E-737-100-15 (1)

SONY_®

ID7000™ Spectral Cell Analyzer

Operator's Guide

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Software Version 2.0

LE-ID7000 Series

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- For non-clinical research use only. Not for use in diagnostic or therapeutic procedures, or for any other clinical purpose.
- The ID7000 Spectral Cell Analyzer is classified as a Class 1 laser product.

The supplied installation disc includes the Operator's Guide for this unit in PDF format. For details, see "Using the PDF Manual" on page 11.

- Reproduction or duplication, in whole or part, of the software or Operator's Guide supplied with the unit, as well as renting or leasing of the software without the authorization of the right holder is prohibited under copyright law.
- Sony assumes no responsibility for damages, loss of income, or any claims from a third party arising out of use of the unit or supplied software.
- The software supplied with the unit cannot be used with any other units.
- Note that the specifications of the unit and supplied software are subject to change for improvement without prior notice.

Software Revision History

Functions Added in Version 2.0

The following functions have been added in ID7000 Software version 2.0.

Electronically signed records

A record of the operation history, called an audit trail, is created automatically and electronically signed for export as a PDF file.

For details, see "Running Sample Acquisition and Analysis".

Expansion of Standardization function

In Standardization mode in previous versions, the voltage value was limited to ST5.0 or lower for standardization between devices. In version 2.0, it is now possible to set the voltage value to the limit value of ST5.0 or higher obtained by Daily QC of the device. This enables more flexible experiments than could be conducted previously, ensuring reproducibility within the unit.

For details, see "Preparing the Instrument" (page 34) and "Configuring Instrument Settings" (page 59).

Sample groups can be configured across different types of plates

A single sample group can now be configured across different types of plates. Samples can be moved to any sample groups within an experiment, regardless of the type of plate.

For details, see "To move a sample to another sample group" (page 56).

Worksheet mode can be configured for each sample

In previous versions, the worksheet mode (Individual mode or Shared mode) could be configured only for sample groups. In this version, the mode can be configured for each sample.

For details, see "Changing the Worksheet Mode for Each Sample" (page 90).

[Setup as Single Positive Control] function can now be configured on the [Analysis] tab

The [Setup as Single Positive Control] function can now be configured on the [Analysis] tab in addition to the [Acquisition] tab.

Sheath line de-bubble function added

It is now possible to de-bubble the lines through which the sheath fluid flows when cleaning the sample probe. However, changes to the hardware are required.

For details, see "De-bubbling the Sheath Line" (page 158).

Internet connection of instrument control computer

ID7000 Software can be operated while the instrument control computer is connected to the Internet, presuming that appropriate information security measures are in place.

For details, see "ID7000 Software Operating Environment" (page 23).

Functions Added in Version 1.2

The following functions have been added in ID7000 Software version 1.2.

Contour plots added

Contour plots can now be created.

For details, see "Editing Plots" (page 69).

Increase in number of functions supporting individual user preferences

The number of functions that support individual user preferences has been increased. Also, preference settings can now be imported and exported so that the preference settings of one user can be used for other users.

For details, see "Configuring User Preference Settings" (page 86) and "[Preference Settings] dialog" (page 111).

Copy and paste function added for instrument settings

You can now copy and paste the instrument settings of a sample group to another sample group.

For details, see "Using Instrument Settings of Another Sample Group" (page 61).

Synchronous adjustment of PMT voltages for all channels in Normal mode

The PMT voltages can be increased or decreased all channels at once in Normal mode.

For details, see "Configuring Instrument Settings" (page 59).

Unmixing Viewer functions expanded

The spillover matrix can now be directly edited using the Unmixing Viewer.

For details, see "Checking Spectral Unmixing Results Summary" (page 82).

Improved operability of preference settings

The current state of the [Instrument Settings] dialog or plot properties dialog can now be saved as your preference settings.

For details, see "Using Instrument Settings of Another Sample Group" (page 61) and "Properties of dot plots, density plots, and histogram plots" (page 139).

Updating spectral reference fluorochrome information

Spectral reference fluorochrome information can now be updated.

For details, see "Updating Spectral Reference Fluorochrome Information" (page 102).

Saving an experiment as a template

An existing experiment can now be saved as a template.

For details, see "Saving an Experiment as a Template" (page 94).

Changing the event rate during acquisition

The event rate can be changed during acquisition in order to reduce the acquisition time.

For details, see step **5** in "Acquiring Data" (page 63).

Spectral overlay function added

A spectral overlay function has been added that allows multiple spectral data to be overlaid on a single plot.

For details, see "Overlaying Multiple Spectral Data" (page 84).

Undo/redo function added

Undo/Redo buttons have been added to the top left of the main window.

For details, see "Undo/Redo Function" (page 85).

Functions Added in Version 1.1.10

The following functions have been added in ID7000 Software version 1.1.10.

Sample keyword function added

Up to ten keywords can be attached to each sample.

For details, see "Attaching Keywords to Each Sample" (page 92).

Expansion to methods for adding fluorochromes

In addition to the existing direct entry method in the [Experiment Designer] window, support has been added for selecting from a list and for importing from the color panel.

For details, see "Adding Fluorochromes for Acquisition" (page 45).

Expansion to methods for stopping data acquisition

It is now possible to set the data acquisition stop conditions and saturation gate event monitoring for other unacquired samples within the same sample group.

For details, see "[Stopping Condition] dialog" (page 131).

Functions Added in Version 1.1

The following functions have been added in ID7000 Software version 1.1.

Consecutive running of Daily QC and Performance QC

Daily QC and Performance QC can now be run consecutively using the added [Start Daily and Performance QC] option.

For details, see "Running Daily QC and Performance QC Consecutively" (page 36).

Spectral unmixing results summary

You can now check the results of spectral unmixing by displaying a list of plots for each fluorochrome combination.

For details, see "Checking Spectral Unmixing Results Summary" (page 82).

Detector voltages adjustable separately

You can now adjust the voltage for each detector channel separately.

For details, see "Adjusting Detector Voltages Separately" (page 89).

Automatic export of acquisition FCS file

FCS file can now be exported automatically as soon as data is acquired.

For details, see "Exporting FCS File Automatically During Acquisition" (page 98).

ID7000 Data Agent application

The "ID7000 Data Agent" application has been added for exporting FCS file. ID7000 Data Agent starts when the FCS file export operation is executed, and its icon is displayed in the task tray.

For details, see "Using ID7000 Data Agent" (page 99).

Batch processing function

A "batch spectral unmixing" function, which performs spectral unmixing for multiple samples at the same time, and a "batch analysis" function, which performs analysis for multiple samples at the same time, have been added.

For details, see "Running Batch Analysis" (page 100).

Added QC criteria settings

User-defined criteria settings can now be configured for Daily QC and Performance QC.

For details, see "Setting QC Criteria" (page 104).

Using the PDF Manual

The Operator's Guide can be read on a computer with Adobe Reader installed.

You can download Adobe Reader free from the Adobe website.

- **1** Open the Operator's Guide folder in the installation disc.
- **2** Select and click on the Operator's Guide that you want to read.

If you have lost or damaged the installation disc, you can purchase a new one from your Sony distributor.

Components and Documentation

Component List

The ID7000 system comprises the following items.

- Main unit (1)
- Tank base (1)
- Safety Guide (1)
- Operator's Guide (this document) (1)
- Installation disc (Operator's Guide, ID7000 Software) (1)
- Power cord (2)
- PC connection cable (1)
- Sheath filter (1)
- Air filter for sheath tank (1)
- Air filter for waste tank (1)
- 10L sheath tank (1)
- 10L sheath tank cap kit (with pickup tube) (1)
- 10L waste tank (1)
- 10L waste tank cap kit (with float sensor) (1)
- Sheath tank connection tube (green) (2)
- Waste tank connection tube (red) (2)
- External tank connection cable (1)
- Auto sampler compatible 5 ml tube rack (1)

For U.S. and Canada customer

Notice about power cord to use

For this equipment (Model LE-ID7000 followed by A, B, C, D, E or F), only use the UL listed power cord. This power cord can be purchased from Sony Biotechnology Inc. as a separate accessory.

Accessory number of power cord: LE-U3E003 Specification of power cord:

15 A, 125 VAC Cable: 60 °C, SJT, VW-1, 300 V, 3×14 AWG, Max 2.4 m long. One end with NEMA 5-15P

Note

This equipment uses two power cords.

Documentation Structure

The system documentation comprises the following manuals.

• Safety Guide Contains safety information, usage precautions, and specifications.

Read carefully before operating the unit.

Operator's Guide (this document) Describes the required preparation and basic operating procedures in order to use the instrument, adjustments and settings of the instrument, and the configuration items of the ID7000 Software windows and dialogs. First, read "Chapter 2 Initial Setup" to perform the required preparation for using the instrument. Next, read "Chapter 3 Operation Flow" to grasp the operating principles, from data acquisition to analysis. For details about operations, see "Chapter 5 Acquisition" and "Chapter 6 Analysis". Subsequently, refer to "Chapter 7 Advanced Operations" for further details, as required.

Overview

Chapter

7

Main Features

The LE-ID7000 series (hereinafter referred to as the ID7000) is a spectral analysis type cell analyzer. Equipped with up to seven lasers and more than 180 channel photodetectors, it can acquire high-accuracy spectral data from various fluorochromes, and supports multi-color analysis of 44 or more colors. The software supports a spectral reference library that greatly improves the workflow for multicolor experiments, by eliminating complicated single-stain acquisition and fluorescence compensation. The unit also incorporates event rate detection and automatic shutdown functions.

Intuitive operation, spectral reference library

Intuitive acquisition and analysis operations through the combination of software and hardware, providing a through analysis of user workflows. Single-stain reference information (described below) is provided using a spectral reference library to achieve more intuitive workflows.

Free from single-stain acquisition

The unit can acquire and quickly analyze a multi-color experiment, omitting the normal single stain and fluorescence compensation steps, by recalling a registered spectral reference from the library using a wizard format.

High-speed acquisition

The hardware supports acquisition at speeds of up to 40,000 eps. The sample rate range of approximately 33.3 μ /min to 200 μ /min enables high-speed sample data acquisition.

Low carryover

A cleaning block mechanism that can move up and down cleans not only the inside of the sample nozzle but also the outside, resulting in low carryover.

Autosampler with active sample cooling

The ID7000 is equipped with an autosampler as standard. It supports 5 ml tube racks (24 tubes), 96-well plates, and 384-well plates. The samples can be cooled to 4 $^{\circ}$ C (39 $^{\circ}$ F) using a Peltier element.

Sample probe agitation function

Individual tubes/wells in tube rack/multi-well plate can be agitated using the sample probe to minimize potential damage to cells. In addition, samples can be agitated during acquisition, enabling stable, long-duration acquisition.

Auto shutdown

The autosampler is equipped with an extra loading station. Up to three 5 ml tubes can be placed in the extra loading station. You can configure the instrument in the software to shut down automatically after acquisition is completed by loading the solutions required for shutdown in the extra station.

Autofluorescence separation

Employs Sony's unique spectral optical technology for traditionally-difficult autofluorescence spectral separation for improved data stability and reliability. In ID7000 Software, you can find the autofluorescence spectrum using a wizard format.

Event rate detection

Detects air bubbles during sample acquisition, and automatically restores the equipment state. Use of the autosampler, 10L tank system, and auto shutdown functions also significantly increases the convenience of automatic acquisition.

Notifications

A sound can be emitted to notify the user at the normal end of a process and when an error/warning occurs during acquisition, QC, and cleaning.

Preference settings

You can configure software preference settings for each login user for added convenience.

Standardization mode

In Standardization mode, the output of each channel is standardized so that the SSC and fluorescence detection sensitivities are the same between multiple instruments. When the standardized settings (ST) are the same in software on multiple instruments, the sensitivity of the instruments is roughly aligned. Standardized data is obtained by running Daily QC, and is updated according to the instrument status. The unit supports standardization of Area and Height data.

Fluorescence compensation (unmixing) algorithms

The ID7000 supports the following spectral unmixing algorithm. This provides high-reproducibility fluorescence separation, quickly and simply.

• WLSM (Weighted Least Squares Method) This is a spectral unmixing algorithm based on the least squares method applicable for the detection principle of a spectral analysis-type cell analyzer. The variances in the signal that is detected generally increases as the fluorescence intensity increases. In weighted least squares method, this effect is appropriately handled by suppressing the variances in the calculated results by solving the least squares method with a weighting condition that permits errors due to high fluorescence intensity.

Virtual filter

The virtual filter function provides simple adjustment of channels, and provides the user flexibility for adding fluorochromes easily as needed. The information for each fluorochrome can be monitored visually in a color palette. The information does not effect unmixing result.

Fluorochromes

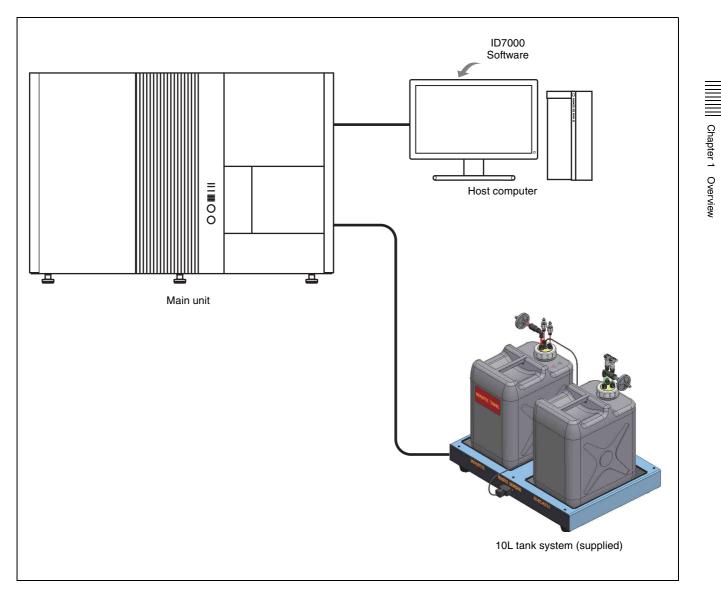
Often-used fluorochromes are configured as presets. You can also register user-specific fluorochromes.

Spectral overlay

Multiple spectral data can be overlaid on a single plot. Also, the displayed spectral data can be exported as a CSV file for further analysis using Microsoft Excel or other software.

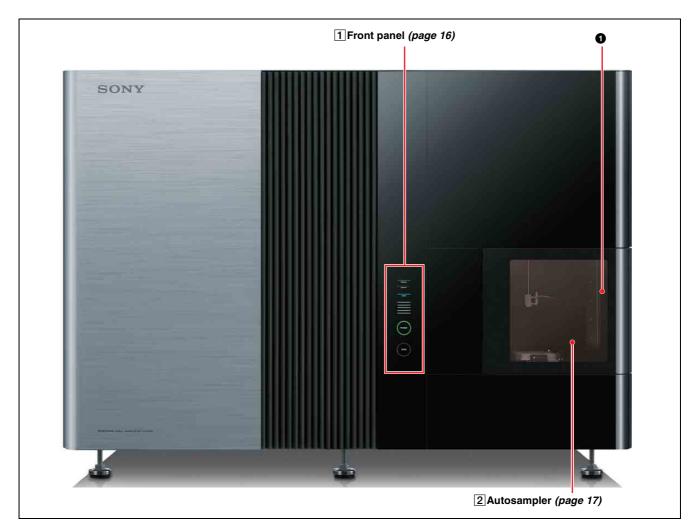
System Configuration

The ID7000 system comprises the ID7000 main unit, 10L tank system, and host computer.



Name and Function of Parts

Front Panel

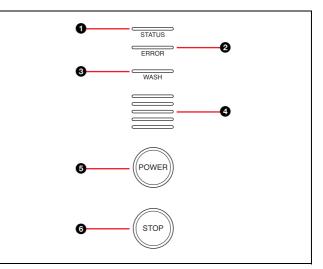


1 Front door

Provides access for loading and removing multi-well plates and sample tubes, and for cleaning.

A safety interlock function prevents acquisition while the door is open.

1 Front panel



1 STATUS indicator

Indicates the status of the main unit. Lights up green when the ID7000 starts up. The front door can be opened and closed when it is lit orange.

2 ERROR indicator

Indicates when an error has occurred in the main unit.

3 WASH indicator

Indicates the fluidics system is being cleaned.

4 DAQ (data acquisition) indicator

During acquisition, it displays a 5-level scale, depending on the event rate.

5 POWER button/indicator

Turns on the power supply to the main unit. When the power supply is on, the indicator light is green. When the power supply is on, press and hold this button for three seconds or longer to turn off the power supply.

6 STOP button/indicator

Stops acquisition if acquisition is in progress. The indicator lights up red.

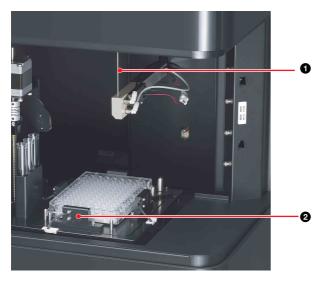
If the acquisition state prevents stopping, the indicator does not turn on, even if the STOP button is pressed.

Тір

Under normal circumstances, the main unit should be shut down from ID7000 Software.

For details, see "Shutting Down the Main Unit" (page 42).

2 Autosampler (with front door open)



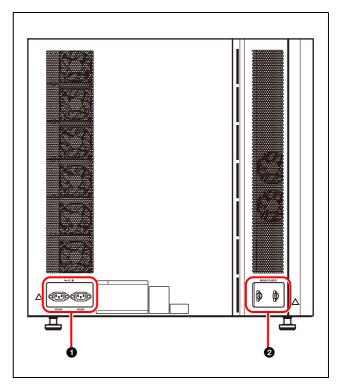
1 Sample probe line

Sample fluid intake from the multi-well plate or sample tube. The sample probe line is a maintenance item.

2 Multi-well plate holder

Loads a multi-well plate or 5 ml tube rack.

Left Side Panel



$\ensuremath{\textcircled{0}}\xspace \sim$ AC IN connector (AC power supply inlet, two locations)

Connects to a standard AC power supply (100-240 VAC, 50/60 Hz). Use only the supplied power cords to connect the ID7000 main unit to a power supply.

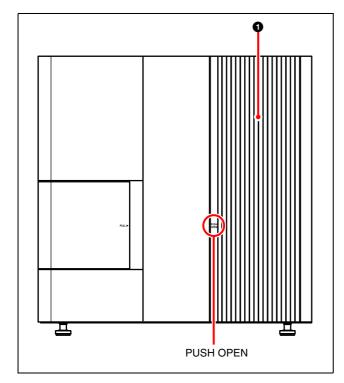
Notes

- Use only within the specified supply voltage range.
- The two power cords must be connected to power outlets which are grounded.

2 Main power switches (two locations)

Turns the power supply for the ID7000 on/off. Push both main power switches up to turn them on, and push the switches down to turn them off. If an electrical fault is detected, the circuit breaker function automatically disconnects the power supply.

Right Side Panel



1 Fluidics maintenance door

Provides access to the fluidics system for performing maintenance.

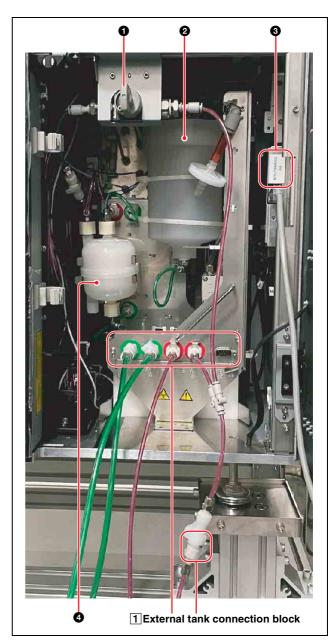
Press PUSH OPEN to open the door.

Notes

Chapter 1 Overview

- Be aware of the sharp door edges when the door is open.
- Press to open the door, and close the door gently. Do not use force to open and close the door.
- When closing the door, make sure that any tubing and cables exit from the bottom. If they exit from other places, tubing may be pinched, leading to clogging, and cables may be become disconnected.

Interior (Right Side Panel)



1 Valve

Used to de-bubble the lines through which the sheath fluid flows when cleaning the sample probe.

For details about operation, see "De-bubbling the Sheath Line" (page 158).

2 Pressurized tank

Supplies sheath fluid to the fluidics system for acquisition. The tank has a capacity of 500 ml.

3 PC CONNECTION connector

Connects directly to the host computer using the supplied PC connection cable.

Notes

• Do not disconnect the PC connection cable during operation. Data acquisition cannot be performed if the cable is disconnected.

• Do not connect any device other than the host computer using the PC connection cable. Device operation is not guaranteed if an external device is connected.

CAUTION

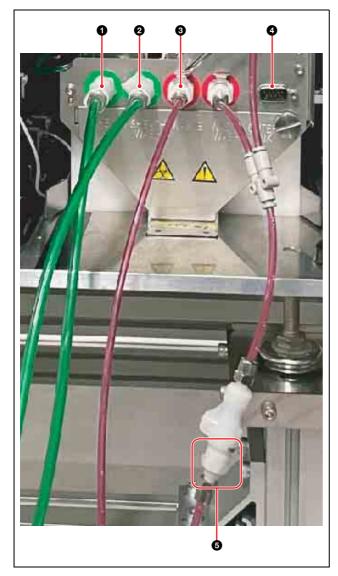
For safety, do not connect the connector for peripheral device wiring that might have excessive voltage to this port.

Follow the instructions for this port.

4 Sheath filter

Filters the sheath fluid flowing between the sheath tank and the fluidics system. The sheath filter is a maintenance item.

1 External tank connection block



1 SHEATH connector

Connects to the 10L sheath tank using the supplied sheath tank connection tube (green). It supplies sheath fluid used for acquisition.

2 SHEATH WASH connector

Connects to the 10L sheath tank using the supplied sheath tank connection tube (green). It supplies sheath fluid used for cleaning the fluidics system.

Тір

No distinction is made between the sheath tank connectors. Both connectors work with either connector on the tank.

3 WASTE connector

Connects to the 10L waste tank using the supplied waste tank connection tube (red). Waste fluid collected during acquisition is passed to the 10L waste tank.

4 OUTER TANK (external tank connection cable) connector

Connects to the MAIN UNIT connector on the 10L tank system using the supplied external tank connection cable.

Тір

No distinction is made between the waste tank connectors. Both connectors work with either connector on the tank.

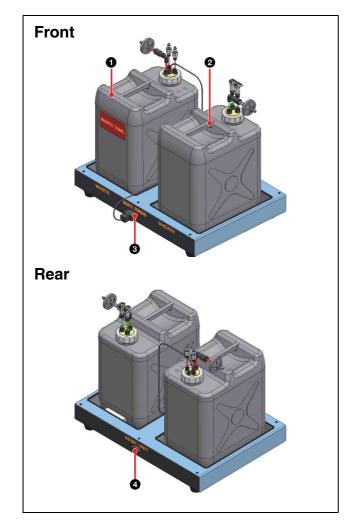
5 WASTE WASH connector

Connects to the 10L waste tank using the supplied waste tank connection tube (red). Waste fluid collected during fluidics system cleaning is passed to the 10L waste tank.

Tip

The supplied fluidics lines and cable are about 5 m (16.4 ft) in length.

10L Tank System (Supplied)



Chapter 1 Overview

1 10L waste tank (WASTE, red)

Collects waste fluid from the internal waste tank and the suction tank in the main unit.

An alarm is generated when the waste fluid rises above a set level detected by a sensor. The tank has a capacity of 10 liters.

The waste tank is a maintenance item.

2 10L sheath tank (SHEATH, green)

Supplies sheath fluid used for acquisition and cleaning the fluidics system.

An alarm is generated when the sheath fluid falls below a set level detected by a sensor.

The tank has a capacity of 10 liters.

The sheath tank is a maintenance item.

3 WASTE SENSOR connector

Connects to the waste sensor cable attached to the cap of the 10L waste tank.

4 MAIN UNIT (external tank connection cable) connector

Connects to the OUTER TANK (external tank connection cable) connector on the main unit using the supplied cable.

DANGER

Waste fluid may contain biological, chemical, or other agents. Always wear gloves and other protective clothing, mask, and goggles, as required, when handling collected samples.

Notes

- Check that fluidics lines and cables are securely connected to prevent leakage and malfunction.
- Check that connecting tubing and cables are not bent, squashed, or crimped.
- Avoid locations where the connecting tubing and cables may become snagged during operation.

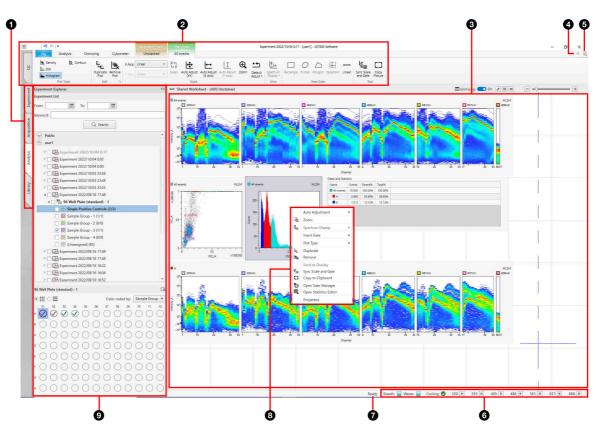
Main Window

Tips

- The menus and buttons displayed vary depending on the user account privileges.
- The screenshots displayed in this document may vary from the actual software.

Operating window

Example: Analysis window



1 Workflow tabs

Selects the various configuration and operation views. **[QC]:** Performs Daily QC and Performance QC.

- **[Experiment]:** Creates experiments used to acquired
- data.
- [Acquisition]: Configures instrument settings, and performs sample acquisition operations.
- [Analysis]: Performs analysis operations on the acquired data.
- [Library]: Used to manage the spectral reference library.

2 Ribbon

Displays the tool buttons that can be used in the displayed window.

The content of the ribbon varies depending on the selected tab.

3 Worksheet

Work area for displaying and editing the results from data acquisition and analysis.

4 Ribbon show/hide control

Clicking alternately shows the ribbon (\heartsuit) or hides the ribbon (\diamondsuit) .

5 Logout

Logs out the currently logged in user and displays the login window (*page 32*).

6 Hardware status

Displays icons indicating the status of the main unit, and the fluid levels in each tank.

When an error or warning occurs, \bigotimes (error) or \blacktriangle (warning) appears.

The laser icons displayed vary with the model.

- **Sheath:** Displays the status of the 10L sheath tank. When the fluid level in the 10L sheath tank becomes low or the main unit is disconnected from the 10L tank system, **A** appears.
- **Waste:** Displays the status of the 10L waste tank. When the 10L waste tank becomes full or the main unit is disconnected from the 10L tank system, **A** appears.

[Cooling]: Displays icons indicating the sample cooling status.

OFF: Cooling is off.

Rotating icon: Cooling is in progress Cooling state is being maintained

- **320:** 😹 Indicates whether the 320 nm laser is on or off.
- **355: *** Indicates whether the 355 nm laser is on or off.
- **405: *** Indicates whether the 405 nm laser is on or off.
- **488:** Indicates whether the 488 nm laser is on or off.
- **561:** Indicates whether the 561 nm laser is on or off.
- **637:** Indicates whether the 637 nm laser is on or off.
- **808:** Minimize Mathematical Base is a constraint of the second s

Tip

The status of the internal waste tank (for cleaning) and the suction tank is not displayed.

7 Status indicator area

Displays the status of ID7000 Software.

8 Context menu

Displays menu commands available when right-clicking an item in the displayed window.

9 Control pane

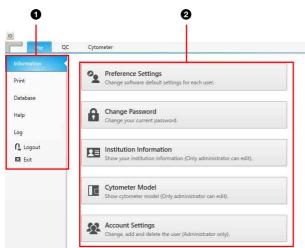
Displays the control buttons for operations that can be performed.

The control pane is a collapsible window element. The panel can be shown or hidden as required.

The available functions vary depending on the displayed window.

[File] tab (ribbon)

Example: [Information] window (for administrator)



1 Menu items

Displays the following menu items.

- **[Information]:** Displays commands used to configure user passwords and institution information, and to add/manage user accounts.
- [Print]: Prints analysis results, QC results, and other reports.

- **[Database]:** Displays commands used to import/export experiments, perform backups, and to check hard disk free capacity.
- **[Help]:** Displays commands used to display the Operator's Guide and to check software version information.
- **[Log]:** Displays commands used to display the error/ warning log.
- **[Logout]:** Logs out the currently logged in user and displays the login window (*page 32*).

[Exit]: Exits ID7000 Software (page 42).

2 Operation area

Displays function buttons and information related to the selected menu item.

ID7000 Software Operating Environment

The host computer must satisfy the following requirements in order to run ID7000 Software.

Operating system (OS): Microsoft Windows 10 Pro for Workstations (4 Cores plus)/Windows 11 Pro
CPU: Intel Xeon GOLD 6248 2.5 GHz 20C/40T or higher
Memory: 256 GB or greater
Network interface: 1000BASE-T (1)
Main storage: 8 TB HDD or greater
Monitor resolution: 1920×1200 or greater
OS language setting: Symbol for the decimal point is "." (period) and the digit separator is "," (comma).

Note

If the decimal point is set to a character other than "." (period), the following error message is displayed when starting ID7000 Software.

Invalid Language setting was detected. Please change "Decimal symbol" setting to ". (period)" according to the Operator's Guide. Press [OK] to close this application.

If this occurs, use the following procedure to change the decimal point to "." (period), then restart ID7000 Software.

- Open [Control Panel] > [Clock, Language, and Region] > [Region and Language].
- **2** Open [Additional settings...] on the [Formats] tab.
- **3** Set [Decimal symbol] to "." (period) on the [Numbers] tab.

Notes

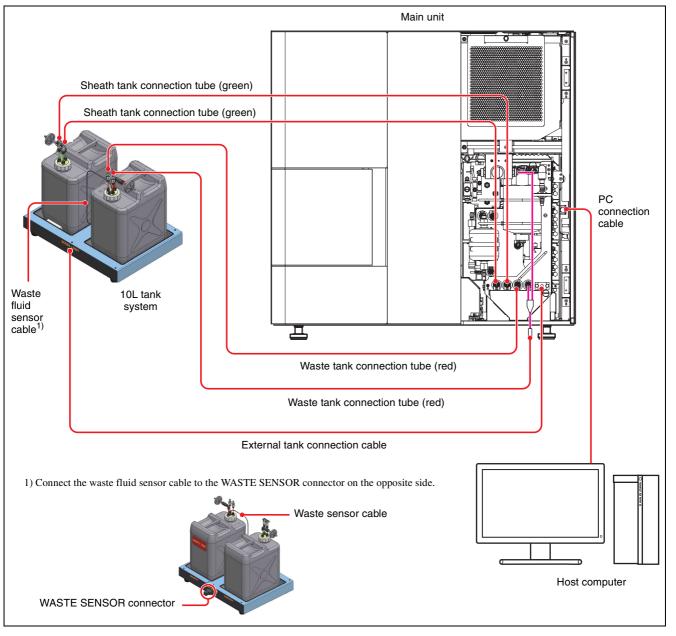
- Do not connect other devices or external networks to the network directly connecting the unit and computer to prevent a loss in measurement performance.
- When connecting the computer to the internet, connect via a system that provides a protection function, such as a router or firewall. If connected without such protection, security issues may occur.
- While using ID7000 software, do not run other applications or any loss intensive services, such as Windows Update, on the computer to prevent a loss in acquisition performance. For details about running applications, contact your computer administrator.

- Implement acquires that ensure information security for use of the computer with peace of mind. Examples: Update antivirus software definition files, run Windows Update regularly, configure a firewall, manage ID and password appropriately, and so on. For details, contact your network administrator or IT department.
- Synchronize your computer's clock regularly to ensure that the time information contained in your records is accurate. For details, contact your network administrator or IT department.
- For details about compatibility-verified antivirus software, contact your Sony distributor.



Checking Device Connections

Check that the main unit, 10L tank system, tank base, and host computer are connected correctly.



Note

Do not install the 10L tank system on top of the ID7000 unit or in a high position.

Logging In as an Administrator

To perform the initial configuration, it is necessary to log in to ID7000 Software as an administrator.

Notes

- Only one user can be logged in at a time.
- While using the unit, do not run other applications or any load intensive services to prevent a loss in measurement performance.
- ID7000 software operates correctly only when it is installed on the host computer controlling the instrument.
- **1** Select ID7000 Software on the Start screen to start the software.

When you launch ID7000 Software for the first time, the software user license agreement appears.

2 Select your country or region and read the license agreement, then select [I accept the terms in the license agreement] and click [OK].

For users in USA

END USER LICENSE AGREEMENT MPORTANT: BEFORE USING THE SOFTW	ADE DIFASE DEAD THIS	â
IND USER LICENSE AGREEMENT ("EULA") (. KOFTWARE YOU ARE AGREEING TO BE BOUN ULA FOR VOURSELF AND ANY ORGANIZA YOU ARE USING THIS SOFTWARE. IF YOU DO OF THIS EULA, YOU MAY NOT USE THE SOFTW	D BY THE TERMS OF THIS TION ON WHOSE BEHALF NOT AGREE TO THE TERMS	
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his EULA is a legal agreement between (i) epresent (You 7) and (ii) Sony Biotechnology overns your rights and obligations regarding OXY and/or its third party licensors (induc heir respective affiliates (collectively, the TT ogether with any updates/upgrades provide n-line or other electronic documentation fi ata files created by operation of such s SOFTWARE').	¹ Inc. ("SONY"). This EULA g the ID7000 software of ding SONY's affiliates) and HIRD-PARTY SUPPLIERS"), ad by SONY, any printed, or such software, and any	

For users in other countries/regions

JSA Japan International	
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LICENCE TERMS This EULA is a legal agreement between (i) you and any entity you represent ("you") and (ii) Sony Imaging Products & Solutions Inc. ("SONT'). This EULA governs your rights and obligations regarding the Drooos ostivare of SONY and/or its third party licensors (including SONT's affiliates) and their respective affiliates (collectively, the "THIRD- NARTY SUPPLIERS"), together with any updates/upgrades provided by SONTs, any printed, on-line or other electronic documentation for such software, and any data like created by operation of such software (collectively, the "SOFTWARE").	
) I accept the terms of the license agreement.	

ID7000 Software launches and the login window appears.

3 Enter your user name and password, then click [OK].

Tips

- The administrator account user name is set to "administrator" by default.
- The Sony field service engineer account user name is set to "sony-service" by default.

D7000 Soft	ware Login	<u>2000</u>	707	×
ID70C	0 Software			
Username:	user1			
Password:	•••••			
	Login	Exit		

If login is successful, the [QC] workflow tab appears.

If more than eight hours have elapsed since the last acquisition, the priming wizard appears. Click [Start] to perform priming.

Tip

If the administrator account password has not been configured, the [Set Password] dialog appears for the administrator account when launching the software. Set the password, then click [OK].

Username:	administrat	10	
*Password:			
*Confirm Password:			

 [New Password]: Enter a new password, comprising a combination of 8 to 20 alphanumeric characters (required item).
 [Confirm Password]: Enter the same password again for confirmation (required item).

If the password for the Sony field service engineer account is not configured, it is set to the same password as the administrator account.

Set Password		>
Username:	sony-service	
*Password:		
*Confirm Password:		
	OK Ca	ncel

Preconfiguring the Instrument

Preconfigure the following items before operating the system.

Setting Institution Information

- **1** Click the [File] tab, then click [Information] in the menu on the left.
- **2** Click [Institution Information].

ine (2C Cytometer
Print	Preference Settings
cont.	Change software default settings for each user.
Database	
Help	Change Password
heip	Change your current password.
Log	
C Logout	Institution Information
🖾 Exit	Show your institution information (Only administrator can edit).
	Cytometer Model
	Show cytometer model (Only administrator can edit).
	Account Settings
	Change, add and delete the user (Administrator only).

The [Institution Information] dialog appears.

3 Enter the name and address of the institution, then click [OK].

ID Institution Inform	ation				\times
Institution:					
Address:					
City:					
State:					
Zip Code:					
Country/Region:					
		0	K	Cancel	

The institution information is registered.

Chapter 2 Initial Setup

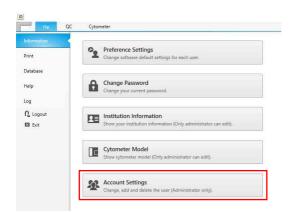
Configuring Users

This section describes how to manage user accounts in ID7000 Software.

Adding Users

Only administrator can add new users.

- 1 Click the [File] tab, then click [Information] in the menu on the left.
- **2** Click [Account Settings].



The [Account Settings] dialog appears.

3 Click [Add].

v jiro xxxd	\$10X.0X Li	e Science Division	Sony	Delete
				Edit
				Edit

The [Add Account] dialog appears.

4 Set each field.

D Add Account	×
*Username:	user2
*Password:	•••••
*Confirm Password:	•••••
*Full Name:	Sony Hanako
*Email:	XXX @XXX XXX
*Department:	Life Science Division
*Institution:	Sony
	Advanced Settings
	OK Cancel

[Username]: Enter the user name, of up to 20 alphanumeric characters, used when logging in to ID7000 Software (required item). [Password]: Enter the password, comprising a combination of 8 to 20 alphanumeric characters, used when logging in to ID7000 Software.
[Confirm Password]: Enter the same password again for confirmation.
[Full Name]: Enter the full name of the user.
[Email]: Enter the email address of the user.
[Department]: Enter the name of the department.
[Institution]: Enter the name of the institution.

5 Click [Advanced Settings].

The [Advanced Settings Dialog] dialog appears.

6 Set each item, then click [OK].



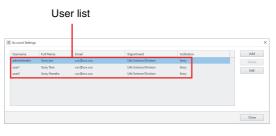
- **[Password Expiration]:** Set the interval before the password expires in the range 1 to 366 days. The default value is 120 days.
- [Account Lockout]: Set the number of times that an incorrect password can be entered in succession before the account is locked in the range 1 to 10. The default value is 5 times.

For details about unlocking a locked account, see "When your account is locked (general users)" (page 33).

Close the [Edit Account] dialog to return to the [Account Settings] dialog.

7 Click [OK].

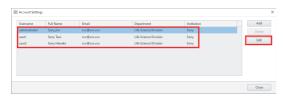
The user is registered in the user list in the [Account Settings] dialog.



Editing User Settings

Only administrator can edit user settings.

1 Select the user you want to edit in the [Account Settings] dialog, then click [Edit].



The [Edit Account] dialog appears.

2 Edit the settings, then click [OK].

Edit Account	
Username:	user2
*Password:	•••••
*Confirm Password:	
*Full Name:	Sony Hanako
*Email:	xxx@xxx.xxx
*Department:	Life Science Division
*Institution:	Sony
Unlock:	
	Advanced Settings
	OK Cancel

For details, see step 4 (page 27) in "Adding Users".

3 Click [Advanced Settings] to configure detailed settings related to passwords, as required.



- **[Password Expiration]:** Set the interval before the password expires in the range 1 to 366 days. The default value is 120 days. Selecting [At Next Login] will require the user to reset the password the next time they log in.
- [Account Lockout]: Sets the number of times that an incorrect password can be entered in succession before the account is locked in the range 1 to 10. The default value is 5 times.

For details about unlocking a locked account, see "When your account is locked (general users)" (page 33).

The account settings are updated.

Changing Passwords

Individual users can change their password used to log in to ID7000 Software.

- 1 Click the [File] tab, then click [Information] in the menu on the left.
- **2** Click [Change Password].

,fie (QC Cytometer
nformation Print	Preference Settings Change software default settings for each user.
Database	
Help	Change Password Change your current password.
Log	
∩_ Logout ⊠ Exit	Institution Information Show your institution information (Only administrator can edit).
	Cytometer Model Show cytometer model (Only administrator can edit).

The [Change Password] dialog appears.

3 Set each item, then click [OK].

ID Change Password		\times
Username:	user1	
*Old Password:	•••••	
*New Password:	•••••	
*Confirm Password:	•••••	
	OK Cancel	

- [Old Password]: Enter the current password (required item).
- **[New Password]:** Enter a new password, comprising a combination of 8 to 20 alphanumeric characters (required item).
- [Confirm Password]: Enter the same password again for confirmation (required item).

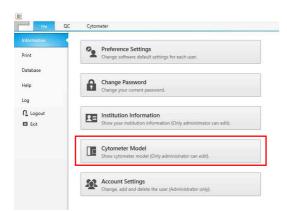
The password is updated.

Switching the Cytometer Model to Control

If a laser is added to the instrument at a later date, the cytometer model for control must be changed in ID7000 Software.

Only administrator can switch the cytometer model.

1 Click the [File] tab, then click [Cytometer Model] in the menu on the left.



The [Cytometer Model] dialog appears.

2 Select the cytometer model to connect to using ID7000 Software, and click [OK].

Selecting the model number enables the sample loader type and configured lasers that are supported by that model in [Configuration].

The cytometer model number can be checked by checking the status of the connected instrument on the [Help] screen (*page 119*).

D Cytometer	Model	×
Model No.:	LE-ID7000B	
Configur	ation	1
Sample	Loader: Auto Sampler	
Laser:	405 / 488 / 561 / 637	
		_
	OK Cancel	

The ID7000 settings are updated for the specified model.

Tip

A warning message is displayed if the cytometer model selected is different from the cytometer connected. This chapter describes the basic operating procedure sequence for operating the instrument, acquisition and analysis.

Тір

In this document, 5 ml tube racks, 96-well plates, and 384-well plates are collectively referred to as "(multi-well) plates," and 5 ml tubes and wells in which sample fluid is placed are collectively referred to as "samples."

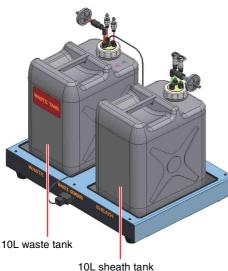
Workflow

Step 1	Check the 10L sheath tank and 10L waste tank (page 31)
	Check the 10L sheath tank and 10L waste tank comprising the 10L tank system.
	+
Step 2	Start the main unit (page 31)
	Turn on the power to start the ID7000.
	+
Step 3	Log in <i>(page 32)</i>
	Turn on the PC, start ID7000 Software, and log in to the ID7000.
	+
Step 4	Prepare the instrument (page 34)
	Run priming, Daily QC, and Performance QC before starting acquisition.
	+
Step 5	Prepare for acquisition (page 38)
	Conduct single stain acquisition and calculate/register a spectral reference, then create an
	experiment to acquire data for the sample.
	+
Step 6	Conduct sample acquisition and analysis <i>(page 39)</i>
	+
Step 7	Run manual cleaning (if continuing acquisition) (page 156)
	Go to step 8 if shutting down the main unit.
	+
Step 8	Shut down (page 42)
	Perform pre-shutdown cleaning and then shut down the main unit.

Checking the 10L Sheath Tank and 10L Waste Tank

Before starting the main unit, check the 10L sheath tank and 10L waste tank in the tank base for the following.10L sheath tank is filled with sheath fluid

- IOL sneath tank is filled with sneat
- 10L waste tank is not full



For details about filling the sheath tank, see "Refilling the 10L Sheath Tank with Sheath Fluid" (page 154).

For details about replacing the 10L waste tank, see "Emptying/Replacing the 10L Waste Tank" (page 153).

Note

Always place the 10L sheath tank on the SHEATH side of the tank base, and the 10L waste tank on the WASTE side.

Tip

An alert message is displayed on the host computer if the fluid level in the sheath tank gets too low during operation. When prompted, select whether to continue or to stop.

Starting the Main Unit

Turn on the power supply and start the ID7000 using the following procedure.

Notes

- Pay close attention to the temperature of the operating environment to ensure instrument performance. (Specifications: 17.5 °C to 27.5 °C (63.5 °F to 81.5 °F))
- Also, be aware of changes in temperature of the operating environment. (Specifications: < 5 °C (< 9 °F))
- **1** Check the level in the tanks within the 10L tank system.

Also, check that both the fluidics lines and the external tank connection cable are connected correctly.

2 Turn on the two main power switches on the lower right of the left side panel.



Main power switches

Tip

During normal operation, the two main power switches should be left turned on. Use the POWER button on the front panel to turn power on/off on a normal basis.

3 Press the POWER button on the front panel of the main unit to turn on the unit.

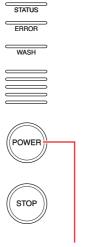
Note

Plates and the 5 ml tube rack should be loaded after the main unit is turned on. If one is already loaded, remove it before turning the main unit on. A startup error will occur if a plate or the 5 ml tube rack is already loaded when the main unit is turned on.

When the unit is turned on, the POWER indicator lamp is lit green.

The STATUS indicator is lit green when the ID7000 starts up.

The front door can be opened and closed when the STATUS indicator is lit orange.



POWER indicator

After instrument startup, wait about two to seven minutes for the unit to become ready for acquisition (time varies with instrument configuration).

Logging In

Turn on the PC, start ID7000 Software, and log in to the ID7000.

Notes

- Only one user can be logged in at a time.
- While using the unit, do not run other applications or any load intensive services to prevent a loss in measurement performance.
- The instrument can only be operated from a computer with ID7000 Software installed.
- To connect with the instrument main unit and run samples, launch ID7000 Software. To run data analysis without starting the instrument main unit, launch ID7000 Software Analysis Mode.
- **1** Select ID7000 Software on the Start screen to start the software.

ID7000 Software launches and the login window appears.

2 Enter your user name and password, then click [OK].

are		

If login is successful, the [QC] workflow tab appears.

If more than eight hours have elapsed since the last priming, the priming wizard appears. Click [Start] to perform priming.

If more than one week has elapsed since the last time the sheath line was de-bubbled, the sheath line debubble wizard appears before priming. Click [Start] to perform the sheath line de-bubble process.

If more than one week has elapsed since the last optics alignment, the optics alignment wizard appears. Select the lot number of the beads to use and click [Start] to run optics alignment.

For details, see "Adjusting the Optical Axis" (page 87).

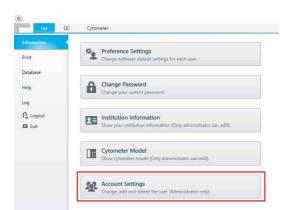
When your account is locked (general users)

A general user account will be locked after repeated unsuccessful attempts to enter a password. When locked, the following message appears on the login screen and account access is disabled.

10700)0 Software	
10100		
	10:22	_
Username:	user2	
Password:	•••••	

If this occurs, use the following procedure to unlock the account. Only administrators can unlock an account.

- 1 Click the [File] tab, then click [Information] in the menu on the left.
- **2** Click [Account Settings].



The [Account Settings] dialog appears.

3 Select the user whose account is locked, then click [Edit].

de Scence Division Sony Edit Edit
Edit
Edit

The [Edit Account] dialog appears.

If the account is locked, a check mark is displayed in the [Unlock] checkbox.

Username:	user2
*Password:	•••••
*Confirm Password:	
*Full Name:	Sony Hanako
*Email:	xxx@xxx.xxx
*Department:	Life Science Division
*Institution:	Sony
Unlock:	
	Advanced Setting

4 Clear the check mark in [Unlock], then click [OK].

*Department:	Life Science Division
*Institution:	Sony
Unlock:	
	Advanced Settings
	OK Cancel

The account is unlocked and the user can now log in again.

5 Click [Close] to close the [Edit Account] dialog.

When your account is locked (administrator account)

The administrator account will be locked after repeated unsuccessful attempts to enter a password. When locked, the "Your account is locked. Please contact your Sony distributor for service." message appears on the login screen and logging in using the administrator account is disabled.

If this occurs, contact your Sony distributor for service.

Preparing the Instrument

This section describes the operation flow for Step 4 in *"Workflow" (page 30)*.

After starting the ID7000, run priming, and then Daily QC and Performance QC daily before starting acquisitions to check the instrument status. Update the QC beads lot, as required.

• Priming

Flush sheath fluid through the system.

• Daily QC

Daily QC uses Align Check beads, and automatically performs the following checks and calibration. In addition to regular checks, it can also be performed at any time if, for example, there is some doubt about acquisition results.

- Instrument status check (detection sensitivity, stability, etc.)
- Flow rate calibration
- Laser delay adjustment
- Standardization adjustments
- Detection sensitivity calibration

• Performance QC

Performance QC uses 8-peak beads, and performs the following checks automatically.

Instrument status check (fluorescence detection performance)

Update QC beads lot

You can use a new beads lot you have obtained by updating the QC beads lot information.

Notes

- Pay close attention to the temperature of the operating environment to ensure instrument performance. (Specifications: 17.5 °C to 27.5 °C (63.5 °F to 81.5 °F))
- Also, be aware of changes in temperature of the operating environment. (Specifications: Change in temperature after Daily QC of $< 5 \,^{\circ}$ C ($< 9 \,^{\circ}$ F))

Tips

- You can also monitor the QC results and history.
- The QC Criteria settings can be changed by users logged in with Administrator privileges. For details, see "Setting QC Criteria" (page 104).
- To acquire data in Standardization Mode, it is recommended that you perform Flow Rate Calibration and Optics Alignment, in that order, before running Daily QC. For details, see "*Calibrating the Fluidics System*" (*page 87*) and "*Adjusting the Optical Axis*" (*page 87*).
- To adjust the instrument to obtain higher accuracy, it is recommended to wait about 30 minutes after turning on the instrument (power button only) to allow the instrument to warm up.

Priming

Prime the fluidics lines by flushing with sheath fluid.

Note

If more than eight hours have elapsed since the last acquisition, the priming wizard appears. Always execute the priming function.

For details, see "Priming" (page 155).

Running Daily QC

Daily QC uses Align check beads.

Tips

- Daily QC takes approximately 12 minutes.
- Store Align Check beads in a cool location, and use them immediately after preparation.
- Daily QC uses Align check beads. Shake the Align Check beads bottle vigorously before use to obtain a uniform bead concentration. Place two drops of beads in a sample tube and mix with 450 µl of DI water or PBS (saline) solution.
- 1 Click [Daily QC] on the [QC] workflow tab.

The [Daily QC] window appears.

2 Click [Start Daily QC].



Note

[Start Daily QC] is enabled only when no plate is loaded.

The [Daily QC Wizard] window appears.

3 Follow the on-screen instructions to run Daily QC.

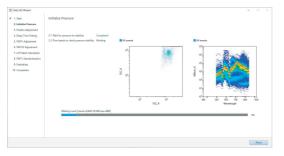
Duity QC Waard		-		
1. Start	Start Daily QC			
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	Start		Cancel	

Note

Use the same beads as the lot number selected in [Bead Lot Number] for QC. QC may not be performed correctly if the beads do not match the lot number.

For details about beads lot files, see "Updating the Beads Lot File" (page 103).

The progress of Daily QC is displayed in the wizard.



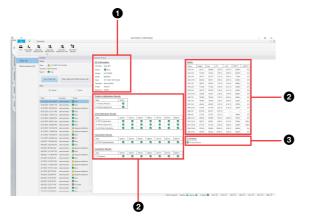
When Daily QC finishes, the QC result is displayed on the [Daily QC Report] sheet.

Checking Daily QC results

The following information is displayed in [Daily QC Report].

For details, see "[Daily QC] Window" (page 121).

• When [History] is selected in [View]



1 QC information area

Displays the following information when QC is performed.

