

QuantStudio™ Absolute Q™ Digital PCR System

INSTALLATION, USE, AND MAINTENANCE

for use with:

QuantStudio™ Absolute Q™ Digital PCR Software v6.3 or later

QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite

Security, Auditing, and E-signature (SAE) v2.2 software or later

Catalog Numbers A52864, A57608, A57609, A57610, A57611, A57612

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Revision H



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

ThermoFisher
S C I E N T I F I C



Revision history: MAN0028562 H (English)

Revision	Date	Description
H	19 January 2026	- QuantStudio™ Absolute Q™ Digital PCR Software was updated to version 6.4, with corresponding updates to the software workflow, instructions, and product information. “Overview of the Absolute Q™ instrument” on page 15, “Install the QuantStudio™ Absolute Q™ Digital PCR System” on page 137, and “Moving the instrument” on page 139 were updated to add the second shipping screw.
G	26 March 2025	“Instruments, software, accessories, and consumables” on page 11 and “Recommended materials not supplied” on page 13 were updated. “DNA preparation guidelines” on page 28 and “RNA preparation guidelines” on page 29 were updated. - QuantStudio™ Absolute Q™ Digital PCR Software was updated to version 6.3.5, with corresponding updates to the software workflow. - QuantStudio™ Absolute Q™ Digital PCR Software download, installation, and update instructions and QuantStudio™ Absolute Q™ Digital PCR Instrument software and firmware update instructions were removed (now available in <i>QuantStudio™ Absolute Q™ Digital PCR Software Installation Guide</i>, see “Related documentation” on page 208). - Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later” was updated to include compatibility with later versions of the software, add a reference to software/firmware compatibility matrix, and include a software download connect specific to the China region. - Appendix G, “Connect Absolute Q™ companion PCs to the network using the bridge PC” was added. - Appendix H, “Data export information package for QuantStudio™ Absolute Q™ Digital PCR Software v6.3 or later.” was added. “Related documentation” on page 208 was updated.
F	15 May 2024	- Multiday hands-free operation information was added (“Product description” on page 10). - Power-cycle instructions were added (Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”, “Add more plates during an AutoRun Work Unit with hotels” on page 148, and “Maintenance” on page 177). - Permissions were updated (“Default permissions and roles” on page 164).
E00	8 March 2024	- The product description was updated (“Product description” on page 10). - The troubleshooting section for AutoRun Suite startations was updated (“Troubleshooting AutoRun Suite implementations” on page 117).
D.0	8 January 2024	- Content about the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite was added. - An instrument indicator status light basic meaning was added (“Instrument indicator status light key” on page 16). - Default SAE permissions and roles were updated (“Default permissions and roles” on page 164).
C.0	6 November 2023	Catalog data for the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite were removed.
B.0	26 September 2023	- Content about the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite was removed. - The instructions to install the desktop software were updated.
A.0	30 June 2023	New record for use of the QuantStudio™ Absolute Q™ Digital PCR System and QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite with v6.3 software.

The information in this guide is subject to change without notice.

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IMPORTANT! Before using this product, read and understand the information in the “Safety” appendix in this document.

Product description

The Applied Biosystems™ QuantStudio™ Absolute Q™ Digital PCR System enables precision quantification of target nucleic acid sequences. Using patented microfluidic array technology, QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates (MAP plates) are loaded with digital PCR (dPCR) reagents, then processed by the QuantStudio™ Absolute Q™ Digital PCR Instrument. Depending on the dPCR assay and protocol, results can be provided in less than 90 minutes. The resulting data are visualized with the QuantStudio™ Absolute Q™ Digital PCR Software.

The Applied Biosystems™ QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite is an integrated system solution that scales precise nucleic acid quantification. With flexible instrument configurations and intelligent automation, it supports multi-day hands-free operation. Combined with the QuantStudio™ Absolute Q™ Digital PCR Software and its tools for multi-plate analysis, the AutoRun Suite increases workflow utilization, efficiency, and throughput.

Instruments, software, accessories, and consumables

The following table describes the products that are required for the installation and operation of the system. Unless otherwise indicated, all materials are available through [thermofisher.com](https://www.thermofisher.com).

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

Item	Cat. No.	Amount
Instrument system		
QuantStudio™ Absolute Q™ Digital PCR System: - QuantStudio™ Absolute Q™ Digital PCR Instrument - Dell™ tower computer with monitor, keyboard, and mouse - QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later	A52864	1 instrument, 1 desktop computer, 1 monitor, and accessories
QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite, single configuration: - QuantStudio™ Absolute Q™ Digital PCR Instrument - Dell™ tower computer with monitor, keyboard, and mouse - QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later - Spinnaker™ Microplate Mover - Momentum™ Workflow Scheduler Software	A57608	1 instrument, 2 desktop computers, 1 monitor, 1 robot, and accessories
QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite, dual configuration: - QuantStudio™ Absolute Q™ Digital PCR Instrument - Dell™ tower computer with monitor, keyboard, and mouse - QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later - Spinnaker™ Microplate Mover - Momentum™ Workflow Scheduler Software	A57609	2 instruments, 3 desktop computers, 1 monitor, 1 robot, and accessories
QuantStudio™ Absolute Q™ AutoRun single configuration upgrade for Absolute Q™: - Dell™ tower computer with monitor, keyboard, and mouse - QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later - Spinnaker™ Microplate Mover - Momentum™ Workflow Scheduler Software	A57610	1 desktop computer, 1 robot, and accessories
QuantStudio™ Absolute Q™ AutoRun dual configuration upgrade for Absolute Q™: - Dell™ tower computer with monitor, keyboard, and mouse - QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later - Spinnaker™ Microplate Mover - Momentum™ Workflow Scheduler Software	A57611	1 desktop computer, 1 robot, and accessories

(continued)

Item	Cat. No.	Amount
QuantStudio™ Absolute Q™ AutoRun single-to-dual configuration upgrade: QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later	A57612	Accessories only
Instrument accessories		
QuantStudio™ Absolute Q™ MAP16 Plate Kit: 12 × QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates 60 × QuantStudio™ Absolute Q™ MAP plate gasket strips 3 mL QuantStudio™ Absolute Q™ Isolation Buffer	A52865	1
QuantStudio™ Absolute Q™ MAP16 Plate Kit and Master Mix: 12 × QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates 60 × QuantStudio™ Absolute Q™ MAP plate gasket strips 3 mL QuantStudio™ Absolute Q™ Isolation Buffer 360 µL Absolute Q™ DNA Digital PCR Master Mix (5X)	A53301	1
QuantStudio™ Absolute Q™ Digital PCR Starter Kit ^[1]	A52732	1
QuantStudio™ Absolute Q™ Nest Accessory for Plate Handler Automation	A57588	1
Reagents		
(Recommended) Absolute Q™ Universal DNA Digital PCR Master Mix (5X) or Absolute Q™ DNA Digital PCR Master Mix (5X)	A72710 A52490	200 reactions
QuantStudio™ Absolute Q™ Isolation Buffer	A52730	1 × 3 mL bottle
Absolute Q™ 1-step RT Digital PCR Master Mix (4X)	A55146	200 reactions

^[1] Required for system installation. See *QuantStudio™ Absolute Q™ Digital PCR Starter Kit User Guide* (Pub No. MAN0025653).

Digital PCR assays

Pre-designed and custom dPCR assays are available for use in dPCR experiments. To obtain the information about the use of pre-designed dPCR assays, see the documentation provided with the assay available at thermofisher.com/dpcr-assays.html or contact your local sales representative.

Use this guide to perform experiments with dPCR assays ordered from Thermo Fisher Scientific or with your custom assay protocols.

Required materials not supplied

Unless otherwise indicated, all materials are available through [thermofisher.com](https://www.thermofisher.com). "MLS" indicates that the material is available from [fisherscientific.com](https://www.fisherscientific.com) or another major laboratory supplier.

Item	Source
Equipment	
Centrifuge, table top	MLS
Adjustable pipettors of various sizes capable of pipetting volumes from 1 µL to 200 µL	MLS
Sterile aerosol barrier (filtered) pipette tips	MLS
Consumables	
Low DNA binding microcentrifuge tubes	MLS
Microcentrifuge tube rack	MLS
Nuclease-free Water	MLS
Non-abrasive, lint-free wipe	MLS
70% ethanol solution in water	MLS

Recommended materials not supplied

Unless otherwise indicated, all materials are available through [thermofisher.com](https://www.thermofisher.com). "MLS" indicates that the material is available from [fisherscientific.com](https://www.fisherscientific.com) or another major laboratory supplier.

Item	Source
Nucleic acid isolation kit	thermofisher.com/magmax or MLS
Spectrophotometer	MLS
Qubit™ Flex Fluorometer	Q33327
KingFisher™ Apex with 96 Deep Well Head	5400930
KingFisher™ Apex with 24 Combi head	5400940
QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite custom table (2 tables and 1 racking system for D-Link and KVM switches)	A57613
PC running Windows™ 10 or 11 (if using the bridge PC to connect one or more Absolute Q™ companion PCs to the network) ^[1]	—

^[1] See Appendix G, "Connect Absolute Q™ companion PCs to the network using the bridge PC".

Software description

The QuantStudio™ Absolute Q™ Digital PCR System and QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite use the following software.

- QuantStudio™ Absolute Q™ Digital PCR Software v6.4 or later—Controls the instrument, performs user-defined experiments, analyzes data generated by the experiment. Parameters such as plate format, optical channels, and thermal conditions for an experiment can be modified as needed before generating data.

You can use the software to perform the following tasks:

- Define the experiment, including sample types, sample groups, replicates, pool sample, dilutions, threshold parameters, experiment notes, and names
- Create run templates and batches to support the AutoRun Suite
- Create and edit protocols
- Run and monitor protocols
- View system status
- View data in plot and tables
- Analyze multiple runs simultaneously using studies
- Generate run reports
- Export data and reports
- Insert and remove MAP plates
- Install the shipping lock screws for transport of the instrument
- Download instrument logs for system troubleshooting
- Security, Auditing, and E-signature (SAE) v2.2 or later software (*Optional*)—Controls security and user access to the software and specific features. For more information, see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”.
- Momentum™ Workflow Scheduler Software—Part of the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite that enables users to define, execute, and monitor scientific processes and workflows in an easy-to-use visual environment. For more information, see Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.

The software is installed during system installation. For more information, see “Download, install, and update the software” on page 138.

Hardware description

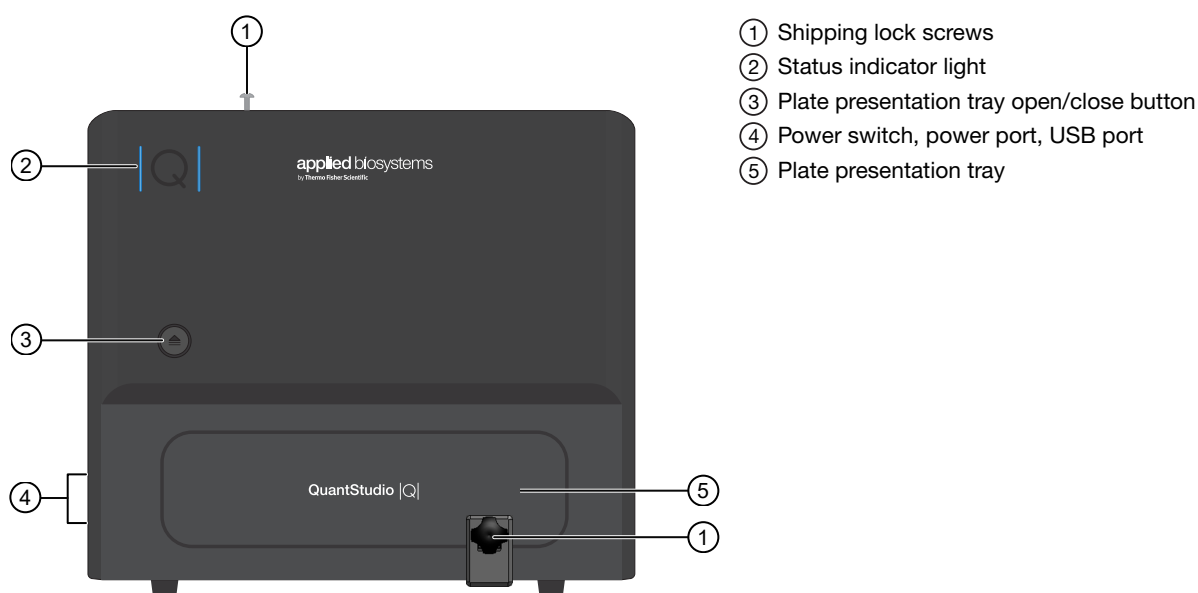
Overview of the Absolute Q™ instrument

The instrument is an integrated processing system compatible with QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates.

A dedicated computer provided with the instrument uses the QuantStudio™ Absolute Q™ Digital PCR Software to operate the instrument and analyze data.

For information about installing the instrument, see Appendix C, “Install, update, and move the QuantStudio™ Absolute Q™ Digital PCR System”.

For information about maintaining the instrument, see Appendix F, “Maintain the instrument”.



The instrument has the following features and functions.

- The plate presentation tray is controlled using a button on the front panel or from within the software. Once a MAP plate is loaded into the tray, it is retracted into the instrument for automated processing.
- An internal barcode scanner verifies the barcodes on the MAP plates.
- An internal compressor and pneumatic subsystem drives the microfluidic array compartmentalization directly within the MAP plate using positive pressure.
- Liquid never contacts any parts in the instrument, so minimal maintenance and cleaning are required.
- The plate nest is thermally controlled to perform PCR thermal cycling.
- The fluorescent optical system is mounted above the MAP plate and scans the MAP plate in up to 5 optical channels before and after PCR.
- Each optical channel is associated with a color and a supported dye (see “QuantStudio™ Absolute Q™ Digital PCR Instrument optical dyes” on page 18).
- A computer integrated into the instrument manages critical runtime activities and stores recent data that have not yet been analyzed.

- During an experiment run, positive pressure is applied to drive and separate the reagent mix into pico-scale microchambers on the MAP plate before starting PCR. PCR occurs in parallel across the entire MAP plate. Each microchamber contains a discrete reaction.
- The microchamber arrays are scanned for fluorescence before and after PCR and are used for data analysis.

Instrument indicator status light key

The vertical bars of the Q symbol on the front of the instrument display the instrument status.

Appearance	Color	Status	Meaning
~ Q ~	White	Flashing	On, initializing – not ready.
Q	Blue	Steady	Idle, ready to connect to Absolute Q™ software. Analyzing a run.
~ Q ~	Blue	Pulsing	Running protocol.
~ Q ~	Yellow	Brief flashing	Plate door open button pushed while door is locked.
Q	Red	Steady	Error, see Appendix A, “Troubleshooting”.

Instrument information

The  **Instrument** screen (accessible from the left navigation pane) in the QuantStudio™ Absolute Q™ Digital PCR Software provides information about the instrument that is connected to the desktop computer.

22050366

TIME STARTED
11:55

ESTIMATED TIME REMAINING

USER
LAB OPERATOR

PLATE BARCODE
M01KE224900353

✓

✓

3

Prime 100%

PCR 100%

Scan 100%



SERIAL NUMBER
22050366

INSTRUMENT SOFTWARE
1.0.45

INSTRUMENT FIRMWARE
0.9.4

LAST CALIBRATION DATE
2023-06-06T18:48:22.714025

CALIBRATION TYPE
Single instrument calibration

PREPARE FOR SHIPPING

CALIBRATE SYSTEM DYES

The instrument software displays the following states for the device.

State	Description
READY	The instrument is ready to run an experiment.
RUNNING	The statistics of the run in progress are displayed. Note: A run can be interrupted from the Instrument screen by clicking STOP .
ERROR	Displays an error if an error condition has occurred.
No instrument found	An instrument is not connected to the desktop computer.
UPDATE	Incompatible instrument configuration detected (see “Download, install, and update the software” on page 138).

QuantStudio™ Absolute Q™ Digital PCR Instrument optical dyes

The following optical dyes are supported for use when selecting optical channels when analyzing experiment runs.

For more information about optical configuration, see “QuantStudio™ Absolute Q™ Digital PCR Instrument Optical Configuration” on page 191.

Channel color	System dyes
Blue	FAM™ dye
Green	VIC™ dye (<i>recommended</i>) HEX™ ^[1,2]
Yellow	ABY™ dye
Red	JUN™ dye ROX™ dye
Dark Red	Cy5™ dye

^[1] For information about HEX™ dye support, contact a Thermo Fisher service and support representative.

^[2] HEX™ data from two instruments cannot be combined into a study, even if the systems are co-calibrated. For more information, see “Multi-plate analysis (studies)” on page 107.

Overview of the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite

The AutoRun Suite uses the Absolute Q™ Digital PCR System integrated with the Thermo Fisher Scientific Spinnaker™ Microplate Mover to manage loading and unloading plates. The AutoRun Suite can be set up with automation using a single or dual Absolute Q™ dPCR instrument configuration to dynamically scale throughput with multi-plate and multi-batch analysis. For more information about using the AutoRun Suite, see Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.

The QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite is available in a single or a dual configuration.

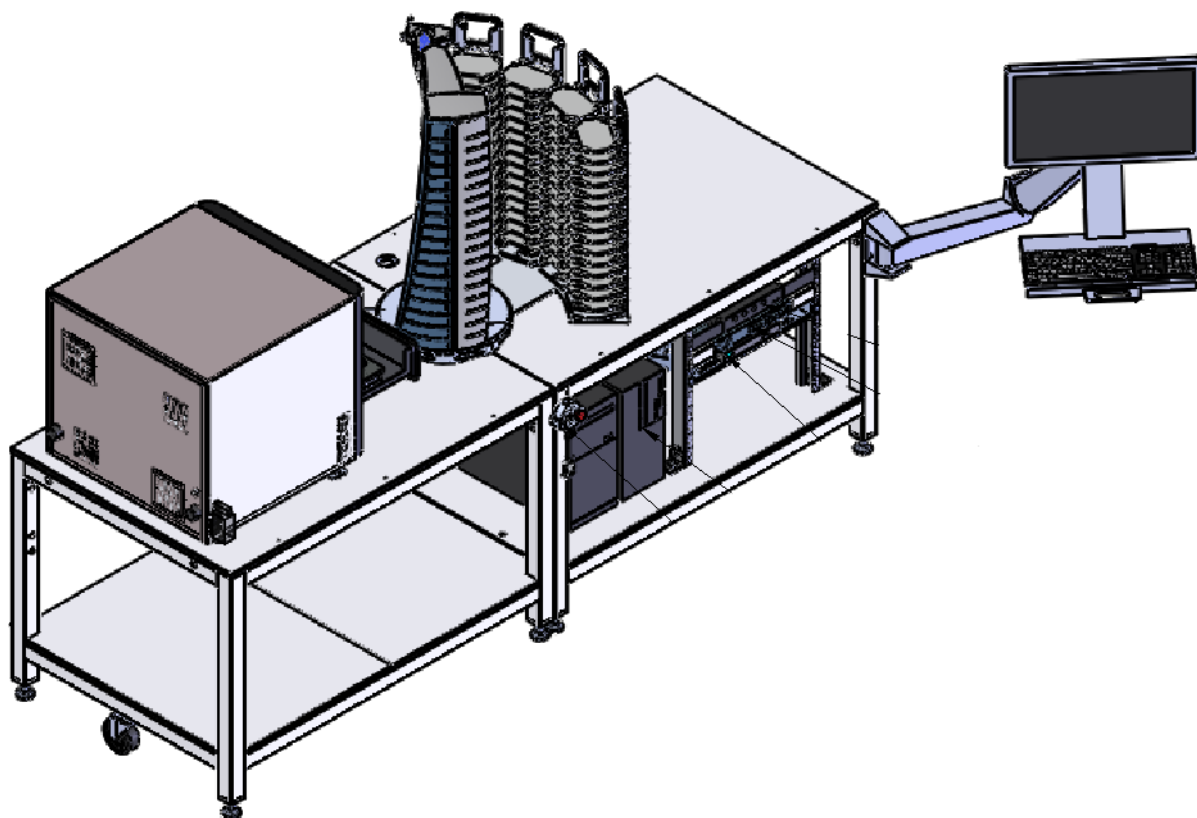


Figure 1 QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite single configuration

The single configuration contains 1 Absolute Q™ system and the Spinnaker™ Microplate Mover with random access hotels or sequential access stacks.

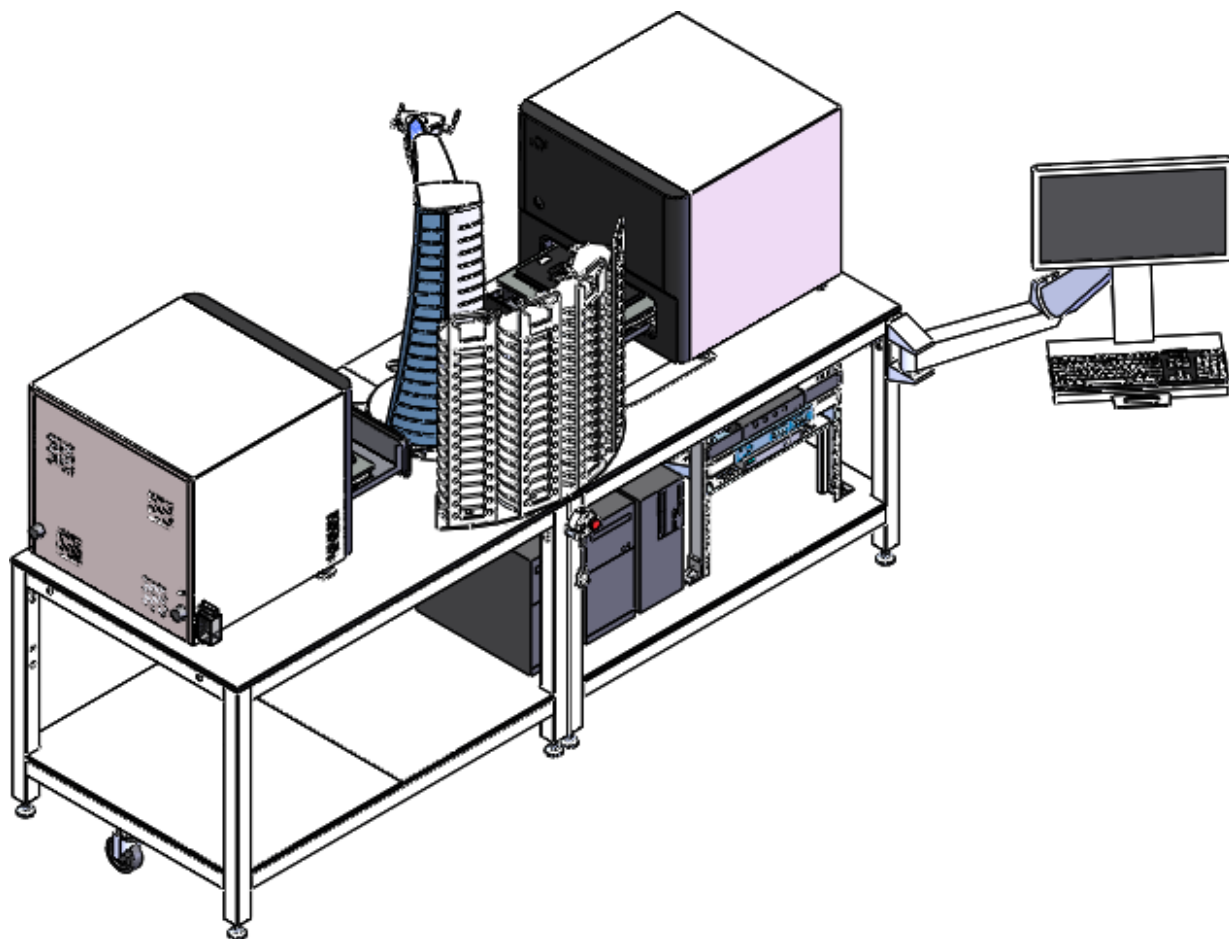


Figure 2 QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite dual configuration

The dual configuration contains 2 Absolute Q™ systems and the Spinnaker™ Microplate Mover with random access hotels or sequential access stacks.

QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates description

The QuantStudio™ Absolute Q™ Digital PCR Instrument uses QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates (MAP plates) for loading samples and running experiments.

IMPORTANT! When disposing of plates, follow all applicable waste regulations controlling the chemicals used in the experiment.

Each MAP plate has the following features.

- Contains 16 wells, 4 columns of 4 wells each, and each experiment must use at least one full column (4 samples).
- Contains 16 digital PCR microchamber arrays that each contain 20,480 fixed volume microchambers where dPCR is performed.
- Can be used in up to 4 experiments, depending on the number of columns used in each experiment. A MAP plate with unused columns can be used with subsequent experiments until all 4 columns have been used.
- Has a standard microtiter plate footprint and is compatible with most plate and liquid handlers.
- Has a label that includes a barcode, product number, and unique serial number. The instrument automatically reads the barcode when the MAP plate is inserted, and the unique serial number is tracked in the results.
- Requires 1 MAP plate gasket strip be placed on each column before insertion into the instrument, regardless of whether the column is being used for the experiment.

Take the following into consideration when using MAP plate gasket strips.

- Following use on a column that was used for sample testing, the MAP plate gasket strip cannot be reused.
- MAP plate gasket strips on unused columns can be reused until the column is used for sample testing.
- MAP plate gasket strips used on column X can be used for all runs on that MAP plate.

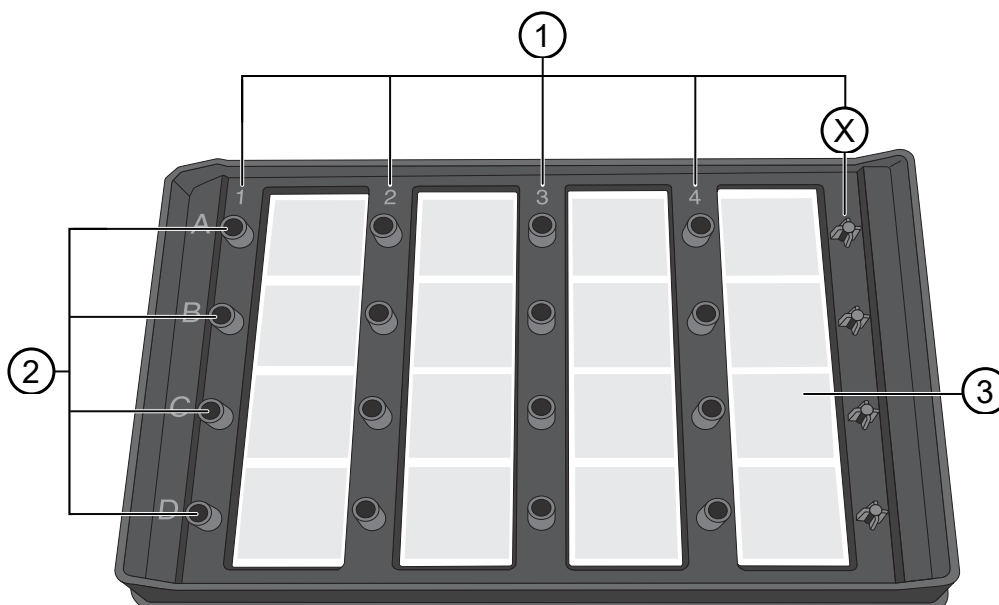


Figure 3 MAP plate without MAP plate gasket strips

- ① Columns 1–4 and column X
- ② Wells A1–D1 associated with column 1
- ③ Microchamber associated with well 4C

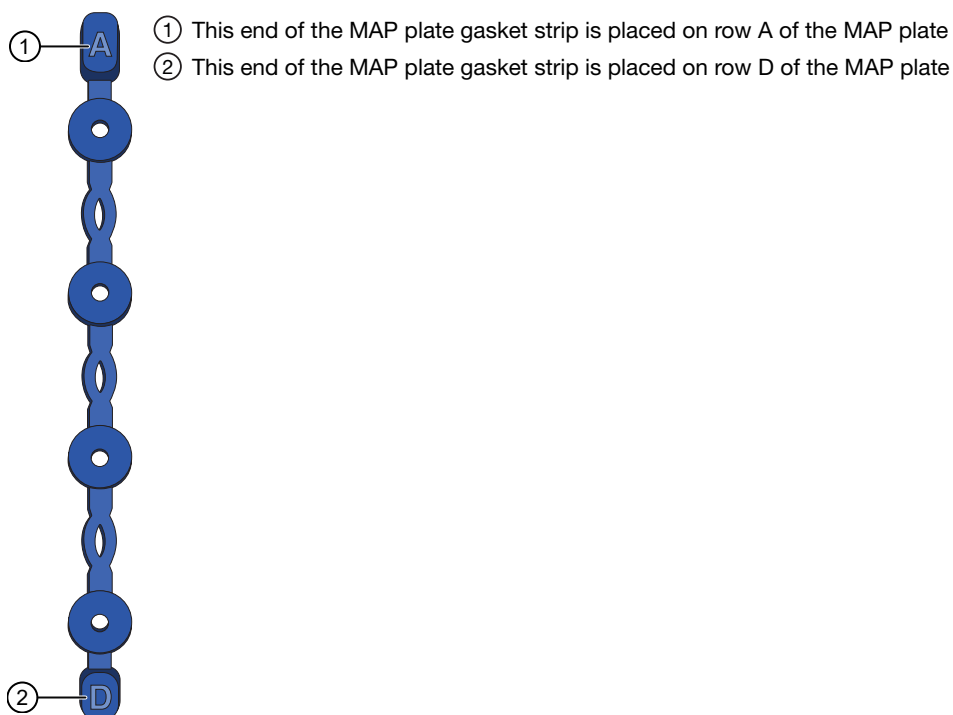


Figure 4 MAP plate gasket strip

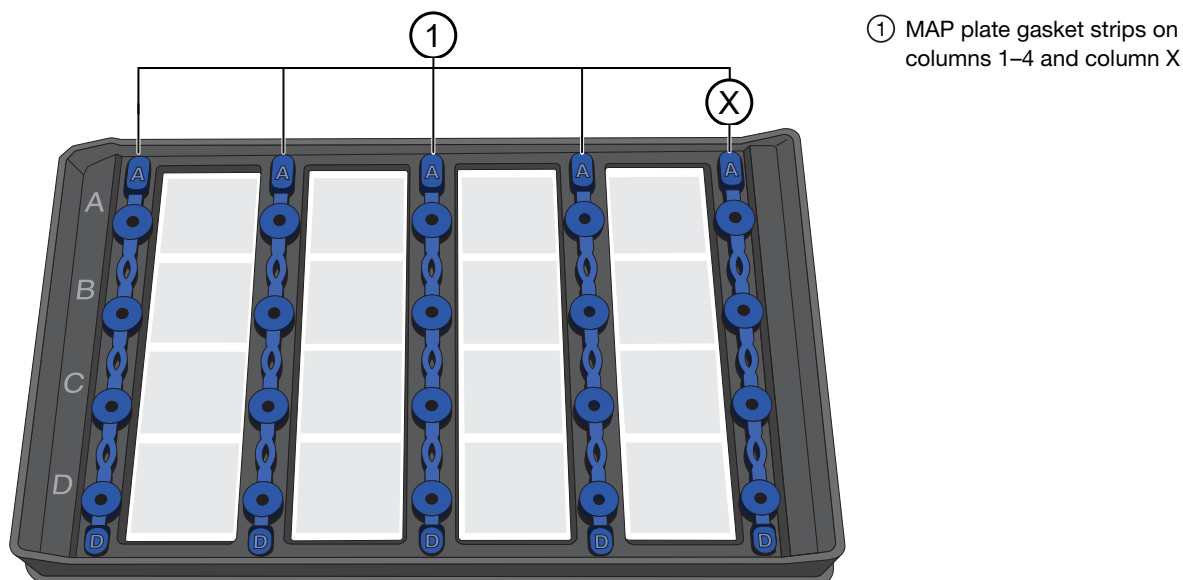


Figure 5 MAP plate with MAP plate gasket strips in place

The following figure shows the dimensions of a MAP plate.

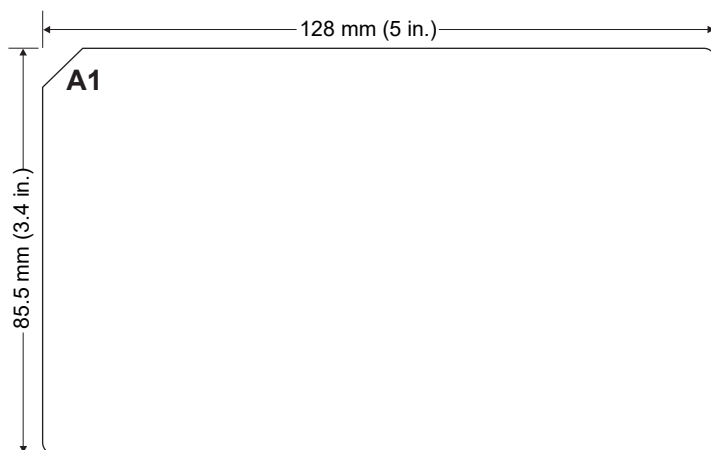


Figure 6 MAP plate dimensions

QuantStudio™ Absolute Q™ MAP16 Digital PCR Plate compatibility

IMPORTANT! The instrument is only compatible with QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates. The instrument can malfunction with third-party plates, which could result in contamination and/or damage to the instrument.

- For best results, we strongly recommend that you use an Absolute Q™ Master Mix and QuantStudio™ Absolute Q™ Isolation Buffer.
- MAP plates are made of injection molded thermoplastic commonly used in other PCR vessels and are generally compatible with most existing reagent kits and components available from third parties. Compatibility of any untested third-party reagent is not guaranteed. Contact technical support for more information on tested reagents (see Appendix K, “Documentation and support”).

QuantStudio™ Absolute Q™ Digital PCR Software security

By default, the QuantStudio™ Absolute Q™ Digital PCR Software does not require login credentials to access the software nor does it restrict access to functions within the software.

To require login credentials and modify access by user roles, see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”.

Network and password security requirements

Network configuration and security

The network configuration and security settings of your laboratory or facility (such as firewalls, anti-virus 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.

Single-plate workflow

This workflow represents running a single experiment on the QuantStudio™ Absolute Q™ Digital PCR Instrument.

Experiment workflow	
	Prepare an experiment (page 27)
	Run digital PCR (page 38)
	Analyze data (page 77)

Automation workflow

The QuantStudio™ Absolute Q™ Digital PCR Software lets you create dPCR runs that can be used on an automated system. This workflow represents the steps in the Absolute Q™ software that are necessary for running dPCR runs using automation. If you are using an automated system other than the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite, see the documentation provided with that system for details on performing runs.

Automation workflow

Prepare an experiment (page 27)

Create and export a template (page 40)

OR

Create and export draft runs (page 54)

Import template or draft run files into your automated system

Perform a run (page 144)

Export completed run data from your automated system (page 144)

Import the run data into the QuantStudio™ Absolute Q™ Digital PCR Software (page 66)

Analyze the imported run data (page 77)



Prepare an experiment

■ Sample preparation workflow	27
■ DNA preparation guidelines	28
■ RNA preparation guidelines	29
■ Sample dilution guidelines	30
■ Prepare the dPCR reaction mix for DNA	30
■ Prepare the dPCR reaction mix for RNA	31
■ Load the reaction mix into the MAP plate	33

This chapter provides a general protocol for preparing experiments using custom dPCR assays ordered at thermofisher.com/dpcr-assays.html or with your custom dPCR assays. For pre-designed dPCR assays, follow the instructions in the assay user guide provided with the assay.

Sample preparation workflow

The following workflow represents preparing a single experiment on the QuantStudio™ Absolute Q™ Digital PCR Instrument.

Note: The procedure for sample preparation can vary depending on application and reagents.

Sample preparation workflow

Review DNA preparation guidelines (page 28)

AND/OR

Review RNA preparation guidelines (page 29)

Review Sample dilution guidelines (page 30)

Prepare the dPCR reaction mix for DNA (page 30)

AND/OR

Prepare the dPCR reaction mix for RNA (page 31)

Load the reaction mix into the MAP plate (page 33)

Run digital PCR (page 38)

DNA preparation guidelines

We recommend the following best practices for the preparation of genomic DNA (gDNA) template or complementary DNA (cDNA) template, for use in digital PCR (dPCR) experiments. Because dPCR experiment strategy and methodology can vary significantly, sample preparation and template quality must be assessed on an individual basis.

For information about DNA isolation kits, go to thermofisher.com/us/en/home/life-science/dna-rna-purification-analysis.html.

Quality of DNA

Use a gDNA or cDNA template that meets the following criteria.

- DNA is extracted from the raw material that you are testing with an optimized protocol.

IMPORTANT!

- Salting-out procedures and crude lysates are not recommended.
 - Failure to adhere to these recommendations can cause PCR amplification failures and clogging of the microfluidics both leading to poor or absent data collection.
-

- DNA contains minimal PCR inhibitors.
- DNA has $A_{260/280}$ ratio ~1.8.
- DNA has an $A_{260/230}$ ratio between 2.0 and 2.2.

The ratio of absorbency at 260 nm and 280 nm is used to evaluate the purity of DNA. A ratio of ~1.8 is generally accepted as “pure” for DNA. A ratio that is appreciably below the expected can indicate the presence of protein, phenol, or other contaminants that absorb strongly at or near 280 nm.

The ratio of absorbency at 260 nm and 230 nm is used as a secondary determinant of nucleic acid purity. The 260/230 values for “pure” nucleic acid are often greater than the respective 260/280 values. Expected 260/230 values are commonly in the range of 2.0 to 2.2. A ratio that is appreciably below the expected can indicate the presence of contaminants that absorb at 230 nm.

Quantity of DNA

The quantity of DNA template added to a dPCR reaction depends on the following factors.

- Concentration of gDNA or cDNA present in each sample.
- Expected number of copies of the target sequence present in the genome or cDNA of your samples.

Before performing digital PCR experiments, consider quantifying the amount of gDNA or cDNA in each sample.

We recommend one of the following methods for quantification, see “Recommended materials not supplied” on page 13.

- Quant-iT™ 1X dsDNA Assay Kit, High Sensitivity using the Qubit™ Flex Fluorometer
- Spectrophotometer

RNA preparation guidelines

We recommend the following best practices for the preparation of RNA template for use in digital PCR (dPCR) experiments. Because dPCR experiment strategy and methodology can vary significantly, sample preparation and template quality must be assessed on an individual basis.

For information about RNA isolation kits, go to thermofisher.com/rnaisolation.

Quality of RNA

Use an RNA template that meets the following criteria.

- RNA must be free of inhibitors of reverse transcription and PCR.
- RNA must be free of RNase activity.
- RNA has $A_{260/280}$ ratio ~2.0.
- RNA has an $A_{260/230}$ ratio between 2.0 and 2.2.

The ratio of absorbency at 260 nm and 280 nm is used to evaluate the purity RNA. A ratio of ~2.0 is generally accepted as “pure” for RNA. A ratio that is appreciably below the expected can indicate the presence of protein, phenol, or other contaminants that absorb strongly at or near 280 nm.

The ratio of absorbency at 260 nm and 230 nm is used as a secondary determinant of nucleic acid purity. The 260/230 values for “pure” nucleic acid are often greater than the respective 260/280 values. Expected 260/230 values are commonly in the range of 2.0 to 2.2. A ratio that is appreciably below the expected can indicate the presence of contaminants that absorb at 230 nm.

Quantity of RNA

- The recommended concentration of RNA for the dPCR reactions is 1–4,000 copies/μL.
- A lower or higher concentration of RNA can be used for the dPCR reactions, depending on the application. We recommend testing a lower or higher concentration of RNA with the application.

Sample dilution guidelines

If a target is present at a sufficiently high concentration in the sample of interest, it is possible that all reaction microchambers will be positive, which prevents the determination of the target concentration. In this case, the sample must be diluted prior to running the dPCR experiment. To determine the optimal dilution for your sample, see “Determine the optimal sample dilution” on page 124.

After you determine the dilution factor you can include it as part of sample setup (see “SETUP tab (templates)” on page 44 or “SETUP tab (draft runs)” on page 59).

Prepare the dPCR reaction mix for DNA

This section provides general information for using the Absolute Q™ Universal DNA Digital PCR Master Mix (5X) and your dPCR assay to prepare a dPCR reaction mix.

For information about preparing the dPCR reaction mix for a pre-designed Absolute Q™ dPCR assay, see the documentation provided with the assay.

Note: MAP plates are made of injection molded thermoplastic commonly used in other PCR vessels and are generally compatible with most existing reagent kits and components available from third parties. Compatibility of any untested third-party reagent is not guaranteed. Contact technical support for more information on tested reagents (see Appendix K, “Documentation and support”).

Prepare the following materials.

- Absolute Q™ Universal DNA Digital PCR Master Mix (5X)
- Nuclease-free water
- Digital PCR assay (40X or 20X)

IMPORTANT!

- Throughout this procedure, protect reagents from light when not in use.
 - For best results, use Applied Biosystems™ TaqMan™ Assays or Absolute Q™ digital PCR assays with the Absolute Q™ Universal DNA Digital PCR Master Mix (5X). Applied Biosystems™ reagents have been tested for in-plate stability at ambient temperature for up to 96 hours, to support automation.
-

1. Thaw and equilibrate all reagents to room temperature before use.

Note: Store reagents on ice when not in use.

2. Vortex the Absolute Q™ Universal DNA Digital PCR Master Mix (5X) and dPCR assay (40X or 20X) at high speed for 10 seconds.

3. Using a benchtop centrifuge, centrifuge the DNA sample at $10,000 \times g$ or the highest speed available for 1 minute, then transfer the supernatant to the reaction mix as indicated in step 4.
4. Combine the following reagents in the order listed.

Reagent	Final concentration	Volume per reaction	Volume per reaction with 10% overage ^[1]
Nuclease-free water	—	Fill to 9 μL	Fill to 10 μL
Absolute Q™ Universal DNA Digital PCR Master Mix (5X)	1X	1.8 μL	2 μL
Digital PCR assay (40X or 20X) ^[2]	1X	0.23 μL (40 X) or 0.45 μL (20X)	0.25 μL (40 X) or 0.50 μL (20X)
DNA Sample	1–11,000 copies/ μL ^[3]	Variable	Variable
Total	—	9 μL	10 μL

^[1] After calculating the number of reactions required, prepare the dPCR mix for the appropriate number of reactions and scale those components by 10% for overage. Dilute the assay accordingly to avoid pipetting less than 1 μL volumes.

^[2] If you are using a dPCR assay with a stock concentration other than 40X or 20X you must manually calculate the volumes based on the concentration you are using.

^[3] A DNA copy and dilution calculator can be found at <http://www.thermofisher.com/DNA-calculator>.

5. Mix the dPCR reagents well using one of the following approaches.
 - Pipette mix 10–20 times. Avoid generating bubbles.
 - Pulse vortex 3–5 times for 1 second each.
6. Centrifuge at $1,000 \times g$ for up to 1 minute to collect the contents at the bottom of the tube.

Proceed to “Load the reaction mix into the MAP plate” on page 33.

Prepare the dPCR reaction mix for RNA

This section provides general information for using the Absolute Q™ 1-Step RT-dPCR Master Mix (4X) and your dPCR assay to prepare a dPCR reaction mix.

For information on preparing the dPCR reaction mix for a pre-designed Absolute Q™ dPCR assay, see the documentation provided with the assay.

Note: MAP plates are made of injection molded thermoplastic commonly used in other PCR vessels and are generally compatible with most existing reagent kits and components available from third parties. Compatibility of any untested third-party reagent is not guaranteed. Contact technical support for more information about tested reagents (see Appendix K, “Documentation and support”).

Prepare the following materials.

- Absolute Q™ 1-Step RT-dPCR Master Mix (4X)
- Nuclease-free water
- Digital PCR assay (20X)

IMPORTANT!

- Throughout this procedure, protect reagents from light when not in use.
- For best results, use Applied Biosystems™ TaqMan™ Assays or Absolute Q™ digital PCR assays with the Absolute Q™ 1-Step RT-dPCR Master Mix (4X).
- Store prepared reactions on ice or at 4°C for up to one hour.

The volume of the dPCR reaction can be adjusted depending on experimental requirements. Scale the components proportionally according to the number of reactions and include 10% overage.

Note:

- Thaw RNA templates on ice or at 4°C.
- Bring the reaction mix to room temperature before loading the MAP plate.

