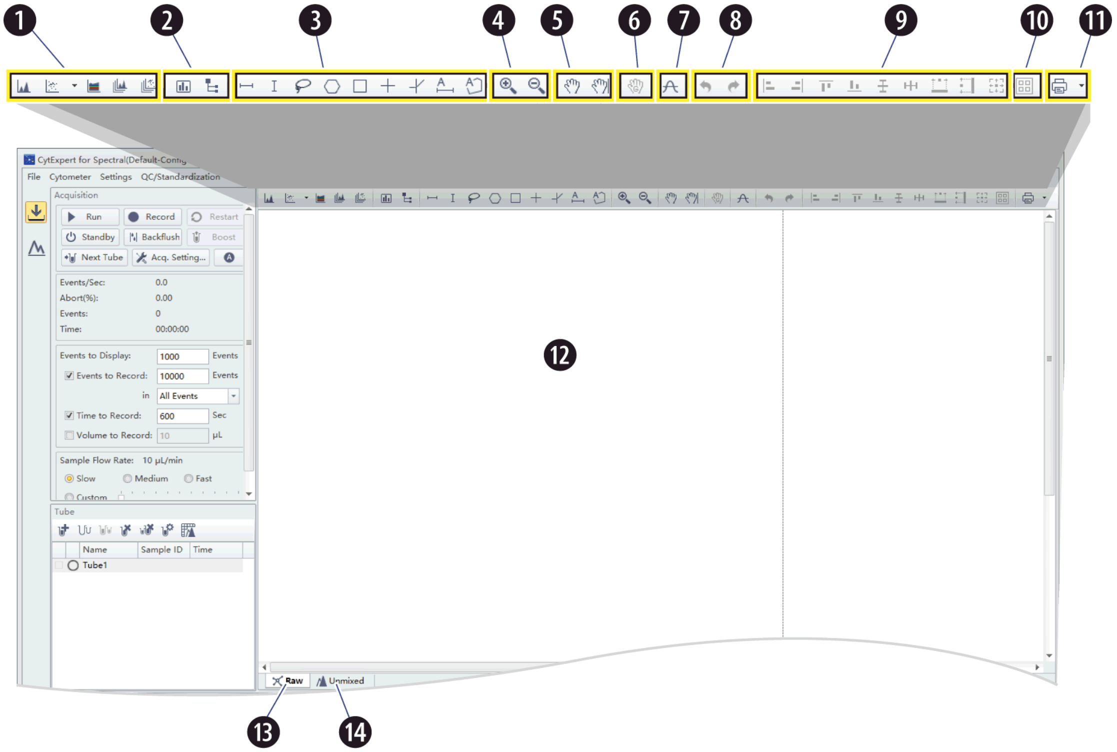
New Spectral Unmixing Experiment Test Tube Management Module [Plate Loader Sample Injection Mode]



Plot Area

The CytExpert for Spectral software supports two types of worksheets, Raw worksheet and Unmixed worksheet. Select the **Raw** or **Unmixed** tab in the bottom of the screen to switch between worksheets. Refer to Figure 2.1 and Figure 2.2.

Figure 2.1 Plot Area **[Raw Worksheet Shown]**



Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00
Events to Display: 1000 Events
☑ Events to Record: 10000 Events in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL
Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

Name	Sample ID	Time
Tube1		

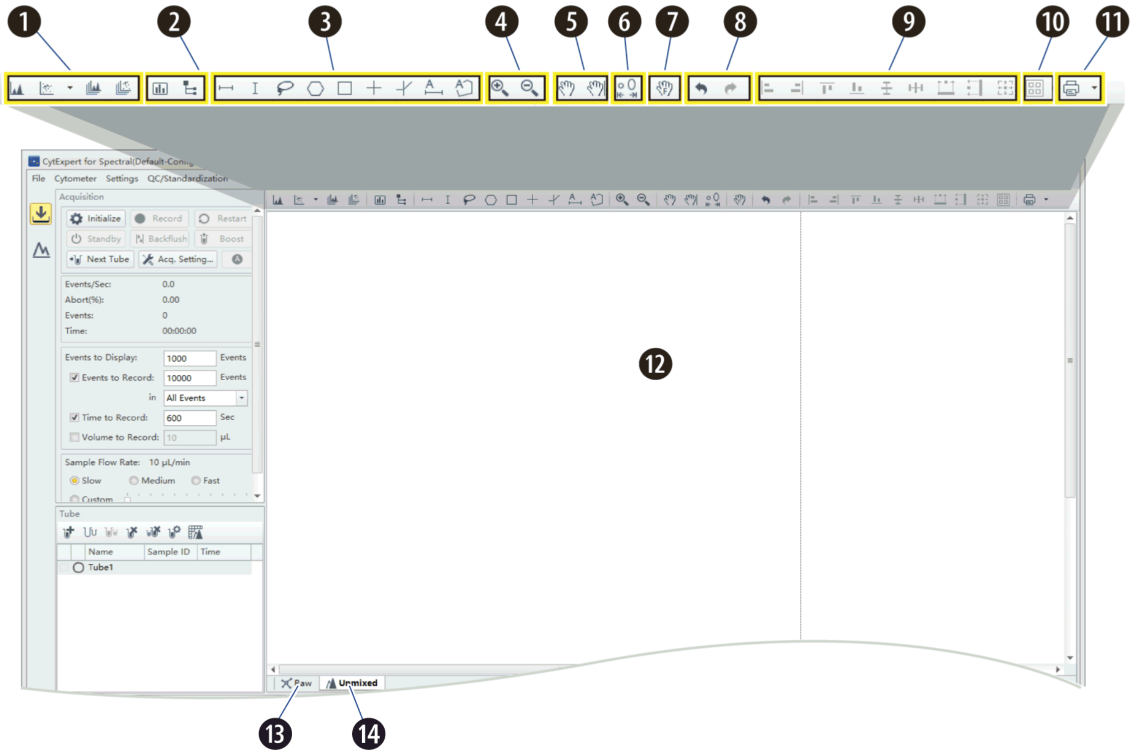
1. **Plot controls.** For creating single or multiple plots, such as dot plots, histograms, density plots, pseudo color plots, contour plots and spectrum plots.
2. **Statistics and hierarchy controls.** For creating statistical and hierarchical charts.
3. **Graphical gating controls.** For creating graphical gates.
4. **Zoom controls.** For zooming in and out within a plot.
5. **Pan axis display controls.** For scaling axis ranges in the plots.
6. **Gain adjustment control.** For increasing and lowering gain adjustments on the plots.

   NOTE The gain adjustment control only works when a sample is running.

7. **Threshold control.** For setting the minimum particle size limit, scatter value, or fluorescence intensity that acquisition will allow.

8. **Undo and redo controls.** For undoing or redoing an action in the drawing area.
9. **Display controls.** For controlling how plots and tables are aligned and arranged.
10. **Rearrange.** For restoring the plots to the default positions.
11. **Printing controls.** For printing and previewing the plot area.
12. **Plot area.** For creating plots and displaying statistics and hierarchy tables.
13. **Raw worksheet.** For editing and displaying raw data, and exporting or printing the results.
14. **Unmixed worksheet.** For editing and displaying unmixed data, and exporting or printing the results.

Figure 2.2 Plot Area **[Unmixed Worksheet Shown]**

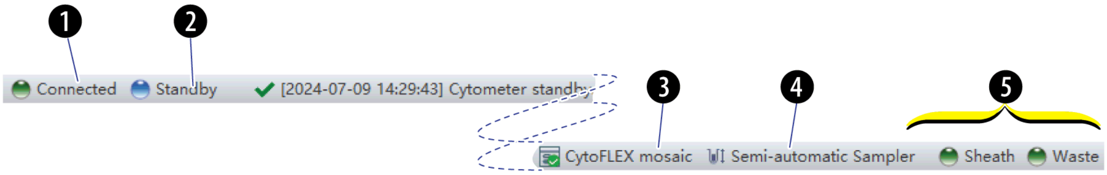


Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00
Events to Display: 1000 Events
☑ Events to Record: 10000 Events in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL
Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

1. **Plot controls.** For creating single or multiple plots, such as dot plots, histograms, density plots, pseudo color plots, and contour plots.
2. **Statistics and hierarchy controls.** For creating statistical and hierarchical charts.
3. **Graphical gating controls.** For creating graphical gates.
4. **Zoom controls.** For zooming in and out within a plot.
5. **Pan axis display controls.** For scaling axis ranges in the plots.
6. **Auto scaling control.** For scaling axis ranges in the plots automatically.
7. **Fine Adjustment control.** For using fine adjustment functions.
8. **Undo and redo controls.** For undoing or redoing an action in the drawing area.
9. **Display controls.** For controlling how plots and tables are aligned and arranged.
10. **Rearrange.** For restoring the plots to the default positions.
11. **Printing controls.** For printing and previewing the plot area.

12. **Plot area.** For creating plots and displaying statistics and hierarchy tables.
13. **Raw worksheet.** For editing and displaying raw data, and exporting or printing the results.
14. **Unmixed worksheet.** For editing and displaying unmixed data, and exporting or printing the results.

Status Bar



1. **Communication connection status.** Displays whether the Cytometer and the Workstation are connected.
2. **Instrument status information.** Displays the status of the Cytometer.
3. **CytoFLEX mosaic status information.** Displays the status of the CytoFLEX mosaic Spectral Detection Module.
4. **Sampler status.** Displays the sample injection mode state. There are two sample injection modes: Semi-automatic sample injection mode and manual sample injection mode.

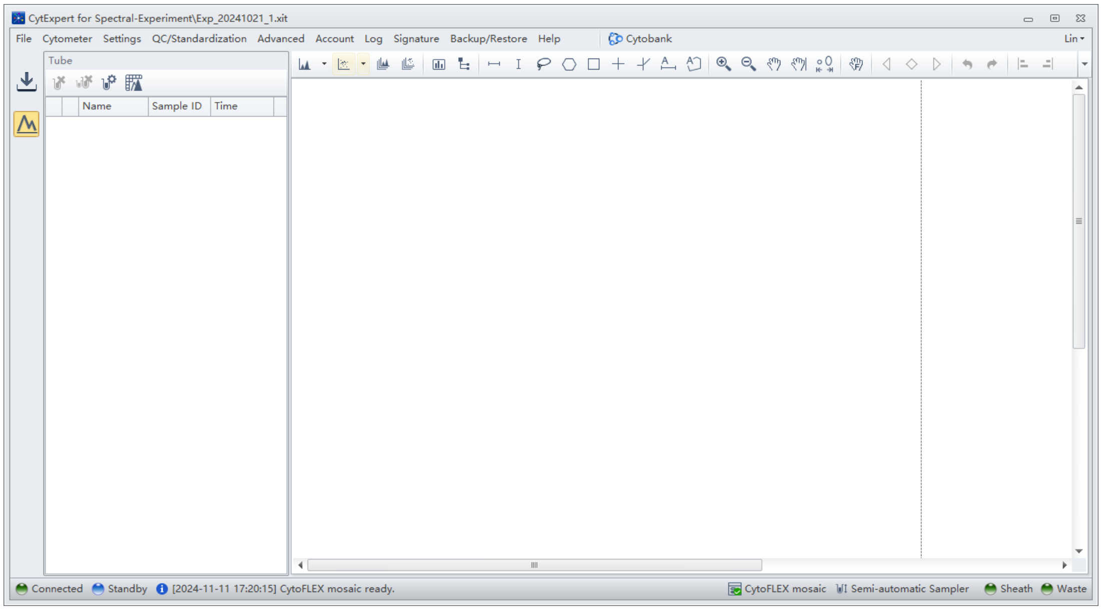
   NOTE CytoFLEX Cytometers equipped with a plate loader have three sample injection modes: Semi-automatic sample injection mode, manual sample injection mode, and plate loader sample injection mode.

5. **Fluid status information.** Displays the liquid level of the Fluid Containers/Cubitainers.

Analysis Screen

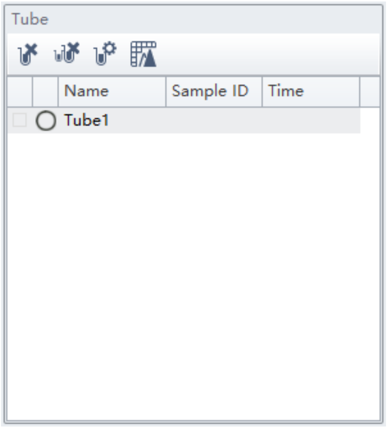
The Analysis screen is similar to the Acquisition screen, without the acquisition control modules.

NOTE Only the unmixed data can be analyzed in the analysis screen.



The Tube management module cannot add new sample tubes. Return to the Acquisition screen to add new sample tubes.

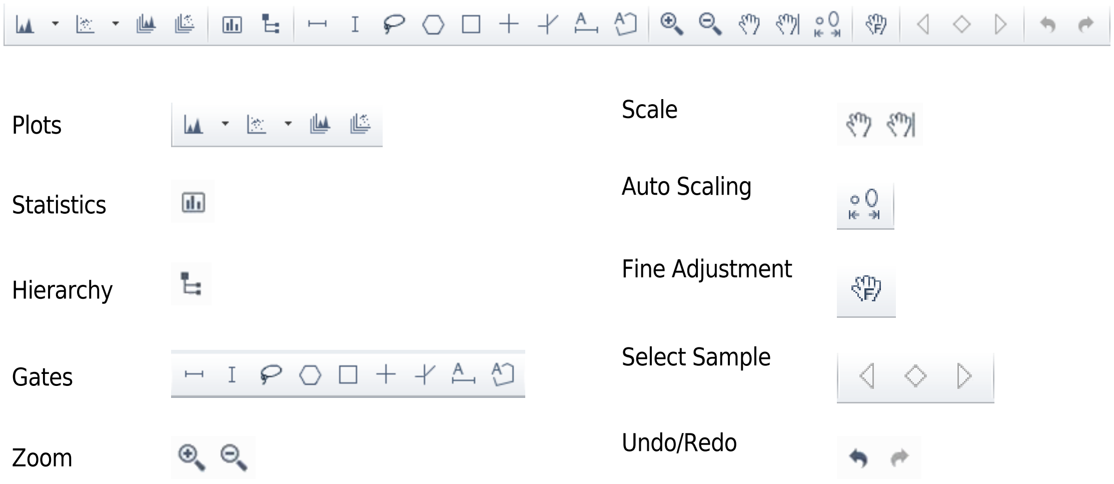
Tube Management



NOTE The 📂 is only available if you have the User Management software option installed. Select 📂 to open a previously created experiment.

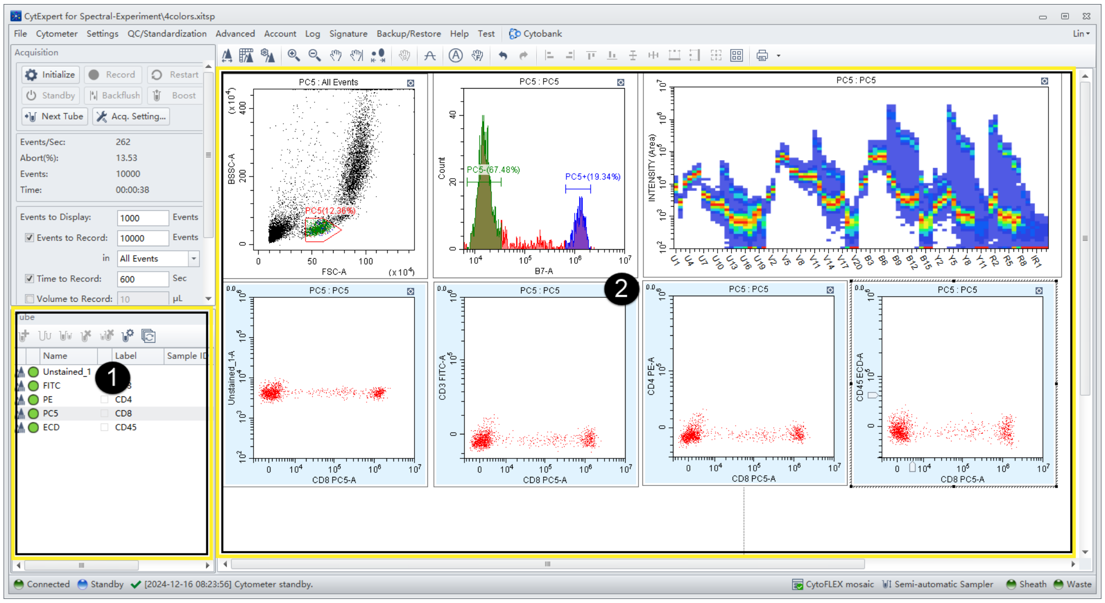
Drawing controls (see Figure 2.3) include the multi-data histograms and graphical display data controls.

Figure 2.3 Drawing Controls Toolbar (Top of Screen)



Spectral Unmixing Experiment Screen

The Spectral Unmixing Experiment screen appears when you open or create a new Spectral Unmixing experiment.


CytExpert for Spectral-Experiment\4colors.xitsp
File Cytometer Settings QC/Standardization Advanced Account Log Signature Backup/Restore Help Test Cytobank
Acquisition
Initialize Record Restart
Standby Backflush Boost
Next Tube Acq. Setting...
Tube
Name Label Sample ID
Unstained_1
FITC
PE CD4
PC5 CD8
ECD CD45
PC5 : All Events
PC5 : PC5
PC5(12.36%)
PC5-(67.48%)
PC5+(19.34%)
FSC-A
BSSC-A
Count
B7-A
INTENSITY (Area)
CD8 PC5-A
Unstained_1-A
CD3 FITC-A
CD4 PE-A
CD45 ECD-A
1
2
CytoFLEX mosaic Semi-automatic Sampler Sheath Waste

Events/Sec: 262
Abort(%): 13.53
Events: 10000
Time: 00:00:38
Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL
Connected Standby ✓ [2024-12-16 08:23:56] Cytometer standby.

1. **Tube management.** Displays sample tubes required for the spectral unmixing experiment.

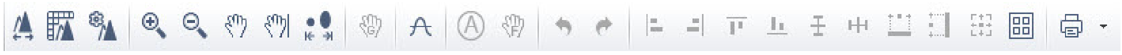
   NOTE The Tube management section of the screen can import saved data (.fcs) files for computational purposes.

2. **Plot area.** Displays plots and spectrum plot used for preview, and unmixing plots and gating.

   NOTE When the unmixing matrix is generated and applied, unmixing plots appear.

Spectral Unmixing Controls

The control area includes the spectral unmixing controls, coordinate pan axis display controls, gain adjustment controls, fine adjustment controls, and the undo and redo controls. The spectral unmixing controls gives you the option of calculating the spectral unmixing value, displaying the spectral unmixing matrix, or changing the spectral unmixing parameters.



Spectral Unmixing		Adjust Gain	
Unmixing Matrix		Threshold	
Spectral Unmixing Setup		Auto Fine Adjustment	Ⓐ
Zoom		Fine Adjustment	
Pan		Undo/Redo	↶ ↷
Single Side Pan		Alignment	
Auto Scaling		Print	

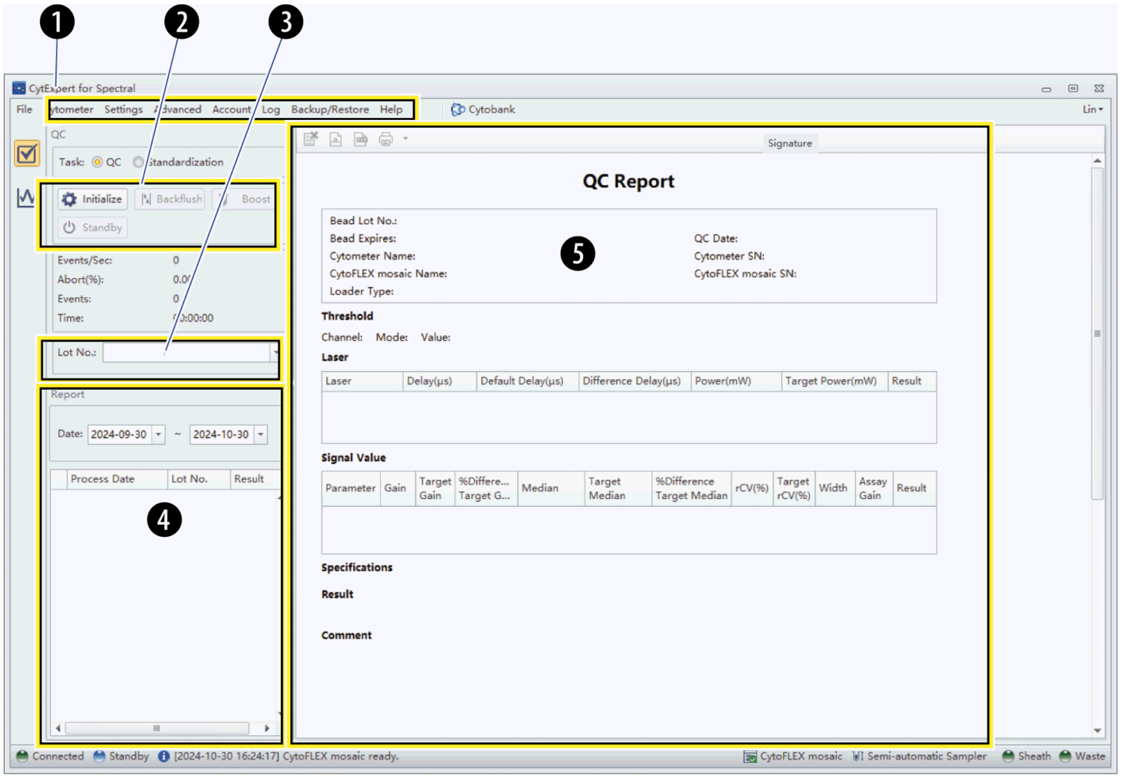
QC Experiment Screen

The Quality Control (QC) Experiment screen appears when you access a QC experiment.

QC Report Screen

Before starting the QC routine, a Settings screen appears.

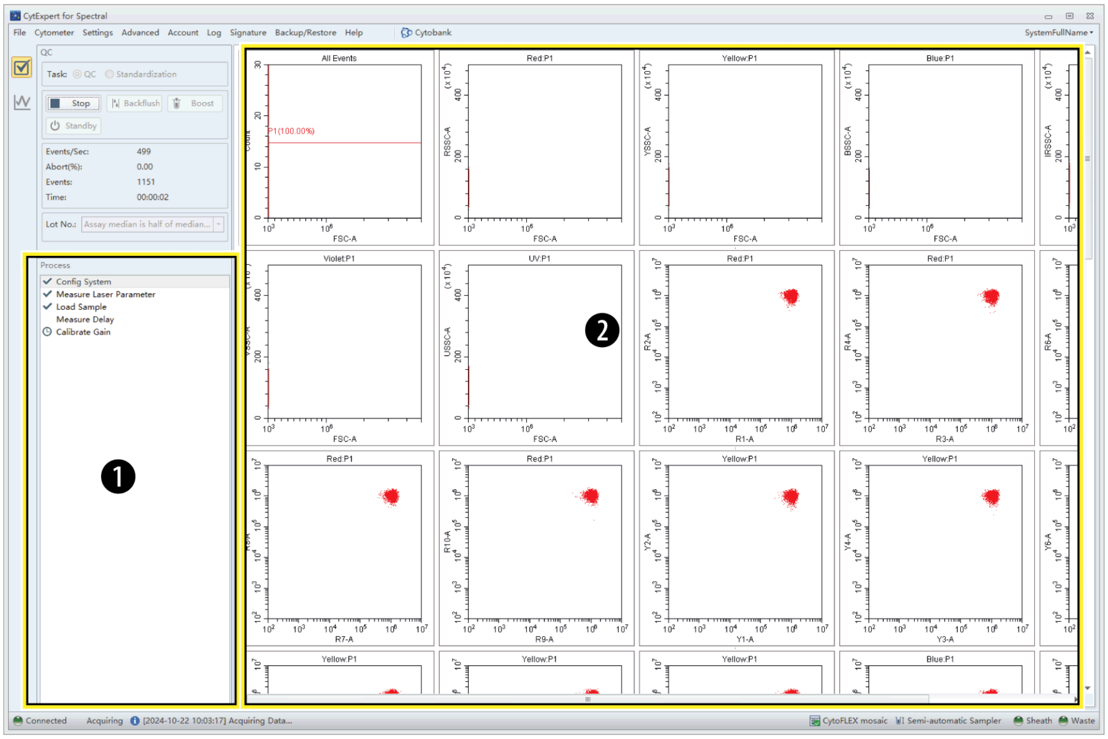
Figure 2.4 QC Report Screen



1. **Menu.** Allows you to configure settings related to QC experiments.
2. **Acquisition control.** Controls sample loading/unloading and data recording.
3. **Lot selection.** Allows you to select the lot number of the QC reagent.
4. **QC results list area.** Displays the time and results of completed QC runs.
5. **QC reports area.** Displays detailed reports for the selected QC experiment.

QC Experiment Screen

When acquiring QC samples, the software opens the QC screen.



Task: ◉ QC ○ Standardization

Events/Sec: 499
Abort(%): 0.00
Events: 1151
Time: 00:00:02

Lot No.: Assay median is half of median...

- ✓ Config System
- ✓ Measure Laser Parameter
- ✓ Load Sample
- Measure Delay
- Calibrate Gain

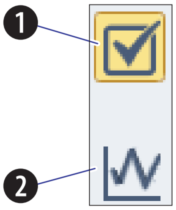
Connected Acquiring [2024-10-22 10:03:17] Acquiring Data...

CytoFLEX mosaic Semi-automatic Sampler Sheath Waste

1. **QC experiment progress indicator.** Displays the QC stage.
2. **Plot area.** Displays the QC plots.

QC Screen Navigation

The Analysis screens have two navigation icons, one for the QC screen and the other for the Levey-Jennings (LJ) charts. Refer to CHAPTER 5, Creating Levey-Jennings Charts.

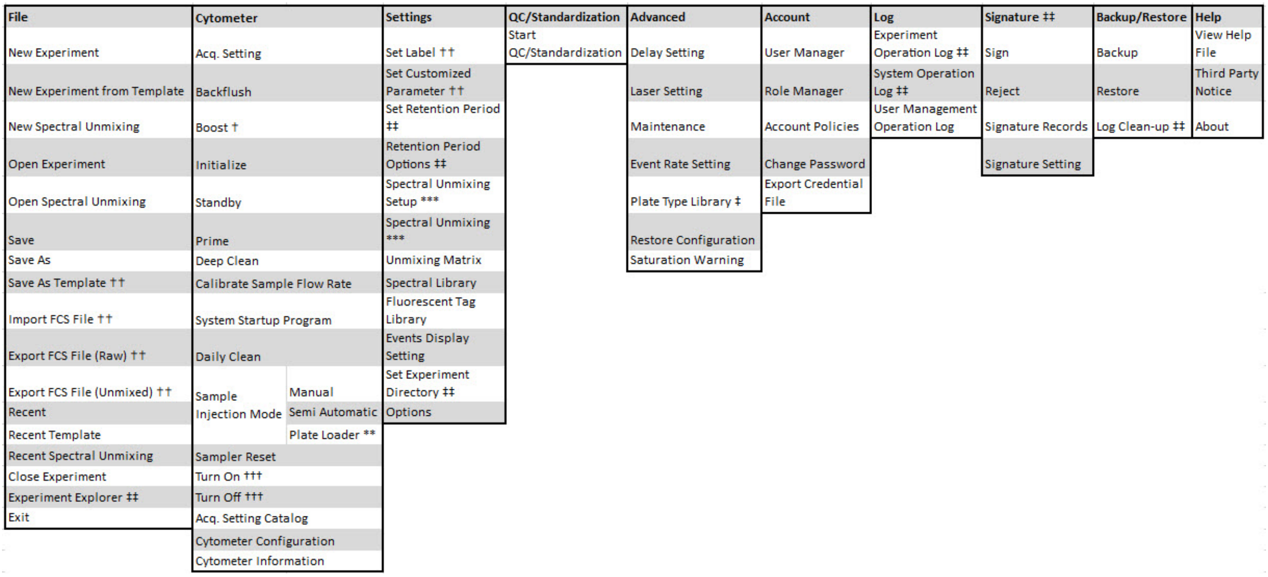


1. **QC screen icon.** Accesses the QC screen.
2. **LJ screen icon.** Accesses the Levey-Jennings (LJ) screen.

Software Menu

The CytExpert for Spectral software contains the following selectable menu items:

Figure 2.5 Software Menu Tree*



File
- New Experiment
- New Experiment from Template
- New Spectral Unmixing
- Open Experiment
- Open Spectral Unmixing
- Save
- Save As
- Save As Template ††
- Import FCS File ††
- Export FCS File (Raw) ††
- Export FCS File (Unmixed) ††
- Recent
- Recent Template
- Recent Spectral Unmixing
- Close Experiment
- Experiment Explorer ‡‡
- Exit

Cytometer
- Acq. Setting
- Backflush
- Boost †
- Initialize
- Standby
- Prime
- Deep Clean
- Calibrate Sample Flow Rate
- System Startup Program
- Daily Clean
- Sample Injection Mode
  - Manual
  - Semi Automatic
  - Plate Loader **
- Sampler Reset
- Turn On †††
- Turn Off †††
- Acq. Setting Catalog
- Cytometer Configuration
- Cytometer Information

Settings
- Set Label ††
- Set Customized Parameter ††
- Set Retention Period ‡‡
- Retention Period Options ‡‡
- Spectral Unmixing Setup ***
- Spectral Unmixing ***
- Unmixing Matrix
- Spectral Library
- Fluorescent Tag Library
- Events Display Setting
- Set Experiment Directory ‡‡
- Options

QC/Standardization
- Start QC/Standardization

Advanced
- Delay Setting
- Laser Setting
- Maintenance
- Event Rate Setting
- Plate Type Library ‡
- Restore Configuration
- Saturation Warning

Account
- User Manager
- Role Manager
- Account Policies
- Change Password
- Export Credential File

Log
- Experiment Operation Log ‡‡
- System Operation Log ‡‡
- User Management Operation Log

Signature ‡‡
- Sign
- Reject
- Signature Records
- Signature Setting

Backup/Restore
- Backup
- Restore
- Log Clean-up ‡‡

Help
- View Help File
- Third Party Notice
- About

* The menu options for **File**, **Cytometer**, **Settings**, and **QC/Standardization** change when you select Start QC/ Standardization. Refer to Figure 2.6.

† **Boost** is only active in the Manual Sample Injection mode.

‡ **Plate Type Library** is only an option if the Plate Loader module is installed and the Plate Loader Sample Injection mode is selected.

** **Plate Loader** is only an option if the Plate Loader module is installed.

*** Only available when the spectral unmixing experiment is open.

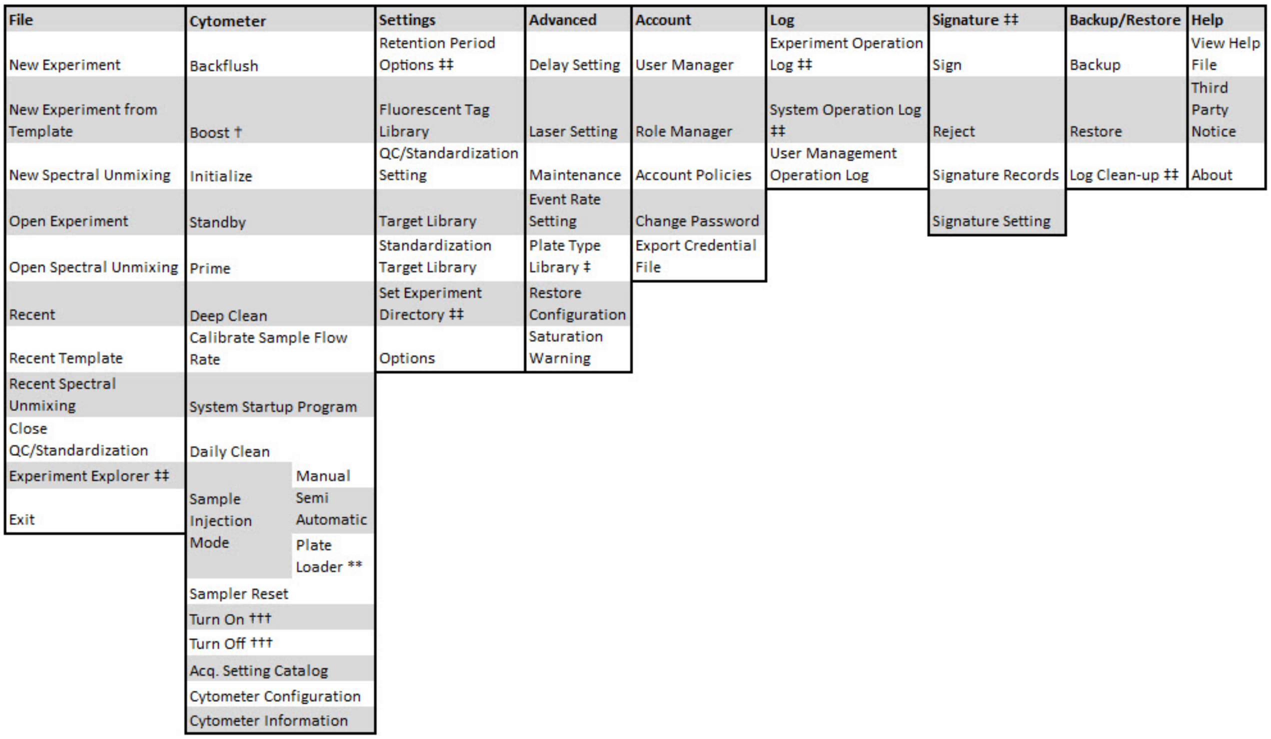
†† Only available when a standard experiment is open.

‡‡ These options are only available if the CytExpert for Spectral Electronic Record Management software option is installed.

††† These options are only available on the CytoFLEX LX flow cytometer.

The Cytobank icon is hyper linked to the Cytobank spotlight page where you can login to the Cytobank Premium server, request a 30-day free trial and access additional information. The Cytobank platform allows you to analyze, manage, and securely share flow cytometry data. FCS files can be uploaded to the platform with the related attachments such as PDF files.

Figure 2.6 QC Software Menu Tree



File	Cytometer	Settings	Advanced	Account	Log	Signature ‡‡	Backup/Restore	Help
New Experiment	Backflush	Retention Period Options ‡‡	Delay Setting	User Manager	Experiment Operation Log ‡‡	Sign	Backup	View Help File
New Experiment from Template	Boost †	Fluorescent Tag Library	Laser Setting	Role Manager	System Operation Log ‡‡	Reject	Restore	Third Party Notice
New Spectral Unmixing	Initialize	QC/Standardization Setting	Maintenance	Account Policies	User Management Operation Log	Signature Records	Log Clean-up ‡‡	About
Open Experiment	Standby	Target Library	Event Rate Setting	Change Password		Signature Setting		
Open Spectral Unmixing	Prime	Standardization Target Library	Plate Type Library ‡	Export Credential File				
Recent	Deep Clean	Set Experiment Directory ‡‡	Restore Configuration					
Recent Template	Calibrate Sample Flow Rate	Options	Saturation Warning					
Recent Spectral Unmixing	System Startup Program							
Close QC/Standardization	Daily Clean							
Experiment Explorer ‡‡	Sample Injection Mode: Manual Semi Automatic Plate Loader **							
Exit	Sampler Reset							
	Turn On †††							
	Turn Off †††							
	Acq. Setting Catalog							
	Cytometer Configuration							
	Cytometer Information							

† **Boost** is only active in the Manual Sample Injection mode.

‡ **Plate Type Library** is only an option if the Plate Loader module is installed and the Plate Loader Sample Injection mode is selected.

** **Plate Loader** is only an option if the Plate Loader module is installed.

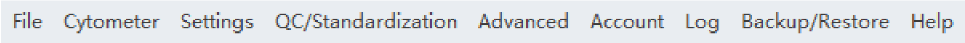
‡‡ These options are only available if the CytExpert for Spectral Electronic Record Management software option is installed.

††† These options are only available on the CytoFLEX LX flow cytometer.

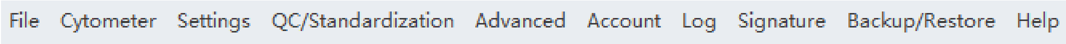
The Cytobank icon is hyper linked to the Cytobank spotlight page where you can login to the Cytobank Premium server, request a 30-day free trial and access additional information. The Cytobank platform allows you to analyze, manage, and securely share flow cytometry data. FCS files can be uploaded to the platform with the related attachments such as PDF files.

Acquisition and Analysis Screen Menu

CytExpert for Spectral - User Management



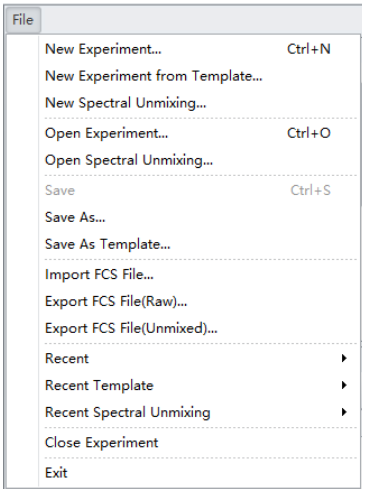
CytExpert for Spectral - Electronic Record Management



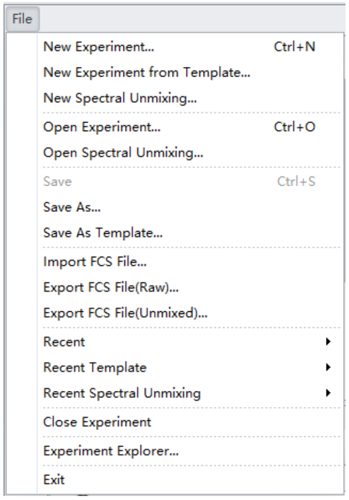
File Menu

For creating new experiments, opening existing experiments, saving new experiments and data, and importing/exporting FCS data files.

User Management



Electronic Record Management

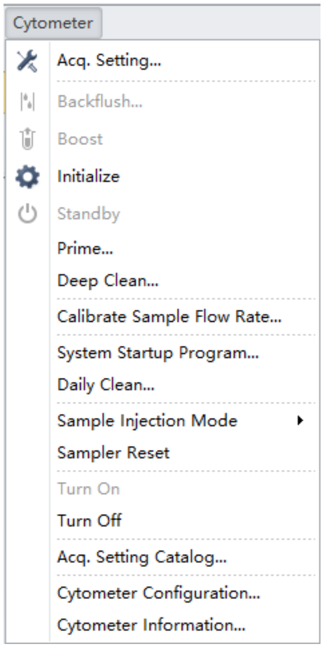


Cytometer Menu

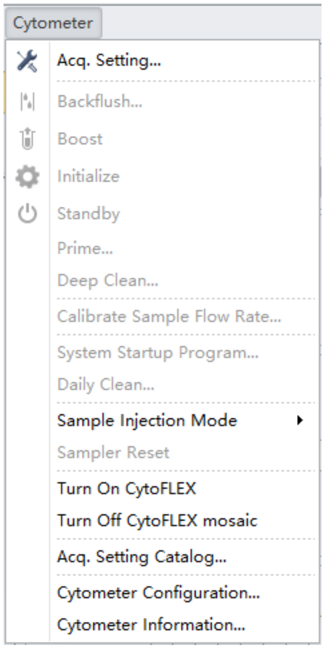
For configuring Cytometer settings and controlling Cytometer functions. Depending on the Cytometer state, certain functions may not be available.

NOTE The **Turn On**, **Turn Off**, **Turn On CytoFLEX mosaic** and **Turn Off CytoFLEX** selections are only available on the CytoFLEX LX.

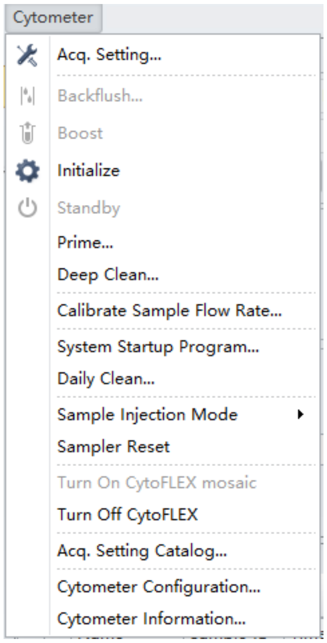
Cytometer On and CytoFLEX mosaic Spectral Detection Module On



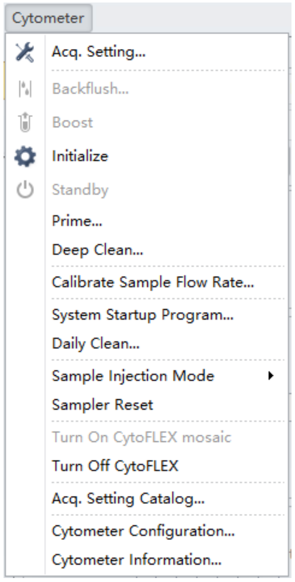
Cytometer On and CytoFLEX mosaic Spectral Detection Module Off



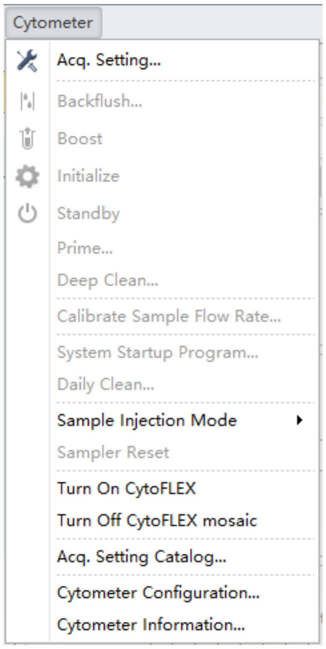
Cytometer On and CytoFLEX mosaic Spectral Detection Module Power Off



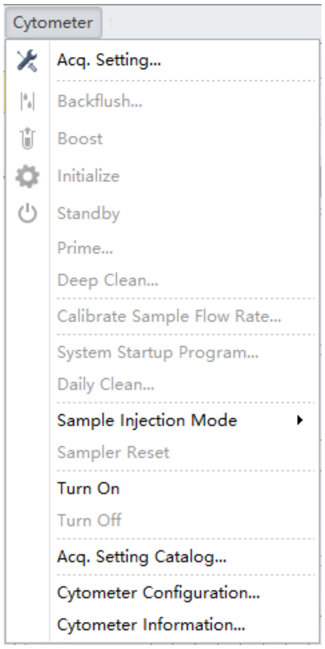
NOTE If the software restarts, the **Turn On CytoFLEX mosaic** selection is not available when the Cytometer is on and the CytoFLEX mosaic Spectral Detection Module is off.



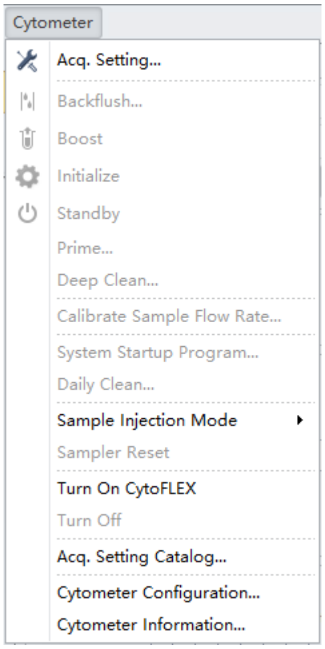
Cytometer Off and CytoFLEX mosaic Spectral Detection Module On



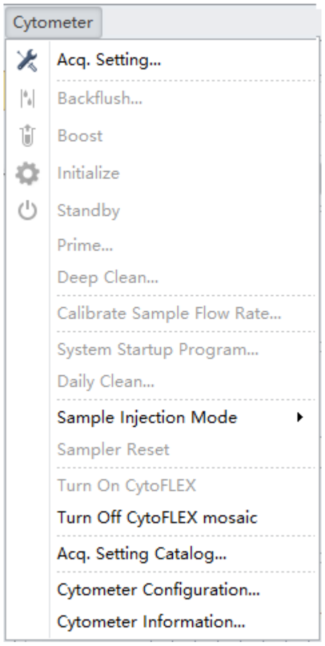
Cytometer Off and CytoFLEX mosaic Spectral Detection Module Off



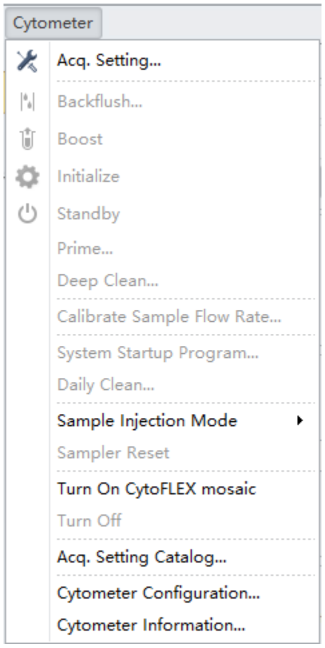
Cytometer Off and CytoFLEX mosaic Spectral Detection Module Power Off



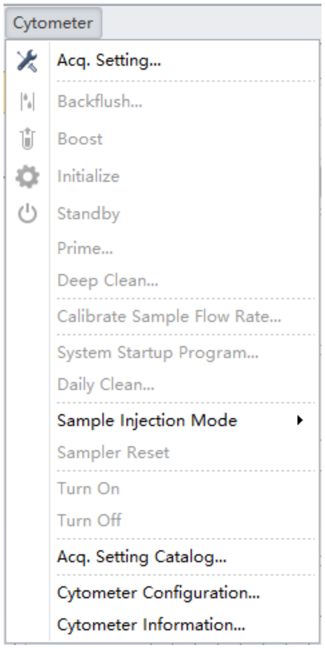
Cytometer Power Off and CytoFLEX mosaic Spectral Detection Module On



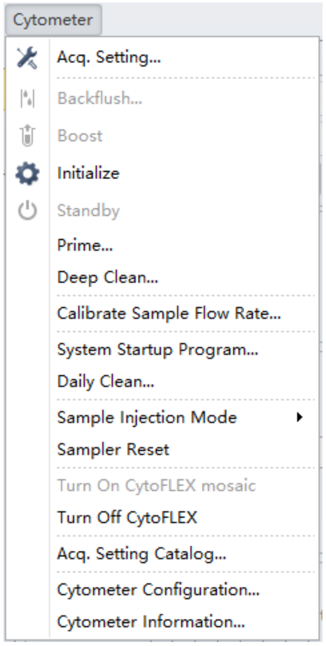
Cytometer Power Off and CytoFLEX mosaic Spectral Detection Module Off



Cytometer Power Off and CytoFLEX mosaic Spectral Detection Module Power Off



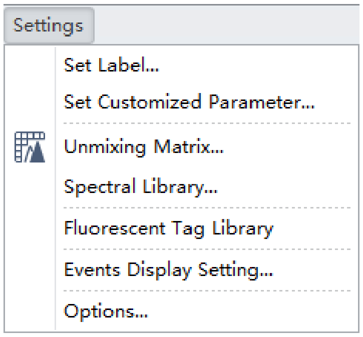
NOTE If the software restarts, the **Turn On CytoFLEX mosaic** selection is not available when the Cytometer is power off and the CytoFLEX mosaic Spectral Detection Module is turned off.



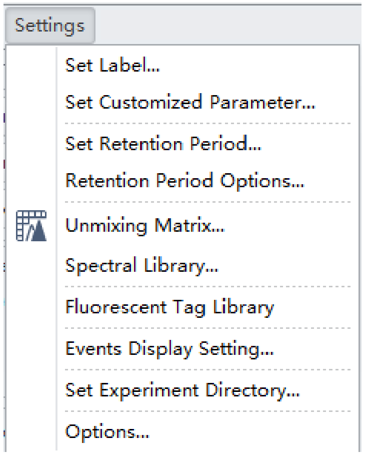
Settings Menu

Used to select and/or change software options and settings.

User Management

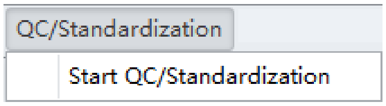


Electronic Record Management



QC/Standardization Menu

Select **Start QC/Standardization** from the QC/Standardization menu to start the QC routine.

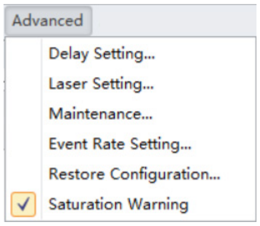


NOTE The QC/Standardization menu is the same for the CytExpert for Spectral User Management and the CytExpert for Spectral Electronic Record Management software options.

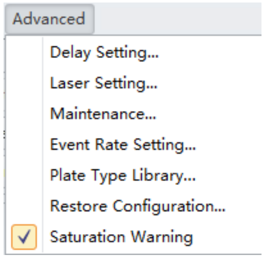
Advanced Menu

Used to access advanced settings for experienced users. Includes laser time delay settings.

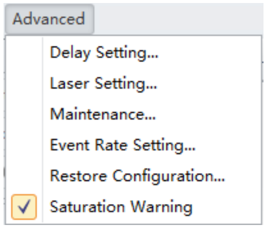
User Management - Semi-Automatic/Manual Sample Injection Mode



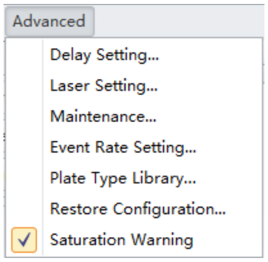
User Management - Plate Loader Sample Injection Mode



Electronic Record Management - Semi-Automatic/Manual Sample Injection Mode



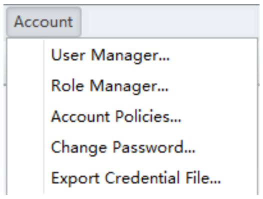
Electronic Record Management - Plate Loader Sample Injection Mode



NOTE To restore the Cytometer and CytoFLEX mosaic Spectral Detection Module configuration file, refer to APPENDIX A, Restoring the Cytometer and CytoFLEX mosaic Spectral Detection Module Configuration File.

Account Menu

Used to for user account management settings.

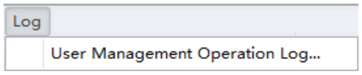


NOTE The Account menu is the same for the CytExpert for Spectral User Management and the CytExpert for Spectral Electronic Record Management software options.

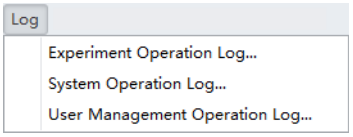
Log Menu

Used to access logs including the Experiment Operation Log, the System Operation Log, and the User Management Operation Log.

User Management

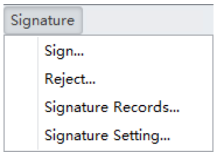


Electronic Record Management



Signature Menu

Used to sign experiment and view signature details.

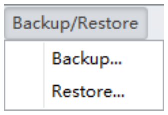


NOTE The Signature menu is only available in the CytExpert for Spectral Electronic Record Management software option.

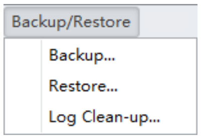
Backup/Restore Menu

Used to backup/restore databases. Refer to CHAPTER 10, Backup and Restore.

User Management

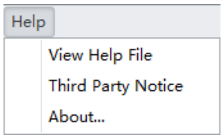


Electronic Record Management



Help Menu

For displaying software version information and system Instructions for Use.



NOTE The Help menu is the same for the CytExpert for Spectral User Management and CytExpert for Spectral Electronic Record Management software options.

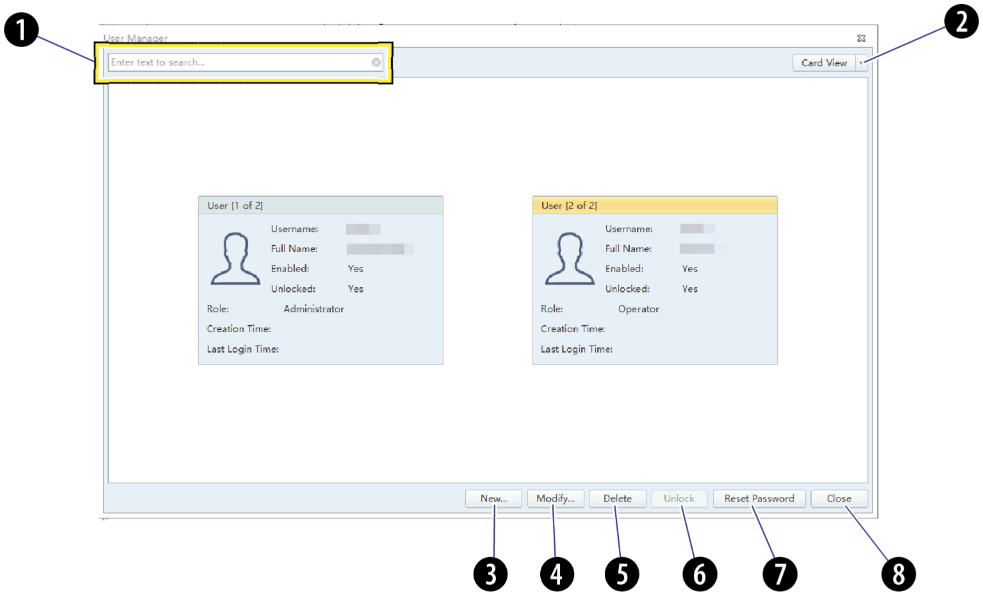
User Management

IMPORTANT Only the Initial Administrator or authorized users can manage users.

User Management is used to create and manage user accounts.

Select **Account** > **User Manager**. The User Manager window appears.

Figure 2.7 User Manager (Card View)



User [1 of 2]
Username: [redacted]
Full Name: [redacted]
Enabled: Yes
Unlocked: Yes
Role: Administrator
Creation Time:
Last Login Time:

User [2 of 2]
Username: [redacted]
Full Name: [redacted]
Enabled: Yes
Unlocked: Yes
Role: Operator
Creation Time:
Last Login Time:

Figure 2.8 User Manager (Grid View)

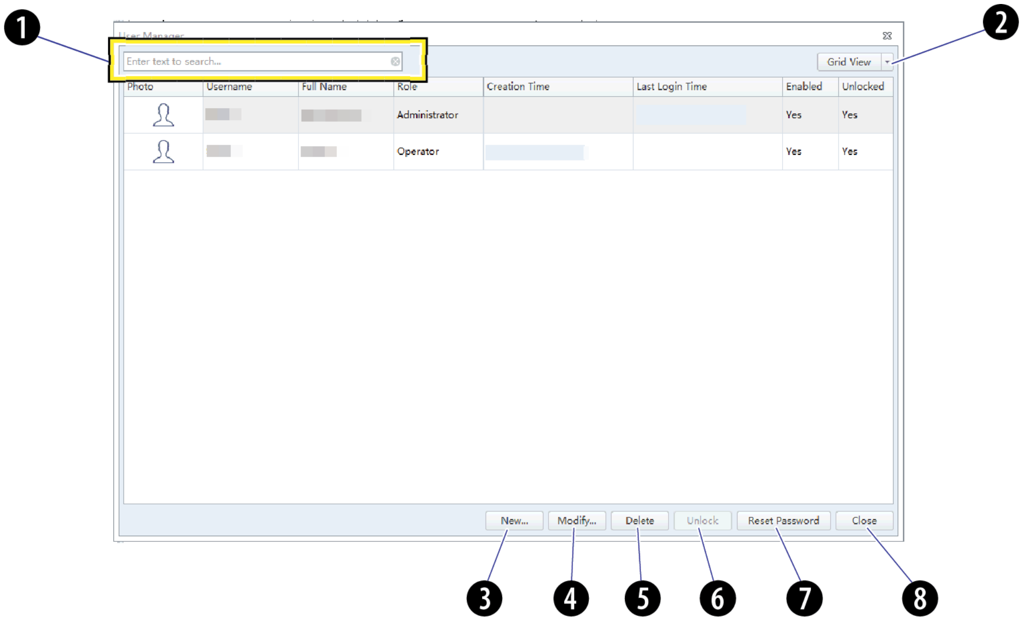


Photo	Username	Full Name	Role	Creation Time	Last Login Time	Enabled	Unlocked
	[redacted]	[redacted]	Administrator		[redacted]	Yes	Yes
	[redacted]	[redacted]	Operator	[redacted]		Yes	Yes

1. **Search text box.** Filters users by username and display name.

2. **View drop-down.** Toggles between Card View (see Figure 2.7) and Grid View (see Figure 2.8).
3. **New.** Used to create a new user profile.
4. **Modify.** Used to modify an existing user profile.
5. **Delete.** Used to delete an existing user profile.
6. **Unlock.** Used to unlock an existing account that has been locked.

   NOTE An account locks after 3 failed password attempts. The number of attempts can be changed by the administrator. Refer to Account Policies.

   NOTE An account automatically unlocks after 30 minutes. The duration can be changed by the administrator. Refer to Account Policies.

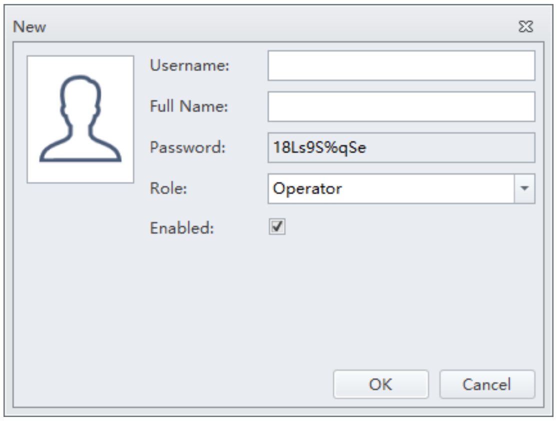
7. **Reset password.** Used to reset an existing user password to a random password.
8. **Close.** Closes the User Manager window.

Creating, Deleting, and Modifying Users in User Manager

The initial system administrator indicating with in the username cannot be deleted or modified.

Creating a New User in User Manager

1 Select **New** in the User Manager window. The New window appears.



New

Username:

Full Name:

Password: 18Ls9S%qSe

Role: Operator

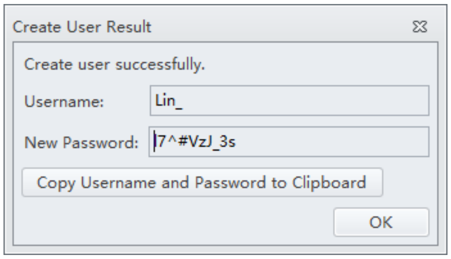
Enabled: ☑

2 Fill in the new user information.

a. Enter the Username.
b. Enter the Full Name.
c. Select the User Role.
d. Select the Enabled checkbox to enable the user.

   NOTE The Enabled checkbox can only be changed by an administrator.

3 Select **OK**. The Create User Result window appears.



Create user successfully.

Username: Lin_

New Password: 7^#VzJ_3s

NOTE Select Copy Username and Password to Clipboard to copy the username and password to a clipboard and inform the user of the change. The user is required to change the new password immediately upon the initial login.

4 Select **OK**.

5 Select **Close**.

Deleting Users in User Manager

IMPORTANT If an account has been used and log information has been generated related to it, the account cannot be deleted, but it can be disabled.

1 Select the user to be deleted in the User Manager window then select **Delete**.

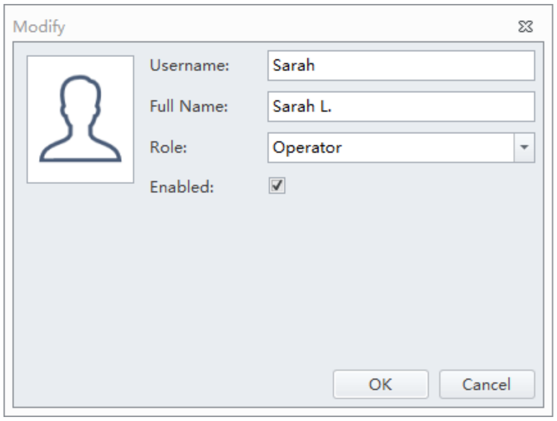
2 The following message appears: *Are you sure to you want to delete the user?* Select **Yes** to confirm.

3 Select **Close** to exit the User Manager window.

Modifying Users in User Manager

IMPORTANT If an account has been used and log information has been generated related to it, the username cannot be modified.

1 Select **Modify** in the User Manager window. The Modify window appears.



Username: Sarah
Full Name: Sarah L.
Role: Operator
Enabled: ☑

NOTE The Initial System Administrator is a system default user and you can only modify its Full Name.

2 Modify the user information as necessary.

NOTE Uncheck the enabled box to disable a user.

3 Select **OK**.

4 Select **Close** to exit the User Manager window.

Unlocking a User Account

Select a *Locked* user in the User Manager window and select **Unlock**.

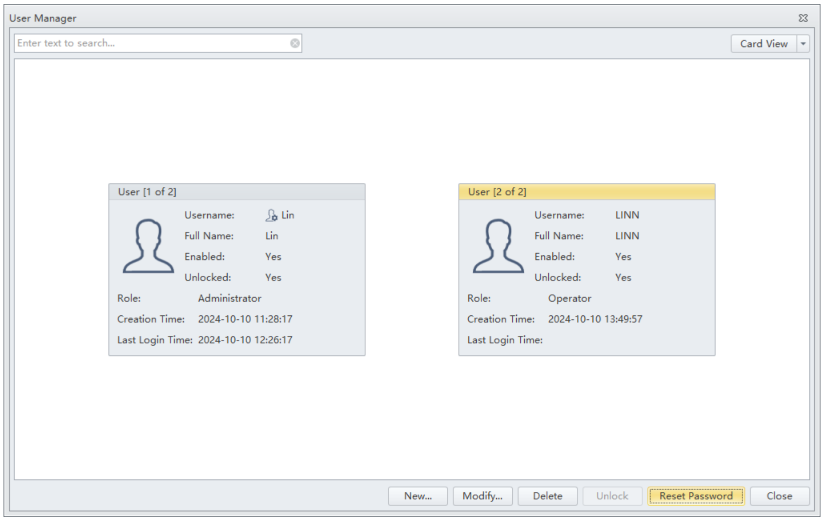
NOTE You cannot unlock an active user.

Resetting a User Password

IMPORTANT Only the administrator or authorized users can reset a password.

---

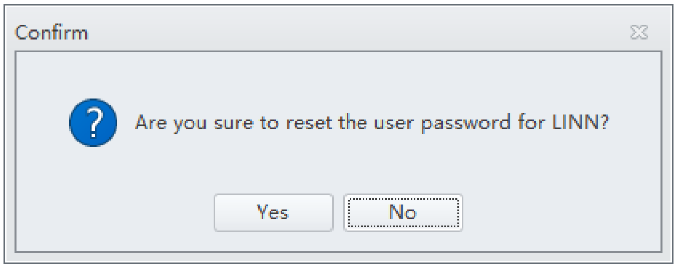
1 Select the user to be reset in the User Manager window and select **Reset Password.**



User [1 of 2]
Username: Lin
Full Name: Lin
Enabled: Yes
Unlocked: Yes
Role: Administrator
Creation Time: 2024-10-10 11:28:17
Last Login Time: 2024-10-10 12:26:17

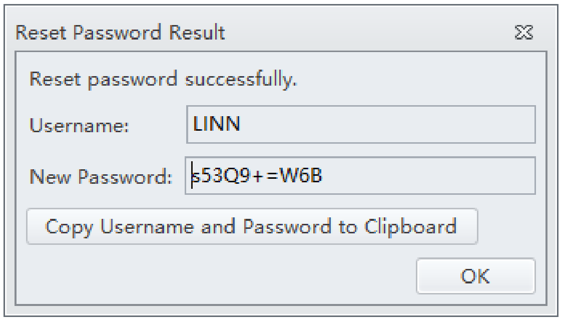
User [2 of 2]
Username: LINN
Full Name: LINN
Enabled: Yes
Unlocked: Yes
Role: Operator
Creation Time: 2024-10-10 13:49:57
Last Login Time:

The confirm window appears.



Are you sure to reset the user password for LINN?

2 Select **Yes**. The Reset Password Result window appears. A new random password is automatically generated.


Reset Password Result

Reset password successfully.

Username: LINN

New Password: s53Q9+=W6B

NOTE Select Copy Username and Password to Clipboard to copy the username and password to a clipboard and inform the user of the change. The user is required to change the new password immediately upon the initial login.

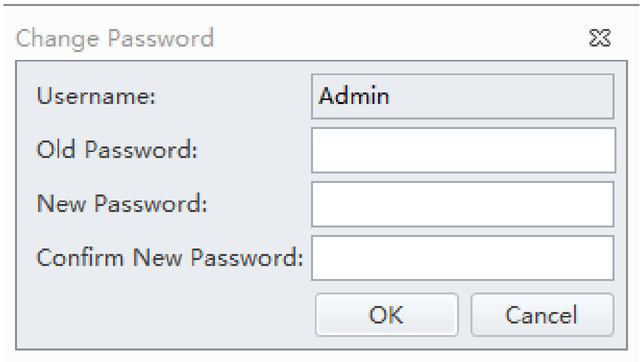
3 Select **OK**.

4 Select **Close** to exit the User Manager window.

Changing a User Password

Beckman Coulter recommends changing your password on a regular basis.

1 Select **Account > Change Password**. The Change Password window appears.


Change Password

Username: Admin

Old Password:

New Password:

Confirm New Password:

2 Enter the current password, the new password, and confirm the new password.

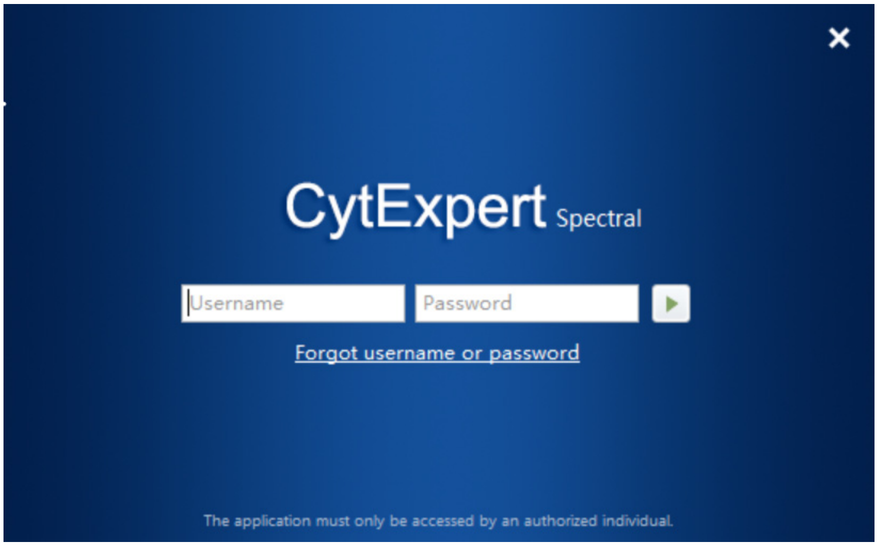
3 Select **OK**.

Forgot Username or Password

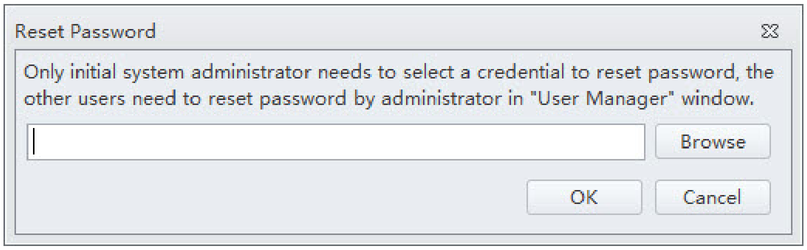
The Forgot username or password function allows only the initial system administrator to reset a password using the system administrator credential file. If you are a common user and forget your password, contact the administrator to reset a password for you. For instructions, refer to Resetting a User Password.

IMPORTANT The initial system administrator password cannot be reset without the system administrator credential file. Beckman Coulter is not responsible for and will not be able to recover your initial system administrator account if the system administrator password is forgotten and the credential file is lost. For instructions on exporting the credential file, refer to Exporting the Credential File.

1 Select **Forgot username or password** on the login window.


CytExpert Spectral
Username
Password
Forgot username or password
The application must only be accessed by an authorized individual.

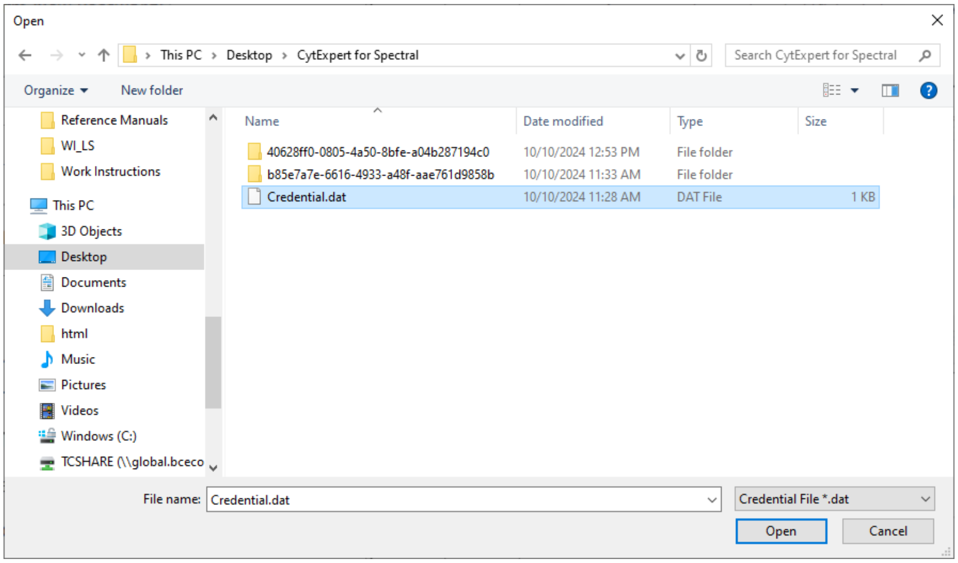
The Reset Password window appears.



Only initial system administrator needs to select a credential to reset password, the other users need to reset password by administrator in "User Manager" window.

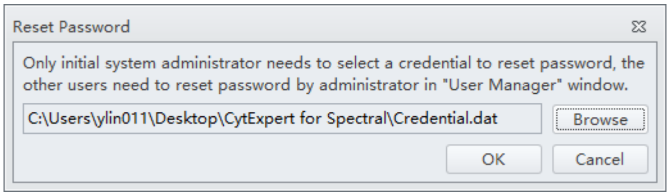
IMPORTANT Find the system administrator credential file from the backup folder when installing the CytExpert or Spectral software. You can also export the credential file by selecting **Export Credential File** from the **Account** menu if you are logged in. Refer to Exporting the Credential File.

2 Navigate to the system administrator credential file and select **Open**.



Name	Date modified	Type	Size
40628ff0-0805-4a50-8bfe-a04b287194c0	10/10/2024 12:53 PM	File folder	
b85e7a7e-6616-4933-a48f-aae761d9858b	10/10/2024 11:33 AM	File folder	
Credential.dat	10/10/2024 11:28 AM	DAT File	1 KB

File name: Credential.dat
Credential File *.dat

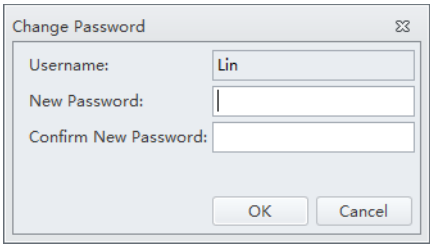


Only initial system administrator needs to select a credential to reset password, the other users need to reset password by administrator in "User Manager" window.

C:\Users\ylin011\Desktop\CytExpert for Spectral\Credential.dat

---

3 Select **OK**. The Change Password window appears.



Username: Lin
New Password:
Confirm New Password:

4 Enter the new password, and confirm the new password.

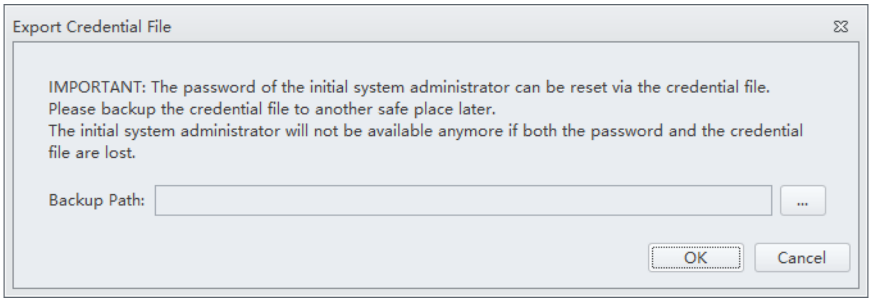
NOTE The new password must contain at least ten digits and at least three of the following character types by default:

- Uppercase characters of European languages (A through Z, with diacritic marks, Greek and Cyrillic characters);
- Lowercase characters of European languages (a through z, sharp-s, with diacritic marks, Greek and Cyrillic characters);
- Base 10 digits (0 through 9);
- Non-alphabetic characters (for example, !, $, #, %);
- Any Unicode character recognized as an alphabetic character but not uppercase or lowercase.

5 Select **OK**.

Exporting the Credential File

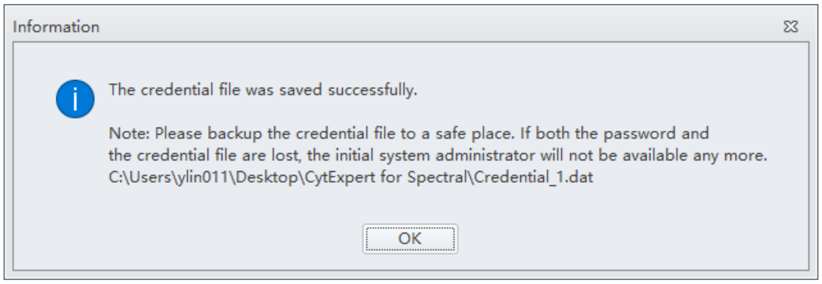
1 Select **Account** > **Export Credential File**. The Export Credential File window appears.



IMPORTANT: The password of the initial system administrator can be reset via the credential file. Please backup the credential file to another safe place later.
The initial system administrator will not be available anymore if both the password and the credential file are lost.

2 Select ... and browse to the desired backup directory to export the credential file.

3 Select **OK**. The following system prompt appears.



The credential file was saved successfully.

Note: Please backup the credential file to a safe place. If both the password and the credential file are lost, the initial system administrator will not be available any more.
C:\Users\ylin011\Desktop\CytExpert for Spectral\Credential_1.dat

4 Select **OK.**

Role Management

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Role Management.

Creating, Deleting, and Modifying User Roles in Role Manager

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Creating, Deleting, and Modifying User Roles in Role Manager.

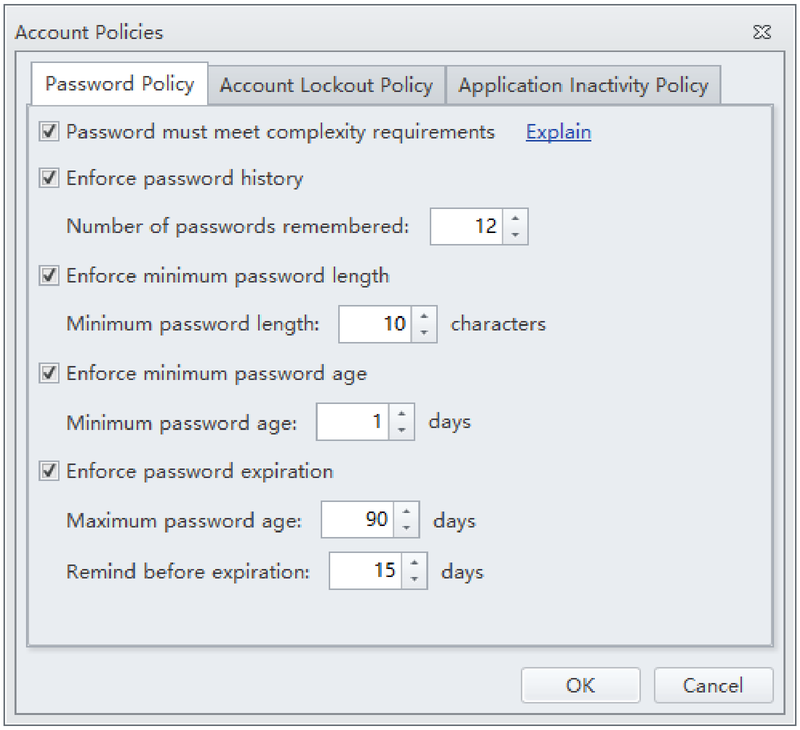
Account Policies

IMPORTANT Only an Administrator or an account has the Access account policies permission can manage account policies.

Account policies is used to define the default properties for the password policy, account lockout policy, and application inactivity policy.

Select **Account** > **Account Policies.** The Account Policies window appears.

Figure 2.9 Account Policies - Password Policy



☑ Password must meet complexity requirements Explain

☑ Enforce password history

Number of passwords remembered: 12

☑ Enforce minimum password length

Minimum password length: 10 characters

☑ Enforce minimum password age

Minimum password age: 1 days

☑ Enforce password expiration

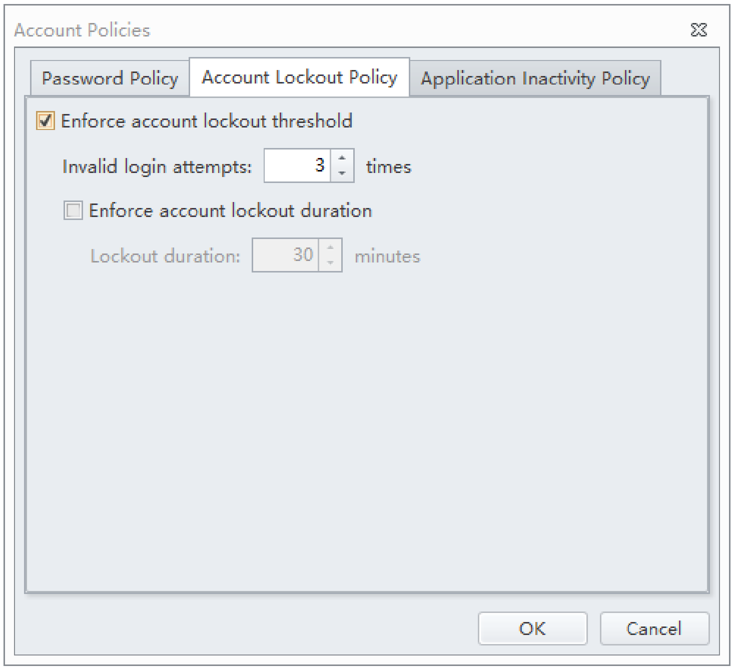
Maximum password age: 90 days

Remind before expiration: 15 days

NOTE The allowable range for each entry is as follows:

- Password History: 12-24 times
- Password Length: 10-12 characters
- Minimum Age for Password: 0-89 days
- Password Expiration: 1-90 days
- Reminder for Expiration: 1-90 days

Figure 2.10 Account Policies - Account Lockout Policy



☑ Enforce account lockout threshold

Invalid login attempts: 3 times

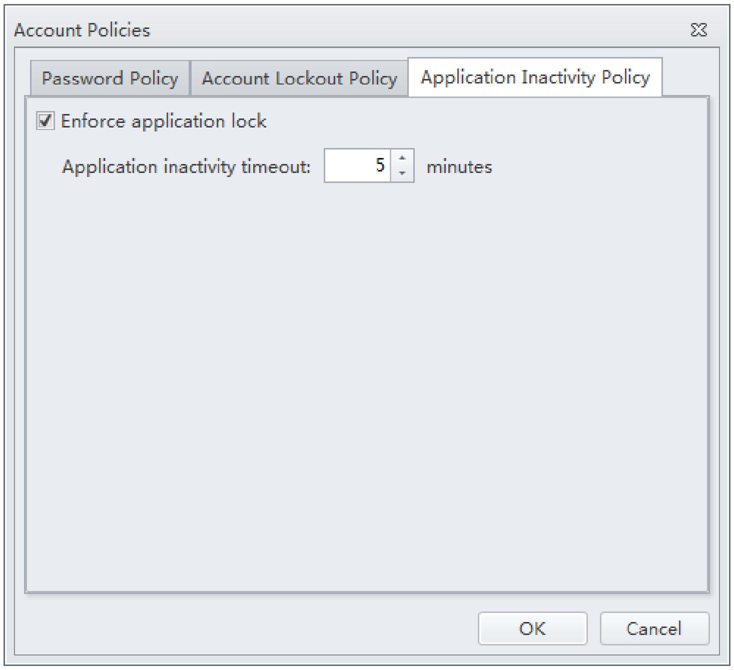
☐ Enforce account lockout duration

Lockout duration: 30 minutes

NOTE The allowable range for each entry is as follows:

- Invalid Login Attempts: 3-5 times
- Lockout Duration: 30-1440 minutes

Figure 2.11 Account Policies - Application Inactivity Policies



☑ Enforce application lock

Application inactivity timeout: 5 minutes

NOTE The allowable range for each entry is as follows:

- Inactivity Duration: 1-15 minutes

User Management Operation Log

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, User Management Operation Log.

Graphic and Gating Styles

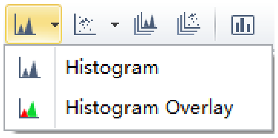
Plots

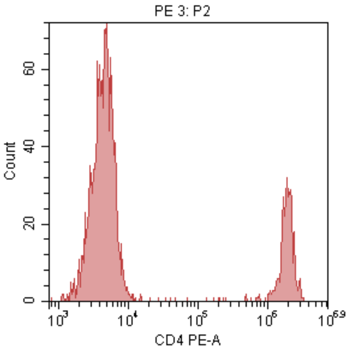
The CytExpert for Spectral software offers a variety of plot formats including:

- Single-parameter plots and histogram overlays
- Dual-parameter plots: dot plots, density plots, pseudo color plots, contour plots, and dot plot overlays
- Spectrum plots

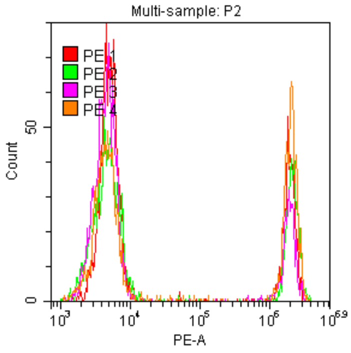
NOTE Histogram Overlays and Dot Plot Overlays can only be created from multiple samples in the Analysis screen. A maximum of 10 samples can be overlaid.

NOTE Spectrum option is only an option in the Raw worksheet.

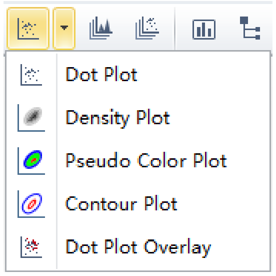


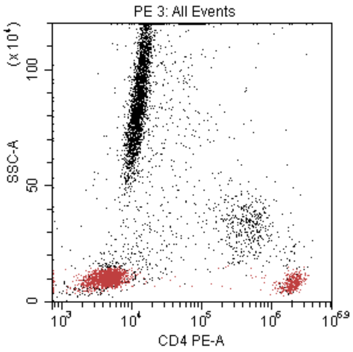


Histogram

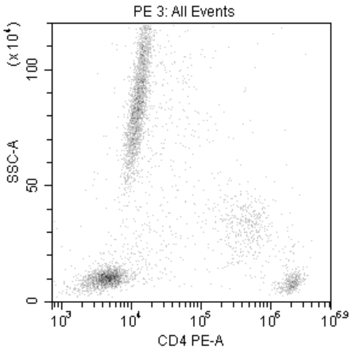


Multi-sample histogram overlays

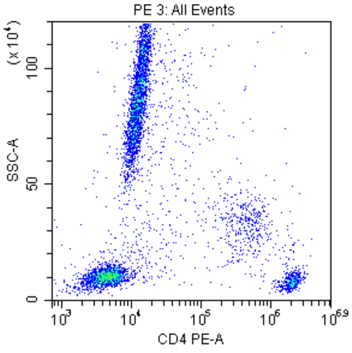




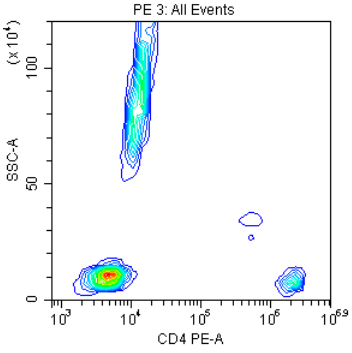
Dot plot



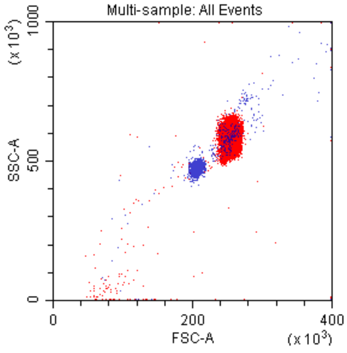
Density plot



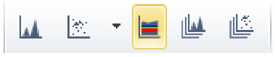
Pseudo color plot

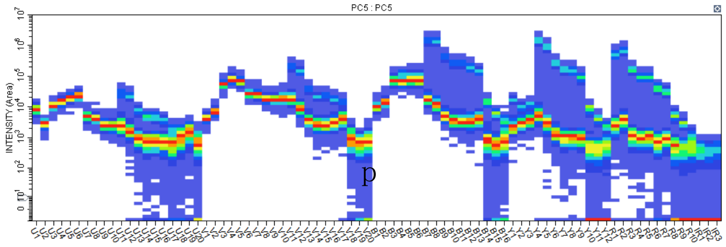


Contour plot



Dot plot overlay





Spectrum plot

Gates

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Gates.

NOTE Gating is not available in spectrum plots.

Plate Type Library

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Plate Type Library.

Adding a Plate Type

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Adding a Plate Type.

Editing a Plate Type

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Editing a Plate Type.

Duplicating a Plate Type

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Duplicating a Plate Type.

Deleting a Plate Type

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 2, Deleting a Plate Type.

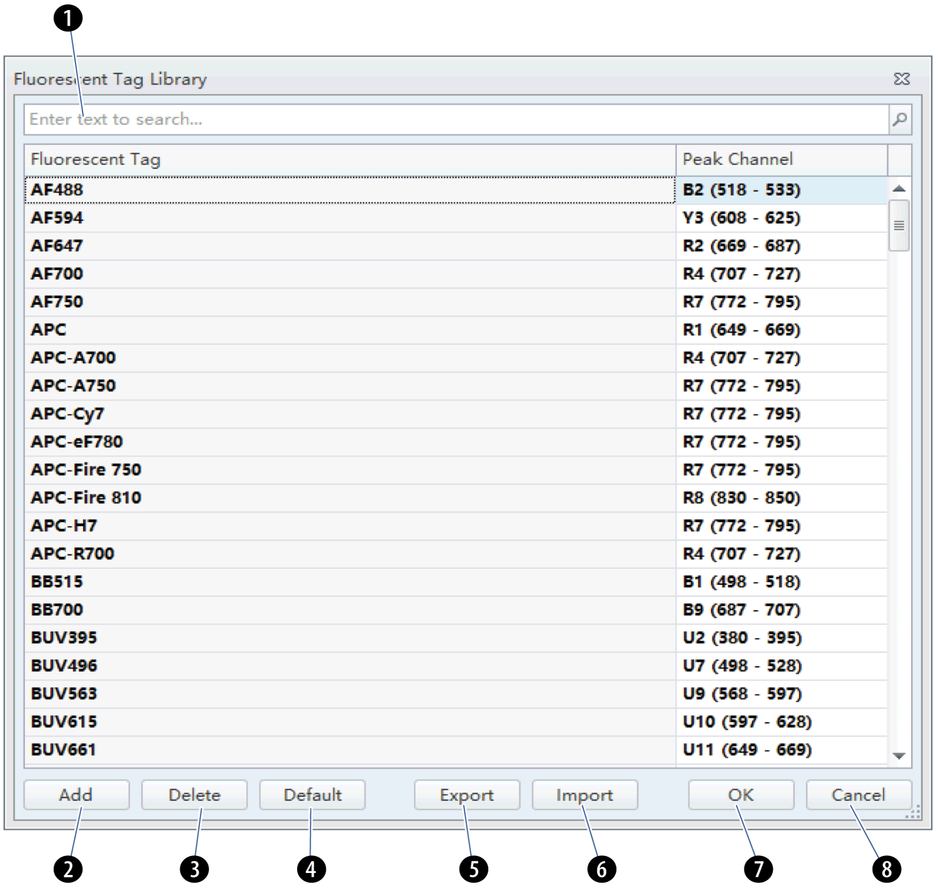
Fluorescent Tag Library

IMPORTANT The maximum number of fluorescent tags allowed in the Fluorescent Tag Library is 500.

The Fluorescent Tag Library is used to manage fluorescent tags. Fluorescent tag can be added, deleted, and edited from the Fluorescent Tag Library.

Select **Settings** > **Fluorescent Tag Library** to access the Fluorescent Tag Library. Refer to Figure 2.12.

Figure 2.12 Fluorescent Tag Library



Fluorescent Tag	Peak Channel
AF488	B2 (518 - 533)
AF594	Y3 (608 - 625)
AF647	R2 (669 - 687)
AF700	R4 (707 - 727)
AF750	R7 (772 - 795)
APC	R1 (649 - 669)
APC-A700	R4 (707 - 727)
APC-A750	R7 (772 - 795)
APC-Cy7	R7 (772 - 795)
APC-eF780	R7 (772 - 795)
APC-Fire 750	R7 (772 - 795)
APC-Fire 810	R8 (830 - 850)
APC-H7	R7 (772 - 795)
APC-R700	R4 (707 - 727)
BB515	B1 (498 - 518)
BB700	B9 (687 - 707)
BUV395	U2 (380 - 395)
BUV496	U7 (498 - 528)
BUV563	U9 (568 - 597)
BUV615	U10 (597 - 628)
BUV661	U11 (649 - 669)

1. **Search text box.** Filters fluorescent tag by tag name and peak channel.
2. **Add.** Creates a new fluorescent tag.
3. **Delete.** Deletes an existing fluorescent tag.
4. **Default.** Applies the default settings.
5. **Export.** Exports fluorescent tag files.

6. **Import.** Imports fluorescent tag files.
7. **OK.** Saves the fluorescent tag.
8. **Cancel.** Cancels the settings.

Adding a Fluorescent Tag

1 Select **Settings** > **Fluorescent Tag Library**. The Fluorescent Tag Library window appears. Refer to Figure 2.12.

IMPORTANT The maximum number of fluorescent tags allowed in the Fluorescent Tag Library is 500.

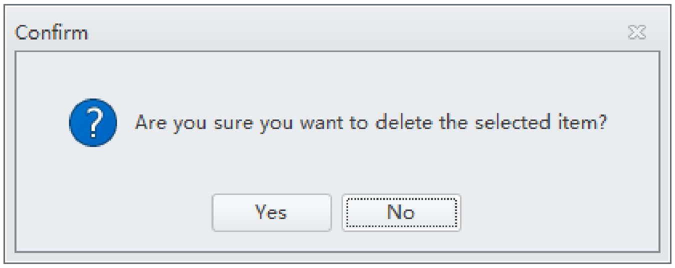
2 Select **Add.** Enter the fluorescent tag name and select the peak channel from the Peak Channel drop-down menu.

3 Select **OK** to exit the Fluorescent Tag Library window.

Deleting a Fluorescent Tag

1 Select **Settings** > **Fluorescent Tag Library**. The Fluorescent Tag Library window appears. Refer to Figure 2.12.

2 Select the fluorescent tag to be deleted, and select **Delete**. The following message appears.



Confirm

Are you sure you want to delete the selected item?

NOTE The default fluorescent tags, shown in bold, cannot be deleted from the Fluorescent Tag Library.

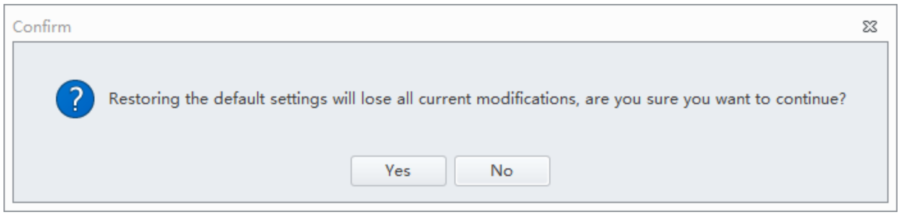
3 Select **Yes.**

4 Select **OK** to exit the Fluorescent Tag Library window.

Restoring the Default Settings

1 Select **Settings** > **Fluorescent Tag Library**. The Fluorescent Tag Library window appears. Refer to Figure 2.12.

2 Select **Default.** The following message appears.



Confirm

Restoring the default settings will lose all current modifications, are you sure you want to continue?

3 Select **Yes**.

4 Select **OK** to exit the Fluorescent Tag Library window.

Exporting Fluorescent Tag Files

1 Select **Settings** > **Fluorescent Tag Library**. The Fluorescent Tag Library window appears. Refer to Figure 2.12.

2 Select **Export** to specify a path and filename for the fluorescent tag file you are saving.

3 Select **Save**.

NOTE The generated file ends in .ft.

4 Select **Close**.

Importing Fluorescent Tag Files

1. Select **Settings** > **Fluorescent Tag Library**. The Fluorescent Tag Library window appears. Refer to Figure 2.12.

2. Select **Import** and locate the path where the fluorescent tag file is saved. Select the corresponding fluorescent tag file (.ft) to import.

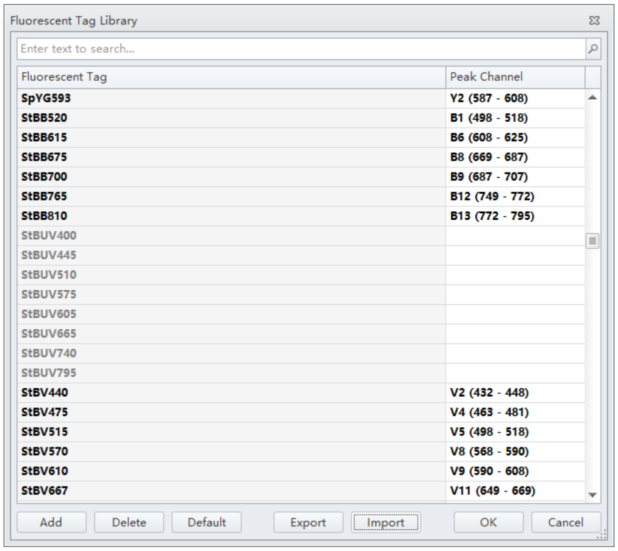
   NOTE If the fluorescent tag exists in the fluorescent tag library, the importing will overwrite the existing fluorescent tag. The system will prompt to ask you to confirm.

IMPORTANT The import failures could occur if:

- The number of fluorescent tags exists in the fluorescent tag library and the imported fluorescent tag file exceeds 500.
- The integrity of the imported fluorescent tag file is compromised.

3. Select **Open** to import.

   NOTE The fluorescent channels imported that are not supported by the software are shown in gray.



Fluorescent Tag	Peak Channel
SpYG593	Y2 (587 - 608)
StBB520	B1 (498 - 518)
StBB615	B6 (608 - 625)
StBB675	B8 (669 - 687)
StBB700	B9 (687 - 707)
StBB765	B12 (749 - 772)
StBB810	B13 (772 - 795)
StBUV400	
StBUV445	
StBUV510	
StBUV575	
StBUV605	
StBUV665	
StBUV740	
StBUV795	
StBV440	V2 (432 - 448)
StBV475	V4 (463 - 481)
StBV515	V5 (498 - 518)
StBV570	V8 (568 - 590)
StBV610	V9 (590 - 608)
StBV667	V11 (649 - 669)

4. Select **Close**.

Editing a Fluorescent Tag

1 Select **Settings** > **Fluorescent Tag Library**. The Fluorescent Tag Library window appears. Refer to Figure 2.12.

2 Select the fluorescent tag to be edited and select the desired peak channel from the Peak Channel drop-down menu.

NOTE The default fluorescent tags' name, shown in bold, cannot be edited. Select **Default** to restore the default settings. Refer to Restoring the Default Settings.

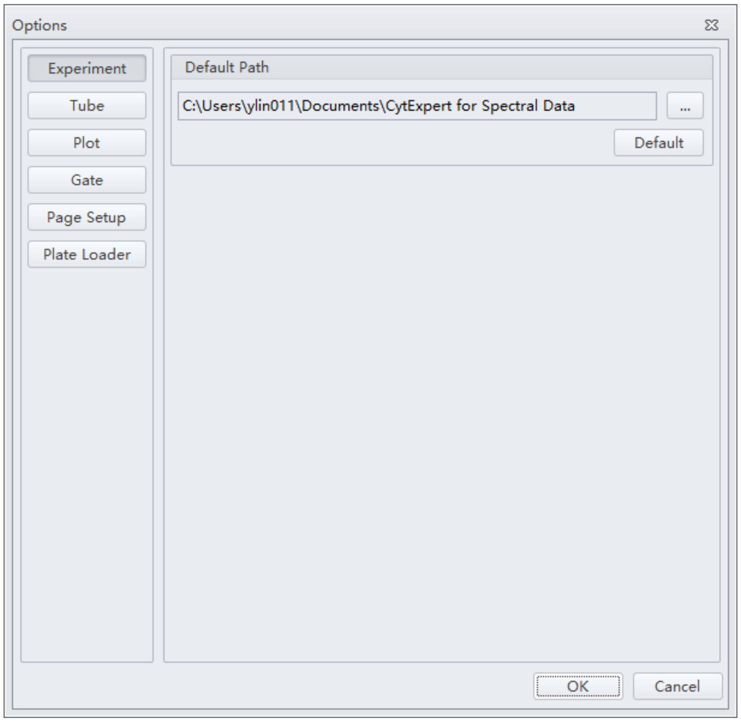
3 Select **Close**.

Software Settings

Select **Settings** > **Options** to configure the software settings.

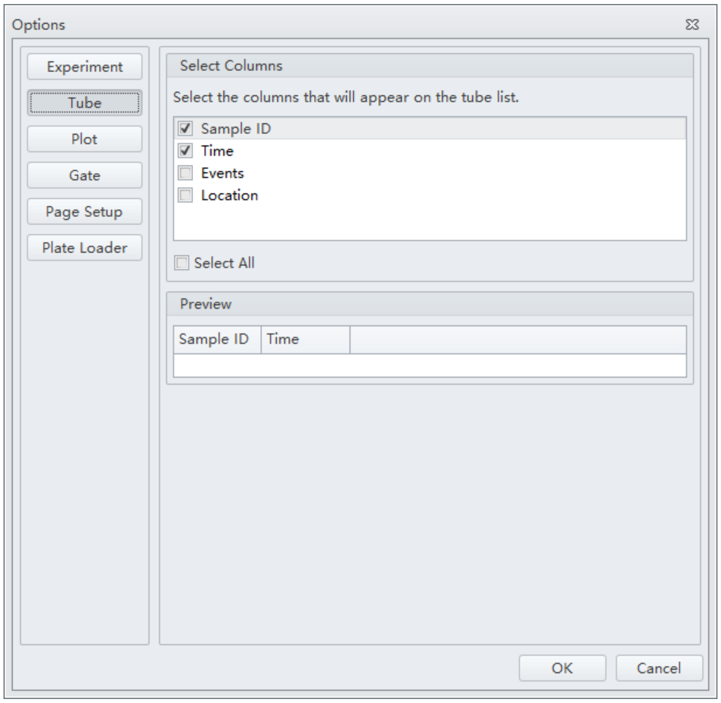
In the **Experiment** settings, you can set the experiment's default save path.

NOTE The Experiment setting is only available if the CytExpert User Management software option is installed.



C:\Users\ylin011\Documents\CytExpert for Spectral Data

In the **Tube** settings, you can select the columns that display in the tube section of the screen.



Select Columns

Select the columns that will appear on the tube list.

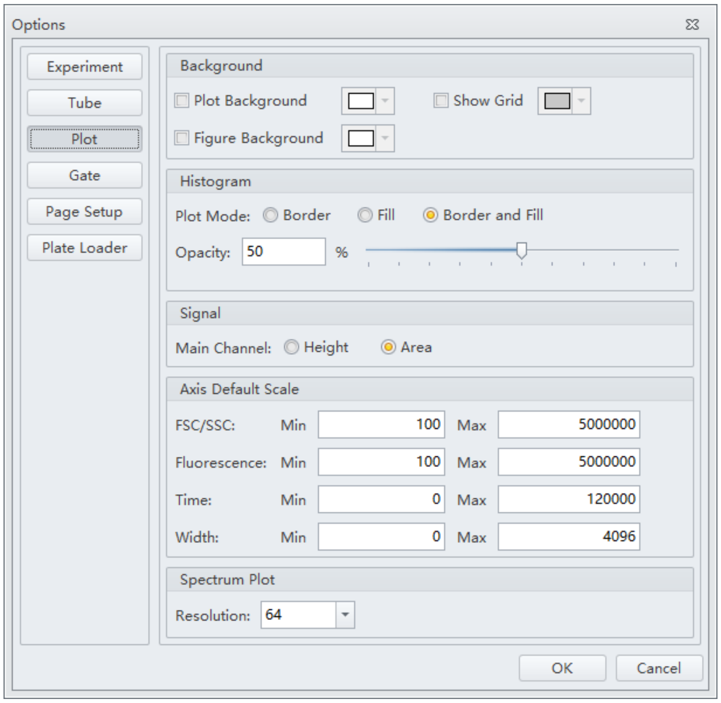
- ☑ Sample ID
- ☑ Time
- ☐ Events
- ☐ Location

☐ Select All

Preview

Sample ID	Time

In the **Plot** settings, you can define the background of the graphics display area, configure the histograms, and set the default signal parameters to either the channel's area or the channel's height. The default is **Area**. You can also set the default axis display range, and set the spectrum plot resolution.



Background

☐ Plot Background ☐ Show Grid

☐ Figure Background

Histogram

Plot Mode: ○ Border ○ Fill ● Border and Fill

Opacity: 50 %

Signal

Main Channel: ○ Height ● Area

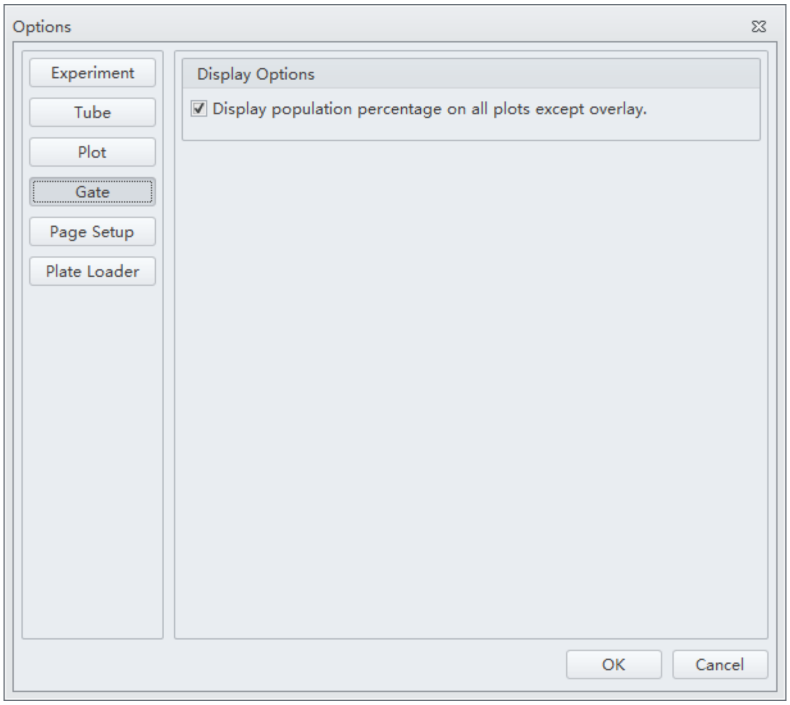
Axis Default Scale

FSC/SSC:	Min	100	Max	5000000
Fluorescence:	Min	100	Max	5000000
Time:	Min	0	Max	120000
Width:	Min	0	Max	4096

Spectrum Plot

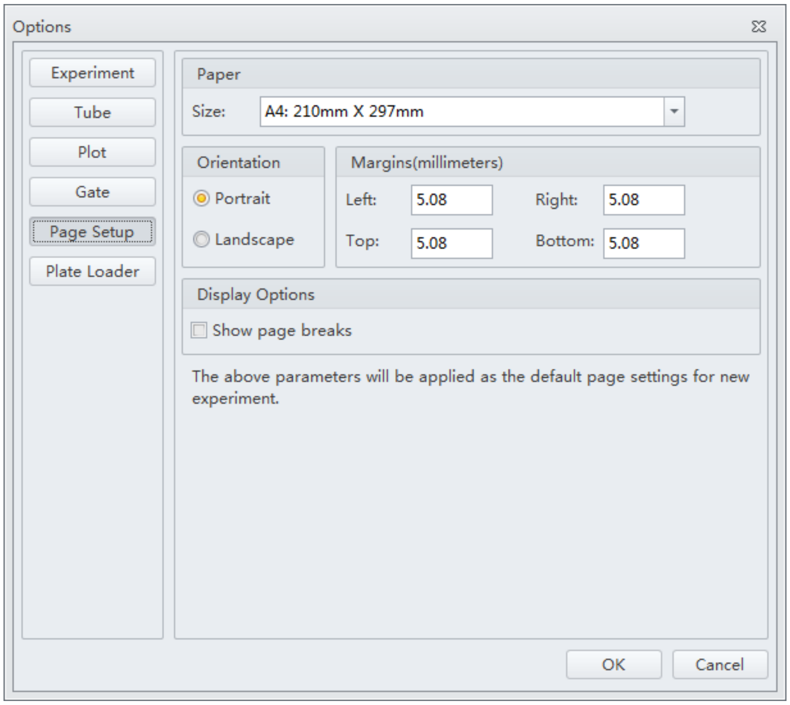
Resolution: 64

In the **Gate** settings, you can choose to display population percentage on all plots except overlay.



☑ Display population percentage on all plots except overlay.

In the **Page Setup** settings, you can change the page size, orientation, margin size, and display options. Select **Show page breaks** to display page boundaries within the Acquisition or Analysis views for simplifying plot arrangement for printing.



Size: A4: 210mm X 297mm

● Portrait
○ Landscape

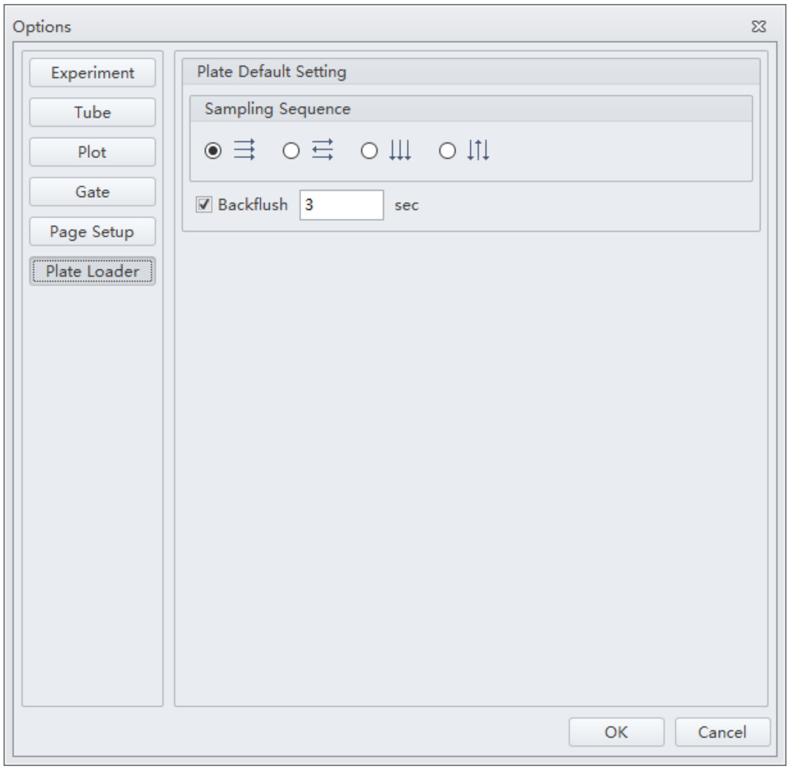
Left: 5.08
Right: 5.08
Top: 5.08
Bottom: 5.08

☐ Show page breaks

The above parameters will be applied as the default page settings for new experiment.

In the **Plate Loader** settings, you can select the plate type, sampling sequence, mix, and backflush settings for the plate loader.

NOTE This setting is only available in the Plate Loader sample injection mode.



● ⇶ ○ ⇄ ○ ↓↓↓ ○ ↓↑↓

☑ Backflush 3 sec

CHAPTER 3

Operation Principles

Overview

This chapter explains how the Cytometer measures scattered light and fluorescence as cells pass through the laser beam.

The illustrations in this chapter are not exact representations of the inside of the Cytometer. They are for explanatory purposes only.

This chapter contains information on:

- Sample Flow
- Laser Beam Shaping
- Cell Illumination
- Light Collection, Separation and Measurement
- Signal Processing
- Data Storage
- Automated Software Features
- Parameters
- Plot Display
- Statistics

Sample Flow

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Sample Flow for detailed instructions.

Sample Loading

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Sample Loading for detailed instructions.

Hydrodynamic Focusing

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Hydrodynamic Focusing for detailed instructions.

Laser Beam Shaping

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Laser Beam Shaping for detailed instructions.

Cell Illumination

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Cell Illumination for detailed instructions.

Forward Scatter

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Forward Scatter for detailed instructions.

Side Scatter and Fluorescent Light

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Side Scatter and Fluorescent Light for detailed instructions.

Light Collection, Separation and Measurement

Forward Scatter Collection

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Forward Scatter Collection for detailed instructions.

Side Scatter and Fluorescent Light Collection

Both side scatter and fluorescence are measured 90 degrees from the laser axis.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Side Scatter and Fluorescent Light Collection for detailed instructions.

Side Scatter

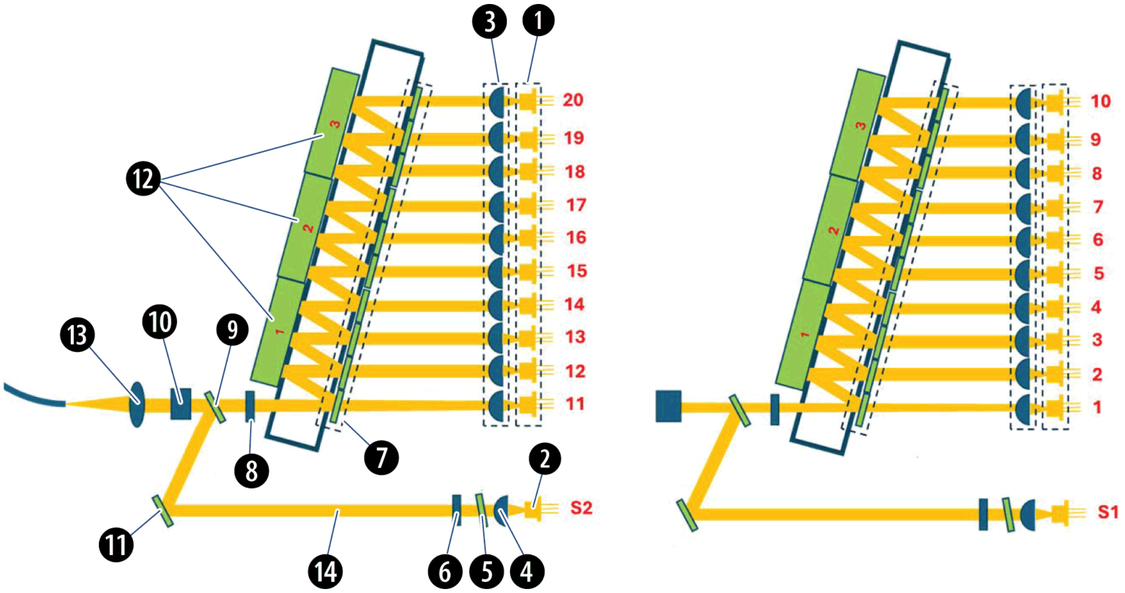
The CytoFLEX mosaic Spectral Detection Module collects SS from each individual laser. It is much more intense than FL.

Side scatter light collected by the objective lens is delivered by fiber optics to a patent-pending design with high performance, solid-state, high efficiency, and low-noise detector arrays.

Fluorescent Light

Fluorescence and scattered light are transmitted by optical fibers to the Wavelength division multiplexer (WDM). Each WDM is a unique detector array that corresponds to a different laser. Refer to CHAPTER 1, Wavelength Division Multiplexer (WDM). Each WDM contains optical filters and detectors for detecting channel fluorescence or scatter from a particular laser. It is necessary to ensure that the filter and software settings match for each channel.

Figure 3.1 Light Path through the WDM with a Single Port



1. Fiber array photo detectors (FAPD)
2. SSC fiber array photo detector (SSC FAPD)
3. Lens
4. Lens
5. SSC filter
6. ND filter
7. Filters
8. Filter
9. 30-degree reflector
10. Filter
11. Mirror
12. Mirror
13. Lens
14. Light path

Signal Processing

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Signal Processing for detailed instructions.

Data Storage

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Data Storage for detailed instructions.

NOTE Sample results can be printed out, saved to removable media, saved to a local hard drive or saved to a network drive. You can store sample results in Flow Cytometry Standard (FCS 3.1 or FCS 3.0) files.

Automated Software Features

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, Automated Software Features for detailed instructions.

Parameters

TIME Parameter

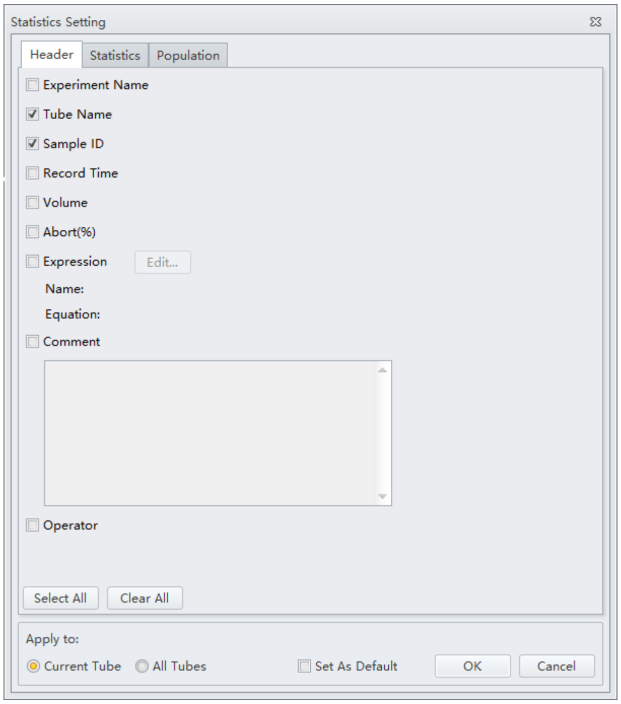
Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 3, TIME Parameter for detailed instructions.

Plot Display

The results of sample analysis appear on the Workstation screen as graphs called plots. Refer to CHAPTER 2, Graphic and Gating Styles.

Statistics

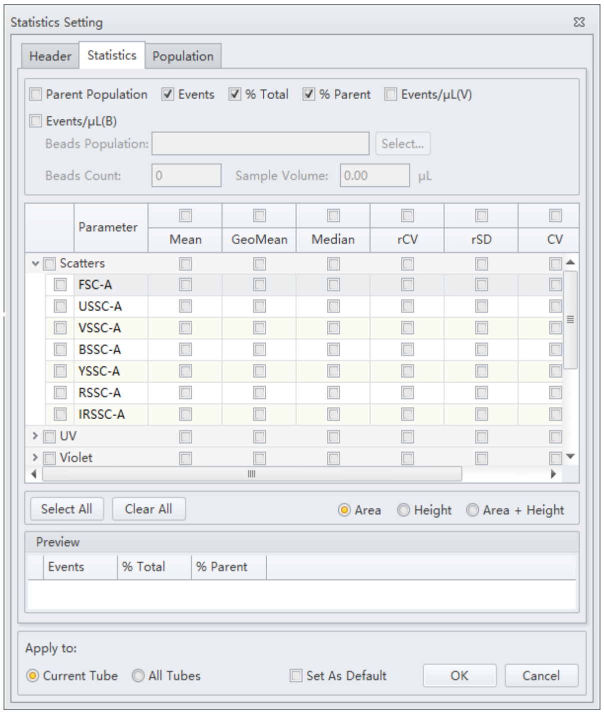
The Statistics Setting window allows you to change the display of the header, statistical elements and cell populations included.



☐ Experiment Name
☑ Tube Name
☑ Sample ID
☐ Record Time
☐ Volume
☐ Abort(%)
☐ Expression

Name:
Equation:
☐ Comment
☐ Operator

Apply to:
● Current Tube ○ All Tubes ☐ Set As Default



☐ Parent Population ☑ Events ☑ % Total ☑ % Parent ☐ Events/µL(V)
☐ Events/µL(B)
Beads Population:

Beads Count: 0 Sample Volume: 0.00 µL

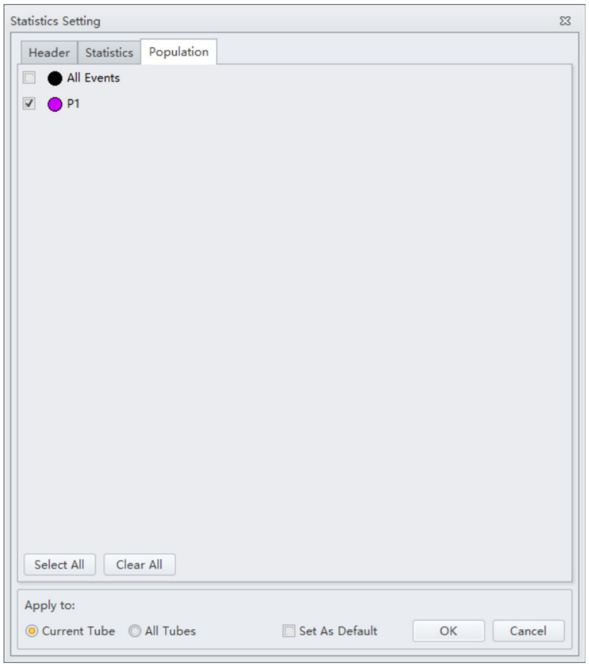
Parameter	☐ Mean	☐ GeoMean	☐ Median	☐ rCV	☐ rSD	☐ CV
☐ Scatters	☐	☐	☐	☐	☐	☐
☐ FSC-A	☐	☐	☐	☐	☐	☐
☐ USSC-A	☐	☐	☐	☐	☐	☐
☐ VSSC-A	☐	☐	☐	☐	☐	☐
☐ BSSC-A	☐	☐	☐	☐	☐	☐
☐ YSSC-A	☐	☐	☐	☐	☐	☐
☐ RSSC-A	☐	☐	☐	☐	☐	☐
☐ IRSSC-A	☐	☐	☐	☐	☐	☐
☐ UV	☐	☐	☐	☐	☐	☐
☐ Violet	☐	☐	☐	☐	☐	☐

● Area ○ Height ○ Area + Height

Preview

Events	% Total	% Parent

Apply to:
● Current Tube ○ All Tubes ☐ Set As Default



☐ ● All Events
☑ ● P1

Apply to:
● Current Tube ○ All Tubes ☐ Set As Default

CHAPTER 4

Daily Startup

Overview

IMPORTANT Verify that the two correct USB configuration keys are securely connected to the computer USB ports. If the USB configuration keys are not connected, the following error message appears: *CytExpert cannot find the license. Please check whether the correct USB configuration key has been plugged in.*

This chapter describes the instrument startup procedure.

Workflow:

Pre-startup inspection ➔ Turn on power ➔ Open software ➔ Initialize instrument

This chapter contains information on:

- Pre-Startup Inspection
- Turning On the Instrument
- Logging into the Software
- Initializing the Instrument

Pre-Startup Inspection

Before using the Cytometer with the CytoFLEX mosaic Spectral Detection Module, perform the following system checks.

Checking Waste and Reagent Levels [4 L Fluid Containers]

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Checking Waste and Reagent Levels [4 L Fluid Containers] for detailed instructions.

Checking Waste and Reagent Levels [10 L Fluid Cubitainers]

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Checking Waste and Reagent Levels [10 L Fluid Cubitainers] for detailed instructions.

Power Source Inspection

Check the power cable located below the power switch on the back of the Cytometer and the CytoFLEX mosaic Spectral Detection Module, and verify it is securely connected to the Cytometer, the CytoFLEX mosaic Spectral Detection Module and the appropriate power source.

Workstation Connections Inspection

Check that the monitor, mouse, keyboard, the Cytometer, and the CytoFLEX mosaic Spectral Detection Module are properly connected to the computer. Refer to Figure 1.3.

Turning On the Instrument

⚠ **CAUTION**

1. **If the Cytometer, CytoFLEX mosaic Spectral Detection Module or Workstation fails to start properly, check first to see whether the power cable and connection cables are properly connected.**
2. **Never shut off the power or disconnect a data cable while the instrument is performing a task. Doing so can result in data loss or damage to the system.**

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Turning On the Instrument for detailed instructions.

1 Turn on the main power switch located on the back of the Cytometer.

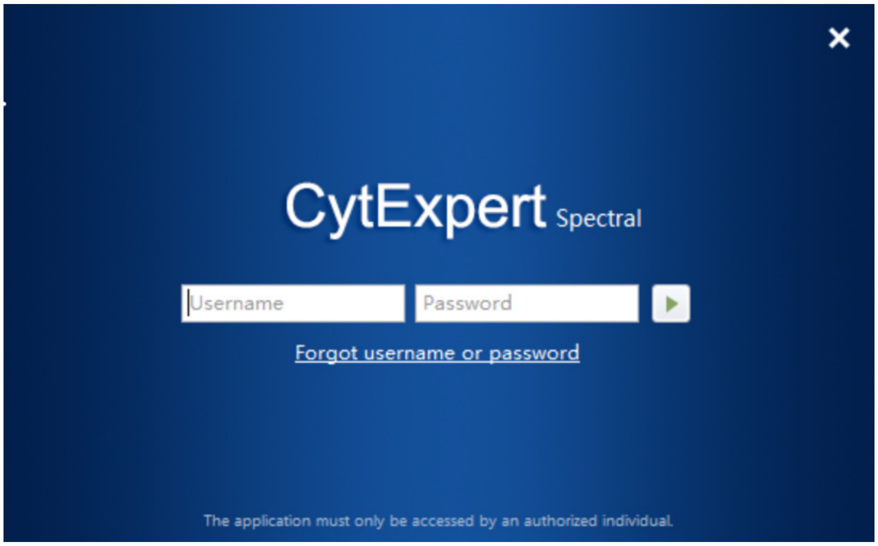
2 Wait for the Cytometer to finish powering on, then turn on the CytoFLEX mosaic Spectral Detection Module using the power switch located on the left side of the CytoFLEX mosaic Spectral Detection Module followed by the power button on the front.

3 Wait for the CytoFLEX mosaic Spectral Detection Module to finish powering on, then turn on the Workstation.

NOTE The CytoFLEX mosaic Spectral Detection Module takes approximately 40 seconds to power on.

Logging into the Software

1 Log in to the Windows operating system and double-click the CytExpert for Spectral desktop icon to open the software, the login window appears.



The application must only be accessed by an authorized individual.

NOTE The default software shortcut appears on the desktop. If you do not see the icon, the default installation path is under C:/Program Files/CytExpert for Spectral. Double-click CytExpert.exe to run the software.

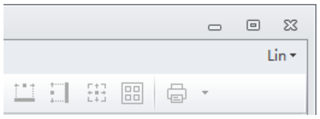
---

2 Enter your username and password.

---

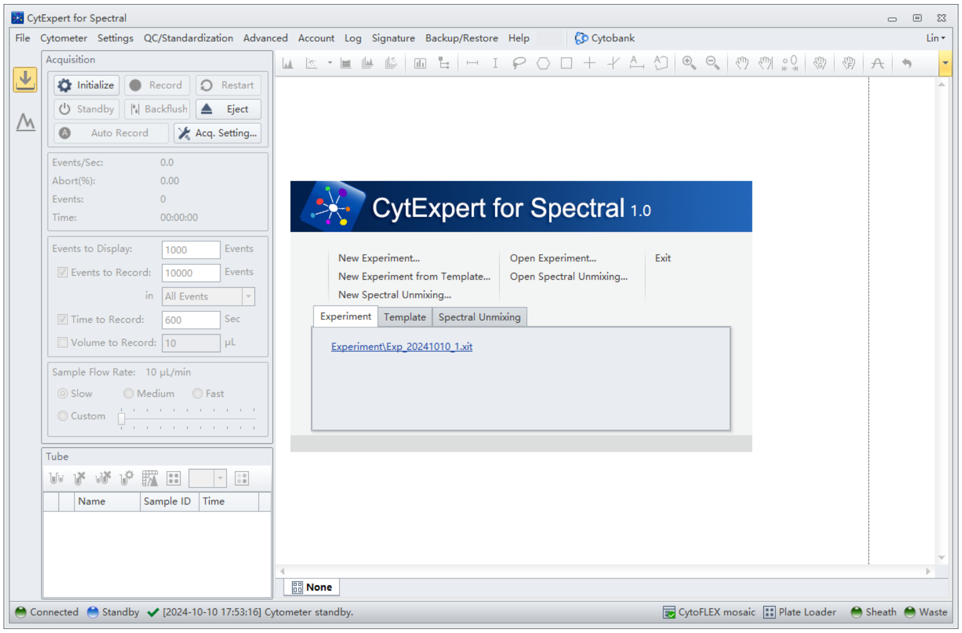
3 Select ▶.