- QC type
- QC result (Pass, Fail, or Requires Attention)
- Model number
- Date
- Operator
- QC execution mode
- Beads lot information

2 QC result display area

Displays execution results of each QC step.

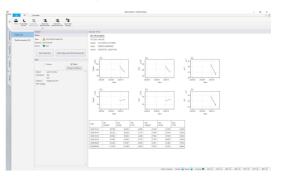
3 Comments display area

Displays the reason if the QC result is "Fail" or "Requires Attention."

• When [Trend] is selected in [View]

Displays a line graph showing the changes of each statistic over time with numerical values in a tabular format in sets of up to nine items.

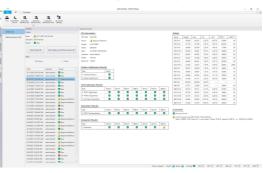
To change the statistics values displayed and the display period, click [Change Conditions] and edit the values in the displayed dialog.



• If Daily QC fails

Check the error description, and perform the following.

- ① Run a flow cell purge (*page 156*) or run priming (*page 155*).
- 2 Run [Flow Rate Calibration] (page 87).
- ③ Run [Optics Alignment] (page 87).
- (4) Check the beads preparation.
- If the Daily QC result is "Requires Attention" Check the alert description, and perform the following.



- ① Check the QC Criteria settings (page 104).
- ② Run a flow cell purge (page 156) or run priming (page 155).
- ③ Run [Flow Rate Calibration] (page 87).
- ④ Run [Optics Alignment] (page 87).
- (5) Check the beads preparation.

Tips

• The "Requires Attention" alert is displayed if the user-defined QC Criteria settings are exceeded or

if the laser is outside specifications (excludes the 488 nm laser).

• Even if "Requires Attention" is displayed, flow rate calibration, laser delay adjustment, standardization adjustment, and detection sensitivity calibration are completed.

Running Performance QC

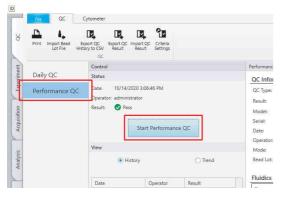
Performance QC uses 8-peak beads.

Tips

- Performance QC takes approximately five minutes.
- Store 8-peak beads in a cool location, and use them immediately after preparation.
- Shake the 8-peak beads bottle vigorously before use to obtain a uniform bead concentration.
 Place two drops of 8-peak beads in a sample tube and mix with 450 µl of DI water or PBS (saline) solution.
- Click [Performance QC] on the [QC] workflow tab.

The [Performance QC] window appears.

2 Click [Start Performance QC].



Note

[Start QC] is enabled only when no plate is loaded.

The [Performance QC Wizard] window appears.

3 Follow the on-screen instructions to run Performance QC.

Performance QC Woard				
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	1-3. Sales final (or Number: Beel or Number) (Null You Null You			
		Stat	Cance	ē

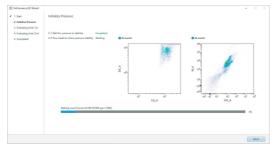
Note

Use the same beads as the lot number selected in [Bead Lot Number] for QC. QC may not be

performed correctly if the beads do not match the lot number.

For details about beads lot files, see "Updating the Beads Lot File" (page 103).

The Performance QC progress is displayed in the wizard.



When Performance QC finishes, the QC result is displayed on the [Performance QC Report] sheet.

Checking Performance QC results

The [Performance QC Report] has two display modes ([History] and [Trend]), where the basic structure is the same as for the [Daily QC Report]. The [Performance QC Report] displays the execution

result of each QC step, and the MESF, Q, B, and other values as statistics to indicate the fluorescence detection performance.

For details, see "[Performance QC] Window" (page 122).

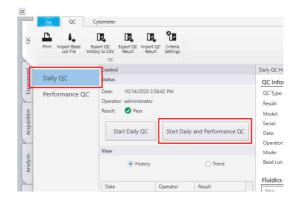
Running Daily QC and Performance QC Consecutively

You can run Daily QC and Performance QC consecutively.

Click [Daily QC] on the [QC] workflow tab.

The [Daily QC] window appears.

2 Click [Start Daily and Performance QC].



Note

[Start Daily and Performance QC] is enabled only when no plate is loaded.

The [Daily and Performance QC Wizard] window appears.

3 Follow the on-screen instructions to run Daily QC and Performance QC.

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1. Stert	Start Daily and Performance QC			
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	Bend Lot Number of AlignCheck Beads: 1402712/44.00 *			
	Bend Lot Number of 8 Peak Bendy: NIX21132V8422 *			
	1-5. Start Daily and Performance QC Confirm that the loader floor is closed, sick Start.			
	2at		Cancel	

Note

Use the same beads as the lot number selected in [Bead Lot Number] for QC. QC may not be performed correctly if the beads do not match the lot number.

For details about the beads lot file, see "Updating the Beads Lot File" (page 103).

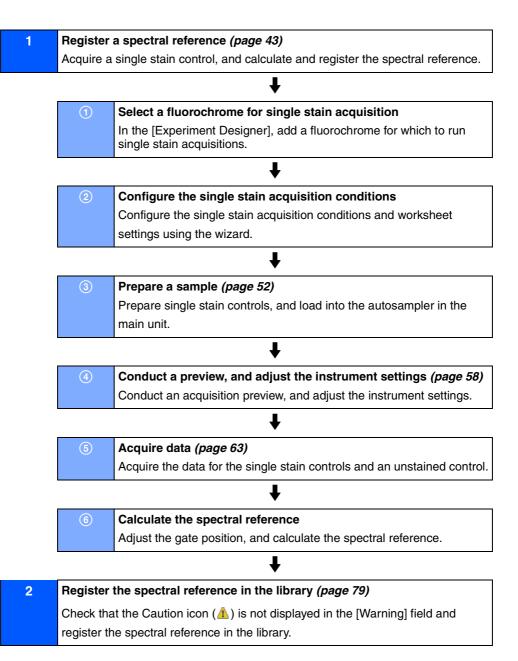
Preparing for Acquisition

This section describes the operation flow for Step 5 in "Workflow" (page 30).

Conduct single stain acquisition and calculate/register the spectral reference, then create an experiment to acquire data for the sample. The single stain acquisition and spectral reference calculation and registration is performed using a wizard.

For details about conducting simple acquisitions immediately, or creating an experiment from an existing experiment, see "Creating a New Experiment using a Template or Existing Experiment" (page 49).

Workflow



Running Sample Acquisition and Analysis

This section describes the operation flow for Step 6 in "Workflow" (page 30).

Experiment Acquisition and Analysis Create an experiment to acquire data for the sample (page 47) 1 Create an experiment to acquire data for the sample using the [Experiment Designer] wizard. 1 2 Edit the experiment (page 54) Add plates and sample groups, and edit the sample information. T 3 Prepare a sample (page 52) Prepare a sample for analysis, and load it into the autosampler in the main unit. 4 Conduct a preview, and adjust the instrument settings Configure the detector and other settings while conducting a preview. Previewing (page 58) Configuring instrument settings (page 59) 5 Run auto acquisition (page 65) Automatically acquire data for the sample. 6 Load a spectral reference Select and load spectral references to use for spectral unmixing from the spectral reference library. Loading an existing spectral reference (page 80) Apply spectral unmixing 7 Applying spectral unmixing (page 80) Adjusting spectral unmixing manually (Spectrum) (page 81) Analyze data 8 Editing plots (page 69) Editing gates (page 72) Customizing the statistics table (page 74)

Export analysis results

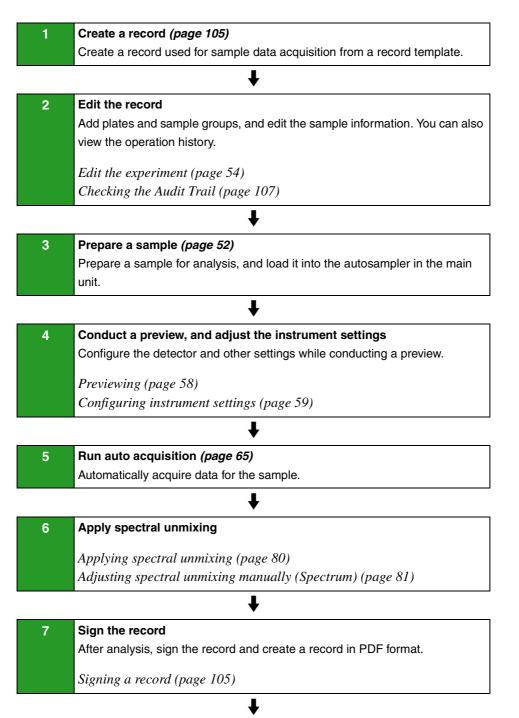
Exporting FCS file (page 97) Exporting statistics table information as a CSV file (page 75) Copying a plot to the clipboard (Copy to Picture) (page 141)

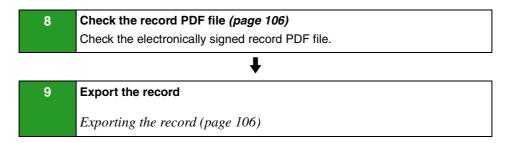
10 Create a template

Saving an experiment as a template (page 94)

Record Acquisition and Analysis

9





Shutting Down the Main Unit

The main unit should be shut down using ID7000 Software under normal circumstances.

Note

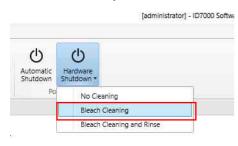
PBS (saline) can be used instead of DI water as the sheath fluid, subject to changes in the instrument settings. When using PBS (saline), [Long Term Storage] should be run before shutting down if you are not going to use the instrument for a week or longer.

For details, see "Maintenance Before Non-use for Extended Periods" (page 158).

Tips

- You can also shut down using the POWER button on the front panel, but it is recommended you shut down from ID7000 Software under normal circumstances.
- If the main unit is turned off and on again using the POWER button on the front panel or the main power switch, it will be necessary to restart ID7000 Software.
- In bleach cleaning, use a solution of approximately 1% sodium hypochlorite solution and sheath fluid.
- A 1% solution of sodium hypochlorite can be created by diluting household bleach by a factor of about 5.
- 1 Click [Hardware Shutdown] in the [Power] group on the [Cytometer] tab of the ribbon, and select [Bleach Cleaning].

To include a rinse cycle, select [Bleach Cleaning and Rinse]. In this case, prepare commercial rinsing fluid solution for flow cytometers beforehand.



The bleach cleaning wizard starts.

2 Follow the on-screen instructions to clean the instrument.

Also, refer to "Cleaning Before Shutting Down" (page 160).

[Bleach Cleaning] takes approximately eight minutes.

3 When cleaning finishes, a message appears. Click [OK].

- **4** Click [Shutdown].
 - B Cytometer Shutdown Blackt Cleaning
 ×

 I . Start
 Cleaning was completed.
 -

The main unit shuts down, and the power supply turns off.

5 Click the [File] tab, then click [Exit] on the left side to exit ID7000 Software.

Fie Q	C Cytometer
formation	
	Preference Settings
Print	Change software default settings for each user.
Database	
**	Change Password
Help	Change your current password.
Log	
N Logout	Institution Information
🖾 Exit	Show your institution information (Only administrator can edit).
	Cytometer Model
	Show cytometer Model Show cytometer model (Only administrator can edit).

ID7000 Software exits.

6 See "*Regular Cleaning*" (*page 160*) for details about cleaning the multi-well plate holder.



This chapter describes how to create and register a spectral reference, and how to create an experiment.

Registering a Spectral Reference

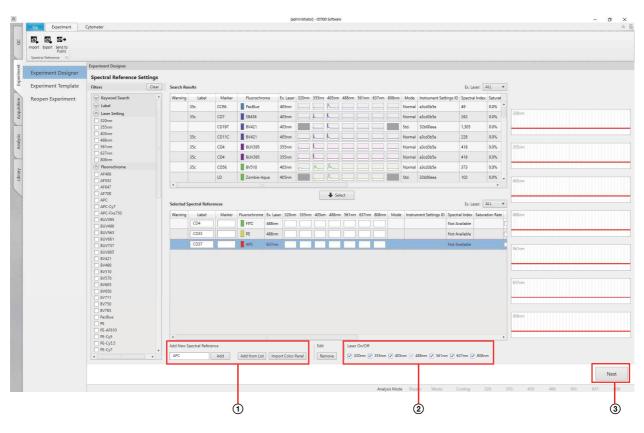
Registering a Spectral Reference using the Wizard

Acquire single stain controls, and calculate and register the spectral references using a wizard.

1 Click the [Experiment] workflow tab, and click [Experiment Designer].

The [Experiment Designer] window appears.

2 Add a fluorochrome for which to acquire a single stain control.

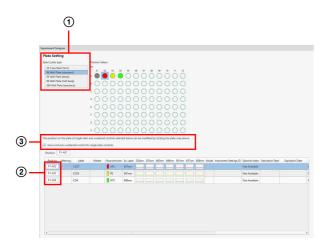


(1) Add the fluorochromes to acquire in [Selected Spectral References].

For details about adding, see "Adding Fluorochromes for Acquisition" (page 45). If a fluorochrome is added by mistake, you can remove it by selecting it in [Selected Spectral References] and clicking [Remove].

② Select the lasers for use.

3 Select the plate type in the [Plate Setting] step in the [Experiment Designer] wizard, then click [Next].



- ① Select the plate type.
- ② Specify the position of the single stain control well.

Enter the position of the well in [Position] in the color palette.

You can enter the position by clicking the well in the plate map.

- To acquire the unstained control, place a check mark in [Use a common unstained control for single stain controls], and enter the position of the unstained control well in [Position]. You can enter the position by clicking the well in the plate map.
- **4** Check the instrument settings and configure the acquisition conditions in the [Instrument Settings] step, then click [Next].

You can also configure all instrument settings using [Import].

strument Settings				
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[Cleaning Mode]: Select the cleaning method used between acquisition of each well.

- [Inner and Outer]: Clean the interior and exterior of the sample probe by backflushing with sheath fluid.
- [Inner Only]: Clean the interior of the sample probe only by backflushing with sheath fluid.
- [No Wash]: Skip sample probe cleaning.

[Acquisition Offset Time]: Set the offset time between the start of data acquisition of each well in units of seconds.

[Low Dead Volume]: To reduce the number of acquisition samples that are discarded, place a check mark in [Enable low dead volume mode].

[Agitation]: Set agitation settings.

Place a check mark in [Enable sample agitation] to enable agitation, and select the agitation method.

[Pattern]: Select the agitation method.

- [Once]: Agitate only once before sample acquisition.
- [Cyclic]: Agitate repeatedly at regular intervals during sample acquisition.
- [Continuous]: Agitate continuously during sample acquisition.
- [Mode]: Select the agitation mode.
- [Normal]: Agitate normally.
- [Short]: Agitate gently

Tips

- When [Low Dead Volume] is enabled, [Agitation] is set to [Once] (fixed). To use [Cyclic] or [Continuous], first disable [Low Dead Volume].
- When [Low Dead Volume] is enabled, the height of each well is detected before the acquisition begins, which increases the overall acquisition time.
- **5** Configure the acquisition order for wells in the [Acquisition Settings] step, then click [Next].

Experiment Designer	
Acquisition Sett	ngs
Order:	$●$ $₹$ Left-To-Right \bigcirc $𝔐$ Top-To-Bottom

[Order]: Select the well acquisition sequence.

6 Enter the metadata for the experiment and sample in the [Information] step, then click [Next].

Experiment Designer	
Information	
Experiment Information	
Name:	Experiment 2022/08/27 21:04
Investigator (optional):	user1
Operator (optional):	user1
Memo (optional):	
Plate Information	
P1 Name:	96 Well Plate (standard) - 1
P2 Name:	
P3 Name:	
P4 Name:	
P5 Name:	
Sample Group Information	
Name:	Single Positive Controls

7 Check the experiment settings in the [Experiment Preview] step, then click [Create Experiment].

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The experiment is created, and the [Acquisition] workflow tab is displayed.

8 Perform single stain acquisitions.

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	Tend And And And	

(1) Prepare samples.

Prepare single stain controls, and load into the autosampler in the main unit.

For details, see "Preparing a Sample" (page 52).

⁽²⁾ Conduct a preview, and adjust the instrument settings.

For details, see "Previewing" (page 58) and "Configuring Instrument Settings" (page 59).

3 Acquire data.

Acquire the data for the single stain controls and an unstained control.

For details, see "Acquiring Data" (page 63).

9 Adjust the gate position, and calculate the spectral reference.

For details, see "Creating a Spectral Reference" (page 76).

Mark Orient Raine Mark Orient Raine Mark							Universal Negative			Spectral Reference	8
Image: Description of the state of	Fluoro	chrome		Add			0	• Ex Laset A	U. *	220an	
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AuteRoomseance *			The cost of					• R (Allandy			
								Autofluore	scence *		

10 Register the spectral reference in the library.

For details, see "Creating a Spectral Reference" (page 76).



Tip

A spectral reference for a fluorochrome with a caution icon (1) displayed in the [Warning] field on the right may have a problem with quality. Adjust the gate settings, reacquire the single stain control, and recalculate.

Adding Fluorochromes for Acquisition

You can add fluorochromes to acquire using three methods.

Entering a fluorochrome name

In the [Experiment Designer] window, enter the name of a fluorochrome in the input field in [Add New Spectral Reference] and click [Add] to add the fluorochrome to the [Selected Spectral References] list.

Add New Spectral Reference			
	Add New Spectral I	Keterence	

Selecting from a list

1 In the [Experiment Designer] window, click [Add from List] in [Add New Spectral Reference].

Add New Spectra	al Reference		
FITC	Add	Add from List	Import Color Panel

The [Add New Fluorochromes from List] dialog appears.

2 Place a check mark in the fluorochrome(s) to add, then click [OK].

Clear the check marks for any unwanted fluorochromes that are selected. In [Ex. Laser], specify a laser to filter the fluorochromes displayed in the list.

Number of selection 11 / 100 Ex.	Laser: ALL 🔻	
AF700	ALL 320nm	320mm
AF750	355nm	
AF790	405nm	
AmCyan	488nm	
APC	561nm	355nm
APC-AF700	637nm 808nm	
APC-AF750	SUSHIM	
APC-Cy5.5		
APC-Cy7		
APC-eFluor780		
APC-Fire750		
APC-Fire810	1	
APC-H7		
APC-iFluor700		488ma
APC-iFluor750		
APC-R700		
APC-Vio770		
BB515		561nm
✓ BB630		
BB660		
BB700		
BB755		637nm \Lambda
BB790	÷	Λ
Fluorochrome Information		
Laser: Emission Spectru	m	808nm

The fluorochrome is added to the list in [Selected Spectral References].

Importing from Color Panel

You can import spectral reference fluorochome data from a list that was initially exported as a CSV file using [Color Panel Settings] > [Export] on the [Color Panel] tab of the [Unmixing Settings] dialog.

1 In the [Experiment Designer] window, click [Import Color Panel] in [Add New Spectral Reference].

Add New Spectral	Reference			
FITC	Add	Add from List	Import Color Panel	

The [Import Color Panel from CSV file] dialog appears.

2 Click [Browse], specify a CSV file, and click [Import].

D Impo	rt Color F	Panel from CSV F	ile					-		×
Input F	ile: C:\	Users\fcm\Desk	top\0513	\sample1.c	5V				Browse	
Line	Marker	Fluorochrome						-		
2		FITC								
3		PE								
4		APC								
	s in 0 line ings in 0									
						Imp	oort		Cancel	

The fluorochrome is added to the list in [Selected Spectral References].

Creating an Experiment

Create an experiment used to acquire data.

Creating a New Experiment using the Wizard

This section describes how to create a new experiment using the [Experiment Designer] wizard in the [Experiment] window.

1 Click the [Experiment] workflow tab, and click [Experiment Designer].



The [Experiment Designer] window appears.

2 Select a spectral reference to use.



- ① Specify keywords and filter criteria to search for a spectral reference.
- ② Select a spectral reference in the [Search Results] list.

You can select more than one spectral reference by pressing and hold the Shift key or Ctrl key and clicking other spectral references.

③ Click [Select].

The selected spectral reference is added to the list in [Selected Spectral References]. You can inspect the spectral waveform by

selecting the row for a spectral reference.

- (4) Click [Next].
- 3 Select the plate type in the [Plate Setting] step in the [Experiment Designer] wizard, then click [Next].

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ete Set																			
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ni stat	d special re	ineries.																	
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4 Check the instrument settings mode in the [Instrument Settings] step, then click [Next].

You can also configure all instrument settings using [Import].

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trument Settings				
Anada U 2010 Anak U 2010 Anak Navel Synthesis Analysis Anatomic Anatomic Analysis Anatomic Anatomic An	Last Ov08 Last Ov08 Didden 200e 400e 400e 300e 300e 400e 500e 500e 500e 500e	- Denote Nameda (Elever U) Stronger (0) Stronger (0) States 40 40 mm 400 40 mm 400	 He Catigit Event Serget Nov Kare Serget Nov Kare Age and on Other Time E Age and on Other Time E Age and on Other Time E	Aspatics lating. Chaning Mails Inter and Outer Aspansion Office Aspansion Aspansion Aspan Aspan
	Aphrice Law Dent Yolume O Aphrice O	Auto Augure Preferences Canterie	Deaning Cleaning Mode Inner and Duter	
		ele omal		

[Cleaning Mode]: Select the cleaning method used between acquisition of each well.

- [Inner and Outer]: Clean the interior and exterior of the sample probe by backflushing with sheath fluid.
- [Inner Only]: Clean the interior of the sample probe only by backflushing with sheath fluid. • [No Wash]: Skip sample probe cleaning.
- [Acquisition Offset Time]: Set the offset time between the start of data acquisition of each well in units of seconds.
- [Low Dead Volume]: To reduce the number of acquisition samples that are discarded, place a check mark in [Enable low dead volume mode]. [Agitation]: Set agitation settings.

Place a check mark in [Enable sample agitation] to enable agitation, and select the agitation method.

[Pattern]: Select the agitation method.

- [Once]: Agitate only once before sample acquisition.
- [Cyclic]: Agitate repeatedly at regular intervals during sample acquisition.
- [Continuous]: Agitate continuously during sample acquisition.

[Mode]: Select the agitation mode.

- [Normal]: Agitate normally.
- [Short]: Agitate gently

Tips

- When [Low Dead Volume] is enabled, [Agitation] is set to [Once] (fixed). To use [Cyclic] or [Continuous], first disable [Low Dead Volume].
- When [Low Dead Volume] is enabled, the height of each well is detected before the acquisition begins, which increases the overall acquisition time.
- **5** Configure the acquisition conditions in the [Acquisition Settings] step, then click [Next].

Experiment Designer			
Acquisition Setti	ngs		
Order:	● ≩ Left-To-Right	○ ∭, Top-To-Bottom	

[Order]: Select the well acquisition sequence.

6 Enter the metadata for the experiment and sample in the [Information] step, then click [Next].

eriment Designer	
nformation	
Experiment Information	
Name:	Experiment 2022/02/03 15:57
Investigator (optional):	administrator
Operator (optional):	administrator
Memo (optional):	
Plate Information	
Plate information P1 Name:	96 Well Plate (standard)
PT Name:	So men more (standard)
P2 Name:	
P3 Name:	
P4 Name:	
P5 Name:	
Sample Group Information	
Name:	Sample Group - 1
Sample Information	5
Name Prefix:	Well

7 Check the experiment settings in the [Experiment Preview] step, then click [Create Experiment].

Topetrunt 1	Eylonister				
R. R. S.					
	Equilment Designer				
xperiment Designer	Experiment Preview				
operment Template	Death				
	Second Lat	Sample Group Information			
leopen Experiment	* 2: 12 Species 2018/00 1140 < 2: 12 March 1011/0010 1140	Rana Sanja Dogi 1 Mongi Sanak Dogi Tang Tang			
	 Employee 1000 Employee 100 	(do bod			
	(S. II head-span				
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				Canod Bad	DI Destribute

The experiment is created, and the [Acquisition] workflow tab is displayed. Proceed to "*Previewing*" (*page 58*).

Creating a New Experiment using a Template or Existing Experiment

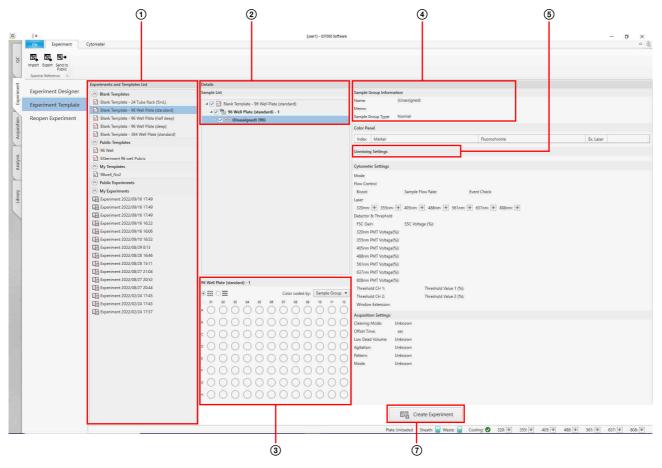
This section describes how to create a new experiment using existing templates and/or recent experiments.

1 Click [Experiment Template] on the [Experiment] workflow tab.

The [Experiment Template] window appears.

2 Create an experiment.

When a sample group is selected in the [Sample List] panel



When an experiment is selected in the [Sample List] panel

E. E.+					
rt Export Send to Public					
octral Reference 15					
	Experiments and Templates List	Details			
periment Designer	🔿 Blank Templates	Sample List	Experiment Information		
periment Template	Blank Template - 24 Tube Rack (5ml.)	🔺 🗹 💼 Blank Template - 96 Well Plate (standard)	Name:	Experiment 2022/10/03 22:35	
	Blank Template - 96 Well Plate (standard)	▲ 🗹 96 Well Plate (standard) - 1	Cytometer Type:	Auto Sampler	
open Experiment	Blank Template - 96 Well Plate (half deep)	▷ 28 (Unassigned) (96)			
	Blank Template - 96 Well Plate (deep)		Experiment Owner:	user1	
	Blank Template - 384 Well Plate (standard)		Investigator (optional):	user1	
	Public Templates 95 Well				
	EXteriment 96 well Pubric		Operator (optional):	user1	
	My Templates		Memo (optional):		
	2 98well_No2				
	Public Experiments				
	My Experiments				
	Experiment 2022/09/16 17:49				
	Experiment 2022/09/16 17:49			Off	
	Experiment 2022/09/16 17:49		Audit Trait	Off	
	Experiment 2022/09/16 16:22		Setting		
	Experiment 2022/09/16 16:06		Mode	Normal	
	Experiment 2022/09/10 16:52		Mode	Normal	
	Experiment 2022/08/29 0:13				
	Experiment 2022/08/28 16:46				
	Experiment 2022/08/28 15:11				
	Experiment 2022/08/27 21:04				
	Experiment 2022/08/27 20:52				
	Experiment 2022/02/24 17:45				
	Experiment 2022/02/24 17:43				
	Experiment 2022/02/24 17:37				
	44				
		Select plate or sample group to show information			
				E Create Experiment	
				Esta create cyperinent	
			Plate U	Inloaded Sheath 🔲 Waste: 🔲 Cooling: 🛇 320: 👫 355: 🕷	405: 🚼 488: 🗭 561: 📳 637: 🗐
					dana dana dana

For details about each item, see "[Experiment Template] Window" (page 126).

(1) Select a template or an experiment you want to use.

You can click an experiment or template in the [Experiment and Templates List] to check the settings.

To create a new experiment, you can select a template under [Blank Templates] that matches the multi-well plate or sample tubes to be used for the experiment.

[My Experiments] displays the experiments belonging to the logged-in user.

- ② Check the contents in the [Sample List] panel. If [Blank Template] is selected, a single multiwell plate is added to the experiment, according to the selected template.
- ③ Check the contents in the [Plate] panel. Clicking a multi-well plate in the [Sample List] panel will display information about the multi-well plate.

If [Blank Template] is selected, nothing will have been configured.

The shape of the well icon will vary depending on the worksheet mode configured for the worksheet.

- (circle): [Shared] mode
- (square): [Individual] mode

- (4) Edit the experiment information. Information about the item selected in the [Sample List] panel is displayed. Selecting an experiment in the [Sample List] panel allows you to edit settings, such as the name of the experiment.
- Check the contents in the [Unmixing Settings] panel.
 Clicking a sample group in the [Sample List] panel will display information about the spectral unmixing applied to the sample group.
 If [Blank Template] is selected, nothing will have been configured.
- 6 Check the instrument configuration mode.
- Click [Create Experiment]. The experiment is created, and the [Acquisition] workflow tab is displayed. Proceed to "Previewing" (page 58).

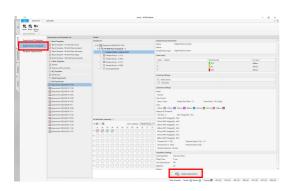
Opening an Existing Experiment

You can open an existing experiment and edit the experiment, for example, to reacquire data or add sample tubes.

1 Click [Reopen Experiment] on the [Experiment] workflow tab.

The [Reopen Experiment] window appears.

2 Select the experiment to open, then click [Reopen in Acquisition].



The selected experiment opens and can now be edited.

For details about each item, see "[Reopen Experiment] Window" (page 127).

Тір

If you select an experiment in [Public Experiments] and click [Reopen Experiment], a copy is created as an experiment belonging to the logged-in user and then opened.

Acquisition



This chapter describes the basic operating procedures for sample acquisition and for monitoring the instrument.

Acquisition can be performed manually or automatically. For details about automatic acquisition, see "Running Auto Acquisition" (page 65).

Preparing a Sample

Place samples for acquisition in a multi-well plate, and load the multi-well plate into the autosampler in the main unit. If using tubes, load the tubes using the supplied 5 ml tube rack.

Note

Always turn the main unit on before loading a multi-well plate or tube rack. Loading a multi-well plate or tube rack with the main unit turned off will result in incorrect operation.

Use only the following multi-well plates and sample tubes in the unit.

Multi-well plates (recommended: Corning)

- 96-well plates (flat bottom, U-bottom, V-bottom)
- 96-well plates (shallow well, round bottom, polypropylene)
- 96-well plates (deep well, round bottom, polypropylene)
- 384-well plates (flat bottom)

Sample tubes (recommended: Corning Falcon)

- 5 ml polystyrene round tubes
- 5 ml polypropylene round tubes

Sample fluid volume

- 96 well-plate: 55 to 250 µl
- 384 well-plate: 40 to 80 µl
- 5 ml tube: 100 to 4,000 µl

Tips

• Always stain samples in accordance with the instructions for the reagents. Also, note that the staining conditions will vary with the application, even when using the same reagents.

• It is recommended that the staining conditions be reviewed in advance (using titration and controls as required) to ensure appropriate staining. If unexpected acquisition results are obtained, check to see whether there is a problem with sample staining.

Notes

- Always remove the covers from multi-well plates and the caps from sample tubes before acquisition.
- Never use cracked or broken multi-well plates or tubes.
- Do not reuse multi-well plates or sample tubes.
- Note the following when using sample tubes.
- Fill with sample fluid within the specified range (above) to prevent spills and damage to the sample tube.
- Remove any fluid droplets or dirt adhering to the sample tube.

Cooling the sample

The unit can cool the multi-well plate holder to a temperature of 4 $^{\circ}$ C (39 $^{\circ}$ F).

Turn cooling on/off using [Cooling] in the [Plate] group on the [Cytometer] tab of the ribbon.

Without the multi-well plate holder, it takes less than 15 minutes to cool to 4 $^{\circ}$ C (39 $^{\circ}$ F).

Notes

- As the holder is cooled to 4 °C (39 °F), water droplets may adhere to the cooling unit, but this does not affect performance.
- When plates and tube racks are loaded into the cooling unit and cooled, surface tension due to water droplets between them and the cooling unit may make it difficult to remove the plate or tube rack. Use caution when removing.
- If a plate or tube rack is cooled before loading it in the cooling unit, many water droplets may appear on the cooling unit, but this does not represent a problem.

Тір

You can start cooling of the sample stage when logging in.

For details about setup, see "Configuring User Preference Settings" (page 86).

Loading a multi-well plate

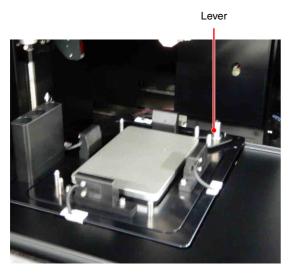
1 Open the front door.

Note

Do not open the front door during acquisition or when instructed not to do so by ID7000 Software.

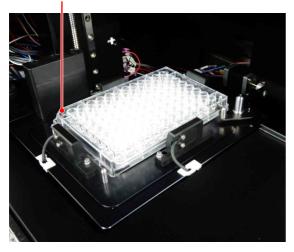
2 Place samples for acquisition in a multi-well plate, and load the multi-well plate onto the cooling unit.

Push the lever on the multi-well plate holder to the back and then position the multi-well plate. Load with sample A01 at front left.



Υ





3 Close the front door.

Loading sample tubes

You can load up to 24 sample tubes.

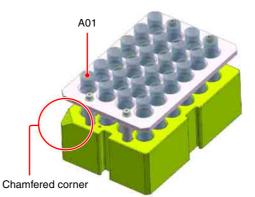
1 Open the front door.



Do not open the front door during acquisition or when instructed not to do so by ID7000 Software.

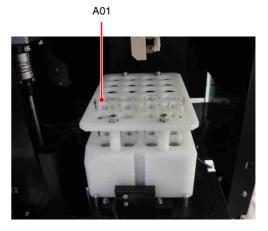
2 Place samples in sample tubes, and put the tubes in the supplied 5 ml tube rack.

The 5 ml tube rack has a chamfered corner for checking the correct orientation. Load the rack with the chamfered corner placed at the front left to load tube A01 at the front left.



3 Load the 5 ml tube rack into the multi-well plate holder in the main unit, with tube A01 placed at front left.

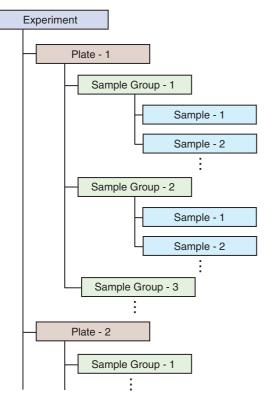
Push the lever on the multi-well plate holder to the back and then load the tube rack. Load with sample A01 at front left.



4 Close the front door.

Editing an Experiment

ID7000 Software allows you to run experiments with the following kind of structure.



Note

In version 1.1 and later, sample groups are shared for each plate of the same type (96-well plates, 384-well plates, 24-tube racks). This means that samples over multiple plates of the same type can be handled as a single sample group.

Adding a Plate

Operations are performed using buttons in the [Plate] group on the [Acquisition] tab of the ribbon. You can perform the same operation from the context menu by right-clicking an experiment in the [Experiment] panel.

					eet Inde		
nmixing	3	Cytomet	er s	Sample	Group - 1		
e,	000	889 X	000	0	888	888	80
Close	New	Delete	Load Template	Layout Editor	Export Layout to CSV File	Import Layout from CSV File	New
ent: 15				Plate		15e	

For details about each button, see "[Plate] group" (page 129) in "[Acquisition] Tab (Ribbon)".

To add a new plate to an experiment

1 Select an experiment in the [Experiment] panel, click [New] in the [Plate] group on the [Acquisition] tab of the ribbon, and select the type of plate to add.

ð	000	889 X	00	0	0000
Save as Template	New	Delete	Load Template	Layout Editor	Export Layout to CSV File
t 15	3	24 Tube R	ack (5mL)		
	5	et			
	9				
	3				
ter front d		884 Well F	late (standa	ard)	

The plate is added to the experiment.



To add a new plate to an experiment using a plate of an existing experiment

You can add a new plate to an experiment using a plate of an existing experiment.

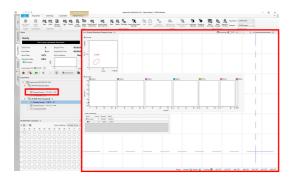
- 1 Select an experiment in the [Experiment] panel, click [New] in the [Plate] group on the [Acquisition] tab of the ribbon, and a new, empty plate to the experiment.
- **2** Select the added plate, and click [Load Template] in the [Plate] group on the [Acquisition] tab of the ribbon.

The [Load Plate Template] dialog appears.

Chapter 5 Acquisition

3 Select the template to apply, and click [Load].

- D Load Plate Template × Select a plate to load Experiment List ▲ 🛄 Experiment 2022/08/29 0:13 a 96 96 Well Plate (standard) - 1 Single Positive Controls (2/2) Sample Group - 1 (1/1) 88 Sample Group - 2 (0/0) 88 Sample Group - 3 (1/1) Sample Group - 4 (0/1) 88 (Unassigned) (92) a 96 Well Plate (standard) - 2 88 Single Positive Controls (0/2) 88 Sample Group - 1 (0/1) 88 Sample Group - 2 (0/0) 88 Sample Group - 3 (0/1) Sample Group - 4 (1/1) 80 (Unassigned) (95) ▲ 🛄 Experiment 2022/08/28 16:46 ⊿ 96 Well Plate (standard) Sample Group - 1 (1/1) 88 (Unassigned) (95) 4 🔚 Experiment 2022/08/28 15:11 4 96. 96 Well Plate (standard) - 1 Cancel Load
- **4** Select the plate on the [Experiment] panel, and check that the settings have been applied.



Editing Sample Information

You can edit metadata for a sample or plate.

 Right-click the plate whose information you want to edit on the [Experiment] panel, and select [Properties] from the context menu.



The plate properties dialog is displayed.

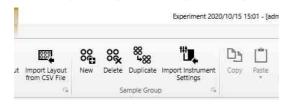
2 Edit each item, then click [Close].

96 Well Plate (standard)	1 - Properties	-		×
Plate Information				_
Туре:	96 Well Plate (standard)			
Name:	96 Well Plate (standard) - 1			
Date Created:	10/15/2020 3:04:23 PM			
Number (optional):				
Memo (optional):			Ĩ	
Acquisition Setting				
Order:	🛛 🛃 Left-To-Right 🔿 🕅 Top-To-Bottom			
	ок	11	Cancel	
		- 14 I.S.		

For details about editing sample metadata using Microsoft Excel or other software, see "Editing and Importing Plate Metadata" (page 93).

Editing a Sample Group

Operations are performed using buttons in the [Sample Group] group on the [Acquisition] tab of the ribbon. You can also perform the same operations by rightclicking a sample group on the [Experiment] panel.



For details about each button, see "[Sample Group] group" (page 130) in "[Acquisition] Tab (Ribbon)".

To add a new sample group to an experiment

Select the plate to which you want to add a sample group on the [Acquisition] panel, then click [New] in the [Sample Group] group on the [Experiment] tab of the ribbon to add a sample group to the experiment.

Exp	Expe	riment 2022/10	0/03 23:56		
	⊿ <mark>96</mark> 96	Well Plate (st	tandard) - 1		
	88	Sample Group	- 1 (0/0)		
	✓ 88	Sample Group	o - 2 (0/0)		
	88	(Unassigned) (9	96)		

Note

Sample groups are shared between multiple plates. This means that samples over multiple plates of the same type can be handled as a single sample group.

To add/remove a sample in a sample group

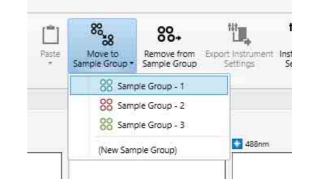
You can add a sample to a sample group or remove a sample from a sample group using the buttons in the [Sample] group on the [Acquisition] tab of the ribbon.



To add a sample to a sample group

Select the sample you want to add in the [Plate] panel, click [Move to Sample Group], and select the destination sample group.

15:22 - [administrator] - ID7000 Software



To remove a sample from a sample group

Select the sample you want to remove in the [Plate] panel, and click [Remove from Sample Group].

To move a sample to another sample group

Select the sample you want to move in the [Plate] panel, click [Move to Sample Group], and select the destination sample group.

You can change the worksheet mode for each sample, as required. For details about configuration, see "Changing the Worksheet Mode for Each Sample" (page 90).

Note

Removing a sample from a sample group will discard all settings and data relating to the sample.

For details about each button, see "[Sample] group" (page 130) in "[Acquisition] Tab (Ribbon)".

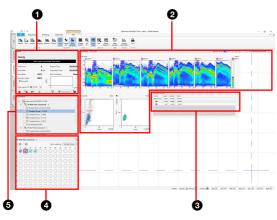
Chapter 5 Acquisition

[Acquisition] Tab Main Functions $\mid 57$

[Acquisition] Tab Main Functions

During data acquisition, the acquired data information is displayed on plots and in the statistics table on the [Acquisition] workflow tab.

This tab controls the data acquisition, recording, and stopping, as appropriate, while monitoring the data acquisition status. You can change the type of plots, change gates, and edit statistics in the [Gates and Statistics] table, as required.



1 [Status] panel

Controls data acquisition.

2 Plots

Displays acquired data in graphical form.

3 [Gates and Statistics] table

Displays statistics calculated automatically from acquired data.

4 [Plate] panel

Displays a list of wells. You can switch the display to list view or map view.

The shape of the well icon will vary depending on the worksheet mode configured for the worksheet.

) (circle): [Shared] mode

(square): [Individual] mode

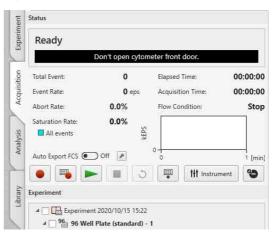
5 [Experiment] panel

Displays the structure of the experiment.

[Status] Panel Functions

The [Status] panel is used to control data acquisition. After data acquisition starts, the elapsed time since the start of data acquisition, the total number of events acquired, and the event rate acquired in events/second (eps) are displayed.

You control acquisition, as required, while monitoring the acquisition status.



[Auto Export FCS]: When enabled, an FCS file is automatically export upon acquisition. The

- (Acquire) and (Auto Acquire) buttons change to (Acquire) and (Auto Acquire).
- Displays the [Auto Export FCS Settings] dialog (page 98).
- (Acquire): Acquires and records data.
- (Auto Acquire): Automatically acquires data for multiple samples configured beforehand in Acquisition Target under the specified acquisition conditions.
- (Preview): Previews the acquisition. It does not start data recording.
- **Stop):** Stops data acquisition manually.
- **o** (**Restart**): During preview, resets the data display.
- (Load) / (Unload): Loads/unloads the multiwell plate.

[Instrument]: Configures settings related to lasers, detector, and flow rate.

Clicking this button displays the [Instrument Settings] dialog.

For details, see "Configuring Instrument Settings" (page 59)).

(Stopping Condition): Defines settings related to the data acquisition stop conditions.

For details, see "[Stopping Condition] dialog" (page 131).

Tip

If an automatic stop condition has been specified, data acquisition stops automatically when the stop condition has been satisfied.

Previewing

You can preview acquisition on the [Acquisition] workflow tab after creating an experiment.

Before starting preview

Check the following items.

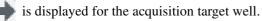
- Check the fluid levels in each of the tanks
- 10L tank system is connected correctly to the external tank connection block
- Click 💼 on the [Status] panel to load a sample.

Experiment	Plate Unload	ed		
tion	Total Event:	0	Elapsed Time:	00:00:00
Acquisition	Event Rate:	0 eps	Acquisition Time:	00:00:00
Ac	Abort Rate:	0.0%	Flow Condition:	Stop
Analysis	Saturation Rate:	0.0%	2	
Ane	Auto Export FCS 💽	Off 🛃	0	1 [mir
	🕘 🖷 🕨	- 🔳 🗧) 💼 🚻 İnstru	ment 🚷
Library	Experiment			

2 Select the plate to acquire on the [Experiment] panel.



3 Select the well/tube to acquire in the [Plate] panel, and click [Set Current Position] in the [Acquisition] tab of the ribbon.

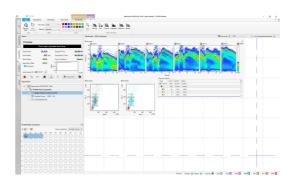


~··· ~ _								c		
• • =					Cold	or code	d by:	Samp	le Gro	γp ₹
02	03	04	05	06	07	08	09	10	11	12
	\bigcirc	С								
• 🔿 🔿	\bigcirc	С								
c () ()	\bigcirc	С								
	\bigcirc	С								
E () ()	\bigcirc	С								
F () ()	\bigcirc	С								
G () ()	\bigcirc	С								
$\square \cap \cap$	\bigcirc	C								

4 Click (Preview) on the [Status] panel.



Previewing starts, and the acquisition status is displayed on the worksheet.



5 Adjust the instrument settings and worksheet settings, as required.

For details, see "Configuring Instrument Settings" (page 59) and "Customizing the Display" (page 63).

6 After completing adjustments, click **•**.

Tips

• You can sort wells by color for the metadata selected in [Color Coded by]. You can select [Sample Group], [Cell], or [Condition] for sorting by color.



• On models equipped with a 320 nm laser, wait about one minute after starting preview for the laser to warm up.

Configuring Instrument Settings

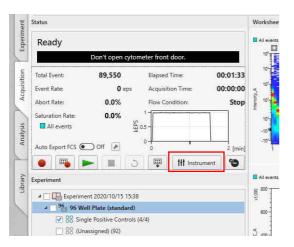
The following instrument settings can be configured on the ID7000.

- Lasers
- Detector
- Flow rate control
- Agitation
- Event viewer
- Cleaning

The instrument settings can be set for each sample group. Changing the settings for a sample in a sample group will change the settings for all unacquired samples belonging to the same sample group. If more than one sample group is configured, you can set different settings for each sample group.

You can also copy and paste the instrument settings of a sample group to apply the settings to another sample group.

1 Select a sample to acquire on the [Plate] panel, and click [Instrument].

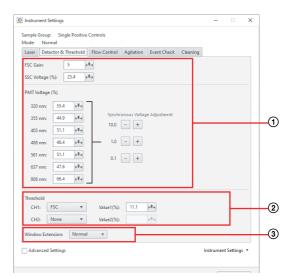


The [Instrument Settings] dialog appears.

2 Place a check mark in the checkbox for the laser(s) to use on the [Laser] tab.

lode:	Normal		i and			ii.	
Laser	Detector & Threshold	Flow Control	Agitation	Event Check	Cleaning		
Laser	On/Off:						
100	320mm						
1	355nm						
~	405nm						
100	488mm						
1	561nm						
1	637nm						
V	808nm						
Advi	anced Settings					Instrument Setting	s •

3 Set the detector gain and threshold levels on the [Detector & Threshold] tab.



(1) Set the forward scatter (FSC), side scatter (SSC), and fluorescence sensitivity.

Clicking displays a slider. Adjust the sensitivity using the slider or enter a value directly.

Increasing the sensitivity will increase the voltage, but the signal may become saturated, preventing correct data acquisition, if the value is too high. You can check for fluorescence signal saturation using [Saturation Rate] in the [Status] panel during acquisition (value is displayed in red when the signal is saturated).

- [FSC Gain]: Set the forward scatter (FSC) gain level.
- [SSC Voltage (%)] ([SSC Voltage (ST)] in Standardization mode): Set the side scatter (SSC) PMT voltage.
- [PMT Voltage (%)] ([PMT Voltage (ST)] in Standardization mode): Set in conjunction with the PMT voltages of each laser channel.

Tip

You can use [+] and [-] under [Synchronous Voltage Adjustment] to increase or decrease all channels at once. (The balance between all laser channels is maintained in Standardization mode.)

Note

In Standardization mode, the configurable PMT voltage range is displayed. Setting a value greater than 5.00 may prevent standardization between instruments.

- ② Set the trigger threshold channel and threshold value.
 - [Threshold] [CH1]: Select the channel for the threshold to trigger detection of events. The forward scatter (FSC) channel is commonly used as the trigger channel to isolate targeted populations from background

noise and cell debris, as it is less susceptible to the effects of background noise.

[Threshold] [Value1(%)]: Sets the threshold value as a percentage of the output signal range.

Clicking displays a slider. Set the value using the slider or enter a value directly.

- [Threshold] [CH2]: Select the fluorescence channel for the threshold to trigger detection of events.
- [Threshold] [Value2(%)]: Set the threshold value as a percentage of the receive signal output range.

Clicking displays a slider. Set the value using the slider or enter a value directly.

Tip

Data is acquired only for events that exceed the threshold value in the channel selected in [Threshold CH]. You can remove background noise and cell debris by raising the threshold, but you may not be able to acquire the desired data if the threshold is too high. In addition, a wider window extension setting is suitable for the higher threshold level.

③ Select the window extension. Select [Narrow] when acquiring small cells, or select [Wide] or [Extra Wide] when acquiring large cells.

Window Extension (WE)	Cell size
Narrow	Up to 3 µm
Normal	Up to 10 µm
Wide	Up to 20 µm
Extra Wide	Up to 40 µm

Тір

If the window extension is too narrow, CV may deteriorate. The above sizes are for reference only, and may vary slightly depending on the instrument settings.

4 Configure the flow rate on the [Flow Control] tab.



[Boost]: Set the boost time.

Increasing the boost time allows more events to flow faster at the start of acquisition.

[Sample Flow Rate] Set the sample flow rate. A lower sample flow rate provides more accurate acquisition. A higher sample flow rate may reduce the time required to accumulate a specific number of events.

[Acquisition Offset Time]: Set the offset time between the start of data acquisition of each well in units of seconds.

5 On the [Agitation] tab, configure the agitation-related settings.

	nent Settings							
Sample Mode:	Group: Single Positive Normal	Controls						
Laser	Detector & Threshold	Flow Control	Agitation	Event Check	Cleaning			
Pa Mi	ation	ontinuous						
Adva	inced Settings					Instrumer	it Setting	s •
							Close	

[Low Dead Volume]: To reduce the dead volume, place a check mark in [Enable low dead volume mode].

[Agitation]: Set agitation settings.

Place a check mark in [Enable sample agitation] to enable agitation, and select the agitation method.

[Pattern]: Select the agitation method.

- [Once]: Agitate only once before sample acquisition.
- [Cyclic]: Agitate repeatedly at regular intervals during sample acquisition.
- [Continuous]: Agitate continuously during sample acquisition.
- [Mode]: Select the agitation mode.
- [Normal]: Agitate normally.
- [Short]: Agitate gently

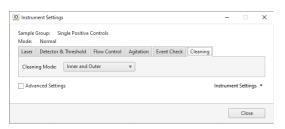
Тір

When [Low Dead Volume] is enabled, [Pattern] is set to [Once] (fixed). To use [Cyclic] or [Continuous], first disable [Low Dead Volume]. **6** On the [Event Check] tab, configure the settings related to event rate drop and air bubble detection.

aser			Litins in		The second second	
	Detector & Threshold	Flow Control	Agitation	Event Check	Cleaning	
Eve	nt Check:					
0	Off					
~	he system will not detect	air bubbles or lo	w event rate.			
	On					
If	the system detects air bu	bbles or low eve	nt rate data a	acquisition is pau	used. The syst	tem will then clean
it	self and recover automat	ically of stop acc	uisition base	d on the Auto A	cquire Prefer	ence Setting.
Aut	o Acquire Preferences:					
Set	Preference for the system	action when an	error is deter	cted during Auto	Acquire.	
0	Stop					
S	ystem will stop Auto Acq	uire.				
	Continue					
۲	ystem will continue Auto	Acquire.				
s	anced Settings					Instrument Settings

[Event Check]: Enable/disable detection of falling event rates and air bubbles.

