applied biosystems

QuantStudio™ Absolute Q™ Digital PCR System

INSTALLATION, USE, AND MAINTENANCE

Catalog Number A52864

Publication Number MAN0025621

Revision E.0

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

ThermoFisher SCIENTIFIC

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For descriptions of symbols on product labels or product documents, go to thermofisher.com/symbols-definition.

Revision history: MAN0025621 E.0 (English)

Revision	Date	Description
E.0	11 April 2023	• The manufacturing site address was updated to Singapore. • The recommended equipment and a link to the recommended kits for nucleic acid isolation were added ("Recommended materials not supplied" on page 10). • Information on nucleic acid isolation was added (see "Nucleic acid isolation" on page 19). • The optical dyes were revised to identify the recommended dyes. • Translations were added to safety information (see "Installation and environmental requirements" on page 84 and Appendix G, "Safety").
D.0	31 August 2022	• New filter feature is available for filtering runs by run type, status, and instrument when selecting a run. • Instructions were added for using the keyboard shortcuts for zooming in and out on pages ("Software features" on page 37). • Instructions were added for preparing DNA samples ("Prepare the DNA samples" on page 19). • Instructions were revised for preparing the dPCR reagent mix ("Prepare the dPCR reaction mix" on page 22). • Instructions were added for creating a custom protocol using the Absolute Q™ Starter or other existing protocols ("Create a custom protocol" on page 29) . • Instructions were revised for editing the sample plate area ("Load the plate and run the protocol" on page 32). • Instructions were revised for editing the sample plate area to modify the run name, select and name samples, and manage groups and group sets ("SETUP page" on page 39). • Instructions were added for deleting a group set ("Delete group sets" on page 53). • Instructions were added for changing a group set name ("Edit group set names" on page 52). • Instructions were revised for navigating plots ("View plots" on page 63). • Instructions were revised for manually setting thresholds ("Set thresholds" on page 66). • Instructions were added for overlaying sample plots with identical thresholds ("Overlay samples" on page 68). • Instructions were added for updating the instrument software ("Update the instrument software" on page 87). • Instructions were revised for adding e-Signatures for plate setup and run results ("Sign data in the software" on page 99). • Instructions were revised for installing the SAE Administrator Console software and Absolute Q™ application profile ("Install the SAE Administrator Console and Absolute Q™ application profile" on page 93). • Information was added and revised regarding auditing at the SAE Administrator Console ("Use audit functions" on page 97). • Information was added to describe the information regarding e-Signature data that is captured at the SAE Administrator Console ("View and review e-Signatures" on page 99). • Information was added regarding system dye calibration ("Maintenance" on page 113). • Information was added regarding troubleshooting actions to take if the connection to the SAE Administrator Console is lost (Appendix E, "Troubleshooting"). • Instructions were added for using FSA files for troubleshooting run and instrument issues ("Field Service Archive files" on page 116).

Revision	Date	Description
C.0	29 September 2021	• Added an appendix documenting the use of Security, Auditing, and E-signature (SAE) v2.2 software with QuantStudio™ Absolute Q™ Digital PCR Software (Appendix C, "Use the software with Security, Auditing, and E-signature (SAE) v2.2"). • Information was added regarding the Security, Auditing, and E-signature (SAE) v2.2 software ("Software description" on page 11). • Information was added for the use of the Security, Auditing, and E-signature (SAE) v2.2 software for system security ("QuantStudio™ Absolute Q™ Digital PCR Software security" on page 17). • Instructions were revised for placing MAP plate gasket strips on the MAP plate ("Load the reagent mix into the MAP plate" on page 23).
B.0	13 September 2021	• Updated graphics in the Analysis page section with optical dye channel information. • Instructions were revised for preparing the dPCR reagent mix with steps to thaw or equilibrate reagents before use and to vortex Absolute Q™ DNA Digital PCR Master Mix (5X) and Digital PCR assay ("Prepare the dPCR reaction mix" on page 22).
A.0	1 September 2021	New publication documenting instrument functions and data analysis features of the QuantStudio™ Absolute Q™ Digital PCR System.

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

Contents

1 Product information

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

Product description

The 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 and then processed by the QuantStudio™ Absolute Q™ Digital PCR Instrument. Depending on the protocol, results can be provided in less than 90 minutes. The resulting data are visualized with the QuantStudio™ Absolute Q™ Digital PCR Software. The QuantStudio™ Absolute Q™ Digital PCR System is for research use only, not for use in diagnostic procedures.

Instruments, kits, consumables, and accessories

The following table describes the products covered in this user guide.

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

Instrument system		
Item	**Cat. No.**	**Amount**
QuantStudio™ Absolute Q™ Digital PCR System: • QuantStudio™ Absolute Q™ Digital PCR Instrument • Dell™ OptiPlex XE3 Tower computer with monitor, keyboard, and mouse	A52864	1 instrument, 1 desktop computer and monitor
Instrument accessories		
Item	**Catalog No.**	**Amount**
QuantStudio™ Absolute Q™ MAP16 Plate Kit includes: • 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™ Digital PCR Starter Kit [1]	A52732	1
Reagents		
Item	**Catalog No.**	**Amount**
Absolute Q™ DNA Digital PCR Master Mix (5X)	A52490	200 reactions
QuantStudio™ Absolute Q™ Isolation Buffer	A52730	(1) 3 mL bottle

[1] The kit is 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. For more information contact your local sales representative or go to **http://www.thermofisher.com/dpcr-assays.html**.

For information on the use of pre-designed dPCR assays, see the documentation provided with the assay available at **http://www.thermofisher.com/dpcr-assays.html**.

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

Required materials not supplied

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

Item	Source
Equipment	
Centrifuge, table top	MLS
Pipettes, P10, P20, and P200	MLS
Filter pipette tips, P10, P20, and P200	MLS
Other consumables	
Low bind microcentrifuge tubes	MLS
Microcentrifuge tube rack	MLS
Nuclease-Free Water	MLS
Microfiber or optical lens cleaning cloth	MLS
70% ethanol in water	MLS

Recommended materials not supplied

Note: For information on the recommended kits for nucleic acid isolation, see **http://thermofisher.com/magmax**.

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

Item	Source
Spectrophotometer	MLS
Qubit™ Flex Fluorometer	Q33327
KingFisher™ Apex with 24 Combi Head	5400940
KingFisher™ Apex with 96 Deep-Well Head	5400930

Software description

The QuantStudio™ Absolute Q™ Digital PCR System uses the following software:

- QuantStudio™ Absolute Q™ Digital PCR Software—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 prior to the start of data generation. The software lets you to perform the following tasks:
  - Define the experiment, including sample types, sample groups, replicates, pool sample, experiment notes, and names
  - Create and edit protocols
  - Run and monitor protocols
  - View system status
  - View data in plot and tables
  - Generate run reports
  - Export data and reports
  - Insert and remove MAP plates
  - Install the shipping lock screw for transport of the instrument
- ThermoFisher Connect Transfer Software (Optional)—Data transmission feature that collects instrument run data to send to Thermo Fisher Scientific to be used for improving the product and user experience.
- Security, Auditing, and E-signature (SAE) v2.2 (Optional)—Controls security and user access to the software and specific features. See Appendix C, “Use the software with Security, Auditing, and E-signature (SAE) v2.2”.

The software is installed during system installation. See “Download and install the desktop software” on page 86.

Instrument hardware description

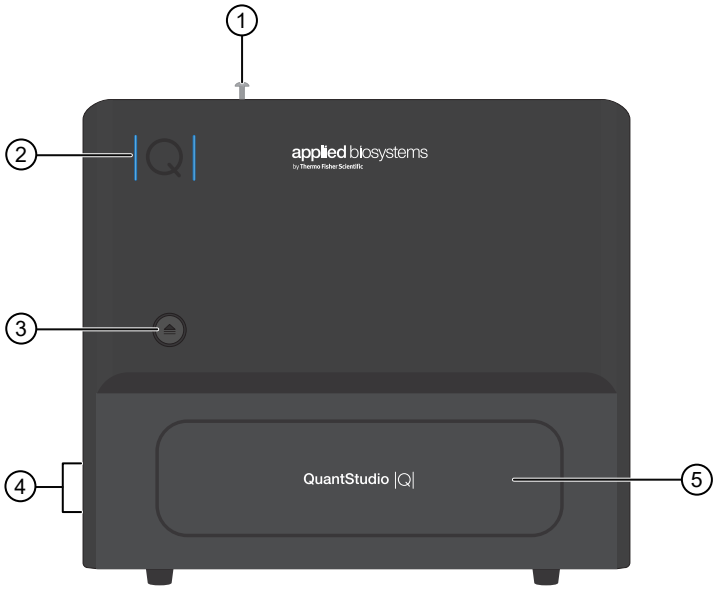
Overview of the instrument

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 on installing the instrument, see Appendix B, “Install, update, and move the QuantStudio™ Absolute Q™ Digital PCR System”.

For information on maintaining the instrument, see Appendix D, “Maintain the instrument”.



1. Shipping lock screw
2. Status indicator light
3. Plate presentation tray open/close button
4. Power switch, power port, USB port
5. Plate presentation tray

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 13.
- 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 microreaction chambers on the MAP plate before starting PCR. PCR occurs in parallel across the entire MAP plate. Each microreaction chamber contains a discrete reaction.
- The microreaction chamber 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	White	Steady	On, not connected to software.
Q	Blue	Steady	On, ready.
~ Q ~	Blue	Pulsing	Running protocol.
– Q –	Yellow	Brief flashing	Plate door open button pushed while door is locked.
Q	Red	Steady	Error, see Appendix E, “Troubleshooting”.

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 on optical configuration, see “QuantStudio™ Absolute Q™ Digital PCR Instrument Optical Configuration” on page 119.

Channel color	System dyes
Blue	FAM™ dye
Green	VIC™ dye *(recommended)* HEX™ dye
Yellow	ABY™ dye
Red	ROX™ dye
Dark Red	CY™5 dye *(recommended)* JUN™ dye

QuantStudio™ Absolute Q™ MAP16 Digital PCR Plates

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 microreaction chamber arrays that each contain 20,480 fixed volume microreaction chambers 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.

Note: MAP plate gasket strips can only be used once per MAP plate.

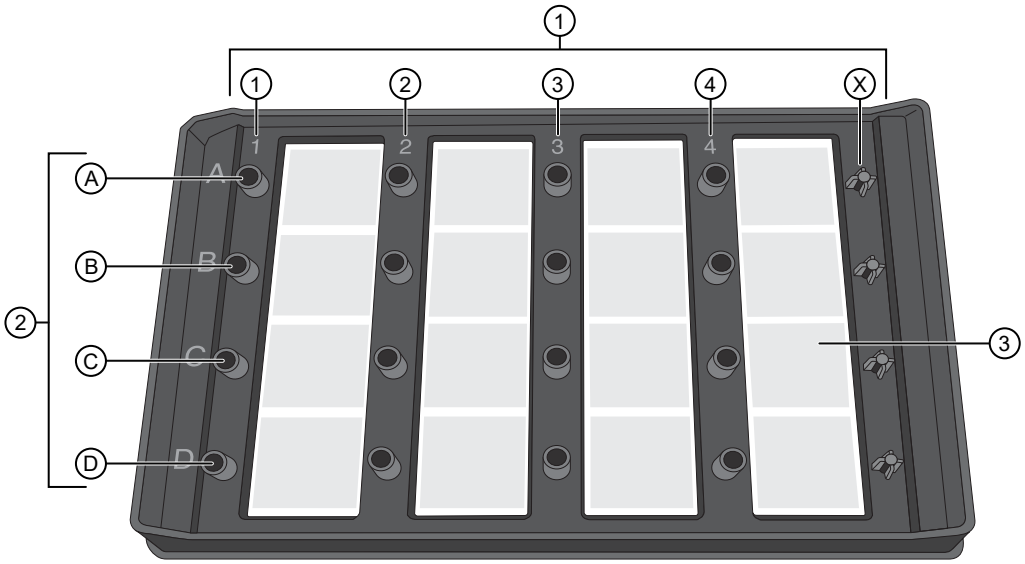


Figure 1 MAP plate without MAP plate gasket strips

1. Columns 1–4 and column X
2. A–D represents wells A1–D1 associated with column 1
3. Microreaction chamber associated with well 4C

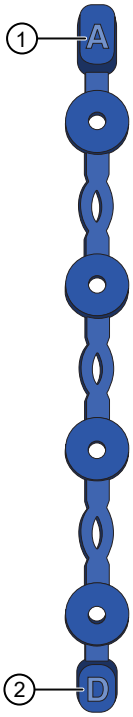


Figure 2 MAP plate gasket strip

① This end of the MAP plate gasket strip is placed on row A of the MAP plate
② This end of the MAP plate gasket strip is placed on row D of the MAP plate

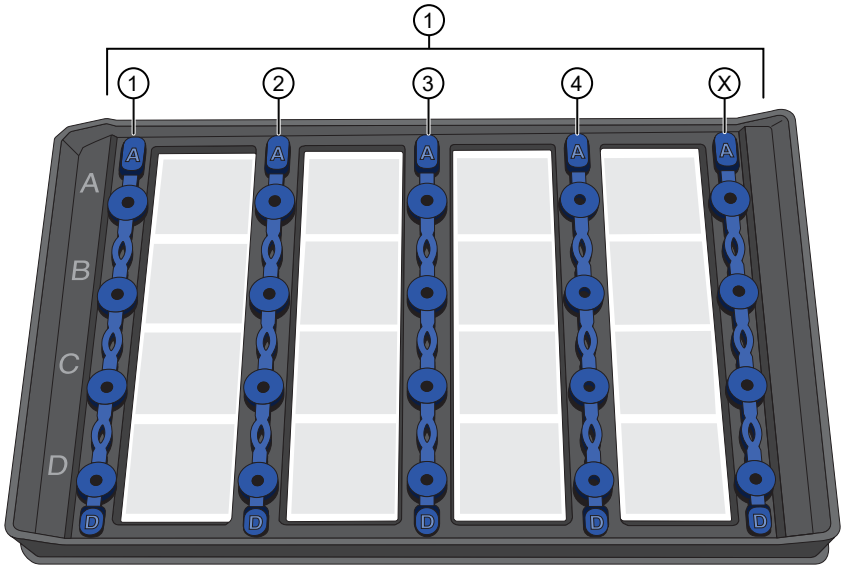


Figure 3 MAP plate with MAP plate gasket strips in place

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

The following figure shows the dimensions of a MAP plate.

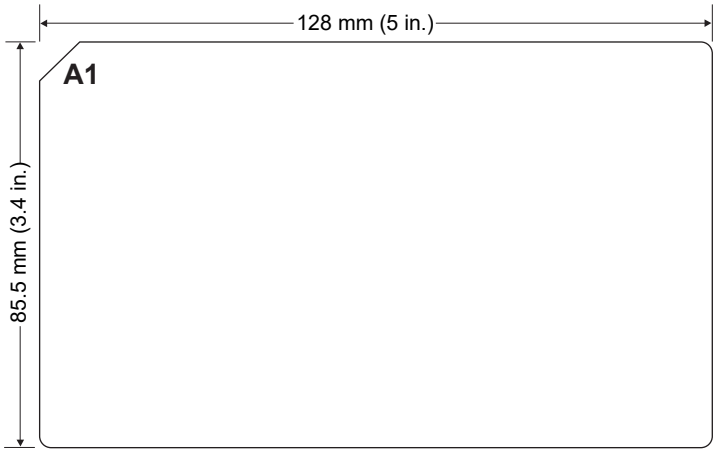


Figure 4 MAP plate dimensions

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 of the instrument.

- For best results, we strongly recommend that you use Absolute Q™ DNA Digital PCR Master Mix (5X) 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 H, “Documentation and support”).

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.

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 C, “Use the software with Security, Auditing, and E-signature (SAE) v2.2”.

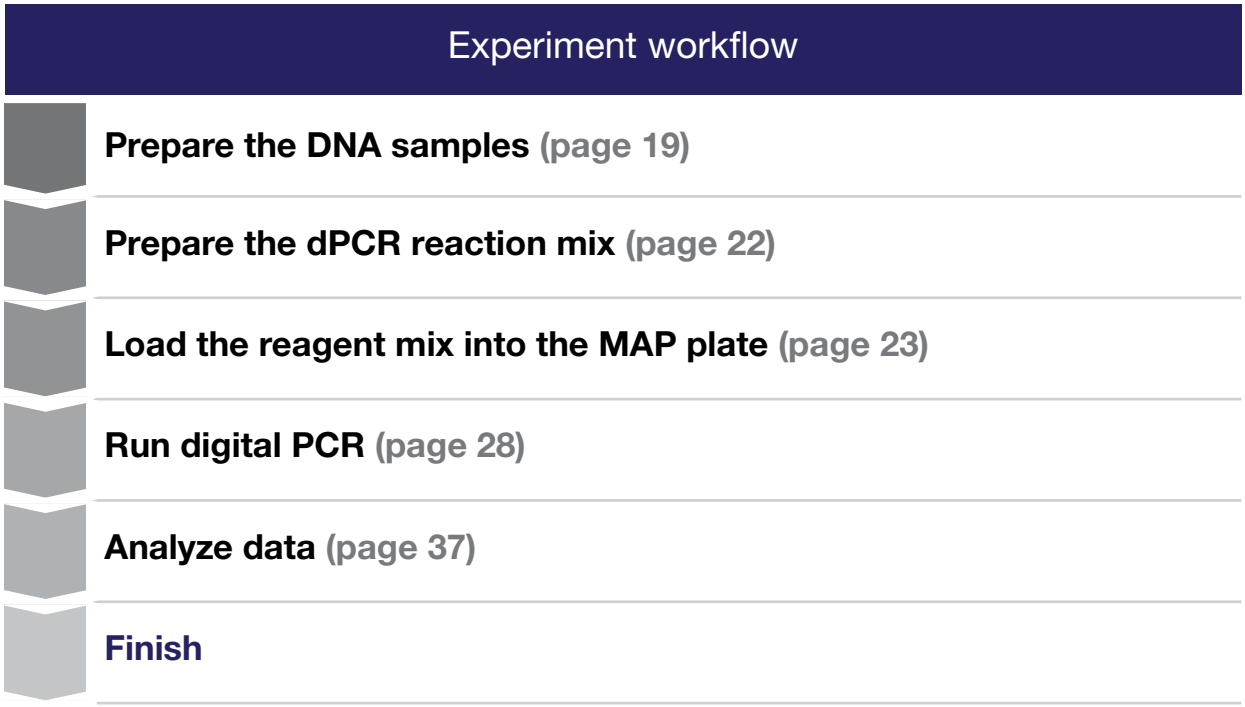
2 Prepare and run an experiment

This chapter provides a general protocol for preparing experiments using custom dPCR assays ordered on **http://www.thermofisher.com/dpcr-assays.html** or with your unique custom dPCR assays. For predesigned dPCR assays, follow the instructions in the user guide for your particular assay.