NOTE The display name of the user that is currently logged in displays in the top, right corner of the software screen.



4 Confirm that the software, the CytoFLEX mosaic Spectral Detection Module and the Cytometer are properly connected.

a. Open the software. The Startup screen appears.



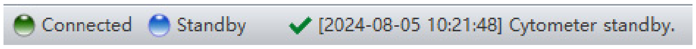
Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
○ Slow ○ Medium ○ Fast
○ Custom

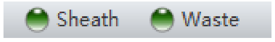
Connected Standby ✓ [2024-10-10 17:53:16] Cytometer standby.

b. Verify that the connection indicator light in the lower left corner of the software screen is green, and *Connected* is displayed. The left side shows the connection status, the middle shows the instrument status, and the right side shows the status details.

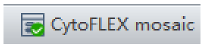


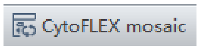
Connected Standby ✓ [2024-08-05 10:21:48] Cytometer standby.

c. Verify that the *Sheath* and *Waste* flow indicators in the lower right corner of the software screen are green indicating that the fluidics system is normal.



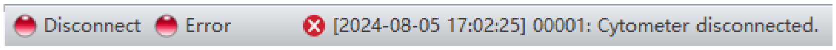
d. Verify that the CytoFLEX mosaic Spectral Detection Module status indicator in the lower right corner of the software screen has a green checkmark indicating that the CytoFLEX mosaic Spectral Detection Module is ready.



NOTE  indicates that the CytoFLEX mosaic Spectral Detection Module is warming up. This process takes approximately 4 minutes.

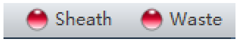
NOTE

- A red connection indicator light indicates that there is a faulty connection. Ensure that the instrument is properly turned on and connected. If necessary, restart both the Cytometer and the Workstation.

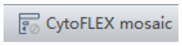


[2024-08-05 17:02:25] 00001: Cytometer disconnected.

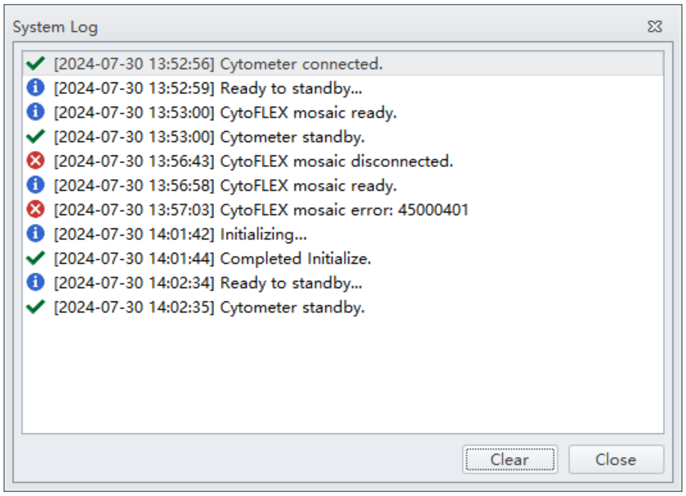
- After the instrument initializes, a warning beep sounds if there is a problem with the fluidics system. If a flow indicator is red and blinking, it means that the fluidics system requires attention.



- When the waste fluid sensor is disconnected, the waste flow indicator shows that the waste container is full or nearly full.
- A gray CytoFLEX mosaic Spectral Detection Module status indicator indicates that there is a faulty connection. Ensure that the CytoFLEX mosaic Spectral Detection Module is properly turned on and connected. If necessary, restart the CytoFLEX mosaic Spectral Detection Module.



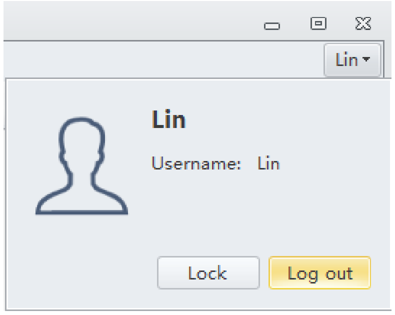
- Select the status information in the lower left to open the system log. Send a copy of the system log to your Beckman Coulter Representative for support if a service call is requested.



```
✓ [2024-07-30 13:52:56] Cytometer connected.
ℹ [2024-07-30 13:52:59] Ready to standby...
ℹ [2024-07-30 13:53:00] CytoFLEX mosaic ready.
✓ [2024-07-30 13:53:00] Cytometer standby.
✖ [2024-07-30 13:56:43] CytoFLEX mosaic disconnected.
ℹ [2024-07-30 13:56:58] CytoFLEX mosaic ready.
✖ [2024-07-30 13:57:03] CytoFLEX mosaic error: 45000401
ℹ [2024-07-30 14:01:42] Initializing...
✓ [2024-07-30 14:01:44] Completed Initialize.
ℹ [2024-07-30 14:02:34] Ready to standby...
✓ [2024-07-30 14:02:35] Cytometer standby.
```

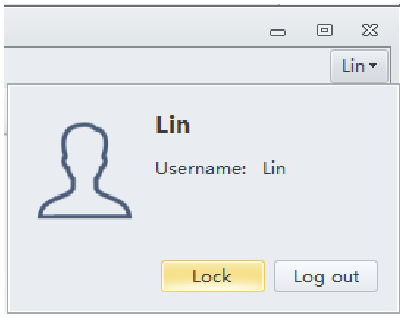
Logging Out of the Software

Select the username displayed in the top-right corner of the software screen and select **Log out.**



Locking the Account

Select the username displayed in the top-right corner of the software screen and select **Lock**.



The account locks automatically if it remains inactive for a specified duration. Refer to CHAPTER 2, Account Policies - Application Inactivity Policies.

Selecting the Proper Sample Injection Mode

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Selecting the Proper Sample Injection Mode for detailed instructions on the following procedures:

- Using Semi-Automatic Injection Mode
- Using Manual Injection Mode

Selecting the Plate Loader Sample Injection Mode [With Plate Loader]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Selecting the Plate Loader Sample Injection Mode [With Plate Loader] for detailed instructions on the following procedure:

- Using Plate Loader Injection Mode

Running the System Startup Program [with the Single Tube Loader]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Running the System Startup Program [with the Single Tube Loader] for detailed instructions.

Running the System Startup Program [With Plate Loader]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Running the System Startup Program [With Plate Loader] for detailed instructions.

Selecting Experiments from the Start Page

Refer to CHAPTER 2, Start Page.

Initializing the Instrument

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Initializing the Instrument for detailed instructions.

CHAPTER 5

Instrument Quality Control and Standardization

Overview

This chapter provides information on performing daily Quality Control (QC) on the CytoFLEX flow cytometer equipped with the CytoFLEX mosaic Spectral Detection Module and how to confirm that the instrument is working properly within the specified parameters. Quality Control allows you to determine whether your instrument can provide adequate signal strength and precision.

This chapter also provides information on performing standardization. CytoFLEX Ready to Use Daily QC Fluorospheres, CytoFLEX Daily IR QC Fluorospheres or any other reference material that is relevant for your application(s) may be used as the standardization sample(s). The system can only recognize a single peak.

Standardization can be used to monitor the Median Fluorescent Intensities (MFI), or target values for scatter and fluorescent parameters that have been defined and optimized for a specific application. All channels in the current configuration, those with/without an assigned QC target, can be tracked as necessary via Standardization since this portion of the CytExpert for Spectral software is used to assess application specific settings. Standardization, however, does NOT replace QC as the Cytometer's optical alignment (rCV statistical analysis), Laser Power and Laser Delay outputs are not measured during the run.

NOTE Beckman Coulter recommends performing QC on a daily basis.

NOTE CytExpert for Spectral QC includes an automated QC routine with Levey-Jennings (LJ) charts tracking and logging.

NOTE CytExpert for Spectral Standardization allows for application-specific settings to be established and applied to future experiments.

NOTE Perform QC on the same day prior to performing the CytExpert for Spectral standardization.

QC Workflow:

Prepare the QC sample → Enter the QC experiment → Select QC target values → Acquire data → Confirm results

Standardization Workflow:

Obtain target acquisition settings → Run standardization sample → Perform standardization → Apply standardization settings

This chapter contains information on:

Quality Control

The QC process verifies important system functions. The system:

1. Measures the laser power of each individual laser and ensures that each laser meets the system specifications.
2. Loads the QC sample and begins to acquire data.
3. Verifies that the actual laser delays match those set in the software and will adjust the delay accordingly.
4. Notifies you if laser delay is >2 µs from the previous setting. The software automatically changes the laser delay setting.

    OR

    Notifies you if laser delay is >5 µs from the previous setting. Manual laser delay adjustments are required. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 12, Setting Laser Delay.
5. Verifies and calibrates the gain settings. If any of these parameters are outside of the operating limits, the system automatically adjusts these parameters. If the system is unable to adjust these parameters to fall within the operating limits, the system notifies you.

Preparing the QC Sample

Required Materials

The following materials are required to complete the QC process:

- CytoFLEX Ready to Use Daily QC Fluorospheres
- CytoFLEX Daily IR QC Fluorospheres (for systems configured with an IR laser)
- CytoFLEX Sheath Fluid or another nonionic antimicrobial sheath fluid
- Sample tubes (12 x 75 mm)
- Vortexer

CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process for detailed instructions.

CytoFLEX Daily IR QC Fluorospheres Preparation Process

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, CytoFLEX Daily IR QC Fluorospheres Preparation Process for detailed instructions.

Preparing the QC Sample [With Plate Loader]

Required Materials

The following materials are required to complete the QC process:

- CytoFLEX Ready to Use Daily QC Fluorospheres
- CytoFLEX Daily IR QC Fluorospheres (for systems configured with an IR laser)
- CytoFLEX Sheath Fluid or another nonionic sheath fluid
- Standard 96-well plate
  - 96-well flat-bottom
  - 96-well V-bottom
  - 96-well U-bottom
- 96-well deep well plate
  - 96-well V-bottom
  - 96-well U-bottom
- Vortexer

CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Preparing the QC Sample [With Plate Loader], CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process for detailed instructions.

CytoFLEX Daily IR QC Fluorospheres Preparation Process

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Preparing the QC Sample [With Plate Loader], CytoFLEX Daily IR QC Fluorospheres Preparation Process for detailed instructions.

Importing Lot-Specific Target Values

Import lot-specific target values for each new lot of CytoFLEX Ready to Use Daily QC Fluorospheres, or CytoFLEX Daily IR QC Fluorospheres.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Importing Lot-Specific Target Values for detailed instructions.

⚠ CAUTION

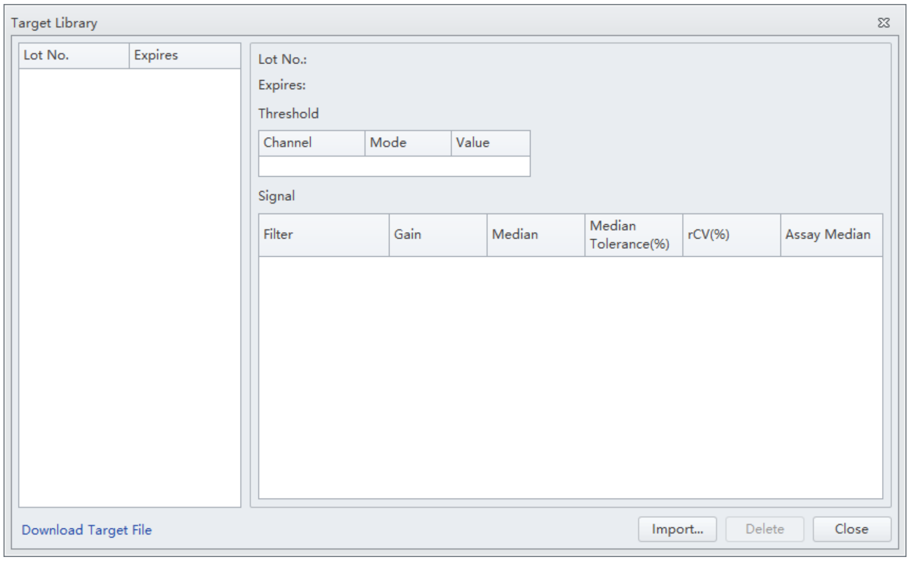
Risk of erroneous QC results. Different target value information correspond to different lot numbers. Selecting the wrong lot number will lead to erroneous QC results.

---

1 Select **Start QC/Standardization** from the QC/Standardization menu.

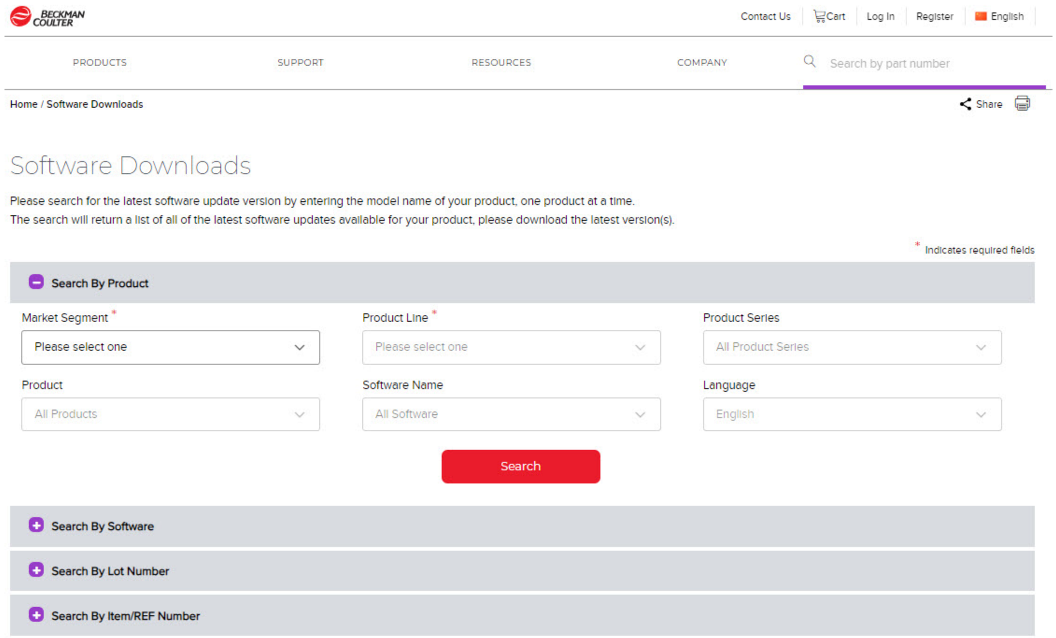
---

2 Select **Target Library** from the Settings menu. The Target Library window appears.



IMPORTANT The Beckman Coulter website may prompt you to select your Region and Country prior to the Beckman Coulter Technical Documents and Software page.

3 Select **Download Target File**. The Beckman Coulter Technical Documents and Software Downloads page appears.



Software Downloads

Please search for the latest software update version by entering the model name of your product, one product at a time.
The search will return a list of all of the latest software updates available for your product, please download the latest version(s).

* Indicates required fields

Search By Product

Market Segment *: Please select one
Product Line *: Please select one
Product Series: All Product Series
Product: All Products
Software Name: All Software
Language: English

NOTE If your CytoFLEX Workstation does not have access to the internet, navigate to https://www.beckmancoulter.com/wsrportal/page/softwareDownloadSearch using a computer with access to the internet and save the file to a USB drive. If the website is not accessible, contact us.

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4 If necessary, register and log in to the Beckman Coulter website.

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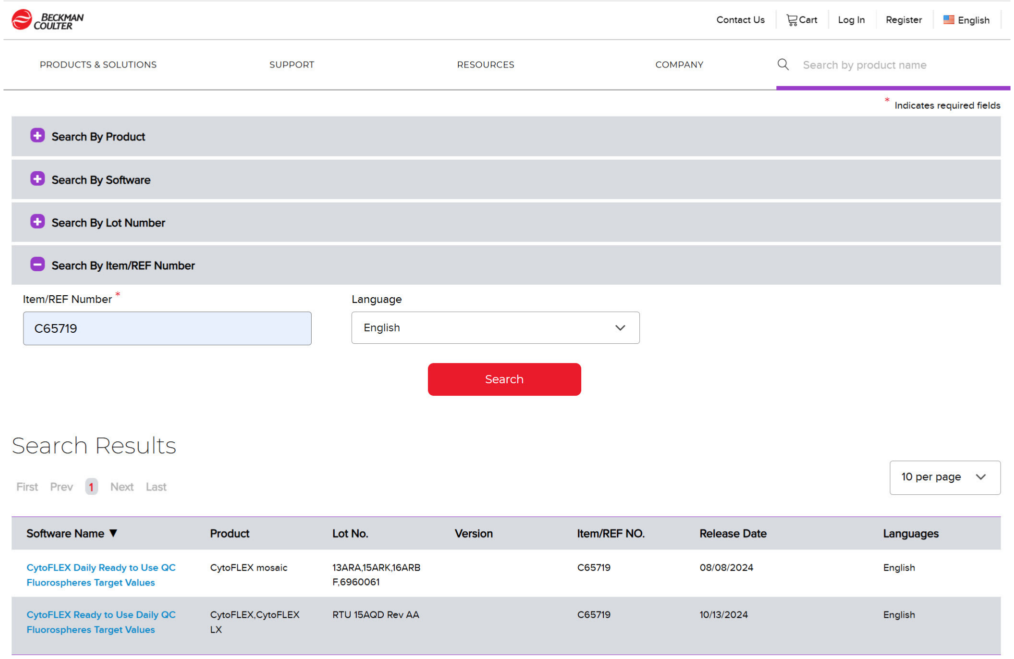
5 In the Search By Product section of the screen, select the following:

- **a.** Select **Research & Discovery** from the Market Segment drop-down menu.
- **b.** Select **Flow Cytometry** from the Product Line drop-down menu.
- **c.** Select **Instruments** from the Product Platform drop-down menu.
- **d.** Select **CytoFLEX mosaic** from the Product drop-down menu.
- **e.** Select **CytoFLEX Ready to Use QC Fluorospheres Target Value**, or **CytoFLEX Daily IR QC Fluorospheres Target Values** from the Software Name drop-down menu.
- **f.** Select **English** from the Language drop-down menu.

---

6 Select **Search**.

7 The search results appear below the Search By Item/REF Number tab.



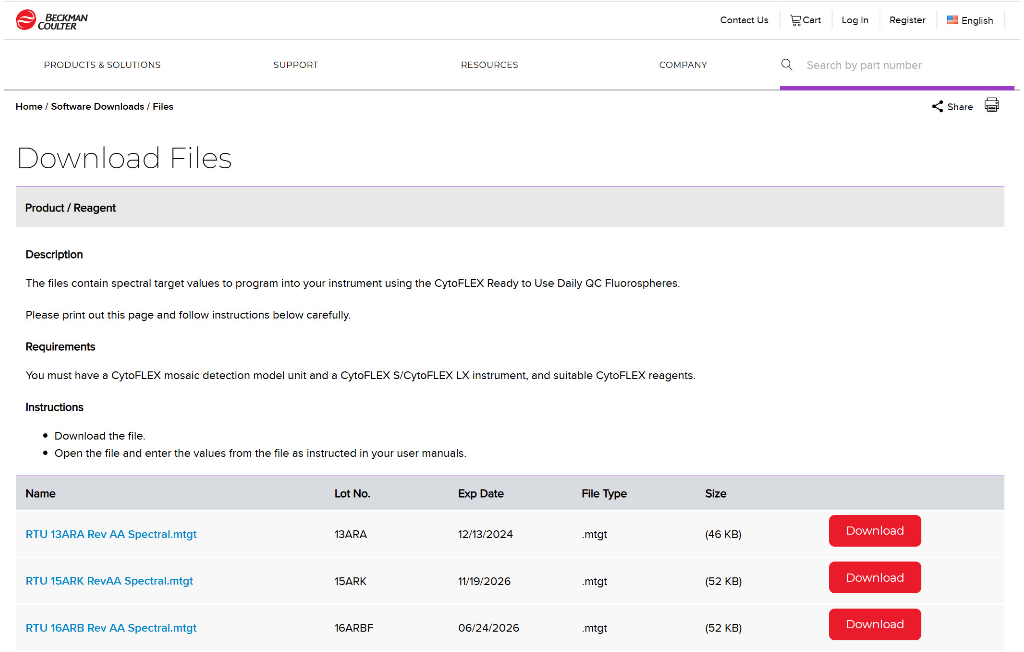
Item/REF Number *: C65719

Language: English

Search Results

Software Name ▼	Product	Lot No.	Version	Item/REF NO.	Release Date	Languages
CytoFLEX Daily Ready to Use QC Fluorospheres Target Values	CytoFLEX mosaic	13ARA,15ARK,16ARBF,6960061		C65719	08/08/2024	English
CytoFLEX Ready to Use Daily QC Fluorospheres Target Values	CytoFLEX,CytoFLEX LX	RTU 15AQD Rev AA		C65719	10/13/2024	English

8 Select **CytoFLEX Daily Ready to Use QC Fluorospheres Target Values**, or **CytoFLEX Daily IR QC Fluorospheres Target Values** under the Software Name column. The CmpytoFLEX Daily Ready to Use QC Fluorospheres Target Values or CytoFLEX Daily IR QC Fluorospheres Target Values page appears.



Download Files

Product / Reagent

Description

The files contain spectral target values to program into your instrument using the CytoFLEX Ready to Use Daily QC Fluorospheres.

Please print out this page and follow instructions below carefully.

Requirements

You must have a CytoFLEX mosaic detection model unit and a CytoFLEX S/CytoFLEX LX instrument, and suitable CytoFLEX reagents.

Instructions

- Download the file.
- Open the file and enter the values from the file as instructed in your user manuals.

Name	Lot No.	Exp Date	File Type	Size	
RTU 13ARA Rev AA Spectral.mtgt	13ARA	12/13/2024	.mtgt	(46 KB)	Download
RTU 15ARK RevAA Spectral.mtgt	15ARK	11/19/2026	.mtgt	(52 KB)	Download
RTU 16ARB Rev AA Spectral.mtgt	16ARBF	06/24/2026	.mtgt	(52 KB)	Download

9. Select **Download** next to the correct lot number from the CytoFLEX Daily Ready to Use QC Fluorospheres Target Values or CytoFLEX Daily IR QC Fluorospheres Target Values page.

10. If the File Download pop up window appears, select **Save** and browse to the desired file path.

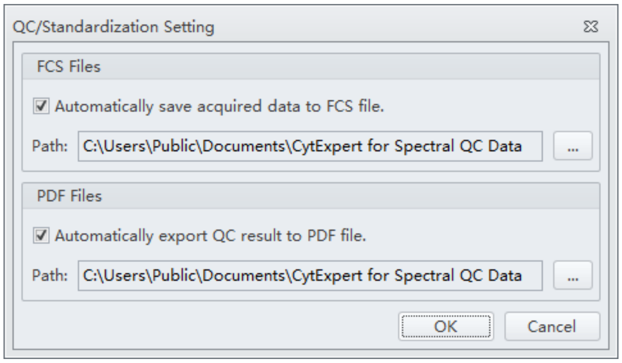
11. Select **Import** from the Target Library window in the CytExpert for Spectral software.

12. Navigate to the file saved in Step 10 and select **Open**.

13. Select **Close** to exit the Target Library window.

Collecting QC Data

QC data and reports are saved by default. Select **QC/Standardization Setting** in the Settings menu to change the default save settings or modify the file path these files are saved to.

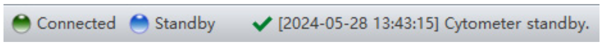


☑ Automatically save acquired data to FCS file.
Path: C:\Users\Public\Documents\CytExpert for Spectral QC Data

☑ Automatically export QC result to PDF file.
Path: C:\Users\Public\Documents\CytExpert for Spectral QC Data

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Collecting QC Data for detailed instructions.

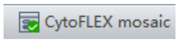
1. Double-click to start the CytExpert for Spectral software.
   a. Ensure that the **Connected** icon on the Status Bar near the bottom-left side of the display is green.



Connected Standby ✓ [2024-05-28 13:43:15] Cytometer standby.

NOTE If the icon is not green, ensure that the Cytometer USB is securely connected to the Workstation and restart the Workstation. Refer to Table 10.1, Troubleshooting [With CytoFLEX mosaic Spectral Detection Module].

b. Ensure that the **CytoFLEX mosaic** icon on the Status Bar near the bottom-right side of the display has a green checkmark.



NOTE If the icon doesn't have a green checkmark, ensure that the network cable is securely connected to the Workstation and restart the Workstation. Refer to Table 10.1, Troubleshooting [With CytoFLEX mosaic Spectral Detection Module].

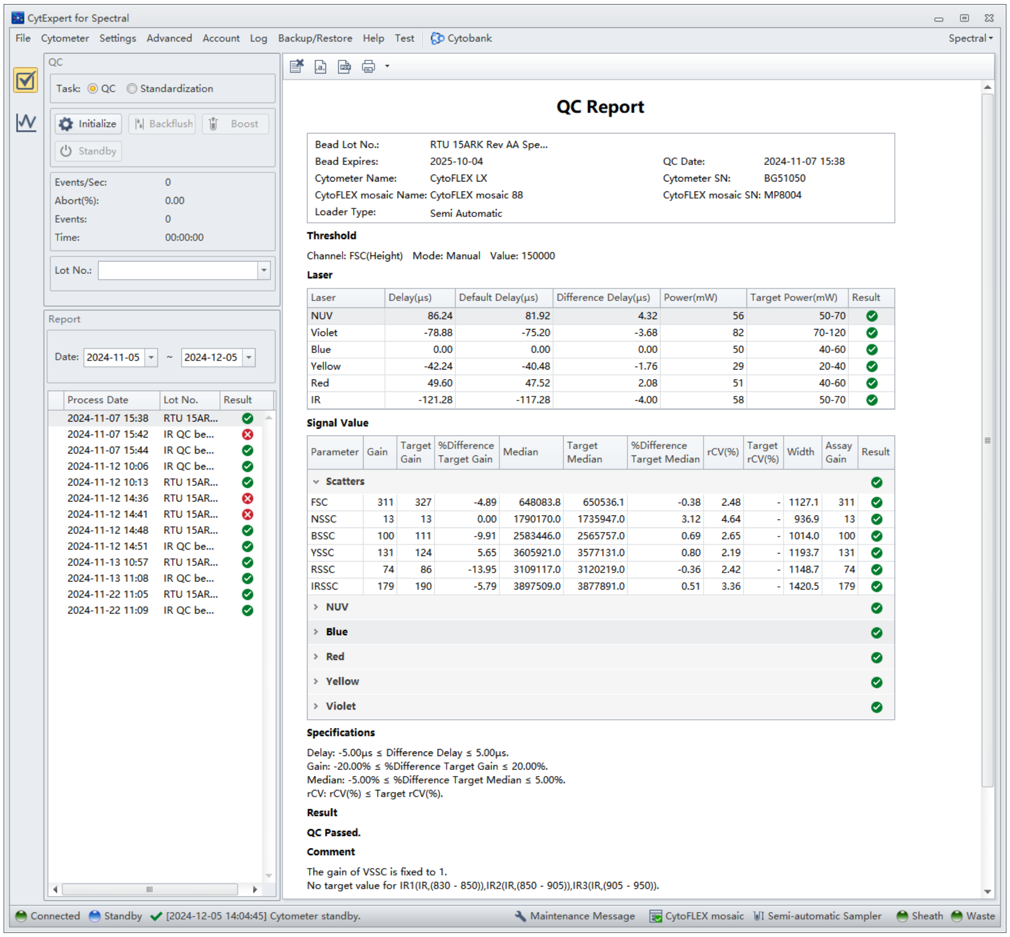
---

2 Verify the laser settings. Refer to CHAPTER 6, Laser Settings.

---

3 Select **Start QC/Standardization** in the QC/Standardization menu to access the QC experiment.

[CytoFLEX LX Shown]



Task: ● QC ○ Standardization

Events/Sec: 0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Lot No.:

Date: 2024-11-05 ~ 2024-12-05

Process Date	Lot No.	Result
2024-11-07 15:38	RTU 15AR...	✓
2024-11-07 15:42	IR QC be...	✗
2024-11-07 15:44	IR QC be...	✓
2024-11-12 10:06	IR QC be...	✓
2024-11-12 10:13	RTU 15AR...	✓
2024-11-12 14:36	RTU 15AR...	✗
2024-11-12 14:41	RTU 15AR...	✗
2024-11-12 14:48	RTU 15AR...	✓
2024-11-12 14:51	IR QC be...	✓
2024-11-13 10:57	RTU 15AR...	✓
2024-11-13 11:08	IR QC be...	✓
2024-11-22 11:05	RTU 15AR...	✓
2024-11-22 11:09	IR QC be...	✓

QC Report

Bead Lot No.: RTU 15ARK Rev AA Spe...
Bead Expires: 2025-10-04
Cytometer Name: CytoFLEX LX
CytoFLEX mosaic Name: CytoFLEX mosaic 88
Loader Type: Semi Automatic
QC Date: 2024-11-07 15:38
Cytometer SN: BG51050
CytoFLEX mosaic SN: MP8004

Threshold

Channel: FSC(Height) Mode: Manual Value: 150000

Laser

Laser	Delay(µs)	Default Delay(µs)	Difference Delay(µs)	Power(mW)	Target Power(mW)	Result
NUV	86.24	81.92	4.32	56	50-70	✓
Violet	-78.88	-75.20	-3.68	82	70-120	✓
Blue	0.00	0.00	0.00	50	40-60	✓
Yellow	-42.24	-40.48	-1.76	29	20-40	✓
Red	49.60	47.52	2.08	51	40-60	✓
IR	-121.28	-117.28	-4.00	58	50-70	✓

Signal Value

Parameter	Gain	Target Gain	%Difference Target Gain	Median	Target Median	%Difference Target Median	rCV(%)	Target rCV(%)	Width	Assay Gain	Result
Scatters											✓
FSC	311	327	-4.89	648083.8	650536.1	-0.38	2.48	-	1127.1	311	✓
NSSC	13	13	0.00	1790170.0	1735947.0	3.12	4.64	-	936.9	13	✓
BSSC	100	111	-9.91	2583446.0	2565757.0	0.69	2.65	-	1014.0	100	✓
YSSC	131	124	5.65	3605921.0	3577131.0	0.80	2.19	-	1193.7	131	✓
RSSC	74	86	-13.95	3109117.0	3120219.0	-0.36	2.42	-	1148.7	74	✓
IRSSC	179	190	-5.79	3897509.0	3877891.0	0.51	3.36	-	1420.5	179	✓
NUV											✓
Blue											✓
Red											✓
Yellow											✓
Violet											✓

Specifications

Delay: -5.00µs ≤ Difference Delay ≤ 5.00µs.
Gain: -20.00% ≤ %Difference Target Gain ≤ 20.00%.
Median: -5.00% ≤ %Difference Target Median ≤ 5.00%.
rCV: rCV(%) ≤ Target rCV(%).

Result

QC Passed.

Comment

The gain of VSSC is fixed to 1.
No target value for IR1(IR,(830 - 850)),IR2(IR,(850 - 905)),IR3(IR,(905 - 950)).

Connected Standby ✓ [2024-12-05 14:04:45] Cytometer standby.

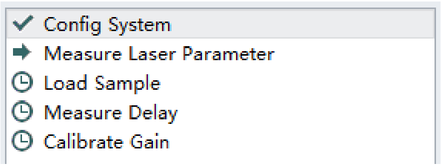
Ensure that the QC bead lot number is selectable in the Lot No. drop down menu. If the lot number is not selectable, refer to Importing Lot-Specific Target Values, then select the proper lot number.

4 Select **Initialize**.

5 Insert the prepared QC sample tube (see CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process) into the tube holder.

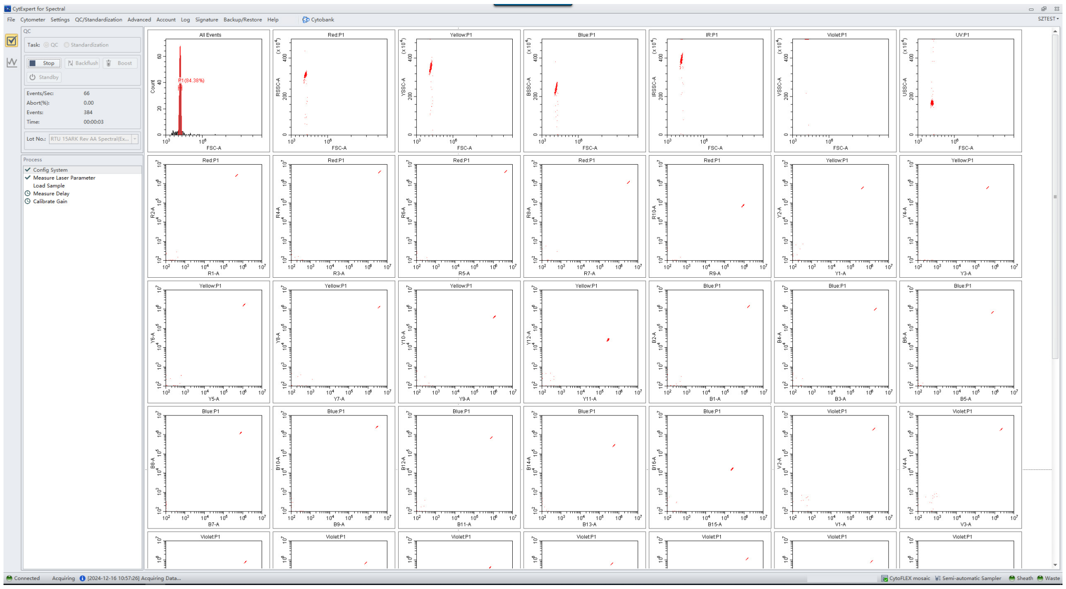
6 Select **Start** to load the sample and begin to run the QC procedure.

Completed processes appear on the left. Plots appear on the right. The QC experiment sequentially detects the system configuration, laser power, laser delay, signal strength, and coefficient of variation.



✓ Config System
➡ Measure Laser Parameter
◷ Load Sample
◷ Measure Delay
◷ Calibrate Gain

During QC, the software automatically seeks the CytoFLEX QC Fluorospheres and computes the results. The software returns to the QC Report screen after the QC run is complete.

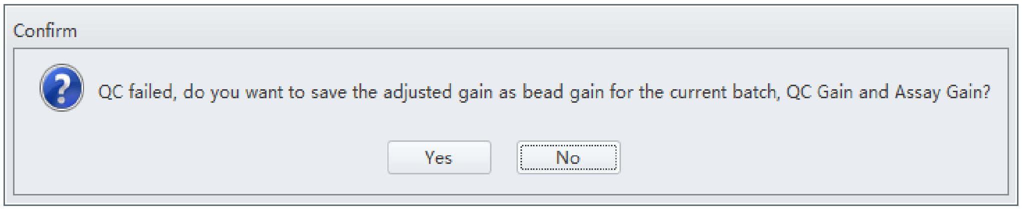


7 If the sampling rate is too low, the Cytometer stops the QC run and displays a prompt that the QC run fails to reach the required event flow rate. This is not considered a QC failure. If this situation occurs, increase the sample concentration by preparing a new tube of CytoFLEX Ready to Use Daily QC Fluorospheres (see CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process) and then perform the experiment.

NOTE Mix the fluorospheres dropper bottle thoroughly before dispensing it into a sample tube.

NOTE If the sample flow rate is not correct, calibrate the sample flow rate before running the QC. For instructions on calibrating the sample flow rate, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 12, Calibrating the Sample Flow Rate or CHAPTER 12, Calibrating the Sample Flow Rate [With Plate Loader].

8 If the lot number of CytoFLEX QC Fluorospheres is new and QC fails, the following software message appears. Select **Yes**.



Confirm

QC failed, do you want to save the adjusted gain as bead gain for the current batch, QC Gain and Assay Gain?

NOTE Target gain values must be established for each new lot number of CytoFLEX QC Fluorospheres. QC could fail up to 3 times upon running each new lot number for the first time until target gain values are established.

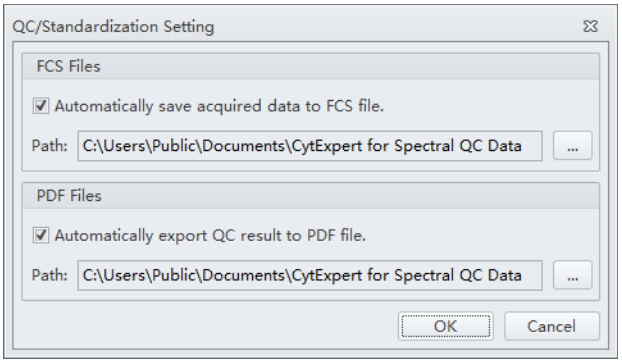
If the lot number of CytoFLEX QC Fluorospheres is NOT new and QC fails, refer to Step 2 of CHAPTER 5, Confirming Results, or CHAPTER 10, Troubleshooting.

If QC passes, proceed to Step 9.

9 Run Daily Clean to remove any residual fluorosphere particles if needed. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Daily Clean.

Collecting QC Data [With Plate Loader]

QC data and reports are saved by default. Select **QC/Standardization Setting** in the Settings menu to change the default save settings or modify the file path these files are saved to.



QC/Standardization Setting

FCS Files

☑ Automatically save acquired data to FCS file.

Path: C:\Users\Public\Documents\CytExpert for Spectral QC Data

PDF Files

☑ Automatically export QC result to PDF file.

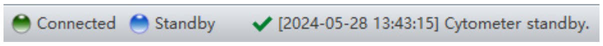
Path: C:\Users\Public\Documents\CytExpert for Spectral QC Data

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Collecting QC Data [With Plate Loader] for detailed instructions.

---

1 Double-click to start the CytExpert for Spectral software.

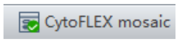
- **a.** Ensure that the **Connected** icon on the Status Bar near the bottom-left side of the display is green.

  ● Connected ● Standby ✓ [2024-05-28 13:43:15] Cytometer standby.

  NOTE If the icon is not green, ensure that the Cytometer USB is securely connected to the Workstation and restart the Workstation. Refer to Table 10.1, Troubleshooting [With CytoFLEX mosaic Spectral Detection Module].

- **b.** Ensure that the **CytoFLEX mosaic** icon on the Status Bar near the bottom-right side of the display has a green checkmark.

  NOTE If the icon doesn't have a green checkmark, ensure that the network cable is securely connected to the Workstation and restart the Workstation. Refer to Table 10.1, Troubleshooting [With CytoFLEX mosaic Spectral Detection Module].

---

2 Verify the laser settings. Refer to CHAPTER 6, Laser Settings.

IMPORTANT Ensure that the QC bead lot number is selectable in the Lot No. drop-down menu. If the lot number is not selectable, refer to CHAPTER 5, Importing Lot-Specific Target Values, then select the proper lot number.

3 Select `Start QC/Standardization` in the QC/Standardization menu to access the QC experiment.

4 Select **Initialize.**

5 Select **Eject.**

6 Insert the prepared QC well plate (see CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process or CytoFLEX Daily IR QC Fluorospheres Preparation Process) into the plate holder.

7 Select Plate Settings... . The Plate Settings window appears.

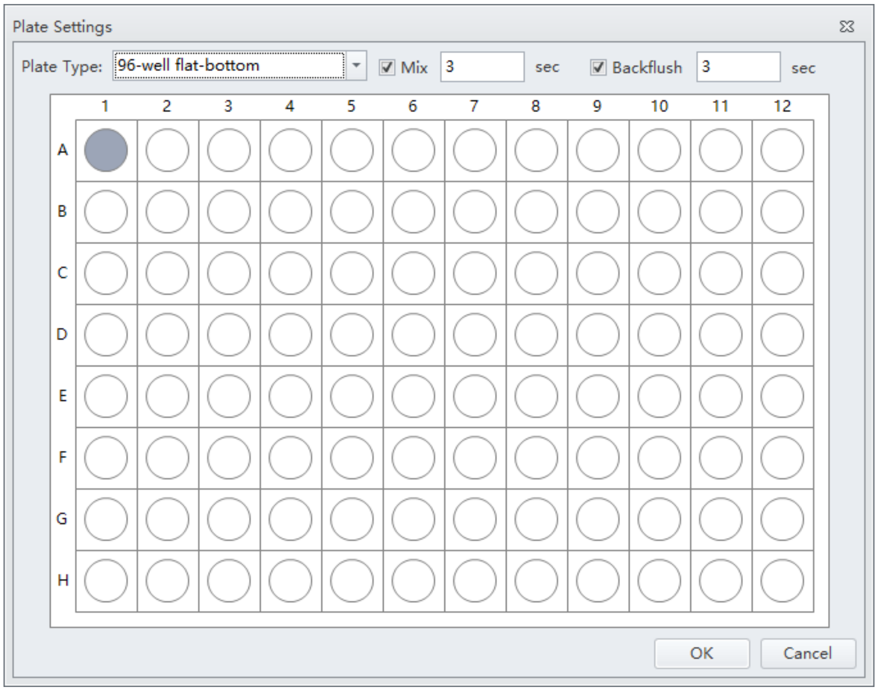


Plate Settings

Plate Type: 96-well flat-bottom ☑ Mix 3 sec ☑ Backflush 3 sec

IMPORTANT Ensure the well position on the plate matches the well position selected in the software.

8 Select the appropriate QC well.

9 Select the desired plate type from the Plate Type dropdown menu.

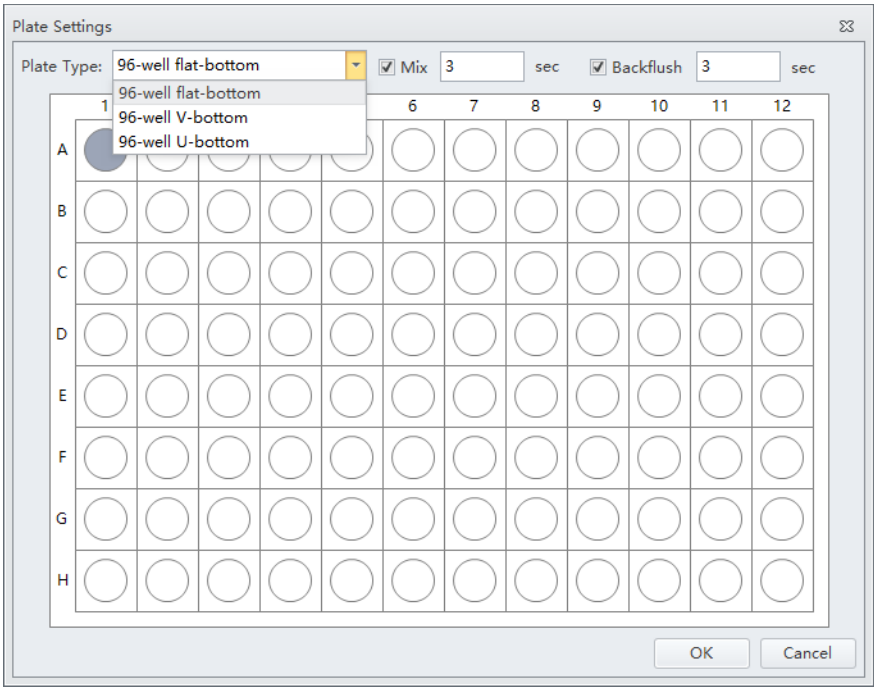


Plate Type: 96-well flat-bottom ☑ Mix 3 sec ☑ Backflush 3 sec

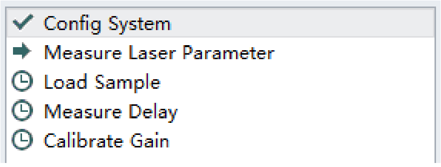
NOTE The available plate types included in the dropdown menu depend on the settings selected in the Plate Library. Refer to CHAPTER 2, Plate Type Library.

10 Select the Mix and Backflush settings in the top of the Plate Settings window.

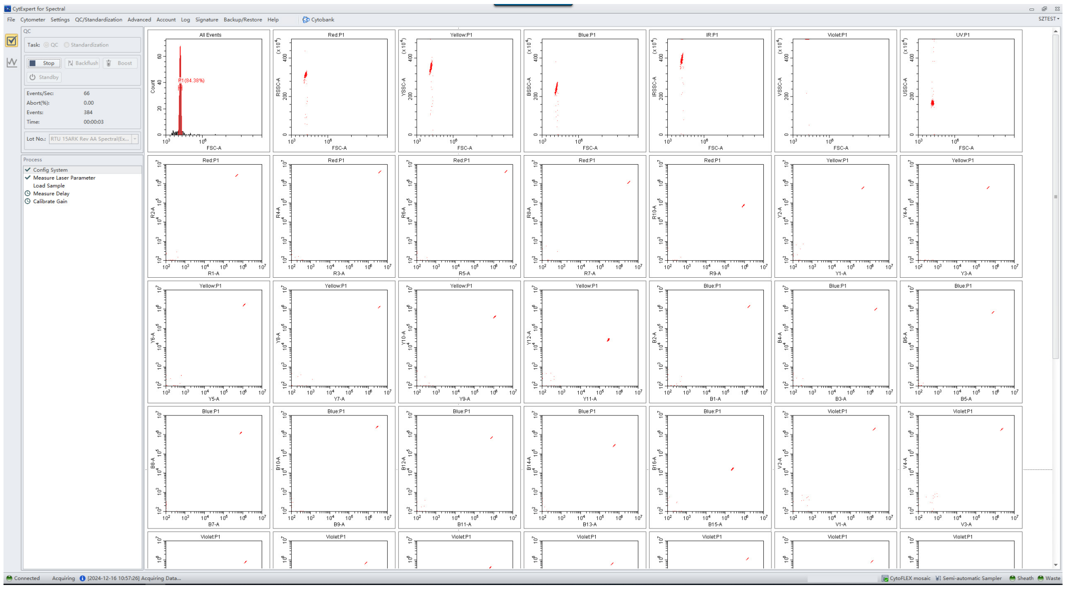
11 Select **OK.**

12 Select **Start** to load the sample and begin to run the QC procedure. The message *Please confirm that the correct plate is placed properly and press OK* appears. Select **OK.**

Completed processes appear on the left. Plots appear on the right. The QC experiment sequentially detects configuration, laser power, laser delay, signal strength, and coefficient of variation.



During QC, the software automatically seeks the CytoFLEX QC Fluorospheres, computes the results and determines whether the gain setting meets the assay target values. The software returns to the QC screen after the QC run is complete.



Events/Sec: 66
Abort(%): 0.00
Events: 384
Time: 00:00:03
Lot No.: RTU 15ARK Rev AA Spectral(Ex...

- ✓ Config System
- ✓ Measure Laser Parameter
- Load Sample
- Measure Delay
- Calibrate Gain

---

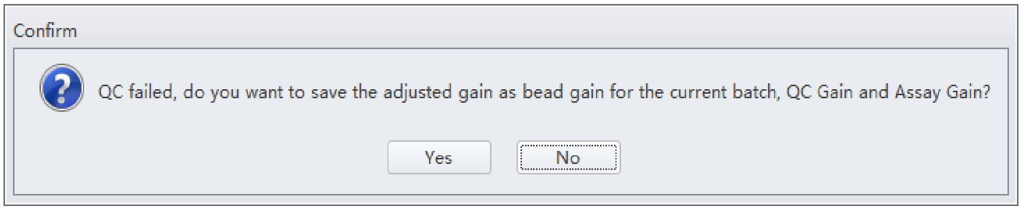
13 If the sampling rate is too low, the Cytometer stops the QC run and displays a prompt that the QC run fails to reach the required event flow rate. This is not considered a QC failure. If this situation occurs, increase the sample concentration by preparing a new tube of CytoFLEX Ready to Use Daily QC Fluorospheres (see CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process) and then perform the experiment.

NOTE Mix the fluorospheres dropper bottle thoroughly before dispensing it into a sample tube.

NOTE If the sample flow rate is not correct, calibrate the sample flow rate before running the QC. For instructions on calibrating the sample flow rate, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 12, Calibrating the Sample Flow Rate or CHAPTER 12, Calibrating the Sample Flow Rate [With Plate Loader].

---

14 If the lot number of CytoFLEX QC Fluorospheres is new and QC fails, the following software message appears. Select **Yes.**



QC failed, do you want to save the adjusted gain as bead gain for the current batch, QC Gain and Assay Gain?

NOTE Target gain values must be established for each new lot number of CytoFLEX QC Fluorospheres. QC could fail up to 3 times upon running each new lot number for the first time until target gain values are established.

If the lot number of CytoFLEX QC Fluorospheres is NOT new and QC fails, refer to Step 2 of CHAPTER 5, Confirming Results, or CHAPTER 10, Troubleshooting.

If QC passes, proceed to Step 15.

---

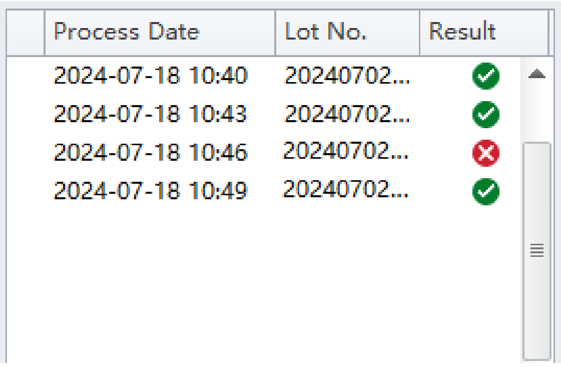
15 Run Daily Clean to remove any residual fluorosphere particles. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Daily Clean [With Plate Loader].

Confirming Results

Select **Start QC/Standardization** in the QC/Standardization menu to return to the QC Setting screen at any time to review completed experiment results.

---

1 Select a QC run from the QC Process list on the left and a QC report appears on the right.



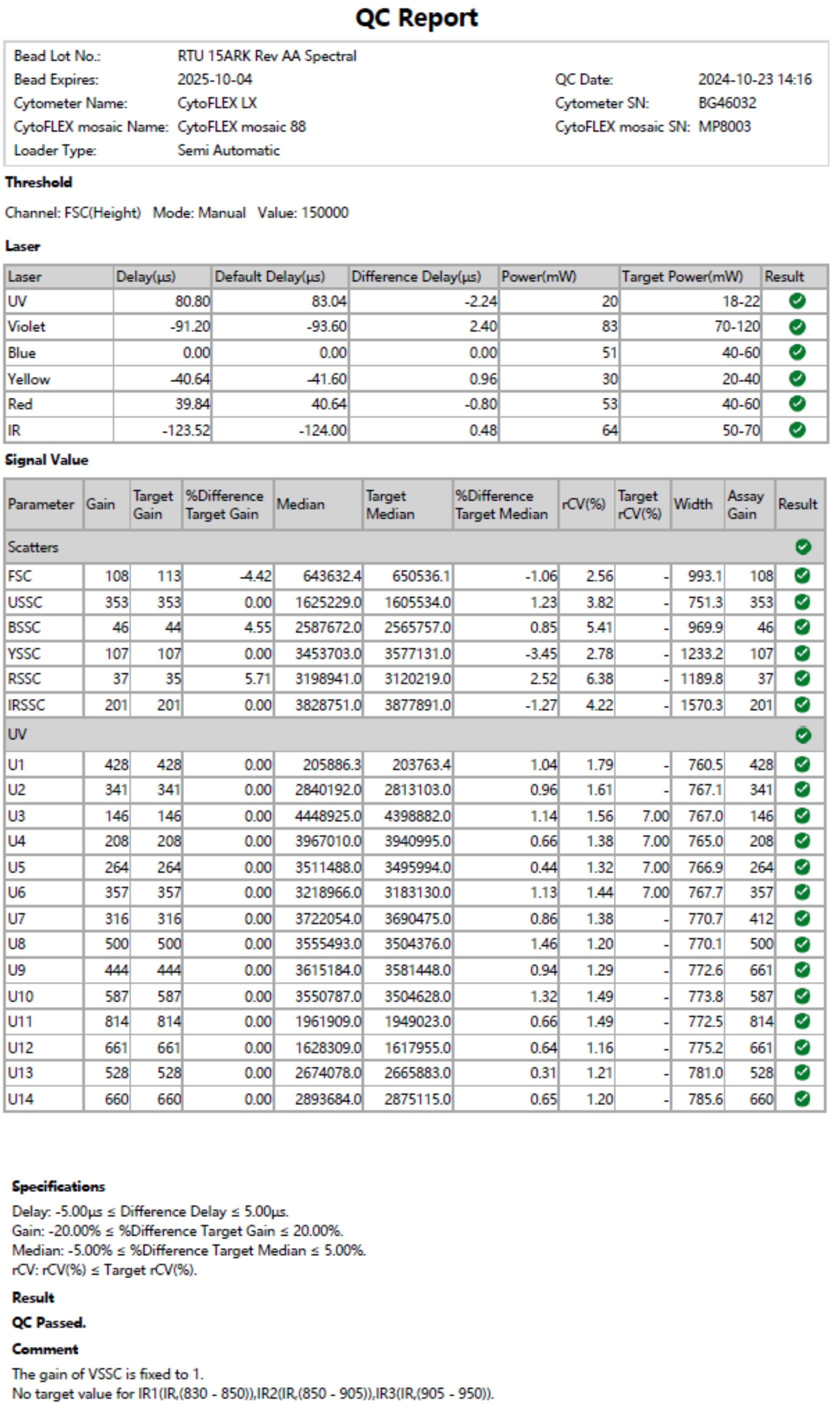
Process Date	Lot No.	Result
2024-07-18 10:40	20240702...	✓
2024-07-18 10:43	20240702...	✓
2024-07-18 10:46	20240702...	✗
2024-07-18 10:49	20240702...	✓

NOTE The results column indicates a passing QC result with a ✓ and a failed QC result with ✗.

QC results must meet the following criteria to pass:

- The gain differences must be ≤20% from the target gain.
- The median fluorescence intensity (MFI) differences must be ≤5% from the target MFI.
- The rCV must be within the target range.

The report area on the right displays detailed experiment results, including laser power, delay, testing conditions, and signal results. The same ✓ and ⊗ symbols are used to indicate each result. For items that fail, values falling outside the prescribed range are displayed in red font. In the Comment area, an explanation appears for each failed item.



QC Report

Bead Lot No.:	RTU 15ARK Rev AA Spectral		
Bead Expires:	2025-10-04	QC Date:	2024-10-23 14:16
Cytometer Name:	CytoFLEX LX	Cytometer SN:	BG46032
CytoFLEX mosaic Name:	CytoFLEX mosaic 88	CytoFLEX mosaic SN:	MP8003
Loader Type:	Semi Automatic		

Threshold

Channel: FSC(Height) Mode: Manual Value: 150000

Laser

Laser	Delay(µs)	Default Delay(µs)	Difference Delay(µs)	Power(mW)	Target Power(mW)	Result
UV	80.80	83.04	-2.24	20	18-22	✓
Violet	-91.20	-93.60	2.40	83	70-120	✓
Blue	0.00	0.00	0.00	51	40-60	✓
Yellow	-40.64	-41.60	0.96	30	20-40	✓
Red	39.84	40.64	-0.80	53	40-60	✓
IR	-123.52	-124.00	0.48	64	50-70	✓

Signal Value

Parameter	Gain	Target Gain	%Difference Target Gain	Median	Target Median	%Difference Target Median	rCV(%)	Target rCV(%)	Width	Assay Gain	Result
Scatters											✓
FSC	108	113	-4.42	643632.4	650536.1	-1.06	2.56	-	993.1	108	✓
USSC	353	353	0.00	1625229.0	1605534.0	1.23	3.82	-	751.3	353	✓
BSSC	46	44	4.55	2587672.0	2565757.0	0.85	5.41	-	969.9	46	✓
YSSC	107	107	0.00	3453703.0	3577131.0	-3.45	2.78	-	1233.2	107	✓
RSSC	37	35	5.71	3198941.0	3120219.0	2.52	6.38	-	1189.8	37	✓
IRSSC	201	201	0.00	3828751.0	3877891.0	-1.27	4.22	-	1570.3	201	✓
UV											✓
U1	428	428	0.00	205886.3	203763.4	1.04	1.79	-	760.5	428	✓
U2	341	341	0.00	2840192.0	2813103.0	0.96	1.61	-	767.1	341	✓
U3	146	146	0.00	4448925.0	4398882.0	1.14	1.56	7.00	767.0	146	✓
U4	208	208	0.00	3967010.0	3940995.0	0.66	1.38	7.00	765.0	208	✓
U5	264	264	0.00	3511488.0	3495994.0	0.44	1.32	7.00	766.9	264	✓
U6	357	357	0.00	3218966.0	3183130.0	1.13	1.44	7.00	767.7	357	✓
U7	316	316	0.00	3722054.0	3690475.0	0.86	1.38	-	770.7	412	✓
U8	500	500	0.00	3555493.0	3504376.0	1.46	1.20	-	770.1	500	✓
U9	444	444	0.00	3615184.0	3581448.0	0.94	1.29	-	772.6	661	✓
U10	587	587	0.00	3550787.0	3504628.0	1.32	1.49	-	773.8	587	✓
U11	814	814	0.00	1961909.0	1949023.0	0.66	1.49	-	772.5	814	✓
U12	661	661	0.00	1628309.0	1617955.0	0.64	1.16	-	775.2	661	✓
U13	528	528	0.00	2674078.0	2665883.0	0.31	1.21	-	781.0	528	✓
U14	660	660	0.00	2893684.0	2875115.0	0.65	1.20	-	785.6	660	✓

Specifications

Delay: -5.00µs ≤ Difference Delay ≤ 5.00µs.
Gain: -20.00% ≤ %Difference Target Gain ≤ 20.00%.
Median: -5.00% ≤ %Difference Target Median ≤ 5.00%.
rCV: rCV(%) ≤ Target rCV(%).

Result

QC Passed.

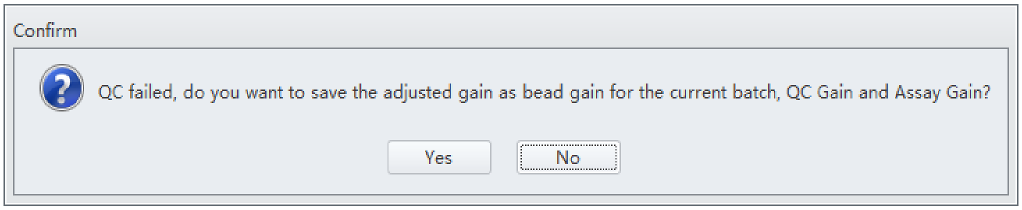
Comment

The gain of VSSC is fixed to 1.
No target value for IR1(IR,(830 - 850)),IR2(IR,(850 - 905)),IR3(IR,(905 - 950)).

2 If QC fails, follow the procedure below:

NOTE When the Confirm window appears, select **Yes** if you are sure that the QC failure is caused by the QC reagent batch difference or you need to use the adjusted gain.

Otherwise, select **No**.



Confirm

QC failed, do you want to save the adjusted gain as bead gain for the current batch, QC Gain and Assay Gain?

a. Verify whether the beads used were within their shelf life and stored in accordance with the appropriate instruction manual.

b. Verify whether the allocated sample tube was prepared as required and correctly positioned.

c. Prime the Flow Cell, and retest. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 12, Priming the Flow Cell.

d. Run the Daily Clean, and retest. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Daily Clean.

e. Run the Deep Clean Procedure, and restart. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Deep Clean Procedure.

f. Repeat Steps c-d.

NOTE If QC fails two times in a row on the same day after repeating Steps a-f, contact us.

3 If necessary, you can select (for CSV format) or (for PDF format) in the top left corner of the report area to export the QC results.

4 Select **Close QC/Standardization** in the File menu to exit the QC screen.

Creating Levey-Jennings Charts

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Creating Levey-Jennings Charts for detailed instructions.

QC Result Manager

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, QC Result Manager for detailed instructions.

Standardization

Ensure that the standardization sample has been run at optimized experiment settings to determine the standardization sample threshold setting as well as median values for all relevant channels.

Preparing the Standardization Sample

Use Beckman Coulter CytoFLEX Ready to Use Daily QC Fluorospheres or any other reference material that is relevant for your application.

Required Materials

The following materials are required to complete the QC process:

- CytoFLEX Ready to Use Daily QC Fluorospheres, or other material applicable for your application

  NOTE Note that the VSSC and NSSC channels are designed for small particle detection. It is not recommended to use the CytoFLEX Ready to Use Daily QC Fluorospheres when performing standardization for the VSSC and NSSC channels.

- CytoFLEX Daily IR QC Fluorospheres (for systems configured with an IR laser)
- CytoFLEX Sheath Fluid
- Sample tubes (12 x 75 mm)
- Vortexer

Preparation Process

For procedures, refer to CytoFLEX Ready to Use Daily QC Fluorospheres Preparation Process and/or CytoFLEX Daily IR QC Fluorospheres Preparation Process.

Generating Target Median Values

Generate lot-specific target values for each new lot of standardization beads.

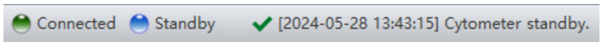
NOTE If you want to generate target median values automatically for all channels from the Acq. Setting Catalog using existing settings in the Acq. Setting Catalog, refer to Creating a New Standardization Item from Acq. Setting Catalog.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Generating Target Median Values for detailed instructions.

---

1 Double-click to start the CytExpert for Spectral software.

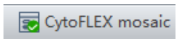
a. Ensure that the **Connected** icon on the Status Bar near the bottom-left side of the display is green.



● Connected ● Standby ✓ [2024-05-28 13:43:15] Cytometer standby.

NOTE If the icon is not green, ensure that the Cytometer USB is securely connected to the Workstation and restart the Workstation. Refer to Table 10.1, Troubleshooting [With CytoFLEX mosaic Spectral Detection Module].

b. Ensure that the **CytoFLEX mosaic** icon on the Status Bar near the bottom-right side of the display has a green checkmark.



CytoFLEX mosaic

NOTE If the icon don’t have a green checkmark, ensure that the network cable is securely connected to the Workstation and restart the Workstation. Refer to Table 10.1, Troubleshooting [With CytoFLEX mosaic Spectral Detection Module].

---

2 Select **New Experiment** to create an experiment.

---

3 Add a tube and edit the tube name. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Changing the Tube Name for detailed instructions.

---

4 Select **Acq. Setting** to input the optimized Gain and Threshold in the relevant channels in the Acq. Setting window. Refer to CHAPTER 6, Configuring Acquisition Settings.

NOTE Use the FSC channel as the trigger channel. The threshold may need to be adjusted to visualize the QC beads populations. If so, record this value for future reference.

---

5 Load the sample tube.

NOTE The sample tube holder accommodates 1.5 mL, 2.0 mL, and 12 x 75 mm sample tubes.

---

6 Create a FSC/SSC dot plot and gate on the singlet bead population. Refer to CHAPTER 6, Creating Plots and Gates.

---

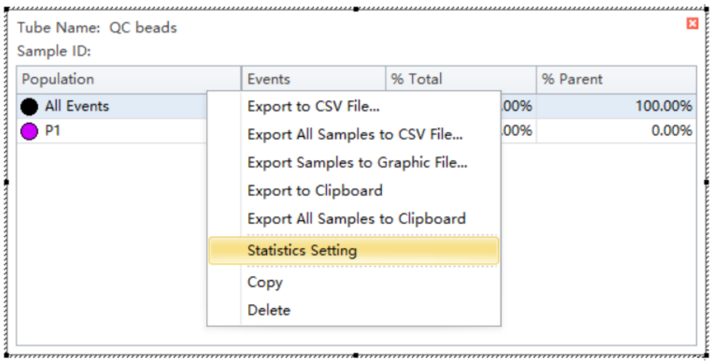
7 Create histograms for each channel and apply the gate.

---

8 Select **Run**.

---

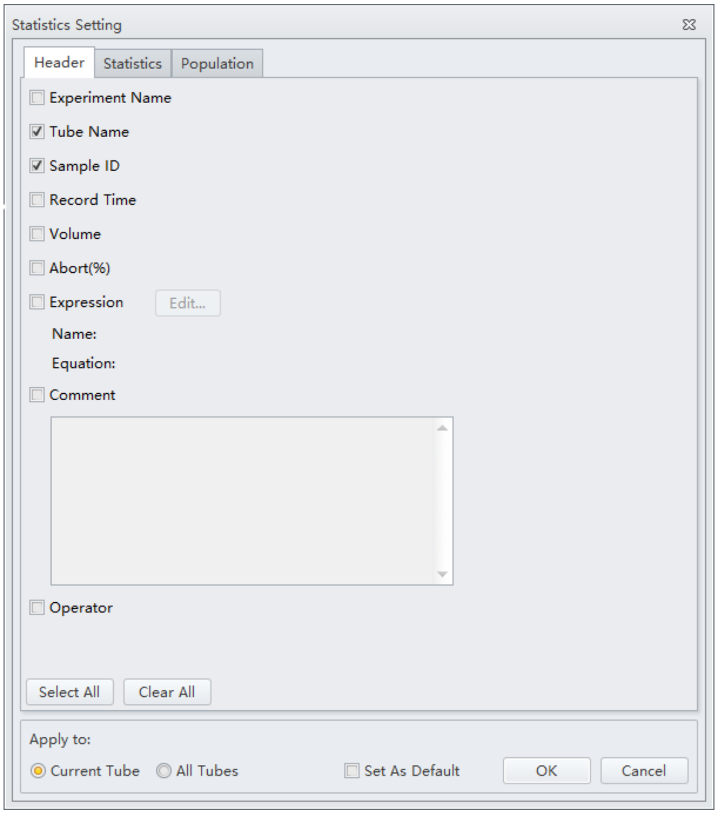
9 Add a statistics table and right-click the table and select **Statistics Setting**. The Statistics Setting window appears.



Tube Name: QC beads
Sample ID:

Population	Events	% Total	% Parent
● All Events		00%	100.00%
● P1		00%	0.00%

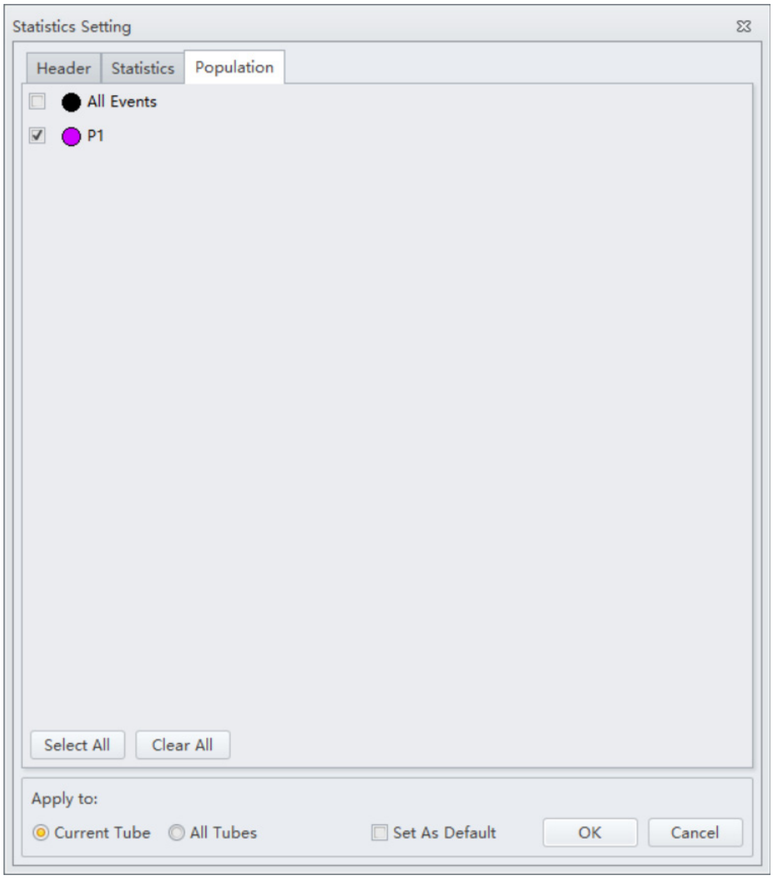
Export to CSV File...
Export All Samples to CSV File...
Export Samples to Graphic File...
Export to Clipboard
Export All Samples to Clipboard
Statistics Setting
Copy
Delete



☐ Experiment Name
☑ Tube Name
☑ Sample ID
☐ Record Time
☐ Volume
☐ Abort(%)
☐ Expression
Name:
Equation:
☐ Comment
☐ Operator

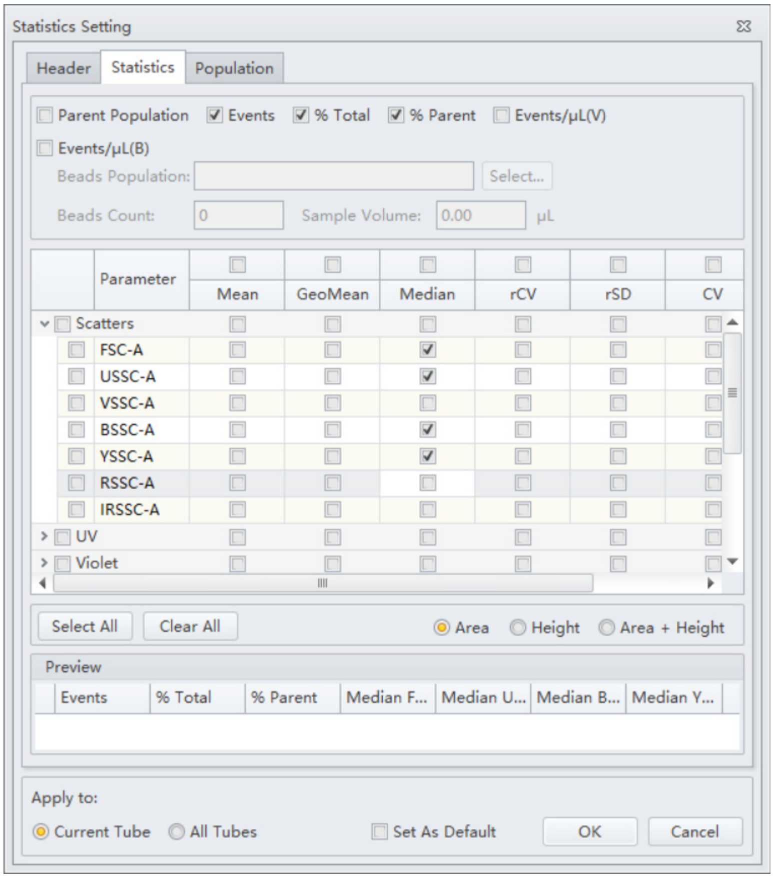
Apply to:
● Current Tube ○ All Tubes ☐ Set As Default

10 Select the **Population** tab and select the relevant population for the tube.



---

11 Select the **Statistics** tab then select the Median Fluorescence value for all parameters used and select **OK.**



☐ Parent Population ☑ Events ☑ % Total ☑ % Parent ☐ Events/µL(V)

☐ Events/µL(B)

Beads Population: Select...

Beads Count: 0 Sample Volume: 0.00 µL

Parameter	☐ Mean	☐ GeoMean	☐ Median	☐ rCV	☐ rSD	☐ CV
☐ Scatters	☐	☐	☐	☐	☐	☐
☐ FSC-A	☐	☐	☑	☐	☐	☐
☐ USSC-A	☐	☐	☑	☐	☐	☐
☐ VSSC-A	☐	☐	☐	☐	☐	☐
☐ BSSC-A	☐	☐	☑	☐	☐	☐
☐ YSSC-A	☐	☐	☑	☐	☐	☐
☐ RSSC-A	☐	☐	☐	☐	☐	☐
☐ IRSSC-A	☐	☐	☐	☐	☐	☐
☐ UV	☐	☐	☐	☐	☐	☐
☐ Violet	☐	☐	☐	☐	☐	☐

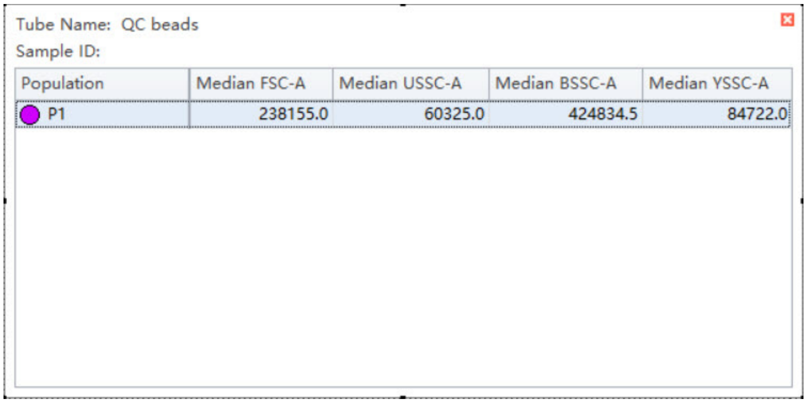
● Area ○ Height ○ Area + Height

Preview

Events	% Total	% Parent	Median F...	Median U...	Median B...	Median Y...

Apply to:

● Current Tube ○ All Tubes ☐ Set As Default



Tube Name: QC beads
Sample ID:

Population	Median FSC-A	Median USSC-A	Median BSSC-A	Median YSSC-A
● P1	238155.0	60325.0	424834.5	84722.0

NOTE The median values are the target settings that will be used for standardization.

---

12 Right-click the statistics table and select **Export to CSV File**.

If Excel is not available, manually record all the median values or take a screen shot.

---

13 Save the experiment.

NOTE Rerun the experiment and create a new standardization file if:

- You change the standardization fluorosphere used.
- The Lot number for the standardization fluorosphere is changed.

Creating a New Standardization Item

The standardization item can be created in two ways:

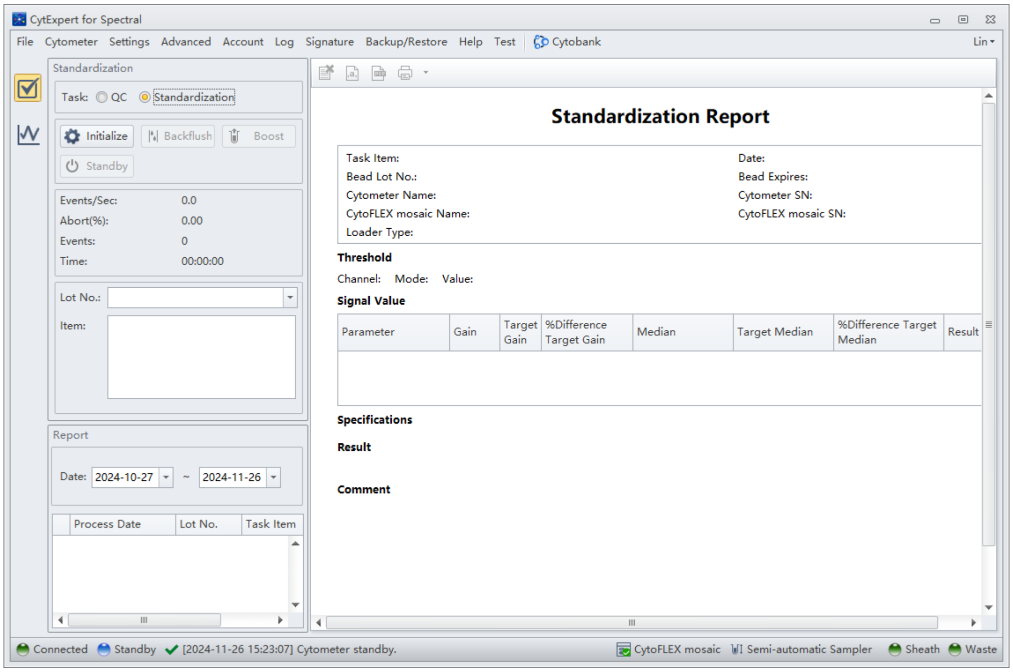
- Add a new standardization item in the Standardization Target Library using the previously generated target median values. Refer to Generating Target Median Values and Adding a New Standardization Item.
- Create a new standardization item from the Acq. Setting Catalog using the previously saved acquisition settings. Refer to CHAPTER 6, Exporting Instrument Settings and Creating a New Standardization Item from Acq. Setting Catalog.

Adding a New Standardization Item

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Adding a New Standardization Item for detailed instructions.

---

1 Select **Start QC/Standardization** in the QC/Standardization menu to access the Standardization screen.



Task: ○ QC ● Standardization

Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Lot No.:
Item:

Date: 2024-10-27 ~ 2024-11-26

Task Item:
Bead Lot No.:
Cytometer Name:
CytoFLEX mosaic Name:
Loader Type:
Date:
Bead Expires:
Cytometer SN:
CytoFLEX mosaic SN:

Threshold
Channel: Mode: Value:

Signal Value

Specifications

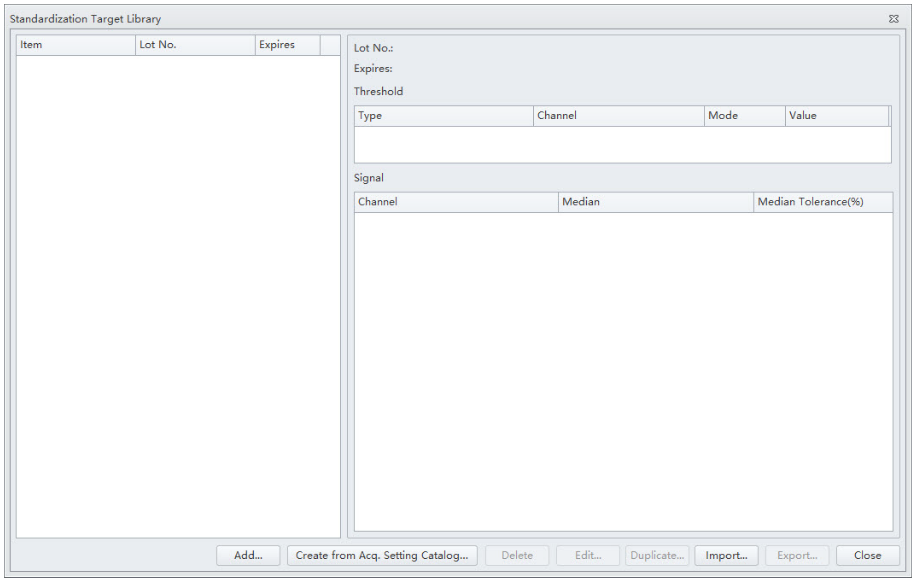
Result

Comment

[2024-11-26 15:23:07] Cytometer standby.

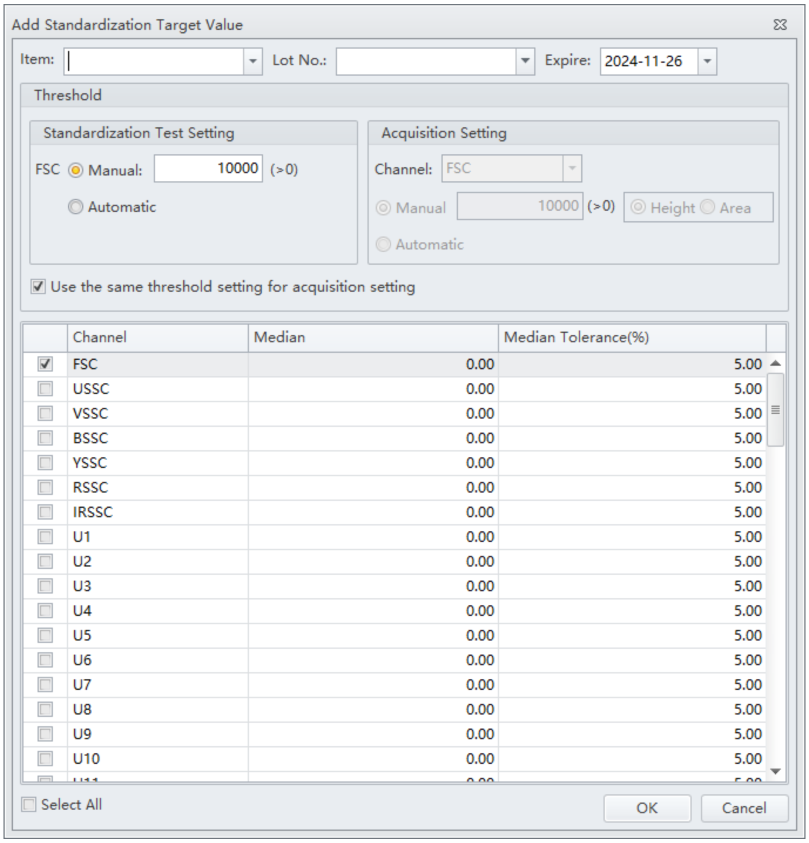
---

2 Select **Settings > Standardization Target Library.** The Standardization Target Library window appears.



---

3 Select **Add.** The Add Standardization Target Value window appears.


Add Standardization Target Value
Threshold
Standardization Test Setting
Acquisition Setting
Select All
OK
Cancel

Item:
Lot No.:
Expire: 2024-11-26

Standardization Test Setting: FSC ● Manual: 10000 (>0) ○ Automatic

Acquisition Setting: Channel: FSC; ● Manual 10000 (>0) ● Height ○ Area; ○ Automatic

☑ Use the same threshold setting for acquisition setting

	Channel	Median	Median Tolerance(%)
☑	FSC	0.00	5.00
☐	USSC	0.00	5.00
☐	VSSC	0.00	5.00
☐	BSSC	0.00	5.00
☐	YSSC	0.00	5.00
☐	RSSC	0.00	5.00
☐	IRSSC	0.00	5.00
☐	U1	0.00	5.00
☐	U2	0.00	5.00
☐	U3	0.00	5.00
☐	U4	0.00	5.00
☐	U5	0.00	5.00
☐	U6	0.00	5.00
☐	U7	0.00	5.00
☐	U8	0.00	5.00
☐	U9	0.00	5.00
☐	U10	0.00	5.00

☐ Select All

---

4 Enter the Item, Lot No. and Expiration date from the drop-down menu located at the top of the Add Standardization Target Value window.

NOTE A single Lot No. can include several Items, but you cannot add duplicate Items under the same Lot No.

NOTE If the Lot No. selected already exists, the Expiration date cannot be edited.

---

5 Select either **Manual** or **Automatic** threshold from the **Standardization Test Setting** or **Acquisition Setting** section of the window.

NOTE If you select **Manual** threshold, enter a value greater than 0, but less than 1,048,575.

NOTE Keep the threshold setting the same as previous Step 4 in Generating Target Median Values.

NOTE Unchecking the *Use the same threshold setting for acquisition setting* checkbox allows you to specify custom threshold settings and when to save the test item into the Acquisition Setting Catalog.

---

6 Enter the target median values saved in Step 11 in Generating Target Median Values into the corresponding channels.

Or copy the median values from the previously exported CSV file in Step 12 in Generating Target Median Values and paste into the corresponding median column,

NOTE Do not set the median tolerance range any lower than 5%.

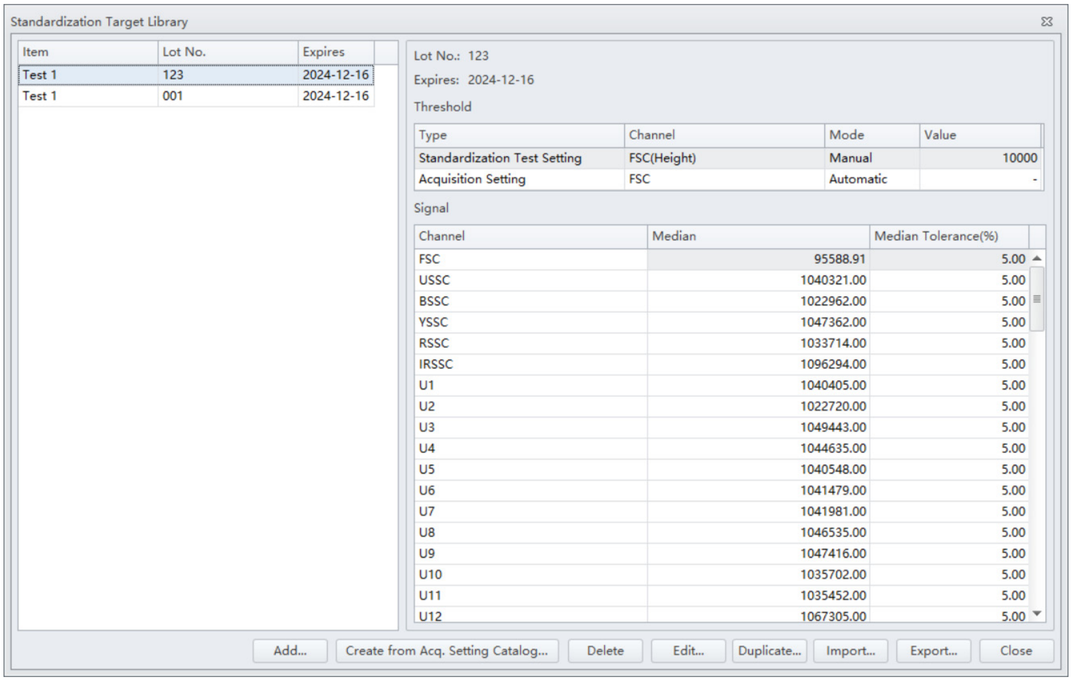
NOTE FSC is a required channel.

NOTE Verify that the target values are entered correctly.

---

7 Select **OK** to save the target value.

The saved results display in the Standardization Target Library window. This item is ready to be run through the Standardization experiment. Refer to Performing the Standardization.


Standardization Target Library
Add...
Create from Acq. Setting Catalog...
Delete
Edit...
Duplicate...
Import...
Export...
Close

Item	Lot No.	Expires
Test 1	123	2024-12-16
Test 1	001	2024-12-16

Lot No.: 123

Expires: 2024-12-16

Threshold

Type	Channel	Mode	Value
Standardization Test Setting	FSC(Height)	Manual	10000
Acquisition Setting	FSC	Automatic	-

Signal

Channel	Median	Median Tolerance(%)
FSC	95588.91	5.00
USSC	1040321.00	5.00
BSSC	1022962.00	5.00
YSSC	1047362.00	5.00
RSSC	1033714.00	5.00
IRSSC	1096294.00	5.00
U1	1040405.00	5.00
U2	1022720.00	5.00
U3	1049443.00	5.00
U4	1044635.00	5.00
U5	1040548.00	5.00
U6	1041479.00	5.00
U7	1041981.00	5.00
U8	1046535.00	5.00
U9	1047416.00	5.00
U10	1035702.00	5.00
U11	1035452.00	5.00
U12	1067305.00	5.00

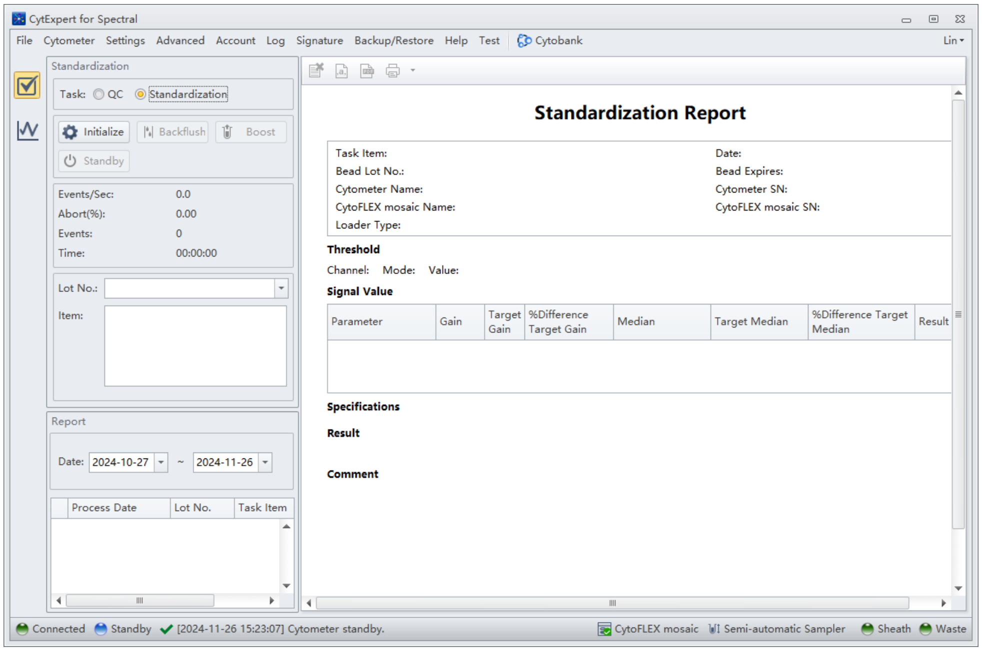
---

8 Select **Close** to exit the Standardization Target Library window,

Creating a New Standardization Item from Acq. Setting Catalog

---

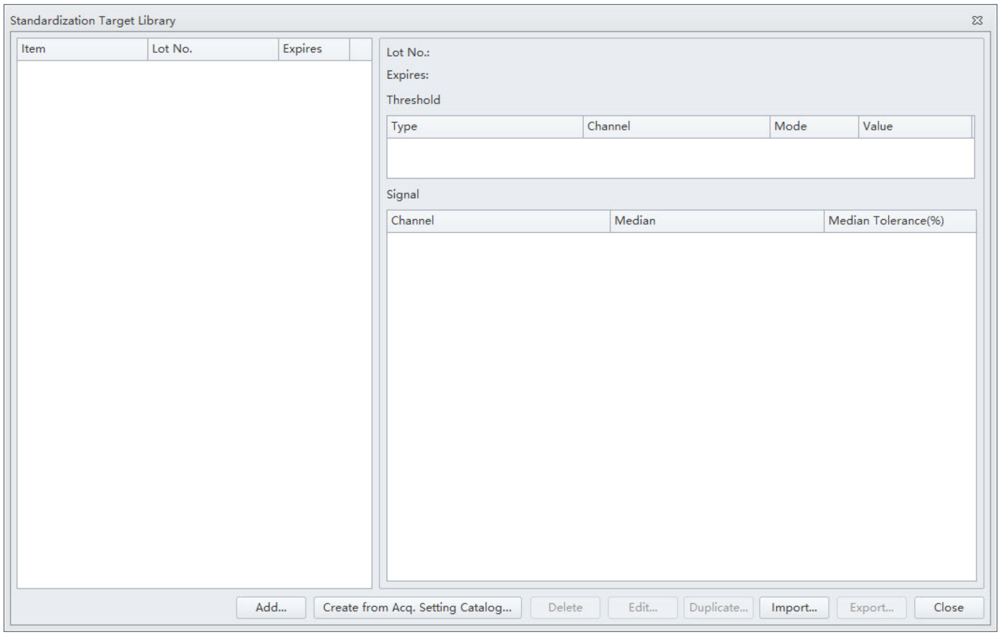
1 Select **Start QC/Standardization** in the QC/Standardization menu to access the Standardization screen.


CytExpert for Spectral
File Cytometer Settings Advanced Account Log Signature Backup/Restore Help Test Cytobank Lin
Standardization
Task: ○ QC ● Standardization
Initialize Backflush Boost
Standby
Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00
Lot No.:
Item:
Report
Date: 2024-10-27 ~ 2024-11-26
Process Date Lot No. Task Item
Standardization Report
Task Item: Date:
Bead Lot No.: Bead Expires:
Cytometer Name: Cytometer SN:
CytoFLEX mosaic Name: CytoFLEX mosaic SN:
Loader Type:
Threshold
Channel: Mode: Value:
Signal Value
Parameter Gain Target Gain %Difference Target Gain Median Target Median %Difference Target Median Result
Specifications
Result
Comment
Connected Standby
CytoFLEX mosaic Semi-automatic Sampler Sheath Waste

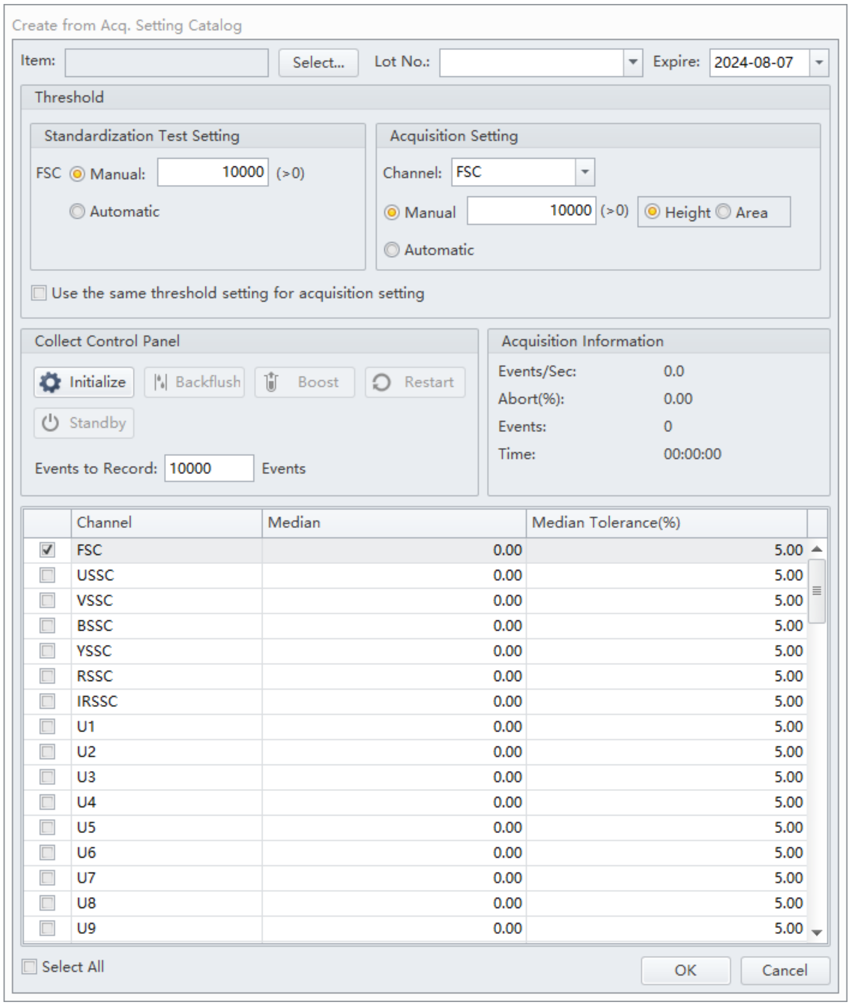
✓ [2024-11-26 15:23:07] Cytometer standby.

---

2 Select **Settings > Standardization Target Library.** The Standardization Target Library window appears.



3 Select Create from Acq. Setting Catalog... . The Create from Acq. Setting Catalog window appears.



Item:
Lot No.:
Expire: 2024-08-07

FSC ● Manual: 10000 (>0)
○ Automatic

Channel: FSC
● Manual 10000 (>0) ● Height ○ Area
○ Automatic

☐ Use the same threshold setting for acquisition setting

Events to Record: 10000 Events

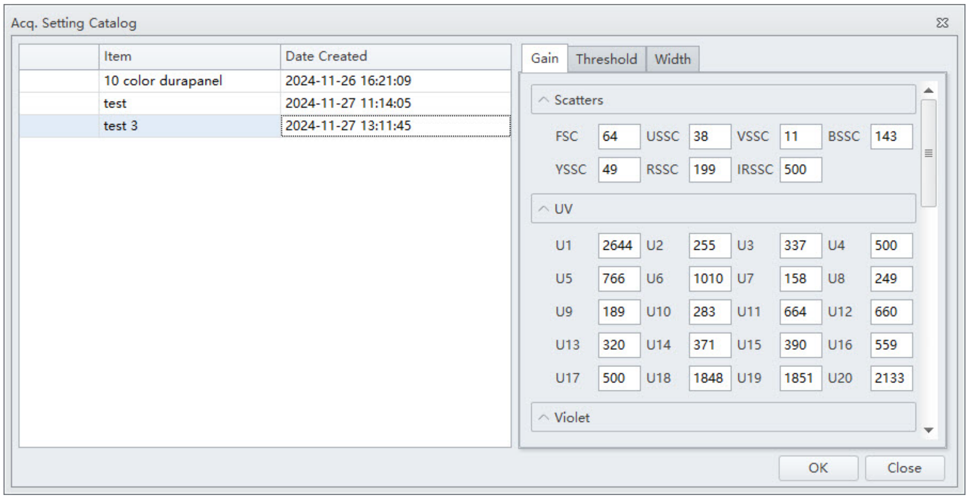
Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

	Channel	Median	Median Tolerance(%)
☑	FSC	0.00	5.00
☐	USSC	0.00	5.00
☐	VSSC	0.00	5.00
☐	BSSC	0.00	5.00
☐	YSSC	0.00	5.00
☐	RSSC	0.00	5.00
☐	IRSSC	0.00	5.00
☐	U1	0.00	5.00
☐	U2	0.00	5.00
☐	U3	0.00	5.00
☐	U4	0.00	5.00
☐	U5	0.00	5.00
☐	U6	0.00	5.00
☐	U7	0.00	5.00
☐	U8	0.00	5.00
☐	U9	0.00	5.00

☐ Select All

---

4 Select Select... located at the top left of the Create from Acq. Setting Catalog window. The Acq. Setting Catalog window appears.



Item	Date Created
10 color durapanel	2024-11-26 16:21:09
test	2024-11-27 11:14:05
test 3	2024-11-27 13:11:45

Scatters

FSC	64	USSC	38	VSSC	11	BSSC	143
YSSC	49	RSSC	199	IRSSC	500		

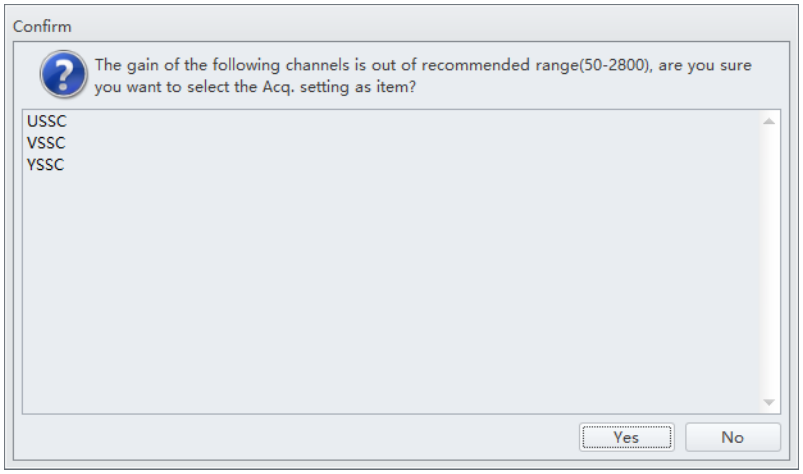
UV

U1	2644	U2	255	U3	337	U4	500
U5	766	U6	1010	U7	158	U8	249
U9	189	U10	283	U11	664	U12	660
U13	320	U14	371	U15	390	U16	559
U17	500	U18	1848	U19	1851	U20	2133

---

5 Select the desired Item and select **OK**.

NOTE If the following prompt appears, select **Yes** to proceed. If you do not want to proceed, select **No**.



The gain of the following channels is out of recommended range(50-2800), are you sure you want to select the Acq. setting as item?

USSC
VSSC
YSSC

---

6 Enter the Lot No. and Expiration date from the drop-down menu located at the top of the Create from Acq. Setting Catalog window.

NOTE A single Lot No. can include several Items, but you cannot add duplicate Items under the same Lot No.

NOTE If the Lot No. selected already exists, the Expiration date cannot be edited.

---

7 Select either **Manual** or **Automatic** threshold from the **Standardization Test Setting** or **Acquisition Setting** section of the window.

NOTE If you select **Manual** threshold, enter a value greater than 0, but less than 1,048,575.

NOTE Unchecking the *Use the same threshold setting for acquisition setting* checkbox allows you to specify custom threshold settings.

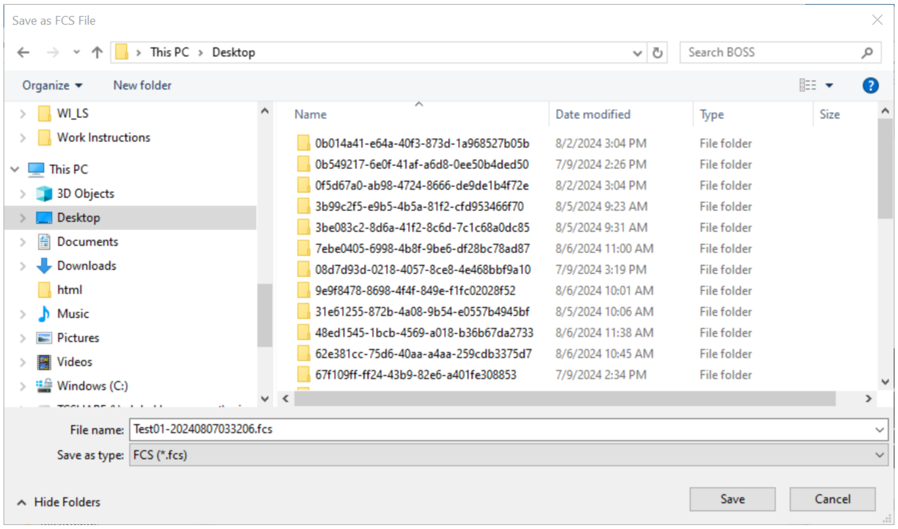
---

8 Select **Initialize** from the Collect Control Panel and select **Record**.

NOTE If you need to set the number of events to record in the Collect Control Panel, enter a value between 2,000 and 100,000.

---

9 Wait for the software to finish recording. The Save as FCS File window appears.



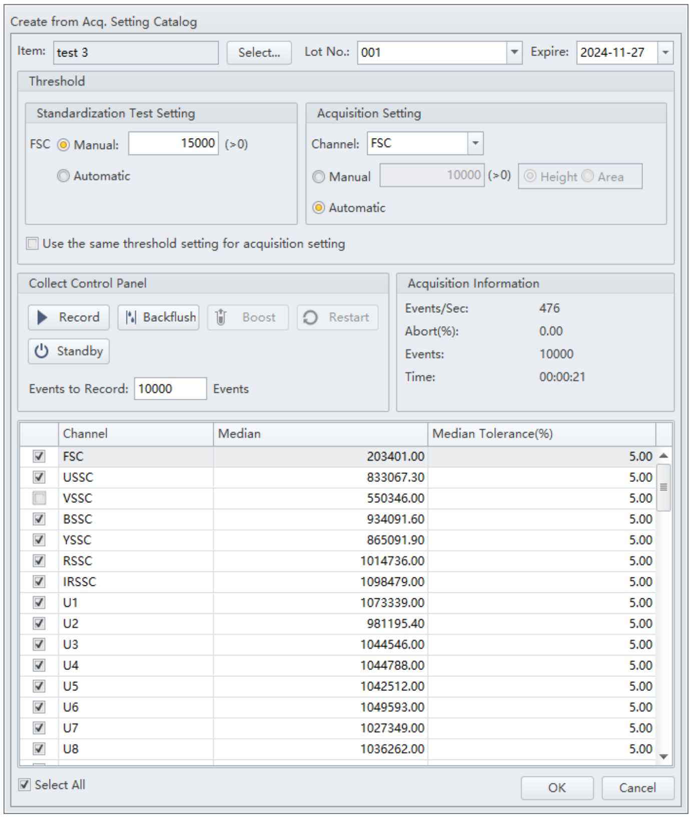
Name	Date modified	Type	Size
0b014a41-e64a-40f3-873d-1a968527b05b	8/2/2024 3:04 PM	File folder	
0b549217-6e0f-41af-a6d8-0ee50b4ded50	7/9/2024 2:26 PM	File folder	
0f5d67a0-ab98-4724-8666-de9de1b4f72e	8/2/2024 3:04 PM	File folder	
3b99c2f5-e9b5-4b5a-81f2-cfd953466f70	8/5/2024 9:23 AM	File folder	
3be083c2-8d6a-41f2-8c6d-7c1c68a0dc85	8/5/2024 9:31 AM	File folder	
7ebe0405-6998-4b8f-9be6-df28bc78ad87	8/6/2024 11:00 AM	File folder	
08d7d93d-0218-4057-8ce8-4e468bbf9a10	7/9/2024 3:19 PM	File folder	
9e9f8478-8698-4f4f-849e-f1fc02028f52	8/6/2024 10:01 AM	File folder	
31e61255-872b-4a08-9b54-e0557b4945bf	8/5/2024 10:06 AM	File folder	
48ed1545-1bcb-4569-a018-b36b67da2733	8/6/2024 11:38 AM	File folder	
62e381cc-75d6-40aa-a4aa-259cdb3375d7	8/6/2024 10:45 AM	File folder	
67f109ff-ff24-43b9-82e6-a401fe308853	7/9/2024 2:34 PM	File folder	

File name: Test01-20240807033206.fcs

Save as type: FCS (*.fcs)

If you want to export the FCS file, navigate to the desired file path and select **Save**. If you do not want to export the FCS file, select **Cancel**.

10 The generated median values display in the Create from Acq. Setting Catalog window.



Item: test 3
Lot No.: 001
Expire: 2024-11-27

FSC ● Manual: 15000 (>0)
○ Automatic

Channel: FSC
○ Manual 10000 (>0) ○ Height ○ Area
● Automatic

☐ Use the same threshold setting for acquisition setting

Events to Record: 10000 Events

Events/Sec: 476
Abort(%): 0.00
Events: 10000
Time: 00:00:21

	Channel	Median	Median Tolerance(%)
☑	FSC	203401.00	5.00
☑	USSC	833067.30	5.00
☐	VSSC	550346.00	5.00
☑	BSSC	934091.60	5.00
☑	YSSC	865091.90	5.00
☑	RSSC	1014736.00	5.00
☑	IRSSC	1098479.00	5.00
☑	U1	1073339.00	5.00
☑	U2	981195.40	5.00
☑	U3	1044546.00	5.00
☑	U4	1044788.00	5.00
☑	U5	1042512.00	5.00
☑	U6	1049593.00	5.00
☑	U7	1027349.00	5.00
☑	U8	1036262.00	5.00

☑ Select All

11 Select **OK** to save the target value and exit the Create from Acq. Setting Catalog window.

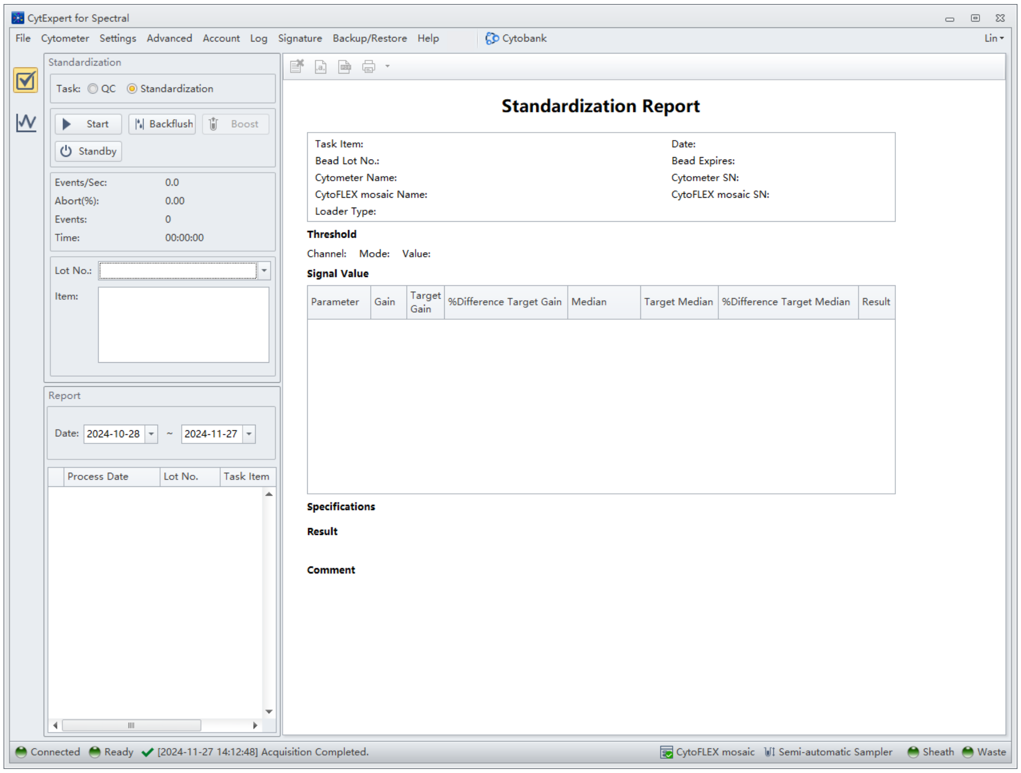
12 Select **Close** to exit the Standardization Target Library window.

Performing the Standardization

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Performing the Standardization for detailed instructions.

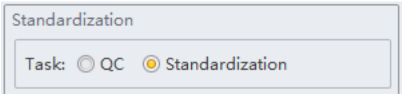
---

1 Select **Start QC/Standardization** in the QC/Standardization menu to access the Standardization screen.

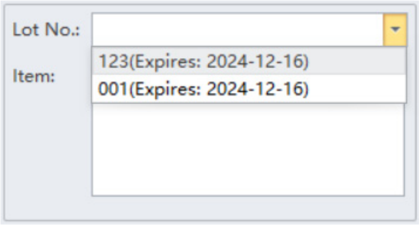


---

2 Select the **Standardization** radio button.



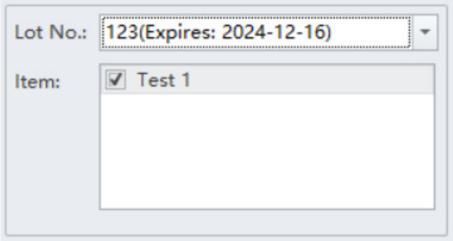
3 Select the correct Lot No. from the Lot No. drop-down menu.



123(Expires: 2024-12-16)
001(Expires: 2024-12-16)

NOTE Ensure the Lot No. corresponds to the standardization sample that generated the target median values.

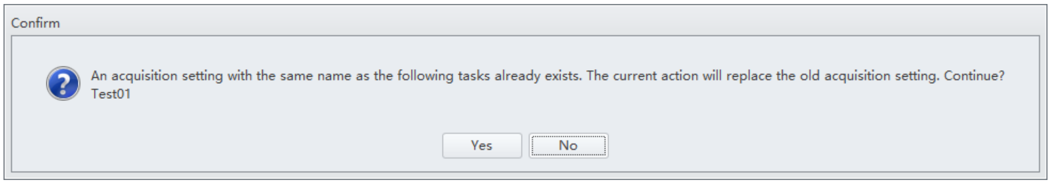
4 Select the Items to be standardized.


Lot No.: 123(Expires: 2024-12-16)

Item: ☑ Test 1

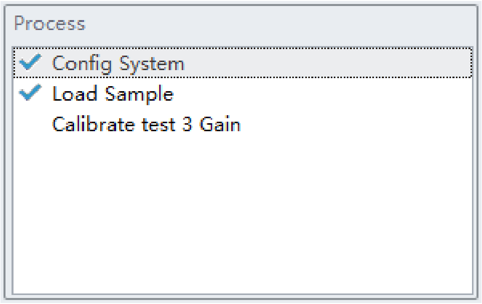
5 Select **Initialize.**

6 Select **Start**. The following software prompt appears. Select **Yes.**



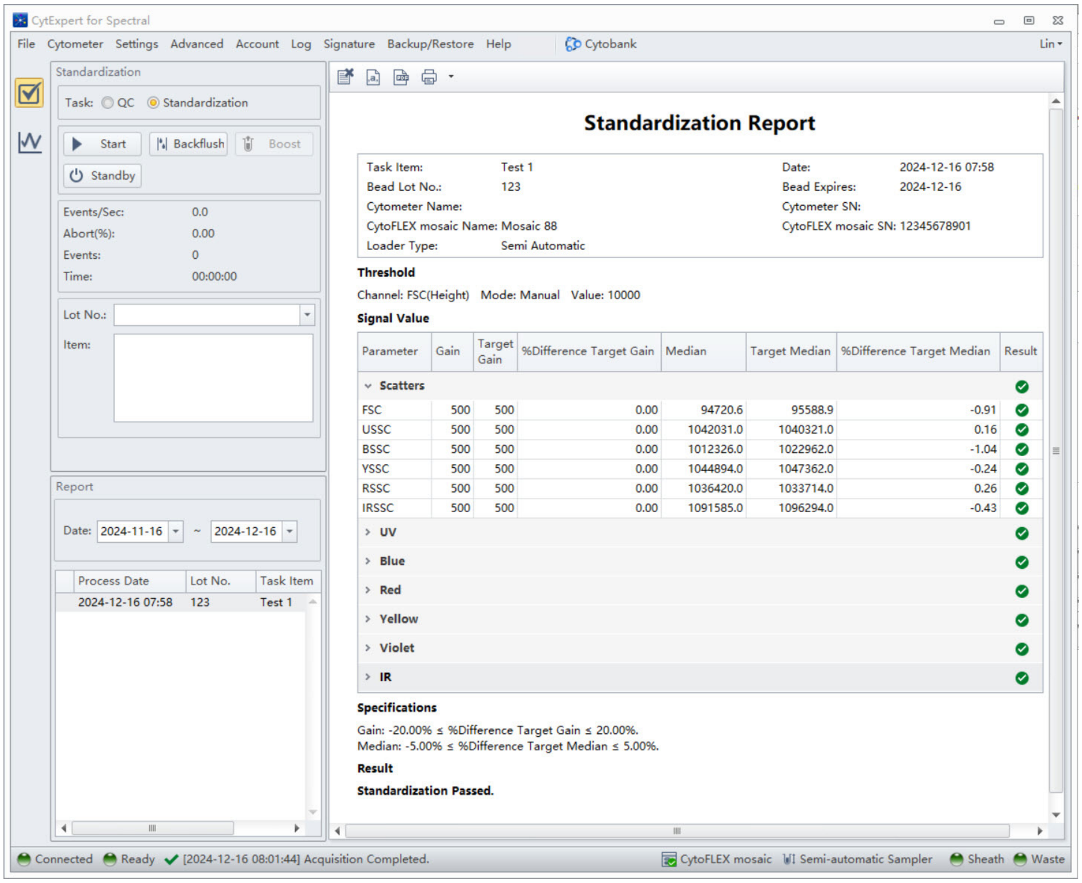
An acquisition setting with the same name as the following tasks already exists. The current action will replace the old acquisition setting. Continue?
Test01

The Process section of the screen displays the process details.



✓ Config System
✓ Load Sample
Calibrate test 3 Gain

Once the process is complete, the standardization results displays in the Process list on the left and a Standardization Report displays on the right.



Task: ○ QC ● Standardization

Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Lot No.:
Item:

Date: 2024-11-16 ~ 2024-12-16

Process Date	Lot No.	Task Item
2024-12-16 07:58	123	Test 1

Standardization Report

Task Item: Test 1
Bead Lot No.: 123
Cytometer Name:
CytoFLEX mosaic Name: Mosaic 88
Loader Type: Semi Automatic
Date: 2024-12-16 07:58
Bead Expires: 2024-12-16
Cytometer SN:
CytoFLEX mosaic SN: 12345678901

Threshold

Channel: FSC(Height) Mode: Manual Value: 10000

Signal Value

Parameter	Gain	Target Gain	%Difference Target Gain	Median	Target Median	%Difference Target Median	Result
Scatters							✓
FSC	500	500	0.00	94720.6	95588.9	-0.91	✓
USSC	500	500	0.00	1042031.0	1040321.0	0.16	✓
BSSC	500	500	0.00	1012326.0	1022962.0	-1.04	✓
YSSC	500	500	0.00	1044894.0	1047362.0	-0.24	✓
RSSC	500	500	0.00	1036420.0	1033714.0	0.26	✓
IRSSC	500	500	0.00	1091585.0	1096294.0	-0.43	✓
UV							✓
Blue							✓
Red							✓
Yellow							✓
Violet							✓
IR							✓

Specifications

Gain: -20.00% ≤ %Difference Target Gain ≤ 20.00%.
Median: -5.00% ≤ %Difference Target Median ≤ 5.00%.

Result

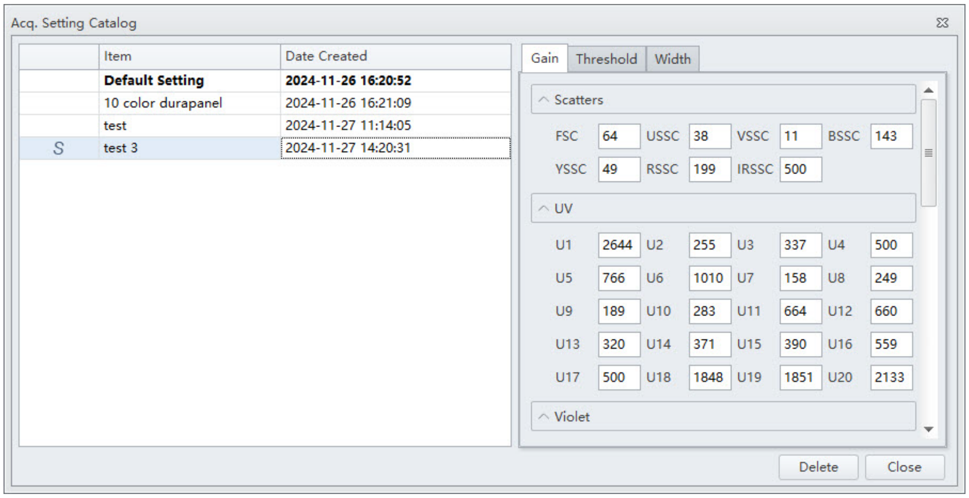
Standardization Passed.

Connected Ready ✓ [2024-12-16 08:01:44] Acquisition Completed. CytoFLEX mosaic Semi-automatic Sampler Sheath Waste

NOTE The result column indicates a passing standardization result with a ✓ and a failed standardization result with ✗.

NOTE The updated Standardization item is added to the Acquisition Catalog automatically and overwrites the previously existing standardized settings for this item.

7 Select **Acq. Setting Catalog** from the Cytometer menu to verify the gain settings. The Acq. Setting Catalog window appears.



Acq. Setting Catalog

	Item	Date Created
	Default Setting	**2024-11-26 16:20:52**
	10 color durapanel	2024-11-26 16:21:09
	test	2024-11-27 11:14:05
S	test 3	2024-11-27 14:20:31

Scatters

FSC 64, USSC 38, VSSC 11, BSSC 143
YSSC 49, RSSC 199, IRSSC 500

UV

U1 2644, U2 255, U3 337, U4 500
U5 766, U6 1010, U7 158, U8 249
U9 189, U10 283, U11 664, U12 660
U13 320, U14 371, U15 390, U16 559
U17 500, U18 1848, U19 1851, U20 2133

Violet

NOTE S designates test items from Standardization.

8 Select **Close** to exit the Acq. Setting Catalog window.

9 Run Daily Clean. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Daily Clean.

10 Select **Close QC/Standardization**.

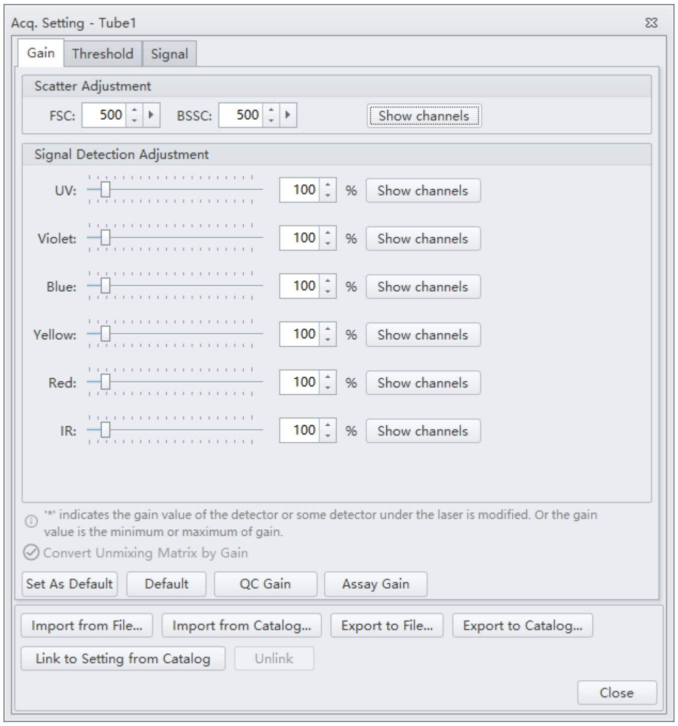
Applying the Standardized Acquisition Settings

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Applying the Standardized Acquisition Settings for detailed instructions.

1 Open an experiment.

---

2 Select **Acq. Setting** from the Cytometer menu. The Acq. Setting window appears.



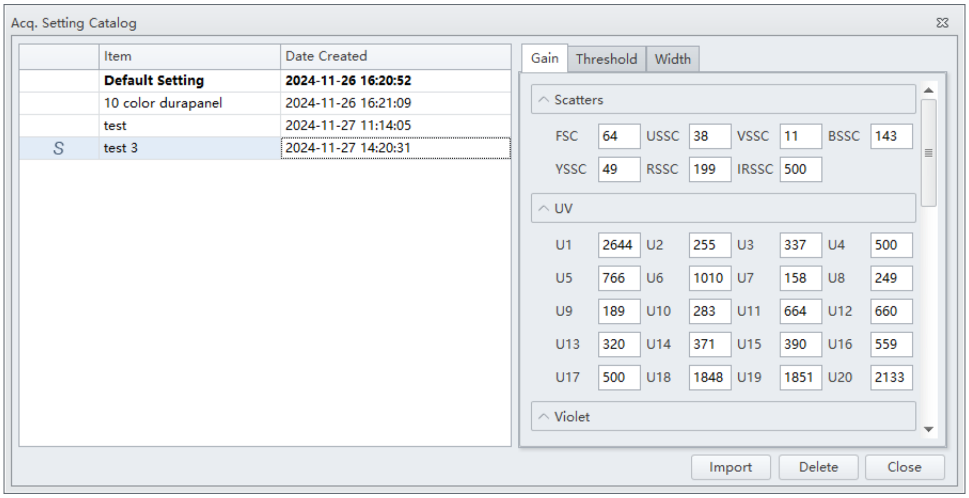
FSC: 500
BSSC: 500

UV: 100 %
Violet: 100 %
Blue: 100 %
Yellow: 100 %
Red: 100 %
IR: 100 %

'*' indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

---

3 Select **Import from Catalog**. The Acq. Setting Catalog window appears.



	Item	Date Created
	Default Setting	**2024-11-26 16:20:52**
	10 color durapanel	2024-11-26 16:21:09
	test	2024-11-27 11:14:05
S	test 3	2024-11-27 14:20:31

Scatters

FSC 64 USSC 38 VSSC 11 BSSC 143
YSSC 49 RSSC 199 IRSSC 500

UV

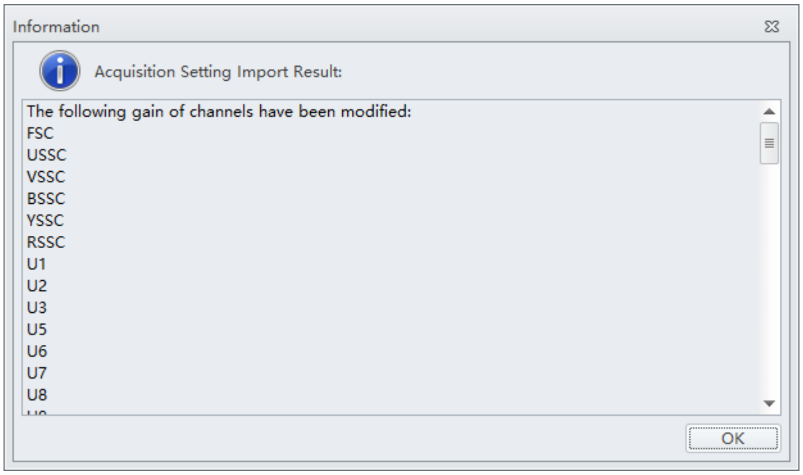
U1 2644 U2 255 U3 337 U4 500
U5 766 U6 1010 U7 158 U8 249
U9 189 U10 283 U11 664 U12 660
U13 320 U14 371 U15 390 U16 559
U17 500 U18 1848 U19 1851 U20 2133

---

4 Browse for the item to import and select **Import.**

The standardized settings are applied to the sample tube.

The Information window appears to notify of the corresponding channels with the changed gain as a result of the Standardization.



The following gain of channels have been modified:
FSC
USSC
VSSC
BSSC
YSSC
RSSC
U1
U2
U3
U5
U6
U7
U8

5 Select **OK**.

Standardization Target Library

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Standardization Target Library for detailed instructions on the following procedures:

- Importing a Standardization Item
- Exporting a Standardization Item

  NOTE The standardization items save as .mstgt file. This file can be used to standardize the settings between different instruments with the same laser configuration.

- Editing Standardization Item Parameters
- Duplicating Standardization Items
- Deleting Standardization Items

CHAPTER 6

Data Acquisition and Sample Analysis

Overview

This chapter contains information on how to use your Cytometer with CytoFLEX mosaic Spectral Detection Module, including data acquisition, analyzing and exporting results.

Workflow:

Create experiment → Adjust instrument settings and gates → Load sample → Record Data → Analyze and export data → Save experiment

This chapter contains information on:

- Creating an Experiment
- Configuring Acquisition Settings
- Load Sample and Record Data
- Analyzing and Exporting Data
- Saving the Experiment

Creating an Experiment

Creating an Experiment [Without Plate Loader]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Creating an Experiment [Without Plate Loader] for detailed instructions.

