QuantStudio[™] 3 and 5 Real-Time PCR Systems INSTALLATION, USE, AND MAINTENANCE

Firmware v1.4

Publication Number MAN0010407 Revision D.0





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Revision	Date	Description
D.0	22 July 2021	Updates – describes new features in Firmware v1.4.x and other changes, including:
		• Updated the name of the cloud-based platform to Thermo Fisher [™] Connect.
		• Added instructions for connecting to the Thermo Fisher [™] Connect cloud-based platform.
		Updated the firewall instrument-to-computer port number.
		Updated licensing information.
		Added Network and password security requirements section
		Added Perform internal validations before software upgrades section
		Added information for updating the firmware.
		 Update operating system to Windows[™] 10 (64-bit).
		 Added the MicroAmp[™] Optical Film Compression Pad as an optional accessory for the 96-well 0.2-mL block when using a MicroAmp[™] Optical 96-Well Reaction Plate, including instructions to load and unload the plate if using this accessory
		• Specified that an instrument profile is only for the instrument. It is not related to any other account for the system or the software.
		 Specified that if the connection between the instrument and the desktop softwa is interrupted during a run, the instrument will complete the run. The EDS file m be transferred manually to the desktop software with a USB drive or through a network drive.
		Changed all references to Bleach, 10% solution to Na-hypochlorite (0.1% v/v) solution
		Updated the calibration and verification schedule.
		 Specified that Background and Dye calibrations must be performed after ROI/Uniformity calibration.
		- Specified ROI/Uniformity calibration invalidates all other calibrations.
		 Added that the Background calibration can be used to obtain the most accurate data for the removal of background fluorescence.
		 Specified that a Background calibration and a Dye calibration do not invalidate all other calibrations.
		 Added a recommendation that RNase P instrument verification be performed after instrument calibrations.
		Added troubleshooting information for RNase P verification.
		 Added troubleshooting information if the connection between the instrument ar the computer is not recognized.
		 Added storage and transport conditions to the environmental requirements.
		 Specified that restoring factory defaults can only be performed by an administrator and specified that the most recent valid ROI/Uniformity, Background, and System Dye calibrations are not deleted when factory default are restored.
C.0	December 2015	Updates – describes new features in Firmware v1.2.x, including:
		Updated workflow to create an instrument profile
		 Streamlined workflow for connectivity to Thermo Fisher[™] Connect
		 Improved display of VeriFlex[™] Zones
		Plate insert reminder before starting run
		Support for 96-well Fast (0.1 mL) plates

(continued)

Revision	Date	Description
B.0	September 2015	Updates – describes new features in Firmware v1.1.x, including:
		• Experiment runs: monitor real-time data, edit cycle number and lock screen during a run, view end plot, and support for 384-well plates
		• File management: access Network folders, navigate folder structures and save templates on instrument, and perform batch actions for file management
		• Instrument configuration: support for Security, Audit, and e-Signature (SAE), smart monitoring, and choice of server region
		Includes information on software feature comparison, definitions of terms, parts of method, network connection options, experiment types, desktop software installation.
A.0	April 2015	New document. Describes installation, operation, and maintenance of the QuantStudio [™] 3 and 5 Real-Time PCR Systems.

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About this guide

Purpose

This guide provides information about installing, using, and maintaining the QuantStudio[™] 3 and 5 Real-Time PCR Systems.

Note: For information and instructions on performing experiments on these systems, see *QuantStudio*[™] *Design and Analysis Desktop Software User Guide* (Pub. No. MAN0010408).



Product information

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Network and password security requirements

Network configuration and security

The network configuration and security settings of your laboratory or facility (such as firewalls, antivirus software, network passwords) are the sole responsibility of your facility administrator, IT, and security personnel. This product does not provide any network or security configuration files, utilities, or instructions.

If external or network drives are connected to the software, it is the responsibility of your IT personnel to ensure that such drives are configured and secured correctly to prevent data corruption or loss. It is the responsibility of your facility administrator, IT, and security personnel to prevent the use of any unsecured ports (such as USB, Ethernet) and ensure that the system security is maintained.

Password security

Thermo Fisher Scientific strongly recommends that you maintain unique passwords for all accounts in use on this product. All passwords should be reset upon first sign in to the product. Change passwords according to your organization's password policy.

It is the sole responsibility of your IT personnel to develop and enforce secure use of passwords.

Perform internal validations before software upgrades

IMPORTANT! If you have a validated workflow, you must perform all internal validations as required by your organization's standard operating procedures before performing a software upgrade.

Update the instrument firmware

If your instrument is not on version 1.4 or later of the firmware you must update the firmware to access the features and updates described in Revision D.0 of the revision history. For more information, see "Revision history" on page 2. For information on updating your firmware, see https://www.thermofisher.com/us/en/home/global/forms/life-science/quantstudio-3-5-software.html.

Prior to updating your firmware, we recommend you perform a backup. For information on performing a backup, see "Backup the instrument" on page 11.

Backup the instrument

- 1. In the home screen, touch (a) Settings > Maintenance and Service > Backup/Restore.
- 2. Touch Backup Instrument.
- 3. Select a location to store the backup settings for your instrument.
- 4. If backing up to a USB drive, insert a USB drive into the front-panel USB port.
- 5. Enter a backup file name, then touch **Done**.
- 6. Select the elements to backup, or leave them all selected.
- 7. Touch Backup.

Instrument hardware description

Instrument overview

The QuantStudio[™] 3 and 5 Real-Time PCR Systems use fluorescence-based polymerase chain reaction (PCR) reagents to perform:

- Quantitative detection of target nucleic acid sequences (targets).
- Qualitative detection of targets (endpoint analysis, genotyping experiments, and presence/absence experiments).
- Qualitative analysis of the PCR product (post-PCR melt curve analysis).



The following fixed-block configurations are available:

QuantStudio [™] 3 Real-Time PCR System	QuantStudio [™] 5 Real-Time PCR System
 96-Well VeriFlex[™] 0.2-mL Block (4 Color) 96-Well VeriFlex[™] 0.1-mL Block (4 Color) 	 96-Well VeriFlex[™] 0.2-mL Block (6 Color De-coupled) 384-Well Block (5 Color) 96-Well VeriFlex[™] 0.1-mL Block (6 Color De-coupled)

The instrument can be run directly from the touchscreen to create and start experiments. To design experiments or to analyze data, the instrument can be integrated with the QuantStudio[™] Design and Analysis Software.

An optional barcode scanner and optional wireless adapter can be purchased separately.

Instrument filters and supported dyes

System dyes

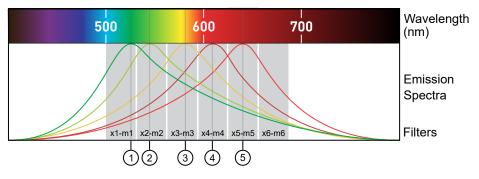
The instruments use a coupled four-color, coupled five-color, or de-coupled six-color filter set that supports the dyes shown in the following table and figure. For more information about available spectral dye calibration kits, contact Support.

Peak filter	Color	Filter wavelength (nm) ^[1]		Factory-calibrated	Evenue eveten duce
Peak Inter		Excitation	Emission	dyes	Example custom dyes
x1-m1	Blue	470 ± 15	520 ± 15	FAM [™] dye, SYBR [™] Green	SYT09
x2-m2	Green	520 ± 10	558 ± 12	VIC [™] dye	JOE [™] dye, HEX [™] dye, TET [™] dye ^[2]
x3-m3	Yellow	550 ± 10	587 ± 10	NED [™] dye, TAMRA [™] dye, ABY [™] dye	Cy [®] 3
x4-m4	Orange	580 ± 10	623 ± 14	ROX [™] dye, JUN [™] dye	Texas Red [™] dye
x5-m5	Red	640 ± 10	682 ± 14	MUSTANG PURPLE [™] dye, Cy [®] 5	LIZ [™] dye
x6-m6	Deep-Red	662 ± 10	711 ± 12	None ^[3]	Cy [®] 5.5

^[1] The central wavelengths are the optimized wavelengths.

[2] The HEX[™] and TET[™] dyes from Thermo Fisher Scientific fall within the emission wavelength range of the system, therefore they can be added and adapted for use on the instrument.

^[3] This filter set currently does not support any dyes supplied by Thermo Fisher Scientific.



- (1) x1-m1 − FAM[™] dye, SYBR[™] Green
- ② x2-m2 − $VIC^{^{\text{\tiny M}}}$ dye
- ③ x3-m3 − ABY[™] dye, NED[™] dye, Cy[®]3, TAMRA[™] dye
- (4) x4-m4 − JUN^{$^{\text{M}}$} dye, ROX^{$^{\text{M}}$} dye, Texas Red^{$^{\text{M}}$} dye
- (5) x5-m5 − Cy[®]5, MUSTANG PURPLE[™] dye

Custom dyes

The instrument can run assays designed with custom dyes.

Custom dyes include:

- Dyes that are not supplied by Thermo Fisher Scientific.
- Dyes or formulations of dyes that are not system dyes for the instrument.

To use a custom dye on the instrument, review the following requirements.

- Calibrate the instrument for the custom dye (see "Calibrate custom dyes" on page 59).
- Ensure that the custom dye excites between 455 672 nm and emits between 505 723 nm.
- Select a custom dye that does not overlap with other dyes used in the run (see the filterwavelength table in "System dyes" on page 12).
- The custom dye must be attached to the 5' end of a short DNA oligonucleotide consisting of the first two bases of the probe sequence without a quencher at the 3' end.

About data collection

The instrument collects raw fluorescence data at different points during the PCR cycle, depending on the type of run performed.

When you create an experiment template (EDT file) in the software, you can customize the optical filter channels through which the instrument collects data. You can specify a filter channel set for all PCR thermal protocols and, optionally, a different filter set for the melt curve stages.

Run type	Experiment type	Data collection point
Real-time	 Standard curve Relative standard curve Comparative C_T (ΔΔC_T) Melt curve 	During the thermal cycling protocol. Typical timing is to collect data at each cycle of a PCR stage or continuously during a melt stage.
Post-PCR (endpoint)	GenotypingPresence/ Absence	 After thermal cycling is completed. For Presence/Absence and Genotyping experiments, data collection before the PCR cycle is optional, but recommended. (Optional) Before thermal cycling starts. Collecting data during the run can confirm genotyping results by viewing traces in allelic discrimination plots or viewing genotyping calls at earlier cycles.

Blocks with VeriFlex[™] Zones

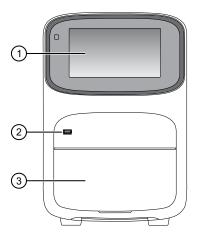
Applied Biosystems[™] VeriFlex[™] technology provides independent temperature zones that offer enhanced functionality and precise control over your real-time PCR runs.

QuantStudio [™] 3 System	QuantStudio [™] 5 System
Three programmable VeriFlex [™] Zones.	Six programmable VeriFlex [™] Zones. ^[1]
$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$

^[1] Only applicable for 96-well 0.2-mL and 0.1-mL blocks.

These independent zones are ideal for real-time PCR optimization or the ability to run multiple experiments in the same run. Unlike standard gradients which give a sigmoidal temperature curve across the columns, blocks with VeriFlex[™] Zones help deliver accurate temperatures across every zone.

Parts of the instrument



1 Touchscreen – Controls the instrument.

(2) USB port – For connection to an external network drive or external data storage device.

③ Instrument drawer – Contains sample plate.

The instrument includes three additional USB ports on the back of the instrument.

Note: The instrument recognizes only one external storage device at a time for data transfer.

Software description

Instrument, desktop, and cloud software features

The instrument software and the QuantStudio[™] Design and Analysis Software (desktop and Thermo Fisher[™] Connect) include the features described below.

Feature	Instrument	Desktop	Thermo Fisher [™] Connect
Use as guest (no sign in)	1	✓ [1]	—
Create templates (unlocked or locked)	_	√	1
Edit unlocked templates	✓	 Image: A set of the set of the	1
Edit locked templates (password assigned by template creator required)	_	1	1
Load system or user-created templates (.edt file) to instrument	1	1	_



(continued)

Feature	Instrument	Desktop	Thermo Fisher [™] Connect
Change experiment settings in template (.edt file) loaded on the instrument	1	_	_
Settings that can be changed in a locked template (no password required):			
Properties: All settings			
Method: No changes allowed			
Plate: Sample names			
Settings that can be changed in an unlocked template:			
Properties: All settings			
Method: All settings			
Plate: Sample names			
Load plate in instrument	1	_	_
Start run	1	1	_
View real-time data during a run	1	1	1
View instrument status (running, calibration needed, and so on)	1	_	1
Analyze results	_	1	1
Set calibration reminders	1	_	✓
Review exported calibration or RNase P verification results	_	1	✓ (anh: DNasa D)
			(only RNase P)

[1] Login required if the Security, Audit, and e-Signature (SAE) module is enabled (QuantStudio[™] 5 System only)

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Term	Definition	Supported in
Folders in Load Experiment My Instrument Public USB Post Read	 Location in which you can store templates (EDT files) on the instrument: My Instrument – Displayed if you are signed in Public – Location in which all experiments run by guest users are stored USB – USB for manual transfer to and from a computer Post Read – If you run an endpoint experiment, the preread experiment is automatically saved in this folder for post-read analysis 	Instrument
Experiment Template File (EDT)	 Default settings for an instrument run, can be modified before instrument run. Two types: Factory-provided, accessed from Open Template (instrument) or Create Experiment (desktop or Thermo Fisher[™] Connect) User-created, accessed from Load Experiment 	Instrument Desktop Thermo Fisher [™] Connect
Experiment Run File (EDS)	Settings and data for a completed instrument run.	Instrument Desktop Thermo Fisher [™] Connect

Folders, templates, experiments, and projects

Template and experiment components	Instrument	Desktop	Thermo Fisher [™] Connect
Properties	 Experiment file name Reagent information (Reagent barcode, Lot#) Plate barcode Data destination (location for auto-transfer of data) Comments (Tags are not used at this time) 	 Experiment file name Plate barcode User name Instrument type Block type Experiment type Chemistry (reagent inform Run mode Comments (Thermo Fisher[™] Connect 	
Method	Thermal cycling conditions	Thermal cycling conditions	



(continued)

Template and experiment components	Instrument	Desktop	Thermo Fisher [™] Connect
Plate	Sample names You cannot edit targets/SNP assays or tasks on the instrument.	Define and assign samples, ta tasks in the Quick Setup and a Plate tab.	rgets or SNP assays, and Advanced Setup panes of the
Run	Start and monitor a run in progress View: time remaining and temperature, method, plots Pause, resume, stop a run	Start and monitor a run in progress View: time remaining and temperature, method, plots	Monitor a run in progress (link to Thermo Fisher [™] Connect Instrument Details) View: time remaining and temperature, method, plots
Results	Not applicable	Review plots	
Export	Not applicable	Export results	

Third-party software

Before installing third-party software on the computer running QuantStudio[™] Design and Analysis Software, confirm that the third-party software will not do the following:

- Restrict Ethernet communication.
- Interfere with instrument or computer operation.

Network connection options

You can connect an instrument to a network or computer in the configurations listed below. For specific information on networking, see "Networking" on page 95.

Direct connection	Local area network (LAN) connection	Thermo Fisher [™] Connect connection
wired	wired or wireless	wired or wireless



Types of runs

Purpose	Description
Standard curve run	
Determines absolute target quantity in samples.	 The software measures amplification of the target in a standard dilution series and in test samples. The software generates a standard curve using data from the standard dilution series. The software uses the standard curve to interpolate the absolute quantity of target in the test samples.
Relative standard cu	ve run
Determines relative target quantity in samples.	 The software measures amplification of the target of interest and of an endogenous control target in a standard dilution series, in a reference (calibrator) sample, and in test samples. The endogenous control is a target that is expressed equally in all samples; examples of endogenous controls are β-actin, GAPDH, and 18S ribosomal RNA. The software can algorithmically incorporate multiple endogenous control targets in relative quantification calculations. The reference sample is used as the basis for relative quantification results (or 1× sample). For example, in a study of drug effects on gene expression, an untreated control is an appropriate reference sample. The software generates standard curves for the target of interest and the endogenous control using data from the corresponding standard dilution series. The software uses the standard curves to interpolate the quantities of the target of interest and the endogenous control in each sample. The target quantity in each sample is then normalized to the sample's endogenous control quantity.
	4. To determine the relative quantity of the target in test samples, the software divides the normalized target quantity in the sample by the normalized target quantity in the reference sample.
Comparative C_t ($\Delta\Delta C$	t) run
Determines relative target quantity in samples.	 The software measures amplification of the target of interest and of an endogenous control target in a reference (calibrator) sample and in test samples. The endogenous control is a target that is expressed equally in all samples; examples of endogenous controls are β-actin, GAPDH, and 18S ribosomal RNA. The software can algorithmically incorporate multiple endogenous control targets in relative quantification calculations. The reference sample is used as the basis for relative quantification results (or 1× sample). For example, in a study of drug effects on gene expression, an untreated control is an appropriate reference sample. The measurements for the target of interest are normalized to the endogenous control. To determine the relative quantity of the target in test samples, the software compares the normalized ΔC_q (ΔC_t or ΔC_{rt}) for the sample to the normalized ΔC_q (ΔC_t or ΔC_{rt}) for the reference sample.



Purpose	Description				
Genotyping run	Genotyping run				
Detects single nucleotide polymorphism (SNP) variants of a target nucleic acid sequence.	 Genotyping runs use preformulated TaqMan[™] SNP Genotyping Assays that include the following components: Two sequence-specific primers for amplification of sequences containing the SNP of interest Two allele-specific TaqMan[™] probes for Allele 1 and Allele 2 The software normalizes the fluorescence of the reporter dyes to the fluorescence of the passive reference dye in each well. The software plots the normalized reporter dye signal of each sample well on an Allelic Discrimination Plot, which contrasts the reporter dye intensities of the allele-specific probes. The software algorithmically clusters the sample data, and assigns a genotype call to the samples of each cluster according to its position on the plot. 				
Presence/absence ru	n				
Determines the presence or absence of a target nucleic acid sequence in a sample.	The software calls the target present or absent based on an algorithmically determined call threshold. (The call threshold is different from the C_t threshold; the C_t threshold is not used to make calls.)				
Melt curve run					
Determines the melting temperature (T_m) of the amplification products of a PCR that used intercalating dyes.	 In the software, melt curve analysis is included in the default run method for any run type that uses intercalating dyes. 1. The software plots a melt curve based on the fluorescence of the dye with respect to change in temperature. 2. Using the melt curve, the software calculates the melting temperature (T_m). 				