Workflow

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

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



Prepare the DNA samples

We recommend the following best practices for the preparation of DNA template (genomic DNA (gDNA) or complementary DNA (cDNA)) 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.

Nucleic acid isolation

Determine the extraction procedure that is optimal for your workflow. High-quality nucleic acids can be isolated from various sample types. Different procedures can be used for downstream dPCR analysis, depending on your workflow. Each procedure obtains nucleic acid at a different concentration, and the concentration of the nucleic acid must be within the dynamic range of the instrument.

For more information on recommended kits for nucleic acid isolation, see “Recommended materials not supplied” on page 10.

Quality of DNA

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

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

  Note: Salting-out procedures and crude lysates are not recommended.

- Does not contain PCR inhibitors
- Has an $A_{260/230}$ and $A_{260/280}$ ratio between 1.7 and 1.9

The ratio of absorbency at 260 nm and 280 nm is used to assess the purity of DNA and RNA. A ratio of ~1.8 is generally accepted as “pure” for DNA. A ratio of ~2.0 is generally accepted as “pure” for RNA. If the ratio is appreciably lower in either case, it may 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 measure of nucleic acid purity. The 260/230 values for “pure” nucleic acid are often higher than the respective 260/280 values. Expected 260/230 values are commonly in the range of 2.0–2.2. If the ratio is appreciably lower than expected, it may 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 10.

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

Sample dilution

If a target is present at a sufficiently high concentration in the sample of interest, it is possible that all reaction replicates will be positive, thereby preventing the determination of the target concentration. In this case, the sample must be diluted prior to running the dPCR experiment.

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 as described in the following sections.

- “Determine the target copy number per genome” on page 20
- “Dilute the extracted genomic DNA to the ideal input range” on page 21

Determine the target copy number per genome

This section provides examples of calculations for determining the target copy number per genome. Other calculation methods can be used. For information on 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.

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

   m = 3.5×10^{-12} gram (g) or 3.5 picogram (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.

Dilute the extracted genomic DNA to the ideal input range

Based on the known target copy number per genome, dilute the samples so that each reaction well contains approximately 0.6 to 1.6 copies of the target sequence.

Sample dilution = target copy number ÷ microchamber volume

For example, if 3.5 pg represents 1 copy of the target sequence in a 432 pL microchamber, dilute the stock human gDNA of the sample to 8.1 ng/µl (3.5÷0.432) in the final dPCR reaction.

The expected result in the dPCR is ~1 copy gene target per reaction well.

Determine the optimal dilution when the target is unknown

If the target copy number per genome is unknown (e.g., 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 ensures that one of the data points is within the optimal digital range.

The 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, baseline, threshold, etcetera.

If tested using real-time PCR, the quantification cycle (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 of 20 µL produces C_q of ~38—96-well plate
- 1 copy in total of 10 µL produces C_q of ~37—384-well plate
- 1 copy in total of 1.5 µL produces C_q of ~34.5—TaqMan™ Array Card
- 1 copy in total of 33 nL produces C_q of ~29—OpenArray™ Plate

Prepare the dPCR reaction mix

This section provides general information for using the Absolute Q™ DNA Digital PCR Master Mix (5X) 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: Other dPCR reagents are compatible with the MAP plates and can be used to create the dPCR reagent mix. For more information on specific applications, contact technical support (see Appendix H, "Documentation and support").

Gather the following materials:

- Absolute Q™ 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, perform the run within one hour of reaction preparation.

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

   Note:
   - Store reagents on ice when not in use.
   - Limit number of reagent freeze/thaw cycles.

2. Pulse vortex the Absolute Q™ 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 × *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™ 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 then 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 by performing one of the following actions:
   - Pipette mix 10–20 times, or
   - Pulse vortex 3–5 times for 1 second each.
6. Centrifuge at 1,000 × *g* for up to 1 minute.
7. Perform the run within one hour of reaction preparation.

Load the reagent 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 run at a time. Columns cannot be reused, but a MAP plate with unused columns can be used for subsequent experiments. When the experiment is complete, if the MAP plate has unused columns, place it back into its pouch for storage.

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.

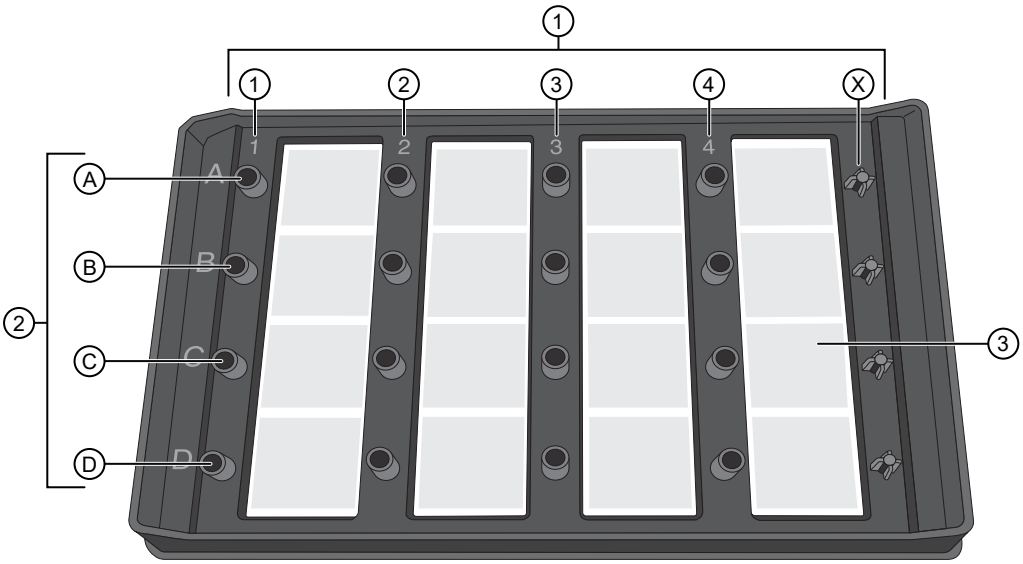


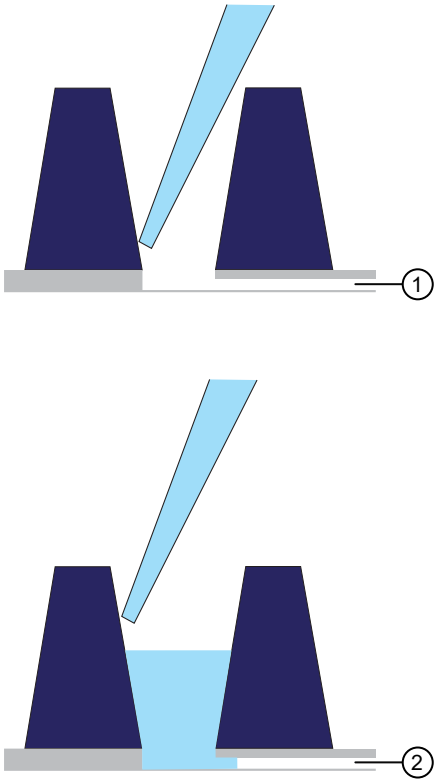
Figure 5 MAP plate without MAP plate gasket strips

(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, at a 45° angle, load 9 µL of the dPCR reagent mix to the bottom of the well. Pipette the mixture only to the first stop to prevent bubble formation.

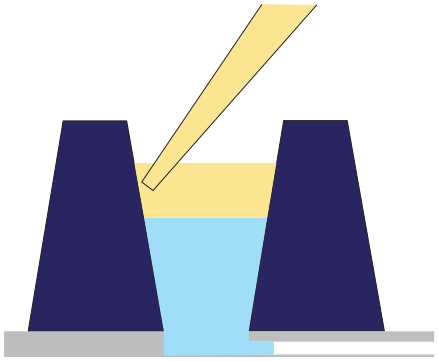
IMPORTANT! Do not contact bottom of well with the pipette tip or puncture the thin film at the bottom of the well.



① Microfluidic channel to the microreaction chamber array
② Reagent remains in the well until the instrument pushes it into the microreaction chamber 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 reagent mix. Carefully overlay the buffer on top of the reagent mix to prevent mixing or bubble formation. Pipette only to the first stop.

   The isolation buffer sits on top of the reagent, 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 on the 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.

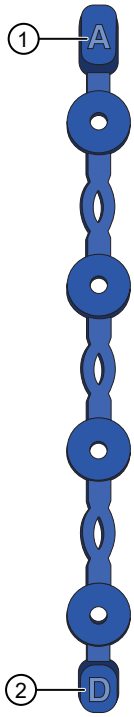


Figure 6 MAP plate gasket strip

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

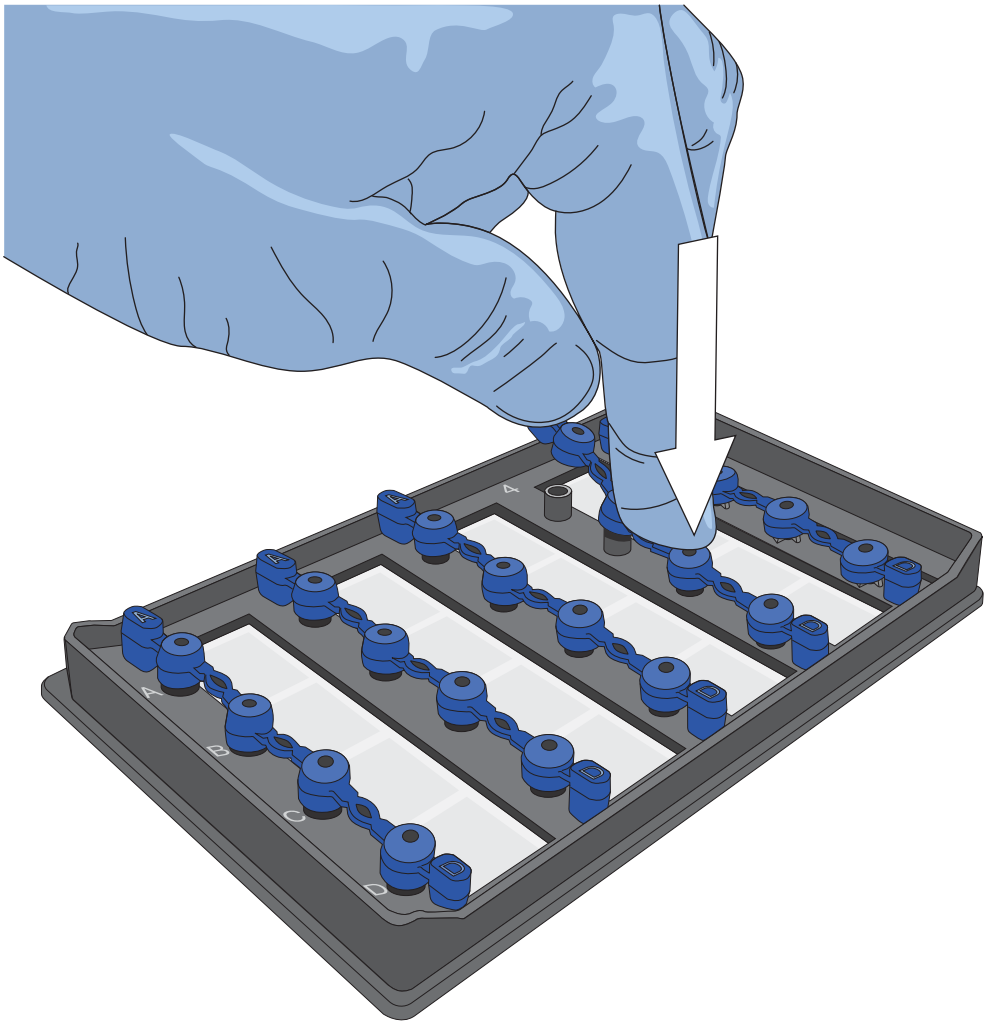


Figure 7 Press the MAP plate gasket strips firmly into place

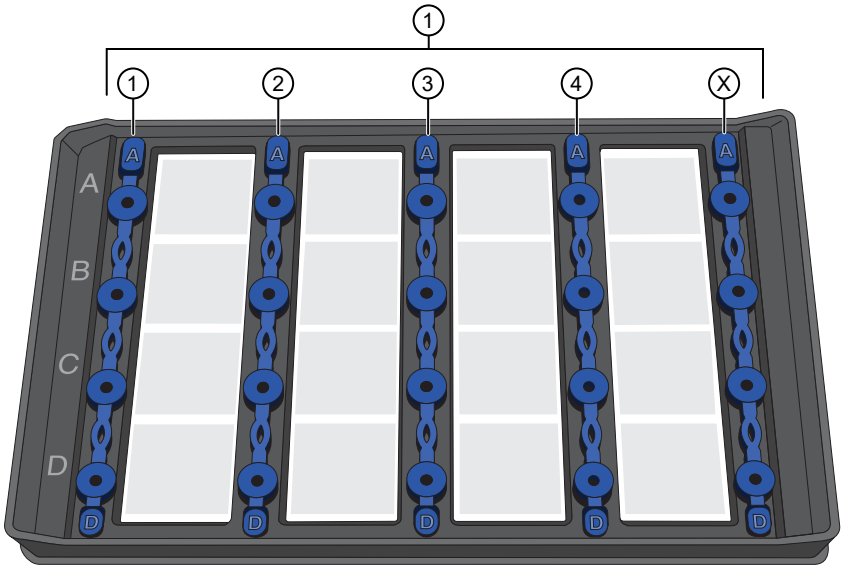


Figure 8 MAP plate with MAP plate gasket strips in place

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.

Run digital PCR

Power on the instrument and computer

IMPORTANT! Prior to powering on the QuantStudio™ Absolute Q™ Digital PCR Instrument, confirm that the shipping lock screw has been removed. Failure to do so can damage the instrument. See “Uninstall the shipping lock screw” on page 89.

1. Confirm that the power cable is connected to an appropriate power source.
2. Confirm that the USB cable is connected from the instrument to the dedicated computer.
3. Power on the dedicated computer and monitor, then start the software.
4. 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 and a ready status appears under the instrument on the **Instrument** page in the QuantStudio™ Absolute Q™ Digital PCR Software.

Create a custom protocol

The QuantStudio™ Absolute Q™ Digital PCR Software is pre-configured with the Absolute Q™ Starter (default) protocol to use as a template for creating your first custom protocol. Protocols created from this default protocol can be edited or used as templates to create additional custom protocols.

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

Note: The default protocol cannot be edited.

Protocols define the following run information:

- Dyes used in each active optical channel
- PCR parameters

1. In the left pane, click to access the **Instrument** page.
2. In the **PROTOCOL** pane, click **PROTOCOL**, then in the **Protocols** screen, select a protocol.

   Note: For your first custom protocol, select the default protocol.

3. Click to create a copy of the protocol.
   A copy of the protocol is added to the protocol list.
4. *(Optional)* Use the following options to make changes to a protocol.

Option	Action
Rename the protocol.	1. Select the protocol, then click . 2. In the name field, type a new name and press Enter.
Delete a protocol.	Select the protocol, then click .
Copy a protocol.	Select the protocol, then click .

5. Select the new protocol, then click **LOAD**.
6. In the **PROTOCOL** pane, click EDIT PROTOCOL.

7. In the **Channels** area, set the optical channels as needed.

Parameter	Action
Active optical channel	Select the check box for each optical channel to be used.
Target dye for active channel	For each active optical channel, select the drop-down to choose the target dye.

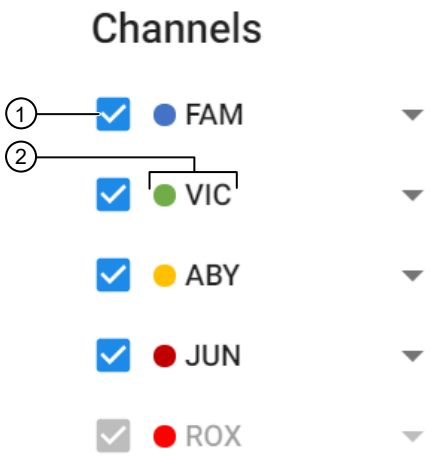


Figure 9 Optical channel dyes

① Dye channel check box
② Dye channel name

8. Modify PCR parameters as needed.

Parameter	Actions
Temperature	• Enter a value in the temperature fields. • Drag the slider bars to adjust the temperature.
Dwell times	Enter in seconds or minutes and seconds in mm:ss format.
Cycles	Set the number of cycles by entering a value into the **Cycles** field.
RNA-RT	Select **RNA-RT** to add an extra temperature step for RNA reverse transcription to cDNA for RNA samples. Not required for DNA samples.
Preheat	Select **Preheat** to add a preheat step. Sometimes called hot start, preheating the samples before PCR helps to reduce non-specific binding at lower temperatures.

(continued)

Parameter	Actions
Two or three-step cycling	Select the **Two Step** drop-down to select 2 or 3 step cycling.
Two-stage PCR cycle	Select **Two Stage PCR** to add a second PCR cycle stage.