- [Off]: Disable detection of falling event rates and air bubbles.
- [On]: Enable detection of falling event rates and air bubbles.
- [Auto Acquire Preferences]: Sets whether automatic acquisition stops or continues if the event rate drops or air bubbles are detected during automatic acquisition.
 - [Stop]: Stops automatic acquisition when the event rate drops or air bubbles are detected.
 - [Continue]: When the event rate drops or air bubbles are detected, cleaning is performed and automatic acquisition continues from the next sample.
- 7 On the [Cleaning] tab, configure the settings related to sample probe cleaning.



[Cleaning Mode]: Select the cleaning method used between acquisition of each well.

- [Inner and Outer]: Clean the interior and exterior of the sample probe by backflushing with sheath fluid.
- [Inner Only]: Clean the interior of the sample probe only by backflushing with sheath fluid.
- [No Wash]: Skip sample probe cleaning.



8 When finished configuration, click [Close].

Using Instrument Settings of Another Sample Group

You can copy and paste the instrument settings of a sample group to apply the settings to another sample group.

Click [Instrument Settings] in the bottom right of the [Instrument Settings] dialog, and select the desired target from the context menu.

60 ⊧ ♥⊣ mW	
	Instrument Settings 🔻
	Copy Instrument Settings Paste Instrument Settings
	Load Preference Settings Save as Preference Settings

[Copy Instrument Settings]: Copies the instrument settings.

[Paste Instrument Settings]: Pastes the copied instrument settings to the selected sample group.

[Load Preference Settings]: Loads the current preference settings as the instrument settings of the current sample group.

[Save as Preference Settings]: Saves the current instrument settings as preference settings.

Monitoring Instrument Status

The instrument status is displayed along the bottom of the window.

During ID7000 operation, you should constantly monitor the instrument status, check for any errors, and check for warnings related to each tank.



For details about hardware status icons, see "7 Hardware status "Hardware status" (page 21)" in "Operating window".

To check detailed instrument status

1

Click [Cytometer Status] in the [Status] group on the [Cytometer] tab of the ribbon.



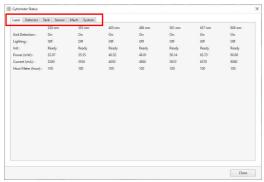
The [Cytometer Status] dialog appears.

2 Click the item to monitor from the tabs at the top.

Information can be displayed for the following items. The displayed information will vary depending on the model.

- Laser
- Detector
- Tank
- Sensor
- Mech (mechanism)

• System



Detailed instrument Information is displayed.

Example: [Sensor]

Notch Filter		Laser Shutter					Value					
561 nm (IN) :	Off	Shutter (OPEN) :	Off				1: 0ff	4-	Off	7.	Off	
561 nm (OUT) :	On	Shutter (CLOSE) :	On				2: Off		Off		Off	
637 nm (IN) :	Off						3: Off		Off			
637 nm (OUT) :	On	Optics Original Position										
808 nm (IN) :	Off	Y-Axis	On				Door					
808 nm (OUT) :	On	Z-Axis	Off				Interlock	0	H.			
Temperature		Pressure					Pump Duty					
Laser ("C) :	0.0	Pressure Tank (kPa) :	0.00	7	0.00	0	LP1 (%) :	0.1	5			
Fluidics (*C) :	0.0	Vecuum Tenk (kPa) :	0.00	2	0.00	0	LP2 (%) :	0.1	5			
Power Supply (*C) :	0.0	Sample (kPa) :	0.00	2	0.00	0	LP3 (%) :	0.1	5			
320 nm PMT (*C) :	32.0	Washing Unit (kPa) :	0.00				LP4 (%):	0.1	5			
355 nm PMT (*C) :	35.5	Notch Shutter Tank (kPa) 179.88	(0	II)		AP1 (%) :	0.1	3			
405 nm PMT (*C) :	40.5						AP2 (%):	0.1	5			
488 nm PMT (*C) :	48.8											
561 nm PMT (*C) :	56.1											
637 nm PMT (*C) :	63.7											
Sample Stage (*C) :	0.0	0										

For details about each item, see "[Cytometer Status] dialog" (page 124).

3 Click [Close] to close the dialog.

Chapter 5 Acquisition

Customizing the Display

You can change the type of plots, modify gates on plots, and customize the content displayed in the [Gates and Statistics] table displayed on the [Acquisition] workflow tab during data acquisition.

You can customize items using the tabs on the ribbon.

Example: [Worksheet Tools] tab

-	Well - A0	1							
卞			Ð,		R	E,	6		D
nap to Grid	Auto Arrange	Change Palette •	80 % •	Show Table	Edit	Export Statistics •	Gate Manager	Overlay Worksheet *	Print
lignmer	nt -	View	N	-	Statist	ics	Gates	Overlay	Print

- 1 In the [View] group, the view of plots can be changed and plots can be zoomed in/out.
- In the [Statistics] group, the [Gates and Statistics] table can be shown/hidden and the content of the statistics table can be customized.
- 3 In the [Gates] group, gates can be modified.
- In the [Overlay] group, spectral data of a selected plot can be overlaid on a plot within the overlay worksheet.

For details about each item, see "[Worksheet Tools] Tab (Ribbon)" (page 135).

Acquiring Data

Select a single well and acquire data for the well.

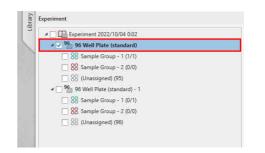
For details about acquiring multiple wells automatically, see "Running Auto Acquisition" (page 65).

For details about data acquisition for creating a spectral reference library, see "Registering a Spectral Reference" (page 43).

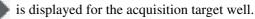
Before starting data acquisition

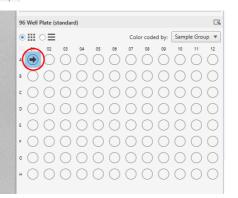
Check the following items.

- Check the fluid levels in each of the tanks
- 10L tank system is connected correctly to the external tank connection block
- **1** Select the plate to acquire on the [Experiment] panel on the [Acquisition] workflow tab.



2 Select the well/tube to acquire in the [Plate] panel, and click [Set Current Position] in the [Acquisition] tab of the ribbon.





3 Configure the acquisition stop condition and instrument settings.

Click [Stopping Condition] in the [Sample] group on the [Acquisition] tab of the ribbon to open the [Stopping Condition] dialog.

Place a check mark in the checkbox for the item you want to configure, then select an acquisition stop condition in the drop-down menu and select a value.

• You can specify a combination of [Total Events], [Acquisition Time], or [Elapsed Time] for the acquisition stop condition. You can also enter a value directly from the keyboard.

- You can also set gates, and set the number of events to stop acquisition for each gate.
- You can configure a combination logic stopping condition by selecting AND or OR.

 Total Events 		10,000	Ŧ	
Acquisition Ti	me v	00:02:00		
Gate Level Stoppin	ng Conditio	on ———		
📝 #1 📕 A		10,000		events
📝 #2 📕 B	×	10,000		events
[]#3 [Ť		Ŧ	events
aturation Gate				

For details about other settings, see "Configuring Instrument Settings" (page 59).

Tips

- The instrument settings can be set for each sample group. Changing the settings for a sample in a sample group will change the settings for all unacquired samples belonging to the same sample group. If more than one sample group is configured, you can set different settings for each sample group.
- You can also open the [Stopping Condition] dialog by clicking 🕤 in the [Status] panel.
- **4** Click (Acquire) on the [Status] panel.

tent	Status			
Experiment	Ready			
		Don't open cyto	meter front door.	
tion	Total Event:	89,550	Elapsed Time:	00:01:33
Acquisition	Event Rate:	0 eps	Acquisition Time:	00:00:00
Ac	Abort Rate:	0.0%	Flow Condition:	Stop
1	Saturation Rate:	0.0%	1-	7
Analysis	All events	LED C	0.5 -	
Ans	Auto Export FCS 💽	Off 🕑	0	2 (min
-	🕘 🖷 🕨	- III (3	111 Instrum	ment 😮
Library	Experiment			
E.	4 🔄 🛄 Experime	nt 2020/10/15 15:38	3	
5	4 96 96 We	ll Plate (standard)		
	Sing	le Positive Controls	(4/4)	

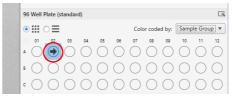
Data acquisition starts, and the acquisition status is displayed on the worksheet.

5 To change the event rate during acquisition, click [Instrument] and change [Sample Flow Rate] on the [Flow Control] tab of the [Instrument Settings] dialog.

Sample: Mode:	Well - A01 Normal						
Laser	Detector & Threshold	Flow Control	Agitation	Event Check	Cleaning		
Acqui	sition Offset Time: 0	😂 sec					
	inced Settings				In	strument S	Settings
Adva	niceu serungs						

6 When data acquisition is completed, select the next well/tube in the well list and repeat steps 2 to 5 to acquire data.

Select the next well/tube according to the configured acquisition sequence.



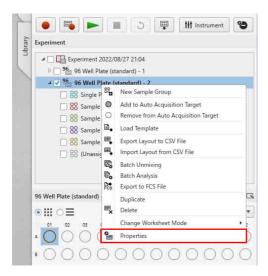
Running Auto Acquisition

You can automatically perform data acquisition using settings configured beforehand.

Configuring Acquisition Conditions for Plates

Set acquisition conditions for each plate.

1 Right-click the plate for auto acquisition on the [Experiment] panel, and select [Properties] from the context menu.



The plate properties dialog is displayed.

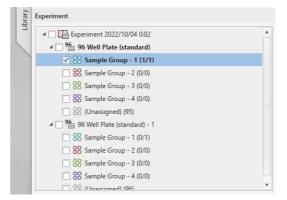
2 Set each item in [Acquisition Settings], then click [Close].

96 Well Plate (standard)	- 1 - Properties	-		×
Plate Information				-
Туре:	96 Well Plate (standard)			
Name:	96 Well Plate (standard) - 1			
Date Created:	10/15/2020 4:35:08 PM			
Number (optional):				
Memo (optional):			-	
ev (1998) is 199				
Acquisition Setting				
Order:) 🛃 Left-To-Right 🔿 🕅 Top-To-Botto	m		
				_
	ОК		Cancel	_

[Order]: Select the well acquisition sequence.

Starting Auto Acquisition

1 Select the plate to acquire on the [Experiment] panel.



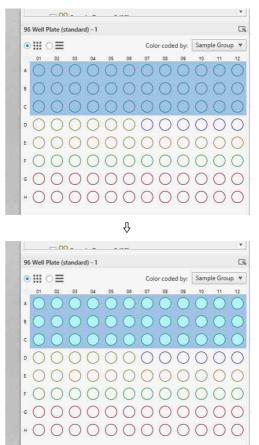
2 Configure the acquisition stop condition and instrument settings.

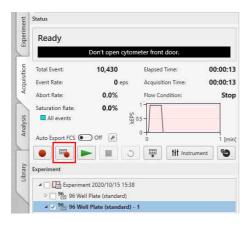
Set the acquisition stop condition and instrument settings for the target samples for auto acquisition.

For details, see step **3** in "Acquiring Data" (page 63).

3 Set the acquisition range.

Select the wells that you want to acquire automatically in the [Plate] panel, and click [Add to Target] in the [Auto Acquire] group on the [Acquisition] tab of the ribbon.

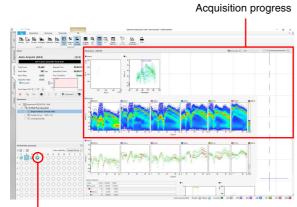




Automatic acquisitions starts.

Monitoring the Progress of Auto Acquisition

During auto acquisition, the settings and progress for the well currently being acquired are displayed.



Current well

To check the data for acquired wells

Click a well whose acquisition has been completed in the [Plate] panel to switch the display to the acquisition data for that well.

Closing an Experiment

After data acquisition has finished, close the experiment.

1 Click [Close] in the [Experiment] group on the [Acquisition] tab of the ribbon.



A confirmation message appears.

2 Click [OK].

The experiment is saved and closed, and the display automatically switches to the [Analysis] workflow tab.

Тір

The experiment is saved automatically when a new experiment is created.

Proceed to "Selecting a Sample for Analysis" (page 67).



Analysis

Selecting a Sample for Analysis

After data acquisition is finished, select the sample for analysis and analyze the acquired data on the [Analysis] workflow tab.

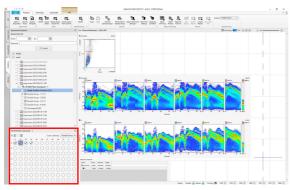
Selecting a Sample for Analysis

- **1** Open the experiment you want to analyze in the experiment list on the [Analysis] workflow tab, and select a plate.
- **2** Select a sample to analyze on the [Plate] panel.

The analysis data is displayed on the worksheet.

Select a plate, then select a well/tube.

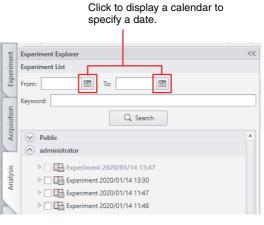
Data is displayed on the worksheet for each well/ tube.



Searching Experiments

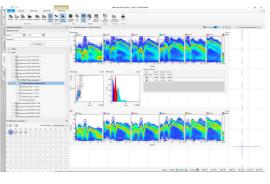
You can search experiments by date and keyword.

1 In [Experiment Explorer] on the [Analysis] workflow tab, specify dates and keywords, and then click [Search].



The experiments are searched using the specified criteria.

2 Click the desired experiment or analysis sample to display the data on the worksheet.



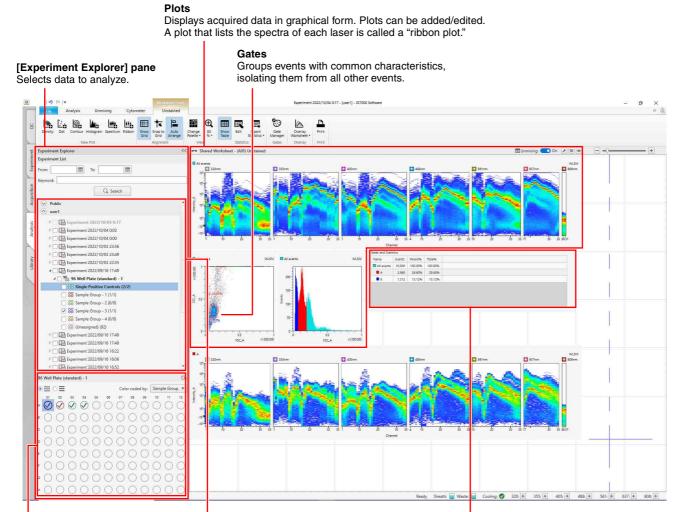
Tip

You cannot open the experiment that is currently open on the [Acquisition] tab. In this case, first close the experiment on the [Acquisition] tab.

For details about closing an experiment, see "Closing an Experiment" (page 66).

[Analysis] Tab Main Functions

You can perform analysis operations on all acquired data on the [Analysis] workflow tab.



[Plate] panel

Displays a list of wells/tubes. You can switch the display to list view or map view.

Experiment list

Displays data for completed experiments and samples for analysis in list view.

[Gates and Statistics] table

Displays statistics calculated automatically from acquired data. The statistics in the table can be customized.

To display data for analysis

Search for the target data in the [Experiment Explorer] on the left, and select the well to analyze.

For details about searching, see "Searching Experiments" (page 67).

Editing Plots

ID7000 Software displays the following types of plots. You can edit the plots, for example, to change the plot type or to change the display color.

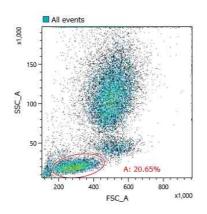
For details about adding gates to a plot, see "Editing Gates" (page 72).

Тір

You can select multiple plots on the worksheet using the Shift key or Ctrl key.

Density plots

Density plots display an event data distribution for two acquisition parameters (bivariate distribution). Density plots use color to show the distribution of the number of detected events having the same value. Highdensity regions are shown in red, and low-density regions are shown in dark blue. Density plots are most useful for displaying populations where the number of detected events is relatively large.

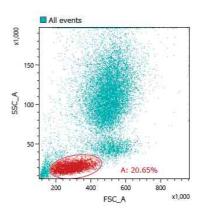


Dot plots

Dot plots display an event data distribution for two acquisition parameters (bivariate distribution). Dot plots are convenient for displaying data with less than 500K events. Events on a dot plot are colored based on their classification (gate and population).

Tips

- As the number of events increases, it may become difficult to discern patterns. In this case, reduce the number of events displayed or use a density plot to view the data.
- You can change the number of events to display using [Display Events] on the [Analysis] tab of the ribbon.

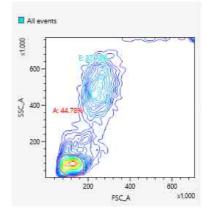


Contour plots

Contours plots display an event data distribution based on the sample data for two acquisition parameters (bivariate distribution).

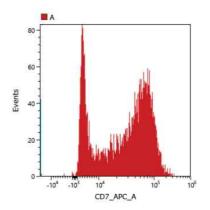
Contour plots show the distribution of event density by density contour lines, where each line connects regions of the same event density.

Contour plots are useful for distinguishing populations with a high disparity yet located in a close proximity. Such regions may be difficult to distinguish in a dot plot because they would appear bunched together.



Histogram plots

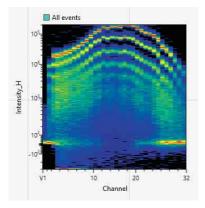
Histogram plots display an event data distribution for a single acquisition parameter (univariate distribution).



Spectrum plots

Spectrum plots display sample data (event data) in up to 35 channels for each laser.

The event signal intensity is displayed on the Y-axis, and the wavelength for events is displayed on the X-axis. The X-axis parameter can be switched to display "Wavelength" or "Channel". The gradation of colors indicates the density of events having the same intensity. Red indicates high density, and dark blue indicates low density. Spectrum plots are useful for identifying the characteristic spectral profile of samples and for detecting when PMT saturation occurs.



Ribbon plots

Ribbon plots display sample data (event data) for each channel for all lasers. The X-axis parameter is "Channel" (fixed).

10 ⁶ * 488nm		🗶 561nm	1		637nm	
105						
10				_		
102	<u>z</u> C.					
104						
10						

Adding a Plot

You can add plots to the worksheet.

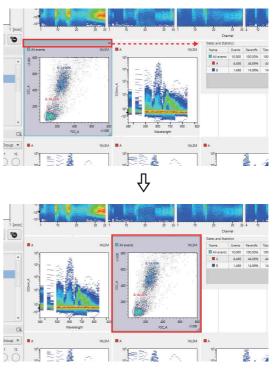
1 Click the type of plot to add in the [New Plot] group on the [Worksheet Tools] tab of the ribbon.

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-	File	A	cquisitior	n U	nmixing	Су	tometer	e	Well - A0
Sc		Ľ.	6	Å.	-0	Щь	Ħ	1=	
	Density	Dot	Contour	Histogram	Spectrum	Ribbon	Show Grid	Snap to Grid	Auto Arrange
1			Ne	ew Plat				Alignme	nt

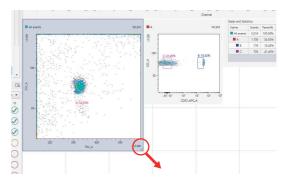
A plot is added to the worksheet. The new plot created represents the [All Events] population.

2 Change the layout of plots, as required.

You can change the position of plots by dragging the title part of target plot and dropping it to a new location.



You can also zoom in/out to change the display size of plots individually by dragging the bottom right of the plot.



Changing Axis Scales

Select a plot on the worksheet, then click the desired axis scale using the buttons in the [Scale] group on the [Plot Tools] tab of the ribbon.

You can also drag the axis on the plot to change the axis scale.



For details about the function of each button, see "[Scale] group" (page 140).

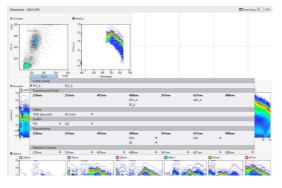
Changing Axis Parameters

Click the axis title and select the desired parameter from the context menu.

Parameters are grouped by [Scatter (Area)], [Fluorochrome (Area)], [Others], [Scatter], [Fluorochrome], and [Detection Channel], and are displayed for each excitation laser.

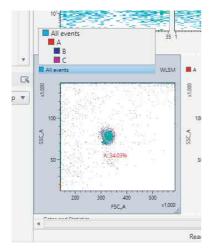
Example: Changing the X axis to FSC area data

Click the X-axis title and select [FSC_A] from the popup menu.



Changing the Source Gate of a Plot

Click the gate name on the top left of a plot and select the root gate from the drop-down list.

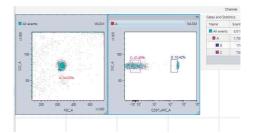


Changing Several Plots Simultaneously

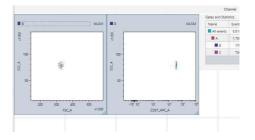
You can select multiple plots on the worksheet and apply the same settings to all plots at the same time. The settings that can applied to more than one plot at the same time include changes to gates and axis parameters (for example, FSC to FITC), changes to axis scales (for example, Max 1×10^4 to 1×10^5), and changes to axis scale type (for example, log to linear).

This section describes changes to the root gate as an example.

Select the target plots, and then select the root gate.



The root gate is changed on the selected plots at the same time.

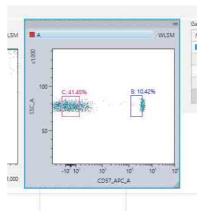


You can apply the same operation to multiple selected plots at the same time using the [Plot Type], [Scale], and [Copy Picture] group buttons on the [Plot Tools] tab of the ribbon.



Changing Plot Type

1 Select a plot whose type you want to change on the worksheet.



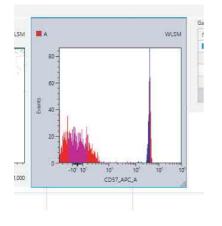
2 Click the target plot type in the [Plot Type] group on the [Plot Tools] tab of the ribbon.

The same operation is available by right-clicking a plot and selecting [Plot type].

For example, the diagram below shows a density plot being changed to a histogram plot.



The type of the selected plot is changed.



Duplicating a Plot

1 Select a plot to be duplicated on the worksheet, then click [Duplicate Plot] in the [Edit] group on the [Plot Tools] tab of the ribbon.

The same operation is available by right-clicking a plot and selecting [Duplicate].

ID	1 57 (24)	-				Work	
1	File	Acquisition	Unmixing	Cyt	ometer	We	ell - A01
S	Density 🙆 Contour		ar <u>li</u> . Le <u>i</u> te		X Axis:	Linear 🔹	
0	🔛 Dot	am	Duplicate Plot		Y Axis:	Linear	
1		Plot Type	Edit	- 60			

The selected plot is duplicated.

2 Edit the duplicated plot, as required, using the buttons on the [Plot Tools] tab of the ribbon.

For details about each button, see "[Plot Tools] Tab (Ribbon)" (page 138).

Removing a Plot

Select a plot to be removed on the worksheet, then click [Remove Plot] in the [Edit] group on the [Plot Tools] tab of the ribbon.

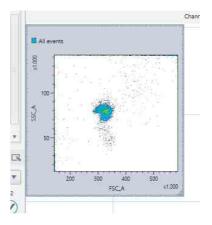
The same operation is available by right-clicking a plot and selecting [Remove].

Editing Gates

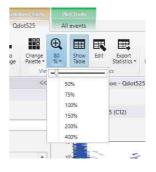
Adding Gates

You can separate events into groups by drawing gates around the events on plots. Gates of different types (Rectangle, Ellipse, Polygon, Quadrant, Linear) are available which can be drawn to match the distribution of events. Adding a gate allows you to display a new plot representing the population of events that fall within the gate.

Select a plot on which to add a gate.



Zoom in or out on the plot, as required, using controls in the [View] group on the [Worksheet Tools] tab of the ribbon to make it easier to draw the gate on the plot.



2 Click the button for desired gate type to surround the target population in the [New Gate] group on the [Plot Tools] tab of the ribbon, and draw a gate on the plot.

Alternatively, right-click the plot, then select [Insert gate] and the desired shape of gate from the context menu.

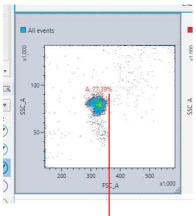
			[admin	istrator] -	ID7000 Soft	ware		
••			0		E	00	<u>لو</u>	[]
Default Adjust •	Spectrum Display *	Rectangle	Ellipse	Polygon	Quadrant	Linear	Sync Scale and Gate	Copy Picture
	View	New Gate					Too	16

[Rectangle]: Adds a rectangle gate. [Ellipse]: Adds an ellipse gate. [Polygon]: Adds a polygon gate.

- [Quadrant]: Adds a quadrant gate. A quadrant gate is added to a plot by clicking the mouse at the intersection point of the four quadrants. Gates "<gate_name>-Q1" to "<gate_name>-Q4" are assigned to the four quadrants on the plot. Drag the intersection point to change the size of the quadrants.
- [Linear]: Adds a linear gate. Linear gates can be added to histogram plots only.

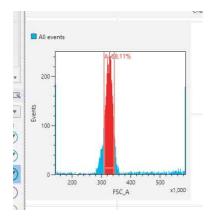
The gate is added to the plot and the population percentage statistic for the gate is displayed.

Example: Polygon gate



Statistic

Example: Linear gate



3 Edit the name of the gate, the displayed statistic, color, and other preferences, as required.

Select the gate, click [Gate Manager] or $\overline{\Gamma}$ in the [Edit] group on the [Gate Tools] tab of the ribbon to display a dialog where you can change settings.



[Gate Manager]: Displays the [Gate Manager] dialog.

For details about each item, see "[Gate Manager] dialog" (page 137).

F: Displays the [Gate Properties] dialog. You can perform the same operation from the right-click context menu.

For details about each item, see "[Gate Properties] dialog" (page 137).

Editing Gates

You can change the shape of and settings for gates on plots.

To change the shape of a gate

Select the gate and drag the drawing handles to change the shape.

For quadrant gates, drag the intersection point of the quadrants to another position to change the gate.

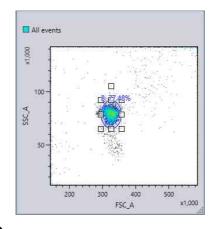
To move a gate

Select the gate and drag the gate to the desired position.

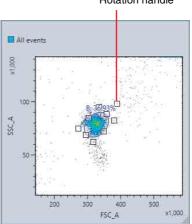
To rotate a gate

You can rotate polygon and ellipse gates.

Select a polygon gate or ellipse gate.



2 Move the rotation handle to rotate the gate in any direction.



Rotation handle

To change gate color and statistics information

Select a gate, then select the desired color in the [Color] group on the [Gate Tools] tab of the ribbon. You can change the font size and the statistic that is displayed for a gate on a plot in the [Label] group.



To change gate settings

Select the gate, click [Gate Manager] or $\boxed{1}$ in the [Edit] group on the [Gate Tools] tab of the ribbon to display the [Gate Manager] dialog or [Gate Properties] dialog, respectively, and then change the settings.



For details about each item, see "[Gate Manager] dialog" (page 137) or "[Gate Properties] dialog" (page 137).

To delete a gate

Select the gate to be deleted, then click [Remove Gate] in the [Edit] group on the [Gate Tools] tab of the ribbon. Alternatively, right-click the gate and select [Remove] from the context menu.

Note

Deleting a gate will delete all data and statistics for the gate, including all child gates and plots derived from the gate.

To move a gate

Right-click the gate you want to move in the statistics table, and select [Cut] from the context menu. Right-click the move destination gate, and select [Paste] from the context menu.

To duplicate a gate

Right-click the gate you want to duplicate in the statistics table, and select [Copy] from the context menu. Right-click the duplicate destination gate, and select [Paste] from the context menu.

Note

Gates cannot be moved to another worksheet. When duplicating gates on other worksheets, some gates may not be duplicated due to the difference of fluorochromes in the color panel.

Customizing the Statistics Table

You can change the statistics items displayed in the statistics table, and export the statistics table as a CSV file.

Editing Items Displayed in the Statistics Table

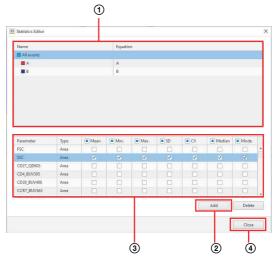
1 Click [Edit] in the [Statistics] group on the [Worksheet Tools] tab of the ribbon.

Alternatively, right-click the [Gates and Statistics] table and select [Open Statistics Editor] from the context menu.



The [Statistics Editor] dialog appears.

2 Edit the items displayed in the [Gates and Statistics] table.



① Select a gate.

The display parameters and statistics values for the selected gate are displayed in the lower area.

- Click [Add] to add statistics for fluorochrome parameters and pulse parameters.
 A new parameter configuration item is added to the bottom with each click. Select [Parameter] and [Type] to select the parameters to display.
- ③ Place a check mark in the checkboxes for the statistics you want to display for each fluorochrome and type parameter.

④ Click [Close] to close the dialog.

Тір

To delete a pulse parameter, select the pulse parameter row and click [Delete].

To copy item settings displayed in the statistics table to another gate

- **1** Select the copy source gate, then right-click and select [Copy] from the context menu.
- **2** Select the copy destination gate, then right-click and select [Paste] from the context menu.

To change the width of columns in the statistics table.

Drag the right edge of a table column to change the width of the column.

Exporting Statistics Table Information as a CSV File

You can export the statistics table as a CSV file. You can also export statistics information for each sample, or for multiple samples.

To export each sample

1 Click [Export Statistics] – [Sample] in the [Statistics] group on the [Worksheet Tools] tab of the ribbon.



The [Export to CSV File] dialog appears.

2 Click (Browse) to specify the folder to save the CSV file, then click [Export].

D Export Statistics	
Output Path	700
	0% Export
	Close

Information for the statistics table displayed on the worksheet is saved in a CSV file.

3 Click [Close] to close the dialog.

Sample CSV file export

	A	в	C	D	E	F	G	н	I	J	K	
1	Name	Events	Parent[%]	Total[%]	Mean	Min.	Max.	SD	CV	Median	Mode	
2	All events	10000	100.00%	100.00%								
3	A	1675	16.75%	16.75%								
4	в	259	2.59%	2.59%								
5												
6												

To export statistics information for multiple samples

You can export statistics information for a sample in the statistics table displayed on the worksheet, together with the completed samples within the same sample group.

 Select the row with the statistics information you want to export in the statistics table, and click [Export Statistics] – [Sample Group] in the [Statistics] group on the [Worksheet Tools] tab of the ribbon.

Alternatively, right-click the row in the statistics table, and select [Export to CSV File] – [Export Group Statistics] from the context menu.



The [Export to CSV File] dialog appears.

2 Click (Browse) to specify the folder to save the CSV file, then click [Export].

All events A A A B B Parameter Type B Man. B Man. B SD B CV B Median B Mode FSC Area V V V V V V V V V V CO CO COSC Area P <th></th> <th></th> <th></th> <th>E</th> <th>quation</th> <th></th> <th></th> <th></th> <th></th> <th></th>				E	quation					
Parameter Type O Man O Max O SD O V O Median O Mode SSC Area V	Alf events									
Parameter Type B Man	🖌 📕 A			:4	í.					
Area V	🖌 🔳 B			B	8					
Area V										
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Area V										
SSC Area V <th>Parameter</th> <th>Type</th> <th>Mean</th> <th>Min.</th> <th>Max.</th> <th>I SD</th> <th>CV.</th> <th>Median</th> <th>Mode</th> <th></th>	Parameter	Type	Mean	Min.	Max.	I SD	CV.	Median	Mode	
C027_08665 Area	FSC	1.1120.00		A STATE OF A	1	~	~	~	×.	
CD4.80/395 Area Image: Constraint of the second se	SSC	Area		1		1	V	V	v	
CD38_BUV496 Area	CD27_QD605	Area								
CCR7_BUV563 Area	CD4_BUV395	Area								
	CD38_BUV496	Area								
		Area								
	CCR7_BUV563			-						17
		1.1								
Output Path C:\Users\fcm\Desktop\0114_bmp\SpectralReference_20200114102121\20200110.csv Browse		sers\fcm\Desl		\SpectralRefe			0110.csv	1	Browse	
Dutput Path C:\Users\fcm\Desktop\0114_bmp\SpectralReference_20200114102121\20200110.csv Browse		sers\fcm\Desi		\SpectralRefe			0110.csv		Browse.	
		sers\fcm\Desl		\SpectralRefe			10110.csv			
Uuguut Path C:USeen/tcm/Deatkop10114_bmpiSpectralReference_2020011410212120200110.csv Browse 0% Export		sers\fcm\Desi		\SpectralRefe			10110.csv			

The statistics information for the samples within the same sample group are saved to a CSV file with the same gate name as the selected row.

3 Click [Close] to close the dialog.

Sample CSV file export

The statistics information for the samples within the same sample group are saved.



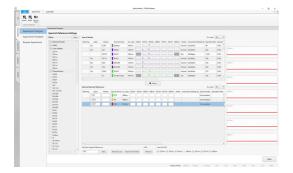
Setting Up Spectral Unmixing

Create a spectral reference and set up spectral unmixing.

Acquiring Single Stain Sample Data

This section describes creating an experiment to acquire the following fluorochromes and acquire single stain sample data.

- FITC
- PE
- APC
- Add the fluorochromes above using the [Experiment Designer] on the [Experiment] workflow tab.



- **2** Create an experiment using the wizard.
- **3** Acquire single stain sample data.

Creating a Spectral Reference

A spectral reference provides information about the shape of the fluorescence spectrum, which is used for spectral unmixing. A spectral reference can be created from acquisition data of a single stain control.

1 Prepare a single stain control using the target fluorochrome.

Notes

- A Positive control (PC) and a Negative control (NC) acquired under the same acquisition conditions are required.
- The Positive and Negative controls differ only in the presence/absence of staining. Basically, the same type of particles must be used. For example, if a single stain beads control is prepared as the Positive control, prepare the same type of beads, unstained, for the Negative control.

2 Set Positive and Negative controls for each fluorochrome.

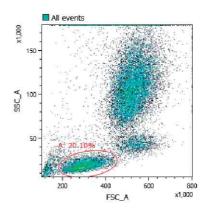
Set gates corresponding to the Positive control and Negative control populations, for the prepared single stain controls and unstained control, and assign to the fluorochromes in the [Unmixing Settings] dialog.

Example:

(1) Add a gate to the density plot for the FITC single stain control.

Display the FITC single stain control, click [Ellipse] in the [New Gate] group on the [Plot Tools] tab of the ribbon, and add the gate to the FSC-SSC density plot.

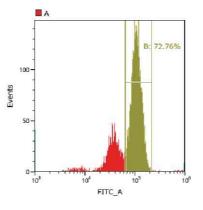
You can adjust the axis scale or zoom the plot, as required, if the gate is difficult to draw.



② Add a histogram plot and add a gate to the FITC positive population.

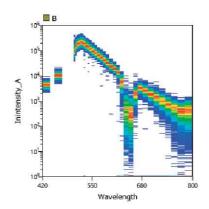
Right-click the gate added in step ①, and select [Create a Child Plot] – [Histogram Plot] to add a histogram, and change the horizontal axis to FITC-A.

Select the histogram plot, click [Linear] in the [New Gate] group on the [Plot Tools] tab of the ribbon, and add a linear gate to the FITC negative population.



Тір

You can display a spectrum chart by rightclicking the gate for the FITC positive population and selecting [Create a Child Plot] – [Spectrum Plot] to check that the gate is set appropriately.



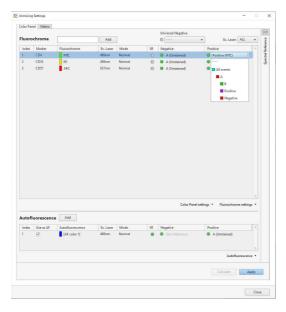
③ Click [Unmixing Settings] in the [Unmixing] group on the [Unmixing] tab of the ribbon.



The [Unmixing Settings] dialog appears.

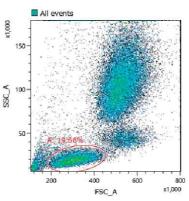
Assign the configured gate to the FITC Positive control.

Click [Positive] for FITC in the fluorochrome list, and select the gate set for the FITC positive population from the displayed list.

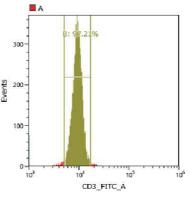


 Set the gate for the unstained control corresponding to the FITC positive population above.

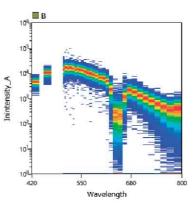
Repeat steps ① to ③ for the unstained control. First, set a gate for the data for the unstained control.



Next, add a histogram plot.



Display a spectrum plot to check that the gate is set appropriately.



Tip

The populations specified for Positive and Negative must basically be the same type of cells. Accordingly, on the FSC-SSC plot, the gate should be set in roughly the same position as the gate for the FITC positive population. If not set correctly, the correct spectral reference will not be obtained due to the effects of autofluorescence of cells.

(6) Assign the configured gate to the FITC Negative control.

	el Matrix							
	chrome	· · · · · · · · · · · · · · · · · · ·	Add			Universal Negative	• Ex. Laser. ALL	Ŧ
Index	Marker	Huorochrome	Ex. Laser	Mode	SR.	Negative	Positive	
1	CD4	FITC	438nm	Normal	0	Negative (FITC)	Positive (FITC)	
2	CD33 CD57	PE APC	438nm 637nm	Normal	0	• · · ·	 Positive (PE) Positive (APC) 	
					0	 Universit Negative Zare Defense.ce All events A Bistive B 		
							ettings * Fluorochrome set	
Autofi	uorescence	Add				Color Panel s	ettings • Pluorochrome set	tings *
Autofl	uorescence Use as AF	Add	Ex. Laser	Mode	SR	Color Panel s	Positive	tings •
			Ex. Laser 488nm	Mode Normal	SR •		-	×
Index	Use as AF	Autofluorescence				Negative	Positive	X

⑦ Assign the gates for Positive and Negative controls for all fluorochromes in the same way.

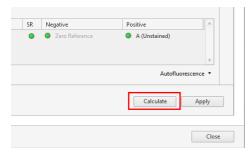
						Universal Negative		
Fluor	ochrome	[Add				w. Ex. Lasen AL	L
Index	Marker	Fluorochrome	Ex. Laser	Mode	SR	Negative	Positive	
1	CD4	FITC	438nm	Normal	0	Negative (FITC)	Positive (FITC)	
2	CD33	PE	438nm	Normal	0	Negative (PE)	Positive (PE)	
3	CD57	APC	637nm	Normal	0	Negative (APC)	Positive (APC)	
Auto	fluorescence	Add				Color Panel	settings • Plusrochrome se	etting
Auto		Add	Ex. Laser	Mode	SR	Color Panel Negative	settings • Fluoreschrome so Positive	etting
			Ex. Laser 488nm	Mode Normal	SR.			etting
Index	Use as AF	Autofluorescence				Negative	Positive	

Тір

When assigning an unstained sample that is common to multiple fluorochromes, you can assign a gate to Universal Negative and then set the Negative of each fluorochrome to Universal Negative to eliminate the need for individual Negative gate settings.

3 Click [Calculate].

The spectral unmixing is calculated.



If a data saturated warning appears

When calculating a spectral reference, the percentage of event data that was saturated at the time of acquisition in the gates set to Positive Gate and Negative Gate is evaluated, and a warning is displayed if it is determined that the quality will be affected. If you want to exclude saturated event data from the spectral reference calculation, place a check mark in [Calculate Spectral Reference without saturated event(s)], and click [Continue].

If a caution mark appears in the [Unmixing] panel

The quality of each spectral reference is evaluated when it is calculated. If a spectral reference conflicts with the following conditions, the quality is deemed to be bad, and \triangle appears for the corresponding fluorochrome in the [Unmixing] panel.

Color Pa	nel Matrix					
Fluoro	ochrome	1	Add			Universal Negativ
Index	Marker	Fluorochrome	Ex. Laser	Mode	SR	Negative
1		FITC	488nm	Normal		A (FITC)
2		PE	488nm	Normal	4	A (FITC)
3		APC	637nm	Normal	1	A (FITC)

The reason can be displayed by hovering the mouse over the \bigwedge icon.

• Spectral index error

A appears if the spectral index value derived from the Area/Height parameters of the data used to calculate the spectral reference is less than 5.

	Universal Negative		
	0 ··· v	Ex. Laser: ALL	٣
SR	Negative	Positive	- A.
A	Negative (FITC)	Positive (FITC)	
	Negative (PE)	Positive (PE)	
	Negative (APC)	Positive (APC)	

Negative value error

 \triangle appears if the negative value in the calculated spectral reference is higher than 10% of the maximum positive value.

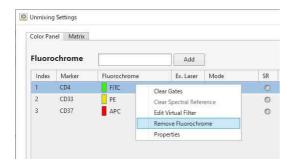
C V Ex. Laser: ALL	-
	٣
SR Negative Positive	· A.'
🛕 🕒 Negative (FITC) 🕒 Positive (FITC)	
🔥 🕒 Negative (PE)	
A Separative (APC) Positive (APC)	

If a conflict occurs, the following types of waveforms are displayed.

	Spectral Reference	<<
Ex. Laser: ALL	488nm	
ive	9 / / I	
A (Well - A01)	marth	Ann
3 (Well - A01)	V	
	561nm	1
	\sim	
		m
	637nm	1.1
		the
		V V

To remove a fluorochrome from the list

1 Right-click the target cell, and select [Remove Fluorochrome].



A confirmation message appears.

2 Click [Yes].

The fluorochrome is removed from the list.

Registering a Spectral Reference in the Library

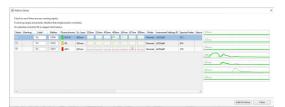
You can register a calculated spectral reference in the spectral reference library.

1 Click [Fluorochrome settings] in the bottom right of the [Unmixing Settings] dialog, and select [Add to Library] from the context menu.

	Color Panel set	tings • Flu	vorachrome settings 🔻
			Load from Library Add to Library
SR	Negative	Positive	A
0	Zero Reference	A (Unst	ained)
			Autofluorescence *
		Calc	ulate Apply

The [Add to Library] dialog appears.

2 Place a check mark for the spectral reference to register in the library, enter required metadata, and click [Add to Library].



The spectral reference is added to the library.

Fluorescence Compensation (Unmixing)

Fluorescence compensation is used to remove fluorescence spillover components from each fluorescence channel to obtain the signal for the target fluorochrome only for each channel.

Loading an Existing Spectral Reference

You can load a spectral reference from the spectral reference library.

- Select a sample group in the [Experiment] panel.
- **2** Click [Load Spectral Reference] on the [Unmixing] tab of the ribbon.



The [Load Spectral Reference] dialog appears.

3 Specify and load a spectral reference.



- Specify the spectral reference search criteria. The search results are displayed in [Search Results].
- ② Select the spectral reference to load. The waveform of the selected spectral reference is displayed on the right. Multiple selections are supported.

Тір

A spectral reference with a caution icon (1) displayed in the [Warning] field may have a problem with quality. Select a spectral reference that does not display the caution icon.

- ③ Click [Select].
 The selected spectral reference is moved to [Selected Spectral References].
- (4) Repeat steps (2) and (3) to move all spectral references to be loaded.
- Click [Load]. The spectral references are loaded into the experiment.

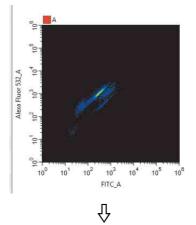
For details about each item, see "[Load Spectral Reference] dialog" (page 134).

Applying Spectral Unmixing

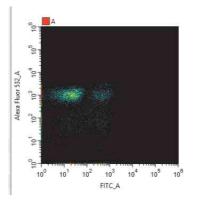
Click [Apply] on the [Unmixing] tab of the ribbon to perform spectral unmixing.



Before unmixing

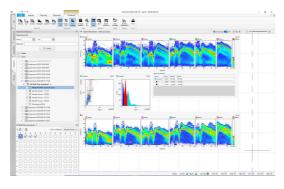


After unmixing



Analyzing data

Use the various tools to analyze the data.



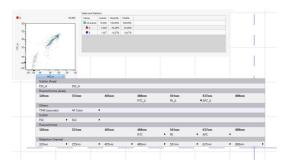
Adjusting Spectral Unmixing Manually (Spectrum)

You can adjust spectral unmixing graphically on the worksheet using a mouse if you want to adjust the spectral unmixing values obtained using the spectral reference.

Тір

Changing the spectral unmixing on a plot will also change the unmixing on other plots having the same fluorochrome as one of its parameters, changing the position of events. Accordingly, you should add plots for the required spectral parameters to the worksheet before adjusting spectral unmixing manually in order to monitor the affect of adjusting the unmixing on each plot.

- **1** Display the worksheet for the well whose spectral unmixing you want to adjust.
- **2** Create a density plot or dot plot, and specify fluorochromes for both the X axis and Y axis.



3 Click [Spectral Reference Adjuster] in the [Spectral Reference] group on the [Unmixing] tab of the ribbon.



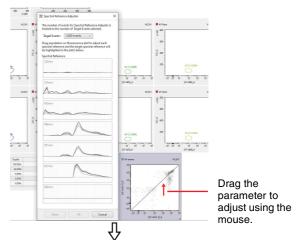
The [Spectral Reference Adjuster] dialog appears. The waveforms of the spectral reference for each fluorochrome are displayed in the dialog.

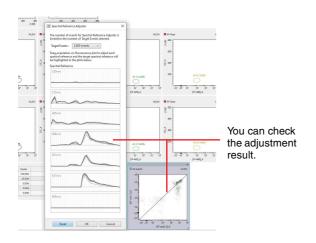
Spectral Refere	nce Adjuster	
	vents for Spectral Ref Imber of Target Event	
Target Events :	5,000 events	T .
pectral reference	on fluorescence plot e and the target spec 1 the plots below.	
pectral Referen	ce	
320nm		
355nm		
Am		
405nm		
\sim		
488nm	$ \land $	-
561nm	~	
637nm		
808nm		

A diagonal line is displayed on the plot.

4 Place the mouse cursor over a plot, and drag the plot to change the unmixing for the target parameter.

The mouse cursor changes to a horizontal doubleended arrow cursor in the left half of the plot, and a vertical cursor in the right half of the plot. Dragging the mouse on the plot that you want to adjust will modify the spectral reference, changing the calculated results. The target spectrum to modify is displayed highlighted in the spectrum display area of the dialog.





5 When unmixing finishes, click [OK] in the [Spectral Reference Adjuster] dialog.

The [Save unmixing matrix] dialog appears.

6 Select a matrix to overwrite in [Matrix List] or enter a matrix name in [Name] to save a new matrix, and click [Save].

Тір

The adjustment result can be saved as a new spectral unmixing matrix or can overwrite an existing spectral unmixing matrix. You can reload the pre-adjustment spectral unmixing matrix using [Load Spectral Reference] on the [Unmixing] tab of the ribbon.

Checking Spectral Unmixing Results Summary

You can check the results of spectral unmixing by displaying a list of plots for each fluorochrome combination.

You can also make manual adjustments to spectral unmixing as required.

- **1** Display the worksheet for the well/tube whose spectral unmixing result you want to check.
- **2** Click [Unmixing Viewer] in the [Spectral Reference] group on the [Unmixing] tab of the ribbon.



The Unmixing Viewer launches.

3 Select a fluorochrome for the X axis in [X Axis].

You can also select the previous or next fluorochrome using [Prev] and [Next], respectively.

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	10mm	
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	Antike Norkalee Select picts and the abbed to the Description	is before
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		Ce

- **4** Adjust the spectral unmixing as required.
 - ① Set [Adjuster] on at the top of the screen. A diagonal line appears on plots that have fluorescence parameters on the X and Y axes.
 - ② Place the mouse cursor over a plot, and drag the plot to change the unmixing for the target parameter.

The mouse cursor changes to a horizontal doubleended arrow cursor in the left half of the plot, and a vertical cursor in the right half of the plot. Dragging the mouse on the plot that you want to adjust will modify the spectral reference, changing the calculated results. The target spectrum to modify is displayed highlighted in the spectrum display area.

5 To add the plot to the worksheet, select the plot and click [Add to Worksheet].

A confirmation dialog appears. Clicking [OK] closes the Unmixing Viewer and then adds the plot to the worksheet.

6 Click [Close] to exit the Unmixing Viewer.

Tip

Changing the spectral unmixing on a plot will also change the unmixing on other plots with the same fluorochrome as one of its parameters, changing the position of events. The Unmixing Viewer displays plots of all fluorochrome combinations, so check the plot display for the corrected fluorochrome axis.

Finding Autofluorescence Populations

You can find the autofluorescence spectrum in acquisition data by searching for the autofluorescence populations.

Tip

The [Autofluorescence] tab of the [Unmixing Settings] dialog and the worksheet are updated after using the [Autofluorescence Finder].

1 Display the worksheet for the well/tube whose autofluorescence populations you want to find.

2 Click [Autofluorescence Finder] in the [Autofluorescence] group on the [Unmixing] tab of the ribbon.

Тою	k Pi	of Tools			Experiment 2020/10/15
1	AI	l events			
	0			P	8
ence	-	Load Spectral Reference	Add to Library	Autofluorescence Finder	Fluorochrome Database
5	Spectral Ref	erence		Autofluorescence	Fluorochrome

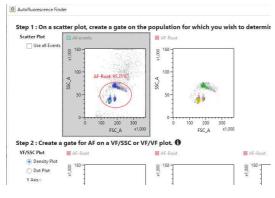
A message is displayed prompting for confirmation that the corresponding well/tube is selected.

3 Check the selected well/tube, then click [OK].

	Confirm	ation		>
	ected the unstained control. not selected, cancel and select the	unstained control then star	Autofluorescence	
in the unstained control is	not selected, cancer and select the	unstanieu control, men star	Hatomaorescenti	e rinder again.

The [Autofluorescence Finder] dialog appears.

4 In [Step 1], add the appropriate gates to the FSC/SSC plot.



Tips

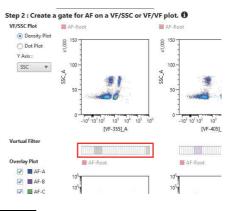
- Gate the corresponding populations to make later adjustment easier.
- You can adjust the axis settings on a plot (maximum value, minimum value, Biexponential axis parameter) by dragging the axis on the plot.
- To select all events without adding a new gate, place a check mark in [Use all Events].

The added gate automatically is reflected as the parent gate in [Step 2].

5 In [Step 2], search for the autofluorescence population while changing the virtual filter ranges.

You can change the wavelength band of the virtual filters to any range using the mouse.

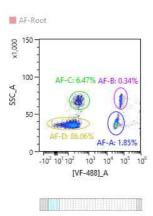
If you want to change the Y-axis of the plot to the wavelength range selected by the virtual filter, select the virtual filter of the desired laser axis from [Y Axis].