1. Vortex the Absolute Q™ 1-Step RT-dPCR Master Mix (4X) and the assay at high speed for 10 seconds.
2. Combine the following reagents in the order listed.

Table 1 dPCR reaction with a 20X assay

Reagent	Final concentration	Volume per reaction (with 10% overage)	Volume for 4 reactions with 10% overage
Nuclease-free water	—	For 10 µL total reaction volume	For 40 µL total reaction volume
Absolute Q™ 1-Step RT-dPCR Master Mix (4X)	1X	2.5 µL	10.0 µL
Digital PCR assay (20X) ^[1]	1X	0.5 µL	2.0 µL
RNA sample	1–4,000 copies/µL	Variable	Variable
Total reaction volume	—	10 µL	40 µL

^[1] Adjust the volume if the assay is a custom assay at a different concentration. Adjust the volume of water to achieve the total reaction volume.

3. Mix the dPCR reagents well by performing one of the following actions.
 - Pipet up and down 10–20 times to mix. Avoid creating bubbles.
 - Pulse vortex 3–5 times for 1 second each.
4. Using a benchtop centrifuge, centrifuge at 10,000 × g for one minute to collect the contents at the bottom of the tube.

Proceed to “Load the reaction mix into the MAP plate” on page 33.

Load the reaction mix into the MAP plate

At a clean lab bench, gather the following materials.

- P10 or P20 pipette and filter pipette tips
- Prepared dPCR reaction mix
- QuantStudio™ Absolute Q™ Isolation Buffer
- MAP plate with sufficient unused columns for the experiment
- MAP plate gasket strips (unused)

IMPORTANT! At least 1 column of the MAP plate must be used for each run and all wells in the column must contain a sample (or water plus isolation buffer if there is insufficient sample to fill all wells). Columns cannot be reused, but a MAP plate with unused columns can be used for subsequent experiments. If the MAP plate has unused columns, when the experiment is complete, place it back into its pouch for storage.

Note: The MAP plate follows SBS standard plate format, allowing for use with an automated liquid handling workflow. For information about potential implementation and guidance, contact your Thermo Fisher Scientific representative or visit <http://www.thermofisher.com/autorun>.

1. Just prior to use, remove the MAP plate from its package.

Note:

- Leave the MAP plate in the package until ready to load sample.
 - Be careful to handle the MAP plate by its frame.
 - Place the MAP plate back into the package when not in use.
-

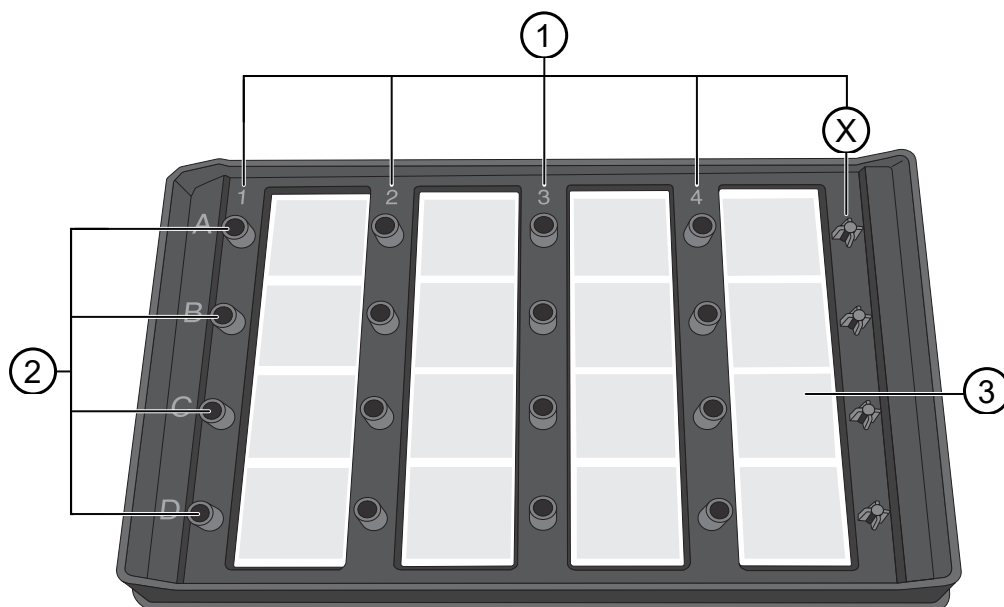


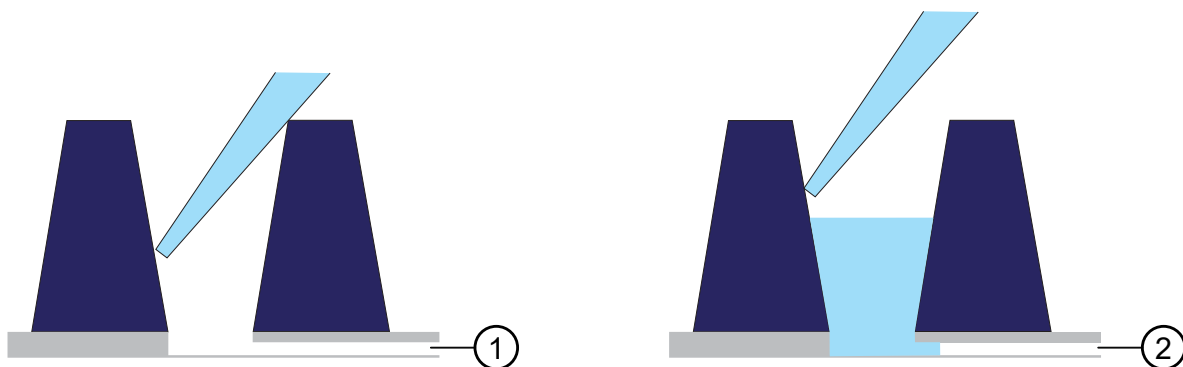
Figure 7 MAP plate without MAP plate gasket strips

- ① Columns 1–4 and column X
- ② A–D represent wells A1–D1 associated with column 1
- ③ Array associated with well 4C

2. Place the MAP plate on a level, dust-free, dry surface.
3. Using a new pipette tip for each well, holding the pipette at a 45° angle, load 9 μ L of the dPCR reaction mix to the bottom of the well. Pipet the mixture only to the first stop to prevent bubble formation.

IMPORTANT! To avoid the transfer of contents from the bottom of the centrifuged dPCR reaction mix tube, do not pipet from the bottom of the tube.

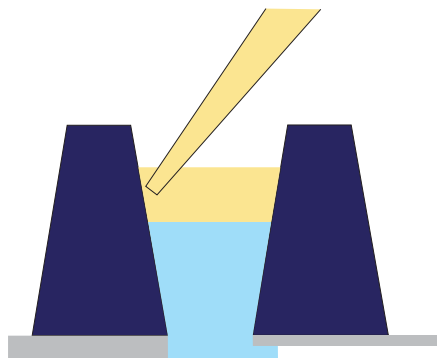
IMPORTANT! Do not contact bottom of well with the pipette tip or puncture the thin film at the bottom of the well.



- ① Microfluidic channel to the microchamber array
- ② Reaction mix remains in the well until the instrument pushes it into the microchamber array during the run

4. Using a new pipette tip for each well, at a 45° angle, load 15 µL of the Absolute Q™ Isolation Buffer on the side of the well above the top of the reaction mix. Carefully overlay the buffer on top of the reaction mix to prevent mixing or bubble formation. Pipet only to the first stop.

The isolation buffer sits on top of the reaction mix, preventing contamination and evaporation.



5. Place a total of 5 MAP plate gasket strips on all 4 columns of wells and the X-shaped posts of column X on the right side of the plate. Orient the MAP plate gasket strip so that the side labeled A–D aligns with rows A–D marked on the plate. Be sure to cover the columns completely and press the MAP plate gasket strips firmly into place.

IMPORTANT! MAP plate gasket strips must be placed on all columns, including unused columns. Failure to do so can produce poor results.

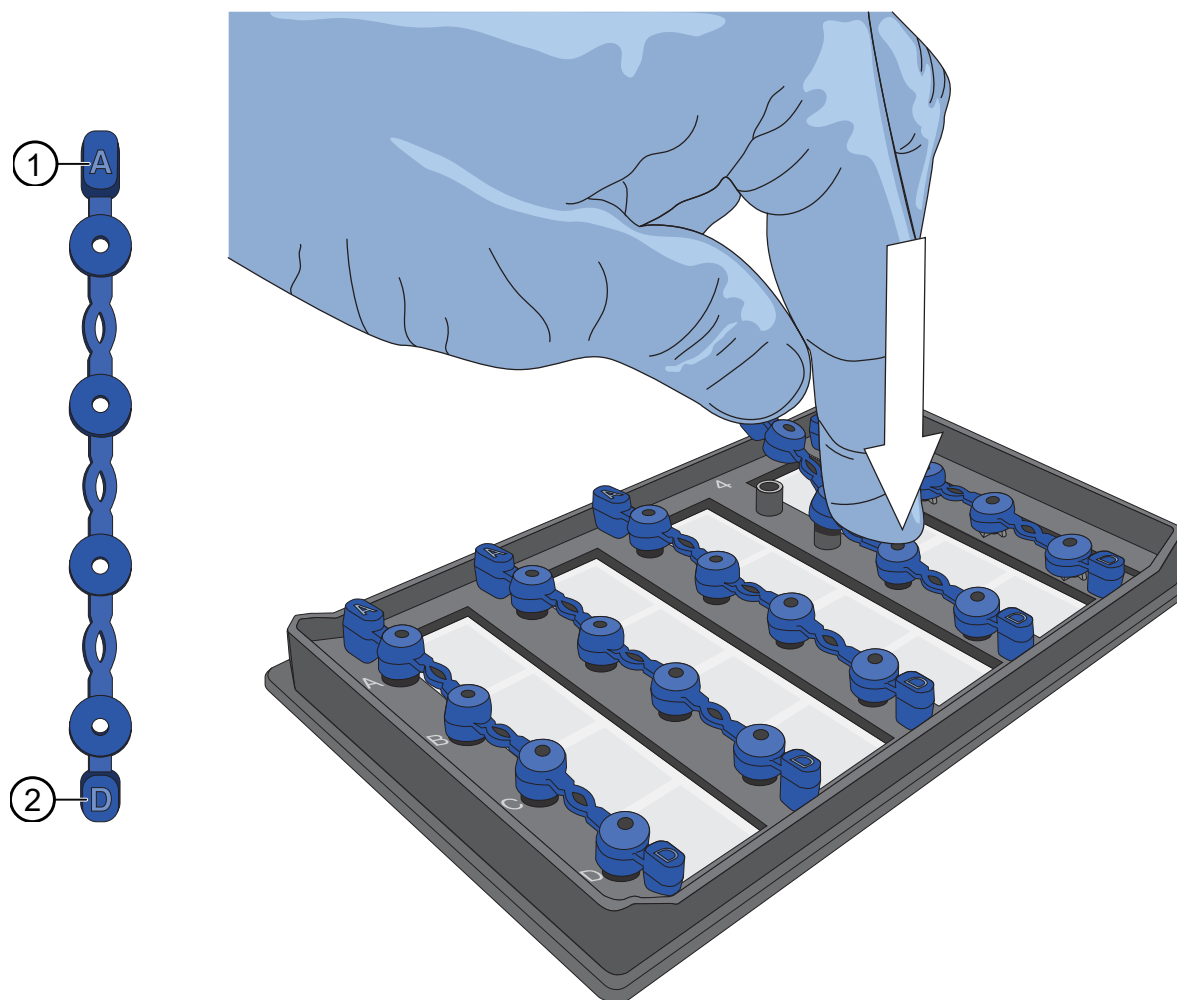


Figure 8 Place the MAP plate gasket strips firmly into place

- ① Place this end of the MAP plate gasket strip on row A
- ② Place this end of the MAP plate gasket strip on row D

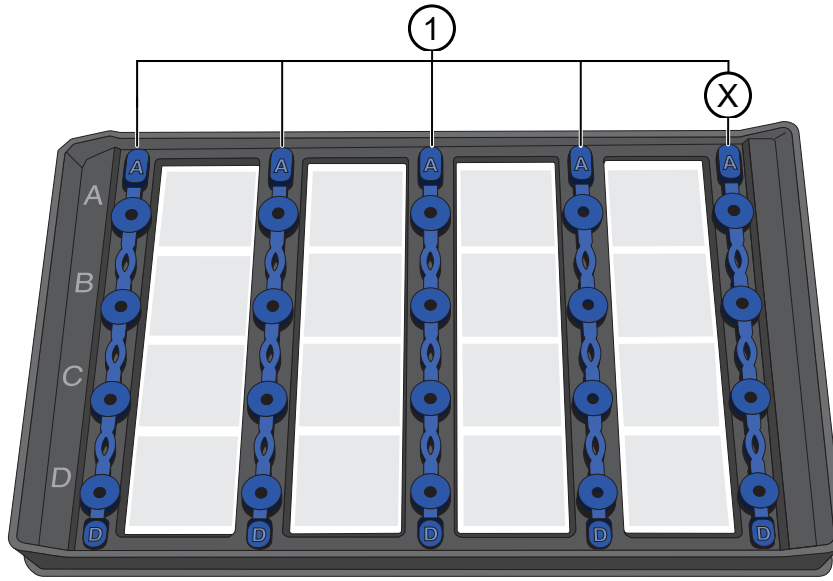


Figure 9 MAP plate with MAP plate gasket strips in place

① MAP plate gasket strips on columns 1–4 and column X

6. Move the MAP plate to the instrument.

IMPORTANT! Do not tip, invert, or shake the filled MAP plate.

■ Power on the instrument and computer	38
■ Dashboard	39
■ Manage templates	40
■ Manage runs	54
■ Manage groups	70

This chapter provides information for running experiments on the QuantStudio™ Absolute Q™ Digital PCR Instrument.

Power on the instrument and computer

IMPORTANT! Before powering on the QuantStudio™ Absolute Q™ Digital PCR Instrument, confirm that the shipping lock screws have been removed. Failure to do so can damage the instrument. For more information, see “Uninstall the shipping lock screws” on page 140.

1. Confirm that the power cable is connected to an appropriate power source.
2. Power on the instrument by moving the power switch located on the left side near the back of the instrument to the **I** position.

Note: The instrument makes a humming noise as it charges the internal compressor.

The bars of the instrument symbol flash white to indicate that the system is initializing. This takes approximately 30 seconds.

The instrument is ready when the status lights are a steady blue.

3. Confirm that the USB 3.0 cable is connected from the instrument to the USB 3.0 port on the dedicated computer.
4. Power on the dedicated computer and monitor, then start the software.

Note: The instrument is ready when ready status appears under the instrument in the **Instrument** screen in the QuantStudio™ Absolute Q™ Digital PCR Software.

Note: If your facility is implementing the Security, Auditing, and e-Signature (SAE) module, you are prompted for a user ID and password. Contact your system administrator to obtain login credentials.

Dashboard

When you launch the QuantStudio™ Absolute Q™ Digital PCR Software, the **Dashboard** screen opens. Use the left navigation pane and the **Dashboard** to access the software functions.

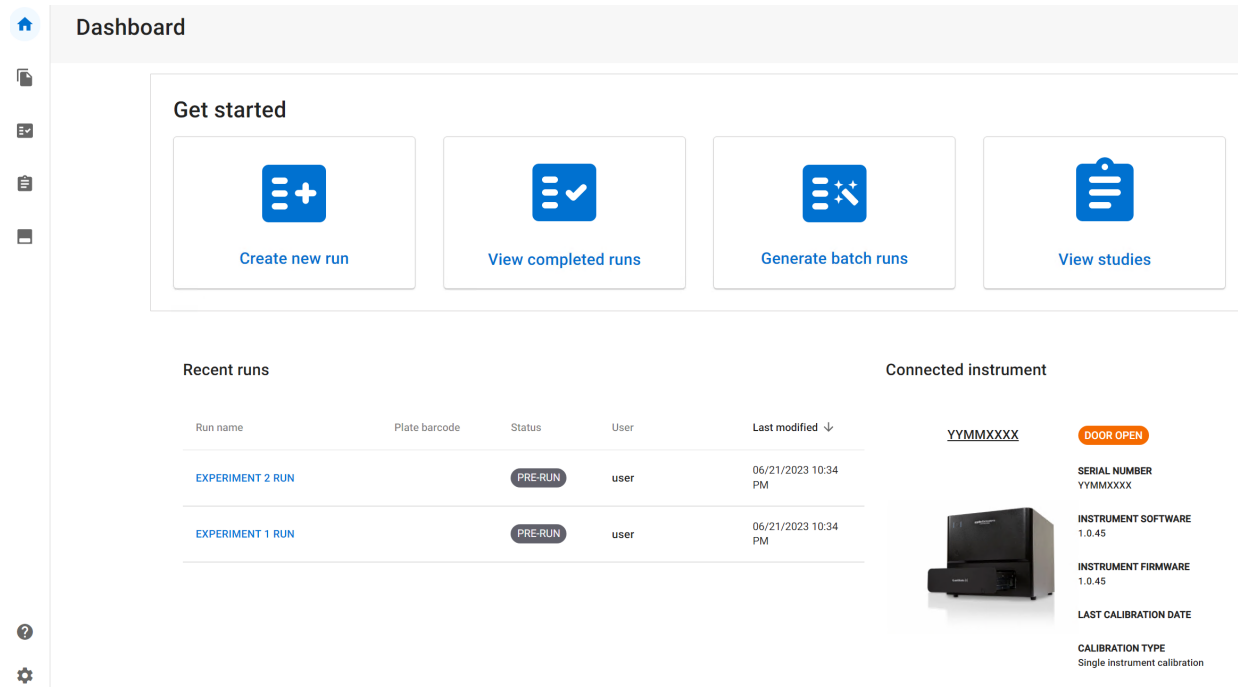


Figure 10 Dashboard with a connected instrument

The left navigation pane provides access to functions from any screen in the software.

- **Dashboard**—Return to the **Dashboard**.
- **Templates**—Open the **Templates** screen (see “Manage templates” on page 40).
- **Runs**—Open the **Runs** screen (see “Manage runs” on page 54).
- **Studies**—Open the **Studies** screen (see “Multi-plate analysis (studies)” on page 107).
- **Instrument**—Open the **Instrument** screen (see “Instrument information” on page 17).
- **Help**—Access the end user license agreement (EULA) and information about the currently installed version of software.
- **System Settings**—Access the **System Settings** screen (see “Configure system settings” on page 131).

The **Dashboard** screen provides the following shortcuts to software functions and system information.

- **Create new run**—Opens the **Create run** dialog, where you can create a new run. You can also create a new run from the **Runs** screen (see “Create a run from the Runs screen” on page 55).
- **View completed runs**—Opens the **Runs** screen to the **COMPLETED** tab that lists all completed runs (see “Open a completed run” on page 79).
- **Generate batch runs**—Opens the **Generate batch runs** dialog, where you can create batch runs from templates (see “Generate batch runs from a template” on page 51).
- **View studies**—Opens the **Studies** screen, where you can view, manage, and create studies (see “Multi-plate analysis (studies)” on page 107).

- **Recent runs**—Lists information about the most recent runs. You can select a run to see more information about the run in the **Runs** screen.
- **Connected instrument**—Provides information about the connected instrument. This information only appears when an instrument is connected.

Manage templates

Templates include protocol parameters and plate configurations that can be used for creating new runs. The software is preconfigured with a default template.

You can perform the following tasks from the **Templates** screen.

- View information about the template, including whether it can be edited or read-only, number of samples defined in the experiment, the user who created the template, and the date and time when the template was last modified.
- Open a template to view and edit protocol and plate setup parameters (see “Edit a template” on page 48).
- **IMPORT TEMPLATES**—Import one or more templates (see “Import templates” on page 49).
- **CREATE TEMPLATE**—Create a template with specific protocol and plate setup parameters (see “Create a template” on page 40).
- **GENERATE BATCH RUNS**—Create batch runs from a single template (see “Generate batch runs from a template” on page 51).
- **EXPORT**—Export one or more templates (see “Export templates” on page 49).
- **RENAME**—Rename a template (see “Rename a template” on page 50).
- **DELETE**—Delete one or more templates (see “Delete templates” on page 50).
- Search for a template by name.
- Create a run from a template (see “Create a run from a template” on page 50).

Create a template

Create a template to define PCR protocol parameters and plate configuration for use in creating runs.

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. In the upper-right corner, click **CREATE TEMPLATE**.
3. When prompted, enter a template name, then click **CREATE TEMPLATE**.
The new template opens to the **PROTOCOL** tab.

Continue with template creation in the **PROTOCOL** and **SETUP** tabs.

Action	Description
Define PCR parameters	PROTOCOL tab (templates) (page 41)
Define samples, sample groups, targets, dyes, analyses, and thresholds	SETUP tab (templates) (page 44)

PROTOCOL tab (templates)

The **PROTOCOL** tab in template creation or editing screen supports the following tasks when configuring the protocol. For detailed instructions, see “Define or edit protocol (PROTOCOL tab—templates)” on page 41.

- Define PCR parameters—Use the controls to set protocol-specific parameters.
- Customize optical channels—Use the toggle switch to enable or disable optical channels.
- **IMPORT PROTOCOL**—Import an existing protocol.
- **EXPORT PROTOCOL**—Export the protocol.
- **GENERATE BATCH RUNS**—Create multiple runs (batch runs) containing plate-specific information such as a barcode and sample information from a template. Batch runs can be used both in non-automated and automated configurations (see “Generate batch runs from a template” on page 51).
- **MAKE A COPY**—Create a copy of the template.

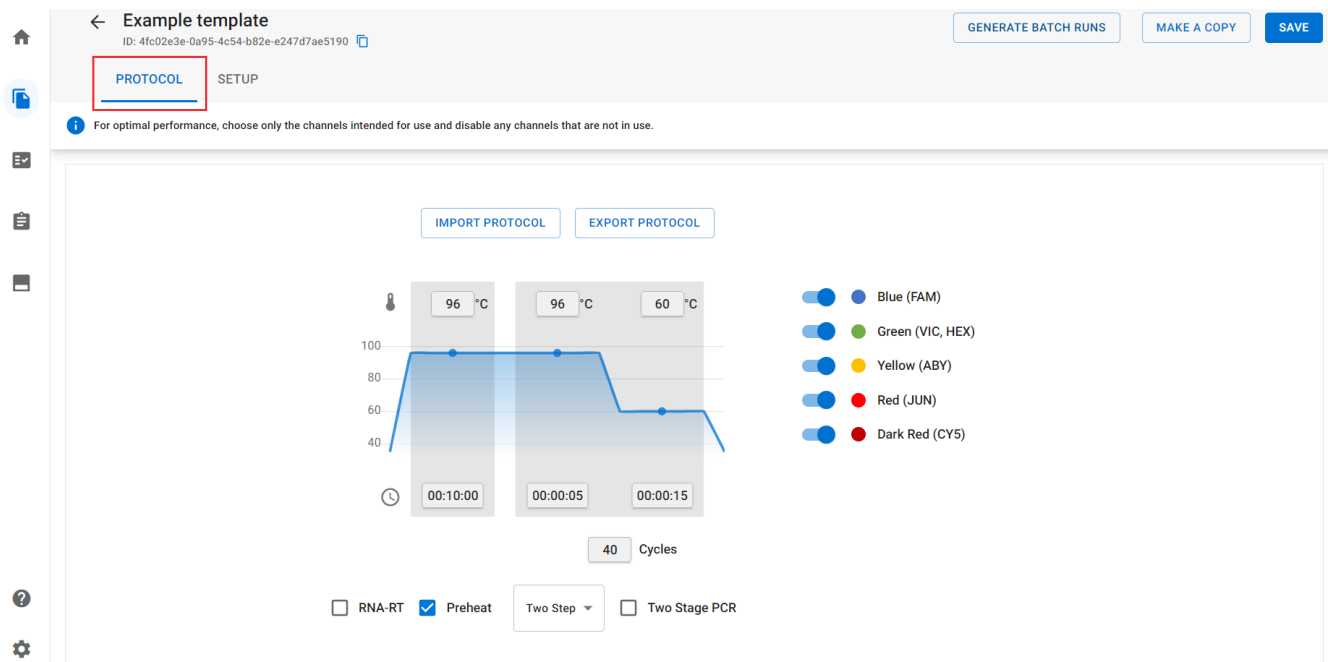


Figure 11 PROTOCOL tab (templates)

Define or edit protocol (PROTOCOL tab—templates)

Before you begin, create a template or open an existing template to edit.

- Create a template (page 40)
- Edit a template (page 48)

1. Open the **PROTOCOL** tab.
2. Modify PCR parameters as needed.

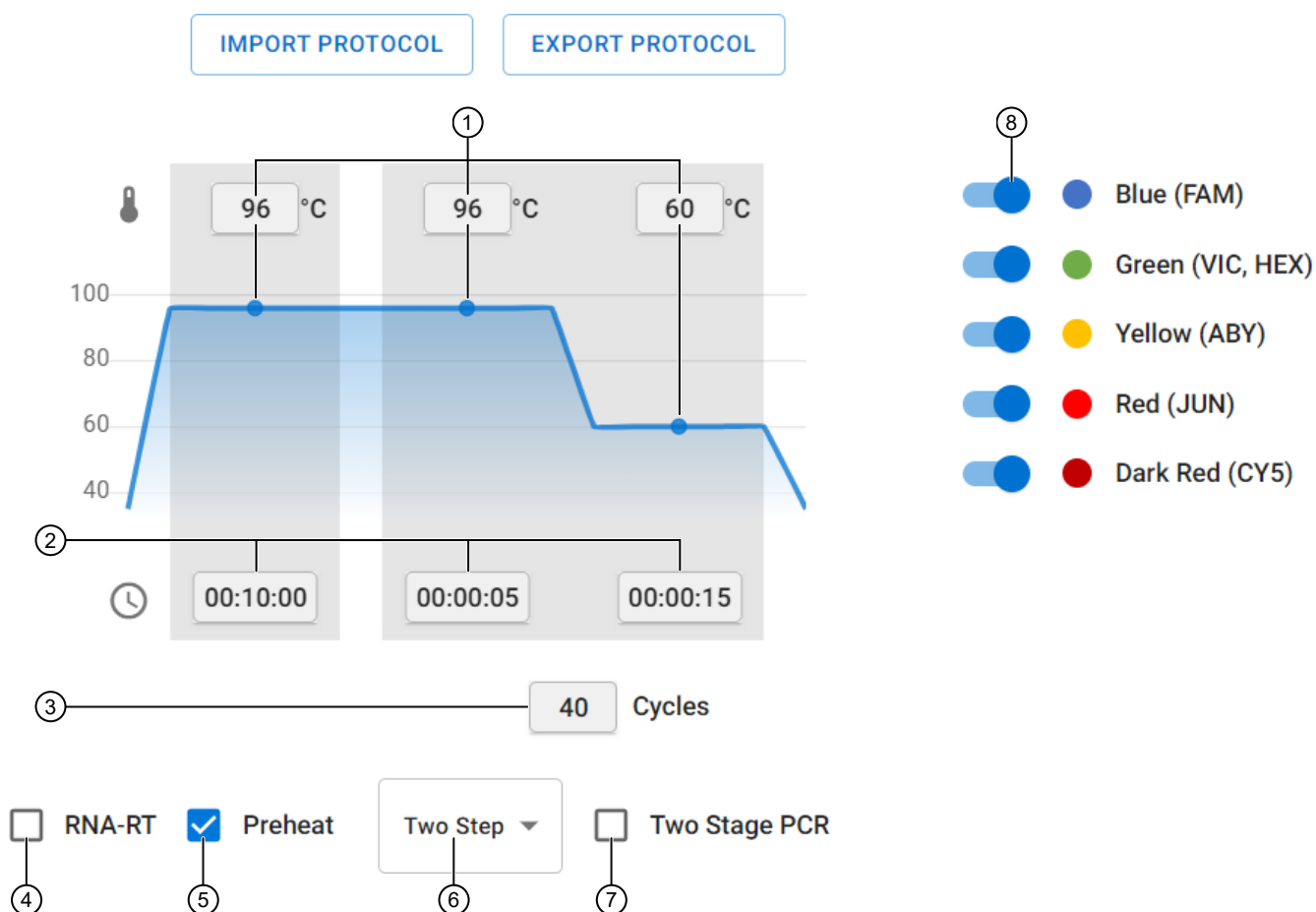


Figure 12 Protocol parameters

- | | |
|-------------------------------------|---|
| ① Temperature fields and slider bar | ⑤ Preheat option |
| ② Time fields | ⑥ Two Step or Three Step cycling option |
| ③ Cycles field | ⑦ Two Stage PCR option |
| ④ RNA-RT option | ⑧ Channel selection options (with corresponding dyes) |

Parameter	Actions
① Temperature	To modify the temperature in one or more steps of the PCR protocol, perform one of the following actions for each step that you want to modify. • Enter a value in the temperature field. • Adjust the temperature by dragging the slider bar up or down.
② Dwell times	Enter time in mm:ss format for each step that you want to modify.
③ Cycles	Set the number of cycles by entering a value into the Cycles field.
④ RNA-RT	<i>(RNA samples only)</i> To add an extra temperature step for RNA reverse transcription to cDNA, select RNA-RT , then adjust the temperature and dwell time as needed.

(continued)

Parameter	Actions
⑤ Preheat	To add a preheat step (sometimes called "hot start"), select Preheat then adjust the temperature and dwell time as needed. Preheating the samples before PCR helps to decrease non-specific binding at lower temperatures.
⑥ Two or three-step cycling	Select Two Step or Three Step cycling from the dropdown list to add or remove an additional cycling step. Adjust the temperature and dwell time as needed.
⑦ Two-stage PCR cycle	To add a second PCR cycle stage, select Two Stage PCR , then adjust the temperature and dwell times as needed.

3. Set the optical channels by toggling the channels on or off (see ⑧).

Note: For optimal performance, select only the channels intended for use and disable any channels that are not in use.

IMPORTANT! Disabling optical channels prevents data from being collected from those channels. Any existing sample groups using these channels are affected. To include a run in a study, all runs must have the same optical collection settings. For information about studies, see "Multi-plate analysis (studies)" on page 107.

4. (Optional) Import an existing protocol.
 - a. Click **IMPORT PROTOCOL**, then click **IMPORT FILE** to open the **File browser**, then navigate to the location of the AQUA file to import.
 - b. Select the file, then click **Open**.
5. (Optional) Export the protocol.
 - a. Click **EXPORT PROTOCOL** to open the **File browser**, then navigate to the location where you want to save the exported protocol AQUA file.
 - b. Click **Export**.
6. (Optional) Create a copy of the current template with a new name.
 - a. In the template parameters screen, click **MAKE A COPY** in the top right corner.
 - b. When prompted, enter the new name in the **Template name** field, then click **MAKE COPY**.
7. In the top-right corner of the screen, click **SAVE** to save the changes.
8. (Optional) To create multiple runs from this template, click **GENERATE BATCH RUNS**.
For more information, see "Generate batch runs from a template" on page 51.

Note: Although batch runs can be created from the **PROTOCOL** tab, this task is typically done after plate setup is complete.

Proceed to the **SETUP** tab to continue with template creation by configuring the MAP plate. For more information, see "SETUP tab (templates)" on page 44.

SETUP tab (templates)

The **SETUP** tab in the template creation or template editing screen supports the following tasks for setting up the MAP plate. For detailed instructions, see “Define or edit plate settings (SETUP tab—templates)” on page 45

- **Customize sample names**—Sample names are user-assigned identifiers for the contents of each loaded well of a plate.
- **Set the dilution factor (DF) denominator**—The dilution factor is the total dilution from the sample to the reaction mix. The value entered in the DF field is the denominator of the dilution factor and must be ≥ 1 .

For example, if 2 μL of the sample is put into a 10 μL reaction mix, the dilution factor is $[2 \mu\text{L sample} / 10 \mu\text{L reaction} = 1/5]$. In this case, the dilution factor denominator that you will enter is 5. If the stock solution is diluted by a factor of 100, then 2 μL of the diluted sample is used in a 10- μL reaction mix, then the dilution factor is $[(1 \mu\text{L stock} / 100 \mu\text{L dilution}) \times (2 \mu\text{L dilution} / 10 \mu\text{L reaction}) = 1/500]$. In this case, the dilution factor denominator that you will enter is 500.

Note: The dilution factor field accepts scientific notation, for example 10×10^{10} .

- **Assign samples to groups**—Groups determine what type of analysis is applied to all samples within a group.
- **ADD GROUP**—Create more groups. See “Manage groups” on page 70.
- **EDIT DYES**—Reassign the dye for the green channel.
- **Export plate setup**—Use to export a blank or configured plate setup CSV file to modify outside of the QuantStudio™ Absolute Q™ Digital PCR Software, then import for use.
- **Import plate setup**—Use to import plate setup files.
- **GENERATE BATCH RUNS**—Create multiple runs containing plate-specific information, such as a barcode or sample information from a template. Batch runs can be used both in non-automated and automated configurations. See “Generate batch runs from a template” on page 51.
- **MAKE A COPY**—Create a copy of the template.
- **ADD NOTE**—Add notes regarding the plate setup.

← New Template
ID: 1f234990-d6ae-40df-a6d8-bc22884c45a1

PROTOCOL **SETUP**

Sample group: Sample group Dilution factor: Dilution factor APPLY TO SELECTION

A	☒ Sample 1 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 5 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 9 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 13 Unknown Dilution factor (DF) = 1/1 Group 1
	☒ Sample 2 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 6 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 10 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 14 Unknown Dilution factor (DF) = 1/1 Group 1
B	☐ Sample 3 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 7 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 11 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 15 Unknown Dilution factor (DF) = 1/1 Group 1
	☐ Sample 4 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 8 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 12 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 16 Unknown Dilution factor (DF) = 1/1 Group 1

Sample groups: EDIT DYES + ADD GROUP

- Group 1
- Group 2
- Group 3

Notes
Enter note... ADD NOTE

Figure 13 SETUP tab for templates

Define or edit plate settings (SETUP tab—templates)

Before you begin, create a template or open an existing template to edit.

- Create a template (page 40)
- Edit a template (page 48)

1. Open the **SETUP** tab.
2. Modify the plate settings as needed.

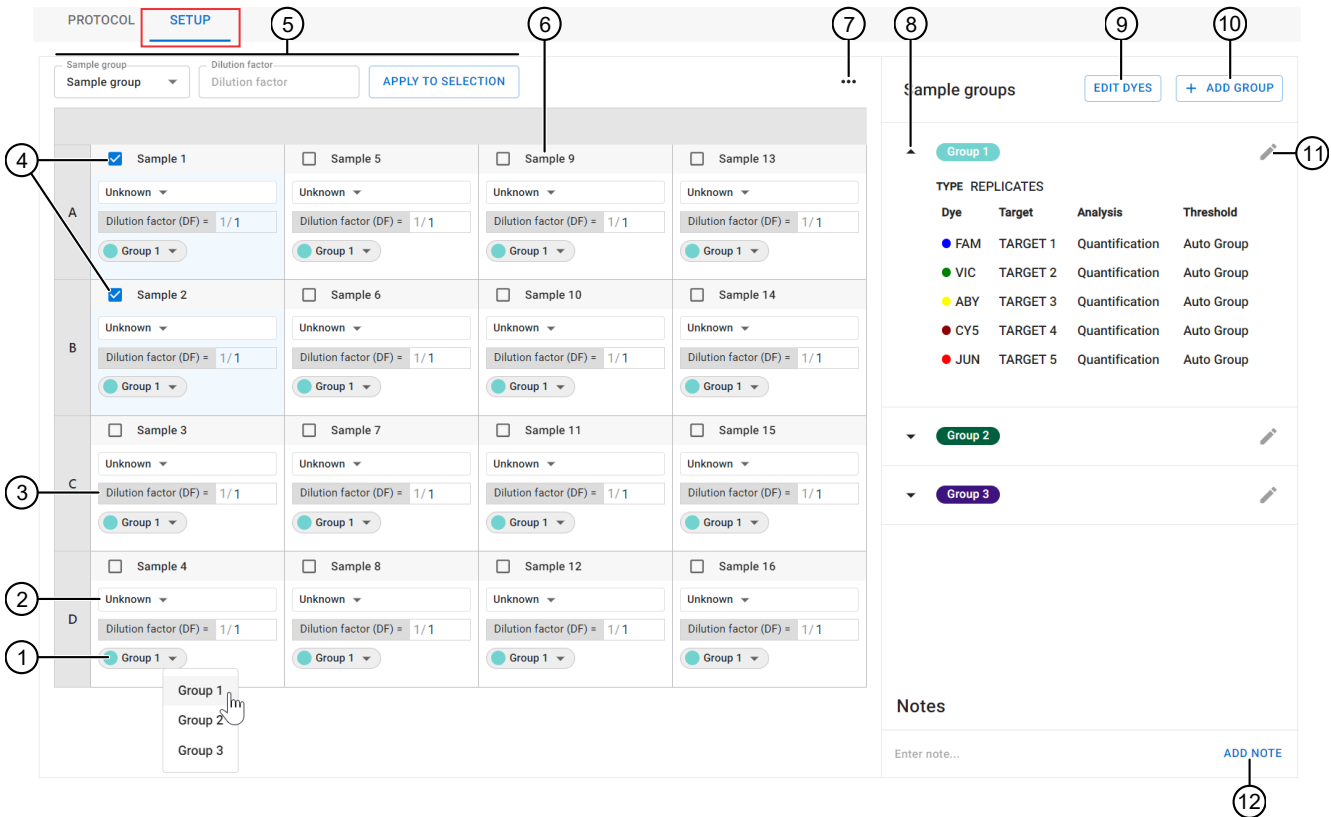


Figure 14 Plate SETUP settings

- | | |
|---|--|
| (1) Group assigned to sample | (7) Import or export plate setup |
| (2) Sample type assigned to sample (view or select) | (8) View group details |
| (3) Sample dilution factor | (9) Edit dye assignment to the green channel |
| (4) Select samples for batch update | (10) Add a sample group |
| (5) Update selected samples | (11) Edit a sample group |
| (6) Sample name (view or edit) | (12) Add notes |

Setting	Action
Sample name	Sample wells are preconfigured with a default name. To change the name, click the sample name, then enter a new name in the field (see ⑥).
Sample type	By default, all samples are assigned an Unknown sample type. Use the sample type dropdown list to assign Unknown , Positive Control , or Negative Control (no template control) sample type to a sample (see ②). Note: Assigning a sample type does not affect the results. The sample type is an attribute of the sample, and therefore only one sample type can be assigned for each sample. The sample type appears in the Analysis screen (samples table), Results ▶ Quantification screen, as well as the Plate Setup and Results sections of the PDF report.

(continued)

Setting	Action
Dilution factor (DF)	Sample wells are preconfigured with a dilution factor of 1. You can change the dilution factor for each sample individually or for multiple samples at once. - For a single sample: enter a value in the Dilution factor field of the sample (see ③). - For multiple samples: select the samples in the sample plate layout, enter a value in the Dilution factor field above the table, then click APPLY TO SELECTION (see ④ and ⑤).
Add a sample group	- Click + ADD GROUP (see ⑩). - In the Add sample group dialog, enter Group name. - Select Individual, Replicates, or Pooling. - Use the switch to toggle channels on or off, then enter the target name into the Target field. - For each active channel, select the analysis type from the Analysis dropdown list. - If you selected CNV Ref as your analysis type, enter the number of copies into the Copies field. - If you selected Variant as your analysis type, select the wild-type reference from the Wild-type ref field. - For each active channel, select the default threshold from the Default threshold dropdown list. If you selected Auto Group or Manual as your default threshold, enter the desired value into the Threshold Guide or Manual value field, respectively. - Click CONTINUE to add the sample group and close the dialog. Alternatively, click ADD & CONTINUE, to add another sample group. For more information, see “Manage groups” on page 70 and “Create groups” on page 72.
Edit or remove a sample group	To determine if you need to edit a sample group, you can view sample group Dye, Target, Analysis, and Threshold information by expanding the group in the Sample groups list (see ⑧). In the Sample groups list, click  (Edit) in the row of a group (see ⑪). - To edit a group—in the Edit sample group dialog, modify group properties, then click CONTINUE. - To delete a group—in the Edit sample group dialog, click DELETE SAMPLE GROUP. For more information, see “Manage groups” on page 70 and “Create groups” on page 72.
Assign samples to groups	By default, all sample wells are assigned to the preconfigured Group 1. You can assign each sample to a group individually or assign multiple samples to the same group at once. - For a single sample: use the Group dropdown list in the sample plate layout to select a new group (see ①). - For multiple samples: select the samples in the sample plate layout, select a new group from the Sample group dropdown list above the table, then click APPLY TO SELECTION (see ④ and ⑤).
EDIT DYES	In the Sample groups list, click EDIT DYES, then use the Dye dropdown list to change the dye assignment for the Green channel. Click CONTINUE (see ⑨).

3. (Optional) Import or export the plate setup. Click ... **(Actions)** in the upper-right corner of the sample plate layout to access the **Import plate setup** and **Export plate setup** options (see ⑥).

Option	Actions
Import plate setup —Import an existing plate setup. You can use the imported plate setup as-is, or modify as needed.	1. Click ... (Actions), then click Import plate setup. 2. In the File browser, navigate to the location of the plate setup CSV file to be imported. 3. Select the plate setup file, then click Open.
Export plate setup —Export the plate setup file to modify or use as-is in other templates or runs.	1. Click ... (Actions), then click Export plate setup. 2. In the File browser, navigate to the location of where to export the plate setup CSV file. 3. Click Save.

4. (Optional) In the **Notes** pane, in the **Enter note** field, enter any notes related to this plate setup, then click **ADD NOTE** (see ⑪).
You can edit or remove existing notes if needed.
5. In the top-right corner of the screen, click **SAVE** to save the protocol and MAP plate configurations.
6. (Optional) Create a copy of the current template with a new name.
- a. In the template parameters screen, click **MAKE A COPY** in the top right corner.
 - b. When prompted, enter the new name in the **Template name** field, then click **MAKE COPY**.
7. (Optional) Click **GENERATE BATCH RUNS** to create multiple runs for this template. See “Generate batch runs from a template” on page 51.

Edit a template

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. If needed, use the search field to find a template.
3. In the list of templates, click the template name, then in the **Select action** dialog, click **Edit template ▶ EDIT TEMPLATE**.
The template opens to the **PROTOCOL** tab.
4. Modify template parameters in the **PROTOCOL** and the **SETUP** tabs as needed.
 - To modify the PCR parameters in the **PROTOCOL** tab, see “PROTOCOL tab (templates)” on page 41.
 - To modify the MAP plate configuration in the **SETUP** tab, see “SETUP tab (templates)” on page 44.
5. In the top-right corner of the screen, click **SAVE** to save the protocol and MAP plate configurations.

Import templates

Use the **IMPORT TEMPLATES** option to import existing templates.

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. In the top right corner, click **IMPORT TEMPLATES**.
3. In the **Import templates** dialog, import one or more DPT files using one of the following import options.

Option	Action
Drag-and-drop template files from a location in your file system.	Using File Explorer , navigate to the DPT file location in your file system, then drag-and-drop the file into the Import templates dialog box.
Browse to locate and add the files.	1. In the Import templates dialog, click IMPORT FILES. 2. In the File browser, navigate to the location of the template DPT file or files to be imported. 3. Select one or more template files, then click Open.

4. When the import completes, in the **Import Status** dialog, click **CLOSE**.
If there are templates that cannot be imported, an error appears in the **Import Status** dialog. Make note of the error so that you can take corrective action on the template, then click **CLOSE** to continue.

The imported templates are added to the **Templates** list.

Export templates

Use the **EXPORT** feature to export templates for use on other Absolute Q™ systems or software installed independently from the instrument, including the AutoRun Suite.

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. If needed, use the search field to find a template.
3. Select the template from the list, then click **EXPORT**.
4. In the **File browser**, navigate to the location on your file system where you want to export the template file or files, select the folder, then click **Select Folder**.

Note: If you are using the AutoRun Suite, export the run template (DPT) files to the D:\Users\Public\Documents\Thermo Scientific\Automation Folder\DPT Templates folder on the Momentum™ computer.

5. When the export completes, click **CLOSE** in the **Export Status** dialog.

Note: If there are templates that cannot be exported, an error appears in the **Status** column. Make note of the error so that you can take corrective action on the template, then click **CLOSE** to continue.

Rename a template

Only one template can be renamed at a time.

Note: To create a copy of an existing template with a new name, use the **MAKE A COPY** option in the template editing workflow (see “Edit a template” on page 48).