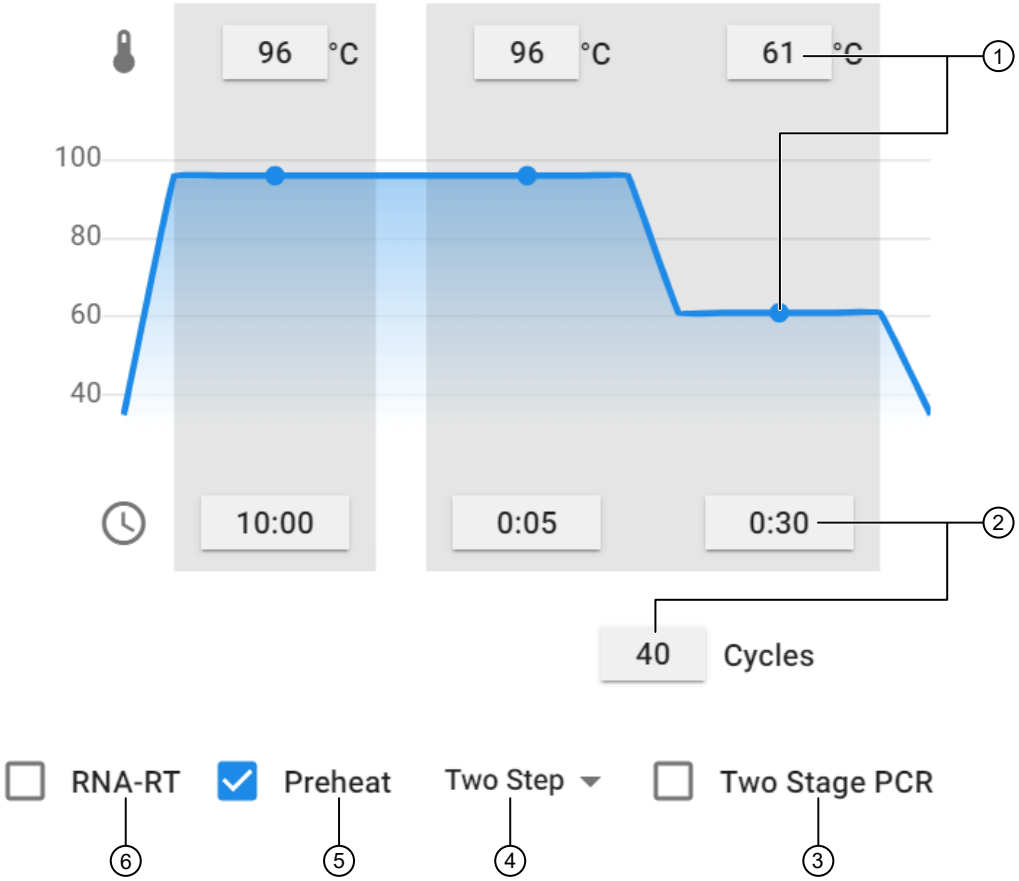


Figure 10 Protocol parameters

① Temperature settings fields and slider bar
② Time fields and cycles field
③ Two-stage PCR setting
④ Two or three step cycling option
⑤ Preheat setting
⑥ RNA-RT setting

9. Select **SAVE**.

Select a protocol for a run

1. In the left pane of the QuantStudio™ Absolute Q™ Digital PCR Software, select to access the **Instrument** page.

2. Select **PROTOCOL**, then perform one of the following tasks:

Task	Actions
Select an existing protocol.	1. Select **PROTOCOL**. 2. Select a protocol from the **Protocols** list. 3. Select **LOAD**.
Edit the loaded protocol.	1. In the **PROTOCOL** pane, click EDIT PROTOCOL. 2. Modify the optical channels and PCR parameters as needed. See Appendix A, “Modify protocols”. 3. Select **SAVE**.
Create a custom protocol	See “Create a custom protocol” on page 29.
Import a protocol.	1. Select **PROTOCOL**. 2. In the **Protocols** list area click . 3. Select **IMPORT FILE**, then navigate to the location of the protocol file. 4. Select the file, then select **Open**. 5. Select the imported protocol from the **Protocols** list. 6. Select **LOAD**.

Load the plate and run the protocol

IMPORTANT! You must clean the plate nest before each run. See “Clean the instrument and plate nest” on page 113.

IMPORTANT! Before running the protocol, make sure your protocol parameters are defined correctly. Protocol parameters cannot be changed after the run. See “Select a protocol for a run” on page 32 or Appendix A, “Modify protocols”.

1. In the left pane, click to access the **Instrument** page.

2. In the sample plate area, use the check boxes above the plate to select the columns to be used in the run.

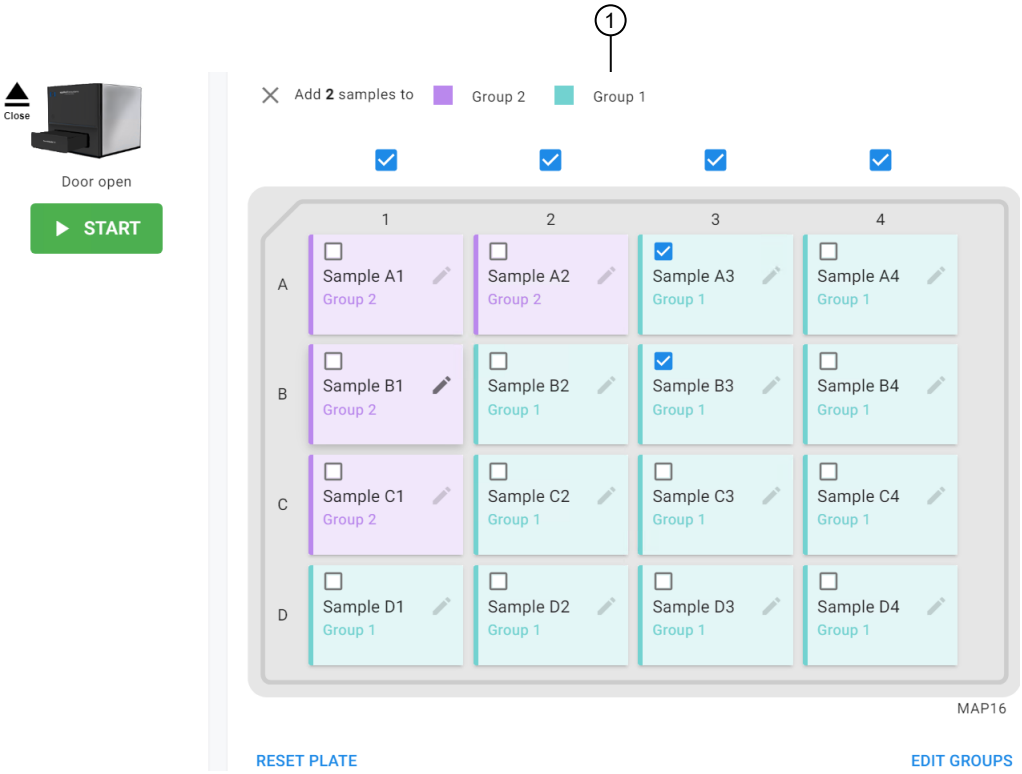
   IMPORTANT! Failure to deselect the columns that are not in use will prevent them from being used in a subsequent run.

3. Use the following options to manage samples for the run.

Option	Actions
Edit a sample name.	1. In the sample plate area, click the ✎ on the sample well, then enter a sample name. 2. Perform one of the following actions to save the sample name. • Click away from the sample name field. • Press enter to save the edit and open to the next sample name in the same column for editing.
Modify an existing group or create additional groups.	1. Below the sample plate area, select **EDIT GROUPS** to open the groups dialog box. 2. Select **NEW GROUP** in the Group list to add a new group. 3. In the **Group name** field, enter or change the name for the group 4. In the **Target DNA** fields, enter or change the name of the DNA target for each active optical channel. 5. From the **Analysis** drop-down, select the analysis type for each optical channel. 6. From the **SAMPLES** area, select one of the following sample grouping options: • **Individual** • **Replicates** • **Pooling** 7. Select **SAVE** in the groups dialog box.

4. Assign samples to a group by performing the following actions.
   a. In the sample plate area select one or more samples to be included in a group.
      Previously defined groups appear above the sample plate grid.

b. Select the group for these samples.



① Group list area

5. *(Optional)* To reset the plate to default values at any time during the plate loading process, select **RESET PLATE**.

6. *(Optional)* In the **Notes** field, enter information regarding this run, then click **ADD NOTE**.

7. Click the **START** button under the instrument icon.

   The instrument door opens to receive the loaded MAP plate.

   IMPORTANT! Confirm that gaskets are placed on all columns of the MAP plate, including unused columns. Failure to do so can produce poor results.

8. When prompted, verify that gaskets have been placed on all wells and on the column X posts on the far right as shown on the screen.

   Note: See callout 5 in the following figure for the location of column X.

9. Carefully load the MAP plate in the plate nest.

   IMPORTANT! Be sure to load the MAP plate gently to avoid damage to the plate nest.

10. Select **CLOSE DOOR**.

    The door closes and the MAP plate bar code is scanned and is displayed in the **Run name** dialog box.

    Note: If the instrument cannot scan the barcode, it can be manually added in the **Plate Barcode** field of **Run name** dialog box.

11. When prompted, enter a **Run name**.

12. Click **RUN**.

    - The run status displays in the left sidebar.
    - While processing the run, the instrument lights slowly pulse blue.
    - When the run is complete, the instrument lights are a steady blue.
    - Data populates the **ANALYSIS** tab on the **Runs** page as it becomes available.

13. When the **Run complete** dialog displays, select the run name to view the final data in the **Runs** page.

    For more information on analyzing experiment results, see Chapter 3, “Analyze data”.

Download a protocol from the Instrument page

You can save a protocol that you have created or modified to use on another computer using the QuantStudio™ Absolute Q™ Digital PCR Software by using the download option on the **Instrument** page.

1. In the left pane, click to access the **Instrument** page.

2. Select **PROTOCOL**, then select a protocol from the **Protocols** list.

3. Above the **Channels** list, click .

4. Select one of following options.
   - **Download Protocol** to download the protocol AQUA file.
   - **Download PNG** to download a graphic representation of the protocol.
5. When prompted, navigate to the location where you want to save the file, then select **Save**.

3 Analyze data

Software features

QuantStudio™ Absolute Q™ Digital PCR Software has the following pages:

- The **Runs** page lets you see experiment results organized by plate run and provides access to the **SETUP**, **ANALYSIS**, and **RESULTS** pages for analyzing and viewing experiment results. See “Runs page” on page 38.
- The **SETUP** page for a run provides controls for analysis options such as sample and target names, sample groups, replicate statistics, pooling, and copy number calculations. See “SETUP page” on page 39.
- The **ANALYSIS** page for a run displays relevant information by samples or groups and provides different options for viewing the data. See “ANALYSIS page” on page 55.
- The **RESULTS** page for a run shows a summary of the run data and provides reporting and data exporting options. See “RESULTS page” on page 73.

You can zoom in or zoom out on any page using the following keyboard shortcuts:

- ***ctrl*+** + to zoom in
- ***ctrl*+** - to zoom out

Runs page

The **Runs** page lets you see experiment results organized by plate run. Recently completed runs require a few minutes to complete the data analysis. The progress of the analysis is displayed in the **Analyzed** column. A green check mark indicates that the analysis is complete.

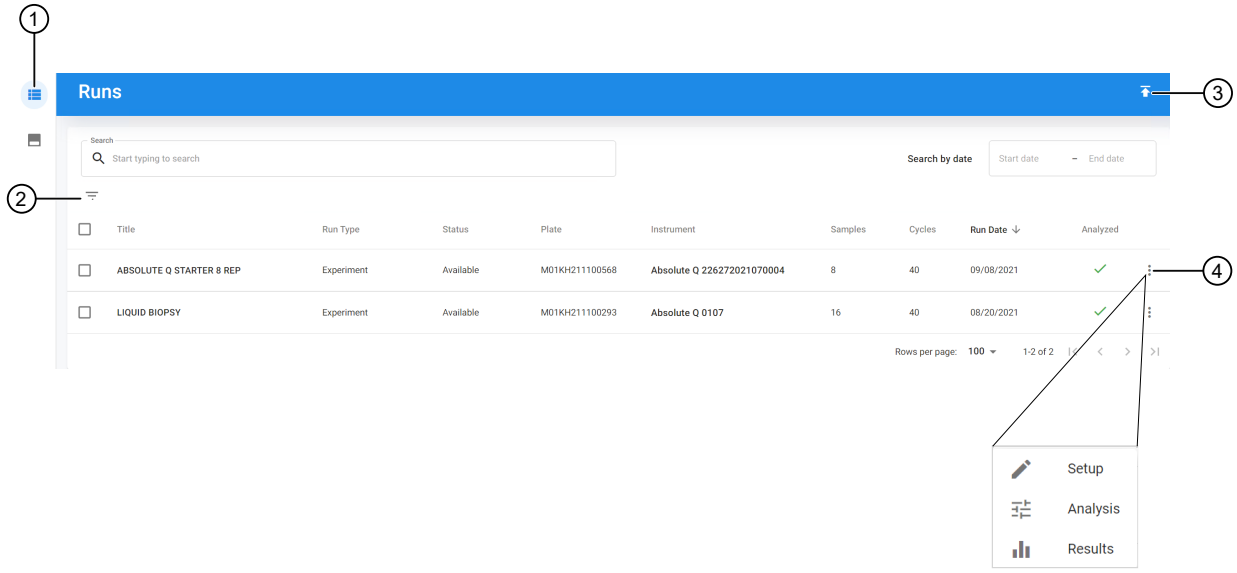


Figure 11 Runs page overview

1. Runs page
2. Filter run criteria
3. Import run
4. Run analysis shortcut menu

When a run is being analyzed, the **ANALYSIS** and **RESULTS** pages periodically update with new information.

Run data can be exported for analysis on another computer running the QuantStudio™ Absolute Q™ Digital PCR Software and imported from runs generated on a different instrument. See “Export and import runs” on page 77.

Select a run

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list.
3. *(Optional)* Use the filter option to find and select a run.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

Proceed to the **SETUP** page (see "SETUP page" on page 39), **ANALYSIS** page (see "ANALYSIS page" on page 55}, and **RESULTS** page (see "RESULTS page" on page 73) to continue with the data analysis of the run.

SETUP page

The **SETUP** page within the run lets you perform the following tasks:

- Change the run name. See "Change the run name" on page 39.
- Name the samples and set the analysis type. See "Name a sample" on page 41.
- Create and assign groups. See "Manage groups" on page 45.
- Save or load previously defined groups. See "Manage group sets" on page 50.
- Download the PCR thermal protocol or an image of the protocol. See "Download a protocol from the SETUP page" on page 43.

Sample names and group analysis can be done before or during the run in the **Instrument** page or after the run in **Runs** ▸ **SETUP** page.

Change the run name

1. In the left pane, click ⊞ to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.

3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click ⚌ located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.
4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.
5. Click ✎ next to the run name field, then enter the new name.
6. Click **SAVE**.

Sample selection and names

Sample names are user-assigned identifiers for the contents of each loaded well of a plate.

Groups determine what type of analysis is applied to all samples within a group.

Selecting a sample displays the analysis of that single sample.

Select samples

1. In the left pane, click ■ to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click ⚌ located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.

5. In the sample plate area use the following options to select samples.

Option	Actions
Select a single sample.	1. Select the sample check box. 2. Click anywhere in the sample area to select it.
Select multiple samples.	1. Click and drag through samples to select multiple samples at once.

6. Click **SAVE**.

Name a sample

1. In the left pane, click to access the **Runs** page.

2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.

3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.
5. In the sample plate area, click the ✎ on the sample well, then enter a sample name.
6. Perform one of the following actions to save the sample name.
   - Click away from the sample name field.
   - Press enter to save the edit and open to the next sample name in the same column for editing.
7. Click **SAVE**.

Hide samples

When samples are hidden, they are treated as if they were never run.

Hidden samples can be revealed. No data is lost by hiding a sample.

Hiding samples can be useful if there is an issue with the reagents, loading, or integrity of the sample.

Hiding samples has the following effect on the information that is included for display:

- No analysis results are shown for the samples.
- The samples are removed from the **RESULTS** page.
- The samples are excluded from reports.
- The samples are excluded from calculations for replicates or pooled results.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.
4. In the plot area, click EDIT ANALYSIS.
5. On the sample to be hidden, click to hide the sample.
6. To reveal a hidden sample, click on the sample.
7. Click **Save**.

Download a protocol from the SETUP page

You can save a protocol that you have created or modified to use on another computer using the QuantStudio™ Absolute Q™ Digital PCR Software by using the download option on the **SETUP** page.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the Protocol area, click ⬇ .

5. Select one of following options.
   - **Download Protocol** to download the protocol AQUA file.
   - **Download PNG** to download a graphic representation of the protocol.

6. When prompted, navigate to the location where you want to save the file, then select **Save**.

Manage groups

After a run is completed, groups are used to define the analysis and results type for reporting for individual samples or sets of samples. Once defined, a group can be edited or deleted.

Note: Only groups without samples can be deleted.

When samples are assigned to a group, they will all have the same definition for the following characteristics of the sample:

- The target DNA associated with each fluorescent dye.
- The analysis type for each optical channel:
  - CNV (Copy Number Variation)—Reporting ratio of CNV/CNV Ref
  - 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.
  - Signal—Absolute quantification
  - Not Used—Ignored in analysis
- Grouping options:
  - Individual—Each sample has a separate result entry.
  - Replicates—The results show the Mean, Standard Deviation, and the CV% of the concentration for all the samples in the group.
  - Pooling—The results treat all of the samples in the group as one large sample.

See the following sections for more information:

- To create groups, see “Create groups” on page 45.
- To edit groups, see “Edit groups” on page 47.
- To delete groups, see “Delete groups” on page 48.
- To add samples to groups, see “Assign samples to groups” on page 49.
- To save group sets, see “Save group sets” on page 50.
- To load group sets, see “Load group sets” on page 51.
- To edit group set names, see “Edit group set names” on page 52.
- To delete group sets, see “Delete group sets” on page 53.