Start, sign on, and configure the instrument

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Installation and instrument verification

Before the first use of the instrument, complete the following tasks:

- Install the instrument (see "Unpack and install the instrument" on page 91).
- Verify instrument performance (see "Perform instrument verification using RNase P plates" on page 53).

Note:

- Instruments are factory calibrated, so no calibration is necessary at installation. However, we recommend that you verify instrument performance before using the instrument.
- Regular calibration and verification should be performed according to the calibration and verification schedule (see "Calibration and verification schedule" on page 43).

Precautions for use



CAUTION! PHYSICAL INJURY HAZARD. Do not remove the instrument cover. There are no components inside the instrument that you can safely service yourself. If you suspect a problem, contact technical support.



CAUTION! FIRE HAZARD. For continued protection against the risk of fire, replace fuses only with listed and certified fuses of the same type and rating as those currently in the instrument.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the sample block temperature can reach 100°C. Allow it to cool to room temperature before handling.



CAUTION! Before using a cleaning or decontamination method other than those recommended by Thermo Fisher Scientific, confirm with Thermo Fisher Scientific that the proposed method will not damage the instrument.

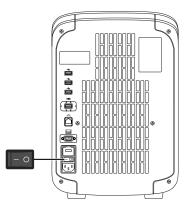


CAUTION! Use flat caps for tubes. Rounded caps can damage the heated cover.

Power on the instrument

- 1. Touch anywhere on the touchscreen to determine if the instrument is in sleep mode. If the home screen is displayed, the instrument is already powered on.
- 2. If the home screen does not display, power on the instrument by pressing the switch on the rear panel.

If left unattended, the instrument automatically enters sleep mode (enabled by default) to conserve power.



2

SSFLCCN8M12 (?)(▲) 2) 1 3 $\left(4\right)$ Set up run Guest (5) QuantStudio® 5 - 96-Well 0.2-mL Block ۲ (9) $\left(6\right)$ Sign In Load Experiment **Open Template** Settings **Run Last** 08/04/2015 03:38 AM (8) (7)(1) Avatar and Instrument name 2 Eject icon 3 Help icon (4) Status dial (5) Current user name; instrument block type 6 Settings button 7 Buttons for accessing .edt files (8) Connectivity icons

Parts of the home screen

(9) Sign In (or My Profile) button

Table 1 Parts of the home screen

Element of the home screen	Function	For more information, see
Avatar and Instrument name	Set by the administrator to uniquely identify instrument.	"Manage the instrument name (Administrator only)" on page 88.
Eject icon	Touch to open or close the instrument drawer.	_
Help icon	Touch to launch the touchscreen Help system to access step-by-step instructions.	_



Element of the home screen	Function	For more information, see
Status dial	 When the instrument is in use – Displays the sample block temperature, the elapsed run time, and the run status. You can swipe the dial to the left or touch > to access real-time views of the run. When the instrument is not in use – Displays Set up run. You can start a run by touching the status dial. When the instrument is locked – Displays a lock icon () within the status dial. 	_
Current user name and block type	Displays the user name of the current signed-in user and the instrument block type. Note: If no user is signed-in, the instrument defaults to the Guest profile.	_
Settings button	Touch (*) Settings to configure, calibrate, or learn about the instrument.	"Configure instrument settings" on page 26.
Buttons for accessing experiment and	Load Experiment – Touch to open a user-created .edt file from a Thermo Fisher [™] Connect account, USB, instrument folder, or network drive.	"Run an experiment from a saved file" on page 30.
template files	Open Template – Touch to open a system .edt template file.	"Create and run an experiment from a template" on page 30.
	 Run Last – Touch to open the last .edt file run on the instrument. If you are signed-in, the file opened will be the last file you ran when signed-in. If you are not signed-in, the file opened will be the last file the guest profile ran. 	"Repeat the last instrument run" on page 31.
Connectivity icons	 ^B – The instrument is connected to a wired network. 	_
Sign In button (My Profile button when a user is signed in)	 Touch ③ Sign In to sign into an instrument profile and link to a Thermo Fisher[™] Connect account. Touch ④ My Profile to change instrument profile settings, link to a Thermo Fisher[™] Connect account, or lock the instrument during a run. 	"Sign in" on page 25.

 Table 1
 Parts of the home screen (continued)

Use the instrument without signing in

If the instrument is configured by an Administrator to allow guest access (❀ Settings ► Manage Users ► Sign In Required set to off), you can use the instrument without signing in.

The following limits are in place if you do not sign in to the instrument:

- All actions are recorded to Guest user profile.
- You have access only to the **Public** folder for selecting and storing experiments.
- You cannot transfer data to Thermo Fisher[™] Connect (only to USB or network drive).

Create a new instrument profile

- 1. In the home screen, touch (a) Sign In, then touch Get Started.
- 2. Touch Name, enter a username, then touch Done.
- 3. Touch PIN Code, enter a four-digit numerical password, then touch Enter.

Note: Touch the Show PIN checkbox to switch PIN display on or off.

- 4. Touch Confirm PIN, then repeat step 2.
- 5. Touch Create profile.
- 6. Sign in to the profile you just created. See "Sign in" on page 25.

Note: To enable access to Thermo Fisher[™] Connect, see "Link an instrument profile to Thermo Fisher[™] Connect" on page 77.

Sign in

Create an instrument profile before signing into the instrument. See "Create a new instrument profile" on page 25.

Note: An instrument profile is a user account specifically for the instrument. It is not related to any other user account for the system or software.

- 1. In the home screen, touch (2) Sign In.
- 2. Touch Sign In, then select your username.
- 3. Enter your PIN, then touch Enter.

Note: To enable access to Thermo Fisher[™] Connect, see "Link an instrument profile to Thermo Fisher[™] Connect" on page 77.

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Sign out

- 1. In the home screen, touch (2) My Profile.
- 2. Touch Sign Out.

Configure instrument settings

Touch (a) **Settings** in the home screen to configure settings as needed.



Touch (2) for step-by-step instructions on configuring settings.

Options	Description	
Instrument Settings	Instrument Settings	
Instrument Name (Administrator only)	Instrument name. If connected to a network, the instrument name must be unique. <i>(Optional)</i> Add an avatar image for the instrument (.jpg, .png, or .gif) from a USB.	
Sleep Mode	Enable the instrument to enter a standby mode after a set length of inactivity.	
Heated Cover Temperature	Set the temperature of the heated cover for instrument operation and standby mode. Enable heated cover to automatically turn off during standby mode.	
Network Drive	Specify a default network location for the signed-in user.	
Insert Plate Reminder	Enable a reminder to insert a plate before starting a run from the instrument.	
OEM Connection Only (Administrator only)	Required for API access to the instrument. When enabled, the desktop cannot connect to the instrument. API access to the instrument is exclusive to authorized OEM partners.	
Cloud Region	Specify the regional server location to access Thermo Fisher [™] Connect.	
(Administrator only)	Note: Once you set a region for the cloud platform, restore factory defaults to change the region.	



Options	Description	
Date/Time	Set time zone and date and time formats.	
Network Connection	Set wired or wireless network connection for the instrument.	
Restore Factory Defaults	Restore the instrument to the factory settings.	
	IMPORTANT! Back up the instrument before restoring factory defaults (see "Backup or restore the instrument" on page 67).	
	 Note: When you restore an instrument to its factory defaults: User profiles and files stored on the instrument are deleted, including all user- created .edt and .eds files and any custom dye and custom melt calibrations. System templates and factory calibrations remain on the instrument. 	
SAE Mode	When enabled the following items apply:	
(Administrator only)	The home screen displays SAE mode.	
(QuantStudio [™] 5 Instrument only)	 Runs must be started from the desktop software. The instrument locks out access to load experiments, open templates, repeat the last run, start a run, change the instrument name, and change the date/time. 	
	Only an administrator can perform calibrations.	
	• A user can monitor a run and view the method from the instrument touchscreen.	
About Instrument	About Instrument	
About Instrument	Displays the instrument Model Name, Block Serial Number, and Firmware Version.	
License Agreement	Displays the End User Software License Agreement and the Limited Product Warranty. You can export the License Agreement to a USB drive.	
Notifications		
_	Enable home screen notifications of instrument errors and software updates. This function is not related to the Notifications function in the cloud software.	
	The number of new, unviewed notifications displays over Settings in the home screen.	
Maintenance and Service		
Software Update	Update the instrument software.	
(Administrator only)		
Monitoring	Enable:	
(Administrator only)	 Remote Monitoring Service to automatically send critical device statistics to Thermo Fisher Scientific (does not monitor or send data). 	
	 Thermo Fisher Scientific Cloud Monitor to allow real-time monitoring of instrument runs from a Cloud account. 	
Instrument Statistics	Displays instrument usage information: Block Cycle Count and LED Life.	



Options	Description	
Calibrations	 Perform calibrations ROI and Uniformity Dye Custom (including Background calibration) View calibration history and set calibration reminders in History and Reminders 	
RNase P Verification	Perform instrument verification using an RNase P plate.	
Self Verification Test	Check the instrument hardware functions.	
Log	View and export Instrument Log.	
Backup / Restore	 Backup Instrument Restore a Backup (Administrator only) 	
Ship Prep Mode	Prep Mode Place the instrument in a safe state for shipping, moving, or long-term storage.	
Run History		
_	 Displays the experiments run on the instrument and whether the data was transferred. Note: Experiments run while a user is signed-in can only be viewed or transferred by that user or an administrator. Touch an experiment to view experiment run details and to transfer or delete .eds file. 	
Manage Users		
Sign In Required (Administrator only)	Enable the restriction of instrument use to only signed-in users (disables Guest profile use).	
Sign Out Timer (Administrator only)	Set the duration of inactivity before a user is automatically signed out.	
Manage Profiles (Administrator only)	 Instrument Cloud All Profiles Displays the profile information for the instrument and the associated Thermo Fisher[™] Connect cloud-based account. 	



Create and run experiments on the instrument

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Workflow

Start the instrument

▼

(Optional) Sign in to your instrument profile *(Optional)* Link to your Thermo Fisher[™] Connect account

▼

Load an existing experiment template (EDT file) or

Create an experiment template (EDT file)

▼

Modify experiment settings as needed

▼

Load the plate in the instrument

▼

Start the run from the instrument or the desktop software

▼

(Optional) Monitor the run from Thermo Fisher[™] Connect

▼

Transfer results (EDS file) to the desktop software or Thermo Fisher[™] Connect, then analyze



Options for running an experiment

Create and run an experiment from a template

In the home screen:

- 1. Touch 🕹 Open Template.
- 2. (Optional) Touch a category in the left column.
- 3. Touch the file name.
- 4. (*Optional*) Enter or edit template properties, including **Run File Name** (EDS file name), **Plate Barcode**, **Reagent Information**, and **Data Destination** (see "Enter or edit template properties").
- 5. (Optional) Edit the run method (see "Edit the run method" on page 33).
 - Add, remove, or edit a step, stage, melt curve, or data collection point.
 - Adjust the heated cover temperature, sample volume, or number of cycles.
 - Configure VeriFlex[™] Zones, ramp rate, and pause settings.
- 6. (*Optional*) Define plate wells with sample names and view **Well ID**, **Targets**, or **Dyes** (see "Define, assign, and view well details").
- 7. Load a plate into the instrument (see "Load and unload the plate" on page 37).
- 8. Touch Start Run.

When prompted, confirm that you inserted a plate.

Note: To disable this reminder, select **Do not show again** or select (a) **Settings > Insert Plate Reminder** in the home screen.

Run an experiment from a saved file

In the home screen:

- 1. Touch 🚵 Load Experiment.
- 2. Touch the appropriate icon to navigate to your file location.
 - For files saved to the guest profile, touch **Wy Instrument > Public**.
 - For pre and post read files, touch **Wy Instrument > Post Read**.
- 3. Touch the file name.
- 4. (Optional) "Manage templates (EDT files)" in either 📼 USB or 📮 My Instrument.
- (Optional) Enter or edit template properties, including Run File Name (EDS file name), Plate Barcode, Reagent Information, and Data Destination (see "Enter or edit template properties").

- 6. (Optional) Edit the run method (see "Edit the run method" on page 33).
 - Add, remove, or edit a step, stage, melt curve, or data collection point.
 - Adjust the heated cover temperature, sample volume, or number of cycles.
 - Configure VeriFlex[™] Zones, ramp rate, and pause settings.
- 7. (*Optional*) Define plate wells with sample names and view **Well ID**, **Targets**, or **Dyes** (see "Define, assign, and view well details").
- 8. Load a plate into the instrument (see "Load and unload the plate" on page 37).
- 9. Touch Start Run.

When prompted, confirm that you inserted a plate.

Note: To disable this reminder, select **Do not show again** or select **③ Settings > Insert Plate Reminder** in the home screen.

Repeat the last instrument run

This feature applies only to runs started from the instrument and is not available for runs started from the desktop software. If you are signed-in, this feature applies to the last run from your instrument profile.

In the home screen:

- 1. Touch 🝰 Run Last.
- (Optional) Enter or edit template properties, including Run File Name (EDS file name), Plate Barcode, Reagent Information, and Data Destination (see "Enter or edit template properties").
- 3. (Optional) Edit the run method (see "Edit the run method" on page 33).
 - Add, remove, or edit a step, stage, melt curve, or data collection point.
 - Adjust the heated cover temperature, sample volume, or number of cycles.
 - Configure VeriFlex[™] Zones, ramp rate, and pause settings.
- 4. (*Optional*) Define plate wells with sample names and view **Well ID**, **Targets**, or **Dyes** (see "Define, assign, and view well details").
- 5. Load a plate into the instrument (see "Load and unload the plate" on page 37).
- 6. Touch Start Run.

When prompted, confirm that you inserted a plate.

Note: To disable this reminder, select **Do not show again** or select (a) **Settings > Insert Plate Reminder** in the home screen.

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Edit an experiment before starting a run

Enter or edit template properties

Access a template (EDT file). For more information, see the following sections:

- "Create and run an experiment from a template" on page 30.
- "Run an experiment from a saved file" on page 30.
- "Repeat the last instrument run" on page 31.

In the Properties screen, touch Edit to enter or edit template properties.

- Edit the file name for the EDS file for the run.
 - a. Touch the Run File Name field.
 - b. Enter the name for the EDS file, then touch Done.
- Enter a plate barcode.
 - a. Touch the Plate Barcode field.
 - b. Enter or scan the plate barcode, then touch **Done**.
- Record reagents and their expiration dates.
 - a. Touch **Reagent Information**.
 - b. Touch Add, or touch an existing reagent, then touch Edit or Delete.
 - c. Touch the Name, Type, Lot #, Reagent Barcode, Part #, or Expiration Date field to enter individual reagent information.
 - d. Touch Done.
- Automatically transfer run data (EDS file) when an instrument run ends.
 - a. Touch for Data Destination.
 - b. Touch the appropriate icon to select a data destination.
 - c. Under the desired data destination, select Automatically transfer experiment.
 - d. Touch Done.
- Enter a comment.
 - a. Touch Comments.
 - **b.** Enter text, then touch **Done**.

Scan a barcode using the optional barcode scanner

The instrument is compatible with an optional Handheld Barcode Scanner (Cat. No. 4488442, purchased separately). The barcode scanner reads Code 128 (alphanumeric), which supports 128 ASCII character barcodes.

- 1. Click the Barcode field.
- 2. Hold the scanner 20–30 cm away from a plate or container label and aim at the center of the barcode, then press the trigger.
- 3. Slowly move the scanning beam across the barcode until the scanner emits a high-pitched tone.

When the scanner scans a barcode, it automatically transmits the following information:

- Transmits the alphanumeric equivalent of the barcode to the barcode field.
- Transmits other reagent information (Lot #, Part #, Expiration Date, etc.)

For more information about the hand-held barcode scanner, see the user documentation provided with the barcode scanner.

Edit the run method

Access a template (EDT file):

- "Create and run an experiment from a template" on page 30.
- "Run an experiment from a saved file" on page 30.
- "Repeat the last instrument run" on page 31.

For an overview of the method as it is graphically represented on the touchscreen, see "Method elements" on page 34.

In the Method screen:

- 1. Touch Edit.
- 2. Touch a field, enter changes, then touch Enter.

Note: Touch-drag \equiv to quickly increase or decrease a step temperature.

3. Touch Manage Steps to add or remove a step, stage, melt curve, or data collection point.

Note: The Manage Steps screen also provides access to configuring VeriFlex[™] Zones, Ramp Rates, and Add Pause settings (see "Configure VeriFlex[™] Zones, ramp rates, and pause settings" on page 35).

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Method elements

	7 8 9
 6 1 Method tab 2 Heated cover temperature 	(6) Number of cycles for the sta(7) Temperature for the step

- ③ Reaction volume
- (4) Stage of thermal protocol
- 5 Step within a stage

- ige
- 8 Hold time for the step
- (9) Data collection point

Manage steps, stages, melt curves, and data collection points

Add or remove a step, stage, melt curve, or data collection point using the Manage Steps option. You can also configure ramp rates and pause settings from the Manage Steps screen (see "Configure VeriFlex[™] Zones, ramp rates, and pause settings" on page 35).

• In the Method screen, touch Edit > Manage Steps.

Option	Procedure
	1. Touch Add/Remove steps > Add steps.
Add a step	 Touch + on the left or right border of a step to add a step before or after the step, respectively.
	3. Enter parameters for the new step, then touch Enter.
	4. Touch Done.
	1. Touch Add/Remove steps ▶ Remove steps.
Remove a step	2. Touch — on the step to be removed, then touch Done.



1	continued)
(continueuj

Option	Procedure	
	1. Touch Add/Remove stages > Add stages.	
	 Touch + on the left or right border of a stage to add a stage before or after the stage, respectively. 	
Add a stage	3. Edit parameters of the new stage in the Method screen. See "Edit the run method" on page 33.	
	4. Touch Done.	
D	1. Touch Add/Remove stages > Remove stages.	
Remove a stage	2. Touch — on the stage to be removed, then touch Done .	
	1. Touch Melt curves ▶ Add melt curve.	
	 Touch + on the left or right border of a step to add a melt curve before or after the step, respectively. 	
	3. Select Continuous or Step and hold.	
Add a melt curve	4. Touch melt curve parameters to edit, then touch Done.	
	5. Touch Done.	
	Note: Depending on the experiment type, there can be restrictions on the addition or placement of melt curves.	
	1. Touch Remove melt curve.	
Remove a melt curve	2. Touch — on the melt curve to be removed, then touch Done.	
Add or remove data	1. Touch Data collection location.	
collection points	2. Touch in to switch data collection on or off.	