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. If needed, use the search field to find a template.
3. Select the template that you want to rename, then click **RENAME**.
4. In the **Rename template** dialog, in the **Template name** field, enter the new name, then click **RENAME**.

The **Templates** list is updated to reflect the new template name.

Delete templates

Multiple templates can be deleted at a time.

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. If needed, use the search field to find a template.
3. From the templates list, select one or more templates, then click **DELETE**.
4. In the **Delete template** dialog, confirm that the selected template or templates are to be deleted by clicking **DELETE TEMPLATE** (if a single template was selected) or **DELETE [# of templates selected] TEMPLATES** (if multiple templates were selected). If you do not want to delete the selected templates, click **DO NOT DELETE**.

Create a run from a template

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. If needed, use the search field to find a template.
3. Click the template name to open the **Select action** dialog, then select **Create run from template**.
4. In the **Run name** field, enter a name for the run, then click **CREATE RUN**.
The **PROTOCOL** tab opens.

Review the **PROTOCOL** and **SETUP** tabs and, if needed, make changes to the protocol and MAP plate configurations. For more information, see “PROTOCOL tab (draft runs)” on page 55 and “SETUP tab (templates)” on page 44.

Generate batch runs from a template

Use the **GENERATE BATCH RUNS** feature to create multiple runs from the same template. These runs can be performed manually or with an automated system that uses a robot to load the plates into the instrument. Each batch run is generated from a single template.

When using an automated system, when the batch run is complete, you must import the run results generated on the automated system into the QuantStudio™ Absolute Q™ Digital PCR Software for analysis.

After you determine how many runs you want to generate from the template, you must create sample assignment files and/or plate barcode files that define plate information for each plate in the batch run.

Note:

- You can use a combination of sample assignment files and plate barcode files in a batch run if the files do not contain duplicate barcode information.
- Examples files can be downloaded from the **Generate batch runs** dialog.

File type	Description
Sample assignment file	• A 2-column CSV file that defines the well number and associated sample name for each well on the plate • Each CSV file represents one plate which corresponds to one run IMPORTANT! If you are using the AutoRun Suite to run batches, the sample assignment file name must include the barcode number. For information about automated workflows, see Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.
Plate barcode file	• A 1-column TXT file that defines the barcode numbers for plates in the run. • The sample names for each well are inherited from the template.

1. In the left navigation pane, click  **Templates** to open the **Templates** screen.
2. If needed, use the search field to find a template.

3. Open the **Generate batch runs** dialog in one of the following ways.

Option	Description
If using the template as-is	Select a template from the list, then click GENERATE BATCH RUNS .
To modify the template before use	1. Click the template name, then in the Select action dialog, click Edit template ► EDIT TEMPLATE 2. In the PROTOCOL and SETUP tabs, make changes to the template as needed, then click SAVE. For more information, see “PROTOCOL tab (draft runs)” on page 55 and “SETUP tab (templates)” on page 44 3. Click GENERATE BATCH RUNS.

Generate batch runs dialog opens, the name of the selected template appears in the **Select template file** field. If needed, you can select a different template at this point.

4. Use the following options to define plate information for the batch run.

Option	Description
ADD BARCODES & SAMPLE ASSIGNMENT FILES —Defines sample name by well.	1. Click ADD BARCODES & SAMPLE ASSIGNMENT FILES. 2. Use the File browser to locate and select the sample assignment CSV files for the plates for this batch run, then click Open. IMPORTANT! If you are using the AutoRun Suite, the sample assignment file name must include the barcode number for the system to find the plate. Failure to include the barcode prevents the run from starting. For information about automated workflows, see Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.
ADD PLATE BARCODE FILE —Defines the barcode numbers of plates used in the run.	1. Click ADD PLATE BARCODE FILE. 2. Use the File browser to locate and select the barcode TXT files for the plates for this batch run, then click Open.

The **Generate batch runs** dialog opens with information from the CSV or TXT files.

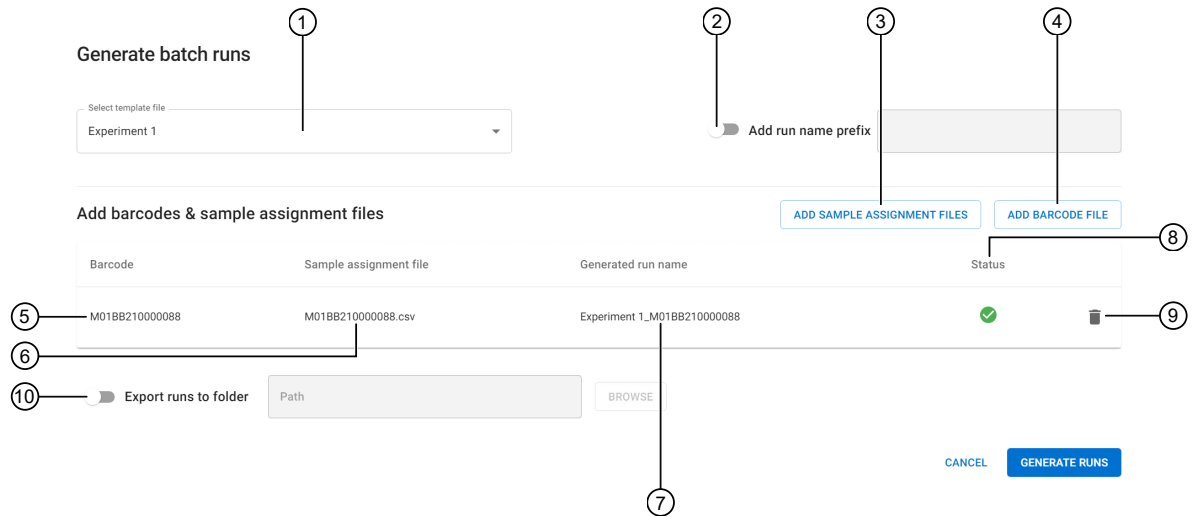


Figure 15 Generate batch runs dialog

5. In the **Generate batch runs** dialog, review or modify run details as needed using the following actions.

Callout	Action	Description
①	Confirm template selection or select another template.	To change the template, select a template from the Select template file dropdown list.
②	Add a prefix to the run name.	To add a prefix to the run name, toggle on the Add run name prefix option, then enter the desired text in the field.
③, ④	Add more plates to a run.	Click ADD SAMPLE ASSIGNMENT FILES or ADD BARCODE FILE , select a file from your file browser, then click Open . ADD SAMPLE ASSIGNMENT FILES option adds more plates to the run using the sample CSV file format. ADD BARCODE FILE option adds more plates to the run using the barcode TXT file format. For more information, see step 4.
⑤	Review the barcode, sample assignment file, and generated run name.	Barcode—Plate barcode number of the plate. Sample assignment file—If imported from sample assignment file, the name of the file includes the path to the location of the file on the network. If imported from barcode file, the name is inherited from the template file. Generated run name—System-assigned run name that contains the format <template_name_barcode>
⑧	View status of each imported file.	✓—Indicates a valid file format. ⚠—Indicates an issue with the file that must be corrected. If present, place the pointer over ⚠ for information about the issue. Take the appropriate corrective action.
⑨	Delete one or more plates from a run.	Click 🗑 (Delete) in the row of a run that you want to delete.

(continued)

Callout	Action	Description
⑩	Export runs to a folder. Note: This feature is used for implementations that use automation. The folder selected for export is the folder from which the automated system can retrieve the information to perform the batch run.	1. Toggle on Export run to folder to designate a location for the batch file information. 2. Click BROWSE, then navigate to the folder on your file system where you want to export runs. 3. Click Select Folder. The Path field is populated with the filepath to the folder. Note: If you are using the AutoRun Suite, export the batch run (DPR) files to the D:\Users\Public\Documents\Thermo Scientific\Automation Folder\DPR Barcoded Templates folder on the Momentum™ computer.

6. Click **GENERATE RUNS** to create the DPR files for each plate.
When the batch run generation is complete, the runs are listed in the **Runs** screen, in the **DRAFT** tab.

- For implementations using automation, completed run data must be imported from the automated system to be used in analysis. For information about importing completed batch run data, see “Import runs” on page 66.

Note: Runs created in the QuantStudio™ Absolute Q™ Digital PCR Software, but performed on an automated system, remain in the **DRAFT** tab of the **Runs** screen with a **DRAFT** status until the run data from the automated system is imported.

- For implementations not using automation, experiments are run one plate at a time. For information about initiating a single-plate run, see “Start a single-plate run” on page 64 .

Manage runs

The **Runs** screen provides information about three run types—**DRAFT**, **COMPLETED**, and **CALIBRATION**.

IMPORTANT! **CALIBRATION** runs are only performed by qualified Thermo Fisher Scientific representatives.

This section provides information about managing **DRAFT** runs. For information about managing **COMPLETED** runs, see “Single-plate analysis (completed runs)” on page 78 and “Multi-plate analysis (studies)” on page 107.

You can perform the following tasks from the **DRAFT** tab.

- View status information for a run.
 - **PRE-RUN**—The run has not been started.
 - **IN PROGRESS**—The run has been started and is not yet complete.
- View information about the draft run, including plate barcode, status of the run, user who created the run, and the dates the run was last modified and exported.

- Open a draft run to modify protocol and plate setup parameters (see “Edit a draft run from the Runs screen” on page 64).
- **CREATE RUN**—Create a new run (see “Create a run from the Runs screen” on page 55).
- **IMPORT RUN**—Import runs created on other Absolute Q™ systems or runs generated on an automated system (see “Import runs” on page 66).
- **START RUN**—Start a run. This function is only available if an instrument is connected to the dedicated computer (see “Start a single-plate run” on page 64).
- **EXPORT**—Export runs for use on other Absolute Q™ systems (see “Export runs” on page 68).
- **RENAME**—Rename a run (see “Rename a run” on page 69).
- **DELETE**—Delete one or more runs (see “Delete runs” on page 69).

Create a run from the Runs screen

Runs are created from templates. If you have not already done so, create a template as described in “Create a template” on page 40.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. In the upper-right corner, click **CREATE RUN**.
3. In the **Create run** dialog, provide the following information, then click **CREATE RUN**.
 - **Run name:** Enter a name for the run.
 - **Template:** Select a template from the list.

The new run opens to the **PROTOCOL** tab.

Review the **PROTOCOL** and **SETUP** tabs and, if needed, make changes to the protocol and MAP plate configurations.

Action	Description
View or edit run protocol	“PROTOCOL tab (draft runs)” on page 55
View or edit MAP plate settings	“SETUP tab (draft runs)” on page 59

PROTOCOL tab (draft runs)

The **PROTOCOL** tab for a draft run (from the **Runs** screen **DRAFT** tab) supports the following tasks when configuring the protocol. For detailed instructions, see “View or edit run protocol (PROTOCOL tab—draft runs)” on page 56.

- Define PCR parameters—Use the controls to set protocol-specific parameters.
- Customize optical channels—Use the toggle switch to enable or disable optical channels.
- **IMPORT PROTOCOL**—Import an existing protocol.
- **EXPORT PROTOCOL**—Export the protocol.

- **SAVE AS TEMPLATE**—Save the run settings as a new template.
- **E-SIGN**—For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software (see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”).



Figure 16 PROTOCOL tab for draft runs

View or edit run protocol (PROTOCOL tab—draft runs)

Before you begin, create or open a draft run.

- Create a run from the Runs screen (page 55)
- Edit a draft run from the Runs screen (page 64)

1. Open the **PROTOCOL** tab.
2. Modify PCR parameters as needed.

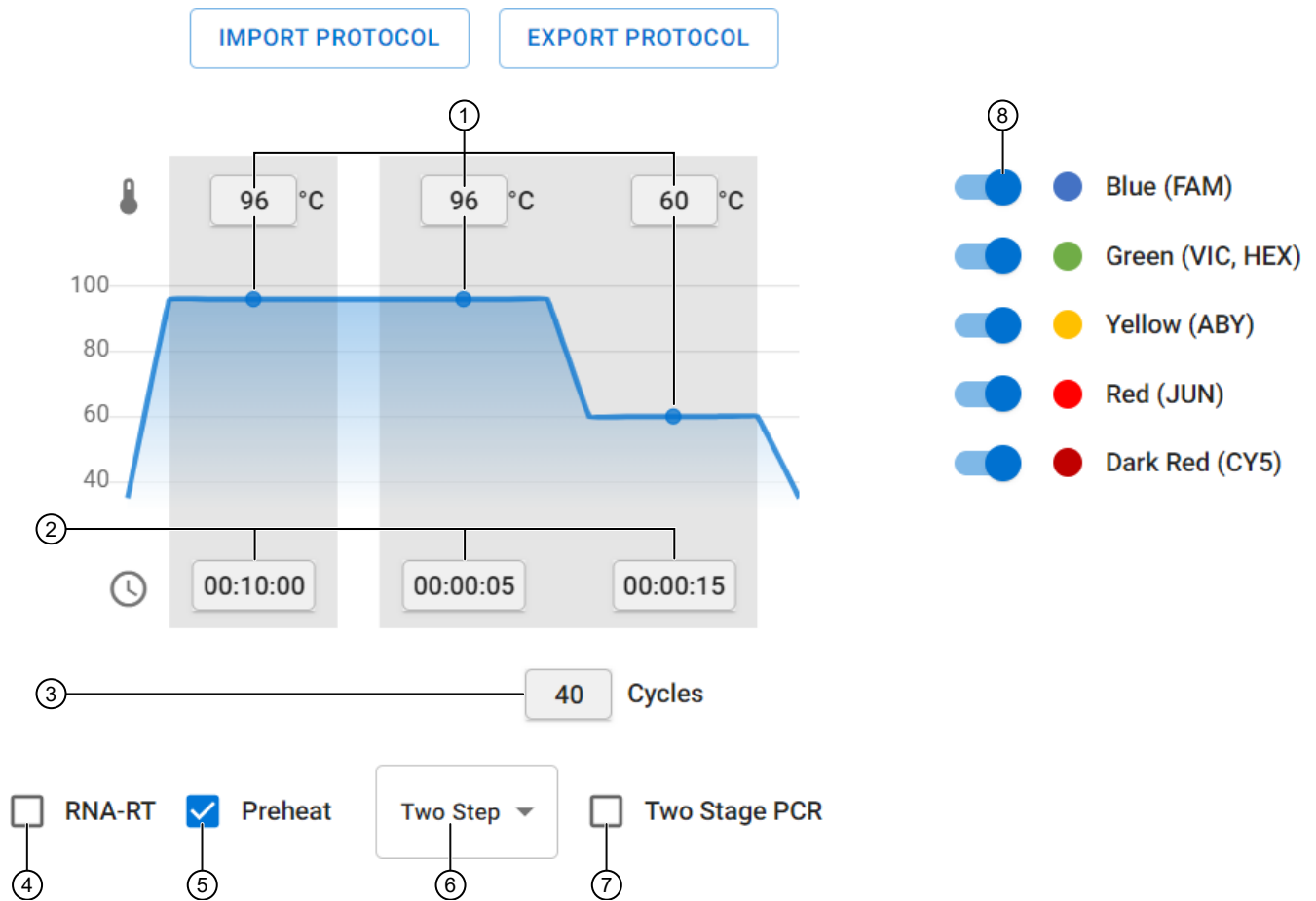


Figure 17 Protocol parameters

- | | |
|-------------------------------------|---|
| ① Temperature fields and slider bar | ⑤ Preheat option |
| ② Time fields | ⑥ Two Step or Three Step cycling option |
| ③ Cycles field | ⑦ Two Stage PCR option |
| ④ RNA-RT option | ⑧ Channel selection options (with corresponding dyes) |

Parameter	Actions
① Temperature	To modify the temperature in one or more steps of the PCR protocol, perform one of the following actions for each step that you want to modify. - Enter a value in the temperature field. - Adjust the temperature by dragging the slider bar up or down.
② Dwell times	Enter time in mm:ss format for each step that you want to modify.
③ Cycles	Set the number of cycles by entering a value into the Cycles field.
④ RNA-RT	<i>(RNA samples only)</i> To add an extra temperature step for RNA reverse transcription to cDNA, select RNA-RT , then adjust the temperature and dwell time as needed.

(continued)

Parameter	Actions
⑤ Preheat	To add a preheat step (sometimes called "hot start"), select Preheat then adjust the temperature and dwell time as needed. Preheating the samples before PCR helps to decrease non-specific binding at lower temperatures.
⑥ Two or three-step cycling	Select Two Step or Three Step cycling from the dropdown list to add or remove an additional cycling step. Adjust the temperature and dwell time as needed.
⑦ Two-stage PCR cycle	To add a second PCR cycle stage, select Two Stage PCR , then adjust the temperature and dwell times as needed.

3. Set the optical channels by toggling the channels on or off (see ⑧).

Note: For optimal performance, select only the channels intended for use and disable any channels that are not in use.

IMPORTANT! Disabling optical channels prevents data from being collected from those channels. Any existing sample groups using these channels are affected. To include a run in a study, all runs must have the same optical collection settings. For information about studies, see “Multi-plate analysis (studies)” on page 107.

4. (Optional) Import an existing protocol.
- Click **IMPORT PROTOCOL**, then click **IMPORT FILE** to open the **File browser**, then navigate to the location of the AQUA file to import.
 - Select the file, then click **Open**.
5. (Optional) Export the protocol.
- Click **EXPORT PROTOCOL** to open the **File browser**, then navigate to the location where you want to save the exported protocol AQUA file.
 - Click **Export**.
6. (Optional) Create a template of the run with a new name.
- Click **SAVE AS TEMPLATE**.
 - When prompted, enter the new template name in the **Template name** field, then click **SAVE AS TEMPLATE**.
7. In the top-right corner of the screen, click **SAVE** to save the changes.
8. (Optional) For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, click **E-SIGN** to provide an e-signature (see “Sign data in the software” on page 170).

Proceed to the **SETUP** tab to continue with run creation by configuring the MAP plate. For more information, see “SETUP tab (draft runs)” on page 59.

SETUP tab (draft runs)

The **SETUP** tab for a draft run (from the **Runs** screen **DRAFT** tab) supports following tasks when setting up the MAP plate. For detailed instructions, see “View or edit plate settings (SETUP tab—draft runs)” on page 60.

- Edit sample names—Sample names are user-assigned identifiers for the contents of each loaded well of a plate.
- Include or exclude plate columns for use in the run.
- Customize sample names—Sample names are user-assigned identifiers for the contents of each loaded well of a plate.
- Set the dilution factor (DF) denominator—The dilution factor is the total dilution from the sample to the reaction mix. The value entered in the DF field is the denominator of the dilution factor and must be ≥ 1 .

For example, if 2 μL of the sample is put into a 10 μL reaction mix, the dilution factor is $[2 \mu\text{L sample} / 10 \mu\text{L reaction} = 1/5]$. In this case, the dilution factor denominator that you will enter is 5. If the stock solution is diluted by a factor of 100, then 2 μL of the diluted sample is used in a 10- μL reaction mix, then the dilution factor is $[(1 \mu\text{L stock} / 100 \mu\text{L dilution}) \times (2 \mu\text{L dilution} / 10 \mu\text{L reaction}) = 1/500]$. In this case, the dilution factor denominator that you will enter is 500.

Note: The dilution factor field accepts scientific notation, for example 10E+10.

- Assign samples to groups—Groups determine what type of analysis is applied to all samples within a group.
- **ADD GROUP**—Create more groups (see “Manage groups” on page 70).
- **EDIT DYES**—Reassign the dye for the green channel for this run.
- **Export plate setup**—Export a blank or configured plate setup CSV file to modify outside of the QuantStudio™ Absolute Q™ Digital PCR Software, then import for use.
- **Import plate setup**—Import plate setup files.
- **Plate barcode**—Assign the barcode of the plate used for the run.
- **ADD NOTE**—Add notes regarding the run.
- **SAVE AS TEMPLATE**—Save the run settings as a new template.
- **E-SIGN**—For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”.

← New Run
ID: 0748b83f-6a02-44ad-85a4-973715dd2373

PROTOCOL **SETUP**

SAVE AS TEMPLATE START RUN SAVE

	1	2	3	4
A	☐ Sample 1 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 5 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 9 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 13 Unknown Dilution factor (DF) = 1/1 Group 1
B	☐ Sample 2 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 6 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 10 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 14 Unknown Dilution factor (DF) = 1/1 Group 1
C	☐ Sample 3 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 7 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 11 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 15 Unknown Dilution factor (DF) = 1/1 Group 1
D	☐ Sample 4 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 8 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 12 Unknown Dilution factor (DF) = 1/1 Group 1	☐ Sample 16 Unknown Dilution factor (DF) = 1/1 Group 1

Plate barcode

Sample groups

EDIT DYES + ADD GROUP

- Group 1
- Group 2
- Group 3

Notes

Enter note... ADD NOTE

Figure 18 SETUP tab for draft runs

View or edit plate settings (SETUP tab—draft runs)

Before you begin, create or open a draft run.

- Create a run from the Runs screen (page 55)
- Edit a draft run from the Runs screen (page 64)

1. Open the **SETUP** tab.
2. (Optional) Modify the plate settings.

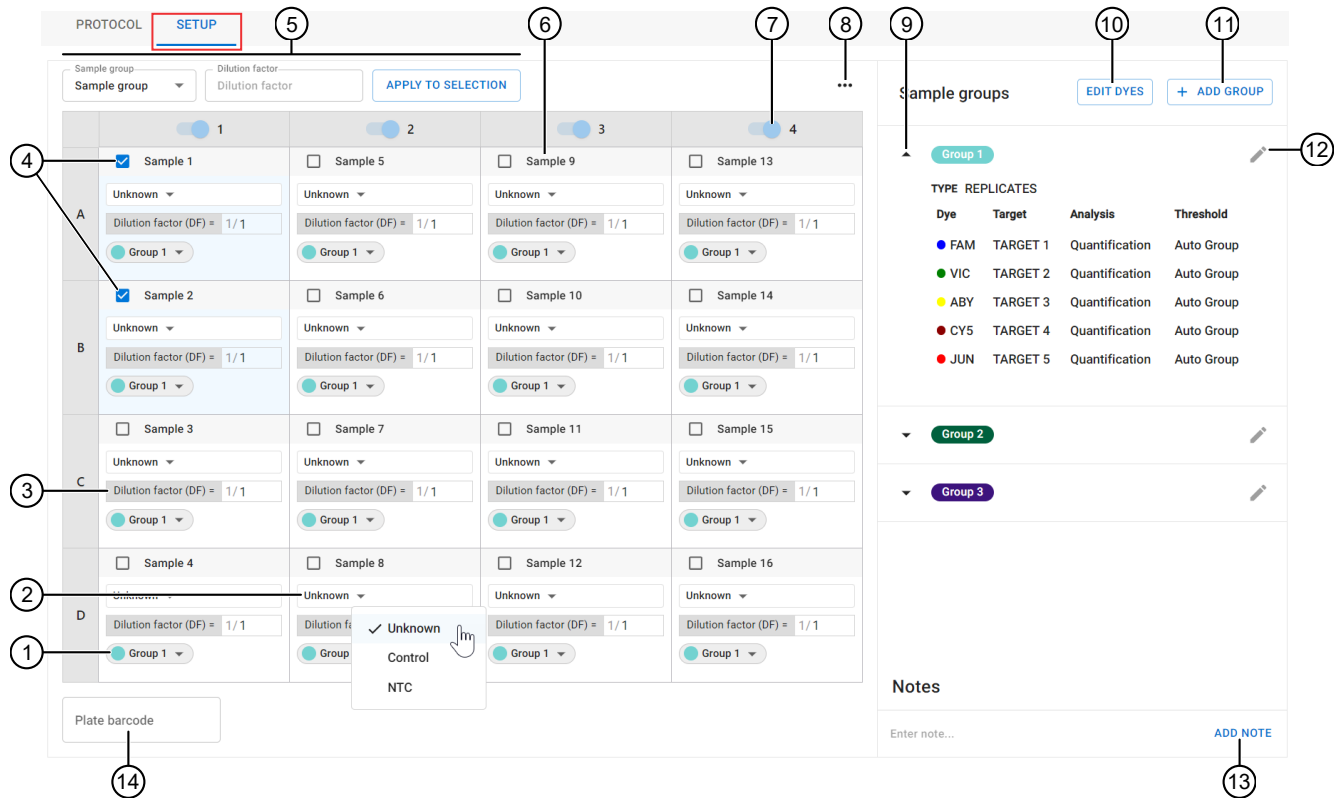


Figure 19 Plate SETUP settings

- | | |
|--|---|
| ① Group assigned to sample | ⑧ Import or export plate setup |
| ② Sample type assigned to sample (view or select) | ⑨ View group details |
| ③ Sample dilution factor | ⑩ Edit dye assignment to the green channel |
| ④ Select samples for batch update | ⑪ Add a sample group |
| ⑤ Update selected samples | ⑫ Edit a sample group |
| ⑥ Sample name (view or edit) | ⑬ Add notes |
| ⑦ Toggle columns on or off to for inclusion in a run | ⑭ Enter the plate barcode to associate the plate with the run |

Setting	Action
Sample name	Sample wells are preconfigured with a default name. To change the name, click the sample name, then enter a new name in the field (see ⑥).
Sample type	By default, all samples are assigned an Unknown sample type. Use the sample type dropdown list to assign Unknown , Positive Control , or Negative Control (no template control) sample type to a sample (see ②). Note: Assigning a sample type does not affect the results. The sample type is an attribute of the sample, and therefore only one sample type can be assigned for each sample. The sample type appears in the Analysis screen (samples table), Results ▶ Quantification screen, as well as the Plate Setup and Results sections of the PDF report.

(continued)

Setting	Action
Dilution factor (DF)	Sample wells are preconfigured with a dilution factor of 1. You can change the dilution factor for each sample individually or for multiple samples at once. - For a single sample: enter a value in the Dilution factor field of the sample (see ③). - For multiple samples: select the samples in the sample plate layout, enter a value in the Dilution factor field above the table, then click APPLY TO SELECTION (see ④ and ⑤).
Add a sample group	- Click + ADD GROUP (see ①). - In the Add sample group dialog, enter Group name. - Select Individual, Replicates, or Pooling. - Use the switch to toggle channels on or off, then enter the target name into the Target field. - For each active channel, select the analysis type from the Analysis dropdown list. - If you selected CNV Ref as your analysis type, enter the number of copies into the Copies field. - If you selected Variant as your analysis type, select the wild-type reference from the Wild-type ref field. - For each active channel, select the default threshold from the Default threshold dropdown list. If you selected Auto Group or Manual as your default threshold, enter the desired value into the Threshold Guide or Manual value field, respectively. - Click CONTINUE to add the sample group and close the dialog. Alternatively, click ADD & CONTINUE, to add another sample group. For more information, see “Manage groups” on page 70 and “Create groups” on page 72.
Edit or remove a sample group	To determine if you need to edit a sample group, you can view sample group Dye, Target, Analysis, and Threshold information by expanding the group in the Sample groups list (see ⑨). In the Sample groups list, click  (Edit) in the row of a group (see ⑫). - To edit a group—in the Add sample group dialog, modify group properties, then click CONTINUE. - To delete a group—click DELETE SAMPLE GROUP. For more information, see “Manage groups” on page 70 and “Create groups” on page 72.
Assign samples to groups	By default, all sample wells are assigned to the preconfigured Group 1. You can assign each sample to a group individually or assign multiple samples to the same group at once. - For a single sample: use the Group dropdown list in the sample plate layout to select a new group (see ①). - For multiple samples: select the samples in the sample plate layout, select a new group from the Sample group dropdown list above the table, then click APPLY TO SELECTION (see ④ and ⑤).

(continued)

Setting	Action
EDIT DYES	In the Sample groups list, click EDIT DYES , then use the Dye dropdown list to change the dye assignment for the Green channel. Click CONTINUE (see ⑩).
Associate a specific plate with the run	In the Plate Barcode field, enter the barcode for the plate associated with this run (see ⑭). Note: The instrument scans the plate for the barcode when the run is initiated. If your process is to ensure that a specific plate is used for this run, enter the barcode for the plate here. If the barcode entered here does not match the scanned number, the software returns an error. If this occurs, load the correct plate or return to the SETUP tab to correct the barcode number. IMPORTANT! If this run file is going to be used as part of a DPR Run in AutoRun, you must enter a barcode. See Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.

3. (Optional) Import or export the plate setup. Click ... **(Actions)** in the upper-right corner of the sample plate layout to access the **Import plate setup** and **Export plate setup** options (see ⑦).

Option	Actions
Import plate setup —Import an existing plate setup. You can use the imported plate setup as-is, or modify as needed.	1. Click ... (Actions), then click Import plate setup. 2. In the File browser, navigate to the location of the plate setup CSV file to be imported. 3. Select the plate setup file, then click Open.
Export plate setup —Export the plate setup file to modify or use as-is in other templates or runs.	1. Click ... (Actions), then click Export plate setup. 2. In the File browser, navigate to the location of where to export the plate setup CSV file. 3. Click Save.

4. In the sample plate pane, at the top of each column of the plate, use the toggle switch to include or omit a column from the run (see ⑥).
- Toggle the switch to the on position to include a column in the run.
 - Toggle the switch to the off position to omit the column from the run.

IMPORTANT!

- Failure to omit the columns that are not in use prevents them from being used in a subsequent run.
- This setting cannot be changed after the run is completed. Ensure that columns are defined correctly before you start a run.

5. (Optional) In the **Notes** pane, in the **Enter note** field, enter any notes related to this plate setup, then click **ADD NOTE** (see ⑫).
- You can edit or remove existing notes if needed.
6. (Optional) Create a template of the current run parameters.
- a. Click **SAVE AS TEMPLATE**.

- b. When prompted, type the new name in the **Template name** field, then click **SAVE AS TEMPLATE**.
7. In the top-right corner of the screen, click **SAVE** to save the changes.
8. (Optional) For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, click **E-SIGN** to provide an e-signature (see “Sign data in the software” on page 170).
9. (Optional) Click **START RUN** to start the run.

Note: This function is only available if the desktop computer is connected to an instrument.

Edit a draft run from the Runs screen

For runs that are in the **DRAFT** tab of the **Runs** screen with a **PRE-RUN** status, you can edit both – the PCR protocol and MAP plate setup.

Note: Protocol parameters cannot be modified for **COMPLETED** runs. Protocol parameters can only be modified for draft runs with a **PRE-RUN** status.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. Click **DRAFT** to open the list of draft runs.
3. (Optional) Use the search field to find a run.
4. Click a run name to open the run protocol and MAP plate configuration, then modify as needed.
 - To modify **PROTOCOL** parameters on a draft run, see “PROTOCOL tab (draft runs)” on page 55.
 - To modify MAP plate configuration, see “SETUP tab (draft runs)” on page 59.
5. In the top-right corner of the screen, click **SAVE** to save the run protocol and MAP plate configuration.

Start a single-plate run

This procedure describes how to start a single-plate run in a non-automated configuration. For information about starting runs using the AutoRun Suite, see Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.

The **START RUN** function is only available when the dedicated computer is connected to an instrument.

IMPORTANT!

- You must inspect and clean the plate nest before each run. See “Clean the Absolute Q™ instrument and plate nest” on page 176.
 - Before running the protocol, ensure that protocol parameters are defined correctly. Protocol parameters cannot be changed after the run. See “PROTOCOL tab (draft runs)” on page 55.
-

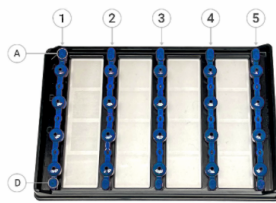
1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. If needed, use the search field to locate a run.
3. Start a run using one of the following options.

Option	Description
Start a run from the Runs screen.	In the DRAFT tab, select a run from the list, then click START RUN .
Start a run from the Runs ► SETUP screen.	In the Runs screen, click a run name to open a run, then from the SETUP tab of the run, click START RUN .

The **Start run** dialog box opens and the instrument door opens to receive the loaded MAP plate.

IMPORTANT! Ensure that gaskets are placed on all columns of the MAP plate, including unused columns. Ensure that gaskets have been placed on all wells and on the column X posts (see ⑤) as shown on the screen. Failure to do so can produce poor results.

FastTest



1. Put all five gaskets on the plate



2. Place the plate on the tray

CLOSE DOOR

- Columns 1–4 at locations ①–④ and column X at location ⑤
- Rows A–D at locations ①–④

4. In the **Plate barcode** field, add the barcode number of the MAP plate.
5. Carefully load the MAP plate in the plate nest.

IMPORTANT! Load the MAP plate gently to avoid damage to the plate nest.

6. Click **CLOSE DOOR**, then click **START RUN**.

The door closes and the MAP plate barcode is scanned.

Note: If the barcode number does not match the number entered, or the instrument cannot scan the barcode, you are prompted to add it in the **Plate barcode** field of the **Start Run** dialog.

When the run has successfully started, the **Runs** screen returns to the **DRAFT** tab and the status of the selected run is updated to **IN PROGRESS**.

7. (Optional) To check the status of the run, in the left navigation pane, click  **Instrument** to open the **Instrument** screen.

The status bar to the right of the run name displays the progress of the run.

TFSABSQ123456789
RUNNING
STOP

TIME STARTED 12:13 PM	ESTIMATED TIME REMAINING 45 MINUTES	USER LAB OPERATOR	PLATE BARCODE CXUY112	KEY VALUE
--------------------------	--	----------------------	--------------------------	--------------

RUN_NAME

✓

2

3

Prime 100%
PCR 50%
Scan 0%



SERIAL NUMBER
TFSABSQ123456789

INSTRUMENT SOFTWARE
1.0.41

INSTRUMENT FIRMWARE
0.9.3

LAST CALIBRATION DATE
AUG 10, 2022

DESKTOP SOFTWARE
6.3.0

IMPORTANT! When using the AutoRun Suite, the Absolute Q™ dedicated computer is only used for instrument control and troubleshooting. Do not use the Absolute Q™ software on the Absolute Q™ dedicated computer when an AutoRun is processing in the Momentum™ software. Doing so may interfere with the AutoRun system.

Import runs

Use the **IMPORT RUN** feature to import runs created on other Absolute Q™ systems or runs generated on an automated system. Runs can be imported from either the **DRAFT** or **COMPLETED** tabs. Completed runs have a ZST file extension.

Note: A run can be imported more than once. Each time the run is imported, it is treated as a separate run and does not overwrite the previously imported data.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. Click **IMPORT RUN**.

3. In the **Import runs** dialog, perform one of the following actions.

Action	Description
Drag-and-drop run files from a location on your file system.	Using File Explorer , locate the DPR, ZIP, or ZST file or files in your file system, then drag-and-drop the file into the Import runs dialog.
Browse to locate and add the run files.	1. In the Import runs dialog, click IMPORT FILES. 2. In the File browser, navigate to the location of the DPR, ZIP, or ZST file or files to be imported. 3. Select one or more run files, then click Open.

4. When the import completes, in the **Import Status** dialog, click **CLOSE**.

Note: If there are runs that cannot be imported, an error appears in the **Import Status** dialog. Make note of the error so that you can take corrective action on the run, then click **CLOSE** to continue.

The imported runs are added to the **Runs** list.

Export runs

Use the **EXPORT** feature to export run files for use on other Absolute Q™ systems, including the AutoRun Suite. Run files can be exported from either the **DRAFT** or **COMPLETED** tabs.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. Use the following options to export runs that have not been started from the **DRAFT** tab or completed runs from **COMPLETED** tab.

Option	Description
Export draft run files. Note: • Runs that have not been run have a DPR file extension. • Draft run files are used as part of the DPR Run workflow for AutoRun. See Appendix D, “Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite”.	1. In the DRAFT tab, use the search field to find the runs for export, then select one or more runs from the list. 2. Click EXPORT. 3. In the File browser, navigate to the location where you want to export the run file or files, then click Select Folder.
Export completed run files. Note: Completed runs have a ZST file extension.	1. In the COMPLETED tab. 2. Use the search field to find the runs for export, or click  to filter the runs by STATUS, INSTRUMENT, or STUDY-COMPATIBLE RUNS. 3. Select one or more runs. 4. Click EXPORT. 5. In the File browser, navigate to the location where you want to export the run file or files. 6. Click Select Folder.

3. When the export completes, in the **Export Status** dialog, click **CLOSE**.

Note: If there are runs that cannot be exported, an error appears in the **Export Status** dialog. Make note of the error so that you can take corrective action on the run, then click **CLOSE** to continue.

Rename a run

Only one run can be renamed at a time. Runs can be renamed from either the **DRAFT** or **COMPLETED** tabs.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. Use the following options to rename runs that have not been started from the **DRAFT** tab or runs that have finished from **COMPLETED** tab.

Option	Action
Rename draft run files. Note: Runs that have not been run have a DPR file extension.	1. In the DRAFT tab, use the search field to find the runs to rename, then select a run from the list. 2. Click RENAME.
Rename completed run files. Note: Completed runs have a ZST file extension.	1. In the COMPLETED tab. 2. Use the search field to find a run to rename, or click  to filter the runs by STATUS, INSTRUMENT, or STUDY-COMPATIBLE RUNS. 3. Select a run, then click RENAME.

3. In the **Rename run** dialog in the **Run name** field, enter the new run name, then click **RENAME**.

The **Runs** list is updated to reflect the new run name.

Delete runs

Multiple runs can be deleted at a time. Runs can be deleted from either the **DRAFT** or **COMPLETED** tabs.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
2. Use the following options to delete runs that have not been started from the **DRAFT** tab or completed runs from **COMPLETED** tab.

Option	Action
Delete draft run files.	1. In the DRAFT tab, use the search field to find the runs to delete, then select a run from the list. 2. Click DELETE.
Delete run files.	1. In the COMPLETED tab. 2. Use the search field to find a run to delete, or click  to filter the runs by STATUS, INSTRUMENT, or STUDY-COMPATIBLE RUNS. 3. Select a run, then click DELETE.

3. In the **Delete run** dialog, confirm that the selected run or runs are to be deleted by clicking **DELETE RUN** (if a single run was selected) or **DELETE [# of runs selected] RUNS** (if multiple runs were selected). If you do not want to delete the selected runs, click **DO NOT DELETE**.

Manage groups

Groups are used to define the analysis and results type for reporting of individual samples or sets of samples. After a group has been defined, it can be edited or deleted.

Note: Only groups without samples can be deleted.

When samples are assigned to a group, the samples within the same group acquire the same definitions for the following characteristics.

Sample grouping

Sample grouping	Description
Individual	Each sample has a separate result entry.
Replicates	The results show the Mean , Standard Deviation , and CV% for the concentration for all the samples in the group.
Pooling	The results handle all samples in the group as one large sample.

Optical channel selections

All samples within the same group will have the same set of optical channels enabled.

Target

All samples within the same group will have the same target DNA associated with each optical channel and fluorescent dye.

Analysis

Analysis type	Option	Description
Absolute quantification	Quantification	Determines the concentration of a target molecule.
CNV	CNV Ref (Copy Number Variation Reference)	The reference target for CNV. Note: The reference target is a gene of known and stable copy number used to calculate the copy number for the gene of interest.
	CNV (Copy Number Variation)	Reporting ratio of CNV/CNV Ref. Note: A reference must be selected when using CNV. If CNV is selected for multiple channels, they will share the same reference.
Variant allele detection (Variant Allele Frequency (VAF))	VAF Wild Type	Wild type is treated as a standard (or common) allele reference to detect if the corresponding variant is present in the sample. Note: • A maximum of 2 VAF Wild Type channels are allowed. • Each VAF Wild Type channel must be assigned to at least one corresponding VAF Variant channel.
	VAF Variant	Variant (mutant) allele is present in the sample. Note: Each channel that is assigned to VAF Variant must reference a corresponding VAF Wild Type channel.

Default threshold

Default threshold	Description
Auto Sample	The software automatically assigns the threshold to each sample independently.
Auto Group	The software aggregates the samples in the group and automatically determines a common threshold.
Manual	Manually set a custom threshold for the channel.

Initial threshold values

The **Threshold Guide** and **Manual value** settings are used for establishing the initial thresholds for analyses by Sample Group **Auto Group** or **Manual** threshold types, respectively.

Default threshold selection	Initial threshold guide setting
Auto Group	Enter the expected mean positive signal for the group into the Threshold Guide field. The analysis algorithm uses the Threshold Guide value to calculate the threshold value for the group.
Manual	Enter the desired threshold value into the Manual value field.
Auto Sample	No initial guide value is required. The software automatically assigns the threshold to each sample independently.

Note: **Threshold Guide** and **Manual value** are initial values only and can be modified during analysis (see “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90).

For instructions about how to manage groups, see the following section.

- To create groups, see “Create groups” on page 72.
- To edit groups, see “Edit groups” on page 74.
- To delete groups, see “Delete groups” on page 74.
- To add samples to groups, see “Assign samples to groups” on page 75.

Create groups

Groups can be created as part of template or run creation workflows in the **SETUP** tab, or as part of the editing workflow after a run or a template was created.

For details about each group setting, see “Manage groups” on page 70.

1. Navigate to the list of templates or runs.
 - Click  **Templates** to open the **Templates** screen.
 - Click  **Runs** to open the **Runs** screen.
2. If needed, use the search field to find a run or template.
3. Open a run or a template.
 - In the **Templates** screen, click the name of the template, then in the **Select action** dialog, select **Edit template**, then click **EDIT TEMPLATE**.
 - In the **Runs** screen, click the name of the run.
4. Navigate to the **SETUP** tab for the template or run.
5. In the **Sample groups** pane, click **+ ADD GROUP**.
6. In the **Group name** field, enter a name for the group.

7. Select **Individual**, **Replicates**, or **Pooling** sample grouping option.
8. (Optional) Toggle an optical channel on or off to include or exclude a channel from the sample group.
9. In the **Target** field for each active optical channel, enter the name of the DNA target.
10. From the **Analysis** dropdown list, select one of the following options for each active optical channel, then select or enter additional information required for the selected analysis type.

Analysis type	Option	Description
Absolute quantification	Quantification	Proceed to step 12.
CNV	CNV	If CNV analysis is selected for one or more channel, select one CNV Ref analysis within the sample group.
	CNV Ref	In the Copies field, enter the number of copies, then proceed to step 12.
Variant allele detection (Variant Allele Frequency (VAF))	VAF Wild Type	If one or more channels are assigned to a VAF Variant analysis, assign at least one channel to VAF Wild Type analysis. Note: • A maximum of 2 VAF Wild Type channels are allowed. • Each VAF Wild Type channel must be assigned to at least one corresponding VAF Variant channel.
	VAF Variant	From the Wild-type ref dropdown list, select the corresponding wild type [channel (target)] reference, then proceed to step 11. Note: Each channel that is assigned to a VAF Variant must reference a corresponding VAF Wild Type channel.

11. (Optional—Variant allele detection (VAF) analyses only) Enter the VAF cutoff for each target.
 - a. Click **VAF CUTOFF** to open the VAF cutoff dialog.
 - b. Toggle the switch on for each target that you want to include in the analysis, then enter the percent (%) threshold value into the **Value** field.
The value must be a percentage in the 0–100 range, expressed to up to 2 decimal places.

If you enable the optional VAF cutoff settings for one or more targets, one of the following statuses are displayed in the **VAF Cutoff** column in the **Variant allele detection** results table:

- Samples with the target below the specified cutoff: **Below <cutoff value>**
- Samples with the target at the specified cutoff: **At <cutoff value>**
- Samples with the target above the specified cutoff: **Above <cutoff value>**

For more information, see “RESULTS tab (completed runs and studies)” on page 103.

12. From the **Default Threshold** dropdown list, select **Auto Sample**, **Auto Group**, or **Manual** for each active optical channel.

13. (Auto Group or Manual default thresholds only) Enter the threshold guide or threshold value.
 - If you selected **Auto Group** as the default threshold, enter the desired value into the **Threshold Guide** field.
 - If you selected **Manual** as the default threshold, enter the desired threshold value into the **Manual value** field.

For more information, see “Manage groups” on page 70.

14. Click **CONTINUE** to add the sample group and close the dialog. Alternatively, click **ADD & CONTINUE**, to add the current sample group and proceed to create another sample group.

Edit groups

Groups can be edited from either the **Templates** or **Runs** screens, as part of the template or run editing workflows.

1. Navigate to the list of templates or runs.
 - Click  **Templates** to open the **Templates** screen.
 - Click  **Runs** to open the **Runs** screen.
2. If needed, use the search field to find a run or template.
3. Open a run or a template.
 - In the **Templates** screen, click the name of the template, then in the **Select action** dialog, select **Edit template**, then click **EDIT TEMPLATE**.
 - In the **Runs** screen, click the name of the run.
4. Navigate to the **SETUP** tab for the template or run.
5. In the **Sample groups** list, click  (**Edit**) in the row of the group you want to modify.
6. In the **Edit sample group** dialog, edit the group settings as needed, then click **CONTINUE**.
For details about each group setting, see “Manage groups” on page 70. For instructions on how to modify group settings, see “Create groups” on page 72.
7. Click **SAVE** in the top-right corner of the screen to save the changes to the template or run.