Create groups

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.
5. Below the sample plate area, select **EDIT GROUPS** to open the groups dialog box.
6. Select **NEW GROUP** in the Group list.
7. In the **Group name** field, enter a name for the group.
8. In the **Target DNA** fields, enter the name of the DNA target for each active optical channel.
9. From the **Analysis** drop-down, select the analysis type for each optical channel.
10. From the **SAMPLES** area, select one of the following sample grouping options:
    - **Individual**
    - **Replicates**
    - **Pooling**
11. Select **SAVE** in the groups dialog box.
12. Select **SAVE** on the **SETUP** page.

Edit groups

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.
4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.
5. Below the sample plate area, select **EDIT GROUPS** to open the groups dialog box.
6. Select the group in the **Name** list.
7. Edit group settings as needed.
8. Select **SAVE** in the groups dialog box.
9. Select **SAVE** on the **SETUP** page.

Delete groups

Only groups that do not contain samples can be deleted.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.
4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.
5. Below the sample plate area, select **EDIT GROUPS** to open the groups dialog box.
6. Select the group in the **Name** list.
7. Click .
8. Select **SAVE** in the groups dialog box.
9. Select **SAVE** on the **SETUP** page.

Assign samples to groups

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

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.
4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.
5. In the sample plate area select one or more samples to be included in a group.
   Pre-defined groups appear above the sample plate grid.
6. Select the group for these samples.
7. Select **SAVE**.

Manage group sets

All the groups defined for a plate can be saved as a named group set.

Saved group sets can be loaded into other runs.

Each group set contains the following information:

- All the groups created in a run, even if they are not applied to a sample
- The location of the samples applied to each group
- The name and color of each group
- The target DNA names in each group
- The analysis type for each target
- The analysis for samples as individual, replicates, or pooling

Group sets do not include the sample names and dye names which will be unaffected by loading a group set.

The QC channel in a run is never changed by loading a group set. For information on the QC channel, see “View by samples” on page 58.

To save a group set, see “Save group sets” on page 50.

To load a group set, see “Load group sets” on page 51.

To edit a group set name, see “Edit group set names” on page 52.

To delete a group set, see“Delete group sets” on page 53.

Save group sets

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the upper-right corner of the sample plate area, click EDIT SETUP to enable editing.

5. From the sample plate grid, select the groups to be included in the Group Set.

6. In the upper-right corner of the page, click .

7. In the **Save Group Set** dialog, enter a name for the Group Set, then select **SAVE**.

8. Select **SAVE** on the **SETUP** page.

Load group sets

When a group set is loaded, the following items apply:

- All existing groups in the run are replaced by the groups in the set and applied to the samples as defined by the group set.
- The targets in each group are assigned to the closest channel matching the run that the group set originated from.
- When a group set containing only a single column is applied to a multi-column run, the group pattern is repeated for each column.

1. In the left pane, click to access the **Runs** page.

2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.

3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click filter located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.
4. In the upper-right corner of the page, click
5. In the **Load Group Set** dialog, choose a group set from the list, then select **LOAD**.

Edit group set names

When a group set is loaded:

- All existing groups in the run are replaced by the groups in the set and applied to the samples as defined by the group set.
- The targets in each group are assigned to the closest channel matching the run that the group set originated from.
- When a group set containing only a single column is applied to a multi-column run, the group pattern is repeated for each column.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click filter located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the upper-right corner of the page, click

5. In the **Load Group Set** dialog, select a group set, then .

6. Edit the group set name, then select .

Delete group sets

When a group set is loaded:

- All existing groups in the run are replaced by the groups in the set and applied to the samples as defined by the group set.
- The targets in each group are assigned to the closest channel matching the run that the group set originated from.
- When a group set containing only a single column is applied to a multi-column run, the group pattern is repeated for each column.

1. In the left pane, click to access the **Runs** page.

2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.

3. *(Optional)* Use the filter option to find and select a run, then select the **SETUP** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the upper-right corner of the page, click

5. In the **Load Group Set** dialog, choose a group set from the list, then select .

ANALYSIS page

The **ANALYSIS** page displays relevant information by samples or groups and provides different options for viewing the data.

Figure 12 ANALYSIS page overview

1. View by samples
2. View by groups
3. View QC channel
4. Export sample data
5. View data by optical channel
6. Select **EDIT ANALYSIS** to edit plot information
7. Toggle **Show Rejects** to show microreaction chambers automatically rejected from analysis and results
8. Toggle to display array view
9. Toggle between **1D Scatter plot**, **Histogram**, and **2D Scatter plot** views

Viewing by samples puts the color channel information into rows for comparison across the color channels. You can view or download data plots for each sample. See “View by samples” on page 58.

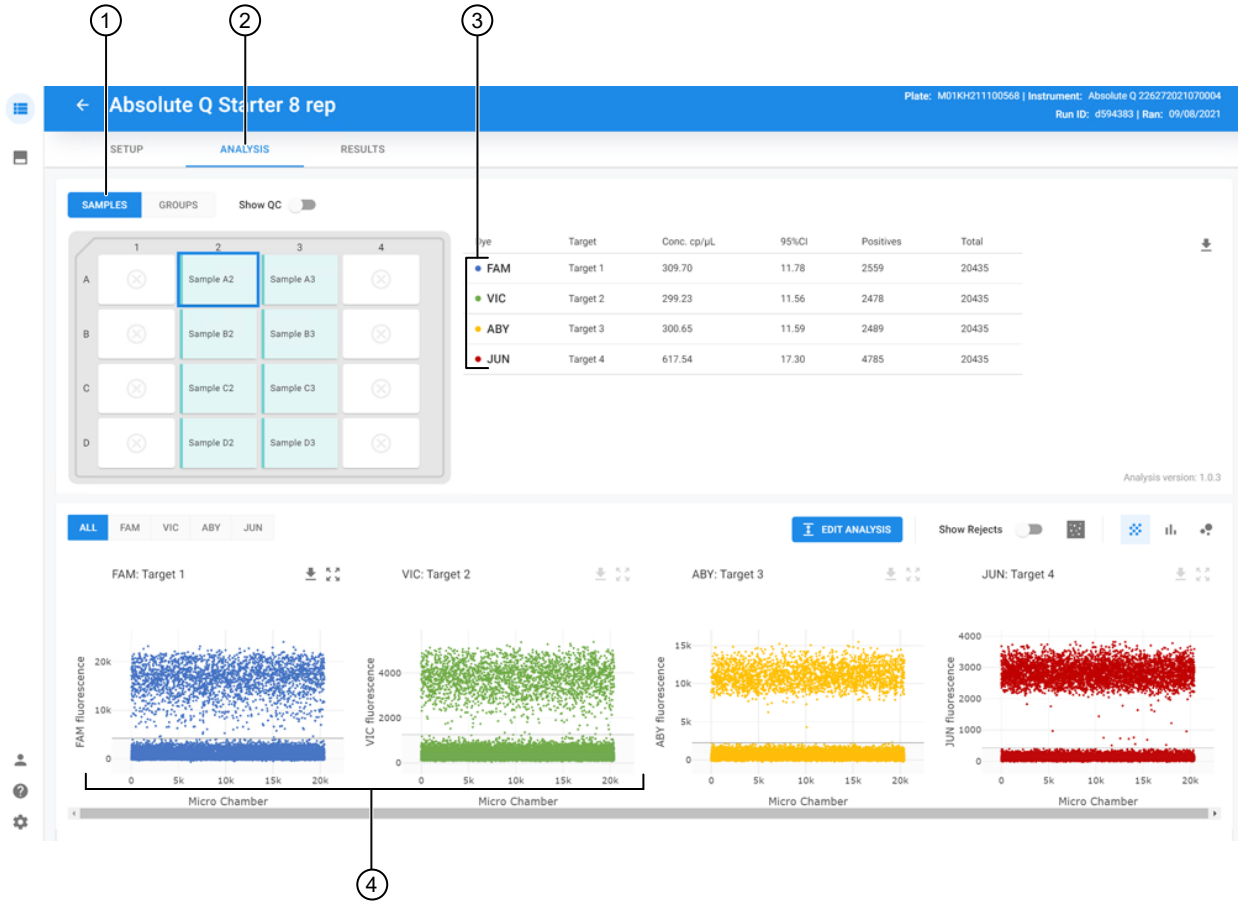


Figure 13 View by samples

① View by samples
② ANALYSIS page
③ Color channels for the selected sample
④ 1D Scatter plots with positive and negative threshold

QC channel data is provided for each sample.

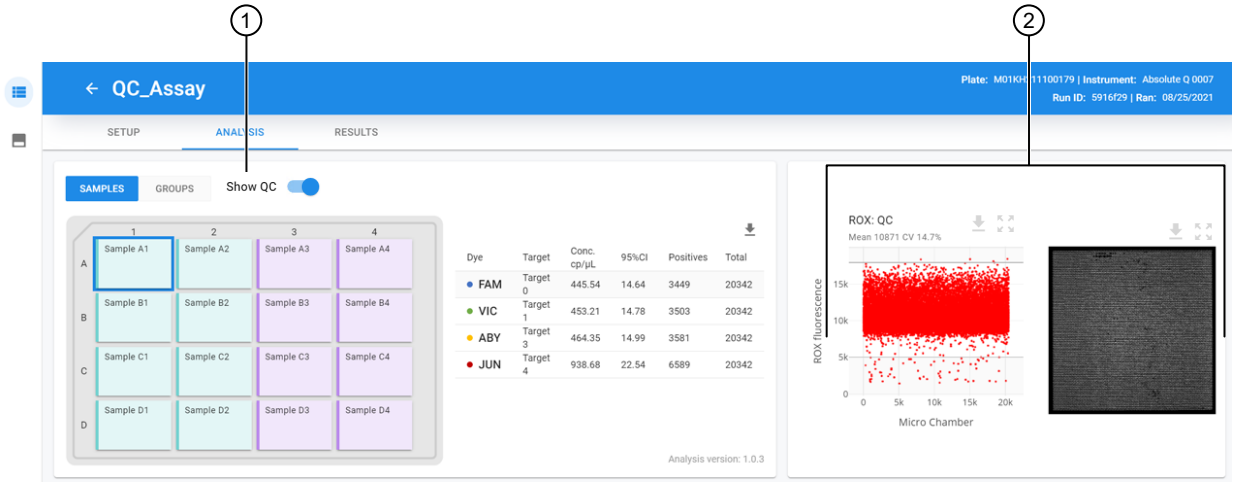


Figure 14 View QC channel data for a sample

① QC channel toggle
② QC channel window

Viewing by **GROUPS** lets you compare color channels across the sample. You can view or download data plots for each group. See “View by groups” on page 60.

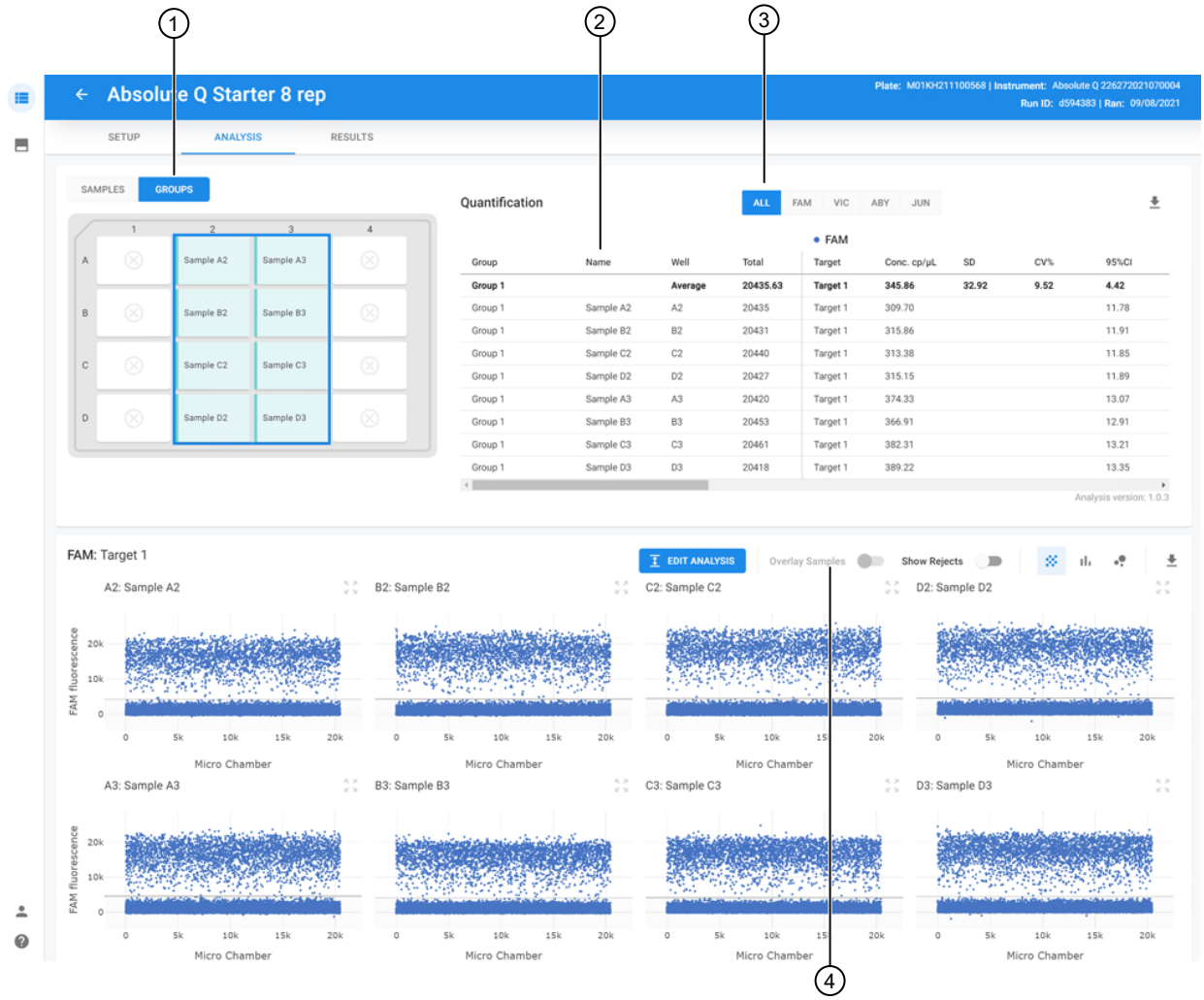


Figure 15 View by groups

① View by groups
② Row for each sample in the selected group
③ Color channel selection
④ Toggle **Overlay Sample** to see all data from a sample target within a group in single plot

View by samples

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. On the **ANALYSIS** page, select **SAMPLES**.
   The information is displayed in the results area using the 1D Scatter plot type.

5. *(Optional)* Use these options to change the results view:

Option	Description
	1D Scatter plot type
	Show Rejects
	Histogram plot type
	2D Scatter plot type
	2D Scatter plot type with heatmap
	Show Arrays

   For information about plots, see “Plot types” on page 62 and “View plots” on page 63.
   For information about showing arrays, see “Show array images” on page 70.

6. *(Optional)* Toggle the QC window by selecting the **QC** toggle.
   The QC window displays the plot and array image for the QC channel (usually ROX™).This quality control data ensures that only properly filled microreaction chambers are used for analysis by evaluating the ROX™ signal for each microreaction chamber.

The top and bottom thresholds are automatically set but can be manually adjusted. The QC plot should have a single level band indicating uniform filling.

7. *(Optional)* Manually adjust the threshold using the EDIT ANALYSIS function.
   For more information about adjusting thresholds, see "Set thresholds" on page 66.

View by groups

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.
4. On the **ANALYSIS** page, select **GROUPS**.
   The information is displayed in the results area using the 1D Scatter plot type.

5. *(Optional)* Use these options to change the results view.

Option	Description
	1D Scatter plot type
	Show Rejects
	Histogram plot type
	2D Scatter plot type
	2D Scatter plot type with heatmap
	Show Arrays

For more information on plots, see “Plot types” on page 62 and “View plots” on page 63.
For more information on showing arrays, see “Show array images” on page 70.

6. *(Optional)* Manually adjust the threshold using the 

function.
For more information on adjusting thresholds, see “Set thresholds” on page 66.

7. *(Optional)* Overlay sample targets with common thresholds within a group, see “Overlay samples” on page 68.

Plot types

There are three plot options:

Option	Description
	1D scatter (signal versus index)
	Histogram
Heatmap	2D Scatter, with optional heatmap (color versus color) — use the Heatmap to display scatter plots as a heatmap. Brighter areas indicate a higher density of plots.

Show Rejects (Show Rejects): display or hide microreaction chambers that have been rejected from the analysis results. Showing rejects does not impact the analysis or results calculations.