Configure VeriFlex[™] Zones, ramp rates, and pause settings

In the Method screen, touch Edit > Manage Steps > Advanced Options.

- To use VeriFlex[™] Zones (96-well blocks only):
 - a. Touch VeriFlex[™] Zones.
 - **b.** Touch ✓ on the step to apply VeriFlex[™] Zones.
 - c. Touch each zone to edit the temperature, then touch Enter. The background colors of the VeriFlex[™] Zones change with the temperature edit. Zones with higher temperatures display a background brighter than those with lower temperatures.
 - d. (Optional) Apply additional VeriFlex[™] Zones.
 - e. Touch Done.
- To edit Ramp Rates:
 - a. Touch Ramp Rates.
 - b. Touch the ramp rates fields.



- c. Touch Enter.
- d. (Optional) Edit additional steps, then touch Done.
- To add a pause into the method:
 - a. Touch Add Pause.
 - **b.** Touch **||** in a stage.
 - c. Enter the pause temperature, and the cycle after which the pause will occur.
 - d. Touch Enter.

The pause is represented by a **P** in the corner of the stage.

e. Touch Done.

Define, assign, and view well details

- 1. Access a template (EDT file). For more information, see the following sections.
 - "Create and run an experiment from a template" on page 30.
 - "Run an experiment from a saved file" on page 30.
 - "Repeat the last instrument run" on page 31.
- 2. In the **Plate** screen, define, assign, and view well details.

Option	Procedure
Define and assign samples in the plate layout view	 Touch IIII. Touch Manage or touch an individual well. Touch the Samples subtab. Select one or more wells, then touch Edit. Enter sample names for the selected wells, then touch Done.
Define and assign samples in the well table view	 Touch ≡. Touch Edit to edit sample names for individual wells. Touch a sample name field, enter a new name, then touch Done.
View target information	 Touch IIII. Touch Manage or touch an individual well. Touch the Targets subtab. Touch Details to view target information, then touch Done.

3. Touch Done to return to the Plate tab.

Load and unload the plate

If a compression pad is not used, see "Load and unload a plate in the instrument" on page 37.

A compression pad is needed for some applications with a 96-well 0.2-mL MicroAmp plate.

See your product documentation to confirm if the use of a compression pad is required.

To load and unload the plate, see "Load and unload a plate in the instrument with a compression pad" on page 38.

Load and unload a plate in the instrument



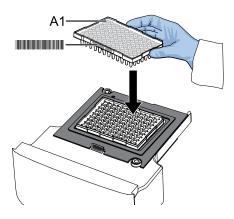
CAUTION! Use flat caps for 0.2-mL tubes and 0.1-mL tubes. Rounded caps can damage the heated cover.

1. Load the plate.

a. Touch a to eject the instrument drawer.

b. Load the plate onto the plate adaptor so that the following criteria are met.

- Position well A1 of the plate in the top-left corner of the plate adapter.
- Ensure that the barcode faces the front of the instrument.



IMPORTANT! The instrument should be used by trained operators who have been warned of the moving parts hazard.

Note: (*For 96-well 0.2-mL blocks only*) Do not remove the black plate adapter before loading a plate or tube strips. If used, tube strips can fit loosely in the adapter, but the heated cover will apply the appropriate pressure to seat the tube strips securely in the adapter.

Note: The 384-well and 96-well Fast (0.1-mL) block configurations do not require a plate adapter.

c. Touch (a) to close the instrument drawer.



Chapter 3 Create and run experiments on the instrument Load and unload the plate

- 2. When the run ends, unload the plate.
 - a. Touch (a) to eject the instrument drawer.
 - b. Remove the plate.
 - c. Touch a to close the instrument drawer.



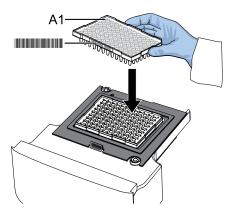
CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. Allow it to cool to room temperature before handling.

Note: If the instrument does not eject the plate, contact Support.

Load and unload a plate in the instrument with a compression pad

Some applications use a compression pad for a 96-well 0.2-mL MicroAmp plate.

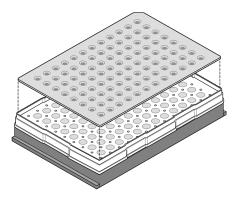
- 1. Load the plate.
 - a. Touch (a) to eject the instrument drawer.
 - b. Load the plate onto the plate adaptor so that the following criteria are met.
 - Position well A1 of the plate in the top-left corner of the plate adapter.
 - Ensure that the barcode faces the front of the instrument.



IMPORTANT! The instrument should be used by trained operators who have been warned of the moving parts hazard.

Note: (*For 96-well 0.2-mL blocks only*) Do not remove the black plate adapter before loading a plate or tube strips. If used, tube strips can fit loosely in the adapter, but the heated cover will apply the appropriate pressure to seat the tube strips securely in the adapter.

c. Before loading the plate in the drawer, place the MicroAmp[™] Optical Film Compression Pad with the gray side down on the surface of the plate to ensure a proper seal between the instrument and the adhesive film.



IMPORTANT! Be careful to place the compression pad with the brown side up and the gray side down, centered on top of the plate. Ensure that the compression pad is free from wrinkles and signs of deterioration prior to use.

- d. Touch a to close the instrument drawer.
- 2. When the run ends, unload the plate.
 - a. Touch (a) to eject the instrument drawer.
 - b. Remove the plate.
 - c. Touch (a) to close the instrument drawer.
 - d. Remove the MicroAmp[™] Optical Film Compression Pad from the plate and store it inside the pack.

IMPORTANT!

- If the compression pad becomes stuck inside the instrument, call service to clean the heated cover.
- Between each use, place the pad back in the pouch so that it does not dry out.
- Each compression pad may be used up to 20 times before discarding. Do not use more than 20 times.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. Allow it to cool to room temperature before handling.

Note: If the instrument does not eject the plate, contact Support.



View, pause, or stop a run

Do any of the following actions during an instrument run.

- View real-time data and plots (see "View real-time data and plots on the instrument touchscreen" on page 40).
- Adjust the display of real-time plots (see "Adjust the display of real-time plots on the instrument touchscreen" on page 41).
 - Touch > or swipe left once to view real-time run method information or to edit the number of cycles.
 - Touch > or swipe left twice to view real-time data and plots.
- Pause or stop an instrument run (see "Pause or stop an instrument run" on page 40).

View real-time data and plots on the instrument touchscreen

- 1. In the instrument home screen, during an instrument run, touch > or swipe left twice.
- 2. Touch Well details.
- 3. Touch Samples, Targets, or Tasks to select a graphical representation of each selection.
- 4. Touch Close to return to the home screen.

Pause or stop an instrument run

In the home screen, during an instrument run:

- 1. Touch > or swipe left once to view real-time run method information.
- 2. Stop or pause the run.
 - Touch Stop Run.
 - Touch **Pause**, then enter a pause temperature.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. If you want to access the plate during a run pause, enter room temperature as the pause temperature and allow the plate to cool to room temperature before handling.

3. (Optional) After pausing a run, touch Edit to change the number of cycles.

Adjust the display of real-time plots on the instrument touchscreen

- 1. In the instrument home screen, during an instrument run, touch > or swipe left twice to view real-time data and plots.
- 2. Touch Zoom.
- **3.** Touch \oplus or \oplus to zoom in or out.
- 4. Touch the arrows to pan left, right, up, or down on the graph.
- 5. Touch Close to return to the default view.

Lock the touchscreen during a run

After you have started a run, you can lock the touchscreen so that other users cannot interfere with instrument operation.

Note: You must be signed-in to use this feature.

- 1. Lock the touchscreen.
 - a. In the home screen, touch (a) My Profile.
 - b. Touch Lock Screen, then touch Lock.
- 2. Unlock the touchscreen.
 - a. Touch anywhere on the touchscreen.
 - b. Touch the **PIN Code** field, then enter your PIN.

Note: The touchscreen automatically unlocks when the run is complete.

Transfer, view, or manage files and results

Transfer EDS files from the instrument home screen

- 1. In the home screen, when a run ends, touch **Transfer File**.
- 2. Select the data destination for the EDS file.
- 3. Navigate to and select a folder.
- 4. Touch OK.
- 5. Touch Transfer.

Note: Touch (a) Settings > Run History to transfer EDS files at any time.



View run history and delete or transfer files from the instrument

In the home screen, touch (Settings > Run History.

- Touch an individual run record to view its details, then complete one of the following actions:
 - Touch **Delete** to delete the run record.
 - Touch **Transfer** to export the run data.
- Touch Manage to select multiple run records for simultaneous viewing, deletion, or transfer.

Note:

- Guests (users not signed-in) can only view guest run records.
- Users signed into their instrument profiles can also view their own run records.
- · Administrators can view all run records.

Note: If the connection between the instrument and the desktop software is interrupted during the run, the instrument still completes the run. However, the run data (EDS file) must be transferred from the instrument to the desktop software using a USB drive or a network drive.

Manage templates (EDT files)

This feature applies to templates (EDT files) on a **USB** or in **My Instrument**. To manage run results files (EDS files), see "Transfer EDS files from the instrument home screen" on page 41.

- 1. In the home screen, touch 📾 Load Experiment.
- 2. Touch 🖙 USB or 📮 My Instrument.
- 3. Touch Manage Files.
- 4. (*Optional*) Navigate through the **My Instrument**, **Public**, **USB**, and **Post Read** folders to access an EDT file. Folder availability depends on your sign-in status.
- 5. Touch one or more files to select for managing.
- 6. Delete or copy the files.
 - Touch **Delete Files**, then confirm deletion.
 - Touch Copy Files, select a file destination, then touch Paste Files.
- 7. Touch Done.



Calibrate and verify instrument performance

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Calibration and verification schedule

The instrument is factory-calibrated and does not require calibration at installation. To ensure optimal performance, perform calibrations at the recommended frequency.

Note: After instrument installation, we recommend performing instrument verification using the provided RNase P plate.

IMPORTANT! Perform calibrations and instrument runs under the environmental conditions that are specified in "Environmental requirements" on page 108. Exposure to extreme temperatures can adversely affect the instrument performance and shorten the life span of the instrument components.

Calibration	Recommended frequency	
ROI/Uniformity	Every two years (recommended)	
	 Always perform new Background and Dye calibrations after an ROI/Uniformity calibration. 	
	Note: Performing an ROI/Uniformity calibration invalidates all other calibrations.	

(continued)

Calibration	Recommended frequency
Background	Every two years (recommended)
	Background calibration can also be performed, as needed:
	 To check for contamination (depends on usage and laboratory conditions).
	 To obtain the most accurate data for the removal of background fluorescence.
	Note: Performing a Background calibration does <i>not</i> invalidate any other calibration.
Dye	Every two years (recommended)
	• During a Dye calibration, only the dyes on the given spectral calibration plate are calibrated.
	Note: Performing a Dye calibration for a given dye plate does <i>not</i> invalidate any other calibration.
RNase P instrument	After installing or moving the instrument
verification	After performing instrument calibrations (recommended)
	As needed to confirm instrument performance

Note: To prepare custom dye plates and to perform custom calibrations, see "Calibrate custom dyes" on page 59.

Calibration descriptions

Calibration description and purpose	Pass Criteria		
ROI/Uniformity			
 The software captures images for each optical filter. The software uses calibration data to map the increase in fluorescence to the plate wells during subsequent runs and to evaluate well-to-well consistency of the signals. 	The image for each filter distinguishes all wells of the plate. Each well in the image is distinct.		
Background			
• The software captures background images for each optical filter in the absence of sample and reagent, and it checks that the fluorescence from each well is below a fluorescence threshold.	The plate images for all filters are free of abnormal fluorescence.		
• The software uses calibration data to remove background fluorescence during a run.			
Note: You can also run this calibration to determine if contamination is related to the sample block or the plate.			



(continued)

Calibration description and purpose	Pass Criteria
Dye	
 The software extracts a spectral profile for each dye standard, then produces a set of spectral profiles plotted as fluorescence vs filter. The software uses calibration data to characterize and distinguish the individual contribution of each dye in the total fluorescence signals collected by the instrument. 	Dye spectra peak within the same filter as their group.

View the calibration status and set reminders

View calibration status and set reminders in the instrument

- 1. In the home screen, touch (a) Settings > Maintenance and Service > Calibrations > History and Reminders.
- 2. In the Calibration Reminders screen, view the status of each calibration type.
- **3.** (*Optional*) Touch a calibration row to view the history of that specific calibration type, then touch **Done**.
- 4. Touch Edit to set the calibration reminder settings. For each calibration type:
 - a. Slide the control **On** to enable the calibration reminder.
 - b. Edit the Exp interval and Remind me fields.
 - c. Touch Save.
- 5. (Optional) To transfer the calibration report, touch **Export** then follow the directions on the screen.
- 6. Touch Done.

View calibration status and set reminders in Thermo Fisher[™] Connect

Note: The calibration reminders feature requires a connection between the instrument and a computer network.

- 1. In Thermo Fisher[™] Connect, click for the **Instrument Connect** page.
- 2. Select any of your registered instruments.



- 3. In the Summary tab:
 - Click + Calibrations to view the status of each calibration type.
 - (Optional) Click + Calibration Reminders to set the calibration reminder time table and enter the notification email address(es).

Note: More than one email address can receive the calibration reminders.

Note: The settings are automatically saved.

- (Optional) In the Downloads section, click Maintenance Summary.pdf to download the calibration status report.
- 4. In the Calibrations History tab:
 - View the history of each calibration type.
 - *(Optional)* Click ± to download the calibration history report.

Perform ROI/uniformity, background, and dye calibrations

Workflow: Calibration

Note: The ROI and uniformity calibrations use the same calibration plate.

Perform an ROI calibration

▼

Perform a uniformity calibration

You are automatically prompted to perform background calibration.

▼

Perform a background calibration

Perform any time that ROI calibrations are current.

▼

Perform system dye calibrations

Perform any time that ROI and background calibrations are current.

▼

(Optional) Perform custom dye calibrations ("Calibrate custom dyes" on page 59) Perform any time that ROI and background calibrations are current.

Prepare a calibration plate

Materials required for calibration plate preparation

- Plate(s) for the calibration that you are performing:
 - ROI plate (the same plate is used for ROI and uniformity calibrations)
 - Background calibration plate
 - Dye calibration plates

Note: We recommend calibrating with all Spectral Dye Calibrations Plates available for your block configuration even if you are not using all the dyes in the plates.

Note: Do not discard the packaging for the calibration plates. Each calibration plate can be used up to 3 times if the following conditions are met:

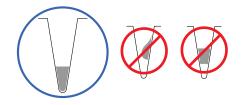
- The plate is stored in its packing sleeve at -15 to -25°C.
- The plate is used within 6 months after opening.
- . The plate is used before the plate expiry date
- Centrifuge with plate adapter; buckets cleaned before use
- Powder-free gloves
- Safety glasses

Thaw, vortex, and centrifuge a calibration plate

- 1. Remove the calibration plate from the freezer, then thaw the plate in its packaging. Keep plates protected from light until you perform the calibration.
 - Thaw each plate for 30 minutes.
 - Use each plate within 2 hours of thawing.

IMPORTANT! Do not remove the plate from its packaging until you are ready to use it. The fluorescent dyes in the wells of calibration plates are photosensitive. Prolonged exposure to light can diminish the fluorescence of the dyes.

- 2. While wearing powder-free gloves, remove the calibration plate from its packaging and retain the packaging. Do not remove the optical film.
- 3. Vortex the plate for 5 seconds, then centrifuge at $750-1,000 \times g$ for 2 minutes.
- 4. Confirm that the liquid in each well is at the bottom of the well and free of bubbles. If it is not, centrifuge the plate again.





IMPORTANT! Keep the bottom of the plate clean. Fluids and other contaminants on the bottom of the plate can contaminate the sample block and cause an abnormally high background signal.

Perform calibrations

1. In the instrument home screen:

Calibration	Touch	
ROI/Uniformity ^[1]	Settings ► Maintenance and Service ► Calibrations ► ROI and Uniformity	
Background ^[2]	 Settings ► Maintenance and Service ► Calibrations ► Custom ► Background 	
Dye		

^[1] Automatically followed by Background calibration.

^[2] Initiate via this route if performing Background calibration only.

2. Follow the instructions on the screen to start the calibration.

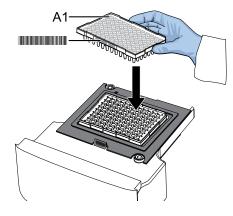
Note: Dye calibration only: Select the Dye Plate to run, then touch Next.

3. Load the plate.

a. Touch (a) to eject the instrument drawer.



- b. Load the plate onto the plate adaptor so that the following criteria are met.
 - Position well A1 of the plate in the top-left corner of the plate adapter.
 - Ensure that the barcode faces the front of the instrument.



IMPORTANT! The instrument should be used by trained operators who have been warned of the moving parts hazard.

Note: (*For 96-well 0.2-mL blocks only*) Do not remove the black plate adapter before loading a plate or tube strips. If used, tube strips can fit loosely in the adapter, but the heated cover will apply the appropriate pressure to seat the tube strips securely in the adapter.

Note: The 384-well and 96-well Fast (0.1-mL) block configurations do not require a plate adapter.

c. Touch (a) to close the instrument drawer.

4. Touch Start.

5. When the run is complete and the screen displays **Calibration Complete**, touch **View Results** to check the calibration status.

Calibration status	Action
Passed	Touch Next to proceed to the next required calibration.
Failed	See "Troubleshoot calibration failure" on page 51.

Note: You can view the calibration images only after the ROI/Uniformity and Background calibrations pass.

- 6. When the run ends, unload the plate.
 - a. Touch (a) to eject the instrument drawer.
 - **b.** Remove the plate.
 - c. Touch (a) to close the instrument drawer.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. Allow it to cool to room temperature before handling.

Note: If the instrument does not eject the plate, contact Support.

7. Return the plate to its original packaging.

Note: Each calibration plate can be used up to 3 times if the following conditions are met:

- The plate is stored in its packing sleeve at –15 to –25°C.
- The plate is used within 6 months after opening.
- · The plate is used before the plate expiry date

View calibration images and transfer results to USB

The instrument performs the ROI, Uniformity, and Background calibrations in sequence. You can view the calibration images after the Background calibration is complete.