Тір

You can adjust the axis settings on a plot (maximum value, minimum value, Biexponential axis parameter) by dragging the axis on the plot. The axis type is fixed to the type set before displaying the [Autofluorescence Finder] dialog.

6 Add a gate for the population to register as the autofluorescence population.



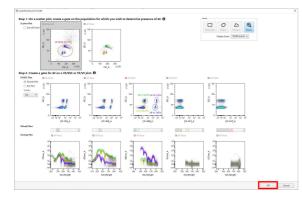
Tip

Up to eight autofluorescence components can be registered as fluorochromes named "AF color 1" up to "AF color 8."

7 Click [OK].

Note

The gate created in the [Autofluorescence Finder] dialog and its corresponding plot are added to the worksheet. Gates created in the [Autofluorescence Finder] are also updated.



A confirmation message appears.

8 Click [OK].

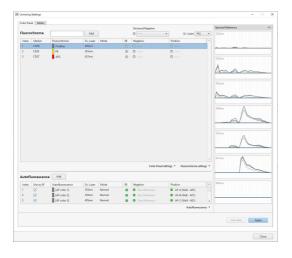
A reconfirmation message appears.

9 Click [Apply].

The worksheet is updated.

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10 Check the calculated spectral reference in the [Unmixing Settings] dialog.



Overlaying Multiple Spectral Data

You can overlay spectral data of multiple plots on a single plot to compare analysis results.

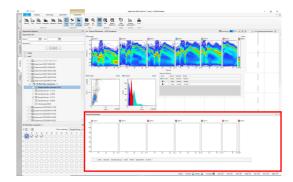
1 Click [Overlay Worksheet] in the [Overlay] group on the [Worksheet] tab of the ribbon and select the worksheet display method for overlays.

E	xperiment 2022	/02/03 9:54 - [administrat	or] - ID7000
۳		<u> </u>	
Gate Manager	Overlay Worksheet *	Print	
Gates	Half Wo	rksheet View	7
Sample Gro	Full Wo	ksheet View	
	Hide Ov	erlay Worksheet	
	Gate Manager Gates	Gate Gates Gates Gates Gates Gates Gates Gates Half Wo Full Work	Gate Worksheet + Print Gates Half Worksheet View

[Half Worksheet View]: Displays the overlay worksheet in the bottom half of the worksheet.[Full Worksheet View]: Displays the overlay worksheet in the full area of the worksheet.

The overlay worksheet is displayed on the worksheet.

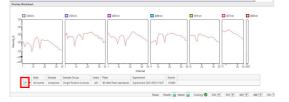
Display example: When [Half Worksheet View] is selected



2 Right-click a spectrum or ribbon plot with the spectral data to be overlaid, and select [Send to Overlay] from the context menu.

The spectral data is displayed in the plot of the overlay worksheet.

You can show/hide the spectral data by turning the checkbox in the legend on/off.





The overlay plot is not saved as a template.

To delete spectral data

- **1** Right-click a row in the legend of the overlay plot and select [Remove].
- **2** Right-click within the overlay plot, and select [Remove All] from the context menu.

To change the spectral data type

Right-click within the overlay plot, and select [Change Data Type] - [Mean] (average), [SD] (standard deviation), or [CV] (coefficient of variance) from the context menu.

To export the overlay plot data as a CSV file

Right-click within the overlay plot, and select [Export Data to CSV File] from the context menu.

To copy the overlay plot and legend to the clipboard

1 Copy the overlay plot to the clipboard in the desired format.

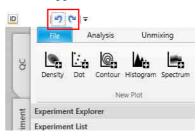
Right-click within the overlay plot, and select [Copy to Clipboard] - [Overlay Plot] or [Overlay Plot with Legend] from the context menu.

2 Paste into another application.

You can paste the image into an external application, such as PowerPoint or Word.

Undo/Redo Function

Undo/redo buttons are located in the title bar of this software. Worksheet actions including those listed below, support the undo/redo function.



- Creating plots or gates
- Deleting plots or gates
- Editing plots or gates
- Resizing/moving worksheet items
- Editing the Gate and Statistics table
- Adjusting the Spectral Reference
- Editing the Virtual Filter

Actions, such as those listed below, do not support the undo/redo function.

- Sync scale and gate
- Change palette
- Changes to the spectrum display
- Edit color
- Changes to the worksheet mode
- Copying and pasting worksheets
- Unmixing apply on/off
- When displaying dialogs, such as Plot/Gate Properties/ Gate Manager/Statistics Editor
- Changes to the zoom factor of the worksheet
- Changes to displayed events
- Adding plots from the Unmixing Viewer or Autofluorescence Finder



Advanced Operations

For details about each item, see "[Preference Settings] dialog" (page 111).

To import/export user preference settings

You can import the preference settings of another user and export your own preference settings.

Click [Preference Settings] in the bottom right of the [Preference Settings] dialog, and select [Import] or [Export] from the context menu.

	Preference Settings		
	Import		
ОК	Export		

Configuring User Preference Settings

Various default function settings can be saved by each user in user preferences. User preferences are loaded automatically, eliminating the need for some configuration settings when you want to perform acquisition, analysis, or sorting. You can also set an audible notification received when data acquisition, QC, or cleaning is completed.

1 Click [Information] on the [File] tab of the ribbon, then click [Preference Settings].

Print	Preference Settings
Print	Change software default settings for each user.
Database	
11.1.	Change Password
Help	Change your current password.
Log	
∩ _ Logout	Re Institution Information
🖾 Exit	Show your institution information (Only administrator can edit).

The [Preference Settings] dialog appears.

2 Set each item.

The values set here become the default values used for each function.

Cytometer Settings	Define cytometer control default settings.	Reset
Experiment Instrument Settings Uxxisheet Data Management Printer Norficulton Account Settings	Service Temperature Control Service Temperature Control Plate Cooling © On _ Off	
		Preference Setting

Calibrating the Instrument

ID7000 Software features the following calibration functions.

- Flow Rate Calibration Adjusts the fluidics system pressure to achieve the optimum sheath fluid flow rate.
- Optics Alignment: Automatically aligns the laser excitation position for the flow cell.

Calibrating the Fluidics System

Flow Rate Calibration uses Align Check beads.

Tips

- Store Align Check beads in a cool location, and use them immediately after preparation.
- Shake the Align Check beads bottle vigorously before use to obtain a uniform bead concentration. Place two drops of Align Check beads in a sample tube and mix with 450 µl of DI water or PBS (saline) solution.
- 1 Click [Flow Rate] in the [Calibration] group on the [Cytometer] tab of the ribbon.

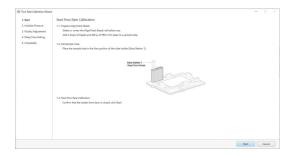
cquisition	Unmixing) Cyto	ometer					
Ц.	$\langle x \rangle$	Q.,	*	°:	S.	l.	ن.	24
Flow Celi Purge	Decontamination	Sheath Line	Optics Alignment	Flow Rate	Agitate	Cooling	Refill and Empty	Long Term Storage
Cleaning		De-bubble	Calibra	tion	Pi	ate	Tank Ma	intenance

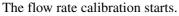
Note

[Flow Rate] is enabled only when no plate is loaded.

The [Flow Rate Calibration Wizard] window appears.

2 Follow the on-screen instructions to load a sample tube containing Align Check beads in the "Extra Station 1" position, and click [Start].





The flow rate calibration progress is displayed in the wizard.

Adjusting the Optical Axis

Optics alignment uses Align Check beads.

Tips

- Store Align Check beads in a cool location, and use them immediately after preparation.
- Shake the Align Check beads bottle vigorously before use to obtain a uniform bead concentration. Place two drops of Align Check beads in a sample tube and mix with 450 µl of DI water or PBS (saline) solution.
- 1 Click [Optics Alignment] in the [Calibration] group on the [Cytometer] tab of the ribbon.



Note

[Optics Alignment] is enabled only when no plate is loaded.

The [Optics Alignment Wizard] window appears.

2 Follow the on-screen instructions to load a sample tube of Align Check beads in the "Extra Station 1" position, select the beads lot number to be used, and click [Start].

Optics Alignment Wash		-		
1. Start	Start Optics Alignment			
2. Migning	1-1. Prepare AlignCheck Beach			
3. Complete	Shake or vortex the AlignCheck Beach vial before use.			
	Add 2 drops of beach and 450 µl of PBS or DI water to a sample tube.			
	1-2. Set Sample Tube			
	Place the sample tube in the first position of the tube holder (Satu Station 1).			
	Erros Station 1			
	Align Check Bearts			
	A CONTRACTOR OF A CONT			
	1-3. Select Beed Lot Number.			
	Bead Lat Number VALD/10/ML02 *			
	1-4. Start Optics Alignment			
	Confirm that the loader host, door is closed, click Start.			
		Start	Cancel	

The optics alignment is executed, and the progress is displayed in the wizard.

You can monitor the count displayed above the progress bar as it increases.

Adjusting the Laser Output

Note

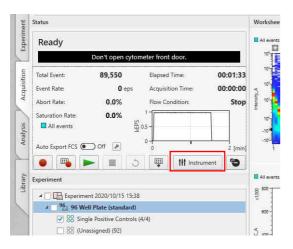
It is not recommended to adjust the laser output in a normal experiment. The output of each laser directly affects the waveform of the spectral reference and the spectral unmixing results.

You can adjust the output of each laser individually.

Тір

You can adjust the laser output when the instrument configuration mode is set to [Normal Mode] only.

1 Select a sample to acquire on the [Plate] panel, and click [Instrument].



The [Instrument Settings] dialog appears.

- **2** Place a check mark in the checkbox for the laser(s) to use on the [Laser] tab, and place a check mark in [Advanced Settings].
- **3** Click [Yes] in the displayed confirmation message dialog.

[Laser Power] is displayed on the [Laser] tab.

4 Adjust the laser output using the slider or enter a value directly.

aser	Detector & Threshold	Flow Control	Agitation	Ev	ent C	heck	Cleaning	Detector	(Advance	d
Laser	On/Off:		Las	er P	ower	:				
~	320nm			20	⊧¥-I	mW				
1	355nm			50	ŀ¥-I	mW				
1	405nm			60	₽ ₩- 1	mW				
1	488nm			40	ı.₩.,	mW				
1	561nm			60	⊧¥-I	mW				
-	637nm			60	ŀ₫-I	mW				
1	808nm			60	1-W-1	mW				
1 Adva	inced Settings						ĩ	nstrument	Settings ·	

5 When finished configuration, click [Close].

Adjusting Detector Voltages Separately

You can adjust the voltage for each detector channel.

Tip

You can adjust the detector voltages when the instrument configuration mode is set to [Normal Mode] only.

1 Select a sample to acquire on the [Plate] panel, and click [Instrument].

nent	Status	Worksh
Experiment	Ready	All ever
	Don't open cytometer front door.	10'
Acquisition	Total Event: 89,550 Elapsed Time: 00:01:33 Event Rate: 0 eps Acquisition Time: 00:00:00	10-
Acqu	Abort Rate: 0.0% Flow Condition: Stop	-Au
Analysis	Saturation Rate: 0.0% 1 All events 40.5 -	10- -10 ⁻¹
Ans	Auto Export FCS Off P 0 2 [min	-10-
~	• 🖷 🕨 🔳 🕉 🐺 🚻 Instrument 🔮	Alleve
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-	4 🗌 🔚 Experiment 2020/10/15 15:38	1008
	4 96 Well Plate (standard)	600 -
	Single Positive Controls (4/4)	
	88 (Unassigned) (92)	5

The [Instrument Settings] dialog appears.

2 Place a check mark in [Advanced Settings].

A confirmation message appears.

3 Read the content of the confirmation message, and then click [Yes] to proceed with the change.

The [Detector (Advanced)] tab appears.

- 4 Click the [Detector (Advanced)] tab.
- 5 Select the laser axis in [Laser] and adjust the voltages by moving the slider for each channel, then click [OK].

Laser	Detector & Threshold	Flow Control	Agitation	Event Check	Cleaning	Detector (Advan
Lasen	320nm 💌					
CH1:	6		C	-121:	0 8	e E
CH2:			C	-122:		E.
CH3:			C	-123:		ē.
CH4:			C	-124:		ē.
CH5:			C	-125:		
CH6:			C	-126:		
CH7:			CI	-127:		ř.
CH8:	7_ 7		C	-128:	(s	i.
CH9:			C	-129:	(s	Ē
CH10:	() s		C	-130:		E.
CH11:	0 s		C	-131:		í.
CH12:	() 8		C	-132:	0 e	i.
CH13:	0 s		C	-133:	E	i -
CH14:	Ū 8		C	-134:	Ū s	i.
CH15:	0 s		C	-135:	e	í.
CH16:	6		C	136:		
CH17:	Ū 8		0	137:		
CH18:	Ū 8					
CH19:	0 s					
CH20:						
🗸 Adva	nced Settings				Ir	nstrument Settings

Note

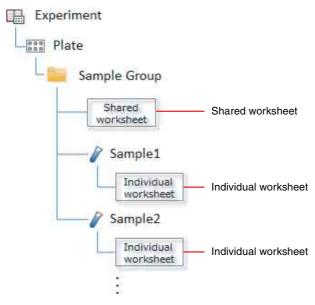
It is not recommended to adjust the detector voltages separately for each channel in a normal experiment. The output balance of each detector directly affects the waveform of the spectral reference and the spectral unmixing results.

Configuring Shared Worksheets

ID7000 Software provides worksheets in the following structure.

A worksheet for a sample group is called a "Shared Worksheet," and a worksheet for a sample is called an "Individual Worksheet."

A shared worksheet allows each sample within the sample group to use the same worksheet.



A shared worksheet supports two sharing modes: [Individual] mode and [Shared] mode.

• [Shared] mode

Refer to the shared worksheet when acquiring and analyzing each sample. After acquisition is completed, the shared worksheet is automatically copied to the corresponding individual worksheet.

For example, if you want to perform acquisition and analysis using the same plot, same gate, and same statistics for all samples within a sample group.

[Individual] mode

Refer to the individual worksheet when acquiring and analyzing each sample.

For example, in cases where a unique gate or plot is required for each sample prior to acquisition or in cases where a unique gate or plot is used for each sample when acquiring multiple single-stained samples within a sample group.

Depending on the purpose of the sample group, the initial worksheet mode can be set for each sample. For details, see "Setting the Initial Worksheet Mode for Each Sample" (page 91).

Changing the Worksheet Mode in Batch

You can change the worksheet mode for all samples within a sample group, a plate, or an experiment at the same time.

1 Right-click an experiment, plate, or sample group and select [Change Worksheet Mode] - [Individual] or [Shared] from the context menu.

When you change the worksheet mode, the [Plate] panel icon also changes.

In [Individual] mode



In [Shared] mode



Тір

If you right-click a sample group and change the worksheet mode of all samples in the sample group from [Shared] to [Individual], a dialog appears which prompts whether or not to copy the shared worksheet settings to the individual worksheets of all samples. Click [Yes] to copy the settings to all samples. Click [No] to change only the worksheet mode and maintain the configured contents of individual worksheets.

Changing the Worksheet Mode for Each Sample

If you want to measure a single positive control (usually a worksheet in [Individual] mode) and multi-stained samples (usually a worksheet in [Shared] mode) in the same sample group, or perform fluorescence compensation between the single positive control and multi-stained samples, you can change the worksheet mode for each sample, for example, when you want to share a matrix or when you want to adjust the gate for a specific sample when the measurement results are out of alignment. 1 Right-click a sample, and select [Properties] from the context menu.

The sample properties dialog appears. Right-click a sample, and select [Change Worksheet Mode] - [Shared] or [Individual] from the context menu to change the worksheet mode.

2 Select [Shared] or [Individual] in [Worksheet Mode], then click [OK].

Sample Information				
Name:	Unstained			
Cell (optional):				
Condition (optional):				
Memo (optional):				
Sample Keywords				
	Name		Value	
Keyword 1:	Project	New Experie	ment 001	
Keyword 2:	Phase	Test 001		
Keyword 3:				
Keyword 4:				
Keyword 5:				
Keyword 6:				
Keyword 7:				
Keyword 8:				
Keyword 9:				
Keyword 10:				
Sample Status				
Status	Acquired	Aborted Events:		
Acquisition Date:	8/27/2022 9:16:09 PM	Abort Count:	0	
Acquisition Condition:		Abort Rate:	0.0%	
Total Events:	10,000	Worksheet Mode:	Individual Not Available	
		Unmixing Cache	HOL AVAILABLE	
Sample Settings		-		
Worksheet Mode:	Individual ○ Shared ≈	•		
Sample Group:	88 Sample Group - 3			•
	Auto acquisition target			

The worksheet mode of the sample is changed.

Sharing a Worksheet as the Shared Worksheet

Right-click a well or sample tube, and select [Copy to Shared Worksheet] from the context menu. The Individual Worksheet for the specified sample is copied as the shared worksheet for the sample group.

Тір

The shared worksheet is displayed by selecting the sample group within [Experiment List].

Copying a Shared Worksheet to the Individual Worksheet of All Samples

Right-click the sample group, and select [Copy to all Individual Worksheets] from the context menu. The shared worksheet of the sample group is copied to the Individual Worksheet of all samples.

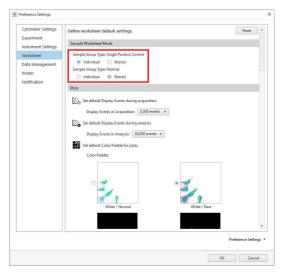
Setting the Initial Worksheet Mode for Each Sample

Depending on the type of sample group, the initial worksheet mode can be set for a sample.

1 Click [Information] on the [File] tab of the ribbon, then click [Preference Settings].

The [Preference Settings] dialog appears.

2 Select [Worksheet] from the menu on the left side, set the initial worksheet mode for samples in each sample group type in [Sample Worksheet Mode], then click [OK].



[Sample Group Type: Single Positive

Control]: Selects the initial worksheet mode for samples when the sample group type is [Single Positive Control].

- [Individual]: Useful when you want to use a unique gate and plot for each sample.
- [Shared]: Useful when you want to use the same gate and plot for samples within a sample group.
- [Sample Group Type: Normal]: Selects the initial worksheet mode for samples when the sample group type is [Normal].
 - [Individual]: Useful when you want to set different gates for different distributions of measurement results for the same sample group.
 - [Shared]: Useful when you want to set a common gate because the distribution of measurements for the same sample group is similar.

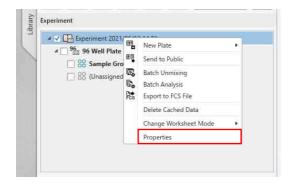
Attaching Keywords to Each Sample

Using the sample keyword function allows you to set keywords for each sample. Up to 10 keyword names can be set for each experiment and a value can be entered in each sample for keywords set in an experiment. The set keyword and value are added as separate keywords for each sample when exporting in FCS format.

Setting Keyword Names for an Experiment

Keyword names are configurable for each experiment.

1 Right-click an experiment on the [Acquisition] tab or [Analysis] tab, and select [Properties] from the context menu.



2

Chapter 7 Advanced Operations

In [Sample Keywords], enter keyword names in [Keyword 1 Name] to [Keyword 10 Name], and click [OK].

A keyword cannot be used if a keyword name is not entered (blank field).

xperiment Properties		
Information		
Name:	Experiment 2022/08/27 21:04	
Cytometer Type:	Auto Sampler	
Date Created:	8/27/2022 9:13:01 PM	
Experiment Owner:	user1	
Investigator (optional):	user1	
Operator (optional):	user1	
Memo (optional):	dia 1	
Citeria Citeria		
Audit Trail:	Off	
Sample Keywords		
Keyword 1 Name:	Project	
Keyword 2 Name:	Phase	
Keyword 3 Name:		
Keyword 4 Name:		
(C)		
Keyword 5 Name:		
Keyword 6 Name:		
Keyword 7 Name:		
Keyword 8 Name:		
Keyword 9 Name:		
Keyword 10 Name:		
Settings		
Mode:	 Standardization Normal 	
Signature		
Date Created:		
Usen		
Signature Name:		
	0	K Cancel

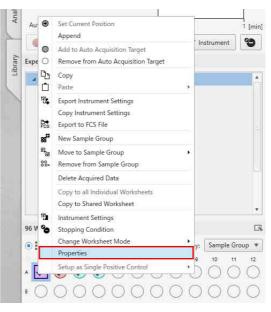
The keyword names are configured in the experiment.

Attaching a Keyword to a Sample

For keyword names configured in an experiment, you can set the value of a keyword for each sample. The keyword value is set using the properties dialog of the sample or the plate layout editor dialog.

Setting in sample properties

1 Right-click a well or tube on the [Acquisition] tab or [Analysis] tab, and select [Properties] from the context menu.



The sample properties dialog appears.

2 In [Sample Keywords], enter a keyword value in [Value] for any of the [Keyword 1 Name] to [Keyword 10 Name] keyword names, and click [OK].

ample Information				
Vame: Cell (optional):	Unstained			
Condition (optional):				-
vlemo (optional):				
imple Keywords				
inpic neyword?	Name		Value	٦
eyword 1:	Project	New Experim	nent 001	
eyword 2:	Phase	Test 001		
eyword 3:		1		
eyword 4:				
leyword 5:		6		
leyword 6:		1		
eyword 7:				
leyword 8:		1		
eyword 9:		4		
leyword 10:				
imple Status				
tatus:	Acquired	Aborted Events:		
equisition Date:	8/27/2022 9:16:09 PM	Abort Count:	0.0%	
equisition Condition	Normal 10,000	Worksheet Mode:	Individual	
otal Events	10,000	Unmixing Cache	Not Available	
imple Settings				
Vorksheet Mode:	 Individual O Shared ee 			
ample Group:	88 Sample Group - 3			Ψ.
	C Auto acquisition target			
			OK	Cancel

The keyword is set for the sample

Setting using the plate layout editor

1 Click [Layout Editor] in the [Plate] group on the [Acquisition] tab or [Analysis] tab of the ribbon.

			Worksheet	Tools			
Cyt	ometer	5	Sample Gro	oup - 1			
	888	888	80	0	000	000	80
Save as emplate	New	Delete	Load Template	Layout Editor	Export Layout to CSV File	Import Layout from CSV File	New
- Tie				Plate		(15)	

The [Layout Editor] dialog appears.

2 In [Sample Keywords], enter a keyword value in [Value] for any of the [Keyword 1 Name] to [Keyword 10 Name] keyword names, and click [Close].

ayout Editor					
Ⅲ ○ 三	Color coded by: Semple Group +	Unstained (A01)			
	2 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	Sample Information			
🗹 🖉 🖉 📿 🔾		Neve	Unstained		
		Cell (aptional):			
		Condition (optional) Memo (optional)			
		Meno coptional)			
		Sample Keywords	Name	136ar	_
		Keyword 1:	Project.	New Experiment 001	_
		Keyword 2:	Phase	Test 001	=
		Keyword 3:			
		Keyword &			
		Keyword 5:		1	
		Keyword 6:		1	
		Keyword 7:			
		Keyword 8:			
		Keyword 9:			
		Keyword 13			
		Sample Status			
			Acquired 8/27/2022 9:16:09 PM	Worksheet Mode Individual Unmixing Cache Net Available	
			1/2//2022 9/1809 PM	Unmang Cache Inter Available	
		Sample Settings			
		Worksheet Mode:	Individual		
		Sample Group:	88 Sample Group - 1		
			Auto acquisition target		
		To select multiple sample	es, hold down the Ctil or Shift ke	y and click samples.	
					Close

The keyword is set for the sample

Editing and Importing Plate Metadata

You can export metadata for a plate to a CSV file, edit the metadata using Microsoft Excel, and then import the edited metadata.

1 In the experiment list, select a plate whose metadata you want to export, and click [Export Layout to CSV File] on the [Acquisition] tab or [Analysis] tab of the ribbon.

You can also perform the same operation by rightclicking the plate and selecting [Export Layout to CSV File] from the context menu.

	100	orksheet	Tools			
Cytomete	er	Well - A	01			
888	800	0	000	000	80	80
Delete	Load Template	Layout Editor	Export Layout to CSV File	Import Layout from CSV File	New	Delete I
		Plate		:15		San

The [Export Layout to CSV File] dialog appears.

2 Specify the file export destination, then click [Export].

Export Layo	ut to CSV File		2
Output File:	D:\data\20200115.csv		Browse
		Export	Cancel

The metadata is exported as a CSV file.

3 Open and edit the exported file in Microsoft Excel or other software.

Example: Plate metadata

1	A	В	с	D	E	F	G	н	1	1	К	L	м	N	0
	Experiment	Plate	Order	Row	Column	Sample G	Sample	Cell	Condition	Мето	Keyword 1	Keyword	2 Keyword	Keyword	Keyword 5
2	Experiment	96 Well	Plate (stand	A	1	Single Pos	Unstained	5			New Elem	Test 001			
3	Experiment	96 Well	Plate (stand	A	2	Single Por	APC				New Elem	Test 002			
4	Experiment	96 Well	Plate (stand	A	3	Single Pos	FITC				New Elem	Test 003			
5	Experimen	96 Well	Plate (stand	A	4	Single Pos	PE				New Elem	Test 004			
6	Experiment	96 Well	Plate (stand	A	5		Well - A0	5							
7	Experiment	96 Well	Plate (stand	A	6		Well - AO	5							
8	Experiment	96 Well	Plate (stand	A	7		Well - A0	7							
9	Experiment	96 Well	Plate (stand	A	8		Well - A0	3							
10	Experiment	96 Well	Plate (stand	A	9		Well - A0	2							
11	Experiment	96 Well	Plate (stand	A	10		Well - A1	0							
12			Plate (stand		11		Well - A1	L							

4 In the experiment list, select a plate whose metadata you want to import, and click [Import Layout from CSV File] on the [Acquisition] tab or [Analysis] tab of the ribbon.

You can also perform the same operation by rightclicking the plate and selecting [Import Layout from CSV File] from the context menu.

X	Marksheel	Tools					
ter	Well - A	401					
	0.000	808	000	80	80	88 488	
Load Template	Layout Editor	Export Layout to CSV File	Import Layout from CSV File	New	Delete	Duplicate	In
	Plate		:54		S	ample Grou	p)

The [Import Layout from CSV file] dialog appears.

5 Specify the metadata file to import, check the file contents, and click [Import].

Updated locations are displayed with a green background, warning locations with a yellow background, and error locations with a red background.

Changes to sample groups are not reflected.

	Single Positive Controls								Kenverd 3 Value	
				Non Demont 001	Text 001					
32	Single Positive Controls	APC .		New Dament 002	Test 002					
8	Single Positive Controls	PTK:		New Dement 003	Test 005					
64	Single Positive Controls	95		New Dement 004	Test 004					
										5
6701.										

The metadata is imported.

Saving an Experiment as a Template

You can save an existing experiment as a shared template (Public Templates) or a private template (My Templates). Saving an experiment as a template allows you to create an experiment with the same settings as the original experiment, even after the experiment has been deleted.

- 1 Select an experiment you want to save as a template on the [Acquisition] tab or [Analysis] tab.
- 2 Click [Save as Template] in the [Experiment] group on the [Acquisition] tab or [Analysis] tab of the ribbon.

-	File	Analys	is U	Inmixing		Cytometer
Х	EQ.	EQ.	ß	<u>888</u>	2 500	0000
0	Open in Acquisition	Send to Public	Save as Template	Delete	Layout Editor	Export Layout to CSV File
1	Exp	eriment				Plate

The [Save as Template] dialog appears.

3 Set each item, then click [OK].

Template Selection:	My Templates 🛛 🔻	
Template Name:	98well_No2	
Audit Trail:	💿 On 🔿 Off	

[Template Selection]: Select the template type. [Template Name]: Enter the name of the template. [Audit Trail]: Turns the audit trail function on/off.

The experiment is saved as a template.

Configuring Acquisition and Analysis Settings

Configuring the Virtual Filter Function

Using the virtual filter function allows you to set the detection channel range for each fluorochrome. These settings are used when spectral unmixing is off.

1 Click [Unmixing Settings] in the [Unmixing] group on the [Unmixing] tab of the ribbon.

ID							Worksheet Trail	le .	
-	/Fie	Analysis	U	nmixing	C)	tometer	PerCP Cy5.5		
Я	Matrix	15 color	Ŧ	88	100	200	125	100	
9	🗹 Use d	common matrix		Calculate Matrix	Apply	Unmixing Settings	Spectral Reference Adjuster		Lo
1			Unmixi	ng	-	-	5	ipectrai Ref	erer

The [Unmixing Settings] dialog appears.

2 On the [Fluorochrome] tab, right-click the fluorochrome whose channels you want to adjust, and select [Edit Virtual Filter] from the context menu.

						Universal Negative			>>
luoro	chrome		Add			0	Ex. Laser:	ALL ¥	ace
Index	Marker	Ruorachrome	Ex. Laser	Mode	SR	Nepative	Positive		Spectral Reference
1	CD38	8UV496	355mm		0	0	0		100
z	CCR7	BUV563	355mm		0	0	0		Spec
3	CD11B	BUV661	355mm		0	0	0		
4	CD14	8UV737	355mm	Normal	•	0	0		
5	1gD	BUV805	355mm		0	0	0		
6	CD11C	BV421	405rm	Normal					
7	CD7	SB. Clear Ga			0	0	0		
8	CCR5		ctral Reference		0	0	0		
9	CD56	BV Edit Vins		al .	•	0	0		
10	CD161	EV	noncerone		0	0	0		
11	CD45R0	BV	s		0	0	0		
12	CCR4	EV605	405mm		0	0	0		
13	CD28	BV650	405 nm		0	0	0		
14	CD10	58702	405mm		0	0	0		
15	CD16	BV711	405mm	Normal	•		•		
16	CD2	BV750	405 nm		0	0	0		
17	CKCR3	BV786	405mm		0	0	0		
18	CD25	AF488	488mm	Normal	۰		0		
19	CD127	AF532	488em		0	0	0		
20	CD45RA	PerCP	488em		0	0	0		
21	CD3	PerCP-Cy5.5	488 nm	Normal	•		0		
22	PD-1	PerCP-elluor710	488em		0	0	0	w.	
						Color	Panel settings • Fluorochron	re settings •	
Autofl	uorescence	btA							
Index	Use as AF	Autofluorescence	Ex.Laser	Mode	SR	Negative	Positive		
							Autoflu	erescence •	
							Calculate	Apply	

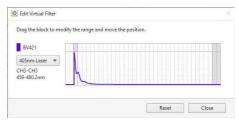
The [Edit Virtual Filter] dialog appears.

3 Set the channels for which the virtual filter is applied.

Dragging either side of the channel marker allows you to extend or reduce the set of contiguous channel markers.

Dragging within a channel cell moves all channel cells.

You can also change the laser to apply.



Tip

To restore the default settings, click [Reset].

4 Click [Close].

The settings are reflected in the fluorochrome list.

5 Click [Close] in the [Unmixing Settings] dialog to close the dialog.

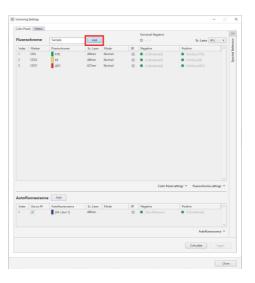
Adding a New Fluorochrome

1 Click [Unmixing Settings] in the [Unmixing] group on the [Unmixing] tab of the ribbon.

ID							Workshiel Ten	le :	
1	File	Analysis	Ur	mixing	Cy	tometer	PerCP Cy5.5		
gc	Matrix:	15 color	•	00	100	20		02	
0	🗹 Use o	common matrix		Calculate Matrix	Apply	Unmixing Settings	Spectral Reference Adjuster		Loa R
1			Urimixir	ng			Ş	ipectrai Refi	aren

The [Unmixing Settings] dialog appears.

2 Enter a fluorochrome name, then click [Add].



The [Add New Fluorochrome] dialog appears.

3 Set each item, then click [OK].

Add New Fluc	rochrome	×		
Name:	Sample			
Laser:	320nm 💌			
Start Channel:	CH1 (360.5-390) 💌			
End Channel: CH1 (360.5-390) 🔻				
Color:	Gray	•		
Copy from Name: Load from Preview:				
	OK Can	cel		

Displays an image representing the spectrum waveform for the currently selected fluorochrome.

[Name]: Enter the name of the fluorochrome.
 [Laser]: Click ▼ and select the laser to use.
 [Start Channel] / [End Channel]: Enter the range of valid channels for the fluorochrome.

[Color]: Click V	and select the display color from
the color picke	er.

Name:	Sample	
Laser:	320nm	¥.
Start Channel:	CH1 (360.5-390)	*
End Channel:	CH1 (360.5-390)	*
Color:	Gray	
Spectrum Imai Blank Copy from Name: Load from Preview:	Available Colors	
	Standard Colors	

[Spectrum Image]: Select the fluorochrome spectrum image waveform.

Selecting the waveform displays a preview.

- [Blank]: Does not display a waveform.
- [Copy from another fluorochrome]: Uses another waveform registered in the database. When selected, enter the name of the fluorochrome.
- **[Load from CSV file]:** Loads waveform information from a CSV file. When selected, click [Browse] to specify the CSV file. The CSV file has the following format.

Wavelength, Intensity Example: 500, 0.01 501, 0.02 502, 0.04 ... 625, 1 ... 799, 0.01 800, 0

The fluorochrome is added.

The added fluorochrome is also automatically added to the fluorochrome drop-down list in the [Unmixing Settings] dialog.

To customize the display color of a fluorochrome

1 Click [Color] in the [Add New Fluorochrome] dialog, then click [Advanced] in the [Available Colors] color picker.



The dialog displays the following palette.

2 Specify a color.

Name:	Sample		
Laser:	320nm		¥
Start Channel:	CH1 (360.5-390)		w.
End Channel:	CH1 (360.5-390)		¥.
Color:	Gray		1
💿 Blank	5		
Copy from		#FF8080	180
Copy from	R	#FF8080	12
Copy from		#FF8080	
Copy from Name:	R	#FF8080	12
Copy from Name:	R	#FF8080	12

3 Click anywhere outside the palette to close the palette.

Exporting an FCS File

- **1** Select the data you want to export in the [Experiment] panel.
- 2 Click [Export to FCS File] on the [Acquisition] tab or [Analysis] tab of the ribbon.

You can also perform the same operation by rightclicking in the [Experiment] panel and selecting [Export to FCS file] from the context menu.



The [FCS File Export] dialog appears.

3 Place a check mark in the plates or sample tubes to export, specify the file format, specify the export destination, and click [Export].

elect Samples	Pulse Type (for fluorescence):
 AI → Esperiment 2020/10/15 18:50 ▲ → Big 98 Well Plate (standard) ▶ → Big Sample Group - 1 (1/1) ↔ ▶ → Big Sample Group - 1 (1/1) ↔ 	Area Area Height Width Data Type: CS File (Virtual Filter applied) (by Sample Group) Urmined FCS File (by Sample Group) Matrix Algorithm
	□ Paw FCS File (not unmixed)
	Optional FCS Text Segment Keywords (by sample):
	Sample name:
	\$INST:
	SEXP:
	SOP:
	SPROJ:
	SSMNO:
	SSRC:
	SCELLS
	SCOM:
CS version: 3.1 v	
ave in: C:\Users\fcm\Desktop	Browse
Export FCS status can be checked on ID7000 Dz	ta Agent icon in Nortification Area on the Task Bar.
1	3 0%

[Pulse type]: Select the pulse type.

- [Data Type]: Select the data type for the FCS file to be exported.
- [FCS File (Virtual Filter applied) (by Sample Group)]: Export the fluorochrome data acquired from the virtual filter without performing spectral unmixing.
- [Unmixed FCS File (by Sample Group)]: Perform spectral unmixing and export the resulting fluorochrome data.
- [Raw FCS File (not unmixed)]: Export the acquired fluorescence spectral data as-is.
- [Optional FCS Text Segment Keywords (by sample)]: Set metadata for each sample.



Keywords and their values set using the sample keyword function are exported automatically.

[FCS version]: Selects the FCS file version. **[Save in]:** Specify the file save destination.

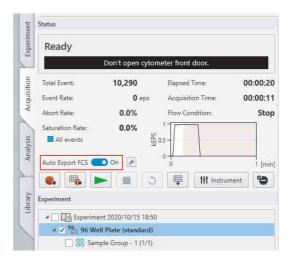
Note

Check the manual of the analysis software, select the FCS file version, and export.

The FCS file is exported.

Exporting FCS File Automatically During Acquisition

On the [Status] panel, set [Auto Export FCS] to On.



The [Auto Export FCS Settings] dialog appears.

2 Set each item, then click [OK].

Auto Expor	t FCS Settings	
Select	data type to export	
	or fluorescence):	
Area	🕝 Height 📝 Width	
Data Type:		
C3.1	(Virtual Filter applied)	
Unmix	PART CONTROL AND THE ADDRESS OF ADDRES	
And Contraction	Inmixing Matrix will be applied to exported data.	
Raw FC	S File (not unmixed)	
Ontional ECS	Text Segment Keywords:	
	n be changed via Plate Properties dialog.	
Sample nar	<u>_ 0. 0. 0.</u>	
SINST:		
SEXP:	administrator	
SOP:	administrator	
SPROJ:	Experiment 2020/10/26 16:30	
\$SMNO:	A01	
SSRC:		
SCELLS:	Well - A01	
SCOM:		
	1	
FCS version:	3.1 w	
Save in:	C:\Users\fcm\Desktop	Browse
	Export FCS status can be checked on ID7000 Data Agent icon	in Nortification Area on the Task Ba
		OK Cancel
		Cancel

[Pulse Type (for fluorescence)]: Select the type of pulse to output for the fluorochrome data.

- [Data Type]: Select the data type for the FCS file to be exported.
 - [FCS File (Virtual Filter applied)]: Export the fluorochrome data acquired from the virtual filter without performing spectral unmixing.

- [Unmixed FCS File]: Perform spectral unmixing and export the resulting fluorochrome data.
- [Raw FCS File (not unmixed)]: Export the acquired fluorescence spectral data as-is.
- [FCS version]: Select the FCS file version.
- [Save in]: Select the file output destination folder.

Tip

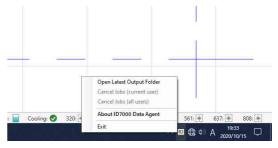
Clicking *on the right side of [Auto Export FCS]* displays the [Auto Export FCS Settings] dialog.

Using ID7000 Data Agent

In ID7000 Software, you can use the ID7000 Data Agent application to export FCS file.

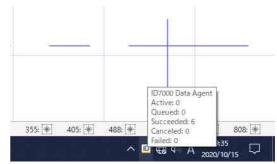
ID7000 Data Agent starts when the FCS file export operation in ID7000 Software is executed, and its icon is displayed in the task tray.

You can right-click the ID7000 Data Agent icon to display a menu and obtain various information related to FCS file export.



Displaying the FCS File Export Status

You can display the FCS file export status by hovering the mouse over the ID7000 Data Agent icon in the task tray.



Opening the FCS File Output Folder

Right-click the ID7000 Data Agent icon and select [Open Latest Output Folder] from the context menu to open the FCS file output folder currently in use or most recently used.

A log file recording the FCS file export status is also exported to the output folder.

Canceling FCS File Export

Right-click the ID7000 Data Agent icon and select [Cancel Jobs (current user)] from the context menu to cancel the unfinished FCS file export process. Note that this operation can only cancel the FCS file export process executed by the user who is currently logged in to the ID7000.

Canceling FCS File Export for All Users

Right-click the ID7000 Data Agent icon and select [Cancel Jobs (all users)] from the context menu to cancel all unfinished FCS file export processes, regardless of the users who initiated the export processes.

This operation is available only when logged in using an administrator account. This menu item is not displayed if logged as a general user.

Displaying ID7000 Data Agent Version Information

Right-click the ID7000 Data Agent icon and select [About ID700 Data Agent] from the context menu to display the version information for ID7000 Data Agent.

Exiting ID7000 Data Agent

Right-click the ID7000 Data Agent icon and select [Exit] from the context menu to exit ID7000 Data Agent. A confirmation message appears when the menu item is selected. Click [Yes] to exit ID7000 Data Agent. If an FCS file export process is in progress, the unfinished export process is canceled.

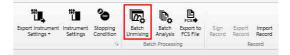
Batch Processing

Running Batch Spectral Unmixing

If an unmixing matrix that can be used for spectral unmixing already exists, the spectral unmixing for multiple samples can be performed at the same time.

1 On the [Experiment] panel, select the experiment, plate, or sample group as the target for batch spectral unmixing, and click [Batch Unmixing] in the [Acquisition] tab or [Analysis] tab on the ribbon.

You can also perform the same operations by rightclicking an experiment, plate, or sample group and selecting [Batch Unmixing] from the context menu.



The [Batch Unmixing] dialog appears.

2 Check the target displayed in [Batch Unmixing], and click [Start].

D Batch Unmixing			
Batch Unmixing: 🚫 Sample Group - 1			0/2
	Start	Abort	Close

When the batch spectral unmixing is completed, a confirmation message appears.

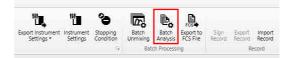
- **3** Click [OK].
- **4** Click [Close] to close the [Batch Unmixing] dialog.

Running Batch Analysis

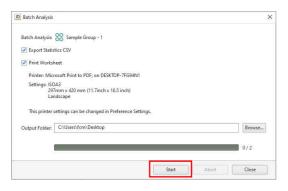
If a worksheet for which analysis has been completed already exists, the analysis for multiple samples can be performed at the same time.

1 On the [Experiment] panel, select the experiment, plate, or sample group as the target for batch analysis, and click [Batch Analysis] in the [Acquisition] tab or [Analysis] tab on the ribbon.

You can also perform the same operations by rightclicking an experiment, plate, or sample group and selecting [Batch Analysis] from the context menu.



2 Check the target displayed in [Batch Analysis], and click [Start].



[Export Statistics CSV]: Exports the contents of the statistics table as a CSV-format file.[Print Worksheet]: Prints the contents of the worksheet.

- **3** Click [Browse] for [Output Folder] and specify the export destination folder.
- 4 Click [Start].

When the batch analysis is completed, a confirmation message appears.

- **5** Click [OK].
- **6** Click [Close] to close the [Batch Analysis] dialog.

Tip

To change the destination printer or paper size for batch analysis, configure the printer and paper settings in [Preference Settings].

For details about preferences, see "[Preference Settings] dialog" (page 111).

Configuring Data Management Settings

You can import/export the experiment database.

For details about backing up the database, see "Backing Up/Restoring the Database" (page 108).

Tip

The menus and buttons displayed vary depending on the user account privileges.

Importing an Experiment

You can import experiment data created using another ID7000 for use on this unit.

1 Click [Import] in the [Database] window on the [File] tab.

tte	QC Cytowneter	(administrator) - 107000 Spectral Analyzer
ormation nt	Import Import experiments from the exported data files.	Hard Disk Drive Capacity 179708 Free Of 188108 Database Capacity
P	Export Export experiments to the files	9.258 Free OF 1008
Logout Exit	Backup Eack up the database (Administrator Only).	
	S Restore Restore the database (Administrator Crity).	
	Reset Reset The eletabase (Administrator Only).	

The [Import Experiment Data] dialog appears.

2 Specify the folder where the experiment data you want to import is stored, then click [Import].

D Import Experi	nent Data	\times
Import File(s):	C:\Users\fcm\Desktop\0818_ID7000\/D7000-20220807 Browse	
	0%	
	Import Abort Close	

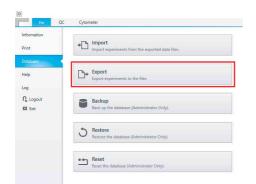
The experiment data is imported.

3 Click [Close] to close the dialog.

Exporting an Experiment

You can export experiment data for use on another ID7000 unit.

1 Click [Export] in the [Database] window on the [File] tab.



The [Export Experiment Data] dialog appears.

2 Select the experiments to be exported, then click [Export].

User: admini	strator 🔻				
Experiment List				Exporting Experiment List	
Experiment P	lame			Experiment Name	
Experim Experim Experim Experim Experim	entt 2020/01/14 20:57 entt 2020/01/14 20:24 entt 2020/01/14 19:36 entt 2020/01/14 19:30 entt 2020/01/14 19:32 entt 2020/01/14 19:32 entt 2020/01/14 19:12 entt 2020/01/14 18:21		Move Move All	Cill Experiment 2020/01/14 20:57 Cill Experiment 2020/01/14 20:24 Cill Experiment 2020/01/14 19:00 Cill Experiment 2020/01/14 19:33	
Output Folder.	D/\deta				Clear
oupurroute		_		0%	on a constant

For details about each item, see "[Export Experiment Data] dialog" (page 118).

The experiment data is exported.

3 Click [Close] to close the dialog.

Managing the Spectral Reference Library

The following operations are supported for the spectral reference library.

- Exporting/importing spectral reference data (see "[Library] Tab (Ribbon)" (page 146))
- Sharing a spectral reference between users (see *"[Search Result] in the [Library] Window"* (page 151))
- Deleting a spectral references from the library (see "[Search Result] in the [Library] Window" (page 151))
- You can update the spectral reference fluorochrome information registered in the spectral reference library.

This operation is available only when logged in using an administrator account.

Select the [Library] workflow tab.

The [Spectral Reference] window appears.

2 Perform the target operation.

8 5. 5. 5.																	
Input Sport Spains																	
¥.	Nes. Do	e) a	sof feads												As Learn Mill	*	
Spectral Reference.	Contract Country Country Country		and the	Maker	Residence.	fe tan	e Mha	. Then a	1700 H	ter V	ten Alber		10.00	Increased Section	e 0 Spectations		
3	 Later 		29	0087	Rentell	Iller								0.00	101		
	C tearbrook		25	00185	N-bucket	2004							10,000	ANDRY	421		
8	line		24	COURS	1.6432	Allen							Normal	0.00	144		
3	40m		23	CREE	1	20.00							These division of	0.00	974		
2	20140		72	0054	140	4700								0.00	254		
8	C 6Dvs		Zh	COLT	0000	Libra								0.00	10		
Apres 1	C Derekens		24	00.4	PLANO	1000	-					-		shallon .	-		
1	ANS:		10	CODE	. S.			100						-resource	424		
	ASSE		-	C209			-	- 10				-		otter	516		
1	T A090		04 194	C010		NAN DOOL	_							denar	514		
3	C AR T ARCOL				PT Davances		-										λ.
	0.0315		Th.	COURS		Nos	-					-		NEEMA	421		
	0.048		100	0008	H-bencia	50 hm								00566	421		
	0.0001		Th	C08	M-Cyll	How								Ability	IN .		
	0.010 0.000		251	CONDRA	NOT.	425.0								4hthtk			
	C 9501		2hr	C006	E/682	Allere									254		
	0.00		2N	C088.	Moet .	62%								x54309	421		
	0.007		25x	COFINE	Aureau	Hos							Normal	449.04	184		
	0.000		25a	COTO	M-C(A3)	How							Farred	Abellety	211		
	Centre		294	400	Re01-0,65	4000							North	ANDER	8		
	0.00		The	cost	and and	42've							Famal	0.00	111		
	- Autor		75	0007	APC .	60-0							North	OTTM	178		
	[m [mano		Ta.	CONT	al al c	625m							Nernal	-IEEEAA	15		
	1900		TN .	conr	ALC:	62%*	1						New	NUM	19		
	T MASS T MOT		Vie .	C06	16.64	-scine		i					Normal	detail	874		
	C R.Cortolida																
	- AuCP AuCP-055 - AuCP-offset10 - 0948																
	905 970 7 Tests 10																

Updating Spectral Reference Fluorochrome Information

1 Right-click the spectral reference you want to update in [Search Result] in the [Library] window, and select [Replace Fluorochrome] from the context menu.

CD197	BV421	405nm		
LD	Zombie-Aqua	405nm		
CD56	BV510	405nm		
CD19	BV510	Expor		
CD25	15400		to Public	
CD38	BUV496	Delet		
CD161	BV480		ce Fluorochrome	
CD127	AF532	Prope 400mm	rties	
CD8	AF488	488nm		
CCR7	BUV563	355nm	M	
CD335	PE-Dazzle594	561nm		ha

The [Replace Fluorochrome] dialog appears.

2 Select the fluorochrome to update from the list, and click [OK].

BUV661		
BUV737		
BUV805		
BV421		
BV480		
BV510		
BV570		1
BV605		
BV650		
BV711		
BV750		
BV785		,
BV785 Fluorochrome Int Laser: 405nm CH4-CH8 493.9-554.2nm	formation Emission Spectrum	

A confirmation message appears.

3 Click [Yes].

The fluorochrome information is updated.

Updating the Beads Lot File

You can update the beads lot information and select the correct lot number before running Daily QC and Performance QC.

Doby QC Illiand		-	
1. Stert	Start Daily QC		
2. Initialize Pressure 3. Fluidics Adjustment 4. Delay Time Setting	1-1. Prepare AdapCheck Brack Status or univer the MilipiCheck Beack with before use. Program a surger laboration of AdapCheck Beack in 420µ4 of DI water or PES.		
5. PMTV Adjustment 6. PMT32 Adjustment	1-2. Set Sample Tube Place the sample tube in the first position of the tube holder (Data Station 1).		
7. A/H Batio Calculation 8. FMTV Standardization			
9. Evaluating	Entra Station 1		
13. Completed			
	1-3 Selet Tani (ut Number) Best (ut Number) VMUTEWILLD		
		2at	Cano

Note

Use the same bead lot as selected in the software to ensure that QC will run correctly. QC may not be performed correctly if the beads do not match the lot number.

Contact your Sony representative for obtaining the bead lot file.

1 On the [QC] workflow tab, click [Import Bead Lot File] on the [QC] tab of the ribbon.

File	QC	Cytometer			
P	å.,	E.			0
Print	Import Bead Lot File	Export QC History to CSV	Export QC Result	Import QC Result	Criteria Settings

The file selection dialog appears.

2 Select a beads lot file (.bl), and click [Open]. The beads lot file is imported.

Setting QC Criteria

You can configure user-defined criteria settings for Daily QC and Performance QC.

1 On the [QC] workflow tab, click [QC Criteria Settings] on the [QC] tab of the ribbon.

-	File	QC	Cytometer			
2	P	١.				0
	Print	Import Bead Lot File	Export QC History to CSV	Export QC Result	Import QC Result	Criteria Settings

The [QC Criteria Settings] dialog appears.