NOTE CytExpert for Spectral software has two installation options, the CytExpert for Spectral User Management software option and the CytExpert for Spectral Electronic Record Management software option.

Creating an Experiment [With Plate Loader]

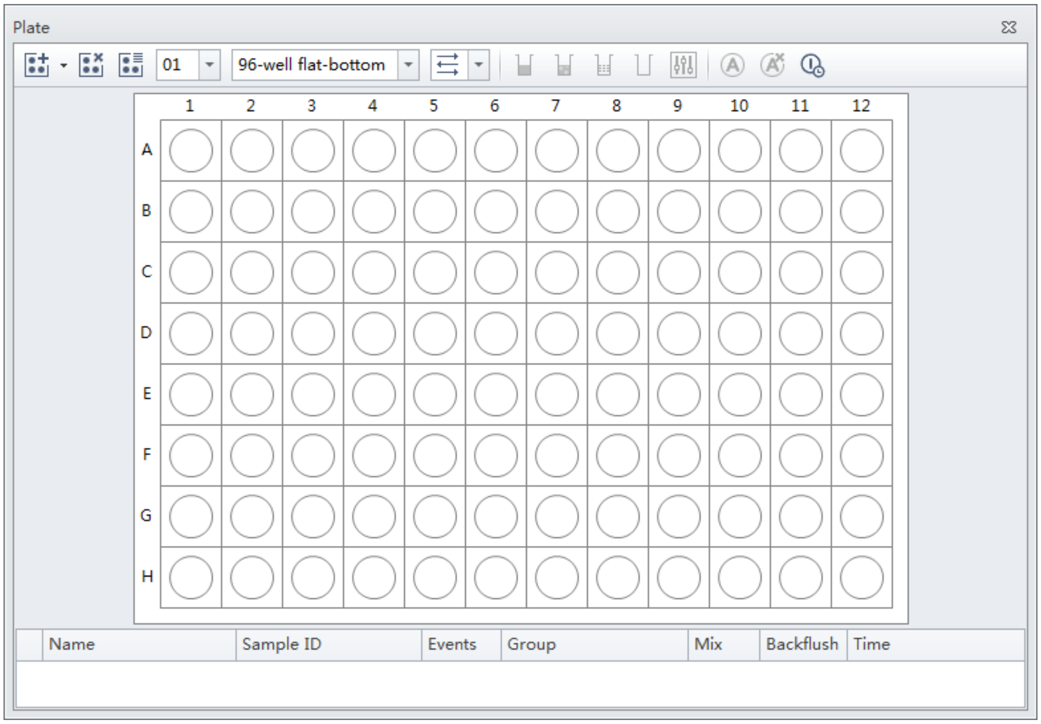
This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Creating an Experiment [With Plate Loader] for detailed instructions.

Setting Sample Wells

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting Sample Wells for detailed instructions.

Once the plate protocol is created, the plate window appears. Refer to Figure 6.1.

Figure 6.1 Plate Window [CytoFLEX LX Shown]

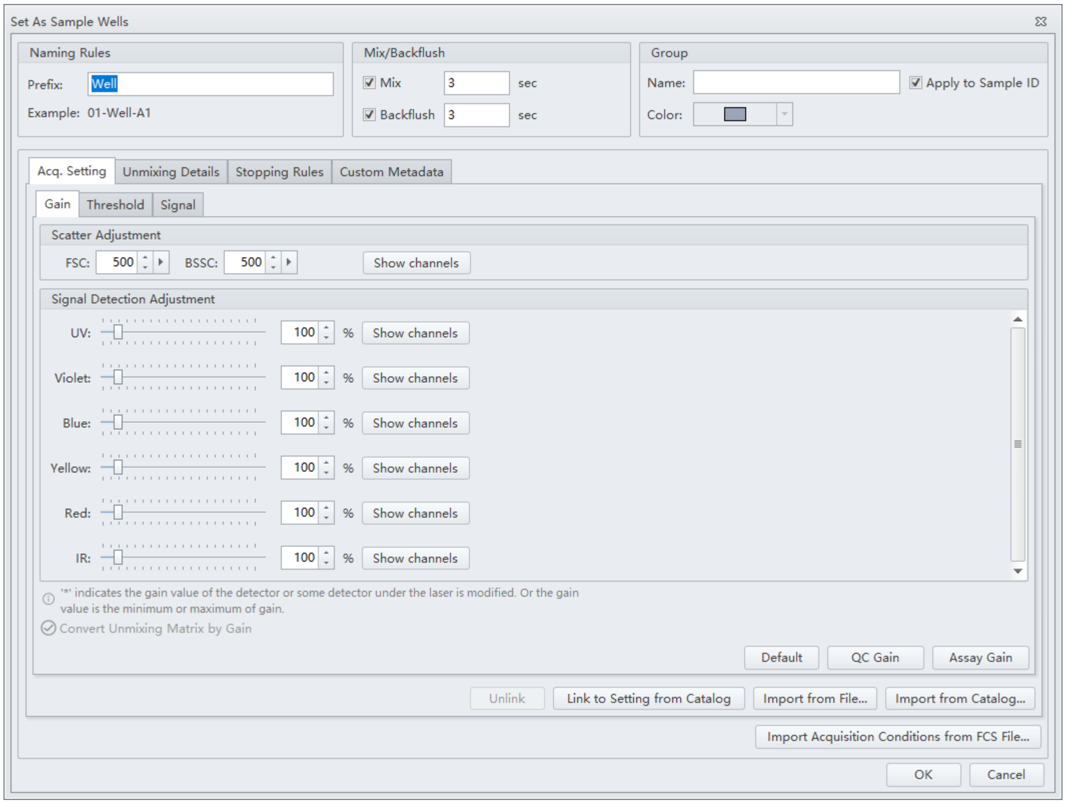


○	Empty well.
●	The well with color is set as sample well, but is not set for Auto Record.
● 3	The well is set as sample well, and is ready for Auto Record. The number labeled at the right bottom shows the order of auto record.
🟠	Cleaning Agent well.
🔵	Deionized water well.

✓	A blue check mark means the data is acquired, not recorded.
✓	A green check mark means the data is recorded.
✗	A red cross mark means the data is recorded by Auto Record mode. But the acquisition is terminated abnormally. For example, the well is skipped or the acquisition is manually stopped.

1 Left-click and drag your mouse to highlight the desired wells or hold the Control key and select each desired well.

2 Select or right-click the selected wells and select **Set As Sample Wells**. The Set As Sample Wells window appears.


Set As Sample Wells

Naming Rules
Prefix: Well
Example: 01-Well-A1

Mix/Backflush
☑ Mix 3 sec
☑ Backflush 3 sec

Group
Name:
☑ Apply to Sample ID
Color:

Scatter Adjustment
FSC: 500
BSSC: 500

Signal Detection Adjustment
UV: 100 %
Violet: 100 %
Blue: 100 %
Yellow: 100 %
Red: 100 %
IR: 100 %

'*' indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.
Convert Unmixing Matrix by Gain

NOTE Select or right-click and select **Set As Empty Wells** to reset selected wells as empty.

3 Enter the name in the Prefix box in the Naming Rules section of the window.

4 Select the desired Mix and Backflush duration from the Mix/Backflush section of the window.

---

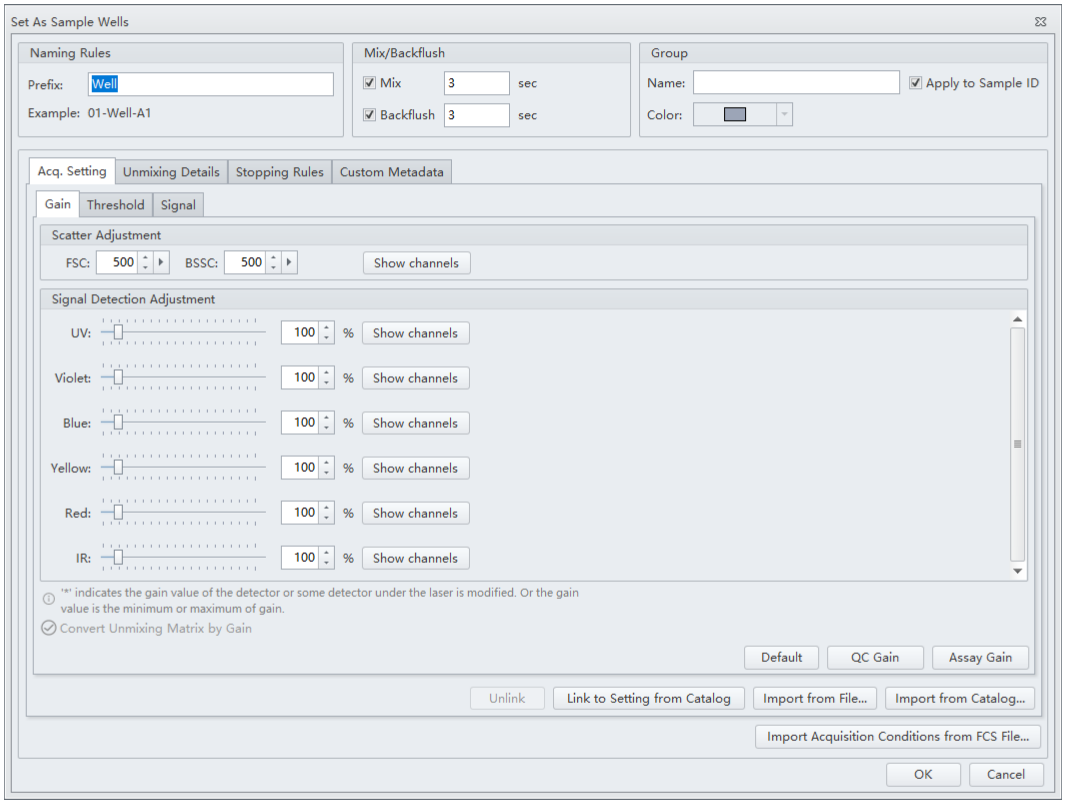
5 Enter the Group Name in the Name box in the Group section of the window.

---

6 Select the sample well color using the color dropdown under the Group section of the window.

---

7 Select the desired acquisition settings under the Acq. Setting tab.



Set As Sample Wells

Naming Rules

Prefix: Well

Example: 01-Well-A1

Mix/Backflush

☑ Mix 3 sec

☑ Backflush 3 sec

Group

Name:

☑ Apply to Sample ID

Color:

Scatter Adjustment

FSC: 500

BSSC: 500

Signal Detection Adjustment

UV: 100 %

Violet: 100 %

Blue: 100 %

Yellow: 100 %

Red: 100 %

IR: 100 %

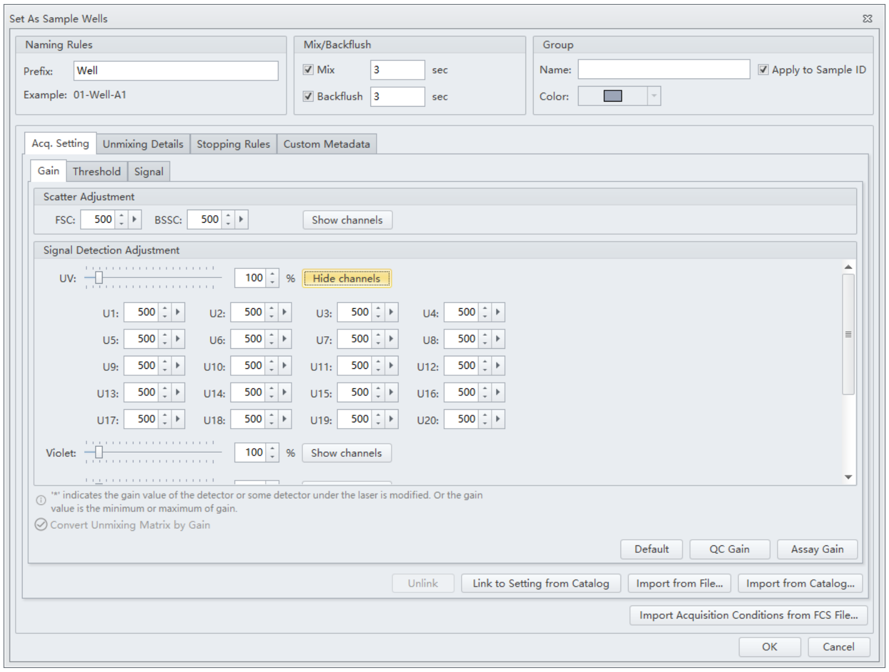
'*' indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

NOTE Select **Import from File** to import the settings from a FCS file.

NOTE If desired, import saved settings/standardization settings from the catalog.

---

8 Select the channels and adjust as needed.



Set As Sample Wells

Naming Rules
Prefix: Well
Example: 01-Well-A1

Mix/Backflush
☑ Mix 3 sec
☑ Backflush 3 sec

Group
Name:
☑ Apply to Sample ID
Color:

Acq. Setting | Unmixing Details | Stopping Rules | Custom Metadata

Gain | Threshold | Signal

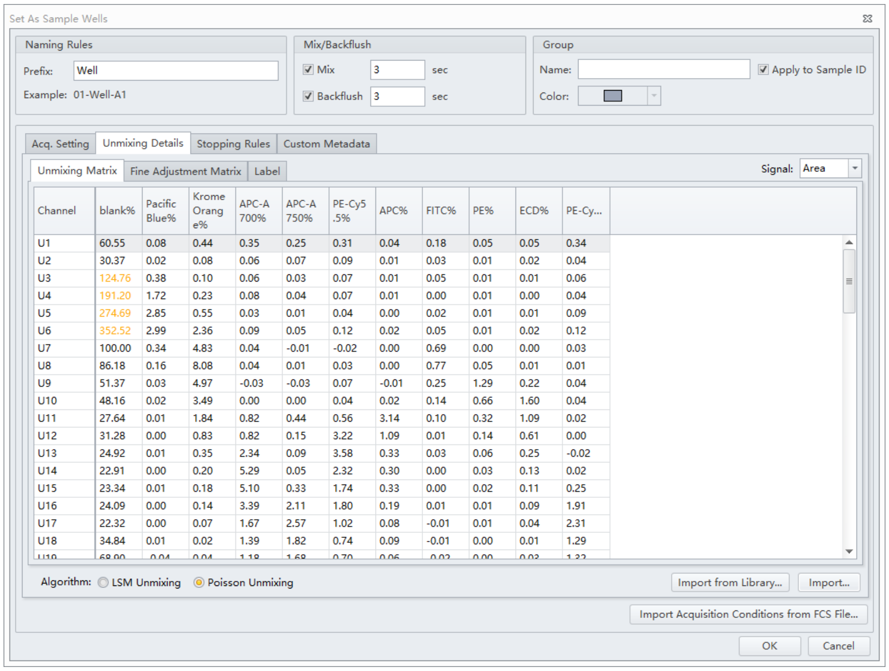
Scatter Adjustment
FSC: 500 BSSC: 500 Show channels

Signal Detection Adjustment
UV: 100 % Hide channels
U1: 500 U2: 500 U3: 500 U4: 500
U5: 500 U6: 500 U7: 500 U8: 500
U9: 500 U10: 500 U11: 500 U12: 500
U13: 500 U14: 500 U15: 500 U16: 500
U17: 500 U18: 500 U19: 500 U20: 500
Violet: 100 % Show channels

"*" indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.
Convert Unmixing Matrix by Gain

---

9 Set unmixing under the Unmixing Details tab. Refer to CHAPTER 7, Unmixing for detailed instructions on setting unmixing.



Set As Sample Wells

Naming Rules
Prefix: Well
Example: 01-Well-A1

Mix/Backflush
☑ Mix 3 sec
☑ Backflush 3 sec

Group
Name:
☑ Apply to Sample ID
Color:

Acq. Setting | Unmixing Details | Stopping Rules | Custom Metadata

Unmixing Matrix | Fine Adjustment Matrix | Label

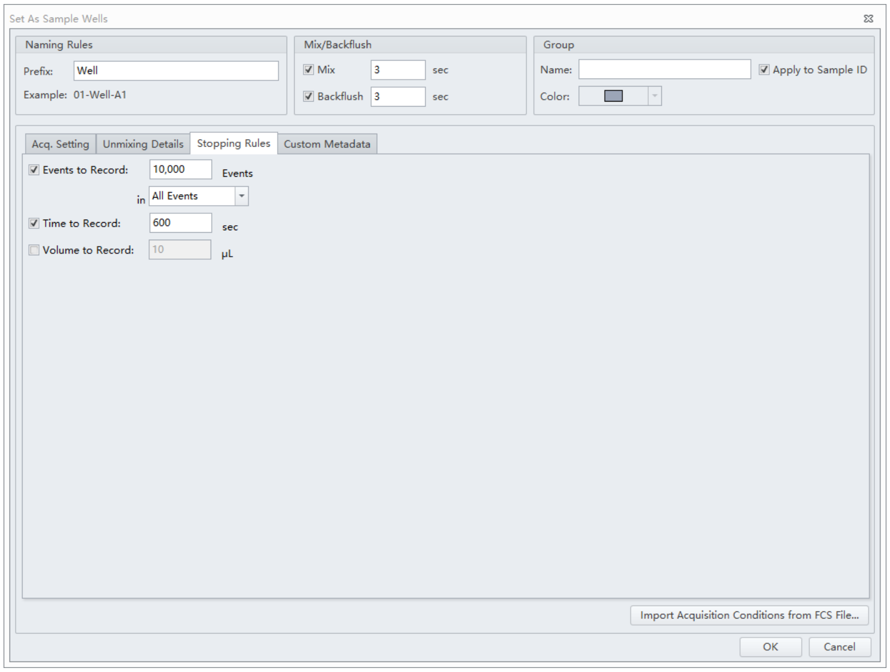
Signal: Area

Channel	blank%	Pacific Blue%	Krome Orange%	APC-A 700%	APC-A 750%	PE-Cy5.5%	APC%	FITC%	PE%	ECD%	PE-Cy...
U1	60.55	0.08	0.44	0.35	0.25	0.31	0.04	0.18	0.05	0.05	0.34
U2	30.37	0.02	0.08	0.06	0.07	0.09	0.01	0.03	0.01	0.02	0.04
U3	124.76	0.38	0.10	0.06	0.03	0.07	0.01	0.05	0.01	0.01	0.06
U4	191.20	1.72	0.23	0.08	0.04	0.07	0.01	0.00	0.01	0.00	0.04
U5	274.69	2.85	0.55	0.03	0.01	0.04	0.00	0.02	0.01	0.01	0.09
U6	352.52	2.99	2.36	0.09	0.05	0.12	0.02	0.05	0.01	0.02	0.12
U7	100.00	0.34	4.83	0.04	-0.01	-0.02	0.00	0.69	0.00	0.00	0.03
U8	86.18	0.16	8.08	0.04	0.01	0.03	0.00	0.77	0.05	0.01	0.01
U9	51.37	0.03	4.97	-0.03	-0.03	0.07	-0.01	0.25	1.29	0.22	0.04
U10	48.16	0.02	3.49	0.00	0.00	0.04	0.02	0.14	0.66	1.60	0.04
U11	27.64	0.01	1.84	0.82	0.44	0.56	3.14	0.10	0.32	1.09	0.02
U12	31.28	0.00	0.83	0.82	0.15	3.22	1.09	0.01	0.14	0.61	0.00
U13	24.92	0.01	0.35	2.34	0.09	3.58	0.33	0.03	0.06	0.25	-0.02
U14	22.91	0.00	0.20	5.29	0.05	2.32	0.30	0.00	0.03	0.13	0.02
U15	23.34	0.01	0.18	5.10	0.33	1.74	0.33	0.00	0.02	0.11	0.25
U16	24.09	0.00	0.14	3.39	2.11	1.80	0.19	0.01	0.01	0.09	1.91
U17	22.32	0.00	0.07	1.67	2.57	1.02	0.08	-0.01	0.01	0.04	2.31
U18	34.84	0.01	0.02	1.39	1.82	0.74	0.09	-0.01	0.00	0.01	1.29

Algorithm: ○ LSM Unmixing ● Poisson Unmixing

---

10 Select **Events to Record**, **Time to Record**, or **Volume to Record** under the Stopping Rules tab.



Set As Sample Wells

Naming Rules
Prefix: Well
Example: 01-Well-A1

Mix/Backflush
☑ Mix 3 sec
☑ Backflush 3 sec

Group
Name:
☑ Apply to Sample ID
Color:

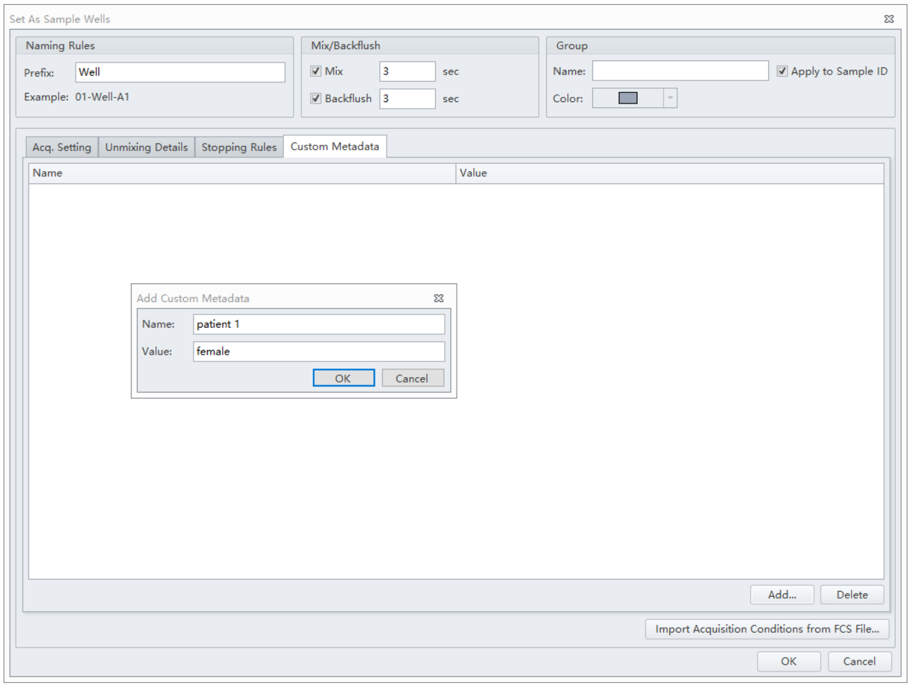
Acq. Setting | Unmixing Details | Stopping Rules | Custom Metadata

☑ Events to Record: 10,000 Events
in All Events
☑ Time to Record: 600 sec
☐ Volume to Record: 10 µL

NOTE Beckman Coulter recommends setting an acquisition time limit to stop the acquisition if the event limit cannot be reached.

---

11 Create the desired name and value under the Custom Metadata tab.



Set As Sample Wells

Naming Rules
Prefix: Well
Example: 01-Well-A1

Mix/Backflush
☑ Mix 3 sec
☑ Backflush 3 sec

Group
Name:
☑ Apply to Sample ID
Color:

Acq. Setting | Unmixing Details | Stopping Rules | Custom Metadata

Name | Value

Add Custom Metadata
Name: patient 1
Value: female

12 Select **OK**.

Modifying Well Settings

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Modifying Well Settings for detailed instructions on the following procedures:

- Copying, Cutting, and Pasting Wells
- Applying Existing Well Settings to Additional Wells
- Moving the Location of a Well

Setting the Label

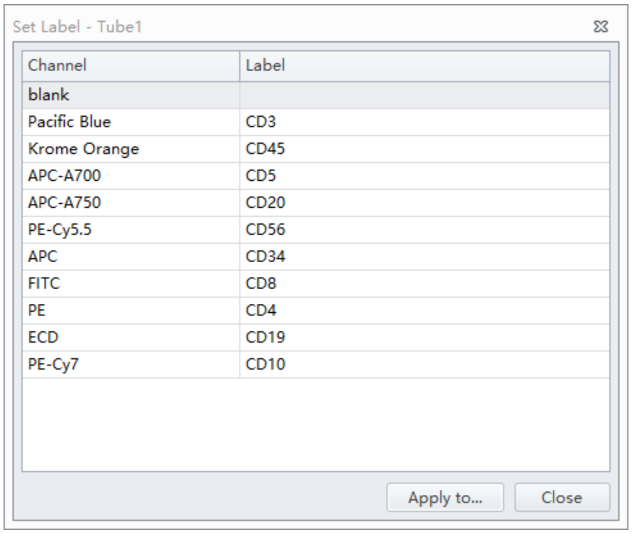
NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting the Label for detailed instructions.

NOTE The Set Label function is not available in spectral unmixing experiments.

NOTE The Set Label function is only available when the tube contains the unmixing matrix. To import unmixing matrix, refer to CHAPTER 7, Importing and Exporting Unmixing Matrix.

1 Select **Set Label** in the Settings menu. The Set Label window appears.

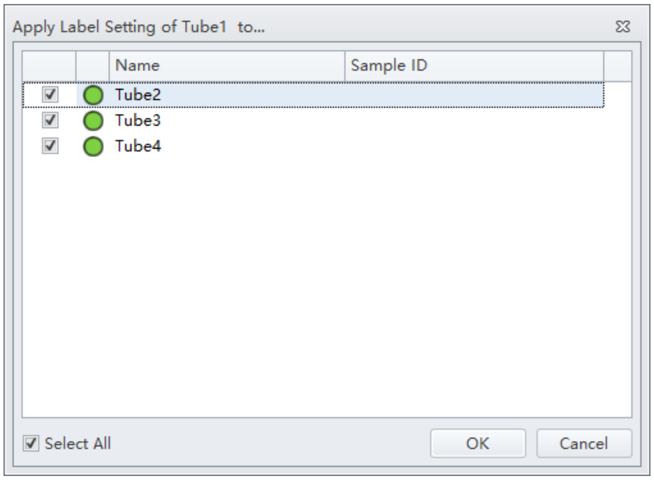
2 In the Set Label window, modify the label as needed.



Channel	Label
blank	
Pacific Blue	CD3
Krome Orange	CD45
APC-A700	CD5
APC-A750	CD20
PE-Cy5.5	CD56
APC	CD34
FITC	CD8
PE	CD4
ECD	CD19
PE-Cy7	CD10

---

3 Select **Apply to.** The Apply Label Setting window appears.



Apply Label Setting of Tube1 to...

		Name	Sample ID
☑	●	Tube2	
☑	●	Tube3	
☑	●	Tube4	

☑ Select All

---

4 Select the tubes to apply the label settings to and select **OK**.

NOTE The **Select All** checkbox allows you to select all of the tubes listed.

---

5 Select **Close** to exit the Set Label window.

Creating Plots and Gates

IMPORTANT The maximum number of elements allowed in an experiment is 200. Elements include plots, statistics tables, and gate hierarchy tables.

IMPORTANT The maximum number of gates allowed in an experiment is 200.

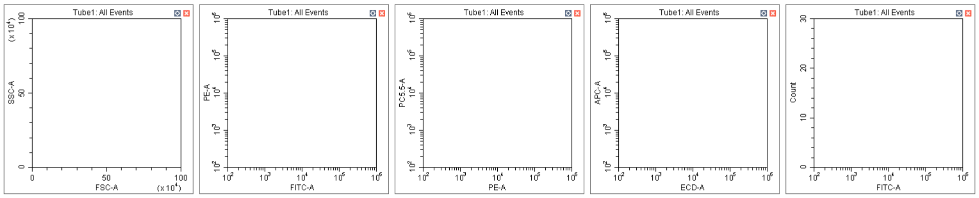
NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Creating Plots and Gates for detailed instructions.

---

1 Use the plotting controls (see Figure 2.3) in the plot area to create plots and gates and to generate graphs as shown.

Use the icons to generate histograms, dot plots, density plots, pseudo color plots, contour plots, and spectrum plots.

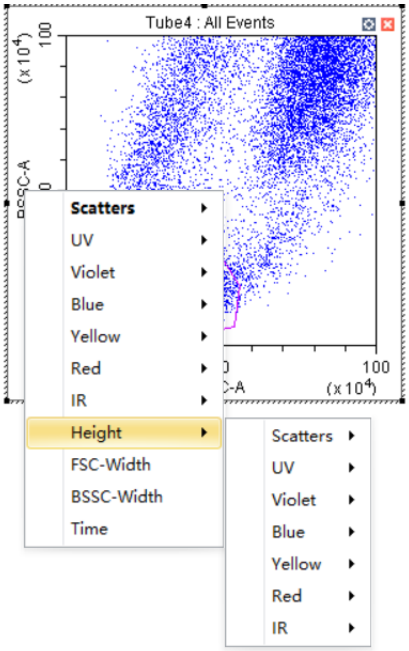
The experiment uses scatter plots, histograms, polygon gating, four-quadrant gating, and line-segment gating.



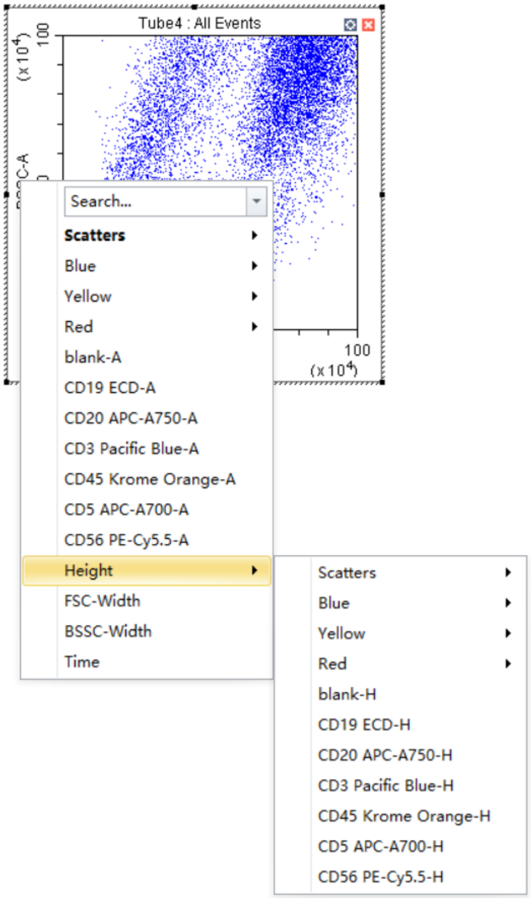
a. After selecting a plot, click and drag the mouse to adjust the position and select and drag the sizing handles at the edge of the graph to adjust the size of the graph.

b. Select an axis name to change which channel is displayed. An “A” after the channel name indicates signal pulse area, while an “H” indicates height. The default setting is “A”.

[Raw Worksheet Shown]



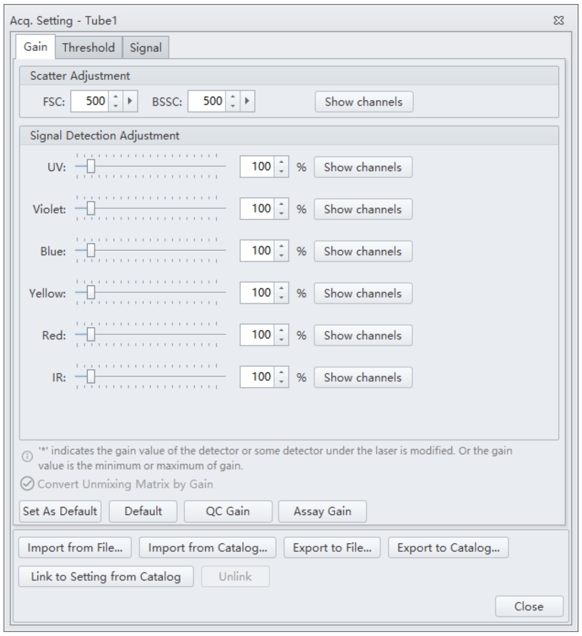
[Unmixed Worksheet Shown]



NOTE To modify the default settings, select **Options** in the Settings menu. The Options window appears. Select **Plot** on the left side of the Options window. Under the Signal section of the window, change the Main Channel default by selecting the **Height** or **Area**.

NOTE When using both Height and Area signals, ensure the gain setting is set to where the Height signal does not reach its upper range.

c. Signal width can be used as a tool for doublet discrimination and to differentiate somatic cell adhesion. If necessary, select Acq. Setting... to open the Acq. Setting window.



FSC: 500 BSSC: 500

UV: 100 %

Violet: 100 %

Blue: 100 %

Yellow: 100 %

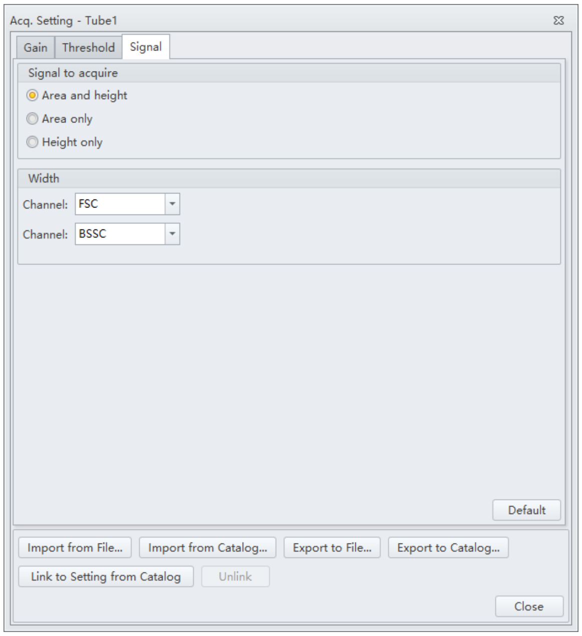
Red: 100 %

IR: 100 %

'*' indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

Convert Unmixing Matrix by Gain

d. Select the **Signal** tab, and select the channel with the required signal width.



Signal to acquire

● Area and height

○ Area only

○ Height only

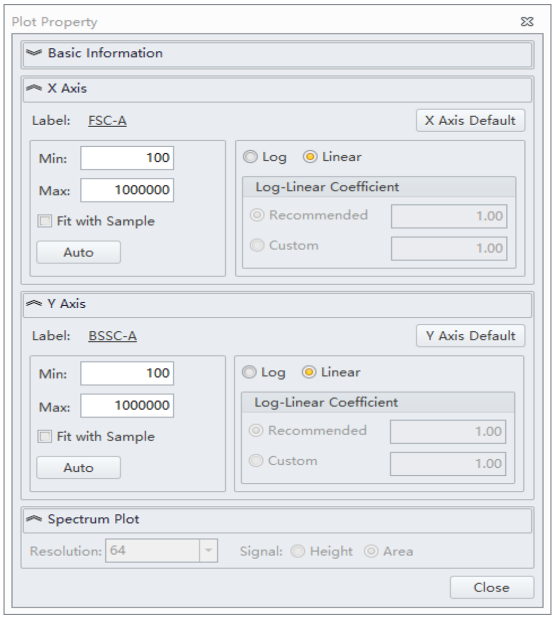
Width

Channel: FSC

Channel: BSSC

e. Plot properties can be configured to display axes in Log, Log-Linear, or Linear format.

1) Double-click the plot or right-click the plot and select **Property** from the drop-down menu. The Plot Property screen appears.



X Axis
Label: FSC-A
Min: 100
Max: 1000000
☐ Fit with Sample
○ Log ● Linear
Log-Linear Coefficient
○ Recommended 1.00
○ Custom 1.00

Y Axis
Label: BSSC-A
Min: 100
Max: 1000000
☐ Fit with Sample
○ Log ● Linear
Log-Linear Coefficient
○ Recommended 1.00
○ Custom 1.00

Spectrum Plot
Resolution: 64
Signal: ○ Height ○ Area

2) Select whether to display the axes in logarithmic or linear format for both the X-axis and Y-axis. Enter a value for log-linear coefficient if the log-linear view is desired.

3) Select **Close**.

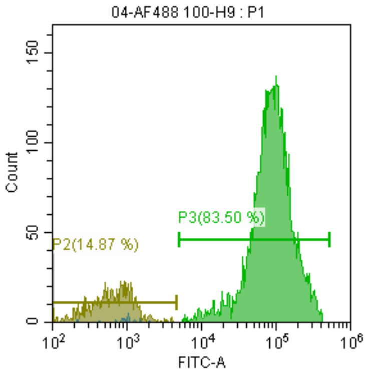
Or

Select the logarithmic axis on the plot. The slider appears. Drag the slider along the axis to change the log-linear coefficient and view events that are not shown, including events with negative values.

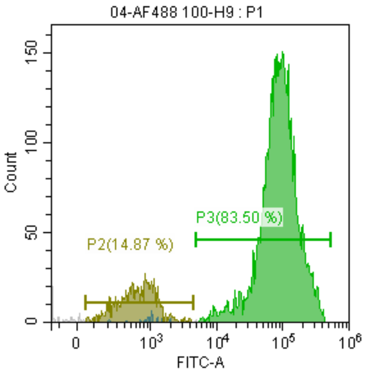
NOTE The log-linear slider is also available during data acquisition.

NOTE To reset the axis back to logarithmic, right-click on the axis and select **Property**. Select **X Axis Default** or **Y Axis Default** to reset the axis.

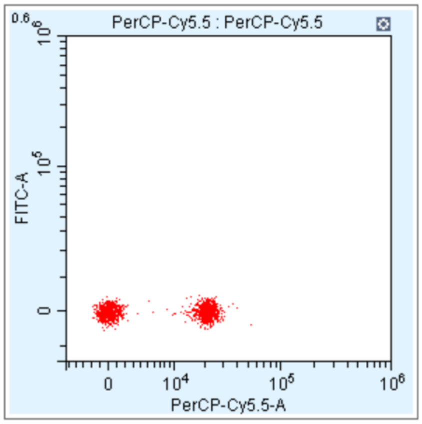
Histogram with logarithmic



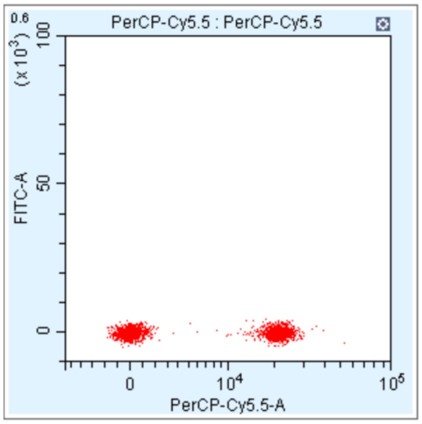
Histogram with log-linear



Dot plot with logarithmic

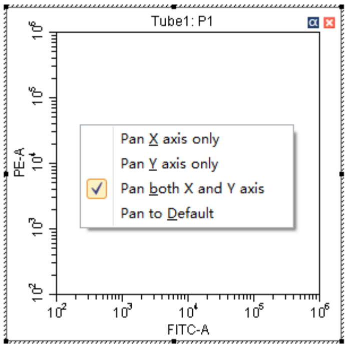


Dot plot with log-linear



f. You can adjust axis ranges using the pan axis display controls located at the top of the screen.

- Select 🔍 🔍 to zoom-in and define which area of a plot to enlarge. The selected area can be magnified to fill the entire graph. By selecting the zoom-out function, you can click on the graph and restore the plot to its original appearance before magnification.
- Select ✋ ✋ to shift the axes. The mouse pointer appears as a hand. It allows you to drag the graph to reveal the axis segment you need.
  - ✋ Pan: Modifies the axis display range dimensions when panning both axes. When the pan control is selected, you can right-click the graph and select which axis you need to adjust when dragging. You can also pan directly to the default axis range.



  - ✋ Single side pan: Modifies the axis display range dimensions when panning one axis.

NOTE Only the low end of the axis can be adjusted by the single side pan tool.

- Select to scale axis ranges in the plots automatically.

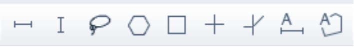
  NOTE This feature is only available in the spectral unmixing experiment and in the unmixed worksheet opened in a standard experiment (see Figure 2.2).

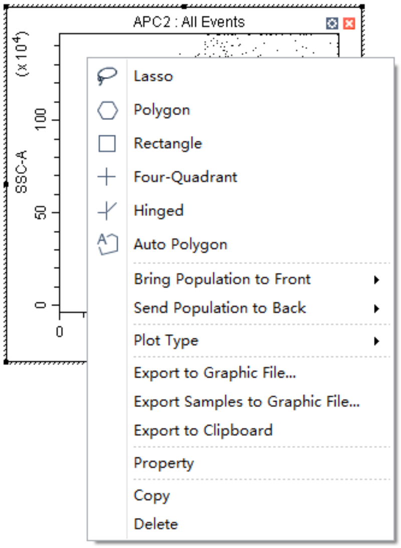
- Double-click the border area of the plot to open the Plot Property window, or right-click the plot, then select **Property** to open the same Plot Property window.
- In the Plot Property window, manually enter the minimum and maximum display values for the X- and Y-axes. You can also select **Fit With Sample** to let the software automatically adjust the lower limit according to the signal and perform the corresponding log-linear transformation. The X- and Y-axes **Default** settings are the default parameters. The default parameters are 100-1,000,000.

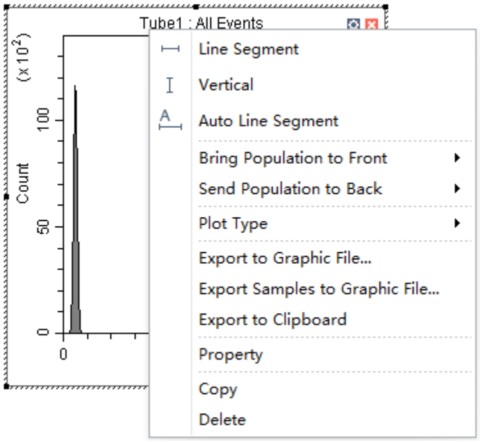
  NOTE Select **Fit With Sample** to identify the signal's lower limit, adjusting automatically as warranted. Selecting this item is recommended whenever the signal appears to be relatively low.

  NOTE Select **Auto** to automatically set the upper and lower display limits of the axes based on the data already collected.

  NOTE Select **Options** in the Settings menu, then select **Plot** to modify the default setting of the axis range under the Axis Default Setting section of the window.

2. To create gates, use the  control buttons or right-click the plot and select the gate type required. Gates can be set according to different requirements to differentiate cell populations.





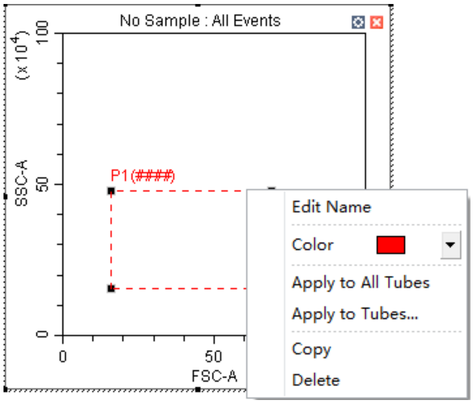
NOTE To add a vertex to a polygon gate:

1. Select the gate.
2. Hover your cursor over the perimeter of the gate until the cursor changes to the hand icon.
3. Select the desired location for the new gate vertex.

NOTE A newly created gate becomes a subset of the plot where it appears. The relationship between parent and progeny/daughter gates can be changed when a displayed gate is subsequently modified.

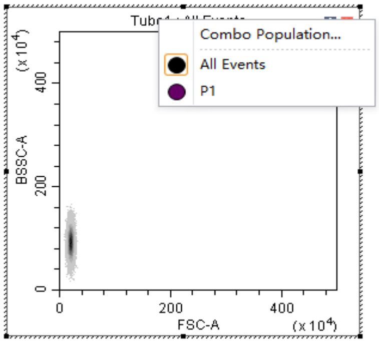
The position of the same gate in different sample tubes may vary. To change the position of a gate and apply the change to all sample tubes accordingly, you can right-click the gate and select **Apply to All Tubes**.

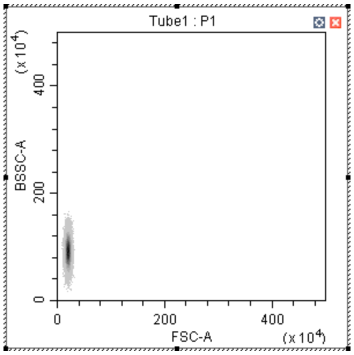
You can also apply the change to select tubes by selecting **Apply to Tubes.**



3 Select the gates to display.

a. Select the heading area of the plot, select the parent population/gate to display in the plot from the drop-down menu. The selected parent gate appears in the heading area of the plot.





NOTE The CytExpert for Spectral software will not list gates which would create circular gating logic.

Figure 6.2 shows all gates defined in the example experiment below. Note that the only gate option in plot 1 of Figure 6.3 is P2 for the following reasons:

- Plot 1 cannot be gated on P1 because P1 is on that plot.
- Plot 1 cannot be gated on P2 because P2 is gated on P1.
- Plot 1 cannot be gated on the P2 OR P1 combo population because the gate logic contains P1.

Figure 6.2 All Gates - Example Experiment

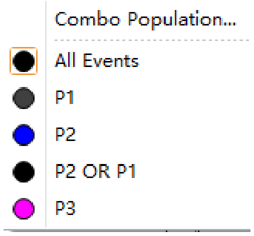
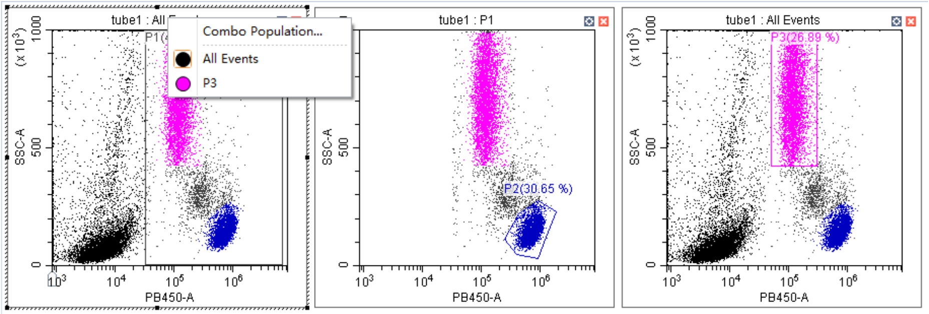
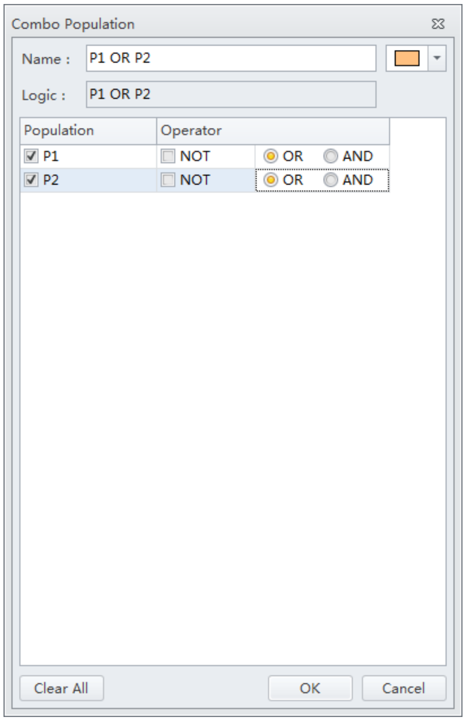


Figure 6.3 Circular Gating Logic - Example Experiment



b. If necessary, you can select the **Combo Population** option from the drop-down menu to create a combination gate, using the Boolean relationships “AND”, “OR”, and “NOT” to produce a new gate. You can also select the population color or change the gate name.



Name : P1 OR P2

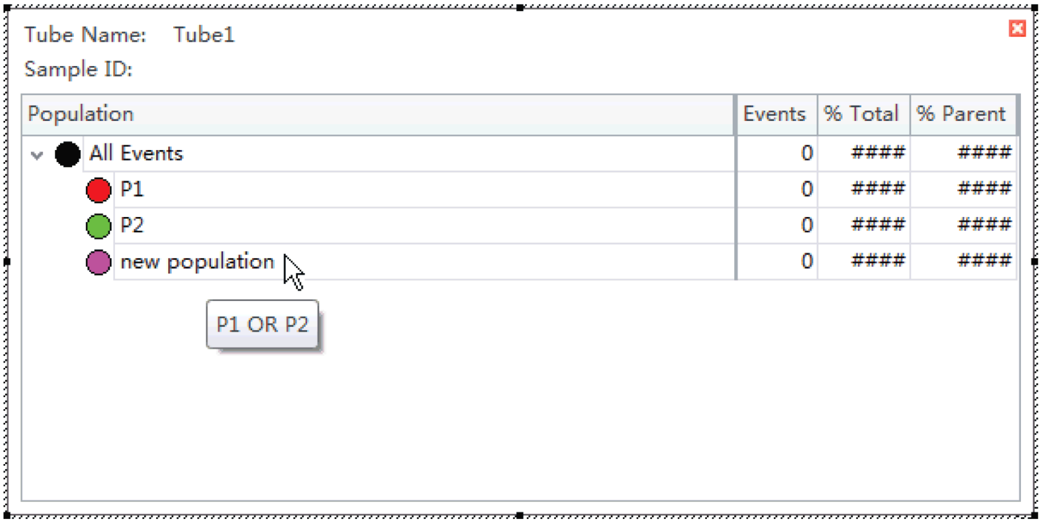
Logic : P1 OR P2

Population	Operator	
☑ P1	☐ NOT	● OR ○ AND
☑ P2	☐ NOT	● OR ○ AND

- “AND” indicates that all selections must be satisfied. For example, “P1 AND P2” means that the data for the newly added gate represent the intersection of P1 and P2.
- “OR” indicates that only one of the selections needs to be satisfied. For example, “P1 OR P2” means that the data for the newly added gate represent the union of P1 and P2.
- “NOT” indicates exclusion from the selection. For example, “NOT P1” means that the data for the newly added gate represent the events that are not part of P1.

4 Select to display the population hierarchy.

The Population Hierarchy function allows you to view how gates rank in relation to one another. To change the display color, double-click the default color and select the desired color from the drop-down color palette. To change the name of each gate, double-click the name of the desired gate. By hovering your mouse pointer over a combo population whose display name has just been changed, you can view its corresponding Boolean logical operation.



Tube Name: Tube1

Sample ID:

Population	Events	% Total	% Parent
● All Events	0	####	####
● P1	0	####	####
● P2	0	####	####
● new population	0	####	####

P1 OR P2

Creating and Adjusting Auto Gates

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Creating and Adjusting Auto Gates for detailed instructions on the following procedures:

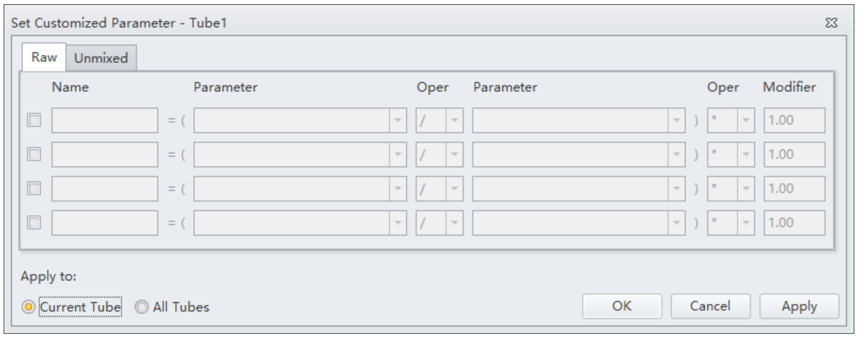
- Turning Auto Recalculate On/Off
- Adjusting Autogate Movement and Extent

Setting Customized Parameters

Set custom parameter to create fluorescence calculations.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting Customized Parameters for detailed instructions.

1. Select **Set Customized Parameter** from the Settings menu. Or, right-click a test tube from the test tube menu and select **Set Customized Parameter**. The Set Customized Parameter window appears.



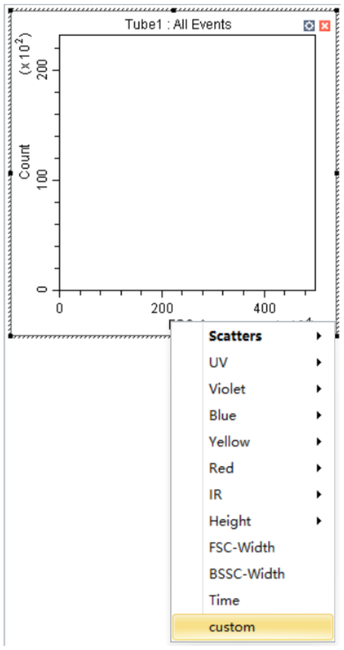
2. Select the **Raw**/**Unmixed** tab on the top of the Set Customized Parameter window as needed.

3. Enter a name for the parameter in the Name section.

4. Select the parameters for calculation from the Parameter dropdown menu.

5. Select the equation operations from the Open dropdown menu.

   The new parameter name is displayed in the list of parameters and statistic items.



Setting Custom Statistics

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting Custom Statistics for detailed instructions.

Configuring Acquisition Settings

NOTE Settings can be imported from the Acquisition Settings Catalog. Refer to CHAPTER 6, Importing Instrument Settings.

If unmixing settings are desired, import the unmixing matrix from the Spectral Library or import the unmixing file. Refer to CHAPTER 7, Importing and Exporting Unmixing Matrix. Unmixing matrix can be imported before or after performing the sample acquisition to generate unmixed data.

Changing the Tube Name

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Changing the Tube Name for detailed instructions.

Laser Settings

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Laser Settings for detailed instructions.

NOTE Lasers can only be enabled and disabled in Conventional Mode.

Setting Laser Target Power Settings [CytoFLEX LX Only]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting Laser Target Power Settings [CytoFLEX LX Only] for detailed instructions.

NOTE Lasers can only be enabled and disabled in Conventional Mode.

Adjusting the Gain

While the instrument is in use, the signal value can be increased or decreased by adjusting the instrument’s gain configuration.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Adjusting the Gain for detailed instructions.

---

1 Select Acq. Setting... on the left side of the screen. The Acq. Setting window appears.

---

2 Select the **Gain** tab in the Acq. Setting window.

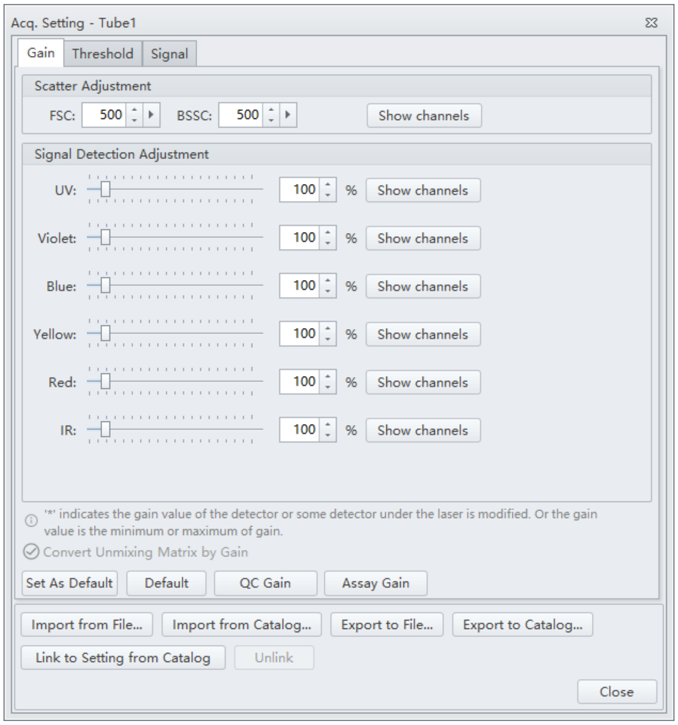
Select or edit the instrument's default gain settings using one of the following methods:

- Edit the gain settings and select **Set As Default** to create a new default settings.
- Select **Default** to return to your saved default settings.
- Select **QC Gain** to use the instrument's QC settings.
- Select **Assay Gain** to use the instrument's assay settings.

NOTE In cases where you do not specify your own default parameters, the Assay settings and default settings are identical.

---

3 Adjust the gain setting of each channel under the **Gain** tab in the Acq. Setting window. Raising the gain increases the signal. Lowering the gain reduces the signal.



FSC: 500
BSSC: 500

UV: 100 %
Violet: 100 %
Blue: 100 %
Yellow: 100 %
Red: 100 %
IR: 100 %

'*' indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

☑ Convert Unmixing Matrix by Gain

NOTE Gain adjustments have a predefined range between 1 and 3,000. For fine adjustments, use the text box and manually enter the gain values as needed.

NOTE The * symbol next to the channel indicates that:

- The gain of the channel has been modified.
- The gain of the channel is set to the minimum or maximum value.

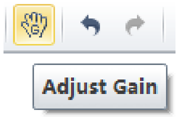
NOTE If you adjust the gain of a laser, the gain of all corresponding channels changes simultaneously.

If the gain of a channel has been modified, it cannot be modified by adjusting the corresponding laser's gain directly.

NOTE Optimize the gain settings according to your own experimental goals. The QC Gain values are only for reference.

NOTE Select **Link to Setting from Catalog** to apply the gain settings directly to skip adjusting the gain setting. Refer to Link to Acq. Setting.

Another option is to use the **Gain Control** button on the tool-bar in the graphic control area to adjust the gain values for cell population data to their desired levels, directly on the plots where the data appears during data collection.

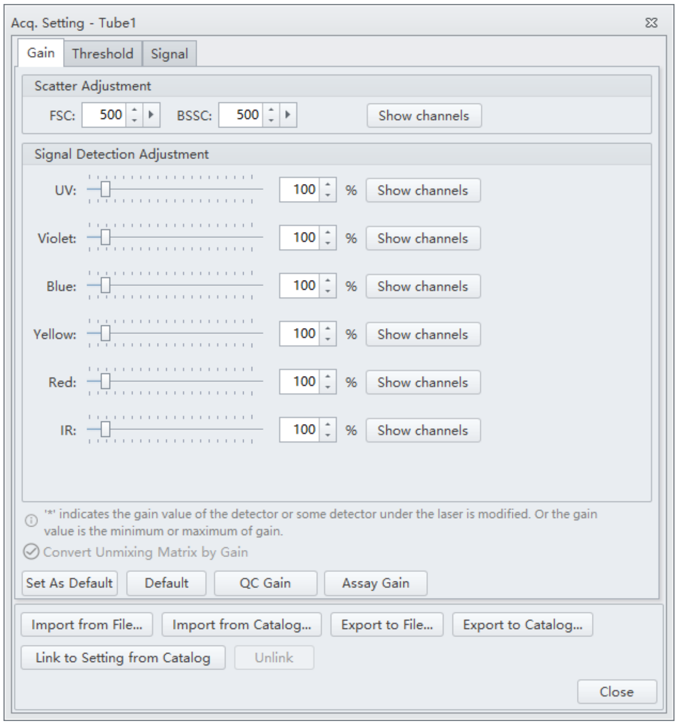


4 If necessary, change the coordinate display range and the plot type.

Link to Acq. Setting

This operation will only affect gain settings, not threshold and width settings.

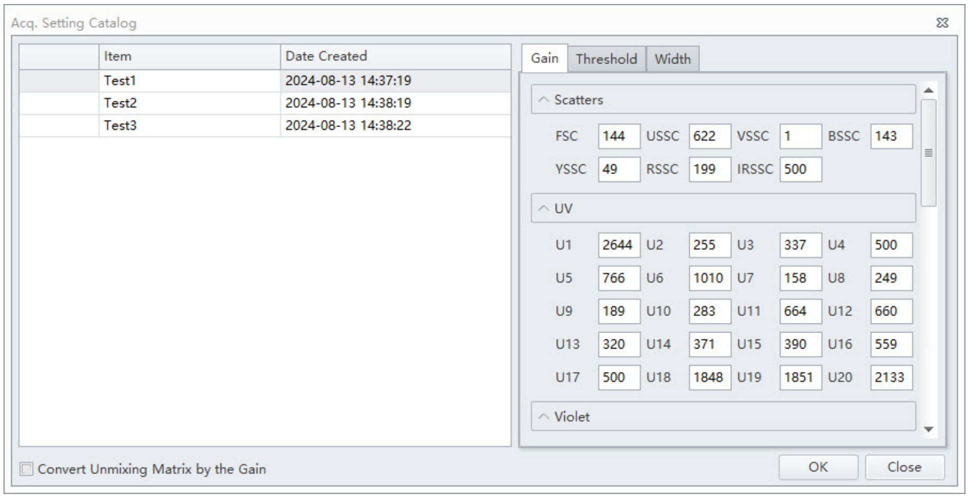
1 Select Acq. Setting... on the left side of the screen. The Acq. Setting window appears.



"*" indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

---

2 Select **Link to Setting from Catalog** in the Acq. Setting window. The Acq. Setting Catalog window appears.



Item	Date Created
Test1	2024-08-13 14:37:19
Test2	2024-08-13 14:38:19
Test3	2024-08-13 14:38:22

Scatters

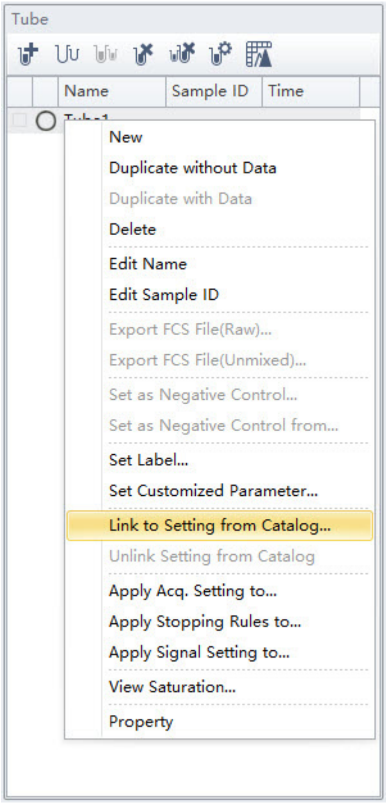
FSC	144	USSC	622	VSSC	1	BSSC	143
YSSC	49	RSSC	199	IRSSC	500		

UV

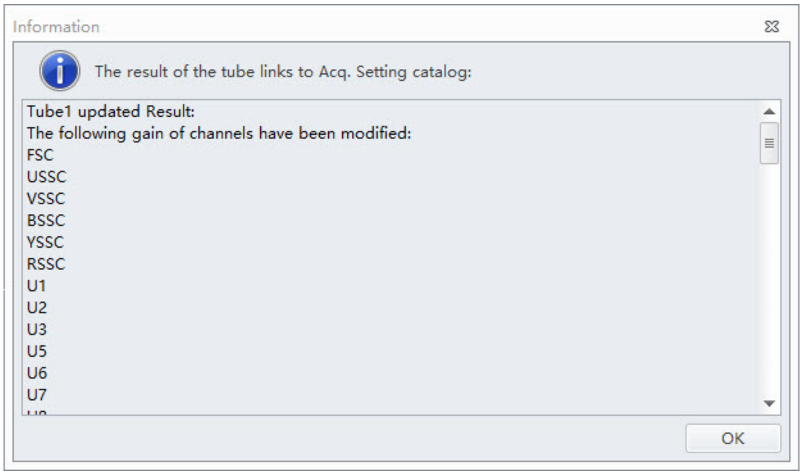
U1	2644	U2	255	U3	337	U4	500
U5	766	U6	1010	U7	158	U8	249
U9	189	U10	283	U11	664	U12	660
U13	320	U14	371	U15	390	U16	559
U17	500	U18	1848	U19	1851	U20	2133

Or

Right-click the desired tube and select **Link to Setting from Catalog** from the drop-down menu. The Acq. Setting Catalog window appears.


Tube
Name
Sample ID
Time
New
Duplicate without Data
Duplicate with Data
Delete
Edit Name
Edit Sample ID
Export FCS File(Raw)...
Export FCS File(Unmixed)...
Set as Negative Control...
Set as Negative Control from...
Set Label...
Set Customized Parameter...
Link to Setting from Catalog...
Unlink Setting from Catalog
Apply Acq. Setting to...
Apply Stopping Rules to...
Apply Signal Setting to...
View Saturation...
Property

3 Select the desired Item to link and select **OK**. The Information window appears.



The result of the tube links to Acq. Setting catalog:

Tube1 updated Result:
The following gain of channels have been modified:
FSC
USSC
VSSC
BSSC
YSSC
RSSC
U1
U2
U3
U5
U6
U7

NOTE Checking the **Convert Unmixing Matrix by the Gain** checkbox allows the unmixing matrix to automatically adjust by gain settings.

4 Select **OK**.

5 Select **Close** to exit the Acq. Setting window.

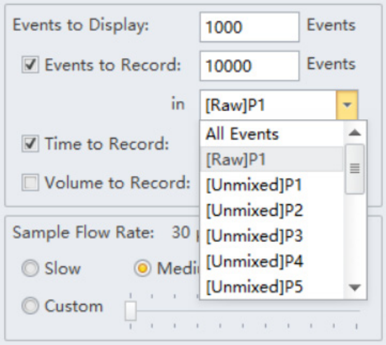
Adjusting the Threshold

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Adjusting the Threshold for detailed instructions.

Setting Collection Conditions

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting Collection Conditions for detailed instructions.

NOTE In the CytExpert for Spectral software, the **Events to Record** also includes a drop down menu where you can select to record either raw data or unmixed data.



Events to Display: 1000 Events
☑ Events to Record: 10000 Events
in [Raw]P1
☑ Time to Record:
☐ Volume to Record:
Sample Flow Rate: 30
○ Slow ● Medi
○ Custom

Setting Plot Display Conditions

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Setting Plot Display Conditions for detailed instructions.

Viewing Saturation [Without Plate Loader]

The saturation warning is an automatic iterative acquisition-and-check process. You can optimize the gain settings according to your own experiment goals to avoid signal saturation.

NOTE The Saturation Warning function is enabled by default. You cannot enable or disable the function during the data acquisition.

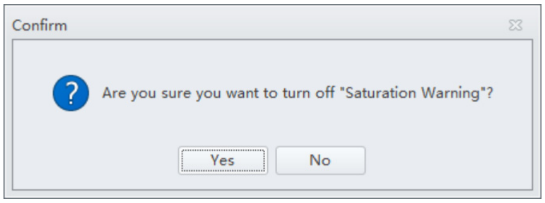
NOTE The Saturation Warning function only applies to the fluorescence channels.

NOTE Adjust the gain settings according to your own experimental goals as necessary to avoid signal saturation. Refer to Adjusting the Gain.

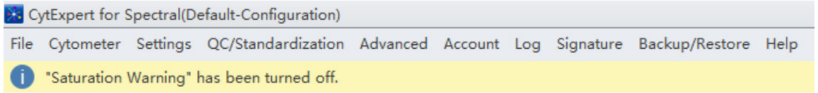
---

1 Confirm that the Saturation Warning function is enabled. If not, select **Saturation Warning** from the Advanced menu to enable it.

NOTE To disable the Saturation Warning function, select **Saturation Warning** from the Advanced menu The following system prompt appears, and select **Yes.** The saturation warning status displays on the top of the software screen.



Are you sure you want to turn off "Saturation Warning"?



"Saturation Warning" has been turned off.

---

2 Create a new experiment and new tube. Refer to Creating an Experiment.

---

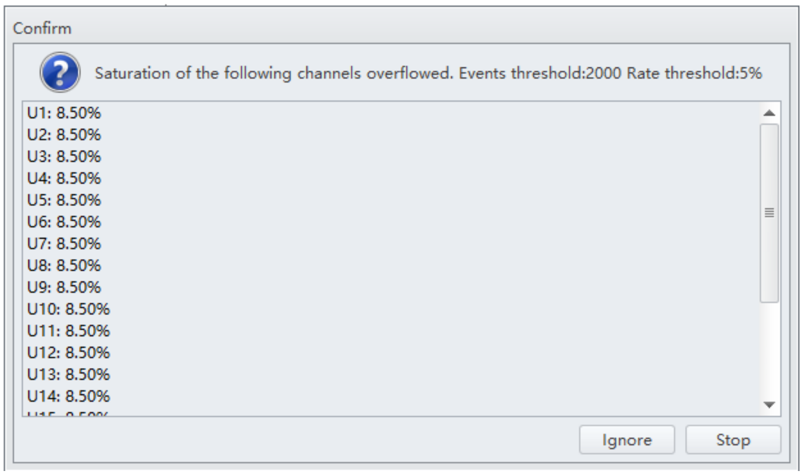
3 Configure the acquisition settings. Refer to Configuring Acquisition Settings.

---

4 Load samples and record data as needed. Refer to Load Sample and Record Data.

---

5 When the acquired or recorded events reach 2,000, the confirmation window appears.



Saturation of the following channels overflowed. Events threshold:2000 Rate threshold:5%

U1: 8.50%
U2: 8.50%
U3: 8.50%
U4: 8.50%
U5: 8.50%
U6: 8.50%
U7: 8.50%
U8: 8.50%
U9: 8.50%
U10: 8.50%
U11: 8.50%
U12: 8.50%
U13: 8.50%
U14: 8.50%

NOTE The default saturation threshold is 5%.

NOTE The acquisition continues when the confirmation window appears.

---

6 Select **Ignore** to close the confirmation window.

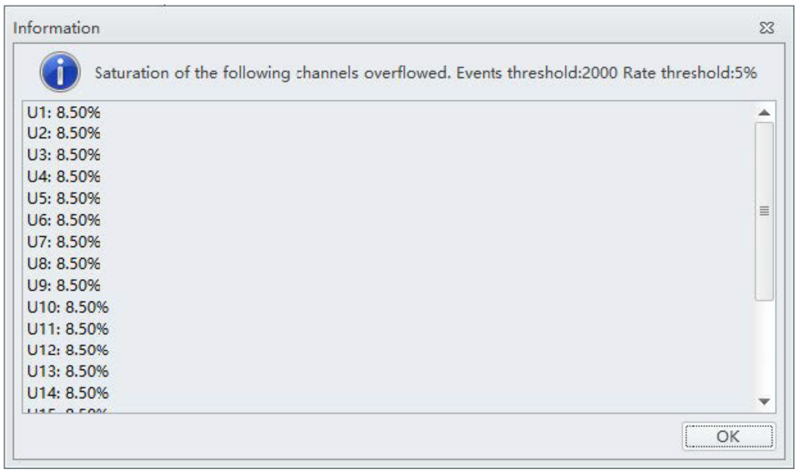
Or

Select **Stop** to stop the acquisition.

NOTE If you select **Ignore**, the confirmation window will no longer prompt.

---

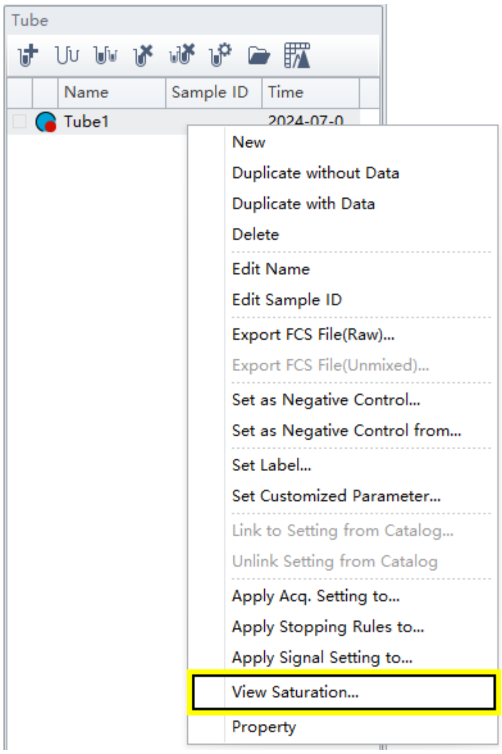
7 When the acquisition completes, the Information window appears. Select **OK**.

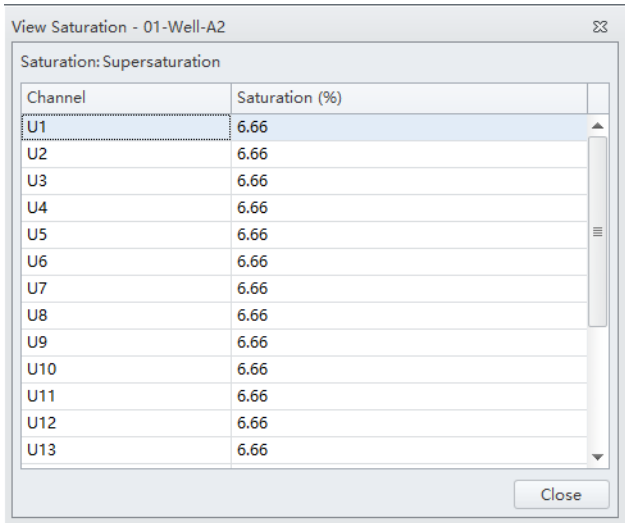


Saturation of the following channels overflowed. Events threshold:2000 Rate threshold:5%

U1: 8.50%
U2: 8.50%
U3: 8.50%
U4: 8.50%
U5: 8.50%
U6: 8.50%
U7: 8.50%
U8: 8.50%
U9: 8.50%
U10: 8.50%
U11: 8.50%
U12: 8.50%
U13: 8.50%
U14: 8.50%

8 Right-click a tube and select **View Saturation.** The View Saturation window appears.


Tube
Name
Sample ID
Time
Tube1
2024-07-0
New
Duplicate without Data
Duplicate with Data
Delete
Edit Name
Edit Sample ID
Export FCS File(Raw)...
Export FCS File(Unmixed)...
Set as Negative Control...
Set as Negative Control from...
Set Label...
Set Customized Parameter...
Link to Setting from Catalog...
Unlink Setting from Catalog
Apply Acq. Setting to...
Apply Stopping Rules to...
Apply Signal Setting to...
View Saturation...
Property



Saturation: Supersaturation

Channel	Saturation (%)
U1	6.66
U2	6.66
U3	6.66
U4	6.66
U5	6.66
U6	6.66
U7	6.66
U8	6.66
U9	6.66
U10	6.66
U11	6.66
U12	6.66
U13	6.66

9 Select **Close.**

Viewing Saturation [With Plate Loader]

The saturation warning is an automatic iterative acquisition-and-check process. You can optimize the gain settings according to your own experiment goals to avoid signal saturation.

NOTE The Saturation Warning function is enabled by default. You cannot enable or disable the function during the data acquisition.

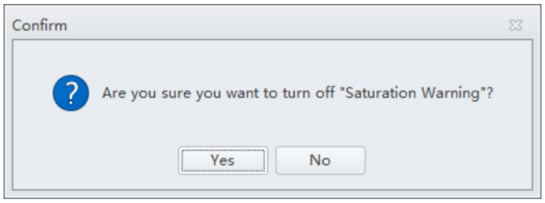
NOTE The Saturation Warning function only applies to the fluorescence channels.

NOTE Adjust the gain settings according to your own experimental goals as necessary to avoid signal saturation. Refer to Adjusting the Gain.

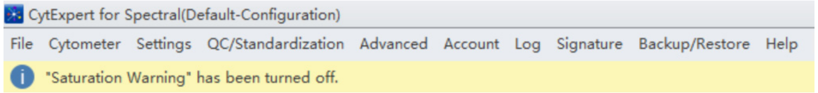
---

1 Confirm that the Saturation Warning function is enabled. If not, select **Saturation Warning** from the Advanced menu to enable it.

NOTE To disable the Saturation Warning function, select **Saturation Warning** from the Advanced menu The following system prompt appears, and select **Yes.** The saturation warning status displays on the top of the software screen.



Are you sure you want to turn off "Saturation Warning"?



"Saturation Warning" has been turned off.

---

2 Create a new experiment and new tube. Refer to Creating an Experiment.

---

3 Configure the acquisition settings. Refer to Configuring Acquisition Settings.

---

4 Load samples and record data as needed. Refer to Load Sample and Record Data.

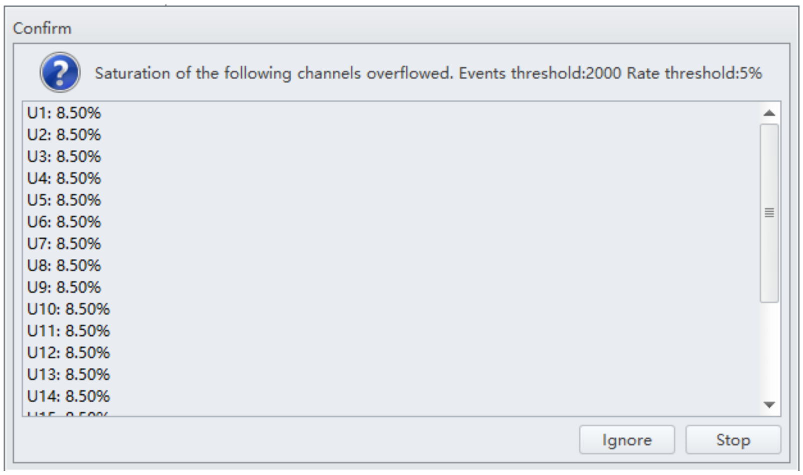
---

5 If you are running a single well, proceed to Step 6.

If you are running a set of wells, skip to Step 9.

---

6 When the acquired or recorded events reach 2,000, the confirmation window appears.



Saturation of the following channels overflowed. Events threshold:2000 Rate threshold:5%

U1: 8.50%
U2: 8.50%
U3: 8.50%
U4: 8.50%
U5: 8.50%
U6: 8.50%
U7: 8.50%
U8: 8.50%
U9: 8.50%
U10: 8.50%
U11: 8.50%
U12: 8.50%
U13: 8.50%
U14: 8.50%

NOTE The default saturation threshold is 5%.

NOTE The acquisition continues when the confirmation window appears.

---

7 Select **Ignore** to close the confirmation window.

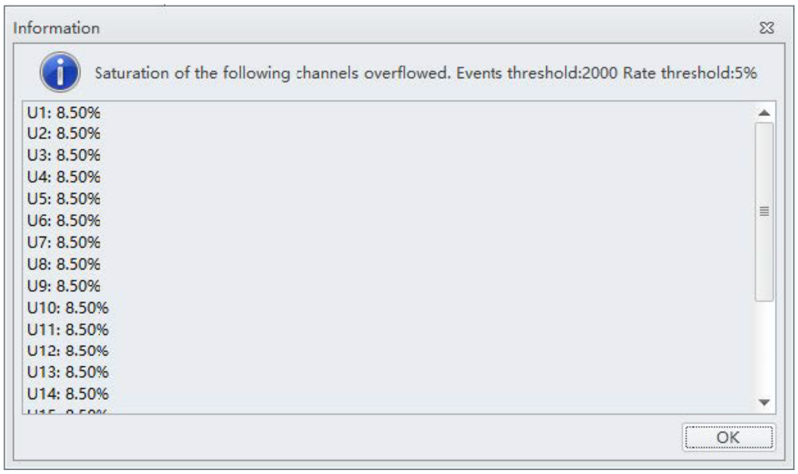
Or

Select **Stop** to stop the acquisition.

NOTE If you select **Ignore**, the confirmation window will no longer prompt until the software completes the current sample tube acquisition.

---

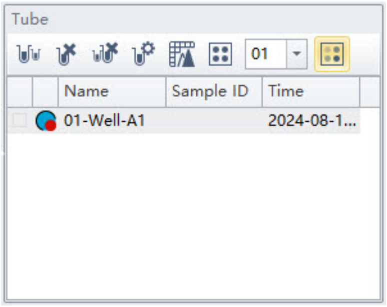
8 When the acquisition completes, the saturation information window appears. Select **OK**.



Saturation of the following channels overflowed. Events threshold:2000 Rate threshold:5%

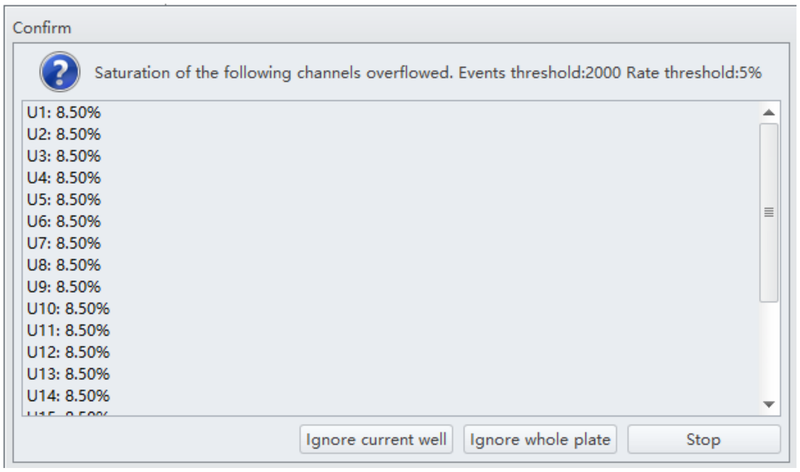
U1: 8.50%
U2: 8.50%
U3: 8.50%
U4: 8.50%
U5: 8.50%
U6: 8.50%
U7: 8.50%
U8: 8.50%
U9: 8.50%
U10: 8.50%
U11: 8.50%
U12: 8.50%
U13: 8.50%
U14: 8.50%

The saturated icon appears in the Tube List next to the tube if the tube data acquired is saturated. Skip to Step 11.



---

9 When the auto recorded events reach 2,000, the confirmation window prompts.



Saturation of the following channels overflowed. Events threshold:2000 Rate threshold:5%

U1: 8.50%
U2: 8.50%
U3: 8.50%
U4: 8.50%
U5: 8.50%
U6: 8.50%
U7: 8.50%
U8: 8.50%
U9: 8.50%
U10: 8.50%
U11: 8.50%
U12: 8.50%
U13: 8.50%
U14: 8.50%

NOTE The default saturation threshold is 5%.

NOTE The acquisition continues when the confirmation window prompts.

10 Select **Ignore current well** or **Ignore whole plate** to close the confirmation window.

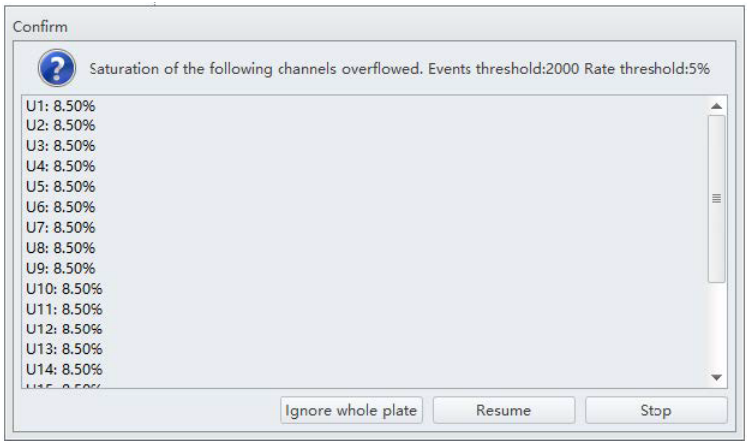
Or

Select **Stop** to stop the acquisition.

NOTE If you select **Ignore whole plate**, the confirmation window will no longer prompt.

NOTE If you select **Ignore current well**, the following confirmation window appears when the current sample tube acquisition completes. The acquisition stops.

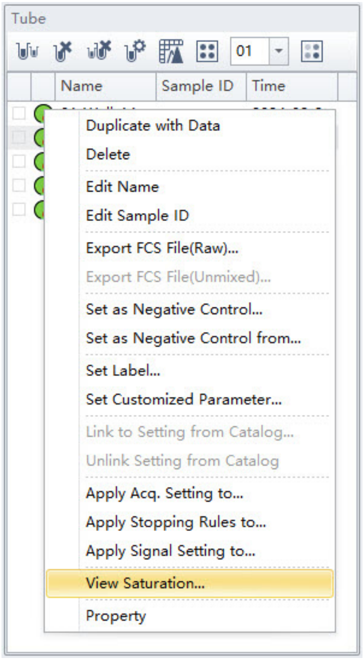
- Select **Ignore whole plate** to continue the acquisition. The confirmation window will no longer prompt.
- Select **Resume** to continue the acquisition. The confirmation window will prompt when the next tube is saturated.
- Select **Stop** to stop the acquisition.

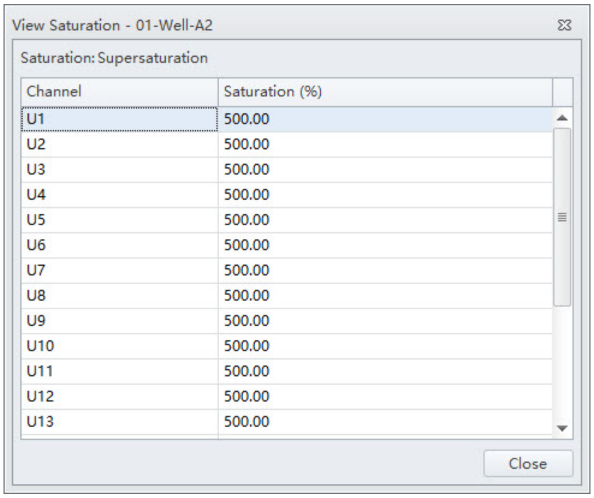


Saturation of the following channels overflowed. Events threshold:2000 Rate threshold:5%

U1: 8.50%
U2: 8.50%
U3: 8.50%
U4: 8.50%
U5: 8.50%
U6: 8.50%
U7: 8.50%
U8: 8.50%
U9: 8.50%
U10: 8.50%
U11: 8.50%
U12: 8.50%
U13: 8.50%
U14: 8.50%

11 Right-click a tube and select **View Saturation.** The View Saturation window appears.


Tube
01
Name
Sample ID
Time
Duplicate with Data
Delete
Edit Name
Edit Sample ID
Export FCS File(Raw)...
Export FCS File(Unmixed)...
Set as Negative Control...
Set as Negative Control from...
Set Label...
Set Customized Parameter...
Link to Setting from Catalog...
Unlink Setting from Catalog
Apply Acq. Setting to...
Apply Stopping Rules to...
Apply Signal Setting to...
View Saturation...
Property



View Saturation - 01-Well-A2

Saturation: Supersaturation

Channel	Saturation (%)
U1	500.00
U2	500.00
U3	500.00
U4	500.00
U5	500.00
U6	500.00
U7	500.00
U8	500.00
U9	500.00
U10	500.00
U11	500.00
U12	500.00
U13	500.00

12 Select **Close.**

Load Sample and Record Data

Before Running Samples

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Before Running Samples for detailed instructions.

NOTE The Detector Configuration function is only available in Conventional Mode.

Sampling and Collecting Data [Without Plate Loader]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Sampling and Collecting Data [Without Plate Loader] for detailed instructions.

NOTE The Compensation function is only available in Conventional Mode.

Sampling and Collecting Data [With Plate Loader]

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Sampling and Collecting Data [With Plate Loader] for detailed instructions on the following procedures:

- Creating a Heat Map [with Plate Loader]
- Refreshing a Heat Map
- Modifying Existing Heat Map Settings
- Deleting an Existing Heat Map
- Exporting a Heat Map

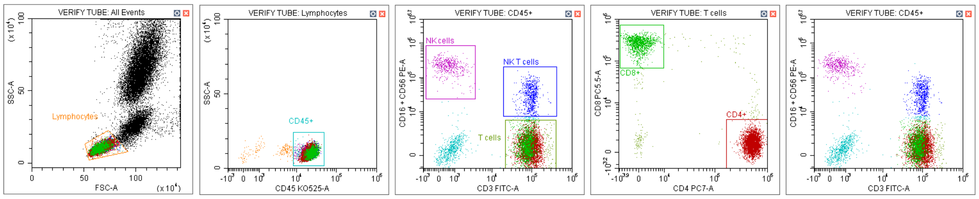
NOTE In the CytExpert for Spectral software, the Heat Map function is only available when the tube contains unmixed data.

Analyzing and Exporting Data

1 Select **Raw/Unmixed** located on the bottom of the plot area to open the raw/unmixed worksheet (see Figure 2.1 and Figure 2.2).

2 Select the sample tube to be analyzed.

3 Establish new gates or adjust the position of existing gates. Refer to Creating Plots and Gates.



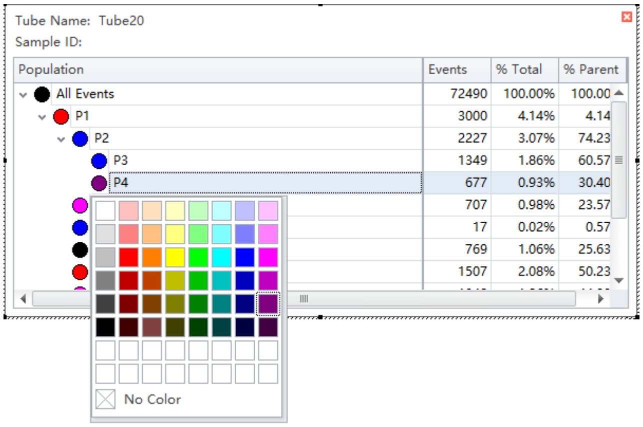
NOTE Changing a gate's position does not affect the positions of other gates already established on a given sample tube. Each test tube individually records the positions of its associated gates. If you need to make a change that concerns all the tubes, you must select the gate, then right-click the correctly positioned gate and select **Apply to All Tubes**.

4 Select . The Gate Hierarchy screen appears.

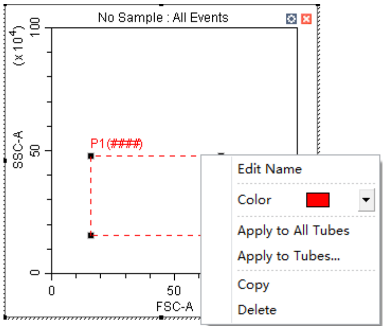
5 Check the relationship between the parent and daughter gates in the Gate Hierarchy window.

NOTE Newly added gates become subsets of populations displayed in plots with existing gates. Name and display color can be modified. Right-click directly on a gate plot to change the name and color.

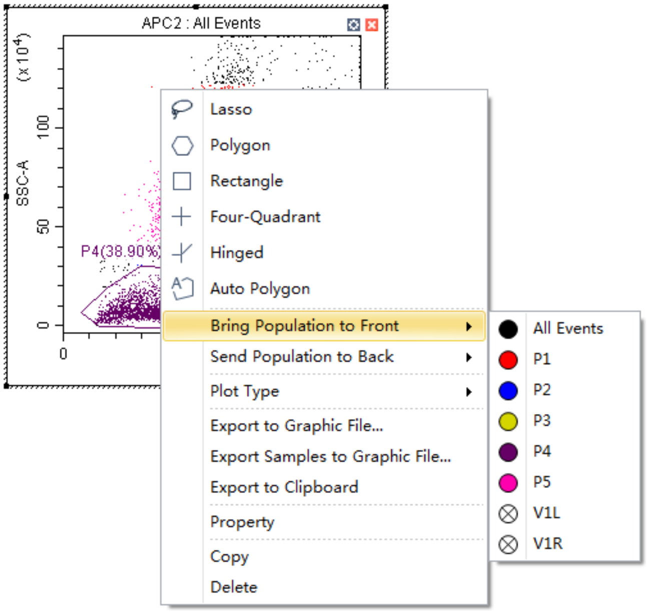
NOTE Select **No Color** to leave the gated events uncolored while retaining the color of the parent populations. By default, the populations defined by a vertical gate, hinged gate, or four-quadrant gate are uncolored.


Tube Name: Tube20
Sample ID:

Population	Events	% Total	% Parent
All Events	72490	100.00%	100.00
P1	3000	4.14%	4.14
P2	2227	3.07%	74.23
P3	1349	1.86%	60.57
P4	677	0.93%	30.40
	707	0.98%	23.57
	17	0.02%	0.57
	769	1.06%	25.63
	1507	2.08%	50.23

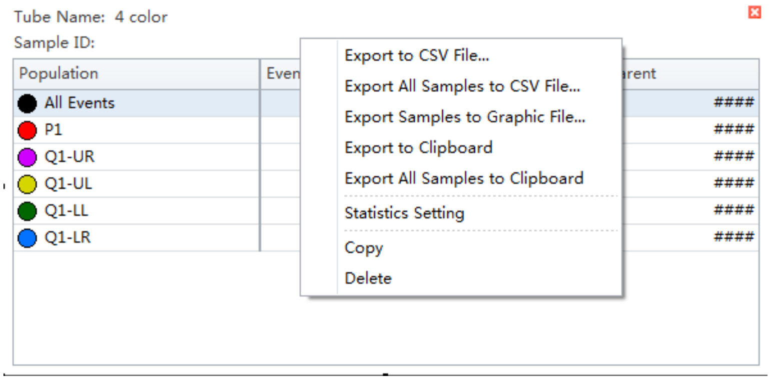


6 Right-click the plot and select **Bring population to front** to make the display color of the specified gate appear in front of all other colors, or select **Send population to back** to hide the display color of the specified gate behind all other colors.

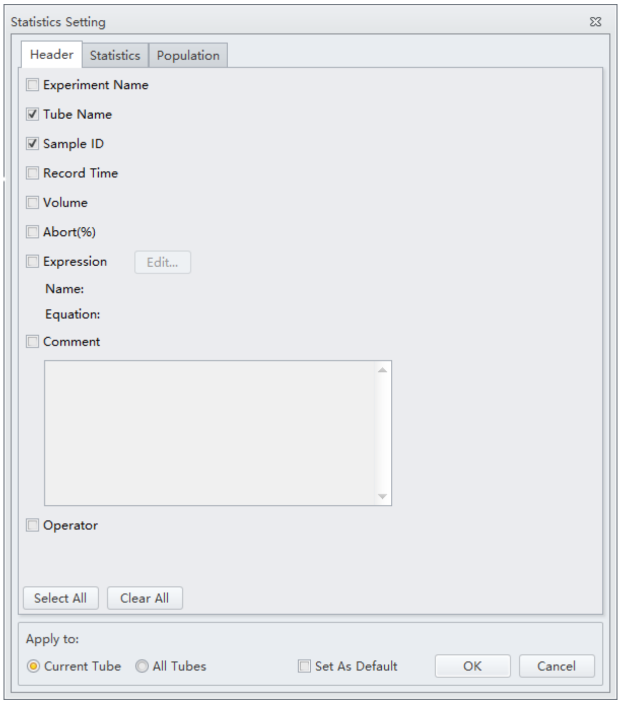


7 Select 📊 in the plot area to generate a statistical table.

8 Right-click the table and select **Statistics Setting** to modify the settings of the statistics display parameters. The Statistics Setting window appears.



The Statistics Setting window allows you to change the display of the header, statistical elements and cell populations included.

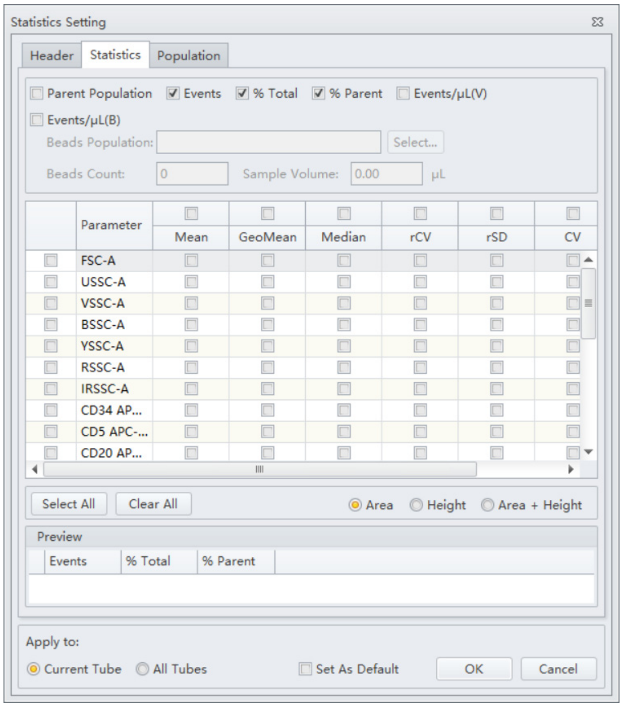


Statistics Setting

Header | Statistics | Population

☐ Experiment Name
☑ Tube Name
☑ Sample ID
☐ Record Time
☐ Volume
☐ Abort(%)
☐ Expression
Name:
Equation:
☐ Comment
☐ Operator

Apply to: ● Current Tube ○ All Tubes ☐ Set As Default



Statistics Setting

Header | Statistics | Population

☐ Parent Population ☑ Events ☑ % Total ☑ % Parent ☐ Events/μL(V)
☐ Events/μL(B)
Beads Population:
Beads Count: 0
Sample Volume: 0.00 μL

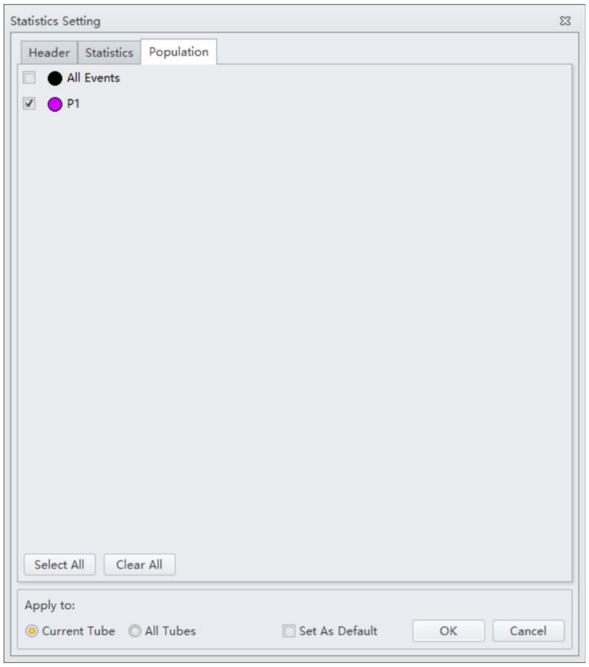
	Parameter	☐ Mean	☐ GeoMean	☐ Median	☐ rCV	☐ rSD	☐ CV
☐	FSC-A	☐	☐	☐	☐	☐	☐
☐	USSC-A	☐	☐	☐	☐	☐	☐
☐	VSSC-A	☐	☐	☐	☐	☐	☐
☐	BSSC-A	☐	☐	☐	☐	☐	☐
☐	YSSC-A	☐	☐	☐	☐	☐	☐
☐	RSSC-A	☐	☐	☐	☐	☐	☐
☐	IRSSC-A	☐	☐	☐	☐	☐	☐
☐	CD34 AP...	☐	☐	☐	☐	☐	☐
☐	CD5 APC-...	☐	☐	☐	☐	☐	☐
☐	CD20 AP...	☐	☐	☐	☐	☐	☐

● Area ○ Height ○ Area + Height

Preview

Events	% Total	% Parent

Apply to: ● Current Tube ○ All Tubes ☐ Set As Default



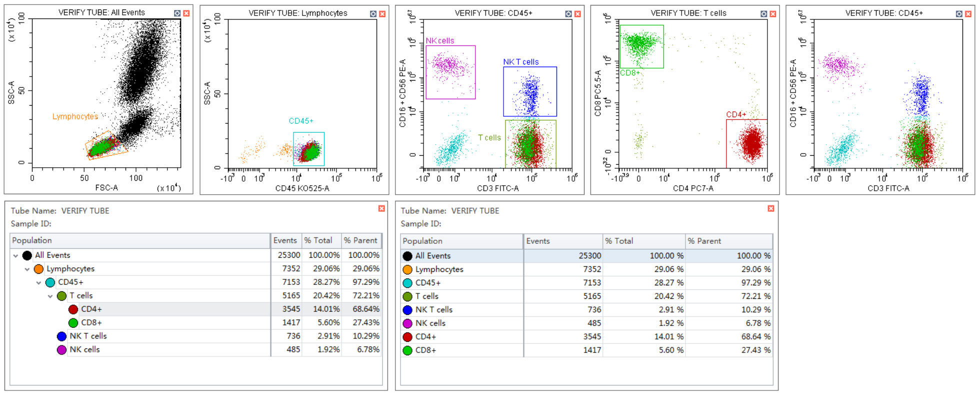
Statistics Setting

Header | Statistics | Population

☐ ● All Events
☑ ● P1

Apply to: ● Current Tube ○ All Tubes ☐ Set As Default

The final generated plots appear as below.



Tube Name: VERIFY TUBE
Sample ID:

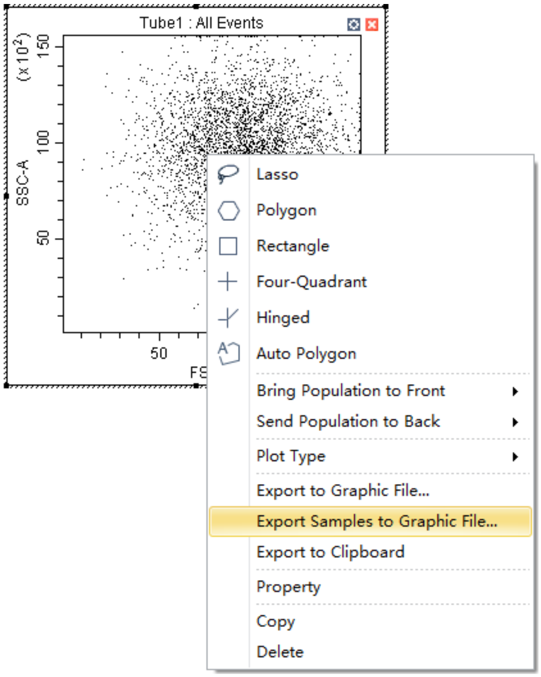
Population	Events	% Total	% Parent
All Events	25300	100.00%	100.00%
Lymphocytes	7352	29.06%	29.06%
CD45+	7153	28.27%	97.29%
T cells	5165	20.42%	72.21%
CD4+	3545	14.01%	68.64%
CD8+	1417	5.60%	27.43%
NK T cells	736	2.91%	10.29%
NK cells	485	1.92%	6.78%

Tube Name: VERIFY TUBE
Sample ID:

Population	Events	% Total	% Parent
All Events	25300	100.00 %	100.00 %
Lymphocytes	7352	29.06 %	29.06 %
CD45+	7153	28.27 %	97.29 %
T cells	5165	20.42 %	72.21 %
NK T cells	736	2.91 %	10.29 %
NK cells	485	1.92 %	6.78 %
CD4+	3545	14.01 %	68.64 %
CD8+	1417	5.60 %	27.43 %

---

9 Right-click a plot and select **Export to Clipboard** or **Export to Graphic File** from the drop-down menu to select an image to export.



- **Export to Clipboard** copies the plot to the clipboard, allowing you to paste it directly into documents in common file formats.

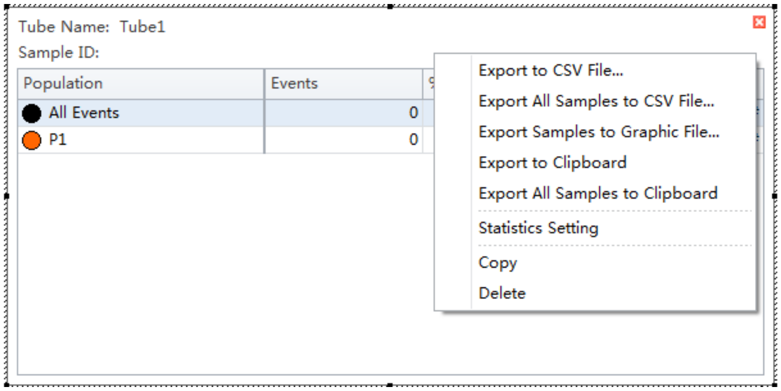
  NOTE Multiple plots can only be copied and pasted into Microsoft Word. If a single plot is copied, this can be pasted into both Microsoft Word or Microsoft PowerPoint.

- **Export to Graphic File** saves the plot as an image file.

  NOTE **Export to Graphic File** can export plots in two selectable file formats. BMP bitmap format and EMF vector format.

---

10 To export statistics, right-click a statistical table to select any one of the available export options.



Tube Name: Tube1
Sample ID:

Population	Events
● All Events	0
● P1	0

Export to CSV File...
Export All Samples to CSV File...
Export Samples to Graphic File...
Export to Clipboard
Export All Samples to Clipboard
Statistics Setting
Copy
Delete

- **Export to CSV File** exports individual tube statistics as a single CSV file.
- **Export All Samples to CSV File** exports all tube statistics as a single CSV file.
- **Export to Clipboard** copies the statistics of an individual sample to the clipboard, allowing you to paste them directly into a Microsoft Excel file or other file formats.
- **Export All Samples to Clipboard** assembles the statistics for all the sample tubes of an experiment and copies them together to the clipboard. From there they can be pasted as a group into a Microsoft Excel file or other file formats.
- **Copy** converts a statistical table into an image format that can be pasted into documents.

---

11 Export the FCS file if necessary. Refer to Exporting FCS Files.

NOTE Ensure that any storage devices used with the instrument are free from viruses. To guard against data loss, Beckman Coulter recommends backing up data on a frequent and regular basis. Beckman Coulter is not liable for any loss of data resulting from computer viruses or damage to hardware.

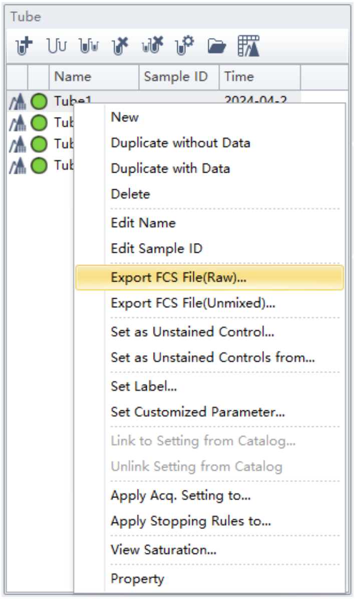
Exporting FCS Files

Exporting Single Tube Files

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Exporting Single Tube Files for detailed instructions.

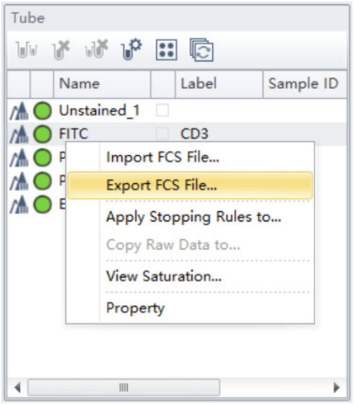
---

1 **[Standard Experiment]:** Right-click the desired tube from the test tube section of the screen and select **Export FCS File (Raw)** or **Export FCS File (Unmixed)**. The Export FCS File window appears.


Tube
Name
Sample ID
Time
Tube1
2024-04-2
New
Duplicate without Data
Duplicate with Data
Delete
Edit Name
Edit Sample ID
Export FCS File(Raw)...
Export FCS File(Unmixed)...
Set as Unstained Control...
Set as Unstained Controls from...
Set Label...
Set Customized Parameter...
Link to Setting from Catalog...
Unlink Setting from Catalog
Apply Acq. Setting to...
Apply Stopping Rules to...
View Saturation...
Property

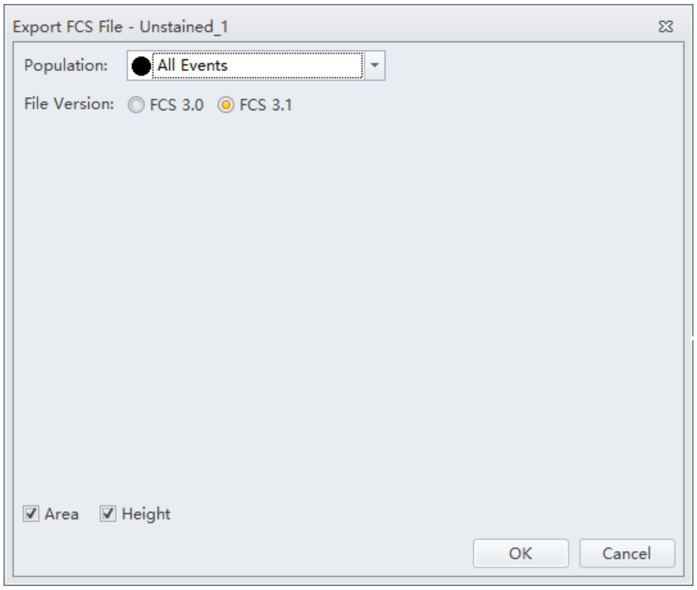
---

2 **[Spectral Unmixing Experiment]:** Right-click the desired tube and select **Export FCS File** from the drop-down menu. The Export FCS File window appears.



---

3 Select the population from the Population drop-down menu.



Export FCS File - Unstained_1

Population: ● All Events

File Version: ○ FCS 3.0 ● FCS 3.1

☑ Area ☑ Height

---

4 Select **Area** and/or **Height**.

---

5 Select the FCS format next to File Version.

NOTE The default setting is FCS 3.1.

---

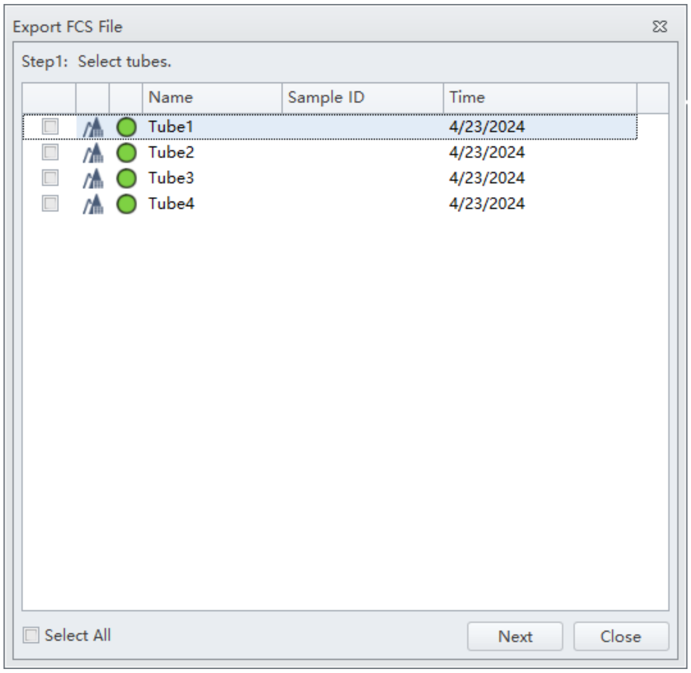
6 Select **OK**. Specify the file path and file name, and select **Save**.

Exporting Multiple FCS Files

NOTE This function is not available in spectral unmixing experiments.

---

1 Select **File** > **Export FCS File (Raw)** or **Export FCS File (Unmixed)**. The Export FCS File window appears.



☐			Name	Sample ID	Time
☐		●	Tube1		4/23/2024
☐		●	Tube2		4/23/2024
☐		●	Tube3		4/23/2024
☐		●	Tube4		4/23/2024

---

2 Select the desired tubes to export.

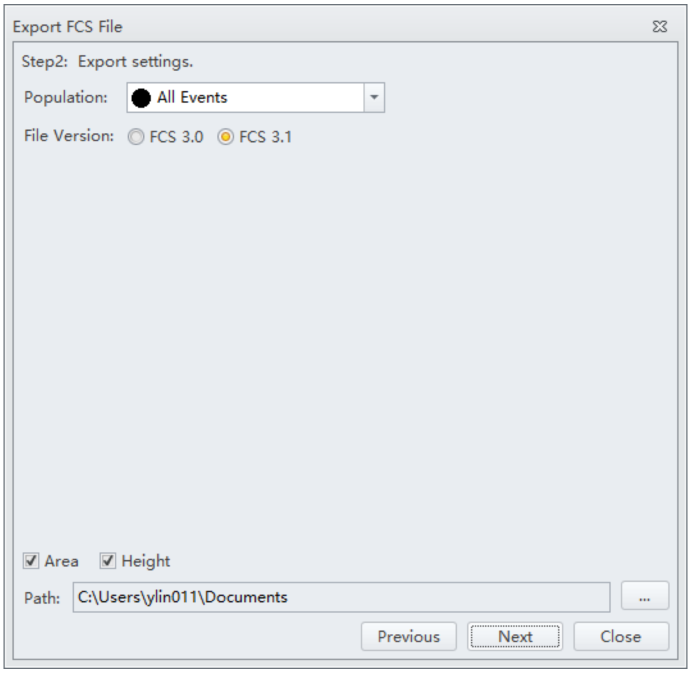
NOTE The **Select All** checkbox allows you to select all of the tubes listed.

---

3 Select **Next**.

---

4 Select the population from the Population drop-down menu.



Export FCS File

Step2: Export settings.

Population: ● All Events

File Version: ○ FCS 3.0 ● FCS 3.1

☑ Area ☑ Height

Path: C:\Users\ylin011\Documents

---

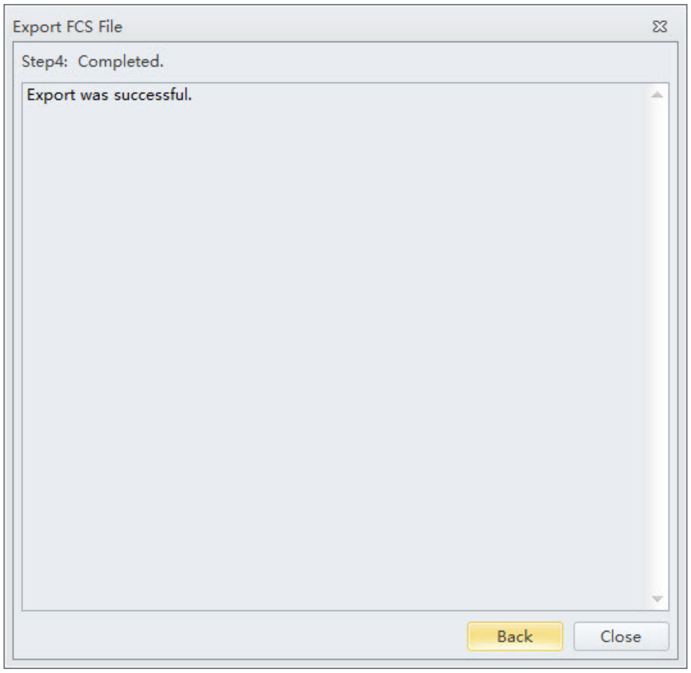
5 Select **Area** and/or **Height**.

---

6 Select the FCS format next to File Version.

NOTE The default setting is FCS 3.1.

7 Select ... to specify the file path, and select **Next**. The following massage appears: *Export was successful.*



Step4: Completed.

Export was successful.

8 Select **Close** to exit the Export FCS File window.

Or

Select **Back** and repeat Steps 1-7 to continue exporting FCS files.

Exporting Plots or the Statistics Table of Multiple Tubes as Picture Files

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Exporting Plots or the Statistics Table of Multiple Tubes as Picture Files for detailed instructions.

Importing and Exporting Instrument Settings

The CytExpert for Spectral software supports importing and exporting instrument settings to facilitate the experiment process. Only instrument settings identical to the current configuration can be imported with current detector settings.

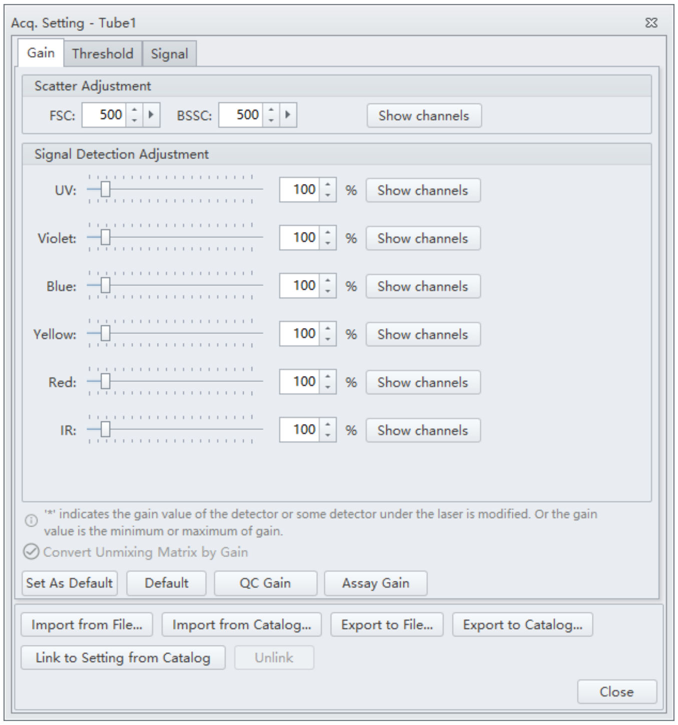
Select Acq. Setting... to edit gain, threshold, and width. These can be imported from an experiment file or from a catalog of instrument settings.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Importing and Exporting Instrument Settings for detailed instructions.

Importing Instrument Settings

---

1 Select the desired sample tube to import and select Acq. Setting.... The Acq. Setting window appears.



FSC: 500
BSSC: 500

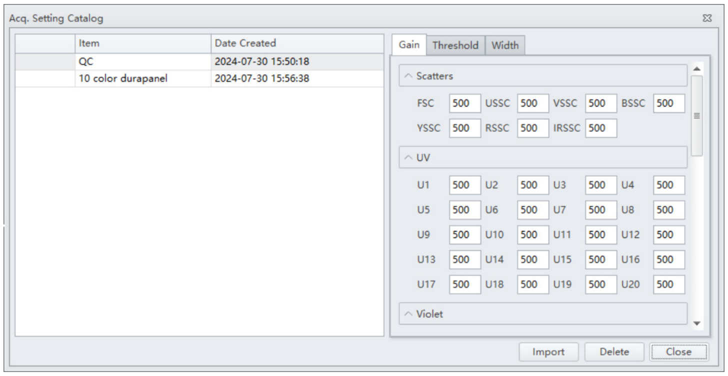
UV: 100 %
Violet: 100 %
Blue: 100 %
Yellow: 100 %
Red: 100 %
IR: 100 %

'*' indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

NOTE Instrument settings can only be imported into tubes where data has not yet been recorded.

2 Select **Import From File**, locate the file with the required instrument settings, or select **Import From Catalog** to import the instrument settings.

[CytoFLEX LX Shown]



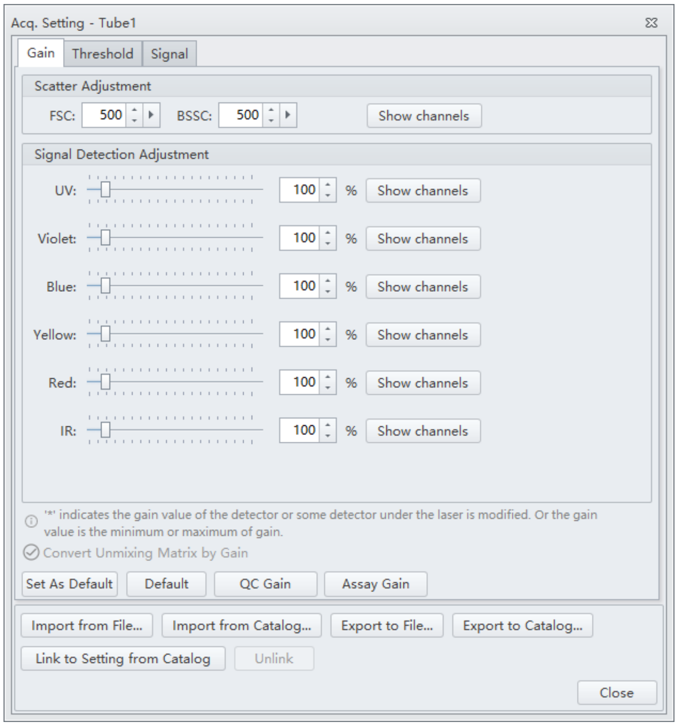
	Item	Date Created
	QC	2024-07-30 15:50:18
	10 color durapanel	2024-07-30 15:56:38

FSC 500 USSC 500 VSSC 500 BSSC 500
YSSC 500 RSSC 500 IRSSC 500
U1 500 U2 500 U3 500 U4 500
U5 500 U6 500 U7 500 U8 500
U9 500 U10 500 U11 500 U12 500
U13 500 U14 500 U15 500 U16 500
U17 500 U18 500 U19 500 U20 500

3 Select **Close**.

Exporting Instrument Settings

1 Select the desired sample tube to export and select Acq. Setting... . The Acq. Setting window appears.



FSC: 500
BSSC: 500
UV: 100 %
Violet: 100 %
Blue: 100 %
Yellow: 100 %
Red: 100 %
IR: 100 %

"*" indicates the gain value of the detector or some detector under the laser is modified. Or the gain value is the minimum or maximum of gain.

2 Select **Export to File** to export a current set of instrument settings. Specify the file path and file name and select **Save**.

NOTE The generated file ends in *.acq.

Or

Select **Export to Catalog**, give a name to the settings to be exported, and export the file to the software's Acquisition Setting Catalog, then select **OK**.

3 Select **Close**.

Importing and Exporting Unmixing Settings

The software supports unrestricted importing and exporting of unmixed data, regardless of whether the sample tube data has already been acquired. Imported unmixing values only cover channels identical with the current instrument configuration. The software automatically adjusts unmixing values according to differences in the gain level.

Printing Graphics

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Printing Graphics for detailed instructions.

Saving the Experiment

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Saving the Experiment for detailed instructions.

Concluding the Experiment

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Saving the Experiment for detailed instructions.

CHAPTER 7
Unmixing

Overview

Spectral unmixing is used to identify the fluorescence signal for each fluorospheres used in a given experiment. The spectral unmixing process is to disassemble sample raw data into single dye channels according to their spectral signatures. This chapter provides procedures for using the CytoFLEX mosaic Spectral Detection Module.

NOTE To switch between Spectral Mode and Conventional Mode, refer to CHAPTER 12, Switching between Spectral Mode and Conventional Mode.

Workflow:

Prepare spectral unmixing samples	➔	Create spectral unmixing experiment	➔	Acquire and record data	➔	Calculate spectral unmixing	➔	Apply spectral unmixing to sample for data analysis

This chapter contains information on:

Preparing the Spectral Unmixing Sample

For the negative control sample and single positive control sample, you can use blood, cell lines, or dedicated compensation beads such as VersaComp Antibody Capture Beads. For details, refer to the appropriate reagent instructions for use. The negative control tube is used to determine the autofluorescence of the sample.

Preparing the Spectral Unmixing Sample [Without Plate Loader]

To perform a spectral unmixing experiment, prepare:

- A single positive control tube for each color present in the panel
- A negative control tube

NOTE Users can add more than one single positive control tube or negative control tube as necessary.

Preparing the Spectral Unmixing Sample [With Plate Loader]

To perform a spectral unmixing experiment, prepare:

- A single positive control well for each color
- A negative control well

NOTE Users can add more than one single positive control tube or negative control tube as necessary.

Creating a Spectral Unmixing Experiment

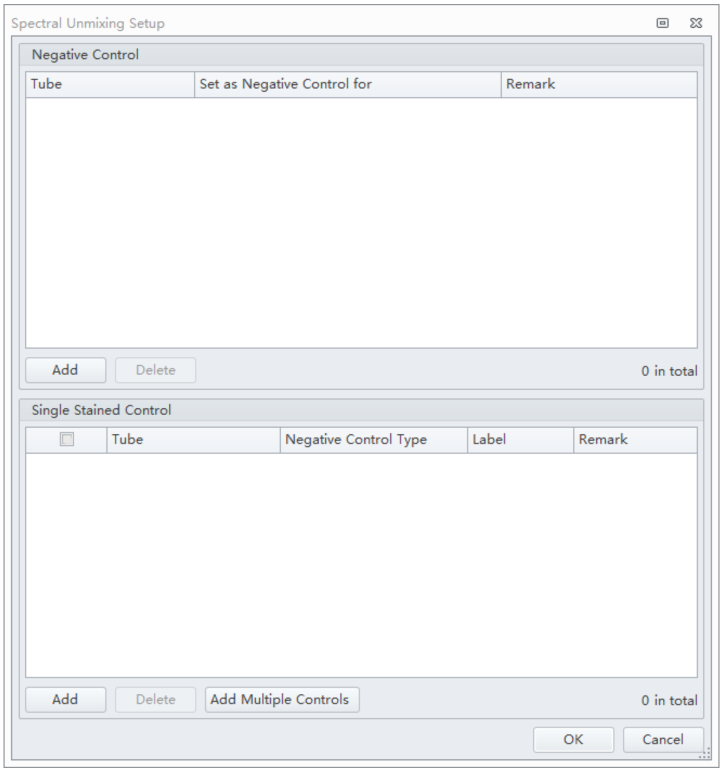
Creating a Spectral Unmixing Experiment [Without Plate Loader]

1. Select **File > New Spectral Unmixing** or **New Spectral Unmixing** on the start page to create a new spectral unmixing experiment.

   NOTE The file name of the newly created spectral unmixing experiment has a ".xitsp" suffix.

---

2 Navigate to the desired file path and select **Save.** The Spectral Unmixing Setup window appears.



NOTE A **Control** is the reference material used for creating the unmixing matrix.

⚠ CAUTION

Risk of erroneous results. Select a negative control tube, according to which the fluorescence background will be set. If there is not a negative control tube available, then the single color tube must have a negative population.

3 Add a negative control tube.

a. Select **Add.**

b. Edit the Tube name and Remark as needed.

c. Select the desired control type from the **Set as Negative Control for** drop-down menu.

> **NOTE** The default control type is **Sample and Single Stained**. If you select **Sample and Single Stained**, the tube will appear in the unmixing matrix and be used for unmixing calculation. If you select **Single Stained Only**, the tube will not appear in the unmixing matrix and not be used for unmixing calculation.

> **NOTE** To add more negative control tubes, repeat Steps a.-c. A maximum of 10 negative control tubes can be added for the spectral unmixing experiments.