- 1. In the Calibration Status screen, touch Details.
- 2. In the **Details** screen, touch a calibration type to view its images and plots.

Calibration	Example results indicating successful calibration
ROI	Green circles around all wells and bright well centers.
Note: Select the desired filter combination from the Filter Set dropdown list.	
Uniformity	Signals from each well following a uniform trend.



(continued)		
Calibration	Example results indicating successful calibration	
Background	Few, if any, signals with abnormally high fluorescence.	
Dye	Signals from each well following a uniform trend, and each dye peaks at the correct filter.	

- In the Calibration Status screen, touch Accept Results or Reject Results.
 Accepting the results saves the calibration data to the instrument and overwrites existing data.
- 4. (Optional) Touch Transfer EDS to transfer the calibration data to a USB.

Troubleshoot calibration failure

Observation	Possible cause	Recommended action
Calibration failed	The plate was improperly prepared.	Ensure the following:
		The correct plate was used for the calibration performed.
		The plate was properly thawed.
		The plate was properly centrifuged.
		The plate was properly sealed.
	The plate is damaged or contaminated.	Check for damage, improper plate seal, or contamination.
		Order a replacement plate. If the replacement plate fails, contact Support.
High fluorescence signal in individual wells	Signals that exceed the limit of normal fluorescence may indicate fluorescent contaminants on the plate or the sample block.	See "Identify contamination" on page 52.
Calibration failed but plate is undamaged	The incorrect plate was used for calibration performed.	Use the plate that matches the calibration performed.



Observation	Possible cause	Recommended action
Calibration failed but plate is undamaged	The plate was improperly prepared.	Repeat the calibration with the plate properly prepared.
(continued)		If the calibration fails again, order a replacement plate. If the replacement plate fails, contact Support.

Identify contamination

Signals that exceed the limit of normal fluorescence may indicate fluorescent contaminants on the calibration plate or the sample block. Common contaminants include ink residue from permanent pens, powder from disposable gloves, and dust.

- 1. View the calibration data and note the wells that failed the calibration.
- 2. Remove the plate from the instrument, rotate the plate 180°, then perform the calibration again.
- 3. Determine the location of the failed wells again as in step 1.

Position of failed wells	Action
Identical	The sample block is contaminated. Decontaminate the sample block (see page 68).
Reversed	The plate is contaminated. Discard the plate, then perform the calibration using a new calibration plate.

4. If the calibration fails after you decontaminate the sample block and replace the plate, contact Support.

Create a background plate (optional)

Whenever possible, use a background plate listed in Appendix C, "Parts and materials". These plates contain a buffer that accurately simulates the reagents used for PCR, and, therefore, produces high-quality calibration data.

If a background plate is not available, you can create one as described below.

Required materials:

- MicroAmp[™] optical 96-well reaction plate
- Optical adhesive cover or optical flat caps
- Pipettor, 200-µL (with pipette tips)
- Powder-free gloves
- Safety glasses
- Deionized water

IMPORTANT! Wear powder-free gloves while creating the background plate.

- 1. Remove a reaction plate from its box and place it on a clean, dry surface.
- 2. Aliquot deionized water to each well of the reaction plate. The recommended volume for a 96-well plate is 10-20 μ L. The recommended volume for 384-well plate is 5 μ L.
- 3. Seal the plate using an optical adhesive cover or optical flat caps.
- 4. Use the plate for background calibration.

Perform instrument verification using RNase P plates

Instruments are factory-calibrated, so calibration is not necessary for initial installation. However, perform instrument verification:

- After installation and before first use of the instrument.
- After performing instrument calibrations. (recommended)
- As needed to confirm instrument performance after calibrations, repair, preventative maintenance, or change to the system.

The instrument requires valid ROI/uniformity, background, and dye calibrations to perform instrument verification.

Instrument verification description

Purpose	Description	Pass criteria
Confirms the performance of the instrument.	Quantifies the number of copies of the human RNase P gene in samples with known concentrations of the corresponding DNA template.	The instrument passes performance specifications if the following inequality is true and the instrument successfully distinguishes between unknown populations A and B.
		$[(C_{tA}) - 3(\sigma_{CtA})] > [(C_{tB}) + 3(\sigma_{CtB})]$
		where:
		• C_{tA} = Average C_t of unknown population A
		 σ_{CtA} = Standard deviation of unknown population A
		• C_{tB} = Average C_t of unknown population B
		 σ_{CtB} = Standard deviation of unknown population B

RNase P instrument verification plate

The RNase P plate contains the reagents necessary for the detection and quantitation of genomic copies of the human RNase P gene (a single-copy gene encoding the RNase moiety of the RNase P enzyme). Each well contains: PCR master mix, RNase P primers, FAM[™] dye-labeled probe, and a known concentration of human genomic DNA template.

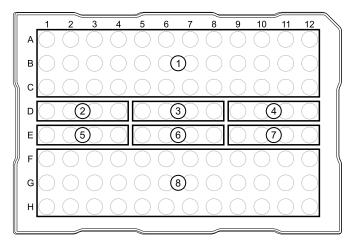


Figure 1 96-well RNase P plate

- (1) Unknown A (5,000)
- 2 NTC (no template control)
- ③ STD 1,250 copies
- (4) STD 2,500 copies

- (5) STD 5,000 copies
- 6 STD 10,000 copies
- (7) STD 20,000 copies
- (8) Unknown B (10,000)

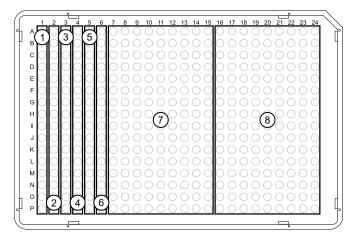


Figure 2 384-well RNase P plate

- (1) NTC (no template control)
- (2) STD 1,250 copies
- ③ STD 2,500 copies
- (4) STD 5,000 copies

- (5) STD 10,000 copies
- 6 STD 10,000 copies
- (7) Unknown A (5,000)
- (8) Unknown B (10,000)

Performance specifications pass criteria

After the run, the software calculates average copy number values and standard deviation values. The instrument passes performance specifications if the following inequality is true and the instrument successfully distinguishes between unknown populations A and B.

 $[(C_{tA}) - 3(\sigma_{CtA})] > [(C_{tB}) + 3(\sigma_{CtB})]$

- C_{tA} = Average C_t of unknown population A
- σ_{CtA} = Standard deviation of unknown population A
- C_{tB} = Average C_t of unknown population B
- σ_{CtB} = Standard deviation of unknown population B

The software automatically adjusts the threshold and omits a defined number of wells from the unknown populations to meet the performance specifications. To view any omitted wells, open the EDS file for the verification in the desktop software.

Prepare an RNase P plate

Materials required for RNase P plate preparation

- RNase P instrument verification plate
- Centrifuge with plate adapter; buckets cleaned before use
- Powder-free gloves
- Safety glasses

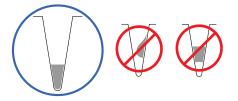
Thaw, vortex, and centrifuge an RNase P plate

IMPORTANT! Expose the RNase P plate to room temperature for no more than 45 minutes, inclusive of thawing and preparation time.

After thawing, the RNase P plate cannot be refrozen.

- 1. Remove the RNase P plate from the freezer, then thaw the plate in its packaging.
 - Thaw the plate for approximately 5 minutes.
 - Use the plate within 30 minutes of thawing.
- **2.** Confirm that the bench, vortex, and centrifuge are clean. Before use, wipe the vortex and centrifuge using a lint-free tissue.
- 3. While wearing powder-free gloves, remove the plate from its packaging.
- 4. Vortex the plate for 5 seconds, then centrifuge at $750-1,000 \times g$ for 2 minutes.

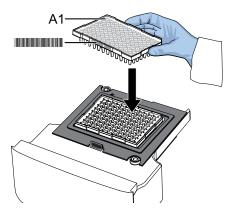
5. Confirm that the liquid in each well is at the bottom of the well and free of bubbles. If it is not, centrifuge the plate again.



IMPORTANT! Keep the bottom of the plate clean. Fluids and other contaminants on the bottom of the plate can contaminate the sample block and cause an abnormally high background signal.

Perform RNase P verification

- 1. In the home screen, touch (a) Settings > Maintenance and Service > RNase P Verification.
- 2. Load the plate.
 - a. Touch (a) to eject the instrument drawer.
 - b. Load the plate onto the plate adaptor so that the following criteria are met.
 - Position well A1 of the plate in the top-left corner of the plate adapter.
 - Ensure that the barcode faces the front of the instrument.



IMPORTANT! The instrument should be used by trained operators who have been warned of the moving parts hazard.

Note: (*For 96-well 0.2-mL blocks only*) Do not remove the black plate adapter before loading a plate or tube strips. If used, tube strips can fit loosely in the adapter, but the heated cover will apply the appropriate pressure to seat the tube strips securely in the adapter.

Note: The 384-well and 96-well Fast (0.1-mL) block configurations do not require a plate adapter.

c. Touch a to close the instrument drawer.



3. Touch Start.

4. When the run is complete and the screen displays **Verification Complete**, touch **View Results** to confirm the status of the run.

Calibration status	Action	
Passed	Instrument is ready for use.	
Failed	See "Troubleshoot verification failure" on page 57.	

- 5. In the RNase P Verification Status screen, touch:
 - Accept Results to save the results to the instrument
 - Reject Results to delete the RNase P verification results
 - Export Results to export the calibration results to a USB
- 6. When the run ends, unload the plate.
 - a. Touch (a) to eject the instrument drawer.
 - b. Remove the plate.
 - c. Touch a to close the instrument drawer.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. Allow it to cool to room temperature before handling.

Note: If the instrument does not eject the plate, contact Support.

Troubleshoot verification failure

Observation	Possible cause	Recommended action
Verification failed	The plate was improperly prepared.	 Ensure the following: The correct plate was used for the verification performed. The plate was properly thawed. The plate was properly centrifuged. The plate was properly sealed.
		Open the data file for the verification in the desktop software or Thermo Fisher [™] Connect to view the flags and troubleshooting details for failed wells.
	The plate is damaged or contaminated.	Check for damage, improper plate seal, or contamination.
		Order a replacement plate. If the replacement plate fails, contact Support.



Observation	Possible cause	Recommended action
High fluorescence signal	The reaction volume is not correct.	Ensure that reaction volumes in the plate are correct and match the volume that is entered in the Method tab.
	Signals that exceed the limit of normal fluorescence may indicate fluorescent contaminants on the plate or the sample block.	Examine the bottom of the reaction plate. If there is contamination, prepare a new plate and run the experiment again.
		Identify the location of contamination on the plate or sample block.
		1. Obtain a background plate.
		2. Follow the procedures that are described in "Identify contamination" on page 52.
Verification failed but plate is undamaged	The incorrect plate was used for verification.	Use the correct RNase P plate for verification.
	The plate was improperly prepared.	Repeat the verification with a new properly prepared plate.
		Note: The verification procedure is an experiment run, so each RNase P plate can only be used once.
		Open the data file for the verification in the desktop software or Thermo Fisher [™] Connect to view the flags and troubleshooting details for failed wells.
		If the verification fails again, order a replacement plate. If the replacement plate fails, contact Support.

Calibrate custom dyes

Custom dyes overview

The instrument can run assays designed with custom dyes. Custom dyes include:

- Dyes that are not manufactured by Thermo Fisher Scientific.
- Dyes or formulations of dyes that are not system dyes for the instrument.
- For custom dye requirements, see "Custom dyes" on page 13.

Workflow: Custom dye calibration

For each custom dye, determine the optimal dye concentration. Use this concentration to prepare all subsequent dye calibration plates.

Use a dilution series to determine an optimal custom dye concentration

Prepare a custom dye dilution plate

▼

Run the dilution plate as an experiment

▼

Determine the optimal dye concentration

▼

Calibrate the custom dye using the optimal concentration

Create a custom dye calibration plate

Add a custom dye to the instrument

▼

Perform a custom dye calibration

Use a dilution series to determine an optimal custom dye concentration

Custom dye dilution guidelines

Prepare a dilution series for each custom dye.

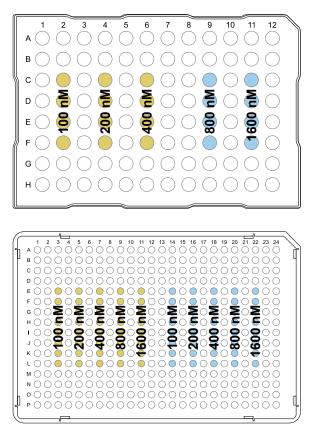
- Target several dye concentrations within a range of 100-2,000 nM.
- Choose a 2- or 3-fold difference in dilution points.
- Dispense 10–20 µL per well for 96-well plates or 5 µL per well for 384-well plates.
- Dilute the dye in buffer compatible with your master mix.
- (*Intercalating dyes only*) Add the appropriate amount of amplified PCR product to generate fluorescence.

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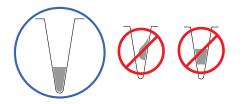
Prepare a custom dye dilution plate

IMPORTANT! Wear powder-free gloves throughout the procedure.

- 1. Prepare a 2- or 3-fold dilution series of the custom dye.
- Dispense aliquots of each dilution into the center of a reaction plate, then seal the plate. A full plate is not needed. See the following figures for suggested replicates.



- 3. Vortex the plate for 5 seconds, then centrifuge at $750-1,000 \times g$ for 2 minutes.
- 4. Confirm that the liquid in each well is at the bottom of the well and free of bubbles. If it is not, centrifuge the plate again.



IMPORTANT! Keep the bottom of the plate clean. Fluids and other contaminants on the bottom of the plate can contaminate the sample block and cause an abnormally high background signal.

4

Run the dilution plate as an experiment

1. Load the plate into the instrument.



CAUTION! The instrument should be used by trained operators who have been warned of the moving parts hazard.

- 2. Set up a genotyping experiment on the instrument.
 - Load an experiment created in the desktop or cloud software.
 - a. In the home screen, touch Load Experiment.
 - b. Select the experiment location, then the experiment file.
 - Create a new experiment on the instrument touchscreen.
 - a. In the home screen, touch Open Template > Genotyping > Genotyping Post.
 - b. (Optional) In the Properties tab, edit the experiment properties.
 - c. In the **Method** tab, set the hold temperature to 60°C with a 2 minute hold and enter the appropriate reaction volume.
 - d. In the **Plate** tab, enter the dilution series information for the appropriate wells.
- 3. Touch Start Run.
- 4. When the run is complete, download the results for analysis.
- 5. Unload the plate from the instrument.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. Allow it to cool to room temperature before handling.

Note: If the instrument does not eject the plate, contact Support.

Determine the optimal dye concentration

Review the dye signal data and select the dilution to use for dye calibration.

1. In the **Results** tab of the desktop or cloud software, select **Raw Data Plot**.

This plot displays the raw fluorescence signal of each optical filter, for individual wells.

Note: The Raw Data Plot cannot be viewed on the instrument touchscreen.

2. For each replicate population of dilutions, select the wells in the Plate Layout to view in the plot.



3. Examine the raw data to identify the wells yielding signals according to the ranges shown in the following table.

Plate type	Acceptable signal range ^[1]
96-well	800,000 to 3,200,000
384-well	400,000 to 2,000,000

^[1] Signal range for the optical filter where the dye is brightest.

- 4. (*Optional*) Export the raw data, then calculate the average fluorescence value for each concentration.
- 5. Select the lowest (optimal) dye concentration that falls within the acceptable signal range.

Calibrate the custom dye

Create a custom dye calibration plate

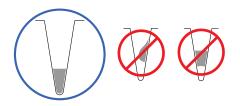
IMPORTANT! Wear powder-free gloves while creating the dye plate.

Create a full plate of the custom dye diluted to the optimal concentration:

1. Dilute the custom dye to the optimal concentration in buffer.

Prepare an adequate volume, using a volume range of 10–20 μ L/well for a 96-well plate or 5 μ L/well for a 384-well plate.

- 2. Pipet the appropriate volume of the diluted custom dye to the plate wells.
- 3. Seal the plate.
- 4. Vortex the plate for 5 seconds, then centrifuge at $750-1,000 \times g$ for 2 minutes.
- 5. Confirm that the liquid in each well is at the bottom of the well and free of bubbles. If it is not, centrifuge the plate again.



IMPORTANT! Keep the bottom of the plate clean. Fluids and other contaminants on the bottom of the plate can contaminate the sample block and cause an abnormally high background signal.



Add a custom dye to the instrument

- 2. Touch Add Custom Dye.
- 3. Enter the dye information:

Field/option	Action	
Custom Dye Name	Enter a name for the custom dye.	
	IMPORTANT!	
	Do not use a system dye name for a custom dye name.	
	Dye names are spacing sensitive and cannot contain special characters.	
Туре	Select:	
	 Reporter—The dye works in conjunction with a quencher dye to report an increase of PCR product. 	
	 Quencher—The dye suppresses the fluorescence of a reporter dye until amplification of PCR product. 	
	• Both —The dye can be used as a reporter or quencher dye.	

4. Touch Save.

IMPORTANT! You must also add the custom dye to the desktop software dye library before creating, running, or analyzing experiments that use the custom dye. For detailed information, see the *QuantStudio*[™] *Design and Analysis Desktop Software User Guide*.

Perform a custom dye calibration

1. Load the plate into the instrument.



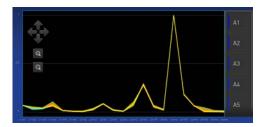
CAUTION! The instrument should be used by trained operators who have been warned of the moving parts hazard.

- **3.** Touch the custom dye to calibrate.
- 4. Review the custom dye information, (optional) make changes, then touch Update.
- 5. Enter the calibration temperature.
- 6. (Optional) Touch Reagents, then enter reagent information.
- 7. Touch Start.

- 8. When the run is complete and the screen displays Calibration Complete, touch View Results ► Details.
- **9.** Review the plot. Passing calibration results show uniform signals with peaks that are aligned with the dye wavelength.

Peak filter	Filter wavelength (nm) ^[1]	
	Excitation	Emission
x1-m1	470 ± 15	520 ± 15
x2-m2	520 ± 10	558 ± 12
x3-m3	550 ± 10	587 ± 10
x4-m4	580 ± 10	623 ± 14
x5-m5	640 ± 10	682 ± 14
x6-m6	662 ± 10	711 ± 12

^[1] The central wavelengths are the optimized wavelengths.