Delete groups

Groups can be deleted from either the **Templates** or **Runs** screens, as part of the template or run editing workflows. Only groups that do not contain samples can be deleted.

1. Navigate to the list of templates or runs.
 - Click  **Templates** to open the **Templates** screen.
 - Click  **Runs** to open the **Runs** screen.
2. If needed, use the search field to find a run or template.

3. Open a run or a template.
 - In the **Templates** screen, click the name of the template, then in the **Select action** dialog, select **Edit template**, then click **EDIT TEMPLATE**.
 - In the **Runs** screen, click the name of the run.
4. Navigate to the **SETUP** tab for the template or run.
5. In the **Sample groups** list, click  (**Edit**) in the row of the group you want to delete.
6. At the bottom of the **Edit sample group** dialog, click **DELETE SAMPLE GROUP**.
7. Click **SAVE** in the top-right corner of the screen to save the changes to the template or run.

Assign samples to groups

Assigning samples to groups defines the analysis and results types for reporting for individual samples or sets of samples.

Samples can be assigned to groups from the **Runs**, **Templates**, or **Studies** screens, as part of the template, run, or study editing workflows.

For instructions on adding groups, see “Create groups” on page 72

1. Navigate to the list of templates, runs, or studies.
 - Click  **Templates** to open the **Templates** screen.
 - Click  **Runs** to open the **Runs** screen.
 - Click  **Studies** to open the **Studies** screen.
2. If needed, use the search field to find a run or template.
3. Open a run, a template, or a study.
 - In the **Templates** screen, click the name of the template, then in the **Select action** dialog, select **Edit template**, then click **EDIT TEMPLATE**.
 - In the **Runs** screen, click the name of the run.
 - In the **Studies** screen, click the name of the study
4. Navigate to the **SETUP** tab.

5. Use one of the following options to assign samples to a group.

Note: By default, all sample wells are assigned to the preconfigured **Group 1**. You can assign each sample to a group individually or assign multiple samples to the same group at once.

Option	Description
Assign each sample to a sample group individually	Use the Group dropdown list in the sample plate layout to assign a new group for each individual sample.
Assign multiple samples to a sample group	1. Select the sample well of each sample that you want to assign to a group. 2. Use the Sample group dropdown list above the sample plate layout to select the sample group that you want to assign to the selected samples. 3. Click APPLY TO SELECTION.

6. Repeat step 5 as needed to assign all samples to groups.
7. Click **SAVE** in the top-right corner of the screen to save the changes to sample group assignments.

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Analysis features

QuantStudio™ Absolute Q™ Digital PCR Software includes the following analysis features.

- Single-plate analysis—In the **Runs** screen, the **COMPLETED** tab contains single-plate experiment results (see “Single-plate analysis (completed runs)” on page 78).
- Multi-plate analysis—The **Studies** screen contains multi-plate results for groups of completed runs and supports the combined analysis of the grouped runs (see “Multi-plate analysis (studies)” on page 107).

Both options support access to the **PROTOCOL**, **SETUP**, **ANALYSIS**, and **RESULTS** tabs for analyzing and viewing experiment results.

- The **PROTOCOL** screen provides the protocol settings used for a completed run (see “PROTOCOL tab (completed runs and studies)” on page 80).
- The **SETUP** screen provides controls for analysis options, including sample and target names, sample groups, optical channels, replicate statistics, pooling, and copy number calculations (see “SETUP tab (completed runs and studies)” on page 81).
- The **ANALYSIS** screen provides relevant information by sample or group of samples and provides options for viewing the data (see “ANALYSIS tab (completed runs and studies)” on page 85).
- The **RESULTS** screen provides a summary of the run data and reporting and data exporting options (see “RESULTS tab (completed runs and studies)” on page 103).

Analysis workflow

The following is a typical workflow for analyzing run data.

Analysis workflow

 Select a completed run (page 79)

OR

Select a study (page 112)

 Review plate setup and modify as needed before starting analysis (page 81) (Optional) Review the PCR protocol (page 80) View the general analysis and adjust group thresholds if needed (page 86) View sample-specific analysis and adjust individual sample thresholds if needed (page 96) Investigate QC flags and adjust QC bounds if needed (page 97) View results, export data, and generate a report (page 103)

Single-plate analysis (completed runs)

You can perform the following tasks in the **COMPLETED** tab in the **Runs** screen to view and analyze single-plate runs. For information about multi-plate analysis, see “Multi-plate analysis (studies)” on page 107.

- View information about the run including plate barcode, status, user who created the run, the instrument used to perform a run, the QuantStudio™ Absolute Q™ Digital PCR Software version used the first time the run was analyzed, and the dates the run was modified and exported.

Note: If a run was analyzed using an earlier version of the QuantStudio™ Absolute Q™ Digital PCR Software, you can manually re-analyze the run using the latest version of the software. For more information, see “Update the analysis version” on page 133.

- Open a run to view run parameters, set analysis parameters, and view results for a single-plate run (see “Open a completed run” on page 79).
- **ADD TO STUDY**—Add runs to a study. Studies are a group of completed runs that facilitate the results of those runs to be analyzed together (see “Add completed runs to a study” on page 109).
- **EXPORT**—Export runs for use on other Absolute Q™ systems (see “Export runs” on page 68).

- **RENAME**—Rename a run (see “Rename a run” on page 69).
- **DELETE**—Delete one or more runs (see “Delete runs” on page 69).
- **IMPORT RUN**—Import runs created on other Absolute Q™ systems or runs generated on an automated system (see “Import runs” on page 66).
- **CREATE RUN**—Create a new run (see “Create a run from the Runs screen” on page 55).

Open a completed run

COMPLETED runs can have the following statuses.

Status	Description
ABORTED	The run execution was aborted on the instrument.
FINISHED	The run completed without error.
WAITING	The run is finished and waiting to be analyzed.
ANALYZING	The run is in the analysis phase.
ERROR	An error has occurred during the run.

1. In the left navigation pane, click  **Runs** to open the **Runs** screen.
By default, the **Runs** screen opens on the **COMPLETED** tab.
2. (Optional) Use the search field or filter options to locate a run.
 - a. Click  above the list of runs.
 - b. Select one or more filters from the filter options, then click **APPLY**.
You can filter by run **STATUS**, the **INSTRUMENT** associated with the run, or **Filter selections by study compatibility** to filter the run list by the study inclusion criteria. For information about study criteria, see “Multi-plate analysis (studies)” on page 107.
The run list refreshes to show runs with the selected filters.
 - c. If needed, click the  to remove a filter from the list of runs.

The list of runs is updated to reflect the search or selected filters.

3. Click the run name, then proceed to view run information and analyze run data.

Action	Instructions
View protocol information	“PROTOCOL tab (completed runs and studies)” on page 80
View or modify plate settings	“SETUP tab (completed runs and studies)” on page 81
Perform analysis	“ANALYSIS tab (completed runs and studies)” on page 85
View and export results	“RESULTS tab (completed runs and studies)” on page 103

PROTOCOL tab (completed runs and studies)

The **PROTOCOL** tab for a completed run provides the following information.

- Protocol settings used for the run
- The date and time a run was completed
- The plate barcode number
- The instrument that was used to complete a run
- The name of the user—based on the implementation (see Table 2)

Note: Within a study, the protocol for each run can be viewed individually by clicking through the list of runs (see “Open a study” on page 112)

Table 2 User name based on implementation

Implementation	User name
Standalone, not using SAE	Unknown (local)
Standalone, using SAE	The user name of the person who performed the run.
Runs imported from other systems	Unknown (remote)
AutoRun	Unknown (remote)

You can perform the following actions in the **PROTOCOL** tab for a completed run or runs in a study. For detailed instructions, see “View, export, or save the protocol (PROTOCOL tab—completed runs and studies)” on page 80.

- **EXPORT PROTOCOL**—Export the protocol.
- **SAVE AS TEMPLATE**—Save the run settings as a new template.
- **E-SIGN**—For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software (see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”).

View, export, or save the protocol (PROTOCOL tab—completed runs and studies)

After a run is completed, you can view, export, or save the protocol as a new template. You cannot modify the protocol for a completed run.

Open a completed run or a study.

- Open a completed run (page 79)
- Open a study (page 112)

1. Open the **PROTOCOL** tab.

2. (Optional) Export the protocol.

a. Click **EXPORT PROTOCOL** to open the **File browser**, then navigate to the location where you want to save the exported protocol AQUA file.

b. Click **Export**.

3. (Optional) Create a template of the run with a new name.
 - a. Click **SAVE AS TEMPLATE**.
 - b. When prompted, enter the new template name in the **Template name** field, then click **SAVE AS TEMPLATE**.
4. (Optional) For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, click **E-SIGN** to provide an e-signature (see “Sign data in the software” on page 170).

SETUP tab (completed runs and studies)

The **SETUP** tab for completed runs provides the following information.

- The date and time a run was completed
- The plate barcode number
- The instrument that was used to complete a run and the calibration
- The name of the user—based on the implementation (see Table 2)
- For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, the **Run ID** appears below the run or study name in the upper-left corner of the screen. For more information see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”.

Note: Within a study, the plate setup for each run can be viewed individually by clicking through the list of runs (see “Open a study” on page 112).

You can make the following changes to the setup of the MAP plate on a completed run or runs in a study. For detailed instructions, see “View or edit plate settings (SETUP tab—completed runs and studies)” on page 82.

- Edit sample names—Sample names are user-assigned identifiers for the contents of each loaded well of a plate.
- Set the dilution factor (DF) denominator—The dilution factor is the total dilution from the sample to the reaction mix. The value entered in the DF field is the denominator of the dilution factor and must be ≥ 1 .

For example, if 2 μL of the sample is put into a 10 μL reaction mix, the dilution factor is $[2 \mu\text{L sample} / 10 \mu\text{L reaction} = 1/5]$. In this case, the dilution factor denominator that you will enter is 5. If the stock solution is diluted by a factor of 100, then 2 μL of the diluted sample is used in a 10 μL reaction mix, then the dilution factor is $[(1 \mu\text{L stock} / 100 \mu\text{L dilution}) \times (2 \mu\text{L dilution} / 10 \mu\text{L reaction}) = 1/500]$. In this case, the dilution factor denominator that you will enter is 500.

Note: The dilution factor field accepts scientific notation, for example 10E+10.

- Assign samples to groups—Groups determine what type of analysis is applied to all samples within a group.
- **ADD GROUP**—Create more groups (see “Manage groups” on page 70).
- Edit or delete sample groups—Edit group name, type, analyses, and dyes or delete unused groups.

- **EDIT DYES**—Reassign the dye for the green channel for this run.

Note: This action is only available for runs that were performed using the same software version that is used for analysis.

- **Export plate setup**—Export a blank or configured plate setup CSV file to modify outside of the QuantStudio™ Absolute Q™ Digital PCR Software.
- **Import plate setup**—Import plate setup files.
- **ADD NOTE**—Add notes regarding the run.
- **SAVE AS TEMPLATE**—Save the run settings as a new template.
- **E-SIGN**—For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, see Appendix E, “Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later”.

Figure 20 SETUP tab for completed runs used in a study

View or edit plate settings (SETUP tab—completed runs and studies)

After a run is completed, you can view and modify the MAP plate settings.

Open a completed run or a study.

- Open a completed run (page 79)
 - Open a study (page 112)
1. Open the **SETUP** tab.
 2. (Studies only) From the **Run name** list, select a run.

3. (Optional) Modify the plate settings.

The screenshot shows the 'SETUP' tab of the software interface. It features a grid of sample wells (A-D, 1-4) with dropdown menus for sample names, sample types, and dilution factors. A right-hand panel shows 'sample groups' with a table of dye assignments and a 'Notes' section at the bottom. Numbered callouts (1-13) point to specific UI elements: 1 points to a sample group dropdown, 2 to a sample type dropdown, 3 to a dilution factor field, 4 to a sample selection checkbox, 5 to an 'APPLY TO SELECTION' button, 6 to a sample name field, 7 to an import/export button, 8 to the 'sample groups' panel header, 9 to an 'EDIT DYES' button, 10 to an 'ADD GROUP' button, 11 to an edit icon for a group, 12 to an 'ADD NOTE' button, and 13 to the 'RUN STARTED' information at the bottom.

Figure 21 Plate SETUP settings

- | | |
|---|--|
| ① Group assigned to sample | ⑧ View group details |
| ② Sample type assigned to sample (view or select) | ⑨ Edit dye assignment to the green channel |
| ③ Sample dilution factor | ⑩ Add a sample group |
| ④ Select samples for batch update | ⑪ Edit a sample group |
| ⑤ Update selected samples | ⑫ Add notes |
| ⑥ Sample name (view or edit) | ⑬ View run information |
| ⑦ Import or export plate setup | |

Setting	Action
Sample name	Sample wells are preconfigured with a default name. To change the name, click the sample name, then enter a new name in the field (see ⑥).
Sample type	By default, all samples are assigned an Unknown sample type. Use the sample type dropdown list to assign Unknown , Positive Control , or Negative (no template control) sample type to a sample (see ②). Note: Assigning a sample type does not affect the results. The sample type is an attribute of the sample, and therefore only one sample type can be assigned for each sample. The sample type appears in the Analysis screen (samples table), Results ▶ Quantification screen, as well as the Plate Setup and Results sections of the PDF report.

(continued)

Setting	Action
Dilution factor (DF)	Sample wells are preconfigured with a dilution factor of 1. You can change the dilution factor for each sample individually or for multiple samples at once. - For a single sample: enter a value in the Dilution factor field of the sample (see ③). - For multiple samples: select the samples in the sample plate layout, enter a value in the Dilution factor field above the table, then click APPLY TO SELECTION (see ④ and ⑤).
Add a sample group	- Click + ADD GROUP (see ⑩). - In the Add sample group dialog, enter Group name. - Select Individual, Replicates, or Pooling. - Use the switch to toggle channels on or off, then enter the target name into the Target field. - For each active channel, select the analysis from the Analysis dropdown lists. - If you selected CNV Ref, enter the number of copies into the Copies field. - If you selected Variant, select the wild-type reference from the Wild-type ref field. - Click CONTINUE to add the sample group and close the dialog. Alternatively, click ADD & CONTINUE, to add another sample group. For more information, see “Manage groups” on page 70 and “Create groups” on page 72.
Edit or remove a sample group	To determine if you need to edit a sample group, you can view sample group Dye, Target, Analysis, and Threshold information by expanding the group in the Sample groups list (see ⑧). In the Sample groups list, click  (Edit) in the row of a group (see ⑪). - To edit a group—in the Add sample group dialog, modify group properties, then click CONTINUE. - To delete a group—click DELETE SAMPLE GROUP. For more information, see “Manage groups” on page 70 and “Create groups” on page 72.
Assign samples to groups	By default, all sample wells are assigned to the preconfigured Group 1. You can assign each sample to a group individually or assign multiple samples to the same group at once. - For a single sample: use the Group dropdown list in the sample plate layout to select a new group (see ①). - For multiple samples: select the samples in the sample plate layout, select a new group from the Sample group dropdown list above the table, then click APPLY TO SELECTION (see ③ and ④).
EDIT DYES	In the Sample groups list, click EDIT DYES, then use the Dye dropdown list to change the dye assignment for the Green channel. Click CONTINUE (see ⑨).
View run information	The following run information is summarized below the plate layout pane: run start date and time, plate barcode, instrument, user, and calibration (see ⑬).

4. (Optional) Import or export the plate setup. Click ... **(Actions)** in the upper-right corner of the sample plate layout to access the **Import plate setup** and **Export plate setup** options (see ⑥).

Option	Actions
Import plate setup —Import an existing plate setup. You can use the imported plate setup as-is, or modify as needed.	1. Click ... (Actions), then click Import plate setup. 2. In the File browser, navigate to the location of the plate setup CSV file to be imported. 3. Select the plate setup file, then click Open.
Export plate setup —Export the plate setup file to modify or use as-is in other templates or runs.	1. Click ... (Actions), then click Export plate setup. 2. In the File browser, navigate to the location of where to export the plate setup CSV file. 3. Click Save.

5. (Optional) In the **Notes** pane, in the **Enter note** field, enter any notes related to this plate setup, then click **ADD NOTE** (see ⑪).
You can edit or remove existing notes if needed.
6. (Optional) Create a template of the current run parameters.
 - a. Click **SAVE AS TEMPLATE**.
 - b. When prompted, type the new name in the **Template name** field, then click **SAVE AS TEMPLATE**.
7. In the top-right corner of the screen, click **SAVE** to save the changes.
8. (Optional) For implementations using Security, Auditing, and E-signature (SAE) v2.2 or later software, click **E-SIGN** to provide an e-signature (see “Sign data in the software” on page 170).

ANALYSIS tab (completed runs and studies)

The **ANALYSIS** tab in a completed run or a study displays plots for samples and sample groups. The sample list provides information about individual samples included in a group and the ability to modify which samples are included in the plots.

Analysis by **Sample Group** enables the comparison of dyes across samples in a group and curation of sample data at the group level. For information about analyzing runs by sample group, see “ANALYSIS tab—View by Sample Group” on page 86.

Analysis by **Sample** enables the curation of sample data at individual sample level. For information about analyzing sample data, see “ANALYSIS tab—View by Sample” on page 96.

ANALYSIS tab—View by Sample Group

Analysis by **Sample Group** compares dyes across the samples in a group.

The **GALLERY** view for a sample group provides an overview of the data for all samples in a group, organized by dye, and supports the following actions on sample data.

- Hide or show a sample in analysis
- Omit or include a sample in analysis
- Investigate QC alerts
- View sample data details
- Download plots

For details about the **GALLERY** view for a sample group, see the following sections:

- For 1D scatter plots, see “Analyze sample groups—1D SCATTER PLOT—GALLERY view” on page 86.
- For 2D scatter plots, see “Analyze sample groups—2D SCATTER PLOT—GALLERY view” on page 92.

The **DETAIL** (1D scatter plots) or **OVERLAY** (2D scatter plots) view for a sample group provides detailed data information for all samples in a group, organized by dye, and supports the following actions on sample data.

- Hide or show a sample in analysis
- Omit or include a sample in analysis
- Investigate QC alerts
- View sample data details
- Adjust thresholds for samples in a group by dye
- Adjust the data range to zoom in or out on an area in the plot
- Download plots

For details about the **DETAIL** (1D scatter plots) or **OVERLAY** (2D scatter plots) view for a sample group, see the following sections:

- For 1D scatter plots, see “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.
- For 2D scatter plots, see “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

For investigating QC data, see “Analysis—QC data and arrays” on page 97.

Analyze sample groups—1D SCATTER PLOT—GALLERY view

For information about analyzing sample groups in **1D SCATTER PLOT—DETAIL** view, see “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.

For information about **2D SCATTER PLOT** features, see:

- “Analyze sample groups—2D SCATTER PLOT—GALLERY view” on page 92.
- “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

To review QC data, see “Analysis—QC data and arrays” on page 97.

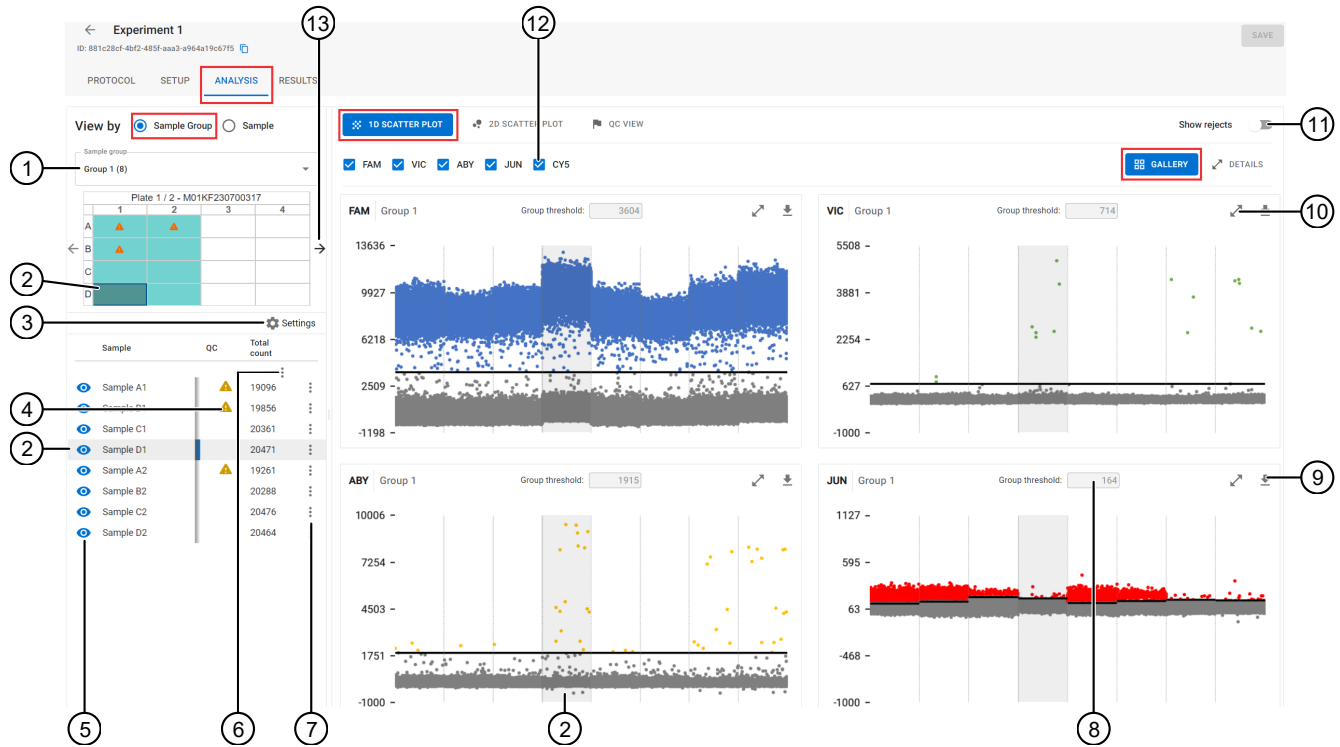


Figure 22 Sample Group—1D SCATTER PLOT—GALLERY view

Table 3 General ANALYSIS tab features

Callout	Feature	Description
①	(View by Sample Group only) Select a sample group.	From the Sample group dropdown list, select a sample group to view data for that sample group.
②	Highlight the sample of interest.	Click a well in a plate, a sample in the samples table, or a sample in the plot view to highlight that sample.
	View information about a sample.	Place the pointer over the sample column in the scatter plot to view information about that sample. Sample—Sample name Well—Well location on the plate Threshold—Threshold of the sample Total—Total number of accepted microchamber reactions less the number of microchambers that have been rejected Plate barcode—Barcode number of the plate Run—Name of the run Instrument—Name of the instrument used for the run
	(View by Sample Group only) Open 1D SCATTER PLOT GALLERY VIEW for a sample.	Right-click a sample column in the scatter plot, then click View Sample . The Analyze by ▶ Sample is selected and 1D SCATTER PLOT GALLERY VIEW opens for the sample.

Table 3 General ANALYSIS tab features (continued)

Callout	Feature	Description
③	Configure the display and order of sample table columns. Note: The Total count column displays the number of accepted microchamber reactions less the number of microchambers that have been rejected	- Click  (Settings) to open Table Settings, then customize the table view by selecting the preferred table settings: Table Columns and the Sort direction of the table columns. Table Organization—select one of the available options to organize sample rows by run (default), sample name, plate barcode. You can also choose not to organize samples by any category. Table Columns—select columns that you want to display in the samples table and modify column order as needed. Sort direction—select to order the samples table by columns (A1, B1, etc.) or rows (A1, A2, etc.). - Click SAVE TABLE SETTINGS to update the samples table with the new settings.
④	View QC flags—Displayed for samples that have either failed or have unusual data.	Place the pointer over the QC flag in the plate view or the samples table to view, then review the warning or error message.  (WARNING)—The sample has unusual data that needs investigation.  (FAILURE)—The sample failed and is automatically omitted from the analysis.  (CRITICAL)—The sample failed and is automatically omitted from the analysis. To view the QC scatter plot for a failed sample, see ⑦. For more information, including analysis of samples that failed QC, see “Analysis—QC data and arrays” on page 97.
⑤	(View by Sample Group only) Show or hide an individual sample in the scatter plots. See also – ②.	In the samples table, click / (Shown/Hidden) in the row of a sample that you want to show or hide, then click  UPDATE PLOTS to view the selection. A sample that is hidden from the scatter plots is designated with  (HIDDEN) flag in the plate view.
⑥	(View by Sample Group for studies only) Show or hide all samples in a plate from the samples table.	In the samples table, in the row of a plate that you want to show or hide, click  (Actions), then select Hide all samples or Show all samples.

Table 3 General ANALYSIS tab features (continued)

Callout	Feature	Description
⑦	Include or omit an individual sample from analysis. See also – ②.	In the samples table, in the row of a sample that you want to omit, click  (Actions), then click Omit sample or Unomit sample , then click SAVE to view the selection. Note: If a sample has a  (CRITICAL) flag, click  (Actions) ▶ View QC to open the QC scatter plot for the sample. For more information, see “Analysis—QC data and arrays” on page 97) For more information about omitting or including failed QC samples, see “Analyze QC data for a sample” on page 100.
⑨	Download a plot for a dye. Plots are saved in PNG format.	In the top-right corner of a plot, click  (Download), navigate to the location to save the file, then click Save .
⑪	Display or hide microchambers that have been rejected from the analysis results.	Use the Show rejects toggle to display or hide the rejects. Showing rejects does not impact the analysis or results calculations.
⑫	Customize dyes to be included in the 1D SCATTER PLOT display.	Select or deselect the dyes to be included or omitted, then click  UPDATE PLOTS to view the selection.
⑬	(Studies only) Switch plate view to a different plate.	Use the arrows to switch between different plates that are included in a study.

Table 4 1D SCATTER PLOT—GALLERY view features

Callout	Feature	Description
⑧	View group threshold.	The group threshold is displayed in the 1D SCATTER PLOT for each dye. By default, AUTO GROUP threshold is displayed. To change the threshold to AUTO SAMPLE or use a MANUAL threshold, see the DETAIL view (“Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90).
⑩	Open the DETAIL view of the results.	To open the DETAIL view of the 1D SCATTER PLOT : - Click  DETAIL in the upper-right corner of the 1D SCATTER PLOT screen. - Click  (Details) in the upper-right corner of the 1D SCATTER PLOT for a dye.

Before performing analysis, complete any modifications that are needed to the sample setup of the plate or plates you are analyzing (see “SETUP tab (completed runs and studies)” on page 81).

1. To access the **ANALYSIS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

By default, when you first open the **ANALYSIS** tab for a run or a study, the first sample group is selected in the **Analyze by** pane and **1D SCATTER PLOT** is populated for each dye.

2. Navigate to the **ANALYSIS** tab, click **1D SCATTER PLOT**, then click **GALLERY** in the top-right corner.
3. Analyze the sample group data using the **ANALYSIS 1D SCATTER PLOT GALLERY** view features described in Table 3 and Table 4.

Proceed to “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.

Analyze sample groups—1D SCATTER PLOT—DETAIL view

For information about analyzing sample groups in **1D SCATTER PLOT—GALLERY** view, including a comprehensive overview of **ANALYSIS** tab features, see “Analyze sample groups—1D SCATTER PLOT—GALLERY view” on page 86.

For information about **2D SCATTER PLOT** features, see:

- “Analyze sample groups—2D SCATTER PLOT—GALLERY view” on page 92.
- “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

To review QC data, see “Analysis—QC data and arrays” on page 97.

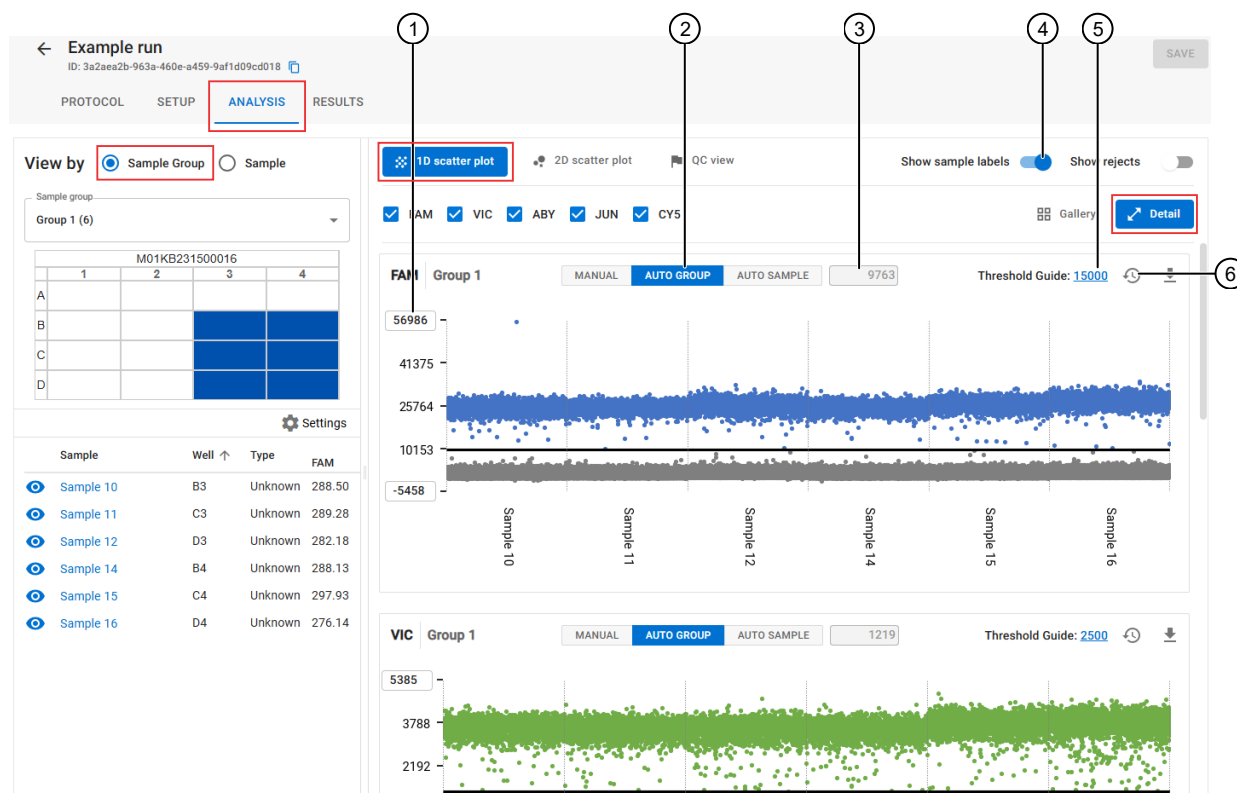


Figure 23 Sample Group—1D SCATTER PLOT—DETAIL view

Table 5 1D SCATTER PLOT—DETAIL view features

Callout	Feature	Description
①	Adjust the data range to zoom in or out on an area in the scatter plot.	1. Enter the upper and lower limits into the upper and lower level fields on the Y-axis. 2. Click SAVE in the upper-right corner of the screen. 3. To return to default Y-axis range, click  (Return) (see ⑥).
② , ③	Select auto-threshold for a sample or a group or manually customize thresholds for each dye.	1. Select the threshold type (see ②). 2. (MANUAL threshold only) Enter the desired threshold value into the field (see ③). 3. Click SAVE in the top right corner of the screen. For more information, see “Select the threshold mode or manually adjust threshold values” on page 113.
④	View or hide sample labels from the scatter plot.	Toggle Show Sample Labels on or off to view or hide the sample labels from the scatter plot.
⑤	Modify the Threshold Guide .	1. Click the Threshold Guide value. 2. In the Edit threshold guide dialog, modify the values for each optical channel as needed, then click DONE. 3. Click SAVE in the top-right corner of the screen. Note: The initial value for the Threshold Guide was defined at the time of template or run creation when creating or editing sample groups. For more information, see: • “Define or edit plate settings (SETUP tab—templates)” on page 45 • “View or edit plate settings (SETUP tab—draft runs)” on page 60 • “Manage groups” on page 70 • “Create groups” on page 72
⑥	Reset scatter plot axis to default.	If changes were made to the Y-axis range, click  (Return) to reset the range to default (see ①).

Before performing analysis, complete any modifications that are needed to the sample setup of the plate or plates you are analyzing. See “SETUP tab (completed runs and studies)” on page 81.

1. To access the **ANALYSIS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

By default, when you first open the **ANALYSIS** tab for a run or a study, the first sample group is selected in the **Analyze by** pane and **1D SCATTER PLOT** is populated for each dye.

- Navigate to the **ANALYSIS** tab, click **1D SCATTER PLOT**, then click **DETAIL** in the top-right corner.
- Analyze the sample group data using the **ANALYSIS 1D SCATTER PLOT DETAIL** view features described in Table 5.

To view general **ANALYSIS** tab features, see Table 3 on page 87.

Analyze sample groups—2D SCATTER PLOT—GALLERY view

For information about analyzing sample groups in **2D SCATTER PLOT—OVERLAY** view, see “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

For information about **1D SCATTER PLOT** features, see:

- “Analyze sample groups—1D SCATTER PLOT—GALLERY view” on page 86.
- “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.

To review QC data, see “Analysis—QC data and arrays” on page 97.

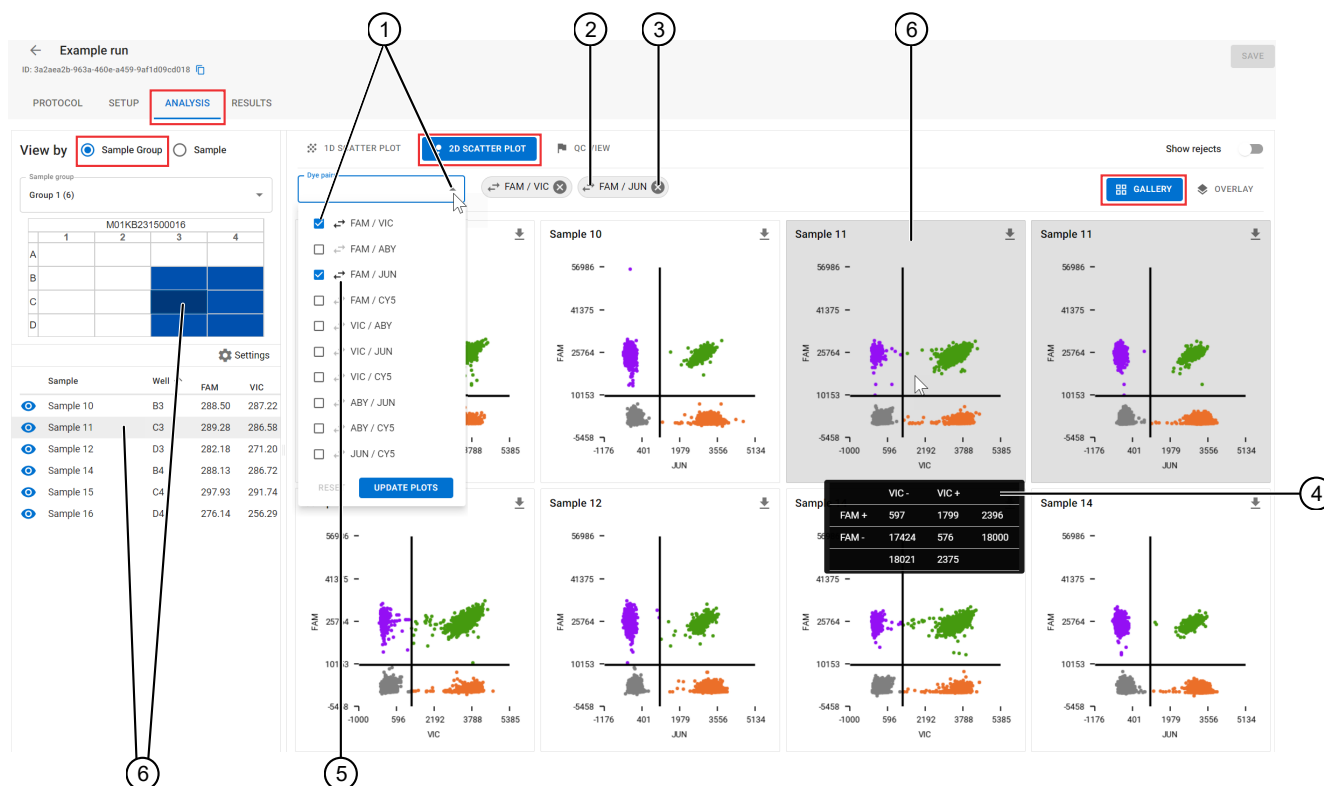


Figure 24 Sample Group—2D SCATTER PLOT—GALLERY view

Table 6 2D SCATTER PLOT—GALLERY view features

Callout	Feature	Description
① , ③	Select the dye pairs to view.	From the Dye pairs dropdown list, select one or more dye pairs, then click  UPDATE PLOTS to generate the 2D plots for the selected dye pairs (see ①) or click RESET to reset the 2D plot selections to the last-saved view. To remove the plots for specific dye pairs, use the Dye pairs dropdown list to de-select the dye pairs, then click  UPDATE PLOTS (see ①). Alternatively, click  next to the dye pair you want to remove, then click  UPDATE PLOTS in the top right corner of the scatter plot pane to generate the 2D plots (see ③). If more than one dye pair is selected, dye pair plots for the same sample will be displayed side-by-side.
② , ⑤	Change X-axis and Y-axis orientation for dyes in a pair.	From the Dye pairs dropdown list, click  (Swap) to swap the dye orientation on the X-axis and Y-axis, then click  UPDATE PLOTS to generate the updated plots (see ②). Alternatively, click  (Swap) next to the selected dye pairs, then click  UPDATE PLOTS in the top right corner of the scatter plot pane to generate the updated plots (see ⑤). Note: You can also adjust the plot axis scale and threshold values in the OVERLAY screen (see page 94).
④	View a summary of total counts for each dye in a pair.	Place the pointer over the scatter plot for a sample to view the breakdown of total counts for each dye in a pair.
⑥	Highlight the sample of interest.	Click a well in a plate, a sample in the samples table, or a sample in the 2D SCATTER PLOT to highlight that sample.

Before performing analysis, complete any modifications that are needed to the sample setup of the plate or plates you are analyzing. See “SETUP tab (completed runs and studies)” on page 81.

1. To access the **ANALYSIS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

By default, when you first open the **ANALYSIS** tab for a run or a study, the first sample group is selected in the **Analyze by** pane and **1D SCATTER PLOT** is populated for each dye.

2. Navigate to the **ANALYSIS** tab, click  **2D SCATTER PLOT**, then click **GALLERY** in the top-right corner.
3. Analyze the sample group data using the **ANALYSIS 2D SCATTER PLOT GALLERY** view features described in Table 6.
To view general **ANALYSIS** tab features, see .

Proceed to “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

Analyze sample groups—2D SCATTER PLOT—OVERLAY view

For information about analyzing sample groups in **2D SCATTER PLOT—GALLERY** view, see “Analyze sample groups—2D SCATTER PLOT—GALLERY view” on page 92.

For information about **1D SCATTER PLOT** features, see:

- “Analyze sample groups—1D SCATTER PLOT—GALLERY view” on page 86.
- “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.

To review QC data, see “Analysis—QC data and arrays” on page 97.

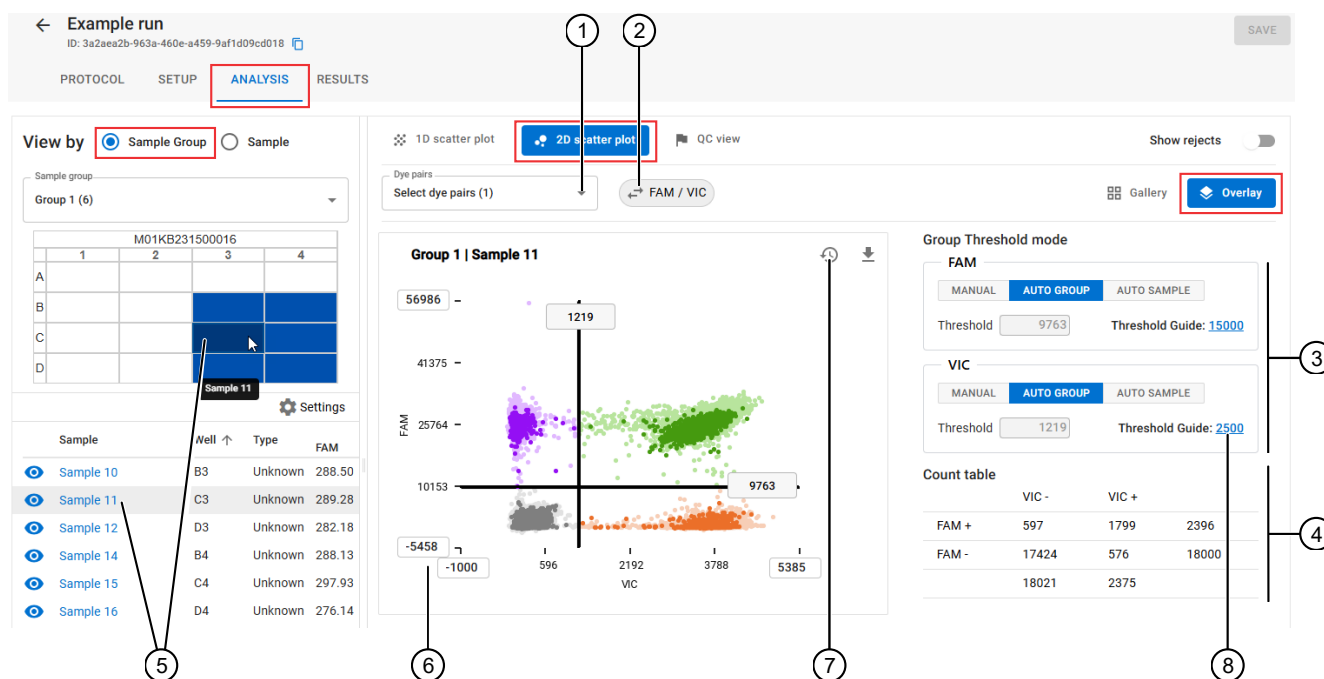


Figure 25 Sample Group—2D SCATTER PLOT—OVERLAY view

Table 7 2D SCATTER PLOT—OVERLAY view features

Callout	Feature	Description
①	Select the dye pair to view.	From the Dye pairs dropdown list, select a dye pair, then click UPDATE PLOTS to generate the 2D plots for the selected dye pairs (see ①) or click RESET to reset the 2D plot selections to the last-saved view Note: Only one dye pair can be selected at a time.
②	Change X-axis and Y-axis orientation for dyes in a pair.	Click ↔ (Swap) to swap the dye orientation on the X-axis and Y-axis (see ②), then click UPDATE PLOTS in the top right corner of the scatter plot pane to generate the updated plots. Alternatively, from the Dye pairs dropdown list (see ①), click ↔ (Swap) to swap the dye orientation on the X-axis and Y-axis, then click UPDATE PLOTS to generate the updated plots or click RESET to reset the 2D plot selection to the last-saved view.

Table 7 2D SCATTER PLOT—OVERLAY view features (continued)

Callout	Feature	Description
③	Select auto-threshold for a sample or a group or manually customize thresholds for each dye.	See “Select the threshold mode or manually adjust threshold values” on page 113. Note: To modify the Threshold Guide , see “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.
④	View the Count table for a sample.	The Count table summarizes total counts for each dye in a pair.
⑤	Navigate to the 2D scatter plot for a different sample.	Click a row in the samples table or a well in the plate view to view the 2D scatter plot for a sample of interest.
⑥	Adjust the data range to zoom in or out on an area in the scatter plot.	1. Enter the upper and lower limits into the upper and lower level fields on the X-axis and Y-axis. 2. Click SAVE in the upper-right corner of the screen.
⑦	Return to default X-axis and Y-axis ranges	To return to default X-axis and Y-axis ranges, click  (Return).
⑧	Modify the Threshold Guide .	1. Click the Threshold Guide value. 2. In the Edit threshold guide dialog, modify the values for each optical channel as needed, then click DONE. 3. Click SAVE in the top-right corner of the screen. Note: The initial value for the Threshold Guide was defined at the time of template or run creation when creating or editing sample groups. For more information, see: • “Define or edit plate settings (SETUP tab—templates)” on page 45 • “View or edit plate settings (SETUP tab—draft runs)” on page 60 • “Manage groups” on page 70 • “Create groups” on page 72

Before performing analysis, complete any modifications that are needed to the sample setup of the plate or plates you are analyzing (see “SETUP tab (completed runs and studies)” on page 81).