2 Click [Daily QC] tab or [Performance QC] tab.

 Use All Result 				
O From:	To:	[]]	Calcul	ste
Last N Passes	30 Please	e set between 2 and 60		
Target CH	rCV Average	Criteria Range	SD Minus	SD Plus
488-FSC	2.81 %	None 💌	N/A	N/A
488-SSC	3.66 %	None 🔻	N/A	N/A
Target CH	rCV Average	Criteria Range	SD Minus	SD Plus
320-CH1	-	None 🔻	N/A	N/A
320-CH2	-	None 🔻	N/A	N/A
320-CH3	-	None 🔻	N/A	N/A
320-CH12	-	None 🔻	N/A	N/A
355-CH1	2.90 %	None 🔻	N/A	N/A
355-CH2	2.66 %	None 🔻	N/A	N/A
355-CH3	2.64 %	None 🔻	N/A	N/A
355-CH12	3.02 %	None 💌	N/A	N/A
405-CH1	1.16 %	None 🔻	N/A	N/A
405-CH2	1.18 %	None 🔻	N/A	N/A
405-CH3	1.23 %	None 🔻	N/A	N/A
405-CH12	2.66 %	None 🔻	N/A	N/A
488-CH12	1.67 %	None 🔻	N/A	N/A
561-CH12	3.96 %	None 🔻	N/A	N/A
637-CH23	2.28 %	None 🔻	N/A	N/A
808-CH36		None 🔻	N/A	N/A
808-CH37	-	None 🔻	N/A	N/A

3 Set the required date range for specifying criteria, and click [Calculate].

The calculation is performed and is reflected in the dialog.

ily QC Perform	ance QC			
 Use All Result 	lt			
O From:	To:		Calcula	ite
Last N Passe	s 30 Pleas	e set between 2 and 60		
Target CH	rCV Average	Criteria Range	SD Minus	SD Plus
488-FSC	2.81 %	±1SD ▼	2.56 %	3.05 %
488-SSC	3.66 %	±2SD ▼	2.82 %	4.50 %
Target CH	rCV Average	Criteria Range	SD Minus	SD Plus
320-CH1		None v	N/A	N/A
320-CH2		None 🔻	N/A	N/A
320-CH3		None 🔻	N/A	N/A
320-CH12		None V	N/A	N/A
355-CH1	2.90 %	±1SD ▼	2.76 %	3.05 %
355-CH2	2,66 %	±1SD ▼	2,56 %	2.76 %
355-CH3	2.64 %	±1SD ▼	2.55 %	2.74 %
355-CH12	3.02 %	±2SD ▼	2.68 %	3.36 %
405-CH1	1.16 %	±3SD ▼	0.82 %	1.50 %
405-CH2	1.18 %	±2SD ▼	0.98 %	1.38 %
405-CH3	1.23 %	±1SD ▼	1.12 %	1.34 %
405-CH12	2.66 %	±2SD ▼	2.20 %	3.12 %
488-CH12	1.67 %	±1SD ▼	1.53 %	1.81 %
561-CH12	3.96 %	±1SD ▼	3,44 %	4.47 %
637-CH23	2.28 %	±3SD ▼	0.30 %	4.26 %
808-CH36	-	None T	N/A	N/A
808-CH30		None T	N/A	N/A
000-CH57	-	None +	N/A	N/A

- **4** Select [Criteria Range] from the pull-down menu.
- **5** Click [OK] to close the dialog.

Tips

- [Criteria Range] can be set to [±1SD], [±2SD], or [±3SD].
- The QC Criteria settings can only be changed by users logged in with Administrator privileges.

Creating a Record with Electronic Signature

A record containing an audit trail can be created automatically and electronically signed for export as a PDF file. A record is an experiment in which the audit trail is automatically recorded.

Creating a Record Template

- **1** Select an experiment you want to save as a template on the [Acquisition] tab or [Analysis] tab.
- 2 Click [Save as Template] in the [Experiment] group on the [Acquisition] tab or [Analysis] tab of the ribbon.



The [Save as Template] dialog appears.

3 Select [On] in [Audit Trail], then click [OK].

Template Selection:	My Templates 💌	
Template Name:	98well_No2	
Audit Trail:	● On Off	
	OK	ncel

The experiment is saved as a template for which the audit trail will be recorded automatically.

Creating a Record

You can create a record using a record template.

1 Create an experiment using a template configured so that an audit trail will be recorded automatically.

You can check the audit trail setting in [Experiment Information] in the [Experiment Template] window.

Experiment Information		
Name:	Experiment 2022/08/29 0:21	
Cytometer Type:	Auto Sampler	
Experiment Owner:	user1	
Investigator (optional):	user1	
Operator (optional):	user1	
Memo (optional):		
Audit Trail:	On	
Setting		
Mode	Normal	

Note

A record can only be created using a record template.

2 Perform acquisition, analysis, and other operations.

The record is saved within the unit.

Signing a Record

You can create a record in PDF format and then electronically sign the file.

1 Select a record that you want to sign electronically in the Experiment Explorer.

Only records created by the user can be electronically signed.

2 Click [Sign Record] in the [Record] group on the [Acquisition] tab or [Analysis] tab of the ribbon.

Ø,	P.	FGS D	E7	E.,	EĮ		Analysis:	50,000 events
Batch Unmixing	Batch Analysis	Export to	Sign Record	Export Record	Import Record	View Audit Trail		
Bat	ch Processi	ng		Re	scord			Display Events

The [Sign Record] dialog appears.

3 Click [Start].

D Sign Record			>
Sign all the samples in the record.			
Signing Record: 🔚 Experiment 2022/10/04 0:16			
			0/4
	Start	Cancel	Close

The [User Authentication Dialog] dialog appears.

D User Authentio	ation Dialog			×
Enter usernam	ne and passwo	ord.		
Username:				
Password:				
		ОК	Close	

After the user name and password are verified, a record PDF file is created and is signed electronically.

- **5** When the completion message appears, click [OK].
- **6** Click [Close] to close the [Sign Record] dialog.

The following information is contained in the electronic signature.

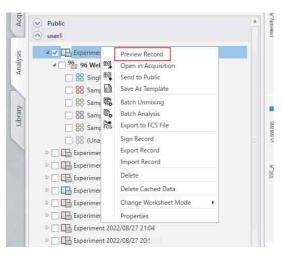
[Date & Time]: Date and time the record was electronically signed

[User]: Electronically signed user name **[Signature Name]:** Type of electronic signature

Checking a Record PDF File

You can check an electronically signed record PDF file.

1 Right-click the electronically signed record on the [Experiment] panel, and select [Preview Record] from the context menu.



The [Preview Record] dialog appears.

2 Check the contents of the record PDF file.



3 Click [Close] to close the [Preview Record] dialog.

Exporting a Record

You can export an electronically signed record. A record contains the following files stored and exported as a combined zip file.

- Record PDF file
- Record exdat file
- Data management file
- 1 Click [Export Record] in the [Record] group on the [Acquisition] tab or [Analysis] tab of the ribbon.

Ω_{α}	P.	FCS	E7	C.,	⊑⊒,	5	Analysis:	50,000 events
Batch Unmixing	Batch Analysis	Export to FCS File	Sign Record	Export Record	Import Record	View Audit Trail		

A confirmation message appears.

2 Click [Yes].

The [Export Record] dialog appears.

3 Specify the record export destination folder, then click [Export].

D Export Record	d Dialog	×
Output Folder:	C\User\\xxx\Deixtop	Browse
	0%	
	Export	Close

The [Set Password] dialog appears.

4 Select whether to configure a password for the exported record file, then click [OK].

ID ID7000 Software		×
Enter password for e	xported record.	
No Password:		
Password:		
Confirm Password:		
	OK Close	

[No Password]: Place a check mark in the checkbox to continue without setting a password.

- **[Password]:** If configuring a password, enter a password comprising a combination of 8 to 20 alphanumeric characters.
- [Confirm Password]: Enter the same password again for confirmation.

Close the [Set Password] dialog to start exporting the record.

5 When exporting is finished, click [Close] to close the dialog.

Importing a Record

You can import the following files stored in a record zip file.

- Record PDF file
- Record exdat file
- Encrypted hash file
- 1 Click [Import Record] in the [Record] group on the [Acquisition] tab or [Analysis] tab of the ribbon.

<i>D</i> .	P.	FCS +	E۷	E.	E ⊒	EJ	Analysis:	50,000 events
Batch Unmixing	Batch Analysis	Export to FCS File	Sign Record	Export Record	Import Record	View Audit Trail		
Bat	ch Processi	ng		Re	cord			Display Events

The [Import Record Dialog] dialog appears.

2 Specify the record file to import, then click [Import].

mport Report	Dialog	
port File(s):	C/UserVxxx/Desktop/user1_Experiment 20220916 1749_20220916224506.aip	Browse
	0%	

If a password was configured for the record file, a dialog for entering a password appears.

3 Enter the password, then click [OK].

D ID7000 Softwar	re	Х
Enter passw	ord for user1_Experiment 20220916 1749_20220916225053.	
Password:	•••••	
	OK Close	

- If the password is verified, the import starts.
- **4** When the completion message appears, click [OK].
- **5** Click [Close] to close the dialog.

Checking the Audit Trail

You can view the contents of the audit trail.

- 1 Select an experiment in the record list whose audit trail you want to view.
- **2** Click [View Audit Trail] in the [Record] group on the [Acquisition] tab or [Analysis] tab of the ribbon.

6	P.	FCS +	E۷	Eļ,	E⊒	E3	Analysis:	50,000 events
Batch Unmixing	Batch Analysis	Export to FCS File	Sign Record	Export Record	Import Record	View Audit Trail		
Bat	ch Processi	ng		Re	scord			Display Events

The [View Audit Trail Dialog] dialog appears.

3 Enter a comment of up to 200 characters in the [Comment] column, as required.

Only the user who performed each operation can enter comments.

ete & Terre	User	Diportment	Pieto	Sample Group	Semple	Well	Messept	Comment	Selfware Vestion	PCNome
10/2022 12:49:36 AM (FEC+09:00)	user1	Experiment 2022/10/1	96 Viel Piele Odender	Sample Group - 1	APC	A02	Shared Worksheek Gate [2] was maved from		Version 23.633321	065K30P-7K05i *
(9/2222 12:49/27 AM (UTC+09:00)	user1	Experiment 2022/13/1	Hi Viel Pate (dandar	Sample Geoup - 1	APC	A02	Shaved Worksheet, Gate [J.] use moved free		Weise 23.032/21	0558709-7666
HV202124047AM (UTC+0900)	user1	Esperiment 2023/13/1	96 Well Plate (standar	Sample Group - 1	APC	AG2	Shaved Worksheet (Dipra] gata [0] was one		Weston 2.8.0 19121	0158702-7669
HV252212:50:00 AM (UTC+09:00)	uper1	Esperiment 2020/10/1	96 Well Plate Otandar	Single Positive Control	92	A05	Selected Matrix was changed from Matrix 2		Westion 2.8.0 19121	0558302-7009
PR252 12:00 12 AM (UTC+09:00)	user1	Experiment 2022/11/1	96 Well Piete Ostender	Sample Group - 1	APC	ACC	Sets was acquired. E100XX events/ Disper-		Version 2.8.0 19121	055830P-7109
(\$2522 12:5533 AM (FEC+09:00)	user1	Esperiment 2022/12/1	96 Viel Piele (Mandar	Single Positive Control	н	A08	Acquisition start.		Version 23.632321	DISKIOP-Wok
(#2022 12:50:50 AM (#TC+09:00)	seert	Experiment 2023/13/1	96 Well Plate (clander	Single Positive Control	95	AGE	Shared Worksheet, Gate [A] use moved from		Webon 2.8.0 10121	0558709-7666
W2922 12:50:55 AM (UTC+09:00)	user1	Esperiment 2020/10/1	96 Well Plate Ottender	Single Positive Control	95	A00	Shared Worksheet, Gate [4] use moved from		Westion 2.8.030321	0058302-3769
4/2522 12:59:58 AM (UTC+89:08)	user1	Experiment 2022/11/1	96 Well Plate Ostender	Single Positive Control	92	A05	Shared Worksheet, Gate [4] was moved from		Weston 2.8.0 19121	0558302-7009
W2022 12:53:22 AM (JPC+09:00)	user1	Experiment 2022/11/1	96 Well Piete Odender	Single Pesitive Control	PE	A05	Bela was suspired. EXITIN events/ Dispejo		Version 2.8.0 19121	065K30P-7K09-
0/2023 12:54 12 AM (UTC+00:00)	san1	Experiment 2023/13/1	Hi Viel Pate (danks	Single Positive Control	FILE	A66	Acquisitor start.		Weise 23.012/21	0158709-3566
R/202112:54-49 AM (UTC+09:00)	seer1	Esperiment 2023/13/1	96 Viell Plate (standar	Single Positive Control	FILC	AG4	Bata was acquired. [10540] events/ (Stapping		Weston 2.8.0 10121	0558709-7666
W20212125315 AM (UTC+0900)	user1	Experiment 2022/11/1	95 Well Plate Ottender	Single Positive Control	rm;	A04	Undo action: Shared Worksheet, Gate [4] or		Weston 2.8.0 19121	0158702-7105
W2522 12:59:05 AM (UTC+09:00)	user1	Experiment 2022/10/1	96 Well Piete Ostender	Sample Group - 1	APC	ACC .	Shared Worksheet (Density) plot of (AI ever		Version 2.8.0 19121	055830P-709
(\$25321256354M (\$FC+0500)	user1	Experiment 2022/13/1	96 Viel Piele Odender	Sample Group - 1	APC	AG2	Shared Worksheets (Density) plot of (All ever		Vesioe 23.612321	0154307-3666
(#202112:56:25 AM (UTC+04:00)	seer1	Esperiment 2023 13/1	96 Viel Plate (standar	Sample Geoup - 1	APC .	AG2	Workshort area was changed.		Webon 23.033/21	0558709-7566
W202112:50:30 AM (UTC+09:00)	user1	Esperiment 2022/13/1	96 Well Plate Ottender	Sample Group - 1	APC .	A02	Shared Worksheet, Pict type of [All events]3		Weston 2.8.0 10121	0158709-7169
W2522 1-02-25 AM JUTC+19-00	uper1	Experiment 2023/10/1					Signed by our loser11		Weston 2.8.0 19121	0558302-7029

[Date & Time]: Date and time of operation
[User]: Name of user who performed an operation
[Experiment]: Experiment name
[Plate]: Plate name
[Sample Group]: Sample group name
[Sample]: Sample name
[Well]: Well position
[Message]: Description of operation
[Comment]: Miscellaneous information entered by the user who performed an operation (as required)
[Software Version]: ID7000 software version
[PC Name]: Name of the computer on which an operation was performed

Items Recorded in a Record PDF File

The following items are recorded in a record PDF file.

- Type of signature
- Electronic signature (user name and date & time)
- User name
- Full name of user
- Name of the computer on which the PDF file was created
- Experiment information
- Experiment structure
- Plate information
- Sample group information
- Sample information
- Instrument settings
- Fluorescence unmixing settings
- Data acquisition settings
- Stop conditions
- Data acquisition results
- Worksheet (plots)
- Worksheet (gates and statistics table)
- Audit trail

Backing Up/Restoring the Database

You can back up the ID7000 Software database. You can also restore experiment data that has been backed up, as required.

You can also delete all data and reset the database. This operation is available only when logged in using an administrator account.

Tips

- Backup/restore operations are supported on NTFSformat file systems. Other format file systems are not supported.
- Backup operations are performed for external storage only.

Backing up the Database

1 Click [Backup] in the [Database] window on the [File] tab.

	QC Cytometer
Information Print	Import experiments from the exported data files.
Database	Indian address and the address and the
Help	Export Export experiments to the files
Log	
∩ Logout ⊠ Exit	Backup Back up the database (Administrator Only).
	C Restore Restore the database (Administrator Only).
	Reset Reset the database (Administrator Only).

The [Backup Database] dialog appears.

2 Click [Browse] and specify the backup data storage destination, then click [Backup].

Backup Databa	se		
USB external sto	age device is recommended.		
The device must	be NTFS formatted.		
Output Folder:	D:\data\Backup		Browse
L		0%	
		Backup	Close

The database backup commences and a progress bar is displayed.

When the database is backed up, a confirmation message appears.

3 Click [OK].

Restoring a Database

Tip

Restoring a database loads the information from a previous database backup, erasing and replacing all information in the current database.

1 Click [Restore] in the [Database] window on the [File] tab.

File QC	Cytometer
Information Print	Import Import experiments from the exported data files.
Database.	
Help	Export Export to the files
Log	
∩ Logout ⊠ Exit	Backup Back up the database (Administrator Only).
	3 Restore Restore the database (Administrator Only).
	Reset Reset Reset the database (Administrator Only).

The [Restore Database] dialog appears.

2 Click [Browse] and specify the backup data to restore, then click [Restore].

ID Restore Data	abase	×
Backup File:	D:\data\Backup\20200115-095443\\D7000DatabaseFu	Browse
	Restore	Close

A confirmation message appears.

3 Click [Yes].

ID7000 Software exits.

4 Launch ID7000 Software.

The database is restored. When finished, a confirmation message appears.

5 Click [OK].

Resetting the Database

You can delete all users, experiments, and analysis results by resetting all data in the database.

Notes

• Resetting the database resets all data, including user accounts, experiments, and analysis results. Before

resetting the database, it is recommended that the database be backed up.

- After resetting the database, ID7000 Software automatically shuts down.
- 1 Click [Reset] in the [Database] window on the [File] tab.

Import Import experiments from the exported data files. Export Export experiments to the files Export Export experiments to the files Export Export experiments to the files
Export experiments to the files Backup
Export experiments to the files Backup
3 Restore Restore the database (Administrator Only).
•↔ Reset
ĺ

A confirmation message appears.

2 Place a check mark in the [Reset Database] checkbox, then click [Reset].

ID Reset Database		×
Reset function deletes all users, experiments and	l results.	
After reset data, ID7000 Spectral Analyzer will be	shutdown automatically.	
Reset database		
	Reset	lose

After the database is reset, ID7000 Software automatically shuts down.

Managing Logs

You can display the error/warning log, and export it as a CSV-format file.

Click the button for the log you want to display in the [Log] window on the [File] tab. The log is displayed in the area on the right.

		Cytus Algoried Leg					
	Optics Alignment Log		Menane	feed	Position Y Basel Lot	Menape	
	en ossisteration of	10 TO 2000 100 TO 10		failed	Contract Contractor		
dane.		3.20.2020 Moth WAM		Faird		Outro, discovert is fields.	
	Flow farm Calibration Log Does for out-adjuster top						
Segnal Bel	Drop and Warning Log Drop and Warning Log						
	Der log Store aus tep Admentistic ortet						

For details about reading logs, see "[Log] Window" (page 120).



Window Description

Tips

- The menus and buttons displayed vary depending on the user account privileges.
- The screenshots displayed in this document may vary from the actual software.

[File] Window

The [File] window is displayed by clicking the [File] tab of the ribbon in the top left of the main window.

For details about [Logout] and [Exit], see "[File] tab (ribbon)" (page 22).

[Information] Window

The [Information] window is displayed by clicking [Information] in the menu on the left side. The [Information] window is used to change login passwords, set institution information, and add/modify user accounts.

Example: Administrator account login

File QC	Cytometer
formation	
rint	Preference Settings Change software default settings for each user.
atabase	
	Change Password
elp	Change your current password.
og	
Logout	Institution Information
3 Exit	Institution information Show your institution information (Only administrator can edit).
	Cytometer Model
	Show cytometer model (Only administrator can edit).
	Account Settings
	Change, add and delete the user (Administrator only).

[Preference Settings]: Various default function

settings can be saved by each user in user preferences. The values set here become the default values used for each function.

Clicking this button displays the [Preference Settings] dialog (*page 111*).

- [Change Password]: Allows you to change the password used to log in to ID7000 Software. Clicking this button displays the [Change Password] dialog (*page 28*).
- **[Institution Information]:** Allows you to specify information about the institution using the instrument.

Clicking this button displays the [Institution Information] dialog (*page 116*).

- **[Cytometer Model]:** Specifies the instrument's model number for control by ID7000 Software. This button is displayed when logged in using a user account, but only an administrator account has permission to change the setting. Clicking this button displays the [Cytometer Model] dialog (*page 29*).
- [Account Settings]: Allows you to configure accounts for those who will use ID7000 Software. This button is displayed only when logged in using an administrator account. Clicking this button displays the [Account Settings]
 - dialog (page 116).

[Preference Settings] dialog

The [Preference Settings] dialog is used to save various default function settings by each user in user preferences.

User preferences are loaded automatically, eliminating the need for some configuration settings when you want to perform acquisition, analysis, or sorting.

The [Preference Settings] dialog is displayed by clicking [Information] – [Preference Settings] on the [File] tab of the ribbon.

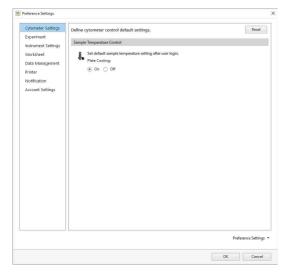
The items that are displayed vary depending on the user account privileges.

Тір

You can click [Reset] to restore the ID7000 Software default values for each setting.

• [Cytometer Settings] tab

Defines default settings related to the instrument.

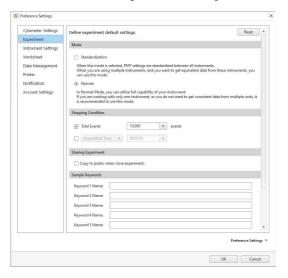


[Sample Temperature Control]

Sets whether to cool the multi-well plate holder to $4 \text{ }^{\circ}\text{C}$ (39 $^{\circ}\text{F}$) after logging in.

• [Experiment] tab

Defines default settings related to experiments.



[Mode]

- Selects the instrument configuration mode.
 - [Standardization]: The output of each channel is standardized so that the SSC and fluorescence detection sensitivities are the same between multiple instruments. When the standardized settings (ST) are the same in software on multiple instruments, the sensitivity of the instruments is roughly aligned. Standardized data is obtained by running Daily QC, and is updated according to the instrument status.
 - **[Normal]:** Select this mode when there is no requirement to correlate data between multiple instruments.

[Stopping Condition]

Selects the data acquisition stop condition and its value. You can also enter a value directly from the keyboard.

- **[Total Events]:** Stops when the number of events reaches the specified value.
- [Elapsed Time]: Acquisition stops when the accumulated elapsed time since the start of data acquisition reaches the specified value.
- [Acquisition Time]: Acquisition stops when the data acquisition time reaches the specified value.

[Sharing Experiment]

[Copy to public when close experiment.]: Place a check mark in the checkbox to copy an experiment to [Public Experiments] when the experiment is closed.

[Sample Keywords]

[Keyword 1 Name] to [Keyword 10 Name]: Sets the sample keyword names to add to new experiments.

• [Instrument Settings]

Defines default settings related to the instrument for data acquisition.

	Define instrument control defaul	t settings.	Reset
Experiment Instrument Settings	Laser		
Worksheet Data Management Printer Notification Account Settings	Later On/Off: V 30mm V 40mm V 40mm V 50mm V 50mm Detector & Thesehold		
	PSC Gain: 17 (# Normal Mode SSC Voltage (%): 26.5 (# PMT Voltage (%): 320 nm: 46.8 (# 335 nm: 46.8 (# 405 nm: 46.8 (#))	- Standerdization Mode	
	488 nm: 46.8 +8-4	488 nm: 3.78 +84 (1.00 - 5.00) 561 nm: 3.78 +84 (1.00 - 5.00)	

[Laser]

[Laser On/Off]: Check the checkboxes of the laser(s) to be used.

[Detector & Threshold]

- Sets detector gain and threshold levels.
 - [FSC Gain]: Sets the forward scatter (FSC) gain level.
 - [SSC Voltage (%)] ([SSC Voltage (ST)] in Standardization mode): Sets the side scatter (SSC) PMT voltage.

Clicking displays a slider. Set the value using the slider or enter a value directly.

[PMT Voltage (%)] (or [PMT Voltage (ST)] in Standardization mode): Sets the PMT voltages of each laser channel in conjunction with each other. Clicking displays a slider. Set the value using the slider or enter a value directly.

[Threshold] [CH1]: Selects the channel for the threshold to trigger detection of events. The forward scatter (FSC) channel is commonly used as the trigger channel to isolate targeted populations from background noise and cell debris, as it is less susceptible to the effects of background noise.

[Threshold] [Value1 (%)]: Sets the threshold value as a percentage of the receive signal output range.

Clicking displays a slider. Set the value using the slider or enter a value directly.

[Threshold] [CH2]: Selects the fluorescence channel for the threshold to trigger detection of events.

[Threshold] [Value2 (%)]: Sets the threshold value as a percentage of the receive signal output range.

Clicking displays a slider. Set the value using the slider or enter a value directly.

[Windows Extension]: Selects the window extension.

Select [Narrow] when measuring small cells, or select [Wide] or [Extra Wide] when measuring large cells.

Use the following guidelines for particle size when specifying the window extension.

- [Narrow]: Up to 3 μm
- + [Normal]: Up to 10 μm
- [Wide]: Up to 20 µm
- [Extra Wide]: Up to 40 µm

[Flow Control]

Configures settings relating to the flow rate.

[Boost]: Sets the boost time.

Increasing the boost time allows more events to flow faster at the start of acquisition.

[Sample Flow Rate]: Sets the sample flow rate.

A lower sample flow rate provides more accurate acquisition. A higher sample flow rate may reduce the time required to accumulate a specific number of events.

[Acquisition Offset Time]: Sets the offset time between the start of data acquisition of each well in units of seconds.

[Agitation]

Sets agitation settings.

[Low Dead Volume]: To reduce the number of acquisition samples that are discarded, place a check mark in [Enable low dead volume mode].

[Enable sample agitation]: Place a check mark in the checkbox to enable the sample fluid agitation function.

[Pattern]: Select the agitation method.

• [Once]: Agitate only once before sample acquisition.

- [Cyclic]: Agitate repeatedly at regular intervals during sample acquisition.
- [Continuous]: Agitate continuously during sample acquisition.

[Mode]: Select the agitation mode.

- [Normal]: Agitate normally.
- [Short]: Agitate gently

Tip

When [Low Dead Volume] is enabled, [Agitation] is set to [Once] (fixed). To use [Cyclic] or [Continuous], first disable [Low Dead Volume].

[Event Check]

Sets whether to perform detection of falling event rates and air bubbles.

- [Off]: Disable detection of falling event rates and air bubbles.
- [On]: Enable detection of falling event rates and air bubbles.

[Auto Acquire Preferences]: Sets whether automatic acquisition stops or continues if the event rate drops or air bubbles are detected during automatic acquisition.

- [Stop]: Stops automatic acquisition when the event rate drops or air bubbles are detected.
- [Continue]: When the event rate drops or air bubbles are detected, cleaning is performed and automatic acquisition continues from the next sample.

[Cleaning]

[Cleaning Mode]: Selects the cleaning method used between acquisition of each well.

- [Inner and Outer]: Clean the interior and exterior of the sample probe by backflushing with sheath fluid.
- [Inner Only]: Clean the interior of the sample probe only by backflushing with sheath fluid.
- [No Wash]: Skip sample probe cleaning.

[Advanced Settings]

[Advanced Settings]: Place a check mark in the checkbox to enable fine adjustment of the laser output power and detectors. Placing a check mark in the checkbox displays a confirmation message. Click [Yes] to enable the following items.

[Laser Power]: Adjusts the output of each laser.

Adjust the laser output using the slider or enter a value directly.

[Detector (Advanced)]: Adjusts the voltage for each detector channel.

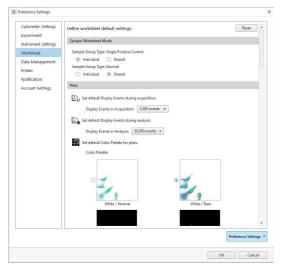
Select the laser axis in [Laser] and adjust the voltages by moving the slider for each channel.



- You can adjust the laser output and detector voltages when the instrument configuration mode is set to [Normal Mode] only.
- It is not recommended to adjust the laser output power and detector voltages separately for each channel in a normal experiment. The output power of each laser and output balance of each detector directly affects the waveform of the spectral reference and the fluorescence unmixing calculations.

• [Worksheet] tab

Defines default settings related to the worksheet.



[Sample Worksheet Mode]

Specifies the worksheet mode for each sample group type.

[Sample Group Type: Single Positive

Control]: Selects the initial worksheet mode for samples when the sample group type is [Single Positive Control].

- [Individual]: Useful when you want to use a unique gate and plot for each sample.
- [Shared]: Useful when you want to use the same gate and plot for samples within a sample group.
- [Sample Group Type: Normal]: Selects the initial worksheet mode for samples when the sample group type is [Normal].
 - [Individual]: Useful when you want to set different gates for different distributions of measurement results for the same sample group.
 - [Shared]: Useful when you want to set a common gate because the distribution of measurements for the same sample group is similar.

[Plots]

- [Display Events in Acquisition]: Selects the number of events to display during data acquisition.
- [Display Events in Analysis]: Selects the number of events to display during data analysis.
- [Color Palette]: Selects the color palette for plots (background color and display of outlying (rare) events).
 - White Background / Normal
 - White Background / Rare Event
 - Black Background / Normal
 - Black Background / Rare Event

[Gate]

[Label Font Size]: Selects the font size of text labels shown on gates. You can check the size in [Preview].

[Default Plots]

Sets the plots added to a new worksheet.

- [Spectrum]: Selects whether to add a spectral plot to a new worksheet.
- **[Scatter]:** Selects the scatter type of plot added to a new worksheet.

[Single Positive Default Plots]

Selects the plot type for single stain Positive control.

[Default Axis Settings for Density/Dot/Contour/ Histogram]

Sets the default axis settings for density plots, dot plots, contour plots, and histogram plots.

- **[FSC Axis Parameters]:** Sets the forward-scatter axis parameters.
 - [Pulse]: Selects the pulse type.
 - [Axis Type]: Selects the default scale type of an axis.
 - [Axis Range]: Sets the axis scale range by specifying the minimum value, negative area, or maximum value according to the axis scale type.

[SSC Axis Parameters]: Sets the side-scatter axis parameters.

- [Pulse]: Selects the pulse type.
- [Axis Type]: Selects the default scale type of an axis.
- [Axis Range]: Sets the axis scale range by specifying the minimum value, negative area, or maximum value according to the axis scale type.

[Fluorescence Axis Parameters]: Sets the fluorochrome data axis parameters.

- [Pulse]: Selects the pulse type.
- [Axis Type]: Selects the default scale type of an axis.
- [Axis Range]: Sets the axis scale range by specifying the minimum value, negative area, or maximum value according to the axis scale type.

[Default Settings for Spectrum / Ribbon Plots]

Sets the default settings for spectrum plots and ribbon plots.

- [X Axis]: Selects the X-axis parameter.
- **[Y Axis]:** Select the Y-axis pulse parameter and scale type.
- [Spectrum Display]: Sets the spectrum display mode.
 - [Display All]: Displays all density data.
 - [Display High Density]: Displays the highincidence spectral information, omitting low-incidence events, to make the display easier to view.

[Default Settings for Contour Plots]

Sets the default settings for contour plots.

- [Method]: Selects the division method.
 - [Probability]: Draw contour lines so that each area between contour lines contains the same number of events.
 - [Linear]: Draws contour lines so that the area between each pair of contour lines contains the equal percentage of the maximum density.
 - [Log]: Draws contour lines so that the area between each pair of contour lines contains a percentage of the maximum density divided logarithmically.
- [Percentage]: Selects percentage division of the contour lines. The larger the number, the larger the distance between contour lines.

[Color Scheme]: Selects the contour colors.

- [Multicolor]: High-density contour lines are shown in red, and low-density contour lines are shown in blue.
- [Black/White]: Display in black and white.
- [Show Outliers]: Place a check mark here to display events outside the outermost contour line as a dot plot.

[Alignment]

Sets whether to display a grid on the worksheet, and whether to enable the position of plots and the statistics table to be quickly aligned to the grid.

- [Auto Arrange]: Automatically aligns the plots and [Gates and Statistics] table on the worksheet.
- [Snap to Grid]: Aligns the plots and [Gates and Statistics] table on the worksheet with the grid when they are moved or added.

[Show Grid]: Displays the grid on the worksheet.

[View]

Defines default settings related to the display. Scale factor: Sets the display size of the worksheet.

• [Data Management] tab

Specifies the export destination folders for various data.

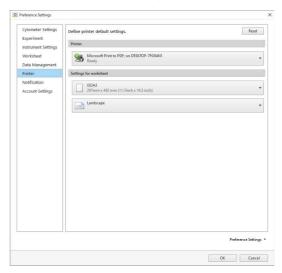
Cytometer Settings	Define data management default settings.		Rese
Experiment Instrument Settings	Default Export Folders		
Worksheet	Default Export Folder for Plate Layout File:	C:\Users\fcm\Desktop	Browse.
Data Management	Default Export Folder for FCS File:	C:\Users\fcm\Desktop	Browse.
Printer	Default Export Folder for Experiment:	C:\Users\fcm\Desktop	Browse.
Notification	Default Export Folder for Spectral Reference:	C:\Users\fcm\Desktop	Browse.
Account Settings	Default Export Folder for Batch Analysis:	C:\Users\fcm\Desktop	Browse

[Default Export Folder for Plate Layout File]: Specifies the destination when exporting the layout to a CSV file (*page 93*).

- [Default Export Folder for FCS File]: Specifies the destination when exporting the layout to an FCS file (*page 97*).
- [Default Export Folder for Experiment]: Specifies the destination when exporting experiment data (*page 118*).
- [Default Export Folder for Spectral Reference]: Specifies the destination when exporting a spectral reference (*page 127*).
- [Default Export Folder for Batch Analysis]: Specifies the output destination for [Export Statistics CSV] (*page 101*).

• [Printer]

Defines settings related to printers.



[Printer]: Specifies the print output destination printer.

[Settings]: Sets the paper size and orientation.

• [Notification] tab

Defines settings related to notifications.

Cytometer Settings Experiment	Define notification d	efault settings.			Reset
xperiment nstrument Settings	Sound Alarm				
Vorksheet Data Management	Repeat Interval: 6	1000			
rinter	Success:	Repeat	*	-	
lotification	Warning:	On	*	•	
ccount Settings	Erron	Repeat	*	•	
	QC				
	Success:	Repeat	¥ 🕨	-	
	Error:	Repeat	•	-	
	Cleaning				
	Success:	Repeat	v	-	
	Error:	Repeat	•		
					Preference Settings *

[Sound Alarm]

Sets an audible notification after data acquisition, QC, or cleaning is completed.

- **[Repeat Interval]:** Sets the repeat interval for emitting the notification sound. You can also enter a value directly from the keyboard.
- [Acquisition]: Selects the notification method when data acquisition is completed, when a warning occurs, or when an error occurs.
- **[QC]:** Selects the notification method when QC is completed or when an error occurs.
- [Cleaning]: Selects the notification method when cleaning is completed or when an error occurs.



• [Account Settings] tab

Configures initial values displayed in the [Add Account] dialog when creating a new user. This items is displayed only when logged in using an administrator account.

Perference Settings: <td

[Password Settings]

Configures settings relating to passwords.

[Password Expiration]: Sets the interval before the password expires in the range 1 to 366 days. The default value is 120 days.

[Lockout Settings]

Configures settings relating to locked accounts.

[Account Lockout]: Sets the number of times that an incorrect password can be entered in succession before the account is locked in the range 1 to 10. The default value is 5 times.

For details about releasing an account lockout, see "When your account is locked (general users)" (page 33).

• [Preference Settings]

You can import the preference settings of another user and export your own preference settings.

[Import]: Imports user data.

[Export]: Exports the current settings as preference settings.

[Institution Information] dialog

The [Institution Information] dialog is displayed by clicking [Institution Information] in the [Information] window.

The [Institution Information] dialog is used to specify information about the institution using the unit.

Institution:	Sony Corporation
Address:	1-7-1 Konan
City:	Minato-ku
State:	Токуо
Zip Code:	108-0075
Country/Region:	Japan

[Institution]: Enter the name of the institution.
[Address]: Enter the street address of the institution.
[City]: Enter the name of the city or town.
[State]: Enter the name of the state or province.
[Zip Code]: Enter the postal code number.

[Account Settings] dialog

The [Account Settings] dialog is displayed by clicking [Account Settings] in the [Information] window. The [Account Settings] dialog is used to configure accounts for those who will use the instrument.

[[]Country / Region]: Enter the name of the country or region.

mame Full Name	Email	Department	Institution	Add
ninistrator Sony jiro	33X\$200.03X	Life Science Division	Sony	Delete
r1 Sony Taro	33X\$X03.33X	Life Science Division	Sony	Edit
r2 Sony Hanako	xxx.xxx@xxx.xxx	Life Science Division	Sony	Ean -

1 User list

Displays the list of users currently registered.

2 User information

Displays information about the user selected in the user list.

3 [Add]

Displays the [Add Account] dialog for adding a new user.

For details, see "Adding Users" (page 27).

4 [Delete]

Deletes the user selected in the user list.

5 [Edit]

Displays the [Edit Account] dialog for editing the account properties for the user selected in the user list.

For details, see "Editing User Settings" (page 28).

6 [Close]

Closes the dialog.

[Print] Window

The [Print] window is displayed by selecting a plot or other object you want to print on the worksheet and clicking [Print] in the menu on the left side.

The [Print] window can print analysis results, statistics, and plots displayed on the worksheet.



Chapter 8 Window Description

1 [Print] button

Sends the print job to the printer.

2 [Print]

Sets the number of copies to print. Place a check mark in the [Use Advanced Settings] checkbox to set advanced printer settings.

3 Print preview

Displays a preview image of the print job.

4 [Printer]

Selects the printer for printing.

5 [Settings]

Sets the paper size and page orientation.

[Database] Window

The [Database] window is displayed by clicking [Database] in the menu on the left side. The [Database] window is used to perform experiment database operations and to check hard disk free capacity.

Example: Administrator account login

	QC Cytometer	
ormation		Hard Disk Drive Capacity
it	Import Import experiments from the exported data files.	1797GB Free Of 1861G
tabase		Database Capacity
lp	Export	9.2GB Free Of 10GB
	Export experiments to the files	
og		
C Logout	Backup	
Exit Exit	Back up the database (Administrator Only).	
	. P	
	3 Restore Restore the database (Administrator Only).	
	e Reset	
	Reset the database (Administrator Only).	

[Import]: Allows you to import experiment data.

Clicking this button displays the [Import Experiment Data] dialog (*page 118*).

You can import experiment data created using another ID7000 for use on this unit.

[Export]: Allows you to export experiment data. Clicking this button displays the [Export Experiment Data] dialog (*page 118*).

You can export experiment data created on this unit for use by another ID7000.

[Backup]: Allows you to back up the database. This button is displayed only when logged in using an administrator account.

Clicking this button displays the [Backup Database] dialog.

Backing up the database allows the data to be restored at a later time, if necessary.

For details, see "Backing Up/Restoring the Database" (page 108).

[Restore]: Allows you to restore a backed up database. This button is displayed only when logged in using an administrator account.

Clicking this button displays the [Restore Database] dialog.

For details, see "Backing Up/Restoring the Database" (page 108).

[Reset]: Allows you to reset the database.

This button is displayed only when logged in using an administrator account.

Clicking this button displays the [Reset Database] dialog (*page 119*).

Note

Resetting the database removes all data, including user accounts, experiments, and analysis results.

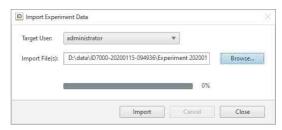
Before resetting the database, it is recommended that the database be backed up.

[Hard Disk Drive Capacity]: Displays the available free space on the hard disk.

[Database Capacity]: Displays the available free space in the database.

[Import Experiment Data] dialog

The [Import Experiment Data] dialog is displayed by clicking [Import] in the [Database] window. The [Import Experiment Data] dialog is used to import data created on another ID7000 into this instrument.



[Target User]: Selects the target user for importing experiment data.

This item is displayed only when logged in using an administrator account.

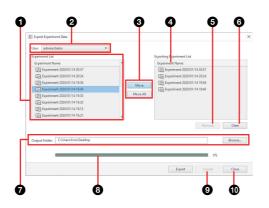
- [Import File(s)]: Specifies the names of files in which the experiment data you want to import is stored.
- **Progress bar:** Displays the import progress.

[Import]: Starts importing.

- [Cancel]: Cancels importing, if in progress.
- [**Close**]: Closes the dialog.

[Export Experiment Data] dialog

The [Export Experiment Data] dialog is displayed by clicking [Export] in the [Database] window. The [Export Experiment Data] dialog is used to export data created on this unit.



1 Experiment list

Displays a list of experiments that satisfy the specified search criteria.

Select the experiment(s) you want to export.

Selecting an experiment in the upper list displays the tubes contained in the experiment in the lower list.

2 User

Selects the owner of the experiments to export.

3 [Move] / [Move All]

Clicking [Move] adds the experiment data selected in the experiment data list to the [Exporting Experiment List]. Clicking [Move All] adds all the experiment data in the experiment data list to the [Exporting Experiment List].

4 [Exporting Experiment List]

Lists the experiments to be exported.

5 [Remove]

Removes the selected experiment from the [Exporting Experiment List].

6 [Clear]

Clears the [Exporting Experiment List].

[Output Folder]

Specifies the export destination folder. Click [Browse] to select the destination folder.

8 Progress bar

Displays the export progress.

9 [Export]

Starts exporting.

① [Close]

Closes the dialog.

[Reset Database] dialog

The [Reset Database] dialog is displayed by clicking [Reset] in the [Database] window. The [Reset Database] dialog is used to delete all users,

experiments, and analysis results, resetting the database.

Notes

- Resetting the database resets all data, including user accounts, experiments, and analysis results. Before resetting the database, it is recommended that the database be backed up.
- After resetting the database, ID7000 Software automatically shuts down.

D Reset Database	×
Reset function deletes all users, experiments and results.	
After reset data, ID7000 Spectral Analyzer will be shutdown a	utomatically.
Reset database	
	Reset Close

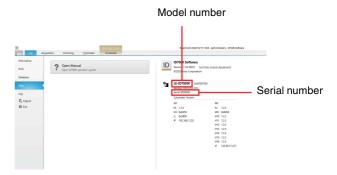
[Reset database]: Place a check mark in the checkbox to enable database reset.

[Reset]: Executes the database reset. **[Close]:** Closes the dialog.

[Help] Window

The [Help] window is displayed by clicking [Help] in the menu on the left side.

The [Help] window is used to display the Operator's Guide (this document), ID7000 Software version information, and the instrument model number, version information, and serial number.



[Open Manual]: Displays the Operator's Guide.

[ID7000 Software]: Displays the ID7000 Software version information, and the instrument model name and version information.

Clicking [End User License Agreement] displays the license agreement.

[Log] Window

The [Log] window is displayed by clicking [Log] in the menu on the left side.

The [Log] window is used to display the following logs.

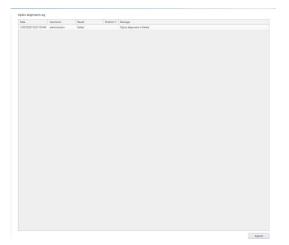


- [Optics Alignment Log]: Displays the optics alignment history.
- [Flow Rate Calibration Log]: Displays the adjusted flow rate history when running [Flow Rate Calibration] and QC.
- [Error and Warning Log]: Displays the error and warning log.

[User Log]: Displays the user usage time log.

Optics Alignment Log

Displays the time, date, name of user, and result for when the optics were aligned.



[Export]: Exports the contents of the log to a CSV-format file.

Clicking this button displays the [Export to CSV File] dialog.

Flow Rate Calibration Log

Displays the time, date, name of user, and result for when the flow rate was calibrated.

[Export]: Exports the contents of the log to a CSVformat file.

Clicking this button displays the [Export to CSV File] dialog.

• Error and Warning Log

Displays the time, date, name of user, category, and other information for when errors and warnings occurred.

Selecting a log entry in the list displays details in the [Description] area.

135 1/	Date	Usemane	Category	Severity	Message
	/20/2020 10:22:38 AM	administrator	Software	Warning	Fale_Bookmark.Resume()Disconnected()) timeout
134 1/	/20/2020 10:22:37 AM	edministrator	Cytometer	free	HandlingInstanceID: ca1613ec-5ce6-45%-b636-fcecf700164cc
233 1/	(20/2020 10:22:37 AM	administrator	Cytometer	Error	HandlingInstanceID: MIDb6eF-ded2-4007-bcHF-e7307c16edcf
122 1/	(20/2020 10.22.37 AA	administrator	Cytometer	Error	HandlingInstanceID: a0xe0485-45e9-4710-b330-65325a745146
191 10	AA 75.52.01 0222/02/	administrator	Cytometer	Information	VultureStatusTransitionToken/Signal#AlError)
130 1/	/20/2020 10:22:37 AM	edministrator	Cytometer	Information	WatureStatus TransitionToken/Signal48(Error)
129 1/	V20/2020 10:22:37 AM	administrator	Cytometer	Information	VultureDatus TransitionFoken.SignalAN(Error)
126 1/	(20/2020 10:22:35 A).	administrator	Cytometer	Information	(LitureStatueTransitionToken.SignalAll(Canceled)
125 V	(20/2220 10:22:25 AA	administrator	Cytometer	Information	VultureStatusTransitionToken.SignalAB[Enor]
120 N	AL 155501 0505/050	administrator	Software	Information	Oreck OpticsAlignment / ControlMoveForwardEactationY
19 10	/20/2020 10:22:31 AA	administrator	Software	Information	Oreck OpticsAlignment / ControlMoveBackEscitationY
118 1/	/20/2020 10:22:31 AM	edministrator.	Software	Information	Check OpticsAlignment : ControlEscitation/WeturnToOrigin
217 1/	(20/2020 10:22:51 AM	administrator	Software	Information	Check OpticsAlignment : Control/VoveForwardEscitationY
10 1)	(20/2220 10:22:31 A&	administrator	Software	Information	Check Opticollignment : ControlNovellackEacitationY
115 V	AL 15:52:01 05:52:05/	administrator	Software	Information	Oreck Optical/Ignment / Control/NoveForwardExcitationY
314 1/	/20/2020 10:22:31 AA	administrator	Cytometer	Information	
13 1/	V20/2020 10:22:30 AM	administrator	Cytometer	Information	VultureStatus TransitionFolien.SignalAII(Error)
112 1/	/20/2020 10:22:50 AM	administrator	Cytometer	trror	HandlingInstanceID: 9918eb09-6d31-418-b751-4d976386d009
211 1/	(20/2220 10:22:30 AA	administrator	Software	Information	Check OpticoAlignment : Control/VoxeForwardEactationY
10 10	AA 06.55.01 0525/05/	administrator	Software	Information	Check Opticallignment - Control/MoveBackEackation/Y
108 1/	AA 06:52.01 02020 AA	administrator	Software	Information	Oreck OpticsAlignment / ControlMoveForwardEscitationY
106 1/	/20/2020 10:21:27 AM	administrator	Software	Information	Check OpticsAlignment : ControlEscitation/WetumToOrigin
205 V	/20/2020 10:21:24 AM	administrator	Software	Information	[CheckWork7ype - EXT_STAGE] NG (AS_WORK_EXISTS: 1h, AS_WORK_HEIGHT: 4E2h)
10 1/	(20/2020 10:17:02 A3		Software	Information	######################################
					Export Delet
					Expert vee
cription					Library Control

- [Category]: Place a check mark in the checkbox for the logs you want to display.
 - [From Cytometer]: Displays logs loaded from the main unit.
 - [From Software]: Displays logs loaded from ID7000 Software.
- **Log list:** Displays the list of log entries loaded from the main unit or ID7000 Software.
- **[Export]:** Exports the contents of the log to a CSV-format file.

Clicking this button displays the [Export to CSV File] dialog.

- **[Delete]:** Deletes the log.
- **[Description]:** Displays information about the log entry selected in the log list.

• User Usage Log

Displays the time when the user logged in/out and the duration of use.

	Full Name	Organization	Login Date	Logout Date	Usage Time		
dministrator			1/15/2020 5/21/23 AM				
dministrator			1/14/2020 7:37:01 PM	1/14/2020 9:58:25 PM	02:01:24		
dministrator			1/14/2020 1.48.06 PM				
iser1			1/14/2020 1/46/56 PM	1/14/2020 147/17 PM	00:00:21		
dministrator			1/14/2020 11:47:27 AM	1/14/2020 1/45/28 PM	01-59-01		
dministrator			1/14/2020 9:54:30 AM	1/14/2020 11:46:26 AM	015156		
uer1			1/14/2020 9:17:16 AM	1/14/2022 9:53:16 AM	00:35:00		
ser1			1/9/2020 6:56:34 PM	1/9/2020 7:16:39 PM	00:20:05		
isert			1/9/2020 644.25 PM	1/9/2020 655:34 PM	00:11:08		
dministrator			1/9/2020 6/29-52 PM	1/9/2020 6:44:05 PM	00:14:14		
ser1			1/9/2020 6/29-06 PM	1/9/2020 6:29:25 PM	00:00:17		
cer1			1/9/2020 6/23/25 PM	1/8/2020 6/28/57 PM	00:05:32		
dministrator			1/9/2020 6(1):11 PM	1/9/2020 6/23/02 PM	00:11:51		
dministrator			1/9/2020 6:08:56 PM	1/9/2020 6:10:01 PM	00/01/05		
iser1			1/8/2020 1:06-37 PM	1/6/2020 1:09:32 PM	00:00:55		
dministrator			12/12/2019 4:11:54 PM	12/12/2019 4:56:28 PM	00:44:34		
dministrator			12/11/2019 2:52:31 PM	12/12/2019 9:22:17 AM	18,29,46		
						Eppert	Dekte

[Export]: Exports the contents of the log to a CSV-format file.

Clicking this button displays the [Export to CSV File] dialog.

[Delete]: Deletes the log.

Тір

Clicking a title cell sorts the log in ascending/descending order.

[QC] Tab

The [QC] tab is displayed by clicking the [QC] workflow tab.

The [QC] tab is used to perform Daily QC and Performance QC.

The [QC] tab has the following menu items.

- [Daily QC] (page 121)
- [Performance QC] (page 122)

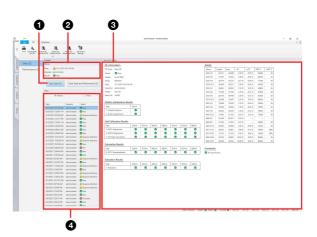
The ribbon has the following tabs.

- [QC] tab (page 123)
- [Cytometer] tab (page 124)

[Daily QC] Window

The Daily QC window is displayed by clicking [Daily QC] in the menu on the left side. The [Daily QC] window is used to run Daily QC and to check QC results.

For details about Daily QC, see "Running Daily QC" (page 34).



[Start Daily QC]

Starts the Daily QC wizard.

2 [Start Daily and Performance QC]

Displays the wizard for running Daily QC and Performance QC consecutively.

3 [Status]

Displays summary information of the most recent Daily QC check.

[Date]: Displays the date and time of Daily QC.

[Operator]: Displays the name of the operator who ran Daily QC.

[Result]: Displays the results of Daily QC.

4 [Daily QC History]

Displays the results for the specified Daily QC.

5 [View]

Selects the content to display in 4 [Daily QC History].

[History]: Select this option when you want to display results for each Daily QC check.