Note: Example dye calibration plot. The peaks for your dye may align with a different filter set.

10. Select an action depending on whether the custom dye calibration passed or failed.

Calibration status	Action	
Passed	Touch Accept Results or Reject Results.	
	Note: Accepting the results saves the calibration data to the instrument and overwrites existing data.	
	• (<i>Optional</i>) Touch Transfer EDS to transfer the calibration data to a USB.	
Failed	Perform the calibration again using a new custom dye plate.See "Troubleshoot calibration failure" on page 51.	

11. Unload the plate from the instrument.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the plate temperature can reach 100°C. Allow it to cool to room temperature before handling.

Note: If the instrument does not eject the plate, contact Support.



Calibrate for a custom melt curve experiment

Note: A custom melt calibration calibrates a custom dye and a melt calibration at the same time.

Before running the custom melt calibration:

- "Add a custom dye to the instrument" on page 63.

In the home screen:

- 1. Touch (a) Settings > Maintenance and Service > Calibrations > Custom > Custom Melt.
- 2. Touch **PCR + Melt** or **Melt only** as appropriate for the kit you are using.
- 3. Load the plate into the instrument.
- 4. Select or add a dye, then select a filter set appropriate for your dye's wavelength (see filterwavelength table below).

Note: Refer to your reagent kit documentation for dye name and wavelength information.

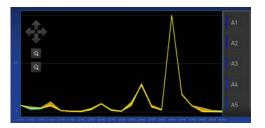
Peak filter	Filter wavelength (nm) ^[1]	
	Excitation	Emission
x1-m1	470 ± 15	520 ± 15
x2-m2	520 ± 10	558 ± 12
x3-m3	550 ± 10	587 ± 10
x4-m4	580 ± 10	623 ± 14
x5-m5	640 ± 10	682 ± 14
x6-m6	662 ± 10	711 ± 12

^[1] The central wavelengths are the optimized wavelengths.

IMPORTANT! If the selected filter set does not match your reagent kit documentation, then the incorrect wavelength may be collected during a run.

- 5. (Optional) Touch Reagents, then enter reagent information.
- 6. Touch Start.
- 7. When the run is complete and the screen displays Calibration Complete, touch View Results ► Details.

8. Review the plot. Passing calibration results show uniform signals with peaks that are aligned with the dye wavelength.



Note: Example dye calibration plot.

The peaks for your dye may align with a different filter set.

9. Select an action depending on whether the custom dye calibration passed or failed.

Calibration status	Action	
Passed	Touch Accept Results or Reject Results.	
	Note: Accepting the results saves the calibration data to the instrument and overwrites existing data.	
	• (<i>Optional</i>) Touch Transfer EDS to transfer the calibration data to a USB.	
Failed	Perform the calibration again using a new custom dye plate.See "Troubleshoot calibration failure" on page 51.	

10. Unload the plate from the instrument.

Note: You must also add the custom dye to the desktop software dye library before creating, running, or analyzing experiments that use the custom dye. For detailed information, see the *QuantStudio*[™] *Design and Analysis Desktop Software User Guide*.

Note: To perform a custom melt experiment, you can either create a Standard Curve or a Custom experiment with melt, then specify the data points per degree in the method.



Maintain the instrument

Backup or restore the instrument	67
Decontaminate the sample block	68
Replace the instrument fuses	71
Power on or off, store, and move	72

IMPORTANT! This chapter contains user maintenance procedures for the instrument. Procedures other than those described in this document must be performed by a qualified Thermo Fisher Scientific service representative.

Backup or restore the instrument

In the home screen, touch (a) Settings > Maintenance and Service > Backup/Restore.

То	Action
Backup	1. Touch Backup Instrument.
	2. Select a location to store the backup settings for your instrument.
	 If backing up to a USB drive, insert a USB drive into the front-panel USB port.
	4. Enter a backup file name, then touch Done.
	5. Select the elements to backup, or leave them all selected.
	6. Touch Backup.
Restore (Administrator	1. Touch Restore a Backup.
only)	2. Select the storage location of the backup settings.
	3. If restoring from a USB drive, insert a USB drive into the front-panel USB port.
	4. Select the backup file, then touch Restore.

Decontaminate the sample block

Perform this procedure to eliminate fluorescent contaminants from the instrument sample block. Contamination is generally evident in failed background calibrations where one or more wells consistently exhibit abnormally high signals.



CAUTION! PHYSICAL INJURY HAZARD. Do not remove the instrument cover. There are no components inside the instrument that you can safely service yourself. If you suspect a problem, contact Support.



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the sample block temperature can reach 100°C. Allow it to cool to room temperature before handling.



CAUTION! Before using a cleaning or decontamination method other than those recommended by Thermo Fisher Scientific, confirm with Thermo Fisher Scientific that the proposed method will not damage the equipment.

Materials required

- Safety glasses
- Powder-free gloves
- Tissue, lint-free
- Cotton or nylon swabs and lint-free cloths
- Pipette (100-µL) with pipette tips
- Deionized water
- Ethanol, 95% solution
- Na-hypochlorite, (0.1% v/v) solution

Clean the sample block



CAUTION! PHYSICAL INJURY HAZARD. During instrument operation, the sample block temperature can reach 100°C. Allow it to cool to room temperature before handling.

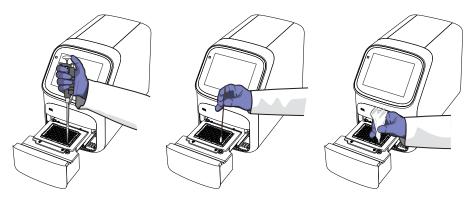
IMPORTANT! Wear powder-free gloves when you perform this procedure.

IMPORTANT! Always use deionized water to rinse wells after cleaning with bleach or ethanol solution.

- 1. Identify the contaminated wells of the sample block (see "Identify contamination" on page 52).
- 2. Prepare the instrument and access the sample block:
 - a. Power off and unplug the instrument, then allow it to cool for 15 minutes.



- **b.** Touch (a) to eject the instrument drawer.
- **3.** Rinse the contaminated wells with deionized water (see "Detailed procedures for cleaning the sample block" on page 70).



- 4. Close the drawer and test the sample block for contamination:
 - a. Push the instrument drawer back in to the instrument.
 - **b.** Plug in, then power on the instrument.
 - c. Perform a background calibration to confirm that you have eliminated the contamination.
- 5. If the contamination remains, clean the contaminated wells using a 95% ethanol solution:
 - a. Repeat step 2 step 3.
 - b. Clean the contaminated wells using a 95% ethanol solution (see "Detailed procedures for cleaning the sample block" on page 70).
 - c. Repeat step 3 step 4 to rinse the sample block with deionized water and to confirm that you have eliminated the contamination.

IMPORTANT! Always use deionized water to rinse wells after cleaning with bleach or ethanol solution.

- **6.** If the contamination still remains, clean the contaminated wells using a Na-hypochlorite (0.1% v/v) solution:
 - a. Repeat step 2 step 3.
 - **b.** Clean the contaminated wells using a Na-hypochlorite (0.1% v/v) solution (see "Detailed procedures for cleaning the sample block" on page 70).
 - c. Repeat step 3 step 4 to rinse the sample block with deionized water and to confirm that you have eliminated the contamination.

IMPORTANT! Always use deionized water to rinse wells after cleaning with Na-hypochlorite or ethanol solution.

7. If the contamination continues to remain, contact Support.

Detailed procedures for cleaning the sample block

IMPORTANT! Use these cleaning procedures *only* in conjunction with the complete decontamination procedure (see "Clean the sample block" on page 68).

- Rinse the sample block with deionized water.
 - a. Pipet a small volume of deionized water into each contaminated well.
 - b. In each well, pipet the water up and down several times to rinse the well.
 - c. Pipet the water to a waste beaker.
 - d. Use a cotton swab to scrub inside of each contaminated well.
 - e. Use a lint-free cloth to absorb the excess deionized water.
- Clean the sample block with 95% ethanol.
 - a. Pipet a small volume of 95% ethanol solution into each contaminated well.
 - b. In each well, pipet the solution up and down several times to rinse the well.
 - c. Pipet the ethanol solution to a waste beaker.

IMPORTANT! Always use deionized water to rinse wells after cleaning with bleach or ethanol solution.

- Clean the sample block with Na-hypochlorite (0.1% v/v) solution.
 - a. Pipet a small volume of Na-hypochlorite (0.1% v/v) solution into each contaminated well.
 - b. In each well, pipet the solution up and down several times to rinse the well.
 - c. Pipet the bleach solution to a waste beaker.

IMPORTANT! Always use deionized water to rinse wells after cleaning with bleach or ethanol solution.

Replace the instrument fuses

CAUTION! FIRE HAZARD. For continued protection against the risk of fire, replace fuses only with listed and certified fuses of the same type and rating as those currently in the instrument.

Materials required

- Fuses (2) 10A, Time-Lag T, 250VAC, 5 × 20mm
- Safety glasses
- Powder-free gloves
- Screwdriver, flathead

Replace the fuses

- 1. Power off and unplug the instrument, then allow it to cool for 15 minutes.
- 2. Using a flat-head screwdriver, unscrew and remove the fuse holder.



3. Remove each fuse from its fuse holder and inspect it for damage. Carbon typically coats the inside of failed fuses.

Good	Failed

4. Replace each failed fuse.

Note: The voltage and amperage ratings are on the fuse holder.

- 5. Install the fuse holder back into the instrument.
- Plug in, then power on the instrument.
 The installation is successful if the instrument powers on.

Note: Fuse failure can result from fluctuations in the supplied power to the system. To prevent further failures, consider installing an electrical protective device, such as a UPS or a surge protector. If issues with the fuse persist, contact Support.

Power on or off, store, and move

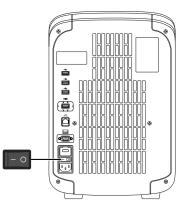
Enable Sleep Mode

- 1. In the home screen, touch (a) Settings > Instrument Settings > Sleep Mode.
- 2. Slide the control **On** to enable sleep mode.
- 3. Touch Edit Time, then enter the time length of inactivity before the instrument enters sleep mode.
- 4. Touch Enter, then touch OK.

Power on the instrument

- 1. Touch anywhere on the touchscreen to determine if the instrument is in sleep mode. If the home screen is displayed, the instrument is already powered on.
- 2. If the home screen does not display, power on the instrument by pressing the switch on the rear panel.

If left unattended, the instrument automatically enters sleep mode (enabled by default) to conserve power.



Power off the instrument

The instrument operates in low-power mode when not in use. However, the instrument can be powered off completely so that the components use no power.

Note: To power off the instrument for >1 week, see "Prepare the instrument to store, move, or ship" on page 73.

- 1. Power off the instrument using the power switch on the back of the instrument.
- 2. Power off the computer.

Prepare the instrument to store, move, or ship

In the home screen:

- 1. Touch (a) Settings > Maintenance and Service > Ship Prep Mode > Next.
- 2. Touch (a) to eject the instrument drawer.
- 3. Load the packing plate or an empty plate, then touch to close the drawer.
- 4. Touch Lock Block.
- 5. Power off the instrument using the power switch on the back of the instrument.

The instrument is now ready to store, move, or ship.

Move the instrument



CAUTION! PHYSICAL INJURY HAZARD. Do not attempt to lift the instrument or any other heavy objects unless you have received related training. Incorrect lifting can cause painful and sometimes permanent back injury. Use proper lifting techniques when lifting or moving the instrument. At least two people are required to lift it.

IMPORTANT! Moving your instrument can create subtle changes in the alignment of the instrument optics. Recalibrate the instrument if necessary.

- Ensure that the surface on which you place the instrument can support at least 35 kg (77 lbs).
- Ensure that the path to transport the instrument is clear of obstructions.
- At least two people are needed to lift and carry the instrument.
- Keep your spine in a good neutral position.
- Bend at the knees and lift with your legs.
- Do not lift an object and twist your torso at the same time.
- Coordinate your intentions with your assistant before lifting and carrying.

IMPORTANT! After moving the instrument, perform an RNase P instrument verification run. If the run fails, perform ROI/ uniformity, background, and dye calibrations.

Return the instrument for service

The service process requires 2 to 3 weeks.

Before returning the instrument for service, perform the following tasks.

- 1. Back up the instrument (see "Backup or restore the instrument" on page 67).
- 2. In the home screen, touch (a) Settings > Instrument Settings > Reset Factory defaults.
- Set the instrument to Ship Prep Mode (see "Prepare the instrument to store, move, or ship" on page 73).

To return the instrument for service, perform the following tasks.

- 1. Contact your local customer care center or technical support group to obtain a copy of the Certificate of Instrument Decontamination, a service notification, a service call number, and packaging materials (if required).
- 2. Follow the instructions in the form to decontaminate the instrument.

IMPORTANT! The instrument must be decontaminated before packing it for shipping.

- 3. Complete and sign a copy of the Certificate of Instrument Decontamination.
- 4. Fax the Certificate of Instrument Decontamination to the customer care center.
- 5. Pack the instrument in the provided packaging and follow the instructions in the table below.

Prepare	Include	Exclude
 Transfer any data files from the instrument. Load an empty plate in the sample block. Use the touchscreen to place the instrument in ship mode. Note: The empty plate and ship mode protects the internal components of the instrument during transport. 	 Instrument Completed and signed Certificate of Instrument Decontamination Note: The instrument will not be accepted for service without a hard copy of the Certificate of Instrument Decontamination. 	 Any accessories, including: Power cord Ethernet cable USB drive Wireless adapter Note: If included with the instrument, these items will be disposed of during service and not returned.

6. Attach the postage provided with the Certificate of Instrument Decontamination to the box, then ship the instrument to the designated facility.



Configure the instrument and manage instrument profiles

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Manage the instrument name (Administrator only)	88
Set the date and time (Administrator only)	89
Manage the Sign Out Timer (Administrator only)	89
	Initial start-up

Initial start-up

Perform these tasks during initial instrument start-up or after you restore factory defaults (see "Restore factory defaults" on page 87).

- 1. (Optional) Select one of the following options.
 - Configure the instrument for either wired connection or wireless network connection (see "Configure the network" on page 87).
 - Touch Neither/Decide Later.
- 2. (Optional) Enter an instrument name (see "Manage the instrument name (Administrator only)" on page 88).
- 3. Set the date and time (see "Set the date and time (Administrator only)" on page 89).
- 4. Select one of the following options.
 - Select a region for Thermo Fisher[™] Connect.
 - Touch **Next** to continue.



- 5. Create an administrator profile (see "Create an administrator instrument profile during initial startup" on page 76).
- 6. Select one of the following options.
 - Link the instrument profile to Thermo Fisher[™] Connect (see "Link an instrument profile to Thermo Fisher[™] Connect" on page 77).
 - Touch Skip.

Manage profiles

То	See
Create a profile	 "Create an administrator instrument profile during initial start-up" on page 76 "Create a new instrument profile" on page 25
Configure a profile	 "Link an instrument profile to Thermo Fisher[™] Connect" on page 77 "Edit an instrument profile" on page 77
View or manage all profiles	"Manage all instrument profiles (Administrator only)" on page 76

Create an administrator instrument profile during initial start-up

During initial start-up, the user is automatically prompted to create an administrator instrument profile. See "Initial start-up" on page 75.

- The first instrument profile that is created during installation is given administrator privileges.
- Administrators can grant administrative privileges to other users. See "Manage all instrument profiles (Administrator only)" on page 76.
- 1. Touch Name, enter a username, then touch Done.
- 2. Touch PIN, enter a four-digit numerical password, then touch Enter.

Note: Touch the Show PIN checkbox to switch PIN display on or off.

- 3. Touch Confirm PIN, then repeat step 2.
- 4. Touch Create profile.

Manage all instrument profiles (Administrator only)

- 1. In the home screen, access the **All Profiles** tab.
 - Touch (2) My Profile > All Profiles.
 - Touch Settings ► Manage Users ► Manage Profiles ► All Profiles.

A list of users, the date the profile was created, and the user type displays.

- 2. Select the instrument profile to edit.
- **3.** Edit the profile.
 - To delete the profile, touch **Delete profile > Delete**.
 - To reset the PIN, touch Reset PIN Reset.
 - The user will be directed to enter a new PIN at the next sign in.
 - To enable or disable administrative privileges, slide the control to **Administrator** or **Standard**, respectively.
- 4. Touch Done.

Edit an instrument profile

1. In the home screen, touch (a) My Profile.

- 2. Touch Edit.
- 3. Select the fields to edit, then make changes.
- 4. Touch Done.

Link an instrument profile to Thermo Fisher[™] Connect

Linking your instrument profile to Thermo Fisher[™] Connect cloud-based platform allows you to access the following functions:

- View an instrument status from the cloud.
- Download templates from your cloud storage to the instrument.
- Transfer run data from the instrument to your cloud storage.

To link an instrument profile to Thermo Fisher[™] Connect, see the following sections:

- "Recommended order to set up profiles" on page 78
- "Overview of local instrument profiles and Thermo Fisher[™] Connect profiles" on page 78
- "Thermo Fisher" Connect instrument profile roles and functions" on page 79
- "Link the instrument to your Thermo Fisher[™] Connect account" on page 80
- "Link a local profile to a Thermo Fisher[™] Connect profile" on page 82
- "Unlink a Thermo Fisher[™] Connect account" on page 83
- "If you link when you are signed in to the instrument" on page 84
- "Change the PIN for a Thermo Fisher[™] Connect profile" on page 85



Recommended order to set up profiles

Task	Description
Create a profile for the instrument	The first profile on the instrument becomes the instrument administrator.
administrator.	See "Create an administrator instrument profile during initial start-up" on page 76.
	A Thermo Fisher [™] Connect administrator is distinct from an instrument administrator.
Select a Thermo Fisher [™] Connect administrator.	The first Thermo Fisher [™] Connect profile to be linked to the instrument is automatically assigned the role of a Thermo Fisher [™] Connect administrator. It is recommended that the first Connect profile to be linked is the one that should be a Thermo Fisher [™] Connect administrator.
	The roles can be updated at a later time on the Thermo Fisher [™] Connect platform.
	Create a local instrument profile. Thermo Fisher [™] Connect -enabled features are not available.
Each person creates a profile.	<i>(Optional)</i> Link a local instrument profile to a Thermo Fisher [™] Connect profile.
	Sign in with a Thermo Fisher [™] Connect profile. Thermo Fisher [™] Connect-enabled features are available.
Assign additional instrument administrators.	Any of the profiles can be assigned the role of instrument administrator. The local profile must be created first or the Thermo Fisher [™] Connect profile must be linked first.