Figure 16 View 1D Scatter plot

① EDIT ANALYSIS option
② Show Rejects option
③ 1D Scatter option

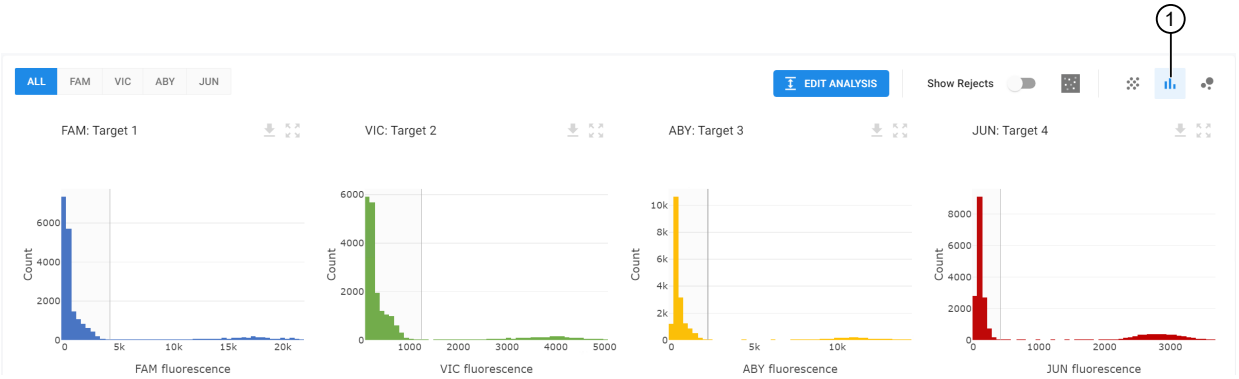


Figure 17 View Histogram plot

① Histogram plot option

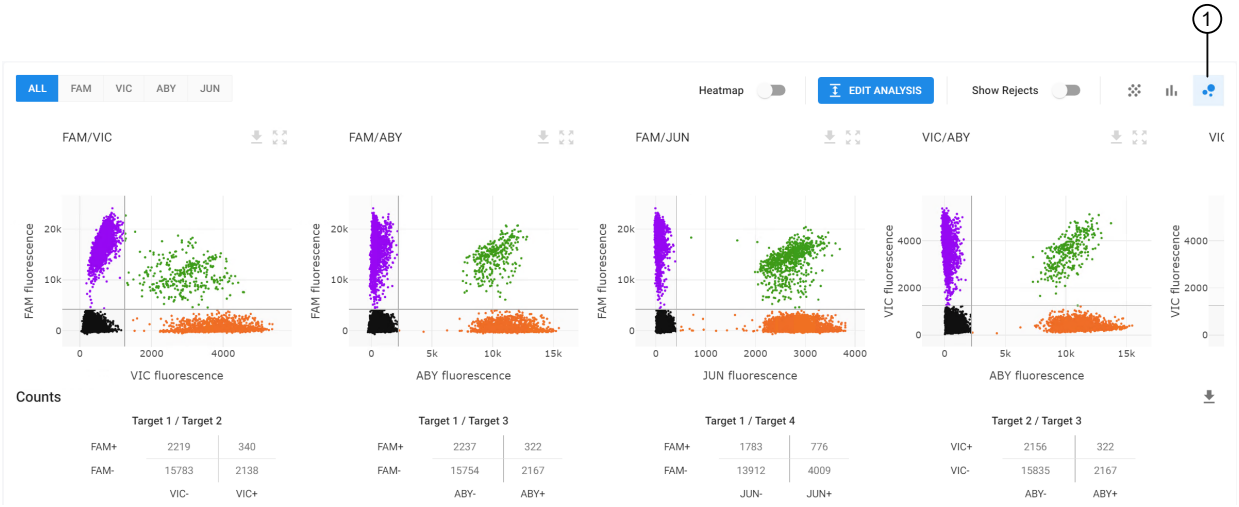


Figure 18 View 2D Scatter plot

① 2D Scatter plot option

View plots

View plot information by zooming in on an area of interest or enlarging the plot.

For information on adjusting thresholds, see “Set thresholds” on page 66.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. Use the following options to view a plot.

Option	Actions
Filter plot area by channel.	By default all channels are displayed in the plot area. • To view to a specific channel, click on the channel name in the upper-left corner of the plot area.
Zoom in on an area of a plot.	1. Hover over a plot, then click and drag the cursor on the area of interest. 2. *(Optional)* To download the selected area of the plot as a PNG image or download data from the plot, click ⬇. 3. *(Optional)* To return the plot to the original scale, click 🔍.
Enlarge a plot.	1. Hover over a plot, then click ⛶. 2. Use the left and right arrows to navigate between the channels. 3. *(Optional)* Hover over the enlarged plot, then click and drag the cursor on an area of interest. 4. *(Optional)* To download the plot as a PNG image or download data from the plot, click ⬇. 5. *(Optional)* To return the plot to the original scale, click 🔍.

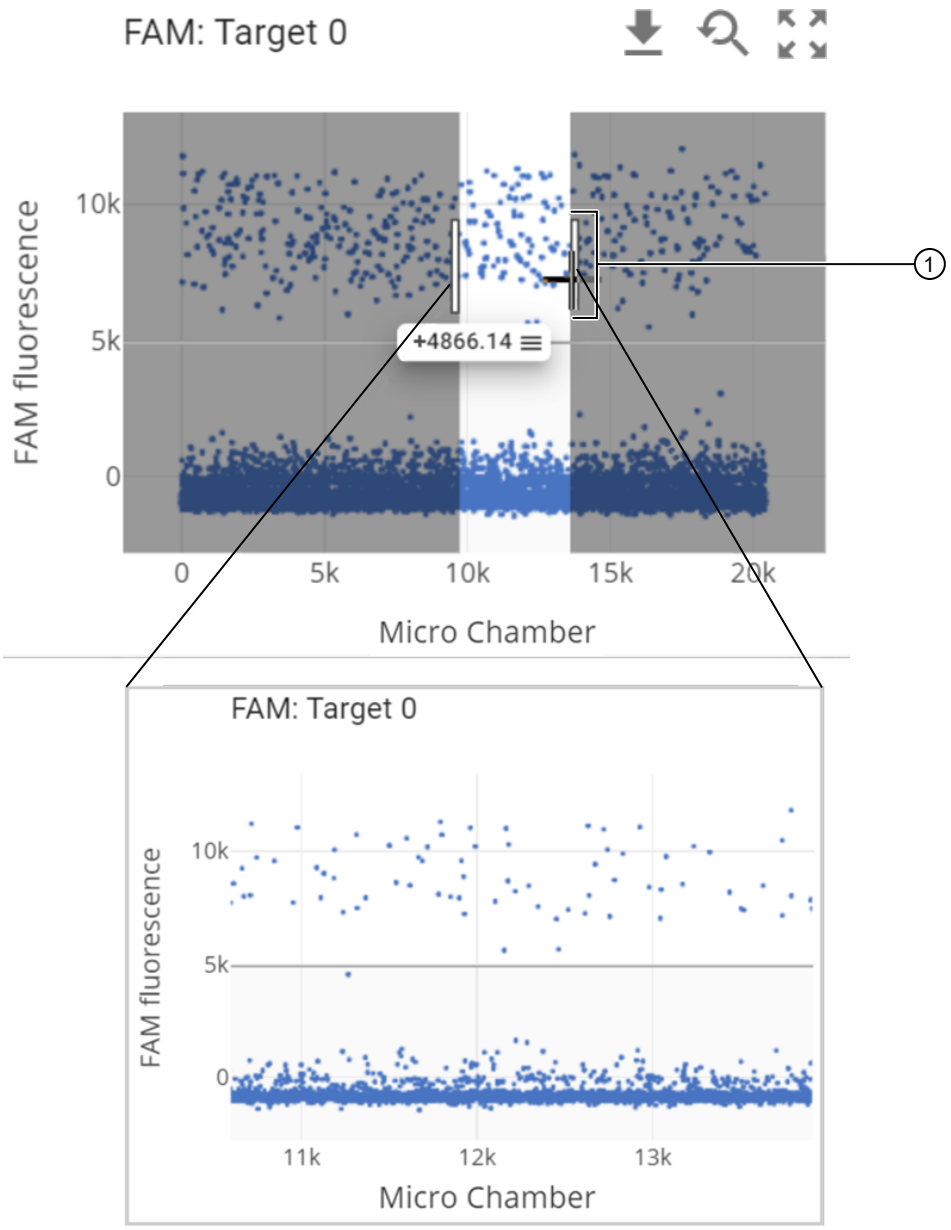


Figure 19 Zoom in on a section of a plot

① Click and drag to zoom in on a selection

Set thresholds

Thresholds on all plots are automatically set during the initial analysis based on the data distribution.

Automatic (default) thresholds have a single grey line indicating the barrier between positive and negative data points.

Thresholds that have been adjusted have a single blue line indicating the barrier between positive and negative data points.

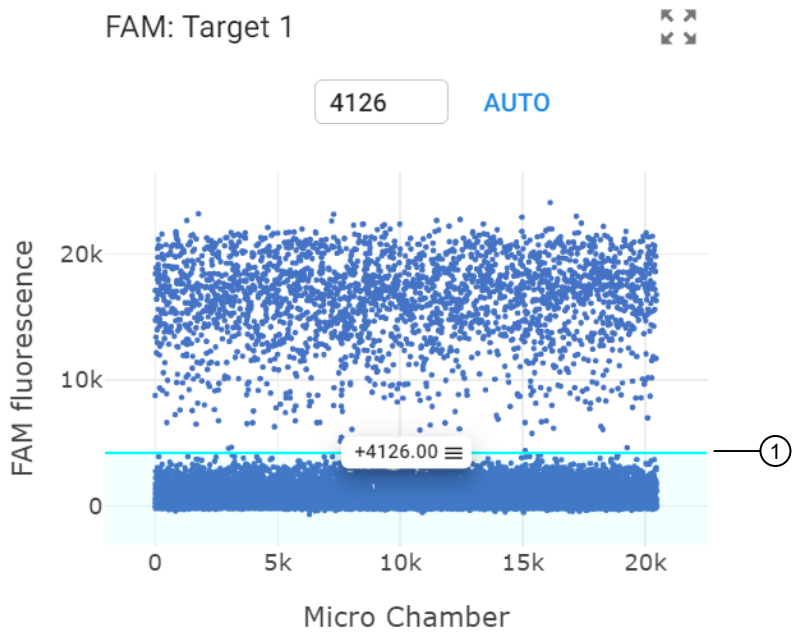


Figure 20 Adjust thresholds

① Blue line indicates threshold has been adjusted.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. To enable editing in the plot area, click EDIT ANALYSIS.
The following options are available in the plot area.

Option	Description
EDIT ANALYSIS	Enable editing of the **Analysis** page.
SAVE	Save changes and exit edit mode.
CANCEL	Discard all changes and exit edit mode.
	Autoscale to return the plot to return the original scale.
AUTO	Auto-threshold to return the plot to the original threshold.
	Enlarge the plot.

5. *(Optional)* Filter the plot area by channel by selecting the channel name in the upper-left corner of the plot area.

6. Adjust the thresholds on a plot by using one of the following options.

Option	Action
Manually enter the threshold values.	Adjust the threshold hold number using the following actions. • Click in the threshold value field above the plot, then enter the new value. • Hover over the threshold value field above the plot, then click the up or down arrows to adjust the value.
Drag the threshold bar.	1. Hover over the threshold line until the threshold value appears. 2. Click on the threshold value, then drag the threshold bar up or down to adjust the value. **Note:** Clicking on the threshold line (not the value) results in a zoom action.

Note: For samples that have been grouped, adjusting a channel threshold changes the threshold value for all samples in the group to the same value.

7. *(Optional)* To revert to the original threshold value automatically calculated by the system, click **AUTO**.

Note: Auto-threshold reverts all the samples that have had thresholds adjusted back to their original system-generated auto-threshold values. Samples in groups that have not been adjusted will not be changed.

8. *(Optional)* To discard all changes and exit the edit mode, click **CANCEL**.

9. To save the changes and exit the edit mode, click **SAVE**.
The system recalculates the thresholds and refreshes the page to display the adjusted thresholds.

Overlay samples

Using the **GROUPS** view on the **ANALYSIS** page, you can overlay sample plots when you want to see all the data from a sample target within a group in a single plot. For example, sample overlay in the 2D plots allows for clustering with a larger microreaction chamber population so it can help provide more confidence in the clustering results (especially when some samples have low positive count) and allows you to apply a common threshold to the entire group at once.

Note: All analysis editing and plot view features are available for use with overlayed samples.

To overlay samples within a target, all samples within that target in the sample group must have a common threshold. For information on adjusting thresholds, see “Set thresholds” on page 66.

1. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.

2. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click ⚌ located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

3. Select **GROUPS**, then in the sample plate area, select the group of interest.

Note: If the Overlay Samples toggle is not available, the sample targets in the selected group do not have common thresholds. To adjust the thresholds, see “Set thresholds” on page 66.

4. In the plot area, select Overlay Samples.

The page refreshes to display the overlayed plots for each sample channel.

Show array images

The **Show Arrays** ( ) option toggles the display of array images for all channels of a sample.

Microreaction chambers with positive data points are colored. Microreaction chambers with negative and rejected data points are gray scale. Often they have a low signal and appear black.

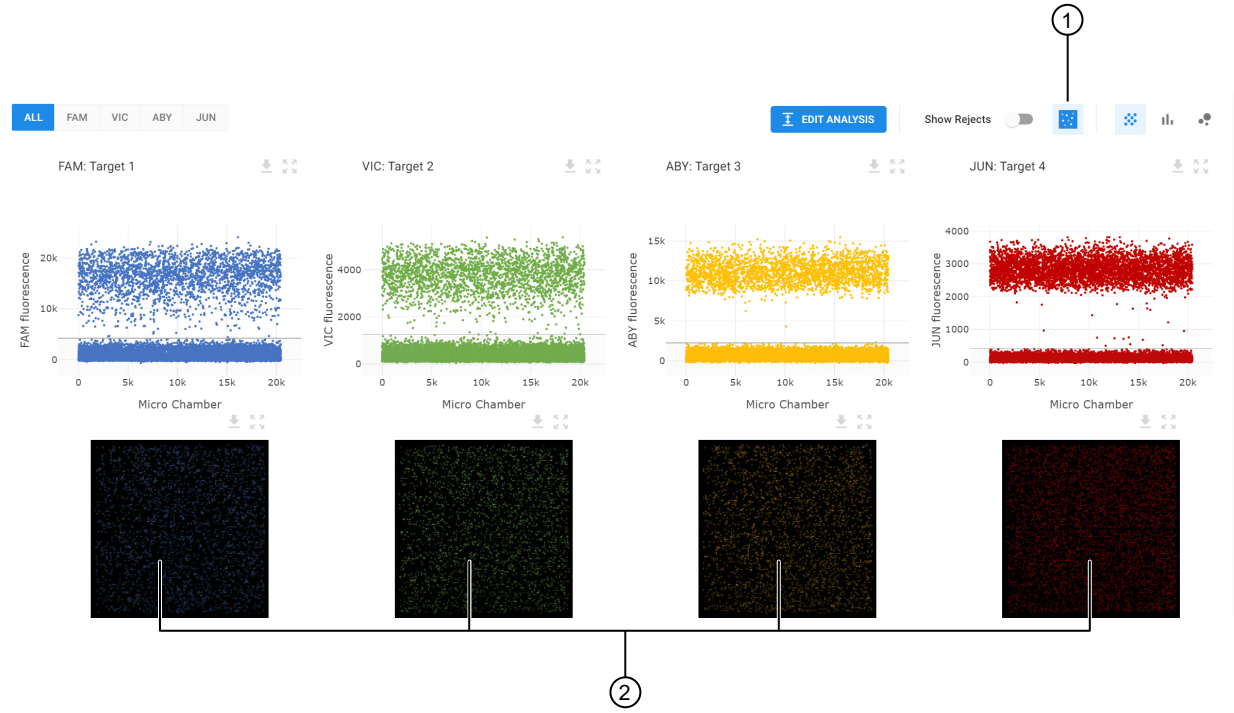


Figure 21 View channel array images

① Show arrays option
② Channel array images

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. On the **ANALYSIS** page, select a sample, then click .

5. Select an array to view an enlarged image.

6. Use the left and right arrows to navigate between arrays.

Export data from the ANALYSIS page

1. In the left pane, click to access the **Runs** page.

2. Use the search fields to find a run or select a run from the list, then select the **ANALYSIS** page.

3. *(Optional)* Use the filter option to find and select a run, then select the **ANALYSIS** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. To export data, perform one of the following tasks:

Option	Actions
Export table data.	1. From the **ANALYSIS** page, in the optical channel table area, click ⬇. 2. Select an export option. • **Copy to Clipboard** — copies the data in HTML format • **Download CSV** — downloads the data in CSV format 3. When prompted, navigate to the location to save the file and select **Save**.
Export plot data.	1. From the **ANALYSIS** page, in the plots area, hover over a plot and click ⬇. 2. Select an export option. • **Download Plot** • **Download Data** 3. When prompted, navigate to the location to save the file and select **Save**.

RESULTS page

The **RESULTS** page displays the results for all the samples in a single table. The values are plotted together below the concentration table.

From the **RESULTS** page, you can:

- View statistical results from the run. See “View results” on page 73.
- View results plots from the run by sample or group. See “View results” on page 73.
- Copy the data to the clipboard in HTML format. See “Export data from the RESULTS page” on page 75.
- Download the data table in CSV format. See “Export data from the RESULTS page” on page 75.
- Generate data reports. See “Generate reports” on page 76.

The presentation of the **RESULTS** page is based on the groups assigned in the **SETUP** page.