When selected, a list of previously run Daily QC checks is displayed. Selecting a Daily QC item displays the QC results on the worksheet.

liew			
 History 		◯ Trend	
Date	Operator	Result	
10/14/2020 2:58:42 PN	administrator	👽 Pass	4
10/14/2020 2:45:23 PN	administrator	1 Requires Attention	
10/14/2020 2:42:51 PN	administrator	😣 Canceled	
10/14/2020 2:31:53 PN	administrator	😣 Fail	
10/14/2020 2:08:21 PN	administrator	Pass	
10/14/2020 11:49:44 A	administrator	Pass	
10/14/2020 7:52:05 AN	administrator	Pass	
10/14/2020 7:43:14 AM	administrator	Pass	

[Trend]: Select this option when you want to display the changes in Daily QC results over time.

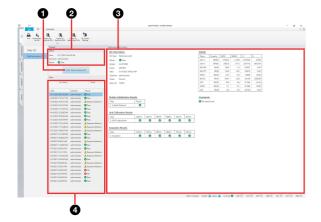
To change the display period and parameters, click [Change Conditions] and configure settings in the *"[Change Conditions] dialog" (page 122)*.

C	History	 Trend 	
		Change Conditions	
Period	Last 3 months		
Parameters	FSC		
	SSC		
Statistics	Height,Area,rCV		
PMT Voltage			

[Performance QC] Window

The [Performance QC] window is displayed by clicking [Performance QC] in the menu on the left side. The [Performance QC] window is used to run Performance QC and to check QC results.

For details about Performance QC, see "Running Performance QC" (page 36).



1 [Start Performance QC]

Starts the [Performance QC] wizard.

2 [Status]

Displays summary information of the most recent Performance QC event.

- [Date]: Displays the date and time of Performance QC event.
- **[Operator]:** Displays the name of the operator who ran Performance QC.

[Result]: Displays the results of Performance QC.

③ [Performance QC Report]

Displays the results for the specified Performance QC.

4 [View]

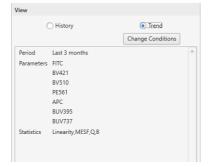
Selects the content to display in **3** [Performance QC Report].

[History]: Select this option when you want to display results for each Performance QC check. When selected, a list of previously run Performance

QC events is displayed. Selecting a Performance QC item displays the QC results on the worksheet.

 History 		◯ Trend
Date	Operator	Result
10/14/2020 3:06:46 PN	administrator	👽 Pass
10/14/2020 2:54:02 PN	administrator	1 Requires Attention
10/14/2020 2:43:44 PN	administrator	🔇 Canceled
10/14/2020 2:19:25 PN	administrator	Pass
10/14/2020 11:58:01 A	administrator	Pass
10/14/2020 8:09:16 AM	administrator	👽 Pass
10/14/2020 8:05:28 AM	administrator	👽 Pass
10/14/2020 8:00:21 AM	administrator	🔀 Fail

[Trend]: Select this option when you want to display the changes in Performance QC results over time. To change the display period and parameters, click [Change Conditions] and configure settings in the *"[Change Conditions] dialog" (page 122).*



[Change Conditions] dialog

The [Change Conditions] dialog is displayed by clicking [Change Conditions] in [Trend] on the [View] pane. You can select the QC results interval and the target parameters in the [Change Conditions] dialog.

Change Condition Period • Last 3 months					×	
🔘 Last 6 months						
Custom	1	im	To:			
Parameters						
FITC						
V421						
V510						
VE561						
APC						
BUV395						
BUV737						
Statistics						Ĺ
🔽 Linearity 🛛	MESF [V Q	🗸 B			
Restore Defaults		OK		Cance	J	

• [Period]

Selects the interval for the QC results you want to display.

When [Custom] is selected, specify the start and end dates to display.

2 [Parameters]

Place a check mark in the checkboxes for the parameters to display. You can select multiple parameters.

3 [Statistics]

Place a check mark in the checkboxes for the statistics to display. You can select multiple statistics.

4 [Restore Defaults]

Restores the display items to the default values.

6 [OK]

Saves the settings and closes the dialog.

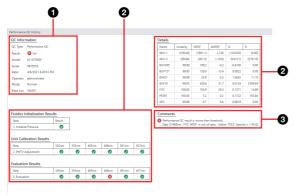
6 [Cancel]

Cancels the settings and closes the dialog.

Checking QC results

The following information is displayed in [Performance QC Report].

When [History] is selected in [View]



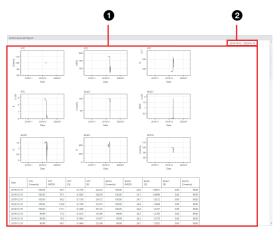
1 Summary information

2 Statistics for all detected channels

You can show/hide the table using [Show Table] on the [QC] tab of the ribbon.

3 Details when QC check fails

When [Trend] is selected in [View]



1 Time charts for statistics in the specified period

2 Display period

[QC] Tab (Ribbon)

The [QC] tab of the ribbon has the following buttons.



[QC] group

These buttons are used to perform operations on QC results.

[Print]: Prints QC results.

Clicking this button displays the [Print] window *(page 117)*.

- [Import Bead Lot File]: Imports the beads lot file for Align Check beads.
- [Export QC Results to CSV]: Exports QC results as a CSV-format file.
- [Export QC Results to XML]: Exports QC results as an XML-format file.
- [Import QC Results from XML]: Imports the values for each channel saved in an XML-format file.
- [QC Criteria Settings]: Sets QC criteria.

For details, see "Setting QC Criteria" (page 104).

[Cytometer] Tab (Ribbon)

The [Cytometer] tab of the ribbon has the following buttons.



[Cleaning] group

These buttons are used to run manual cleaning, set cleaning times, and other cleaning operations.

[**Priming]:** Primes the fluidics lines by backflushing with sheath fluid.

This takes approximately nine minutes. During priming, a progress bar and status are displayed, allowing you to check the progress and current state.

- [Probe Wash]: Starts cleaning of the probe. This takes approximately 15 seconds. During cleaning, the status is displayed, allowing you to check the current state.
- [Flow Cell Purge]: Backflushes a large volume of sheath fluid through the flow cell. This takes approximately four minutes.

During cleaning, a progress bar and status are displayed, allowing you to check the progress and current state.

[Decontamination]: Starts cleaning with bleach.

- [Bleach Cleaning]: Starts cleaning of the sample line, sample probe, and flow cell using a sodium hypochlorite (bleach) solution. This takes approximately eight minutes. During cleaning, a progress bar and status are displayed, allowing you to check the progress and current state.
- [Bleach Cleaning and Rinse]: Starts cleaning of the sample line, sample probe, and flow cell using a sodium hypochlorite (bleach) solution, then rinses the fluidics system using rinse fluid. This takes approximately nine minutes. During cleaning, a progress bar and status are displayed, allowing you to check the progress and current state.

[De-bubble] group

This button is used to run the de-bubble process.

[Sheath Line]: De-bubbles the sheath line. Clicking this button displays the [Sheath Line De-bubble Wizard] wizard. Follow the on-screen instructions.

[Calibration] group

These buttons are used to calibrate the Instrument.

[Optics Alignment]: Automatically aligns the laser excitation position for the flow cell.

[Flow Rate]: Adjusts the fluidics system pressure to achieve the optimum sheath fluid flow rate.

For details, see "Calibrating the Instrument" (*page* 87).

[Plate] group

This button is used for operations on plates.

[Agitate]: Agitates the sample.

[Cooling]: Cools the sample holder to a temperature of 4 °C (39 °F).

[Tank Maintenance] group

These buttons are used for 10L tank system maintenance.

[Refill and Empty]: Used to refill the 10L sheath tank and replace the 10L waste tank. Clicking this button displays the [Tank Maintenance Wizard]. Follow the on-screen instructions.

For details, see "Refilling the 10L Sheath Tank with Sheath Fluid" (page 154) and "Emptying/Replacing the 10L Waste Tank" (page 153).

[Long Term Storage]: If PBS (saline) is used as the sheath fluid, this performs maintenance before shutting down if you plan not to use the instrument for a week or longer.

For details, see "Running the [Long Term Storage] Process" (page 158).

[Status] group

This button is used to monitor instrument status.

[Cytometer Status]: Monitors the instrument status. Clicking this button displays the [Cytometer Status] dialog (*page 124*).

[Cytometer Status] dialog

The [Cytometer Status] dialog is displayed by clicking [Cytometer Status] on the [Cytometer] tab of the ribbon. Selecting an item from the tabs on the top displays the status of that item.

• [Laser]

Displays the status of the lasers. The installed lasers vary depending on the model.

aser Detector 1		Mech System					
	320 nm	355 nm	405 nm	488 nm	561 nm	637 nm	808 nm
Unit Detection :	On	On	On	On	On	On	On
Lighting :	Off	Off	Off	Off	Off	Off	Off
Init :	Ready	Ready	Ready	Ready	Ready	Ready	Ready
Power (mW) :	32.07	35.55	40.52	48.81	56.14	63.73	80.86
Current (mA) :	3200	3550	4050	4880	5610	6370	8080
Hour Meter (hour) :	100	100	100	100	100	100	100

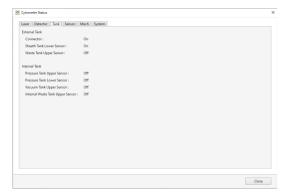
• [Detector]

Displays the status of the detector.

ser Detector. Tan	k Sensor M	ech System			
>					
FSC Gain :	Level5				
ит					
SSC HV :	Oh	405 nm VB1 HV :	Oh		
320 nm UV1 HV :	Oh	405 nm VB2 HV :	Oh		
320 nm VB1 HV :	Oh	405nm VB3 HV :	Oh		
320 nm VB2 HV :	Oh	405 nm 32ch HV :	Oh		
320 nm 32ch HV :	Oh	488 nm 32ch HV :	Oh		
355 nm UV1 HV :	Oh	561 nm 32ch HV :	Oh		
355 nm VB1 HV :	Oh	637nm 32ch HV :	0h		
355 nm VB2 HV :	Oh	808nm IR1 HV :	0h		
355 nm 32ch HV :	Oh	808nm IR2 HV :	Oh		

• [Tank]

Displays the status of the tanks.



• [Sensor]

Displays the status of the various sensors.

Laser Detector Tank	Sensor	Mech System												
Notch Filter		Laser Shutter					Valve							
561 nm (IN) :	Off	Shutter (OPEN) :	Off				1:	Off	4:	Off	7:	Off		
561 nm (OUT) :	On	Shutter (CLOSE) :	On				2:	Off	5:	Off	8:	Off		
637 nm (IN) :	Off						3:	Off	6:	Off	9:	Off		
637 nm (OUT) :	On	Optics Original Position												
808 nm (IN) :	Off	Y-Axis	Off				Door							
808 nm (OUT) :	On	Z-Axis	Off				Inte	nlock :	Of	f)				
Temperature		Pressure					Pump	Duty						
Laser (*C) :	0.0	Pressure Tank (kPa) :	0.00	7	0.00	0	LP1	(%);	0.0					
Ruidics (*C):	0.0	Vacuum Tank (kPa) :	0.00	2	0.00	0	LP2	(%):	0.0					
Power Supply (*C) :	0.0	Sample (kPa) :	0.00	7	0.00	0	LP3	(%):	0.0					
320 nm PMT (*C) :	32.0	Washing Unit (kPa) :	0.00				LP4	(%):	0.0					
355 nm PMT (*C) :	35.5	Notch Shutter Tank (kPa)	179.88	(0	ff)		AP1	(%):	0.0					
405 nm PMT (*C) :	40.5						AP2	(%):	0.0					
488 nm PMT (*C) :	48.8													
561 nm PMT (*C) :	56.1													
637 nm PMT (*C) :	63.7													
Sample Stage (*C) :	0.0	0												

• [Mech] (mechanism)

Displays the status of the loader and other mechanisms.

ample Loader X-Axis		Washing Unit		
Origin :	Off	Origin :	Dff	
Limit (CW) :	Off	Limit (CW) :		
Limit (CCW):	Off	Limit (CCW) :	Dff	
Probe Detection :	Off			
ample Loader Y-Axis				
Origin :	Off			
Limit (CW) :	Off			
Limit (CCW):	Off			
Probe Detection :	Off			
ample Loader Z-Axis				
Origin :	Off			
Limit (CW) :	Off			
Limit (CCW):	Off			
Work Height :	Off			
Probe Collision :	On			

• [System]

Displays the system status.

Cytometer Status	
Laser Detector Tank Sensor Mech System	
Cytometer Status	
Status : Eject	
Sheath Setting	
Sheath Fluid : DI Water	

[Power] group

These buttons are used to shut down the instrument.

- [Automatic Shutdown]: Shuts down the main unit and turns off the power supply automatically after acquisition and tube cleaning finishes.
- [Hardware Shutdown]: Shuts down the main unit and turns off the power supply.

You can also run cleaning processes before the main unit shuts down. Click this button and select the cleaning to perform from the drop-down menu.



- [No Cleaning]: Shuts down the main unit without performing cleaning.
- **[Bleach Cleaning]:** Performs cleaning using a sodium hypochlorite (bleach) solution, followed by flushing using sheath fluid, and then shuts down the main unit.
- **[Bleach Cleaning and Rinse]:** Performs cleaning using a sodium hypochlorite (bleach) solution and rinsing using rinsing fluid, followed by flushing using sheath fluid, and then shuts down the main unit.

[Experiment] Tab

The [Experiment] tab is displayed by clicking the [Experiment] workflow tab.

The [Experiment] tab is used to create/register a spectral reference, and to create an experiment.

The [Experiment] tab has the following menu items.

- [Experiment Designer] (page 126)
- [Experiment Template] (page 126)
- [Reopen Experiment] (page 127)

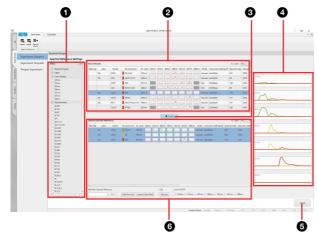
The ribbon has the following tabs.

- [Experiment] tab (page 127)
- [Cytometer] tab (page 124)

[Experiment Designer] Window

The [Experiment Designer] window is displayed by clicking [Experiment Designer] in the menu on the left side.

The [Experiment Designer] window is used to create a new experiment using the wizard.



[Filters]

Filters the search for a spectral reference.

2 [Search Results]

Displays the spectral reference search results in list view.

3 [Select]

Clicking this button selects a spectral reference used for acquisition from the [Search Results] list.

4 Waveform display

Displays a magnified view of the waveforms for the selected spectral reference.

5 [Next]

Displays the [Experiment Designer] wizard.

6 [Selected Spectral References]

Displays a list of spectral references to use for acquisition.

You can create a spectral reference and register it in the library.

To remove a spectral reference from the list, select the target spectral reference row and click [Remove].

For details about creating and registering a spectral reference, see "Registering a Spectral Reference" (page 43).

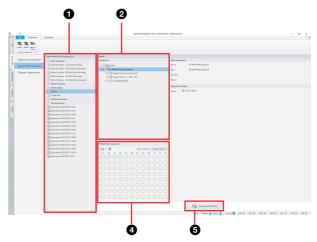
[Experiment Template] Window

The [Experiment Template] window is displayed by clicking [Experiment Template] in the menu on the left side.

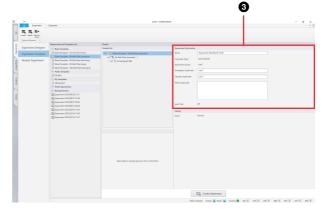
The [Experiment Template] window is used to create a new experiment using templates.

For details about creating an experiment, see "Creating a New Experiment using the Wizard" (page 47).

When a sample group is selected in the [Sample List] panel



When an experiment is selected in the [Sample List] panel



1 [Experiments and Templates List]

Displays a list of templates, and experiments that can be used as templates.

Click \bigcirc / \bigcirc to show/hide the list.

[Blank Templates]

Displays blank templates for each type of plate.

[Public Templates]

Displays templates registered as shared public templates.

[My Templates]

Displays private templates registered by the user.

[Public Experiments]

Displays shared experiments. These experiments can be used as templates.

[My Experiments]

Displays experiments created by the logged in user. These experiments are private, and cannot be used by other users.

2 [Sample List]

Displays the structure of the experiment. Place a check mark in the corresponding checkbox to enable the use of a plate, sample group, or well in the experiment.

Selecting a plate, sample group, or well will display detailed settings for that item on the right.

③ [Experiment Information]

Enter information about the experiment, as required.

4 [Plate] panel

Displays the wells/tubes contained in the plate selected in the [Sample List] panel in list view. You can switch the display to list view or map view.

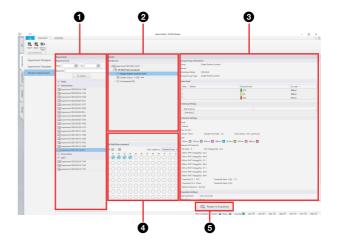
5 [Create Experiment]

Creates a new experiment with the specified settings. Clicking this button displays the [Acquisition] workflow tab (*page 128*).

[Reopen Experiment] Window

The [Reopen Experiment] window is displayed by clicking [Reopen Experiment] in the menu on the left side.

The [Reopen Experiment] window is used to open an existing experiment, reacquire data, add plates, and other operations.



1 [Experiment List]

Displays the list of previous experiments. You can search experiments by date and keyword. Select the experiments you want to edit in this list.

2 [Sample List] panel

Displays the structure of the experiment in list view. Selecting a plate, sample group, or well will display detailed settings for that item on the right.

3 [Experiment Information]

Information about the item selected in the [Sample List] panel is displayed.

4 [Plate] panel

Displays the wells contained in the plate selected in the [Sample List] panel in list view.

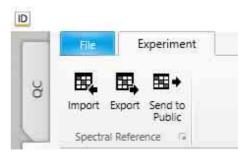
You can switch the display to list view or map view.

(Open in Acquisition)

Opens the selected experiment. Clicking this button displays the [Acquisition] workflow tab (*page 128*).

[Experiment] Tab (Ribbon)

The [Experiment] tab of the ribbon has the following buttons.



[Spectral Reference] group

[Import]: Imports spectral reference data saved in a CSV-format file.

[Export]: Exports the spectral reference to a CSV-format file.

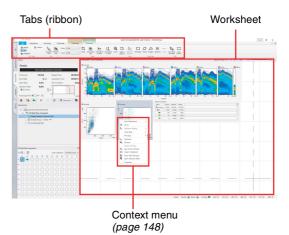
- **[Send to Public]:** Shares the selected spectral reference as a public spectral reference. Clicking this button registers the selected spectral reference as a shared spectral reference.
- Displays the [Properties] dialog for the selected spectral reference.

[Acquisition] Tab

The [Acquisition] tab is displayed by clicking the [Acquisition] workflow tab.

The [Acquisition] tab is used to configure template settings and to perform data acquisition.

The [Acquisition] tab has the following structure.



The ribbon has the following tabs (tabs vary depending on the current selection).

- [Acquisition] tab (page 128)
- [Unmixing] tab (*page 132*)
- [Cytometer] tab (page 124)
- [Worksheet Tools] tab (page 135)
- [Plot Tools] tab (page 138)
- [Gate Tools] tab (page 141)

Tip

The [Cytometer] tab has the same ribbon as the [QC] tab.

[Acquisition] Tab (Ribbon)

The [Acquisition] tab of the ribbon has the following buttons.



[Acquire] group

[Set Current Position]: Assigns the target well for preview and data acquisition.

[Auto Acquire] group

[Add to Target]: Selects the target wells for automatic acquisition.

[Experiment] group

These buttons are used to perform operations related to experiments.

[New]: Creates a new experiment.

Clicking this button saves and closes the currently open experiment, and switches the display to the [Create Experiment] window.

For details, see "[Experiment Template] Window" (page 126).

[Close]: Closes the experiment.

Clicking this button saves the currently open experiment, and switches the display to the [Analysis] workflow tab.

[Save as Template]: Saves the experiment selected in the [Experiment] panel as a template. Clicking this button displays the [Save as Template]

dialog.

For details, see "Saving an Experiment as a Template" (page 94).

Solution: Displays the [Properties] dialog for the selected experiment.

[Plate] group

These buttons are used to add and delete plates in an experiment.

[New]: Adds an empty plate to an experiment.

- [Delete]: Deletes the plate selected in the [Experiment] panel.
- [Load Template]: Applies template settings to the plate selected in the [Experiment] panel.
- [Layout Editor]: Tool used for editing metadata. Clicking this button displays the plate layout editor dialog (*page 129*).
- **[Export Layout to CSV File]:** Exports the metadata for the plate or sample group selected in the experiment list as a CSV-format file. Clicking this button displays the [Export to CSV File] dialog (*page 93*).

[Import Layout from CSV File]: Imports the metadata for the plate or sample group selected in the experiment list.

Clicking this button displays the [Import Layout from CSV File] dialog (*page 93*).

□ Displays the [Plate Properties] dialog (*page 129*) for the selected plate.

Plate Properties dialog

The plate properties dialog is used to edit plate information and to configure settings for data acquisition.

late Information	96 Well Plate (standard)	
Name:	96 Well Plate (standard) - 1	
Date Created:	10/16/2020 10:14:00 AM	
Number (optional):		
Memo (optional):		
cquisition Setting		-

1 [Plate Information]

You can edit information about a plate.
[Type]: Displays the type of plate.
[Name]: Enter the name of the plate.
[Date Created]: Displays the creation date.
[Number (optional)]: Enter the plate number, as required.

[Memo (optional)]: Enter a comment, as required.

2 [Acquisition settings]

Configures settings for data acquisition. [**Order**]: Select the well acquisition sequence.

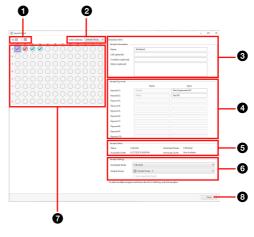
③ [OK]

Saves the settings and closes the dialog.

4 [Cancel]

Cancels the settings and closes the dialog.

Plate layout editor dialog



1 View

You can switch the display to list view or map view.

[Color coded by]

You can sort wells by color for selected metadata.

- Sample Group
- Cell
- Condition

3 [Sample Information]

You can edit information about a sample.

[Name]: Enter the name of the well.

[Cell (optional)]: Enter information about the cell, as required.

[Condition (optional)]: Enter the condition of the sample, as required.

[Memo (optional)]: Enter a comment, as required.

4 [Sample Keywords]

You can edit the sample keywords for the keyword names set in an experiment.

[Name]: Displays the keyword names configured in the experiment.

[Value]: Enter the value of a sample keyword.

5 [Sample Status]

Displays the acquisition status of the sample.
[Status]: Displays the acquisition status of the sample.
[Acquisition Date]: Displays the date of acquisition.
[Worksheet Mode]: Displays the worksheet mode.
[Unmixing Cache]: Indicates whether spectral unmixing data exists.

6 [Sample Settings]

Displays settings relating to sample acquisition.

- [Worksheet Mode]: Selects the worksheet mode for the selected sample.
- **[Sample Group]:** Selects the sample group to which the sample belongs.
- [Auto acquisition target]: Place a check mark in the checkbox to the set as the auto acquisition target.

7 Wells

Displays the status each well using icons.

- : Data has not been acquired yet.
- 🕃: Data acquisition is in progress.
- S: Data acquisition has been completed.
- (): Target well for preview or data acquisition.
- : Target well for auto acquisition.
- Well in which the event rate has dropped or air bubbles are detected.

The shape of the icon will vary depending on the worksheet mode.

- (circle): [Shared] mode
 - (square): [Individual] mode

8 [Close]

Closes the dialog.

[Sample Group] group

These buttons are used to create, delete, and perform other actions related to sample groups.

[New]: Creates a new sample group.

Clicking this button creates a new sample group in the plate selected in the [Experiment] panel.

[Delete]: Deletes the sample group selected in the [Experiment] panel.

- **[Duplicate]:** Duplicates the sample group selected in the [Experiment] panel.
- [Import Instrument Settings]: Imports instrument settings.
- **Displays the [Properties] dialog for the selected** sample group.

Sample group properties dialog

The sample group properties dialog is used to configure metadata for a sample group.

ample Group Informati	on		
Name:	Sample Group - 1		
Date Created:	8/27/2022 9:13:01 PM		
Memo (optional):			
ample Group Settings			
ample Group Settings Sample Group Type:	 Single Positive Control 	Newsl	
sample Group type:	Single Positive Control	Normal	

1 [Sample Group Information]

You can Edit information about a sample group. [Name]: Enter the name of the sample group. [Date Created]: Displays the creation date. [Memo (optional)]: Enter a comment, as required.

2 [Sample Group Settings]

Configures settings for a sample group. [Sample Group Type]: Selects the sample group type.

3 [OK]

Saves the settings and closes the dialog.

4 [Cancel]

Cancels the settings and closes the dialog.

[Sample] group

These buttons are used for operations on wells.

[Copy] / [Paste]: Copies/pastes the selected well.[Move to Sample Group]: Moves the selected well to another sample group.

[Remove from Sample Group]: Removes the selected well from a sample group.

[Export Instrument Settings]: Exports instrument settings to a file.

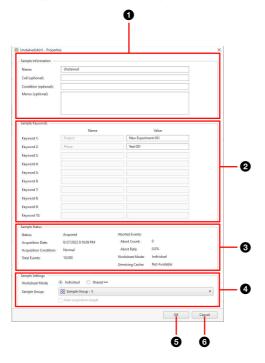
[Instrument Settings]: Configures settings related to the lasers, detector, and flow rate. Clicking this button displays the [Instrument Settings] dialog.

For details, see "Configuring Instrument Settings" (page 59).

[Stopping Condition]: Specifies the data acquisition stop condition. Clicking this button displays the [Stopping Condition] dialog. Displays the [Properties] dialog for the selected well.

Well Properties dialog

The Well Properties dialog is used to configure the sample group, targets for auto acquisition, and metadata.



() [Sample Information]

You can edit information about a sample.

[Name]: Enter the name of the well.

[Cell (optional)]: Enter information about the cell, as required.

[Condition (optional)]: Enter the condition of the sample, as required.

[Memo (optional)]: Enter a comment, as required.

2 [Sample Keywords]

You can edit the sample keywords for the keyword names set in an experiment.

[Name]: Displays the keyword names configured in the experiment.

[Value]: Enter the value of a sample keyword.

3 [Sample Status]

Displays the acquisition status of the sample.

[Status]: Displays the acquisition status of the sample. [Acquisition Date]: Displays the date of acquisition. [Acquisition Condition]: Displays the acquisition

stop condition.

- [Normal]: Indicates acquisition ended successfully.
- [Low Event Rate]: A low event rate or reducing event rate was detected.
- [Air]: Air bubbles have been detected.
- [Elapsed Time Limit]: The upper limit of data acquisition time has been reached.

[Total Events]: Displays the number of events.

[Aborted Events]: Displays the number of aborted events and their percentage.

[Worksheet Mode]: Displays the worksheet mode.

[Unmixing Cache]: Indicates whether spectral unmixing data exists.

4 [Sample Settings]

Displays settings relating to sample acquisition. [Worksheet Mode]: Selects the worksheet mode. [Sample Group]: Selects the sample group to which the sample belongs.

[Auto acquisition target]: Place a check mark in the checkbox to the set as the auto acquisition target.

6 [OK]

Saves the settings and closes the dialog.

6 [Cancel]

Cancels the settings and closes the dialog.

[Stopping Condition] dialog

Total Events		10,000		
Acquisition Tin	ne v	00:02:00		
Gate Level Stoppin	g Conditio	on		
AND T				
✓ #1 A		10,000		events
😿 #2 🔲 B	Ŧ	10,000		events
11=3	Ϋ́		Ŧ	events
turation Gate All events •	tion			

[Stopping Condition]

Specifies the data acquisition stop condition. Place a check mark in the checkbox to enable the stop condition.

When multiple conditions are specified, acquisition stops when any one of the conditions is satisfied.

Stop condition: Selects the data acquisition stop condition and its value. You can also enter a value directly from the keyboard.



- **[Total Events]:** Stops when the number of events reaches the specified value.
- **[Elapsed Time]:** Acquisition stops when the accumulated elapsed time since the start of data acquisition reaches the specified value.
- [Acquisition Time]: Acquisition stops when the data acquisition time reaches the specified value.

[Gate Level Stopping Condition]: Specifies the number of events before stopping for each gate. Place a check mark against the target gate or [All events], and select the number of events before stopping. You can also enter a value directly from the keyboard.

Data acquisition stops when the number of events detected reaches the specified value.

When conditions are specified for multiple gates, you can specify an AND or OR condition. When AND is used, acquisition stops when the specified conditions for all gates are satisfied. When OR is used, acquisition stops when the specified conditions for any one gate are satisfied.

- [Saturation Gate]: Specifies the target gate to monitor events for which to detect saturation status during data acquisition.
- [Sync Stopping Condition]: Sets the data acquisition stop conditions and saturation gate event monitoring configured in the dialog for other unacquired samples within the same sample group.

This function is enabled in [Individual] worksheet mode.

[Batch Processing] group

These buttons are used to run spectral unmixing and analysis on multiple samples at the same time.

[Batch Unmixing]: Runs batch spectral unmixing for selected experiments, plates, and sample groups. Clicking this button displays the [Batch Unmixing] dialog.

For details, see "Running Batch Spectral Unmixing" (page 100).

[Batch Analysis]: Runs batch analysis for selected experiments, plates, and sample groups.

Clicking this button displays the [Batch Analysis] dialog.

For details, see "Running Batch Analysis" (page 100).

[Export to FCS File]: Exports acquisition data for the selected experiments, plates, and sample groups as an FCS file.

Clicking this button displays the [FCS File Export] dialog.

For details, see "Exporting an FCS File" (page 97).

[Record] group

These buttons are used for records and electronic signatures.

For details about operation, see "Creating a Record with Electronic Signature" (page 105).

[Sign Record]: Creates a record in PDF format and then electronically signs the file.

[Export Record]: Exports a record. Clicking this button displays the [Export Record] dialog.

[Import Record]: Imports an exported record into the unit. Clicking this button displays the [Import Record] dialog.

[View Audit Trail]: Displays the audit trail.

[Display Events] group

Sets the number of events that are displayed on plots.

[Acquisition]: Selects the number of events to display during data acquisition.

Acquisition:	5,000 events	2
·	500 events	
Analysis:	1,000 events	
1	5,000 events	
	10,000 events	
	100,000 events	
	1 sec	
	2 sec	
	5 sec	
	10 sec	

[Analysis]: Selects the number of events to display during analysis.

Acquisition:	5,000 events *
Analysis:	50,000 events *
0	10,000 events
	50,000 events
	100,000 events
	500,000 events
	1,000,000 events
	5,000,000 events
	10,000,000 events
	16,000,000 events
	Full events

[Unmixing] Tab (Ribbon)

The [Unmixing] tab of the ribbon has the following buttons.



[Unmixing] group

These buttons are used to calculate the spectral unmixing matrix and to apply the matrix.

[Matrix]: Selects the spectral unmixing matrix.

[Use common matrix]: Place a check mark in the checkbox to apply the spectral unmixing matrix to all the samples in the sample group.

[Calculate Matrix]: Calculates the spectral unmixing matrix.

[Apply]: Selects whether spectral unmixing is applied to plots on the worksheet.

Clicking this button toggles unmixing on/off. This setting is reflected on the unmixing settings.

[Unmixing Settings]: Used to adjust the spectral unmixing.

Clicking this button displays the [Unmixing Settings] dialog (*page 133*).

[Spectral Reference] group

These buttons are used to adjust spectral references and register spectral references in the library.

- [Spectral Reference Adjuster]: Enables manual adjustment of spectral unmixing by adjusting the spectral reference of the selected matrix. Clicking this button displays the [Spectral Reference Adjuster] dialog (*page 81*).
- [Unmixing Viewer]: Displays the results of spectral unmixing in a list of plots for each fluorochrome combination.

Clicking this button displays the [Unmixing Viewer] dialog.

For details, see "Checking Spectral Unmixing Results Summary" (page 82).

[Load Spectral Reference]: Loads the spectral

reference from the spectral reference library into the selected matrix.

Clicking this button displays the [Load Spectral Reference] dialog (*page 134*).

[Add to Library]: Registers the spectral reference in the library.

Clicking this button displays the [Add to Library] dialog.

[Autofluorescence]

This button is used to find autofluorescence populations.

[Autofluorescence Finder]: Finds the

autofluorescence spectrum in acquisition data by searching for the autofluorescence populations. Clicking this button displays the [Autofluorescence Finder] dialog (*page 83*).

[Fluorochrome]

This button is used to manage the fluorochrome database.

[Fluorochrome Database]: Edits the fluorochrome database.

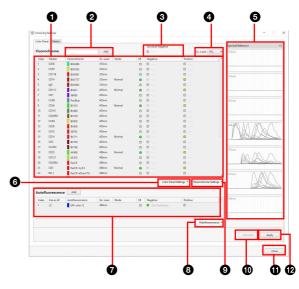
Clicking this button displays the [Fluorochrome Database] dialog (*page 135*).

[Unmixing Settings] dialog

The [Unmixing Settings] dialog is displayed by clicking [Unmixing Settings] on the [Unmixing] tab of the ribbon. The [Unmixing Settings] is used to configure and adjust spectral unmixing.

• [Color Panel] tab

Used for editing the fluorochrome list and creating spectral references.



1 Fluorochrome list

For details, see "Creating a Spectral Reference" (page 76).

2 [Add]

Adds a fluorochrome to the fluorochrome list.

3 [Universal Negative]

Assigning a gate to Universal Negative and then setting the Negative of each fluorochrome to Universal Negative eliminates the need for individual Negative gate settings.

• [Ex. Laser]

Specify a laser to filter the fluorochromes displayed in the list.

5 Waveform display

Displays a magnified view of the waveforms for the spectral reference selected in the fluorochrome list.

6 [Color Panel Settings]

Displays the following menu items. [Import]: Imports saved fluorochrome list data. [Export]: Exports fluorochrome list data as a CSV file.

7 [Autofluorescence]

Use to edit the autofluorescence list. [Add]: Adds an autofluorescence color to the autofluorescence list.

8 [Autofluorescence]

Displays the following menu items.

[Import]: Imports the autofluorescence spectral reference data from a file.

Selecting this menu item displays the [Import Spectral Reference] dialog.

[Export]: Exports the autofluorescence spectral reference data to a file.

9 [Fluorochrome Settings]

Displays the following menu items.

[Load from Library]: Loads a spectral reference from the library.

Selecting this menu item displays the [Load Spectral Reference] dialog.

[Add to Library]: Registers the spectral reference, with check mark in the fluorochrome list, in the library. Selecting this menu item displays the [Add to Library] dialog.

① [Calculate]

Calculates the spectral unmixing matrix.

(Close]

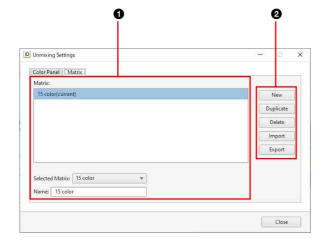
Closes the dialog.

(Apply)

Selects whether spectral unmixing is applied to plots on the worksheet.

• [Matrix] tab

Use to edit the spectral unmixing matrix.



1 [Matrix] list

Displays the list of registered matrices. You can also rename matrices, as required.

[Selected Matrix]: Selects the matrix. [Name]: Enter the name of the matrix.

2 Editing buttons

[New]: Adds a new matrix to the [Matrix] list. [Duplicate]: Duplicates the selected matrix in the [Matrix] list.

[Delete]: Deletes the selected matrix in the [Matrix] list. [Import]: Imports a matrix saved in a CSV-format file.

[Export]: Exports the selected matrix to a CSV-format file. The spectral references, spectral unmixing matrix, and other information are saved in the CSV file.

[Calculate], [Apply], and [Close] are common to all tabs in the dialog. See "[Color Panel] tab" (page 133).

[Load Spectral Reference] dialog

The [Load Spectral Reference] dialog is displayed by clicking [Load Spectral Reference] on the [Unmixing] tab of the ribbon.

The [Load Spectral Reference] dialog is used to load an existing spectral reference from another experiment.

For details, see "Loading an Existing Spectral Reference" (page 80).



1 Search area

Specify keywords and filter criteria to search for a spectral reference.

[Keyword Search]: Searches by keyword.

- Enter a fluorochrome name or marker, then click [Search] to start searching.
- [Filters]: Filters the search using search conditions with check marks.

Tip

Clicking [Clear] removes the check marks from all checkboxes.

2 [Search Results]

Displays the search results. Select the spectral reference to load from the list.

3 [Select]

Adds the spectral reference selected in [Search Results] to [Selected Spectral References].

4 [Selected Spectral References]

Lists the spectral references to load. Selecting a spectral reference displays the spectral waveform below the list. Multiple selections are supported.

[Remove]: Removes the selected spectral reference from the list.

6 [Load]

Loads the spectral references in [Selected Spectral References] into the experiment.

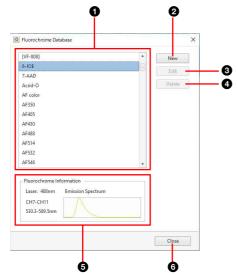
6 [Cancel]

Cancels the operation and closes the dialog.

[Fluorochrome Database] dialog

The [Fluorochrome Database] dialog is displayed by clicking [Fluorochrome Database] on the [Unmixing] tab of the ribbon.

The [Fluorochrome Database] dialog allows you to edit the fluorochrome database.



1 Fluorochrome list

Fluorochromes supplied with the unit.

- [Preset Fluorochrome]: Fluorochromes supplied with the unit. These fluorochromes are available to all users.
- **[Administrator]:** Displays all users and user groups. This item is displayed when logged in using administrator accounts only.
- **Logged-in user:** Custom fluorochromes added by each user. These fluorochromes can be used by other users, but only the user who added the fluorochrome or an administrator may edit the fluorochrome. This item is displayed when logged in using general user accounts only.

2 [New]

Registers a new fluorochrome. Clicking this button displays the [Add New Fluorochrome] dialog.

For details about the [Add New Fluorochrome] dialog, see "Configuring the Virtual Filter Function" (page 95).

3 [Edit]

Clicking this button displays the [Edit New Fluorochrome] dialog where the user can edit registered custom fluorochromes.

4 [Delete]

Deletes the fluorochrome selected in the fluorochrome list.

5 Fluorochrome information

Displays information about the fluorochrome selected in the fluorochrome list.

6 [Close]

Closes the dialog.

[Worksheet Tools] Tab (Ribbon)

The [Worksheet Tools] tab of the ribbon has the following buttons.



[New Plot] group

These buttons are used to add a new plot.

For details about adding plots, see "Adding a Plot" (page 70).

[Density]: Adds a new density plot.
[Dot]: Adds a new dot plot.
[Contour]: Adds a new contour plot.
[Histogram]: Adds a new histogram plot.
[Spectrum]: Adds a new spectrum plot.
[Ribbon]: Adds a new ribbon plot.

[Alignment] group

These buttons are used to display a grid on the worksheet and to align displayed items with the grid.

[Show Grid]: Displays the grid on the worksheet.

- **[Snap to Grid]:** Aligns the plots and [Gates and Statistics] table on the worksheet with the grid when they are moved or added.
- [Auto Arrange]: Automatically aligns the plots and [Gates and Statistics] table on the worksheet.

[View] group

These buttons are used to make settings related to the display of plots.

[Change Palette]: Changes the display style of the selected plot.

Click this button and select the display style. Example:



- White background
- White background with highlighted outlying (rare) events
- Black background
- Black background with highlighted outlying (rare) events

Highlighting outlying events is useful for identifying and analyzing rare events.

Zoom%: Changes the display size of plots.

The current zoom factor is displayed below the icon.

This setting is applied to all plots. Click this button and select the zoom factor, or use the slider to set the zoom factor.



[Statistics] group

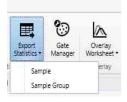
These buttons are used to make settings related to the statistics table.

[Show Table]: Shows/hides the statistics table. [Edit]: Edits the statistics table.

Clicking this button displays the [Statistics Editor] dialog (*page 136*).

[Export Statistics]: Exports the contents of the statistics table as a CSV-format file.

Clicking this button and selecting [Sample] or [Sample Group] displays the [Export Statistics] dialog (*page 137*).

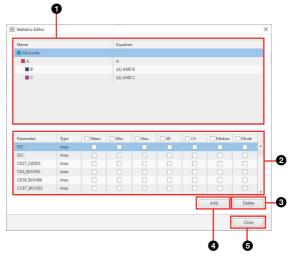


- **[Sample]:** Exports statistics information for each sample.
- **[Sample Group]:** Exports statistics information for each sample group.

[Statistics Editor] dialog

The [Statistics Editor] dialog is displayed by clicking [Edit] in the [Worksheet Tools] tab of the ribbon. The [Statistics Editor] dialog is used to add statistics to the statistics table.

For details about settings, see "Customizing the Statistics Table" (page 74).



1 Gate list

Displays the hierarchy of gates added to the displayed sample tube.

The top level in the hierarchy is represented by [All Events] (ungated parent population).

2 Statistics list

Displays the statistics values for the selected gate in the gate list.

Place a check mark in the checkbox for the statistics you want to add to the statistics table.

You can select the following statistics.

[Mean]: Mean value in the gated population.

- The mean value can also be used as a guide when adjusting the spectral unmixing spillover matrix manually.
- [Min.]: Minimum value in the gated population.
- [Max.]: Maximum value in the gated population.
- **[SD]:** Standard deviation of events from the mean value of the gated population.
- **[CV]:** Coefficient of variation of the distribution in the gated population.

[Median]: Median value in the gated population. The median value can also be used as a guide when adjusting the spectral unmixing spillover matrix manually if there are a large number of outlying events (rare points).

[Mode]: Mode value in the gated population. Corresponds to the highest peak on a histogram plot.

3 [Delete]

Deletes the pulse parameter selected in the statistics list.

4 [Add]

Adds pulse parameters to the statistics list. A new parameter configuration item is added to the bottom with each click. Select [Parameter] and [Type], and select the parameters to display.

5 [Close]

Closes the dialog.

[Export Statistics] dialog

The [Export Statistics] dialog is displayed by clicking [Export Statistics] – [Sample] or [Sample Group] on the [Worksheet Tools] tab of the ribbon.

The [Export Statistics] dialog is used to export the statistics table as a CSV-format file.

Output Path	D:\data\ID7000-20200115-094936\20200110.csv		
		0%	Export
			112

[Output Path]: Specifies the output folder for saving CSV files. Click ____ to select a folder.

Progress bar: Displays the export progress.[Export]: Starts exporting.[Close]: Closes the dialog.

[Gates] group

This button is used to manage and configure gates.

For details about adding gates, see "Editing Gates" (page 72).

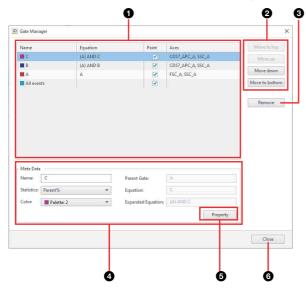
[Gate Manager]: Manages and configures the gates added to the selected sample tube.

Clicking this button displays the [Gate Manager] dialog (*page 137*).

[Gate Manager] dialog

The [Gate Manager] dialog is displayed by clicking [Gate Manager] on the [Worksheet Tools] tab of the ribbon.

The [Gate Manager] dialog is used to manage and configure the gates added to the selected sample tube.



1 Gate list

Displays a list of gates added to the selected sample tube. The following information is displayed for each gate. **[Name]:** Displays the name of the gate.

[Equation]: Displays the equation of the gate.

[Paint]: Placing a check mark in the corresponding checkbox adds color to the display of dot plots.

[Axes]: Displays the X and Y axes of the plot.

2 Change order

Changes the order of the gate selected in the list. The order is used to change the coloring of events for each gate on dot plots.

[Move to top]: Moves the gate to the top.

[Move up]: Moves the gate up one row each time the button is clicked.

[Move down]: Moves the gate down one row each time the button is clicked.

[Move to bottom]: Moves the gate to the bottom.

3 [Remove]

Deletes the gate selected in the list.

4 [Meta Data]

Displays information about the gate selected in the list. **[Name]:** Displays the name of the gate. You can edit the information, as required.

[Statistics]: Displays the statistics parameter configured for the gate. You can edit the information, as required.

Parent%	Ŧ
Events	
Parent%	
Total%	

- **[Total%]:** Proportion of events in the gated population as a percentage of the total number of events ([All Events]).
- **[Parent%]:** Proportion of events in the gated population as a percentage of the number of events in the parent population.
- **[Events]:** Number of events in the gated population.

[Color]: Displays the color of the gate. You can edit the information, as required.



[Parent Gate]: Displays the name of the parent gate. **[Equation]:** Displays the equation of the gate.

[Expanded Equation]: Displays the equation in the format "(Parent gate) AND Equation." Note that this item is omitted if the parent is "All Events."

5 [Property]

Displays the [Gate Properties] dialog (page 137).

6 [Close]

Closes the dialog.

[Gate Properties] dialog

The [Gate Properties] dialog is displayed by clicking [Property] in the [Gate Manager] dialog or by clicking in the [Edit] in the [Gate Tools] tab of the ribbon. The [Gate Properties] dialog displays information about the selected gate. You can change the setting of each parameter, as required.

D Gate Properties	с	
Name:	Show on the chart	
Statistics:	Parent% ✓ Show on the chart	Ŧ
Color:	Palette: 2	Ŧ
Parent gate:	A	v
Equation:	С	
Expanded Equation:	(A) AND C	
		Close

[Name]: Displays the name of the gate.

Place a check mark in the checkbox to display the name of the gate on the plot.

[Statistics]: Selects the statistics parameter configured for the gate.

Place a check mark in the checkbox to display the statistics parameter on the plot.

Parent%	
Events	
Parent%	
Total%	

- **[Total%]:** Proportion of events in the gated population as a percentage of the total number of events ([All Events]).
- **[Parent%]:** Proportion of events in the gated population as a percentage of the number of events in the parent population.
- **[Events]:** Number of events in the gated population.

[Color]: Selects the color of the gate.

Palette: 0	
Standard colors	
🔲 All e 📕 🔳 📕 📕 📕	
A User defined colors	
	//
A Edit Selected Color	

[Parent Gate]: Displays the name of the parent gate. [Equation]: Displays the equation of the gate. This parameter cannot be modified.

[Expanded Equation]: Displays the equation in the format "(Parent gate) AND Equation." Note that this item is omitted if the parent gate is "All Events." This parameter cannot be modified.

[Close]: Closes the dialog.

[Overlay] group

This button is used to display spectral data for the selected plots overlaid.

[Overlay Worksheet]: Displays the overlay worksheet.

- [Half Worksheet View]: Displays the overlay worksheet in the bottom half of the worksheet.
- [Full Worksheet View]: Displays the overlay worksheet in the full area of the worksheet.
- [Hide Overlay Worksheet]: Hides the overlay worksheet.

For details, see "Overlaying Multiple Spectral Data" (page 84).

[Print] group

This button is used to print the worksheet.

Note

The print function is unavailable during data acquisition.

[**Print]:** Prints the content displayed on the worksheet. Clicking this button displays the [Print] window (*page 117*).

[Plot Tools] Tab (Ribbon)

The [Plot Tools] tab of the ribbon has the following buttons.



For details about operations, see "Editing Plots" (page 69).

[Plot Type] group

These buttons are used to change the plot type. This item can be applied to multiple selected plots on the worksheet.

[Plot Type]: Changes the type of the selected plot.

Density	Contour
🔀 Dot	
Histogram	
Plo	t Type

Tip

Spectrum plots cannot be modified.

[Edit] group

These buttons are used to duplicate and delete plots.

[Duplicate Plot]: Duplicates the selected plot within the worksheet.

[Remove Plot]: Deletes the selected plot.

Displays the properties of the selected plot. You can change the setting of each parameter, as required. Clicking this button displays the [Plot Properties] dialog (page 139).

[Plot Properties] dialog

The [Plot Properties] dialog displays information about the selected plot. You can change the setting of each parameter, as required.

• Properties of dot plots, density plots, and histogram plots

Title:		1		
Source:	All events	¥		
Plot Type: D	ot Plot	¥		
X Axis				
Parameter:	FSC	v	Minimum:	1
	Area	v	Maximum:	709020
Axis Type:	Linear	v	Label:	FSC_A
Axis iype:				Axis Settings 🔹
- Y Axis				Axis Settings 🔹
244 0	SSC	¥	Minimum:	1
Y Axis	SSC Area	*	Minimum: Maximum:	Axis Settings • 1 770000
Y Axis	E SANTA S	* *		1
– Y Axis – Parameter:	Area		Maximum:	1 770000

1 Basic information

Displays the following information. You can change the settings, as required.

[Title]: Title of the plot.

[Source]: Population of the plot (parent gate). **[Plot Type]:** Type of plot.

2 [X Axis]

Displays information about the X axis of the plot. You can change the settings, as required.

[Parameter]: Parameter.

[Axis type]: Type of axis.

- You can select the following axis types.
- Linear: Performs a linear transformation between the acquisition values and the channel numbers (bins) used to acquire data (0 to 256). Linear scales are typically used to display forward scatter and back scatter parameters on plots.
- Logarithmic: Display cells that exhibit fluorescence with intensities across a wide dynamic range. Log scales help to make individual peaks easier to visualize. However, log scales cannot adequately represent data with a low mean and high variance.
- Biexponential: Combination of a linear scale (at the lower end of the axis, around zero) and a logarithmic scale at the higher end of the axis, with an algorithmic transition between the two scales. A biexponential scale allows events close to or below zero to be plotted.

[Minimum]: Minimum value of the scale.

[Maximum]: Maximum value of the scale. **[Label]:** Axis label displayed on the graph.

[Options]

[Negative Area]: Sets negative area. [Extra Negative Area]: Sets extra negative area.

[Axis Settings]

[Save as Preference Settings]: Saves the current state as preference settings.

3 [Y Axis]

Displays information about the Y axis of the plot. For details about each item, see [X Axis] (2).

4 [Close]

Closes the dialog.

Properties for contour plots

This section describes the basic information that applies to contour plots only.

For details about other properties, see "Properties of dot plots, density plots, and histogram plots" (page 139).

Тір

During acquisition the contour plot is shown as a density plot. After acquisition is completed, the plot will change into a contour plot.

Title:				
iource:	🗖 All events	¥.		
lot Type:	Contour Plot	٣	Method:	Probability
			Percentage:	5
			Color Scheme:	Multicolor
			Show Outliers:	
X Axis				
Parameter:	FSC	v	Minimum:	1
	Area	v	Maximum:	709020
Axis Type:	Linear	٣	Label:	FSC_A
				Axis Settings 🔹
Y Axis —				
Parameter:	SSC	٧	Minimum:	1
	Area	٣	Maximum:	770000
Axis Type:	Linear	¥	Label:	SSC_A
				Axis Settings 🔹

Contour plot basic information

Displays the following information. You can change the settings, as required.

[Method]: Division method.

• [Probability]: Draws contour lines so that the area between each pair of contour lines contains the same number of events.

- [Linear]: Draws contour lines so that the area between each pair of contour lines contains the equal percentage of the maximum density.
- [Log]: Draws contour lines so that the area between each pair of contour lines contains a percentage of the maximum density divided logarithmically.

[Percentage]: Selects percentage division of the contour lines. The larger the number, the larger the distance between contour lines.

[Color Scheme]: Contour colors.

- [Multicolor]: High-density contour lines are shown in red, and low-density contour lines are shown in blue.
- [Black/White]: Display in black and white.