> **NOTE** To delete a tube, select the desired tube and select **Delete**.

4. Add a single stained control tube.
   1. Select **Add**.
   2. Select the desired fluorescent tag from the **Tube** drop-down menu.

      NOTE To manage the fluorescent tag, refer to CHAPTER 2, Fluorescent Tag Library.

   3. Select the desired control type from the **Negative Control Type** drop-down menu.

      NOTE The default control type is **Internal Negative Control**. The unstained control tube can be selected if needed.

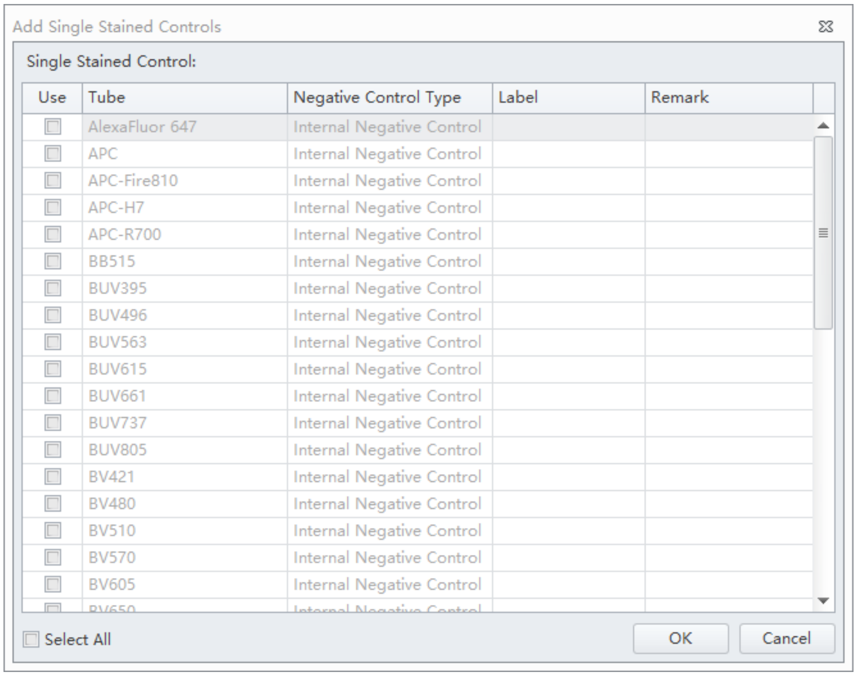
   4. Edit the Label and Remark as needed.

      NOTE Label information is retained in the unmixing matrix. Label information will be applied to the corresponding channels when the unmixing matrix is imported for the use in other experiments.

      NOTE To add more single stained control tubes, repeat Steps a.-d. A maximum of 60 single stained control tubes can be added for the spectral unmixing experiments.

      NOTE To delete a tube, select the checkbox to the left of the desired tube and select **Delete**.

      NOTE To add multiple single stained control tubes, select **Add Multiple Controls**. The Add Single Stained Controls window appears.



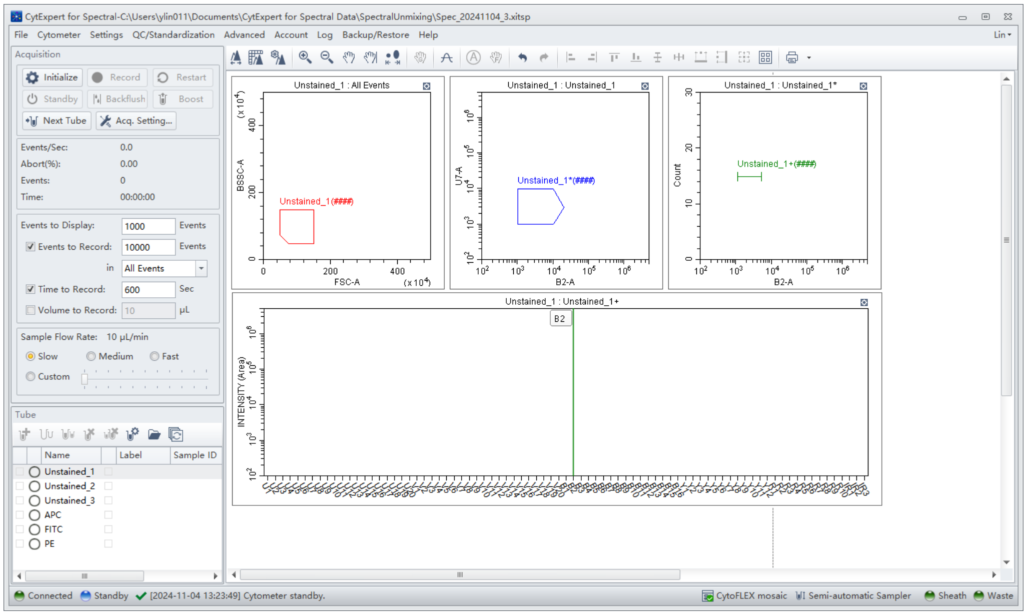
Use	Tube	Negative Control Type	Label	Remark
☐	AlexaFluor 647	Internal Negative Control		
☐	APC	Internal Negative Control		
☐	APC-Fire810	Internal Negative Control		
☐	APC-H7	Internal Negative Control		
☐	APC-R700	Internal Negative Control		
☐	BB515	Internal Negative Control		
☐	BUV395	Internal Negative Control		
☐	BUV496	Internal Negative Control		
☐	BUV563	Internal Negative Control		
☐	BUV615	Internal Negative Control		
☐	BUV661	Internal Negative Control		
☐	BUV737	Internal Negative Control		
☐	BUV805	Internal Negative Control		
☐	BV421	Internal Negative Control		
☐	BV480	Internal Negative Control		
☐	BV510	Internal Negative Control		
☐	BV570	Internal Negative Control		
☐	BV605	Internal Negative Control		
☐	BV650	Internal Negative Control		

Select the **Use** checkbox to add the desired tube. Select the desired control type from the Negative Control Type drop-down menu. Edit the Label and Remark as needed. Select **OK**.

NOTE The **Select All** checkbox allows you to select all of the tubes listed.

5 Select **OK**.

After confirmation, the software automatically generates the following unmixing spectral experiment.



CytExpert for Spectral-C:\Users\ylin011\Documents\CytExpert for Spectral Data\SpectralUnmixing\Spec_20241104_3.xitsp

Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

Name	Label	Sample ID
Unstained_1		
Unstained_2		
Unstained_3		
APC		
FITC		
PE		

Connected Standby [2024-11-04 13:23:49] Cytometer standby.

6 Confirm that the gating and peak channel are appropriate. Use the pan tool to adjust the axis scale so that the sample signal appears in a suitable position. Adjust the gate so that it encloses the target cell population. Refer to CHAPTER 6, Creating Plots and Gates.

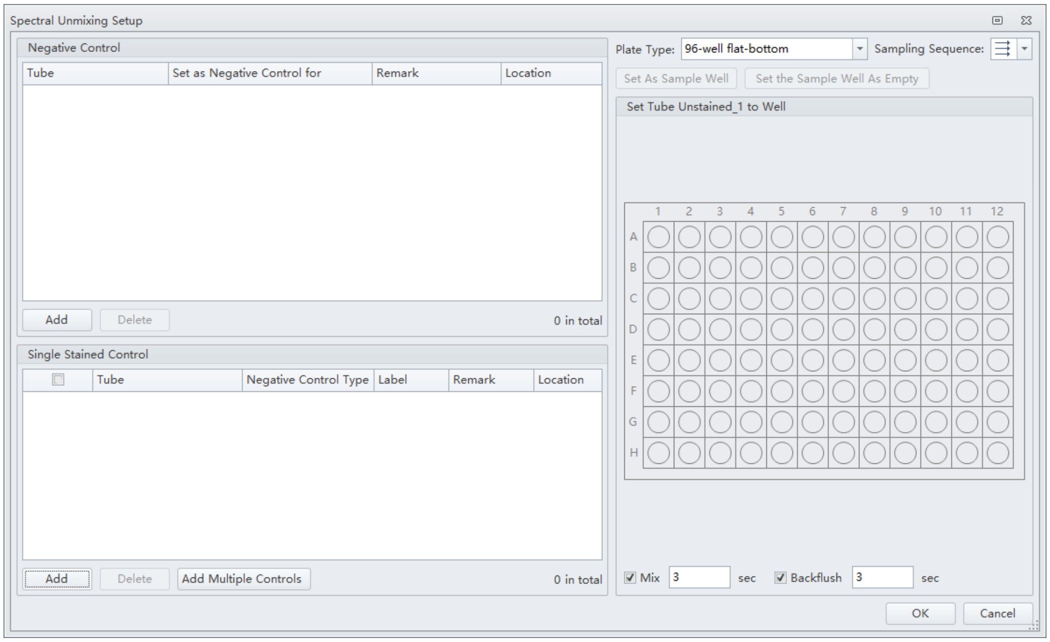
Creating a Spectral Unmixing Experiment [With Plate Loader]

1 Select **File** > **New Spectral Unmixing** or **New Spectral Unmixing** on the start page to create a new spectral unmixing experiment.

NOTE The file name of the newly created spectral unmixing experiment has a ".xitsp" suffix.

---

2 Navigate to the desired file path and select **Save**. The Spectral Unmixing Setup window appears.



0 in total

0 in total

Plate Type: 96-well flat-bottom

☑ Mix 3 sec

☑ Backflush 3 sec

NOTE A **Control** is the reference material used for creating the unmixing matrix.

⚠ CAUTION

Risk of erroneous results. Select a negative control tube, according to which the fluorescence background will be set. If there is not a negative control tube available, then the single color tube must have a negative population.

3 Add a negative control tube.

a. Select **Add**.

b. Edit the Tube name and Remark as needed.

c. Select the desired control type from the **Set as Negative Control for** drop-down menu.

NOTE The default control type is **Sample and Single Stained**. If you select **Sample and Single Stained**, the tube will appear in the unmixing matrix and be used for unmixing calculation. If you select **Single Stained Only**, the tube will not appear in the unmixing matrix and not be used for unmixing calculation.

d. Select the plate type, sampling sequence, mix and backflush settings for the tube.

e. Select **Set As Sample Well** to assign well location.

NOTE To add more negative control tubes, repeat Steps a.-e. A maximum of 10 negative control tubes can be added for the spectral unmixing experiments.

4 Add a single stained control tube.

a. Select **Add**.

b. Select the desired fluorescent tag from the **Tube** drop-down menu.

NOTE To manage the fluorescent tag, refer to CHAPTER 2, Fluorescent Tag Library.

c. Select the desired control type from the **Negative Control Type** drop-down menu.

NOTE The default control type is **Internal Negative Control**. The unstained control tube can be selected if needed.

d. Edit the Label and Remark as needed.

NOTE Label information is retained in the unmixing matrix. Label information will be applied to the corresponding channels when the unmixing matrix is imported for the use in other experiments.

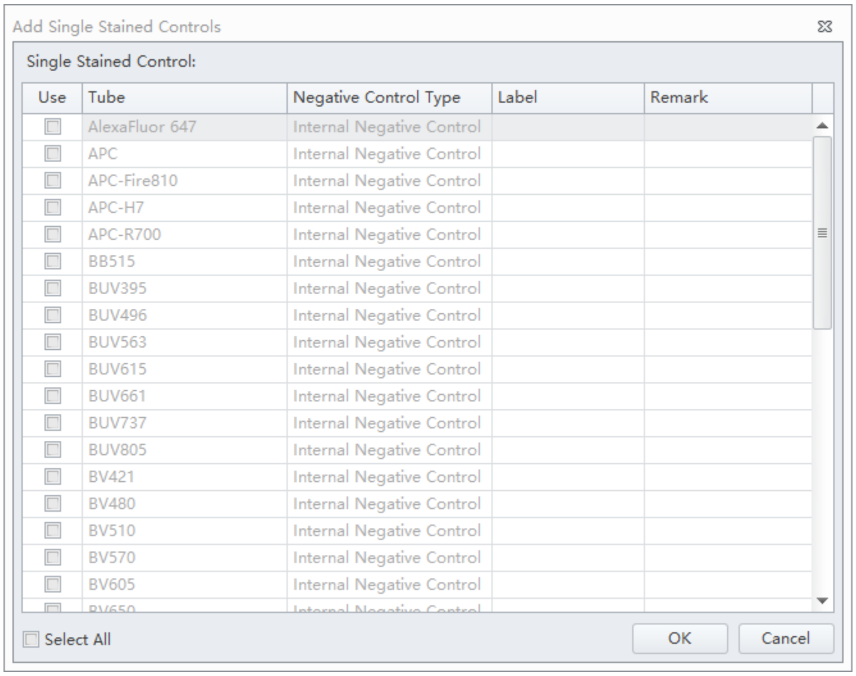
e. Select the plate type, sampling sequence, mix and backflush settings for the tube.

f. Select **Set As Sample Well** to assign well location.

NOTE To add more single stained control tubes, repeat Steps a.-f. A maximum of 60 single stained control tubes can be added for the spectral unmixing experiments.

NOTE To delete a tube, select the checkbox to the left of the desired tube and select **Delete**.

NOTE To add multiple single stained control tubes, select **Add Multiple Controls**. The Add Single Stained Controls window appears.



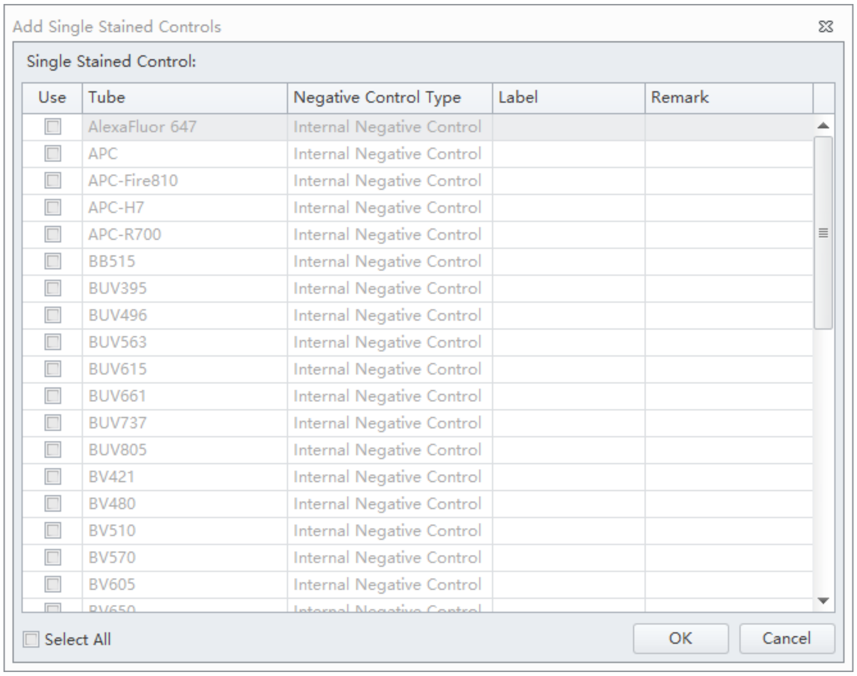
Use	Tube	Negative Control Type	Label	Remark
☐	AlexaFluor 647	Internal Negative Control		
☐	APC	Internal Negative Control		
☐	APC-Fire810	Internal Negative Control		
☐	APC-H7	Internal Negative Control		
☐	APC-R700	Internal Negative Control		
☐	BB515	Internal Negative Control		
☐	BUV395	Internal Negative Control		
☐	BUV496	Internal Negative Control		
☐	BUV563	Internal Negative Control		
☐	BUV615	Internal Negative Control		
☐	BUV661	Internal Negative Control		
☐	BUV737	Internal Negative Control		
☐	BUV805	Internal Negative Control		
☐	BV421	Internal Negative Control		
☐	BV480	Internal Negative Control		
☐	BV510	Internal Negative Control		
☐	BV570	Internal Negative Control		
☐	BV605	Internal Negative Control		

Select the **Use** checkbox to add the desired tube. Select the desired control type from the Negative Control Type drop-down menu. Edit the Label and Remark as needed. Select **OK**.

NOTE The **Select All** checkbox allows you to select all of the tubes listed.

---

5 If you need to add multiple single stained control tubes, select **Add Multiple Controls**. The Add Single Stained Controls window appears.



Use	Tube	Negative Control Type	Label	Remark
☐	AlexaFluor 647	Internal Negative Control		
☐	APC	Internal Negative Control		
☐	APC-Fire810	Internal Negative Control		
☐	APC-H7	Internal Negative Control		
☐	APC-R700	Internal Negative Control		
☐	BB515	Internal Negative Control		
☐	BUV395	Internal Negative Control		
☐	BUV496	Internal Negative Control		
☐	BUV563	Internal Negative Control		
☐	BUV615	Internal Negative Control		
☐	BUV661	Internal Negative Control		
☐	BUV737	Internal Negative Control		
☐	BUV805	Internal Negative Control		
☐	BV421	Internal Negative Control		
☐	BV480	Internal Negative Control		
☐	BV510	Internal Negative Control		
☐	BV570	Internal Negative Control		
☐	BV605	Internal Negative Control		

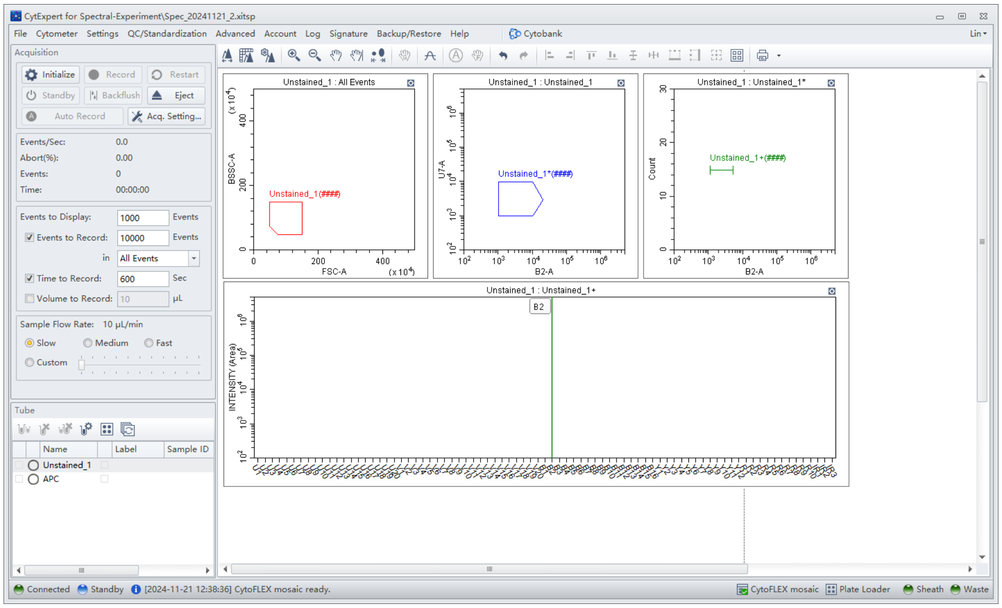
Select the **Use** checkbox to add the desired tube. Select the desired control type from the Negative Control Type drop-down menu. Edit the Label and Remark as needed. Select **OK**.

NOTE The **Select All** checkbox allows you to select all of the tubes listed

Select the plate type, sampling sequence, mix and backflush settings for all the tubes added.

6 Select **OK**.

After confirmation, the software automatically generates the following unmixing spectral experiment.



Events/Sec: 0.0
Abort(%): 0.00
Events: 0
Time: 00:00:00

Events to Display: 1000 Events
☑ Events to Record: 10000 Events in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 µL

Sample Flow Rate: 10 µL/min
● Slow ○ Medium ○ Fast
○ Custom

○ Unstained_1
○ APC

Connected Standby [2024-11-21 12:38:36] CytoFLEX mosaic ready.

---

7 Confirm that the gating and peak channel are appropriate. Use the pan tool to adjust the axis scale so that the sample signal appears in a suitable position. Adjust the gate so that it encloses the target cell population. Refer to CHAPTER 6, Creating Plots and Gates.

---

8 Before acquiring data, ensure the plate has been loaded properly. Data can be acquired as a single well or as a set of wells. Refer to CHAPTER 6, Sampling and Collecting Data [With Plate Loader].

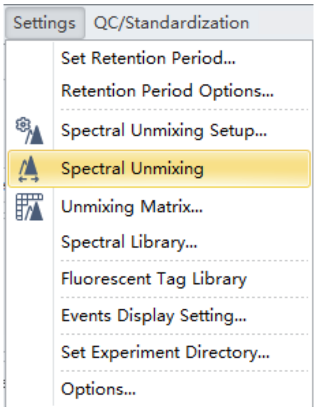
Loading Sample and Recording Data

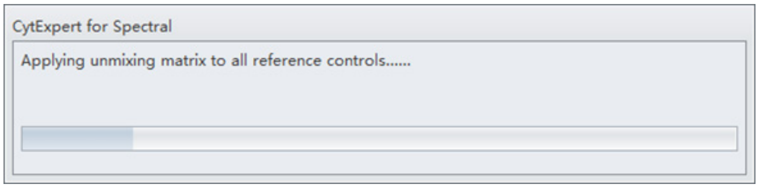
Refer to CHAPTER 6, Load Sample and Record Data for detailed instructions on loading sample and recording data.

NOTE If you change the acquisition settings of a negative control tube, the new acquisition settings will apply to all relevant single stained control tubes without data.

Calculating Unmixing Values

1 Select or select **Spectral Unmixing** from the Settings menu to calculate the spectral unmixing values. The following system prompt appears.

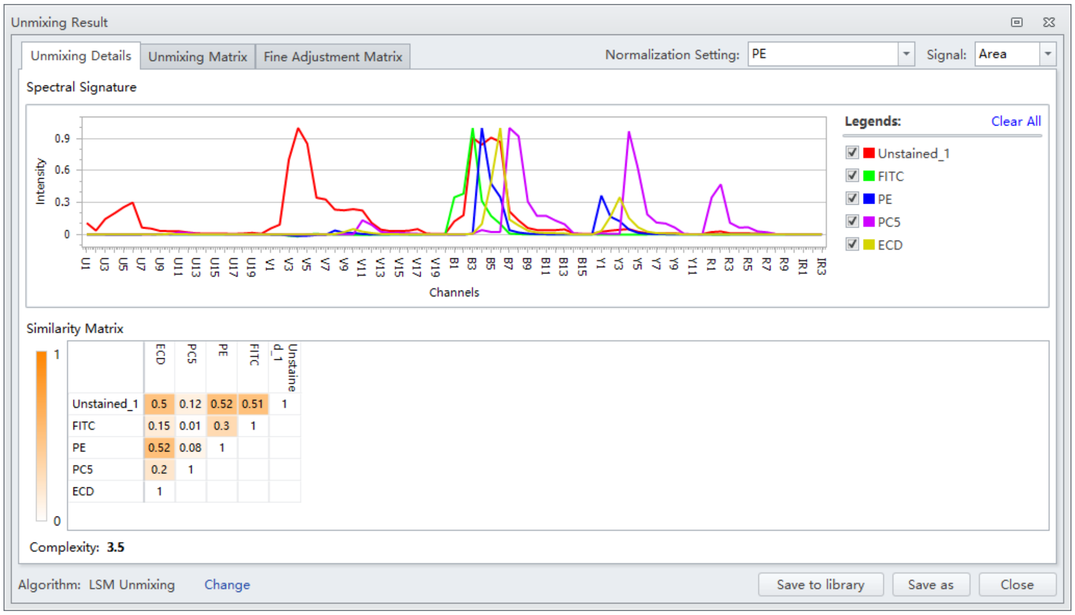




Applying unmixing matrix to all reference controls......

---

2 The Unmixing Result window appears.



Normalization Setting: PE

Signal: Area

	ECD	PC5	PE	FITC	Unstained_1
Unstained_1	0.5	0.12	0.52	0.51	1
FITC	0.15	0.01	0.3	1	
PE	0.52	0.08	1		
PC5	0.2	1			
ECD	1				

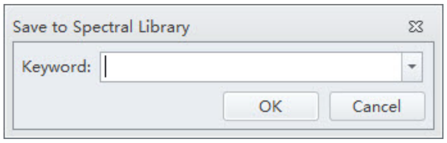
Complexity: **3.5**

Algorithm: LSM Unmixing

3 Select **Save As** to export the unmixing matrix as a .unmix file and specify where to save it.

NOTE The unmixing matrix can also be imported for use in other experiments.

4 Select **Save to Library** to save the unmixing settings in the spectral library. The Save to Spectral Library window appears.



5 Specify the Keyword and select **OK**.

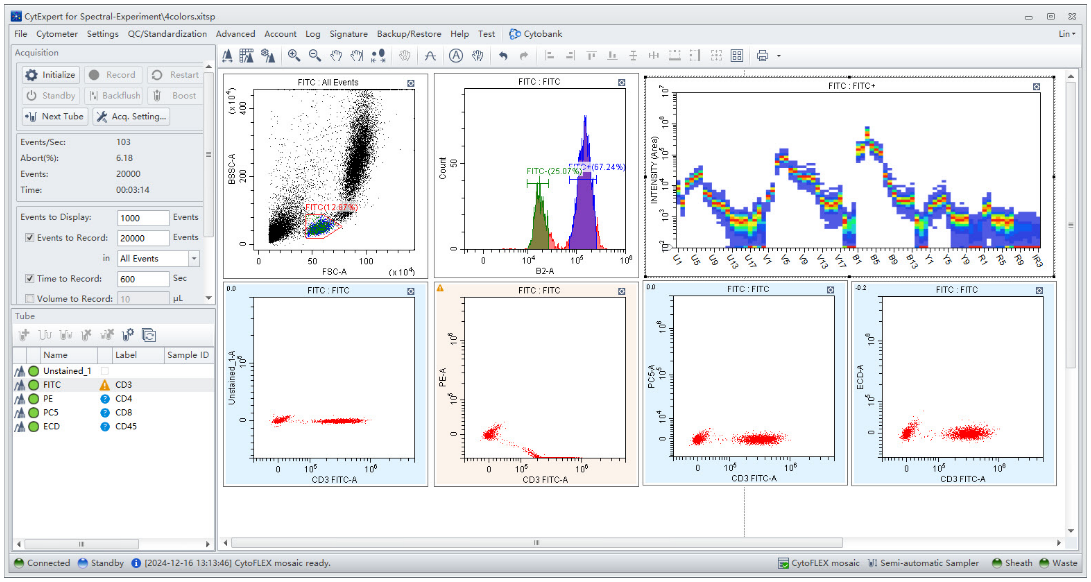
NOTE The settings stored in the spectral library are specific to the detector configuration. The spectral library can only be applied when the detector configuration are the same.

6 Select **Close**.

NOTE At any time, saved spectral unmixing experiments can be reopened and the unmixing values can be recalculated.

Confirming Unmixing Results

1 Select a sample tube from the tube list on the left side of the screen and the unmixing plots appear on the plot area.



Events/Sec: 103
Abort(%): 6.18
Events: 20000
Time: 00:03:14

Events to Display: 1000 Events
☑ Events to Record: 20000 Events
in All Events
☑ Time to Record: 600 Sec
☐ Volume to Record: 10 μL

Name		Label	Sample ID
Unstained_1	☐		
FITC	⚠	CD3	
PE	?	CD4	
PC5	?	CD8	
ECD	?	CD45	

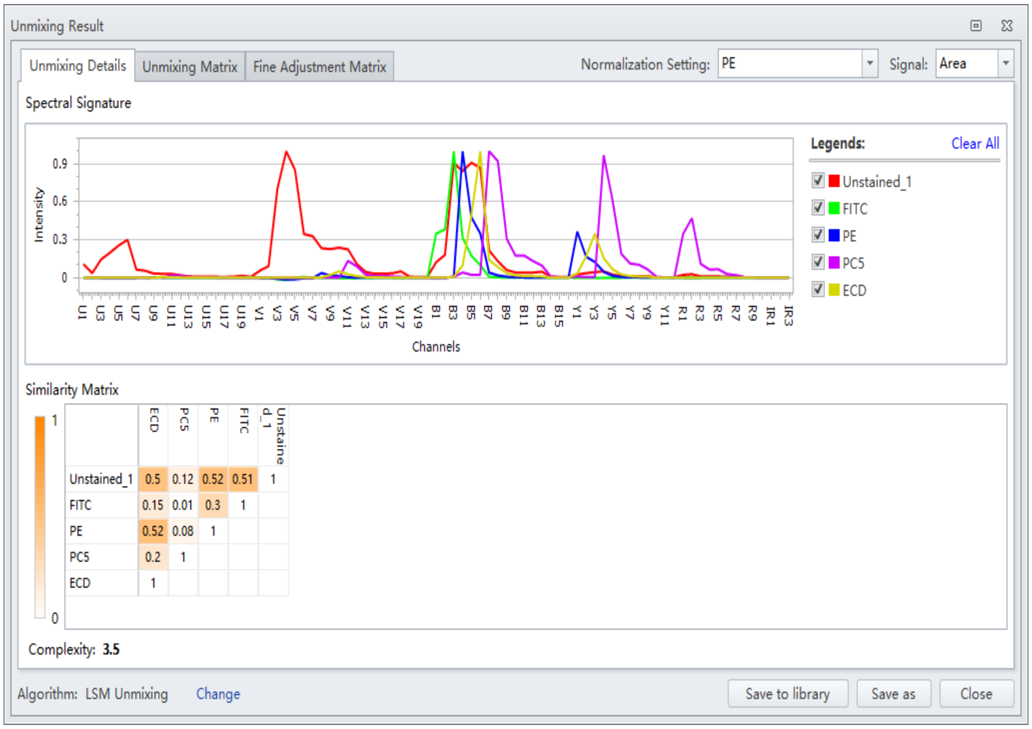
Connected Standby [2024-12-16 13:13:46] CytoFLEX mosaic ready.

NOTE The unmixing results appears on the right of the Name column. The ⚠ indicates that there is an unmixing gap. The ? indicates that the unmixing gap should be recalculated and refreshed. Refer to Fine Adjustment.

NOTE The software uses the positive-negative method to calculate the unmixing gap.

$$GAP_{(y\ to\ x)} = \frac{Y_{(Positive)} - Y_{(Negative)}}{X_{(Positive)} - X_{(Negative)}}$$

2 Select or select **Spectral Unmixing** from the Settings menu. The Unmixing Result window appears.



Normalization Setting: PE

Signal: Area

Similarity Matrix

	ECD	PC5	PE	FITC	Unstained_1
Unstained_1	0.5	0.12	0.52	0.51	1
FITC	0.15	0.01	0.3	1	
PE	0.52	0.08	1		
PC5	0.2	1			
ECD	1				

Complexity: 3.5

Algorithm: LSM Unmixing

NOTE The **Change** button in the lower left of the Unmixing Result window allows you to change the unmixing algorithm. The two options currently offered are **LSM Unmixing** and **Poisson Unmixing**.

NOTE The **Normalization Setting** drop-down menu in the top right of the Unmixing Result window allows you to select the tube gain settings to apply to the unmixing matrix.

NOTE The **Signal** drop-down menu in the top right of the Unmixing Result window allows you to select which signal type to use, **Height** or **Area**.

NOTE In the Unmixing Details window:

- The spectral signature chart displays the unmixing matrix values based on the peak channel. Use the **Legends** checkbox to choose which unmixing control's spectral signature to show.
- The similarity index is a number between 0-1. The CytExpert for Spectral software displays the value in orange as a visual clue. The darker the shade, the higher the similarity.
- The complexity index is a measure of how distinguishable a collection of spectral signatures are from each other when unmixed together. The complexity index is used to assess the stability of a linear system.

NOTE In the Unmixing Matrix window, the CytExpert for Spectral software displays the unmixing values in either blue, or black, or orange, or red as a visual clue.

- Blue denotes the value falls below 0, which requires special attention.
- Black denotes the value falls within 0 - 100, which is acceptable.

NOTE In the Fine Adjustment Matrix window:

- The **Use** checkbox applies the fine adjustment to the selected sample.

NOTE The Fine Adjustment range is from -500 to 500 depending on the spillover percentage calculated. The CytExpert for Spectral software displays the value in either black, or orange, or red as a visual clue.

- Black denotes the value falls within 0 - 99.99, which is acceptable.
- Orange denotes the value falls within 100 - 399.99, or -99.99 to -0.01, which requires special attention.
- Red denotes the value is within 400 - 500 or -500 to -100, which is unacceptable.

---

3 If there is an unmixing gap, follow the procedure below:

a. Verify whether the allocated single stained control tube was prepared as required and correctly positioned.

b. Verify that the gating and population hierarchy in each tube were correct.

NOTE The minimum recommended number of events in the specified gate is 50.

c. Verify that the peak channel selected in each tube was appropriate.

d. Repeat Calculating Unmixing Values to recalculate and export the unmixing results.

---

4 **Optional:** Use the Fine Adjustment function to adjust the unmixing gaps as needed. Refer to Fine Adjustment.

Saving the Spectral Unmixing Experiment

Selecting **Save** in the File menu allows you to save the experiment.

Selecting **Save As** and saving the experiment under a different name allows you to create a backup.

At any time, saved spectral unmixing experiments can be reopened and the unmixing values recalculated.

Concluding the Experiment

Conclude the experiment as follows:

- Select **Standby** to return the instrument to the standby state.
- Select **File > Close Experiment** to clear the experiment and return to the Start Page.

NOTE If changes were made to the experiment, the software prompts you to save the latest changes in the experiment before returning to the Start Page.

- Shut down the system. Refer to CHAPTER 9, Daily Shutdown.

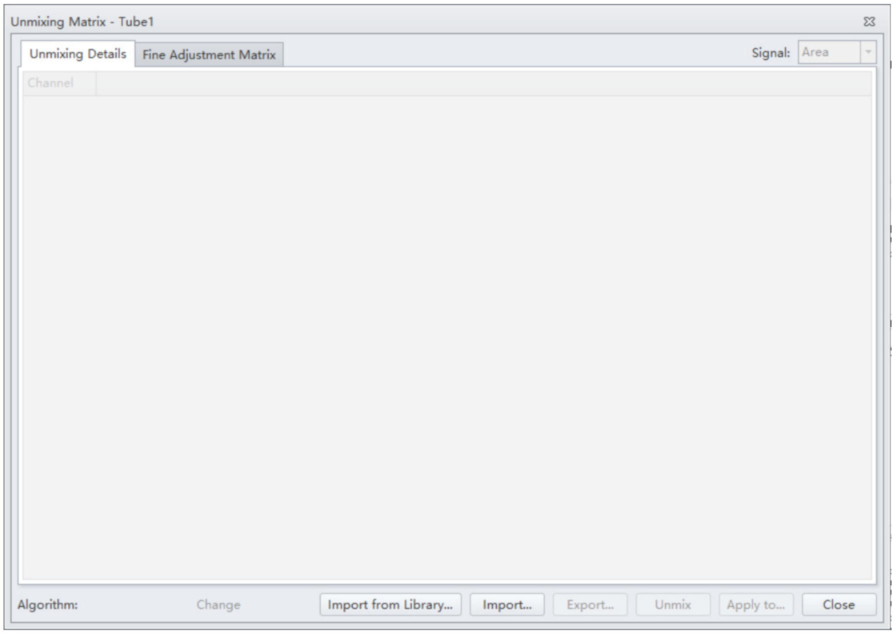
Adjusting Spectral Unmixing

Importing and Exporting Unmixing Matrix

Importing Unmixing Settings from Unmixing Matrix Files

1 Create a new experiment or open a saved experiment. Refer to CHAPTER 6, Creating an Experiment.

2 Select the desired sample tube and select or select **Settings** > **Unmixing Matrix**. The Unmixing Matrix window appears.



---

3 Select **Import** and locate the path where unmixing matrix files are saved. Select the corresponding unmixing matrix file (.unmix) to import the unmixing values.

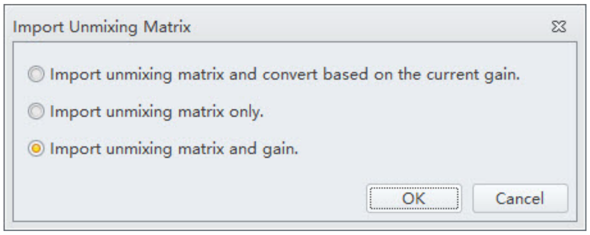
NOTE To import unmixing values from the spectral library, refer to Importing Unmixing Settings from the Spectral Library.

Both methods allow you to apply the gain independent unmixing capability. For example, importing the unmixing values with or without recalculating the unmixing matrix based on the gain settings.

NOTE Fine adjustment matrix is included in the unmixing matrix, and can be imported with the unmixing matrix files (.unmix).

---

4 After opening the desired unmixing file, the Import Unmixing Matrix window appears.



Import Unmixing Matrix

○ Import unmixing matrix and convert based on the current gain.
○ Import unmixing matrix only.
● Import unmixing matrix and gain.

Select one of the following:

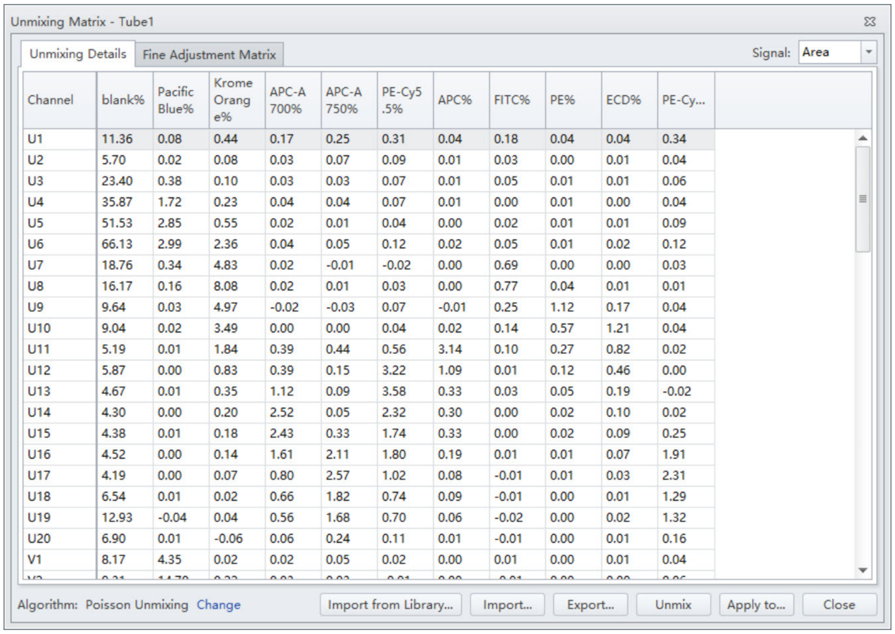
- Import unmixing matrix and convert based on the current gain.
- Import unmixing matrix only.
- Import unmixing matrix and gain.

NOTE

- If the tube does not have any data when importing unmixing values calculated from other instrument settings, the software prompts you to select whether the gain settings must be imported as well. Select **Yes** to import fluorescence channel gains settings along with the rest of the data. Select **No** to allow the CytExpert for Spectral software to adjust the unmixing matrix values based on the current gain settings.
- If the tube does have data when importing unmixing values from other instrument settings, the software prompts you to select whether the unmixing values are adjusted based on the current gain settings.
- It is important to note that automatic adjustments to unmixing values calculated from other instrument gain settings could result in incorrect unmixing. Always review the data after importing unmixing values to ensure the sample is unmixed properly.

---

5 Select **OK**. The imported unmixing matrix appears in the Unmixing Matrix window.



Signal: Area

Channel	blank%	Pacific Blue%	Krome Orange%	APC-A 700%	APC-A 750%	PE-Cy5 .5%	APC%	FITC%	PE%	ECD%	PE-Cy...
U1	11.36	0.08	0.44	0.17	0.25	0.31	0.04	0.18	0.04	0.04	0.34
U2	5.70	0.02	0.08	0.03	0.07	0.09	0.01	0.03	0.00	0.01	0.04
U3	23.40	0.38	0.10	0.03	0.03	0.07	0.01	0.05	0.01	0.01	0.06
U4	35.87	1.72	0.23	0.04	0.04	0.07	0.01	0.00	0.01	0.00	0.04
U5	51.53	2.85	0.55	0.02	0.01	0.04	0.00	0.02	0.01	0.01	0.09
U6	66.13	2.99	2.36	0.04	0.05	0.12	0.02	0.05	0.01	0.02	0.12
U7	18.76	0.34	4.83	0.02	-0.01	-0.02	0.00	0.69	0.00	0.00	0.03
U8	16.17	0.16	8.08	0.02	0.01	0.03	0.00	0.77	0.04	0.01	0.01
U9	9.64	0.03	4.97	-0.02	-0.03	0.07	-0.01	0.25	1.12	0.17	0.04
U10	9.04	0.02	3.49	0.00	0.00	0.04	0.02	0.14	0.57	1.21	0.04
U11	5.19	0.01	1.84	0.39	0.44	0.56	3.14	0.10	0.27	0.82	0.02
U12	5.87	0.00	0.83	0.39	0.15	3.22	1.09	0.01	0.12	0.46	0.00
U13	4.67	0.01	0.35	1.12	0.09	3.58	0.33	0.03	0.05	0.19	-0.02
U14	4.30	0.00	0.20	2.52	0.05	2.32	0.30	0.00	0.02	0.10	0.02
U15	4.38	0.01	0.18	2.43	0.33	1.74	0.33	0.00	0.02	0.09	0.25
U16	4.52	0.00	0.14	1.61	2.11	1.80	0.19	0.01	0.01	0.07	1.91
U17	4.19	0.00	0.07	0.80	2.57	1.02	0.08	-0.01	0.01	0.03	2.31
U18	6.54	0.01	0.02	0.66	1.82	0.74	0.09	-0.01	0.00	0.01	1.29
U19	12.93	-0.04	0.04	0.56	1.68	0.70	0.06	-0.02	0.00	0.02	1.32
U20	6.90	0.01	-0.06	0.06	0.24	0.11	0.01	-0.01	0.00	0.01	0.16
V1	8.17	4.35	0.02	0.02	0.05	0.02	0.00	0.01	0.00	0.01	0.04

Algorithm: Poisson Unmixing

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6 If necessary, select Apply to... to apply the unmixing values to the selected test tubes.

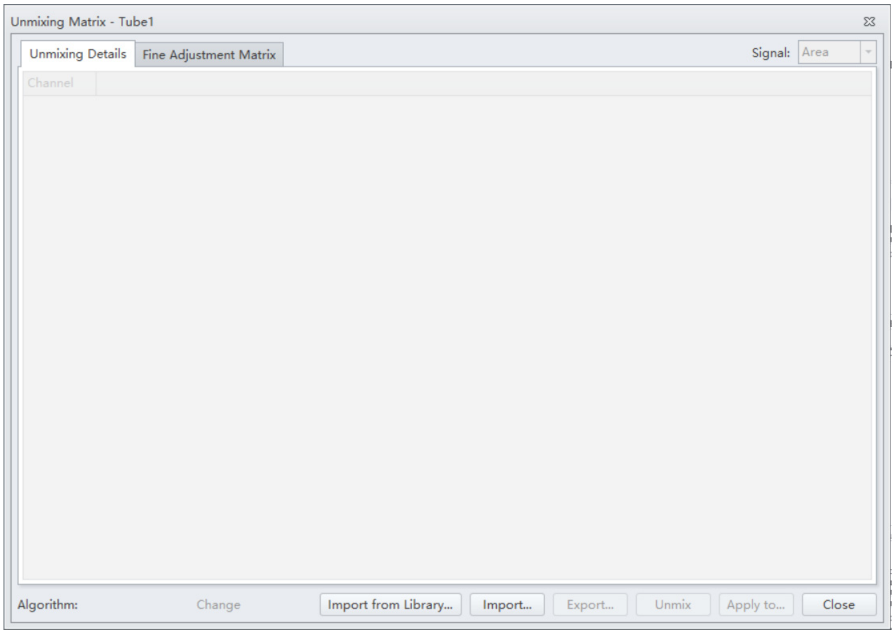
---

7 Select **Close** to exit the Unmixing Matrix window.

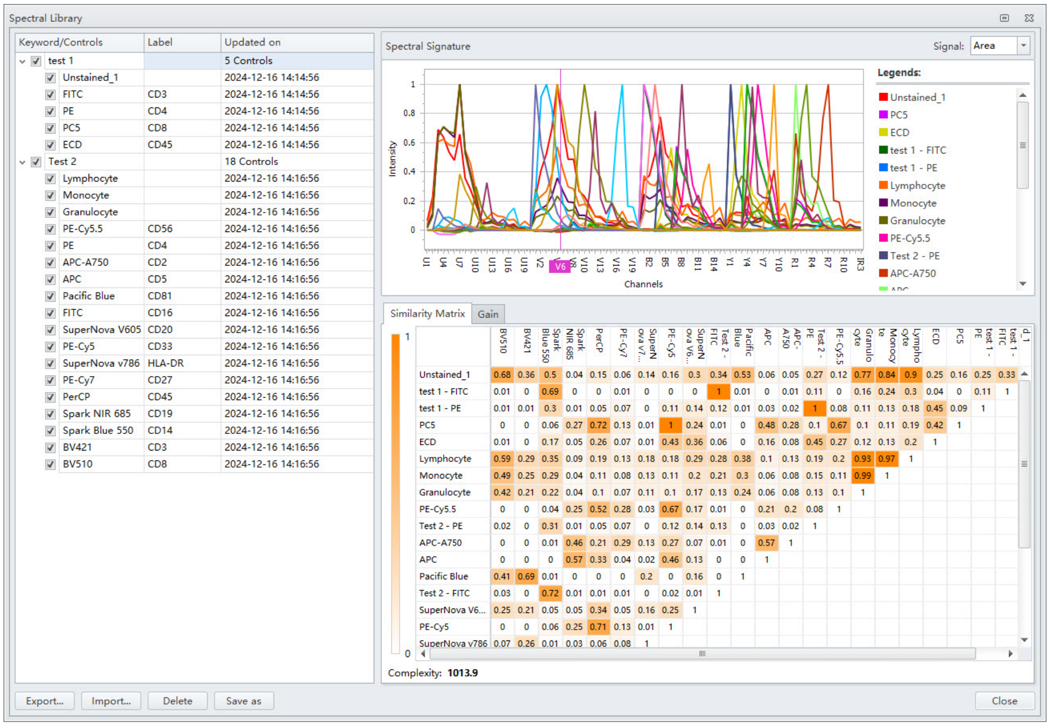
Importing Unmixing Settings from the Spectral Library

1 Create a new experiment or open a saved experiment. Refer to CHAPTER 6, Creating an Experiment.

2 Select the desired sample tube and select or select **Settings > Unmixing Matrix**. The Unmixing Matrix window appears.



3 Select Import from Library... . The Spectral Library window appears.



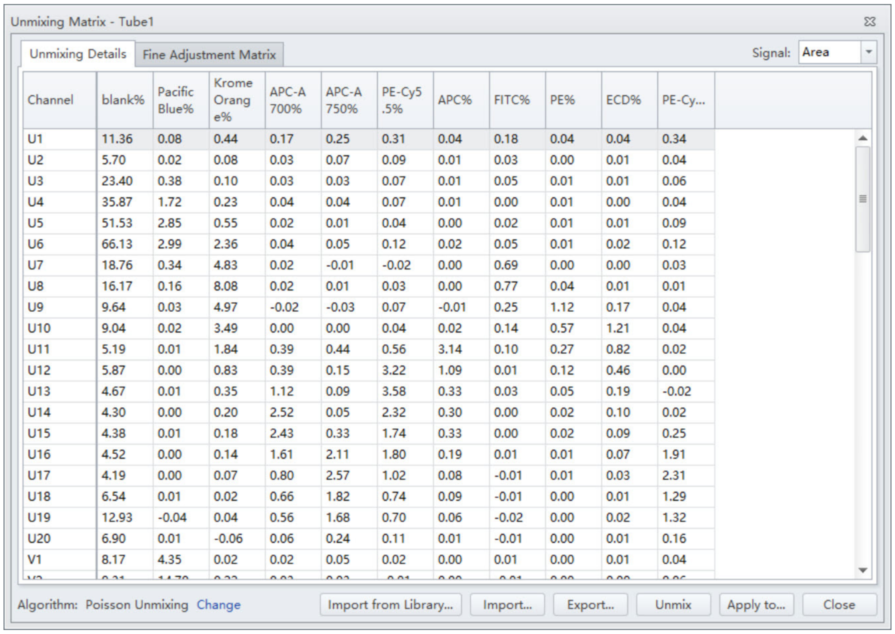
Keyword/Controls	Label	Updated on
☑ test 1		5 Controls
☑ Unstained_1		2024-12-16 14:14:56
☑ FITC	CD3	2024-12-16 14:14:56
☑ PE	CD4	2024-12-16 14:14:56
☑ PC5	CD8	2024-12-16 14:14:56
☑ ECD	CD45	2024-12-16 14:14:56
☑ Test 2		18 Controls
☑ Lymphocyte		2024-12-16 14:16:56
☑ Monocyte		2024-12-16 14:16:56
☑ Granulocyte		2024-12-16 14:16:56
☑ PE-Cy5.5	CD56	2024-12-16 14:16:56
☑ PE	CD4	2024-12-16 14:16:56
☑ APC-A750	CD2	2024-12-16 14:16:56
☑ APC	CD5	2024-12-16 14:16:56
☑ Pacific Blue	CD81	2024-12-16 14:16:56
☑ FITC	CD16	2024-12-16 14:16:56
☑ SuperNova V605	CD20	2024-12-16 14:16:56
☑ PE-Cy5	CD33	2024-12-16 14:16:56
☑ SuperNova v786	HLA-DR	2024-12-16 14:16:56
☑ PE-Cy7	CD27	2024-12-16 14:16:56
☑ PerCP	CD45	2024-12-16 14:16:56
☑ Spark NIR 685	CD19	2024-12-16 14:16:56
☑ Spark Blue 550	CD14	2024-12-16 14:16:56
☑ BV421	CD3	2024-12-16 14:16:56
☑ BV510	CD8	2024-12-16 14:16:56

	BV510	BV421	Spark Blue 550	Spark NIR 685	PerCP	PE-Cy7	SuperNova v7...	PE-Cy5	SuperNova V6...	Test 2 - FITC	Pacific Blue	APC	APC-A750	Test 2 - PE	PE-Cy5.5	Granulocyte	Monocyte	Lymphocyte	ECD	PC5	test 1 - PE	test 1 - FITC	d_1
Unstained_1	0.68	0.36	0.5	0.04	0.15	0.06	0.14	0.16	0.3	0.34	0.53	0.06	0.05	0.27	0.12	0.77	0.84	0.9	0.25	0.16	0.25	0.33	
test 1 - FITC	0.01	0	0.69	0	0	0.01	0	0	0	1	0.01	0	0.01	0.11	0	0.16	0.24	0.3	0.04	0	0.11	1	
test 1 - PE	0.01	0.01	0.3	0.01	0.05	0.07	0	0.11	0.14	0.12	0.01	0.03	0.02	1	0.08	0.11	0.13	0.18	0.45	0.09	1		
PC5	0	0	0.06	0.27	0.72	0.13	0.01	1	0.24	0.01	0	0.48	0.28	0.1	0.67	0.1	0.11	0.19	0.42	1			
ECD	0.01	0	0.17	0.05	0.26	0.07	0.01	0.43	0.36	0.06	0	0.16	0.08	0.45	0.27	0.12	0.13	0.2	1				
Lymphocyte	0.59	0.29	0.35	0.09	0.19	0.13	0.18	0.18	0.29	0.28	0.38	0.1	0.13	0.19	0.2	0.93	0.97	1					
Monocyte	0.49	0.25	0.29	0.04	0.11	0.08	0.13	0.11	0.2	0.21	0.3	0.06	0.08	0.15	0.11	0.99	1						
Granulocyte	0.42	0.21	0.22	0.04	0.1	0.07	0.11	0.1	0.17	0.13	0.24	0.06	0.08	0.13	0.1	1							
PE-Cy5.5	0	0	0.04	0.25	0.52	0.28	0.03	0.67	0.17	0.01	0	0.21	0.2	0.08	1								
Test 2 - PE	0.02	0	0.31	0.01	0.05	0.07	0	0.12	0.14	0.13	0	0.03	0.02	1									
APC-A750	0	0	0.01	0.46	0.21	0.29	0.13	0.27	0.07	0.01	0	0.57	1										
APC	0	0	0	0.57	0.33	0.04	0.02	0.46	0.13	0	0	1											
Pacific Blue	0.41	0.69	0.01	0	0	0	0.2	0	0.16	0	1												
Test 2 - FITC	0.03	0	0.72	0.01	0.01	0.01	0	0.02	0.01	1													
SuperNova V6...	0.25	0.21	0.05	0.05	0.34	0.05	0.16	0.25	1														
PE-Cy5	0	0	0.06	0.25	0.71	0.13	0.01	1															
SuperNova v786	0.07	0.26	0.01	0.03	0.06	0.08	1																

Complexity: **1013.9**

4 In the Keyword/Controls column, the corresponding unmixing values can be selected for each channel. The unmixing values of the same keyword/controls can also be selected using the drop-down menus in the Keyword/Controls column.

5 Select **OK.** The imported unmixing matrix appears in the Unmixing Matrix window.



Signal: Area

Channel	blank%	Pacific Blue%	Krome Orange%	APC-A 700%	APC-A 750%	PE-Cy5 .5%	APC%	FITC%	PE%	ECD%	PE-Cy...
U1	11.36	0.08	0.44	0.17	0.25	0.31	0.04	0.18	0.04	0.04	0.34
U2	5.70	0.02	0.08	0.03	0.07	0.09	0.01	0.03	0.00	0.01	0.04
U3	23.40	0.38	0.10	0.03	0.03	0.07	0.01	0.05	0.01	0.01	0.06
U4	35.87	1.72	0.23	0.04	0.04	0.07	0.01	0.00	0.01	0.00	0.04
U5	51.53	2.85	0.55	0.02	0.01	0.04	0.00	0.02	0.01	0.01	0.09
U6	66.13	2.99	2.36	0.04	0.05	0.12	0.02	0.05	0.01	0.02	0.12
U7	18.76	0.34	4.83	0.02	-0.01	-0.02	0.00	0.69	0.00	0.00	0.03
U8	16.17	0.16	8.08	0.02	0.01	0.03	0.00	0.77	0.04	0.01	0.01
U9	9.64	0.03	4.97	-0.02	-0.03	0.07	-0.01	0.25	1.12	0.17	0.04
U10	9.04	0.02	3.49	0.00	0.00	0.04	0.02	0.14	0.57	1.21	0.04
U11	5.19	0.01	1.84	0.39	0.44	0.56	3.14	0.10	0.27	0.82	0.02
U12	5.87	0.00	0.83	0.39	0.15	3.22	1.09	0.01	0.12	0.46	0.00
U13	4.67	0.01	0.35	1.12	0.09	3.58	0.33	0.03	0.05	0.19	-0.02
U14	4.30	0.00	0.20	2.52	0.05	2.32	0.30	0.00	0.02	0.10	0.02
U15	4.38	0.01	0.18	2.43	0.33	1.74	0.33	0.00	0.02	0.09	0.25
U16	4.52	0.00	0.14	1.61	2.11	1.80	0.19	0.01	0.01	0.07	1.91
U17	4.19	0.00	0.07	0.80	2.57	1.02	0.08	-0.01	0.01	0.03	2.31
U18	6.54	0.01	0.02	0.66	1.82	0.74	0.09	-0.01	0.00	0.01	1.29
U19	12.93	-0.04	0.04	0.56	1.68	0.70	0.06	-0.02	0.00	0.02	1.32
U20	6.90	0.01	-0.06	0.06	0.24	0.11	0.01	-0.01	0.00	0.01	0.16
V1	8.17	4.35	0.02	0.02	0.05	0.02	0.00	0.01	0.00	0.01	0.04

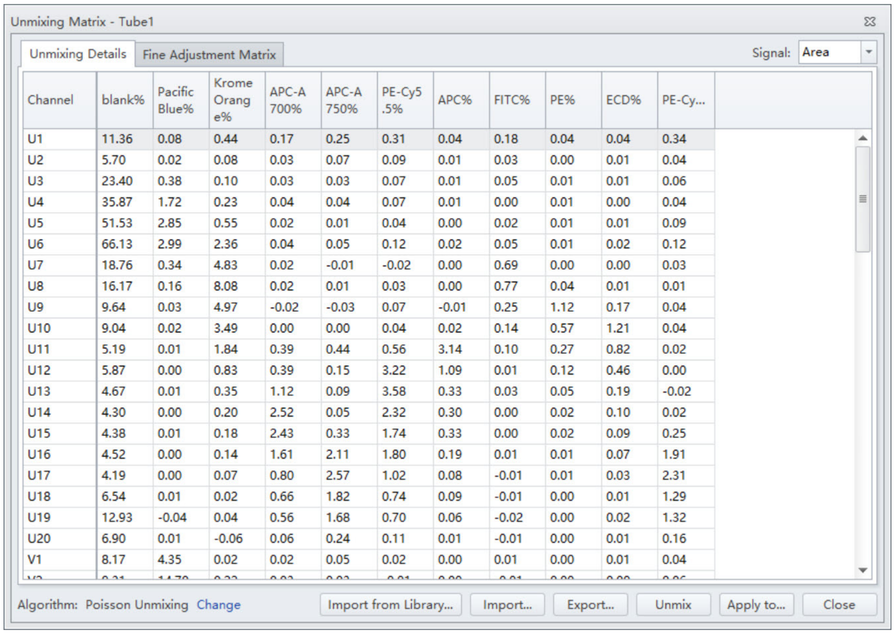
Algorithm: Poisson Unmixing Change

6 If necessary, select Apply to... to apply the unmixing values to the selected test tubes.

7 Select **Close**.

Exporting Unmixing Settings to Unmixing Matrix Files

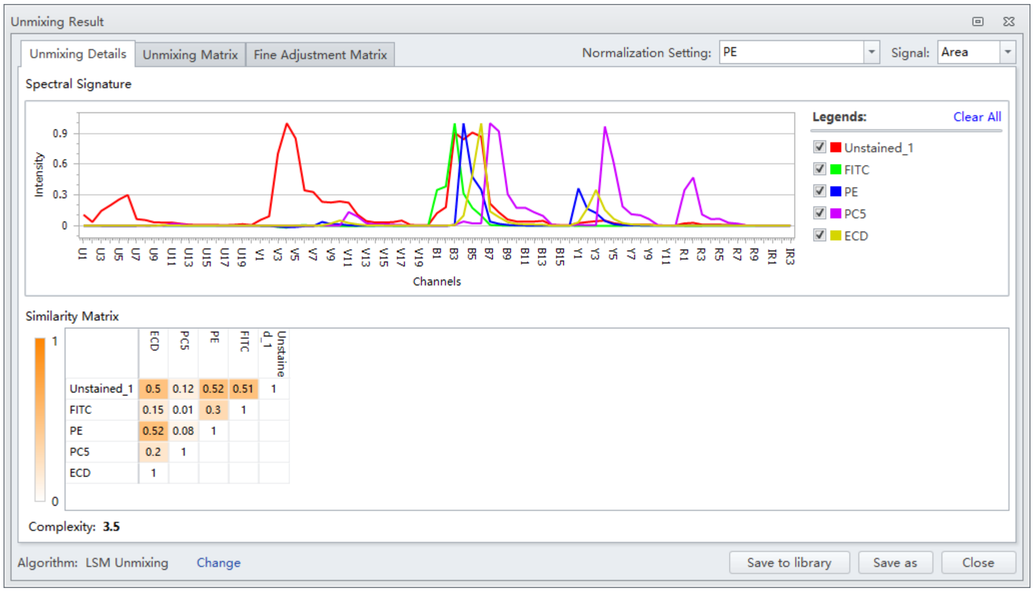
1 **[Standard Experiment]:** Select the desired tube that contains the unmixing matrix and select or select **Settings** > **Unmixing Matrix**. The Unmixing Matrix window appears.



Channel	blank%	Pacific Blue%	Krome Orange%	APC-A 700%	APC-A 750%	PE-Cy5.5%	APC%	FITC%	PE%	ECD%	PE-Cy...
U1	11.36	0.08	0.44	0.17	0.25	0.31	0.04	0.18	0.04	0.04	0.34
U2	5.70	0.02	0.08	0.03	0.07	0.09	0.01	0.03	0.00	0.01	0.04
U3	23.40	0.38	0.10	0.03	0.03	0.07	0.01	0.05	0.01	0.01	0.06
U4	35.87	1.72	0.23	0.04	0.04	0.07	0.01	0.00	0.01	0.00	0.04
U5	51.53	2.85	0.55	0.02	0.01	0.04	0.00	0.02	0.01	0.01	0.09
U6	66.13	2.99	2.36	0.04	0.05	0.12	0.02	0.05	0.01	0.02	0.12
U7	18.76	0.34	4.83	0.02	-0.01	-0.02	0.00	0.69	0.00	0.00	0.03
U8	16.17	0.16	8.08	0.02	0.01	0.03	0.00	0.77	0.04	0.01	0.01
U9	9.64	0.03	4.97	-0.02	-0.03	0.07	-0.01	0.25	1.12	0.17	0.04
U10	9.04	0.02	3.49	0.00	0.00	0.04	0.02	0.14	0.57	1.21	0.04
U11	5.19	0.01	1.84	0.39	0.44	0.56	3.14	0.10	0.27	0.82	0.02
U12	5.87	0.00	0.83	0.39	0.15	3.22	1.09	0.01	0.12	0.46	0.00
U13	4.67	0.01	0.35	1.12	0.09	3.58	0.33	0.03	0.05	0.19	-0.02
U14	4.30	0.00	0.20	2.52	0.05	2.32	0.30	0.00	0.02	0.10	0.02
U15	4.38	0.01	0.18	2.43	0.33	1.74	0.33	0.00	0.02	0.09	0.25
U16	4.52	0.00	0.14	1.61	2.11	1.80	0.19	0.01	0.01	0.07	1.91
U17	4.19	0.00	0.07	0.80	2.57	1.02	0.08	-0.01	0.01	0.03	2.31
U18	6.54	0.01	0.02	0.66	1.82	0.74	0.09	-0.01	0.00	0.01	1.29
U19	12.93	-0.04	0.04	0.56	1.68	0.70	0.06	-0.02	0.00	0.02	1.32
U20	6.90	0.01	-0.06	0.06	0.24	0.11	0.01	-0.01	0.00	0.01	0.16
V1	8.17	4.35	0.02	0.02	0.05	0.02	0.00	0.01	0.00	0.01	0.04

Algorithm: Poisson Unmixing Change

[Spectral Unmixing Experiment]: Select the desired tube and select or select **Settings** > **Unmixing Matrix**. The Unmixing Result window appears.



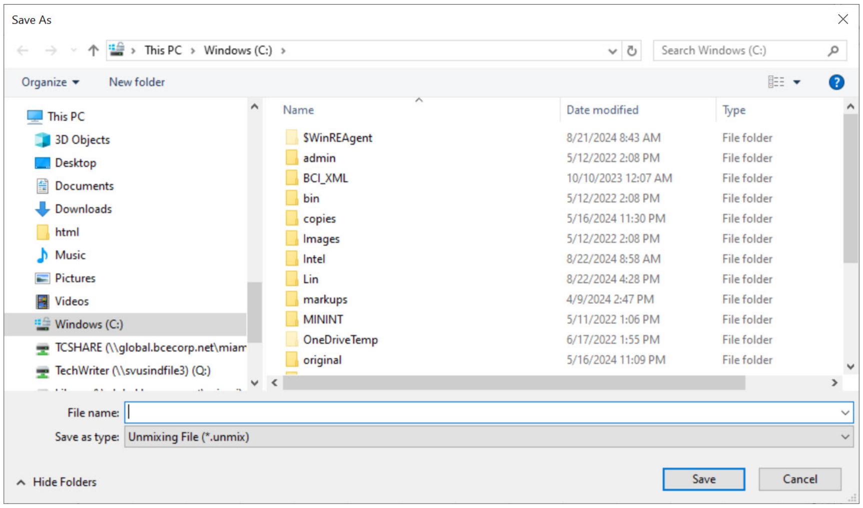
	ECD	PC5	PE	FITC	Unstained_1
Unstained_1	0.5	0.12	0.52	0.51	1
FITC	0.15	0.01	0.3	1	
PE	0.52	0.08	1		
PC5	0.2	1			
ECD	1				

Complexity: **3.5**

Algorithm: LSM Unmixing Change

2 **[Standard Experiment]:** Select **Export**. Specify a path and filename for the unmixing file you are saving and select **Save**.

[Spectral Unmixing Experiment]: Select **Save as**. Specify a path and filename for the unmixing file you are saving and select **Save**.



Name	Date modified	Type
$WinREAgent	8/21/2024 8:43 AM	File folder
admin	5/12/2022 2:08 PM	File folder
BCI_XML	10/10/2023 12:07 AM	File folder
bin	5/12/2022 2:08 PM	File folder
copies	5/16/2024 11:30 PM	File folder
Images	5/12/2022 2:08 PM	File folder
Intel	8/22/2024 8:58 AM	File folder
Lin	8/22/2024 4:28 PM	File folder
markups	4/9/2024 2:47 PM	File folder
MININT	5/11/2022 1:06 PM	File folder
OneDriveTemp	6/17/2022 1:55 PM	File folder
original	5/16/2024 11:09 PM	File folder

File name:
Save as type: Unmixing File (*.unmix)

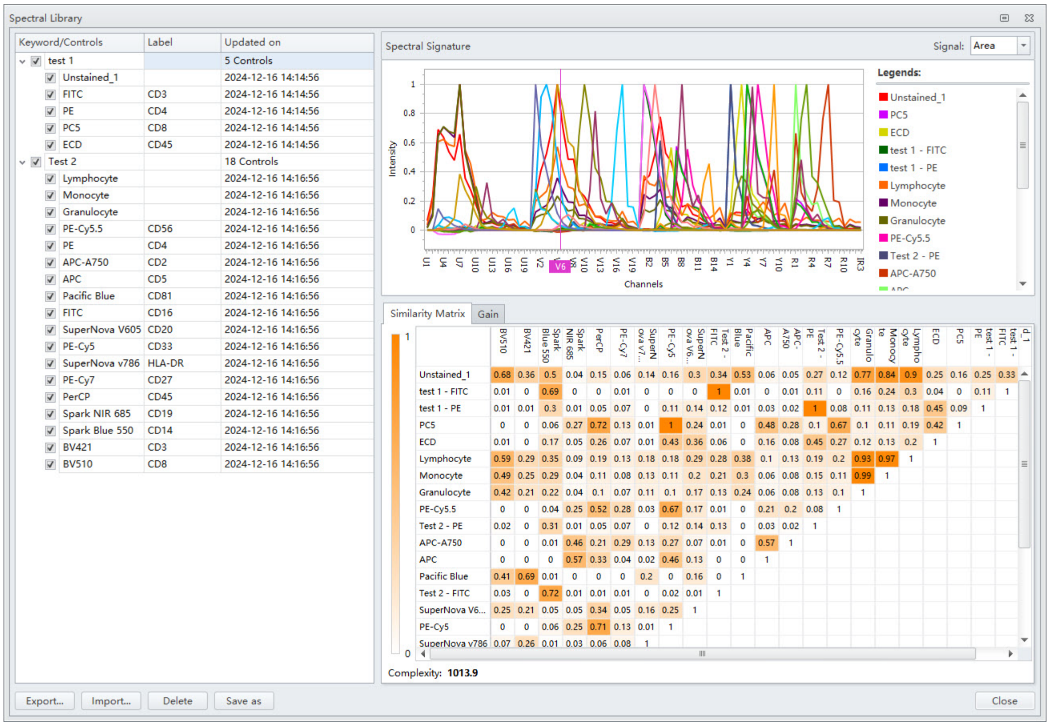
NOTE The generated file ends in .unmix.

3 Select **Close**.

Exporting Unmixing Settings to Unmixing Matrix Files from the Spectral Library

---

1 Select **Settings > Spectral Library**. The Spectral Library window appears.



Keyword/Controls	Label	Updated on
☑ test 1		5 Controls
☑ Unstained_1		2024-12-16 14:14:56
☑ FITC	CD3	2024-12-16 14:14:56
☑ PE	CD4	2024-12-16 14:14:56
☑ PC5	CD8	2024-12-16 14:14:56
☑ ECD	CD45	2024-12-16 14:14:56
☑ Test 2		18 Controls
☑ Lymphocyte		2024-12-16 14:16:56
☑ Monocyte		2024-12-16 14:16:56
☑ Granulocyte		2024-12-16 14:16:56
☑ PE-Cy5.5	CD56	2024-12-16 14:16:56
☑ PE	CD4	2024-12-16 14:16:56
☑ APC-A750	CD2	2024-12-16 14:16:56
☑ APC	CD5	2024-12-16 14:16:56
☑ Pacific Blue	CD81	2024-12-16 14:16:56
☑ FITC	CD16	2024-12-16 14:16:56
☑ SuperNova V605	CD20	2024-12-16 14:16:56
☑ PE-Cy5	CD33	2024-12-16 14:16:56
☑ SuperNova v786	HLA-DR	2024-12-16 14:16:56
☑ PE-Cy7	CD27	2024-12-16 14:16:56
☑ PerCP	CD45	2024-12-16 14:16:56
☑ Spark NIR 685	CD19	2024-12-16 14:16:56
☑ Spark Blue 550	CD14	2024-12-16 14:16:56
☑ BV421	CD3	2024-12-16 14:16:56
☑ BV510	CD8	2024-12-16 14:16:56

Signal: Area

	BV510	BV421	Spark Blue 550	Spark NIR 685	PerCP	PE-Cy7	SuperNova v7...	PE-Cy5	SuperNova V6...	Test 2 - FITC	Pacific Blue	APC	APC-A750	Test 2 - PE	PE-Cy5.5	Granulocyte	Monocyte	Lymphocyte	ECD	PC5	test 1 - PE	test 1 - FITC	d_1
Unstained_1	0.68	0.36	0.5	0.04	0.15	0.06	0.14	0.16	0.3	0.34	0.53	0.06	0.05	0.27	0.12	0.77	0.84	0.9	0.25	0.16	0.25	0.33	
test 1 - FITC	0.01	0	0.69	0	0	0.01	0	0	0	1	0.01	0	0.01	0.11	0	0.16	0.24	0.3	0.04	0	0.11	1	
test 1 - PE	0.01	0.01	0.3	0.01	0.05	0.07	0	0.11	0.14	0.12	0.01	0.03	0.02	1	0.08	0.11	0.13	0.18	0.45	0.09	1		
PC5	0	0	0.06	0.27	0.72	0.13	0.01	1	0.24	0.01	0	0.48	0.28	0.1	0.67	0.1	0.11	0.19	0.42	1			
ECD	0.01	0	0.17	0.05	0.26	0.07	0.01	0.43	0.36	0.06	0	0.16	0.08	0.45	0.27	0.12	0.13	0.2	1				
Lymphocyte	0.59	0.29	0.35	0.09	0.19	0.13	0.18	0.18	0.29	0.28	0.38	0.1	0.13	0.19	0.2	0.93	0.97	1					
Monocyte	0.49	0.25	0.29	0.04	0.11	0.08	0.13	0.11	0.2	0.21	0.3	0.06	0.08	0.15	0.11	0.99	1						
Granulocyte	0.42	0.21	0.22	0.04	0.1	0.07	0.11	0.1	0.17	0.13	0.24	0.06	0.08	0.13	0.1	1							
PE-Cy5.5	0	0	0.04	0.25	0.52	0.28	0.03	0.67	0.17	0.01	0	0.21	0.2	0.08	1								
Test 2 - PE	0.02	0	0.31	0.01	0.05	0.07	0	0.12	0.14	0.13	0	0.03	0.02	1									
APC-A750	0	0	0.01	0.46	0.21	0.29	0.13	0.27	0.07	0.01	0	0.57	1										
APC	0	0	0	0.57	0.33	0.04	0.02	0.46	0.13	0	0	1											
Pacific Blue	0.41	0.69	0.01	0	0	0	0.2	0	0.16	0	1												
Test 2 - FITC	0.03	0	0.72	0.01	0.01	0.01	0	0.02	0.01	1													
SuperNova V6...	0.25	0.21	0.05	0.05	0.34	0.05	0.16	0.25	1														
PE-Cy5	0	0	0.06	0.25	0.71	0.13	0.01	1															
SuperNova v786	0.07	0.26	0.01	0.03	0.06	0.08	1																

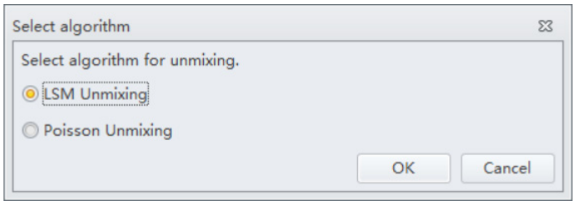
Complexity: **1013.9**

---

2 In the Keyword/Controls column, select the desired unmixing values to export.

---

3 Select **Save as**. The Select algorithm window appears.



Select algorithm for unmixing.

● LSM Unmixing

○ Poisson Unmixing

Select one of the following:

- LSM Unmixing
- Poisson Unmixing

---

4 Select **OK**.

5 Specify a path and filename for the unmixing matrix file you are saving, and select **Save**.

NOTE The generated file ends in *.unmix.

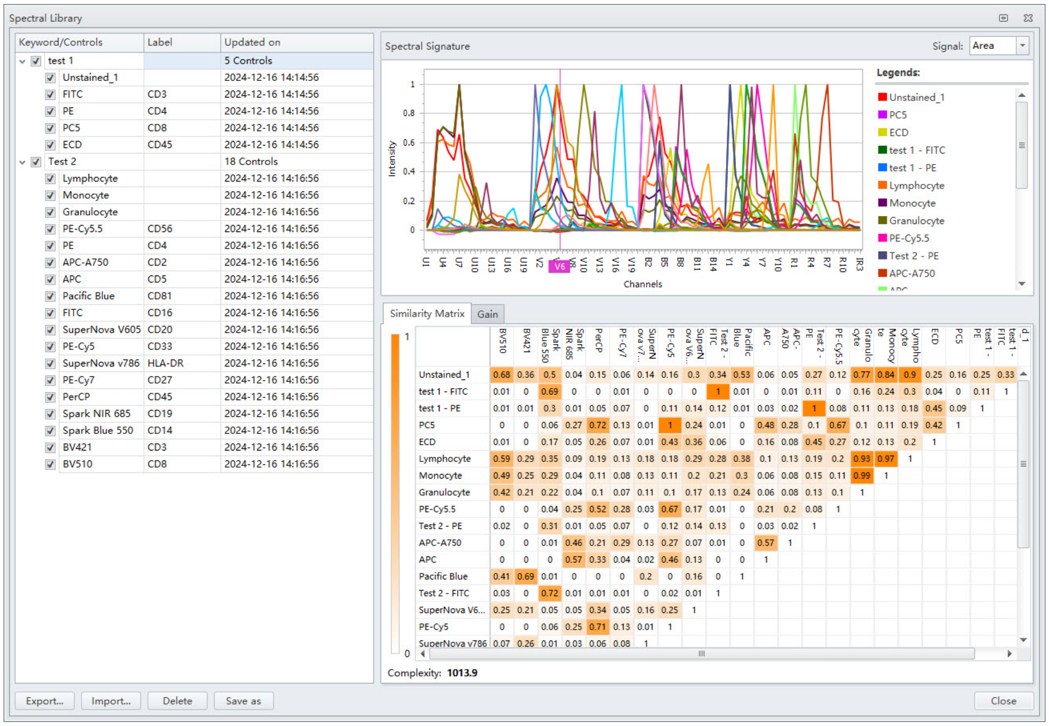
6 Select **Close**.

Managing Spectral Library

Unmixing values can be managed in the Spectral Library.

Importing Spectral Library Files

1 Select **Settings** > **Spectral Library**. The Spectral Library window appears.



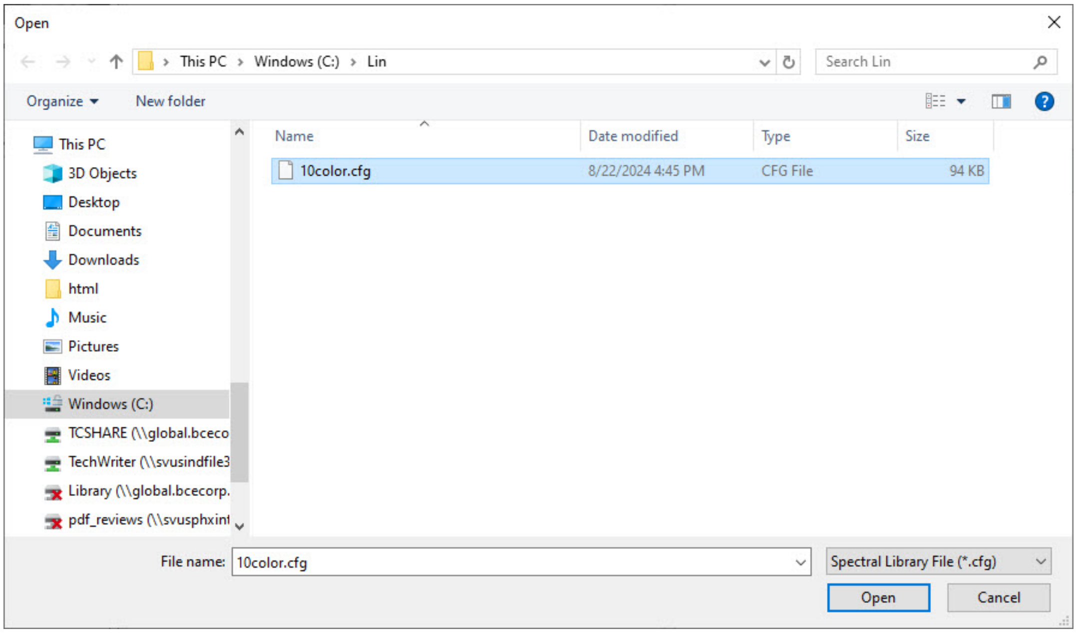
Keyword/Controls	Label	Updated on
☑ test 1		5 Controls
☑ Unstained_1		2024-12-16 14:14:56
☑ FITC	CD3	2024-12-16 14:14:56
☑ PE	CD4	2024-12-16 14:14:56
☑ PC5	CD8	2024-12-16 14:14:56
☑ ECD	CD45	2024-12-16 14:14:56
☑ Test 2		18 Controls
☑ Lymphocyte		2024-12-16 14:16:56
☑ Monocyte		2024-12-16 14:16:56
☑ Granulocyte		2024-12-16 14:16:56
☑ PE-Cy5.5	CD56	2024-12-16 14:16:56
☑ PE	CD4	2024-12-16 14:16:56
☑ APC-A750	CD2	2024-12-16 14:16:56
☑ APC	CD5	2024-12-16 14:16:56
☑ Pacific Blue	CD81	2024-12-16 14:16:56
☑ FITC	CD16	2024-12-16 14:16:56
☑ SuperNova V605	CD20	2024-12-16 14:16:56
☑ PE-Cy5	CD33	2024-12-16 14:16:56
☑ SuperNova v786	HLA-DR	2024-12-16 14:16:56
☑ PE-Cy7	CD27	2024-12-16 14:16:56
☑ PerCP	CD45	2024-12-16 14:16:56
☑ Spark NIR 685	CD19	2024-12-16 14:16:56
☑ Spark Blue 550	CD14	2024-12-16 14:16:56
☑ BV421	CD3	2024-12-16 14:16:56
☑ BV510	CD8	2024-12-16 14:16:56

Signal: Area

	BV510	BV421	Spark Blue 550	Spark NIR 685	PerCP	PE-Cy7	SuperNova v7...	PE-Cy5	SuperNova V6...	Test 2 - FITC	Pacific Blue	APC	APC-A750	Test 2 - PE	PE-Cy5.5	Granulocyte	Monocyte	Lymphocyte	ECD	PC5	test 1 - PE	test 1 - FITC	d_1
Unstained_1	0.68	0.36	0.5	0.04	0.15	0.06	0.14	0.16	0.3	0.34	0.53	0.06	0.05	0.27	0.12	0.77	0.84	0.9	0.25	0.16	0.25	0.33	
test 1 - FITC	0.01	0	0.69	0	0	0.01	0	0	0	1	0.01	0	0.01	0.11	0	0.16	0.24	0.3	0.04	0	0.11	1	
test 1 - PE	0.01	0.01	0.3	0.01	0.05	0.07	0	0.11	0.14	0.12	0.01	0.03	0.02	1	0.08	0.11	0.13	0.18	0.45	0.09	1		
PC5	0	0	0.06	0.27	0.72	0.13	0.01	1	0.24	0.01	0	0.48	0.28	0.1	0.67	0.1	0.11	0.19	0.42	1			
ECD	0.01	0	0.17	0.05	0.26	0.07	0.01	0.43	0.36	0.06	0	0.16	0.08	0.45	0.27	0.12	0.13	0.2	1				
Lymphocyte	0.59	0.29	0.35	0.09	0.19	0.13	0.18	0.18	0.29	0.28	0.38	0.1	0.13	0.19	0.2	0.93	0.97	1					
Monocyte	0.49	0.25	0.29	0.04	0.11	0.08	0.13	0.11	0.2	0.21	0.3	0.06	0.08	0.15	0.11	0.99	1						
Granulocyte	0.42	0.21	0.22	0.04	0.1	0.07	0.11	0.1	0.17	0.13	0.24	0.06	0.08	0.13	0.1	1							
PE-Cy5.5	0	0	0.04	0.25	0.52	0.28	0.03	0.67	0.17	0.01	0	0.21	0.2	0.08	1								
Test 2 - PE	0.02	0	0.31	0.01	0.05	0.07	0	0.12	0.14	0.13	0	0.03	0.02	1									
APC-A750	0	0	0.01	0.46	0.21	0.29	0.13	0.27	0.07	0.01	0	0.57	1										
APC	0	0	0	0.57	0.33	0.04	0.02	0.46	0.13	0	0	1											
Pacific Blue	0.41	0.69	0.01	0	0	0	0.2	0	0.16	0	1												
Test 2 - FITC	0.03	0	0.72	0.01	0.01	0.01	0	0.02	0.01	1													
SuperNova V6...	0.25	0.21	0.05	0.05	0.34	0.05	0.16	0.25	1														
PE-Cy5	0	0	0.06	0.25	0.71	0.13	0.01	1															
SuperNova v786	0.07	0.26	0.01	0.03	0.06	0.08	1																

Complexity: **1013.9**

2 Select **Import** and locate the path where spectral unmixing settings are saved. Select the corresponding spectral library file to import.



3 Select **Open** to import.

4 Select **Close**.

Exporting Spectral Library Files

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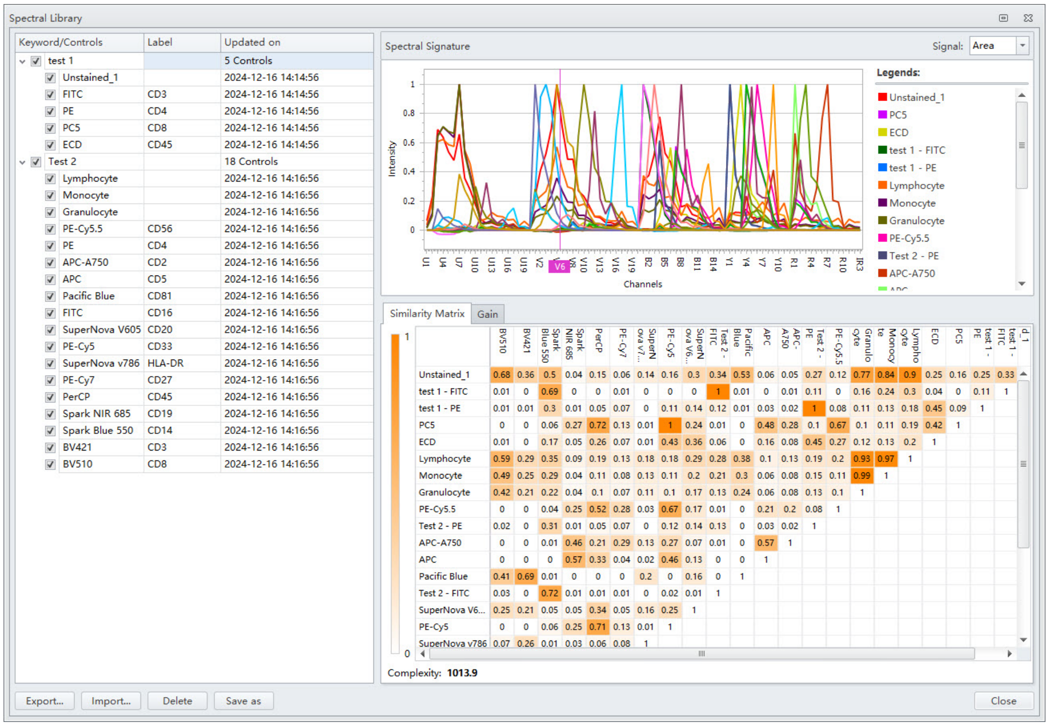
1 Select **Settings > Spectral Library.** The Spectral Library window appears.



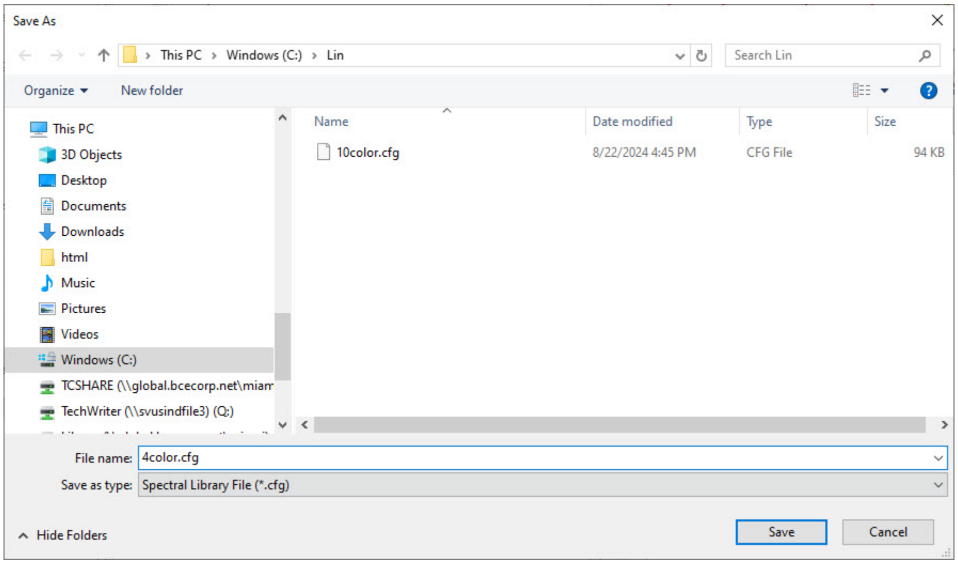
Keyword/Controls	Label	Updated on
☑ test 1		5 Controls
☑ Unstained_1		2024-12-16 14:14:56
☑ FITC	CD3	2024-12-16 14:14:56
☑ PE	CD4	2024-12-16 14:14:56
☑ PC5	CD8	2024-12-16 14:14:56
☑ ECD	CD45	2024-12-16 14:14:56
☑ Test 2		18 Controls
☑ Lymphocyte		2024-12-16 14:16:56
☑ Monocyte		2024-12-16 14:16:56
☑ Granulocyte		2024-12-16 14:16:56
☑ PE-Cy5.5	CD56	2024-12-16 14:16:56
☑ PE	CD4	2024-12-16 14:16:56
☑ APC-A750	CD2	2024-12-16 14:16:56
☑ APC	CD5	2024-12-16 14:16:56
☑ Pacific Blue	CD81	2024-12-16 14:16:56
☑ FITC	CD16	2024-12-16 14:16:56
☑ SuperNova V605	CD20	2024-12-16 14:16:56
☑ PE-Cy5	CD33	2024-12-16 14:16:56
☑ SuperNova v786	HLA-DR	2024-12-16 14:16:56
☑ PE-Cy7	CD27	2024-12-16 14:16:56
☑ PerCP	CD45	2024-12-16 14:16:56
☑ Spark NIR 685	CD19	2024-12-16 14:16:56
☑ Spark Blue 550	CD14	2024-12-16 14:16:56
☑ BV421	CD3	2024-12-16 14:16:56
☑ BV510	CD8	2024-12-16 14:16:56

	BV510	BV421	Spark Blue 550	Spark NIR 685	PerCP	PE-Cy7	SuperNova v7...	PE-Cy5	SuperNova V6...	Test 2 - FITC	Pacific Blue	APC	APC-A750	Test 2 - PE	PE-Cy5.5	Granulocyte	Monocyte	Lymphocyte	ECD	PC5	test 1 - PE	test 1 - FITC	d_1
Unstained_1	0.68	0.36	0.5	0.04	0.15	0.06	0.14	0.16	0.3	0.34	0.53	0.06	0.05	0.27	0.12	0.77	0.84	0.9	0.25	0.16	0.25	0.33	
test 1 - FITC	0.01	0	0.69	0	0	0.01	0	0	0	1	0.01	0	0.01	0.11	0	0.16	0.24	0.3	0.04	0	0.11	1	
test 1 - PE	0.01	0.01	0.3	0.01	0.05	0.07	0	0.11	0.14	0.12	0.01	0.03	0.02	1	0.08	0.11	0.13	0.18	0.45	0.09	1		
PC5	0	0	0.06	0.27	0.72	0.13	0.01	1	0.24	0.01	0	0.48	0.28	0.1	0.67	0.1	0.11	0.19	0.42	1			
ECD	0.01	0	0.17	0.05	0.26	0.07	0.01	0.43	0.36	0.06	0	0.16	0.08	0.45	0.27	0.12	0.13	0.2	1				
Lymphocyte	0.59	0.29	0.35	0.09	0.19	0.13	0.18	0.18	0.29	0.28	0.38	0.1	0.13	0.19	0.2	0.93	0.97	1					
Monocyte	0.49	0.25	0.29	0.04	0.11	0.08	0.13	0.11	0.2	0.21	0.3	0.06	0.08	0.15	0.11	0.99	1						
Granulocyte	0.42	0.21	0.22	0.04	0.1	0.07	0.11	0.1	0.17	0.13	0.24	0.06	0.08	0.13	0.1	1							
PE-Cy5.5	0	0	0.04	0.25	0.52	0.28	0.03	0.67	0.17	0.01	0	0.21	0.2	0.08	1								
Test 2 - PE	0.02	0	0.31	0.01	0.05	0.07	0	0.12	0.14	0.13	0	0.03	0.02	1									
APC-A750	0	0	0.01	0.46	0.21	0.29	0.13	0.27	0.07	0.01	0	0.57	1										
APC	0	0	0	0.57	0.33	0.04	0.02	0.46	0.13	0	0	1											
Pacific Blue	0.41	0.69	0.01	0	0	0	0.2	0	0.16	0	1												
Test 2 - FITC	0.03	0	0.72	0.01	0.01	0.01	0	0.02	0.01	1													
SuperNova V6...	0.25	0.21	0.05	0.05	0.34	0.05	0.16	0.25	1														
PE-Cy5	0	0	0.06	0.25	0.71	0.13	0.01	1															
SuperNova v786	0.07	0.26	0.01	0.03	0.06	0.08	1																

Complexity: **1013.9**

---

2 Select **Export** to specify a path and filename for the unmixing file you are saving.



Name	Date modified	Type	Size
10color.cfg	8/22/2024 4:45 PM	CFG File	94 KB

File name: 4color.cfg
Save as type: Spectral Library File (*.cfg)

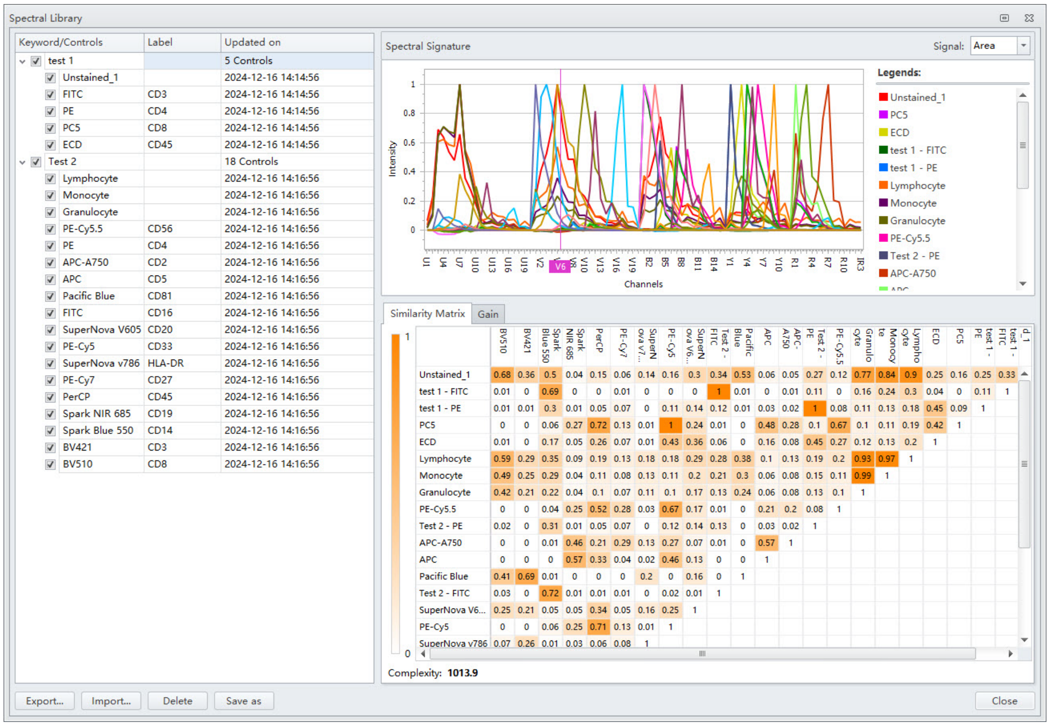
3 Select **Save.**

NOTE The generated file ends in .cfg.

4 Select **Close.**

Deleting Spectral Library Files

1 Select **Settings > Spectral Library**. The Spectral Library window appears.



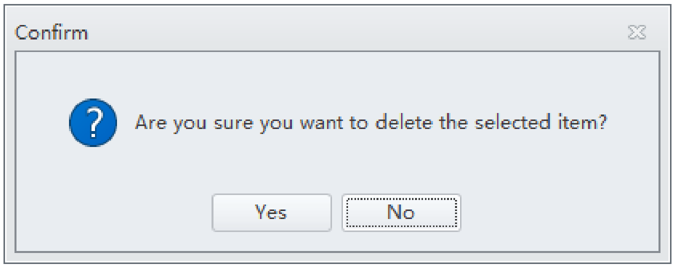
Keyword/Controls	Label	Updated on
☑ test 1		5 Controls
☑ Unstained_1		2024-12-16 14:14:56
☑ FITC	CD3	2024-12-16 14:14:56
☑ PE	CD4	2024-12-16 14:14:56
☑ PC5	CD8	2024-12-16 14:14:56
☑ ECD	CD45	2024-12-16 14:14:56
☑ Test 2		18 Controls
☑ Lymphocyte		2024-12-16 14:16:56
☑ Monocyte		2024-12-16 14:16:56
☑ Granulocyte		2024-12-16 14:16:56
☑ PE-Cy5.5	CD56	2024-12-16 14:16:56
☑ PE	CD4	2024-12-16 14:16:56
☑ APC-A750	CD2	2024-12-16 14:16:56
☑ APC	CD5	2024-12-16 14:16:56
☑ Pacific Blue	CD81	2024-12-16 14:16:56
☑ FITC	CD16	2024-12-16 14:16:56
☑ SuperNova V605	CD20	2024-12-16 14:16:56
☑ PE-Cy5	CD33	2024-12-16 14:16:56
☑ SuperNova v786	HLA-DR	2024-12-16 14:16:56
☑ PE-Cy7	CD27	2024-12-16 14:16:56
☑ PerCP	CD45	2024-12-16 14:16:56
☑ Spark NIR 685	CD19	2024-12-16 14:16:56
☑ Spark Blue 550	CD14	2024-12-16 14:16:56
☑ BV421	CD3	2024-12-16 14:16:56
☑ BV510	CD8	2024-12-16 14:16:56

	BV510	BV421	Spark Blue 550	Spark NIR 685	PerCP	PE-Cy7	SuperNova v7...	PE-Cy5	SuperNova V6...	Test 2 - FITC	Pacific Blue	APC	APC-A750	Test 2 - PE	PE-Cy5.5	Granulocyte	Monocyte	Lymphocyte	ECD	PC5	test 1 - PE	test 1 - FITC	d_1
Unstained_1	0.68	0.36	0.5	0.04	0.15	0.06	0.14	0.16	0.3	0.34	0.53	0.06	0.05	0.27	0.12	0.77	0.84	0.9	0.25	0.16	0.25	0.33	
test 1 - FITC	0.01	0	0.69	0	0	0.01	0	0	0	1	0.01	0	0.01	0.11	0	0.16	0.24	0.3	0.04	0	0.11	1	
test 1 - PE	0.01	0.01	0.3	0.01	0.05	0.07	0	0.11	0.14	0.12	0.01	0.03	0.02	1	0.08	0.11	0.13	0.18	0.45	0.09	1		
PC5	0	0	0.06	0.27	0.72	0.13	0.01	1	0.24	0.01	0	0.48	0.28	0.1	0.67	0.1	0.11	0.19	0.42	1			
ECD	0.01	0	0.17	0.05	0.26	0.07	0.01	0.43	0.36	0.06	0	0.16	0.08	0.45	0.27	0.12	0.13	0.2	1				
Lymphocyte	0.59	0.29	0.35	0.09	0.19	0.13	0.18	0.18	0.29	0.28	0.38	0.1	0.13	0.19	0.2	0.93	0.97	1					
Monocyte	0.49	0.25	0.29	0.04	0.11	0.08	0.13	0.11	0.2	0.21	0.3	0.06	0.08	0.15	0.11	0.99	1						
Granulocyte	0.42	0.21	0.22	0.04	0.1	0.07	0.11	0.1	0.17	0.13	0.24	0.06	0.08	0.13	0.1	1							
PE-Cy5.5	0	0	0.04	0.25	0.52	0.28	0.03	0.67	0.17	0.01	0	0.21	0.2	0.08	1								
Test 2 - PE	0.02	0	0.31	0.01	0.05	0.07	0	0.12	0.14	0.13	0	0.03	0.02	1									
APC-A750	0	0	0.01	0.46	0.21	0.29	0.13	0.27	0.07	0.01	0	0.57	1										
APC	0	0	0	0.57	0.33	0.04	0.02	0.46	0.13	0	0	1											
Pacific Blue	0.41	0.69	0.01	0	0	0	0.2	0	0.16	0	1												
Test 2 - FITC	0.03	0	0.72	0.01	0.01	0.01	0	0.02	0.01	1													
SuperNova V6...	0.25	0.21	0.05	0.05	0.34	0.05	0.16	0.25	1														
PE-Cy5	0	0	0.06	0.25	0.71	0.13	0.01	1															
SuperNova v786	0.07	0.26	0.01	0.03	0.06	0.08	1																

Complexity: **1013.9**

2 In the Keyword/Controls column, the corresponding unmixing values can be selected for each channel. The unmixing values of the same keyword/controls can also be selected using the drop-down menus in the Keyword/Controls column.

3 Select **Delete**. The following message appears:



Are you sure you want to delete the selected item?

4 Select **Yes**.

5 Select **Close**.

Fine Adjustment

The fine adjustment can be adjusted in an experiment in three ways: auto fine adjustment, manual fine adjustment, and fine adjustment matrix.

Auto Fine Adjusting

Select the populations that need to be adjusted in the bivariate plot. Select Ⓐ from the graphic control area to use auto fine adjustment.

NOTE Select **Refresh All Unmixing Gaps** ↻ to recalculate and refresh all unmixing gaps after using the fine adjustment.

NOTE The auto fine adjustment function is only available in the spectral unmixing experiment.

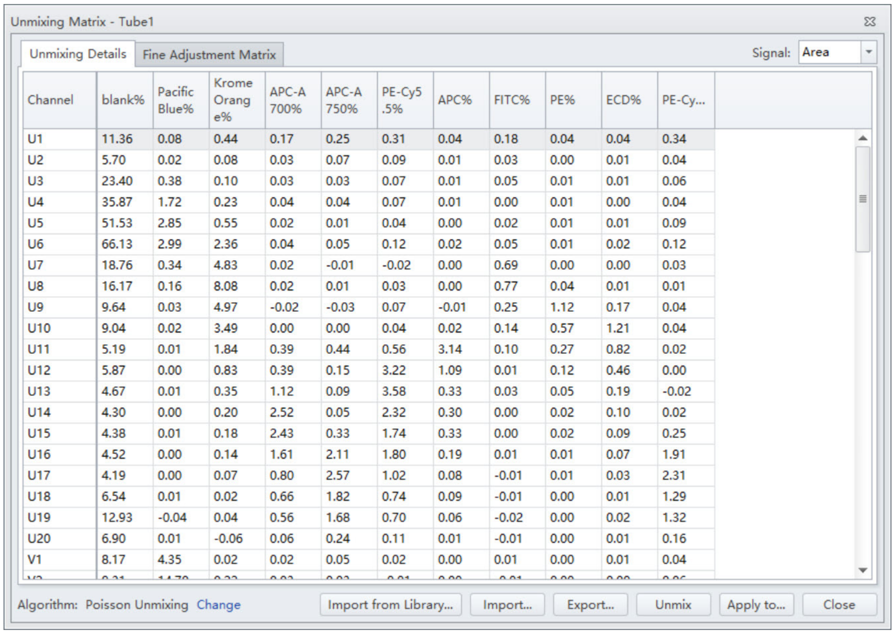
Manual Fine Adjustment

Select the populations that need to be adjusted in the bivariate plot. Select ✋ from the graphic control area, then click and drag the mouse pointer inside the plot to adjust. In a standard experiment, you can drag the mouse pointer vertically and/or horizontally. In a spectral unmixing experiment, you can only drag the mouse pointer vertically.

NOTE Select **Refresh All Unmixing Gaps** ↻ to recalculate and refresh all unmixing gaps after using the fine adjustment.

Fine Adjustment Matrix

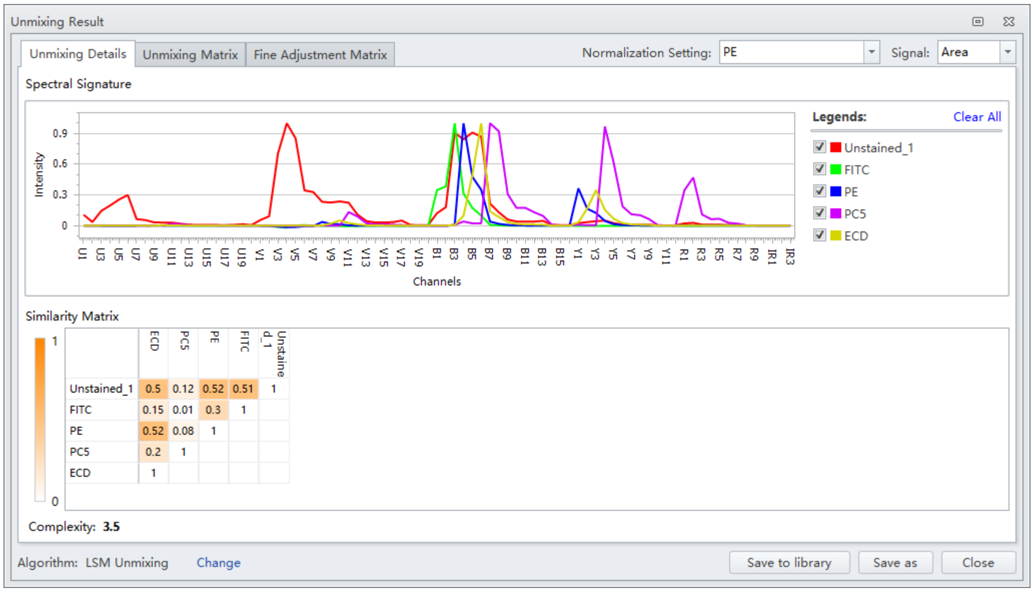
1 **[Standard Experiment]:** Select the desired tube that contains the unmixing matrix and select or select **Settings** > **Unmixing Matrix**. The Unmixing Matrix window appears.



Channel	blank%	Pacific Blue%	Krome Orange%	APC-A 700%	APC-A 750%	PE-Cy5 .5%	APC%	FITC%	PE%	ECD%	PE-Cy...
U1	11.36	0.08	0.44	0.17	0.25	0.31	0.04	0.18	0.04	0.04	0.34
U2	5.70	0.02	0.08	0.03	0.07	0.09	0.01	0.03	0.00	0.01	0.04
U3	23.40	0.38	0.10	0.03	0.03	0.07	0.01	0.05	0.01	0.01	0.06
U4	35.87	1.72	0.23	0.04	0.04	0.07	0.01	0.00	0.01	0.00	0.04
U5	51.53	2.85	0.55	0.02	0.01	0.04	0.00	0.02	0.01	0.01	0.09
U6	66.13	2.99	2.36	0.04	0.05	0.12	0.02	0.05	0.01	0.02	0.12
U7	18.76	0.34	4.83	0.02	-0.01	-0.02	0.00	0.69	0.00	0.00	0.03
U8	16.17	0.16	8.08	0.02	0.01	0.03	0.00	0.77	0.04	0.01	0.01
U9	9.64	0.03	4.97	-0.02	-0.03	0.07	-0.01	0.25	1.12	0.17	0.04
U10	9.04	0.02	3.49	0.00	0.00	0.04	0.02	0.14	0.57	1.21	0.04
U11	5.19	0.01	1.84	0.39	0.44	0.56	3.14	0.10	0.27	0.82	0.02
U12	5.87	0.00	0.83	0.39	0.15	3.22	1.09	0.01	0.12	0.46	0.00
U13	4.67	0.01	0.35	1.12	0.09	3.58	0.33	0.03	0.05	0.19	-0.02
U14	4.30	0.00	0.20	2.52	0.05	2.32	0.30	0.00	0.02	0.10	0.02
U15	4.38	0.01	0.18	2.43	0.33	1.74	0.33	0.00	0.02	0.09	0.25
U16	4.52	0.00	0.14	1.61	2.11	1.80	0.19	0.01	0.01	0.07	1.91
U17	4.19	0.00	0.07	0.80	2.57	1.02	0.08	-0.01	0.01	0.03	2.31
U18	6.54	0.01	0.02	0.66	1.82	0.74	0.09	-0.01	0.00	0.01	1.29
U19	12.93	-0.04	0.04	0.56	1.68	0.70	0.06	-0.02	0.00	0.02	1.32
U20	6.90	0.01	-0.06	0.06	0.24	0.11	0.01	-0.01	0.00	0.01	0.16
V1	8.17	4.35	0.02	0.02	0.05	0.02	0.00	0.01	0.00	0.01	0.04

Algorithm: Poisson Unmixing Change

[Spectral Unmixing Experiment]: Select the desired tube and select or select **Settings** > **Unmixing Matrix**. The Unmixing Result window appears.



Normalization Setting: PE

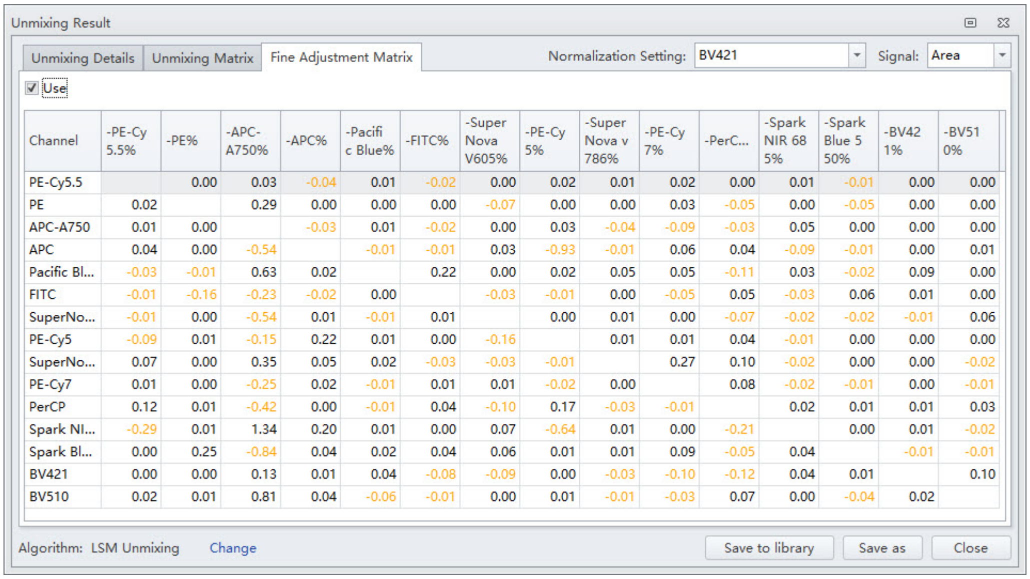
Signal: Area

	ECD	PC5	PE	FITC	Unstained_1
Unstained_1	0.5	0.12	0.52	0.51	1
FITC	0.15	0.01	0.3	1	
PE	0.52	0.08	1		
PC5	0.2	1			
ECD	1				

Complexity: 3.5

Algorithm: LSM Unmixing Change

2 Select the **Fine Adjustment Matrix** tab, and adjust the fine adjustment value as needed.



Normalization Setting: BV421
Signal: Area

☑ Use

Channel	-PE-Cy5.5%	-PE%	-APC-A750%	-APC%	-Pacific Blue%	-FITC%	-Super Nova V605%	-PE-Cy5%	-Super Nova v786%	-PE-Cy7%	-PerC...	-Spark NIR 685%	-Spark Blue 550%	-BV421%	-BV510%
PE-Cy5.5		0.00	0.03	-0.04	0.01	-0.02	0.00	0.02	0.01	0.02	0.00	0.01	-0.01	0.00	0.00
PE	0.02		0.29	0.00	0.00	0.00	-0.07	0.00	0.00	0.03	-0.05	0.00	-0.05	0.00	0.00
APC-A750	0.01	0.00		-0.03	0.01	-0.02	0.00	0.03	-0.04	-0.09	-0.03	0.05	0.00	0.00	0.00
APC	0.04	0.00	-0.54		-0.01	-0.01	0.03	-0.93	-0.01	0.06	0.04	-0.09	-0.01	0.00	0.01
Pacific Bl...	-0.03	-0.01	0.63	0.02		0.22	0.00	0.02	0.05	0.05	-0.11	0.03	-0.02	0.09	0.00
FITC	-0.01	-0.16	-0.23	-0.02	0.00		-0.03	-0.01	0.00	-0.05	0.05	-0.03	0.06	0.01	0.00
SuperNo...	-0.01	0.00	-0.54	0.01	-0.01	0.01		0.00	0.01	0.00	-0.07	-0.02	-0.02	-0.01	0.06
PE-Cy5	-0.09	0.01	-0.15	0.22	0.01	0.00	-0.16		0.01	0.01	0.04	-0.01	0.00	0.00	0.00
SuperNo...	0.07	0.00	0.35	0.05	0.02	-0.03	-0.03	-0.01		0.27	0.10	-0.02	0.00	0.00	-0.02
PE-Cy7	0.01	0.00	-0.25	0.02	-0.01	0.01	0.01	-0.02	0.00		0.08	-0.02	-0.01	0.00	-0.01
PerCP	0.12	0.01	-0.42	0.00	-0.01	0.04	-0.10	0.17	-0.03	-0.01		0.02	0.01	0.01	0.03
Spark NI...	-0.29	0.01	1.34	0.20	0.01	0.00	0.07	-0.64	0.01	0.00	-0.21		0.00	0.01	-0.02
Spark Bl...	0.00	0.25	-0.84	0.04	0.02	0.04	0.06	0.01	0.01	0.09	-0.05	0.04		-0.01	-0.01
BV421	0.00	0.00	0.13	0.01	0.04	-0.08	-0.09	0.00	-0.03	-0.10	-0.12	0.04	0.01		0.10
BV510	0.02	0.01	0.81	0.04	-0.06	-0.01	0.00	0.01	-0.01	-0.03	0.07	0.00	-0.04	0.02	

Algorithm: LSM Unmixing

3 Select **Close**.

4 Select **Refresh All Unmixing Gaps** to recalculate and refresh all unmixing gaps.

CHAPTER 8

Data Review

Overview

This chapter discusses how to use the Analysis screen to analyze data. Data can be analyzed using any computer equipped with the CytExpert for Spectral software. No online connection is required.

Workflow:

Import experiment or data	➔	Plot and configure statistics	➔	Export results

This chapter contains information on:

- Copying Experiments and Importing Data
- Setting the Plots and Statistics
- Calculating Sample Volume and Concentration
- Adjusting Unmixing Settings
- Exporting Results

Copying Experiments and Importing Data

Copying a Previously Acquired Experiment

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 8, Copying a Previously Acquired Experiment.

Importing Previously Acquired Data

The CytExpert for Spectral software can import and analyze compatible FCS data files acquired by other CytoFLEX flow cytometers.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 8, Importing Previously Acquired Data for detailed instructions.

---

1 Create a new experiment or open a saved experiment. Refer to CHAPTER 6, Creating an Experiment.

2. **[Standard Experiment]:** Select **File** > **Import FCS File,** specify the file path and select **Open**.

   Imported data files appear in the Tube screen.

   The Ⓕ symbol in front of each data tube indicates that the data tube is an imported data file. Imported data files are copied and saved in the folder where the current experiment data files are saved.

3. **[Spectral Unmixing Experiment]:** Right-click the desired tube and select **Import FCS Files** from the drop-down menu. Specify the file path and select **Open**.

   Imported data files appear in the Tube screen.

   The Ⓕ symbol in front of each data tube indicates that the data tube is an imported data file. Imported data files are copied and saved in the folder where the current experiment data files are saved.

Setting the Plots and Statistics

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 8, Setting the Plots and Statistics for detailed instructions on the following procedures:

- Opening the Analysis Screen
- Creating Histogram and Dot Plot Overlays

For configuring gates and generating statistics, refer to CHAPTER 6, Data Acquisition and Sample Analysis.

Calculating Sample Volume and Concentration

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 8, Calculating Sample Volume and Concentration for detailed instructions.

Adjusting Unmixing Settings

Data unmixing can be carried out at any time. You can select the desired tube in the tube list on the left side of the screen and select or select **Settings** > **Unmixing Matrix**. Refer to CHAPTER 7, Adjusting Spectral Unmixing for detailed instructions on adjusting unmixing settings.

Exporting Results

Refer to CHAPTER 6, Data Acquisition and Sample Analysis.

CHAPTER 9

Daily Shutdown

Overview

This chapter provides procedures for shutting down the CytoFLEX instrument equipped with the CytoFLEX mosaic Spectral Detection Module.

Workflow:

Prepare the cleaning solution	➔	Clean the instrument	➔	Turn the instrument off

This chapter contains information on:

- Preparing the Cleaning Solution
- Running Daily Clean
- Shutting Down the Instrument
- Auto Shutdown [CytoFLEX LX Only]

Preparing the Cleaning Solution

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 9, Preparing the Cleaning Solution for detailed instructions.

Running Daily Clean

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 9, Running Daily Clean for detailed instructions.

Shutting Down the Instrument

1. Shut down the Cytometer. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 9, Shutting Down the Instrument for detailed instructions on shutting down the Cytometer.

2. Long press the soft button on the front of the Cytometer mosaic Spectral Detection Module to turn off the CytoFLEX mosaic Spectral Detection Module.

3 **Optional:** Turn off the main power switch on the left of the Cytometer mosaic Spectral Detection Module.

Auto Shutdown [CytoFLEX LX Only]

You can set up the system to automatically shutdown the Cytometer.

To schedule an auto shutdown after acquisition, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 6, Creating an Experiment.

To schedule an auto shutdown during Daily Clean, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Daily Clean [With Plate Loader].

CHAPTER 10

Troubleshooting

Overview

IMPORTANT In addition to the information stated, never disassemble the instrument or have it repaired by unauthorized personnel. Beckman Coulter bears no responsibility for any problems arising from the unauthorized repair of the instrument.

This chapter introduces solutions to common problems. If there is a problem, follow the information in this chapter to carry out self-inspection. If the problem cannot be resolved, contact us.

This chapter contains information on:

Precautions/Hazards

Laser Related Hazards

Beckman Coulter design and manufacture of the instrument complies with the requirements governing the use and application of a laser specified in regulatory documents issued by the:

- U.S. Department of Health and Human Services
- Center for Devices and Radiological Health (CDRH)
- International Electrotechnical Commission (IEC)

In compliance with these regulatory documents, every measure has been taken to ensure the health and safety of users and laboratory personnel from the possible dangers of laser use.

Use the instrument according to the information in the manuals.

Use controls or adjustments or performance of procedures other than those specified herein might result in hazardous radiation exposure.

To ensure your safety, the Cytometer lasers are covered with protective shields. Do not remove these shields.

No user-serviceable assemblies are accessible. Do not attempt to remove the laser or open it. The instrument has components that are dangerous to the operator. If any attempt has been made to defeat a safety feature, or if the instrument fails to perform as described in its manuals, disconnect the power and contact us.

Laser Beam Hazards

The CytoFLEX Platform flow cytometer can be configured with up to 6 solid-state diode lasers that are capable of producing laser light at the following levels:

- 355 nm, 20 mW solid-state diode laser
- 375 nm, 60 mW solid-state diode laser
- 405 nm, 80 mW solid-state diode laser
- 488 nm, 50 mW solid-state diode laser
- 561 nm, 30 mW solid-state diode laser
- 638 nm, 50 mW solid-state diode laser
- 808 nm, 60 mW solid-state diode laser

A laser beam is a unique light source that shows characteristics different from conventional light sources. The safe use of the laser depends upon familiarity with the instrument and the properties of coherent, intense beams of light.

⚠ **WARNING**

Risk of personal injury. The laser beam can cause eye damage if viewed either directly or indirectly from reflective surfaces (such as a mirror or shiny metallic surfaces). To prevent eye damage, avoid direct exposure to the laser beam. Do not view it directly or with optical instruments.

Indirect contact with the laser beam from reflective surfaces (such as jewelry or a screwdriver) is called specular reflection and might also cause damage.

For these reasons, it is important to:

- Limit access to the Cytometer to trained and experienced personnel.
- Never attempt to remove a shield housing a laser and defeat interlocks.
- Never remove a warning label.
- Contact us if a label is missing or unclear.

Laser Warning Labels

⚠ **WARNING**

Risk of personal injury from radiation exposure. Never remove the shield surrounding a laser. Never remove covers and defeat interlocks.

CDRH-approved and IEC compliant labels are also placed near or on those covers that when removed might expose laser radiation. If necessary, a cover with a CDRH-approved or IEC compliant label must be removed by a qualified Beckman Coulter Representative only.

Refer to the following figures for the locations of the CDRH-approved and IEC compliant labels:

See Figure 10.1 for the Laser Warning Label on the Cytometer optical bench.

See Figure 10.2 and Figure 10.3 for the Laser Warning Label on the optical bench (located Inside the Cytometer).

See Figure 10.4 for the Laser Warning Label on the 355 nm Laser.

See Figure 10.5 and Figure 10.6 for the Laser Warning Labels on the Cytometer Back Cover.

See Figure 10.7 for the Laser Warning Label on the CytoFLEX mosaic Spectral Detection Module Back Cover.

The laser product is classified as CLASS 1 when all protective measures are in place. This product complies with 21 CFR Parts 1040.10 and 1040.11 as well as EN60825-1. See Figure 10.1.

Figure 10.1 Laser Warning Label on the Laser Optical Bench [**CytoFLEX LX**]

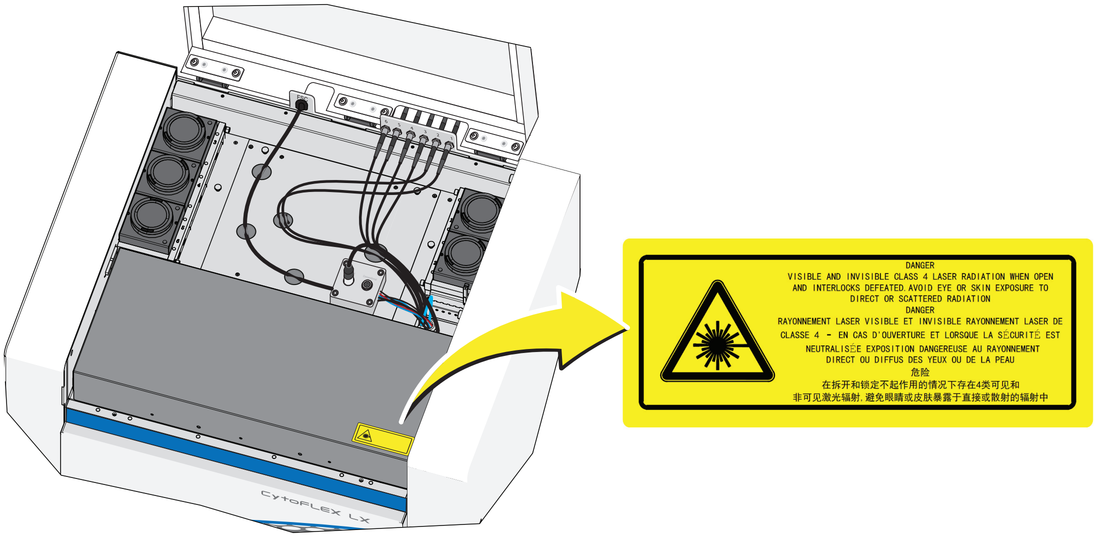

DANGER
VISIBLE AND INVISIBLE CLASS 4 LASER RADIATION WHEN OPEN AND INTERLOCKS DEFEATED. AVOID EYE OR SKIN EXPOSURE TO DIRECT OR SCATTERED RADIATION
DANGER
RAYONNEMENT LASER VISIBLE ET INVISIBLE RAYONNEMENT LASER DE CLASSE 4 - EN CAS D'OUVERTURE ET LORSQUE LA SÉCURITÉ EST NEUTRALISÉE EXPOSITION DANGEREUSE AU RAYONNEMENT DIRECT OU DIFFUS DES YEUX OU DE LA PEAU
危险
在拆开和锁定不起作用的情况下存在4类可见和非可见激光辐射,避免眼睛或皮肤暴露于直接或散射的辐射中
CytoFLEX LX

Figure 10.2 Laser Warning Label within the Optical Bench (Located Inside the Cytometer) [**CytoFLEX S**]

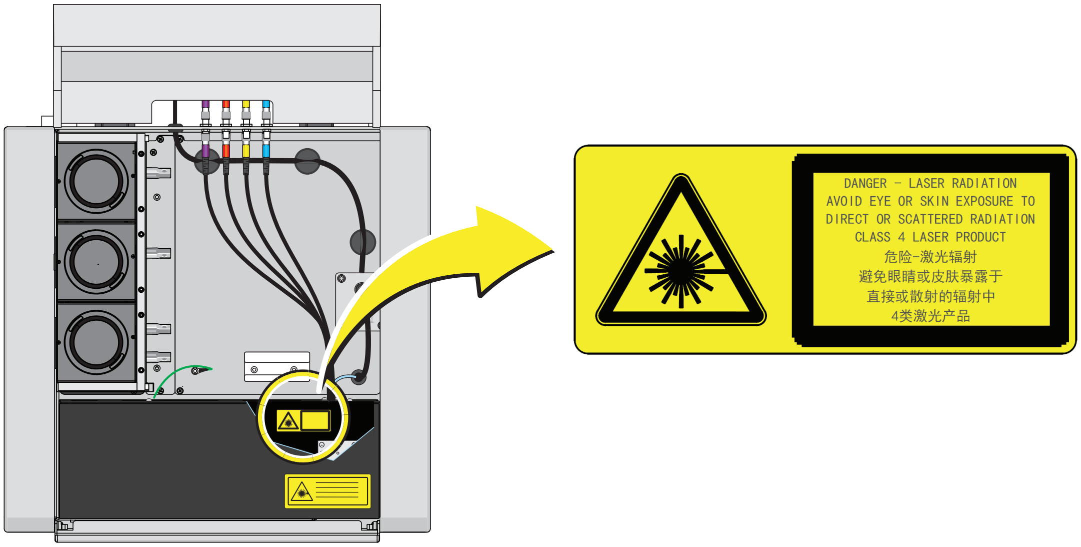
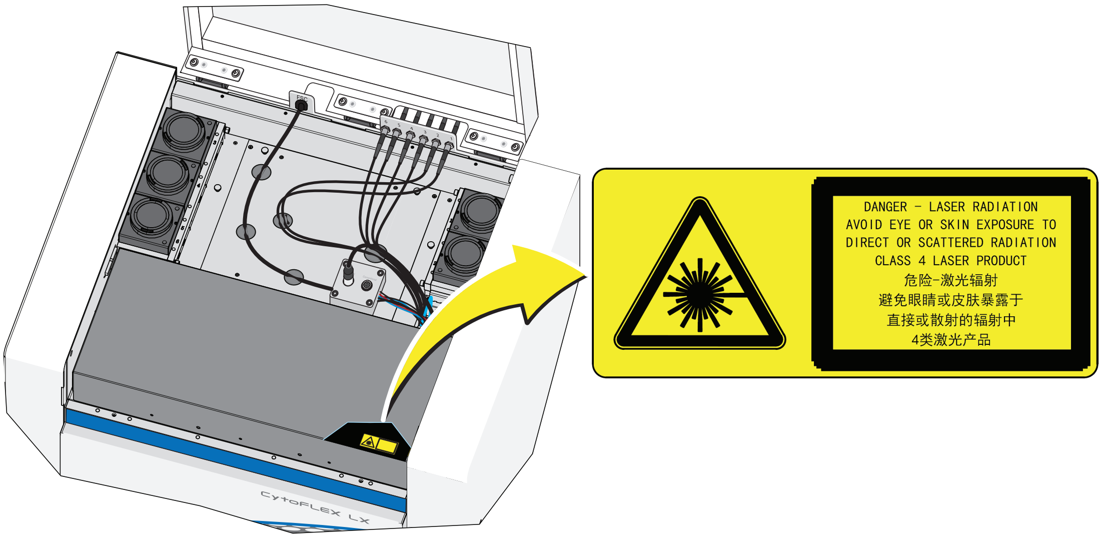
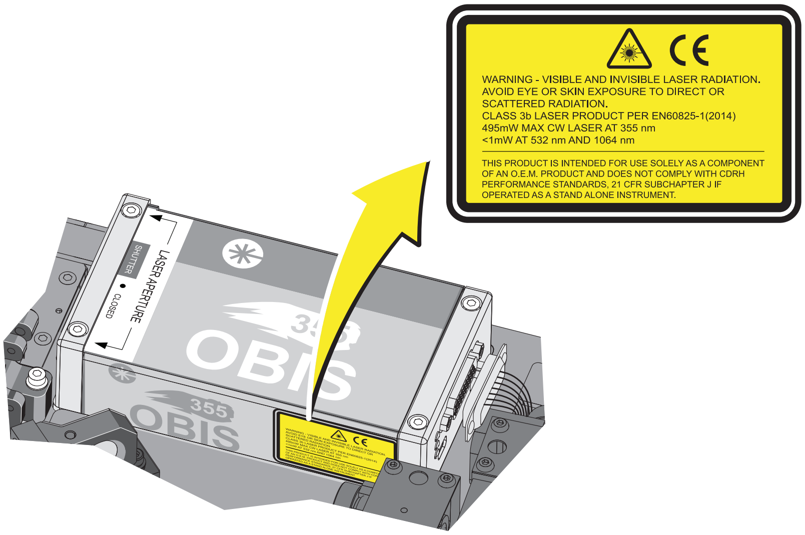


Figure 10.3 Laser Warning Label within the Optical Bench (Located Inside the Cytometer) [**CytoFLEX LX**]



DANGER – LASER RADIATION
AVOID EYE OR SKIN EXPOSURE TO
DIRECT OR SCATTERED RADIATION
CLASS 4 LASER PRODUCT
危险-激光辐射
避免眼睛或皮肤暴露于
直接或散射的辐射中
4类激光产品

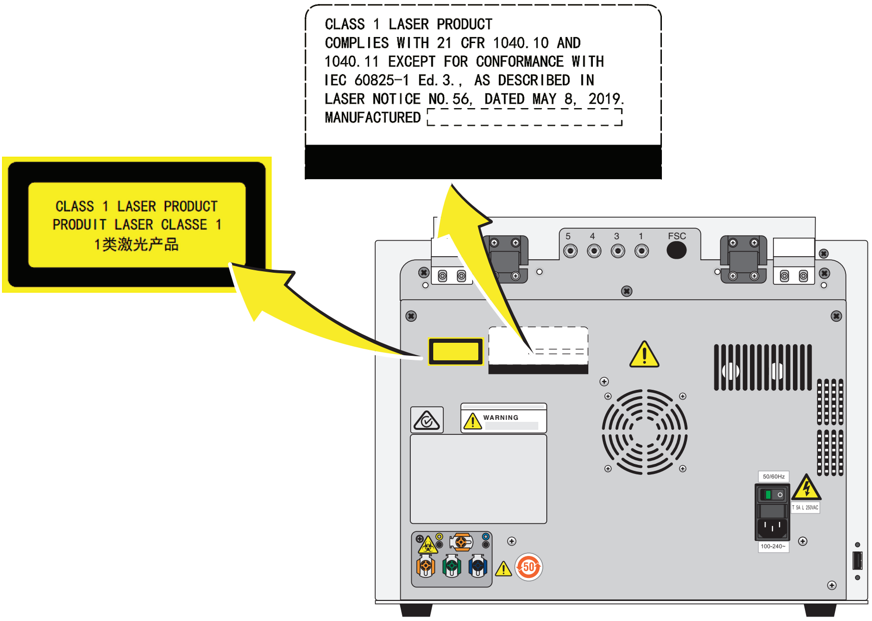
Figure 10.4 Laser Warning Label on the 355 nm Laser [**CytoFLEX LX**]



WARNING - VISIBLE AND INVISIBLE LASER RADIATION.
AVOID EYE OR SKIN EXPOSURE TO DIRECT OR
SCATTERED RADIATION.
CLASS 3b LASER PRODUCT PER EN60825-1(2014)
495mW MAX CW LASER AT 355 nm
<1mW AT 532 nm AND 1064 nm

THIS PRODUCT IS INTENDED FOR USE SOLELY AS A COMPONENT
OF AN O.E.M. PRODUCT AND DOES NOT COMPLY WITH CDRH
PERFORMANCE STANDARDS, 21 CFR SUBCHAPTER J IF
OPERATED AS A STAND ALONE INSTRUMENT.