1. To access the **ANALYSIS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

By default, when you first open the **ANALYSIS** tab for a run or a study, the first sample group is selected in the **Analyze by** pane and **1D SCATTER PLOT** is populated for each dye.

2. Navigate to the **ANALYSIS** tab, click  **2D SCATTER PLOT**, then click **OVERLAY** in the top-right corner.
3. Analyze the sample group data using the **ANALYSIS 2D SCATTER PLOT OVERLAY** view features described in Table 7.

To view general **ANALYSIS** tab features, see Table 3 on page 87.

ANALYSIS tab—View by Sample

Analysis by **Sample** compares dyes for each individual sample and allows you to visualize and understand the behavior of a single sample.

The **GALLERY** view for a sample provides an overview of the data for each individual sample, organized by dye, and supports the following actions on sample data.

- Navigate between samples in a plate (see Figure 26).
- Omit or include a sample in analysis
- Investigate QC alerts
- View sample data details
- Download plots

For details about the **GALLERY** view for a sample, see the following sections:

- For 1D scatter plots, see “Analyze sample groups—1D SCATTER PLOT—GALLERY view” on page 86.
- For 2D scatter plots, see “Analyze sample groups—2D SCATTER PLOT—GALLERY view” on page 92.

The **DETAIL** (1D scatter plots) or **OVERLAY** (2D scatter plots) views for a sample provides detailed data information for each individual sample, organized by dye, and supports the following actions on sample data.

- Navigate between samples in a plate (see Figure 26).
- Omit or include a sample in analysis
- Investigate QC alerts
- View sample data details
- Adjust thresholds for samples in a group by dye
- Adjust the data range to zoom in or out on an area in the plot
- Download plots

For details about the **DETAIL** (1D scatter plots) or **OVERLAY** (2D scatter plots) for a sample, see the following sections:

- For 1D scatter plots, see “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.
- For 2D scatter plots, see “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

For investigating QC data, see “Analysis—QC data and arrays” on page 97.

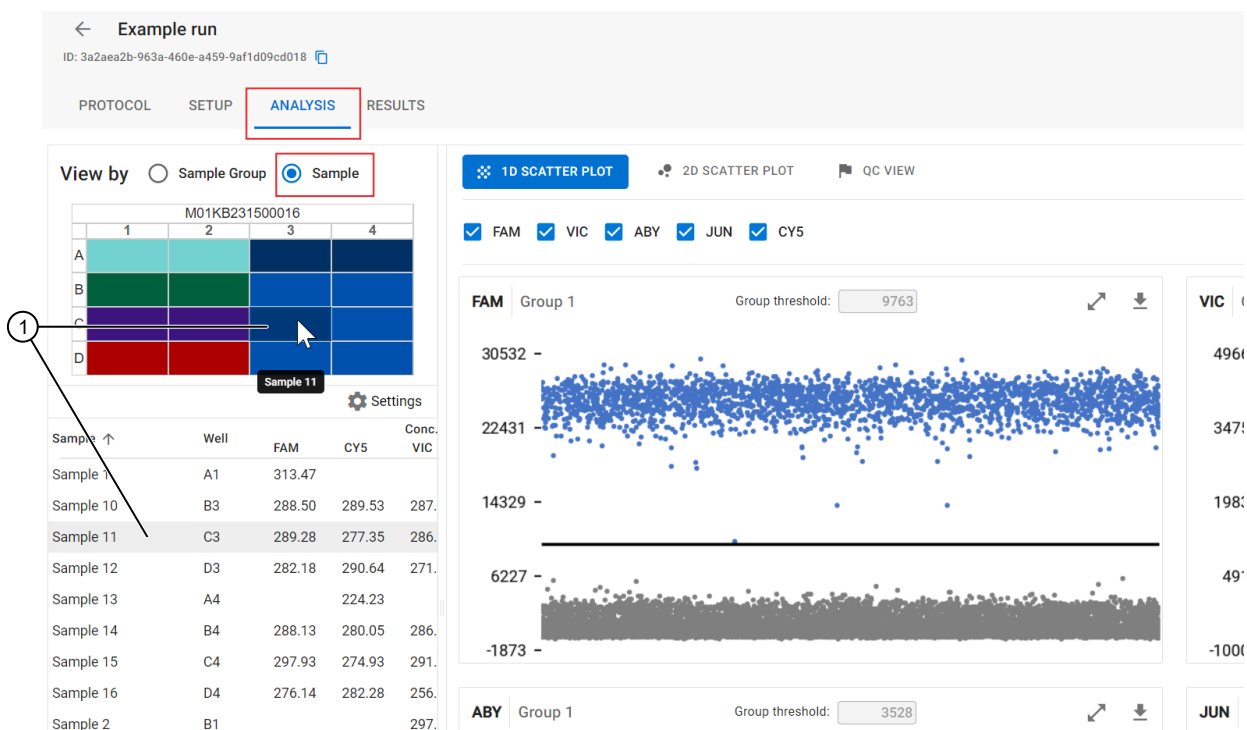


Figure 26

- ① Click a sample in the sample plate or the samples table to display the plots for that sample.

Analysis—QC data and arrays

The **QC VIEW** in the **ANALYSIS** tab displays a QC (quality control) scatter plot for the QC dye–ROX™ for sample groups and samples. Additionally, the **QC VIEW** provides array images and count data summaries for all dyes that were selected for a sample. The QC data helps ensure that only properly filled microchambers are used for analysis by evaluating the ROX™ signal for each microchamber. The upper and lower QC bounds are automatically set by the software and can also be manually adjusted for each sample. An exemplary QC scatter plot should have a single level band, indicating uniform filling.

- For details about **QC VIEW** for a sample group, see “Analyze QC data for a sample group” on page 99.
- For details about **QC VIEW** for individual samples, see “Analyze QC data for a sample” on page 100.

QC flags are used to mark samples with QC failures and warnings. The following QC flags can appear in the plate and table views.

QC flag	Description
 (WARNING)	A sample with a warning flag alerts that there is something unusual about the sample that requires further investigation to determine if the sample should be omitted from the analysis or have its QC bounds or dye thresholds adjusted. If needed, you can manually omit or include a sample with a warning flag in the analysis. Samples with a QC warning can be displayed in 1D SCATTER PLOT, 2D SCATTER PLOT, and QC VIEW in both, View by Sample and View by Sample Group analyses. Samples with a QC warning will also display the concentration values in the samples table in the Analysis tab and the Results tab. For information about how to omit or include samples in an analysis and hide or show samples in the scatter plots, see Table 3.
 (FAILURE)	By default, a sample with a QC failure flag is automatically omitted from the analysis (see “Configure system settings” on page 131). If needed, you can manually include a sample with a QC failure in the analysis. If you manually select to include a sample with a QC failure in the analysis, you can also manually select to show or hide that sample in the scatter plots. Samples with a QC failure can be displayed in the 1D SCATTER PLOT, 2D SCATTER PLOT, and QC VIEW in the View by Sample analysis. For information about how to omit or include samples in an analysis and hide or show samples in the scatter plots, see Table 3. IMPORTANT! We do not recommend including samples with QC failures in analyses because this action can affect the auto-group threshold and the related calculations/results.
 (CRITICAL)	A sample with a critical QC flag is automatically omitted from the analysis and cannot be manually included. Samples with the critical QC flag can only be displayed in the 1D SCATTER PLOT and QC VIEW in the View by Sample analysis. 2D SCATTER PLOT is not supported for samples with the critical QC flag.

QC flags are accompanied by specific messages that explain why a sample was flagged. For more information about the QC failure and warning messages that can occur, including recommended actions, see “QC failure and warning messages” on page 129.

Analyze QC data for a sample group

The **ANALYSIS QC VIEW** for a sample group provides an overview of QC data for all samples in a group.

To adjust QC scatter plots for individual samples and to view array images for all dyes for a sample, see “Analyze QC data for a sample” on page 100.



Figure 27 Sample Group—QC VIEW

Table 8 QC VIEW—View by Sample group

Callout	Feature	Description
①	Adjust the data range to zoom in or out on an area in the scatter plot.	1. Enter the upper and lower limits into the upper and lower level fields Y-axis. 2. Click SAVE in the upper-right corner of the screen. 3. To return to default Y-axis range, click Default range above the scatter plot.
②	View the upper and lower QC bounds for all samples in a group.	Each sample can have unique upper and lower QC bounds. You can adjust the QC bounds for each sample individually in the QC VIEW for a sample (see “Analyze QC data for a sample” on page 100).
③	Highlight the sample of interest.	Click a well in a plate, a sample in the samples table, or a sample in the QC VIEW scatter plot to highlight that sample.

Table 8 QC VIEW—View by Sample group (continued)

Callout	Feature	Description
③	View information about a sample.	Place the pointer over the sample column in the scatter plot to view information about that sample. • Sample—Sample name • Well—Well location on the plate • Total—Total number of accepted microchamber reactions less the number of microchambers that have been rejected • Plate barcode—Barcode number of the plate • Run—Name of the run • Instrument—Name of the instrument used for the run

1. To access the **ANALYSIS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

By default, when you first open the **ANALYSIS** tab for a run or a study, the first sample group is selected in the **Analyze by** pane and **1D SCATTER PLOT** is populated for each dye.

2. Navigate to the **ANALYSIS** tab, then click  **QC VIEW**.
The QC scatter plot for the QC dye–ROX™ is displayed for all samples in the group.
3. In the **Analyze by** pane, ensure that **Sample group** is selected.
4. Analyze the sample group QC data using the **ANALYSIS QC VIEW** features described in Table 8.
To view general **ANALYSIS** tab features, see Table 3 on page 87.

Analyze QC data for a sample

The **ANALYSIS QC VIEW** for individual samples generates a QC scatter plot for the QC dye–ROX™ as well as array images and count data summaries for all optical channels that were selected for a sample.

Array images for microchambers with positive data points are colored. Array images for microchambers with negative and rejected data points are gray scale. Microchambers that have a low signal yield images that can appear black.

To view a QC scatter plot for all samples in a group, see “Analyze QC data for a sample group” on page 99.

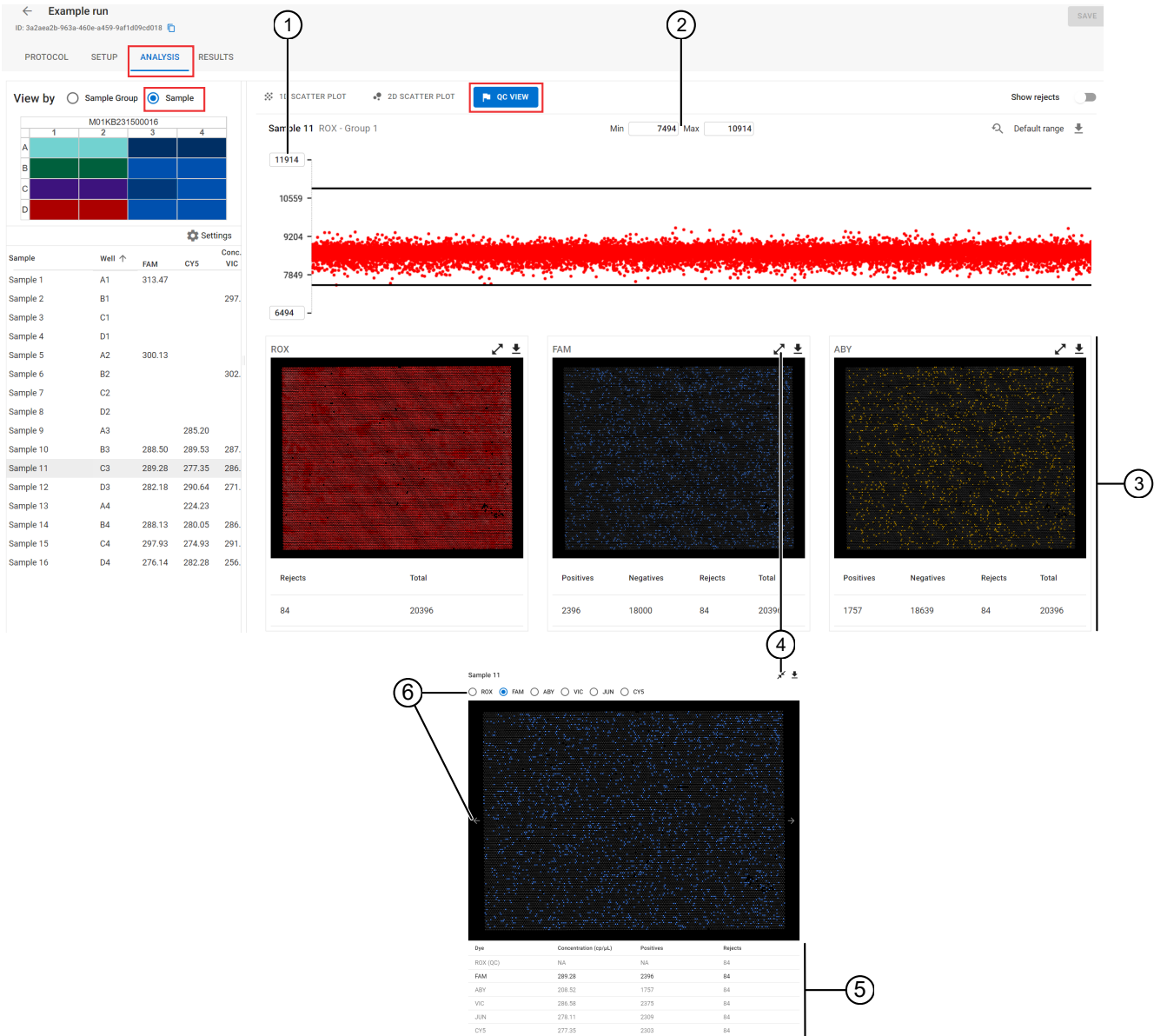


Figure 28 Sample—QC VIEW

Table 9 QC VIEW—View by Sample

Callout	Feature	Description
①	Adjust the data range to zoom in or out on an area in the scatter plot.	1. Enter the upper and lower limits into the upper and lower level fields Y-axis. 2. Click SAVE in the upper-right corner of the screen. 3. To return to default Y-axis range, click Default Range.

Table 9 QC VIEW—View by Sample (continued)

Callout	Feature	Description
②	Adjust the upper and lower QC bounds for an individual sample.	- Above the QC scatter plot, enter the lower QC bound into Min field and upper QC bound into Max. - Click SAVE in the upper-right corner of the screen to save the changes and update the plot. When the new bounds are set for an individual sample, the QC plot for the sample group is also updated to reflect the changes (see “Analyze QC data for a sample group” on page 99).
③, ④	View array images and data for each dye for a sample.	Array images are shown for each dye for a plate well (see ③). You can expand the image for each dye by clicking  (Expand) to view details for the specific dye in a sample (see ④). For each dye for a sample, the number of counts for Positives, Negatives, Rejects, and Total is summarized below each array image.
④, ⑤, ⑥	View the enlarged array image and dye-specific metrics for all dyes in a sample.	When expanded (see ④), an enlarged array image for the specific dye opens. Dye-specific metrics are displayed below the image (see ⑤). The metrics for the selected dye are displayed in black. You can use the arrows to navigate between the enlarged array images of all dyes in a sample or select a specific dye from the options above the image (see ⑥).

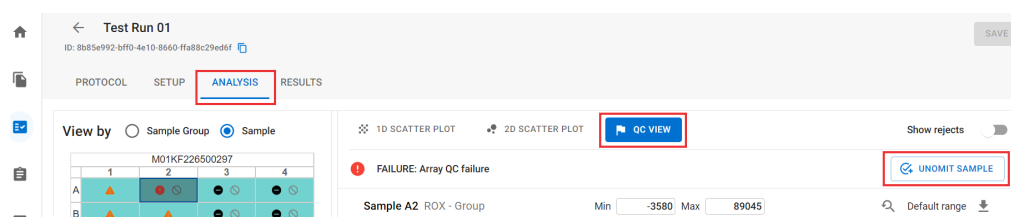
- To access the **ANALYSIS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

By default, when you first open the **ANALYSIS** tab for a run or a study, the first sample group is selected in the **Analyze by** pane and **1D SCATTER PLOT** is populated for each dye.

- Navigate to the **ANALYSIS** tab, then click  **QC VIEW**.

For samples that failed QC and have been omitted from the analysis, click  (**Actions**) ▶ **View QC** in the samples table to navigate to the  **QC VIEW**. To include the omitted sample in the analysis, click **UNOMIT SAMPLE** above the QC scatter plot. To omit a sample from the analysis, click **OMIT SAMPLE** above the QC scatter plot. Click **SAVE** to update the analysis.



You can also use the  (**Actions**) menu in the samples table to omit or include a sample with a failed QC in the analysis (see Table 3 on page 87).

3. In the **Analyze by** pane, ensure that **Sample** is selected.
4. Analyze the sample QC data using the **ANALYSIS QC VIEW** features described in Table 9.
To view general **ANALYSIS** tab features, see Table 3 on page 87.

RESULTS tab (completed runs and studies)

The **RESULTS** tab for a completed run or a study summarizes the results for samples and sample groups in a single table. In addition, the concentration values and their 95% confidence intervals (95%CI) are plotted for each sample or group, organized by dye.

Use the **RESULTS** tab to perform the following actions:

- View results by group and sample.
- View results plots by group or sample.
- Generate data reports in PDF format.
- Download the data table in CSV format using one or a combination of the following download options:
 - Results summary
 - Fluorescence
 - Sample based multichannel
 - Channel based multichannel

For details, see “View results” on page 104.

For information about the formulas used to calculate results, see “Computation of results” on page 127.

For descriptions of the results table headers / data fields, see “Definitions of data fields” on page 186.

The presentation of the **RESULTS** page is based on the data included in analysis in the **ANALYSIS** tab.

View results

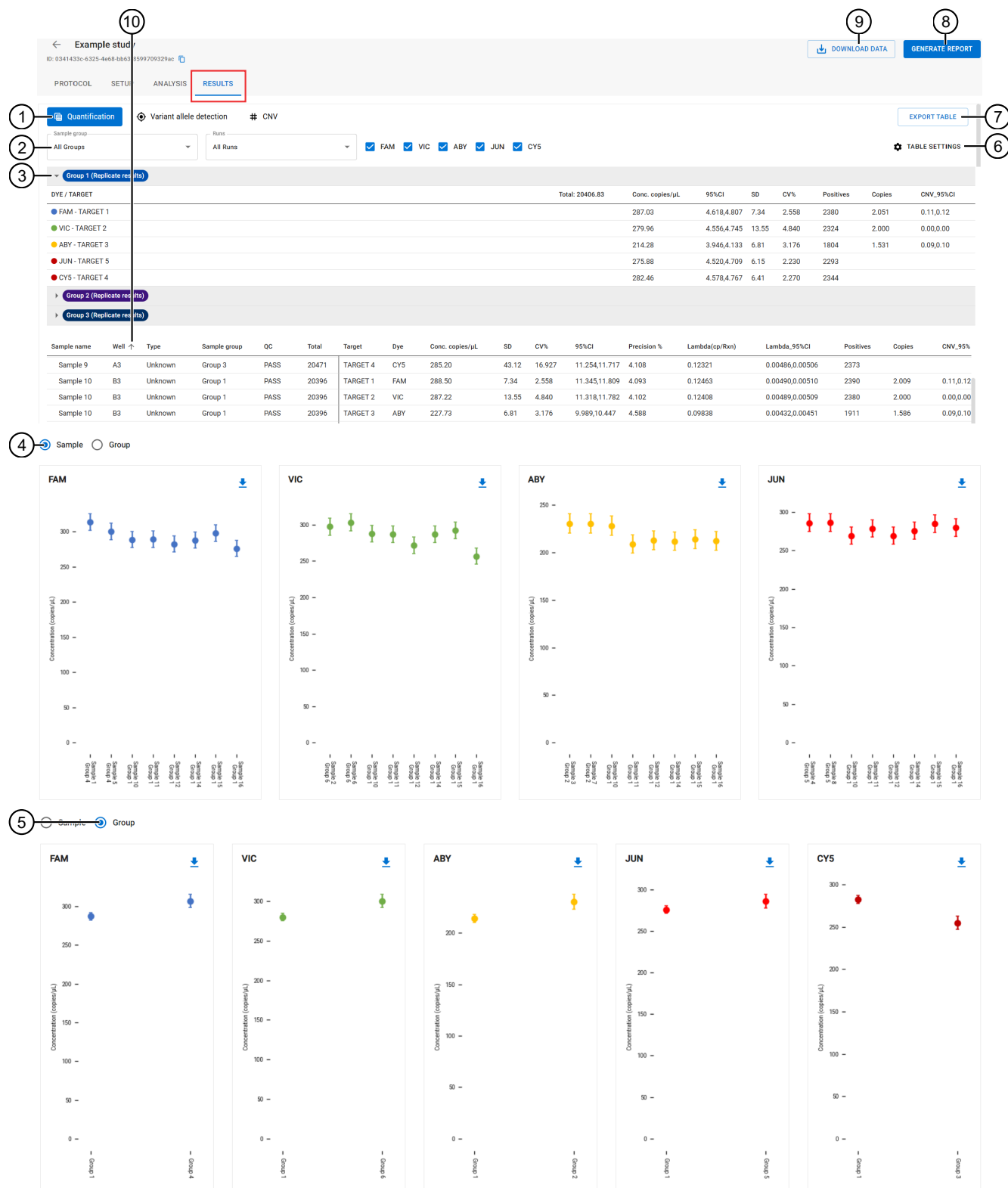


Figure 29 RESULTS tab - completed run or study

Table 10 RESULTS tab features

Callout	Feature	Actions
①	Switch between Quantification , Variant allele detection , and CNV results to focus on the data for a specific analysis type.	Click Quantification, Variant allele detection, or CNV to view the tables for each results type. Each table can be exported as a CSV file (see callout ⑦). Note: - If there is no sample group present with the CNV analysis type, the CNV tab is disabled. <i>(Variant allele detection (VAF) analyses only)</i> If there are multiple sample groups that include the VAF analysis type, the variant allele detection table and the exported CSV file will display the data for each group separately.
②	<i>(Quantification results only)</i> Filter the Quantification results table by sample groups, runs, and dye channels.	Use the dropdown lists and checkboxes to filter the results. You can select one or multiple criteria at once. - From the Sample group dropdown list, select one or more groups to filter the results by group, or select All Groups. Click Done. <i>(Studies only)</i> From the Runs dropdown list, select one or more groups to filter the results by group, or select All Runs. Click Done. - Select the checkbox for one or more dyes, as needed.
③	View group summary data.	Click ► (Expand) to expand the group summary table and review the group summary data for each group.
④	View the concentration results plots by sample.	Select Sample to view concentrations by sample based on the filters selected in ②.
⑤	View the concentration results plots by group.	Select Group to view concentrations by group based on the filters selected in ②.
⑥	<i>(Quantification results only)</i> Configure the display and order of the columns for sample information and the results.	- Click ⚙ TABLE SETTINGS to open the Results Table Settings. - Configure the column order and selections in the SAMPLE and RESULTS tabs. - Select the checkbox in the each row of each column that you want to display in the table, or select Select all to select all columns. - Click and drag in the row of a column to change the column order. - Click SAVE TABLE SETTINGS to save the changes and view the modified table.

Table 10 RESULTS tab features (continued)

Callout	Feature	Actions
⑦	Export the results table in CSV format.	Click EXPORT TABLE, then select a folder in your local storage. One of the following CSV files is exported to the designated folder and reflects the same data and order as it appears in the software. • The Quantification table is exported as <code>quant.csv</code>. • The Variant allele detection table is exported as <code>variantallele.csv</code>. • The CNV table is exported as <code>cnv.csv</code>. <i>(Quantification table only)</i> The exported CSV file reflects all the filters applied in ②, the custom table column configurations applied in ⑥, and the order of data sorted in ⑨. <i>(All tables)</i> The exported CSV file reflects the same data and order as it appears in the software. For example, if you sorted the columns, as described in ⑩, the CSV will reflect the sorted order of data.
⑧	Generate a report.	1. Click GENERATE REPORT. 2. In the Generate report dialog, make the desired selections of Sections to be included in the report, then in the Filter by list, select All Groups to view the results for all groups, or select one or more groups individually. 3. Click GENERATE REPORT. When the report is generated, the file browser window opens. 4. In the file browser window, navigate to the location where you want to save the report, then click SAVE. The report is saved in PDF format and includes the following information, depending on the selections made in step 2. • <i>(Study reports only)</i> Study summary and list of runs • SAE • Protocol and plate setup for each run • List of omitted samples • Separate results tables for each sample • Results by Group • Results by Sample • Signal quantification for each dye

Table 10 RESULTS tab features (continued)

Callout	Feature	Actions
⑨	Download data.	1. Click  DOWNLOAD DATA. 2. In the Download options dialog, select the data to include in the download bundle. 3. Click DOWNLOAD SELECTED. The file browser opens. 4. Select the folder where you want to save the data, then click SELECT FOLDER. The data files are saved to the designated location in CSV format. For more information, see “QuantStudio™ Absolute Q™ Digital PCR Software – Export” on page 184.
⑩	Sort the table columns.	Click a column header to sort the results table by the ascending or descending order of data in that column. Click the table header again to reverse the order. Note: Each sample is displayed per channel. For example, if Sample A1 has results for 4 channels (FAM, VIC, ABY, JUN), the table will display 4 separate rows for A1, each corresponding to a different channel.

1. To access the **RESULTS** tab, open a completed run or a study according to the analysis type.

Analysis type	Procedure
Single-plate analysis (completed runs)	See “Open a completed run” on page 79.
Multi-plate analysis (studies)	See “Open a study” on page 112.

2. Navigate to the **RESULTS** tab.
3. View the results using the features described in Table 10.

Multi-plate analysis (studies)

Studies are a group of completed runs from multiple plates that enable the results of those runs to be analyzed together and include the following features.

- Studies can include up to 24 runs.
- Changes made to runs added to a study do not affect the original run.
- Samples from runs in a study that have identical group setups are merged into a single group for analysis.

IMPORTANT! To be included in a study, runs must meet the following criteria.

- All runs must be created with the same software version.
 - Each run must have completed successfully and have a **FINISHED** status.
 - Each run must have used the same optical collection settings in the run protocol.
 - For standalone and AutoRun single configuration systems, the same instrument must be used to collect the data.
 - For AutoRun dual configuration systems, the instruments must have been co-calibrated and confirmed optically compatible by a qualified Thermo Fisher field service engineer.
 - Data from AutoRun co-calibrated instruments can only be combined into a study if the calibration date of the instruments is the same at the time the run was completed.
-

Studies can be created in one of the following ways.

- From the **Runs** screen, see “Add completed runs to a study” on page 109.
- From the **Studies** screen, see “Create a study” on page 108.

You can perform the following tasks from the **Studies** screen.

- Open a study to view run parameters, set analysis parameters, and view results (see “Open a study” on page 112).
- **CREATE STUDY**—Create a study (see “Create a study” on page 108).
- Add completed runs to a study (see “Add completed runs to a study” on page 109).
- **IMPORT**—Import studies created on other Absolute Q™ systems (see “Import studies” on page 110).
- **EXPORT**—Export studies for analysis on other Absolute Q™ systems (see “Export studies” on page 111).
- **RENAME**—Rename a study (see “Rename a study” on page 111).
- **DELETE**—Delete one or more studies (see “Delete studies” on page 112).

Manage studies

Create a study

Studies can be created from the **Studies** screen or from the **Runs** screen **COMPLETED** tab.

For information about creating a study from the **COMPLETED** tab of the **Runs** screen, see “Add completed runs to a study” on page 109.

1. In the left navigation pane, click  **Studies** to open the **Studies** screen.
2. Navigate to the list of completed runs.

Option	Description
If you have one or more studies in the Studies list	In the upper-right corner of the screen, click CREATE STUDY , then in the Select runs to create study dialog, click GO TO COMPLETED RUNS .
If there are no existing studies in the Studies list	Click GO TO COMPLETED RUNS .

The **Runs** screen opens to the **COMPLETED** tab.

Proceed to “Add completed runs to a study” on page 109.

Add completed runs to a study

To analyze samples in multiple runs as a group, you can add multiple runs to a study.

Completed runs are added to studies from the **Runs** screen, **COMPLETED** tab.

IMPORTANT! To be included in a study, runs must meet the following criteria.

- All runs must be created with the same software version.
 - Each run must have completed successfully and have a FINISHED status.
 - Each run must have used the same optical collection settings in the run protocol.
 - For standalone and AutoRun single configuration systems, the same instrument must be used to collect the data.
 - For AutoRun dual configuration systems, the instruments must have been co-calibrated and confirmed optically compatible by a qualified Thermo Fisher field service engineer.
 - Data from AutoRun co-calibrated instruments can only be combined into a study if the calibration date of the instruments is the same at the time the run was completed.
-

1. In the **Runs** screen, in the **COMPLETED** tab, identify the first run that you want to include in the study.

Option	Description
Use the search field to find a run.	Enter run information in the search field. Search for runs using the following information. • Run name • Plate barcode • Instrument name
Use the filter options to find completed runs.	1. Click  above the list of runs. 2. In the Filter runs dialog, select the FINISHED status, then if needed, select the instrument. 3. Click APPLY.

2. Select a run from the filtered run list.
3. Use the filter option to find and select runs that meet study-compatible criteria.
 - a. Click  above the list of runs.
 - b. In the **Filter runs** dialog, select **Filter selections by study compatibility**, then click **APPLY**.
The run list is updated to show runs that meet study-compatible criteria.
 - c. (Optional) If needed, click the  (**Remove**) to remove a filter selection.
4. Select the run or runs to be added to your study, then click **ADD TO STUDY**.

5. In the **Add to Study** dialog, select one of the following options.

Option	Description
Create a new study	1. Select Create new study. 2. In the Study name field, enter a unique name for the study. 3. Click ADD RUNS TO STUDY.
Add to an existing study	1. Select Add to existing study. 2. From the Study name dropdown list, select a study. 3. Click ADD RUN TO STUDY (if one run was selected) or ADD RUNS TO STUDY (if multiple runs were selected).

The selected runs are added to a new study or an existing study. The software opens the **Studies** screen to the **SETUP** tab for the study.

Note: If runs that are added to a study include sample groups with identical names but different analysis threshold settings, the software automatically creates new sample groups to separate out the results for each group. Review the individual sample group settings in the **SETUP** tab and threshold parameters in the **ANALYSIS** tab in the **1D scatter plot Detail** view or **2D scatter plot Overlay** view to determine the sample group identity. If needed, rename the sample groups to create clarity. For more information, see:

- “View or edit plate settings (SETUP tab—completed runs and studies)” on page 82
- “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90
- “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94

Import studies

Use the **IMPORT STUDIES** feature to import studies created on other Absolute Q™ systems. Study files have a ZIP extension.

1. In the left navigation pane, click  **Studies** to open the **Studies** screen.
2. In the top-right corner of the screen, click **IMPORT STUDIES**.
3. In the **Import studies** dialog, import studies using one of the following options.

Option	Action
Drag-and-drop study files from a location on your file system.	Using File Explorer , locate the ZIP file or files in your file system, then drag-and-drop the file into the Import study dialog.
Browse to locate and add study files.	1. In the Import study dialog, click IMPORT FILE. 2. In the File browser, navigate to the location of the ZIP file or files to be imported. 3. Select one or more study files, then click Open.

4. When the import completes, in the **Import Status** dialog, click **CLOSE**

Note: If there are studies that cannot be imported, an error appears in the **Import Status** dialog. Make note of the error so that you can take corrective action on the run, then click **CLOSE** to continue.

The imported studies are added to the **Studies** list.

Export studies

Use the **EXPORT** feature to export studies for use with other Absolute Q™ systems.

1. In the left navigation pane, click  **Studies** to open the **Studies** screen.
2. If needed, use the search field to locate the studies for export.
3. Select one or more studies from the list, then click **EXPORT**.
4. In the **File browser**, navigate to the location of where to export the study, then click **Select Folder**.
5. When the export completes, in the **Export Status** dialog, click **CLOSE**.

Note: If there are studies that cannot be exported, an error appears in the **Export Status** dialog. Make note of the error so that you can take corrective action on the study, then click **CLOSE** to continue.

Rename a study

Only one study can be renamed at a time.

1. In the left navigation pane, click  **Studies** to open the **Studies** screen.
2. If needed, use the search field to locate a study.
3. Select a study from list, then click **RENAME**.
4. In the **Rename study** dialog, in the **Study name** field, enter a new name for the study, then click **RENAME**.

The **Studies** list is updated to reflect the new study name.

Delete studies

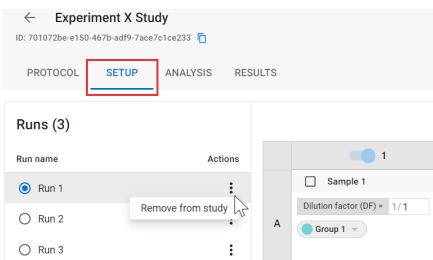
Multiple studies can be deleted at a time.

1. In the left navigation pane, click  **Studies** to open the **Studies** screen.
2. If needed, use the search field to locate studies to be deleted.
3. From the **Studies** list, select one or more studies, then click **DELETE**.
4. When prompted, confirm that the selected study or studies are to be deleted by clicking **DELETE STUDY** (if a single study was selected) or **DELETE [# of studies selected] STUDIES** (if multiple studies were selected). If you do not want to delete the selected studies, click **DO NOT DELETE**.

Open a study

Use the **Studies** screen to open a study to view the grouped runs and prepare for analysis.

1. In the left navigation pane, click  **Studies** to open the **Studies** screen.
2. If needed, use the search field to locate the studies.
3. (Optional) To view the list of runs contained in a study, click ► **(Expand)** in the row of a study.
4. Click the study name to open the study.
By default, the study opens to the **SETUP** tab for the first run in the study.
5. Review the list of runs in the study and modify as needed.

Action	Description
View the protocol and plate setup of a run in a study	In the SETUP tab or the PROTOCOL tab, select the run from the Runs list to view.
Remove a run from a study	In the Runs list, in the row of a run, click  (Actions) ► Remove from study. 

Review the protocol and plate setup details, then continue to analysis.

- To view protocol information, see “**PROTOCOL** tab (completed runs and studies)” on page 80.
- To view or modify plate settings, see “**SETUP** tab (completed runs and studies)” on page 81.
- To perform analysis, see “**ANALYSIS** tab (completed runs and studies)” on page 85.
- To view and export results, see “**RESULTS** tab (completed runs and studies)” on page 103.

Select the threshold mode or manually adjust threshold values

When analyzing the results by sample or sample group, the following threshold mode settings are available in QuantStudio™ Absolute Q™ Digital PCR Software.

Table 11 Threshold mode settings

Threshold mode	Description
Software-defined threshold—View by Sample Group	
AUTO SAMPLE	The software applies the threshold to each sample in a group individually for the dye. The auto sample threshold cannot be modified.
AUTO GROUP	The software applies the group threshold to all samples in the group for the dye. Note: The default threshold values were defined when the sample group was set up during template or run creation. For more information see “Manage groups” on page 70.
MANUAL	Enables a custom threshold to be applied to a sample group for the dye. Note: The default threshold values were defined when the sample group was set up during template or run creation. For more information see “Manage groups” on page 70.
Software-defined threshold—View by Sample	
AUTO	The software applies the sample-specific threshold to the sample for the dye. Selecting AUTO pins the threshold for the sample and isolates it from group threshold changes.
FROM GROUP	The software applies the group threshold to the sample for the dye.
MANUAL	Enables custom threshold to be set to a sample group or a sample for the dye. Selecting MANUAL pins the threshold for the sample and isolates it from group threshold changes.

To adjust the threshold mode:

1. Navigate to the **ANALYSIS** tab for a run or a study, then open one of the following scatter plot screens.
 - In the **ANALYSIS** tab, click  **1D SCATTER PLOT**, then click  **DETAIL** in the top-right corner.
For more information, see “Analyze sample groups—1D SCATTER PLOT—DETAIL view” on page 90.
 - In the **ANALYSIS** tab, click  **2D SCATTER PLOT**, then click **OVERLAY** in the top-right corner.
For more information, see “Analyze sample groups—2D SCATTER PLOT—OVERLAY view” on page 94.

2. Select the threshold mode.

- (View by Sample Group) Above the **1D SCATTER PLOT** for a dye or in the **2D SCATTER PLOT Threshold mode** pane, select **MANUAL**, **AUTO GROUP**, or **AUTO SAMPLE**.

Note: When **AUTO SAMPLE** is selected as the threshold mode, the single threshold cannot be displayed in this view.

- (View by Sample) Select **MANUAL**, **FROM GROUP**, or **AUTO**.

IMPORTANT! Selecting the **MANUAL** or **AUTO** mode pins the threshold for the sample and isolates it from group threshold changes. To clear pinned thresholds for all samples in a group, proceed to step 4.

3. (Optional) Modify the threshold values.

Analysis type	Threshold	Description
Sample Group	MANUAL	Enter a value in the threshold field or drag the threshold bar in the 1D or 2D scatter plot to the desired value.
	AUTO GROUP	In the  1D SCATTER PLOT  DETAILS screen, adjust the Threshold Guide value for each channel as needed. Note: The analysis algorithm uses the Threshold Guide value to adjust the computation of the threshold value for the group.
	AUTO SAMPLE	Cannot be modified.
Sample	Manual	Enter a value in the threshold field or drag the threshold bar in the 1D or 2D scatter plot to the desired value.
	From Group	Cannot be modified.
	Auto	Cannot be modified.

4. (Optional; View by Sample Group only) If the sample group contains one or more samples with pinned thresholds, you can clear all pinned thresholds by clicking **CLEAR PINNED THRESHOLDS** in the top-right corner of the scatter plot pane.

Clearing all pinned individual sample thresholds returns the threshold to the group mode for the sample and sample group.

5. Click **SAVE** at the top-right corner of the screen.



Troubleshooting

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Troubleshooting Absolute Q™ standalone implementations

Observation	Possible cause	Recommended action
QuantStudio™ Absolute Q™ Digital PCR Software is not connecting, front panel LEDs are white	Internal instrument software connection error.	Power off the instrument, then unplug both USB and power cables from instrument. Wait 30 seconds, then plug in the power cord and power on the instrument. When solid blue LED is seen, then plug in the USB cable.
QuantStudio™ Absolute Q™ Digital PCR Software is not connecting, front panel LEDs are blue	Poor USB cable connection.	Confirm that a USB 3.0 cable is used and that it is plugged into the USB 3.0 port on the desktop computer.
	Corrupt software.	Uninstall, then reinstall the software (see “Download, install, and update the software” on page 138).
Front panel LEDs are red	Instrument error.	Power cycle the instrument using the power switch. 1. Power off the instrument. 2. Unplug both USB and power cables from instrument. 3. Wait 30 seconds, then plug in the power cord and power on the instrument. 4. When solid blue LED is seen, then plug in the USB cable. 5. If the LED is red, contact Technical Support.
The run Status displays as DISCONNECTED (QuantStudio™ Absolute Q™ Digital PCR Software v6.3.1 or earlier only)	Port 8000 is blocked.	If a firewall or other application is using port 8000, remove it or use a different port for the firewall or other application. For connection limitations, follow the IT Checklist.
The run Status is stuck at RUNNING after the run has completed	The PC was unable to release the TCP ports.	Restart the companion PC.
Pressure leak error	Missing or damaged gaskets.	Make sure that all 5 columns of gaskets are present.
		Replace any damaged gaskets.
Barcode not found	Plate in backwards.	Well A1 should be at the top left of the plate tray.

Observation	Possible cause	Recommended action
Barcode not found (continued)	Missing or unreadable barcode label.	Enter the barcode manually if it is human readable.
Connection to the standalone SAE Administrator Console is lost	Power outage.	Restore power to the SAE Administrator Console.
	Cables have become disconnected.	Confirm all cables are properly connected.
	Hardware failure of the SAE Administrator Console.	Uninstall and reinstall the QuantStudio™ Absolute Q™ Digital PCR Software to continue use of the QuantStudio™ Absolute Q™ Digital PCR System without SAE. 1. Consult with your organization's policies and procedures regarding operation without SAE enabled before continuing. 2. At the desktop computer, shut down the QuantStudio™ Absolute Q™ Digital PCR Software. 3. Uninstall the QuantStudio™ Absolute Q™ Digital PCR Software. IMPORTANT! To prevent data loss, select Keep during uninstall to preserve the existing database. 4. Install the QuantStudio™ Absolute Q™ Digital PCR Software (see “Download, install, and update the software” on page 138). IMPORTANT! To prevent data loss, select Update during installation of the software to preserve the existing database.
Communication between the instrument and the Absolute Q™ computer is interrupted or inconsistent	Another application may be causing a communication conflict.	Ensure that only the QuantStudio™ Absolute Q™ Digital PCR Software and if applicable SAE Administrator Console are installed on the dedicated computer. Uninstall any other applications. For connection limitations, follow the IT Checklist.
	Hardware failure.	Confirm all cables are properly connected
		Confirm that both devices have power.
		Contact thermofisher.com/support .

Troubleshooting AutoRun Suite implementations

Observation	Possible cause	Recommended action
Not enough disk space error	The Momentum™ computer D drive is full.	1. Navigate to D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Data Output on the Momentum™ computer. 2. Back up and delete files to create more storage space. 3. Click OK on the error message in the Momentum™ software, then initiate a new Work Unit.
Precondition failed error	The companion PC memory is full.	1. Navigate to the computer of the affected Absolute Q™ instrument. 2. Navigate to C:\ProgramData\Applied Biosystems\QuantStudio Absolute Q\Digital PCR Software 6\fsa. 3. Back up and delete files to create more storage space. 4. Navigate back to the Momentum™ computer. 5. Select Restart the Run Template operation.
Absolute Q™ instrument LED is red when an AutoRun is processing in the Momentum™ software	Instrument error.	1. Bring the affected Absolute Q™ instrument offline in the Momentum™ software. 2. Power cycle the Absolute Q™ instrument. 3. Bring the affected Absolute Q™ instrument online in the Momentum™ software. 4. When prompted by the Momentum™ software, select Restart the Access operation. 5. If the error continues to appear, contact Technical Support.
Absolute Q™ instrument LED is red when an AutoRun is not processing	Instrument error.	1. Power cycle the Absolute Q™ instrument. 2. If the error continues to appear, contact Technical Support.
OpenDoorException error	Instrument error.	1. Bring the affected Absolute Q™ instrument offline in the Momentum™ software. 2. Power cycle the Absolute Q™ instrument. 3. Bring the affected Absolute Q™ instrument online in the Momentum™ software. 4. Select Restart the Access operation.
CloseDoorException error	Instrument error.	1. Confirm if the door is jammed. Press the door button on the respective Absolute Q™ instrument. 2. If the door operates as expected, select the Restart the Access operation option. The system attempts to continue.

Observation	Possible cause	Recommended action
CloseDoorException error (continued)	The door is jammed on the Absolute Q™ instrument.	1. Confirm if the door is jammed. Press the door button on the respective Absolute Q™ instrument. 2. If the door is jammed, turn the Absolute Q™ instrument off and contact Technical Support Note: If using a dual configuration system, select Take the "AbsoluteQLeft" device offline or Take the "AbsoluteQRight" device offline to continue the run using the other instrument. Then, if prompted, select Keep the AbsoluteQ device offline.
PressureLeakException error	Pressure test failure during plate loading.	1. Inspect the plate and ensure that all gaskets are applied correctly, then place the plate back into the Absolute Q™ nest 2. Select Restart the Run Template operation to attempt to restart the plate. 3. If the error is displayed again, select Abort this iteration of the experiment to skip the iteration entirely and continue with the remaining plates in the batch. 4. If the error continues to appear for subsequent plates, contact Technical Support.
Barcode is not detected error	Barcode read error.	1. Inspect the plate and ensure that it is in the correct orientation in the Absolute Q™ nest 2. Select Restart the Run Template operation to attempt to restart the plate run. 3. If the error is displayed again, select Abort this iteration of the experiment to skip the iteration entirely and continue with the remaining plates in the batch. 4. If the error continues to appear for subsequent plates, contact Technical Support.
(404) Not Found error	Instrument connection error.	1. Ensure the instrument is powered on and inspect all cables and connections. 2. Bring the affected Absolute Q™ instrument offline in the Momentum™ software. 3. Power cycle the Absolute Q™ instrument. 4. Bring the affected Absolute Q™ instrument back online in the Momentum™ software. 5. Select Restart the Access operation. 6. If the error continues to appear, contact Technical Support.