[Show Outliers]: Place a check mark here to display events outside the outermost contour line as a dot plot.

• Properties for spectrum plots

This section describes [X Axis] only.

For details about other properties, see "Properties of dot plots, density plots, and histogram plots" (page 139).

Tip

The plot type for a spectrum plot cannot be changed.

				tle:
		•	All events	ource:
		÷.	bbon Plot	ot Type: 🛛 Ri
				X Axis
Ť	Minimum:	×	Channel	Parameter:
37	Maximum:	*		
G# 1 1.				
Channel Axis Settings 🔹	Label:	¥	Custom	Axis Type:
Channel	Label:	W	Custom	
Channel	Label: Minimum:	¥	Custom	Y Axis
Channel Axis Settings 🔹				Y Axis
Channel Axis Settings ▼ -154	Minimum:	*	Intensity.	Axis Type: Y Axis

[X Axis]

Displays information about the X axis of the plot. You can change the settings, as required.

[Parameter]: Parameter.

- [Axis type]: Type of axis. This parameter cannot be modified.
- [Minimum]: Minimum value of the scale. This parameter cannot be modified.
- [Maximum]: Maximum value of the scale. This parameter cannot be modified.

[Label]: Axis label displayed on the graph.

[Axis Settings]

[Save as Preference Settings]: Saves the current state as preference settings.

Тір

The range that can be specified using scale minimum and maximum values depends on the parameter. The minimum and maximum values of [Wavelength] and [Channel] are fixed.

[Scale] group

These buttons are used to make settings related to the scale of the X and Y axes.

This item can be applied to multiple selected plots on the worksheet.

[X Axis] / [Y Axis]: Selects the type for the X and Y axes.

For details about other properties, see [X Axis] (2) for the properties of dot plots, density plots, and histogram plots (page 139).

[Swap]: Swaps the X axis and Y axis parameters.

[Auto Adjustment (XY)]: Automatically adjusts the scale of the X axis and Y axis.

- [Auto Adjustment (X Axis)]: Automatically adjusts the scale of the X axis.
- [Auto Adjustment (Y Axis)]: Automatically adjusts the scale of the Y axis.

[Zoom]: Changes the magnification of plots. Click this button to turn on zoom, and then drag around the portion of the plot you want to zoom in on to magnify that portion.

[Default Adjust]: Restores the scales to the default view.



Tips

- Automatically adjusting the axis scale places the minimum and maximum values of the population near the ends of the X axis and Y axis.
- The automatic adjustment function adjusts the axis when the button is clicked. During data acquisition, events that are detected after the axes are automatically adjusted may be off-scale. In this case, readjust the axes as required.

[View] group

These buttons are used to make settings related to the display of plots.

[Spectrum Display]: Sets the spectrum display mode.

- [Display All]: Displays all events.
- [Display High Density]: Displays the highincidence spectral information, omitting lowincidence events, to make the display easier to view.

[New Gate] group

These buttons are used to add a new gate.

For details about adding gates, see "Editing Gates" (page 72).

[Rectangle]: Adds a rectangle gate.

[Ellipse]: Adds an ellipse gate.

[Polygon]: Adds a polygon gate.

- **[Quadrant]:** Adds a quadrant gate. Quadrant gates are added with the intersection point of the quadrants at the position of the mouse click.
- [Linear]: Adds a linear gate to a histogram plot. Linear gates are added by dragging the mouse horizontally on the histogram plot. The height of the reference line is for display only, and has no effect on the value of the linear gate.

[Tool] group

[Sync Scale and Gate]: Synchronizes the scale and gates of all plots within a sample group.

[Copy Picture]: Copies the selected plot or [Gates and Statistics] table to the clipboard for pasting in other software, for example to produce reports.

This item can be applied to multiple selected plots on the worksheet.

[Gate Tools] Tab (Ribbon)

The [Gate Tools] tab of the ribbon is displayed when a gate is selected in a plot or in the statistics table. The [Gate Tools] tab has the following buttons.



For details about operations, see "Adding a Plot" (page 70).

[Edit] group

These buttons are used to edit the selected gate.

[Remove Gate]: Deletes the selected gate. [Gate Manager]: Manages and configures the selected

gate. Clicking this button displays the [Gate Manager] dialog.

The [Gate Manager] dialog can also be accessed from the [Worksheet Tools] tab of the ribbon. See "[Gate Manager] dialog" (page 137).

Displays the properties of the selected gate. You can change the setting of each parameter, as required. Clicking this button displays the [Gate Properties] dialog (*page 137*).

[Label] group

These buttons make settings related to gate labels.

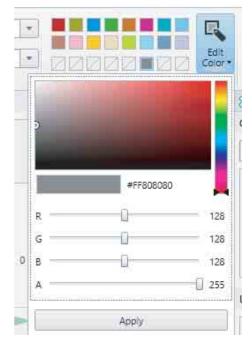
[Font Size]: Selects the font size of text labels. [Statistic]: Selects the statistic that is displayed beside gates on plots.

[Color] group

This group is used to change the colors of gates.

Color palette: Changes the color of the selected gate. [**Edit Color**]: Changes a color in the palette to a custom color.

Click a color in the color palette, click the [Edit Color] button, then select a new color, and click [Apply].



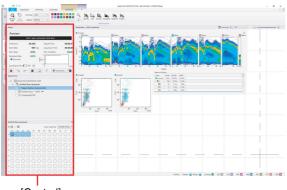
[New Child Plot] group

These buttons are used to add a new child plot.

The function and use of each button is the same as the [New Plot] group on the [Worksheet Tools] tab of the ribbon. See "[New Plot] group" (page 135) on the [Worksheet Tools] tab.

Control Pane

The control pane is used to perform acquisitions and to set acquisition conditions. The control pane comprises two parts: the [Control] area on the left containing panels for various operations, and the [Settings] area on the right containing panels for setting acquisition conditions.



[Control] area

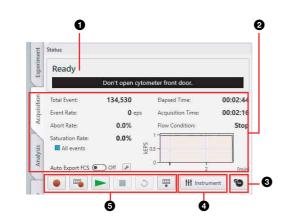
The following panels are displayed in the areas.

- [Control] area
 - [Status] panel (page 142)
 - [Experiment] panel (*page 143*)- [Plate] panel (*page 143*)

[Status] panel

The [Status] panel is used to check the status of the main unit, and to perform data acquisition operations.

For details about operations, see "Previewing" (page 58).



1 Instrument status

Displays the status of the instrument. During warm-up and cleaning, the remaining time is displayed on the right hand side.

Displays the following status.

- Warming Up
- QC (quality check)
- Disconnected
- Maintenance (calibration and other tasks)
- Error
- Cleaning
- Ready
- Preview
- Acquire (data acquisition mode)
- Plate Unloaded
- Auto Acquire
- Acquisition Offset
- Agitate
- Please wait... (during preparation)

2 Data acquisition status

Displays the status of data acquisition.

- [Total Events]: Displays the total number of events detected during data acquisition.
- [Elapsed Time]: Displays the cumulative elapsed time since the start of data acquisition.
- [Event Rate]: Displays the rate events are detected during data acquisition in events per second (eps). (Value range: 0 eps to 40,000 eps) Changes in the event rate are also displayed as a graph.
- [Acquisition Time]: Displays the duration of data acquisition.
- [Abort Rate]: Displays the rate events are aborted during data acquisition as a percentage.
- [Flow Condition]: Displays the fluidics status. [Stop]: Data acquisition is stopped.

[Adjusting]: The sample pressure is being adjusted.

- [Stable]: The sample pressure is stable.
- [Low Event Rate]: The event rate is low or is reducing.

[Air]: Air bubbles have been detected.

- **[Saturation Rate]:** Displays the rate saturated events are detected during data acquisition as a percentage. You can specify the target gate for which to calculate the percentage. When a gate is specified, the name of the gate is displayed below the title.
- [Auto Export FCS]: Exports an FCS file automatically upon data acquisition.

For details, see "Exporting FCS File Automatically During Acquisition" (page 98).

3 (Stopping Condition)

Defines settings related to the data acquisition stop conditions.

For details, see "[Stopping Condition] dialog" (page 131).

4 [Instrument]

Configures settings related to lasers, detector, and flow rate.

Clicking this button displays the [Instrument Settings] dialog.

For details, see "Configuring Instrument Settings" (page 59).

6 Operation buttons

- (Acquire): Acquires and records data.
- **(Auto Acquire):** Runs auto acquisition.
- **(Preview):** Previews the acquisition. It does not start data recording.
- **Stop):** Stops data acquisition manually.
- **O** (**Restart):** During preview, resets the data display.
- (Load) / (Unload): Loads/unloads the multiwell plate.

[Experiment] panel

The [Experiment] panel displays the structure of the experiment.

An experiment contains the following structural components.

Experiment	
 Experiment 2020/10/15 15:38 96 Well Plate (standard) 	A
Single Positive Controls (4/4)	
🗌 🎖 Sample Group - 1 (0/0) 👄	
☐ 88 Sample Group - 2 (0/0) ←>	
🔜 🍪 Sample Group - 3 (0/0) 🚥	
🔄 😽 Sample Group - 4 (0/0) 🖙	
🗔 🍪 Sample Group - 5 (0/0) 👄	

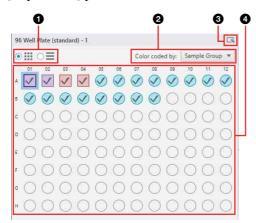
Experiment:

96 : Plate

- 88: Wells not belonging to a sample group
- 88 88 88 : Sample group

[Plate] panel

The [Plate] panel displays the wells of the plate selected in the [Experiment] panel.



1 View

You can switch the display to list view or map view.

2 [Color Coded by]

You can sort wells by color for selected metadata.

- Sample Group
- Cell
- Condition

8 💽

The [Plate] panel is displayed in a separate window.

4 Wells

Selecting a well opens the worksheet for conducting data acquisition or analysis.

The status of each well is displayed using the following icons.

C: Data has not been acquired yet.

- 🕃 : Data acquisition is in progress.
- S: Data acquisition has been completed.
- (): Target well for preview or data acquisition.
- : Target well for auto acquisition.

Well in which the event rate has dropped or air bubbles are detected.

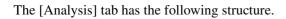
The shape of the icon will vary depending on the worksheet mode.

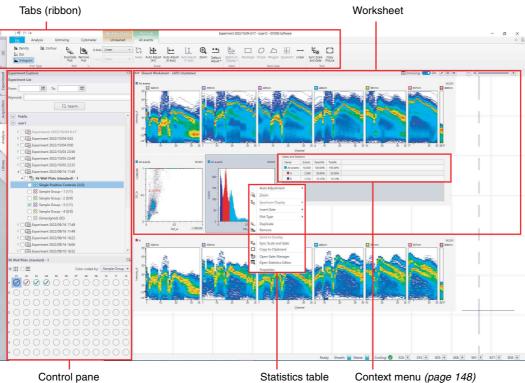
(circle): [Shared] mode

(square): [Individual] mode

[Analysis] Tab

The [Analysis] tab is displayed by clicking the [Analysis] workflow tab. The [Analysis] tab is used to perform analysis operations on the acquired data.





(page 147)

Control pane (page 145)

The ribbon has the following tabs (tabs vary depending on the current selection).

- [Analysis] tab (page 144)
- [Unmixing] Tab (page 132)
- [Cytometer] tab (page 124)
- [Worksheet Tools] tab (*page 135*)
- [Plot Tools] tab (*page 138*)
- [Gate Tools] tab (page 141)

Tip

The tabs, other than the [Analysis] tab, are the same as for the [QC] tab and [Acquisition] tab of the ribbon.

[Analysis] Tab (Ribbon)

The [Analysis] tab of the ribbon has the following buttons.

[Experiment] group

These buttons are used to perform operations related to experiments.

[Open in Acquisition]: Opens the experiment selected in the [Experiment List] panel in the [Acquisition] workflow tab to edit the experiment and conduct acquisition.

[Send to Public]: Shares the displayed experiment as a public experiment.

Click this button to make a clone of the displayed experiment in the Public (shared) folder.

- [Save as Template]: Saves the experiment selected in the [Experiment] panel as a template.
- Opens a dialog for editing information about the experiment selected in the [Experiment List].

[Plate] group

These buttons allow you to delete a plate from an experiment or edit plate settings.

[Delete]: Deletes the plate selected in the [Experiment] panel.

[Layout Editor]: Edits the plate layout selected in the [Experiment] panel.

Clicking this button displays the [Layout Editor] tab of the [Plate Properties] dialog (*page 129*).

[Export Layout to CSV File]: Exports the metadata for the plate selected in the [Experiment List] panel as a CSV-format file.

Clicking this button displays the [Export to CSV File] dialog (*page 129*).

[Import Layout from CSV File]: Imports the metadata for the plate selected in the [Experiment List] panel.

Clicking this button displays the [Import Layout from CSV File] dialog (*page 129*).

I: Opens a dialog for editing information about a plate.

[Sample Group] group

These buttons are used for operations on sample groups.

- [Delete]: Deletes the sample group selected in the [Experiment] panel.
- General Contraction in the sample group.

[Sample] group

These buttons are used for operations on wells.

For details, see "[Sample] group" (page 130) on the [Acquisition] tab.

[Batch Processing] group

These buttons are used to run fluorescence unmixing and analysis on multiple samples at the same time.

For details, see the "[Batch Processing] group" (page 132) on the [Acquisition] tab.

[Record] group

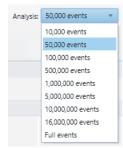
These buttons are used for records and electronic signatures.

For details, see the "[Record] group" (page 132) on the [Acquisition] tab.

[Display Events] group

Sets the number of events that are displayed on plots.

[Analysis]: Selects the number of events to display on plots.



Control Pane

The control pane is used to select a sample for analysis. The control pane is a collapsible window element. You can click [<<] / [>>] to show/hide the pane as required.

The following panels are displayed in the control pane. • [Experiment List] panel (*page 67*)

[Library] Tab

The [Library] tab is displayed by clicking the [Library] workflow tab.

The [Library] tab is used to edit spectral references.

The [Library] tab has the following structure.

	[Filters] pane				[Search Results]											
						openner.	205.79	0.031-		00-07	0150540					
5.5.	ombr												T			
ROT NOT	Ren Der	and look											1		to been Die	
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	S Label	254	0.04	B1000	355an	Ē	1			Ê				43120354	410	
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	Elber Alfen	25	007	1000	40m		1	K					tered.	shatte	262	
	C ablon	254	00103	1 man	dim		1	I					terral (shelida	228	N-
	C Minn	35e	0016	Petter	40m			N					land	abolide	41	
	Non	154	054	BIN08	355nm				A.,				iord	0035eV	Not Aulist	
	C Renateure	1%	0256	8018	4510								tored .	43:02:54	3.0	
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	C M60 C M30	124	00102	B1621	40m				2				level.	1000	The Average	
	C MC	Sec.	6000	ar 485	4800									abolide	496	10-m
	LACC/1 BACR	254	0014	BIVER.	355nm								icend	43(39)54	20	
	0 8214% 0 82042	254	C2+64	100	465nm								iord	alczółów	40	
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	C 95-055 C 95-07	24	000	FE-67410	385mm									ale di la la	40	
	6-beside															

The ribbon has the following tabs.

- [Library] tab (page 146)
- [Cytometer] tab (page 124)

[Library] Tab (Ribbon)

The [Library] tab of the ribbon has the following buttons.



[Spectral Reference] group

Used for operations on spectral references.

[Import]: Imports spectral reference data saved in a file.[Export]: Exports the spectral reference data to a file.[Send to Public]: Shares the spectral reference selected in the list as a public spectral reference.

Edits information related to the spectral reference selected in the list.

[Spectral Reference Properties] dialog

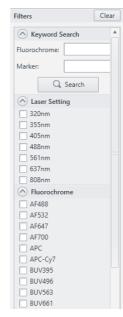
The [Spectral Reference Properties] dialog is used to configure metadata properties

Spectral Reference Informa	
Name:	BUV395
Label:	35c
Marker:	CD4
Lasen	320nm: 🎇 355nm: 🎇 405nm: 🎇 488nm: 🚼 561nm: 🚼 638nm: 🎇 808nm:
Mode:	Normal
Instrument Settings ID	a3cd3b5e
Spectral Index:	Area: 418
	Height: 421
Saturation Rate:	
Expiration Date:	2020/08/29
Library:	Private
Registration User:	administrator
Cell Type:	
Clone:	a
Species:	
Manufacturer:	
Lot Number:	
Acquisition Date:	2019/08/29 10:39:02
Calculation Date:	2019/11/27 11:47:12
Registration Date:	2019/12/11 14:55:21
Spectral Reference ID:	5cbfec40-fd73-413e-bb84-b08dd276ab31
Experiment Name:	35 23 15 color cyto Experiment 5/28/2019 3:05 PM NEO
Plate Name:	96 Well Plate (standard) - 1
Sample Group Name:	Sample Group - 1
Matrix Name:	35 color
	OK Cancel

[Filters] Pane

The [Filters] pane filters the search for a spectral reference.

The search results for filters with a check mark are displayed in [Search Results].



[Search Results]

Displays the spectral reference search results in list view. You can filter by lasers using the drop-down menu at the top right.

You can check the waveform in the area on the right by selecting a spectral reference in the list.

faming.	Label	Markar	Fluorochrome	Dr. Later	322011	355rm	405em	458nm	Stine	60mm	008rm	Mode	Instrument Settings (D	Spectral Index: Set	
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	294	C011C	81321	25.00								Normal	absiliele	228	~
	25e	C07	38436	dition.								Norrel	aboliste	242	
	254	00110	81021	ative								Norral	abalide	228	N-
	354	CC86	Patho	ation			N					Norral	abolide	49	1
	154	CD4	835385	155mm				A				Normal	+#255e64	Not Analytick	
	254	C056	81510	dire								Normal	absible	m	405-m
	154	00110	81121	dlive				2				Normal	18355404	Not Available	
	154	00110	8/121	dition				2				Normal	x8355e01	Not Available	
	26e	CD25	1448	425.00								Normal	abilitie	416	Albert
	264	CD38	80.036	255mm								Normal	alizable	313	Alleren
	264	00164	avise	425-00								Normal	ahulikie	453	
	264	CD127	AF522	diam.								Normal	aluikie	190	
	154	C025	A-488	482-00								Noval	uR155w04	416	
	154	C056	81510	dition.								Nonnal	cR355e01	m	
	154	C025	A-428	diam.								Normal	28155e01	436	
	154	C056	81510	405m				<u>A.</u>				Normal	c8355e01	10	
	154	C056	8 1510	42510								Normal	c8255e01	372	
	254	CCR7	8,4563	255 vm								Normal	alicitzie	155	
	254	CD93	8	SEtim								Nonnal	alkolikóe	615	
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	254	CD45R0	01570	455 ve.								Nonal	aloolitide	122	
	254	CCR4	ev585	455nm								Nonnal	aloibte	274	
	25c	C004	PG-42510	SEtine-								Nonnal	aloolib5e	492	
		2000		1000										AN	

Worksheet

Toolbar

The following tools are available on the top right of the worksheet.



[Unmixing]: Selects whether spectral unmixing is applied to plots on the worksheet.

This setting is reflected on the unmixing settings. This function is the same as [Apply] (*page 132*) on the [Unmixing] tab of the ribbon.

Used to adjust the spectral unmixing. Clicking this button displays the [Unmixing Settings] dialog (page 133).

This function is the same as [Unmixing Settings] (*page 132*) on the [Unmixing] tab of the ribbon.

 Enables manual adjustment of spectral unmixing by adjusting the spectral reference of the selected matrix. Clicking this button displays the [Spectral Reference Adjuster] dialog (page 81).

This function is the same as [Spectral Reference Adjuster] (*page 133*) on the [Unmixing] tab of the ribbon.

• You can check the results of spectral unmixing by displaying a list of plots for each fluorochrome combination.

Clicking this button displays the [Unmixing Viewer] dialog.

This function is the same as [Unmixing Viewer] (*page 133*) on the [Unmixing] tab of the ribbon.

For details, see "Checking Spectral Unmixing Results Summary" (page 82).

- /+: Zoom in/out on the worksheet.

Statistics Table

The [Gates and Statistics] table displays the following information.

Name	Events	Parent%	Total%	
All events	5,019	100.00%	100.00%	
A	1,708	34.03%	34.03%	
B B	374	21.90%	7.45%	
C	1,327	77.69%	26.44%	

Displays the names of gates and parameters.

2 [Events]

Displays the number of events.

3 [Parent%]

Displays the proportion of events of each gate relative to its parent.

• [Total %]

Displays the proportion of events of each gate relative to the total of all events.

5 Gate hierarchy

Displays the hierarchical relationship between gates. The color to the left of the gate name shows the color of the gates on plots.

Тір

You can change the statistics items displayed for each gate in the statistics table.

See "Customizing the Statistics Table" (page 74).

Context Menu

The context menu is displayed by right-clicking items in the control pane or plots on the worksheet, and provides a shortcut for common operations.

Experiments

The following menu commands are displayed when right-clicking an experiment icon in the [Experiment] panel (*page 143*).

- [Preview Record]: Displays an electronically signed record PDF file. This menu item is displayed only for an experiment (record) configured with [Audit Trail] set to [On].
- [New Plate]: Adds a new plate. This function is the same as [Plate] [New] (*page 129*) on the [Acquisition] tab of the ribbon.
- [Send to Public]: Shares the experiment as a public experiment. This function is the same as [Send to Public] (*page 148*) on the [Analysis] tab of the ribbon.
- **[Save as Template]:** Saves the experiment selected in the [Experiment] panel as a template. This function is the same as [Save as Template] on the [Acquisition] tab and [Analysis] tab of the ribbon.
- [Batch Unmixing]: Runs spectral unmixing on multiple samples at the same time.
- [Batch Analysis]: Runs analysis on multiple samples at the same time.
- [Export to FCS File]: Exports FCS file. This function is the same as [Export to FCS File] on the [Acquisition] tab and [Analysis] tab of the ribbon.
- [Sign Record]: Creates a record in PDF format and then electronically signs the file. This function is the same as [Record] - [Sign Record] on the [Acquisition] tab and [Analysis] tab of the ribbon.
- [Export Record]: Exports a record. This function is the same as [Record] [Export Record] on the [Acquisition] tab and [Analysis] tab of the ribbon.
- [Import Record]: Imports a record. This function is the same as [Record] [Import Record] on the [Acquisition] tab and [Analysis] tab of the ribbon.
- [Delete Cached Data]: Deletes cached data. This deletes all fluorescence compensation data within an experiment, regardless of the unmixing matrix selection status, to free up hard disk space by that amount.
- [Change Worksheet Mode]: Changes the worksheet mode.
- [**Properties**]: Displays the properties for the selected experiment.

Plates

The following menu commands are displayed when right-clicking a plate in the [Experiment] panel (*page 143*).

- **[New Sample Group]:** Creates a new sample group. This function is the same as [Sample Group] – [New] (*page 130*) on the [Acquisition] tab of the ribbon.
- [Add to Auto Acquisition Target]: Sets all the wells contained in a sample group in a plate as targets for auto acquisition.

[Remove from Auto Acquisition Target]: Removes all the wells contained in a sample group in a plate as targets for auto acquisition.

[Load Template]: Applies template settings to the selected plate. This function is the same as [Plate] – [Load Template] (*page 129*) on the [Acquisition] tab of the ribbon.

[Export Layout to CSV File]: Exports the metadata for the selected plate as a CSV-format file. This function is the same as [Plate] – [Export Layout to CSV File] (*page 129*) on the [Acquisition] tab of the ribbon.

- [Import Layout from CSV File]: Imports the metadata for the selected plate. This function is the same as [Plate] – [Import Layout from CSV File] (*page 129*) on the [Acquisition] tab of the ribbon.
- [Batch Unmixing]: Runs spectral unmixing on multiple samples at the same time.
- [Batch Analysis]: Runs analysis on multiple samples at the same time.
- [Export to FCS File]: Exports FCS file. This function is the same as [Export to FCS File] on the

[Acquisition] tab and [Analysis] tab of the ribbon. [**Duplicate]:** Duplicates the selected plate.

- [**Delete**]: Deletes the selected plate.
- [Change Worksheet Mode]: Changes the worksheet mode.
- [**Properties**]: Displays the properties for the selected plate.

Sample Groups

The following menu commands are displayed when right-clicking a sample group in the [Experiment] panel (*page 143*).

[Remove from Auto Acquisition Target]:

Removes all the wells contained in a sample group as targets for auto acquisition.

- [Batch Unmixing]: Runs spectral unmixing on multiple samples at the same time.
- **[Batch Analysis]:** Runs analysis on multiple samples at the same time.

[Export to FCS File]: Exports FCS file. This function is the same as [Export to FCS File] on the [Acquisition] tab and [Analysis] tab of the ribbon.

- [Delete]: Deletes the selected sample group.
- [Copy to all Individual Worksheets]: Copies the shared worksheet to the individual worksheet of all samples.
- [Import Instrument Settings]: Imports instrument settings.
- [Paste Instrument Settings]: Pastes copied instrument settings into the current sample group. This function is the same as [Paste Instrument Settings] (*page 61*) in the menu displayed by clicking [Instrument Settings] in the bottom right of the [Instrument Settings] dialog.
- **[Instrument Settings]:** Displays the [Instrument Settings] dialog. This function is the same as [Instrument Settings] (*page 142*) on the [Status] panel.
- [Stopping Condition]: Displays the [Stopping Condition] dialog. This function is the same as [Stopping Condition] (*page 131*) on the [Status] panel.
- [Change Worksheet Mode]: Changes the worksheet mode.
- [Sample Group Type]: Changes the sample group type.
- [**Properties**]: Displays the properties for the selected sample group.

Wells

The following menu commands are displayed when right-clicking a well in the [Plate] panel (*page 143*).

- [Set Current Position]: Assigns the target well for preview and data acquisition. This function is the same as [Acquire] – [Set Current Position] (*page 128*) on the [Acquisition] tab of the ribbon.
- [Append]: Appends added data acquisition results for the selected well.
- [Add to Auto Acquisition Target]: Selects the target wells for automatic acquisition. This function is the same as [Auto Acquire] – [Add to Acquisition Target] (*page 128*) on the [Acquisition] tab of the ribbon.

[Remove from Auto Acquisition Target]: Removes the selected well from the list of wells for automatic acquisition.

[Copy] / [Paste]: Copies/pastes the selected well. This function is the same as [Sample] – [Copy]/[Paste] (*page 130*) on the [Acquisition] tab of the ribbon.

- [Export Instrument Settings]: Exports instrument settings to a file.
- [Copy Instrument Settings]: Copies the instrument settings.
- [Export to FCS File]: Exports FCS file. This function is the same as [Export to FCS File] on the [Acquisition] tab and [Analysis] tab of the ribbon.

[[]Add to Auto Acquisition Target]: Sets all the wells contained in a sample group as targets for auto acquisition.

[New Sample Group]: Adds a sample group to an experiment. This function is the same as [Sample Group] – [New] (*page 130*) on the [Acquisition] tab of the ribbon.

[Move to Sample Group]: Moves the selected well to another sample group. This function is the same as [Sample] – [Move to Sample Group] (*page 130*) on the [Acquisition] tab of the ribbon.

[Remove from Sample Group]: Removes the selected well from a sample group. This function is the same as [Sample] – [Remove from Sample Group] (*page 130*) on the [Acquisition] tab of the ribbon.

[Delete Acquired Data]: Deletes the data acquired for the selected well.

[Copy to all Individual Worksheets]: Copies the shared worksheet to the individual worksheet of all samples.

[Copy to Shared Worksheet]: Sets the worksheet for the selected sample as the shared worksheet.

[Instrument Settings]: Displays the [Instrument Settings] dialog. This function is the same as [Instrument Settings] (*page 142*) on the [Status] panel.

[Stopping Condition]: Displays the [Stopping Condition] dialog. This function is the same as [Stopping Condition] (*page 131*) on the [Status] panel.

- [Change Worksheet Mode]: Changes the worksheet mode.
- [**Properties**]: Displays the properties for the selected plot.

[Setup as Single Positive Control]: Sets a sample as a single stain Positive control.

Plots

The following menu commands are displayed when right-clicking a plot on the worksheet.

- [Auto Adjustment]: Automatically adjusts the scale of the X axis and Y axis for the selected plot. This function is the same as [Auto Adjustment (XY)] (*page 140*) in the [Plot Tools] tab of the ribbon.
- **[Zoom]:** Changes the magnification of the selected plot. This function is the same as [Zoom] (*page 140*) in the [Plot Tools] tab of the ribbon.
- [Spectrum Display]: Sets the spectrum display mode. This function is the same as [Spectrum Display] (*page 140*) on the [Plot Tools] tab of the ribbon.
- **[Insert Gate]:** Adds a gate to the selected plot. This function is the same as [New Gate] (*page 141*) in the [Plot Tools] tab of the ribbon. Select the gate type from the submenu.
- [Plot Type]: Changes the type of the selected plot. This function is the same as [Plot Type] (*page 138*) in the [Plot Tools] tab of the ribbon. Select the plot type from the submenu.

[Duplicate]: Duplicates the selected plot within the worksheet. This function is the same as [Duplicate Plot] (*page 138*) in the [Plot Tools] tab of the ribbon.

[**Remove]:** Removes the selected plot from the worksheet. This function is the same as [Remove Plot] (*page 138*) in the [Plot Tools] tab of the ribbon.

[Send to Overlay]: Displays the spectral data of the selected plot in the overlay worksheet.

[Sync Scale and Gate]: Synchronizes the scale and gates of all plots within a sample group.

[Copy to Clipboard]: Copies the selected plot to the clipboard for pasting in other software, for example to produce reports.

[Open Gate Manager]: Displays the [Gate Manager] dialog (*page 137*). This function is the same as [Gate Manager] (*page 137*) in the [Worksheet Tools] tab of the ribbon.

[Open Statistics Editor]: Displays the [Statistics Editor] dialog (*page 136*). This function is the same as [Edit] (*page 136*) in the [Worksheet Tools] tab of the ribbon.

[**Properties**]: Displays the properties for the selected plot.

Gates

The following menu commands are displayed when right-clicking a gate in a plot.

[Create a Child Plot]: Adds a child plot for the selected gate. This function is the same as [New Child Plot] (*page 141*) in the [Gate Tools] tab of the ribbon.

[Assign Gate]: Assigns the selected gate as the following gate types.

- Positive Gate (page 76)
- Negative Gate (page 76)
- Stopping Condition Gate (page 112)
- Saturation Rate Gate (page 142)
- [Auto Adjustment (Biexponential]: Automatically adjusts the scale of the X axis and Y axis for the selected plot. This function is the same as [Auto Adjustment (XY)] (*page 140*) in the [Plot Tools] tab of the ribbon.
- **[Remove]:** Deletes the selected gate. This function is the same as [Remove Gate] (*page 141*) in the [Gate Tools] tab of the ribbon.
- [**Properties**]: Displays the properties for the selected gate.

[Gates and Statistics] Table

The following menu commands are displayed when right-clicking the statistics table on the worksheet.

[Create a Plot]: Adds a parent plot for the selected gate.

- [Assign Gate]: Assigns the selected gate as the following gate types.
 - Positive Gate (page 76)

- Negative Gate (page 76)
- Stopping Condition Gate (page 112)
- Saturation Rate Gate (*page 142*)
- [Auto Adjustment (Biexponential)]: Automatically adjusts the scale of the X axis and Y axis for the selected plot. This function is the same as [Auto Adjustment (XY)] (*page 140*) in the [Plot Tools] tab of the ribbon.
- [Copy] / [Paste]: Copies/pastes the selected gate to duplicate a gate.
- [Cut] / [Paste]: Cuts/pastes the selected gate to move a gate.
- [Remove]: Deletes the selected gate or statistic (multiple selection supported).
- [Export Statistics]: Exports the contents of the statistics table as a CSV-format file. This function is the same as [Export Statistics] (*page 136*) in the [Worksheet Tools] tab of the ribbon.
- **[Open Gate Manager]:** Displays the [Gate Manager] dialog (*page 137*). This function is the same as [Gate Manager] (*page 137*) in the [Worksheet Tools] tab of the ribbon.
- **[Open Statistics Editor]:** Displays the [Statistics Editor] dialog (*page 136*). This function is the same as [Edit] (*page 136*) in the [Worksheet Tools] tab of the ribbon.
- [**Properties]:** Displays the properties for the statistics table.

Worksheet

The following menu commands are displayed when right-clicking blank space on the worksheet.

- [Show Table]: Shows/hides the statistics table. This function is the same as [Show Table] (*page 136*) on the [Worksheet Tools] panel.
- [Auto Arrange Items]: Automatically aligns the plots and [Gates and Statistics] table on the worksheet. This function is the same as [Auto Arrange] (*page 135*) on the [Worksheet Tools] panel.
- [Align Items to Grid]: Aligns the plots and [Gates and Statistics] table on the worksheet with the grid.
- [Show Grid]: Displays the grid on the worksheet. This function is the same as [Show Grid] (*page 135*) on the [Worksheet Tools] panel.
- **[Open Gate Manager]:** Displays the [Gate Manager] dialog (*page 137*). This function is the same as [Gate Manager] (*page 137*) in the [Worksheet Tools] tab of the ribbon.
- **[Open Statistics Editor]:** Displays the [Statistics Editor] dialog (*page 136*). This function is the same as [Edit] (*page 136*) in the [Worksheet Tools] tab of the ribbon.
- [Copy to all Individual Worksheets]: Copies the shared worksheet to the individual worksheet of all samples.
- [Copy to Shared Worksheet]: Sets the worksheet for the selected sample as the shared worksheet.

Templates

The following menu commands are displayed when right-clicking a template in the [Experiment & Template List] panel in the [Experiment] window.

[Delete]: Deletes the selected template.

[**Properties**]: Displays the properties for the selected template.

[Color Panel] Tab of [Unmixing Settings] Dialog

The following menu commands are displayed when right-clicking a fluorochrome in the fluorochrome list.

[Open Positive Sample]: Displays the sample worksheet for the gate configured as the Positive gate.

[Open Negative Sample]: Displays the sample worksheet for the gate configured as the Negative gate.

[Clear Gates]: Clears the gate assignments for the selected fluorochrome.

[Clear Spectral Reference]: Deletes the spectral reference for the selected fluorochrome.

[Edit Virtual Filter]: Edits the virtual filter.

[Remove Fluorochrome]: Removes the selected fluorochrome from the list.

[**Properties**]: Displays the properties for the selected spectral reference.

[Search Result] in the [Library] Window

[Export]: Exports the spectral reference data to a file. **[Send to Public]:** Shares the spectral reference as a public spectral reference. This function is the same as

public spectral reference. This function is the same as [Send to Public] (*page 148*) on the [Analysis] tab of the ribbon.

[Delete]: Deletes the selected spectral reference. [Replace Fluorochrome]: Updates the selected fluorochrome information.

[**Properties**]: Displays the properties for the selected spectral reference.

[History] List on the [QC] Tab

- [Set selected QC Result to baseline]: Sets the selected QC result as the baseline. An asterisk (*) is added to the QC results when set as the baseline.
- [Clear baseline QC Result]: Clears the QC results configured as the baseline.



Maintenance

This chapter describes the routine maintenance procedures required to maintain the performance and longevity of the ID7000.

DANGER

Fluids may contain biological, chemical, or other agents. Always wear gloves and other protective clothing, mask, and goggles, as required, when performing maintenance.

Maintenance Schedule

The maintenance procedures outlined in this manual should be performed according to the schedule below.

Maintenance Checks at Startup

Maintenance item	Schedule	See
Waste fluid disposal Replacing the 10L waste tank	 Check that the tank is not full when starting up and shutting down Replace when nearly full or when prompted in ID7000 Software 	<i>"Emptying/ Replacing the 10L Waste Tank" (page 153)</i>
Refilling the sheath tank	 Check that the tank is full when starting up Refill when prompted in ID7000 Software 	"Refilling the 10L Sheath Tank with Sheath Fluid" (page 154)

Maintenance During Operation

Cleaning the fluidics system

Maintenance item	Schedule	See
Priming	When starting upAs required	"Priming" (page 155)
Cleaning the probe	 As required If more than eight hours have elapsed since the last acquisition 	<i>"Cleaning the Probe Manually" (page 156)</i>
Purging the flow cell	 When clogging of the flow channel is suspected As required 	"Purging the Flow Cell" (page 156)
De-bubbling the sheath line	 When contamination between samples is suspected As required 	<i>"De-bubbling the Sheath Line" (page 158)</i>

Maintenance Checks at Shutdown

Cleaning the fluidics system

Maintenance item	Schedule	See
Cleaning the sample line and sample probe	 As required 	<i>"Cleaning Using Bleach Solution"</i> (page 157)
Shutdown cleaning	 When shutting down at the end of the day 	<i>"Cleaning Before Shutting Down"</i> (page 160)

General cleaning

Maintenance item	Schedule	See
Cleaning the multi-well plate holder	 When shutting down 	"Regular Cleaning" (page 160)
Cleaning the exterior and 5 ml tube rack	 When shutting down 	"Regular Cleaning" (page 160)

Weekly Maintenance

Maintenance item	Schedule	See
Cleaning the sheath tank	• Weekly	"Refilling the 10L Sheath Tank with Sheath Fluid" (page 154) Discard any remaining unused fluid in the 10L sheath tank, wash tank with about 500 ml of sheath fluid, then refill tank with sheath fluid.

Periodic Maintenance

Maintenance item	Schedule	See
Replacing the 10L waste tank and internal waste tank air filter	MonthlyAs required	"Replacing Tank Air Filters" (page 162)
Replacing the sheath filter	 Every 3 to 12 months As required 	"Replacing the Sheath Filter" (page 161)

Miscellaneous

Maintenance item	Schedule	See
Removing/ reconnecting the tank base	As required	"Removing/ Reconnecting the Tank Base" (page 163)

Emptying/Replacing the 10L Waste Tank

Check that the 10L waste tank is not full daily before starting. If it is full, dispose of the waste fluid or replace the tank with a new tank. Also, the waste fluid must be disposed or the 10L waste tank must be replaced when a message appears in ID7000 Software stating that the waste tank is full.

A fluid-level (Full) detection sensor detects whether the 10L waste tank is full or not, which can be monitored using ID7000 Software.

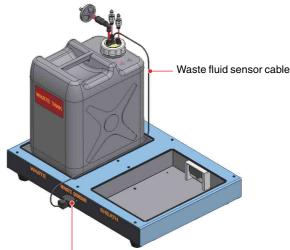
Note

When handling the 10L waste tank, always follow the laboratory rules for the handling of waste fluids.

WARNING

Waste fluid may contain virulent or hazardous substances that may be highly contagious or toxic, depending on the sample under test. Always wear gloves and other protective clothing, mask, and goggles, as required, when handling waste fluid.

- **1** Turn off the two main power switches on the lower right of the left side panel.
- **2** Disconnect the waste fluid sensor cable attached to the cap of the 10L waste tank from the WASTE SENSOR connector on the front of the tank base.



WASTE SENSOR connector

Notes

- Do not touch the connector of the waste tank connection tube.
- When disconnecting the 10L waste tank fluid sensor cable, do not apply excessive force to the fittings or connectors, and take care not to pull the cable with force.

3 Remove the tank 10L waste tank, with cap attached, from the tank base, then dispose of the waste fluid.

Notes

- When moving the 10L waste tank, take care not to pull the waste fluid sensor cable.
- Dispose of the 10L waste tank and waste fluid in accordance with the laboratory rules, and local ordinances and regulations.
- **4** Place an empty 10L waste tank or a new 10L waste tank on the WASTE side of the tank base.
- **5** Connect the connector of the waste fluid sensor cable attached to the cap of the 10L waste tank to the WASTE SENSOR connector on the front of the tank base.

Notes

- Make sure that the waste tank connection tube (waste fluid) is connected securely to the connector on the cap of the 10L waste tank, and that the cap is securely fastened on the 10L waste tank to prevent leakages.
- Always place the 10L waste tank on the WASTE side of the tank base.
- Do not autoclave the 10L waste tank. The shape of the 10L waste tank will deform if placed in an autoclave.
- If the air filter attached to the 10L waste tank becomes clogged, air cannot escape the waste tank as it fills with waste fluid, which may cause incorrect operation. The air filter may become clogged if it gets wet. Do not tilt or otherwise jostle the 10L waste tank if it is close to full.

Refilling the 10L Sheath Tank with Sheath Fluid

Before starting the unit each day, replace any unused sheath fluid remaining in the 10L sheath tank. Also, if the fluid level in the sheath tank becomes low during operation, refill the tank with sheath fluid.

Note

Use only filtered DI water or ultrapure water in the 10L sheath tank. The use of PBS (saline) instead of DI water or ultrapure water is supported, however this requires changes to the instrument setup. If you wish to use PBS (saline), contact your Sony distributor beforehand.

Precautions when handling sheath fluid

- Once a week, discard any remaining unused sheath fluid, wash tank with about 500 ml of sheath fluid, then refill tank with fresh sheath fluid.
- Use sheath fluid uncontaminated by bacteria or biological matter to control the growth of bacteria.

Perform the following procedure with the power supply turned on and ID7000 Software running. To refill the sheath tank before turning the main unit on, start from step **3**.

1 Click [Refill and Empty] in the [Tank Maintenance] group on the [Cytometer] tab of the ribbon.

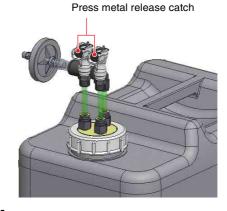
The [Tank Maintenance Wizard] window appears.

2 Click [Start].

Follow the instructions according to the wizard.

3 Press the metal release catch and withdraw the line connectors (two places).

Place the ends of the lines in a container, such as a beaker.



4 Remove the 10L sheath tank from the tank base to allow easy access to the cap of the sheath tank.

5 Place the cap (with nozzle, connector, and tube attached) on a metal plate or other container to prevent sheath fluid spills from the nozzle.



Note

Place the metal plate in a safe position where it cannot be accidentally moved or bumped.

6 Dispose of the remaining sheath fluid in the tank.

Note

Dispose of the sheath fluid in accordance with the laboratory rules and local ordinances and regulations.

- **7** Fill the tank with fresh sheath fluid, and attach the cap.
- **8** Place the 10L sheath tank on the SHEATH side of the tank base, insert the 10L sheath tank line connectors, and push them in until they lock into place.

Clean any fluid from the connector and outside surfaces of the tank.

9 Run a dummy sample and check for leaks around the tube connector, as required.

Priming

You can prime the fluidics system manually. At the start of each day, or after eight hours has elapsed, the priming wizard will appear. Click [Start] in step **2** to perform priming.

This takes approximately nine minutes.

1 Click [Priming] in the [Cleaning] group on the [Cytometer] tab of the ribbon.

The [Priming] wizard appears.

2 Click [Start].

Priming starts.

During priming, a progress bar and status are displayed, allowing you to check the progress and current state.

Note

If [Long Term Storage] processing has been performed, the priming that is executed the first time is different from normal priming. Instead, priming for recovery from [Long Term Storage] is performed. Follow the instructions in the wizard.

Chapter 9 Maintenance

Cleaning the Probe Manually

After acquisition, the probe is cleaned automatically. Under normal circumstances, manual cleaning is usually not required, but can be performed if necessary. During probe cleaning, the fluidics system is backflushed using sheath fluid.

This takes approximately 15 seconds.

1 Click [Probe Wash] in the [Cleaning] group on the [Cytometer] tab of the ribbon.

A confirmation message appears.

2 Click [Yes].

Sheath fluid backflushes the system. The current status and a progress bar are displayed on the bottom left of the window.

For details about cleaning manually using bleach, see "Cleaning Using Bleach Solution" (page 157).

Purging the Flow Cell

Purging the flow cell backflushes a large volume of sheath fluid through the flow cell. The flow cell should be purged whenever foreign matter or air bubbles are suspected in the flow cell. This takes approximately five minutes.

1 Click [Flow Cell Purge] in the [Cleaning] group on the [Cytometer] tab of the ribbon.

The [Flow Cell Purge] wizard appears.

2 Click [Start].

Cleaning starts.

During cleaning, a progress bar and status are displayed, allowing you to check the progress and current state.

Cleaning Using Bleach Solution

Clean the sample fluid fluidics system regularly using a protein-dissolving bleach to disinfect the lines that carry cells and other material for analysis. Disinfect the fluidics system on a regular basis, or as required, especially if running samples that contain, for example, infectious substances.

The sample line, sample probe, and flow cell should be cleaned using a 1% concentration sodium hypochlorite (bleach) solution.

DANGER

A 1% concentration of sodium hypochlorite (bleach) solution is a hazardous substance that may cause serious damage to the eyes and skin. Always wear gloves, mask, goggles, and other protective clothing, as required, when handling.

Tips

- After cleaning with sodium hypochlorite (bleach) solution, a large volume of sheath fluid used for normal cleaning is used to remove all traces of bleach from the system.
- A 1% solution of sodium hypochlorite can be created by diluting household bleach by a factor of about 5.
- 1 Click [Decontamination] in the [Cleaning] group on the [Cytometer] tab of the ribbon, and select [Bleach Cleaning].

Tip

To include a rinse cycle, select [Bleach Cleaning and Rinse]. In this case, prepare commercial rinsing fluid solution for flow cytometers beforehand.

Note

The bleach cleaning sequence cannot be stopped once it is started. Once started, the sequence must be followed to completion.

2 Place 2 ml of 1% concentration Sodium Hypochlorite (bleach) solution in a 5 ml round tube.

Follow the on-screen instructions.

3 Click [Start] to begin cleaning.

The system is cleaned using sodium hypochlorite (bleach) solution and sheath fluid (DI water), in that order.

During cleaning, the current status and a progress bar are displayed in the bottom left of the window.

4 When finished, remove the sample tube.

Notes

- Always fill the 5 ml round tube with the specified volume of sodium hypochlorite (bleach) solution when cleaning with bleach.
- If sample fluid contains a high concentration of contaminants, clean the fluidics system using sodium hypochlorite solution (bleach) after acquisition.

De-bubbling the Sheath Line

If any air enters the sheath line, the flow of sheath fluid may become poor, adversely affecting the exterior cleaning of the sample probe. When contamination between samples is suspected, de-bubble the sheath line.

1 Click [Sheath Line] in the [De-bubble] group on the [Cytometer] tab of the ribbon.

The sheath line de-bubble wizard appears.

2 Click [Start].

Follow the on-screen instructions in the wizard.

Note

For details about the location of the valve required for operation during the wizard, see "*Interior (Right Side Panel*)" (*page 18*) in "Name and Function of Parts".

Maintenance Before Non-use for Extended Periods

After performing preparatory maintenance, shut down the unit.

For details about shutting down, see "Cleaning Before Shutting Down" (page 160).

If using DI water as the sheath fluid

Shut down the instrument using the normal procedure, even if not using it for an extended period.

If using PBS (saline) as the sheath fluid

When the instrument is not used for an extended period, there is a possibility that components in the PBS (saline) may precipitate and cause clogging. If you do not plan to use the instrument for a week or longer, use the [Long Term Storage] procedure to replace the PBS (saline) with DI water in the fluidics flow channels.

For details, see "Running the [Long Term Storage] Process" (page 158).

Note

This instrument is designed for the use of DI water as the sheath fluid, but PBS (saline) is also supported subject to setup changes. Contact your Sony distributor for the required setup changes.

Running the [Long Term Storage] Process

If you do not plan to use the instrument for a week or longer, use the following procedure to replace the PBS with DI water in the fluidics flow channels. This takes approximately 40 minutes.

1 Click [Long Term Storage] in the [Tank Maintenance] group on the [Cytometer] tab of the ribbon.

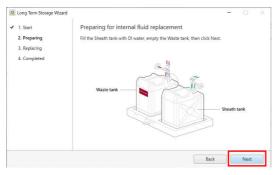


The [Long Term Storage] wizard appears.

2 Click [Start].

3 Fill the 10L sheath tank with DI water, empty the 10L waste tank, and click [Next].

Discard any remaining PBS (saline) in the 10L sheath tank, and then fill the tank with DI water.



Note

Dispose of the sheath fluid and waste fluid in accordance with the laboratory rules and local ordinances and regulations.

The [Long Term Storage] process starts. During the process, a progress bar and status are displayed, allowing you to check the progress and current state.

Note

Shut down the instrument promptly after [Long Term Storage] is executed. If refilling the 10L sheath tank with sheath fluid during this interval, use DI water, not PBS (saline).

When ready to use the instrument again

After the [Long Term Storage] process has been performed, the priming that is performed the first time the instrument is subsequently turned on is a recovery from [Long Term Storage]. Follow the instructions in the wizard.

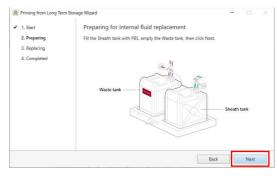
This takes approximately 15 minutes.

1 Click [Start].

Priming from Long Tern	n Storage Wizard		×
1. Start	Start recovery from long term storage		
2. Preparing	Replace internal fluid with PBS. It will take about 10 minutes.		
3. Replacing			
4. Completed			

2 Fill the 10L sheath tank with PBS (saline), empty the 10L waste tank, and click [Next].

Discard any remaining DI water in the 10L sheath tank, and then fill the tank with PBS (saline).



Note

Dispose of the sheath fluid and waste fluid in accordance with the laboratory rules and local ordinances and regulations.

Recovery from [Long Term Storage] starts. During the process, a progress bar and status are displayed, allowing you to check the progress and current state.

Cleaning Before Shutting Down

You should clean the fluidics system before shutting down and turning off the power to the main unit. The preshutdown cleaning is performed using a software wizard. When cleaning is completed, the main unit shuts down automatically.

Clean the main unit using a solution of approximately 1% bleach followed by DI water before shutting down.

DANGER

A 1% concentration of sodium hypochlorite (bleach) solution is a hazardous substance that may cause serious damage to the eyes and skin. Always wear gloves, mask, goggles, and other protective clothing, as required, when handling.

Tip

A 1% solution of sodium hypochlorite can be created by diluting household bleach by a factor of about 5.

1 Click [Hardware Shutdown] in the [Power] group on the [Cytometer] tab of the ribbon, and select [Bleach Cleaning].



The shutdown cleaning wizard opens.

2 Follow the on-screen instructions to clean the instrument.

[Bleach Cleaning] takes approximately eight minutes.

The status appears in the lower left corner of the window.

Tip

To include a rinse cycle after cleaning with a Sodium Hypochlorite (bleach) solution, select [Bleach Cleaning and Rinse].

When cleaning is completed, a message appears to notify you that the unit will shut down.

3 Click [OK].

The main unit shuts down, and the power supply turns off.

Cleaning the Main Unit

Regular Cleaning

Clean the exterior, 5 ml tube holder, and multi-well plate holder (or sample tube holder) using water and ethanol. Cleaning all surfaces prevents the spread of biological matter to ensure instrument performance.

Precautions when handling ethanol

- Ensure the laboratory is well ventilated during cleaning with ethanol. Ethanol vapor can cause irritation of the eyes, skin and respiratory tract, loss of coordination, drowsiness, and in sufficient concentration, unconsciousness.
- Never place near a naked flame or other hazardous material. Ethanol vapor is highly flammable at normal room temperature.