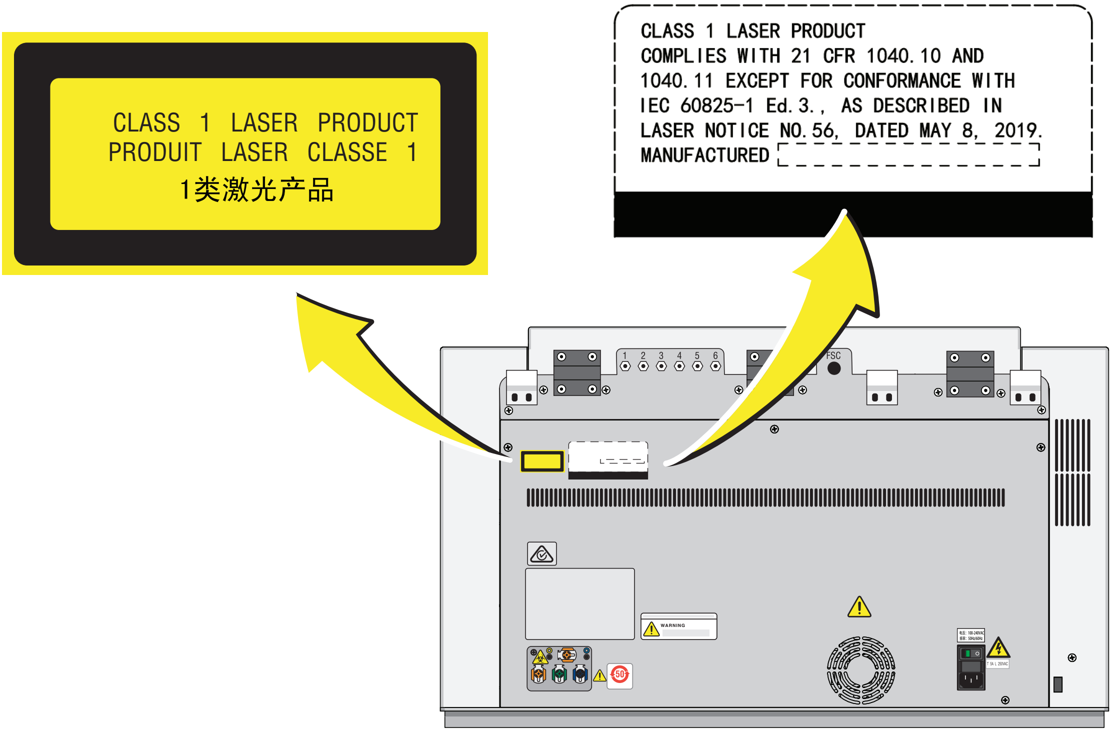
Figure 10.5 Laser Warning Labels on the Cytometer Back Cover [**CytoFLEX S**]



CLASS 1 LASER PRODUCT
COMPLIES WITH 21 CFR 1040.10 AND
1040.11 EXCEPT FOR CONFORMANCE WITH
IEC 60825-1 Ed.3., AS DESCRIBED IN
LASER NOTICE NO.56, DATED MAY 8, 2019.
MANUFACTURED

CLASS 1 LASER PRODUCT
PRODUIT LASER CLASSE 1
1类激光产品

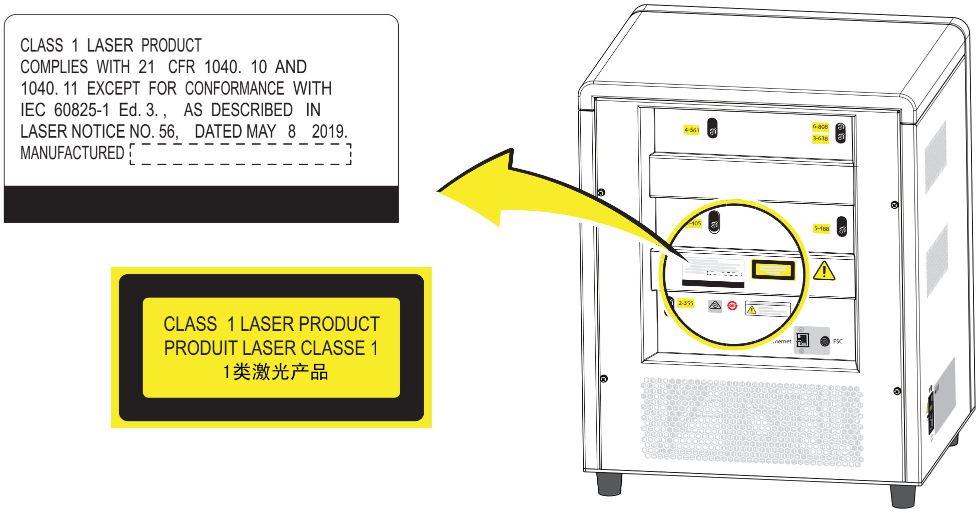
Figure 10.6 Laser Warning Labels on the Cytometer Back Cover [**CytoFLEX LX**]



CLASS 1 LASER PRODUCT
PRODUIT LASER CLASSE 1
1类激光产品

CLASS 1 LASER PRODUCT
COMPLIES WITH 21 CFR 1040.10 AND
1040.11 EXCEPT FOR CONFORMANCE WITH
IEC 60825-1 Ed.3., AS DESCRIBED IN
LASER NOTICE NO.56, DATED MAY 8, 2019.
MANUFACTURED

Figure 10.7 Laser Warning Labels on the CytoFLEX mosaic Spectral Detection Module Back Cover


CLASS 1 LASER PRODUCT
COMPLIES WITH 21 CFR 1040. 10 AND
1040. 11 EXCEPT FOR CONFORMANCE WITH
IEC 60825-1 Ed. 3. , AS DESCRIBED IN
LASER NOTICE NO. 56, DATED MAY 8 2019.
MANUFACTURED

CLASS 1 LASER PRODUCT
PRODUIT LASER CLASSE 1
1类激光产品

Disposal Precaution

WARNING

Risk of biohazardous contamination if you have skin contact with the waste container, its contents, and its associated tubing. The waste container and its associated tubing could contain residual biological material and must be handled with care. Clean up spills immediately. Dispose of the contents of the waste container in accordance with your local regulations and acceptable laboratory procedures.

Use universal precautions when working with pathogenic materials. Means must be available to decontaminate the instrument and to dispose of biohazardous waste.

Troubleshooting Table

Table 10.1 lists problems that you could encounter while running the CytoFLEX flow cytometer with the CytoFLEX mosaic Spectral Detection Module, the probable causes of each problem, and the corrective actions. These problems are listed alphabetically in the Index, under the primary entry “troubleshooting”.

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 10, Troubleshooting Table for detailed instructions on the Cytometer troubleshooting.

Table 10.1 Troubleshooting [With CytoFLEX mosaic Spectral Detection Module]

Problem	Probable Cause	Corrective Action
The CytoFLEX mosaic Spectral Detection Module cannot be turned on.	• The CytoFLEX mosaic Spectral Detection Module is turned off in the Cytometer menu. [CytoFLEX mosaic 88] • The power switch is in the off position and the Turn On selection will not function in the Cytometer menu. [CytoFLEX mosaic 88] • The power cable is not securely connected. • The fuse is blown.	**1.** Ensure the power switch is in the on position on the back of the CytoFLEX mosaic Spectral Detection Module. **2.** Select **Turn On** in the Cytometer menu. [CytoFLEX mosaic 88] **3.** Ensure that the power cable is securely connected to the back of the CytExpert mosaic Spectral Detection Module. **4.** If the problem persists, contact us.
The CytExpert for Spectral software is not responsive after the software has been launched.	The digital signature validation is timing out and failing due to a malfunction in the DNS server or firewall.	**1.** Type **Registry Editor** from the Windows search bar to locate the Register Editor. **2.** Select **Register Editor** to open the Register Editor window. **3.** Select **HKEY_CURRENT_USER** > **SOFTWARE** > **Microsoft** > **Windows** > **CurrentVersion** > **WinTrust** > **Trust Providers** > **Software Publishing**. **4.** Double click State to open the Edit DWORD Value window. **5.** Change the Value data to **23e00**. **6.** Select the **Hexadecimal** radio button. **7.** Select **OK**. **8.** Close the Registry Editor window. **NOTE** Problems resulting from incorrect Registry Editor settings may require the operating system reinstallation.
The connection indicator light in the lower right corner of the software screen is gray and displays *Disconnected*.	• Data connection error • FAPD data connection error • The CytoFLEX mosaic Spectral Detection Module is not turned on. • The CytoFLEX mosaic Spectral Detection Module's power cable is disconnected.	**1.** Ensure that the network cable is securely connected to the back of the Cytometer and the back of the Workstation. Refer to Figure 1.3 or Figure 1.6. **2.** Restart the software. Restart the Workstation. Refer to CHAPTER 4, Initializing the Instrument. **3.** Turn on the CytoFLEX mosaic Spectral Detection Module using the power switch on the back of the CytoFLEX mosaic Spectral Detection Module. **4.** Verify that the power cable is securely connected to the back of the CytoFLEX mosaic Spectral Detection Module. **5.** If the problem persists, contact us.

Backup and Restore

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 10, Backup and Restore.

Backup

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 10, Backup.

Restore

This software function is the same in both the CytExpert software and the CytExpert for Spectral software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 10, Restore.

Log Cleanup

Use Log Cleanup to delete Experiment Operation Logs and System Operation Logs before a selected date.

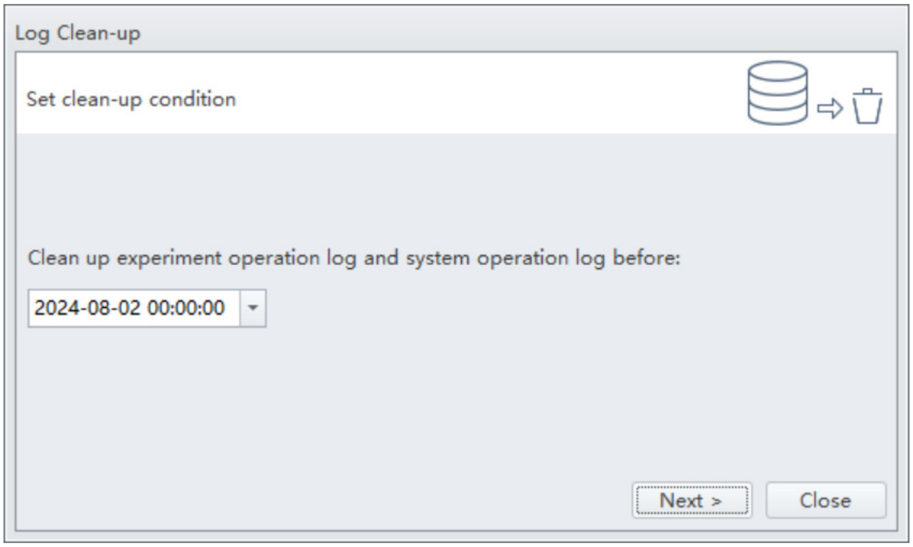
NOTE These procedures are only available if you have the Electronic Record Management software option installed.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 10, Log Cleanup.

1. Ensure all experiment files are closed.

---

2 Select **Backup/Restore > Log Clean-up.** The Log Clean-up window appears.



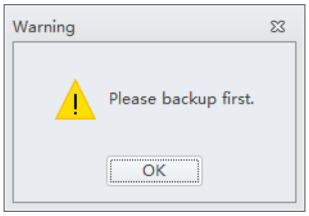
Log Clean-up

Set clean-up condition

Clean up experiment operation log and system operation log before:

2024-08-02 00:00:00

Backup the data and try again if the operation system prompts the following message:

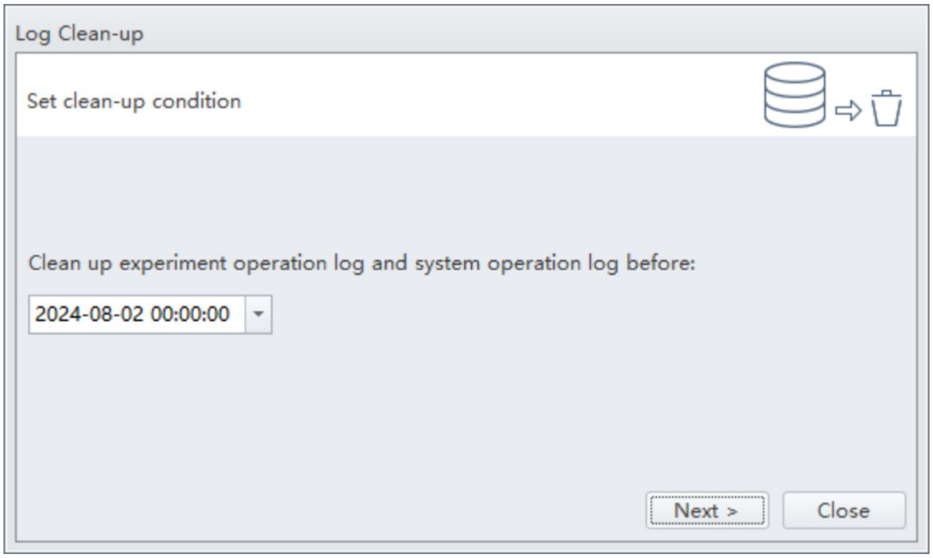


Warning

Please backup first.

---

3 Select the desired date and time.



Log Clean-up

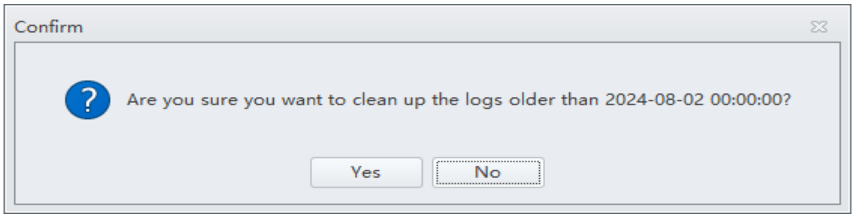
Set clean-up condition

Clean up experiment operation log and system operation log before:

2024-08-02 00:00:00

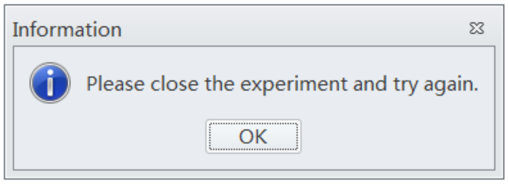
---

4 Select **Next.** The Confirm window appears.



Are you sure you want to clean up the logs older than 2024-08-02 00:00:00?

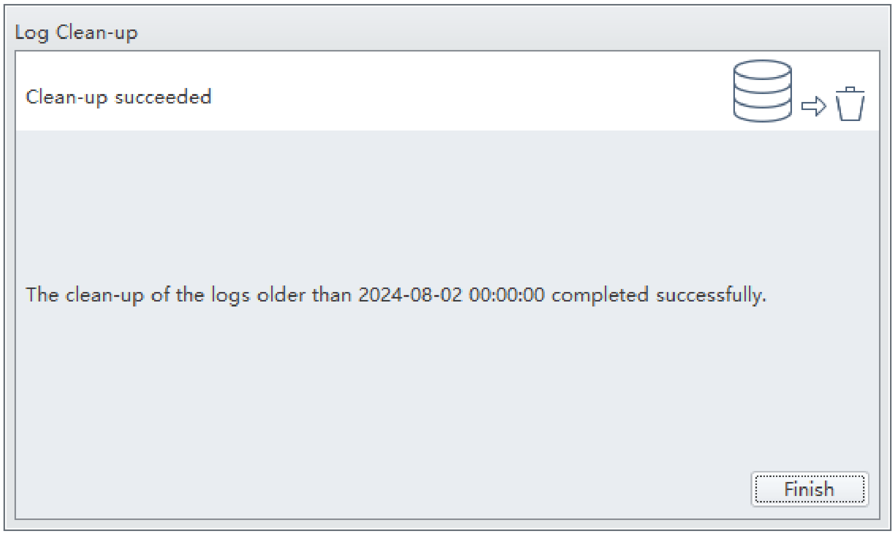
Close the experiment and try again if the operation system prompts the following message:



Please close the experiment and try again.

---

5 Select **Yes.** The Clean-up succeeded window appears.



Clean-up succeeded

The clean-up of the logs older than 2024-08-02 00:00:00 completed successfully.

---

6 Select **Finish.**

CHAPTER 11

Cleaning Procedures

Overview

This chapter describes how to carry out certain routine and nonscheduled cleaning procedures. Proper cleaning can help extend the service life of the instrument and ensure experimental accuracy. When conducting any cleaning, take all necessary biosafety precautions and use proper personal protective equipment.

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Cleaning Procedures for detailed instructions on the following procedures:

- Routine Cleaning
  - Daily Clean
  - Daily Clean [With Plate Loader]
  - Cleaning the Sample Station
  - Deep Clean Procedure
  - Cleaning the 4 L Sheath Fluid Container
  - Cleaning the 4 L Waste Container
- Nonscheduled Cleaning
  - Surface Cleaning and Disinfection
  - Preparing the Instrument for Transport or Storage

This chapter contains information on:

- Preparing the Instrument for Transport or Storage

Preparing the Instrument for Transport or Storage

When the instrument is to be transported or is not to be used for 30 days or more, complete the emptying processes to prevent instrument damage and to reduce the possibility of biological contamination. Contact us if you have any questions.

1 Prepare the CytoFLEX for transportation or storage. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Preparing the Instrument for Transport or Storage for detailed instructions.

2 Power down the CytoFLEX mosaic Spectral Detection Module and disconnect all the cables.

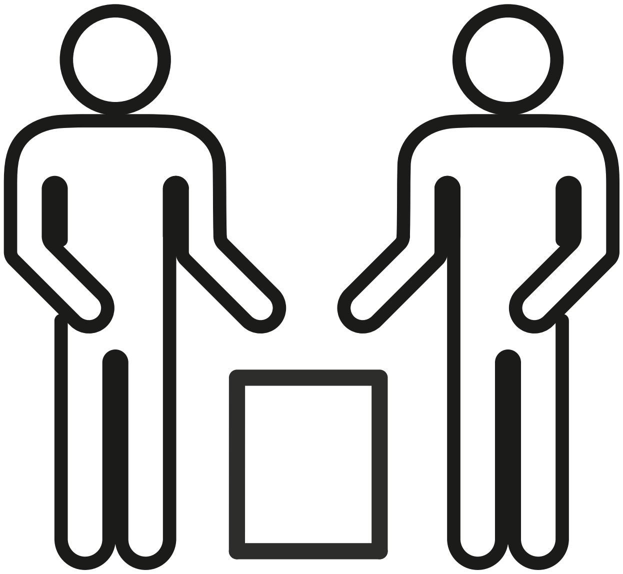
3 If the CytoFLEX mosaic Spectral Detection Module is to be transported or stored, put the instrument into the Beckman Coulter supplied packing and comply with the requirements in APPENDIX A, Instrument Transportation and Storage, regarding correct placement during transportation and storage.

Lifting and Carrying Instructions [CytoFLEX S and CytoFLEX LX]

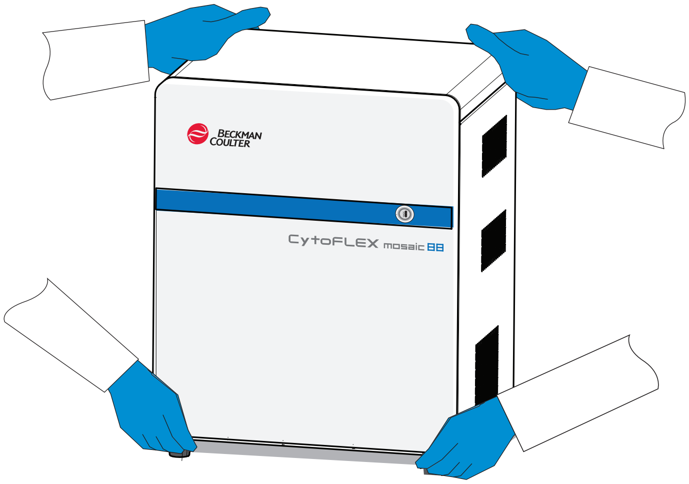
Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 11, Lifting and Carrying Instructions for detailed instructions.

Lifting and Carrying Instructions [CytoFLEX mosaic Spectral Detection Module]

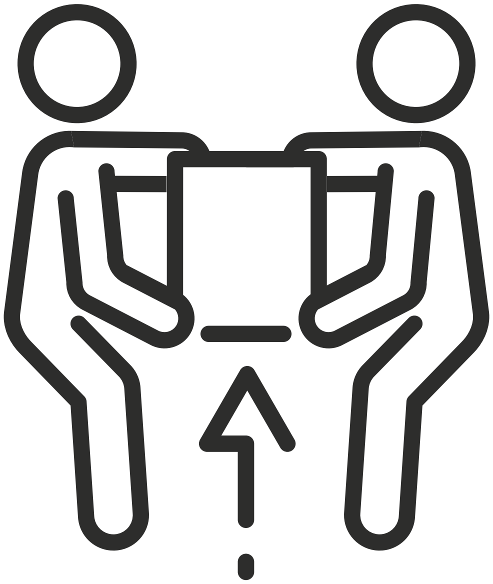
1 Position a person on the left and right sides of the CytoFLEX mosaic Spectral Detection Module.



2 Grasp the CytoFLEX mosaic Spectral Detection Module as shown in the figure below.



3 Gently lift the CytoFLEX mosaic Spectral Detection Module as shown in the figure below.



⚠ WARNING

Risk of personal injury. Use caution when lowering the CytoFLEX mosaic Spectral Detection Module to avoid pinching fingers.

4 Lower the CytoFLEX mosaic Spectral Detection Module to its designated location.

CHAPTER 12

Replacement/Adjustment Procedures

Overview

This chapter describes how to carry out certain routine and nonscheduled maintenance procedures. Proper maintenance can help extend the service life of the instrument and ensure experimental accuracy. When conducting any maintenance work, take all necessary biosafety precautions.

IMPORTANT In addition to parts specifically discussed, for all replacement parts, use only parts provided by Beckman Coulter to ensure proper functioning of the instrument. Never disassemble any part of the instrument without prior authorization. Beckman Coulter assumes no responsibility for any instrument problems resulting from the use of any part not authorized by Beckman Coulter for use with the instrument.

Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 12, Replacement/Adjustment Procedures for detailed instructions on the following procedures:

- Routine Replacement/Adjustment
  - Front Cover Removal and Reinstallation
  - Right-Side Cover Removal and Reinstallation
  - Filling the 4 L Sheath Fluid Container
  - Replacing the 10 L Sheath Fluid Cubitainer
  - Emptying the 4 L Waste Container
  - Emptying the 10 L Waste Cubitainer
  - Managing the Maintenance Reminder

    NOTE If you have previously installed the CytExpert software, the Maintenance data will automatically synchronize between the CytExpert Software and the CytExpert for Spectral Software.

  - Adding the Deep Clean Solution
  - Replacing the Sheath Fluid Filter
  - Unclogging the Sample Probe [With Sample Injection Mode Control Kit]
  - Unclogging the Sample Probe [Without Sample Injection Mode Control Kit]
  - Replacing the Sample Probe and/or the Sample Peristaltic Pump Tubing
  - Replacing the Sample Probe Assembly [With Plate Loader]
  - Changing the Sample Probe from the Single Tube Sample Station to the Plate Loader [CytoFLEX With Plate Loader]

  - Changing the Sample Probe from the Plate Loader to the Single Tube Sample Station [CytoFLEX With Plate Loader]
  - Inspecting the Liquid Flow Path for Leaks
  - Priming the Flow Cell
  - Replacing the Plate Holder [With Plate Loader]
  - Plate Loader Module Removal and Reinstallation [With Plate Loader]
  - Changing the Event Rate Setting

- Nonscheduled Replacement/Adjustment
  - Calibrating the Sample Flow Rate
  - Calibrating the Sample Flow Rate [With Plate Loader]
  - Setting Laser Delay
  - Replacing the Fuse
  - Replacing the Sheath Fluid Harness and/or Waste Harness
  - Changing Sample Mixing and Backflush Settings
  - Calibrating the Plate Position [With Plate Loader]

This chapter contains information on:

  - Switching between Spectral Mode and Conventional Mode

Switching between Spectral Mode and Conventional Mode

The CytoFLEX flow cytometer equipped with a CytoFLEX mosaic Spectral Detection Module supports two work modes: Conventional Mode and Spectral Mode.

- **Spectral Mode:** Attach the CytoFLEX mosaic Spectral Detection Module to the CytoFLEX flow cytometer to enable its spectral flow functionality and allow users to run more complex assays and use spectral unmixing technology for spectral overlap correction. Refer to Switching to Spectral Mode for detailed instructions on switching the instrument from Conventional Mode to Spectral Mode.
- **Conventional Mode:** Adjust how the optical fibers are connected to the Cytometer to use the Cytometer's optical components for collecting optical signals. Refer to Switching to Conventional Mode for detailed instructions on switching the instrument from Spectral Mode to Conventional Mode.

Table 12.1 Optical Fibers with Indicators **[CytoFLEX S]**

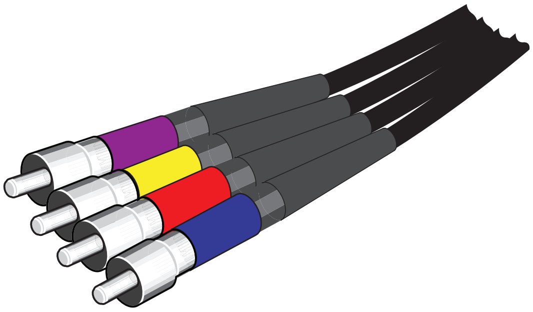
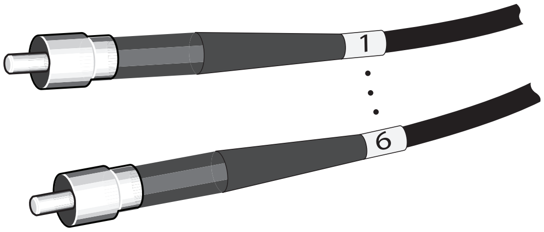
Optical Fibers	Indicator	Name
	Violet	Violet fiber
	Yellow	Yellow fiber
	Red	Red fiber
	Blue	Blue fiber

Table 12.2 Optical Fibers with Indicators **[CytoFLEX LX]**

Optical Fibers	Indicator	Name
	1	Violet fiber
	2	UV/NUV fiber
	3	Red fiber
	4	Yellow fiber
	5	Blue fiber
	6	IR fiber

Switching to Conventional Mode

⚠ **CAUTION**

Risk of data integrity damage.

- **Verify that the optical fibers are securely connected to the WDM. A loose connection can alter the optical path and affect fluorescence.**
- **Do not kink the optical fibers.**

---

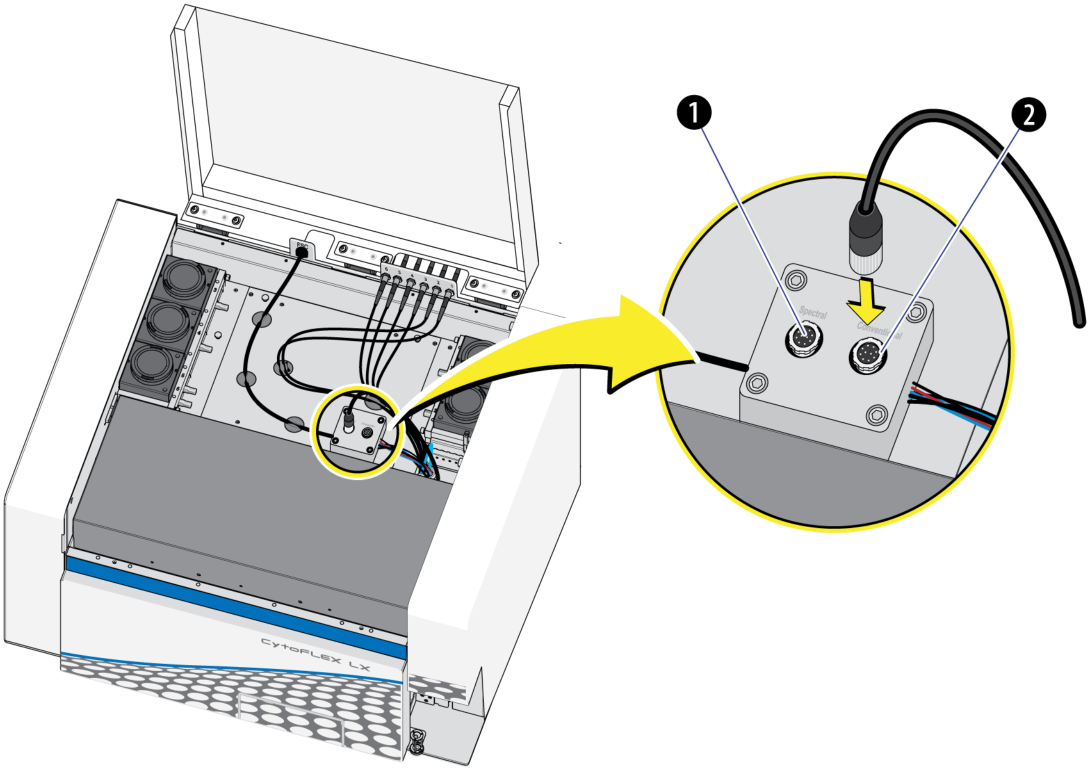
1 Confirm that the instrument is in the standby state or that the instrument and CytoFLEX mosaic Spectral Detection Module are turned off.

---

2 Open the top cover.

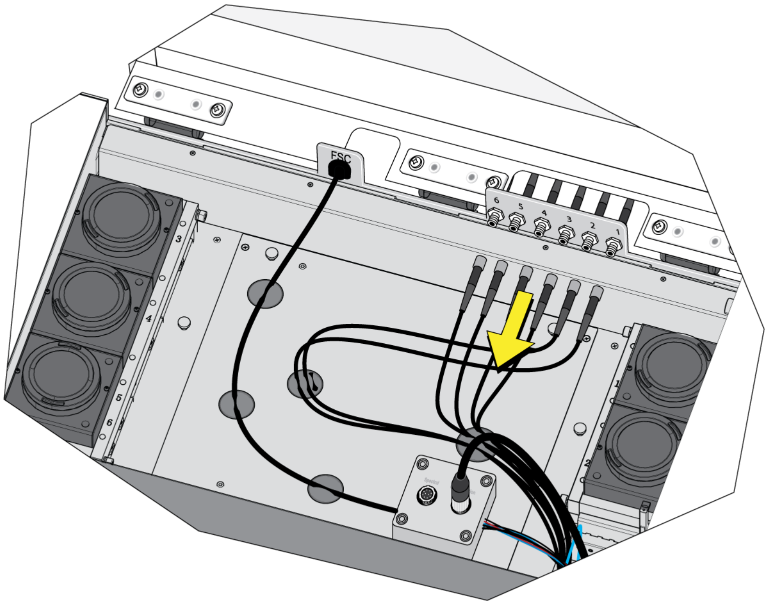
3. Locate the Forward Scatter Switch and move the FSC connector from the Spectral Port **(1)** to the Conventional Port **(2)**.

[CytoFLEX LX Shown]

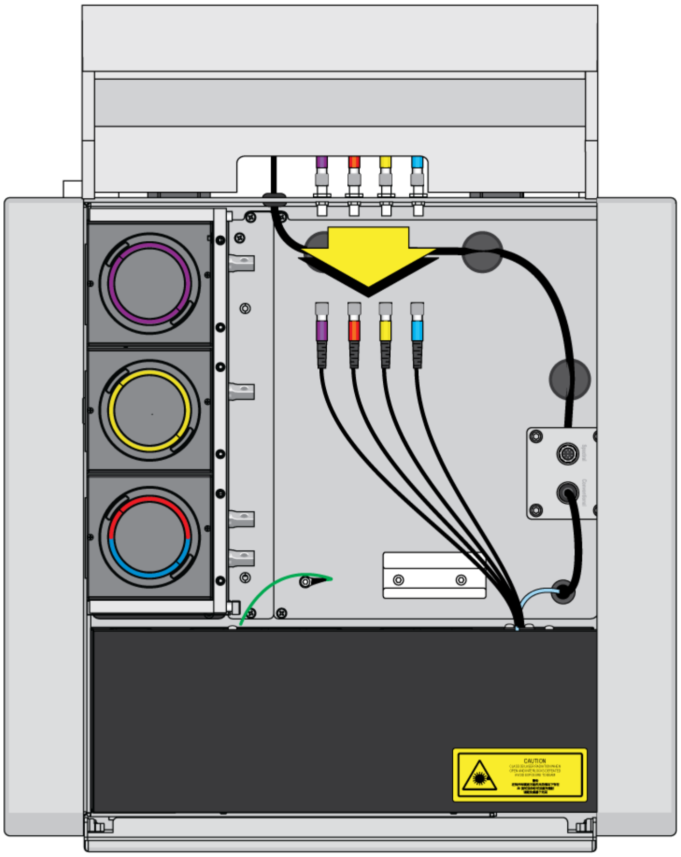


4 Disconnect the rest of the optical fibers.

[CytoFLEX LX Shown]

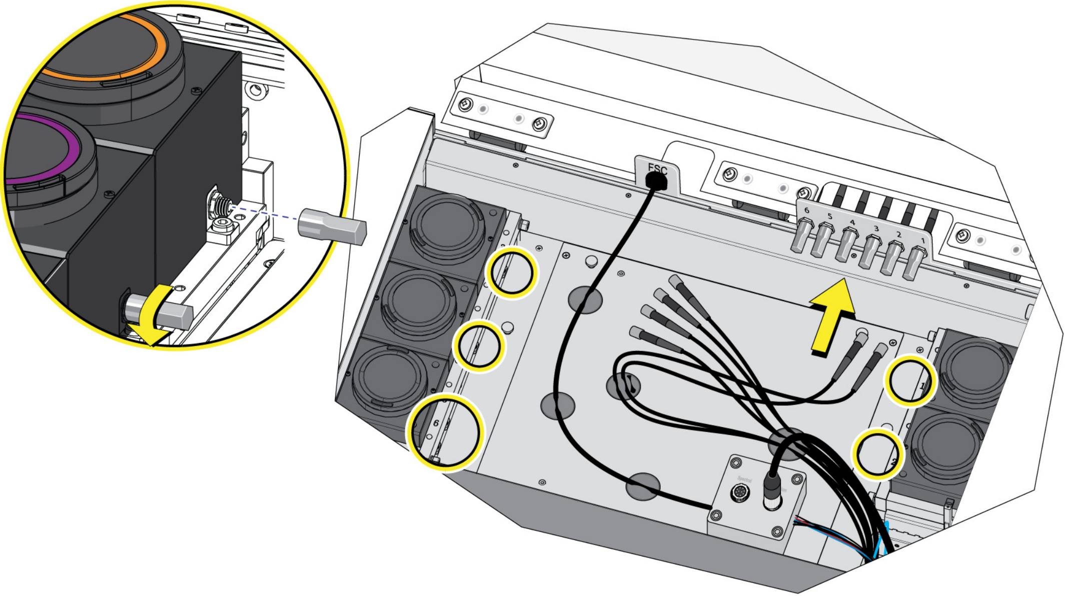


[CytoFLEX S Shown]

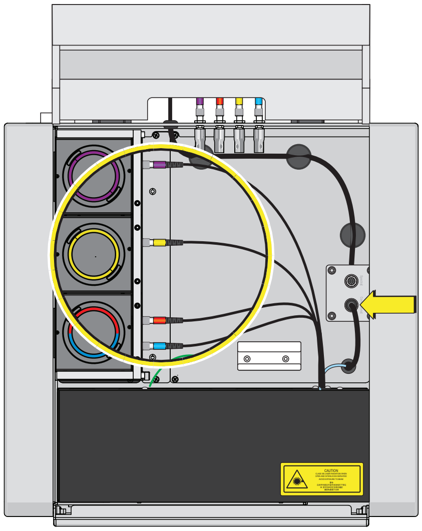


5 Remove the connector protective caps from the WDM optical fiber connectors, and connect them to the adapter optical fiber connectors.

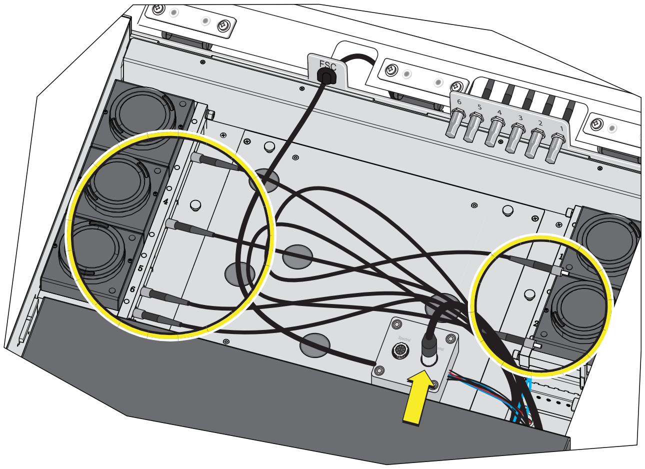
[CytoFLEX LX Shown]



6 Connect the correct optical fibers to the corresponding WDMs. Ensure the indicator number or color of the optical fiber matches the corresponding WDM. Refer to Table 12.1 and Table 12.2.



CytoFLEX S



CytoFLEX LX

7 Close the top cover.

8 Log out of the CytExpert for Spectral software. Refer to CHAPTER 4, Logging Out of the Software.

9 Log into the CytExpert software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Logging into the Software for detailed instructions.

10 Initialize the instrument. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Initializing the Instrument for detailed instructions.

NOTE If the instrument is turned off in Step 1, turn on the instrument. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Turning On the Instrument for detailed instructions.

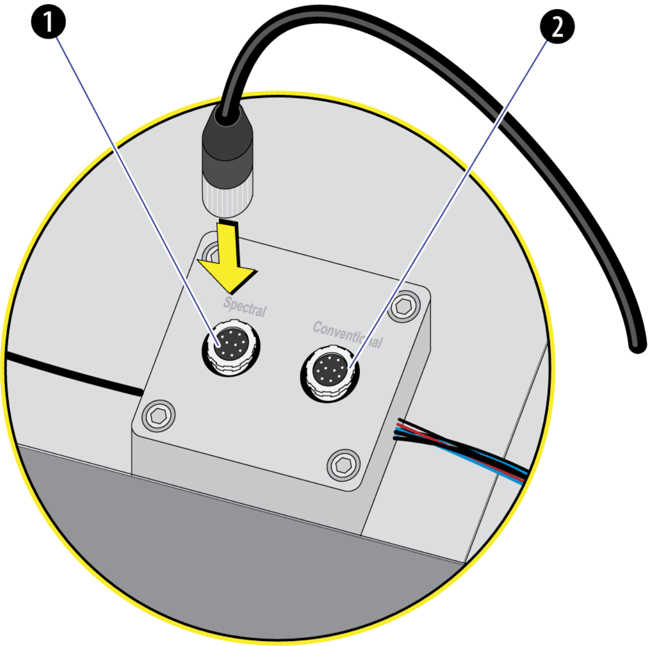
11 Run QC. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 5, Quality Control for detailed instructions.

Switching to Spectral Mode

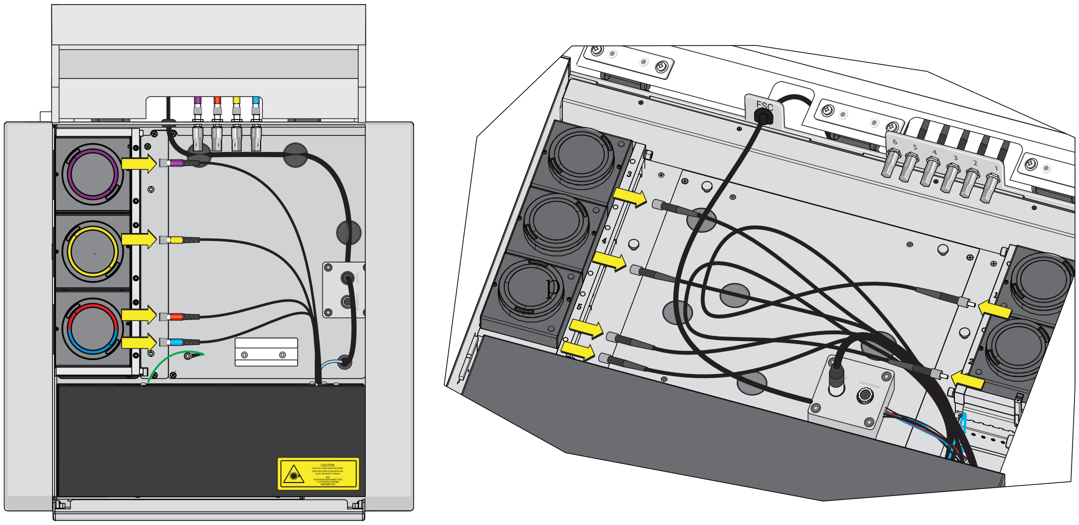
1 Confirm that the instrument is in the standby state or that the instrument is turned off.

2 Open the top cover.

3 Locate the Forward Scatter Switch and move the FSC connector from the Conventional Port **(2)** to the Spectral Port **(1)**.

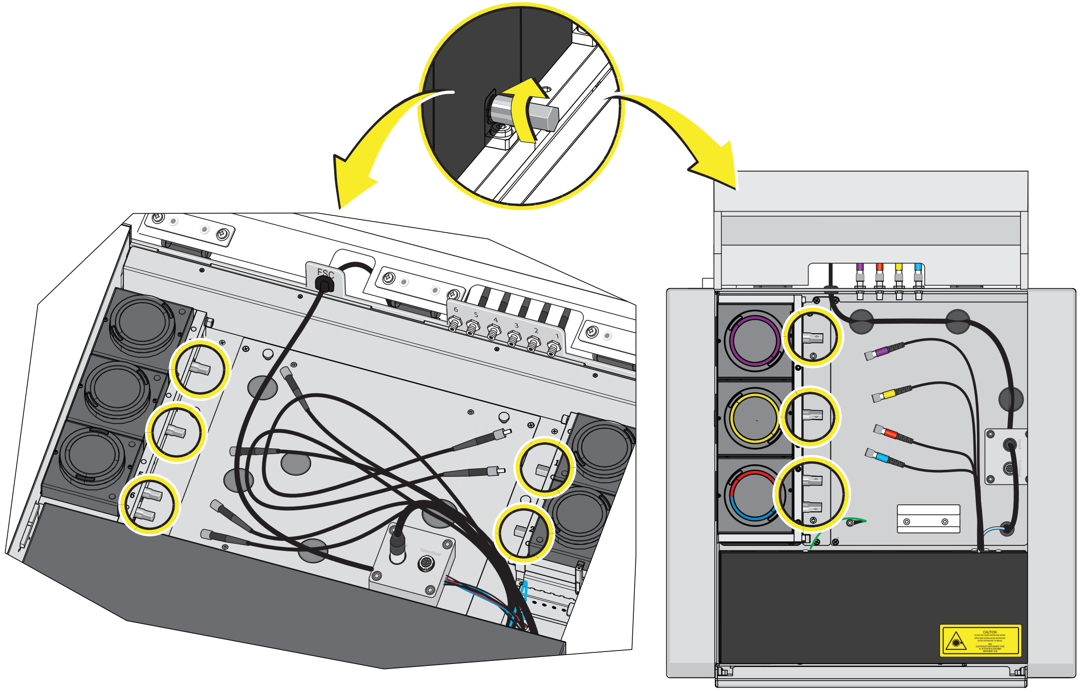


4 Disconnect the rest of the optical fibers.



CytoFLEX S **CytoFLEX LX**

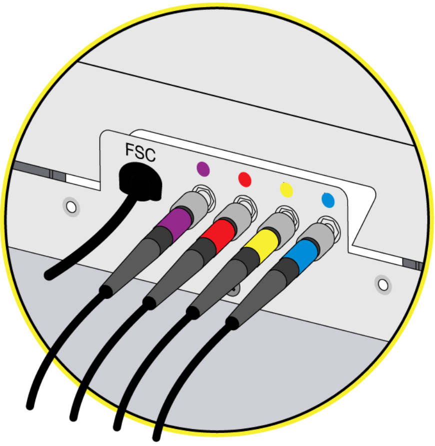
5 Remove the connector protective caps from the adapter optical fiber connectors, and connect them to the WDM optical fiber connectors.



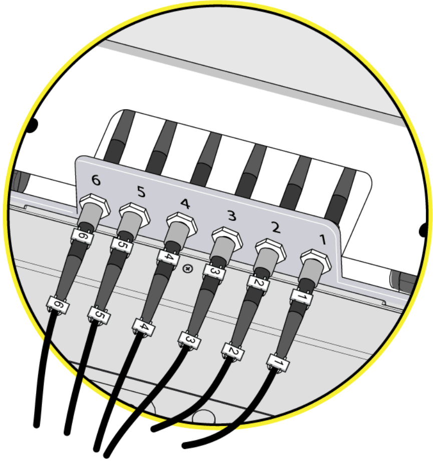
CytoFLEX LX

CytoFLEX S

6 Connect the correct optical fibers to the adapter optical fiber connectors. Ensure the indicator number or color of the optical fiber matches the corresponding adapter optical fiber connectors. Refer to Table 12.1 and Table 12.2



CytoFLEX S



CytoFLEX LX

7 Close the top cover.

8 Turn on the instrument and the CytoFLEX mosaic Spectral Detection Module. Refer to CHAPTER 4, Turning On the Instrument.

9 Log out of the CytExpert software. Refer to the *CytoFLEX Platform Instructions for Use* manual, CHAPTER 4, Logging Out of the Software for detailed instructions.

10 Log into the CytExpert for Spectral software. Refer to CHAPTER 4, Logging into the Software.

11 Initialize the instrument. Refer to CHAPTER 4, Initializing the Instrument.

12 Run QC. Refer to CHAPTER 5, Quality Control.

APPENDIX A

Instrument Installation

Overview

[CytoFLEX S]: Your instrument may have been shipped directly to your laboratory, in which case you will need to set up and connect the Cytometer and the Workstation. Refer to this chapter for the instrument installation procedure.

Validate the environment ➔ Open the package ➔ Install the instrument ➔ Prepare to boot

[CytoFLEX LX]: The CytoFLEX LX is installed by your Beckman Coulter Service Representative. Do not open the box or crate. Wait for a qualified Beckman Coulter Service Representative.

[CytoFLEX mosaic 63/88]: The CytoFLEX mosaic 63/88 is installed by your Beckman Coulter Service Representative. Do not open the box or crate. Wait for a qualified Beckman Coulter Service Representative.

This chapter contains information on:

- Instrument Transportation and Storage
- Installation Environment Validation
- Unpacking the Instrument and Inspecting the Materials for Defects or Omissions
- Installing the Instrument and Connecting the Equipment
- CytExpert for Spectral Software Installation Options
- Installing the CytoFLEX for Spectral Software
- Reinstalling the CytExpert for Spectral Software
- Restoring the Cytometer and CytoFLEX mosaic Spectral Detection Module Configuration File

Instrument Transportation and Storage

Refer to CHAPTER 11, Preparing the Instrument for Transport or Storage, prior to transportation or storage.

Attention to the following items is required when transporting or storing the instrument:

- Take caution to protect the instrument from exposure to rain or sunlight.
- Always place the instrument on a flat, stable surface, and take note of the symbol for *this side up*.
- Ambient temperature: -10 to 55 °C.
- Ambient humidity: < 80% RH.
- To prevent extrusion, the load on top cannot exceed 100 kg.

- CytoFLEX S Cytometer gross weight 27 kg; transport the instrument using only appropriate equipment so as to guard against personal injury.
- CytoFLEX LX Cytometer gross weight 103 kg; transport the instrument using only appropriate equipment so as to guard against personal injury.
- CytoFLEX mosaic 63 gross weight 68 kg; transport the instrument using only appropriate equipment so as to guard against personal injury.
- CytoFLEX mosaic 88 gross weight 71 kg; transport the instrument using only appropriate equipment so as to guard against personal injury.

Installation Environment Validation

IMPORTANT This instrument is intended for indoor use only.

Verify whether the installation environment satisfies the following requirements:

Worktable

⚠ CAUTION

Risk of instrument damage. Place the instrument on a level surface. Failing to do so places the system is in danger of toppling and can result in damage. Take all necessary precautions throughout the process of storing or transporting the instrument.

- The tabletop must be smooth and level.
- Minimum tabletop load bearing capacity [CytoFLEX S with CytoFLEX mosaic 63]: 90 kg.
- Minimum tabletop load bearing capacity [CytoFLEX LX with CytoFLEX mosaic 88]: 140 kg.
- The tabletop must not vibrate or shake.
- Minimum tabletop dimensions [CytoFLEX S with CytoFLEX mosaic 63]: 120 cm x 80 cm; minimum vertical space above tabletop: 100 cm.
- Minimum tabletop dimensions [CytoFLEX LX with CytoFLEX mosaic 88]: 200 cm x 80 cm; minimum vertical space above tabletop: 100 cm.
- Position the instrument in such a manner that it will facilitate disconnection of the power cable at the instrument end.

Ventilation and Cleaning

IMPORTANT If necessary, use ventilation equipment, but airflow must not be allowed to blow directly on the system, as it can affect the reliability of the data.

- Ensure that the working environment is well ventilated for proper heat dissipation.
- Maintain a clearance of at least 20 cm from the back of the instrument for heat dissipation.
- Keep the environment as dust free as possible.

- Avoid direct exposure to sunlight.
- Avoid placing near heat sources or exposing to drafts.
- Avoid corrosives or flammable gases.

Power Source

⚠ **DANGER**

Risk of electric shock and/or instrument damage. Ensure that the power source is properly grounded. Improper grounding can cause electric shock and damage the system. Verify that the output voltage of the power outlet conforms to the system requirements and that a 5 A, time delay, T 5 AL 250 VAC fuse is installed. To prevent personal injury, Beckman Coulter recommends using a power source designed to protect against electrical shock.

⚠ **CAUTION**

Possible instrument damage could occur if you use an extension cord or a power strip to connect the Cytometer. Always plug the Cytometer into a dedicated outlet with an isolated ground.

The power source requirements are as follows:

- This instrument has been tested to and meets all applicable requirements for CE Marking.
- This instrument complies with the emission and immunity requirements described in IEC 61326-1.
- This equipment has been designated and tested to CISPR 11 Class A. In a domestic environment it may cause radio interference, in which case, you may need to take measures to mitigate the interference.
- It is advised that the electromagnetic environment should be evaluated prior to operation of the device.
- Do not use this device in close proximity to sources of strong electromagnetic radiation (unshielded intentional RF sources), as these may interfere with the proper operation.
- 100-240 volts, 50/60 Hz, 3-wire power cable, well grounded.
- Amperage not less than 10 A.
- The system requires a well-grounded power outlet (150 VA normal, 250 VA max) to provide the necessary power.
- Distance from system to socket less than 1.5 m.

Power consumption of the Plate Loader is <30 W.

Temperature and Humidity

⚠ CAUTION

Risk of instrument damage and/or erroneous results. To ensure reliability, the system must be operated in the specified environment, within the required temperature and humidity ranges. If the ambient temperature or humidity level falls outside the ranges mentioned above, use appropriate air conditioning.

- **CytoFLEX S:** Ambient temperature: 15-27°C with fluctuations of no more than <±2°C per hour.
- **CytoFLEX LX:** Ambient temperature: 15-30°C with fluctuations of no more than <±2°C per hour.
- **CytoFLEX mosaic Spectral Detection Module:** Ambient temperature: 15-30°C with fluctuations of no more than <±2°C per hour.
- Relative humidity: 15% RH-80% RH, non-condensing.

Waste Disposal

⚠ WARNING

Risk of biohazardous contamination if you have skin contact with the waste container, its contents, and its associated tubing. The waste container and its associated tubing might contain residual biological material and must be handled with care. Clean up spills immediately. Dispose of the contents of the waste container in accordance with your local regulations and acceptable laboratory procedures.

The waste line from the Cytometer is connected to a waste container/cubitainer. Dispose of the system's waste in accordance with your local regulations and acceptable laboratory procedures.

The waste line supplied with the instrument can be connected to an open drain. If you use an open drain, mechanically secure the waste tube into the drain so the tube cannot accidentally come out of the drain. This prevents spillage.

Unpacking the Instrument and Inspecting the Materials for Defects or Omissions

⚠ CAUTION

Possible instrument damage could occur if you uncrate the instrument, install it, or set it up especially when you are in doubt of an instrument damage during shipment. Keep the instrument in its package until your Beckman Coulter Representative unpacks it for installation and setup.

When you receive your instrument, carefully inspect all cartons. If you see signs of mishandling or damage, file a claim with the carrier immediately. If separately insured, file the claim with the insurance company.

Take care to store the instrument in a suitable environment where it can remain in the proper position.

[CytoFLEX S and CytoFLEX LX]: Check that the following components on the packing list are present:

- Cytometer
- Cables
- Computer
- Mouse
- Keyboard
- Monitor
- Fluid Container holder
- Sheath fluid container [CytoFLEX S]
- Waste container
- Sheath fluid tubing
- Waste tubing
- USB configuration key
- Software USB

NOTE For the computer related components, refer to the manufacturer's manual.

[CytoFLEX mosaic Spectral Detection Module]: Check that the following components on the packing list are present:

- CytoFLEX mosaic
- Ethernet cable
- Power cable
- FSC cables
- USB configuration key
- Software USB

- Optical Fibers
- Top Cover and hinge board
- WDM Fixed block
- Cable ties
- FSC connect box
- FSC cable strain relief bush
- Connector protective covers
- Laser cover [CytoFLEX mosaic 88]
- Fiber digital wire clips [CytoFLEX mosaic 88]

Installing the Instrument and Connecting the Equipment

⚠ CAUTION

Risk of personal injury. Do not transfer the instrument from one location to another after the instrument installation is completed by service.

Installing the Instrument and Connecting the Equipment [CytoFLEX S]

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX A, Installing the Instrument and Connecting the Equipment [CytoFLEX] for detailed instructions. For connecting the instrument, refer to Figure 1.3.

Installing the Instrument and Connecting the Equipment [CytoFLEX LX]

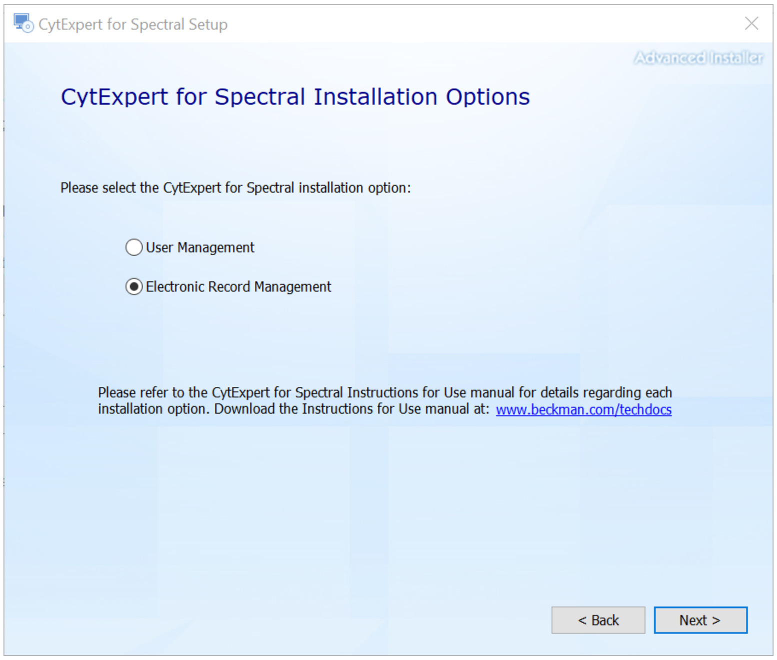
The CytoFLEX LX must be must be installed by a trained Beckman Coulter service engineer. For connecting the instrument, refer to Figure 1.6.

Installing the CytoFLEX mosaic Spectral Detection Module

The CytoFLEX mosaic Spectral Detection Module must be installed by a trained Beckman Coulter service engineer. For connecting the instrument, refer to Figure 1.3, Figure 1.6 and Figure 1.10.

CytExpert for Spectral Software Installation Options

CytExpert for Spectral software has two installation options upon install.



CytExpert for Spectral Installation Options

Please select the CytExpert for Spectral installation option:

○ User Management

● Electronic Record Management

Please refer to the CytExpert for Spectral Instructions for Use manual for details regarding each installation option. Download the Instructions for Use manual at: www.beckman.com/techdocs

- **CytExpert for Spectral User Management software option.** User Login is required to run the system. Contains features and functionality that facilitates user and role management.
- **CytExpert for Spectral Electronic Record Management software option.** User Login is required to run the system. Contains features and functionality that facilitates compliance with 21 CFR Part 11 guidelines for Electronic Records and Signatures.

Installing the CytoFLEX for Spectral Software

The CytExpert for Spectral software is installed by Beckman Coulter service.

Reinstalling the CytExpert for Spectral Software

IMPORTANT The CytExpert software and the CytExpert for Spectral software can be installed on the same computer, but you cannot use the CytExpert for Spectral software and the CytExpert software together.

Use this procedure to:

- Change the software option installed. Refer to CytExpert for Spectral Software Installation Options for the differences between each software option available.
- Reinstall the same version of software.
- Upgrade your software to a higher version.

NOTE If you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX A, Reinstalling the CytExpert Software.

⚠ CAUTION

Risk of data loss. Reinstalling the CytExpert for Spectral software could overwrite your database. Ensure you backup your database prior to software reinstallation.

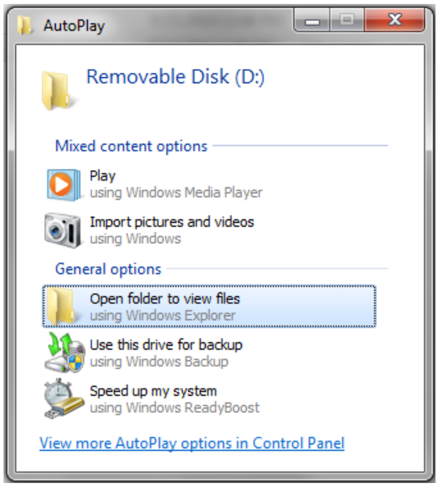
Uninstalling the Software

IMPORTANT Ensure you backup all of your data before uninstalling the software.

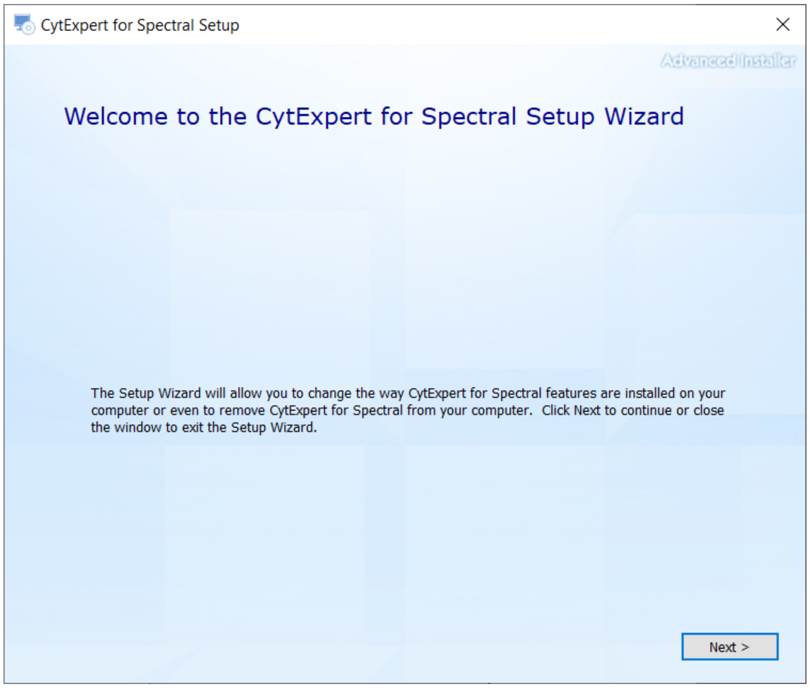
1 Backup your CytExpert for Spectral data if you previously had the User Management or Electronic Record Management software option installed. Refer to CHAPTER 10, Backup and Restore.

2 Insert the software USB into the computer.

NOTE If the Autoplay window appears, select **Open folder to view files**.



3 Select **CytExpert for Spectral Setup_x.x.x.exe**. The Welcome to the CytExpert for Spectral Setup Wizard window appears.

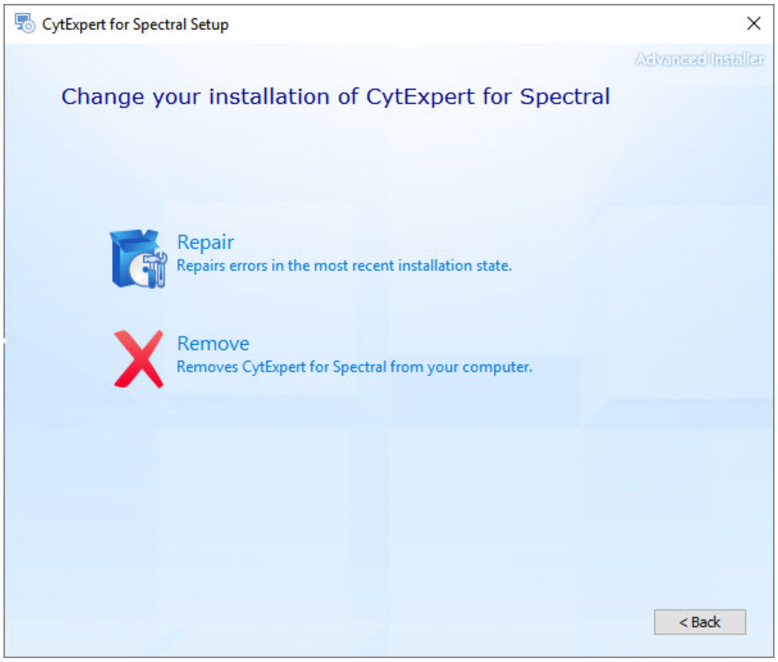

CytExpert for Spectral Setup
Advanced Installer
Welcome to the CytExpert for Spectral Setup Wizard
Next >

The Setup Wizard will allow you to change the way CytExpert for Spectral features are installed on your computer or even to remove CytExpert for Spectral from your computer. Click Next to continue or close the window to exit the Setup Wizard.

A User Account Control message may display the message *Do you want to allow this app to make changes to your device?* Select **Yes** to continue.

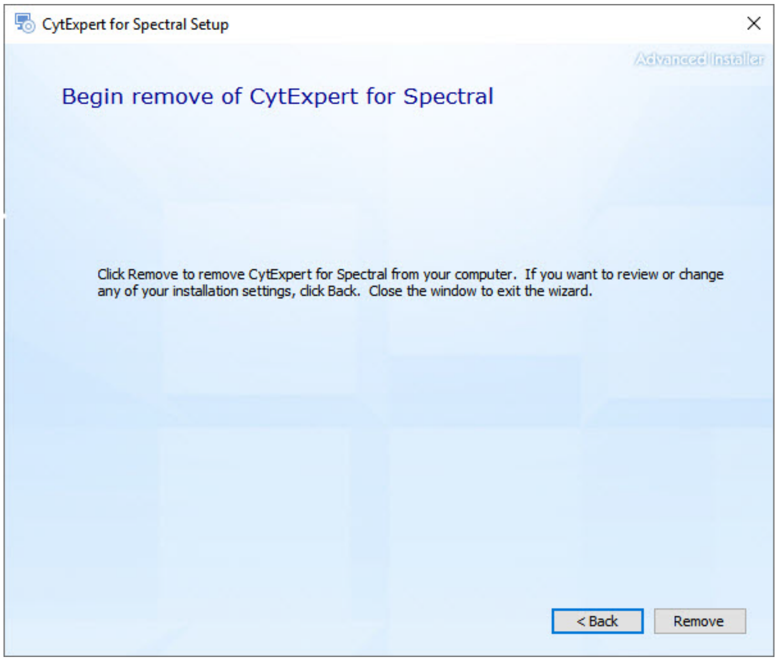
---

4 Select **Next.** The Change your installation of CytExpert for Spectral screen appears.


CytExpert for Spectral Setup
Advanced Installer
Change your installation of CytExpert for Spectral
Repair
Repairs errors in the most recent installation state.
Remove
Removes CytExpert for Spectral from your computer.
< Back

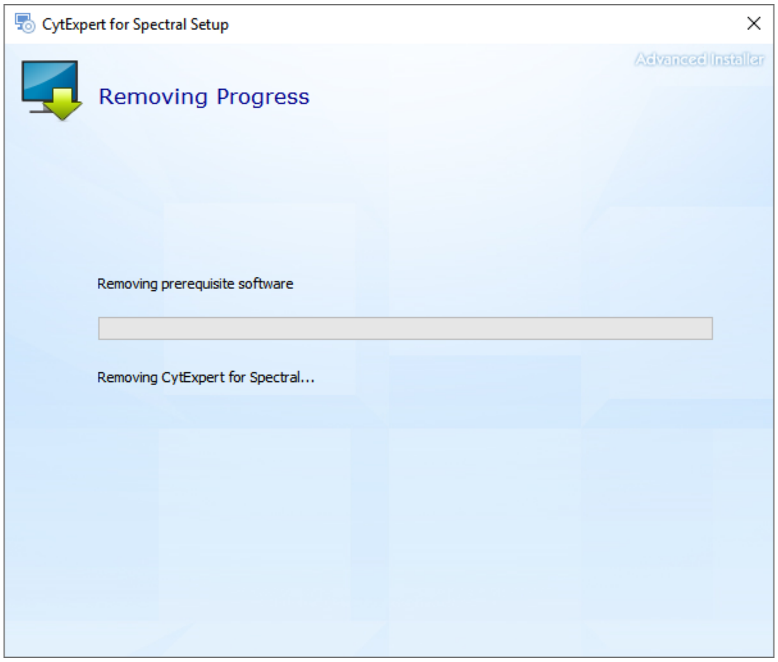
---

5 Select **Remove.** The Begin remove of CytExpert for Spectral screen appears.


CytExpert for Spectral Setup
Advanced Installer
Begin remove of CytExpert for Spectral

Click Remove to remove CytExpert for Spectral from your computer. If you want to review or change any of your installation settings, click Back. Close the window to exit the wizard.

6 Select **Remove**. The Removing Progress window appears.

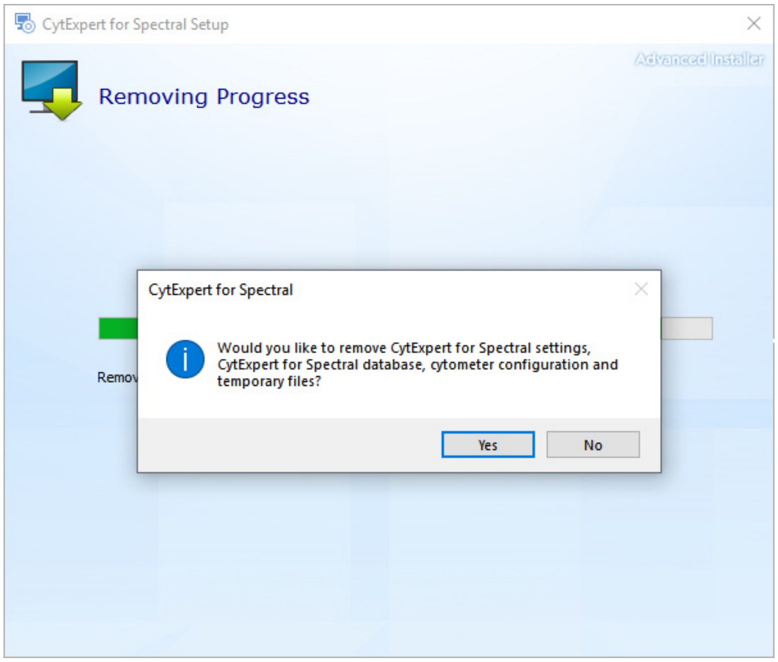


Removing prerequisite software

Removing CytExpert for Spectral...

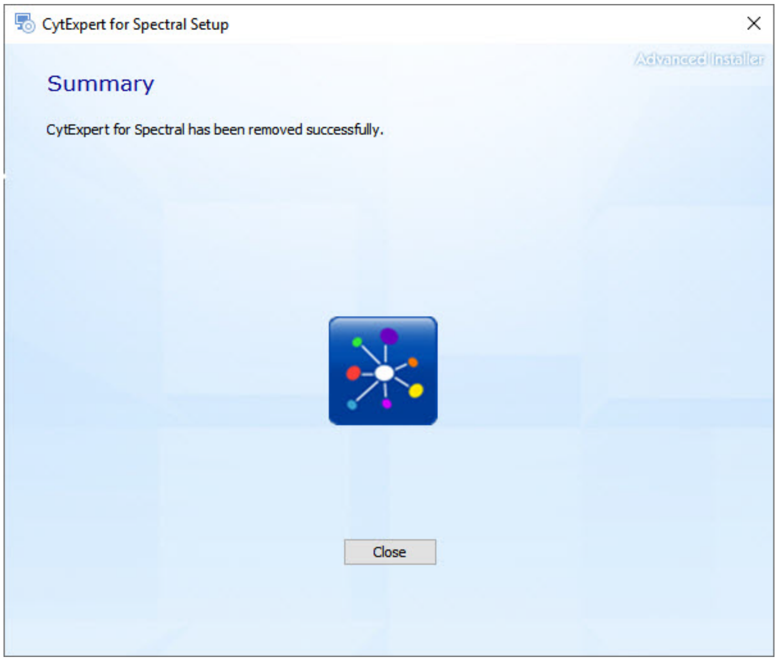
IMPORTANT If you select **Yes** to remove the CytExpert for Spectral settings, CytExpert for Spectral database, cytometer configuration, and temporary files, your settings, database, configuration files, and temporary files will be overwritten and you will need to reinstall your cytometer configuration and restore any databases you might have.

7 The message *Would you like to remove CytExpert for Spectral settings, CytExpert for Spectral database, cytometer configuration and temporary files?* appears. Select **No**.



Would you like to remove CytExpert for Spectral settings, CytExpert for Spectral database, cytometer configuration and temporary files?

8 When software removal is complete, the software displays the message *CytExpert for Spectral has been removed successfully.*



Summary

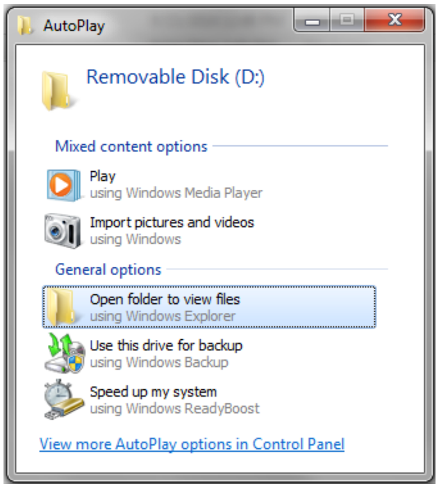
CytExpert for Spectral has been removed successfully.

9 Select **Close.**

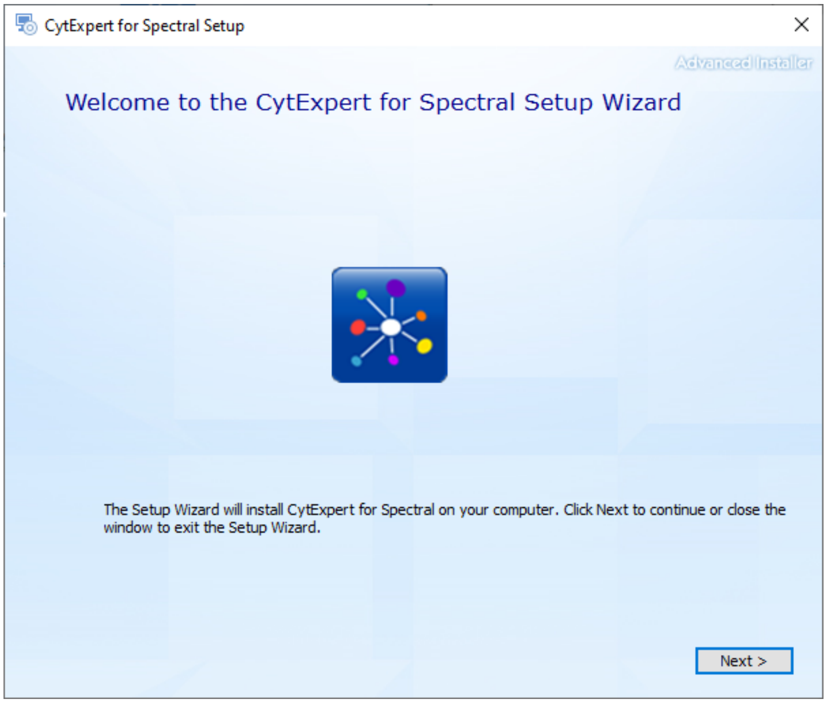
Reinstalling the CytExpert for Spectral Software

1 Insert the software USB into the computer.

NOTE If the Autoplay window appears, select **Open folder to view files.**



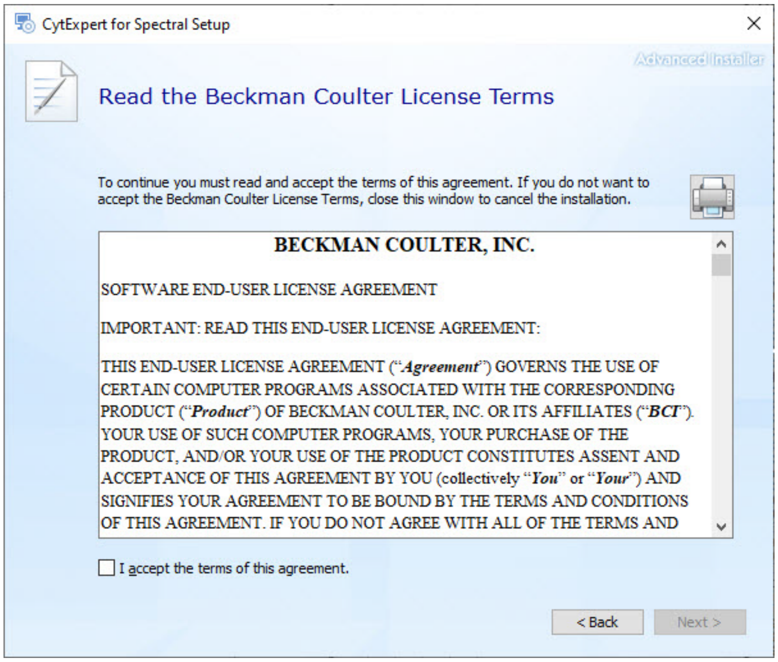
2 Select **CytExpert for Spectral Setup_x.x.x.exe.** The Welcome to the CytExpert for Spectral Setup Wizard window appears.


CytExpert for Spectral Setup
Advanced Installer
Welcome to the CytExpert for Spectral Setup Wizard
Next >

The Setup Wizard will install CytExpert for Spectral on your computer. Click Next to continue or close the window to exit the Setup Wizard.

A User Account Control message may display the message *Do you want to allow this app to make changes to your device?* Select **Yes** to continue.

3 Select **Next**. The Beckman Coulter License Terms window appears.



Read the Beckman Coulter License Terms

To continue you must read and accept the terms of this agreement. If you do not want to accept the Beckman Coulter License Terms, close this window to cancel the installation.

BECKMAN COULTER, INC.

SOFTWARE END-USER LICENSE AGREEMENT

IMPORTANT: READ THIS END-USER LICENSE AGREEMENT:

THIS END-USER LICENSE AGREEMENT (“***Agreement***”) GOVERNS THE USE OF CERTAIN COMPUTER PROGRAMS ASSOCIATED WITH THE CORRESPONDING PRODUCT (“***Product***”) OF BECKMAN COULTER, INC. OR ITS AFFILIATES (“***BCI***”). YOUR USE OF SUCH COMPUTER PROGRAMS, YOUR PURCHASE OF THE PRODUCT, AND/OR YOUR USE OF THE PRODUCT CONSTITUTES ASSENT AND ACCEPTANCE OF THIS AGREEMENT BY YOU (collectively “***You***” or “***Your***”) AND SIGNIFIES YOUR AGREEMENT TO BE BOUND BY THE TERMS AND CONDITIONS OF THIS AGREEMENT. IF YOU DO NOT AGREE WITH ALL OF THE TERMS AND

☐ I accept the terms of this agreement.

---

4 Read the Beckman Coulter Customer End User License Agreement.

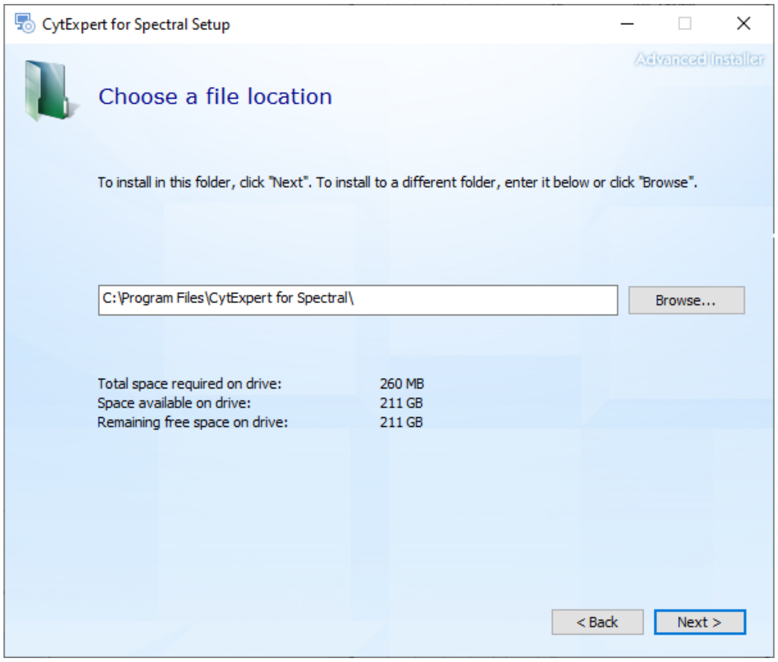
---

5 Select the *I accept the terms of this agreement* checkbox.

NOTE The checkbox is not selectable until you scroll all the way to the end of the agreement.

---

6 Select **Next**. The Choose a file location window appears.



Choose a file location

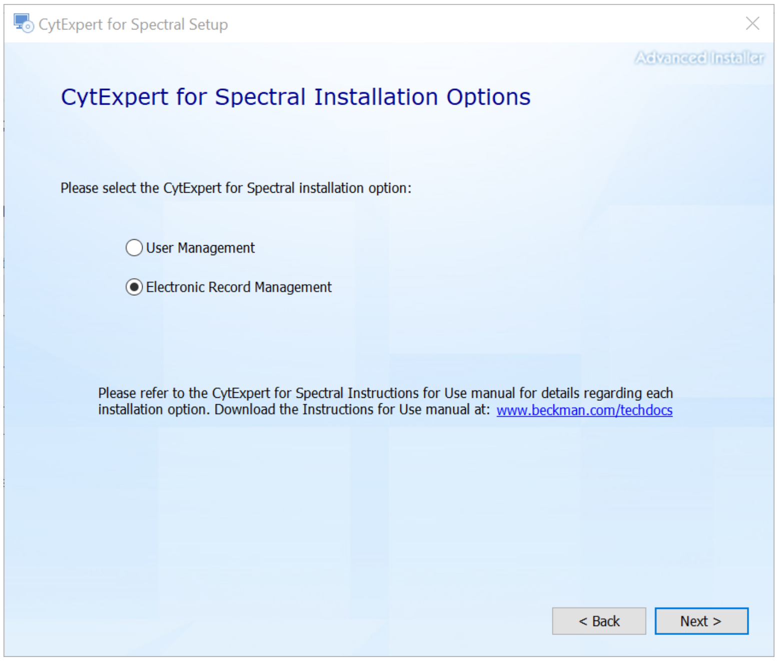
To install in this folder, click "Next". To install to a different folder, enter it below or click "Browse".

C:\Program Files\CytExpert for Spectral\

Total space required on drive:	260 MB
Space available on drive:	211 GB
Remaining free space on drive:	211 GB

---

7 Select **Next**. The Installation Options window appears.



CytExpert for Spectral Installation Options

Please select the CytExpert for Spectral installation option:

○ User Management

● Electronic Record Management

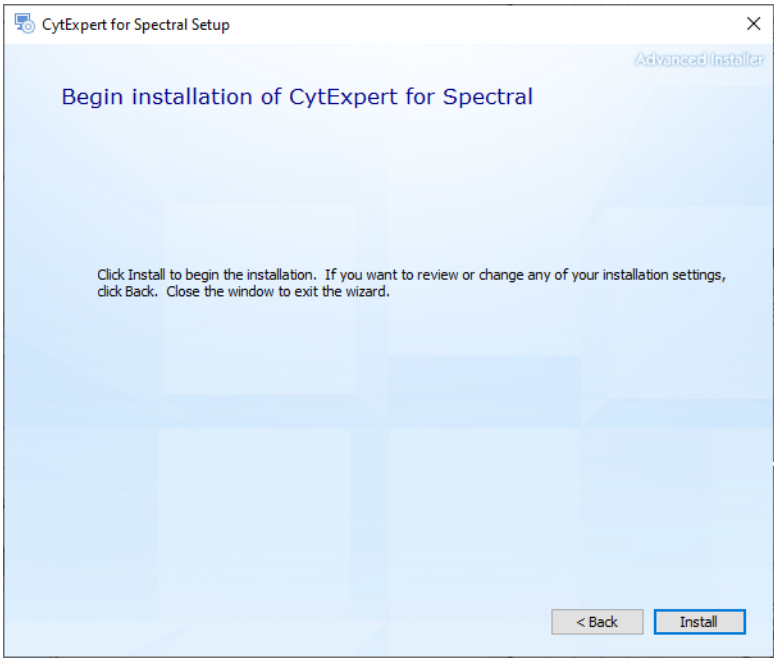
Please refer to the CytExpert for Spectral Instructions for Use manual for details regarding each installation option. Download the Instructions for Use manual at: www.beckman.com/techdocs

---

8 Select the desired installation option. Refer to CytExpert for Spectral Software Installation Options.

---

9 Select **Next**. The Begin installation of CytExpert for Spectral window appears.

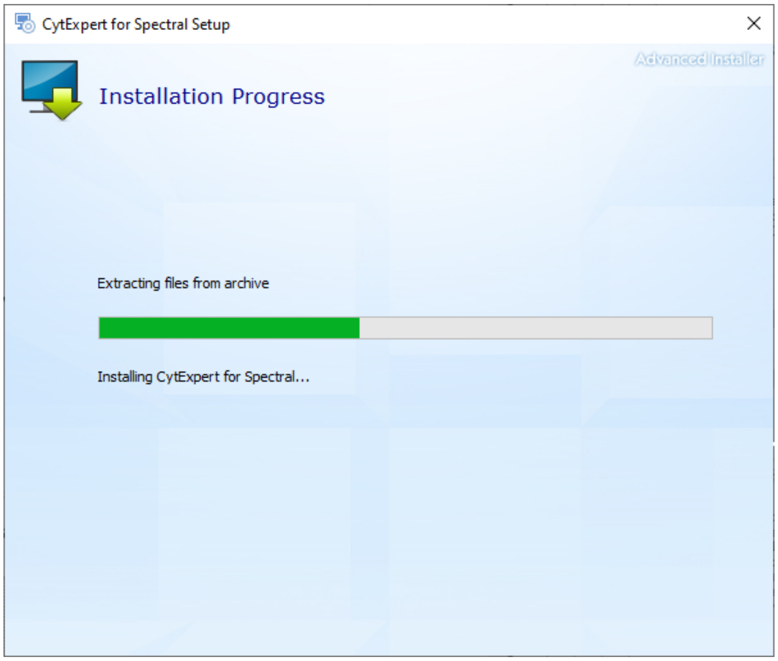


Begin installation of CytExpert for Spectral

Click Install to begin the installation. If you want to review or change any of your installation settings, click Back. Close the window to exit the wizard.

---

10 Select **Install** to begin installing the software. The Installation Progress window appears.

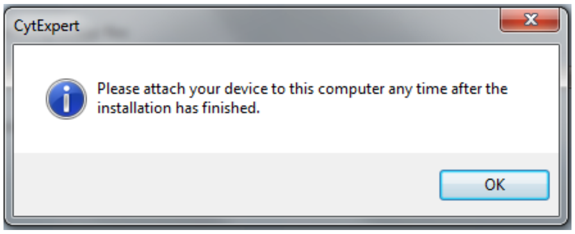


Installation Progress

Extracting files from archive

Installing CytExpert for Spectral...

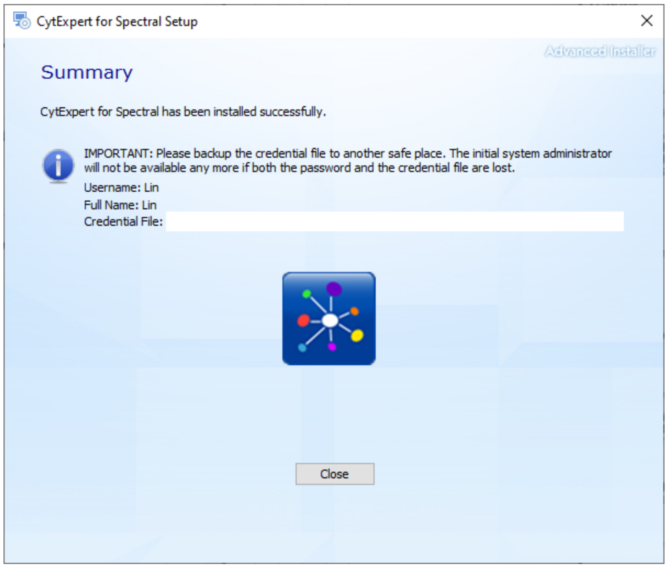
11 If the following software prompt appears, select **OK**.



Please attach your device to this computer any time after the installation has finished.

NOTE The term “device” in this message refers to the cytometer.

12 Wait for the software to finish installing. The install complete window appears.



Summary

CytExpert for Spectral has been installed successfully.

IMPORTANT: Please backup the credential file to another safe place. The initial system administrator will not be available any more if both the password and the credential file are lost.

Username: Lin

Full Name: Lin

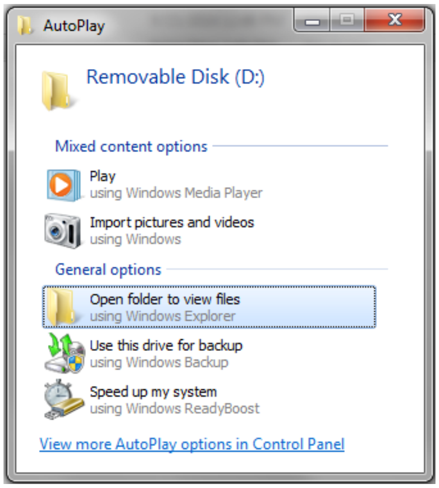
Credential File:

13 Select **Close** to finish the CytExpert for Spectral software installation.

Restoring the Cytometer and CytoFLEX mosaic Spectral Detection Module Configuration File

1 Insert the software USB into the computer.

NOTE If the Autoplay window appears, select **Open folder to view files**.

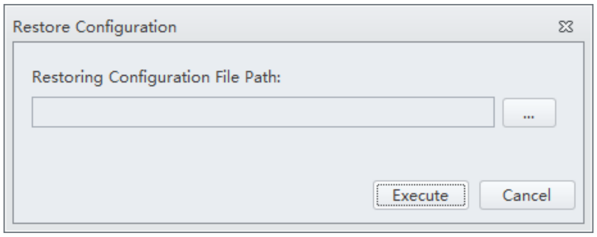


---

2 Open the CytExpert for Spectral software. Refer to CHAPTER 4, Logging into the Software.

---

3 Select **Advanced** > **Restore Configuration.** The Restore Configuration window appears.



NOTE Ensure the experiment is closed.

---

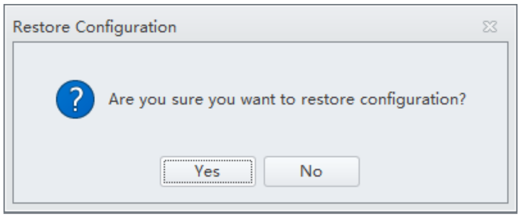
4 Select ... and browse to the desired configuration file folder from your Software USB.

---

5 Open the Cytometer configuration file **CytExpert for Spectral_x.x.xxx_Config_Backup_.bak**.

---

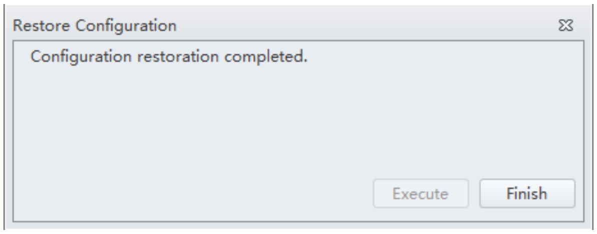
6 Select **Execute**. The Restore Configuration window appears.



Are you sure you want to restore configuration?

---

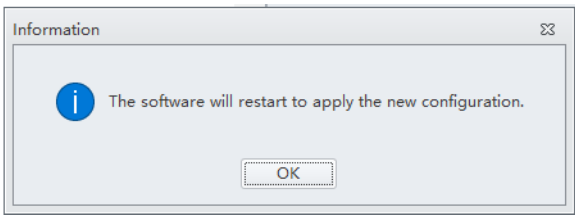
7 Select **Yes** to start restore the configuration.



Configuration restoration completed.

---

8 Select **Finish**.



The software will restart to apply the new configuration.

NOTE If the CytExpert software is installed, its configuration will automatically synchronized with the restored configuration. Restart the CytExpert software to apply the restored configurations.

---

9 Select **OK**.

APPENDIX B

CytExpert for Spectral Electronic Record Management

Overview

IMPORTANT You must have the CytExpert for Spectral Electronic Record Management software option installed to use the features listed below. Refer to APPENDIX A, CytExpert for Spectral Software Installation Options.

Beckman Coulter's CytoFLEX with CytExpert for Spectral software version 1.0 and higher contains features and functionality that facilitates compliance with 21 CFR Part 11 guidelines for Electronic Records and Signatures. This electronic record management includes controls for user identification, permissions, electronic signatures, data integrity, operation and experiment logs and audit trails. CytExpert for Spectral software version 1.0 and higher contains a database that uses checksum matching to prevent tampering of the records and files that are indexed in the Closed File System.

This chapter contains information on:

- Software Menu
- Experiment Management
- Log
- Electronic Signature
- User Management

Software Menu

The CytExpert for Spectral Electronic Record Management software option includes additional software menu items that are not available in the CytExpert for Spectral User Management software option. Refer to Figure 2.5 and Figure 2.6 for comprehensive software menu trees and details on which menu item applies to each software option.

NOTE If your CytoFLEX instrument is not equipped with a CytoFLEX mosaic Spectral Detection Module or if you are using Conventional Mode, refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX B, Software Menu.

Experiment Management

⚠ **CAUTION**

Risk of file corruption. Do not add, delete, or modify data from the Windows Explorer directory. Manage all data changes using Experiment Explorer to ensure file indexing remains intact.

Closed File System

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX A, Closed File System for detailed instructions.

Experiment Directory Management

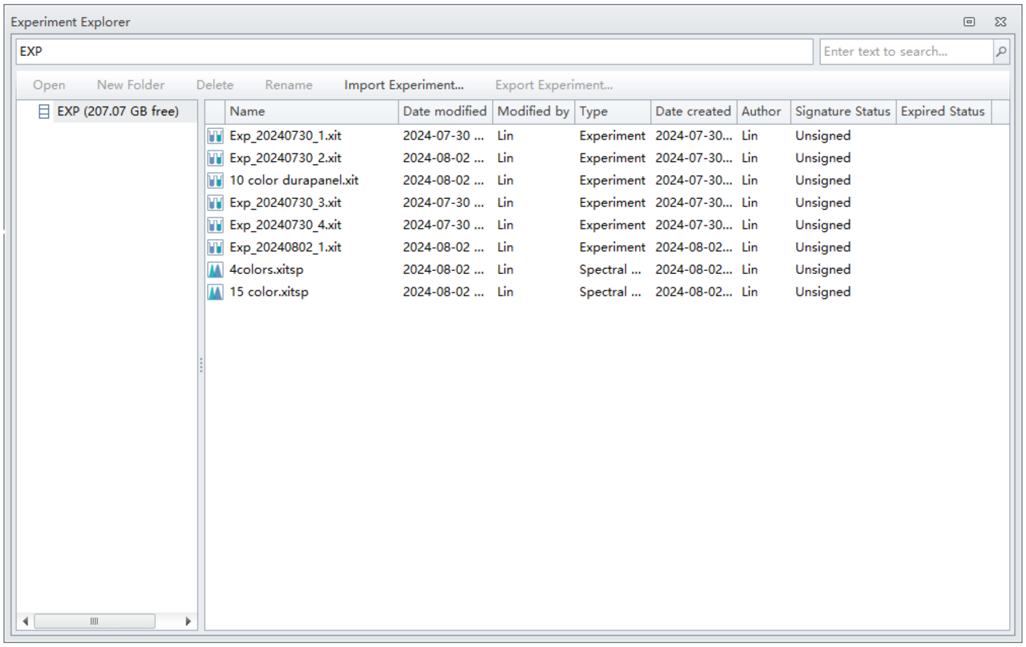
Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX A, Experiment Directory Management for detailed instructions.

Folder Hierarchy Management

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX A, Folder Hierarchy Management for detailed instructions.

Experiment Related Operations

The Experiment Explorer dialog appears in place of the Windows File Explorer in the following operations: New/Open Experiment, New/Open Spectral Unmixing, Save As, Save As Template, Recent Template, and New Experiment from Template.



Name	Date modified	Modified by	Type	Date created	Author	Signature Status	Expired Status
Exp_20240730_1.xit	2024-07-30 ...	Lin	Experiment	2024-07-30...	Lin	Unsigned	
Exp_20240730_2.xit	2024-08-02 ...	Lin	Experiment	2024-07-30...	Lin	Unsigned	
10 color durapanel.xit	2024-08-02 ...	Lin	Experiment	2024-07-30...	Lin	Unsigned	
Exp_20240730_3.xit	2024-07-30 ...	Lin	Experiment	2024-07-30...	Lin	Unsigned	
Exp_20240730_4.xit	2024-07-30 ...	Lin	Experiment	2024-07-30...	Lin	Unsigned	
Exp_20240802_1.xit	2024-08-02 ...	Lin	Experiment	2024-08-02...	Lin	Unsigned	
4colors.xitsp	2024-08-02 ...	Lin	Spectral ...	2024-08-02...	Lin	Unsigned	
15 color.xitsp	2024-08-02 ...	Lin	Spectral ...	2024-08-02...	Lin	Unsigned	

Importing an Experiment/Template

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX B, Importing an Experiment/Template for detailed instructions on importing experiment (.xit), spectral unmixing (.xitsp), or experiment from template (.xitm) files into the system.

Exporting an Experiment/Template

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX B, Exporting an Experiment/Template for detailed instructions on exporting experiment (.xit), spectral unmixing (.xitsp), or experiment from template (.xitm) files from the system.

Log

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX B, Log for detailed instructions on the following:

- Experiment Operation Log
- System Operation Log
- User Management Operation Log

Electronic Signature

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX B, Electronic Signature for detailed instructions on the following procedures:

- Signing Experiments
- Rejecting Experiment
- Setting the Signature Retention Period
- Signature Setting
- Printing an Experiment Signature

User Management

User Administration

Logging In and Out of the Software

Refer to CHAPTER 4, Logging into the Software and CHAPTER 4, Logging Out of the Software.

Locking the Account

Refer to CHAPTER 4, Locking the Account.

Role Management

Refer to CHAPTER 2, Role Management.

User Management

Refer to CHAPTER 2, User Management.

Account Policies

Refer to CHAPTER 2, Account Policies.

APPENDIX C

Sample Injection Mode Control Kit

Overview

The Sample Injection Mode Control Kit is a mechanical knob installed by your service engineer that enables users to switch between the Plate Loader sample injection mode and the Semi-Automatic or manual sample injection mode. The switch eliminates the need to manually re-route the tubing.

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX C, Sample Injection Mode Control Kit for detailed instructions on the following procedures:

- Performance Characteristics [With the Sample Injection Mode Control Knob]
- Sample Injection Mode Control Kit Components
- Switching the Sample Probe from the Single Tube Sample Station to the Plate Loader [With Sample Injection Mode Control Knob]
- Switching the Sample Probe from the Plate Loader to the Single Tube Sample Station [With Sample Injection Mode Control Knob]

APPENDIX D

Deep Well Plate

Specimen Collection Plate Specifications

Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX D, Specimen Collection Plate Specifications for detailed instructions.

APPENDIX E

Cyber Security

Overview

The following procedures are the only validated and approved recommendations for cyber privacy and security. Contact your IT professional for assistance.

- Changes below require Administrator level Windows access.
- Before connecting to an external device such as a hard drive, USB, or DVD/CD, verify it does not have a virus or malware.

  NOTE Beckman Coulter recommends using multiple anti-virus software to ensure that all viruses or malware can be identified.

- Close all unnecessary applications while operating the CytExpert for Spectral software.

⚠ CAUTION

Risk of security vulnerabilities and privacy issues. Use the USB and Ethernet ports with caution. Unauthorized or improper use of these ports will lead to potential information security risks. Beckman Coulter recommends disabling all the unused USB and Ethernet ports on the instrument computer. Ensure that only trusted and authorized devices are connected to the active ports.

To prevent unauthorized access and data breaches, Beckman Coulter recommends checking the use of the USB and Ethernet ports on a regular basis.

⚠ CAUTION

System integrity could be compromised and operational failures could occur if:

- **You introduce software that is not authorized by Beckman Coulter into your computer. Only operate your system's computer with software authorized by Beckman Coulter.**
- **You apply domain policies which alter the default configuration.**
- **You alter the system in a manner other than the approved changes outlined below.**

IMPORTANT Beckman Coulter Life Sciences has assembled a global product security team to assess vulnerabilities and determine responses in a coordinated Vulnerability Disclosure (CVD) process. If you encounter a potential threat to cyber security, please contact us via https://www.beckman.com/about-us/compliance/coordinated-vulnerability-disclosure.

This chapter contains information on:

- Drive Encryption
- Protection from Malware Software
- Security Updates
- User Management
- System Hardening
- Remote Access

Drive Encryption

Your Windows 10 system is equipped with BitLocker. Use BitLocker disk encryption software to prevent unauthorized access to your hard drive, refer to BitLocker Keys. For instructions on BitLocker Encryption or BitLocker Decryption, see Enabling BitLocker or BitLocker Decryption.

IMPORTANT BitLocker is the only approved encryption method.

BitLocker Keys

⚠ CAUTION

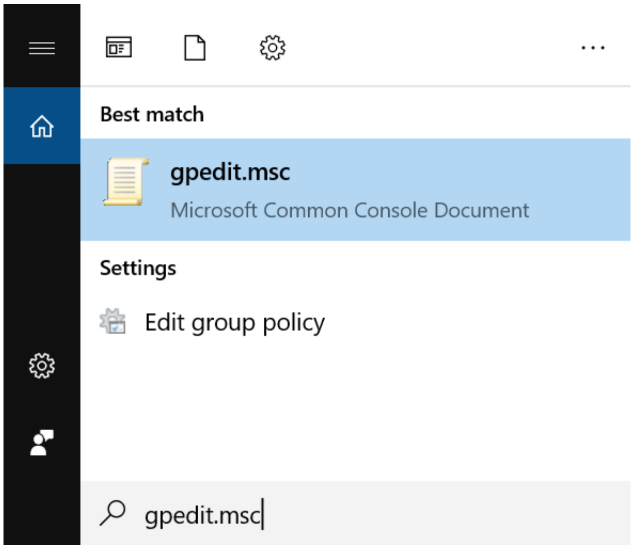
Risk of data loss. Store your BitLocker key in a safe place. For instructions on saving your BitLocker key, refer to Backing Up Your Recovery Key. If you lose your BitLocker Key, you will lose access to your entire hard drive including all of your data. Beckman Coulter is not responsible for security keys and will not be able to recover your data off your hard drive. Work with your IT department in creating and storing the keys in a safe place.

Changing Encryption Strength Level for BitLocker

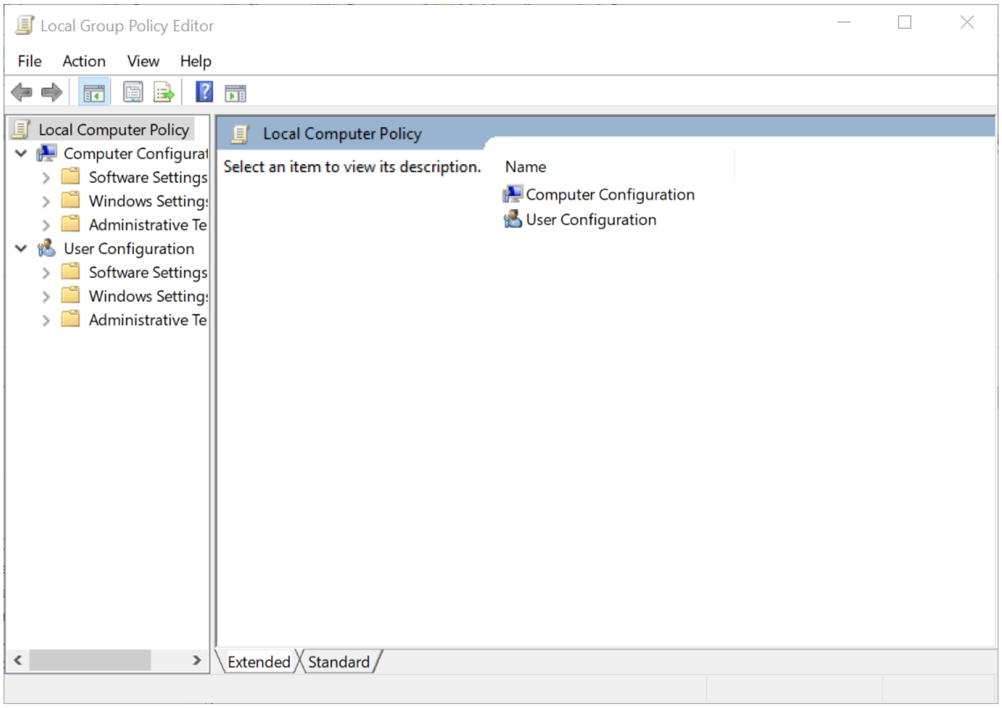
BitLocker defaults to XTS-AES 128-bit encryption method. To alter the encryption, follow the steps below before enabling BitLocker. Changes to the encryption level will only impact the encryption of new volumes. Work with your IT department to change the Encryption Strength Level.

Changing the encryption strength for BitLocker

1 Type **gpedit.msc** from the Windows search bar to locate the gpedit Control Panel.

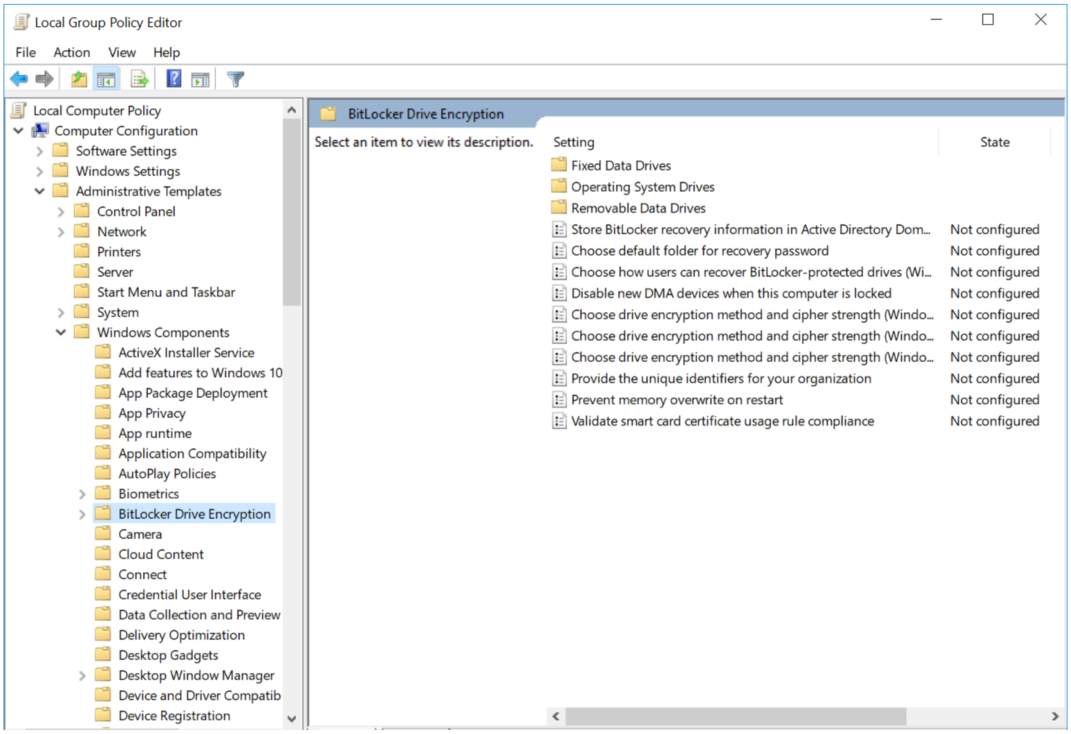


2 Click **gpedit.msc**. The Local Group Policy Editor window appears.



---

3 Navigate to **Computer Configuration** > **Administrative Templates** > **Windows Components** > **BitLocker Drive Encryption**.

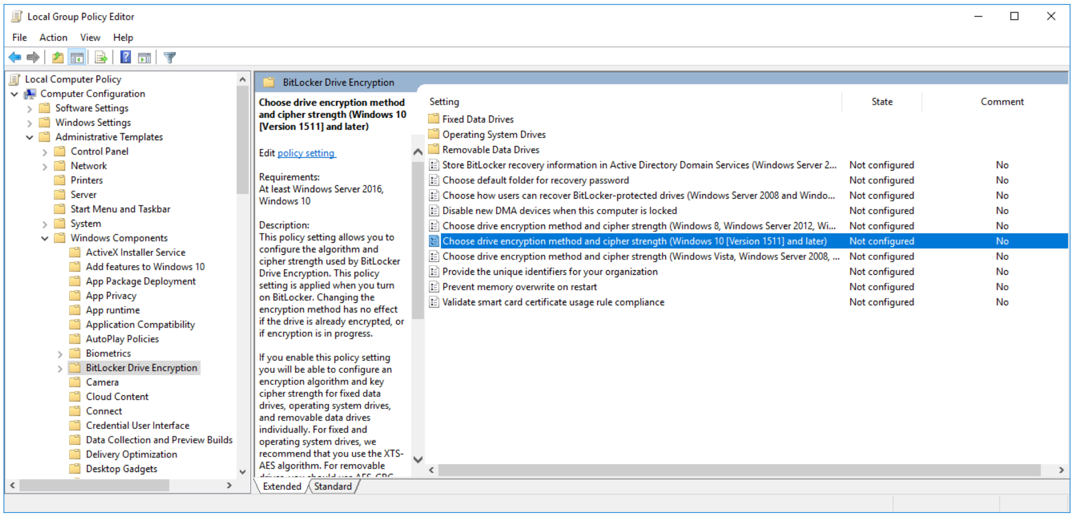


Select an item to view its description.

Setting	State
Fixed Data Drives	
Operating System Drives	
Removable Data Drives	
Store BitLocker recovery information in Active Directory Dom...	Not configured
Choose default folder for recovery password	Not configured
Choose how users can recover BitLocker-protected drives (Wi...	Not configured
Disable new DMA devices when this computer is locked	Not configured
Choose drive encryption method and cipher strength (Windo...	Not configured
Choose drive encryption method and cipher strength (Windo...	Not configured
Choose drive encryption method and cipher strength (Windo...	Not configured
Provide the unique identifiers for your organization	Not configured
Prevent memory overwrite on restart	Not configured
Validate smart card certificate usage rule compliance	Not configured

---

4 Double-click *"Choose drive encryption method and cipher strength (Windows 10.....)"*.



Choose drive encryption method and cipher strength (Windows 10 [Version 1511] and later)

Edit policy setting

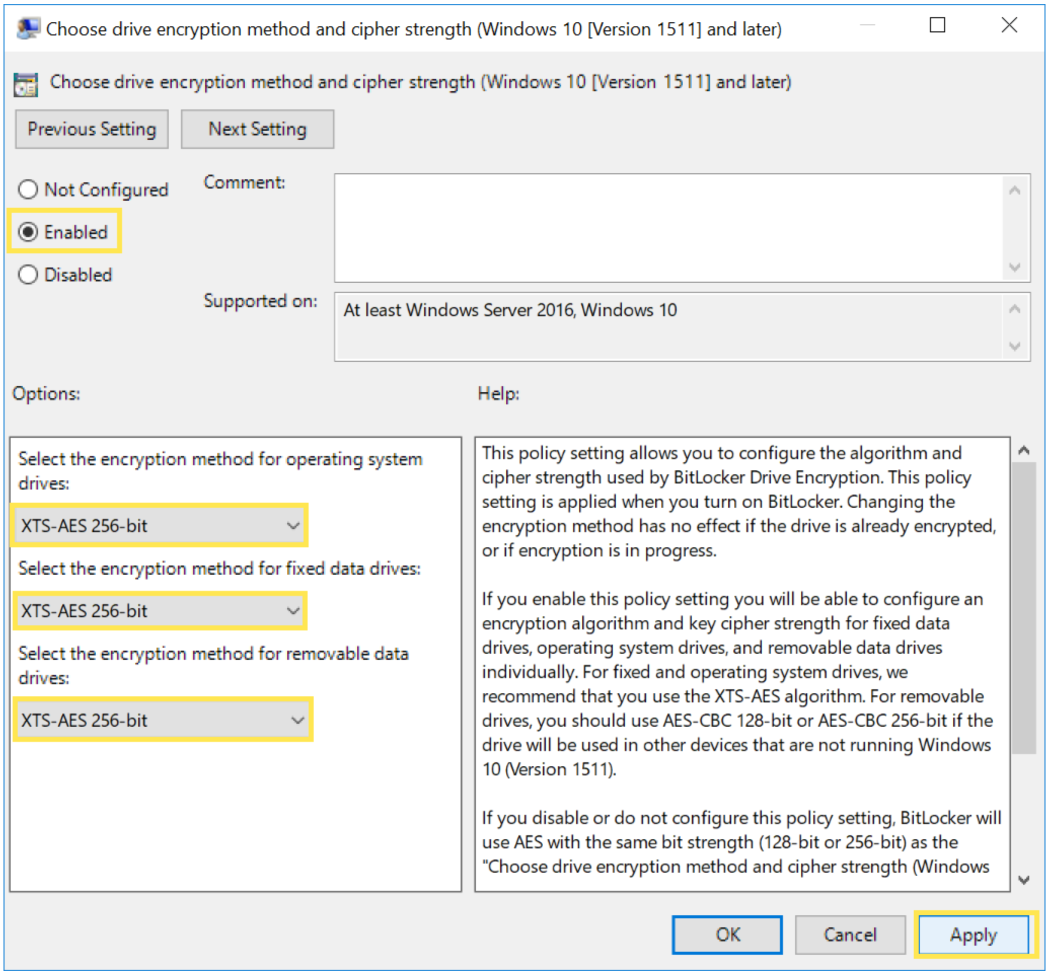
Requirements:
At least Windows Server 2016, Windows 10

Description:
This policy setting allows you to configure the algorithm and cipher strength used by BitLocker Drive Encryption. This policy setting is applied when you turn on BitLocker. Changing the encryption method has no effect if the drive is already encrypted, or if encryption is in progress.

If you enable this policy setting you will be able to configure an encryption algorithm and key cipher strength for fixed data drives, operating system drives, and removable data drives individually. For fixed and operating system drives, we recommend that you use the XTS-AES algorithm. For removable

Setting	State	Comment
Fixed Data Drives		
Operating System Drives		
Removable Data Drives		
Store BitLocker recovery information in Active Directory Domain Services (Windows Server 2...	Not configured	No
Choose default folder for recovery password	Not configured	No
Choose how users can recover BitLocker-protected drives (Windows Server 2008 and Windo...	Not configured	No
Disable new DMA devices when this computer is locked	Not configured	No
Choose drive encryption method and cipher strength (Windows 8, Windows Server 2012, Wi...	Not configured	No
Choose drive encryption method and cipher strength (Windows 10 [Version 1511] and later)	Not configured	No
Choose drive encryption method and cipher strength (Windows Vista, Windows Server 2008, ...	Not configured	No
Provide the unique identifiers for your organization	Not configured	No
Prevent memory overwrite on restart	Not configured	No
Validate smart card certificate usage rule compliance	Not configured	No

5 Select **Enabled** and use the dropdown arrows to select the encryption method for the operating system drive, fixed data drive, and for the removable data drives.


Choose drive encryption method and cipher strength (Windows 10 [Version 1511] and later)
Choose drive encryption method and cipher strength (Windows 10 [Version 1511] and later)
Previous Setting
Next Setting
Comment:
Options:
Help:
OK
Cancel
Apply

○ Not Configured
● Enabled
○ Disabled

Supported on: At least Windows Server 2016, Windows 10

Select the encryption method for operating system drives: XTS-AES 256-bit

Select the encryption method for fixed data drives: XTS-AES 256-bit

Select the encryption method for removable data drives: XTS-AES 256-bit

This policy setting allows you to configure the algorithm and cipher strength used by BitLocker Drive Encryption. This policy setting is applied when you turn on BitLocker. Changing the encryption method has no effect if the drive is already encrypted, or if encryption is in progress.

If you enable this policy setting you will be able to configure an encryption algorithm and key cipher strength for fixed data drives, operating system drives, and removable data drives individually. For fixed and operating system drives, we recommend that you use the XTS-AES algorithm. For removable drives, you should use AES-CBC 128-bit or AES-CBC 256-bit if the drive will be used in other devices that are not running Windows 10 (Version 1511).

If you disable or do not configure this policy setting, BitLocker will use AES with the same bit strength (128-bit or 256-bit) as the "Choose drive encryption method and cipher strength (Windows

NOTE The default is set to XTS-AES 128-bit.

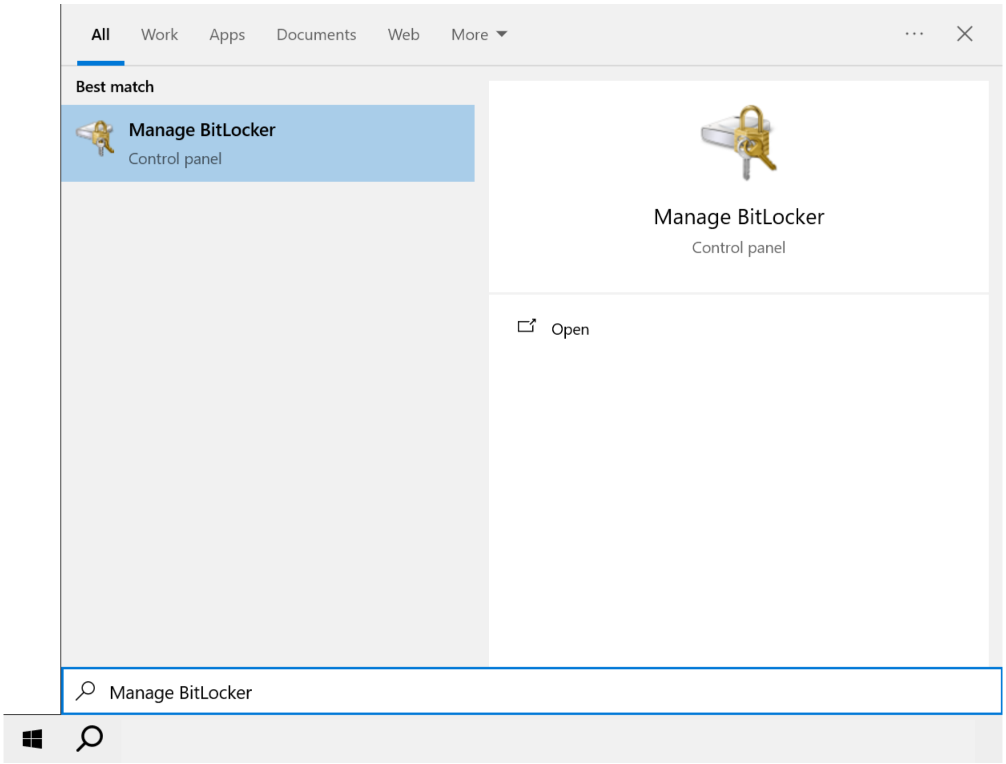
6 Select **Apply**.