Observation	Possible cause	Recommended action
Cannot locate file error	DPR file not found for corresponding plate barcode.	1. Navigate to the DPR Barcoded Templates folder and confirm that a DPR file exists for the plate barcode being run. The file name should contain the barcode. 2. Select Restart the Run File operation. 3. If the error is displayed again, select Abort this iteration of the experiment to skip the iteration entirely and continue with the remaining plates in the batch. 4. If the error continues to appear for subsequent plates, contact Technical Support.
Spinnaker operation did not complete error	Plate is not present in nest when expected.	1. Confirm a plate is present in the expected location as indicated in the Momentum™ message window. 2. Select Retry the grip check sub-step of the Get operation. 3. If the error is displayed again, select one of the following options: • Manually move the plate to the nest, then select Manually complete the Get operation. • Select Abort this iteration of the experiment to skip the iteration entirely and continue with the remaining plates in the batch • If the error continues to appear for subsequent plates, contact Technical Support.
	The Spinnaker™ instrument movement is interrupted.	1. Inspect and clear any physical obstructions on the deck. 2. Select Restart the Get operation. 3. A new Restore Mover Spinnaker Momentum™ message may appear. If the message appears, select Restore & Continue. The Spinnaker™ instrument will home one or multiple motion axes before attempting to restart the operation.
	The emergency stop button is accidentally pressed.	1. Ensure that the Red Reset button is popped up. Twist button if it is depressed. 2. Confirm the DC Power switch is set to ON. 3. Press the MOTOR ON button. When the MOTOR ON button turns green, the Spinnaker™ instrument is fully powered. 4. Confirm that the Spinnaker™ instrument is online in the Momentum™ software. 5. Select Restart the Get operation.
TimeoutException error	Instrument error.	1. Bring the affected Absolute Q™ instrument offline in the Momentum™ software. 2. Power cycle the Absolute Q™ instrument. 3. Bring the affected Absolute Q™ instrument back online in the Momentum™ software. 4. Select Restart the Access operation. 5. If the error continues to appear, contact Technical Support.



Observation	Possible cause	Recommended action
Other instrument errors	Instrument error.	1. Bring the affected Absolute Q™ instrument offline in the Momentum™ software. 2. Power cycle the Absolute Q™ instrument. 3. Bring the affected Absolute Q™ instrument back online in the Momentum™ software. 4. Select an option to restart the operation. 5. If the error continues to appear, contact Technical Support.



Field Service Archive files

Field Service Archive (FSA) files contain information regarding runs and instrument usage that can be used for troubleshooting unexpected run results and instrument performance. The following table provides information on the FSA files that can be captured from the QuantStudio™ Absolute Q™ Digital PCR System.

Note: For implementations using AutoRun Suite, FSA files are automatically exported to the Momentum™ computer. See “Files generated during an AutoRun Work Unit” on page 149.

File type and file name format	Description
Data {run_name}_{short_run_id}_{YYYY_MM_DD}_data.fsa	• Used for troubleshooting issues with a run • Automatically created with each run • File size can be large: >1GB • 20 most recent files retained
Log {run_name}_{short_run_id}_{YYYY_MM_DD}_logs.fsa	• Used for troubleshooting issues with a run • Automatically created with each run • File size is ~120 MB • 70 most recent files retained
System {YYYY_MM_DD}_system.fsa	• Used for troubleshooting system issues not related to a run • Contains logs from the desktop computer and instrument computer at the time of capture • Created on demand • File size is ~120 MB • The file is not automatically deleted

Capture and transfer data and log FSA files

Data and log FSA files are used for troubleshooting unexpected results or instrument failure during a run.

Note: Only use these instructions when instructed by a Thermo Fisher Scientific support representative.

1. Capture and transfer data and log FSA files using one of the following options based on your implementation.

Implementation	Action
Standalone (non-automated) Absolute Q™ system.	1. On the desktop computer, open the Start Menu. 2. Locate the shortcut to the QuantStudio Absolute Q 6 Field Service Archives folder by performing one of the following actions. • In the search field, type QuantStudio Absolute Q 6 Field Service Archives. • Scroll through the application list. 3. Click on the shortcut to open the archive files folder, then select the data and log files for the run in question. For example: • <i>Absolute Q Starter Chemistry Run 31_8ea5ccfc_2022_04_01_data.fsa</i> • <i>Absolute Q Starter Chemistry Run 31_8ea5ccfc_2022_04_01_logs.fsa</i>
AutoRun Suite	1. On the Momentum™ computer, use the file browser to navigate to D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Data Output. 2. Select the file name for the run in question. FSA files use the following naming convention. <run_name>_AutoRun_<iso8601_localtime>_<barcode>.zip

2. Send the files to your Thermo Fisher Scientific support representative for analysis using a file transfer program of your choosing.



Capture and transfer system FSA files

System FSA files are used for troubleshooting system issues not related to a run, for example if the plate tray is malfunctioning. A system FSA file is created on demand using the QuantStudio™ Absolute Q™ Digital PCR Software on the dedicated computer for the QuantStudio™ Absolute Q™ Digital PCR Instrument.

Note: Only use these instructions when instructed by a Thermo Fisher Scientific support representative.

Before you begin, ensure that the QuantStudio™ Absolute Q™ Digital PCR Software is connected to the QuantStudio™ Absolute Q™ Digital PCR Instrument that is powered on.

1. In the QuantStudio™ Absolute Q™ Digital PCR Software, select  **System**, then in the **Download system logs** area, select **DOWNLOAD SYSTEM FSA**.
The system log file is created and a **File Explorer** window opens with the system log file name pre-populated in the file name field.
2. Navigate to a folder of your choice, then click **Save**.
3. Send the files to your Thermo Fisher Scientific support representative for analysis using a file transfer program of your choice.



Supplemental information

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Determine the optimal sample dilution

Is the target copy number per genome is known?	Proceed to
Yes	Determine the optimal dilution when the target is known (page 124)
No	Determine the optimal dilution when the target is unknown (page 126)

Determine the optimal dilution when the target is known

In a dPCR experiment, gDNA samples are diluted to a limiting quantity, to the extent that most individual PCR reactions contain either zero or one target molecule. If the target copy number per genome is known, dilute the extracted DNA to the optimal input range using the following workflow.

1. Determine the target copy number per genome (page 124)
2. Dilute the extracted genomic DNA to the ideal input range (page 125)

Determine the target copy number per genome

This section provides example calculations for determining the target copy number per genome. Other calculation methods can be used. For information about the human genome, see "On the length, weight and GC content of the human genome", Piovesan et al. BMC Res Notes (2019) 12:106 https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6391780/pdf/13104_2019_Article_4137.pdf.

1. If the source or species of the gDNA is known, using a genome size checker tool, determine the size of the genome. The lengths of several common genome models can be found at: <http://www.thermofisher.com/DNA-calculator>.

The size checker estimate of the single human genome is 3.2×10^9 bp (haploid).

2. Using the size of the genome determined in step 1, calculate the genome mass using the following formula.

$m = (n) (1.096 \times 10^{-21} \text{ g/bp})$, where 'm' is the genome mass, and 'n' is the genome size in base pairs

The following example calculates the mass of the human genome using the estimate of 3.2×10^9 bp (haploid) for (n).

$$m = (3.2 \times 10^9 \text{ bp}) (1.096 \times 10^{-21} \text{ g/bp})$$

$$m = 3.5 \times 10^{-12} \text{ g or } 3.5 \text{ pg}$$

3. Using the mass of the genome calculated in step 2, refer to a public database of genomic variants to identify the copies of the target sequence per single genome. For example, <http://dgv.tcag.ca/dgv/app/home>.

The following example determines the genomic copy ratio to the mass of the human genome of the RNase P gene (single exon RPPH1 gene) located on chromosome 14 cytoband 14q11.2. (chr.14:20343370 on build GRCh38).

RNase P gene copies per haploid human genome mass: 1 copy/3.5 pg.

In other words, 1 copy of the RNase P target sequence can be found in every 3.5 pg of human DNA. This example is relevant to any gene that is present at the normal rate of one copy per haploid genome (two copies per diploid genome) and provides a basis to perform a dPCR experiment to determine the optimal digital range.

Note: The digital range is the dynamic range of a dPCR instrument.

Dilute the extracted genomic DNA to the ideal input range

1. Based on the known target copy number per genome, dilute the samples to be within the dynamic range of the digital platform. The optimal digital range is such that each reaction well contains approximately 0.6–1.6 copies of the target sequence.

Note: A much wider range of concentrations can be run, however the concentration between 0.6 and 1.6 copies achieves the greatest confidence.

To calculate the desired dilution point, divide the target copies per reaction by the volume of each microchamber's reaction, for example 0.6 to 1.6 copies divide by 0.000432 μL .

2. Take the resulting copies/ μL of reaction mix and divide by the copies per picogram value determined in "Determine the target copy number per genome" on page 124 to arrive at a genomic pg/ μL concentration target.

For example:

$$\frac{[12,926.96 \text{ pg gDNA}]}{([1.6 \text{ cp RNase P}] / [0.000432 \mu\text{L}])} \times (p3.5 \text{ pg gDNA} / [1 \text{ cp RNase P}])$$

- Once your target concentration is identified, construct a dilution series to dilute your genomic material to the correct final concentration. For an example of serial dilution, see Table 12.

IMPORTANT! Be sure to include the final addition of your sample into the reaction mix as the final dilution step.

Table 12 Serial dilution example

Tube	μL from previous dilution	μL diluent	Total tube μL	Conc. (pg/μL)	Total dilution factor
Stock	—	—	—	6.10×10^7	4.7×10^2
D1	3.28	196.72	200	1.00×10^6	
D2	10.00	190.00	200	5×10^4	
Reaction mix	2.59	7.41	10	12962.96	

- The total dilution factor shown in Table 12 can be used during run setup to automatically back-calculate the stock concentration from the dPCR concentration results. The dilution factor can be calculated by dividing the stock concentration by the final reaction mix concentration.

Determine the optimal dilution when the target is unknown

If the target copy number per genome is unknown (for example, for a locus of unknown copies per genome or RNA of unknown expression level), we recommend that you determine the optimal dilution by preparing a dilution series of the sample that includes three to four data points above and below the expected digital range. This helps ensure that one of the data points is within the optimal digital range.

The quantification cycle (C_q) value is a function of concentration, therefore 1 copy target sequence in different reaction volumes produces different C_q values. Additionally, the actual C_q value in real time PCR always depends on the primary analysis parameters set by the user (for example, baseline, and threshold).

If tested using real-time PCR, the C_q values can be used to estimate the target molecule input for the points of the dilution series prior to dPCR.

- 1 copy in total volume of 20 μL produces C_q of ~38 (96-well plate)
- 1 copy in total volume of 10 μL produces C_q of ~37 (384-well plate)
- 1 copy in total volume of 1.5 μL produces C_q of ~34.5 (TaqMan™ Array Card)
- 1 copy in total volume of 33 nL produces C_q of ~29 (OpenArray™ Plate)

Computation of results

The following table describes formulas that are used to calculate results.

Table 13 Computation of results

Result	Formula(e)	Description
Lambda(cp/Rxn) Lambda (copies/reaction)	$\lambda = -\ln(z/n)$	λ —the average number of molecules per microchamber or reaction
Lambda_95%CI Lower and upper 95% confidence intervals for Lambda (copies/reaction)	$\Delta_{lower} = \lambda \left[1 - e^{\frac{-1.96\sqrt{e^\lambda - 1}}{\lambda\sqrt{n}}} \right]$ $\Delta_{upper} = \lambda \left[e^{\frac{1.96\sqrt{e^\lambda - 1}}{\lambda\sqrt{n}}} - 1 \right]$	z —the number of empty microchambers in the sample n —the number of accepted (non-rejected) microchambers
Precision %	$P = 100 \times \max(\Delta_{lower}, \Delta_{upper}) / \lambda$ Where, $\Delta_{lower} = \lambda \left[1 - e^{\frac{-1.96\sqrt{e^\lambda - 1}}{\lambda\sqrt{n}}} \right]$ $\Delta_{upper} = \lambda \left[e^{\frac{1.96\sqrt{e^\lambda - 1}}{\lambda\sqrt{n}}} - 1 \right]$	P —the spread of the confidence level around λ compared to the true λ value $\max$ —the higher of the two values
Conc. copies/μL Concentration (copies/μL)	$C = \frac{\lambda}{V} \times DF$	C —the sample concentration in copies per microliter V —the volume of each microchamber (432 pL)
95%CI Lower and upper 95% confidence intervals for Concentration (copies/μL)	$\Delta_{lower} = \frac{\lambda}{V} \left[1 - e^{\frac{-1.96\sqrt{e^\lambda - 1}}{\lambda\sqrt{n}}} \right] \times DF$ $\Delta_{upper} = \frac{\lambda}{V} \left[e^{\frac{1.96\sqrt{e^\lambda - 1}}{\lambda\sqrt{n}}} - 1 \right] \times DF$	DF —the dilution factor specified during plate setup
Copy # or Copies (for each sample) Copy Number Variation	$CNV = N_r \times \frac{\lambda_t}{\lambda_r}$	CNV —the estimated copy number N_r —the number of copies per genome of the reference target
Copies (group summary table, see Figure 29 callout ③) Average copy number for the group	The software algorithm uses the concentration values to calculate the average group copy number. First, the software calculates the average target concentration from all the target concentrations and the average reference concentration from all the reference concentrations. The software then uses the average target and reference concentrations to calculate the copy number (copies) for the group.	λ_t —the estimated average number of molecules per microchamber of the target λ_r —the estimated average number of molecules per microchamber of the reference

Table 13 Computation of results (continued)

Result	Formula(e)	Description
CNV_95% CI Lower and upper 95% confidence intervals for copy numbers	$\Delta_{lower} = \left[e^{\ln \frac{\lambda_t}{\lambda_r}} - e^{\ln \frac{\lambda_t}{\lambda_r} - 1.96 \sigma_R} \right] N_r$ $\Delta_{upper} = \left[e^{\ln \frac{\lambda_t}{\lambda_r} + 1.96 \sigma_R} - e^{\ln \frac{\lambda_t}{\lambda_r}} \right] N_r$ Where, $\sigma_R = \sqrt{\frac{1 - e^{-\lambda_t}}{n \lambda_t^2 e^{-\lambda_t}} + \frac{1 - e^{-\lambda_r}}{n \lambda_r^2 e^{-\lambda_r}}}$	CNV —the estimated copy number N_r —the number of copies per genome of the reference target λ_t —the estimated average number of molecules per microchamber of the target λ_r —the estimated average number of molecules per microchamber of the reference
% VAF Variant Allele Frequency (VAF, %)	$VAF = (M_t / (M_t + M_r)) \times 100$	VAF —Variant Allele Frequency (%) M —the estimated molecules per reaction
95 % CI VAF Lower and upper 95% confidence intervals for VAF	$\Delta_{lower} = VAF - 100 \times p_L$ $\Delta_{upper} = 100 \times p_U - VAF$ Where, $p_L = \text{Beta}^{-1}(0.025; M_t, M_r + 1)$ $p_U = \text{Beta}^{-1}(0.975; M_t + 1, M_r)$	n —the total number of microchambers (partitions) analyzed λ —the average number of molecules per microchamber t —designates the variant channel r —designates the wild-type channel
Log % VAF Note: Defined only if VAF > 0.	$\text{Log \% VAF} = \log_{10}(VAF)$	Beta^{-1} —inverse Beta distribution

QC failure and warning messages

The following table defines the QC flags that can occur and recommended corrective actions.

Table 14 QC messages

QC status (flag)	QC message	Description	Recommendation
 CRITICAL	CRITICAL: Image registration error in [Dye name(s)]	The microchamber array cannot be located or identified with sufficient accuracy. This can be caused by excessive dust or debris on the plate or if the microchamber array was filled poorly.	• Ensure that any unused dye channels are disabled. For example, if the assay only contains FAM™ and VIC™ dyes, disable the other dye channels. • Ensure environmental cleanliness. • Ensure reaction mixture is made up properly^[1].
 FAILURE	FAILURE: Array QC failure	QC problem or problems with the sample have been detected that prevent accurate analysis. Potential causes include loading issues or other failure modes.	Severe failures are rare but can occur. • Ensure environmental cleanliness. • Vortex and centrifuge reagents thoroughly in each step and prior to loading samples^[1]. • Ensure that any unused dye channels are disabled. For example, if the assay only contains FAM™ and VIC™ dyes, disable the other dye channels.
 WARNING	WARNING: Bridging between microchambers	A statistically improbable string of positive microchambers adjacent to each other that can indicate that amplifiable material is being shared between microchambers have been detected. This can be caused by pipetting errors or setting incorrect thresholds.	• Ensure that only 9 µL of reaction mixture is loaded into each sample well and pipettes are calibrated. • Ensure that thresholds for all dyes are set accurately.
 WARNING	WARNING: Camera saturation in: [Dye name(s)]	Microchambers with high fluorescence that saturate the camera detector have been detected. This can be caused by a high concentration of probes.	To resolve the saturation, reduce the concentration of probes ^[1] .

Table 14 QC messages (continued)

QC status (flag)	QC message	Description	Recommendation
 WARNING	WARNING: High rejection rate from QC bounds	A high number of microchambers rejected due to signal levels outside of QC channel bounds are detected. This could be caused by incorrect filling or incorrect bounds.	• Ensure that the minimum and maximum bounds in the QC Dye are set accurately. • Ensure that the correct master mix and correct dilution of master mix are used.
 WARNING	WARNING: Uneven distribution of positive <dye name>	Microchambers exhibit irregular spatial distribution of positive signals, which can indicate contamination, uneven loading, or debris affecting fluidics and imaging.	• Verify proper sample preparation and handling procedures. • Clean the working area to minimize contamination. • Adjust the dye thresholds.
 WARNING	WARNING: Overloaded array in: [Dye name(s)]	No negative microchambers were detected. This impacts computation based on Poisson statistics. Possible causes: • Samples were run at a high concentration through a suboptimal dilution. • Incorrect thresholds were set in the affected dyes.	• Ensure that normal concentration is correct. • Ensure that thresholds for the overloaded dyes are set correctly.
PASS	PASS: No QC issues found	No widespread problems with the sample have been detected. The number of rejected microchambers, if any, are in acceptable bounds.	Proceed with regular analysis workflow.

^[1] If failures or warnings reoccur, save the data FSA files for use in troubleshooting.

Configure system settings

Systems settings are optional and are configured based on your implementation of the system.

1. In the left navigation pane, click  **System Settings** to open **System Settings**.
2. Configure each setting as needed for your implementation.

Setting	Action
Enable security When toggled on, this setting requires users to sign in with an ID and password. Note: For implementations using the AutoRun Suite, security is configured using the Momentum™ Workflow Scheduler Software.	Perform one of the following actions. • To enable, toggle the switch to the on position. • To disable, toggle the switch to the off position. If enabled, perform the following actions in the SAE connection settings dialog. 1. In the IP address field, enter the IP address of the SAE Administrator Console. 2. In the Port field, enter the port number of the SAE Administrator Console. 3. Click TEST CONNECTION to confirm successful communication to the SAE Administrator Console. 4. Enter a user name and password for an SAE system administrator. For implementations using the Security, Auditing, and E-signature (SAE) v2.2 or later software, use this setting to configure the connection to the SAE Administrator Console.
Remote API settings For implementations using automation other than the AutoRun Suite, use this setting to configure access to the Remote API using an access code. The Remote API settings provides the following information. • Port—System assigned Port for the Remote API • Fingerprint—The hash of the SSL certificate. This is used as an identifier to ensure connection to a valid Remote API server For information about using automation with a third party system, see <i>QuantStudio™ Absolute Q™ Digital PCR System Remote API User Guide</i> (Pub No.MAN0028059)	Perform the following actions. 1. To enable, toggle the switch to the on position. 2. To enter an access code, perform one of the following actions. • Click GENERATE ACCESS CODE to create a system generated access code. • In the Access Code field, manually enter an access code. 3. Click SAVE in the top-right corner of the screen. 4. Close the QuantStudio™ Absolute Q™ Digital PCR Software, then restart the computer for the settings to take affect. Note: Failure to restart the computer prevents the instrument from connecting to the automated system.

(continued)

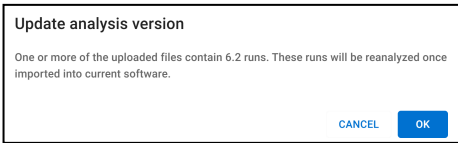
Setting	Action
Batch export location For implementations using automation, use this setting to set a file system location for export of batched runs.	Click BROWSE , then navigate to the location on the file system where batched run files are to be exported.
Run import location Use this setting to set the file system location for runs that are available for import.	Click BROWSE , then navigate to the location on the file system where run files for import are located.
Auto export completed runs Use this setting to set the data types and file system location for the export of information from manual runs. The following data files are available for export. • ZST files—Run information in ZST file format. • Run data CSV files—Run information in Excel™ spreadsheet file format. • Run results CSV files—Run results information in Excel™ spreadsheet file format.	Perform the following actions for each data type to be exported. 1. Select each data type to be exported. Note: The Results summary CSV table format was updated for QuantStudio™ Absolute Q™ Digital PCR Software v6.4 and later. To download the Results summary CSV in v6.3 format, select the 6.3 format (legacy). 2. Click BROWSE, then navigate to the location on the file system where the run files are to be exported.
Storage management Use the setting to determine the number of runs to save before the oldest run is deleted.	Perform the following actions to customize the number of runs to save before deletions. 1. To enable, toggle the switch to the on position. 2. In the Runs to retain field, enter the number of runs to save.
Download system logs Use this option to download Field Service Archive (FSA) system logs for use in troubleshooting.	1. Click DOWNLOAD SYSTEM FSA. 2. In the File explorer, navigate to the location to save the files, then click SAVE. 3. Send the files to your Thermo Fisher Scientific support representative for analysis using a file transfer program of your choosing. Note: For more information about Field Service Archive files, see “Field Service Archive files” on page 121
Enable pressure leak detection <i>(Recommended)</i> Toggle this option on to enable the pressure leak detection warnings.	To navigate to the Enable pressure leak detection setting, expand the Advanced settings actions at the bottom of the screen. Toggle the switch on or off as needed.
Automatically omit failed QC samples <i>(Recommended)</i> Toggle this option on to set the software to automatically omit the samples with flagged with QC failure (🔴) from all analyses.	To navigate to the Automatically omit failed QC samples setting, expand the Advanced settings actions at the bottom of the screen. Toggle the switch on or off as needed.

3. When all settings needed for your implementation are configured, click **SAVE** in the top-right corner of the screen.

Update the analysis version

If a run was analysed using QuantStudio™ Absolute Q™ Digital PCR Software version earlier than 6.4, the run can be re-analyzed using the latest QuantStudio™ Absolute Q™ Digital PCR Software version.

Importing a completed run that was analyzed using QuantStudio™ Absolute Q™ Digital PCR Software version earlier than 6.4 automatically triggers the following configuration changes.

Imported run QuantStudio™ Absolute Q™ Digital PCR Software analysis version	Configuration changes made upon import into QuantStudio™ Absolute Q™ Digital PCR Software version 6.4
6.2	The Update analysis version dialog appears.  - Clicking OK automatically triggers re-analysis. Note: You cannot view the original analysis. - Clicking Cancel cancels the run import. Upon successful import, the JUN™ targets are remapped to the Red (JUN) channel. For more information, see “QuantStudio™ Absolute Q™ Digital PCR Instrument Optical Configuration” on page 191.
6.3	Upon successful import, the JUN™ targets are remapped to the Red (JUN) channel. For more information, see “QuantStudio™ Absolute Q™ Digital PCR Instrument Optical Configuration” on page 191. When you open the imported run for the first time, you can select to view the original analysis or update the analysis to the latest version of the QuantStudio™ Absolute Q™ Digital PCR Software (see step 1–step 3 below).

1. In the left navigation pane, click  **Runs**, then navigate to the **COMPLETED** tab.
2. Click a run name to open a run.
If a run was last analyzed using QuantStudio™ Absolute Q™ Digital PCR Software version 6.3, you are prompted to update the analysis version.
3. In the **Update analysis version** dialog, select one of the following options.

Option	Description
UPDATE ANALYSIS	The run is re-analyzed using the latest version of the QuantStudio™ Absolute Q™ Digital PCR Software. No further action is required.
VIEW ORIGINAL ANALYSIS	The run is not re-analyzed and the original analysis is displayed. When you elect to view the original analysis, a banner appears at the top of the page that persists until the run is re-analyzed using the latest version of the QuantStudio™ Absolute Q™ Digital PCR Software. To dismiss the banner, make any modification to the run, then click SAVE . Saving the run triggers reanalysis using the latest version of the QuantStudio™ Absolute Q™ Digital PCR Software.



Install, update, and move the QuantStudio™ Absolute Q™ Digital PCR System

■ Installation and environmental requirements for standalone instruments	135
■ Installation and environment requirements for the AutoRun Suite	137
■ Install the QuantStudio™ Absolute Q™ Digital PCR System	137
■ Download, install, and update the software	138
■ Moving the instrument	139

Installation and environmental requirements for standalone instruments

The room where the instrument is installed must be kept within the following operational environment conditions.

Condition	Acceptable range
Installation site	For indoor laboratory use only (Applicable pollution degree 2)
Operating temperature and humidity	15-30°C (60-85°F), 0-80% RH
Storage temperature and humidity	5-40°C (40-105°F), 0-80% RH
Vibration	Do not place the instrument adjacent to strong vibration sources. Excessive vibration during use can affect instrument performance.
Altitude	Up to 6,500 ft (2,000 m)



(continued)

Condition	Acceptable range
Input voltage tolerance	+/-10%
Over voltage category	II

- Installation time: <10 minutes
- Required materials: scissors or a strap cutter
- Space requirement: The instrument is approximately 0.6 m (2 ft) cubed. The presentation drawer must not be obstructed and extends approximately 200 mm (8 in) from the front panel of the instrument when open. The power and USB connections are on the left side near the back of the instrument.
- Ensure that the fan vents on the back and bottom of the instrument are not obstructed.

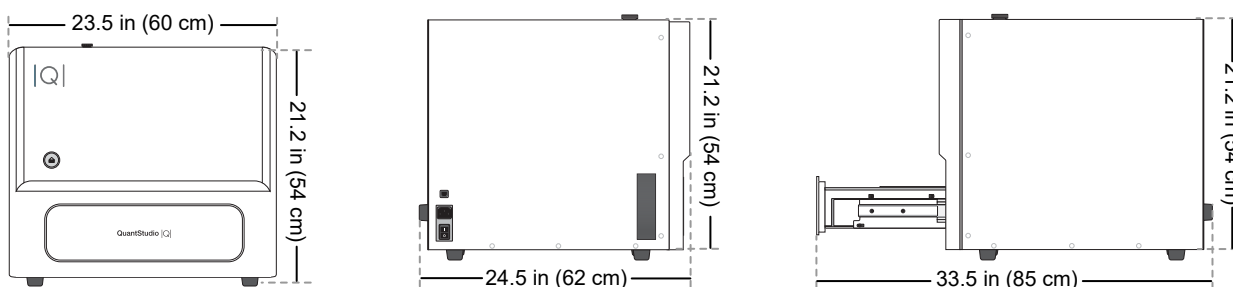


Figure 30 Instrument dimensions

IMPORTANT! Keep all packaging materials in good condition, as they are required if the instrument needs to be returned for any reason.



WARNING! The instrument requires 2–3 people for moving. Moving the system alone may result in serious injury.



AVERTISSEMENT ! Le déplacement de l'instrument nécessite 2 à 3 personnes. Si vous déplacez le système seul, vous risquez de vous blesser gravement.



Installation and environment requirements for the AutoRun Suite

The room where the instrument is installed must be kept within the following operational environment conditions.

Ensure that the fan vents on the back and bottom of the instrument are not obstructed.

Condition	Acceptable range
Installation site	For indoor laboratory use only (Applicable pollution degree 2)
Operating temperature and humidity	15–30°C (60–85°F), 20–80% RH
Storage temperature and humidity	5–40°C (40–105°F), 20–80% RH
Vibration	Do not place the instrument adjacent to strong vibration sources. Excessive vibration during use can affect instrument performance.
Altitude	Up to 6,500 ft (2000 m)
Input voltage tolerance	+/-10%
Over voltage category	II

IMPORTANT! Keep all packaging materials in good condition, as they are required if the instrument needs to be returned for any reason.



WARNING! The instrument requires 2–3 people for moving. Moving the system alone may result in serious injury.



AVERTISSEMENT ! Le déplacement de l'instrument nécessite 2 à 3 personnes. Si vous déplacez le système seul, vous risquez de vous blesser gravement.

Install the QuantStudio™ Absolute Q™ Digital PCR System

IMPORTANT! Ensure that the installation location meets the power and environmental requirements specified in “Installation and environmental requirements for standalone instruments” on page 135.

1. With 2–3 people, carefully unbox the instrument by cutting the straps and lifting the top of the box off using the hand holes.
Do not cut or damage any of the packaging. Keep all packaging as it is required for returns or service requests.
2. Carefully place the instrument on a flat, stable surface with no adjacent vibration sources.



3. Position the instrument so that there is access to the power and USB connectors on the left side of the system.
4. Once the instrument is in place, remove the two shipping lock screws, one screw at the top of the instrument and one door lock screw.
 - a. With the power off, unscrew the shipping lock screws.
 - b. Insert the provided white plastic caps into screw holes.

For more information on removing the shipping lock screws, see “Uninstall the shipping lock screws” on page 140.

IMPORTANT! To prevent damage to the instrument, the shipping lock screws must be removed before powering on the instrument.

Keep the shipping lock screw in case the instrument needs to be moved or returned for service.

5. Confirm that the power switch is in the OFF, O, position and then connect the power cable to the instrument and a suitable power source.
6. Set up the dedicated computer and monitor near the instrument.
7. Use the power cable to connect the dedicated computer to a suitable power source.
8. Connect the keyboard and mouse to the back of the dedicated computer.
9. Power on the dedicated computer.

IMPORTANT! To prevent damage to the instrument, only power on the dedicated computer at this step, do not power on the instrument.

10. Install the software onto the dedicated computer. See “Download, install, and update the software” on page 138.
11. When the software installation is complete, use the USB cable to connect the instrument to the dedicated computer.
12. Power on the instrument by moving the power switch located at the left side near the back to the I position.

Wait approximately 30 seconds for the instrument to initialize.
13. Once connected to the software, check that there are no errors reported.

The system is ready for use.

Download, install, and update the software

For instructions about downloading, installing, and updating the QuantStudio™ Absolute Q™ Digital PCR Software, including information about updating the embedded QuantStudio™ Absolute Q™ Digital PCR Instrument software and firmware, see the *QuantStudio™ Absolute Q™ Digital PCR Software Installation Guide* (Pub. No. MAN1001443).



Moving the instrument

IMPORTANT! When moving the instrument, the shipping lock screws must be manually installed before moving the unit and manually removed after transport. Moving the instrument without the shipping lock screws in place can cause damage to the instrument.

IMPORTANT! When moving the instrument, make sure there is no plate in the instrument as it can become dislodged and jam mechanical parts during instrument transport.

IMPORTANT! Use clinging plastic wrap or painter's tape to secure the tray door and prevent it from opening during transport.

Note: It is not recommended that FSE pack or unpack the instrument.

Install the shipping lock screws

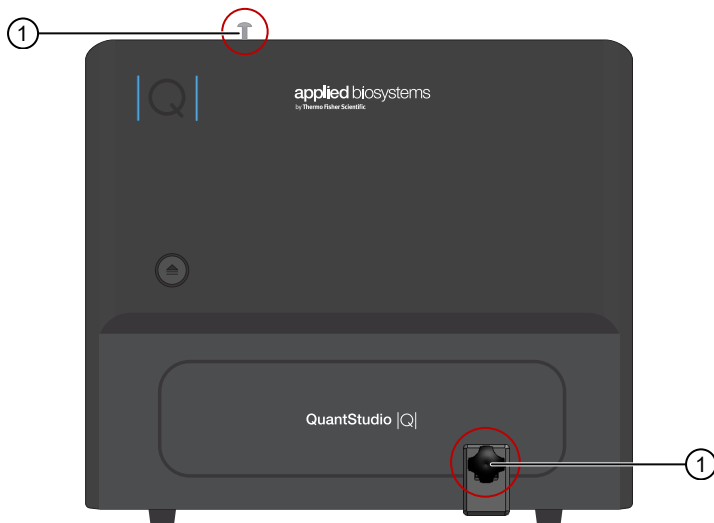
1. Power on the instrument.
2. Launch the QuantStudio™ Absolute Q™ Digital PCR Software.
3. Open the plate door to ensure there is no plate loaded. If a plate is loaded, remove it.
4. Close the plate door.
5. In the left navigation pane, click **Instrument** to access the **Instrument** screen.
6. Click on the instrument, then click **Prepare for Shipping**.
Wait until a message stating *Ready for Shipping* appears before proceeding.
7. Remove the white plastic plug from each shipping screw hole, one at the top of the instrument and one on the plate door. Place each plug in the bag attached to the shipping screws.
8. Insert each shipping screw into the corresponding shipping screw hole and screw it finger-tight. Do not over tighten.
If the shipping screws cannot be screwed in, then the shipping screw offsets might need to be recalibrated. Follow the procedure to Calibrate the shipping lock screws.
9. Close the QuantStudio™ Absolute Q™ Digital PCR Software, then power off the instrument.



Uninstall the shipping lock screws

IMPORTANT! Perform this task before powering on the instrument.

1. Ensure that the power is off and the instrument is not plugged into a power source.
2. Unscrew the two shipping lock screws, one at the top of the instrument and one on the plate door.



① Shipping lock screws

3. Insert the white plastic cap into each shipping lock screw hole.
The instrument is now ready for power-up and use.



Automate runs with the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite

■ Momentum™ Workflow Scheduler Software key concepts	142
■ Power on and start the AutoRun Suite	143
■ Perform an AutoRun using the Spinnaker™ Microplate Mover	144
■ Add more plates during an AutoRun Work Unit with hotels	148
■ Files generated during an AutoRun Work Unit	149
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■ Momentum™ Security	150

The AutoRun Suite uses the Absolute Q™ Digital PCR system with the Thermo Fisher Scientific Spinnaker™ Microplate Mover to manage loading and unloading plates in an automated workflow. The Momentum™ Workflow Scheduler Software running on a dedicated computer, lets you manage AutoRun multi-plate batches using the Spinnaker™ Microplate Mover.

IMPORTANT! For optimal system performance and stability, it is recommended to power-cycle the Absolute Q™ Digital PCR instrument(s) before starting each Work Unit. Multiple batches can be added to a single Work Unit when using a hotel configuration, but it is recommended not to exceed 80 plates on a single configuration or 160 plates on a dual configuration in a single Work Unit. To power-cycle the AutoRun Suite, refer to “Power off the AutoRun Suite” on page 150.

IMPORTANT! When using the AutoRun Suite, the Absolute Q™ dedicated computer is only used for instrument control and troubleshooting. Do not use the Absolute Q™ software on the Absolute Q™ dedicated computer when an AutoRun is processing in the Momentum™ software. Doing so may interfere with the AutoRun system.

Momentum™ Workflow Scheduler Software key concepts

The Momentum™ Workflow Scheduler Software lets you automate experiment runs using the Spinnaker™ Microplate Mover and the Absolute Q™ instrument. This section provides information about terminology used in the Momentum™ software when scheduling an AutoRun Work Unit.

- **Database**—The Momentum™ database file (SDF) contains a set of devices and experiments for a specific system. There are 4 types of databases available with AutoRun.
 - Single Configuration with Hotels
 - Dual Configuration with Hotels
 - Single Configuration with Stacks
 - Dual Configuration with Stacks
- **Work Unit**—A set of operations that Momentum™ processes according to the order they appear in the **Work Queue**.
- **Batch**—A defined number of plates being run with one specific experiment type. A hotel configuration supports adding multiple batches in a **Work Unit** and each batch can be assigned a different priority level.
- **Experiment**—A set of operations designed for a workflow. The AutoRun databases have 2 types of workflows.
 - **DPT Runs**—These runs use a single protocol defined in a run template file (DPT) to perform the run on each plate in a Momentum™ batch. For information about creating run templates, see “Manage templates” on page 40.
 - **DPR Runs**—These runs use a separate draft run file (DPR) for each plate. The DPR file is associated with the plate using the plate barcode. This enables different protocols to be run on individual plates in the same Momentum™ batch. For information about creating multiple DPR files for use in the Momentum™ batch, see “Generate batch runs from a template” on page 51. For information about creating a single DPR file, see “Create a run from the Runs screen” on page 55.

The Momentum™ Workflow Scheduler Software Dashboard lets you manage AutoRun batches.

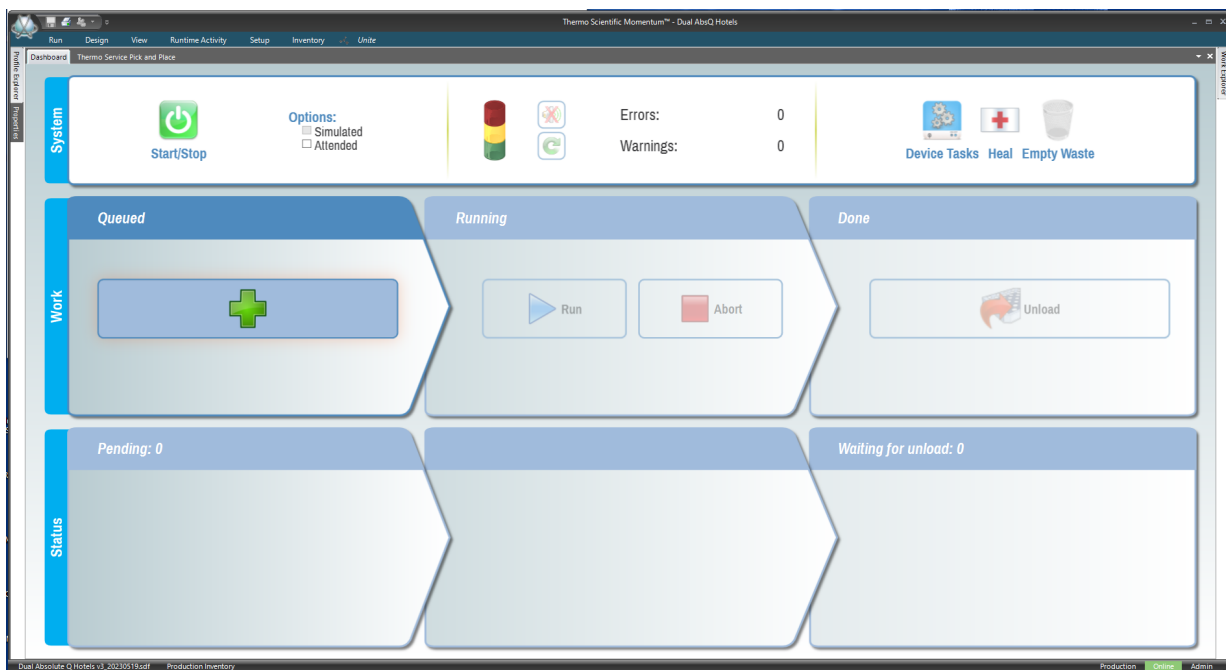


Figure 31 Momentum™ Workflow Scheduler Software Dashboard

Power on and start the AutoRun Suite

IMPORTANT! When using the AutoRun Suite, the Absolute Q™ dedicated computer is only used for instrument control and troubleshooting. Do not use the Absolute Q™ software on the Absolute Q™ dedicated computer when an AutoRun is processing in the Momentum™ software. Doing so may interfere with the AutoRun system.

One monitor, keyboard, and mouse controls all of the dedicated computers that are part of the AutoRun Suite. Use the following options to switch between the computers.

Computer	Keyboard shortcut
Momentum™	On the keyboard, press <CTRL>, <CTRL>, 1.
Absolute Q™ left computer	On the keyboard, press <CTRL>, <CTRL>, 2.
Absolute Q™ right computer (dual configurations only)	On the keyboard, press <CTRL>, <CTRL>, 3.

1. Power on the Absolute Q™ dedicated computer running the QuantStudio™ Absolute Q™ Digital PCR Software and sign in using the local administrator account.

Note: For dual configurations, perform this task on both dedicated computers.

2. Power on the Spinnaker™ dedicated computer running the Momentum™ Workflow Scheduler Software using the steps that follow.
 - a. Switch the monitor to the dedicated computer for the Spinnaker™ (using the Momentum™ Workflow Scheduler Software) and power on the computer.

- b. Sign in using the local administrator account.
 - c. On the desktop of the dedicated computer, click the Momentum™ shortcut to start the software.
3. Power on the Spinnaker™ instrument using the steps that follow.
 - a. On the Spinnaker™ power puck, ensure that the red **RESET** button is popped-up.
 - b. Move the **DC POWER** switch on the front of the power puck to the on position.
 - c. Press the **MOTOR ON** button on the top of the power puck.
The Spinnaker™ is ready when the **MOTOR ON** light is a steady green.
4. Power on the Absolute Q™ instrument by moving the power switch on the left side near the back of the instrument to the I position.

Note: The instrument makes a humming noise as it charges the internal compressor.

The bars of the instrument symbol flash white to indicate that the system is initializing. This takes approximately 30 seconds.

The Absolute Q™ instrument is ready when the status lights are a steady blue and a ready status appears under the instrument in the **Instrument** screen in the QuantStudio™ Absolute Q™ Digital PCR Software.

Note: For dual configurations, perform this task on both Absolute Q™ instruments.

Perform an AutoRun using the Spinnaker™ Microplate Mover

IMPORTANT! When using the AutoRun Suite, the Absolute Q™ dedicated computer is only used for instrument control and troubleshooting. Do not use the Absolute Q™ software on the Absolute Q™ dedicated computer when an AutoRun is processing in the Momentum™ software. Doing so may interfere with the AutoRun system.

During an AutoRun Work Unit using the Spinnaker™, the Momentum™ software processes plates in the following order for hotels and stacks.

- **Hotels**—For each of the 4 hotels, plates are processed starting with Hotel 1 from Nest 1 on the bottom of the hotel, to Nest 15 on the top of the hotel proceeding sequentially to Hotel 4. Completed plates are placed into an open hotel nest.

The maximum number of plates per run for a hotel configuration is 60.

Note: This can be changed if additional batches are added to the Work Unit during the run. See “Add more plates during an AutoRun Work Unit with hotels” on page 148.

- **Stacks**—For each of the 2 input stacks, plates are processed starting with Stack 1 from top to bottom, then Stack 2 top to bottom. Completed plates are placed into Stack 3 and Stack 4.

The maximum number of plates per run for a stack configuration is 80.

Before performing an AutoRun Work Unit using the Momentum™ software, the DPT and DPR runs must be planned using the Absolute Q™ software that is installed on the Momentum™ dedicated computer or on a separate computer.

- For information about creating run templates, see “Manage templates” on page 40.
- For information about creating multiple DPR files for use in the Momentum™ batch, see “Generate batch runs from a template” on page 51.

IMPORTANT! Before starting an AutoRun, ensure that the plate nest accessory has been properly installed into the Absolute Q™ instrument plate drawer. Contact your Thermo Fisher representative for assistance.

1. Place each MAP plate into a plate nest in the hotels or stacks using the orientation shown in Figure 32.

Note: Figure 32 represents a plate loaded in a hotel plate nest. The same orientation is used for stack plate nests.