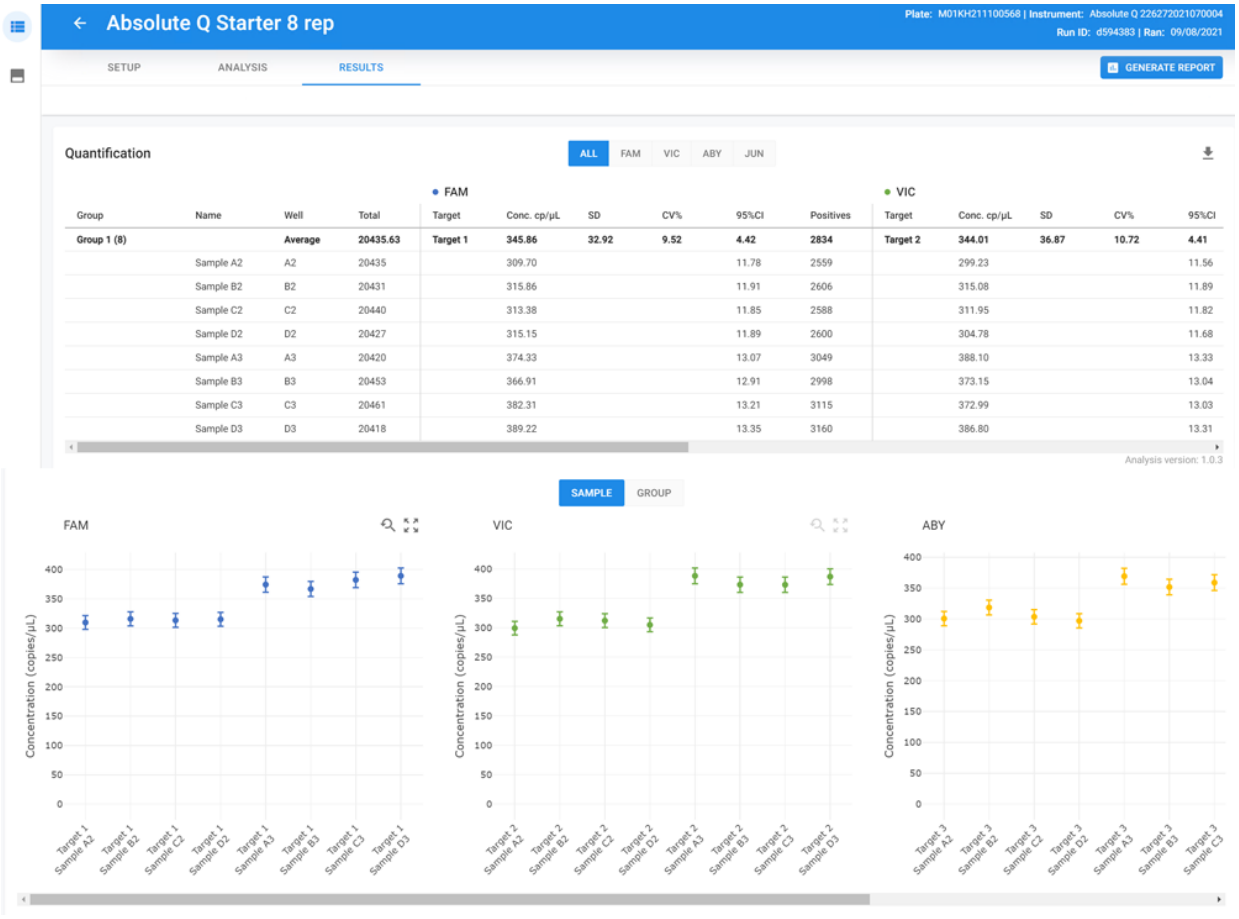


Figure 22 View Results

View results

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **RESULTS** page.

3. *(Optional)* Use the filter option to find and select a run, then select the **RESULTS** page.
   a. Click ⚌ located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. To view results, perform one of the following tasks:

Option	Actions
View results for all optical channels.	1. From the list of active optical channels above the concentration table, select **ALL** to view all channels. 2. Use the scroll bar at the bottom of the concentration table to scroll through concentration table data for all channels. 3. Use the scroll bar on the side of the page to scroll down to see the plot data for all channels. 4. Toggle between **SAMPLE** and **GROUP** to change the display of the plot data. 5. Use the scroll bar at the bottom of the plot area to scroll through the plot data for all channels.
View results for a specific optical channel.	1. From the list of active optical channels above the concentration table, select the desired channel. 2. Use the scroll bar on the side of the page to scroll down to see the concentration table data for the channel. 3. Use the scroll bar on the side of the page to scroll down to see the plot data for the channel. 4. Toggle between **SAMPLE** and **GROUP** to change the display of the plot data.

Export data from the RESULTS page

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **RESULTS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **RESULTS** page.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.

4. To export data, perform one of the following tasks.

Option	Actions
Export concentration data.	1. From the **RESULTS** page, in the concentration table area, click ⬇. 2. Select an export option. • **Copy to Clipboard** — copies the data in HTML format. • **Download CSV** — downloads the table data shown in CSV format. • **Multi-channel CSV** — downloads table data not seen in the **RESULTS** page that provides concentration data on target combinations in CSV format. 3. When prompted, navigate to the location to save the file and select **Save**.
Export plot data.	1. From the **RESULTS** page, in the plots area, hover over a plot and click ⬇. 2. Select an export option. • **Download Plot** — downloads plot data as a PNG file. • **Download Data** — downloads the plot data in CSV format. 3. When prompted, navigate to the location to save the file and select **Save**.

Generate reports

The **GENERATE REPORT** option uses the Report Builder to create and export reports as PDF files.

Note: Hidden samples are excluded from reports. You must unhide any samples that you need included in a report. To unhide a sample, see “Hide samples” on page 42.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find a run or select a run from the list, then select the **RESULTS** page.
3. *(Optional)* Use the filter option to find and select a run, then select the **RESULTS** page.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. From the **RESULTS** page, select **GENERATE REPORT** to open **Report Builder**.
   The **Report Builder** dialog box opens and **All Groups** are selected by default to be included in the report.

5. *(Optional)* Select **Select Groups**, then from the list of groups, select the check box next to each group to be included in the report. Any combination of groups can be selected.

6. *(Optional)* Select **QC Channel** to include the QC channel data for all samples.

7. Select **BUILD**.

Export and import runs

Runs can be exported to a ZST file that can be transferred and imported into QuantStudio™ Absolute Q™ Digital PCR Software running on a different computer.

The ZST run file contains all analysis options:

- Group details
- Sample names
- Thresholds

Run files are approximately 30 MB in size.

To export a run, see “Export a run” on page 78.

To import a run, see “Import a run” on page 79.

Export a run

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find the runs for export.
3. *(Optional)* Use the filter option to find and select runs for export.
   a. Click located above the run list.
   b. Select one or many filters from the following list of filter options.

   Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

   c. Click away from the filter list to view the run list based on your filter selections.
   d. *(Optional)* Click the on the filter name to remove a filter selection from the list of runs.
4. In the list of runs, select the check boxes of the runs to be included in the export.
5. In the upper-right side of the run list, click .
6. When prompted, enter a name for the export file and navigate to the locate where you want to save the run, then select **Save**.

Import a run

1. In the left pane, click to access the **Runs** page.
2. In the upper-right corner of the **Runs** page click .
3. Perform one of the following options:

Option	Action
Drag the ZST file into the **Import your result file** window from Windows™ File Explorer.	1. Using Windows™ File Explorer, navigate to the location of the ZST file to import. 2. Drag and drop the file into the **Import your result file** window.
Navigate to the ZST file from the **Import your result file** window.	1. Select **IMPORT FILE** and navigate to the location of the ZST file to import. 2. Select the file, then select **Open**.

Delete a run

IMPORTANT! Deleting a run is permanent. You cannot restore a deleted run.

1. In the left pane, click to access the **Runs** page.
2. Use the search fields to find the runs to be deleted.
3. (*Optional)* Use the filter option to find and select runs to be deleted.
   a. Click located above the run list.

b. Select one or many filters from the following list of filter options.

Note: If no filters are selected, all runs are displayed.

Filter	Option
Run Type	• **Experiment**—A typical user-generated run. • **System Calibration**—A run performed by a Thermo Fisher representative to calibrate the instrument.
Status	• **Active**—A dye calibration run currently being used as the calibration for the attached instrument. • **Available**—An experiment run available in the software. • **Obsolete**—A dye calibration run not currently being used as the calibration for the attached instrument. • **Unknown**—The status of a run cannot be determined because the instrument is not currently attached, or does not contain the requisite data.
Instrument	Instruments associated with the run data either generated in or imported into the system.

c. Click away from the filter list to view the run list based on your filter selections.

d. *(Optional)* Click the ⊗ on the filter name to remove a filter selection from the list of runs.

4. In the list of runs, select the check boxes of the runs to be deleted.

5. In the upper-right side of the run list, click 🗑.

6. When prompted to delete the run, select **DELETE**.

A

Modify protocols

Existing protocols can be edited or used as templates to create additional custom protocols.

Protocols define the following run information:

- Dyes used in each active optical channel
- PCR parameters

1. In the left pane, click to access the **Instrument** page.
2. Use one of the following options to select a protocol.

Option	Action
Select the loaded protocol.	In the **PROTOCOL** pane, click EDIT PROTOCOL to modify the loaded protocol.
Select a protocol from the list of available protocols.	1. In the **PROTOCOL** pane, click **PROTOCOL**. 2. In the **Protocols** screen, select a protocol, and click **LOAD**. 3. In the **PROTOCOL** pane, click EDIT PROTOCOL to modify the loaded protocol.

3. Modify optical channels as needed.

Parameter	Action
Active optical channel	Select the check box for each optical channel to be used.
Target dye for active channel	For each active optical channel, select the drop-down to choose the target dye.

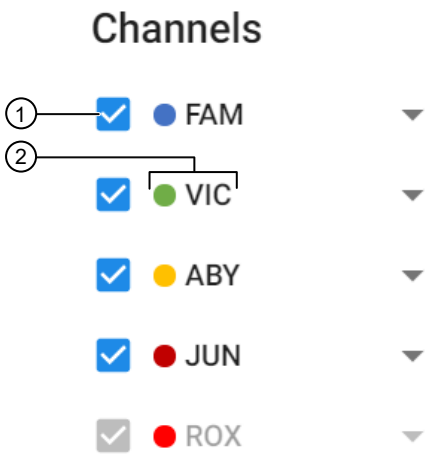


Figure 23 Optical channel dyes

① Dye channel check box
② Dye channel name

4. Modify PCR parameters as needed.

Parameter	Actions
Temperature	• Enter a value in the temperature fields. • Drag the slider bars to adjust the temperature.
Dwell times	Enter in seconds or minutes and seconds in mm:ss format.
Cycles	Set the number of cycles by entering a value into the **Cycles** field.
RNA-RT	Select **RNA-RT** to add an extra temperature step for RNA reverse transcription to cDNA for RNA samples. Not required for DNA samples.
Preheat	Select **Preheat** to add a preheat step. Sometimes called hot start, preheating the samples before PCR helps to reduce non-specific binding at lower temperatures.

(continued)

Parameter	Actions
Two or three-step cycling	Select the **Two Step** drop-down to select 2 or 3 step cycling.
Two-stage PCR cycle	Select **Two Stage PCR** to add a second PCR cycle stage.

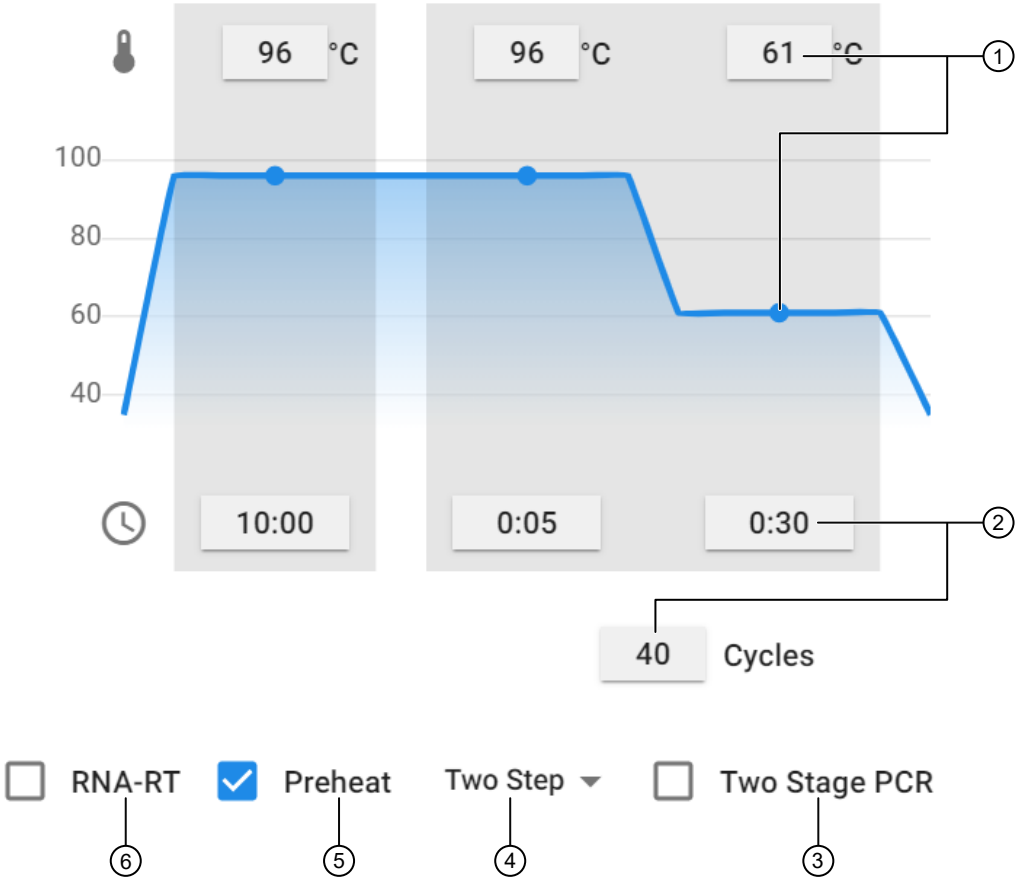


Figure 24 Protocol parameters

① Temperature settings fields and slider bar
② Time fields and cycles field
③ Two-stage PCR setting
④ Two or three step cycling option
⑤ Preheat setting
⑥ RNA-RT setting

5. Select **SAVE**.

B

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

Installation and environmental requirements

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 (2000 m)
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.

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" on page 84.

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 shipping lock screw on the top of the instrument.
   a. With the power off, unscrew the shipping lock screw on the top of the instrument.

   b. Insert the provided white plastic cap into the screw hole.

      For more information on removing the shipping lock screw, see "Uninstall the shipping lock screw" on page 89.

      IMPORTANT! To prevent damage to the instrument, the shipping lock screw 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. Turn on the power to the dedicated computer.

10. Install the software onto the dedicated computer. See “Download and install the desktop software” on page 86.

11. When the software installation is complete, use the USB cable to connect the instrument to the dedicated computer.

12. Turn on the power to 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 and install the desktop software

Computer requirements for the desktop software

Install the QuantStudio™ Absolute Q™ Digital PCR Software on the computer provided by Thermo Fisher Scientific, and use it to control the instrument. Thermo Fisher Scientific does not support the use of customer-provided computers to control the instrument.

However, you can install the QuantStudio™ Absolute Q™ Digital PCR Software on a customer-provided computer to use the software to import run data for analysis. Minimum requirements for a customer-provided computer are:

- Operating system—Windows™ 10 (64-bit)
- Dell™ OptiPlex XE3 Tower computer

Download the desktop software

1. Go to **https://www.thermofisher.com/us/en/home/global/forms/life-science/quantstudio-absolute-q-software.html**.

2. Download each software package.

Install the desktop software

1. Use a Windows™ Administrator account to log in to the computer on which you are installing the desktop software.

2. For each software package perform the following actions:
   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**.

3. Start the QuantStudio™ Absolute Q™ Digital PCR Software.

4. When prompted, accept the End User License Agreement.

5. When prompted, accept or decline the Privacy Statement that allows Thermo Fisher Scientific to use the ThermoFisher Connect Transfer Software to collect instrument run data.
   - If you accept, ThermoFisher Connect Transfer Software data transmission is activated.
   - If you decline, ThermoFisher Connect Transfer Software data transmission is not activated.

Note: Connect Transfer can be deactivated at any time by a user with administrator privileges within the QuantStudio™ Absolute Q™ Digital PCR Software by accessing the Help menu (?), opening the Privacy Statement, and declining data collection.

Update the instrument software

If the software and/or firmware on the instrument is not compatible with the desktop computer's software, you are automatically prompted to update the instrument software.

1. In the left pane of the QuantStudio™ Absolute Q™ Digital PCR Software, select to access the **Instrument** page.
   A dialog box appears with the following message, *Software update required to use this instrument*.

2. Click **UPDATE**.

3. In the **Absolute Q™ Instrument Setup** dialog box, click **Install**.

Note: The instrument information and setup file are pre-populated and cannot be changed.

4. When the update is complete, click **Finish**.

Note: When the update is complete, the instrument automatically restarts.

Moving the instrument

IMPORTANT! When moving the instrument the shipping lock screw must be manually installed before moving the unit, and manually removed after transport. Moving the instrument without the shipping lock screw 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.

Install the shipping lock screw

1. Power on the instrument.
2. Start 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 pane, select ⬓ to access the **Instrument** page.
6. Click on the instrument and select **Prepare for Shipping**.
   Wait until a message stating *Ready for Shipping* appears before proceeding.
7. Remove the white plastic plug from the shipping screw hole and place it in the bag attached to the shipping screw.
8. Insert the shipping screw and screw it finger tight. Do not over tighten.
9. Close the software and power off the instrument.

① Shipping lock screw

Uninstall the shipping lock screw

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 shipping lock screw from the top of the instrument.

① Shipping lock screw

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

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

The Security, Auditing, and E-signature (SAE) v2.2 software (SAE Administrator Console) is only compatible with the QuantStudio™ Absolute Q™ Digital PCR System.

For more information on Security, Auditing, and E-signature (SAE) v2.2, including definitions of accounts and roles, see the *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

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.

Depending on the way that your SAE administrator configures these features:

- 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 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 • 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 93)

Connect to the SAE server (page 94)

Enable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software (page 94)

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 standalone computer that is connected to the Absolute Q™ dedicated computer and optional Absolute Q™ companion computers
- SAE and Absolute Q™ software that is installed on the Absolute Q™ dedicated computer and is connected to optional Absolute Q™ companion computers
- SAE and Absolute Q™ software that is installed on an Absolute Q™ companion computer that is connected to the Absolute Q™ dedicated computer and other optional Absolute Q™ companion computers

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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 Absolute Q™ software that you are using.

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1. To download the SAE server and SAE Administrator Console software and Absolute Q™ application profile go to **https://www.thermofisher.com/us/en/home/global/forms/life-science/quantstudio-absolute-q-software.html**.

2. Install the SAE server and SAE Administrator Console software 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. At the SAE Administrator Console, an SAE administrator must install the application profile for the Absolute Q™ software before SAE can be used.

   The application profile contains default settings for the Absolute Q™ software.

   For information on installing application profiles, see the *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

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 server.

   Note: If using a dynamic IP address, enter the hostname instead of the IP address to prevent the loss of a connection.
3. (*Optional*) Click **Test Connection** to confirm that the connection information is correct.
4. Click **Save**.

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 94).
- Close all protocol or analyzed run files.

1. In the QuantStudio™ Absolute Q™ Digital PCR Software, select ⚙ **System** ▸ **Enable Security**.
2. 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 95.

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 112.