7 Select **OK** and close all windows.

8 Restart the computer and proceed to Enabling BitLocker.

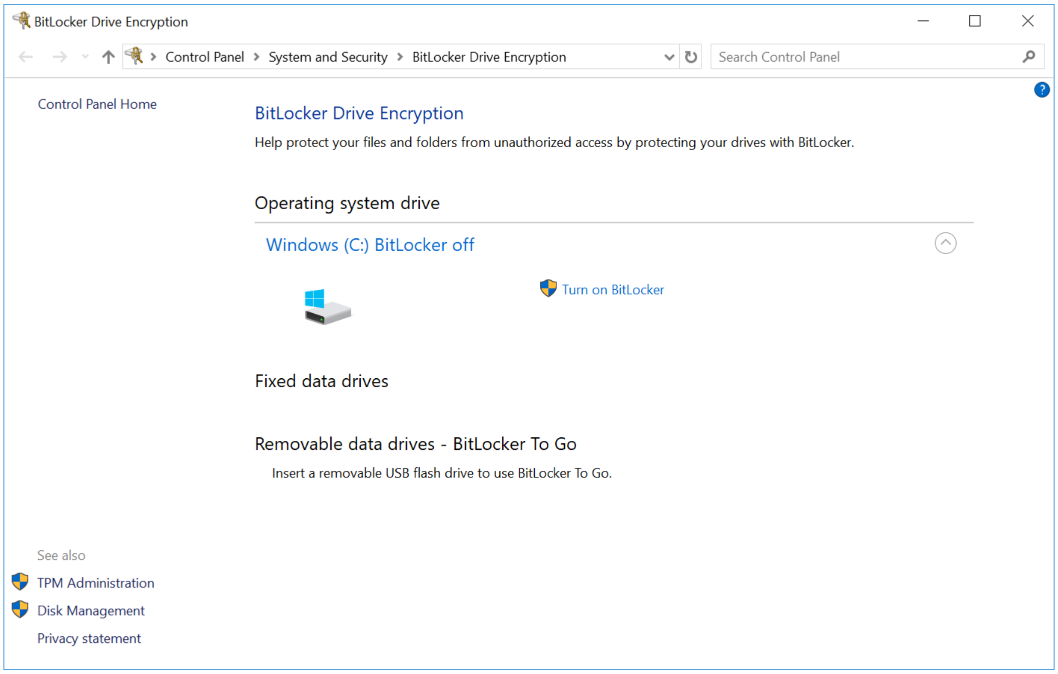
Enabling BitLocker

1 Type **Manage BitLocker** from the Windows search bar to locate the **Manage BitLocker Control Panel.**



---

2 Double-click **Manage BitLocker** to open the BitLocker Drive Encryption window.



BitLocker Drive Encryption

Control Panel > System and Security > BitLocker Drive Encryption

Search Control Panel

Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

Operating system drive

Windows (C:) BitLocker off

Turn on BitLocker

Fixed data drives

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

See also

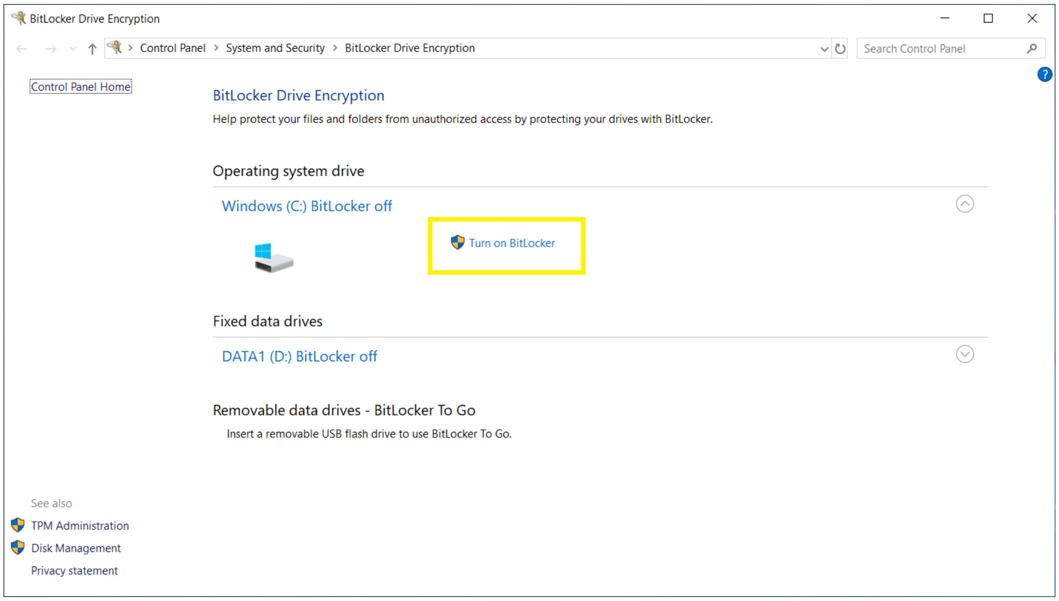
TPM Administration

Disk Management

Privacy statement

---

3 Select **Turn on BitLocker** from the Control Panel screen.



BitLocker Drive Encryption

Control Panel > System and Security > BitLocker Drive Encryption

Search Control Panel

Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

Operating system drive

Windows (C:) BitLocker off

Turn on BitLocker

Fixed data drives

DATA1 (D:) BitLocker off

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

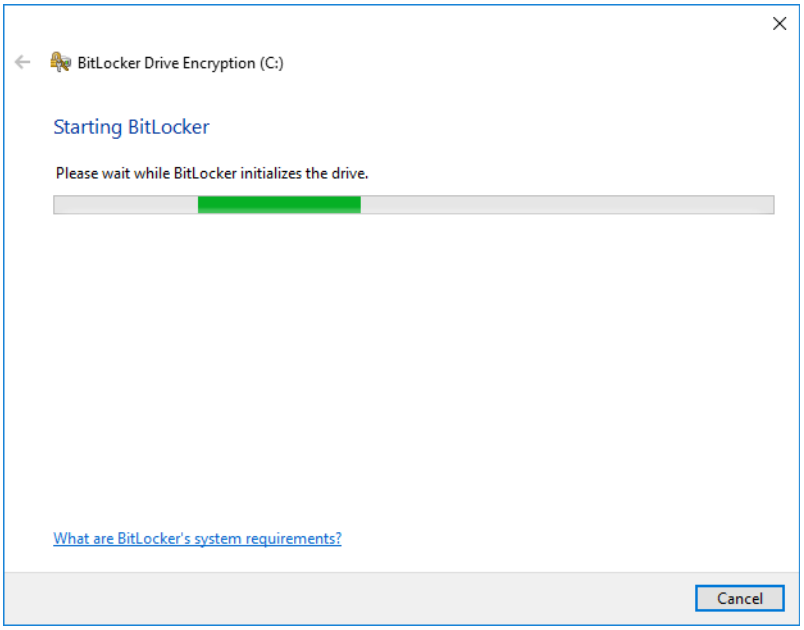
See also

TPM Administration

Disk Management

Privacy statement

4 BitLocker verifies that your PC meets the requirements.



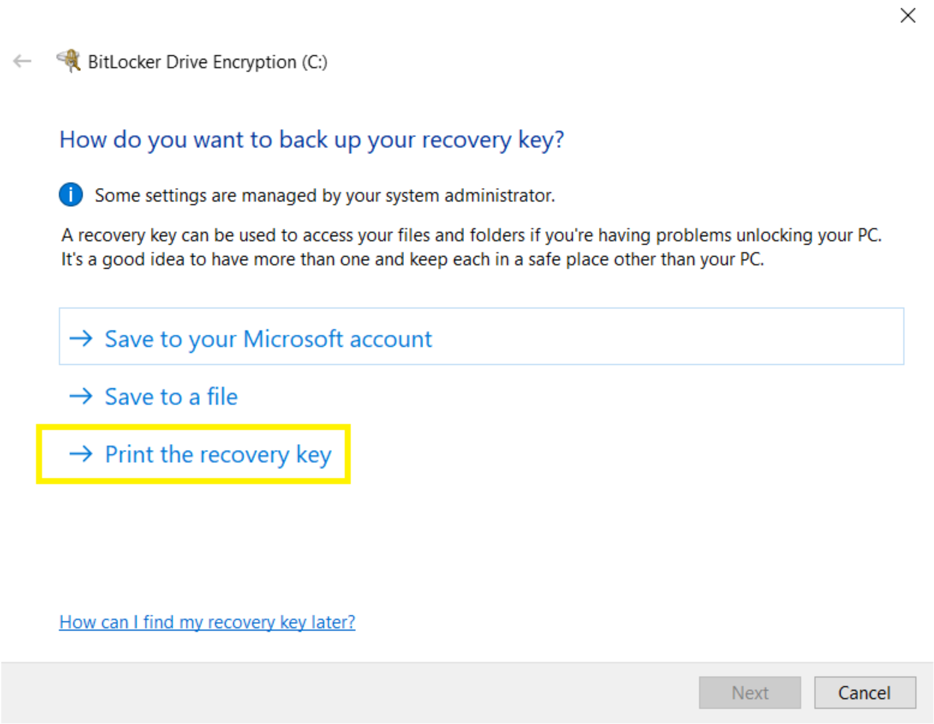
BitLocker Drive Encryption (C:)

Starting BitLocker

Please wait while BitLocker initializes the drive.

What are BitLocker's system requirements?
Cancel

5 Select **Print the recovery key** to back up the recovery key. For instructions, refer to Backing Up Your Recovery Key.



BitLocker Drive Encryption (C:)

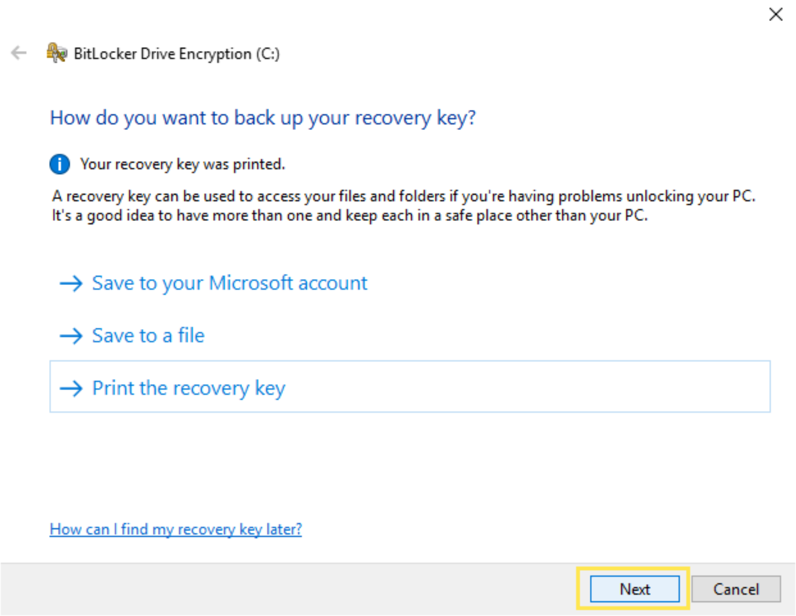
How do you want to back up your recovery key?

Some settings are managed by your system administrator.

A recovery key can be used to access your files and folders if you're having problems unlocking your PC. It's a good idea to have more than one and keep each in a safe place other than your PC.

→ Save to your Microsoft account
→ Save to a file
→ Print the recovery key
How can I find my recovery key later?
Next
Cancel

6 Select **Next.**



BitLocker Drive Encryption (C:)

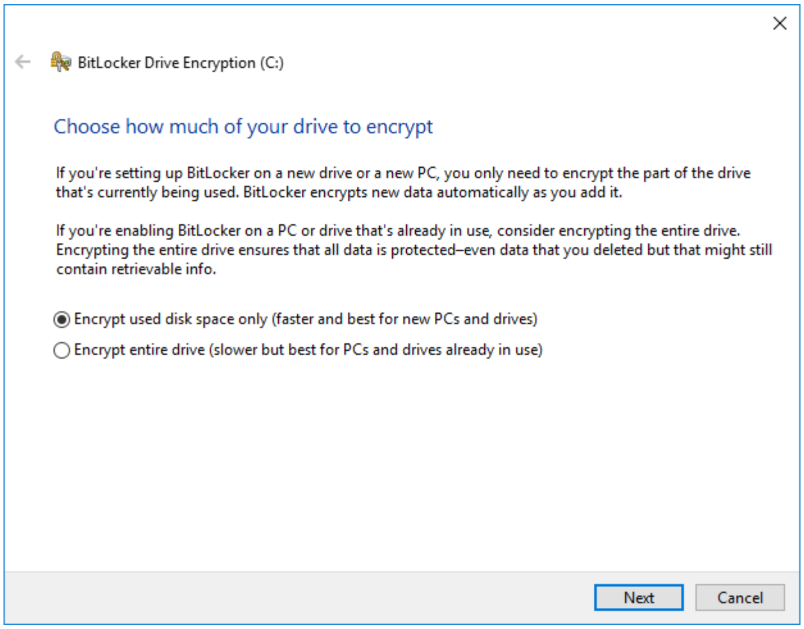
How do you want to back up your recovery key?

Your recovery key was printed.

A recovery key can be used to access your files and folders if you're having problems unlocking your PC. It's a good idea to have more than one and keep each in a safe place other than your PC.

→ Save to your Microsoft account
→ Save to a file
→ Print the recovery key
How can I find my recovery key later?
Next
Cancel

7 Select *"Encrypt used disk space only (faster and best for new PCs and drives)"*, then select **Next.**



BitLocker Drive Encryption (C:)

Choose how much of your drive to encrypt

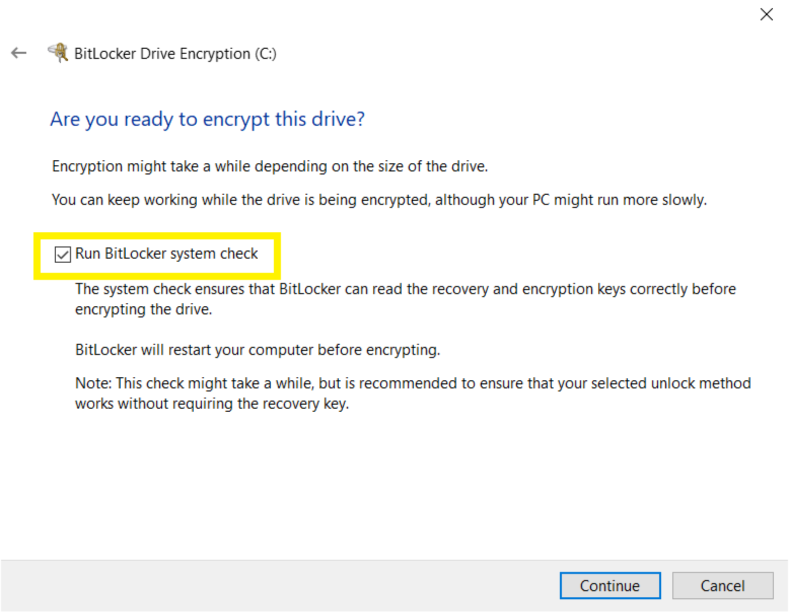
If you're setting up BitLocker on a new drive or a new PC, you only need to encrypt the part of the drive that's currently being used. BitLocker encrypts new data automatically as you add it.

If you're enabling BitLocker on a PC or drive that's already in use, consider encrypting the entire drive. Encrypting the entire drive ensures that all data is protected–even data that you deleted but that might still contain retrievable info.

● Encrypt used disk space only (faster and best for new PCs and drives)
○ Encrypt entire drive (slower but best for PCs and drives already in use)

IMPORTANT The system check ensures that BitLocker can read the recovery and encryption keys correctly before encrypting the drive. This check might take some time, but it is recommended to ensure that your selected unlock method works without requiring the recovery key.

8 Select *"Run BitLocker system check"*, then select **Start encrypting**.



Are you ready to encrypt this drive?

Encryption might take a while depending on the size of the drive.

You can keep working while the drive is being encrypted, although your PC might run more slowly.

☑ Run BitLocker system check

The system check ensures that BitLocker can read the recovery and encryption keys correctly before encrypting the drive.

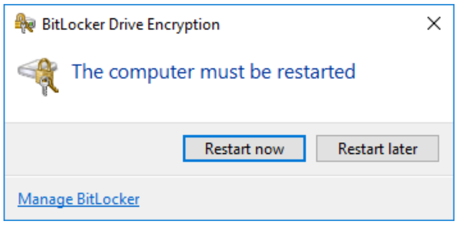
BitLocker will restart your computer before encrypting.

Note: This check might take a while, but is recommended to ensure that your selected unlock method works without requiring the recovery key.

NOTE BitLocker restarts your computer before encrypting.

---

9 Select **Restart now** to start the BitLocker Encryption.



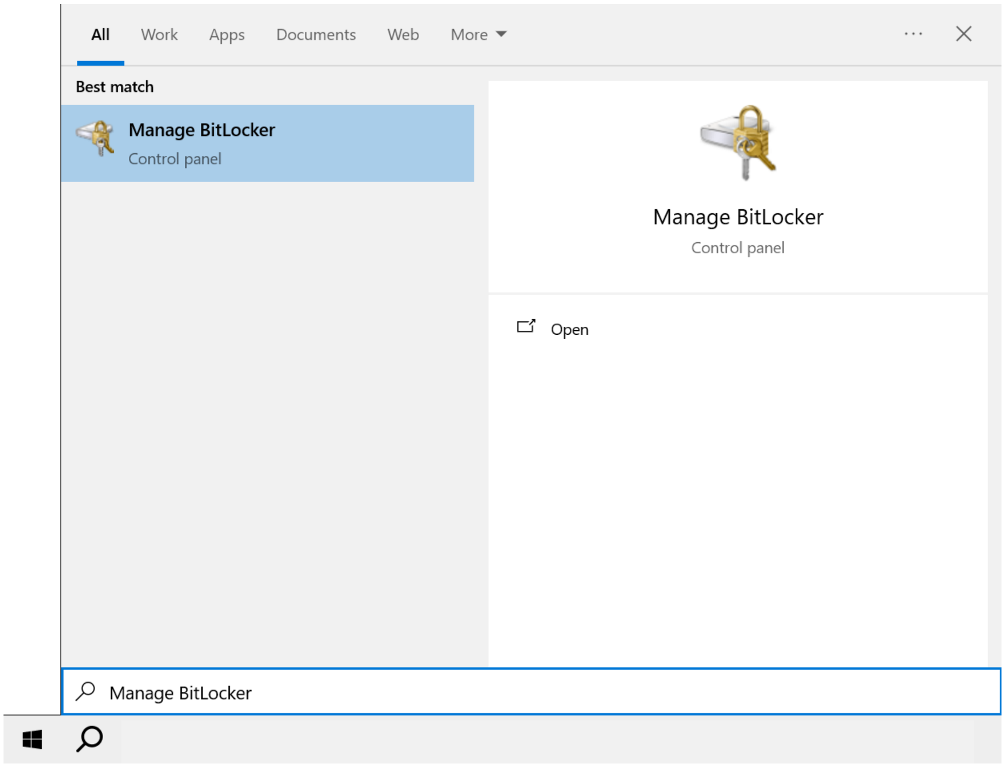
The computer must be restarted

---

10 Enter your password in the Password screen.

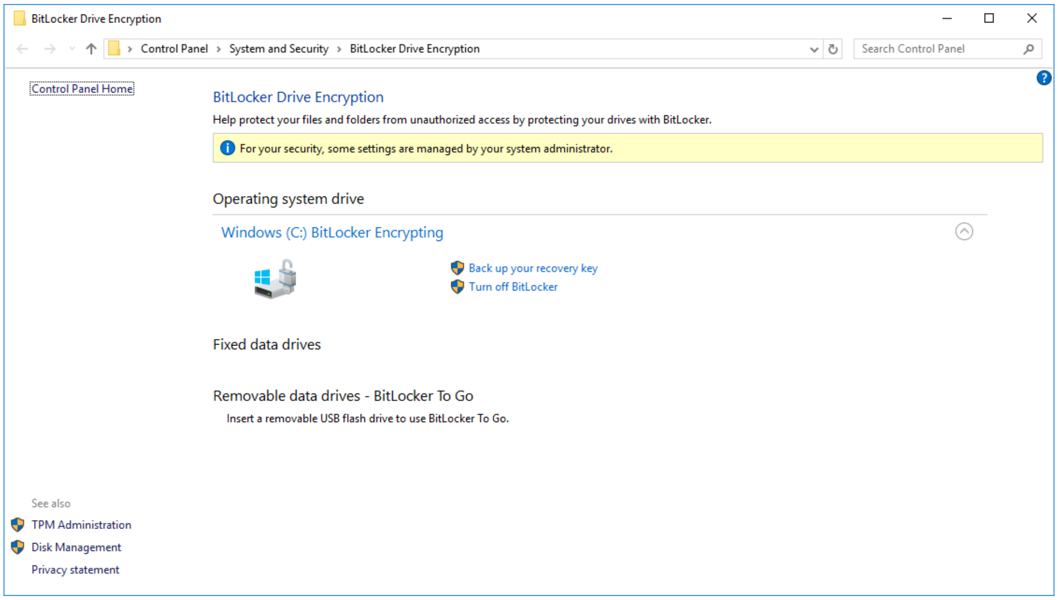
NOTE After logging in you will see a Window that says **Encrypting in Progress**.

11 Open the **Manage BitLocker Control Panel** from the Windows search bar.



Manage BitLocker

12 Under Operating System Drive, you will see the BitLocker is Encrypting the drive.


BitLocker Drive Encryption
Control Panel › System and Security › BitLocker Drive Encryption
Search Control Panel
Control Panel Home
Back up your recovery key
Turn off BitLocker
See also
TPM Administration
Disk Management
Privacy statement

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive

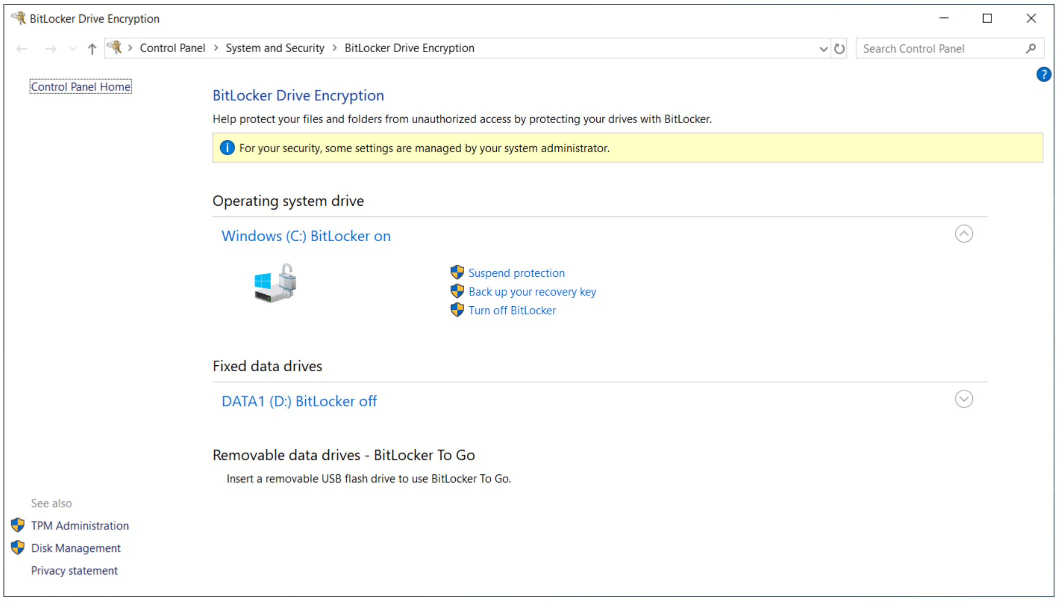
Windows (C:) BitLocker Encrypting

Fixed data drives

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

13 Once the BitLocker has encrypted the drive, the BitLocker will be on.


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive

Windows (C:) BitLocker on

- Suspend protection
- Back up your recovery key
- Turn off BitLocker

Fixed data drives

DATA1 (D:) BitLocker off

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

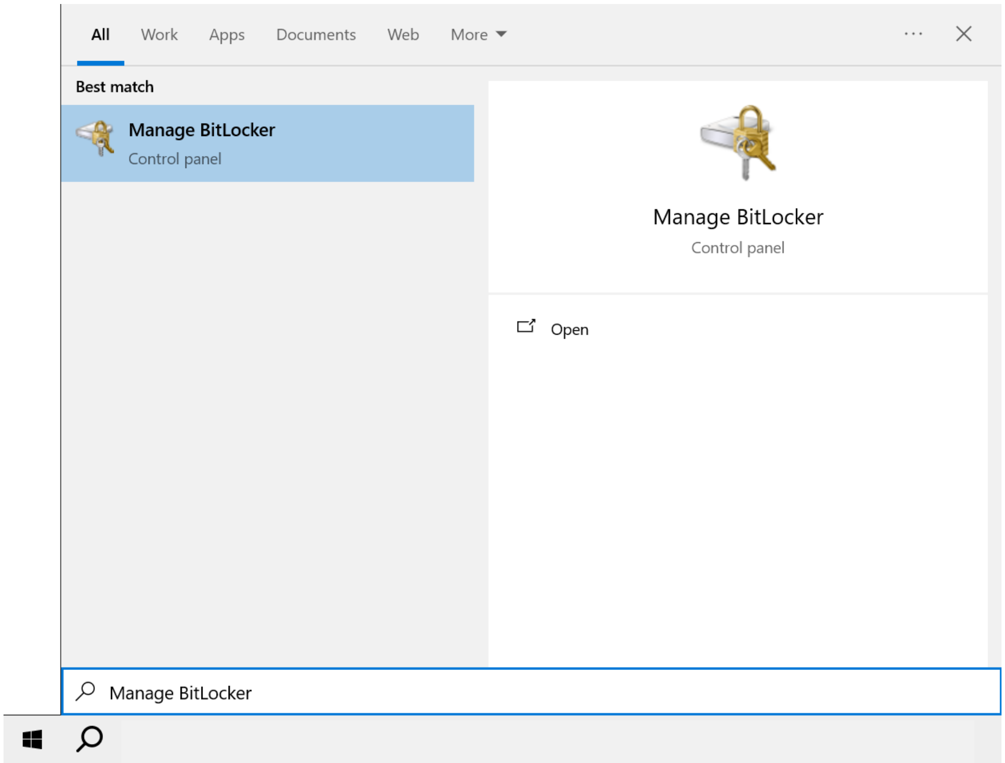
Backing Up Your Recovery Key

IMPORTANT The Back Up Your Recovery Key to your Microsoft Account is not supported at this time.

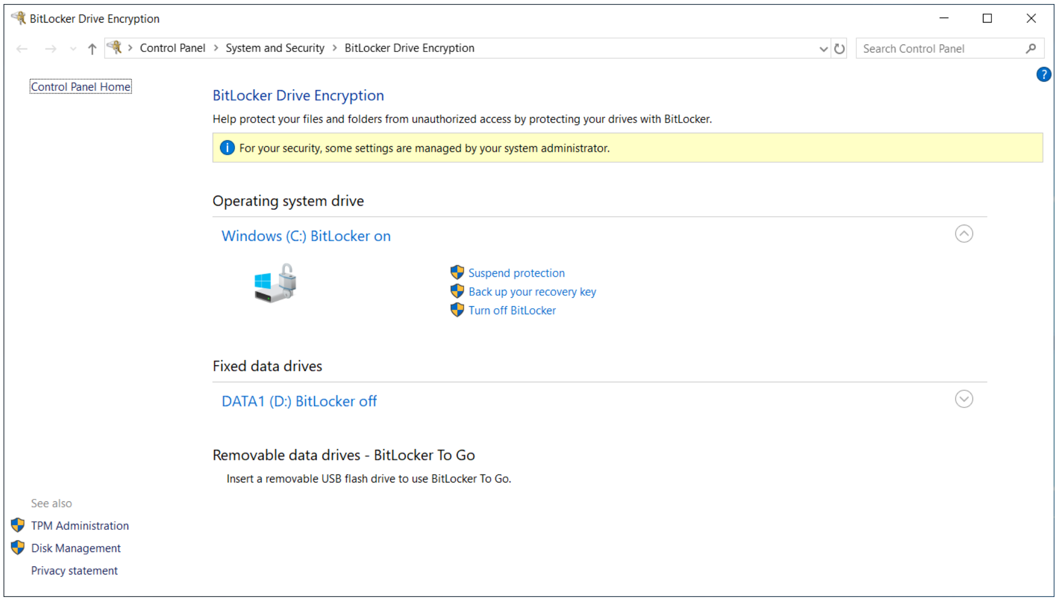
IMPORTANT The Recovery Key can only be saved to a USB or external drive. You cannot save to a location on your PC.

---

1 Type **Manage BitLocker** from the Windows search bar to locate the **Manage BitLocker Control Panel.**



2 Double-click **Manage BitLocker** to open the BitLocker Drive Encryption window.


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive

Windows (C:) BitLocker on

Suspend protection
Back up your recovery key
Turn off BitLocker

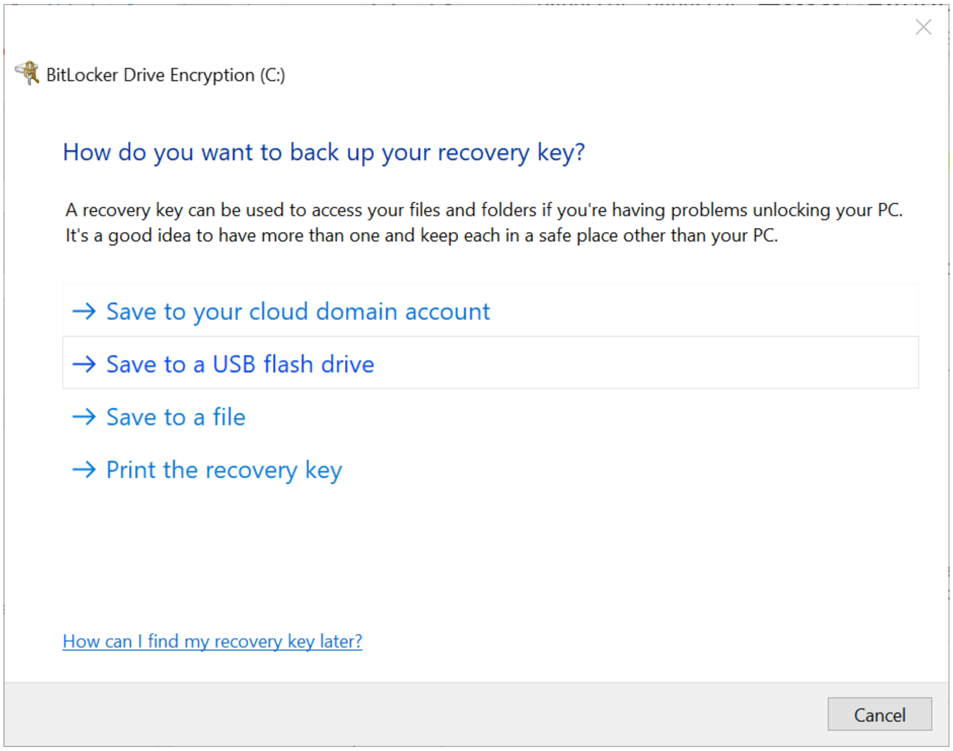
Fixed data drives

DATA1 (D:) BitLocker off

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

3 Select *"Backup your recovery key"*. The following window appears.


BitLocker Drive Encryption (C:)

How do you want to back up your recovery key?

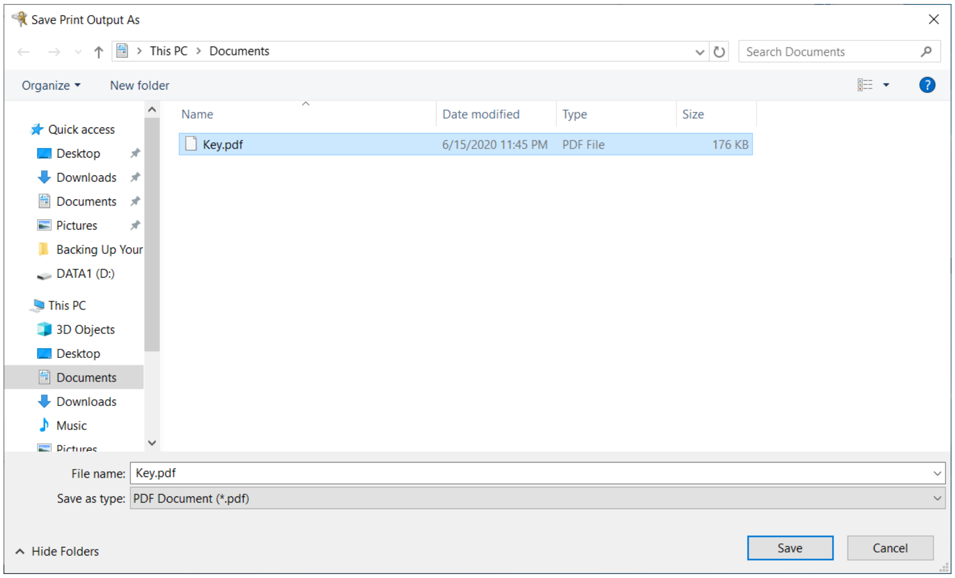
A recovery key can be used to access your files and folders if you're having problems unlocking your PC. It's a good idea to have more than one and keep each in a safe place other than your PC.

→ Save to your cloud domain account
→ Save to a USB flash drive
→ Save to a file
→ Print the recovery key
How can I find my recovery key later?
Cancel

IMPORTANT The Recovery Key can only be saved to a USB or external drive. You cannot save to a location on your PC.

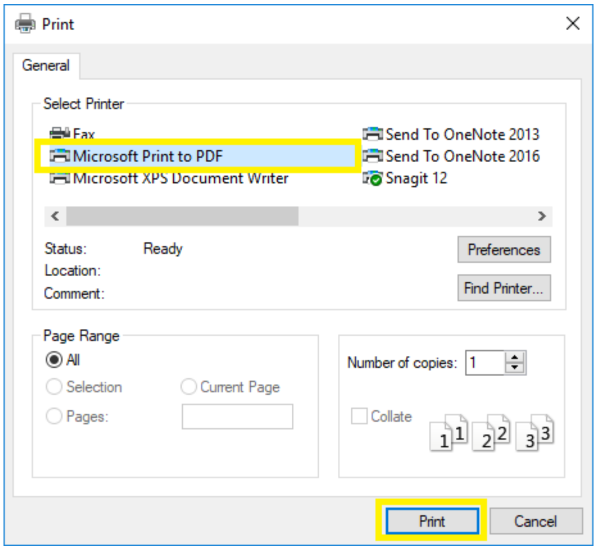
4 Select a destination to save the Recovery Key.

- **Save to a file**: Select "*Save to a file*". The following screen appears.



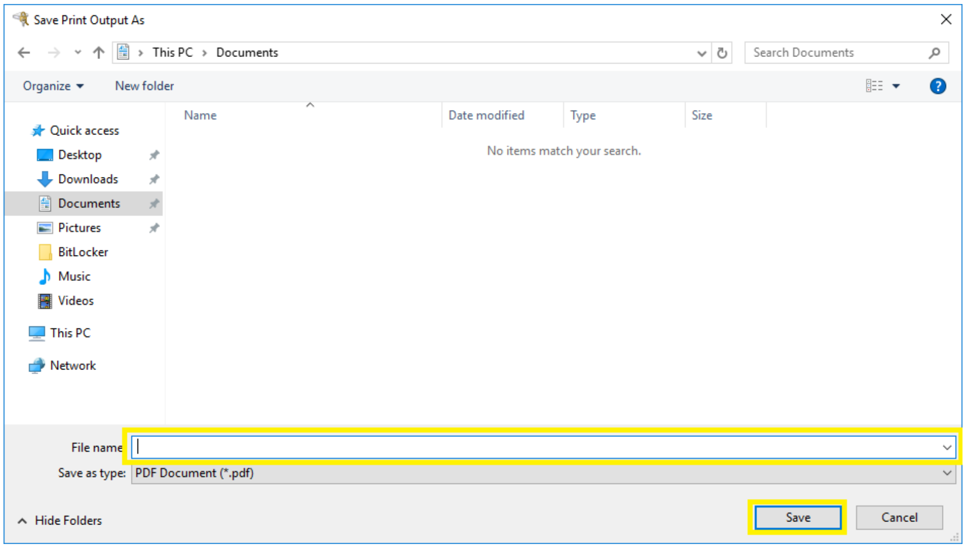
File name: Key.pdf
Save as type: PDF Document (*.pdf)

- **Print the recovery key**: Select "*Print the recovery key*". The following screen appears.



Status: Ready
Location:
Comment:
Number of copies: 1

1. Select "*Microsoft Print to PDF*", and then select **Print**.



No items match your search.

File name:
Save as type: PDF Document (*.pdf)

2. Select the location that you want the file saved. In the File name, type **BitLocker Key** and in the Save as type, ensure that it is listed as .pdf.

---

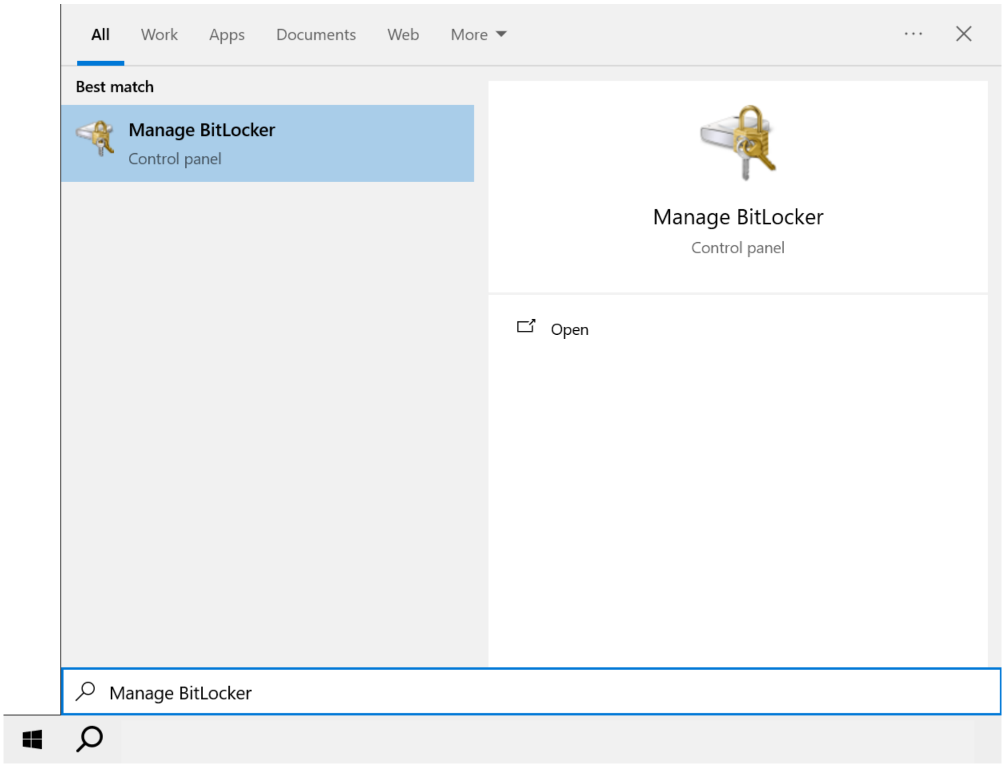
5 Select **Save** then select **Finish**.

BitLocker Decryption

IMPORTANT Turning off the BitLocker feature decrypts all the data on the drive. Decrypting your operating system means that you have removed BitLocker from your system. The BitLocker key will change each time BitLocker is re-enabled. Only the latest BitLocker key will work. Previous BitLocker keys will be invalid and will not unlock the system. Please note this is a time-consuming process. However, you can also suspend BitLocker instead of decrypting the drive, see Suspending BitLocker Drive Encryption.

Turning off BitLocker

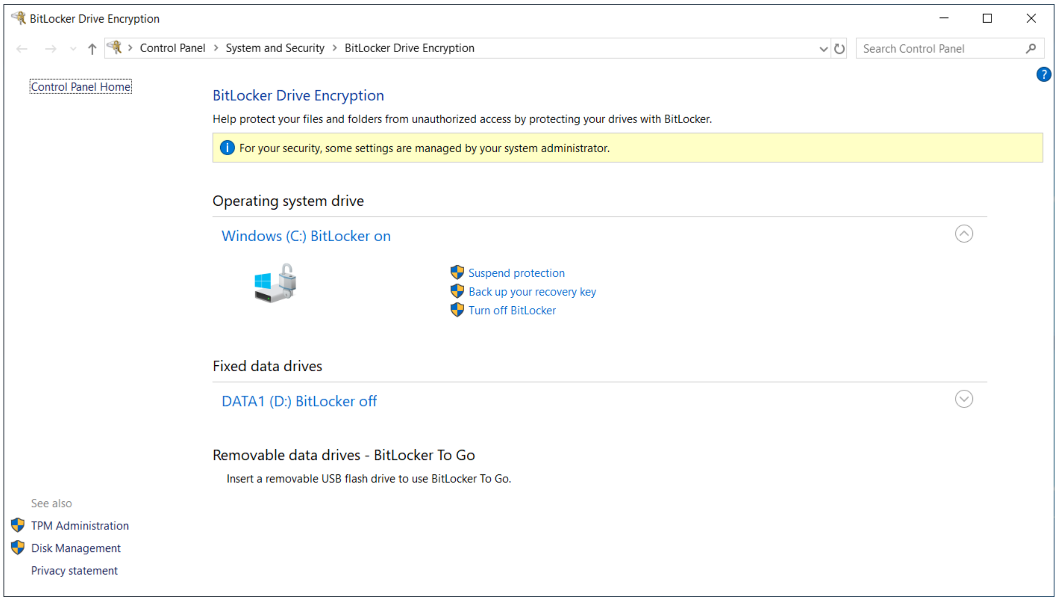
1 Type **Manage BitLocker** from the Windows search bar to locate the **Manage BitLocker Control Panel**.



Manage BitLocker

---

2 Double-click **Manage BitLocker** to open the BitLocker Drive Encryption.


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home
BitLocker Drive Encryption
Operating system drive
Windows (C:) BitLocker on
Suspend protection
Back up your recovery key
Turn off BitLocker
Fixed data drives
DATA1 (D:) BitLocker off
Removable data drives - BitLocker To Go
See also
TPM Administration
Disk Management
Privacy statement

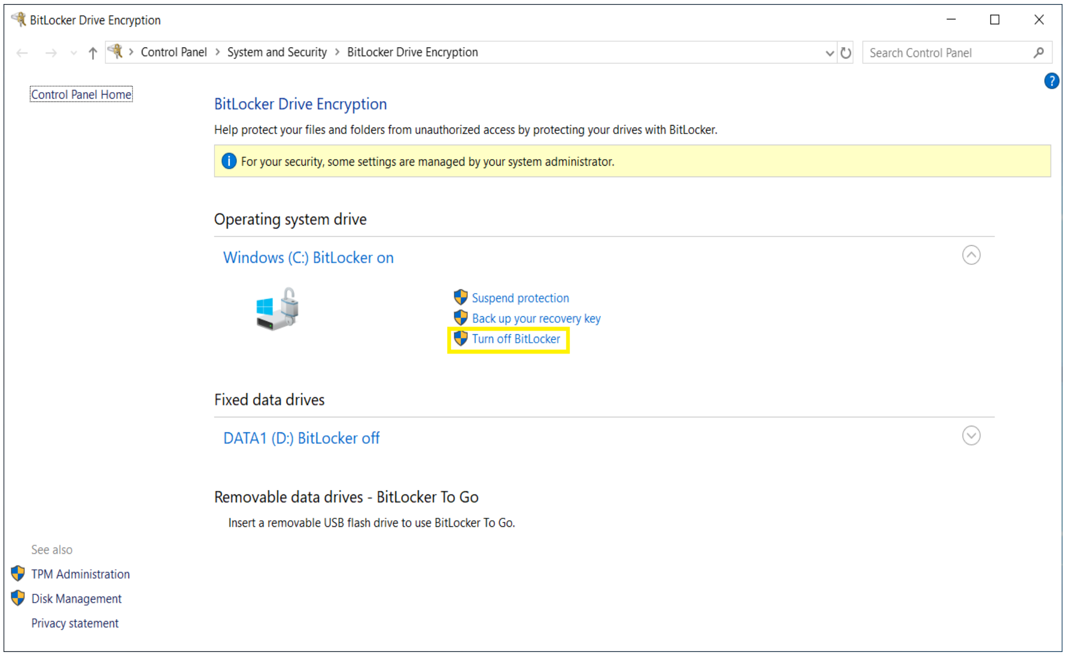
Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Insert a removable USB flash drive to use BitLocker To Go.

---

3 Select **Turn off BitLocker**.


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home
BitLocker Drive Encryption
Operating system drive
Windows (C:) BitLocker on
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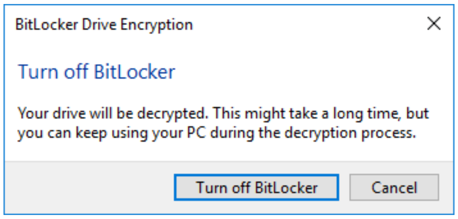
See also
TPM Administration
Disk Management
Privacy statement

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Insert a removable USB flash drive to use BitLocker To Go.

The following message appears.



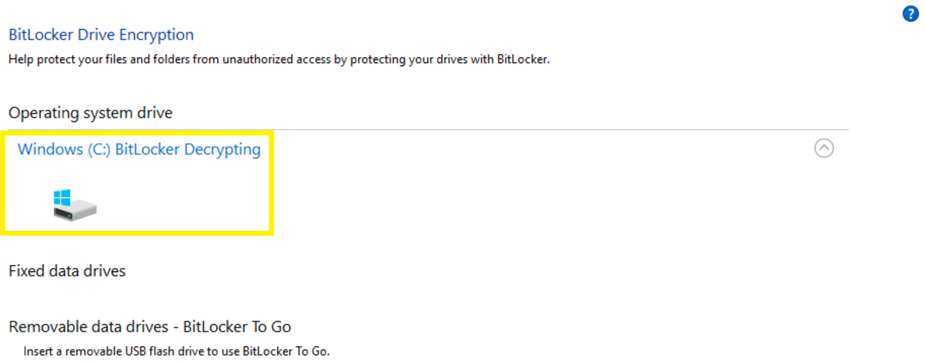
BitLocker Drive Encryption

Turn off BitLocker

Your drive will be decrypted. This might take a long time, but you can keep using your PC during the decryption process.

IMPORTANT Drive decryption might take a long time, but you can keep using your PC during the decryption process.

4 Select **Turn Off BitLocker**. The BitLocker Drive Decryption screen displays.



BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

Operating system drive

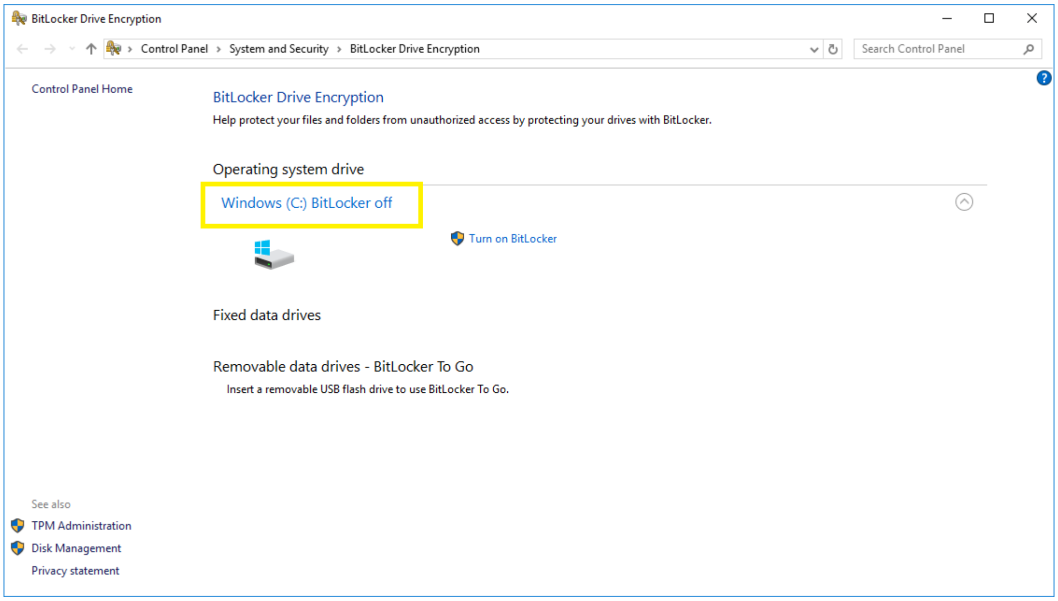
Windows (C:) BitLocker Decrypting

Fixed data drives

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

After the decryption has completed, the following screen displays.


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

Operating system drive

Windows (C:) BitLocker off

Turn on BitLocker

Fixed data drives

Removable data drives - BitLocker To Go

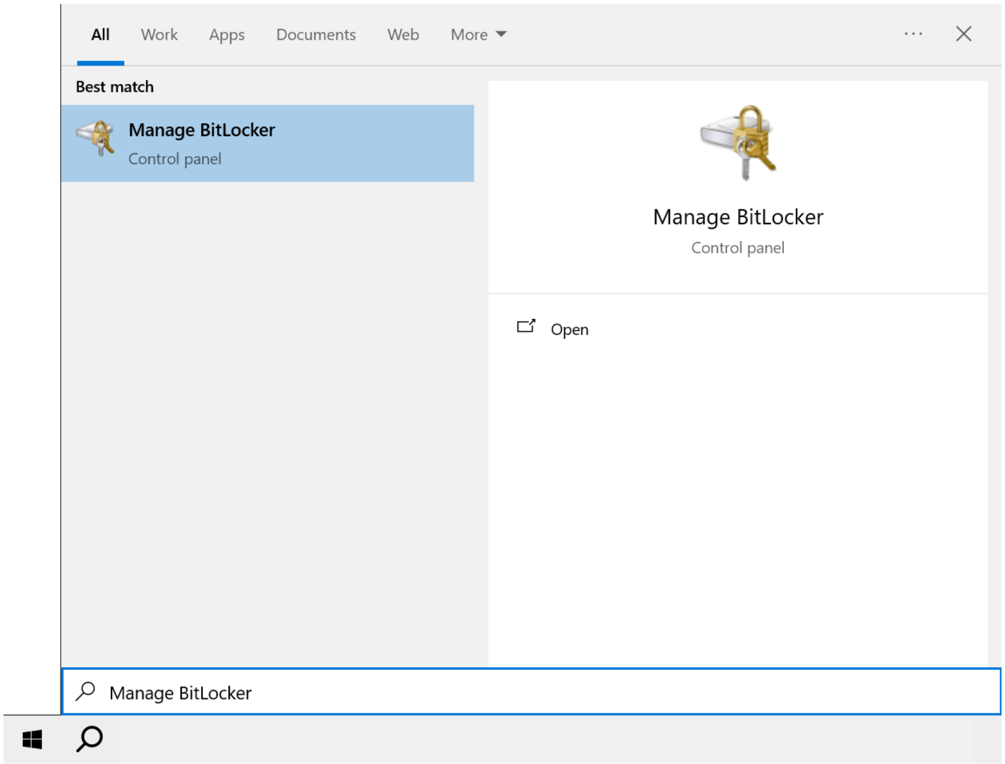
Insert a removable USB flash drive to use BitLocker To Go.

Suspending BitLocker Drive Encryption

IMPORTANT Suspending BitLocker Drive Encryption is a temporary method for removing BitLocker protection without decrypting the Windows drive. Use Suspend BitLocker only if you need to update the BIOS firmware or Windows boot loader/startup files that requires access to the drive in a pre-boot environment. This will help prevent BitLocker from locking the drive and can avoid a lengthy decryption process.

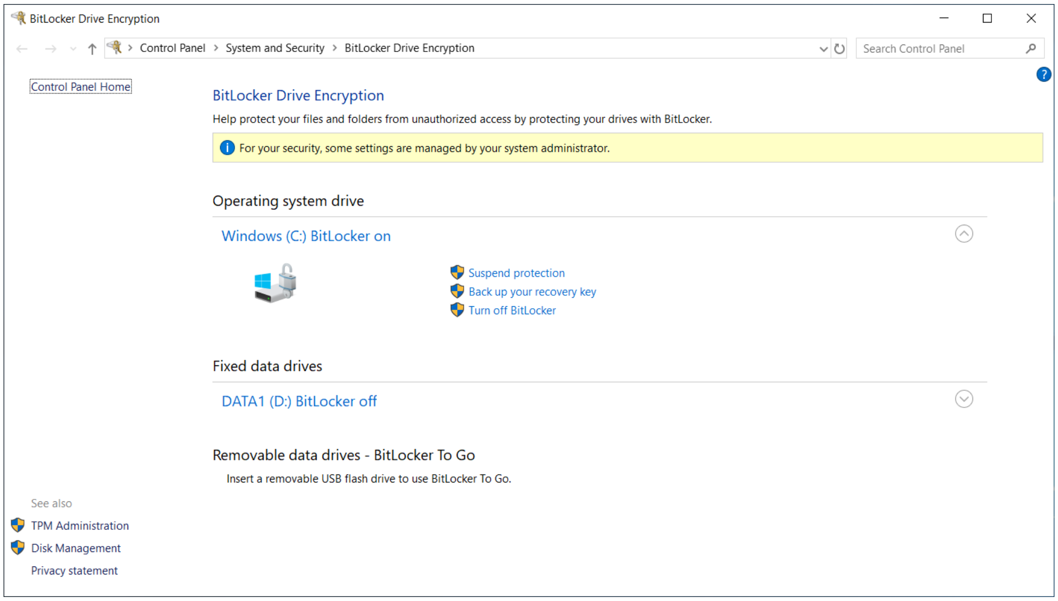
Suspending BitLocker Protection

1 Type **Manage BitLocker** from the Windows search bar to locate the **Manage BitLocker Control Panel**.



Manage BitLocker

2 Double-click **Manage BitLocker** to open the BitLocker Drive Encryption window.


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive

Windows (C:) BitLocker on

Suspend protection
Back up your recovery key
Turn off BitLocker

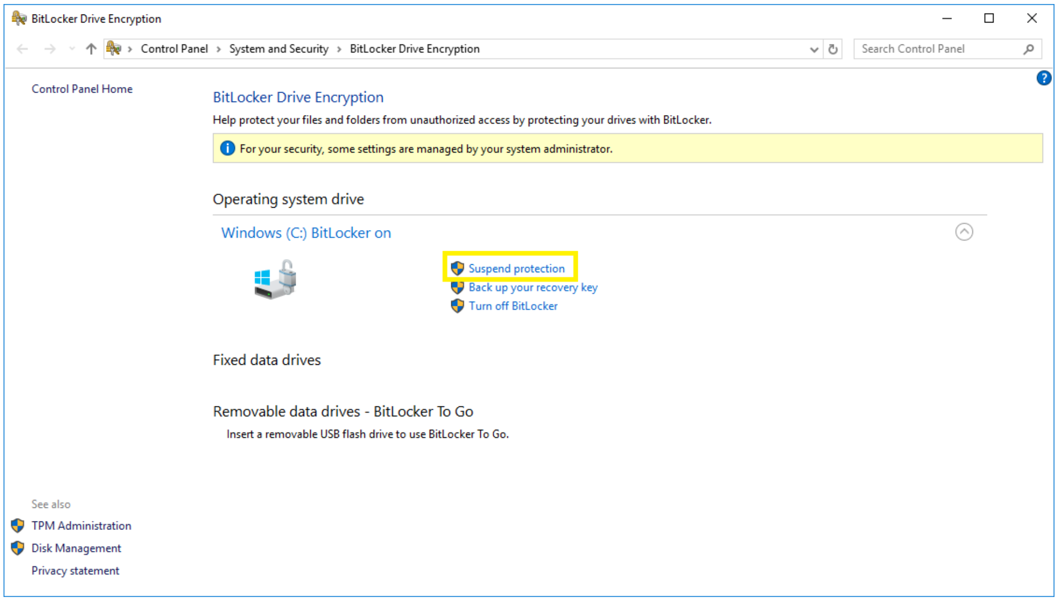
Fixed data drives

DATA1 (D:) BitLocker off

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

3 Select **Suspend Protection.**


BitLocker Drive Encryption
Control Panel > System and Security > BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive

Windows (C:) BitLocker on

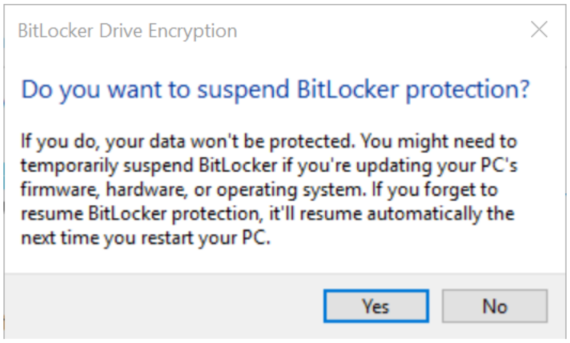
Suspend protection
Back up your recovery key
Turn off BitLocker

Fixed data drives

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

A confirmation message “*Do you want to suspend BitLocker protection?*” displays.



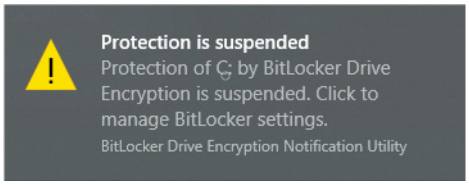
BitLocker Drive Encryption

Do you want to suspend BitLocker protection?

If you do, your data won't be protected. You might need to temporarily suspend BitLocker if you're updating your PC's firmware, hardware, or operating system. If you forget to resume BitLocker protection, it'll resume automatically the next time you restart your PC.

IMPORTANT If you choose to suspend BitLocker, your data will not be protected.

4 If you want to proceed, select **Yes** and the following popup information will display.



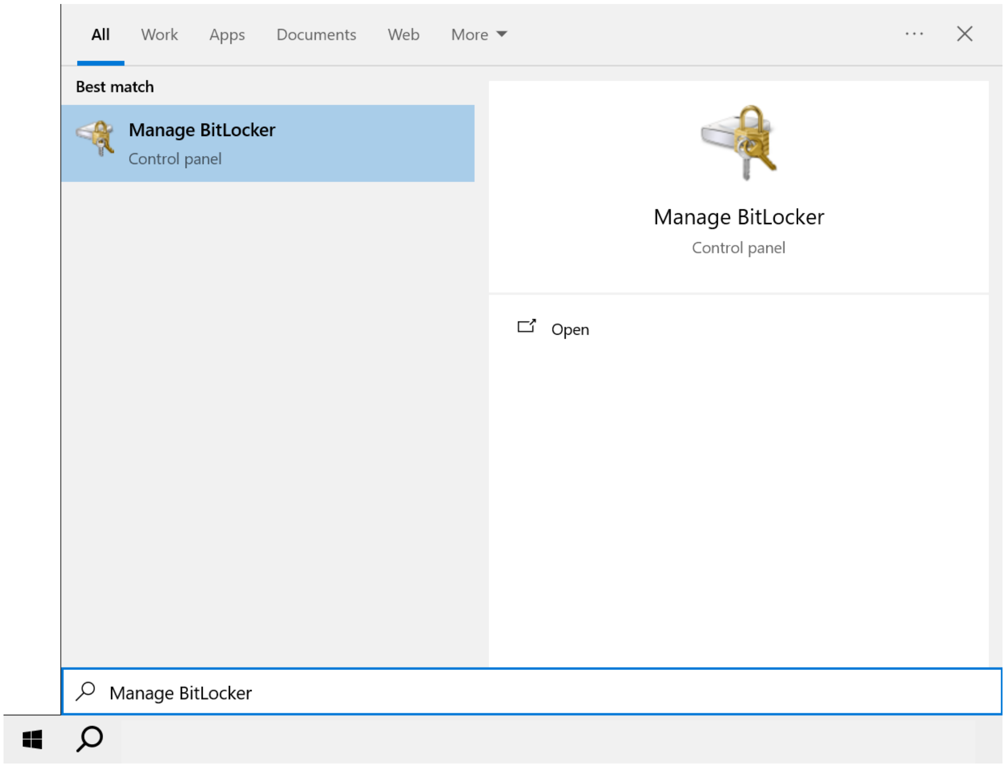
Protection is suspended
Protection of C: by BitLocker Drive Encryption is suspended. Click to manage BitLocker settings.
BitLocker Drive Encryption Notification Utility

If you do not want to proceed, select **No**.

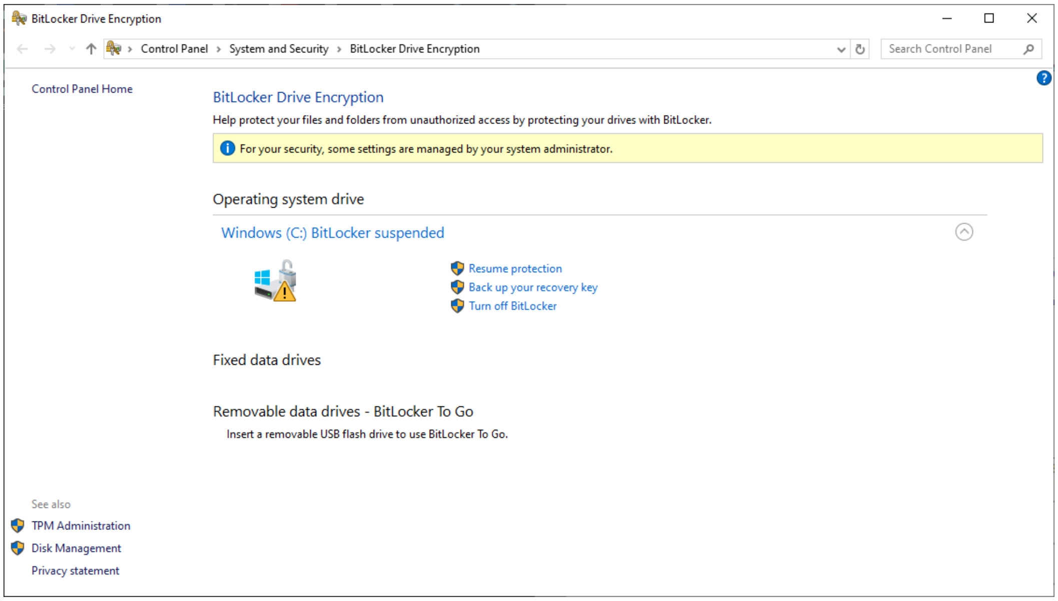
NOTE There are certain situations where you may want to suspend BitLocker such as updating your PC’s firmware, hardware, or operating system. If you forget to resume BitLocker after these updates are completed, BitLocker will automatically resume the next time you restart your PC.

Restarting BitLocker Protection

1 Type **Manage BitLocker** from the Windows search bar to locate the **Manage BitLocker Control Panel.**



2 Double-click **Manage BitLocker** to open the BitLocker Drive Encryption window.


BitLocker Drive Encryption
Control Panel › System and Security › BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive

Windows (C:) BitLocker suspended

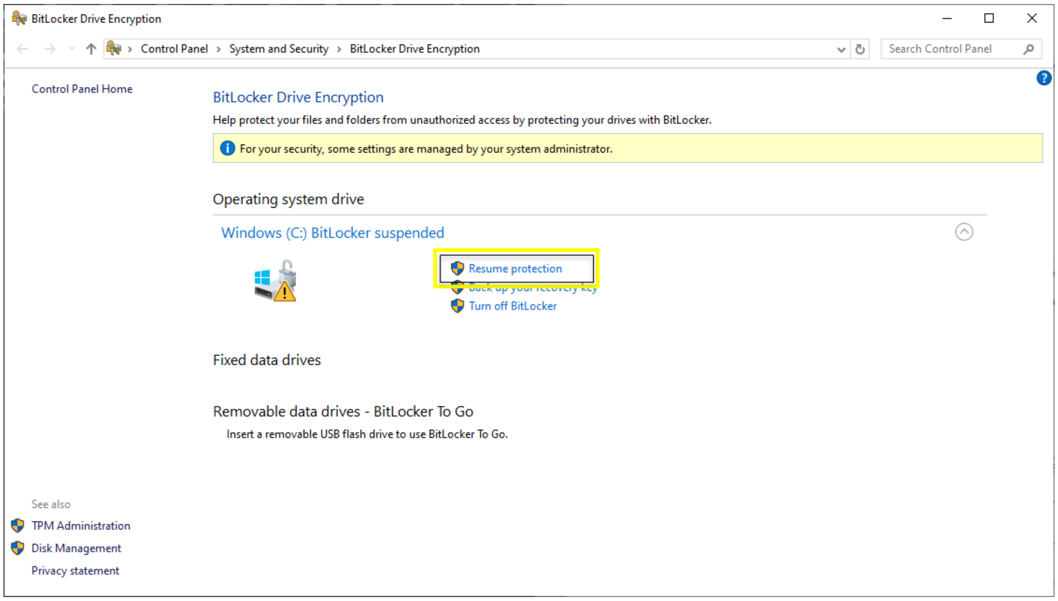
Resume protection
Back up your recovery key
Turn off BitLocker

Fixed data drives

Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

3 Select **Resume Protection**.



BitLocker Drive Encryption
Control Panel › System and Security › BitLocker Drive Encryption
Search Control Panel
Control Panel Home

BitLocker Drive Encryption

Help protect your files and folders from unauthorized access by protecting your drives with BitLocker.

For your security, some settings are managed by your system administrator.

Operating system drive
Windows (C:) BitLocker suspended
Resume protection
Back up your recovery key
Turn off BitLocker
Fixed data drives
Removable data drives - BitLocker To Go

Insert a removable USB flash drive to use BitLocker To Go.

See also
TPM Administration
Disk Management
Privacy statement

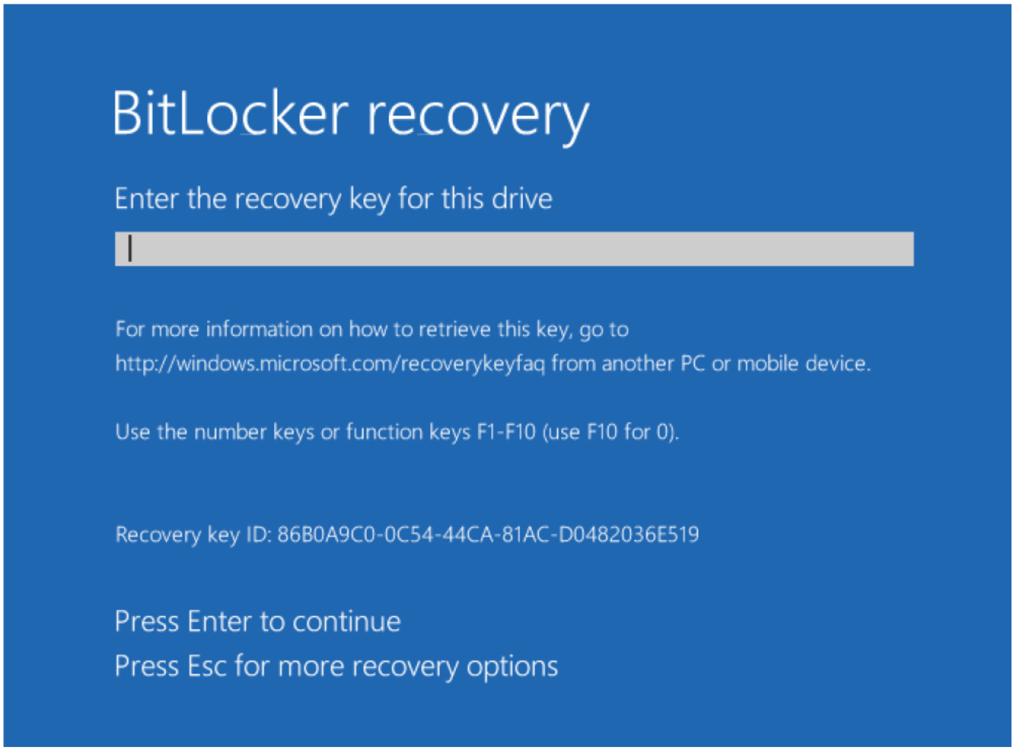
Recovering Your BitLocker Key

There might be situations where the BitLocker Key is requested from your computer, such as:

- Your drive is placed onto another computer
- Computer requests your recovery key; it may be due to one of the following but is not limited to:
  - Hardware changes
  - Firmware changes
  - Other Windows updates

BitLocker Recovery

1 BitLocker Recovery window will be displayed upon powering on your system.



BitLocker recovery

Enter the recovery key for this drive

For more information on how to retrieve this key, go to http://windows.microsoft.com/recoverykeyfaq from another PC or mobile device.

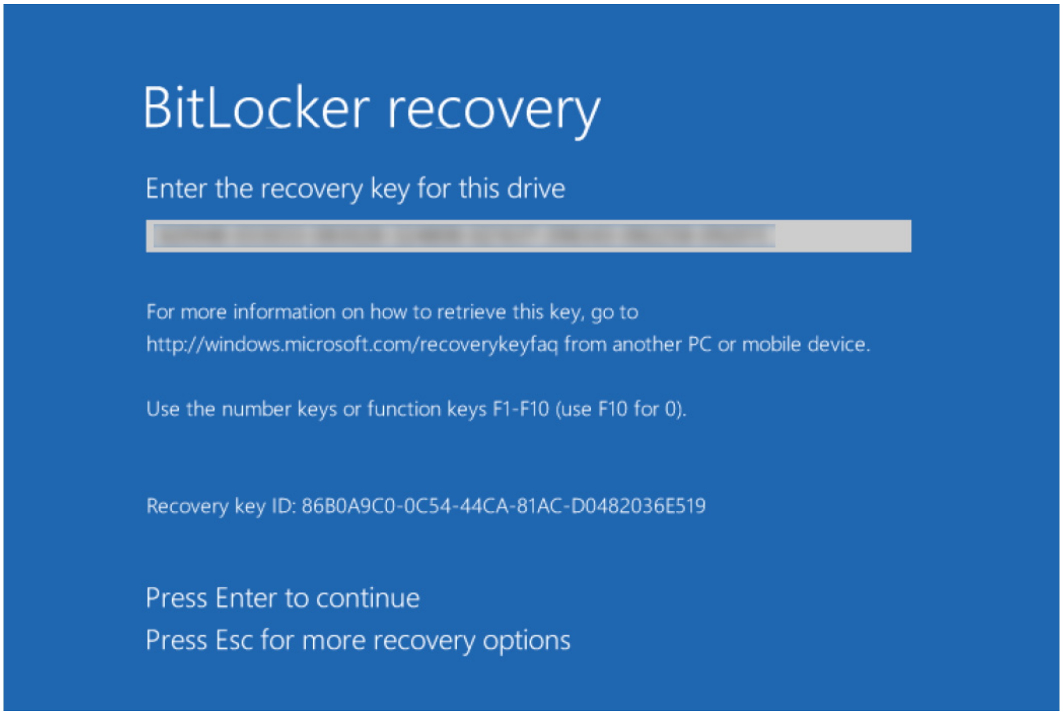
Use the number keys or function keys F1-F10 (use F10 for 0).

Recovery key ID: 86B0A9C0-0C54-44CA-81AC-D0482036E519

Press Enter to continue

Press Esc for more recovery options

2 Enter your key from your saved text file or printed copy, then select **Enter**.



BitLocker recovery

Enter the recovery key for this drive

[redacted]

For more information on how to retrieve this key, go to
http://windows.microsoft.com/recoverykeyfaq from another PC or mobile device.

Use the number keys or function keys F1-F10 (use F10 for 0).

Recovery key ID: 86B0A9C0-0C54-44CA-81AC-D0482036E519

Press Enter to continue
Press Esc for more recovery options

3 Windows will reboot your computer.

Protection from Malware Software

⚠ **CAUTION**

The instrument workstation will be vulnerable to malware, viruses, and unauthorized access if the instrument PC is connected directly to the laboratory network without installation of all approved OS and malware patches.

IMPORTANT Only the version of the Trellix® Application Control (Version 8.4.1) has been validated to work with the CytoFLEX instrument.

For more information on Configuration of Trellix Application Control, refer to Trellix Application Control Guide.

Installing Trellix Application Control

IMPORTANT The Trellix Application Control Software (Version 8.4.1), which has been proved compatible with the software, should be purchased separately and downloaded from Trellix.

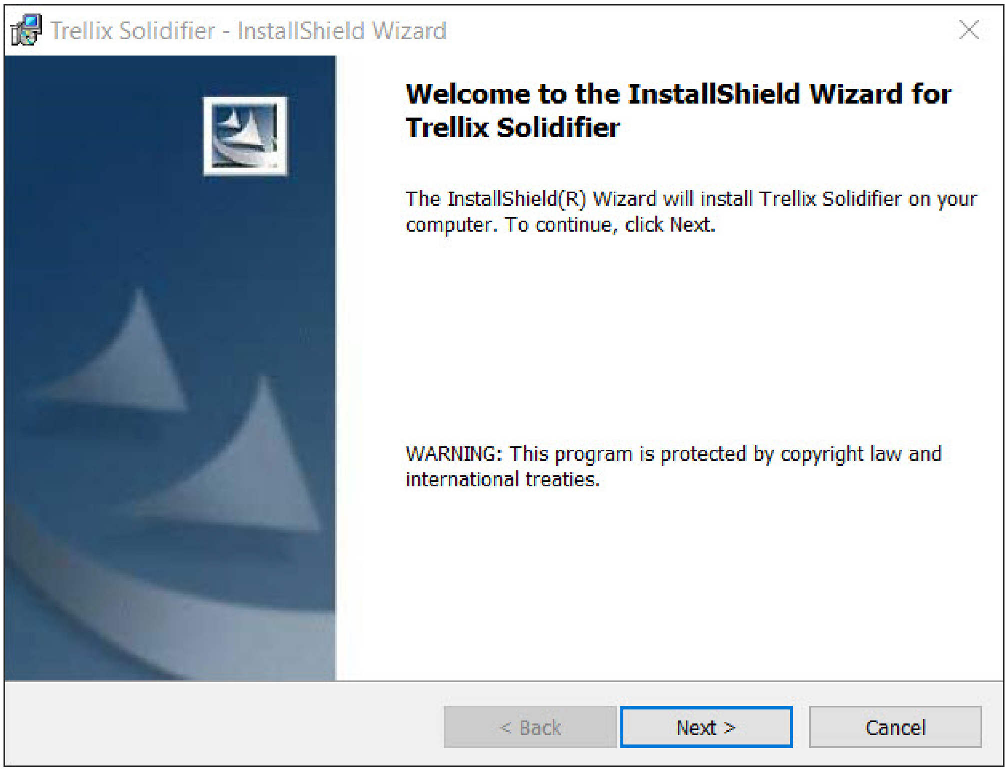
The following procedure can assist you to install the Trellix Application Control software. For more instructions, access the Trellix for help.

---

1 Unzip the Trellix Application Control file **SOLIDCORxxx-xxx_WIN.zip**

---

2 Right-click **Setup-win-8-10-2012-amd64-xxx.exe**, and select **Run as administrator**. The Trellix Installer opens.



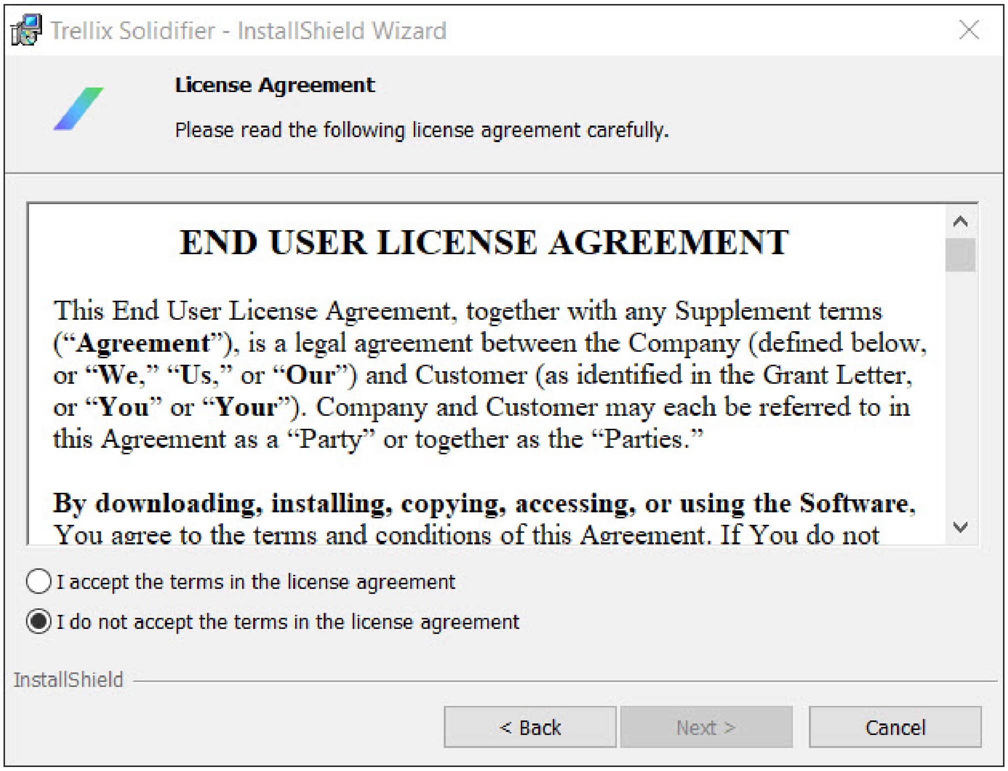
Trellix Solidifier - InstallShield Wizard

Welcome to the InstallShield Wizard for Trellix Solidifier

The InstallShield(R) Wizard will install Trellix Solidifier on your computer. To continue, click Next.

WARNING: This program is protected by copyright law and international treaties.

3 Select **Next**. The License Agreement displays.



License Agreement

Please read the following license agreement carefully.

END USER LICENSE AGREEMENT

This End User License Agreement, together with any Supplement terms (“**Agreement**”), is a legal agreement between the Company (defined below, or “**We**,” “**Us**,” or “**Our**”) and Customer (as identified in the Grant Letter, or “**You**” or “**Your**”). Company and Customer may each be referred to in this Agreement as a “Party” or together as the “Parties.”

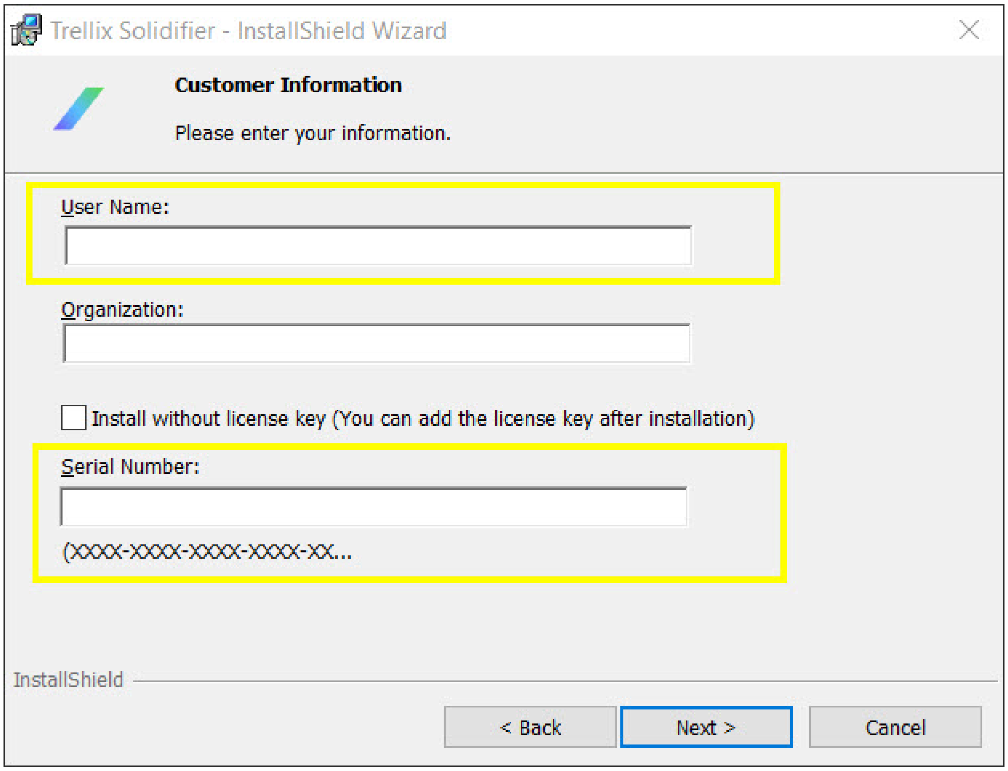
By downloading, installing, copying, accessing, or using the Software, You agree to the terms and conditions of this Agreement. If You do not

○ I accept the terms in the license agreement

● I do not accept the terms in the license agreement

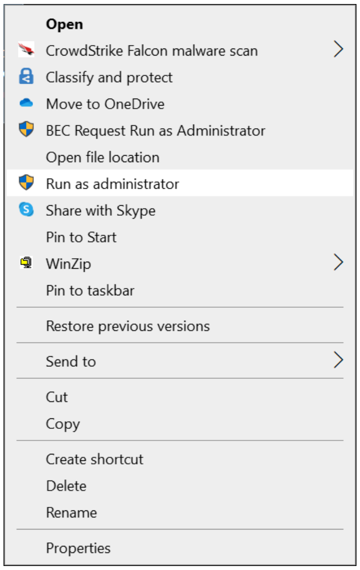
Read the license agreement and select **I accept the terms in the license agreement**, then select **Next.**

4 Enter the username and the serial number. Select **Next**.

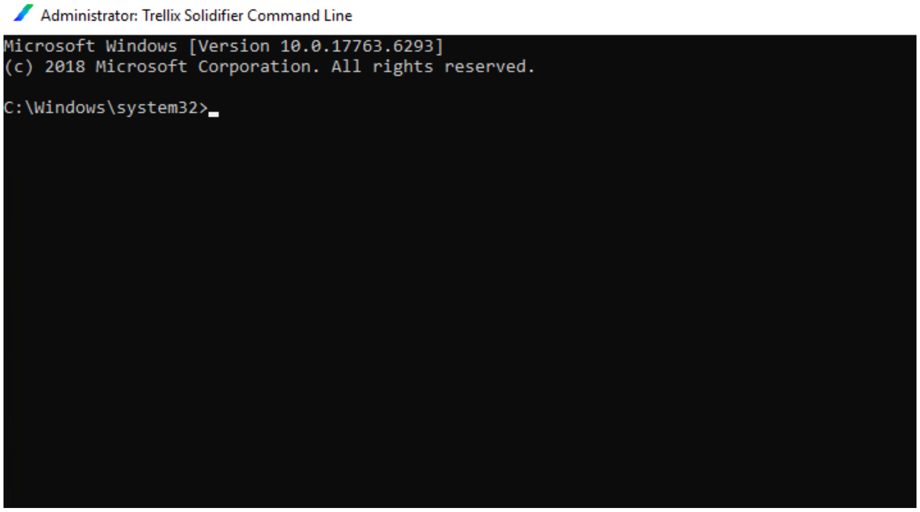

Trellix Solidifier - InstallShield Wizard
Customer Information
Please enter your information.
User Name:
Organization:
☐ Install without license key (You can add the license key after installation)
Serial Number:
(XXXX-XXXX-XXXX-XXXX-XX...
InstallShield
< Back
Next >
Cancel

If you select **Install without license key**, follow the procedures below to add the license key after the installation.

a. Right-click Trellix Solidifier... on your desktop, and select **Run as administrator**.

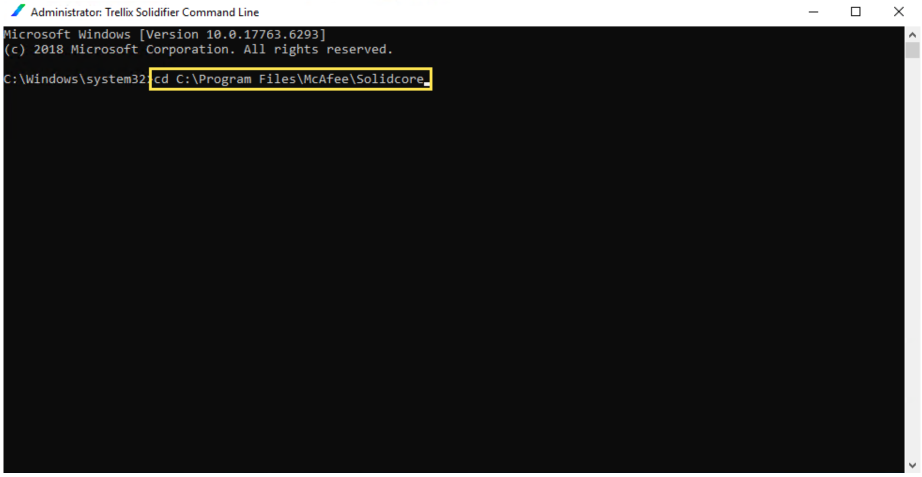


The following window displays.



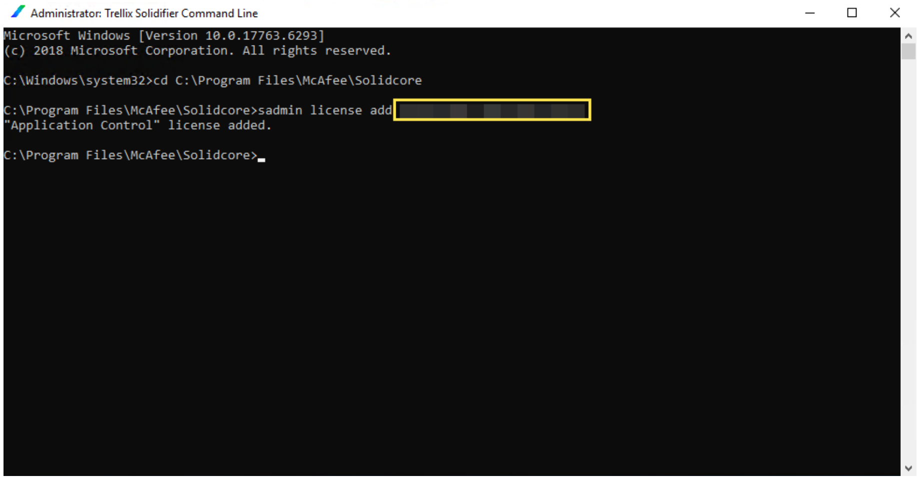
```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>
```

b. Enter **cd C:\Program Files\McAfee\Solidcore**.



```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore
```

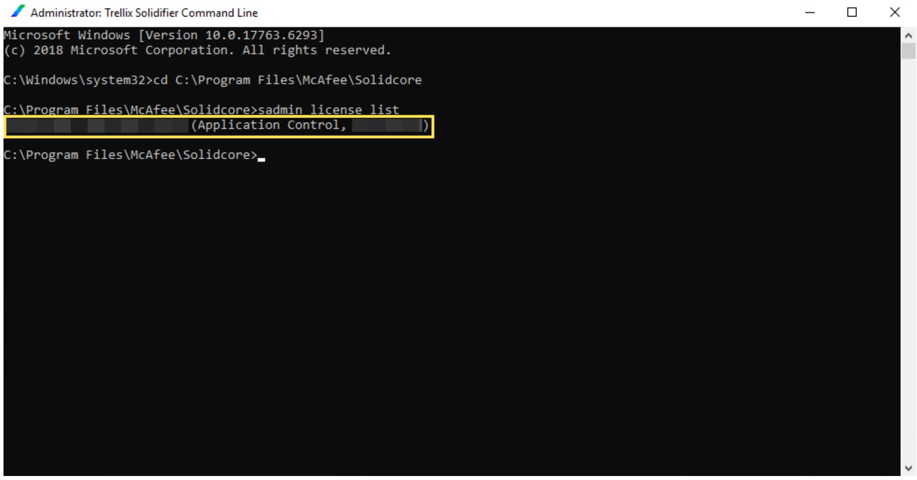
c. Enter the license key in the console.



Administrator: Trellix Solidifier Command Line

```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore

C:\Program Files\McAfee\Solidcore>sadmin license add [redacted]
"Application Control" license added.

C:\Program Files\McAfee\Solidcore>_
```

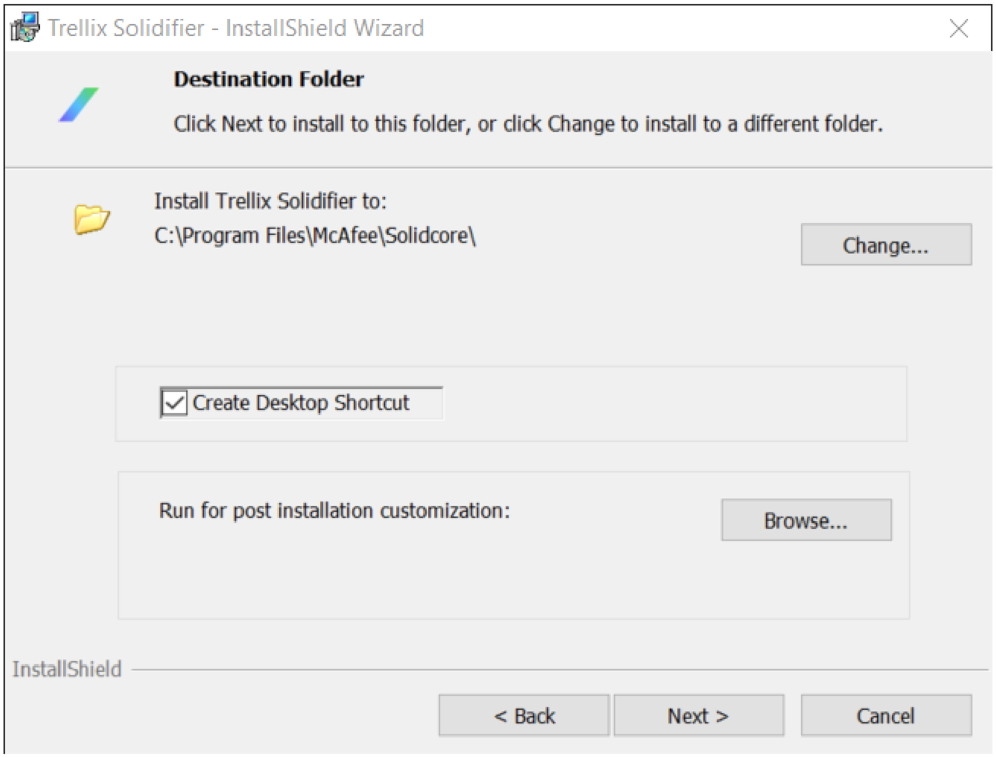
The license key is added.



Administrator: Trellix Solidifier Command Line

```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore

C:\Program Files\McAfee\Solidcore>sadmin license list
[redacted] (Application Control, [redacted])

C:\Program Files\McAfee\Solidcore>_
```

5 Select the destination folder and select **Next**.



Trellix Solidifier - InstallShield Wizard

Destination Folder

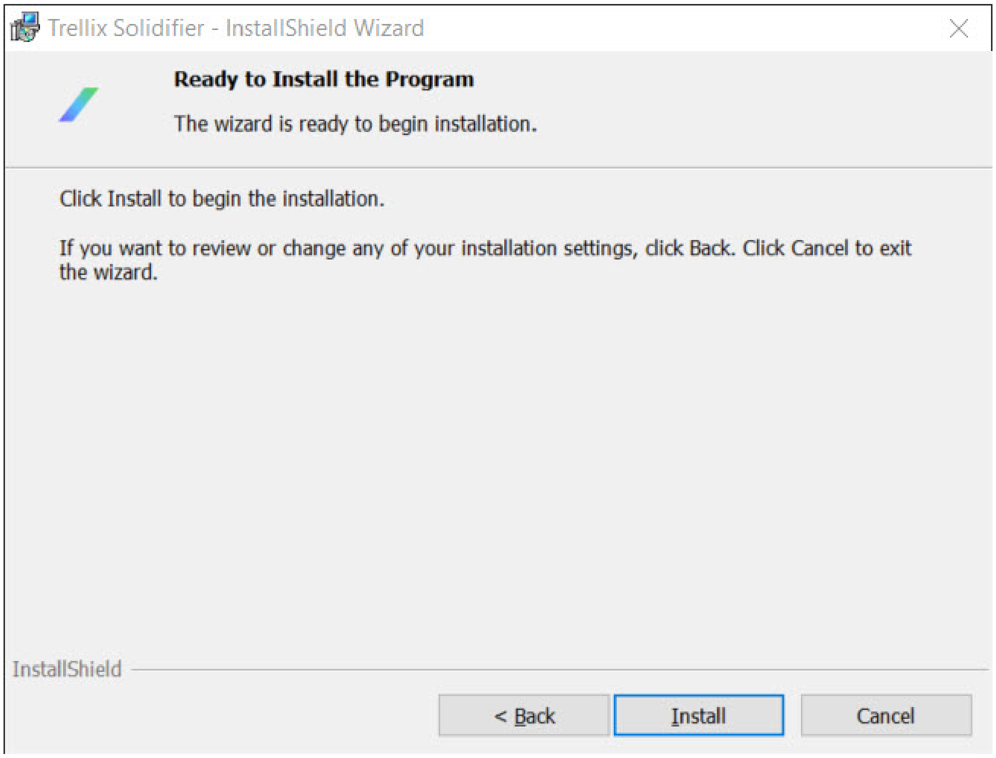
Click Next to install to this folder, or click Change to install to a different folder.

Install Trellix Solidifier to:
C:\Program Files\McAfee\Solidcore\

☑ Create Desktop Shortcut

Run for post installation customization:

6 Select **Install.** The system starts to install Trellix Solidifier

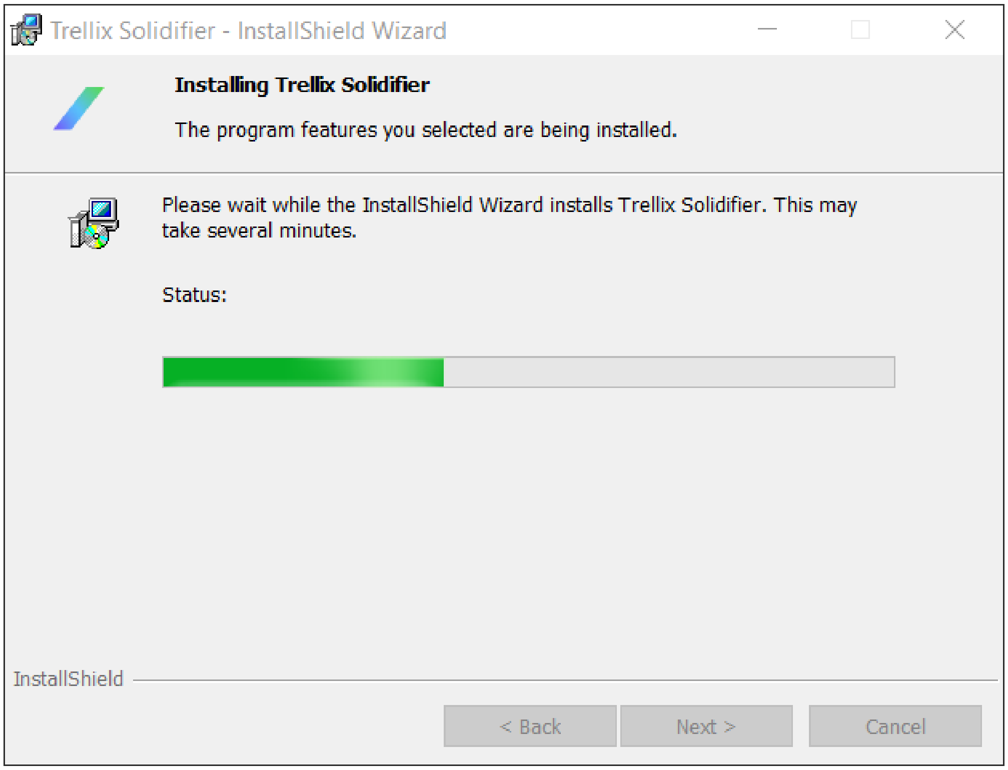


Ready to Install the Program

The wizard is ready to begin installation.

Click Install to begin the installation.

If you want to review or change any of your installation settings, click Back. Click Cancel to exit the wizard.



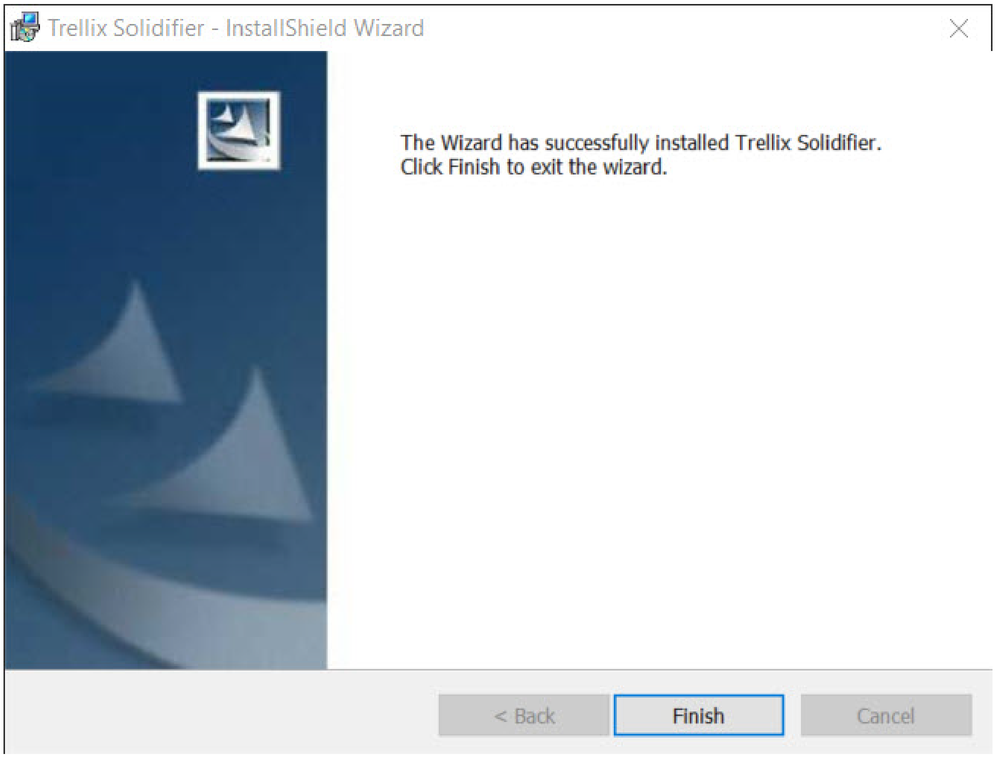
Installing Trellix Solidifier

The program features you selected are being installed.

Please wait while the InstallShield Wizard installs Trellix Solidifier. This may take several minutes.

Status:

7 Wait for the Trellix Solidifier to finish installing. Select **Finish**.



The Wizard has successfully installed Trellix Solidifier.
Click Finish to exit the wizard.

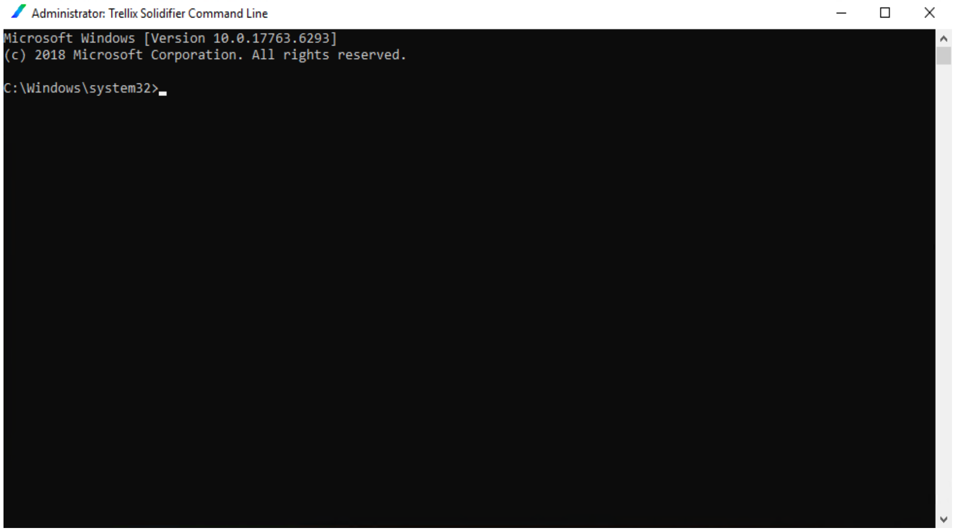
Enabling Trellix Application Control

The enabled Trellix Application Control Software prevents your system from executing unauthorized applications on your instrument workstation.

IMPORTANT Ensure all the necessary applications have been installed prior to enabling the Trellix Application Control.

---

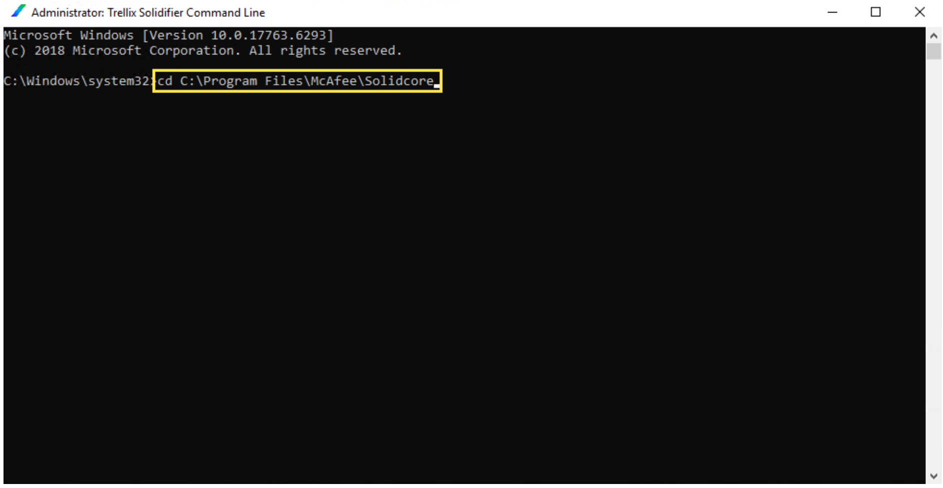
1 Right-click Trellix Solidifier... on your desktop, and select **Run as administrator**. The following window displays.



```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>_
```

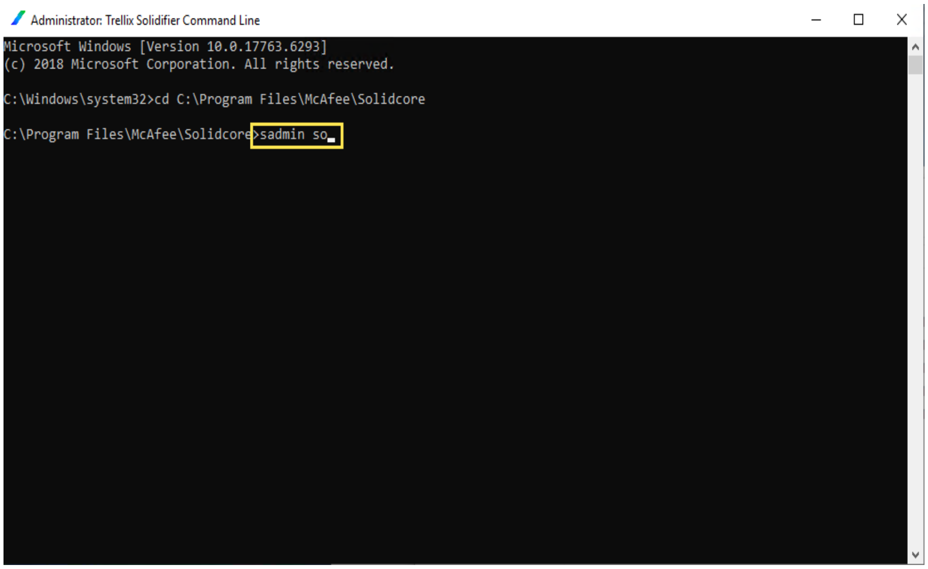
---

2 Enter **cd C:\Program Files\McAfee\Solidcore**.

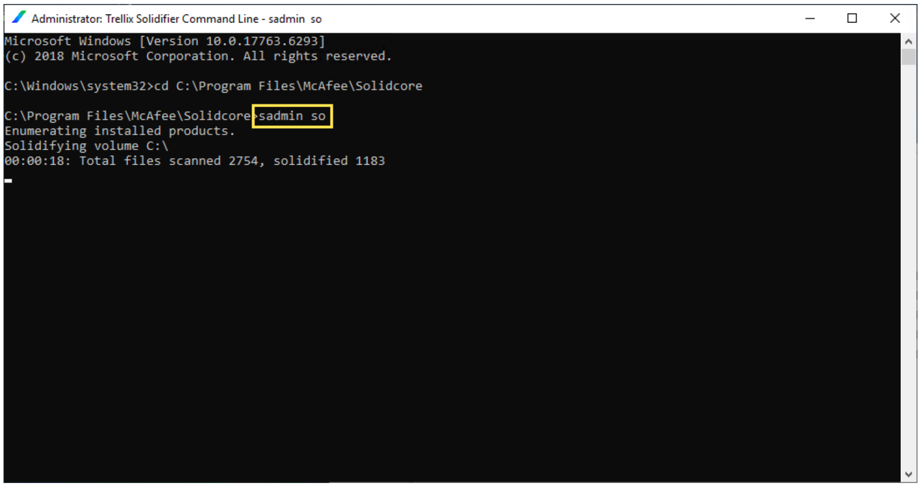


```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore_
```

3 Enter **sadmin so** in the console to scan the system and create an allowlist. The solidification process takes about 30 minutes depending on your system size and CPU performance.

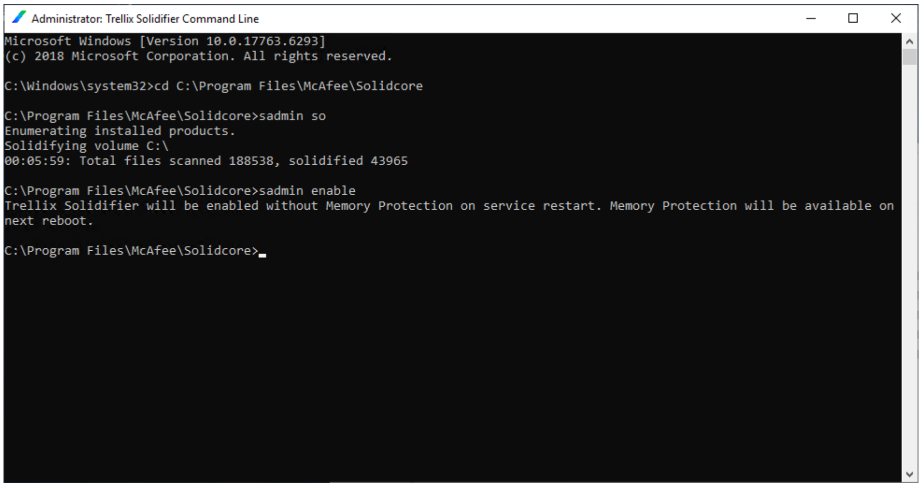


```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore

C:\Program Files\McAfee\Solidcore>sadmin so_
```



```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore

C:\Program Files\McAfee\Solidcore>sadmin so
Enumerating installed products.
Solidifying volume C:\
00:00:18: Total files scanned 2754, solidified 1183
_
```

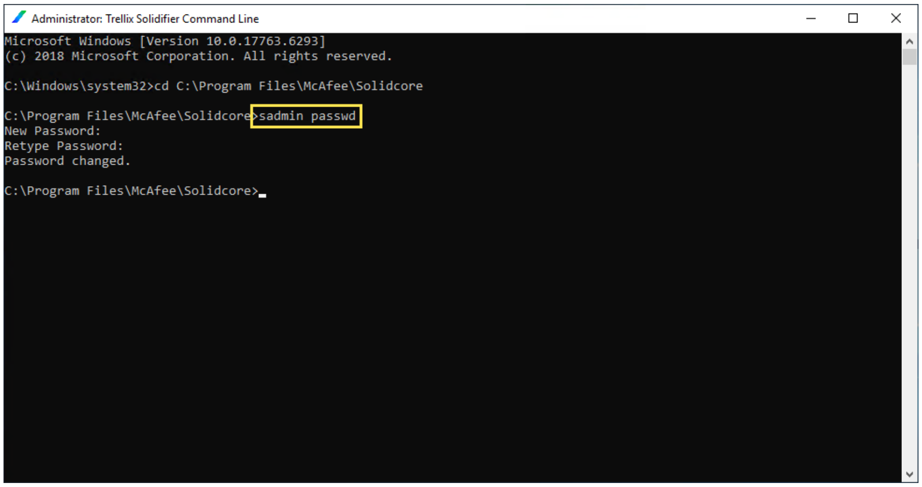
4 Enter **sadmin enable** to enable the Trellix Application Control.



```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore

C:\Program Files\McAfee\Solidcore>sadmin so
Enumerating installed products.
Solidifying volume C:\
00:05:59: Total files scanned 188538, solidified 43965

C:\Program Files\McAfee\Solidcore>sadmin enable
Trellix Solidifier will be enabled without Memory Protection on service restart. Memory Protection will be available on
next reboot.

C:\Program Files\McAfee\Solidcore>_
```

IMPORTANT Beckman Coulter recommends setting a password to manage the authority of enabling/disabling the Trellix Application Control.

5 Enter **sadmin passwd** to set a password to lock the Trellix Application Control authority.



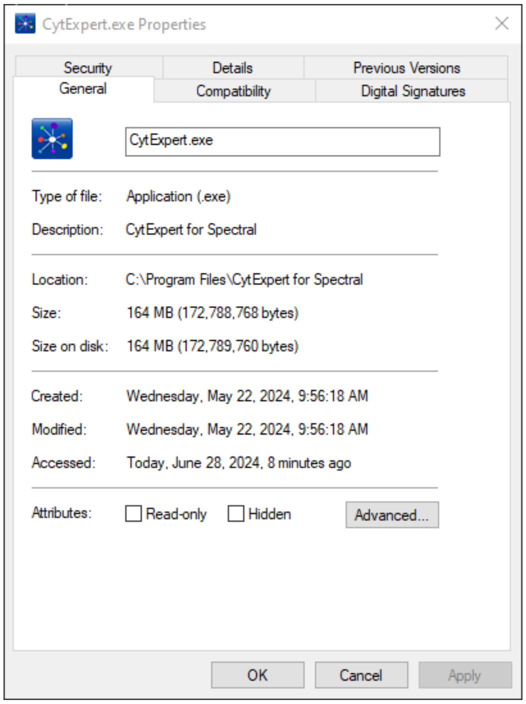
```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>cd C:\Program Files\McAfee\Solidcore

C:\Program Files\McAfee\Solidcore>sadmin passwd
New Password:
Retype Password:
Password changed.

C:\Program Files\McAfee\Solidcore>_
```

6 Restart the system.

Exporting the Certificate

Follow procedures below to export the certificate of CytExpert for Spectral software into your PC.

1 Right-click on the application **CytExpert.exe**, and select **Properties**. The following window displays.



CytExpert.exe

Type of file: Application (.exe)

Description: CytExpert for Spectral

Location: C:\Program Files\CytExpert for Spectral

Size: 164 MB (172,788,768 bytes)

Size on disk: 164 MB (172,789,760 bytes)

Created: Wednesday, May 22, 2024, 9:56:18 AM

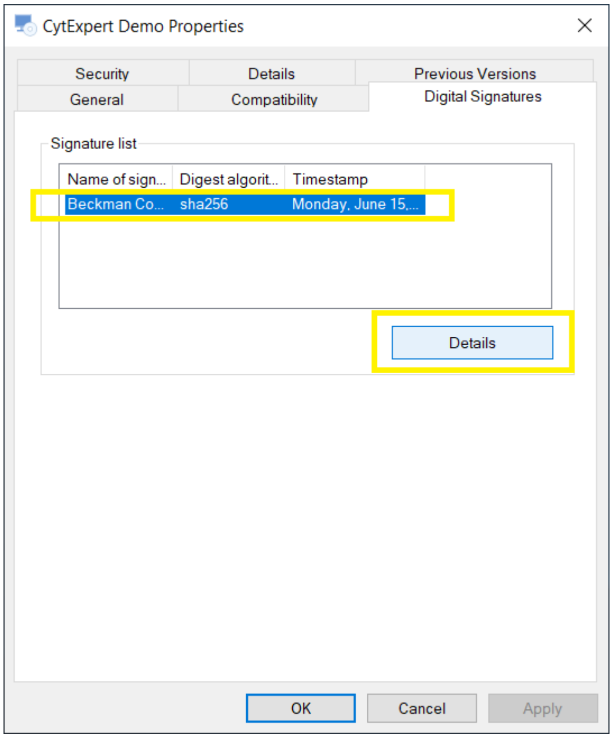
Modified: Wednesday, May 22, 2024, 9:56:18 AM

Accessed: Today, June 28, 2024, 8 minutes ago

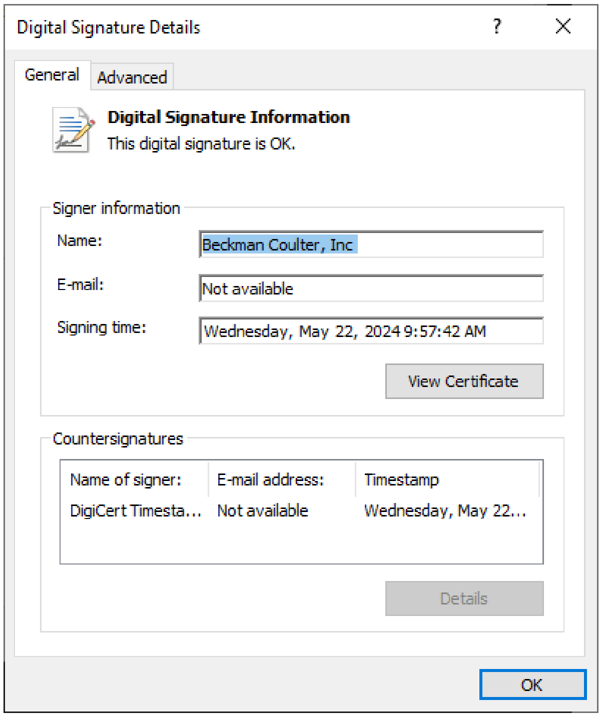
Attributes: ☐ Read-only ☐ Hidden

---

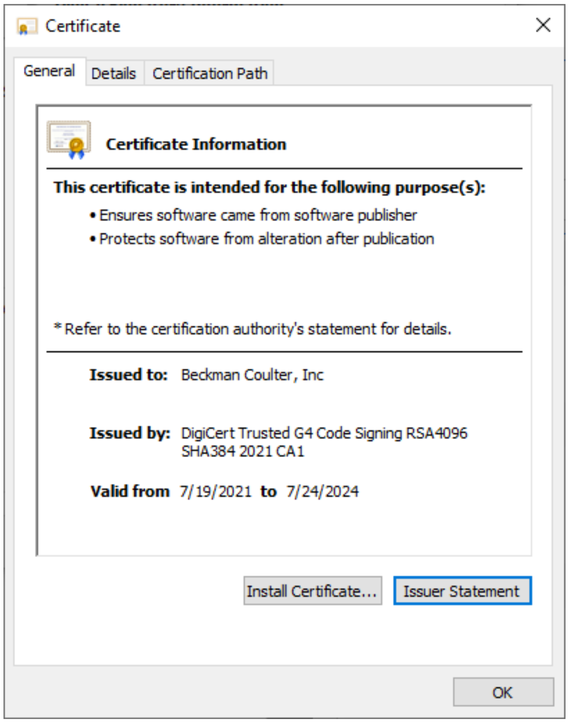
2 Select the **Digital Signatures** tab. Then select the signature and select **Details** to view the signature information.



The Digital Signature Details window displays.



3 Select **View Certificate.**



Certificate Information

This certificate is intended for the following purpose(s):

- Ensures software came from software publisher
- Protects software from alteration after publication

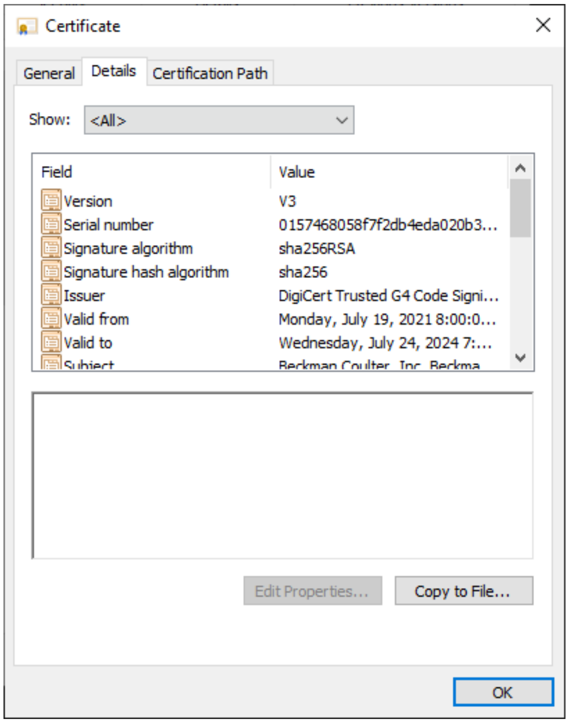
* Refer to the certification authority's statement for details.

Issued to: Beckman Coulter, Inc

Issued by: DigiCert Trusted G4 Code Signing RSA4096 SHA384 2021 CA1

Valid from 7/19/2021 **to** 7/24/2024

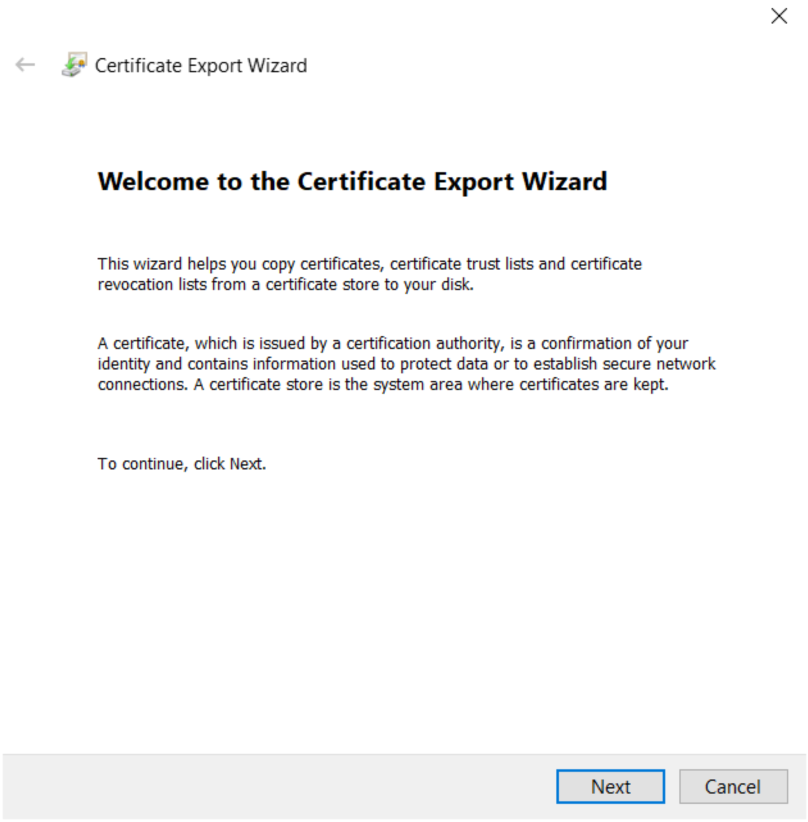
4 Select the **Details** tab and then select **Copy to File.**



Show: <All>

Field	Value
Version	V3
Serial number	0157468058f7f2db4eda020b3...
Signature algorithm	sha256RSA
Signature hash algorithm	sha256
Issuer	DigiCert Trusted G4 Code Signi...
Valid from	Monday, July 19, 2021 8:00:0...
Valid to	Wednesday, July 24, 2024 7:...
Subject	Beckman Coulter, Inc, Beckma

The Certificate Export Wizard window appears.



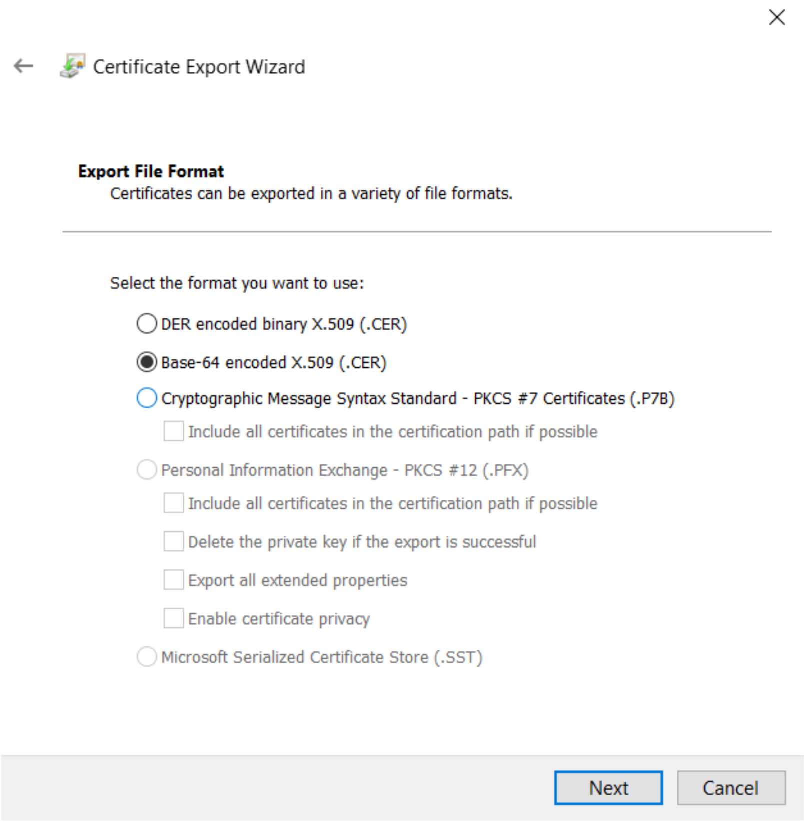
Welcome to the Certificate Export Wizard

This wizard helps you copy certificates, certificate trust lists and certificate revocation lists from a certificate store to your disk.

A certificate, which is issued by a certification authority, is a confirmation of your identity and contains information used to protect data or to establish secure network connections. A certificate store is the system area where certificates are kept.

To continue, click Next.

5 Select **Next** to export the certificate.



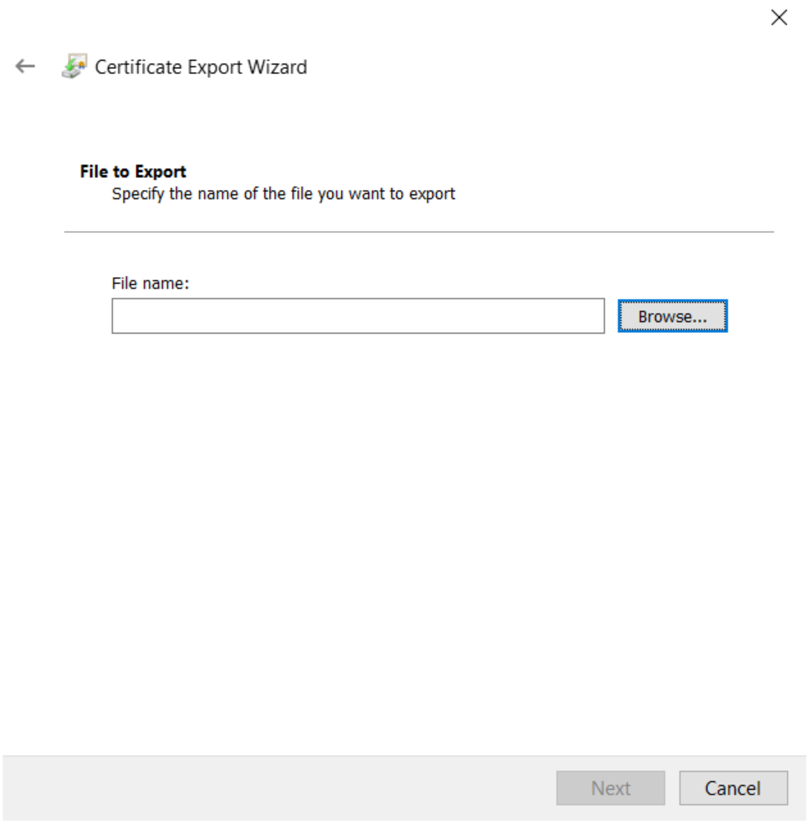
Export File Format

Certificates can be exported in a variety of file formats.

Select the format you want to use:

- ○ DER encoded binary X.509 (.CER)
- ● Base-64 encoded X.509 (.CER)
- ○ Cryptographic Message Syntax Standard - PKCS #7 Certificates (.P7B)
  - ☐ Include all certificates in the certification path if possible
- ○ Personal Information Exchange - PKCS #12 (.PFX)
  - ☐ Include all certificates in the certification path if possible
  - ☐ Delete the private key if the export is successful
  - ☐ Export all extended properties
  - ☐ Enable certificate privacy
- ○ Microsoft Serialized Certificate Store (.SST)

6 Select **Base-64-encoded X.509** and then select **Next.**

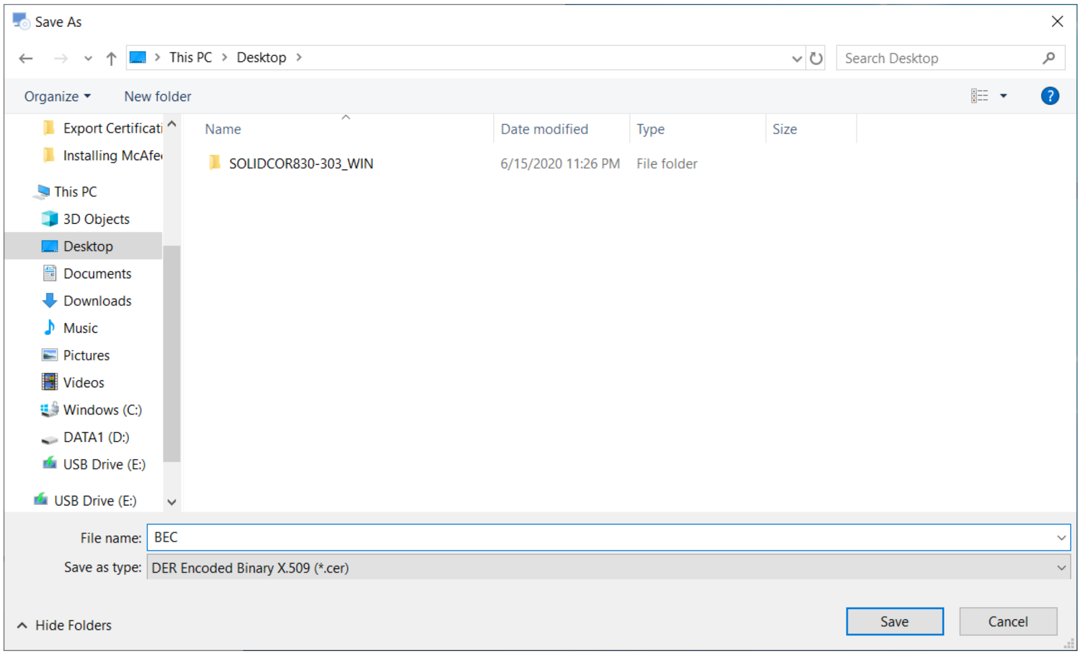


File to Export

Specify the name of the file you want to export

File name:

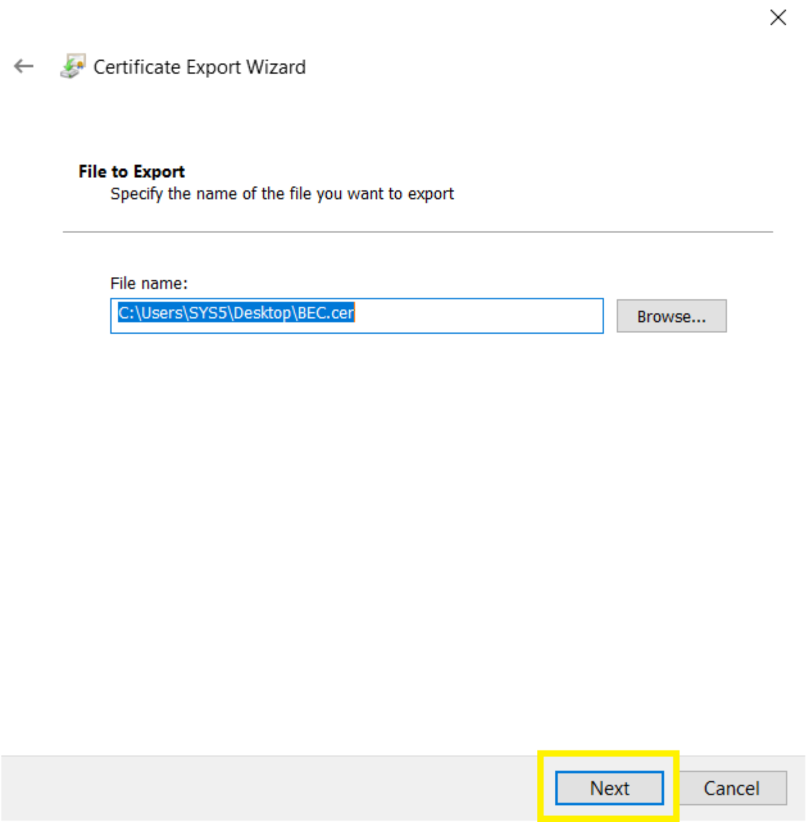
7 Select a destination to save the certificate and then select **Save.**



Name	Date modified	Type	Size
SOLIDCOR830-303_WIN	6/15/2020 11:26 PM	File folder	

File name: BEC
Save as type: DER Encoded Binary X.509 (*.cer)

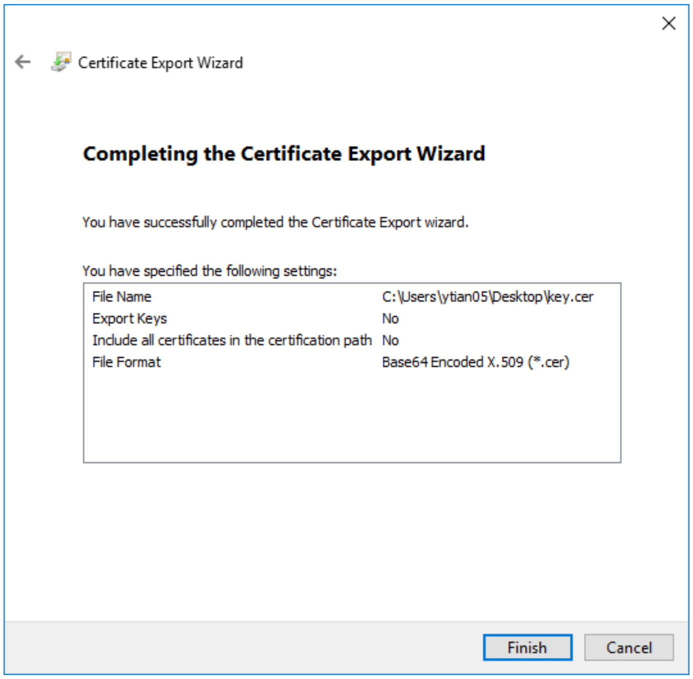
8 Select **Next**.