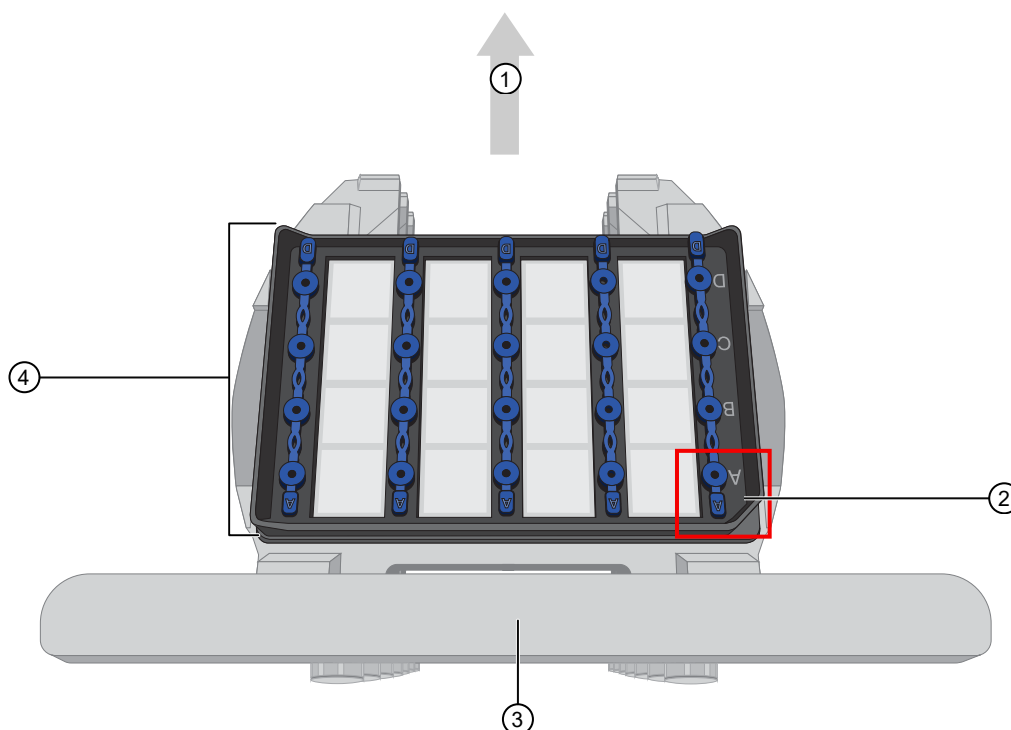


Figure 32 MAP plate in a hotel

- ① Orientation to the Spinnaker™
- ② Position of well A-1
- ③ The hotel or stack handle
- ④ Side of the MAP plate with barcode

2. If the run files were created on a different computer using the Absolute Q™ software, copy them into the correct folder for the type of run you are performing.
 - For the DPT Runs workflow, copy the DPT files to D:\Users\Public\Documents\Thermo Scientific\Automation Folder\DPT Templates
 - For the DPR Runs workflow, copy the DPR files to D:\Users\Public\Documents\Thermo Scientific\Automation Folder\DPR Barcoded Templates.

Note: Runs that are created on this computer are exported to these locations as part of the run creation process.

3. If security is enabled, sign in with a user name and password.
 For information about security, see “Momentum™ Security” on page 150.
4. In the Momentum™ software, click **Dashboard**
5. (Optional) In the **System, Options** area, click **Attended**.

IMPORTANT! Only click **Attended** if you are monitoring the run and want to respond to any error messages that may appear.

If **Attended** is not selected, the software manages all errors except fatal errors that require operator intervention.

6. To bring the instruments online, in the **System** area, click **Start/Stop**.
 For a single Absolute Q™ configuration, one server window opens. For a dual Absolute Q™ configuration, two server windows open.
7. Minimize the server window or windows.

IMPORTANT! Be sure to only minimize the server window, do not close it.

8. (Optional) If using only one Absolute Q™ instrument during a run on a dual configuration system, perform the following actions.
 - a. Click **Device Tasks**.
 A list of the devices that are online appears.
 - b. To take an Absolute Q™ instrument offline, in **Device Tasks**, click the power button icon next to the instrument name, then click **Done**.
9. In the **Work, Queued** area, click  to add a work unit.
10. On the **Add/Remove Work** screen, click **Experiments**.
 Three experiment options are available.
 - **Start DPR Run**
 - **Start DPT Run**
 - **Test Thermo Service Pick and Place**—This option is only to be performed by a qualified Thermo Fisher field service engineer.

11. Based on the workflow you are running, click **Start DPR Run** or **Start DPT Run**.



12. In the **Experiment Details**, **Iterations** field, enter the number of plates you are running, then click  **Add**.
 13. In the **Load Work Unit <#>** dialog, ensure that the plates are loaded correctly, then click **Verify**.
 14. On the **Add/Remove Work**, **Work Queue** area, click **Done**.
The work unit appears in the **Status, Pending** area.
 15. (Optional) To rename the batch in the work unit, perform the following actions.
 - a. In the right-pane, click **Work Explorer**.
 - b. In the batch within the work unit, click .
 - c. In the **Name** field, enter the new name for the batch, then click **OK**.
-
- Note:** If the **Work Explorer** is not visible, click  in the upper-left of the screen, then click **Reset View**.
-
16. (Optional) For hotel configurations only, for multiple batch runs, you can set the priority for each batch using the **Resource Conflict Priority** field.
 - a. In the right-pane, click **Work Explorer**.
 - b. In the batch for the work unit, click .
 - c. In the **Resource Conflict Priority** field, enter a value from 0–10 to set the priority of these plates in the run, then click **OK**.
Priority level ranges from 0–10, 0 is the highest priority and 10 is the lowest priority. 10 is the default setting.
-
- Note:** Batches set with the same priority level are processed sequentially.
-
17. To start the run, in the **Running** area, click **Run**.
The work unit moves to the **Status, Progress** status box.
 18. For DPT Runs, when prompted for the template file, navigate
to `D:\Users\Public\Documents\Thermo Scientific\Automation Folder\DPT Templates`, select the DPT file for this run, then click **OK**.
When the run is complete, the work unit moves to the **Status, Waiting for unload** box.
 19. In the **Work, Done** box, click **Unload**.
The **Unload Work Unit <#>** dialog lists the plates that are expected to be removed.
 20. Physically remove the plates shown on the list from the hotel or stack, then click **Verify**.



Add more plates during an AutoRun Work Unit with hotels

In configurations using hotels you can add more plates during a run by adding a new batch to the work unit. You can also set a priority level for the batch if it must be run before other batches in the current run.

IMPORTANT! For optimal system performance and stability, it is recommended to power-cycle the Absolute Q™ Digital PCR instrument(s) before starting each Work Unit. Multiple batches can be added to a single Work Unit when using a hotel configuration, but it is recommended not to exceed 80 plates on a single configuration or 160 plates on a dual configuration in a single Work Unit.

IMPORTANT! Do not attempt to load more than 60 plates onto the system at one time. Doing so can lead to aborted runs.

Although you can only load 60 plates at a time with a hotel configuration, by unloading completed plates from the batch in progress and adding a batch to a work unit, you can increase the number of plates for this run.

1. (Optional) If there are 60 plates currently loaded in the system, unload completed plates before adding plates for the new batch.
 - a. In the **Work, Done** area, click **Unload**.
 - b. When prompted, unload the plates that have been completed to create room for the plates in the new batch.
2. In the right-pane, click **Work Explorer**.

Note: If the **Work Explorer** is not visible, click  in the upper-left of the screen, then click **Reset View**.

3. Click  located above the work unit name to add a batch.
4. In the **Experiment to run** dropdown in the **Add to Batch to Work Unit** dialog, select the type of experiment to run with the batch.
 - **Start DPR Run**
 - **Start DPT Run**
5. (Optional) In the **Batch Name** field, change the name of the batch.
6. In the **Number of Iterations** field, enter the number of plates to add to the work unit.
7. (Optional) In the **Resource Conflict Priority** field, enter a value from 0–10 to set the priority of these plates in the run.

Priority level ranges from 0–10, 0 is the highest priority and 10 is the lowest priority. 10 is the default setting.



8. In the **Start Constraint** list select an option for starting this batch.

- **As soon as possible**
- **At a specific time**
- **Delay start by**
- **Delay after start of another experiment**
- **Delay after completion of another experiment**

Note: **As soon as possible** is the most commonly used option. When selected, this batch starts when the current batch completes if both batches have the same **Resource Conflict Priority**. If these plates must be run prior to the completion of the current batch, set the **Resource Conflict Priority** to a lower value than the current batch or batches.

9. Click **OK** to add the batch to the work unit.

The batch is added to the queue and is run based on the **Resource Conflict Priority**.

10. In the **Load Work Unit <#>** dialog, reference the nests where the plates will be loaded, place the new plates into the corresponding nests, then click **Verify**.

11. For DPT runs, when prompted for the template file, navigate to `D:\Users\Public\Documents\Thermo Scientific\Automation Folder\DPT Templates`, select the DPT file for this run, then click **OK**.

Files generated during an AutoRun Work Unit

The following table provides information about the files generated during an AutoRun Work Unit and where they are located on the Momentum™ computer.

File	Description	Export location	Source
ZST	Absolute Q™ completed run data file.	D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Data Output	Result of an Absolute Q™ run.
FSA ZIP	Field Service Archive files used for troubleshooting. See “Field Service Archive files” on page 121.	D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Data Output	Result of an Absolute Q™ run.
CSV ZIP	Analyzed results files.	D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Data Output	Result of an Absolute Q™ run.
Momentum™ Run Status Report	Summary of status and locations of plates run during a Work Unit.	D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Momentum Run Status Reports	Result of a completed work unit in Momentum™.



Power off the AutoRun Suite

1. To take the devices offline, in the Momentum™ **Dashboard, System** area, click **Start/Stop**.
The icon changes from bright green to gray to indicate that the devices are offline.
2. On the Momentum™ dedicated computer, close the Momentum™ software and Absolute Q™ software, if open.
3. On the Absolute Q™ dedicated computer or computers, close the Absolute Q™ software.
4. To power off the Spinnaker™ instrument, move the **DC POWER** switch on the front of the power puck to the off position.
5. To power off the Absolute Q™ instrument or instruments, move the power switch on the left side near the back of the instrument to the O position.
6. Power off all the dedicated computers.

Momentum™ Security

When the Momentum™ security feature is enabled, each user has a user account, and must sign in using this account to use the system.

There is no limit to the number of user accounts that can be created.

The sections that follow provide information about the different account types, managing users, enabling security, and signing in and out of the software.

Momentum™ User Accounts

There are three types of conventional user accounts.

- Operator. See “Momentum™ operator accounts” on page 151
- Expert. See “Momentum™ expert accounts” on page 151
- Administrator. See “Momentum™ administrator accounts” on page 151

Additionally, there is a super-user “Admin” account that is used by system administrators and Thermo Fisher Scientific personnel when installing, maintaining, and troubleshooting the system.

For information about managing user accounts, see “Manage user accounts” on page 153.



Momentum™ operator accounts

The operator account has limited access to the features in Momentum™. Operators can perform actions related to initiating and managing runs, but cannot edit the settings or profile elements for runs.

Operators can perform the following actions.

- View profiles, processes, and experiments
- Bring the system online and offline
- Create work units and add batches
- Monitor runs
- Verify that plates have been loaded and unloaded
- View and address warning and error messages
- Bring instruments online and offline
- Initiate tasks

Momentum™ expert accounts

Experts have access to more features than operators, but cannot adjust security settings, or create, edit, or delete security accounts.

Experts can perform the following actions.

- Perform all actions permitted by an operator account. See “Momentum™ operator accounts” on page 151.
- Create and edit profiles, processes, and experiments.

Momentum™ administrator accounts

Administrator accounts have access to all the features that an expert account permits, as well as managing security settings, and user accounts.

Administrators can perform the following actions.

- Perform all actions permitted by operator and expert accounts. See “Momentum™ operator accounts” on page 151 and “Momentum™ expert accounts” on page 151.
- Enable and disable Momentum™ security. See “Enable or disable security” on page 153.
- Create, edit, and delete user accounts. See “Manage user accounts” on page 153.
- Set password properties. See “Manage password settings” on page 152.
- Activate and disable user accounts. See “Activate or disable a user account” on page 155.

Manage password settings

Only administrators and users logged in as the Administrator super-user can change password parameters.

1. From the **Setup** ribbon, **Security** section, click **Administer**.
2. In the **Preferences** menu, select **Restrictions**.
3. In the **User Restrictions** dialog, modify the password parameters according to the policies and procedures of your organization, then click **OK**.

Parameter	Description
Password Retries	The maximum number of login attempts that can be made before an account is locked. Default retries are 10. Retries can be set from 3–50. To unlock an account, it must be reactivated. See “Activate or disable a user account” on page 155.
Lock Timeout (minutes)	The amount of time (in minutes) the software waits for an action from the user before locking the current session and requiring the user to sign in again. The default is 15, but can range from 1 to 1,440 (twenty-four hours). Enter 0 to disable the timeout.
Password Expiration (days)	The amount of time (in days) before a password expires and must be changed. The default is 90 days, however, 30 to 366 days are allowed. Enter 0 to disable password expiration.
Minimum Number of Characters	Default: 8 Range allowed: 6–50
Minimum Number of Lowercase Alpha Characters	Default: 1 Range allowed: 1–50
Minimum Number of Uppercase Alpha Characters	Default: 1 Range allowed: 1–50
Minimum Number of Digits	Default: 1 Range allowed: 1–50
Minimum Number of Special Characters	Default: 1 Range allowed: 0–50



Enable or disable security

IMPORTANT! When security is initially enabled, ensure to record the current password for the administrator account. Failure to do so prevents users from opening Momentum™ until other security accounts have been created.

1. From the **Setup** ribbon, **Security** section, click **Administer**.
The **Security** editor opens in the main window and the main toolbar displays the **Administration** options.
The **Administration, Preferences** menu shows **Enabled** if security is enabled or **Disabled** if security is disabled.
2. To change the security setting from its current setting, in the **Preferences** menu, click **Enabled** or **Disabled**.
3. In the **Security Selection** dialog, select the setting you want to use, then click **OK**.
 - **Enable**—Requires users to sign on to the system with administrator assigned credentials.
 - **Disable**—Users are not required to sign on the system.
4. When enabling security, when prompted, sign in with a user name and password, then click **Login**.

Manage user accounts

There is no limit to the number of user accounts that an administrator may create.

Create user accounts

Only administrators and users logged in as the Administrator super-user can create user accounts. Each user account must have a valid user name and password.

1. From the **Setup** ribbon, **Security** section, click **Administer**.
2. In the **Preferences** menu, select **Filter**, then ensure it is set to **Active**.
3. In the **Users** menu, select **New**.
4. In the **New User** screen enter the following user parameters.

Note: Required fields are noted with an .

Parameter	Action
User Role	From the dropdown list, select the user role. • Administrator • Expert • Operator
User Name	Enter the user name of the user.



(continued)

Parameter	Action
Password	Enter a password that complies with the policies and procedures of your organization.
Confirm Password	Re-enter the password.
Choose Picture	1. Select Choose Picture. 2. In the file browser, navigate to the location of the image. 3. Select the image, then click OK.
User must change password at next login	Select this option to force the user to change their password the next time they log in.
Full Name	Enter the full name of the user.
E-mail	Enter the email address of the user.
Phone	Enter the phone number of the user.

5. Click **OK** to save the user information and activate the user account.

Edit user accounts

Only administrators and users logged in as the Administrator super-user can edit user accounts.

Only active accounts can be modified. See “Activate or disable a user account” on page 155.

1. From the **Setup** ribbon, **Security** section, click **Administer**.
2. In the **Preferences** menu, select **Filter** and ensure it is set to **Active**.
3. In the **Users** menu, select the user account, then select **Edit**.
4. Modify the user account settings as needed. See “Create user accounts” on page 153.
5. When the changes are complete, click **OK**.

Change user password

All user account types can change their password as long as it complies with the policies and procedures of your organization.

1. In the Quick Access toolbar in the upper-left corner of the screen, select the **User Tasks** button, and then select **Change Password**.
2. In the **Old Password** field, enter the current password for the account.
3. In the **New Password** field, enter a password that complies with the policies and procedures of your organization.

Note: The new password must be different from the last six passwords used for this user account.



4. In the **Confirm Password** field, re-enter the new password. It must match the new password to save the change.
5. Select **Change** to save the new password.

Activate or disable a user account

Only administrators and users logged in as the Administrator super-user can activate or disable a user account.

1. From the **Setup** ribbon, **Security** section, click **Administer**.
2. In the **Users** menu, select the user account, then select **Edit**.
The user list is displayed.
3. Select a user, then choose one of the following options.

Option	Action
Disable	Select to disable, but not delete the account. The account is removed from the active account list and placed on the disabled list.
Activate	Select to reactivate a disabled account or an account that has been locked due to failed password entry. The account is moved from the disabled account list to the active account list.

4. When the changes are complete, click **OK**.

Delete user accounts

Only administrators and users logged in as the Administrator super-user can delete user accounts.

Take the following into consideration when deleting a user account.

- Deleting a user account sets the account status to deleted and removes it from the list of useable accounts in the AutoRun system.
- Deleted accounts cannot be reactivated.
- User names from deleted accounts cannot be reused when creating a new user account.
- Only active accounts can be deleted.

1. From the **Setup** ribbon, **Security** section, click **Administer**.
2. In the **Preferences** menu, select **Filter** and ensure it is set to **Active**.
3. In the **Users** menu, select the user account you want to delete.
4. When prompted, select **Delete**.

Sign in and out of Momentum™ software

If security is enabled, all users must sign in using a valid user security account to access the Momentum™ software.

Take the following actions into consideration when security is enabled.

- When the Momentum™ software is closed, the current user is automatically signed out.
- A user can sign out without closing the Momentum™ software to allow another user to sign in.
- The current session can be locked so that the Momentum™ software cannot be used until the current user signs out or another account is used to sign in, ending the current session.

Sign into Momentum™

1. Start Momentum™ by clicking the desktop icon.

If security is enabled, the Login dialog box opens.

Note: The status bar in the lower-right corner of the page displays the name of the user who is currently logged in.

2. Enter a valid user name and password, then click **Login**.

Sign out, lock, or switch users

1. In the **Quick Access** toolbar in the upper-left corner of the screen, select the **User Tasks** button, and then select **Lock/Switch User**.
2. In the **Login** dialog, perform one of the following options.

Option	Action
Sign in under a different user security account.	Enter a user name and password, then click Login . The previous user session ends and the new user is logged in.
Lock the session of the current user and prevent others from using it.	Leave the Login dialog open and make no changes. The current session cannot be used by other users until one of the following actions is taken in the Login dialog. • The current user enters their user name and password. • A different user enters their user name and password which ends the current session and starts a new one with the new user.



Use the software with Security, Auditing, and E-signature (SAE) v2.2 or later

■ Overview of the SAE Administrator Console components	157
■ Enable SAE functions	159
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The Security, Auditing, and E-signature (SAE) v2.2 or later software (SAE Administrator Console) is only compatible with the QuantStudio™ Absolute Q™ Digital PCR System. For more information, see the *QuantStudio™ Absolute Q™ Systems Software/Firmware Compatibility and User Documents Reference* (Pub. No. MAN0025886).

For more information on Security, Auditing, and E-signature (SAE) v2.2 or later software, including definitions of accounts and roles, see the user guide for your version of the software (“Related documentation” on page 208).

Overview of the SAE Administrator Console components

The SAE Administrator Console includes three components.

- SAE Administrator Console that an administrator uses to configure the module.
- SAE server that stores settings, user accounts, and audit records.

Note: The SAE server and SAE Administrator Console software are installed simultaneously on the same computer during installation.

- SAE screens in an application (sign in and audit that a user interacts with). QuantStudio™ Absolute Q™ Digital PCR Software is an application.

The SAE Administrator Console provides the following SAE functionality in the QuantStudio™ Absolute Q™ Digital PCR Software.

- **System security**—Controls user sign in and access to functions.
- **Auditing**—Tracks changes and actions performed by users.
- **E-signature**—Allows users to provide an electronic signature (user name and password) when performing certain functions.

Take the following into consideration based on the way that your SAE administrator configures the system.

- Some features and functions that are described in this guide may not be accessible to you.
- You may see dialog boxes and prompts when you use the software.

Overview of the QuantStudio™ Absolute Q™ Digital PCR Software functionality when SAE functions are enabled

The following features are active when SAE functions are enabled in the QuantStudio™ Absolute Q™ Digital PCR Software.

- Users must sign in with an SAE user account to use QuantStudio™ Absolute Q™ Digital PCR Software.
- Both audit objects and audit actions are tracked in the SAE Administrator Console. Audit actions are tracked automatically, audit objects are viewable when enabled.
- Run setup and software functions for a user are determined by the SAE application profile and user account settings.

Recommendations for SAE passwords

Thermo Fisher Scientific recommends enabling a password policy for SAE user accounts with the following minimum number of characters:

- Administrative users: 12 characters
- Non-administrative users: 8 characters

The use of a password manager is recommended in order to help to create secure passwords.

SAE functions not supported by the QuantStudio™ Absolute Q™ Digital PCR Software

The following SAE functions are not supported by the QuantStudio™ Absolute Q™ Digital PCR Software.

Function	Option not supported
System > Other Settings	• Open file from non-SAE system • Client offline sign in • Offline sign in threshold
Audit history	Instrument Run Records



(continued)

Function	Option not supported
e-Signature	• Ability to add e-Signature meanings • Ability to delete e-Signature meanings • Ability to configure actions that require e-Signature • Ability to control/configure e-Signature rights by user role • Ability to control reasons available for e-Signature • Ability to control/configure data to be signed for each e-Signature meaning Signed data in the e-Signature Records PDF report generated by the SAE Administrator Console does not contain any objects. To create a report with this information, print the result report from the QuantStudio™ Absolute Q™ Digital PCR Software. • Ability to control/configure number of signatures (by role) for each action requiring e-Signature

Enable SAE functions

Workflow

Enable SAE functions

Install the SAE Administrator Console and Absolute Q™ application profile (page 160)

Connect to the SAE server (page 161)

Enable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software (page 162)



Install the SAE Administrator Console and Absolute Q™ application profile

The following configurations of SAE server and SAE Administrator Console software are supported.

- SAE installed on a stand-alone computer that is connected to the Absolute Q™-dedicated computer and optional Absolute Q™ analysis-dedicated computers

Computer	Function	Software	Provider
SAE stand-alone computer	SAE server	SAE Administrator Console	Customer or Thermo Fisher Scientific
Absolute Q™ dedicated computer	Computer connected to the QuantStudio™ Absolute Q™ Digital PCR Instrument	QuantStudio™ Absolute Q™ Digital PCR Software	Thermo Fisher Scientific
(Optional) Analysis computer(s)	Analyzing digital PCR data	QuantStudio™ Absolute Q™ Digital PCR Software	Customer or Thermo Fisher Scientific

- SAE and QuantStudio™ Absolute Q™ Digital PCR Software that is installed on the Absolute Q™-dedicated computer and is connected to optional Absolute Q™ analysis-dedicated computers

Computer	Function	Software	Provider
Absolute Q™ dedicated computer	SAE server and computer connected to the QuantStudio™ Absolute Q™ Digital PCR Instrument	SAE Administrator Console and QuantStudio™ Absolute Q™ Digital PCR Software	Thermo Fisher Scientific
(Optional) Analysis computer(s)	Analyzing digital PCR data	QuantStudio™ Absolute Q™ Digital PCR Software	Customer or Thermo Fisher Scientific

IMPORTANT! Before installing the application profile, see the release notes for compatibility information to ensure you are installing the Absolute Q™ application profile that is compatible with the version of QuantStudio™ Absolute Q™ Digital PCR Software that you are using.

- To download the SAE Administrator Console software and Absolute Q™ application profile go to the software download page specific to your region:

Region	Go to:
Global (except China)	thermofisher.com/us/en/home/global/forms/life-science/quantstudio-absolute-q-software.html
China	thermofisher.cn/cn/zh/home/technical-resources/software-downloads/quantstudio-absolute-q-digital-pcr-system.html

- Click **Get software**.
- Sign in using your thermofisher.com username and password.

Note: If you do not have a thermofisher.com account, click **Create account** in the sign in screen, then follow the instructions.

- Download the software package.



2. Install the SAE server and SAE Administrator Console software on a computer with a static IP address (*recommended*) or a dynamic IP address.
 - a. Unzip the downloaded software.
 - b. Double-click **setup.exe**
 - c. Follow the **InstallShield Wizard** prompts to install the software.
 - d. Select **Typical** as the setup preference, then click **Next**.
 - e. Click **Finish**.

Note: The SAE server and SAE Administrator Console software are installed simultaneously during installation.

3. In the SAE Administrator Console, an SAE administrator must install the application profile for the QuantStudio™ Absolute Q™ Digital PCR Software before SAE can be used.

The application profile contains default settings for the QuantStudio™ Absolute Q™ Digital PCR Software.

For information about installing application profiles, see the user guide for your version of the software (“Related documentation” on page 208).

Connect to the SAE server

1. In the QuantStudio™ Absolute Q™ Digital PCR Software, select  **System ▶ SAE Connection Settings**.
2. Enter the IP address and port number of the SAE Administrator Console.

If the SAE Administrator Console is installed on the same computer as the QuantStudio™ Absolute Q™ Digital PCR Software, enter *localhost*.

If the SAE Administrator Console is installed on a different computer from the QuantStudio™ Absolute Q™ Digital PCR Software, enter the IP address of the computer on which the SAE Administrator Console is installed.

Note: If using a dynamic IP address, enter the hostname instead of the IP address to prevent the loss of a connection (see “Determine the hostname” on page 162).

The port number is the firewall port. See “Firewall ports that must be open” on page 162.

3. Click **Test Connection** to confirm that the connection information is correct.
4. Click **Save**.



Determine the hostname

If the SAE Administrator Console is on a separate computer from the application and a dynamic IP address is used, the hostname is recommended instead of the IP address. This helps to prevent the loss of a connection between the SAE Administrator Console and the application

1. In the Windows™ search bar, enter **cmd** to open the **Command Prompt**.
2. Enter **hostname**, then press **Enter**.

The hostname of the computer is displayed in the **Command Prompt**.

Firewall ports that must be open

The following ports must be open for the operating system on the computer that is running the SAE Administrator Console.

SAE Administrator Console version	Port	Condition
v2.0	8201	• Instrument-to-SAE Administrator Console server connection • Computer-to-SAE Administrator Console server connection^[1]
v2.1 and later	8443	• Instrument-to-SAE Administrator Console server connection • Computer-to-SAE Administrator Console server connection^[1]

^[1] If the software is installed on a different computer than the SAE Administrator Console.

Open firewall ports

- To open a port for Microsoft Defender™, add inbound rules for the port, and apply to all profiles.
- To open a port for Norton Internet Security™, use the **Settings** menu to open the port.

Enable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software

This procedure requires an SAE administrator account.

Before you enable SAE functions in the QuantStudio™ Absolute Q™ Digital PCR Software, you must complete the following tasks:

- Connect to the SAE server (see “Connect to the SAE server” on page 161).
 - Close all protocol or analyzed run files.
1. In the QuantStudio™ Absolute Q™ Digital PCR Software, navigate to **⚙ System Settings**.
 2. Toggle the **Enable security** on (see “Configure system settings” on page 131).
 3. Enter your SAE administrator account user name and password, then click **Sign In**.

The SAE administrator account is automatically signed into the software after SAE functions are enabled. The SAE user name is displayed in the upper-right corner of the software menu bar. All users must sign into the software while SAE functions are enabled.



To sign out of the SAE administrator account in the Absolute Q™ software, see “Sign out of the software using an SAE account” on page 163.

Note: Signing out of the SAE administrator account does not disable SAE functions in the Absolute Q™ software. To disable SAE functions in the Absolute Q™ software, see “Disable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software” on page 175.

Sign into QuantStudio™ Absolute Q™ Digital PCR Software using an SAE account

Sign in for the QuantStudio™ Absolute Q™ Digital PCR Software is only required if SAE functions are enabled by an SAE administrator (see “Enable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software” on page 162).

1. In the QuantStudio™ Absolute Q™ Digital PCR Software sign in screen, enter your SAE user name and password.
2. Click **Sign In**.

The user name of the SAE account that is signed in to the software appears in the menu bar.

Sign out of the software using an SAE account

1. In the lower-left corner of the left pane, click .
2. Click **Sign Out**.

Change your SAE account password

Note: External user account (External/Federated LDAP repository accounts) passwords cannot be changed in the QuantStudio™ Absolute Q™ Digital PCR Software, they can only be changed in their respective repository.

1. In the lower-left corner of the left pane, click .
2. Click **Change Password**.
3. Enter the password information, then click **OK**.



Default permissions and roles

The SAE Administrator Console provides the following default permissions and roles. You can use the default roles when you create SAE user accounts or create custom roles in the Security, Auditing, and E-signature (SAE) Administrator Console v2.2 or later (see the user guide for your version of the software (“Related documentation” on page 208)).

- Administrator
- Technician
- Scientist
- Service

IMPORTANT! Permissions for a role apply to all user accounts that are assigned to the role.

The roles and associated user-configurable permissions are listed in the following table. You can also double-click the role in the **Roles** tab to display the list of permissions.

Note: The **No Privileges** role is used by the software when you set up user repositories. Do not assign this role to a user account.

Function	Description	Role			
		Administrator	Scientist	Technician	Service
Miscellaneous					
Service access	Access to the instrument service menu.	Yes	No	No	Yes
System settings	Access to the system menu.	Yes	No	No	Yes
Generate report	Create analysis reports.	Yes	Yes	Yes	Yes
E-SIGN run	Place an electronic signature on a run.	Yes	Yes	No	No
E-SIGN study	Place an electronic signature on a study.	Yes	Yes	No	No
Edit notes	Edit notes on plate setup.	Yes	Yes	Yes	Yes
Accept or reject calibration results	Accept or reject the results provided with an instrument calibration.	Yes	No	No	Yes
Presets Management					
Create template	Create a template for a run.	Yes	Yes	Yes	Yes
Import template	Import a template from another system.	Yes	Yes	Yes	Yes
Export template	Export a template to another system.	Yes	Yes	Yes	Yes
Rename template	Rename an existing template.	Yes	Yes	Yes	Yes
Delete template	Delete a template from the system.	Yes	Yes	No	Yes
Save as template	Save a run as a template.	Yes	Yes	Yes	Yes



(continued)

Function	Description	Role			
		Administrator	Scientist	Technician	Service
Create a batch run	Create multiple runs from the same template.	Yes	Yes	Yes	Yes
Edit protocol—templates	Change protocol settings in an existing template.	Yes	Yes	Yes	Yes
Assign samples and groups—templates	Assign samples and groups to wells on the plate in a template.	Yes	Yes	Yes	Yes
Edit groups and dye settings—templates	Modify groups and dye settings in an existing template.	Yes	Yes	Yes	Yes
Edit plate samples and rename samples—templates	Modify samples in an existing template.	Yes	Yes	Yes	Yes
Instrument Control					
Start run	Choose a protocol and start and stop instrument runs.	Yes	Yes	Yes	Yes
Stop run	Stop a run in progress.	Yes	Yes	No	Yes
Software or firmware update for instrument	Update the instrument software and firmware.	Yes	No	No	Yes
Pre-Run					
Edit protocol	Change protocol settings on a draft run.	Yes	Yes	Yes	Yes
Assign samples and groups	Assign samples to set groups or load a group set in a run.	Yes	Yes	Yes	Yes
Edit groups and dye settings	Modify groups and dye settings on a draft run.	Yes	Yes	Yes	Yes
Edit plate samples and rename samples	Modify samples on a draft run.	Yes	Yes	Yes	Yes
Run analysis					
Change thresholds	Change channel thresholds in a run.	Yes	Yes	No	Yes
Edit groups and dye settings	Edit group definitions including dye settings in a run.	Yes	Yes	No	Yes
Edit and rename samples	Change sample names in a run.	Yes	Yes	Yes	Yes
Assign samples and groups	Assign samples to set groups or load a group set in a run.	Yes	Yes	Yes	Yes



(continued)

Function	Description	Role			
		Administrator	Scientist	Technician	Service
Omit or include samples	Include or omit samples from an analysis in a run.	Yes	Yes	No	Yes
Run management					
Delete run	Delete a run from the database.	Yes	No	No	Yes
Import run	Import runs to and from ZST or ZIP files.	Yes	Yes	Yes	Yes
Export run	Export runs to and from ZST files.	Yes	Yes	Yes	Yes
Rename run	Change the name of the run.	Yes	Yes	Yes	Yes
Study Analysis					
Change thresholds	Change sample and group thresholds during study analysis.	Yes	Yes	No	Yes
Edit groups	Edit groups contained in a study.	Yes	Yes	No	Yes
Edit plate samples and rename samples	Edit and rename samples in a study.	Yes	Yes	Yes	Yes
Omit or include samples	Include or omit samples from a study.	Yes	Yes	No	Yes
Study Management					
Import study	Import studies from other systems.	Yes	Yes	Yes	Yes
Export study	Export studies to ZIP files.	Yes	Yes	Yes	Yes
Rename study	Change the name of a study.	Yes	Yes	No	Yes
Delete study	Delete a study.	Yes	Yes	No	Yes
Create a study and add runs to a study	Create and add runs to studies.	Yes	Yes	No	Yes

Use audit functions

The following sections provide information on using SAE auditing functions.



Specify audit reason

Depending on how the audit settings are configured in the SAE Administrator Console, the **Enter Audit Reason** screen may appear when you make changes to a protocol or an analyzed run in the QuantStudio™ Absolute Q™ Digital PCR Software to prompt you to select an audit reason from the drop down list, or add a custom reason.

Note: **Custom Reason** is not displayed if audit settings are configured to require users to select a reason.

For more information on configuring audit settings, see the user guide for your version of the software (“Related documentation” on page 208).

View audit records

For instructions to view audit action records for a protocol or an analyzed run, see the user guide for your version of the software (“Related documentation” on page 208).

For a list of actions that are audited, see “Actions that are audited” on page 168.

For instructions to view audit object records of a specific run, see “View audit object records” on page 167.

View audit object records

Use the following steps to view the audit object record of a specific run by using the Run ID for the run.

1. In the QuantStudio™ Absolute Q™ Digital PCR Software, select the desired run.
2. In the upper-left corner of the run page, click  next to the **Run ID** to copy the **Run ID** to the clipboard.
3. At the SAE Administrator Console perform the following steps.
 - a. Select **Audit History > Application Object Records**.
 - b. Select **Enable Application Objects Filtering**.
 - c. In the **Object name** field, paste the **Run ID** that you copied in step 2.
 - d. Click **Search**.

The information regarding the run appears in results area of the **Audit History** screen.

Note: For assistance in interpreting audit history data, contact Technical Support.



Actions that are audited

The actions are audited and listed in the action records regardless of whether audits are enabled or disabled.

The following user actions are audited.

Function	Actions audited
Miscellaneous	• EULA accept or decline • Sign in • Sign out • Save system settings • Update instrument software/firmware version • Open and/or close instrument door (exact action with user name)
Templates	• Create, edit, or save a template • Save as template (when creating/editing a template) • Import or export protocol (when creating/editing a template) • Create a sample group (when creating/editing a template) • Edit dyes (when creating/editing a template) • Change optical settings (when creating/editing a template) • Add, edit, or delete notes in setup • Import template or templates • Export template or templates • Rename template • Delete template or templates • Generate batch runs
Runs	• Create, edit, and save a run • Save a run as a template (when creating/editing a run and viewing a completed run) • Import protocol (when creating/editing a run) • Export protocol (when creating/editing a run) • Update sample group assignment • Edit dyes (when creating/editing a run) • Add, edit, or delete notes in setup • Import run or runs • Export run or runs • Rename run • Delete run or runs • Start or stop run on the instrument • Start or stop a calibration run • Add a run or runs to a study

(continued)

Function	Actions audited
Runs—changes during analysis	• Update sample group assignment • Create or delete a sample group • Export protocol • Edit dyes • Add, edit, or delete notes in setup • Change threshold (both group threshold and threshold for a dye channel) • Omit sample • Pin or unpin threshold • Generate a report for a run • Download data for a run • Accept or reject calibration results • Generate a report for calibration run
Studies	• Create a study • Import study or studies • Export study or studies • Rename study • Delete study or studies • Add a run or runs to a study
Studies—changes during analysis	• Update sample group assignment • Create or delete a sample group • Export Protocol • Edit Dyes • Add, edit, or delete Notes in Setup • Change threshold (both group threshold and threshold for a dye channel) • Omit sample • Pin or unpin threshold • Generate a report for a run • Download data for a run • Accept or reject calibration results • Generate a report for calibration run

Export audit records

For information on exporting audit records for a protocol or an analyzed run, see the user guide for your version of the software (“Related documentation” on page 208).



Sign data in the software

An e-signature can optionally be added for plate setup and run results on the **Runs** and **Studies** pages.

1. Select from the following options to provide an e-signature for plate setup and run results.

Option	Actions
Runs screen, DRAFT tab—Signing for plate protocol and setup.	1. In the left navigation pane, click Runs to open the Runs screen. 2. Use the search field to find a run or select a run from the list.
Runs screen, COMPLETED tab—Signing for protocol, setup, and results of the run.	1. In the left navigation pane, click Runs to open the Runs screen. 2. Navigate to the COMPLETED tab. 3. Use the search field to find a run or select a run from the list.
Studies screen—Signing for protocol, setup, and results of the study.	1. In the left navigation pane, click Studies to open the Studies screen. 2. Use the search field to find a study or select a study from the list.

2. Click **E-SIGN**, then select one of the following options from the dropdown list to indicate the meaning of the e-signature.
 - Reviewed & approved setup
 - Reviewed & approved results
3. Enter your user name and password.
4. Click **E-SIGN**.

If a run is signed and unmodified, the signature appears on reports that are created using the **GENERATE REPORT** feature (see “View results” on page 104).

For information on how to view e-signature data in the SAE software, see *View and report audit and e-Signature records* in the the user guide for your version of the software (“Related documentation” on page 208).

View and review e-Signatures

For information on how to view e-Signature data, see *View and report audit and e-Signature records* in the the user guide for your version of the software (“Related documentation” on page 208).

The sections that follow provide detailed information for reviewing e-Signature data.

- For information on plate setup e-Signature data, see “Review plate setup e-Signature information” on page 171.
- For information on plate results e-Signature data, see “Review plate results e-Signature information” on page 173.



Review plate setup e-Signature information

The sections that follow provide descriptions of the information provided in the e-Signature plate setup record for draft runs and run templates. Optionally, this information can be printed.

Signature metadata

This section provides information regarding the signature metadata for each e-Signature plate setup record.

Table 15 Signature metadata

Object	Description
Meaning	The e-Signature option selected.
Signed Date	The date of e-Signature.
Signed By	The name of user.
Host ID	The instrument name.
Full Name	The user name.
Status	The status of the signature: - CURRENT: Valid - OBSOLETE: Invalid
Role	The role assigned to the user who performed the run.

Protocol information

This section provides information regarding the **protocol** section of the e-Signature plate setup record.

Table 16 Protocol details

Object	Description
ScanRed	Status of True indicates this optical channel was enabled. Status of False indicates this optical channel has been disabled.
ScanGreen	Status of True indicates this optical channel was enabled. Status of False indicates this optical channel has been disabled.
ScanYellow	Status of True indicates this optical channel was enabled. Status of False indicates this optical channel has been disabled.
ScanDarkRed	Status of True indicates this optical channel was enabled. Status of False indicates this optical channel has been disabled.
RNAStep_Duration	The duration of RNA-RT step (<i>optional</i>).
RNAStep_Temperature	The temperature of RNA-RT step (<i>optional</i>).



Table 16 Protocol details (continued)

Object	Description
PCRPreheat_Duration	The duration of pre-heat step (optional).
PCRPreheat_Temperature	The temperature of pre-heat step (optional).
PCR_Stage(1/2)_Step(1/2/3)_Duration	The duration of indicated stage and step.
PCR_Stage(1/2)_Step(1/2/3)_Temperature	The temperature of indicated stage and step.
Name	The name of protocol.

Plate channel information for each sample

This section provides information regarding the channels used in the **plate** section of the e-Signature plate setup record. If the channel was not used, the detail will reflect **None** in all data points. The figure that follows depicts a partial record.

Table 17 For each color — blue, green, yellow, red, and dark red

Object	Description
Channel	The name of the target.
Type	The analysis type selected.
Threshold	The predefined analysis threshold.
Selection	The dye selected for the channel.

Additional plate information

This section provides information regarding the additional information provided in the **plate** section of the e-Signature plate setup record.

Table 18 Other plate information

Object	Description
DilutionFactor	Total dilution from sample to reaction mix.
CNVRefNum	The number of copies of the reference genome.
Name	The sample name.
Group	The group name.
GroupType	the group analysis setting



Run metadata

The section provides information regarding the **run name** section of the e-Signature plate setup record.

Table 19 Run metadata

Object	Description
run name	The name given to the run at the instrument.
Columns	Columns enabled for the run.
LastEditedEPOCH	Epoch time stamp of the run.
Barcode	Plate barcode number.

Review plate results e-Signature information

The sections that follow provide descriptions of the information provided in the e-Signature plate results record for completed runs and studies. Optionally, this information can be printed.

Signature metadata

This section provides information regarding the signature metadata for each e-Signature plate results record.

Table 20 Signature metadata

Object	Description
Meaning	E-Signature option selected.
Signed Date	Date of e-Signature.
Signed By	Name of user.
Host ID	Instrument name.
Status	Status of the signature: - CURRENT: Valid - OBSOLETE: Invalid

Results by group

This section provides information regarding the **groups** section of the e-Signature plate results record. A column is included for each dye used.

Table 21 For each group, for each dye

Object	Description
Total	One of the following options: - If replicates, this is the group average of microchambers. - If pooled, this is the total pooled microchambers.



Table 21 For each group, for each dye (continued)

Object	Description
Positive	Group positive microchambers.
Conc. (cp./uL)	Group concentration in copies per microliter.

Results for samples

This section provides information regarding the **samples** section of the e-Signature plate results record. A column is included for each dye used.

Table 22 For each sample, for each dye

Object	Description
Total	Sample total microchambers.
Positive	Sample positive microchambers.
PosThresh	Analysis threshold input by user.

Run metadata

The section provides information regarding the **run name** section of the e-Signature plates result record.

Table 23 Run metadata

Object	Description
run name	The name given to the run at the instrument.
Columns	Columns enabled for run.
LastEditedEPOCH	Epoch time stamp of the run.
Barcode	Plate barcode number.



Disable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software

This procedure requires an SAE administrator account.

IMPORTANT! Disable SAE functions in the QuantStudio™ Absolute Q™ Digital PCR Software before uninstalling the SAE Administrator Console.

Close all plate files and data files.

1. In QuantStudio™ Absolute Q™ Digital PCR Software, select  **System ▶ Disable Security**.
2. Enter the password of the SAE administrator account, then click **Sign In**.



Maintain the instrument

■ Clean the Absolute Q™ instrument and plate nest	176
■ Clean the Spinnaker™ Microplate Mover, hotels, and stacks	176
■ Maintenance	177

Clean the Absolute Q™ instrument and plate nest

All surfaces should be dry and free of dust and lint before operation.

Clean the outside of the instrument with a damp, lint-free cloth using one of the following solutions:

- Mild soap
- 70% ethanol in water

Clean the plate nest surface gently with a lint-free cloth (microfiber cloth or optical lens cleaning cloth) using 70% ethanol in water. Do not wipe the grooves that surround the plate nest.

IMPORTANT! The plate nest is covered in a thin graphite sheet. This sheet is susceptible to scratches and may impact results if it is damaged. It is important to only wipe the graphite surface with lint-free wipes or use air-dusters. Contact technical support if this surface becomes damaged (see Appendix K, “Documentation and support”).

Clean the Spinnaker™ Microplate Mover, hotels, and stacks



CAUTION! The Spinnaker™ must be de-energized before it can be cleaned. Failure to do so may result in injury.



MISE EN GARDE ! Le robot Spinnaker™ doit être mis hors tension avant de pouvoir être nettoyé. Le non-respect de cette consigne peut entraîner des blessures corporelles.

Use the steps that follow to help ensure that the Spinnaker™ robot is free of dust or debris.

1. De-energize the robot motors by pressing the **Motor Off** button.
2. Remove power to the robot by setting the **DC Power** switch to **OFF**.
3. Ensure that the power has been removed from the robot by disconnecting the power cord from the power outlet.

4. Lightly dab a sponge or soft, lint-free cloth in a solution of water mixed with a mild detergent. Ensure that the sponge or cloth is only damp (well-wrung and not dripping) before continuing.

Note: A mild isopropyl alcohol-based cleaning agent can also be used.

5. Gently wipe the surface of the robot, removing any dust or debris that may have accumulated.

IMPORTANT! Be sure to avoid direct contact with electrical components, including the power connection on the robot base.

6. If cleaning a robot connected to a base plate that supports random-access hotels and/or sequential-access stacks, remove the hotels and/or stacks and clean them separately using the same sponge or cloth and cleaning solution.
7. Before replacing the hotels and/or stacks, clean and dry the base plate.
8. After ensuring that the base plate is dry, replace any previously removed hotels and/or stacks.