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 94).

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 SAE Administrator Console v2.2 (see the *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468)).

- Administrator
- Scientist
- Technician
- Service

IMPORTANT! SAE 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.	No	No	No	Yes
Application administration	Access to application administration menus.	Yes	No	No	Yes
Generate report	Create analysis reports.	Yes	Yes	Yes	Yes
Instrument Control					
Edit protocol	Edit run protocols.	Yes	Yes	No	Yes
Start run	Choose a protocol and start and stop instrument runs.	Yes	Yes	Yes	Yes
Run analysis					
Change thresholds	Change channel thresholds.	Yes	Yes	No	Yes
Edit groups	Edit group definitions.	Yes	Yes	No	Yes
Rename samples	Change sample names.	Yes	Yes	Yes	Yes
Assign samples	Assign samples to set groups or load a group set.	Yes	Yes	Yes	Yes
Hide samples	Show or hide samples from an analysis.	Yes	Yes	No	Yes
Run management					
Delete run	Delete a run from the database.	Yes	No	No	Yes
e-sign run	Place an electronic signature on a run.	Yes	Yes	No	Yes
Import run	Import and export runs to and from ZST files.	Yes	Yes	No	Yes
Edit run	Edit run features.	Yes	Yes	No	Yes
Security Configuration					
Configure security and auditing	Configure security and auditing in the SAE Administrator Console.	Yes	No	No	No
Audit History					
View action records	View action records.	Yes	No	No	No

(continued)

Function	Description	Role			
		Administrator	Scientist	Technician	Service
View system configuration	View system configuration records.	Yes	No	No	No
View application object records	View application object records.	Yes	No	No	No
View instrument run records	Edit run features.	Yes	No	No	No

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 *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

View audit records

For instructions to view audit action records for a protocol or an analyzed run, see the *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

For a list of actions that are audited, see “Actions that are audited” on page 98.

For instructions to view audit object records of a specific run, see “View audit object records” on page 98.

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.
   For information on selecting a run, see “Select a run” on page 38.
2. In the upper-right corner of the **Run** screen, hover over the **Run ID** and click **Copy to 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 your Thermo Fisher representative.

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:

- Sign in success
- Sign out
- Sign in failure
- Start Run
- Stop Run
- Accept Calibration
- Reject Calibration

Export audit records

For information on exporting audit records for a protocol or an analyzed run, see the *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

Sign data in the software

An e-Signature can optionally be added for plate setup and run results.

1. Chose from the following options to provide an e-Signature for run setup, plate setup, and run results.

Option	Actions
Run setup, **Instrument** page	• In the left pane, click to access the **Instrument** page.
Plate setup, **SETUP** page	1. In the left pane, click to access the **Runs** page. 2. Use the search fields to find a run or select a run from the list, then select the **SETUP** page.
Run results, **RESULTS** page	1. In the left pane, click to access the **Runs** page. 2. Use the search fields to find a run or select a run from the list, then select the **RESULTS** page.

2. Depending on the page you are on, click one of the following options to add your e-Signature:
   - RUN page — E-SIGN AND RUN
   - SETUP page — E-SIGN SETUP
   - RESULTS page — E-SIGN RESULTS
3. Select one of the following options from the drop-down list to indicate the meaning of the e-Signature.
   - Reviewed and Approved Plate Setup
   - Reviewed and Approved Plate Results
4. Enter your user name and password.
5. Click **Sign**.

If a run is signed and unmodified, the signature appears on reports that are created using **GENERATE REPORT**.

For information on how to view e-signature data in the SAE software, see *View and report audit and e-Signature records* in the *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

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 *SAE Administrator Console v2.0 or later User Guide for PCR systems* (Pub. No. MAN0017468).

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 100.
- For information on plate results e-Signature data, see “Review plate results e-Signature information” on page 107.

Review plate setup e-Signature information

The sections that follow provide descriptions of the information provided in the e-Signature plate setup record. Optionally, this information can be printed.

Signature metadata

This section provides information regarding the signature metadata for each e-Signature plate setup record.

Signature metadata

Object	Description
Meaning	E-Signature option selected
Signed Date	Date of e-Signature
Signed By	Name of user
Host ID	Instrument name
Full Name	User name
Status	Status of the signature: • CURRENT: Valid • OBSOLETE: Invalid
Role	The role assigned to the user who performed the run

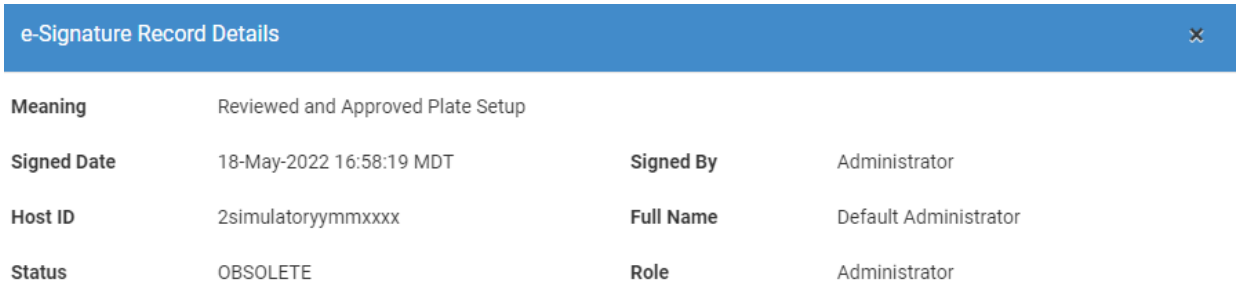

Figure 25 Signature details

Protocol information

This section provides information regarding the **protocol** section of the e-Signature plate setup record.

Protocol details

Object	Description
ScanChannelOne – ScanChannelFive	Up to five channels specified to scan. Each channel reflects the optical dye used. • FAM™ dye • VIC™ dye *(recommended)* or HEX™ dye • ABY™ dye • ROX™ dye • CY™5 dye *(recommended)* or JUN™ dye
RNAStep_Duration	Duration of RNA-RT step *(optional)*
RNAStep_Temperature	Temperature of RNA-RT step *(optional)*
PCRPreheat_Duration	Duration of pre-heat step *(optional)*
PCRPreheat_Temperature	Temperature of pre-heat step *(optional)*
PCR_Stage(1/2)_Step(1/2/3)_Duration	Duration of indicated stage and step
PCR_Stage(1/2)_Step(1/2/3)_Temperature	Temperature of indicated stage and step
Name	Name of protocol

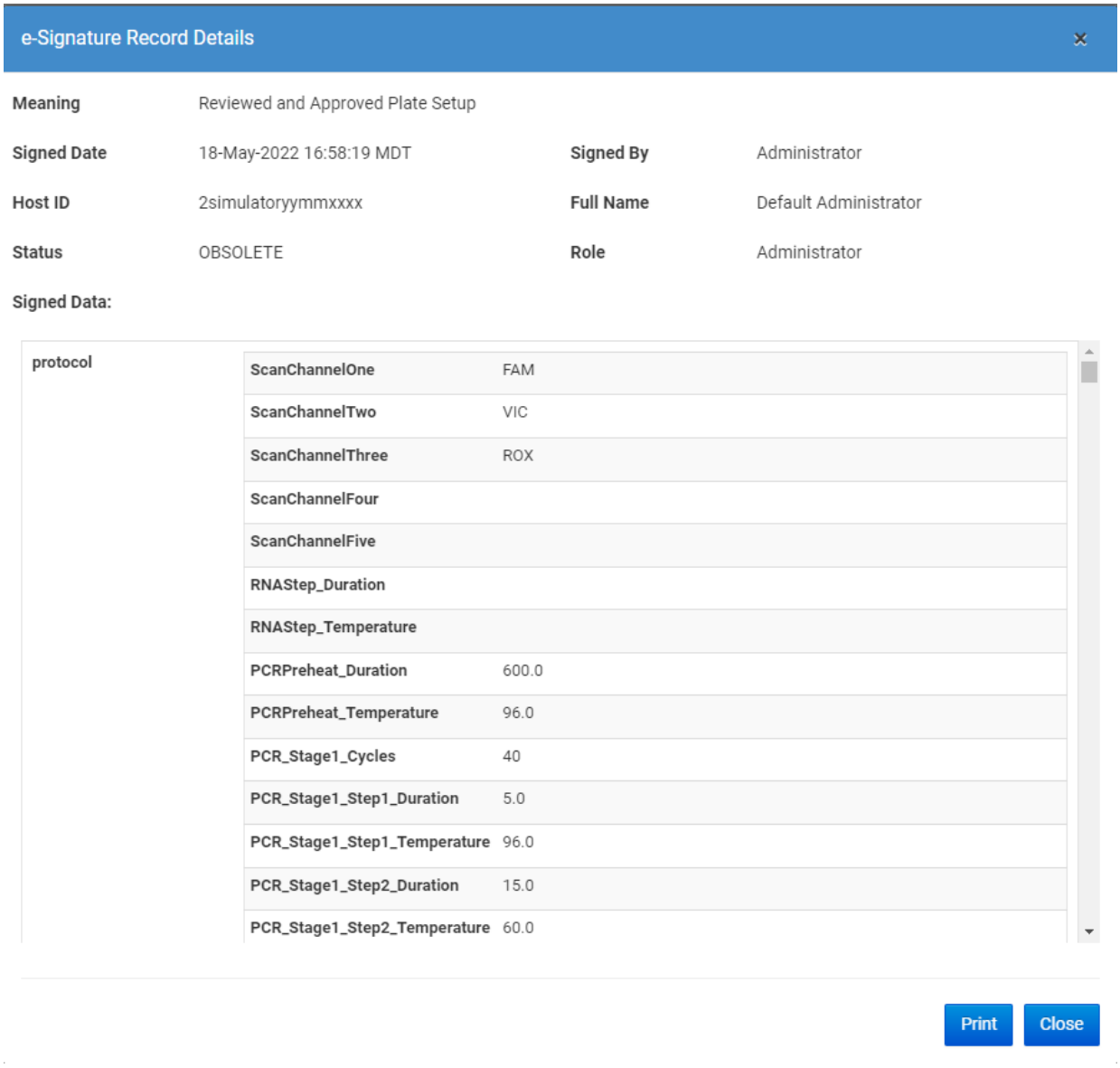

Figure 26 Plate setup — protocol

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.

For each color — blue, green, yellow, red, and dark red

Object	Description
Channel	Name of the target
ChannelType	Channel analysis setting
ChannelMaximum	For red only, (QC) maximum QC channel value
ChannelMinimum	For red, (QC) minimum QC channel value For other colors, the threshold dividing positive and negative numbers

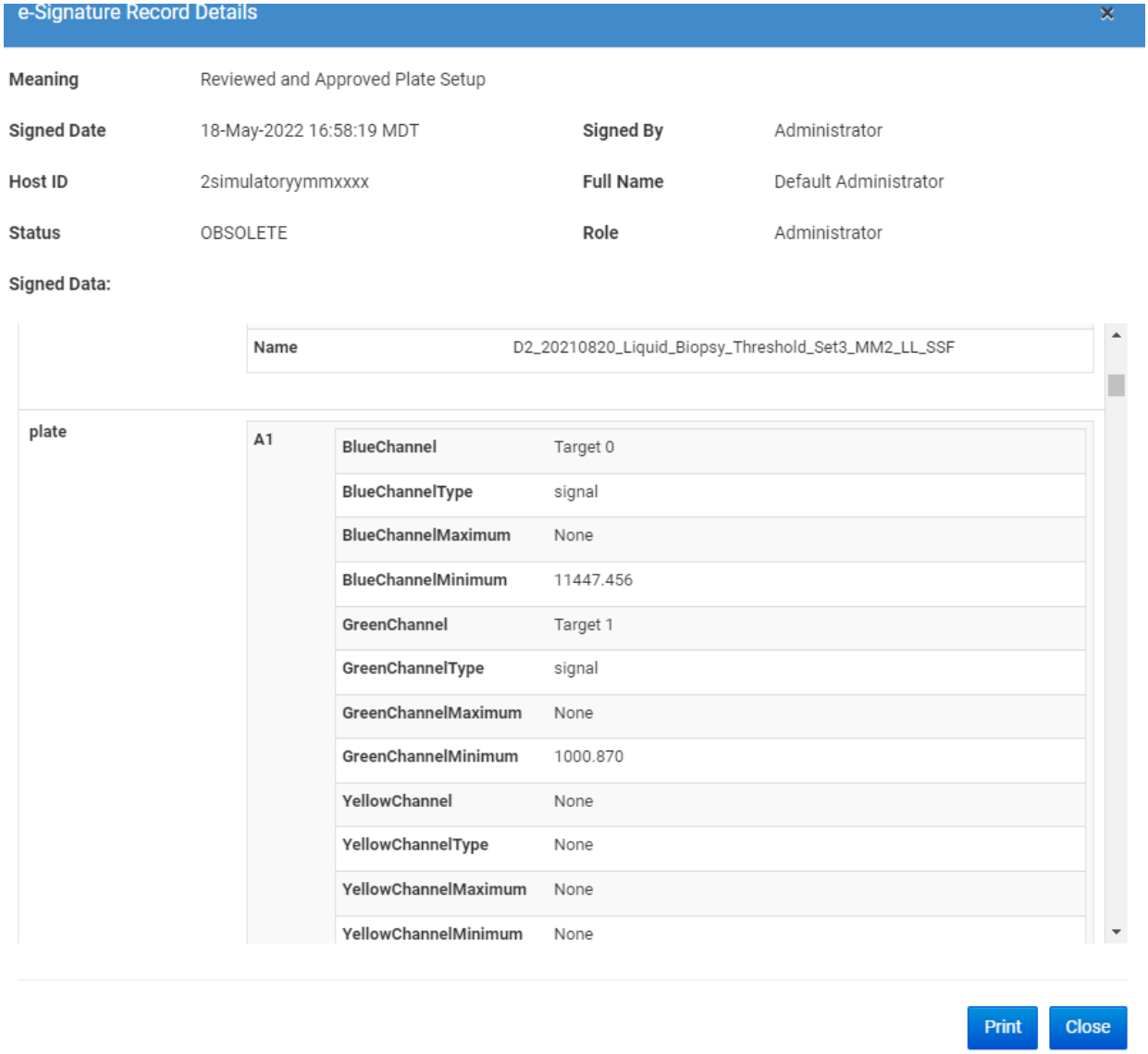

Figure 27 Plate channel detail by channel color

Additional plate information

This section provides information regarding the additional information provided in the **plate** section of the e-Signature plate setup record.

Other plate information

Object	Description
CNVReferenceNumber	If CNV statistics are used for this channel, the reference factor specified, otherwise None.
Name	Sample name
Group	Group name
GroupType	Group analysis setting
Hidden	Sample hidden in analysis: • TRUE • FALSE

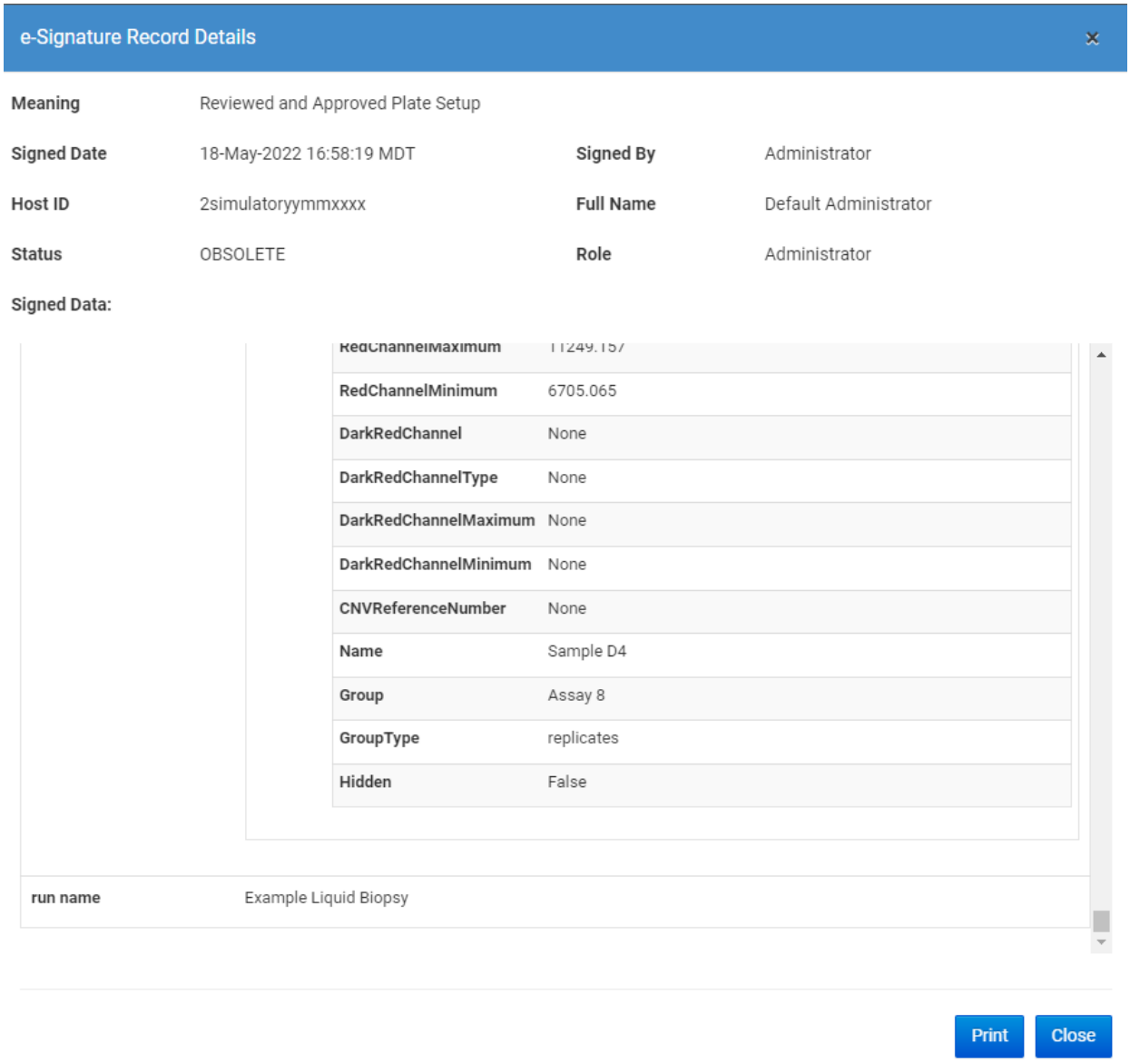

Figure 28 Plate information

Run metadata

The section provides information regarding the **run name** section of the e-Signature plate setup record.