Overview of local instrument profiles and Thermo Fisher[™] Connect profiles

- Local instrument profile
 - Experiment template files and data files are stored on the instrument.
 - Experiment template files and data files can be transferred to the desktop software.
 - Email notifications are not available.
 - Separate profiles are required for each instrument if multiple instruments are used.
- Thermo Fisher[™] Connect profile
 - Experiment template files and data files are stored on the instrument.
 - Experiment template files and data files can be transferred to the Thermo Fisher[™] Connect platform or the desktop software.
 - Email notifications are available.
 - The same profile can be used for multiple instruments.

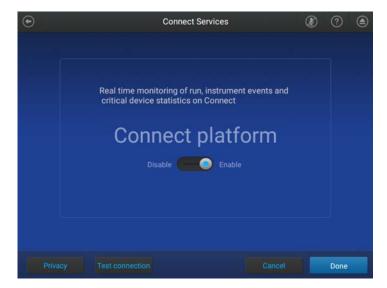
Thermo Fisher[™] Connect instrument profile roles and functions

The first user who links their local instrument profile to their Thermo Fisher[™] Connect account is assigned a Thermo Fisher[™] Connect profile with administrator role.

Instrument profile	Location	Functions allowed
Standard	Thermo Fisher [™] Connect	 Create, save, open, import, and run template files Create and modify run settings View and export data files
Administrator	Thermo Fisher [™] Connect	 All the permissions of a local administrator profile, plus the following functions performed in Thermo Fisher[™] Connect: See a list of all the Thermo Fisher[™] Connect profiles that are linked to the instrument Assign Thermo Fisher[™] Connect administrator roles to one or more users Remove a user from an instrument Disconnect the instrument from Thermo Fisher[™] Connect Change the instrument name

Test the connection to the Thermo Fisher[™] Connect platform

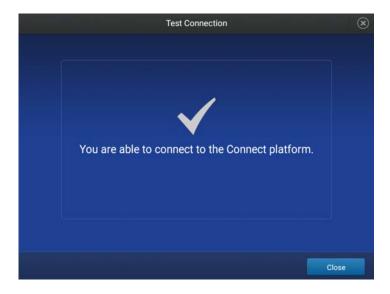
 In the home screen, tap (a) (Settings) ➤ Maintenance and service ➤ Connect services. The Connect Services screen is displayed.



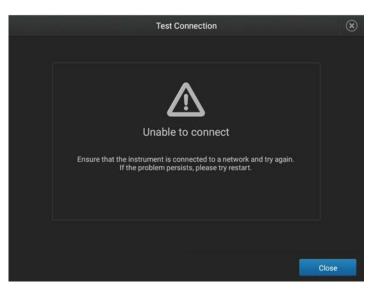
2. Tap Test connection.

If the connection can be established, **You are able to connect to the Connect platform** will be displayed.





If the connection cannot be established, Unable to connect will be displayed.



3. Tap Close.

Link the instrument to your Thermo Fisher[™] Connect account

This section describes using your Thermo Fisher[™] Connect account when you use the instrument for the first time.

The first time you use your Thermo Fisher[™] Connect account on an instrument, you will be prompted to create a four-digit numerical PIN. This PIN is to use when signing in to the instrument with your Thermo Fisher[™] Connect account. It will apply to all other instruments when you use your Thermo Fisher[™] Connect account. This does not change the password when signing in to your Thermo Fisher[™] Connect account on a browser.

After the PIN is set up on the instrument, it must be changed with your Thermo Fisher[™] Connect account on a browser, see "Create an administrator instrument profile during initial start-up" on page 76.

The first time the instrument is used with a Thermo Fisher[™] Connect account, a region must be selected.

- 1. In the Sign In screen, tap Get started ➤ Create a local instrument file ➤ Connect to your Thermo Fisher[™] Connect account.
- 2. (Optional) Select the appropriate region.

Option	Description
China	For users in China
U.S.	For users in any country other than China

3. Tap a connection option.

Option	Action
Mobile devices	Note: Before selecting this option, install and sign in to the Thermo Fisher [™] Connect application on your mobile device.
	On the instrument: 1. Tap [] Mobile devices.
	 Hold the camera on your mobile device over the QR code that is displayed on the touchscreen. Tap Close.
PC	 A link code is displayed on the instrument. On a computer: 1. Access the Thermo Fisher[™] Connect platform. 2. Click Add instrument. 3. Select QuantStudio.
	4. Enter the link code.

4. *(Optional)* In the Enter PIN screen, tap the PIN (4 digits required) field, enter a four-digit numerical PIN, then tap Enter.

Tap the **Show PIN** checkbox to show or hide the PIN.

- 5. (Optional) Tap the Confirm PIN field, enter the four-digit numerical PIN again, then tap Enter.
- 6. Tap Done.

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Link a local profile to a Thermo Fisher[™] Connect profile

This section describes linking a local instrument profile to a Thermo Fisher[™] Connect profile, if a local instrument profile was created first.

The first time you use your Thermo Fisher[™] Connect account on an instrument, you will be prompted to create a four-digit numerical PIN. This PIN is to use when signing in to the instrument with your Thermo Fisher[™] Connect account. It will apply to all other instruments when you use your Thermo Fisher[™] Connect account. This does not change the password when signing in to your Thermo Fisher[™] Connect account on a browser.

After the PIN is set up on the instrument, it must be changed with your Thermo Fisher[™] Connect account on a browser, see "Change the PIN for a Thermo Fisher[™] Connect profile" on page 85.

The first time the instrument is used with a Thermo Fisher[™] Connect account, a region must be selected.

For a detailed description of the profile, see "If you link when you are signed in to the instrument" on page 84.

(Optional) Test the connection, see "Test the connection to the Thermo Fisher[™] Connect platform" on page 79.

- In the home screen, tap () (Profile). The My Profile screen is displayed.
- Tap Connect ➤ Link Account. The Connect to Connect Platform screen is displayed.
- 3. (Optional) Select the appropriate region.

Option	Description
China	For users in China
U.S.	For users in any country other than China

4. Tap a connection option.

Option	Action
Mobile devices	Note: Before selecting this option, install and sign in to the Thermo Fisher [™] Connect application on your mobile device.
	On the instrument:
	1. Tap 🛄 Mobile devices.
	2. Hold the camera on your mobile device over the QR code that is displayed on the touchscreen.
	3. Tap Close.
	A link code is displayed on the instrument.
	On a computer:
	1. Access the Thermo Fisher [™] Connect platform.
	2. Click Add instrument.
	3. Select QuantStudio.
	4. Enter the link code.

5. *(Optional)* In the Enter PIN screen, tap the PIN (4 digits required) field, enter a four-digit numerical PIN, then tap Enter.

Tap the **Show PIN** checkbox to show or hide the PIN.

- 6. (Optional) Tap the Confirm PIN field, enter the four-digit numerical PIN again, then tap Enter.
- 7. Tap **Done**.

Unlink a Thermo Fisher[™] Connect account

Unlinking a Thermo Fisher[™] Connect is done from the Thermo Fisher[™] Connect platform on your computer.

For more information about unlinking a Thermo Fisher[™] Connect account, see "If you link when you are signed in to the instrument" on page 84.

- 1. Sign in to your account on the desktop Thermo Fisher[™] Connect platform.
- 2. In the left pane, click **(Instrument)**.
- 3. Select the instrument, then click **m** Disconnect.
- 4. Tap Confirm.

QuantStudio $^{^{
m M}}$ 3 and 5 Real-Time PCR Systems Installation, Use, and Maintenance Guide

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If you link when you are signed in to the instrument

In this scenario, your local instrument profile name is created manually on the instrument before you link. Your local instrument profile name differs from your Thermo Fisher[™] Connect instrument profile name.

Phase	Steps that occur
Before you link:	 You enter your local instrument profile name in the Sign In screen. Your local instrument profile (<i>UserABC</i>) is displayed in the home screen of the instrument. All plates and results that you create are accessible only when you are signed in with your local instrument profile.
When you link:	 You link your local instrument profile (see "Link a local profile to a Thermo Fisher[™] Connect profile" on page 82). If this is the first time you link, a Thermo Fisher[™] Connect instrument profile is created using the <i>FirstNameLastInitial</i> of the user name from your thermofisher.com account. Example: <i>User1@thermofisher.com</i> First name is <i>User</i>, Last name is <i>Gray</i>. Thermo Fisher[™] Connect account username is <i>User G</i>. Your local instrument profile (<i>UserABC</i>) is linked to your Thermo Fisher[™] Connect account (<i>User1@thermofisher.com</i>). Your Thermo Fisher[™] Connect instrument profile (<i>User G.</i>) replaces your local instrument profile.
After you link:	 Your Thermo Fisher[™] Connect instrument profile (User G.) and is displayed in the home screen of the instrument. Plate files and data files from your local instrument profile can be transferred to Thermo Fisher[™] Connect. New plate files and data files are saved under your Thermo Fisher[™] Connect instrument profile.
If your Thermo Fisher [™] Connect account is unlinked:	 Your local instrument profile (<i>UserABC</i>) is displayed in the home screen of the instrument. Plate files and data files that were saved under your Thermo Fisher[™] Connect instrument profile are accessible under your local instrument profile. Plate files and data files are saved under local instrument profile and can be copied to Thermo Fisher[™] Connect (see "Manage templates (EDT files)" on page 42 and "Transfer EDS files from the instrument home screen" on page 41). Your local instrument profile name (<i>UserABC</i>) is available for selection in the Sign In screen.

Change the PIN for a Thermo Fisher[™] Connect profile

This section describes changing a PIN for a Thermo Fisher[™] Connect profile. To change a PIN for a local instrument profile, see "Manage all instrument profiles (Administrator only)" on page 76.

- 1. Sign in to Thermo Fisher[™] Connect on a browser.
- 2. Click Update PIN number.
- 3. In the **Update PIN number** dialog box, enter a new PIN, then enter the new PIN a second time to confirm it.
- 4. Click Send.

The 4-digit PIN to sign in to instruments with your Thermo Fisher[™] Connect profile is updated. This change applies to all of the instruments that you access with your Thermo Fisher[™] Connect profile.

Enable SAE mode (Administrator only)

Security, Audit, and e-Signature (SAE) functions are only available for the QuantStudio[™] 5 System.

The SAE module works with the instrument and desktop software to record and restrict various user activities. Audits and e-Signatures are tracked in the desktop software.

In the home screen:

- 1. Touch (a) Settings > Instrument Settings > SAE Mode.
- 2. Slide the control **On** to enable SAE on the instrument.
- 3. Touch OK.

The following instrument functions are disabled in SAE mode.

• Start a run from the instrument.

Note: All instrument runs must be started from desktop software that has SAE functions enabled.

- Edit an instrument name.
- Edit an instrument date or time.
- Restore to factory defaults.
- Perform a software update.



Require instrument profile sign-in (Administrator only)

- 1. In the home screen, touch (a) Settings > Manage Users > Sign In Required.
- 2. Slide the control On.

Only signed-in users are allowed to access the instrument for any task, including access to **Settings**.

3. Touch Done.

Enable remote instrument monitoring (Administrator only)

- 1. In the home screen, touch (a) Settings > Maintenance and Service > Monitoring.
- 2. Configure the instrument monitoring settings.

Option	Description
Remote Monitoring Service	Slide the control On to enable the Remote Monitoring Service . Enabling the Remote Monitoring Service allows the instrument to automatically notify Thermo Fisher Scientific support teams in real time of potential instrument issues. The service monitors and sends general <i>instrument</i> data, but the service does not monitor or send <i>customer</i> data.
Thermo Fisher [™] Connect Monitor	Slide the control On to enable users to access real-time monitoring of amplification plots from the Thermo Fisher [™] Connect dashboard (cloud-based platform).

3. Touch OK.

Update instrument software (Administrator only)

- 1. In the home screen, touch (a) Settings > Maintenance and Service > Software Update.
- 2. Select the location of the update files.
- 3. When prompted, confirm your request to update the software.

Restore factory defaults

This procedure can only be performed by an Administrator.

IMPORTANT! Back up the instrument before restoring factory defaults (see "Backup or restore the instrument" on page 67).

- 1. In the home screen, touch (a) Settings > Instrument Settings > Restore Factory Defaults.
- 2. Touch Restore Factory Defaults.
- 3. Power Off, then power On the instrument to apply the change.
 - Instrument profiles and files stored on the instrument are deleted, including all EDS files and any custom dye calibrations.
 - The most recent valid ROI/Uniformity, Background, and system dye calibrations are not deleted.

After restoring factory defaults, perform initial start-up tasks. See "Initial start-up" on page 75.

Configure the network

IMPORTANT! Configure the instrument for either wired connection or wireless connection but not both.

- 器 "Set up a wired connection" on page 87

Set up a wired connection

- 1. In the home screen, touch (a) Settings > Instrument Settings > Network Connection > & Wired.
- 2. In the **Network Configuration** screen, touch a radio button to connect to a network either by **DHCP** or by a **Static IP** address.

Option	Action
Automatic	Touch DHCP.
Manual	 Touch Static IP. Enter the IP address for the instrument, the Subnet Mask, the Default Gateway, and <i>(optional)</i> the Primary DNS Server and the Secondary DNS Server. Addresses are in the form of X.X.X.X, where each X is a number from 0 to 255.

6



Note: Ask your system administrator if the IP address is assigned statically or dynamically. For static addresses, you need to know the **IP address** for the instrument, the **Subnet Mask**, and the **Default Gateway**.

3. Touch OK.

Set up a wireless connection

- 2. Select a network or touch **Join other network**, enter the network password, then touch **Enter**. If you select **Join other network**, enter the network name and security type.
- 3. Touch Join to continue.
- 4. In the Network Connection Complete screen, touch Next.
- 5. Touch OK.
- 6. In the Network Configuration screen, edit the Network details or touch Done.

Select a region for Thermo Fisher[™] Connect (Administrator only)

During the initial startup, you will be automatically prompted to select a region for Thermo Fisher[™] Connect. Return to the selection through (a) **Settings**, if necessary.

- 1. In the home screen, touch (a) Settings > Instrument Settings > Thermo Fisher Connect Region.
- 2. Touch a region.

The server used to store your data is located in this region.

3. Touch OK.

Manage the instrument name (Administrator only)

- 1. In the home screen, touch (a) Settings > Instrument Settings > Instrument Name.
- 2. Touch the **Instrument Name** field, enter an instrument name, then touch **Done**.
- 3. Touch OK.

Set the date and time (Administrator only)

- 1. In the home screen, touch (*) Settings > Instrument Settings > Date/Time.
- 2. Select a time zone from the dropdown list.
- 3. Select a date format.
 - a. Touch Date Format, then select the preferred date format.
 - **b.** Touch **Next**, touch the date field, then enter the date.
 - c. Touch Enter, then touch Done.
- 4. Select a time format.
 - a. Touch Time Format.
 - **b.** Slide the control to select a 12-hour or 24-hour clock.
 - c. Touch Next, touch the time field, then enter the time.
 - d. Touch Enter, then touch Done.
- 5. Touch Done.

Manage the Sign Out Timer (Administrator only)

- 1. In the home screen, touch (a) Settings > Manage Users > Sign Out Timer.
- 2. Touch the **Edit Time** field, then enter the desired duration of inactivity before automatic user sign out.
- 3. Touch Enter, then touch Done.



Install and connect the instrument to a network

Workflow: Install and connect to a network	90
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Workflow: Install and connect to a network

Perform all steps in "Before you begin" on page 91

▼

"Unpack and install the instrument" on page 91

▼

"Power on and follow the startup wizard" on page 92

▼

"Perform instrument verification using RNase P plates" on page 53

▼

"Connect the computer to the instrument directly or to a LAN" on page 92

Before you begin

This section provides instructions for customer installation of the computer and the instrument. For installation by a field service engineer (FSE), contact Support to order a service call.

Before starting the installation, complete the following tasks:

- Determine the configuration for your instrument and obtain the network information you need (see "Networking" on page 95).
- Review site requirements (see *QuantStudio*[™] 3 and 5 Real-Time PCR Systems Site Preparation Guide (Pub. No. MAN0010405).
- Review instrument and computer connections (see "Instrument and computer connections" on page 93).

Unpack and install the instrument

- 1. Prepare the installation site as described in the *QuantStudio*[™] 3 and 5 Real-Time PCR Systems Site Preparation Guide (Pub. No. MAN0010405).
- 2. Follow the pre-printed instructions on the instrument box to unpack the instrument, accessories, and reference documentation. Save the packing material for future use or recycle it.

The instrument box contains:

- Reference documentation: Welcome note, unpacking and set up instructions card, system documentation insert
- One instrument
- Accessories: power cable, Ethernet cable, USB drive, reaction tube retainer
- Shipping plate

Note: Save the shipping plate but do not use it to operate the instrument.

If you ordered the wireless adapter, it is provided separately.

- 3. Place the instrument on the bench.
- 4. Plug the power cable into the power port on the back panel of the instrument, then plug the cable into an electrical receptacle.
- Connect your instrument as required by your network configuration (see "Supported options for instrument and computer connections" on page 95 and "Instrument and computer connections" on page 93).
 - Connect an Ethernet cable to the Ethernet port on the back panel of the instrument, then connect the cable to a computer or a networking port.

or

• Connect wirelessly via the wireless adapter.



Note: Do not connect the High Power USB WiFi Module (Cat. No. A26774) to the instrument if it is connected to a network by an Ethernet cable. Configuring the instrument for both wired and wireless connection can interfere with instrument operation.

Power on and follow the startup wizard

- **1.** Power on the instrument.
- 2. Follow the startup wizard through the following tasks:
 - Select the language for the instrument.
 - Accept the license agreement.
 - Select a **Wired** or **Wireless** networking option, as needed for your configuration (see "Supported options for instrument and computer connections" on page 95).
 - Configure the instrument date and time.
 - Create an administrator instrument profile.

Note: You can perform any of the steps above at a later time if you do not have the information needed to complete the startup screens. See "Configure instrument settings" on page 26.

IMPORTANT! Before using the instrument for the first time, we recommend that you perform instrument verification using RNase P plates (see "Perform instrument verification using RNase P plates" on page 53).