File to Export
Specify the name of the file you want to export

File name: C:\Users\SYS5\Desktop\BEC.cer

9 Select **Finish** when the success message displays.



Completing the Certificate Export Wizard

You have successfully completed the Certificate Export wizard.

You have specified the following settings:

File Name	C:\Users\ytian05\Desktop\key.cer
Export Keys	No
Include all certificates in the certification path	No
File Format	Base64 Encoded X.509 (*.cer)

Importing BEC Certificate

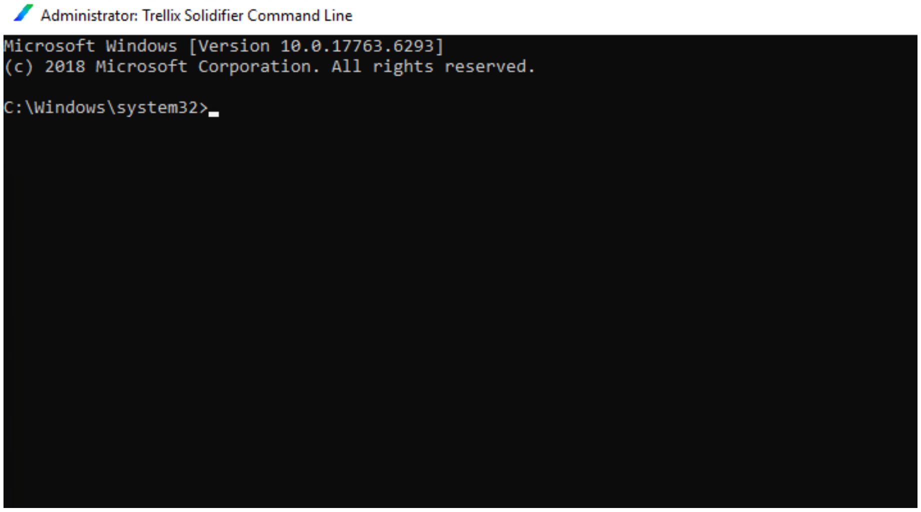
Follow the procedures below to add the certificate of CytExpert for Spectral software into the Trellix Certificate list. All the certified applications will be regarded as trusted and authorized by the Trellix Application Control.

IMPORTANT Trellix Application Control recognize only X.509 certificates.

1 Copy the certificate *BEC.cer* from the CytExpert for Spectral software disk into your instrument PC.

---

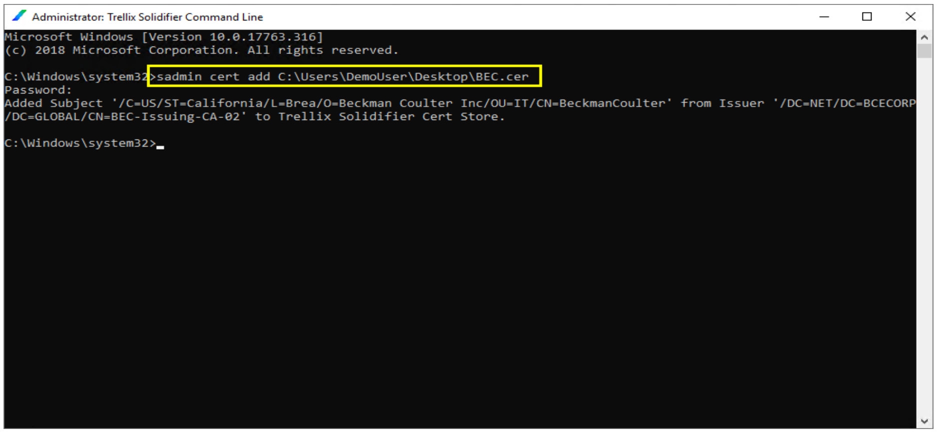
2 Right-click Trellix Solidifier… on your desktop, and select **Run as administrator**. The following window displays.



```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>_
```

---

3 Enter **sadmin cert add C:\BEC.cer** to add the certificate.

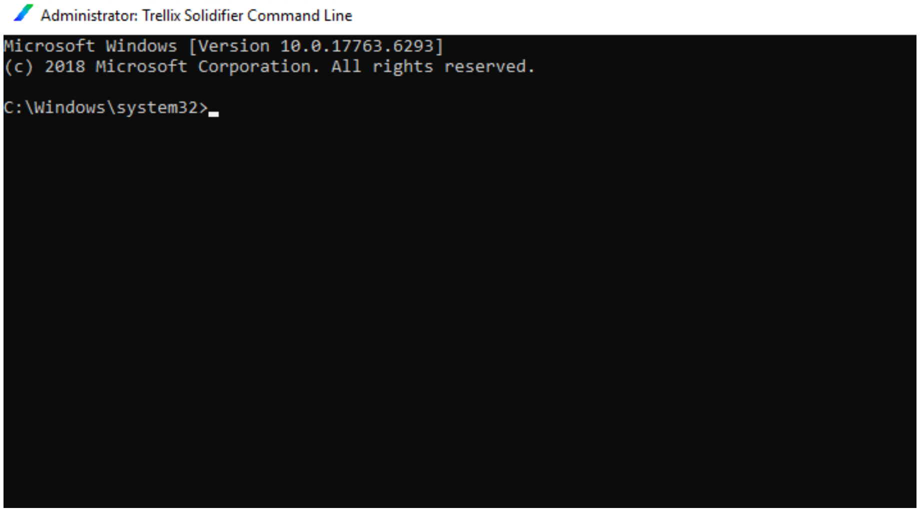


```
Microsoft Windows [Version 10.0.17763.316]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>sadmin cert add C:\Users\DemoUser\Desktop\BEC.cer
Password:
Added Subject '/C=US/ST=California/L=Brea/O=Beckman Coulter Inc/OU=IT/CN=BeckmanCoulter' from Issuer '/DC=NET/DC=BCECORP
/DC=GLOBAL/CN=BEC-Issuing-CA-02' to Trellix Solidifier Cert Store.

C:\Windows\system32>_
```

Switching Trellix Application Control to Update Mode

The update mode allows the Trellix Application Control to scan the instrument workstation and add the newly-installed application into the allowlist automatically. Switch the Trellix Application Control to the update mode when you execute scheduled or emergency changes on your instrument workstation.

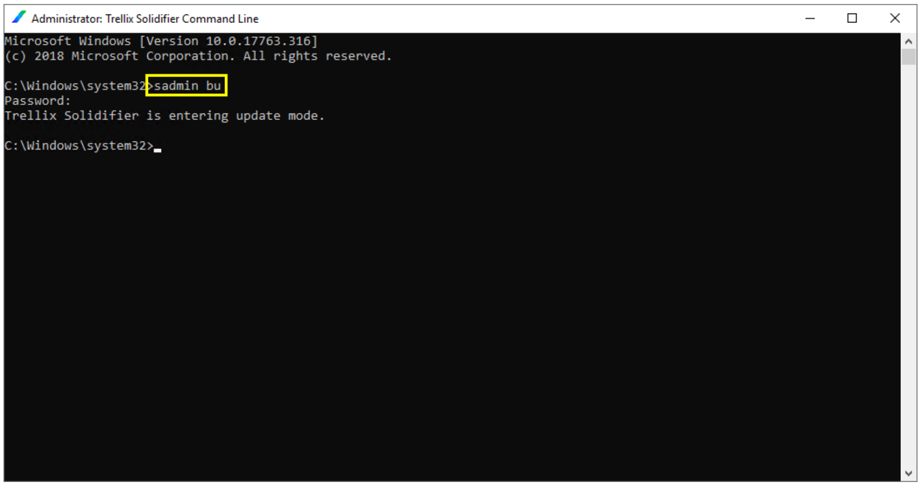
1 Right-click Trellix Solidifier… on your desktop, and select **Run as administrator**. The following window displays.



Administrator: Trellix Solidifier Command Line

```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>_
```

2 Enter **sadmin bu** to switch the Trellix Application Control to Update Mode.

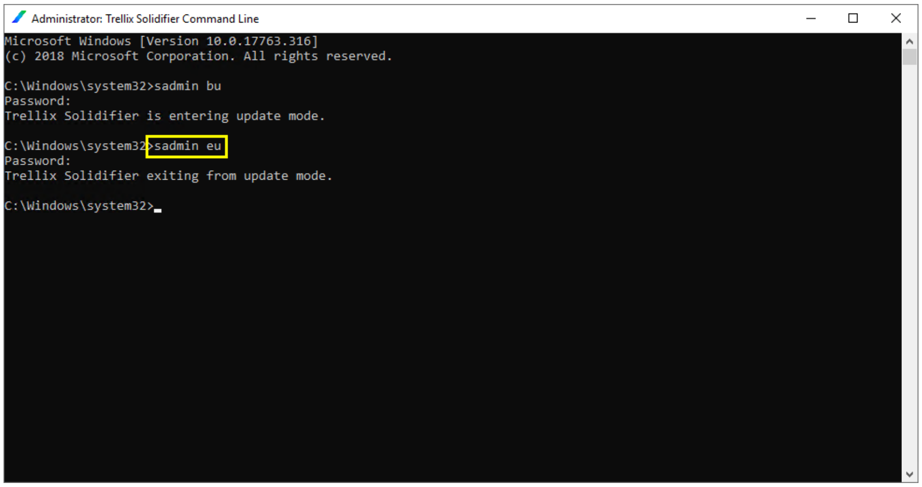


Administrator: Trellix Solidifier Command Line

```
Microsoft Windows [Version 10.0.17763.316]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>sadmin bu
Password:
Trellix Solidifier is entering update mode.

C:\Windows\system32>_
```

3 Execute scheduled or emergency changes, patch installations, or software updates for your instrument workstation.

4 Enter **sadmin eu** to exit the Update Mode.



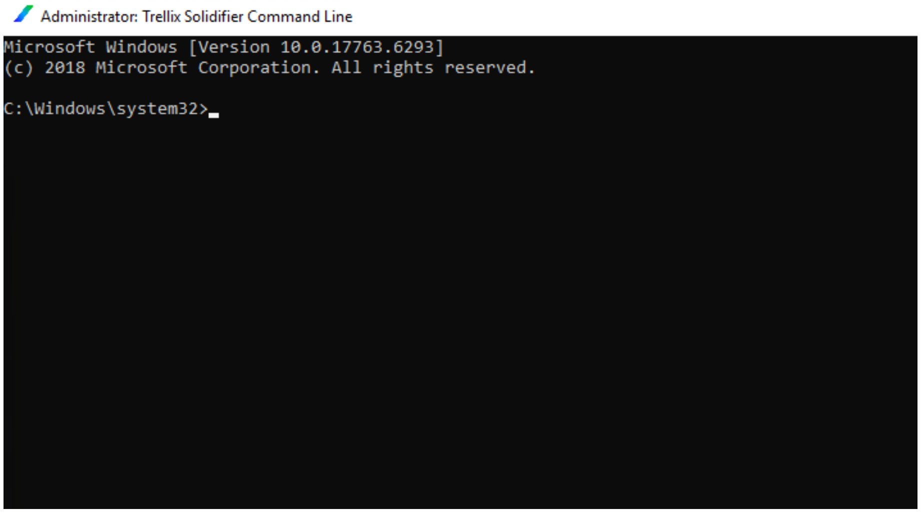
```
Microsoft Windows [Version 10.0.17763.316]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>sadmin bu
Password:
Trellix Solidifier is entering update mode.

C:\Windows\system32>sadmin eu
Password:
Trellix Solidifier exiting from update mode.

C:\Windows\system32>_
```

Disabling Trellix Application Control

IMPORTANT Disabling Trellix Application Control is a temporary method in exceptional cases, for example, Windows or firmware changes. This will help stop Trellix Application Control from blocking unauthorized applications. Beckman Coulter recommends enabling the Trellix Application Control in most cases.

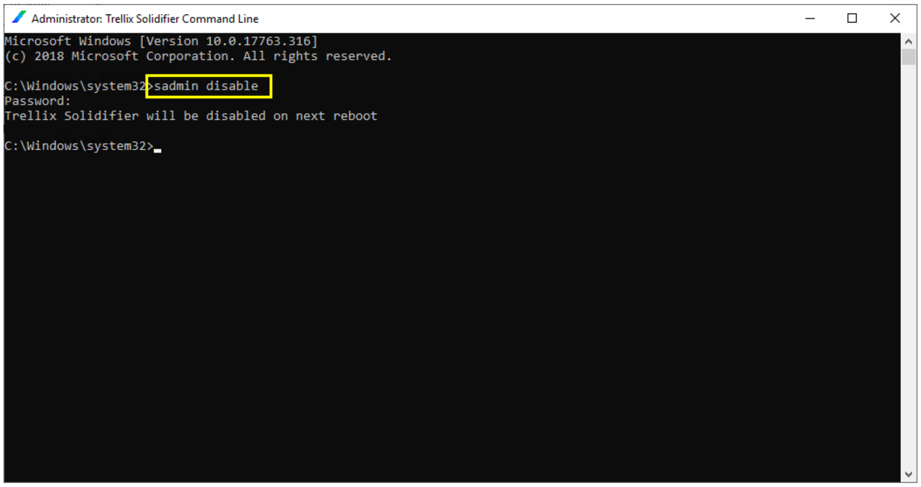
All the applications installed in this mode will not be included in the Trellix allowlist.

1 Right-click Trellix Solidifier... on your desktop, and select **Run as administrator**. The following window displays.



```
Microsoft Windows [Version 10.0.17763.6293]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>_
```

2 Enter **sadmin disable** in the console.

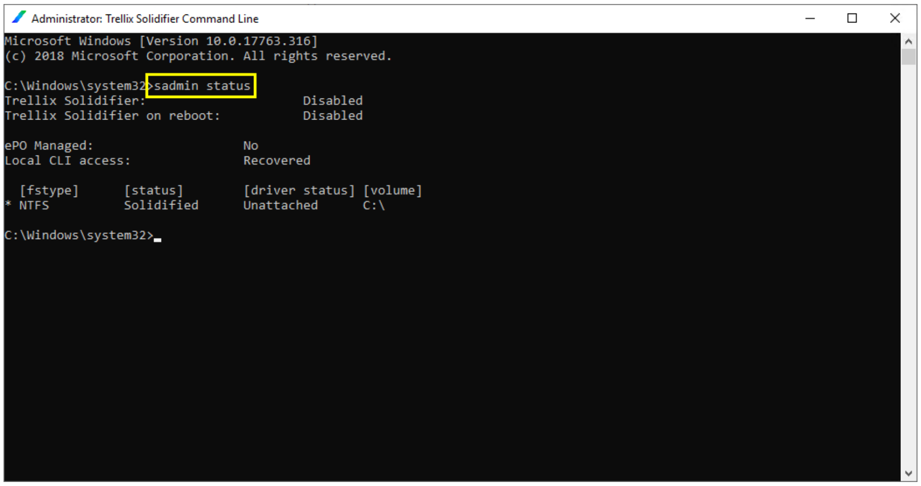


```
Microsoft Windows [Version 10.0.17763.316]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>sadmin disable
Password:
Trellix Solidifier will be disabled on next reboot

C:\Windows\system32>_
```

3 Select **Enter**.

4 Restart the System.

5 Enter **sadmin status** to view the status of Trellix Application Control.



```
Microsoft Windows [Version 10.0.17763.316]
(c) 2018 Microsoft Corporation. All rights reserved.

C:\Windows\system32>sadmin status
Trellix Solidifier:                     Disabled
Trellix Solidifier on reboot:           Disabled

ePO Managed:                    No
Local CLI access:               Recovered

  [fstype]      [status]        [driver status] [volume]
* NTFS          Solidified      Unattached      C:\

C:\Windows\system32>_
```

Security Updates

⚠ WARNING

Risk of software incompatibility. Only security updates posted to the Beckman Coulter website should be installed on the instrument controller PC. Do not install patches from any other source.

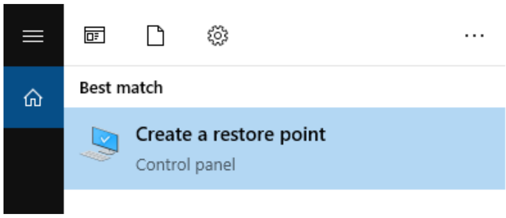
Install the update on one instrument and verify functionality before installing on other instruments.

IMPORTANT For information on the currently supported Windows build and security updates, consult the Release Notes of the latest validated update posted to the website. Refer to Downloading Security Updates.

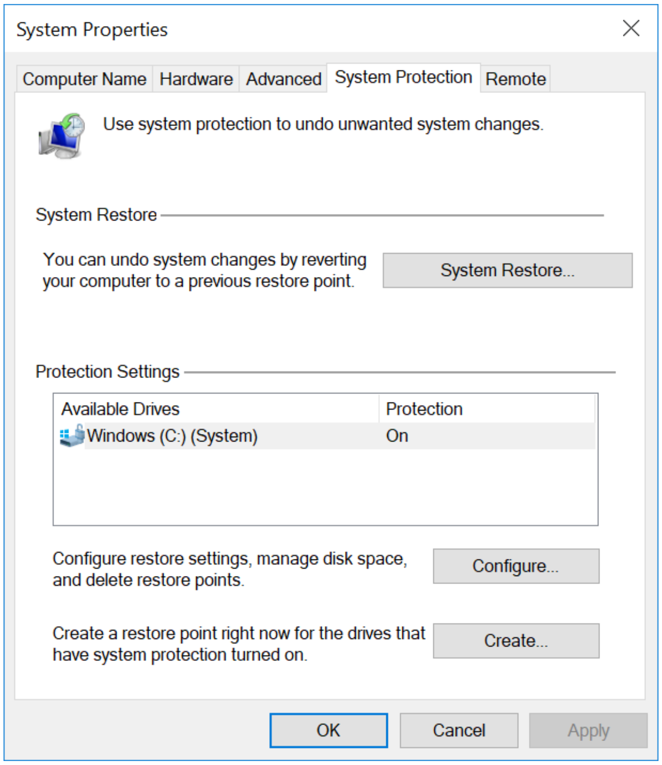
IMPORTANT Before installing the security updates, ensure you have a recent backup of the instrument database and experiment files (see CHAPTER 10, Backup and Restore) and the current BitLocker key (see Backing Up Your Recovery Key). Ensure the backups are saved to an encrypted external media or secured network drive.

Enabling System Protection

1 In the Start Menu, type **Restore** to locate the **Create a Restore Point**.



2 Double-click **Create a Restore Point** to open the System Properties window.



Use system protection to undo unwanted system changes.

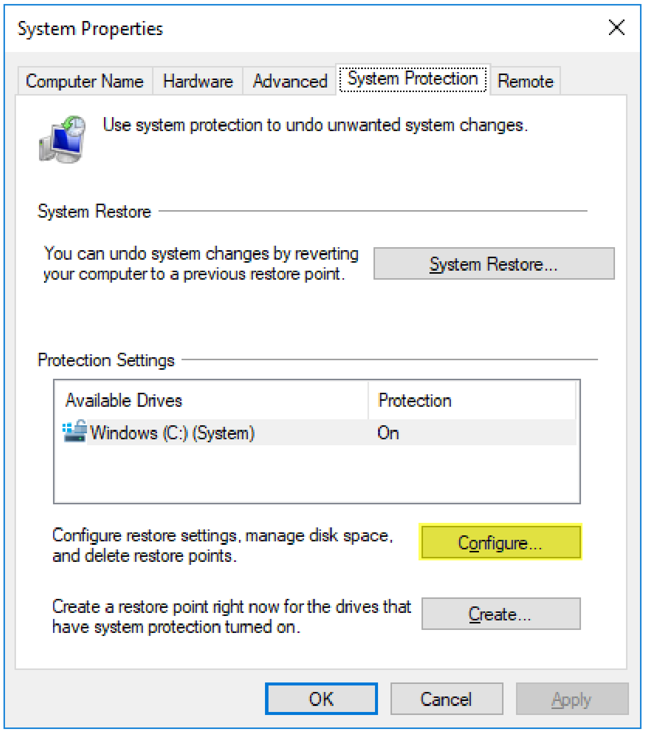
You can undo system changes by reverting your computer to a previous restore point.

Available Drives	Protection
Windows (C:) (System)	On

Configure restore settings, manage disk space, and delete restore points.

Create a restore point right now for the drives that have system protection turned on.

3 Select **Configure.**



Use system protection to undo unwanted system changes.

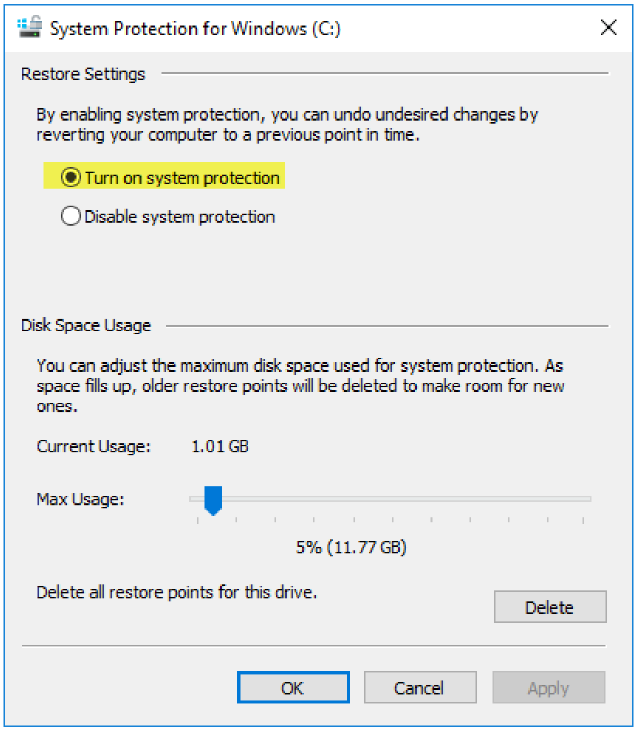
You can undo system changes by reverting your computer to a previous restore point.

Available Drives	Protection
Windows (C:) (System)	On

Configure restore settings, manage disk space, and delete restore points.

Create a restore point right now for the drives that have system protection turned on.

The following window appears.



By enabling system protection, you can undo undesired changes by reverting your computer to a previous point in time.

● Turn on system protection

○ Disable system protection

You can adjust the maximum disk space used for system protection. As space fills up, older restore points will be deleted to make room for new ones.

Current Usage: 1.01 GB

Max Usage: 5% (11.77 GB)

Delete all restore points for this drive.

4 Select **Turn on system protection**, then select **Apply**.

5 Close the System Properties window by selecting **OK**.

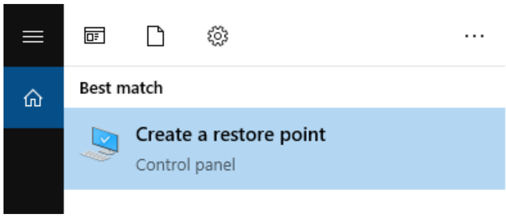
Creating a Restore Point

A restore point is used to remove all software and update installations that occurred after it was created. If installing an update on your PC causes any loss of functionality, the restore point can be used to place the system back into the operable state present before the update installation. In all cases, a system restore with any restore point would not cause loss of any data (i.e., LMD files) on the system.

⚠ CAUTION

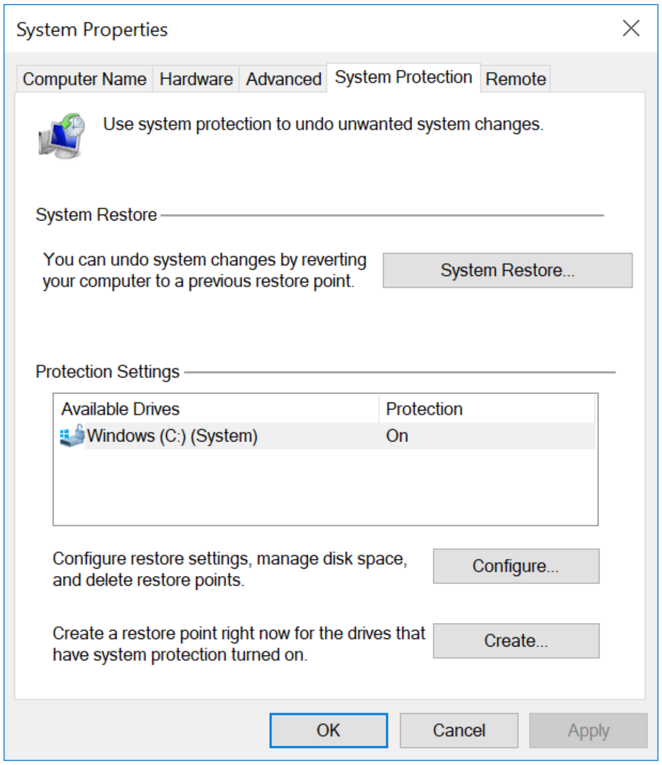
Risk of unexpected software modification. To ensure a system recovery point is available that is indicative of the most recent software configuration, the user should create a system restore point immediately prior to running the security update installer. Otherwise, a user would need to utilize the most recent automatically created restore point which might be up to 72 hours old and not include all recent software modifications.

1 In the Start Menu, type **Restore** to locate the **Create a Restore Point**.



---

2 Double-click **Create a Restore Point** to open the System Properties window.



Use system protection to undo unwanted system changes.

You can undo system changes by reverting your computer to a previous restore point.

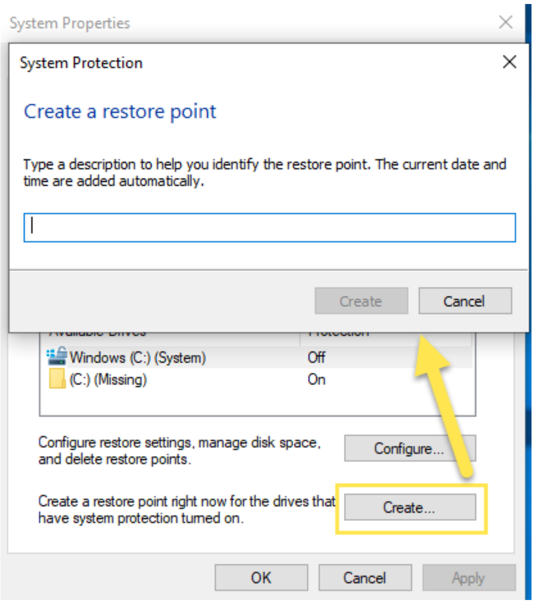
Available Drives	Protection
Windows (C:) (System)	On

Configure restore settings, manage disk space, and delete restore points.

Create a restore point right now for the drives that have system protection turned on.

---

3 Select **Create.**



Create a restore point

Type a description to help you identify the restore point. The current date and time are added automatically.

Windows (C:) (System)	Off
(C:) (Missing)	On

Configure restore settings, manage disk space, and delete restore points.

Create a restore point right now for the drives that have system protection turned on.

4 Type a name for the restore point and select **Create**.

5 Wait for the restore point creation to complete and then select **Close**.

6 Close the System Properties window by selecting **OK**.

Downloading Security Updates

In order to download and install security updates, you will need to access the Beckman Coulter website at www.beckmancoulter.com.

IMPORTANT Account registration is required for new users.

1 Go to www.beckmancoulter.com.

2 Using the menu bar in the in the upper right hand corner of the screen, hover over **Login**, then scroll down to select **Software Downloads.**

3 To search by product:

- **a.** Select **Research & Discovery** from the Market Segment dropdown menu.
- **b.** Select **Flow Cytometry** from the Product Line. dropdown menu.
- **c.** Select **Instruments** from the Product Series dropdown menu.
- **d.** Select **CytoFLEX mosaic** from the Product dropdown menu.

4 Select **Search**.

IMPORTANT Print out the Release Notes for each update as they will not be available when the next update is released. The Component Name section of the release notes identifies the Microsoft updates that are included in the download. This information may be requested by your IT team and will be needed to recover from a failed update.

5 Each security update is cumulative and only the most recent validated set of updates will be available for download. Select the first file in the results to view the release notes and then select Download.

If more than one update is listed in the search results, select Back, and repeat this step for each update.

If the security updates cannot be not downloaded directly to the instrument controller system, transfer the downloaded files via an encrypted external media or secured network drive to the instrument controller PC.

Notification Options

To receive notification when a new security update is validated and available for download, you must sign up for an account on www.beckmancoulter.com and follow the procedure for setting up subscriptions.

Sign up for subscriptions as outlined below:

- Select the **Document Language** consistent with your IFU language.
- Select **Research & Discovery** from Market Segment.
- Select **Flow Cytometry** from Product Line.
- Select **Instruments** from Product Series.
- Select **CytoFLEX mosaic** from Product.

If you do not sign up for product notifications, you will have to check periodically for newly released security updates, see Downloading Security Updates.

NOTE Beckman Coulter will only validate and post updates for Windows 10 LTSC 2019.

Installing the Security Updates

⚠ **CAUTION**

Risk of software incompatibility. Beckman Coulter can only support security updates on unmodified systems. Application of domain policies or other configuration changes may impact compatibility.

IMPORTANT If more than one update is available for your product, install the updates in the order indicated by the sequence number as outlined in the file naming convention below.

RSU-<Operating System> <Product Name>-<Year><Month>_<Sequence #>_<Sequence Revision#>

Example: RSU-W10LTSC-CytoFLEX for Spectral-2025Feb_1_0

1. Ensure you have a recent copy of your BitLocker key, refer to Backing Up Your Recovery Key.

2. Login to Windows as a user with Administrator access.

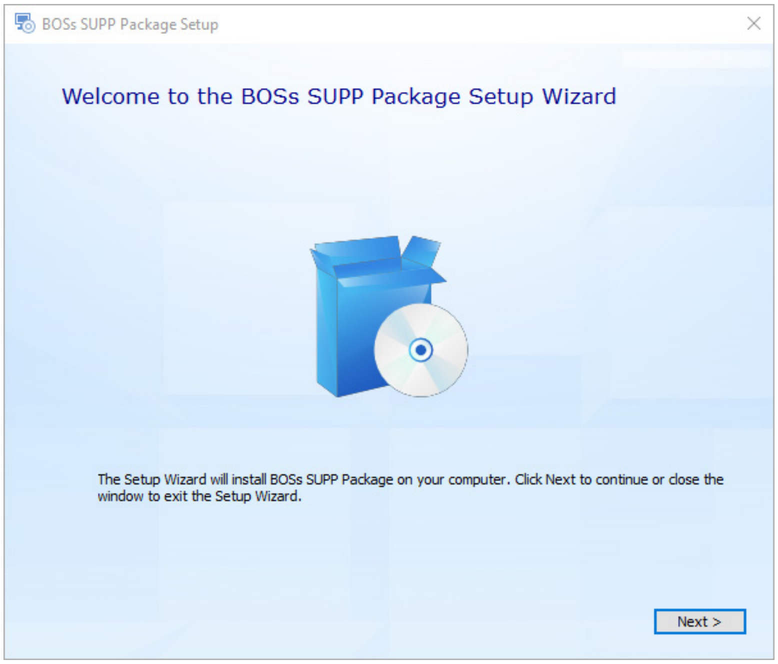
3. Close all program and save open files.

---

4 Power down the instrument.

---

5 Open the first security update file (For example: RSU-W10LTSC-CytoFLEX for Spectral-2025Feb_1_0) on the Instrument Controller PC by double-clicking on the file. The BOSS SUPP Package Setup window appears.



BOSs SUPP Package Setup

Welcome to the BOSs SUPP Package Setup Wizard

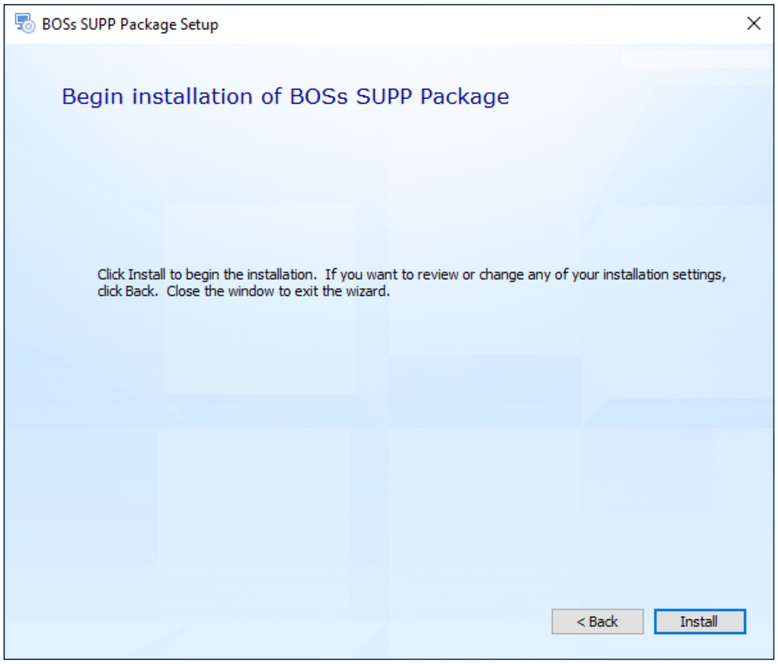
The Setup Wizard will install BOSs SUPP Package on your computer. Click Next to continue or close the window to exit the Setup Wizard.

If the Microsoft Windows Defender Smart Screen prompt appears, select **More Info** and **Run Anyway** to launch the installer.

A User Account Control message may display the message *Do you want to allow this app to make changes to your device?* Select **Yes** to continue.

NOTE The security update installer will automatically detect and install only the necessary updates.

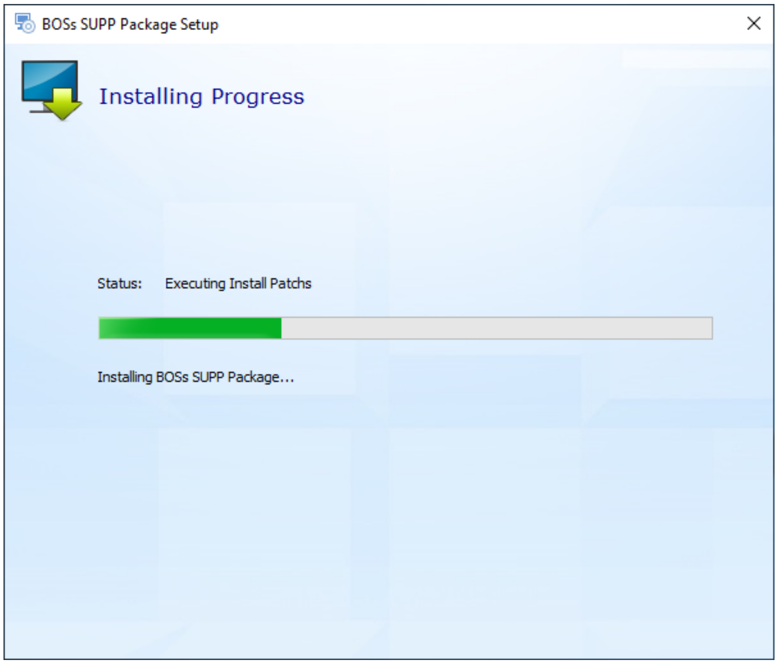
6 Select **Next.** The Begin installation of BOSS SUPP Package window appears.



Begin installation of BOSs SUPP Package

Click Install to begin the installation. If you want to review or change any of your installation settings, click Back. Close the window to exit the wizard.

7 Select **Install** to begin installing the security updates. The Installing Progress window appears. This process may take several minutes.

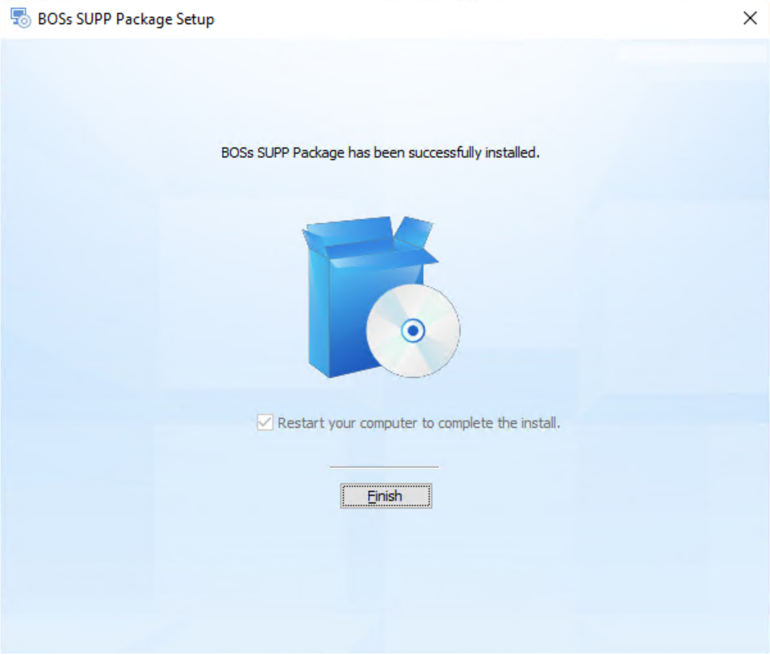


Installing Progress

Status: Executing Install Patchs

Installing BOSs SUPP Package...

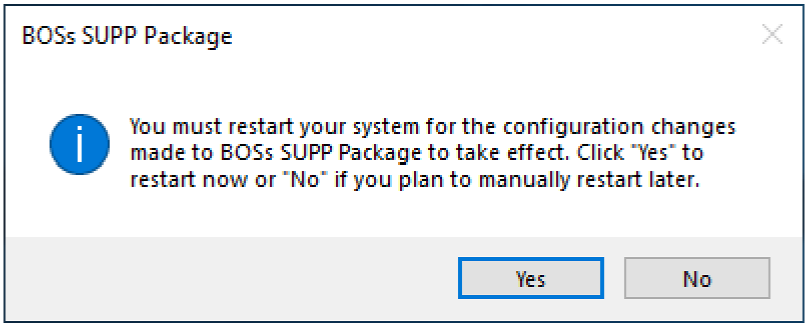
8 Wait for the security updates to finish installing. The install complete window appears.



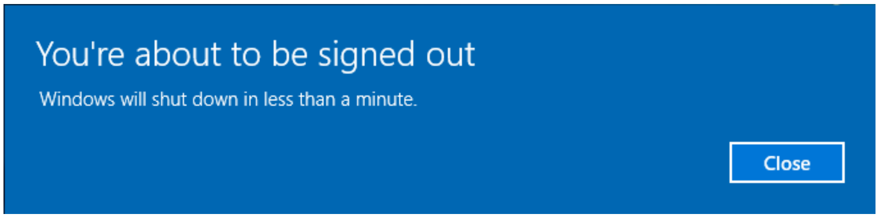
BOSs SUPP Package has been successfully installed.

☑ Restart your computer to complete the install.

9 Select **Finish**. When prompted to restart the system, select **Yes**.



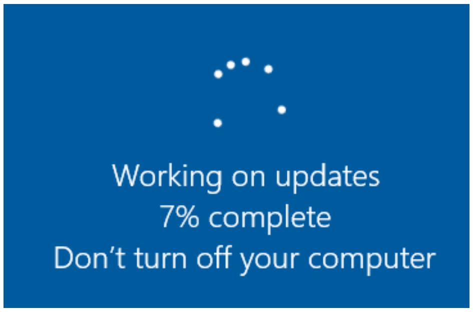
You must restart your system for the configuration changes made to BOSs SUPP Package to take effect. Click "Yes" to restart now or "No" if you plan to manually restart later.



You're about to be signed out

Windows will shut down in less than a minute.

10 Wait while the system completes the installation of the security updates. This process may take several minutes and will return you to the Windows login screen.



Working on updates
7% complete
Don't turn off your computer

11 Install any additional downloaded security updates (sequence numbers above 1) by repeating Steps 5 to 9.

12 Verify functionality of instrument software before installing security updates on any other system. If functionality is impaired, see Recovering from Failed Security Updates.

Recovering from Failed Security Updates

These steps below should only be followed if the instrument software or operating system functionality is impaired after a security update was applied.

1 Power down the instrument and close all programs. Do not turn your workstation off.

2 In the start menu, type view update and select **View your Update History**.

3 Select **Uninstall Updates.**

4 Highlight the update listed under Microsoft Windows that matches the cumulative security update KB number in the release notes. Refer to Component Name section of the Release Notes, see Step 5, Downloading Security Updates.

5 Select **Uninstall** and select **Yes** when prompted to verify the uninstall. This process may take several minutes to complete.

6 When prompted to restart, select **Restart Now.**

7 Once the PC has been rebooted, login and verify Microsoft update has been removed by repeating Steps 2 and 3. Verify the KB selected in Step 4 is no longer listed.

8 Power on the instrument and verify restored software functionality.

- If software functionality has been restored, continue using your instrument without the update installed. Contact us for troubleshooting the update incompatibility.
- If software functionality has not been restored and the PC was not provided by Beckman Coulter, contact laboratory IT for system restore options.
- If software functionality has not been restored and the PC was provided by Beckman Coulter, contact us for system restore.

User Management

⚠ CAUTION

Risk of unauthorized access and modification. CytoFLEX workstation transmits unencrypted data to any network printer. To avoid unauthorized access to transmitted data, ensure adequate network protection mechanism are in place.

⚠ CAUTION

Risk of unauthorized access and modification. To protect your workstation from unauthorized access and modification, please refer to the following recommendations.

Beckman Coulter recommends that you use role-based access control policies to manage user accounts on workstations. Assigning appropriate roles to users accessing workstations is the responsibility and obligation of workstation administrators. Table E.1 is a good practice recommended by Beckman Coulter to manage user accounts according to the use requirements of the workstation and combined with Windows built-in user groups.

Table E.1 Role-based Access Control Policies

Requirements for Using a Workstation	Windows Built-in User Groups	Role Example
Responsible for the daily maintenance of the workstation and have full control authority over the workstation. Can install, uninstall software, install security updates, restore the system, and manage user accounts on workstation.	Administrators	**1.** IT Administrator **2.** Device Administrator
The workstation is used daily to control the cytometer to complete collection, QC, analysis and other work.	User	**1.** Operator **2.** Experimenter

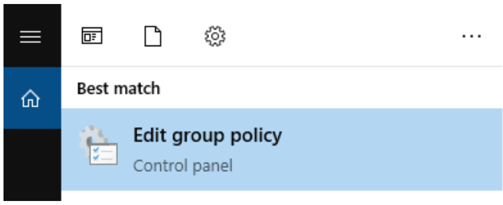
System Hardening

Enabling Automatic Log-Off

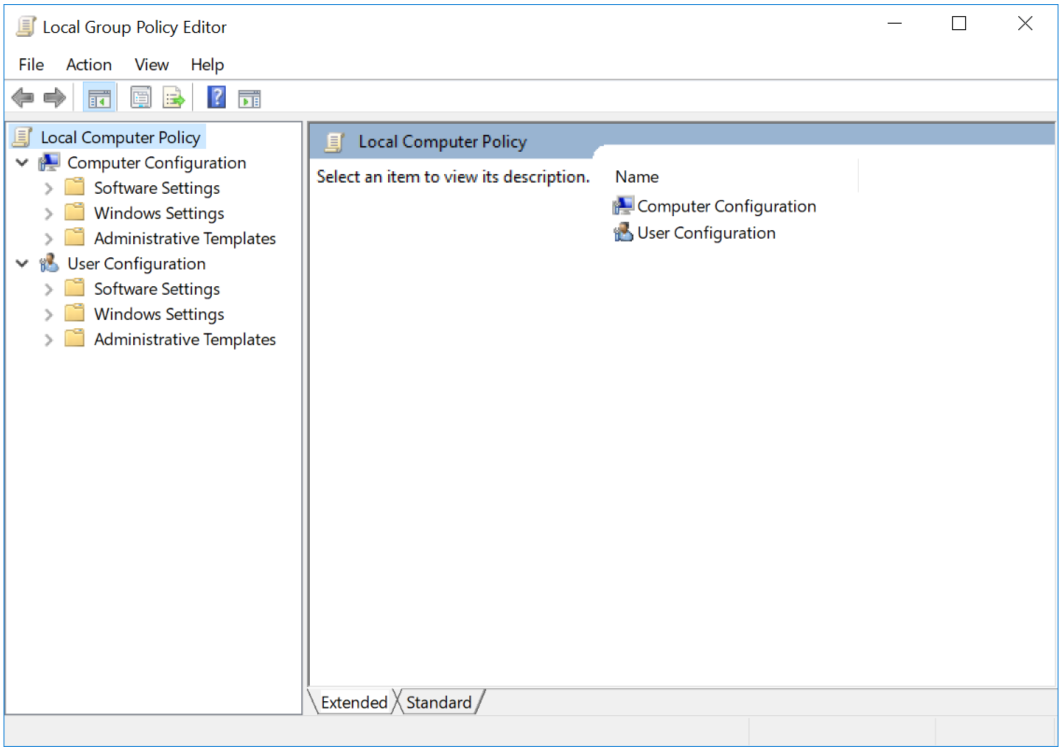
IMPORTANT Automatic log-off can protect your instrument PC from unauthorized operation when you leave the PC temporarily. Follow the steps below to enable the Screen Saver protection.

Enabling Screen Saver Protection

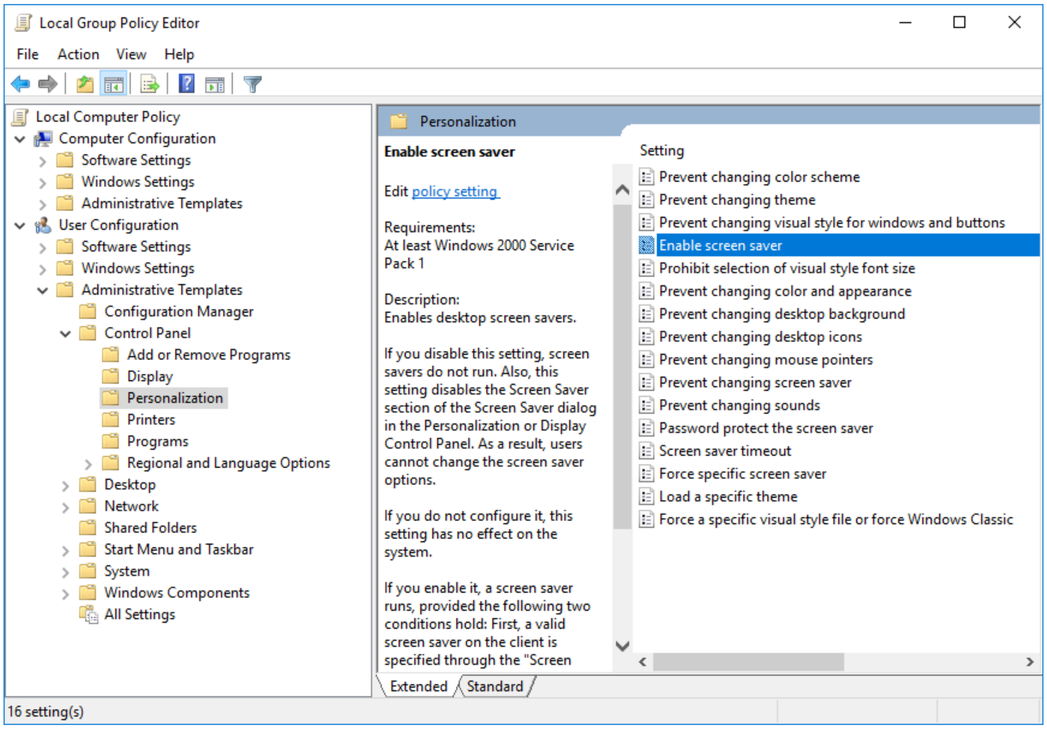
1 Type **Edit group policy** from the Windows search bar to locate the Edit group policy editor.



2 Select **Edit group policy**. The Local Group Policy Editor window displays.



3 Navigate to **User Configuration** > **Administrative Templates** > **Control Panel** > **Personalization.**



Enable screen saver

Edit policy setting

Requirements:
At least Windows 2000 Service Pack 1

Description:
Enables desktop screen savers.

If you disable this setting, screen savers do not run. Also, this setting disables the Screen Saver section of the Screen Saver dialog in the Personalization or Display Control Panel. As a result, users cannot change the screen saver options.

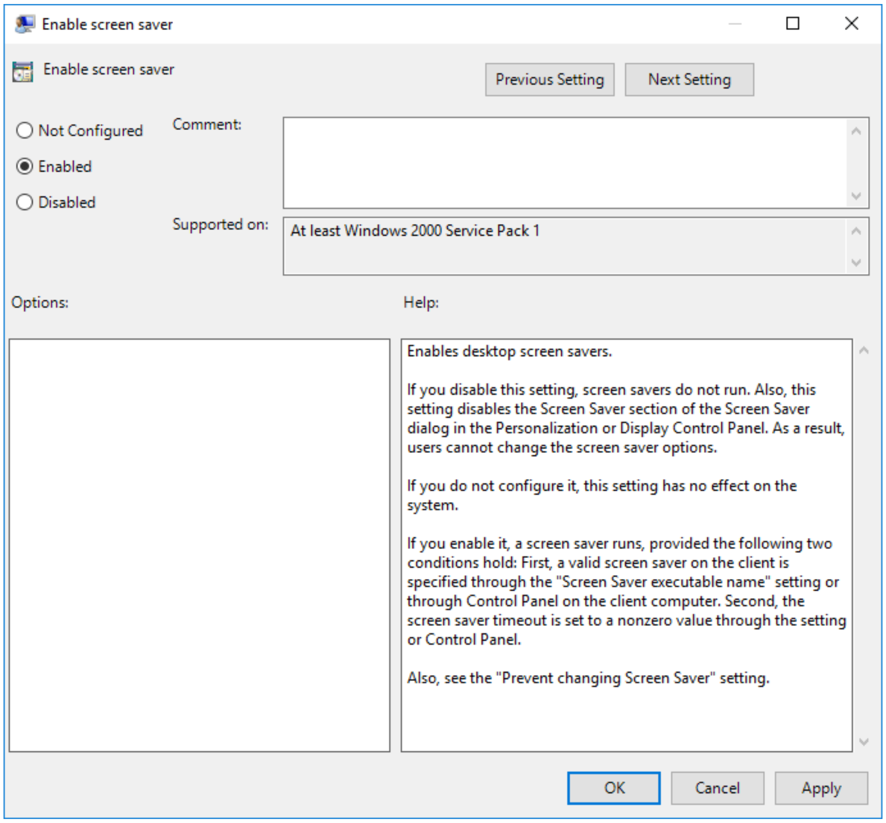
If you do not configure it, this setting has no effect on the system.

If you enable it, a screen saver runs, provided the following two conditions hold: First, a valid screen saver on the client is specified through the "Screen

Setting
- Prevent changing color scheme
- Prevent changing theme
- Prevent changing visual style for windows and buttons
- Enable screen saver
- Prohibit selection of visual style font size
- Prevent changing color and appearance
- Prevent changing desktop background
- Prevent changing desktop icons
- Prevent changing mouse pointers
- Prevent changing screen saver
- Prevent changing sounds
- Password protect the screen saver
- Screen saver timeout
- Force specific screen saver
- Load a specific theme
- Force a specific visual style file or force Windows Classic

---

4 Select **Enable screen saver.** The following window appears.



Supported on: At least Windows 2000 Service Pack 1

Enables desktop screen savers.

If you disable this setting, screen savers do not run. Also, this setting disables the Screen Saver section of the Screen Saver dialog in the Personalization or Display Control Panel. As a result, users cannot change the screen saver options.

If you do not configure it, this setting has no effect on the system.

If you enable it, a screen saver runs, provided the following two conditions hold: First, a valid screen saver on the client is specified through the "Screen Saver executable name" setting or through Control Panel on the client computer. Second, the screen saver timeout is set to a nonzero value through the setting or Control Panel.

Also, see the "Prevent changing Screen Saver" setting.

---

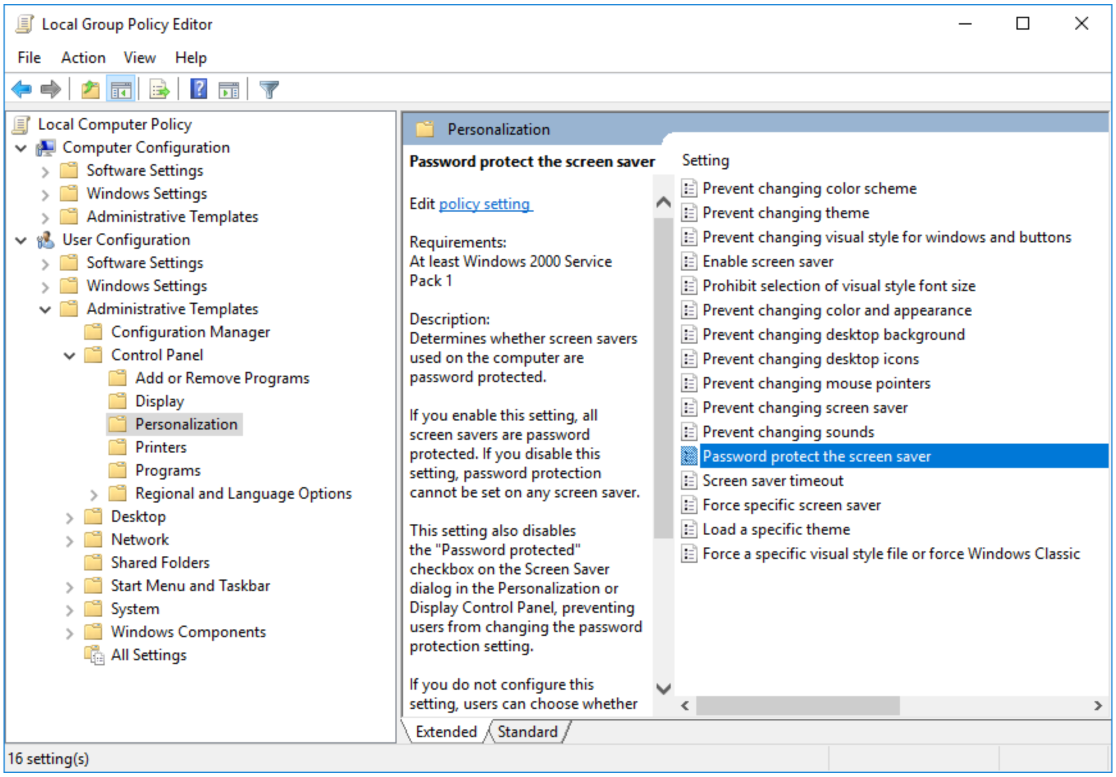
5 Select **Enabled** and **Apply**.

---

6 Select **OK** to close the Enable screen saver window.

---

7 Select **Password protect the screen saver** from the Local Group Policy Editor window.



- Local Computer Policy
  - Computer Configuration
    - Software Settings
    - Windows Settings
    - Administrative Templates
  - User Configuration
    - Software Settings
    - Windows Settings
    - Administrative Templates
      - Configuration Manager
      - Control Panel
        - Add or Remove Programs
        - Display
        - Personalization
        - Printers
        - Programs
        - Regional and Language Options
      - Desktop
      - Network
      - Shared Folders
      - Start Menu and Taskbar
      - System
      - Windows Components
      - All Settings

Personalization

Password protect the screen saver

Edit policy setting

Requirements:
At least Windows 2000 Service Pack 1

Description:
Determines whether screen savers used on the computer are password protected.

If you enable this setting, all screen savers are password protected. If you disable this setting, password protection cannot be set on any screen saver.

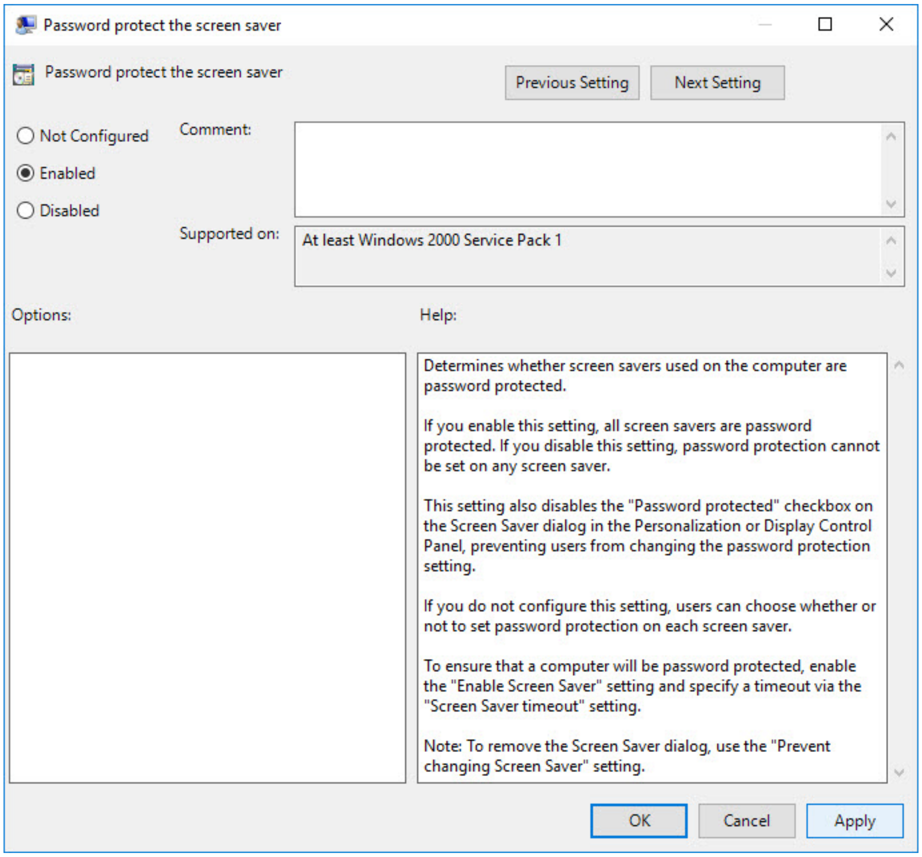
This setting also disables the "Password protected" checkbox on the Screen Saver dialog in the Personalization or Display Control Panel, preventing users from changing the password protection setting.

If you do not configure this setting, users can choose whether

Setting
- Prevent changing color scheme
- Prevent changing theme
- Prevent changing visual style for windows and buttons
- Enable screen saver
- Prohibit selection of visual style font size
- Prevent changing color and appearance
- Prevent changing desktop background
- Prevent changing desktop icons
- Prevent changing mouse pointers
- Prevent changing screen saver
- Prevent changing sounds
- Password protect the screen saver
- Screen saver timeout
- Force specific screen saver
- Load a specific theme
- Force a specific visual style file or force Windows Classic

16 setting(s)

8 Select **Enabled** and **Apply**.



○ Not Configured

● Enabled

○ Disabled

Supported on: At least Windows 2000 Service Pack 1

Determines whether screen savers used on the computer are password protected.

If you enable this setting, all screen savers are password protected. If you disable this setting, password protection cannot be set on any screen saver.

This setting also disables the "Password protected" checkbox on the Screen Saver dialog in the Personalization or Display Control Panel, preventing users from changing the password protection setting.

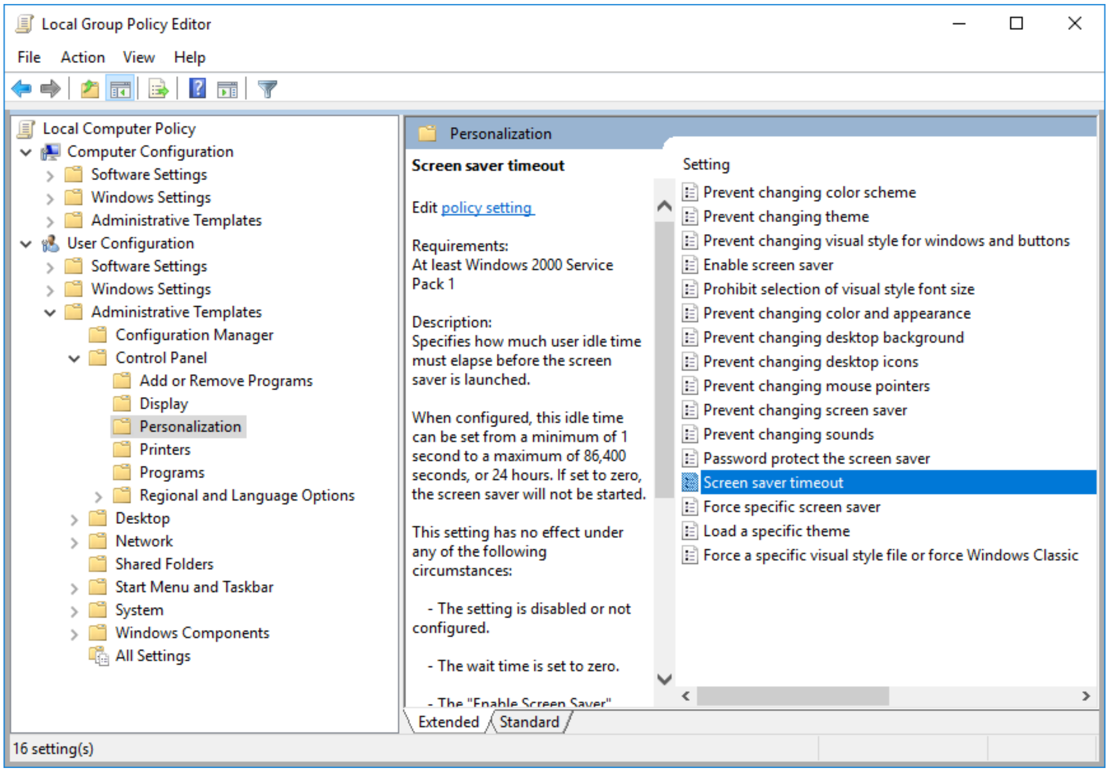
If you do not configure this setting, users can choose whether or not to set password protection on each screen saver.

To ensure that a computer will be password protected, enable the "Enable Screen Saver" setting and specify a timeout via the "Screen Saver timeout" setting.

Note: To remove the Screen Saver dialog, use the "Prevent changing Screen Saver" setting.

9 Select **OK** to close the window.

10 Select **Screen saver timeout** from the Local Group Policy Editor window.



Screen saver timeout

Edit policy setting

Requirements:
At least Windows 2000 Service Pack 1

Description:
Specifies how much user idle time must elapse before the screen saver is launched.

When configured, this idle time can be set from a minimum of 1 second to a maximum of 86,400 seconds, or 24 hours. If set to zero, the screen saver will not be started.

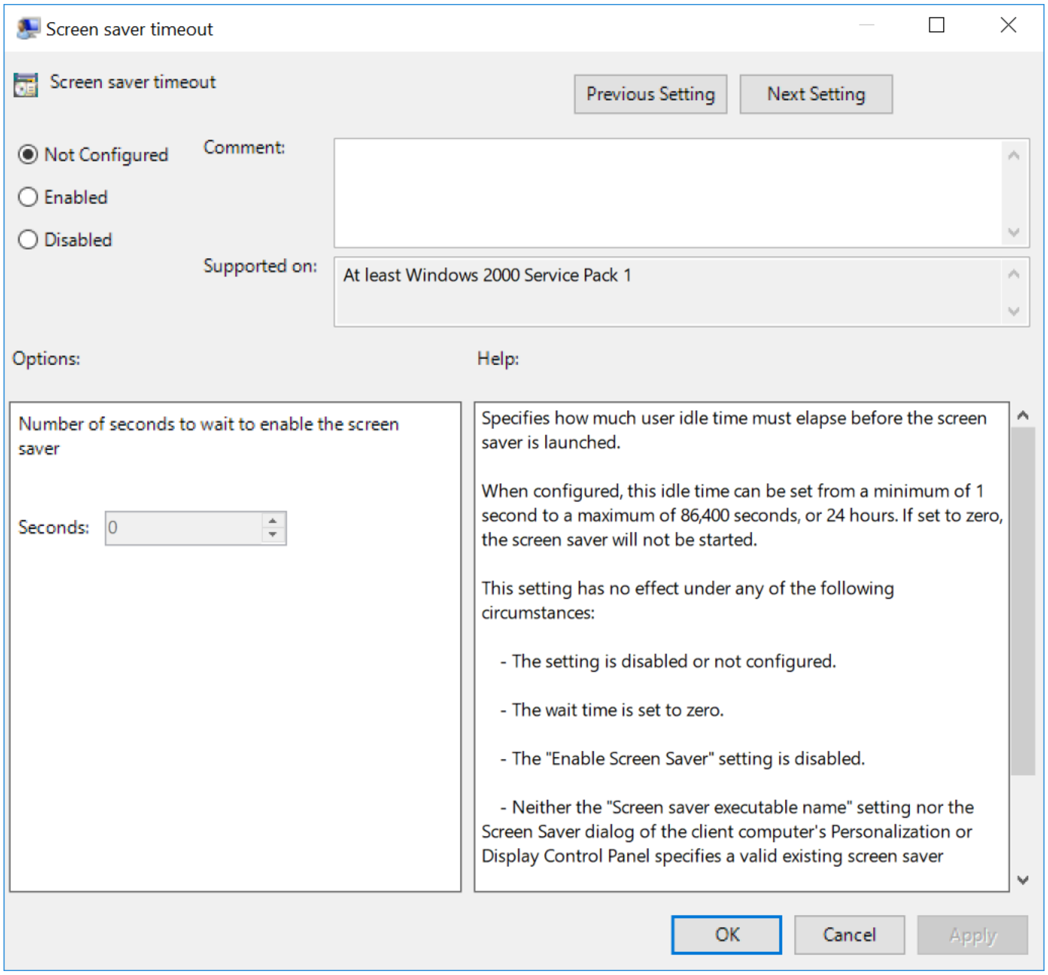
This setting has no effect under any of the following circumstances:

- The setting is disabled or not configured.

- The wait time is set to zero.

- The "Enable Screen Saver"

The Screen saver timeout window appears.



Screen saver timeout

Screen saver timeout

● Not Configured
○ Enabled
○ Disabled

Comment:

Supported on: At least Windows 2000 Service Pack 1

Options:

Number of seconds to wait to enable the screen saver

Seconds: 0

Help:

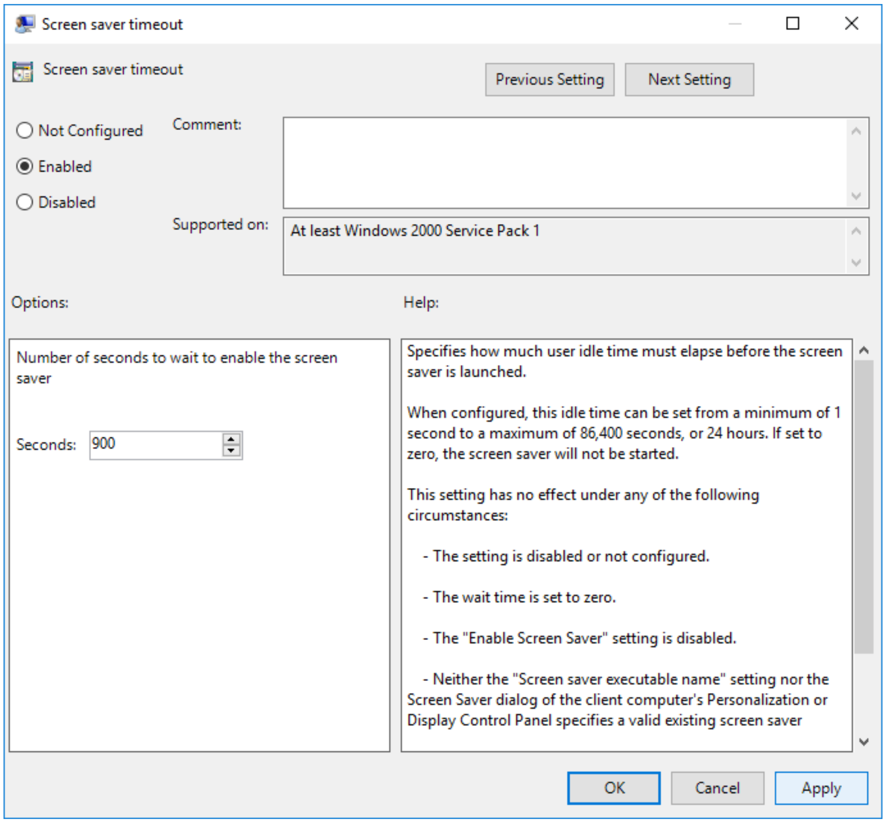
Specifies how much user idle time must elapse before the screen saver is launched.

When configured, this idle time can be set from a minimum of 1 second to a maximum of 86,400 seconds, or 24 hours. If set to zero, the screen saver will not be started.

This setting has no effect under any of the following circumstances:

- The setting is disabled or not configured.
- The wait time is set to zero.
- The "Enable Screen Saver" setting is disabled.
- Neither the "Screen saver executable name" setting nor the Screen Saver dialog of the client computer's Personalization or Display Control Panel specifies a valid existing screen saver

11 Select **Enabled** and **Apply**.



○ Not Configured
● Enabled
○ Disabled

Supported on: At least Windows 2000 Service Pack 1

Number of seconds to wait to enable the screen saver

Seconds: 900

Specifies how much user idle time must elapse before the screen saver is launched.

When configured, this idle time can be set from a minimum of 1 second to a maximum of 86,400 seconds, or 24 hours. If set to zero, the screen saver will not be started.

This setting has no effect under any of the following circumstances:

- The setting is disabled or not configured.

- The wait time is set to zero.

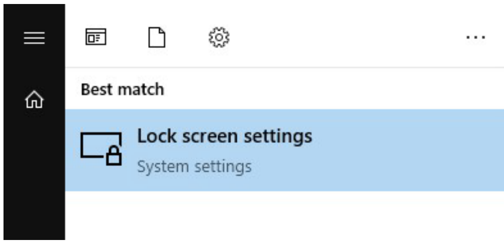
- The "Enable Screen Saver" setting is disabled.

- Neither the "Screen saver executable name" setting nor the Screen Saver dialog of the client computer's Personalization or Display Control Panel specifies a valid existing screen saver

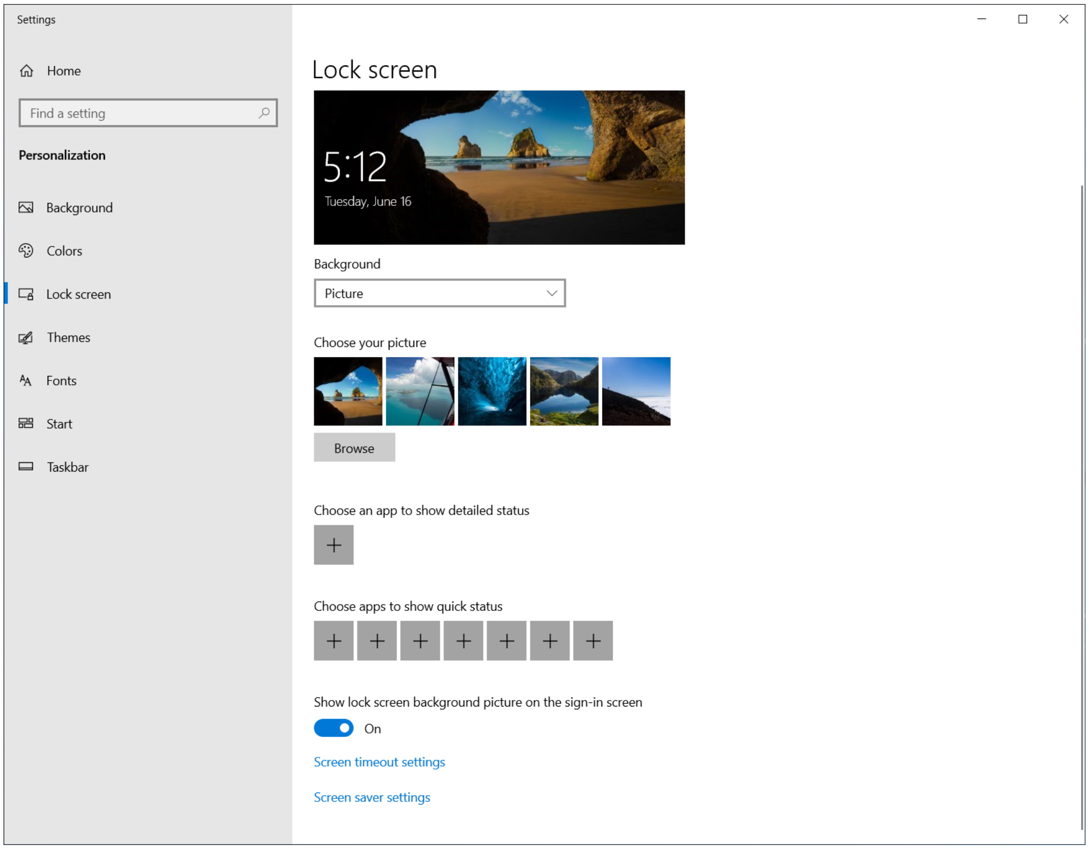
12 Select **OK** to close the window.

Setting Lock Screen

1 Type **Lock screen settings** from the Windows search bar to locate **Screen saver settings**.



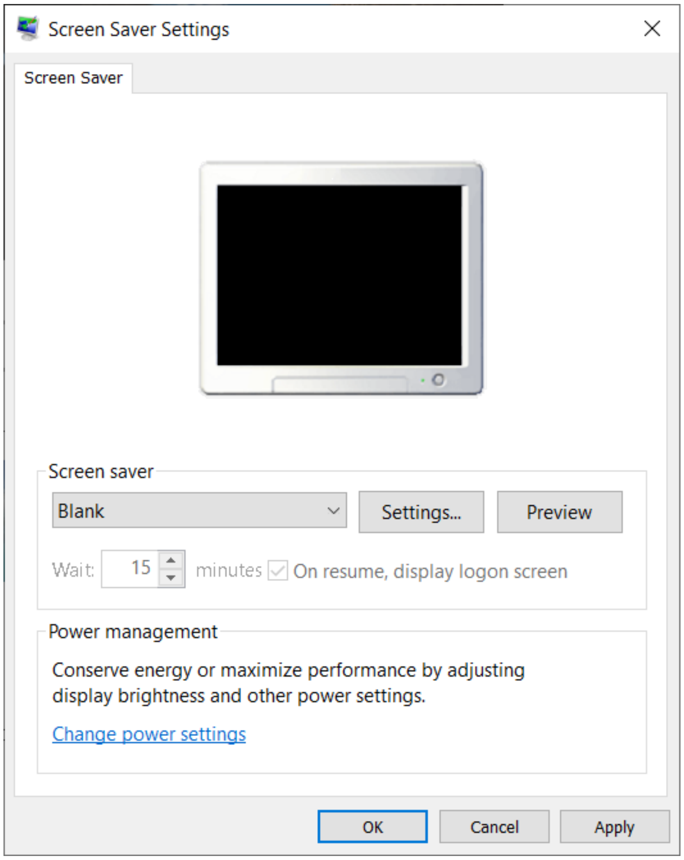
The Settings window appears.



Background
Picture

Show lock screen background picture on the sign-in screen
On

2 Scroll down to the bottom and select **Screen saver settings**. The Screen Saver Settings window appears.

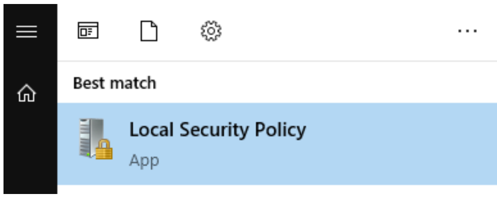

Screen Saver Settings
Screen Saver
Screen saver
Blank
Settings...
Preview
Wait: 15 minutes ☑ On resume, display logon screen
Power management
Conserve energy or maximize performance by adjusting display brightness and other power settings.
Change power settings
OK
Cancel
Apply

3 Select **Apply**.

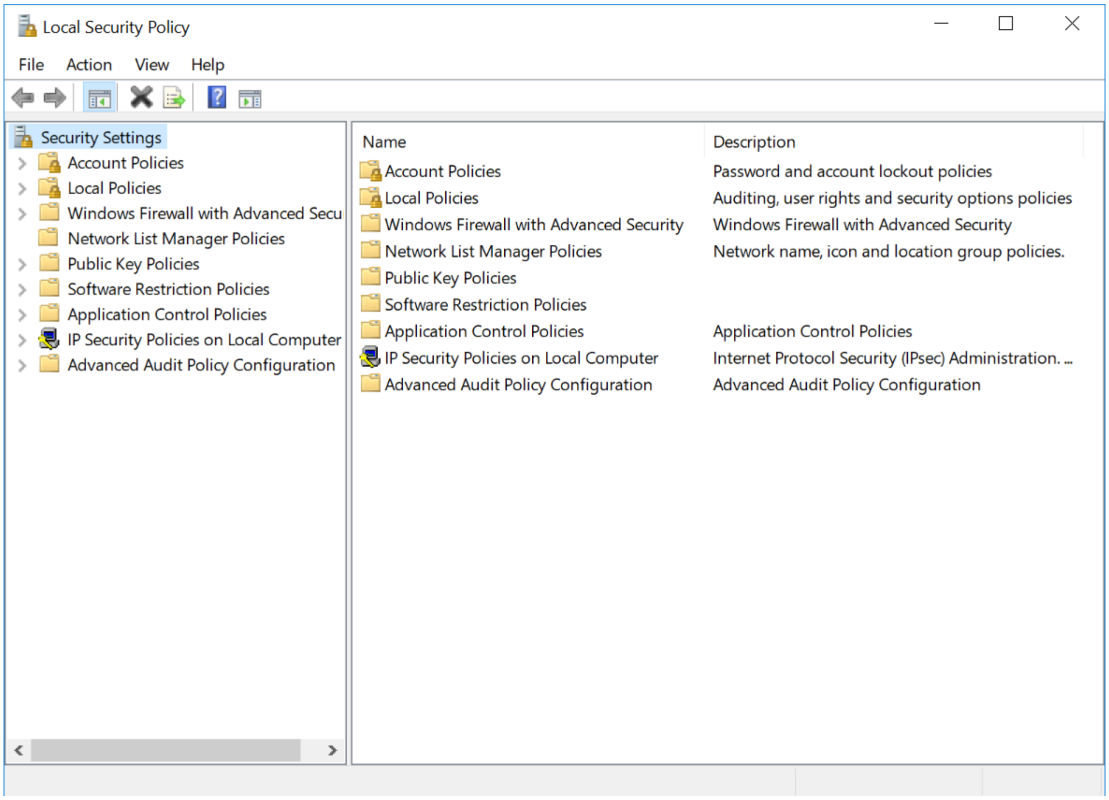
4 Select **OK** to close the window.

Setting Account Lockout Policy

1 Type **Local Security Policy** from the Windows search bar to locate the **Local Security Policy**.

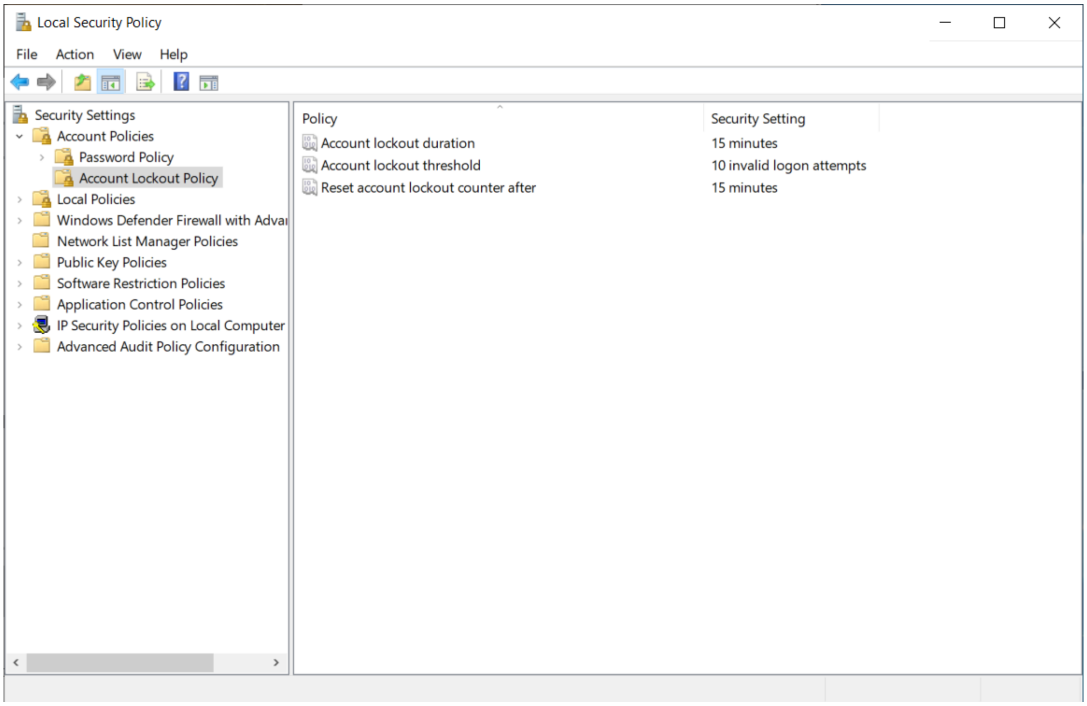


2 Double-click **Local Security Policy** to open the Local Security Policy window.


Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Local Policies
Windows Firewall with Advanced Secu
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Computer
Advanced Audit Policy Configuration

Name	Description
Account Policies	Password and account lockout policies
Local Policies	Auditing, user rights and security options policies
Windows Firewall with Advanced Security	Windows Firewall with Advanced Security
Network List Manager Policies	Network name, icon and location group policies.
Public Key Policies	
Software Restriction Policies	
Application Control Policies	Application Control Policies
IP Security Policies on Local Computer	Internet Protocol Security (IPsec) Administration. ...
Advanced Audit Policy Configuration	Advanced Audit Policy Configuration

3 Navigate to **Security Setting** > **Account Policies** > **Account Lockout Policy.** Configure the Security Settings as below.

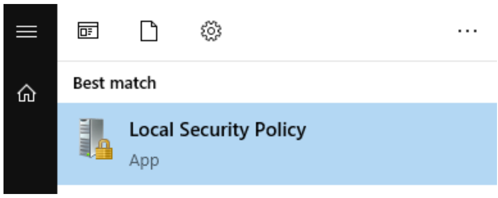

Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Password Policy
Account Lockout Policy
Local Policies
Windows Defender Firewall with Advai
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Computer
Advanced Audit Policy Configuration

Policy	Security Setting
Account lockout duration	15 minutes
Account lockout threshold	10 invalid logon attempts
Reset account lockout counter after	15 minutes

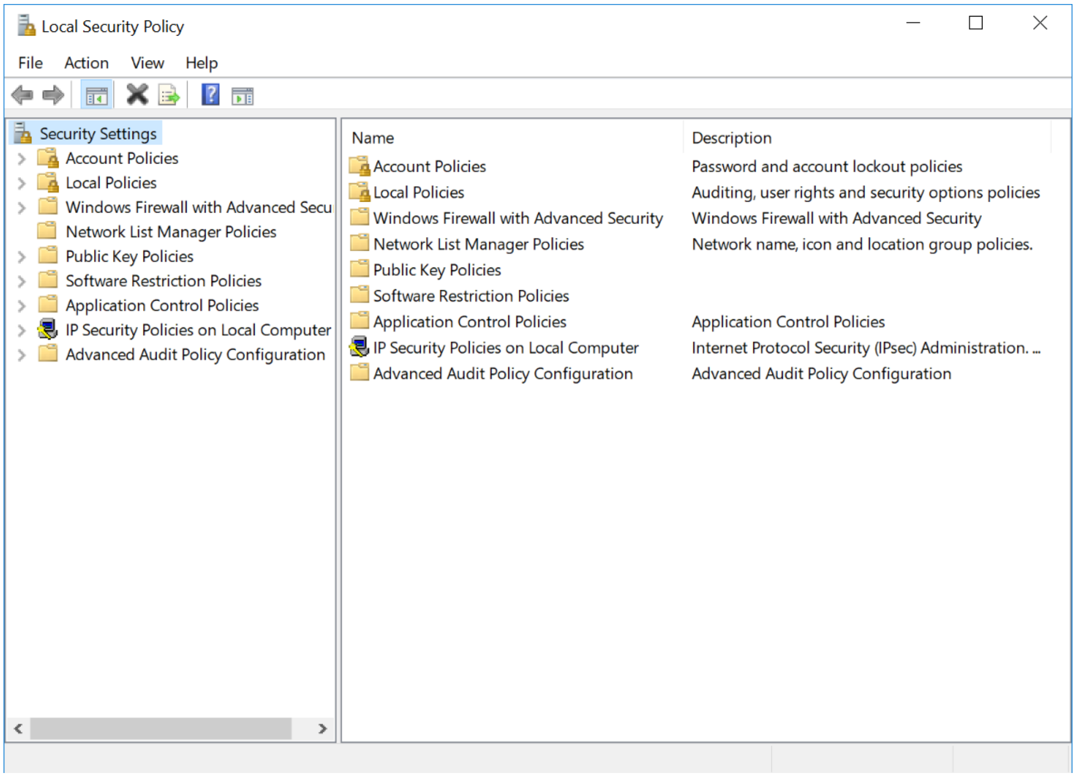
NOTE You must configure the **Account lockout threshold** first.

Setting Password Policy

1 Type **Local Security Policy** from the Windows search bar to locate the **Local Security Policy.**



2 Double-click **Local Security Policy** to open the Local Security Policy window.


Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Local Policies
Windows Firewall with Advanced Secu
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Computer
Advanced Audit Policy Configuration

Name	Description
Account Policies	Password and account lockout policies
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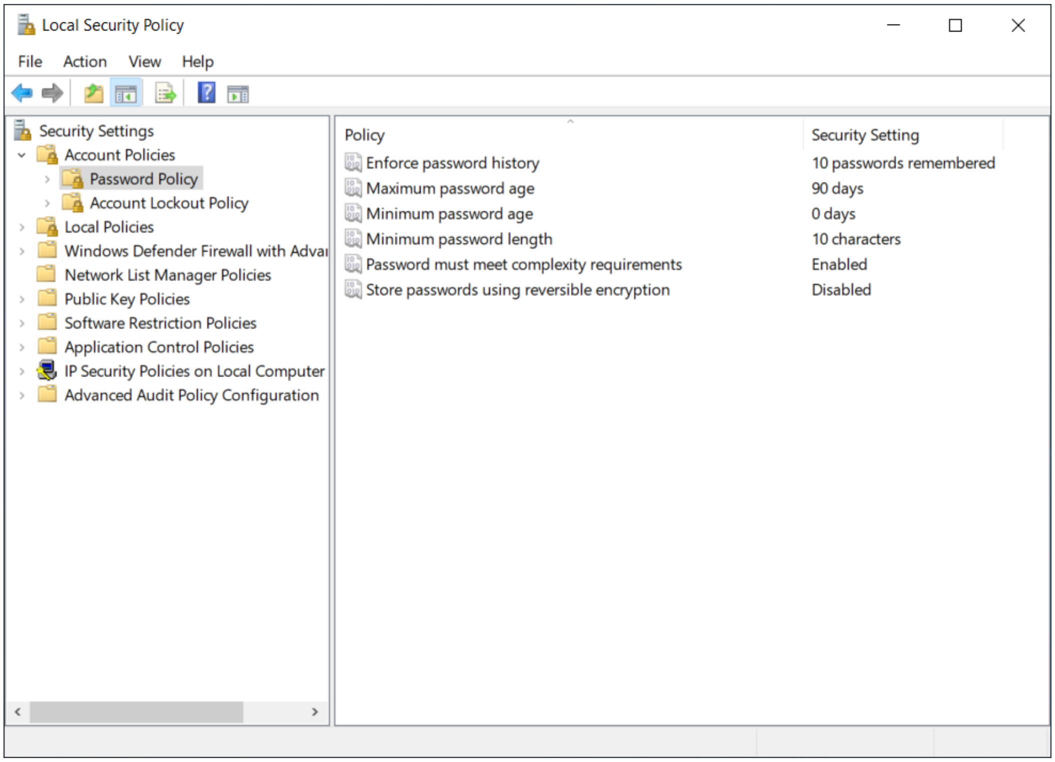
Public Key Policies	
Software Restriction Policies	
Application Control Policies	Application Control Policies
IP Security Policies on Local Computer	Internet Protocol Security (IPsec) Administration. ...
Advanced Audit Policy Configuration	Advanced Audit Policy Configuration

3 Navigate to **Security Setting** > **Account Policies** > **Password Policy**.


Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Password Policy
Account Lockout Policy
Local Policies
Windows Defender Firewall with Advai
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Computer
Advanced Audit Policy Configuration

Policy	Security Setting
Enforce password history	10 passwords remembered
Maximum password age	90 days
Minimum password age	0 days
Minimum password length	10 characters
Password must meet complexity requirements	Enabled
Store passwords using reversible encryption	Disabled

4 Customize the password policy according to the following.

- **Enforce password history:** 10 passwords remembered
- **Maximum password age:** 90 days
- **Minimum password age:** optional
- **Minimum password length:** 10 characters
- **Password must meet complexity requirements:** enabled

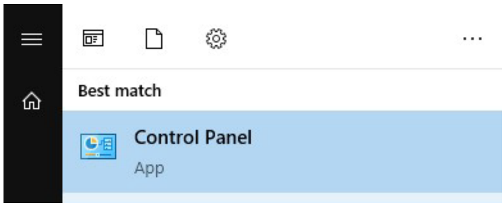
  The password must contain all four of the following character types:

  - letter in upper case (A -Z)
  - letter in lower case (a -z)
  - numbers (0-9)
  - distinctive character (i.e.,,!, @, #, $,%, ^, &).

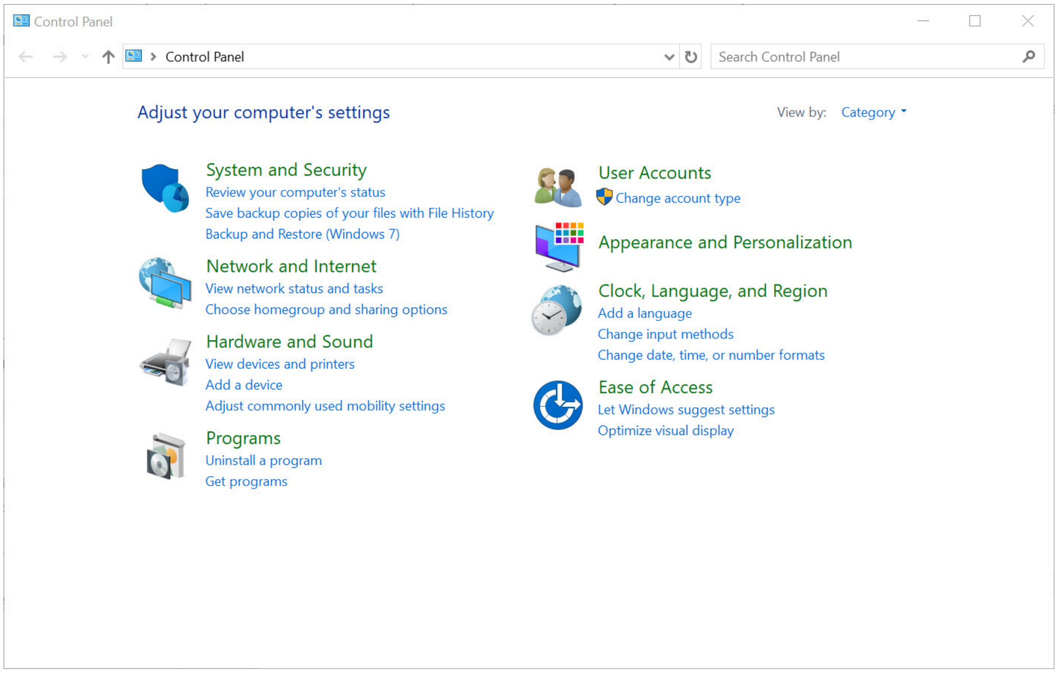
- **Store passwords using reversible encryption:** optional

Disabling Remote Access

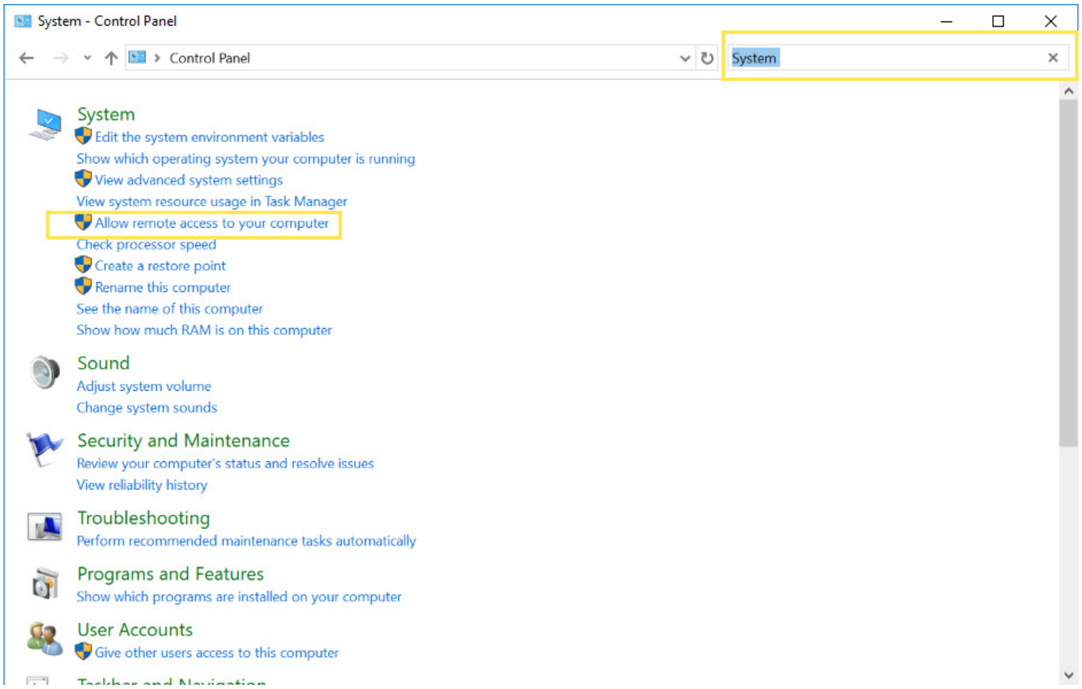
1 Type **Control Panel** from the Windows search bar to locate the **Control Panel** app.



2 Select **Control Panel** to open the Control Panel window.


Control Panel
Control Panel
Search Control Panel
Adjust your computer's settings
View by: Category
System and Security
Review your computer's status
Save backup copies of your files with File History
Backup and Restore (Windows 7)
Network and Internet
View network status and tasks
Choose homegroup and sharing options
Hardware and Sound
View devices and printers
Add a device
Adjust commonly used mobility settings
Programs
Uninstall a program
Get programs
User Accounts
Change account type
Appearance and Personalization
Clock, Language, and Region
Add a language
Change input methods
Change date, time, or number formats
Ease of Access
Let Windows suggest settings
Optimize visual display

3 Type **System** from the search bar to locate **Allow remote access to your computer**.



System - Control Panel

Control Panel

System

System
- Edit the system environment variables
- Show which operating system your computer is running
- View advanced system settings
- View system resource usage in Task Manager
- Allow remote access to your computer
- Check processor speed
- Create a restore point
- Rename this computer
- See the name of this computer
- Show how much RAM is on this computer

Sound
- Adjust system volume
- Change system sounds

Security and Maintenance
- Review your computer's status and resolve issues
- View reliability history

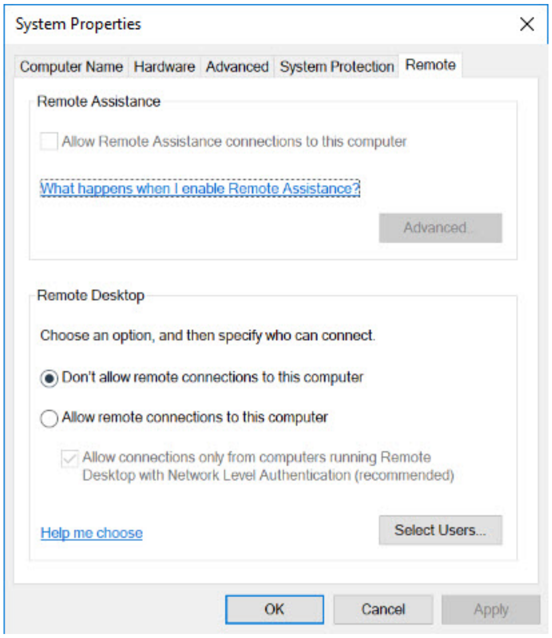
Troubleshooting
- Perform recommended maintenance tasks automatically

Programs and Features
- Show which programs are installed on your computer

User Accounts
- Give other users access to this computer

---

4 Select **Allow remote access to your computer.** The System Properties window appears.



System Properties

Computer Name | Hardware | Advanced | System Protection | Remote

Remote Assistance

☐ Allow Remote Assistance connections to this computer

What happens when I enable Remote Assistance?

Advanced...

Remote Desktop

Choose an option, and then specify who can connect.

● Don't allow remote connections to this computer

○ Allow remote connections to this computer

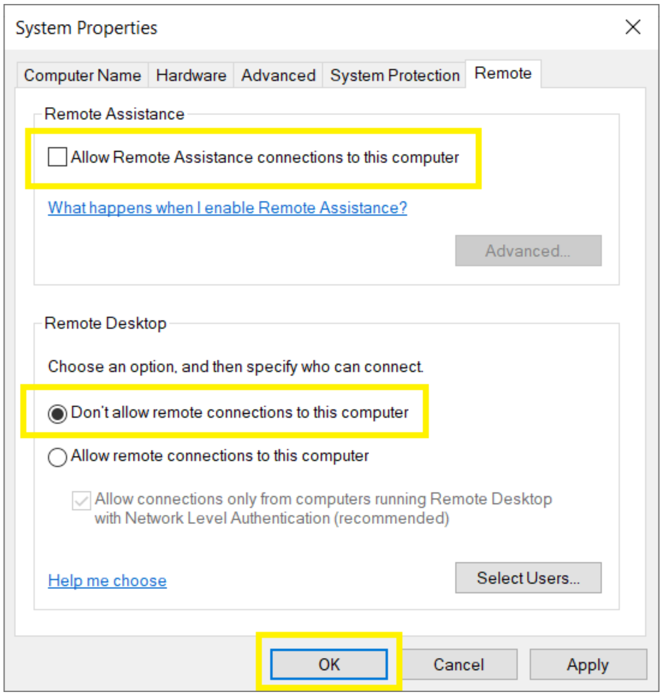
☑ Allow connections only from computers running Remote Desktop with Network Level Authentication (recommended)

Help me choose

Select Users...

OK | Cancel | Apply

5 Select **Don't allow remote connections to this computer** from the Remote tab.



System Properties

Computer Name | Hardware | Advanced | System Protection | Remote

Remote Assistance

☐ Allow Remote Assistance connections to this computer

What happens when I enable Remote Assistance?

Advanced...

Remote Desktop

Choose an option, and then specify who can connect.

● Don't allow remote connections to this computer

○ Allow remote connections to this computer

☑ Allow connections only from computers running Remote Desktop with Network Level Authentication (recommended)

Help me choose

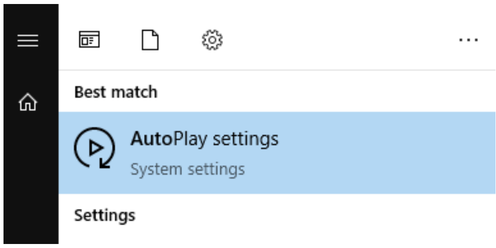
Select Users...

OK Cancel Apply

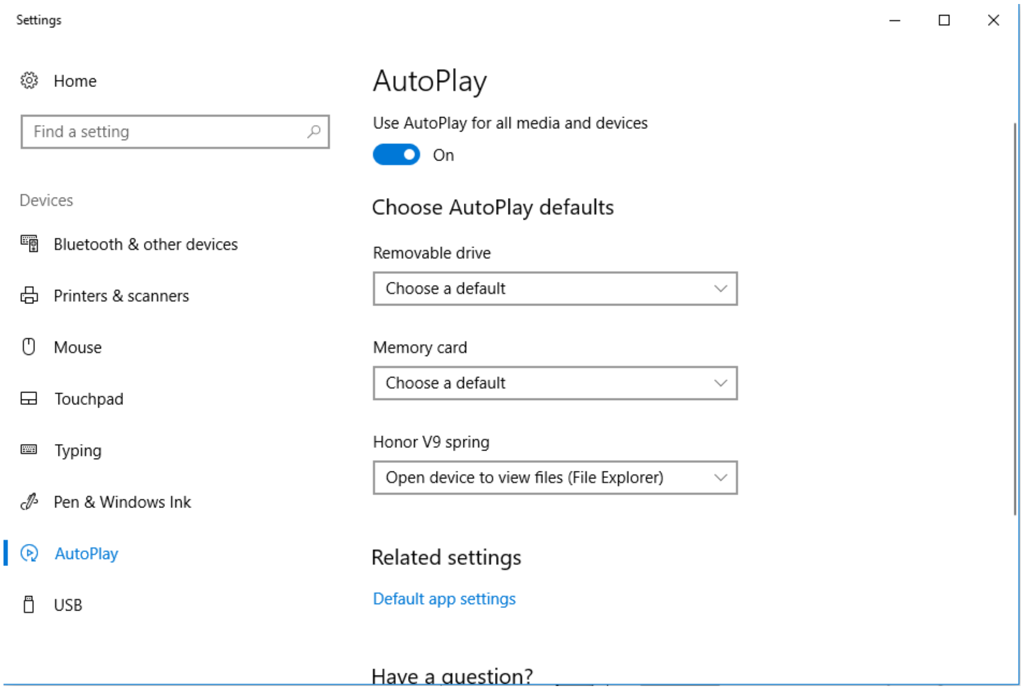
6 Select **OK** to close the window.

Disabling Auto Play

1 Type **Auto Play settings** from the Windows search bar to locate the **Auto Play settings.**



2 Select **Auto Play settings.** The Settings window appears.



AutoPlay

Use AutoPlay for all media and devices: On

Choose AutoPlay defaults

Removable drive: Choose a default

Memory card: Choose a default

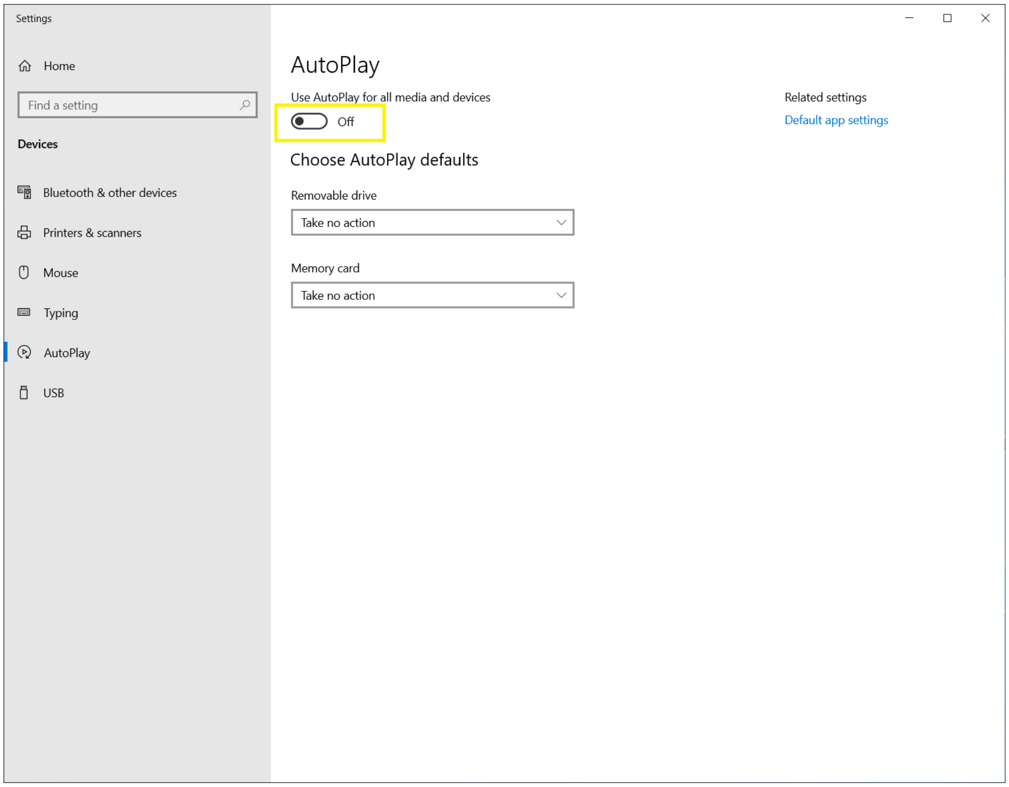
Honor V9 spring: Open device to view files (File Explorer)

Related settings

Default app settings

Have a question?

3 Select **Off** to turn off Auto Play.



Use AutoPlay for all media and devices: Off

Removable drive: Take no action

Memory card: Take no action

4 Close the window.

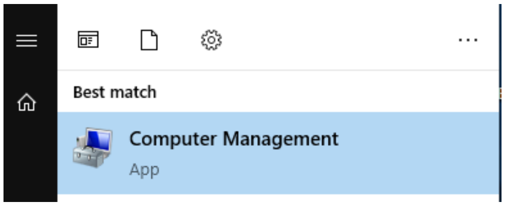
Disabling Unnecessary Services

It is recommended to disable all the unnecessary services while operating the CytExpert for Spectral software.

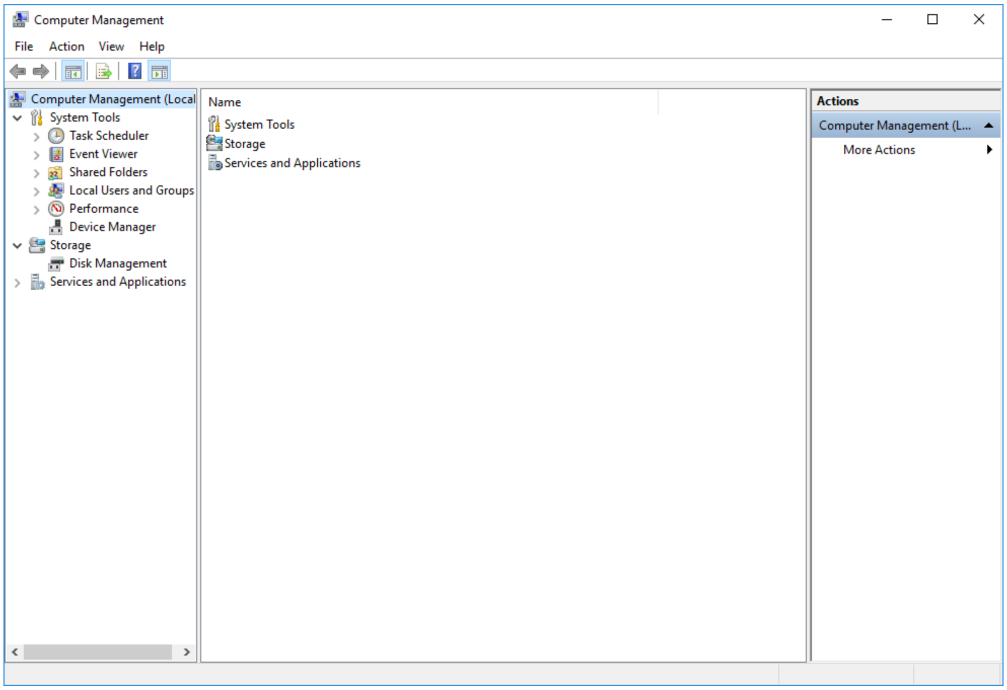
Table E.2 List of Unnecessary Services

Bluetooth Support Service	Microsoft FTP Service (If available)	Windows Media Player Network Sharing Service
Geo-location Service	Microsoft iSCSI Initiator Service	Windows Mobile Hotspot Service
Infrared Monitor Service	Remote Procedure Call (RPC) Locator	WinHTTP Web Proxy Auto-Discovery Service
Internet Connection Sharing	Routing and Remote Access	Xbox Live Game Save
Link-Layer Topology Discovery Mapper	SSDP Discovery	Xbox Live Networking Service
LxssManager (If available)	UPnP Device Host	

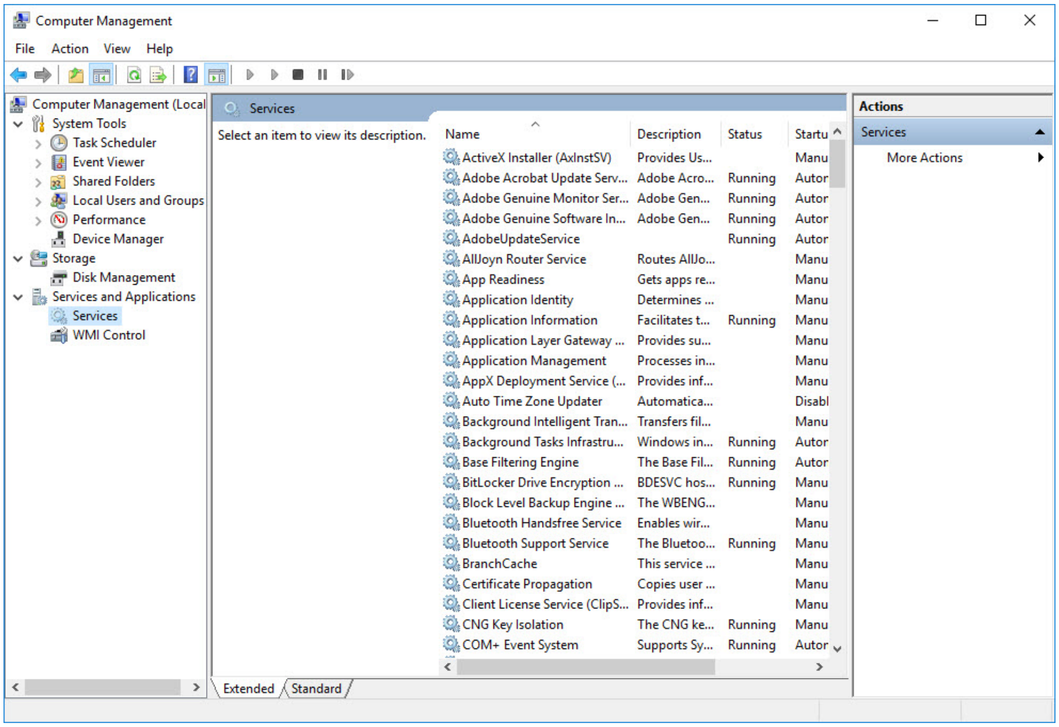
1 Type **Computer Management** from the Windows search bar to locate the Computer Management App.



2 Select **Computer Management**.



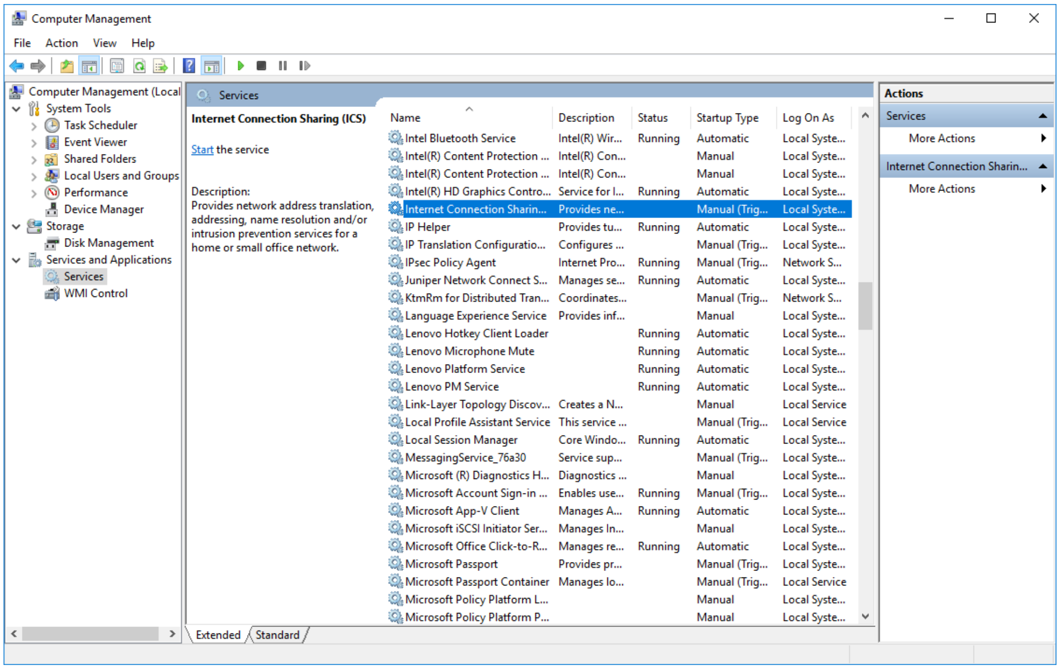
3 Navigate to **Services and Applications** > **Services**.



Select an item to view its description.

Name	Description	Status	Startu
ActiveX Installer (AxInstSV)	Provides Us...		Manu
Adobe Acrobat Update Serv...	Adobe Acro...	Running	Autor
Adobe Genuine Monitor Ser...	Adobe Gen...	Running	Autor
Adobe Genuine Software In...	Adobe Gen...	Running	Autor
AdobeUpdateService		Running	Autor
AllJoyn Router Service	Routes AllJo...		Manu
App Readiness	Gets apps re...		Manu
Application Identity	Determines ...		Manu
Application Information	Facilitates t...	Running	Manu
Application Layer Gateway ...	Provides su...		Manu
Application Management	Processes in...		Manu
AppX Deployment Service (...	Provides inf...		Manu
Auto Time Zone Updater	Automatica...		Disabl
Background Intelligent Tran...	Transfers fil...		Manu
Background Tasks Infrastru...	Windows in...	Running	Autor
Base Filtering Engine	The Base Fil...	Running	Autor
BitLocker Drive Encryption ...	BDESVC hos...	Running	Manu
Block Level Backup Engine ...	The WBENG...		Manu
Bluetooth Handsfree Service	Enables wir...		Manu
Bluetooth Support Service	The Bluetoo...	Running	Manu
BranchCache	This service ...		Manu
Certificate Propagation	Copies user ...		Manu
Client License Service (ClipS...	Provides inf...		Manu
CNG Key Isolation	The CNG ke...	Running	Manu
COM+ Event System	Supports Sy...	Running	Autor

4 Double-click the service you want to disable.



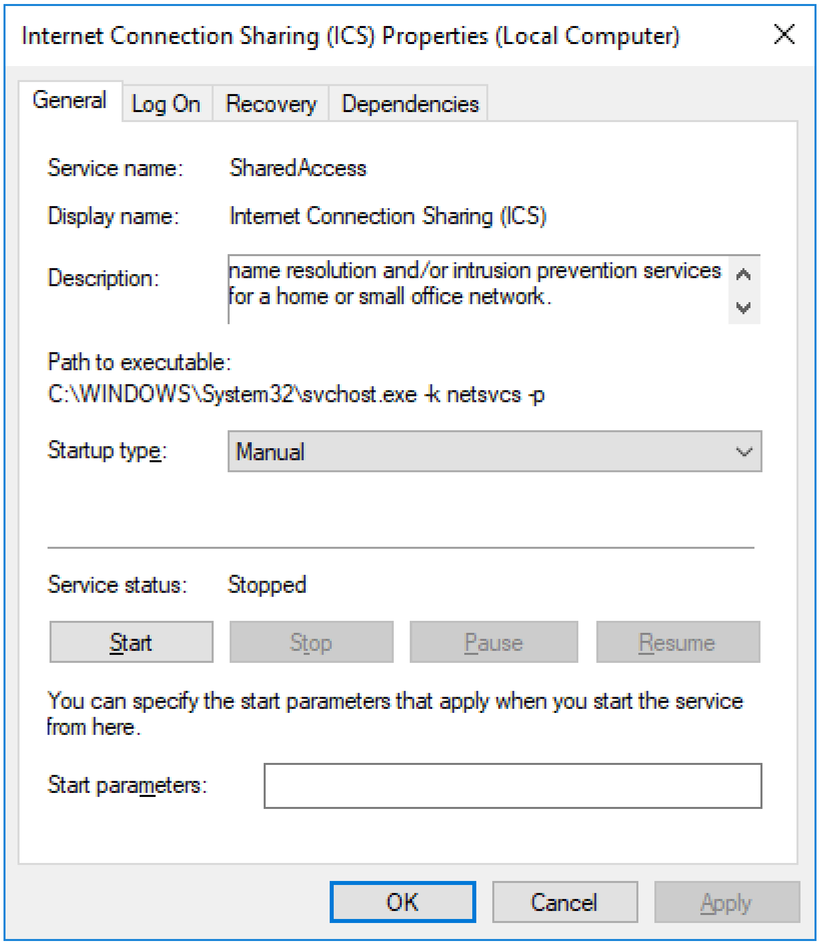
Internet Connection Sharing (ICS)

Start the service

Description:
Provides network address translation, addressing, name resolution and/or intrusion prevention services for a home or small office network.

Name	Description	Status	Startup Type	Log On As
Intel Bluetooth Service	Intel(R) Wir...	Running	Automatic	Local Syste...
Intel(R) Content Protection ...	Intel(R) Con...		Manual	Local Syste...
Intel(R) Content Protection ...	Intel(R) Con...		Manual	Local Syste...
Intel(R) HD Graphics Contro...	Service for I...	Running	Automatic	Local Syste...
Internet Connection Sharin...	Provides ne...		Manual (Trig...	Local Syste...
IP Helper	Provides tu...	Running	Automatic	Local Syste...
IP Translation Configuratio...	Configures ...		Manual (Trig...	Local Syste...
IPsec Policy Agent	Internet Pro...	Running	Manual (Trig...	Network S...
Juniper Network Connect S...	Manages se...	Running	Automatic	Local Syste...
KtmRm for Distributed Tran...	Coordinates...		Manual (Trig...	Network S...
Language Experience Service	Provides inf...		Manual	Local Syste...
Lenovo Hotkey Client Loader		Running	Automatic	Local Syste...
Lenovo Microphone Mute		Running	Automatic	Local Syste...
Lenovo Platform Service		Running	Automatic	Local Syste...
Lenovo PM Service		Running	Automatic	Local Syste...
Link-Layer Topology Discov...	Creates a N...		Manual	Local Service
Local Profile Assistant Service	This service ...		Manual (Trig...	Local Service
Local Session Manager	Core Windo...	Running	Automatic	Local Syste...
MessagingService_76a30	Service sup...		Manual (Trig...	Local Syste...
Microsoft (R) Diagnostics H...	Diagnostics ...		Manual	Local Syste...
Microsoft Account Sign-in ...	Enables use...	Running	Manual (Trig...	Local Syste...
Microsoft App-V Client	Manages A...	Running	Automatic	Local Syste...
Microsoft iSCSI Initiator Ser...	Manages In...		Manual	Local Syste...
Microsoft Office Click-to-R...	Manages re...	Running	Automatic	Local Syste...
Microsoft Passport	Provides pr...		Manual (Trig...	Local Syste...
Microsoft Passport Container	Manages lo...		Manual (Trig...	Local Service
Microsoft Policy Platform L...			Manual	Local Syste...
Microsoft Policy Platform P...			Manual	Local Syste...

The following window appears.



Internet Connection Sharing (ICS) Properties (Local Computer)

Service name: SharedAccess

Display name: Internet Connection Sharing (ICS)

Description: name resolution and/or intrusion prevention services for a home or small office network.

Path to executable:
C:\WINDOWS\System32\svchost.exe -k netsvcs -p

Startup type: Manual

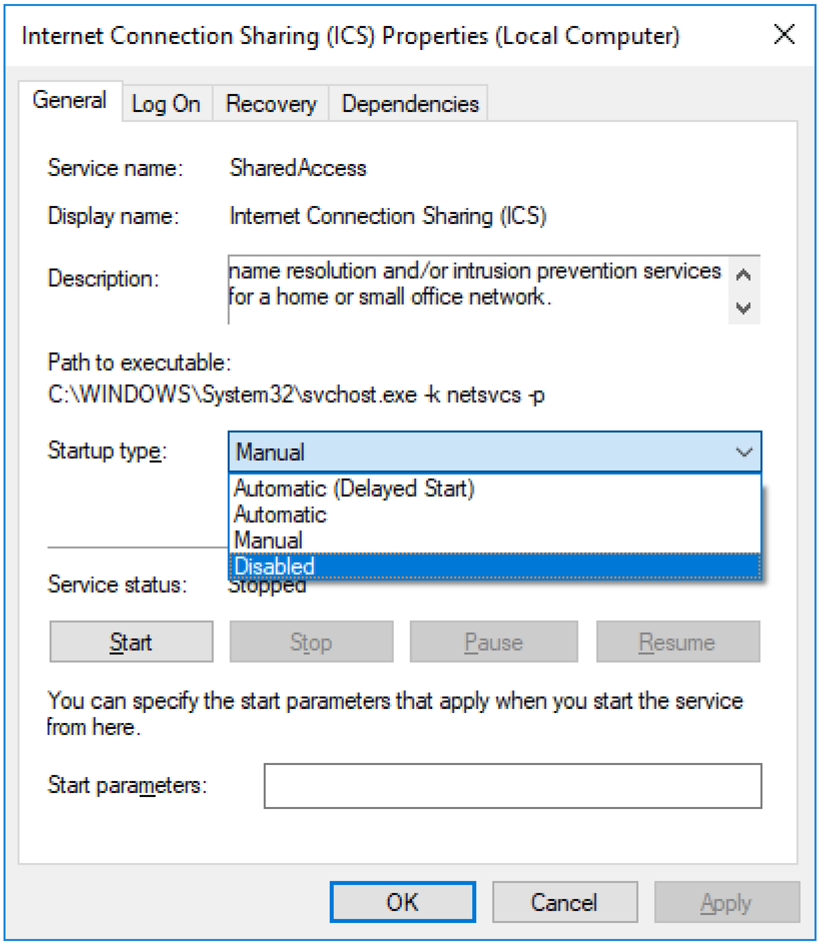
Service status: Stopped

You can specify the start parameters that apply when you start the service from here.