When the robot is clean and dry, operation can resume.

Maintenance

For best results when using the instrument, the following practices are recommended:

- The plate nest must be inspected and cleaned before each run.
- For implementations using the AutoRun Suite, the plate nest and hotels or stacks must be cleaned before starting each work unit.
- For implementations using the AutoRun Suite, regularly backup, then delete the contents of D:\Users\Public\Documents\Thermo Scientific\Automation Folder\Data Output on the Momentum™ computer.
- Ensure that the fan vents on the back and bottom of the Absolute Q™ instrument are not obstructed.
- Ensure that system dyes are calibrated on a yearly basis.

Note: A warning message appears in the **Instrument** screen 45 days before dye calibration expiration. If the dyes are not calibrated within that time frame, a warning message appears indicating that the calibration has expired and remains in the **Instrument** screen until the dyes are calibrated.

IMPORTANT! System dye calibration must only be performed by qualified field service engineers. Attempting to calibrate dyes without the assistance of a field service engineer can compromise run data for analysis.

- For implementations using the AutoRun Suite, power cycle the Absolute Q™ Digital PCR instrument, the Spinnaker™ Microplate Mover, and the corresponding computers weekly.

IMPORTANT! For implementations using QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite, the system requires annual maintenance and qualification. For more information, contact your Thermo Fisher Scientific representative.

For information about maintenance and service plans, contact technical support (see Appendix K, “Documentation and support”).



Connect Absolute Q™ companion PCs to the network using the bridge PC

■ Network guidelines	179
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■ Network configuration	180
■ Connect one or more Absolute Q™ companion PCs to the network using the bridge PC	180

To satisfy site-specific IT requirements, one or more QuantStudio™ Absolute Q™ Digital PCR Systems can be connected to the customer network using the bridge PC. That is, the bridge PC is a PC that is used to establish a connection between one or more Absolute Q™ companion PCs and the customer network. This setup allows the IT department to provision the bridge PC with the necessary security settings and software without compromising the functionality of the QuantStudio™ Absolute Q™ Digital PCR System.

Network guidelines

- We do not recommend connecting the Absolute Q™ companion PC that is used for controlling the QuantStudio™ Absolute Q™ Digital PCR Instrument to the internet.
- We do not recommend installing extraneous software on the Absolute Q™ companion PC.

Hardware requirements

When connecting one Absolute Q™ System:	
• QuantStudio™ Absolute Q™ Digital PCR System • Bridge PC running Windows™ 10 or 11 • Cat5e / Cat6 / Cat6a Ethernet cable	Use an Ethernet cable to make the following connections: 1. Connect the Absolute Q™ companion PC to the bridge PC. 2. Connect the bridge PC to the network.



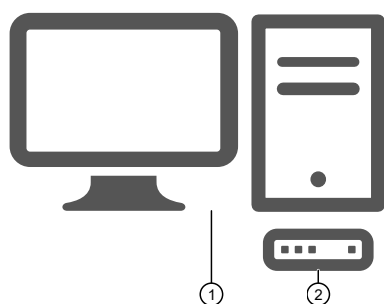
When connecting multiple Absolute Q™ Systems:

- QuantStudio™ Absolute Q™ Digital PCR Systems
- Bridge PC running Windows™ 10 or 11
- Gigabit Ethernet Switch
- Cat5e / Cat6 / Cat6a Ethernet cable

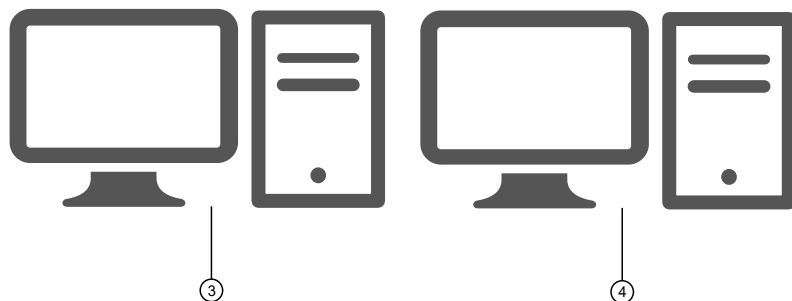
Use an Ethernet cable to make the following connections:

1. Connect each Absolute Q™ companion PC to the Gigabit Ethernet Switch.
2. Connect the Gigabit Ethernet Switch to the bridge PC.
3. Connect the bridge PC to the network.

Network configuration



- ① Bridge PC with:
\\BridgeShare-1
\\BridgeShare-2
- ② Router



- ③ Absolute Q™-1 with:
\\BridgeShare-1
- ④ Absolute Q™-2 with:
\\BridgeShare-2

Connect one or more Absolute Q™ companion PCs to the network using the bridge PC

On an Absolute Q™ companion PC:

1. Press **Windows + R** to open the **Run** dialog box.
2. In the **Run** dialog box, in the **Open** field, enter `ncpa.cpl`, then click **OK**.
3. Navigate to **Network Connections** to view the available network connections on the PC.
4. In the **Network Connections** window, right-click the network adapter that is being used to connect the Absolute Q™ companion PC to the bridge PC, then select **Properties**.
The **Properties** window for the selected network adapter opens.



5. In the **Networking** tab, select **Internet Protocol Version 4 (TCP/IPv4)** from the list, then click **Properties**.
6. In the **Internet Protocol Version 4 (TCP/IPv4) Properties** window, select the **Use the following IP address** option button, then enter the following information:

Field	Input
IP address	172.16.0.2 Note: For each additional Absolute Q™ companion PC, increment the last digit of the IP address. For example, when setting up an additional Absolute Q™ companion PC, enter 172.16.0.3 into the IP address field, and so on.
Subnet mask	255.255.255.0

7. Click **OK** to close the **Properties** windows, then close the **Network Connections** window.
8. For each additional Absolute Q™ companion PC, repeat step 1–step 7.

Note: Ensure that a unique IP address is used for each Absolute Q™ companion PC, as described in step 6.

On the bridge PC:

9. Press **Windows + R** to open the **Run** dialog box.
10. In the **Run** dialog box, in the **Open** field, enter `ncpa.cpl`, then click **OK**.
11. Navigate to **Network Connections** to view the available network connections on the PC.
12. In the **Network Connections** window, right-click the network adapter that is being used to connect the Absolute Q™ companion PCs to the bridge PC, then select **Properties**.
13. In the **Networking** tab, select **Internet Protocol Version 4 (TCP/IPv4)** from the list, then click **Properties**.
14. In the **Internet Protocol Version 4 (TCP/IPv4) Properties** window, select the **Use the following IP address** option button, then enter the following information:

Field	Input
IP address	172.16.0.1
Subnet mask	255.255.255.0

15. Click **OK** to close the **Properties** windows, then close the **Network Connections** window.

On an Absolute Q™ companion PC:

16. Use the **INSTR-ADMIN** account to sign in to the Absolute Q™ companion PC.
17. On the PC desktop, create a new folder named **BridgeShare**.
18. Right-click the **BridgeShare** folder, then select **Properties**.



19. In the **BridgeShare Properties** window, in the **Sharing** tab, click **Advanced Sharing**.
20. In the **Advanced Sharing** window, select **Share this folder**.
21. Click **Permissions**, then in the **Permissions for BridgeShare** window, click **Add**.
22. In the **Select Users or Groups** window, in the **Enter the object names to select** field, enter **INSTR-ADMIN**, then click **Check Names**.
23. In the **Permissions for BridgeShare** window, in the **Group or user names** pane, select **INSTR-ADMIN** user from the list, then in the **Permissions for INSTR-ADMIN** pane, select **Allow** in the **Full Control** row.
24. Click **OK** to close the **Permissions for BridgeShare** and **BridgeShare Properties** windows.

On the bridge PC:

25. Open **File Explorer**.
26. In the address field, enter the Absolute Q™ companion PC IP address, such as \\172.16.0.2.

IMPORTANT! Each Absolute Q™ companion PC has a unique IP address. Ensure that you enter the IP address that was designated in step 6 for the corresponding Absolute Q™ companion PC.

27. When prompted, enter the **INSTR-ADMIN** and password, then click **OK**.
28. Right-click the **BridgeShare** folder, then select **Map network drive**.
29. In the **Map network drive** window, select the drive from the **Drive** dropdown list, then click **Finish**.
The **BridgeShare** folder on the Absolute Q™ companion PC is mapped to the selected drive, and is now accessible from the bridge PC.
30. For each additional Absolute Q™ companion PC, repeat step 25–step 29.

The **BridgeShare** folder can now be used to transfer data between Absolute Q™ companion PCs and the bridge PC. Data transfer can be performed manually or by using the **Auto export completed runs** QuantStudio™ Absolute Q™ Digital PCR Software feature.



Data export information package for QuantStudio™ Absolute Q™ Digital PCR Software v6.3 or later.

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Data flow overview

This chapter provides information about the import and export functions of the QuantStudio™ Absolute Q™ Digital PCR Software that facilitate the integration of the QuantStudio™ Absolute Q™ Digital PCR Software with the Laboratory Information System (LIS) or Laboratory Information Management System (LIMS). Figure 33 describes the key integration points between the LIS/LIMS, middleware, and the QuantStudio™ Absolute Q™ Digital PCR Software.

Each LIS/LIMS workflow is unique and may require a different data configuration for integration. The information provided is for reference use only. For more information, contact your local instrument service group (see “Customer and technical support” on page 209) or e-mail:

instrumentservices@thermofisher.com.

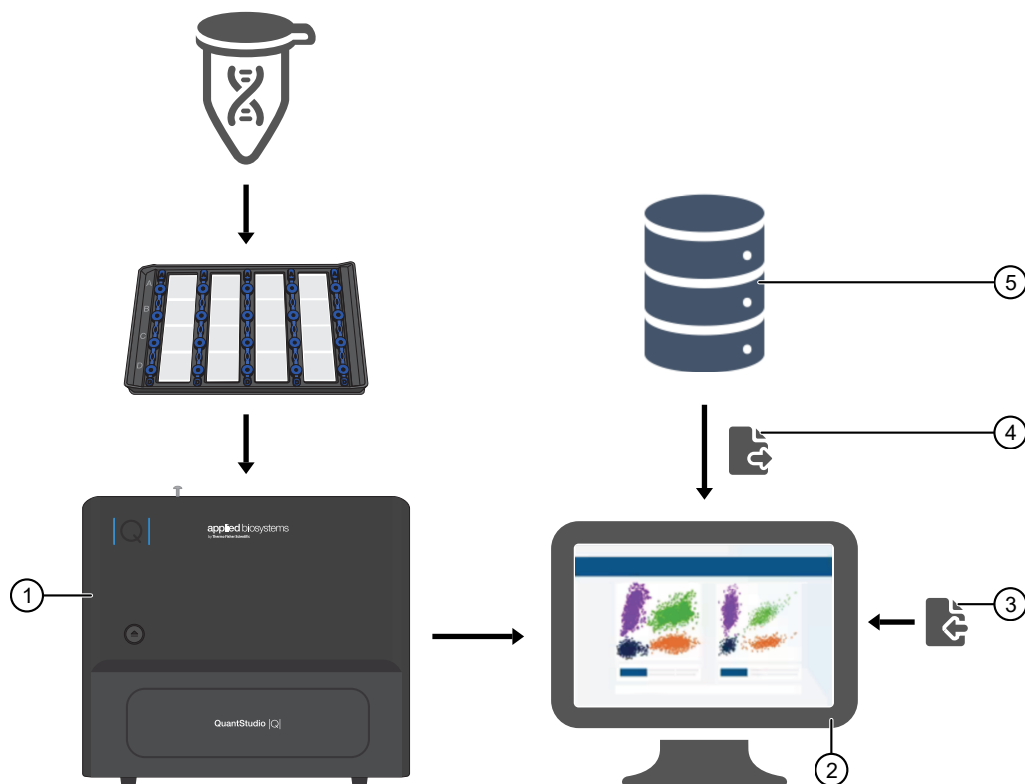


Figure 33 Data workflow

- ① QuantStudio™ Absolute Q™ Digital PCR System
- ② QuantStudio™ Absolute Q™ Digital PCR Software
- ③ File imports
- ④ Exported results
- ⑤ LIS/LIMS database

QuantStudio™ Absolute Q™ Digital PCR Software – Export

The export function of the QuantStudio™ Absolute Q™ Digital PCR Software can be performed at a sample level or a group (pre-defined collection of samples) level for a run or for a multi-plate analysis, otherwise known as a study. The exported analyses and result parameters also depend on the analysis type that is selected during the experiment setup. The following table provides a summary of data, analysis, and sample types that can be exported from the QuantStudio™ Absolute Q™ Digital PCR Software.

Type	Description
Data:	
Sample-level	Refers to results from the runs based on a single sample and analysis type selected.
Group-level	Refers to results from all samples in a selected group, including relevant statistical parameters such as standard deviation (SD), coefficient of variation (CV%), and 95% confidence interval (95%CI).



(continued)

Type	Description
Sample:	
Individual	Each sample has a separate result entry.
Replicates	The results show the Mean, Standard Deviation, and CV% for the concentration for all the samples in the group.
Pooling	All samples in the group are treated as one large sample.
Analysis:	
CNV	Reports the ratio of copy number variation (CNV) to CNV Ref.
CNV Ref	The reference target for CNV. $\text{CNV Ref} = ([\text{sample concentration}] \times [\text{dilution factor}]) \div [\text{reference concentration}]$
Quantification	Absolute quantification.
VAF Wild Type (variant allele detection)	Reports the concentration for the standard (or common) allele (WT) that is used to detect if the corresponding variant is present in a sample.
VAF Variant (variant allele detection)	Reports the concentration for the variant (or mutant) allele (VA) present in the sample; uses the WT and VA data to calculate variant allele frequency (VAF).
Not used	Ignored in the analysis: • No analysis for the sample • No results shown • Sample excluded from the report • Sample excluded from the calculations for individual, replicates, and pooling sample types

The QuantStudio™ Absolute Q™ Digital PCR Software provides the following options for exporting data. You can select one or multiple options to include in the download bundle.

Note: The column names in the CSV match those displayed in the Quantification table, but do not include the custom column configuration or sorting.

Option	Description
Results summary	Summary of the analysis (concentration values) Note: The Results summary CSV table format was updated for QuantStudio™ Absolute Q™ Digital PCR Software v6.4 and later. To download the Results summary CSV in v6.3 format, select the 6.3 format (legacy) .
Fluorescence	Fluorescence data for each dye channel
Sample based multichannel	Multi-channel results organized by sample
Channel based multichannel	Multi-channel results organized by channel



QuantStudio™ Absolute Q™ Digital PCR Software – Import

The import function of the QuantStudio™ Absolute Q™ Digital PCR Software facilitates the import of sample information and barcode information for creating batch run files. When plate barcodes are imported for creating batch run files, the software generates default sample names, in which case the import of sample information is optional. The software supports the following types of data:

- Sample information
- Barcode information

For more information, see Chapter 3, “Run digital PCR” and Chapter 4, “Analyze data”.

Definitions of data fields

For information about how the software algorithm calculates the results, see “Computation of results” on page 127.

Field	Content type	Description
95% CI	Number	95% confidence interval of the reported concentration of a target across wells [lower value, upper value]
95 % CI VAF	Number	95% confidence interval VAF delta [lower delta value, upper delta value].
% VAF	Number	Percent variant allele frequency (VAF) of variant (mutant) allele (VA) compared to the wild type (WT) allele.
(Variant allele detection data) [Dye name] Conc. (WT/VA)	Number	Concentration in copies per μ L. Note: In the Variant allele detection table, the analysis type is displayed next to the dye name. For example: FAM Conc. (WT) vs. ABY Conc. (VA) , where WT refers to wild type and VA refers to variant.
Channels	Text	Optical dye channels
CNV_95% CI	Number	95% confidence interval delta for copy number [lower delta value, upper delta value]
Conc. copies/ μ L	Number	Calculated absolute concentration in copies per microliter
Copies	Number	Copy number of the target of interest based on reference copy number
Copies per microliter	Number	Calculated number of targets per microliter
Count	Number	Number of microchambers with signal above the threshold
CV%	Number	Coefficient of variation of the reported concentration of a target across wells
Date	Text	Date in YYYY/MM/DD format
Dilution factor	Number	Dilution factor



(continued)

Field	Content type	Description
Dye	Text	Currently supported dyes by the software - FAM™, VIC™, HEX™, ABY™, JUN™, Cy5™, and ROX™ (QC dye)
Group	Text	Name of a collection of samples
Instrument	Text	The name of the instrument on which the run is performed
Labels	Text	Optical dye channels
Lambda (cp/Rxn)	Number	Average number of molecules per microchamber
Lambda_95% CI	Number	95% confidence interval delta for lambda (copies per reaction) [lower delta value, upper delta value]
Plate	Alphanumeric	Barcode assigned to the plate
Plate barcode	Alphanumeric	Barcode assigned to the plate
Positives	Number	Number of microchambers with signal above the threshold
Precision%	Number	Spread of confidence level of the reported concentration of a target
QC	Text	Quality result (PASS / WARNING / FAIL / CRITICAL)
Reject	Boolean	Annotation for each microchamber that designates whether the signal is above (accept/true) or below (reject/false) the set threshold
Run	Text	Run name
Run name	Text	Run name
Sample	Text	Unique sample identifier such as sample name, Accession ID, or Sample ID
Sample group	Text	The group that the sample is assigned to.
Sample name	Text	Sample name
SD	Number	Standard deviation of reported concentration of a target across wells
Target	Text	Target names assigned to a dye within a group
Threshold	Number	Threshold value for a given target, expressed as <dye name>_threshold
Total	Number	Number of microchambers used in the analysis
Type	Text	Sample type: Unknown, Positive Control, or Negative Control
Units	Text	Units for the data (for example: Conc. cp/μL)



(continued)

Field	Content type	Description
VAF Cutoff	Text	VAF result. The VAF Cutoff column lists the following values in the Variant allele detection results table: • Below <VAF cutoff>—samples with target below the specified VAF cutoff. • At <VAF cutoff>—samples with target at the specified VAF cutoff. • Above <VAF cutoff>—samples with target above the specified VAF cutoff. The VAF cutoff values are specified when creating or editing a sample group at the time of template or run creation or after a run is completed. For more information, see “Manage groups” on page 70.
Well	Alphanumeric	Sample position range, from A1 to D4



Product specifications

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QuantStudio™ Absolute Q™ Digital PCR Instrument specifications

Dimensions (unpacked)	620 mm (l) x 600 mm (w) x 540 mm (h) 24.5 in (l) x 23.5 in (w) x 21.2 in (h)
Dimensions (packaged)	860 mm (l) x 860 mm (w) x 790 mm (h) 33.5 in (l) x 34 in (w) x 30 in (h)
Weight	Approximately 60 kg, 132 lbs
Connections	Power, USB 3.0 (to dedicated computer)
Cooling mode	Forced convection
Illumination	Red, Blue, Phosphor Green high-power LED
Optical channels	5 (fixed configuration)
Power input	100-240 V, 50-60Hz
Power rating	1200-1600 W
Rated current	12 A (110V), 8.5 A (230 V)
Maximum noise level	70 dB



QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite specifications

Table 24 QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite, single configuration

Dimensions (unpacked)	168 cm (l) x 65 cm (w) x 80 cm (h) 66 in (l) x 25.6 in (w) x 31.5 in (h)
Weight	Approximately 85 kg, 187 lbs
Cooling mode	Forced convection
Illumination	Red, Blue, Phosphor Green high-power LED
Optical channels	5 (fixed configuration)
Power input	100-240 V, 50-60Hz
Power rating	1200-1600 W
Rated current	12 A (110V), 8.5 A (230 V)
Maximum noise level	70 dB

Table 25 QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite, dual configuration

Dimensions (unpacked)	239 cm (l) x 80 cm (w) x 80 cm (h) 93.9 in (l) x 31.5 in (w) x 31.5 in (h)
Weight	Approximately 145 kg, 319 lbs
Cooling mode	Forced convection
Illumination	Red, Blue, Phosphor Green high-power LED
Optical channels	5 (fixed configuration)
Power input	100-240 V, 50-60Hz
Power rating	1200-1600 W
Rated current	12 A (110V), 8.5 A (230 V)
Maximum noise level	70 dB

Dedicated computer requirements

Operating system	Windows™ 10 (64-bit) or later
Computer	Dell™ Tower



QuantStudio™ Absolute Q™ Digital PCR Instrument Optical Configuration

The QuantStudio™ Absolute Q™ Digital PCR Instrument comes in a single optical configuration and is pre-calibrated during manufacturing. It can be field calibrated for enhanced spectral compensation.

Note:

- For information about HEX™ dye support, contact a Thermo Fisher Scientific and support representative.
- A warning message appears in the **Instrument** screen 45 days before dye calibration expiration. If the dyes are not calibrated within that time frame, a warning message appears indicating that the calibration has expired and remains on the **Instrument** screen until the dyes are calibrated.
- Contact technical support to request custom calibration service (see Appendix K, “Documentation and support”).

The following optical configuration is default in QuantStudio™ Absolute Q™ Digital PCR Software version 6.4 and later:

Color (channel)	Excitation filter peak	Emission filter peak	System dyes
Blue	466	520	FAM™ dye
Green	514	560	VIC™ dye (<i>recommended</i>) HEX™ dye ^[1,2]
Yellow	549	589	ABY™ dye
Red	589	625	JUN™ dye ROX™ dye
Dark Red	630	684	Cy5™ dye

^[1] For information about HEX™ dye support, contact a Thermo Fisher Scientific service and support representative.

^[2] HEX™ data from two instruments cannot be combined into a study, even if the systems are co-calibrated. For more information, see “Multi-plate analysis (studies)” on page 107.

When using templates, draft runs, or completed runs and studies from QuantStudio™ Absolute Q™ Digital PCR Software version 6.3 or earlier, the software version 6.4 and later adapts the protocol and setup settings to account for the conflicting differences in the configuration as described in the following table.



Channels and dye selections from software v6.3 or earlier	Adaptation in software v6.4 and later
Templates and draft runs	
The Dark Red channel was enabled and assigned to JUN™.	In the SETUP tab, in the Edit sample group dialog, the Dark Red (CY5) channel is disabled. You can edit your channel (dye) selections in the PROTOCOL tab. You can also create new sample groups in the SETUP tab and enable the desired channels.
The Dark Red channel was enabled and assigned to Cy5™.	In the SETUP tab, in the Edit sample group dialog, the Red (JUN) channel is disabled. You can edit your channel (dye) selections in the PROTOCOL tab. You can also create new sample groups in the SETUP tab and enable the desired channels.
The Dark Red channel was disabled.	In the PROTOCOL tab, both the Dark Red (CY5) and Red (JUN) channels are disabled and cannot be enabled. In the SETUP tab, in the Edit sample group dialog, both the Dark Red (CY5) and Red (JUN) channels are not available for selection.
Completed runs and studies	
The Dark Red channel was enabled and assigned to JUN™.	In the PROTOCOL, the Dark Red (CY5) channel is disabled and cannot be enabled. In the SETUP tab, in the Edit sample group dialog, the Dark Red (CY5) channel is not available for selection.
The Dark Red channel was enabled and assigned to Cy5™.	In the PROTOCOL, the Red (JUN) channel is disabled and cannot be enabled. In the SETUP tab, in the Edit sample group dialog, the Red (JUN) channel is not available for selection.
The Dark Red channel was disabled.	In the PROTOCOL tab, both the Dark Red (CY5) and Red (JUN) channels are disabled and cannot be enabled. In the SETUP tab, in the Edit sample group dialog, both the Dark Red (CY5) and Red (JUN) channels are not available for selection.



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, visit thermofisher.com/support.



AVERTISSEMENT ! SÉCURITÉ GÉNÉRALE. L'utilisation de ce produit d'une manière non spécifiée dans le manuel d'utilisation peut entraîner des blessures ou endommager l'instrument ou l'appareil. Assurez-vous que toute personne utilisant ce produit est formée aux pratiques générales de sécurité pour les laboratoires et aux informations de sécurité fournies dans le présent document.

- Avant d'utiliser un instrument ou un dispositif, lisez et assimilez les informations de sécurité figurant dans le manuel d'utilisation fourni par le fabricant de l'instrument ou du dispositif.
- Avant de manipuler des produits chimiques, lisez et assimilez toutes les fiches de données de sécurité (FDS) applicables et utilisez les équipements de protection individuelle appropriés (gants, blouses, lunettes de protection, etc.). Pour consulter les fiches de données de sécurité, rendez-vous sur le site thermofisher.com/support.

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.

Standard safety symbols

Symbol and description	
	CAUTION! Risk of danger. Consult the manual for further safety information.
	CAUTION! Caution, air inlet.
	CAUTION! Hot surface.
	CAUTION! Potential biohazard.

Symbole et description	
	MISE EN GARDE ! Risque de danger. Consulter le manuel pour d'autres renseignements de sécurité.
	MISE EN GARDE ! Risque de choc électrique.
	MISE EN GARDE ! Surface chaude.
	MISE EN GARDE ! Danger biologique potentiel.

Control and connection symbols

Symbols and descriptions	
	On (Power)
	Off (Power)
	Protective conductor terminal (main ground)

Conformity symbols

Conformity mark	Description
    	Indicates conformity with safety requirements for Canada and U.S.A.
	INDICATES CONFORMITY WITH THE WEEE DIRECTIVE 2012/19/EU.  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 réduire l'empreinte écologique résultant de l'élimination des composants électroniques, ne les jetez pas dans les déchets municipaux non triés. Respectez les réglementations locales en matière de déchets pour un traitement approprié et contactez le service clientèle pour en savoir plus sur les solutions responsables.

Location of safety labels

Label and location



Figure 34 Detection cell cover (front view, door open)

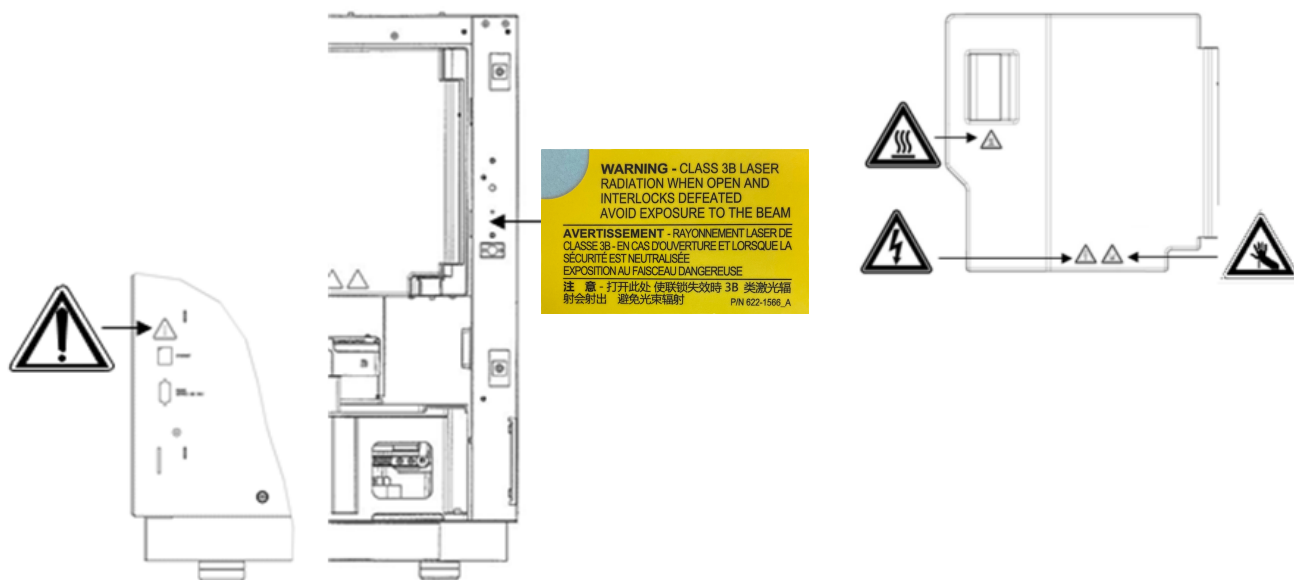


Figure 35 Rear panel (left) oven door (right)

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.



MISE EN GARDE ! Ne retirez pas les couvercles de protection de l'instrument. Si vous retirez les panneaux de protection des instruments ou si vous désactivez les dispositifs de verrouillage, vous risquez de courir de graves dangers, comme, par exemple, un choc électrique, une exposition au laser, un écrasement ou une exposition à des produits chimiques.

Spinnaker™ Microplate Mover general safety



CAUTION! When working around the Spinnaker™ Microplate Mover, ensure that the following safety requirements are met.

- Only trained and authorized personnel may work with or around the robot.
- The robot is a fully automated device that may move at any time while under the control of the Momentum™ Workflow Scheduler Software or another system automation management application. While it is under the control of the software, remain outside the workspace of the robot and do not attempt to handle the robot. Doing so may result in injury.
- Spills may occur while the robot is transporting a container. This may be due to collision with operators or other obstructions, an improperly taught location, or human error while positioning containers for the robot to transport. If the robot is being used to transport hazardous materials, before beginning operation, ensure that all precautions necessary to protect operators from exposure are in place.



MISE EN GARDE ! Lorsque vous travaillez autour du Spinnaker™ Microplate Mover, assurez-vous de respecter les consignes de sécurité suivantes.

- Seul le personnel formé et autorisé peut travailler avec ou autour du robot.
- Le robot est un dispositif entièrement automatisé pouvant se déplacer à tout moment sous le contrôle de Momentum™ Workflow Scheduler Software ou d'une autre application de gestion de l'automatisation du système. Restez en dehors de l'espace de travail du robot pendant qu'il est sous le contrôle du logiciel et n'essayez pas de le manipuler. Cela peut entraîner des blessures.
- Des déversements peuvent se produire lorsque le robot transporte un récipient. Cela peut être dû à une collision avec des opérateurs ou d'autres obstacles, à un emplacement mal enseigné ou à une erreur humaine lors du positionnement des récipients à transporter par le robot. Si le robot est utilisé pour transporter des matières dangereuses, il convient de s'assurer avant le début de l'opération que toutes les précautions nécessaires à la protection des opérateurs contre l'exposition sont en place.

Hot Surface



CAUTION! Hot surface. During instrument operation, the temperature of the plate nest can be as high as 100°C. The instrument has a software interlock to prevent the door from opening if the plate nest temperature is over 45°C, but if the system appears to be malfunctioning use caution when operating near the plate nest.



MISE EN GARDE ! Surface chaude. En cours de fonctionnement, la température des plaques peut atteindre 100°C. L'instrument est doté d'un logiciel de verrouillage qui empêche l'ouverture de la porte si la température des plaques est supérieure à 45°C. Toutefois, si le système semble présenter un dysfonctionnement, soyez prudent lorsque vous travaillez à proximité des plaques.

Air inlet



CAUTION! Air inlet. Air inlet is only suitable for atmospheric air and not pressurized gas. Do not connect flammable gas to the air inlet port. Do not restrict air inlet port.



MISE EN GARDE ! Arrivée d'air. L'arrivée d'air ne convient qu'à l'air atmosphérique et non aux gaz sous pression. Ne raccordez pas de gaz inflammable à l'orifice d'arrivée d'air. Veillez à ne pas obstruer l'orifice d'arrivée d'air.

Physical injury



CAUTION! Moving and Lifting Injury. 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.



MISE EN GARDE ! Blessures causées par le déplacement et le soulèvement. Soulever de manière inappropriée peut provoquer des lésions dorsales douloureuses et permanentes.

Éléments à prendre en compte avant de soulever ou de déplacer l'instrument ou ses accessoires:

- Selon le poids, deux personnes ou plus peuvent être nécessaires pour déplacer ou soulever l'instrument.
- Si vous décidez de soulever ou de déplacer l'instrument après son installation, n'essayez pas de le faire seul, sans un équipement approprié et sans avoir recours à des techniques appropriées.
- Assurez-vous d'avoir une prise sûre et confortable sur l'instrument ou l'accessoire.
- Assurez-vous que le chemin entre l'endroit où se trouve l'objet et l'endroit où il est déplacé est libre de tout obstacle.
- Ne soulevez pas un objet et ne pivotez pas votre torse en même temps. Tenez votre colonne vertébrale dans une position bien droite en vous relevant.
- Les participants doivent coordonner leurs mouvements avant de soulever et de porter.
- Pour les petits colis, au lieu de soulever l'objet de son emballage, inclinez soigneusement le carton sur le côté et maintenez-le immobile pendant que quelqu'un d'autre fait glisser le contenu hors du carton.

Electrical safety



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.



AVERTISSEMENT ! Veiller à utiliser une alimentation électrique appropriée. Pour garantir le fonctionnement de l'instrument en toute sécurité :

- Brancher le système sur une prise électrique correctement mise à la terre et de puissance adéquate.
- S'assurer que la tension électrique est convenable.
- Ne jamais utiliser l'instrument alors que le dispositif de mise à la terre est déconnecté. La continuité de la mise à la terre est impérative pour le fonctionnement de l'instrument en toute sécurité.



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



AVERTISSEMENT ! Cordons d'alimentation électrique. Utiliser des cordons d'alimentation adaptés et approuvés pour raccorder l'instrument au circuit électrique du site.



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



AVERTISSEMENT ! Déconnecter l'alimentation. Pour déconnecter entièrement l'alimentation, détacher ou débrancher le cordon d'alimentation. Placer l'instrument de manière à ce que le cordon d'alimentation soit accessible.

Cleaning and decontamination



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

- No decontamination or cleaning agents are used that can react with parts of the equipment or with material that is contained in the equipment. Use of such agents could cause a HAZARD condition.
- The instrument is properly decontaminated a) if hazardous material is spilled onto or into the equipment, and/or b) before the instrument is serviced at your facility or is sent for repair, maintenance, trade-in, disposal, or termination of a loan. Request decontamination forms from customer service.
- Before using any cleaning or decontamination methods (except methods that are recommended by the manufacturer), confirm with the manufacturer that the proposed method will not damage the equipment.



MISE EN GARDE ! Nettoyage et décontamination. Utiliser uniquement les méthodes de nettoyage et de décontamination indiquées dans la documentation du fabricant destinée aux utilisateurs. L'opérateur (ou toute autre personne responsable) est tenu d'assurer le respect des exigences suivantes:

- Ne pas utiliser d'agents de nettoyage ou de décontamination susceptibles de réagir avec certaines parties de l'appareil ou avec les matières qu'il contient et de constituer, de ce fait, un DANGER.
- L'instrument doit être correctement décontaminé a) si des substances dangereuses sont renversées sur ou à l'intérieur de l'équipement, et/ou b) avant de le faire réviser sur site ou de l'envoyer à des fins de réparation, de maintenance, de revente, d'élimination ou à l'expiration d'une période de prêt (des informations sur les formes de décontamination peuvent être demandées auprès du Service clientèle).
- Avant d'utiliser une méthode de nettoyage ou de décontamination (autre que celles recommandées par le fabricant), les utilisateurs doivent vérifier auprès de celui-ci qu'elle ne risque pas d'endommager l'appareil.

Spinnaker™ Microplate Mover safety during maintenance



CAUTION! When performing maintenance tasks on the robot, observe the following guidance. Failure to do so may result in serious injury or death.

- Do not attempt to disassemble the supply adapter of the mover or any other electrical components. Doing so may result in a lethal electric shock.
- Before cleaning the mover, ensure that it has been completely de-energized and disconnected from the power supply.



MISE EN GARDE ! Respectez les consignes suivantes lorsque vous effectuez des opérations d'entretien sur le robot. Le non-respect de cette consigne peut entraîner de graves blessures ou la mort.

- N'essayez pas de démonter l'adaptateur d'alimentation du moteur ou tout autre composant électrique. Cela pourrait entraîner un choc électrique mortel.
- Assurez-vous que le moteur a été complètement mis hors tension et déconnecté de l'alimentation électrique avant de le nettoyer.

Instrument component and accessory disposal



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 réduire l'empreinte écologique résultant de l'élimination des composants électroniques, ne les jetez pas dans les déchets municipaux non triés. Respectez les réglementations locales en matière de déchets pour un traitement approprié et contactez le service clientèle pour en savoir plus sur les solutions responsables.

Safety and electromagnetic compatibility (EMC) standards

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

Safety standards

Reference	Description
EU Directive 2011/65/EU & Commission Delegated Directive (EU) 2015/863	European Union “RoHS Directive” – Restriction of hazardous substances in electrical and electronic equipment
IEC 61010-1	<i>Safety requirements for electrical equipment for measurement, control, and laboratory use – Part 1: General requirements</i>
IEC 61010-2-010	<i>Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials</i>
IEC 61010-2-081	<i>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</i>

EMC standards

Reference	Description
EMC EN 61326-1	<i>Electrical Equipment for Measurement, Control and Laboratory Use – EMC Requirements – Part 1: General Requirements</i>
FCC Class A equipment Caution	This device complies with Part 15 of the FCC rules. Operation is subject to the following two conditions: 1. This device may not cause harmful interference, and 2. This device must accept any interference received, including interference that may cause undesired operation.
FCC Part 15 Subpart B (47 CFR)	<i>U.S. Standard Radio Frequency Devices</i> 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.

Environmental design standards

Reference	Description
Directive 2012/19/EU	European Union “WEEE Directive”—Waste electrical and electronic equipment
Directive 2011/65/EU	European Union “RoHS Directive”—Restriction of hazardous substances in electrical and electronic equipment
SJ/T 11364-2014	“China RoHS” Standard—Marking for the Restricted Use of Hazardous Substances in Electronic and Electrical Products For instrument specific certificates, visit our customer resource page at www.thermofisher.com/us/en/home/technical-resources/rohs-certificates.html .

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.



AVERTISSEMENT ! PRÉCAUTIONS GÉNÉRALES EN CAS DE MANIPULATION DE PRODUITS CHIMIQUES. Pour minimiser les risques, veiller à ce que le personnel du laboratoire lise attentivement et mette en œuvre les consignes de sécurité générales relatives à l'utilisation et au stockage des produits chimiques et à la gestion des déchets qui en découlent, décrites ci-dessous. Consulter également la FDS appropriée pour connaître les précautions et instructions particulières à respecter :

- Lire et comprendre les fiches de données de sécurité (FDS) fournies par le fabricant avant de stocker, de manipuler ou d'utiliser les matériaux dangereux ou les produits chimiques. Pour obtenir les FDS, se reporter à la section « Documentation et support » du présent document.
- Limiter les contacts avec les produits chimiques. Porter des équipements de protection appropriés lors de la manipulation des produits chimiques (par exemple : lunettes de sûreté, gants ou vêtements de protection).
- Limiter l'inhalation des produits chimiques. Ne pas laisser les récipients de produits chimiques ouverts. Ils ne doivent être utilisés qu'avec une ventilation adéquate (par exemple, sorbonne).
- Vérifier régulièrement l'absence de fuite ou d'écoulement des produits chimiques. En cas de fuite ou d'écoulement d'un produit, respecter les directives de nettoyage du fabricant recommandées dans la FDS.
- Manipuler les déchets chimiques dans une sorbonne.

- Veiller à utiliser des récipients à déchets primaire et secondaire. (Le récipient primaire contient les déchets immédiats, le récipient secondaire contient les fuites et les écoulements du récipient primaire. Les deux récipients doivent être compatibles avec les matériaux mis au rebut et conformes aux exigences locales, nationales et communautaires en matière de confinement des récipients.)
- Une fois le récipient à déchets vidé, il doit être refermé hermétiquement avec le couvercle fourni.
- Caractériser (par une analyse si nécessaire) les déchets générés par les applications, les réactifs et les substrats particuliers utilisés dans le laboratoire.
- Vérifier que les déchets sont convenablement stockés, transférés, transportés et éliminés en respectant toutes les réglementations locales, nationales et/ou communautaires en vigueur.
- **IMPORTANT !** Les matériaux représentant un danger biologique ou radioactif exigent parfois une manipulation spéciale, et des limitations peuvent s'appliquer à leur élimination.

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.



AVERTISSEMENT ! Risque biologique potentiel. En fonction des échantillons utilisés sur cet instrument, la surface peut être considérée comme présentant un risque biologique. Utilisez des méthodes de décontamination appropriées lorsque vous travaillez en présence de risques biologiques.



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
[cdc.gov/labs/bmbi](https://www.cdc.gov/labs/bmbi)
- Laboratory biosafety manual, fourth edition. Geneva: World Health Organization; 2020 (Laboratory biosafety manual, fourth edition, and associated monographs)
[who.int/publications/i/item/9789240011311](https://www.who.int/publications/i/item/9789240011311)



AVERTISSEMENT ! RISQUE BIOLOGIQUE. Les échantillons biologiques tels que les tissus, les fluides corporels, les agents infectieux et le sang de l'homme et d'autres animaux sont susceptibles de transmettre des maladies infectieuses. Effectuez tous vos travaux dans des installations correctement équipées et dotées du matériel de sécurité approprié (par exemple, des dispositifs de confinement physique). L'équipement de sécurité peut également inclure des articles de protection personnelle, tels que des gants, des manteaux, des blouses, des couvre-chaussures, des bottes, des respirateurs, des masques faciaux, des lunettes de sécurité ou des lunettes de protection. Les personnes doivent être formées conformément aux exigences réglementaires applicables et aux exigences de l'entreprise ou de l'institution avant de travailler avec des matières potentiellement dangereuses. Respectez toutes les réglementations locales, nationales et/ou provinciales applicables. Les références suivantes proposent des recommandations générales pour la manipulation d'échantillons biologiques dans un environnement de laboratoire.

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 6th Edition, HHS Publication No. (CDC) 300859, révision de juin 2020
[cdc.gov/labs/bmbi](https://www.cdc.gov/labs/bmbi)
- Laboratory biosafety manual, fourth edition. Genève: Organisation mondiale de la santé; 2020 (Laboratory biosafety manual, fourth edition, et monographies associées)
[who.int/publications/i/item/9789240011311](https://www.who.int/publications/i/item/9789240011311)



Documentation and support

■ Related documentation	208
■ Customer and technical support	209
■ Limited product warranty	209

Related documentation

Document	Publication number	Description
<i>QuantStudio™ Absolute Q™ Digital PCR Software Installation Guide</i>	MAN1001443	Describes how to download, install, and update the QuantStudio™ Absolute Q™ Digital PCR Software, including instructions for updating the embedded QuantStudio™ Absolute Q™ Digital PCR Instrument software and firmware.
<i>QuantStudio™ Absolute Q™ Digital PCR Starter Kit User Guide</i>	MAN0025653	Describes the setup, use, and analysis of runs using the QuantStudio™ Absolute Q™ Digital PCR Starter Kit assay (Cat. No. A52732).
<i>QuantStudio™ Absolute Q™ Digital PCR System Site Preparation Guide</i>	MAN0026431	Describes the site preparation required for installing the QuantStudio™ Absolute Q™ dPCR System.
<i>QuantStudio™ Absolute Q™ Digital PCR System IT Checklist</i>	MAN0028309	Describes the IT setup requirements for successful installation of the QuantStudio™ Absolute Q™ dPCR System, and for the effective support of the instrument by Thermo Fisher Scientific.
<i>QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite Site Preparation Guide</i>	MAN0029736	Describes the site preparation required for installing the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite.
<i>QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite IT Checklist</i>	MAN0029237	Describes the IT setup requirements for successful installation of the QuantStudio™ Absolute Q™ dPCR System, and for the effective support of the QuantStudio™ Absolute Q™ AutoRun Digital PCR Suite instrument by Thermo Fisher Scientific.
<i>QuantStudio™ Absolute Q™ Digital PCR System Remote API User Guide</i>	MAN0028059	Describes the Remote API for the automation of the QuantStudio™ Absolute Q™ dPCR System.



(continued)

Document	Publication number	Description
<i>QuantStudio™ Absolute Q™ Systems Software/Firmware Compatibility and User Documents Reference</i>	MAN0025886	Describes the compatibility of the QuantStudio™ Absolute Q™ Digital PCR Software with SAE Administrator Console, Momentum™ Workflow Scheduler Software, and QuantStudio™ Absolute Q™ Digital PCR Instrument software and firmware.
<i>SAE Administrator Console v2 User Guide for PCR systems</i>	MAN0017468	Describes the setup and use of the Security, Auditing, and E-signature (SAE) module.
<i>SAE Administrator Console v3 User Guide</i>	MAN1000386	

Note: For additional documentation, see “Customer and technical support” on page 209.

Customer and technical support

Visit [thermofisher.com/support](https://www.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 its affiliates 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 questions, contact Life Technologies at www.thermofisher.com/support.