Note

Always wear biology laboratory gloves and other protective clothing, mask, and goggles.

- **1** Turn off the two main power switches on the lower right of the left side panel.
- **2** Moisten a clean, soft, lint-free laboratory-use cloth with clean water, and wipe the multi-well plate holder (or sample tube holder).
- **3** Spray a clean, soft, lint-free laboratory-use cloth with ethanol from a spray bottle, and wipe the multi-well plate holder (or sample tube holder).

If the exterior of the main unit is dirty, clean using a cloth sprayed with ethanol.

4 Clean the 5 ml tube holder using a cloth sprayed with ethanol in the same way.

For light dust etc. on the exterior, wipe with a soft, lint-free cloth.

Notes

- Do not operate ID7000 Software or hardware while cleaning the multi-well plate holder.
- Check 10L tank system for leaks and spills regularly.
- Do not use any substances other than water and ethanol for cleaning.

Replacing the Sheath Filter

Replace the sheath filter in the fluidics module regularly to prevent contamination due to bacteria or dust from entering the fluid flow.



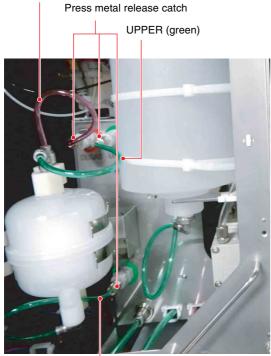
Sheath filter

Perform the following procedure with the main unit shut down and power turned off.

1 Open the fluidics maintenance door on the right side, check that the sheath fluid flow is stopped, and then disconnect the filter lines from the UPPER, LOWER, and DEGAS connectors.

Press the metal release catch and withdraw each connector.

DEGAS (red)



LOWER (green)

The sheath filter connectors have stop valves that prevent leakage of fluid when disconnected.

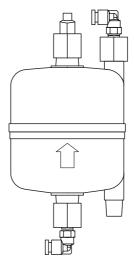
2 Remove the sheath filter from the retaining clamp, and discard the filter.

Note

Discard the sheath filter in accordance with the laboratory rules, and local ordinances and regulations.

3 Place a new sheath filter in the retaining clamp.

Mount the filter with the arrow on the side of the filter facing up.



4 Connect the three fluidics lines from the sheath filter to the corresponding connectors.



LOWER (green)

Connect the line from the bottom of the filter to the LOWER connector, the line from the top of the filter

to the UPPER connector, and the DEGAS connector, in that order. Insert each connector and push it until it locks into place. A click sound occurs when the connectors lock into place.

- **5** Fasten the sheath filter retaining clamp.
- **6** Turn on the power supply to the main unit and start ID7000 Software.
- **7** Prime the fluidics system.

See "Priming" (page 155).

- **8** Close the fluidics maintenance door.
- **9** When finished, click [Close].

Replacing Tank Air Filters

The 10L sheath tank, 10L waste tank, and internal waste tank are fitted with an air filter. The filter allows air to flow into or out of the tank as the fluid level in the tank changes.

The air filter on each tank uses the same type of connector. This section describes the replacement procedure using the sheath tank as an example.

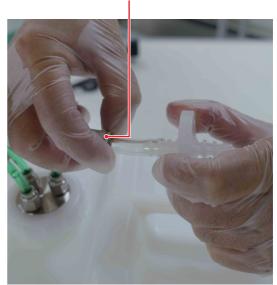
Notes

- The air filter on each tank uses the same type of connector, but the filter type varies. Use only the correct filter for each tank.
- Always wear biology laboratory gloves and other protective clothing, mask, and goggles.
- **1** Shut down the main unit.

Make sure the main unit is completely shut down before proceeding.

2 Grasp the connector, and push in and hold the quick release collet using one hand, then pull out the short length of tube using your other hand.

Exert gentle force if the tube feels difficult to pull out from the connector.



Quick release collet

Υ



3 Insert the replacement air filter tube into the connector and push firmly into place.

Note

The filter may become clogged if fluid gets inside the filter, adversely affecting operation. If any fluid enters the filter during maintenance, replace the filter immediately.

Removing/Reconnecting the Tank Base

If you wish to move the tank base after installation, it may be necessary to disconnect and reconnect the tank base.

Note

If you wish to move the ID7000 main unit, contact your Sony distributor. Do not attempt to move it yourself. The main unit contains high-precision optical and laser instruments that require alignment after installation that only your Sony distributor can provide.

- **1** Turn off the two main power switches on the lower right of the left side panel.
- **2** Disconnect the four tank connection tubes from the 10L tanks.

Press the metal release catch of each connection tube and withdraw the tube.

Notes

- Do not touch the surfaces of the connectors that come into contact with fluids. Take care not to spill any fluid.
- Always wear gloves and other protective clothing, mask, and goggles, as required, when handling waste fluid.
- **3** After moving the tank base, reconnect the four tank connection tubes to the 10L tanks.

Insert each connector and push until it locks into place.

4 Turn on the two main power switches on the lower right of the left side panel of the main unit, then press the POWER button on the front panel to turn on the power supply.

Check for any fluid spills, and clean as necessary.

5 Check the status of the fluid level sensors in the hardware status in the bottom right of the main window in ID7000 Software.

Check the cable electrical connection if the status information does not appear.

Miscellaneous

Appendix

Lineup

- 320 nm laser
- 355 nm laser
- 405 nm laser
- 488 nm laser
- 561 nm laser
- 637 nm laser
- 808 nm laser

Model	No. of lasers	Laser wavelengths (nm)
LE-ID7000A	3	405, 488, 637
LE-ID7000B	4	405, 488, 561, 637
LE-ID7000C	5	355, 405, 488, 561, 637
LE-ID7000D	6	355, 405, 488, 561, 637, 808
LE-ID7000E	6	320, 355, 405, 488, 561, 637
LE-ID7000F	7	320, 355, 405, 488, 561, 637, 808

The fluorescence spectrum that can be detected varies depending on the wavelength of the lasers.

Troubleshooting

Check these items before calling for service. If the problem still persists after carrying out the recommended actions, contact your Sony distributor.

Problem	Possible cause	Recommended solution
Events are not detected or the even rate is abnormally low when the instrument appears	The threshold value is too high.	Decrease the threshold value, and try acquiring again. For details setting the threshold value, see "Configuring Instrument Settings" (page 59).
to be performing acquisitions normally.	Sample flow rate is low.	Increase the sample flow rate, and try acquiring again. For details about setting the sample flow rate, see "Configuring Instrument Settings" (page 59).
	The trigger channel setting is inappropriate.	 Specify a detectable signal for the desired sample as the trigger channel. Check that the signal specified for the trigger channel is a detectable signal (lasers are on, detection voltages, etc.). For details about setting the trigger channel and detectors, see "Configuring Instrument Settings" (page 59).
	The sample concentration is low.	Adjust the sample to the appropriate concentration by increasing the sample, and try acquiring again.
	The sample has precipitated.	Stir the sample thoroughly before acquisition.
	The flow cell is clogged or contains trapped air.	Click [Flow Cell Purge] in the [Cleaning] group on the [Cytometer] tab of the ribbon to backflush the flow cell. If the problem persists, click [Priming] in the [Cleaning] group on the [Cytometer] tab of the ribbon to prime the fluidics system.
	A leak has occurred in the sample flow.	If a leak is confirmed visually, contact your Sony distributor for service.
	The optical alignment is inappropriate.	Run Daily QC. If the result is "Failed," run [Optics Alignment] and [Flow Rate Calibration]. For details, see "Adjusting the Optical Axis" (page 87) and "Calibrating the Fluidics System" (page 87).
Event rate is too high.	The threshold value is too low.	Increase the threshold value, and try acquiring again. For details setting the threshold value, see "Configuring Instrument Settings" (page 59).
	Sample flow rate is high.	Decrease the sample flow rate, and try acquiring again. For details about setting the sample flow rate, see "Configuring Instrument Settings" (page 59).
	The sample concentration is high.	Adjust the sample to the appropriate concentration by diluting the sample, and try acquiring again.

Problem	Possible cause	Recommended solution
Daily QC, Performance QC, Flow Rate Calibration, or Optics Alignment fail to pass.	The beads used for QC have passed their expiry date or were not stored in the proper state.	 Check the expiry date of the beads used for QC, and use only beads prior to their expiry date. Check the beads used for QC are stored correctly.
	Incorrect beads were used or lot information was not selected correctly.	 Use only genuine beads for QC. Set the lot information for the beads used for QC. For details about settings, see "Updating the Beads Lot File" (page 103).
	Preparation of the beads used for QC was inappropriate.	Prepare the specified beads using the specified dilution procedure, and use the beads as soon as possible.
	The flow cell is not clean.	 Click [Flow Cell Purge] in the [Cleaning] group on the [Cytometer] tab of the ribbon to backflush the flow cell. If the
	The flow cell is clogged or contains trapped air.	 problem persists, click [Priming] in the [Cleaning] group on the [Cytometer] tab of the ribbon to prime the fluidics system. Click [Decontamination] in the [Cleaning] group on the [Cytometer] tab of the ribbon, and select [Bleach Cleaning]. <i>For details, see "Cleaning Using Bleach Solution" (page 157).</i>
	30 minutes or more has elapsed since preparation using Align Check beads.	Perform preparation again.
	The optical alignment is inappropriate.	Run Daily QC. If the result is "Failed," run [Flow Rate Calibration] and [Optics Alignment] again, in that order. For details, see "Calibrating the Fluidics System" (page 87) and "Adjusting the Optical Axis" (page 87).
Normal fluorescence spectrum is not observed.	The Fluorescence PMT Voltage is too low.	Increase the Fluorescence PMT Voltage until the fluorescence signal is detected. For details about setup, see "Configuring Instrument Settings" (page 59).
	The signal is saturated because the Fluorescence PMT Voltage is too high.	Increasing the Fluorescence PMT Voltage too much can cause signal saturation, preventing the PMT signal output from changing. (You can check the signal saturation in [Saturation Rate].) In this case, reduce the Fluorescence PMT Voltage and readjust settings to find the optimum operating point. For details about setup, see "Configuring Instrument Settings" (page 59).
	The required lasers are not turned on.	Check that the lasers required for excitation of the fluorochromes to observe are turned on. Turn lasers on if turned off.
	The status of the sample is poor.	Prepare the sample again using the appropriate protocols.
	The target cells for observation are not detected.	Check the threshold value and the gate position, and check that the stained cell population is enclosed correctly.
One population appears separated.	The Biexponential axis setting is inappropriate.	If the setting for the Negative area of the Biexponential axis is inappropriate, the population will appear jammed up against the plot axis origin. Check the setting for the Negative area.
Fluid leaks from the sample suction nozzle.	The flow cell is clogged.	Click [Flow Cell Purge] in the [Cleaning] group on the [Cytometer] tab of the ribbon to backflush the flow cell.
	Air is entering via the connector coupling.	Reconnect the connector, and check to make sure air is not entering via the coupling.
Sample tube does not fit correctly.	The sample tube is non- conforming.	Use only the specified verified sample tubes. The use of non- conforming sample tubes may damage the sample probe.
Sample tube hits the sample probe.		
"Disconnected" is displayed in ID7000 Software, even though the computer and instrument are connected and power is turned on.	The instrument, computer, or ID7000 Software was not started correctly.	Exit ID7000 Software, and turn the instrument and computer off. Check the connections, then turn the instrument and computer on, and start ID7000 Software.

Problem	Possible cause	Recommended solution
Fluidics component (pump, fluid connectors, etc.) is	The valve seals have deteriorated due to age.	Clean areas where leakages occur, and contact your Sony distributor for service.
leaking fluid.		Note Always wear gloves and other protective clothing, mask, and goggles, as required, when cleaning spills and leakages.
CV of acquired data is high.	The flow cell is not clean. The flow cell is clogged or contains trapped air.	 Click [Flow Cell Purge] in the [Cleaning] group on the [Cytometer] tab of the ribbon to backflush the flow cell. If the problem persists, click [Priming] in the [Cleaning] group on the [Cytometer] tab of the ribbon to prime the fluidics system. Click [Decontamination] in the [Cleaning] group on the [Cytometer] tab of the ribbon, and select [Bleach Cleaning]. For details, see "Cleaning Using Bleach Solution" (page 157).
	Sheath filter or air filter is clogged.	Replace the sheath filter or air filter. For details about replacing, see "Replacing the Sheath Filter" (page 161) and "Replacing Tank Air Filters" (page 162).
	The status of the sample is poor.	Prepare the sample again using the appropriate protocols.
	The optical alignment is inappropriate.	Run Daily QC. If the result is "Failed," run [Optics Alignment] and [Flow Rate Calibration]. For details, see "Adjusting the Optical Axis" (page 87) and "Calibrating the Fluidics System" (page 87).
Large numbers of spurious events appear on a plot.	The sheath filter is not clean.	Replace the sheath filter. For details about replacing, see "Replacing the Sheath Filter" (page 161).
	The threshold value is too low.	Increase the threshold value, and try acquiring again. For details setting the threshold value, see "Configuring Instrument Settings" (page 59).
	The fluidics tubes are not clean.	Click [Flow Cell Purge] in the [Cleaning] group on the [Cytometer] tab of the ribbon to backflush the flow cell. If the problem persists, click [Priming] in the [Cleaning] group on the [Cytometer] tab of the ribbon to prime the fluidics system.
	The sheath fluid is contaminated by bacteria.	Discard old sheath fluid, clean the 10L sheath tank, and then refill the tank with fresh sheath fluid. Run the priming function when finished.
Error message is displayed in ID7000 Software.	For details about resolvir	ng each error message, see "Error Messages" (page 169).
No output from the lasers (laser temperature error).	The temperature of the laser module exceeds the allowed range.	If the laser operating temperature exceeds 40 °C (104 °F), equivalent to an ambient operating temperature of 35 °C (95 °F), the lasers shutdown automatically and an error message is displayed. To clear the error, shut down the main unit and wait for the ambient temperature to cool sufficiently before restarting the main unit. If the error persists, even after allowing the ambient temperature to cool below 35 °C (95 °F), contact your Sony distributor for service. If the ambient temperature drops below 10 °C (50 °F), the lasers shut down automatically and an error message is displayed. To clear the error, shut down the main unit and wait for the ambient temperature to increase sufficiently before restarting the main unit. If the error persists, even after the temperature has risen above 17.5 °C (64 °F), contact your Sony distributor for service.
Waste fluid is not being collected in the 10L waste tank.	The waste fluid connection tubes (red) are not connected correctly.	Waste fluid may be leaking due to an unconnected or unsecured waste fluid connection tube on the WASTE or WASTE WASH connector. Check the connections are secure.
	Waste tank air filter is clogged. (10L waste tank is bulging at the sides).	Replace the waste tank air filter. For details about replacing, see "Replacing Tank Air Filters" (page 162).

Problem	Possible cause	Recommended solution
Sheath fluid is not being drawn into the instrument from the 10L sheath tank.	The sample line is not connected correctly.	Sheath fluid may not be drawn into the unit due to an unconnected or unsecured sheath fluid connection tube (green) on the SHEATH or SHEATH WASH connector. Check the connections are secure.
	The sheath filter is clogged.	Replace the sheath filter. For details about replacing, see "Replacing the Sheath Filter" (page 161).
ID7000 Software operation is slow.	The number of events displayed is high.	Reduce the number of events to display to improve performance.
	The number of plots, gates, and statistics is high.	Delete the plots, gates, and statistics not required for the current acquisition to improve performance.
	A multicolor spectral unmixing calculation is in progress.	Turn off spectral unmixing, when it is not required, to improve performance.
	There are a large number of plates in a single experiment.	ID7000 Software operation may become slower as the number of plates in a single experiment increases. Reduce the number of plates in an experiment, for example, by splitting the experiment.
	The number of experiments in ID7000 Software is high.	ID7000 Software operation may become slower as the number of experiments displayed in ID7000 Software increases. Export any experiments not required for reference purposes, and then delete the experiments from ID7000 Software.
Contamination between samples is suspected.	Air has entered the sheath line.	If any air enters the sheath line, the flow of sheath fluid may become poor, adversely affecting the exterior cleaning of the sample probe. If this occurs, de-bubble the sheath line. <i>For details about operation, see "De-bubbling the Sheath Line"</i> (page 158).

Error Messages

This section lists the error messages that may be displayed in ID7000 Software. When an error occurs, check these items for the actions to take to resolve each error. If the problem still persists after carrying out the recommended actions, make a note of the error message and then contact your Sony distributor.

ID	Error message	Solution
00-102	An unexpected error has occurred.	A system error was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-111	External tank system is not detected. Follow the operator's guide to fix the connection with the external tank system.	After the main unit shuts down, check the electrical connection between the main unit and the external tank system, and then restart the main unit. If the problem persists, contact your Sony distributor for service.
00-115	Water leakage is detected.	Open the fluidics maintenance door on the right side, and check for leakages. If the problem persists, contact your Sony distributor for service.
00-120	The laser initialization timed out. Please check whether room temperature is within the specification.	The ambient operating temperature may have exceeded the limit. After the main unit shuts down, allow the ambient temperature to fall before restarting the unit. If the problem persists at temperatures that do not exceed 35 °C (50 °F), contact your Sony distributor for service.
00-121	System power supply error on the MD4 board.	After the main unit shuts down, wait a while before
00-122	AC/DC power supply error on the MD4 board.	restarting. If the problem persists, contact your Sony distributor for
00-166	Failed to update EEPROM on LDD board.	service.
00-168	Communication error on the MD4 board.	
00-169	Communication error on the MD1 board.	
00-170	Communication error on the OMD board.	
00-171	Communication error on the MD3 board.	
00-172	Communication error on the LDD board (CPU1).	
00-173	Communication error on the MD3 board (CPU2).	
00-174	No Reply error on the MD4 board.	
00-175	No Reply error on the MD1 board.	
00-176	No Reply error on the OMD board.	
00-177	No Reply error on the MD3 board (CPU1).	
00-178	No Reply error on the LDD board (CPU1).	
00-179	No Reply error on the MD3 board (CPU2).	
00-189	The model number is invalid.	
00-190	The laser configuration is invalid.	
00-196	Flash write error.	
00-212	Warning for the unsuccessful communication on MD4 board.	
00-213	Warning for the unsuccessful communication on MD1 board.	
00-214	Warning for the unsuccessful communication on OMD board.	
00-215	Warning for the unsuccessful communication on MD3 board.	
00-216	Warning for the unsuccessful communication on LDD board (CPU1).	
00-217	Warning for the unsuccessful communication on MD3 board (CPU2).	

ID	Error message	Solution
00-228	The sheath filter may need to be exchanged.	Sheath filter is due for scheduled replacement. Contact your Sony distributor for service.
00-261	The optical stage might have reached its limit range of motion.	A system error was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-1018	Temperature of MD4 power board is too high. Please check whether room temperature is within the specification.	The main unit may be overheating. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-1047	Failed to update EEPROM on MD1 board.	After the main unit shuts down, wait a while before
00-1048	Failed to update EEPROM on OMD board.	restarting. If the problem persists, contact your Sony distributor for
00-1049	Failed to update EEPROM on MD3 board.	service.
00-1051	Notch 637 in movement error.	
00-1052	Notch 637 out movement error.	
00-1068	Notch 808 in movement error.	
00-1069	Notch 808 out movement error.	
00-2000	Stop button was pressed.	The STOP button on the main unit was pressed.
00-2005	Internal waste tank is full.	After the main unit shuts down, wait a while before
00-2006	Fan1 on MD4 board is not working properly.	restarting. If the problem persists, contact your Sony distributor for
00-2007	Fan2 on MD4 board is not working properly.	service.
00-2008	Fan3 on MD4 board is not working properly.	
00-2009	Temperature is too high in fluidics area. Please check whether room temperature is within the specification.	The main unit may be overheating. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-2012	Temperature of the sheath filter area is low. Please check whether room temperature is within the specification.	The main unit may be too cold. After the main unit shuts down, allow the temperature to rise before restarting. If the problem persists at temperatures exceeding 10 °C
00-2013	Temperature of MD4 power board is low. Please check whether room temperature is within the specification.	(50 °F), contact your Sony distributor for service.
00-3000	Washing unit movement error.	Check that the sample probe is not bent, and wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3001	Auto sampler movement error.	After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3002	Sample probe collision is detected.	Check that the sample probe is not bent, and wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3003	Work height detection error.	After the main unit shuts down, load the cooling unit. If the problem persists, contact your Sony distributor for service.
00-3004	Notch 561 in movement error.	After the main unit shuts down, wait a while before
00-3005	Notch 561 out movement error.	restarting. If the problem persists, contact your Sony distributor for service.
00-3006	Sample probe detection error, the probe might be bent.	Check that the sample probe is not bent, and wait a while before restarting. If the problem persists, contact your Sony distributor for service.

ID	Error message	Solution
00-3008	Door is open. Please close cytometer door. The cytometer was terminated to avoid further accidents.	After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3009	Auto sampler x-axis sensor error.	
00-3010	Auto sampler y-axis sensor error.	
00-3012	Temperature of the laser area is too high. Please check whether room temperature is within the specification.	The main unit may be overheating. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3013	Temperature of the laser area is too low. Please check whether room temperature is within the specification.	The main unit may be too cold. After the main unit shuts down, allow the temperature to rise before restarting. If the problem persists at temperatures exceeding 10 $^{\circ}$ C (50 $^{\circ}$ F), contact your Sony distributor for service.
00-3014	Auto sampler overrun error.	After the main unit shuts down, wait a while before
00-3016	Washing unit overrun error.	restarting. If the problem persists, contact your Sony distributor for service.
00-3017	Sample probe detection error.	Check whether there are any droplets or deformation on the sample probe. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3018	Failed to run pump LP4.	A device fault (pump LP4) was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3019	Failed to run pump LP2.	A device fault (pump LP2) was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3020	Failed to run pump LP1.	A device fault (pump LP1) was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3021	Failed to run pump AP1.	A device fault (pump AP1) was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3022	Failed to run pump AP2.	A device fault (pump AP2) was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3023	Failed to run pump LP3.	A device fault (pump LP3) was detected. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3025	Failed to update EEPROM on MD3 board (CPU2).	After the main unit shuts down, wait a while before
00-3030	Motor movement error.	restarting. If the problem persists, contact your Sony distributor for
00-3031	Thermal sensor error.	service.
00-3032	Auto sampler sensor error in initialization.	1
00-3034	Washing unit sensor error in initialization.	1
00-3035	Discharge time out.	The pressurized tank discharge operation has timed out. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.

ID	Error message	Solution
00-3036	Communication error on the LDD board (CPU2).	After the main unit shuts down, wait a while before
00-3037	No Reply error on the LDD board (CPU2).	restarting. If the problem persists, contact your Sony distributor for
00-3038	Communication error on the MCY board.	service.
00-3041	Failed to Shutter Close.	After the main unit shuts down, wait a while before
00-3042	Failed to Shutter Open.	restarting. If the problem persists, contact your Sony distributor for
00-3043	Charging air tank time out.	service.
00-3044	No Reply error on the MCY board.	
00-3045	Command_B Error on the Deck (488nm).	
00-3046	Command_B Error on the Deck (405nm).	
00-3047	Command_B Error on the Deck (637nm).	
00-3048	Command_B Error on the Deck (561nm).	
00-3049	Command_B Error on the Deck (355nm).	
00-3050	Command_B Error on the Deck (320nm).	
00-3051	Command_B Error on the any Deck.	
00-3052	Command_L Error on the Deck (488nm).	
00-3053	Command_L Error on the Deck (405nm).	
00-3054	Command_L Error on the Deck (637nm).	
00-3055	Command_L Error on the Deck (561nm).	
00-3056	Command_L Error on the Deck (355nm).	
00-3057	Command_L Error on the Deck (320nm).	
00-3065	Temperature error of the 488nm laser. Please check whether room temperature is within the specification.	The ambient operating temperature may be too high (too low). After the main unit shuts down, allow the ambient temperature to fall (rise) before restarting the unit.
00-3066	Temperature error of the 405nm laser. Please check whether room temperature is within the specification.	If the problem persists at temperatures that do not exceed 27.5 °C (81.5 °F), contact your Sony distributor for service.
00-3067	Temperature error of the 637nm laser. Please check whether room temperature is within the specification.	
00-3068	Temperature error of the 561nm laser. Please check whether room temperature is within the specification.	
00-3069	Temperature error of the 355nm laser. Please check whether room temperature is within the specification.	
00-3070	Temperature error of the 808nm laser. Please check whether room temperature is within the specification.	
00-3071	Temperature error of the 320nm laser. Please check whether room temperature is within the specification.	

ID	Error message	Solution
00-3079	Failed to detect the 488nm laser.	After the main unit shuts down, wait a while before
00-3080	Failed to detect the 405nm laser.	restarting. If the problem persists, contact your Sony distributor for
00-3081	Failed to detect the 637nm laser.	service.
00-3082	Failed to detect the 561nm laser.	
00-3083	Failed to detect the 355nm laser.	
00-3084	Failed to detect the 808nm laser.	
00-3085	Failed to detect the 320nm laser.	
00-3086	Air tank pressure is too low.	
00-3089	Failed to run the optical Y motor.	
00-3090	Failed to run the optical Z motor.	
00-3091	Failed to run the washing motor.	
00-3092	Failed to run the stage X motor.	
00-3093	Failed to run the stage Y motor.	
00-3094	Failed to run the stage Z motor.	
00-3097	Power supply error on MD4 board 5V.	
00-3098	Power supply error on MD4 board 3.3V.	
00-3099	12V power supply error on LDD board.	
00-3100	24V power supply error on LDD board.	
00-3101	12V power supply error on OMD board.	
00-3102	24V power supply error on OMD board.	
00-3103	12V power supply error on MD3 board.	
00-3104	24V power supply error on MD3 board.	
00-3105	5V power supply error on MD3 board.	
00-3106	12V power supply error on MD1 board.	
00-3107	24V power supply error on MD1 board.	
00-3108	5V DC power supply error on MD1 board.	
00-3110	High temperature detected on the multi-well plate holder. Please check whether room temperature is within the specification.	The main unit may be overheating. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-3113	Temperature of the Deck (488nm) area is too high. Please check whether room temperature is within the specification.	
00-3114	Temperature of the Deck (405nm) area is too high. Please check whether room temperature is within the specification.	
00-3115	Temperature of the Deck (637nm) area is too high. Please check whether room temperature is within the specification.	
00-3116	Temperature of the Deck (561nm) area is too high. Please check whether room temperature is within the specification.	
00-3117	Temperature of the Deck (355nm) area is too high. Please check whether room temperature is within the specification.	
00-3118	Temperature of the Deck (320nm) area is too high. Please check whether room temperature is within the specification.	

ID	Error message	Solution
00-3119	Model config on MCY and SKY do not match.	After the main unit shuts down, wait a while before
00-3120	The 488nm laser error is detected.	restarting. If the problem persists, contact your Sony distributor for
00-3121	The 405nm laser error is detected.	service.
00-3122	The 637nm laser error is detected.	
00-3123	The 561nm laser error is detected.	
00-3124	The 355nm laser error is detected.	
00-3125	The 808nm laser error is detected.	
00-3126	The 320nm laser error is detected.	
00-4000	Remove the sample plate or tube rack.	Remove the multi-well plate or tube rack, and close the door.
00-4001	Internal sheath tank is empty.	After the main unit shuts down, wait a while before
00-4002	Internal vacuum tank is full.	restarting. If the problem persists, contact your Sony distributor for service.
00-4006	Door is open.	The door is open. Close the door. Initialize the auto sampler.
00-4007	Sample probe collision is detected.	A collision with the sample probe occurred. Initialize the auto sampler.
00-4010	No plate or rack	The multi-well plate or tube rack could not be detected. Load the multi-well plate or tube rack.
00-4011	Failed to control sample pressure. Please execute priming.	The sample suction pressure is not stable. Run system priming.
00-4012	Failed to control sample pressure. Please execute priming.	If the problem persists, contact your Sony distributor for service.
00-4013	Washing unit step-out.	A position error of the washing unit was detected. Initialize the washing unit.
00-4015	External sheath tank is empty.	Refill the sheath tank with sheath fluid.
00-4016	External waste tank is full.	Dispose of the waste fluid from the waste tank.
00-4017	Replacement of the pump LP4 is recommended.	The pump running time exceeds 10,000 hours. Contact
00-4018	Replacement of the pump LP2 is recommended.	your Sony distributor for service.
00-4019	Replacement of the pump LP1 is recommended.	
00-4020	Replacement of the pump AP1 is recommended.	
00-4021	Replacement of the pump AP2 is recommended.	
00-4022	Replacement of the pump LP3 is recommended.	The pump running time exceeds 4,800 hours. Contact your Sony distributor for service.
00-4023	Auto sampler overrun occurred.	A position error of the auto sampler unit was detected. Initialize the auto sampler unit.
00-4025	Washing unit overrun occurred.	A position error of the washing unit was detected. Initialize the washing unit. If the problem persists, contact your Sony distributor for service.
00-4028	Refill time out. Please check the sheath line.	The refill operation for the pressurized tank has timed out. Shut down the main unit, check the connections of the sheath tank connection tubes (green), and then restart the main unit. If the problem persists, contact your Sony distributor for service.
00-4029	Failed to control sheath pressure by the pressure tank.	Sheath fluid flow is unstable. Run system priming.
00-4030	Failed to control sheath pressure by the vacuum tank.	
00-4031	Failed to control washing unit pressure. Replacement of air filter for waste tank is recommended.	The waste tank air filter may be clogged. Replacement of the air filter is recommended. If the problem persists, contact your Sony distributor for service.

ID	Error message	Solution
00-4033	Failed to control washing unit pressure.	A pressure error was detected during cleaning. Replacement of the waste tank air filter is recommended. If the problem persists, contact your Sony distributor for service.
00-4034	Replacement of the loader cable is recommended.	The number of motion cycles of the motor cable exceeds the endurance limit. Contact your Sony distributor for service.
00-4039	External sheath tank is empty.	Refill the sheath tank with sheath fluid.
00-4040	External waste tank is full.	Dispose of the waste fluid from the waste tank.
00-4041	Warning for the unsuccessful communication on LDD board (CPU1).	After the main unit shuts down, wait a while before restarting.
00-4044	Warning for the unsuccessful communication on MCY board.	If the problem persists, contact your Sony distributor for service.
00-4048	Fan on MCY board is not working properly.	
00-4049	Fan on SKY board is not working properly.	
00-4050	Fan on the Deck (488nm) is not working properly.	
00-4051	Fan on the Deck (405nm) is not working properly.	
00-4052	Fan on the Deck (637nm) is not working properly.	
00-4053	Fan on the Deck (561nm) is not working properly.	
00-4054	Fan on the Deck (355nm) is not working properly.	
00-4055	Fan on the Deck (320nm) is not working properly.	
00-4056	Fan on the LD_COMMON is not working properly.	
00-4057	Fan on 488LD is not working properly.	
00-4058	Fan on 405LD is not working properly.	
00-4059	Fan on 637LD is not working properly.	
00-4060	Fan on 561LD is not working properly.	
00-4061	Fan on 355LD is not working properly.	
00-4062	Fan on 808LD is not working properly.	
00-4063	Fan on 320LD is not working properly.	
00-4064	Fan on sample temperature controlling radiator is not working properly.	
00-4065	Fan on AS MD3 is not working properly.	
00-4066	Nozzle agitation is not working properly.	Check that the sample probe is not bent, and wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-4074	Temperature of the laser area is high. Please check whether room temperature is within the specification.	The main unit may be overheating. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-4087	Jackal versions do not match.	After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-4088	Failed to cool the multi-well plate holder. Please disable cooling control.	Temperature of the sample stage cannot be adjusted. After the main unit shuts down, wait a while before restarting. If the problem persists, contact your Sony distributor for service.
00-4090	Failed to control sample pressure. Please execute priming.	The sample suction pressure is not stable. Run system priming. If the problem persists, contact your Sony distributor for service.
01-0100	Failed to align optical axis. Please run Flow Rate Calibration, then try again.	Optics alignment failed. Run flow rate calibration.

ID	Error message	Solution
01-0200	Failed to adjust PMT voltage. Please run Flow Cell Purge or Decontamination or Optics Alignment, then try again.	PMT voltage calibration failed. Run flow cell purge or optics alignment.
01-0300	Failed to adjust PMT voltage. Please run Flow Cell Purge or Decontamination or Optics Alignment, then try again.	PMT voltage calibration failed. Run flow cell purge or optics alignment.
01-1100	Optics alignment position is far from the latest position. Bubble may cause this issue, so Flow Cell Purge and trying again are recommended.	The result of optics alignment are significantly different from the previous alignment. Run flow cell purge, and then run optics alignment again.
01-2001	Temperature in fluidics area is out of range for Daily QC. Please check whether room temperature is within the specification.	The sheath fluid is outside the rated temperature range, and Daily QC cannot be started. Check that the room temperature is within the rated range of the unit.
01-2002	Temperature in fluidics area is out of range for Flow Rate Calibration. Please check whether room temperature is within the specification.	The sheath fluid is outside the rated temperature range, and Flow Rate Calibration cannot be started. Check that the room temperature is within the rated range of the unit.
01-2003	Temperature in fluidics area is out of range for acquisition. It may affect performance of the instrument. Are you sure you want to start acquisition?	The sheath fluid is outside the rated temperature range, which may adversely affect unit performance. Check that the room temperature is within the rated range of the unit before starting data acquisition.
01-2004	Temperature in fluidics area is out of range for auto acquisition. It may affect performance of the instrument. Are you sure you want to start auto acquisition?	
01-2005	Temperature in fluidics area is out of range for preview. It may affect performance of the instrument. Are you sure you want to start preview?	
01-2101	Temperature in fluidics area has changed 5°C/9°F from latest Daily QC had passed. It may affect performance of the instrument. Please run Daily QC, then try again.	The temperature of the surroundings has changed significantly since Daily QC was run. Run Daily QC again.

US FDA 21 CFR Part 11 Support

ID7000 software supports functions relevant to 21 CFR Part 11. The software in the table below refers to the ID7000 software.

Section	Requirement	Compliance
11.10	Controls for Closed Systems Persons who use closed systems to create, modify, maintain, or transmit electronic records shall employ procedures and controls designed to ensure the authenticity, integrity, and, when appropriate, the confidentiality of electronic records, and to ensure that the signer cannot readily repudiate the signed record as not genuine. Such procedures and controls shall include the following:	
11.10(a)	Validation of systems to ensure accuracy, reliability, consistent intended performance, and the ability to discern invalid or altered records.	The software is released following validation and verification testing conducted by Sony. IQ and OQ procedures and QC functions are available for system validation.
11.10(b)	The ability to generate accurate and complete copies of records in both human readable and electronic form suitable for inspection, review, and copying by the agency. Persons should contact the agency if there are any questions regarding the ability of the agency to perform such review and copying of the electronic records.	The software provides both electronic and human readable formats of the records (e.g., worksheets, reports, audit trails, user logs). Once exported outside the system, the user must take needed steps to ensure protection of data and records.
11.10(c)	Protection of records to enable their accurate and ready retrieval throughout the records retention period.	Records are secured within a hidden folder structure controlled by an indexing database. All records are retained until the user with appropriate privileges decides to delete records. Records that are exported from the software must be managed by the user according to prevailing policies and processes to ensure data protection.
11.10(d)	Limiting system access to authorized individuals.	The software requires a password for all users when logging in. Each user has a defined role, associated with access privileges. Users and their roles must be managed by administrators.
11.10(e)	Use of secure, computer-generated, time-stamped audit trails to independently record the date and time of operator entries and actions that create, modify, or delete electronic records. Record changes shall not obscure previously recorded information. Such audit trail documentation shall be retained for a period at least as long as that required for the subject electronic records and shall be available for agency review and copying.	The software automatically generates a secure, computer-generated, time-stamped audit trail of all user actions that affect the record. Once turned on, users cannot modify, deactivate or delete the audit trail. The audit trail is maintained with the record through the lifetime of said record.
11.10(f)	Use of operational system checks to enforce permitted sequencing of steps and events, as appropriate.	The device restricts what users can do based on the state of the system and the permissions associated with the account which is logged in.
11.10(g)	Use of authority checks to ensure that only authorized individuals can use the system, electronically sign a record, access the operation or computer system input or output device, alter a record, or perform the operation at hand.	The software is accessed using a valid username and password. Based on the role and privileges associated with the account, the software provides the users with the authority to carry out particular functions. These privileges include tasks such as electronic signature application to a record, deletion of record or export of records.
11.10(h)	Use of device (e.g., terminal) checks to determine, as appropriate, the validity of the source of data input or operational instruction.	The software has been designed to require specific inputs for given fields of information - and it includes error messaging capability that informs the user when invalid information is input into a software field. The software also confirms the validity of the record by attaching a checksum value to the record when it is exported. Before a record can be imported, the software checks the value and restricts the import if the value has changed since export.

Section	Requirement	Compliance
11.10(i)	Determination that persons who develop, maintain, or use electronic record/electronic signature systems have the education, training, and experience to perform their assigned tasks.	Sony can provide user training and documentation for operation of the system and software. It is the responsibility of the user's organization to ensure training of staff and providing needed information on the record/electronic signature operating procedure based on their own policies and procedures.
11.10(j)	The establishment of, and adherence to, written policies that hold individuals accountable and responsible for actions initiated under their electronic signatures, in order to deter record and signature falsification.	This is the responsibility of the user's organization. Written policies and procedures should be defined that outline the importance of electronic signatures with respect to individual responsibility and the consequences of tampering for both the company and the individual.
11.10(k)	Use of appropriate controls over systems documentation including: (1) Adequate controls over the distribution of, access to, and use of documentation for system operation and maintenance. (2) Revision and change control procedures to maintain an audit trail that documents time-sequenced development and modification of systems documentation.	The software has a built-in help function that displays user manuals and is available to all users. The user manuals incorporated in the help function will be updated appropriately with system software updates. All updates are made available to end users to ensure optimal system operation through the field support organization.
11.30	Controls for Open Systems	
	Persons who use open systems to create, modify, maintain, or transmit electronic records shall employ procedures and controls designed to ensure the authenticity, integrity, and, as appropriate, the confidentiality of electronic records from the point of their creation to the point of their receipt. Such procedures and controls shall include those identified in § 11.10, as appropriate, and additional measures such as document encryption and use of appropriate digital signature standards to ensure, as necessary under the circumstances, record authenticity, integrity, and confidentiality.	The software is designed to operate as a closed system and therefore does not apply.
11.50	Signature Manifestations	
11.50(a)	Signed electronic records shall contain information associated with the signing that clearly indicates all of the following: (1) The printed name of the signer; (2) The date and time when the signature was executed; and (3) The meaning (such as review, approval, responsibility, or authorship) associated with the signature.	All signed records (electronic and human readable forms) within the software contain the username of the signer, the date and time of the signing and the authorship.
11.50(b)	The items identified in paragraphs $(a)(1)$, $(a)(2)$, and $(a)(3)$ of this section shall be subject to the same controls as for electronic records and shall be included as part of any human readable form of the electronic record (such as electronic display or printout).	
11.70	Signature/Record Linking	
	Electronic signatures and handwritten signatures executed to electronic records shall be linked to their respective electronic records to ensure that the signatures cannot be excised, copied, or otherwise transferred to falsify an electronic record by ordinary means.	Records are created in PDF format and electronic signatures are applied via PDF functionality. The PDF function ensures that electronic signatures cannot be reused.
11.100	General Requirements	
11.100(a)	Each electronic signature shall be unique to one individual and shall not be reused by, or reassigned to, anyone else.	The software ensures that every combination of username and password is unique to the user. If the username is disabled, it cannot be reused for other users. It is the responsibility of the user's organization to verify the identity of all users who use the software.

Section	Requirement	Compliance
11.100(b)	Before an organization establishes, assigns, certifies, or otherwise sanctions an individual's electronic signature, or any element of such electronic signature, the organization shall verify the identity of the individual.	Responsibility of the user's organization.
11.100(c)	Persons using electronic signatures shall, prior to or at the time of such use, certify to the agency that the electronic signatures in their system, used on or after August 20, 1997, are intended to be the legally binding equivalent of traditional handwritten signatures. (1) The certification shall be submitted in paper form and signed with a traditional handwritten signature, to the Office of Regional Operations (HFC-100), 5600 Fishers Lane, Rockville, MD 20857. (2) Persons using electronic signatures shall, upon agency request, provide additional certification or testimony that a specific electronic signature is the legally binding equivalent of the signer's handwritten signature.	Responsibility of the user's organization.
11.200	Electronic Signature Components and Controls	
11.200(a)	Electronic signatures that are not based upon biometrics shall: (1) Employ at least two distinct identification components such as an identification code and password. (i) When an individual executes a series of signings during a single, continuous period of controlled system access, the first signing shall be executed using all electronic signature components; subsequent signings shall be executed using at least one electronic signature component that is only executable by, and designed to be used only by, the individual. (ii) When an individual executes one or more signings not performed during a single, continuous period of controlled system access, each signing shall be executed using all of the electronic signature components. (2) Be used only by their genuine owners; and (3) Be administered and executed to ensure that attempted use of an individual's electronic signature by anyone other than its genuine owner requires collaboration of two or more individuals.	The software requires a unique username and password to apply an electronic signature. Both components are necessary for each signing, separately from login.
11.200(b)	Electronic signatures based upon biometrics shall be designed to ensure that they cannot be used by anyone other than their genuine owners.	The software does not support biometric signatures.
11.300	Controls for Identification Codes/Passwords Persons who use electronic signatures based upon use of identification codes in combination with passwords shall employ controls to ensure their security and integrity. Such controls shall include:	
11.300(a)	Maintaining the uniqueness of each combined identification code and password, such that no two individuals have the same combination of identification code and password.	No two individuals can have the same combinations of username and password. The combinations of username and password must be unique.
11.300(b)	Ensuring that identification code and password issuances are periodically checked, recalled, or revised (e.g., to cover such events as password aging).	Passwords are set to expire when they are first created or reset. The password expiration period can be set by the administrator.
11.300(c)	Following loss management procedures to electronically deauthorize lost, stolen, missing, or otherwise potentially compromised tokens, cards, and other devices that bear or generate identification code or password information, and to issue temporary or permanent replacements using suitable, rigorous controls.	Administrators can delete accounts or change compromised passwords as needed. The user must reset their password on the first login after the change to ensure that the administrator does not own the password.

Section	Requirement	Compliance
11.300(d)	Use of transaction safeguards to prevent unauthorized use of passwords and/or identification codes, and to detect and report in an immediate and urgent manner any attempts at their unauthorized use to the system security unit, and, as appropriate, to organizational management.	After a specified number of consecutive failed login attempts, the software locks out the problematic user. The specified number of attempts can be set by the administrator to suit the operation of the user's organization.
11.300(e)	Initial and periodic testing of devices, such as tokens or cards, that bear or generate identification code or password information to ensure that they function properly and have not been altered in an unauthorized manner.	Periodic checks are the responsibility of the user. Administrators are encouraged to periodically check for access restrictions and unauthorized data manipulation.

Replacement Parts

For details about purchasing replacement parts, contact your Sony distributor.

Item	Part number
Align Check Beads	LE-AE700510
8-peak beads	LE-AE700522
Autosampler compatible 5 ml tube rack	LE-A8101
Sheath filter kit	LE-U5F001
10L Sheath tank	LE-U5F006
10L Waste tank	LE-U5F007
10L Sheath tank cap set	LE-U5F008
10L Waste tank cap set	LE-U5F009
Sheath tank connection tube	LE-U8F001
Waste tank connection tube	LE-U8F002
External tank connection cable	LE-U8F003
Sheath tank air filter	LE-U3F014
Waste tank air filter	LE-U3F011A
10L tank system	LE-K5105

Software Licenses

GPL

This product uses "U-Boot" software under the GPL license. The customer is entitled to obtain, modify, and redistribute the source code for the software.

Source code can be obtained from the

"\Others\SourceCode" folder on the supplied installation disc.

However, Sony cannot accept inquiries regarding the content of these source codes.

For license information, refer to the "\Others\License" folder on the supplied installation disc.

Apache License

This product uses the Apache license software, "ZXing," "Unity," and "Prism Framework." For license information, refer to the \Others\License folder on the supplied installation disc.

Microsoft Public License (Ms-PL)

This product uses the Microsoft Public License (Ms-PL) software, "CommonServiceLocator," "Enterprise Library," "WPF Toolkit," and "DotNetZip." For license information, refer to the \Others\License folder on the supplied installation disc.

The MIT License (MIT)

This product uses the MIT License software, "Fluent Ribbon Control Suite." For license information, refer to the \Others\License folder on the supplied installation disc.

Third-party Software

This product uses third-party software, "Entity Framework," ".NET Framework," "Visual C++ Redistributable Packages," "Microsoft SQL Server," and "DioDocs" (Copyright (C) 2018 GrapeCity Inc.). For license information, refer to the \Others\License folder on the supplied installation disc.

Software End User License Agreement

For Users in USA

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You agree to comply with all applicable local, federal and state laws, rules and regulations (including, without limitation, those relating to healthcare). You agree to take sole responsibility for compliance with the Health Insurance Portability and Accountability Act of 1996, as amended, and the regulations relating thereto, and agree to hold SONY harmless and indemnify SONY from any and all claims or damages arising out of your failure so to do. For the avoidance of doubt, SONY shall not have any access to, or obligations with respect to, any protected health information of any patient as a result of this Agreement.

You represent and warrant that you, and the organization for whom you work, have not, and will not employ or contract with any person sanctioned or excluded from participation with any federal, state or local health care program (including without limitation Medicare or Medicaid) or debarred from contracting with any federal or state agency. Furthermore, you represent and warrant that you have routinely checked all employees and contractors against the Department of Health and Human Services Office of Inspector General's List of Excluded Individuals/Entities and the General Services Administration's Excluded Parties list System at least as frequently as required or recommended by any health care program or state agency with jurisdiction over you or the company with which you work.

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Except as prohibited by applicable law, you agree to indemnify and hold harmless SONY and the THIRD-PARTY SUPPLIERS and their respective subsidiaries, affiliates, officers and employees, from any loss, claim or demand, including reasonable attorneys' fees, made by any third party due to or arising out of your use of the SOFTWARE and/or the DEVICE (including, without limitation, any software vulnerability caused by such use), your violation of this EULA or your failure to fulfill your responsibility under this EULA.

UPDATES

From time to time, SONY or the THIRD-PARTY SUPPLIERS may automatically update or otherwise modify the SOFTWARE, including, but not limited to, for purposes of enhancement of security functions, error correction and improvement of functions, at such time as you interact with SONY's or third parties' servers, or otherwise. Such updates or modifications may delete or change the nature of features or other aspects of the SOFTWARE, including, but not limited to, functions you may rely upon. You acknowledge and agree that such updates are made at SONY's sole discretion and that SONY may condition continued use of the SOFTWARE upon your complete installation or acceptance of such update or modifications. Any updates/modifications shall be deemed to be, and shall constitute part of, the SOFTWARE for purposes of this EULA. By acceptance of this EULA, you consent to such update/modification.

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SONY shall not be liable for any delay or failure to perform resulting from causes outside its reasonable control, including, but not limited to, acts of God, war, terrorism, riots, embargos, acts of civil or military authorities, fire, floods, accidents, strikes or shortages of transportation facilities, fuel, energy, labor or materials.

ENTIRE AGREEMENT, WAIVER, SEVERABILITY

This EULA as amended and modified by SONY from time to time, constitutes the entire agreement between you and SONY with respect to the SOFTWARE. The failure of SONY to exercise or enforce any right or provision of this EULA shall not constitute a waiver of such right or provision. If any part of this EULA is held invalid, illegal, or unenforceable, that provision shall be enforced to the maximum extent permissible so as to maintain the intent of this EULA, and the other parts will remain in full force and effect.

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The United Nations Convention on Contracts for the International Sale of Goods shall not apply to the SOFTWARE or this EULA. Furthermore, this EULA will not be governed or interpreted in any way by referring to any law based on the Uniform Computer Information Transactions Act (UCITA) or any other act derived from or related to UCITA. This EULA, any disputes or claims arising out of or related to this EULA and your relationship with SONY under this EULA, shall be governed by and construed in accordance with the laws of the State of California, United States, without regard to its conflict of laws principles. Any and all disputes or claims arising out of or relating to this EULA or your relationship with SONY under this EULA shall be determined by arbitration in San Jose, California, before three (3) impartial arbitrators, in accordance with the laws of the State of California, United States, without regard to its conflict of laws principles. If you reside in the United States, the arbitration shall be administered by JAMS and the arbitrators shall be selected pursuant to the rules and procedures of JAMS. If you reside outside the United States, the arbitration shall be administered by ICC and the arbitrators shall be selected pursuant to the rules and procedures of ICC. The parties shall bear their own costs and expenses, including attorneys' fees, but the arbitrators may, in the award, allocate all of the administrative costs of the arbitration, including the fees of the arbitrators, against the party who did not prevail. The decision of the arbitrators shall be final and nonappealable. Judgment on any award may be entered in

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Notwithstanding anything contained in this EULA to the contrary, you acknowledge and agree that any violation of or non-compliance with this EULA by you will cause irreparable harm to SONY, for which monetary damages would be inadequate, and you consent to SONY obtaining any injunctive or equitable relief that SONY deems necessary or appropriate in such circumstances. SONY may also take any legal and technical remedies to prevent violation of and/or to enforce this EULA, including, but not limited to, immediate termination of your use of the SOFTWARE, if SONY believes in its sole discretion that you are violating or intend to violate this EULA. These remedies are in addition to any other remedies SONY may have at law, in equity or under contract.

TERMINATION

Without prejudice to any of its other rights, SONY may terminate or suspend your access or use of the SOFTWARE and/or terminate this EULA if you fail to comply with any of its terms. In case of such termination, you must: (i) cease all use, and destroy any copies, of the SOFTWARE, and certify the same to SONY upon SONY's request; (ii) comply with the requirements in the section below entitled "Your Account Responsibilities". Any terms and conditions that by their nature or otherwise reasonably should survive expiration or termination of this EULA shall survive.

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UPDATES

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GOVERNING LAW AND JURISDICTION

The United Nations Convention on Contracts for the International Sale of Goods shall not apply to the SOFTWARE or this EULA. This EULA shall be governed by the laws of Japan, without regards to conflict of laws provisions. Any dispute arising out of this EULA shall be subject to the exclusive venue of the Tokyo District Court in Japan, and the parties hereby consent to the venue and jurisdiction of such courts.

EQUITABLE REMEDIES

Notwithstanding anything contained in this EULA to the contrary, you acknowledge and agree that any violation of or non-compliance with this EULA by you will cause irreparable harm to SONY, for which monetary damages would be inadequate, and you consent to SONY obtaining any injunctive or equitable relief that SONY deems necessary or appropriate in such circumstances. SONY may also take any legal and technical remedies to prevent violation of and/or to enforce this EULA, including, but not limited to, immediate termination of your use of the SOFTWARE, if SONY believes in its sole discretion that you are violating or intend to violate this EULA. These remedies are in addition to any other remedies SONY may have at law, in equity or under contract.

TERMINATION

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