Connect the computer to the instrument directly or to a LAN

This section describes the direct wired connection of the computer to the instrument or to a LAN.

- 1. Connect an Ethernet cable from the instrument or a LAN to the computer.
- 2. Power on the computer, then log in using a Windows[™] Administrator account.
- 3. Open the Control Panel, then open the Network and Sharing Center.
- 4. In the left pane of the Network and Sharing Center window, click Change Adapter Settings.
- 5. Right-click on Broadband Connection, then select Properties.
- 6. In the Networking tab, select Internet Protocol Version 4 (TCP/IPv4).
- 7. Click Properties.

- 8. In the Internet Protocol Version 4 (TCP/IPv4) Properties dialog box, select one of the following options.
 - Obtain an IP address automatically
 - Use the following IP address
- 9. If Use the following IP address was selected, enter the IP address.
- 10. Close all dialog boxes by clicking **OK**, then re-start the computer.

Instrument and computer connections

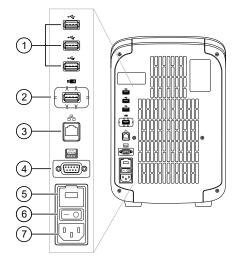


Figure 3 Instrument back panel

- 1 USB ports
- (2) WiFi USB port-Connect USB wireless adapter for wireless network access (ordered separately)
- ③ Ethernet Port—RJ45 port for 100/1,000 Mbps Ethernet communication with the instrument
- (4) RS232 Port-For service use only
- 5 Fuse Cover
- 6 Power Switch
- 7 Power Port-100 to 240 VAC





Appendix A Install and connect the instrument to a network Download and install the desktop software

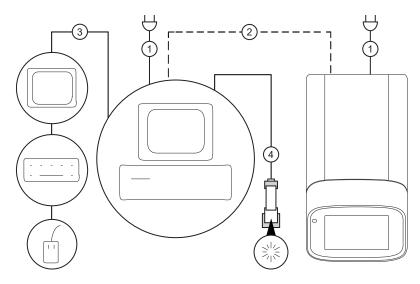


Figure 4 Instrument-to-computer connections (barcode scanner connected to the computer)

Minitower configuration

- (1) Detachable power supply cord compatible with local power supply receptacle.
- (2) Connection between the computer and the instrument.
- ③ Connection between the computer and the monitor, keyboard, and mouse.
- (4) Connection between the computer and the *(optional)* handheld barcode scanner.

Download and install the desktop software

Computer requirements for the desktop software

If you purchased a computer provided by Thermo Fisher Scientific, you can install the desktop software and use it to control the instrument.

This instrument uses QuantStudio[™] Design and Analysis Software v1.5.x. QuantStudio[™] Design and Analysis Software v2.x can also be used to setup and analyze files.

Thermo Fisher Scientific does not support the use of customer-provided computers to control the instrument.

However, if desired, you can install the desktop software on a customer-provided computer and use the software to create templates and analyze data. Minimum requirements for a customer-provided computer are:

- Operating system Windows[™] 10 (64-bit)
- Processor Pentium[®] 4 or comparable
- Memory-4 GB RAM
- Hard drive 500 GB
- Monitor 1280 × 1024 resolution

Download the desktop software

1. Sign in to your account at thermofisher.com.

Note: If you do not have an account, create an account.

- 2. Go to thermofisher.com/quantstudio3-5softwaredownloads.
- 3. Download the software and the example files.

Install the desktop software

- 1. Use a Windows[™] Administrator account to log in to the computer on which you are installing the desktop software.
- 2. Unzip the downloaded software and example files.
- 3. Double-click setup.exe.
- 4. Follow the InstallShield Wizard prompts to install the software.
- 5. Accept the License Agreement.
- 6. Select Typical as the setup preference, then click Next.
- 7. If you are installing the software on a computer provided by Thermo Fisher Scientific, install the software on the D:\ drive. If you are installing on a customer-provided computer, install the software in your preferred location.
- 8. Click Finish.

Networking

IMPORTANT! This section provides general networking information. It *does not* provide adequate detail to integrate the instrument into all possible network architectures. Because a network may contain advanced features (such as a firewall or network domains), we recommend that you consult a network administrator before connecting the instrument to your laboratory network.

Supported options for instrument and computer connections

We support the following direct, networked (LAN–local area network), or Thermo Fisher[™] Connect configurations. Configurations other than those listed are not recommended. Select a configuration that meets the needs of your laboratory's instrument, software, and workflow requirements.

IMPORTANT! Do *not* connect *both* an Ethernet cable and the Wi-Fi module (Cat. No. A26774) to the instrument. Configuring the instrument for both wired and wireless connection can interfere with instrument operation.



Note: For detailed information about networking your instrument, see the *QuantStudio*[™] 3 and 5 *Real-Time PCR Systems IT Checklist* (Pub. No. MAN0013738).

Direct configuration option

- A computer provided by Thermo Fisher Scientific with the QuantStudio[™] Design and Analysis Software
- Computer-to-instrument connection:
 - Direct, wired connection between the computer and the instrument using an Ethernet cable

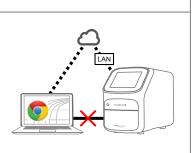
IMPORTANT! A direct instrument-to-computer connection *cannot* be combined with the LAN or the Thermo Fisher[™] Connect configurations.

Networked configuration option

- A computer provided by Thermo Fisher Scientific with the QuantStudio[™] Design and Analysis Software
- The computer and instrument must have the same subnet mask within the same network.
- Computer-to-LAN connection (select an option):
 - Wired connection to the network using an Ethernet cable or -
 - Wireless connection to the network
- Instrument-to-LAN connection (select an option):
 - Wired connection to the network using an Ethernet cable or -
 - Wireless connection to the network using the instrument Wi-Fi module

Thermo Fisher[™] Connect configuration option

- Internet access and a computer with the Chrome[™] web browser to access Thermo Fisher[™] Connect
- Computer-to-Thermo Fisher[™] Connect connection (select an option):
 - Wired connection to the internet using an Ethernet cable or -
 - Wireless connection to the internet
- Instrument-to-Thermo Fisher[™] Connect connection (select an option):
 - Wired connection to the network using an Ethernet cable -or -
 - Wireless connection to the network using the instrument Wi-Fi module





Control and monitor networked instruments

When the instrument is connected to a network:

- Computers on the network that are running the desktop software can control the instrument. Networked instruments can be controlled by only one computer at a time.
- Instruments linked to Thermo Fisher[™] Connect cannot be controlled remotely. However, you can perform the following functions:
 - Remotely access Thermo Fisher[™] Connect to create (and analyze) experiments.
 - From the instrument, download the experiments and start a run.
 - Monitor a run from Thermo Fisher[™] Connect in real time.

Ethernet port overview

The Ethernet port of the instrument supports:

- Static IP network service with subnet mask, primary and secondary data network service (DNS), and default gateway settings, or dynamic host configuration protocol (DHCP) network service.
- mDNS/DNS for local domains.

Note: Because mDNS is limited to direct network connections, an instrument configured for mDNS may not be visible to other nodes that are separated by a router, hub, or another network device.

 IPv4 linknlocal (IPV4LL) in the RFC (also known as Automatic Private IP Addressing [APIPA] or Internet Protocol Automatic Configuration [IPAC]).

Note: When an instrument is set for DHCP, APIPA is automatically enabled, and the instrument provides an IP address when no address is supplied by the DHCP server.

Firewall ports that must be open

Ports	Condition
80/443	Standard ports for instrument-to-Thermo Fisher [™] Connect and computer-to- Thermo Fisher [™] Connect connections
mDNS, 7443	Instrument-to-computer connection
mDNS, 5353	Instrument discovery



Appendix A Install and connect the instrument to a network *Networking*

Networking guidelines and best practices

- Consult a network administrator before connecting the instrument to a network.
- To enable the full functionality of the software, the computer requires a network connection.
- Open the firewall port for the instruments to be discovered. See "Firewall ports that must be open" on page 97.
- Observe the restrictions to mDNS and Autodiscovery.

The instrument supports mDNS but only when the instrument and computer share a direct network connection and are within the same subnet. Network computers that are separated from the instrument by a router, hub, or another network device may not be able to access the instrument by its host name.

- Confirm the uniqueness of the instrument name.
 - The instrument name must be unique within the subnet. The desktop software can automatically discover instruments on the link-local network.
 - The instrument does not test the uniqueness of the instrument name within the subnet when it is set.



Troubleshooting

Observation	Possible cause	Recommended action
Inconsistent communication between instrument and computer or instrument and Thermo Fisher [™] Connect	The instrument is configured for <i>both</i> wired and wireless network connection.	Ensure only one connectivity option is plugged into the instrument (either an Ethernet cable or a wireless adapter, but not both).
Thermo Fisher Connect		Configure for wired <i>or</i> wireless network connection.
	Weak or unstable internet connection, especially if	Change the configuration to a wired connection.
	configured for wireless.	Use a wireless network with a stronger or more consistent signal.
The connection between the instrument and the computer is	The connection is not fully established.	Power the instrument off, then power it on again.
not recognized	If using a networked configuration, the instrument and computer are not on the same subnet mask.	Contact your information technologies department to have them ensure that the instrument and computer are on the same subnet mask.
	If using a networked configuration, the instrument or computer has an invalid IP address.	Contact your information technologies department to have them ensure that the IP addresses are valid.
Insufficient disk space message	Insufficient disk space to save a run.	 In the home screen, touch Settings ▶ Run History ▶ Manage.
		 Delete or transfer experiments from the instrument.
The touchscreen is black	The instrument is in sleep mode.	Touch anywhere on the instrument touchscreen.
	The instrument is not powered on.	If you touch the instrument touchscreen and it remains black, check if the instrument is powered on. The power switch is located on the rear panel of the instrument.
		If the instrument does not power on, check that the instrument is properly plugged in.
		If the instrument does not power on and the instrument is properly plugged in, contact Support.
Forgot PIN for instrument profile	Non-administrator forgot instrument profile PIN.	See "Manage all instrument profiles (Administrator only)" on page 76.



Observation	Possible cause	Recommended action
Forgot PIN for instrument profile (continued)	Administrator forgot instrument profile PIN.	Have another administrator reset the PIN for the forgotten-PIN profile (see "Manage all instrument profiles (Administrator only)" on page 76).
		If there is not another administrator profile on the instrument, you must restore factory defaults (see "Restore factory defaults" on page 87).



Parts and materials

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384-well consumables	102
Consumables (96-well, 0.1-mL format)	103
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General-use materials and consumables	104

Kits, consumables, accessories, and reagents

Unless otherwise indicated, all materials are available through thermofisher.com.

Store all calibration and RNase P plates at –20°C. All other items can be stored at 15–30°C. Use all materials by the expiration date on the packaging.

Catalog numbers that appear as links open the web pages for those products.

Consumables (96-well, 0.2-mL format)

Consumable	Amount	Cat. No.
MicroAmp [™] Optical 8-Cap Strips	300 strips	4323032
MicroAmp [™] Optical 8-Tube Strip, 0.2 mL	125 strips	4316567
MicroAmp [™] Optical Tube without Cap, 0.2 mL	2,000 tubes	N8010933
MicroAmp [™] 96-Well Tray/Retainer Set for Veriti [™] Systems	10 trays	4381850
Miere Amm TM Optical OC Wall Departies Plate	10 plates	N8010560
MicroAmp [™] Optical 96-Well Reaction Plate	500 plates	4316813
Miere Amp [™] Optical OC Wall Departies Dista with Deveeds	20 plates	4306737
MicroAmp [™] Optical 96-Well Reaction Plate with Barcode	500 plates	4326659
MicroAmp [™] EnduraPlate [™] Optical 96-Well Clear Reaction Plates with	20 plates	4483354
Barcode	500 plates	4483352
MicroAmp [™] EnduraPlate [™] Optical 96-Well Clear GPLE Reaction Plates with Barcode	20 plates	4483348



(continued)

Consumable	Amount	Cat. No.
MicroAmp [™] EnduraPlate [™] Optical 96-Well Clear GPLE Reaction Plates with Barcode	500 plates	4483351
MicroAmp [™] EnduraPlate [™] Optical 96-Well Fast Clear Reaction Plates with Barcode	20 plates	4483485
MicroAmp [™] EnduraPlate [™] Optical 384-Well Clear Reaction Plates with Barcode	20 plates	4483285
MicroAmp [™] Optical Adhesive Film Kit	1 kit	4313663
MicroAmp [™] Optical Film Compression Pad (Optional, used for some applications)	5 each	4312639

Instrument verification or calibration plate	Cat. No.
TaqMan [™] RNase P Instrument Verification Plate, 96-Well 0.2-mL	4432382
Region of Interest (ROI) and Background Plates, 96-Well 0.2-mL (2 plates)	4432364
QuantStudio [™] 3/5 10-Dye Spectral Calibration Kit, 96-Well 0.2-mL (contains all 3 spectral calibration plates listed below)	A26343
QuantStudio [™] 3/5 Spectral Calibration Plate 1 (FAM [™] , VIC [™] , ROX [™] , and SYBR [™] dyes), 96-Well 0.2-mL	A26331
QuantStudio [™] 3/5 Spectral Calibration Plate 2, 96-Well 0.2-mL (ABY [™] , JUN [™] , and MUSTANG PURPLE [™] dyes)	A26332
QuantStudio [™] 3/5 Spectral Calibration Plate 3, 96-Well 0.2-mL (TAMRA [™] , NED [™] , and Cy [®] 5 dyes)	A26333

384-well consumables

Consumable	Contents	Cat. no.
MicroAmp [™] Optical 384-Well Reaction Plate with Barcode	50 plates	4309849
	500 plates	4326270
	1000 plates	4343814
MicroAmp [™] EnduraPlate [™] Optical 384-Well Reaction Plate with	20 plates	4483285
Barcode (clear)	500 plates	4483273
MicroAmp [™] Optical Adhesive Film Kit	1 kit	4313663

Calibration or instrument verification plate	Cat. no.
384-Well Region of Interest (ROI) and Background Plates (2 plates)	4432320
QuantStudio [™] 3 or 5 10-Dye Spectral Calibration Kit, 384-Well	A26341
384-Well Spectral Calibration Plate 1 (containing FAM [™] , VIC [™] , ROX [™] , TAMRA [™] , and SYBR [™] dyes)	A26334
384-Well Spectral Calibration Plate 2 (containing ABY [™] , JUN [™] , MUSTANG PURPLE [™] , NED [™] , and Cy [®] 5 dyes)	A26335
384-Well TaqMan [™] RNase P Instrument Verification Plate	4455280

Consumables (96-well, 0.1-mL format)

Consumable	Contents	Cat. no.
MicroAmp [™] Optical 8-Cap Strip	300 strips	4323032
MicroAmp [™] Optical Fast 8-Tube Strip (0.1 mL)	125 strips	4358293
MicroAmp [™] Optical Fast Tube with Cap (0.1 mL)	1000 tubes	4358297
MicroAmp [™] 96-Well Tray for VeriFlex [™] Block	10 trays	4379983
MicroAmp [™] Optical 96-Well Fast Reaction Plate (0.1 mL)	10 plates	4346907
MicroAmp [™] EnduraPlate [™] Optical 96-Well Fast Reaction Plate with	20 plates	4481194
Barcode (clear) (0.1 mL)	500 plates	4483494
MicroAmp [™] Optical Adhesive Film Kit	1 kit	4313663

Calibration or instrument verification plate	Cat. no.
96-Well Fast Region of Interest (ROI) and Background Plates (2 plates)	4432426
QuantStudio [™] 3 or 5 10-Dye Spectral Calibration Kit, 96-Well 0.1-mL	A26342
96-Well 0.1-mL Spectral Calibration Plate 1 (containing FAM [™] , VIC [™] , ROX [™] , and SYBR [™] dyes	A26336
96-Well 0.1-mL Spectral Calibration Plate 2 (containing ABY [™] , JUN [™] , and MUSTANG PURPLE [™] dyes)	A26337
96-Well 0.1-mL Spectral Calibration Plate 3 (containing TAMRA [™] , NED [™] , and Cy [®] 5 dyes)	A26340
96Well Fast TaqMan [™] RNase P Instrument Verification Plate	4351979

QuantStudio[™] 3 and 5 Real-Time PCR Systems Installation, Use, and Maintenance Guide

Accessories

Item	Amount	Cat. No.
MicroAmp [™] Multi Removal Tool	1 tool	4313950
MicroAmp [™] Cap Installing Tool (handle style)	1 tool	4330015
MicroAmp [™] Optical Adhesive Film	25 films	4360954
	100 films	4311971
MicroAmp [™] Adhesive Film Applicator	5 applicators	4333183
RT-PCR Grade Water	10 × 1.5 mL tubes	AM9935
Handheld Barcode Scanner	1 scanner	4488442
High Power USB WiFi Module	1 module	A26774

General-use materials and consumables

The following general-use materials and consumables are required to calibrate, maintain, and operate the instrument.

Unless otherwise indicated, all materials are available through **thermofisher.com**. "MLS" indicates that the material is available from **fisherscientific.com** or another major laboratory supplier.

Material/Consumable	Source
Centrifuge with 96-well plate buckets	MLS
Cotton or nylon swabs and lint-free cloths	MLS
Ethanol, 95% solution	MLS
Optical clear adhesive film for PCR	MLS
Na-hypochlorite, 0.1% v/v solution	
Pipettors, 100-μL and 200-μL (with pipette tips)	MLS
Powder-free gloves	MLS
Safety glasses	MLS
Screwdriver, flathead	MLS
Tissue, lint-free	MLS
Deionized water	MLS



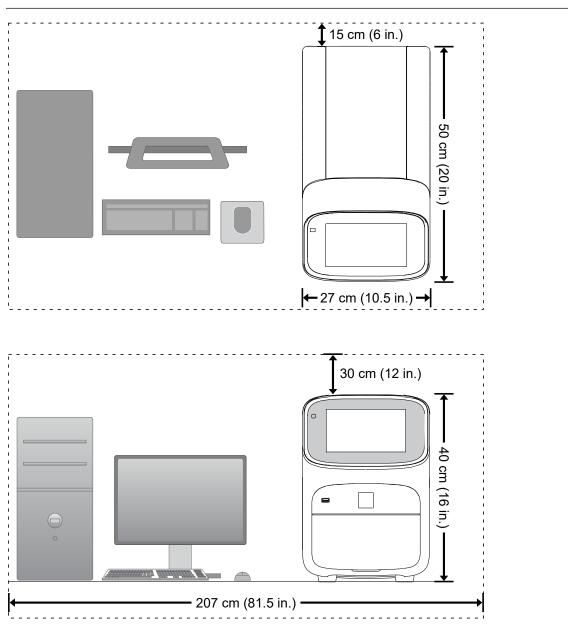
Instrument specification and layout

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Configured system dimensions

Allow space for the configured instrument. A typical setup with a co-located minitower computer is shown below.