Start parameters:

---

5 Select **Disabled** from the Startup type dropdown.



Internet Connection Sharing (ICS) Properties (Local Computer)

Service name: SharedAccess

Display name: Internet Connection Sharing (ICS)

Description: name resolution and/or intrusion prevention services for a home or small office network.

Path to executable:
C:\WINDOWS\System32\svchost.exe -k netsvcs -p

Startup type: Manual

Service status: Stopped

You can specify the start parameters that apply when you start the service from here.

Start parameters:

---

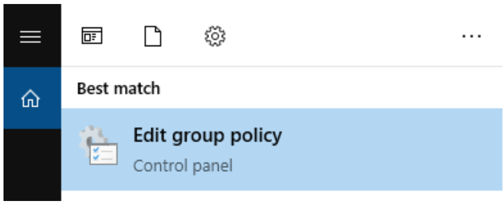
6 Select **Apply**.

---

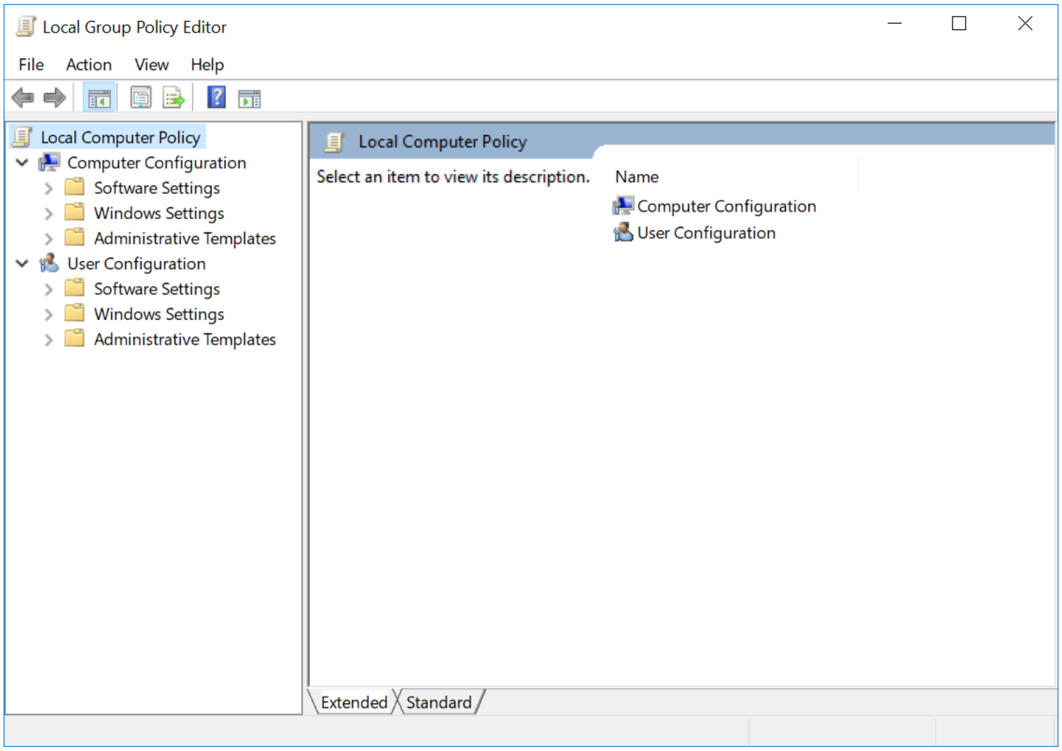
7 Select **OK** to close the window.

Disabling Unauthorized Applications

1 Type **Edit group policy** from the Windows search bar to locate the **Local group policy editor** control panel.



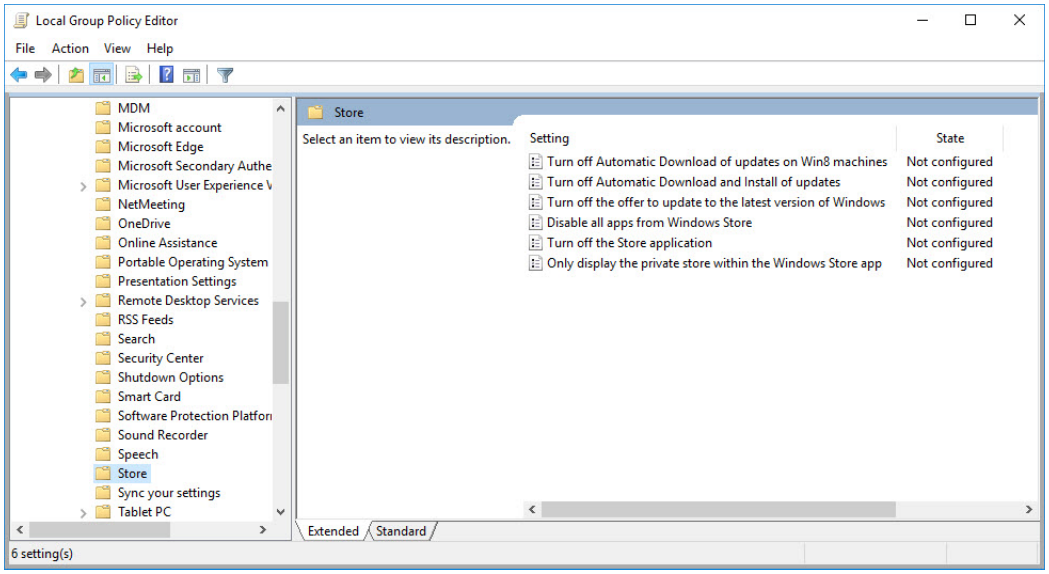
The Local Group Policy Editor window displays.



Select an item to view its description.

---

2 Navigate to **Computer Configuration > Administrative Templates > Windows Components > Store**.

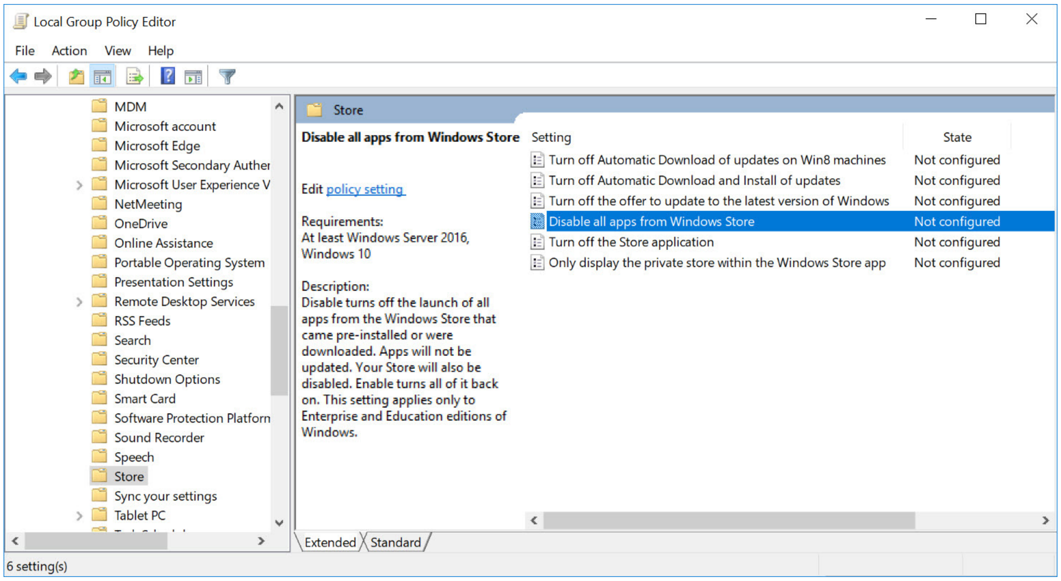


Select an item to view its description.

Setting	State
Turn off Automatic Download of updates on Win8 machines	Not configured
Turn off Automatic Download and Install of updates	Not configured
Turn off the offer to update to the latest version of Windows	Not configured
Disable all apps from Windows Store	Not configured
Turn off the Store application	Not configured
Only display the private store within the Windows Store app	Not configured

---

3 Select **Disable all apps from Windows Store**.



Disable all apps from Windows Store

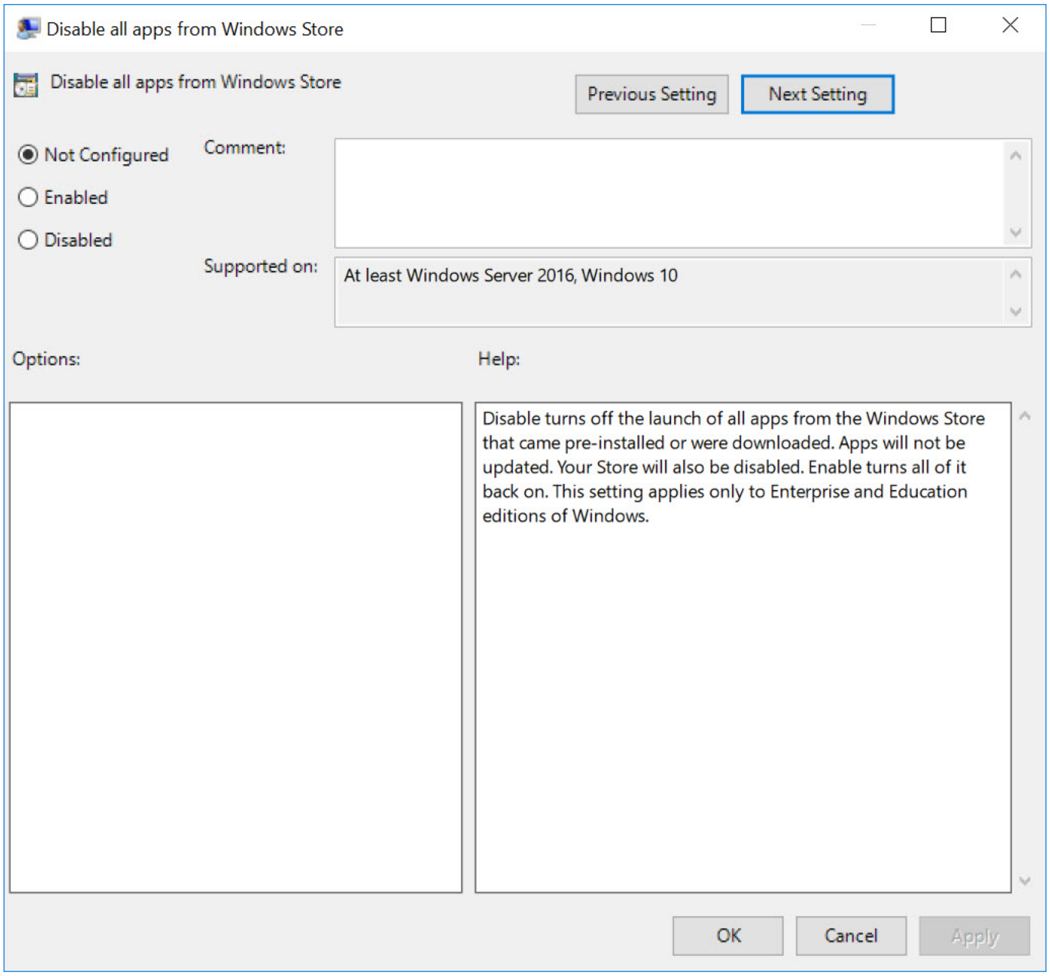
Edit policy setting

Requirements:
At least Windows Server 2016, Windows 10

Description:
Disable turns off the launch of all apps from the Windows Store that came pre-installed or were downloaded. Apps will not be updated. Your Store will also be disabled. Enable turns all of it back on. This setting applies only to Enterprise and Education editions of Windows.

Setting	State
Turn off Automatic Download of updates on Win8 machines	Not configured
Turn off Automatic Download and Install of updates	Not configured
Turn off the offer to update to the latest version of Windows	Not configured
Disable all apps from Windows Store	Not configured
Turn off the Store application	Not configured
Only display the private store within the Windows Store app	Not configured

The following window appears.



At least Windows Server 2016, Windows 10

Disable turns off the launch of all apps from the Windows Store that came pre-installed or were downloaded. Apps will not be updated. Your Store will also be disabled. Enable turns all of it back on. This setting applies only to Enterprise and Education editions of Windows.

---

4 Select **Enabled** and **Apply**.

---

5 Select **OK** to close the window.

---

6 Disable the following applications. Refer to Steps 3-5.

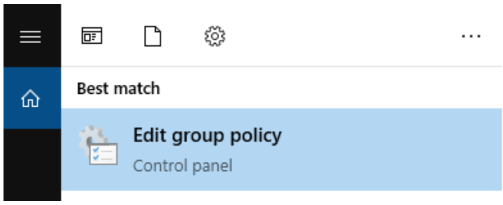
- **Turn off Automatic Download and Install of updates.**
- **Turn off the offer to update to the latest version of Windows.**
- **Turn off the Store application.**

Enabling SMB Digitally Sign Communications

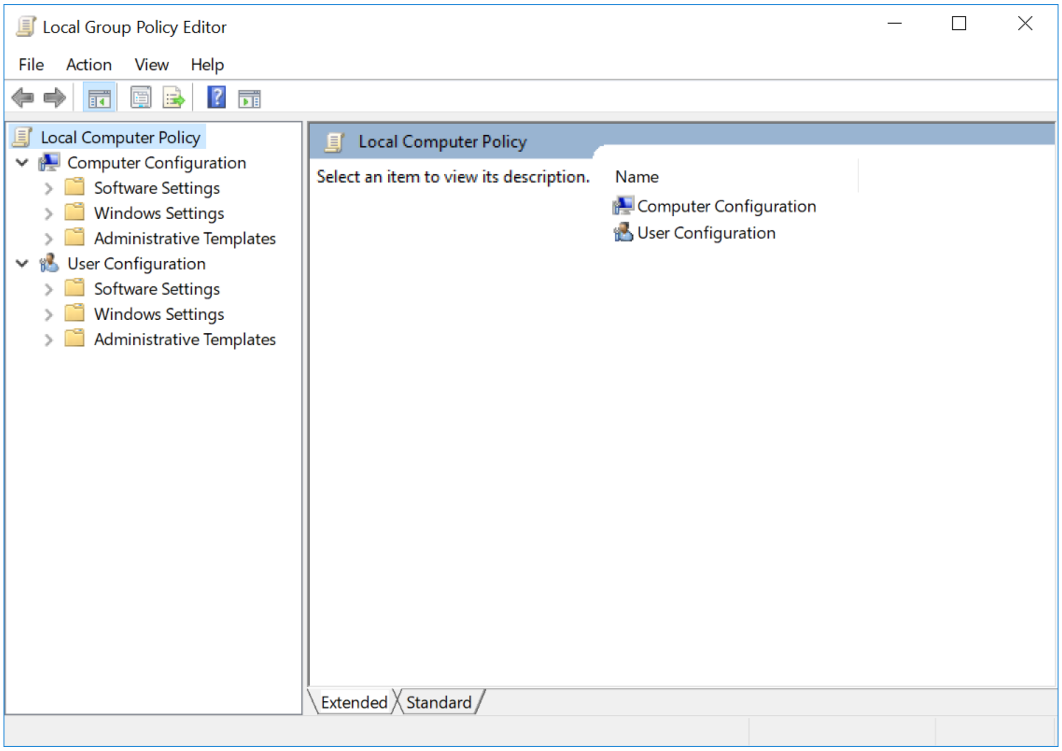
The Server Message Block (SMB) protocol provides the basis for file and print sharing and many other networking operations, such as remote Windows administration. To prevent man-in-the-middle attacks that modify SMB packets in transit, follow the instructions below to enable the SMB Digitally Sign Communications.

---

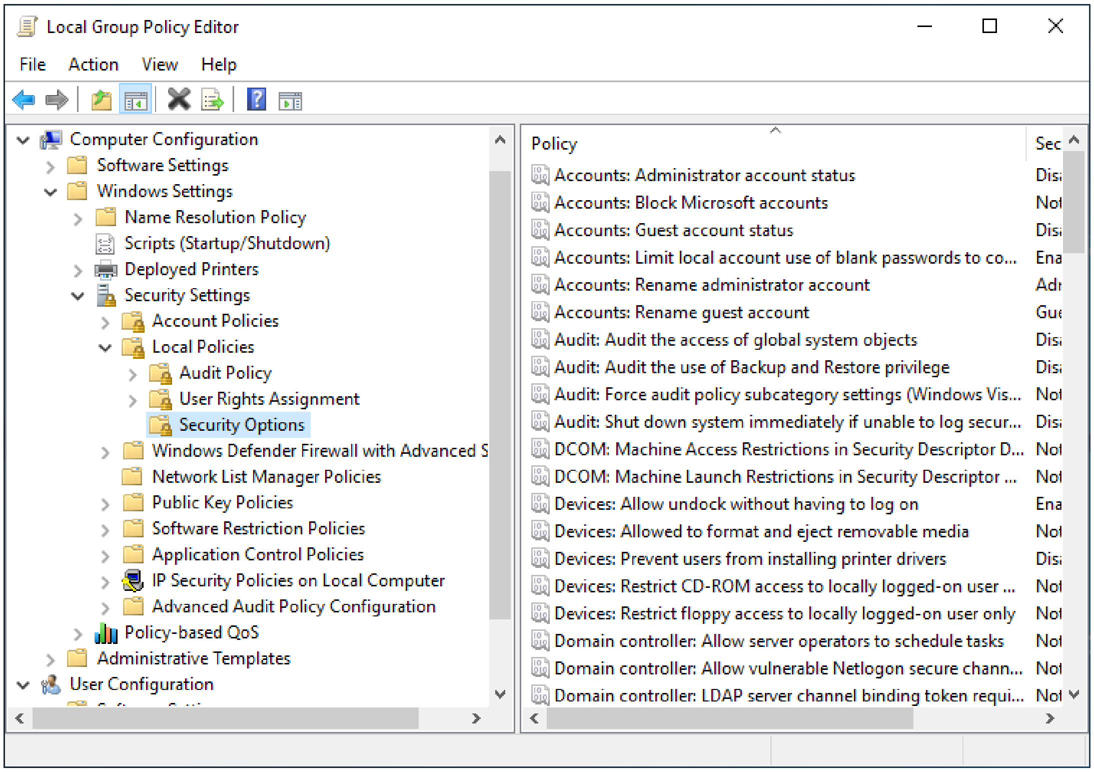
1 Type **Edit group policy** from the Windows search bar to locate the **Local group policy editor** control panel.



The Local Group Policy Editor window displays.



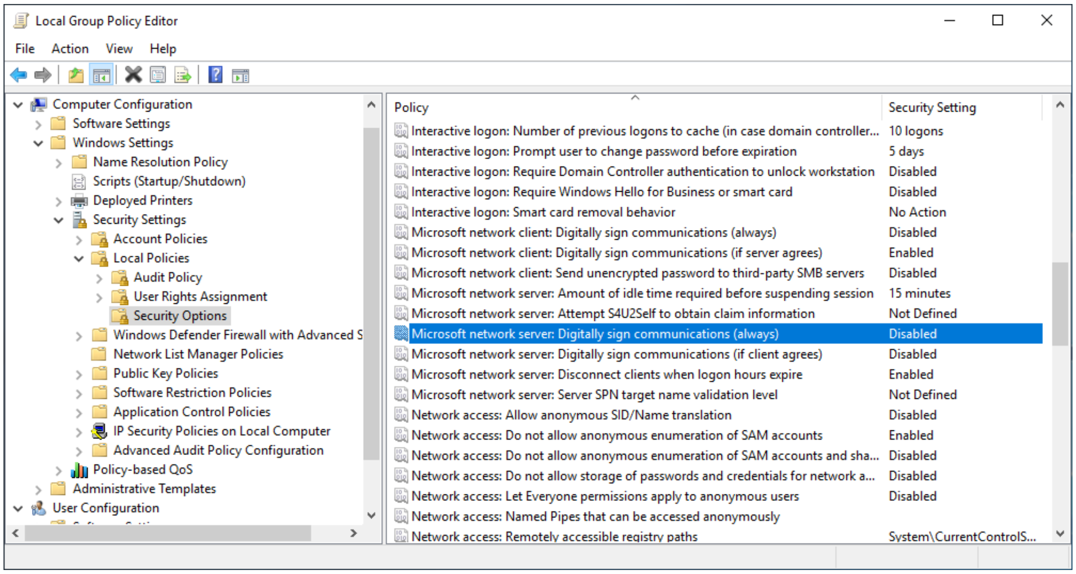
2 Navigate to **Computer Configuration** > **Windows Settings** > **Security Settings** > **Local Policies** > **Security Options.**



Policy	Sec
Accounts: Administrator account status	Dis
Accounts: Block Microsoft accounts	Not
Accounts: Guest account status	Dis
Accounts: Limit local account use of blank passwords to co...	Ena
Accounts: Rename administrator account	Adr
Accounts: Rename guest account	Gue
Audit: Audit the access of global system objects	Dis
Audit: Audit the use of Backup and Restore privilege	Dis
Audit: Force audit policy subcategory settings (Windows Vis...	Not
Audit: Shut down system immediately if unable to log secur...	Dis
DCOM: Machine Access Restrictions in Security Descriptor D...	Not
DCOM: Machine Launch Restrictions in Security Descriptor ...	Not
Devices: Allow undock without having to log on	Ena
Devices: Allowed to format and eject removable media	Not
Devices: Prevent users from installing printer drivers	Dis
Devices: Restrict CD-ROM access to locally logged-on user ...	Not
Devices: Restrict floppy access to locally logged-on user only	Not
Domain controller: Allow server operators to schedule tasks	Not
Domain controller: Allow vulnerable Netlogon secure chann...	Not
Domain controller: LDAP server channel binding token requi...	Not

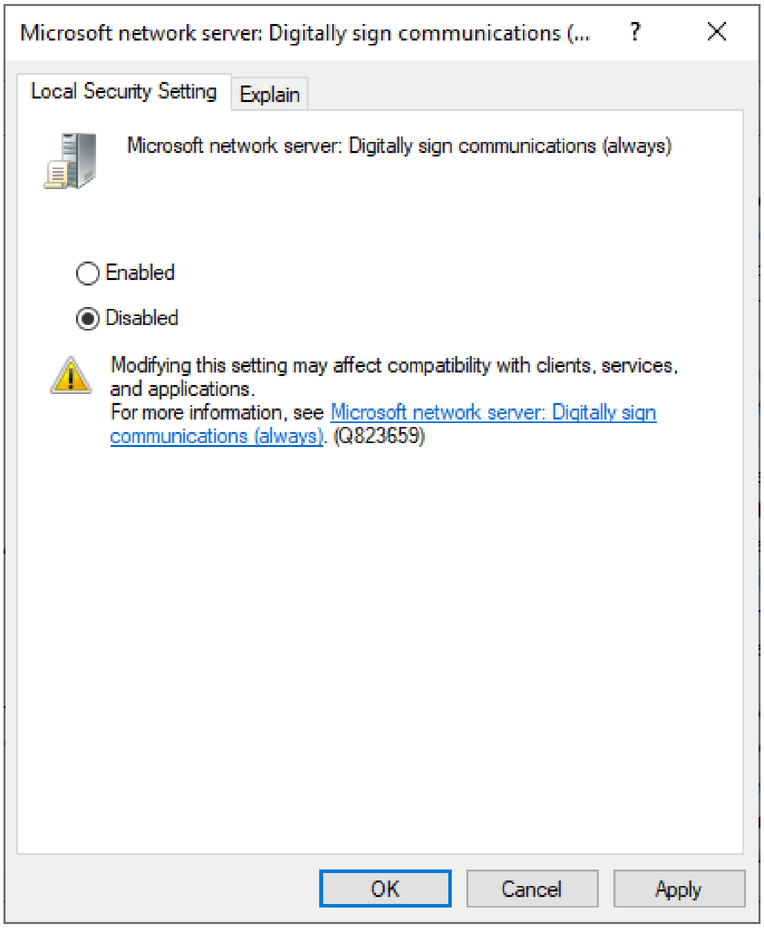
---

3 Select **Microsoft network server: Digitally sign communications (always).**



Policy	Security Setting
Interactive logon: Number of previous logons to cache (in case domain controller...	10 logons
Interactive logon: Prompt user to change password before expiration	5 days
Interactive logon: Require Domain Controller authentication to unlock workstation	Disabled
Interactive logon: Require Windows Hello for Business or smart card	Disabled
Interactive logon: Smart card removal behavior	No Action
Microsoft network client: Digitally sign communications (always)	Disabled
Microsoft network client: Digitally sign communications (if server agrees)	Enabled
Microsoft network client: Send unencrypted password to third-party SMB servers	Disabled
Microsoft network server: Amount of idle time required before suspending session	15 minutes
Microsoft network server: Attempt S4U2Self to obtain claim information	Not Defined
Microsoft network server: Digitally sign communications (always)	Disabled
Microsoft network server: Digitally sign communications (if client agrees)	Disabled
Microsoft network server: Disconnect clients when logon hours expire	Enabled
Microsoft network server: Server SPN target name validation level	Not Defined
Network access: Allow anonymous SID/Name translation	Disabled
Network access: Do not allow anonymous enumeration of SAM accounts	Enabled
Network access: Do not allow anonymous enumeration of SAM accounts and sha...	Disabled
Network access: Do not allow storage of passwords and credentials for network a...	Disabled
Network access: Let Everyone permissions apply to anonymous users	Disabled
Network access: Named Pipes that can be accessed anonymously	
Network access: Remotely accessible registry paths	System\CurrentControlS...

The following window appears.


Microsoft network server: Digitally sign communications (...
?
Local Security Setting
Explain
OK
Cancel
Apply

Microsoft network server: Digitally sign communications (always)

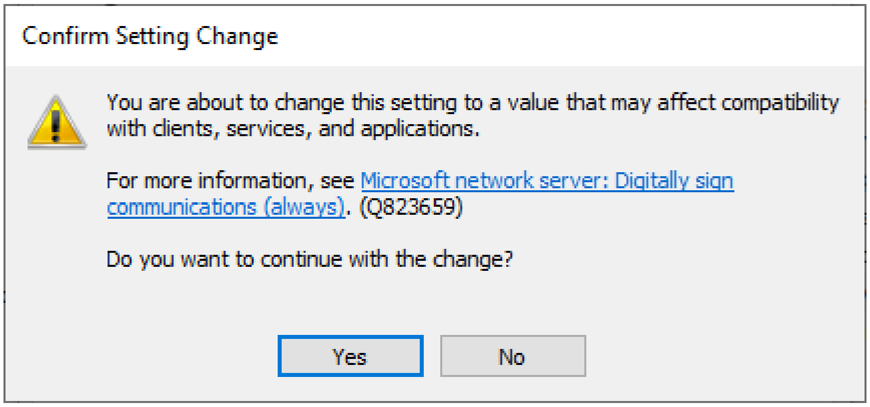
○ Enabled

● Disabled

Modifying this setting may affect compatibility with clients, services, and applications.
For more information, see Microsoft network server: Digitally sign communications (always). (Q823659)

---

4 Select **Enabled** and **Apply.** The Confirm window appears.



You are about to change this setting to a value that may affect compatibility with clients, services, and applications.

For more information, see Microsoft network server: Digitally sign communications (always). (Q823659)

Do you want to continue with the change?

---

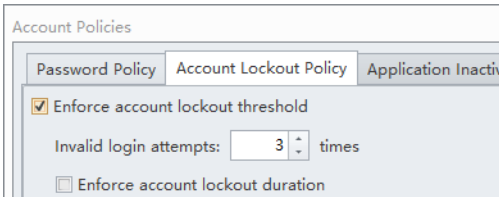
5 Select **Yes.** Then select **OK** to close the window.

Enabling Audit Object Access

This audit object access policy allows the Windows system to record an event for the CytoFLEX mosaic database if the security auditing setting for both the CytExpert for Spectral database and CytExpert for Spectral service database are enabled. When the CytExpert for Spectral database files and/or the CytExpert for Spectral service database are deleted, an event will be automatically created in the Windows security logs.

---

1 Type **Local Security Policy** from the Windows search bar to locate the **Local Security Policy** control panel.



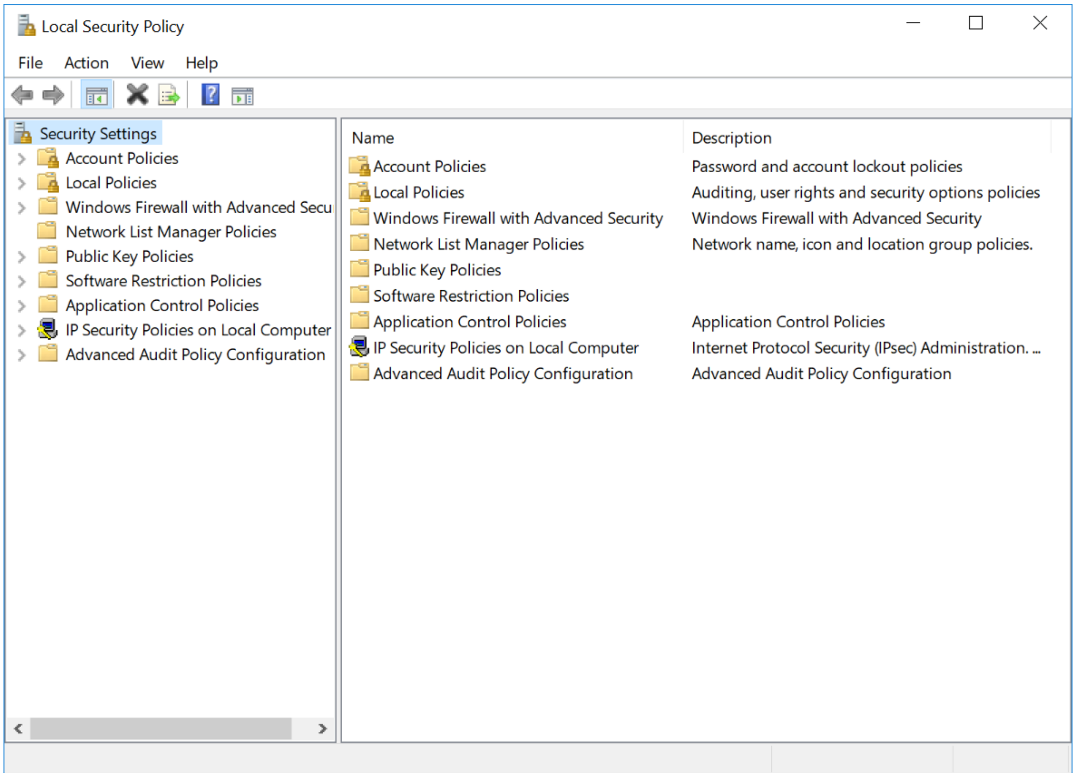
☑ Enforce account lockout threshold

Invalid login attempts: 3 times

☐ Enforce account lockout duration

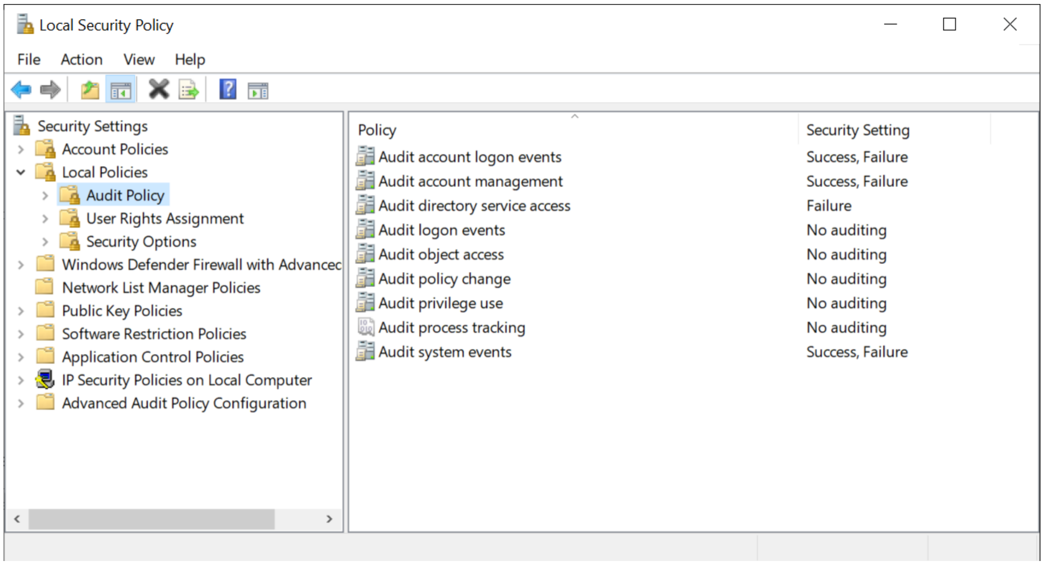
---

2 Double-click **Local Security Policy** to open the Local Security Policy window.


Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Local Policies
Windows Firewall with Advanced Secu
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Computer
Advanced Audit Policy Configuration

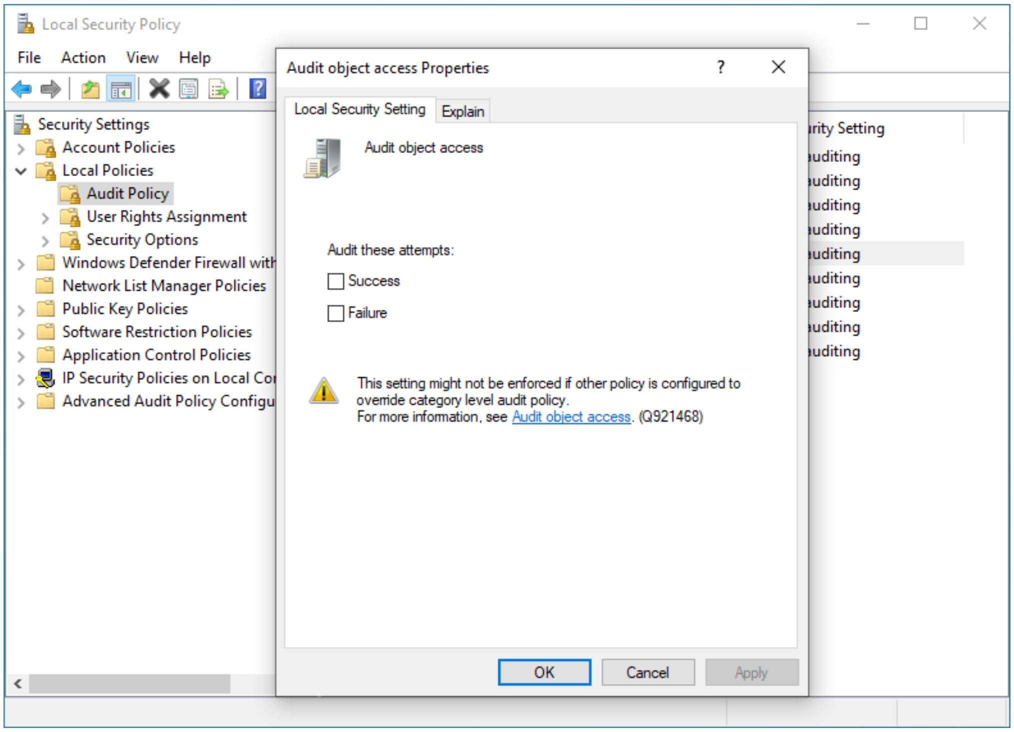
Name	Description
Account Policies	Password and account lockout policies
Local Policies	Auditing, user rights and security options policies
Windows Firewall with Advanced Security	Windows Firewall with Advanced Security
Network List Manager Policies	Network name, icon and location group policies.
Public Key Policies	
Software Restriction Policies	
Application Control Policies	Application Control Policies
IP Security Policies on Local Computer	Internet Protocol Security (IPsec) Administration. ...
Advanced Audit Policy Configuration	Advanced Audit Policy Configuration

3 Navigate to **Security Settings** > **Local Policies** > **Audit Policy**.


Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Local Policies
Audit Policy
User Rights Assignment
Security Options
Windows Defender Firewall with Advancec
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Computer
Advanced Audit Policy Configuration

Policy	Security Setting
Audit account logon events	Success, Failure
Audit account management	Success, Failure
Audit directory service access	Failure
Audit logon events	No auditing
Audit object access	No auditing
Audit policy change	No auditing
Audit privilege use	No auditing
Audit process tracking	No auditing
Audit system events	Success, Failure

4 Double-click **Audit object access** to open the Audit object access Properties window.



Audit object access

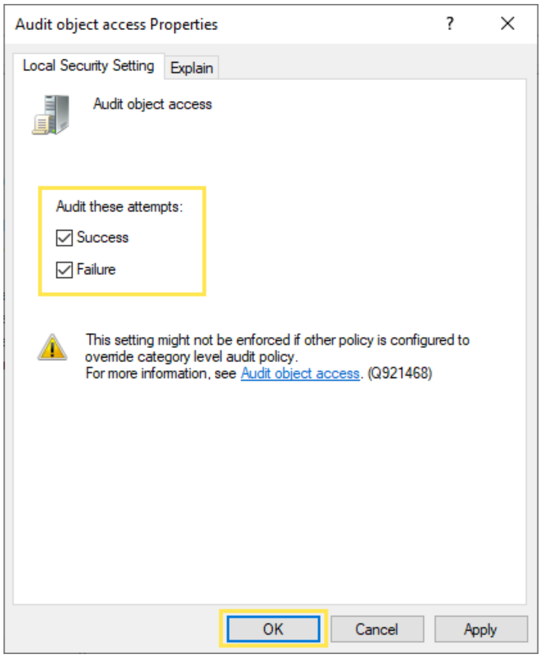
Audit these attempts:

☐ Success

☐ Failure

This setting might not be enforced if other policy is configured to override category level audit policy.
For more information, see Audit object access. (Q921468)

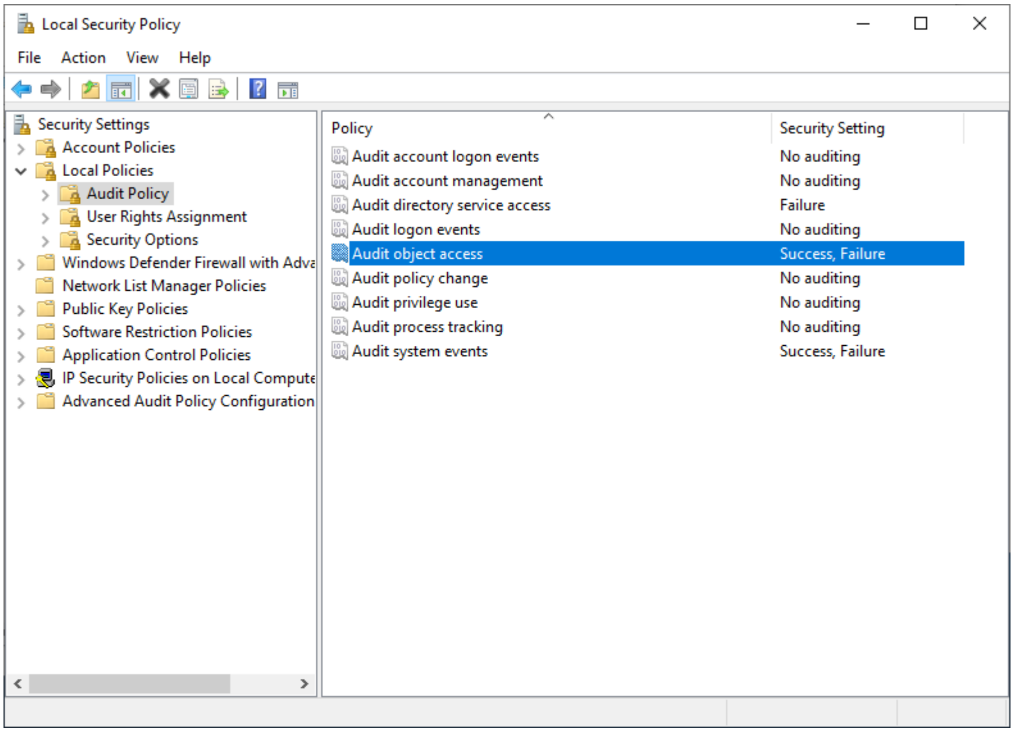
5 Select **Success** and **Failure**, and select **OK** to confirm.



Audit these attempts:
☑ Success
☑ Failure

This setting might not be enforced if other policy is configured to override category level audit policy.
For more information, see Audit object access. (Q921468)

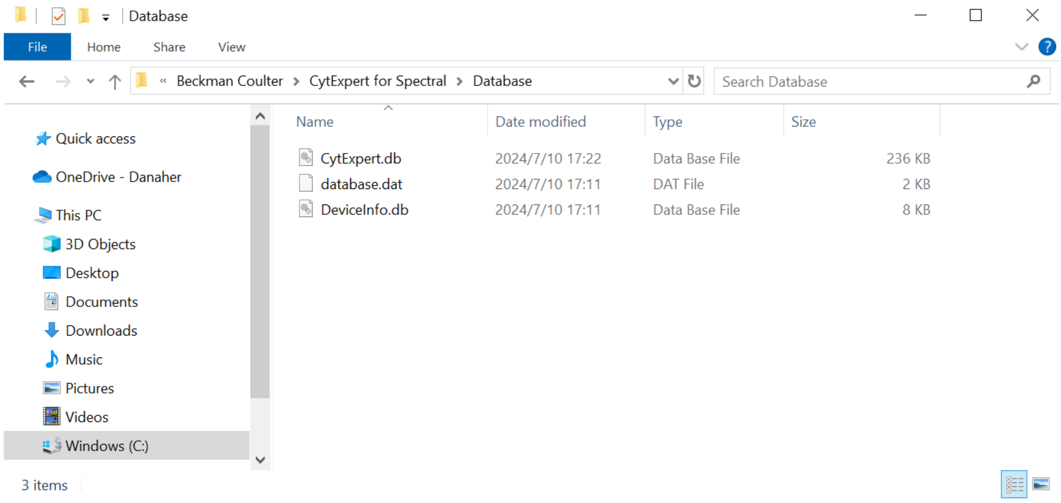
The security setting for audit object access is enabled.


Local Security Policy
File
Action
View
Help
Security Settings
Account Policies
Local Policies
Audit Policy
User Rights Assignment
Security Options
Windows Defender Firewall with Adva
Network List Manager Policies
Public Key Policies
Software Restriction Policies
Application Control Policies
IP Security Policies on Local Compute
Advanced Audit Policy Configuration

Policy	Security Setting
Audit account logon events	No auditing
Audit account management	No auditing
Audit directory service access	Failure
Audit logon events	No auditing
Audit object access	Success, Failure
Audit policy change	No auditing
Audit privilege use	No auditing
Audit process tracking	No auditing
Audit system events	Success, Failure

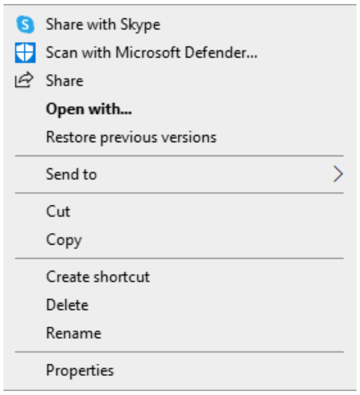
6 Close the Local Security Policy window.

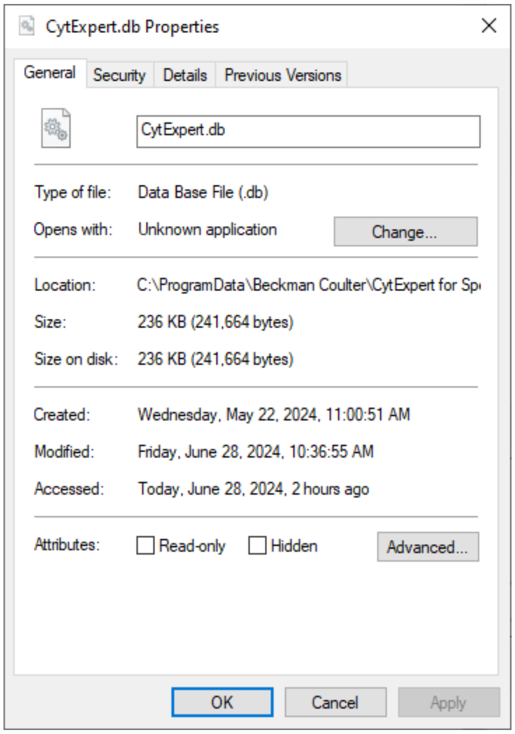
7 Navigate to **Windows (C:)** > **ProgramData** > **Beckman Coulter** > **CytExpert for Spectral** > **Database**.



Name	Date modified	Type	Size
CytExpert.db	2024/7/10 17:22	Data Base File	236 KB
database.dat	2024/7/10 17:11	DAT File	2 KB
DeviceInfo.db	2024/7/10 17:11	Data Base File	8 KB

8 Right-click the **CytExpert.db** database file, and select **Properties** from the drop-down menu to open the CytExpert.db Properties window.





CytExpert.db

Type of file: Data Base File (.db)

Opens with: Unknown application

Location: C:\ProgramData\Beckman Coulter\CytExpert for Sp

Size: 236 KB (241,664 bytes)

Size on disk: 236 KB (241,664 bytes)

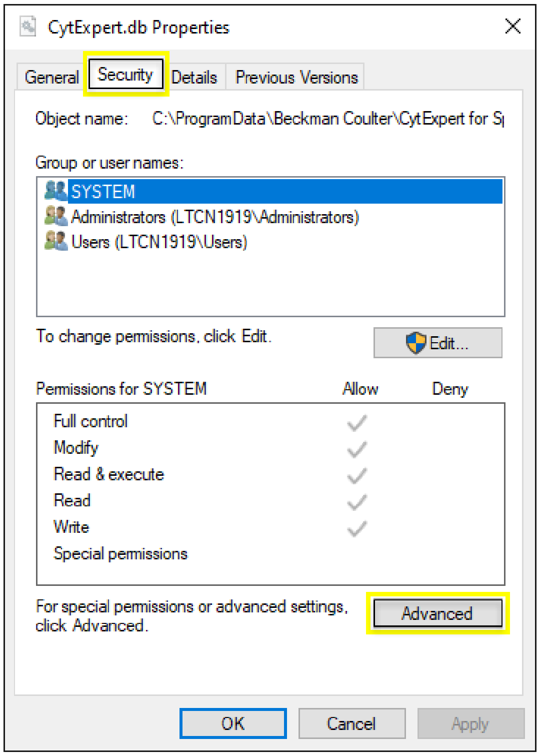
Created: Wednesday, May 22, 2024, 11:00:51 AM

Modified: Friday, June 28, 2024, 10:36:55 AM

Accessed: Today, June 28, 2024, 2 hours ago

Attributes: ☐ Read-only ☐ Hidden

9 Select the **Security** tab, and select **Advanced.**



CytExpert.db Properties

Object name: C:\ProgramData\Beckman Coulter\CytExpert for S

Group or user names:

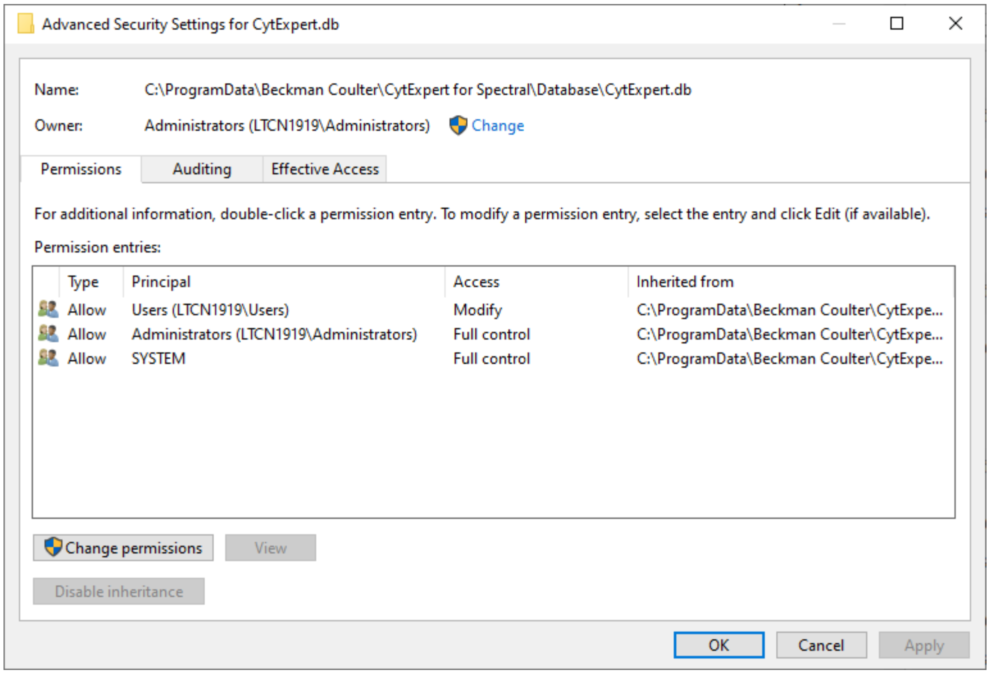
- SYSTEM
- Administrators (LTCN1919\Administrators)
- Users (LTCN1919\Users)

To change permissions, click Edit.

Permissions for SYSTEM	Allow	Deny
Full control	✓	
Modify	✓	
Read & execute	✓	
Read	✓	
Write	✓	
Special permissions		

For special permissions or advanced settings, click Advanced.

The following window appears.



Advanced Security Settings for CytExpert.db

Name: C:\ProgramData\Beckman Coulter\CytExpert for Spectral\Database\CytExpert.db

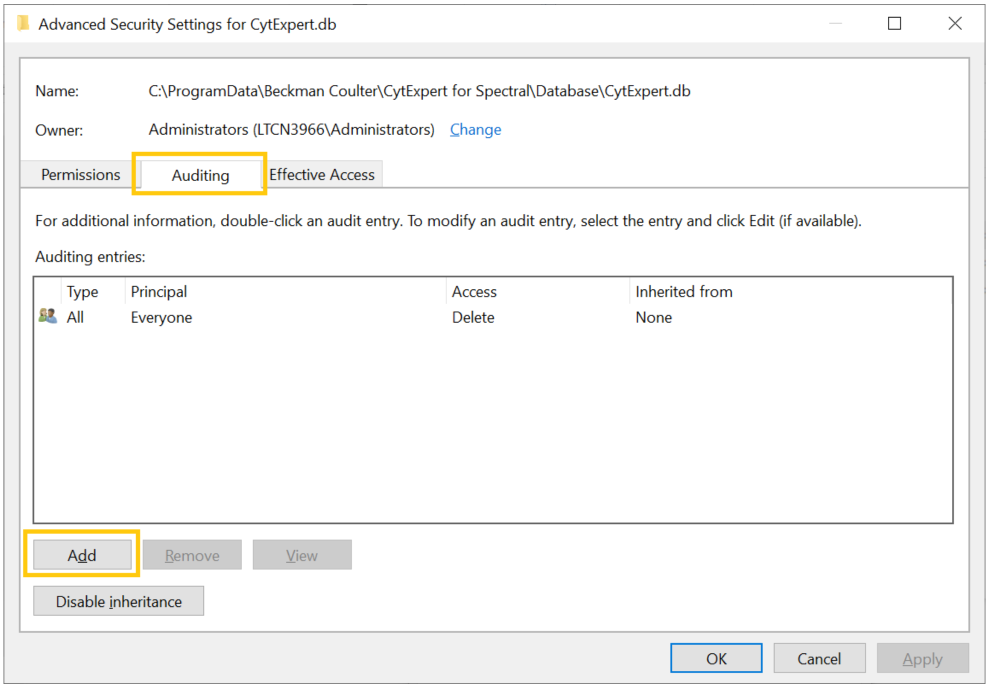
Owner: Administrators (LTCN1919\Administrators) Change

For additional information, double-click a permission entry. To modify a permission entry, select the entry and click Edit (if available).

Permission entries:

Type	Principal	Access	Inherited from
Allow	Users (LTCN1919\Users)	Modify	C:\ProgramData\Beckman Coulter\CytExpe...
Allow	Administrators (LTCN1919\Administrators)	Full control	C:\ProgramData\Beckman Coulter\CytExpe...
Allow	SYSTEM	Full control	C:\ProgramData\Beckman Coulter\CytExpe...

10 Select the **Auditing** tab and select **Add**.



Advanced Security Settings for CytExpert.db

Name: C:\ProgramData\Beckman Coulter\CytExpert for Spectral\Database\CytExpert.db

Owner: Administrators (LTCN3966\Administrators) Change

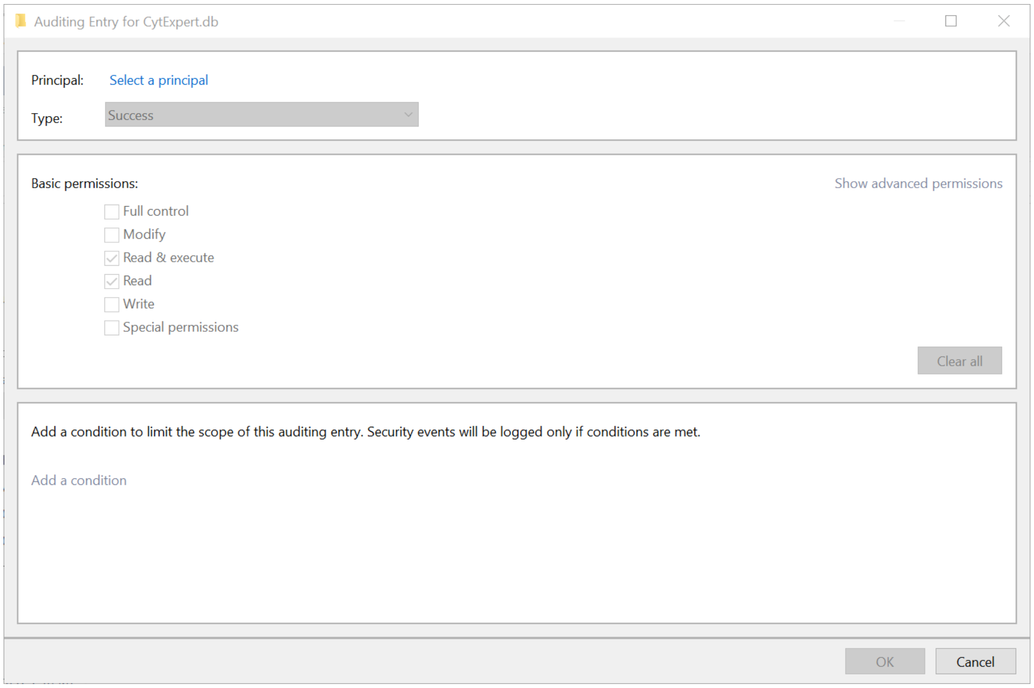
For additional information, double-click an audit entry. To modify an audit entry, select the entry and click Edit (if available).

Auditing entries:

Type	Principal	Access	Inherited from
All	Everyone	Delete	None

NOTE You must be an administrator or have been given the appropriate privileges to view the auditing properties of this object.

The following window appears.



Auditing Entry for CytExpert.db

Principal: Select a principal

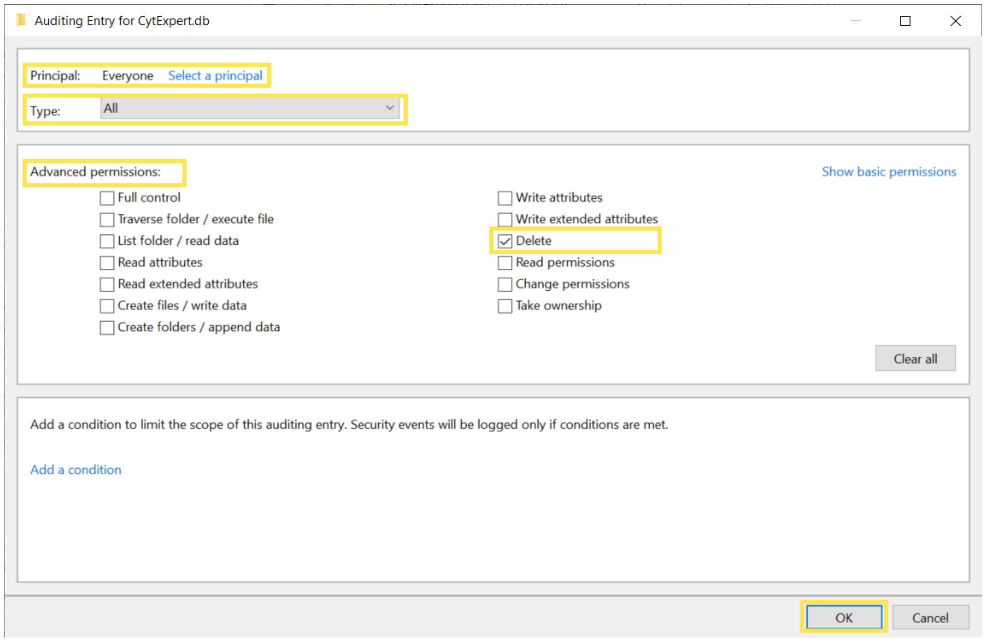
Type: Success

Basic permissions:

- ☐ Full control
- ☐ Modify
- ☑ Read & execute
- ☑ Read
- ☐ Write
- ☐ Special permissions

Add a condition to limit the scope of this auditing entry. Security events will be logged only if conditions are met.

11 Configure the security settings as below.



Auditing Entry for CytExpert.db

Principal: Everyone Select a principal

Type: All

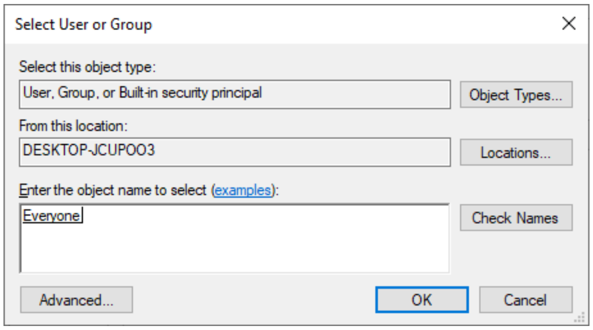
Advanced permissions: Show basic permissions

☐ Full control
☐ Traverse folder / execute file
☐ List folder / read data
☐ Read attributes
☐ Read extended attributes
☐ Create files / write data
☐ Create folders / append data
☐ Write attributes
☐ Write extended attributes
☑ Delete
☐ Read permissions
☐ Change permissions
☐ Take ownership

Add a condition to limit the scope of this auditing entry. Security events will be logged only if conditions are met.

Add a condition

a. Click **Select a principal**. The Select User or Group window appears.



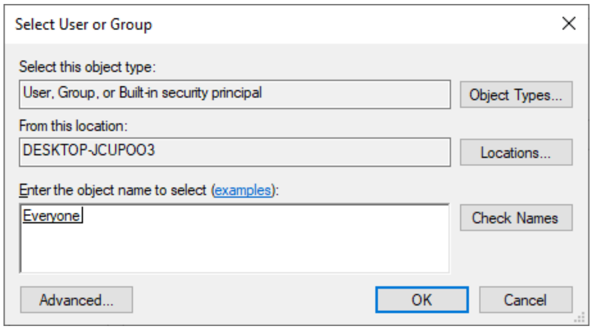
Select User or Group

Select this object type:
User, Group, or Built-in security principal

From this location:
DESKTOP-JCUPOO3

Enter the object name to select (examples):
Everyone

b. Enter **Everyone** in the object name field and select **OK**.



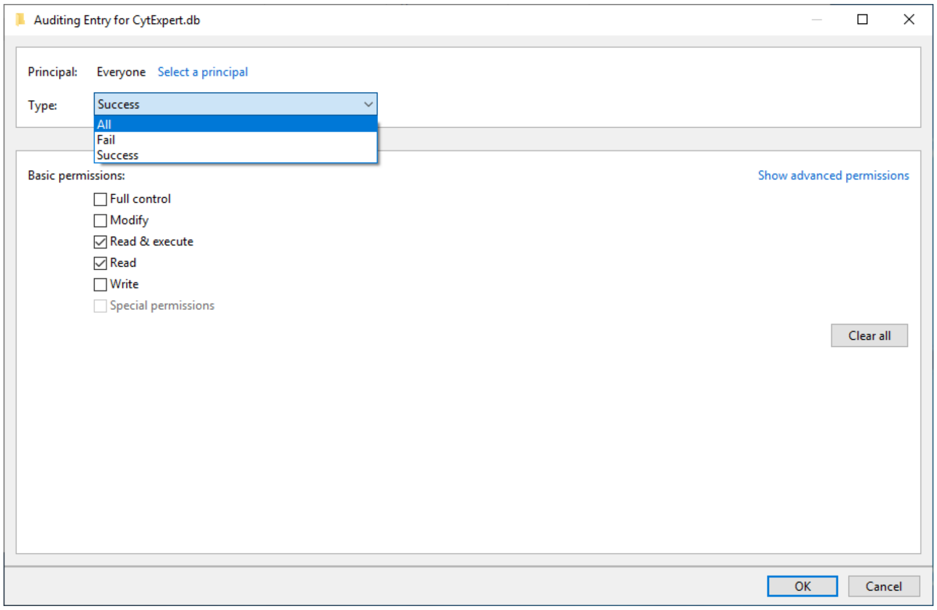
Select User or Group

Select this object type:
User, Group, or Built-in security principal

From this location:
DESKTOP-JCUPOO3

Enter the object name to select (examples):
Everyone

c. Select the Type **All**.



Auditing Entry for CytExpert.db

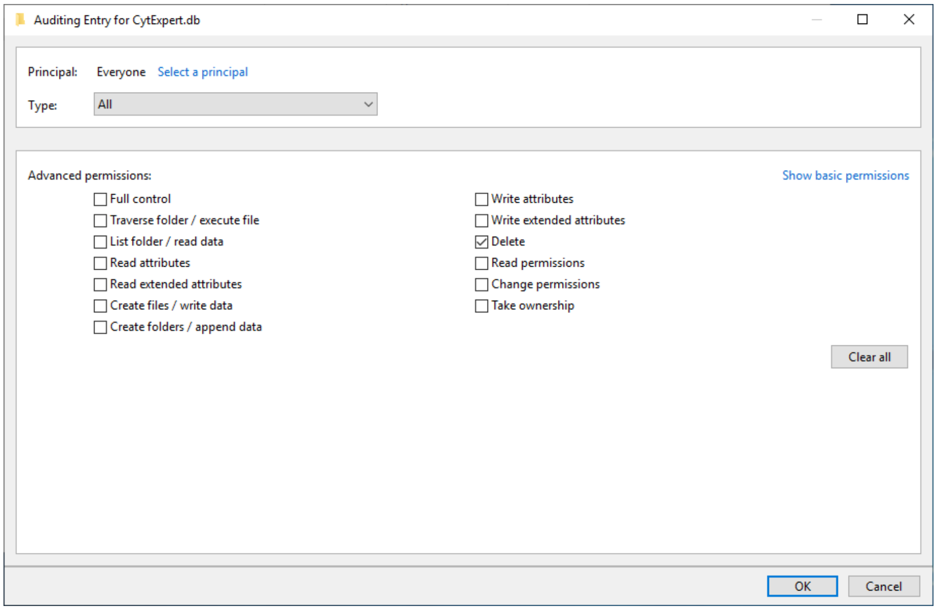
Principal: Everyone Select a principal

Type: Success
- All
- Fail
- Success

Basic permissions:

☐ Full control
☐ Modify
☑ Read & execute
☑ Read
☐ Write
☐ Special permissions

d. Select **Show advanced permissions.** The following window appears.



Auditing Entry for CytExpert.db

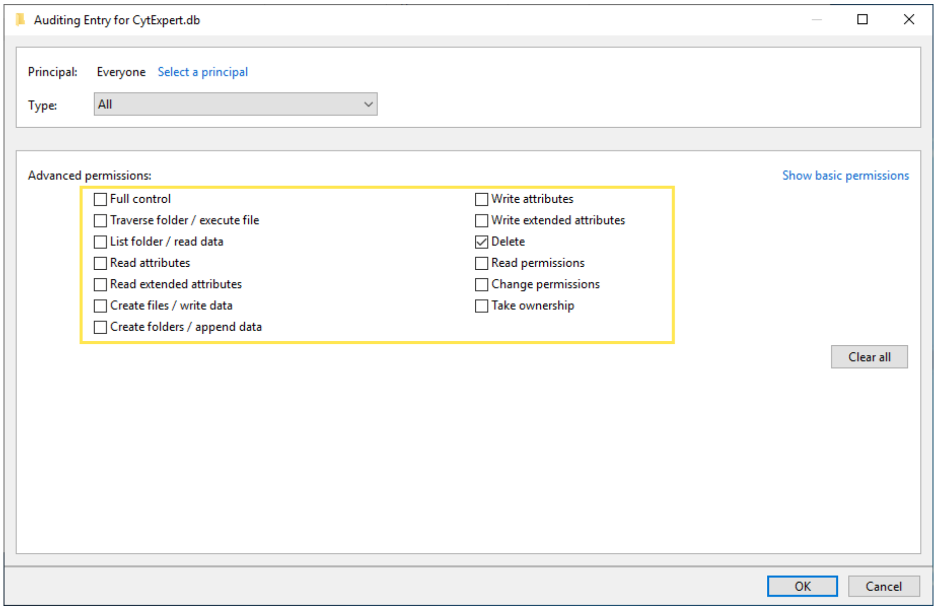
Principal: Everyone Select a principal

Type: All

Advanced permissions:

☐ Full control
☐ Traverse folder / execute file
☐ List folder / read data
☐ Read attributes
☐ Read extended attributes
☐ Create files / write data
☐ Create folders / append data
☐ Write attributes
☐ Write extended attributes
☑ Delete
☐ Read permissions
☐ Change permissions
☐ Take ownership

e. Set the advanced permissions and Select **OK**.



Auditing Entry for CytExpert.db

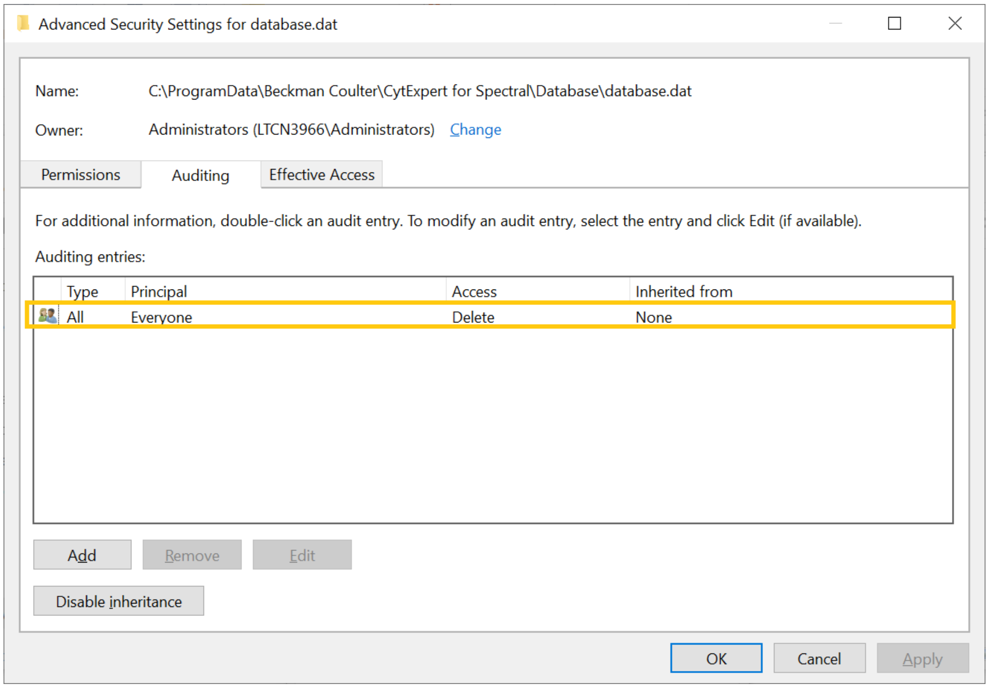
Principal: Everyone Select a principal

Type: All

Advanced permissions:

☐ Full control
☐ Traverse folder / execute file
☐ List folder / read data
☐ Read attributes
☐ Read extended attributes
☐ Create files / write data
☐ Create folders / append data
☐ Write attributes
☐ Write extended attributes
☑ Delete
☐ Read permissions
☐ Change permissions
☐ Take ownership

The security settings for CytoFLEX for Spectral database is enabled.



Advanced Security Settings for database.dat

Name: C:\ProgramData\Beckman Coulter\CytExpert for Spectral\Database\database.dat

Owner: Administrators (LTCN3966\Administrators) Change

For additional information, double-click an audit entry. To modify an audit entry, select the entry and click Edit (if available).

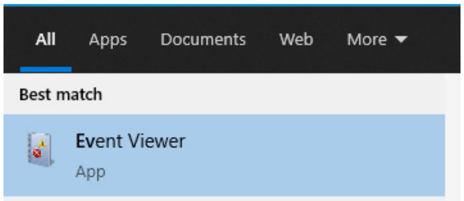
Auditing entries:

Type	Principal	Access	Inherited from
All	Everyone	Delete	None

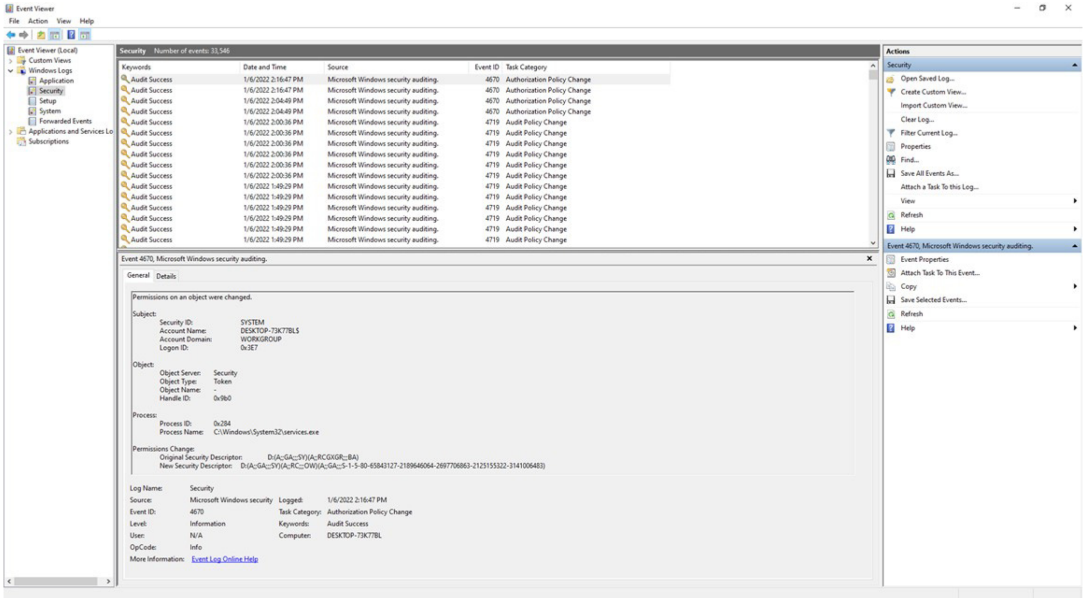
12 Select **OK**.

Viewing Windows Security Logs

1 Type **Event Viewer** from the Windows search bar to open the Event Viewer window.



2 Navigate to **Windows Logs** > **Security**.



Security Number of events: 33,546

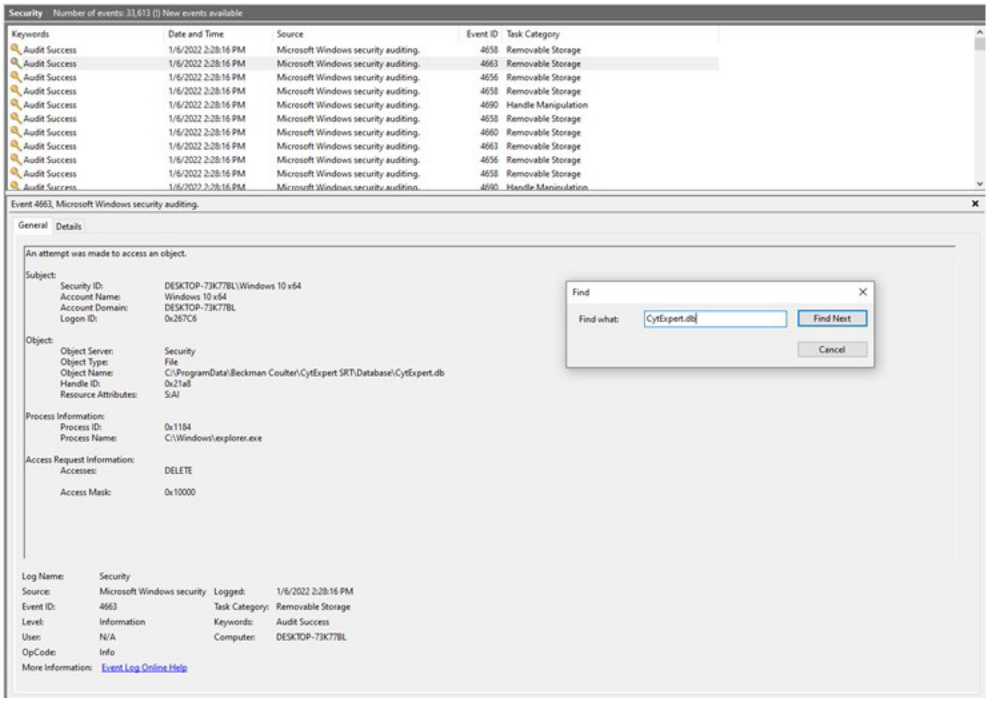
Keywords	Date and Time	Source	Event ID	Task Category
Audit Success	1/6/2022 2:16:47 PM	Microsoft Windows security auditing.	4670	Authorization Policy Change
Audit Success	1/6/2022 2:16:47 PM	Microsoft Windows security auditing.	4670	Authorization Policy Change
Audit Success	1/6/2022 2:04:49 PM	Microsoft Windows security auditing.	4670	Authorization Policy Change
Audit Success	1/6/2022 2:04:49 PM	Microsoft Windows security auditing.	4670	Authorization Policy Change
Audit Success	1/6/2022 2:00:36 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 2:00:36 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 2:00:36 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 2:00:36 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 2:00:36 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 2:00:36 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 1:49:29 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 1:49:29 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 1:49:29 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 1:49:29 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 1:49:29 PM	Microsoft Windows security auditing.	4719	Audit Policy Change
Audit Success	1/6/2022 1:49:29 PM	Microsoft Windows security auditing.	4719	Audit Policy Change

Event 4670, Microsoft Windows security auditing.

```
Permissions on an object were changed.

Subject:
        Security ID:            SYSTEM
        Account Name:           DESKTOP-73K77BL$
        Account Domain:         WORKGROUP
        Logon ID:               0x3E7

Object:
        Object Server:  Security
        Object Type:    Token
        Object Name:    -
        Handle ID:      0x9b0

Process:
        Process ID:     0x284
        Process Name:   C:\Windows\System32\services.exe

Permissions Change:
        Original Security Descriptor:       D:(A;;GA;;;SY)(A;;RCGXGR;;;BA)
        New Security Descriptor:    D:(A;;GA;;;SY)(A;;RC;;;OW)(A;;GA;;;S-1-5-80-65843127-2189646064-2697706863-2125155322-3141006483)
```

```
Log Name:         Security
Source:           Microsoft Windows security   Logged:          1/6/2022 2:16:47 PM
Event ID:         4670                         Task Category:   Authorization Policy Change
Level:            Information                  Keywords:        Audit Success
User:             N/A                          Computer:        DESKTOP-73K77BL
OpCode:           Info
More Information: Event Log Online Help
```

3 Select **Find** from the right Actions area and type **CytExpert.db** to quickly find the security logs for CytExpert for Spectral database.



```
Security   Number of events: 33,613 (!) New events available

Keywords        Date and Time           Source                                Event ID  Task Category
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4658      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4663      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4656      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4658      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4690      Handle Manipulation
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4658      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4660      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4663      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4656      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4658      Removable Storage
Audit Success   1/6/2022 2:28:16 PM     Microsoft Windows security auditing.  4690      Handle Manipulation
```

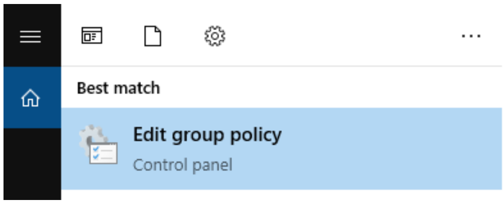
```
Event 4663, Microsoft Windows security auditing.

An attempt was made to access an object.

Subject:
        Security ID:            DESKTOP-73K77BL\Windows 10 x64
        Account Name:           Windows 10 x64
        Account Domain:         DESKTOP-73K77BL
        Logon ID:               0x267C6

Object:
        Object Server:          Security
        Object Type:            File
        Object Name:            C:\ProgramData\Beckman Coulter\CytExpert SRT\Database\CytExpert.db
        Handle ID:              0x21a8
        Resource Attributes:    S:AI

Process Information:
        Process ID:             0x1184
        Process Name:           C:\Windows\explorer.exe

Access Request Information:
        Accesses:               DELETE

        Access Mask:            0x10000

Log Name:          Security
Source:            Microsoft Windows security   Logged:          1/6/2022 2:28:16 PM
Event ID:          4663                         Task Category:   Removable Storage
Level:             Information                  Keywords:        Audit Success
User:              N/A                          Computer:        DESKTOP-73K77BL
OpCode:            Info
More Information:  Event Log Online Help
```

Find what: CytExpert.db

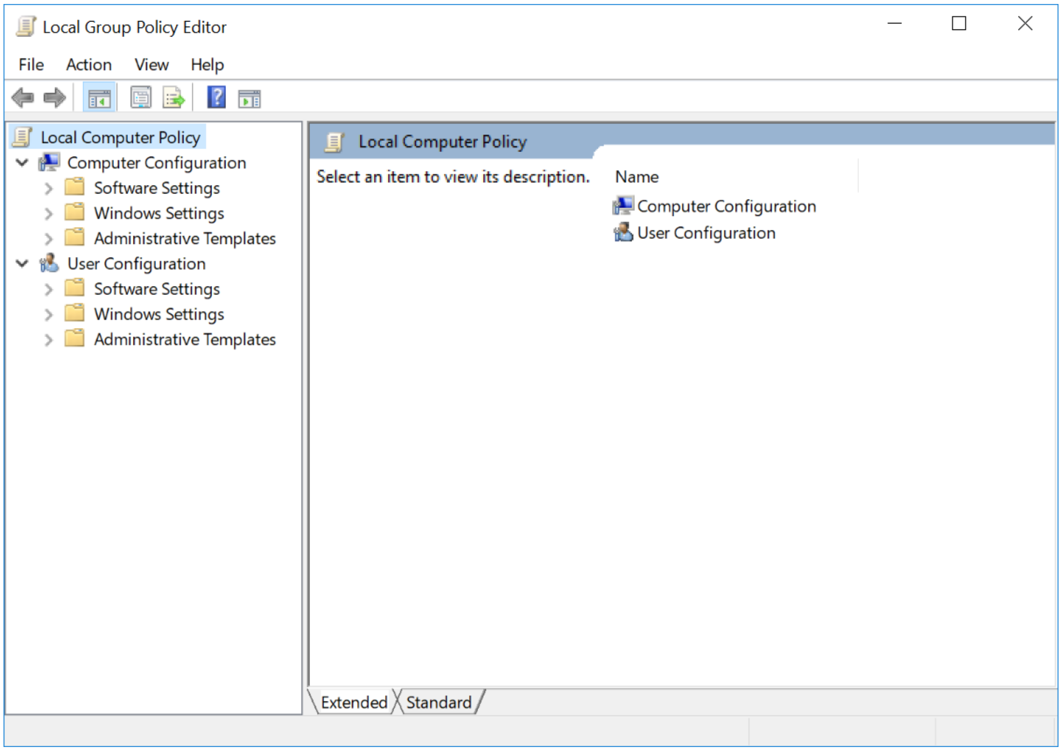
Type **CytoCtrlService.db** and select **Find Next** to quickly find the security logs for the CytoCtrlService database.

Enabling Installation Restriction

1 Type **Edit group policy** from the Windows search bar to locate the **Edit group policy editor** control panel.

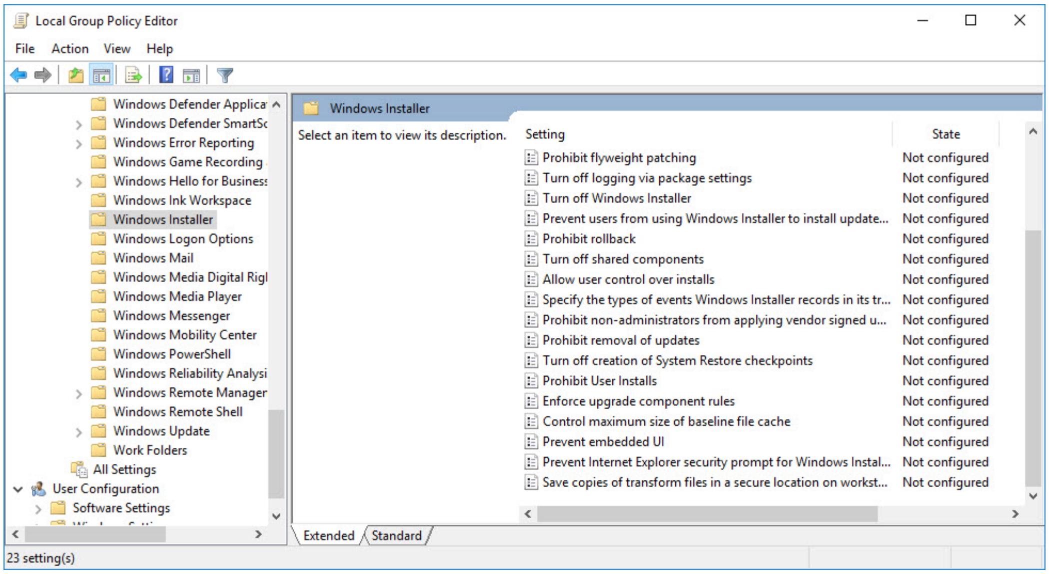


2 Select **Edit group policy editor.** The Local Group Policy Editor window displays.



Select an item to view its description.

3 Navigate to **Computer Configuration** > **Administrative Templates** > **Windows Components** > **Windows Installer.**

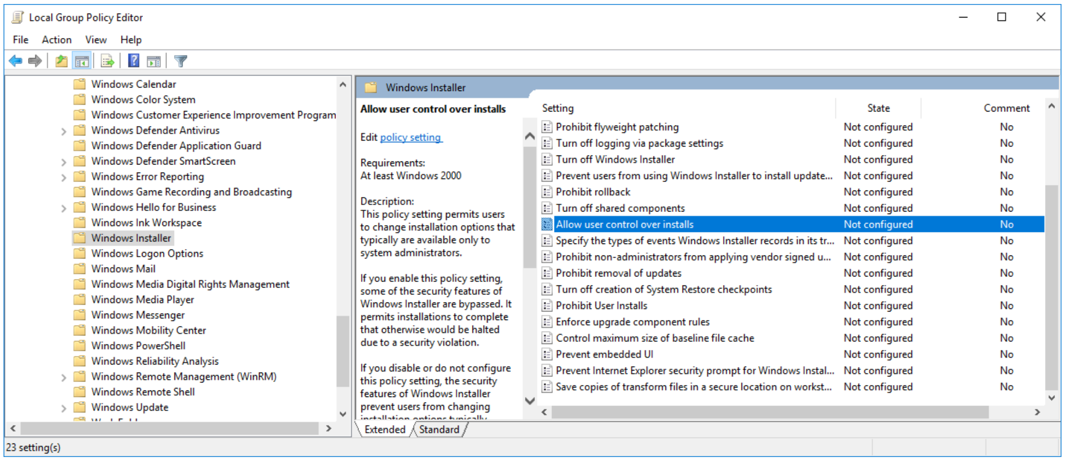


Select an item to view its description.

Setting	State
Prohibit flyweight patching	Not configured
Turn off logging via package settings	Not configured
Turn off Windows Installer	Not configured
Prevent users from using Windows Installer to install update...	Not configured
Prohibit rollback	Not configured
Turn off shared components	Not configured
Allow user control over installs	Not configured
Specify the types of events Windows Installer records in its tr...	Not configured
Prohibit non-administrators from applying vendor signed u...	Not configured
Prohibit removal of updates	Not configured
Turn off creation of System Restore checkpoints	Not configured
Prohibit User Installs	Not configured
Enforce upgrade component rules	Not configured
Control maximum size of baseline file cache	Not configured
Prevent embedded UI	Not configured
Prevent Internet Explorer security prompt for Windows Instal...	Not configured
Save copies of transform files in a secure location on workst...	Not configured

23 setting(s)

4 Select **Allow user control over installs.**


Local Group Policy Editor
File
Action
View
Help
Windows Calendar
Windows Color System
Windows Customer Experience Improvement Program
Windows Defender Antivirus
Windows Defender Application Guard
Windows Defender SmartScreen
Windows Error Reporting
Windows Game Recording and Broadcasting
Windows Hello for Business
Windows Ink Workspace
Windows Installer
Windows Logon Options
Windows Mail
Windows Media Digital Rights Management
Windows Media Player
Windows Messenger
Windows Mobility Center
Windows PowerShell
Windows Reliability Analysis
Windows Remote Management (WinRM)
Windows Remote Shell
Windows Update
Windows Installer
Extended
Standard

Allow user control over installs

Edit policy setting

Requirements:
At least Windows 2000

Description:
This policy setting permits users to change installation options that typically are available only to system administrators.

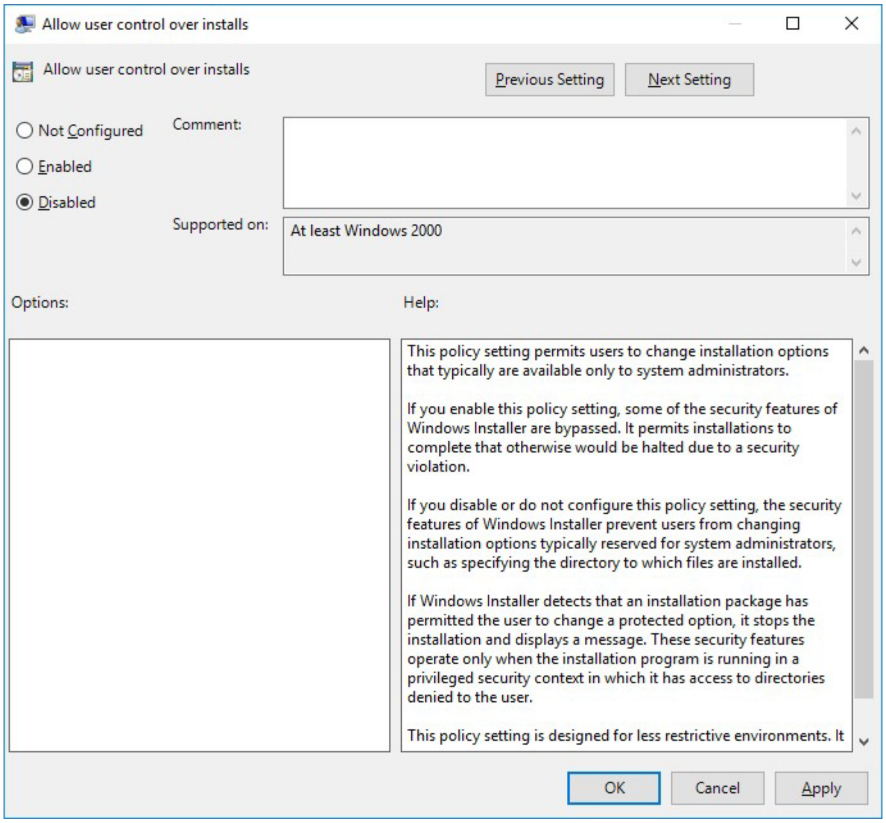
If you enable this policy setting, some of the security features of Windows Installer are bypassed. It permits installations to complete that otherwise would be halted due to a security violation.

If you disable or do not configure this policy setting, the security features of Windows Installer prevent users from changing

Setting	State	Comment
Prohibit flyweight patching	Not configured	No
Turn off logging via package settings	Not configured	No
Turn off Windows Installer	Not configured	No
Prevent users from using Windows Installer to install update...	Not configured	No
Prohibit rollback	Not configured	No
Turn off shared components	Not configured	No
Allow user control over installs	Not configured	No
Specify the types of events Windows Installer records in its tr...	Not configured	No
Prohibit non-administrators from applying vendor signed u...	Not configured	No
Prohibit removal of updates	Not configured	No
Turn off creation of System Restore checkpoints	Not configured	No
Prohibit User Installs	Not configured	No
Enforce upgrade component rules	Not configured	No
Control maximum size of baseline file cache	Not configured	No
Prevent embedded UI	Not configured	No
Prevent Internet Explorer security prompt for Windows Instal...	Not configured	No
Save copies of transform files in a secure location on workst...	Not configured	No

23 setting(s)

The following window appears.


Allow user control over installs
Allow user control over installs
Previous Setting
Next Setting
Comment:
Options:
Help:
OK
Cancel
Apply

○ Not Configured
○ Enabled
● Disabled

Supported on: At least Windows 2000

This policy setting permits users to change installation options that typically are available only to system administrators.

If you enable this policy setting, some of the security features of Windows Installer are bypassed. It permits installations to complete that otherwise would be halted due to a security violation.

If you disable or do not configure this policy setting, the security features of Windows Installer prevent users from changing installation options typically reserved for system administrators, such as specifying the directory to which files are installed.

If Windows Installer detects that an installation package has permitted the user to change a protected option, it stops the installation and displays a message. These security features operate only when the installation program is running in a privileged security context in which it has access to directories denied to the user.

This policy setting is designed for less restrictive environments. It

---

5 Select **Disabled** and **Apply**.

---

6 Select **OK** to close the window.

7 Disable **Always install with elevated privileges.** Refer to Steps 3-5.

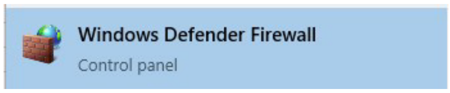
Enabling Microsoft Defender Firewall

IMPORTANT The Instrument cannot work properly if the necessary network ports are disabled. The communication between the CytExpert for Spectral software and the Instrument requires the following network ports to be enabled.

Outbound:

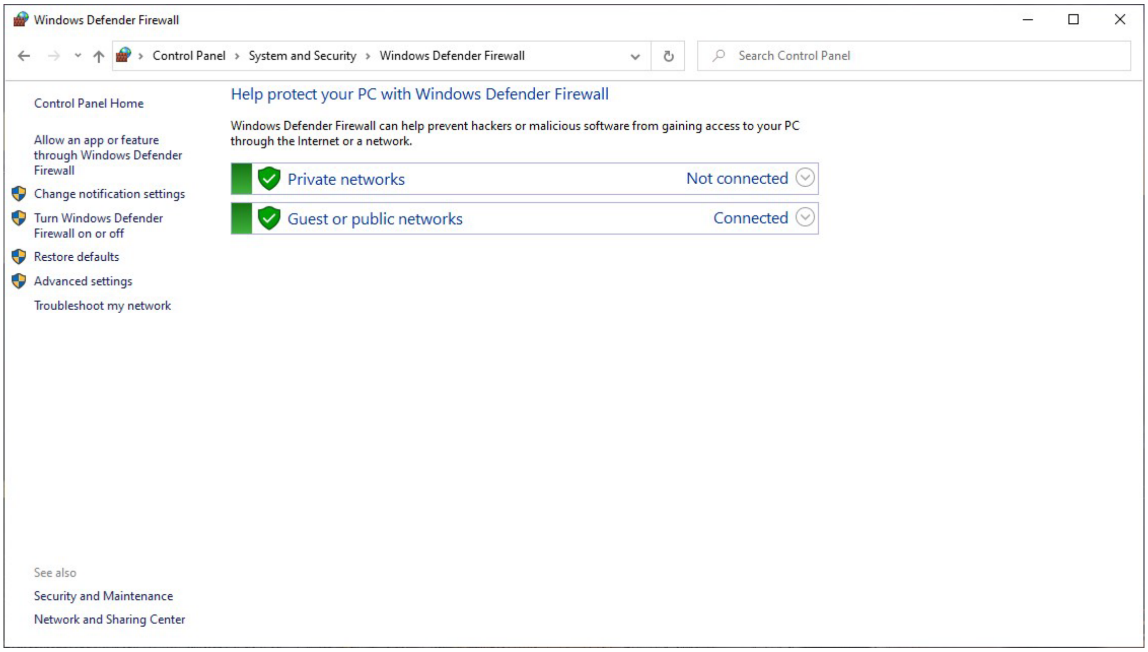
- TCP/192.168.76.18:1521
- TCP/192.168.76.18:1522
- TCP/192.168.76.18:1523
- TCP/192.168.76.18:1524
- TCP/192.168.76.18:1525
- TCP/192.168.76.18:60298

1 Type **Windows Defender Firewall** from the Windows search bar to locate the Windows Defender Firewall app.



---

2 Select **Turn Windows Defender Firewall on or off.**


Windows Defender Firewall
Control Panel › System and Security › Windows Defender Firewall
Search Control Panel
Control Panel Home
Allow an app or feature through Windows Defender Firewall
Change notification settings
Turn Windows Defender Firewall on or off
Restore defaults
Advanced settings
Troubleshoot my network
See also
Security and Maintenance
Network and Sharing Center

Help protect your PC with Windows Defender Firewall

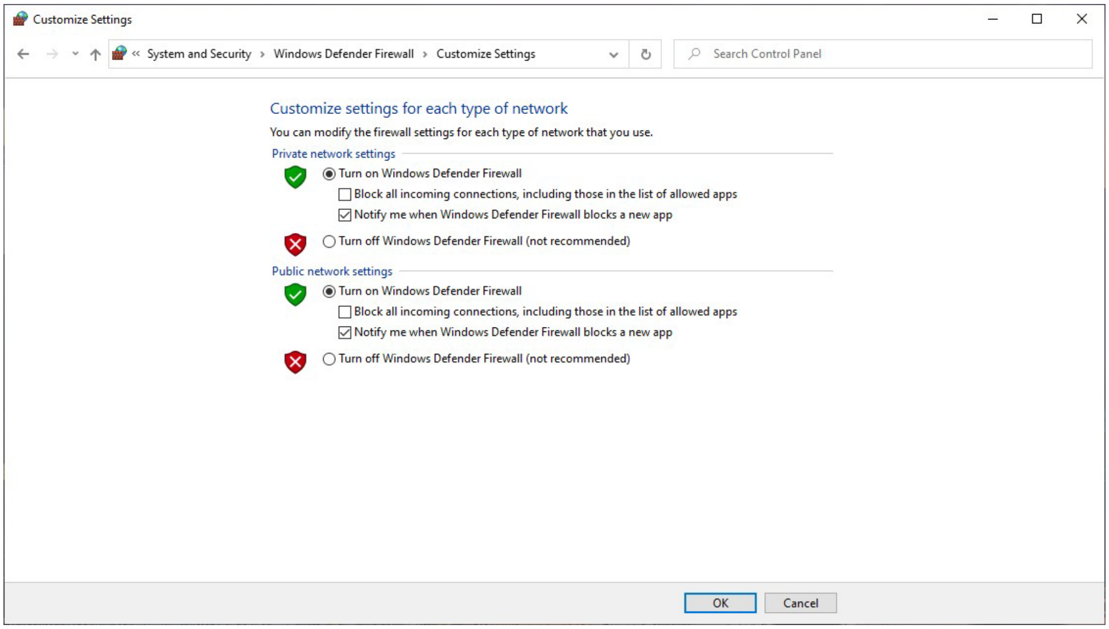
Windows Defender Firewall can help prevent hackers or malicious software from gaining access to your PC through the Internet or a network.

Private networks — Not connected

Guest or public networks — Connected

---

3 Select **Turn on Windows Defender Firewall** for both Private network and Public network settings.


Customize Settings
« System and Security › Windows Defender Firewall › Customize Settings
Search Control Panel
OK
Cancel

Customize settings for each type of network

You can modify the firewall settings for each type of network that you use.

Private network settings

- ● Turn on Windows Defender Firewall
  - ☐ Block all incoming connections, including those in the list of allowed apps
  - ☑ Notify me when Windows Defender Firewall blocks a new app
- ○ Turn off Windows Defender Firewall (not recommended)

Public network settings

- ● Turn on Windows Defender Firewall
  - ☐ Block all incoming connections, including those in the list of allowed apps
  - ☑ Notify me when Windows Defender Firewall blocks a new app
- ○ Turn off Windows Defender Firewall (not recommended)

---

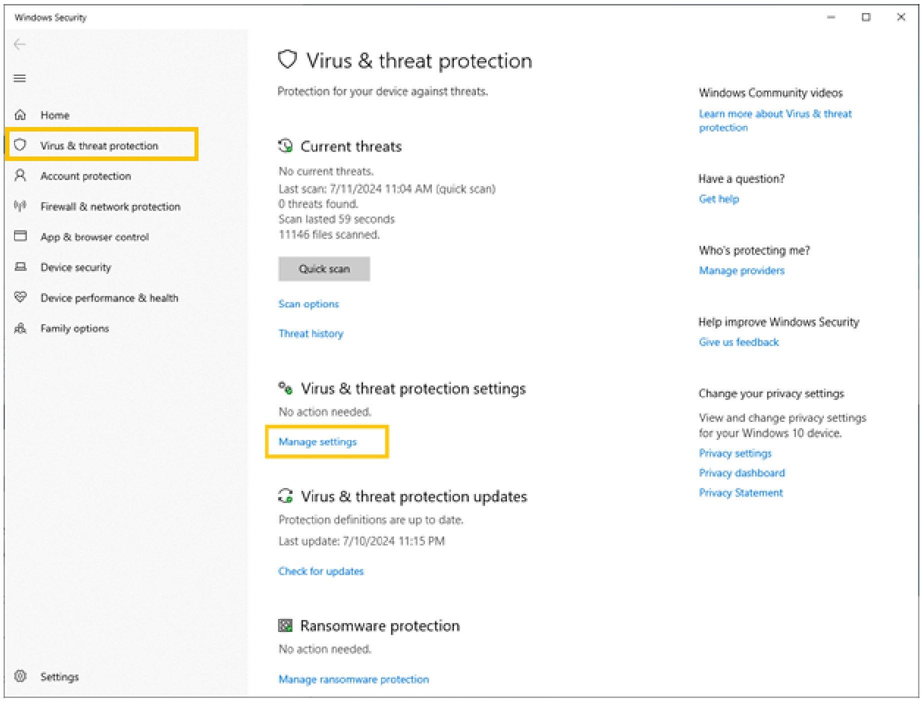
4 Select **OK.**

Enabling Microsoft Defender

1 Type **Windows Defender settings** from the Windows search bar.

2 Double-click **Windows Defender settings** to open the Windows Security window.

3 Select **Virus & threat protection**, and select **Manage settings**. The Virus & threat protection settings window appears.



Protection for your device against threats.

No current threats.
Last scan: 7/11/2024 11:04 AM (quick scan)
0 threats found.
Scan lasted 59 seconds
11146 files scanned.

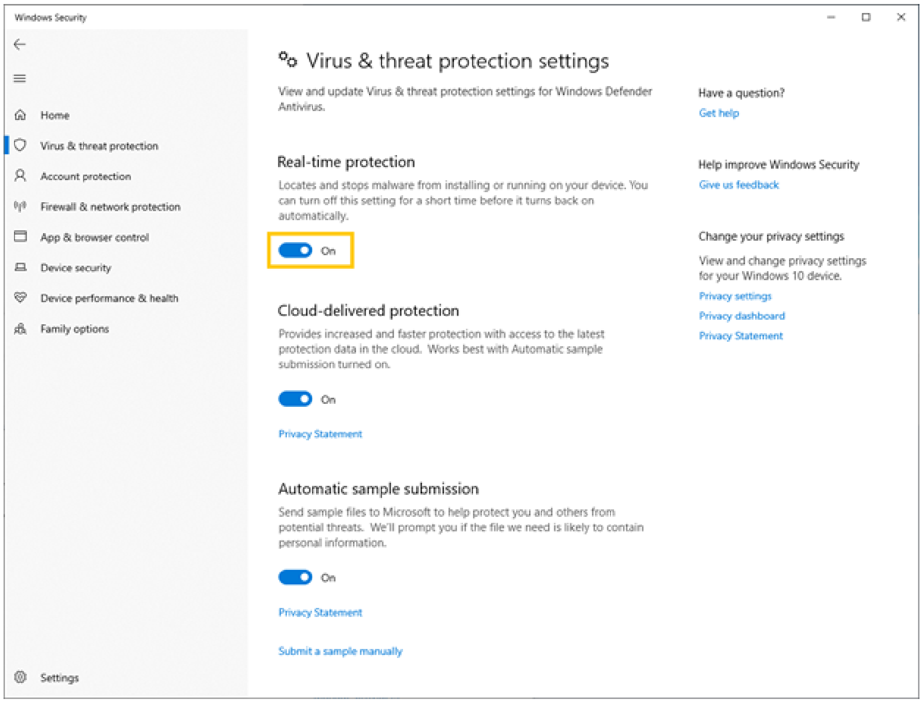
No action needed.

Protection definitions are up to date.
Last update: 7/10/2024 11:15 PM

No action needed.

View and change privacy settings for your Windows 10 device.

4 Ensure the **Real-tine protection** is on.



Virus & threat protection settings

View and update Virus & threat protection settings for Windows Defender Antivirus.

Real-time protection

Locates and stops malware from installing or running on your device. You can turn off this setting for a short time before it turns back on automatically.

On

Cloud-delivered protection

Provides increased and faster protection with access to the latest protection data in the cloud. Works best with Automatic sample submission turned on.

On

Privacy Statement

Automatic sample submission

Send sample files to Microsoft to help protect you and others from potential threats. We'll prompt you if the file we need is likely to contain personal information.

On

Privacy Statement

Submit a sample manually

Have a question?
Get help

Help improve Windows Security
Give us feedback

Change your privacy settings

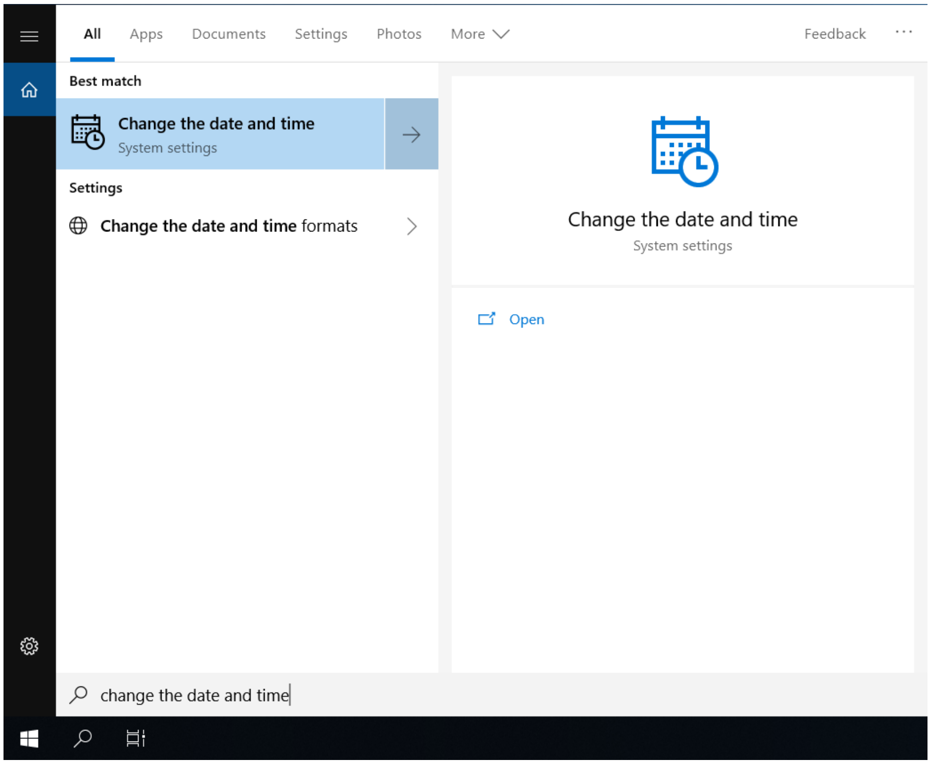
View and change privacy settings for your Windows 10 device.

Privacy settings
Privacy dashboard
Privacy Statement

Enabling Network Time Protocol

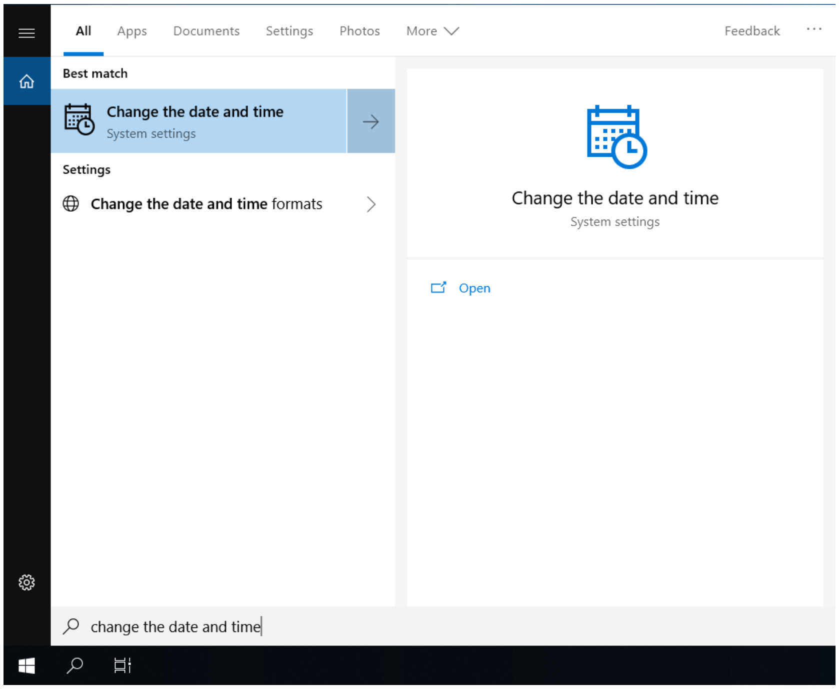
To ensure the accuracy of date and time information included in logs, follow the instructions below to enable the Network Time Protocol.

1 Type **Change the date and time** from the Windows search bar.



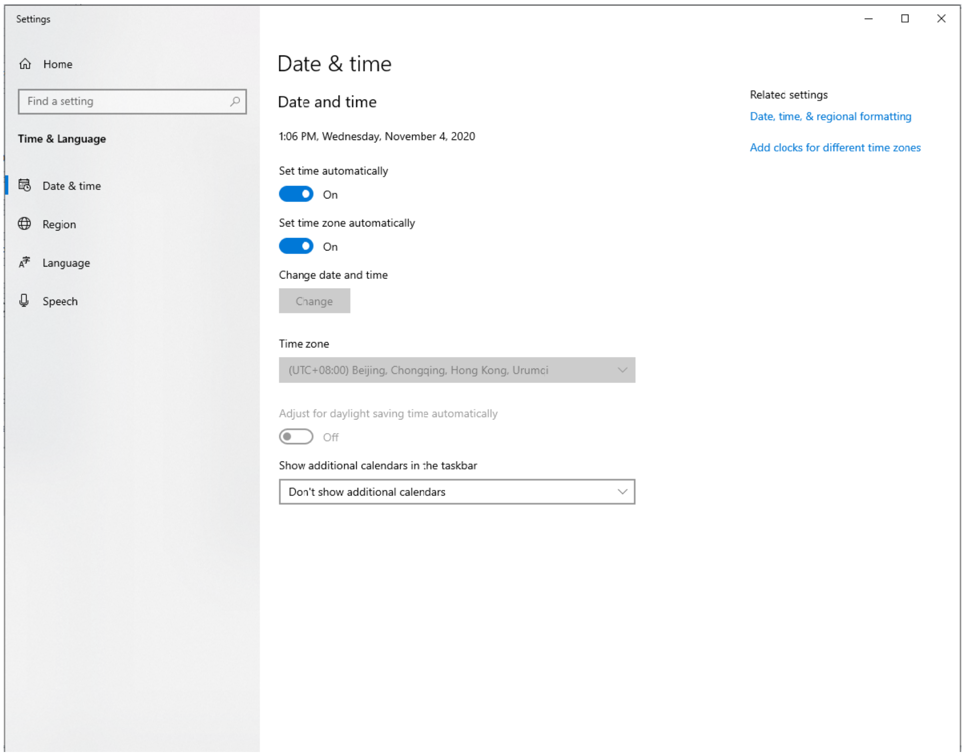
change the date and time

2 Double-click **Change the date and time.**



change the date and time

The Date and Time Settings window appears.



Date & time

Date and time

1:06 PM, Wednesday, November 4, 2020

Set time automatically: On

Set time zone automatically: On

Change date and time

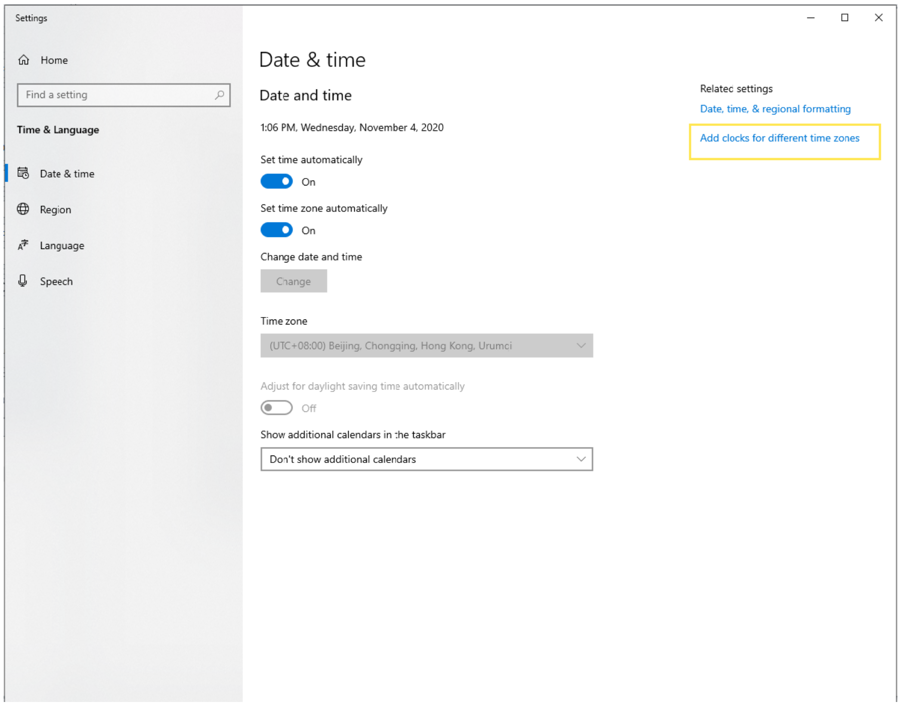
Time zone: (UTC+08:00) Beijing, Chongqing, Hong Kong, Urumqi

Adjust for daylight saving time automatically: Off

Show additional calendars in the taskbar: Don't show additional calendars

NOTE Ensure that the **Set time zone automatically** and **Set time zone automatically** are on.

3 Select **Add clocks for different time zones** to access the Date and Time window.



Date & time

Date and time

1:06 PM, Wednesday, November 4, 2020

Set time automatically: On

Set time zone automatically: On

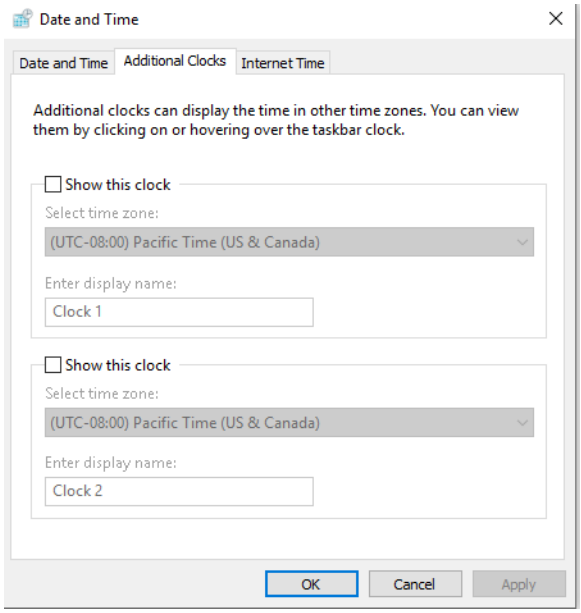
Change date and time

Time zone: (UTC+08:00) Beijing, Chongqing, Hong Kong, Urumqi

Adjust for daylight saving time automatically: Off

Show additional calendars in the taskbar: Don't show additional calendars

The Date and Time window displays.



Additional clocks can display the time in other time zones. You can view them by clicking on or hovering over the taskbar clock.

☐ Show this clock

Select time zone: (UTC-08:00) Pacific Time (US & Canada)

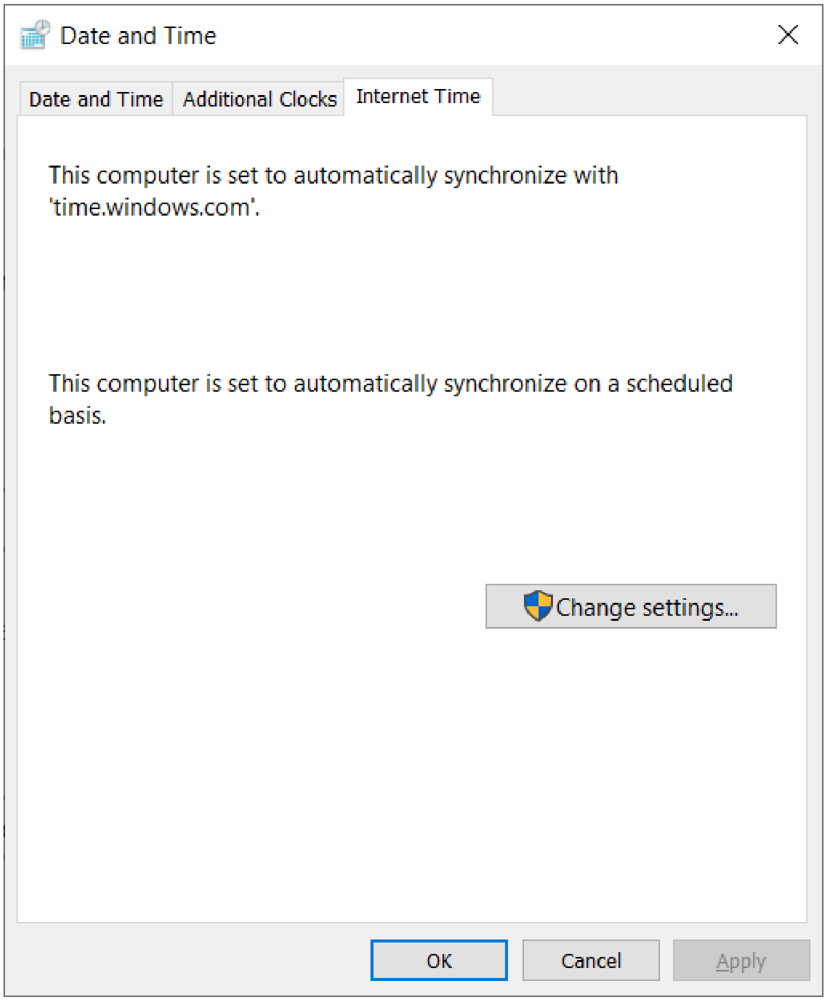
Enter display name: Clock 1

☐ Show this clock

Select time zone: (UTC-08:00) Pacific Time (US & Canada)

Enter display name: Clock 2

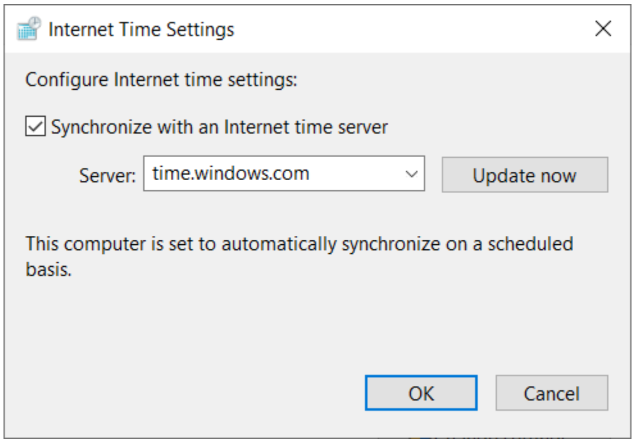
4 Select the Internet Time tab from the Date and Time window, and then select **Change settings** from the Internet Time tab.



This computer is set to automatically synchronize with 'time.windows.com'.

This computer is set to automatically synchronize on a scheduled basis.

The Internet Time Settings window displays.



Configure Internet time settings:

☑ Synchronize with an Internet time server

Server: time.windows.com

This computer is set to automatically synchronize on a scheduled basis.

5 Select **Synchronize with an internet time server**, and select **OK**. The PC is set to automatically synchronize with the internet time server.

Remote Access

IMPORTANT Use BeckmanConnect for remote control over the CytoFLEX instrument. For instructions on using BeckmanConnect, refer to the BeckmanConnect website.

APPENDIX F

Table of Hazardous Substances

Table of Hazardous Substances

The Hazardous Substances Names and Concentration is shown in Table F.1 and Table F.2.

NOTE Refer to the *CytoFLEX Platform Instructions for Use* manual, APPENDIX H, Table of Hazardous Substances for the tables of hazardous substances of the CytoFLEX S and CytoFLEX LX.

Table F.1 有毒有害物质名称及含量的标识格式 Table of Hazardous Substances Name and Concentration [CytoFLEX mosaic 63]

电子电气产品号码 **EEP Part Number: D10641**	产品名称 **Product Name: CytoFLEX mosaic Spectral Detection Module 63** 产品型号 **Product Model Number: CytoFLEX mosaic**									
部件名称 Component Name	有毒有害物质或元素 Hazardous Substances Name									
	铅 (Pb)	汞 (Hg)	镉 (Cd)	六价铬 (Cr^{6+})	多溴联苯 (PBB)	多溴二苯醚 (PBDE)	邻苯二甲二乙酯 (DEHP)	乙血压 (BBP)	舒张压 (DBP)	二异丁基苯酚 (DIBP)
金属件 Metal Part	O	O	O	O	O	O	O	O	O	O
塑料件 Plastic Part	O	O	O	O	O	O	O	O	O	O
玻璃制品 Glass Part	O	O	O	O	O	O	O	O	O	O
橡胶制品 Rubber Part	O	O	O	O	O	O	O	O	O	O
印刷电路板 Printed Circuit Assembly	O	O	O	O	O	O	O	O	O	O
金属（底盘/螺钉/组件/护罩）Metal (Chassis/Screw/Subassemblies/Shields)	O	O	O	O	O	O	O	O	O	O
线缆及连接器 Wire/Cable/Connectors	O	O	O	O	O	O	O	O	O	O
其他电子件 Other Electrical	O	O	O	O	O	O	O	O	O	O
标签 Labeling	O	O	O	O	O	O	O	O	O	O
包装材料 Package	O	O	O	O	O	O	O	O	O	O

This table is prepared in accordance with the provisions of SJ/T 11364

O：表示该有毒有害物质在该部件所有均质材料中的含量均在GB/T 26572标准规定的限量要求以下

x: 表示该有毒有害物质至少在该部件的某一均质材料中的含量超出GB/T 26572标准规定的限量要求

（企业可在此处，根据实际情况对上表中打"×"的技术原因进行进一步说明)

O: Indicates that the toxic or hazardous substances contained in all of the homogenous materials for this part is below the limit requirements in GB/T 26572.

X: Indicates that the toxic or hazardous substance contained in at least one of the homogenous materials used for this part in above the limit requirement in GB/T 26572.

(Enterprise may further provide in this box technical explanation for marking "X" based on their actual conditions)

Table F.2 有毒有害物质名称及含量的标识格式 Table of Hazardous Substances Name and Concentration [CytoFLEX mosaic 88]

电子电气产品号码 **EEP Part Number: D10638**	产品名称 **Product Name: CytoFLEX mosaic Spectral Detection Module 88** 产品型号 **Product Model Number: CytoFLEX mosaic**									
部件名称 Component Name	有毒有害物质或元素 Hazardous Substances Name									
	铅 (Pb)	汞 (Hg)	镉 (Cd)	六价铬 (Cr^{6+})	多溴联苯 (PBB)	多溴二苯醚 (PBDE)	邻苯二甲二乙酯 (DEHP)	乙血压 (BBP)	舒张压 (DBP)	二异丁基苯酚 (DIBP)
金属件 Metal Part	O	O	O	O	O	O	O	O	O	O
塑料件 Plastic Part	O	O	O	O	O	O	O	O	O	O
玻璃制品 Glass Part	O	O	O	O	O	O	O	O	O	O
橡胶制品 Rubber Part	O	O	O	O	O	O	O	O	O	O
印刷电路板 Printed Circuit Assembly	O	O	O	O	O	O	O	O	O	O
金属（底盘/螺钉/组件/护罩）Metal (Chassis/Screw/Subassemblies/Shields)	O	O	O	O	O	O	O	O	O	O
线缆及连接器 Wire/Cable/Connectors	O	O	O	O	O	O	O	O	O	O
其他电子件 Other Electrical	O	O	O	O	O	O	O	O	O	O
标签 Labeling	O	O	O	O	O	O	O	O	O	O
包装材料 Package	O	O	O	O	O	O	O	O	O	O

This table is prepared in accordance with the provisions of SJ/T 11364

○：表示该有毒有害物质在该部件所有均质材料中的含量均在GB/T 26572标准规定的限量要求以下

x: 表示该有毒有害物质至少在该部件的某一均质材料中的含量超出GB/T 26572标准规定的限量要求

（企业可在此处，根据实际情况对上表中打“×”的技术原因进行进一步说明)

O: Indicates that the toxic or hazardous substances contained in all of the homogenous materials for this part is below the limit requirements in GB/T 26572.

X: Indicates that the toxic or hazardous substance contained in at least one of the homogenous materials used for this part in above the limit requirement in GB/T 26572.

(Enterprise may further provide in this box technical explanation for marking “X” based on their actual conditions)

Abbreviations

The following list is a composite of the symbols, abbreviations, acronyms, and reference designators either used in this manual or related to the information in it. When the same abbreviation (or reference designator) is used for more than one word (or type of component), all meanings relevant to this manual are included, separated by semicolons.

' — foot

“ — inch

% — percent

°C — degrees Celsius

°F — degrees Fahrenheit

± — plus or minus

< — less than

> — greater than

≤ — less than or equal to

µ — micron

µL — microliters

µm — micrometer

A — ampere

AC — alternating current

APC — Allophycocyanin

APC-A700 — Allophycocyanin-Alexa Fluor 700 tandem dye

APC-A750 — Allophycocyanin-Alexa Fluor 750

APC-Cy7 — Allophycocyanin-Cyanin 7

API — Application Programming Interface

Acq. — Acquisition

BCI — Beckman Coulter Incorporated

BMP — bitmap

BP — band-pass filter

CDRH — Center for Devices and Radiological Health

CFSE — carboxyfluorescein succinmidyl ester

cm — centimeters

CSV — comma separated value

CV — coefficient of variation

DNA — deoxyribonucleic acid

DW — deep well

ECD — Energy Coupled Dye

EFUP — Environmentally friendly Use Period

EMC — Electromagnetic Compatibility

EMF — enhanced metafile format

EMR — electromagnetic radiation

FAPD — Fiber Array Photo Detector

FCS — flow cytometry standard

FITC — Fluorescein isothiocyanate

FLn — Fluorescence Number

FSC — forward scatter

GB — gigabyte

GHz — gigahertz

Gr Wt — gross weight

H — humidity

Hz — hertz

IEC — International Electrotechnical Commission

IR — infrared

kg — kilograms

KO — Krome Orange

LED — light emitting diode

L — liter

LJ — Levey-Jennings

LWH — length, width, height

m — meter

MB — megabyte

MFI — median fluorescence intensity

MHz — megahertz

min — minute

mL — milliliter

mm — millimeter

mW — milliwatt

NA — numerical aperture

NaClO — sodium hypochlorite solution

NaN_3 — sodium azide

nm — nanometer

Nt Wt — net weight

PB — Pacific Blue dye

PC5 — Phycoerythrin-Cy5 tandem dye

PC5.5 — Phycoerythrin-Cy5.5 tandem dye

PC7 — Phycoerythrin-Cy7 tandem dye

PE — Phycoerythrin

PEEK — polyether ether ketone

PerCP — Peridinin-Chlorophyll

PI — Propidium Iodide

PN — part number

QC — quality control

RAM — random access memory

rCV — robust coefficient of variation

RH — relative humidity

RoHS — Restriction of Hazardous Substances Directive

RPTM — real-time messaging protocol

S/N — serial number

SNR — signal to noise ratio

SSC — side scatter

USB — universal serial bus

UV — ultraviolet

V — volts

VA — volt-ampere

VAC — voltage alternating current

VSSC — violet side scatter

WDM — wavelength division multiplexer

W — watts

Index

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Related Documents

CytoFLEX mosaic Instructions for Use
PN D17052

CytoFLEX Platform Instructions for Use
PN B49006

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