Run metadata

Object	Description
run name	The name given to the run at the instrument

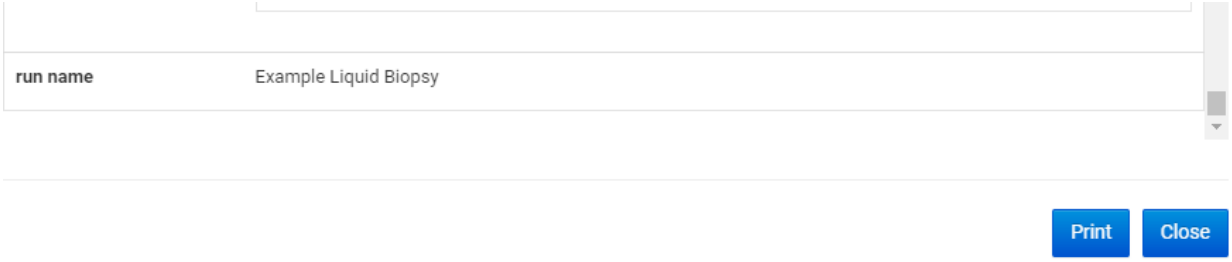

Figure 29 Run name

Review plate results e-Signature information

The sections that follow provide descriptions of the information provided in the e-Signature plate results record. Optionally, this information can be printed.

Signature metadata

This section provides information regarding the signature metadata for each e-Signature plate results record.

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

Figure 30 Signature information

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.

For each group, for each dye

Object	Description
name	Group name
std_conc	Concentration standard deviation of analysis group
cv_conv	Coefficient of variation of analysis group (where 100% = 1.00)
CI_95	95% percent confidence interval for concentration for analysis group
Total	One of the following options: • If replicates, this is the group average of microchambers • If pooled, this is the total pooles microchambers
Positive	Group positive microchambers
percent_cv	Coefficient of variation of analysis group (where 100% = 100)
cp_num	Calculated copy number (if available)
cp_CI_95	95% confidence interval of calculated copy number (if available)
Conc.(cp./uL)	Group concentration in copies per microliter

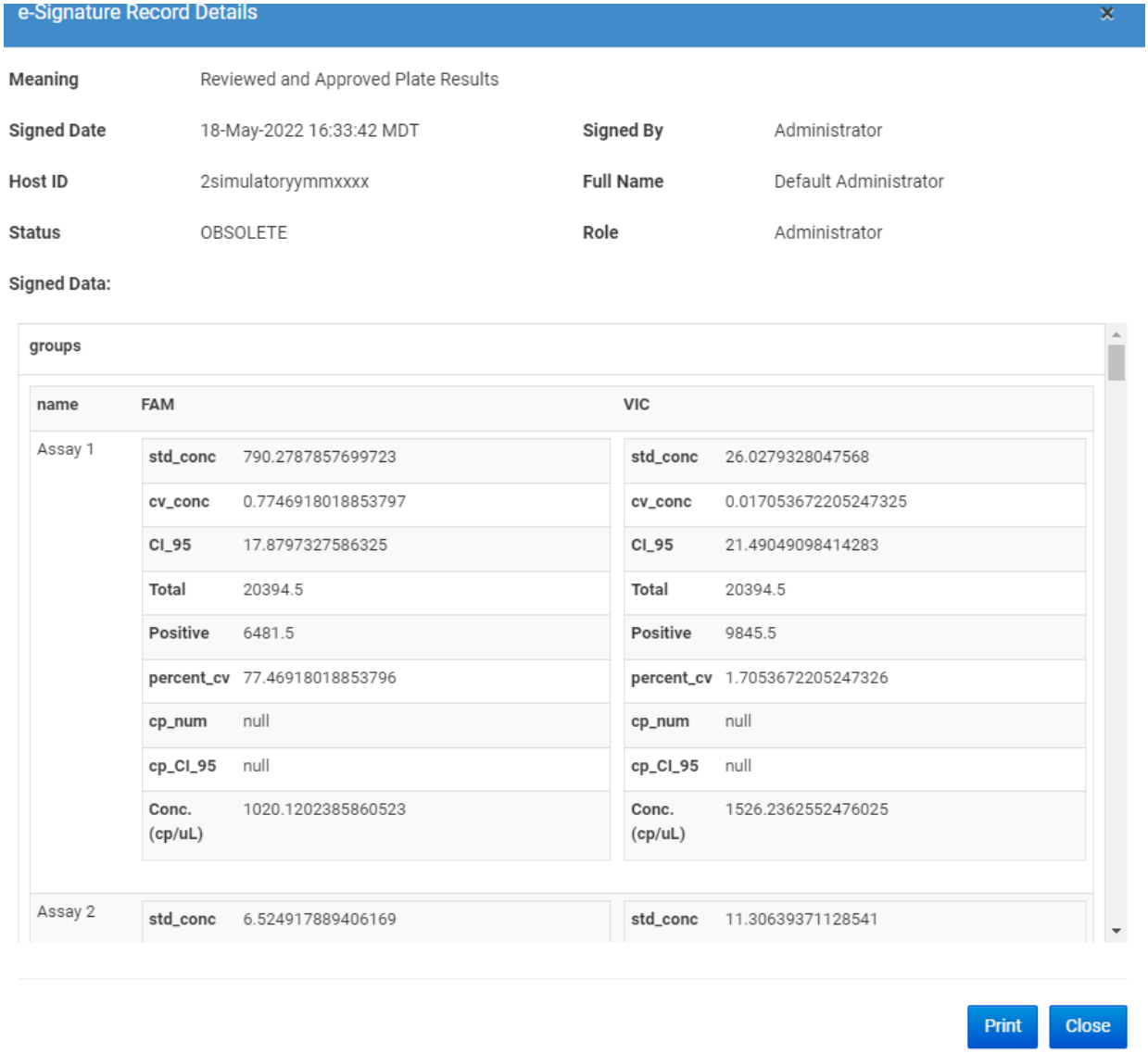

Figure 31 Plate results by group

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.

For each sample, for each dye

Object	Description
name	Sample name
std_conc	Concentration standard deviation of analysis group
cv_conv	Coefficient of variation of analysis group (where 100% = 1.00)
CI_95	95% percent confidence interval for concentration for analysis group

For each sample, for each dye *(continued)*

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
Positive	Group positive microchambers
percent_cv	Coefficient of variation of analysis group (where 100% = 100)
cp_num	Calculated copy number (if available)
cp_CI_95	95% confidence interval of calculated copy number (if available)
Conc.(cp./uL)	Group concentration in copies per microliter

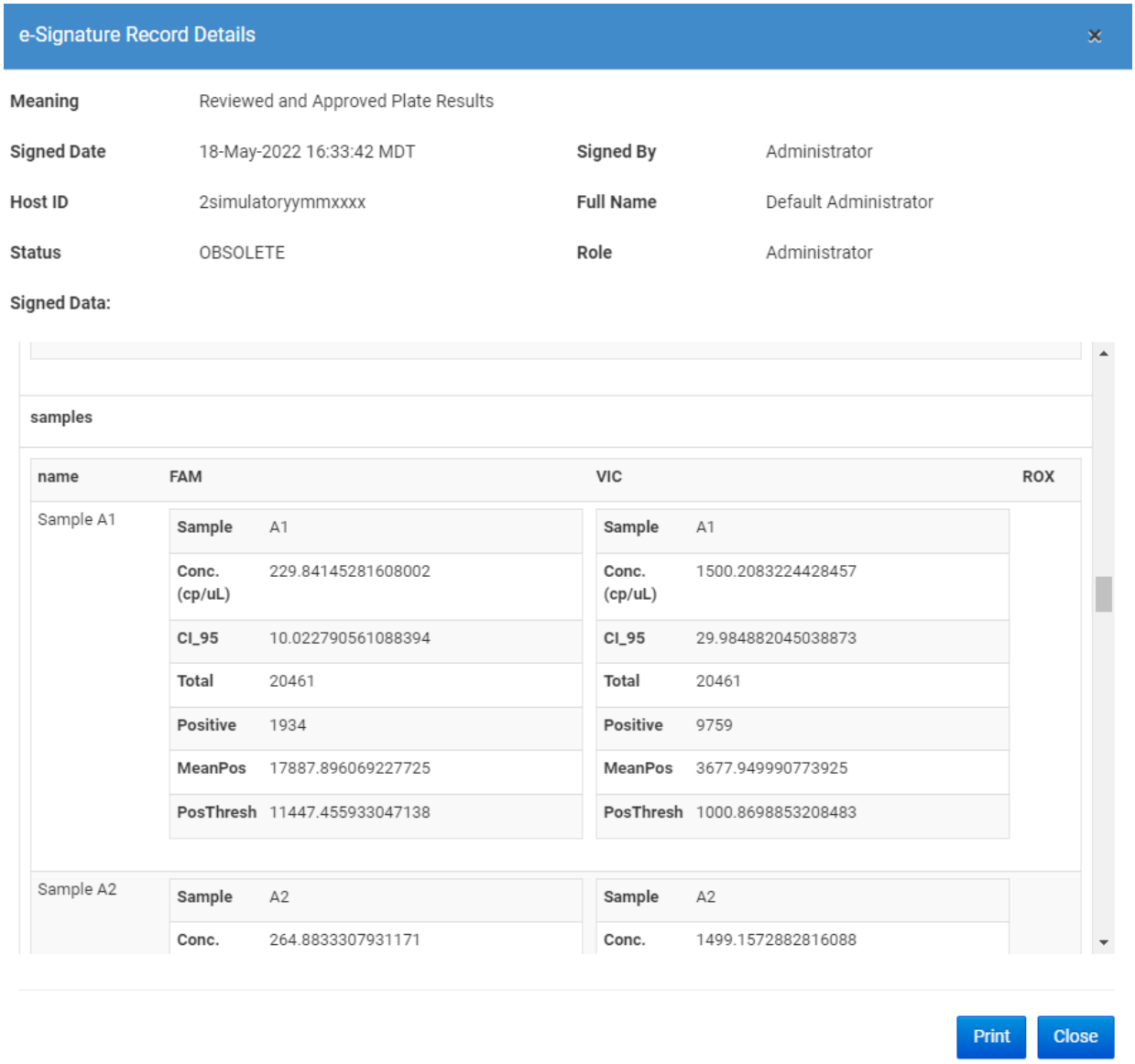

Figure 32 Plate results by samples

Run metadata

The section provides information regarding the **run name** section of the e-Signature plates result record.

Run metadata

Object	Description
run name	The name given to the run at the instrument

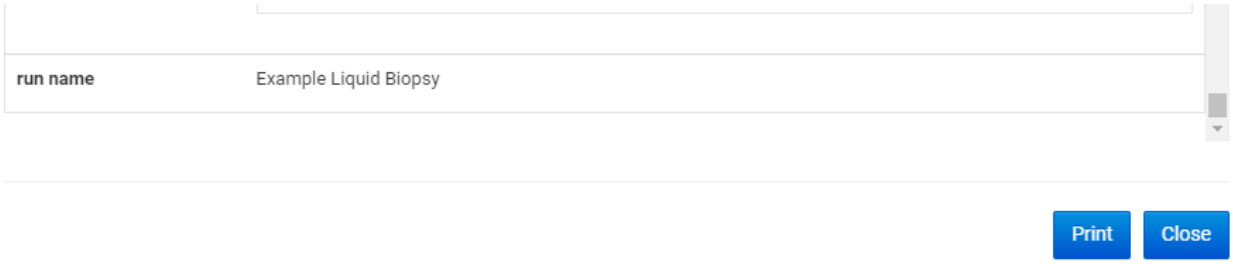

Figure 33 Run name

Disable SAE functions in QuantStudio™ Absolute Q™ Digital PCR Software

This procedure requires an SAE administrator account.

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**.

D

Maintain the instrument

Clean the 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 H, “Documentation and support”.

Maintenance

For best results when using the instrument, the following practices are recommended:

- The plate nest must be cleaned before each run.
- Ensure that the fan vents on the back and bottom of the instrument are not obstructed.
- Ensure that system dyes are calibrated on a yearly basis.

  Note: A warning message appears on the **Instrument** page 45 days prior to 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** page 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 may compromise run data for analysis.

For information on maintenance and service plans, contact technical support (see Appendix H, “Documentation and support”).

E Troubleshooting

Observation	Possible cause	Recommended action
QuantStudio™ Absolute Q™ Digital PCR Software is not connecting, front panel LEDs are white	Software connection error.	Power cycle system using power switch on the side of the instrument.
		Uninstall and re-install the software.
QuantStudio™ Absolute Q™ Digital PCR Software is not connecting, front panel LEDs are blue	Poor USB cable connection.	Power off the instrument. Unplug the power and USB cables from the instrument. Wait 10 seconds. Plug the power and USB cables back in to the instrument and dedicated computer. Power on and connect.
Front panel LEDs are red	Instrument error.	Power cycle the instrument using the power switch.
The Run status displays as DISCONNECTED	Port 8000 is blocked.	If a firewall or other application is using port 8000, remove it or use a different port.
Pressure leak error	Missing or damaged gaskets.	Make sure that all 5 columns of gaskets are present.
		Replace any damaged gaskets.
Instrument makes noise and LEDs are white one minute after power up	Instrument firmware startup error.	Power off the instrument. Unplug the power cable from the instrument. Wait 10 seconds. Plug the power cable back in and power on the instrument.
Barcode not found	Plate in backwards.	Well A1 should be at the top left of the plate tray.
	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.

Observation	Possible cause	Recommended action
Connection to the standalone SAE Administrator Console is lost *(continued)*	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 exising database. 4. Install the QuantStudio™ Absolute Q™ Digital PCR Software, see “Install the desktop software” on page 86. **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 dedication computer. Uninstall any other applications.
	Hardware failure.	Confirm all cables are properly connected
		Confirm that both devices have power.
		Contact **thermofisher.com/support**.
		1. Confirm all cables are properly connected.. 2. Confirm that both devices have power. 3. Contact **thermofisher.com/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.

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 • Contains raw images and the run ZST file • 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 • Contains logs from the QuantStudio™ Absolute Q™ Digital PCR Software • Contains logs from the instrument computer software and hardware not related to the camera • 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 support representative.

1. On the desktop computer, open the **Start Menu**.
2. Find 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:

   - *Absolute Q Starter Chemistry Run 31_8ea5ccfc_2022_04_01_data.fsa*
   - *Absolute Q Starter Chemistry Run 31_8ea5ccfc_2022_04_01_logs.fsa*

4. Send the files to your Thermo Fisher 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.

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

1. In the QuantStudio™ Absolute Q™ Digital PCR Software, select **System** ▸ **Download System Logs**.
   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 support representative for analysis using a file transfer program of your choosing.

F

Product Specifications

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) x30 (h)
Weight	Approximately 60 kg, 132 lbs
Connections	Power, USB 3.0 (to dedicated computer)
Cooling mode	Forced convection
Illumination	Rax, 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™ OptiPlex XE3 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: HEX™ dye is not supported for field calibration.

Note: A warning message appears on the **Instrument** page 45 days prior to 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** page until the dyes are calibrated.

#	Color	Excitation filter peak	Emission filter peak	System dyes
1	Blue	466	520	FAM™ dye
2	Green	514	560	VIC™ dye *(recommended)* HEX™ dye
3	Yellow	549	589	ABY™ dye
4	Red	589	625	ROX™ dye
5	Dark Red	630	684	CY™5 dye *(recommended)* JUN™ dye

Safety

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 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 triées. 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 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.

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.

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 triées. 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	*Safety requirements for electrical equipment for measurement, control, and laboratory use – Part 1: General requirements*
IEC 61010-2-010	*Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials*
IEC 61010-2-081	*Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-081: Particular requirements for automatic and semi-automatic laboratory equipment for analysis and other purposes*

EMC standards

Reference	Description
EMC EN 61326-1	*Electrical Equipment for Measurement, Control and Laboratory Use – EMC Requirements – Part 1: General Requirements*
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)	*U.S. Standard Radio Frequency Devices* This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

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
  www.cdc.gov/labs/pdf/CDC-BiosafetymicrobiologicalBiomedicalLaboratories-2020-P.pdf
- Laboratory biosafety manual, fourth edition. Geneva: World Health Organization; 2020 (Laboratory biosafety manual, fourth edition, and associated monographs)
  www.who.int/publications/i/item/9789240011311

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
  www.cdc.gov/labs/pdf/CDC-BiosafetymicrobiologicalBiomedicalLaboratories-2020-P.pdf
- Laboratory biosafety manual, fourth edition. Genève: Organisation mondiale de la santé; 2020 (Laboratory biosafety manual, fourth edition, et monographies associées)
  www.who.int/publications/i/item/9789240011311

Documentation and support

Related documentation

Document	Publication number	Description
QuantStudio™ Absolute Q™ Digital PCR Starter Kit User Guide	MAN0025653	Describes the setup, use, and analysis of runs using the QuantStudio™ Absolute Q™ Digital PCR Starter Kit assay. (Catalog No. A52732)
QuantStudio™ Absolute Q™ Digital PCR System Site Preparation Guide	MAN0026431	Describes the site preparation required for installing the QuantStudio™ Absolute Q™ dPCR System.
SAE Administrator Console v2.0 or later User Guide for PCR systems	MAN0017468	Describes the setup and use of the Security, Auditing, and E-signature (SAE) module.

Note: For additional documentation, see “Customer and technical support” on page 131.

Customer and technical support

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

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

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

Limited product warranty

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