Instrument clearances

During instrument installation and maintenance, it is necessary to access the back of the instrument. If the back of the instrument faces a wall, ensure that there is sufficient clearance on the bench to rotate the instrument for access.

IMPORTANT! For safety, the power outlet for the instrument must be accessible.

Component	Тор	Front	Sides	Back
Instrument	30 cm (12 in.)	30 cm (12 in.)	15 cm (6 in.)	15 cm (6 in.)
Computer ^[1]	_	15 cm (6 in.)	_	15 cm (6 in.)

^[1] Co-locating the computer with the instrument is optional.

Electrical requirements



WARNING! For safety, the power outlet used for powering the instrument must be accessible at all times. See "Instrument clearances" on page 107 for information about the space needed between the wall and the instrument. In case of emergency, you must be able to immediately disconnect the main power supply to all the equipment. Allow adequate space between the wall and the equipment so that the power cords can be disconnected in case of emergency.



WARNING! Use of accessories, transducers, and cables other than those specified or provided by the manufacturer of the equipment could result in increased electromagnetic emissions or decreased electromagnetic immunity of this equipment and result in improper operation.

- Electric receptacle with grounding capability
- Maximum power dissipation: ~960 W (not including computer and monitor)
- Mains AC line voltage tolerances must be up to ±10 percent of nominal voltage

Device	Rated voltage	Circuit required	Rated frequency	Rated power
Instrument	100–240 ±10% VAC ^[1]	10 A	50/60 Hz	960 W
Computer (laptop)	100–240 ±10% VAC	10 A	50/60 Hz	90 VA
Computer (desktop)	100–240 ±10% VAC	10.4	50/60 Hz	125 VA
Monitor	100-240 ±10% VAC	10 A		65 VA

^[1] If the supplied power fluctuates beyond the rated voltage, a power line regulator may be required. High or low voltages can adversely affect the electronic components of the instrument.



Environmental requirements

Table 2 Environmental requirements

Condition	Acceptable range
Installation site	Indoor use only
Electromagnetic interference	Do not use this device in close proximity to sources of strong electromagnetic radiation (for example, unshielded intentional RF sources). Strong electromagnetic radiation may interfere with the proper operation of the device.
Altitude	Between sea level and 2000 m (6500 ft) above sea level
Operating conditions	 Humidity: 15–80% relative humidity (noncondensing) Temperature: 15°C to 30°C (59°F to 86°F) Note: For optimal performance, avoid rapid or extreme fluctuations in room
	temperature.
Storage and transport	Humidity: 20–80% relative humidity (noncondensing)
conditions	 Temperature: -30°C to 60°C (-22°F to 140°F)
Thermal output	During operation, the net thermal output, based on the actual current draw of the instrument, is expected to be approximately 960 W (3275 Btu/h).
Vibration	Ensure that the instrument is not adjacent to strong vibration sources, such as a centrifuge, pump, or compressor. Excessive vibration will affect instrument performance.
Pollution degree	The instrument has a Pollution Degree rating of II. The instrument may only be installed in an environment that has nonconductive pollutants such as dust particles or wood chips. Typical environments with a Pollution Degree II rating are laboratories and sales and commercial areas.
	The noise output of the instrument is \leq 60 dB when running.
Other conditions	Ensure the instrument is located away from any vents that could expel particulate material onto the instrument components.
	Avoid placing the instrument and computer adjacent to heaters, cooling ducts, or in direct sunlight.

Network requirements

The instrument:

- Is factory-configured for IPv4 TCP/IP communication and includes an Ethernet adapter (100/1,000 Mbps) with an RJ45-type connector for integrating the device into a local area network (LAN).
- Can alternatively be configured for wireless networking (High Power USB WiFi Module required, sold separately as an optional accessory).

The instrument can be configured for either wired or wireless networking, not both.

If a Thermo Fisher Scientific service representative is to install the instrument:

- If the instrument will be connected to a LAN, an active, tested network jack must be in place before the scheduled installation date.
- A representative from your information technologies department must be available during the installation to help connect the instrument to your network.

Required materials to network the instrument:

- Wired-Ethernet cable of sufficient length with RJ45 connectors
 - CAT5 cable for a 100 Mbps network connection
 - CAT5e or CAT6 cable for a 1,000 Mbps network connection

-or-

• Wireless—High Power USB WiFi Module (Cat. No. A26774, sold separately)



Safety

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WARNING! GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
- Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, and so on). To obtain SDSs, see the "Documentation and Support" section in this document.

Symbols on this instrument

Symbols may be found on the instrument to warn against potential hazards or convey important safety information. In this document, the hazard symbol is used along with one of the following user attention words:

- **CAUTION!**—Indicates a potentially hazardous situation that, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices.
- **WARNING!**—Indicates a potentially hazardous situation that, if not avoided, could result in death or serious injury.
- **DANGER!**—Indicates an imminently hazardous situation that, if not avoided, will result in death or serious injury.

Symbol	English	Français
Â	Caution, risk of danger Consult the manual for further safety information.	Attention, risque de danger Consulter le manuel pour d'autres renseignements de sécurité.

(continued)

Symbol	English	Français
<u>A</u>	Caution, risk of electrical shock	Attention, risque de choc électrique
	Moving parts	Parties mobiles
	Caution, hot surface	Attention, surface chaude
Ś	Potential biohazard	Danger biologique potentiel
	Ultraviolet light	Rayonnement ultraviolet
×	Potential slipping hazard	Danger de glisser potentiel
	On	On (marche)
\bigcirc	Off	Off (arrêt)
\bigcirc	On/Off	On/Off (marche/arrêt)
Ċ	Standby	En attente
	Earth (ground) terminal	Borne de (mise à la) terre
	Protective conductor terminal (main ground)	Borne de conducteur de protection (mise à la terre principale)
\sim	Terminal that can receive or supply alternating current or voltage	Borne pouvant recevoir ou envoyer une tension ou un courant de type alternatif
\sim	Terminal that can receive or supply alternating or direct current or voltage	Borne pouvant recevoir ou envoyer une tension ou un courant continu ou alternatif

(continued)

Symbol	English	Français		
X	Do not dispose of this product in unsorted municipal waste	Ne pas éliminer ce produit avec les déchets usuels non soumis au tri sélectif.		
	★ CAUTION! To minimize negative environmental impact from disposal of electronic waste, do not dispose of electronic waste in unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provision and contact customer service for information about responsible disposal options.	MISE EN GARDE ! Pour minimi- ser les conséquences négatives sur l'environnement à la suite de l'éli- mination de déchets électroniques, ne pas éliminer ce déchet électro- nique avec les déchets usuels non soumis au tri sélectif. Se conformer aux ordonnances locales sur les dé- chets municipaux pour les disposi- tions d'élimination et communiquer avec le service à la clientèle pour des renseignements sur les options d'élimination responsable.		

Conformity symbols

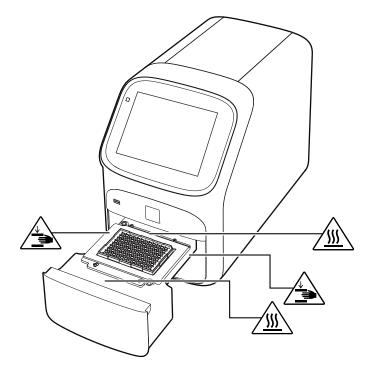
Conformity mark	Description
C	Indicates conformity with safety requirements for Canada and U.S.A.
CE	Indicates conformity with European Union requirements for safety and electromagnetic compatibility.
	Indicates conformity with Australian standards for electromagnetic compatibility.

Safety alerts on this instrument

Additional text may be used with one of the symbols described above when more specific information is needed to avoid exposure to a hazard. See the following table for safety alerts found on the instrument.

English	Français		
CAUTION! Hazardous chemicals. Read the Safety Data Sheets (SDSs) before handling.		MISE EN GARDE ! Produits chimiques dangereux. Lire les fiches signalétiques (FS) avant de manipuler les produits.	
CAUTION! Hazardous waste. Refer to SDS(s) and local regulations for handling and disposal.		MISE EN GARDE ! Déchets dangereux. Lire les fiches signalétiques (FS) et la régle- mentation locale associées à la manipulation et à l'élimination des déchets.	

Location of safety labels on the instrument



Safety information for instruments not manufactured by Thermo Fisher Scientific

Some of the accessories provided as part of the instrument system are not designed or built by Thermo Fisher Scientific. Consult the manufacturer's documentation for the information needed for the safe use of these products.

Instrument safety

General



CAUTION! Do not remove instrument protective covers. If you remove the protective instrument panels or disable interlock devices, you may be exposed to serious hazards including, but not limited to, severe electrical shock, laser exposure, crushing, or chemical exposure.



CAUTION! Solvents and Pressurized fluids. Wear eye protection when working with any pressurized fluids. Use caution when working with any polymeric tubing that is under pressure:

- Extinguish any nearby flames if you use flammable solvents.
- Do not use polymeric tubing that has been severely stressed or kinked.
- Do not use polymeric tubing with tetrahydrofuran or nitric and sulfuric acids.
- Be aware that methylene chloride and dimethyl sulfoxide cause polymeric tubing to swell and greatly reduce the rupture pressure of the tubing.
- Be aware that high solvent flow rates (~40mL/min) may cause a static charge to build up on the surface of the tubing and electrical sparks may result.

Physical injury



CAUTION! Moving and Lifting Injury. The instrument is to be moved and positioned only by the personnel or vendor specified in the applicable site preparation guide. Improper lifting can cause painful and permanent back injury.

Things to consider before lifting or moving the instrument or accessories:

- Depending on the weight, moving or lifting may require two or more persons.
- If you decide to lift or move the instrument after it has been installed, do not attempt to do so without the assistance of others, the use of appropriate moving equipment, and proper lifting techniques.
- Ensure you have a secure, comfortable grip on the instrument or accessory.
- Make sure that the path from where the object is to where it is being moved is clear of obstructions.
- Do not lift an object and twist your torso at the same time. Keep your spine in a good neutral position while lifting with your legs.
- Participants should coordinate lift and move intentions with each other before lifting and carrying.
- For smaller packages, rather than lifting the object from the packing box, carefully tilt the box on its side and hold it stationary while someone else slides the contents out of the box.



CAUTION! Moving Parts. Moving parts can crush, pinch and cut. Keep hands clear of moving parts while operating the instrument. Disconnect power before servicing.

WARNING! Do not attempt to lift or move the instrument without the assistance of others. Use appropriate moving equipment and proper lifting technique, improper lifting may result in serious injury.



Electrical safety

WARNING! Fuse Installation. Before installing the instrument, verify that the fuses are properly installed and the fuse voltage matches the supply voltage. Replace fuses only with the type and rating specified for the unit. Improper fuses can damage the instrument wiring system and cause a fire.



WARNING! Ensure appropriate electrical supply. For safe operation of the instrument:

- · Plug the system into a properly grounded receptacle with adequate current capacity.
- Ensure the electrical supply is of suitable voltage.
- Never operate the instrument with the ground disconnected. Grounding continuity is required for safe operation of the instrument.



WARNING! Power Supply Line Cords. Use properly configured and approved line cords for the power supply in your facility.



WARNING! Disconnecting Power. To fully disconnect power either detach or unplug the power cord, positioning the instrument such that the power cord is accessible.

Cleaning and decontamination



CAUTION! Cleaning and Decontamination. Use only the cleaning and decontamination methods specified in the manufacturer's user documentation. It is the responsibility of the operator (or other responsible person) to ensure the following requirements are met:

- No decontamination or cleaning agents are used that could cause a HAZARD as a result of a reaction with parts of the equipment or with material contained in the equipment.
- The instrument is properly decontaminated a) if hazardous material is spilled onto or into the equipment, and/or b) prior to having the instrument serviced at your facility or sending the instrument for repair, maintenance, trade-in, disposal, or termination of a loan (decontamination forms may be requested from customer service).
- Before using any cleaning or decontamination methods (except those recommended by the manufacturer), users should confirm with the manufacturer that the proposed method will not damage the equipment.

Safety and electromagnetic compatibility (EMC) standards

The instrument design and manufacture complies with the following standards and requirements for safety and electromagnetic compatibility.

Safety compliance

Reference	Description
EU Directive 2014/35/EU	European Union "Low Voltage Directive"
IEC 61010-1 EN 61010-1 UL 61010-1 CSA C22.2 No. 61010-1	Safety requirements for electrical equipment for measurement, control, and laboratory use – Part 1: General requirements
IEC 61010-2-010 EN 61010-2-010	Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials
IEC 61010-2-081 EN 61010-2-081	Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes

EMC

Reference	Description
Directive 2014/30/EU	European Union "EMC Directive"
EN 61326-1/ IEC 61326-1	Electrical Equipment for Measurement, Control and Laboratory Use – EMC Requirements – Part 1: General Requirements
AS/NZS CISPR 11	Limits and Methods of Measurement of Electromagnetic Disturbance Characteristics of Industrial, Scientific, and Medical (ISM) Radiofrequency Equipment
ICES-001, Issue 4	Industrial, Scientific and Medical (ISM) Radio Frequency Generators
FCC Part 15 Subpart B (47 CFR)	U.S. Standard Radio Frequency Devices

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

QuantStudio[™] 3 and 5 Real-Time PCR Systems Installation, Use, and Maintenance Guide



Environmental design

Reference	Description
Directive 2012/19/EU	European Union "WEEE Directive" - Waste electrical and electronic equipment
EU Directive 2011/65/EU & Commission Delegated Directive (EU) 2015/863	European Union "RoHS Directive" – Restriction of hazardous substances in electrical and electronic equipment
Directive 2006/66/EC	European Union "Battery Directive"

China EEP Hazardous Substances Information						
Component	QuantStudio [™] 3 and 5 Real-Time PCR Systems					
Name			Hazardous	s Substances		
	(Pb)	(Hg)	(Cd)	(Cr(VI))	(PBB)	(PBDE)
PCBA	Х	0	0	0	0	0

SJ/T11364. This table is compiled according to SJ/T 11364 standard.

O: GB/T26572.

Indicates that the concentration of the hazardous substance in all homogeneous materials for the part is below the relevant threshold of the GB/T 26572 standard.

X: GB/T26572.

Indicates that the concentration of the hazardous substance in at least one homogenous material of the part is above the relevant threshold of the GB/T 26572 standard.

Chemical safety



WARNING! GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below. Consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the "Documentation and Support" section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with sufficient ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer cleanup procedures as recommended in the SDS.
- Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if needed) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.



WARNING! HAZARDOUS WASTE (from instruments). Waste produced by the instrument is potentially hazardous. Follow the guidelines noted in the preceding General Chemical Handling warning.



WARNING! 4L Reagent and Waste Bottle Safety. Four-liter reagent and waste bottles can crack and leak. Each 4-liter bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position.

Biological hazard safety

WARNING! Potential Biohazard. Depending on the samples used on this instrument, the surface may be considered a biohazard. Use appropriate decontamination methods when working with biohazards.



WARNING! BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Conduct all work in properly equipped facilities with the appropriate safety equipment (for example, physical containment devices). Safety equipment can also include items for personal protection, such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses, or goggles. Individuals should be trained according to applicable regulatory and company/ institution requirements before working with potentially biohazardous materials. Follow all applicable local, state/provincial, and/or national regulations. The following references provide general guidelines when handling biological samples in laboratory environment.

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 6th Edition, HHS Publication No. (CDC) 300859, Revised June 2020 https://www.cdc.gov/labs/pdf/CDC-BiosafetymicrobiologicalBiomedicalLaboratories-2020-P.pdf
- Laboratory biosafety manual, fourth edition. Geneva: World Health Organization; 2020 (Laboratory biosafety manual, fourth edition and associated monographs)
 www.who.int/publications/i/item/9789240011311



Documentation and support

Related documentation

Document	Publication number	Description
QuantStudio [™] 3 and 5 Real-Time PCR Systems Installation, Use, and Maintenance Guide	MAN0010408	Describes the hardware and software and provides information on preparing, using, maintaining, and troubleshooting the system.
QuantStudio [™] Real-Time PCR System Help	MAN0010422	Describes the instrument touchscreen and provides procedures for configuration, calibration, and performing a run.
QuantStudio [™] Design and Analysis Desktop Software Command-Line Application Guide	MAN0010409	Describes how to use the command-line software application and provides the procedure to automate the creation of new experiment files and export data from existing files.
QuantStudio [™] Design and Analysis Desktop Software User Guide	MAN0010408	Describes how to design and perform experiments using the software.
QuantStudio [™] Design and Analysis Desktop Software Help	MAN0010415	Describes the software and provides procedures for common tasks.
QuantStudio [™] 5 Real-Time PCR System SAE Admin Console Help	MAN0010417	Describes the SAE Administrator Console and provides procedures for common tasks.
QuantStudio [™] 5 Real-Time PCR System SAE Admin Console User Guide	MAN0010410	Describes how to use the SAE Administrator Console.
QuantStudio [™] Design and Analysis Cloud Software Help	MAN0010414	Describes the Cloud version of the software and provides procedures for common tasks.
QuantStudio [™] 3 and 5 Real-Time PCR Systems Site Preparation Guide	MAN0010405	Explains how to prepare your site to receive and install the system. Intended for personnel who schedule, manage, and perform the tasks required to prepare the site for installation.

Note: For additional documentation, see "Customer and technical support" on page 122.

Obtain information from the Help system

The instrument has a Help system that describes how to use each feature of the touchscreen. Touch (?) on the instrument touchscreen to access the Help system.

Customer and technical support

Visit thermofisher.com/support for the latest service and support information.

- Worldwide contact telephone numbers
- Product support information
 - Product FAQs
 - Software, patches, and updates
 - Training for many applications and instruments
- Order and web support
- Product documentation
 - User guides, manuals, and protocols
 - Certificates of Analysis
 - Safety Data Sheets (SDSs; also known as MSDSs)

Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at www.thermofisher.com/us/en/home/ global/terms-and-conditions.html. If you have any questions, please contact Life Technologies at www.thermofisher.com/support.

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